Grassland at the heart of circular and sustainable food systems

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Foreword

We would like to welcome all delegates of the European Grassland Federation 29th General Meeting to Caen, France. The previous EGF General Meeting held in France took place in La Rochelle in 2002. During these last two decades, food systems have been increasingly challenged for their impacts on climate change, loss of biodiversity and other environmental issues, while at the same time the question of food security in Europe is being raised together with successive health and political crises. Therefore, the main theme of the EGF 2022 General Meeting is *Grassland at the heart of circular and sustainable food systems*. This EGF meeting will consider the contributions of grasslands to the development of circular, healthy and sustainable food systems. Grasslands are widely acknowledged for their role in preserving natural resources and biodiversity and in soil carbon sequestration, but at the same time ruminants, the livestock that are used to manage and utilize grassland for food production, are blamed for their emissions of methane and their inefficient use of resources. As the expectations regarding food systems are multi-faceted and because the importance of each service provided by grasslands varies according to the stakeholder's visions, local context and farming practices, achieving the objectives requires the search for new compromises. The analysis of the relationship between services, their drivers encompassing economic, social, biological and biotic regulatory processes and the search for compromises will be the keystone of this meeting.

The meeting has five themes: (1) Putting grasslands into perspective; (2) Highlighting the bundles of services provided by grasslands; (3) Using biodiversity to reduce vulnerability and increase resilience of grassland-based systems; (4) Looking for synergy between animals, grasslands and crops; (5) Illustration of initiatives for the transfer and co-construction of innovations on and for grassland.

There are five mid-conference tours organized in Normandy to discover the high value habitats and attractive landscape of wet grasslands, the dairy and beef production from grassland-based systems, and the diversity of animal-based products derived from grassland. In addition, there is a visit to a horse farm as Normandy is the primary French region for horse breeding. The post-conference tour will visit Omaha beach and an impressive American cemetery, the Mont St Michel Bay with an amazing crossing of the bay by foot, sheep flocks grazing on salt-marsh grassland, and a visit to the old city of Rennes.

The General Meeting is organized by INRAE and the University of Caen Normandy. They develop a wide range of research projects including ecology, plant and animal science, environmental and social sustainability, grassland and grazing management, system analysis and whole value chain perspectives.

We would like to thank all authors for their contributions, numerous reviewers for their valuable remarks which have helped to ensure the high quality of the papers presented, the members of the scientific and organizing committees, the secretary of EGF, and our sponsors and all delegates attending the conference.

We wish that the 29th General Meeting of EGF will provide novel insights for grassland science and stimulate fruitful discussions and networking and that all participants will have enjoyable days in Normandy.

Dr Jean-Louis Peyraud	Luc Delaby	Marie Pascale Prudhomme
President, European Grassland Federation	*Chair Scientific Committee*	*Secretary Organizing committee*

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Theme 1.
Putting grasslands into perspective

Why and how to support the supply of non-provisioning ecosystem services by European grasslands through the Common Agricultural Policy?

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Abstract

This paper analyses why and how European grasslands should and could be supported through the Common Agricultural Policy (CAP). We first recall the importance and diversity of grassland in the European Union. This diversity reflects different climatic, historical and socio-economic contexts, and different management practices that result in heterogeneous climatic and environmental impacts. It is mainly because grassland provides non-provisioning ecosystem benefits that this farmland use should and could be supported by public policies. Unfortunately, the CAP, including the policy that will apply over the five-year period 2023-2027, does not sufficiently protect or encourage the environmental benefits that grassland can provide. We then show how simple principles of public economics may be called to support non-provisioning ecosystem services of grassland. We conclude by highlighting some trade-offs that such an orientation of the CAP might induce, and by discussing its compatibility with the European Green Deal ambition and objectives.

Keywords: European Union, grassland, climate, environment, public economics, payments for climatic and environmental services

Introduction

European agriculture has been governed by the Common Agricultural Policy (CAP) for sixty years. The CAP was initially a production, productivity and income-support policy. The first two dimensions have lost their importance. The income support dimension is now completed by an increasing social issue linked to the unequal distribution of budgetary aids between products, farms and countries. Environmental and then climatic objectives and instruments have been added progressively since around 30 years ago, and the first major reform of the CAP was in 1992. More recently measures have been added that are targeted on climate change mitigation and adaptation. The European Green Deal (GD) launched in December 2019 aims to make the European Union (EU) 'the world's first climate-neutral continent by 2050' by proposing a new decarbonized sustainable growth model for the EU (EC, 2019). It includes all environmental dimensions as well as health and social justice issues, including those linked to agricultural and food systems. To that end, it adopts a whole food chain approach encompassing objectives and actions not only on the agricultural supply side but also on the food demand side where it encourages a shift towards healthy and environmentally friendly diets. Making the CAP compatible with the GD questions this policy; more specifically its focus on agriculture only, the choice of policy objectives, and the choice of policy instruments (Guyomard *et al.,* 2020).

Within that general framework, this paper analyses specifically why and how grassland should and could be supported through the CAP. After this introduction, the importance and diversity of grassland in the EU is recalled. The diversity of grassland management intensity reflects variable climatic, historical and socio-economic contexts, and results in different environmental impacts. It is mainly because grassland provides non-provisioning benefits that this farmland use should and could be supported by public policies. Unfortunately, the CAP, including the policy that will apply over the five-year period 2023-2027, does not sufficiently protect or encourage the environmental benefits that grassland can provide. We then show how simple principles of public economics may be called to support non-provisioning ecosystem services of grassland. We conclude by highlighting some trade-offs that such an orientation of the CAP might induce, and by discussing its compatibility with the GD ambition and objectives.

Importance and diversity of grasslands in the European Union

According to the European Commission (EC) Regulation 1307/2013, permanent grassland and permanent pasture, hereafter called simply permanent grassland, are types of 'land used to grow grasses or other herbaceous forage naturally (self-seeded) or though cultivation (sown) and that has not been included in the crop rotation of the holding for five years or more' (EC, 2013). Permanent grassland differs from temporary grassland of less than five years of age included in the rotation. Both types of grassland may benefit from decoupled and coupled direct aids of the first pillar of the CAP. Before the EC defined permanent grassland and temporary grassland, first in 2004 (EC, 2004) and more precisely in 2013 (EC, 2013), both types of grassland were defined in rather vague terms (Reheul *et al.,* 2004). However, the EC definition is not without drawbacks, including from a public policy perspective, since it does not distinguish grassland categories according to the non-provisioning ecosystem services that they provide, particularly as these non-provisioning services should be the basis of climatic and environmental measures of the CAP. The problem is exacerbated by the fact that other classifications of grassland have been developed that rely on other environmental criteria, for example, on high nature value grassland vs seeded grassland (Baltic Environmental Forum, 2017).

Approximately 50% of European land (4.1 million square km in 2018) is farmed, and approximately one-third of this farmed area is covered by permanent grassland, with substantial differences between the various Member States (MS). According to the EC (2021), the 2018 share of permanent grassland in the total agricultural area was equal to 57.7% in Ireland, 34.2% in the Netherlands, 32.9% in Luxembourg and 28.2% in Belgium, but only to 5.5% in Sweden and 5.7% in Finland. In a general way, this share is lower in regions where climatic conditions are harsh, notably in northern Europe (EC, 2021). In Mediterranean Europe, permanent pastures are also a very important land use, for example 31.5% of the total agricultural area in Portugal. There is even an increasing trend in this land use in less favourable regions because of extensification and the decline in crop production. In mountainous areas, there is a decline in the total area of extensive permanent grazing. However, this still does not compensate for the increase resulting from extensification in the more marginal areas. These figures are subject to uncertainty, some estimates suggesting more important shares of permanent grassland (Peyraud *et al.,* 2012).

Permanent grassland areas and their shares of total agricultural area have declined since the early 1970s. They continued to decrease after 2000 with differences according to countries and sub-periods. Furthermore, where their importance increased, it was not because of a 'positive' investment choice in extensive livestock production but more because of a decline of crop production systems and extensification on the more peripheral and less productive soils. Permanent grassland areas increased between 2015 and 2018 in most MS, notably in northern, central and eastern countries, as well as in the United Kingdom; it was stable in western countries and declined in southern Europe of Greece, Italy, and the South of Spain (Mosquera-Losada *et al.* (2019). This last sub-period shows that the third greening measure of the CAP, that applies from 2015 and obliges maintaining the ratio of permanent grassland in total utilized agricultural area (UAA), is not a success in all regions. Since a large part of grassland is permanent grassland, permanent and total grassland areas evolve similarly. In contrast with permanent and total grassland evolutions, temporary grassland has either been maintained or has increased since the early 2000s (Mosquera-Losada *et al.,* 2019). In part, this is due to the replacement of decreased permanent grassland by increased temporary grassland. In southern Europe, the registered increase in permanent pastures is often associated with a declining productivity of soils and related abandonment of former farm systems (annual crops and/or temporary grassland), and seldom with an investment in permanent pastures valorising the ecosystem services they provide and their fundamental climatic mitigation role.

The diversity of grassland also derives from the variability of grassland management intensity that relates to three main factors: mowing frequency, fertilization, and grazing pressure and management (Blüthgen *et al.,* 2012). From that perspective, Estel *et al.* (2018) combined maps of mowing frequency, livestock distribution and grassland management frequency to define six clusters of similar grassland management intensity (Table 1). They summarized their work by highlighting three main results: first, 'highest grassland intensity [in clusters 1 and 2] occurs in regions with the highest grassland productivity'; second, 'lower grassland management [in clusters 3 and 4] was often found in socio-economically marginal regions facing rural depopulation and abandonment'; third, the lowest intensity clusters 5 and 6 separate West Europe from East Europe with, in addition, a link to long fallow periods or land abandonment. This kind of work could be used to regionalize farm policy measures aimed at supporting incomes of cattle producers or encouraging the provision of environmental services by grassland. This would require combining the map of grassland management intensity with maps of income distribution and environmental services. Both are likely to vary in function of grassland management intensity.

European grasslands should be supported on the basis of the non-provisioning ecosystem services they provide

Strengths and weaknesses of European livestock

Livestock production in the European Union (EU) is increasingly criticised because of climatic, environmental, health and animal welfare arguments (Buckwell and Nadeu, 2018; Guyomard *et al.,* 2021). Criticisms relate notably to emissions of greenhouse gases (GHG), ammonia, nitrogen oxides and volatile organic compounds. According to the European Environment Agency (EEA), agricultural GHG emissions represented 11% of EU-28 emissions in 2018 under the form of three gases: methane (55%) linked to farm animal digestion (enteric fermentation) and manure digestion, nitrous oxide (43%) linked mainly to nitrogen fertilization, and carbon dioxide (2%). While European GHG emissions declined by one quarter between 1990 and 2013 (because of the herd decline in Eastern European countries after the fall of communist regimes, increased feed efficiencies and improved fertilization practices), they have been slightly increasing since 2013. As a result, an extension of the short-term trend until 2030 shows that EU agriculture is not on the right track to contribute to the Green Deal objective of zero

Table 1. The six grassland clusters defined by Estel *et al.* (2018).

Cluster	Grassland management intensity	Determinants of grassland management intensity[1]			Main locations
		1	2	3	
1	High	++	-	-	Northern and southern Germany, the Netherlands, Ireland, United Kingdom
2	High	++	+	+	North-West Europe, notably in Ireland, Wales, the Netherlands, northern France
3	Medium	+	-	+	Ireland, northern and central France, United Kingdom, northern and central Spain, Greece
4	Medium	+	-	--	The Alps and the Pyrenees
5	Low	--	-	--	Northern Ireland, United Kingdom, the Netherlands, Germany, Hungary
6	Low	-	-	-	Northern and southern Germany, United Kingdom, the Extremadura, mountainous areas (e.g. Carpathians, central France), eastern Poland, Latvia

[1] Mowing frequency (determinant 1), livestock density (determinant 2), grassland management frequency (determinant 3); the deviation from the global mean of each determinant can be greater than 1 (++), between 0 and 1 (+), between -1 and 0 (-), and lower than -1 (--).

GHG net emissions by 2050 (Figure 1). Furthermore, inventoried agricultural GHG emissions do not include fossil fuel consumption in equipment and buildings, and fossil energy used for the synthesis of mineral fertilizers. Critics also extend to the land consumption associated with livestock production, as livestock agriculture requires more land than crops to provide the same level of output, if expressed in just terms of calories or protein. They also include health considerations related to the impact of the use of antibiotics in livestock on antimicrobial resistance, zoonosis risks, and adverse health effects linked to an excessive consumption of animal products, notably red and processed meats. Finally, animal production is increasingly questioned because of animal welfare considerations.

Specifically, cattle livestock systems relying on intensive grazing practices have negative effects on many ecosystem services, notably on biodiversity of plants (Herrero-Jáuregui and Oesterheld, 2018; Olff and Ritchie, 1998), insects (Takagi and Miyashita, 2014; Van Klink *et al.,* 2015) and other animal species (we do not address here the question of intensive monogastric systems). Such cattle livestock systems are often linked to strong decline in vegetation biomass, and many plant species cannot tolerate the very intensive biomass removal, in particular in unproductive environments where plant regrowth is slow (Olff and Ritchie, 1998). Trampling by grazing animals and associated soil compaction can decrease plant biodiversity (Olff and Ritchie, 1998). In addition to antimicrobial resistance issues associated with the over-use of antibiotic medicine, use of parasiticide medicine in livestock also has negative effects on biodiversity, especially on arthropod populations (Floate *et al.,* 2005).

However, too often, critics forget the other side of the coin. The economic, social and cultural importance of EU livestock production can be illustrated by three figures: around 40% of European agricultural production is provided by animal production, more than 50% of agricultural holdings have farmed animals, and European livestock farms account for several million direct jobs – 4 million in 2010 according to the Animal Task Force (2017). Some livestock systems, notably grassland-based extensive systems, provide environmental benefits by sequestering carbon, improving water and soil quality, preserving biodiversity and maintaining diversified open landscapes (Dumont *et al.* (2019). Many of them are classified as High Nature Value (HNV) farming systems, for example the extensive grazing under open tree cover in the sylvo-pastoral systems in Iberia (Bugalho *et al.,* 2018; Pinto-Correia *et al.,* 2018). As HNV, these grassland-based systems provide a series of ecosystem services far beyond climatic and environmental benefits. Farmed animals recycle biomass and protein that cannot directly be used as human food (Mottet *et al.,* 2017). In particular, ruminants use grasslands or other lands that cannot be cultivated and hence cannot be directly used to produce food for human consumption. Meat products

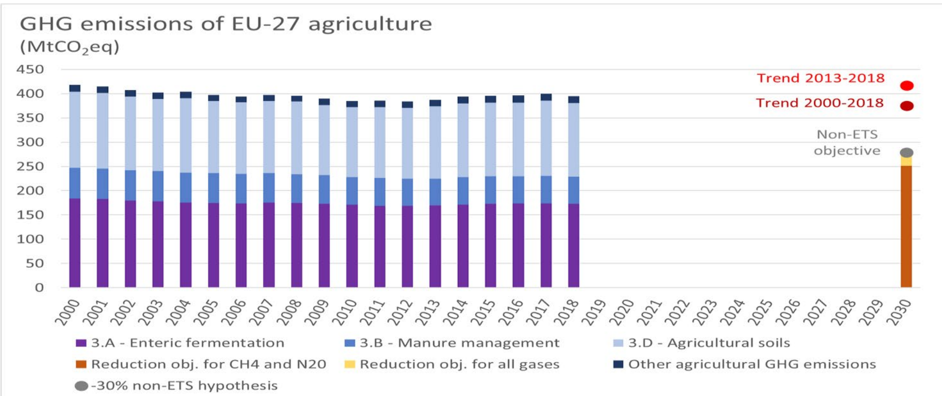

Figure 1. EU-27 Agricultural GHG emissions in million tons of carbon dioxide equivalent (MtCO_2eq), evolution 2000-2018 and projections to 2030 (Guyomard *et al.,* 2020 from EEA data).

consumed in accordance with nutritional recommendations provide proteins of high nutritional quality (containing the nine essential amino acids), micronutrients and bioactive components. Likewise, dairy products provide calcium, iron and magnesium that are crucial for bone development.

Why support grasslands?

Many of the positive and negative impacts of animal production and consumption are public goods or public bads (i.e. positive and negative externalities) that are not – or only very partially – taken into account by private markets. It follows that there is very likely an under-provision of public goods and an excessive provision of public bads. These market failures open the door for public policies in order to reduce the damage and increase the benefits linked to animal production and consumption.

From that perspective, Guyomard *et al.* (2021) recommend an enhanced application of the polluter-pays principle (PPP) targeted on negative climatic, environmental and nutritional externalities. This could be achieved by taxing the latter, which would send the right price signal to all actors in the food chain, from producers to final consumers, in a context where current prices highlight inadequate pricing of animal products (Pieper *et al.,* 2020). They point out that it will be very difficult to obtain a political agreement on a European taxation scheme. They then show that the same climatic and environmental outcome on the supply side could achieved by a second-best policy relying on the current instrumentation of the CAP, more specifically by effectively reinforcing climatic and environmental requirements that a producer should respect to receive CAP subsidies (for more details on CAP conditionality, see the section below). This policy of penalizing negative externalities in accordance with the PPP would increase the legitimacy of its counterpart, the provider-gets principle (PGP), which will aim at increasing the provision of amenities, in particular those linked to grassland-based systems. It is because grassland systems may provide non-provisioning services – by sequestering carbon, saving natural resources and improving their quality, protecting biodiversity and maintaining diversified open landscapes that are more resilient to extreme weather events such as wildfires – that they could and should be supported by public policies. However, this may create tensions between different policy tools (Pinto-Correia and Azeda, 2017). A direct consequence of the PGP is that corresponding incentive payments should be proportionate to provided amenities: the greater the services, the higher the payments. This means that it is time to shift from a logic of cost compensation based on an obligation of practices to a logic of payments for climatic and environmental services based on an obligation of results/impacts (Herzon *et al.,* 2018). There are at least two problems with this approach. First, it requires the measurement of climatic and environmental services that vary according to grassland systems. It also requires reliable results-based indicators. Second, strict proportionality to services that are provided may lead, because of the CAP budget constraint, to insufficient payments per hectare in regions where viable alternatives to grassland-based systems do exist and where it is very important to maintain the latter from a climatic and environmental point of view.

Before explaining how grassland could be efficiently supported through the CAP, it is necessary to explain how the current CAP supports – or not – grassland in the general framework of regulations applied to animal production.

Support to European grasslands through the CAP

A very brief history of the CAP

The CAP is 60 years old (Chatellier and Guyomard, 2022). The policy aimed at increasing agricultural productivity thanks to technical progress and sectoral restructuring, increasing farm incomes, stabilizing agricultural markets, and ensuring sufficient food availabilities and reasonable food prices for European consumers. The instruments used to achieve these objectives were the guarantee of production prices (thanks to public purchases) completed by trade instruments, more specifically variable import duties to limit imports from third countries and variable export subsidies to bring back European export prices to world prices. During this first period, they were no explicit environmental objectives and instruments.

This first CAP was an unquestionable success if the latter is measured against productive objectives, allowing notably the EU to become a net exporter of agricultural and agri-food products despite successive enlargements to countries that were often net importers. The first drawbacks appeared from the mid-1970s and became more significant in the 1980s. They were linked to the downward trend in real farm incomes, the increase of CAP budgetary expenditure, the uneven distribution of support among products, producers and countries, and the first negative impacts of modernized, intensified, simplified and specialized crop and livestock systems on natural resources and the environment. Specific and sectoral measures were adopted during the 1980s to address these challenges, notably by introducing milk quotas in 1984 and crop set-aside a few years later. However, it is mainly because of the external pressure that the CAP was reformed in 1992, the EU being, rightfully, accused of competing unfairly on international markets thanks to this policy.

The 1992 CAP reform has defined the path that we are still on today, 30 years later. The path chosen was to progressively eliminate trade-distorting instruments by suppressing guaranteed prices and export subsidies, and by disciplining import measures. These provisions allowed the EU to sign, two years later, the Uruguay Round Agreement on Agriculture (URAA). The latter still governs world agricultural trade in a context where the current round (Doha Round) has not yet been completed. However, bilateral agreements are multiplying. Like the URAA, the latter do not include significant commitments aimed at better protecting natural resources and reducing agricultural GHG emissions. Price tools (supported by both the consumer and the taxpayer) were replaced by direct aids (supported by the taxpayer) that were progressively disconnected from production choices and volumes. Although around 10% of direct aids remain coupled to production, essentially in the sectors of small and large ruminants (notably for suckler cows), the decoupling process of income support measures, initialised in 1992 and continued in subsequent reforms in 1999 and 2003, is now achieved with European prices close to world prices although generally higher thanks to tariff and non-tariff barriers on imports. However, CAP measures adopted since 1992 have failed to significantly reduce agricultural GHG emissions and curb biodiversity loss in agro-ecosystems. This raises the central question of the future of decoupled income support direct aids (that still today represent the lion's share of CAP budget, around 65% with disparities among MS), in a context where the issue of unequal distribution of this budget has not been really resolved despite several policy changes aimed to address this question (2008 (2013 and 2022) and in a context where climate and environment issues are more and more important and urgent.

The progressive integration of environmental objectives and measures in the CAP

From 1992, the CAP has progressively included environmental objectives and measures focused first on diffuse pollutions, and more recently on climate change and biodiversity. Until now, success was very limited and mainly local (Dupraz and Guyomard, 2020).

CAP payments for Less Favoured Areas (LFA) were introduced in the 1970s. Their primary objective was to support incomes of farmers located in these areas with, in addition, positive environmental benefits linked to the maintenance of agricultural use of land and diversified landscapes. The payments have benefitted cattle livestock producers who are proportionally more numerous in LFA.

Implementation of Agri-Environment Schemes (AES) became obligatory for MS from 1992. Very quickly, the different MS have developed a large range of AES on a large spectrum of ecological objectives (Uthes and Matzdorf, 2013). AES were renamed Agri-Environment and Climate Schemes (AECS) from

2015 to reflect the addition of climate change as an explicit objective. AECs cover today around 25% of European farmland. Defined at the level of each MS, they include (1) system measures at the farm scale (for example, in France, measures targeted on grazing and pastoral systems that aim at maintaining practices favourable to the environment: low stoking rate, no pesticide use on permanent grassland, no ploughing of permanent grassland areas); (2) localized action measures at the scale of the parcel of a set of parcels (measures targeted on the preservation of wetlands, the protection of biodiversity, the quality of soils, water or landscapes); and (3) non-zoned measures targeted on the protection of bees, endangered plant species and breeds, etc. AECS are voluntary instruments. Despite some well-documented drawbacks (Cullen *et al.,* 2018), AECS can be efficient instruments. However, very often, farmers chose measures that were less damaging from an economic point of view and that have low environmental benefits (Azeda *et al.,* 2021). A supplementary weakness is that AECS only compensate farmers for income forgone linked to the use of more environmentally friendly practices or systems. Conversion subsidies to organic farming are a specific example of AECS.

The second environmental tool of the CAP was introduced in 2003 and applied from 2005. From that date, the granting of first-pillar income support direct subsidies is conditional on compliance with (1) minimum Statutory Management Requirements (SMR) related to public, plant and animal health, food safety, animal welfare, and environmental protection,; and (2) Good Agricultural and Environmental Conditions (GAEC) corresponding to basic farmland management rules. Unlike AECS, cross-compliance covers the whole agricultural area. Until now, cross-compliance was, however, not sufficiently constraining to significantly reduce the negative environmental footprint of European agriculture.

The third environmental instrument was introduced in 2013. It conditions the granting of 30% of first-pillar direct aids to the respect of three criteria relative to the maintenance of permanent grasslands, minimal crop diversification and a minimal preservation (5%) of agroecological infrastructure such as trees, hedges, wetlands, etc. More specifically, the ratio of permanent grassland to agricultural land should not decrease in each MS, with a 5% margin of flexibility. The criterion can be applied at the regional level. In addition, each MS must designate areas of environmentally sensitive permanent grassland that cannot be ploughed or converted. These sensitive areas cover around 18% of permanent grassland, mainly (more than 95%) within Natura 2000 areas since only six MS have designated such sensitive areas outside Natura 2000 areas. This third environmental instrument (called the greening payment) has been heavily criticised for its low environmental efficiency. According to the European Court of Auditors (ECA), 'greening as currently implemented is unlikely to provide significant benefits for the environment and climate' with too weak requirements to significantly change practices and systems (ECA, 2017). In particular, the Court shows that the ratio of permanent grassland on total farmland has slightly increased from 28.6% in 2007-2014 to 30.1% in 2016. However, changes in the percentage value mask a decrease of 3 million ha in permanent grassland between the two periods (from 47 to 44 million ha), the ratio increase being linked to the more important decrease in the denominator (from 164 to 145 million ha). The Court adds that the permanent grassland criterion must be better targeted and 'should focus on parcels with a high carbon content already accumulated in the soil, many of which are likely to be located outside Natura 2000 areas' (ECA, 2017). In the CAP that will apply from January 2023 over the five-year period 2023-2027, greening will be suppressed and corresponding requirements included in cross-compliance renamed conditionality.

The green architecture of the 2023-2027 CAP

The green architecture of the 2023-2027 CAP is displayed in Figure 2, and compared with that of the current CAP (Figure 2). It will continue to combine mandatory and voluntary measures. Mandatory instruments correspond to conditionality that includes current cross-compliance and greening for

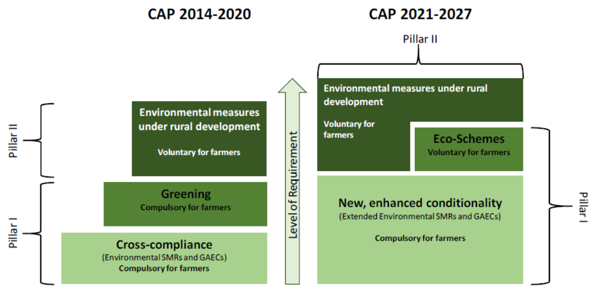

Figure 2. The green architecture of the current (2014-2020) and future (2021-2027) CAP (Lotz *et al.,* 2019).

globally unchanged constraint levels. Voluntary instruments include Pillar 2 AECS with only marginal changes with respect to today, and a new environmental instrument in Pillar 1 called eco-scheme.

Eco-schemes have several features in common with AECS. Both measures are targeted on the three climatic and environmental specific objectives of the CAP. Both are granted per hectare. Both are mandatory for countries but optional for beneficiaries. They differ by the fact that eco-schemes are an instrument of the first pillar and thus are fully funded by the European budget, while AECS are co-funded by national/regional authorities because they belong to the second pillar. Like AECS, eco-scheme aids could be granted in compensation for extra costs incurred or income foregone induced by the adoption of more environmentally friendly practices. However, although MS will very likely not use this possibility in an important way, eco-scheme aids could also be designed as fixed top-payments to basic income support aids. This alternative should be encouraged because it opens the door for the implementation of climatic and environmental payments more explicitly linked to the provision of corresponding services. Unfortunately, like conditionality and AECS, the first drafts of CAP National Strategic Plans (NSP) are not very ambitious from a climatic and environmental point of view (Runge *et al.,* 2002). In particular, according the EEB (European Environmental Bureau) and BirdLife International (2022), 'only two countries (Czech Republic and Finland) score well on [the grassland protection] dimension, indicating that most national CAP plans will lack strong enough measures and targets to protect and sustainably manage grasslands. This is notably the case for Austria, France, Hungary, Ireland, Lithuania, Portugal and Spain.'

How to support European grasslands thanks to the CAP?

Grasslands and conditionality

It is of considerable concern that the conditionality requirements of the next CAP will be hardly more demanding than cumulated commitments of cross-compliance and greening (Guyomard *et al.,* 2020).

Grasslands are targeted through GAEC #1 which aims to maintain permanent grassland areas on the basis of a ratio of permanent grassland on total agricultural area at national, regional, sub-regional or holding level, with the tolerance of -5% relative to a base period that can be either 2015 or 2018 (at the choice of the MS). GAEC #1 replaces the eponymous greening measure of the current CAP that has been heavily criticised (ECA, 2017). Grasslands are also targeted through GAEC #9, which prohibits the ploughing of permanent grassland in protected sensitive areas. To date, the latter were, with rare exceptions, restricted to permanent grassland areas in Natura 2000 zones. Preserving and increasing permanent grassland play a key role in the maintenance and creation of carbon sinks. Except in Natura 2000 areas, conditionality requirements will not prevent a permanent grassland area from being ploughed provided that an equivalent area is converted into permanent grassland, with associated negative impacts for both carbon storage and biodiversity since the older permanent grassland would be expected to have higher carbon storage and biomass diversity (Lotz *et al.,* 2019). We thus recommend strengthening GAEC #1 by an application at the level of the holding with a reduced tolerance of say -2% and using a reference period that is not updated (i.e. using 2015 as the reference period). For GAEC #9, each MS should have a stronger ambition. This could be done by starting from areas currently protected by this measure (following a no-backsliding principle) and by gradually increasing the surfaces over the period 2023-2027 on the basis of an action plan that will be defined, monitored and controlled in each NSP.

In addition, it is important that all farms and all farmland uses include non-productive areas and elements dedicated to biodiversity preservation (Guyomard *et al.,* 2020; Meredith and Kollenda, 2021). In the framework of the 2023-2017 CAP, this requirement is restricted to arable farms and lands though GAEC #8. The latter should be extended to all types of farms and farmland uses with adjustments of coefficients used to weight the different ecological focus areas in order to better coincide with biodiversity services they provide (Pe'er *et al.,* 2017).

Grasslands and incentives

Enhanced GAEC #1 would define the basis level below which the PPP would apply through conditionality, and above which the PGP would apply through an eco-scheme specific measure targeted on the remuneration of non-provisioning services provided by grasslands Ideally, the payment should be proportional to services that cover carbon storage, biodiversity protection, water regulation and quality, and soil quality. A simplified and immediately operational scheme would distinguish six types of grasslands corresponding to increasing levels of services; more specifically: (1) temporary grassland without legumes, (2) temporary grassland with legumes, (3) permanent grassland between 5 and 10 years without legumes, (4) permanent grassland between 5 and 10 years with legumes, (5) permanent grassland above 10 years without legumes, and (6) permanent grassland above 10 years with legumes (Figure 3). Three payment levels will be proposed (blue bars) with a bonus (orange bars) for legumes.

The y-axis of Figure 3 corresponds to basic payment levels and bonuses for legumes, but is not detailed in so far as where it depends on budgetary constraints. In addition, the scheme should take into account the fact that it is likely to be more important to increase the climatic and environmental quality of grasslands where the latter is threatened by grassland intensification and/or crop conversion, than to support the

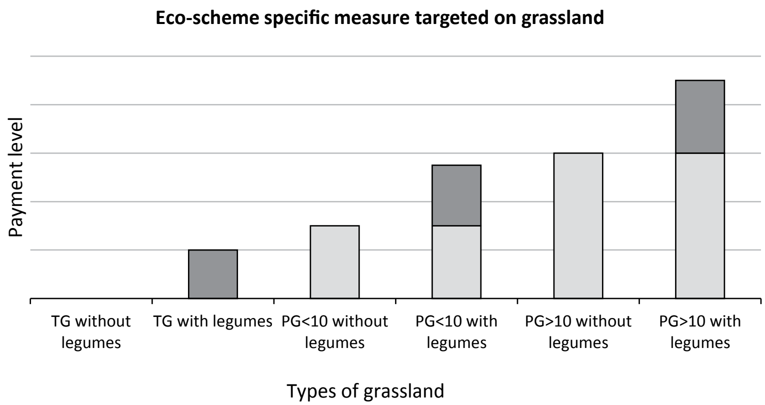

Figure 3. Definition of an ecoscheme specific measure targeted on the supply of non-provisioning services by different types of grasslands.

incomes of livestock producers relying on extensive management of grasslands in regions where there are no or only very few alternatives to permanent grassland. The income support objective must be targeted by CAP income support instruments, notably CAP payments for Less Favoured Areas (LFA) that were introduced in the 1970s. Although the main objective of LFA payments is to offset additional costs and/or lower incomes in these areas, they can also offer environmental benefits by limiting farmland abandonment and maintaining an agricultural activity beneficial for the environment thanks to natural constraints enforcing extensive management practices and systems, notably permanent grassland-based systems.

The climatic and environmental urgency implies there is a need for better protection of grasslands and for increased supply of the non-provisioning services they can provide. It is urgent to do so as quickly as possible. Coupled with enhanced conditionality, the grassland eco-scheme measure proposed above aims to respond to this urgency. Demonstrators whose objectives would be to enable better quantification of the non-provisioning services that the different types of grassland provide should complete the measure. This would make it possible to adjust the proposed grassland eco-scheme measure progressively (types of grassland that should be taken into account, likely by differentiating them regionally, payment levels). This would also make it possible to generalize payments for providing climatic and environmental outcomes based on an obligation of results (impacts). This opens the door for such payments to be funded not only by the taxpayer through the CAP, but also by the intermediate and final user through bilateral contracts or ecological service markets.

Concluding remarks: How to manage trade-offs? Is grassland protection in line with the Green Deal ambition and objectives?

Any CAP reform that would be (more) ambitious from a climatic and environmental point of view should explicitly address the potential trade-offs that such an ambition could entail, notably a potential trade-off between climatic and environmental objectives and economic impacts. This trade-off is too often used as a pretext for not moving (the *status quo*) while many proponents of a strong climatic and environmental CAP ignore the economic dimension of sustainability. Many action levers could be used to alleviate the trade-off by playing on the length of the transition period, by exploiting all productivity gain sources including precision farming and genetics, by increasing vertical (along food chains) and horizontal (among actors within territories) solidarities, etc. In addition, new sources of incomes based on the development of results-based payments for climatic and environmental services and the use of pollution and health savings that would be generated by more environmentally-friendly and healthier agricultural and food systems are promising avenues to explore. Pilot demonstrators defined and implemented in various contexts, with clear ecological targets, validated result-based indicators and dedicated budgets, are a promising avenue for progress, allowing for learning, notably by experimenting and doing.

Are our recommendations for grassland protection and the remuneration of grassland services compatible with the European GD? The latter sets ambitious quantitative targets implying significant reductions in the use of pesticides (-50% by 2030), fertilizers (-20%) and antibiotics (-50%), and large increases in agricultural land under organic farming (25%) and in high-diversity landscape features (10%). European agriculture is not on the right track to meet these targets, and the 2023-2027 CAP will very likely be insufficient to reverse unfavourable trends (Guyomard *et al.,* 2020). Unlike the CAP, the GD is not restricted to farm aspects. It rightfully adopts a whole food-chain approach by pointing out that an increasing proportion of the European population does not comply with dietary recommendations. Policies that are much more ambitious are needed in this area with health/nutrition and ecological benefits in the centre. This means that the GD ambition could not been achieved without both supply and demand side policies. Many policy instruments relying on education, information campaigns, food labels and fiscal tools can be used to change inadequate consumption patterns (Guyomard *et al.,* 2021). These demand side measures will aim to reduce the excessive consumption of animal products, including red meat and dairy products. This means that they will try to take into account public costs associated with inadequate food patterns (penalization of negative externalities), in accordance with supply measures we propose for the remuneration of the non-provisioning services that grasslands may provide (recognition of positive externalities).

At the time we were writing this paper (March 2022), the war in Ukraine was accelerating changes that we could not have foreseen just some weeks previously. Against that dramatic framework, one may fear that environmental and social objectives, budgets and instruments take a back seat. However, 'it is only backing up to better change'. The resilience of European food systems requires urgent and important changes, including by addressing their direct and indirect (such as fertilizer cost) dependency on fossil resources. In that perspective, grassland-based systems are also an opportunity that justifies public support.

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References

Animal Task Force (ATF) (2017) Why is European animal production important today? ATF, Facts and Figures, Brussels, Belgium, 5 p.

Azeda C., Guiomar N., Medeiros J.P., Godinho S. and Pinto-Corretia T. (2021) The ambiguous role of agri-environment-climate measures in the safeguarding of High Nature Value Farming Systems. The case of the Montado in Portugal. *Agriculture, Ecosystems and the Environment* 319, 107562.

Baltic Environmental Forum (2017) How do grasslands benefit humans: Introduction to grassland ecosystem services. LIFE+ Environment Policy and Governance project 'Integrated planning tool to ensure viability of grasslands' (LIFE Viva Grass, project No. LIFE13 ENV/LT/000189).

Blüthgen N., Dormann C.F., Prati D., ... and Weisser W.W. (2012) A quantitative index of land-use intensity in grasslands: Integrating mowing, grazing and fertilization. *Basic and Applied Ecology* 13, 207-220.

Buckwell A. and Nadeu E. (2018) What is the safe operating space for EU livestock? Report, RISE Foundation, Brussels, Belgium, 108 p.

Bugalho M., Pinto-Correia T. and Pulido F. (2018) Human use of natural capital generates cultural and other ecosystem services in *montado* and *dehesa* oak woodlands. In Paracchini M.L., Zingari P.C. and Blasi C. (eds.), Re-connecting natural and cultural capital, contributions from science and policy. Publications Office of the European Union, Luxembourg, Luxembourg.

Chatellier V. and Guyomard H. (2022) A brief story of the CAP. In: Détang-Dessendre C, and Guyomard H. (eds.), What Common Agricultural Policy tomorrow? Editions Quae, Versailles, France.

Cullen P., Dupraz P., Moran J., Murphy P., O'Flaherty R., O'Donoghue C., O'Shea R. and Ryan M. (2018) Agri-environment scheme design: past lessons and future suggestions. *EuroChoices* 17, 26-30.

Dupraz P. and Guyomard H. (2019) Environment and climate in the Common Agricultural Policy (CAP). *EuroChoices* 18, 18-25.

European Environmental Bureau (EEB) and BirdLife International (2022) CAP Strategic Plans – are they likely to deliver on given promises?

European Commission (EC) (2004) Commission Regulation (EC) N° 796/2004. Brussels, Belgium.

European Commission (EC) (2013) Regulation (EU) N° 1307/2013. Brussels, Belgium.

European Commission (EC) (2019) Communication from the Commission to the European Parliament, the European Council, the Council, the European Economic and Social Committee and the Committee of the Regions. The European Green Deal. COM (2019) 640 final, 24 p. + Annex.

European Commission (EC) (2021) Land cover statistics. Brussels, Belgium. Available at: https://ec.europa.eu/eurostat/statistics-explained/index.php?title=Land_cover_statistics

European Court of Auditors (ECA) (2017) Greening: a more complex income support scheme, not yet environmentally effective. Special Report 21/2017, 63 p.

European Environment Agency (EEA) (2020b) EEA greenhouse gas – data viewer. Data viewer on greenhouse gas emissions and removals, sent by countries to UNFCCC and the EU Greenhouse Gas Monitoring Mechanism (EU Member States). Last modified 02 June 2020.

Estel S., Mader S., Levers C., Verburg P.H., Baumann M. and Kuemmerle T. (2018) Combining satellite data and agricultural statistics to map grassland management intensity in Europe. *Environmental Research Letters* 13, 074020.

Floate K.D., Wardhaugh K.G., Boxall A.B.A., Sherratt T.N. (2005) Fecal residues of veterinary parasiticides: Non target effects in the pasture environment. *Annual Review of Entomology* 50, 153-179.

Guyomard H., Bureau J.C., Chatellier V., Détang-Dessendre C., Dupraz P., Jacquet F., Reboud X., Réquillart V., Soler L.G. and Tysebaert M. (2020) The Green Deal and the CAP: policy implications to adapt farming practices and to preserve the EU's natural resources. Research for AGRI Committee, European Parliament, Policy Department for Structural and Cohesion Policies, Brussels, 211 p. (+ annexes).

Guyomard H., Bouamra-Mechemache Z., Chatellier V., Delaby L., Détang-Dessendre C., Peyraud J.-L. and Réquillart V. (2021) Review: Why and how to regulate animal production and consumption: The case of the European Union. *Animal* 15(1), 100283.

Herrero-Jáuregui C. and Oesterheld M. (2018) Effects of grazing intensity on plant richness and diversity: a meta-analysis. *Oikos* 127, 757-766.

Herzon I., Birge T., Allen B., Povellato A., Vanni F., Hart K., Radley G., Tucker G., Keenleyside C., Oppermann R., Underwood E., Poux X., Beaufoy G. and Pra J. (2018) Time to look for evidence: Results-based approach to biodiversity conservation on farmland in Europe. *Land Use Policy* 71, 347-354.

Lotz B., Monschauer Y. and Schäfer M. (2019) Climate-friendly design of the EU Common Agricultural Policy. Navigant Discussion Paper (final version) prepared for the Federal Ministry for the Environment, Nature Conservation and Nuclear safety (BMU), Berlin, 35 p.

Meredith S. and Kollenda E. (2021) What green ambition of the CAP reform can still be salvaged? Briefing note on the Common Agricultural Policy trilogue negotiations, IEEP, Brussels, Belgium.

Mosquera-Losada M.R., Santiogo-Freijnanes J.J., Ferreiro-Dominguez N, Rofrigue-Rigueiro J. and Rigueiro-Rofriguez A. (2019) Report with the maps of the different permanent grassland. EU project GO-GRASS, Deliverable 1.1.

Mottet A., De Haan C., Falcucci A., Tempio G., Opio C. and Gerber P.J. (2017) Livestock: On our plates or eating at our table? A new analysis of the feed/food debate. *Global Food Security* 14, 1-8.

Olff H., Ritchie M.E. (1998) Effects of herbivores on grassland plant diversity. *Trends in Ecology & Evolution* 13, 261-265.

Pe'er G., Zinngrebe Y., Hauck J., Schindler S., Dittrich A., Zingg S., Tscharntke T., Oppermann R., Sutcliffe L.M.E., Sirami C., Schmidt J., Hoyer C., Schleyer C. and Lakner S. (2016) Adding some green to the greening: improving the EU's Ecological Focus Areas for biodiversity and farmers. *Conservation Letters* 10(5), 12333. https://doi.org/10.1111/conl.12333

Peyraud J.L., Peeters A. and De Vliegher A. (2012) Place et atouts des prairies permanentes en France et en Europe. *Fourrages* 211, 195-204.

Pieper M., Michalke A. and Gaugler T. (2020) Calculation of external climate costs for food highlights inadequate pricing of animal products. *Nature Communications* 11, 1-13.

Pinto-Correia T. and Azeda C. (2017) Public policies creating tensions in Montado management models: insights from farmers' representations. *Land Use Policy* 64, 76-82.

Pinto-Correia T., Guiomar N., Ferraz-de-Oliveira M.I., Sales-Baptista E., Rabaça J., Godinho C., Ribeiro N., Sá Sousa P., Santos P., Santos-Silva C., Simões M.P., Belo A., Catarino L., P. Costa P., Fonseca E., Godinho S., Azeda C., Almeida M., Gomes L., Lopes de Castro J., Louro R., Silvestre M. and Vaz M. (2018). Progress in identifying High Nature Value Montados: relating biodiversity to grazing and stock management. *Rangeland Ecology and Management* 71, 612-625.

Runge T., Latacz-Lohmann U., Schaller L., Todorova K., Daugbjerg C., Termansen M., Liira J., Le Gloux F., Dupraz P., Leppanen J., Fogorasi J., Zita V.E., Berzina I., Schulp C., Bradfield T., Hennessy T., Targetti S., Viaggi S., Majewski E., Pecurul M., Prokofieva I., Velazquez J.B., Bouriaud L. and Baciu F. (2022). Implementation of eco-schemes in fifteen European Union Member States. *EuroChoices* (in press).

Takagi S. and Miyashita T. (2014) Scale and system dependencies of indirect effects of large herbivores on phytophagous insects: A meta-analysis. *Population Ecology* 56(3), 435-445.

Uthes S. and Matzdorf B. (2013) Studies on agri-environmental measures: A survey of the literature. *Environmental Management* 51(1), 251-266.

Van Klink R., Schrama M., Nolte S., Bakker J.P., WallisDeVries M.F. and Berg M.P. (2015) Defoliation and soil compaction jointly drive large-herbivore grazing effects on plants and soil arthropods on clay soil. *Ecosystems* 18, 671-685.

Permanent grassland and ruminants are a key component of the agroecological transition in Europe – findings from the 'Ten Years For Agroecology' scenario

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Abstract

While the importance of permanent grasslands (PG) for the conservation of high nature value habitats is often acknowledged, their role in most of the main sustainable food system scenarios published in the last few years is not made explicit. In the best case, the place of permanent grassland in scenarios and the policy agenda is their conservation; in the worst-case scenario, they are simply replaced by afforested land or cropland, considered as a better option when focusing on greenhouse gas (GHG) emissions only. In this paper, we defend the idea that several misinterpretations of the positive and negative impacts of ruminants, the 'natural' users of PG, and explain why these latter are poorly addressed by most scenarios. Based on the findings of an agroecological scenario for Europe – TYFA, standing for Ten Years For Agroecology – in which extensive permanent grassland and ruminants play a prominent role for biodiversity conservation, nitrogen cycle and climate change mitigation and adaptation altogether, we call for a research agenda that would better inform the specific role of PG in the provision of ecosystem services – and in particular those depending on nitrogen management.

Keywords: permanent grassland, scenario, agroecology, biodiversity, climate change, livestock, nitrogen

Introduction

Here we defined permanent grassland (PG) as herbaceous and non-herbaceous (ligneous) vegetation used for grazing and/or mowing, provided that it has not been ploughed for 5 years or more. It thus includes rangeland, such as garrigue, moorland, etc. This definition is consistent with the most recent one in the Common Agricultural Policy. In Europe, these PG have an ambivalent position in the agri-environmental debate. On the one hand, they are acknowledged as paramount for natural resource conservation. Their role in sustainable water resource management – and, reciprocally, the impact of their subsequent ploughing in releasing important quantities of nitrates and carbon – is well known (Strebel *et al.*, 1989). In addition, when extensively managed, with low stocking rates, their irreplaceable role for biodiversity conservation in High Nature Value systems is also well understood (Veen *et al.*, 2009). The list of services they provide puts PG at the centre of many reflections on multifunctional agriculture in Europe (Ryschawy *et al.*, 2017; Schils *et al.*, 2022). These attributes also explain why they are protected through the CAP regulation for instance.

On the other hand, while the importance of permanent grassland is recognized, their 'natural' managers, namely ruminants, are subject to strong criticism. They are blamed for several reasons, ranging from high demand for water (through the amount of water needed to produce 1 kg of beef meat) to inefficient use of feed and thus of land, compared with other grain-fed livestock, poultry and pigs (Herrero *et al.*, 2013). But the strongest case against herbivores probably concerns their methane emissions (Steinfeld *et al.*, 2006). In short, permanent grasslands are praised for their biodiversity and the ecosystem services they provide, most visible at a local scale, but ruminants fed by them are blamed for their impact on climate change, generally when considered at a global scale (Garnett *et al.*, 2017). A recent report issued from a joint workshop between IPBES and IPCC (Pörtner *et al.*, 2021) put it in a similar way: PG needs to be conserved and not converted to produce bioenergy; but beef and dairy consumption should be reduced to a minimum. How PG will then be managed remains an open question in such a perspective.

This paper takes a broader perspective to further explore this tension and explore in particular the role that PG and ruminants could play in the functioning of sustainable food systems. Our analysis is based on a review of recent sustainable food system scenarios published in the EU over the last five years (Duru *et al.*, 2021), and more particularly the TYFA scenario (Poux and Aubert, 2018). In the first part, we present a review of how existing scenarios address the role of PG and ruminants and point out several limits. The second part presents the conceptual framework underpinning the TYFA scenarios along with its key results. Overall, we show under which conditions the apparent contradictions between PG (desirable) and ruminants (undesirable) can be overcome. The conclusion identifies key science and policy areas to further investigate for the design of fully sustainable food and agricultural systems, in which PG and ruminants can play a determining role in the European context.

The role of PG in food system scenarios

An agenda dominated by climate change mitigation

In the EU, the agriculture-environment debate has become dominated by climate issues, even though biodiversity conservation, and the role PG could play therein (Simoncini *et al.*, 2019), – is not at all absent. As such, climate mitigation is, by large and far, considered as the top priority of most sustainable food system scenarios published recently. On the contrary, biodiversity is considered as a 'bonus' that is addressed only by a limited number of scenarios adopting a multifunctional perspective (Duru *et al.*, 2021).

The analysis of this set of scenarios has been extensively explained (Duru *et al.*, 2021). It shows that to reach their objectives, the vast majority of the scenarios rely on (a) a reduction of livestock – including ruminants; (b) carbon sequestration through afforestation; (c) important land use changes through simultaneous increases in yields and afforestation.

Except TYFA, which will be presented further in this paper, and Future Nordic Diet, the other scenarios consider permanent grassland mostly as potential land to be taken for afforestation. Assumptions range from nearly all PG being afforested (with a net C gain) to some PG being conserved for biodiversity reasons – but with no key role in food production. This well illustrates the tensions between the need to conserve PG; and the objective of reducing greatly the ruminant herd. In some case, ruminants are also replaced by biogas in an expected win-win-win prospect (grassland + bioenergy + methane emission reduction).

The limits of the prevailing approaches with regards to permanent grassland

A first type of scenario attributes a very limited role to PG, through to no role at all (#1, 2, 4, 5, 8, 9, 10, 11 and 12 in Table 1), for two major reasons. The first reason is that ruminants are harmful for the climate and for human health and can be easily substituted by other, more climate efficient, sources of food. The second reason is that despite their potential biodiversity interest, permanent grassland afforestation is considered as a no-regret option as it could store more carbon, provide renewable fuel (although there is a contradiction in terms between storing carbon and using it in renewable energy) and support biodiversity conservation, assuming that forested areas are also good for biodiversity by nature, as distinct to that role as provided by permanent grassland. This framing leads to a strong reduction of both ruminants and permanent grassland – or even their total disappearance – even though such extreme assumptions are not necessarily spelled out.

Table 1. Scenarios analysed in (Duru *et al.*, 2021) – Note that TYFA has been issued by the authors of the present paper and will be presented further.

#	Name of the scenario	Year of issue	Level of analysis	Authors/sponsor(s)
1	Achieving net zero farming's 2040 goal	2019	United Kingdom	National Farmers Union
2	Climate neutrality in 2050	2017	Denmark	Danish Food and Agricultural Council
3	Future Nordic Diet	2017	Denmark, Sweden, Norway, Finland	Karlsson *et al.*
4	Achieving net zero	2020		Haut conseil pour le climat
5	Pathways to sustainable land-use and food systems	2019	17 territories of the EU	FABLE Coalition/IIASA
6	Scenarios for an ecological transition for Walloon farming)	2019	Wallonia	Université Catholique de Louvain
7	TYFA	2018 / 2019	European Union	IDDRI, AScA
8	Net Zero emissions in agriculture	2019	European Union	IEEP/ECF
9	Long term strategy for Europe	2018	European Union	IIASA (Globiom)
10	Vision 2050	2014	France	ADEME
11	Global Warming of 1.5° IPCC Special report	2018	World	IPCC (GIEC)
12	Afterres	2011/2016	France	SOLAGRO

A second type of scenarios/models (#3, 6, 7 and 12) better recognizes the role of PG for biodiversity, when extensively managed, i.e. (a) in the absence of mineral nitrogen fertilizer and (b) when stocking densities are adapted to the natural primary productivity. Ruminants are also acknowledged as providers of milk and meat, with a genuine important nutritional role in ensuring adequate calcium intake. The issue is to find the right balance between their positive role and the need to limit their place in the food system, both in diet and in methane emissions. Modelling assumptions help to set the maximum number of hectares and livestock head to conserve an acceptable envelope of permanent grassland for landscape and biodiversity while minimizing the share of ruminant-based food in the diet, for the sake of reducing methane emissions.

A first conceptual limit in the first type of scenarios described is that they overlook the role of extensive permanent grassland in biodiversity conservation, especially in the European context. When they intend to care for biodiversity, they implicitly assume that a land use change from grassland to forest can bring another type of biodiversity, different in nature from the one present in PG, but comparable in value. While in some places, moving from intensively managed PG to forest will indeed improve the biodiversity, this vision does not consider the fact that biodiversity value stands on the diversity of types of habitats. It also ignores the long term processes that have made PG so important for biodiversity conservation in Europe (Pärtel *et al.*, 2005), and the fact that just under 30% of all habitats the EU has set to conserve as per the Biodiversity Convention are indeed dependent upon extensive livestock systems, and thus PG (Halada *et al.*, 2011). In short, replacing most high nature value permanent grasslands with forests, even forest with biodiversity interest, would decrease Europe's overall biodiversity. More fundamentally, this interpretation misses the fact that extensive PG also contribute to maintain biodiversity within agroecosystems and thus provide fundamental ecosystem services, amongst which are pollination and pest control (Dainese *et al.*, 2019). Last but not least, such a vision does not consider the important cultural dimensions dealing with open landscapes, including well-being in open landscapes, and the heritage associated with high quality products obtained from animals fed on extensive PG.

A second conceptual limit regarding how PG are considered lies with their role in nutrient cycles – both N and C – and hence their role in climate change mitigation. In most scenarios, the claim for limitation of the share of PG and ruminants in the food system rests on the following assumptions: that ruminants emit more methane than the CO_2 equivalent that can be stored in PG, whereas forests have a net positive carbon balance (Garnett *et al.*, 2017). This simplistic approach has at least two limitations:

- Most models do not account for the nitrogen (N) supply from permanent grassland while Eurostat estimates that PG supply 2/3 of the overall proteins consumed by EU livestock (Eurostat, 2021). Yet PG, when legumes are abundant, provides symbiotic nitrogen and the impact on the environment is much lower than nitrogen from synthetic fertilizers; we therefore consider PG to be fundamental N suppliers and that extensive mixed ruminant systems to be unique conveyors of such organic N to arable land – the impact of which being, in temperate areas, considerably less important than that of mineral fertilizers (Buendia *et al.*, 2019) (see below). As such, most models tend to give a distorted image of the impact of methane emissions against that of N_2O (Lynch *et al.*, 2021).
- With regards to methane, most reasoning assimilates the impact on climate to the annual emission level alone, calculated through the use of an 'equivalent CO_2' impact over a period, generally 100 years. This simplification does not consider the short life of methane, the warming impact of which is very strong but the effect does not last (a 'flash' effect), due to the fact that half of the molecules decay into CO_2 + H_2O after a period of 12 years. When the methane comes from a biogenic cycle (i.e. a present cycle, as opposed to the release of fossil methane), which is the case in agriculture, the decay of CH_4 into CO_2 at the end of the cycle is 'compensated' by the initial fixed CO_2 in the cycle through photosynthesis. This peculiarity completely alters the understanding of the impact of methane emissions on climate change: a stable level of emissions of biogenic methane from a sector does not increase global temperature (livestock in our case, but the same applies to irrigated crops, such as rice). Reciprocally, an increase or a decrease in emissions will respectively lead to higher temperature (increase the powerful flash effect of CH_4) or, when decreasing, to a so-called 'cooling effect' (Allen *et al.*, 2018b; Lynch *et al.*, 2021). Finer analysis taking into account the heating power of the flash effect of methane on oceans results in a neutral impact of methane emissions being obtained for changes of -10 to -15% in 12 years (Allen *et al.*, 2018a).

This rapid overview of existing scenarios shows that the links between the management of the carbon cycle (and the understanding of its impact on climate change), the nitrogen cycle, yields and overall land use change should be better analysed together. In particular, any gains arising from greatly reducing the numbers of ruminants should be set against the nitrogen budget that they contribute to: when it comes to replacing synthetic N by organic N from manure and the subsequent carry-over effect, there is a balance with methane emissions to better factor in. This is what we propose to do in the next section.

TYFA: another outlook on biodiversity, livestock and permanent grassland

The conceptual basis of TYFA

Against this backdrop, the modelling underpinning the TYFA scenario (Poux and Aubert, 2018) was developed to better take into account biodiversity conservation issues *within* agricultural landscapes, along with climate change. As such, it puts the emphasis on two central dimension of agroecosystem management: the absolute level of synthetic inputs used (e.g. Geiger *et al.*, 2010), and the level of landscape heterogeneity (e.g. Fahrig *et al.*, 2011). Within this perspective, PG and ruminants are given specific attention for their role in both N/C cycles and landscape structuration. This approach is rooted particularly in the High Nature Value (HNV) conceptual framework (Lomba *et al.*, 2014; Strohbach *et al.*, 2015), whose contribution to a better understanding of the role of extensive livestock systems and semi-natural vegetation as the backbone of biodiversity in European open landscapes has been instrumental.

To explore how to resolve tensions between biodiversity and natural resources conservation, and climate change mitigation, the TFYA scenario rests on a biomass-balance model (called TYFAm hereafter) organized around five compartments between which material and energy flow, and which are connected systemically (Figure 1):

1. Crop production, resulting from a certain European land use (distributed between arable land, permanent crops, permanent grasslands and agro-ecological infrastructures: hedges, trees, ponds, stony habitats, sunken paths) and the associated yields.
2. Livestock production, fed by a fraction of crop production, some of which may compete with human food (for example cereals), while the rest does not (grasslands and co-products).
3. Demand for food, which is the result of individual eating habits and a given level of population growth in Europe, and is covered by both European production and imported products.
4. Non-food/industrial demand for biomass (energy and biomaterials), which can once again be covered by a mix of European production and imports.
5. Finally, the nitrogen flows associated with the functioning of and interactions between the first four compartments largely determine the level of soil fertility. The analysis of N flows takes into account the different types of inputs (synthetic nitrogen, animal feed imports, symbiotic fixation, transfers by manure) and exports (livestock and crop production).

For each compartment, the TFYA scenario proposes detailed and quantitative assumptions (on yield, crop rotation, input-output ratio, human diets...). This set of assumptions is outlined below (Figure 2 and detailed in Poux and Aubert, 2018). It aims at addressing the following key questions: within the 'European farm', what level of production is compatible with the multiple objectives of biodiversity conservation and climate mitigation? Is this level of production sufficient to feed Europeans or to generate a surplus, and under which conditions in terms of their diets?

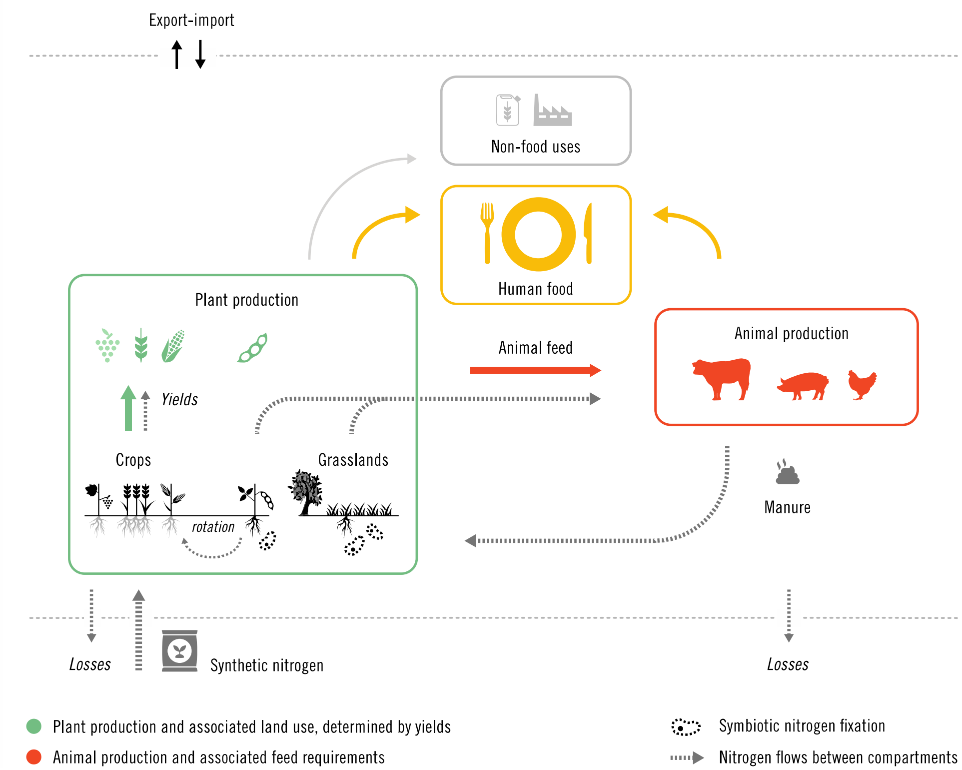


Figure 1. Logical structure of the model underpinning the TFYA scenario.

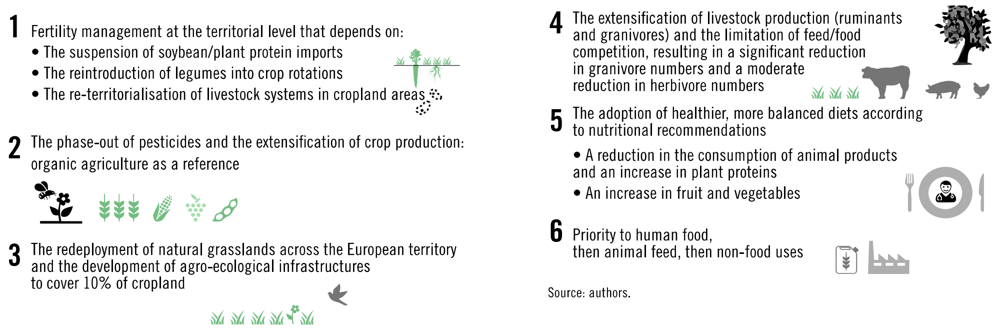


Figure 2. Key assumptions of the TYFA scenario.

The role of PG in TYFA: a key component for multifunctionality

PG play a central role in the overall TYFA scenario for both food production and the provision of ecosystem services, under the assumption that they are extensively managed (see above). These conditions are central for two reasons: the first is that mineral fertilization of PG is negatively correlated with the number of flora and thus fauna species. This is all the more important as extensive PG are statistically the most significant land use category under semi-natural vegetation, the importance of which for ecosystem services provision in agro-systems is well acknowledged (Garibaldi *et al.*, 2020). The second reason is that the absence of mineral N fertilization fosters spontaneous N fixation through free and symbiotic bacteria in PG ecosystems, thus maximizing the natural inflow of this element in a large share of agro-ecosystems.

The capacity of PG to provide food and ecosystem services is determined by the balance between: (a) the maximum amount of N such extensive PG can fix through symbiotic fixation – with an upper limit determined by the share of legumes in the flora community and the overall yield, and (b) the subsequent net N export from ruminants in the form of milk and meat, and mostly as manure.

Our calculations show that this balance is tight. TYFA modelling assumes a net export of around 90-100 kg N ha^{-1} PG $year^{-1}$, based on PG yields of, on average at the EU level, 5 tons of dry matter ha^{-1} and a 30% share of legumes in PG (following empirical results and a meta-analysis, presented respectively in Bignal (2000) and Smit *et al.* (2008)). On the other hand, current models suggest an N supply from symbiotic fixation of around 70-80 kg ha^{-1} (see below our assumption of net export). However, such models are statistical and calibrated on data covering a wide variety of grasslands, amongst which some are fertilized (as shown in Einarsson *et al.*, 2021) and thus do not reflect the specificity of truly semi-natural grassland. More generally, analysis of N fluxes in extensive systems has been understudied and quantified and the apparent shortage in N supply from symbiotic fixation can be covered by N aerial deposition and fixation by free living bacteria (Roper and Gupta, 2016). All in all, there is a need for further research in this field, on most occasions extensive PG are conceptually considered as grasslands with a 0 value on a gradient ranging from 0 to say 180 uN ha^{-1} or more. We defend the idea that a low-input PG ecosystem develops an N-fixation mechanism that is functionally different from a PG with a high mineral-N legacy or input.

This analysis based on the N cycle helps to make more explicit the key ecosystem services that PG provide in the TFYA scenario in addition to their existence value for biodiversity conservation purposes. At the landscape level, low-input management of PG will go along with the provision of non-polluted water due to the absence of pesticide use and the highly limited risks of N runoff under low-fertilized PG.

At the level of the whole agroecosystem, considered as a combination of PG and semi-natural components with arable, typical of mixed systems, N provision under an organic form (through N fixation and its subsequent transfer to arable land in the form of animal manure) has a double benefit. Because the release of mineral N (through mineralization of organic N) is slow and scarce, it first respects the soil ecosystem functioning, provided adapted tillage (e.g. superficial ploughing or no-ploughing when possible), while mineral-N fertilization will alter this latter. This, together with the assumption of pesticide-free cropping system, in turn allows the conservation of soil life and avoids inhibiting nitrogen fixation in cropping systems (Fox *et al.*, 2007), thus enhancing this function in rotations.

Another benefit of organic N fertilization is the reduction of volatilization, in the temperate context, as mentioned above (IPCC, 2019). The 2.6 times less N_2O emitted from organic fertilizers compared with that from mineral fertilizers is highly significant for the primary GHG emitted by agriculture as a whole. Note that underground N takeover after a legume in a crop rotation is considered to have hardly no climate impact.

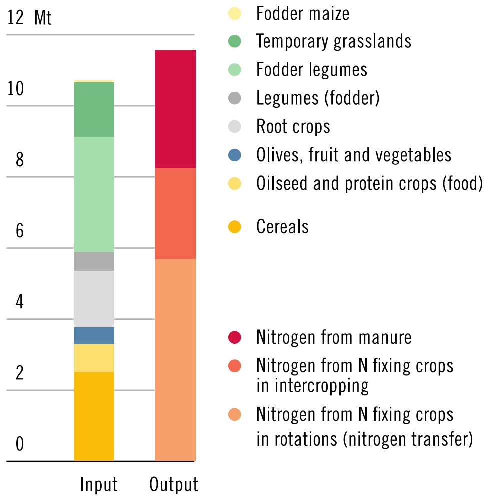
All in all, the climate performance of TYFA relies on three levers: (1) the reduction of the ruminant herd (dairy cows, cattle and small ruminants) by 18%; (2) an overall reduction in N application (from over 20 Mt of N under the form of synthetic N and manure in 2010 to 3.3 Mt or organic N in the form of manure by 2050) due to the extensification of vegetal production and a greater Nitrogen use efficiency, and (3) the integral shift from mineral fertilizers to organic N fertilizers.

The production constraint through the N reading

Despite the negative impacts of mineral N, one cannot envisage a (too) low level of this element in absolute terms. In the context of temperate agriculture, this element is the main limiting factor in terms of plant nutrition when water availability is not the limitation. Thus, a balance must be found between minimizing the environmental impacts, or even better providing a bulk of positive environmental services on the one hand, and providing enough food for the population on the other hand. The challenge is to maximize the naturally fixed N flows entering the cropping system. In this perspective, there are two major sources: the flows inside the cropping systems, through legumes in rotation – including N-fixing cover-crops – and the flows from outside, namely through fertility transfers from semi-vegetation areas; essentially PG when it comes to nitrogen. In modelling N flows at the EU scale, we found that the share of N that could inflow from legumes in cropping systems was not sufficient to cover the needs calculated for the provision of a sustainable diet (see next section). Indeed, there are three limits in the share of legumes in the cropping systems: the needs for food supply (we do not grow pulses/legumes above that needed for human and animal consumption), the possibility to set N-fixing cover-crops, depending on the share of spring crops (no such cover-crops between two winter crops), and an agronomic limit, considering that above 25-30% of legumes in a rotation would entail pathologies (fungal attack).

TYFA N balance (Figure 3) shows that N-supply from N-fixing crops in rotation (including intercropping and temporary grasslands) can cover up to 70% of crop needs. This result is indirectly confirmed by Barbieri *et al.* (2021). While they indeed demonstrate that a fully organic agricultural system would result in a N shortage of around 40% at the global level, they did not consider any supply of N from PG, as illustrated in Figure 4.

The remaining 30% of N-needs are covered by manure (see Figure 3). Of this, 20% comes from monogastric and 80% from ruminant systems – assuming a ratio of 60% of N in manure from ruminants transferable to crops. The net N transfer of PG to cropland finally depends on the share of grass from PGs in the overall feed ration. On that matter, TYFA's assumptions are that, on average, PGs provide 70% of the feed needed (in dry matter) for all ruminants (dairy systems, cattle, small ruminants). This means a net N transfer from PG to cropland of just under 20% of N-crop needs.



Source: TYFAm.

Figure 3. TYFA nitrogen balance.

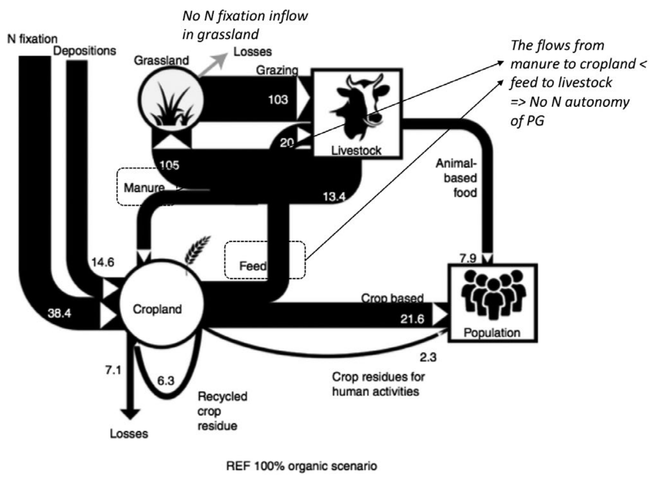


Figure 4. N flows representation in (Barbieri, 2021), amounts in TgN. Italics, arrows and dashed box are our addition. In this figure there is no N fixation accounted for in PG, and thus no transfer to cropland, unlike TYFA.

As such, PG play a key role in closing the organic N cycle, with cascading positive consequences on production, biodiversity, natural resources management and, as developed further, climate. This bulk of key services largely stands on the sustainable N cycle induced by the complementarity between PG and arable, which must then be taken together. A 'function' of arable is thus to value the exported N from PG and – by the way – allow a maximum biodiversity in these latter. Here, it should be noted that this approach implies a redistribution of PG at the EU level in order to generalize mixed livestock-cropping systems. This means a 'de-specialization' in areas today dominated by PG but where crop production is possible and had existed recently (typically in hill areas) on the one hand, and reintroduction of PG in areas specialized in crop production today on the other hand. Many areas would remain under the main use of PG (i.e. mountains, wetlands), but they are statistically of minor importance. All in all, the scenario relies on a strong assumption that would deserve further exploration: that the area of PG replaced by crops (in today's grassy areas) equals the one of crops replaced by PG, with a net compensation of the C and N released. This geographical shift implies more productive PG and less productive crops at the end.

In this ecological functioning, ruminants are also important as they provide the service of N transfer, if one can label it this way. In TYFA as compared with other scenarios, ruminants have three key characteristics that make them genuinely interesting: (1) they transfer N without use of machinery, and thus of fuel, (2) they provide organic N, the value of which has been extensively described previously, and (3) they provide food produced from vegetation that is non-edible for humans.

The dietary dimension of PG and ruminants

The need to significantly reduce the intake of proteins from animal origin in OECD countries, in order to address climate change mitigation at global level, is a widely acknowledged result (Clark *et al.*, 2020). The dietary discussion has thus become a key issue in scenarios dealing with this matter, and TYFA makes no exception about this and assumes halving the intake of meat and dairy products. The issue then is the best balance between ruminants and granivores in the meat supply.

Two main analyses can be found in this domain. The first fundamentally considers that ruminants combine two major drawbacks: (1) they are the less efficient way of producing food from a climate perspective (as measured in terms of CO_2 eq emissions/kcal), (2) and they can be replaced by other food in the human diet. The EAT-Lancet report (Willett *et al.*, 2019) can be identified as a flagship paper in this stream, aligned with a series of climate-friendly scenarios based on the minimization of food produced by ruminants. In this framing, there is no argument for producing any calorie of meat in general and even less from ruminants, and the best diet for climate and for most other indicators is the vegan one.

TYFA takes place in another outlook on the analysis of sustainability of the food systems. It positions itself in the stream of work considering that one major advantage of ruminants is their ability to feed on non-edible vegetation, in other terms the PG at the core of this paper (as proposed in Van Zanten *et al.* (2016) and Van Zanten *et al.* (2018)). In this perspective, while all agricultural land is not suitable for arable production, meat and dairy food produced from PG is not an inefficient land use but, on the contrary, represents the best one for non-arable land (see also Van Kernebeek *et al.*, 2016). In a wider perspective, this complementary source of calories reduces the need to produce food on the scarce area of arable land and thus reduces the need to intensify on this area. Indeed, when taking into account the fundamental difference between feeding animals with grains vs non-edible feed, ruminants become the most efficient land users, provided that they indeed are fed on PG (Mottet *et al.*, 2017; Wilkinson, 2011).

This core of assumptions meets the findings of van Selm *et al.* (2022). They propose, by factoring-in this use of 'non-edible' land, alternative sustainable diets and land-use compared with the EAT-Lancet one of Willett *et al.* (2019). A balanced share of ruminants/PG leads to better performances, including performance in terms of GHG emissions, compared to diets minimizing this ruminant share, e.g. the reference EAT-Lancet diet.

The issue then is to design a diet valuing the optimal number of ruminants, based on the best use of existing PG. In the case of TYFA, the 'best use' means their extensive management for the reasons described above (i.e. maximization of N entry). Going beyond this threshold, meaning more ruminants fed by grains/pulses or expanding PG at the expense of scarce arable land, would indeed decrease the efficiency of the food system. In TYFA, it happens that conserving the overall envelope of PG and reducing their average productivity to 4.5 tonnes of dry matter ha^{-1} on average (assuming that this corresponds to the productivity of semi-natural no-input PG) leads to a reduction of dairy production of 34% and to the quasi-maintenance of ruminants for meat due to the extensification of the dairy livestock, meaning more meat by-produced per tonne of milk at the end (roughly: replacing one cow at 10,000 l milk $year^{-1}$ by two cows at 5,000 l $year^{-1}$ doubles the number of calves l^{-1} milk produced). It should be noted that while the share of grass in the ruminants' diet increases, dairy cows are complemented by feed from crop systems and cakes.

All in all, the resulting diet increases the share of ruminants' meat from 15% in 2010 to 35%, thus leaving the majority of meat supply to pigs and poultry – both in absolute and relative terms: meat from monogastrics still amount to 56 g $person^{-1}$ day^{-1} in the scenario (vs 140 g today), which represents roughly 65% (vs 80% today). Monogastric are indeed key to 'transform' pulses in rotation into organic N – in short: replacing the imported soya by EU-grown legumes – thus contributing to symbiotic N fixation in the arable share of EU agricultural land. As such, TYFA's diet, and other diets in similar modelling, is not vegan but 'flexitarian', with a relatively high share of red meat as compared with other healthy diets (Figure 5).

Conclusion: a revision of the role of PG and ruminants in the sustainability agenda and the needs to further analysis on the science and policy agenda

Permanent grasslands, as used by ruminants, are currently seen more as a problem to deal with than any sort of solution. In the best case, they are restricted to areas where they are the only possible land use, where a certain density of ruminants can be accepted for the list of services they provide. In the worst-case scenario, they are replaced by forests or energy crops. We have discussed the conceptual limitations of such approaches and proposed a scenario based on an alternative framing of issues, combining an original biodiversity/PG/ruminants/climate change nexus. Using the biomass model developed in TYFA, we have factored in a scenario for the EU that leads to positive outcomes for a healthy diet, for biodiversity, natural resources and climate (Figure 6). We insist on the need to adopt a healthier diet, halving the intakes of meat and dairy products to achieve this.

The role of PG and ruminants in this model should not be understood for their only sake, but also for what they bring to the crop systems. A key idea is, in a sense, that the semi-natural functioning of PG provides a series of services (nitrogen fixation, organic matter, biological auxiliaries) that can be transferred in some ways to the cropping systems by ruminants. We have also discussed how the impact of ruminants on climate change should be revised and put in a wider analysis in which the trade-off with N management should be accounted for.

On this basis, we argue for a complete change in the European policies orientation and design. Firstly, there is a need to rebalance the agenda in favour of biodiversity conservation linked to farming and livestock rearing. More precisely, the targets should go beyond existing HNV areas – which nevertheless

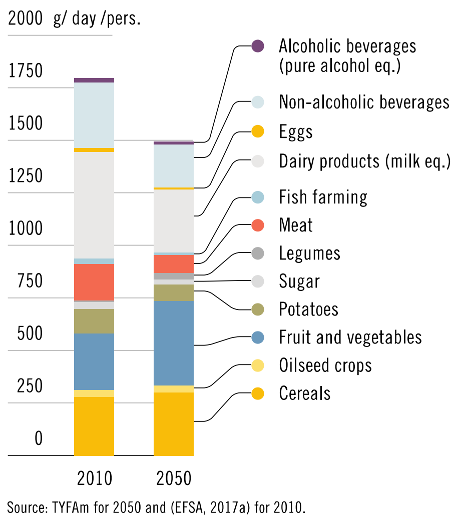


Figure 5. TYFA's assumptions on diets compared to 2010 (EU averages).

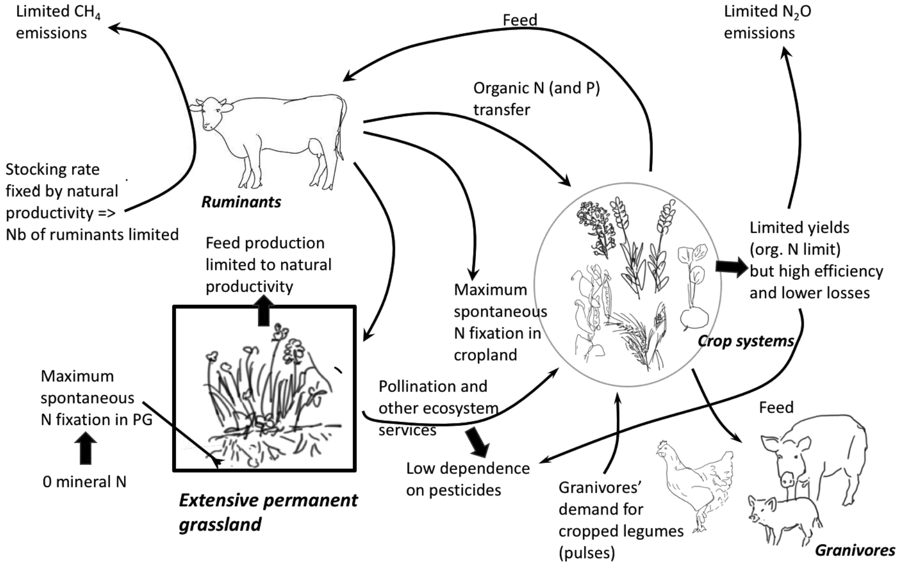


Figure 6. Synthesis of the role of extensive PG in TYFA in the wider agricultural system. Arrows shows the main components and causal relationship of the reasoning.

are a goal in themselves – and hedges and other linear and punctual landscape features meant to address biodiversity conservation in policy schemes such as the CAP cross-compliance. Voluntary and well-funded policies should support the development of extensive PG and associated extensive ruminants livestock, included – and to some extent in priority – in areas where such types of livestock systems have disappeared or are on the way to doing so. Through the fertility transfer allowed in such mixed systems, we identify a path for more autonomous and resilient systems. As these lines are written (in March 2022, at the start of the Russo-Ukrainian war), we strongly argue that this orientation is probably the most resilient one to lower the dependence of the whole agriculture on fossil energy, and that fostering natural N fixation in PG is a major issue for EU food sovereignty, meeting the biodiversity and climate agendas.

Paving the way for policies in this direction opens up different research perspectives. We organize them with regards to the level of organization they are dealing with:

- The farming system (grassland management, livestock management and breeding, integrated mixed farming...), with focus on a better understanding of what takes place in the soil in terms of carbon, N cycle and fixation (as a matter of fact, this is poorly addressed in the literature when it comes to extensive grassland).
- The landscape and the territory, with design of cooperation at this level reflecting integrated land use, meeting environmental and economic goals. This level includes social sciences (participatory science for instance).
- The food chain, and notably its capacity to fully value the assets resulting from integrated PG/crops systems, which is not from far the case today.
- The wider policy design, which is currently neglecting the case for PG despite the fact that it could meet most of the goals assigned to a sustainable food system. In this field, there is a need to better value the methodological frames showing the biodiversity, climate and health performances of integrated mixed systems against those based on reductionist metrics. Making such methodologies influential for policy decision-making needs to link them in a sound socio-economic analysis, revealing the overall benefits for the EU and the rest of the World.

References

Allen M.R., Cain M., Lynch J., *et al.* (2018a) *Climate metrics for ruminants*. Oxford Martin Programme on Climate Pollutants, Oxford, UK, 4 p.

Allen M.R., Shine K.P., Fuglestvedt J.S., *et al.* (2018b) A solution to the misrepresentations of CO_2-equivalent emissions of short-lived climate pollutants under ambitious mitigation. *NPJ Climate and Atmospheric Science* 1 (1), 16.

Barbieri P., Pellerin S., Seufert V., *et al.* (2021) Global option space for organic agriculture is delimited by nitrogen availability. *Nature Food* 2 (5), 363-372.

Bignal E. (2000) The environmental impact of dairy production in the EU' practical options for the improvement of the environmental impact. *European Commission (DGXI) CEAS*, 1179.

Buendia E.C., Guendehou S., Limmeechokchai B., *et al.* (2019) *2019 Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories.* IPCC, Geneva, Switzerland.

Clark M.A., Domingo N.G.G., Colgan K., *et al.* (2020) Global food system emissions could preclude achieving the 1.5° and 2°C climate change targets. *Science* 370, 705-708.

Dainese M., Martin E.A., Aizen M.A., *et al.* (2019) A global synthesis reveals biodiversity-mediated benefits for crop production. *Science Advances* 5 (10), 13.

Duru M., Aubert P.-M., Couturier C., *et al.* (2021) Scénarios de systèmes alimentaires à l'horizon 2050 au niveau européen et français: quels éclairages pour les politiques publiques? *Revue AE&S* 11 (1).

Einarsson R., Sanz-Cobena A., Aguilera E., *et al.* (2021) Crop production and nitrogen use in European cropland and grassland 1961-2019. *Scientific Data* 8 (1), 288.

Fahrig L., Baudry J., Brotons L., *et al.* (2011) Functional landscape heterogeneity and animal biodiversity in agricultural landscapes. *Ecology Letters* 14 (2), 101-112.

Fox J.E., Gulledge J., Engelhaupt E., *et al.* (2007) Pesticides reduce symbiotic efficiency of nitrogen-fixing rhizobia and host plants. *Proceedings of the National Academy of Sciences of the USA* 104 (24), 10282-10287.

Garibaldi L.A., Oddi F.J., Miguez F.E., *et al.* (2020) Working landscapes need at least 20% native habitat. *Conservation Letters* 14, e12773.

Garnett T., Godde C.C., Muller A., *et al.* (2017) *Grazed and confused? Ruminating on cattle, grazing systems, methane, nitrous oxide, the soil carbon sequestration question – and what it all means for greenhouse gas emissions*. Food Climate Research Network, Oxford, UK, 127 p.

Geiger F., Bengtsson J., Berendse F., *et al.* (2010) Persistent negative effects of pesticides on biodiversity and biological control potential on European farmland. *Basic and Applied Ecology* 11 (2), 97-105.

Halada L., Evans D., Romão C., *et al.* (2011) Which habitats of European importance depend on agricultural practices? *Biodiversity and Conservation* 20 (11), 2365-2378.

Herrero M., Havlík P., Valin H., *et al.* (2013) Biomass use, production, feed efficiencies, and greenhouse gas emissions from global livestock systems. *Proceedings of the National Academy of Sciences of the USA* 110 (52), 20888-20893.

IPCC (2019) *Climate Change and Land. An IPCC special report on climate change, desertification, land degradation, sustainable land management, food security, and greenhouse gas fluxes in terrestrial ecosystems*. WMO, UNEP, Geneva, Switzerland.

Lomba A., Guerra C., Alonso J., *et al.* (2014) Mapping and monitoring High Nature Value farmlands: Challenges in European landscapes. *Journal of Environmental Management* 143, 140-150.

Lynch J., Cain M., Frame D., *et al.* (2021) Agriculture's contribution to climate change and role in mitigation is distinct from predominantly fossil CO_2-emitting sectors. *Frontiers in Sustainable Food Systems* 4 (300).

Mottet A., de Haan C., Falcucci A., *et al.* (2017) Livestock: On our plates or eating at our table? A new analysis of the feed/food debate. *Global Food Security* 14, 1-8.

Pärtel M., Bruun H.H. and Sammul M. (2005) Biodiversity in temperate European grasslands: origin and conservation. *Grassland Science in Europe* 10, 1-14.

Pörtner H.-O., Scholes R., Agard J., *et al.* (2021) *IPBES-IPCC co-sponsored workshop report on biodiversity and climate change*. IPBES & IPCC, Bonn, Germany, 28 p.

Poux X. and Aubert P.-M. (2018) *Ten Years for Agroecology in Europe: a multifunctional agriculture for healthy eating. Findings from the Ten Years For Agroecology (TYFA) modelling exercise*. Iddri, Paris, France. Available at: https://www.iddri.org/sites/default/files/PDF/Publications/Catalogue%20Iddri/Etude/201809-ST0918EN-tyfa.pdf.

Roper M. and Gupta V. (2016) Enhancing non-symbiotic N fixation in agriculture. *Open Agriculture Journal* 10 (1), 7-27.

Ryschawy J., Martin G., Moraine M.*, et al.* (2017) Designing crop–livestock integration at different levels: Toward new agroecological models? *Nutrient Cycling in Agroecosystems* 108 (1), 5-20.

Schils R.L.M., Bufe C., Rhymer C.M.*, et al.* (2022) Permanent grasslands in Europe: Land use change and intensification decrease their multifunctionality. *Agriculture, Ecosystems & Environment* 330, 107891.

Simoncini R., Ring I., Sandström C.*, et al.* (2019) Constraints and opportunities for mainstreaming biodiversity and ecosystem services in the EU's Common Agricultural Policy: Insights from the IPBES assessment for Europe and Central Asia. *Land Use Policy* 88, 104099.

Smit H.J., Metzger M.J., Ewert F. (2008) Spatial distribution of grassland productivity and land use in Europe. *Agricultural Systems* 98 (3), 208-219.

Steinfeld H., Gerber P., Wassenaar T.*, et al.* (2006) *Livestock's long shadow – environmental issues and options*. FAO, Rome, Italy.

Strebel O., Duynisveld W. and Böttcher J. (1989) Nitrate pollution of groundwater in western Europe. *Agriculture, Ecosystems & Environment* 26 (3-4), 189-214.

Strohbach M.W., Kohler M.L., Dauber J.*, et al.* (2015) High Nature Value farming: From indication to conservation. *Ecological Indicators* 57, 557-563.

Van Kernebeek H.R., Oosting S.J., Van Ittersum M.K.*, et al.* (2016) Saving land to feed a growing population: consequences for consumption of crop and livestock products. *International Journal of Life Cycle Assessment* 21 (5), 677-687.

Van Selm B., Frehner A., de Boer I.J.M.*, et al.* (2022) Circularity in animal production requires a change in the EAT-Lancet diet in Europe. *Nature Food* 3 (1), 66-73.

Van Zanten H., Meerburg B., Bikker P.*, et al.* (2016) Opinion paper: The role of livestock in a sustainable diet: a land-use perspective. *Animal* 10 (4), 547-549.

Van Zanten H.H., Herrero M., Van Hal O.*, et al.* (2018) Defining a land boundary for sustainable livestock consumption. *Global Change Biology* 24 (9), 4185-4194.

Veen P., Jefferson R., De Smidt J.*, et al.* (eds.), (2009) *Grasslands in Europe of high nature value.* KNNV Uitgeverij, Zeist, the Netherlands.

Wilkinson J. (2011) Re-defining efficiency of feed use by livestock. *Animal* 5 (7), 1014-1022.

Willett W., Rockström J., Loken B.*, et al.* (2019) Food in the Anthropocene: the EAT-Lancet Commission on healthy diets from sustainable food systems. *The Lancet* 393, 447-492.

Other stakeholders than farmers contribute to diversify the management of (peri-)urban grasslands

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Abstract

Peri-urban and urban grasslands can be managed by farmers but also by public structures or service companies that specialize in the maintenance of these areas. Each manager has specific objectives but faced with the biodiversity crisis, these practices are changing. This study explores the diversity of management practices on grassland areas in urban and peri-urban areas according to the type of stakeholder involved. We surveyed 26 stakeholders (11 farmers, 7 public structures and 8 private actors outside agriculture) managing grassland areas in and around the city of Angers, France. The survey explores mainly grazing and mowing practices. A multiple correspondence analysis and a hierarchical ascending classification defined eight groups of management of (peri-)urban grasslands: three groups of mown-only grasslands, two groups that were grazed exclusively, two groups with both grazing and mowing, and finally, unmanaged grassland. The different types of managers have preferential management practices: public structures prefer summer mowing; farmers often have a mixed use of grasslands and do not often have grazing all year round, unlike private non-agricultural companies. However, all the stakeholders implement a diversity of management methods for their plots and the diversity of grassland management is higher when considering stakeholders other than farmers. The next step is to evaluate how this diversity of management could influence the floristic biodiversity of (peri-)urban grasslands.

Keywords: peri-urban, grasslands, management, typology

Introduction

As a result of the strong dynamics of urban expansion, agricultural areas are more in contact with urban and peri-urban areas. Grassland areas managed by breeders stand alongside those managed by public structures or specialized service companies. All of these grasslands in urban and peri-urban areas represent biodiversity hotspots (Cochard *et al.*, 2017) and, faced with the biodiversity crisis (IPBES, 2019), managers are changing their practices (cessation of pesticides, differentiated management, etc.). As each manager has different constraints and objectives (forage production, management of green spaces for recreational purposes, decrease of labour time ...), this study seeks to assess the link between the type of manager and the management practices of the grassland of urban and peri-urban areas.

Materials and methods

We surveyed 26 stakeholders managing grassland areas in and around the commune of Angers, Loire Valley, France: 11 farmers, 7 public structures and 8 private actors outside of agriculture. We conducted a semi-directive survey with each of them, exploring all the grassland management they use and we excluded all intensive management (over 6 cuts a year), which results in 110 managements to analyse. We described grazing practices (type of animals, grazing method, period of presence of the animals ...), mowing practices (number and periods, usage of the cut, height before and after mowing ...) as well as fertilization practices and other amendments. Each stakeholder may use the same management in several grassland areas but they did not have time to describe all the plots they have to manage, so it was impossible to analyse the relation between plot characteristics and management. The survey data were analysed using a multiple correspondence analysis. Different management practices were then grouped using a hierarchical ascending classification. These analyses were performed using the FactoMineR module for R (Lê *et al.*, 2008). For the management categories representing more than 10% of the sample

($n \geq 11$), we looked for the existence of a link with the type of stakeholder implementing them via a Chi^2 test with the Yates correction.

Results and discussion

The results for the types of management are based on the two first axes of the multiple correspondence analysis. These explain 31 and 13% of the variability, respectively. The first axis is mainly driven by the dichotomy between mown and grazed grasslands. Grazed grasslands on this axis are managed with unproductive animals or small ruminants, and under continuous grazing. On the second axis managements with grazing and mowing are separated, and grazing is done with dairy cows or cattle and the grasslands are fertilized.

According to the classification, we identify 8 management groups for urban and peri-urban grasslands. Three groups correspond to mown-only grasslands: several annual non-exported mowings (n=8), one summer hay mowing (n=33), and one autumn mowing left in place (n=5). Two groups of exclusive grazing management are identified and distinguish between plots grazed year-round (including winter) often by sheep or goats (n=11), and plots grazed freely by heifers, dry cows or horses from spring to autumn (n=22). Two groups concern plots with both grazing and mowing (mixed management), differentiating between areas where the first use is mowing in summer and then free grazing in autumn (n=17), and those where the first use is rotational grazing in spring and hay mowing in late spring or refusals in autumn (n=10). Finally, the last management is left to nature without human intervention (n=4).

The management practices identified can be interpreted in terms of intensity of use. Within each of the major categories, we can distinguish management with more or less resource extraction: multiple mowing vs single mowing with or without export, grazing all year round or only during the period of grass growth, first use in the spring or starting only in the summer. And each stakeholder combines these different intensities of use to meet his objectives. This is in line with the results obtained by Roche *et al.* (2010) or Martel *et al.* (2013) on grassland management methods in Brittany and in the Jura.

Table 1 shows the cross-tabulations between type of manager and management category with numbers greater or equal to 11. The corrected Chi-square test shows a significantly different distribution of practices between stakeholders (X^2=20.95, P<0.001). The communities prefer summer mowing and are not very active in the management of grazing with heifers or dry cows. Farmers often have mixed use and do not often graze all year round, unlike the 'green space' companies. However, all the stakeholders implement a diversity of management methods for their plots.

The link between management categories and the type of manager can be explained fairly well by the constraints and objectives assigned to the grasslands managed by these different stakeholders. Indeed, farmers aim to feed their herds and need to build up stocks while feeding productive and unproductive animals. Local authorities are often obliged to provide only one mowing in order to limit workload and

Table 1. Cross-tabulations between type of manager and management category with numbers greater than or equal to 11.

Management	Farmers	Private actors outside farmers	Local authorities
Summer mowing	18	6	9
Mixed management	17	0	0
Grazing by cows	17	5	0
Grazing 12 months a year	2	6	3

realize it before the summer season to allow the use of the grasslands by the city inhabitants (Daniel *et al.*, 2021). Finally, the private companies surveyed were mainly companies that use eco-grazing and tend to mobilize small animals that are more robust and easier to transport than cattle (Eychenne *et al.*, 2020).

Other objectives were also expressed during the interview, like biodiversity or aestheticism, but we were not able to relate these objectives to management classes because the objective was given to too many (biodiversity) or too few (aestheticism) numbers of managements. It would be also important to include the general characteristics (area, form, soil, slope …) of the managed plots to explain the management.

As all the stakeholders have preferences in management, the global diversity of management of peri-urban grasslands is increased when we consider all the type of managers of grasslands. This diversity contribute to the increase of the landscape complexity with is related to a better resilience of ecosystems (Tscharnke *et al.*, 2012).

Conclusions

This work confirms that dairy and cattle farmers have specific grassland management. Other stakeholders involved in (peri)-urban grassland management have practices rarely operated by farmers. These practices can be important for resilience of ecosystems. Future work will aim at evaluating the floristic biodiversity of grasslands under each of these managements in order to help stakeholders to include this objective in the choice of the practices.

References

Cochard A., Pithon J., Jagaille M., Beaujouan V., Pain G. and Daniel H. (2017) Grassland plant species occurring in extensively managed road verges are filtered by urban environments. *Plant Ecology and Diversity* 10(2-3), 217-229.

Daniel H., Moinardeau C., Hédont M., Martel G., Beaujouan V., Pithon J. and Bulot A. (2021) Des espaces herbacés pour favoriser la biodiversité en ville: quels enjeux de gestion ? Retours d'expériences collectées auprès de gestionnaires. Colloque REVER, 29 June-1 July 2021.

Eychenne C., Bories O. and Noûs C. (2020) (Éco)pâturage, (éco)pastoralisme: la gestion de l'espace par les troupeaux, éléments d'analyse et de compréhension, Carnets de géographes. Available at: http://journals.openedition.org/cdg/6086.

IPBES (2019) *Global assessment report on biodiversity and ecosystem services of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services.* Brondizio E.S. *et al.* (eds.) IPBES secretariat, Bonn, Germany. 1148 pages. https://doi.org/10.5281/zenodo.3831673

Lê S., Josse J. and Husson F. (2008) FactoMineR: an R package for multivariate analysis. *Journal of Statistical Software* 25(1), 1-18.

Martel G., Herzog F. and Huguenin-Elie O (2013) Influences of local policies and opportunities on farmers' strategies and grassland management. 64th annual meeting of the European Association for Animal Production.

Roche B., Lanoë E., Le Cœur D., Thenail C. Martel G.(2010) Diversité des systèmes d'élevage et des modes d'exploitation des prairies: quelles conséquences sur la biodiversité ? *Rencontres Recherches Ruminants* 17, 41-44.

Tscharntke T., Tylianakis J.M., Rand T.A., Didham R.K., Fahrig L., Batáry P., …, and Westphal C. (2012) Landscape moderation of biodiversity patterns and processes – eight hypotheses. *Biological Reviews* 87, 661-685. https://doi.org/10.1111/j.1469-185X.2011.00216.x

Regulatory and social context linked to European grassland in a bioeconomy context (GO-GRASS)

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Abstract

Grasslands are effective for carbon sequestration as they store carbon mostly underground, compared to forest carbon storage which is mostly in woody biomass and leaves. Grasslands are also key for socioeconomic development in EU rural areas. As part of the Horizon 2020 GO-GRASS project, this study aims to provide an analysis of the CAP 2014-2020 and offer recommendations to promote grasslands within the different EU strategies and policies. Permanent grassland is outstanding as a key type of land to be funded; grazing activities are beneficial, enhancing biodiversity and favouring water protection, and under-grazing could lead to problems related to inadequate biomass management. Mowing is highlighted as an option linked to bioeconomy activities like those proposed by GO-GRASS on grass-based businesses. However, only Measure 16 supports valued products from grassland and it is implemented in only a few RDPs. Concerning the lack of measures and policies supporting permanent grasslands, it is recommended to: (1) boost knowledge transfer through demonstration fields and extension services; (2) establish measures promoting cooperation and adequate land management; and (3) turning arable land into grassland to take advantage of its environmental benefits.

Keywords: CAP, permanent grassland, rural development, green deal, sustainability

Introduction

The European Commission has acknowledged that climate change and environmental degradation are as major threats. In this context, grasslands stand out as effective for promoting carbon sequestration through their substantial underground carbon storage (Dass *et al.*, 2018). In addition, grasslands are basic for the rural economy and have a role in addressing depopulation in EU rural areas, a problem that EU and national governments are having to deal with. The European Green Deal and related strategies intend to build an EU economy based on sustainability and competitiveness. The Common Agricultural Policy (CAP) is the main driver of agricultural policy in the EU, funding the different types of land and also livestock by means of the coupled measures. The CAP is based on conditionality, Pillar I and Pillar II, requiring a set of norms that need to be fulfilled by a farmer to access funding. Payments linked to Pillar I are funded by the European Agricultural Guarantee Fund, while payments linked to Pillar II are partly funded by national governments with a 50-85% range depending on the country (Mosquera-Losada *et al.*, 2016). Pillar I entails the highest amount of allocated budget managed through payment rights linked to arable crops, permanent grassland (PG) (defined as grasslands over 5-years old) and permanent crops as land-use categories. In addition to direct payments, the 'Payment for agricultural practices beneficial for the climate and the environment' (Greening) represents 30% of the payments for the Member States (MS). On the other hand, Pillar II is related to rural development and involves different measures promoted by MS. Considering the aforementioned, grasslands are supported by CAP in both Pillars I and II. The aim of this study is to analyse the current CAP measures that foster grasslands at the different international-EU levels, including socioeconomic aspects influencing the implementation of future policies.

Materials and methods

To develop this study, an in-deep review of EUROSTAT data was performed to draw a picture of the current social situation of farms in Europe. These data included information at the NUTS 2 level on permanent pasture and permanent grassland (PG), livestock on farms, young and female farm holders, and holders' tenant evolution for different years ranging from 2000-2016. In addition, metadata analysis was carried out through CAP Pillar II mainly, since Pillar I application presents no significant differences among countries and information is more easily accesible. For the analysis of Pillar II, an evaluation of the different rural development programmes (RDPs) was developed by employing deeple translation tool and a set of keywords associated with grasslands, allowing us to identify the most relevant practices linked to the measures developed in the current 118 RDPs. Nine RDPs correspond to general frames of some MS with more than one RDP region (France, Germany, Italy and Spain) and five for the overseas regions of France. The present study is focused on the remaining 104 regional RDPs of the continental EU and surrounding islands.

Results and discussion

Pillar I

Considering the differences existing between countries on land eligibility for direct payments, both arable land (with temporary grassland) and PG/permanent pasture could be linked to grassland use according to their definition in the Regulation (EU) 1307/2013. The declaration of the PG area as eligible relies on the MS that can adapt a pro-rata system based on the recognition of annual-herbaceous species (self-seeded) and woody dominated grassland vegetation through its recognition as traditional practices and habitat conservation. In addition, as a consequence of the midterm review of the current CAP, the OMNIBUS regulation (2018) acknowledges shrubs/trees producing animal feed (Table 1).

For greening payments, crop diversification affects farms with large cropland areas. The presence of permanent grassland or crops linked to grasses or other herbaceous forage on a farm makes crop diversification already fulfilled. Greening is not compulsory on those holdings: (1) where more than 75% of the arable land is used for the production of grasses or other herbaceous forage selected by the MS; (2) more than 75% of the eligible agricultural area is permanent grassland, used for the production of grass or other herbaceous forage; (3) more than 50% of the areas of arable land declared were not declared by the farmer in his aid application of the previous year; and (4) that are situated in areas north of 62°N latitude or certain adjacent areas. Attending to this, farms with high share of permanent grasslands fulfil the greening requirements. Member States shall ensure the ratio of PG to the total agricultural area declared by farmers will not decrease by more than 5% compared to a reference ratio established by MS.

Pillar II

The 2014-2020 RDPs are composed of 16 measures, common to all MS, consisting of sub measures also common for the MS that are specified through operations, that are designed by each RDP in order to be

Table 1. Member states that extended the definition of Permanent grasslands (PG) as established local practices arguing traditional practices, conservation of habitats and through the OMNIBUS regulation.

Reason – Countries	CY	DE	EL	ES	FR	IE	IT	PT	SE	UK	BG	HR	LT	SK
Traditional practices	X	X	X	X	X	X	X	X	X	X				
Habitat conservation		X	X	X					X					
OMNIBUS = if ploughed non-PG	X	X	X	X			X				X	X	X	X
OMNIBUS – PG may include shrub/trees for animal feed if herbaceous remain			X	X	X			X						

more precise. The whole analysis of the RDPs shows that 1,518 operations within the 16 measures were developed for the 28 EU countries (Figure 1).

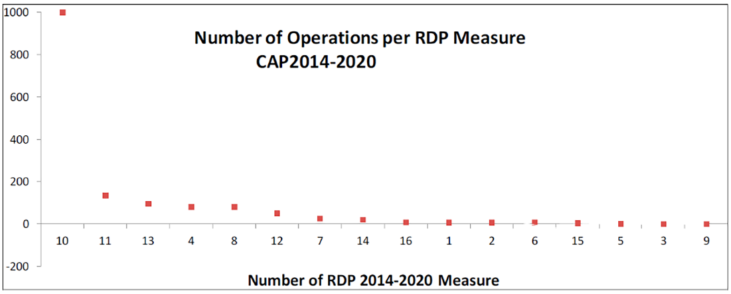

Figure 1. Number of operations per RDP Measure.

Measure 10 is the most employed measure of the RDP, aiming at the enhancement of ecosystem services (biodiversity, erosion …) but also at improved management. All regions except the Balearic Islands and Hesse used this measure to promote grasslands. Measures 11 (promoting grazing systems within organic farming) and 13 (animal welfare in extensive farming) follow Measure 10 on operations promoting grasslands. Only eight RDPs activate Measures 1 and 2 as part of the knowledge transfer improvement linked to grasslands in Europe. Measure 4 is related to the restoration of grasslands from under- and over-grazing. Measures 5 and 6 are used to promote grassland by a few RDPs, while Measure 7 pursues enhanced energy saving to reorient pasture management and enhance cultural and natural heritage of pastures and meadows, including silvopasture, mountain and summer pasture to promote biodiversity. Measure 8 associates grazing and grassland promotion in forest areas, of which Measure 8.2 is the most popular linked to silvopasture, an agroforestry practice, to reduce forest fires. Measure 15 promotes silvopastoralism in forest areas. Finally, Measure 16 is unique in linking grasslands to bioeconomy through cooperation, where products are valorised when linked to grasslands.

Socioeconomy of grassland farms

The share of PG on different EU farms showed clear decreases until 2007, with a progressive increase since 2016, mainly in UK, Sweden, NW Spain and France, which is indicative of high availability of grasslands for alternative uses linked to the bioeconomy. The owner's age is high, a consequence of the ageing farming population, while numbers of female holders, though increasing in recent years, remain low.

Conclusions

Permanent grassland is one of the most important types of land to be funded by the EU due to the large surface it occupies and the multiple ecosystem services it provides, such as carbon sequestration. Measure 16 relates the promotion of valued products from grassland, in contrast with the EU acknowledgement of grazing activities being key to maintain and enhance biodiversity and protect water. Considering the policies and socioeconomic analysis, three recommendations can be listed: (1) to boost knowledge transfer through demonstration fields and extension services; (2) to establish measures promoting cooperativism and adequate land management; and (3) turning arable land into grassland to exploit its environmental benefits.

References

Dass P., Houlton B.Z., Wang Y. and Warlind D. (2018) Grasslands may be more reliable carbon sinks than forests in California. *Environmental Research Letters* 13, 074027. https://doi.org/10.1088/1748-9326/AACB39

Mosquera-Losada, M.R., Santiago Freijanes, J.J., Pisanelli, A., *et al.* (2016) Extent and success of current policy measures to promote agroforestry across Europe. Deliv. 8.23 for EU FP7 AGFORWARD 613520. 95 pp.

Variability of European farming systems relying on permanent grasslands across biogeographic regions

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Abstract

The relevance of permanent grasslands (PG) for a large share of European farms is high, and yet understudied. We used single-farm records from the FADN (Farm Accountancy Data Network) database 2017, which included 41,926 farms-with-PG to characterize PG-based farming systems. Each farm was assigned to one class in terms of: (1) main livestock species/category; (2) stocking rate on total farmland; (3) PG share; (4) biogeographic region (BGR). We carried out a Multi Correspondence Analysis (MCA) on the resulting classification, which explained 20% of the variance. The five BGR separated well in the first two MCA dimensions. Alpine farms were predominantly related to beef cattle, with relatively low stocking rates, and intermediate to high PG shares. Atlantic farms also revealed high PG shares, but were linked to higher stocking rates and 'Mixed bovine' and 'Dairy cow' farming. The dominance of farms without livestock in the Boreal BGR resulted in generally very low stocking rates and showed a limited importance of PG. Continental farms were not clearly related to one specific livestock category or a stocking rate, but consistently showed a share of 10-30% PG per farm. Finally, the Mediterranean BGR separated from the others, being dominated by sheep and goat farming.

Keywords: Europe, farming system, grassland management, livestock species, meadow, pasture, stocking rate

Introduction

Farming systems (FS) are the result of environmental conditions, historic and cultural factors, policies, and management practices. Based on these regional differences, the ability of different FS to deliver ecosystem services (ES) can vary widely (Ribeiro *et al.*, 2021). Therefore, it is important to recognise which factors differentiate FS from each other, to address further actions to improve productivity and sustainability, create resilience, optimize farm profitability, and deliver ES for the society (Santos *et al.*, 2021). Permanent grasslands (PG) provide high-quality fodder and are able to deliver a variety of important ES (Roy and Potschin, 2018). A first attempt to implement a FS typology considering the role of PG within farms was provided by Hercule *et al.* (2017), but based only on grassland share within farms and animal stocking density. To overcome these limitations, we implemented a new FS typology within the H2020 project 'SUPER-G' (Developing SUstainable PERmanent Grassland systems and policies), aiming to identify the main FS that rely on PG, with a view to assessing the extent to which different FS deliver multiple ES. Livestock species, stocking rate, PG share, and the biogeographic region (BGR) were selected as discriminating factors, and their role in differentiating European FS is discussed here.

Materials and methods

A dataset containing single farm records was retrieved from the 2017 Farm Accountancy Data Network (FADN) and used as a representative sample of European farms. The main advantage of working with FADN data, is the A subset of records including only farms with PG was selected, which included 41,926 farms located in 1063 NUTS3 regions belonging to 28 European countries. Each farm was assigned to a class according to four descriptors (i.e. qualitative variables). The first variable was the main livestock species/category, i.e. the species or category accounting for more than 75% of livestock units (LU) on the farm, selected among: beef cattle; milking cows; mixed bovines (i.e. farms with both beef cattle and milking cows); sheep and goats; mixed ruminants (i.e. farms with bovines - either beef cattle, or milking cows, or both - and sheep or goats); mixed and others (i.e. farms with other livestock species such as horses or pigs together or not with bovines, sheep, or goats). The second variable was the stocking rate on total utilised agricultural area (UAA) of a farm, calculated as the ratio between LU of main domestic herbivores (i.e. bovines, sheep, goats, and equines) and the UAA, which resulted in four classes (<0.5; 0.5-1; 1-2; >2 LU ha^{-1}). The third variable was the PG share of the UAA, divided into five classes (<10; 10-30; 30-50; 50-70; >70%), and the fourth variable was the BGR where the farm was located (i.e. Alpine, Atlantic, Boreal, Continental, or Mediterranean). The resulting dataset containing the four qualitative variables was used to perform a multiple correspondence analysis (MCA). The analysis was carried out in R (v. 4.0.3, R Core Team 2020), using 'FactoMineR' package (Husson *et al.*, 2016).

Results and discussion

The FADN database proved to be effective to explore farm variability throughout Europe, due to the vast amount of available data covering all regions of EU-28. The first two dimensions of the MCA explained 12.6 and 9.1% of the total variance, respectively (Figure 1). The five BGR separated quite well in the first two MCA dimensions. More specifically, the typical FS of the Alpine BGR was mainly related to beef cattle, relatively low stocking rates, and intermediate to high PG share per farm in line with Sturaro *et al.* (2009), highlighting the extensiveness of FS. The Atlantic BGR also showed high PG shares but, compared to the Alpine BGR with higher stocking rates and more 'Dairy cow' farms as shown by Stypinski (2011), indicating more intensive FS. Farms without or with mixed livestock dominated in the Boreal BGR and were associated with very low stocking rates and a very low PG share per farm. This is likely determined on the one hand by agricultural intensification, leading to spread of temporary grasslands at the cost of PG, and on the other hand by abandonment of extensive PG (Aune *et al.*, 2018). The majority of the continental farms were mostly related to 10-30% PG share class but not clearly to a specific livestock category or stocking rate, which is probably due to the high variability of environmental and socio-economic conditions of this BGR. Finally, the Mediterranean BGR, in the upper part of the plot, clearly separated from the other BGRs, being strongly related to the presence of small ruminants on farm. Indeed, sheep and goats are the species mostly kept in the Mediterranean area (Porqueddu *et al.*, 2017), due to their ability to exploit low quality forage.

Conclusions

The FS typology developed for this study provides a selection of factors that can be used to distinguish farm types that rely on PG according to their level of management intensity, and the delivery of associated ES. Such a typology helps understand the variability of farming systems across BGR of Europe and the role of PG in supporting each of them. The typology could also be important for grading farms according to their ability to deliver ES to the society, while promoting the development of sustainable management practices and agri-environment schemes. Future research should also consider the variability in distinct types of PG, between and within BGR, as a key factor shaping ES delivery of FS at farm level.

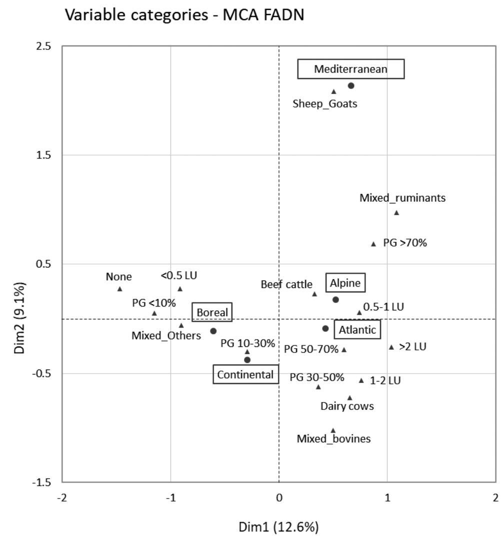

Figure 1. Relationships between variable classes in the first two MCA-dimensions performed on the FADN database. The variance explained by each dimension is reported in brackets. LU, livestock units, PG, permanent grasslands.

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References

Aune S., Bryn A. and Hovstad K.A. (2018) Loss of semi-natural grassland in a boreal landscape: impacts of agricultural intensification and abandonment. *Journal of Land Use Science* 13(4), 375-390.

Hercule *et al.* (2017) Une typologie pour présenter la diversité des territoires d'élevages en Europe. INRA Prod Anim. 20147, 30(4), 285-302.

Husson F., Josse J., Le S., Mazet J. and Husson M.F. (2016) Package 'FactoMineR'. *An R package* 96, 698.

Porqueddu C., Melis R.A.M., Franca A., Sanna F., Hadjigeorgiou I. and Casasús Pueyo I. (2017) The role of grasslands in the less favoured areas of Mediterranean Europe. *Grassland Science in Europe* 22, 3-22.

R Core Team (2020) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Ribeiro P.P.D.C.F., Canadas M.J.P.R., de Vinha Novais A.M.C., Moreira F.M.R.F. and Lomba Â.C.D.A.R. (2021) Explaining farming systems spatial patterns: A farm-level choice model based on socioeconomic and biophysical drivers. *Agricultural Systems* 191, 103140.

Roy H.-Y. and Potschin M.B. (2018) Common international classification of ecosystem services (CICES) V5.1 and guidance on the application of the revised structure. *European Environment Agency* 53.

Santos J.L., Moreira F., Ribeiro P.F., Canadas M.J., Novais A. and Lomba A. (2021) A farming systems approach to linking agricultural policies with biodiversity and ecosystem services. *Frontiers in Ecology and the Environment* 19(3), 168-175.

Stypinski P. (2011) The effect of grassland-based forages on milk quality and quantity. *Agronomy Research* 9 (Special Issue II), 479-488.

Sturaro E., Cocca G., Gallo L., Mrad M. and Ramanzin (2009) Livestock systems and farming styles in Eastern Italian Alps: an on-farm survey. *Italian Journal of Animal Science* 8(4), 541-554.

Towards protein self-sufficiency for both dairy and beef cattle in western France

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Abstract

Ruminant diets require rations rich in energy and protein. Maize silage forms the basis of many rations in western France but it has a low content of digestible protein and must be combined with feedstuffs rich in protein. Concentrate costs have a direct impact on farm profitability. Therefore, farmers seek to produce more feed resources on their farms to be more independent of increasing world market prices. Two programs, called OPTIALIBIO on organic production and 4AGEPROD on both conventional and organic production, have been implemented. Increasing protein self-sufficiency has been tested at farm scale for both dairy and beef cattle. Compared with perennial ryegrass-white clover swards, multispecies swards including tall fescue and red clover for grazing produced higher yields of dry matter (+2.5 t DM ha^{-1} $year^{-1}$) and protein (1.5 t crude protein ha^{-1} $year^{-1}$). For silage or hay, the best mixtures were composed of red clover and grasses. The research for improving protein self-sufficiency in both dairy and beef cattle, by including alternative forages is a real opportunity for farmers in western France. Economic simulations underline a risk associated with increased self-sufficiency as costs usually increase as a result of switching towards multispecies swards.

Keywords: dairy cow, beef cattle, protein, feed, self-sufficiency

Introduction

Ruminant nutrition is mainly based on energy and protein. Roughages represent the biggest share of the average daily intake of a dairy cow. Maize silage is the most important one in Western France dairy systems. Nevertheless, maize has a low content in digestible protein and must be combined with feedstuffs rich in protein, very often rapeseed meal or imported soybean meal. Thus, given the fact that profitability of cattle production is proportionally linked to the concentrate costs, farmers seek to produce as much as possible their feed resources on their farms (Brocard *et al.*, 2015). Therefore, through two programs called 4AGEPROD and OPTIALIBIO, increasing protein self-sufficiency has been tested for both dairy and beef cattle.

Materials and methods

Four experimental farms in western France have been involved in this work over 4 years: two for dairy cows (2×79 cows over 4 winters for conventional production, and 2×29 cows over 2 winters for organic production) and two for beef production (4×14 young bulls over 2 periods (in Etablières 2×16 young bulls, and 2×16 heifers over 2 periods in Mauron). OPTIALIBIO focused on different pasture mixtures in a 100% pasture management for dairy production to improve feeding self-sufficiency in organic farms (Madeline *et al.*, 2016). Six mixtures (M1 to M6) were tested in comparison with two controls (Figure 1). Modalities were repeated three times within the plot. For 5 years, and for each cycle, data on available biomass, floristic composition by blends and the food values were collected. In 4AGEPROD the valorisation of harvested grass on farm was studied for both conventional and organic production. Two modalities were compared: late or classic cut versus early cut. For these modalities, trials were

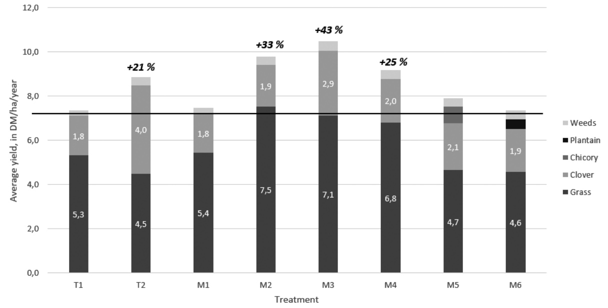

Figure 1. Average yields of pasture mixtures over 5 years (T1: ryegrass + white clover; T2: hybrid ryegrass + white clover; M1: ryegrass + white clover + hybrid clover; M2: ryegrass + fescue + white clover + hybrid clover; M3: ryegrass + fescue + white clover + red clover; M4: ryegrass + 2 types of fescue + white clover + hybrid clover; M5: ryegrass + white clover + chicory; M6: ryegrass + white clover + plantain).

implemented with groups of animals, either dairy (Brocard *et al.*, 2019, 2020) or beef cattle. For each trial, comparison was realized between a control group with a low level of dietary self-sufficiency versus an experimental group with a higher self-sufficiency. Economic simulations were made to compare the modalities. The complementarity of the programs implemented makes it possible to study different types of forage (pasture, hay, wrapped bales, silage), different plant species (alfalfa, perennial ryegrass, hybrid ryegrass, white clover, red clover, crimson clover, plantain) and different types of animals (dairy cows, young bulls, heifers, cull cows) from various breeds (Holstein, Limousin, Charolais).

Results and discussion

For OPTIALIBIO, the addition of a new forage species to perennial ryegrass-white clover showed no increased yield over the 5 years. Despite slightly lower measured digestibility for multispecies grasslands, the differences in feed values were not significant. There was a difference between T1 and T2 as T2 produced 21% more DM than T1. Diversification of the types of clover, especially by the addition of red clover, made it possible to improve both yield and proportion of legumes relative to white clover. The yield of multi-species meadows was higher than the yield of perennial ryegrass-white clover, except for one modality (M1). These mixtures produced an average of +2.5 t DM ha^{-1} $year^{-1}$ over T1 (Figure 1). These results are in line with the conclusions of previous trials in less favourable areas for the growth of grass, and in particular for perennial ryegrass (Roca Fernandez *et al.*, 2014). These three mixtures (M2, M3, and M4) produced on average between 1.47 and 1.65 t of CP ha^{-1}, or +270 to +440 kg, over the perennial ryegrass.

In dairy production, alfalfa silage and grass silage were tested in the 4AGEPROD program. Results showed no statistical differences in milk, fat content and protein content as long as the share of alfalfa silage in the diet did not exceed 20%. When using 40%, milk production significantly dropped by 10%. However, the soybean meal requirements can be reduced by 50%. The use of alfalfa silage decreased feeding costs by a maximum of 5 € 1000 $litres^{-1}$ of milk. This impacts on the final income over feed costs, which represent € 2,500 per year for a farm producing 500,000 litres per year.

For grass silage in conventional agriculture, the early harvest system led to 5 cuts per year with a reduced yield per ha in two years out of four (average -1.5 t DM ha^{-1}), compared to the late cut. Important variations between years were noticed. However, the early harvested forage always offered higher nutritive values both in energy and protein. Thus, the yields of net energy and protein per ha were globally improved, reaching 9,392 UFL and 1.64 t CP ha^{-1} (respectively +18% and +32% compared to late cut). Farm incomes over feed costs were slightly affected (-2 € 1000 $litres^{-1}$) in a bad year but increased +11 € 1000 $litres^{-1}$ in a good year. However, this technique does not involve a change of system and can therefore be implemented quickly and simply but with more work required when harvesting (2 h day^{-1}). In organic production, the search for better fodder quality has significantly improved milk performance (raw milk +3.3 kg cow^{-1} day^{-1}). This has repercussions on the income over feed cost (+14 € 1000 $litres^{-1}$).

In beef production, results on young bulls and heifers for fattening indicate that it is possible to reach total autonomy by replacing up to 100% of the protein-rich concentrate by wrapped alfalfa while maintaining equivalent growths and also improving the margin over feed cost (+38 € $head^{-1}$). Due to high variability of harvested grass, some trials also found a significant decrease in performance for bulls fed on alfalfa. The margin on feed costs is then reduced (8%). Growth performance is sometimes negatively impacted, especially when the energy density of the ration is reduced due to the introduction of legume silages (as shown by Bastien *et al.*, 2016). Further work is necessary to get new and more conclusive insights regarding the economic aspects for consideration in this investigation.

Conclusions

In organic production, chicory and plantain did not show any interesting effect on yields or feed values compared to ryegrass-white clover. All multi-species pastures showed increased annual yields in this study, compared to a classic ryegrass-white clover pasture. An average gain of +2.2 t DM ha^{-1} $year^{-1}$ was observed. Energy and protein production per hectare were higher for all multi-species pastures. These results show that multi-species pastures are convincing and can be considered as a solution to improve self-sufficiency of dairy farms. In addition, improving farm protein self-sufficiency in dairy and beef cattle production farms by including alternative harvested forages is a real opportunity for farms in western France. Whether from grass silage or alfalfa in different forms, the gain of protein autonomy is possible, going from 39 to 72% with alfalfa silage and 67 to 73% for grass silage. This reaches 100% on organic production.

Acknowledgements

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References

Bastien D., Benoit M. and Valence S. (2016) Engraissement de jeunes bovins charolais avec de l'ensilage d'herbe. *Idele, collection Résultats* n#00 16 301 030, 66p.

Brocard V., Belot P.-E., Foray S., Perrot C. and Rouillé B. (2015) High output farming systems in Europe: the French case. *Grassland Science in Europe* 20, 26-39.

Brocard V., Cloet E., Tranvoiz E. and Rouillé B. (2019) Ryegrass-red clover mixtures: impact of harvest stage for silage on yield and nutritional value. *Grassland Science in Europe* 24, 213.

Brocard V., Cloet E., Tranvoiz E. and Rouillé B. (2020) Ryegrass and red clover mixtures for dairy cows: impact of harvest stage for silage on intake, production and income. *Grassland Science in Europe* 25, 221-223.

Madeline L., Drieu C., Martin G. and Philibert A. (2016) Organic cattle breeding systems: food self-sufficiency and resilience to climatic hazards. *Rencontres Recherches Ruminants* 23, 267.

Roca-Fernandez A.I., Peyraud J-L., Delaby L., Lassalas J. and Delagarde R. (2014) Interest of multi-species swards for pasture-based milk production systems. *Grassland Science in Europe* 19, 728-730.

Contrasted evolution of grassland area across Europe in the past decades to promote grass-based business opportunities in rural areas (GO-GRASS)

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Abstract

Rural depopulation demands that there is clear support from authorities to revitalize the rural economy. The H2020 GO-GRASS project aims to create new business opportunities in rural EU by boosting knowledge and analysing the potential for replication of business models. In order to achieve the general objectives, this study seeks to provide an analysis of the current situation for permanent and temporary grassland in Europe. The methodology employed was the creation of maps based on all data and years available in the databases of Corine Land Cover (CLC) and LUCAS. According to CLC, northern and southern countries of Europe increased the proportion of grassland. However, in recent years, most of eastern and northern countries increased their grassland proportion, while in southern and western countries it was reduced. On the other hand, LUCAS reflects a clear reduction of permanent grasslands in western Europe, but an increase in some eastern and northern countries with a low percentage of permanent grasslands. The central countries, as evidenced by both databases, show clearly reduced proportions of grasslands. In contrast, temporary grassland has increased all over Europe. Finally, grazed and silvopasture areas were maintained, and livestock presence appears to be clearly specialized, with larger animals more associated with northern and central countries and the smaller animals with the South.

Keywords: bioeconomy, grass, rural development, Green Deal, sustainability, land use

Introduction

Permanent and temporary grassland are defined in the EC regulation 1307/2013 of the Common Agricultural Policy (CAP). Grasslands can receive payments from the CAP linked to 'arable crops' with temporary grasslands, or as 'permanent grassland/pasture' (Mosquera-Losada *et al.*, 2016). The new definition of permanent grassland recognizes all types of permanent grasslands across European biogeographic regions including the possibility of recognizing 'self-seeded' (annual herbaceous species) and 'grasses and other herbaceous forage' linked to southern summer drought conditions.

Materials and methods

To develop this study, data were gathered and maps were created based on the years available in the databases of Corine land cover (CLC) and Land Use/Cover Area frame Survey (LUCAS) following the methodology of Mosquera-Losada *et al.* (2016) to calculate changes.

The CLC database is based on polygons. The minimum surface mapping unit is 25 ha, while the linear elements collected are those with a width of at least 100 m when silvopasture areas were considered. The CLC includes permanent grasslands but also the concept of Natural grasslands (areas with herbaceous vegetation covering >50% surface). LUCAS corresponds to a Eurostat Survey checking 1,100,000 points separated 2 km north-south and east-west in EU countries with photo interpretation techniques. After a selection was visited *in situ* by the LUCAS surveyors (about 330,000 points the last survey in 2018). LUCAS show two cover and two uses. In addition, there are other observations as Land Management, informing if there are signs of grazing or not. LUCAS points do not have a minimum unit to be mapped, the only condition is on the selected coordinates. Grassland corresponds to LUCAS B55 (temporary grassland – artificial pastures – crop rotation ≤5 years) and E classes (included permanent grassland – no crop rotation ≥5 years).

Data mapping

CLC was used in raster format with a 1 ha pixel^{-1} resolution (biogeographical issue). With the open application QGIS, Version 3.10.1-A Coruña, the zonal stats were acquired in the shapefile with the EU NUTS2 areas. LUCAS data are points in CSV format. GIS software was employed to join attributes by location and after Excel filtering to select points with the main or secondary cover as grassland and cluster in NUTS2 regions.

Results and discussion

When analysing the evolution of grassland in Europe in the last decades we have two different perspectives on data source and time frame (Figure 1). LUCAS includes data from 2009-2018 indicating that in most of western Europe the area percentage of grasslands had reduced while in the eastern part of Europe it was maintained or increased. On the other hand, CLC covers a longer period from 1990-2018 offering not so clear patterns but reflecting that in most of Spain, Germany, Ireland, Estonia, Rumania, Ukraine, part of Denmark and Poland the proportion of grasslands in their areas had increased, whereas in France, Italy, Greece and most of the Eastern countries it had reduced. If splitting is based on shorter time frames, it is observed that from 1990 to 2000 grassland area generally decreased while from 2012 to 2018 it increased. This may be explained partly by the fact that CAP payments are linked to land use and not to production but also to the fact that from 2013 the CAP regulations asked countries not to reduce their permanent grassland area by more than 5%.

Analysis of the permanent grassland evolution in Europe in the 9 years to 2018 (Figure 2) reveals that most of the zones in the western part of Europe saw a decrease in the proportion of permanent grasslands while in the eastern and northern parts of Europe the situation is more contrasted. During 2006-2009 there was a clear reduction of permanent grassland in Europe, even more marked for the period 2012-2015, but during the period 2015-2018 most of the areas were either maintained or increased, probably as a consequence of the 5% maintenance of the permanent grassland promoted by the Greening of the CAP for this type of land use. This increase occurred in the northern countries where permanent grassland extent rates were low.

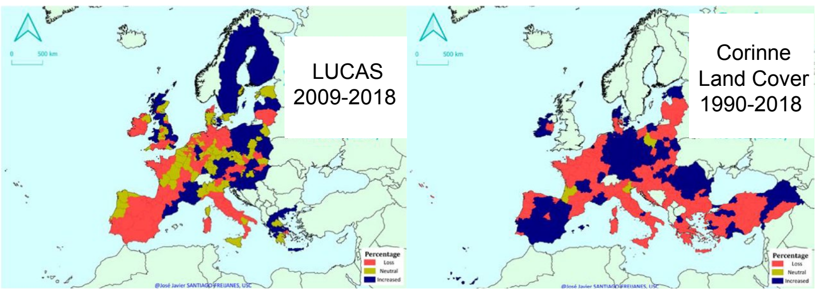

Figure 1. LUCAS and CLC grassland evolution in percentage. LUCAS comprises the 2009-2018 period and CLC 1990-2018 with red for the loss of grassland; yellow for a neutral situation and blue for the increase in grassland surface. LUCAS ranges from -5% (loss) to 5% (increase) while CLC ranges from -2.5 to 2.5%.

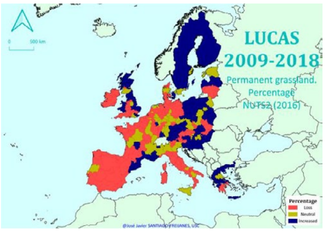

Figure 2. Permanent grassland evolution in the 2009-2018 period, with red for the loss; yellow for neutral and blue for increase, being Neutral situation -2.5 to 2.5%.

When dealing with temporary grassland, the situation is the opposite. Temporary grassland shows a generalized increase all over Europe (Figure 3), with the exception of the 2012-2015 period. Nevertheless, it is important to highlight that the extent of temporary grassland is very low.

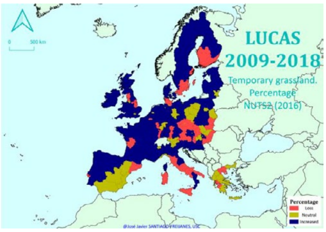

Figure 3. Temporary grassland evolution in the 2009-2018 period, with red for the loss; yellow for neutral and blue for increase, Neutral situation being -0.5 to 0.5%.

In Europe, grasslands can be grazed or harvested. Permanent grasslands are mainly grazed and harvested when possible, whereas temporary grasslands are more usually harvested or cut and grazed.

According to the LUCAS data on sites with grazing evidence, grazing seems to be more associated with the western part of Europe whereas grazing is scarce in the eastern or northern parts of Europe. This could be related to the low proportion of permanent grassland in North-western Europe. As a general pattern, grazed areas were maintained and even increased between 2009 and 2018. In addition, the areas of unmanaged natural grasslands have increased in most parts of Europe, probably as a consequence of population ageing and land abandonment.

Conclusions

Based on data analysis from 1990 to 2018, the total grassland proportion in northern and southern areas of Europe has increased while in the central areas of Europe it has reduced. Nevertheless, when considering the timeframe 2009 to 2018, linked to the LUCAS dataset, most of the eastern and northern countries of Europe have increased their proportion of grassland while those in southern and western Europe have reduced it. Permanent grasslands are so far the most important type of grasslands in Europe, with a higher presence than temporary grassland. Permanent grasslands were clearly reduced in the western part of Europe, while increasing in some eastern and northern EU countries that have a low percentage of permanent grasslands. In contrast to the situation for permanent grassland, a generalized increase of the area of temporary grassland was found all over Europe.

References

Mosquera-Losada M.R., Santiago-Freijanes J.J., Pisanelli A. *et al.* (2016) Extent and success of current policy measures to promote agroforestry across Europe. AGFORWARD project. Deliverable 8.1.

Mosquera-Losada M.R., Santiago-Freijanes J.J., Ferreiro-Domínguez N., Rodríguez-Rigueiro F.J. and Rigueiro-Rodríguez A. (2019) Report with the maps of different permanent grasslands GO-Grass project. Deliverable 1.1.

Citizen and consumer attitudes to grassland landscapes in Europe

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Abstract

Permanent grassland (PG) landscapes offer an important mix of ecosystem services (ES) which include habitat provision, carbon sequestration, water quality protection, food production and cultural activities, among others. In policies relevant to PG management, citizen and consumer demand for ES are less well considered than the supply of ES. In order to develop sustainable grassland food systems, it is important to understand attitudes of citizens and consumers to ensure sustainable management of PGs, and the balance of ES they provide, in order to develop sustainable grassland food systems. This study focuses on the work of researchers in the H2020 SUPER-*G* project (Sustainable Permanent Grassland Systems and Policies) to identify priorities and preferences of citizens for ES provision from PG landscapes in a comparison across five biogeographic zones in Europe. We present results of an international online survey (n=3,184) conducted with a nationally representative sample of citizens in five European countries (UK, Spain, Sweden, Czech Republic and Switzerland) in 2021. Structural equation modelling is used to model the drivers of public attitudes to grassland landscapes, including understanding factors that predict behavioural intentions to spend time in grasslands, and purchase products from grassland. We discuss the results in the context of the processes required to co-develop sustainable policy options for PG, and the role of citizen priorities in the development of agri-environment policy.

Keywords: ecosystem services (ES), permanent grassland (PG), citizens and consumer attitudes, sustainable management, agri-environmental policy, quantitative analysis

Introduction

Permanent grasslands (PG) are multifunctional landscapes that produce multiple benefits for environment and society through delivery of a variety of ecosystem services (ES). Management of PG is often governed by policies that consider a narrow range of ES while not fully considering societal demand for ES. However, as more holistic policies are to be implemented, including linking financial support for farmers to production of public goods, there is a need for a greater coherence when considering public attitudes to, and preferences for, ES from PG. This is to ensure that agricultural policy and farm practices align with societal priorities and respond adequately to the development of citizens' attitudes and values. Attitudes are a 'deeply held mental stance' that connect to preferences for, and perceptions of land or landscape, as well as the way that people attach meaning and value to it (Swanwick, 2009). Previous studies have explored citizen attitudes to grassland landscapes in relation to landscape preference, perceptions of cultural ES, and willingness to pay for ES, often in specific regions including mountains, and marginal or protected landscapes. Multiple factors have been found to affect peoples' perceptions and attitudes, including age, gender and education, as well as rural-urban residency (Martín-López *et al.*, 2012), and environmental interest (Schmitt *et al.*, 2021). However, there has been little consideration of the drivers that underpin positive or negative attitudes to PG across different socio-economic contexts, and climatic regions in Europe, including influences on behavioural intentions associated with PG. In this study, we aim to understand the drivers of citizen attitudes towards, and values associated with PG in five European countries (Spain, UK, Switzerland, Czech Republic and Sweden), representing five biogeographic regions, and a variety of PG types within each region, in order to make comparisons between different geographical and demographic populations.

Materials and methods

A total of 3,184 participants from the 5 European countries were quota sampled based on age, gender and socio-economic class, and rural versus urban residency. Participants were nationally representative of the population for each country, with 620 citizens surveyed via a 30-minute, self-directed online questionnaire, administered through a social research agency, in each country. We used a structured questionnaire (translated and back translated into local languages), with closed ended questions. Five-point agree-disagree Likert scales were used to measure attitudes and values towards: (1) threats to the countryside; (2) personal benefits from the countryside; (3) social trust; (4) management of the environment; (5) attitude to the environment (using the Environmental Attitude Inventory (EIA); Milfont and Duckitt, 2010); and (6) consumer attitudes towards PG products. Likert scales using priority measures were used to explore (7) attitudes towards ES, and the overall behavioural intention of consumers; to (8) buy products from PG; and (9) spend time in grasslands and meadows. Nine hypotheses (Figure 1) outlining the relationship between influential socio-economic factors, attitudes to PG, and behavioural intentions from a citizen and consumer perspective were tested by applying factor analysis, principal component analysis, followed by structural equation modelling (SEM) based on partial least squares approach (SEM-PLS).

Results and discussion

The full results of the SEM process (analysing the drivers of citizens' attitudes) will be reported in the final paper. Here we discuss some preliminary results. Across the whole survey, we found that there were significant differences between the responses of citizens in each country, indicating that there are different perceptions and attitudes to PG across Europe. Across the five countries the highest proportion of respondents indicated that they visit the countryside for recreation and leisure, with up to a quarter of

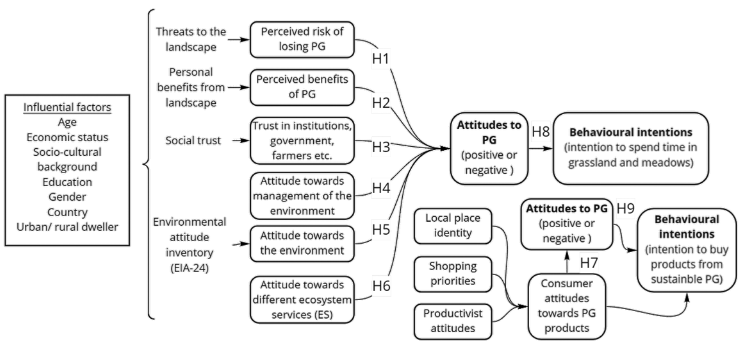

Figure 1. Conceptual model and hypotheses testing. H_1 – Perceived risk of losing PG will influence the attitudes of consumers and citizens towards PG. H_2 – More perceived benefits of PG will positively influence the attitudes of consumers and citizens towards PG. H_3 – Increased trust in institutions and government will positively influence attitudes to PG H_4 – More positive attitudes towards the management of the environment will positively influence attitudes to PG. H_5 – More positive attitudes to the environment will positively influence attitudes to PG. H_6 – More positive attitudes towards different ES will positively influence attitudes to PG. H_7 – More positive consumer attitudes towards PG products will positively influence their attitudes towards PG. H_8 – A positive attitude towards PG will lead to positive behavioural intention to buy products from PG. H_9 – A positive attitude towards PG will lead to a positive behavioural intention to spend time in grasslands and meadows.

respondents in each country saying they lived/ worked in the countryside (highest in Czech Republic, 26%), and between one-fifth and a quarter of respondents never visiting the countryside (except Spain, 37%). Respondents in the UK rated affective responses to grasslands linked to 'beauty' and 'enjoyableness' higher (more positive) than respondents from other countries. Czech respondents rated 'pleasantness' and 'goodness' higher than other countries, and Swedish respondents rated 'value' and 'interest' higher. Overall, we can infer that the majority of the respondents viewed grasslands and meadows as pleasant, good, valuable, interesting, beautiful and enjoyable. When asked about their intention to visit meadows and grassland, the majority of the respondents noted that they intend to spend time in grassland and think it is easy to spend time there. In terms of sustainability of food, respondents from Spain and Sweden noted that it is easy for them to identify sustainably produced food in general. However, they noted that it is less easy for them to buy sustainably produced food, and indeed is not a food purchasing priority. In general, the majority of respondents from all countries mentioned that trees were an important part of visiting the countryside (average 85% of respondents), followed by feeling happy, having a fascinating landscape, and a sense of peacefulness, and plenty to discover. The presence of meadows and pastures and open landscape were important when visiting the countryside for a slightly lower proportion of respondents (average 76 and 75%); however, varied landscapes were also seen as important (average 78% of respondents). The most likely problems in the countryside were attributed to bad behaviour by visitors (highest in the UK, 74%), lack of young farmers taking over farming (highest in Spain, 72%), and misuse of chemical fertilizers (highest in UK, 70%). Conversion of pasture or meadows to forest or woodland, and too many livestock causing damage to the land were of least concern (on average less than 35% agree they are a problem). The level of agreement differed between countries investigated but the greatest concern in terms of perceived risk was common across five countries. Our results so far show that affective responses of citizens are linked to non-monetary ES associated with PG. In our forthcoming analysis we will use a SEM to assess the relationships between drivers of attitudes, and attitudes, as well as behavioural intentions, for: (1) citizen attitudes towards ES associated with PG; and (2) consumer attitudes to the products of PG.

Conclusions

This study gives an insight into the drivers behind citizen and consumer attitudes to PG, including in relation to behavioural intentions to spend time in, and buy products from PG. Results will be relevant for using value and perceptions of PG as a basis for current and future public policy and practice design.

References

Martín-López B., Iniesta-Arandia I., García-Llorente M., Palomo I., Casado-Arzuaga I., Amo D.G.D., ... and Montes C. (2012) Uncovering ecosystem service bundles through social preferences. *PLoS ONE* 7(6), e38970.

Milfont T.L. and Duckitt J. (2010) The environmental attitudes inventory: A valid and reliable measure to assess the structure of environmental attitudes. *Journal of Environmental Psychology* 30(1), 80-94.

Schmitt T.M., Martín-López B., Kaim A., Früh-Müller A. and Koellner T. (2021) Ecosystem services from pre-alpine grasslands: Matches and mismatches between citizens' perceived suitability and farmers' management considerations. *Ecosystem Services* 49, 101284.

Swanswick C. (2009) Society's attitudes to and preferences for land and landscape. *Land Use Policy* 26, S62-75.

Are we talking about the same thing? Stakeholder perspectives on grassland management intensity

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Abstract

Grassland management crucially influences the delivery of ecosystem services from permanent grasslands. Variability in management practices is often described along a gradient from 'low intensity' to 'high intensity'. These terms are likely to carry different meanings across European regions that differ in environmental and socio-economic conditions as well as between different groups of stakeholders. We conducted an online survey among grassland stakeholders asking them to characterise what they consider as 'low', 'intermediate' and 'high'-intensity management in terms of cutting frequency, grazing intensity, and nitrogen fertilization. The answers of the 125 respondents revealed high variability in the thresholds between management intensity levels. Professional background ('agriculture' vs 'ecology/ conservation') explained only a small percentage of the variability. The biogeographical region on which the respondents' expertise was based also influenced the evaluation of management practices. Our survey exposed the hidden problem of communicating about grassland management across regions and professional backgrounds, and identifies a need for a common terminology when making general recommendations for sustainable grassland management.

Keywords: biogeographical regions, management intensity, permanent grassland

Introduction

European permanent grasslands differ greatly in their contribution to agricultural production, biodiversity conservation and other ecosystem services. This variability is strongly related to differences in management, often described as a gradient from 'low intensity' to 'high intensity'. While these terms are widely used as a shorthand when communicating about permanent grasslands, they are likely to carry different meanings across biogeographical regions differing in environmental or socio-economic conditions as well as between different groups of stakeholders. To quantify the agreement or disagreement in interpreting management intensity levels, we surveyed stakeholders, asking them to characterize their understanding of 'low', 'intermediate' and 'high' management intensity of European permanent grasslands. We expected answers to show high variability and to be influenced by the professional and geographic background of the stakeholders.

Materials and methods

We conducted an online survey from June – December 2019, recruiting respondents through the professional networks of partners within the European Union (EU) H2020 project 'SUPER-*G*'. Respondents characterized what they considered typical of 'low', 'intermediate' and 'high' management intensity of permanent grasslands by providing lower and upper thresholds for three management practices: (1) cutting frequency in grassland that is exclusively mown, not grazed (numbers of cuts per year); (2) grazing intensity in grassland that is exclusively grazed, not mown (livestock-unit (LU) grazing days per hectare and year, one LU corresponding to 500 kg live weight); and (3) rate of total nitrogen (N) fertilizer application, including mineral or organic fertilizers and animal excreta during grazing. As ranges were allowed to overlap, we calculated two threshold values for each respondent and management practice as the means of the given ranges: one between low and intermediate and one between intermediate and high management intensity.

Respondents were also asked to specify the country or countries on which their expertise in grassland management was based and whether their professional background was in 'agriculture', 'ecology/conservation' or in both. For each management practice and threshold, we used a linear regression model to test the effect of professional background, parameterised as a factor with three levels. Furthermore, we assigned respondents' countries of expertise to one or more of six biogeographical regions and calculated means and standard deviations for the six management intensity thresholds separately for each region represented.

Results and discussion

A total of 125 respondents from 26 countries answered the survey, with the numbers of answers differing among cutting frequency (n=123), grazing intensity (n=84) and nitrogen fertilization (n=82). Of the respondents, 67 specified their background as 'agriculture', 36 as 'ecology/conservation' and 16 as both. The mean thresholds between 'low' and 'intermediate' management intensities were 1.9 annual cuts, 238 LU grazing days ha^{-1} y^{-1} and 63 kg N ha^{-1} total N fertilization. The corresponding mean thresholds between 'intermediate' and 'high' management intensities were 3.2 cuts, 467 LU grazing days ha^{-1} y^{-1} and 149 kg ha^{-1} total N fertilization. In all cases, individual answers varied widely around these means (Figure 1).

A professional background in 'agriculture' vs 'ecology/conservation' significantly influenced both thresholds for cutting frequency and the low/intermediate threshold for N fertilization, with a background in 'ecology/conservation' being associated with slightly lower thresholds than one in 'agriculture' (Figure 1). This background, however, explained no more than 13.4% of the variance observed. Geographical background also appeared to influence responses (Figure 2). Mean thresholds were generally lowest for respondents from the Mediterranean, Pannonian, and Boreal biogeographical regions, where summer drought or long winters restrict vegetation season length and productivity. Standardizing management intensity categories by growing conditions might address this source of variability but holds its own methodological challenges.

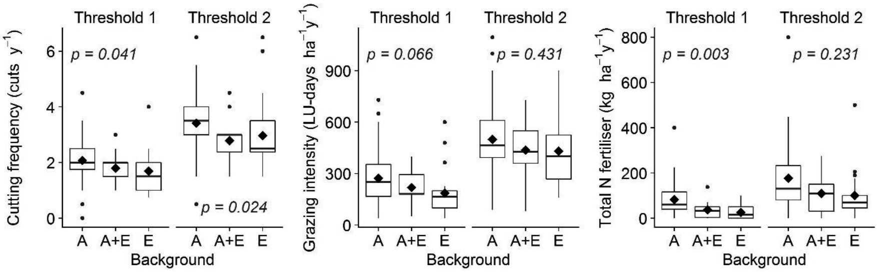

Figure 1. Thresholds between low and intermediate (threshold 1) and between intermediate and high (threshold 2) management intensity of permanent grassland, depending on respondents' background (A: 'agriculture' and/or E: 'ecology/conservation').

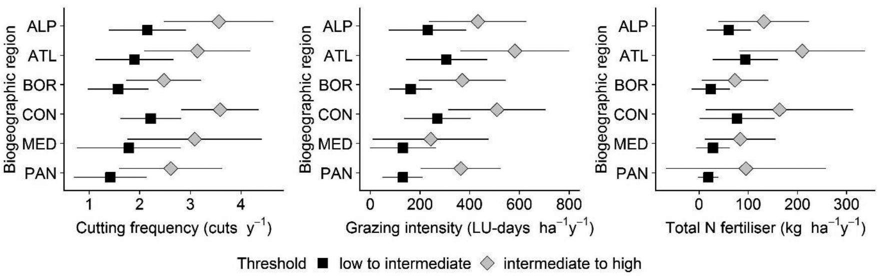

Figure 2. Thresholds between low and intermediate as well as between intermediate and high management intensity of permanent grassland, depending on the biogeographical regions of the respondents. Means ± standard deviation. ALP: Alpine, ATL: Atlantic, BOR: Boreal, CON: Continental, MED: Mediterranean, PAN: Pannonian.

Few alternatives for characterizing agriculturally managed grasslands across Europe exist, besides attempts to calculate continuous gradients in land-use intensity (Blüthgen *et al.*, 2012). Terms such as 'semi-natural' or 'unimproved' versus 'improved' grasslands have sometimes been defined more stringently (e.g. Peeters *et al.,* 2014), but still suffer from similar ambiguities in meaning and thresholds. The EU EUNIS habitat classification (EEA, 2021), which is based on phytosociological classification, distinguishes only two subclasses ('dry or moist' and 'wet' within the class of 'Agriculturally improved grasslands' (Code V31), which represents most agriculturally managed permanent grasslands in Europe.

Conclusions

Our survey revealed that the terms 'low-intensity' and 'high-intensity' grassland management carried widely differing meanings among European stakeholders, making them insufficient for communicating across regions and stakeholder groups. It is thus important to acknowledge this difficulty when communicating about European permanent grasslands. Concerted actions to refine existing terminologies could facilitate knowledge transfer about permanent grassland among European stakeholders.

Acknowledgements

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References

Blüthgen N., Dormann C.F., Prati D.A., Klaus V.H., Kleinebecker T., Hölzel N., ... and Weisser W. (2012) A quantitative index of land-use intensity in grasslands: Integrating mowing, grazing and fertilization. *Basic and Applied Ecology* 12, 207-220.

EEA (2002) Europe's biodiversity – biogeographical regions and seas. *EEA Report* 1/2002.

EEA (2021) *EUNIS habitat classification*. Available at: https://www.eea.europa.eu/data-and-maps/data/eunis-habitat-classification-1.

Peeters A., Beaufoy G., Canals R.M., De Vliegher A., Huyghe C., Isselstein J., ... and Wilkins R.J. (2014) Grassland term definitions and classifications adapted to the diversity of European grassland-based systems. *Grassland Science in Europe* 19, 743-750.

The influence of circular agriculture on the financial performance of dairy farms in the Netherlands

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Abstract

Circular agriculture is a solution to the depletion of soil, water and raw materials and the increasing global temperatures. The objective of this study was to generate insight into the influence of circular agriculture on the financial performance of dairy farms. This insight can guide dairy farm management. Data from 238 Dutch dairy farmers were analysed with a linear regression, t-test and MANOVA. Circular farms had a higher margin than non-circular farms. Livestock sales, concentrate costs and transport costs were the main influencing factors. For all farms, a positive relationship was found between grazing and the margin, and between protein autonomy and the margin. A negative relationship was found between CO_2 emissions and the margin. Circular agriculture combines environmental and financial benefits by practising grazing, by optimizing the amount of concentrates fed as well as optimizing N and P-use efficiency at farm level.

Keywords: costs, circular dairy farming, financial performance, margin, sustainable agriculture

Introduction

In 2018, the Dutch Ministry of Agriculture, Nature and Food Quality (LNV), published a vision, 'Valuable and Connected', on transition to circular agriculture under the expectation that this transition would instigate more sustainable use of raw materials and meet society's desire for sustainable dairy farming. Stuiver and Verhoeven (2010) defined circular agriculture as the optimization of production with selective use of external inputs, long-term income generation and respect for natural systems. The transition to circular agriculture is hampered by legislation and regulations and an unclear revenue model (Maij *et al.*, 2019). Successful transition is expected to have a positive impact on the environment and society, but is it is important for farmers to know whether it is financially sound to proceed with the transition to circular agriculture. In addition, understanding which factors influence the financial performance can help improve farm management. The objective of this study is to generate insight into the influence of circular agriculture on the financial performance of dairy farms.

Materials and methods

This study used data from 238 anonymous Dutch dairy farms, all of which are clients of Dirksen Management Support (DMS) and mainly located in the centre of the Netherlands. The dataset contained the annual accountancy report and the Annual Nutrient Cycle Assessment (ANCA, Dutch: Kringloopwijzer) for 2019. The Life Cycle Analysis (LCA) regulations and the Product Environmental Foodprint Category Rules (PEFCR) apply to all calculations of the ANCA (Van Dijk *et al.*, 2019). This study defined circular agriculture based on the vision statement of the Ministry of Nature, Agriculture and Food Quality (2018), operationalized with values of the Milieukeur Foundation (SMK) (2020). SMK is a certification institute that develops, manages and tests sustainability criteria. The farms were divided into a circular and non-circular group based on the criteria of Table 1 that can be found in the ANCA. Only farms that complied with all the requirements of Table 1 were selected as circular farms.

The data were analysed with the programme RStudio version 3.6.2. Before the analysis, the data were checked for appropriateness given the type of analysis. A multiple linear regression provided insight into the

Table 1. Technical aspects defining circular farms.[1]

Technical aspects of circular farms	
Grazing	Yes
Protein autonomy (%)	≥50
CO_2 emission (g kg of milk^{-1})	≤1199
N soil surplus (kg ha^{-1})	≤150
Permanent grassland (% of farm area)	≥40
Renewable energy	Yes
Natural vegetation (% of farm area)	≥5
NH_3 emission (kg ha^{-1})	≤80

[1] All technical aspects have been adopted from the vision statement of the Ministry of Nature, Agriculture and Food Quality (2018) and the Milieukeur Foundation (2020).

Table 2. Fifteen independent financial parameters defining the financial performance of the farms.[1]

Financial parameters (€ 100 kg^{-1} FPCM)	
Milk sales	(Hire of) machinery
Livestock sales	Transport (fuel) costs
Other revenues	Livestock costs
Silage costs	Labour costs
Concentrate costs	Other costs
Fertilizer costs	Overhead
Crop protection costs	Margin
Purchased seed	

[1] The financial parameters are derived from Chen and Holden (2018) and March *et al.* (2017).

relationship between the margin and the technical aspects; for this analysis no division was made between circular and non-circular farms. After the multiple linear regression, the farms were divided into two groups. Farmers do not always correctly fill in the proportion of natural vegetation in the ANCA, as it is difficult to register and has little added value for farmers. The other parameters are considered to be reliable. As natural vegetation was therefore unlikely to be a reliable selection criterion, a population Y1 (of which circular farms n=9, non-circular farms n=229) with natural vegetation selection, and a population Y2 (of which circular farms n=39, non-circular farms n=199) without natural vegetation selection were made. A t-test provided insight into whether there was a difference between the margins of circular farms and non-circular farms for both populations. A Wilcoxon rank sum test with continuity correction was performed to reduce the effect of outliers. To substantiate any differences in the margins between circular and non-circular farms, a MANOVA was carried out with the parameters described in Table 2.

Results and discussion

The relationships between the margin and the technical aspects presented in Table 2 are shown in Table 3. Y1 showed no difference in the margin between circular and non-circular farms. When natural vegetation was not included as a selection criterion (Y2), there was a difference between the margin of circular and non-circular farms (P=0.006, Wilcoxon rank-sum test is performed). A MANOVA provided insight into which financial parameters contributed to the difference in the margin. For Y2, livestock sales (P=0.05), concentrate costs (P=0.003) and transport costs (P=0.05) contributed to differences in the margin. The study of Ma *et al.* (2022) also showed that lower feed costs and young livestock costs contribute to higher net profits in cooperative crop-livestock systems.

In this study, it is expected that feeding less concentrate contributes to the correlation between lower CO_2 emissions and a higher margin, since the amount of concentrate fed contributes largely to the amount of CO_2 emissions in the calculation methodology of the ANCA (Van Dijk *et al.* (2019). Circular farms showed management with a high milk production (>10,000 kg fat and protein corrected milk

Table 3. Multiple linear regression analysis between the margin and the technical aspects.[1]

Variable	Estimate	*T*-value	*P*-value
Intercept	1.002	0.261	0.794
Margin and grazing	0.013	2.554	0.011*
Margin and protein autonomy (%)	0.058	2.018	0.045*
Margin and CO_2 emissions (g kg milk^{-1})	-0.006	-2.380	0.018*
Margin and N soil surplus (kg ha^{-1})	0.002	0.340	0.735
Margin and permanent grassland (%)	-0.007	-0.554	0.580
Margin and renewable energy	0.007	0.895	0.372
Margin and natural vegetation (%)	0.009	0.169	0.866
Margin and NH_3 emission (kg ha^{-1})	0.022	0.821	0.412

[1] Multiple R^2=0.072, Adjusted R^2=0.040. * = (P<0.05).

(FPCM)) and higher N- and P-use efficiency at farm level than non-circular farms. This study analysed the data based on the definition of circular agriculture given by the Dutch government. The results are influenced by these selection criteria and the corrections that were carried out for (non-)circular farms. The results of this study apply to the Dutch definition of circular agriculture only. The research showed no difference in the margins when natural vegetation was included as a selection criterion. It should be noted that the size of the population of Y1 (n=9) made it more difficult to demonstrate effects. Many studies assume that farmers strive for maximum profit. However, they may be motivated by other aspects, for example the recognition of other farmers or animal welfare (Kristensen and Jakobsen, 2011).

Conclusions

Circular agriculture combines environmental and financial benefits by practising grazing, by optimizing the amount of concentrates fed, optimizing N- and P-use efficiency at farm level, as well as increasing farm efficiency (maximum output with optimal input and minimum waste). Circular agriculture results in a higher margin (selection on natural vegetation not taken into account) and contributes to the financial performance of Dutch dairy farms.

References

Chen W. and Holden N.M. (2018) Bridging environmental and financial costs of dairy production: A case study of Irish agricultural policy. *Science of the Total Environment* 615, 597-601.

Kristensen E. and Jakobsen E.B. (2011) Challenging the myth of the irrational dairy farmer; understanding decision-making related to herd health. *New Zealand Veterinary Journal* 59, 1-7.

Ma Y., Hou Y., Dong P., Velthof G.L., Long W., Ma L., Ma W., Jiang R. and Oenema O. (2022) Cooperation between specialized livestock and crop farms can reduce environmental footprints and increase net profits in livestock production. *Journal of Environmental Management* 302 Part A, 113960.

Maij H., Baarsma B., Koen, C., Van Dijk G., Van Trijp H., Volberda H. ... and Thus S. (2019) *Goed boeren kunnen boeren niet alleen*. Ministerie van Landbouw, Natuur en Voedselveiligheid, Den Haag, the Netherlands, 64 pp.

March M.D., Shalloo L., Roberts, D.J. and Ryan W. (2017) Financial evaluation of Holstein Friesian strains within composite and housed UK dairy systems. *Livestock Science* 200, 14-22.

Milieukeur Foundation (2020) *Certificatieschema melk.* Stichting Milieukeur (PlanetProof), Den Haag, the Netherlands, 34 pp.

Ministry of Nature, Agriculture and Food Quality (2018) *Agriculture, Nature and Food: Valuable and Connected.* Rijksoverheid, Den Haag, NL, pg. 21.

Stuiver M. and Verhoeven F. (2010) *Kringlooplandbouw, op weg naar geborgde bedrijfsspecifieke milieuresultaten.* Alterra Wageningen UR, Wageningen, the Netherlands, 18 pp.

Van Dijk W., Schröder J.J., Sebek L.B., Oenema J., Conijn J.G., Vellinga Th., ... and Verloop K. (2019) *Rekenregels van de KringloopWijzer 2019.* Wageningen University & Research, Wageningen, the Netherlands, 134 pp.

Theme 2.
Bundles of services provided by grasslands

Ecosystem services provided by semi-natural and improved grasslands – synergies, trade-offs and bundles

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Abstract

Although grasslands have an extensive global coverage and are important contributors to fodder production, they have received less attention as providers of multiple ecosystem services (ES). In this paper, we investigate the utility of the ES framework for grassland management in Europe. We focus on semi-natural grasslands (SNG) and improved grasslands (IG); both are widespread in European agriculture. We present an overview of the ES delivered by these two grassland types and their potential synergies, trade-offs and bundles. We show that SNG are able to generate a wider range of ES than IG, and that trade-offs between ES exist in both grassland types. For example, SNG are good in providing habitat for biodiversity, pollination, biological control and cultural services, but are poorer in biomass production and for increasing water infiltration, whereas IG produce higher quantities of biomass for fodder but contribute less to cultural services. Both IG and SNG are likely needed for the long-term sustainability of food production, but a larger effort towards landscape-scale management is needed to balance the provision of ES. Applying the ES concept to grasslands in farming systems could be valuable if used in an informed way, leveraging ecologically and economically grassland management for sustainable livestock farming systems in Europe.

Keywords: biodiversity, multifunctionality, stakeholders, supply and demand, sustainable farming

Introduction

Grasslands cover about 40% of the Earth's terrestrial surface and represent about 65% of the world's agricultural land area (Dengler *et al.*, 2020). Compared to forests, for example, grasslands have received much less attention in the multiple ecosystem service (ES) framework (Bargett *et al.*, 2021; Diaz *et al.*, 2015). This is unfortunate, as grasslands are important for food production and extensively managed grasslands also contribute to the maintenance of high biodiversity and key ecological processes (e.g. pollination or water regulation) at local and landscape scales (Dengler *et al.*, 2020). They also have outstanding cultural values, e.g. as legacies of ancient land use systems, their beauty (Plieninger *et al.*, 2015), and they are classified as 'high natural value habitats' by the EU.

As recently as just 100 years ago most European livestock still grazed semi-natural grassland within multifunctional and high natural and cultural value pasture systems (Hartel *et al.*, 2018, Velado-Alonso *et al.*, 2020), whereas currently a large part of the livestock production occurs on technologically improved, monofunctional grasslands or croplands (Naylor *et al.*, 2005). As a consequence, European grasslands went through a sharp decline during the last century (Bargett *et al.*, 2021). On the other hand, the lack of management and abandonment of grasslands that are still left may pose a major threat to people worldwide, especially those societies which rely directly on the multiple ES of grasslands such as food, fuel, fibre and medicinal products, as well as their multiple cultural values (Bengtsson *et al.*, 2019).

Three main types of grasslands can be differentiated within agricultural production systems: natural, semi-natural, and agronomically improved grasslands (Bengtsson *et al.*, 2019; Bullock *et al.*, 2011; Lemaire *et al.*, 2011; Peeters *et al.*, 2014). Natural grasslands form the grassland biomes and are natural areas mainly created by processes related to fire and wildlife grazing (Bengtsson *et al.*, 2019), but are also used by livestock. Semi-natural grasslands (SNG) are pastures with a long-term history of traditional management (Dengler *et al.*, 2020). In Europe, SNG represent an important component of the cultural and natural heritage. Within the EU`s Common Agricultural Policy they are recognized as High Nature Value farmlands and are listed as Annex I habitats in the Habitats Directive. SNG typically require livestock grazing as well as a certain degree of direct human management (e.g. scrub control, mowing) for their maintenance and will generally be encroached by shrubs and trees if abandoned (Queiroz *et al.*, 2014). Improved grasslands (IG) are pastures resulting from ploughing, i.e. former arable fields, and sowing agricultural varieties or non-native grasses with high production potential. The term 'improved' refers to modern, technological capital-intensive management, including artificial fertilization, monoculture and/or high density of livestock (Pilgrim *et al.*, 2010). There is no clear threshold between SNG and IG, and with time an IG may become a SNG depending on, e.g. nutrient status, humidity, availability of typical grassland species in the seed bank or in the surrounding landscape, and management (Dengler *et al.*, 2020). Due to the great variety of grassland types and their environmental, ecological, historical, technological and socio-cultural contexts within which they evolved, identifying universal ES supply and ES demand patterns is important and challenging, yet still largely unaddressed (e.g. Bengtsson *et al.*, 2019; Hartel *et al.*, 2018; Herzon *et al.*, 2022).

The aim of this paper is to show the utility of the ES framework for sustainable grassland management in Europe. We start by presenting the ES framework related to farmland management. Then we present an overview of the individual or bundles of ES delivered by European grasslands. We focus on semi-natural grasslands (SNG) and improved grasslands (IG), which are the two major contributors to European (Figure 1). Through a literature survey, as well as our own research experience, we selected the most common ES related to grassland and analysed the synergies, trade-offs and bundles in the SNG and IG types. We end by suggesting future research directions and how managing ES in an informed way may increase the sustainability of future livestock farming systems in Europe.

The ecosystem service framework

Ecosystem services are the goods and the direct and indirect benefits people obtain from ecosystems (Haines-Young and Potschin, 2018; MA, 2005). The concept of ES, nonetheless, still generates controversy in the research community (Richter *et al.*, 2021), and the frameworks adopted, e.g. MA (2005) or CICES (see Haines-Young and Potschin, 2018) and/or the assessment approaches can differ among users (Richter *et al.*, 2021). Despite these fundamental discussions, the scientific community has intensively delved into the associations between different ES and the ecosystem features such as biodiversity, ecosystem functions

A B

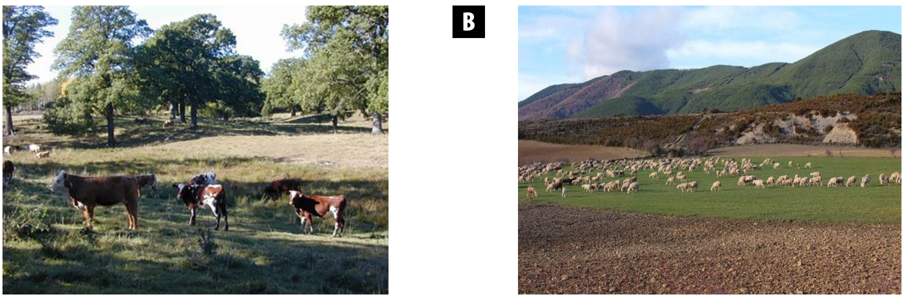

Figure 1. Illustration of an (A) semi-natural grassland (SNG) in Sweden, and (B) an improved grassland (IG) in Spain.

and structures, but also in society as a tool to inform decision-making for sustainable management (Saidi and Spray, 2018). Many studies have focused on the assessment of individual ES, on trade-offs (antagonistic relationship between two or several ES) and synergies (synergetic relationship between two or several ES) across ES, or on identifying and analysing associations among multiple services, referred to as bundles of ES. A bundle of services is understood as a set of services that occur together across space and time. Bundles of services are often sought for in decision-making, because they could improve the management actions to favour as many ES as possible (e.g. Raudsepp-Hearne *et al.*, 2010).

The association among services may, however, vary depending on the ecosystem type and its use. In intensively managed agricultural systems, the increase of provisioning services (food and meat production, timber production) often occurs at the cost of regulating services (e.g. water cycle regulation, soil fertility, carbon sequestration, biological control) and supporting services (biodiversity) (Foley *et al.*, 2005; MA, 2005). Both spatial and temporal aspects are crucial for understanding ES generation. This scale dependence will hence affect the trade-offs, synergies and bundles of services as they are generated at different scales (Kleijn *et al.*, 2011). Although it is clear that the flow of ES is not static, this has gained little attention, both in science and in practice (Rau *et al.*, 2020).

To understand the ES value of grasslands both the supply (the capacity of an ecosystem to produce a service) and demand (the amount and type of services demanded by people, but also considering the potential future demands) should be acknowledged (Lamarque *et al.*, 2011). Still, most studies of ES do not distinguish between the ES supply and demand (Yahdjian *et al.*, 2015), nor aim to address ES mismatches. The benefits (welfare gains) of a particular ecosystem service largely depends on how different actors in society perceive or attach value to that ecosystem service, which could originate conflicts between stakeholders with different interests.

Grasslands can generate a wide diversity of ES (e.g. D'Ottavio *et al.*, 2016; Bengtsson *et al.*, 2019; Sollenberger *et al.*, 2019; Zhao *et al.*, 2020), of which we selected the twelve most relevant for IG and SNG to be included in the literature survey. The selection of ES was based on the following criteria: (1) to capture key examples of provisioning, regulating and cultural ES categories, and (2) to represent the different biocultural regions of Europe. Supporting services have often been excluded from ES assessments (Khan, 2020; Haines-Young and Potschin, 2018; Price, 2014) and were therefore not considered in this paper. Additionally, biodiversity is discussed in relation to the regulating service habitat provision.

ES generated from semi-natural and improved grasslands

There was a clear difference in provision of ES between SNG and IG, where SNG showed a higher potential to generate most of the services at relatively high levels, compared to IG (Table 1). Some ES, like water-related ES and carbon storage and sequestration, have been frequently studied, especially for IG, but not much within the ES framework (Sollenberger *et al.*, 2019). Information about the generation of pollination and biological control was especially poor for IG. It was clear that ES generation depends on size of grassland, scale of analysis and the landscape context in which the grassland is located.

Most of the indicators within the ES framework assessed in this literature overview concerned regulating services and less among cultural and provisioning services (Table 1). Biomass production was found to be higher in IG than in SNG, although differences occurred depending on location (Sollenberger *et al.*, 2019). Grassland productivity is generally increased by technological innovations, such as irrigation and fertilization, and is linked to the intensity of use like frequency of cuts and livestock unit per area. Since IG often are former arable fields, biomass production is generally much higher due to fertilization. The ability for SNG to act as habitat providers, especially to support high biodiversity, is well documented, although differences occur depending on local biophysical conditions (Dengler *et al.*, 2020 Kok *et al.*,

Table 1. Most important ecosystem services generated from improved (IG) and semi-natural grasslands (SNG).[1]

Ecosystem services	Confidence term	Comments	Reference
Plant biomass production (Fodder production)	WE	Generally higher production in IG than in SNG	Zisenis *et al.*, 2011
Wild products	EI	SNG are better providers than IG, mostly due to historical ecological knowledge and values	Sucholas *et al.*, 2017; Torralba *et al.*, 2018; Vári *et al.*, 2020
Habitat provision (maintaining nursery population and habitats)	WE, IC	SNG are better providers, but few studies are conducted in IG	Dengler *et al.*, 2020; Wilson *et al.*, 2012
Pollination (pollination of crops and wildflowers)	WE, IC	Few studies directly relate SNG and IG to crop production. SNG important for pollination in the landscape	Werling *et al.*, 2014; Taki *et al.*, 2010
Biological control (pest control for increase crop production)	EI	Few studies directly relate SNG and IG to crop production	Jonsson *et al.*, 2014
Carbon capture (carbon sequestration through photosynthesis)	IC, UR	Carbon capture is generally higher in IG, but results are inconclusive and site dependent	Sollenberger *et al.*, 2019; Chang *et al.*, 2021
Carbon storage (Carbon sink in the soil)	WE	Carbon storage is higher in SNG	Dlamini *et al.*, 2016; Sollenberger *et al.*, 2019
Erosion control (reducing run-off and stabilizing soil)	IC	Long-term permanent vegetation in SNG may prevent run-off and stabilization of soils, in contrast to IG	Pligrim *et al.*, 2010; Fu *et al.*, 2011
Water quantity (infiltration and storage capacity)	UR	Potentially important but site dependent	Sollenberger *et al.*, 2019; Posthumus *et al.*, 2010; Guo *et al.*, 2020
Water quality (cleaning water through infiltration)	EI, IC	Potentially provided by SNG but could be decreased in IG	Cadman *et al.*, 2013; Sollenberger *et al.*, 2019
Tourism/recreation (possibilities for recreation)	EI, IC	Clearly linked to high levels of biodiversity and multifunctionality of SNG, but less clear with IG	Hönigova *et al.*, 2012; Martino and Muenzel, 2018
Cultural heritage (historical activities, legacies and biological values)	WE	Cultural heritage is highly related to SNG but not to IG	Fischer *et al.*, 2008; Lindborg *et al.*, 2008; Bullock *et al.*, 2011

[1] Provisioning services (grey), regulating services (light grey) and cultural services (white). The confidence terms listed are based on the four-box model for qualitative communication according to IPBES procedure (IPBES, 2018). WE = well established; EI = established but incomplete; UR = unresolved; IC = inconclusive.

2020a). They are also important sources for maintaining and dispersing organisms at the landscape scale (Dengler *et al.*, 2020). Improved grasslands, having higher nutrient levels and high yielding varieties, do not reach the same high level of biodiversity. That may, however, change with time if IG soils lose nutrients as they become permanent (Sexton and Emery, 2020). Sowing of specific forbs or legumes (often occurring in SNG) could also increase biodiversity significantly without jeopardizing forage quality or ruminant heath (Hamacher *et al.*, 2021).

Pollination and biological control generated in SNG may have a direct positive effect on the overall richness of insects and also on agricultural production adjacent to the grassland (Jonsson *et al.*, 2014; Taki *et al.*, 2010; Werling *et al.*, 2014). Some studies show a positive effect of SNG in the landscape on the number of pollinators, and in contrast, a decline in plants that rely on them may occur if pollinator populations decline (Potts *et al.*, 2010). However, Wood *et al.* (2013) showed that although abundance of flowering plants was similar between legumes and forbs, the abundance and diversity of pollinator groups were greater in legumes than in forbs. It should be noted that rare pollinator species were nonetheless found in the wildflower habitat only.

The role of grasslands for climate change mitigation is frequently discussed, especially carbon storage in permanent grassland, as they may store large amounts in the soil (Lal, 2004; Smith, 2014; Susanna *et al.*, 2010). Although, carbon sequestration increases by increased nutrient inputs (e.g. Kätterer *et al.*, 2012; Sollenberger *et al.*, 2019; but see Skinner, 2013), carbon capture and storage in grassland soils is reduced by intensive grazing (Dlamini *et al.*, 2016). Natural grasslands converted into improved sown grasslands may increase the organic C sequestration, whereas conversion into silvopasture increased the mineral-associated C fraction (Adewopo *et al.*, 2015). This suggests that management intensification promotes soil C sequestration, and integration of trees improves soil C stability. Hence, carbon storage is concluded to be higher in SNG than in IG, whereas carbon capture capacity can be higher in IG. However, if IG are ploughed, large amounts of C are released. While storage capacity is high after conversion of arable land to grasslands (Franzluebbers and Stuedemann, 2009), that capacity is limited in time until soil reaches a saturation limit (Smith, 2014). One way to increase the C stock in IG grassland soils is to reduce grazing pressure as the reduction of carbon by grazing is only partly compensated for by addition by mineral fertilizers (Eze *et al.*, 2018). This will also reduce greenhouse gas emissions generated from livestock grazing in grassland (Manzana and White, 2019).

The water-related ES is currently of minor importance in European grasslands, but this has a potential to increase (Lamarque *et al.*, 2011). In general, water quality is negatively affected by fertilizers (increased N and P) and high stocking rates increasing the nutrient load in runoff (Sollenberger *et al.*, 2019). This suggests that IG, with high amounts of nutrients in the soil and high stocking rates, will have a negative impact on water quality. The water quality in SNG is generally thought to be better and could be improved in certain locations where water capture and increased infiltration could mitigate negative effects from heavy rains (Cadman *et al.*, 2013). Floodplain meadows and coastal grasslands, although both are becoming increasingly rare SNG types in Europe, are highly relevant in regulating floods and mitigating the negative impacts by rivers and sea. Coastal grasslands mitigate the impacts of sea-level rise and buffer against intensified coastal erosion processes (Posthumus *et al.*, 2010). Permanent grasslands also contribute greatly in mitigating erosion (Pligrim *et al.*, 2010), especially in comparison with cropland (Cerdan *et al.*, 2010). Since permanent vegetation is very efficient in preventing run-off and stabilizing soils, SNG are, in general, more efficient in erosion control than IG with a shorter history of vegetation establishment.

Although many SNG are protected for their high cultural and biological values, recreational activities are often related to the broader landscape, making it difficult to separate the role of semi-natural grasslands from that of improved grassland (Bullock *et al.*, 2011) and the overall heterogeneity of the landscape. In contrast to IG, SNG are appreciated for their cultural heritage linked to extensive use and traditional management (Fischer *et al.*, 2008; Lindborg *et al.*, 2008). Many are located on ancient sacred places such as burial mounds and have been kept open by livestock for thousands of years (Lindborg *et al.*, 2008). Traditional management of grasslands in terms of hay-making has played an important role in social cohesion among villagers and still does (Stenseke 2009). Wild products (e.g. mushrooms, berries, medicinal plants, fruits from shrubs and trees) from SNG were highly appreciated in the past across Europe (Vari *et al.*, 2020). Nowadays, the importance of wild products is declining, partly due to formal regulations of SNG, and partly due to changes in society, culture and values, resulting in less of these products on the market.

Trade-offs and synergies between ES and bundles of ES

Clear trade-offs and synergies could be detected in both SNG and IG (Figure 2). For example, SNG are good in providing wild food, habitat for biodiversity, pollination, biological control and cultural services, but are relatively poor in capturing carbon and improving water quantity and quality (Figure 2). In comparison, IG produce higher quantities of biomass and contribute less to pollination and carbon capture. The trade-off between biomass production and natural habitat provision (enhancing biodiversity) is established as a fundamental trade-off (Foley *et al.*, 2005, Kok *et al.*, 2021b). Experimental studies have shown that an increased number of plants may increase primary production. In an experiment including herbaceous grassland species richness it was found that annual soil C storage increased as the number of species increased (Tilman *et al.*, 2006). However, such experiments are difficult to translate into real-world systems as they only include a relatively low number of species (1-16 species), whereas species richness in SNG can be as high as 60 plant species in one square meter (Wilson *et al.*, 2012). In general, the relationship between species richness and soil C accumulation was inconsistent in the published literature, although greater biodiversity increases resistance to disturbance by stabilizing grassland productivity and productivity-dependent ES (Isbell *et al.*, 2015).

Bundles of ES from SNG were analysed by Bengtsson *et al.*, (2019) who detected three bundles: one dominated by water ES and also including plant biomass production, a second comprised of a number of cultural ES connected to livestock production, and a third bundle consisted of the regulating services pollination and biological control, indirectly linked to biodiversity. This shows that bundles are formed across the classical categories by MA (2005), and are related to physical geography of the SNG, the indicators representing and the ecological functions underpinning the ES. Although not statistically analysed, somewhat similar trends were also noted in our overview, where habitat provision, pollination, biocontrol, cultural heritage and soil storage in SNG could form a bundle, as a result of historical management (e.g. Herzog *et al.*, 2022). Potential bundles for IG were not equally clear, although biomass production and carbon capture are clearly linked. The ES found within each bundle are likely to be suitable to be managed together.

Supply and demand of ecosystem services

Research has shown that the extent to which ES translates into social wellbeing depends on the policies, formal and informal institutions, and power relationships within the local community (determines the access to ES), as well as individually held values (Horcea-Milcu *et al.*, 2016). Several tensions and conflicts around ES at the level of the society and management-induced changes can be associated with the above-mentioned factors (Bernues *et al.*, 2016). For example, in Romania social tensions arise

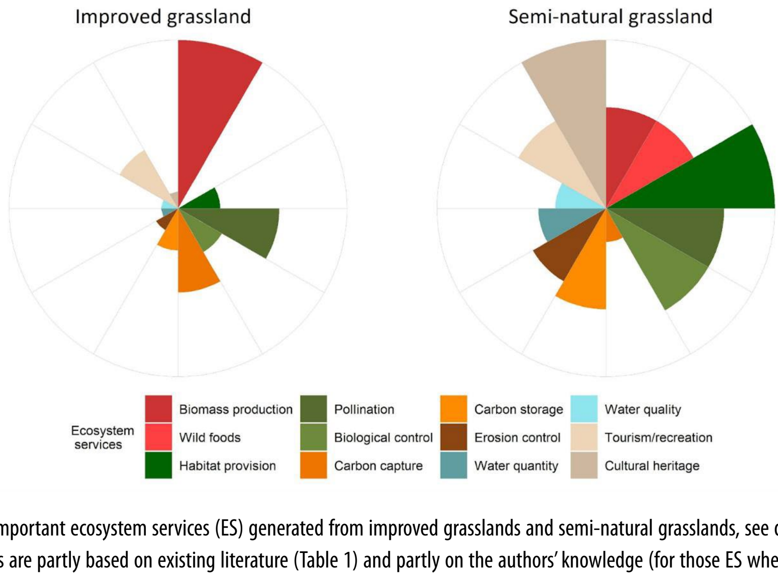

Figure 2. Most important ecosystem services (ES) generated from improved grasslands and semi-natural grasslands, see definition Table 1. The ES estimates are partly based on existing literature (Table 1) and partly on the authors' knowledge (for those ES where the confidence term is low, Table 1).

around the overgrazing of pastures and the illegal access of shepherds to SNG used as hay fields. In Sweden, stone walls are important cultural components of SNG that are perceived as barriers towards increasing the farming economy, mitigating intensification (Stenseke *et al.*, 2009). Furthermore, the loss of wood-pasture systems across Europe is related to policy misfit, change of value systems at the level of the society related to ES, and the spatial dynamic of human population and infrastructure (Plieninger *et al.*, 2015).

Differences in the knowledge and perception of ES may also generate conflicts among stakeholders regarding preference for ES (Bernués *et al.*, 2016, Dingkuhn *et al.*, 2020) and those discrepancies may exist between the demands and actual supply of ES (Dingkuhn *et al.*, 2020). For instance, Bernués *et al.* (2016) showed that farmers have a greater knowledge about ES than non-farmers (particularly for regulating services), as well as on the multiple relationships between agricultural practices (e.g. diversifying crops, no pesticide use, optimal stocking rate) and multiple regulating/cultural services (e.g. water quality, soil fertility, landscape quality) and biodiversity. They intuitively understood that an intermediate intensity of management (SNG grasslands, rather than IG) could improve the delivery of multiple ES. Non-farmers, in contrast, attached greater importance to cultural ES that were discussed in bundles, such as recreation and tourism, aesthetic value of the landscape and spiritual, educational and cultural values. Divergent views were also observed around the relationships between farming and conservation policies (Bernués *et al.*, 2016; Kok *et al.*, 2020b; Pascual *et al.*, 2021), where farmers are usually sceptical of conflicting species (predators in particular). Another ES where discrepancies often appear is the aesthetic quality of agricultural landscapes, where local people prefer higher levels of human intervention and agricultural activity, whereas the general population prefer more 'natural' landscapes (Bernués *et al.*, 2014, 2015; Faccioni *et al.*, 2019). Consensus does, however, exist between herders and scientists, agreeing that sparse trees and shrubs are beneficial for the grassland environment, biodiversity and for livestock (Molnar *et al.*, 2020).

Stakeholders groups are usually presented as homogenous, but diversity can be great within those groups in terms of socio-economic and psychographic characteristics (Rodríguez-Ortega *et al.*, 2019; Spash *et al.*, 2009). The subjective perceptions and interests of stakeholders for ES can vary across regions, socio-economic and policy contexts, and cultural backgrounds (Randall, 2002). However, more general patterns were also found across three mountain agroecosystems in Europe (Bernués *et al.* (2019): (1) the willingness to pay for the provision of ES exceeded in all cases the real level of public support, (2) further abandonment and intensification of agriculture were clearly rejected by the public, and (3) increasing the provision of biodiversity and specially regulating ES (prevention of forest fires, maintenance of water quality and soil fertility) always produced welfare gains for society. However, the optimal level of delivery was context dependent and people perceived trade-offs between ES across policy scenarios (e.g. provision of quality food products and regulating services).

Many ES constitute public goods that do not have a market price, and therefore farmers do not have economic incentives to produce them. SNG grasslands are appreciated due to the aesthetic and recreational value, but while farmers maintain these landscapes, the profit goes to others, e.g. the tourism industry. IG do not deliver those aesthetic and recreational services, but IG do give higher revenues from higher productivity levels. Hence, a transition to IG becomes attractive from an economic viewpoint. That becomes a market failure that needs to be addressed with adequate policies and support for grasslands that underpin the delivery of ES, i.e. SNG. In order to make the concept of ES operational and useful for land management, the current agro-environmental policies should be replaced by more specific and targeted policies, such as Payments for Ecosystem Services (PES). PES should be based on concrete biodiversity objectives or on the agricultural practices at farm level that provide these biodiversity objectives or more generally, bundles of ES directly favouring ES, like e.g. habitat provision, pollination and biological control (Rodríguez Ortega *et al.*, 2016). These authors designed a PES system based on the relationships between agricultural practices and ES and showed that maintaining grasslands and applying grazing management practices delivered higher bundles of ES.

Another way to incentivize farmers is by transferring social demands into farmers' economies through proper value chains (i.e. food products and services linked to grassland territories) and support from financial institutions (Ripoll-Bosch and Schoenmaker, 2021). Consumers, and society at large, increasingly hold 'ethical' concerns about the model of agriculture and the food chain. For example, the concerns for animal welfare and the environment constitute two of the main future trends with regard to meat consumption and aligns with the call to eat 'less but better' meat (Sahlin *et al.*, 2020) This opens up an opportunity to move from the standard bulk production to differentiated and value-added products, to develop novel value chains and quality labels valuing 'extrinsic quality attributes' (i.e. quality attributes based on production system) and to add to consumer well-being. To do so, it is necessary to expand the farm-to-fork frame to a wider one 'landscape-to-fork', which incorporates the structure and functioning of natural and SNG into the value chain (Bernués *et al.*, 2016). This expanded framework would mean that value chains become more circular, helping to close nutrient and energy cycles. The lower the dependence on external inputs (i.e. higher dependence on solar energy), the more circular the production system.

Conclusions

Grasslands are highly valuable ecosystems providing ES relevant for long-term sustainability of food production, farming systems and for general well-being of communities across the world. However, management of grasslands strongly determines their capacity to deliver multiple ES. Improved grasslands (IG) are 'designed' to maximize food production and other ES have not been traditionally considered in high-intensity management systems. Conversely, semi-natural grasslands (SNG) are characterized by more balanced provision of different ES. This was well reflected in our literature overview, where significant trade-offs emerged between biomass production and provision of other services in IG.

Both IG and SNG are likely to be needed for long-term sustainability of food production, but significantly more effort must be put into landscape-scale spatial configuration of farming systems to ensure a more balanced provision of ES. Given the significant decrease in the area of SNG over the past century in Europe, it is highly relevant that remaining SNG are valued and maintained, and that degraded or destroyed SNG are restored in farming landscapes. In addition, IG management should be steered towards a more balanced provision of ES. For that, ES bundles – associations among multiple services – can provide information about necessary shifts in management practices of IG. For example, trade-offs related to high livestock intensity could be reduced if the density is kept below carrying capacity, resulting in improved water quality, carbon storage, erosion prevention and enhance pollinator diversity, and improve cultural ES.

The scale at which ES are provided is important. Trade-offs and synergies may vary when scale changes, as the provision is highly dependent on the underpinning ecological functions which operate at different spatial scales. When managing population-based ecosystem services, like pollination, as well as water regulation and erosion control, synergies become more prominent when interventions are implemented at larger landscape scales. Hence, local conditions, as well as the wider landscape context and configuration, are highly important to be considered in order to avoid trade-offs and to promote synergies. Further, a clear focus on bundles of services, e.g. 'water-biomass production-erosion control' or 'habitat provision-pollination-biological control', could increase multiple ES supply and facilitate management of both SNG and IG grasslands. Approaches based on bundles of ES can allow us to describe the performance of grasslands concerning the evolution of trade-offs and synergies, and to evaluate their biological, technical, cultural and economic determinants. This knowledge can help to inform decision-making concerning the payment of non-market services, like agro-environmental schemes. We stress that the application of the ES concept to grasslands in farming systems should be used in an informed way. ES is certainly crucial for leveraging ecologically and economically sustainable grassland management but its effectiveness as a conceptual tool will depend on local biophysical conditions, a wide range of social features and dynamics as well as the knowledge and the intent of its user.

References

Adewopo J.B., Silveira M.L., Xu S., Gerber S., Sollenberger L.E. and Martin T. (2015) Management intensification impacts on particle-size soil carbon fractions in subtropical grasslands: Evidence from 13C natural abundance. *Soil Science Society of America Journal* 79, 1198-1205.

Bardgett R.D., Bullock J.M, Lavorel S. *et al.* (2021) Combating global grassland degradation. *Nature Reviews Earth & Environment* 2, 720-735.

Bengtsson J., Bullock J.M., Egoh B., Everson C., Everson T., O'Connot T., O'Farrell P.J., Smith H.G. and Lindborg R. (2019) Grasslands – more important for ecosystem services than you might think. *Ecosphere*, https://doi.org/10.1002/ecs2.2582

Bernués A., Rodríguez-Ortega T., and Olaizola A.M. (2016) From landscape to fork: value chains based on ecosystem services. *Options Méditerranéennes A* 115, 317-326.

Bernués A., Rodríguez-Ortega T., Ripoll-Bosch R. and Alfnes F. (2014) Socio-cultural and economic valuation of ecosystem services provided by mediterranean mountain agroecosystems. *PLoS One* 9, e102479. https://doi.org/10.1371/journal.pone.0102479

Bernués A., Rodríguez-Ortega T., Alfnes F., Clemetsen M. and Eik L.O. (2015) Quantifying the multifunctionality of fjord and mountain agriculture by means of sociocultural and economic valuation of ecosystem services. *Land Use Policy* 48, 170-178. https://doi.org/10.1016/j.landusepol.2015.05.022

Bernués A., Alfnes F., Clemetsen M., Eik L.O., Faccioni G., Ramanzin M., Ripoll-Bosch R., Rodríguez-Ortega T., and Sturaro O. (2019) Exploring social preferences for ecosystem services of multifunctional agriculture across policy scenarios. *Ecosystem Services* 39, 101002. https://doi.org/10.1016/j.ecoser.2019.101002

Bullock J.M., *et al.* (2011) Chapter 6: Semi-natural grasslands. In: *UK NEA, The UK National Ecosystem Assessment*, UNEP-WCMC, Cambridge, UK, pp. 161-196.

Cadman M., de Villiers C., Lechmere-Oertel R. and McCulloch D. (2013) *Grasslands Ecosystem Guidelines: landscape interpretation for planners and managers*. South African National Biodiversity, Institute, Pretoria, South Africa.

Cerdan O., *et al.* (2010) Rates and spatial variations of soil erosion in Europe: a study based on erosion plot data. *Geomorphology* 122, 167-177.

Chang J., Ciais P., Gasser T., Smith P. *et al.* (2020) Climate warming from managed grasslands cancels the cooling effect of carbon sinks in sparsely grazed and natural grasslands. *Nature Communications* 12, 118.

D'Ottavio P., Francioni, M. *et al.* (2018) Trends and approaches in the analysis of ecosystem services provided by grazing systems: A review. *Grass and Forage Science* 73, 15-25.

Dengler J., Biurrun I., Steffen Boch S., Dembicz I. and Török P. (2020) *Grasslands of the Palaearctic Biogeographic Realm: Introduction and Synthesis Encyclopedia of the World's Biomes 1*. https://doi.org/10.1016/B978-0-12-409548-9.12432-7

Diaz S., *et al.* (2015) The IPBES Conceptual Framework – connecting nature and people. *Current Opinion in Environmental Sustainability* 14, 1-16.

Dingkuhn E.L., Wezel A., Bianchi F.J., Groot J.C., Wagner A. and Yap H.T. (2020) A multi-method approach for the integrative assessment of soil functions: application on a coastal mountainous site of the Philippines. *Journal of Environmental Management* 264, 110461.

Dlamini P., ChivengeP. and Chaplot V. (2016) Overgrazing decreases soil organic carbon stocks the most under dry climates and low soil pH: A meta-analysis shows. *Agriculture, Ecosystems & Environment* 221, 258-269.

Eze S., Palmer S.M. and Chapman P.J. (2018) Soil organic carbon stock in grasslands: effects of inorganic fertilizers, liming and grazing in different climate settings. *Journal of Environmental Management* 223, 74-84.

Faccioni G., Sturaro E., Ramanzin M. and Bernués A. (2019) Socio-economic valuation of abandonment and intensification of Alpine agroecosystems and associated ecosystem services. *Land Use Policy* 81, 453-462. https://doi.org/10.1016/j.landusepol.2018.10.044

Fischer M., Rudmann-Maurer K., Weyand A. and Stöcklin J. (2008) Agricultural land use and biodiversity in the Alps - How cultural tradition and socioeconomically motivated changes are shaping grassland biodiversity in the Swiss Alps. *Mountain Research and Development* 28, 148-155.

Foley J., Defries R., Asner G.P. *et al.* (2005) Global consequences of land use. *Science* 309, 570-574.

Franzluebbers A.J. and Stuedemann J.A. (2009) Soil-profile organic carbon and total nitrogen during 12 years of pasture management in the Southern Piedmont USA. *Agriculture, Ecosystems & Environment* 129, 28-36.

Fu B.J., Liu Y., Lu Y.H., He C.S., Zeng Y. and Wu B.F. (2011) Assessing the soil erosion control service of ecosystems change in the Loess Plateau of China. *Ecological Complexity* 8, 284-293.

Guo X., Dai L., Li Q., Qian D., Cao G., Zhou H. and Du Y. (2020) Grazing significantly reduces soil water storage in alpine grasslands on the Qinghai-Tibet Plateau. *Sustainability* 12(6), 2523. https://doi.org/10.3390/su12062523

Haines-Young R. and Potschin M. (2018) *Common international classification of ecosystem services (cices) v5.1 guidance on the application of the revised structure*. Fabis Consulting Ltd., UK.

Hartel T., Fagerholm N., Torral, B.A. and Plieninger T. (2018) Forum: social-ecological system archetypes for European rangelands. *Rangeland Ecology & Management* 5, 536-544.

Herzon I., Raatikainen K.J., Helm A., Rusin S., Wehn S. and Eriksson O. (2022) Semi-natural habitats in the European boreal region: Caught in the socio-ecological extinction vortex? *Ambio.* https://doi.org/10.1007/s13280-022-01705-3

Horcea-Milcu A-I., Leventon J., Hanspach J. and Fischer J. (2016) Disaggregated contributions of ecosystem services to human well-being: a case study from Eastern Europe. *Regional Environmental Change* 16, 1779-1791.

Höonigova I. *et al.* (2012) *Survey on grassland ecosystem services. Report to the EEA – European Topic Centre on Biological Diversity*. Nature Conservation Agency of the Czech Republic, Prague, Czech Republic.

IPBES (2018) *Summary for policymakers of the regional assessment report on biodiversity and ecosystem services for Europe and Central Asia of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services.* IPBES, Bonn, Germany.

Isbell F., Craven D., Connolly J., Loreau M., Schmid B., Beierkuhnlein C., *et al.* (2015) Biodiversity increases the resistance of ecosystem productivity to climate extremes. *Nature* 527, 315-316.

Jonsson M., Bommarco R., Ekbom B. *et al.* (2014) Ecological production functions for biological control services in agricultural landscapes. *Methods in Ecology and Evolution* 5, 243-252.

Khan M.S.I. (2020) Supporting ecosystem services: concepts and linkages to sustainability. In: Leal Filho W. *et al.* (eds.) *Life on land, Encyclopedia of the UN Sustainability goals*. https://doi.org/10.1007/978-3-319-71065-5_34-1

Kleijn D., Rundlöf M., Scherper J., Smith H. and Tscharntke T. (2011) Does conservation on farmland contribute to halting the biodiversity decline? *Trends in Ecology & Evolution* 26, 474-481.

Kok A., de Olde E.M., de Boer I.J.M. and Ripoll-Bosch R. (2020a) European biodiversity assessments in livestock science: A review of research characteristics and indicators. *Ecological Indicators* 112, 105902. https://doi.org/10.1016/j.ecolind.2019.105902

Kok A., Oostvogels V.J., de Olde E. and Ripoll-Bosch M.R. (2020b) Balancing biodiversity and agriculture: Conservation scenarios for the Dutch dairy sector. *Agriculture, Ecosystems & Environment* 302, 107103. https://doi.org/10.1016/j.agee.2020.107103

Lal R. (2004) Soil carbon sequestration impacts on global climate change and food security. *Science* 304, 1623-1627.

Lamarque P., Tappeiner U., Turner C. *et al.* (2011) Stakeholder perceptions of grassland ecosystem services in relation to knowledge on soil fertility and biodiversity. *Regional Environmental Change* 11, 791-804.

Lemaire G., Hodgson J. and Chabbi A. (eds.) (2011) *Grassland productivity and ecosystem services.* CABI Publishing, Wallingford, UK.

Lindborg R., Bengtsson J., Berg Å., *et al.* (2008) A landscape perspective on conservation of semi-natural grasslands. *Agriculture, Ecosystems and Environment* 125, 213-222.

Lindborg R., GordonL.J., Malinga R., Bengtsson J., Peterson G., Bommarco R., Deutsch L., Gren A., Rundlöf M., and Smith H.G. (2017) How spatial scale shapes the generation and management of multiple ecosystem services. *Ecosphere* 8, e01741.

Millennium Ecosystem Assessment (MA) (2005) *Ecosystems and human well-being. Synthesis.* World Resources Institute, Washington, DC, USA. Available at: http://www.millenniumassessment

Manzan P. and White S.R. (2019) Intensifying pastoralism may not reduce greenhouse gas emissions: wildlife-dominated landscape scenarios as a baseline in life-cycle analysis. *Climate Research* 77, 91-97. https://doi.org/10.3354/cr01555

Martino S. and Muenzel D. (2018) The economic value of high nature value farming and the importance of the Common Agricultural Policy in sustaining income: The case study of the Natura 2000 Zarandul de Est (Romania) *Journal of Rural Studies* 60, 176-187.

Molnár Z., Kelemen A. *et al.* (2020) Knowledge co-production with traditional herders on cattle grazing behaviour for better management of species-rich grasslands. *Journal of Applied Ecology* 57, 1677-1687. https://doi.org/10.1111/1365-2664.13664

Naylor R., Steinfeld H., Falcon W., Galloway J. *et al.* (2005) Losing the links between livestock and land. *Science* 310, 1621-1622.

Pascual U., Adams W.M., Díaz S. *et al.* (2012) Biodiversity and the challenge of pluralism. *Nature Sustainability* 4, 567-572.

Peeters A., Beaufoy G., Canals R.M., De Vliegher A., Huyghe C., Isselstein J., Jones G. *et al.* (2014) Grassland term definitions and classifications adapted to the diversity of European grassland-based systems. *Grassland Science in Europe* 19, 743-750.

Pilgrim, E.S. *et al.*, (2010) Interactions among agricultural production and other ecosystem services delivered from European temperate grasslands. *Advances in Agronomy* 109, 117-154.

Plieninger T., Hartel T., Martın-Lopez B. *et al.* (2015) Wood-pastures of Europe: geographic coverage, social-ecological values, conservation management, and policy implications. *Biological Conservation* 190, 70-79.

Posthumus H. and Morris J. (2010) Implications of CAP reform for land management and runoff control in England and Wales. *Land Use Policy* 27*,* 42-50.

Potts S.G., Biesmeijer J.C., Kremen C., Neumann P., Schweiger O. and Kunin W.E. (2010) Global pollinator declines: Trends, impacts and drivers. *Trends in Ecology and Evolution* 25, 345-353.

Price C. (2014) Regulating and supporting services and disservices: customary approaches to valuation, and a few surprising case-study results. *New Zealand Journal of Forestry Science* 44, S5. https://doi.org/10.1186/1179-5395-44-S1-S5

Queiroz C., Beilin R., Folke C. and Lindborg R. (2014) Farmland abandonment: Threat or opportunity for biodiversity conservation? *Frontiers in Ecology and the Environment* 12, 288-296.

Randall A. (2002) Valueing the outputs of multifunctional agriculture. *European Review of Agricultural Economics* 29, 289-307. https://doi.org/10.1093/eurrag/29.3.289

Rau A-L., Burkhardt V., Dorninger C., Hjort C., Ibe K., Keβler L., Kristensen J.A., McRobert A., Sidemo-Holm W., Zimmermann H., Abson D.J., von Wehrden H. and Ekroos J. (2010) Temporal patterns in ecosystem services research: A review and three recommendations. *Ambio* 49, 1377-1393. https://doi.org/10.1007/s13280-019-01292-w

Raudsepp-Hearne C., Peterson G.D. and Bennett E.M. (2010) Ecosystem service bundles for analyzing tradeoffs in diverse landscapes. *Proceedings of the National Academy of Sciences of the USA* 107, 5242-5247.

Richter F., Pierrick J., El Benni N., Lüscher A., Buschmann N. and Klaus V.H. (2021) A guide to assess and value ecosystem services of grasslands. *Ecosystem Services* 52, 101376. https://doi.org/10.1016/j.ecoser.2021.101376

Ripoll-Bosch R. and Schoenmaker D. (2021) *Impact of finance on biodiversity: How agricultural business models get financed and promoted. Erasmus Platform for Sustainable Value Creation*, Working Paper. Accessible at: https://www.rsm.nl/fileadmin/Images_NEW/Erasmus_Platform_for_Sustainable_Value_Creation/20220214_Impact_of_finance_on_biodiversity_-_How_agric

Rodríguez-Ortega T., Bernués A. and Alfnes F. (2019) Psychographic profile affects willingness to pay for ecosystem services provided by Mediterranean high nature value farmland. *Ecological Economics* 128, 232-245.

Sahlin K.R., Röös E. and Gordon L. (2020) Less but better meat is a sustainability message in need of clarity. *Nature Food* 1, 520-522.

Saidi N. and Spray C.J. (2018) Ecosystem services bundles: Challenges and opportunities for implementation and further research. *Environmental Research Letters* 13, 113001.

Skinner R.H. (2013) Nitrogen fertilization effects on pasture photosynthesis, respiration, and ecosystem carbon content. *Agriculture Ecosystems & Environment* 172, 35-41.

Smith P. (2014). Do grasslands act as a perpetual sink for carbon? *Global Change Biology* 20, 2708-2711.

Sollenberger L.Y., Kohmann M.M., Dubeux Jr. J.C.B. and Silveira,M.L. (2019) Grassland management affects delivery of regulating and supporting ecosystem services. *Crop Science* 59, 441-459.

Soussana J.F., Tallec T. and Blanfort V. (2010) Mitigating the greenhouse gas balance of ruminant production systems through carbon sequestration in grasslands. *Animal* 4, 334-350.

Spash C.L., Urama K., Burton R., Kenyon W., Shannon P. and Hill G. (2009) Motives behind willingness to pay for improving biodiversity in a water ecosystem: economics, ethics and social psychology. *Ecological Economics* 68, 955-964.

Stenseke M. (2009) Local participation in cultural landscape maintenance: lessons from Sweden. *Land Use Policy* 26, 214-223.

Sucholas J., Greinwald A., Heinrich M. and Luick R., (2017) Naturally occurring medicinal plants as ecosystem service of extensively used grassland. *Grassland Science* 22, 624-626.

Taki H., Okabe K., Yamaura Y., Maeto K. *et al.* (2010) Effects of landscape metrics on Apis and non- Apis pollinators and seed set in common buckwheat. *Basic and Applied Ecology* 11, 594-602.

Tilman D., Hill J. and Lehman C. (2006) Carbon-negative biofuels from low-input high-diversity grassland biomass. *Science* 314, 1598-1600.

Torralba M., Fagerholm N., Hartel T., Moreno G. and Plieninger T. (2018) A social-ecological analysis of ecosystem services supply and trade-offs in European wood-pastures. *Science Advances* 4, eaar217. https://doi.org/10.1126/sciadv.aar2176

Vári A., Arany I., Kalóczkai A., Kelemen K., Papp J. and Czúcz B. (2020) Berries, greens, and medicinal herbs – mapping and assessing wild plants as an ecosystem service in Transylvania (Romania). *Journal of Ethnobiology and Ethnomedicine* 16, 13. https://doi.org/10.1186/s13002-020-0360-x

Velado-Alonso E., Morales-Castilla I., Rebollo S. and Gómez-Sal A. (2020) Relationships between the distribution of wildlife and livestock diversity. *Diversity and Distribution* 26, 1264-1275. https://doi.org/10.1111/ddi.13133

Werling B.P. *et al.* (2014) Perennial grasslands enhance biodiversity and multiple ecosystem services in bioenergy landscapes. *Proceedings of the National Academy of Sciences of the USA* 111, 1652-1657.

Wilson J.B., Peet J.P.K., Dengler J. and Pärtel M. (2012) Plant species richness: the world records. *Journal of Vegetation Science* 23, 796-802.

Wood T.J., Smith B.M., Hughes B., Gill J.A. and Holland J.M. (2013) Do legume-rich habitats provide improved farmland biodiversity resources and services in arable farmland? *Aspects of Applied Biology* 118, 239-246.

Yahdjian L., Sala O.E. and Havstad K.M. (2015) Rangeland ecosystem services: shifting focus from supply to reconciling supply and demand. *Frontiers in Ecology and Environment* 13, 44-51.

Zhao Y., Liu Z. and Wu J. (2020) Grassland ecosystem services: a systematic review of research advances and future directions. *Landscape Ecology* 35, 793-814. https://doi.org/10.1007/s10980-020-00980-3

Zisenis M., Richard D., Vačkář D., Lorencová E. *et al.* (2011) *Survey on grassland ecosystem services in the Czech Republic and literature review*. ETC/BD report to the EEA.

Multifunctionality and diversity of livestock grazing systems for sustainable food systems throughout the world: what can we learn for Europe?

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Abstract

Throughout the world, livestock grazing systems (LGS) include and provide livelihoods for many rural populations. These LGS are represented in a wide variety of agroecological contexts and offer a huge variety of system organization. They contribute to sustainable food systems by providing multiple products including low-cost edible proteins and energy, draft power, outputs (carbon and soil nutrient regulation, landscape and biodiversity maintenance), roles (local development support in harsh environments, contribution to the circular economy) and benefits to population (revenue, employment and cultural assets). These multiple functions can be described through a multifunctional conceptual model specified for LGS. Applied to cases in Africa, Asia, Latin America and Europe, the framework enables the assessment of these systems in a holistic manner that includes four dimensions: production, social, environmental and local development. These dimensions and associated local indicators demonstrate the potential important contribution LSG makes to sustainable food systems. Management of trade-offs between these functions may be improved using, such a model in a multi-stakeholder approach. Some of the functions and balance between them might have been overlooked in the consideration of European food systems.

Keywords: livestock grazing systems, multifunctionality, diversity, sustainable food systems

Introduction

Livestock grazing systems (LGS) are where 90% of ruminant diets are composed of forage grazed from natural or cultivated grasslands, according to FAO and ILRI (Robinson *et al.*, 2011). LGS play a significant role in livestock production, accounting for 39% of global domestic ruminant numbers and 30% of animal derived proteins (Mottet *et al.,* 2017, 2018). One and a half billion hectares of land usually unsuitable for cropping due to poor rainfall, soil fertility and topography are utilized by LGS, as is 54% of the total terrestrial landscape. Much of this (28M km^2) is in desert or marginal xeric shrubland areas (ILRI *et al.,* 2021). Many of these systems are dependent on both the mobility of livestock and people (socio-ecological systems) as they take advantage of the spatial and temporal variability in forage production throughout the year. These mobile systems rely on natural resources and processes, e.g. existing forage, water source, manure from livestock and associated high human capital input. The large land footprint of LGS and associated management of livestock results in impacts on the ecosystem dynamics which result in a diversity of functions for both the environment and human well-being at different scales and dimensions (production, economics, cultural, environmental, local development, etc.). These functions are not always considered when assessing the impact of LGS although attempts to take a whole-of-system approach have been undertaken using the 'Ecosystem Services' framework focused at the ecosystem scale (Huang *et al.,* 2015). The prolific debates that occurred during the 2021 UN Food Systems Summit have led to several so-called 'Coalitions' that have to be implemented by states and civil society, many of them dealing with livestock issues. They have confirmed that Food Systems are now a global issue and that industrialized countries cannot represent a model for the rest of the world, but also have a lot to learn from the Global South and its diversity of farming systems, particularly about herbivore breeding. In this paper, we shall consider LGS, tackled here in a larger definition than FAO, within the context of a multifunctionality framework that makes transparent the many functions derived from LGS, and we document the results of its application through global case studies. The hypothesis is that the multiple functions of LGS, demonstrated in a diversity of global contexts, will inform the description and identify pathways for sustainable food system development potentially overlooked in past agriculture simplification within Europe.

Why apply the multifunctionality concept to livestock grazing systems?

Current methods of assessing the different functions of LGS oversimplify and underestimate the impact. We hypothesize that the use of the concept of 'Multifunctionality of agriculture,' which was developed during the 1990s (Caron *et al.,* 2008; Hervieu *et al.*, 2002a,b; Huang *et al.,* 2015; UNCED, 1992) is a better way for developing a more exhaustive assessment of the different functions of LGS. Through this multifunctionality (MF) methodological approach, we seek to show that LGS have an important role to play in Sustainable Food System development worldwide. The MF considers the diversity of functions needed to assess the real impact of agriculture at local, regional and international levels including production outputs, economic (employment, infrastructure and services development, financial fluxes, etc.), environmental (landscape management, GHG emissions, soil fertility, biodiversity and nutrient fluxes, etc.). Due to their large terrestrial footprint from local to global scale, LGS have significant impacts on ecosystem dynamics (biodiversity, nutrient cycling, land degradation, etc.) and climate change (GHG emissions, carbon sequestration) (Steinfeld *et al.,* 2006). LGS also support massive amounts of social groups and populations throughout the world (ILRI *et al.,* 2020), providing revenues, livelihoods, social and cultural assets. In this regard, the MF framework has been adopted by Action Network 2 'Restoring value to grassland' within the Global Agenda for Sustainable Livestock (GASL), as the relevant approach to use with multiple stakeholders to describe, evaluate and discuss the different functions provided by LGS. This MF framework fits well with the global framework on Sustainable Development Goals (SDG) proposed by the UN 2030 program as the multiple functions of LGS relate to at least 8 SDGs out of the existing 17 (1: no poverty, 2: zero hunger, 5: gender equality, 6: clean water, 8: decent work and economic growth, 12: responsible consumption and production, 13: climate action, 15: life on land). Finally, considering the contribution of LGS to the emergent concern of sustainable food systems (SFS) debated during the September 2021 UN conference, the MF framework will allow the identification of crucial functions that might inform the main principles supporting SFS: environmentally friendly, easy access, availability, food security, food quality.

Building a multifunctionality conceptual model to support local livestock grazing systems dynamics

A multi-stakeholder participative modelling approach was developed to ensure that a broad diversity of contexts and worldviews informed a common framework applicable to the diversity of LGS global contexts. Participants included researchers from a range of disciplines related to LGS from seven different countries (Argentina, Brazil, France, Mongolia, Senegal, New Zealand, Vietnam), agribusiness, farmers and policy makers. An iterative approach was applied to ensure the robustness of the framework which consisted of: (1) a literature review that created the base platform for conceptual model construction at the first workshop (May 2016) followed by (2) interviews with ten French farmers, and later with local stakeholders at sites of five of the different countries, and (3) two further workshops (July 2016, December 2017) with several rangeland experts focused on clarifying definitions, discussions on the structure of the conceptual model and testing its robustness with respect to a set of indicators defined to assess the impact of livestock from a variety of perspectives. The resulting multifunctionality of LGS conceptual model (CM) consists of four dimensions (productive, social, local development and environmental) within which entities (farmers, livestock, pastures, products, atmosphere, water, infrastructures, organizations, etc.) and processes (trading, feeding, producing, consuming, building, earning, etc.) have been identified, chosen and described in the UML language with their associated indicators (Figure 1 and indicators in the Case study descriptions). From 2017 onwards, the CM has been applied to a variety of case studies, documented below, from which iterative improvement and enrichment of the CM has occurred via practical experience.

Case studies from South America, Asia, Africa and Europe

The cases documented below illustrate how the multifunctionality framework has been applied with different tools and methodology to a diversity of contexts and issues regarding sustainable development of LGS throughout the world.

Improving holistic comprehension of multifunctional goods and services provided by pastoral ecosystems: Puna de Jujuy, Argentina

Context. The Puna (3,500 m a.s.l.) is a high plateau located in a dry area (100 to 300 mm year^{-1} rainfall), very windy with high daily and annual temperature fluctuations. The vegetation is sparse, mainly shrubby steppe and archipelagos of very productive but sparsely distributed wetlands. In these harsh environments there are limited possibilities for agriculture (Quiroga Mendiola and Cladera, 2018). The aim of this work is to promote the values of this high altitude pastoral system as it is a producer of multiple goods and services. We organized the case study into the four dimensions of the MF framework (productive, social, local development and environmental) through a multidisciplinary team approach to facilitate a holistic analysis of the whole system.

Material and methods. A domestic unit (DU) analysis scale was chosen as it is the first economic step of natural grasslands management and knowledge transmission, and because relevant indicators and measures already existed at this scale for use in the approach. Four dimensions were addressed: (1) productive dimension: the main local product is meat (llama, sheep or goat) sold in the local market (formal and informal); indicators are: (a) \$ kg^{-1} of meat produced year^{-1}; (b) kg meat sold year^{-1}; (c) livestock diversity: number of animal species/flock; and (d) number of strategies against drought. (2) Social dimension: we assessed (a) the number of family members living and working in the household (productive unit persistence and knowledge transmission); and (b) number of local organizations in which the DU participates. (3) Local development dimension: (a) annual income (US\$ of meat) and (b) the number and diversity of marketing channels for the meat produced. (4) Environmental dimension: kg DM ha^{-1} year^{-1} (carbon capture and forage provision), vegetation cover (water and temperature regulation), plant diversity and richness (biodiversity maintenance).

Results. The meat production ranged from 487 to 2,272 kg meat^{-1} year^{-1} DU^{-1}, of which 50% was for self-consumption and 50% for sale (Echenique *et al.*, 2015; Paz *et al.,* 2011). The herds have two animal species and 3 to 5 diverse strategies were applied to face drought (changing grazing sites, changing flock composition, reducing flock size, buying fodder from outside the area, and finding new ways of agreement between herder's families) (Quiroga Mendiola, 2015a). This demonstrates the family's capacity to produce meat for self-consumption and for other consumers, and also the different knowledge and strategies for various animal species per flock and diverse landscape management. Concerning the social dimension, the stability or fragility of the family was demonstrated as they are made up of 1 to 5 members that remain in the production unit and are linked to 1 to 4 local market organizations. These networks provide diverse and flexible opportunities such as negotiation capacities, improving selling prices, information access, etc. (Alcoba *et al.*, 2018). The local development dimension generates annual income of US$1,194 to US$6,289 (local and country wealth generation) and access to diverse marketing channels (actions of the cooperative to sell the meat outside the territory, selling most of the meat for Christmas, Easter or social events, selling some animals to an intermediary or local trader, sale or exchange of meat with neighbouring families or other members of the community) showing the generation of wealth and family and community resilience (Alcoba *et al.*, 2018). The environmental dimension shows a forage production of 300 kg DM ha^{-1} $year^{-1}$, with 65 to 73% vegetation cover and genetic richness and plant diversity conservation in a sustainable way (Molina, 2011; Quiroga Mendiola *et al.*, 2010, 2015b).

Conclusions. The multifunctional goods and services that the pastoralist agroecosystem provides were acknowledged and analysed, and were better captured by a multidisciplinary team to provide a more comprehensive understanding of the system complexity. The application of the MF approach allowed us to measure and integrate several indicators in the four different dimensions, analyse diverse herder strategies to cope with this kind of environment and be more resilient to shocks, and to make transparent and place a value on the system's multifunctionality.

Improving grassland system multifunctionality by natural regeneration of native trees for the implementation of a silvopastoral system for beef production in Brazil

Context. In Latin America, extensive systems are the most common management for cattle ranching, and traditionally are based on monoculture forages and low stocking rates (Chara *et al.*, 2017). Deforestation is part of the process applied to implement monoculture pastures in large areas and in different biomes of tropical countries, including Brazil. This practice improves profits in the short-term, but after many years the soil fertility, biodiversity and stocking rate capacity are reduced, and consequently also the farmers' income. Pasture degradation has taken place on approximately 100 million hectares all over Brazil. Silvopastoral systems which incorporate trees and shrubs in pastures increase the amount of biomass per unit of area and provide other ecosystem services. Silvopastoral systems aim to promote sustainable intensification of land while also increasing the vegetation and animal biodiversity, water use efficiency and biomass production, and respecting animal welfare, compared with systems based on traditional monoculture forages (Mauricio *et al.*, 2019). The objective of this study is to demonstrate that natural regeneration of native trees and bushes associated with grass forages is one sustainable option to implement a multifunctional silvopastoral system (SPS) in Brazil.

Material and methods. Several seminars were organized where a demonstration farm (1,000 ha in Maranhao State, Brazil) was used to illustrate and discuss the SPS practices (natural regeneration) with farmers, ONG (Brazilian Centre for sustainable livestock – CBPS), local extension services, researchers and students under the coordination of the Federal University of Sao Joao Del-Rei Brazil. The four dimensions of the MF approach were applied to the SPS, as SPS deliver a range of functions including high production (meat ha^{-1} $year^{-1}$), social improvements (jobs and financial stability), environment (biodiversity, animal welfare) and local development (livestock business and sustainable practices).

Results. It was demonstrated that the profit from the SPS has steadily increased in comparison with traditional monoculture systems based exclusively on *Brachiaria*. In addition, high biodiversity, fauna and flora from silvopastoral practices has positively changed the farm landscape, which has been enhanced through soil conservation, forage biomass and animal welfare. The seminars fostered discussions and clarified several technical points among stakeholders (farmers, technicians and students) which facilitated the practical changes towards implementation of the SPS.

Conclusions. The multifunctionality and multi-stakeholder approach used in this case study (farmers, researchers, extension services and students) increased the adoption of the system by other farmers (1000 visitors per year) and consequently improved the sustainability of livestock production in the region. It is expected that the economic, social and environmental benefits of the SPS could be used for further policies and payment for ecosystem services.

Multifunctionality of the Tibetan grassland system

Context. Known as the 'Water Tower of Asia', the Qinghai-Tibetan Plateau is an irreplaceable source of water for the billions of people living downstream. The Plateau is a vast plain at over 4,000 m above sea level and surrounded by mountain ranges. Its unique geological history and high-elevation environment makes it the centre of origin for a rich number of plants and animals. The world's largest grazing system, i.e. *Kobresia* grasslands, covers an extensive 450,000 km^2 of the Plateau and has been formed by pastoralism over the past 8,000 years (Miehe *et al.*, 2019). The dominant plant species, *Kobresia pygmaea*, is a sedge less than 4 cm high, adapting well to the grazing of livestock (*ibid*.). Having a thick turf layer and a dense root mat, *Kobresia* grasslands are resistant to yak trampling and have high water retention capacity. Due to steady population growth and production-orientated agricultural policies during the 'people's commune' time, there was a substantial increase of livestock numbers from the 1950s to 1980s. Although overgrazing posed threats to the provisioning of ecosystem services of the *Kobresia* grasslands, the total number of livestock has declined continuously following a series of grassland protection and restoration policies launched during the past two decades. In the *Kobresia* grasslands of Qinghai, the stocking rate from 2003-2012 was 15.41 million sheep units, showing a drop of 21.3% compared with 1988-2002 (Zhang *et al.*, 2014). Further decline of livestock numbers is projected to take place responding to intensifying urbanization and subsidized land-use extensification. Previous studies on Tibetan avian assemblages (Li *et al.*, 2018) have found that small-farming pastoralism can keep the grassland landscape, slow down the encroachment of shrubs, e.g. *Potentilla fruticosa*, and create habitats for open-grassland specialists. In the regional development planning of Qinghai Province (2021), the main function of *Kobresia* grasslands is water and biodiversity conservation. However, more than 80% of the population in this region are still subsistence pastoralists (National Development and Reform Commission, 2013) and for local pastoral communities, the provisioning service of the grasslands remains prominent. Prioritizing *Kobresia* grasslands' environmental functions in the national policies often leads to the question: in which conditions can the grasslands' environmental and economic functions be realized in synergy?

Material and methods. Using the MF framework, we examined the impact of yak grazing on biodiversity and landscape structure in Nyanpo Yutse of the eastern Qinghai-Tibetan Plateau. Using Unmanned Aerial Vehicles, we obtained high-resolution (15-cm level) landscape imageries of 45 km^2 and calculated landscape heterogeneity indices (Fritz *et al.*, 2018). We conducted two breeding-season bird surveys in 140 sample plots. To measure yak grazing intensity, we first conducted participatory mapping of the pasture boundaries, and then counted herd size grazed on each of the 140 sample plots. Finally, we developed statistical models to test the threshold of grazing intensity that can best sustain the multifunctionality of the livestock grazing system.

Results. Our study found no significant correlation between livestock grazing intensity with bird diversity, while the landscape mosaic created by yak grazing had a positive impact on bird species richness. Particularly, human built structures, including Tibetan prayer flags, increased the vertical complexity of the landscape, and formed a keystone structure (Tews *et al.*, 2004) to sustain high-diversity bird assemblages. Among pastures where livestock grazing intensity is lower than 1 sheep unit ha^{-1}, there was pronounced species replacement among sample sites, indicating that species having varied habitat requirements could coexist in the landscape where extensive pastoralism was practised.

Conclusions. Our study demonstrated that extensive pastoralism will benefit the Plateau's biodiversity conservation through maintaining the heterogenous structure of the landscape. The multifunctionality of the Tibetan grassland system should be acknowledged and supported: the *Kobresia* grasslands are not only a wilderness region that matters for biodiversity and water conservation, but also a precious cultural landscape where tangible and intangible values of nature and society intertwine.

Community-based conservation provides a platform for maintaining multifunctional use of Mongolian mountain-forest steppe ecosystems

Context. Mongolian rangelands account for 73% of the country's territory that directly support the livelihood of over 300,000 pastoralists, around 10% of the population (NSO, 2020). Half of all Mongolians benefit from the economic activity generated from pastoralism. The most common livestock types include sheep, cows, yaks, goats and horses. Mongolia's rangelands encompass three major ecological zones: mountain-forest-steppe, steppe, and desert-steppe (Hilbig, 1995). Khoid Mogoin Gol Teel Local Protected Area (KMG-T LPA) in Bulgan soum (district), Arkhangai aimag (province), occupies 137,000 ha of mountain forest-steppe. One-third of the LPA is covered by forests (44,830 ha) that host rich biodiversity including globally endangered species such as musk deer, saker falcon, steppe eagle, red deer, and Mongolian marmot (Marshall-Stochmal *et al.,* 2020). As of 2020, the LPA provided forage for over 34,000 livestock reared by over 200 herder households residing within the KMG-T. Due to its proximity to the Arkhangai centre and the central road to the western region, KMG-T LPA has been affected by illegal logging, poaching, forest fires and overgrazing. Therefore, the Bulgan Soum Government took KMG-T under local protection in 2017, and the Zoological Society of London (ZSL) facilitated the management of the LPA from 2018.

Methods and methods. Multiple stakeholders, including the Bulgan Soum Government, the Union of Conservation Communities (UCC) uniting 15 herder organizations, Aimag Environment & Tourism Department (ETD), Aimag Forestry Unit (FU), Aimag Ecological Police (EP) and the ZSL have been co-managing KMG-T LPA. Their differing roles in the use of rangelands and partnerships for maintaining ecosystems were analysed using the multifunctionality framework and associated indicators.

Results. Concerning the social dimension, over 270 herders (162 households) joined the UCC with increased participation in natural resource management and positive attitudes towards nature engaging in conservation activities. Poverty rates decreased (from 0.115 to 0.084) with increased access to financial services through Village Saving and Loan Associations (VSLAs) and a rise in the average household income (553,837 MNT to 963,224) (IRIM, 2021). Concerning the environment, thanks to 17 volunteer rangers conducting SMART patrolling in their areas, KMG-T became a zero-poaching area with a substantial reduction in illegal logging. The UCC reintroduced marmots, whose population increased 36% over three years, and the population of musk deer and red deer remained stable (IRIM, 2021). UCC members fenced 8.3 ha of forest areas supporting natural regeneration and reforested 3 ha areas. For local development, the UCC's conservation inspired other communities in Bulgan Soum and the Government leading to the establishment of five more herder groups (ZSL, 2021). Most of the tree planting and waste cleaning activities in Bulgan are being handled by UCC members. Environment and Forestry units collaborated with UCC herders to clean forests in over 30 ha and firewood was supplied to Aimag residents, raising around 60 million MNT over three years (ZSL, 2021). Concerning production, with increased income the livestock production in KMG-T was slightly increased (8%), including cattle (20%), horses (7%), sheep (2%) and goats (3%) (IRIM, 2021). Besides livestock production, UCC members diversified their incomes by introducing new businesses such as tourism, haymaking, vegetable growing and briquette-making (ZSL, 2021).

Conclusions. The case study confirmed the applicability of selected indicators across four dimensions of the multifunctionality framework in the complex Mongolian livestock system (LS). Specifics to Mongolia included additional new indicators proposed by herders that reflect their perspectives shaped by the nature of extensive LS and pastoral cultures. Social and environmental dimensions were a more pronounced feature for Mongolian LS compared to local development and production dimensions. The government partners and CBOs found the multifunctionality concept and the indicators useful for M&E and Planning for rangeland management specifically measuring progress towards SDG 1, 3, 5, 6, 8, 13, 15, and 17.

Dairy-oriented agropastoral system in northern Senegal: thinking multifunctionality of milk production in a semi-arid environment

Context. Milk production in Senegal is mainly from pastoral systems (Corniaux *et al.*, 2012). However, this production is too seasonal and dispersed to provide a significant supply to dairy industries and therefore very poorly collected by local industries, most of which prefer to use imported milk powder, mainly because of the lack of competitiveness of local milk, which is still very expensive as a raw material. In northern Senegal, the department of Dagana is experiencing a dynamic in the local milk sector due to the development, since 2007, of an industrial dairy that uses local milk (Bourguoin *et al.*, 2018). This company is faced with seasonal hazards and strong variability in production from year to year. The Dagana milk innovation platform (PIL), created at the end of 2014, brings together all the stakeholders involved in the local milk value chain (breeders, farmers, collectors, processors, NGOs, public institutions) to work on scenarios for the sector's development. Since 2018, a reflection has been carried out on the means to ecologically intensify milk production by relying on local agricultural and natural resources. The objective is to better understand the local milk production potential by adopting a perspective on the multifunctionality of this sector in the Sahelian pastoral system.

Material and methods. Starting from the MF conceptual model, this work consisted of co-constructing a computer simulator with PIL stakeholders that is capable of modelling zootechnical, ecological, agricultural, socioeconomic and geographical parameters (Delay *et al.*, 2021). This model reproduces the production conditions of livestock farmers in the Sahelian strip living in the vicinity (50-60 km radius) of a river that irrigates intensive agriculture on its banks. Workshops enabled the stakeholders to put forward various hypotheses on the organization of the sector and to discuss the constraints of each type of stakeholder. A first workshop for the general public focused on the role of biomass flows in the sustainability of pastoral dairy systems. A second workshop focused on the organization of the milk collection system with local stakeholders in order to achieve greater efficiency and social inclusion.

Results. The milk potential of the Richard-Toll dairy basin was estimated at between 2,000 and 10,000 litres day^{-1} according to different seasons and three levels of productivity: pure pastoral, intensified pastoral and intensified pastoral with stabling (Cesaro *et al.*, 2020). During the workshop discussions, the stakeholders considered that these estimated potentials were credible because they were sufficiently close to the reality on the ground (collection varying between 3,500 and 9,500 litres day^{-1} between 2018 and 2021), accounting for local fodder resources. To this end, the objective of exploiting the milk potential efficiently and sustainably requires cooperation between actors in several sectors (rice, sugarcane and milk). Nevertheless, rules of access to agricultural areas by livestock farmers must be discussed between actors to allow the circulation of biomass on a territorial scale. Maximum scenario estimates the material flow at 4,000 tonnes of dry fodder (rice and sugar cane) and 2,000 tonnes of agriculture by-products (rice bran). Moreover, dairy intensification may also induce equity in the allocation of natural and economic resources between groups of herders and have social (concentration of resources) and environmental (concentration of herds) consequences. Intensive pastoral farms produce 3 to 4 times more milk than a traditional pastoral system but need 8 times more inputs. Cattle prolificacy is also 3 times higher in intensive pastoral farms than traditional ones. This new distribution may increase the differentiation between herders living near agricultural areas and those living in sylvopastoral areas.

Conclusions. The use of the concept of multifunctionality (Ickowicz *et al.*, 2018) during the simulation workshops allowed the stakeholders to see what levels of interdependence should be considered to achieve sustainable dairy intensification scenarios and to better comprehend and understand the points of view of the other stakeholders in the territory and the compromises to be sought.

Grazing livestock system in mountainous north-west Vietnam as a sustainable option for local development

Context. In the mountains of north-western Vietnam, smallholder livestock farms rely heavily on natural pastures for animal feed (cattle, buffalo). However, livestock grazing systems are considered insufficiently intensive to meet the national increase in meat consumption and reduce import dependency, and to provide sufficient income to value chain stakeholders to contribute to poverty reduction. Livestock farming is in competition for space and resources with other economic activities (fruit and forestry plantations), or environmental protection (forest protection). These systems therefore remain weakly supported by local government, and are not considered in the livestock development strategies. Reconsidering the multiple functions of mountainous grazing systems at landscape level might change the assessment of their role in local development strategies.

Material and methods. This study has quantified the multiple contributions of the grazing systems to the sustainable development of farms and territories using the example of livestock farms in Quai Nua commune in Dien Bien Province. In this mountainous commune, extensive grazing systems coexist with livestock systems in the process of intensification with trough feeding, forage production and fattening systems. The approach was to identify indicators from the multifunctionality framework on the four dimensions covering the herd: the farm, the community and the landscape and the services and value chain scales. The indicators were used in discussions on the contribution of livestock grazing systems to sustainable development with a diversity of local stakeholders (livestock farmers, agricultural extension staff, representatives of the livestock cooperative, stakeholders of the beef value chain).

Results. This study produced references on the contribution of livestock grazing systems to sustainable development in the study Quai Nua Commune. Concerning the production dimension, livestock grazing systems produce about 49% of the beef production and about 48% of the meat integrated into the beef value chain (fresh meat, meal and dried meat typical of this region). For the environmental dimension, livestock grazing systems support soil organic fertility and production of the cropping systems through about 18% of the manure produced at communal level. Permanently stalled livestock provide the remainder. The contribution of these systems to landscape management has not been assessed. The livestock grazing systems contribute to local development with 11% of the profits of actors in the beef value chain (collector, slaughter man, restaurant, and processor). Other profits come from more intensive livestock systems and monogastric livestock. In total, 66% of farm workers are directly linked to these systems. However, although consumers show a preference for meat from grazing systems, the products from these systems have not been differentially marketed. Using the MF framework allowed the identification of different points of view. Livestock farmers attach importance to income and low input production. They also emphasize the importance of the social links that exist between farmers who graze (through sharing time to supervise animals at pasture). Finally, in addition to the function of bank savings (to cover accidents, planned events), these herds also provide for needs during social events (weddings, funerals, etc.). Agricultural extension officers explain that livestock grazing systems contribute to the livelihoods and standard of living of the population, providing an opportunity for work in a region that lacks it. Although these systems contribute clearly to poverty alleviation, the other actors of the beef value chain still focus on the functions of meat production and quality products.

Conclusions. These discussions highlight the full complementarities of the contributions of livestock grazing systems to production and economy, but also to the social dimension and to local development in the Province. Grasslands, essential for animal feed, contribute significantly to meat production, job creation, income and profits along the beef value chain. It seems necessary to ensure a visible, logical and sustainable approach to the management of grasslands to support animal production and the sustainable development of territories where livestock grazing systems are part.

What is at stake about assessment of multi-functionality of grazing systems in French Mediterranean mountains?

Context. Landscapes of most of the mountain regions of the Mediterranean area in Europe have been strongly shaped by pastoral farming, while this activity also contributes to the cultural identity of these areas. Livestock farming in these regions relies on grazing and it co-exists with the dynamics of livestock farming that relies on intensification and the associated increasing contribution of industrial feed. These regions also face deep socio-economic changes in the move from rural to residential and the tourism economy (Garde *et al.*, 2014). As a consequence, public lands, a main component of grazed areas in the Mediterranean, support multiple uses that livestock farmers have to deal with. Meanwhile, environmental management of so called 'semi-natural areas' and the contribution of grazing to biodiversity has become of concern while these constitute a reservoir for endangered species like wolves. The Agri-environmental scheme promoted by the European common agricultural policy has amplified this concern and put emphasis on grazing practices. Concern related to the future of livestock grazing goes beyond the environmental dimension alone and also addresses considerations for contributing to cultural identity, maintaining landscape (two dimensions strongly related to tourism activity) as well as delivering food products rooted in the local economy. Social concerns include how to enhance the interaction between residents and the promotion of inclusiveness. These dynamics indicate the complexity of the social-ecological system and to explore the future of the livestock grazing system within this dynamic requires dialogue with all stakeholders involved across scale from the sector to the territory (Zahm, 2008).

Material and methods. Our hypothesis is that applying a multifunctionality approach of LGS will support discussions between stakeholders in their dialogue on a sustainable future for livestock farming activities in territories. We interviewed stakeholders involved in livestock activities in the Provence-Alpes-Côte-d'Azur region, e.g. livestock farmers and their representative, farm and pastoralism advisers, food chains operators, local elected persons, representatives of nature protection associations, protected area managers, local development associations, etc. These interviews included considerations on the diversity of livestock farming, the main recent changes, difficulties with ensuring the future of farm activities or interacting with other land users. We then organized three focus groups to deliberate on this future. Short videos of the interviews, where the different points of view of stakeholders were captured, helped to organize the dialogue around the dimensions making it easier for participants to express their views.

Results. Among actors closely related to farming activities (farmers, pastoral advisers, meat-sector operators and protected area managers) the main questions regarding multifunctionality dealt with the trade-off between the abilities to use the LSG system for the preservation of forage resources, as a marketing advantage for specific pastoral products, and advocacy for the usefulness of pastoral systems to foster biodiversity of natural grazed areas (i.e. justifying strong public support as elaborated within the second pillar of the European CAP). In a wider arena of discussion, involving actors of the local community, questions dealing with protection of remarkable or endangered species related to pastoral ecosystems were embedded within a wider spectrum of questions including the maintenance of local identity, high value tourism economic operations, as well as contributing in designing and reinforcing social interactions at local level. Reinforcing diversity of participation is required, especially the inclusion of citizen associations and consumers. It appears also that putting emphasis on short supply chains is a lever to reinforce the perception of livestock activities within the territory as it helps to maintain dialogues and interaction between local society and farmers while allowing farmers to keep control on maintaining consistencies for their systems and the meanings of their jobs (Lasseur and Dupré, 2018).

Conclusions. Using the multifunctionality approach enabled the reduction of misunderstanding between stakeholders about what could be the future of LGS. The MF approach also enabled the participation and dialogue that underlines the positive outcomes and interactions of embedding a large spectrum of stakeholders when dealing with reinforcement of territorial sustainability with the contribution of livestock farming activities.

Transversal analysis of multifunctionality

The opportunity to apply the multifunctionality common framework to a global range of contexts has demonstrated the power of the approach. Table 1 summarizes the indicators used in the case studies and the results of multifunctionality-based local debates and analysis.

Creating a space and process for multi-stakeholders to hear, respond and decide

In all cases the MF framework provided a common language and forum to make transparent the world views of the participating stakeholders, and through this for them to come to a common understanding of management, policy and adoption of management practice. This was aided by the defining of local indicators ascribed to each of the four dimensions of the framework to account for the context and the diverse world views of stakeholders. The choice of indicators and the inclusion of the stakeholders in the process ensured that the process was relevant for the context. To populate the diversity of indicators requires a range of qualitative and quantitative methods, to gain a baseline and then to test the impact of policy and management options. Gaining data is not always easy and requires the use of a range of expertise to populate and analyse the information.

Multifunctionality LGS conceptual model applicable to a variety of contexts

The cases show (Table 2) that the MF framework is operational and relevant to a diversity of contexts and issues. Nevertheless, the processes and tools developed and designed may be as diverse as workshops, brainstorming, surveys, participatory films, action research processes, participatory simulation models to analyse and identify the four dimensions, their entities and processes and their indicators. The heuristic significance of our approach relies in maintaining a consistency between its relevance for each case study as well as its contribution to global debates on livestock farming facing climate change, biodiversity erosion, food security and poverty and alleviation of inequities. In Europe, where past development of livestock systems was mainly driven by economic and some main environmental concerns (pollution, climate change), rethinking livestock development through its contribution within territories to social interactions and solidarity, cultural life, biodiversity conservation, economic networks and infrastructures would be facilitated using this MF framework. Following the monogastric model, many herbivore-farming systems have been unplugged from the local resources as a result of using industrial livestock feed often supplied by components coming from abroad and assessed by only its economic efficiency. These livestock systems have thus lost their links with their social and ecological environment and are more and more criticised by their neighbours as well as by environmental or animal activists. Such communities have lost the link with domestic animals, which is part of the Western culture, considering their environment as wild and forgetting that most of the European landscapes have been produced by centuries of livestock husbandry and cannot be maintained without it! Our purpose, by using the

Table 1. A summary of the indicators used in the case studies and the results of the application of the multifunctional approach.

Case	Social indicators	Environmental indicators	Production indicators	Local development indicators	Results
Argentina; the Puna high altitude, dry pastoralism	household members; number of local organisations in supply chain	biodiversity; plant cover; dry matter production	kg meat sold; diversity of livestock; drought strategies	annual income; number and diversity of marketing channels	Strategies for resilience based on social networks and diversity of livestock species related to the local supply chain and household participants linked to local wealth generation. The grazing system maintains vegetation condition and diversity with cover regulating soil temperature and water.
Brazil; Maranhao, silvo-pastoral systems	employment; profit	biodiversity; animal welfare	kg meat ha^{-1} $year^{-1}$	number of businesses	Greater profit achieved compared with monoculture, with potential for further gain with payments for additional ecosystem services provided, i.e. increased biodiversity of flora and fauna, and enhanced soil conservation. Animal welfare was enhanced.
Senegal; Ferlo, rangeland based dairy milk platform	social inclusion; collaboration between forage producers	biomass production	biomass flows; efficiency of milk collection; litres of milk day^{-1}	networks of biomass supply; milk income; milk value chain development	Exploration of three scenarios of dairy intensification identified the trade-offs between outputs and inputs and social and environmental consequences and assisted in sector strategy development.
Mongolia; Bulgan forest steppes, conservation coexisting with livestock systems	household income; participating families; diverse employment	increased numbers of existing species; reintroduction of species	livestock production	new business opportunities	The positive uptake by herder households of conservation related employment alleviated poverty and improved environmental outcomes while not diminishing existing livestock systems.
Vietnam; Dien Bien, mountain beef systems development	household income; social networks; insurance; cultural activities; employment	soil organic matter and fertility	percentage of beef supplied inputs	profit going to actors in the value chain	Results showed the contribution that extensive beef production brings to the household, community, and local development in comparison to other livestock systems and cropping activities.
China; Qinghai plateau, conservation with livestock systems	tibetan buddhism cultural relationship with nature	landscape heterogeneity indices; bird biodiversity	yak grazing intensity		The landscape mosaic created by yak grazing had a positive impact on bird species richness. Extensive pastoralism and related culture coexist with improved environmental outcomes.
France; PACA, agro pastoral systems in Mediterranean mountain area	still to be defined, using the simulation model, in interacting loops between local stakeholders	same	same	same	Identifying relevant actors and activities LGS have to interact with to foster sustainability of socio ecological system; identify processes and properties of LGS putted into questions and identification of levers of public actions to be settled.

Multifunctionality framework, aims to formulate scientific evidence about the other dimensions linked to livestock grazing systems in the diverse faces of their environment. The diversity of cases above show how this Western story is at work in many other parts of the world, generating tensions between increasing the production, specializing the workers, changing the breed, seeking for markets and the traditional place animals have in the family or the community (like in Senegal or Vietnam). The Argentinian, Tibetan and Mongolian cases illustrate the importance of these links on which the social dimension is based and that only slight and cautious changes are introduced. On the other hand, when changes have already happened, as in Brazil and France, people are seeking new arrangements between livestock farming and their human and ecological environments. The Multifunctional framework allows in this way to understand the complexity of each situation and what makes it able to change, by mobilizing the same levers but differently. It allows us to overcome the fact that each situation is different; yes, they are, but following a common framework which represents the essence of livestock farming all over this world.

Supporting sustainability through different scales

The MF framework has also shown its robustness when applied to different scales, household (Puna), farm (Brazil), landscape (Tibet, France), local (Mongolia) and sector (Vietnam, Senegal) and different socio-ecological contexts ranging from communal, migratory, individual and sedentary systems. In the discussion processes among stakeholders, it appeared clearly that multiple scales must be managed and represented to build a holistic and collective understanding of system and territorial sustainability.

MF framework to articulate activities in territories

Our target was to build a strong common conceptual framework in order to overcome the singularities of each case study in order to demonstrate the role of LGS beyond the strict animal production. It confirms that everywhere in such contrasted situations, LGS is not an isolated activity, as some other economic activity could be: LGS, due to its large landscape footprint is closely linked to a specific area, which provide its resources but which is also used by other stakeholders. Sustainable management of territories needs articulating and facilitating synergies between activities and sectors in order to collectively design

Table 2. A summary of the utility of the multifunctional (MF) approach by case.

Case	Utility of Multifunctional approach
Argentina, Puna	The resilience and adaptive capacity of the Puna herders at the household and community level was able to be explored through the application of the four dimensions. Successful implementation of the approach required a multidisciplinary team which for this context was not the norm, thus building the capacity to tackle such complex socio-ecological issues.
Brazil, Maranhao	Exposing a range of actors including students, farmers and agribusiness to the holistic analysis using the MF approach of the silvo-pastoral system has had a positive impact on adoption of practices by farmers. Students have gained a greater understanding of the complexity of the system and how it works.
Senegal, Ferlo	Building and using a simulation model based on the MF approach contributed to a facilitated dialogue between stakeholders to find solutions to share resources and find synergies between actors and biomass fluxes.
Mongolia, Bulgan	The development of local indicators was key in ensuring the MF approach was relevant. In this case, not all four dimensions were equal with greater emphasis being on the social and environmental. The approach was appreciated by planners to assist in development of policy.
Vietnam, Dien Bien	The MF framework facilitated dialogue between actors based on common indicators showing the complementarity of different agricultural systems toward sustainable development of the territory and reaching the objective of food supply for the population
China, Tibet	The MF approach demonstrated that the Kobresia grasslands are a cultural landscape where nature and society interact to the benefit of the environment and the wellbeing of people.
France, PACA	The MF approach allows to identify main relevant dimensions of LGS putted into questions by local actors to contribute to the sustainability of local socio ecosystem and it promotes local device to settle dialogue and allow identification of levers to foster mid-term co-evolution

the future for which the Multifunctionality framework helps to organize discussions on priorities and trade-offs.

Managing diverse points of views and trade-offs

The cases demonstrate the relationship between the dimensions and the dilemmas involved in attempting to deliver a balanced outcome across the different dimensions. What is very clear is the multiple functions LGS deliver and how this delivery is mediated through human intervention. No longer is it acceptable to focus only on productivity or environment alone when considering these systems, but to acknowledge, value and respect the interrelated multiple functions.

The multiple functions of LGS are still present but fragile

The diversity of cases analysed show that in most of the contexts where traditional LGS are in place, the diversity of functions within the four dimensions are really operating (see list of indicators identified) and support the viability and sustainability of the socio-ecosystems. But it appears also that faced with economic and policy dynamics, some of these functions might be endangered calling into question the sustainable future of an important part of the local society and even of the environment.

Conclusions

The MF framework applied to a diversity of livestock grazing systems has shown at the landscape level the existence of strong and operational interactions between production, social, environmental and local development impacts that support the sustainability of these socio-ecosystems. This interweaving of functions has the opportunity to identify the policy and practice to be prioritized to ensure that all are achieved simultaneously and equitably. Central to the delivery of these functions are people and their wellbeing and associated institutions. As we address the issues related to food sovereignty and security, we can take a holistic approach as demonstrated in these cases to align land governance, resource access, cultural identity and rural livelihoods. This is a means to secure sustainable food systems (SFS), including livestock grazing systems, well rooted in territories through multi-sectorial synergies, delivering local goods and services but oriented toward larger value chains and trade.

This brief round-the-world trip illustrates as well the diversity of LGS in different geographical, historical and political contexts also its consistency as a human ancestral activity based on our societies' interactions with the natural world through the mediation of domestic animals. Considering herbivores, this has generated a diversity of breeds, each of them well adapted to the environment in which their breeders are living, allowing them through multiple interactions with their environment to adapt their practices to the availability of resources, diversity and variability in space and time. However, in most industrialized countries – but not only – we notice a strong homogenizing dynamic, particularly in cattle, and standardization of breeding conditions considering only how to optimize meat or milk production and forgetting the other livestock functions which start to be contested by several social movements. Alternatives and new pathways are sought to overcome this industrialized vision of livestock farming, but in a context that has changed and could generate new conflicts as the French case illustrates. Taking advantage of what has been illustrated in other parts of the world about livestock farming thanks to our common framework, we developed a very systemic and dynamic point of view. LGS is at the core of the links between human societies and the natural world: it is still obvious when it is close to a traditional situation like in Argentina, Tibet and Mongolia, it needs careful management when the process of change is ongoing, like in Senegal and Vietnam, and it has to be rebuilt when the transformation has been done, and is not considered as plenty satisfying, like in Brazil and France. Thus can we in Europe reverse this global standardization starting from this only Western model and take advantage of the lessons from the Global South, as illustrated in this paper, to reinvent and redesign multifunctional and sustainable LGS, well integrated and adapted to the diversity of territories in our continent?

References

Alcoba L.N., Chavez, M.F. and Vittar, M.C. (2018) *Las Carniceras intermediarias. Sujetos clave en la trama socio-productiva y comercial de la carne de llama, cordero y cabra de la Puna y Quebrada jujeñas*. Ed. INTA IPAF REG NOA, Buenos Aires, Argentina, 76 pp.

Bourgoin, J., Corniaux, C., Touré, L. and Cesaro, J.D. (2018) *Atlas des dynamiques observées dans le bassin de collecte de la Laiterie du Berger*. CIRAD, 32 p.

Caron, P., Reig, E., Roep, D., Hediger, W., Le Cotty, T., Barthélemy, D., Hadyńska, A., Hadyński, J., Oostindie, H.A. and Sabourin, E. (2008) Multifunctionality: refocusing a spreading, loose and fashionable concept for looking at sustainability? *International Journal of Agricultural Resources, Governance and Ecology* 7(4/5), 301-318.

Cesaro, J.D., Vall, E., Gaye, M., Müller, J.P., Diaw, A. and Ickowicz, A. (2020) *Estimation spatialisée du potentiel laitier autour de Richard-Toll et scénarios d'évolution des systèmes de collecte*. CIRAD 10 p.

Chara J., Barahona R., Murgueitio E., Debilitz K., Reyes E., Mauricio R.M., Molina J.J. and Flores M.X. (2017) Integrating landscapes: agroforestry for biodiversity conservation and food sovereignty. In: Montagnini F. (ed.). *Integrating Landscapes: Agroforestry for Biodiversity Conservation and Food Sovereignty. 1st ed*. Springer International Publishing, Cham, Switzerland, pp. 1-22.

Corniaux C., Alary V., Gautier D. and Duteurtre G. (2012) Producteur laitier en Afrique de l'Ouest: une modernité rêvée par les techniciens à l'épreuve du terrain. *Autrepart* (3), 17-36.

Delay E., Müller J.P., Cesaro J.D., Valls-Fox H. and Ickowicz A. (2021) Empowering a model for improved multi-actor discussions: case study from Dairy production in Sahelian agro-pastoral systems. In: *1st conference GAMA Days 2021*.

Echenique M., Chavez M.F., Vittar M.E. and Longoni, A. (2015) *Análisis de la dinámica del stock ganadero de la Puna de Jujuy, su importancia productiva actual, problemáticas y posibles techos productivos*. Ed. INTA, San Salvador de Jujuy.

FAO (2018) *World livestock: Transforming the livestock sector through Sustainable Development Goals*. FAO, Rome, Italy.

Fritz A., Li L., Storch I. and Koch B. (2018) UAV-derived habitat predictors contribute strongly to understanding avian species–habitat relationships on the Eastern Qinghai-Tibetan Plateau. *Remote Sensing in Ecology and Conservation* 4(1), 53-65.

Garde L., Dimanche M. and Lasseur J. (2014) Permanence and changes in pastoral farming in the southern alps. *Journal of Alpine Research* 102 (2). https://doi.org/10.4000/rga.2416

Hervieu B (2002a) La multifonctionnalité de l'agriculture: genèse et fondement d'une nouvelle approche conceptuelle de l'activité agricole. *Cahiers Agriculture* 11, 415-419.

Hervieu B. (2002b) La multifonctionnalité: un cadre conceptuel pour une nouvelle organisation de la recherche sur les herbages et les systèmes d'élevage. *Fourrages* 20, 219-226.

Hilbig, W. (1995) *The vegetation of Mongolia.* SPB Academic Publishing, Amsterdam, the Netherlands.

Huang J., Tichit M., Poulot M., Darly S., Li S., Petit C. and Aubry C. (2015) Comparative review of multifunctionality and ecosystem services in sustainable agriculture. *Journal of Environmental Management* 149, 138-147.

Ickowicz A., Blanfort V., Forman S., Hubert B., Lasseur J., Marguerat C., Tourrand J-F., VelascoGil G. and Wedderburn L. (2018) Why and how address multifunctionality of pastoral farming systems? In: Blanfort V. and Lasseur J. (eds.) *Multifunctionality of pastoralism: linking global and local strategies through shared visions and methods*. International Rangeland Congress Workshop 2016. GASL, Montpellier, France, 127 pp.

ILRI, IUCN, FAO, WWF, UNEP and ILC (2021) *Rangelands Atlas*. ILRI, Nairobi, Kenya, 42 pp.

IRIM (2021). Conserving forest steppe ecosystems and endangered species through community-led network: Endline study report. Retrieved from Ulaanbaatar.

Lasseur J. and Dupré L. (2017) Entre production d'agneaux, incitations environnementales et inscription dans le local: débats au sein du monde professionnel et reconfigurations des activités d'élevage dans les Alpes du Sud. *Pour* 231, 157-165.

Li J., Wang D., Yin H., Zhaxi D., Jiagong Z., Schaller G.B., Mishra C., Mccarthy T.M., Wang H. and Wu L. (2014) Role of Tibetan Buddhist monasteries in snow leopard conservation. *Conservation Biology* 28(1), 87-94.

Li L., Tietze D.T., Fritz A., Lü Z., Bürgi M. and Storch I. (2018) Rewilding cultural landscape potentially puts both avian diversity and endemism at risk: A Tibetan Plateau case study. *Biological Conservation* 224, 75-86.

Marshall-Stochmal F., Schulte to Buhne H., Lopes M., Merson S., Ganbaata, E. and Pettorelli N. (2020). Assessing changes in land cover and vegetation productivity for threatened forest-steppe ecosystems: a case study in Arkhangai province, Mongolia. *Mongolian Journal of Biological Sciences* 18(1).

Mauricio R.M., Ribeiro R.S., Paciullo D.S.C., Cangussú M.A., Murgueitio E., Chará J. and Estrada M.X.F. (2019) Silvopastoral systems in Latin America for biodiversity, environmental, and socioeconomic improvements. In: Lemaire G., Carvalho P.C.D.F., Kronberg S. and Recous S. (eds.) *Agroecosystem diversity: reconciling contemporary agriculture and environmental quality*. Elsevier, Amsterdam, the Netherlands, pp. 287-297.

Miehe G., Schleuss P.-M., Seeber E., Babel W., Biermann T., Braendle M., Chen F., Coners H., Foken T. and Gerken T. (2019) The Kobresia pygmaea ecosystem of the Tibetan highlands – Origin, functioning and degradation of the world's largest pastoral alpine ecosystem: Kobresia pastures of Tibet. *Science of the Total Environment* 648, 754-771.

Molina, D. (2011) *Sistema de Pastoreo y Estado Actual de los Recursos Vegetales en una Comunidad de la Puna salteña. El Caso de Cobres – Departamento La Poma*. Tesis de Grado, FCN, Universidad Nacional de Salta.

Mottet A., de Haan C., Falcucci A., Tempio G., Opio C. and Gerber P. (2017) Livestock: on our plates or eating at our table? A new analysis of the feed/food debate. *Global Food Security* 14, 1-18.

Mottet A., Teillard F., Boettcher P., DeBesi G. and Besbes B. (2018) Review: Domestic herbivores and food security: current contribution, trends and challenges for a sustainable development. *Animal* 12 (s2), 188-198. https://doi.org/10.1017/S1751731118002215

National Development and Reform Commission (2013) The second phase planning of the Qinghai Sanjiangyuan ecological protection and construction project.

NSO (2020) National Statistical Information Service. Available at: https://www.1212.mn/stat.aspx?LIST_ID=976_L10_1

Paz R., Lamas H., Echazú F., Sosa Valdéz F. and Califano L. (2011). *Diversidad, mercantilización y Potencial productivo de la Puna jujeña (Argentina).* Ed. INTA – EEA Abra Pampa.

Qinghai Provincial Government (2021) 14th five-year plan for the National Economic and Social Development of Qinghai Province.

Quiroga Mendiola M. (2015a) '*Donde no se puede sembrar*.' La triple espacialidad pastoril en Suripujio, Puna de Jujuy, Argentina. In: Benedetti A. and Tomasi J. (eds.) *Espacialidades altoandinas. Nuevos aportes desde la Argentina. Tomo I: Miradas hacia lo local, lo comunitario, lo doméstico*. Pp: 227-256, Editorial de la Facultad de Filosofía y Letras de la UBA, 352 pág.

Quiroga Mendiola M. (2015b) Pastores de la Puna: manejo sustentable de pastizales naturales en las tierras altas de Jujuy, Argentina. In: Martínez Carretero, E. (ed.) *Restauración ecológica en la diagonal árida de la Argentina 2.* 1ra ed., pp. 159-178.

Quiroga Mendiola M. and Cladera J. (2018) Ganadería en la Puna argentina. In: Grau R., Izquierdo A., Babot, J. and Grau A. (eds.) L*a Puna argentina geografía y ecología natural y cultural*. Serie de Conservación de la Naturaleza 24. Ed. Fundación Miguel Lillo, San Miguel de Tucumán, Argentina.

Quiroga Mendiola M., Briones V., De Gracia J. and Sánchez N.E. (2010) *Conservación y pastoralismo: Experiencias de Investigación Participativa en el Parque Nacional Los Cardones, Valles Calchaquíes, Argentina*. Ed. Universidad Nacional de Salta, 204 pp.

Quiroga Mendiola M., Saravia A.G.Y and Longoni A. (2018) Andean mutual breeding, multifunctional services and pastoral strategies against drought in the Argentine Puna Grasslands. In: Blanfort V. and Lasseur J. (eds.) 2018. *Multifunctionality of pastoralism: linking global and local strategies through shared visions and methods*. GASL Action Network 'Restoring value to grassland, GASL, Montpellier, France, pp. 90-97.

Robinson T.P., Thornton P.K., Franceschini G., Kruska R.L., Chiozza F., Notenbaert A., Cecchi G., Herrero M., Epprecht M., Fritz S., You L., Conchedda G. and See L. (2011) *Global livestock production systems*. Food and Agriculture Organization of the United Nations (FAO) and International Livestock Research Institute (ILRI), Rome, Italy, 152 pp.

Steinfeld H., Gerber P., Wassenaar T., Castel V., Rosales M. and de Haan C. (2006) Livestock's long shadow. environmental issues and options. Food and Agriculture Organization of the United Nations (FAO), LEAD initiative, Rome, Italy. 390 pp.

Tews J., Brose U., Grimm V., Tielborger K., Wichmann M.C., Schwager M., *et al.* (2004) Animal species diversity driven by habitat heterogeneity/diversity: the importance of keystone structures. *Journal of Biogeography* 31(1), 79-92.

UNCED (1992) Agenda 21d an action plan for the next century. United Nations Conference on Environment and Development, New York, NY, USA.

Wedderburn L., Ickowicz A., Mauricio R.M., Quiroga Mendiola M., Blanchard M., Le Thi Thanh H., Hubert B., Lasseur J., Blanfort V. and Müller J-P. (2021) Restoring value to grassland initiative: to maintain the environmental and economic value of grasslands and to promote their social and cultural functions. Joint XXIV International Grassland Congress XI International Rangeland Congress, 25-29 October 2021, Nairobi, Kenya.

Zahm F., Viaux P., Vilain L., Girardin P. and Mouchet C. (2008) Farm sustainability assessment using the IDEA method. From the concept of farm sustainability to case studies on French farms. *Sustainable Development* 16, 271-281. https://doi.org/10.1002/SD.380

Zhang L., Fan J., Shao Q., Tang F., Zhang H. and Li Y. (2014) Changes in grassland yield and grazing pressure in the Three Rivers headwater region before and after the implementation of the eco-restoration project. *Acta Prataculturae Sinica* 23(5), 116-123.

ZSL (2021). ZSL Mongolia Representative Office Report 2018-2020 (in Mongolian). Retrieved from Ulaanbaatar.

Coupling the benefits of grassland crops and green biorefining to produce protein, products and services for the green transition

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Abstract

Biorefining forages can produce protein that can substitute, e.g. to replace imported soybean meal. Forage crops can deliver high yields of biomass as well as protein with a well-balanced amino acid profile. In grass from unfertilized permanent grassland, focus has to be on the fibre part of the grass due to a low protein yield. Changing from annual crops to perennial grassland will decrease N-leaching and greenhouse gas emissions. With current techniques, 40% of the forage protein can be recovered in a protein concentrate with around 50% protein. In addition, a fibre fraction containing 15-18% protein can be produced and used as ruminant feed, bioenergy, or further biorefined into chemical building blocks or bio-materials. Experiments have shown that biorefined grassland protein can substitute for soybean meal for poultry and pigs without negative effects on animal performance. The fibre fraction seems suitable for ruminant feeding. The first industrial scale biorefineries on green biomass for feed and bioenergy are now established in Denmark, while more research is needed to evaluate the protein quality for food applications, and in addition a full EFSA approval. The green biorefinery concept opens new markets for grassland and opportunities for increasing grassland area and achieve the connected ecosystem services.

Keywords: sustainability, perennial grasses and clover, biorefining, protein, fibre

Introduction

A fully developed bioeconomy will require a complete utilization of agricultural biomass, not only for food and feed but also for chemical compounds, fibres and bioenergy. This will require both a larger total biomass production and higher utilization of residues. However, in Europe, it may be difficult to increase productivity in existing cropping systems without also increasing environmental impacts, and the concept of 'sustainable intensification (more with less)' is contested in much recent literature. Van Grinsven *et al.* (2015) proposed to focus on 'sustainable extensification (less with less)' in Europe. In contrast, Jørgensen and Lærke (2016) proposed a change in cropping systems for Northwestern Europe from annual crop rotations into grassland, which holds the potential for increasing biomass yield, reducing environmental impact, and a European production of protein to substitute the high current import of soy. This would support the EU Protein Strategy that otherwise has a focus on increased protein seed production (EU Commission, 2018).

The idea of utilizing leaf-protein-concentrates as a protein source for animal or human consumption is not new; it dates back to the early 20th century where pioneering efforts led to significant amounts of research and pilot-scale development (Pirie, 1942). Throughout the 20th century and well into the 21st there have been multiple attempts and supporting research to facilitate commercial success of green biorefineries in Denmark (Pedersen *et al.*, 1979) and internationally (Chiesa and Gnansounou, 2011; Houseman and Connell, 1976; Näsi and Kiiskinen, 1985; Pirie, 1978; Pisulewska *et al.*, 1991). However, these early evaluations did not value the environmental benefits by changing cropping systems, utilizing surplus grasslands and substituting imports of soy products from other continents with high carbon footprints. Such environmental effects have attained much higher political focus over the last decades and national and EU legislation, such as the Water Framework Directive, Nitrate Directive and recent climate policies, stipulates the needs for improvements.

The combination of the techno-economic and environmental potential of producing high carbon capture in grasslands with the recent developments in biorefinery techniques is the novelty of the concept of 'Green Biorefinery'. Our aim is to develop and document win-win solutions with positive business economy, environmental benefits, no or negative iLUC, and improved self-sufficiency of protein concentrates, as a novel opportunity to solve several of our grand challenges in a sustainable way.

The development of new crop production systems combined with green biorefineries is not just about technical development of the production circle. It is also important to discuss the total land-use in relation to societal demands for environment, climate, recreation and biodiversity. This discussion has been supported by several land-use and technology scenarios (Gylling *et al.*, 2016; Larsen *et al.*, 2017; Mortensen and Jørgensen, 2022). They show that the bioeconomy may contribute significantly to additional reductions in nitrate leaching and greenhouse gas emissions but the scale of reductions depends a lot on the way agriculture is combined with the biobased energy and material sector. The development of the land-use towards either sustainable intensification or extensification and a higher share of nature has shown important determinants for the potential size of the bioeconomy and for emission reductions.

Grassland crops are the most sustainable agricultural crops, however, with a limited market

Compared with annual grain and seed crops, the production of perennial grassland crops reduces significantly the losses of nutrients, the need for pesticides, and it supports soil carbon build-up (Cadoux *et al.,* 2014; Chen *et al.,* 2022; Manevski *et al.,* 2018; Pugesgaard *et al.*, 2015). Even though water quality will be improved by perennial cropping systems with longer growing season, water quantity (surplus for ground water and river discharge) may be reduced due to a higher annual evapotranspiration than from annual crops. However, there is an increased water infiltration capacity in perennial compared with annual crops due to more macropores (Franzluebbers *et al.*, *2*014), which can reduce the loss by water run-off. The potential for securing a water supply for a long growing period is highest in humid Northwestern Europe. Here, grasses and legumes can capture solar radiation more efficiently than annual grain and seed crops, in which a considerable part of the growing season is used for crop ripening, harvest, soil tillage and sowing (Cadoux *et al.*, 2014; Dohleman and Long 2009; Pugesgaard *et al.*, 2015). Accordingly, Manevski *et al.* (2017) measured an interception of approx. double the amount of Photosynthetically Active Radiation in perennial grassland compared with in annual cropping systems, which translated into approx. double the amount of crop harvest in the grasses (Figure 1). Tall fescue and *Festulolium* varieties seem to be the most productive grasses at Northwestern European latitudes (Becker *et al.*, 2020; Cougnon *et al.*, 2017).

It is not new knowledge that grasslands are environmentally and climate friendly. However, the problem has been (1) that these ecosystem benefits are not economically valorised, and (2) that market outlets for increased grassland areas were almost limited to the ruminant sector. There is some use of grass for biogas production (Pehme *et al.*, 2017) but to avoid severe indirect land-use-change effects there is a need for envisaging also new food products from grasslands in order to harvest more of the ecosystem services associated with them. The extraction of protein to feed monogastric animals or for direct human consumption will open a market for more grassland, which will increase carbon sequestration per land unit and lower environmental impacts.

Perennial grass and legume crops on intensive arable land

Intensive grass production on arable land can be managed to optimize protein concentration and quality for extraction in a biorefinery. Numerous factors influence protein content, extractability and yield per ha. Some of the most important are plant species, harvest time, fertilization, and leaf/stem ratio. Research at Aarhus University has investigated the quality of protein with regard to its availability to animals using

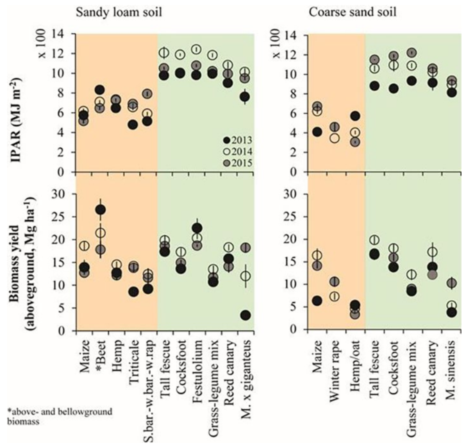

Figure 1. Interception of photosynthetically active radiation (IPAR) in annual (orange shade) and perennial (green shade) crops during 2013-2015 on two soil types at AU (Manevski *et al.*, 2017).

the Cornell Net Carbohydrate and Protein System (CNCPS) (Solati *et al.*, 2017; Thers *et al.*, 2021). With regards to plant species, total protein recovery into concentrate was highest for the legumes (Thers *et al.*, 2021) but this may depend on grass fertilization optimization. Solati *et al.* (2017) found that the estimated extractable protein (g kg^{-1} dry matter (DM)), defined as the sum of the easily available protein fractions B1+B2, was significantly highest in white clover and alfalfa (Figure 2C). However, if in addition the more cell wall attached protein fraction B3 can be extracted, white clover had the highest extractable protein content amongst all species (Figure 2D). Due to the higher biomass productivity of red clover, it showed the highest productivity of protein per ha.

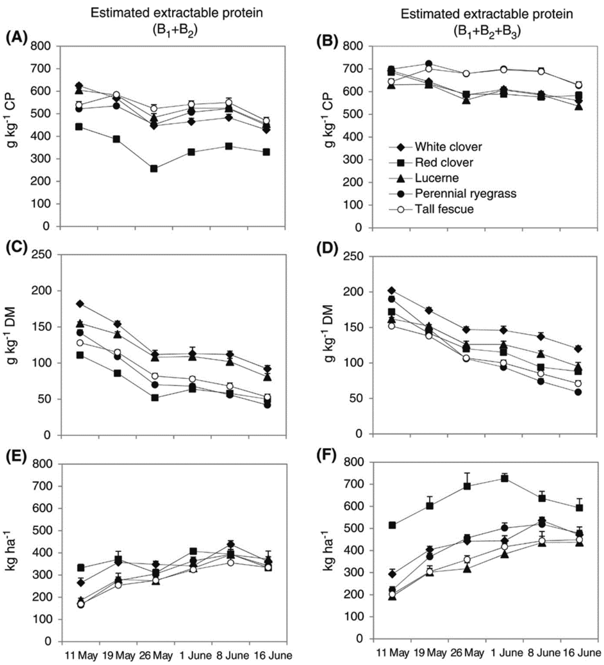

Figure 2. Extractable protein around the first cut in spring defined as easily available (B1 + B2) (A, C, E) and potentially extractable (B1 + B2 + B3) (B, D, F) in legume and grass species (Solati *et al.*, 2017).

The chemical composition, and in particular the protein content, depends on N fertilization. In grass-clover mixtures, N fertilization does not influence total protein yield much, while the yield of protein in pure ryegrass increases significantly with increased N-fertilization (Jørgensen *et al.,* 2021). Thus, the protein to carbohydrate ratio is high in grasses that are cut frequently and supplemented with N fertilizer, while protein content in grass-clover only varies a little depending on N fertilization.

Perennial grassland in an extensive production system

If long-term grasslands are not fertilized, only a very moderate DM yield of 2-4 t ha^{-1} $year^{-1}$ can be expected after a few years of harvest (Nielsen, 2012). In addition, grass from unfertilized meadows usually has low nitrogen and protein concentrations and is therefore not suitable for protein extraction. Alternatively, the use of the grass biomass for biogas production can be considered, and this may present positive LCA-results, if there is no alternative use of the grassland, e.g. animal grazing (Pehme *et al.*, 2017). Another option supporting the bioeconomy is to use the fibres from grasses with low protein content for paper, packaging, animal bedding, biochar, etc. (www.go-grass.eu).

The attainable yield of permanent grassland on organic soils depends on type of species and cultivars, sward age, annual harvest frequency and fertilization rates (Nielsen, 2021). On well-drained areas, fertilized permanent grassland is expected to produce the same yield as grass in rotation for several years after establishment. However, if not well-drained, the typical DM production is estimated to be between 70 and 80% of grass in rotation (Nielsen, 2012). The cultivation of flood-tolerant species, e.g. reed canary grass, *Festulolium* and tall fescue on wet or temporarily flooded organic soils, also known as paludiculture, has documented high annual yields of up to 10-19 t DM ha^{-1} (Jørgensen *et al.*, 2021; Kandel *et al.*, 2013, 2016; Nielsen *et al.*, 2021b). This is comparable to productivity of grass in rotation on drained soils under similar fertilization rates of 160 – 240 kg N ha^{-1} $year^{-1}$.

For biorefining, the protein content in grass biomass depends on nitrogen availability, frequency and timing of cutting, similar to the systems on intensive arable land. Recent research has found crude protein contents of up to 2.9-3.4 t ha^{-1} $year^{-1}$, and precipitated protein concentrates of up to 1.2-2.2 t ha^{-1} $year^{-1}$, for reed canary grass and tall fescue, cultivated on wet organic soils (Nielsen *et al.*, 2021a). Optimal timing of harvest seems to remain the most critical factor for biomass and protein yields.

Green biorefining and its main products

Green Biorefining is a fundamental concept that 'represents the sustainable processing of green bio-mass into a spectrum of marketable products and energy' (McEniry and O'Kiely, 2014). In other words, Green Biorefining is a technology platform that integrates a variety of different sustainable solutions in order to produce everything from food and feed to biomaterials, biofuels and bioenergy. The Green Biorefining has an inherent focus on products containing proteins or amino acids, which is due to the high protein productivity of green crops.

In order to utilize the high protein content of green biomass for monogastric animal feed, an efficient separation process platform is needed. Several unit operations and steps are involved in the processing of fresh green biomass, before the desired protein concentrate can be separated. The major steps involved are shredding/maceration, fractionation, precipitation and separation. An overview of these process steps and the protein separation platform is presented in Figure 3.

The yields and mass distribution between the different processing streams depends on a long list of parameters and can vary to a large extent. Figure 4 shows the typical ranges of DM and crude protein yields following a Green Biorefining separation process like the one in Figure 3. Depending on the processing conditions, the extractability of the protein in the green biomass and the efficiency at the

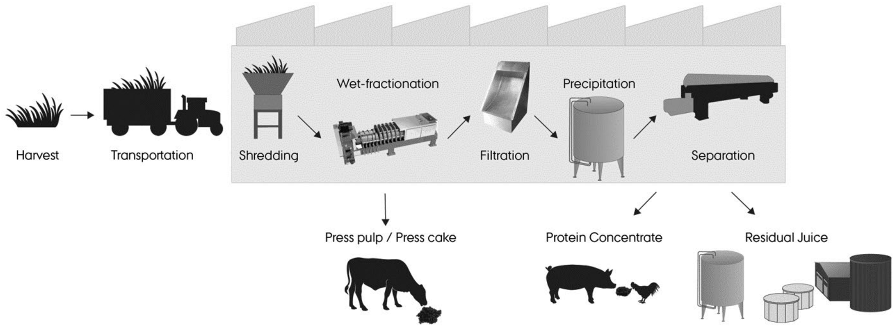

Figure 3. The green biorefinery protein separation platform. Unit operations and process steps involved in separation of protein from green biomass in the green Biorefining platform (Source: Jacobsen, 2020)

biorefinery, 30-50% of the biomass DM and up to 40-60% of protein will be pressed out in the juice fraction, leaving the remaining fractions.

From the juices fraction, 10-20% of the original DM and 20-60% of the original protein can be precipitated and end in the precipitated protein-rich fraction, and the rest goes to the residual juice (Damborg *et al.*, 2020; and also unpublished results from L. Stødkilde). These ranges of mass and protein distribution are not ultimate but illustrate the possibilities for optimization of the process according to what the desired outcome is with respect to protein yield and process cost.

The development in Denmark during recent years has been focused around the processing of fresh green biomass, as opposed to processing of silage grass. The main products in focus have been a protein-rich concentrate that can substitute soybean meal in feed mixtures for monogastric animals, a press cake fibre-rich product for ruminant feed and/or biogas production, and a residual juice for biogas and nutrient recycling.

Protein

The protein fraction is considered the most valuable of the three main products, and much focus has been on increasing the amount of protein isolated and on the concentration and nutritional quality of this protein. Protein concentration in the green protein concentrates that were initially around 35% of

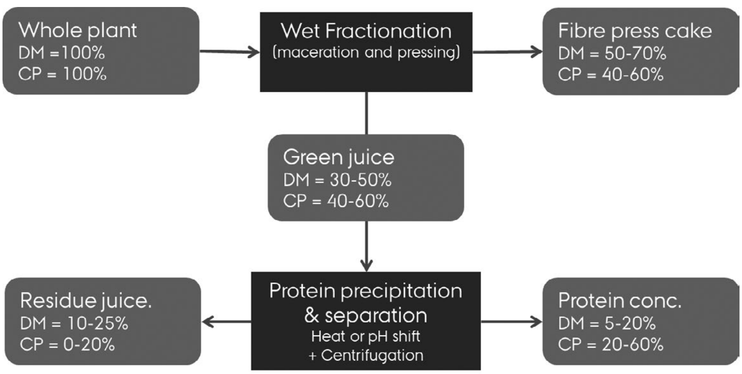

Figure 4. Schematic overview and typical dry matter (DM) and crude protein (CP) yields in the fractionation of green biomass into the three process streams of fibre press cake, residual juice and protein concentrate. The numbers are mass balance % (weight per weight in input material) (M. Ambye-Jensen and estimates from Damborg *et al.*, 2020).

DM has now been improved to more than 60% protein, and the nutritional quality of the protein has increased concurrently (Stødkilde *et al.*, 2021). Table 1 shows the chemical composition of green protein concentrates with 46 and 56% protein in DM (Stødkilde *et al.*, 2021).

The amino acid composition in both green protein batches in Table 1 are similar to the amino acid composition in soybean meal, while the lipid content is highest in the green protein. It is worth noting that roughly 50% of the fatty acids in the green protein consist of alpha-linolenic acid (C18:13n3). In addition, the fibre content is high in the green protein. In agreement with previous rat studies, where it was shown that the digestibility increased with increasing protein content in the green protein (Stødkilde *et al.*, 2019), the *in vitro* digestibility (EDOMi) was higher in the green protein with the highest protein content, but still lower than in soybean meal.

Regarding the nutritional value of the protein as feed for pig and poultry, the first Danish feeding experiment was performed with organic broilers fed with a relative low crude protein diet containing concentrate (36% CP). With this protein concentrate it was possible to substitute 8% of the diet, primarily soy press cake, (13% of the CP) with protein concentrate extracted from organic grass-clover, without affecting growth performance (Stødkilde *et al.*, 2020). However, larger inclusions challenged feed intake and growth rate due to the low protein content and the correspondingly high content of insoluble dietary fibres in the protein extract, which to a large extent are indigestible.

In addition, two feeding experiments have been performed with growing-finishing pigs with a protein concentrate containing 46 and 56% protein (Table 1), respectively. Both protein concentrates showed a well-balanced amino acid composition, slightly lower in lysine than soybean but higher in most other essential amino acids. With these concentrates, pigs performed equally, irrespective of inclusion rate (Stødkilde *et al.*, 2021). The highest inclusion rate of 15% of the traditional feed (up to 41% of the crude protein) with grass-clover protein still secured a feed intake, growth and feed utilization similar to a control group with soybean meal as the dominating protein source. In both experiments, daily weight gains above 1000 g were achieved.

In both of the broiler experiments the n3 fatty acids increased from 6.7 to 11.8% of total fatty acids in breast meat, and in the pig experiment the increase was from 1.17 to 3.12% of total fatty acids in *Longissimus dorsi* (Stødkilde *et al.*, 2020, 2021).

Table 1. Chemical composition of green protein with 46 and 56% protein respectively, and dehulled soybean meal.

On DM basis	Protein, 46%	Protein, 56%	Soybean meal
DM, %	97.4	92.32	87.2
Crude protein, %	45.8	56.2	52.4
Lipids, %	10.6	13.8	2.9
Ash, %	12.1	8.30	8.14
Total dietary fibre, %	29.7	n.a.[1]	n.a.[1]
Amino acids, g/16 g N			
Lys	5.76	5.75	6.29
Met	2.27	2.03	1.36
Met + Cys	2.73	2.72	2.79
Thr	5.02	4.60	4.06
Trp	2.42	2.21	1.38
EDOMi[2], %	67.9	72.8	77.8

[1] n.a. = not analysed.

[2] EDOMi = enzyme digestibility of organic matter at the ileum.

Regarding the possibilities for using the protein in human food, a recent review concluded that high quality leaf protein may be incorporated into food for humans (Møller *et al.*, 2021). RuBisCO and alfalfa protein show the most promising functional properties with respect to solubility and foaming properties, making it a potential substitute for animal protein ingredients. Thus far, when considering leaf protein for food the focus has been on proteins from alfalfa and sugar beet leaves. However, the RuBisCO protein is very well preserved among different plant species in terms of protein sequence and structure, which is why RuBisCO obtained from leafy plants, such as grasses and clovers, may have similar functional properties. At the same time, RuBisCO shows relatively low allergenicity, so a purified RuBisCO product may serve as a potential source for highly challenged multi-allergenic population. However, it is unclear whether proteins other than RuBisCO present in different green biomass have allergenic properties, and this needs to be resolved. Today, alfalfa protein is approved in food applications, but only based on a limited daily intake. There is still some way to go to describe the full matrix, both for alfalfa and other green biomass. Different anti-nutritional factors are present in different plant species and they need to be quantified in each specific case as they may concentrate in the protein fraction and affect nutrient bioavailability. Any new protein product produced from either alfalfa, clover or grass needs an EFSA approval before the protein can be used in food products (Møller *et al.*, 2021).

Fibre press cake

Around half of the plant crude protein will distribute to the fibre press cake (pulp), and the composition of amino acids in this fraction is similar to the composition in the whole plant (Damborg *et al.*, 2018). As a considerable proportion of the protein retained in the pulp is fibre-associated, the pulp is expected to be suitable for ruminants. Chemical analysis of the pulp revealed a fraction with a higher DM concentration than in the whole crop (plant), similar crude protein concentration and lower crude ash concentration (Table 2). *In vitro* ruminant digestibility tended to be lower for the pulp, as expected due to a large proportion of soluble organic matter removed upon juice extraction. When expressed as digestible organic matter (DOM) as a proportion of DM, the difference disappeared for white clover and perennial ryegrass because the ash content is also reduced during the extraction step.

Regarding the nutritional value of the pulp as feed for ruminants, one feeding experiment with cows has been published so far (Damborg *et al.*, 2018), and several ensiling experiments have been performed. Despite the low residual sugar in the pulp, the pulp generally ensiles very well, probably because the buffer capacity also is low due to the relatively low mineral content (Hansen *et al.*, 2020). Contrary to the *in vitro* and *in situ* digestibility analyses of the pulp by Damborg *et al.* (2018), the *in vivo* digestibility of CP and neutral detergent fibre was greater for pulp silage diets compared with grass-clover silage diets. This observation can likely be explained by the physical processing of the pulp in the screw-press

Table 2. Chemical composition of red clover, perennial ryegrass, alfalfa and white clover plant and the resulting pulp (Damborg *et al.*, 2018).[1]

Plant species	**Fraction**	**DM, %**	**Crude protein, % of DM**	**Crude ash, % of DM**	***In vitro* OM digestibility, %**	**DOM, %**
Red clover	plant	16.6	20.5	9.06	65.4	59.4
	pulp	43.5	19.8	6.63	57.9	54.0
Perennial ryegrass	plant	19.9	16.7	8.63	74.4	67.9
	pulp	41.4	16.4	5.11	69.9	66.3
Alfalfa	plant	19.6	20.5	8.86	61.9	56.4
	pulp	39.9	19.8	5.80	56.6	53.2
White clover	plant	15.8	26.7	10.4	77.4	69.4
	pulp	41.2	26.8	7.23	74.3	68.9
P-value		<0.001	0.44	<0.001	0.046	0.21

[1] All values are mean of three harvests (Nov 2013, Jun 2014, Sep 2014).

during biorefining, which disintegrates the fibres and increases the accessibility for the rumen microbes, thus increasing the degradability of the fibre and fibre-bound nutrients. This higher feed utilization was reflected in a higher energy-corrected milk yield from cows fed pulp silage compared to grass clover silage (Table 3). The results imply that extraction of protein from grassland plants can increase the digestibility of the fibre part of grassland plants. A Finnish study investigated the effects of including pulp made from silage that substituted up to 50% of the grass silage on feed intake, rumen fermentation, diet digestion and milk production in dairy cows. In this study no effect on milk yield (37 kg energy corrected mild yield) was detected (Savonen *et al.*, 2020).

The fibre pulp has numerous alternative applications than just ruminant feed, and the possibilities for adding further biorefining technologies are many. This includes, e.g. bioenergy production through anaerobic digestion to biogas or pyrolysis to synthesis gas, bio-oil and biochar, biomaterials or biochemicals. In the project Grass Biochar, it is investigated how Green Biorefining can be integrated with pyrolysis of the fibre pulp. The pyrolysis will produce renewable energy to supply the heat for protein precipitation and drying of the protein concentrate, as well as a high-quality biochar. Large-scale production of biochar from the grass fibre will open up significant potentials for creating bioenergy with carbon capture and storage (BECCS) solutions in combination with green biorefineries.

Using the fibre pulp for fibre-based biomaterials is another valuable application. This approach is in fact the main aim for all of the existing Green Biorefineries that process silage instead of fresh green biomass. Biowert in Brensbach, Germany (www.biowert.de) produces grass-based insulation material and grass fibre enforced bio-plastic, a biocomposite material suitable for injection moulding or extrusion applications. Newfoss in Uden in the Netherlands (www.newfoss.com) produces insulation materials and fibres for paper and packaging. The project SinProPack in DK has recently started the investigations and development of producing biobased packaging for the takeaway market out of the fibre pulp from green biorefineries and another project, Høsttek, has started developing sustainable fibreboards of the fibre pulp.

Common to both fibre pulp utilization for bioenergy or biomaterials is that it is an advantage if the fibre is depleted of its protein content. Thus, efficient extraction of protein at the green biorefineries poses no negative impact on these applications. However, for the application where the fibre pulp is utilized for ruminant animal feed, there is a lower limit of how little protein should be left in the pulp.

Table 3. Feeding experiment with pulp to dairy cattle compared with grass clover (Damborg *et al.*, 2018).[1]

Feed	Pulp silage	Grass clover silage
DM, %	28	52
Crude protein, %	18	16
Ash, %	9.3	9.4
NDF, %	45	39
In vitro dig. OM, %	70	72
DM intake, kg/day	23.0	22.7[2]
ECM, kg/day	37.0	33.5[2]
In vivo digestibility		
OM, %	73	70[2]
NDF, %	63	54[2]
Protein, %	66	60[2]

[1] ECM = energy corrected milk yield; OM = organic matter; DM = dry matter; NDF = neutral detergent fibre.

[2] Significant different from pulp silage.

Residual juice fraction

The residual juice remaining after the protein precipitation from the green juice is characterized by a low DM content (5-8%), a variable but high content of soluble carbohydrates and minerals, while the crude protein (10-20% of DM) only contains half the amount expressed as true protein, the other half being various non-protein nitrogen compounds (NPN) (Damborg *et al.*, 2020). The specific composition of the residual juice depends on a number of factors including both the processing steps involved in the Green Biorefining separation platform, especially the precipitation method, as well as type-, maturity- and growth conditions of the green biomass input.

The application for anaerobic digestion of the residual juice is a straightforward opportunity, especially in Denmark, which has a significant biogas industry. Many of the biogas plants in Denmark could benefit from an extra substrate with a low, but easily digested, solids concentration in order to co-digest with fibrous agricultural residues such as deep litter, cow manure and straw from cereal grain and grass seed production. This is for example the case at Ausumgaard, the first commercial green biorefinery in Denmark (https://ausumgaard.dk/baeredygtig-energi/graesprotein/), which has a large biogas facility where both the residual juice and the fibrous pulp from the biorefinery can be digested. The use of residual juice for anaerobic digestion has been evaluated in terms of technical, economic and environmental sustainability (Corona *et al.*, 2018; Djomo *et al.*, 2020; Feng *et al.*, 2021; Jensen and Gylling, 2018; Santamaria-Fernandez *et al.*, 2018). If the residual juice cannot be co-digested in an existing anaerobic digestion plant, it is a much cheaper and more efficient solution to install a packed bed reactor, as shown by Feng *et al.* (2021). Here residual juice was efficiently digested as a sole substrate at low retention time (5.5 days) and therefore a much smaller reactor size and capital investment is needed. An obvious advantage for anaerobic digestion of the residual juice from green biorefineries is that the inorganic nutrients will be led directly into an existing recirculation of nutrients, as the digestate from anaerobic digestion is spread on agricultural land as fertilizer, already in the current system.

The residual juice could potentially be used for much more than bioenergy, before nutrients are recirculated to the crop production. (1) Historically, the valuable products from residual juice/brown juice from green biomass processing has been focused around amino acids and lactic acid. Several studies and commercial activities have looked into the production of amino acid concentrates (Ecker *et al.*, 2012) or specific amino acids such as L-lysine (Andersen and Kiel, 2000; Thomsen *et al.*, 2015). (2) In the few existing green biorefineries that are processing silage grass, the juice is used for bioenergy through biogas production (Biowert) or its amino acid, organic acids and inorganic nutrient content are used primarily as fertilizer products, concentrated through membrane filtration technology. (3) When processed in the Danish base case setup (Figure 4) the residual juice will be high in carbohydrates and inorganic nutrients. This combination has high potential as a substrate for fermentation applications in the biotech industry, producing products such as building block biochemicals, single cell protein or high value secondary metabolites. In order to achieve a good fermentation substrate, it is an advantage to reduce the volume and increase the concentration of the carbohydrates as well as other macronutrients present in the residual juice. This is experimented in the current refinery platform by membrane filtration.

Future perspectives

The perspectives of simultaneously securing farmers licence to produce and creating a new biobased industry that can supply local products for the green transition are supporting the development of green biorefining in Denmark. In the recent financial bill, a significant budget was set aside for both R&D and for the support of new commercial activities. Several EU projects are supporting the development on a broader scale. There is still much to optimize in order to be able to produce the preferred raw material for a biorefinery, in contrast to the earlier single focus on feed quality for ruminants. Such issues include:

- plant breeding for optimal protein extractability and quality;
- development of efficient harvest planning and logistics to continuously deliver good quality green biomass to the biorefineries;
- process development and optimization at the biorefineries to achieve constant high yields of protein concentrates with constant high digestibility and nutritional value;
- value creation of the press cake fibre and the residual juice to achieve a better overall business case for the green biorefinery;
- technology integration for cascade utilization of side streams and residues;
- development of flexible biorefinery solutions, where input biomass and output products can change according to seasonal variation and market conditions, in order to achieve efficient use of production facilities all year round;
- valorisation of the benefits from grassland production on climate and environment.

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References

Andersen M. and Kiel P. (2000) Integrated utilisation of green bio-mass in the green biorefinery. *Industrial Crops and Products* 11, 129.

Becker T., Isselstein J., Jurschik R., Benke M. and Kayser M. (2020) Performance of modern varieties of *Festuca arundinacea* and *Phleum pratense* as an alternative to *Lolium perenne* in intensively managed sown grasslands. *Agronomy* 10, 540. https://doi.org/10.3390/agronomy10040540.

Cadoux S., Ferchaud F., Demay C., Boizard H., Machet J.M., Fourdinier E., Preudhomme M., Chabbert B., Gosse G. and Mary B. (2014) Implications of productivity and nutrient requirements on greenhouse gas balance of annual and perennial bioenergy crops. *GCB Bioenergy* 6, 425-438. https://doi.org/10.1111/gcbb.12065

Chen J., Lærke P.E. and Jørgensen U. (2022) Land conversion from annual to perennial crops: A win-win strategy for biomass yield and soil organic carbon and total nitrogen sequestration. *Agriculture, Ecosystems and Environment* 330, 107907. https://doi.org/10.1016/j.agee.2022.107907

Chiesa S. and Gnansounou E. (2011) Protein extraction from biomass in a bioethanol refinery – possible dietary applications: Use as animal feed and potential extension to human consumption. *Bioresource Technology* 102(2), 427-436.

Corona A., Ambye-Jensen M., Vega G.C., Hauschild M.Z. and Birkved M. (2018) Techno-environmental assessment of the green biorefinery concept: Combining process simulation and life cycle assessment at an early design stage. *Science of the Total Environment* 635, 100-111.

Cougnon M., De Swaef T., Lootens P., Baert J., De Frenne P., Shahidi R., Roldan-Ruiz I. and Reheul D. (2017) *In situ* quantification of forage grass root biomass, distribution and diameter classes under two N fertilisation rates. *Plant and Soil* 411, 409-422. https://doi.org/10.1007/s11104-016-3034-7

Damborg V.K., Jensen S.K., Weisbjerg M.R., Adamsen A.P. and Stødkilde L. (2020) Screw-pressed fractions from green forages as animal feed: Chemical composition and mass balances. *Animal Feed Science and Technology* 261, 114401.

Damborg V.K., Stødkilde L., Jensen S.K. and Weisbjerg M.R. (2018) Protein value and degradation characteristics of pulp fibre fractions from screw pressed grass, clover, and lucerne. *Animal Feed Science and Technology* 244, 93-103.

Djomo N.S., Knudsen M.T., Martinsen L., Andersen M.S., Ambye-Jensen M., Møller H.B. and Hermansen J.E. (2020) Green proteins: An energy-efficient solution for increased self-sufficiency in protein in Europe. I. *Biofuels, Bioproducts and Biorefining* 14 3, 605-619.

Dohleman F.G. and Long S.P. (2009) More productive than maize in the Midwest: how does Miscanthus do it? *Plant Physiology* 150, 2104-2115.

Ecker J., Schaffenberger M., Koschuh W., Mandl M., Böchzelt H.G., Schnitzer H., Harasek M. and Steinmüller H. (2012) *Green biorefinery upper Austria – Pilot Plant operation. Separation and Purification Technology* 96, 237-247. https://doi.org/10.1016/j.seppur.2012.05.027

EU Commission (2018) Report from the Commission to the Council and the European Parliament on the development of plant proteins in the European Union. COM(2018) 757 final, 16 pp.

Feng L., Ward A.J., Ambye-Jensen M. and Møller H.B. (2021) Pilot-scale anaerobic digestion of by-product liquid (brown juice) from grass protein extraction using an un-heated anaerobic filter. *Process Safety and Environmental Protection* 146, 886-892.

Franzluebbers A.J., Sawchik J. and Taboada M.A. (2014) Agronomic and environmental impacts of pasture-crop rotations in temperate North and South America. *Agriculture Ecosystems and Environment* 190, 18-26.

Gylling M., Jørgensen U., Bentsen N.S., Kristensen I.T., Dalgaard T., Felby C. and Johansen V.K. (2016) The + 10 million tonnes study: increasing the sustainable production of biomass for biorefineries. Department of Food and Resource Economics, University of Copenhagen, Frederiksberg, Denmark. Available at: https://ifro.ku.dk/publikationer/saerlige_ifro-udgivelser/10miotons/

Hansen N.K., Kristensen, T., Johansen M. and Weisbjerg M.R. (2020) Effect of regrowth period for perennial ryegrass on yield and nutritive value of grass. *Grassland Science in Europe* 25, 267-269.

Houseman R.A. and Connell J. (1976) Utilization of products of green-crop fractionation by pigs and ruminants. *Proceedings of the Nutrition Society* 35, 213-220.

Jacobsen E. (2020) Fermentation of concentrated brown juice from a green biorefining platform. Production of high value carotenoids using recombinant oleaginous yeast – An experimental and industrial, techno-economic case study. MSc thesis at Aarhus University, Department of Biological and Chemical Engineering, Aarhus, Denmark.

Jensen J.D. and Gylling M. (2018) Økonomiske vurderinger i forhold til værdikæden for Grøn Bioraffinering. Institut for Fødevare- og Ressourceøkonomi, Københavns Universitet, Copenhagen, Denmark, 11 pp.

Jørgensen U. and Lærke P.E. (2016) Perennial grasses for sustainable European protein production. In: Barth S., Murphy-Bokern D., Kalinina O., Taylor G. and Jones M. (eds.) *Perennial biomass crops for a resource-constrained world.* Springer Publishing, Belin, Germany, pp. 33-41. https://doi.org/10.1007/978-3-319-44530-4_3

Jørgensen U., Kristensen T., Jørgensen J.R., Kongsted A.G., De Notaris C., Nielsen C., Mortensen E.Ø., Ambye-Jensen M., Jensen S.K., Stødkilde-Jørgensen L., Dalsgaard T.K., Møller A.H., Grøn Sørensen C., Asp T., Lehmann Olsen F. and Gylling M. (2021) Green biorefining of grassland biomass. DCA report No. 193 Available at: https://dcapub.au.dk/djfpublikation/index.asp?action=showandid=1480

Kandel T.P., Elsgaard L., Andersen M.N. and Lærke P.E. (2016) Influence of harvest time and frequency on light interception and biomass yield of *Festulolium* and tall fescue cultivated on a peatland. *European Journal of Agronomy* 81, 150-160.

Kandel T.P., Elsgaard L., Karki S. and Lærke P.E. (2013) Biomass yield and greenhouse gas emissions from a drained fen peatland cultivated with reed canary grass under different harvest and fertilizer regimes. *BioEnergy Research* 6 (3), 883-895.

Larsen S., Bentsen N.S., Dalgaard T., Jorgensen U., Olesen J.E. and Felby C. (2017) Possibilities for near-term bioenergy production and GHG-mitigation through sustainable intensification of agriculture and forestry in Denmark. *Environmental Research Letters* 12. https://doi.org/10.1088/1748-9326/aa9001

Manevski K., Laerke P.E., Jiao X., Santhome S. and Jørgensen U. (2017) Biomass productivity and radiation utilisation of innovative cropping systems for biorefinery. *Agricultural and Forest Meteorology* 233, 250-264.

Manevski K., Laerke P.E., Olesen J.E. and Jorgensen U. (2018) Nitrogen balances of innovative cropping systems for feedstock production to future biorefineries. *Science of the Total Environment* 633, 372-390. https://doi.org/10.1016/j.scitotenv.2018.03.155

McEniry J. and O'Kiely P. (2014) Developments in grass-/forage-based biorefineries, in: *Advances in Biorefineries: Biomass and waste supply chain exploitation*. Woodhead Publishing Limited, pp. 335-363. https://doi.org/10.1533/9780857097385.1.335.

Møller A.H., Hammershøj M., Passos N.H.M., Tanambell H., Stødkilde L., Ambye-Jensen M., Danielsen M., Jensen S.K. and Dalsgaard T.K. (2021) Biorefinery of green biomass. How to extract and evaluate high quality leaf protein for food? *Journal of Agricultural and Food Science* 69, 14341-14357.

Mortensen E.Ø. and Jørgensen U. (2022) Danish agricultural biomass production and utilization in 2030. Memorandum from DCA – Danish Centre for Food and Agriculture, 24 pp. Available at: https://pure.au.dk/portal/files/257489198/Danish_agricultural_biomass_production_2030_DCA_memorandum_2022.pdf.

Näsi M. and Kiiskinen T. (1985) Leaf protein from green pulse crops and nutritive-value of legume protein-concentrates for poultry. *Journal of Agricultural Science in Finland* 57, 117-123.

Nielsen C.K., Elsgaard, L., Jørgensen U. and Lærke P.E. (2021b) Mitigation potential of paludiculture for five different Danish peatland sites under controlled water tables – a mesocosm study. Renewable resources from wet and rewetted peatlands. In: *Proceedings of the RRR2021 conference*. Greifswald, Germany, p. 43.

Nielsen C.K., Stødkilde L., Jørgensen U. and Lærke P.E. (2021a) Effects of harvest and fertilization frequency on protein yield and extractability from flood-tolerant perennial grasses cultivated on a fen peatland. *Frontiers in Environmental Science* 9:619258. https://doi.org/10.3389/fenvs.2021.619258.

Nielsen L. (2012) Baggrundsnotat: Udbytteniveauer ved forskellig drift af lavbundsjord, 7 p. Available at: http://ifro.ku.dk/fotos/media_library/foi/docs/publikationer/rapporter/eksterne_rapporter/ti_mio_tons_plan/bpl4.

Pedersen E.J.N., Witt N., Mortensen J. and Sørensen C. (1979) Fractionation of green crops and preservation of pressed crops and juice. I: Ensiling of pressed crops. *Tidsskrift for Planteavl* 1509.

Pehme S., Veromann E. and Hamelin L. (2017) Environmental performance of manure co-digestion with natural and cultivated grass – A consequential life cycle assessment. *Journal of Cleaner Production* 162, 1135-1143. https://doi.org/10.1016/j.jclepro.2017.06.067.

Pirie N. (1942) Green leaves as a source of proteins and other nutrients. *Nature* 149, 251. https://doi.org/10.1038/149251a0

Pirie N.W. (1978) *Leaf protein and its by-products in human and animal nutrition* (2nd ed.). Cambridge University Press, Cambridge, UK.

Pisulewska E., Hanczakowski P., Pisulewski P. and Szymczyk B. (1991) The changes of the yield, composition and nutritive value of leaf protein extracted from vetch and cereal mixtures during three years cultivation. *Journal of the Science of Food and Agriculture* 55, 197-205.

Pugesgaard S., Schelde K., Larsen S.U., Lærke P.E. and Jørgensen U. (2015) Comparing annual and perennial crops for bioenergy production influence on nitrate leaching and energy balance. *Global Change Biology Bioenergy* 7, 1136-1149.

Santamaria-Fernández M., Molinuevo-Salces B., Lübeck M. and Uellendahl H. (2018) Biogas potential of green biomass after protein extraction in an organic biorefinery concept for feed, fuel and fertilizer production. *Renewable Energy* 129, 769-775.

Savonen O., Franco M., Stefanski T., Mäntysaari P., Kuoppala K. and Rinne M. (2020) Grass silage pulp as a dietary component for high-yielding dairy cows. *Animal* 14(7), 1472-1480. https://doi.org/10.1017/S1751731119002970

Solati Z., Jørgensen U., Eriksen J. and Søegaard K. (2017) DM yield, chemical composition and estimated extractable protein of legume and grass species during the spring growth. *Journal of the Science of Food and Agriculture* 97(12), 3958-3966.

Stødkilde L., Ambye-Jensen M. and Jensen S.K. (2020) Biorefined grass-clover protein composition and effect on organic broiler performance and meat fatty acid profile. *Journal of Animal Physiology and Animal Nutrition* 104, 1757-1767.

Stødkilde L., Ambye-Jensen M. and Jensen S.K. (2021) Biorefined organic grass-clover protein concentrate for growing pigs: effect on growth performance and meat fatty acid profile. *Animal Feed Science and Technology* 276, 114943. https://doi.org/10.1016/j.anifeedsci.2021.114943

Stødkilde L., Damborg V., Jørgensen H., Lærke H. and Jensen S. (2019) Digestibility of fractionated green biomass as protein source for monogastric animals. *Animal* 13(9), 1817-1825. https://doi.org/10.1017/S1751731119000156

Thers H., Stødkilde L., Jensen S.K. and Eriksen J. (2021) Linking protein quality in biorefinery output to forage crop crude protein input via the Cornell Net Carbohydrate and Protein System. *Applied Biochemistry and Biotechnology* 193, 2471-2482.

Thomsen M.H. and Kiel P. (2008) Selection of lactic acid bacteria for acidification of brown juice (grass juice), with the aim of making a durable substrate for L-lysine fermentation. *Journal of the Science of Food and Agriculture* 88 (6), 976-983.

Van Grinsven, H.J.M., Erisman, J.W., de Vries, W. and Westhoek, H. (2015) Potential of extensification of European agriculture for a more sustainable food system, focusing on nitrogen. *Environmental Research Letters* 10, 2.

Vils E., Jensen S.K., Bache J.K. and Vinther J. (2020) Grønprotein, Hestebønne og Rapskager til Slagtesvin. Meddelelse nr. 1214. In: *SEGES Svineproduktion, Den rullende Afprøvning*.

The influence of nitrogen fertilization and legume species on the forage quality of multicomponent sown meadows

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Abstract

Field trials were carried out with the aim to investigate forage yield quality of sown legume-grass swards under three nitrogen fertilization rates: N0, N60 and N120. Mixtures were composed of 50% legumes and 50% grasses (G). The use of multicomponent grass-legume mixtures containing red clover *Trifolium pratense* (Tp) and lucerne *Medicago sativa* (Ms) is a traditional practice in Latvia, but fodder galega *Galega orientalis* (Go) is grown for a relatively short period. Dry matter (DM) yield was analysed for the following quality indices and mineral contents: crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), phosphorus (P), potassium (K) and calcium (Ca). The legume species used in swards determined the quality and the mineral content of DM yield. The mixtures containing lucerne (Ms+G) and galega (Go+G) had a higher CP content, the red clover mixture (Tp+G) had a lower ADF content, and lucerne-containing mixture (Ms+G) achieved a higher Ca content. A significant N fertilization effect on the phosphorus and potassium content was established.

Keywords: herbage quality, mineral content, grass-legume mixture, nitrogen fertilization

Introduction

Legumes allow improved sustainability and stability of agroecosystems, and provide the cheapest source of nitrogen (Wilkins and Vidrih, 2000). Although nitrogen (N) fertilization contributes to increased dry matter (DM) yield, it also has a negative effect on the content of legumes in the sward (Soegaard and Nielsen, 2012). Crude protein content is closely connected with the proportion of legumes in the sward. Using different grass-legume mixtures, the protein content could be more affected by the type of seed mixture than by the N level applied (De Vliegher and Carlier, 2008; Meripold *et al.*, 2016). Some researchers have found no significant effect of species or mixtures on the nutritive value of herbage, while increasing the rates of N fertilization can cause a significant increase in crude protein (CP) (Moloney *et al.*, 2016). The objective of this research was to determine the influence of N fertilization and legume species on the crude protein, fibre and mineral element content in herbage of multicomponent grass-legume mixtures.

Materials and methods

Field trials were conducted at three experimental sites in Latvia. At each site (with same experimental design and condition) the same mixtures were sown in June 2014: without a cover crop, in three replications. Plot size was 10 m^2. The following grass combinations were used in mixtures: *Festuca arundinacea, ×Festulolium loliaceum,* and *Lolium×boucheanum* in equal parts (G). Mixtures were composed of *Trifolium pratense* 50% and grasses 50% (Tp+G); *Medicago sativa* 50% and grasses 50% (Ms+G); and *Galega orientalis* 50% and grasses 50% (Go+G). The following fertilization treatments were used for all mixture types (MT): P78, and K90, and three N fertilization levels: N0, $N60_{(30+30)}$, and $N120_{(60+60)}$ kg ha^{-1}. Swards were cut three times during the vegetation season. The CP content of DM yield was determined by modified Kjeldahl method; neutral detergent fibre (NDF) and acid detergent fibre (ADF) were determined by the van Soest method, and mineral elements P, K and Ca were analysed by atomic absorption spectrometry. Data were analysed with two-way analysis of variance; differences between means were detected by LSD at $P<0.05$ (MS Excel for Windows, 2003).

Results and discussion

Significant differences in the content of CP, Ca and ADF in the harvested DM were found between mixture types (Table 1). Galega is reported to be a highly valuable legume, providing fodder rich in protein and bio-active substances (Baležentiene, 2008). The CP content determined in our trials (121.0 g kg^{-1}, average for three fertilization rates) for galega-containing mixture was not very high, due to the large grass proportion in swards (especially in the first year of sward use). The mixtures containing lucerne (Ms+G) and galega (Go+G) had significantly higher CP content in comparison with the red clover mixture (Tp+G).

Legumes accumulated more Ca than grasses, and Ca content in mixture DM is closely connected with legume content (Adamovics and Gutmane, 2018). A significantly higher Ca content in DM yield was found for the mixture containing lucerne. There were no significant NDF content differences between the mixtures and N fertilization rates. No significant N fertilization effect was found on the CP, Ca and ADF content in the DM, but a significant N fertilization effect on the P and K content was established. Increasing N fertilizer application rate from 0 to 120 kg ha^{-1} contributed to a significant increase in P content for all mixtures.

The proportion of legumes in swards had a significant positive correlation with the content of CP and Ca in the DM yield (Figure 1). A significant ($P<0.01$) negative correlation was established between the proportion of legumes in swards and NDF ($r=-0.46$). The CP content in DM yield had a significant ($P<0.01$) positive correlation with the content of P and Ca in DM yield (Figure 2). The NDF content had a significant ($P<0.01$) negative correlation with the content of K ($r=-0.47$), Ca ($r=-0.58$) and CP ($r=-0.39$) in DM yield. A significant ($P<0.01$) positive correlation ($r=0.42$) was established between the fibre fractions NDF and ADF.

Table 1. Quality of grass-legume swards on average for three production years.[1]

Mixture type (MT)	N rate, kg ha^{-1} (N)	Content in DM, g kg^{-1}					
		CP	NDF	ADF	Ca	P	K
Tp+G	N0	94.8	512.2	323.9	7.7	2.52	23.45
	N60	103.3	538.5	343.0	6.7	2.73	25.65
	N120	106.3	544.6	326.1	8.0	2.73	22.25
Mean Tp+G		101.5±5.0	531.8±10.8	331±6.0	7.5±0.50	2.66±0.07	23.78±0.60
Ms+G	N0	131.0	517.0	359.0	11.5	2.72	23.60
	N60	115.9	561.4	364.4	8.4	2.77	24.75
	N120	132.7	543.6	375.5	11.7	2.88	23.55
Mean Ms+G		126.5±5.5	540.6±9.8	366.3±5.4	10.5±0.70	2.79±0.05	23.97±0.37
Go+G	N0	121.3	536.4	360.6	7.2	2.55	23.10
	N60	107.1	567.2	363.6	6.1	2.67	23.98
	N120	134.4	570.9	360.7	7.5	2.93	23.30
Mean Go+G		121±7.1	558.1±7.3	361.6±5.0	6.9±0.44	2.71±0.08	23.46±0.28
$LSD_{0.05}$ N		n.s.	n.s.	n.s.	n.s.	0.20	1.02
$LSD_{0.05}$ MT		17.14	n.s.	16.07	1.57	n.s.	n.s.
$LSD_{0.05}$ N/MT		n.s.	n.s.	n.s.	n.s.	n.s.	n.s.

[1] n.s. = not significant.

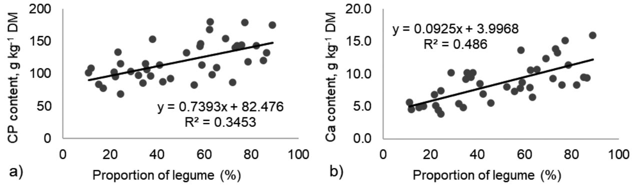

Figure 1. Relationship ($P<0.01$) between (A) CP content in herbage DM and the proportion of legumes in the sward, and (B) Ca content in herbage DM and the proportion of legumes in the sward.

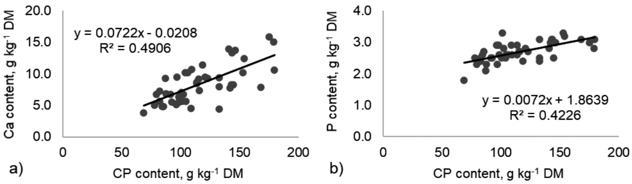

Figure 2. Relationship ($P<0.01$) between (A) Ca content, (B) P content and CP content in the herbage DM.

Conclusions

Significant differences in the content of CP, ADF and Ca in DM yields were found between mixture types. No significant N fertilization effect on the CP, NDF, ADF and Ca contents were found.

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References

Adamovics A. and Gutmane I. (2018) The influence of nitrogen fertilizer and legume content on the quality of multi-species swards. *Grassland Science in Europe* 23, 111-113.

Balezentiene L. (2008) Evaluation of galega suitability for cattle feeding. *Grassland Science in Europe* 13, 777-779.

De Vliegher A. and Carlier L. (2008) Potential of fodder legumes under intensive farming conditions in Flanders. *Grassland Science in Europe* 13, 236-238.

Meripold H., Tamm U., Tamm S., Vosa T. and Edesi L. (2016) Yields and feed value of different fodder galega-grass mixtures. *Grassland Science in Europe* 21, 464-466.

Moloney T., Sheridan H., O'Riordan E. G., O'Kiely P. (2016) Nitrogen fertilization effects on multi-species and *Lolium perenne* yields and composition under silage management. *Grassland Science in Europe* 21, 188-190.

Soegaard K. and Nielsen K.A. (2012) White and red clover in highly productive short-lasting grassland mixtures. *Grassland Science in Europe* 17, 172-174.

Wilkins R.J. and Vidrih T. (2000) Grassland for and beyond 2000. *Grassland Science in Europe* 5, 9-17.

Response of tall fescue and orchardgrass to deficit irrigation

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Abstract

Two species of cool season grasses – tall fescue, cv. 'Fawn' and orchardgrass cv. 'Tekapo') – were tested under arid zone condition. Both were sown in the experimental fields of Qassim University, Saudi Arabia, and subjected to four levels of water supply: 25, 60, 75 and 100% of the calculated crop evapotranspiration, respectively. Growth parameters, such as plant height, fresh matter yield and dry matter yield were measured, and the water use efficiency was calculated. Subsequent analyses revealed that water stress had an adverse impact on plant growth, whereby water use efficiency was maximized when water supply corresponded to 75% of evapotranspiration. Moreover, tall fescue was better able to withstand the water stress, as its water use efficiency remained unchanged irrespective of the water supply.

Keywords: tall fescue, orchardgrass, water deficit, irrigation

Introduction

Most of the fresh water around the globe is used for food production and is projected to increase even further to maximize the crop yield (Davis *et al.*, 2017). Forage crops are particularly significant water consumers, as they may remain in the field for several years and require ample irrigation, which may be problematic in arid regions. Thus, extensive research has been conducted to identify cultivars that can thrive under arid conditions. Empirical evidence indicates that tall fescue cv. 'Fawn' is one of the cool season forage cultivars that can grow in soils with up to 8,000 ppm of NaCl (Al-Ghumaiz, 2013; Al-Ghumaiz and Motawei, 2011; Al-Ghumaiz *et al.*, 2017; Motawei and Al-Ghumaiz, 2012). However, its response to deficit irrigation under arid conditions is not sufficiently explored. In this work, tall fescue and orchardgrass cv. 'Tekapo' are compared in terms of their ability to withstand different levels of water deficit.

Material and methods

Tall fescue (*Festuca arundinacea*, cv. 'Fawn') and orchardgrass (*Dactylis glomerata*, cv. 'Tekapo') were sown in the experimental fields of College of Agricultural and Veterinary Medicine, Qassim University, Saudi Arabia (26°17'51.5'N; 43°45'54.9'E). For each variety, seeding rate was 20 kg ha^{-1}, and plots were subdivided into 1 m^2 experimental subunits (1 m apart) to apply four irrigation levels. In accordance with the randomized complete block experimental design, each combination was replicated three times. After calculating crop evapotranspiration using the FAO Penman Monteith method (Allen, 1998), four levels of water supply were determined as follows: control (C) with 100% of the calculated crop evapotranspiration, and three treatments, with 25% (SD), 60% (AD), and 75% (MD) of the calculated crop evapotranspiration. To meet the study aims, plant height (H), fresh weight (FMY), and dry weight (DMY) were measured and the water use efficiency (WUE) was calculated (Geerts and Raes, 2009). Significance of the studied factors was tested using the ANOVA and their means were compared using the least significant differences (LSD) method.

Results and discussion

The analyses revealed that water deficit levels exerted adverse influence on the measured growth parameters for both species. However, the ANOVA results show this effect was significant only for plant height (Table 1) as control samples from both cultivars were significantly taller than those that received suboptimal irrigation. When different treatments were compared using the LSD test, no statistically significant differences in H emerged between C and MD, as well as AD and SD (Table 1). Similar trends were noted for both FMY and DMY, whereby none of the treatments yielded significant differences. However, closer examination of DMY values revealed that, when water supply was not drastically reduced (MD and AD treatments) even though the plant water content decreased, growth remained relatively unaffected, which was not the case in the SD treatment. These observations are in line with the results reported by Asay *et al.* (2001). Water use efficiency (WUE) also revealed different trends, whereby it was the highest in controls, followed by the MD, SD, and finally AD treatment (but none of the differences were statistically significant).

The ANOVA results further indicated that the interaction between the plant species and the deficit level was significant, suggesting a variation in the response of the two cultivars to the imposed water deficits. Moreover, although 'Tekapo' plants in the C and MD group had a higher WUE, the values declined sharply as the water supply was reduced further. Conversely, WUE calculated for 'Fawn' was relatively constant irrespective of the water supply, as shown in Table 2.

Table 1. Statistical analysis of the effects of the different water deficit treatments.[1]

Treatment	Plant height	FMY (kg ha^{-1})	DMY (kg ha^{-1})	WUE (kg m^{-3})
Plant species (Sp)				
Fawn	15.42a	1,467.5^{a}	512.9^{a}	0.092^{a}
Tekapo	11.85^{a}	1,324.0^{a}	468.3^{a}	0.087^{a}
Significance	n.s.	n.s.	n.s.	n.s.
Water deficit level (WD)				
C	17.50^{a}	2,558.3^{a}	875.2^{a}	0.107^{ab}
MD	15.33^{ab}	1,736.7^{b}	541.8^{b}	0.114^{a}
AD	12.67^{b}	823.2^{c}	362.2^{bc}	0.0629^{b}
SD	11.83^{b}	465^{c}	183.3^{c}	0.0761^{b}
Significance	**	***	***	*
Sp × WD	**	*	***	**

[1] n.s. = not significant; * P<0.05; ** P< 0.01 *** P<0.001.

Table 2. The effect of interaction of the two factors.

		C	MD	AD	SD
WUE (kg m^{-3})	Tekapo	0.127^{a}	0.135^{a}	0.032^{d}	0.056^{c}
	Fawn	0.087^{b}	0.092b	0.093^{b}	0.097^{b}
H (cm)	Tekapo	17.3^{b}	16.0^{b}	11.0c	9.7^{d}
	Fawn	20.0^{a}	15.7^{b}	13.3^{c}	12.7^{c}
DMY (kg ha^{-1})	Tekapo	971.3^{a}	742.0^{b}	186.0^{e}	149.3^{e}
	Fawn	776.0^{b}	569.4^{c}	323.4^{d}	217.3^{d}
FMY (kg ha^{-1})	Tekapo	3,040.0^{a}	2,066.7^{b}	423.3^{d}	340.0^{d}
	Fawn	2,076.7^{b}	1,406.7^{c}	619.3^{d}	590.0^{d}

Conclusions

Two species of cool season grasses were tested under arid conditions in the experimental fields of Qassim University, Qassim, KSA, by subjecting the plants to different levels of water stress (25, 60, and 75% of the calculated crop evapotranspiration) and comparing their key growth parameters and water use efficiency with controls (irrigated at 100% of the calculated crop evapotranspiration level). The results showed that, while both cultivars were adversely affected by insufficient irrigation, *Festuca arundinacea*, cv. 'Fawn' was better able to withstand arid conditions than *Dactylis glomerata* cv. 'Tekapo'.

References

Al-Ghumaiz N.S. (2013) Response of five introduced forage grass cultivars to salinity stress under irrigation. *Grassland Science in Europe* 18, 282-284.

Al-Ghumaiz N.S., Abd-Elmoniem E.M. and Motawei M.I. (2017) Salt tolerance and K/Na ratio of some introduced forage grass species under salinity stress in irrigated areas. *Communications in Soil Science and Plant Analysis* 48(12), 1494-1502.

Al-Ghumaiz N.S. and Motawei M.I. (2011) Productivity, forage quality and presence of dehydrin genes in some introduced pasture grass cultivars growing under heat stress in central region of Saudi Arabia. *Crop Science* 5(8), 1001-1006.

Allen R.G., Pereira L.S., Raes D. and Smith M. (1998) Crop evapotranspiration guidelines for computing crop water requirements – FAO irrigation and drainage paper 56. *FAO, Rome*, *300*(9), D05109.

Asay K.H., Jensen K.B. and Waldron B.L. (2001) Responses of tall fescue cultivars to an irrigation gradient. *Crop Science* 41(2), 350-357.

Davis K.F., Rulli M.C., Garrassino F., Chiarelli D., Seveso A. and D'Odorico P. (2017) Water limits to closing yield gaps. *Advances in Water Resources* 99, 67-75.

Geerts S. and Raes D. (2009) Deficit irrigation as an on-farm strategy to maximize crop water productivity in dry areas. *Agricultural Water Management* 96(9), 1275-1284. https://doi.org/10.2135/cropsci1996.0011183X003600020026x

Motawei M.I. and Al-Ghumaiz N. (2012) Genetic diversity in some introduced pasture grass cultivars revealed by inter-simple sequence repeats (ISSR) markers. *African Journal of Biotechnology* 11(15), 3531-3536.

Long-term changes in carbon content and chemical properties of soil in grassland plots fertilized with cattle slurry and mineral fertilizer

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Abstract

The objective of this long-term study (16 years) was to determine the effects of repeated inputs of different types of fertilizer on a grassland in the Atlantic coastal region of Galicia (NW Spain). The study included the following fertilization treatments: mineral fertilizer (calcium ammonium nitrate), cattle slurry applied in bands (on the soil surface), cattle slurry injected at a depth of 3-5 cm, and a control treatment without nitrogen (N). Soil parameters were monitored during several consecutive years in order to determine the changes in the total carbon (C), organic matter (OM) and total N contents, as well as in other fertility-related parameters. The inputs of cattle slurry applied by both techniques (banding and injection) led to a gradual increase in the OM, C and N contents in the surface layer of the soil throughout the study period. The increases were greater than those yielded by mineral fertilizer. Eleven years after the start of the study, the OM and C contents had increased significantly, by 17 and 21%, relative to those yielded by the mineral fertilizer; after 16 years the increases were smaller, at 8 and 7%, respectively. The slurry inputs also increased the soil pH and effective exchange capacity significantly and decreased the acid saturation in the exchange complex.

Keywords: cow slurry, calcium ammonium nitrate, slurry application method

Introduction

The application of livestock slurry to grassland is a common agricultural practice in the study area (Galicia, NW Spain). Slurry is applied to farmland as a source of plant nutrients and also as a means of recycling organic waste produced on farms. The application of organic manure to land alters the chemical properties of soil, with different effects on plant nutrient supply and nutrient loss. Grassland soils are capable of sequestering atmospheric CO_2, but C sequestration is regulated by complex biogeochemical processes, which are, in turn, affected by management practices (e.g. nutrient fertilization) and environmental factors. The main objective of this study was to determine whether long-term (16 years) application of dairy cattle slurry to grassland in which grass forage is harvested in consecutive years affects the chemical properties and C-fixing capacity of the soil.

Materials and methods

The study was carried out between 2005 and 2020 at the CIAM Research Centre (NW Spain), on a silt loam soil classified as Humic Cambisol. The climate of the study area is classified as humid-temperate. In October 2004, a ryegrass-clover sward was established on the site. The trial was conducted using a completely randomized block design with three replicates of each of four treatments: no nitrogen (N) fertilizer (control, C), mineral fertilizer (calcium ammonium nitrate, CAN) and slurry application, by either surface banding (BS) or injection (3-5 cm depth, 15 cm between lines, IS). Between 2005 and 2020, the fertilizers were applied in spring and/or autumn 32 times, with a mean dose for the slurry of 53 m^3 ha^{-1} $year^{-1}$. Each dose of mineral fertilizer was adjusted to supply the same amount of total N as the slurry. Grass forage was cut in consecutive years to simulate grassland harvesting. Soil samples were collected in the first year of the study (09/2005) and in various different years thereafter (03/2008, 12/2010, 04/2014, 10/2018). The soil samples were analysed to determine the OM, total C and total N contents, pH, available elements, P (Olsen), K (extracted with 1N ammonium acetate, pH 7), exchangeable cations (Ca, Mg, Na, K) and exchangeable acidity (EA). Samples obtained in October 2020 from the upper layer (0-10 cm) and deeper layers (0-30, 30-60 and 60-90 cm) were analysed to determine the apparent density in order to estimate the C stocks. Analysis of variance was used to compare the effects of the different treatments and was applied using SPSS software, version 21. Any differences between means were determined using a post-hoc Duncan's test at a significance level of $P<0.05$.

Results and discussion

The inputs of cattle slurry by both techniques (surface banding and injection) gradually increased the OM, C and N contents in the upper soil layer (0-10 cm) throughout the study period, to a greater extent than yielded by the mineral fertilizer (Figure 1). Thus, 11 years after the start of the study, the OM and C contents were respectively 17 and 21% higher than those yielded by the mineral fertilizer; after 14 years, the increases were 20 and 28%, and at the end of the study (after 16 years), the differences were smaller, at 8 and 7%, respectively. The N contents were also higher than those yielded by the mineral fertilizer, with increases of 27, 24 and 9% after 11, 14 and 16 years, respectively. No differences in the parameters were observed in relation to the method of application of the slurry.

Addition of slurry led to an increase in the soil pH (Table 1) and a decrease in the acid saturation in the exchange complex. The Ca and Mg contents in the exchange complex increased after the application of slurry over 16 years. The Ca/Mg and K/Mg ratios remained optimal after the application of slurry, whereas they appeared imbalanced in the control and mineral fertilizer treatments. Regarding the accumulation of Na in the slurry-treated plots, the low percentage of 2% in the exchange complex would not be expected to cause any problems related to physical or chemical properties.

The estimated C stocks after 16 years are shown in Table 2. There were no significant differences in C stocks between the different soil layers (0-30, 30-60 and 60-90 cm) in relation to the different fertilizer treatments. However, for the C stocks in the 0-90 cm layer, the slurry yielded some increases (not

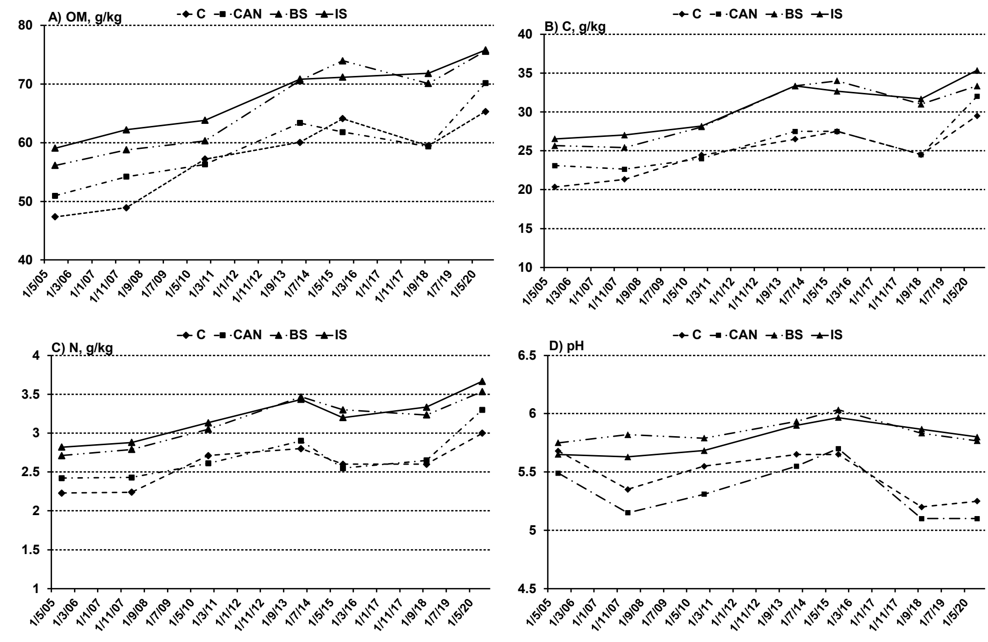

Figure 1. Changes in organic matter (A), carbon (B), nitrogen (C) and pH (D) in the upper soil layer (0-10 cm).

Table 1. Chemical properties at 0-10 cm soil depth at the start and end of the experiment.[1]

	Treatment	Cation exchange								
		AE (cmol/kg)	Ca^{2+}	Mg^{2+}	Na^{+}	K^{+}	CEC	Olsen P (mg/kg)	K	AS (g/100g)
26/09/2005	C	1.00	3.74	0.68	0.10c	0.69	6.19	34	271	16.1
	CAN	0.95	3.73	0.73	0.,07bc	0.77	6.29	35	299	15.4
	BS	0.53	5.37	1.00	0.15ab	0.79	7.85	33	309	7.3
	IS	0.70	5.09	0.94	0.16ab	0.82	7.70	33	320	9.3
	Mean	0.80	4.48	0.84	0.10	0.77	7.01	34	300	12.0
	P-value	ns	ns	ns	*	ns	ns	ns	ns	ns
14/10/2020	CAN	1.15a	3.77b	0.61c	0.11b	0.84a	6.51b	48a	328	18.16a
	M	1.40a	4.00b	0.76b	0.12b	0.50b	6.76b	34b	197	20.51a
	BS	0.13b	7.66a	1.54a	0.20a	0.69ab	10.20a	29b	269	1.26b
	IS	0.13b	7.41a	1.47a	0.19a	0.74ab	9.94a	31b	292	1.32b
	Mean	0.70	5.71	1.09	0.16	0.69	8.35	35	271	10.3
	P-value	***	**	***	**	ns	**	**	ns	***

[1] AE = acid exchange; CEC = effective cation exchange capacity; AS = acid saturation. Different letters indicate significant differences between treatments. ns = not significant; ** $P<0.01$; *** $P<0.001$.

Table 2: Carbon stocks (Mg C ha^{-1}) at different soil depths.[1]

	C stocks					
	0-10 cm	10-30 cm	0-30 cm	30-60 cm	60-90 cm	0-90 cm
C	40.8	36.9c	77.7b	10.6	2.1	90.4
CAN	38.1	47.0a	85.1a	15.4	3.8	104.3
BS	43.0	40.8bc	83.8a	24.3	11.8	119.8
IS	41.2	43.2ab	84.5a	22.9	11.0	118.4
Mean	40.8	42.0	82.8	18.3	7.2	108.2
P-value	ns	*	*	ns	ns	ns

[1] ns = not significant; ** $P<0.01$; *** $P<0.001$.

statistically significant) relative to CAN fertilizer and control, particularly due to the accumulation of C in the 30-60 and 60-90 cm layers. Therefore, in addition to the upper 0-10 cm layer, deeper layers (to 90 cm) should also be considered in calculating C stocks (Klumpp and Fornara, 2018).

Conclusions

Relative to fertilization with CAN, application of cattle slurry to grassland subjected to consecutive forage harvesting improved some chemical properties of the soil, such as the OM content, N content and exchangeable cations, reduced the need for soil amendments and favoured C fixation in the soil. No differences were observed in relation to the method of application of the slurry. In conclusion, fertilization of grassland with cattle slurry can yield ecological and environmental benefits.

Acknowledgements

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References

Klumpp K. and Fornara D.A. (2018) The carbon sequestration of grassland soils – climate change and mitigation strategies. *Grassland Science in Europe* 23, 509-519.

Current research view about nitrous oxide uptake in agricultural soils

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Abstract

Agricultural nitrous oxide (N_2O) emissions account for 60% of the total N_2O emissions. For sustainable agriculture, it is essential to reduce the emissions of N_2O from agricultural soils. Since decades, agricultural soils are a substantial N_2O source; however, there are emerging studies that also show there is uptake of N_2O by agricultural soils. These studies, showing the net uptake of atmospheric N_2O by agricultural soils, bring a series of research questions and scientific curiosity to the frontline. This is because they report fluxes of N_2O, which are in contrast with the traditional view that agricultural soils are always N_2O sources. In this abstract, we discuss the current knowledge of soil N_2O uptake in general and its current consensus with regard to agricultural ecosystems, the methodology to assess soil N_2O uptake, and future research needed to better understand the N_2O flux dynamics in agricultural ecosystems.

Keywords: sustainable agriculture, nitrous oxide uptake, nitrogen cycle, denitrifiers

Introduction

Use of synthetic nitrogen (N) fertilizers in agriculture is expected to increase by 2 to 4 fold by 2050 to meet the global food demand for the increasing human population (Tilman *et al.*, 2001). Besides increases in food and forage production, addition of fertilizer N to agricultural soils also boosts nitrification and denitrification, two major soil N transforming processes contributing to the emissions of nitrous oxide (N_2O) – a greenhouse gas ~300 times stronger than carbon dioxide (Firestone and Davidson, 1989). Therefore, for sustainable agriculture, reduction of agricultural N_2O emissions is paramount. To reduce the agricultural N_2O footprint, measures such as improving nitrogen use efficiency in animal and crop production while maintaining their yield, reducing the N fertilizer use and promoting a suitable dietary choice have been suggested (Reay *et al.*, 2012). However, to meet the goal of reducing the agricultural N_2O footprint seems still a challenging job for science and for policymakers.

N_2O uptake in agricultural ecosystems

In the biosphere, the only biological mechanism known to contribute to the soil N_2O sink is N_2O reduction to dinitrogen (N_2) via the nitrous oxide reductase enzyme, being encoded by the *nosZ* gene (Hallin *et al.*, 2018). The traditional understanding is that N_2O reduction is carried out exclusively by denitrifiers possessing the *nosZ* gene (Firestone and Davidson, 1989), organisms that are defined as *nosZ* clade I. This view was recently challenged with the discovery of N_2O reduction by non-denitrifiers, so called atypical *nosZ* or *nosZ* clade II organisms (Jones *et al.*, 2013; Sanford *et al.*, 2012). Compared to organisms of *nosZ* clade I those of *nosZ* clade II show higher functional diversity, and besides catalyzing denitrification and non-denitrifier respiration, they also represent electron sinks, and mediate detoxification processes in soils (Shan *et al.*, 2021). The multifunctional ecological services and the substantial abundance of organisms with *nosZ* clade II genes in the terrestrial environment have led to the new paradigm that highlights the soil's potential to act as N_2O sink (Jones *et al.*, 2013). This has called scientific researchers globally to work on constraining the understanding of the N_2O sink capacity of agricultural soils, for decades considered as N_2O emission hotspots. A negative N_2O flux, i.e. soil N_2O uptake, in any type of agricultural soils sounds counterintuitive. One's assumption would rather be that agricultural soils should be net N_2O emitters because of the amendment of N fertilizer – a primary substrate for microbial N_2O production. However, while constraining agricultural N_2O emissions is a challenging task, there have been an increasing number of studies showing uptake of atmospheric N_2O by agricultural soils (Figure 1). These studies therefore contrast the traditional view (i.e. that all agricultural soils are N_2O sources) and indicate that N_2O flux dynamics of agricultural soils require further investigation, especially caused by the new paradigm regarding the functional diversity of organisms of the *nosZ* clade II (Shan *et al.*, 2021). Soils represent a highly complex matrix, supporting tight interactions between N_2O metabolizing microbes (clade I and II) and nitrifiers, and soil properties such as soil pH, mineral N availability, soil moisture and oxygen status, soil temperature, and, most importantly, the quality and quantity of soil organic carbon – a primary substrate for N_2O respiring microbes (Hallin *et al.*, 2018). All the above-mentioned soil variables regulate the activities of N_2O respiring microbes, thus defining the direction of N_2O flux between soils and the atmosphere (Hallin *et al.*, 2018).

From dry oxic soils (Flechard *et al.*, 2005; Wu *et al.*, 2013) to soils with high substrate availability (e.g. mineral N) and enzymatic activities related to denitrification (Wen *et al.*, 2016), soil uptake of N_2O has been reported, indicating that not only wet soil conditions promote these organisms (Hallin *et al.*, 2018). Previously wet conditions with low oxygen and high soil organic carbon availability were thought to support the soil N_2O uptake. Moreover, recently a novel finding has shown that soyabean (*Glycine max* L. Merr.) – a globally grown leguminous food crop, can substantially reduce atmospheric N_2O to N_2 via *nosZ* gene (Itakura *et al.*, 2013), thus opening a new avenue to research for increasing agricultural N_2O emission mitigation with other leguminous types of crop species, such as alfalfa and clovers, which are an important leguminous forage crops globally. Therefore, to better constrain our agricultural N_2O footprint, a thorough scientific investigation is clearly required to answer why (e.g. ecological benefits or niche differentiation between N_2O producers vs N_2O reducers, see Hallin *et al.* (2018)), how (e.g. mechanistic pathways of clade I vs clade II microbes in tandem with soil variables), where (e.g. mineral vs organic soils, croplands vs grasslands, bulk vs rhizosphere soils, legume vs non-legume crops, and roots vs root nodules) and when (e.g. vegetative vs reproductive vs senescence phase, wet vs dry soils, and high vs low mineral N) in agricultural ecosystems soil N_2O uptake occurs in tandem with soil N_2O emissions.

Methodologies to understand agricultural soil N_2O uptake and future prospects

From laboratory to *in situ* experiments using ^{15}N labelled N_2O, so called $^{15}N_2O$ pool dilution approaches (Wen *et al.*, 2016; Yang *et al.*, 2011), from quantitative polymerase chain reactions (qPCR) to omics techniques (e.g. metagenomics) (Jones *et al.*, 2013; Sanford *et al.*, 2012), and from static chamber to eddy covariance techniques (Shurpali *et al.*, 2016) have been applied to investigate N_2O flux dynamics, associated microbial communities, and pathways underlying N_2O uptake in agricultural soils. These techniques have been proven to be robust, and particularly the omics methods have been suggested to be very useful, especially when applied together with flux measurement methods, as the omics methods

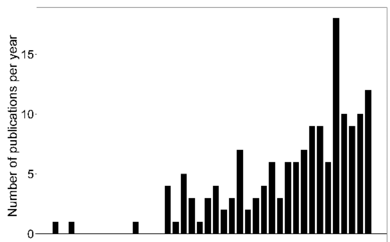

Figure 1. Number of studies reporting N_2O uptake in agricultural soils.

directly assess the soil microbes possessing *nosZ* genes and can reveal relative contributions of different functional groups (e.g. clade I vs clade II) involved in soil N_2O reduction (Hallin *et al.*, 2018; Shan *et al.*, 2021). One very important aspect to focus on in the future is to assess the role of other leguminous food and forage crops in N_2O reduction, as this process has been shown clearly for soybean (Itakura *et al.*, 2013). More importantly, future studies should consider combining $^{15}N_2O$ pool dilution approaches (laboratory and *in situ*) and omics methodologies, along with the assessment of important soil physicochemical characteristics, in the presence and absence of crops of interest, while assessing agricultural soils for net N_2O uptake. Our ongoing research in Finland focuses on understanding the N_2O reduction capacity of a leguminous-grassland ecosystem cultivated with red clover (*Trifolium pratense* L. cv. Ilte) and timothy (*Phleum pratense* L. cv. Nuutti), by using $^{15}N_2O$ pool dilution assays in laboratory and *in situ* conditions along with the omics approach.

Conclusions

To better constrain the global and regional N_2O budget, we need to emphasize both emissions and uptake of N_2O in agricultural ecosystems, especially the latter, which is still neglected in current earth system models of N_2O fluxes. The cause and explanation behind any negative N_2O flux at a given space and time should be addressed by using relevant methodology. Only when a proper understanding of N_2O uptake in agricultural soil is established will we be able to better estimate the agricultural N_2O footprint and develop proper N_2O mitigation strategies for agriculture.

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References

Firestone M.K. and Davidson E.A. (1989) Microbiological basis of NO and N_2O production and consumption in soil. In A.O. Andreae and D.S. Schimel (eds.) *Exchange of trace gases between terrestrial ecosystems and the atmosphere.* Wiley, pp. 7-21.

Flechard C.R. *et al.* (2005) Bi-directional soil/atmosphere N_2O exchange over two mown grassland systems with contrasting management practices. *Global Change Biology* 11, 2114-2127.

Hallin S. *et al.*, (2018) Genomics and ecology of novel N_2O-reducing microorganisms. *Trends in Microbiology* 26, 43-55.

Itakura M. *et al.* (2013) Mitigation of nitrous oxide emissions from soils by *Bradyrhizobium japonicum* inoculation. *Nature Climate Change* 3, 208-212.

Jones C.M. *et al.* (2013) The unaccounted yet abundant nitrous oxide-reducing microbial community: a potential nitrous oxide sink. *The ISME Journal* 7, 417-426.

Reay D.S. *et al.* (2012) Global agriculture and nitrous oxide emissions. *Nature Climate Change* 2, 410-416.

Sanford R.A. *et al.*, (2012) Unexpected nondenitrifier nitrous oxide reductase gene diversity and abundance in soils. *Proceedings of the National Academy of Sciences of the USA* 109, 19709-19714.

Shan J. *et al.*, (2021) Beyond denitrification: The role of microbial diversity in controlling nitrous oxide reduction and soil nitrous oxide emissions. *Global Change Biology* 27, 2669-2683.

Shurpali N.J. *et al.* (2016) Neglecting diurnal variations leads to uncertainties in terrestrial nitrous oxide emissions. *Scientific Reports* 6, 25739.

Tilman D. *et al.* (2001) Forecasting agriculturally driven global environmental change. *Science* 292, 281-284.

Wen Y. *et al.* (2016) Disentangling gross N_2O production and consumption in soil. *Scientific Reports* 6, 36517.

Wu D. *et al.* (2013) N_2O consumption by low-nitrogen soil and its regulation by water and oxygen. *Soil Biology and Biochemistry* 60, 165-172.

Yang W.H. *et al.* (2011) A test of a field-based ^{15}N-nitrous oxide pool dilution technique to measure gross N_2O production in soil. *Global Change Biology* 17, 3577-3588.

Utility value of grasslands in a legally protected area depending on the management

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Abstract

Active protection of meadow communities in national parks is an important element of maintaining their ecosystem functions. Field studies were carried out in Biebrza National Park in Poland (mid-June 2010-2011). The aim of the research was to determine the effect of mowing and grazing on the vegetation of meadow communities. The assessed area was divided into three parts: I – non-mown, II – mown every 2-4 years, III – mown once a year and grazed by Konik horses. Sward cover, species composition, yield of fresh and dry mass, fodder-value score (FVS) and nutritional value of the sward were estimated. The yields of fresh mass from non-mown and mown parts were similar, while that from the area grazed by Konik horses was significantly lower. Floristic composition of non-mown and mown meadows differed only in the proportion of some groups of species. *Molinia caerulea* and *Juncus* sp. had the highest cover in the sward on the non-mown meadow. There were also communities with *Salix* sp. and *Betula pubescens.* On the mown meadow *Carex* sp. dominated, while *Salix* sp. and *M. caerulea* achieved lower shares. There were two communities on the part grazed by Konik horses, one was *Carex* sp. dominant, and one *Agrostis canina* dominant. The sward of all parts of the meadow showed poor FVS and low nutritional value. Although mowing is necessary to prevent succession, grazing even at a very low stocking rate additionally contributes to diversifying the plant communities.

Keywords: biodiversity, cessation of grasslands utilization, feeding value, grazing, moving, national park areas

Introduction

In the last 70 years, the grassland areas in Europe have gradually declined (European Commission, 2020). Deterioration of a naturally valuable habitat occurs as a result of either intensification or abandonment of management practices (Török *et al*., 2018). The succession of invasive species is particularly visible in areas where traditional extensive management (mowing and grazing) has ceased. This threatens, among other things, fen meadows due to the low profitability of their usage and harsh environmental conditions (e.g. high water level). Recent years have increasingly seen the introduction of free-range grazing in areas of high natural value to preserve habitats and maintain or restore the open landscape. While the key role in the protection of lowland grasslands in Europe is attributed to cattle, horses also have a great ability to feed even on low-quality swards (Prache *et al*., 1998). Among the primitive breeds, Polish primitive horses (Koniks) are particularly predisposed for this (Chodkiewicz 2020). The need to use grazing as a form of natural environment protection became a reason for the establishment in 2004 of Koniks breeding reserve in Biebrza National Park (BNP, north-eastern Poland). The aim of the research was to determine the effect of mowing and grazing on the vegetation of meadow communities in Grzędy in BNP.

Materials and methods

The study was conducted in Biebrza National Park (BNP, situated in north-eastern Poland) in the Central Basin North protected district in 2010-2011. The assessed area was divided into three parts: I – non-mown (the last cutting was carried out seven years before the start of the research), II – mown every 2-4 years (the last mowing was carried out 4 years before the research), III – mown once a year and grazed by Konik horses (26 horses in the year, 0.55 LU ha^{-1}) which began 5 years before the research. Horses stayed on the pastures all year long. The vegetative season in the valley lasts 205 days on average. The climate in this area is continental with features of hemi-boreal and is among the coldest in Poland.

During the field studies (mid-June), along transects (one in each part of the meadow) 12 patches of vegetation every 100 m were separated. Within each patch, the sward cover (%) and the range of the main plant height (cm) were estimated in duplicate. In order to investigate the effect of management on the botanical composition of the sward, 500 g samples of fresh mass were taken, and the cover of individual species in the sward was assessed (% in dry mass). The yield of fresh mass (DY) and dry mass (DM) were evaluated according to the formula: mean height of sward (cm) × 0.6 (conversion factor for fresh mass) × sward cover (%). The fodder value score (FVS) defined by Filipek (1973) and content of crude protein (CP), crude fibre (CF), ash-free neutral detergent fibre (NDF), digestibility of organic and dry matter (chemical analyses) were determined. Statistical analyses (one-way ANOVA) of the obtained data (sward height, DY, DM) were performed for all managements.

Results and discussion

The vegetation of the investigated area was classified as *Molinia* meadow. The assessed parts of the meadow clearly differed in terms of cover and height of sward (Figure 1). The sward cover of grass communities, regardless of the method of utilization, was small and ranged from 51% on the grazed area to 62% on the mown meadow. The sward was the highest on the non-mown part (average 70 cm) and the lowest on the grazed area (average 45 cm).

As a result, the available yields of fresh and dry mass in the summer period (June) from non-mown as well as mown meadows were similar (approx. 20.0 Mg ha^{-1} and 9.0 Mg ha^{-1} of DM, respectively). In turn, the yields from the area grazed by Konik horses were significantly lower (11.0 Mg ha^{-1} of DY and 5.0 Mg ha^{-1} of DM). Floristic composition of the patches of parts of the meadow I (non-mown) and II (mown every 2-4 years), was similar, but clearly differed in the proportion of some species or groups of species (Table 1).

Molinia caerulea L. (approx. 33%) and *Carex* sp. (approx. 24%) had the highest cover values in the non-mown sward (I). The occurrence of plant communities with the participation of *Salix rosmarinifolia* L., *S. cinerea* L., *S. aurita* L. and *Betula pubescens* Ehrh. (approx. 16%) was also noticeable, which proves the initiated succession. Dicotyledonous plants had a large cover, among them were observed legally protected orchids (*Dactylorhiza incarnata* (L.) Soó, *Platanthera bifolia* (L.) *Rich.*), while the least were *Phragmites australis* (Cav.) Trin. Ex Steud and other grasses (*Calamagrostis stricta* (Timm) Koeler). On

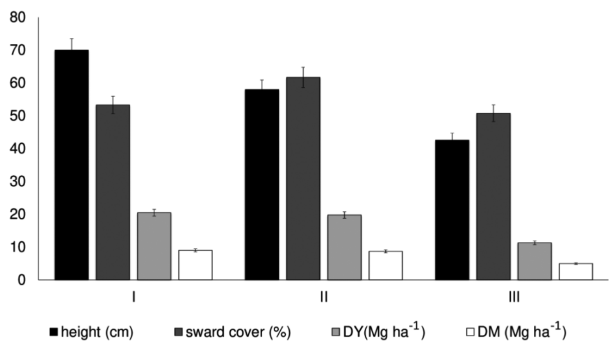

Figure 1. Height and cover of sward, disposable yield of fresh mass (DY) and dry mass yield (DM) of investigated parts of the meadow: I – non-mown, II – mown every 2-4 years, III – mown once a year and grazed by Konik horses.

Table 1. Botanical composition of sward on assessed parts of the meadow: I – non-mown, II – mown every 2-4 years, III – mown once a year and grazed by Konik horses (%).

Object	*Salix* sp.	*Betula pubescens*	*Phragmites australis*	*Molinia caerulea*	*Deschampsia caespitosa*	*Agrostis canina*	Other grasses	*Carex* sp.	*Juncus* sp.	*Dicotyledonous*
I	14.4	1.4	4.8	32.6	0.0	0.0	4.7	23.6	0.0	18.5
II	8.4	0.0	2.8	10.5	0.0	0.0	0.1	63.0	0.0	15.2
III	0.3	0.0	0.5	0.0	4.6	11.6	1.0	49.2	9.3	23.5

the other hand, *Carex* sp. dominated on the sporadically mown meadow (II) (over 60%), while the share of *Salix* sp. was two times lower, and *Molinia caerulea* L. three times lower. On the part of the meadow grazed by Konik horses (III) two communities were distinguished – with dominance of sedges (*Carex panicea* L. and *Carex flava* L.) and *Agrostis canina* L. (Chodkiewicz 2020). According to Kotlarz *et al.* (2010) extensive meadow not fertilized and late mown gives hay with very high share of plants without, low or moderate fodder value score. In our study the sward of mown and non-mown meadows showed a poor utility value (FVS approx. 2), while on the grazed part it was slightly better (FVS 3-4), but still poor. Regardless of the management, the sward of all grass communities was characterized by low nutritional value. This was due to the high content of crude fibre (CF), ash-free neutral detergent fibre (NDF) and low digestibility of organic and dry matter.

Conclusions

The management method significantly affected grassland vegetation and yields on the studied area. All parts of the meadow clearly differed in cover and height of the sward. Regardless of the management, the sward of all grass communities was of low nutritional value.

References

Chodkiewicz A. (2020) Advantages and disadvantages of Polish primitive horse grazing on valuable nature areas – A review. *Global Ecology and Conservation* 21, e00879 https://doi.org/10.1016/j.gecco.2019.e00879

European Commission (2020) Report from the Commission to the Council and the European Parliament: The State of Nature in the European Union. https://www.eea.europa.eu/publications/state-of-nature-in-the-eu-2020

Filipek J. (1973) Project of classification of meadow and pasture plants based on the numbers of utility value. *Postępy Nauk Rolniczych* 20(4), 59-68. (Abstract in English)

Kotlarz A., Stankiewicz S. and Biel W. (2010) Botanic and chemic composition of hay from semi-natural meadow and its nutritive value for horses. *Acta Scientiarum Polonorum, Zootechnica* 9(4), 119-128. (Abstract in English)

Prache S., Gordon I.J. and Rook A.J. (1998) Foraging behaviour and diet selection in domestic herbivores. *Annales de Zootechnie* 47, 335-345.

Török P., Janišová M., Kuzemko A., Rūsiņa S. and Stevanović Z.D. (2018) Grasslands, their threats and management in eastern Europe. In: Squires V.R., Dengler J., Hua L., Feng H. (eds.) *Grasslands of the World: Diversity. Management and Conservation*. CRC Press, Boca Raton, FL, USA, pp. 64-88.

Permanent grassland ecosystems services: farmer perceptions

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Abstract

As part of the Horizon 2020 SUPER-*G* project, we carried out a survey of 50 grassland farmers in Lorraine and Normandy to assess their understanding and knowledge of ecosystem services. Ecosystem services define the goods and services that humans can derive from ecosystems, directly or indirectly, to ensure human well-being (food, water quality, landscapes, etc.) (Tibi *et al.*, 2018). In general, livestock farmers understood the importance of the production ecosystem service as well as the environmental goods and services commonly mentioned in various communications and regulations, namely: carbon storage, water quality, biodiversity, and water flow regulation (Van den Pol-van Dasselaar *et al.*, 2019). However, a main finding of the survey was that a significant proportion (88%) of the 50 farmers was not aware of the term 'ecosystem services'.

Keywords: ecosystem services, permanent grassland, biodiversity, carbon storage, water quality

Introduction

It is generally accepted that the term 'ecosystem services' is not well known within the farming community. Nevertheless, many farmers recognize the multitude of (non-production) services that permanent grassland (PG) provides, particularly biodiversity and carbon storage. This paper investigates the main factors that encourage grassland livestock farmers to provide and enhance ecosystem services on grassland, such as legislation, personal choice, traditional practice, and income diversification.

Materials and methods

Fifty farmers were included in a PG ecosystem services questionnaire survey in 2019: 30 in Lorraine and 20 in Normandy (northern France). 66% were dairy farmers and 33% beef farmers. The qualitative survey was divided into nine sections: (1) farm information, (2) permanent grassland on your farm, (3) soil management, (4) grass management, (5) sward management on improved PG, (6) sward management on unimproved PG, (7) ecosystem services, (8) economics, and (9) innovation and knowledge exchange. This paper focuses on the results from section 7 – ecosystem services. The aim of the survey was to gather information from the farms within the networks regarding their PG management practices, business profitability and their views on the delivery of ecosystem services.

Results and discussion

In general, the term 'ecosystem services' was not well known (Table 1). Nevertheless, the services themselves are generally well understood by farmers who are aware of the importance of their pastures and meadows for the environment, particularly for water and carbon cycles. Supporting biodiversity and pollination were also recognized as important ecosystem services provided by PG. 'Leisure and tourism' services were rarely acknowledged by the farmers.

The farmers were questioned on their practices on permanent grasslands and what ecosystem services would be provided by their practices. Moreover, we asked them about their main motivation to develop this practice. The motivations reported were very diverse, ranging from purely regulatory (especially for the impact on water quality, a service that is very much covered by public policies), to personal choice or to tradition (often reported for providing an attractive landscape and/or biodiversity).

Table 1. Perception of ecosystem services by 50 French farmers.

		Lorraine (n=30)	Normandy (n=20)
Knowledge of the term 'ecosystem services'	Yes	13%	10%
	No	87%	90%
For you, what is the level of your grassland's contribution to these ecosystem services? (Proportion of response 'important and intermediate supply' for each ecosystem services on their farm. Other possible choice low-level supply or no supply)	Forage production	100%	100%
	Carbon storage	96%	100%
	Water quality	88%	90%
	Biodiversity	81%	100%
	Soil erosion	77%	90%
	Pollination	50%	80%
	Leisure, tourism	35%	50%

Table 2. Motivation of ecosystem services by the farmers interviewed.

Practice	Farms employing the practice (Lorraine, n = 30)	Farms employing the practice (Normandy, n =20)	Ecosystem Service produced according to farmers perception	Main reason to employ the practice (Lorraine)	Main reason to employ the practice (Normandy)
Manure plans	17	7	water quality	legislation (100%)	legislation (100%)
Avoiding manure spreading at times of high risk	13	4	water quality	legislation (100%)	legislation (50%)
Maintaining hedges	27	19	biodiversity, carbon storage, erosion control, wind protection, use of wood	usual practices (63%), personal choice (52%), legislation (37%), investment aid (11%)	personal choice (68%), usual practices (32%), tradition of the locally typical *bocage* landscape
Agroforestry	16	8	biodiversity, carbon storage, erosion regulation, production, leisure and tourism	usual practices (69%), personal choice (38%), legislation (13%), investment aid (13%)	personal choice (88%), legislation (12%)
Maintaining species-rich permanent grasslands	16	4	biodiversity, carbon storage, erosion control, production	usual practices (88%), personal choice (19%), income diversification (6%)	personal choice (100%), usual practices (25%)
Buffer strip along watercourse	22	4	biodiversity, carbon storage, erosion control	legislation (100%), usual practices (55%), personal choice (9%)	legislation (50%), personal choice (25%), usual practices (25%)

Legislation is often the main reason for a change of practice. For example, buffer strips along watercourses first appeared for regulatory reasons in 2005, but they are now accepted as 'normal' on 55% of farms and claimed as a personal choice on 9% of farms. Among the widely adopted practices, maintaining hedges and agroforestry in meadows were strongly linked to traditional practices in rural areas (63% for hedges, 69% for agroforestry) and were often associated with grazing practices and the need to provide shelter for grazing animals. Regulations were rarely reported as a motivation for adopting or maintaining these two practices (37% for hedges, 13% for agroforestry). Finally, we underline the existence of local funding opportunities (region, department or county) that triggered tree replanting for around 10% of farmers, which shows that this financial aid was important in the decision-making of farmers.

Conclusions

Only 10% of the 50 farmers questioned knew the term 'ecosystems services', but all farmers were aware of the specific grassland services listed. Many farming practices already exist for supporting the services related to securing water quality and reducing the farm's carbon footprint. These following measures have been identified to favour ecosystem services:

- Aid for adopting and maintaining sustainable farming practices: compensatory measures for the sward renovation.
- Aid for investment for machinery supporting the maintenance of grassland areas and aid for planting hedges.
- Aid for the ecological transition and reduction of GHG emissions, in particular through the sale of carbon credits to companies in the strongly emitting sectors (Cantarel, 2011).
- Biodiversity is an issue that has historically been confined to specific land zoning, in particular Natura 2000 areas. Apart from these areas where compensatory measures existed, the biodiversity issue is now increasingly considered in the territories. Since 2015 the region is in charge of the biodiversity preservation. This is evidenced by the many 'Blue and Green Network' projects aimed at maintaining or recreating ecological continuities (French Ministry for ecological transition, 2019).

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References

Cantarel A. (2011) Impacts du changement climatique sur les bilans de carbone et de gaz à effet de serre de la prairie permanente en lien avec la diversité fonctionnelle. Sciences agricoles. Université Blaise Pascal - Clermont-Ferrand II.

Tibi A. and Therond O. (2018) *Services écosystémiques fournis par les espaces agricoles – Évaluer et caractériser*, Editions Quae, 188pp.

Van den Pol-van Dasselaar A., Kaemena F. and Bastiaansen-Aantjes L. (2019) *Grassland use in Europe – a syllabus for young farmers,* Chapter 5. Editions Quae, pp. 109-130.

Using crowd-sourced data to quantify cultural ecosystem services provided by grasslands in Auvergne, France

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Abstract

Livestock farming systems (LFS) provide multiple services to people including cultural ecosystem services (CES). It is assumed that LFS improve the cultural and recreational attractiveness of landscape by maintaining grasslands that offer wide-open views. Recently, crowd-sourced data derived from social media emerged as an important component in CES studies. They offer massive amounts of data covering broad spatial and temporal scales. This study examines the level of recreational use of each type of land cover across the Auvergne region in France, based on 5,568 geo-located trails from three social media platforms (Flick, NaturaList, and Wikiloc). It quantitatively analysed the pattern of CES potential supply (% of each land cover in Auvergne) and use (% of land cover in areas visited by users). Results show that on average the use of grasslands within visitors' trails is 7% higher than the potential supply, whereas the use of agricultural land is 14% lower. It suggests a significant demand for grassland is high, and a low demand for agricultural lands.

Keywords: cultural ecosystem services, social media, grasslands, livestock, recreation

Introduction

Ecosystem services (ES) are defined as 'the contributions of ecosystem structure and function to human well-being' (Costanza *et al.*, 2017). They include provisioning, regulating, and cultural services. In Europe, most grassland ecosystems result from an interaction of natural and human processes, including livestock grazing (Lemaire *et al.*, 2011). These ecosystems are valued for diverse reasons in response to their benefits to people. They range from forage provision and climate regulation to recreation and leisure activities (Le Clec'h *et al.*, 2019). Understanding interactions between public and grassland ecosystems, and their benefits for well-being, can improve decision making about best practices to preserve them. However, grasslands are generally under-appreciated in CES assessment compared to other habitats, such as forests (Diaz *et al.*, 2015).

Traditionally, the survey questionnaire is the most common tool used in CES studies. However, these methods are tedious, cost and/or time consuming, and mostly used for local assessments (Cheng *et al.*, 2019). At a large scale, social media data are more consistent, geo-localized, and increasingly applied (Ghermandi and Sinclair, 2019). Social media data have been applied to explore human-nature interaction in several ecosystems ranging from marine ecosystems to urban parks. However, relatively little research has been conducted in the quantification of CES related to grasslands and rural landscapes. Therefore, this study aims at quantitatively analysing the importance of grasslands in outdoor activities of three types of social media users (sportive, naturalist, and photographer). The case study concerns the Auvergne region in France.

Materials and methods

Data and study area

Crowd-sourced georeferenced data used in this study were retrieved from three distinct social media platforms: Flickr, NaturaList, and Wikiloc. Flickr is popular with amateur and professional photographers, Wikiloc is the most popular and active outdoor activities platform in European Mediterranean countries (https://www.wikiloc.com), and NaturaList is the largest platform for volunteers to record and share their fauna and flora observations in France (faune-france.org). Flickr and Wikiloc data were retrieved using Application Programming Interfaces (API), R and Python environments. NaturaList data are about bird observations and they were obtained from LPO Auvergne (Ligue pour la Protection des Oiseaux). We combined three diverse platforms in order to include a diversity of profiles in our study. To comply with privacy policies all data were anonymised and any unnecessary data deleted.

The Auvergne region is located around 45°42′ N and 3°18′ E in the centre of France. The region has an area of 26,132 km^2 and is currently home to around 1.3 million inhabitants. The mosaic of natural ecosystems present in the area (geological features, grassland, forest...) together with rural livestock farming, pastures landscape and related traditions (e.g. cheese making), makes it a pertinent study area.

Data analysis

To understand where social media users go on their trips, we used a new approach based on GPS-based trails available in each platform (one trail = one statistical individual). After a preliminary filtering and elimination of trails steps with atypical forms following Callau *et al.*, (2020), a total of 5,568 trails were kept for the analysis. The majority are from NaturaList (n=5,188) uploaded by 462 users, followed by Wikiloc (n=329) for 165 users and Flickr accounts for n=51 trails, uploaded by 29 users (Figure 1A). We used ArcGIS 10.8 to extract the land cover surrounding each trail with a buffer of 100m based on the data issued from the Corine land cover dataset of 2018 (Figure 1B).

Then, we quantitatively assessed the pattern of CES potential supply through the percentage of each land cover in Auvergne. These percentages were weighted based on road and population density, to account for potential differences of accessibility of the land cover types. We then compared the differences between the potential supply and their use, and we considered that differences indicated a specific demand for a given land cover type (i.e. the demand for grasslands is high if this type of land has higher percentages in trails than in the potential supply).

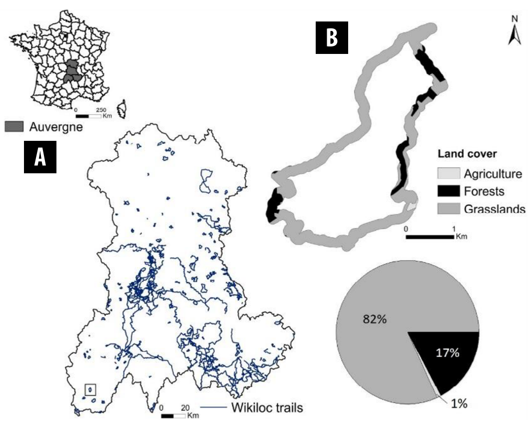

Figure 1. (A) Study area and Wikiloc trails of 2019, (B) example of % of land cover within in a trail.

Results and discussion

Table 1 shows the comparison between land cover used by visitors of each platform and the potential supply in Auvergne. Grasslands were attractive for users of diverse platforms (on average +7%), according to platforms, the frequentation of grasslands increased significantly (value = +4, $P<0.001$) in NaturaList trails compared to the potential supply, in Wikiloc trails (value= +6, $P<0.001$) and in Flickr trails (value= +12, $P<0.01$). In contrast, agricultural lands were unattractive (on average -14%) compared to the potential supply, this non-frequentation was found to be significant in the three platforms ($P<0.001$). Wikiloc trails were made with more forest lands than NaturaList and Flickr trails, while NaturaList trails include more agricultural land and water bodies than Flickr and Wikiloc. It should be mentioned in addition that no land cover type is entirely avoided, which suggests that there are no accessibility differences between them. This tends to validate our approach as accessibility differences may have caused biases in visited areas.

Table 1. Comparison between land cover used by visitors of each social media platform and the potential supply in Auvergne.[1]

	Social media (use %)			
Land cover	**Flickr**	**NaturaList**	**Wikiloc**	**Potential supply % Auvergne**
Forests	28.31	20.28***	42.50***	26.32
Grasslands	45.33**	37.43***	39.79***	33.66
Agricultural land	23.54***	36.15***	16.17***	39.13
Rocks	0***	0.08***	0.10	0.002
Wetlands	0***	0.48***	0.04	0.009
Water bodies	2.82	5.58***	1.405	0.88

[1] For each comparison social media vs potential supply, *** $P<0.001$, ** $P<0.01$, * $P<0.05$ (one-simple t-test).

Conclusions

With the advent of social media data, it is now possible to more easily quantify CES related to diverse types of land cover. Our findings show that grasslands were visited more by social media users than agriculture in their recreation activities. It suggests a high demand for grassland CES and a low demand for agricultural lands. More investigation is needed to explain the motivation behind grasslands frequentation, by integrating, for example, textual metadata in the analysis.

References

Beudou J., Martin G. and Ryschawy J. (2017) Cultural and territorial vitality services play a key role in livestock agroecological transition in France. *Agronomy for Sustainable Development* 37, 36.

Callau A., Pérez-Albert Y. and Serrano D. (2020) Quality of GNSS Traces from VGI: A data cleaning method based on activity type and user experience. *ISPRS International Journal of Geo-Information* 9(12), 727.

Cheng X., Van Damme S., Li L. and Uyttenhove P. (2019) Evaluation of cultural ecosystem services: a review of methods. *Ecosystem Services* 37.

Costanza R., de Groot R., Braat L., Kubiszewski I., Fioramonti L., Sutton P., … and Grasso M. (2017) Twenty years of ecosystem services: How far have we come and how far do we still need to go? *Ecosystem Services* 28, 1-16.

Diaz S., Demissew S., Carabias J., Joly C., Lonsdale M., Ash N., … and Zlatanova D. (2015) The IPBES Conceptual Framework – connecting nature and people. *Current Opinion in Environmental Sustainability* 14, 1-16.

Ghermandi A. and Sinclair M. (2019) Passive crowdsourcing of social media in environmental research: a systematic map. *Global Environmental Change* 55, 36-47.

Le Clec'h, S., Finger R., Buchmann N., Gosal A., Hörtnagl L., Huguenin-Elie O., … and Huber R. (2019) Assessment of spatial variability of multiple ecosystem services in grasslands of different intensities. *Journal of Environmental Management* 251, 109372.

Lemaire G., Hodgson J. and Chabbi A. (2011) *Grassland productivity and ecosystem services*. CABI, Wallingford, UK.

Does herbage protein, fibre, sugar and energy content have an effect on plant mineral content?

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Abstract

The intricacies of interactions between minerals at the soil, plant and animal level are complex. It has been reported that the mineral content of both soil and herbage have decreased over the decades due to changes in agricultural practice and increased herbage yields. The objective of this study was to investigate the relationship between herbage nutritional quality and mineral content. Grass samples were collected over two years and analysed for mineral content and herbage quality. Data were analysed using a Pearson's correlation. Despite fifty-one statistically significant ($P<0.05$) relationships being observed, only ten were deemed of moderate importance. Moderate positive relations were observed between crude protein and magnesium (Mg), sodium (Na), copper (Cu) and zinc (Zn). Moderate negative relationships were observed between water soluble carbohydrates and Mg, calcium (Ca), Na, Cu, Zn and sulphur (S).

Keywords: herbage quality, mineral content, simulated grazing

Introduction

Grassland covers 90% of the agricultural land in Ireland and is the main feed source for ruminants. Minerals are essential in animal nutrition to support optimal performance, reproduction and health. It has been reported that the mineral content of both the soils and in crops have decreased over the decades due to increases of yields and changes in agricultural practices (Ekholm *et al.*, 2007; Guyot *et al.*, 2009). With grass being the sole feedstuff for many ruminants during the grazing season it is important to know the mineral content of this herbage. Herbage quality throughout the grazing season changes so it is important to determine if there is a relationship between herbage quality and mineral content. The objective of this study was to investigate if there are relationships between the nutritional quality and the mineral content of herbage.

Materials and methods

This study was undertaken at the Agri-Food and Biosciences Institute (AFBI), Hillsborough, Northern Ireland. Grass samples were taken from experimental plots as described by Chesney *et al.* (2020). These were 2 × 3 m plots that were harvested based on a 21-day rotation to simulate the optimum grazing interval. The plots were perennial ryegrass (*Lolium perenne)* dominated, as determined by four separate botanical surveys undertaken throughout the year (April, June, August and October). Each sample contained approximately 1 kg of fresh matter from a pooled sample from three plots that were cut to a height of 4-5 cm using an Agria-mower. Herbage samples (n=168) were collected over from April to October in 2018 and 2019 from plots at six different sites. Samples for mineral analysis were collected monthly and analysed using an inductively coupled plasma optical emission spectroscopy (ICP-OES) analysis for phosphorus (P), Mg, Ca, Na, potassium (K), chloride (Cl^-), manganese (Mn), Cu, Zn, selenium (Se), cobalt (Co), iron (Fe) aluminium (Al), molybdenum (Mo), S and lead (Pb) content, whilst iodine was analysed using an inductively coupled plasma mass spectrometry (ICP-MS) analysis. A replicate of the same sample was analysed for nutritional quality (dry matter (DM), CP, acid degradable fibre (ADF), water soluble carbohydrates (WSC) and metabolizable energy (ME)) using near infra-red spectrometry (NIRS). To establish if there were relationships a Pearson's correlation was undertaken.

Results and discussion

Of the potential 85 relationships examined, there were 51 that were deemed significant ($P < 0.05$) based on a Pearson's correlation (Table 1). A. Gordon (AFBI, pers. comm) classifies no relationship, a weak relationship, a moderate relationship and strong relationship as having an R value of 0.00-0.24, 0.25-0.49, 0.5-0.74 and 0.75-1.00, respectively. Therefore, there were only ten moderate and 26 weak relationships identified. Moderate positive relations were observed between CP and Mg, Na, Cu and Zn. Moderate negative relationships were observed between WSC and Mg, Ca, Na, Cu, Zn and S. Although the relationships between mineral content and ADF or ME were weak or non-significant they were seen to mirror each other (if ADF was positive, ME negative with a similar r value) for mineral parameters in the majority of cases. Mg, Zn and Cu are all vital minerals in protein synthesis in the plant (Broadley *et al.*, 2007; Verbruggen and Hermans, 2013; Yruela, 2005). WSC was seen to have a negative correlation with Zn, Mg and Cu, which can be related to the fact that WSC and CP of the grass plant are known to be negatively correlated and carbohydrates are used for protein synthesis (Marschner, 2012). This therefore means that as the sugar content in the leaf builds up the rate of photosynthesis reduces and therefore the CP of the plant falls. The negative relationship between Ca and WSC can be related to the fact the Ca is needed to transport WSC to other parts of the plant and is an important part of the cell wall, so as growth increases there is a decrease in the WSC and the mobilization of Ca (Joham, 1957; Marschner, 2012). This relationship between S and WSC, as when S is deficient, carbohydrates build up in plants as there is no synthesis of amino acids or other materials needed for protein synthesis and therefore the WSC cannot be used (Marschner, 2012). Sodium was seen to increase the uptake of nutrients and increases protein content of the plants (Varga and Ducsay, 2003).

Table 1. Pearson's correlation between mineral content and herbage quality factors.[1]

		DM (%)	CP (% DM)	ADF (% DM)	WSC (% DM)	ME (MJ kg DM^{-1})
P (g kg^{-1})	*R*	-0.34	0.31	0.29	-0.34	-0.28
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001
Mg (g kg^{-1})	*R*	-0.47	0.66	0.41	-0.73	-0.41
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001
Ca (g kg^{-1})	*R*	-0.42	0.43	0.38	-0.59	-0.38
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001
Na (g kg^{-1})	*R*	-0.27	0.59	0.15	-0.53	-0.14
	Sig.	<0.001	<0.001	0.06	<0.001	0.07
K (g kg^{-1})	*R*	-0.11	-0.06	-0.08	0.09	0.09
	Sig.	NS	NS	NS	NS	NS
Cl^- (g kg^{-1})	*R*	0.45	-0.40	-0.27	0.48	0.27
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001
Mn (mg kg^{-1})	*R*	0.07	0.12	-0.22	0.03	0.21
	Sig.	NS	NS	NS	NS	NS
Cu (mg kg^{-1})	*R*	-0.39	0.60	0.20	-0.60	-0.21
	Sig.	<0.001	<0.001	0.01	<0.001	0.01
Zn (mg kg^{-1})	*R*	-0.38	0.54	0.22	-0.55	-0.22
	Sig.	<0.001	<0.001	0.005	<0.001	0.004
Se (mg kg^{-1})	*R*	-0.05	0.09	-0.01	-0.09	0.01
	Sig.	NS	NS	NS	NS	NS
Co (mg kg^{-1})	*R*	-0.16	0.14	-0.01	-0.20	-0.02
	Sig.	NS	NS	NS	NS	NS
I (mg kg^{-1})	*R*	0.06	-0.09	-0.18	0.14	0.17
	Sig.	NS	NS	NS	NS	NS
Fe (mg kg^{-1})	*R*	-0.16	0.06	0.00	-0.08	-0.02
	Sig.	NS	NS	NS	NS	NS
Al (mg kg^{-1})	*R*	-0.14	0.08	-0.04	-0.11	0.01
	Sig.	0.07	NS	NS	NS	NS
Mo (mg kg^{-1})	*R*	0.10	-0.18	-0.20	0.26	0.19
	Sig.	NS	0.02	0.01	<0.001	0.01
S (g kg^{-1})	*R*	-0.32	0.28	0.47	-0.50	-0.47
	Sig.	<0.001	<0.001	<0.001	<0.001	<0.001
Pb (mg kg^{-1})	*R*	-0.15	0.22	0.01	-0.27	-0.04
	Sig.	0.06	0.005	NS	<0.001	NS

[1] NS = not significant.

Conclusions

In the analysis of this data set a number of relationships were seen between DM, CP, ADF, WSC and ME and herbage mineral content, with the exception of K, Se and Al.

References

Broadley, M. R., White, P. J., Hammond, J. P., Zelko, I. and Lux, A. (2007) Zinc in plants. *New Phytologist* 173, 677-702.

Chesney, L., Scollan, N., Gordon, A., McConnell, D.A. and Lively, F.O. (2020) Investigating the effect of nitrogen application level on grass production and quality under simulated grazing. *Grassland Science in Europe* 25, 55-57.

Ekholm, P., Reinivuo, H., Mattila, P., Pakkala, H., Koponen, J., Happonen, A., Hellström, J. and Ovaskainen, M.-L. (2007) Changes in the mineral and trace element contents of cereals, fruits and vegetables in Finland. *Journal of Food Composition and Analysis* 20, 487-495.

Guyot, H., Saegerman, C., Lebreton, P., Sandersen, C. and Rollin, F. (2009) Epidemiology of trace elements deficiencies in Belgian beef and dairy cattle herds. *Journal of Trace Elements in Medicine and Biology* 23, 116-123.

Joham, H. E. (1957) Carbohydrate distribution as affected by calcium deficiency in cotton. *Plant Physiology* 32, 113-117.

Marschner, P. (2012) *Marschner's mineral nutrition of higher plants.* Academic Press, London, UK.

Varga, L. and Ducsay, L. (2003) Influence of sodium humate on the yield and quality of green pepper. *Horticultural Science* 30, 116-120.

Verbruggen, N. and Hermans, C. (2013) Physiological and molecular responses to magnesium nutritional imbalance in plants. *Plant and Soil* 368, 87-99.

Yruela, I. (2005) Copper in plants. *Brazilian Journal of Plant Physiology* 17, 145-156.

Fate of recently fixed C in plant-soil monoliths from permanent grasslands

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Abstract

Permanent grasslands are recognized for their ability to store C, but little is known about the role of C rhizodeposition in the C storage, particularly for multispecies permanent grasslands. A study was carried out on semi-natural grasslands located in French Natural Regional Parks (Normandie Maine and Lorraine) to analyse the relationships between soil organic content (SOC), C rhizodeposition and C storage. Monoliths from three low SOC and three high SOC fields were used to perform a 5-day $^{13}CO_2$-labelling experiment to measure rhizodeposition and storage of photosynthetic-C. At the end of the labelling period, the amount of labelled-C recovered in the soil was correlated to the amount of C fixed by the monolith during the labelling period, showing the strong link between photosynthesis and rhizodeposition. In addition, a high proportion of labelled-C, from 24 to 54% depending on the grassland, was allocated to the soil compartment. Twelve weeks later, between 6 and 63% of the C fixed during the labelling period was recovered in the soil. This proportion increased with the clay content of the soil but a strong variation was observed between soils with high clay content, indicating that this edaphic factor was not the only driver.

Keywords: C storage, rhizodeposition, ^{13}C-labelling, soil organic matter, permanent grassland

Introduction

Grasslands are recognized as ecosystems with a high potential for mitigating increasing atmospheric carbon dioxide (CO_2) (Soussana and Lemaire, 2014). This C input is due to the transfer of atmospheric C by plants into the soil, either as root and shoot litter after plant death or as C released by living roots through rhizodeposition (Pausch and Kuzyakov, 2018). Rhizodeposition regulates a wide range of ecological soil functions and produces dramatic changes in the biological, chemical and physical nature of the rhizosphere (Nguyen, 2003). The review by Pausch and Kuzyakov (2018) showed that grasslands allocate more C belowground (33%) than crops (20%). However, most studies concerning grasslands have been performed for monospecific culture or young highly fertilized grasslands (Henneron *et al.*, 2020). The aims of this work were to quantify the C rhizodeposition by permanent grasslands and to study its relationship with C storage and soil organic matter content (SOM). Moreover, we aimed to determine the effect of soil traits (clay and SOC) on C input.

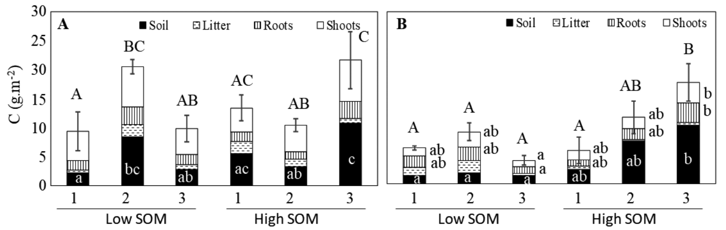
Materials and methods

The study was realized with monoliths taken from six mown grasslands, over 40 years old, located in two Regional Natural Parks selected for the Ademe REACTIFF project 'P^2C' (Morvan-Bertrand *et al.*, 2019). Three grasslands ('Low SOM') were characterized by low SOM (5 to 10% in 0-10 cm topsoil) and low clay content (15 to 25%). Three others ('High SOM') had high SOM (13.5 to 17.5%) and high clay content (44 to 49%). For the labelling experiment, in each grassland, 12 monoliths (8 cm diameter × 30 cm deep) were sampled along a line every 10 m with a corer. Monoliths were kept for 1 night at 5 °C, placed in PVC tubes (same size), transferred into the greenhouse and shoots were cut at 5 cm height. Plants were grown for 6 to 8 weeks with additional artificial lighting providing 400 μmol m^{-2} of photosynthetically active radiation (PAR) at plant height with a photoperiod of 16 h and a temperature of 22/18±2 °C (day/night). Six to eight weeks after the beginning of the acclimation period, 8 monoliths from each field were introduced into a labelling chamber. Control monoliths were harvested for ^{13}C

natural abundance determination before the labelling procedure. Monolith labelling lasted for 5 d and the procedure was similar to that used by Kante *et al.* (2021). At the end of the labelling period, 4 monoliths from each grassland were harvested immediately (T0) and the others were transferred into the greenhouse for 12 weeks (T12). For each sampling point, plant shoots were cut and the litter was harvested. Roots were carefully separated from the soil. Plant tissues and soil were dried at 60 °C until constant mass and ball milled. C and N amounts and ^{13}C enrichments in plant compartments (shoot, litter, root) and soil were determined using an isotope ratio mass spectrometer (IRMS, Isoprime, GV Instruments). The amount of labelled C recovered in the soil at the end of the labelling period was used to estimate C rhizodeposition during the labelling period. The amount of labelled C in the soil recovered in the soil 12 weeks after the end of the labelling period (T12) was used to estimate C storage capacity of soil. Comparisons of amounts of labelled C between stations were performed using one-way ANOVAs (R version 3.5.0).

Results and discussion

A strong effect of the original grassland of the monoliths on the total amount of labelled C recovered at the end of the 5-d labelling was observed (F=5.05, P<0.01). This amount ranged from 9.33 g m^{-2} (Low SOM1) to 21.61 g m^{-2} (High SOM3) (Figure 1A). On average, 40% of the labelled C was recovered in the shoots. Similar proportions are generally observed for temporary grasslands; this is lower than those observed for crops (Pausch and Kuzyakov, 2018). Only 15% of this newly assimilated C was recovered in the roots and 9% in the litter, and the differences between grasslands were not significant in any of the plant compartments (shoot, root and litter). The proportion of labelled C recovered belowground (root+soil, mean 51%) is high compared with previous studies (Pausch and Kuzyakov, 2018; Saggard and Hedley, 2001). This result can be explained by the high root:shoot ratio in such old grasslands (mean 2.20 compared with less than 1 in most of the studies) and the length of the labelling period (5 days compared with 1 day for most of the studies). At the end of the labelling period, the proportion of newly assimilated C recovered in the soil varied strongly (F=7.4; P<0.001) from 24 (Low SOM1) to 54% (High SOM3) and the monoliths exhibiting the greater amounts of labelled C in the soil were also those with the greater amounts of total labelled C (Figure 1A; Low SOM2 and High SOM3). Consequently, a strong correlation was observed between the amount of C fixed per monolith and the amount of labelled C recovered in the soil at the end of the labelling period (r=0.7873, P<0.001), showing the strong link between photosynthesis and C rhizodeposition. However, the original SOM content had no significant effect and does not appear to discriminate grasslands when focusing on C photosynthetic fixation and rhizodeposition.



SOM (% DM)	5.5	3.8	3.1	13.5	14.3	13.0	5.5	3.8	3.1	13.5	14.3	13.0
Clay (% DM)	15.9	21.2	24.3	44.0	46.3	48.7	15.9	21.2	24.3	44.0	46.3	48.7

Figure 1. Amounts of labelled C recovered at the end of the 5-day labelling period (T0, A) or 12 weeks after the end of the labelling period (T12, B) in the various plant compartments and in soil for each grassland. The data represent the mean values (± standard error, n=4). Average SOM and clay content (% DM) for each grassland are given below the graphs. Means with similar letters (upper case letters for total amounts, lower case letters for compartments) are not significantly different (P<0.05).

The amounts of labelled C incorporated during the labelling period and recovered in each compartment 12 weeks later (T12) are shown in Figure 1B. As expected, these amounts decreased in shoots between the two harvests, due to respiration and C transfer belowground. However, a significant amount of the C fixed during the labelling period was still recovered in the shoots. This amount was also strikingly high in roots (average 2.02 g m^{-2}). These results contrast with those obtained with most pulse labelling studies showing the quick release of labelled C from plant organs (Kaštovská and Šantrůčková, 2007; Pausch and Kuzyakov, 2018). It is likely that the length of the labelling period has allowed a significant proportion of labelled C to be stored in long-term reserves and cell-wall compounds. The amount of labelled C remaining in the soil varied strongly among grasslands, from 4.08 g m^{-2} (Low SOM3) to 17.63 g m^{-2} (High SOM3) (Figure 1B). A correlation was observed between the amount of soil labelled C and SOM (r=0.511, P<0.05) showing the strong link between C inputs in the soil of grasslands and their original SOM. In line with numerous studies showing the major impact of clay on soil organic matter stabilization (Paul, 2016), we observed a correlation between the amount of soil labelled C at the end of the experiment and clay content (r=0.459, P<0.05). However, the strong variation observed between soils with high clay content (Figure 1B, High SOM) indicates that this edaphic factor was not the only driver.

Conclusions

This work shows that great amounts of C can be rhizodeposited and stored by permanent grasslands and evidenced a strong link between photosynthesis and rhizodeposition. These amounts vary greatly from one grassland to another depending not only on the soil texture but also on other drivers which remain to be determined.

Acknowledgements

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References

Henneron L., Cros C., Picon-Cochard C., Rahimian V. and Fontaine S. (2020) Plant economic strategies of grassland species control soil carbon dynamics through rhizodeposition. *Journal of Ecology* 108, 528-545.

Kante M., Riah-Anglet W., Cliquet J.B. and Trinsoutrot-Gattin I. (2021) Soil enzyme activity and stoichiometry; linking soil microorganism resource requirement and legume carbon rhizodeposition. *Agronomy,* doi.org/10.3390/agronomy11112131

Kaštovská E. and Šantrůčková H. (2007) Fate and dynamics of recently fixed C in pasture plant–soil system under field conditions. *Plant and Soil* 300, 61-69.

Morvan-Bertrand A., Amiaud B., Mischler P., Klumpp K., Vécrin R., Husse S., Louault F. Cliquet J.B., Lemauviel-Lavenant S. (2019) P^2C, la plante: pilote de la capture et du transfert de carbone vers le sol des prairies – *Rapport final.* ADEME, Angers, France. 41 pp.

Nguyen C. (2003) Rhizodeposition of organic C by plants: mechanisms and controls. *Agronomie* 23, 375-396.

Paul A. (2016) The nature and dynamics of soil organic matter: plant inputs, microbial transformations and organic matter stabilization. *Soil Biology and Biochemistry* 98, 109-126.

Pausch J. and Kuzyakov Y. (2018) Carbon input by roots into the soil: quantification of rhizodeposition from roots to ecosystem scale. *Global Change Biology* 24, 1-12.

Soussana J.F. and Lemaire G. (2014) Coupling carbon and nitrogen cycles for environmentally sustainable intensification of grasslands and crop-livestock systems. *Agriculture, Ecosystems & Environment* 190, 9-1.

Microbiota diversity of the phyllosphere of pastures plants

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Abstract

The diversity of microorganisms located at the leaf surface is largely unexplored in pastures whereas it constitutes a source of microbial diversity for raw milk. Our objective was to investigate the microbial diversity of the phyllosphere of three dominant plant species from permanent grassland managed by cattle grazing. Leaf samples of *Lolium perenne*, *Holcus lanatus* and *Trifolium repens* were collected at three different times of the year in two grasslands. Total bacterial DNA from the leaf surface (phylloplane) was extracted and a 16S-based metagenomic approach was performed. The localization of microorganisms at the leaf surface was studied by scanning electron microscopy. *Methylobacterium* was the dominant bacterial genus and its abundance varied among plant species. The structure of the bacterial community was determined mainly by the identity of the host species and was related to the total water-soluble carbohydrate and sucrose contents of the leaves. This work constitutes the first insight into the microbiota of the phylloplane in relation to the biochemical composition of leaves of different grassland species.

Keywords: microbiota, phyllosphere, grassland, water-soluble carbohydrates, grazing

Introduction

The phyllosphere surface is estimated to represent over 10^8 km^2 over the globe (Lindow and Brandl, 2003). Thus, leaves provide a very large microbial habitat, where bacteria are often found in numbers averaging 10^4 to 10^5 cells mm^{-2} of leaf surface (up to 10^8 cells g^{-1} of leaf), outnumbering the cells of the plants themselves (Lindow and Brandl, 2003; Penuelas and Terradas, 2014). Given the great plant diversity of some grasslands, and in particular of permanent grasslands, their phyllosphere might constitute a huge reservoir of microbial diversity. Its composition and the factors which influence its fluctuations are barely known. The aim of the current study was to investigate the structure of the phyllosphere microbiota of three pasture plant species.

Materials and methods

Sample collection was performed in two permanent grasslands of the INRAE Experimental Domain of Le Pin-au-Haras (Normandy, France) that are managed by cattle grazing. In each pasture, three 3 sampling zones were defined, and sampling campaigns took place in 2016 at three timepoints, in early summer, late summer and autumn. A total of 54 samples (2 pastures × 3 zones × 3 plant species × 3 periods) were collected. For each sample, plant tissues 5 cm above ground were harvested from 3 plant species (*Holcus lanatus*, *Lolium perenne* and *Trifolium repens*). Per sampled plant, 30 g of fresh plant tissue were sampled for bacterial analysis and 5 g for biochemical analysis. To localise and observe the microbial communities on leaf surfaces, microscopy analysis was conducted. Leaf tissues were harvested at the same date and on the same plant species. For *H. lanatus* and *L. perenne*, a 1-cm long blade section located at 1 cm above the ligule was harvested from the last mature leaf of a tiller. For *T. repens*, the central leaflet was harvested of the last mature leaf of a stolon. Leaves were fixed with 1% glutaraldehyde in 0.1 M phosphate buffer pH 7.0 for one week at 4 °C. Samples were rinsed in 0.1 M phosphate buffer pH 7.0, and then dehydrated to critical point in 70-100% progressive ethanol bath (CPD 030 LEICA Microsystem). The cells were pulverized with platinum and observed under a JEOL 6400F scanning electron microscope (JEOL, Croissy sur Seine, France). For bacterial DNA analysis, 30 g of leaves were immersed in 270 ml PBS buffer containing 0.1% Tween 20 in sterile stomacher bags. The bags were shaken at 110 rpm for 5 min at room temperature before a sonication step of 30 s (2 s pulses at 40 W and 1 s pause between pulses). The suspensions were centrifuged at 4,500 rpm for 5 min at 4 °C and the pellet was washed once in 10 ml PBS buffer and centrifuged again at 4,500 rpm for 5 min at 4 °C. Total DNA was extracted from the pellets using the PowerFood microbial DNA kit (Mobio). Amplification of the V1-V3 region (~ 500 bases) of the 16S rRNA gene, amplicon libraries construction using the InView™ Microbiome Profiling 2.0 service, and Illumina MiSeq paired-end sequencing (2×300 pb) were performed at GATC Biotech (Konstanz, Germany). Sequence analysis was performed using the Galaxy-supported pipeline FROGS (Escudie *et al.*, 2018) and statistical analysis using Phyloseq R package implemented in FROGS (FROGSSTAT). Statistical analysis of alpha diversity was performed using the Wilcoxon test in R (3.5.4). The Bray-Curtis distance was used to obtain dissimilarity matrices and the major gradient in bacterial community structure among samples was summarized with a nonmetric multidimensional scaling ordination (NDMS) with an analysis of the correlation between the ordination factors and water-soluble carbohydrate (WSC) and sucrose content. WSC extraction and analysis were carried out as described in Volaire *et al.* (2020).

Results and discussion

The scanning electron micrographs revealed a great epiphytic microbial diversity on the lower leaf surface (Figure 1). Microorganisms are solitary or on aggregates. Many bacteria were present with a large diversity of form and length of cells such as rod-shaped bacillus-like cells and other coccus-like cells. The three plants also hosted fungal hyphae and spores, and yeasts. The 16S rRNA gene-based metagenomics analysis exhibited similar trends in genera composition for *L. perenne*, *H. lanatus*, and *T. repens*. Differences in microbial community structure among the three plant species appeared by Principal Coordinates Analysis (PCoA) based on Bray-Curtis distances (Figure 2A). The phyllosphere bacterial community composition varied among plant species whereas it did not vary according to the pasture and to the collecting period. The phyllosphere bacterial communities of *T. repens* samples were dominated by *Methylobacterium*. *Sphingomonas* and *Pedobacter* were the second and third most abundant genera in the *T. repens* microbiome. The bacterial microbiota community composition of *L. perenne* and *H. lanatus* were more closely related, with *Hymenobacter, Methylobacterium* and *Pedobacter* genera each adding up 4 to 19% of the sequences depending on the sample. Observed species numbers based on OTUs were significantly higher for *H. lanatus* than for the other two plants, indicating a more diverse bacterial community on *H. lanatus* leaves. The *L. perenne* bacterial community was significantly the least diverse of the three plant species. According to NMDS ordination (Figure 2B), when the plant species were taken together, the structure of the microbiota was significantly driven by the levels of total WSC (P=0.002; R^2=25.6%; permutational multivariate ANOVA on Bray-Curtis distances) and sucrose (P=0.023; R^2=14.2%).

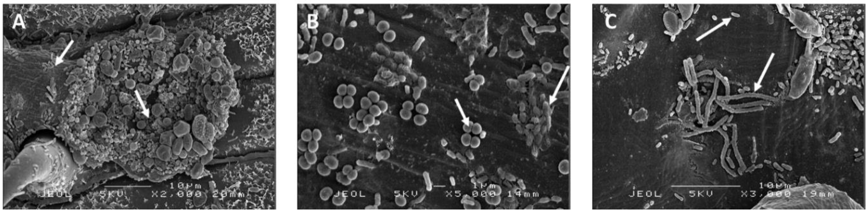

Figure 1. Scanning electron microphotographs illustrating the diversity of microorganisms colonizing the lower leaf surface of *Holcus lanatus* (A), *Lolium perenne* (B) and *Trifolium repens* (C).

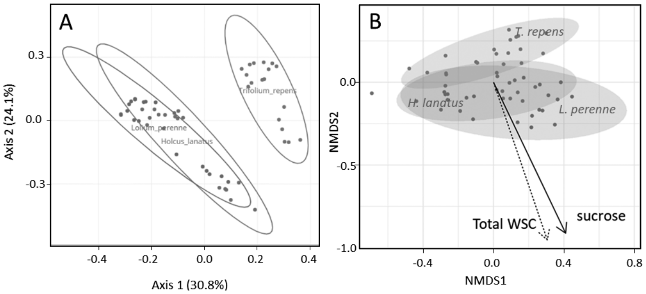

Figure 2. PCoA plots of the phyllosphere of the three plants (A) and nonmetric multidimensional scaling (NMDS) ordination of variation in bacterial community structure based on Bray-Curtis distances in relation with total WSC and sucrose content (B).

Conclusions

Our results show that the microbiota composition of pasture plant leaf is mainly determined by plant species identity as observed among tree species by Laforest-Lapointe *et al.* (2017). Differences in microbiota composition among plant species might depend on plant phenotypic traits, such as leaf morphology. Another main driver of the bacterial community of the phylloplane of grassland was the total water-soluble carbohydrate (WSC) and sucrose contents of the leaves. This result, together with the known roles of soluble carbohydrates for microbe nutrition and plant immune responses, suggests that microbial community of the phylloplane is correlated to soluble carbohydrate composition of the leaves.

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References

Escudie F., Auer L., Bernard M., Mariadassou M., Cauquil L., Vidal K., Maman S., Hernandez-Raquet G., Combes S. and Pascal G. (2018) FROGS: Find, Rapidly, OTUs with Galaxy Solution. *Bioinformatics* 34, 1287-1294.

Laforest-Lapointe I., Messier C. and Kembel S.W. (2017) Tree leaf bacterial community structure and diversity differ along a gradient of urban intensity. *mSystems* 2, 1-16.

Lindow S.E. and Brandl M.T. (2003) Microbiology of the phyllosphere. *Applied and Environmental Microbiology* 69, 1875-1883.

Penuelas J. and Terradas J. (2014) The foliar microbiome. *Trends in Plant Science* 19, 278-280.

Volaire F., Morvan-Bertrand A., Prud'homme M.-P., Benot M.-L., Augusti A., Zwicke M., Roy J., Landais D. and Picon-Cochard C. (2020) The resilience of perennial grasses under two climate scenarios is correlated with carbohydrate metabolism in meristems. *Journal of Experimental Botany* 71, 370-385.

Prediction of hay digestibility from its assessment on the fresh forage and drying time

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Abstract

Organic matter digestibility (OMd) of hay depends mainly on OMd of the corresponding fresh forage. Relationship between OMd in fresh and conserved forages have been used to estimate the feed value of conserved grass forages in INRAE Feed tables since 1989. During this period, evolution of machinery in hay making as well as climate change may have modified the relationships. The aim of this work was to revisit and refine the prediction of hay OMd from fresh forage assessment. Twenty plots of multi-species grasslands were studied in three semi-mountain areas. The drying time (DT) was recorded from cutting to baling or entrance in the barn of the hay. Pepsin cellulase dry matter digestibility (PCDMD) was measured on both fresh forages and hays to predict OMd. The OMd of hay was highly and positively related to the corresponding OMd measured on fresh forage (R^2=0.91; RMSE = 0.016). The addition of DT in the prediction equation slightly improved its accuracy (R^2=0.94; RMSE = 0.013). The coefficient of DT in the prediction equation was negative consistently with the negative correlation of DT with OMd of hay (R Spearman = -0.66; P<0.01). Finally, OMd of hay depends firstly on the OMd in the corresponding fresh forage, in relation with maturity stage and botanical composition of the grassland, and secondly on the drying time between cutting and hay baling.

Keywords: hay, fresh forage, digestibility, drying time

Introduction

Organic matter digestibility (OMd) is the major determinant of forage feed value. Its early prediction at harvesting of the forage allows farmers to know in advance the forage value when preparing conserved-grass based diets fed to animals in winter. Relationship between OMd in fresh and conserved forages are used to estimate the feed value of conserved grass forages in INRAE Feed tables (Baumont *et al.,* 2018; Demarquilly *et al.,* 1989). Since the 1980s, climate and machinery in haymaking have evolved (Deroche *et al.*, 2020; Hartmann *et al.,* 2013; Savoie, 2001) and may have modified these relationships. The aim of this work was to revisit and refine the prediction of hay OMd from fresh forage assessment.

Materials and methods

During 2017, twenty plots of multi-species grasslands, including four sown plots intensively and 16 permanent pasture plots extensively managed, were studied in three semi-mountainous areas: Friburg foothills (Switzerland; n=4), Jura (France; n=8) and Massif Central (France, n=8). The plots were studied during the 1st vegetation cycle except four of the eight plots in Massif Central that were studied during the 2nd vegetation cycle. For each plot, representative samples of fresh forage were collected at cutting and of the corresponding hay after drying and storage that lasted between 149 and 348 days (261±79.6 days on average). The drying time (DT) was recorded for each plot between cutting and baling for sun dried hays (n=16) or between cutting and the entrance in the barn for barn dried hays (n=4). Dry matter (DM), ash, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), and pepsin cellulase dry matter digestibility (PCDMD) according to Aufrère *et al.* (2007) were measured on all fresh forage and hay samples. OMd was predicted from PCDMD on both hay and fresh forage (Aufrère *et al.*, 2007). Correlations between OMd of hay and chemical criteria of fresh forage or DT were calculated using Spearman's correlation. Stepwise linear regression procedure (SAS 5.1) was used to establish prediction equations of OMd of hay from chemical constituents of fresh forage and DT.

Results and discussion

On average, the grasslands contained 75.9±10.9% of grass species (between 57.2 and 93.9%), 7.6±9.4% of legumes (0.2-35.8%) and 16.5±9.1% of forbs (2.5-35.0%). The average stage of maturity at cutting was heading (between stem elongation and flowering) and onset of stem elongation for 1st and 2nd vegetation cycle respectively.

As a result of partial consumption of soluble constituents of plants during the drying process, mean NDF and ADF content increased, whereas CP content decreased between fresh forage and hay (Table 1). As a consequence, OMd of hays decreased compared to the corresponding fresh forage (Table 1) in accordance with previous observations (Demarquilly *et al.*, 1989; Baumont *et al.,* 2018). The range of variation of OMd in fresh forages and hays was high (8.5 points of digestibility) which makes necessary to establish prediction equations (Table 1). Correlations of OMd of hay with OMd of fresh forage were highly positive, and negative with NDF and ADF (Table 1), as found by Andueza *et al.* (2019). The OMd of hay was also negatively correlated with DT (R=-0.655; P<0.01).

Moreover, OMd of fresh forage explained a high part of variability of OMd of hay (R^2=0.909; RMSE = 0.0163) (Table 2), which confirms the previous relationship established by Demarquilly *et al.* (1989). On this dataset the prediction of OMd of hay from the ADF content of the fresh forage is of similar accuracy (Table 2). The introduction of hay DT improved significantly but only slightly the accuracy of the prediction equation of hay OMd, whether from OMd or ADF of the corresponding fresh forage (Table 2). This may be explained by the fact that (1) the accuracy of OMd prediction from single variable is already high, and (2) the DT between cutting and harvesting hay was short in this study due to good weather conditions (2.24±0.497 and 1.70±0.320 days for 1st and 2nd vegetation cycle, respectively).

Conclusions

This study confirms that OMd of hay depends firstly on the OMd and the fibre content of the corresponding fresh forage measured at cutting, in relation with the maturity stage and botanical composition of the grassland. When the drying time between cutting and hay baling is short (below 3 days), this criterion improves only slightly the accuracy of the prediction of the OMd of the hay from the characteristics of the fresh forage. A larger number of samples should allow to strengthen these relationships.

Table 1. Chemical composition and OMd of the fresh forages and the corresponding hays from multi-species grassland plots (n=20), and Spearman's correlation coefficients between OMd of hay and chemical constituents of fresh forages.[1]

	Fresh forage				Hay				Correlation with OMd of hay
	Mean	SD	Min	Max	Mean	SD	Min	Max	
DM (g kg^{-1})	241	34.3	179	325	914	2.46	908	918	-0.666**
Ash (g kg^{-1} DM)	80.4	15.5	57.9	112	76.2	8.97	62.2	93.2	0.474*
CP (g kg^{-1} DM)	116	19.0	93.8	152	107	17.9	81.2	138	0.627**
NDF (g kg^{-1} DM)	541	67.1	411	654	565	70.1	433	678	-0.958***
ADF (g kg^{-1} DM)	282	43.9	209	365	296	43.8	228	376	-0.941***
OMd (g g^{-1})	0.710	0.0520	0.617	0.795	0.666	0.0528	0.571	0.751	0.910***

[1] SD = standard deviation; Min = minimum; Max = maximum; OMd = organic matter digestibility predicted by pepsin cellulase dry matter digestibility (Baumont *et al.*, 2018). Significance: *** P<0.001; ** P<0.01; * P<0.05.

Table 2. Prediction models of OMd of hay (H) (g g^{-1}) by OMd, chemical constituents measured on the corresponding fresh forage (FF) and hay DT (n=20).[1]

Model components	R^2	RMSE
OMd_H = -0.0223 + 0.970 × OMd_FF (g g^{-1})	0.909	0.0163
OMd_H = 0.9907 – 0.00115 ×ADF_FF (g kg^{-1} DM)	0.915	0.0158
OMd_H = 0.1010 + 0.862 × OMd_FF (g g^{-1}) – 0.0219 × DT (days)	0.936	0.0133
OMd_H = 1.0115 – 0.1006 × ADF_FF (g kg^{-1} DM) – 2.823 × DT (days)	0.956	0.0111

[1] RMSE = root mean square error.

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References

Andueza D., Picard F., Pradel P. and Theodoridou K. (2019) Feed value of barn-dried hays from permanent grassland: a comparison with fresh forage. *Agronomy* 9, 273.

Aufrère J., Baumont R., Delaby L., Peccatte J.R., Andrieu J., Andrieu J.P. and Dulphy J.P. (2007) Prévision de la digestibilité des fourrages par la méthode pepsine-cellulase. Le point sur les équations proposées. *INRA Productions Animales* 20, 129-136.

Baumont R., Tran G., Chapoutot P., Maxin G., Sauvant D., Heuzé V., Lemosquet S. and Lamadon A. (2018) INRA feed tables used in France and temperate areas. In: Nozière P., Sauvant D. and Delaby L. (eds.) *INRA feeding system for ruminants.* Wageningen Academic Publishers, Wageningen, the Netherlands, pp. 441-458.

Demarquilly C., Andrieu J.P., Michalet-Doreau B. and Sauvant D. (1989) Measurement of the nutritive value of feeds. In: Jarrige R. (eds.) *Ruminant nutrition: Recommanded Allowances & Feed Tables,* John Libbey Eurotext, Paris, France, pp. 193-212.

Deroche B., Pradel P. and Baumont R. (2020) Long-term evolution and prediction of feed value for permanent mountain grassland hay: Analysis of a 32-year data set in relation to climate change. *Grass and Forage Science* 75(1), 18-27.

Hartmann D.L., Klein Tank A.M.G., Rusticucci M., Alexander L.V., Brönnimann S., Charabi Y., Dentener F.J., Dlugokencky E.J., Easterling D.R., Kaplan A., Soden B.J., Thorne P.W., Wild M. and Zhai P.M. (2013) Observations: Atmosphere and surface. In: *Climate Change 2013: The physical science basis. Contribution of working group I to the fifth assessment report of the intergovernmental panel on climate change.* Cambridge University Press, Cambridge, UK, pp. 159-254.

Savoie P. (2001) Intensive mechanical conditioning of forages: A review. *Canadian Biosystems Engineering* 43, 2.1-2.12.

Prediction of water-soluble carbohydrate contents in hay from their content in fresh forage and drying time

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Abstract

Soluble carbohydrates influence the forage feed value. Total water-soluble carbohydrates (WSC) content and composition, particularly fructans, may change during grass drying after cutting. We aimed to predict WSC and fructans contents in hay from their content measured in fresh forage and the time of drying, which is known to induce a decrease in the content of soluble constituents. Twenty plots of multi-species grasslands were studied in three semi-mountain areas. Biochemical components were measured on fresh forage at cutting and the corresponding hay. The drying time (DT) was recorded from cutting to baling or entrance to the hay barn. WSC content was 187±64.9 and 167±63.7 g kg^{-1} dry matter (DM), and fructans content was 111±47.5 and 89.6±37.9 g kg^{-1} DM, in fresh forage and hay respectively, the decrease being non-significant. WSC and fructans content of hay can be predicted by their content in fresh forage (R^2=0.76 and 0.55; RMSE=32.4 and 26.2 g kg^{-1} DM, respectively). DT was negatively correlated with WSC and fructans content of hay and significantly improved the accuracy of the prediction equations (R^2=0.89 and 0.76; RMSE=21.5 and 18.7 g kg^{-1} DM respectively). Finally, soluble carbohydrate content of hay depends mainly on their content in the fresh forage and to a lesser extent on the DT between cutting and hay baling.

Keywords: hay, fresh forage, water soluble carbohydrate, fructans, drying time

Introduction

Total water-soluble carbohydrates (WSC) influence positively grass digestibility (Humphreys, 1989), and thus forage feed value. Fructans constitute the major part of WSC in temperate grasslands (Chatterton *et al.,* 1989). During drying, plants continue to breathe and a part of soluble carbohydrates (McGechan, 1989) are consumed modifying WSC content and their composition (Ould-Ahmed *et al.,* 2015; Peng *et al.,* 2018). To date, no prediction model of WSC content in hay exists. We aimed to predict WSC and fructans contents of hay from their content measured on fresh forage and considering the drying time (DT).

Materials and methods

During 2017, twenty plots of multi-species grasslands, including four sown plots intensively and 16 permanent pasture plots extensively managed, were studied in three semi mountain areas: Friburg foothills (Switzerland; n=4), Jura (France; n=8) and Massif Central (France, n=8). The plots were studied during the 1st vegetation cycle except four of the eight plots in Massif Central that were studied during the 2nd vegetation cycle. For each plot, representative samples of fresh forage were collected at cutting and of the corresponding hay after drying and storage that lasted between 149 and 348 days (261±79.6 days on average). DT was recorded for each plot between cutting and baling for sun dried hays (n=16) or between cutting and the entrance in the barn for barn dried hays (n=4). Dry matter (DM), ash, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), WSC and fructans contents were measured on all fresh forage and hay samples. Correlations between WSC or fructans contents of hay and chemical criteria of fresh forage or DT were calculated using Spearman's correlation. Stepwise linear regression procedure (SAS 5.1) was used to establish prediction equations of WSC and fructans content of hay from chemical constituents of fresh forage and DT.

Results and discussion

On average, the grasslands contained 75.9±10.9% of grass species (between 57.2 and 93.9%), 7.6±9.4% of legumes (0.2-35.8%) and 16.5±9.1% of forbs (2.5-35.0%). The average stage of maturity at cutting was heading (between stem elongation and flowering) and onset of stem elongation for 1st and 2nd vegetation cycle, respectively.

Due to good weather conditions DT between cutting and harvesting hays were short (2.24±0.497 and 1.70±0.320 days for 1st and 2nd vegetation cycle, respectively). The mean values of WSC, fructans and CP content decreased between fresh forage and hay, whereas NDF and ADF content increased (Table 1), in accordance with literature (Peng *et al.,* 2018). The decrease was not significant for WSC and fructans. This may be due to the short DT since it has been shown that WSC and fructans content in grass forage did not decrease during a 24-hour wilting period (Ould-Ahmed *et al.,* 2015). The range of variation of chemical constituents' contents were high, particularly for WSC and fructans contents that varied in a ratio of more than 1 to 3 (Table 1). This high variability may be linked to the different botanical composition, maturity stage and drying conditions between plots (temperature, light) which modify the kinetics of evolution of soluble carbohydrate contents during drying (Ould-Ahmed *et al.,* 2015). Spearman's correlation analysis showed that the WSC and fructans contents of hay were highly negatively correlated with the NDF and ADF contents and positively with the WSC and fructans contents of the corresponding fresh forages (Table 1).

Stepwise linear regression showed that the WSC and fructans contents of hay were best predicted by their contents measured on the corresponding fresh forage (Table 2). Accuracy of the prediction equations was highly improved by the addition of DT (respectively +13.0% and +21.1%) having a negative coefficient, consistently with the negative correlation of DT with these criteria measured on hay (-0.640, $P<0.01$ and 0.713, $P<0.001$ for WSC and fructans respectively). Part of the variability remains unexplained and could come from the storage period or the drying conditions (temperature, light).

Table 1. Chemical composition (in g kg^{-1} DM, except in g kg^{-1} for dry matter) of fresh forages and the corresponding hays from multi-species grassland plots (n=20), and Spearman's correlation coefficients between WSC and fructans contents in hay and chemical constituents of fresh forages.[1]

Variables	Fresh forage (FF)				Hay				WSC of hay	Fructans of hay
	Mean	SD	Min	Max	Mean	SD	Min	Max	Correlation with FF	
DM	241	34.3	179	325	914	2.46	908	918	-0.620**	-0.63**
Ash	80.4	15.5	57.9	112	76.2	8.97	62.2	93.2	0.420†	0.427†
CP	116	19.0	93.8	152	107	17.9	81.2	138	0.469*	0.480*
NDF	541	67.1	411	654	565	70.1	433	678	-0.907***	-0.880***
ADF	282	43.9	209	365	296	43.8	228	376	-0.866***	-0.824***
WSC	187	64.9	100	323	167	63.7	108	308	0.735***	0.710***
Fructans	111	47.5	57.4	227	89.6	37.9	58.7	184	0.690**	0.671**

[1] SD = standard deviation; Min = minimum; Max = maximum. Significance: *** $P<0.001$; ** $P<0.01$; * $P<0.05$; † $P<0.10$.

Table 2. Prediction equations of WSC and fructans contents in hay (H) from WSC and fructans content in the corresponding fresh forage (FF) and DT (n=20).[1]

	Model components	R^2	RMSE
WSC_H (g kg^{-1} DM)	7.8 + 0.854 × WSC_FF (g kg^{-1} DM)	0.756	32.35
	165.2+ 0.701 × WSC_FF (g kg^{-1} DM) - 59.1 × DT [(3)] (days)	0.886	21.52
Fructans_H (g kg^{-1} DM)	23.9 + 0.590 × Fructans_FF (g kg^{-1} DM)	0.547	26.24
	112.8 + 0.501 × Fructans_FF (g kg^{-1} DM) - 37.09 × DT (days)	0.758	18.66

[1] RMSE = root mean square error.

Conclusions

This study demonstrated that soluble carbohydrate content of hay depends mainly on their content in the corresponding fresh forage and to a lesser extent on the drying time between cutting and hay baling. Longer DT will induce higher decrease in soluble carbohydrate in the hay. The effect of storage period and drying conditions should be studied to try to improve these predictions.

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References

Chatterton N.J., Harrison P.A., Bennett J.H. and Asay K.H. (1989) Carbohydrate partitioning in 185 accessions of gramineae grown under warm and cool temperatures. *Journal of Plant Physiology* 134, 169-179.

Humphreys M.O. (1989) Water-soluble carbohydrates in perennial ryegrass breeding. III. Relationships with herbage production, digestibility and crude protein content. *Grass and Forage Science* 44, 423-430.

McGechan M.B. (1989) A review of losses arising during conservation of grass forage: part 1, field losses. *Journal of Agricultural Engineering Research* 44, 1-21.

Ould-Ahmed M., Decau M.-L., Morvan-Bertrand A., Prud'homme M.-P., Lafrenière C. and Drouin P. (2015) Fructan, sucrose and related enzyme activities are preserved in timothy (*Phleum pratense* L.) during wilting. *Grass and Forage Science* 72, 64-79.

Peng K., Huang Q., Xu Z., McAllister T., Acharya S., Mueller-Harvey I., Drake C., Cao J., Huang Y., Sun Y., Wang S. and Wang Y. (2018) Characterization of condensed tannins from purple prairie clover (*Dalea purpurea* vent.) conserved as either freeze-dried forage, sun-cured hay or silage. *Molecules* 23, 586.

Effects of Ca:Mg ratio and pH on soil properties and grass N yield in drained peat soil

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Abstract

In three dairy grasslands on peat, minerals were added to manipulate the soil Ca:Mg ratio with or without effect on pH. The responses of soil properties and grass N yield were measured. $CaCO_3$ application led to higher soil Ca:Mg ratio and pH_{KCl} compared to the untreated control, decreased N_{total} and C_{total}, and increased P availability. Grass N yield increased in the first year by 21 kg N ha^{-1} whereas soil N_{total} decreased by 380 kg N ha^{-1} in the same period. $MgCO_3$ reduced the Ca:Mg ratio, had little influence on soil parameters and no effect on grass N yield. In contrast, $CaSO_4$ and $MgSO_4$ did not influence pH_{KCl} but reduced grass N yield in most cases. We conclude that grass N yield was not linked with changes in Ca:Mg ratio but with soil pH. To avoid potentially large soil losses of C and N, the current agricultural advice on pH management in peat grasslands should be better adapted to local edaphic characteristics.

Keywords: grassland, lime, gypsum, kieserite, soil pH, N_{total}, C_{total}

Introduction

Reducing soil organic matter (SOM) decomposition while maintaining sufficient grass production for dairy farming is a major challenge in drained peat soils. From a large data set in 20 dairy grasslands on peat, including grass N uptake and the natural variation in soil properties, Deru *et al.* (2019) concluded that the Ca:Mg ratio in the topsoil was the best single soil parameter predicting the unfertilized grass N yield (a proxy for soil N supply). Their results suggested that a higher Ca:Mg ratio may increase the uptake of mineralized N by grassland due to improved soil structure, rooting and water availability, in line with the work of Dontsova and Norton (2002), without increasing the N mineralization itself. This raised the question whether the soil Ca:Mg ratio in peat grasslands can be manipulated to influence the grass N uptake without affecting SOM decomposition.

Materials and methods

A study was carried out on three peat grasslands with different initial soil Ca:Mg ratios (6.9, 4.0, 2.9). Four minerals were added to increase or decrease the Ca:Mg ratio: two with an expected effect on pH ($CaCO_3$ and $MgCO_3$) and two without such an effect ($CaSO_4$ and $MgSO_4$). Amounts were based on applications of 2,500 kg Ca ha^{-1} and 760 kg Mg ha^{-1}, resulting in higher CO_3 or SO_4 applications in the treatments with Ca compared to those with Mg. In each grassland, a randomized block experiment with five treatments (including an untreated control) in four blocks was laid out. During the application year (2014), the farmers continued their normal grassland management. In 2015 and 2016, the experiment was not grazed and no fertilizer was applied, but grass was harvested four times per year, including weighing and sampling for dry matter and total N. In February 2015, soil Ca:Mg ratio was measured. In October 2015, soil pH_{KCl}, C_{total}, N_{total} and P availability (P_{AL}) were measured in 0-10 cm. Additional information on methods is given in Deru *et al.* (2021).

Results and discussion

Soil Ca:Mg ratio was influenced by treatments at the three grasslands as expected (increase with added Ca and decrease with added Mg; Table 1). Soil pH_{KCl} was increased by added CO_3 containing minerals, but not by SO_4 containing minerals. C_{total} was reduced by CO_3 (equivalent to a loss of 2.8 Mg C ha^{-1}) but not by SO_4. N_{total} and P_{AL} were influenced only by $CaCO_3$; N_{total} negatively (equivalent to a loss of 380 kg N ha^{-1}), and P_{AL} positively.

Unfertilized grass N yield was increased in the $CaCO_3$ treatment with 21 kg N ha^{-1} compared to the control (yielding 203 kg N ha^{-1}) during the first year following mineral addition, but not in the subsequent year (Figure 1). Thus, the extra grass N yield after liming with $CaCO_3$ was only 6% of the reduction in the previously mentioned soil N stock (N_{total}). The minerals containing SO_4 reduced grass N yield in both years. Regression analysis showed no correlation between Ca:Mg ratio and unfertilized grass N yield, but grass N yield was positively correlated with pH.

Conclusions

Addition of Ca- and Mg-containing minerals in peat grasslands influenced the soil properties especially for $CaCO_3$, the treatment with the highest CO_3 input. Contrary to our hypothesis, grass N yield was primarily linked with changes in soil pH and not with changes in Ca:Mg ratio. Grass N yield increased (+21 kg N ha^{-1}) one year after applying the $CaCO_3$ mineral, but the coinciding strong decrease in the soil N stock (-380 kg N ha^{-1}) indicated low utilization of the (extra) mineralized N and a disproportional environmental risk of increasing the pH of peat soils. The results of our experiment do not support Ca:Mg ratio as a useful measure of soil quality for increased herbage production in peat grasslands without extra losses of C and N. To avoid those losses, the agricultural pH advice for peat grasslands should be better adapted to the local soil properties that influence SOM decomposition, such as initial pH and P availability. Moreover, advice should be specific in terms of the type and quantity of mineral to be used, based on the expected effect on pH and SOM mineralization.

Table 1. Treatment effects on soil chemical properties. Means with the same superscript are not significantly different (α=0.05).

Parameter	*P*-value	Control	$CaCO_3$	$CaSO_4$	$MgCO_3$	$MgSO_4$
Ca:Mg ratio	<0.001	4.6^c	6.4^d	6.3^d	3.8^b	2.5^a
pH_{KCl}	<0.001	4.7^a	6.1^c	4.7^a	5.0^b	4.8^a
C_{total} (g 100 g^{-1})	0.008	20.9^b	20.5^a	21.1^b	20.4^a	21.1^b
N_{total} (g 100 g^{-1})	0.006	1.70^b	1.63^a	1.68^b	1.67^b	1.69^b
P_{AL} (mg P_2O_5 100 g^{-1})	0.002	45.1^a	50.0^b	43.6^a	46.0^a	45.1^a

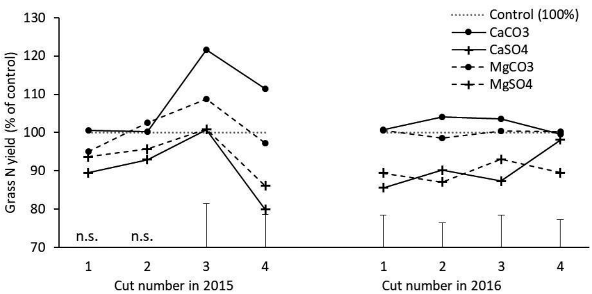

Figure 1. Unfertilized grass N yield (kg N ha^{-1}) per cut in 2015 and 2016, expressed as a percentage of the control plots. Vertical bars on the x-axis represent the LSD per cut in case of significant ($P \leq 0.05$) treatment effect. Mean values of the control plots for each consecutive cut are 72, 44, 62 and 40 kg N ha^{-1} in 2015 and 76, 57, 45 and 43 kg N ha^{-1} in 2016.

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References

Deru J.G.C., Bloem J., de Goede R., Hoekstra N., Keidel H., Kloen H., Nierop A., Rutgers M., Schouten T., van den Akker J., *et al.* (2019) Predicting soil N supply and yield parameters in peat grasslands. *Applied Soil Ecology* 134, 77-84. https://doi.org/10.1016/j.apsoil.2018.10.018

Deru J.G.C., Hoekstra N., van Agtmaal M., Bloem J., de Goede R., Brussaard L. and van Eekeren N. (2021) Effects of Ca:Mg ratio and pH on soil chemical, physical and microbiological properties and grass N yield in drained peat soil. *New Zealand Journal of Agricultural Research*. https://doi.org/10.1080/00288233.2021.1990087

Dontsova K.M. and Norton L.D. (2002) Clay dispersion, infiltration, and erosion as influenced by exchangeable Ca and Mg. *Soil Science* 167(3), 184-193. https://doi.org/10.1097/00010694-200203000-00003

Cattle slurry degradability influences soil organic carbon stock dynamics

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Abstract

In recent years, farmers have increasingly been using organic fertilizers instead of mineral fertilizers for forage production. Organic fertilizers can contribute additional inputs of carbon to the soil. The objective of this work was to measure the capacity of exogenous organic matter (EOM) to increase the rate of soil organic carbon (SOC) over time. To this end, the percentage of decomposable EOM and resistant EOM (DEOM and REOM) of different cattle slurries was analysed. RothC model was run considering experimentally obtained DEOM and REOM values. Results showed that the degradability of slurry could influence SOC dynamics. SOC after 36 years of simulation differed by 10 Mg C ha^{-1} between slurries. In conclusion, amendment degradability of EOM can affect the rate of SOC stock over time.

Keywords: grassland, RothC, soil organic carbon, slurry, DEOM, REOM

Introduction

Organic fertilizers have been identified as a strategy to mitigate climate change (Matsuura *et al.*, 2021). Organic manures that originate in livestock systems are highly variable in their composition and manure handling methods. This variability can affect their decomposability and soil organic carbon (SOC) dynamics over the years. A complex matrix of agents is involved in the transformation of exogenous organic matter (EOM) in manure into SOC (Franzluebbers, 2005). Lashermes *et al.* (2009) developed a method to estimate the potential rate of incorporated carbon of EOM on soil (I_{ROC}). Peltre *et al.* (2012) showed that I_{ROC} could be related to the degradability of farmyard manure in the Rothamsted Carbon model (RothC, Microsoft Excel version). In the study presented here, we simulate how decomposable EOM (DEOM) and resistant EOM (REOM) of cattle slurry added to the soil could determine the rate at which C can be stored in soils.

Materials and methods

Degradability of amendments was determined by incubating soils mixed with EOM following Lashermes *et al.* (2009). Soil used for the incubation experiments was sampled from permanent grassland at 30 cm depth. Soil pH was 6.25, with a loam texture (25% clay), 2.2% total C and 0.17% total N content. Fresh soil was sieved through a 2 mm grid. Slurries from four different cattle farms were used for the incubation experiments. Slurries differed in physicochemical properties (Table 1), probably due to differences in digestibility, slurry management, cleaning practices, etc. To characterize exogenous organic matters (EOM), they were fractionated into soluble, hemicellulose-, cellulose- and lignin-like fractions (SOL, HEM, CEL and LIC, respectively) using the Van Soest method (Van Soest and Wine, 1967). Mixtures of soil and EOM were made by adding the same content of C in all treatments (0.2 g C kg^{-1}) to study C mineralization of EOMs. Three replicated samples were measured after 3 days of incubation in sealed jars at 28 °C (C_{3d}). The C-CO_2 was trapped in 10 ml of 0.5 M NaOH and determined by colourimetry (AFNOR, 2018). The percentage of organic carbon in samples was determined by dry combustion using an elemental analyser (LECO TruSpec® CHN-S (AE:1457)). The SOL, CEL and LIC fractions and the C_{3d} were used to calculate which portion of the amendment would be stored as SOC along the time, express as I_{ROC} indicator (Lashermes *et al.*, 2009):

$$I_{ROC} = 44.5 + 0.5\ SOL + 0.2\ CEL + 0.7\ LIC - 2.3\ C_{3d} \quad (1)$$

The RothC model was used to simulate SOC dynamics. It is partitioned into five basic compartments: inert organic matter (IOM), decomposable plant material (DPM), resistant plant material (RPM), microbial biomass (BIO) and humified organic matter (HUM) (Coleman and Jenkinson, 1996). Decomposable and resistant materials of farmyard manure (FYM) are referred to as DEOM and REOM *per* Mondini *et al.* (2018). Thus, I_{ROC} was converted to DEOM and REOM following Peltre *et al.* (2012), which were introduced in RothC. The results were compared with the observed calibrated data of previous studies for grassland soils with default FYM values using a coefficient of determination (R^2=0.7562, data not shown).

Results and discussion

Results showed significant differences in degradability between cattle slurries. A reference simulation was included, resulting from RothC calculations using default values for FYM (49% of DEOM and 49% of REOM). REOM fraction, which is susceptible to transformation to SOC, was in the range of 32-43%, being significantly higher in Slurries 1 and 2 than slurries 3 and 4 (Table 1).

Figure 1 shows model outputs resulting from default and experimental degradability parameters used for slurries from 1984 to 2020. Slurries 1 and 2 presented higher REOM values than slurries 3 and 4, which contributed to increase SOC stock along the time in these treatments. Differences captured by the model with respect to default value were observed from 1995. This deviation increased continuously, reaching 6.05 Mg C ha^{-1} by 2020. Simulations with slurries 3 and 4 showed a lower SOC stock content than the reference. Additionally, the difference of SOC in relation to standard simulation was constant from 1988, with an average of 2.23 Mg C ha^{-1}. As expected, slurries with more resistant organic matter content contributed to increased SOC stocks (Zimmermann *et al.*, 2007). On the other hand, HUM fraction represents an important part of the SOC stock (Xu *et al.*, 2011). In fact, in RothC model, the RPM fraction decomposes faster (0.3 $year^{-1}$) than the HUM fraction (0.02 $year^{-1}$) (Coleman and Jenkinson, 1996). Thus, even if slurries 1 and 2 showed lower REOM values than those used for FYM in RothC, SOC stock calculations were offset by a significantly higher HUM fraction. As a result, SOC after 36 years of simulation differed in 10 Mg C ha^{-1} between slurries.

Conclusions

In conclusion, cattle slurries could present differences in their decomposability that affect SOC stock evolution. This is important to take into account for degradability parameters in order to produce better estimates of SOC stock evolution.

Table 1. Characterization of decomposable exogenous organic matter (DEOM) and resistant exogenous organic matter (REOM) of slurries and reference value of FYM in RothC.[1]

Treatment	DM (%)	TOC (% DM)	DEOM (% TOC)	REOM (% TOC)	HUM (% TOC)
RothC			49^a	49^a	2
Slurry 1	7.0^a	41.8^a	50^a	43^b	7
Slurry 2	11.5^b	51.1^b	50^a	42^b	8
Slurry 3	8.9^c	46.9^c	63^b	33^c	4
Slurry 4	9.2^c	46.0^c	63^b	32^c	5

[1] Dry matter (DM), total organic carbon (TOC) and compartment of humified organic matter (HUM). Newman-Keuls tests was done for multiple comparisons, same letter represent that there is no significant differences between treatments (*P*=0.05).

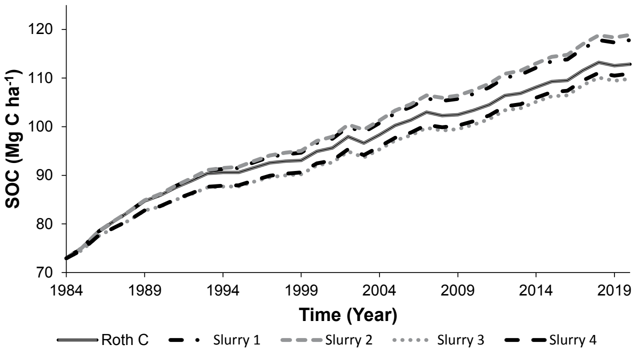

Figure 1. SOC evolution during 1983-2020 simulation of grassland soil. Different lines were simulated treatments with different DEOM and REOM.

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References

AFNOR (2018) FD U44-163. Amendements organiques et supports de culture – Caractérisation de la matière organique par la minéralisation potentielle du carbone et de l'azote. AFNOR, Paris, 33 pp.

Coleman K. and Jenkinson D.S. (1996) RothC-26.3 - A model for the turnover of carbon in soil. In: Powlson D.S., Smith P. and Smith J.U. (eds.) *Evaluation of Soil Organic Matter Models*, NATO ASI Series (Series I: Global Environmental Change), Springer, Berlin, Germany, pp. 237-246.

Franzluebbers A.J. (2005) Organic residues, Decomposition. In: Hillel D. (ed.) *Encyclopedia of Soils in the Environment*, Elsevier, Columbia University, New York, NY, USA, pp. 112-118.

Lashermes G., Nicolardot B., Parnaudeau V., Thuriès L., Chaussod R., Guillotin M. L., Linères M., Mary B., Metzger L., Morvan T., Tricaud A., Villette C. and Houot S. (2009) Indicator of potential residual carbon in soils after exogenous organic matter application. *European Journal of Soil Science* 60, 297-310.

Matsuura S., Kazama R., Hibino H., Funatsu M. and Hojito M. (2021) Manure application in managed grasslands can contribute to soil organic carbon sequestration: evidence from field experiments across Japan. *Regional Environmental Change* 21, 76.

Peltre C., Christensen B.T., Dragon S., Icard C., Kätterer T. and Houot S. (2012) RothC simulation of carbon accumulation in soil after repeated application of widely different organic amendments. *Soil Biology & Biochemistry* 52, 49-60.

Van Soest P.J. and Wine R.H. (1967) Use of detergents in the analysis of fibrous feeds. IV. Determination of plant cell-wall constituents. *Journal of Association of Official Analytical Chemists* 50, 50-55.

Xu X., Liu W. and Kiely G. (2011) Modeling the change in soil organic carbon of grassland in response to climate change: Effects of measured versus modelled carbon pools for initializing the Rothamsted Carbon model. *Agriculture, Ecosystems & Environment* 140, 372-381.

Zimmermann M., Leifeld J., Schmidt M.W.I., Smith P. and Fuhrer J. (2007) Measured soil organic matter fractions can be related to pools in the RothC model. *European Journal of Soil Science* 58, 658-667.

Pre-grazing herbage mass and post-grazing sward height: grass production and quality

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Abstract

Herbage production influences profitability in grass-based ruminant systems. This study aimed to determine the effects of pre-grazing herbage mass (PGHM: 1,500 or 2,500 kg DM ha^{-1}) and post-grazing sward height (PGSH: 4 or 6 cm) on herbage production and nutritive quality. Charolais steers rotationally grazed *Lolium perenne*-dominant swards (13.7 ha) for 222 days and herbage production was measured via pre-grazing herbage mass (lawnmower cuts) and post-grazing sward height (platemeter) on the grazing area. Concurrently, on a separate area, herbage production was measured for the same treatments on cutting plots (5×2 m) (lawnmower; not grazed). There was no PGHM × PGSH interaction for herbage production. Grazing rotation cycle length and herbage production (kg DM ha^{-1}) were greater for PGHM-2,500 than PGHM-1,500 ($P<0.001$), and for PGSH-4 than PGSH-6 ($P<0.01$). Herbage production differences within PGHM and PGSH treatments were proportionately greater on the cutting plots (0.17 and 0.12, respectively) than the grazing area (0.08 and 0.05, respectively). On the grazing area, herbage organic matter digestibility was greater for PGHM-1,500 than PGHM-2,500 and did not differ between PGSH treatments. In conclusion, grazing a higher pasture mass and a lower sward residual increased herbage production.

Keywords: grazing, herbage accumulation, herbage nutritive quality, pasture growth, regrowth interval, steers

Introduction

Increased herbage production and its nutritive quality can positively increase stocking capacity and animal live-weight gain respectively. Increasing regrowth interval (pre-grazing herbage mass, PGHM) can increase herbage production and subsequently reduce fertilization costs (O'Riordan, 1997), but can also reduce herbage nutritive quality, potentially reducing animal performance (McEvoy *et al.*, 2009). Many studies have independently investigated the effect of either PGHM (McEvoy *et al.*, 2009) or post-grazing sward height (PGSH) (Doyle *et al.*, 2021a) on herbage production and nutritive quality but few experiments have examined the combined effect of PGHM and PGSH on these respective variables. The objective of this study was to investigate the effect of PGHM and PGSH on herbage production and its nutritive quality in a rotational stocking system.

Materials and methods

The grazing area comprised five adjacent land blocks totalling sixty permanent paddocks. Within land block paddocks were assigned to one of twelve grazing area farmlets, balanced for initial herbage supply. Each paddock was further sub-divided into three sub-paddocks. Farmlet (1.14 ha each) was randomly assigned to a two (PGHM: 1,500 or 2,500 kg dry matter (DM) ha^{-1} above 4 cm) × two (PGSH: 4 or 6 cm compressed height) factorial arrangement of treatments. Twelve grazing groups of Charolais steers were randomly assigned to farmlet and rotationally grazed *Lolium perenne*-dominant swards for 222 days. The difference between pre-grazing (lawnmower cuts at 4 cm; 5×0.53 m cutting strip) and post-grazing (estimated via rising plate meter; (cm × 242) – 804) herbage mass were recorded at each rotation and were summed to calculate annual herbage production, as detailed in Doyle *et al.* (2021a). Herbage in excess of grazing requirements was removed as silage and its yield determined. Herbage production for each sub-paddock was further sub-divided into what was grazed, removed as silage, or herbage remaining as a 'closing cover' at the end of the season. Herbage samples were obtained from every pre-grazing cut (above the grazing horizon) for each grazing group and herbage organic matter digestibility (OMD) and chemical composition was determined as described by Doyle *et al.* (2021a). Concurrently, in a separate area, herbage production was measured for the same treatments on designated cutting plots (5×2 m) (not grazed). The grazing treatments were replicated four times in a fully randomized complete block design. Herbage was cut (lawnmower) to the assigned PGSH (4 or 6 cm) when PGHM was equivalent to the grazing study, and the quantity of herbage DM removed at each cut was recorded and summed to calculate annual herbage production.

Herbage growth and nutritive data were statistically analysed using the MIXED procedure of SAS where the experimental unit was sub-paddock (for herbage growth on the grazing area), cut plot, or grazing group (herbage nutritive value on the grazing area), as appropriate. The model contained fixed effects for PGHM, PGSH and their interactions. Data averaged per sub-paddock were weighted for frequency of grazing (i.e. the number of times the sub-paddock was defoliated). Differences between means were tested for significance using the PDIFF statement and adjusted by Tukey, as appropriate.

Results and discussion

On the grazing area mean PGHM was 1,687 and 2,683 kg DM ha^{-1} (P<0.001) and mean PGSH was 4.1 and 6.0 cm (P<0.001). There were no PGHM × PGSH interactions for any herbage production variables (Table 1). Grazing rotation cycle length (days) and herbage production (kg DM ha^{-1}) were greater for PGHM-2,500 than PGHM-1,500 (P<0.001) and for PGSH-4 than PGSH-6 (P<0.01). Herbage production differences within PGHM and PGSH treatments were proportionately greater on the cut plots (0.17 and 0.12, respectively) than the grazing area (0.08 and 0.05, respectively). It is hypothesized that these differences are due to the prolonged grazing residency time for PGHM-2,500 than PGHM-1,500 (3.8 vs 2.2 days; P<0.001) and PGSH-4 than PGHS-6 (3.6 vs 2.4 days; P<0.001), which is suggested to reduce herbage regrowth due to the depletion of water-soluble carbohydrate reserves under prolonged grazing (Fulkerson and Donaghy, 2001). On the grazing area, the quantity of herbage consumed ha^{-1} did not differ within PGHM and PGSH treatments. The quantity of excess herbage removed ha^{-1} as silage did not differ between PGHM treatments, but was greater (P<0.05) for PGSH-4 than PGSH-6. Mean closing farmlet pasture supply was greater for PGHM-2,500 than PGHM-1,500 (943 vs 716 kg DM ha^{-1}, respectively) and for PGSH-4 than PGSH-6 (929 vs 731 kg DM ha^{-1}, respectively). This additional herbage can be used to increase the length of the grazing season or herbage availability the following spring.

There were PGHM × PGSH interactions for herbage neutral detergent fibre (NDF) (P<0.05) and crude protein (CP) concentration (Table 1). For NDF concentration, 2,500-4 was greater than 1,500-4, but 1,500-6 and 2,500-6 did not differ. For CP concentration, 2,500-6 was greater than 2,500-4, but 1,500-4 and 1,500-6 did not differ. Herbage OMD was lower (P<0.001) for PGHM-2,500 than PGHM-1,500; consequently, PGHM-2,500 reduced animal live-weight gain at pasture (-0.06 kg day^{-1}) (Doyle *et al.*, 2021b). Herbage OMD did not differ significantly between PGSH treatments, which is similar to Doyle *et al.* (2021b). Despite this, PGSH-6 had a greater live-weight gain at pasture (+0.16 kg day^{-1}) due to a greater dry matter intake (+0.75 kg DM day^{-1}) (Doyle *et al.*, 2021).

Conclusions

Grazing a higher pasture mass and a lower sward residual increased herbage production; differences between treatments were greater on mechanically cut rather than grazed plots.

Table 1. Effect of pre-grazing herbage mass (PGHM – 1,500 or 2,500 kg dry matter (DM) ha^{-1}) and post-grazing sward height (PGSH – 4 or 6 cm) on herbage production and chemical composition on the grazing area.

Variable	PGHM 1,500		PGHM 2,500		SEM	Significance		
	PGSH 4	PGSH 6	PGSH 4	PGSH 6		PGHM	PGSH	PGHM×PGSH
Grazing area								
Number of grazing rotation cycles	6.2	7.6	3.8	4.7	0.11	***	***	NS
Regrowth interval (days)	31.7	26.5	52.1	44.8	0.84	***	***	NS
Average growth rate[2]	49.3	45.8	55.2	53.3	1.07	***	*	NS
Total herbage production	11,506	10,688	12,095	11,861	179.8	***	**	NS
of which grazed	8,421	8,254	8,751	8,748	461.4	NS	NS	NS
of which removed as silage	2,633	1,608	2,487	1,926	338.0	NS	*	NS
Cutting plots								
Number of rotation cycles	7	8	5	6	.	.	.	.
Regrowth interval (days)	28	24	42	36	.	.	.	.
Average growth rate[2]	51.8	46.8	63.4	55.7	1.31	***	***	NS
Herbage production (kg DM ha^{-1})	12,228	10,672	14,148	12,643	299.8	***	***	NS
Herbage nutritive value								
OMD (g kg^{-1})	801^{a}	793^{a}	765^{b}	$782^{a,b}$	5.9	**	NS	0.06
CP (g kg^{-1} DM)	182^{a}	177^{a}	150^{c}	159^{b}	2.1	***	NS	**
NDF (g kg^{-1} DM)	400^{c}	$405^{b,c}$	438^{a}	$422^{a,b}$	4.6	***	NS	*
ADF (g kg^{-1} DM)	226^{b}	225^{b}	$240^{a,b}$	253^{a}	3.8	***	NS	0.07

[1] The pasture supply remaining in the pasture at the end of the year.

[2] Expressed in kg DM ha^{-1} day^{-1}; OMD = *in vitro* organic matter digestibility, CP = crude protein, NDF = neutral detergent fibre, ADF = acid detergent fibre, WSC = water soluble carbohydrates; Ash = crude ash; SEM = standard error of the mean for PGHM × PGSH; means within a row with different superscript letters differ (P<0.05); NS = non-significant, * P<0.05, ** P<0.01, *** P<0.001.

References

Doyle P.R., McGee M., Moloney A.P., Kelly A.K. and O'Riordan E.G. (2021a) Effect of post-grazing sward height, sire genotype and indoor finishing diet on steer intake, growth and production in grass-based suckler weanling-to-beef systems. *Animals* 11, 2623.

Doyle P.R., McGee M., Moloney A.P., Kelly A.K. and O'Riordan E.G. (2021b) Effect of pre-grazing herbage mass, post-grazing sward height and finishing diet on performance of late-maturing beef steers. *Journal of Animal Science* 99 (Supp 3), 296.

Fulkerson W.J. and Donaghy D.J. (2001) Plant-soluble carbohydrate reserves and senescence-key criteria for developing an effective grazing management system for ryegrass-based pastures: a review. *Australian Journal of Experimental Agriculture*, 41(2), 261-275.

McEvoy M., O'Donovan M., Kennedy E., Murphy J.P., Delaby L. and Boland T.M. (2009) Effect of pregrazing herbage mass and pasture allowance on the lactation performance of Holstein-Friesian dairy cows. *Journal of Dairy Science* 92, 414-422.

O'Riordan E.G. (1997) Effect of nitrogen application rate and grass regrowth interval on annual herbage production. *Agricultural Research Forum*, 133-134.

Vertical distribution of herbage chemical composition: effect of pre- and post- grazing sward height

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Abstract

This study aimed to determine the effects of pre-grazing herbage mass (PGHM: 1,500 or 2,500 kg DM ha^{-1}) and post-grazing sward height (PGSH: 4 or 6 cm) on the vertical distribution of the sward chemical composition and digestibility in *Lolium perenne*-dominant swards rotationally grazed from mid-March to late-October. Pre-grazing herbage samples were taken from ground level in May, June and September and cut into layers, one below the grazing horizon (i.e. below 4 or 6 cm) and subsequent 4 cm layers above the grazing horizon. Organic matter digestibility and crude protein concentration decreased and neutral detergent fibre, acid detergent fibre and ash concentrations increased from the top to the base of the plant at each sampling month ($P<0.001$). There was a PGSH × layer interaction for acid detergent fibre (ADF) concentration in September, whereby PGSH-6 had a higher ADF concentration than PGSH-4 below the grazing horizon, but concentration did not differ between PGSH treatments above the grazing horizon. In conclusion, chemical composition differed between layers. For a given chemical constituent, the ranking between layers was unaffected by season, PGHM and PGSH.

Keywords: grazing horizon, nutritive value, perennial ryegrass, regrowth interval, sward horizons, organic matter digestibility

Introduction

Herbage defoliation in rotational and strip-grazing systems occurs through removal of successive layers of the sward (Wade, 1991). As the chemical composition of each layer in the sward can change throughout the year due to grazing management (Delagarde *et al.*, 2000), this has a key impact on nutrient intake of the grazing animal. The effect of pre-grazing herbage mass (PGHM) on the vertical distribution of sward chemical composition is well documented (Delagarde *et al.*, 2000); however, few studies have investigated the interactive effect of PGHM and post-grazing sward height (PGSH) on the vertical distribution of sward chemical composition and digestibility throughout the grazing season. Therefore, the objective of this experiment was to investigate the effect of PGHM and PGSH on the vertical distribution of the sward chemical composition, and how it changes during the grazing season.

Materials and methods

Lolium perenne-dominant paddocks were assigned to a two (PGHM >4 cm: 1,500 or 2,500 kg DM ha^{-1}) × two (PGSH: 4 or 6 cm compressed height) factorial arrangement of treatments, which were rotationally stocked by 3 replicated grazing groups per treatment of suckler-bred Charolais steers from 21 March to 29 October (222 days). Paddocks received 150 kg chemical nitrogen ha^{-1}. Pre-grazing herbage samples were cut from ground level using a scalpel at 15 random areas throughout a paddock for each grazing group in May (vegetative stage), June (reproductive stage) and September (post-reproductive stage). Samples were composited in the laboratory while still maintaining their straight vertical distribution. A 500 g sub-sample was placed under a guillotine blade and cut from ground level to the grazing horizon (4 or 6 cm) and cut into 4 cm layers above the grazing horizon until the top of the canopy was reached; layer 1 represented the bottom of the plant. Accordingly, PGSH-6 was cut into layers of 0-6 cm (layer 1), 6-10 cm (layer 2), 10-14 cm (layer 3), 14-18 cm (layer 4), 18-22 cm (layer 5) etc. Each layer was dried at 40 °C to a constant weight. The first five layers (from ground level) were individually ground, and the remaining layers were grouped together and ground (Wiley mill; 1 mm aperture; Arthur H. Thomas, Philadelphia, PA, USA) in preparation for chemical analysis as described by Doyle *et al.* (2021). Data were statistically analysed for each of the three-monthly measurement periods using the MIXED procedure of SAS, where the experimental unit was grazing group. The model contained fixed effects for PGHM, PGSH, sward horizon layer and their interactions. Differences between means were tested for significance using the PDIFF statement and adjusted by Tukey, as appropriate.

Results and discussion

There were PGHM × PGSH interactions for herbage CP concentration in May (P<0.05), and NDF and ADF concentrations in June (P<0.01) and September (P<0.001) (Table 1). There were no PGHM × PGSH × layer or PGHM × layer interactions. However, there was very large variation in chemical composition form the top to the base of the sward. Overall, organic matter digestibility (OMD) and crude protein (CP) concentrations decreased (P<0.001) and neutral detergent fibre (NDF), acid detergent fibre (ADF) and crude ash concentrations increased (P<0.001) from the top (layer 6) to the base (layer 1) of the sward for each sampling month (Table 1), as observed previously, and likely reflects the proportion of leaf, stem and dead tissues in these layers (Delagarde *et al.*, 2000). There was a PGSH × layer interaction for ADF concentration in September, whereby PGSH-6 had a greater (P<0.05) ADF concentration than PGSH-4 below the grazing horizon (layer 1), but did not differ above the grazing horizon (layer 2 to 6). This implies that fresh herbage regrows from the defoliation point and older dead herbage had accumulated to the base of the sward. In practical terms this means a PGSH of 6 vs 4 cm does not negatively impact the nutrient value of consumed herbage, which agrees with Doyle *et al.* (2021).

Table 1. Effect of pre-grazing herbage mass (1,500 and 2,500 kg dry matter (DM) ha^{-1}) and post-grazing sward height (4 or 6 cm) on the sward vertical distribution of chemical composition and *in vitro* digestibility in May, June and September.[1]

	Grazing treatment (PHM-PGSH)					Layer							Significance			
	1,500-4	1,500-6	2,500-4	2,500-6	SEM	1	2	3	4	5	6	SEM	PGHM	PGSH	Layer	PGHM*PGSH
May																
OMD (g kg^{-1})	809	794	810	804	6.0	691	792	827	838	833	843	7.4	NS	NS	***	NS
CP (g kg^{-1} OM)	146^{a}	160^{a}	125^{b}	117^{b}	5.3	103	110	127	146	160	174	6.5	***	NS	***	*
NDF (g kg^{-1} OM)	482	501	502	498	8.8	575	512	482	473	476	457	9.5	NS	NS	***	NS
ADF (g kg^{-1} OM)	236	253	257	268	4.2	317	275	255	242	231	200	5.1	***	**	***	NS
Ash (g kg^{-1} OM)	102	102	97	102	5.1	171	100	89	88	82	75	6.2	NS	NS	***	NS
June																
OMD (g kg^{-1})	784	771	762	768	6.5	669	757	778	800	809	812	7.9	0.06	NS	***	NS
CP (g kg^{-1} OM)	193	192	131	145	5.2	119	133	166	188	195	189	6.4	***	NS	***	NS
NDF (g kg^{-1} OM)	466^{a}	565^{b}	558^{b}	583^{b}	12.9	628	595	555	530	493	458	15.8	***	***	***	**
ADF (g kg^{-1} OM)	265^{a}	309^{b}	314^{bc}	326^{c}	3.8	359	336	314	291	271	250	5.7	***	***	***	***
Ash (g kg^{-1} OM)	113	120	102	101	5.2	145	104	104	107	98	96	6.3	**	NS	***	NS
September																
OMD (g kg^{-1})	771	767	750	766	6.1	670	749	781	791	794	797	7.5	0.09	NS	***	NS
CP (g kg^{-1} OM)	162	174	162	165	5.2	121	129	155	180	198	212	6.6	NS	NS	***	NS
NDF (g kg^{-1} OM)	469^{a}	499^{b}	554^{d}	527^{c}	6.0	556	534	510	498	495	480	7.4	***	NS	***	***
ADF (g kg^{-1} OM)[2]	264^{a}	287^{b}	319^{c}	289^{b}	4.0	308	305	297	297	272	260	5.0	***	NS	***	***
Ash (g kg^{-1} OM)	125	115	113	111	5.2	184	116	103	101	97	93	6.4	NS	NS	***	NS

[1] OMD = *in vitro* organic matter digestibility, CP = crude protein, NDF = neutral detergent fibre, ADF = acid detergent fibre, WSC = water soluble carbohydrates, Ash = crude ash; Layer 1 = bottom of the plant, layer 6 = top of the plant, SEM = standard error of the mean. Means within a row with different superscript letters differ (P<0.05); NS = non-significant, * P<0.05, ** P<0.01, ***P<0.001.

[2] PGSH × layer interaction (P<0.05): values of 298 vs 319, 303 vs 306, 298 vs 296, 308 vs 286, 280 vs 265 and 264 vs 257 for PGSH-4 vs PGSH-6 in layers 1, 2, 3, 4, 5 and 6, respectively.

Conclusions

Chemical constituents differed between layers. For a given chemical constituent, the ranking between layers was unaffected by season, PGHM and PGSH. However, the ADF differences between PGSH treatments did differ between horizon layers towards the end of the grazing season.

References

Delagarde R., Peyraud J.L., Delaby L. and Faverdin P. (2000) Vertical distribution of biomass, chemical composition and pepsin-cellulase digestibility in a perennial ryegrass sward: interaction with month of year, regrowth age and time of day. *Animal Feed Science and Technology* 84, 49-68.

Doyle P., McGee M., Moloney A.P., Kelly A.K. and O'Riordan E.G. (2021) Effect of post-grazing sward height, sire genotype and indoor finishing diet on steer intake, growth and production in grass-based suckler weanling-to-beef systems. *Animals* 11, 2623.

Wade M.H. (1991) Factors affecting the availability of vegetative *Lolium perenne* to grazing dairy cows with special reference to sward characteristics, stocking rate and grazing method. Doctoral dissertation, Rennes, France.

Economic loss of the provisioning service in uplands due to disruption of traditional management

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Abstract

Profound socioeconomic changes in mountain areas, as evidenced by depopulation, rural abandonment or modernization of farms, have promoted a decline in the number of domestic herbivores in upland areas whose sustainable management promotes floristic communities of high diversity. In the western Pyrenees, the maintenance of open land has been complemented by the use of fire through pastoral burnings. However, the decrease of biomass consumption by hervibores encourages a more frequent use of fire in herbaceous areas where otherwise necromass is accumulating. These management regimes favour the spread of the native tall-grass *Brachypodium rupestre*. The expansion of this unpalatable grass generates degraded and low-diversity grasslands, decreasing the provisioning ecosystem service. This research economically quantified the loss of the provisioning service of high-quality food for livestock, implementing the substitution economic approach based on contrasted floristic inventories (high- vs low-diversity grasslands) and detailed information to determine the number of feed rations lost in degraded areas. Economic valuation of natural resources may be helpful in raising awareness among stakeholders and to encourage environmental policies that prevent grassland degradation.

Keywords: high-mountain grasslands, ecosystem services, *Brachypodium rupestre*

Introduction

Many grasslands in Europe are located in high-mountain areas and are characterized by seasonal production concentrated in summer under an extensive farming system. Humans have managed these grasslands with the main aim of providing food for domestic herbivores, but there are other goods that grasslands provide, encompassed within the ecosystem service concept (MEA, 2005). The changes occurring in the primary sector have led to a drastic decrease in the number of grazing animals, favouring the accumulation of (dead) biomass and encouraging farmers to use burning to reduce fuel loads. The subsequent decoupling of traditional fire and grazing regimes has a direct effect on the floristic composition. Specifically in the Aezkoa valley (Spain), a significant loss of floristic diversity has been detected in relation to the expansion of the tall-grass *Brachypodium rupestre* (Múgica *et al.*, 2021).

Materials and methods

Our study was conducted in the Aezkoa valley, located in the western Spanish Pyrenees. This valley encompasses high-value grasslands, which grow between 800-1,400 m a.s.l., and cover 2,147 ha. These natural grasslands are included in the SAC Roncesvalles-Selva de Irati.

For the economic evaluation, we applied the substitution approach. This method provides an economic value based on the replacement cost associated with the environmental resource lost, considering the least costly alternative and requiring a deep knowledge of the evaluated resource. The method includes two types of analysis: replacement cost and resource equivalency. We adopted the latter, which considers economic estimates to compensate farmers for the loss of the environmental resource. The quantity of the lost resource was represented by number of feed rations for livestock (Champ *et al.*, 2017; Chapman *et al.*, 2018). For this purpose, we used field floristic surveys done in high- (HD) and low-diversity (LD) grasslands using the point quadrat method (8 transect lines per plot). The data collected allowed for the estimation of the pastoral forage value (PFV) based on the assignment of an index of specific quality (ISQ) of values between 0 and 5 considering the palatability (Jouglet, 1999) and the specific contribution (SC) of each species present in each inventory: [PFV = 0.2 The PFV is linked to the energy value (EV) by applying a conversion coefficient (k) adjusted by regional specificities of altitude, in Aezkoa k ranging from 50 to 60.

Based on a previous study (Ferrer and Canals, 2008), we created a map combining *B. rupestre* covers and slopes in order to assess the viable surface for grazing, discarding steep slopes (>15°). In turn, intermediate slopes (7-15°) and flat areas (<7°) were assigned to grazing only with sheep or mixed flocks (sheep, horses and cows), respectively. The difference between HD and LD grasslands allows estimation of the energy loss caused by the expansion of *B. rupestre*. The balanced ration of food that animals would receive each day in a hypothetical indoor feeding (150 days with 2019 prices) was designed using the software INRAtion-PrévAlim (INRA, 2017-2019). We used data on the current censuses of animals and their reproductive stage (maintenance, pregnancy, etc.), for estimating the food intake (kg dry matter day^{-1}) and the energy supply (FU day^{-1}) required to fulfil their needs. We complemented the valuation approach by performing a sensitivity analysis, which focused on: (a) the variation in ISQ of *B. rupestre* (from 0 to 1), and (b) the rising of the ration quality (low vs high-cost ration).

Results and discussion

Eight hundred sampling points were inventoried in the study region. The average number of species in HD (*Br* cover <50%) and LD (*Br* cover >50%) grasslands were 22 and 11 respectively, and the Shannon index was 2.51 and 1.55, reflecting the loss of diversity caused by *B. rupestre* spreading. The difference in energy lost between HD and LD grasslands was 413.6 FU ha^{-1} (Figure 1A) and the total degraded area with a potential for grazing was 200.05 ha out of 287.76 ha studied. The total daily cost per ration and the number of lost rations were calculated for sheep and mixed flocks; therefore the total loss of the provisioning value was 21,146 € (106 € ha^{-1} year^{-1}), when the replacement ration was the least costly and the ISQ of *B. rupestre* was 0.5 (Figure 1B). The sensitivity analysis varied between 10,925 €, considering ISQ=1 and the cheapest replacement ration, and 33,399 €, considering ISQ=0 and the more expensive replacement ration (Figure 1C).

The disruption of traditional management regimes also causes damage to the local biodiversity due to the expansion of species with strong competitive ability (clonal reproduction, tall height, clumped density, etc.). The recovery of degraded areas is associated with specific problems (slopes, altitude, accessibility, etc.) and therefore restoration measures are limited. Recurrent prescribed burnings of the steepest areas without subsequent grazing generate controversy. In the particular situation of the loss of floristic diversity in the Aezkoa grasslands, efforts on a local scale should focus on the areas more likely to recover, by supporting regular and guided grazing. Using external inputs instead of grazing may also increase the competition for using lowlands as well as fluctuations of food prices, volatility of international markets, the deterioration of animal health and quality products (Durán *et al.*, 2020).

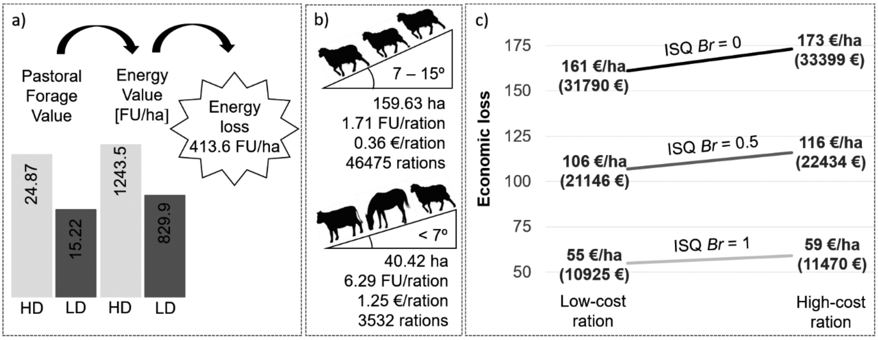

Figure 1. (A) Methodology followed to calculate the loss of energy value between HD and LD grasslands. (B) Results of crossed data of *Brachypodium rupestre* covers and slopes, the energy value and prices of each type of flock and (C) the range of the sensitivity analysis given different *B. rupestre* covers.

Conclusions

In natural grasslands where the loss of floristic diversity is related to a decrease in the palatability of the plant community and its energy value, it is possible to evaluate the loss of the provisioning ecosystem service by applying the economic substitution method, using floristic inventories and estimating the loss of food rations that need to be substituted by external forage. This approach helps to economically evaluate a severe environmental problem and could be an effective tool to raise awareness among stakeholders and the population affected by this change.

References

Champ P.A., Boyle K.J. and Brown T.C. (2017) A primer on nonmarket valuation. *The economics of non-market goods and resources*. Springer, Dordrecht, the Netherlands.

Chapman D., Steen C. and Özdemiroglu E. (2018) *Economic valuation for equivalency analysis. Equivalency methods for environmental liability: Assessing damage and compensation under the European environmental liability directive*. Springer, Heidelberg, Germany.

Durán M., Canals R.M., Sáez J.L., Ferrer V. and Lera-López F. (2020) Disruption of traditional land use regimes causes an economic loss of provisioning services in high-mountain grasslands. *Ecosystem Services* 46, 101200.

Ferrer V. and Canals R.M. (2008) Proyecto de ordenación de los recursos pascícolas forestales del Monte Aezkoa nº 1 del CUP. Consultoría Belardi and Universidad Pública de Navarra, Pamplona, Spain.

INRA (2017-2019) *INRAtion PrévAlim. Logiciel de rationnement pour bovins, ovins et caprins*. ed. INRA, France.

Jouglet J. (1999) *Les végétations des alpages des Alpes Françaises du Sud. Guide technique pour la reconnaissance et la gestion des milieux patures d'altitude*. Cemagref et Aten, France.

MEA (2005) *Ecosystems and human well-being: synthesis*. Island Press, Washington DC, USA.

Múgica L., Canals R.M., San Emeterio L. and Peralta J. (2021) Decoupling of traditional burnings and grazing regimes alters plant diversity and dominant species competition in high-mountain grasslands. *Science of the Total Environment* 790, 147917.

Identifying cropping strategies for sustainable ley farming systems based on legumes

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Abstract

Legume species are recognized as an important component for production of high-protein forage. In Sweden, red clover (RC; *Trifolium pratense* L.) is the main legume species in mixed swards whereas lucerne (LU; *Medicago sativa* L.) has been confined to drier regions with high soil pH. One constraint of RC is the poor persistence originating from root rot caused by several soil-borne pathogens. The objective was to identify strategies for sustainable legume cropping in production cycles. Persistence and production of RC and LU grown in mixed swards with timothy (TI; *Phleum pratense* L.) were compared at one field site where RC and LU were grown for two years, and thereafter re-established with RC or LU as pre-crop; four combinations in total. We present data from three cuts of the first production year of the second cycle (2019). The prevalence of root rot was assessed visually and the abundance of *Fusarium avenaceum*, *Phoma* spp. and *Cylindrocarpon destructans* was estimated with molecular analyses. Red clover showed high disease scores when grown after RC and LU, whereas LU also was infected but at a significantly lower level. In conclusion, LU in mixture with TI can maintain competitive production but propagates pathogenic fungi.

Keywords: disease severity index, lucerne, red clover, root rot

Introduction

Cultivation of domestic protein crops is of great importance for strengthening the competitiveness and improving business management in animal production. Ruminants are able to use the nutrients in forage through symbiosis with microbes in the rumen, hence the forage crop is the main source of protein and fibre supply. However, the weak persistence of RC has for a long-time reduced mixed ley production since the red clover plants have died during the winters of first and second harvest years. Likewise, legume stands are an important part of the nitrogen supply in organic farming, which is based on biological fixation of nitrogen from the air, and which mainly takes place through the symbiotic fixation in the root nodules of the legume plant. The development, persistence and production capacity of the legume plants are therefore crucial for organic farming. The aim of the project was to identify and develop strategies for sustainable production of locally produced protein forage.

Materials and methods

A large-plot field trial, without replicates, was sown on 6 May 2015 at Ullberga farm northeast of Nyköping (N 58° 50', E 16° 53'). The soil characteristics were: 19% clay, 1.8% soil carbon, pH 6.7 and total soil nitrogen (0-30 cm) of 1.35 g kg^{-1}. The average temperature is 7.5 °C on a yearly basis, and average annual precipitation is 674 mm (sv.climate-data.org). Five treatments with different ley species were sown with oats as nurse crop and each treatment represents one crop rotation (Table 1). Two treatments were red clover (RC; *Trifolium pratense* L. cv. SW Vicky) sown in mixture with timothy (TI; *Phleum pratense* L cv. Lischka) and two were (LU; *Medicago sativa* L. cv. Power 4.2) sown in mixture with TI (cv. Lischka). The fifth treatment, farmer's choice (FC), consisted of 20 ley species, included lucerne, birdsfoot trefoil, white clover, timothy, perennial ryegrass, red fescue, meadow fescue and herbs. The seed rate was 8 kg ha^{-1} of RC, 17 kg ha^{-1} of LU and 8 kg ha^{-1} of TI, while the seed rate of the farmer's choice was 23 kg ha^{-1}. The seeds of LU were inoculated with the bacterium *Sinorhizobium meliloti* (Nitragen Gold) and seeds of BFT were inoculated with *Rhizobium loti*. No plant nutrients were added. The nurse crop was oats (180 kg ha^{-1}) and the size of each large plot was 7.2×50 m. After the harvest in 2017, the experimental area was ploughed under, except for the plot with the farmer's choice of ley mixture. New mixed swards with legumes and timothy were sown in August 2017 with winter wheat as nurse crop.

Yield and dry matter content were determined by cutting the above-ground biomass in four 0.5 m^2 squares in each plot at a hight of 5 cm and dried in 48 °C for 48 h. During the first harvest year of the second cycle (2019), ten plants from each plot, including the FC (4^{th} year) were carefully uprooted, washed, and split with a scalpel. External and internal damage (discoloration) was visually assessed according to Rufelt (1986), where 0 is healthy roots and 4 represents all dark roots. External and internal disease severity indices (DSI_E and DSI_I) and the proportion of roots with external and internal infection (disease incidence, DI_E and DI_I) were calculated for each treatment (Almquist *et al.*, 2016).

A pooled sample for each treatment was analysed for the abundance of the three root rot pathogens *Fusarium avenaceum*, *Cylindrocarpon destructans* and *Phoma* spp. (Almquist *et al.*, 2016). The abundance of each pathogen is expressed as the number of gene copies per 10^6 copies of the plant *cox* gene.

Results and discussion

The forage yields of the first harvest year in the second cycle are presented here. First-cut yields were about 5 Mg dry matter (DM) for all treatments but the yield of LU + TI were significantly higher than that of RC + TI in the two other cuts (Table 2). The total yield of LU grown after RC was 12.5 Mg ha^{-1}, the yield of LU grown after LU was 13.6 Mg, the yield of RC grown after RC was 7.4 Mg ha^{-1} and the yield of RC grown after LU was 8.6 Mg ha^{-1}.

Lucerne was affected by root rot to the same extent when sown after both LU and RC (Table 3). Likewise, RC was severely affected by root rot when sown after both LU and RC, showing that the fungi are present in the soil, but LU is more tolerant than RC to the root rot fungi. Lucerne plants assessed from the plot of FC (4^{th} harvest year) was more affected by root rot than the LU plants sown in the second cycle, which highlights the susceptibility of LU to root rot even though it is more tolerant than RC.

The RT-qPCR analyses also showed that there were higher amounts of *Phoma* spp. and smaller amounts of *C. destructans* in LU than in RC, while *F. avenaceum* were only present in the LU roots in the Farmer's choice treatment (Table 3).

Table 1. Experimental setup for the large plot field trial at Ullberga, Nyköping Sweden with five crop rotations established in 2015, with red clover (RC) or lucerne (LU) grown in mixed swards with timothy (TI) as well as farmer's choice (FC) of grass species, legumes, and herbs.[1]

Year	Crop rotation 1	Crop rotation 2	Crop rotation 3	Crop rotation 4	Crop rotation 5
2015	Oats and LU + TI	Oats and LU + TI	Oats and RC + TI	Oats and RC + TI	Oats and seeds of FC
2016	LU + TI HY 1	LU + TI, HY 1	RC + TI, HY 1	RC + TI HY 1	FC, HY 1
2017	LU + TI, HY 2	LU + TI, HY 2	RC + TI, HY 2	RC + TI, HY 2	FC, HY 2
2018	Winter wheat and LU + TI	Winter wheat and RC + TI	Winter wheat and RC + TI	Winter wheat and LU + TI	FC, HY 3
2019	LU + TI, HY 1	RC + TI, HY 1	RC + TI, HY 1	LU + TI, HY 1	FC, HY 4

[1] HY = harvest year.

Table 2. Harvest yields and dry matter content (DM) from the first harvest year of the second cycle, 2019 at the large plot field trial at Ullberga, Nyköping.[1]

	Cut 1 kg DM ha^{-1}	DM Cut 1 %	Cut 2 kg DM ha^{-1}	DM Cut 2 %	Cut 3 kg DM ha^{-1}	DM Cut 3 %	Total yield kg DM ha^{-1}	Average DM %
LU after LU	5,433	22.3a	4,240a	28.4a	2,873a	23.3	12,547a	24.7a
RC after LU	4,867	16.4b	2,273b	25.0b	1,467b	22.7	8,607b	21.4a
RC after RC	4,500	17.0b	1,767b	26.6ab	1,100b	22.7	7,367b	22.1a
LU after RC	5,693	23.1a	4,893a	28.5a	3,047a	24.4	13,633a	25.4a
FC (LU)	4,847	23.1a	4,120a	26.5ab	2,907a	24.5	11,873a	24.7a
P-value	ns	<0.001	<0.001	0.007	<0.001	ns	<0.001	0.044
Coeff.of *variance*	9.8	4.8	17.2	4.6	12.5	5.1	6.8	13.7

[1] Dates of cuts in 2019; cut 1: 8 June, cut 2: 29 July; cut 3: 21 September. RC= red clover, LU= lucerne, FC= Farmer's choice (lucerne). ANOVA-procedure were used for the statistical analyses. Different characters indicate significant differences according to Tukey's HSD test ($P<0.05$).

Table 3. External (E) and internal (I) disease severity index (DSI), disease incidence (DI) and abundance of *Fusarium avenaceum* (F.a.), *Phoma* spp. (Ph.) and *Cylindrocarpon destructans* (C.d.), at the large plot field trial at Ullberga, Nyköping, during the first harvest year of the second cycle, 2019.[1,2]

Treatment	DSI_E	DSI_I	DI_E	DI_I	F.a.	Ph.	C.d.
Lucerne after lucerne	35	15	90	60	0.0	3,674	328
Red clover after lucerne	70	38	100	90	0.0	211	2,480
Red clover after red clover	73	35	100	90	0.0	980	2,278
Lucerne after red clover	35	23	100	80	0.0	451	294
Farmer's choice (lucerne)	53	25	100	80	19.0	10,056	4,389

[1] The crop rotation of farmer's choice was harvested for the fourth year, established in 2015.

[2] The abundance of each pathogen is expressed as the number of gene copies per 10^6 copies of the plant *cox* gene.

Conclusions

Lucerne in mixture with timothy can maintain competitive production during dry weather conditions, whereas yields of red clover in mixture with timothy are lower than that of lucerne. The root rot fungi *Phoma* spp. and *C. destructans* occur in the roots of both examined legume species, but lucerne seem to be more tolerant to developing disease symptoms. However, lucerne maintains the pathogen inocula in the soil for the next forage crop. Lucerne is an underestimated choice of legume forage species to include in sustainable ley production in Sweden.

References

Almquist C., Stoltz E., Wallenhammar A.C. (2016) Incidence of root pathogens associated to clover root rot in Sweden. *Grassland Science in Europe* 21, 786-788.

Rufelt S. (1986) Studies on *Fusarium* root rot of red clover (*Trifolium pratense* L.) and the potential for its control. Doctoral thesis. Swedish University of Agricultural Sciences. Department of Plant and Forest Protection, Uppsala, Sweden. 33 pp.

Fertilization of grass-clover leys with mineral N and slurry: effect on clover dynamics, N_2-fixation and nitrate leaching

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Abstract

The aim of this study was to examine the N response over two years of slurry and mineral N fertilization on clover dynamics, the quantitative N_2-fixation (qBNF) using the ^{15}N isotope dilution method, and subsequent effect on nitrate leaching. Two- and four-species mixtures of grass-clover received increasing rates of N fertilizer (0-480 kg available N ha^{-1}) either of mineral N only or combined with application of cattle slurry. Fertilization significantly reduced clover proportion and N_2-fixation. The qBNF representing an input of 193-286 kg N ha^{-1} in the unfertilized treatment declined to 16-163 kg N ha^{-1} in the swards amended with 480 kg N ha^{-1}. Due to organic N in slurry, the N surplus – N input minus output – was significantly affected by fertilizer type and less by application rates. However, we did not find a convincing correlation between N surplus and nitrate leaching, neither was nitrate leaching dependent of fertilizer type. Nitrate leaching, ranging between10 and 105 kg N ha^{-1}, was best described by a quadratic function. We concluded that up to a fertilization rate of 240 kg N ha^{-1}, low leaching levels could be achieved, independently of the fertilizer type.

Keywords: red clover, white clover, temporal variation, nitrogen balance, surplus

Introduction

In dairy farming, cattle slurry is typically applied on grasslands as a basic dressing and any use of mineral fertilizer would be in addition to that. In total, a rate of 300 kg N ha^{-1} is often applied as standard in Denmark, with no consideration for the clover content of grass-clover leys. When clover content is low, this rate is too low, and when clover content is high, it is too high. Since, fertilizer N addition generally decreases N_2-fixation activity and quantities (Nesheim *et al.*, 1990), non-optimized fertilization may have a negative impact on resource use, the herbage quality, the environment, and the economy in many cases. The aim of this study was to examine response to N fertilizer application in clover dynamics, the proportion of N derived from the atmosphere (%Ndfa), the quantitative N_2-fixation (qBNF) and nitrate leaching under fertilized grass-clover leys.

Materials and methods

The experiments were conducted at two farmers' fields (hereafter 'SW' and 'MW') located on sandy soils in the Western part of Denmark over two years in 2018 and 2019 (Kristensen *et al.,* 2022). The leys were composed of 10% white clover (*Trifolium repens* L.) and 90% of different varieties of perennial ryegrass (*Lolium perenne* L.) at the SW site, and a mixture of 11% red clover (*Trifolium pratense* L.), 7% white clover, 37% perennial ryegrass and 45% festulolium (*Festulolium braunii,* K.A.) at the MW site. At both sites, ten treatments in four replicates were arranged in randomized plot design. Plots (12×3 m) were fertilized with increasing N levels from 0 to 480 kg plant available N ha^{-1}, either as mineral fertilizer (N-S 27-4, 13.5% ammonium-N, 13.5% nitrate-N) or combined with a basic application of acidified cattle slurry (120 kg available N ha^{-1}). These were applied in spring and after each cut (Table 1). In addition to the N fertilizer, all plots received basic fertilization P, K, S, Mg in spring.

The %Ndfa of clovers in the ley mixtures was estimated using the ^{15}N isotope dilution method (Fried and Middelboe, 1977). The botanical composition, yields and N uptake were determined at each cut, as well

Table 1. Nitrogen application treatments distribution during the growth season in experiment with grass-clover leys.

	Annual N rate	Spring		After 1st cut		After 2nd cut	After 3rd cut
		Slurry	Mineral	Slurry	Mineral	Mineral	Mineral
	kg inorganic N ha^{-1}						
Control	0						
Mineral N	60		60				
	120		80		40		
	240		120		80	40	
	360		150		120	60	30
	480		150		120	120	90
Slurry + mineral N	120	60		60			
	240	60	60	60	20	40	
	360	60	90	60	60	60	30
	480	60	90	60	60	120	90

(Fontaine *et al.*, unpublished data). An N balance was established based on inputs which corresponded to the fertilizer amendment (mineral N and/or slurry available and organic N), N_2 fixation and atmospheric deposition, and outputs which corresponded to the removed N fraction measured in the harvested biomass. Nitrate leaching was measured from suction cups installed at 1 m depth in each plot during the period September-March.

Results and discussion

Fertilization significantly increased the overall yields ($P\leq0.001$) and N uptakes ($P\leq0.001$) of the leys. The increase was generally the result of a significant rise in grass yield larger than a decrease in clover yield. Two types of %Ndfa responses were observed with increasing N fertilization: (a) a relatively flat response with high %Ndfa across N levels (>80% when the N level was below 200-300 kg available N ha^{-1}), and (b) a linear decrease with increasing N (at MW site in treatment with mineral N only application). The qBNF of the harvested biomass in plots without fertilization was 193-286 kg N ha^{-1} and reduced to 16-79 kg N ha^{-1} after application of 480 kg N ha^{-1}. A lower reduction related to the slurry application in comparison to mineral N only was observed at MW site ($P\leq0.05$), where qBNF were maintained around 155-163 kg N ha^{-1}.

The surplus (difference between inputs and output in the N balance) was in the range -11 to 51 kg N ha^{-1} in the control and similar in the treatments with mineral N only application. Surplus was, however, much higher for treatments receiving slurry N, because of the organic N applied (slurry contained ca. 46% organic N that was added to the input of the balance).

Nitrate leaching varied between 3 and 117 kg N ha^{-1} (Figure 1). Leaching curves as a function of fertilizer application had a slight decrease or slow start from 0 to 120-240 kg N ha^{-1} and an increase afterwards. This was confirmed by the fit of the quadratic equation ($0.37\leq R^2\leq0.86$). Nitrate leaching was fertilizer rate dependent ($P\leq0.001$) but independent of fertilizer type ($P=0.242$).

Although it is widely considered that N excess is a good predictor of N leaching under an annual crop, particularly in sandy soils, this was not the case under a perennial grass-clover crop. A simple N balance based on input and output could not predict nitrate leaching.

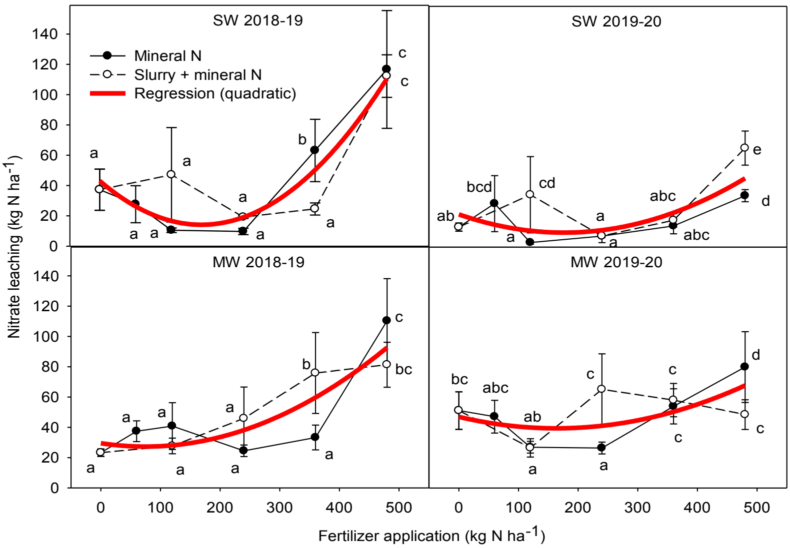

Figure 1. Annual nitrate leaching at both sites and years as function of N fertilizer rate and fitted regression. Error bars show standard errors (n=4). Different letters indicate significant differences between treatments, based on repeated measurements ANOVA.

Conclusions

Nitrogen fixation from clover represented an input of circa 200-300 kg N ha^{-1}. However, moderate fertilization of a grass–clover sward ensures the farmers have productive and stable grass yields, even if the clover proportion reduces. A split N fertilization rate up to 200-300 kg available N ha^{-1} was therefore optimal for uptake by grass with minimal impact on legume N_2-fixation (>80%Ndfa) and nitrate leaching (<40 kg N ha^{-1}). This was particularly true when slurry was added at modest N levels (i.e. at 120 kg available N ha^{-1}).

References

Fried M. and Middelboe V. (1977) Measurement of amount of nitrogen fixed by a legume crop. *Plant and Soil* 47, 713-715. https://doi.org/10.1007/BF00011042

Kristensen R.K., Fontaine D., Rasmussen J. and Eriksen J. (2022) Contrasting effects of slurry and mineral fertilizer on N2-fixation in grass-clover mixtures. *European Journal of Agronomy* 133, 126431. https://doi.org/10.1016/j.eja.2021.126431

Nesheim L., Boller B.C., Lehmann J. and Walther U. (1990) The effect of nitrogen in cattle slurry and mineral fertilizers on nitrogen fixation by white clover. *Grass and Forage Science* 45, 91-97. https://doi.org/10.1111/j.1365-2494.1990.tb02186.x

Modelling studies of beef and dairy farming in boreal environments: a review

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Abstract

Process-based simulation models are increasingly used in livestock agriculture as an adjunct to field studies and are time and cost effective if properly validated with field data. In boreal grasslands, models can effectively estimate biomass production but have been less successful in assessing soil organic carbon (SOC) and greenhouse gas (GHG) emissions from soil and livestock. Given the potential role of model simulations in management planning this situation needs addressing. We conducted a review of current research, identifying the models most used in agricultural research including options for grassland, livestock, and soil carbon flux simulations. Unvalidated simulations were filtered out, and validation outcomes of the remainder were scored as 'poor', 'fair', 'good' or 'excellent'. Research gaps and potential model weaknesses were identified creating a basis for future model validation work for boreal grassland agriculture.

Keywords: ecophysiological modelling, validation, boreal agriculture

Introduction

Boreal regions are expected to experience increasing climatic and land use changes in the coming decades (Unc *et al.*, 2021), the rapidity of which will make mistakes in adaptation costly. Ecosystem models are a key tool in studying the complex interacting C and N processes in agricultural systems (Jones *et al.*, 2016), permitting the integration and interpretation of multiple variables, and can be implemented more quickly and at lower cost than field experiments. Models have been successfully implemented within Europe, North America, Australia and elsewhere (Höglind *et al.*, 2020; Lugato *et al.*, 2015), and although reviews of model performance have been carried out (Brilli *et al.*, 2017), few have focused on boreal and similar grassland systems. Given this deficit, the structured selection, calibration, and validation of models for simulating C and N exchange and related outcomes in boreal grasslands is being carried out in conjunction with the Ministry of Agriculture and Forestry (MMM), Academy of Finland, and EU regional funding agencies for carbon neutrality in the beef and dairy sectors, at Luke Maaninka (Natural Resources Institute Finland). The data collected will be used to validate and calibrate existing models, and assist tool selection for the soil type, management and required outputs. To guide these efforts, a literature review was conducted and validation data for GHG, soil and production outputs in boreal grassland systems was extracted and summarized.

Materials and methods

Literature searches were conducted using the Web of Science Database ® and Google Scholar between 4 May and 30 July 2021. Topic (grasslands, greenhouse gas emissions, soil carbon, methane, carbon dioxide, nitrogen dioxide) and regional keywords (boreal, northern, Nordic, Scandinavia, Finland, Sweden, Norway, Canada, Iceland, United Kingdom, Ireland) were used. Articles that reported field experiments and model validation data were included, and of an initial 65 papers, ten contained validation data for seven models (Table 1). An assessment method was adapted from Despotovic *et al.* (2016), He *et al.* (2020) and Moriasi *et al.* (2015): model performance was graded as 'poor', 'fair', 'good' or 'excellent'. Validation results from each article were extracted and scored from 1 to 4 on this scale (Table 2), and mean scores across methods were calculated. A final score was calculated as the sum of the validation scores.

Table 1. Summary of scores given for each model for variables validated.[1]

Variable		BASGRA_N	CATIMO	DNDC	FASSET	IFSM	PaSim	STICS
GHG	NEE	-	-	-	-	-	fair	-
	CO2	-	-	good	-	-	-	-
	N2O	-	-	good	fair	-	-	-
	NO3	-	-	-	fair	-	-	-
Soil	C seq	-	-	good	-	-	-	-
	Mineral N	-	-	-	good	-	-	-
	N leaching	-	-	poor	-	-	-	-
	N uptake	-	-	-	-	excellent	-	-
Crop	Crop yield	-	-	-	-	fair	-	-
	DM	good	poor	fair	-	good	excellent	fair
	CP	good	-	-	-	-	-	-
	NDF	-	-	-	-	fair	-	-
	LAI	-	-	-	-	-	excellent	-
	Fert v DM	poor	poor	-	-	-	-	fair
Number of studies		1	1	4	2	2	1	2
Overall model score		7	2	12	7	11	9	4

[1] Values represent the mean of scores given to each validation method where more than one was used to assess the variable. Data extracted from Abdalla *et al.*, 2011; Berntsen *et al.*, 2006; Chatskikh *et al.*, 2005; Congreves *et al.*, 2016; He *et al.*, 2020; Höglind *et al.*, 2020; Jégo *et al.*, 2015; Korhonen *et al.*, 2018; Rafique *et al.*, 2011; Vuichard *et al.*, 2007.

Table 2. Criteria for assessing results using the range of validation models used in the literature cited here.[1]

Validation method	Poor	Fair	Good	Excellent
Average relative error (ARE)	≥40%	39-21%	20-11%	≤10%
Mean absolute error (MAE)	≥4.0	3.0-3.9	2.0-2.9	≤1.9
Mean error (ME)	≥20.0	10.0-19.9	4.0-9.9	≤3.9
Normalized average relative error (NARE)	≥±15%	≤±11-15%	≤±2-10%	±1%
Normalized mean error (NME)	≥20%	15-20%	11-15%	≤10%
Normalized root mean square error (NRMSE)	≥40%	39-21%	20-11%	≤10%
Nash-Sutcliffe efficiency (NSE)	≤0.50	0.51-0.70	0.71-0.80	≥0.81
Pearson's correlation (R)	≤0.2	0.3-0.4	0.5-0.7	0.8-1.0
Coefficient of determination (R2)	≤0.2	0.3-0.4	0.5-0.7	0.8-1.0
Root mean square error (RMSE)	≥40	20-39	Nov-19	0-10
Relative Root mean square error (rRMSE)	≥0.30	0.21-0.30	0.11-0.20	≤0.10
Willmott's d	0.1-0.3	0.4-0.5	0.6-0.9	1.0
Relative mean bias error (rMBE)	≥±0.3	±0.2	±0.1	±0.01

[1] Values were given a score: poor = 1, fair = 2, good = 3, and excellent = 4.

Results and discussion

Model validation results varied considerably. DNDC, IFSM and PaSim performed well, though for DNDC this was partly due to the number of studies found and the inclusion of a Canadian soil-adapted version of the model; *v.Can*, in Congreves *et al.* (2016), although GHG emissions and C sequestration performed better than other outcomes. IFSM's performance was good for N uptake and reasonable for biomass, and PaSim's crop growth simulations stood out notably. Biomass production simulations from BASGRA_N seem promising, and FASSET satisfactorily simulated N_2O and NO_3. STICS biomass and fertilizer response simulations were satisfactory, but CATIMO performed poorly over all (Table 1).

Conclusions

Evidence of model validation for grasslands in boreal climates is patchy and there remains a high degree of uncertainty. The literature examined showed considerable gaps and lack of consistency in model testing. Systematic testing and validation of key outcomes in current models can identify strengths and weaknesses, allowing confident model selection for specific situations in boreal grassland ecosystems.

References

Abdalla M., Kumar S., Jones M., Burke J. and Williams M. (2011) Testing DNDC model for simulating soil respiration and assessing the effects of climate change on the CO2 gas flux from Irish agriculture. *Global Planetary Change* 78, 106-115.

Berntsen J., Grant R., Olesen J.E., Kristensen I.S., Vinther F.P., Molgaard J.P. and Petersen B.M. (2006) Nitrogen cycling in organic farming systems with rotational grass-clover and arable crops. *Soil Use and Management* 22, 197-208.

Brilli L., Bechini L., Bindi M., Carozzi M., ..., and Bellocchi G. (2017) Review and analysis of strengths and weaknesses of agro-ecosystem models for simulating C and N fluxes. *Science of the Total Environment* 598, 445-470.

Chatskikh D., Olesen J.E., Berntsen J., Regina K. and Yamulki S. (2005) Simulation of effects of soils, climate and management on N_2O emission from grasslands. *Biogeochemistry* 76, 395-419.

Coleman K., Jenkinson D.S., Crocker G.J., Grace P.R., Klír J., Körschens M., Poulton P.R. and Richter D.D. (1997) Simulating trends in soil organic carbon in long-term experiments using RothC-26.3. *Geoderma* 81, 29-44.

Congreves K.A., Grant B.B., Dutta B., Smith W.N., Chantigny M.H., Rochette P. and Desjardins R.L. (2016) Predicting ammonia volatilization after field application of swine slurry: DNDC model development. *Agriculture, Ecosystems and Environment* 219, 179-189.

Despotovic M., Nedic V., Despotovic D. and Cvetanovic S. (2016) Evaluation of empirical models for predicting monthly mean horizontal diffuse solar radiation. *Renewable and Sustainable Energy Reviews* 56, 246-260.

He W., Dutta B., Grant B.B., Chantigny M.H., Hunt D., Bittman S., Tenuta M., Worth D., VanderZaag A., Desjardins R.L. and Smith W.N. (2020) Assessing the effects of manure application rate and timing on nitrous oxide emissions from managed grasslands under contrasting climate in Canada. *Science of the Total Environment* 716, 135374.

Höglind M., Cameron D., Persson, T., Huang, X. and van Oijen, M. (2020) BASGRA_N: A model for grassland productivity, quality and greenhouse gas balance. *Ecological Modelling* 417, 10892.

Jégo G., Rotz C.A., Bélanger G., Tremblay, G.F., Charbonneau, É. and Pellerin, D. (2015) Simulating forage crop production in a northern climate with the Integrated Farm System Model. *Canadian Journal of Plant Sciences* 95, 745-757.

Jones J.W., Antle J.M., Basso B., Boote, K.J., Conant, R.T., Foster, I., Godfray, H.C.J., Herrero, M., Howitt, R.E., Janssen, S., Keating, B.A., Munoz-Carpena, R., Porter, C.H., Rosenzweig, C. and Wheeler, T.R., (2016) Brief history of agricultural systems modeling. *Agricultural Systems* 155, 240-254.

Korhonen P., Palosuo T., Persson T., Höglind M., Jégo G., Van Oijen M., Gustavsson A.M., Bélanger G. and Virkajärvi P. (2018) Modelling grass yields in northern climates – a comparison of three growth models for timothy. *Field Crops Research* 224, 37-47.

Lugato E., Bampa F., Panagos P. and Montanarella L. (2015) Potential carbon sequestration of European arable soils estimated by modelling a comprehensive set of management practices. *Global Change Biology* 20, 3557-3567.

Moriasi D.N., Gitau M.W., Pai N. and Daggupati P. (2015) Hydrologic and water quality models: performance measures and evaluation criteria. *Transactions ASABE* 58, 1763-1785.

Rafique R., Peichl M., Hennessy D. and Kiely G. (2011) Evaluating management effects on nitrous oxide emissions from grasslands using the process-based DeNitrification-DeComposition (DNDC) model. *Atmospheric Environment* 45, 6029-6039.

Unc,A., Altdorff D., Abakumo, E., Adl S., Baldursson S., Bechtold M., Cattani D.J., Firbank L.G., Grand S. and Borchard N. (2021) Expansion of agriculture in northern cold-climate regions: a cross-sectoral perspective on opportunities and challenges. *Frontiers in Sustainable Food Systems* 5, 663448.

Vuichard N., Soussana J.F., Ciais P., Viovy N., Ammann C., Calanca P., Clifton-Brown J., Fuhrer J., Jones M. and Martin C. (2007) Estimating the greenhouse gas fluxes of European grasslands with a process-based model: 1. Model evaluation from *in situ* measurements. *Global Biogeochemical Cycles* 21, GB1004.

Effects of management factors and additive treatments on grass silage quality

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Abstract

The objective was to evaluate the fermentation quality, aerobic stability and microbial quality of grass silages produced under different management factors. Timothy and meadow fescue grass were harvested in two consecutive cuts and ensiled using five different silage additives. Additional management factors included normal, loose or delayed compaction in the first cut and two distinct dry matter (DM) levels in the second cut. Delayed compaction resulted in higher ensiling losses than normal, loose being intermediate. Higher DM content restricted silage fermentation. All additives improved silage quality similarly irrespective of compaction method, but the additive effects were in general greater in low than in high DM silages.

Keywords: compaction, dry matter content, fermentation, *Phleum pratense*, wilting, *Festuca pratensis*

Introduction

Ensiling is a common way of preserving forage, and thereby provide green biomass for ruminants in spite of the seasonality in grass growth. Preservation quality of grass silage greatly affects losses during storage, aerobic stability during the feed-out period and voluntary feed intake of animals. Silage quality in practice is still a concern, and this emphasizes the need to continuously develop it to ensure economic performance and safety of the food chain based on ruminants. The development and use of various silage additives aims at controlling or modulating fermentation pattern in silage to improve quality (Muck and Kung, 1997). Apart from the effects of silage additives, silage quality and nutritional values are influenced by the dry matter (DM) content, which can be manipulated by the extent of wilting modified by prevailing weather conditions. Controlling silage DM content may become more challenging due to more extreme weather events related to climate change. Another important management factor affecting silage fermentation and subsequent quality is the packing density of the biomass (Kung *et al.*, 2018), which is a result of the compaction process necessary to increase density and remove oxygen from the interior of the silage. The needs for top quality silage are thus evident and the current study evaluated the fermentation quality, aerobic stability and microbial quality of grass silages produced under different management factors.

Materials and methods

A timothy (*Phleum pratense*) and meadow fescue (*Festuca pratensis*) sward was harvested in the first and second cuts during summer 2020 in Jokioinen (60°48'N, 23°29'E), Finland. On both occasions, the grass was cut, precision chopped using farm scale machinery and transported to the laboratory without any additive. In the first cut, grass was wilted for 24 h before silage making. A factorial 3×5 design of experimental treatments was used, comprising three compactions: (1) normal compaction, (2) loose compaction, (3) delayed compaction and covering, where silos were filled similarly as in normal compaction, but left uncovered and then applying additional compaction and covering 24 h later; and five additive treatments: (1) control (C), as a negative treatment without additive, (2) formic acid based additive (FA; AIV Ässä Na, Eastman Chemical Company, Oulu, Finland at 5 l tonnes^{-1}), (3) homofermentative strains of the lactic acid bacteria (LAB) *Lactobacillus plantarum* and *Pediococcus pentosaceus* (HO; Bonsilage, Schaumann Agri International GmbH, Pinneberg, Germany at 1 g tonnes^{-1}), (4) heterofermentative strains of the LAB *Lactobacillus buchneri*, *Lactobacillus plantarum* and *Lactobacillus plantarum* (HE; Feedtech Silage F600, DeLaval, Tumba, Sweden at 1 g $tonnes^{-1}$) and (5) salt based additive (SA; Safesil Pro, Salinity AB, Göteborg, Sweden at 5 l $tonnes^{-1}$). In the second cut, silages were prepared with two distinct DM contents. For the lower DM, grass was ensiled 2 h after harvesting, whereas for the higher DM, grass was artificially wilted in a forced-air open-circuit drier using air temperature of 30 °C for 3 h. The same additives as in the first cut were used for both DM levels. All silages were ensiled in cylindrical ca. 10 litre plastic silos using 3 replicates per treatment. After a 3-month ensiling period, the silage samples were taken and analysed for chemical composition, fermentation quality, aerobic stability and microbial quality. Data were analysed using a SAS MIXED procedure with experimental treatment factors as fixed effects and replicates as a random effect separately for first and second cut.

Results and discussion

The DM content in the first cut was 358 g kg^{-1}, and in the second cut 223 and 517 g kg^{-1}. Fermentation coefficients of the raw materials were 50, 39 and 74, respectively, for the first cut, and the low and high DM contents in the second cut. Similar microbial quality patterns were found in both cuts, except for mould content, which was slightly higher in the fresh forage of the second cut. Type of compaction did not have significant effects on silage fermentation profile, but delayed compaction resulted in higher ensiling losses than normal, with loose being intermediate (Table 1). Further, no interactions were found between compaction types and additive treatments. Higher DM content clearly restricted silage fermentation, and the additive effects were, in general, greater in low than in high DM silages (Table 2). All additives improved silage quality in both cuts. Fermentation was restricted by FA resulting in greater amount of water-soluble carbohydrates and lower amount of lactic acid, while no differences were found among other additive treatments for those parameters. Aerobic stability was higher for FA and SA, followed by HE, while C and HO had the lowest aerobic stability, not differing from each other. No effect of compaction or additive treatment was found for the microbial quality parameters. The most efficient additives in lowering the pH of silages were both inoculants at both DM contents (Table 2), while higher pH values were found for C, SA and FA in low DM, and SA and FA in high DM. FA application resulted in silages with the lowest concentrations of ammonia N in both DM levels (similar to HO in high DM), but the values for other additives were also moderate indicating that they were also well preserved.

Table 1. Chemical composition, fermentation quality, aerobic stability and ensiling losses of first cut mixed timothy and meadow fescue silage treated with different silage and using normal, loose or delayed compaction.[1]

Compaction	Normal					Loose					Delayed					SEM	*P*-value	
Additive	C	FA	HO	HE	SA	C	FA	HO	HE	SA	C	FA	HO	HE	SA		Comp	Add
Dry matter (DM), g kg^{-1}	373	378	383	409	374	368	364	399	407	356	399	390	413	396	384	18.1	0.306	0.151
pH	4.31^{abc}	4.33^{ab}	4.20^{c}	4.29^{abc}	4.24^{abc}	4.26^{abc}	4.35^{a}	4.22^{bc}	4.32^{abc}	4.23^{abc}	4.28^{abc}	4.34^{ab}	4.23^{abc}	4.30^{abc}	4.28^{abc}	0.024	0.762	<0.001
Ammonia N, g kg^{-1}N	57^{abc}	43^{abc}	52^{abc}	52^{abc}	60^{abc}	59^{abc}	41^{c}	51^{abc}	55^{abc}	62^{ab}	52^{abc}	42^{bc}	47^{abc}	55^{abc}	62^{a}	3.8	0.781	<0.001
Water soluble carbohydrates, g/kg DM	13^{b}	63^{a}	14^{b}	9^{b}	14^{b}	12^{b}	47^{a}	15^{b}	11^{b}	13^{b}	14^{b}	51^{a}	16^{b}	8^{b}	13^{b}	5.9	0.728	<0.001
Ethanol, g kg^{-1}DM	9.4^{a}	1.3^{b}	7.7^{a}	7.8^{a}	2.7^{b}	9.2^{a}	1.1^{b}	6.9^{a}	7.7^{a}	3.2^{b}	8.1^{a}	1.3^{b}	6.9^{a}	8.3^{a}	3.3^{b}	0.58	0.842	<0.001
Lactic acid, g kg^{-1}DM	80.2^{a}	44.6^{b}	80.7^{a}	74.6^{a}	75.8^{a}	76.0^{a}	43.6^{b}	75.2^{a}	76.3^{a}	77.0^{a}	78.7^{a}	49.5^{b}	79.2^{a}	74.3^{a}	70.5^{a}	3.42	0.773	<0.001
Acetic acid, g kg^{-1}DM	12.3^{b}	6.4^{c}	11.8^{b}	16.6^{a}	15.2^{a}	11.9^{b}	6.3^{c}	12.3^{b}	16.4^{a}	16.5^{a}	12.1^{b}	6.3^{c}	11.3^{b}	15.6^{a}	16.7^{a}	0.54	0.657	<0.001
Propionic acid, g kg^{-1}DM	0.09^{c}	2.68^{ab}	0.08^{c}	0.09^{c}	0.09^{c}	0.09^{c}	2.88^{a}	0.10^{c}	0.09^{c}	0.10^{c}	0.09^{c}	2.6^{b}	0.08^{c}	0.10^{c}	0.09^{c}	0.053	0.191	<0.001
Butyric acid, g kg^{-1} DM	0.02	0	0.01	0	0	0.04	0	0.03	0.01	0.01	0.10	0	0.01	0.01	0.01	0.023	0.436	0.090
Aerobic stability (2 °C), hours	111^{cd}	320^{ab}	112^{cd}	282^{abc}	360^{a}	93^{d}	293^{ab}	110^{cd}	251^{abcd}	360^{a}	146^{bcd}	322^{ab}	184^{abcd}	284^{abc}	360^{a}	34.4	0.230	<0.001
Ensiling losses, g kg^{-1}initial DM	17.2^{abc}	8.4^{d}	18.1^{ab}	18.0^{ab}	11.0^{cd}	19.2^{ab}	7.8^{d}	17.5^{abc}	22.6^{a}	10.9^{cd}	22.2^{a}	11.0^{cd}	22.0^{a}	23.8^{a}	14.3^{bcd}	1.31	<0.001	<0.001
Density, kg m^{-3}	504^{a}	475^{abcd}	480^{ab}	482^{ab}	482^{ab}	386^{de}	369^{e}	365^{e}	389^{cde}	402^{bcde}	468^{abcd}	519^{a}	453^{abcde}	470^{abcd}	477^{abc}	17.0	<0.001	0.492

[1] C = control; FA = formic + propionic acid; HO = homofermentative strains of lactic acid bacteria; HE = heterofermentative strains of lactic acid bacteria; SA = salt-based additive; SEM = standard error of the mean; Comp = effect of compaction; Add = effect of additive; no interaction effect was found between Comp and Add. Values with different letter in a row are significantly different at 5% Tukey test.

Table 2. Chemical composition, fermentation quality, aerobic stability, ensiling losses and microbial quality of second cut mixed timothy and meadow fescue silage treated with different silage additives at different dry matter contents.[1]

Dry matter (DM) level	Low					High					SEM	*P*-value		
Additive	**C**	**FA**	**HO**	**HE**	**SA**	**C**	**FA**	**HO**	**HE**	**SA**		**DM**	**Add**	**DM×Add**
DM, g kg^{-1}	221^{b}	228^{b}	218^{b}	223^{b}	226^{b}	526^{a}	521^{a}	512^{a}	512^{a}	513^{a}	3.9	<0.001	0.104	0.231
pH	3.96^{d}	3.94^{d}	3.83^{e}	3.81^{e}	4.00^{d}	4.30^{b}	4.60^{a}	4.16^{c}	4.24^{bc}	4.30^{b}	0.020	<0.001	<0.001	<0.001
Ammonia N, g kg^{-1}N	57^{a}	28^{e}	41^{cd}	55^{ab}	62^{a}	42^{cd}	27^{e}	35de	43^{c}	48^{bc}	1.6	<0.001	<0.001	<0.001
Water soluble carbohydrates, g kg^{-1}DM	29^{d}	32^{d}	30^{d}	23^{d}	34^{d}	82^{b}	172^{a}	90^{b}	62^{c}	82^{b}	3.6	<0.001	<0.001	<0.001
Ethanol, g kg^{-1}DM	4.5^{b}	7.2^{a}	4.1^{bc}	3.5^{bcd}	1.2^{e}	2.0^{cde}	0.7^{e}	1.8^{de}	2.0^{cde}	1.0^{e}	0.42	<0.001	<0.001	<0.001
Lactic acid, g kg^{-1}DM	93.5^{b}	54.8^{d}	114.0^{a}	97.0^{b}	86.6^{b}	52.5^{d}	19.1^{e}	68.7^{c}	60.2^{cd}	51.5^{d}	2.49	<0.001	<0.001	0.245
Acetic acid, g kg^{-1}DM	15.3^{d}	22^{a}	12.6^{ef}	20.1^{b}	17.1^{c}	10.7^{g}	6.3^{i}	8^{h}	13.8^{de}	11.8^{fg}	0.32	<0.001	<0.001	<0.001
Propionic acid, g kg^{-1}DM	0.19^{c}	3.32^{a}	0.21^{c}	0.25^{c}	0.15^{c}	0.08^{c}	1.98^{b}	0.08^{c}	0.08^{c}	0.10^{c}	0.050	<0.001	<0.001	<0.001
Butyric acid, g kg^{-1}DM	0.05^{a}	0.06^{a}	0.03^{a}	0.03^{a}	0.01^{a}	0.02^{a}	0.01^{a}	0.02^{a}	0.02^{a}	0.02^{a}	0.010	0.013	0.398	0.180
Aerobic stability (2 °C), h	94^{d}	237^{bc}	54^{d}	112^{d}	336^{a}	244^{bc}	326^{ab}	222^{c}	336^{a}	336^{a}	18.2	<0.001	<0.001	<0.001
Ensiling losses, g kg^{-1}initial DM	9.3^{cd}	12.3^{abcd}	8.7^{cd}	9.5^{cd}	6.6^{d}	16.3^{ab}	9.9^{bcd}	14.7^{abc}	18.3^{a}	16.8^{a}	1.32	<0.001	0.262	0.001
Density, kg $^{m-3}$	696^{a}	680^{a}	670^{a}	687^{a}	641^{a}	484^{b}	442^{b}	463^{b}	490^{b}	442^{b}	22.4	<0.001	0.211	0.890
Moulds, log cfu g^{-1}	10^{3a}	10^{3ab}	10^{3ab}	10^{3ab}	10^{2b}	10^{2b}	10^{2b}	10^{2b}	10^{2b}	10^{2b}	10^{2}	0.001	0.080	0.136

[1] C = control; FA = formic + propionic acid; HO = homofermentative strains of lactic acid bacteria; HE = heterofermentative strains of lactic acid bacteria; SA = salt-based additive, SEM = standard error of the mean; cfu = colony-forming unit; Add = effect of additive; DM = effect of dry matter level; DM × Add = interaction between dry matter level and additive effects. Values with different letter in a row are significantly different at 5% Tukey test.

Conclusions

Silage management factors, such as compaction and especially DM level, combined with the use of additives, greatly affected the quality of grass silages. It is possible that the pilot scale silos were not able to mimic the effects of compaction compared with practical situations where air ingress into the silos during the in-silo fermentation and feed-out time would likely affect fermentation quality and aerobic stability to a greater extent. In practical situations, operative guidelines of good silage production, including proper compaction, should be followed although the effects in the current study were minor. Use of additives improved the quality of grass silage, but different additives modified silage quality in different ways. Increasing DM content and use of FA effectively restricted silage fermentation.

References

Kung Jr L., Shaver R.D., Grant R.J. and Schmidt R.J. (2018) Silage review: Interpretation of chemical, microbial, and organoleptic components of silages. *Journal of Dairy Science* 101, 4020-4033.

Muck R. and Kung L. (1997) *Effects of silage additives on ensiling*. In: Proceedings from the Silage: Field to Feedbunk North American Conference, Hershey, PA, February 1997. pp. 187-200. Ithaca: Northeast Regional Agricultural Engineering Service.

Inclusion of chicory in grass-clover mixtures enhances leys productivity and herbage quality compared to monocultures

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Abstract

The use of herbs in mixtures with grasses and legumes is an innovative approach implemented in grassland-based forage systems in some European countries. The aim of this study was to determine the effects of the inclusion of chicory in grass-legume mixtures on the yield and herbage quality of sward leys, compared with monocultures, at two sites of Wielkopolska region, Poland. The experiment was established in 2015 at Brody (Luvisol soil) and Szelejewo (Cambisol soil) on 10 m^2 plots in a simplex design to define four monocultures and eight mixtures of the four species (*Lolium perenne* L., *Trifolium pratense* L., *Trifolium repens* L. and *Cichorium intybus* L.). During two study years (2016-2017) herbage biomass in the whole plot was harvested three times per year to determine annual dry matter yield. Concentrations of the protein, sugars, crude ash, calcium and magnesium in herbage were also determined. The above-ground biomass of two- and four-species mixtures was significantly higher than that obtained from monocultures. The herbage yield collected from leys with chicory in our study was higher on Luvisols than on Cambisols.

Keywords: grass-legume mixture, chicory, ley farming, herbage quality

Introduction

Grass-legume mixtures such as ryegrass-clover swards are commonly used in temporary agricultural grassland, because the herbage yield of a mixture can exceed that of its best-performing species when grown in a pure stand (transgressive overyielding) (Finn *et al.*, 2013). One innovative approach in the composition of multi-species swards is the addition of herbs in grass-legume mixtures. Recent studies have identified that chicory-containing mixtures improve productivity, which can be a promising strategy for enhancing agricultural output and forage quality in European temporary grasslands (Cong *et al.*, 2018). The productivity of the grass-legume mixtures is affected by environmental, biotic and management factors. One of them is the soil type. The aim of this study was to determine the effects of inclusion of chicory in grass-legume mixtures on leys productivity and herbage quality compared to monocultures in conditions of two soil types.

Materials and methods

A study was carried out during 2016-2017 at two sites of the Wielkopolska region in Poland: at Brody Experimental Station (52°43'24'N, 16°30'31'E) of the Poznan University of Life Sciences, and Szelejewo Breeding Station (51°86'34'N, 17°15'18'E) of Danko Plant Breeding Ltd. The experiment was established in early autumn 2015 on two soil types: (1) Luvisols (pH_{KCl} 6.2, total N of 0.13%, P_2O_5 of 29.4 mg 100 g^{-1}, K_2O 17.9 mg 100 g^{-1}, Mg 5.8 mg 100 g^{-1}) at Brody, and (2) Cambisols (pH_{KCl} 5.9, total N 0.10%, P_2O_5 22.0 mg 100 g^{-1}, K_2O 10.2 mg 100 g^{-1}, Mg 6.7 mg 100 g^{-1}) in Szelejewo on 10 m^2 (1×10 m) plots in a simplex design (Ramseier *et al.*, 2005) to define four monocultures (Mono) and eight mixtures consisting of varying proportions of the four species (*Lolium perenne – Lp*, *Trifolium pratense – Tp*, *Trifolium repens – Tr* and *Cichorium intybus – Ci*). The 8 mixtures consisted of (1) four mixtures dominated in turn by each species (Domi-66.7% of dominant and 11.1% of each of the other species), and (2) three mixtures composed of two species (Bi-50% *Ci* and 50% *Lp*, *Tr* or *Tp*) and (3) the Centroid community (25% of each species). The monocultures and mixtures were sown according to seed rate in pure stands recommended in Poland (*Lp*-30, *Tp*-20, *Tr*-15, *Ci*-5 kg ha^{-1}). The sward was managed by cutting three times each year. Fertilizer was applied each year at the rate of 90 kg N ha^{-1} (30 kg ha^{-1} in spring and 30 after 1st and 2nd regrowths), 60 kg P ha^{-1}, and 90 kg K ha^{-1}. The yearly mean temperature and total precipitation for 2016 and 2017 at Brody were 9.7, 9.0 °C and 622, 764 mm, and at Szelejewo 10.0, 9.2 °C, and 721, 765 mm. For each plot, biomass of aboveground vegetation was measured at each harvest. This was done by cutting the whole plot to a height of 5 cm and determining the fresh weight. A subsample of this material was taken, its fresh weight was determined and the material was dried at 65 °C to constant weight to measure dry matter (DM). The samples collected for DM were ground to pass through a sieve of 1 mm of mesh size and used for forage quality analysis. Crude protein (based on total N content by Kjeldahl × 6.25), water soluble carbohydrates (WSC; colorimetric method by Dubois), crude ash, calcium and magnesium in DM of herbage were analysed. Statistical analysis of the total annual yield data was carried out according to simplex model (Finn *et al.*, 2013). The analysis of evenness gradient was performed; this analysis is based on classifying the 12 mono/mixture plots into 3 groups: L – low (monocultures, 4 plots), M – medium (mixtures dominated by one species, 4 plots), H – high (two species and Centroid mixtures, 4 plots). Differences in herbage quality between mono/mixtures levels were tested using Tukey's *post hoc* test (R software).

Results and discussion

The mixtures generally had significantly higher annual yields than the monocultures at both study sites: on Luvisols by 29.4% and on Cambisols by 31.6% (Table 1). The annual DM yield on Luvisols over the two years was largest in Domi *Ci*, Domi *Lp* and Domi *Tp* mixtures, 17,564, 15,138 and 15,119 kg ha^{-1}, while on Cambisols, Centroid and Bi *Tp-Ci* performed well, 13,385 and 12,975 kg ha^{-1}, respectively. The soil type influenced the productivity of chicory-containing grass-legume mixtures. Higher DM yields were obtained on Luvisols in comparison with Cambisols. On average for all *Ci* mixtures, in 2016 the DM yield was 12,295 kg ha^{-1} vs 7,460 kg ha^{-1} and in 2017 it was 15,461 kg ha^{-1} vs 15,216 kg ha^{-1}. The data in Table 1 show consistent positive effects of increasing plant evenness. In our study, the yield of four- and two-species mixtures exceeded that expected from monoculture performances, either on Luvisols (P<0.001) or Cambisols (P<0.01). This effect has been reported by several authors, e.g. Tilman *et al.* (1996) and Finn *et al.* (2013). There was also a highly significant effect of year for both soil types (P<0.001), but no effect of evenness × year interaction.

Species composition affected the concentrations of protein, WSC, crude ash, calcium and magnesium in herbage (Table 2). The protein content was higher in swards containing clovers, but only Mono *Tr* and Bi *Tr-Ci* differed significantly between all treatments. In the *Lp* swards (Mono, Bi and Domi) a higher WSC content in herbage was determined, opposite to that of clover-containing swards. Clover and chicory mono/mixtures produced higher mineral concentrations (in total, Ca and Mg) than *Lp*. This corroborates Pirhofer-Walzl *et al.* (2011) who suggested that including forbs in ryegrass-clover mixtures

Table 1. Total annual dry matter (DM) yield (kg ha^{-1}) at three levels of evenness for two soil types (averaged over years 2016-2017).[1]

Item	Luvisols				Cambisols			
Evenness	L	M	H	Mixed	L	M	H	Mixed
Mean	10,952	15,263	13,078	14,171	8,774	11,619	11,467	11543
Values for comparing means of evenness	L vs M	L vs H	M vs H	L vs Mixed	L vs M	L vs H	M vs H	L vs Mixed
SED	1,091	1,024	1,134	825	1,132	1,062	1,176	855
t-value	3.95	2.08	1.93	3.90	2.51	2.54	0.13	3.24
P-value	***	*	ns	***	*	*	ns	**

[1] SED = standard error of a difference between two means. ns = non-significant, * P<0.05, ** P<0.01, *** P<0.01.

Table 2. Concentration of selected elements in the herbage (g kg^{-1} DM) depending on the species composition (averaged over years 2016-2017, harvests and soil types).

Treatment	Crude protein	WSC	Crude ash	Calcium	Magnesium
Mono *Lp*	151.3 abc	123.7 b	78.8 a	10.3 a	2.1 a
Mono *Tp*	169.4 bcde	73.6 a	78.7 a	20.0 e	2.8 c
Mono *Tr*	235.5 f	64.3 a	93.3 abc	15.9 bcde	2.3 ab
Mono *Ci*	135.2 a	69.9 a	125.5 d	19.1 de	2.8 bc
Bi *Lp-Ci*	134.7 a	88.6 a	100.1 bc	14.8 bcd	2.4 abc
Bi *Tp-Ci*	155.9 abc	60.9 a	110.3 cd	18.8 cde	2.8 bc
Bi *Tr-Ci*	194.5 e	59.4 a	112.4 cd	17.4 bcde	2.6 bc
Centroid	163.4 abcd	71.2 a	109.5 bcd	13.6 ab	2.4 abc
Domi *Lp*	139.2 ab	84.3 a	95.4 abc	13.8 ab	2.3 abc
Domi *Tp*	183.3 de	72.8 a	89.6 ab	13.5 ab	2.5 abc
Domi *Tr*	175.3 cde	63.3 a	95.7 abc	15.6 bcd	2.5 abc
Domi *Ci*	147.7 abc	80.5 a	95.9 abc	14.6 bc	2.4 abc

[1] Means with different lowercase letters are significantly different ($P<0.05$) using Tukey's *post hoc* test.

not only can enhance herbage production but also improve animal nutrition by providing sufficient dietary mineral supply to ruminants.

Conclusions

The above-ground biomass of two and four-species mixtures containing chicory were significantly higher than that obtained from monocultures. The herbage yield collected from leys with chicory in the sward was higher on Luvisols than on Cambisols. The herbage quality was affected by species diversity. We conclude that increasing species diversity by selecting appropriate grass-legume mixtures with inclusion of chicory adapted to specific soils enhances DM yield and herbage quality of leys in comparison to monocultures.

References

Cong W-F., Suter M., Lüscher A. and Eriksen J. (2018) Species interactions between forbs and grass-clover contribute to yield gains and weed suppression in forage grassland mixtures. *Agriculture, Ecosystems and Environment* 268, 154-161.

Finn J.A., Kirwan L., Connolly J., Sebastià M.T., Helgadóttir A., Baadshaug O.H., Bélanger G., Black A., Brophy C., Collins R.P., Čop J., Dalmannsdóttir S., Delgado I., Elgersma A., Fothergill M., Frankow-Lindgberg B.E., Ghesquiere A., Golińska B., Goliński P., Grieu P., Gustavsson A.M., Höglind M., Huguenin-Elie O., Jørgensen M., Kadžiuliene Z., Kurki P., Llurba R., Lunnan T., Porqueddu C., Suter M., Thumm U. and Lüscher A. (2013) Ecosystem function enhanced by combining four functional types of plant species in intensively managed grassland mixtures: a 3-year continental-scale field experiment. *Journal of Applied Ecology* 50, 2, 365-375.

Pirhofer-Walzl K., Søegaard K., Hogh-Jensen H., Eriksen J., Sanderson M.A., Rasmussen J. and Rasmussen J. (2011) Forage herbs improve mineral composition of grassland herbage. *Grass and Forage Science* 66, 415-423.

Ramseier D., Connolly J. and Bazzaz F. (2005) Carbon dioxide regime, species identity and influence of species initial abundance as determinants of change in stand biomass composition in five-species communities: an investigation using a simplex design and RGRD analysis. *Journal of Ecology* 93, 502-511.

Tilman D., Wedin D. and Knops J. (1996) Productivity and sustainability influenced by biodiversity in grassland ecosystems. *Nature* 379, 718-720.

Extensively managed grasslands: productivity after more than a decade without fertilizer input

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Abstract

With the progressing intensification of agriculture and the subsequent vanishing of pollinating insects, the importance of semi-natural grasslands as insect habitats is increasing. However, such grasslands are threatened by succession following abandonment due to lack of economic profitability. Therefore, we investigated the effect of stocking rate on pasture productivity in a long-term extensive grazing trial. We hypothesize that, due to the closed nutrient cycle, no reduction in productivity occurs in extensively managed grasslands. Stocking information and liveweights of Fleckvieh cows grazing in three different stocking rates based on a target sward height (M=6 cm, L=12 cm, VL=18 cm), recorded from 2005 to 2020, were used to calculate the metabolizable energy in GJ (ME_{total}) provided by the pasture and the animal grazing days in livestock units (LUGD, 1 LU = 500 kg), describing the stocking rate and annual grazing season duration. Linear mixed-effects modelling was performed, to test the effect of experimental design and year on ME_{total} and LUGD. The interaction of year and treatment significantly affected ME_{total} ($F=12.81$, $df=30$, $P<0.0001$) and LUGD ($F=16.85$, $df=30$, $P<0.0001$). The decrease in ME_{total} and LUGD from 2005 to 2020 might be caused by the redistribution of nutrients into less preferred grazing patches.

Keywords: sustainable extensification, extensive grassland, pasture productivity, grazing management, stocking rate, Fleckvieh

Introduction

With the progressing intensification of agriculture and the subsequent vanishing of biodiversity, the importance of extensively managed semi-natural grasslands as habitat is increasing. However, such grasslands are threatened by succession following abandonment due to their lack of economic profitability (Isselstein *et al.*, 2005). Nevertheless, extensively managed grasslands promote biodiversity targets and require less labour and input than intensively managed grassland. Therefore, maintaining extensive grasslands particularly for ruminant production could be a viable strategy to improve food security in the long term - without trade-offs with crop production for human consumption (Van Zanten *et al.*, 2018). However, few studies exist on the long-term effects of different stocking rates on livestock productivity on extensively managed grasslands. This study investigates the hypothesis that in extensive grazing systems pasture productivity in terms of livestock production remains constant in the long term.

Materials and methods

It is part of the long-term grazing experiment 'FORBIOBEN' (Isselstein *et al.*, 2007) and comprises the period from 2005 to 2020. The experimental setup is a one-factorial randomized block design with three replications comparing three stocking treatments, i.e. moderate (M), lenient (L), and very lenient (VL) stocking, on nine 1-ha paddocks. Paddocks were continuously stocked with Fleckvieh beef cows (pregnant, non-lactating) during the growing season from April to October, with a mid-summer resting period of varying length. The treatments are defined by stocking rates based on a target compressed sward height (CSH) of 6 cm for M, 12 cm for L, and 18 cm for VL. The CSH was measured biweekly in 50 locations per paddock using a rising plate meter (30 cm disk diameter, 200 g disk weight). Animals were removed from or added to a paddock when the mean CSH was below or exceeded the target, respectively (put-and-take system), and weighed regularly. A stocking minimum of two cows per paddock was required to meet animal welfare guidelines. The pasture productivity was assessed as secondary production in terms of livestock performance. Livestock unit grazing days (LUGD, where 1 LU = 500 kg LW) were calculated per paddock and year based on regular live weight (LW) measurements of the cows. Subsequently, requirements in metabolizable energy for maintenance and production (ME_{total}) in GJ ME ha^{-1} y^{-1} were calculated based on Baker (2004). Linear mixed-effects modeling was carried out, with year, stocking treatment, and their interaction, as well as block considered as fixed factors, and paddock as the random factor.

Results and discussion

The interaction of year and treatment significantly affected LUGD (F=16.85, df=30, P<0.0001). Estimated means varied significantly between stocking treatments M and VL in all years but usually did not vary significantly between M and L or L and VL (Figure 1A). As a general pattern, LUGD decreased from the first 3 to the last 3 years by a factor of 0.50, 0.47, and 0.53 in M, L, and VL, respectively. But no clear trend is visible. The ME_{total} was significantly affected by the interaction of year and treatment (F=12.81, df=30, P<0.0001) and decreased from the first 3 to the last 3 years by a factor of 0.48, 0.45, and 0.48 in M, L, and VL, respectively (Figure 1B). But no clear trend is visible. ME_{total} in M was always

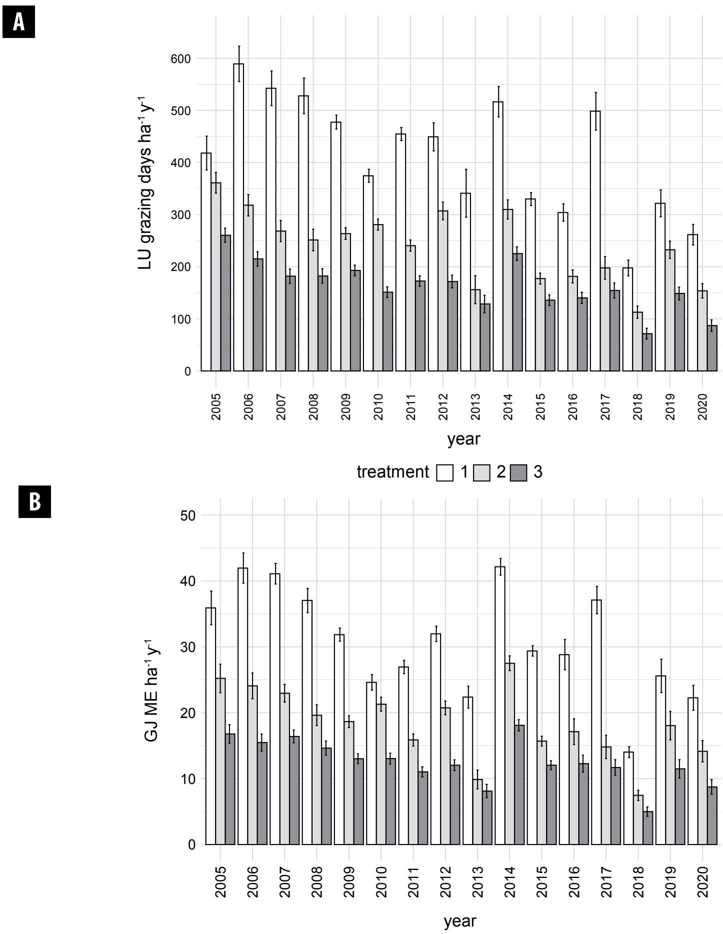

Figure 1. (A) Estimated mean ± SE of livestock unit grazing days (LUGD ha^{-1} y^{-1}) and (B) total metabolizable energy (GJ ME ha^{-1} y^{-1}) supplied by pastures in three stocking treatments moderate (M), lenient (L), and very lenient (VL) during 2005 to 2020.

significantly greater than in VL, and significantly larger than in L in most years. The difference in ME_{total} provided in L and VL was not significant in most years. With average stocking rates of 413, 238, and 164 LUGD ha^{-1} y^{-1} in M, L, and VL, respectively, all treatments should be considered as extensive grazing. However, as both ME_{total} and LUGD were mostly larger in M compared to L and VL, it can be concluded that the most intensive stocking in the present study led to and maintained the highest livestock productivity. Similarly, evaluations in the same experiment concluded that productivity and soil potassium concentration as well as the proportion of short patches, which showed the highest biodiversity, increased with stocking rate (Ebeling *et al.*, 2020; Tonn *et al.*, 2019). It is known that long-term extensive grazing causes a shift in the vegetation and increases heterogeneity as a consequence of herbage selection, creating patches with different vegetation. These patches differ in productivity (Ebeling *et al.*, 2020), soil nutrient concentration, and phytodiversity (Tonn *et al.*, 2019). We based our initial hypothesis on the assumption that grazed grasslands present closed nutrient cycles and, therefore, lead to maintained productivity. However, we found no compelling support for this hypothesis in the present study. Reasons to explain a decline in productivity, aside from climatic factors, could be the export of nutrients in the body tissue of cattle and nutrient redistribution within the pasture.

Conclusions

This study presents a unique investigation into animal performance in extensive grazing systems in temperate climates with varying stocking rates over a course of 16 years. Pasture productivity, based on livestock unit grazing days, and provision of metabolizable energy, varied throughout the study period and between stocking treatments.

References

Baker R.D. (2004) Estimating herbage intake from animal performance. In: Penning P. (ed.) *Herbage Intake Handbook*, British Grassland Society, Reading, UK, pp. 95-120.

Ebeling D., Tonn B. and Isselstein J. (2020) Primary productivity in patches of heterogeneous swards after 12 years of low-intensity cattle grazing. *Grass and Forage Science* 75, 398-408.

Isselstein J., Jeangros B. and Pavlu V. (2005) Agronomic aspects of extensive grassland farming and biodiversity management. In: Lillak R. *et al.* (eds.) *Grassland Science in Europe* 10*,* 211-220.

Isselstein J., Griffith B.A., Pradel P. and Venerus S. (2007) Effects of livestock breed and grazing intensity on biodiversity and production in grazing systems. 1. Nutritive value of herbage and livestock performance. *Grass and Forage Science 62*(2), 145-158.

Tonn B., Densing E.M., Gabler J. and Isselstein J. (2019) Grazing-induced patchiness, not grazing intensity, drives plant diversity in European low-input pastures. *Journal of Applied Ecology* 56(7), 1624-1636.

Van Zanten H.H.E., Herrero M., Van Hal O., Röös E., Muller A., Garnett T., ... and De Boer I.J.M. (2018) Defining a land boundary for sustainable livestock consumption. *Global Change Biology* 24(9), 4185-4194.

Mineral concentration in fractions of green forages after screw-pressing

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Abstract

The concentrations of minerals (K, Ca, Mg, P, Zn and Cu) were analysed in the fractions whole plant, pulp, green juice, green protein, and brown juice from biorefining of white clover, red clover, lucerne, and perennial ryegrass. Forages collected in July, September, and November were separated by screw-pressing and green protein was extracted from the green juice by heat precipitation. Following centrifugation, all samples (n=52) were analysed for minerals. The concentrations of K, Mg, and Cu were lower in pulp (10.0 g kg^{-1} of DM, 1.16 g kg^{-1} DM, and 4.2 mg kg^{-1} of DM, respectively) compared to the whole plant (22.6 g kg^{-1} of DM, 1.95 g kg^{-1} of DM, and 6.61 mg kg^{-1} of DM, respectively), whereas Zn and Cu in green protein (54.5 and 21.0 mg kg^{-1} of DM, respectively) were more than double the whole plant concentrations (25.9 and 6.61 mg kg^{-1} of DM, respectively). Especially green protein of perennial ryegrass had five times higher concentration of Cu than that of whole plant. Concentrations of K, Mg and P were higher in brown juice (48.9, 4.14, and 6.02 g kg^{-1} of DM, respectively) compared to the whole plant (22.6, 1.95, and 3.28 g kg^{-1} of DM, respectively), whereas the concentrations in green protein were similar to the whole plant.

Keywords: biorefining, mineral, pulp, green juice, green protein, brown juice

Introduction

From an environmental point of view, locally grown protein sources as alternatives to soybean meal are highly desired. In northern Europe, green forages such as grass, lucerne and clover give high yields of crude protein, but in order to utilize the protein efficiently in monogastric animals, soluble protein has to be up-concentrated and separated from the fibre fraction. This is done during a biorefining process using, e.g. a screw-press followed by protein precipitation and centrifugation of green juice to obtain a green protein concentrate. The process leaves a side-stream of pulp and brown-juice. Other compounds such as minerals might be recovered in each of the fractions depending on the solubility and binding properties of the individual mineral to, e.g. protein and fibre. Surplus amounts of minerals (e.g. P) might have negative environmental effects, whereas naturally occurring Zn in the green protein could be beneficial for animal health. Knowledge, however, on mineral content in the biorefining fractions is very limited and is needed in order to optimally utilize the different fractions. The aim of this study was to determine the concentration of minerals in whole plant, pulp, green juice, green protein, and brown juice after screw-pressing and protein precipitation of white clover, red clover, lucerne and perennial ryegrass.

Materials and methods

Forage samples, weighing 2-5 kg, of white clover (*Trifolium repens* L., var. Klondike and Silvester; WC), red clover (*Trifolium pratense* L., var. Rajah and 'Suez'; RC), lucerne (*Medicago sativa* L., var. Creno; LU), and perennial ryegrass (*Lolium perenne* L., var. Trocadero and Calvano 1; PR) were sampled (July, September, and November) from the experimental farm at Aarhus University, Foulum, Denmark, and immediately frozen at -20 °C until processing. Sampling and processing are described in detail in Damborg *et al.* (2020). The plant material was thawed overnight at 5 °C before being processed at room temperature in a lab-scale twin-screw press (82 rpm) without prior chopping. Pulp and green juice obtained from processing were stored at -20 °C until further processing or analysis. Thawed green juice was heated in a two-step procedure to precipitate protein. First the green juice was heated to a targeted 60 °C in a water bath and kept at that temperature for 20-30 seconds, after which it was cooled to 5 °C. The green juice was centrifuged (1,950×*g* for 10 min) and the pellet was defined as the green protein. The supernatant was heated again to 80 °C and kept at that temperature for 20-30 s before it was cooled to 5 °C and centrifuged (1,950×*g* for 10 min). The supernatant was defined as the brown juice. The pellet was defined as white protein, but the sample amount was too small for mineral analysis. The concentrations of K, Ca, Mg, P, Zn, and Cu were analysed by ICP in the fractions whole plant, pulp, green juice, green protein, and brown juice. The statistical analyses were conducted in R using a linear model, which included sampling time (n=3), forage type (n=4), fraction (n=5), and the interaction between forage type and fraction as fixed effects. Results are given as least square means and largest standard error of mean (SEM) is provided. Pairwise comparison of means was made using Tukey's test. Fractions of green protein and brown juice from November were not available due to limited sample material.

Results and discussion

Sampling time had no effect on mineral concentrations. The mineral composition of the four different forages (Table 1) reflected expected concentrations (NorFor, 2021). Across forages, K was higher ($P<0.01$ for both) in green and brown juice (48.5 and 48.9 g kg^{-1} DM, respectively) compared to the whole plant (22.6 g kg^{-1} DM), whereas no difference was found between green protein and whole plant, reflecting the high water solubility of this element. The concentration of Ca was more than twice as high ($P<0.01$) in LU compared to PR (16.1 vs 6.76 g kg^{-1} DM, respectively). The Ca is both fibre-bound and located in enzymes, which could explain why the concentration of Ca in individual fractions differed less from the whole plant compared to that of the other minerals. Mg is more water-soluble than Ca, which explains the higher ($P<0.01$ for both) concentrations found in green and brown juice (3.46 and 4.14 g kg^{-1} DM) relative to the whole plant across forages (1.95 g kg^{-1} DM). Moreover, Mg concentration in brown juice, especially for RC and PR, was 2.5 times higher ($P<0.01$ for both) than in the whole plant. In contrast, Mg concentration was 40% lower ($P<0.01$) in pulp (1.16 g kg^{-1} DM) compared to the whole plant across forages. Across forages, P is mainly present as either inorganic PO_4^{3-} or in phospholipids. The inorganic PO_4^{3-} follows the liquid phase, which could explain why the P concentration in green and brown juice (5.71 and 6.02 g kg^{-1} DM, respectively) was almost twice as high ($P<0.01$ for both) as in the whole plant (3.28 g kg^{-1} DM). The P concentration in green protein, which has a reactive high lipid content, was similar to that of the whole plant, probably because of the phospholipids found in the green protein. Zn is often associated to proteins as part of many enzymes, and can explain the more than twice as high Zn concentration ($P < 0.01$) in the green protein compared to the whole plant (54.5 vs 25.9 mg kg^{-1} DM). Cu is also a part of many enzymes and can explain the three times higher ($P<0.01$) concentration of Cu in green protein compared to the whole plant across species (21.0 vs 6.61 mg kg^{-1} DM, respectively). The exception was WC, where the Cu concentration in green protein was less different from that of the whole plant. In green protein of PR, the Cu concentration was five times higher ($P<0.01$) than that of the whole plant of PR (Table 1).

Conclusions

The investigation showed that biorefining of grass and legume forages resulted in differentiation in mineral composition between the different fractions. On DM basis, the concentration of all minerals analysed (K, Ca, Mg, P, Zn, and Cu) was either significantly or numerically lower in pulp compared to the whole plant, and for Zn and Cu, the concentration was higher in green protein compared to the whole plant. Brown juice had a lower concentration of Cu and a higher concentration of K, Mg, and P compared to the whole plant. Concentration of Ca was more equally distributed between fractions than the other minerals.

Table 1. Concentration of minerals in white clover, red clover, lucerne, and perennial ryegrass, and in the five fractions; whole plant, pulp, green juice, green protein, and brown juice, produced during the juice extraction and heat precipitation.[1]

Forage	Fraction	K	Ca	Mg	P	Zn	Cu
White clover	Whole plant	25.9	11.6	2.00[b]	3.30	21.8	6.26[b]
	Pulp	11.2	10.2	1.36[b]	2.20	17.4	4.11[b]
	Green juice	51.6	13.4	3.17[a]	5.49	30.7	11.0[a]
	Green protein	21.2	12.0	2.28[ab]	2.87	36.1	13.1[a]
	Brown juice	41.0	12.6	3.44[a]	5.12	24.6	5.99[b]
Red clover	Whole plant	18.7	12.3	2.24[cd]	2.78	30.9	8.43[c]
	Pulp	8.32	10.3	1.24[d]	1.73	25.9	5.93[cd]
	Green juice	44.7	15.6	3.67[b]	4.62	41.4	13.0[b]
	Green protein	17.2	12.5	2.44[c]	2.33	65.0	22.0[a]
	Brown juice	57.6	17.4	5.43[a]	5.62	48.2	3.38[d]
Lucerne	Whole plant	17.3	15.5	1.82[cd]	3.07	24.3	6.37[c]
	Pulp	8.77	12.7	1.06[d]	2.13	18.7	3.89[cd]
	Green juice	40.8	22.7	3.78[a]	5.21	37.4	12.9[b]
	Green protein	20.8	16.2	2.26[bc]	4.17	64.6	22.2[a]
	Brown juice	31.1	13.4	3.16[ab]	3.55	21.6	0.298[d]
Perennial ryegrass	Whole plant	28.5	5.00	1.75[cd]	3.98	26.6	5.39[c]
	Pulp	11.7	3.74	0.988[d]	2.35	16.2	2.73[c]
	Green juice	56.8	6.96	3.23[b]	7.52	34.4	11.0[b]
	Green protein	31.2	9.05	2.78[bc]	5.69	52.2	26.6[a]
	Brown juice	65.8	9.06	4.56[a]	9.78	53.3	2.46[c]
SEM[2]		7.42	1.985	0.312	0.925	7.46	1.261
P-value	Forage	0.02	<0.01	0.03	<0.01	0.01	0.02
	Fraction	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
	Forage × Fraction	0.57	0.18	0.01	0.14	0.24	<0.01

[1] The values for K, Ca, Mg, and P are given as g kg^{-1} DM, and the values for Zn and Cu are given as mg kg^{-1} DM. Values within each column and forage with different subscript letters differ ($P<0.05$).

[2] Largest standard error of mean.

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References

Damborg V.K., Jensen S.K., Weisbjerg M.R., Adamsen A.P. and Stødkilde L. (2020) Screw-pressed fractions from green forages as animal feed: chemical composition and mass balances. *Animal Feed Science and Technology* 261: 114401.

NorFor (2021) Feed table. Available at: http://www.norfor.info/feed-table/.

Nitrate concentrations of leachate collected from perennial ryegrass and plantain monocultures after dairy cow urination

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Abstract

Nitrate losses from agricultural systems continue to be a significant issue in Europe, particularly on ruminant grazing focused farms. Recently, studies have indicated that, compared to perennial ryegrass (PRG, *Lolium perenne* L.), ribwort plantain (PL, *Plantago lanceolata* L.) can reduce the levels of nitrate leached from soil horizons in established swards (>18 months), particularly during high rainfall seasons. A lysimeter study was established in Ireland to investigate whether such effects are seen across differing soil types within one year of sward establishment. Monoculture swards of PL and PRG were established in fallow lysimeters in May 2020; 4 replicates of each were sown to both free-draining (FD) and poorly-draining (PD) soil types. Urine collected from lactating dairy cows was applied to each lysimeter in October 2020. Leachate was collected from each lysimeter at two-week intervals until March 2021. Data analysis was conducted using SAS 9.4. Leachate from the FD soil had a higher nitrate nitrogen (P<0.001) concentration for both species over the collection period; no difference in leachate nitrate nitrogen concentration was found between plant species within soil type. These results indicate little influence of plant species on soil nitrate leaching in the establishment year.

Keywords: nitrate loss, reduction, soil type, *Lolium perenne, Plantago lanceolata*

Introduction

Nitrate (NO_3) losses from agricultural systems are a significant issue in Europe, as they represent a significant source of groundwater and waterway pollution. This is of particular importance in grazing-focused grassland systems, where NO_3 leaching is problematic due to losses of soil nutrients from the farm system. This includes the Irish dairy sector which, similar to New Zealand (NZ), places a large emphasis on utilizing fresh pasture in the dairy cow's diet. While such systems are shown to be economically and environmentally efficient, they rely upon high nitrogen-(N) input perennial ryegrass pastures (PRG, *Lolium perenne* L.) for a majority of the herbage supply. Furthermore, NO_3 losses from dairy cow urine patches represent a significant point source of pollution with an N load of 500-1000 kg N ha^{-1} (Cameron *et al.*, 2013). Data on actual NO_3 losses from Irish grassland systems are limited although it has been shown that soil water from grazing systems can often breach maximum allowable concentrations of NO_3 (Ryan *et al.*, 2006); soil type can also influence the level of NO_3 in soil water (Creamer and O'Sullivan, 2018). Agriculture has previously been shown to be responsible for 90% of aquatic N loading in Ireland and, more recently, the intensification of Irish dairy grazing systems may contribute to greater pollution pressure on Irish waterways (O'Boyle *et al.*, 2017). Studies from NZ have shown swards of ribwort plantain (PL, *Plantago lanceolata* L.) to be an effective strategy in reducing soil NO_3 losses with a reduction of up to 50% in leachate water NO_3 concentrations when compared with PRG swards under similar conditions (Welten *et al.*, 2019). The objective of the current study is to investigate the effectiveness of PL to reduce NO_3 leaching from two different soil types over the winter period in Ireland within one year post sowing.

Materials and methods

A lysimeter study was established at Teagasc Moorepark (52°16'N, 8°26'W) in May 2020. Sixteen lysimeters (0.07 m^2 diameter, 0.7 m depth) were sown with either PRG (cv. AberChoice) or PL (cv. Tonic) monocultures. Of the sixteen total lysimeters eight were of a Cambisol (free draining; FD) soil type and eight were of a Gleysol (poorly draining; PD) soil type. The species were distributed evenly between soil types to give four replicates of a 2×2 factorial designed experiment. Chemical fertilizer (44.3 kg N ha^{-1}) was applied to all lysimeters post sowing to aid establishment in June and July of 2020 with no fertilizer applied thereafter. Urine was collected from cows grazing PRG and white clover (*Trifolium repens* L.) pasture on 5 October and analysed for N content. In order to mimic a typical dairy cow urination event 0.7 l of urine was applied to each lysimeter on 12 October, providing a total N loading of 4888 mg N/ lysimeter or 698 kg N ha^{-1}. Background levels of NO_3 from soil leachate water were determined prior to urine application and used as a control within the trial. Leachate was collected in 10 l vessels and volume was measured fortnightly until March 2021 when NO_3 concentrations returned to background levels. Sub-samples of leachate were analysed for N contents within 48 h of collection; total oxidized nitrogen (TON) and nitrite (NO_2) concentrations of leachate were measured spectrophotometrically and NO_3 levels were determined by subtracting NO_2 from TON. Total NO_3 leached and peak NO_3 concentrations of leachate were analysed using a mixed model in SAS 9.4 where plant species, soil type and associated interactions were included as fixed effects; repetition was a random variable.

Results and discussion

Total rainfall over the trial period was 671 mm, which was over 100 mm more than the 30-year average (1981-2010) for the site. The volume of leachate drained (38.0 l average) was similar across all treatments. Soil type was associated with total NO_3 loss; when averaged across both plant species total NO_3 lost in leachate from the PD and FD soil types was 148.3 and 290.0 mg, respectively; peak NO_3 concentration in leachate from PD soil was 36% of that recorded from the FD soil type (Figure 1). Soil type can have an impact on NO_3 leaching and aerobic soils, such as Cambisol, are most susceptible to NO_3 leaching (Creamer and O'Sullivan, 2018). There was no association between plant species and NO_3 collected in leachate. Such results disagree with those of Welten *et al.* (2019) who found large differences in NO_3 concentrations of leachate collected from lysimeters containing either PL or PRG. However, the current study involved plants established <1 year prior to urine application whereas previous trials generally used

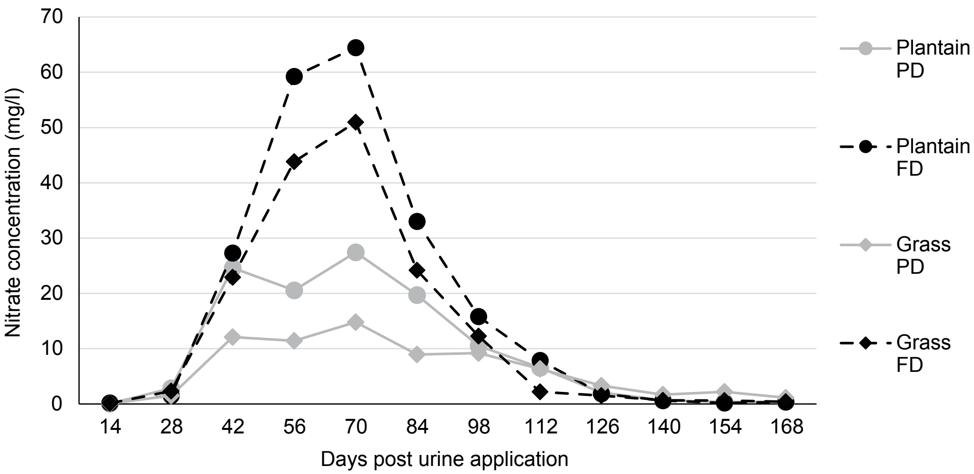

Figure 1. Nitrate concentration (mg/l) of leachate from free and poorly drained soil type and perennial ryegrass and plantain monoculture lysimeters at 14-day intervals post urine application.

swards >1.5 years old at time of urine application (Welten *et al.*, 2019, Carlton *et al.*, 2019). Greater rooting depth and increased winter growth are the mechanisms by which PL can utilize more N in soil and hence reduce N losses; plant N uptake data are not available for the current analysis but lack of root development within the shorter time period of the current study may have reduced the capacity of PL to utilize N as it leached through the soil horizons. In agreement with the current work, a mesocosm study on the effects of PL and PRG on soil and leachate NO_3 concentrations found no association between PL and NO_3 up to day 148 post establishment (Pijlman *et al.*, 2020). Such results pose an issue for the use of PL as a nitrate leaching mitigation strategy in newly established swards, as there are persistency issues with herbs, such as PL, in mixed-species sward grazing systems (Lee *et al.*, 2015). The current study will continue to analyse the effects of PL and soil type on NO_3 leaching through subsequent seasons and urine application events.

Conclusions

Within the first year post establishment PL was not associated with either total NO_3 leached or the peak NO_3 concentrations in leachate following a dairy cow urination event during a high rainfall season in Ireland. This may be an issue if PL were to be deployed to mitigate NO_3 leaching as the first year post establishment could represent a significant proportion of the lifetime of the plant within a sward. Further work is required to determine lifetime persistence of PL under grazing and whether or not there is a leaching mitigation potential for PL post the establishment year and the factors that influence such mitigation.

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References

Cameron K., Di H.J. and Moir J. (2013) Nitrogen losses from the soil/plant system: a review. *Annals of Applied Biology* 162, 145-173.

Carlton A.J., Cameron K.C., Di H.J., Edwards G.R. and Clough T.J. (2019) Nitrate leaching losses are lower from ryegrass/white clover forages containing plantain than from ryegrass/white clover forages under different irrigation. *New Zealand Journal of Agricultural Research* 62, 150-172.

Creamer R. and O'Sullivan L. (2018) *The soils of Ireland*, Springer, Berlin, Germany.

Lee J.M., Hemmingson N.R., Minnee E.M. and Clark C.E. (2015) Management strategies for chicory (*Cichorium intybus*) and plantain (*Plantago lanceolata*): impact on dry matter yield, nutritive characteristics and plant density. *Crop and Pasture Science* 66, 168-183.

O'Boyle S., Wilkes R., McDermott G. and Longphuirt S.N. (2017) Will recent improvements in estuarine water quality in Ireland be compromised by plans for increased agricultural production? A case study of the Blackwater estuary in southern Ireland. *Ocean & Coastal Management* 143, 87-95.

Pijlman J., Berger S.J., Lexmond F., Bloem J., Van Groenigen J.W., Visser E.J., Erisman J.W. and Van Eekeren N. (2020) Can the presence of plantain (*Plantago lanceolata* L.) improve nitrogen cycling of dairy grassland systems on peat soils? *New Zealand Journal of Agricultural Research* 63, 106-122.

Ryan M., Brophy C., Connolly J., McNamara K. and Carton O.T. (2006) Monitoring of nitrogen leaching on a dairy farm during four drainage seasons. *Irish Journal of Agricultural and Food Research* 45, 115-134.

Welten B.G., Ledgard S.F., Judge A.A., Sprosen M.S., McGowan A.W. and Dexter M.M. (2019) Efficacy of different temperate pasture species to reduce nitrogen leaching from cattle urine applied in different seasons: a soil lysimeter study. *Soil Use and Management* 35, 653-663.

Analysis of the nutritive value of various *Festulolium* hybrids assessed at different harvesting times

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Abstract

Festulolium hybrid grasses are relatively new components of grassland management practices in Hungary as well as in the whole of Europe. Due to scarcity of information concerning the nutritive value of these hybrids, a substantially important parameter in the evaluation of grass species/varieties is lacking. To address this issue, a study was embarked upon to compare the feeding value of F_1-hybrids with that of Italian ryegrass (*Lolium multiflorum*) and tall fescue (*Festuca arundinacea)*. For assessing the nutritive value index of the harvested grass, FT-NIR analyser was used. In light of our findings it can be stated that the harvesting time and the varieties had a significant impact on the bulk of the analysed parameters such as: dry matter (DM) yield, DM, crude protein, crude fibre, aNDFom, acid detergent fibre, acid detergent lignin (ADL), $dNDF_{om48}$, content, DM yield and digestibility data (OMd, $aNDF_{omd48}$). The interaction between harvesting time and varieties was found to have a statistically significant effect on DM, ADL and $aNDF_{omd48}$. Our findings may contribute to optimize the forage supply of ruminants in the Central/Eastern-European region.

Keywords: *Festulolium*, harvesting time, nutrient content

Introduction

Viewed globally, *Festulolium* hybrids are being grown increasingly to supply ruminants with feed. These hybrids are formed from crossing *Lolium* and *Festuca* species. They are characterized by palatability, higher yields, improved drought tolerance and better digestibility. *Festulolium* hybrids possess excellent biotic and abiotic stress tolerance and may therefore represent a good source for improving the performance of prospective genetic stocks (Ghesquière *et al.*, 2010). Due to global climate change there is an increasing need to base crop production on plants with higher drought tolerance so that superseding silage maize and other fodder plants, which currently provide the basis for feeding ruminants, can become possible. According to Ghesquière (1996), in addition to their high productivity, these grasses have an excellent forage value and highly acceptable. In ensuring grass-based forage supply, *Festulolium* species hybrids are promising. During our experiments, the quality of the high-yield stages with respect to the preparation of haylage was investigated.

Materials and methods

The site of the experiments was the Institute of Forage Production, HUALS (46°41'03'N; 18°11'00'). The soil type of the experimental area is limestone chernozem. In the experiment an Italian ryegrass (IR; *Lolium multiflorum*), a tall fescue (TF; *Festuca arundinacea*) and four different *Festulolium* hybrids (Fh) were tested, two of which were *Lolium* type (Fh1 and Fh2) and the other two were *Festuca* type (Fh3 and Fh4). The number of replicates was four. Following soil preparation and fertilization (300 kg ha^{-1} N:P:K 4:17:30; 150 kg ha^{-1} CAN 27), the seeds were sown in 1.5-45 m blocks on 14 September 2017. The plantation received CAN 27 fertilizer at a dose of 200 kg ha^{-1} during the spring season on 12 March 2018. The seed dose was adjusted to 40 kg ha^{-1}, and the plants were not watered throughout the experiment. The accumulated precipitation was 91.4 mm until harvest, which represented a relatively dry spring. The dry matter yield was assessed to determine the optimal harvesting time. Sampling was performed with a 1 m^2 dummy frame randomly placed at 4 replicates at each of 3 sampling times. Samplings were effectuated on 26 April, 4 May and 11 May. Mowing times and phenological phases were as follows: BBCH 34 (Node 4^{th} at least 2 cm above node 3^{rd}), 26 April; BBCH 41 (Early boot stage: flag leaf sheath extending), 04 May; BBCH 49 (First awns visible, in awned forms only, 05 May; BBCH 59 (End of heading: inflorescence fully emerged). The range of the stubble height was 8 to 10 cm. Subsequently, for the assessment of the nutritive value index of the harvested grass, a Q-Interline Quant FT-NIR analyser was used (dataset developed by the BLGG AgroXpertus, Wageningen, the Netherlands). The parameters examined were: dry matter (DM), crude protein (CP), crude fibre (CF), amylase treated ash corrected neutral detergent fibre (aNDFom), acid detergent fibre (ADF), acid detergent lignin (ADL), digestible organic matter (OMd_{48}), 48 h *in vitro* digestibility of aNDFom ($aNDFomd_{48}$) and determined digestible aNDFom by 48 h *in vitro* incubation ($daNDFom_{48}$). The results were evaluated by two-factorial variance analysis. Variety effects and differences between variants at each sampling time point were determined by one-way variance analysis.

Results and discussion

Based on the results of the two-factorial analysis of variance, the effect of cultivar/hybrid and mowing time was significant for all presented nutritive parameters ($P\leq0.001$). This finding IS in accordance with our previous results (Hoffmann *et al.*, 2020) which revealed the effect of hybrid and mowing time being significant for the green and dry matter yield as well as the estimated silage yield ($P\leq0.001$). The combined effect of cultivar and time ($P\leq0.05$) proved significant for the dry matter yield. Significant differences were observed in DM yield at each mowing time (Table 1). Based on our results, the parameters examined differed significantly at each cutting time *(*$P\leq0.05$) with the exception of NDF and ADF. Within each mowing time point, the Fh2 *Lolium*-type hybrid had the significantly lowest DM content and that of the *Festuca*-type Fh4 was significantly the highest. These findings were observed at all three cutting times. In terms of CP content, the Fh 3 *Lolium*-type hybrid had significantly higher CP content at the first cutting as compared to the other treatments. Only that of tall fescue did not differ significantly. At subsequent mowing times, the protein content decreased substantially, albeit Fh3 still had the highest protein content. During the experiment, the CF content varied between 250-320 g kg^{-1}. Fh4 had the lowest crude fibre content in the first two cuts. The crude fibre content of IR changed little. The content of IR NDF hardly changed as a function of cutting time. The increase was greater between the first and second mowing times, while the difference between the 2^{nd} and 3^{rd} cuts was minimal. There was no significant difference in NDF and ADF content between the different (harvest time). In the second and third cutting, the difference between Fh4 and tall fescue was significant. Regarding ADL, similarly to TF, Fh3 had the highest value in all three cuts, whereas IR and *Lolium*-type Fh1 had the lowest ADL values. *Festulolium* hybrids showed the best OMd values, while TF possessed the weakest one. The aND_{Fomd48}, and $daND_{Fom48}$ were best for *Festulolium* and TF; IR was the weakest performing in this respect.

Conclusions

Based on our previous results (Hoffmann *et al.*, 2020), in addition to IR, the cultivation of Fh2 and Fh3 *Lolium*-type hybrids seems the most promising due to their higher dry matter yield. TF cultivated separately, but also combined with other species, is recommended for grass mixtures. The first cutting time may be a good choice, especially based on the ADL values obtained, while the second and third cutting times appear to be more suitable for IR Fh1 and Fh2 mixtures. It can be established that the highest feeding value should be taken into account to determine the appropriate cutting time (Hoffmann *et al.*, 2020). Thus, the first mowing appears to be ideal from nutritional point of view, whereas if the highest yield is to be attained, the third cutting seems expedient. Therefore, in view of satiating the high nutritive value requirement of forage given to high-yielding milking dairy cows, the first cutting is suggested while the second mowing is proposed for cows with lower milk production, beef or sheep, leading to apparently a good compromise.

Table 1. Dry matter yield, dry matter content and nutritive parameters of different grass varieties at different harvest dates.[1]

Sampling dates	Hp	DM yield kg ha^{-1}	DM g kg^{-1}	CP g kg^{-1}	CF g kg^{-1}	NDF g kg^{-1}	ADF g kg^{-1}	ADL g kg^{-1}	OMd48 %	NDFd48 %	dNDF48 g kg^{-1}
Italian ryegrass IR	1	6.37a	161.9a	135.3a	264.7a	536.0a	285.0a	20.3a	79.2a	71.8a	385.0a
Festulolium Fh1	1	5.55b	145.4a	161.3ab	261.7a	539.3a	286.0a	18.7a	81.8b	77.9bc	419.7bc
Festulolium Fh2	1	5.39b	125.3b	181.7bc	268.7a	555.3a	291.3a	19.0a	82.7b	78.2b	434.0b
Festulolium Fh3	1	4.44c	151.5a	223.0c	269.0a	564.3a	288.3a	25.3b	77.9a	75.6c	426.7b
Festulolium Fh4	1	2.38d	200.8c	128.0ad	256.5a	527.5a	283.5a	20.5ab	78.9a	74.1ac	390.5ac
Tall fescue TF	1	5.30b	165.9a	190.7bc	268.0a	560.3a	302.0a	23.0ab	78.4a	76.2bc	427.3b
Standard error		0.2402	9.8877	19.612	9.2414	14.161	10.662	2.3671	1.0801	1.1467	12.528
Italian ryegrass IR	2	7.80a	193.1a	103.0a	288.3ab	562.3ab	313.0ab	25.3a	73.6ab	60.4a	339.7a
Festulolium Fh1	2	7.31a	169.6b	127.7ab	289.3ab	578.3ab	317.7ab	27.0ab	74.8ab	65.1b	376.3bd
Festulolium Fh2	2	6.98ab	150.3c	148.0bc	283.0a	575.7ab	310.7ab	24.3a	76.8a	68.2b	392.7bc
Festulolium Fh3	2	6.17b	185.2a	161.3c	294.7ab	594.3a	314.7ab	31.7bc	73.1b	68.0b	404.3c
Festulolium Fh4	2	3.35c	212.2d	116.0a	277.3a	543.0b	298.0a	25.0a	75.1ab	67.0b	363.0d
Tall fescue TF	2	5.56c	170.2b	170.0c	307.7b	589.3a	335.3b	28.3c	71.8b	65.3b	383.3b
Standard error		0.3813	6.4164	11.9567	11.1687	18.6805	15.5646	2.3333	1.6218	1.9857	8.1012
Italian ryegrass IR	3	6.68a	232.9a	85.0a	282.7a	554.7a	303.7a	28.7a	72.6ab	57.6a	319.7a
Festulolium Fh1	3	6.67a	188.0b	115.0b	301.7bc	585.3bc	328.3bc	28.3a	71.7ac	59.7ac	349.3b
Festulolium Fh2	3	5.60b	168.8b	145.0c	291.3ab	587.0bcd	314.7ab	29.3a	73.9b	63.0bd	370.0b
Festulolium Fh3	3	4.29c	243.3a	150.3c	302.0bc	606.3bd	331.0bc	36.0b	70.4c	61.1bc	370.0b
Festulolium Fh4	3	2.98d	228.5a	113.7ab	293.0ab	570.0ac	319.3ab	27.3a	73.8b	64.1d	365.7b
Tall fescue TF	3	5.13bc	222.2a	106.7ab	318.0c	609.3d	349.0c	34.0a	70.5b	59.7ac	364.0b
Standard error		0.4681	11.3563	13.2665	8.1377	10.7772	9.6282	1.8459	0.8041	1.3192	10.2523

[1] Hp = Harvesting period; Hp 1 is 26 April; 2 is 4 May; 3 is 11 May. Standard errors refer to the columns. Values within each column with different subscript letters differ ($P \leq 0.05$).

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References

Ghesquière M., Emile J.C., JadasHécart J., Mousset C., Traineau R. and Poisson C. (1996) First *in vivo* assessment of feeding value of Festulolium hybrids derived from Festuca arundinacea var. glaucescens and selection for palatability. *Plant Breeding* 115(4), 238-244.

Ghesquière M., Humphreys M. and Zwierzykowski Z. (2010) Festulolium hybrids: results, limits and prospects. In: *Sustainable use of genetic diversity in forage and turf breeding.* Springer, Dordrecht, the Netherlands, pp. 495-507.

Hoffmann R., Pal-Fam F., Keszthelyi S. and Halasz A. (2020) Yield of Festulolium hybrids at different phenological stages during primary growth *Proceedings of the 28th General Meeting of the European Grassland Federation* (*Grassland Science in Europe* 25, pp. 82-84.

Kopecký D., Loureiro J., Zwierzykowski Z., Ghesquière M. and Doležel J. (2006) Genome constitution and evolution in Lolium× Festuca hybrid cultivars (Festulolium). *Theoretical and Applied Genetics* 113(4), 731-742.

Effect of grazing method on grass quality change during a 24-hour period

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Abstract

Changes in grass quality during day and night can be relevant for both research and practice. In research a representative sample of the provided grass is essential for reliable results, and for practice it can affect management decisions. In a zero-grazing system, grass is brought to the barn two to three times a day and afterwards fed in small portions which might lead to a decay of grass quality. In a pasture, fresh grass quality can change during the day and night under the influence of sunlight and selection by the cows. In a grazing trial on the Dairy Campus, it was examined how grass quality was affected over time. In the zero-grazing system, a sample was taken on 7 occasions during a 24-h period to assess the quality. In the 24-h grazing system, fresh grass samples were taken from the same plot around 8 am and 6 pm. The samples were analysed on feed quality. It appeared that mowing fresh grass twice a day and providing it in small portions over the day did not result in a significant change in feed quality. During grazing, it was found that in the evening the sugar content was higher and the crude protein content lower. There was no significant difference in the other components. These results show that time of sampling fresh grass in pasture needs to be taken into account in grazing research.

Keywords: grass quality, nutrient value, fresh grass, daytime, grazing, zero grazing

Introduction

In an ongoing multi-annual trial at research innovation centre Dairy Campus (Leeuwarden, the Netherlands), chemical composition of fresh grass over time was examined. Part of the experiment was a comparison of two treatment groups: a 24-h grazing system and feeding 100% fresh grass in the stable (zero grazing). The trial was carried out in three periods of two weeks (May, July and September). The chemical composition of grass may change during the day (and night) as a result of grazing and the day-night rhythm (Abrahamse *et al.*, 2009). Additionally, in a zero-grazing system grass quality might change as a result of storing it on the feeding alley (Melvin and Simpson, 1963). The effects of day and night grazing (selection) and zero-grazing on chemical composition were examined in this study.

Materials and method

The fresh grass samples were taken for the objects 100% grazing (W) and 100% fresh grass in the barn (zero-grazing, Z) on 14 consecutive days and during three periods (May, July and September) in 2020 and 2021. In the object W, the cows grazed daily on a strip of fresh grass. Fresh grass samples were taken at 8 am and 6 pm from the field grazed at that day. For Z in the same period, fresh grass was mowed twice daily (around 8 am and 4 pm) and unloaded into the feeding alley. From this grass stock, the roughage intake control (RIC) bins were filled 8 times a day and fresh grass samples were taken every refill. All samples were examined with the NIRS method for crude protein (CP), crude fibre (CF), crude ash, total sugar, digestibility coefficient organic matter (DC-OM) and neutral detergent fibre (NDF). The data from object W were statistically analysed using ANOVA, with moment (morning or evening) and period as explanatory variables, and day within a period as block effect. The data from object Z were analysed with REML to estimate the period effect, with date as random factor and moment of mowing as explanatory (fixed) factor. A distinction was made between the grass that was mowed in the morning and the grass that was mowed in the afternoon, because there may be a difference in the starting situation. A regression analysis was used to estimate the trend over the morning and afternoon period separate.

Results

Table 1 shows that there was a significant effect of period when comparing the fresh grass samples from 100% grazing ($P<0.001$), but not in the same way for all components. The digestibility and sugar content were highest in spring, while NDF, CP and CF were the lowest. There was a significant difference ($P<0.001$) between morning and afternoon samples for CP, sugar and NDF. In all periods NDF and CP were the highest in the morning, while sugar was highest in the afternoon. Only sugar ($P<0.001$) and CP ($P=0.005$) showed a significant interaction effect between period and moment; in July the differences between morning and evening were the smallest (Table 1). For the Z-object, there was a significant period effect ($P<0.001$) for all parameters examined. The nutritional value is dependent on time during the grazing season. The regression analysis only showed a significant trend over time for sugar content, which increased around the second mowing time ($P=0.05$). Figure 1 shows the development in sugar content for the 8 feeding moments (time 4 or 5 afternoon mowing) and the three periods for the Z-object.

Discussion

For fresh grass in the pasture, the contents of sugar, CP and NDF appeared to differ between the morning and the evening, which is in line with Smit and Elgersma (2004). This is probably caused by two components that cannot be separated in this analysis: change in composition under the influence of day-night rhythm and change due to selective grazing (not all of the allowed grass was taken up at the end of the day). During the day, sunlight initiates photosynthesis, resulting in an increase in the sugar

Table 1. Differences in nutrient value of fresh grass between morning and evening samples.[1]

Period	May		July		September		Overall		LSD
Moment	am	pm	am	pm	am	pm	am	pm	
VC_OM	86.1	85.6	81.1	76.9	81.3	81.2	82.8 ns	81.2 ns	3.61
CP	197	147	201	168	232	185	210 ^{b***}	167 ^{a***}	14.76
CF	195	193	235	232	225	221	219 ns	215 ns	9.86
Sugar	161	229	99	139	73	132	111 ^{a***}	167 ^{b***}	17.1
NDF	508	466	572	538	571	528	550 ^{b***}	511 ^{a***}	13.69

[1] Overall values with different subscript letters differ between pm and am. ns = not significant, *** $P<0.001$.

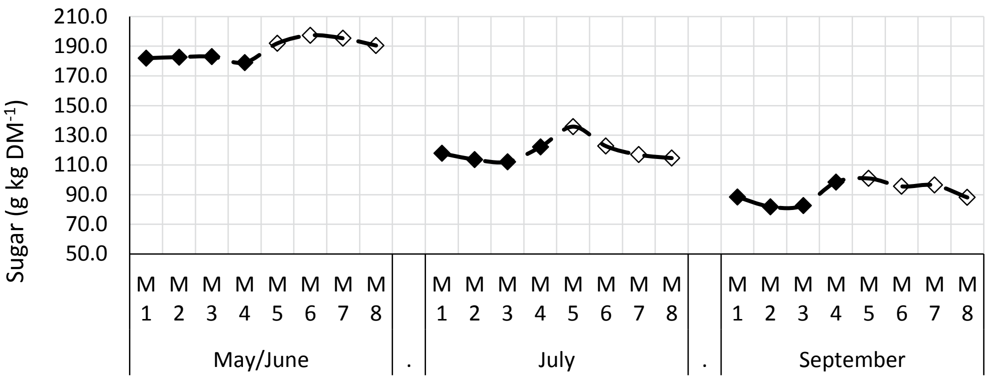

Figure 1. Effect of storage time on the feeding alley (M 1-8, $P<0.001$, LSD=3.99) on sugar content of fresh grass mown in two batches over three periods (May/June, July and September, $P<0.001$, LSD=14.3). Closed dots mown at 8 a.m. open dots mown at 4 pm.

content in the grass and a lower CP content. However, this is not expected to affect animal performance (Gibbs *et al.*, 1998). No clear significant trend in change of feed value was observed during the storage of grass in the feeding alley after it was mowed in the morning or in the afternoon, except for a slight decrease in sugar content. Similar results were found in other drying experiments (Melvin and Simpson, 1963). With two moments of mowing a day and approximately 10 h of storage time there was no decline in nutritional value.

Conclusions

A different feed quality can be obtained depending on the (starting) time of grazing (morning or evening) and on the moment of sampling. This can be relevant for grazing experiments, for example to explain the effect of grass quality on methane excretion, although the effect on animal performance is not expected to be significant. Nevertheless, the moment of sampling needs to be taken into account in order to collect representative grass samples. Results of this study indicate that the temporary storage of grass after mowing has no negative consequences for the nutritional value if the storage time does not exceed approximately 10 h.

References

Abrahamse P.A., Tamminga S. and Dijkstra J. (2009) Effect of daily movement of dairy cattle to fresh grass in morning or afternoon on intake, grazing behaviour, rumen fermentation and milk production. *Journal of Agricultural Science* 147(6), 721-730.

Gibb M.J., Huckle C.A. and Nuthall R. (1998) Effect of time of day on grazing behaviour by lactating dairy cows. *Grass and Forage Science* 53, 41-16.

Melvin J.F. and Simpson B. (1963). Chemical changes and respiratory drift during the air drying of ryegrass. *Journal of the Science of Food and Agriculture* 14(4), 228-234.

Smit H.J. and Elgersma A. (2004) Diurnal fluctuations in vertical distribution of chemical composition in a perennial ryegrass (*Lolium perenne* L.) sward during the season. *Grassland Science in Europe* 9, 951-953.

Presumed yield benefit of grassland renewal is offset by loss of soil quality

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Abstract

An important motivation for farmers to renew grassland swards (by ploughing and reseeding) is to introduce the most recent grass cultivars that give high yields and high forage quality. However, grassland renewal may affect soil quality negatively due to ploughing. The aims of this study were (1) to compare grass productivity and soil chemical quality of young and old grasslands, and (2) to investigate the relation between soil chemical quality and grass productivity. On clay soil in the north of the Netherlands we measured grass productivity and soil parameters of 'young' (5-15 years without grassland renewal) and 'old' (>15 years without grassland renewal) grasslands, located as pairs at ten different dairy farms. We selected grasslands with at least 70% desirable grasses (i.e. *Lolium perenne* and *Phleum pratense*). We found a lower herbage nitrogen (N) yield in young grassland and no significant difference compared with old grassland in terms of herbage dry matter yield and fertilizer N response. The soil of young grassland contained less soil organic matter (SOM), carbon (C-total) and nitrogen (N-total) compared to the old grassland soil. Grass productivity was positively correlated with SOM, N-total and C-total. The current management practice of renewing grassland after 10 years without considering the botanical composition is counter-productive.

Keywords: grass productivity, nitrogen, permanent grassland, soil organic matter

Introduction

On clay soils in the Netherlands, permanent grasslands are renewed (i.e. destroyed by herbicides, ploughed and reseeded) on average once every 10 years (Russchen, 2005). An important motivation for grassland renewal is to introduce the most recently developed perennial ryegrass varieties (*Lolium perenne*) to increase feed production and quality. Plant breeding programmes have accomplished a yield increase of 3% per decade plus enhanced digestibility, and many studies have shown that, with this increase, reseeding is economically attractive for farmers (Chaves *et al.*, 2009; Sampoux *et al.*, 2011). However, these studies are based on comparison of older grass varieties with new varieties sown at the same time on the same field. Possible effects of loss of soil quality due to ploughing have not been taken into account. The main objective of this study was to compare grass productivity and soil chemical quality of young (5-15 years) with old (>15 years) permanent grassland on clay soil.

Materials and methods

The study was conducted in 2014 at ten conventional dairy farms on marine clay soil in the north of the Netherlands. At each farm a young and an old grassland were selected based on the following criteria: seeded with the most recently developed commercially available perennial ryegrass varieties at time of renewal, no clover seeded, with no visual soil compaction, and at the time of sampling containing at least 70% desirable grasses (i.e. *Lolium perenne* and *Phleum pratense*). On each grassland, an experimental plot of 15×9 m was laid out with three 10×3 m sub-plots fertilized with 0, 150 or 300 kg N ha^{-1} yr^{-1}. The remaining 5×9 m sub-plot was not fertilized and was used to measure SOM, N-total and C-total in April 2014. Grass was harvested four times in 2014, weighed and sampled for dry matter (DM) and total N analysis. See Iepema *et al.* (2020) for details on the experimental lay-out, measurements and statistical analyses.

Results and discussion

The N yield without N fertilization (NY_{N0}) was significantly lower in the young grasslands compared to the old grasslands. The other grassland productivity parameters, i.e. grassland dry matter yield without nitrogen fertilization (DMY_{N0}), grassland dry matter yield response to nitrogen fertilization (DMY-res) and grassland nitrogen response to nitrogen fertilization (NY-res) were not different between the young and old grasslands (Table 1).

The average age difference between young and old grasslands was 16 years. Assuming an increase of 0.3% per year in productivity by genetic improvement, the average increase in productivity in these 16 years should theoretically be 4.8%. However, we found a (non-significant) decrease in DMY_{N0} of 9% of the young compared to the old grasslands. This is the productivity without N fertilization. According to Dutch legislation, farmers on a clay soil were allowed to fertilize their grasslands with 345 kg available N ha^{-1} $year^{-1}$ in 2014. At the N application rate of 345 kg N ha^{-1} $year^{-1}$, we also did not find a difference in the DMY (based on the DMY_{N0} and DMY-res per field) of young (on average 16.2 Mg DM ha^{-1}) and old grasslands (on average 16.3 Mg DM ha^{-1}). This finding is in line with the study of Hopkins (1990) who found higher productivity of *Lolium perenne* reseeds only at fertilizer-N rates of 450 and 900 kg N ha^{-1} $year^{-1}$. Apparently, the genetic potential of the most recently developed varieties sown in the young grasslands in our study was offset by the loss of soil quality.

The topsoil (0-10 cm) of the young grassland contained significantly lower soil organic matter, C-total and N-total than the topsoil of old grassland (Table 1), indicating C and N losses in topsoil due to ploughing. SOM and related soil parameters were strongly correlated with grass productivity parameters (Table 2).

Conclusions

Our study confirms that when grassland contains at least 70% desirable grasses, grass productivity does not increase after renewal on the long-term, most likely because of loss of SOM as a result of ploughing. In the past, dairy farmers could compensate for this loss of soil quality through extra fertilization. However, due to current legislative prescriptions, fertilization is limited and this makes such compensation less feasible. Therefore, a strict recommendation to renew all grasslands after 10 years to improve productivity

Table 1. Grass productivity and soil parameters in the 0-10 cm depth of the young (n=10) and old (n=10) grasslands on marine clay soil.[1]

Parameter	Unit	Young grassland		Old grassland		
		mean	SD	mean	SD	*P*-value
Grass age	years without cultivation	9	4	25	4	<0.001
DMY_{N0}	Mg DM ha^{-1} $year^{-1}$	9.2	2	10.2	1	0.154
DMY-res	kg DM kg N^{-1}	20	6	18	4	0.250
NY_{N0}	kg N ha^{-1} $year^{-1}$	172	50	198	21	0.034
NY-res	kg N kg N^{-1}	0.68	0.06	0.64	0.09	0.198
Soil organic matter	g. 100 g dry $soil^{-1}$	10.7	3.3	13.3	2.2	0.031
C-total	g C kg dry $soil^{-1}$	45.2	18	61.0	12	0.002
N-total	g N kg dry $soil^{-1}$	4.82	1.7	6.28	1.2	<0.001

[1] Means, standard deviations and *P*-values are based on a paired T-test. DMY_{N0}: grassland dry matter yield without nitrogen fertilization, DMY-res = grassland dry matter yield response to nitrogen fertilization; NY_{N0} = nitrogen yield without nitrogen fertilization; NY-res = grassland nitrogen response to nitrogen fertilisation; SD = standard deviation.

Table 2. Pearson correlations between grass productivity parameters and topsoil parameters (measured in 0-10 cm soil layer).

Grass productivity parameter	SOM	N-total	NSC±	C-total
DMY_{N0}	0.65**	0.70**	0.71**	0.67**
DMY-res	-0.78 ***	-0.79***	-0.81***	-0.77***
NY_{N0}	0.75 ***	0.80***	0.81***	0.77***
NY-res	-0.29	-0.28	-0.27	-0.30

[1] NSC± = nitrogen supply capacity calculated from N-total, corrected for grassland age according to the formulas of the Dutch grassland fertilization guideline. ** 0.001<*P*<0.01; ****P*<0.001.

can be considered obsolete. When the introduction of high yielding grassland varieties is necessary, the focus should be on oversowing (i.e. non-destructively adding grass seeds to the existing sward) rather than renewing the grassland. Moreover, farm management should focus on minimizing the need for renewal by good grassland management, e.g. maintaining desirable grasses by grazing, adequate fertilization, irrigation and preventing soil compaction (De Boer *et al.*, 2018).

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References

Chaves B., De Vliegher A., Van Waes J., Carlier L. and Marynissen B. (2009) Change in agronomic performance of *Lolium perenne* and *Lolium multiflorum* varieties in the past 40 years based on data from Belgian VCU trials. *Plant Breeding* 128, 680-690.

De Boer H.C., Deru J.G.C. and Van Eekeren N. (2018) Sward lifting in compacted grassland: effects on soil structure, grass rooting and productivity. *Soil and Tillage Research* 184, 317-325.

Hopkins A., Gilbey J., Dibb C., Bowling P.J. and Murray P.J. (1990) Response of permanent and reseeded grassland to fertilizer nitrogen. 1. Herbage production and herbage quality. *Grass and Forage Science* 45, 43-55.

Iepema G., Deru J.G.C., Bloem J., Hoekstra N., de Goede R., Brussaard L. and van Eekeren N. (2020) Productivity and topsoil quality of young and old permanent grassland: an on-farm comparison. *Sustainability* 12, 2600.

Russchen H.J. (2005) *Pasture Renovation: reseeding and crop rotation in practice (in Dutch)*. Plant Research International, Wageningen UR, Wageningen, the Netherlands.

Sampoux J.-P., Baudouin P., Bayle B., Béguier V., Bourdon P., Chosson J.-F., Deneufbourg F., Galbrun C., Ghesquière M., Noël D., Pietraszek W., Tharel B. and Viguié A. (2011) Breeding perennial grasses for forage usage: An experimental assessment of trait changes in diploid perennial ryegrass (*Lolium perenne* L.) cultivars released in the last four decades. *Field Crops Research* 123, 117-129.

Extending grassland age for climate change mitigation and adaptation on clay soils

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Abstract

Permanent grassland soils can act as a sink for carbon and may therefore positively contribute to climate change mitigation and adaptation. We compared young (5-15 years since latest grassland renewal) with old (>15 years since latest grassland renewal) permanent grassland soils in terms of carbon stock, carbon sequestration, drought tolerance and flood resistance. In old grassland soils we found a higher carbon stock (62 Mg C ha^{-1}) than in young grassland soils (51 Mg C ha^{-1}). The carbon sequestration rate tended to be higher (not statistically significant) in young (average 3.0 Mg C ha^{-1} $year^{-1}$) than old (1.6 Mg C ha^{-1} $year^{-1}$) grassland soils. Regarding potential drought tolerance, we found larger soil moisture and lower soil bulk density in old than in young grassland soils. Old grassland soils were also more resistant to heavy rainfall. We conclude that by extending grassland age on clay soil, farmers can contribute to climate change mitigation and adaptation ecosystem services.

Keywords: carbon sequestration, ecosystem services, permanent grassland, soil carbon stock, water infiltration

Introduction

On clay soils in the Netherlands, permanent grasslands are renewed (i.e. destroyed by herbicides, ploughed and reseeded) on average once every 10 years (Russchen, 2005). Ploughing of grassland for renewal significantly reduces soil carbon stocks (Linsler *et al.*, 2013; Necpálová *et al.*, 2014). In addition to climate change mitigation by preventing loss of carbon due to ploughing, permanent grassland can potentially play a role in climate change adaptation by increased drought tolerance and flood resistance. We tested the effect of permanent grassland age (young versus old grassland) on carbon stock and carbon sequestration rate in the topsoil (0-10 cm soil layer) as this can contribute to climate change mitigation. Additionally, we investigated the effect of permanent grassland age on soil parameters that can influence the resistance to periods of drought and the resistance to excess rainfall. Resistance to excess rainfall and periods of drought are both aspects of climate change adaptation.

Materials and methods

The study was conducted in 2014, at 10 dairy farms on marine clay soil in the north of the Netherlands. At each farm, a young (5-15 years since grassland renewal) and an old (> 15 years since grassland renewal) grassland were selected. On each grassland, a non-fertilized plot of 5×9 m was used to determine soil quality parameters in April 2014 and in December 2018 (only C-total). See Iepema *et al.* (2021) for details on the experimental lay-out, the measurements and the statistical analyses.

Results and discussion

By combining the observations from 2014 and 2018, we found a significant positive curvilinear relationship between age of the sward and carbon stock in the 0-10 cm soil layer. In the first years after grassland renewal, carbon stock increased relatively fast and after approximately ten years the line curved to a flatter response (Figure 1).

Potential drought resistance as indicated by soil moisture content, soil bulk density and soil structure, was significantly higher for old than for young grassland topsoils (Table 1). Rooting, the fourth parameter indicating potential drought resistance, was not significantly different between young and old grassland. However, we measured in old grassland a greater percentage of root tips from the total number of root tips at 10 cm (64%) in comparison with young grassland (57%). It might be that in old grassland, moisture and nutrients are more concentrated in the 0-10 cm soil layer, causing a larger proportion of root tips at 10 cm, which also contributes to a better soil structure in this soil layer. The old grassland soils showed the potential for greater resistance to heavy rainfall in comparison with the young grassland soils, as indicated by the larger water infiltration rate and a higher number of macropores (statistically significant only at 20 cm soil depth; Table 1). Macropores at this soil layer increase infiltration capacity under the subsoil (Jarvis *et al.*, 2017).

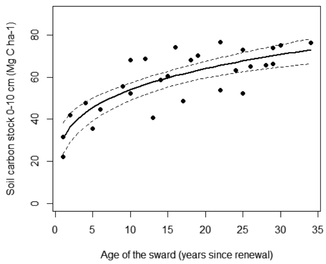

Figure 1. Carbon stock in the 0-10 cm soil layer (measured in 2014 and 2018) as a function of grassland age (years since renewal) with 95% confidence interval (dotted lines). The black line shows the model $y = 30.7 * x^{0.27}$; $R^2 = 0.67$.

Table 1. Characteristics of the topsoil (0-10 cm depth) related to climate change mitigation and adaptation of the young (n=10) and old (n=10) grasslands on marine clay soil.[1]

Parameter	**Unit**	**Young grassland**		**Old grassland**		***P*-value**
		mean	**SD**	**mean**	**SD**	
Grass age	Yrs without cult.	9	4	25	4	<0.001
Soil carbon stock	Mg C ha^{-1}	51	16	62	9	0.013
Carbon sequestration rate[2]	Mg C ha^{-1} $year^{-1}$	3.0	2	1.6	1	0.145
Parameters indicating potential drought resistance						
Soil moisture content	volume %	28.6	3.2	31.7	2.8	0.007
Soil bulk density	g cm^{-3}	1.16	0.1	1.03	0.1	<0.001
Crumbs	% of total weight	67.3	20	80.3	15	0.011
Angular blocky elements	% of total weight	12.2	14	4.6	8	0.052
Root tip density at 10 cm	Number dm^{-2}	109	16	118	40	0.524
Root tip density at 20 cm	Number dm^{-2}	81	17	65	23	0.185
Proportion of root tips at 10 cm (from the total number of root tips)	%	57	7	64	4	0.002
Parameters indicating potential excess rainfall resistance						
Water infiltration rate	mm min^{-1}	3.7	6	11.1	13	0.033
Macropores at 10 cm	number dm^{-2}	3.5	2	5.3	3	0.175
Macropores at 20 cm	number dm^{-2}	1.5	1	3.4	2	0.013

[1] Means, standard deviations, *P*-values based on a paired T-test. SD = standard deviation; Yrs without cult. = years without cultivation.

[2] Carbon sequestration rate was only measured on 6 young and 6 old grasslands.

Conclusions

The carbon sequestration rate of young grassland topsoil was larger than that of old grasslands, yet carbon is also still sequestered in the topsoil of grassland older than 30 years. Extending grassland age on clay soils can positively contribute to climate change mitigation and adaptation, but by how much warrants further study.

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References

Iepema G., Deru J.G.C., Bloem J., Hoekstra N., de Goede R., Brussaard L. and van Eekeren N. (2020) Productivity and topsoil quality of young and old permanent grassland: an on-farm comparison. *Sustainability* 12, 2600.

Iepema G., Hoekstra N.J., de Goede R., Bloem J., Brussaard L. and van Eekeren N. (2021) Extending grassland age for climate change mitigation and adaptation on clay soils. *European Journal of Soil Science* 73, e13134.

Jarvis N., Forkman J., Koestel J., Kätterer T., Larsbo M. and Taylor A. (2017) Long-term effects of grass-clover leys on the structure of a silt loam soil in a cold climate. *Agriculture, Ecosystems & Environment* 247, 319-328.

Linsler D., Geisseler D., Loges R., Taube F. and Ludwig B. (2013) Temporal dynamics of soil organic matter composition and aggregate distribution in permanent grassland after a single tillage event in a temperate climate. *Soil and Tillage Research* 126, 90-99.

Necpálová M., Li D., Lanigan G., Casey I.A., Burchill W. and Humphreys J. (2014) Changes in soil organic carbon in a clay loam soil following ploughing and reseeding of permanent grassland under temperate moist climatic conditions. *Grass and Forage Science* 69, 611-624.

Russchen H.J. (2005) *Pasture Renovation: reseeding and crop rotation in practice (in Dutch)*. Plant Research International, Wageningen UR, Wageningen, the Netherlands.

Possibilities for estimating the yield and quality of forage harvested from meadows of high natural value with non-destructive methods

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Abstract

Non-destructive methods and tools, such as the Rising Plate Meter (RPM), are increasingly being used to measure the quantity and quality of grassland swards on European farms. The aim of our study was to assess herbage yield and its quality in periodically excessively wet meadows, on a low, multi-species sward with patches of protected plants, using non-destructive methods. The study was carried out in 2019-2021, in eleven extensive meadows located in the valley of Rządza river (central Poland). Two methods of yield evaluation were compared: (1) sward height and sward density measurement, and yield calculation according to Kostuch, and (2) the RPM. Sward botanical composition was identified by the Braun-Blanquet method and its quality using the fodder value score according to Filipek. We discuss the problems with precise measurements of sward height due to sward heterogeneity. Although non-destructive methods and tools can be used to assess the quantity and quality of yields in extensive meadows of high nature value, use of the RPM needs further calibration.

Keywords: multi-species sward, permanent meadow, sward height, yield, feeding value score

Introduction

Non-destructive methods and tools for measuring the quantity and quality of pastures have recently become more and more used on European farms, particularly those with intensive grazing systems (McMurphy *et al.*, 2021). In Poland, farmers estimate the yields of permanent grassland most often based on the number of bales of hay or haylage harvested. One simple method for measurement of herbage yield uses the Rising Plate Meter (RPM), sometimes known as the Grasshopper system (Hart *et al.*, 2019). It is usually used in pastures to assess feed available for grazing. In Poland, the area of permanent pastures is small, accounting for only 2.6% of total agricultural land, while permanent meadows comprise 19.1%. Wet meadows with patches of protected species are especially valuable. Many of them are now highly endangered, and the most important threat is abandonment of management. The aim of this study was to evaluate the natural and productive value of these meadows and to assess the possibility of using some non-destructive methods (including RPM) to estimate the quantity and quality of the yield from such valuable meadows.

Materials and methods

The study was carried out in 2019-2021, in eleven extensive meadows located in the valley of the upper section (over a length of 12 km) of the Rządza river (Jakubów commune, central Poland). The meadows are periodically wet, as a result of groundwater rising to the surface, especially in spring and after periods of heavy rainfall. The loamy sand soils are mainly acidic (pH_{KCl} = 4.01-5.59), with a very low P (8.04-21.79 mg kg^{-1}) and K (24.04-62.72 mg kg^{-1}) content. The meadows have not been fertilized for several years. They differed in the frequency of mowing: five meadows were cut once a year, three were cut twice a year, one was cut every two years and two were fallow for 4 years. Vegetation was identified, based on an analysis of phytosociological relevés (Braun-Blanquet method) made in the first half of June each year. Each reading represented an area of 100 m^2. Natural values of the meadows were determined with the total number of species and their abundance. Shannon's diversity index values (H') were calculated. The production value of the meadows was assessed on the basis of the first-cut yield and its fodder value. Two methods of yield evaluation were compared: (1) the height of main sward mass and the degree of soil coverage with plants (sward density) were determined with a ruler (1 cm = 1%) in more than 12 places. Then the DM yields (Y) were calculated using the Kostuch (1982) formula: $Y = (r - s) \times 0.6 \times (z/100) \times 0.2$, where Y – DM yield [t ha^{-1}], r – height of the main sward mass, s – cutting height, 0.6 – yield of 1 cm sward (green hay) per 1 ha [t] at 100% sward density, z – sward density [%], 0.2 – conversion of green hay to DM (5 kg of green hay = 1 kg of DM), (2) using the rising plate meter (RPM) (in 2020-2021), which measures the height of compressed sward and calculates the available DM ha^{-1} using the conversion equation. This method takes into account the grass density as well as height. The quality of yield was determined using the fodder value score (FVS) according to Filipek (1973). The correlation coefficients were calculated to determine the strength of relationship between the results of sward height and yielding obtained with the two methods.

Results and discussion

In the studied meadows, 111 vascular plant species were found, including three protected species: *Iris sibirica* L. (VU) (in one patch), *Dactylorhiza majalis* (Rchb.) P.F. Hunt et Summerh. (NT), and *D. incarnata* (L.) Soó (NT). Among the total number of species there were 21 grass species, 11 species from *Fabaceae*, 60 non-legume forbs and 19 species from *Cyperaceae*, *Juncaceae* and *Equisetaceae*. A greater number of species was found in 1-cut meadows as compared to 2-cut meadows or to meadows which were fallow for 4 years (Table 1). Grasses and non-leguminous forbs made up most of the vegetation cover. Species characterized by the highest (V) class of phytosociological constancy and cover coefficient (up to 25-50%) were: *Holcus lanatus* L., *Ranunculus acris* L., *Lychnis flos-cuculi* L., *Anthoxanthum odoratum* L. *Poa pratensis* L., *Festuca rubra* L., *Rumex acetosa* L., *Ranunculus repens* L., *Plantago lanceolata* L., *Festuca pratensis* Huds. and *Deschampsia caespitosa* (L.) P. Beauv. The presence of *A. odoratum* L. and

Table 1. Differentiation of the share of plant groups, natural and production values depending on the frequency of mowing.

Group of plants [%]	Frequency of mowing			
	Twice a year	Once a year	Every 2 years	Fallow for 4 years
Grasses	44.5	40.1	49.6	45.4
Legumes	5.3	6.8	9.8	2.4
Sedges, rushes and horsetails	12.7	12.4	8.1	2.5
Non-legume forbs	37.5	40.7	32.5	49.7
Natural value				
Number of species				
Mean in patch	33	36	37	34
Range in patch	28-40	25-47	33-40	26-36
H' index	1.84-3.15	2.50-3.33	2.24-2.93	2.19-3.08
Production value				
Sward height [cm] (ruler)	18.6-49.4	27.0-48.6	38.6-71.6	36.0-62.2
Sward density (%)	65-90	45-80	45-55	45
Yield [kg DM ha^{-1}] (Kostuch)	1,224.0-3,196.8	1,890.0-4,081.0	2,217.6-3,596.4	1,674.0-3,091.5
Sward height [cm] (RPM)	7.3-11.2	10.1-15.0	14.2-19.8	10.0-11.8
Yield [kg DM ha^{-1}] (RPM)	2,456.8-3,440.0	3,164.5-4,380.4	4,196.5-5,582.4	3,126.6-3,586.3
FVS	3.22-5.14	2.02-5.18	5.12-6.23	4.69-5.50

Luzula campestris (L.) DC. indicates a significant deterioration of the habitat, which was confirmed by soil tests. The study showed that the vast majority of plants belonged to the *Molinio-Arrhenatheretea* class, characteristic for semi-natural meadow communities, and to the *Scheuchzerio-Caricetea nigrae* class, whose species were located in the lower terrain. The high values of the Shannon-Wiener diversity index (2.5 - 3.33) indicate high biodiversity. Most of the meadows are characterized by low yields of moderately quality (FVS 3-6), and a good value was obtained in one case only. The highest yields with the best value were distinguished by the meadows with the highest share of grasses, located in the most favourable water and soil conditions. It was found that both the sward heights and yields obtained using both methods (RPM and the Kostuch) were positively correlated ($r = 0.669$ and $r = 0.619$ respectively). When analysing the sward height, it was noticed that the RPM showed lower values than those measured with a ruler, while the inverse relationship was found in relation to the yields. The RPM overestimated the results, especially when the sward was high and the share of weeds with thick stems was greater (e.g. *Cirsium palustre* (L.) Scop., *Centaurea jacea* L.). There was also a problem with uneven swards, empty spaces and dense tufts of grasses, sedges or rushes. This is in line with the conclusions of Werner *et al.* (2021) who concluded that the application potential of the RPM might be limited and needs further adaptation for use with heterogenous short-grass swards.

Conclusions

Wet meadows with patches of protected species located in the Rządza River valley are characterized by high floristic diversity and low yields of moderate quality. Both the sward height and yield values obtained using both methods (RPM and Kostuch) were positively correlated. The RPM overestimated yields when the sward was high and included species with thick stems. There was also a problem with uneven swards, empty spaces and dense tufts of grasses, sedges or rushes. The use of RPM in such meadows needs further calibration.

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References

Filipek J. (1973) Project of classification of meadow and pasture plants on the basis of numbers of utility value [in Polish]. *Postępy Nauk Rolniczych* 4, 59-68.

Hart L., Oudshoorn F., Latsch R. and Umstatter C. (2019) How accurate is the Grasshopper® system in measuring dry matter quantity of Swiss and Danish grassland? In: 9th European Conference on Precision Livestock Farming, Cork, Ireland, 26-29 August 2019, pp. 188-193.

Kostuch R. (1982) Estimating and measuring valuation of yields from meadows and pastures [in Polish]. *Wiadomości Melioracyjne i Łąkarskie* 6, 126-128.

Murphy D.J., Murphy M.D., O'Brien B. and O'Donovan M. (2021) A review of precision technologies for optimising pasture measurement on Irish grassland. *Agriculture* 11, 600.

Werner J., Stumpe C., Höhn D., Bateki C. and Dickhöfer U. (2021) Case study on monitoring sward height and available biomass with a rising plate meter on pastures of dairy farms in Southwest Germany. *Grassland Science in Europe* 26, 98-100.

Stress caused by extreme weather conditions reflects on the nutritive value of grass

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Abstract

With climate change, extreme weather conditions are becoming more common causing challenges to grass quality. The aim of our study was to measure effect of air temperature and soil moisture on nutritive value of timothy and meadow fescue. In a greenhouse experiment, plants were exposed to three temperatures (cold, optimum, hot) and for three soil moisture levels (dry, optimum, wet) for aftermath growth for 2 weeks before cutting. Dry matter (DM) yield, nutritive value and mineral content of the grass were analysed. Increases in temperature and soil moisture increased the amount of DM yield and decreased digestibility in both species. In general, soil moisture had a larger effect on measured parameters than air temperature. The largest effect of soil moisture was seen in reducing sugars and phosphorus (P). When soil moisture was raised from optimum to wet, increase in P concentration was relatively larger than the increase in yield. The concentration of sugars decreased with increasing air temperature and soil moisture, indicating reduced stress level of the plants. The level of crude protein was adequate to support normal grass growth. Based on the results, the biggest stressor was drought, although cold also limited yield formation.

Keywords: *Phleum pratense*, *Festuca pratensis*, drought, temperature, feed value

Introduction

With climate change, extreme weather conditions are becoming more common and presenting challenges to grass growth and nutritional quality. Combinations of abiotic stresses, such as cold and drought, are commonly believed to have an additive negative effect on crop physiology and dry matter (DM) yield compared to situations when they occur separately, but they may also have an ameliorative effect on the plant (Loka *et al.,* 2018). The physiological stress response of grass depends on grass survival strategy in stressful environments, and this may vary between species (Okamoto *et al.,* 2011). Thus, Loka *et al.* (2018) suggested that it would be important to study the effects of combined stresses closely aligned to real-field conditions. The aim of this study was to measure effects of air temperature and soil moisture and their combinations on nutritive value of timothy (*Phleum pratense*) and meadow fescue (*Festuca pratensis* Huds) in a greenhouse experiment.

Material and methods

The effect of soil moisture and air temperature on DM yield, nutritive value and mineral content in the second cut of timothy and meadow fescue was studied in a greenhouse experiment. The soil (44% sand, 45% loam+clay) for the experiment was collected from the field, screened, limed and fertilized with 50 mg/kg P, 250 mg/kg N and 155 mg/kg K. Timothy and meadow fescue were sown as pure stands in jars (4.4 kg, 18 cm Ø, height 15 cm, 20 seeds per jar, 36 jars of both grass). Grasses were grown in the greenhouse for 2 months, cut, and vernalized at 10 °C degrees for 3 months. After vernalization the grasses were cut to 7 cm and fertilized with 250 mg/kg N and 155 mg/kg K at each cut. Growing period before the first cut lasted 30 days at 17/12 °C with 18/6 h diurnal light/dark cycle. After the first cut, the jars were grown for 3 weeks at 17/12 °C, followed by stress treatments for two weeks. After stress period the grasses were cut to 7 cm and DM yield was measured, and samples sent for analysis. Stress treatments included three temperature and soil moisture levels. Temperature levels were: +10/+2 °C (cold), +18/+12 °C (optimum) and +25/+17 °C (warm) in 16/8 h diurnal cycle. The temperature sum during the growing period was 269, 376 and 478 degrees under cold, optimum and warm respectively. Soil moisture levels during the two-week stress period were: 40% (dry), 50-70% (optimum) and 100% (wet) of the field capacity, respectively. Grass nutritive value was measured by near-infrared spectroscopy (NIR) method (Valio Ltd.) and mineral content by inductively coupled plasma spectroscopy (ICP). Results of the experiment were analyses using ANOVA (SAS 9.4., *Glimmix*-procedure).

Results and discussion

The effect of temperature on grass yield and quality was clear. The DM yield of grasses grown in cold and wet conditions was on average 25% higher than in cold and dry conditions, but less than half that produced in warm and wet growing conditions (Table 1). As is usual in greenhouse experiments, the proportion of digestible organic matter (D-value) of the forage was high. Increasing temperature and temperature sum lowered the D-value in both species, and the effect was greatest in wet soil conditions, especially for timothy. Under cold conditions, soil moisture had no effect on the D-value.

Table 1. Dry matter yield (g DM m^{-2}), crude protein (CP), reducing sugars (RS), neutral detergent fibre (NDF), D-value and P and K concentration, g kg DM^{-1} in forage harvested at the second cut of timothy and meadow fescue growth in cold, optimum and warm temperature (Temp) and dry, optimum and wet soil moisture (Moist) level.[1]

Temp	Moist	DM g/m^2	CP g kg	RS/DM	NDF	D-value	P	K
(Sp) Timothy								
Cold	dry	386	187	197	409	744	1.99	34.3
	optimum	495	192	223	398	734	2.15	27.6
	wet	477	206	166	422	743	3.09	34.8
Optimun	dry	456	174	257	389	742	1.58	24.7
	optimum	628	189	199	427	738	1.92	26.8
	wet	832	193	139	477	715	3.04	32.2
Warm	dry	582	178	163	453	709	1.78	28.0
	optimum	851	181	139	471	695	2.13	32.3
	wet	1143	192	66	537	662	3.21	36.3
(Sp) Meadow fescue								
Cold	dry	402	191	229	395	743	1.97	33.2
	optimum	551	201	166	419	759	2.20	37.9
	wet	512	201	176	432	742	3.03	33.8
Optimun	dry	450	182	211	402	761	1.66	31.8
	optimum	822	199	151	452	753	2.03	34.4
	wet	779	195	120	484	738	2.56	35.0
Warm	dry	480	189	123	458	721	2.01	35.8
	optimum	975	194	88	505	705	2.51	39.2
	wet	1182	194	59	528	696	2.82	39.2
Mean		667	191	159	448	728	2.31	33.2
SEM		70	7.1	12.6	7.8	6.5	0.124	1.72
P values	Sp	ns	*	***	**	***	ns	***
	Temp	***	ns	*	**	***	ns	ns
	Sp×Temp	ns	ns	*	ns	ns	ns	ns
	Moist	***	**	***	***	***	***	**
	Sp×Moist	ns	ns	***	**	ns	**	**
	Temp×Moist	***		***	***	*	ns	ns
	Sp×Temp×Moist	ns	ns	*	ns	ns	ns	ns

[1] Significance: *** $P<0.001$, ** $P<0.01$, * $P<0.05$, ns = non-significant; SEM = standard error of the mean.

The CP concentration of the grass corresponded to the CP concentrations of field experiments (Termonen *et al.,* 2020), i.e. the grass did not suffer from N deficiency. However, the effect of the limited N supply can be seen in decreasing CP concentration when growth conditions are favourable for grass growth. Based on the RS concentration describing the stress level of the grass, the biggest stressor was drought, although cold temperature also limited grass yield and increased sugar content. This is also reflected in a negative correlation between RS and NDF (r=-0.91, P<0.001). The amount of RS was at its highest at optimum temperature when drought limited growth. In silage production low sugar content in herbage presents a risk for successful silage conservation (Davies *et al.*, 1998).

Increasing soil moisture favoured the uptake of P by both species (Table 1). The P content, especially in timothy, increased proportionally more than the DM yield from dry to wet soil moisture conditions. Water is essential for the grass to have access to soil P. Furthermore, P cannot move in the soil over long distances and therefore the root system must be adequate to provide a sufficient contact with the P on soil particles. Temperature had no effect on P uptake, unlike, for example, barley, where in previous Finnish experiments cold has made P uptake more difficult (Ylivainio and Peltovuori, 2018). There was also a significant difference in K uptake between the species and soil moisture levels. Meadow fescue had high concentrations of K in warm conditions, increasing with soil moisture level, which may be nutritionally harmful (Rérat *et al.,* 2009).

Conclusions

In general, soil moisture had a larger effect than air temperature on the forage parameters measured here. The largest effect of soil moisture was seen in RS and P. Changes in RS concentrations caused by weather conditions may present challenges for successful biological conservation of silage, and the risk of changes in forage mineral concentrations must also be considered in relation to impacts on animal health.

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References

Davies D.R., Merry R.J., Williams A.P., Bakewell E.L., Leemans D.K and Tweed J.K.S. (1998) Proteolysis during ensilage of forages varying in soluble sugar content. *Journal of Dairy Science* 81, 444-453.

Loka D., Harper J., Humphreys M., Gasior D., Wootton-Beard P., Gwynn-Jones D., Scullion J., Doonan J., Kingston-Smith A., Dodd R., Wang J., Chadwick D., Hill P., Jones D., Mills,G., Hayes F., and Robinson D. (2019) Impacts of abiotic stresses on the physiology and metabolism of cool-season grasses: A review. *Food Energy Security* 8, e00152. https://doi.org/10.1002/fes3.152

Okamoto H., Ishii K. and An P. (2011) Effects of soil moisture deficit and subsequent watering on the growth of four temperate grasses. *Grassland Science* 57, 192-197. https://doi.org/10.1111/j.1744-697X.2011.00232.x

Rérat M., Philipp A., Hess H.D. and Liesegang A. (2009) Effect of different potassium levels in hay on acid-base status and mineral balance in periparturient dairy cows. *Journal of Dairy Science* 92, 6123-6133.

Termonen M., Korhonen P., Kykkänen S., Kärkönen A., Toivakka M., Kauppila R. and Virkajärvi P. (2020) Effects of nitrogen application rate on productivity, nutritive value and winter tolerance of timothy and meadow fescue cultivars. *Grass and Forage Science* 75, 111-126.

Ylivainio K and Peltovuori T. (2018) Phosphorus acquisition by barley (*Hordeum vulgare* L.) at suboptimal soil temperature. *Agricultural and Food Science* 12, 453-461.

Soil carbon sequestration potential of grass-clover leys: effect of grassland proportion and organic fertilizer

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Abstract

Grass-clover leys may contribute to mitigation of climate change by soil organic carbon (SOC) sequestration, but quantitative documentation of this is extremely rare. This study aims to quantify the effect of grassland proportion and fertilization on SOC storage in the topsoil layer (0-20 cm). For this objective, we used a long-term field experiment including plots with contrasting grassland proportions (2 or 4 years of grass-clover in a six-course rotation) and cattle slurry input during 2005 to 2018. A multiple regression model including grassland proportion (grass-clover 31%- 69%), slurry input (0 to 1.45 t C ha^{-1} yr^{-1}) and the initial SOC stock explained 47% of the variation in the SOC stock change from 2005 to 2018. The SOC stock based on equivalent soil mass (SOC $stock_{FM}$) increased 3.18 Mg C ha^{-1} when increasing grassland proportion by 25% during the 13 years. Applied slurry-C contributed with 11% to soil C sequestration, while an annual slurry-N input corresponding to 150 kg total-N ha^{-1} or 1.5 t C ha^{-1} increased SOC $stock_{FM}$ by 2.16 Mg C ha^{-1}. Our results may be used for climate change accounting at an industry, regional or farm scale, in analysis of the climate impact of dairy products and for modelling SOC sequestration.

Keywords: cattle slurry, grass-clover ley, soil carbon storage

Introduction

Soil C sequestration is an important climate change mitigation strategy and can play a role in offsetting global fossil CO_2 emissions (Smith *et al.*, 2020). Increasing the grassland proportion in crop rotations is among the most substantial climate mitigation measures in modern agriculture. However, an accurate quantification of SOC sequestration potential of grass-clover leys is challenging as long-term experiments with frequent soil sampling and crop rotations with varying grassland proportion are extremely rare. Here we examine the effect of grassland proportion and cattle slurry input on SOC storage in the topsoil layer.

Materials and methods

The organic dairy crop rotation experiment was established in 1987 at Foulumgaard Experimental Station, Aarhus University, Denmark (56°29'N, 09°34'E). The soil is a loamy sand with 7.7% clay. A six-year rotation with two years of grass-clover was introduced in 1987 replacing a cereal-dominated rotation. From 2006 the six-year rotation was split in two crop rotations differing in grassland proportions, being 1/3 under rotation 1 (2G; barley, two years of grass-clover, barley for wholecrop silage with Italian ryegrass undersown, oat with ryegrass undersown and finally barley/pea), and 2/3 under rotation 2 (4G; barley, four years of grass-clover and barley for wholecrop silage with Italian ryegrass undersown). Each of the six fields in the two rotations was divided into two blocks, in which five treatments with varying cattle slurry input were randomly allocated. Cattle slurry was applied in spring. Soil was sampled in the 0-20 cm soil layer in 1986, 1993, 2005, 2009 and 2018, air-dried and then archived. Soils from 1986 and 1993 were no longer available, but had previously been analysed for SOC by high-temperature dry combustion. Archived soil samples were crushed, sieved (<2 mm) and analysed for SOC by dry combustion at 950 °C using a Vario Max Cube. The SOC content is expressed as g 100 g^{-1} oven-dry soil (105 °C for 24 h). In autumn 2020, three undisturbed soil cores (100 cm^3) were extracted from the 6-10 cm soil layer in each plot. This depth was chosen to best reflect the 0-20 cm ploughing depth. The soil cores were oven-dried at 105 °C for 24 h and the calculated bulk density corrected for mass and volume of >2-mm particles. The

>2-mm particles was determined after wet sieving and drying. The SOC stock was based on equivalent soil mass (SOC $stock_{FM}$) as described in Smith *et al.* (2020). Multiple regression was applied using the R-project software package Version 3.4.0 (R Foundation for Statistical Computing).

Results and discussion

Figure 1 shows the SOC $stock_{FM}$ in topsoil sampled during 1986 to 2018 from plots with either 2yr-grass-clover in the 6 year crop rotation throughout the period or 2yr-grass-clover followed by 4yr-grass-clover (2006-2018). As expected, no significant differences were observed between plots in 1986, 1993 and 2005 (i.e. 2yr-grass-clover in both rotations). Increasing the grassland age to 4yr-grass-clover increased the SOC $stock_{FM}$ significantly as compared to continuing with 2yr-grass-clover (2009 and 2018, Figure 1). Under continuous 2yr-grass-clover, the SOC $stock_{FM}$ increased with 2.56 Mg C ha^{-1} (0.37 Mg C ha^{-1} yr^{-1}) and 2.78 Mg C ha^{-1} (0.23 Mg C ha^{-1} yr^{-1}) from 1986 to 1993 and 1993 to 2005, respectively. When changing the grassland age from 2 to 4 years, the SOC $stock_{FM}$ increased with 2.65 Mg C ha^{-1} (0.20 Mg C ha^{-1} yr^{-1}) (Figure 1). When the experiment was established in 1987 the initial SOC content was 1.61% C (42.5 Mg C ha^{-1}) and the field had mainly been used for cereal cropping. At this initial SOC content a change from cereal cropping to a crop rotation with 2yr-grass-clover hence increased the SOC $stock_{FM}$. Notably, the SOC accrual rate was highest in the initial years following the management change similar to the findings of Smith (2014) for conversion of cropland to grassland. The crop rotation with 2yr-grass-clover throughout the period seemed to have reached steady-state conditions in 2005, whereas SOC $stock_{FM}$ with increased grassland proportion continued to increase linearly (Figure 1).

Grassland proportion, cattle slurry input and initial SOC content at plot level during 2006-2018 was used to explain the change in SOC $stock_{FM}$ from 2005 to 2018:

Change in SOC $stock_{FM}$ = 12.74 (P<0.001) x Grassland proportion + 0.11 (P<0.05) × Slurry-C input – 0.09 (P=0.33) × SOC $stock_{FM\text{-}initial}$ – 1.82 (P=0.66), R^2=0.47 (1)

where grassland proportion is the amount of grass-clover in the crop rotation (0.00-1.00), slurry input is the amount of soil C added in cattle slurry and SOC $stock_{FM\text{-}initial}$ is the SOC stock in 2005. The model explained 47% of the variation in the SOC $stock_{FM}$ change from 2005 to 2018. Figure 2 shows the effect of grassland proportion in the crop rotation and organic fertilization on the annual change in SOC $stock_{FM}$ from 2005 to 2018 based on the model parameters.

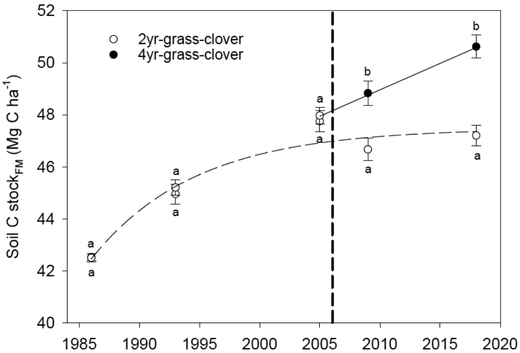

Figure 1. Soil organic carbon (C) stock (0-20 cm) for the two crop rotations during 1986 to 2018. During 1987 to 2006, the 6 year crop rotation comprised 2yr-grass-clover. From 2006 and onwards the experiment was split into two parts; one continued with 2yr-grass-clover, while the grass-clover proportion was increased to 4 years in the other. The standard error of the mean is indicated (n=60). Within years, letters denote statistical significance between crop rotations at P<0.05.

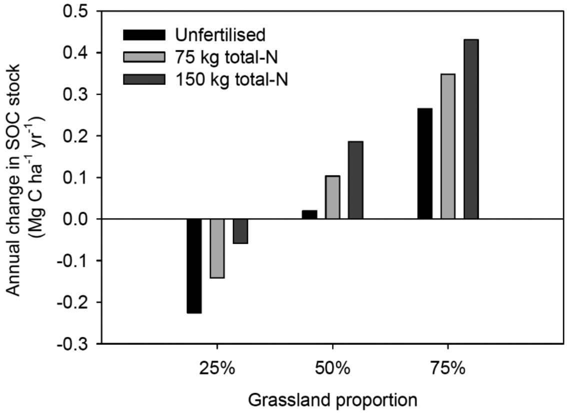

Figure 2. Simulated annual changes in soil organic carbon (C) stocks (0-20 cm) for contrasting grassland proportions in the crop rotation and different cattle slurry input at an average initial SOC stock. The simulation is based on Equation 1.

Conclusions

The SOC $stock_{FM}$ increased by 5.02 Mg C ha^{-1} (0.25 Mg C ha^{-1} yr^{-1}) when converting cereal cropping to a crop rotation with 2yr-grass-clover in a 6 year rotation. The steady-state condition was reached after 20 years. Based on a multiple regression model for the period 2005 to 2018 the SOC $stock_{FM}$ increased by 3.18 Mg C ha^{-1} (0.24 Mg C ha^{-1} yr^{-1}) when increasing grassland proportion by 25%. Slurry-C contributed with 11% to soil C sequestration. An annual increase in slurry input equivalent to 150 kg total-N ha^{-1} (1.5 t C ha^{-1}) increased SOC $stock_{FM}$ by 2.16 Mg C ha^{-1} (0.17 Mg C ha^{-1} yr^{-1}). These soil C sequestration potentials may be used for climate change accounting.

References

Smith P. (2014) Do grasslands act as a perpetual sink for carbon? *Global Change Biology* 20, 2708-2711.

Smith P., Soussana J-F., Angers D. Schipper L., Chenu C., Rasse D.P. *et al.* (2020) How to measure, report and verify soil carbon change to realize the potential of soil carbon sequestration for atmospheric greenhouse gas removal. *Global Change Biology* 26, 219-241.

Changes in sugar concentration over the day and the season in green forages

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Abstract

The aim was to investigate the variation in sugar concentration over the day in green forages in two seasons. Plots with perennial ryegrass, timothy, tall fescue, meadow fescue, orchard grass and lucerne were established in spring 2020, at Foulum, Denmark. First regrowth was sampled in August and the second regrowth was sampled in October, three times over the day (morning, noon and evening) on three days. All samples (n=126) were analysed for glucose, fructose, sucrose and fructans, and the sum were set as total water soluble carbohydrates (WSC). Across species, WSC concentration was lower in October than in August (74.3 vs 120 g kg^{-1} of dry matter (DM)). Generally, lucerne and orchard grass had lowest WSC concentrations, whereas perennial ryegrass had the highest. Increase in sucrose concentration was the main driver for increase in aboveground plant WSC concentration over the day.

Keywords: grass, lucerne, water soluble carbohydrates, diurnal variation, animal nutrition

Introduction

All plants produce simple sugars during photosynthesis, a process activated by solar radiation. The photosynthetic cells use a part of the sugar for respiration, but sugar in surplus is transported to parts of the plants with high energy demand or for storage for later use. The transport of sugar in the plant is primarily in the form of sucrose, which is not a reducing sugar. In many plant species, the excess energy is stored as starch, but in cool-season C_3 grasses, energy is stored as fructans. As part of growth and maturation of the plants, the sugars are also incorporated into structural carbohydrates, a process which is accelerated by increased temperature. Therefore, many factors such as temperature, radiation, maturation, day length and species affect the composition of carbohydrates in plants and will contribute to variation over day and season. Sufficient sugar is essential for a good preservation when making silage for cows, but within equine nutrition, sugars, especially fructans, in grass have been associated with an increased incidence of laminitis (Van Eps and Pollitt, 2006). The aim of this study was therefore to investigate the variation in water soluble carbohydrate (WSC) concentration in different green forages over the day in two seasons under Danish conditions.

Materials and methods

As part of a larger study, 60 plots (effective plot size: 27 m^2) with different grass and legume species were established on 22 April 2020 at AU Foulum (56°29'N, 9°35'E), Tjele, Denmark. Seven of these plots were selected for the current study, including perennial ryegrass (*Lolium perenne* L.) both a diploid (var. 'Bovini', DPR) and a tetraploid (var. 'Sherlock', TPR) variety, timothy (*Phleum pratense* L., var. 'Radde', TIM), tall fescue (*Festuca arundinacea* Schreb., var. 'Ferguson', TF), meadow fescue (*Festuca pratensis* Huds., var. 'Schwetra', MF), orchard grass (*Dactylis glomerata* L., var. 'Kobako', OG) and lucerne (*Medicago sativa* L., var. 'Cigale', LUC). The primary growth, except for LUC, was harvested and removed on 19 June. Due to a poor and uneven establishment, the plots with TIM, MF and OG were reseeded on 24 June. In August, at three days (6, 10 and 13 Aug.), samples (~500 g) from the plots were collected manually with hand-held scissors to a stubble height of 7 cm at 06:00, 12:00 and 18:00 hour each day. After the last sampling day in August, all plots were fully harvested, and the regrowth were sampled at three days in October (1, 5 and 8 Oct.) at 08:00, 12:00 and 16:00 hour each day. Immediately after aboveground plant sample collection, the samples were dried (60 °C, 48 h) after removal of any weed.

Dried samples (n=126) were milled to 0.5 mm and analysed for glucose, fructose, sucrose and fructans using a sequential enzymatic colorimetric method after extraction with a 0.1 M acetate buffer (Larsson and Bengtsson, 1983). The sum of the four analytes were considered as total WSC. Statistical analyses were performed in R (version 4.1.1) using a linear mixed model (*lmer*) including species (n=7), period (n=2), day (n=3) and time (n=3) as main effects as well as their two- and three-way interaction, except species × period × day. The three samples taken at the same plot within day were considered as repeated measurements with a compound symmetry covariance structure. *P*-values are presented in Table 1.

Results and discussion

Across species, WSC was lower in October than in August (Table 2; 74.3 vs 120 g kg^{-1} of DM across species). In both periods, TPR had highest and LUC had lowest WSC concentration. For LUC, WSC concentration did not differ between periods, whereas MF was the species that differed the most between periods (149 and 62.2 g WSC kg^{-1} of DM in August and October, respectively). LUC did not contain any fructans because legumes store excess energy as starch rather than fructans as is the case for the grasses. For the grasses, OG had a lower concentration of fructans, both absolute and relative to WSC (11% of WSC) compared to the others (>30% of WSC). In all species, sucrose concentration increased as expected (Pelletier *et al.*, 2010) from morning to evening (Table 3) with highest increase in absolute amount in TF (+24.1 g kg^{-1} of DM) and lowest increase in OG (+10.6 g kg^{-1} of DM). For TF, MF and LUC, there was only a numeric increase in glucose concentration over day, but also only a minor increase was observed for the other species. Even though the time between morning and evening was shorter in October than in August (-4 h due to reduced day length), the increase in glucose, fructose and sucrose over the day was higher in October than August (Table 4); no increase was observed for fructose in August. In both periods, sucrose concentration was the main driver for increase in aboveground plant WSC concentration over the day, as sucrose concentration increased by 58.7% in August (from 31.5 to 50.0 g kg^{-1} of DM) and by 90.6% in October (19.2 to 36.6 g kg^{-1} of DM). Additionally, concentration of fructans decreased over the day in August, but stayed stable over the day in October. In both periods, sampling day affected the increase in sucrose over the day ($P_{P\times T\times D}$ <0.01, Table 1) due to differences in solar radiation on the sampling days (Jørgensen, 2021).

Conclusions

The concentration of sugars in green forage depended on species and time of harvest, and was generally higher in August than in October. Sucrose was the main sugar component driving the increase in WSC concentration over the day, and sucrose increased more in October than in August from morning to evening.

Table 1. *P*-values for the effect of the variables species, period, day and time and their interactions from the model analysing concentrations of glucose, fructose, sucrose, fructans and total WSC[1] in green forages.

	P-values[2]										
	S	P	D	T	S×P	S×D	S×T	P×D	P×T	D×T	P×D×T
Glucose	< 0.01	< 0.01	0.04	< 0.01	< 0.01	0.59	0.10	0.89	0.07	< 0.01	0.23
Fructose	< 0.01	< 0.01	0.29	< 0.01	0.35	0.86	0.39	0.88	< 0.01	< 0.01	0.27
Sucrose	< 0.01	< 0.01	0.06	< 0.01	< 0.01	0.50	< 0.01	0.02	0.05	< 0.01	< 0.01
Fructans	< 0.01	< 0.01	0.03	0.38	< 0.01	0.13	0.37	0.03	0.03	0.03	0.07
Total WSC	< 0.01	< 0.01	0.16	< 0.01	< 0.01	0.21	0.57	0.27	0.02	< 0.01	0.01

[1] WSC = sum of glucose, fructose, sucrose and fructans.

[2] S = species, P = period, D = day, T = time. The remaining 3-way interactions were not significant.

Table 2. Concentrations of glucose, sucrose, fructans and total WSC (g kg^{-1} of dry matter) in regrowth above 7 cm of different species in August and October across sampling day and sampling time.[1,2]

		Species							
Analyte	**Period**	**DPR**	**TPR**	**TIM**	**TF**	**MF**	**OG**	**LUC**	**SEM**
Glucose	August	$17.4^{c, x}$	$18.5^{c, x}$	$27.4^{d, x}$	$14.6^{bc, x}$	$15.1^{bc, x}$	$12.6^{b, x}$	$5.66^{a, x}$	0.95
	October	$12.3^{b, y}$	$11.5^{b, y}$	$9.22^{b, y}$	$9.24^{b, y}$	$8.00^{ab, y}$	$8.00^{ab, y}$	$4.19^{a, x}$	
Sucrose	August	$43.3^{bc, x}$	$54.7^{d, x}$	$35.6^{ab, x}$	$49.9^{cd, x}$	$44.6^{bc, x}$	$25.8^{a, x}$	$28.1^{a, x}$	1.99
	October	$24.9^{ab, y}$	$28.5^{bc, y}$	$25.9^{abc, y}$	$35.7^{c, y}$	$25.4^{ab, y}$	$17.4^{a, y}$	$27.7^{bc, x}$	
Fructans	August	$71.5^{c, x}$	$73.5^{c, x}$	$65.2^{c, x}$	$44.8^{b, x}$	$73.2^{c, x}$	$7.14^{a, x}$	$-2.38^{a, x}$	3.08
	October	$52.6^{cd, y}$	$56.7^{d, y}$	$19.1^{b, y}$	$40.2^{c, x}$	$18.1^{b, y}$	$4.73^{ab, x}$	$-2.31^{a, x}$	
Total WSC	August	$153^{cd, x}$	$170^{d, x}$	$141^{bc, x}$	$126^{b, x}$	$149^{bcd, x}$	$61.3^{a, x}$	$39.7^{a, x}$	4.82
	October	$106^{c, y}$	$114^{c, y}$	$62.4^{b, y}$	$98.2^{c, y}$	$62.2^{b, y}$	$41.1^{ab, y}$	$36.0^{a, x}$	

[1] WSC = sum of glucose, fructose, sucrose and fructans; DPR = diploid perennial ryegrass; TPR = tetraploid perennial ryegrass, TIM = timothy, TF =tall fescue, MF = meadow fescue; OG = orchard grass; LUC = lucerne; SEM = standard error of the mean.

[2] Values within each row with different subscript letters (a-d) differ ($P<0.05$). Values within each column for each analyte with different subscript letters (x,y) differ ($P<0.05$).

Table 3. Concentrations of glucose and sucrose (g kg^{-1} of dry matter) in regrowth above 7 cm of different species at different time points during the day across sampling period and sampling day.

		Species							
Analyte	**Time**	**DPR**	**TPR**	**TIM**	**TF**	**MF**	**OG**	**LUC**	**SEM**
Glucose	Morning	$13.0^{cde, x}$	$13.5^{de, x}$	$16.0^{e, x}$	$11.1^{bcd, x}$	$8.97^{b, x}$	$9.15^{bc, x}$	$3.87^{a, x}$	0.85
	Noon	$15.2^{cd, xy}$	$15.4^{cd, xy}$	$17.9^{d, x}$	$12.2^{bc, x}$	$13.4^{bc, y}$	$11.0^{b, x}$	$5.07^{a, x}$	
	Evening	$16.4^{d, y}$	$16.1^{cd, y}$	$21.0^{e, y}$	$12.5^{bc, x}$	$12.3^{b, y}$	$10.8^{b, x}$	$5.83^{a, x}$	
Sucrose	Morning	$27.6^{cd, x}$	$34.1^{d, x}$	$22.8^{abc, x}$	$31.3^{d, x}$	$26.2^{bcd, x}$	$16.5^{a, x}$	$18.9^{ab, x}$	1.75
	Noon	$32.9^{bc, y}$	$38.0^{cd, x}$	$27.6^{ab, y}$	$41.7^{d, y}$	$33.1^{bc, y}$	$21.2^{a, y}$	$26.1^{ab, y}$	
	Evening	$41.8^{b, z}$	$52.6^{cd, y}$	$41.9^{b, z}$	$55.5^{d, z}$	$45.7^{bc, z}$	$27.2^{a, z}$	$38.6^{b, z}$	

[1] See footnotes of Table 2.

Table 4. Concentrations of glucose, fructose, sucrose, fructans and total WSC (g kg^{-1} of dry matter) in regrowth above 7 cm at different time points in August and October across species and sampling day.[1,2]

		Time of day			
Analyte	**Period**	**Morning**	**Noon**	**Evening**	**SEM**
Glucose	August	$14.5^{a, x}$	$16.6^{b, x}$	$16.6^{b, x}$	0.45
	October	$7.04^{a, y}$	$9.19^{b, y}$	$10.6^{c, y}$	
Fructose	August	$15.2^{a, x}$	$16.3^{a, x}$	$15.8^{a, x}$	0.40
	October	$9.70^{a, y}$	$11.9^{b, y}$	$13.1^{c, y}$	
Sucrose	August	$31.5^{a, x}$	$39.4^{b, x}$	$50.0^{c, x}$	0.94
	October	$19.2^{a, y}$	$23.6^{b, y}$	$36.6^{c, y}$	
Fructans	August	$51.3^{b, x}$	$46.3^{ab, x}$	$45.1^{a, x}$	1.73
	October	$25.8^{a, y}$	$26.7^{a, y}$	$28.5^{a, y}$	
Total WSC	August	$113^{a, x}$	$119^{a, x}$	$128^{b, x}$	2.47
	October	$62.1^{a, y}$	$71.7^{b, y}$	$89.2^{c, y}$	

[1] WSC = sum of glucose, fructose, sucrose and fructans; SEM = standard error of the mean.

[2] Values within each row with different subscript letters (a-d) differ ($P<0.05$). Values within each column for each analyte with different subscript letters (x,y) differ ($P<0.05$).

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References

Jørgensen L. (2021) Kulhydrater i græsmarksafgrøder. MSc Thesis. Aarhus University, Denmark, 73 pp. Available at: https://pure.au.dk/portal/files/219298394/Specialeafhandling_Louise.pdf

Larsson K. and Bengtsson S. (1983) Determination of readily available carbohydrates in plant material. Methods report No. 22, National Laboratory of Agricultural Chemistry, Uppsala, Sweden.

Pelletier S., Tremblay G.F., Bélanger G., Bertrand A., Castonguay Y., Pageau D. and Drapeau R. (2010) Forage nonstructural carbohydrates and nutritive value as affected by time of cutting and species. *Agronomy Journal* 102, 1388-1398.

Van Eps A.W. and Pollitt C.C. (2006). Equine laminitis induced with oligofructose. *Equine Veterinary Journal* 38, 203-208.

Ecosystem service research in grasslands at 31 experimental farms, networks and demonstration platforms across Europe

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Abstract

Grassland research stations and experimental farms are essential for applied grassland science and related outreach activities. A large proportion of the experiments conducted at these stations aims to test methods to optimize the ecosystem services (ES) delivered by permanent grasslands. We used the framework of the multi-actor research project SUPER-*G* to assess a selection of experiments recently conducted, completed (since 2011), and planned at 31 European experimental research stations, farms, or networks. We further provide an overview of the ES measured at these experiments. Results show that on average two ES were assessed per experiment. The most frequent ES measured were *production* (87% of all experiments), followed by the supporting ES *biodiversity* (59%) and *climate regulation* (33%). Our overview on ES research at European research stations highlights that permanent grassland is a multifunctional ecosystem that provides many benefits to society. Yet, research considering more than two ES is still relatively rare and should thus be strengthened in the future.

Keywords: permanent grassland, ecosystem services, biodiversity, food production, climate change adaptation, research communication

Introduction

Grassland can provide a wide range of essential ecosystem services (ES; Richter *et al.*, 2021). However, permanent grassland (PG) and its ES are threatened by several factors such as abandonment, conversion to arable, and drought (Schils *et al.*, 2020). Furthermore, management intensification changes ES provisioning within PGs by favouring production-related ES over so-called public ES like recreation and cultural values (Allan *et al.*, 2015). Thus, research initiatives all over Europe aim at optimizing the (simultaneous) delivery of (multiple) grassland ES. Research stations such as experimental farms and demonstration platforms are important for applied grassland science and related outreach activities. The experiments conducted at these stations indicate emerging strategies to sustain and promote grassland ES in Europe. Here, we aim to present an overview of current ES research on PGs across European research stations and experimental farms.

Materials and methods

We used the framework of the H2020 project SUPER-*G* 'Developing Sustainable Permanent Grassland Systems and Policies' to assess grassland-focused experiments recently conducted, completed (since 2011), and planned at 31 experimental farms, networks, and demonstration platforms (referred to as *stations* in what follows) across Europe. Thirteen survey partners from twelve countries (CH, CZ, DE, ES, FR, HU, IT, ME, NL, PL, SL, UK) were asked to report details of their station(s) and the associated experiments. Partners included agricultural and ecological research institutions, consultants and extension services. Beside general information such as the grassland types involved in the research, we assessed the main aim of each experiment and the ES measured therein. Main aims were grouped into six categories: (1) *risks and threats*, i.e. the need to adapt PG to risks such as drought or flooding; (2) *biodiversity* including biodiversity assessments, ecological restoration, reintroduction of grazing after abandonment, etc.; (3) *food production and PG management* aiming at improving yields or quality and optimizing PG management via e.g. productive multi-species swards and fertilizer trials; (4) *innovative methods testing and development* such as the use of drones and satellites to estimate yields; (5) *environment* focused on improving environmental performance of PG such as carbon sequestration, greenhouse gas (GHG) mitigation, and erosion control; (6) *other* (e.g. animal welfare). Note, that experimental station representatives were requested to select only one main aim for each experiment. Although this might be difficult for highly interdisciplinary experiments, partners interviewed did not question or criticise this approach, so the method seems to be appropriate and results robust. To translate measurements in the experiments into final ES (Richter *et al.*, 2021), we associated each category with the following: (1) *production* (grass, meat or milk yield); (2) habitat for *biodiversity* (assessments of any taxa); (3) *climate regulation* (e.g. C sequestration, GHG mitigation); (4) *water quality* (e.g. nitrate leaching); (5) *flood control* (e.g. rainwater infiltration and regulation); (6) e*rosion control* (e.g. soil surface protection); (7) r*ecreation and aesthetics* (various cultural values).

Results and discussion

In total, the survey encompassed 70 experiments from 31 stations in five biogeographic zones, i.e. Alpine, Atlantic, Continental, Mediterranean and Pannonian. At each station, experiments were conducted on an average of two (range 1-3) out of six distinct grassland types, classified using the system outlined in Tonn *et al.* (2020). The most frequently researched grassland types were low intensity PG (at 71% of the stations), followed by intermediate intensity PG (48%), and high intensity PG (29%). In addition, frequently renewed PG, unmanaged PG and PG with a high cover of woody species (according to Tonn *et al.*, 2020) were studied at a smaller proportion of sites (16% and less). Work in 77% of stations was focused on conventional (as opposed to organic) PG, including both high and low input systems. At 13% of the stations both conventional and organic PGs were studied, and 10% focused on organic PG. Of the 70 experiments, 22 had been completed (since 2011), 49 were ongoing, and one was about to start. The most frequent main aims of these experiments were *production and management* (47%), *environment* (41%), and *risks and threats* to PG (13%; Figure 1). 29% of the experiments focused on one ES, while the largest proportion (34%) assessed two ES. More ES, either three or four, were measured in 23 and 10% of the experiments, respectively. The maximum of five ES, out of the maximum possible of seven, were studied in only one experiment (1%). At 3% of the experiments, none of the inquired ES were measured as these experiments were focusing on animal welfare. The most frequent ES measured were *production* (87%), followed by *biodiversity* (59%), and *climate regulation* (33%). All other ES were only rarely assessed (Figure 2). Our results underline that grassland research stations do not only examine production, but also consider other crucial ES of PG, which is in line with grassland being a highly multifunctional ecosystem (Allan *et al.*, 2015; Richter *et al.*, 2021).

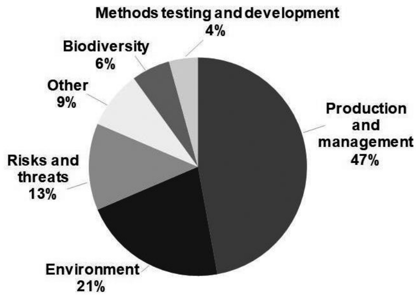

Figure 1. Main aims of 70 grassland experiments at 31 research stations in Europe.

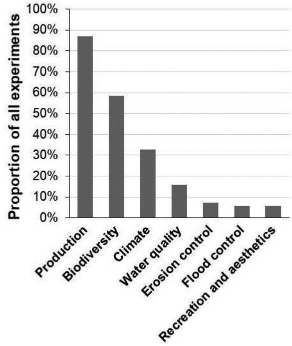

Figure 2. Grassland ecosystem services measured in the 70 experiments.

Conclusions

Our assessment shows that in addition to production-related ES a diverse range of other ES derived from PG are being studied at European research stations. However, only a small proportion of the experiments included in this study were found to have more than two ES being evaluated simultaneously, and cultural services were seldom assessed. Our work suggests that to truly research the multifunctional benefits of management practices, simultaneous monitoring of a wider array of grassland ES should be considered. This would also help assessing potential trade-offs among the many relevant grassland ES. Consequently, despite the promising advancement regarding grassland ES research, we suggest to further strengthen multidisciplinary research efforts on grassland ES multifunctionality.

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References

Allan E., Manning P., Alt F., Binkenstein J., Blaser S., Blüthgen N. ... and Fischer M. (2015) Land use intensification alters ecosystem multifunctionality via loss of biodiversity and changes to functional composition. *Ecology Letters* 18, 834-843.

Richter F., Jan P., El Benni N., Lüscher A., Buchmann N. and Klaus V.H. (2021) A guide to assess and value ecosystem services of grasslands. *Ecosystem Services* 52, 101376.

Schils R.L.M., Newell Price P., Klaus V.H., Tonn B., Hejduk S., Stypinski P. ... and Korevaar H. (2020) European permanent grasslands mainly threatened by abandonment, heat and drought, and conversion to temporary grassland. *Grassland Science in Europe* 25, 553-555.

Tonn B., Bausson C., Ten Berge H., Buchmann N., Bufe C., Eggers S. ... and Newell Price P. (2020) A management-based typology for European permanent grasslands. *Grassland Science in Europe* 25, 412-414.

How professional stakeholders perceive the current and future relevance of grassland ecosystem services in Switzerland

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Abstract

We conducted a (non-representative) online survey in Switzerland to reveal professional stakeholders' perceptions and attitudes towards ecosystem services (ES) provided by permanent grassland. According to the 398 respondents, erosion control, soil fertility, feed production, habitat provision (for biodiversity), and groundwater protection currently represent the most relevant grassland ES. Regarding the future, stakeholders assumed particularly climate regulation (carbon storage), soil fertility, and groundwater protection to increase in relevance. The majority (84%) of respondents stated that grassland ES are insufficiently recognized by society. Almost three quarters of the stakeholders associated grassland management intensification with a loss of ES multifunctionality, and 60% expected higher ES multifunctionality of organic compared to conventional grassland. Our survey revealed that strategies to ensure and to value grassland ES by society and by future agricultural policies might be welcome and supported by stakeholders in Switzerland.

Keywords: agricultural policy, ecosystem service, intensification, multifunctionality, permanent grassland, stakeholder questionnaire

Introduction

Ecosystem services (ES) comprise all goods and services ecosystems provide to humans. They are the basis of our daily food supply and wellbeing. Due to continuing ecosystem degradation and biodiversity loss, we must seek strategies how to sustain ES for future generations (Brondizio *et al.*, 2019). Particularly in countries with a high share of permanent grassland, such as Switzerland, grassland ES play an important role for economy and society. The simultaneous provision of different ES is referred to as multifunctionality, and grasslands are considered particularly multifunctional (Bengtsson *et al.*, 2019). Despite recent advances in grassland ES research (e.g. Allan *et al.*, 2015), there is still a lack of knowledge on how management strategies and environmental changes affect grassland ES (Klaus *et al.*, 2020). Such information is particularly important if agricultural policy aims at promoting ES and multifunctionality in the future. Thus, research regarding the current and future relevance of grassland ES and on their main drivers is urgently needed. For this reason, we launched a survey among professional stakeholders to obtain their opinions on the relevance of ES, the societal recognition of grassland ES and the challenges to maintaining multifunctionality.

Materials and methods

The German-language online questionnaire was sent to professional stakeholders with a link to agriculture or environmental protection in Switzerland, i.e. participants of national agricultural meetings and conferences. In total, 398 stakeholders answered the survey, but due to the unstructured invitation we cannot assess the representativeness of the sample. Emails were sent out on January 15, 2020, and the online link was available until March 16, 2020. The questionnaire consisted of several sections. First, a brief definition of ES was given before stakeholders were asked to rate the overall (1) current and (2) future importance of 14 ES of permanent grassland (Figure 1). This was followed by asking whether ES of permanent grassland were (1) sufficiently recognized by society as a whole, and (2) sufficiently considered in the current Swiss direct payment system. After defining multifunctionality, stakeholders were asked whether (1) intensification of grassland management reduces the multifunctionality of a grassland stand, and whether (2) organically managed grassland has a higher multifunctionality than conventionally managed grassland. Finally, stakeholders were asked about their place of work (*country*) and their field of work (*agriculture* or *nature conservation/ environmental protection* or both or *other*). To test for significant differences among fields of work, a Kruskal-Wallis test was used in combination with a Dunn test with Benjamini-Hochberg correction. For this, Likert-scale responses were converted into numerical data. All *don't know* responses were excluded. Group differences are reported if both tests yielded $P<0.05$. All analyses were done with R.

Results and discussion

Of the 398 participants, 93% worked in Switzerland, and 7% in Germany or Austria. About 66% of the participants worked in the field of *agriculture*, 14% in *nature conservation/ environmental protection* (10% in both, 10% in *other*). Of the 14 given ES, eleven were rated *very important* to *rather important* by over 75% (Figure 1). Two soil functions, erosion control and soil fertility, were considered particularly important, followed by feed production, habitat provision (biodiversity), and groundwater protection. Regarding their future relevance, the six ES rated as currently most relevant were all considered to become more important in the future (Figure 1), except feed production, for which the majority expected no change in relevance. The conservative attitude of stakeholders to rate the future importance of ES similarly to the current relevance was also observed in a previous ES study in the EU (Van del Pol-van Dasselaar *et al.*, 2013). Over 80% of the participants answered that society does not sufficiently recognize permanent grassland ES (42% *no*, 42% *rather no*, 12% *rather yes*, 3% *yes*, 1% *don't know*). There was also a tendency to disagree that ES are sufficiently considered by the current Swiss direct payment system (only responses from people working in Switzerland considered; 16% *no*, 33% *rather no*, 30% *rather yes*, 10% *yes*, 11% *don't know*). Almost three quarters of the participants answered that the intensification of grassland management would reduce its multifunctionality (34% *yes*, 40% *rather yes*, 13% *rather no*, 11%

Current relevance	Erosion control	Soil fertility	Feed production	Habitat (biodiversity)	Groundwater protection	Habitat for pollinators	Nutrient cycling
Very relevant	82	82	72	69	68	55	47
Rather relevant	14	14	22	26	27	37	42
Rather irrelevant	1	2	3	3	3	4	7
Very irrelevant	1	1	1	1	2	1	1
Future relevance							
Increasing	59	72	26	57	70	61	34
Same	38	25	57	41	28	36	60
Decreasing	1	2	16	1	1	1	4

Current relevance	Climate regulation	Biological pest control	Aesthetics	Recreation and Tourism	Traditional cultural landscape	Edible/ medicinal plants	Non-feed biomass
Very relevant	47	42	27	22	20	12	6
Rather relevant	34	44	52	54	42	32	19
Rather irrelevant	14	10	18	20	29	47	52
Very irrelevant	2	2	3	4	8	9	23
Future relevance							
Increasing	73	55	32	33	10	15	16
Same	22	41	59	62	59	68	51
Decreasing	2	2	7	3	29	14	29

Figure 1. Current and future relevance of 14 ecosystem services (ES) in Switzerland, as seen by 398 professional stakeholders (in %). ES listed from highest to lowest values for *very relevant*. Sum is <100% as *don't know* responses are not shown. Grey shading from max (darkest) to min (lowest).

no, 3% *don't know*). However, there were significant differences among stakeholder groups. Stakeholders associated with *nature conservation/ environmental protection*, *agriculture* & *nature conservation/ environmental protection* and *other* answered with 94, 85 and 82% *yes* or *rather yes,* respectively, while stakeholders associated with *agriculture* did so with 66%. In an ES study in Germany, however, Allan *et al.* (2015) found grassland intensification to have a clear negative effect on ES multifunctionality, which is linked to a reduction in plant diversity. Most participants stated that organic grassland has a higher multifunctionality than conventional grassland (24% *yes*, 36% *rather yes*, 16% *rather no*, 22% *no*, 3% *don't know*). There were again significant differences according to stakeholder groups. Those associated with *nature conservation/ environmental protection* answered *yes* and *rather yes* to 92%, while 62% and 51% of the stakeholders associated with *agriculture* and *nature conservation/ environmental protection* and *agriculture* agreed with this, respectively. As studies on the effects of organic grassland farming on multifunctionality are lacking, it is important that this question is being addressed (Klaus *et al.*, 2020).

Conclusions

Our survey revealed that almost all ES of Swiss permanent grassland were perceived as important, underlining the need to protect grassland multifunctionality. We further conclude that despite some differences among stakeholder groups, strategies to ensure, recognize and value grassland ES by society and by future agricultural policies might be widely welcomed.

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References

Allan E., Manning P., Alt F., Binkenstein J., Blaser S., Blüthgen N., ... and Fischer M. (2015) Land use intensification alters ecosystem multifunctionality via loss of biodiversity and changes to functional composition. *Ecology Letters* 18, 834-843.

Bengtsson J., Bullock J.M., Egoh B., Everson C., Everson T., O'Connor T., ... and Lindborg R. (2019) Grasslands - more important for ecosystem services than you might think. *Ecosphere* 10, e02582.

Brondizio E.S., Settele J., Díaz S. and Ngo H. T. (eds.) (2019) *Global Assessment Report on Biodiversity and Ecosystem Services of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services*. IPBES Secretariat, Bonn, Germany.

Klaus V.H., Richter F., Buchmann N., Jan P., El Benni N. and Lüscher A. (2020) Effects of organic farming on ecosystem services and multifunctionality in Switzerland: the ServiceGrass project. *Grassland Science in Europe* 25: 505-507.

Van del Pol-van Dasselaar A., Golinski P., Hennessy D., Huyghe C., Peyraud J.L. and Stienezen M.W.J. (2013) Appreciation of current and future functions of grassland by international stakeholders in Europe. *Grassland Science in Europe* 18, 219-221.

Intense drainage improves N balance in a ley experiment

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Abstract

In Norway, the effect of drainage on grassland yields has received little attention for decades. Low levels of drainage may be a reason for low grassland production. Therefore, a drainage experiment was established in a western Norwegian ley, on a sandy silt soil with a high capacity for water storage. The plots had six- and twelve-metres drain spacing, as well as an undrained treatment. For each drainage treatment there were two or three cuts per year, and fertilization of 190 or 290 kg N yr^{-1} ha^{-1}. Drainage intensity gave a small significant increase in yield. N loss in drainage water increased with drainage intensity. The small herbage yield increase is unlikely by itself to justify drainage, but the drainage installation might still be worthwhile due to increased N efficiency and a more manageable risk of compaction. Precise quantification of the hydrological effects is hard to make due to the inherent soil variability.

Keywords: drain spacing, N-leaching, dry matter yield, N-yield

Introduction

In Norway, soil drainage has received little attention for decades, which may be a reason for low grassland productivity (Haukås and Berger, 2018). Western Norway is a rainy area, with annual precipitation of 2000 mm or more being common. It is a grassland area characterized by small and dispersed fields. Soils are predominantly sandy and silty, mostly of glacial or colluvial origin, and rich in organic matter with 16% classified as organic soil (Lågbu *et al.*, 2018). Drainage is an important intervention to remove excess water from the soil, thus providing a more suitable environment for plant growth and farming operations.

A drier soil is less vulnerable to compaction by farm machinery, although this also depends on the soil type. Sands are usually less vulnerable than silt, clay or organic soils. Silt and peat soils are especially at risk as they have a weak ability to recover from compaction. The consequences of compaction may include smaller pores (Rivedal *et al.*, 2016), stunting roots (Colombi *et al.*, 2018) and restricting the volume available for uptake of plant nutrition. Smaller pores decrease air content and hamper water and gas transport, favouring denitrification (Dobbie and Smith, 2001), and depressing yields. Compaction may thus restrict yields and decrease N-use efficiency. This restricts the farmer, who may have to choose between sub-optimal times for applying fertilizer and harvesting, or risks long-term damage to the soils due to compaction. Both choices result in a lowered productivity.

An increase in subsurface drainage intensity removes excess water more quickly, and might result in higher yields, nitrogen balance and more flexibility in farming operations. One of the objectives of this field experiment was to study the effect of increased drainage intensity on yield and N balance.

Materials and methods

The experiment was conducted in Askvoll, Norway (61°20 N, 5°6 E), from 2014-2017. Average annual precipitation during the experimental period was 2,500 mm yr^{-1} and mean temperature of 8.5 °C. Both temperature and precipitation were roughly 10% higher than the 1991-2020 climatic average for the nearest meteorological station (Fureneset, 6 km distant).

The main soil type was a Mollic to Umbric Gleysol, from sandy silt to silty sand, with a high organic matter content (10-20% LOI) in the topsoil. In the autumn of 2013, drainage pipes were installed 1-1.2

m below the soil surface. Drains were 50 m long and had 0.057 m diameter. Only the top half of the drainage pipe was perforated, and saw dust was used as filter material. Tipping buckets were installed at the pipe outlets to measure discharge. Volume proportional composite water samples were collected every fortnight and analysed for total-N, ammonium and nitrate.

The field was sown in the spring of 2014, with seed mixtures containing different proportions of timothy (*Phleum pratense* L.) for normal two-cut and intensive three-cut regimes (Table 1). Within each cutting regime three different drainage intensities were established: high, with 6 m drain spacing; low, with 12 m drain spacing; and no subsurface drainage. Within each of the drainage intensity treatments two levels of N fertilization were applied on sub-sections: F1 (190 kg N ha^{-1}) and F2 (290 kg N ha^{-1}). A tractor weighing 6.8 Mg passed over the whole area each spring and after each cut, to simulate a realistic amount of traffic for grassland farming. Due to wet conditions in 2017 only two cuts were taken from the 'three-cut' regime treatment.

Herbage yield was measured for three years (2015-2017) from four permanent parallel harvesting plots (18 m^2) within each fertilizer plot. DMY was determined by drying grass samples at 60 °C for 48 h. Samples were analysed by near infrared analysis (NIR) to determine energy and protein concentrations (Fystro and Lunnan, 2006). Nitrogen content was determined by dividing protein concentration by 6.25.

Data from field trials were analysed as a mixed model by multi-factorial analysis using PROC MIXED in SAS (SAS 9.4, SAS Institute Inc., Cary NC, USA). Effects of management, drainage and fertilization were considered as fixed factors.

Results and discussion

DMY were significantly higher from drained (8.68 Mg ha^{-1} yr^{-1}) compared to undrained soils (8.01 Mg ha^{-1} yr^{-1}), but no significant difference was detected between the 6 m and 12 m drain spacings. As a mean of 3 years, DMY increased with the narrower drain spacing at two cuts, but not at three. The two-cut 6 m drain spacing regime used nitrogen more efficiently than the other managements. The nitrogen budget as a mean of 1 May 2016 – 15 April 2017 and 2 May 2017 – 16 April 2017 is given in Table 2. Nitrogen removed through natural drainage between the drains (or in the undrained treatment) remains unquantified.

Only in the two-cut 6 m drain spacing regime does potential evapotranspiration and water in tipping bucket add up to over 50% of precipitation, and thus seepage remains a dominant form of drainage. The differences in drainage between two and three cuts are believed to be mostly due to inherent soil variability. High soil variability between treatments and single drains complicates predicting the quantitative effect of a drainage intervention.

Table 1. Experimental treatments.

	Seed mixture and amount	Fertilizer (kg N ha^{-1})	
		Moderate	High
Two cuts	70% timothy, 20% meadow fescue (*Festuca pratensis* L.), 10% smooth meadow-grass (*Poa pratensis* L.)	110 in spring, 80 after 1st cut	170 in spring, 120 after 1st cut
Three cuts	45% timothy, 15% meadow fescue, 15% smooth meadow-grass, 15% perennial ryegrass (*Lolium perenne* L.) and 10% festulolium (ryegrass type)	100 in spring, 60 after 1st cut, 30 after 2nd cut	140 in spring, 90 after 1st cut, 60 after 2nd cut

Table 2. Nitrogen budget as mean for the hydrological years 2015-2016 and 2016-2017.

Nitrogen (kg ha^{-1} yr^{-1})	12 m drain spacing				6 m drain spacing			
	2 cuts		3 cuts		2 cuts		3 cuts	
Applied	190	290	190	290	190	290	190	290
Removed in yield	95	135	95	127	144	173	94	135
Lost in drain pipes	6	7	7	10	7	14	8	9
Unaccounted for	89	148	88	153	39	103	88	146

Conclusions

The yield increase caused by drainage is likely to be too small to justify the intervention by itself, especially as soil heterogeneity increases the uncertainty of its effect. The possible increased nitrogen efficiency might justify it, by increasing economic and ecological sustainability. Other methods for securing drier soils might be preferrable, such as landscaping the grasslands to favour surface runoff in addition to in-soil drainage.

As more intense drainage also results in drier soil conditions; it may also allow for a greater flexibility for the farmer's timing of field operations, although this is dependent on soil type. This was not tested in the present experiment, but might allow for a more high yielding management with less risk of harming the soil. Testing for this effect in future experiments could give a more realistic picture of the incentives for the farmer to drain her fields.

References

Colombi T., Torres L.C., Walter A. and Keller T. (2018) Feedbacks between soil penetration resistance, root architecture and water uptake limit water accessibility and crop growth – A vicious circle. *Science of the Total Environment* 626, 1026-1035.

Dobbie K.E. and Smith K.A. (2001) The effects of temperature, water-filled pore space and land use on N_2O emissions from an imperfectly drained gleysol. *European Journal of Soil Science* 52, 667-673.

Fystro G. and Lunnan T. (2006) Analysar av grovfôrkvalitet på NIRS. *Bioforsk FOKUS* 1(3), 180-181.

Hansen S., Bernard M.-E., Rochette P., Whalen J.K. and Dörsch P. (2014) Nitrous oxide emissions from a fertile grassland in Western Norway following the application of inorganic and organic fertilizers. *Nutrient Cycling in Agroecosystems* 98, 71-85.

Haukås T. and Berger M. (2018) DRAINIMP – Økonomi i drenering, *NIBIO Rapport* 4 (152).

Lågbu R., Nyborg Å.A. and Svengård-Stokke S. (2018) Jordsmonnstatistikk i Norge. *NIBIO Rapport* 4 (13).

Rivedal S., Riley H., Lunnan T., Børresen T., Øpstad S. and Stürite I. (2016) Verknad av traktorkøyring på engavling og jordfysiske forhold. *NIBIO Rapport* 2 (145).

Ecosystem services provided by wet grasslands through extensive livestock farming

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Abstract

The conservation of wet grasslands is conditioned by the maintenance of extensive livestock farming. Highlighting the ecosystem services they provide appears as a promising way to maintain agricultural activities. The Regional Natural Park 'Marais du Cotentin et du Bessin' is one of the pilot sites of a national project aimed at maintaining farmers in wetlands. A multi-service approach was performed to compare a set of wet grassland habitats. We evaluated services belonging to three categories: supporting services (floristic biodiversity and conservation value of habitats), regulating services (soil carbon stocks and water quality), and provisioning services (productivity and fodder quality). Contrasted habitats provide contrasting bundles of services, and some trade-offs (e.g. water quality decreasing in some habitats with high soil carbon stocks), or synergies (e.g. floristic diversity increasing forage digestibility) have been identified. It is difficult to improve the set of services provided by each habitat but it appears more relevant to maintain a diversity of habitats resulting from different water level and management in order to obtain ecosystem services of high-level at the landscape scale.

Keywords: wet grassland, plant diversity, soil C stocks, water quality, forage quality

Introduction

Wetland areas have declined during the last century in Europe mainly as the result of agriculture intensification. Wet grasslands that have not been drained are still threatened by abandonment which would lead them to become wet woodlands. Wet grasslands represent areas that are complicated to exploit because of the low bearing capacity of the soils and the difficulty of coping with early or prolonged flooding. Their conservation depends closely on the maintenance of extensive livestock farming (Lemauviel-Lavenant and Sabatier, 2017). Nevertheless, wet grasslands provide quality fodder for livestock (Tasset *et al.*, 2019). They constitute high value habitats for plants and animals (Hayes *et al.*, 2015), particularly for breeding waders. They are involved in water quantity and quality regulation (Maltby and Acreman, 2011). They also have an important role in climate regulation through the huge carbon stocks they store in their soils (Adhikari *et al.*, 2009). These different ecosystem services should not be studied in isolation from each other. Reconciling agricultural and environmental benefits is fundamental to preserve both. The analysis of multiple ecosystem services is therefore a promising way to maintain agricultural activities in wet grasslands to ensure their conservation and the services they provide.

Material and methods

The Regional Natural Park 'Marais du Cotentin et du Bessin' is located in Normandy, France and consists of huge marsh areas mainly exploited by cows, by grazing or mowing. This Park was chosen as one of the pilot sites in a national project aimed at maintaining farmers in wetlands. A set of wet grasslands was sampled in 2020 to compare nine locally common habitats (3 grasslands for each) either in peat soils or peaty gley soils. All the grasslands are managed in an extensive way without fertilization. For each grassland, we evaluated indices belonging to three categories of ecosystem services. (1) Supporting services: the cover of plant species was estimated in four 1 m^2 quadrats to calculate plant diversity indices. An index of patrimonial value was obtained on the basis of the conservation status of species and their specificity to wetlands. (2) Regulating services: soils were sampled (4×0-15 cm) to measure organic matter (OM) content by mass loss on ignition. Ionic contents, as indices of soil water quality, have been assessed in water extracts (5 g soil/10 ml pure water). (3) Provisioning services: above-ground biomass was cut at 5 cm in mid-June or mid-July according to farmer practices to assess production of the first cut, crude protein content, pepsin-cellulase digestibility and an index of energy (net energy for dairy production) (Baumont *et al.*, 2007).

Results and discussion

Bundles of services are presented here as flowers in which each petal corresponds to an index of ecosystem service (Figure 1). A great contrast emerges between the habitats. Peat habitats are obviously characterized by the highest C stocks (OM reaching to 52% in the topsoil for *Cirsio-Shoenetum*). Among the peaty gley soil habitats, the most intensive ones (twice mown or mown then grazed) have the highest forage quality but the lowest first-cut production but, unlike the other habitats, this does not represent the annual production. They are also the ones that provide the lowest supporting services. Among supporting services, the plant diversity indices do not appear to be positively correlated with the patrimonial value of the plant community. Indeed, in the peat soils, stresses, mainly anoxia and oligotrophy, act as a filter which only allows specialist species to develop, thereby reducing diversity, and consistent with the humped back model theory (Grime, 2001). Among the forage quality indices, all positively correlated, the crude protein content is negatively correlated with the production obtained in the first cut (r=-0.69, P<0.001). Another trade-off appears among regulation services. For all habitats, water quality can be considered as very good when considering phosphate and ammonium contents. Sulfate contents show very high values for a set of peat soil habitats. The very high values obtained may indicate a mineralization of the soil organic matter and thus the loss of C stocks which generally appears when the water decreases in peatlands (Blodau *et al.*, 2004). Nitrate content was high

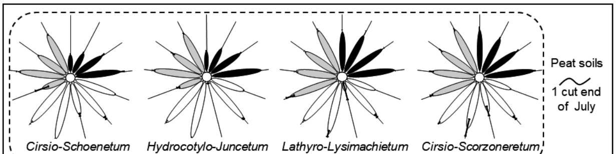

Figure 1. Bundles of ecosystem services provided by a set of different habitats of wet grasslands. Each 'petal' corresponds to an index of ecosystem service (mean, se, n=3 per habitat). NB: As high ionic contents may be considered as dis-services, the graduation of these petals is reversed.

in the *Hordeo lolietum* which corresponds to the less humid habitat. An interesting synergy between plant diversity and forage quality was highlighted by significant positive correlations between all the floristic diversity indices and forage digestibility and energy. Higher diversity is often associated with a higher proportion of forbs, which are characterized by higher digestibility than grasses in late-cut situations encountered in marshes (Tasset *et al.*, 2019).

Conclusions

Neither of the habitats can provide a perfect bundle of services as trade-offs exist among services. Each habitat is the result of edaphic conditions, water constraints (partly driven by the management of water regulation structures) as well as a long history of management. Maintaining extensive management and a high water level, which condition the conservation of specific habitats providing high levels of certain services, appears essential.

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References

Adhikari S., Bajracharaya R.M. and Sitaula B.K. (2009) A review of carbon dynamics and sequestration in wetlands. *Journal of Wetlands Ecology* 2, 42-46.

Baumont R., Dulphy J.P., Sauvant D., Meschy F., Aufrère J. and Peyraud J.L. (2007) Alimentation des bovins, ovins et caprins. Besoins des animaux. Valeurs des Aliments. Quæ, Versailles, France.

Blodau C., Basiliko N. and Moore T.R. (2004) Carbon turnover in peatland mesocosms exposed to different water table levels. *Biogeochemistry* 67, 331-351.

Grime J.P. (2001) *Plant strategies, vegetation processes and ecosystem properties*. Wiley, Chichester, UK. 417 pp.

Hayes M., Boyle P., Moran J. and Gormally M. (2015) Assessing the biodiversity value of wet grasslands: can selected plant and insect taxa be used as rapid indicators of species richness at a local scale. *Biodiversity and Conservation* 24, 2535-2549.

Lemauviel-Lavenant S. and Sabatier R. (2017) Multiple services delivered by semi-natural lands depend on breeding activities. *Productions Animales* 30, 351-362.

Maltby E. and Acreman M.C. (2011) Ecosystem services of wetland: pathfinder for a new paradigm. *Hydrological Sciences Journal* 56, 1341-1359.

Tasset E., Boulanger T., Diquélou S., Laîné P. and Lemauviel-Lavenant S. (2019) Plant trait to fodder quality relationships at both species and community levels in wet grasslands. *Ecological Indicators* 97, 389-397.

Fertilizer regime modifies grassland sensitivity to interannual climate variability

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Abstract

Improved understanding of the interactions between management practices and climate variability is critical for the development of sustainable grassland management and the provision of bundles of ecosystem services in a changing environment. Here we used a long-term fertilizer field trial to examine the impacts of climate variability (annual temperature, rainfall) on an upland grassland subjected to a gradient of nutrient availability. We tested for the effect of climatic indices, fertilizer regime, and their interactions, on annual biomass production, forage quality (crude protein content, digestibility) and plant diversity (species richness, equitability). During the 15-year study period we recorded significant interannual variation in both climate and grassland properties. We found that fertilizer treatments, mean annual temperature and annual precipitation all affected the grassland variables in this study, but interactions between climate and management treatment were generally limited. Contrary to expectations, interactions were driven by the PK rather than the NPK treatment. These results highlight the importance of management for projected responses to future climate change in models of grassland function.

Keywords: upland grasslands, production, forage quality, rainfall variability, nitrogen

Introduction

Managed grasslands are an integral part of livestock farming systems and provide multiple provisioning, regulation and cultural services, as well as representing reservoirs of plant and animal biodiversity (Bengtsson *et al.*, 2019). Increasing evidence suggests that the type and intensity of grassland management practices (mowing/grazing, fertilizer inputs) may condition grassland yield responses to variation in abiotic drivers such as water availability via changes in plant community composition and associated plant traits (Bharath *et al.*, 2020). For example, high nitrogen inputs could enhance grassland sensitivity to drought due to the increased water demand in high biomass systems or root:shoot allocation patterns adapted to light, rather than below-ground resource acquisition. This is of particular relevance in the context of increasing periods of drought/water stress and climate variability predicted with ongoing climate change (IPCC, 2021). Improved understanding of the interactions between management practices and climate variability is a necessary step in the development of sustainable grassland management and the identification of agricultural 'best practices' to improve the resistance of grassland forage production in a changing environment. In the present work, we investigate the interactive effects of fertilizer regime and climate variability (temperature, rainfall) on yield, forage quality and plant diversity in an upland grassland subjected to a gradient of nutrient availability treatments over a 15-year period, and examine whether high N inputs modify grassland response patterns to interannual climate variation.

Materials and methods

The field experiment is located in the Massif-central region in France (45°43023″ N, 03°1021″ E, 880 m a.s.l., mean annual temperature 8.7 °C, annual rainfall 770 mm) on a Cambisol soil, and forms part of the ANAEE-F ACBB long-term agroecosystems management trial set up onsite in 2005. Prior to the experiment establishment, the site was managed for hay production with mineral fertilizer inputs (average 200 kg N ha^{-1} yr^{-1}) for over 10 years. In 2003 and 2004, the site was mown three times per year without any fertilizer input. From 2005, three treatments were applied: NPK (251 kg N ha^{-1}, 28 kg P ha^{-1}, 177 kg K ha^{-1}); PK (21 kg P ha^{-1}, 128 kg K ha^{-1}); and 'None' (no fertilization), with four field repetitions (field size 400 m^2). Each treatment is mown three times per year and fertilizer application is fractioned, with inputs in early spring and then after the first and the second cuts. At each cut, biomass is sampled at a height of 5.5 cm above soil surface (four quadrats of 0.36 m^2 per field), oven-dried (60 °C for 48 h) and weighed. Dry samples are then ground and analysed for total N content (Dumas method) and dry matter digestibility (DMD) with near infrared spectroscopy (NIRS, Foss). Botanical surveys are carried out each year in May using the transect method to determine species presence/absence at 40 points per field. In the present study, we analysed data for 2006-20. Biomass production, DMD and crude protein content (N*6.25) were expressed as annual values based on the three cuts per year and weighted (by mass) means. Species relative frequency was calculated per field, and within-plot species richness (RS) and evenness were estimated (Pielou, 1972) (no data for 2009). Annual precipitation and mean air temperature data were obtained from an onsite meteorological station (INRAE CLIMATIK). Effects of treatment (fixed factor), climatic variable (covariate) and any interactions were assessed using GLM models.

Results and discussion

Climate indices varied over the 15-year study period; mean annual temperature ranged from 7.2-9.8 °C (mean 8.77, CV 7.9%), whereas annual rainfall ranged from 585-990 mm (mean 789 mm, CV 13.2%). Warmer years also tended to be drier years (marginally significant negative correlation between the two climatic indices). During the study, the NPK treatment showed consistently higher production and forage quality, but lower diversity (evenness) compared to the PK and 'No Fertilizer' treatments (Table 1).

Increasing mean annual temperature was generally associated with a decrease in biomass production and crude protein content, but an increase in grassland diversity (species number, evenness) (Table 1, Figure 1). However, the magnitude of temperature effects on biomass production varied depending on fertilizer treatment, with greater temperature-induced decreases recorded in the PK treatment compared to NPK or 'No Fertilizer' (Figure 1).

Table 1. Directionality of responses of grassland properties to climatic variation and fertilizer treatment.[1]

Variable	Mean annual temperature	Total annual rainfall	Fertilizer treatment
Biomass production (g/m^2)	(Figure 1)	↗	NPK > PK > None
Crude protein (%)	↘	(Figure 1)	NPK > PK = None
Dry matter digestibility (%)	n.s.	↘	NPK < PK = None
Species number	↗	↘	NPK < PK < None
Evenness	↗	(Figure 1)	NPK < PK = None

[1] Results represent significant main effects based on GLM analysis (no results are presented for climate variables where fertilizer treatments interact with climate, cf. Figure 1; n.s., non-significant climate effects). Fertilizer treatment rankings apply only for cases with no Tmt × Climate interactions.

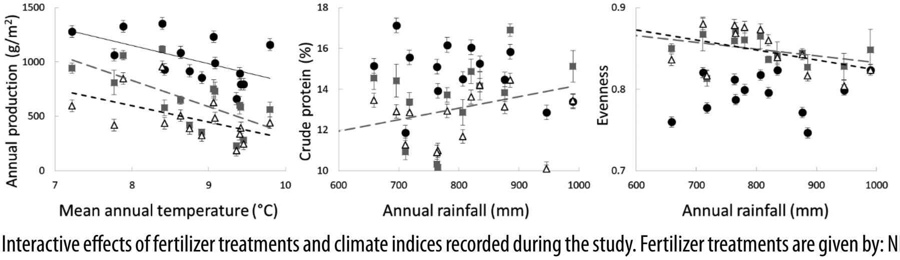

Figure 1. Interactive effects of fertilizer treatments and climate indices recorded during the study. Fertilizer treatments are given by: NPK, black filled circles; PK, grey filled squares, 'None', open triangles.

Digestibility showed no response to temperature, in line with results of previous studies (Dumont *et al.*, 2015). In parallel, annual rainfall had significant positive effect on biomass production but a negative effect on forage digestibility and species richness (Table 1). Effects of annual rainfall on crude protein content and evenness varied depending on fertilizer treatment: crude protein content increased with increasing rainfall in the PK treatment alone, whereas evenness showed a negative relationship with annual rainfall in the PK and 'No Fertilizer' treatments (Figure 1). Contrary to expectations, all interactions recorded between fertilizer treatment and climate index were driven by the PK rather than the NPK treatment. This result may reflect an increased abundance of legume species in the PK treatment, as *Trifolium repens* is known to be sensitive to rainfall deficit (Komainda *et al.*, 2019). Future work should examine responses at different temporal scales and the possible role of belowground responses and species asynchrony in buffering grassland properties against climatic variability.

Conclusions

Interactions between fertilizer treatment and climatic variables assessed at the annual scale were not confined to one particular grassland property or climatic index, but remained relatively limited. In general, interactions were driven by responses in the PK treatment rather than the NPK treatment, possibly linked to greater sensitivity of legumes to climatic variation.

References

Bengtsson J., Bullock J.M., Egoh B., Everson C., Everson T., O'Connor T., O'Farrell P.J., Smith H.G. and Lindborg R. (2019) Grasslands – more important for ecosystem services than you might think. *Ecosphere* 10(2), e02582.

Bharath S., Borer E.T., Biederman L.A., Blumenthal D.M., Fay P.A., Gherardi L.A., Knops J. M.H., Leakey A.D.B., Yahdjian L. and Seabloom E.W. (2020) Nutrient addition increases grassland sensitivity to droughts. *Ecology* 101(5), e02981.

Deroche B., Pradel P. and Baumont R. (2020) Long-term evolution and prediction of feed value for permanent mountain grassland hay: Analysis of a 32-year data set in relation to climate change. *Grass Forage Science* 75, 18-27.

Dumont B., Andueza D., Niderkorn V., Lüscher A., Porqueddu C. and Picon-Cochard C. (2015) A meta-analysis of climate change effects on forage quality in grasslands: specificities of mountain and Mediterranean areas. *Grass and Forage Science* 70, 239-254.

Komainda M., Küchenmeister K., Küchenmeister F., Breitsameter L., Wrage-Mönnig N., Kayser M. and Isselstein J. (2019). Forage legumes for future dry climates: Lower relative biomass losses of minor forage legumes compared to *Trifolium repens* under conditions of periodic drought stress. *Journal of Agronomy and Crop Science*. 205, 460-469.

Pielou E.C. (1972) Niche width and niche overlap: a method for measuring them. *Ecology* 53, 687-692.

Effect of nitrogen fertilization and cutting height on greenhouse gas exchange on a boreal grassland

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Abstract

Grasslands have a potential for increased carbon (C) sequestration through enhanced biomass production by optimizing the nitrogen (N) fertilization rate or cutting height. We investigated the effect of these factors on the greenhouse gas (GHG) exchange including carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O) on a timothy-meadow fescue mixture over two years (2020-2021). Two intensively managed experiments were established on mineral soil in Central Finland in 2018. The effect of N-fertilization was studied at levels of 0 (= no N fertilization), 150, 300 kg N ha^{-1} year^{-1} and at the cutting height at 6 and 12 cm over three harvests at silage stage. Greenhouse gas exchange was measured weekly during the growing season (May - September) by chamber methods and biweekly during winter by chamber or snow gradient method from five replicate plots on each treatment. N-fertilization increased the N_2O emissions but also CO_2 uptake. CH_4 exchange was insignificant in all treatments. Unfertilized plots were net sources of GHGs in both years while fertilized plots were net sinks during the first year but small net sources in the following year. Higher cutting height increased the CO_2 uptake in the first year, but the opposite was observed the following year as a result of weather conditions.

Keywords: grass, carbon, nitrogen, greenhouse gas, emission

Introduction

Agricultural soils, especially perennial grasslands have a potential for carbon sequestration through increased soil C input. N-fertilization increases soil C storage in intensively managed grasslands due to changing soil microbial activity and C allocation of plant (Poeplau *et al.*, 2018, Fornara *et al.*, 2013). On the other hand, N-fertilization can increase greenhouse gas N_2O release from soil as well as an effect of increased decomposition rates of soil organic matter (SOM). Higher cutting height increases the above ground C input to soil and can also affect the root biomass (Thornton and Millard, 1996). In addition to these effects, both measures have an impact on the grass yield and consequently on the animal production per ha. Currently there is urgent need for assessing the implication of climate friendly cultivation techniques on fluxes of CO_2, N_2O and CH_4 together with their effect on grass productivity.

Materials and methods

The study site is located in eastern Finland (63°09'N, 27°20'E). Soil type is sandy loam with 6% organic matter in the 0-20 cm soil layer. There were two randomized complete block design experiments with five replicate plots for each treatment. The swards were established in 2018 with a mixture of timothy (*Phleum pratense* L. and meadow fescue *Festuca pratensis* Huds.) We studied the N-fertilization at levels of 0, 150, 300 kg N ha^{-1} year^{-1} and the effect of cutting height at 6 and 12 cm with annual N-fertilization level of 240 kg N ha^{-1}. The grass harvest to silage stage was performed three times over the growing season (May – September) and harvested dry matter yields were determined.

Annual greenhouse gas exchange was measured from May 2019 until April 2021. During snow-free seasons, CH_4 and N_2O emissions were determined using a dark static chamber (60×60×30 cm) method with permanent collars (60×60×20 cm) installed in soil (Lind *et al.*, 2019, 2020). CO_2 exchange (net ecosystem exchange and ecosystem respiration) was measured during the snow-free season with a transparent polycarbonate chamber and opaque aluminium chamber (60×60×30 cm), equipped with a fan and an ice-water cooling system to keep the chamber temperature close to the prevailing air temperature. An infrared gas analyser (LI-COR, 850) was used to analyse the CO_2 concentrations in the chamber. Air temperature and photosynthetically active radiation (PAR) inside the chamber were recorded during the measurements. N_2O and CH_4 emissions were determined by the dark static chamber method. During the measurement four gas samples were taken from the headspace of the chamber from 5 to 35 minutes after closing and the gas concentrations were analysed with a gas chromatograph (GC) (7890B GC, Agilent Technologies, USA). Instantaneous fluxes were calculated from the change in the gas concentration in the chamber headspace. Diurnal CO_2 exchange was modelled using PAR and other environmental variables (Lind *et al.*, 2020). During snow-covered seasons, the snow gradient method (Maljanen *et al.*, 2003) was used to determine CO_2, CH_4 and N_2O emissions through the snowpack.

Results and discussion

Weather conditions during the studied years were considerably different. During the first growing season (1 May-30 Sept) the precipitation was clearly lower than the long-term average (LTA, 309) but temperature sum was close to LTA (1214). In the second growing season both temperature and precipitation sum were higher than LTA (Table 1).

The different weather conditions affected the total GHG balance and crop yield. The preliminary results of the total balance, including annual greenhouse gas emissions and crop yield are shown in Figure 1. The results show a large variation between the two years with contrasting weather conditions. Therefore, we cannot make strict conclusions how the N-fertilization rates will change the total balance. However, plots without any additional N-fertilization were both net GHG/C sources and produced poorly during both years despite the differences in weather conditions.

Table 1. Temperature sum and precipitation sum during the periods of five months (1 May-30 Sept) in years 2019 and 2020.

Year	Temperature sum (°C)	Precipitation sum (mm)	Δ Temperature[1]	Δ Precipitation[1]
2019	1,248	202	34	-107
2020	1,325	344	111	35

[1] The difference (Δ) between seasonal temperatures and long-term average (1981-2010) values are also shown.

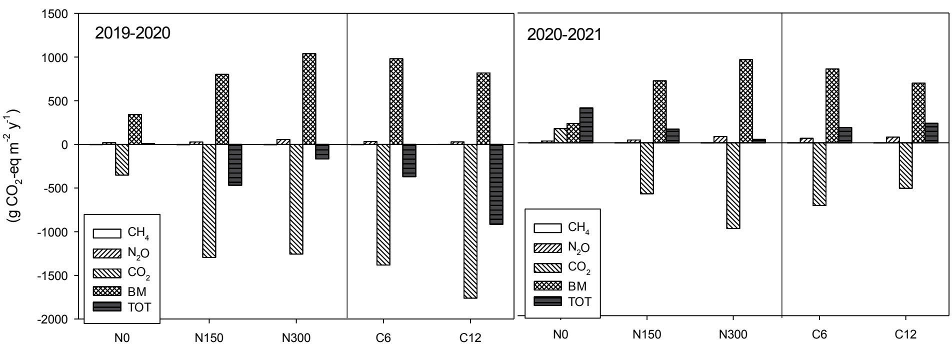

Figure 1. GHG and carbon balance on the studied site in fertilization experiment (N0, N150 and N300, corresponding N-fertilization at levels of no N-fertilization, 150 and 300 kg N ha^{-1} $year^{-1}$) and in cutting height experiment (C6 and C12, corresponding cutting height 6 and 12 cm) during the two years (from May to April). Bars show the annual balance as CO_2 equivalents for each component (CH_4 = methane, N_2O = nitrous oxide, CO_2 = net carbon dioxide exchange, BM = biomass and TOT = total balance). Statistical differences between years and treatments have not yet been calculated for these preliminary data.

Conclusions

We conclude that GHG balance of a boreal grassland is highly dependent on the weather conditions during the growing season. Based on these first two contrasting years no clear conclusions can yet be made how cutting height or N-fertilization rate affects the GHG balance of a boreal grassland. The final conclusion will require results of the third production year as well as a renovation year. Net CO_2 exchange and yield C played a major role in the GHG/C balance, whereas the role of N_2O was minor and the role of CH_4 was negligible.

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References

Fornara D.A., Banin L. and Crawley M.J. (2013) Multi-nutrient vs nitrogen-only effects on carbon sequestration in grassland soils. *Global Change Biology* 19, 3848-3857.

Lind S.E., Virkajärvi P., Hyvönen N.P., Maljanen M., Kivimäenpää M., Jokinen S., Antikainen S., Latva M., Räty M., Martikainen P.J. and Shurpali N.J. (2020). Carbon dioxide and methane exchange of a perennial grassland on a boreal mineral soil. *Boreal Environment Research* 25, 1-17.

Lind S.E., Maljanen M., Hyvönen N.P., Kutvonen J., Jokinen S., Räty M., Virkajärvi P., Martikainen P.J. and Shurpali N.J. (2019) Nitrous oxide emissions from perennial grass cropping systems on a boreal mineral soil. *Boreal Environment Research* 24, 215-232.

Maljanen M., Liikanen A., Silvola J., Martikainen P.J. (2003) Measuring N_2O emissions from organic soils with closed chamber or soil/snow N_2O gradient methods. *European Journal of Soil Science* 54, 625-631.

Thornto B. and Millard P. (1996) Effects of severity of defoliation on root functioning in grasses. *Journal of Range Management* 49, 443-447.

Poeplau C., Zopf D., Greiner B., Geerts R., Korvaar H., Thumm U., Don A., Heidkamp A. and Flessa H. (2018) Why does mineral fertilization increase soil carbon stocks in temperate grasslands? *Agriculture, Ecosystems and Environment* 265, 144-155.

Productivity and regrowth of grasses and legumes for biorefining of protein – effects of defoliation and fertilizer regimes

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Abstract

Productivity of perennial plants integrates photosynthesis, soil nitrogen (N) uptake and internal remobilization, but remains poorly understood for different species subjected to fertilizer and defoliation regimes. A field experiment started in 2019 on sandy soil in Denmark with either fertilized grasses (perennial ryegrass, tall fescue), unfertilized legumes (alfalfa, red clover) or their fertilized mixture (grass-legume), each defoliated at high (2 weeks), medium (4 weeks) or low (6 weeks) frequency intervals, at either 7-9 or 12-14 cm height. Differences between means for the first production year 2020 were evaluated by mixed-effects model fitted to annual biomass and N content. The largest biomass was obtained by tall fescue (11.8-14.2 Mg ha^{-1}) defoliated at medium to low frequency and grass-legume mixture (12.5-13.3 Mg ha^{-1}) defoliated at medium to low frequency, regardless of N fertilization and defoliation height. For N contents, the systems with red clover (395-440 kg N ha^{-1}) and grass-clover mixture (360-400 kg N ha^{-1}) defoliated at high to medium frequency were significantly more productive than the others. This study provides novel insights in perennial productivity modulated in dynamic terms by management and suggests improved integration of environmental and economic sustainability of perennial systems targeting biorefining of feed protein.

Keywords: aboveground, biomass, carbon, multi-factor, nitrogen, tall fescue, seasonal

Introduction

Perennial herbaceous plants after defoliation follow one or a combination of two 'reserve-dependent' regrowth strategies: either to photosynthesize with their remaining leaves, or halt root growth and remobilize stored carbohydrates when defoliation is severe. Productivity and nutritive values thus depend on optimal defoliation and fertilization regimes that modify the source-sink balance to promote best carbon (C) and nitrogen (N) assimilation and allocation for rapid compensatory regrowth (Wang *et al.*, 2021). The main objective of this work was to quantify the effect of species, fertilization, cutting frequency and height of cut on productivity and regrowth of perennial grasses and legumes targeting biorefining of feed protein.

Materials and methods

A field experiment started in 2019 on sandy loam soil in Denmark with grasses (perennial ryegrass, tall fescue) fertilized with 300 or 500 kg N ha^{-1} yr^{-1}, unfertilized legumes (alfalfa, red clover), or their mixture (grass-legume) receiving 300 kg N ha^{-1} yr^{-1} as split amounts after each defoliation with high, medium or low frequency at either 7-8 or 10-12 cm height. Total number of treatments was thus 42 (Table 1). Management of irrigation, plant protection and other nutrients followed optimal practice. Each treatment was laid out on 12×1.5 m plots randomized in four blocks (replicates) separated by 6 m. Field ploughing was on 15 March 2019 and sowing on 15 May. Plots were cut with a plot harvester (Haldrup F-55, Germany) according to height treatment on 7 Aug; thereafter, until October 2019, plots were managed to ensure plant establishment. In 2020, the first production year, treatments proceeded according to the plan (Table 1). Representative biomass samples at each harvest were dried at 60 °C to constant weight, ground, and subsamples (2 g) analysed for N content in an elemental analyser.

Table 1. List of the main treatments and their levels in the field experiment in Denmark.

Plant species/system:		**Nitrogen (N) fertilizer (kg ha^{-1} year^{-1}):**	
G1	Perennial ryegrass (*Lolium perenne* var. Betty)	N0	0 (L1 and L3)
G2	Tall fescue (*Festuca arundinacea* var. Swaj)	N1	300 kg N ha^{-1} (G1, G2 and L2)
L1	Alfalfa (*Medicago sativa* var. SW Nexus)	N2	500 kg N ha^{-1} (G1 and G2)
L2	Grass-legume mix (G1+G2+ L1+ L3)		
L3	Red clover (*Trifolium pratense* var. Taifun)		
Defoliation frequency:		Defoliation height:	
F1= 2 weeks, F2=4 weeks, F3=6 weeks		H1=7-8 cm or H2=10-12 cm	

Mean annual biomass yield (dry matter basis) and N contents in 2020 were separated by treatment using a linear mixed-effects model:

$$Y_{ijkm} = \mu + P_i + F_j + H_k + N_m + (P_i \times F_j \times H_k \times N_m) + B_n + e_{ijkm}$$

where Y is either annual biomass or N content, μ is the overall mean, P, F, H and N are effects of, respectively, plant species, defoliation frequency, height and N fertilization, i, j, k and m are their levels (Table 1), B_n is random effect of block (n=4) and e is residual variation.

Results and discussion

For both biomass and N contents for the first production year, all single factors were highly significant (Table 2). For biomass yield, defoliation frequency significantly interacted with defoliation height, with plant species also added to this interaction, whereas interactions with N fertilizer were close to, but not, significant. For N contents, significant interactions involved defoliation frequency × N fertilizer, including plant species and defoliation height added to three-way interactions.

The statistical model depicted individual differences between treatments, with statistically similar results for many treatments in both biomass and N contents, but the high defoliation frequency treatments were not among the most productive. Instead, the greatest biomass values were obtained by tall fescue (11.8-14.2 Mg ha^{-1}) and grass-clover mixture (12.5-13.3 Mg ha^{-1}), all harvested at 4 and 6 weeks frequency, although red clover harvested at 2 or 4 weeks was in the adjacent group (11 Mg ha^{-1}). On the other hand, the systems yielding statistically the lowest biomass (4-7 Mg ha^{-1}) were those involving perennial ryegrass harvested mostly at 6 weeks and alfalfa at all defoliation frequencies, without apparent effect of defoliation height and N fertilization.

Table 2. Significance of the treatment factors involved in the field experiment in Denmark.

Biomass yield				**Nitrogen content**			
Factors	**F value**	**Pr(>F)**		**Factors**	**F value**	**Pr(>F)**	
Species	153.8	$<2.2\times10^{-16}$	***	Species	170.9	$<2.2\times10^{-16}$	***
Frequency (of cut)	65.2	$<2.2\times10^{-16}$	***	Frequency (of cut)	82.6	$<2.2\times10^{-16}$	***
Nitrogen (fertilizer)	22.2	6.5×10^{-6}	***	Nitrogen (fertilizer)	131.6	$<2.2\times10^{-16}$	***
Height (of cut)	42.0	1.9×10^{-9}	***	Height (of cut)	23.0	4.5×10^{-6}	***
Frequency:Height	6.6	0.0018	**	Frequency:Height	3.5	0.0322	*
Species:Frequency:Height	2.6	0.0099	**	Species:Frequency:Height	2.1	0.0431	*
Frequency:Nitrogen (fertilizer)	2.4	0.0929	.	Frequency:Nitrogen (fertilizer)	3.3	0.0393	*
Species:Nitrogen (fertilizer)	3.7	0.0555	.	Species:Frequency:Nitrogen (fertilizer)	3.7	0.0379	*

High defoliation frequency (2 and 4 weeks) played a significant and promoting role for N content, with the largest N amounts obtained by red clover (395-440 kg N ha^{-1}) and grass-clover mixture (360-400 kg N ha^{-1}), defoliated at both 2 and 4 weeks, which did not differ significantly from tall fescue defoliated at 4 or 6 weeks (300-362 kg N ha^{-1}). Interestingly, these amounts for tall fescue were obtained at fertilizer level N1 (300 kg N ha^{-1}), whereas at N2 (500 kg N ha^{-1}) the amounts were significantly lower (260-275 kg N ha^{-1}), which indicates low efficiency of this grass for rapid N uptake during regrowth, despite ample N available from fertilizer. Other studies have shown that when nutrients are available, plants can preferentially allocate more carbohydrate to roots for storage to balance sink competition between newly expanded leaves and roots, as found for grass for *Stipa*, a genus similar to *Festuca* (Zhang *et al.*, 2021).

The spring cut of all systems contributed notably (5-26%) to the annual biomass and the following two cuts had the largest contribution, but only for medium and low defoliation frequency, cumulating up to 65 and 90%, respectively (Figure 1). This information is relevant for the design of the biorefinery requiring stable and notable supply of biomass.

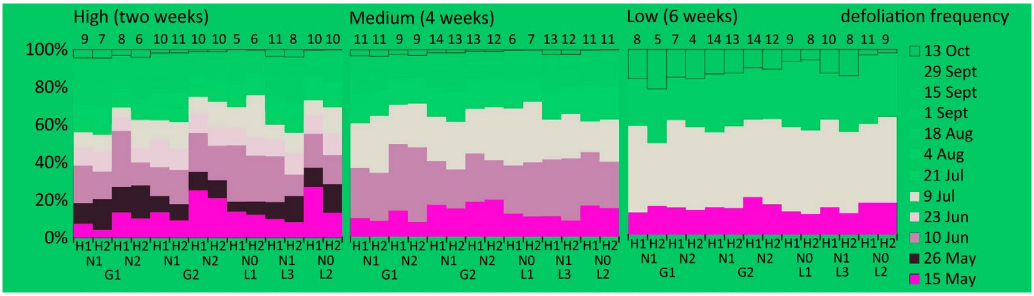

Figure 1. Contribution of defoliation in annual biomass of the systems for the first production year. Treatment codes on the x-axis are shown in Table 1.

Conclusions

Tall fescue and grass-clover mixtures, harvested at 4 or 6 week intervals, were the most productive in terms of biomass, whereas red clover harvested at 2 to 4 weeks yielded the largest N contents. The 2021 data will reveal the robustness of these findings.

Acknowledgements

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References

Wang R., Cresswell T., Johansen M.P. *et al.* (2021) Reallocation of nitrogen and phosphorus from roots drives regrowth of grasses after defoliation under deficit irrigation and nitrogen enrichment. *Journal of Ecology* 109, 4071-4080.

Zhang Z., Gong J., Li X., *et al.* (2021) Underlying mechanism on source-sink carbon balance of grazed perennial grass during regrowth: Insights into optimal grazing regimes of restoration of degraded grasslands in a temperate steppe. *Journal of Environmental Management* 227, 111439.

Effects of stabilized urea fertilizer on nitrate concentration in fresh grass and on silage quality

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Abstract

Stabilized urea (SU) fertilizer has lower NH_3 emissions than conventional urea and lower N_2O emissions than calcium ammonium nitrate (CAN), but its effects on grass and silage quality are not fully understood. High nitrate levels in ensiled grass, especially at later harvests, have been associated with poor fermentation characteristics. This study compared the effects of SU with CAN on the nitrate concentration and buffering capacity (BC) of fresh grass, and on subsequent silage quality. Nitrate levels in fresh grass samples (of <1000 mg kg^{-1} were observed when the ratio of Nitrate (mg kg^{-1}): Yield (kg DM ha^{-1}) was less than approximately 0.30 (in all Early-season, most Mid-season but no Late-season samples for ensilement). Herbage nitrate levels were high in late season, but did not differ between CAN and SU. Nitrate levels in Late-season fresh grass were very strongly correlated (r=0.82: P<0.001) with crude protein and moderately correlated (r=0.52: P=0.010) with BC while there were no strong correlations between nitrate levels and silage quality parameters. Fertilizer type had no effect on silage quality parameters including pH, ethanol, water soluble carbohydrates or dry matter digestibility.

Keywords: stabilized urea, nitrate levels, grass quality, silage quality

Introduction

The application of N fertilizers to agricultural soils is a major source of N_2O emissions which are a potent greenhouse gas (GHG) (Roche *et al.*, 2016). Whereas calcium ammonium nitrate (CAN) emits more N_2O than unamended urea (Watson *et al.*, 2009), unamended urea fertilizers lose more NH_3 through volatilization than CAN (Forrestal *et al.*, 2016). The 'Making Ammonia Visible' report (DAERA, 2017) stated that 91% of all NH_3 emissions in Northern Ireland (NI) come from agriculture and it recommended the use of SU fertilizers (also known as treated urea or protected urea) instead of CAN. High nitrate levels in ensiled grass, especially at later harvests, have been associated with poor fermentation characteristics (Spoelstra, 1985). Research has demonstrated that using stabilized urea (SU) fertilizer, instead of CAN, reduces N_2O emissions (Harty *et al.*, 2016) but its effects on nitrate levels in fresh grass and silage quality are less well known. This study compared the effects of SU with CAN on the nitrate concentration and BC of fresh grass, and on subsequent silage quality in 'Early', 'Mid' and 'Late' season.

Materials and methods

The study was undertaken in 2018 and 2019 at AFBI Hillsborough (54°27′N, 6°04′W) in a field which was re-seeded in 2013 with a seed mixture comprising of intermediate and late maturing perennial ryegrass (*Lolium perenne*) varieties and white clover (*Trifolium repens*). All herbage samples were from managed plots (5×1.5 m) harvested to a stubble height of 4 cm using an Agria mower (scythe width 1.1 m) as per a '3 cut' silage harvest system. The design was a randomized block comprising 4 replicates of 18 treatments in a 3×6 factorial design. The 18 treatments comprised of 3 fertilizer treatments (CAN, SU and a no fertilizer Control) with 6 harvesting intervals per cut (at 2, 3, 4, 5, 6 and 7 weeks post fertilizer application) for 3 cuts per year (1^{st}, 2^{nd} and 3^{rd} cuts) and repeated over 2 years. The CAN fertilizer was SulfaCAN (26.6:0:0:12.5S) and the SU was a KαN product (38:0:0:17.5S) as urea with the urease inhibitor, nBTPT, plus sulphur. Following January soil sampling, fertilizers were applied in March – July as per RB209 guidelines (AHDB, 2010) for soil P and K Index 2 at 120, 100 and 100 kg N ha^{-1} for the 1^{st}, 2nd and 3rd 'silage cuts', respectively. Fresh grass samples were analysed using near infrared spectroscopy (NIRS) for dry matter (DM), nitrate, buffering capacity (BC) and crude protein (CP). The silage cut at each 'Week No 7' was chopped and placed into 6 kg mini pipe silos and a 2 kg sample was analysed after 100 days by NIRS for dry matter digestibility (DMD) and by chemical analysis for volatile corrected organic dry matter (VCODM), volatiles, NH3 N/ Total N, pH, CP, lactic acid (LA), ethanol, acid detergent fibre (ADF), ash and water soluble carbohydrates (WSC). Analysis of Variance (VSNI, 2017) was applied to assess the fixed effects of week, cut, fertilizer treatment and their interactions, blocking on year and the replicated experiment within year. Predictions from the General Linear Model (GLM) were calculated for unbalanced comparisons where required using the Genstat regression model. Pearson's correlation coefficient, *r*, was calculated with probability values in Genstat. The strength of association for absolute values of *r* (both positive and negative) was described as follows: moderate (0.40-0.59), strong (0.60-0.79) or very strong (0.80-1) as per Swinscow (1997).

Results and discussion

Nitrate levels in fresh grass samples of <1000 mg kg^{-1} were observed when the ratio of Nitrate (mg kg^{-1}):Yield (kg DM ha^{-1}) was less than approximately 0.30 (Figure 1). There was no significant treatment × week interaction between ratios for CAN and SU indicating similar rates of nitrate uptake and usage. Herbage nitrate levels were higher in late season but did not differ between CAN and SU. Nitrate levels in late season fresh grass were very strongly correlated (r=0.82: P<0.001) with CP and moderately correlated (r=0.52: P=0.010) with BC but there were no strong correlations between nitrate levels and silage quality parameters, demonstrating that higher nitrate levels in late season did not affect successful silage production. Fertilizer type (SU vs CAN) had no significant effect on silage quality parameters including VCODM, NH_3 N/Total N, pH, CP, LA, ethanol, ADF, ash, WSC, or DMD (Table 1). Treatment effects only existed between the control plots and the N fertilized plots. There were no treatment effects between acetic acid and propanol on control or N fertilized plots. No significant treatment effects were observed for silage DMD between CAN and SU at the cut level or overall.

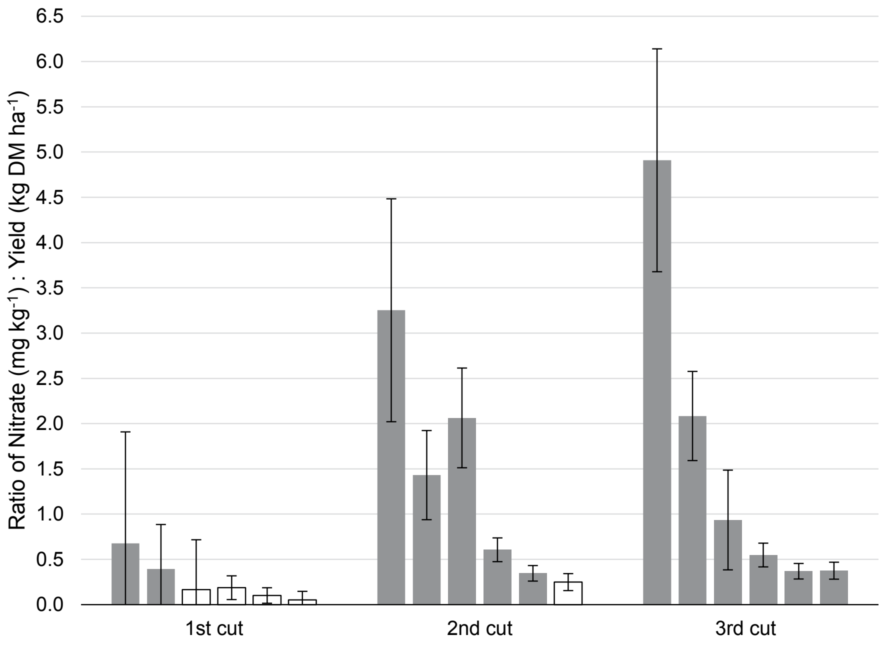

Figure 1. Mean ratio of nitrate (mg kg^{-1}) to yield (kg DM ha^{-1}) in fresh herbage in weeks 2-7 (columns), cuts 1-3 for CAN and SU fertilizer treatments combined in 2018 and 2019. Standard error bars shown, n=256, average s.e.d. = 0.29, white columns indicate ratio <0.30.

Table 1. Effect of fertilizer N type on silage quality parameters in 2018 and 2019.[1]

	Treatment			*P*-value	SED
	Control	CAN	SU		
VCODM (g kg^{-1})	269[a]	223[b]	223[b]	0.003	14.5
NH_3 /N (g kg^{-1} Total N)	44[a]	61[b]	61[b]	< 0.001	3.1
pH	3.88[a]	3.92[b]	3.95[b]	0.021	0.02
CP (g kg^{-1} DM)	103[a]	146[b]	148[b]	< 0.001	4.6
LA (g kg^{-1} DM)	73[a]	106[b]	100[b]	< 0.001	5.3
Ethanol (g kg^{-1} DM)	36.7[a]	24.1[b]	17.9[b]	0.006	5.7
ADF (g kg^{-1} DM)	271[a]	292[b]	293[b]	< 0.001	5.7
Ash (g kg^{-1} DM)	84[a]	90[b]	93[b]	< 0.001	2.1
WSC (g kg^{-1} DM)	74[a]	15[b]	14[b]	< 0.001	7.3
DMD	718[a]	745[b]	735[ab]	0.013	9.1

[1] Significant differences between treatments are suffixed with a or b superscript. Data are means of 1st and 3rd cuts in 2018 and means of 1st, 2nd and 3rd cuts in 2019. SED = standard error of a difference.

Conclusions

The effects of SU fertilizer were similar to those of CAN with regard to nitrate concentration in fresh grass at both week and cut level and with regard to silage quality from early, mid and late season harvests. In multi-cut silage systems SU can therefore be considered a suitable replacement for CAN.

Acknowledgements

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References

AHDB (2010) Section 8: Grass*, Nutrient Management Guide* (RB209). Agriculture and Horticulture Development Board, Kenilworth, UK, pp. 177-214.

DAERA (2017) *Making Ammonia Visible,* Expert Working Group on Sustainable Agricultural Land Management for N. Ireland. pp. 1-101.

Forrestal P.J., Harty M.A., Carolan R., Lanigan G., Watson C.J., Laughlin R.J., McNeill G., Chambers B. and Richards K.G. (2016) Ammonia emissions from stabilised urea fertiliser formulations in temperate grassland, *Soil Use Management* 32, 92-100.

Harty M.A., Forrestal P.J., Watson C.J., McGeough K.L., Carolan R., Elliot C., Krol D., Laughlin R.J., Richards K.G. and Lanigan G.J. (2016) Reducing nitrous oxide emissions by changing N fertiliser use from calcium ammonium nitrate (CAN) to urea based formulations. *Science of the Total Environment* 563-564, 576-586.

Roche L., Forrestal P.J., Lanigan G.J., Richards K.G., Shaw L.J. and Wall D.P. (2016) Impact of fertiliser nitrogen formulation, and N stabilisers on nitrous oxide emissions in spring barley. *Agriculture, Ecosystems & Environment* 233, 229-237.

Spoelstra S.F. (1985) Nitrate in silage. *Grass and Forage Science* 40, 1-11.

Swinscow T.D.V. (1997) 11, Correlation and regression, *Statistics at Square One*, BMJ Publishing Group 1997. Available at: https://www.bmj.com/about-bmj/resources-readers/publications/statistics-square-one/11-correlation-and-regression

VSNI (2017) *Genstat for Windows* 19th Edition, VSN International, Hemel Hempstead, UK.

Watson C.J., Laughlin R.J. and McGeough K.L. (2009) Modification of nitrogen fertilisers using inhibitors: opportunities and potentials for improving nitrogen use efficiency, *Proceedings. The International Fertiliser Society,* York, UK. pp. 1-40.

Effects of fertilization on the yield and nutritive value of bromegrass mixture with legumes

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Abstract

Alaska brome (cv. Hakari) and smooth brome (cv. Lehis) were grown in pure stands and in combination with lucerne (cv. Karlu) and red clover (cv. Varte) in a field trial during 2017-2020 in treatments with fertilization or without fertilization (organic). In fertilized stands Alaska brome and smooth brome plots were cut three times in the summer, and received a total of 200 kg ha^{-1} N (80-60-60 kg ha^{-1} after each cut). Autumn fertilizer (7-20-28) was also given at 300 kg ha^{-1} ($N_{21}P_{60}K_{84}$) in the fertilization treatments. The average dry matter yields (DMY) in organic pure seedings were 2.9 Mg ha^{-1} for smooth brome and 2.5 Mg ha^{-1} for Alaska brome. In the organic mixture with lucerne, DM yields were 6.0 and 5.8 t ha^{-1}. The forage with the highest nutritive value was obtained when growing grasses in the organic mixture with red clover: digestible dry matter (DDM) 621-677 g kg^{-1} DM, metabolizable energy (ME) 9.1-10.2 MJ kg^{-1} DM. The lucerne showed similar results. The nitrogen fertilizer used in the fertilized treatment increased the DMY and the protein content of the forage. Alaska brome had a better nutritional value than smooth brome. The smooth brome mixture was less digestible due to the higher fibre content.

Keywords: Alaska brome, smooth brome, grass mixtures, forage yield, forage nutritive value

Introduction

The increasing role of sustainable grassland-based ruminant systems in Europe highlights the use of sown multi-species swards and stresses the need for comprehensive studies on the influence of grassland management strategies in different local conditions (Peyraud *et al.*, 2014). An optimal combination of suitable grass and legume companion species is needed to obtain high N-use efficiency, high herbage yield and high contents of nutritive compounds in grass-legume mixtures (Elgersma and Søegaard, 2015). Alaska brome-grass (Alaska brome *Bromus sitchensis* Trin. in Bong.) is a relatively new species in Estonia. It has been investigated and cultivated here only very recently (Tamm *et al.,* 2018). The nutritive value is highest when the first cut is taken at early heading and budding or at the beginning of flowering. When choosing legumes for grass-clover mixtures, the rate of phenological development of the species, persistency and nutritive value should be considered. Earlier results have shown that growing red clover in mixtures with grasses improved the nutritive value and ensiling properties of the crop (Tamm, 2017). The aim of this study is to compare production abilities and forage quality of two *Bromus* species in pure stand and in mixture with lucerne and red clover cultivars in Estonian growing conditions.

Materials and methods

The experimental field was established in 2017 in Saku, Estonia (latitude 57° 25'). The study included data from two years (period 2017-2020). The trial plots were established on a typical soddy-calcareous soil where the agrochemical indicators were as follows: pH_{KCl} 7.4 (ISO 10390); soil carbon content C_{org} 3.0% (Tyurin method) and concentrations of lactate soluble phosphorus (P) and potassium (K) of 53 and 97 mg kg^{-1} (Mehlich III method).

The trial was established without fertilization. The sowing rate of Alaska brome cv. Hakari *(Bs)* and smooth brome (*Bromus inermis* Leysser) cv. Lehis *(Bi)* was 20 kg ha^{-1}, lucerne (*Medicago sativa* Lam) cv. Karlu (*Ms)* 12 kg ha^{-1} and red clover cv. Varte *(Tp)* 10 kg ha^{-1}. The trials were established with split-plot design in four replicates. Two different fertilization systems were compared in the experiment: (A) non-fertilized treatment (Alaska brome, smooth brome pure seeding and grass-red clover) and lucerne mixtures, and (B) fertilized treatment (Alaska brome, smooth brome pure seeding with 200 kg N ha^{-1} in three applications (80+60+60 kg ha^{-1}). Autumn fertilizer (7-20-28) was also given at 300 kg ha^{-1} ($N_{21}P_{60}K_{84}$) in treatment B. The crop was cut by scythe, weighed, and samples taken for analyses and determination of botanical composition. A three-cut system was used during harvest. First cuts were taken between 28 May and 2 June. Second cuts were taken between 3 July and 18 July. Third cuts were taken during 2 September to 18 September. Effective temperatures over 5 °C (in April – September) were 1,729 °C in 2018, 1,459 °C in 2019, and 1,394 °C in 2020. Rainfall (April – September) was 266 mm in 2018, 351 mm in 2019 and 459 mm in 2020. The following data were collected in this experiment: dry matter (DM) yield, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), metabolizable energy (ME) content and digestible dry matter (DDM). Statistical analyses (ANOVA and Fisher's LSD) were carried out by Agrobase 20™.

Results and discussion

Over the three years, a significant difference in the DM yields was found in plots using the different fertilization technologies. The highest DM yields of Alaska brome and smooth brome were obtained with pure seeding supplied with N fertilizer, with the 3-year average yields of 10.1 and 11.0 Mg ha^{-1}, respectively. The 3-year average DM yield in the case of non-fertilized pure seeding was 2.9 Mg ha^{-1} for smooth brome and 2.5 Mg ha^{-1} for Alaska brome.

The 3-year average DM yields were 9.2 and 7.8 Mg ha^{-1} for smooth brome-lucerne and Alaska brome-lucerne in the fertilized treatment.

The botanical composition in the mixtures of Alaska brome-lucerne and Alaska brome-red clover (non-fertilized) were grasses 52%, lucerne 44% and red clover 41%. In the mixtures with smooth brome-lucerne and smooth brome-red clover (non-fertilized) grasses averaged 54%, lucerne 44% and red clover 36%. N fertilizer increased grasses and reduced legume proportion in the mixture. The CP in the first cut of A treatment with smooth brome and Alaska brome was low (97-108 g kg^{-1} DM) and in the B treatment it was the highest (141-145 g kg^{-1}DM) (Table 2). The N used in the fertilized treatment increased the forage yield and CP concentration. Lucerne and red clover mixture increased the CP concentration compared

Table 1. The DM yield (Mg ha^{-1}) of Alaska brome *(Bs)*, smooth brome *(Bi)* and in the mixtures with lucerne *(Ms)* and red clover *(Tp)* in 2018-2020.[1]

Species	Treatment	2018	2019	2020	Average
Bs	A	3.8^{e}	1.6^{h}	2^{g}	2.5^{e}
	B	6.1^{b}	7.2^{c}	17^{a}	10.1^{ab}
Bs/Ms	A	4.0^{de}	5.8^{de}	7.7^{d}	5.8^{c}
	B	4.3^{d}	6.9^{c}	12.2^{c}	7.8^{bc}
Bs/Tp	A	4.5^{d}	5.6^{c}	4.3^{e}	4.8^{cd}
	B	4.9^{c}	6.3^{d}	7.7^{d}	6.3^{c}
Bi	A	3.5^{f}	2.5^{g}	2.8^{e}	2.9^{de}
	B	6.4^{a}	9.5^{a}	17^{a}	11^{a}
Bi/Ms	A	3.4^{f}	6.1^{de}	8^{d}	5.9^{e}
	B	3.9^{e}	7.7^{b}	16.1^{b}	9.2^{b}
Bi/Tp	A	3.4^{f}	4.3^{f}	3.3^{f}	3.7^{de}
	B	3.9^{e}	5.9^{de}	8.1^{d}	6^{c}
LSD 95%		0.29	0.49	0.54	1.5

[1] Different lower case letters within column are significantly different ($P<0.05$; ANOVA, Fisher LSD test).

Table 2. The average nutritive value of the Alaska brome *(Bs)* and smooth brome *(Bi)* red clover *(Tp)* lucerne (*Ms)* mixtures at first and second cut during 2018-2020.[1]

Species	Treatment	First cut				Second cut			
		CP g kg^{-1}	NDF g kg^{-1}	DDM g kg^{-1}	ME MJ kg^{-1}	CP g kg^{-1}	NDF g kg^{-1}	DDM g kg^{-1}	ME MJ kg^{-1}
(Bs)	A	108^{cd}	540^{bc}	655^{b}	9.6^{bc}	97^{d}	630^{ab}	610^{cd}	8.9^{c}
	B	142^{ab}	620^{a}	635^{d}	9.3^{cd}	144^{bc}	673^{a}	599^{d}	9.0^{bc}
(Bs)/(Ms)	A	172^{a}	447^{e}	679^{a}	10.2^{a}	167^{ab}	502^{cd}	636^{b}	9.4^{abc}
	B	158^{ab}	477^{de}	671^{ab}	10.0^{ab}	178^{ab}	489^{d}	643^{ab}	9.5^{ab}
(Bs)/(Tp)	A	130^{bcd}	474^{de}	677^{a}	10.2^{a}	117^{cbd}	547^{c}	621^{bc}	9.1^{abc}
	B	140^{b}	505^{cd}	666^{ab}	10.0^{ab}	142^{bc}	513^{cd}	637^{ab}	9.4^{abc}
(Bi)	A	102^{d}	566^{b}	634^{d}	9.3^{cd}	107^{d}	601^{b}	617^{bc}	9.0^{bc}
	B	145^{ab}	641^{a}	618^{d}	9.1^{d}	141^{cb}	662^{a}	609^{cd}	9.2^{abc}
(Bi)/(Ms)	A	164^{ab}	482^{de}	665^{ab}	9.9^{ab}	184^{a}	466^{d}	656^{a}	9.6^{a}
	B	164^{ab}	475^{de}	666^{ab}	10.0^{ab}	187^{a}	477^{d}	649^{ab}	9.5^{ab}
(Bi)/(Tp)	A	136^{bc}	494^{cd}	660^{b}	9.9^{ab}	148^{b}	477^{d}	652^{ab}	9.6^{a}
	B	135^{bc}	506^{cd}	659^{b}	9.8^{ab}	144^{bc}	499^{cd}	644^{ab}	9.5^{ab}

[1]Different lower case letters within column are significantly different ($P<0.05$; ANOVA, Fisher LSD test).

to pure seeding in the case of the non-fertilized treatment (supporting the results of Tamm *et al.*, 2018). In the mixtures smooth brome was less competitive than Alaska brome, thus the CP concentration of the Alaska brome-red clover mixtures was lower than that in the Alaska brome-lucerne mixture. Lower CP concentrations (97-108 g kg^{-1} DM) were found in treatments without N fertilizer in first and second cuts of grasses. An indicator of the nutritive value of the forage is the NDF, which helps to account for the feed intake potential of forage. In all cuts the NDF values of the mixtures were lower than those of the pure grass variants because the concentration of the cell walls was higher in the grasses than in the lucerne and red clover. All grass-legume mixtures had higher DDM (659-679 g kg^{-1} DM) in the first cut than the second cut. Metabolizable energy value was greater in grass-legume mixtures than in the pure grasses.

Conclusions

As a 3-year average, there was no difference in dry matter yield between the two bromegrass species. In the non-fertilized treatments the lucerne mixtures with grasses had high DM yields. Fertilization increased DM yield and improved the forage nutritional value, especially in the case of pure grasses. The legumes ensured that the forage was of high metabolizable energy value with high digestible dry matter content in both fertilization treatments.

References

Elgersma A. and Søegaard K. (2015) Productivity and herbage quality in two-species grass-legume mixtures under cutting. Grassland and forages in high output dairy farming systems. *Grassland Science in Europe* 20, 401-403.

Peyraud J.L., Van den Pol-van Dasselaar A., Collins R.P., Huguenin-Elie O., Dillon P. and Peeters A. (2014) Multi-species swards and multi-scale strategies for multifunctional grassland-base ruminant production systems: An overview of the FP7-MultiSward Project. *Grassland Science in Europe* 19, 695-715.

Tamm U., Meripõld H., Tamm S. and Edesi L. (2018) The nutritive value of Alaska brome and tall fescue forage using different growing technologies. *Grassland Science in Europe* 23, 363-365.

Phyllospheric bacteria alter sugar content and sucrose transporter expression in ryegrass

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Abstract

In perennial ryegrass (*Lolium perenne* L.), the sucrose lateral transporter LpSUT1 retrieves sucrose from the apoplast and the fine tuning of its expression should allow the plant to regulate the growth of phyllosphere bacteria which feed on sugars leaking from the plant apoplast. *Curtobacterium* and *Hafnia* strains were isolated from the microbiota of the phyllosphere of permanent grassland grazed by cattle. Bacterial suspensions were sprayed on leaves of *L. perenne* grown in a greenhouse. One and seven days after the bacterial supply, fructans and sucrose contents as well as *Lp-SUT1* transcript levels were measured in the base of elongating leaves, where meristematic cells act as a strong sink for sucrose. Irrespective of the strain, bacteria had no significant effect on fructan mobilization while one strain of *Hafnia* decreased the sucrose content and up-regulated *LpSUT1* expression. These results suggest that some phyllospheric bacteria sprayed on leaf blades compete with the host plant for sugars and manage a long-distance effect in elongating leaf bases, which is in favour of an increased sucrose lateral transport to compensate for the additional sink of carbon created by the presence of bacteria.

Keywords: Phyllosphere bacteria, sucrose transporter, *Lolium perenne*, fructans, sugars

Introduction

Phyllosphere (i.e. the parts of terrestrial plants above the ground) is considered as a harsh environment for the bacteria that can feed on sucrose leaking out in the cell wall continuum (the apoplast) of the plant. In perennial ryegrass (*Lolium perenne*), which is of major agro-economic importance in temperate grasslands and which can harbor microorganisms that contribute to the quality of raw dairy products, this leak requires a step of temporary translocation of the sucrose prior to loading at high concentration in the phloem. This is achieved through the presence of active sucrose transporters like LpSUT1 (Berthier *et al.*, 2014). In sink tissues such as elongating leaf bases, LpSUT1 enables the efficient import of sucrose from phloem to parenchyma cells, which supports active growth (Figure 1). As such, fine-tuning the expression of *Lp-SUT1* sucrose transporter should be of upmost importance to sequester sucrose in the plant and prevent its leakage to the phyllosphere, which may allow the plant to regulate bacterial growth. Despite the increased knowledge on sucrose transport in plants (Jeena *et al.*, 2018), the possible link between leaf colonization by phyllospheric bacteria and transcriptional regulation of sucrose transporters like Lp-SUT1 could be relevant but has never been investigated before. The sugar contents and the transcriptional regulation of *Lp-SUT1* were therefore assessed in elongating leaf blades after the spraying of leaf blades with two *Curtobacterium* and two *Hafnia* strains that are both non-pathogenic and that were isolated from herd-grazed grassland. These bacteria could therefore be ingested by cattle or be found on the udder and may participate to dairy-raw milk quality.

Materials and methods

Bacteria strains were isolated from the surface of leaves sampled in grassland plots grazed by dairy cows at the INRAE experimental domain of Le-Pin-au-Haras (Normandy, France). Two strain of *Curtobacterium* and two of *Hafnia* were selected with one strain capable of using sucrose and the other incapable of using

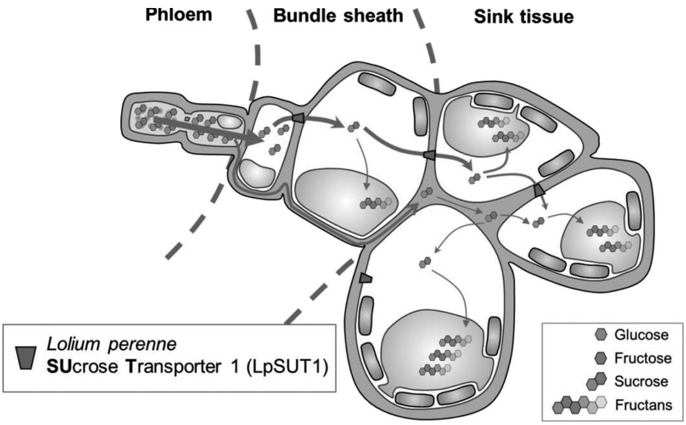

Figure 1. Schematic representation of the elongating leaf base tissue which acts as a sink. LpSUT1 is a transmembrane sucrose transporter located all along the path and responsible for the lateral transport of sucrose from phloem through peri-vascular bundle sheath and towards parenchyma sink tissues. Sucrose can therefore be accumulated as fructans within vacuoles, especially in parenchyma.

it for each genus. Each strain was abundantly sprayed on the shoot of the plants (Figure 1B). Samples were harvested immediately (*t*=0 day) and after 1 and 7 days. Mature leaf blades, their associated leaf sheaths and the elongating leaf bases at the centre of each tiller were harvested separately. For control (only sprayed with water) and each treatment, samples were harvested in four replicates corresponding to four different plants and were immediately frozen in liquid nitrogen to be stored at -80 °C until sugar content and *LpSUT1* transcript level analysis. Sugar analysis was carried out as described by Lothier *et al.* (2007), thanks to High Performance Liquid Chromatography (HPLC). RNA isolation was carried out using Qiagen® RNeasy Mini Kit.and RT-PCR analysis as described by Berthier *et al.*, 2014). Statistical analysis was performed using R Software (ver. 3.6.3) to test the normality (Shapiro-Wilk) and variance homogeneity (Bartlett) of the data and to perform a 1 factor ANOVA followed by a pairwise comparison (Tukey test). Sugar contents and *Lp-SUT1* transcript levels are displayed as the mean ± SE of the values obtained when subtracting the values of the control (Tween 2%) to the values corresponding to the treated plants (bacteria) of the same harvest time (day 1 or 7).

Results and discussion

The spraying with *Curtobacterium* strains had almost no effect on fructans content in elongating leaf bases during the time-course of the experiment while with *Hafnia 1* and *Hafnia 2* it increased and decreased fructans content, respectively (Figure 2). All strains increased sucrose content by 19% on day 1 while *Hafnia* strains decreased sucrose content on day 7.

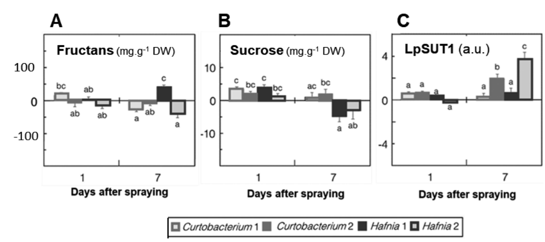

Figure 2. Fructans (A) and sucrose (B) relative contents, and *LpSUT1* relative expression (C) in elongating leaf bases 1 and 7 days after bacteria spraying. Results are presented as the difference between the control plants that where only sprayed with water at day 0 and the treated plants. When larger than the symbols, error bars represent ± standard error of the mean for n=4. Different letters above bars indicate significant differences between treatment ($P<0.05$, Tukey t-test or Dunn's test for multiple comparisons).

Lp-SUT1 transcript levels were slightly increased in elongating leaf bases by both *Curtobacterium* strains on day 1 while a 2 and 4-fold increase was respectively measured in response to *Curtobacterium* 2 and *Hafnia* 2 on day 7. This strongly suggests that these bacteria strains were able to induce a more efficient lateral transport of sucrose in elongating leaf bases. Because this tissue is an active sink for sucrose, this should lead to a better retrieval of sucrose from the phloem sap and a more efficient sequestration of sugars by the host plant, which could regulate by this way both the amount and structure of the phyllospheric microbiota.

Conclusions

Spraying bacteria altered the metabolism and distribution of plant sugars in tissues that were not directly exposed to them. Induction of expression of the sucrose lateral transporter LpSUT1 could be in favour of increased competitiveness for sucrose from the host plant in order to compensate for the sugar sink strength created by bacteria growing on leaf blades. It could also be a host plant driven mechanism to make better use of sucrose and then control the growth of phyllospheric bacteria by decreasing potential sucrose leakage.

Acknowledgements

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References

Berthier A., Meuriot F., Dédaldéchamp F., Lemoine R., Prud'homme M-P. and Noiraud-Romy N. (2014) Identification of a new sucrose transporter in rye-grass (LpSUT2): Effect of defoliation and putative fructose sensing. *Plant Physiology and Biochemistry* 84, 32.

Jeena G.S., Kumar S. and Shukla R.K. (2018) Structure, evolution and diverse physiological roles of SWEET sugar transporters in plants. *Plant Molecular Biology* 100, 35.

Lothier J., Lasseur B., Djoumad A., Le Roy K., Van Laere A., Prud'homme M-P., Barre P., Van den Ende W. and Morvan-Bertrand A. (2007) Cloning, gene mapping and functional analysis of a fructan 1-exohydrolase (1-FEH) from *Lolium perenne* rather implicated in fructan synthesis than in fructan mobilization. *Journal of Experimental Botany* 58, 1969.

Quantifying the permanent grassland erosion and flood mitigation impact in the Mediterranean climate

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Abstract

Land use change (LUC) is identified as one of the main drivers of soil erosion. However, very little information exists on the relation between land use and erosion over longer time periods and regional scales. We quantified the LUC in southern Spain over 62 years, examining its effect on soil erosion and assessing the mitigation role of permanent grassland (PG). The historical assessment was developed modelling the RUSLE's C-factor by the Monte Carlo Method (MCM). Future LUC scenarios were developed by a complete conversion of PG to cropland (PC), permanent crop (PP) and forest and natural area (FP). Despite the intensification of agriculture, no significant variation is observed in cumulative erosion at a regional scale. The underlying reasons for this resilience are multifold but can be mainly attributed to the fact that a small proportion of the total surface, 20%, dominates the total erosion, 67%. Potential LUC scenarios illustrate the importance of PG for erosion mitigation, as the CP and PP scenarios show an abrupt increase of regional erosion by 13 and 14%, while FP shows a small reduction of erosion close to 0%. This allows quantifying the erosion mitigation offered by maintaining the PG and should be considered for future agricultural policy.

Keywords: erosion, land use change, permanent grassland

Introduction

Human-induced land use change (LUC) is often identified as one of the main drivers of accelerated soil erosion. Erosion depends on different environmental factors, but land use (LU) and land management are definitely the main variables that can rapidly change over time and that are directly controlled by human action. Rapid land use change and intensification have led to strongly increased erosion rates after the second half of the 19th century. Recently, soil erosion by water was identified as the major soil threat in the European Union (EU) (Panagos *et al.*, 2015) with a soil loss rate of 9.7×10^8 t ha^{-1} y^{-1}. Within the EU, the Mediterranean countries are the most susceptible to erosion and comprise 49% of the EU's total annual soil erosion. In the Mediterranean, PG includes natural grasslands and agroforestry land uses (European Environment Agency, 2019). The CORINE land use classification defines natural grassland as constituted by a permanent grassland (PG) with low human pressure and productivity, and agroforestry as the typical oak-woodland savanna, called Dehesa or Montado, made up of 10-30% tree species cover (*Quercus suber, Quercus rotundifolia)*. This study aims to quantify the long-term effect of LULC on soil erosion on a regional scale in southern Spain, with particular attention to the role of PGs. For this, the specific objectives are: (1) to quantify the current importance of PG for soil conservation; (2) to calculate soil erosion rates for past, future and potential land use scenarios.

Materials and methods

This study focuses on the southern region of Spain, Andalusia, which extends over 8,370,150 ha. The regional climate is Mediterranean with a mean annual rainfall of 586 mm, a mean annual temperature of 14.7 °C, and a mean reference evapotranspiration of 830 mm. Forestry and agriculture are the dominant land uses while PG covers ~14% of the land. The land use classification used for this study is derived from the CORINE land cover (CLC) maps, available from 1990 to 2018. CLCs have been reclassified into four LUs: permanent crop, PG, and forest and natural area. Artificial surface and wetland and water bodies CLCs have been excluded because they do not generate soil erosion. To extend the analysis of historical land use to 1956, the land cover map of southern Spain in 1956 was used and reclassified into the same LU classes. To stress the impact of the LUC on soil erosion we assumed that the all the RUSLE's factor were constant over the studied periods except for the C-factor. Deeper analysis on the historical variation of R factor have been carried out by Vanwalleghem *et al.* (2011), justifying this assumption. When any given pixel changed its land use over time, a new value was then assigned using a Monte Carlo approach and based on this original frequency distribution of C-values. Finally, the RUSLE model was applied calculate the erosion rate the for each year in the historical series. To underline the PG erosion mitigation role at regional scale, three potential extreme LULC scenarios have been developed applying a total change of the PG area to: Permanent crop (PP); cropland (CP); forest and natural area (FP).

Results and discussion

Between 1956 and 1990, abrupt changes occurred for cropland and PG land uses: PG lost 53% of its area; permanent crop decreased by 15%; cropland, forest and natural area increased by 23% and 33% respectively (Figure 1).

This was a period of intense change in agricultural practices in southern Spain, characterized by a significant rural exodus and the introduction of mechanization. Approximately 40% of the rainfed cereal area was cultivated under fallow, whereas in 1990 this practice had disappeared almost completely. A second phase of LULC, which affected the entire region, occurred between 2006 and 2012. PG and permanent crop areas increased by 43 and 20%, while forest and cropland decreased abruptly by 11 and 16%. Finally, between 2012 and 2018, LU distribution remained practically unchanged. Between 1956 and 2018, despite the big variation of erosion rate within the different LUs, the cumulative erosion rate at regional scale remained steady. The highest peak was reached in 1990 (6.86×10^7 t y^{-1}), and the lowest in 2000 (6.49×10^7 t y^{-1}). This resilient behaviour with respect to erosion can be attributed to two main reasons. Despite significant LULC, the erosion behaviour between categories that replace each other is similar, so LULC often does not result in changes in soil erosion rates. In 2018, PG occupies 13.8% of the regional surface area and contributes less than 1% to the regional cumulative erosion. In the (PP) and (CP) scenarios, the mean erosion rate of the converted area raises respectively from 4.2 t ha^{-1} y^{-1} to 9.7 and 11.5 t ha^{-1} y^{-1}, increasing the regional cumulative erosion rate of 13% and 14% (Table 1). The total conversion of PG to forest and natural area (FP) does not imply significant changes.

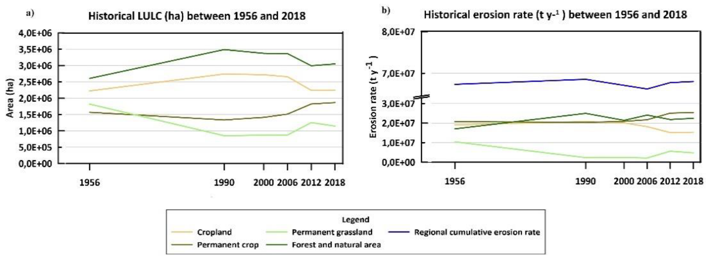

Figure 1. (A) Historical LULC and (B) erosion rate change between 1956 and 2018 in Andalusia.

Table 1. Soil erosion change of the potential LULC scenarios compared to 2018.

Scenario	Regional changes		
	Mean erosion (t ha^{-1})	Cumulative erosion (t ha^{-1} y^{-1})	Cumulative erosion rate change (%)
2018	0	0	0
PP	+1.1	+8.8x10^6	+13%
CP	+1.1	+9.3x10^6	+14%
FP	0	-8.1x10^4	0%

Conclusions

This study analysed the land use and erosion dynamics over the last 62 years at the regional scale of Andalusia, concluding that despite important changes the erosion rate is surprisingly resilient. However, our analysis shows how PG plays an important role on the regional cumulative erosion mitigation, as the total conversion of PG to permanent crop and cropland can raise the regional cumulative erosion of 13% and 14%. These results support the importance given to PG and their conservation in the potential eco-schemes that are being developed by EU member states.

Acknowledgements

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References

Panagos P., Borrelli P., Meusburger K., Alewell C., Lugato E. and Montanarella L. (2015) Estimating the soil erosion cover-management factor at the European scale. *Land Use Policy* 48, 38-50. https://doi.org/10.1016/j.landusepol.2015.05.021

Vanwalleghem T., Amate J.I., de Molina M.G., Fernández D.S. and Gómez J.A. (2011) Quantifying the effect of historical soil management on soil erosion rates in Mediterranean olive orchards. *Agriculture Ecosystems & Environment* 142, 341-351. https://doi.org/10.1016/j.agee.2011.06.003

Effects of additives on grass silage protein quality

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Abstract

To produce high-quality silages, chemical and biological additives are widely used to promote lactic acid fermentation and restrict proteolytic activities. The present experiment aimed to evaluate the effects of different silage additives on N compounds and fermentation products in grass silage under varying ensiling conditions. Therefore, a third grassland cut was wilted within one day to about 230 and 310 g kg^{-1} dry matter (DM), respectively. Plant material either had no additive (control), or was treated with *Lactobacillus plantarum* (LAC; 1 g t^{-1}), and a sodium nitrite-HMTA mixture (KL; 3 l t^{-1}). The material was ensiled in jars and stored for 90 days. The LAC treatment resulted in a significantly faster pH decline within the first 10 days in both the low and high DM silages, compared to the control and KL treatments. The non-protein-N (NPN) concentration at day-10 was lowest for the LAC treatments. However, after 90 days of fermentation, no significant differences in NPN concentration were found between silage treatments. Ammonia-N formation was reduced most effectively by the KL treatment, both after 10 days and 90 days, regardless silage DM. Hence, KL proved to be an adequate approach to improve the protein quality of silages.

Keywords: silage, additive, proteolysis, ammonia nitrogen, dry matter, pH

Introduction

The import of grain legumes from the southern hemisphere has raised questions about sustainability and security of supply due to increasing dependency, and therefore local protein production must evolve in response to societal, political and environmental pressures. Therefore, the use of on-farm-grown protein gets more attention whereby grassland crops are the most available homegrown protein source for dairy and beef production (Lüscher *et al.*, 2014). However, ensiling of high protein forages often results in an inefficient utilization of nitrogen and increasing excretion of N to the environment by ruminants because of an extensive protein breakdown during storage. The proteolytic process during ensiling can be attributed to the combined action of both, plant proteases and microbial enzymes (Fijałkowska *et al.*, 2015).

The use of inoculants, such as *Lactobacillus plantarum*, as well as chemical additives are well proven methods to maintain the feeding value of the fresh plant material and to limit protein degradation. The main objective of this publication is to examine the effects of two different types of additives on silage and protein quality under unfavourable ensiling conditions.

Materials and methods

The third cut of a perennial grassland stand at heading stage was harvested using a disc mower at the Bavarian State Research Center for Agriculture (Poing, Germany). The grass cut was characterized for crude protein, -fibre, -ash and water-soluble carbohydrates by 241, 252, 94 and 32 g kg^{-1} dry matter (DM), respectively. The lactic acid bacteria (LAB) count was 2.5×10^{7}. After mowing, the grass was collected from the field and evenly spread on plastic film to wilt the plant material to the two intended DM concentrations. The plant material was wilted at the same day under good weather conditions to 230 and 310 g kg^{-1} DM, respectively, and ensiled without an additive (control) or with *Lactobacillus plantarum* (LAC; 1g t^{-1} dissolved in 2 l of water) and sodium nitrite-HTMA

(KL; 3 l t^{-1}). The trial was done in triplicate in 1.75 l jars. After 10 days, as also 90 days, samples were taken and analysed for nitrogen compounds, crude protein fractions, fermentation pattern, DM losses and pH. All statistical analyses were implemented using SAS 9.4. Orthogonal contrast tests (PROC GLM statement CONTRAST; $P<0.05$) were performed to verify linear effects of silage additive on chemical constituents. In addition, an analysis of variance (PROC GLM) was used to test the effect of DM on chemical constituents.

Results and discussion

Based on the chemical analyses, the wilted material was classified as difficult to ensile. The fermentability coefficient (FC) was 26 for low DM and 32 for high DM. However, the wilting treatment affected fermentation pattern and silage pH values only to a slight extent (Table 1). Both the extent of protein hydrolysis and deamination of amino acids to NH_3-N were not significantly affected by shifting the silage DM ($P>0.05$). These results do not accord with the findings of Edmunds *et al.* (2014), as they determined a decreased protein degradation due to a rapid and intense wilting process. However, Edmunds *et al.* (2014) noted that wilting only has a relevant influence on proteolytic processes above 350 g kg^{-1} DM. In addition, the difference between the two DM levels is comparatively small, which impairs the expression of effects.

Silage pH was reduced faster as well as deeper by the application of LAC, compared to the control silages, but lactic acid concentration was not affected by the treatment after 90 days of ensiling. In comparison with the control silages, LAC treatment also did not significantly inhibit the formation of NPN compounds and NH_3-N. According to Winters *et al.* (2000), the enzymatic protein degradation is most effectively inhibited by a rapid lowering of the silage pH due to an extensive formation of lactic acid. The plant material in this experiment only contained small quantities of soluble carbohydrates. Besides, high concentrations of lactic acid bacteria were found on harvested plant material, resulting in a decreased impact of the inoculant on the total lactic acid formation and silage pH after 90 days.

Table 1. Effect of silage additives and dry matter (DM) level on crude protein composition and fermentation characteristics (g kg^{-1} DM) after 90 days of ensiling (unless stated).

Item	Low DM			High DM			SEM	*P*-values		
	Control	LAC	KL	Control	LAC	KL		low DM	high DM	DM
DM (g kg^{-1})	230	241	228	302	308	316	1.27	0.184	<0.001	<0.001
pH (3 d)	4.83	4.46	5.32	5.88	4.45	5.05	0.16	0.045	0.018	0.066
pH (10 d)	4.80	4.49	5.11	4.88	4.43	5.20	0.04	<0.001	<0.001	0.252
pH	4.54	4.48	4.60	4.54	4.56	4.43	0.01	0.008	<0.001	0.013
Lactic acid	59.9	60.4	62.6	63.2	66.2	69.1	2.70	0.496	0.468	0.037
Acetic acid	27.7	24.9	25.3	27.0	29.8	17.0	1.12	0.150	<0.001	0.154
Butyric acid	0.00	0.00	0.00	0.00	0.00	0.00				
NEL (MJ kg^{-1} DM)	6.26	6.36	6.43	6.43	6.40	6.42	0.03	0.001	0.626	0.010
Crude protein composition (g kg^{-1} total N)										
NPN (10 d)	469	417	457	471	435	445	4.4	0.019	0.266	0.424
NPN	540	521	527	538	548	529	5.1	0.085	0.053	0.054
True protein	46.0	47.9	47.3	46.2	45.2	47.1	0.45	0.085	0.023	0.054
NH_3-N (10 d)	375	464	103	301	484	179	2.1	<0.001	<0.001	0.763
NH_3-N	562	515	397	562	619	322	3.4	0.005	<0.001	0.728

The slowest pH decline within the first 10 d was determined in silages treated with KL, but NH_3-N accumulation was lowest in the treatment, regardless of the DM level. These results are consistent with the findings of Gomes *et al.* (2021), who observed reduced NH_3-N concentrations when silages were treated with the chemical additive KL. Furthermore, Gomes *et al.* (2021) observed that the application of KL prior to ensiling not only reduced deamination of amino acids but also reduced DM losses during fermentation. Comparable data were also gathered in this experiment, as shown in Figure 1.

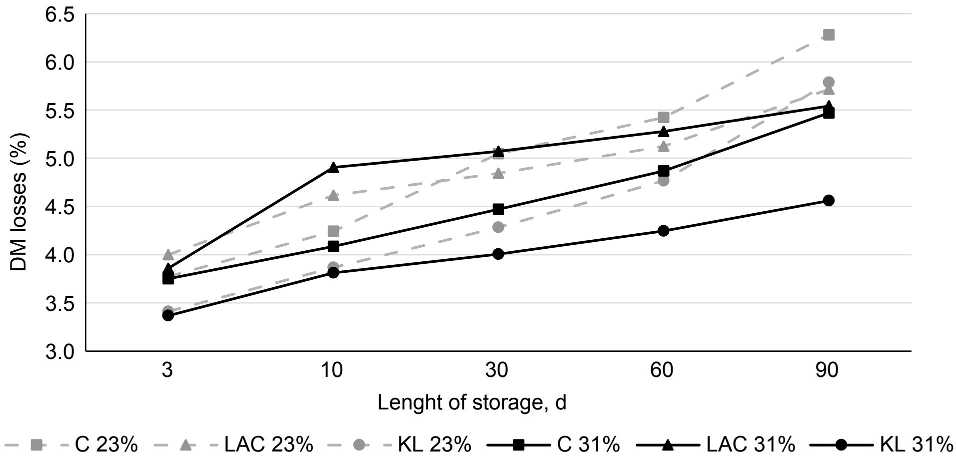

Figure 1. Effect of silage additive and DM level on fermentation losses.

Conclusions

The ensiling of grass, which was characterized by a low FC, did not result in an extensive reduction of silage quality as well as true protein. Increasing DM concentration had neither a significant effect on pH value and fermentation pattern of the silages nor on the extent of proteolytic activities. While the LAC treatment led to a faster pH decrease within the first 10 d of ensiling compared to the remaining treatments, the KL treatment was the most effective in terms of restricting deamination, even under unfavourable ensiling conditions.

References

Edmunds B., Spiekers H., Südekum K.H., Nussbaum H., Schwarz F.J. and Bennett R. (2014) Effect of extent and rate of wilting on nitrogen components of grass silage. *Grass and Forage Science* 69(1), 140-152.

Fijałkowska M., Pysera B., Lipiński K. and Strusińska D. (2015) Changes of nitrogen compounds during ensiling of high protein herbages – a review. *Annals of Animal Science*, 15(2), 289-305.

Gomes A. L. M., Auerbach H.U., Lazzari G., Moraes A., Nussio L.G., Jobim C.C. and Daniel J.L.P. (2021) Sodium nitrite-based additives improve the conservation and the nutritive value of guinea grass silage. *Animal Feed Science and Technology* 279, 115033.

Lüscher A., Mueller-Harvey I., Soussana J.F, Rees R.M., Peyraud J.L. (2014) Potential of legume-based grassland-livestock systems in Europe: A review. *Grass and Forage Science* 69, 206-228.

Winters A.L., Cockburn J.E., Dhanoa M.S. and Merry R.J. (2000). Effects of lactic acid bacteria in inoculants on changes in amino acid composition during ensilage of sterile and non-sterile ryegrass. *Journal of Applied Microbiology* 89(3), 442-452.

Feed value of pulp from fresh and ensiled grass-clover forage

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Abstract

Biorefinery of forages produces a protein-rich press juice, and a fibre-rich pulp. Use of ensiled in addition to fresh forage gives the potential to use the biorefinery all year round but the ensiling of forage can affect the quality of biorefined products. The aim was to evaluate the feed value of the pulp from fresh compared to ensiled timothy-red clover forage. Forage from first cut was pre-wilted and chopped at 28% dry matter (DM). One part of the fresh forage was kept intact (F) and another part was refined to pulp, which was ensiled in bales (FP). A third part of the fresh forage was ensiled in bales (S) and the final silage was refined to pulp (SP). The pulps had higher concentrations of DM and neutral detergent fibre (NDF) with a greater increase in SP compared to FP. DM intakes of FP and SP were lower compared to F and S, which were related to higher NDF concentrations of the pulps. SP had lower *in vivo* digestibility of organic matter (OM) compared to S, whereas no difference was found between F and FP. Thus, biorefinery of fresh or ensiled forage affects the chemical composition and, consequently, *in vivo* OM digestibility of the pulps differently.

Keywords: biorefinery, digestibility, fibre, forage, protein, ruminant

Introduction

Grasslands play a vital role in ensuring a sustainable future for global agriculture both as feed for livestock and to diminish carbon dioxide and nitrous oxide emissions to the atmosphere by being carbon sinks (Poeplau, 2020). Biorefinery of grasslands diversifies the utilization of grasslands by producing a protein-rich press juice and a fibre-rich pulp by mechanical pressing (Savonen *et al.*, 2020). Use of both ensiled and fresh forage gives all-year round use of the biorefinery but ensiling of the forage can affect the quality of biorefined products. The aim of this study was to evaluate the feed value of the pulp from fresh compared to ensiled timothy-red clover forage.

Materials and methods

Forage consisting of 66% timothy (*Phleum pratense* L.) and 44% red clover (*Trifolium pratense* L.) of dry matter (DM) was mown, pre-wilted and chopped at 28% DM at Sötåsen Agricultural High School, Töreboda, Sweden (N 58°41', E 14°8') in the first cut on 3 June 2020. The maturity stage of timothy varied form stem elongation to heading stage whereas the maturity stage of red clover varied from leaf-to-stem elongation stage. One part of the fresh forage was kept intact (F) and another part was refined to pulp, which was ensiled in hard-pressed roundbales (FP). A third part of the fresh forage was ensiled in hard-pressed roundbales (S) and the final silage was biorefined to pulp (SP) in a screw press (Cir-Tech, Skærbæk, Denmark). The four treatments were fed to eight wethers at SLU, Skara, Sweden in a duplicated 4×4 Latin square. The wethers were crosses of Suffolk, Texel or Swedish Finewool, were 7 months old, weighed 60 (standard deviation (SD) 6.4) kg and had a body condition score of 3.3 (SD 0.22) at start of the experiment. Each of the four periods was 4 weeks long, starting with an adaptation period of 14 days before 7 days of registration of *ad libitum* intake, when the wethers were housed in individual pens. During the last 7 days, the wethers were fed individually, at 80% of *ad libitum* intake, in metabolic cages. After a 3-day adaptation to the restricted feeding, total daily collection of faeces occurred during 4 days. Composited daily samples of feed, orts and faeces from each period were analysed for nutrient contents according to conventional methods. Crude protein (CP) fractions (A, B_1, B_2, B_3 and C) based on degradability characteristics according to the Cornell Net Carbohydrate and Protein System (Sniffen *et al.*, 1992) were determined according to Licitra *et al.* (1996). Feed quality data were analysed by analysis of variance for a randomized block design using the mixed procedure of SAS version 9.3, including fixed effects of ensiling, refining and ensiling × refining and random effect of block. Data on feed intake and *in vivo* apparent digestibility were analysed for a duplicated 4×4 Latin square using the same procedure and fixed effects as for the feed quality data with addition of period and random effects of animal within square and square. PSignificant differences between least-square (LS) means were done with Tukey-Kramer adjustment at $P \leq 0.05$ and tendency to significance at $0.05 \leq P \leq 0.10$.

Results and discussion

Pressing of forages in a biorefinery leaves much of the soluble nutrients in the juice fraction, resulting in increased concentrations of DM, neutral detergent fibre (NDF) and acid detergent fibre (ADF), in FP and SP compared to F and S, respectively, with a greater increase in SP compared to FP (Table 1). The water-soluble carbohydrate (WSC) concentration was lower in FP compared to F ($P=0.025$) and part of this difference was caused by fermentation of WSC during ensiling of FP in addition to the WSC extraction to the press juice. No difference in WSC content was found between S and SP, because the WSC already had been fermented mainly to lactic acid in S (data not shown). Some of the non-protein nitrogen (NPN; fraction A) in S was apparently extracted to the press juice, resulting in a smaller proportion of the NPN in the SP compared to S. However, SP contained larger proportions of fraction B_2, which mostly is rumen degradable, and fraction C, which is the ADF-bound protein and is considered to be indigestible, compared to S. No such differences were found between F and FP (Table 1).

Intakes of DM and OM by wethers fed FP and SP were lower compared to wethers fed F and S, which are related to the higher NDF concentrations of the pulps compared to their original forages (Tables 1 and 2). The NDF intake was lower for the pulps than for the intact forages, which shows that rumen fill limited intake. According to Allen (2000), forage NDF concentration is the main factor limiting intake in forage-based diets because of its slow passage rate. Furthermore, *in viv*o digestibility of NDF and ADF was not affected by the mechanical pressing of F and S. *In vivo* apparent digestibility of CP were lower for FP and SP compared to F and S and this difference was driven by SP. Thus, the decreased *in vivo* and *in vitro* OM digestibility of SP compared to S is to a large extent related to the decreased CP digestibility of SP (Tables 1 and 2).

Conclusions

Biorefinery of fresh and ensiled forage affects the chemical composition and, consequently, *in vivo* apparent OM digestibility of the pulps differently.

Table 1. Chemical composition of forage (F), forage pulp (FP), silage (S) and silage pulp (SP), n=4.[1,2]

	Treatment				SEM	*P*-value		
	F	FP	S	SP		Ensiling (E)	Refining (R)	E × R
DM, g/kg	271^{d}	349^{b}	285^{c}	464^{a}	2.6	< 0.001	< 0.001	< 0.001
Ash, g/kg DM	68^{b}	64^{c}	71^{a}	48^{d}	0.6	< 0.001	< 0.001	< 0.001
aNDFom, g/kg DM	471^{c}	542^{b}	461^{c}	629^{a}	7.1	< 0.001	< 0.001	< 0.001
ADFom, g/kg DM	263^{d}	320^{b}	297^{c}	383^{a}	6.8	< 0.001	< 0.001	0.018
ADL, g/kg DM	26	39	30	36	2.5	0.913	0.002	0.168
iNDF, g/kg NDF	183^{a}	154^{b}	143^{bc}	136^{c}	4.4	< 0.001	< 0.001	0.011
IVOMD, g/kg OM	792^{b}	803^{b}	851^{a}	770^{b}	18.9	0.226	0.008	0.002
WSC, g/kg DM	$192^{(a)}$	$89^{(b)}$	$88^{(b)}$	$68^{(b)}$	22.5	0.013	0.015	0.070
CP, g/kg DM	142	135	130	100	10.8	0.046	0.099	0.284
Fraction A, % of CP	42.4^{b}	44.0^{b}	61.2^{a}	40.3^{b}	3.69	0.053	0.019	0.009
Fraction B_1, % of CP	3.1	1.6	2.7	1.7	0.72	0.785	0.074	0.624
Fraction B_2, % of CP	35.3^{ab}	27.4^{bc}	26.3^{c}	37.9^{a}	2.04	0.703	0.374	< 0.001
Fraction B_3, % of CP	15.9	22.4	6.2	12.8	1.07	< 0.001	< 0.001	0.968
Fraction C, % of CP	3.3^{b}	4.6^{b}	3.6^{b}	7.3^{a}	0.44	< 0.001	< 0.001	0.003

[1] LS means in a row with different superscript letters differ significantly at $P \leq 0.05$. LS means in a row with different superscripts letters between brackets (a,b) tends to differ at $0.05 \leq P \leq 0.10$.

[2] aNDFom = ash-free neutral detergent fibre with addition of amylase to the detergent solution, ADFom = ash-free acid detergent fibre, ADL = acid detergent lignin, iNDF = *in vitro* indigestible NDF at 240 h incubation, IVOMD = *in vitro* organic matter (OM) digestibility, WSC = water soluble carbohydrates, CP = crude protein; A = NPN, B_1 = buffer-soluble protein, B_2 = neutral detergent-soluble protein, B_3 = acid detergent-soluble protein, C = acid-detergent insoluble protein.

Table 2. Intake and *in vivo* apparent digestibility of forage (F), forage pulp (FP), silage (S) and silage pulp (SP) fed to wethers, n=8.[1,2]

	Treatment				SEM	*P*-value		
	F	FP	S	SP		Ensiling (E)	Refining (R)	E × R
Body weight (BW), kg	64.0	62.1	62.7	63.1	4.90	0.872	0.408	0.243
Intake								
DM, kg/day	1.71	1.33	1.50	1.06	0.184	0.011	<0.001	0.788
DM, % of BW	2.63	2.13	2.38	1.67	0.132	0.003	<0.001	0.316
OM, kg/day	1.60	1.24	1.39	1.01	0.172	0.013	<0.001	0.883
NDF, kg/day	0.81	0.74	0.76	0.66	0.092	0.072	0.026	0.611
NDF, % of BW	1.23	1.19	1.21	1.04	0.069	0.106	0.067	0.229
CP, g/day	201	171	187	114	22.7	0.011	0.007	0.103
Digestibility, %								
DM	72.8^{a}	69.9^{a}	72.8^{a}	65.2^{b}	1.11	0.045	<0.001	0.044
OM	74.3^{a}	71.6^{a}	74.5^{a}	67.1^{b}	1.01	0.046	<0.001	0.030
NDF	68.3	69.2	66.3	66.0	1.39	0.070	0.835	0.663
ADF	62.9	64.9	65.9	64.8	1.78	0.414	0.791	0.393
CP	64.3	59.6	59.7	49.1	2.07	0.001	0.001	0.170

[1] LS means in a row with different superscript letters differ significantly at $P \leq 0.05$.

[2] See footnote 2 at Table 1 for definitions of abbreviations.

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References

Allen M.S. (2000) Effects of diet on short-term regulation of feed intake by lactating dairy cattle. *Journal of Dairy Science* 83(7), 1598-1624.

Licitra G., Hernandez T.M. and Van Soest P.J. (1996) Standardization of procedures for nitrogen fractionation of ruminant feeds. *Animal Feed Science and Technology* 57(4), 347-358.

Poeplau C. (2020) Grassland soil organic carbon stocks as affected by management intensity and climate change. *Grassland Science in Europe* 25, 375-383.

Savonen O., Franco M., Stefanski T., Mantysaari P., Kuoppala K. and Rinne M. (2020) Grass silage pulp as a dietary component for high-yielding dairy cows. *Animal* 14(7), 1472-1480.

Sniffen C.J., O'Connor J.D., Van Soest P.J., Fox D.G. and Russell J.B. (1992) A net carbohydrate and protein system for evaluating cattle diets: II. Carbohydrate and protein availability. *Journal of Animal Science* 70, 3562-3577.

Mechanical loosening of grasslands – a risk to ecosystem services or a restorative practice?

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Abstract

Well-managed grasslands can deliver a range of goods and services, including storing carbon, supporting habitat, regulating water flows and providing food and clean water. However, when grassland soils are compacted many of these services can be compromised. Mechanical loosening of grassland soils through spike aeration or 'sward lifting' is often promoted as a means of improving soil structure and restoring multiple ecosystem services. This paper presents results from a recent study on the effects of mechanical loosening on grass yields and water infiltration and discusses the results in the context of similar research carried out over the past decade. Several studies have demonstrated that mechanical loosening of 'moderately compacted' soil can substantially increase water infiltration rate (4- to 10-fold) and can result in improved grass yield. However, the implications for surface runoff and flooding risk are unclear, and overall grass yield effects appear to vary by site, year and season. Furthermore, one study has indicated short-term impacts on soil earthworm populations and no long-term effect on nitrous oxide emissions. This paper discusses the pros and cons of mechanical loosening within grassland systems and proposes guidance to help farmers and advisers in their decision making.

Keywords: ecosystem services, soil structure, soil compaction, mechanical loosening

Introduction

In northern and western Europe, climate change is projected to result in warmer, wetter winters and a higher frequency of extreme weather events, such as intense rainfall. This has implications for the frequency and intensity of flooding events in future. The management of permanent grassland (PG), which covers ca. 34% of the European agricultural area, can have an important influence on flooding risk. Management approaches that improve soil porosity and storage may help reduce flooding risk in some catchments. Options such as the introduction of herbs and legumes as 'multi-species swards' or soil mechanical loosening through spike aeration or 'sward lifting' are often promoted as a means of improving soil structure and function including increasing grass yield. This paper presents results from a field experiment to investigate the effect of mechanical loosening on water infiltration rates and forage dry matter yield on a fine-textured grassland soil. The results are discussed and compared with results from previous experiments investigating similar hydrological and production issues.

Materials and methods

The study was carried out on permanent grassland at Cockle Park Farm, Northumberland in north-east England (55.22N, 1.68W). The soil type is a heavy clay loam (27-30% clay), seasonally waterlogged, slowly permeable soil. The field had been in grass for seven years and mainly used for dairy cattle grazing. The field experiment was set up, with three replicates of three randomized treatments, on nine plots of 0.25 ha (ca. 100×25 m, total 2.25 ha): (1) base grass-clover mix (G-C); (2) grass clover plus deep rooting herb and legume mix (HL); and (3) base grass clover mix with mechanical loosening (G-C + L). The base grass clover mix contained 90% perennial ryegrass and 10% white clover, while the grass-clover plus deep rooting herb and legume mix contained 20% sainfoin, 15% bird's-foot trefoil (*Lotus corniculatus*), 10% chicory (*Cichorium intybus*), 5% yarrow (*Achillea millefolium*), 15% plantain (*Plantago lanceolata*), 5%

common bent (*Agrostis capillaris*), 10% sweet vernal grass (*Anthoxanthum odoratum*), 10% crested dog's tail (*Cynosurus cristatus*), 5% white clover (*Trifolium repens*) and 5% wild red clover (*Trifolium pratense* var. *pratense*). All mixes were established in April 2019. Plots 3, 5 and 7 (treatment 3) were mechanically loosened on 11/09/20 using a sward lifter with leading discs, tines and a packer roller. The tine spacing was 76 cm, with tines set to 25 cm depth to disrupt a compact layer at 10-20 cm depth; i.e. score 4 – compact – according to Visual Evaluation of Soil Structure (VESS). Manufactured fertilizer was applied in spring 2021 for a target annual dry matter yield of 5-6 tonnes according to fertilizer recommendations for England and Wales (AHDB, 2020). The manufactured nitrogen fertilizer rate was 80 kg N ha^{-1}. In April 2021, saturated water infiltration rates were measured at three randomly selected locations on each plot using a double ring infiltrometer. To measure grass yield, two 1.5×20 m cuts were taken on each plot with a Haldrup grass harvester on 26/05/21, 23/07/21 and 08/10/21. Conventional analysis of variance (ANOVA) was used to test for statistical differences between the contrasting replicated treatments.

Results and discussion

There were no statistical differences in saturated water infiltration rates between the G-C mix and HL mix. However, mechanical loosening resulted in a ten-fold increase in saturated water infiltration rates, seven months post-loosening (Figure 1). Previous studies have also shown 4- to 10-fold increases in saturated water infiltration rates following mechanical loosening of 'moderately compacted', medium-textured grassland soils (e.g. Newell Price *et al.*, 2014) with evidence of more roots in the lower topsoil. These effects can persist for two to five years post-loosening and have significant implications for the ability to graze the land in the autumn and spring. However, it is not certain whether the increased water infiltration and improved soil structure also results in increased soil water storage and reduced flooding risk in grass-dominated catchments. At Cockle Park in 2021, despite some signs of earlier growth on the mechanically loosened plots, there were no differences in grass yield between treatments for any of the three cuts or for total annual dry matter yield (P=0.86; mean = 5.62 t dry matter ha^{-1}). This reflects previous studies that have shown inconsistent yield effects from mechanical loosening of grassland. Results from European studies have been variable, with both yield increases and decreases measured, as well as variability in effects through the season (e.g. Boer *et al.*, 2018). However, results do suggest that mechanical soil loosening can be effective in improving soil structure and increasing grass yields where soil compaction has been positively identified and mechanical alleviation is effectively carried out. Where no compaction is identified at the outset of field trials/experiments, it appears that soil loosening reduces soil penetration resistance (i.e. the force needed to penetrate the soil) but can result in a reduction in grass yield due to sward and root damage (e.g. Frost, 1988). Research on the effects of mechanical loosening on greenhouse gas emissions and soil biodiversity has been limited. However, studies at two sites (Newell Price *et al.*, 2015) found no long-term effect on nitrous oxide emissions, but a negative effect of mechanical loosening on the abundance and biomass of anecic (deep-burrowing) earthworms, lasting up to two years post-loosening when studied at one site only (Lees *et al.*, 2016). This latter finding although at a single site has serious implications for important macrofauna that are increasingly thought to form a crucial part in sustainable grassland systems.

Conclusions

Mechanical loosening can result in significant increases in water infiltration rates on mineral grassland soils that can last for three years or more. This can improve soil drainage and enable early and late season grazing that can benefit the farm economy, reducing the need for conserved or imported forage and manure management. However, effects on grass yield are inconsistent and it is highly likely that soil macrofauna are negatively impacted. The focus of grassland soil management should therefore be on compaction avoidance and mechanical loosening should only be carried out when clear signs of soil compaction have been identified.

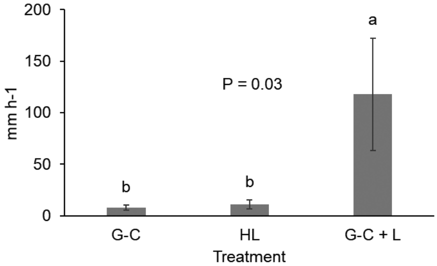

Figure 1. Mean (± standard error) saturated water infiltration rates (mm h^{-1}) on the grass-clover (G-C), deep rooting herb and legume (H&L) and grass-clover with mechanical loosening (G-C + L) plots. Different letters indicate statistically significant differences.

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References

AHDB (2020) *Nutrient Management Guide (RB209)*, Section 3, Grass and forage crops. 40 pp.

De Boer H.C., Deru J.G.C. and Van Eekeren N. (2018) Sward lifting in compacted grassland: effects on soil structure, grass rooting and productivity. *Soil and Tillage Research* 184, 317-325.

Frost J.P. (1988) Effects on crop yields of machine traffic and soil loosening. Part 1. Effects on grass yield of traffic frequency and date of loosening. *Journal of Agricultural Engineering Research* 39, 301-312.

Lees K.J., McKenzie A.J., Newell Price J.P., Critchley C.N., Rhymer C.M., Chambers B.J. and Whittingham M.J. (2016) The effects of soil compaction mitigation on below-ground fauna: How earthworms respond to mechanical loosening and power harrow cultivation. *Agriculture, Ecosystems and Environment* 232, 273-282.

Newell Price J.P., Balshaw H and Chambers B.J. (2014) Managing grasslands to mitigate flooding risk. In: Hopkins A. *et al.* (eds.) *EGF at 50: The Future of European Grassland. Grassland Science in Europe* 19, 270-272.

Newell Price J.P., Balshaw H., Critchley N., McKenzie A., Thorman R., Whittingham M.J. and Chambers B.J. (2015) Characterisation of soil structural degradation under grassland and development of measures to ameliorate its impact on biodiversity and other soil functions. Prepared as part of Defra project BD5001, 132 pp.

Scaling-up innovative grass-based products and services

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Abstract

The bio-based products sector is a priority area with high potential for future growth, re-industrialization, and addressing societal challenges in agriculture and beyond. We employ a multiple case study approach with eighteen innovative grass-based businesses to investigate conditions in the business environment that facilitate or hamper the scaling-up of innovative grass-based products and services. Our analysis reveals that the comprehensive use of grass and green fodder to produce bio-based products, such pulp, paper and plastics is becoming increasingly important across Europe. Yet, fundamental changes in rules and regulations, funding mechanisms and consumer awareness are required to balance the competition with the established fossil-based economy and make the business environments more supportive. This study aims at supporting the development of novel grass-based business models in rural areas, strengthening the bioeconomy and contributing to European policy objectives.

Keywords: grassland, bioeconomy, grass-based products, innovation, regulations, consumers

Introduction

Bioeconomy transitions in Europe open new perspectives for the transfer and co-construction of innovations based on grasslands. Grasslands have been traditionally associated with animal husbandry and the provision of the feed base for grazing livestock. Today, and increasingly so in the future, the potential of grass feedstock for green biorefining and its processing into marketable products and energy can provide new business opportunities in rural areas and address a series of societal and consumer needs (Orozco *et al.*, 2021). Yet, despite the urgent need to harness local resources and develop bio-based businesses and value chains, the full potential of grass remains widely untapped and grass-based products remain opaque in Europe's economy. This challenge gives rise to the question of how to accelerate the development of novel and alternative grass-based businesses and their potential replication. To fill this gap, we investigated 18 successful grass-based business models and found several misalignments, mainly related to competition of bio-based products with the established economic system relying on fossil resources. Our empirical findings can help to identify bottlenecks and business opportunities to improve overall growth of the grass-based industry.

Materials and methods

This study is based on a multiple case study approach (Yin, 2009). Case studies were considered appropriate due to the relatively new research field of alternative grass-based businesses. After an extensive search for grass-based business cases on 'Web of Science', we selected a set of 18 successful grass-based businesses located in different countries of Europe. We based our sampling criterion on the innovativeness of the business models in terms of the use of grass resources, the key activities for making use of grass resources, the value propositions, products and services provided different from livestock (i.e. not milk, meat, cheese, or other traditional grass-based products), as well as the customer segments, business channels, and revenue streams. Thus, the selected cases operate with grass and green fodder as their main feedstock but exhibit diversity in terms of contextual conditions, conversion processes, end-products and users.

Results and discussion

Within our empirical cases (n=18), grass-based paper was the most recurrent product (22%), followed by bioenergy (21%) and plastic (17%). Other products include fibre boards, fertilizers, feed protein, drinking straws and seeds. From our sample, most businesses engage in a multiple-product approach in which grass is processed into a product, and this product is used at least once more in material form before disposal. In line with previous research (McEniy and O'Kiely, 2014), we found that producing several products facilitates the efficient utilization of the whole plant and can create sufficient revenues to cover feedstock and subsequent processing costs. Such businesses might play a critical role in sustainability transitions by developing new products, services and business models and contributing towards the formation of new industries (Köhler *et al.*, 2019). Supportive institutional frameworks are necessary to enable these innovations to materialize (Lange *et al.*, 2021). While our empirical observations reveal that some biogeographical regions have designed policies to support the development of a circular and bio-based economy, most companies argued that there is not enough support for innovative grass-based products, and that they still experience barriers related to stricter policy regulations and administrative procedures. This seems to contradict bioeconomy policies at EU and global level that aim to promote alternative grass-based products.

Innovation in agriculture is a key process for sustainable development. However, scaling up innovative grass-based businesses requires, inter alia, investment and access to financial resources. A lack of direct funds to promote alternative grass-based products at the European level was reported. Instead, funds provided for livestock products through the animal headage payment within the coupled payments may cause competition for the use of the grass on livestock farms. Thus, funding mechanisms that incorporate and promote the specific benefits generated by grass producing and processing companies are needed. Practitioners call for clear regulations that specifically support the developing grass-based industry.

Consumers also have a decisive role in bioeconomy transitions. Yet, there is a lack of confidence or trust regarding the quality of bio-based products, compared to their fossil-based counterparts, which hampers the market uptake of grass-based products. From this perspective, product certification and providing clear information on the quality, usability, production methods and the materials used in the production process could have a positive impact on consumers' willingness to choose grass-based products. Consumers and their agency can also stimulate companies to innovate and to supply more resource-efficient goods and services. Appropriate price signals and adequate labelling with clear information on the verified quality and sustainability of grass-based products emerge as necessary conditions for the development of a grassland bioeconomy.

Conclusions

The potential benefits of multifunctionality in grassland agriculture to provide a diverse number of products and ecosystem services has been increasingly recognized (Weigelt *et al.*, 2009). Thus, it can be expected that services, goods and functions of grassland will become more important. Yet, a key challenge for grassland farming is to design production systems and management measures in such a way that the multiple functions and services are adequately fulfilled or provided (Wehn *et al.*, 2018). Given these realities, the growing significance of grasslands underlines the necessity for both empirical and conceptual research on the co-evolution of bioeconomy transitions and grassland-based innovations.

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References

Kleijn D. and Sutherland W.J. (2003) How effective are European agri-environment schemes in conserving and promoting biodiversity? *Journal of Applied Ecology* 40(6), 947-969. https://doi.org/10.1111/j.1365-2664.2003.00868.x

Köhler J., Geels F.W., Kern F., Markard J., Onsongo E., Wieczorek A., Alkemade F., Avelino F., Bergek A., ... and Wells P. (2019) An agenda for sustainability transitions research: State of the art and future directions. *Environmental Innovation and Societal Transitions* 31, 1-32. https://doi.org/10.1016/j.eist.2019.01.004

Lange L., Connor K.O., Arason S., Bundgård-Jørgensen U. ... and Vieira H. (2021) Developing a sustainable and circular bio-based economy in EU: by partnering across sectors, upscaling and using new knowledge faster, and for the benefit of climate, environment and biodiversity, and people and business. *Frontiers in Bioengineering and Biotechnology* 8, 619066. https://doi.org/10.3389/fbioe.2020.619066

McEniy J. and O'Kiely P. (2014) Developments in grass-/forage-based biorefineries. In: *Advances in Biorefineries* Elsevier, Amsterdam, the Netherlands, pp. 335-363. https://doi.org/10.1533/9780857097385.1.335

Orozco R., Mosquera-Losada M.R., Rodriguez J., Adamseged M.E. and Grundmann P. (2021) *Supportive business environments to develop grass bioeconomy in Europe.* https://doi.org/10.3390/su132212629

Wehn S., Burton R., Riley M., Johansen L., Hovstad K.A. and Rønningen K. (2018) Adaptive biodiversity management of semi-natural hay meadows: The case of West-Norway. *Land Use Policy* 72, 259-269. https://doi.org/10.1016/j.landusepol.2017.12.063

Weigelt A., Weisser W.W., Buchmann N., and Scherer-Lorenzen M. (2009). Biodiversity for multifunctional grasslands: Equal productivity in high-diversity low-input and low-diversity high-input systems. *Biogeosciences* 6(8), 1695-1706. https://doi.org/10.5194/bg-6-1695-2009

Yin R.K. (2009). *Case study research: Design and methods* (4th ed). Sage Publications, Thousand oaks, CA, USA.

Reed canary grass and tall fescue from marginal land as substrates for the bio-economy

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Abstract

Mixtures of grass and clover can be productive and sustainable for industrial and energy purposes. However, not all parts of the biomass are suitable as industrial substrate and there is a need for methods to separate fibrous parts from other biomass. Biomass production was evaluated in two-cut systems at two sites on marginal land. One site is Alnarp in Lomma, southern Sweden on a former pasture on coarse soil. The other is Röbäcksdalen in Umeå, northern Sweden on agricultural silty soil with poor drainage. Reed canary grass *Phalaris arundinacea* L. (2 varieties) and tall fescue, *Festuca arundinacea* Schreb. were sown, either in monocultures, or with clovers, in a randomized design. In addition, we sowed four mixtures in larger plots on another field to make round bale silage for drying and fractionation experiments using a cyclone process. Reed canary grass was very competitive and outcompeted tall fescue and clover where it thrived. However, at Alnarp tall fescue dominated in parts of the plots where the soil was too dry for reed canary grass. We present production data from two to three growing seasons along with evaluation of the cyclone process.

Keywords: reed canary grass, tall fescue, biomass fractionation

Introduction

Biomass for the bioeconomy needs to be produced in a sustainable way and on soil not normally is used for food production. Mixtures of grass and clover are productive and sustainable and production systems for different end-uses need to be developed for marginal land cropping. The first objective of this paper is to compare two grass species grown in monoculture or in a more diverse species mix in two harvest systems with either early or late first harvest. Different parts of the biomass, e.g. stems and leaves have different properties. There is a need for methods to separate the fibre-rich stem from the leaf fraction. The second objective is to evaluate a cyclone process for drying and separating grass materials. The fractions produced can be used as raw material for fibre production, fibreboard production, insulation material, adsorbents, feed, or raw material for biogas.

Material and methods

Two field experiments were conducted on marginal land in Alnarp, southern Sweden lat. 55.66 N; 13.10 E (gravelly soil) and Röbäcksdalen northern Sweden lat. 63.81 N; 20.24 E (silty soil with poor drainage). Triplicate plots were established for each treatment in a split plot design with harvest system (two cuts with early or late first cut) on the main plots and species mixes on the sub plots. At each site, two varieties of reed canary grass (RCG), one a forage variety (var. Lara) and one an energy variety (SWRF5004 at Alnarp and SW Bamse in Röbäcksdalen) and tall fescue var. Swaj (TF) were grown in monoculture. All three grass varieties were also grown in mixture (MIX) with red clover *Trifolium pratense* L. and Alsike clover *Trifolium hybridum* L. At both sites, the clovers did not survive the first year. At Alnarp this was caused by drought and weed problems, and at Röbäcksdalen, by ice cover in the first winter due to repeated thawing and freezing. Re-establishment of clovers was tried at both sites, but this was successful only at Röbäcksdalen. A Haldrup forage harvester was used to cut the biomass on a 1.5 m wide strip in the middle of the plots to assess yield.

At Röbäcksdalen in 2018, we also established four larger non-replicated plots at another field with poor drainage. These were monoculture of RCG Lara, monoculture of TF Swaj and intercrops of RCG var. Lara and Alsike clover (AC) var. Frida and TF and Red clover (RC) var. Betty. Biomass from this field was round-baled and wrapped in plastic for silage.

Round-baled RCG from the first harvests of 2019 that had been stored outdoors for nine months was processed in a cyclone (Tikka *et al.*, 2007). The process produces two fractions, an accept fraction (mainly stems) and a reject fraction (mainly leaves). All materials were cut to approximately 30 mm before fed into the cyclone. About 30 kg material was processed for each setting. Optimization of the cyclone process was done with a full factorial experimental design where feeding rate was varied between 40 and 100 kg h^{-1} and air temperature between 40 °C and 80 °C. The dry matter content of the material fed to the process was about 60%. NCSS 2020 was used to make repeated measures ANOVA models of harvest data for each site separately. Tukey Kramer tests were used for comparisons between treatments.

Results and discussion

Average biomass yields were higher at Röbäcksdalen, 4,443 kg DM ha^{-1}, than at Alnarp, 2,756 kg DM ha^{-1} for each harvest. At Röbäcksdalen, there were significant differences between the species mixtures (Table 1), between harvest occasions and several significant interactions. MIX had higher yields than RCG SW Bamse according to Tukey Kramer's comparison between all treatments. This could be a sign of overyielding, but since RCG comprised 0.90-0.97 of the total biomass in the mixed plots and clover biomass was only 0.01-0.03 of the total biomass this is not very likely. TF had higher yield than RCG in the 2020 first harvest because of more ice damage in RCG than in TF. However, RCG recovered fast and had higher yields than tall fescue both in the 2020 second-harvest and the 2021 first-harvest, making differences in total yield between the monocultures small. Total annual yield did not differ between early and late first cut although there were differences in each cut.

The experiment at Alnarp was very uneven, probably due to uneven water availability in the soil. Thus, there were no significant differences between the species and harvest systems (Table 2). However, two interactions were significant: harvest system × species mixes and harvest system × harvest occasion. RCG SWRF5004 had higher yield than TF and MIX in the early first-cut system, while all species mixtures with RCG had higher yield than TF in the late-harvest system. The yields were very low the first year, but in the second and third year the first harvests were better. However, the regrowth was always poor due to drought and the second harvests yielded too little biomass to be profitable. In MIX, RCG dominated over TF in the wetter parts of the field and TF dominated where RCG did not thrive.

Ice cover also damaged the large field at Röbäcksdalen in winter 2020. Tall fescue had so little biomass in the first harvest of 2020 that the two plots yielded only one bale with TF together (Table 2). However, it recovered partly in 2021 although yields were lower than for RCG.

The cyclone optimization trials showed that the 'accept fraction' increases with increased feeding rate of the raw material. Higher temperature also enables a higher feeding rate and gives a product with higher dry matter content. Preliminary data show an increase in cellulose content of the accept fraction compared to the start material.

Conclusions

Reed canary grass and tall fescue can be productive crops for the bioeconomy when grown in monoculture and fertilized with digestates, and cyclone processing of the ensiled grass can reduce the moisture content and increase cellulose content.

Table 1. Probabilities, p, for no difference in biomass yield between treatments. Repeated Measures ANOVA for each site.[1]

Factor term	Röbäcksdalen (p)	Alnarp (p)
Harvest system	0.763	0.471
Species mixes	*0.008*	0.300
Harvest system × Species mixes	0.966	*0.042*
Harvest occasion	*0.0006*	*0.000000*
Harvest system × Harvest occasion	*0.005*	*0.001*
Species mixes × Harvest occasion	*0.000001*	0.155
Three way interaction	0.541	0.597

[1] $P<0.05$ is in italic.

Table 2. Biomass yields, kg DM ha^{-1}, in larger production plots 2020-2021.

Species mix	2020 spring harvest	2020 first harvest	2021 first harvest
RCG	5,622	3,410	9,045
TF	3,657	Less than 1 bale	4,822
RCG and AC	4,478	2,010	6,971
TF and RC	4,561	Less than 1 bale	6,616

Table 3. Process settings and results from the cyclone optimization trials.

Feeding rate (kg h^{-1})	Temperature (°C)	Accept fraction (%)	Accept DM (%)
44.4	40	47.3	88.8
61.2	40	61.5	87.4
73.8	40	73.1	86.9
42.6	60	48.9	89.2
61.2	60	59.7	88.6
81.6	60	71.4	86.8
43.8	80	46.7	93.2
78	80	74	91.2
99	80	80	90.3

Acknowledgements

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Reference

Tikka J., Lindfors N.-O. and Bäcklund E. (2007) Learning from nature: the tornado process. *The 6th International Heavy Minerals Conference 'Back to Basics', The Southern African Institute of Mining and Metallurgy.*

Robust cattle valorise ecosystem services of marginal grassland

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Abstract

Semi-natural, marginal pastures offer a plethora of ecosystem services but they are often underused in modern agriculture. We analysed if robust cattle valorise these services more efficiently than highly productive cattle. We assessed anatomy, feeding and movement behaviour of Highland cattle (HC) as a model for robust cattle and compared it to the medium-productive Original Brown and high-productive Angus×Holstein in a controlled experiment in the Swiss Alps. Additionally, we investigated the vegetation of 25 pastures of HC with adjacent pastures of highly productive cattle. HC differed significantly from productive breeds: (1) HC were significantly lighter, but had large claws and covered less distance. Consequently, trampling pressure was lower and trampling-adapted plant species were rarer on HC pastures. Since these plants outcompete more-susceptible species, biodiversity was higher on HC pastures. (2) HC grazed least selectively and foraged unattractive plants, whereas high-productive cattle preferred nutrient-rich, easily digestible forage. Thereby, HC reduce problematic plants. (3) HC used the pasture most evenly and exploited different resources. (4) The productive breeds lost weight on the marginal pastures, whereas HC gained weight, indicating a more efficient roughage conversion. Robust cattle make efficient use of marginal grassland, thereby valorising these pastures and promoting biodiversity.

Keywords: biodiversity, cattle breeds, forage selection, movement behaviour, productivity

Introduction

Marginal pastures offer a plethora of ecosystem services, such as biomass production, outstanding biodiversity and landscape aesthetic for recreation and tourism. These services are valorised and maintained by livestock (Martin-Collado *et al.*, 2019). However, due to their comparably small biomass production and low forage quality, these pastures are difficult to integrate into modern intensive agriculture. Consequently, they are underused and abandoned. The abandonment of marginal land may be enforced by output-orientated livestock breeding which enormously changed livestock characteristics during the last century. In cattle, high-productive, specialised dairy or beef cattle emerged. They differ from low-productive, traditional breeds in appearance and productivity, but probably also in anatomy, movement and foraging behaviour. Such differences could have far-reaching consequences for pasture vegetation and the way in which cattle valorise ecosystem services of marginal grasslands.

Materials and methods

Sub-study I (Pauler *et al.*, 2020a; Pauler *et al.*, 2020b) investigated three cattle breeds representing different levels of productivity: (1) low-productive Highland cattle, (2) dual-purpose Original Brown and (3) high-productive Angus×Holstein crossbreed. The cattle simultaneously grazed three types of heterogeneous subalpine pastures in the Swiss Alps (2026 m asl.). Individual body weight and claw base area were measured. To analyse the movement behaviour, we recorded speed, space use evenness and step frequency using GPS tracking and pedometers. We visually observed foraging behaviour of each cow by recording selected plants and compared biomass proportions of each plant species before and after grazing. Differences among breeds were tested by Tukey Range Tests. Preference or avoidance for different plant species were derived from the coefficients of a linear, mixed-effects model. Sub-study II (Pauler *et al.*, 2019) explored long-term breed effects on pasture vegetation. We conducted an observational vegetation study in Switzerland and Germany. At 25 sites, pastures grazed by Highland cattle for at least 5 years, were compared to similar, adjacent pastures of more productive cattle. We recorded the percentage cover of all plant species, assigned them to indicator values of trampling and grazing tolerance (Briemle *et al.*, 2002) and analysed data by generalized linear mixed-effects models.

Results and discussion

Breeds differed consistently with respect to almost all factors analysed. Especially Highland cattle differed from the two more productive breeds significantly, while there was only little divergence between Original Brown and Angus×Holstein cattle: Highland cattle were significantly lighter (358 kg) than Original Brown (582 kg) and Angus×Holstein (679 kg). Claw base was smaller in Highland cattle, but it was relatively large compared to body weight (Figure 1A). Hence, physical pressure to the ground is lower in Highland cattle. Accordingly, we found significantly less trampling-adapted plant species on Highland cattle pastures (Figure 2A).

GPS and pedometers indicated that Highland cattle moved least, but used the space most evenly (Figure 1B). The more productive a breed was, the higher the forage selectivity and step frequency. Highland cattle foraged most evenly (Figure 1C) and thereby chose the diet of lowest quality. Since they were least choosy while foraging, they needed to walk shortest distances, as they just fed on what was in close proximity to their mouth. Thereby, they additionally reduce trampling pressure. Original Brown and Angus×Holstein foraged more broad-leafed grasses and legumes than Highland cattle (Figure 3), while nutrient-poor species, woody or grazing-adapted plants were consumed by Highland cattle much more frequently. The silver thistle ('Ca. acau' in Figure 3), for example, was clearly avoided by Angus×Holstein, whereas Highland cattle were indifferent. Accordingly, grazing adapted plants were significantly less abundant on pastures grazed by Highland cattle for at least 5 years (Figure 2B,C).

Highland cattle pastures were significantly more species-rich than comparable pastures of productive cattle (Figure 2D) and these differences increased with the duration a pasture was grazed by the breeds. This finding is well explained by the lower trampling pressure and the less selective foraging behaviour of Highland cattle, which prevent highly competitive species from overgrowing more susceptible plants.

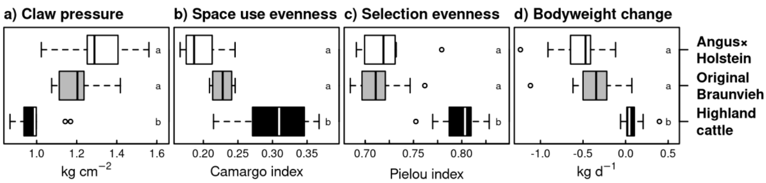

Figure 1. Differences in grazing-relevant characteristics of three cattle breeds. Different letters indicate significant differences among breeds (P<0.05).

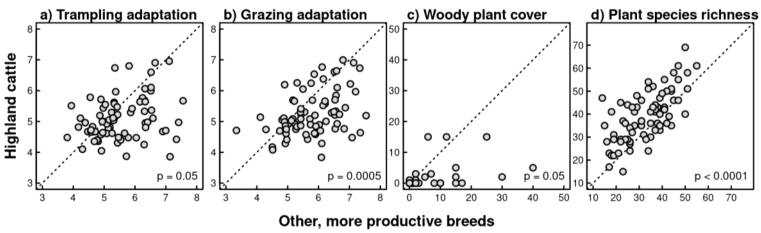

Figure 2. Vegetation indices of paired pastures grazed by Highland cattle or productive breeds.

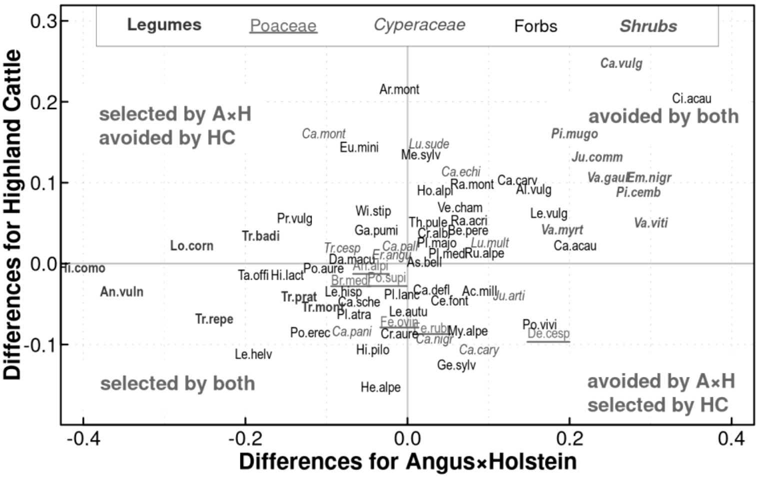

Figure 3. Preference and avoidance of plant species by Highland Cattle (HC; y-axis) and Angus× Holstein (A×H; x-axis), measured as differences in biomass proportions before and after grazing. Negative values indicate preference, positive values avoidance.

On nutrient-poor pastures, cattle commonly lose body weight. Although Highland cattle chose a diet of lower forage quality, they gained weight (0.08 kg d^{-1}), whereas the other two breeds lost 0.3 kg d^{-1} (Original Brown) and 0.6 kg d^{-1} (Angus×Holstein; Figure 1D). Highland cattle compensated the lower energy intake by their unhurried movement behaviour, their warming fur and likely by a more efficient food conversion of the fibre-rich diet.

Conclusions

Robust cattle such as Highland cattle are able to cope with the low forage quality of marginal pastures and make efficient use of them (provisioning ecosystem services). They preserve the biodiversity of semi-natural grasslands most efficiently (supporting services) and maintain open landscapes for recreation and tourism (cultural services). Hence, robust breeds are an ideal option to valorise ecosystem services of semi-natural, marginal pastures.

References

Briemle G., Nitsche S. and Nitsche L. (2002) Nutzungswertzahlen für Gefäßpflanzen des Grünlandes. *Schriftenreihe für Vegetationskunde* 38(2), 203-225.

Martin-Collado D., Boettcher P. and Bernués A. (2019) Opinion paper: livestock agroecosystems provide ecosystem services but not their components – the case of species and breeds. *Animal* 13, 2111-2113. https://doi.org/10.1017/S1751731119001277

Pauler C.M., Isselstein J., Berard J., Braunbeck T. and Schneider M.K. (2020a) Grazing allometry: anatomy, movement, and foraging behavior of three cattle breeds of different productivity. *Frontiers in Veterinary Science* 7, 1-17. https://doi.org/10.3389/fvets.2020.00494

Pauler C.M., Isselstein J., Braunbeck T. and Schneider M.K. (2019) Influence of Highland and production-oriented cattle breeds on pasture vegetation: a pairwise assessment across broad environmental gradients. *Agriculture, Ecosystems & Environment* 284, 1-11. https://doi.org/10.1016/j.agee.2019.106585

Pauler C.M., Isselstein J., Suter M., Berard J., Braunbeck T. and Schneider, M.K. (2020b) Choosy grazers: influence of plant traits on forage selection by three cattle breeds. *Functional Ecology* 34, 980-992. https://doi.org/10.1111/1365-2435.13542

Robust cattle, sheep and goats in green alder shrubs – or how to preserve mountain pastures

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Abstract

Green alder shrubs (*Alnus viridis*) increasingly overgrow mountain pastures and impair ecosystem services by loss of biodiversity, eutrophication and greenhouse gas emissions. Over the centuries, grazing livestock, especially goats, preserved these ecosystems by impeding shrub expansion. Nowadays, livestock numbers are decreasing on remote mountain pastures and goat farming has become unprofitable. A grazing experiment tested if robust cattle and sheep can replace goats as antagonists of green alder and if the available fodder is sufficient. GPS tracking and vegetation mapping were used to analyse movement behaviour and debarking activity of Dexter cattle, Engadine sheep and Pfauen goats. The forage quality of green alder and its understorey was unexpectedly high. Cattle used the space least evenly, preferred flat slopes and open pastures, spent least amount of time in green alder and did not debark any green alder branches. Engadine sheep visited the shrubs nearly as often as goats, but preferred flat slopes and short vegetation. Unexpectedly, this sheep breed debarked a significantly higher share of green alder branches than goats. Dexter cattle cannot replace goats for fast green alder clearance but they may impede shrub encroachment in the long term. Engadine sheep are well suited to recreate biodiverse semi-natural open pastures and maintain their ecosystem services.

Keywords: abandonment; biodiversity; grazing; robust breeds; green alder

Introduction

Green alder shrubs (*Alnus viridis*) increasingly overgrow mountain pastures in the European Alps but hinder natural forest succession. The main reason for shrub expansion is a reduction of farming activities, especially of goat grazing. Green alder is a nitrogen-fixing pioneer shrub and comes along with numerous negative effects including loss of appealing landscape, eutrophication of soils and downstream waters, and emission of greenhouse gases (Bühlmann *et al.*, 2016). Moreover, the understorey vegetation of green alder is species-poor and dominated by only a few broad-leaved herbs (Zehnder *et al.*, 2020). We therefore aimed to test if hardy breeds of more economically-attractive livestock species, cattle and sheep, can replace goats as green alder antagonists.

Materials and methods

A grazing experiment was set up using Dexter cattle, Engadine sheep and Pfauen goats on subalpine, shrub-encroached pastures in the eastern Swiss Alps (46°34'N, 9°50'E; 1,900-2,200 m a.s.l.). The shrub layer consisted of green alder (98%) and elderberry (2%). All chosen breeds were of low productivity and adapted to roam steep terrain and to feed on low-quality forage. We observed two cattle herds, two sheep herds and one goat herd grazing 15 paddocks. Each paddock was grazed twice, but not by different livestock species. To assess the interaction between animal type and the vegetation we measured various parameters: (1) Digestibility of green alder leaves and bark; herbage biomass and digestibility of green alder understorey, of fertile and of nutrient-poor pastures (measured in exclusion cages: 1.2×1.2 m). (2) Movement of animals was monitored by GPS trackers at a frequency of 10s using the methodology of Homburger *et al.* (2015). (3) After each rotation, areas encroached by green alder were systematically searched for signs of de-barking. We counted undamaged and damaged branches, recorded their location and calculated their ratio. Significance of differences was tested by pairwise comparison with Tukey contrasts.

Results and discussion

Green alder stands are an underestimated forage resource

Because of the high elevation, annual biomass yield was low (Table 1). However, the understorey vegetation of green alder produced 1.5 t ha^{-1} on average, and therefore it ranged between that of fertile pastures (2.3 t ha^{-1}) and nutrient-poor pastures (0.9 t ha^{-1}). In addition, measurements by Wiedmer and Senn-Irlet (2006) indicated an annual production of around 3.8 t of green alder leaves and 1 t ha^{-1} of bark. There was no significant difference in *in vitro* digestibility between the understorey vegetation of green alder and the vegetation of open pastures. The digestibility of green alder leaves was slightly lower ($P<0.05$) than for nutrient-poor pastures and understorey vegetation. The crude protein content of alder understorey and leaves was higher ($P<0.05$) than in any other forage type measured. This is explained by the additional input of symbiotically fixed nitrogen provided by green alder (Bühlmann *et al.*, 2016). Commonly, green alder and its associated vegetation are assumed to be of low forage quality. However, the relatively high productivity and the high digestibility and protein content show that this vegetation type provides an underestimated forage resource for adapted low-productive ruminants.

Ruminant species differ in feeding behaviour

All three ruminant species exploited the areas encroached by green alder (Figure 1). However, they differed in space-use evenness (Camargo evenness: cattle=0.39; sheep=0.52; goats=0.47). Cattle preferred flat slopes and open pastures more clearly than sheep and goats (relative presence in green alder stands: cattle=0.55; sheep=0.76; goats=0.80). Cattle were observed foraging on understorey vegetation, leaves and buds, but they did not debark green alder branches. Unexpectedly, Engadine sheep debarked green alder branches frequently, especially at the edge of the stand, where they could access the shrubs more easily than in the centre. Debarked branches die off within a year, because they lose their transport capacity for assimilates. Thereby, debarking represses green alder stands in the long term. Goats showed almost no preference for open pastures over dense green alder stands. They consumed alder leaves and buds but debarked it less frequently than Engadine sheep (sheep=7.4% of green alder branches; goats=0.8%). In contrast, the bark of the few elderberry trees (*Sorbus aucuparia*) growing in the green alder stands was almost completely stripped. Goats consumed the bark of elderberry immediately when released to the paddocks, whereas they debarked green alder only when very little elderberry was left over.

Table 1. Annual biomass yield, *in vitro* digestibility of organic matter and crude protein content in the dry matter of different vegetation types and plant parts of green alder.[1]

Vegetation type	Annual yield (t ha^{-1})	Digestibility (g kg^{-1} DM)	Crude protein (g kg^{-1} DM)
Fertile and nitrophilous pastures	2.25±0.89b	487±114bc	117±37a
Nutrient-poor pastures and wetlands	0.93±0.52a	531±60c	133±34a
Green alder understorey vegetation	1.53±0.89ab	559±75c	190±39b
Green alder leaves	3.8*	439±54b	211±21b
Green alder bark	1.04*	163±12a	78.1±8.8a

[1] Shown are mean values ± one standard deviation. Different letters indicate significant differences of pairwise comparison with Tukey contrasts at 5% level. * Estimates measured and published in Wiedmer and Senn-Irlet (2006).

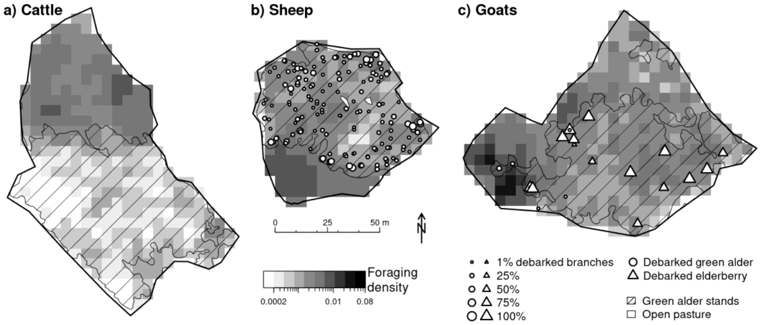

Figure 1. Foraging density (i.e. the relative number of GPS locations classified as foraging per grid cell) and proportion of debarked branches in exemplarily selected paddocks grazed by Dexter cattle (a), Engadine sheep (b) and Pfauen goats (c). For locations without debarking, no symbols are shown.

Conclusions

The forage provided by green alder stands is generally underrated in the nutrition of adapted low-productive ruminants. In contrast to previous assumptions by practitioners and scientists, green alder stands are a valuable forage resource in marginal mountain areas.

Cattle have the smallest direct impact on green alder, but they exploit the understorey vegetation and can open up green alder areas for other types of animals. They cannot replace goats for fast green alder clearance, but they are able to make use of the forage available in green alder stands. Engadine sheep actively counteract green alder expansion by consuming its bark. Hence, they provide an attractive option for regaining open pastures, but they mainly stay within the edge of dense alder stands. Since goats prefer other woody species over green alder and debark them first, they must be kept under high grazing pressure to drive back green alder shrubs. Otherwise, they may only hinder the regeneration of late-successional forest.

All ruminants observed were able to exploit the forage available in green alder stands and thereby, they may at least slow down shrub expansion. Hence, hardy breeds are an important tool to maintain biodiverse, open pastures and mitigate the negative environmental effects of green alder encroachment.

References

Bühlmann T.H., Hiltbrunner E. and Körner C. (2016) Shrub expansion of *Alnus viridis* drives former montane grassland into nitrogen saturation. *Ecosystems* 19, 968-985.

Homburger H., Lüscher A., Scherer-Lorenzen M. and Schneider M.K. (2015) Patterns of livestock activity on heterogeneous subalpine pastures reveal distinct responses to spatial autocorrelation, environment and management. *Movement Ecology* 3, 1-15.

Wiedmer E. and Senn-Irlet B. (2006) Biomass and primary productivity of an *Alnus viridis* stand – a case study from the Schächental valley, Switzerland. *Botanica Helvetica* 116, 55-64.

Zehnder T., Lüscher A., Ritzmann C., Pauler C.M., Berard J., Kreuzer M. and Schneider M.K. (2020) Dominant shrub species are a strong predictor of plant species diversity along alpine pasture-shrub transects. *Alpine Botany* 130, 141-156.

Agrivoltaism, in search of the right coupling between energy production and management of ruminant herds

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Abstract

Fostered by a favourable national strategic framework, renewable energies are booming in France, particularly ground-based photovoltaic production. Access to degraded land is increasingly complicated and limited, managers are then turning to agricultural land. As the use of agricultural land for development projects is highly regulated, developers of photovoltaic power plants have an obligation to set up agrivoltaic projects combining electricity production activities and agricultural activities. The co-activity requires taking into account the issues of the different stakeholders, and a reflection on the arrangements to be planned from the design of the project.

Keywords: agrivoltaic, pasture, ruminants, solar parks

Introduction

The French energy and climate strategy presented in November 2018 set the ambitious goal of achieving carbon neutrality by 2050. It is based on two strategies: the National Low-Carbon Strategy, and the Multiannual Energy Programming (PPE), which sets the priorities for action in the energy sector for the coming decade. While the previous Multi-Year Energy Programme of 2016 had set a target for 2018 of 10.2 GW, the PPE presented in 2018 goes further, as the goal is to double photovoltaic capacity by 2023 (to reach 18.2 to 20.2 GW) and to multiply them by 3 or 4 by 2028 (to reach 35 to 45 GW).

A new activity lacking references

Most photovoltaic-livestock co-activity projects in France involve sheep farming, but schemes involving cattle are emerging thanks to technological innovations in solar array design.

Since 2019, the French livestock institute (Idele) has been providing its knowledge of ruminant production systems to companies in the solar energy sector. Its activity covers different levels of project support, from design support to optimize grazing within the solar park, to the study of animal behaviour and performance, and the grass production under solar arrays through experimental systems intended to shed light on the technical and economic questions raised by agrivoltaic.

Although the scientific literature highlights a number of advantages for agrivoltaic systems, both for the operator and the farmer and his animals, there are still gaps on the impacts of this co-activity. From the agronomic point of view, what are the effects of full shade on biomass production and how does it impact on botanical species richness? From the animal husbandry side, how does this new environment impact animal performance and reproduction? How does grazing under solar arrays impact animal behaviour and welfare? From the farmers' point of view, what does it change in terms of the working conditions and is it an efficient source of income diversification? These are some of the questions that need to be answered quickly.

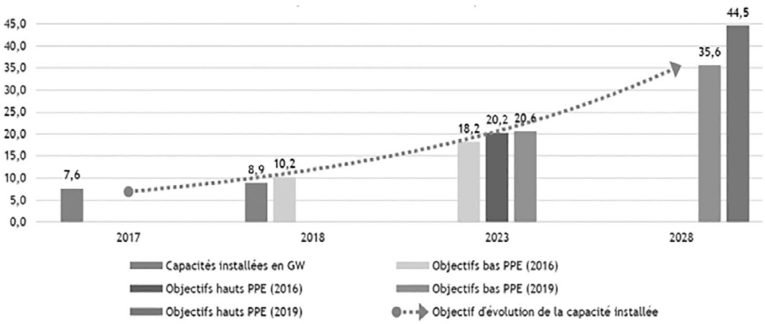

Figure 1. Development objectives for solar energy production in France (GW) (Ademe, présentation au colloque INES, 2019).

It is therefore essential to develop project monitoring and to continue experimentation on the basis of tried and tested protocols. Data, observations and experiments are needed in different pedoclimatic contexts and with different types of solar equipment to build a strong reference system.

Building up from the first observations to support future projects

After three years of project support and collaboration with several companies in the agrivoltaic sector (NEOEN, TSE and VOLTALIA) and in partnership with the Sheep Farmers' Federation (FNO), the French livestock institute (Idele) has set the framework for further research by producing a practical guide focusing on the co-activity of photovoltaic production with ruminant. This document constitutes the technical basis of this reflection and provides insights for a smart partnership between the solar operator and the farmer: from the design of the power plant to the management of the grazing system, including the partnership aspect.

This guide formulates simple recommendations so that the projects are set up to enhance the co-activity and not only the photovoltaic production. It aims to disseminate recommendations that can be used before the building of the power plant, in order to increase the success of the project especially for farmers. The recommendations put forward in this guide are based on feedback from farmers currently grazing in solar parks and good grazing management practices.

This guide offers technical recommendations concerning:

- the choice of photovoltaic equipment and its installation conditions;
- the addition of equipment specific to the livestock activity;
- the strategy for managing the vegetation cover;
- the implementation of a grazing technique adapted to the management objectives;
- the partnership arrangements between the farmer and the solar manager.

Conclusions and outlook

The acceptability and development of agrivoltaic systems depends largely on the ability to combine the technical and economic interests of farmers and solar managers, while limiting the negative effects on agricultural production or land management. The presence of arrays generating more or less mobile shadows according to the technologies deployed, and the creation of an electromagnetic context raise questions that need to be clarified in order to ensure the success of future projects and to reassure farmers in the management of their land and flocks with this new equipment. For solar managers, grassland often represents opportunities linked to its importance in certain regions and the relative ease of setting up equipment, unlike other agricultural production (vineyard, vegetables, arboriculture or even field crops). Although the construction of power plants is therefore easier on grassland areas, they have an impact on livestock farming practices. The shade produced by the arrays, and the microclimate that this could generate, are often put forward as a comfort for the animals and as a way to adapt to a hotter climate. Depending on the context, this shade can be a protection against evapotranspiration and therefore a factor in maintaining plant production in summer or, on the contrary, a limitation of radiation and a factor in reducing photosynthesis. The questions are therefore numerous and diverse, which is why Idele and its partners are currently mobilized through monitoring and experimental systems in the search for references on this topic.

References

Andrew A.C. (2020) Lamb growth and pasture production in agrivoltaic production system. For the degree of Honors Baccalaureate of Science in Biology presented on August 21, 2020.

Armstrong A., Ostle N.J. and Whitaker J. (2016) Solar Park microclimate and vegetation management effects on grassland carbon cycling. *Environmental Research Letters* 11(7), 074016.

Barron G.A. (2019) Agrivoltaics provide mutual benefits across the food-energy-water nexus in drylands. *Nature Sustainability* 2, 848-855.

Dupraz C., Marrou H., Talbot G., Dufour L., Nogier A. and Ferard, Y. (2011) Combining solar photovoltaic panels and food crops for optimising land use: towards new agrivoltaic schemes. *Renewable Energy* 36(10), 2725-2732.

Ehret M., Graß R. and Wachendorf M., (2015) The effect of shade and shade material on white clover/perennial ryegrass mixtures for temperate agroforestry systems. *Agroforestry Systems* 89, 557-570.

Hernandez R.R., Easter S.B., Murphy-Mariscal M.L., Maestre F.T., Tavassoli M., Allen E.B., Barrows C.W., Belnap J., Ochoa-Hueso R., Ravi S. and Allen M.F. (2014) Environmental impacts of utility-scale solar energy. *Renewable and Sustainable Energy Reviews* 29, 766-779.

Kirilov A., Vasilev E., Pachev I. and Stoycheva I. (2013) Changements dans la composition d'une association luzerne – dactyle dans les conditions d'un parc agro-photovoltaïque. *Prairies, systèmes fourragers et changement climatique.* Journées AFPF 2013, pp. 196-197.

Madej L. (2020) Dynamique végétale sous l'influence de panneaux photovoltaïques sur 2 sites prairiaux pâtures. *Milieux et Changements globaux*, hal-03121955.

Pang K., Van Sambeek JW., Navarrete-Tindall NE., Lin C-H., Jose S. and Garrett HE. (2017) Responses of legumes and grasses to non-moderate, and dense shade in Missouri, USA. I. Forage yield and its species-level plasticity. *Agroforestry Systems* 93, 11-24.

Sharpe K.T., Heins B.J., Buchanan E.S. and Reese M.H. (2021) Evaluation of solar photovoltaic systems to shade cows in a pasture-based dairy herd, *Journal of Dairy Science* 104(3), 2794-2806.

Semchenko M., Lepik M., Gotzenberger L. and Zobel K. (2012) Positive effect of shade on plant growth: amelioration of stress or active regulation of growth rate? *Journal of Ecology* 100, 459-466.

A systematic review of threats in permanent grassland cultural ecosystem services

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Abstract

The degradation of ecosystems happens at an unprecedented rate, threatening the provision of ecosystem services and ultimately limiting human well-being. We conducted a systematic literature review evaluating the threats surrounding cultural ecosystem services (CES, namely recreation and landscape aesthetics) in European permanent grasslands. We classified threats into underlying causes, direct threats and consequences, and solutions that have been suggested for their mitigation. We screened 13,719 papers on their relevance, of which 77 studies in 71 articles were extracted and qualitatively analysed. We found the most common threats to be land-use and management change processes, while recreational activities also created negative feedback loops, affecting the ecosystem, biodiversity and CES themselves. Suggested solutions were most commonly socio-economic and institutional measures to improve rural populations' livelihood and improved communication with relevant stakeholders. With those tools, the continued supply of CES can be guaranteed, as they play a crucial role in reconnecting people with nature and thus ensuring future human well-being.

Keywords: direct drivers, Europe, nature conservation, rural development, tourism

Introduction

In the ecosystem services literature, cultural ecosystem services (CES) have only recently become more prominent, shifting the focus from an economic-centred to a socio-ecological approach (Plieninger *et al.,* 2015). CES are defined as the non-material benefits for people and their well-being from recreational and aesthetic experience, spiritual and educational values (MEA, 2005). Through their prevailing trade-off relation with other services (Allan *et al.,* 2015), and accelerating global challenges such as the degradation of ecosystems, climate change or the COVID-19 pandemic (IPBES, 2018; 2020), CES are increasingly under pressure. For this systematic review, we evaluated the threats on recreation and landscape aesthetics in permanent grasslands described in the literature, assessing what consequences they may have and what solutions have been suggested for their mitigation.

Materials and methods

The Scopus and CAB Abstract databases were searched on 5 November 2019 for CES in European permanent grasslands. We read 196 potentially relevant papers out of 13,719 retrieved articles and selected 71 papers that fit our scope, out of which 77 studies were extracted. A substantial part of the data analysis was the identification and classification of threats, followed by a qualitative analysis. We distinguished four levels of threat interactions, namely underlying causes, direct threats, consequences, and suggested solutions to prevent or mitigate negative effects. As threats not only impact beneficial ecosystem services but may also be caused by their usage, we differentiated between threats affecting CES (*direct threats* to *CES*) and threats from CES that affect grassland ecosystems and their services (*direct threats* from *CES*). Due to the lack of detailed descriptions of the studied grassland types in the original papers, we were unable to compare threats according to grassland types.

Results and discussion

By far the most direct threats to CES came from land-use and management changes (e.g. abandonment, intensification, building up), the predominant driver during the second half of the 20th century (Stoate *et al.,* 2009). Underlying causes were primarily socio-economic, institutional and demographic. Another major threat was the perception of nature during recreational activities. When nature is considered as a decorative background for human activities rather than an intrinsic, alive and distinct value, it may lead to the destruction of the environment (Syrbe and Grunewald, 2017). Natural threats related to climate change were minor in our analysis, reported only six times as having adverse effects on recreation. The reduction of suitable areas for skiing and natural afforestation due to a rise in temperatures might hit the tourism industry substantially in the future (IPCC, 2014). The most widely suggested solutions were to ensure local livelihoods, preserve a healthy environment and develop a sustainable, local socio-economic system. Furthermore, CES might be the most important communication channel to raise awareness for ecosystem protection due to its close links to human well-being.

The negative impacts of CES on grasslands were predominantly driven by the high demand for recreation. Touristic activities such as hiking, skiing and vehicle use were the most mentioned threats caused by recreation, mainly affecting vegetation, soil and wildlife, but also the recreational and aesthetic quality of an area (Syrbe and Grunewald, 2017). Further pressures consisted of the development of tourist facilities (e.g. roads and ski lifts) and accommodation. Among the less-mentioned direct threats were hunting tourism and artificial snow. With increasingly snow-poor winters due to a changing climate, skiing facilities are likely to move to higher altitudes (IPCC, 2014), thus extending negative impacts into the susceptible high-Alpine zone (IPBES, 2018). Therefore, developing strategies and recognising new recreational locations for providing quality outdoor recreation will be essential (Askew and Bowker, 2018). Suggested solutions included tourism regulation and economic and regulatory tools to avoid ecosystem overuse. Further, land-use planning, the development of conservation strategies and improved communication and education were mentioned.

Conclusions

Land-use and management change was the most dominant threat to CES, while the demand for recreation was the biggest threat from CES to the ecosystem and other services. The studies reviewed in the present study showed the need for a multi-actor approach with integrated rural development and traditional knowledge in grassland management. We further found that there are still considerable knowledge gaps regarding the effects of new challenges on CES, such as the recent COVID-19 pandemic or climate change. To date, there are only a few studies about such threats, yet climate change is expected to become a decisive future driver (IPBES, 2018). The discussed mitigation tools would guarantee the continued provision of CES and will help reconnect people with nature, thus ensuring future human well-being.

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References

Allan E., Manning P., Alt F., Binkenstein J., Blaser S., Blüthgen N., Böhm S., Grassein F., *et al.* (2015) Land use intensification alters ecosystem multifunctionality via loss of biodiversity and changes to functional composition. *Ecology Letters* 18, 834-843.

Askew A.E., and Bowker J.M. (2018) Impacts of climate change on outdoor recreation participation: Outlook to 2060. *Journal of Park and Recreation Administration* 36, 97-120.

Chan K.M.A., Guerry A.D., Balvanera P., Klain S., Satterfield T., Basurto X., Bostrom A., Chuenpagdee R., *et al.* (2012) Where are cultural and social in ecosystem services? A framework for constructive engagement. *BioScience* 62, 744-756.

Daniel T.C., Muhar A., Arnberger A., Aznar O., Boyd J.W., Chan K.M.A., Costanza R., Elmqvist T., *et al.* (2012) Contributions of cultural services to the ecosystem services agenda. *Proceedings of the National Academy of Sciences of the USA* 109: 8812-8819.

IPBES (2018) The IPBES regional assessment report on biodiversity and ecosystem services for Europe and Central Asia. Rounsevell M., Fischer M., Torre-Marín Rando A., and Mader A. (eds.). Secretariat of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services, Bonn, Germany 836 pp.

IPBES (2020) Workshop report on biodiversity and pandemics of the Intergovernmental Platform on Biodiversity and Ecosystem Services. Daszak P., Amuasi J., das Neves C.G., Hayman D., Kuiken T., Roche B., Zambrana-Torrelio C., *et al.* (eds.) Secretariat of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services, Bonn, Germany. 100 pp.

IPCC (2014) Impacts, adaptation, and vulnerability: contribution of working group II to the fifth assessment report of the Intergovernmental Panel on Climate Change. Barros V.R., Field C.B., Dokken D.J., Mastrandrea M.D., Mach K.J., *et al.* (eds.) Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, UK, 1820 pp.

Kosanic A. and Petzold J. (2020) A systematic review of cultural ecosystem services and human wellbeing. *Ecosystem Services* 45, 101-168.

Millennium Ecosystem Assessment (2005) Ecosystems and human well-being: Synthesis. Island Press, Washington, DC, USA. 141 pp.

Plieninger T., Bieling C., Fagerholm N., Byg A., Hartel T., Hurley P., López-Santiago C.A., Nagabhatla N., *et al.* (2015) The role of cultural ecosystem services in landscape management and planning. *Current Opinion in Environmental Sustainability* 14, 28-33.

Stoate C., Báldi A., Beja P., Boatman N. D., Herzon I., van Doorn A., de Snoo G. R., Rakosy L., *et al.* (2009) Ecological impacts of early 21st century agricultural change in Europe - A review. *Journal of Environmental Management* 91, 22-46.

Syrbe R.U., and Grunewald K. (2017) Ecosystem service supply and demand-the challenge to balance spatial mismatches. *International Journal of Biodiversity Science, Ecosystem Services and Management* 13, 148-161.

Trade-offs between services rendered by semi-natural grasslands of the Vosges massif (France)

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Abstract

Grasslands are at the heart of multiple expectations on the part of farmers and society. The objective of this study was to assess the services provided by semi-natural grasslands, and the trade-offs between these services. Starting from a survey of 150 grasslands in the Vosges massif, we selected 58 that have been monitored for two consecutive years (2018-2019). Grassland services were assessed through measurements (M) and indicator calculations (IND) including dry matter production (M), feed value and anti-oxidant content of grass (M), physicochemical composition of the soil (M), carbon sequestration (M), floristic biodiversity (M), cost of production and replacement (M), product quality (cheese and meat) (IND), animal health (IND), pollinator value (IND), ecological conservation status (IND). The study of the trade-offs between these services shows that there is no binary opposition between economic value and environmental value. On the contrary, certain environmental services may be associated with the economic interest of livestock farmers.

Keywords: ecosystem service, fodder production, economy, product quality, animal health, biodiversity

Introduction

Semi-natural grasslands can provide many services to farmers and society (Boval and Dixon, 2012), but their economic value and their ability to provide services to society are often considered incompatible. However, many studies show that there may be an economic interest in conserving these species-rich grasslands (Bengtsson *et al.*, 2019; Plantureux, 2020). The analysis of the trade-offs between the services rendered by these grasslands is essential to justify their conservation. In this study, the objective was to study these trade-offs on a large number of services, including fodder production and biodiversity, but also looking at economic aspects, animal health and product quality (milk and meat).

Materials and methods

We surveyed 150 permanent grasslands from the Vosges Mountains (North-Eastern France), and we characterized each grassland using phytosociological and agronomic classifications (Mesbahi *et al.*, 2020). We selected the 20 most important grassland classes, and for each of these we selected about 3 representative grasslands. Elevation ranged from 184 to 1,222 m a.s.l and soil pH from 4.2 to 8, grasslands were cut and/or grazed and N-fertilization varied from 0 to 259 kg ha^{-1} (mineral and organic fertilization, and animal deposition). In 2018 and 2019, we realized botanical relevés within areas with homogeneous vegetation types. Grass samples were taken 4 times per year in six 0.5 m^2 quadrats per grassland, except in the absence of grass growth. Grassland services were assessed through direct measurements or observations (M) and indicator calculations (IND), as detailed in Table 1. Principal Component Analysis (PCA) was performed in order to study the links between services.

Results and discussion

Five groups of variables appeared (Figure 1): Group 1 = grass production (code in Table 1: GP1, GP2) associated with biodiversity (B1, B2) and anti-infective potential (AH1), Group 2 = forage quality (GF1, GF2), Group 3 = product quality (QP1, QP2, QP3, QP4), 4), Group 4 = flexibility (GP3) and oligotrophilous richness (B4), and Group 5 = biodiversity (B3,B6) and production costs (E).

Table 1. Evaluated services.[1]

Service	Evaluation	Code	Abbreviation	Variable
Grass production	M	GP1	Yield	annual dry matter production
	IND	GP2	VP	pastoral value
	IND	GP3	Flexibility	grass production flexibility
	IND	GP4	Earliness	grass production earliness
Grass feed value	M	GF1	UFL	forage energy content
	M	GF2	PDIN	forage protein content
	IND	GF3	Milk_pot	potential milk production
Biodiversity	M	B1	SpecRich	plant species richness
	IND	B2	Shannon	Shannon diversity index
	IND	B3	Conserv_status	ecological conservation status
	M	B4	oligod_sample	oligotrophilous species richness
	M	B5	Family_rich	plant family richness
	IND	B6	ValeurPoli	pollinator value
C sequestration	M	C	SoilCarb_030	C content (top 30 cm soil)
Economy	M	E	Cost/DMT	production cost in €/DM ton
Quality of products	IND	QP1	Aroma	cheese aromatic value
	IND	QP2	Cheese_Texture	cheese texture
	IND	QP3	Cheese_Fat_color	cheese fat colour
	IND	QP4	Meat_antiox	meat antioxidant faculty
	IND	AH1	AnimHealth_antiox	antioxidant value
Animal health	M	AH2	IC50Trol_DPPH	forage antioxidant analysis
	IND	AH3	AnimHealth_antiinf	anti-infection value

[1] M = measured or calculated from observations; IND = indicators.

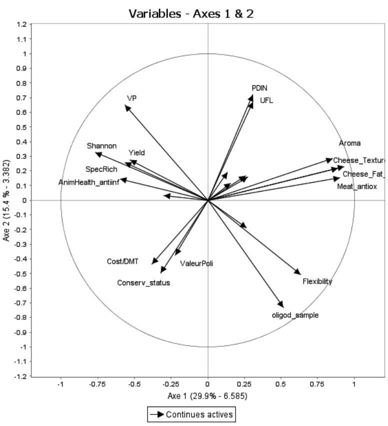

Figure 1. Principal Component Analysis on services rendered by grasslands (n=58) in the Vosges mountains (France). Abbreviations are detailed in Table 1.

It is thus observed that there is no binary opposition between economic and environmental variables. On the contrary, production is associated with specific richness (Group 1), and flexibility with oligotrophilous species richness (Group 4). The opposition between Group 3 (product quality) and Group 5 (biodiversity) is explained by the effect of grazing. This mode of exploitation promotes the quality of the products (favourable impact of fresh grass consumption) but is not favourable to flowering species. The presence in Group 5 of the cost of production (E) might seem surprising because grazing is known as the least expensive method of harvesting. The variable (E) is calculated by dividing cost by the yield, and pastures are penalized here by their lower yield. As found by Grace *et al.* (2016) we found no relations between biodiversity and forage quality, but our results challenged previous studies highlighting a trade-off between biodiversity and yield (Le Clec'h *et al.*, 2019) possibly due to the larger environmental gradient but smaller number of grasslands we studied.

For each of the 58 grasslands in the sample, a unique combination of services is ultimately observed, with environmental and economic strengths and weaknesses.

Conclusions

The trade-off between the services provided by the permanent grasslands is not limited to an opposition between environmental services and production or economic services. The nature of the trade-offs varies from one grassland plot to another. This observation should encourage the conservation of a diversity of types of grassland within a farm or territory, in order to jointly provide a range of services. However, more studies are needed to generalize our observations to other geographical areas.

References

Bengtsson J., Bullock J.M., Egoh B., Everson C., Everson T., O'Connor T., O'Farrell P.J., Smith H.G. and Lindborg R. (2019) Grasslands - more important for ecosystem services than you might think. *Ecosphere* 10, e02582.

Boval M. and Dixon R.M. (2012) The importance of grasslands for animal production and other functions: a review on management and methodological progress in the tropics. *Animal* 6, 748-762.

Grace C., Boland T.M., Sheridan H., Lott S., Brennan E., Fritch R. and Lynch M.B. (2016) The effect of increasing pasture species on herbage production, chemical composition and utilization under intensive sheep grazing. *Grass and Forage Science* 73 (4), 852-864.

Le Clec'h S., Finger R., Buchmann N., Gosal A.S., Hörtnagl L., Huguenin-Elie O., Jeanneret P., Lüscher A., Schneider M.K. and Huber R. (2019) Assessment of spatial variability of multiple ecosystem services in grasslands of different intensities. *Journal of Environmental Management* 251, 109372.

Mesbahi G., Michelot-Antalik A., Goulnik J. and Plantureux S. (2020) Permanent grassland classifications predict agronomic and environmental characteristics well, but not ecological characteristics. *Ecological Indicators* 110, 105956.

Plantureux S. (2020) Reconciling production and biodiversity in management of pastures and grasslands. In: Barberi P. and Moonen A-C. (eds.) *Reconciling Agricultural Production and Biodiversity Conservation*. pp. 38 et seq.

Grass dry matter yield and plant nutrient removal following fertilization with wood ash and digestate

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Abstract

Alongside their use in forage production, grasses can be used successfully for bio-energy, and help in uptake and recovery of nutrient-overloaded soils and promote the re-use of plant nutrients via bioenergy waste products applied as fertilizers. Dry matter yield (DMY) of above-ground biomass and removal of plant nutrients (NPK) by harvest of reed canary grass (RCG) (*Phalaris arundinacea* L.) and *Festulolium* (×*Festulolium)* were investigated using two harvest regimes: one-cut (delayed harvest) and two-cut mowing. Four fertilizer options with an equivalent NPK rate ($N_{100}P_{35}K_{133}$) were chosen: mineral fertilizers (MF); wood ash (WA), digestate used once (D1) and digestate used twice (D2) per season. Missing plant nutrients in wood ash and digestate treatments, if necessary, were compensated using mineral fertilizers. A control with no-fertilizer use was also included. The DMY and plant nutrient removal were largely influenced by grass species, fertilizer treatment and mowing regime. The highest DM yields for both species was obtained from WA and MF treatments. Among the species, RCG was the more productive. Although no significant differences in plant nutrient removal between mowing regimes were found for *Festulolium*, for RCG there was significantly lower nutrient removal under the one-cut (delayed) mowing treatment.

Keywords: *Festulolium*, reed canary grass, mowing regime

Introduction

Perennial grasses have many advantages due to their multifunctionality and high productivity under different soil and climate conditions. In addition to forage for livestock, grass biomass is good feedstock for energy production and can contribute greatly to saving soil plant nutrients. The potential for re-use of plant nutrients is becoming increasingly important with rising costs and need for saving resources. It is therefore important to explore the use of alternative fertilizers, including the waste products of bioenergy such as wood ash and digestate, as these products are rich in plant nutrients (Koszel and Lorencowicz, 2015; Fuzesi *et al.*, 2015). The aim was to study the effect of different fertilizers with equivalent NPK rate on grass dry matter yield (DMY) and nutrient removal in different grass mowing regimes.

Materials and methods

An experiment was carried out on a fine sandy loam soil with pH_{KCl} 5.9-6.5, moderate OM content (3.2%), high phosphorus and moderate plant available potassium content. Four different treatments for reed canary grass (RCG) and *Festulolium*, with equivalent amounts of plant nutrients ($N_{100}P_{35}K_{133}$) were compared: mineral fertilizers (MF), wood ash (WA), digestate used once (D1) or twice (D2) a season, and a no-fertilizer control. Full rate of fertilizers was applied in early spring after vegetation recovery, with except in treatment D2 (split application of digestate), where the other half was used in the autumn after mowing. Mineral fertilizers were used for balancing nutrients in the digestate and WA treatments, the same as MF treatment (ammonium nitrate, superphosphate, potassium chloride) (Table 1).

Table 1. Plant nutrients supplemented with mineral fertilizers, to reach $N_{100}P_{35}K_{133}$.

Treatment	2012 (sowing year)			2013-2015 (harvest years)		
	N	P	K	N	P	K
WA	49.5	9.6	0	99.0	26.6	0
Digestate	0	0	10.8	0	16.6-28.8	44.8-72.2

DMY and NPK removal data were collected for one-cut or delayed harvest at autumn (1-CMR) and two-cut mowing regime – 1st cut at full heading, 2nd at the plant senescence in autumn (2-CMR) for three years of use. ANOVA was used to analyse results and F-test for the assessment of significance of means at $LSD_{0.05}$.

Results and discussion

DMY was significantly affected by fertilization treatment, mowing regime and grass species. The application of all fertilizers produced considerably higher DMY than the no-fertilizer option (Figure 1). The best results, averaged over the three years, were obtained from mineral fertilizer (MF) and wood ash (WA), with no significant differences between them: 8.01 and 8.11 t ha^{-1} for RCG; 5.96 and 6.18 t ha^{-1} for *Festulolium*, respectively. The application of both full rate (D1) and split rate (D2) of digestate gave a significant increase in yield with no significant differences between them. DMY by mowing regimes varied depending on the grass species and year of use; however, there was higher DMY on average for both species under the one-cut mowing regime (1-CMR) than two-cut mowing (2-CMR), with 5.81 t ha^{-1} and 5.12 t ha^{-1}, respectively. Overall, taking into account treatments and mowing regimes, the RCG produced significantly higher DMY (6.47 t ha^{-1}). Factor analysis showed that the greatest effect on the DMY was fertilizer, 56.6% on average for both species. The next important factors were species (27.7%) and mowing regime (4.1%).

NPK removal from the soil with harvest during the whole research period varied largely between year and treatment and it was closely related to DMY and its chemical content. The greatest effect was with grass species and mowing regime. Nitrogen (N) removal in 2-CMR on average per year for RCG varied from 54.0 kg ha^{-1} N (no-fertilizer) to 142.9 kg ha^{-1} N (WA). In 1-CMR, using delayed harvesting, N removal was about two times lower and varied from 39.0 kg ha^{-1} N (no-fertilizer) to 69.1 kg ha^{-1} N (MF) (Table 2). For *Festulolium* the N removal was considerably lower, mainly determined by DMY differences. It was very similar in both mowing regimes: from 23.2 kg ha^{-1} N (no-fertilizer) to 73.3 kg ha^{-1} N (WA) using 2-CMR, and from 22.1 kg ha^{-1} N (no-fertilizer) to 78.7 kg ha^{-1} (MF) using 1-CMR.

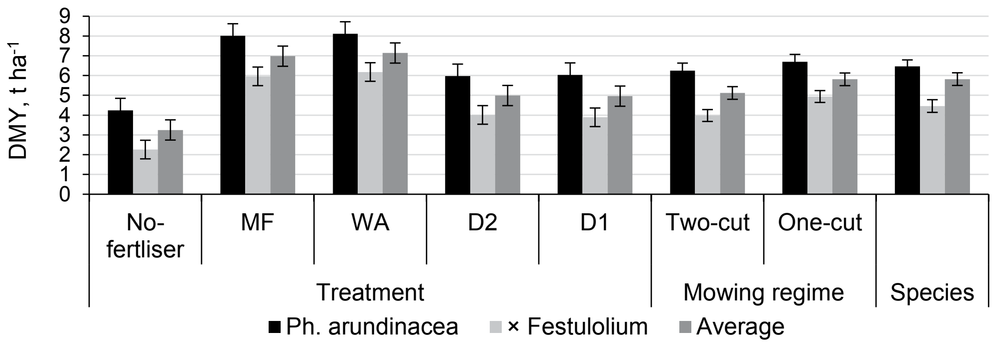

Figure 1. DMY in t ha^{-1} under the influence of different factors, average yield over three years under two different mowing regimes.

Table 2. Plant nutrient removal by harvested above-ground biomass, kg ha^{-1} year^{-1}.

Species	Treatment	Nitrogen (N)		Phosphorus (P)		Potassium (K)	
		two-cut mowing	one-cut mowing	two-cut mowing	one-cut mowing	two-cut mowing	one-cut mowing
Phalaris arundinacea L.	No-fertilizer	54±3.2	39±3.1	11±0.6	10±0.8	77±4.6	38±3.0
	MF	113±3.1	69±0.4	21±0.6	11±0.1	172±4.5	59±0.4
	WA	143±8.3	68±5.2	21±1.3	12±0.9	168±9.8	62±4.7
	D1	95±6.1	56±2.7	17±1.1	10±0.5	142±9.5	58±2.8
	D2	81±4.3	41±0.8	16±0.8	9±0.2	126±7.2	67±1.4
×*Festulolium*	No-fertilizer	23±2.2	22±2.4	4±0.4	3±0.3	36±3.5	32±3.5
	MF	67±1.5	79±4.1	8±0.2	6±0.3	90±2.1	108±5.6
	WA	73±4.7	77±2.8	9±0.5	5±0.2	96±5.9	112±4.1
	D1	45±1.6	42±2.8	7±0.3	4±0.3	74±2.6	71±4.8
	D2	39±1.5	37±2.1	7±0.3	4±0.3	71±2.7	44±2.6

Removal of phosphorus (P), compared to the N, was significantly lower It also varied relatively less between mowing regimes: from 10.6 kg ha^{-1} P (no-fertilizer) to 20.9 kg ha^{-1} P (WA) using 2-CMR; and from 9.7 kg ha^{-1} P (no-fertilizer) to 11.5 kg ha^{-1} P (WA) using 1-CMR for RCG. P removal with DMY of *Festulolium* was on average two times lower in comparison with RCG.

Potassium (K) removals were highest, they exceeded N removals approximately 1.5-2 times. Significant differences were found both among mowing regimes and grass species. The greatest K amount (172.1 kg ha^{-1} K per year) was removed by RCG under treatment 2-CMR (with MF), whereas under 1-CMR the highest K removal (62.3 kg ha^{-1} K) was found in WA. For *Festulolium*, the highest K removal in both mowing regimes was found in WA treatment: 95.5 and 111.7 kg ha^{-1} K in 2-CMR and 1-CMR, respectively (Table 2). Application of WA and MF resulted in a significantly higher K removal, compared to those of both digestate treatments, where K removal under 2-CMR ranged between 126.1 kg ha^{-1} K (D2) and 141.5 kg ha^{-1} K (D1). In general, for RCG in all fertilizer treatments, 2-3 times less K was removed under 1-CMR than 2-CMR. In contrast to RCG, K removals by mowing regimes in *Festulolium* did not differ significantly. This clearly demonstrates the ability of RCG to transfer nutrients from aboveground biomass to roots during senescence.

Conclusions

The DMY and plant nutrient removal was largely influenced by grass species, fertilizer and mowing regime. Higher yields were produced by (1) RCG, using one-cut mowing regime, and (2) both species with WA and MF treatments. At the same time, we found that RCG under a one-cut mowing regime extracted much less nutrients. Results showed significant differences in NPK uptake and utilization between the two grass species and confirm the possibility to reduce fertilizer consumption in rhizomatous grasses using delayed mowing regime.

References

Fuzesi I., Heil B. and Kovacs G. (2015) Effects of wood ash on the chemical properties of soil and crop vitality. *Acta Silvatica at Lignaria Hungarica* 11 (1), 55-64.

Koszel M. and Lorencowicz E. (2015) Agricultural use of biogas digestate as a replacement fertilizers. *Agriculture and Agricultural Science Procedia* 7, 119-124.

Provisioning ecosystem services of fertilized meadows and pastures differ in their response to organic management

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Abstract

The provisioning Ecosystem Services (ES) of organically managed grasslands could be compromised, compared to grasslands under non-organic management, due to restrictions regarding mineral fertilization. We investigated this by measuring forage yield increase per day and feed value in 25 pairs of organic and non-organic fertilized meadows (mown) and pastures (grazed) in the canton of Solothurn (Switzerland). Lower forage yield and feed value in organic pastures were related to lower phosphorus (P) in topsoil compared to non-organic pastures. However, in meadows, organic management had no effect on forage yield and feed value as soil P was hardly affected by organic management. From these findings we conclude that forage provision does not considerably differ between organic and non-organic meadows, but in pastures we see potential indications of nutrient limitation under organic management. Future research should thus assess organic pasture management in more detail to close this production gap.

Keywords: ecosystem services, grassland, organic management, provisioning services, weeds

Introduction

Organic grasslands do not receive synthetic pesticides or mineral fertilizers (Swiss Federal Council, 2018). This could lead to reduced yields (i.e. provisioning Ecosystem Services; ES). However, organic management could potentially help the grasslands to provide a wider range of other, non-monetary ES (e.g. Knudsen *et al.*, 2019; Mäder *et al.*, 2002;). In this study, we focus on the provisioning ES of grasslands in Swiss agriculture, aiming to find out whether organic grasslands have lower biomass yields and lower feed quality than non-organically managed grasslands.

Materials and methods

Intensively managed meadows (n=26) and intensively managed pastures (n=24) within the *ServiceGrass* project in the Canton of Solothurn (Switzerland) were included in this study. The grassland sites belong to 18 organic and 18 non-organic farms, with one organic farm always in close vicinity to a non-organic farm, resulting in a spatially balanced design. In summer 2021, soil cores were taken to 20 cm depth (20 cores pooled per grassland) and analysed for soil phosphorus (P) concentrations (Olsen extraction). Interviews were conducted with the farmers to gather information about fertilization practices. Utilizable nitrogen (N) fertilization was calculated from this information according to Swiss regulations (Richner *et al.*, 2017). Aboveground biomass was sampled between mid-May and mid-June 2021, with four pooled samples of an area of 50×50 cm per site, dried and subsequently weighed. Forage increment per day was calculated as biomass (g) per growing day (days since 1 March) per m^2 (hereafter *forage increase*). Feed value was calculated as an indicator value (Briemle and Dierschke, 2002), using mean species cover from two 2×2 m vegetation relevés per site. *t*-tests were conducted to identify differences among organic and conventional grasslands in soil P, N fertilization, forage increase and feed value. To analyse the effect of organic management on forage increase and feed value via changes in soil P and N fertilization, a structural equation model (SEM) was computed with the R (R Core Team, 2021) package lavaan (Rosseel, 2012). The full model was specified as shown in Figure 1A, first without and then with a multigroup comparison of pastures vs meadows. These two models were compared using the Akaike information criterion (AIC) and sample-size adjusted Bayesian information criterion (BIC). Subsequentially, non-significant pathways were sequentially deleted from the model to achieve a most robust final SEM.

Results and discussion

Over all plots, forage increase tended to be 15% smaller in organic compared to non-organic managed grasslands (mean=3.9 vs 4.7; P>0.1, t-test), as was feed value (mean=6.6 vs 7.1, P=0.033, t-test). The AICc for the SEM including a group comparison of pasture and meadow (Figure 1B) was lower than for the SEM without the group comparison (AIC=527 vs 533, BIC=498 vs 522), indicating that the responses of forage increase and feed value to the environmental, fertilizer and management variables studied here differed between meadows and pastures.

For *pastures* (predominantly grazed), the SEM showed a marginally significant negative effect of organic management on soil P (standardized coefficient -0.33; P=0.062; Figure 1B). The mean soil P differed quite strongly, with 22 mg kg^{-1} in organic and 41 mg kg^{-1} in non-organically managed pastures (P=0.049; t-test). Soil P was additionally influenced by N fertilization (stand. coeff. 0.38; P=0.029). However, N fertilization itself was not influenced by organic management (mean=46.3 vs 79.8 kg N ha^{-1} organic vs non-organic, P>0.1, t-test). This lack of an effect of organic management on N fertilization compared to the direct effect of organic management on soil P could be due to soil P showing the effect of fertilizing events from past years, whereas N fertilization merely reflects fertilization in 2020, the year of the farmer interviews. Soil P in turn positively influenced forage increase (stand. coeff. 0.54, P=0.002) and feed value (stand. coeff. 0.74, P<0.001), leading to 45% lower mean forage increase in organic (2.7 g day^{-1}) than in non-organic (4.1 g day^{-1}; P=0.073, t-test) and mean feed value (6.4 vs 7.1; P=0.055, t-test). Thus, in pastures, organic management influenced forage increase and feed value via lower soil P.

In *meadows* (predominantly cut), the SEM showed no evidence for organic management influencing either soil P or N fertilization (Figure 1B). Indeed, organic and non-organic meadows did not differ strongly in N fertilization (mean=86.7 vs 107.5 kg N ha^{-1}; P>0.1, t-test) and soil P (mean=32.2 vs 40.7 mg kg^{-1}; P>0.1, t-test). As a consequence, no evidence for any effect of organic management on forage increase and feed value was detected in the SEM (Figure 1B) and means for forage increase (5.1 and 5.2 g day^{-1}, respectively) and feed value (6.8 and 7.1) were quite comparable between organic and non-organic meadows (both P>0.1, t-tests). N fertilization in meadows, in contrast to pastures, did not influence soil P, but both factors had direct positive effects on forage increase and feed value (Figure 1B).

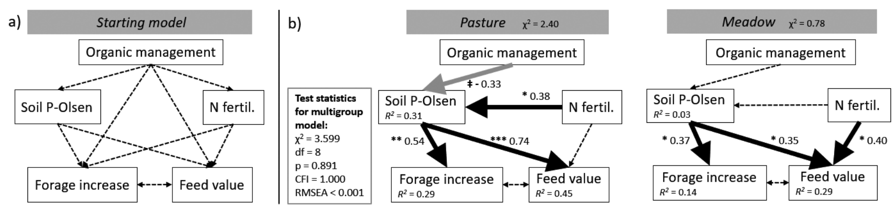

Figure 1. (A) full starting model for structural equation modelling (SEM); and (B) final multigroup SEMs comparing the influence of organic management on forage increase (g day^{-1}) and feed value in pastures vs meadows with statistics for the overall multigroup model on the left of the path-models. Dashed lines indicate non-significant pathways. For significant pathways, standardized coefficients are displayed next to the arrows, grey solid arrows indicating negative, black solid arrows positive effects. Significance levels of the coefficients (*P-value*): 0.1 ≥ '‡' ≥ 0.05 ≥ '*' ≥ 0.01 ≥ '**' ≥ 0.001 ≥ '***'.

The yield reductions in organically managed grasslands found in other studies (Mäder *et al.*, 2002; Oberson *et al.*, 2013; Steinwender *et al.*, 2000) fits to our overall results averaged over pastures and meadows. Interestingly, we find different responses of the two management types, with meadows showing no reduction and pastures a strong reduction in forage increase and forage yield. The meadow response is similar to findings of Klaus *et al.* (2013) regarding no significant yield differences in organic vs non-organic grasslands in Germany. Our results suggest a necessity to differentiate between predominantly grazed pastures and predominantly cut meadows when assessing the interrelated drivers of provisioning ES. This will also help to investigate the reasons for lower soil P in organic pastures.

Conclusions

Our findings suggest that in the studied region forage increase and feed value were not compromised in organic compared to non-organic intensively managed meadows. However, in intensively managed pastures, lower forage yield and quality were related to differences in soil P due to organic management. Further research investigating the reasons for lower soil P is necessary to understand and resolve this issue and to close the yield gap in organic pastures, and to understand why meadows differed in their response.

Acknowledgements

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References

Briemle G. and Dierschke H. (2002) *Kulturgrasland*. Ulmer, Stuttgart, Germany.

Klaus V. H., Kleinebecker T., Prati D., Gossner M. M., Alt F., Boch S., … and Hölzel N. (2013) Does organic grassland farming benefit plant and arthropod diversity at the expense of yield and soil fertility? *Agriculture, Ecosystems and Environment* 177, 1-9.

Knudsen M.T., Dorca-Preda T., Djomo S. N., Peña N., Padel S., Smith L. G., … and Hermansen J.E. (2019) The importance of including soil carbon changes, ecotoxicity and biodiversity impacts in environmental life cycle assessments of organic and conventional milk in Western Europe. *Journal of Cleaner Production* 215, 433-443.

Mäder P., Fließbach A., Dubois D., Gunst L., Fried P. and Niggli U. (2002) Soil fertility and biodiversity in organic farming. *Science* 296, 1694-1697.

Oberson A., Frossard E., Bühlmann C., Mayer J., Mäder P. and Lüscher A. (2013) Nitrogen fixation and transfer in grass-clover leys under organic and conventional cropping systems. *Plant and Soil*, 371, 237-255.

R Core Team (2021) R: a language and environment for statistical computing. R foundation for Statistical Computing, Vienna, Austria.

Richner W., Flisch R., Mayer J. and Schlegel P. (2017) *4/Eigenschaften und Anwendung von Düngern*.

Rosseel Y. (2012) lavaan: An R Package for Structural Equation Modeling. *Journal of Statistical Software* 48, 1-36.

Steinwender R., Gruber L., Schauer A., Guggneberger T., Häusler J. and Sobotik M. (2000) Vergleich zwischen biologischer und konventioneller Wirtschaftsweise im Grünlandbetrieb. *Die Bodenkultur* 52, 267-281.

Swiss Federal Council (2018). Verordnung über die biologische Landwirtschaft und die Kennzeichnung biologisch produzierter Erzeugnisse und Lebensmittel vom 22. September 1997.

Net climate impacts of sustainable intensification measures in Boreal crop-livestock system

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Abstract

Sustainable intensification (SI) in agriculture aims to increase crop yields and animal production without negative environmental effects and use of additional land area. In this study we estimated the impacts of potential SI-options in cropping and animal husbandry on carbon (C) inputs into soil, soil organic carbon (SOC) stocks and total greenhouse gas (GHG) emissions in Boreal crop-livestock farming system. The examined options included increasing crop yields and decreasing grain proportion in the diet of dairy cows. We used agricultural statistics and literature to calculate C inputs, the Yasso07 soil model to estimate changes in SOC stocks, and Carbon Calculator to estimate total GHG emissions. According to our results, animal processes produced the highest share of GHG emissions. Emissions from the SOC stock changes originated mainly from cultivation of organic soils. Results indicate that SI-options that allow for reductions in land area needed for feed production have significant potential to reduce GHG emissions when the reductions are allocated to organic soils.

Keywords: C footprint, farming practices, greenhouse gas emissions, dairy production, life cycle assessment

Introduction

Intensification in agriculture is expected to achieve food security for the growing population. Sustainable intensification (SI) is needed when intending to reduce the negative impacts on environment (Thomson *et al.*, 2019; Garnett *et al.*, 2013). In the northern cropping systems, key SI-options are improved crop rotations, liming and drainage, sowing techniques, novel high-yielding and robust cultivars, and forage grass mixtures which aim to improve the soil growing conditions and have the potential to increase yields (Lehtonen *et al.*, 2018). In addition to this, changing the diet of cattle to include a high forage proportion would intensify the field use, as the potential yield of grass is two-fold compared with grain in Finland. The smaller fodder production area would allow for other uses for the arable land released from the fodder production. Implementing such SI-options could thus increase C inputs to soil and increase SOC stocks.

The aim of this study was to assess the net climate impacts of different SI-options and their realistic combinations in boreal crop-livestock farming systems. The examined SI-options were: (1) yield increases achieved through improved soil growing conditions, and (2) decrease grain proportion in the diet of dairy cow. These were compared with a baseline representing current typical management practices on Finnish dairy production system.

Materials and methods

We analysed the effects of SI-options at a dairy farm in the Northern Savonia region in Finland, representing typical farm and land use in the region. There were 140 dairy cows on the farm. In the baseline scenario, the concentrate proportion was 45%, and the total cultivated area required for fodder production was 225 ha, of which 125 ha was allocated to grasslands, 50 ha to oats, and 50 ha to barley.

Of the total production area, 80% was on mineral soils and 20% on organic soils, allocated equally to different crops. In the baseline, the crop yields for silage, oats and barley were 7,000 kg, 3,400 kg, and 3,400 kg dry weight per ha, respectively. Energy-corrected milk production was 9,697 kg per year. In the SI-scenarios, the assessed yield increases were +10% and +20% for all cultivated crops, with increases attributed to investments in improved soil growing conditions, for the two diet alternatives. For simulations, the system boundary was set to the 'farm gate' in all cases. We assessed and compared net impacts of different SI-options on (1) required cultivation areas of different crops, (2) C inputs into soils, and (3) total GHG emissions on farm and at product level. For all cases, the dry matter intake was fixed, and the milk production yield was constant over the scenarios. As a result, the implementation of the scenarios allowed the area of feed production to be reduced. In S1, S2 and S3 the examined production area was equal to baseline and the area released through implementing SI-options was allocated to production of green fallow. In S4, S5 and S6 the area released through implementing SI-options was allocated away from the production in organic soils, which reduced the total production area (Table 1).

For the assessment, we used agricultural statistics and literature to calculate C inputs, the Yasso07 soil model (Tuomi *et al.*, 2011) to estimate SOC stock changes of mineral soils, and Carbon Calculator (Tuomisto *et al.*, 2013) to estimate total GHG emissions. The effects of implementing SI-options on total emissions were assessed in comparison with the baseline. Time spans of 20 years and 100 years were considered.

Results and discussion

The total farm-level GHG emissions, including changes in the SOC stock, varied from 3,225 Mg CO_2-eq. yr^{-1} to 3575 Mg CO_2-eq. yr^{-1} in the scenarios where the considered production area was similar to baseline (Figure 1). Animal processes, including enteric fermentation and manure management, produced the highest GHG emissions. Most of the emissions from the SOC stock change originated from cultivation of organic soils. The land area released from fodder production due to changes in the cattle diet and yield increase was highest under the S6-scenario, at 56 ha (25% of the total production area). Releasing this area from production on organic soils would replace emissions with a SOC change, from 1,102 Mg CO_2 yr^{-1} (emission in the baseline) to -26 Mg CO_2 yr^{-1} (a C sink), and thus reduce total emissions considerably (Figure 1). The utilized assessment model (Carbon Calculator), however, was unable to incorporate the impacts of selected diets on enteric fermentation. Also, the fixed values for milk production yield and dry matter intake over the different diets are rather unlikely, and might add uncertainties into the results. Accordingly, the linkage between diet composition, emissions from animals and carbon stock changes need further investigations.

Table 1. Characteristics of cattle diet, crop yields and total production area in scenarios.

Scenario	Share of concentrated feed in the diet (%)	Yields	Production area (ha)
Baseline (BL)	45	Current	225
S1	45	+20 %	225
S2	30	Current	225
S3	30	+20 %	225
S4	45	+20 %	188
S5	30	+10 %	184
S6	30	+20 %	169

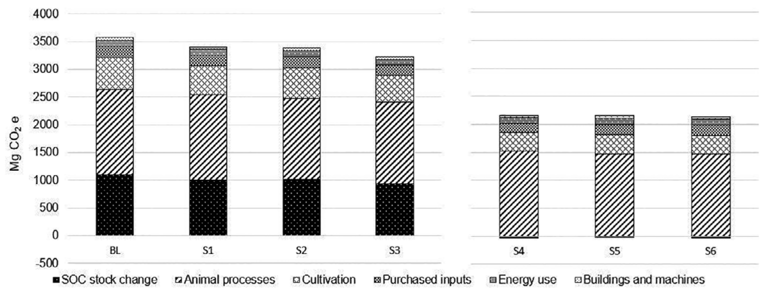

Figure 1. Greenhouse gas emission sources from dairy farm in baseline (BL) and scenarios S1-S6.

Conclusions

The results of the study clearly show the importance of organic soils as a source of GHG emissions for boreal crop-livestock farming systems. SI-options that allow for reductions in land area needed for feed production have significant potential to reduce GHG emissions when the reductions are allocated to organic soils. This way the efforts made to improve the production also serve the targets of climate change mitigation.

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References

Garnett T., Appleby M.C., Balmford A., Bateman I.J., Benton T.G., Bloomer P., Burlingame B., Dawkins M., Dolan L. and Fraser D. (2013) Sustainable intensification in agriculture: premises and policies. *Science Magazine* 341, 33-34.

Lehtonen H., Palosuo T., Korhonen P. and Liu X. (2018) Higher crop yield levels in the North Savo Region – means and challenges indicated by farmers and their close stakeholders. *Agriculture* 8, 93.

Thomson A.M., Ellis E.C., Grau H.R., Kuemmerle T., Meyfroidt P., Ramankutty N., Zeleke G. (2019) Sustainable intensification in land systems: trade-offs, scales, and contexts. *Current Opinion in Environmental Sustainability* 38, 37-43.

Tuomi M., Rasinmäki J., Repo A., Vanhala P. and Liski J. (2011) Soil carbon model Yasso07 graphical user interface. *Environmental Modelling & Software* 26, 1358-1362.

Tuomisto H., De Camillis C., Leip A., Nisini L., Pelletier N. and Haastrup P. (2015) Development and testing of a European Union-wide farm-level carbon calculator. *Integrated Environmental Assessment and Management* 11, 404-416.

Legume biomasses produce high protein yields in a green biorefinery concept

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Abstract

Sustainable sources of protein are in great need for both food and feed use. The protein in forage legumes is bound to a fibre matrix and not efficiently used by pigs, poultry or directly by humans. Legumes can produce high yields of protein per hectare as well as many ecosystem services. The green biorefinery concept can be used to separate the protein from the plant biomass to be used in novel applications. In this study, red clover, white clover, goat's rue, faba bean and pea were harvested as green crops and mechanically separated into liquid and solid fractions. Various pretreatments were used to mimic the potential practical large-scale processes in which the fresh green plants were compared with freezing-and-melting, and drying-and-rehydrating in terms of yields. The liquid yields ranged from 69 to 79% using an efficient twin-screw press. On average 54% of crude protein was extracted into the liquid, and the liquid crude protein concentration averaged 291 g kg^{-1} dry matter. Pneumatic press, on the other hand, was less efficient since the liquid yield was substantially lower, but clearly increased after pre-treatments due to the breakage of plant cells.

Keywords: protein recovery, twin screw press, pneumatic press, liquid-solid separation, forage legume, grain legume

Introduction

Sustainable sources of protein are in great need for both food and feed use. The protein in green biomass is bound to a fibre matrix and therefore not efficiently used by pigs, poultry or directly by humans, thus being currently mostly used as feed for ruminants. Forage and grain legumes harvested at an immature growth stage provide potential raw material for further processing. The green biorefinery concept can be used to separate the protein from green biomass to be used in novel applications for feed use, both for ruminants and monogastrics (Keto *et al.*, 2021), or even directly for humans. Forage legumes can produce high yields of protein per hectare as well as many ecosystem services. Finding new ways to utilize them in the food chain requires basic knowledge on their yields, safety (secondary metabolites, bioactivities, pesticide use, hygienic quality), suitability regarding food technology and acceptance by consumers as well as regulatory approvals. Of the materials used here, only pea sprouts are currently used for human consumption. The aim of the current evaluation was to screen various legume species for their suitability as feedstocks for green biorefineries.

Materials and methods

Green biomass of red clover (*Trifolium pratense*, var. Selma), white clover (*Trifolium repens*, var. Lena), goat's rue (*Galega orientalis,* var. Gale), pea (*Pisum sativum*, var. Hulda) and faba bean (*Vicia faba*, var. Kontu) were harvested from primary growth in year 2021 in Jokioinen, Finland (60°48′N 23°29′E). Additionally, red clover (var. Saija) was also harvested from regrowth in Siikajoki, Finland (64°40′N 25°06′E). Raw material samples were analysed for chemical composition using routine analytical methods of the Luke laboratory (Savonen *et al.* 2020). The biomass of each species was separated into liquid and solid fractions using a twin-screw press (300 g batch) or a pneumatic press (100 g batch; for details see Franco *et al.*, 2019). The biomass samples were processed (1) fresh immediately after harvesting from the field, (2) frozen and melted, and (3) dried and rehydrated into the original dry matter (DM) concentration using tap water in order to evaluate the effect of pretreatments on the efficiency of extraction. For practical reasons, treatments were not applied to all plant materials (e.g. no liquid was produced from fresh faba bean and pea using the pneumatic press). Experimental replication was not included in this screening study, so no statistical evaluation was conducted.

Results and discussion

The legume biomasses used in the current study are described in Table 1. The aim was to use early maturity stage of the plants with relatively high moisture and crude protein (CP) concentrations to facilitate the mechanical separation of proteins. However, the maturity stages and varieties were not totally optimized (e.g. rather late maturity stage of pea, and both grain legumes being varieties intended for seed production). There is indeed large variation in plant biomass composition, and various agronomic and management factors in biomass production have a lot of scope for optimization. The composition of the liquids recovered after pressing with a twin-screw press showed high variability which originated from the differences in raw materials (Table 1). Goat's rue had the highest CP concentration both in raw material and in the extracted liquid, while pea had the lowest concentrations of both, which may have been affected by the late maturity stage of pea at harvest. A meta-analysis conducted in Finland using grass silages as raw materials resulted in liquid DM and CP concentrations of 103 g kg^{-1} and 231 g kg^{-1} DM (Franco *et al.* 2019). The DM concentration is in good agreement with the current results (average 96 g kg^{-1}), but liquid CP concentration was clearly higher in the liquids originating from these legumes (on average 291 g kg^{-1} DM) due to the higher CP concentration of the raw materials used in the current data set.

The high variability in ash yields (e.g. over 100% ash recovery for pea) is probably associated with the inaccuracies in measuring and analysing the small volumes used. The use of the efficient press demonstrates the high potential to extract soluble nutrients from green biomass. If we assume a biomass yield of 10,000 kg DM ha^{-1}, a CP concentration of 200 g kg^{-1} DM and a protein extraction rate of 50% (average value for the current data set was 54%), the amount of CP in the harvested liquid could be 1000 kg ha^{-1}, which is comparable to the yields of protein crops under northern European environmental conditions. The remaining fractions from the green biorefinery may be used for numerous purposes such as feeds for ruminants, biogas, soil amendment, bedding or other materials.

Table 1. Composition of the raw material and liquids of the evaluated legume biomasses and the extraction rates using twin screw press.

	Red clover, 1st cut	Red clover, 2nd cut	White clover	Goat's rue	Pea	Faba bean
Date of harvest in year 2021	16 June	18 August	16 June	9 June	6 August	13 August
Raw materials						
Dry matter (DM), g kg^{-1}	171	121	171	128	278	188
Ash (g kg^{-1} DM)	110	106	113	83	49	62
Crude protein (g kg^{-1} DM)	193	199	224	270	131	234
Liquids						
DM, g kg^{-1}	110	47	100	62	161	98
Ash (g kg^{-1} DM)	150	100	150	62	121	96
Crude protein (g kg^{-1} DM)	272	301	262	442	195	274
Extraction rates (%) of fresh legume biomass						
Liquid	69	79	70	74	71	72
DM	44	31	41	35	41	37
Ash	61	27	41	27	102	57
Crude protein	63	43	55	58	61	44

Figure 1 shows results of pre-treatments and type of liquid separation method on liquid yields of red clover and goat's rue. The pneumatic press was very inefficient when fresh biomass was fractioned, but significant improvements were achieved in the freezing-and-melting and the drying-and-rewetting methods. The increase can be attributed to the breakage of cell walls during these processes. It is notable that the frozen samples did not perform as well in crude protein recovery as they did in liquid extraction. This was caused by lower CP concentration in the liquid, and this effect was consistent between the two types of presses.

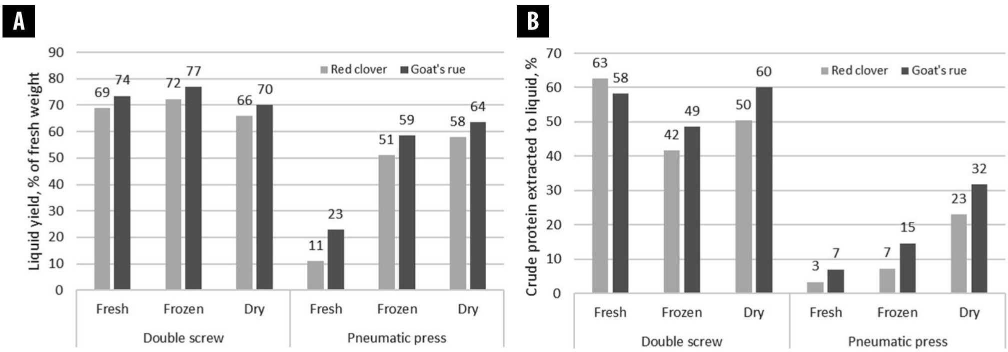

Figure 1. Effect of pre-treatments (frozen and melted; dried and rehydrated (dry)) on the liquid yields (A) and percentage of original crude protein extracted into the liquid (B) of red clover (1st cut) and goat's rue using two different methods for liquid separation when compared to the freshly processed biomass.

Conclusions

Various species of legume biomass have potential to produce large protein yields per area cultivated. The pre-treatments used here, i.e. freezing-and-melting, and drying-and-rewetting, increased the liquid yields compared to fresh material, particularly when the less efficient pneumatic press was used. This can be considered beneficial, because preserving the biomass allows the green biorefinery to operate year-round, and preservation of the produced fractions can be done in smaller batches. Further optimization of biomass production including aspects such as varieties within species, fertilization and harvesting regime would be needed, and this can be supported by previous agronomic knowledge. Further, the possibilities to include green biomass-based proteins into food products for direct human use should be evaluated.

References

Franco M., Hurme T., Winquist E. and Rinne M. (2019) Grass silage for biorefinery – A meta-analysis of silage factors affecting liquid-solid separation. *Grass and Forage Science* 74: 218-230.

Keto L., Perttilä S., Särkijärvi S., Immonen N., Kytölä K., Alakomi H.-L., Hyytiäinen-Pabst T., Tsitko I., Saarela M. and Rinne M. (2021) Effect of silage juice feeding on pig production performance, meat quality and gut microbiome. *Livestock Science* 254: 104728.

Savonen O., Franco M., Stefanski T., Mäntysaari P., Kuoppala K. and Rinne M. (2020) Grass silage pulp as a dietary component for high yielding dairy cows. *Animal* 14, 1472-1480.

Benefits of adaptative multi-paddock grazing – implementation in French livestock production systems

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Abstract

The beef industry and livestock production systems are at the centre of debates questioning GHG emissions and the environment more generally. To improve sustainability and to highlight the contributions of the beef sector has thus become a national and international goal of involved actors. Along with other solutions, grazing management techniques, such as adaptive multi-paddock grazing (AMP), have been suggested to provide services related to production, carbon sequestration, biodiversity, landscape, farmers' workloads and many other aspects at the farm level. Though, a number of studies have compared different grazing systems around the world (e.g. rotational vs continuous), only few have performed integral multi-criteria analyses. Hence, there is urgent need to assess advantages and disadvantages of climate-smart grazing practices based on common and comparable indicators (e.g. ecosystem services). In the present study, we will compare three different grazing systems including AMP, implemented in 27 commercial farms on three pedo-climatic regions. For each grazing system we examine grassland production (quantity and quality), carbon sequestration, biodiversity, and technical-economic results. This work will provide relevant information for grazing management aiming to achieve desired environmental and economic goals, and will put forward interesting grazing systems to meet multiple challenges (services).

Keywords: cow, grazing system, services, carbon sequestration

Introduction

Grassland systems have strong assets to meet societal expectations related to livestock (Michaud *et al.*, 2020). Indeed, the presence of sustainable vegetal cover over time, and grazing as the basis of feeding system, are suggested to be beneficial for beef cattle, its well-being, farm economics and environment (Mann and Sherren, 2018). There are several grazing systems that are an integrated combination of animal, plant, soil, economic and social features, and management objectives designed to achieve specific goals or results (Sollenberger *et al.*, 2020). Among those combinations, adaptive multi-paddock grazing (AMP) has been proposed to provide services such as equivalent production as continuous or slow rotational grazing, better carbon sequestration, better biodiversity, improvement of the landscape organization and many other aspects on the farm (Mosier *et al* 2021; Shrestha *et al.*, 2020; Teague and Barnes 2017; Teague *et al.*, 2013). AMP grazing uses high livestock densities for short durations between long periods of forage rest to stimulate accelerated grass growth. Thus, AMP grazing is characterized by rotational grazing of multiple paddocks, adapting animal loading according to herbage growth, starting with late grazing season at a vegetation height of 12 cm or more (elongation stage), and moving animals to paddocks that have not been assigned a rest period when half of initial herbage mass is consumed (Teague *et al.*, 2011). The herd is moved back when the paddock has had sufficient time for defoliated plants to fully recover. This approach leaves adequate residual plant biomass in grazed paddocks to maintain high growth rates for plants and high forage quality for animals. The rest period depends on soil-climate conditions and regrowth and can vary from 40 to 180 days between grazing events. The aim of this project is to analyse the effects of different grazing systems on carbon sequestration, plant biodiversity and grassland production and quality, as well as the sustainability of production systems. In the context of French beef production systems, this study will deliver intelligent, goal-directed key data on practices to valorise and optimize grazing management to achieve sustainable goals. The results will provide a better understanding on biophysical processes, and how to adjust them to answer to the challenges of production systems in terms of climate adaptation and societal expectations.

Materials and methods

In order to assess the effect of major French grazing systems on the provision of services, literature analyses were carried out using national census data on livestock production systems, including grey and peer reviewed literature. In more detail, literature was screened to identify key elements such as the definition of French grazing practices, animal stocking rates, grazing duration, paddock number, vegetation height at grazing and fertilization. As for associated services, the literature research aimed to identify measurable indicators (i.e. services and methods used) with regard to livestock farming systems, being a compromise between ecological, agricultural and territory approaches (Dernat *et al.*, 2020; Ryschawy *et al.*, 2015). Collected information was used to setup an AMP monitoring project.

Results and discussion

In brief, state-of-the-art literature analyses allowed to highlight (1) major grazing categories and markers to characterize them (Table 1) and (2) identify measurable service bundles permitting to compare different grazing systems. According to the analyses of French grazing systems (Table 1), a number of choices have been made to setup the AMP-monitoring project. For instance, study area; the project will be implemented in three contrasted pedo-climatic regions in France: Normandy, East of France and central France. In each area, three commercial farms have been chosen for implementing AMP grazing and allowing a comparison with a neighbouring practice; continuous grazing (GC), rotational grazing (RG). In total, 27 farms will be monitored for 3 years. Each farm will be examined at several scale levels:

- *Animal scale*. A batch of heifers will be monitored on each farm throughout the project, grazing on studied grasslands. Animal feeding, health and well-being of the batch will be followed, using operational tools.
- *Paddock scale.* Fields dedicated to AMP, CC and RG grazing will be evaluated over the grazing season concerning their botanical composition, biomass production, and nutritive value. Further, each of the followed grass fields will be analysed twice (at the beginning and the end of the project, T0 and T36) for soil organic stocks (0-60 cm), pH and soil minerals (N, P, K). For soil analyses, a 'control' on each farm will be used.
- *System scale.* For each farm, technical-economic surveys will be carried out. Likewise, feed intake of the herd will be defined and quantified by the HerbValo method (Delagarde *et al.*, 2018), and farmers additional workload related to AMP grazing management will be quantified.

Table 1. Main grazing practices in France (literature analyses and agricultural census data).

Grazing systems	Continuous (Leray *et al.*, 2017)	Rotational 'Slow' (Leray *et al.*, 2017)	AMP US (Mosier *et al.*, 2021)
Meadow characteristics	permanent/natural/temporary	permanent/temporary	NAN
Instantaneous livestock density (LU ha^{-1})	Lower than 0.5 LU ha^{-1} to 1.8 LU ha^{-1}	Depends on the type of rotation: 10-50	60-460
Global surface (are LU^{-1})	30-80	25-60	0.03-0.1
Grazing duration per paddock (days)	90-200	3-mei	1-2
Resting period (days)	No resting time (winter)	20-40	45-90
Number of paddocks available	1-3	5-15	5-50
Entry mark; vegetation height cm)	7 to 20	8-15	15 or more
Exit mark; vegetation height (cm)	the animals stay on the same paddock throughout the year	3-6	10 (50% of the entrance biomass)
Fertilization N (Kg ha^{-1})	0-200	0-150	0-90; ideally zero
Plant development stage (entry)	vegetative stage of grass sward	vegetative stage of grass sward	elongation or more

References

Delagarde R., Robic Y., Leurent-Colette S. and Delaby L. (2018) HerbValo – a method for calculating annual pasture utilisation by dairy cows at paddock level. *Grassland Science in Europe* 23, 968-970.

Dernat S. (2020) *Un modèle basé sur les systèmes socio-écologiques*. INRAe. Available at: https://www6.inrae.fr/la-grange/Le-modele-La-Grange/Un-modele-base-sur-les-systemes-socio-ecologiques

Mann C. and Sherren K. (2018) Holistic management and adaptive grazing: a trainers' view. *Sustainability* 10(6), 1848.

Leray O., Doligez P., Jost J., Pottier E. and Delaby L. (2017) Présentation des différentes techniques de pâturage selon les espèces herbivores utilisatrices. *Fourrages* 229, 11-16.

Michaud A., Plantureux S., Baumont R. and Delaby L. (2020) Grasslands, a source of richness and support for innovation for more sustainable and acceptable ruminant livestock farming. *INRAE Productions Animales* 33(3), 153-172.

Mosier S., Apfelbaum S., Byck P., Calderon F., Teague R., Thompson R. and Cotrufo M.F. (2021) Adaptive multi-paddock grazing enhances soil carbon and nitrogen stocks and stabilization through mineral association in southeastern US grazing lands. *Journal of Environmental Management* 288, 112409.

Ryschawy J., Tichit M., Bertrand S., Allaire G., Plantureux S., Aznar O., ... and Disenhaus C. (2015). Comment évaluer les services rendus par l'élevage? Une première approche méthodologique sur le cas de la France. *Productions animales* 28(1), 23-38.

Shrestha B M., Bork E.W., Chang S X., Carlyle C.N., Ma Z., Döbert T.F., ... and Boyce M.S. (2020) Adaptive multi-paddock grazing lowers soil greenhouse gas emission potential by altering extracellular enzyme activity. *Agronomy* 10 (11), 1781.

Sollenberger L.E., Aiken G.E. and Wallau M.O. (2020) Managing grazing in forage-livestock systems. In: *Management Strategies for Sustainable Cattle Production in Southern Pastures*. Academic Press, Cambridge, MA, USA, pp. 77-100.

Teague W.R., Dowhower S.L., Baker S.A., Haile N., DeLaune P.B. and Conover D.M. (2011) Grazing management impacts on vegetation, soil biota and soil chemical, physical and hydrological properties in tall grass prairie. *Agriculture, Ecosystems & Environment* 141(3-4), 310-322.

Teague R., Provenza F., Kreuter U., Steffens T. and Barnes M. (2013) Multi-paddock grazing on rangelands: why the perceptual dichotomy between research results and rancher experience? *Journal of Environmental Management* 128, 699-717.

Teague R. and Barnes M. (2017) Grazing management that regenerates ecosystem function and grazingland livelihoods. *African Journal of Range & Forage Science* 34(2), 77-86.

Delivery of ecosystem services from permanent grasslands in Europe: a systematic review

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Abstract

Permanent grasslands cover 34% of the European Union's agricultural area and are vital for the delivery of essential ecosystem services. Over recent decades, permanent grasslands have suffered a significant decline and land use change continues to threaten their area. We performed a systematic review on the multifunctionality of permanent grasslands in Europe, examining the effects of land use change and management practices on 18 ecosystem service indicators. Based on the evidence in 696 out of 70,456 screened papers, we found that both land use change and intensification decreased multifunctionality. A lower management intensity was associated with benefits for biodiversity, climate regulation and water purification, but had a negative effect on the provision of high-quality animal feed. Increasing the number of species in the sward enhanced multifunctionality of permanent grassland without significant trade-offs such as losses in production. We suggest that a combined approach of protection and management extensification will help secure multiple benefits from permanent grasslands.

Keywords: agro-ecology, land use change, management, multifunctionality

Introduction

Permanent grasslands cover 34% of the European Union's agricultural area and are vital for human wellbeing as they contribute to a wide variety of essential ecosystem services. For centuries, permanent grasslands have been the basis for livestock production on farms all over Europe. However, over the past decades, permanent grasslands have suffered a significant decline and land use change continues to threaten their area (Schils *et al.*, 2020). In addition to the provision of feed, permanent grasslands sustain a broad range of additional ecosystem services, including climate regulation through carbon sequestration, preservation of biodiversity and cultural values, protection against erosion and flooding, and pollination of food crops (e.g. Bengtsson *et al.*, 2019). For European permanent grasslands we have a restricted understanding of land use and management effects on multifunctionality. Here, we analyse the body of, mainly monodisciplinary, studies across Europe in a comprehensive multidisciplinary systematic literature review with a focus on experimental contrasts in land use and management aspects. Our study addressed two central research questions: first, what are the reported effects of land use change on the delivery of ecosystem services? Second, what are the reported effects of intensification and specific management options on the delivery of ecosystem services by permanent grassland?

Materials and methods

We systematically searched the scientific literature for grassland studies on 18 indicators of ecosystem services in Europe, published in the English language between 1980 and 2019. The indicators were pollinators, threatened species, and plant richness for *biodiversity*; nitrous oxide, methane and carbon dioxide emissions, and carbon sequestration for *climate regulation*; nitrogen, phosphorus in groundwater and surface water for *provision of fresh water*; recreational value and aesthetics for *cultural values*; hydraulic conductivity, bulk density, soil loss and runoff for *erosion and flood control;* energy and protein content, and forage yield for *provision of animal feed.* In our study, permanent grasslands are defined as land used to grow grasses or other herbaceous forages that has not been included in a crop rotation for a duration of five years or longer.

We screened 70,456 papers and retained 696 papers that contained at least one of eight experimental contrasts, either in land use (permanent grassland versus cropland, forest or temporary grassland) or in management options (sward renewal, defoliation frequency, nitrogen input, legume presence, and number of sward species). The 696 papers contained 1,032 eligible experimental contrasts. For each contrast, we registered the outcome, i.e. the effect of the contrast on the value of the ecosystem service indicator: no conclusion, favourable, neutral, unfavourable. For the analysis, outcomes were transformed to numerical values (favourable=1, neutral=0, unfavourable=-1). More details are presented in Schils *et al.* (2022).

Results

Most of the extracted papers included in this review were identified in regions where over 40% of the utilized agricultural area was covered by permanent grasslands. Around two thirds of the extracted papers originated from the Atlantic or Continental biogeographic regions.

We found consistent trade-offs in the reported outcomes between indicators for feed on the one hand, and non-feed ecosystem services on the other, for three management intensity indicators, i.e. nitrogen input, increasing defoliation frequency and grass renewal (Figure 1). The reported outcomes of increased number of species in the sward showed mainly favourable effects on the indicators for *biodiversity, cultural values* and *water purification* and mixed effects on *provision of animal feed* (not shown). Grass renewal showed significant favourable effects on forage yield, but no consistent effect on forage quality. In contrast, we found that grassland renewal significantly increased nitrous oxide emissions and nitrogen losses to water. Considering land use change, we found that most studies reported favourable outcomes for maintaining permanent grasslands compared to conversion to croplands across all ecosystem service indicators, apart from forage yield and energy content (not shown).

Discussion and conclusions

The outcomes of our review suggest that, in spite of apparent changes in human dietary preferences, the protection of permanent grasslands in Europe has to be prioritized to prevent further losses of the area and their ecosystem services. At the same time, in view of the need to reduce ruminant livestock's impact on climate change and the apparent benefits of lower management intensity on biodiversity and water quality, the time seems ripe to increase support for a reduced management intensity on existing European permanent grasslands.

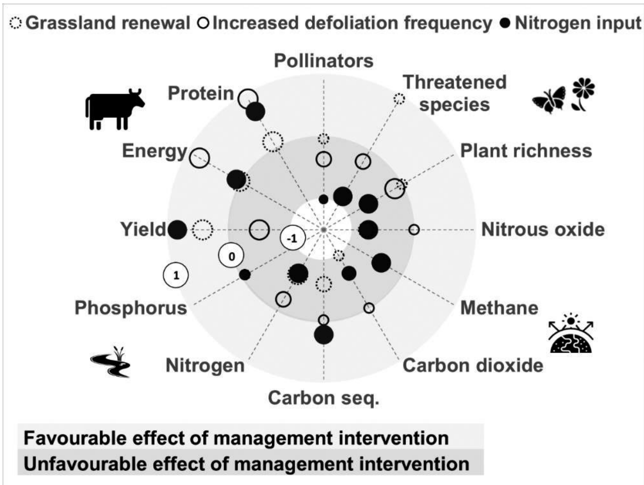

Figure 1. Effects of management intervention on indicators for ecosystem services. The boundary between the shaded zones represents a mean score of 0. Light shaded outer zone represents a favourable score for permanent grassland (moving outwards, the mean score increases from 0 to 1), darker shaded inner zone represents an unfavourable score (moving inwards, the mean score decreases from 0 to -1). Dot size indicates number of underlying cases (small: <5 cases, medium: 5-9 cases. Large: >9 cases).

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References

Bengtsson J., Bullock J.M., Egoh B., Everson C., Everson T., O'Connor, ... and Lindborg R. (2019) Grasslands – more important for ecosystem services than you might think. *Ecosphere* 10, 1-20.

Schils R.L.M., Newell Price P., Klaus V.H., Tonn B., Hejduk S., Stypinski P. ... and Korevaar H. (2020) European permanent grasslands mainly threatened by abandonment, heat and drought, and conversion to temporary grassland. *Grassland Science in Europe* 25, 553-555.

Schils R.L.M., Bufe C., Rhymer, C.M., Francksen R.M., Klaus V.H, Abdalla M.... and Newell Price J.P. (2022) Permanent grasslands in Europe: land use change and intensification decrease their multifunctionality. *Agriculture Ecosystems and Environment* 330, 107891.

Ecosystem services of pre-Alpine grasslands – the effects of climate change and management

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Abstract

Managed grasslands are an important part of the pre-Alpine landscape. Besides feed provision, they fulfil important services like carbon and nitrogen storage, water retention, and habitats for wildlife. Changing climatic conditions and different management systems may affect, e.g. water fluxes, biogeochemical processes and species composition, and hence ecosystem services of grasslands. This study analysed the effects of climate change and management on dry matter yields, soil organic carbon and N storage, and greenhouse gas emissions in pre-Alpine grasslands by combining experimental and modelling studies. We utilized data of the German TERENO pre-Alpine observatory from 2012-2020. Large soil monoliths (lysimeters) were translocated down an elevation gradient to simulate climate change and are operated with two management intensities (differing in cuts and fertilization). Data analyses were accompanied by simulation studies with the biogeochemical model LandscapeDNDC to realize a spatial upscaling. Results indicate a greater effect of management on yields and soil organic carbon and nitrogen stocks compared to climate change effects. Intensively managed lysimeters under climate change conditions had the highest average yields, but showed a pronounced decrease during droughts while extensively managed lysimeters were more robust against these climate extremes. Process understanding and the consideration of different services is necessary to develop sustainable management strategies for these grasslands.

Keywords: biomass, soil organic carbon, nitrogen, lysimeters, process-based model

Introduction

Managed grasslands are a major land use in pre-Alpine regions and fulfil a variety of ecosystem services like feed provision for livestock, carbon (C) and nitrogen (N) storage, water retention and habitat for wildlife (Bengtsson *et al.*, 2019; Reynolds and Frame, 2005; White *et al.*, 2000). These services may be affected by climate and management changes. A profound knowledge about these effects is needed to develop sustainable management strategies. This study aims to analyse the effects of climate change and management on dry matter (DM) yields, soil organic carbon (SOC) and N stocks, as well as greenhouse gas (GHG) emissions of pre-Alpine grasslands by combining experimental and modelling studies.

Materials and methods

This study was conducted in the TERENO pre-Alpine observatory in southern Germany (Kiese *et al.,* 2018). The observatory adopts a space-for-time approach, where large undisturbed soil monoliths (lysimeters) were translocated down an elevation gradient to simulate climate change with a temperature increase and precipitation decrease. These grassland lysimeters are operated with two management intensities (ext = extensive: 3 cuts and 1-2 slurry fertilizations; int = intensive: 4-5 cuts and 4-5 slurry fertilizations) according to the local farmers' practice. Various environmental parameters like air temperature (T_{air}), precipitation (P), soil temperature and moisture, drainage, and GHG fluxes (via automatic chamber systems) are monitored. Furthermore, DM yields as well as C and N content of the harvested biomass are determined for each cut and lysimeter. We analysed annual DM yields of lysimeters from the highest elevation site (CTRL = control; 860 m a.s.l.; P = 1,295 mm a^{-1}; T_{air} = 6.9 °C) and of lysimeters from the highest elevation site translocated to the lowest elevation site (CC = climate change; 600 m a.s.l.; P = 962 mm a^{-1}; T_{air} = 8.9 °C) from 2012-2020. Additionally, we studied GHG fluxes (CH_2, CH_4, N_2O) of lysimeters from the medium elevation site (CTRL2; 770 m a.s.l.; P = 1,111 mm a^{-1}; T_{air} = 8.8 °C) compared to lysimeters from the medium elevation site translocated to the low elevation site from 2016-2018. Each treatment (e.g. CTRL_ext) was represented by 3 replicates.

Analyses of experimental data were accompanied by simulation studies with the biogeochemical model LandscapeDNDC (Haas *et al.*, 2013). These modelling studies offer the possibility to extent the spatial scale (e.g. regional analysis) and test different climate and management scenarios. Here, we show some applications for the Ammer region.

Results and discussion

Annual DM yield

Mean annual DM yields of intensively managed lysimeters were significantly higher than those of extensively managed lysimeters, but showed also higher annual variations (Figure 1). There was no significant difference between the mean annual DM of the CTRL and CC treatment (*P*>0.4), either for extensive or for intensive management.

GHG emissions

During the study period, the analysed soils acted as a N_2O source and a CH_4 sink. Annual CO_2 emissions ranged from 15.1 to 24.6 t C ha^{-1} a^{-1} and ecosystem respiration was strongly temperature driven. Climate change conditions significantly increased CO_2 and N_2O emissions, while management did not significantly affect these GHG emissions (Figure 2).

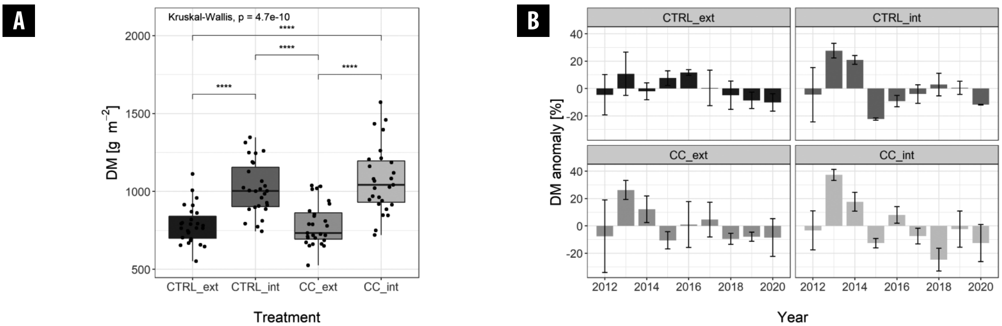

Figure 1. Annual DM yields per treatment (A; significantly different means are indicated: **** *P*≤0.0001) and (B) time series of annual DM yield anomalies per treatment for TERENO pre-Alpine lysimeters (2012-2020).

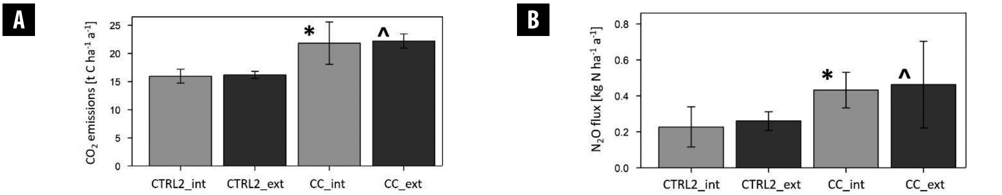

Figure 2. (A) Mean annual CO_2 emissions (ecosystem respiration) and (B) mean annual N_2O fluxes per treatment for TERENO pre-Alpine lysimeters (2016-2018). * indicates a significant difference of CC_int compared to CTRL2_int; ^ indicates a significant difference between CC_ext compared to CTRL2_ext.

Modelling

Simulation results and other experimental studies (Kühnel *et al.*, 2019) show that SOC and N stocks strongly depend on management. Historical fertilization with solid manure resulted in a build-up of humus and associated C and N stocks in these pre-Alpine grasslands. The currently widespread fertilization with liquid manure with its lower C content as well as climate change lead to a decrease in SOC and N stocks. Regional model applications (Figure 3) can be used for risk assessment (e.g. identifying areas of concern due to nitrate leaching) and the optimization of grassland management (e.g. via simulating different management scenarios).

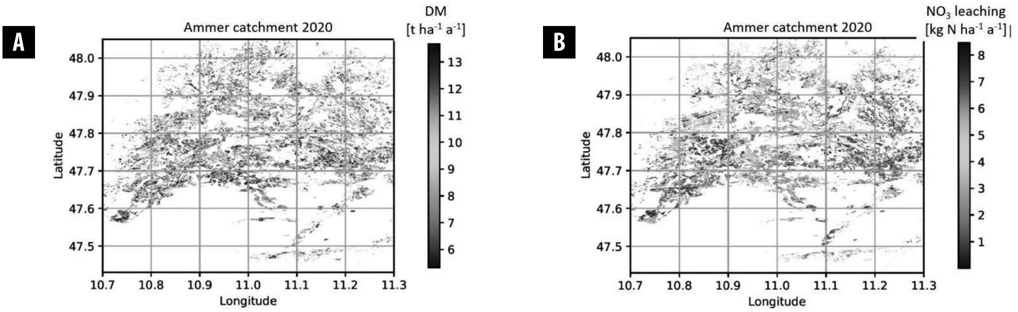

Figure 3. Results for the regional application of the LDNDC model in the Ammer catchment for 2020 under current management: DM yield [t ha^{-1} a^{-1}] (A) and nitrate leaching [kg N ha^{-1} a^{-1}] (B).

Conclusions

Our results show that grassland management and climate change differently affect yields and ecosystem matter fluxes in our study region. Therefore, a thorough understanding of processes, drivers and their interaction is needed to develop sustainable management strategies for pre-Alpine grasslands that consider different ecosystem services. Studies combining experimental and modelling approaches can help to improve this understanding.

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References

Bengtsson J., Bullock J.M., Egoh B., Everson C., Everson T., O'Connor, T., ..., and Lindborg R. (2019) Grasslands- more important for ecosystem services than you might think. *Ecosphere* 10: e02582

Haas E., Klatt S., Fröhlich A., Kraft P., Werner C., Kiese R., ..., and Butterbach-Bahl K. (2013) LandscapeDNDC: a process model for simulation of biosphere–atmosphere–hydrosphere exchange processes at site and regional scale. *Landscape Ecology* 28, 615-636.

Kiese R., Fersch B., Baessler C., Brosy C., Butterbach-Bahl K., Chwala C., ..., Schmid H.P. (2018) The TERENO pre-Alpine observatory: integrating meteorological, hydrological, and biogeochemical measurements and modeling. *Vadose Zone Journal* 17, 180060.

Kühnel A., Garcia-Franco N., Wiesmeier M., Burmeister J., Hobley E., Kiese R., ..., Kögel-Knabner I. (2019) Controlling factors of carbon dynamics in grassland soils of Bavaria between 1989 and 2016. *Agriculture, Ecosystem & Environment* 280, 118-128.

Reynolds S. and Frame J. (2005) *Grasslands: developments, opportunities, perspectives*. CRC Press, Boca Raton, FL, USA.

White R.P., Murray S. and Rohweder M. (2000) *Pilot analysis of global ecosystems – Grassland ecosystems*. World Resources Institute, Washington, DC, USA.

Piloting resource-efficient grass silage production on fifty dairy farms

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Abstract

The European Union's Farm to Fork strategy aims to enhance environmentally friendly management practices, improve nitrogen-use efficiency (NUE) and reduce risk of nitrate leaching. Nutrient balances and NUEs were studied in grass silage production on 50 dairy farms and 100 fields around Finland (2019-2021). Pilot farmers followed a grassland standard, where plant nutrition was optimized based on soil analysis and plant requirement. Grass yield was measured and analysed for quality as well as for macro- and micronutrient contents. The analysis results were used for the calculation of field-specific nutrient balances and NUEs. The average grass yield was 9,350±440 kg DM ha^{-1} and the crude protein content reached the target level each year. Nitrogen (N) balances were negative and NUEs were high, ranging from 89±8 to 127±16 between the different harvests. The Product Carbon Footprint (PCF) of grass silage (CO_2-e t^{-1} $DMkg^{-1}$) was significantly affected by both the NUE and grass yields. Based on our results we discuss how farmers can ensure good quality yields and high NUEs under variable growing conditions in conventional farming systems.

Keywords: dairy farms, grass, nutrient use efficiency, nutrient balance, silage

Introduction

The largest dairy company in Finland, Valio Ltd., has set a goal to reach zero carbon footprint of milk by 2035. In grass silage production greenhouse gas (GHG) emissions will be reduced by more resource-efficient production of high-quality grass silage, increased use of recycled nutrients as well as carbon farming methods. In a mainly indoor-housing system of milk production the main sources of GHG emissions are methane from enteric fermentation and manure management, and nitrous oxide emissions from manure and mineral fertilizer nitrogen (Flysjö *et al.*, 2011). Thus, the optimized use of nitrogen inputs plays an important role in reaching the target of zero carbon footprint of milk. Nutrient balances (N, P) and the NUE can be used as indicators of successful nutrient management. The aim of this farm-scale study was to evaluate the effect of optimized plant nutrition on grass silage yield and quality as well as on nutrient balances and NUE.

Materials and methods

Fifty Valio Ltd. dairy farms located between 60°47 to 64°13 N were selected as Carbo pilot farms (Figure 1). Carbo pilot farms were equipped with Vaisala AWS310 weather stations, SoilScout soil sensors (at 5, 15, 25 cm depth) measuring soil moisture and temperature, DG60 axel scales as well as with Yara Atfarm satellite services for the creation of variable rate mineral fertilizer application maps. Soil samples from each pilot field were analysed for macro- and micronutrients and soil health (Eurofins, Finland). Grass mixtures were classified into four groups: grass mixtures, grass mixtures with clover, multiple-species mixtures (more than four different species) and grass mixture including *Festuca arundinacea* var. *aspera*. The optimal harvesting time was predicted by taking grass samples for the analysis of digestibility one to four times prior to each cut (Valio Ltd. laboratories). Grass yield was measured, and a representative sample was analysed for nutritional quality including macro- and micronutrient contents. Field-specific nutrient balances and NUE was calculated based on these measurements. The Product Carbon Footprint

(PCF) of grass silage (CO_2-e t^{-1} $kg^{-1}DM^{-1}$) was calculated for 16 pilot fields with common PCF methods and using IPCC (2019) methods and emission factors where relevant.

Results and discussion

Carbo pilot farmers followed the grassland standard where plant nutrition and harvesting time was optimized to gain high yield with good quality. One third of the used seed mixtures had red clover. Pure grass mixtures dominated in north-eastern and northern Finland, whereas multispecies mixtures and clover containing mixtures were more typical in southern Finland, where severe drought periods were common. Total nitrogen use was 200 to 240 kg N ha^{-1} for two to three cuts, respectively. On average 30% of N and 60% of the P was derived from organic fertilizers. High NUE was ensured by balanced crop nutrition. The target value for digestibility was 680 to 700 g kg^{-1} DM^{-1} and for crude protein content 130-160 g kg^{-1} DM^{-1}.

Total grass silage yield ranged from 8,760 to 9,820 kg DM ha^{-1} 2019-2021 (Table 1) and the target values for quality parameters were reached each year for all harvests. The variation in grass yield as well as nutrient balances and NUE was significant when some of the fields experienced severe droughts (2nd cut 2019 and 2021) or damage from winter kill (winter 2019-2020). The N balance is suggested as a tool to predict N leaching risk in crop production. The acceptable N balance in grass silage production has been set to +60 kg N ha^{-1} $year^{-1}$ (Salo *et al.*, 2013). This maximum N balance was reached at 325 kg N ha^{-1} in an N response trial (Termonen *et al.*, 2020). In pilot fields the N balances were negative with the exception of 2020 when it was slightly positive with +5 kg N ha^{-1} (Table 1). In 2019 and 2021 the severe summer droughts resulted in yield losses and positive N balances for the 2nd cut (15 and 19.7 kg ha^{-1}, respectively). It is noteworthy that the surplus N was utilized by the 3rd cut where the N balance was again negative (-24.6 in 2019 and -27.5 kg ha^{-1} in 2021). Due to the negative N balances, the NUE was also high, ranging from 97 to 110 across the cuts. At present there is no set target values for NUE in grass production. For winter wheat the desirable NUE is between 50 and 80, where values over 80 refer to soil mining of N. The average silage grass yield in Finland is around 5000 kg DM ha^{-1} (Luke, 2020-2021) and the average use of N is 140 kg ha^{-1} (Composed from the Statistics of Finnish Food Authority, 2018). Based on these values, the average N balance in grass silage production is around 36 and NUE 74.

The PCF varied between 132 and 1239 kg CO_2e t^{-1} kg^{-1} DM^{-1} between farms, fields and individual cuts. Fertilization dominated the PCF, and a lower PCF was found in the plots and cuts that had a higher and NUE. Grass yield also significantly affected PCF. This means that improved fertilization management (in particular N management) is of major importance for reducing PCFs in these systems.

Figure 1. The location of the 50 pilot farms in Finland.

Table 1. Average values of grass yield, N- and P-balances of all pilot fields in years 2019-2021.[1]

	Cut	2019	2020	2021	Mean	SD
Yield (t DM ha^{-1})	1	4,241.5	3,375.5	4,231.1	3,949.4	497.0
	2	3,229.2	3,417.9	2,672.7	3,106.6	387.4
	3	2,356.2	1,964.6	2,556.0	2,292.3	300.9
	all cuts	9,826.9	8,758.0	9,459.8	9,348.2	443.4
N-balance	1	-3.9	15.4	-2.3	3.1	10.7
	2	15.0	-0.2	19.7	11.5	10.4
	3	-24.6	-10.3	-27.5	-20.8	9.2
	all cuts	-13.4	5.0	-10.0		
P-balance	1	-4.7	1.0	-2.5	-2.1	2.9
	2	2.5	-2.3	3.3	1.2	3.0
	3	-7.8	-3.2	-4.1	-5.0	2.4
	all cuts	-10.0	-4.6	-3.3		

[1] Grass silage was cut 2 to 3 times in the growing season.

Conclusions

Grass silage production with optimized plant nutrition ensured high yields with desired quality for pilot farmers. As a result, NUE was improved, risk for N leaching reduced and the PCF was low. Thus, optimized grass silage production can have a significant contribution to carbon neutrality in milk production.

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References

Flysjö A., Henriksson M., Cederberg C., Ledgard S. and Englund J-E. (2011) The impact of various parameters on the carbon footprint of milk production in New Zealand and Sweden. *Agricultural Systems* 104, 449-469.

IPCC (2019) Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories. Calvo Buendia E., Tanabe K., Kranjc A., Baasansuren J., Fukuda M., Ngarize S., Osako A., Pyrozhenko Y., Shermanau P. and Federici S. (eds.). IPCC, Switzerland.

Salo T., Turtola E., Virkajärvi P., Saarijärvi K., Kuisma P., Tuomisto J., Turakainen M. (2013) Nitrogen fertilizer rates. N balances and related risk of N leaching in Finnish agriculture. *MTT Report* 102, 1-37. http://www.mtt.fi/mttraportti/pdf/mttraportti102.pdf.

Termonen M., Korhonen P., Kykkänen S., Kärkönen A., Toivakka M., Kauppila R. and Virkajärvi P. (2020) Effects of nitrogen application rate on productivity, nutritive value and winter tolerance of timothy and meadow fescue cultivars. *Grass and Forage Science* 75, 111-126.

Long-term grassland productivity with and without ploughing

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Abstract

Several studies conclude that permanent and temporary swards are equally productive, given equal management. In Norway, one experimental field trial has been maintained since 1974 (Fureneset; 61°18'N, 5°4'E). This ongoing experiment includes long-term/permanent ley (no-tillage over 25 and 45 years) next to temporary leys reseeded regularly. The objective of the study was to test reseeding/renovation methods that may maintain long-term forage productivity. We hypothesized that sod-seeding after chemical fallowing improves grassland productivity equally to that from reseeding after ploughing. In 2017, the frequently ploughed treatments, and half of the 25-year-old sward, were renewed by ploughing and reseeding with grass-clover seed mixtures. The second half of the 25-year-old sward was chemically fallowed and sod-seeded. The treatments included three different fertilizer strategies: mineral fertilizer (210 N kg ha^{-1}) and cattle slurry in combination with mineral fertilizer (210 and 340 kg total-N kg ha^{-1}). On average for four production years (2018-2021) the dry matter yield (DMY) of permanent sod-seeded 25-year-old ley was about 11 t ha^{-1}, and these yields were equal to swards renewed by ploughing and reseeding.

Keywords: forage, ley, permanent, sod-seeding, yield

Introduction

More than 90% of crops grown on Norwegian soils are turned into food through animal production systems. Sward establishment by ploughing, cultivation and sowing is energy demanding and time consuming. Moreover, Norwegian grassland farming is often located in marginal areas with unfavourable weather conditions resulting in limiting ploughing and reseeding activities. Therefore, there is a need for alternative renovation strategies, particularly in long-term grasslands where productive grass species have given way to herbs and weeds (Lundekvam and Myhr, 1975). Jones and Roberts (1989) concluded that reseeding without ploughing might increase yields by up to 30% if successful. In Canada, improving naturalized pastures by sod-seeding with legumes has been accepted as an inexpensive renovation method (Graves *et al.*, 2012). Here, we present results from an experimental field trial at Fureneset (61°18'N 5°4'E) in West Norway. This trial, which has been maintained since 1974, includes plots which have been maintained without ploughing for over 45 and 25 years, as well as frequently ploughed and reseeded treatments. We hypothesized that sod-seeding of long-term grassland maintains or increases permanent grassland productivity equally as well as renewal of long-term grassland by ploughing and reseeding.

Materials and methods

The long-term trial was established at NIBIO research stations in West Norway, Fureneset, Vestlandet (61°18'N 5°4'E; 15 m a.s.l.) in 1974. The soil has developed on morainic material and consists of a medium peaty topsoil that merges with a poorly drained subsoil. Until 2016, the trials included four main-plot treatments with different sward ages established with three replicates per trial. These were: PG, Permanent grassland established in 1974; S-PG, Semi-permanent grassland established in 1992; LEY-6, 6-year ley; and LEY-3, 3-year ley.

In 2016, the S-PG treatment was either renewed by chemical fallowing and ploughing (S-PGp) or chemical fallowing and direct sod-seeding (S-PGs). LEY-6 and LEY-3 were also ploughed, reseeded and the production period extended to 12 (LEY-12) and 6 years (LEY-6), respectively. A grass-clover mixture was used in all plots renewed by ploughing and sod-seeding. Extremely high precipitation in summer 2016 limited sward establishment and a new direct seeding was carried out in 2017. Repeated sod-seeding was carried out in the 25-year-old S-PGs plots in 2020. Three different fertilization practices were included on sub-sub-plots. Nitrogen (N) applied in the form of mineral fertilizer only (MF; 210 kg N ha^{-1}) and cattle slurry combined with mineral fertilizer (CS+MF; 210 and 340 kg N ha^{-1}). Plant biomass was harvested according to common practice in the region, three times during the growing season. In 2017, however, only one cut was performed in the renewed plots because of repeated sod-seeding and excessive precipitation in the second part of the growing season. The herbage yields were determined after each cut. The data were analysed by general linear model and one-way ANOVAs. For pairwise comparisons of treatments, Tukey's post-hoc test was used.

Results and discussion

On average for four production years, the renewed semi-permanent grassland and reseeded leys had significantly higher forage yield than the permanent grassland (>45 years without ploughing and reseeding), regardless of fertilization strategy ($P<0.001$; Figure 1A). The DM yields from reseeded treatments were 1.2 to 1.3-fold higher than from PG treatments. The results from this field experiment suggest that cultivated PG can maintain good and stable forage production under appropriate fertilization practice over several decades. This agrees with several studies, which have concluded that under equal management conditions, permanent and temporary swards are equally productive (Hopkins *et al.*, 1990; Nevens and Reheul, 2003). Acceptable drainage and lime conditions are important to maintain good yields over time (Lundekvam and Myhr, 1975).

Results from our study support our hypothesis that sod-seeding can be a good alternative to ploughing. In the two first production years after renewal there were no differences in forage DM yields (Figure 1B). After repeated sod-seeding, S-PGs yielded significantly more than S-PGp ($P<0.05$), particularly in the first and second cuts. Assessment of botanical composition showed that forage biomass contained more timothy (*Phleum pratensis*) and perennial ryegrass (*Lolium perenne*) in S-PGs than in S-PGp treatments (data not shown). This may be a plausible explanation for significantly greater yields in S-PGs than in S-PGp (Figure 1B). Thus, if sod-seeding is successful, it may also provide environmental benefits as grasslands may store significant amounts of C (Soussana *et al.*, 2004).

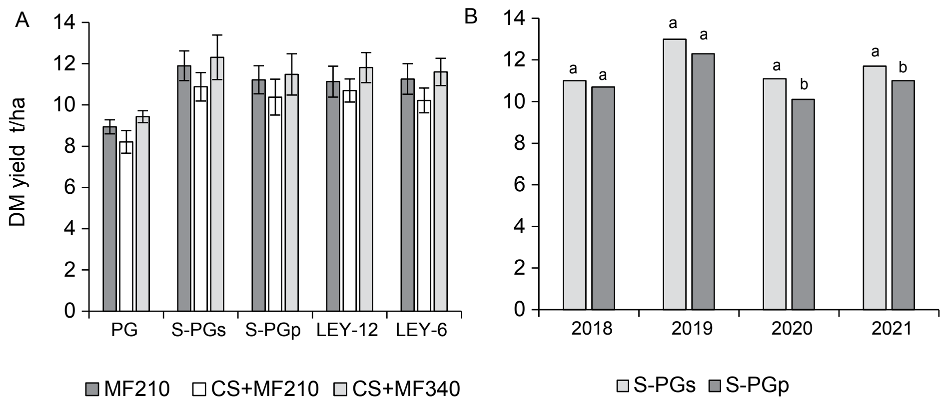

Figure 1. Average DM yield for four production years for permanent grassland (PG; A), semi-permanent grassland sod-seeded (S-PGs) and reseeded after ploughing (S-PGp; A and B) and reseeded ley in 2016/2017 (LEY-6 and LEY12; (A) fertilized with mineral fertilizer only (210 MF) or cattle slurry in combination with mineral fertilizer (210 CS+MF and 340 CS+MF). Different letters denote significant differences within renewal strategies.

Fertilization strategy significantly affected forage production. In particular, treatment 210 CS+MF had significantly lower forage DM yield ($P<0.05$) than treatments 210 MF and 340 CS+MF. On average, for all production years, the DM yields of treatment 210 MF were equal to the 340 CS+MF treatment (data not shown). The lowest level of N applied in spring as cattle slurry resulted in the lowest yields, indicating that only a part of N from cattle slurry is available for the first cut. However, fertilization strategies that include cattle slurry might be a good management practice and might give more advantages than disadvantages in the long-term.

Conclusions

Our findings show that permanent grasslands are productive and can give good yields over several decades. However, both reseeding by ploughing and also without ploughing essentially increased sward productivity compared to that of permanent grassland. Thus, sod-seeding can be a good alternative to ploughing.

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References

Graves M.E., McLean N., Jones G. and Martin R.C. (2012) Pasture and sheep performance response to sod-seeding red clover (Trifolium pratense L.) or white clover (Trifolium repens L.) into naturalized pastures in eastern Canada. *Animal Feed Science and Technology* 177, 7-14.

Hopkins A., Gilbey J., Dibb C., Bowling P.J. and Murray P.J. (1990) Response of permanent and reseeded grassland to fertilizer nitrogen 1. Herbage production and herbage quality. *Grass and Forage Science* 45, 43-55.

Jones E.L. and Roberts J.E. (1989) Sward maintenance of Lolium multiflorum by slurry seeding. *Grass and Forage Science* 44, 27-30.

Lundekvam H.E. and Myhr K. (1975) Forsøk med fornying av gamal eng på Vestlandet i åra 1965-1972. *Forskn.Fors. Landbr*. 22, 135-156 (in Norwegian).

Nevens F. and Reheul D. (2003) Permanent grassland and 3-year leys alternating with 3 years of arable land: 31 years of comparison. *European Journal of Agronomy* 19, 77-90.

Soussana J.F., Loiseau P., Vuichard N., Ceschia E., Balesdent J., Chevallier T. and Arrouays D. (2004) Carbon cycling and sequestration opportunities in temperate grasslands. *Soil Use and Management* 20, 219-230.

Microelement contents in soil, plants and animal tissues of a selected mountainous habitat

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Abstract

In order to determine microelement contents in soil, plants and animals, tissue and excrement samples were collected from the research area. Plant material was collected from fallow grasslands with a history of varying intensity of agricultural management. Average contents of microelements in meadow sward were typical of acidic soils. Levels of copper, zinc and iron were all very low. The examined materials showed that forage for animals from the examined sites varied in terms of its contents of micronutrient, but this did not exclude the occurrence of their deficiencies, or excess in the case of heavy metals.

Keywords: forage quality, wild animals, microelements

Introduction

Agricultural abandonment on many marginal grazing areas has had profound impacts on the nature conservation value and landscape integrity throughout much of Europe (Hopkins and Holz, 2006). In addition to environmental functions such as biodiversity, fallow agricultural land beside forest areas constitutes the fodder base for deer (Zarzycki and Szewczyk, 2013). The quality of the forage correlates with the chemical composition of the soil, and nutrient deficiencies that affect the health or rate of growth of ruminant animals are generally more widespread than those affecting grass growth (Kopeć, 2002; Whitehead, 2000). This study attempts to determine the relationships between contents of selected microelements in soil, and their concentrations in plants and animal tissues. Roe deer were selected as the diagnostic species due to their specific life routine. Roe deer (*Caprelous capreolus* L.) is a species meeting a number of criteria which are required for sensitive bioindicator animals, because of the confirmed correlation between the level of environmental pollution and the degree of accumulation of toxic compounds in the tissues (Cygan-Szczegielniak and Stasiak, 2022). Roe deer is considered an example of a species with a strong attachment to its habitat, with little tendency to migrate (Ossi *et al.*, 2020). It can therefore be assumed that there are significant relationships between the content of microelements in soil, plants and the animals that eat these plants.

Materials and methods

The research was carried out on semi-natural grassland communities located in the Czarny Potok valley near Krynica. The research area covered an area of approx. 50 ha. Based on observations of places of appearance and feeding of roe deer, 16 locations were selected. Soil, plant and animal excrement samples were collected from spring 2019 to the summer 2021. Samples after preliminary treatment (drying, grinding) were analysed for the content of selected minerals. Animal samples in the form of a liver fragment (11 pieces) from the research area were obtained with the help of a hunting club operating in the research area. In order to determine the content of elements in the samples from the studied area, plant material was collected and, after mineralization and transfer to solution, was analysed using the inductively coupled plasma mass spectrometry (ICP-MS) technique.

Results and discussion

The area covered by the research is very diverse in terms of micronutrient contents (Table 1). The contents of microelements in soils of fallow land did not exceed the standard values for unpolluted soils (Lipinski, 2013). Increased contents of Pb, Zn, Cu, Cr and Ni in some of the samples were presumably related to the source rock. This and its mineralogical composition, is a factor determining the amount of microelements, especially in the case of soils subject to mountain weathering. The content of microelements also depends on the granulometric composition and the amount of organic matter (Letkowska and Bogacz, 2000).

Table 1. The content (mg kg^{-1}) and variability (V) of microelements in tested soils.

Item	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Min.	0.05	8.2	1.6	3,533	43.1	3.81	4.71	23.18
Max.	1.41	17.1	29.6	6,832	326.3	12.5	30.8	74.8
Mean	0.52	16.13	17.76	6,432	271.7	10.60	16.9	62.6
V (%)[1]	30	31	13	10	26	40	78	29

[1] V % = SD × 100/Mean.

The average content of trace elements (Table 2) in the sward is typical of swards on acidic soils. Very low levels were found for copper, zinc, and also iron. The coefficients of variation in the content of trace elements are small, which indicates a slight variation within the sites. Cu is a component of enzymes involved in iron metabolism, and deficiency of this element could cause anaemia (Whitehead, 2000).

Table 2. The content of micronutrients (mg kg^{-1}) in the DM biomass of fallow grasslands.

Item	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Min.	0.19	0.13	1.96	17.72	141.13	0.53	0.23	23.22
Max	0.46	0.42	4.29	80.38	285.64	0.99	0.53	46.87
Mean	0.31	0.32	3.56	45.23	224.42	0.73	0.42	34.81
V (%)[1]	22	22	15	29	20	17	18	22

[1] V % = SD × 100/Mean.

The contents of trace elements in the examined animal tissues were characterized by high variability (Table 3). According to Tajchman *et al.* (2020, 2021) animals living in an uncontaminated area can have higher concentrations of some heavy metals than values reported from industrial regions. The high coefficient of variation and the small size of the population do not allow for the formulation of unambiguous relationships in the soil-plant-animal chain. Cygan-Szczegielniak and Stasiak (2022) found some cases of higher levels of heavy metal contents in tissues of female roe deer. Age-related differences in the content of individual metals were also confirmed but the directions of changes were inconsistent. The chemical composition of animal excrements (Table 4) appeared less variable.

Table 3. Variation in the chemical composition of roe-deer liver samples (mg kg^{-1}).

Item	Cd	Cr	Cu	Fe	Mn	Ni	Pb	Zn
Min.	0.24	0.91	8.40	113	6.66	0.18	0.45	39.9
Max.	6.64	58.27	279	3,539	51.07	35.32	14.65	3,721
Mean	1.73	17.74	69.44	379	19.18	10.85	1.50	180
V (%)[1]	83	80	92	103	49	80	136	275

[1] V % = SD × 100/Mean.

Table 4. Variation in the chemical composition of roe-deer faeces samples (mg kg^{-1}).

Item	Cd	Cr	Cu	Fe	Mn	Ni	Zn
Min.	0.064	0.56	5.11	132	66,6	1.03	26.1
Max.	2.025	54.5	34.0	3,753	2,967	31.74	222
Mean	0.650	12.8	12.5	459	865	9.45	81.1
V (%)[1]	70	83	57	132	92	66	56

[1] V % = SD × 100/Mean.

Conclusions

The analysis of mineral compounds, including heavy metal contents in animal tissues, can be used for the evaluation of exposure of wild animals to these pollutants. High coefficients of variation in the chemical composition of soil, plants and animal tissues do not allow for an unambiguous formulation of the relationship between animals and the quality of the habitat. However, this research showed that there was no risk of heavy metals presenting a hazard to roe-deer in the research area. A possible natural higher content of heavy metals in soils could be related to the food chain of animals with a limited territorial range. The results of the analysis of animal tissues could also be partially modified by the influence of winter feeding of free-living animals.

References

Cygan-Szczegielniak D. and Stasiak K. (2022) Effects of age and sex on the content of heavy metals in the hair, liver and the *longissimus lumborum* muscle of roe deer *Capreolus capreolus* L. *Environmental Science and Pollution Research* 29, 10782-10790. https://doi.org/10.1007/s11356-021-16425-6

Hopkins A. and Holz B. (2006) Grassland for agriculture and nature conservation: production, quality and multi-functionality. *Agronomy Research* 4(1), 3-20.

Kopec M. (2002) Causes of mountain meadow soil chemical degradation in long-term fertiliser experiment. *Rostlinna Výroba* 48, 159-166.

Letkowska A. and Bogacz A. (2000) Zawartość mikroelementów w glebach odłogowanych Dolnego Śląska (Microelement contents in fallowed soils of Lower Silesia), *Zesz. Probl. Post. Nauk Rol.* 2000 z. 471: 379-386

Lipinski W. (2013). Zasobność gleb Polski w mikroelementy (Abundance of microelements in Polish soils). *Studia i raporty IUNG-PIB*, Z. 24(8), 121-131. https://doi.org/10.26114/sir.iung.2013.34.09

Ossi, F.,Ranc, N.,Moorcroft, P., Bonanni, P. and Cagnacci, F. (2020) Ecological and behavioral drivers of supplemental feeding use by roe deer *Capreolus capreolus* in a peri-urban context. *Animals* 10, 2088. https://doi.org/10.3390/ani10112088

Tajchman, K., Ukalska-Jaruga, A., Bogdaszewski M., Pecio, M. and Dziki-Michalska, K. (2020) Accumulation of toxic elements in bone and bone marrow of deer living in various ecosystems. a case study of farmed and wild-living deer. *Animals* 20, 2151. https://doi.org/10.3390/ani10112151

Tajchman, K., Ukalska-Jaruga, A., Bogdaszewski, M. *et al.* (2021) Comparison of the accumulation of macro- and microelements in the bone marrow and bone of wild and farmed red deer (*Cervus elaphus*). *BMC Veterinary Research* 17, 324. https://doi.org/10.1186/s12917-021-03041-2

Whitehead D. C. (2000) *Nutrient elements in grassland: soil–plant–animal relationships*. CABI Publishing, Wallingford, UK.

Zarzycki J. and Szewczyk W. (2013) Impact of abandonment on the floristic composition of permanent grassland and grassland created on former arable land. *Grassland Science in Europe* 18, 421-423.

Establishment and production of lucerne in Sweden is affected by inoculation product choice

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Abstract

Lucerne (*Medicago sativa* L.) is an important perennial forage legume in Sweden, but its potential cultivation area is constrained by uncertainty of successful establishment. This study aimed to identify management practices that could lead to improved establishment of lucerne. Lucerne cultivar SW Nexus was grown at four different locations in southern Sweden over two establishment/production cycles. At all locations except Svalöv, lucerne had not previously been cultivated in the plot location for at least seven years. One control (C), one standard rhizobia inoculation treatment (SI), three micronutrient (M1-M3) and eight inoculation treatments (I1–I8) were assessed for their effects on plant growth and development. At Svalöv, where lucerne had previously been grown, there was no effect of any of the treatments. The largest contrast between inoculation treatments was at Rådde in the first year, where the best treatment yielded 12,000 kg DM ha^{-1} across three harvests, nearly twice that of the control. There was no evidence that the soil-applied micronutrients improved yield at any location. In conclusion, inoculation is essential at locations where there is no history of lucerne cultivation, and choice of inoculation product can affect establishment and yield.

Keywords: agricultural management, field experiment, forage legume, nodulation, yield

Introduction

Lucerne (*Medicago sativa* L.) is an important crop in Sweden, but somewhat constrained in its potential cultivation area by issues with establishment. It has specific environmental requirements for growth, including soils with high pH, adequate drainage, sufficient macro and micronutrients, and specific rhizobia (Xu *et al.*, 2016). If these conditions are met then it can fix atmospheric nitrogen, persist, and be highly productive and drought tolerant (Li *et al.*, 2019). However, failure to provide these conditions can result in establishment failure. The objective of this paper is to identify inoculation treatments that could lead to improved lucerne establishment. The general hypothesis is that where lucerne has not previously been cultivated, there will be significant differences between inoculation products/techniques.

Materials and methods

The experiment was conducted at four sites in southern Sweden: Svalöv (Skåne), Tenhult (Småland), Rådde (Västergötland) and Lilla Böslid (Halland), over two establishment and production years (2019/2020 and 2020/2021). At Tenhult, Rådde and Lilla Böslid, lucerne had not previously been cultivated in the plot location for at least seven years. Svalöv was a control site, where lucerne was known to have been grown recently. The experiment had a randomized complete block design with three replicates in 2019/2020 and four replicates in 2020/2021. The sowing plot sizes were 9.60×1.13 m at Svalöv, 15.4×1.75 m at Tenhult, 12.0×1.75 m at Rådde and 12×1.5 m at Lilla Böslid. The adjacent plots were separated by a buffer zone (0.5 m). After field preparation, macronutrient fertilizers (P, K and S) were applied according to soil tests and standard recommendations.

The experimental treatments (Table 1) included one control (C), one standard inoculant (SI), three SI combined with single micronutrient treatments (M1–M3) and eight alternative inoculant treatments (I1–I8) (seven in experiment 1). Lucerne seeds (SW Nexus) were sown as a monoculture (without a cover crop) with ten rows per plot (nine at Svalöv). Inoculants of individual treatment were prepared to

Table 1. Details of individual treatments in field experiments.

Code	Treatments	Details
C	Control, no treatment	
SI	Nitragin Gold	Novozymes A/S, Bagsvaerd, Denmark
M1	SI + Molybdenum	Novozymes A/S, Bagsvaerd, Denmark
M2	SI + Cobalt	Novozymes A/S, Bagsvaerd, Denmark
M3	SI + Boron	Novozymes A/S, Bagsvaerd, Denmark
I1	SI 5× rate	Novozymes A/S, Bagsvaerd, Denmark
I2	SAS Gold	Jouffray-Drillaud, Cisse, France
I3	SAS GR01	Jouffray-Drillaud, Cisse, France
I4	SAS Life	Jouffray-Drillaud, Cisse, France
I5	Thermoseed + SI	Lantmännen BioAgri, Uppsala, Sweden
I6	Pellifix	Legume Technology, East Bridgford, UK
I7	LegumeFix + Lime coating	Legume Technology, East Bridgford, UK
I8	Prolime 100	Prolime AG, Laingsburg, USA

manufacturers' instructions. All management operations, such as weed and pest control, were performed according to standard practices.

Lucerne was harvested with a stubble height of approximately 8 cm, three times per production year. It was harvested in the establishment year only if there was enough biomass. The harvest plot size was 8.80, 11.0, 10.4 and 12.5 m^2 at Svalöv, Tenhult, Rådde and Lilla Böslid, respectively. Biomass samples were collected from each plot and dried at 105 °C until a constant weight was reached, to determine the dry matter (DM) content and calculate the DM yield of each harvest. Treatment results are means of three replicates. The statistical analyses were conducted separately for each site using Proc Glimmix in SAS (version 9.4, SAS Institute Inc., Cray, USA). Differences among treatments were determined using Tukey's test at a significance level of $P<0.05$.

Results and discussion

In the first establishment/production year (2019/2020), lucerne DM yield showed significant variation among different treatments at Tenhult and Rådde, where lucerne had not previously been cultivated, confirming that Rhizobia inoculation is essential for sites without lucerne cultivation history (Jauregui *et al.*, 2019). Lucerne at Svalöv was not harvested in the establishment year, as biomass was lower due to an earlier cut to remove weeds. There were no significant differences between yield treatments at Svalöv in the production year (Figure 1). At Tenhult, in the establishment year and at harvest 1 of the production year, SI, two micronutrient and four alternative inoculant treatments achieved significantly higher yield than the control (C), but no treatments were significantly different from each other. For harvest 2, SI, one micronutrient and five alternative inoculant treatments yielded higher than C, and the best treatment yielded significantly higher than the worst one. No significant differences were found in harvest 3. For the total yield, all but one treatment were more productive than C. At Rådde, in the first establishment year, four alternative inoculant treatments achieved significantly higher yield than C, and the two best alternative inoculant treatments yielded significantly higher than SI. For three harvests of the production year, all treatments yielded significantly higher than C, alternative inoculant treatments yielded higher than SI and micronutrient treatments; there were no significant differences among SI and micronutrient treatments, suggesting that applying micronutrients to the soils was not effective for increasing lucerne production. At Lilla Böslid, there were very few differences between treatments; results not shown due to the influence of weeds at this site in 2019/2020.

In the second establishment/production year (2020/2021), variations among treatments at all sites were less than the first year (Figure 2). At Rådde, in the second establishment year, only one alternative inoculant treatment (I4) yielded higher than C, with no significant differences among other treatments. No significant differences were observed at any harvest (H1–H3) in the production year. At Lilla Böslid, the difference in yield among treatments were not significant at any harvest. The reason for distinct differences between the two establishment years is unknown but could be due to pre-existing soil rhizobia or contamination between plots. This suggests that fields for inoculation experiments should be chosen carefully, and the effects of inoculation may vary between different fields and years. Similar to the first year, there were no significant differences among treatments at Svalöv in the second year (Figure 2). The 2020/2021 experiment at Tenhult was discarded due to the poor establishment.

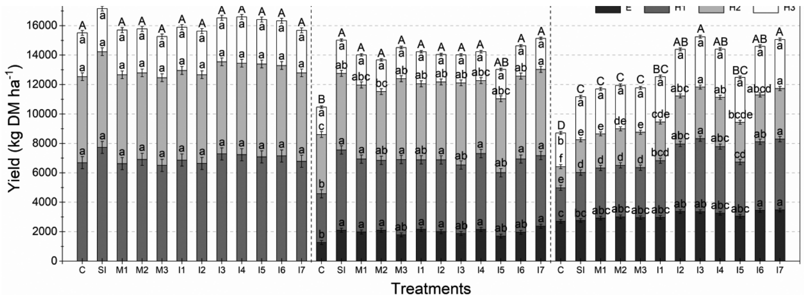

Figure 1. Dry matter yield of lucerne in response to different treatments, at three sites in southern Sweden (year 2019/2020). E: establishment year. H: harvest.

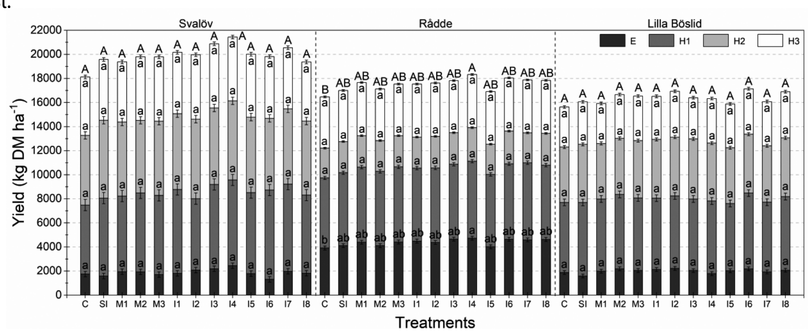

Figure 2. Dry matter yield of lucerne in response to different treatments, at three sites in southern Sweden (year 2020/2021). E: establishment year. H: harvest.

Conclusions

This study suggested that alternative inoculants were better than SI, particularly in the first production year at Rådde. There was no added benefit of soil-applied micronutrients at any site. Where there is recent history of successful lucerne cultivation (Svalöv), lucerne could be well established and productive without pre-inoculation of seed before sowing. However, effects of inoculation vary between different fields and years and therefore seed treatment could serve as an insurance for a well-established and productive lucerne crop.

References

Jauregui J.M., Mills A., Black D.B.S., Wigley K., Ridgway H.J. and Moot D.J. (2019) Yield components of lucerne were affected by sowing dates and inoculation treatments. *European Journal of Agronomy* 103, 1-12.

Li Q., Zhou D., Denton M.D. and Cong S. (2019) Alfalfa monocultures promote soil organic carbon accumulation to a greater extent than perennial grass monocultures or grass-alfalfa mixtures. *Ecological Engineering* 131, 53-62.

Xu K., Wang H., Li X., Liu H., Chi D. and Yu F. (2016) Identifying areas suitable for cultivation of *Medicago sativa* L. in a typical steppe of Inner Mongolia. *Environmental Earth Sciences* 75, 341.

Estimating biorefinery output from forage crops via the Cornell Net Carbohydrate and Protein System

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Abstract

The biorefinery technology aiming at green biomass protein extraction for monogastric animals is increasing and this raises the need to identify suitable inputs. Forage crops have previously been evaluated by the Cornell Net Carbohydrate and Protein System (CNCPS), and the results used as proxies for extractable protein, but validation is lacking. Validation and application of the method are the aims of this study with two experiments. Experiment I (validation) included spring cut of two grasses (perennial ryegrass and tall fescue) and three legumes (lucerne, red and white clover) processed in a lab-scale refinery. The initial biomass, the pulp fraction and the precipitated protein concentrate were CNCPS analysed. Total recovery in concentrate was highest for legumes, which points to an advantage of these species in protein extraction setups. In experiment II (application), four annual cuts during two full seasons for the two grasses and three legumes were CNCPS analysed and translated into potential extraction of protein concentrate by applying the results from experiment I. Results show that red clover and lucerne had the highest protein concentrate yield per ha (835 and 803 kg CP ha^{-1}) and reveal that the entire season needs attention for optimization.

Keywords: CNCPS, crude protein, grass, red clover, white clover, lucerne

Introduction

Green forage plants, such as legumes and grasses have long been used as high-quality protein sources for ruminants, while monogastrics generally cannot efficiently utilize these protein sources (Buxton, 1996; Stødkilde *et al.*, 2019). Protein concentrate obtained from green biomass of grasses and forage legume species has shown promising properties both in terms of protein concentration and balanced amino acid composition (Damborg *et al.*, 2020). However, possible forage crop species, and their combinations, management, etc. are numerous and thus a fast evaluation of biorefinery potentials is needed. CNCPS divides crude protein (CP) into fractions, e.g. based on solubility, why relative recovery in refinery output products expectedly differ between these fractions. Here we (1) validate the capability of applying the CNCPS to estimate potential protein concentrate biorefinery output from forage crops in experiment I, and (2) apply the method on a full year field trial on forage species following a four-cut strategy in experiment II.

Materials and methods

Field trials were carried out at Foulumgård (56°30′N, 9°35′E), Denmark. Forage crops for experiment I were sampled in 2016 (May 17, May 24, May 31, and June 6) and subsamples were analysed according to CNCPS into A, B1, B2, B3 and C fractions according to the protein properties and solubility (Licitra *et al.*, 1996; Tylutki *et al.*, 2008) (see further in Thers *et al.* (2021)). Legume species were white clover (*Trifolium repens* L.), red clover (*Trifolium pratense* L.), and lucerne (*Medicago sativa* L.) and the grass species were perennial ryegrass (*Lolium perenne* L.) and tall fescue (*Festuca arundinacea* L.). Samples were processed in a lab scale biorefinery and the protein concentrate and pulp were CNCPS analysed. Based on CNCPS protein fractions before and after refinery, the species-specific recovery of plant CNCPS protein fractions into the biorefinery protein concentrate output was calculated.

In experiment II the same five species were included for full-season CP yield evaluation following a four-cut strategy in 2015 and 2016 (cut in May, July, August and October at 7 cm height). The two grasses were fertilized at three nitrogen (N) levels (175, 350 and 525 kg N ha^{-1}), whereas legumes were non-N-fertilized, resulting in a total of nine forage crop treatments. Subsamples were analysed according to the CNCPS method. The relative flow of CNCPS protein fractions from plants into refinery output derived from experiment I was applied to experiment II results and in that way potential full-season refinery output was estimated.

Statistical tests were done using R (R Core Team, 2020) and test of differences between treatments and species was performed by the lme mixed linear model in the nlme package followed by a post hoc Tukey test ($\alpha = 0.05$) using the glht function in the multcomp package.

Results and discussion

For the total CP recovery (Table 1), the legumes showed significantly higher recovery than grasses in the protein concentrate (and opposite for the pulp protein recovery). Biorefinery managers will presumably focus on the concentrate output since this has the higher economic value, and thus, these results point to an advantage of legumes as compared to grasses.

Plant crude protein yield from experiment II showed that red clover had the highest CP yield in 2015, whereas the highest N fertilizer level of tall fescue and lucerne gave the highest yield in 2016 (Figure 1). When the recovery obtained from experiment I was applied on the plant CP yield, red clover (2015) and lucerne (2016) had the highest potential protein concentrate output (Figure 2).

Table 1. Percentage of plant CP that is recovered in the protein concentrate.[1]

Species	A (%)	B1 (%)	B2 (%)	B3 (%)	C (%)	Total (%)
White clover	22 (4)AB	33 (1.4)A	51 (5)A	8 (1.2)B	12 (5)A	35 (3)A
Red clover	17 (3)AB	45 (12)A	58 (6)A	21 (5)A	5 (2.1)B	35 (5)A
Lucerne	26 (1.1)A	20 (4)A	58 (3)A	8 (1.3)B	7 (0.7)AB	39 (1.9)A
Per. ryegrass	16 (2.1)B	22 (9)A	29 (2.5)B	22 (3)A	6 (1.9)AB	23 (2.0)B
Tall fescue	19 (1.5)AB	38 (12) A	33 (15) B	18 (5) A	6 (2.1) AB	26 (5) B

[1] The recovered CP is shown as total (in column 6) and as divided on CNCPS fractions (i.e. A, B1, B2, B3, and C) for each of the five forage species. Values are means of 8 samples for each species (average of four harvest dates). Numbers in parenthesis indicate standard errors. Letters indicate significance in the vertical direction ($P<0.05$).

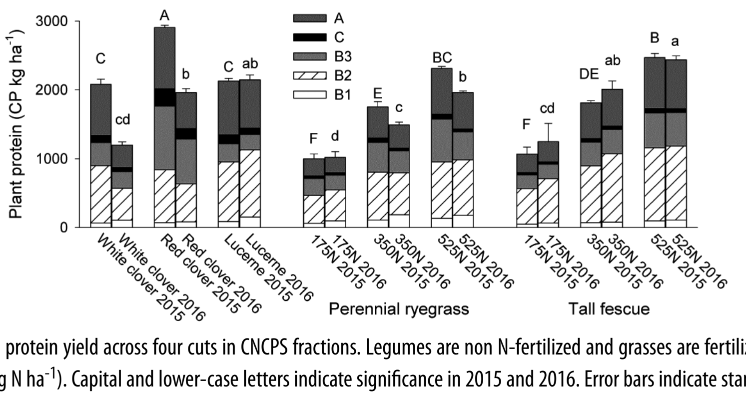

Figure 1. Total crude protein yield across four cuts in CNCPS fractions. Legumes are non N-fertilized and grasses are fertilized at three levels (175, 350 and 525 kg N ha^{-1}). Capital and lower-case letters indicate significance in 2015 and 2016. Error bars indicate standard error for the full bar (all CNCPS fractions).

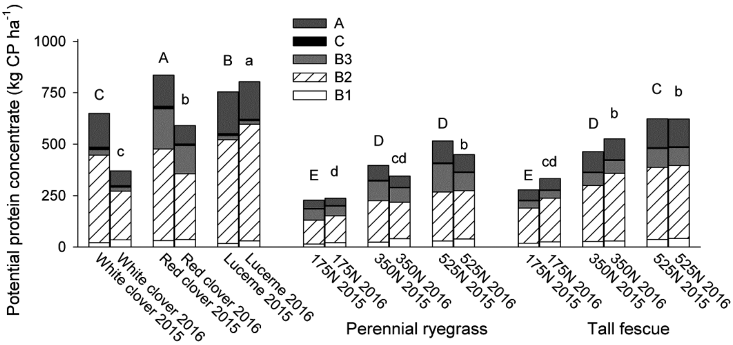

Figure 2. Potential extraction of protein concentrate across four cuts. Legumes are non N-fertilized and grasses are fertilized at three levels (175, 350 and 525 kg N ha^{-1}). Capital and lower-case letters indicate significance in 2015 and 2016. Error bars indicate standard error for the full bar (all CNCPS fractions).

Conclusions

Accounting for potential recovery of plant CP into protein concentrate revealed that red clover and lucerne yielded the highest potential for protein concentrate. The non-N fertilized legumes thus exceeded highly fertilized grass species. Further, a higher proportion of the soluble B2 fraction could be extracted from the legumes compared to the grasses, pointing to a potentially higher quality of protein concentrate for monogastric animals.

Acknowledgements

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References

Buxton, D.R. (1996) Quality-related characteristics of forages as influenced by plant environment and agronomic factors. *Animal Feed Science and Technology* 59, 37-49.

Damborg, V.K., Jensen, S.K., Weisbjerg, M.R., Adamsen, A.P. and Stødkilde, L. (2020) Screw-pressed fractions from green forages as animal feed: Chemical composition and mass balances. *Animal Feed Science and Technology* 261, 114401.

Licitra, G., Hernandez, T.M. and Van Soest, P.J. (1996) Standardization of procedures for nitrogen fractionation of ruminant feeds. *Animal Feed Science and Technology* 57, 347-358.

R Core Team (2020) R: A language and environment for statistical computing. Available at: https://www.r-project.org/.

Stødkilde, L., Damborg, V.K., Jørgensen, H., Lærke, H.N. and Jensen, S.K. (2019) Digestibility of fractionated green biomass as protein source for monogastric animals. *Animal* 13, 1817-1825.

Thers, H., Stødkilde, L., Jensen, S.K. and Eriksen, J. (2021) Linking protein quality in biorefinery output to forage crop crude protein input via the Cornell net carbohydrate and protein system. *Applied Biochemistry and Biotechnology* 193, 2471-2482.

Tylutki, T.P., Fox, D.G., Durbal, V.M., Tedeschi, L.O., Russell, J.B., Van Amburgh, M.E., ... and Pell, A.N. (2008) Cornell Net Carbohydrate and Protein System: A model for precision feeding of dairy cattle. *Animal Feed Science and Technology* 143, 174-202.

A farm level decision support tool to quantify ecosystem service delivery from permanent grassland

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Abstract

The existence and management of permanent grassland (PG) is key to the delivery of multiple ecosystem services (ES) across Europe. The development of a farm-level decision support tool (DST) is being undertaken as part of the EU-H2020 project 'SUPER-*G*' to help inform farmers' decision process. The aim of the tool is to provide the farmer (user of the tool) with an overview of the various ES delivered through the management of PG within their farm. A multi-actor approach, with discussions between farmers, landowners and their advisers, non-governmental organizations and researchers was undertaken to develop the tool. The user needs to answer a series of questions which will calculate values for six ES. These ES indicators are then combined using simple additive weighting to provide the user with scores for six ES. This paper assesses the farmer perception of currently available tools and introduces the proposed alternative tool. Feedback from farmers revealed current tools did not fulfil requirements, and there was appetite for a new ES tool. The new tool was demonstrated to a working group of farmers and was found to provide useful, intuitive feedback on farm ES. The working groups' feedback will be integrated into the building of the tool.

Keywords: ecosystem service, decision support

Introduction

The SUPER-*G* project aims to improve the understanding of the distribution and state of permanent grasslands (PG) in Europe and their ability to deliver a range of ES. This is being achieved through meeting specific objectives, one of which is the development of a farm level decision support tool (DST) for the management of PG to enhance productivity and the delivery of ES to society. A review of existing DSTs was carried out by Rankin and Lively (2020) and identified farmer demand for an alternative custom-built tool that simultaneously addresses several ES. Furthermore, generally few DSTs were found in Eastern Europe and it was concluded that existing DSTs should be tested in this area (Rankin and Lively, 2020). This paper assesses the farmer perception of currently available tools and introduces the proposed alternative tool. Feedback from users will be used to assess whether the development of a new tool is necessary, and if so, how the user responds to the custom tool to direct further development of the tool.

Materials and methods

The novel DST is being created following a user-based design process, which includes four steps: information gathering, development, evaluation, and implementation (Abelse *et al.*, 1998). Information gathering was undertaken to identify a short-list of agri-environmental indicators for the delivery of six ecosystem services (ES) covered by the project: food, wool and biomass production; climate regulation; biodiversity; landscape aesthetics, flood and erosion control; and water quality. During ideation, it was agreed that a suitable method would be simple additive weighting (SAW; Nurmalini and Rahim, 2017). Thus, each ES indicator will be assigned a percentage weighting, with increasing percentage indicating increasing importance for the final ES. The user will answer a series of questions to calculate the score for each ES indicator. The ES indicator score will be normalized to a score between 0 and 1 and multiplied by the ES indicator weighting to give a user score. The normalized score would also be presented on a chart to allow the user to benchmark against expected values. A score for each ES would be calculated by summing all user scores for each parameter pertaining to the ES. The report generated by the tool would provide an easy and intuitive traffic light system to identify where no improvement (green), moderate improvement (amber) or urgent action (red) was required (Figure 1). Suggestions for remedial action that the farmer can implement to improve the score will be provided. The tool will be provided free of charge to the end user.

Two working groups were set up, one in Hungary and one in Northern Ireland (NI). In Hungary, existing farm decision support tools were reviewed by Hungarian SUPER-*G* members together with a farm adviser who was in contact with local farmers. In NI, a group of farmers were invited to an online meeting to discuss existing tools and preview the proposed custom-built tool. Further options of whether the tool should allow users to register a profile to allow access from multiple devices, and whether the tool should collect user data to allow future benchmarking and research analyses of user inputs were discussed. Feedback was received in open discussion and through an online survey.

Results and discussion

In NI, 28 farmers took part in the working group. Of the respondents, 38% were dairy farmers, 38% were beef farmers, 19% were sheep farmers and 3% stated that they were 'other farm type'. Half of the respondents were from lowland farms, with 27% from disadvantaged areas and 23% from severely disadvantaged areas. 15% of respondents already used multiple farm management software tools and 29% of respondents did not use any software. Of those who used software, the same proportion (74%) of respondents used commercial software packages as used freely available software provided by government extension services. The Hungarian group reported that farmers were more likely to depend on advisors and word of mouth and generally did not use software DSTs.

In total, 11 DSTs were discussed (nine by the Hungarian team, three by the NI working group). Four tools were discarded because they were published in a language other than English, the working language of the consortium. A further four tools were excluded as they did not suit the farming system employed by the respondents or were too expensive. The Hungarian working group found the most suitable tool was a virtual fencing technology, which could potentially be used to manage ES, but it would not directly measure any impacts or monitor change. The NI working group concluded the tools presented were not

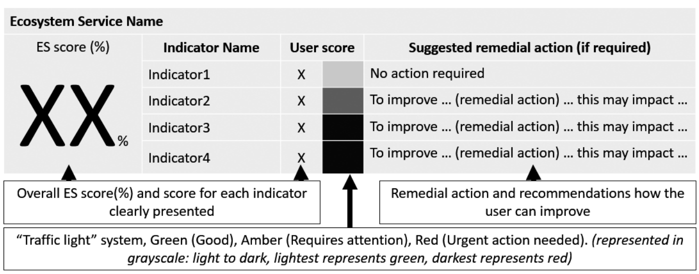

Figure 1. Example report presented to working group. One such table would be produced for each ecosystem service. Final user score given in colours of a traffic-light system.

applicable to all land types and only had limited influence on the daily decisions made when managing a farm. It was agreed that none of the tools fulfilled the role of a dedicated ES analysis and a custom tool was required for this specific purpose.

The proposed SUPER-*G* tool was presented to the NI working group. Table 1 shows that none of the respondents replied negatively to the proposed features of the tool. Some respondents were 'unsure' about the helpfulness of the tools. This suggests that these users were not comfortable interpreting the report. Thus it is essential that when introducing a novel reporting system there are clear instructions on how the information should be understood and used. The majority of users were happy to share their farm information for the generation of future benchmarking scales (provided data was anonymous and GDPR regulations followed).

Table 1. End user feedback on the proposed report generated by a custom designed ecosystem services decision support tool.

	Yes (% (n))[1]	No (% (n))[1]	Unsure (% (n))[1]
Is the overall ecosystem service score helpful?	73 (22)	0 (0)	27 (8)
Is the traffic light system for each indicator helpful?	76 (22)	0 (0)	24 (7)
Is the benchmarking chart intuitive?	79 (22)	0 (0)	21 (6)
Would you be happy to anonymously share your answers to develop future benchmarking scales?	89 (24)	0 (0)	11 (3)

[1] Denotes the proportion (%) of respondents who selected that answer and the number, n, in brackets.

Both the Hungarian and NI working groups suggested in that incorporating maps of the user's farm into the tool would increase its utility. Furthermore, prepopulating the tool with information (e.g. from existing farm management tools) would make it easier and more convenient to use. The large geographical and legislative area that the proposed tool is covering means it is not feasible to introduce maps or integration with existing systems whilst remaining free of charge to the end user. However, the tool will be designed to be agile and, if subsequently taken under custody of more localised areas, the tool will be equipped to incorporate these suggestions.

Conclusions

Feedback from multiple potential end users has shown an appetite for a tool which is specific to grassland ES. Subsequent workshop feedback has shown that the proposed tool is suitable and promising. A pilot of the tool will be built and evaluated by experts and farmer user groups. Where working group suggestions could not be included, they have been recorded and considered for future iterations and developments within the tool.

Acknowledgements

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References

Abelse E.G. *et al.* (1998). A user-based design process for web sites. *Internet Research* 8(1), 39-48.

Newell-Price, P., *et al.* (2020). Quantifying ecosystem services on permanent grassland - A short list of agri-environmental indicators. Horizon 2020 SUPER-*G* project (Grant Agreement No.: 774124) Deliverable report 2.2b. 25pp.

Nurmalini and Rahim R. (2017). Study approach of simple additive weighting for decision support system. *International Journal of Scientific Research in Science and Technology* 3, 541-544.

Rankin J. and Lively F. (2020) Set of requirements for a farmer DST that would attract maximum farmer adoption Horizon 2020 SUPER-*G* project (Grant Agreement No.: 774124) Deliverable report 5.2b.

Holistic environmental assessment of high nature value farming systems in Europe

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Abstract

Many life cycle assessment (LCA) studies comparing environmental impacts of different beef production systems are incomplete as they exclude biodiversity impacts and soil carbon stock changes. This study aims to assess the environmental impact of ruminant production on semi-natural grasslands or so-called high nature value (HNV) farms at the European level. We collected data for 24 HNV farms in Europe (in Finland, Estonia and France). The studied farms are extensive beef, sheep and goat production systems. We used LCA to assess the potential environmental impact of HNV farms according to global warming potential (GWP_{100}), eutrophication, fossil fuels and water use, by using the Solagro carbon calculator and OpenLCA software. Results showed that HNV farming systems have the potential to maintain unique biodiversity, act as carbon sinks, reduce greenhouse gas emissions and reduce nutrient losses and water use while producing animal-derived food. There were significant differences between HNV farms among countries in their greenhouse gas emissions at the farm level ($tCO_2eq\ ha^{-1}$) and N inputs (kg N ha^{-1}). Better regional understanding of the environmental impact performance of HNV farming systems in relation to sustainable ruminant production will be achieved as the undergoing study progresses.

Keywords: biodiversity, LCA, carbon calculator, carbon storage, sustainable ruminant production

Introduction

Livestock production systems vary greatly along the gradient of production intensity, which is likely to influence the overall environmental impact. Although intensive production has shown to result in lower greenhouse gas emissions (GHG) at the product level, extensive production is known to produce other environmental benefits such as biodiversity maintenance or carbon storage (Garnett, 2010), which are not commonly included in life cycle assessment (LCA)-based studies. When the discourse around livestock production sustainability focuses mainly on the global warming potential (GWP), and biodiversity and carbon storage gains are not properly accounted for, there is a high risk of depreciating other mitigation opportunities that are alternatives to intensifying production.

High nature value (HNV) farming systems are extensive production systems known for supporting farmland areas in Europe 'where agriculture is a major land use and where that agriculture supports, or is associated with, either a high species and habitat diversity or the presence of species of European conservation concern or both (Andersen *et al.*, 2003). No research to date has estimated the potential of HNV production systems across the continent in their sustainable ruminant production. The objective of the study is to assess the environmental impact of 24 HNV farms in three regions in Europe in terms of GWP_{100}, eutrophication and depletion of resources such as fossil fuels and water.

Material and methods

Our dataset corresponds to HNV type 1 farms (i.e. farms that utilize semi-natural vegetation for grazing and/or hay production). A total of 24 farms enrolled in the study: 18 beef cattle, 2 sheep, 2 dairy, 2 beef-and-sheep combined. The assessment of the environmental impact was based on a yearly cycle production system estimated upon 5-year average farm data.

We assessed the potential environmental impact of HNV farms by applying the LCA method using two types of software: the Solagro carbon calculator and OpenLCA 1.10. We applied the ReCiPe Midpoint 2016 (H) impact method to estimate GWP_{100}, fossil resource scarcity, land use, fresh water and marine eutrophication for the 24 farms in Finland, Estonia and France. We applied AWARE method for regionalized water use. The system boundary applied in this study was from cradle to farm gate. We estimate the contribution from farming practices such as manure management, and six environmental impact parameters: GHG emissions at the farm and product level ($tCO_2eq\ ha^{-1}$ and $tCO_2eq\ t^{-1}$ LW), total N inputs and outputs (N kg ha^{-1}), total C storage (tC). Biodiversity scores will be added as the study progresses. We will apply SALCA-BD approach to assess biodiversity in HNV farms. We ran ANOVA to test statistical differences between HNV farms within and between countries and Kruskal-Wallis test to assess the differences between farming practices among the conjoint of HNV farms.

Results and discussion

The environmental impact of HNV farms showed a wide variation between and within countries. There were significant differences in relation to GHG emissions at the farm level (kgCO2eq ha^{-1}), N inputs (kgN ha^{-1}) between countries ($P \leq 0.001$ and $P \leq 0.01$, respectively). Average values for GWP_{100} were marginally significantly different ($P \leq 0.08$) between countries. Similarly, there were no significant differences at the product level ($kgCO_2eq\ kg^{-1}$ LW).

Most of the environmental impact in terms of GWP_{100} occur at the farm (Garnett *et al.*, 2017). Our results showed enteric fermentation to contribute most to the average overall emission of 46%, followed by mineral fertilization, and indirect and direct N_2O emissions as 25, 22 and 13%, respectively. HNV farming practices such as circulation of on-farm manure and utilization of cover crops in temporary-grassland fields reduce nutrient loses. Therefore, the application of external inputs, i.e. mineral fertilizers, in HNV farms appeared to cause marginally significant differences between farms in the overall emissions ($P \leq 0.09$). However, HNV systems tend to have negative N balances compared to organic systems (Röös *et al.*, 2018) resulting in low eutrophication values (2.4 kg Neq kg^{-1} LW). Similarly, our results showed low water use values (5.68 $m^3\ kg^{-1}$ LW) caused mainly by the use of natural water sources in HNV farming systems.

The utilization of semi-natural grasslands and permanent grasslands reduces the requirements of purchasing feed. This reduces the overall emissions, as our results suggested ($P \leq 0.006$) and also contributes to carbon storage (Torres-Miralles, unpublished data) Therefore, intensive practices such as application of mineral fertilizers or feed purchases tend to negatively influence the overall performance of HNV farming systems.

There is, however, a wide range of performance among the HNV farms. HNV beef and sheep production had average levels of GWP_{100} at 18.67 and 18.63 kg $CO_2eq\ kg^{-1}$ LW, respectively. Farms with the highest GWP_{100} at the product level corresponded to those that have started their production recently or that retain the animals longer on farm premises, as is the case of two Finnish farms. When such farms (two out of eleven) are excluded, the average GWP_{100} falls to 2.3 kg $CO_2eq\ kg^{-1}$ LW per t of LW, lower than the mainstream Finnish beef production systems, 32.1 kg $CO_2eq\ kg^{-1}$ LW per kg of LW (Hietala *et al.*, 2021). Compared to other farms under mainstream production, according to other European studies, HNV beef have lower GWP_{100} (Nguyen *et al.*, 2010). However, GWP_{100} of beef products may not be comparable when livestock environmental assessments operate with different scopes and are potentially based on global averages.

Further analysis is required to reveal the nuances of the performance of HNV farms in relation to sustainable production. However, our results suggest that product-based environmental impact assessments alone may not reveal a complete sustainability picture of farming systems, HNV included. We demonstrate that, in order to assess sustainability for ruminant production systems, LCA assessments should account for biodiversity and carbon storage, and be framed in the sustainability discourse around farming practices. Assuming that a drastic reduction of animal products is necessary due to the unsustainability of western dietary patterns (Röös *et al.*, 2017), HNV farms, despite their lower yields, have the potential to supply sufficient animal source foods while supporting environmental benefits.

Conclusions

The relationship between the environmental impacts and associated benefits in livestock production is not simple. HNV farms, due to their circularity practices, tend to act as carbon sinks, maintain biodiversity, perform with low eutrophication and water use while reducing the overall GWP and produce animal-sourced food. This study contributes to attempts to quantify the potential of extensive ruminant production to minimize GWP while maintaining biodiversity and other environmental benefits.

References

Andersen, E., Baldock, D., Bennett, H., Beaufoy, G., Bignal, E., Brouwer, F. *et al.* (2003) Developing a high nature value indicator. Report for the European Environment Agency. Copenhagen, Denmark.

Garnett, T. (2010) Intensive versus extensive livestock systems and greenhouse gas emissions. Food Climate Research Network briefing paper.

Hietala, S., Heusala, H., Katajajuuri, J. M., Järvenranta, K., Virkajärvi, P., Huuskonen, A. and Nousiainen, J. (2021) Environmental life cycle assessment of Finnish beef-cradle-to-farm gate analysis of dairy and beef breed beef production. *Agricultural Systems* 194, 103250.

Nguyen, T.L.T., Hermansen, J.E. and Mogensen, L. (2010) Environmental consequences of different beef production systems in the EU. *Journal of Cleaner Production*, 18(8), 756-766.

Röös, E., Bajželj, B., Smith, P., Patel, M., Little, D. and Garnett, T. (2017) Greedy or needy? Land use and climate impacts of food in 2050 under different livestock futures. *Global Environmental Change* 47, 1-12.

Röös, E., Mie, A., Wivstad, M., Salomon, E., Johansson, B., Gunnarsson, S., Wallenbeck, A., Hoffmann, R., Nilsson, U., Sundberg, C. and Watson, C.A. (2018) Risks and opportunities of increasing yields in organic farming. A review. *Agronomy for Sustainable Development* 38(2), 1-21.

The repeatability of perennial ryegrass grazing efficiency as measured by Residual Grazed Height

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Abstract

A key grassland management strategy is to graze to low post-grazing height (4 cm) to maximize grass utilization and maintain/increase sward quality in future rotations. Perennial ryegrass varieties differ in their ability to be grazed to low post-grazing heights, as indicated from on-farm variety assessments and plot studies. Residual grazed height has been developed as a measure of perennial ryegrass variety grazing efficiency and has been included as a trait within the 2021 pasture profit index (PPI). The PPI is a variety selection tool used by the Irish seed industry. Variety grazing efficiency has been evaluated since 2015, but only now enough data exist from a number of studies to investigate the long-term variety grazing efficiency. The current study analysed data from four perennial ryegrass plot evaluations. Moderate to strong correlations were found between trials and evaluation years indicating that variety grazing efficiency is a repeatable trait. Such results provide confidence to the seed industry when making variety selections decisions.

Keywords: perennial ryegrass, variety, grazing efficiency, residual grazed height

Introduction

Perennial ryegrass (*Lolium perenne* L.; PRG) is the predominantly sown forage species in Ireland, producing large quantities (>15 t dry matter (DM) ha^{-1}) of high quality dry matter for animal production. Irish dairy farms operate a 300-day grazing season where grazed grass accounts for 80% of the cow's diet (O'Brien *et al.*, 2018). A key grassland management strategy is to graze to low post-grazing heights to maximize grass utilization and maintain/increase sward quality in future rotations. On-farm variety evaluations conducted in Ireland have provided an innovative new data set with which to investigate variety performance in a more varying and reflective environment compared to mechanically cut evaluation trials (Byrne *et al.*, 2017). Variety feedback from participating commercial farmers in the on-farm evaluations reported that certain PRG varieties were easier to graze to lower post-grazing heights (i.e. these varieties displayed greater grazing efficiency) and that varieties displaying improved grazing efficiency were demanded. Additionally, farmers highlighted that no indication of a variety's grazing efficiency was available within the pasture profit index (PPI) prior to sowing. The PPI is a variety selection tool used by the Irish grassland industry when making variety selection decisions. It is made up of a number of traits that influence the profitability of dairy farms in Ireland (O'Donovan *et al.*, 2016). In 2021, a new grazing utilization sub-index was introduced to the PPI. The sub-index uses data from variety grazing trials conducted at Teagasc Moorepark over a minimum of 2 years (Tubritt *et al.*, 2021). A number of varieties have been examined under a number of varying harvest and sowing years. The aim of this paper was to evaluate the repeatability of grazing efficiency among PRG varieties.

Materials and methods

Grazing efficiency evaluations took place in Teagasc Moorepark, Co. Cork, Ireland (50°70' N, 8°16' W). Average rainfall for the area is 1000 mm and average temperature ranges from 5.3 to 15.5 °C. Four studies evaluating grazing efficiency conducted over varying years were used in the analysis. Three of these studies (Study 1, 2, and 3) had greater than 2 years harvest data and were used for between-years comparisons. Study 4 had one year of harvest data and was included with the other studies to determine correlations between trials (sowing years). The protocol used was common across all studies. The protocol consisted of PRG monoculture plots sown in a complete randomized block design. These plots were then managed under a rotational grazing system where dairy cows grazed plots when the average cover across the plots was 1,400 kg DM ha^{-1} (above 3.5 cm). Prior to grazing, individual pre-grazing height was recorded on each plot using a rising plate meter (Jenquip, Fielding, New Zealand). A herd of cows then grazed the plots with the cows given free choice to graze whichever variety plot they wished. When the average post-grazing height across the plots was 4 cm, cows were removed and individual post-grazing height was recorded from each plot. Increasing pre-grazing height was shown to increase post-grazing height thereby creating bias by using post-grazing height as the sole measure of grazing efficiency. To account for this a regression model was created using SAS 9.4 to predict the post-grazing height of each variety. This model was:

Postheight = Trial(year) + harvest + Trial(block) + pregrazing height (Tubritt *et al.*, 2020)

The predicted post-grazing height of a variety was then subtracted from the actual post-grazing height of the same variety. This figure is termed the residual grazed height (RGH). Where a variety's actual post-grazing height is lower than predicted, the RGH value is negative which is indicative of high grazing efficiency. Where a variety's actual post-grazing height is higher than predicted, the RGH value is positive which indicates the variety has low grazing efficiency.

Repeatability of grazing efficiency was determined between harvest years (i.e. within trial) and sowing years (i.e. between varieties common to separate trials). The SAS procedure Proc Corr was used to determine the correlations in variety RGH between harvest years and between varieties with differing sowing years. Pearson's and Spearman's rank correlations were derived from the analysis. Correlations between years within each study were calculated using annual variety RGH and correlating these values between each evaluation year (i.e. Year 1 RGH of variety X was correlated to Year 2 RGH of variety X). The second analysis determined correlations for varieties common between studies.

Results and discussion

RGH was found to be moderately correlated between harvest years for both Pearson's and Spearman's rank correlation. The average Pearson's and Spearman's rank correlation between years for all studies was 0.61 and 0.64 respectively, with minimal variation between studies (Table 1). Variable growth conditions occurred between years with drought conditions experienced in 2018 reducing annual DM yield by 3t DM ha^{-1} (PastureBase Ireland, 2020). Despite this, grazing efficiency correlations between years remained moderately strong indicating that variety grazing efficiency is poorly influenced by weather conditions. Variety growth habit influences grazing efficiency and is controlled by a variety's genetics rather than meteorological conditions, which may explain why yearly correlations are stronger than those recorded for herbage yield (Mitchell, 1956).

Reseeding of pasture is an expensive investment on commercial farms. The PPI is designed to give an indication of variety performance and must be repeatable on-farm. Moderate to strong correlations were

Table 1. Average Pearson's and Spearman's rank correlations between evaluation years within trials.

Trial codes (years)	Study 1 (2015-2018)	Study 2 (2017-2019)	Study 3 (2019-2021)
Pearson's correlation	0.58 (P<0.001)	0.61 (P<0.005)	0.62 (P<0.004)
Spearman's rank correlation	0.60 (P<0.001)	0.67 (P<0.001)	0.64 (P<0.002)

found between trials evaluating grazing efficiency for the same varieties (Table 2). This indicates that variety RGH is repeatable between trials, and varieties should perform similarly on-farm under animal grazing. Including data from additional trial(s) to create an average RGH value improves the accuracy and consistency of variety grazing efficiency, with correlations between trials and average RGH in the range 0.86 to 0.94 (P<0.001).

Table 2. Pearson's correlation between perennial ryegrass Residual Grazed Height evaluated in separately sown plot evaluations (differing in sowing years) and the average residual grazed height value for each variety across all trials.

Trial	Study 1	Study 2	Study 3	Study 4	Average
Study 1	-	0.84 (14)[1]	0.51 (8)	1.0 (2)	0.94 (21)
Study 2	-	-	1.0 (2)	0.64 (3)	0.93 (3)
Study 3	-	-	-	0.83 (5)	0.91 (5)
Study 4	-	-	-	-	0.86 (6)

[1] Values in brackets indicate the number of varieties common between trials.

Conclusions

Perennial ryegrass variety grazing efficiency is a repeatable trait over years. Moderate correlations were found between trials for variety grazing efficiency indicating that single sowing years are relatively accurate in their evaluation. Evaluating varieties over additional sowing year(s) provides a robust indication of a variety's grazing efficiency.

References

Byrne N., Gilliland T.J., McHugh N., Delaby L., Geoghegan A. and O'Donovan M. (2017) Establishing phenotypic performance of grass varieties on Irish grassland farms. *Journal of Agricultural Science* 155, 1633-1645.

Mitchell K. (1956) Growth of pasture species under controlled environment. 1. Growth at various levels of constant temperature. *New Zealand Journal of Science and Technology, Section A* 38, 203-215.

O'Brien D., Moran B. and Shalloo L. (2018) A national methodology to quantify the diet of grazing dairy cows. *Journal of Dairy Science* 101, 8595-8604.

O'Donovan M., McHugh N., McEvoy M., Grogan D. and Shalloo L. (2016) Combining seasonal yield, silage dry matter yield, quality and persistency in an economic index to assist perennial ryegrass variety selection. *Journal of Agricultural Science* 155, 556-568.

PastureBase Ireland (2020). *Average grass growth in Ireland.* Available online.

Tubritt T., Delaby L., Gilliland T.J. and O'Donovan M. (2020) The relationship between the grazing efficiency and the production, morphology and nutritional traits of perennial ryegrass varieties. *Journal of Agricultural Science* 158, 583-593.

Tubritt T., Shalloo L., Gilliland T., McHugh N. and O'Donovan M. (2021) Incorporation of the grazing utilization subindex and new updates to the Pasture Profit Index. *Journal of Dairy Science* 104, 10841-10853.

Evaluating differences in grazing offtake of perennial ryegrass (*Lolium perenne* L.) under rotational sheep grazing

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Abstract

Forage intake is known to be linked to animal performance. The aim of this study was to evaluate the potential for using grazing offtake to detect differences between perennial ryegrass varieties of the same ploidy and maturity categories, as well as enable an evaluation of consistency for this trait between trials. Four separate plot-based trials (intermediate-heading diploid, intermediate-heading tetraploid, late-heading diploid and late-heading tetraploid) were sown in 2018, 2019 and 2020, including 8 varieties each of perennial ryegrass, and rotationally grazed by 15 hoggets across a 1-2 year period depending on sowing year. Two common varieties were included in all three sowing years to allow for comparison of these two varieties across years; other varieties included were unique to each trial. Pre-grazing and post-grazing compressed sward heights were measured with a rising plate meter, with the difference presented as a measure of offtake. No significant differences were detected in mean annual grazing offtake between varieties in 10 of the 12 grazing efficiency trials investigated. Significant differences in offtake between varieties were observed, however, in 2 of the 3 late tetraploid trials (2019 and 2020 sowings; $P<0.01$ and $P<0.05$, respectively). A multi-trial, over-year analysis of the intermediate tetraploid data revealed a significant difference in seasonal grazing offtake between the two common varieties, with one tetraploid variety grazed more in July than the other in two of the three field trials. These results support the view that ryegrass evaluation should be carried out over a number of years and seasons due to environmental variability.

Keywords: grazing efficiency, yield, variety testing, perennial ryegrass

Introduction

Grazing efficiency has been defined as the proportion of herbage ingested by grazing animals relative to that presented (Tubritt *et al.*, 2020), and has been used to describe how suitable a variety is for grazing. Grazing efficiency has increased in importance in recent years along with a recognition that perennial ryegrasses can differ in their levels of grazing efficiency, and that this is a desirable trait. This study aimed to evaluate variability for grazing offtake between varieties of similar heading date and ploidy, as well as assess the potential for breeding for this trait in perennial ryegrass with the aim of increasing grazing efficiency.

Materials and methods

Grazing offtake studies were carried out at AFBI Loughgall (54°27′N, 6°04′W) over three grazing seasons from 2019-2021. Twelve separate trials were sown: four trials in each year from 2018-2020, including one trial each containing material of intermediate diploid, intermediate tetraploid, late diploid and late tetraploid origin. Each trial contained eight varieties of perennial ryegrass, all of which derived from the AFBI grass-breeding programme (either pre- or post-commercialized). The same two common varieties were included in each of the trials sown depending on maturity and ploidy. Varieties were sown in a randomized complete block design with 3 replicates each (24 plots per trial). Each plot measured 2×4.5 m (9 m^2). Diploid and tetraploid varieties were sown at 25 and 37 kg seed ha^{-1}, respectively, to account for differences in seed size. Fertilizer was applied on 4 occasions throughout each season, (total of 288 kg

N ha^{-1}; 156 kg K_2O ha^{-1}). Plots were rotationally grazed using 30 hoggets in total divided into 2 groups of 15 each. Plots were grazed based on visual estimates of approximate mean sward height; grazing was initiated at 7-9 cm and ceased at 4 cm. A total of 54 grazing events occurred in total. Plots were mown back throughout the season after each grazing event to avoid carry over of non-grazed material into the next grazing event. Pre- and post-grazing sward heights were measured by assessing mean compressed sward height (function of sward height and density) using a rising plate meter (Jenquip, EC09). Trials were continued for two growing seasons each, except for trials sown in 2020, which were assessed for one full season. Offtake was calculated as the difference between pre-grazing and post-grazing height. Differences in offtake were analysed using analysis of variance, both for each trial (over-year) and across trials (multi-trial, over-year) to assess the effects of variety using TrialWizard statistical analysis software. Randomization was designed to enable Nearest Neighbour analysis to adjust for environmental variability in the field. Means were separated by Fisher's least significant difference (LSD) test at $P<0.05$.

Results and discussion

No significant differences were detected in offtake between varieties in 10 of the 12 grazing efficiency trials investigated. Significant differences were observed, however, in two of the three late tetraploid trials (2019 and 2020 sowings; $P<0.01$ and $P<0.05$, respectively). The apparent small differences between varieties would suggest that there may be low levels of variation within elite breeding populations for grazing offtake, once major sources of variation such as heading date and ploidy are removed. This could reduce the potential for breeding for this trait based on screening alone, although the high variability observed may also have obscured varietal differences. This study contrasts with others that have detected significant differences between perennial ryegrass varieties in grazing efficiency amongst and between varieties of differing ploidy and heading date (Byrne *et al.*, 2018; O'Donovan and Delaby; 2005; Tubritt *et al.*, 2020). It should be noted, however, that the trials in this study differed from those carried out previously: animal type; smaller size of plots; and mechanical defoliation between grazing events. Regarding the two common varieties included, there were no significant differences in mean annual offtake. However, a multi-trial, over-year analysis of intermediate tetraploid offtake revealed a seasonal effect, whereby one variety had a greater offtake than the other in July (Table 1). This significant difference was present in two of the three trials investigated (2018 and 2019 sowings), suggesting that this was a true varietal effect (Table 2). The two intermediate varieties included in these trials differed in heading date by a period of 10 days, which may explain the differences observed in mid-summer. Dry matter digestibility (DMD) is known to decrease following maturation, with earlier heading varieties tending to have lower DMD compared with later heading varieties at the same time point later in the season. Herbage digestibility has been previously shown to correlate with grazing efficiency of varieties across ploidy and heading date groups (Byrne *et al.*, 2018).

Table 1. Over-trial and over-year analysis of seasonal differences between pre-and post-grazing heights of two common intermediate tetraploid varieties sown in 2018, 2019 and 2020.

Variety	Heading date	April (cm)	May (cm)	June (cm)	July (cm)	August (cm)	September (cm)	October (cm)
Variety A	20-May	7.6	7.1	3.9	2.9	5.9	4.6	3.3
Variety B	30-May	6.8	6.3	4.3	5.1	5.6	5.4	2.9
cv. (plots)		21.9	34.3	17	25.8	26.5	17.9	38.7
LSD (0.05)					1.00			
F Varieties		-0.7	-0.6	-1.1	21.6	-0.1	-3.9	-0.4
P-value[1]		NS	NS	NS	$P<0.001$	NS	NS	NS

[1] NS = not significant.

Table 2. Over-year analyses of differences between pre-and post-grazing heights in July of two common intermediate tetraploid varieties sown in 2018, 2019 and 2020 (single trial and multi-trial).

Variety	Heading date	Sown 2018	Sown 2019	Sown 2020	Multi-trial (2018-2020)
Variety A	20-May	1.4	3.6	4.7	2.9
Variety B	30-May	4.4	6.0	3.9	5.1
cv. (plots)		19.4	20.0	23.3	25.8
LSD (0.05)		1.1	1.7		1.0
F Varieties		6.2	3.2	-1.8	21.6
P-value[1]		*P*<0.001	*P*<0.001	NS	*P*<0.001

[1] NS = not significant.

Conclusions

These studies revealed few differences in grazing offtake between varieties of the same ploidy and of the same heading date category, suggesting that genetic variability for grazing offtake may be low. The data from these trials will now be further analysed to assess the impact of differences in pre-grazing height on grazing offtake and the link between grazing offtake and grazing efficiency. The low variability for grazing offtake noted in this trial suggests that breeding efforts for beneficial traits linked to grazing may be more successful if the focus is shifted to known traits that increase grazing efficiency, such as digestibility, free leaf lamina or tiller mass (Byrne *et al.*, 2018), rather than testing for grazing traits directly. Seasonal differences were observed, however, in some of the trials investigated, suggesting that differences can be detected using methods such as described here. These differences highlight the need for multi-trial analyses to account for variability.

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References

Byrne N., Gilliland T.J., Delaby L., Cummins D. and O'Donovan M. (2018) Understanding factors associated with the grazing efficiency of perennial ryegrass varieties. *European Journal of Agronomy* 101, 101-108.

O'Donovan M. and Delaby L. (2005) A comparison of perennial ryegrass cultivars differing in heading date and grass ploidy with spring calving dairy cows grazed at two different stocking rates. *Animal Research* 54(5), 337-350.

Tubritt T., Delaby L., Gilliland T. and O'Donovan M. (2020) An investigation into the grazing efficiency of perennial ryegrass varieties. *Grass and Forage Science* 75(3), 253-265.

TrialWizard. Available at: https://www.beerepootconsultancy.com/trialwizard/.

Theme 3.
Using biodiversity to reduce vulnerability and increase resilience of grassland based systems

Using plant diversity to reduce vulnerability and increase drought resilience of permanent and sown productive grasslands

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Abstract

Climate change associated with a greater variability of inter- and intra-annual droughts and the occurrence of extreme events, act in combination to present challenges for semi-natural and sown productive grasslands in Europe. Successful plant strategies under drought strongly depend on stress intensity. Drought resistance to maintain leaf growth under moderate stress exhibits trade-offs with drought survival after cessation of growth under life-threatening drought conditions. Substantial intra-specific variability exists in key forage grasses originating from the Mediterranean to the cool-temperate climates, and represents a great potential for adaptation of future ecotypes and cultivars to a larger range of drought intensities. Plant species diversity offers an opportunity to stabilize forage production in two ways. First, growth reduction under stress is significantly smaller for diverse compared to simple plant communities because diverse communities offer the opportunity to include drought-resistant (or drought-surviving) species. Second, positive interactions among species increase ecosystem functioning of more diverse plant communities under moderate drought, allowing them to compensate for drought-induced yield reductions. Currently, available cultivars of perennial forage species adapted to dry climate are still rare, and only a few forage species are used in productive systems. Thus, both intra- and inter-specific plant diversity should be better valued to reduce vulnerability and increase resilience of productive grasslands.

Keywords: drought stress severity, resistance, survival, insurance effect, ecosystem functioning, complementarity

The wide environmental range of global grassland distribution demonstrates its huge adaptation potential

Grasslands cover about 40% of the world's land area (White *et al.*, 2000) and are among the most important agroecosystems delivering services ranging from forage supply for ruminants and soil carbon storage to habitats of high biodiversity. Reflected by the pedo-climatic conditions, these grasslands include a large variety of ecosystems such as steppe vegetation, savannah, tundra, alpine grassland and temperate grasslands. Moreover, large areas of land across temperate regions that would otherwise be covered by shrubs and trees are maintained as grassland by regular cutting and/or grazing. In short, grasslands can thrive across a vast range of pedo-climatic conditions and extremes, where shrubs and trees cannot grow and/or other agricultural systems are not economically profitable. This strongly suggests that grasslands benefit from plant strategies and ecological processes that ensure that they can grow, survive, resist, recover from, and/or adapt to strongly differing mean environmental conditions and to a multitude of extremes of environmental conditions experienced at different locations. With global climate change, both the mean and variation in climatic conditions are predicted to change (Orlowsky and Seneviratne, 2012), which will give rise to a change in biotic (e.g. weeds, disease, pests) and abiotic stresses (e.g. timing of the seasons, increased incidence of severe weather events, such as summer drought, heatwave, extreme cold, waterlogging).

Objectives

Although climate change will also affect natural and semi-natural (managed at low intensity, less productive, often species-rich) grasslands, we focus here on the effects of drought stress on productive grasslands that are highly modified and generally fertilized to maximize the production of aboveground forage yield and quality. Studies of semi-natural grasslands are referenced to underpin ecological theory and to highlight some specificities of the productive grasslands. The productive grasslands are sown with a low diversity (or even monoculture) of selected species (and cultivars) or are permanent grasslands with a relatively low plant diversity, both offering adaptation through changes in (1) species and (2) genotype composition, as well as (3) their diversity. We focus on the whole range of drought stress, from severe and predictable under Mediterranean climates to moderate and less predictable in temperate to cool climates, suggesting that a range of adaptive strategies are required.

A diversity of plant strategies to face drought

Better understanding of the adaptive strategies of forage plants to face drought is crucial to efficiently manage grasslands and breed cultivars that enhance the resilience of grasslands, i.e. a sufficient post-stress recovery to achieve a comparable post- vs pre-drought productivity. The two major plant response strategies under moderate and severe water deficit are 'drought resistance', i.e. the maintenance of leaf growth and biomass production, and 'drought survival', i.e. the plant ability to survive after growth cessation due to severe life-threatening drought (Volaire, 2018). Drought resistance is more relevant under moderate drought stress while drought survival is key under severe drought stress (Figure 1), but they can both enhance post-drought recovery and therefore resilience of plant communities.

Regarding plant strategies (Figure 1), 'dehydration escape' allows plants to shorten and complete the reproductive cycle before the onset of drought, e.g. annuals overcome drought as desiccation-tolerant seeds. For plants subjected to water deficit, drought resistance is associated mainly with a 'dehydration avoidance' strategy that maximizes water uptake and/or minimizes water loss to maintain high leaf water content and turgor ensuring growth maintenance. In contrast, drought survival is associated with a 'dehydration tolerance' strategy, allowing plants to tolerate moderate tissue dehydration in leaves and meristems. In some cases, dehydration tolerance rests on 'summer dormancy', which is an endogenous controlled strategy that reduces or stops meristem activity to render it relatively insensitive to growth-promoting signals (Volaire and Norton, 2006). Finally, the 'embolism resistance' strategy prevents xylem conduits from becoming air-filled or embolized under negative pressure (hydraulic failure) and hence underpins plant survival as drought drastically intensifies.

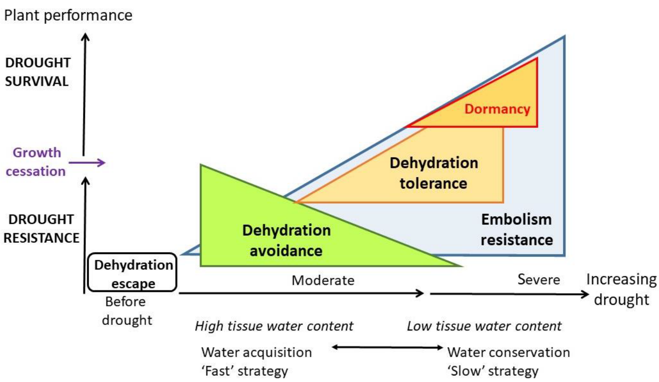

Figure 1. Plant strategies to face increasing water deficit. Dehydration avoidance enhances drought resistance (maintenance of growth at moderate drought), while dehydration tolerance, dormancy and embolism resistance enhance drought survival (Volaire, 2018).

To grow or to survive: a drought resistance vs drought survival trade-off

Most strategies can be combined to some extent, depending on species and populations. Growth maintenance (dehydration avoidance) is associated with fast water use under moderate drought ('water spender' strategy), while plant survival after growth cessation (dehydration and embolism tolerance, dormancy) is associated with slow water use under severe drought ('water saver' strategy). To 'stay green' and keep growing in dry conditions may contribute to depleting soil water and thus make plants extremely vulnerable to an extended and extreme drought (Zhao *et al.*, 2017). Consequently, dehydration avoidance is a strategy that does not enable plant survival under severe drought (Yates *et al.*, 2019). 'Knowing when not to grow' or 'when to senesce' enhance survival in the face of potentially lethal conditions. This is the case for summer dormancy, which confers to genotypes of some grass species the endogenous ability to cease or reduce aerial growth and senesce irrespective of the water supply in summer (Gillespie and Volaire, 2017). Summer dormancy confers superior survival after severe and repeated summer droughts (Norton *et al.*, 2006a,b), revealing that the endogenous and programmed ability to stop growth (or strongly reduce growth) during the drought period is the most efficient response to maximize plant drought survival.

Decoupling plant responses, i.e. growth under favourable summers/winters and plant survival under harsh summers/winters (Keep *et al.*, 2021), showed a general trade-off between seasonal growth potential and seasonal dehydration survival in 385 European populations of ryegrass (*Lolium perenne* L.). Three groups of ryegrass populations were identified according to their origin and contrasting strategies to face seasonal stresses, revealing a trade-off between dehydration avoidance and dehydration tolerance strategies. Populations from northern sites, where low-to-moderate summer drought occurs, mostly had a dehydration avoidance strategy and could maintain growth during summer without being threatened by drought. In contrast, populations from the southern sites, where intense summer drought occurs, had a dehydration-tolerant or a dehydration-escape strategy and survived prolonged drought by reducing their growth potential. Endogenous reduced seasonal growth potentials are phenological adaptations that can be regarded as dormancy levels. They were also identified within European populations of cocksfoot, *Dactylis glomerata* L. (Bristiel *et al.*, 2017), raising a possible generalized adaptive seasonal pattern within herbaceous species. Thus, the balance between productivity and stress survival is becoming a central issue in plant breeding for drought (and frost) survival (Ergon *et al.*, 2018; Volaire *et al.*, 2014) and therefore grassland resistance and resilience.

Plant diversity offers opportunities to stabilize forage production under drought stress

Lessons from semi-natural grasslands illustrate the stabilizing effect of diversity

The role of diversity in promoting resistance and resilience of ecosystem function in the face of environmental disturbance is well-established in ecosystems and experiments based in semi-natural grasslands (e.g. Craven *et al.*, 2016). For example, a meta-analysis of 46 plant association experiments by Isbell *et al.* (2015) revealed that biomass of 'low-diversity communities with one or two grassland plant species changed by approximately 50% during [severe] weather events, whereas that of high-diversity communities with 16-32 species changed by only approximately 25%'. Which ecological processes underlie such benefits of diversity? The 'insurance effect' refers to multiple biological processes that result in a stabilising effect of biodiversity on ecosystem function when subjected to environmental disturbance. The insurance effect includes: (1) the 'portfolio effect' which arises from independent (or sufficiently decoupled) fluctuations in species' abundances over time; (2) beneficial effects of biodiversity on both the mean and the variability of ecosystem properties, and; (3) spatial variability between patches or locations in heterogeneous landscapes (adapted from Loreau *et al.*, 2021). A key question is: do insurance effects occur in productive grasslands?

Species diversity in productive grasslands enhanced ecosystem function under drought

In productive temperate grasslands, where primarily resistance to moderate drought stress is targeted, there are few manipulations of environmental levels to test the role of diversity in maintaining ecosystem function under stress. Under experimentally imposed drought, species diversity enhanced *yield stability* (Grange *et al.*, 2022; Haughey *et al.*, 2018), reflecting the insurance effects of diversity through reduced temporal variance or mean-to-variance ratio. There is some evidence that the stabilizing effect in more diverse communities was caused by asynchrony of species' growth (Haughey *et al.*, 2018). Enhanced yield stability of mixtures compared to monocultures was also found in the AgroDiversity experiment among 16 sites with different climates (Schaub *et al.*, 2020). In the meta-analysis by Isbell *et al.* (2015), 24 of the 46 experiments contained monocultures and mixtures of two grasses and two legumes, so their conclusion that diversity confers higher resistance and stability in biomass productivity is also relevant to simple mixtures in productive grassland communities.

Under drought, more diverse forage mixtures were associated with higher (or at least equal) *yield* than less diverse mixtures or monocultures (overyielding; Hofer *et al.*, 2016; Komainda *et al.*, 2020; Skinner *et al.*, 2004), reflecting that positive complementarity effects on biomass production also occur under drought. Some studies have even shown that these positive effects were so strong that drought-stressed mixtures at least attained the yield of the average of the *rainfed* monocultures (Finn *et al.*, 2018; Grange *et al.*, 2021; Hofer *et al.*, 2016). Thus, growing mixtures instead of monocultures can mitigate negative effects of (moderate) drought. The use of drought-resistant forage species in such mixtures helps to partly overcome the limitations in nutrient uptake arising as a consequence of soil water limitation. Resistance to moderate drought has been shown to occur by sustained symbiotic N_2 fixation in legumes (Hofer *et al.*, 2017) or by increased resource uptake from deeper soil layers (Hoekstra *et al.*, 2015). Importantly, both beneficial species interactions and species' asynchrony are not mutually exclusive and can act simultaneously to increase stability in more diverse communities (Haughey *et al.*, 2018), including under conditions of environmental disturbance.

Improved drought-resistance by mixing species can also occur through the 'portfolio effect'. If mixtures contain at least one species that contributes substantially to community yield and that can cope with stress-induced reductions in growth (decoupled from other species performances), the overall community performance under drought is improved. This may be an important yield stabilising process in mixtures of legumes which were found to be drought resistant and grasses which showed a strong recovery after drought stopped (Hahn *et al.*, 2021; Haughey *et al.*, 2018; Hofer *et al.*, 2016). Interestingly, the portfolio effect can arise solely through (statistical) averaging of species performances over time (Loreau *et al.*, 2021). This has been little studied in productive grasslands. One line of evidence for the occurrence of averaging would be switching in the rank order of monoculture yields over time (and/or space), and especially switching in the identity of the best-performing monoculture. Such switching effects have been demonstrated to be important in the AgroDiversity experiment, which was conducted across 31 different international locations and broad climatic gradients (Finn *et al.*, 2013). Importantly, given that many agronomic studies compare mixture performance against the best-performing monoculture (which is selected in retrospect), and if switching continues over multiple years (and sites), then the relative benefit of mixtures would be expected to increase in comparison to the highest-yielding monoculture over that time period (and spatial scale). In such retrospective comparisons, the selection of the 'best-performing' monoculture enjoys the benefit of hindsight; however, past performance is not always a good predictor of future performance, and even less so when the future has more variable conditions.

Finally, the positive effects of species diversity on productivity and stability are context-dependent and may weaken under severe drought stress. For instance, complementarity effects (Barry *et al.*, 2018) that enable species-rich mixtures to achieve higher yields than monocultures during a moderate drought were not detected for the recovery and resilience of grass communities subjected to a severe drought (Barkaoui *et al.*, 2016). Low levels of soil water can make resource-partitioning among species inefficient. Most expectations are based on vertical segregation of root systems (Oram *et al.*, 2018), assuming that deep soil horizons represent an 'unused' pool of resources by shallow-rooted species, giving an opportunity to use additional water with deep-rooting species. However, deep soil horizons may completely dry out under a severe drought, making surface horizons the only ones with possible water recharge by episodic rainfall, therefore selecting shallow-rooted species only and limiting the complementarity effects. Similarly, facilitation, another facet of complementarity (Wright *et al.*, 2017), usually expected to positively affect productivity with increasing environmental severity (He *et al.*, 2013), may collapse among herbaceous species in areas prone to severe drought (Michalet and Pugnaire, 2016). Nevertheless, the portfolio effect should support the recovery capacity and resilience of species-rich mixtures subjected to severe drought (Kreyling *et al.*, 2017).

Saturation of diversity effects in semi-natural grassland experiments

The evidence given above suggests that species diversity is key to increase drought resistance and resilience of permanent and sown productive grasslands. In the following sections, we evaluate more closely the diversity-ecosystem function relationship. We discuss specific strategies to maximize the 'performance-enhancing effect' of diversity and the degree of diversity needed for adaptation of mixtures to drought stress.

Across a range of studies in semi-natural experimental grasslands, the yield benefits of adding species saturate at a relatively low number (Tilman *et al.*, 1997, 2014). Both theory (Tilman *et al.*, 1997) and empirical research (Hector *et al.*, 1999; Isbell *et al.*, 2017; Naeem *et al.*, 1994) have demonstrated a declining rate of increase in the overall diversity effect with increasing species richness. For example, in the BIODEPTH experiment (Hector *et al.*, 1999), the average biomass increase from doubling the number of species was approximately 80 g m^{-2}. This means that adding one species to a monoculture increased yields by ca. 23%, yet, adding one species to a four-species mixture increased yields by only 5%, and adding a further species to an eight-species mixture improved yields by 2%. The same principle in the performance-diversity relationship was also shown in two of the largest and longest-running biodiversity experiments, which are at Jena (Scherber *et al.*, 2010; Weisser *et al.*, 2017) and Cedar Creek (Tilman *et al.*, 2001), and for ecosystem processes such as community respiration, plant material decomposition, nutrient and water retention (Naeem *et al.*, 1994; Tilman *et al.*, 2014). A first reason to explain saturation of overall performance is the 'selection effect'. In the case of a random selection of species for the assembly of experimental communities, mixtures with a higher number of species have a higher likelihood of containing the most productive species, which shifts the performance towards the potential maximum (assuming that the most productive species becomes dominant in that community). A second reason for saturation comes from niche theory (Tilman *et al.*, 1997, 2014). Although more diverse communities have a higher chance for niche complementarity among particular species, the amount of unused resources gets increasingly smaller. Thus, the potential benefit of species interactions becomes smaller with increasing species richness. Moreover, species interactions can also be neutral or negative (e.g. Hofer *et al.*, 2016; Husse *et al.*, 2017), and the probability for the latter to occur might also increase with increasing species richness.

In productive grassland mixtures, diversity effects saturate even faster

Productive grasslands can show strong responses to diversity

Given the considerable differences between them, it is not necessarily the case that principles from semi-natural grasslands translate to productive grasslands. Over the past 20 years, however, research on forage mixtures in productive grasslands has provided strong evidence that legume-based mixtures with up to five species improve grassland performance (or compare well) relative to the respective monocultures. This has been observed in several responses, including yield, weed suppression, nitrogen yield, yield stability, forage quality, nitrous oxide emissions intensity and overall multifunctionality (Cong *et al.*, 2018; Connolly *et al.*, 2018; Cummins *et al.*, 2021; Finn *et al.*, 2013; Küchenmeister *et al.*, 2012; Lüscher *et al.*, 2014; Suter *et al.*, 2015, 2017, 2021c). This performance-enhancing effect of diversity is one of the key insurance effects to generate a stabilising effect of diversity on ecosystem function in a fluctuating environment such as severe drought events (Haughey *et al.*, 2018; Hofer *et al.*, 2016;. Most of the recent research on mixture benefits uses a modest number of species and less is known regarding mixture gains from more species-rich mixtures with >10 species (but see Jing *et al.*, 2017; Sanderson *et al.*, 2004; Tracy and Sanderson, 2004a).

In productive grassland mixtures the diversity response to yield saturates even faster than in low productive grasslands

In the applied context of production-oriented systems, the saturation of yield is expected to occur even faster than in semi-natural grassland communities. This is because forage mixtures can be designed according to the following principles: (1) selecting the best-performing species – generally evaluated in monoculture – for use in mixtures, which ensures high performance at a lower number of species; (2) targeting species that maximize complementarity for desired functions, which enhances performance without the need for many species; (3) selecting species that maintain a stable community composition over time or that respond to adaptive management to ensure this (Lüscher *et al.*, 2011). Indeed, saturation has been demonstrated for forage mixtures (including herbs), where there were no or only marginal yield increases beyond two species in mixtures compared to nine species (Grace *et al.*, 2018), three species compared to eight (Lorenz *et al.*, 2020), three species compared to nine (Sanderson, 2010), four species compared to five (Moloney *et al.*, 2020a), four species compared to six (Grange *et al.*, 2021), and six species compared to 15 (Tracy and Sanderson, 2004a). In line with the trend for rapid saturation, the average beneficial interaction effect in a six-species mixture containing herbs was only marginally greater than that of a four-species grass-legume mixture (Grange *et al.*, 2021). Contradictory results also show increases in yield from two species in a mixture compared to five (Skinner *et al.*, 2004), and from ten species compared to twelve (due to high-yielding lucerne in the twelve-species mixture, Jing *et al.*, 2017).

Importantly, all of these studies focused on yield alone, and, all else being equal, more species diversity is likely to be needed to simultaneously sustain multiple ecosystem functions: (1) other than yield, (2) over longer time scales, and (3) over more variable environmental conditions (Isbell *et al.*, 2011, 2015, 2017; Lefcheck *et al.*, 2015). Although forage yields often do not, or only marginally, differ between high-yielding grass-clover swards and more complex mixtures, intra-annual yield stability (Lorenz *et al.*, 2020), weed suppression (Tracy and Sanderson, 2004b), and resource availability to pollinators (Cong *et al.*, 2020) can be enhanced by higher diversity. Analyses of forage quality from more complex mixtures indicate that although it can be reduced compared to grass-legume stands (Jing *et al.*, 2017), there are multiple examples where complex mixtures have similar or higher forage quality regarding, amongst others, crude protein and digestibility (Grace *et al.*, 2018; Moloney *et al.*, 2020b; Sanderson, 2010). On grazed multi-species swards, dry matter intake, milk production and soil C accumulation were enhanced and N losses reduced compared to more simple swards (reviewed in Jaramillo *et al.*, 2021).

Compared to the scale of the challenge posed by climate change and the demand for more environmentally sustainable farming practice, the science underpinning the potential benefits of multi-species swards should become a stronger focus of future research. There is still plenty to learn about the extent to which mixture benefits are affected by specific combinations of species rather than species richness, management practices (especially grazing), cultivar diversity, as well as variation in environmental conditions, such as soil type, fertility and moisture level. Cultivar selection and adaptive management to promote persistence in mixtures also deserve further attention. It is still not well established whether more, and how many, species in production-oriented grasslands are needed to simultaneously sustain multiple ecosystem functions, such as resistance and resilience to extreme weather events, soil C sequestration or conservation of faunal diversity. There is an indication that trade-offs can occur among different functions (Grange *et al.*, 2022), and that a distinct mixture and management can maximize either production or a variety of ecosystem services related to sustainability (Savage *et al.*, 2021).

For productive species with good forage quality, the range of traits for functional complementarity and drought adaptation is quite limited

A key to higher mixture performance is the targeted and designed combination of species with functional complementary in terms of relevant traits with the aim to increase total resource acquisition and resource use efficiency (Frankow-Lindberg, 2012; Gross *et al.*, 2007; Mason *et al.*, 2020; Tilman *et al.*, 2014). In forage grassland, substantial yield gains can be achieved by the distinct combination of grasses that have efficient resource uptake, in particular of N, and legumes with their ability for symbiotic N_2 fixation (Frankow-Lindberg and Dahlin, 2013; Nyfeler *et al.*, 2011; Pirhofer-Walzl *et al.*, 2013). A further way of achieving complementarity has been identified in the different temporal development of species over years (Finn *et al.*, 2013; Nyfeler *et al.*, 2009) and within the growing season (Husse *et al.*, 2016). By segregating the periods during which species acquire resources, the total biomass production of mixtures is enhanced by more complete resource use over time. Finally, combining species with differing rooting depth allows for increased yields through spatial complementarity in resource uptake (Husse *et al.*, 2017), although the evidence for yield gains by vertical niche differentiation is inconsistent (Hoekstra *et al.*, 2015; Mommer *et al.*, 2010; Oram *et al.*, 2018; Pirhofer-Walzl *et al.*, 2013).

Comparing the complementarity benefits of specific combinations of plant functional traits or functional types (the *identity* of the species present) with those achieved by species richness *per se* (the *number* of species present), effects of identity were generally at least as large or clearly larger than those of richness (Komainda *et al.*, 2020; Mokany *et al.*, 2008; Skinner *et al.*, 2004). This has led several authors to conclude that low to intermediate levels of species richness are sufficient to reach an optimal balance of multiple ecosystem services, but that these species should exhibit functional contrasts in growth habit and phenology (Küchenmeister *et al.*, 2012; Lüscher *et al.*, 2011; Storkey *et al.*, 2015; Tracy and Sanderson, 2004a). Establishing distinct combinations of many forage species with complementary traits to optimize mixture performance in terms of both forage yield and quality seems to be challenging, probably due to the increasing complexity in isolating the effect of a single trait in a mixture with an increasing number of species with multiple traits. Even less is known about how species functional complementarity can help to adapt mixtures to environmental stress caused by severe weather events. Functional complementarity should be relevant under moderate environmental stress and in productive environments, where resource partitioning allows species to reduce competitive interactions (Gross *et al.*, 2007). Conversely, under severe environmental disturbance or nutrient-poor environments, the importance of functional complementarity has been shown to decrease (Mason *et al.*, 2011) and facilitative processes among species should become more relevant (He *et al.*, 2013; Maestre *et al.*, 2009). For example, nurse plants can cast shade and lead to lower transpiration demands of neighbouring plants under heat and drought (Holmgren *et al.*, 2010). However, the evidence for such effects in grasslands is rare (Martorell *et al.*, 2015), and we are not aware of any study demonstrating facilitation under severe drought for forage plants of productive grasslands.

Valuing and applying inter- and intra-specific variability

Valuing intra-specific variability

To assess the relative importance of dehydration avoidance vs tolerance of each species, plants should be compared by combining tests in shallow soils (expression of dehydration tolerance) and deep soils (drought avoidance through water uptake) under different drought intensities. For instance, the dehydration tolerance of cocksfoot is higher than that of tall fescue (*Festuca arundinacea* Schreb.), which primarily relies on dehydration avoidance through an efficient and deep rooting system (Poirier *et al.*, 2012). However, intra-specific variability was comparable or even higher than inter-specific variability for these two perennial grass species under field conditions. Moreover in both species, the dehydration tolerance was greater for the summer-dormant Mediterranean and semi-arid populations than for the non-dormant temperate populations (Volaire, 2008). The intra-specific variability of cocksfoot (Shihan *et al.*, 2022) and perennial ryegrass (Keep *et al.*, 2021) analysed along environmental gradients allowed mapping of the current and future areas for adaptation of Mediterranean populations under a climate scenario. Areas suitable for the expression of, and adaptation to, summer dormancy are predicted to extend northwards under climate change for cocksfoot and the Mediterranean types of perennial ryegrass (Keep *et al.*, 2021; Shihan *et al.*, 2022). Available cultivars of Mediterranean perennial forage species adapted to a dry climate are rare (<2%) (Lelièvre and Volaire, 2009). It is thus required to better identify and valorize the role of this genetic diversity by (i1) tapping into the Mediterranean and semi-arid genetic resources, (2) testing plant material for summer growth potential (summer dormancy levels) possibly associated to dehydration tolerance, and (3) measuring thresholds of dehydration tolerance in standardized conditions, i.e. soil water potential leading to 50% plant mortality (Norton *et al.*, 2016; Volaire *et al.*, 2014) or embolism resistance (Volaire *et al.*, 2018).

Valuing inter-specific variability

To assess how species diversity is used in today's European sown grasslands and to exploit adaptation of forage production in ley-farming systems to drier conditions, a survey was conducted. As an easy-to-derive proxy for plant species' suitability for growth under wet/dry conditions, we chose the Ellenberg indicator value for moisture (F: 'Feuchte' in German; Ellenberg and Leuschner, 2010), and values were derived from the TRY database (Kattge *et al.*, 2020). We are aware that, for more quantitative analyses, Ellenberg indicator values would be too coarse a proxy. Figure 2 compares the means and ranges of F indicator values for distinct sets of plant species. The potential range of F indicator values of grassland species available on the commercial market are shown with the two sets of species 'Central Europe wild types' (116 species) and 'EU common catalogue' (33 species). Both of these sets span a range of seven units ranging from an F value of two (very dry) to nine (wet, often water-saturated). However, if one compares the species' sets of 'recommended varieties' from six countries (where such lists were available), only a small fraction of the diversity potential is currently utilized in sown, production-oriented grasslands. This is indicated by both the small number of species on the recommended lists (often below 10) and the narrow range of their F indicator values (with the exception of CH, Suter *et al.*, 2021a). Surprisingly, the same picture is evident with the 'production mixtures' (multi-species mixtures that are recommended for productive grassland). Even though these mixtures were specifically designed to meet distinct growth conditions (wet or dry but mostly in temperate environments), the means of F indicator values for mixtures (1) differ only little (0.40 units at maximum) and (2) are at about the centre of the total scale (balanced conditions). In addition, (3) the ranges covered by the individual mixture's component species are small, covering at most two units (4 to 6). The only exception were the 'biodiversity mixtures' designed for improvement of biodiversity rather than forage production. They contain a high number of species, differ distinctly in the mixtures' mean F indicator value (1.40 units between wet and dry), and the species within each mixture cover a large range of F indicator values (up to 7 units).

The survey described above strongly suggests that currently only a small part of the inter-specific variability with respect to moisture conditions is utilized in sown grasslands. This may be due to several reasons such as (1) positive diversity effects saturating at low species numbers in the mixture (see above), (2) trade-offs between growth maintenance under moderate drought and plant survival under severe drought (discussed above), between drought resistance and forage quality (e.g. perennial ryegrass vs tall fescue),

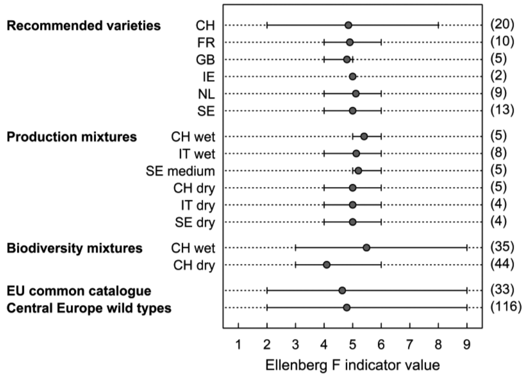

Figure 2. Ellenberg indicator values for moisture (F: 'Feuchte') for different sets of plant species: as national recommended lists of forage species varieties (six countries available), traded forage mixtures for production (three selected countries), mixtures to sustain biodiversity in Switzerland, and the EU common catalogue of agricultural varieties as well as the traded wild type species in Central Europe. Mean (Ÿ) and range displayed; number of species included on the right. 1 = extremely dry, soils that often dry out; 9 = wet, often water-saturated. The species lists, on which the figure is based, can be received from the authors on demand.

or drought resistance and suitability for specific management aims such as grazing (e.g. white clover (*Trifolium repens* L.) vs alfalfa (*Medicago sativa* L.)). An important, more general reason for the relatively small and highly specific set of plant species utilized in intensive forage production is that plant species have to be adapted to (very) high frequencies of defoliation (mowing or grazing), which is a prerequisite for high forage quality and high yields in digestible energy and protein (Huguenin-Elie *et al.*, 2018). Thus, we deem that only species with specific plant traits at the fast end of the 'fast-slow' plant economics spectrum (Reich *et al.*, 1997) seem suitable for production-oriented grasslands in temperate climates. This survey strongly suggests that the shortage of adapted plant material for areas with increasing severe droughts is still insufficiently addressed.

We see two possible strategies to exploit inter-specific diversity to increase drought resistance and resilience of productive grassland at the farm scale. A first strategy would be to increase 'within-field diversity' by designing a more complex mixture that can adapt to different drought conditions. This could be achieved by combining species with distinctly differing moisture requirements (i.e. a mixture with a large range in Figure 2). However, in practice, it is hard to envisage an adequate combination of many species that fulfils the multiple demands of productive grasslands regarding interspecific competition (persistence), complementarity (Suter *et al.*, 2021c) as well as management suitability (for grazing, cutting, silage) (Suter *et al.*, 2021b). A second strategy would be to increase 'among-field diversity' enabling an insurance effect by growing a variety of simpler mixtures (or monocultures), each adapted to different drought conditions (i.e. different mixture means in Figure 2). In this strategy, it might be easier to combine and maintain the persistence of a suitable set of species within each mixture regarding management requirements of the plants, but management of the different fields might be more complex. These strategies can be implemented in sown grassland with the targeted composition of the seed mixture(s) sown and in permanent productive grassland by managing species composition and richness through overseeding, self reseeding, and/or type and intensity of management. Both strategies can be applied not only at the farm scale but also at a regional scale.

Conclusions

Grasslands cover a wide range of global pedo-climatic conditions. They can thrive under harsh growth conditions where other agricultural systems are not economically viable. This demonstrates the considerable adaptation potential of grasslands. The literature reviewed here provides evidence that both intra- and inter-specific diversity have great potential to contribute to the adaptation of permanent and sown productive grassland to drought stress and variability in weather conditions. The choice of a successful strategy to adapt to drought strongly depends on the type of stress. Under severe stress that occurs regularly (as in the Mediterranean summer), drought survival, accompanied by cessation of growth during the stress period, is key to enable fast recovery after the stress has ceased. Under less severe drought and unpredictable weather conditions (as in cool-temperate climates), complementarity is of primary importance in two ways: complementarity in resource use increases ecosystem functioning during the periods of moderate stress and complementarity in water requirements allows for robustness to fluctuating water availability. Nevertheless, both intra- and inter-specific variability seem not sufficiently valued today and undoubtedly are a pillar for adaptation of productive grassland to future conditions. In this context a crucial point is that even a small increase in diversity from monocultures to two- to six-species mixtures already delivers substantial benefits. Diversity of genotypes and/or species on a farm/in a region can be achieved in two ways: either 'within fields' by growing the same complex plant community on all sites, or 'among fields' by growing different (simpler) plant communities on different sites. Both these strategies are easily feasible in sown grassland leys through the targeted composition of the seed mixture. In the long run, they are also applicable in productive permanent grasslands. Future research on the value of diversity in productive grasslands needs to include the interactions of drought stress with other factors (other stresses, soil type, management), and improve measurement of the effect of diversity on multiple ecosystem services (multifunctionality) that include agronomic, environmental and socio-economic responses.

References

Barry K.E., Mommer L., van Ruijven J., Wirth C., Wright A. J., Bai Y., ... and Weigelt A. (2018) The future of complementarity: disentangling causes from consequences. *Trends in Ecology and Evolution* 34, 167-180.

Barkaoui K., Roumet C. and Volaire F. (2016) Mean root trait more than root trait diversity determines drought resilience in native and cultivated Mediterranean grass mixtures. *Agriculture, Ecosystems and Environment* 231, 122-132.

Bristiel P.M., Gillepsie L., Østrem L., Balachowski J.A., Violle C. and Volaire F. (2017) Experimental evaluation of the robustness of the growth–stress tolerance trade–off within the perennial grass *Dactylis glomerata. Functional Ecology* 32, 1944-1958.

Cong W., Dupont Y.L., Søegaard K. and Eriksen J. (2020) Optimizing yield and flower resources for pollinators in intensively managed multi-species grasslands. *Agriculture, Ecosystems and Environment* 302, 107062.

Cong W.-F., Suter M., Lüscher A. and Eriksen J. (2018) Species interactions between forbs and grass-clover contribute to yield gains and weed suppression in forage grassland mixtures. *Agriculture, Ecosystems and Environment* 268, 154-161.

Connolly C., Sebastià M.-T., Kirwan L., Finn J.A., Llurba R., Suter M. ... and Lüscher A. (2018) Weed suppression greatly increased by plant diversity in intensively managed grasslands: A continental-scale experiment. *Journal of Applied Ecology* 55, 852-862.

Craven D., Isbell F., Manning P., Connolly J., Bruelheide H., Ebeling A. ... and Eisenhauer N. (2016) Plant diversity effects on grassland productivity are robust to both nutrient enrichment and drought. *Philosophical Transactions of the Royal Society B – Biological Sciences* 371, 20150277.

Cummins S., Finn J.A., Richards K.G., Lanigan G.J., Grange G., Brophy C., ... and Krol D.J. (2021) Beneficial effects of multi-species mixtures on N_2O emissions from intensively managed grassland swards. *Science of the Total Environment* 792, 148163.

Ellenberg H. and Leuschner C. (2010) *Vegetation Mitteleuropas mit den Alpen*, 6th edn. Ulmer, Stuttgart, Deutschland.

Ergon A., Seddaiu G., Korhonen P., Virkajarvi P., Bellocchi G., ... and Volaire F. (2018) How can forage production in Nordic and Mediterranean Europe adapt to the challenges and opportunities arising from climate change? *European Journal of Agronomy* 92, 97-106.

Finn J.A., Kirwan L., Connolly J., Sebastià M.T., Helgadottir A., Baadshaug O.H., ... and Lüscher A. (2013) Ecosystem function enhanced by combining four functional types of plant species in intensively managed grassland mixtures: a 3-year continental-scale field experiment. *Journal of Applied Ecology* 50, 365-375.

Finn J.A., Suter M., Haughey E., Hofer D. and Lüscher A. (2018) Greater gains in annual yields from increased plant diversity than losses from experimental drought in two temperate grasslands. *Agriculture, Ecosystems and Environment* 258, 149-153.

Frankow-Lindberg B.E. (2012) Grassland plant species diversity decreases invasion by increasing resource use. *Oecologia* 169, 793-802.

Frankow-Lindberg B.E. and Dahlin A.S. (2013) N_2 fixation, N transfer, and yield in grassland communities including a deep-rooted legume or non-legume species. *Plant and Soil* 370, 567-581.

Gillespie L. and Volaire F. (2017) Are winter and summer dormancy symmetrical seasonal adaptive strategies? The case of temperate herbaceous perennials. *Annals of Botany* 119, 311-323.

Grace C., Boland T.M., Sheridan H., Lott S., Brennan E., Fritch R. and Lynch M.B. (2018) The effect of increasing pasture species on herbage production, chemical composition and utilization under intensive sheep grazing. *Grass and Forage Science* 73, 852-864.

Grange G., Brophy C. and Finn J.A. (2022) Drought and plant diversity effects on the agronomic multifunctionality of intensively managed grassland. In: *Grassland at the heart of circular and sustainable food systems* (eds. L. Delaby, R. Baumont, V. Brocard, S. Lemauviel-Lavenant, S. Plantureux, F. Vertes and J.L. Peyraud). *Grassland Science in Europe* 27, 403-405.

Grange G., Finn J.A. and Brophy C. (2021) Plant diversity enhanced yield and mitigated drought impacts in intensively managed grassland communities. *Journal of Applied Ecology* 58, 1864-1875.

Gross N., Suding K.N., Lavorel S. and Roumet C. (2007) Complementarity as a mechanism of coexistence between functional groups of grasses. *Journal of Ecology* 95, 1296-1305.

Hahn C., Lüscher A., Ernst-Hasler S., Suter M. and Kahmen A. (2021) Timing of drought in the growing season and strong legacy effects determine the annual productivity of temperate grasses in a changing climate. *Biogeosciences* 18, 585-604.

Haughey E., Suter M., Hofer D., Hoekstra N.J., McElwain J.C. and Lüscher A. (2018) Higher species richness enhances yield stability in intensively managed grasslands with experimental disturbance. *Scientific Reports* 8, 15047.

He Q., Bertness M.D. and Altieri A.H. (2013) Global shifts towards positive species interactions with increasing environmental stress. *Ecology Letters* 16, 695-706.

Hector A., Schmid B., Beierkuhnlein C., Caldeira M.C., Diemer M., Dimitrakopoulos P.G., ... and Lawton J.H. (1999) Plant diversity and productivity experiments in European grasslands. *Science* 286, 1123-1127.

Hoekstra N.J., Suter M., Finn J.A., Husse S. and Lüscher A. (2015) Do belowground vertical niche differences between deep- and shallow-rooted species enhance resource uptake and drought resistance in grassland mixtures? *Plant and Soil* 394, 21-34.

Hofer D., Suter M., Buchmann N. and Lüscher A. (2017) Nitrogen status of functionally different forage species explains resistance to severe drought and post-drought overcompensation. *Agriculture, Ecosystems and Environment* 236, 312-322.

Hofer D., Suter M., Haughey E., Finn J.A., Hoekstra N.J., Buchmann N. and Lüscher A. (2016) Yield of temperate forage grassland species is either largely resistant or resilient to experimental summer drought. *Journal of Applied Ecology* 53, 1023-1034.

Holmgren M. and Scheffer M. (2010) Strong facilitation in mild environments: the stress gradient hypothesis revisited. *Journal of Ecology* 98, 1269-1275.

Huguenin-Elie O., Delaby L., Klumpp K., Lemauviel-Lavenant S., Ryschawy J. and Sabatier R. (2018) The role of grasslands in biogeochemical cycles and biodiversity conservation. In: *Improving Grassland and Pasture Management in Temperate Agriculture* (eds. A. Marshall and R. Collins). 1-29. Burleigh Dodds Science Publishing, Cambridge, UK.

Husse S., Huguenin-Elie O., Buchmann N. and Lüscher A. (2016) Larger yields of mixtures than monocultures of cultivated grassland species match with asynchrony in shoot growth among species but not with increased light interception. *Field Crops Research* 194, 1-11.

Husse S., Lüscher A., Buchmann N., Hoekstra N.J. and Huguenin-Elie O. (2017) Effects of mixing forage species contrasting in vertical and temporal nutrient capture on nutrient yields and fertilizer recovery in productive grasslands. *Plant and Soil* 420, 505-521.

Isbell F., Adler P.R., Eisenhauer N., Fornara D., Kimmel K., Kremen C., ... and Scherer-Lorenzen M. (2017) Benefits of increasing plant diversity in sustainable agroecosystems. *Journal of Ecology* 105, 871-879.

Isbell F., Calcagno V., Hector A., Connolly J., Harpole W.S., Reich P.B., ... and Loreau M. (2011) High plant diversity is needed to maintain ecosystem services. *Nature* 477, 199-202.

Isbell F., Craven D., Connolly J., Loreau M., Schmid B., Beierkuhnlein C., ... and Eisenhauer N. (2015) Biodiversity increases the resistance of ecosystem productivity to climate extremes. *Nature* 526, 574-577.

Jaramillo D.M., Sheridan H., Soder K. and Dubeux J.C.B. (2021) Enhancing the sustainability of temperate pasture systems through more diverse swards. *Agronomy* 11, 1912.

Jing J., Soegaard K., Cong W.-F. and Eriksen J. (2017) Species diversity effects on productivity, persistence and quality of multispecies swards in a four-year experiment. *PLoS ONE* 12, e0169208.

Kattge J., Bönisch G., Díaz S., Lavorel S., Prentice I.C., Leadley P., … The Nutrient Network (2020) TRY plant trait database – enhanced coverage and open access. *Global Change Biology* 26, 119-188.

Keep T., Sampoux J.P., Barre P., Blanco-Pastor J.L., Dehmer K.J., Durand J.L., … and Volaire F. (2021) To grow or survive: Which are the strategies of a perennial grass to face severe seasonal stress? *Functional Ecology* 35, 1145-1158.

Komainda M., Küchenmeister F., Küchenmeister K., Kayser M., Wrage-Mönnig N. and Isselstein J. (2020) Drought tolerance is determined by species identity and functional group diversity rather than by species diversity within multi-species swards. *European Journal of Agronomy* 119, 126116.

Kreyling J., Dengler J., Walter J., Velev N., Ugurlu E., Sopotlieva D., … and Jentsch A. (2017) Species richness effects on grassland recovery from drought depend on community productivity in a multisite experiment. *Ecology Letters* 20, 1405-1413.

Küchenmeister F., Küchenmeister K., Wrage N., Kayser M. and Isselstein J. (2012) Yield and yield stability in mixtures of productive grassland species: Does species number or functional group composition matter? *Grassland Science* 58, 94-100.

Lefcheck J.S., Byrnes J.E.K., Isbell F., Gamfeldt L., Griffin J.N., Eisenhauer N., … and Duffy J.E. (2015) Biodiversity enhances ecosystem multifunctionality across trophic levels and habitats. *Nature Communications* 6, 6936.

Lelièvre F. and Volaire F. (2009). Current and potential development of perennial grasses in rainfed Mediterranean farming systems. Crop Science 49, 2371-2378.

Loreau M., Barbier M., Filotas E., Gravel D., Isbell F., Miller S.J., … and Thompson P.L. (2021). Biodiversity as insurance: from concept to measurement and application. *Biological Reviews* 96, 2333-2354.

Lorenz H., Reinsch T., Kluβ C., Taube F. and Loges R. (2020) Does the admixture of forage herbs affect the yield performance, yield stability and forage quality of a grass clover ley? *Sustainability* 12, 5842.

Lüscher A., Soussana J.F. and Huguenin-Elie O. (2011) Role and impacts of legumes in grasslands for high productivity and N gain from symbiotic N_2 fixation. In: Lemaire G., Hodgson J. and Chabbi A. (eds.). *Grassland Productivity and Ecosystem Services.* CABI, Wallingford, UK, pp. 101-107.

Lüscher, A., Mueller-Harvey, I., Soussana, J.F., Rees, R.M. and Peyraud, J.L. (2014) Potential of legume-based grassland-livestock systems in Europe: A review. *Grass and Forage Science* 69, 206-228.

Maestre F.T., Callaway R.M., Valladares F. and Lortie C.J. (2009) Refining the stress-gradient hypothesis for competition and facilitation in plant communities. *Journal of Ecology* 97, 199-205.

Martorell C., Almanza-Celis C.A.I., Pérez-García E.A. and Sánchez-Ken J.G. (2015) Co-existence in a species-rich grassland: Competition, facilitation and niche structure over a soil depth gradient. *Journal of Vegetation Science* 26, 674-685.

Mason N.W., de Bello F., Dolezal J. and Leps J. (2011) Niche overlap reveals the effects of competition, disturbance and contrasting assembly processes in experimental grassland communities. *Journal of Ecology* 99, 788-796.

Mason N.W.H., Orwin K.H., Lambie S., Waugh D., Pronger J., Carmona C.P. and Mudge P. (2020) Resource-use efficiency drives overyielding via enhanced complementarity. *Oecologia* 193, 995-1010.

Michalet R. and Pugnaire F.I. (2016) Facilitation in communities: underlying mechanisms, community and ecosystem implications. *Functional Ecology* 30, 3-9.

Mokany K., Ash J. and Roxburgh S. (2008) Functional identity is more important than diversity in influencing ecosystem processes in a temperate native grassland. *Journal of Ecology* 96, 884-893.

Moloney T., Sheridan H., Grant J., O'Riordan E.G. and O'Kiely P. (2020a) Yield of binary- and multi-species swards relative to single-species swards in intensive silage systems. *Irish Journal of Agricultural and Food Research* 59, 12-26.

Moloney T., Sheridan H., Grant J., O'Riordan E.G. and O'Kiely P. (2020b) Conservation efficiency and nutritive value of silages made from grass-red clover and multi-species swards compared with grass monocultures. *Irish Journal of Agricultural and Food Research* 59, 150-166.

Mommer L., van Ruijven J., de Caluwe H., Smit-Tiekstra A.E., Wagemaker C.A.M., Ouborg N.J., … and de Kroon H. (2010) Unveiling below-ground species abundance in a biodiversity experiment: a test of vertical niche differentiation among grassland species. *Journal of Ecology* 98, 1117-1127.

Naeem S., Thompson L.J., Lawler S.P., Lawton J.H. and Woodfin R.M. (1994) Declining biodiversity can alter the performance of ecosystems. *Nature* 368, 734-737.

Norton M., Malinowski D. and Volaire F. (2016) Plant drought survival under climate change and strategies to improve perennial grasses. A review. *Agronomy for Sustainable Development* 36, 29.

Norton M.R., Lelievre F. and Volaire F. (2006a) Summer dormancy in *Dactylis glomerata* L., the influence of season of sowing and a simulated mid-summer storm on two contrasting cultivars. *Australian Journal of Agricultural Research* 57, 565-575.

Norton M.R., Volaire F. and Lelievre F. (2006b) Summer dormancy in *Festuca arundinacea* Schreb., the influence of season of sowing and a simulated mid-summer storm on two contrasting cultivars. *Australian Journal Agricultural Research* 57, 1267-1277.

Nyfeler D., Huguenin-Elie O., Suter M., Frossard E. and Lüscher A. (2011) Grass-legume mixtures can yield more nitrogen than legume pure stands due to mutual stimulation of nitrogen uptake from symbiotic and non-symbiotic sources. *Agriculture Ecosystems and Environment* 140, 155-163.

Nyfeler D., Huguenin-Elie O., Suter M., Frossard E., Connolly J. and Lüscher A. (2009) Strong mixture effects among four species in fertilized agricultural grassland led to persistent and consistent transgressive overyielding. *Journal of Applied Ecology* 46, 683-691.

Oram N.J., Ravenek J.M., Barry K.E., Weigelt A., Chen H., Gessler A., ... and Mommer L. (2018) Below-ground complementarity effects in a grassland biodiversity experiment are related to deep-rooting species. *Journal of Ecology* 106, 265-277.

Orlowsky B. and Seneviratne S.I. (2012) Global changes in extreme events: regional and seasonal dimension. *Climatic Change* 110, 669-696.

Pirhofer-Walzl K., Eriksen J., Rasmussen J., Høgh-Jensen H., Søegaard K. and Rasmussen J. (2013) Effect of four plant species on soil ^{15}N access and herbage yield in temporary agricultural grasslands. *Plant and Soil* 371, 313-325.

Poirier M., Durand J.L. and Volaire F. (2012) Persistence and production of perennial grasses under water deficits and extreme temperatures: importance of intraspecific vs. interspecific variability. *Global Change Biology* 18, 3632-3646

Reich P.B., Walters M.B. and Ellsworth D.S. (1997) From tropics to tundra: Global convergence in plant functioning. *Proceedings of the National Academy of Sciences of the USA* 94, 13730-13734.

Sanderson M. (2010) Nutritive value and herbage accumulation rates of pastures sown to grass, legume, and chicory mixtures. *Agronomy Journal* 102, 728-733.

Sanderson M.A., Skinner R.H., Barker D.J., Edwards G.R., Tracy B.F. and Wedin D.A. (2004) Plant species diversity and management of temperate forage and grazing land ecosystems. *Crop Science* 44, 1132-1144.

Savage J., Woodcock B.A., Bullock J.M., Nowakowski M., Tallowin J.R.B. and Pywell R.F. (2021) Management to support multiple ecosystem services from productive grasslands. *Sustainability* 13, 6263.

Schaub S., Buchmann N., Lüscher A. and Finger R. (2020) Economic benefits from plant species diversity in intensively managed grasslands. *Ecological Economics* 168, 106488.

Scherber C., Eisenhauer N., Weisser W.W., Schmid B., Voigt W., Fischer M., ... and Tscharntke T. (2010) Bottom-up effects of plant diversity on multitrophic interactions in a biodiversity experiment. *Nature* 468, 553-556.

Shihan A., Barre P., Copani V., Kallida R., Østrem L., Testa G., ... and Volaire F. (2022) Induction and potential role of summer dormancy to enhance persistence of perennial grasses under warmer climates. *Journal of Ecology* (in press).

Skinner R.H., Gustine D.L. and Sanderson M.A. (2004) Growth, water relations, and nutritive value of pasture species mixtures under moisture stress. *Crop Science* 44, 1361-1369.

Storkey J., Doering T., Baddeley J., Collins R., Roderick S., Jones H. and Watson C. (2015) Engineering a plant community to deliver multiple ecosystem services. *Ecological Applications* 25, 1034-1043.

Suter D., Frick R. and Hirschi H.U. (2021a) Liste der empfohlenen Sorten von Futterpflanzen 2021-2022. *Agroscope Transfer* 360, 16 pp.

Suter D., Rosenberg E. and Frick R. (2021b) *Standardmischungen für den Futterbau. Revision 2021-2024.* AGFF, Zürich, Switzerland, 16 pp.

Suter M., Connolly J., Finn J.A., Loges R., Kirwan L., Sebastià M.-T. and Lüscher A. (2015) Nitrogen yield advantage from grass-legume mixtures is robust over a wide range of legume proportions and environmental conditions. *Global Change Biology* 21, 2424-2438.

Suter M., Hofer D. and Lüscher A. (2017) Weed suppression enhanced by increasing functional trait dispersion and resource capture in forage ley mixtures. *Agriculture, Ecosystems & Environment* 240, 329-339.

Suter M., Huguenin-Elie O. and Lüscher A. (2021c) Multispecies for multifunctions: combining four complementary species enhances multifunctionality of sown grassland. *Scientific Reports* 11, 3835.

Tilman D., Isbell F. and Cowles J.M. (2014) Biodiversity and ecosystem functioning. *Annual Review of Ecology, Evolution, and Systematics* 45, 471-493.

Tilman D., Lehman C.L. and Thomson K.T. (1997) Plant diversity and ecosystem productivity: Theoretical considerations. *Proceedings of the National Academy of Sciences of the USA* 94, 1857-1861.

Tilman D., Reich P.B., Knops J., Wedin D., Mielke T. and Lehman C. (2001) Diversity and productivity in a long-term grassland experiment. *Science* 294, 843-845.

Tracy B.F. and Sanderson M.A. (2004a) Productivity and stability relationships in mowed pasture communities of varying species composition. *Crop Science* 44, 2180-2186.

Tracy B.F. and Sanderson M.A. (2004b) Forage productivity, species evenness and weed invasion in pasture communities. *Agriculture, Ecosystems and Environment* 102, 175-183.

Volaire F. (2008) Plant traits and functional types to characterise drought survival of pluri-specific perennial herbaceous swards in Mediterranean areas. *European Journal of Agronomy* 29, 116-124.

Volaire F. (2018) A unified framework for plant drought adaptive strategies: across scales and disciplines. *Global Change Biology* 24, 2929-2938.

Volaire F. and Norton M. (2006) Summer dormancy in perennial temperate grasses. *Annals of Botany* 98, 927-933.

Volaire F., Barkaoui K. and Norton M. (2014) Designing resilient and sustainable grasslands for a drier future: Adaptive strategies, functional traits and biotic interactions. *European Journal of Agronomy* 52, 81-89.

Volaire F., Lens F., Cochard H., Xu H., Chacon Doria L., Bristiel P., … and Picon-Cochard C. (2018) Embolism and mechanical resistances play a key-role in dehydration tolerance of a perennial grass *Dactylis glomerat*a L.. *Annals of Botany* 122, 325-336.

Weisser W.W., Roscher C., Meyer S.T., Ebeling A., Guangjuan A., Allan E., … and Tscharntke T. (2017) Biodiversity effects on ecosystem functioning in a 15-year grassland experiment: Patterns, mechanisms, and open questions. *Basic and Applied Ecology* 23, 1-73.

White R.P., Murray S. and Rohweder M. (2000) *Pilot Analysis of Global Ecosystems: Grassland Ecosystems*. World Resources Institute, Washington, DC, USA.

Wright A.J., Wardle D.A., Callaway R. and Gaxiola A. (2017) The overlooked role of facilitation in biodiversity experiments. *Trends in Ecology and Evolution* 32, 383-390.

Yates S., Jaskune K., Liebisch F., Nagelmuller S., Kirchgessner N., Kölliker R., … and Studer B. (2019) Phenotyping a dynamic trait: leaf growth of perennial ryegrass under water limiting conditions. *Frontiers in Plant Science* 10, 9.

Zhao Y., Gao J.H., Kim J.I., Chen K., Bressan R.A. and Zhu J.K. (2017) Control of plant water use by ABA induction of senescence and dormancy: an overlooked lesson from evolution. *Plant and Cell Physiology* 58, 1319-1327.

Diversification increases the resilience of European grassland-based systems but is not a one-size-fits-all strategy

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Abstract

Diversification of grassland-based systems is highly valued in agroecology, organic farming and other forms of regenerative agriculture. For Mediterranean, mountain and lowland areas, we illustrate that diversification of grassland types, livestock, products and farm labour allows coping with market, climatic and workforce-related risks. However, diversification is not a one-size-fits-all strategy. Farmers' technical skills and ability to re-organize and monitor the system must be considered to avoid ineffectiveness of the diversified system. Moreover, it is essential to account for site-specific conditions so that the ecological processes to be optimized can provide the expected benefits. Diversification occurs on different levels, from grassland and feed resources to the entire farm activity. There may be trade-offs among these different levels impairing grassland ecosystem services. For instance, if diversification of farm activities dilutes the workforce, simplified grassland management can lead to the loss of vegetation communities of high ecological value. In contrast, case-adapted diversification benefits from local opportunities, available resources and external supports to secure the system and favour sustainable resource management. Thereby, diversification preserves grassland ecosystem services and enhances farm socio-economic resilience to withstand perturbations.

Keywords: agroecology, biodiversity, ecosystem services, mixed grazing, site-adapted management, trade-offs

Introduction

Over long periods of time, grassland-based systems were adapted to local conditions and external inputs were limited. Therefore these systems were inevitably diverse and self-supplied. In the 20th century, agricultural industrialization and the global market led to systems intensification and specialization. In lowlands and on medium-altitude plateaux, specialization has increased the predominance of high-yielding fertilized swards and grass-legume mixtures over less intensively managed grasslands. Although productive, these high-yielding grassland-based systems are increasingly vulnerable to climate change (Melts *et al.*, 2018; Stampfli *et al.*, 2018). In uplands, where animals graze on steep slopes and sometimes in more remote areas, grasslands are threatened by partial abandonment due to lack of economic profitability and high opportunity costs.

Nowadays, the challenge is to replace the old paradigm based on simplification and standardization of production systems for optimizing productivity per unit of human labour, with a new paradigm emphasizing diversification at field, farm, and landscape scale to optimize productivity per unit of natural resource and provide a number of ecosystem services (Lemaire *et al.*, 2015). Diversification is also widely employed by traditional livestock systems (López-i-Gelats *et al.*, 2011). Dumont *et al.* (2020) have demonstrated that production and ecosystem services provided by animal production systems are grounded in grassland type and feed resource diversity, herd variability (inter-individual, inter-breed or inter-specific) and farm-scale interactions. Herd and product diversification also imply changes in sales management and in work organization (Martin *et al.*, 2020). System redesign can even lead to a diversification of farm activities beyond the food-producing role of agriculture (López-i-Gelats *et al.*, 2011).

Diversification has the potential to reduce the vulnerability of grassland-based systems. Vulnerability not only depends on the exposure and sensitivity to risks, but also on the ability to adapt to or recover from perturbations (Smit and Wandel, 2006). Walker *et al.* (2004) defined resilience as the capacity of a system to absorb perturbations and reorganize while undergoing changes to maintain its function. Darnhofer (2014) has discussed that resilience covers the buffer, adaptive and transformative capabilities of any system. Buffer capability denotes the ability of a system to assimilate a perturbation without changing its structure or function; adaptive capability that of temporarily adjusting to change while staying in the current stability domain; and transformative capability implies transition to a new system. Biggs *et al.* (2012) proposed a hump-shaped relationship between the level of system diversity and the resilience of ecosystem services. This suggests that there is a theoretical diversity optimum, below which low diversity limits the buffer and adaptive capabilities of the system. Beyond the optimum, system resilience would be compromised by being too complex. Farmers become unable to monitor and integrate all possibilities and interconnections into their analysis and consequently, the system will 'stagnate'.

Building on Biggs *et al.* (2012), we apply the hump-shaped relationship between diversification and resilience to grassland-based systems and develop a critical understanding of diversification on different levels. We discuss how the diversification of grassland types, livestock, products and farm labour can improve resilience in grassland-based systems. Meanwhile, we identify diversification trade-offs and risks leading to poor grassland management and therefore to a decrease in production and ecosystem services resilience. Finally, we briefly discuss how private insurance and public support are complementary levers to consider for achieving resilience. Some of the illustrative examples in this article emphasize the specificities of mountain and Mediterranean systems. These areas face environmental constraints that limit grassland productivity, so that grassland-based systems represent the main, and sometimes only, agricultural option. These marginal areas can thus have a pioneering role in developing concepts of diversification, which can also be applied in lowlands. Conversely, we hardly deal with integrated crop-livestock diversification, where literature is abundant (Lemaire *et al.*, 2015; Regan *et al.*, 2017), nor with landscape-scale diversification (Fahrig *et al.*, 2011).

Diversification of grassland types, feed resources and grassland management

Plant species diversity can increase grassland resilience (Lüscher *et al.*, 2022), for example by buffering drought events (e.g. Grange *et al.,* 2021) due to a broad range of plant traits. However, the diversity of a single meadow or pasture can hardly buffer all potential perturbations to which it is exposed. Diversification of grassland types is therefore needed to further increase resilience. In intensively managed grazing systems, grassland type diversification can be achieved by cultivating drought-resistant mixtures (Lüscher *et al.*, 2022), including for instance sainfoin (Kölliker *et al.*, 2017) on some fields of a farm. These resistant mixtures are not the most productive ones in average years, but they reduce the variability of biomass yield in dry years and thereby increase farm resilience. French beef or sheep grazing farmers also diversified forage resources by cultivating additional fodder crops such as corn silage and cover crops. It allowed them to increase and stabilize the quantity of fodder harvested per livestock unit, but farm income was neither higher nor less variable due to higher stocking density and production costs (Mosnier *et al.*, 2013). Diversification of fodder resources thus does not necessarily increase farm resilience but should be considered as part of global risk management at farm scale. Fodder type diversification has further advantages and can benefit animal performance as the result of improved pasture nutritive value, of increased daily intake when animals are offered a more diversified diet, and of parasite control thanks to tannin-rich plant species, such as sainfoin (Dumont *et al.*, 2020).

Another possible source for diversification could be the integration of 'poor agronomic value' grasslands on wet areas (which increases system resilience in dry years) or shallow soils (useful in wet years). Conservation of such semi-natural grasslands with a generally high ecological value would thus also generate benefits for fodder system resilience. In Mediterranean silvopastoral systems, grassland management creates a balanced mix of trees, species-rich pastures and marginal habitats improving animal performance and welfare (Moreno *et al.*, 2018). Silvopastoral systems also preserve and increase biodiversity at farm and landscape scales, especially in transhumant systems. Trees and shrubs providing fodder and shade, favour the adaptation of these ecosystems to climate change and thereby increase their resilience. For instance, leaves and acorns of oak trees are used as forage supplement in Iberian dehesas. In Mediterranean wood pastures, livestock benefits from browsing pollard trees, shrubs or pruned branches. Releasing domestic pigs in wooded areas (so-called pannaging) is still practised for fattening pigs with acorns, beechmast, chestnuts or other nuts in dehesas and montados. Moreover, the introduction of trees into specialized crops and farming systems increases soil carbon sequestration and creates microclimates under the canopy, which limit water evaporation and offer insolation to plants and livestock. This diversification of radiation, micro-topographic parameters (such as slope, exposure, convexity and concavity) and soil parameters (such as pH) enhances the diversity of grassland types (Franca *et al.*, 2016).

Beyond the diversification of fodder system, adapting management intensity can enhance grassland resilience. For instance, Vogel *al*. (2012) found that resilience of grasslands to summer drought depends on management intensity. The higher the mowing frequency in years of drought events, the lower the biomass yield in the subsequent year. Nyfeler *et al.* (2011) demonstrated that if grasslands are heavily fertilized, nitrogen yield increases. However, the additional yield is not provided by the grassland ecosystem itself, but by the external input alone, leading to a decrease in nitrogen use efficiency. Hence, a decrease in land-use intensity and of mineral fertilization could benefit the resilience of grasslands (Melts *et al.*, 2018; Stampfli *et al.*, 2018). Diversification towards site-adapted management allows for the use of each grassland type at an appropriate intensity and provides the benefits of supporting and regulating services. Grassland diversification often comes along with lower intensity of use, which may reduce forage quality of some pastures and meadows. This trade-off may be addressed by grazing lower yielding animals, such as non-lactating dairy cows, on grasslands managed at lower intensity. Differentiated grassland types under site-adapted management can be harvested at different dates of the vegetation period. This reduces farmer's workload at peak times, and permits the use of agricultural machinery of lower volume and price. Finally, diversification of the grassland mosaic increases landscape aesthetics, which improves the perception of grazing systems by society.

Diversification of grazing livestock and herd management

Due to their nutritional requirements and morphological and digestive capacities, cattle, sheep, horses and goats have contrasting abilities to graze on short swards, digest roughage and detoxify forb secondary compounds. Mixed grazing with different livestock species can thus increase overall pasture use, due to the complementary of feeding niches and grazing facilitation processes (Dumont *et al.*, 2012; Martin *et al.*, 2020). Mixed grazing can sometimes produce the most species-rich and structurally diverse swards (Loucougaray *et al.*, 2004) and enhance levels of ecosystem services provided by grassland-based systems (Wang *et al.*, 2019). Animal growth usually benefits from mixed grazing. D'Alexis *et al.* (2014) reported enhanced lamb growth and meat production per hectare in mixed grazing systems of sheep and cattle. Jerrentrup *et al.* (2020) confirmed this result and reported an additional increase in suckler cow weight gain under mixed grazing. The pattern of animal growth according to sheep-cattle ratio is hump-shaped with a plateau, which offers a wide range of ratios resulting in maximal, or quasi maximal, animal performances. For this reason, fine-tuning the sheep-cattle ratio is not needed to take full advantage of mixed grazing. Thus, the need for continuous monitoring and corrective adjustments of livestock species ratio is eliminated and leaves the farmer free to focus on other tasks (Joly *et al.*, 2021).

Due to dilution effects, mixed grazing is also an efficient strategy to reduce parasitic nematode infection in small ruminants (Marley *et al.*, 2006) and horses (Forteau *et al.*, 2020), which is likely to decrease treatment frequency, associated drug resistance and veterinary costs, and to reduce the negative environmental side effects of drug metabolites on dung beetle assemblages (Sands and Wall, 2018). Thanks to their two sets of incisors, horses graze close to the ground and maintain stable sward patches of high nutritive value (Dumont *et al.*, 2012). Cattle are excluded from these short lawns where they cannot meet their daily requirements, and switch to tall grass areas where they graze close to horse dungs and reduce sward parasite burden (Figure 1A). Thereby, feed resources are used more equally. However, an alternate stocking of cattle and horses grazing together on a mesophile grassland provided animals with high-quality regrowth on the short patches. Consequently, cattle avoided tall areas with reproductive and dead grass, which limited their consumption of strongyle larvae near the patches of horse dung (Figure 1B). This can explain why no significant benefits of mixed grazing on horse parasitism was measured (Fleurance *et al.*, 2022), and illustrates that co-grazing requires appropriate management to provide its expected benefits.

Mixed farming systems are gaining interest to reduce inputs and production costs, and as a risk management strategy. Recent surveys in cattle-sheep farms of the French Massif central have confirmed that farmers mention the stability of farm economic performance and an efficient use of grassland resources as the main benefits of mixed grazing systems (Mugnier *et al.*, 2021). Mosnier *et al.* (2022) simulated that mixed farms have fewer work peaks, lower global warming potential and nitrogen balance, lower production costs and higher and more stable net incomes than specialized farms. In the case of a sheep-cattle mix, sheep production benefits more from the presence of cattle on the farm than cattle benefit from the presence of sheep, which may encourage sheep farmers to diversify more than a beef farmer. However, farm diversification may also reduce the performance of the production process due to an increasing complexity of farming systems (De Roest *et al.,* 2018) and to a limited time that farmers can spend on each activity. Some mixed organic beef-sheep farmers indeed justified the low performance of their sheep flock because it was not their priority (Mosnier and Moufid, 2021). Modifying the ewe-cow

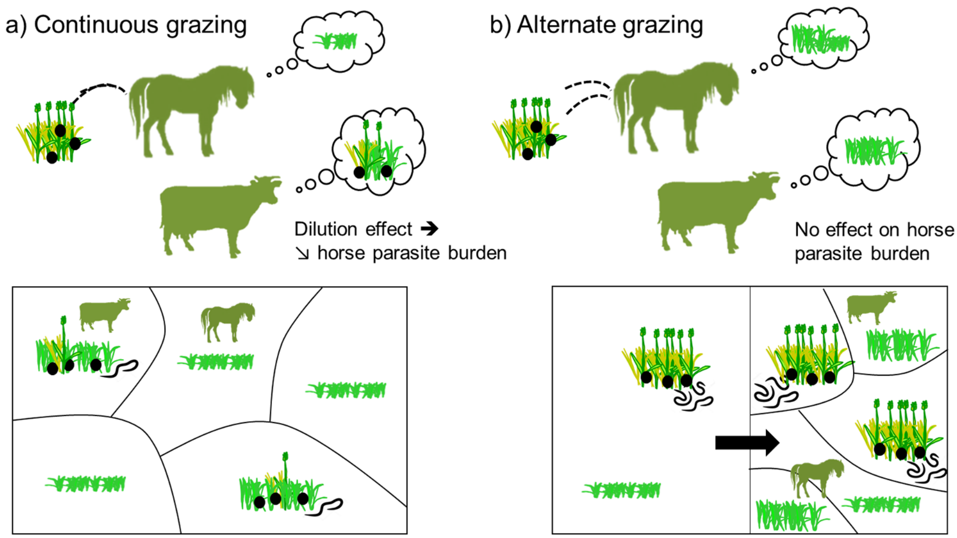

Figure 1. Contrasting effects of mixed grazing by cattle and horses on horse parasite burden and plot use according to pasture management: (A) continuous grazing (adapted from Forteau *et al.*, 2020); (B) alternate grazing between two subplots (Fleurance *et al.*, 2022). Under continuous grazing, cattle were excluded from short lawns and switch to tall areas where they graze close to horse dung, which was not the case under alternate grazing. Horse dung and nematode larvae are represented in each plot.

ratio, usually by adjusting the number of ewes, is the main adaptation leverage used by mixed farmers to cope with market, climatic and workforce-related risks in the short and medium term (Mugnier *et al.*, 2021; Nozières *et al.*, 2011). Although farmers usually consider high workload as a constraint, they also mentioned the pleasure of varied work and the flexibility of work organization reducing overlaps between calving and lambing periods, among the advantages of mixed farming (Mugnier *et al.*, 2021). In contrast, the benefits of mixed grazing for reducing parasitic nematode infection were not mentioned by these farmers, who instead feared disease transmission among species. Also in mixed cattle-horse systems, two thirds of the mixed farmers surveyed by Forteau *et al.* (2020) were not aware of the benefits of mixed grazing for parasite nematode management.

Beneficial diversification of livestock type does not necessarily imply grazing of different species simultaneously. Case-adapted management can also include dual-purpose breeds or cross breeding to ideally balance productivity and a sustainable use of available resources (Phocas *et al.*, 2016). Moreover pasture management and resilience can be improved by keeping a 'service herd' of a hardy breed. These low-productive animals still show a number of adaptive traits. Light and big-footed Highland cattle cause less pressure on the ground. Thereby, they increase grassland resistance against erosion and allow a site-adapted use of steep slopes (Figure 2A), wet pastures and shallow soils. Pauler *et al.* (2020b) showed that low-productive Highland cattle consume more thistles and woody plants (Figure 2C) than high-productive cattle. Thereby, Highland cattle increase plant species richness (Figure 2B), pasture quality and reduce workload needed for pasture management (Pauler *et al.*, 2019). Similar findings were presented for low-productive Engadine sheep, consuming green alder shrubs in subalpine-systems most efficiently and thereby hindering shrub encroachment and its numerous negative environmental effects (Pauler *et al.,* 2022). Under a low-nutritive value diet, so-called low-productive cattle gain more weight than high-productive cattle (Pauler *et al.*, 2020a). Consequently, a diversification of livestock increases farm resilience in years of low forage quality. Finally, though the low output and the additional workload of managing a service herd may prevent farmers from diversifying their herd, the products of a service herd can benefit from the system's 'positive image' and be directly sold on-farm.

Product and farm labour diversification

Transformative changes that enhance the resilience of pasture-based ruminant systems to market price fluctuations can include product diversification, development of an on-farm processing enterprise and short-distribution channels (Martin *et al.*, 2020). Beyond product diversification, transformative changes can diversify farm activities beyond the food-producing role of agriculture (e.g. agritourism) and the full-time dedication of family members to farming activity (López-i-Gelats *et al.*, 2011).

Product diversification can be achieved by adding pigs or poultry with a short production cycle, to ruminant livestock systems. This diversification allows a more regular cash inflows and more stable incomes as cattle and monogastric meats are sold onto different markets. Moreover, offering a diversified range of product for sale also facilitates the use of short supply channels and was shown to enhance the demand for local beef and pig meat and consumers' willingness to pay in rural areas of central France (Vollet and Saïd, 2018). Combining monogastric and cattle production can thus be seen as part of the securization strategy of farmers thereby indirectly enhancing farm resilience. Among 17 organic mixed-species farms from French Massif central and Occitany, the two economically most efficient farms associated beef cattle to monogastrics and had a processing enterprise on-farm (Steinmetz *et al.*, 2021). Conversely, beef systems with large monogastric production units that sell the animals to cooperatives were highly dependent on external inputs, which led to high excess of nitrogen per hectare without gaining economic efficiency. Due to this high dependence on external inputs, pig production did not reduce income variability compared to specialized cattle farms in this case (Mosnier and Moufid, 2021). Diversification also enhances the need for new technical skills and sometimes high initial investments for

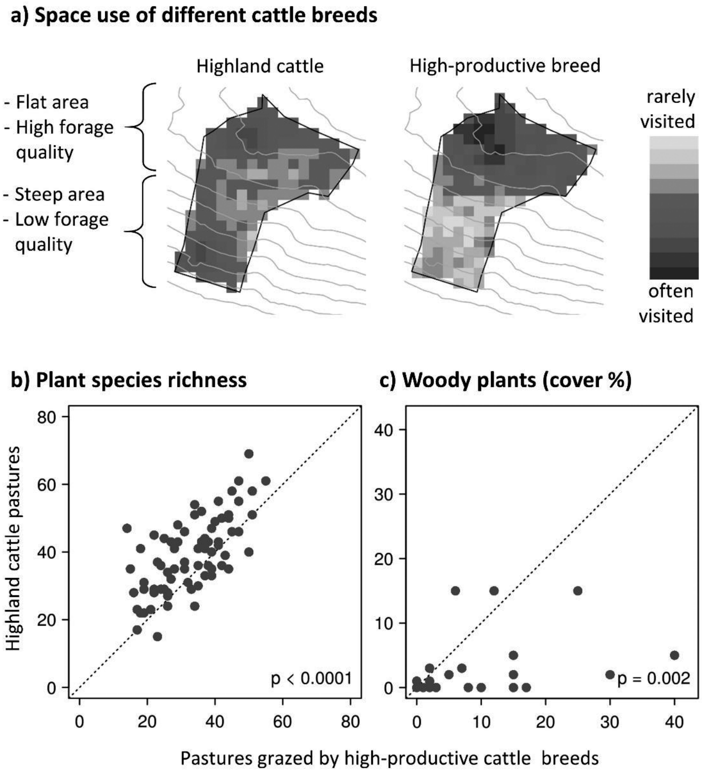

Figure 2. Distribution of two cattle breeds in Swiss upland pastures and its consequences on vegetation community. (A) Low-productive Highland cattle used the available resources on steep pastures of heterogeneous vegetation more evenly than the high-productive breed (Pauler *et al.*, 2020a). (B) Thereby, they increased plant species richness, as measured at 50 paired pastures along a broad environmental gradient, and (C) reduced shrub encroachment, leading to higher pasture resilience. *P*-values represent the significance of the breed effect in a generalized mixed-effect model (Pauler *et al.*, 2019).

animal housing and waste management that can act as strong inhibitors of farm diversification (Dumont *et al.*, 2020). Moreover, there is a risk that farmers become less concerned with grassland management if cereals and pulses are available on the farm. This could in turn negatively impact grassland nutritive value and biodiversity.

Another illustrative example of feed resources, product and farm labour diversification was reported by Vagnoni and Franca (2018). A dairy sheep farm located in the hilly territory of Sardinia diversified their production from an intensive foraging system based on temporary grasslands and irrigated grain-cereal crops towards semi-natural grasslands by exploiting the germination of native seedbank and over-seeding of annual self-reseeding legumes and grasses. Sheep breed and farm stocking density were not modified. This diversification toward site-adapted fodder production reduced production cost significantly, increased the share of species-rich grasslands, reduced environmental impact (-43% kg CO_2-eq per hectare of utilized agricultural area) and increased soil C sequestration by 63% (Arca *et al.*, 2021). With the aim of creating more added value, the whole milk produced on-farm was used to produce Pecorino di Osilo cheese, which is included in the ministerial list of typical Italian agri-food products. This shift in the product sold led to farm labour diversification, as it required know-how on cheese making and marketing (direct selling), and highlighted the role of young family members employed in the renewed farming system.

Diversification is all the more important in regions prone to pastoral abandonment (López-i-Gelats *et al.*, 2011). The particular diversification strategy realized on a farm depends on access and allocation of pastoral resources, such as land, labour, livestock, and capital. Labour diversification outside agriculture, such as agritourism or off-farm employment, can be an attractive option for pastoral households in scenic landscapes. On the one hand, this kind of diversification disconnects farm income from climatic and economic risks (López-i-Gelats *et al.*, 2011). On the other hand, it may increase land abandonment if the additional income is not reinvested into pastoral farming activities. There is also a risk that due to the additional workload outside agriculture, marginal grasslands of high ecological value are poorly managed and finally lost. In the Catalan Pyrenees, there is a gradual transition from sheep to cattle and even horse production due to the low economic profitability of sheep farming. Sheep- and cattle-grazing preserve the species-rich *Arrhenatherion elatioris* community that is typical of cut (or cut and grazed) meadows (Figure 3). Extensive horse production associated with abandonment of mowing requires very little workforce (López-i-Gelats *et al.*, 2015) and are part of a simplifying management regime, which is triggering a transition away form the typical *Arrhenatherion elatioris* community (Figure 3). Thus, while the diversification of labour outside agriculture may enhance the resilience of pastoral households, it also removes resources traditionally devoted to the livestock farming activity and thus threatens grasslands of high ecological value.

External supports to foster system diversification

As diversification can reach its limits, external supports must be considered. Public support and private insurance are important complementary levers to be considered to help farmers achieve sustainable and resilient grassland-based production. Public support such as environmental payments could reduce farm vulnerability by increasing farm income in all situations. In addition, a public safety net compensates farmers in cases of extreme events (climate, market or animal health issues) in several EU countries, which reduces the risk of significant economic loss. However, particular attention must be paid to the conditions of these payments to prevent them becoming a disincentive to farmers from managing normal risks themselves (Tangerman, 2011), and to encourage specialized, capital- or input-intensive systems. For instance, the per hectare and per animal head subsidies did not increase diversification but favoured farm enlargement and simplification of practices (Veysset *et al.*, 2014), which may result in poor grassland management.

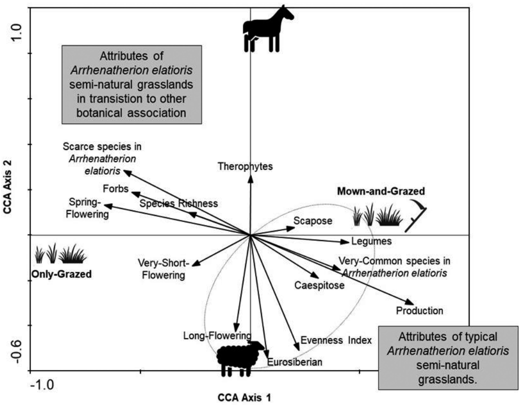

Figure 3. Plot scores for the first two axes of a canonical correspondence analysis for species' composition and vegetation community structure of *Arrhenatherion elatioris* semi-natural grasslands of the Catalan Pyrenees under simplification practices resulting from the diversification of pastoral household labour outside agriculture: horse grazing and abandonment of mowing (adapted from López-i-Gelats *et al.*, 2015). Arrows represent different botanical parameters.

In the most recent narrative of the Common Agricultural Policy 'CAP for public goods', public subsidies will target more specifically the habitat and cultural services provided by semi-natural grasslands, as they fulfil important functions for biodiversity, recreation opportunities, and scenic and cultural landscapes (e.g. open grassland in Swiss silvopastoral landscapes: Huber *et al.*, 2013). Moreover, European agriculture receives subsidies that encourage livestock farmers to diversify their sources of income to limit further land abandonment in upland areas and retain people in more remote regions (e.g. Pardini and Nori, 2011).

Farmers are also encouraged to take out private insurance. Multi-peril grassland insurance scheme can also reduce the variability and the probability of low farm income (Finger and Calanca, 2011). Conversely, many farmers are reluctant to subscribe to such insurances as they find them too costly and prefer to rely on on-farm options and the public safety net. As the cost of self-insurance increases for important and rare losses (Mosnier, 2015), insurances could be an interesting option (Clarke and Dercon, 2009), particularly if the public safety net is reduced. However, they should not be considered without assessing beforehand the opportunities provided by the diversification of grassland types, livestock, products and farm labour on each farm.

Conclusions

Diversification on different levels allows addressing risks of different nature. Numerous benefits arise from this diversification for economic viability (Figure 4) and environmental goals such as input reduction and habitat conservation. Supporting diversification aims at site-adapted management to maintain extensive grassland-based systems in marginal areas. But diversification is not a one-size-fits-all strategy. Local conditions and farmer requirements must be considered. Moreover, there are trade-offs and levels of substitution between different levels of diversification. For instance, if farmer strategy leads to diversified activities, the workforce could be diluted and farmers therefore run the risk of managing each activity less well. This could negatively affect the potential of grassland biodiversity to stabilize and deliver ecosystem services. A case-adapted diversification that fits with the available workforce and benefits from local opportunities allows the preservation of grassland biodiversity, while enhancing farm socio-economic resilience, and its mitigation and adaptation potential to climate change and other perturbations. These opportunities are enabled by processes such as experimenting, knowledge sharing, farmer networking and cooperating, which are developing in European grassland-based systems and worldwide.

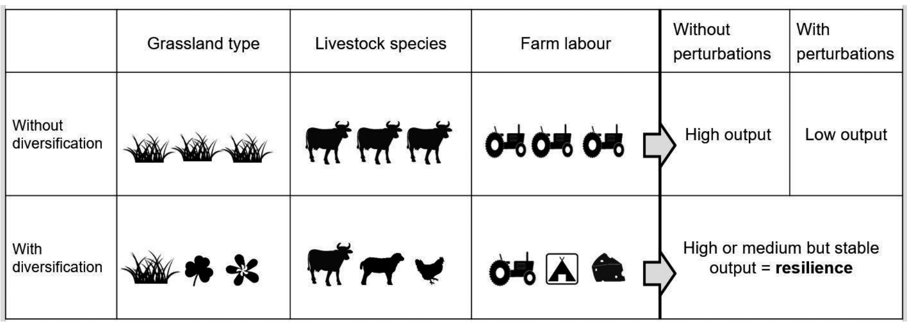

Figure 4. An overall representation of the diversification of European grassland-based systems and of its effects on resilience.

References

Arca P., Vagnoni E., Duce P. and Franca A. (2021) How does soil carbon sequestration affect greenhouse gas emissions from a sheep farming system? Results of a life cycle assessment case. *Italian Journal of Agronomy* 16, 1789.

Biggs R., Schlüter M., Biggs D., Bohensky E.L., Burn Silver S., Cundill G., ... and West P.C. (2012) Towards principles for enhancing the resilience of ecosystem services. *Annual Review of Environment and Resources* 37, 421-448.

Clarke D.J. and Dercon S. (2009) *Insurance, credit and safety nets for the poor in a world of risk*. DESA Working Paper No. 81, Department of Economics and Social Affairs, United Nations, 16 pp. Available at: https://www.un.org/esa/desa/papers/2009/wp81_2009.pdf

D'Alexis S., Sauvant D. and Boval M. (2014) Mixed grazing systems of sheep and cattle to improve liveweight gain: a quantitative review. *Journal of Agricultural Science* 152, 655-666.

Darnhofer I. (2014) Resilience and why it matters for farm management. *European Review of Agricultural Economics* 41, 461-484.

De Roest K., Ferrari P. and Knickel K. (2018) Specialization and economies of scale or diversification and economies of scope? Assessing different agricultural development pathways. *Journal of Rural Studies* 59, 222-231.

Dumont B., Rossignol N., Loucougaray G., Carrère P., Chadoeuf J., Fleurance G., ... and Yavercovski N. (2012) When does grazing generate stable vegetation patterns in temperate pastures? *Agriculture, Ecosystems and Environment* 153, 50-56.

Dumont B., Puillet L., Martin G., Savietto D., Aubin J., Ingrand S., ... and Thomas M. (2020) Incorporating diversity into animal production systems can increase their performance and strengthen their resilience. *Frontiers in Sustainable Food Systems* 4, 109.

Fahrig L., Baudry J., Brotons L., Burel F.G., Crist T.O., Fuller R.J., ... and Martin J.L. (2011) Functional landscape heterogeneity and animal diversity in agricultural landscapes. *Ecology Letters* 14, 101-112.

Finger R. and Calanca L. (2011) Risk management strategies to cope with climate change in grassland production: an illustrative case study for the Swiss plateau. *Regional Environmental Change* 11, 935-949.

Fleurance G., Sallé G., Lansade L., Wimel L. and Dumont B. (2022) Comparing the effects of horse grazing alone or with cattle on horse parasitism and vegetation use in a mesophile pasture. *Grass and Forage Science* (in press)

Forteau L., Dumont B., Sallé G., Bigot G. and Fleurance G. (2020) Horses grazing with cattle have reduced strongyle egg count due to the dilution effect and increased reliance on macrocyclic lactones in mixed farms. *Animal* 14, 1076-1082.

Franca A., Caredda S., Sanna F., Fava F. and Seddaiu G. (2016) Early plant community dynamics following overseeding for the rehabilitation of a Mediterranean silvopastoral system. *Journal of Grassland Science* 62, 81-91.

Grange G., Finn J.A. and Brophy C. (2021) Plant diversity enhanced yield and mitigated drought impacts in intensively managed grassland communities. *Journal of Applied Ecology* 58, 1864-1875.

Huber R., Briner S., Peringer A., Lauber S., Seidel R., Widmer A., ... and Hirschi C. (2013) Modeling social-ecological feedback effects in the implementation of payments for environmental services in pasture-woodlands. *Ecology and Society* 18, 41.

Jerrentrup S.J., Komainda M., Seither M., Cuchillo-Hilario M., Wrage-Mönnig N. and Isselstein J. (2020) Diverse swards and mixed grazing of cattle and sheep for improved productivity. *Frontiers in Sustainable Food Systems* 3, 125.

Joly F., Benoit M., Martin R. and Dumont (2021) Biological operability, a new concept based on ergonomics to assess the pertinence of ecosystem services optimization practices. *Ecosystem Services* 50, 101320.

Kölliker R., Kempf K., Malisch C.S. and Lüscher A. (2017) Promising options for improving performance and proanthocyanidins of the forage legume sainfoin (*Onobrychis viciifolia* Scop.). *Euphytica* 213, 179.

Lemaire G., Gastal F., Franzluebbers F. and Chabbi A. (2015) Grassland-cropping rotations: An avenue for agricultural diversification to reconcile high production with environmental quality. *Environmental Management* 56, 1065-1077.

López-i-Gelats F., Milán M.J. and Bartolomé J. (2011) Is farming enough in mountain areas? Farm diversification in the Pyrenees. *Land Use Policy* 28, 783-791.

López-i-Gelats F., Rivera-Ferre M.G., Madruga-Andreu C. and Bartolomé Filella J. (2015) Is multifunctionality the future of mountain pastoralism? Lessons from the management of semi-natural grasslands in the Pyrenees. *Spanish Journal of Agricultural Research* 13, e0307.

Loucougaray G., Bonis A. and Bouzillé J.B. (2004) Effects of grazing by horses and/or cattle on the diversity of coastal grasslands in western France. *Biological Conservation* 116, 59-71.

Lüscher A., Barkaoui K., Finn J.A., Suter D., Suter M. and Volaire F. (2022) Using plant diversity to reduce vulnerability and increase drought resilience of permanent and sown productive grasslands. In: *Grassland at the heart of circular and sustainable food systems* (eds. L. Delaby, R. Baumont, V. Brocard, S. Lemauviel-Lavenant, S. Plantureux, F. Vertes and J.L. Peyraud). *Grassland Science in Europe* 27, 309-322.

Marley C.L., Fraser M.D., Davies D.A., Rees M.E., Vale J.E. and Forbes A.B. (2006). The effect of mixed or sequential grazing of cattle and sheep on the faecal egg counts and growth rates of weaned lambs when treated with anthelmintics. *Veterinary Parasitology* 142, 134-141.

Martin G., Barth K., Benoit M., Brock, C., Destruel M., Dumont B., … and Primi R. (2020) Potential of multi-species livestock farming to improve the sustainability of livestock farms: A review. *Agricultural Systems* 181, 102821.

Melts I., Lanno K., Sammul M., Uchida K., Heinsoo K., Kull T. and Laanisto (2018) Fertilising semi-natural grasslands may cause long-term negative effects on both biodiversity and ecosystem stability. *Journal of Applied Ecology* 55, 1951-1955.

Moreno G., Aviron S., Berg S., Crous-Duran J., Franca A., García de Jalón S., … and Burgess P. J. (2018) Agroforestry systems of high nature and cultural value in Europe: provision of commercial goods and other ecosystem services, *Agroforestry Systems* 92, 877-891.

Mosnier C. (2015) Self-insurance and multi-peril grassland crop insurance: the case of French suckler cow farms. *Agricultural Finance Review* 75, 533-551.

Mosnier C. and Moufid N. (2021) Assessing and reducing the vulnerability of mixed organic cattle-sheep farms. *Deliverable 5.2 Core Organic project MixEnable* Available at: https://orgprints.org/id/eprint/42975/

Mosnier C., Lherm M., Devun J. and Boutry A. (2013) Sensibilité des élevages bovins et ovins viande aux aléas selon la place des prairies dans les systèmes fourragers. *Fourrages* 213, 11-20.

Mosnier C., Benoit M., Minviel J.J. and Veysset P. (2022) Does mixing livestock farming enterprises improve farm and product sustainability? *International Journal of Agricultural Sustainability*. https://doi.org/10.1080/14735903.2021.1932150

Mugnier S., Husson C. and Cournut S. (2021) Why and how farmers manage mixed cattle–sheep farming systems and cope with economic, climatic and workforce-related hazards. *Renewable Agriculture and Food Systems* 36, 344-352.

Nozières M.O., Moulin C.H. and Dedieu B. (2011) The herd, a source of flexibility for livestock farming systems faced with uncertainties? *Animal* 5, 1442-1457.

Nyfeler D., Huguenin-Elie O., Suter M., Frossard E. and Lüscher A. (2011) Grass-legume mixtures can yield more nitrogen than legume pure stands due to mutual stimulation of nitrogen uptake from symbiotic and non-symbiotic sources. *Agriculture, Ecosystems & Environment* 140, 155-163.

Pardini A. and Nori M. (2011) Agro-silvo-pastoral systems in Italy: integration and diversification. *Pastoralism: Research, Policy and Practice* 1, 26.

Pauler C.M., Isselstein J., Braunbeck T., Schneider M.K. (2019) Influence of Highland and production-oriented cattle breeds on pasture vegetation: a pairwise assessment across broad environmental gradients. *Agriculture, Ecosystems & Environment* 284, 1-11.

Pauler C.M., Isselstein J., Berard J., Braunbeck T. and Schneider M.K. (2020a) Grazing allometry: anatomy, movement, and foraging behavior of three cattle breeds of different productivity. *Frontiers in Veterinary Science* 7, 494.

Pauler C.M., Isselstein J., Suter M., Berard J., Braunbeck T. and Schneider M.K. (2020b) Choosy grazers: influence of plant traits on forage selection by three cattle breeds. *Functional Ecology* 34, 980-992.

Pauler C.M., Zehnder T., Staudinger M., Lüscher A., Kreuzer M., Berard J. and Schneider M.K. (2022) Thinning the thickets: Foraging of hardy cattle, sheep and goats in green alder shrubs. *Journal of Applied Ecology* DOI: 10.1111/1365-2664.14156

Phocas F., Belloc C., Bidanel J., Delaby L., Dourmad J.Y., Dumont B., … and Brochard M. (2016) Review: Towards the agroecological management of ruminants, pigs and poultry through the development of sustainable breeding programs: II. Breeding strategy. *Animal* 10, 1760-1769.

Regan J.T., Marton S., Barrantes O., Ruane E., Hanegraaf M., Berland J., … and Nesme T. (2017) Does the recoupling of dairy and crop production via cooperation between farms generate environmental benefits? A case-study approach in Europe. *European Journal of Agronomy* 82, 342-356.

Sands B. and Wall R. (2018) Sustained parasiticide use in cattle farming affects dung beetle functional assemblages. *Agriculture Ecosystems & Environment* 265, 226-235.

Smit B. and Wandel J. (2006) Adaptation, adaptive capacity and vulnerability. *Global Environmental Change* 16, 282-292.

Stampfli A., Bloor J.M.G., Fischer M. and Zeiter M. (2018) High land-use intensity exacerbates shifts in grassland vegetation composition after severe experimental drought. *Global Change Biology* 24, 2021-2034.

Steinmetz L.,Veysset P., Benoit M. and Dumont (2021) Ecological network analysis to link interactions between system components and performances in multispecies livestock farms. *Agronomy for Sustainable Development* 41, 42.

Tangermann S. (2011), *Risk management in agriculture and the future of the EU's common agricultural policy.* ICTSD, Issue Paper (34).

Vagnoni E. and Franca A. (2018) Transition among different production systems in a Sardinian dairy sheep farm: Environmental implications. *Small Ruminant Research* 159, 62-68.

Veysset P., Benoit M., Laignel G., Bébin D., Roulenc M. and Lherm M. (2014) Analyse et déterminants de l'évolution des performances d'élevages bovins et ovins allaitants en zones défavorisées de 1990 à 2012. *INRA Productions Animales* 27, 49-64.

Vogel A., Scherer-Lorenzen M. and Weigelt A. (2012) Grassland resistance and resilience after drought depends on management intensity and species richness. *PLoS ONE* 7, e36992.

Vollet D. and Said S. (2018) Vers l'identification de paniers de biens et de services liée à la demande locale dans les territoires d'élevage: illustration à partir de la Planèze de Saint Flour et du bocage bourbonnais. *Géocarrefour* 92/3. https://doi.org/10.4000/geocarrefour.11155

Walker B., Holling C.S., Carpenter S.R. and Kinzig A. (2004) Resilience, adaptability and transformability in social-ecological systems. *Ecology and Society* 9, 5.

Wang L., Delgado-Baquerizo M., Wang D., Isbell F., Liu J., Feng C., ... and Liu C. (2019) Diversifying livestock promotes multidiversity and multifunctionality in managed grasslands. *Proceedings of the National Academy of Sciences of the USA* 116, 6187-6192.

Biodiversity assessed through different metrics to evaluate grassland ecosystem services in Massif Central

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Abstract

Grasslands are biodiversity hotspots in Europe and provide a number of ecosystem services (ES) to farmers and society. Grasslands are also some of the most threatened ecosystems on earth, due to land use change and now climate change. Theoretical and empirical models suggest that plant diversity could play a key role on forage biomass and stability, but there is little evidence that this occurs under farming conditions. However, rising temperature, drier summers and increasing frequency of climatic extremes due to climate change may alter biodiversity-ES relationships. Therefore, how biodiversity mediates ES in changing conditions remains uncertain. We used data from 100 field plots from farms located in Massif Central (France). We built regression models to understand the interplay between climate, management, plant diversity and ES delivery (including biomass production, forage quality, carbon stock, habitat quality for pollinators and plant rarity) to understand how plant diversity can be used as a driver to modulate bundles of ESs. We found pervasive interactions between climate and biodiversity or management and biodiversity on most studied ecosystem services. We discuss under which conditions plant diversity can be used to optimize ES delivery in grasslands along climatic and management gradients.

Keywords: plant diversity, climate change, forage, carbon stock, fertilization

Introduction

Grasslands in French Massif Central provide multiple ecosystem services (ES), to society, and represent 70% of agricultural area in this region (Atlas cartographique, 2016). ES are defined as 'the contributions that ecosystems make to human well-being' (Haines-Young, 2011). Climate change (CC) previsions of IPCC predict a shift of climates towards the poles and climate hazards to become more frequent in forthcoming decades (Lee *et al.*, 2021), which will impact the structure of vegetation communities (Alexander *et al.*, 2018). Biodiversity plays a key role in maintaining ecosystem functions that support the provisioning of multiple ecosystem services (Cardinale *et al.*, 2012). Emerging evidence suggests that the effect of biodiversity loss on ecosystems can be as strong as the effect of CC, although climate may also alter the biodiversity-ecosystem services relationship (Garcia Palacios *et al.*, 2018). Yet, there is still no consensus on the relationship between grassland diversity and ES delivery in real world ecosystems (Hagan *et al.*, 2021). Considering the decline of biodiversity and its variability around the globe, this relationship should be defined at a local scale. Here, we aim to understand how plant diversity drives the delivery of multiple ES in grasslands and how climate and local management practices modulate biodiversity-ES relationships. Our goal is to identify under which conditions plant diversity can be used as a lever to mitigate the effect of CC on ES delivery.

Materials and methods

We analysed data from 100 grassland plots from Massif Central farms. Data were collected in either 2008 and 2009, 2014 and 2015 or 2016 and 2017. Those data include elevation data, measurements of soil attributes (pH, C/N and sand) and agricultural practices (management regime: grazing vs mowing, and total nitrogen fertilization ranging from 0 to 132 kg N ha^{-1}). The effects of climate were assessed through the altitude gradient (ranging from 272 to 1,448 m). Plant diversity was assessed through the Braun-

Blanquet method, which is based on the relative area covered by each plant species in at least two 49×49 cm quadrats per plot at flowering peak. From these surveys, two plant diversity metrics were calculated at quadrat scale: plant species richness and the equitability in different functional groups (calculated with Shannon index applied to five groups of plants: grasses, legumes, graminoids, forbs and ligneous).

We built linear models of five ES measured in the field. We first built a model for annual biomass production (biomass in tons per hectare) as a biotic provisioning ES (Haines-Young and Potschin, 2018) that was assessed as the sum of three cuts above 5 cm during spring, in four 70×70 cm quadrats, isolated in 2 cages per plot. We then built a model of forage quality that was assessed using nitrogen concentration in 500 g grass samples collected for biomass production that were analysed by infrared spectroscopy after calibration by analytical methods. We also built models for two regulation services: carbon stock in the soil, measured in the first 10 cm, and the habitat value of grasslands for pollinators (average of estimated attractiveness to pollinators mark of each plant species multiplied by its relative cover area from Braun-Blanquet recordings). The fifth ES we considered is a cultural service, plant species rarity in the context of Massif Central. It was estimated using the local abundance of rare species in context of Massif Central using regional data base from the Conservatoire Botanique du Massif Central.

We evaluated using regression models how climate (elevation), grassland management (mowing vs pasture, fertilization) and local plant diversity (plant species richness, functional group diversity) modulate ES delivery. All ES and predictors were scaled before building the models. The most parsimonious linear models were chosen for each ES according to AIC with dredge function from MuMin package in R.

Results and discussion

Plant diversity metrics were significant in most models and influenced ES in different ways. First, biodiversity could be directly correlated to ES as shown in carbon stock model, where functional equitability had a direct positive effect (i.e. without interaction with other variables). Fertilization also had a direct positive effect and it did not interact with biodiversity. This result suggested that functional equitability and fertilization could be used together to increase carbon stock in the soil, at least in the amplitude of fertilization of our sample.

For other ES, biodiversity effects were modulated by climate (Figure 1). For example, functional equitability interacted with elevation on pollination service. Functional equitability increased with habitat quality for pollinators only at low elevation. The opposite pattern happened for plant rarity. Functional equitability was negatively correlated to plant rarity indicator at low altitude but this correlation turned positive as elevation increased (Figure 1).

Third, biodiversity effects could also be modulated by management practices. In forage quality, the two plant diversity metrics interacted differently with fertilization (Figure 2). Beneficial effect of functional

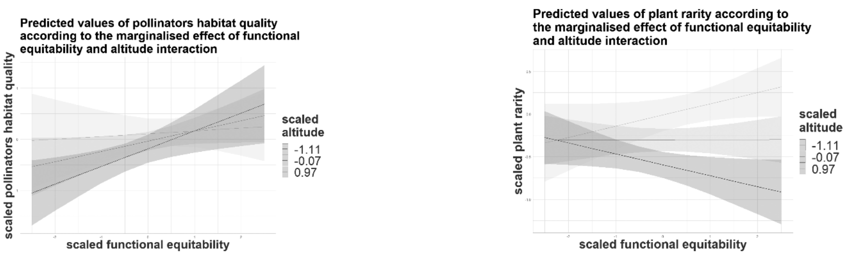

Figure 1. Representations of the isolated effects of functional equitability and altitude interaction in pollinators habitat quality and plant rarity models.

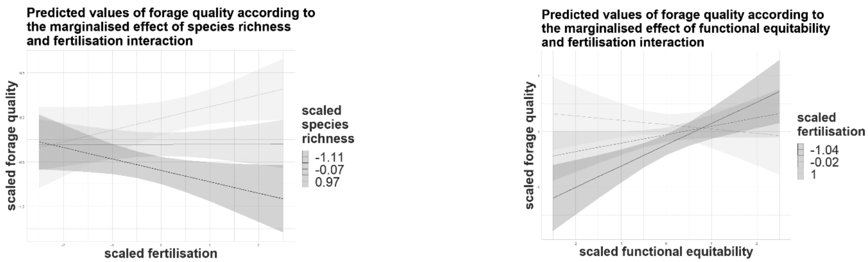

Figure 2. Representations of the isolated interaction effects with both plant diversity metrics in forage quality model.

equitability was only observed at low fertilization level. For species richness, we observed a positive significant interaction with fertilization suggesting synergistical effect of species richness and fertilization on forage quality.

Finally, biodiversity had no significant effect on biomass production, regardless of the biodiversity metric used. Since we used linear models, this result does not eliminate the possibility of a non-linear relationship between these variables (Grime, 1973). Elevation for climate substitution worked for temperature (average daily temperature from 2000 to 2019 (Le Moigne, 2002) and elevation were correlated at -87%) more than rainfall (average yearly rainfall from 2000 to 2019 and elevation are only correlated at 41%). A way to improve models would be to use directly temperature and rainfall gradients to assess the effect of climate change.

Conclusions

Linear models for ES showed different roles played by both plant diversity metrics in ES delivery. Since plant diversity has significant direct effect in some ES, or appears significant in interactions with fertilization or elevation/climate, it seems that taking account of different plant diversity metrics suggests different levers or combination of levers to increase ES individually in the context of climate change. We also found that the use of different biodiversity metrics can help understand how to optimize grassland management to adapt to climate change as seen in the forage quality model where species richness and functional equitability interact differently with fertilization.

References

Alexander, J.M. *et al.* (2018) Lags in the response of mountain plant communities to climate change. *Global Change Biology* 24(2), 563-579.

Atlas cartographique (2016) *Le SIDAM et la COPAMAC*. Available at: https://www.sidam-massifcentral.fr/massif-central/atlas-cartographique/.

Grime J.P. (1973) Competitive exclusion in herbaceous vegetation. *Nature* 242(5396): 344-347.

Hagan J.G., Vanschoenwinkel B. and Gamfeldt L. (2021) We should not necessarily expect positive relationships between biodiversity and ecosystem functioning in observational field data. *Ecology Letters*. https://onlinelibrary.wiley.com/doi/abs/10.1111/ele.13874

Haines-Young R. (2011) Roy Haines-Young Centre for Environmental Management School of Geography, University of Nottingham Nottingham, NG7 2RD, UK, 17.

Haines-Young R. and Potschin M. (2018) Common International Classification of Ecosystem Services (CICES) V5.1 and Guidance on the Application of the Revised Structure, 53.

Le Moigne P. (2002) *Description de l'analyse des champs de surface sur la France par le système Safran. Note de centre GMME, Météo-France.*

Lee, J.Y. *et al.* (2021) Future global climate: scenario-based projections and near-term information. In *Climate Change 2021: The Physical Science Basis. Contribution of Working Group I to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change.* Cambridge University Press, Cambridge, UK.

Response of different grass-based mixtures to weather conditions in the Netherlands

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Abstract

The productivity of grasslands is affected by the response of plants to different weather conditions. Climate change will increase the occurrence of extreme weather events, such as droughts and floods, highlighting the importance of increased resilience of production systems. Multispecies swards are thought to be more resilient to climate change. Therefore, the aim of this experiment was to determine the grass growth of different multispecies swards under varying weather conditions such as drought and high temperatures. For this purpose four different mixtures were sown, namely: perennial ryegrass with herbs, tall fescue with herbs, perennial ryegrass with white clover, perennial ryegrass with red clover. Grass was harvested every four weeks using the Corral and Fenlon methodology and was mown at 4 cm or 9 cm. Results show that, under the average weather conditions in the Netherlands of 2021, grass mixtures with herbs had a higher herbage growth in early grazing season and perennial ryegrass with red clover had a higher grass growth mid-summer. Grass mown at 9 cm had a higher grass growth in the period between half of June and end of June. Grass mown at 4 cm had a higher grass growth in the last two weeks of August.

Keywords: grass growth, resilience, multispecies swards, grass mixtures, cutting height

Introduction

Grasslands provide a variety of ecosystems services, such as carbon sequestration, biodiversity and relatively inexpensive high-quality feed for livestock. In the Netherlands, 50% of agricultural land is grassland (CBS, 2021). The Dutch dairy industry has established a committee to issue an advisory report on how the dairy farming sector in the Netherlands could become land-based by 2040. An important step in that advice is that, in 2025, 65% of the protein in the ration must come from the farmer's own farm. Increased grass production can contribute to that goal. The productivity of grasslands is affected by the response of plants to different weather conditions. Climate change will increase the occurrence of extreme weather events, such as droughts and floods and high temperatures, highlighting the importance of increased resilience of production systems. Multispecies swards are thought to be more resilient to climate change (Finn *et al.*, 2018). The aim of this study was therefore to determine the response of different multispecies swards to weather conditions such as drought and high temperatures in the Netherlands. For this purpose growth rates of different mixtures were monitored during the growing season of 2021.

Materials and methods

The study used a randomized block design with a factorial arrangement of two treatments (grass mown at 4 cm and at 9 cm above soil surface) in two replicates during the 2021 grazing season (April – October) in four paddocks differing in botanical composition at the organic research farm of Aeres University of Applied Sciences in Dronten, the Netherlands. The first mixture (LP Herb) consisted of *Lolium perenne* + seven types of herbs; the second mixture (FA Herb) consisted of *Festuca arundinacea* + seven types of herbs; the third mixture (LP White) consisted of *L. perenne* + *Trifolium repens* (white clover); and the fourth mixture (LP Red) consisted of *L. perenne* + *Trifolium pratense* (red clover). The herbs were similar for LP Herb and FA Herb, namely: (1) *T. pratense* (red clover); (2) *T. repens* (white clover); (3) *Onobrychis viciifolia* (common sainfoin); (4) *Carum carvi* (caraway); (5) *Cichorium intybus* (common chicory); (6) *Lotus corniculatus* (bird's-foot trefoil); and (7) *Plantago lanceolata* (ribwort plantain). LP Red was sown in autumn 2020, LP Herb and FA Herb were sown autumn 2018, and LP White was sown autumn 2014. Herbage growth was measured using the Corral and Fenlon (1978) methodology which estimates growth on a 4-week harvest interval. Four series of plots are harvested in rotation, spaced a week apart so that there is a routine of harvesting a constant number of plots on the same day each week (= cutting moment). The data are used to construct growth curves showing the rate at which dry matter (DM) is produced each week (kg DM ha^{-1}) of the growing season, on swards which are being harvested every four weeks. The quadratic equation for growth rate in week *t* is:

$$Rate_t = (A_1Y_t + A_2Y_{t-1} + A_3Y_{t-2} + A_4Y_{t-3}) / 28$$

where Y_t, Y_{t-1}, Y_{t-2} and Y_{t-3} are the harvested yields at the ends of weeks t, t-1, t-2 and t-3, respectively, and $A_1 = 7/16$, $A_2 = 5/16$, $A_3 = 3/16$ and $A_4 = 1/16$, with greater weight given to plots nearer harvest. Fresh herbage was dried for 48 h at 60 °C and DM content was determined. For analysis, a mixed design ANOVA was performed with cutting height (4 cm, 9 cm) and mixture (FA Herb, LP Herb, LP White, LP Red) as between-subject variables. Cutting moment was taken as within subject variable with all the weekly grass growth measures as dependent variable. Post hoc analyses were performed on significant multilevel effects and interactions. Bonferroni correction was applied when applicable. Alpha was set at 0.05.

Results and discussion

Weather conditions in the Netherlands in the period between April and November 2021 could be characterized as average. There was no precipitation deficit, and after very hot summers in preceding years, the mean temperature in the 2021 summer (17.7 °C) was similar to the long-term average of 17.5 °C (KNMI, 2021).

Our results (Figure 1) show a significant main effect of cutting moment ($F(28,224)=446.866$, $P<0.001$, $\eta^2=0.794$) and mixture ($F(3,8)=94.594$, $P<0.001$, $\eta^2=0.061$). The two-way interactions of cutting moment × mixture ($F(84,224)=676.086$, $P<0.001$, $\eta^2=0.107$) and cutting moment × height ($F(28,224)=9.022$, $P<0.001$, $\eta^2=0.016$) were also found to be significant.

The interaction of cutting moment × mixture was driven by the significant differences in grass growth in the period between 13 April 2021 and 8 June 2021 and during the period 20 July 2021 and 10 August 2021 between the mixtures FA Herb – LP Red, FA Herb – LP White, LP Herb – LP Red and LP Herb – LP White (all $P<0.05$). LP Red had a significantly higher amount of herbage growth per day than the other mixtures in the period between 6 July 2021 and 17 August 2021. The lower growth rate of LP Red in early spring can be explained by the history of this field. It had previously been arable land before being converted to grassland in autumn 2020. Post-hoc analyses show that differences in herbage growth in the period between 15 June 2021 and 29 June 2021, and between 24 August and 31 August 2021, between cutting height of 4 cm and cutting height of 9 cm were significant (all $P<0.05$). Swards mown at 9 cm had greater herbage growth in the period between mid June and end of June ($P<0.005$). Grass mown at 4 cm had a greater amount of growth in the last two weeks of August ($P<0.05$).

More experiments are recommended since the weather conditions during the experimental period were similar to long-term average with no extreme weather events, such as droughts and floods.

Conclusions

Results showed that in the year 2021, with average weather conditions, some differences in herbage growth rates between different mixtures and between different cutting heights were present, but they were relatively small. Continuation of experiments is recommended to test the resilience of different grass mixtures under Dutch conditions in the long term.

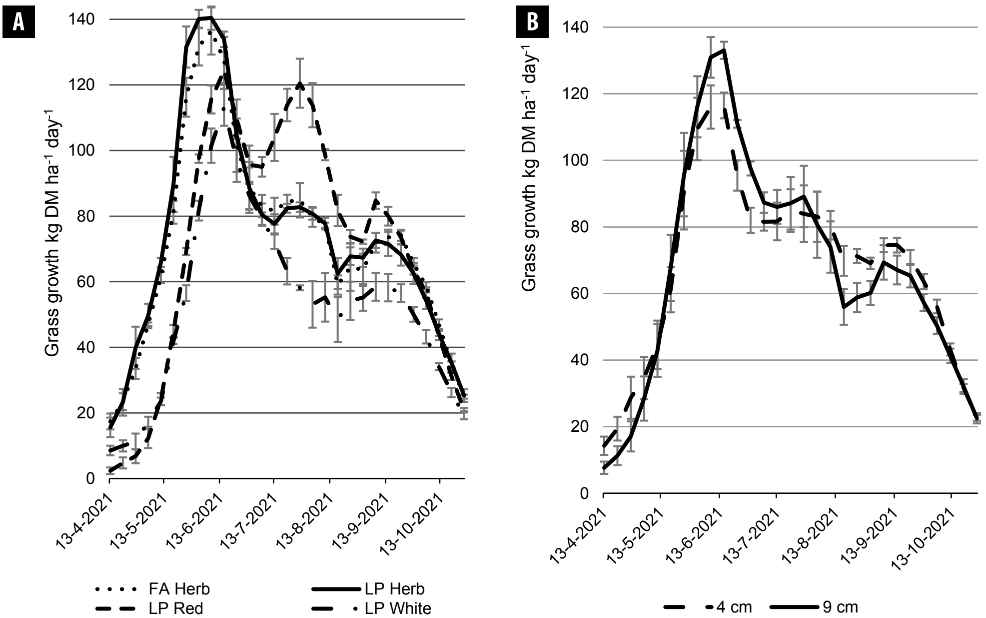

Figure 1. (A) Herbage growth per mixture, and (B) herbage growth per cutting height during the growing season of 2021. The data shown are means (± standard error).

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References

CBS (2021. Regionale kerncijfers Nederland. Available at: https://opendata.cbs.nl/statline/#/CBS/nl/dataset/70072ned/table?ts=1637570264743

Corral, J. and Fenlon, J.S. (1978) A comparative method for describing the seasonal distribution of production from grasses. *Journal of Agricultural Sciences* 91, 61-67. https://doi.org/10.1017/S0021859600056628

Finn, J.A., Suter, M., Haughey, E., Hofer, D. and Lüscher, A. (2018) Greater gains in annual yields from increased plant diversity than losses from experimental drought in two temperate grasslands. *Agriculture, Ecosystems and Environment* 258, 149-153. https://doi.org/10.1016/j.agee.2018.02.014

KNMI (2021). KNMI Data platform. Available at: https://dataplatform.knmi.nl/

Multispecies swards exceed the productivity of perennial ryegrass monocultures under a beef rotational grazing system

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Abstract

Decreasing reliance on fertilizer N is key to enhancing the sustainability of pasture-based systems. Three sward types were investigated in a farmlet experiment for their dry matter (DM) yield potential: (1) *Lolium perenne* (PRG; 205 kg N ha^{-1} a^{-1}); (2) PRG and *Trifolium repens* (PRGWC; 90 kg N ha^{-1} a^{-1}); (3) multispecies sward consisting of PRG, *Phleum pratense*, *T. repens*, *T. pratense*, *Cichorium intybus* and *Plantago lanceolata* (MSS; 90 kg N $ha^{-1}a^{-1}$). Farmlets were stocked with 20 weanling and 20 yearling beef steers (2.5 LU ha^{-1}) originating from the dairy herd. Herbage samples were collected for DM yield and botanical composition determination at each grazing. In their first year (2020) swards produced annual DM yields of 10,339, 10,753 and 12,744 kg DM ha^{-1} respectively, despite reduced N application to PRGWC and MSS swards. In MSS, herbs contributed more and legumes less, to DM yield early in the season than in mid/late season. These results indicate the potential for more botanically diverse swards to enhance dry matter production under much reduced nitrogen fertilizer application rates relative to PRG monocultures.

Keywords: multispecies swards, sustainability, legumes, forage herbs, nitrogen

Introduction

Reduced reliance on chemical fertilizer inputs is key to addressing several sustainability challenges associated with ruminant livestock production systems. This is reflected in the EU Farm to Fork Strategy, which has committed to a 20% reduction in the use of chemical fertilizers across EU Member States by 2030 (EC, 2020). High output grass-based systems in temperate regions are generally reliant on *Lolium perenne* L. (perennial ryegrass) monocultures that have many desirable agronomic qualities (Lee *et al.,* 2010) but are heavily dependent on unsustainable inputs of nitrogen (N) to achieve high dry matter (DM) yields (Dillon *et al.,* 2021). Legume inclusion in swards reduces the requirement for fertilizer N and can also result in improved DM production and feed value relative to perennial ryegrass monocultures. Further increasing sward diversity, multispecies swards have been found to be beneficial in achieving high DM yields (Grace *et al.*, 2018) while also enhancing animal health and performance (Grace *et al.*, 2019) relative to perennial ryegrass monocultures. Thus, multispecies swards appear to have the potential to address many concerns regarding the sustainability of livestock production systems. While research on multispecies has been undertaken in dairy systems, these are not necessarily transferable to beef production systems due to longer residence time in paddocks, for example. Data regarding multispecies swards' performance under a dairy-calf to beef rotational leader-follower grazing system is lacking. Therefore, the objective of this study is to better elucidate the potential of multispecies and perennial ryegrass-white clover relative to perennial ryegrass swards managed under a dairy calf to beef system. Sward types are compared in terms of annual and seasonal DM production and the contribution of the different species components of the swards to DM production over the growing season.

Materials and methods

The study was undertaken on the University College Dublin, Lyons Farm Long-Term Grazing Platform (53°29 ′N, 6°53 ′W), a 24 ha site, established in July 2019 to develop measures to enhance the sustainability of Irish grass-based agricultural systems. The site is divided into three 8-ha farmlets, with each farmlet having one of three sward types: (1) *L. perenne* monoculture receiving 205 kg N ha^{-1} a^{-1} (PRG), (2) *L. perenne* and *Trifolium repens* sward receiving 90 kg N $ha^{-1}a^{-1}$ (PRGWC) and (3) a six-species sward (*L. perenne, Phleum pratense, T. repens, Trifolium pratense*, *Cichorium intybus* and *Plantago lanceolata*) receiving 90 kg N ha^{-1} a^{-1} (MSS) and grazed on a 1 ha paddock scale. Each farmlet was stocked at 2.5 LU ha^{-1}, consisting of 20 weanlings (<1 years) followed by 20 yearling steers (>1 years) from the dairy herd. The data presented were collected over the growing season, March to November 2020. Cattle grazed the sward and silage was cut to a target post-grazing residual height of 4 cm above ground level for the PRG and PRGWC farmlets and 6 cm for the MSS farmlet. Following the first rotation 50% of each farmlet area was closed for silage; in addition, covers above 1,800 kg DM ha^{-1} were taken as surplus bales. Pre-grazing herbage mass above grazing residual was calculated before stock entered each paddock (target: 1,500 kg DM ha^{-1}), by cutting three 2.65 m^2 strips in representative areas of each paddock with a Honda mower, weighing using a field scale (Jadever®), and drying a sub-sample. Compressed sward height was measured before and after stock grazed by taking 30 measurements with a rising plate meter (Jenquip®). Annual and seasonal yield was adjusted for herbage growth rate while animals were resident in paddocks, as per Doyle *et al.* (2021). Botanical composition was determined by the dry-weight rank method (Jones and Hargreaves, 1979), involving a 500 g fresh sample of herbage taken from each paddock prior to grazing. Samples were sorted into sown and unsown species and the DM of the biomass represented by each was calculated. For analysis purposes, the grazing season was divided into early-season (March-May), mid-season (June-August) and late-season (September-November). Data were analysed as a complete randomized block design using the mixed model procedure (PROC MIXED, SAS, version 9.4, Inst. Inc., Cary, NC) with individual paddocks serving as the experimental unit. The statistical model used for DM yield comparison included the fixed effect of sward type, season and the random effect of paddock within block. Botanical compositions were analysed by sward type and included the fixed effect of season and the random effect of paddock within block. Mean comparisons were made using LSMEANS and adjusted with the TUKEY statement for multiple comparisons.

Results and discussion

In 2020, swards of PRG, PRGWC and MSS produced yields of 10,339, 10,753 and 12,744 kg DM $ha^{-1}a^{-1}$ respectively, despite the latter two sward types receiving only approximately 44% of the nitrogen fertilizer applied to the PRG swards. MSS produced higher annual herbage yields than PRG ($P<0.05$) and late-season yields than PRG/PRGWC ($P<0.05$). Mean annual pre-grazing sward heights of MSS were higher than the PRG/PRGWC swards ($P<0.01$). The MSS treatment had a shorter rotation length and more annual rotations than PRG/ PRGWC swards ($P<0.05$; Table 1). The number of rotations achieved was 37% higher for MSS than for PRG or PRGWC swards, likely a result of less severe grazing management and increased dry-matter intake. While sward type had a significant influence on herbage yield, this is coupled with management, i.e. the more frequent grazing but less severe grazing residual of MSS may have contributed to its higher yield compared to PRG or PRGWC, independently from species composition as a greater leaf area remained to photosynthesize. In MSS, herbs contributed more and legumes less, to DM yield early in the season (40 and 16%, respectively) than in mid (29 and 33%) and late seasons (22 and 34%; $P<0.05$), while the grass component of the sward remained consistent throughout the growing season. PRGWC tended to have a higher weed burden than MSS ($P<0.1$), which is important given farmers' concerns about lack of availability of post-emergence herbicides for herb-containing swards.

Conclusions

Reducing reliance on fertilizer N is key to enhancing the sustainability of grassland systems. In their first year, multispecies swards produced greater yields than PRG, despite reduced N application, highlighting their potential in reducing N dependence in grassland agriculture.

Table 1. Mean biomass production grazing parameters and weed burden of the three farmlets in 2020.[1]

	PRG	PRGWC	MSS	SEM	*P*-value
Annual yield (kg DM ha^{-1})	10,339^{a}	10,753^{ab}	12,744^{b}	1190	<0.05
Early-season (March to May)	4,548	4,583	4,344	854	ns
Mid-season (June to August)	4,113^{x}	4,554^{xy}	5,774^{y}	878	<0.10
Late-season (September to November)	1,678^{a}	1,616^{a}	2,627^{b}	210	<0.05
Pre-grazing herbage mass[2] (kg DM ha^{-1})	1,631	1,542	1,431	104	ns
Pre-grazing sward height[2] (cm)	10.3	9.5	11.1	0.6	ns
Rotation length[2] (days)	23.1^{a}	24.0^{a}	18.6^{b}	1.7	<0.05
No. rotations	4.6^{x}	4.6^{x}	6.3^{y}	0.2	<0.01
Weed burden (g kg DM^{-1})	23.2^{xy}	40.4^{x}	20.6^{y}	6.2	<0.10

[1] ns = non-significant (P>0.10). Within rows means of differing superscript (a,b) letters differ significantly (P<0.05). Within rows means of differing superscript (x,y) letters tend to differ (P<0.10). SEM = standard error of the mean.

[2] Exclude the harvest of silage.

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References

Dillon E., Donnellan T., Moran B. and Lennon J. (2021) *Teagasc National Farm Survey 2020; Preliminary Results*. Agricultural Economics and Farm Surveys Department Rural Economy Development Programme Teagasc. Agriculture and Food Development Authority. Available at: https://www.teagasc.ie/media/website/publications/2021/National-Farm-Survey-2020-Preliminary-Results.pdf.

Doyle P.R., McGee M., Moloney A.P., Kelly A.K. and O'Riordan E.G. (2021) Effect of post-grazing sward height, sire genotype and indoor finishing diet on steer intake growth and production in grass-based suckler weanling-to-beef systems. *Animals* 11, 2623.

European Commission (2020) *Farm to Fork Strategy – for a fair healthy and environmentally-friendly food system.* Communication from the Commission to the European Parliament, the Council, the European Economic and Social Committee and the Committee of the Regions. Available at: https://ec.europa.eu/info/sites/default/files/communication-annex-farm-fork-green-deal_en.pdf.

Grace C., Boland T.M., Sheridan H., Lott S., Brennan E., Fritch R. and Lynch M.B. (2018) The effect of increasing pasture species on herbage production chemical composition and utilization under intensive sheep grazing. *Grass and Forage Science* 73, 852-864.

Grace C., Lynch M.B., Sheridan H., Lott S., Fritch R. and Boland T.M. (2019) Grazing multispecies swards improves ewe and lamb performance. *Animal* 13, 1721-1729.

Jones R.M. and Hargreaves J.N.G. (1979) Improvements to the dry-weight-rank method for measuring botanical composition. *Grass and Forage Science* 34, 181-189.

Lee J.M., Donaghy D.J., Sathish P. and Roche J.R. (2010) Perennial ryegrass regrowth after defoliation-physiological and molecular changes. *Proceedings of the New Zealand Grassland Association* 72, 127-134.

Effect of N fertilization on the biomass of soil fungal groups in production grasslands

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Abstract

Nutrient fertilizer application is common in production grasslands as it boosts primary production. We established two 5-year field experiments in the south of Sweden (Lanna, Alnarp). These sites included a two-factorial experiment, i.e. four plant mixtures and two or three levels of nitrogen (N) fertilizer application. The phospholipid- and neutral lipid fatty acid (PLFA, NLFA) analysis was used to estimate the biomass of fungal groups (saprotrophic and arbuscular mycorrhizal fungi, SF and AMF, respectively). At Alnarp, N application was associated with a significant decrease of the AMF biomass, but no trends were seen at Lanna. Similarly, the ratio of AMF/SF decreased with fertilization at Alnarp but not at Lanna. Only one of the plant mixtures (the grass-legume mixture) had an effect in the unfertilized plots, decreasing AMF at Alnarp. Our findings suggest that the impact of N fertilization on soil fungal biomass, specifically the reduction in the abundance of AMF, is site dependent with this group being shown to be more sensitive than SF to N fertilizer application.

Keywords: soil microbial biomass, ley production, diversification, inorganic fertilization

Introduction

Nutrient fertilizer application is generally expected to lead to priming, i.e. a short-term response of microorganisms to easily available nutrients, with the subsequent immediate immobilization and microbial growth before the nutrients eventually become mineralized into plant available forms. This response is thought to more likely involve saprotrophic microorganisms than obligate symbionts, such as arbuscular mycorrhizal fungi. The abundance of AMF has been reported to decrease with N fertilization (Jach-Smith and Jackson, 2018), but contradictory results are also reported, where N fertilization sometimes is found to influence just the AMF community structure (Chen *et al.*, 2014). AMF are generally not selected for by the plant host when nutrients are in excess (Hammer *et al.*, 2011), as is the case under management with mineral fertilization. Plant species diversity enriches AMF functional diversity (Guzmán *et al.*, 2020), but the impact on saprotrophic fungi (SF) is variable. Here we report on the impact of different levels of both plant diversity and mineral N fertilizer application on the soil fungal community of production grasslands. Specifically, we hypothesized that AMF are: (1) more abundant in unfertilized compared to fertilized ley production plots; and (2) promoted by increasing plant species diversity.

Materials and methods

Two field experiments (Alnarp and Lanna) were established in 2013 in the south of Sweden, which has a temperate climate (annual average: 10 °C and 660 mm precipitation). Both sites included a two-factorial experiment, i.e. four plant species mixtures (Table 1) and two N fertilization levels (0 and 60 kg ha^{-1} yr^{-1}), with an additional level (120 kg ha^{-1} yr^{-1}) at Alnarp. Both experiments were replicated in four blocks with a complete randomized factorial design. In the summer of 2018, each experimental plot was divided into four subplots for taking four soil cores (ø 2.5 cm) to a depth of 20 cm and four 0.25 m^2 squares for estimated plant biomass and to make a plant community inventory. The plant biomass and soil sub-samples were homogenized per plot, and soil samples sieved (2 mm) before all were stored at -20 °C. Plant and soil total N, P and C content and soil texture were analysed. Phospho- and neutral lipid fatty acids (PLFAs and NLFAs) were extracted from the soil samples using the protocol presented by Frostegård *et al.* (1993) and quantified by gas chromatography with a flame ionization detector, and both SF and AMF biomass were estimated. Treatment impact on the microbial biomarkers was analysed with an ANOVA with Tukey's post hoc test. Correlations were detected using parametric Pearson correlation test (P<0.05).

Results and discussion

Both experimental sites had a sandy soil texture: loamy sand at Alnarp and sandy loam at Lanna. The total soil C, N and P were not affected by the applied treatments, but Alnarp showed higher values of all the parameters, especially P.

At Lanna there was a greater amount of AMF biomass than at Alnarp, but no effect of fertilization was detected at the former, while mineral-N fertilizer application significantly decreased the AMF biomass iat Alnarp (9.4, 4.4 and 1.3 nmol g^{-1} for 0, 60 and 120 kg N, respectively) (Figure 1A, B). At this site, the AMF biomass was negatively correlated with the total soil N content (R=-0.29, P<0.05). AMF stimulation under N deficiency in P-rich soils, have been described (Blanke *et al.*, 2005). The total plant N content (%) was positively correlated with the AMF biomass at Alnarp (R=0.48, P<0.001), possibly because the AMF can favour N uptake. The SF biomass was similar at both sites, and no impact of N fertilization was detected (Figure 1C, D). The total plant P content was positively correlated with the

Table 1. Plant species mixtures sown at both experimental sites.

PM1	*Dactylis glomerata* (100%)
PM2	*Phalaris arundinacea* (33%), *Festuca arundinacea* (33%), *D. glomerata* (33%)
PM3	*Medicago sativa* (12.5%), *Trifolium hybridum* (12.5%), *Trifolium repens* (12.5%), *Galega orientalis* (12.5%), PM2 (50%)
PM4	Commercial diverse meadow seed mixture (from 'Pratensis') (75%), PM3 (25%)

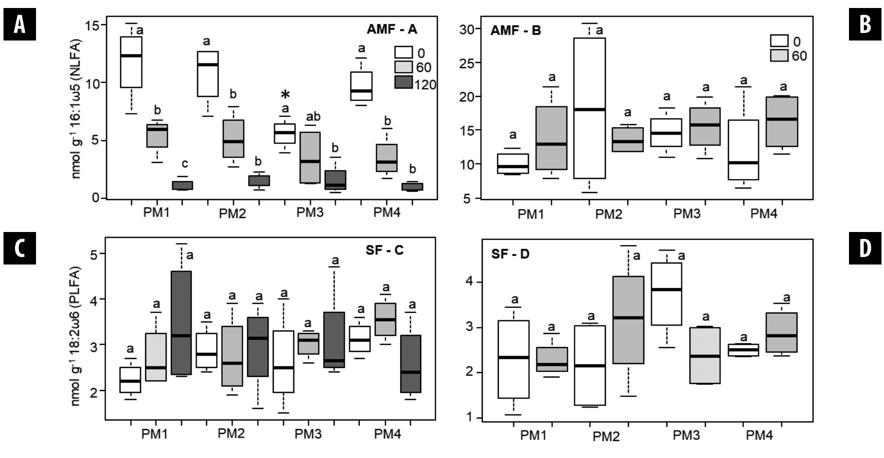

Figure 1. The amount (nmol g^{-1}) of biomarkers of arbuscular mycorrhizal fungi (AMF) and saprotrophic fungi (SF) in relation to fertilization levels (0, 60, 120 kg ha^{-1}) and plant mixtures (PM) at the Alnarp (A, C) and Lanna (B, D) experiments. Different letters and * indicate significant differences in fungal biomass under different fertilization levels and plant mixtures, respectively.

SF biomass at Lanna (R=0.38, P<0.05). This correlation could indicate the presence of P-solubilizing fungi, which constitute about 0.1-0.5% of the total fungal populations in soils (Mehta *et al.*, 2019). The different plant species mixtures did not affect fungal biomass, except for the grass-legume mixture (PM3), which had an effect only in the unfertilized plots, decreasing the AMF biomass at Alnarp (P<0.05, Figure 1A). The interactions between plant diversity and soil microorganisms are major determinant of a plant's influence on ecosystem function. Though other studies have reported that fungal biomass increases significantly with plant diversity (Eisenhauer *et al.*, 2017), such a relationship was not seen in our experiment. The site-specific response of the fungal biomass to the applied treatments might be related to the differences in soil properties or to the previous land use at the site.

Conclusions

Our findings suggest that the response of soil fungal biomass to mineral N fertilizer application has a strong site-specific component, and the reduction in the abundance of AMF, which was more sensitive than SF, only occurs under specific soil conditions.

Acknowledgements

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References

Blanke, V., Renker, C., Wagner, M., Füllner, K., Held, M., Kuhn, A.J. and Buscot, F. (2005) Nitrogen supply affects arbuscular mycorrhizal colonization of *Artemisia vulgaris* in a phosphate-polluted field site. *New Phytologist* 166, 981-992.

Chen Y.L., Zhang X., Ye J.S., Han H.Y., Wan S.Q. and Chen B.D. (2014) Six-year fertilization modifies the biodiversity of arbuscular mycorrhizal fungi in a temperate steppe in Inner Mongolia. *Soil Biology and Biochemistry* 69, 371-381.

Eisenhauer, N., Lanoue, A., Strecker, T., Scheu, S., Steinauer, K., Thakur, M.P. and Mommer, L. (2017) Root biomass and exudates link plant diversity with soil bacterial and fungal biomass. *Scientific Reports* 7, 44641.

Frostegård Å., Bååth E. and Tunlid A. (1993) Shifts in the structure of soil microbial communities in limed forests as revealed by phospholipid fatty acid analysis. *Soil Biology and Biochemistry* 25, 723-730.

Guzmán A., Montes M., Hutchins L., De La Cerda G., Yang P., Kakouridis A., ... and Kremen C. (2021) Crop diversity enriches arbuscular mycorrhizal fungal communities in an intensive agricultural landscape. *New Phytologist* 231, 447-459.

Hammer E.C., Pallon J., Wallander H. and Olsson P.A. (2011). Tit for tat? A mycorrhizal fungus accumulates phosphorus under low plant carbon availability. *FEMS Microbiology Ecology* 76, 236-244.

Jach-Smith L.C. and Jackson R.D. (2018) N addition undermines N supplied by arbuscular mycorrhizal fungi to native perennial grasses. *Soil Biology and Biochemistry* 116, 148-157.

Mehta P., Sharma R., Putatunda C. and Walia A. (2019). Endophytic fungi: role in phosphate solubilization. In: Singh B. (ed.) *Advances in Endophytic Fungal Research. Fungal Biology*. Springer, Cham, Switzerland.

Grazing heights and nitrogen applied in warm season pastures do not change forage production and species diversity in the following cool season pastures in a subtropical environment

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Abstract

Increasing plant species diversity in a pastoral environment provides several benefits over monocultures. Despite this, factors such as grazing height and nitrogen fertilization applied during a season can change sward diversity and leave a legacy in herbage production and species diversity in forage crops cultivated in the following seasons. Thus, we hypothesized that different strategies of summer pasture management can change the diversity of species and yield of forage species growing in subsequent cool season. We carried out an experiment in a complete randomized block design in a 2×3 factorial arrangement, with two heights of pasture management (17 or 23 cm) associated with three nitrogen rates (50, 150 and 250 kg N ha^{-1}), all applied in the warm season only (November-April, Southern Hemisphere), while in the subsequent cool season, pastures were managed according to a single strategy, with a pre-grazing height of 20 cm. The forage accumulation of cool season species averaged 5,650±680 kg dry matter (DM) ha^{-1} in 2020 and 4,600±440 kg DM ha^{-1} in 2021, regardless of the grazing height or nitrogen rates applied in summer. There was no legacy effect of preceding management of warm season pastures (grazing height and nitrogen rates) on subsequent forage production and species diversity of a cool season pasture cultivated in a subtropical environment.

Keywords: biodiverse pasture, forage accumulation, Shannon index

Introduction

Mixing forage species in pastoral environments has the potential for a higher (or at least equivalent) production when compared to monocultures (Grace *et al.*, 2019). In these environments, factors such as nitrogen fertilization and canopy height can modulate not only forage production, but also the botanical composition of the pasture, as they mediate resource competition among species (Tamele *et al.*, 2018; Miqueloto *et al.*, 2020). In several subtropical climate environments, such as in Southern Brazil, there is a great variability in temperature but a relative stability in rainfall (Cfb climate; Koppen classification) allowing C_3 and C_4 grasses and legumes to grow in the same area, but with forage production peaks occurring at different periods (Sbrissia *et al.*, 2017). Therefore, management strategies applied during a specific growth period could, theoretically, carry an effect in the subsequent pastures growing in the same area. In this sense, considering a mixed pastoral environment under a subtropical climate, we hypothesized that combinations of summer pasture management (grazing heights and nitrogen rates) can change the diversity and yield of forage species growing in a subsequent cool season.

Materials and methods

This two-year study (December 2019 to October 2021) was conducted at Santa Catarina State University, Lages, BR (27°47'04"S, 50°18'13"W). The region has a temperate Cfb climate (Köppen–Geiger classification) with mild summers, cool winters and a well-distributed rainfall throughout the year (Alvares *et al.*, 2013). The pastures of our experiment were composed of six species. During the warm season (November-April in Southern Hemisphere), pastures were mainly composed of forage peanut (*Arachis pintoi* cv. Amarillo), kikuyu grass (*Cenchrus clandestinus* Hochst. ex Chiov.) and tifton 85 (*Cynodon* spp.), and during the cool season (May-October), birdsfoot trefoil (*Lotus corniculatus* L. cv. São Gabriel), white clover (*Trifolium repens* L. cv. Zapican) and annual ryegrass (*Lolium multiflorum* L. cv. La Estanzuela 284) were the predominant species. To test the legacy effect of summer management on the herbage production and species diversity of the following cool season crop we adopted the procedures as described: during the summer the main factors were two pre-grazing canopy heights (17 or 23 cm) and nitrogen rates (50, 150 and 250 kg N ha^{-1}) assigned in a factorial arrangement totalling 6 treatments with three replicates each. During the cool season the pastures were subjected to a single management strategy (pre-grazing height of 20 cm and 50 kg of N ha^{-1} applied in the annual ryegrass tillering stage). By using this management strategy any differences in forage production or species diversity during the cool season could be attributable to the previous summer management. Forage accumulation in the cool season was measured by the difference between pre- and post-grazing forage mass, which were estimated with a calibrated rising plate meter (RPM). The botanical composition of the sward was evaluated by cutting and separating species from three samples per paddock (0.5 m^2) collected close to the ground. This procedure was performed in two opportunities during the cool season (August and October) in order to characterize the canopy at the middle and end of the species' growing season. From these data, the Shannon Diversity Index (H′) of the paddocks was calculated. Forage diversity and accumulation data were subjected to analysis of variance, means were compared by Tukey test when $P<0.10$ using the statistical package 'agricolae' (De Mendiburu, 2009) of RStudio software (R Core Team, 2020).

Results and discussion

There was not an effect of previous warm pastures management on forage accumulation of cool season species, with average values of 5,650±680 kg DM ha^{-1} in 2020 and 4,600±440 kg DM ha^{-1} in 2021. Positive legacy effects on the production of an annual ryegrass monoculture were reported by Fox *et al.* (2020); however, the legacy effect observed by these authors occurred due to the percentage of legumes in the area prior to ryegrass sowing, since the residues was shown to have high initial mineralization rates. In contrast, the summer pastures in our experiment were composed almost exclusively of stoloniferous grasses (90%), which are known to have residues with high C:N ratio and lower mineralization rate. Moreover, *C. clandestinus* and *Cynodon* are characterized as resource-exploitative C_4 grasses, with high nutrient absorption capacity and rapid growth (Cruz *et al.*, 2002) which possibly stimulated the use of total available N during the warm season. At the beginning of the cool season, the diversity index, expressed by the Shannon Index (H'), was lower (average of 0.59 in 2020 and 0.66 in 2021) because the canopy was dominated by annual ryegrass, while at the end of the cool season there was a favourable climatic condition for the reestablishment of white clover and warm season species and the species diversity was similar among treatments in both years (average of 1.20 in 2020 and 0.83 in 2021; Figure 1).

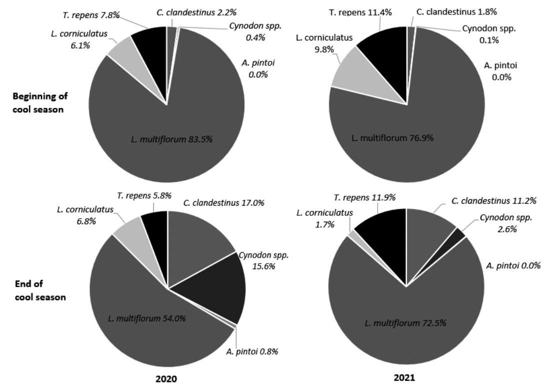

Figure 1. Botanical composition (% of species) at the beginning and end of cool season period in the years 2020 and 2021 in a mixed pasture in Southern Brazil.

Conclusions

There was no legacy effect of preceding management of warm season pastures (grazing height and nitrogen rates) on subsequent herbage production and species diversity of a cool season pasture cultivated in a subtropical environment.

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References

Alvares C.A., Stape J.L., Sentelhas P.C., Gonçalves J.L. de M. and Sparovek G. (2013) Köppen's climate classification map for Brazil. *Meteorologische Zeitschrift* 22, 711-728.

Cruz, P., Duru M., Therond O., Theau J.P., Ducourtieux C. *et al.* (2002) Une nouvelle approche pour caractériser les prairies naturelles et leur valeur d'usage. *Fourrages* 172, 335-354.

De Mendiburu F. (2009). Una herramienta de analisis estadistico para la investigacion agricola. Thesis. Universidad Nacional de Ingenieria (UNI-PERU), Peru.

Fox A., Suter M., Wildmer F. and Lüscher A. (2020) Positive legacy effect of previous legume proportion in a ley on the performance of a following crop of *Lolium multiflorum*. *Plant and Soil* 447(2), 497-506.

Grace C., Lynch M.B., Sheridan H., Lott S., Fritch R. and Boland T.M. (2019) Grazing multispecies swards improves ewe and lamb performance. *Animal* 13, 1721-1729.

Miqueloto T., Winter F.L., Bernardon A., Cavalcanti H.S., Medeiros Neto C de., Martins C.D.M. and Sbrisia A.F. (2020) Canopy structure of mixed kikuyu grass-tall fescue pastures in response to grazing management. *Crop Science* 60, 499-506.

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Sbrissia A.F., Duchini P.G., Echeverria J.R., Miqueloto M., Bernardon A. and Américo L.F. (2017) Produção animal em pastagens cultivadas em regiões de clima temperado da América Latina. *Archivos Latinoamericanos de Producción Animal* 25, 47-60.

Tamele O.H., Lopes de Sá O.A.A., Bernardes T.F., Lara M.A.S. and Casagrande D.R. (2018) Optimal defoliation management of brachiaria grass-forage peanut for balanced pasture establishment. *Grass and Forage Science* 73, 522-531.

Grazing heights do not change forage yield in a biodiverse pasture

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Abstract

Increasing the biodiversity of pastures has been considered an important tool to increase the functionality of pastoral environments without compromising their productive capacity. Despite this very welcome benefit, it is important to consider that grazing management strategies and nitrogen are strong modulators of botanical composition in more complex plant communities. We hypothesized that grazing height changes the forage yield in biodiverse pastures regardless of the nitrogen rates applied (50 to 250 kg of N ha^{-1}). The experiment was carried out in two seasons (2020-2021) and treatments were assigned to a complete randomized block design in a 2×3 factorial design, with two pre-grazing canopy heights (17 or 23 cm) associated with three nitrogen rates (50, 150 and 250 kg N ha^{-1}). Swards were composed of two grasses (*Cenchrus clandestinus* and *Cynodon* spp.) and three legumes (*Arachis pintoi*, *Trifolium repens* and *Lotus corniculatus*). The two grasses maintained similar proportions between them and together make up around of 85% of the canopy composition in all treatments. The highest nitrogen rate increased the forage accumulation. No effect of grazing height on herbage production was observed. These results highlight that more biodiverse pastures can be managed in a range of canopy heights without compromising their productive capacity regardless of the amount of nitrogen applied (50 to 250 kg of N ha^{-1}).

Keywords: legumes, nitrogen rate, grazing management

Introduction

Pasture-based production systems have become highly specialized. These pastures are usually restricted to monocultures or binary grass-legume mixed swards. However, with the growing demand for more sustainable production models, the use of more complex pastures has emerged as an option, since these ecosystems have shown to present greater production stability, resistance, and resilience to extreme climate events (Hofer *et al.*, 2016) and more efficiency in the use of resources (Tilman *et al.*, 2006) when compared to monocultures. It is well accepted that in monocultures there is a range of grazing heights where forage accumulation is relatively constant (Sbrissia *et al.*, 2018) which brings some flexibility in decision-making of grazing management at farm level. Despite this, in mixed swards both grazing height and nitrogen fertilization are strong modulators of the botanical composition of the pasture (Egan *et al.*, 2018; Tamele *et al.*, 2018), and not necessarily promote the same flexibility in forage production as those observed for grasses monocultures. Based on the above, we hypothesized that grazing height management (17 and 23 cm) changes forage yield in pastures composed of 5-species and subjected to nitrogen fertilization rates ranging from 50 to 250 kg of N ha^{-1}.

Material and methods

The experiment was carried out at the Santa Catarina State University, Lages, BR (27°47'04"S; 50°18'13"W) for two years (2020-2021). The region, according to the Köppen-Geiger classification, has a Cfb climate (temperate) with mild summers, cool winters, and rainfall is well distributed throughout the year (Alvares *et al.*, 2013). The experimental area was established in February 2018 with forage peanut (*Arachis pintoi* cv. Amarillo), kikuyu grass (*Cenchrus clandestinus* Hochst. ex Chiov.) and tifton 85 (*Cynodon* spp.), while in the cool (May-October) season of 2019 birdsfoot trefoil (*Lotus corniculatus* L. cv. São Gabriel), white clover (*Trifolium repens* L. cv. Zapican) and annual ryegrass (*Lolium multiflorum* L.

cv. La Estanzuela 284) were established. At the end of the cool season of 2019 all paddocks (315 m^2 each) were grazed, determining the beginning of the experimental period (December 2019). The experiment was assigned to a complete randomized block design in a 2×3 factorial scheme and three replications. The treatments consisted of two canopy heights (17 and 23 cm) associated with three nitrogen rates (50, 150 and 250 kg N ha^{-1}). The experiment was carried out during the warm season (December/November-April) of 2019/2020 and 2020/2021, while in the cool season, when the warm species were seriously damaged by frosts, the plots were overseeded with annual ryegrass. The stocking method used was intermittent grazing with post-grazing height targets corresponding to 60% of the pre- grazing canopy heights. The forage accumulation was obtained by the difference between the pre-grazing forage mass of 'cycle 2' and the post-grazing forage mass of 'cycle 1'. Forage mass before and after grazing was determined using a rising plate meter (RPM), previously calibrated for this pastoral environment. The botanical composition was measured from the cut of three samples per paddock (0.5 m^2) collected close to the ground. This procedure was performed in mid-summer (January) and early-autumn (April). Data were subjected to analysis of variance and means compared by Tukey test when P<0.10. The package used was 'agricolae' (De Mendiburu, 2009) from RStudio software (R Core Team, 2020).

Results and discussion

Herbage production was affected by nitrogen rates (P<0.10) and no effect was observed for grazing heights (P=0.66) or interaction between grazing height and nitrogen rates (Figure 1). This result may be related to the maintenance of a similar botanical composition between treatments, with kikuyu grass and tifton-85 making up >85% of the canopy composition. This condition possibly allowed for compensatory mechanisms between tiller size and density (Mathew *et al.*, 1995) to be expressed, resulting in a similar forage accumulation in a range of management heights. Similar results were found for kikuyu grass monocultures, which maintained similar forage accumulation between 15 and 25 cm (Sbrissia *et al.*, 2018). Thus, despite kikuyu grass and tifton-85 presenting different functional traits (Barreta *et al.*, 2021), size/density compensation mechanisms seem to operate in a similar way for both species when the plants occur together in a biodiverse pasture sward. A greater herbage accumulation (around 35%) was observed when pastures were fertilized with the greatest nitrogen rate (250 kg N ha^{-1}) compared with the lowest rate (50 kg N ha^{-1}) in both years (Figure 1). The difference in herbage production between the two years was due to water shortage in 2020. The greater accumulation of forage at the highest N dose was an expected result, since the grasses used in this experiment are characterized by high nutrient absorption capacity and rapid growth (Cruz *et al.*, 2002). Despite this, it was expected that at the lowest dose of N, the legumes would contribute more to the forage mass; however, the values were below 15% in all treatments (data not shown).

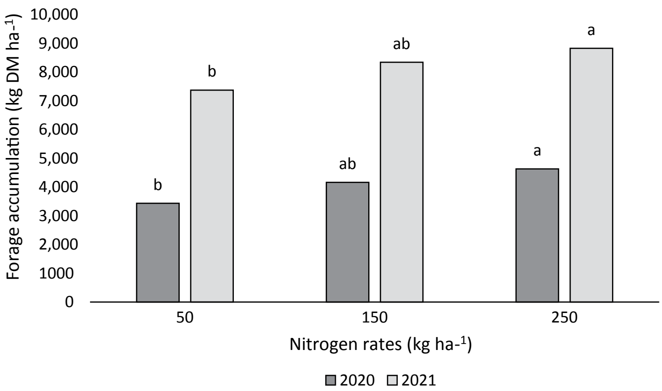

Figure 1. Herbage production (kg dry matter (DM) ha^{-1}) in mixed pastures receiving different nitrogen rates during the summer seasons (November-April) of 2020 and 2021.

Conclusions

Regardless of nitrogen rates (range 50 to 250 kg N ha^{-1}) grazing heights ranging from 17 to 23 cm do not change herbage production in a 5-species mixed sward. These results highlight that more biodiverse pastures can be managed in a range of canopy heights without compromising their productive capacity regardless of the amount of nitrogen applied (within the range 50 to 250 kg of N ha^{-1}).

Acknowledgements

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References

Alvares C.A., Stape J.L., Sentelhas P.C., Gonçalves J.L. de M. and Sparovek G. (2013) Köppen's climate classification map for Brazil. *Meteorologische Zeitschrift* 22, 711-728.

Barreta D.A., Piran F., Winter F.L., Méndez V.M.Y., Gislon F.C.S. and Sbrissia A.F. (2021) Are *Pennisetum clandestinum* and *Cynodon* spp. functionally different? 56th Annual Meeting of the Brazilian Society of Animal Science, Florianópolis, Brazil.

De Mendiburu F. (2009) Una herramienta de analisis estadistico para la investigacion agricola. Thesis. Universidad Nacional de Ingenieria (UNI-PERU), Peru.

Egan, M., Galvin, N. and Hennessy, D. (2018) Incorporating white clover (*Trifolium repens* L.) into perennial ryegrass (*Lolium perenne* L.) swards receiving varying levels of nitrogen fertilizer: Effects on milk and herbage production. *Journal of Dairy Science* 101, 3412-3427.

Heringer, I. and Moojen, E.L. (2002) Potencial produtivo, alterações da estrutura e qualidade da pastagem de milheto submetida a diferentes níveis de nitrogênio. *Revista Brasileira de Zootecnia* 31, 875-882.

Hofer D., Suter M., Haughey E., Finn J.A., Hoekstra N.J., Buchmann N. and Lüscher A. (2016) Yield of temperate forage grassland species is either largely resistant or resilient to experimental summer drought. *Journal of Applied Ecology* 53, 1023-1034.

Matthew C., Lemaire G., Sackville Halmilton N.R. and Hernádez-Garay A. (1995) A modified self-thinning equation to describe size/density relationships for defoliated swards. *Annals of Botany* 76, 579-587.

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/.

Sbrissia A.F., Duchini P.G., Zanini G.D., Santos G.T., Padilha D.A. and Schmitt D. (2018) Defoliation strategies in pastures submitted to intermittent stocking method: underlying mechanisms buffering forage accumulation over a range of grazing heights. *Crop Science* 58, 945-954.

Tamele O.H., Lopes de Sá O.A.A., Bernardes T.F., Lara M.A.S. and Casagrande D.R. (2018) Optimal defoliation management of brachiaria grass–forage peanut for balanced pasture establishment. *Grass Forage Science* 73, 522-531.

Tilman, D., Reich, P.B. and Knops, J.NM.H. (2006) Biodiversity and ecosystem stability in a decade-long grassland experiment. *Nature* 441, 629-632.

Crop rotation effect on red clover persistence in mixed grass-clover leys

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Abstract

Red clover persistence is critical for the production and quality of grass-clover leys. It is affected by management and diseases that cause decline over time. In the long-term field experiment R8-71, established in Sweden in the 1950s, red clover was included in a grass-clover mixture in different cropping systems across three sites. The grass-clover mixture was established by undersowing in barley in six-year crop rotations; the barley was followed by two, three or five years of ley. We investigated whether red clover persistence decreased with increased proportion of grass-clover in the rotation. The proportion of clover in the first harvest year decreased with each crop rotation cycle in the five-year ley (P=0.032), but no such effect was found with the other rotations. The three-year ley did not decrease the rate of red clover decline compared to the five-year ley within a crop rotation cycle on a yearly average. For both crop rotations, the red clover decline started to be noticeable in the second harvest year. Results indicate that longer breaks between leys could improve clover proportion the first harvest year, but not persistence the following harvest years.

Keywords: *Trifolium pratense*, red clover proportion, cropping systems, long-term field experiment, performance, root rot

Introduction

Red clover (*Trifolium pratense* L.) is commonly used in Swedish leys where it contributes with an increase in crude protein concentration without having to add extra nitrogen fertilizer. An issue with red clover is its poor persistence which can be affected by several different factors. This study aimed to determine the effect of crop rotation ley frequency on red clover persistence using data from the Swedish long-term field experiment R8-71. The hypothesis was that the red clover proportion would decrease at a faster rate within a crop rotation cycle and over time if the proportion of ley increased.

Materials and methods

The long-term field experiment was established in Northern Sweden in the 1950s, and the data used for this analysis are from 1963 to 1986. It was conducted at three different sites; Offer (63.14°N, 17.75°E), Röbäcksdalen (63.81°N, 20.24°E) and Ås (63.25°N, 14.56°E) (Zhou *et al.*, 2019). Four crop rotations were included that represented different directions within agriculture, ranging from livestock farming to crops more focused on human consumption (Table 1). One of the crop rotations did not include any harvests of ley (D) and was therefore excluded from this analysis. The field experiment consisted of two replicates at each site, totalling 48 plots per site. All leys were harvested twice per season except the second harvest year in the two-year ley (C) which was only harvested once. Because of this, only the first harvest year of each cycle could be compared between crop rotations A, B and C. Crop rotations A and B were also compared over three harvest years to identify differences in persistence. Each management strategy was adapted to the different crops included. For more details on management and how botanical composition was determined refer to Bergqvist (2021).

Table 1. The six-year rotation for the four different crop rotations at Offer, Röbäcksdalen and Ås.

A	Barley + undersown ley, ley 1, ley 2, ley 3, ley 4, ley 5
B	Barley + undersown ley, ley 1, ley 2, ley 3, oat + peas, rape seed
C	Barley + undersown ley, ley 1, ley 2, winter rye, potato, oat + peas
D	Barley + undersown ley, fallow, winter rye, peas, potato, carrots/swedes

For statistical analysis of the dataset, linear mixed-effect models were fitted using the software programmes JMP Pro 16.0 and SAS 9.4. The model included the explanatory factors; harvest, harvest year, crop rotation and site to explain the response of variable red clover proportion. Trend over time was modelled by a continuous explanatory variable x, defined as x = Year – 1963. Year was included as a random-effects factor, which allowed random deviations from the trend.

Results and discussion

Red clover proportion in the first harvest year of each cycle was affected by crop rotation over time, as shown in Figure 1. The two crop rotations A and B, with five and three harvest years of ley, respectively, had slopes that were significantly different from crop rotation C that had two harvest years of ley. Both crop rotations with a larger proportion of ley had a declining trend in proportion over time, but it was only significant for the crop rotation with five harvest years of ley (P=0.032), though the rotation with three harvest years of ley was close to being significant (P=0.051).

The crop rotation with a five-harvest-year ley (A) was expected to have a decline in red clover proportion over time, but it was not expected that the three-harvest-year ley (B) would have a similar trend. This suggests that if the time period between leys is less than three years there will be no effect of differences in crop rotation. A possible reason for the decrease in the three- and five-harvest-year ley is infection by clover rot (*Sclerotinia trifoliorum*). Clover rot is commonly most severe when infecting younger plants in the establishment year and first-harvest-year (Ylimäki, 1967).

In Figure 2, the interaction between harvest and harvest year is displayed (P<0.001). The three- and five-year ley systems did not significantly differ from one another within the same harvest year, which is why in this figure the two are merged. The decline over time is most likely caused by root rot, as the decrease in red clover proportion starts in the second harvest year with a following sharp decrease in the third harvest year. The presence of the pathogens causing root rot is not only a consequence of a poor crop rotation since they can survive on a broad range of hosts (Rufelt, 1979). If the decrease is due to root rot,

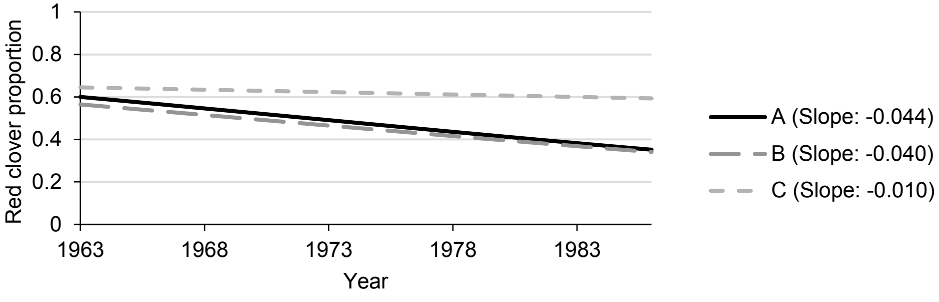

Figure 1. The change in red clover proportion in the first harvest year over time (1963-1986) in mixtures with timothy, as affected by the different crop rotations A-C with 5-, 3- and 2-years of ley, respectively. Values are averaged across harvests 1 and 2 and sites Offer, Röbäcksdalen and Ås (for explanation of A, B and C, see Table 1).

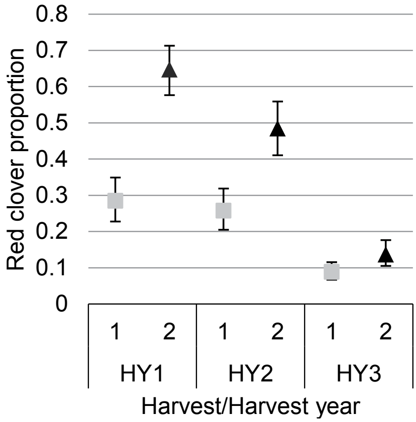

Figure 2. Effect of harvest and harvest year on red clover proportion in mixtures with timothy. Values are averaged across crop rotations A and B; sites Offer, Röbäcksdalen, Ås; and years 1963-1986. The squares and triangles represent the Least squares means of harvest one and two respectively. Error bars indicate 95% confidence interval.

this explains why there is no difference between the three- and five- harvest-year leys. In areas free from red clover diseases, red clover plants can survive for at least eight years (McBratney, 1987).

Conclusions

Red clover performance in the first harvest year of each crop rotation cycle was only benefitted by crop rotation to a certain extent. It is however uncertain if the effect is due to the length of the ley or the gap between leys in a crop rotation, or both. The persistence of red clover over three harvest years was not affected by differences in crop rotations.

Acknowledgements

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References

Bergqvist S. (2021) *Long-term effects of cropping system on red clover proportion and crude protein concentration in mixed leys*. Master's thesis.Swedish University of Agricultural Sciences, Department of Crop Production Ecology, Uppsala, Sweden.

McBratney J.M. (1987) Effect of fertilizer nitrogen on six-year-old red clover/perennial grass swards. *Grass and Forage Science* 42, 147-152.

Rufelt S. (1979) *Root rot of red clover, prevalence, cause and importance in Sweden*. Swedish University of Agricultural Sciences. Uppsala, Sweden. Plant Protection Reports, Agriculture 9. (Summary in English).

Ylimäki A. (1967) Root rot as a cause of red clover decline in leys in Finland. *Journal of the Agricultural Research Centre* 6 (18, Suppl. 1).

Zhou Z., Palmborg C., Ericson L., Dryler K., Lindgren K., Bergkvist G. and Parsons D. (2019) A 60-years old field experiment demonstrates the benefit of leys in the crop rotation. *Acta Agriculturae Scandinavica*, *Section B – Soil & Plant Science* 69(1), 36-42.

Assessing resilience of lucerne cultivars to drought stress in Wisconsin, United States

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Abstract

Increasingly, droughts challenge lucerne production and it is critically important to develop cultivars resilient to drought in the context of a changing climate. Forage yield of twenty-four lucerne cultivars was evaluated over two years under normal rain and drought stress using rainout shelters. Resilience was evaluated as the dry matter yield of stress plot divided by that of no-stress plot. Cultivars were not different for mean resilience across years, but some cultivars had consistently higher resilience values than others. Resilience was negatively associated with forage yield in normal conditions.

Keywords: alfalfa, resilience, climate change, drought stress

Introduction

Lucerne (*Medicago sativa* L.) is the most important perennial forage legume, produced primarily for high-quality hay and silage for cattle (Undersander *et al.*, 2011). Lucerne growers (dairy and beef farmers) across the US are facing a challenge of losing lucerne area caused by extreme climatic conditions. The lowest lucerne area of hay harvested, and total production was due to the drought in 2012, which was the driest year in the last 15 years across United States (NOAA, 2021). It is necessary to better understand resilience of lucerne cultivars to climate stresses. Resilience is defined as the ability of a system to withstand and keep producing under a crisis. A previous analysis of historical data suggested that lucerne cultivars may differ in their resilience to drought across locations (Picasso *et al.*, 2019). This study aims to quantify the resilience of lucerne cultivars to drought stress in experimental field conditions.

Materials and methods

Lucerne drought-stress field experiments were sown on 23 May 2019 in Arlington, Wisconsin, USA (43°18′9.47″ N, 89°20′43.32″ W). The treatment design was a factorial with two factors: (1) drought stress with two levels: normal rainfall and rainout shelter; and (2) lucerne cultivar with 24 levels (modern cultivars selected based on their diverse morphological traits from three major seed companies: Corteva, FGI, and S&W, and two historical cultivars as controls). The experimental design was a split-plot, with a completely randomized block design with two replications for the main plot (drought stress) and cultivar as the split-plot. Each rainout shelter consisted of three low tunnels located side by side, with movable plastic covers on both sides of the shelter. Forecasted rainfall events greater than 2 mm were excluded (i.e. rainout shelters were closed before the expected rain) after the first lucerne harvest each year. However, if severe storms or high winds were forecast, the rainout shelters were kept open to protect the experimental infrastructure. The decision to allow full rain before the first harvest was to ensure that the subsequent drought stress was not extreme, and we could harvest forage biomass throughout the entire growing season. Forage biomass was harvested three times per year over two production years (May 27, Jun 14, Aug 24 in 2020, and May 25, July 12, Sept 7 in 2021). Subsample of harvested material of each split-plot was collected and weighed before and after being dried at 65 °C to determine dry matter. Resilience for each lucerne cultivar by block was determined by dividing the yield under drought stress by yield under normal rainfall. Analysis of variance was used for forage yield using year, drought stress, cultivar, their interactions, and block as fixed effects. For resilience, the analyses of variance considered year, cultivar, their interaction, and block as fixed effects. Differences were considered significant at $P \leq 0.05$.

Results and discussion

Drought stress (i.e. % of rain excluded) during June-August was 49% in 2020 and 46% in 2021 (Table 1). Therefore, the goal of mimicking mild drought conditions in the summer was achieved, which is a likely scenario with current climate predictions. High intensity storm events were not excluded, which is also consistent with current climate predictions.

Lucerne yields under drought stress were lower than under normal rainfall only at the third and annual total harvests in 2020, when normal rainfall was similar to the historic average. In 2021, when rainfall was below average, no differences were detected (Table 2).

Cultivars were different in yields at each harvest in both years. Drought stress resulted in lower yields than normal rainfall in the third harvest in 2020. No interaction between cultivar and drought stress was found any year. Resilience was relatively high overall, and not different among cultivars across years (Table 3), ranging from 0.67 to 0.94 in 2020, and 0.51 to 1.2 in 2021.

However, some cultivars were consistently higher in resilience values than others (e.g. VERNAL, a historic control). The experiment lacked power to identify differences in resilience of cultivars, given that only two replications were used.

Lucerne cultivar resilience was negatively associated with yield under normal rainfall across two production years (Figure 1). This is consistent with results from historical analyses (Picasso *et al.*, 2019). Cultivars with higher yield potential under normal rainfall yielded similar to other cultivars with lower potential during drought stress.

Table 1. Rainfall (mm) and % stress (excluded/total) in each season of the experiment.

Variable	2020	2021
Rainfall March-May, before first harvest (mm)	133	123
Rainfall June-August, between first and third harvest (mm)	311	238
Excluded rain June-August (mm)	153	110
% Stress during June-August	49%	46%

Table 2. Mean DM forage yield (kg ha^{-1}) and resilience to drought (RD) per harvest, for the stress experimental period, and annual across 24 cultivars.[1]

Variable	2020			2021		
	Drought stress	Normal rainfall	RD	Drought stress	Normal rainfall	RD
First harvest (May) – no drought stress applied	2,866	2,994		3,861	3,678	
Second harvest (July)	3,582	4,317	0.82	3,199	3,487	0.92
Third harvest (August)	1,664b	2,692a	0.68	1,172	1,507	0.78
Total during stress period (Second + Third)	5,246	7,009	0.75	4,371	4,994	0.88
Annual total harvest (First + Second + Third)	8,153b	10,003a	0.82	8,233	8,671	0.95

[1] Means with different letters are different for that variable within each year.

Table 3. Sum of second and third forage harvests DM yield (kg ha^{-1}) and resilience to drought (RD) for that period of five lucerne cultivars (for space limitations 19 other cultivars were omitted).[1]

Cultivar	2020			2021		
	Drought stress	Normal rainfall	RD	Drought stress	Normal rainfall	RD
VERNAL	5,076	5,414	0.94	4,234	4,261	0.99
ONEIDAVR	4,689	6,082	0.77	3,873	4,702	0.82
S&W-5	5,731	7,887	0.73	4,774	4,918	0.97
Alforex-1	4,982	7,092	0.70	3,643	4,506	0.81
FGI-8	5,626	7,953	0.71	4,314	5,407	0.80

[1] No differences were found.

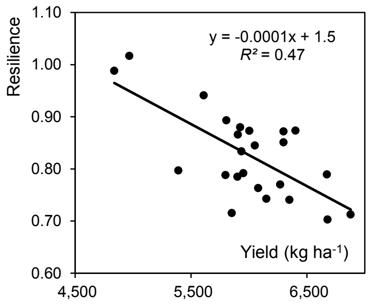

Figure 1. Resilience vs yield (second and third harvests) under normal rainfall of 24 lucerne cultivars in two years.

Conclusions

This study provided experimental evidence of a negative association between forage yield under normal conditions and resilience to drought stress. Increasing resilience in forage production will require addressing this trade-off in lucerne breeding programs.

Acknowledgements

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References

NOAA (2021) National Centers for Environmental information, Climate at a Glance: U.S. Time Series, Palmer Drought Severity Index (PDSI). Available at: http://www.ncdc.noaa.gov/cag.

Picasso V.D., Casler, M.D. and Undersander, D. (2019) Resilience, stability, and productivity of alfalfa cultivars in rainfed regions of North America. *Crop Science* 59, 800-810.

Undersander D., Cosgrove D., Cullen E., Grau C., Rice M.E., Renz M., *et al.* (2011) Alfalfa management guide. ASA, CSSA, SSSA, Madison, WI, USA. https://doi.org/10.2134/2011.alfalfamanagementguide.

Overseeding and rehabilitation of degraded upland grasslands after *Arvicola terrestris* outbreaks

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Abstract

Outbreaks of grassland rodent (water vole: *Arvicola terrestris*) populations can cause dramatic grassland damage, impacting grassland structure and function, as well as the provision of ecosystem services. There is a pressing need to identify effective management techniques which promote grassland recovery after rodent disturbance, and clear guidelines on soil preparation and species mixtures are currently lacking. We have set up a field-scale experiment to examine the interactive effects of mechanical soil treatments and seed mixtures on an upland permanent grassland experiencing an *A. terrestris* outbreak. The ultimate objective is to determine which management practices satisfy two key criteria: (1) sufficient hay production to support a dairy herd during the winter; and (2) limited adverse effects on plant community recovery and longer-term grassland biodiversity. We test two types of soil preparation (with or without use of a disc cultivator) in combination with five overseeding treatments (no seed, annual species in monoculture, mixtures of annual species), and record biomass production, forage quantity and quality and soil properties over a two-year period. Aerial pictures and trapping are used to estimate rodent population density and dynamics. First results are presented and suggest effects of both seed mix and soil preparation treatments on hay quantity and quality.

Keywords: water vole, grassland recovery, management practices, seed mix, soil preparation

Introduction

Permanent grasslands are the main source of fodder for livestock in upland and mountain areas, both for grazing and/or hay production. Aside from climate-driven threats to grassland structure and function, such as drought-induced decreases in grassland productivity, European grasslands also face risks of recurrent rodent outbreaks, with implications for farming sustainability (Jacob *et al.,* 2020). For example, the fossorial water vole *Arvicola terrestris* lives in underground burrows in the grasslands of mountainous areas and is characterised by population outbreaks every 5-9 years (Berthier *et al.,* 2014). These dramatic increases in rodent abundance may have long-lasting effects on plant communities, grassland productivity and forage quality (Quéré *et al.,* 1999, Nicod *et al.,* 2020). A variety of chemical (rodenticides) and agrotechnical approaches can be used for rodent management, but once high-density populations of rodents are present, field control of rodents is forbidden in France (Arrêté du 14 mai 2014). In such situation, there is therefore a pressing need to identify management techniques that promote short-term forage production and longer-term plant community recovery. Overseeding techniques are one promising option for the renovation of degraded grasslands, offering the possibility to ensure sufficient hay production for winter feeding while limiting adverse effects on grassland biodiversity, but clear guidelines on soil preparation and seed mixtures are currently lacking. Here we aimed to examine the importance of soil disturbance and diversity of seed mix for the diversity and forage production in overseeded grasslands exposed to high levels of rodent damage.

Materials and methods

We implemented a field-scale experiment at the INRAE-Herbipole experimental farm (https://doi.org/10.15454/1.5572318050509348E12) in the Massif Central region of France (45.30°N, 2.84°E, 1080 m a.s.l.), following a rodent outbreak on an 18 ha area of permanent grassland in early 2021. Interactive effects of mechanical soil treatments and seed mixtures were assessed using two types of soil preparation (SP) (with or without disc cultivator [DC]), and five overseeding treatments (OT): no seed [Control]; *Avena strigosa* 50 kg ha^{-1} [A]; *Avena* + *Lolium multiflorum* 15 kg ha^{-1} [AL]; *Avena* + *Vicia sativa* 25 kg ha^{-1} [AV]; *Avena* + *L. multiflorum* 12.5 kg ha^{-1} + *Trifolium incarnatum* 6 kg ha^{-1} + *V. sativa* 4 kg ha^{-1} + *Trifolium squarrosum* 2.5 kg ha^{-1} [MIX]. Overseeding treatments represented a gradient of diversity, and all sown species were annuals known to be able to grow at the study site, with a limited risk of regrowth the following year. Each experimental treatment represented an area ranging from 1.1 to 1.4 ha. In the two weeks prior to seeding and after harvesting, rodent populations (RP_1 and RP_2) were estimated by trapping (TopCat ©, Andermatt, France) all rodents in a 400 m^2 area over a 5-h period in each treatment. Aerial pictures were taken in all treatments to estimate soil cover rate before seeding (SCR_1) and after harvesting (SCR_2), defined as the proportion of visible fresh soil. Seeding was carried out using a 3 m-large seed drill on 26 May. On 27 July aboveground biomass was determined in each treatment (eight 50×50 cm quadrats cut to a height of 5 cm, samples oven-dried for 48 h at 60 °C then weighed). Plots were mowed on 9 August and forage yields were estimated for ground-cured hay by weighing the amount of dry hay on the ground in three 20 m^2 squares per treatment. Hay samples were analysed using NIRS techniques, and the feed value was estimated using existing equations (INRA, 2018). Data on hay quantity and quality were statistically analysed using general linear models, with soil treatment and seed mix as fixed effects. Rodent numbers were regressed against levels of soil cover and levels of soil cover were also compared over time using linear models.

Results and discussion

Delattre and Giraudoux (2009) estimate that at over 200 rodents ha^{-1} the situation can be considered as critical for the plots. The presence of *A. terrestris* was high in all treatment plots (337±129 rodent ha^{-1}) and rodent numbers increased by 61% during the trial, suggesting that rodent populations were still in a growth phase. Average fresh soil cover prior to seeding was 37.4±16.8% across plots and SCR_2 was 51.4±10.8%. SCR_1 was marginally correlated with RP_1 (P=0.06) but SCR_2 showed no relationship with RP_1 or RP_2, suggesting that these are not good indicators of rodent population size and dynamics at the plot scale. Soil preparation had no effect on SCR_2, but SCR_2 was positively correlated to SCR_1 (P=0.002).

Effects of seed mix on yield varied depending on soil preparation treatment (SP×OT interaction) (Table 1). With the exception of AV, all seeding treatments showed decreased yields with the disc cultivator (-23% on average). Differences between biomasses and yields of the different seed mixes correspond to losses at harvesting (-16% on average). The most important losses were recorded for AV (-35%, P=0.03), probably due to the loss of *V. sativa* leaves during tedding; AL and MIX treatments did not show significant yield losses (P>0.1).

Forage mineral content, reflecting soil presence in the hay samples, was lower in DC treatments for all seed mixes except AV (Table 1). At the same time, the disc cultivator treatment generally increased forage nitrogen content and decreased cellulose content, resulting in higher NEL levels.

Table 1. Production and nutritive values of forage from the different experimental treatments.[1]

	Soil preparation		Overseeding treatment					*P*-value[2]		
	Control	Disc	Control	A	AL	AV	MIX	SP	OT	SP×OT
Biomass (t ha^{-1})	2.67	2.27	2.64	2.86	2.64	2.10	2.11	*	**	NS
Yield (t ha^{-1})	2.36	1.83	2.02	2.31	2.99	1.35	1.82	***	***	***
Minerals (g kg^{-1})	98	96	94	85	107	102	97	NS	***	*
CP[3] (g kg^{-1})	104	117	108	95	112	123	113	**	**	NS
Cellulose (g kg^{-1})	324	316	312	338	328	301	321	NS	***	NS
NEL[3] (Mcal kg^{-1})	1.36	1.43	1.37	1.36	1.36	1.44	1.41	*	NS	NS

[1] Results for GLMM are shown; soil preparation treatment is given by SP, overseeding treatment is given by OT [A = *Avena strigosa*; AL = A + *Lolium multiflorum*; AV = A + *Vicia sativa*; MIX = AL + *Trifolium incarnatum* + *V. sativa* + *Trifolium squarrosum*].

[2] *** $P<0.001$; ** $P<0.01$; * $P<0.05$; NS (not significant) $P\geq0.05$.

[3] CP = crude protein; NEL = net energy for lactation.

Conclusions

Preliminary results suggest that both soil preparation and seed mix composition have a significant effect on forage quantity and quality following overseeding. Decreases in yield induced by the disc cultivator were at least partly compensated for by higher quality hay. Acceptable fodder quantity and quality were obtained with simple mixes (A, AL) and were not significantly improved by seeding with a more diverse mixture. Subsequent measurements will examine whether effects of overseeding on hay production persist in time and will address impacts on plant community recovery. However, impacts of the treatments have to be balanced by the fact that *A. terrestris* population seemed to be in a growth phase and that seeding may extend the length of the outbreak. Economic analyses are also required to determine the cost-effectiveness of using a disc cultivator as part of the overseeding procedure.

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References

Arrêté du 14 mai 2014 relatif au contrôle des populations de campagnols nuisibles aux cultures ainsi qu'aux conditions d'emploi des produits phytopharmaceutiques contenant de la bromadiolone (2014) NOR: AGRG1300885A.

Berthier K., Piry S., Cosson J.F., Giraudoux P., Foltête J.C., Defaut R., Truchetet D. and Lambin X. (2014) Dispersal, landscape and travelling waves in cyclic vole populations. *Ecology Letters* 17, 53-64.

Delattre P. and Giraudoux P. (2009) *Le campagnol terrestre: Prévention et contrôle des populations*, Editions QUAE, Versailles, France, 263 pp.

INRA (2018) *Alimentation des ruminants*, Editions QUAE, Versailles, France, 728 pp.

Jacob J., Imholt C., Caminero-Saldaña C., Couval G., Giraudoux P., Herrero-Cofreces S., Horvath G., Luque-Larean J.J., Tkaflec E. and Wymenga E. (2020) Europe-wide outbreaks of common voles in 2019. *Journal of Pest Science* 93, 703-709.

Nicod C., Couval G., Giraudoux P. and Gillet F. (2020) Vole disturbance and plant community diversity in a productive hay meadow. *Acta Oecologica* 106, 103585.

Quéré J.P., Garel J.P., Rous C., Pradier B. and Delattre P. (1999) Estimer les dégâts du campagnol terrestre en prairie naturelle. *Fourrages* 158, 133-147.

Flowers in the grassland – management for nature based dairy farming

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Abstract

Grassland management is the key to improve biodiversity and the provision of ecosystem services in permanent grasslands. An overview on the effects of current management practices on species rich grasslands on dairy farms in the Netherlands is, however, lacking. In this research we aim to quantify the relationship between plant diversity, yield and grass quality along a gradient of management intensity. The Alblasserwaard region in the Netherlands is the case study region, where we collect field data and carry out farmers' interviews. Data from the first year of the research provide a clear relationship between plant diversity, yield, grass quality and management intensity. High management intensity of grasslands supports low plant diversity but results in high yields and high grass quality. Moderate management intensity supports intermediate plant diversity, a moderate yield and good grass quality. Moderate management may be of interest for implementation in dairy farming systems on a larger scale. Grasslands with highest plant diversity and a distinctive botanical composition were managed extensively. Grass quality and yields in extensively managed grasslands were low.

Keywords: management intensity, plant biodiversity, yield and grass quality

Introduction

There is growing concern about the widespread decline of biodiversity in permanent grasslands managed for dairy production, but an integrated understanding on the effects of current management and the intensity of the management on plant species richness, herbage yield and grass quality on Dutch dairy farms on peat soils is lacking. Currently, the majority of grasslands on peat soils in the Netherlands are managed at high intensity allowing for the production of high yields of grass of high quality. In these grasslands plant species diversity is low, thereby failing to support biodiversity of higher trophic levels such as birds and pollinators (Tanis *et al.*, 2020) and the provision of multiple ecosystem services is limited. Intensive management of grasslands on peat soils can lead to enhanced soil subsidence and increased greenhouse gas emissions (Smolders *et al.*, 2019). For the last 20 years agri-environmental schemes targeted bird protection or conservation of plant species richness in grasslands. Nowadays the focus has shifted to combined benefits of species rich grasslands for dairy production, biodiversity support and ecosystem services provision. Dairy cooperatives in the Netherlands started paying premium prices to farmers who manage species rich grasslands. The Alblasserwaard region has some species rich grasslands, mostly part of nature reserves, organic farms or agri-environmental contracts. Many farmers in the Alblasserward are interested in increasing biodiversity on their land. In the 'green circle' network of the Alblasserwaard, multiple stakeholders from policy, science, a dairy cooperative, individual farmers, a bank, nature organizations and the agri-environmental cooperative collaborate to support the transition to nature-based farming. In our research we aim to find the relationship between plant diversity, yield and grass quality along a gradient of management intensity on a wide range of permanent grasslands on dairy farms on peat soils in the Alblasserwaard in the Netherlands.

Materials and methods

The grasslands assessed in this project are located in the Alblasserwaard (51°52'14.39' N 4°48'1.79' E) and Vijfheerenlanden (51°54'15.59' N 5°05'5.40' E) in the provinces of South Holland and Utrecht in the Netherlands, where the average grass growing season is 310 days with an average temperature of 10 °C and average annual precipitation of 900 mm. The main soil types of the region are peat soils with a clay layer of various thicknesses (0-50 cm) on top. The altitude of the grasslands in this region is between 0 to 1.75 m below sea level.

In preparation of the field work, in spring 2020 we selected 3 permanent grasslands with high plant diversity, 6 permanent grasslands with intermediate plant diversity, and 4 permanent grasslands with low plant diversity according to the Dutch grassland typology (Schippers *et al.*, 2012). The management of the grasslands was stable for at least 10 years. Measurements on herbage yield and grass quality were carried out when farmers planned their regular cutting or grazing activities. Before grazing, four enclosure cages (1.2×4.2 m) were placed in the field. Per field four samples were taken with a mower (1.2×5 m) at 5 cm cutting height per defoliation event. Grass height was measured with the rising plate meter before and after mowing. Grass samples were oven dried at 70 °C and a fresh sample was sent for analysis (Weende analysis, Tilly and Terry and sugar content analysis). Botanical composition was assessed in October 2020 by the Braun Blanquet method on four vegetation plots (5×5 m) within each field. The information on the botanical composition was used to calculate the Shannon diversity index and the percentage of different plant groups represented in the total yield. To quantify management intensity of grasslands (Blüthgen *et al.*, 2012), detailed information on defoliation intensity by cutting or grazing regimes, grazing intensity and information on the amount and quality of fertilization on the grasslands was obtained through interviews with the farmers. This information was converted into kg per hectare of nitrogen (N) and phosphorus (P) fertilization per year. In summer 2020 manure samples were taken and analysed for N and P content.

Results

In the first year we observed a clear relationship between plant diversity, yield and management intensity (Table 1). Grasslands receiving high levels of animal manure and artificial fertilizer (411 kg N ha^{-1} yr^{-1} and 73 kg P ha^{-1} yr^{-1}) supported a low plant diversity but reached a high yield (15 t dry matter (DM) ha^{-1} yr^{-1}). These grasslands are more frequently cut and utilized at a younger growth stage, leading to high energy content in the grass (883 VEM energy kg^{-1} DM^{-1}) and a high protein yield (2,836 kg crude protein ha^{-1} yr^{-1}) (Figure 1). Management of grasslands with intermediate plant diversity is more variable. On many of these grasslands first mowing dates are delayed for bird protection until mid-June. They are utilized by cutting and grazing, and fertilization is mainly as solid manure. Three out of six grasslands are under organic certification. The yield (9 t DM ha^{-1} yr^{-1}), with energy grass content (767 VEM energy kg^{-1} DM^{-1}) and a crude protein yield (1,274 kg CP ha^{-1} yr^{-1}), was composed of grasses (60%), herbs (34%) and legumes (5.7%), other plants (0.4%) and sedges (0.3%). Highest content of manganese (702 mg kg^{-1} dm^{-1}) and zinc (315 mg kg^{-1} dm^{-1}) were measured in the grass. The proportion of grasslands with intermediate plant diversity ranges from 18 to 100% between farms. Extensively managed grasslands received no manure (two grasslands) or only limited nutrient inputs that were excreted during grazing (one grassland). These extensively managed grasslands are characterized by a unique botanical composition (Shannon diversity index 2.53) with the highest plant species richness (22.5). However, yield (5 t DM ha^{-1} yr^{-1}), protein yield (3,564 kg CP ha^{-1} yr^{-1}) and energy content in the grass (683 VEM energy kg^{-1} DM^{-1}) were all low. In these grasslands, herbs (48%) and legumes (4.3%) dominated the sward. Other plant groups were grasses (41%), sedges (5.5%) and other plants (1.2%).

Table 1. The average management intensity, yield and information on botanical composition of grasslands with low, intermediate and high plant diversity in the Alblasserwaard sampled in 2020.[1]

	High plant diversity	Intermediate plant diversity	Low plant diversity
Number of fields	3	6	4
Defoliation intensity (number of cutting and grazing events per year)	2.7	4.3	6*
Grazing intensity (livestock unit days per ha per year)	30.8	127	0*
N fertilization of manure, artificial fertilizer and excretion during grazing per year (kg N ha^{-1} $year^{-1}$)	45.2	102.7	411*
P fertilization of manure, artificial fertilizer and excretion during grazing per year (kg P ha^{-1} $year^{-1}$)	11.7	30.1	73*
Yield (kg DM ha^{-1} $year^{-1}$)	5,082	8,679	14,795
Plant richness (number of plants per plot)	22.5	17.8	10.6
Shannon diversity index	2.53	2.18	*

[1] Incomplete data set or data set not yet processed (indicated by *).

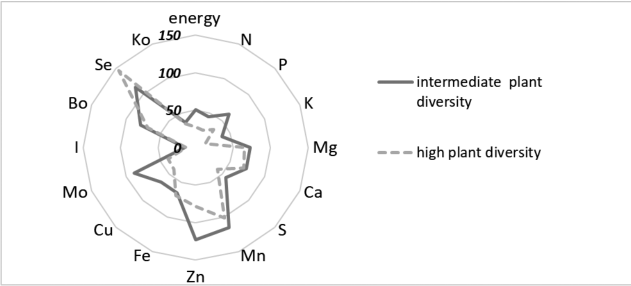

Figure 1. Results from grasslands with intermediate plant diversity (intermediate management) and high plant diversity (extensive management) are compared to grasslands with low plant diversity (intensive management). The results of grasslands with low plant diversity are set to 100%. Yield characteristics, nutrient and macronutrients are expressed as % content of the yield.

Conclusions

Management intensity was positively related to grass yield and quality, while it was negatively related to botanical diversity. Hence there is a trade-off between grassland yield and quality, and grassland biodiversity. Farmers tend to combine a few fields with extensively managed grasslands with more fields of intensively managed grasslands. Extensively managed grasslands supported a unique botanical composition. Farming at intermediate management intensity, reaching intermediate level of plant diversity, may be an interesting option for dairy farming systems with a sufficiently large farm size to reconcile production and biodiversity outcomes.

References

Blüthgen N., Dormann C.F., Prati D., Klaus V. H., Kleinebecker T., Hölzel N., ... and Weisser W.W. (2012) A quantitative index of land-use intensity in grasslands: Integrating mowing, grazing and fertilization. *Basic and Applied Ecology* 13(3), 207-220. https://doi.org/10.1016/j.baae.2012.04.001

Schippers W., Bax I. and Gardenier M. (2012) *Ontwikkelen van Kruidenrijk Grasland*. Available at: https://library.wur.nl/WebQuery/groenekennis/2029832

Smolders *et al.* (2019) Kansen voor krabbenscheer in voedselrijke sloten van het veenweidegebied |B-WARE. Available at: https://www.b-ware.eu/node/7272

Tanis M.F., Marshall L., Biesmeijer J.C. and van Kolfschoten L. (2020) Grassland management for meadow birds in the Netherlands is unfavourable to pollinators. *Basic and Applied Ecology* 43, 52-63. https://doi.org/10.1016/j.baae.2019.12.002

Strong beneficial effects of grassland sward diversity on reducing nitrous oxide emissions and emissions intensity

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Abstract

Decoupling nitrous oxide (N_2O) emissions from grassland production systems will be fundamental to meeting legislative greenhouse gas (GHG) reduction targets and thus mitigating climate change. In recent years, there has been increased uptake of multi-species swards in intensive production systems due to the associated multi-functional benefits including increased nitrogen use efficiency and yield production. In this field trial, we monitored N_2O emissions and forage yield for a full year from different sward types including monocultures and mixtures of grasses, legumes and herbs. A simplex experimental design was used to model the effect of species identity and diversity on N_2O emissions and emissions intensity (the cost of N_2O per unit nitrogen (N) or dry matter (DM) produced). Higher inorganic N fertilizer application resulted in significant increases in N_2O emissions. Species identity rather than interactions was the main driver of N_2O emissions with higher legume proportion significantly increasing N_2O emissions. Regarding emissions intensity, the same N yield or DM yield could have been produced from the 6-species mixture as a *L. perenne* monoculture (both receiving 150 kg N ha^{-1} $year^{-1}$) while reducing N_2O emissions by 41 and 24%, respectively. Overall, this study solidifies the role of multi-species swards in climate smart grassland production systems.

Keywords: multi-species swards, nitrous oxide, emissions intensity

Introduction

Nitrous oxide is a potent greenhouse gas with 265 times the global warming potential of carbon dioxide, inorganic nitrogen N fertilizer being a major source of emissions (from both production and land application). Conventional intensive grassland monocultures are highly dependent on inorganic N inputs to maintain yields. More recently, there has been increased interest in multi-species swards (MSS) (diverse swards containing mixtures of grasses, legumes and herbs). Multi-species swards can maximize yield production (Finn *et al.*, 2018) and improve nitrogen use efficiency (Suter *et al.*, 2021). Regarding GHG environmental impact, emissions intensity is a useful measure of sustainability, denoting the 'cost' of emissions per unit output. Prior to this study, there has been no long-term (1 year or more) assessments of N_2O emissions in relation to sward diversity at field scale, considering emissions intensity and/or using a full simplex design. The main objective of this experiment was to quantify the effect of grassland sward composition on N_2O emissions and emissions intensity for a full year.

Materials and methods

Nitrous oxide emissions and forage yield were monitored on a plot experiment for a full year from March 2018 – March 2019. Nitrous oxide emissions were quantified using static chamber methodology. Additionally, N_2O emissions were divided by either N yield or DM yield to calculate emissions intensity. Plots consisted of systematically varying sward compositions containing 1-6 species from three distinct functional groups: grass (*Lolium perenne* and *Phleum pratense*), legume (*Trifolium pratense* and *Trifolium repens*) and herb (*Cichorium intybus* and *Plantago lanceolata*). All plots were irrigated according to historical rainfall patterns during the summer of 2018 to avoid extreme drought conditions. A simplex experimental design was used to assess the impact of species identity and to determine if interactions occurred between species and/or functional groups that resulted in either positive (synergistic) or negative (antagonistic) effects (Kirwan *et al.*, 2009) on responses (N_2O-N, N yield-scaled and DM yield-scaled N_2O emissions). Inorganic N fertilizer was applied in calcium ammonium nitrate form at a uniform rate of 150 kg N ha^{-1} $year^{-1}$ divided into five split applications. There was an additional 300 kg N ha^{-1} $year^{-1}$ *L. perenne* monoculture treatment to offer a high N input comparison, typical of conventional, intensive production systems.

Results and discussion

There was a strong effect of N fertilizer on N_2O emissions as increasing N application to a *L. perenne* monoculture from 150 to 300 kg N ha^{-1} $year^{-1}$ significantly increased N_2O emissions by 56% (Figure 1). These results are in line with numerous previous findings that highlight the inefficiency of applying inorganic N beyond plant requirements whereby excessive pools of N in the soil system are subsequently leached or lost into the atmosphere as N_2O (e.g. Cardenas *et al.*, 2019).

Regarding sward composition, species identity rather than interactions was found to be the main determinant of N_2O emissions. Nitrous oxide emissions increased with greater legume proportion (rather than grass or herb) within the sward (Figure 2). It is worth noting that N fertilizer application was not reduced to account for N input through biological N fixation and N input through the decomposition of N-rich plant residues. This should be considered when using legumes as cover crops to mitigate against excessive N_2O losses.

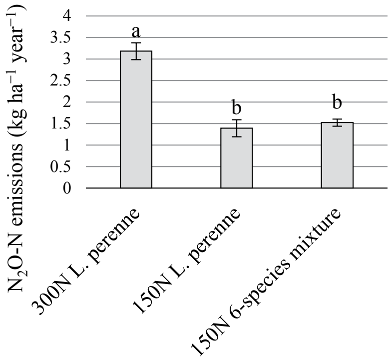

Figure 1. Annual N_2O emissions (N_2O-N emissions (kg ha^{-1} $year^{-1}$).

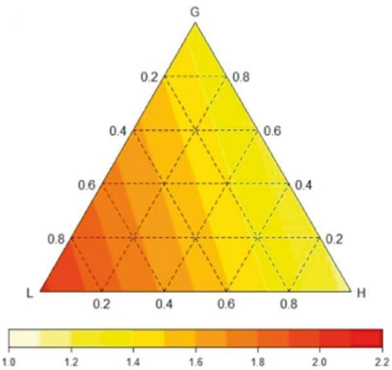

Figure 2. Ternary diagram of annual N_2O emissions according to functional group proportion.

Of notable importance was the significantly reduced N-yield scaled emissions intensity of the 6-species mixture compared to *L. perenne* at higher and equal N inputs (Figure 3). The 6-species mixture also had lower DM yield-scaled emissions intensity than *L. perenne* at 300 kg N ha^{-1} $year^{-1}$ (Figure 4). These results derive from the higher yield produced by the 6-species mixture at equal or lower level of N input, thus lowering the amount of N_2O emitted per unit forage produced. This is crucial for agricultural production systems that target the optimum balance between maximum food production and minimum negative impact on the environment.

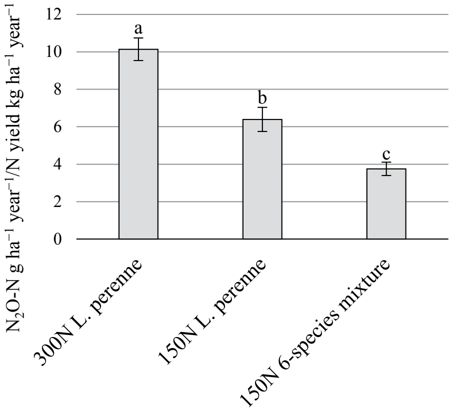

Figure 3. Nitrogen yield-scaled N_2O emissions (N_2O-N g ha^{-1} $year^{-1}$ / N yield kg ha^{-1} $year^{-1}$).

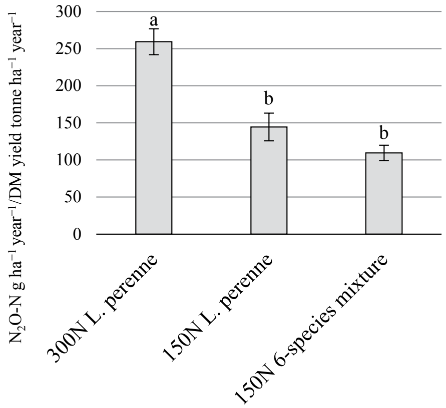

Figure 4. Dry matter yield-scaled N_2O emissions (N_2O-N g ha^{-1} $year^{-1}$ / DM yield tonne ha^{-1} $year^{-1}$).

Conclusions

This study provides further evidence that multi-species swards have an important role to play in improving the sustainability of intensively managed grassland production. Findings also reiterate the inefficiency of applying inorganic N fertilizer beyond plant requirements.

References

Cardenas, L.M., Bhogal, A., Chadwick, D.R., McGeough, K., Misselbrook, T., Rees, R.M., Thorman, R.E., Watson C.J., Williams J.R., Smith K.A. and Calvet S. (2019) Nitrogen use efficiency and nitrous oxide emissions from five UK fertilized grasslands. *Science of the Total Environment* 661, 696-710.

Finn J.A., Suter M., Haughey E., Hofer D. and Lüscher A. (2018) Greater gains in annual yields from increased plant diversity than losses from experimental drought in two temperate grasslands. *Agriculture, Ecosystems & Environment* 258, 149-153.

Kirwan L., Connolly J., Finn J.A., Brophy C., Lüscher A., Nyfeler D. and Sebastià M.T. (2009) Diversity-interaction modeling: estimating contributions of species identities and interactions to ecosystem function. *Ecology* 90(8), 2032-2038.

Suter M., Huguenin-Elie O. and Lüscher A. (2021) Multispecies for multifunctions: combining four complementary species enhances multifunctionality of sown grassland. *Scientific Reports* 11(1), 3835.

Coexistence of geese and grassland – new grassland mixtures tolerating geese grazing

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Abstract

Climate change results in longer growing season, benefitting forage crop production in northern Norway. Wild goose populations take advantage of the increased access to this high-quality feed. European goose populations are increasing, triggering conflicts and economical losses for farmers. A warmer climate may open for higher yielding seed mixtures, with better tolerance against goose grazing. We tested eight different seed mixtures by adding five forage species in various combinations to a traditional, commercial seed mixture in a randomized block design, three replicates. Goose grazing was simulated by weekly cutting small plots (0.25 m^2) fixed within 10.5 m^2 larger plots. Cumulated biomass in the weekly cut small plots was compared to total yields from the large plots, harvested twice according to normal practice. No significant differences in biomass accumulation between seed mixtures of the weekly cut plots were identified, possibly due to large variation between replicates, harvest years and cutting regime. However, results indicate that several of the new mixtures containing *Dactylis glomerata* are higher yielding and tolerate intensified cutting better than the traditional mixtures. This suggests that traditional, commercial seed mixtures are not the best for grasslands subjected to intensive geese grazing.

Keywords: goose grazing, Northern Norway, *Dactylis glomerata*, field study, simulated grazing

Introduction

In Northern Norway, agriculture is climatically marginal, limited by a short growing season and low temperatures. Forage production for sheep, cattle and dairy is the main form of production. However, observed and predicted climate change has resulted in a longer thermal growing season, which may give the potential for increased yields and the possibility to introduce new, promising forage species (Olesen and Bindi, 2002; Uleberg *et al.*, 2014).

Climate change also benefits European goose populations with higher reproductive success, better winter survival and earlier migration (Fox and Madsen, 2017; Tombre *et al.*, 2008). Their increasing populations intensify the conflicts with agricultural interests, as they prefer to graze on forage crops instead of their natural habitats. This results in economic losses for many farmers as they lose yields for winter fodder for livestock (Bjerke *et al.*, 2021; Olsen *et al.*, 2017). In a field experiment in Tromsø, Northern Norway, we investigated if new seed mixtures tolerate intensive clipping, simulating intensive goose grazing, better than traditional mixtures.

Materials and methods

In 2019, a randomized block design field trial was established in Tromsø (69°40'N), Norway. Five new forage species were added in various combinations to a basic commercial seed mixture, designed for grazing and silage (C1, Table 1): *Dactylis glomerata*, *Festuca arundinacea*, × *Festulolium*, *Bromus inermis* and *Trifolium pratense* (MIX 3-8). Also, a commercial mixture designed for silage was tested (C2). Botanical composition was visually estimated in percentage per plot. In the following two years, large silage plots (10.5 m^2) were cut twice per season, the custom for the region. Goose grazing was simulated by weekly cutting small plots (0.25 m^2) fixed in the large plots. Cutting (clipping) is commonly used to simulate goose grazing in agricultural lands (e.g. Conover, 1988; Fox *et al.*, 1998). The location of the small plots was moved between years.

Cutting was performed 8 times (from 17 June) in 2020 and 13 times (from 1 June) in 2021. Cumulated biomass in weekly cut plots was compared to total biomass of plots cut twice and variation between mixtures were analysed using program MINITAB 19, ANOVA, mixed effects model (Minitab, 2020).

Results and discussion

No statistically significant differences in biomass were identified between mixtures, either in plots cut according to normal practice, or weekly cut plots (Table 2). This was possibly due to high variation between repetitions and years. When present, *D. glomerata* rapidly became dominant and, if not present in the mixture, the plots were largely dominated by *F. pratensis* or *F. arundinacea* (Table 3). *D. glomerata* is a strong competitor (Carlen, 1994) and dominates as other species disappear. Despite its high abundancy in the commercial seed mixture (Table 1), *P. pratense* was only present in low quantities in the plots (Table 3).

Winter 2019-20 was characterized by a long-lasting snow cover, and *P. pratense* especially was damaged by snow mould in fields adjacent to the experiment. It is therefore possible that this also affected this field trial. As seen in other goose grazing experiments from the region, annual weather conditions affect sward development in spring and will give contrasting results over years (Bjerke *et al.*, 2021). In 2020, the small plots were cut eight times weekly. As shown in Table 2, mixtures C1 and C2 were lowest yielding in plots cut twice, but had among the highest percentage of remaining yields in cut plots. Since the cutting

Table 1. Mixtures included in the experiment. C1 and C2 are commercial, MIX 3-8 are experimental mixtures added to C1.[1]

Mixture	Species included in mixture
C1	*Phleum pratense* (50), *Festuca pratensis* (20), *Poa pratensis* (15), *Trifolium repens* (5), *Trifolium pratense* (10)
C2	*P. pratense* (80), *F. pratensis* (20)
MIX 3	C1 (50) + *Dactylis glomerata* (25) + × *Festulolium* (25)
MIX 4	C1 (50) + *D. glomerata* (25) + *Bromus inermis* (25)
MIX 5	C1 (50) + *D. glomerata* (25) + *Festuca arundinacea* (25)
MIX 6	C1 (50) + *D. glomerata* (25) + *T. pratense* (25)
MIX 7	C1 (50) + *F. arundinacea* (25) + *B. inermis* (25)
MIX 8	C1 (50) + *B. inermis*, *F. arundinacea*, × *Festulolium*, *D. glomerata* (12.5 each)

[1] Numbers in brackets denote weight portion (%) of total seed weight.

Table 2. Total kg dry matter (DM) ha^{-1} for both harvest years in twice cut, large plots (T.cut) and cumulated yield in weekly cut, small plots (W.cut).[1]

	2020 (8 cuts)			2021 (13 cuts)			Mean both year		
Mixture	T.cut	W.cut	% rem	T.cut	W.cut	% rem	T.cut	W.cut	% rem
C1	7,800	3,770	48.3	8,870	2,640	29.8	8,330	3,210	38.5
C2	7,380	3,750	50.8	8,200	2,350	28.7	7,780	3,050	39.2
MIX 3	8,540	3,520	41.2	8,060	3,060	38.0	8,290	3,290	39.7
MIX 4	7,840	3,920	50.0	7,910	2,490	31.5	7,870	3,200	40.7
MIX 5	8,460	3,650	43.1	8,750	2,660	30.4	8,600	3,150	36.6
MIX 6	8,350	3,380	40.5	8,630	3,140	36.4	8,480	3,260	38.4
MIX 7	8,460	3,700	43.7	8,730	2,530	29.0	8,590	3,110	36.2
MIX 8	8,400	3,720	44.3	8,800	2,820	32.0	8,600	3,270	38.0
P-value	0.12	0.77		0.45	0.42		0.17	0.98	

[1] % rem means remaining dry matter harvest in small plots compared to large plots.

Table 3. Mean botanical composition in big plots for the eight mixtures.[1]

Mixture	*Phleum pratense*	*Festuca pratensis*	*Dactylis glomerata*	*Festuca arundinacea*	*Trifolium pratense*	*Trifolium repens*	× *Festulolium*	*Bromus inermis*
C1	20	68			2	3		
C2	8	83						
MIX3	4	9	79		1		5	
MIX4	4	8	84					2
MIX5	4	5	81	7				
MIX6	5	8	59		25			
MIX7	18	25		47	2	1		4
MIX8	8	13	65	4	1		4	1

[1] Visually analysed at 2nd cut, 2nd harvest year in 2021.

treatment could start earlier in 2021, thirteen cuts were performed. In plots cut twice, higher yields were harvested in seven of eight mixtures, probably reflecting the earlier onset of spring this year. However, compared to 2020, the intense cutting regime affected the mixtures to a varying degree. Lowest remaining yields were seen in mixtures C1, C2 and MIX 7 (% rem, 2021). None of these mixtures contained *D. glomerata* (Table 1), which may indicate a lower tolerance against intense goose grazing.

Conclusions

Although not statistically significant, the results indicate that seed mixtures containing *Dactylis glomerata* better tolerate intensive grazing over prolonged periods of time, but results may vary between years.

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References

Bjerke J.W., Tombre I.M., Hanssen M. and Olsen A.K.B. (2021) Springtime grazing by Arctic-breeding geese reduces first- and second-harvest yields on sub-Arctic agricultural grasslands. *Science of the Total Environment* 793, 148619.

Carlen C. (1994) Root competition and shoot competition between *Festuca pratensis* Huds. and *Dactylis glometata* L. Doctoral Thesis. ETH Zürich, Switzerland, pp.112.

Conover, M.R. (1988) Effect of grazing by Canada geese on the winter growth of rye. *Journal of Wildlife Management* 52, 76-80.

Fox A.D. and Abraham K.D. (2017) Why geese benefit from the transition from natural vegetation to agriculture. *Ambio* 46, 188.197.

Fox A.D. Kristiansen J.N., Stroud D.A. and Boyd H (1998) The effects of simulated spring goose grazing on the growth rate and protein content of *Phleum pratense* leaves. *Oecologia* 116, 154-159.

Minitab Inc. (2020) MINITAB. Available at: http://www.minitab.com/en-US/products/minitab/.

Olesen J.E. and Bindi M. (2002) Consequences of climate change for European agricultural productivity, land use and policy. *European Journal of Agronomy* 16, 239-262.

Olsen A.K.B., Bjerke J.W. and Tombre I.M. (2017) Yield reductions in agricultural grasslands in Norway after springtime grazing by pink-footed geese. *Journal of Applied Ecology* 54, 1836-1746.

Tombre I.M., Høgda K.A., Madsen J. Griffin L.R., Kuijken E., Shimmings P., Rees E. and Verscheure C. (2008) The onset of spring and timing of migration in two arctic nesting goose populations: the pink-footed goose *Anser brachyrhynchus* and the barnacle goose *Branta leucopsis*. *Journal of Avian Biology* 39, 691-703.

Tombre I.M., Oudman T., Shimmings P., Griffin L. and Prop J. (2019) Northward range expansion in spring-staging barnacle geese is a response to climate change and population growth, mediated by individual experience. *Global Change Biology* 25, 3680-3693.

Uleberg E. and Dalmannsdottir S. (2018) Klimaendringenes påvirkning på landbruket i Norge innenfor ulike klimasoner. *NIBIO Rapport 4,* 75 pp 44.

Impact of trees on the growth of the herbaceous layer of Sahelian savannah. A UAV based approach

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Abstract

Sahelian savannah is composed of an annual herbaceous layer and a sparse tree community. The trees have a strong impact on the biomass and the species composition of the herbaceous layer due to microclimate and increase in fertility. In this work, we evaluated the impacts of distance of the tree on the herbaceous layers. We used an RGB UAV to produce a biomass map and then evaluated the distance of the tree impacts. In 2020 in a rangeland in northern Senegal, three grass measurements were made every second day during the growth season combined with a UAV flight made with a parrot Anafi drone. At each date we produced a biomass map and evaluated the distance of the tree impact using geostatic method. We obtained a calibration between UAV and field measurement with an R^2 equal to 0.64. The impacts of the tree ranged from 5 m at the beginning of the wet season to 15 m at the end of the wet season. This work shows the impact of tree distance on the grass layer in a savannah. The evaluation of this impact could be helpful for the management of the tree layers to increase the quantity of grass for the pastoralism.

Keywords: UAV, Savannah, tree impacts, biomass, grass

Introduction

In the Sahel region, the natural vegetation is the main source of food for livestock and plays an essential role for the local population (Ndiaye *et al.*, 2014). The ecosystem is a savannah composed of tree communities and annual herbaceous species. The trees improve the physical conditions of the environment and have a positive impact on the development and structure of the herbaceous layer (Akpo *et al.*, 1997; Elie *et al.*, 2003; Grouzis *et al.*, 1991). However, little is known about the distance of trees impact in these ecosystems. This study uses a drone and geostatistical approach to investigate the distance of influence of trees on the growth of the herbaceous layer in the Sahelian savannah.

Material and methods

Data were collected on a 1 ha plot during the 2020 rainy season (19 July 2020 to 17 September 2020). A drone flight and measurement of biomass in three plots each of 1 m^2, distributed respectively under the crown of a tree, at the edge of the crown, and at a distance from the edge of the crown equal to the height of the tree, were carried out every two days. These plots were rotated among the trees in the field until all four azimuths of trees were covered. The tree species in the field are *Balanites aegyptiaca* and *Vachellia tortilis*. They are between 2.3 and 8.8 m in height with an aerial cover of 6.4%. The drone flights were done at 60 m altitude, with a speed of 2 m s^{-1}, and 90% of overlap rate between images, on a double grid of 100×100 m and the angle of inclination of the camera fixed at 80°. The drone images were processed with PIX4DMapper software using the 3Dmaps analysis (Bossoukpe *et al.*, 2021a,b). Stepwise regression was used for the calibration between the 3D mapping data (Red (R), Green (G), Blue (B), DSM and vegetation indices; Table 1) and field measurements with the R software using Stepwise regression. Biomass maps were produced after calibration using QGIS.

Next, the variogram was calculated on the biomass maps to investigate the spatial variability of biomass around trees. The effect of tree distance (from the crown) refers to a parameter named 'Range' which was geostatistically analysed in R, using the libraries gstat and sp.

Table 1. Vegetation indices.

Acronym	Name	Formula	Source
VARI	visible atmospherically resistant index	(Green – Red) / (Green + Red - Blue)	Gitelson *et al.*, 2002
EXG	excess of green	Green – 0.39 × Red – 0.61 × Blue	Woebbecke *et al.*, 1995
GLI	green leaf index	(2 × Green – Red – Blue) / (2 × Green + Red + Blue)	Louhaichi, Borman and Johnson, 2001)
NDGRI	normalized difference green red index	(Red – Green) / (Red + Green)	Escadafal and Huete, 1991
NDBRI	normalized difference blue red index	(Red – Blue) / (Red + Blue)	Bossoukpe *et al.*, 2021a,b
NDBGI	normalized difference blue green index	(Green – Blue) / (Green + Blue)	Shimada *et al.*, 2012

$$\text{Range} = \sum_{i=0}^{nst} C_i . \Gamma_i \text{ (h)}$$

T_i(h) are the pool of i=0,nst structures, where the 0^{th} nested structure is the nugget effect by convention, is the contribution of the i^{th} structure, and each structural variogram (i=1,...,nst) is defined by seven parameters - three angles and three ranges (that define anisotropy) and a shape (often spherical, exponential or Gaussian).

Results and discussion

The results of the calibration show that the predictive variables of the biomass variation are the red, green, blue (statistically non-significant) spectra and the NDBRI index. Except the blue reflectance (P=0.07), results show the Red (P=3.1e^{-9}), Green (P=8.9e^{-7}) and NDBRI (P=0.02) contributed significantly to the predictive equation. The model obtained from the stepwise is significant with P=3.95e^{-13}, and R^2=0.64 according to the following predictive equation for biomass variation:

DM = -0.0054(Red) + 0.0047(Green) + 0.0032(Blue) + 0.78(NDBRI)-0.29.

The importance of vegetation indices to study biomass has been demonstrated (Lussem *et al.*, 2018). Establishing relationships between RGB reflectance, vegetation indices, and DSM data by using empirical linear methods to predict biomass variation reduces errors in establishing the predictive equation for RGB mosaic calibration.

This study incorporates a temporal dimension of tree impact. Performing variograms shows that the effect of the tree is not oriented and the distance of the impact of the trees on the grass varies during the season. The tree positively influences the variation of the herbaceous layer up to a minimum distance of 5 meters in August and a maximum of 15 m from the crown (Figure 1). These results are consistent with those of Roupsard *et al.* (2020) who used spectral indices (NDVI and MSAVI2) taken by drone and geostatistics to assess the distance of influence of *Faidherbia albida* on millet crop yield. Their results show that *Faidherbia albida* no longer has an effect on the millet crop beyond a distance of 17 m from the crown. The effect of the tree outside the crown may be explained by its ability to improve the soil quality by providing litter and root residues that help maintain soil organic matter levels and improve fertility (Young, 1995) at the plot or cropping system level.

Conclusions

The acquisition of multispectral images by drone associated with appropriate processing methods allows us to study the variation of the herbaceous layer and the interaction between woody and herbaceous species. This study shows the importance of the impact of the trees in Sahelian savannahs, and can help in the management of tree density in pastoral rangelands to positively affect the availability of fodder for livestock and improve the resilience of ecosystems to disturbances, particularly climatic disturbances.

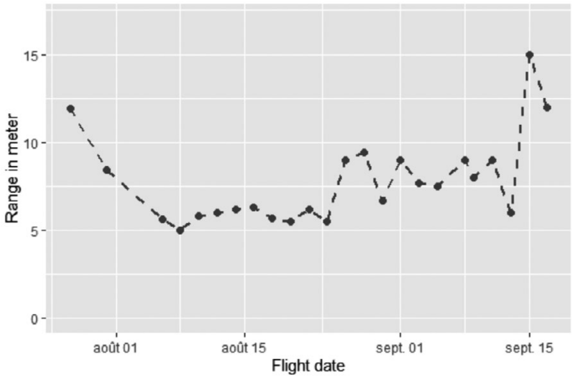

Figure 1. Variation in tree influence distance (Range) from the crown edge on the plot over the season.

However, these results could be improved by considering, in the study, the specificities related to each tree such as specific diversity, height and age.

References

Akpo L.E., Samb P.I. and Grouzis M. (1997) Effet du couvert des arbres sur la structure spécifique de la strate herbacée en savane subhumide soudanienne (Sénégal, Afrique de l'Ouest). *Conservatoire et Jardin Botaniques de Genève*, 52(2), 287-299.

Bossoukpe M., Faye E., Ndiaye O., Diatta S., Diatta O., Diouf A.A., Dendoncker M., Assouma H. and Taugourdeau S. (2021a) Low-cost drones help measure tree characteristics in the Sahelian savanna. *Journal of Arid Environments* 187(March 2020), 104449. https://doi.org/10.1016/j.jaridenv.2021.104449

Bossoukpe M., Ndiaye O., Diatta O., Diatta S., Audebert A., Couteron P., ... and Taugourdeau S. (2021b) Un drone pour l'évaluation de la phytomasse ligneuse et herbacée des savanes sahéliennes. *Revue D'élevage et de Médecine Vétérinaire des Pays Tropicaux* 74, 199-205.

Escadafal R. and Huete A.R. (1991) Étude des propriétés spectrales des sols arides appliquée à l'amélioration des indices de végétation obtenus par télédétection. *CR Acad. Sci. Paris* 312 (2), 1385-1391.

Elie A.L., Bada F. and Grouzis M. (2003) Diversité de la végétation herbacée sous arbre : variation selon l'espèce ligneuse en milieu sahélien. *Candollea*, 58(2), 515-531.

Gitelson, A.A., Kaufman, Y.J., Stark, R. and Rundquist D. (2002) Novel algorithms for remote estimation of vegetation fraction. *Remote Sensing of Environment* 80, 76-87.

Grouzis M., Nizinski J. and Akpo E. (1991) L'arbre et l'herbe au Sahel : influence de l'arbre sur la structure spécifique et la production de la strate herbacée, et sur la régénération des espèces ligneuses. *ORSTOM*.

Louhaichi M., Borman M.M. and Johnson D.E. (2001). Spatially located platform and aerial photography for documentation of grazing impacts on wheat. *Geocarto International* 16(1), 65-70.

Lussem U., Bolten A., Gnyp M.L., Jasper J. and Bareth G. (2018) Evaluation of RGB-based vegetation indices from UAV imagery to estimate forage yield in Grassland. *ISPRS Archives* 42(3), 1215-1219. https://doi.org/10.5194/isprs-archives-XLII-3-1215-2018

Ndiaye O., Diallo A., Wood S.A. and Guisse A. (2014) Structural diversity of woody species in the Senegalese semi-arid zone – Ferlo. *American Journal of Plant Sciences* 5(03), 416-426.

Roupsard O., Audebert A., Ndour A.P., Clermont-Dauphin C., Agbohessou Y., ... and Leroux L. (2020). How far does the tree affect the crop in agroforestry? New spatial analysis methods in a Faidherbia parkland. *Agriculture Ecosystems & Environment 296*(March), 106928. https://doi.org/10.1016/j.agee.2020.106928

Shimada S., Matsumoto J., Sekiyama A., Aosier B., Yokohana M. (2012) A new spectral index to detect Poaceae grass abundance in Mongolian grasslands. *Advances in Space Research* 50, 1266-1273.

Woebbecke D.M., Meyer G.E., Von Bargen K. and Mortensen D.A. (1995) Color indices for weed identification under various soil, residue, and lighting conditions. *Transactions of the ASAE* 38(1), 259-269.

Young A. (1995) *Agroforesterie pour la conservation du sol.* ICRAF. 183 p.

Can grassland vegetation be estimated from smartphone pictures collected by citizen scientists?

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Abstract

The methods used to assess diversity rely on time-consuming and laborious field samplings that limit their application at a regional scale. There is a need for methods that can be used to make rapid assessments of the composition of grassland plant communities. Data collected through citizen science have the advantage of covering large spatial and temporal extents. This study intends to evaluate the feasibility of assessing grassland diversity from pictures collected by a photographic approach using smartphones and measures of picture heterogeneity. We analysed two sets of pictures from Mediterranean and Temperate permanent grasslands. The heterogeneity of the pictures was measured by the Mean information Gain index (MIG) and anisotropy in the Hue and Value channels of JPEG pictures. Pearson correlations and linear mixed-effects models were used to assess their relationships with species composition as assessed by visual determination. The MIG of value channel was positively correlated with the percentage of grasses in Temperate (r=0.364) and Mediterranean grasslands (r=0.400). Linear mixed effect models showed that MIG calculated on the value channel is mainly affected by the percentage of grasses which modulates the MIG relationship with diversity due to the negative correlation between grass cover and plant diversity irrespective of the environment. The MIG index has potential to be applied in a citizen science approach, but the accuracy of the method was found to be comparatively low. Further research is therefore needed.

Keywords: mean information gain, anisotropy, functional groups, picture analysis

Introduction

The diversity and structure of grasslands are essential for ecosystem service delivery as it affects productivity and ecosystem stability. Therefore, its assessment and monitoring are of major interest for scientists and grassland managers. The current methods to assess diversity rely on time-consuming and laborious field samplings carried-out by experts. This is a major limitation to covering large extensions and performing assessments at a regional scale. Although remote sensing by satellites can be an asset to solve these issues, the coarse spatial resolution is nowadays a major constraint because of large within-field variation. The use of simple and rapid indices in combination with citizen science has the advantage of covering large spatial and temporal extents that cannot be afforded by other means. Proulx *et al.* (2008), based on a study in forests, proposed a measure of the texture of pictures known as Mean Information Gain (MIG) that can be related to the structural complexity of ecosystems. The MIG calculated from grey-tone pictures has been previously tested in grassland communities as a measure of diversity and heterogeneity, and showed promising results (Bonin *et al.*, 2014; Proulx *et al.*, 2014). However, further investigation is needed to confirm if this index can be used effectively to assess grassland in different conditions using simple smartphones as common tools in Citizen Science approaches. This study, therefore, aimed at assessing the potential of MIG calculated from the hue and value channels of pictures taken by regular smartphones to infer the structure and composition of grasslands. To this end, pictures were collected from two experiments in Temperate and Mediterranean grasslands in Europe.

Materials and methods

For Temperate grasslands, we collected pictures from the Forbioben experiment in Relliehausen, Lower Saxony (Germany) in the autumn of 2021. It consists of a one factorial randomized block design with three treatments (stocking intensities of grazing) and three replicates divided into nine paddocks (Jerrentrup *et al.*, 2014). In each paddock, ten 1 m^2 subplots (90 subplots in total) were evaluated for compressed sward height, Shannon diversity, and functional group composition using visual estimates of the percentage of cover. In these subplots, a picture was taken from a side view (45°) at 1 m distance to the centre of the plot. For the Mediterranean grasslands twenty-one 10×10 m plots were set at two different farms in the Cordoba province (9 and 12 per farm), Andalusia (Spain) in the spring of 2019. In each plot, four sampling plots of 0.4×0.4 m (84 subplots in total) were set and evaluated for sward height, Shannon diversity and functional group composition calculated by weighing the dry mass (24 h, 105 °C) of each species. In this case, the pictures were taken from top view at 1 m distance from the centre of the subplot.

The MIG measures the information gained by looking at the value of pixels in the neighbourhood of a particular pixel (Proulx *et al.*, 2008). If the distribution of pixels is uniform, MIG equals 0, while a random distribution of pixels would lead to values of MIG close to 1. Clustered patterns would yield intermediate values. The anisotropy is calculated as the ratio of $MIG_{horizontal}/MIG_{vertical}$. We used two regular smartphones to take pictures of 3000x4000 pixels. Before calculating the MIG and anisotropy, the RGB pictures were converted to the HSV channels (hue, saturation, and value). Since saturation and value are highly correlated, we used just value for the analyses. The hue channel defines the colour, while the value channel measures the quantity of light a pixel received. The relationships between picture heterogeneity (measured by MIG and anisotropy), functional group composition, structure and diversity of grasslands was firstly assessed by Pearson correlations. We used the percentage of grasses, sward height and Shannon index as proxies for functional group composition, structure, and diversity respectively. In addition, we evaluated if the picture heterogeneity inferred by MIG H and V can be explained by the percentage of grasses, sward height and plant diversity (Shannon index). We fitted linear mixed-effects models separately for MIG H and MIG V in each grassland ecosystem using height, percentage of grasses, and Shannon index as fixed continuous predictor effects. The categorical grazing intensity treatment as well as block were set as fixed and the sampling plot as a random effect for models of the Temperate grassland to account for the experimental design. For Mediterranean data, the 10×10 m plot label was set as a fixed effect to account for differences between sites and the sampling plot was used as a random effect.

Results and discussion

Anisotropy showed no correlation with the studied variables and was therefore not investigated further. The results of the linear-mixed-effects models showed that the percentage of grasses affected the MIG V in both Temperate (df=1; $F=5.6$; $P<0.05$) and Mediterranean grasslands (df=1; $F=5.9$; $P<0.05$) for which height was also significant (df=1; $F=5.9$; $P<0.05$). The MIG V showed a positive correlation with the percentage of grasses in both types of grasslands (Table 1). The percentage of grasses explained 12 and 15% (univariate R^2_{adj}; $P<0.001$) of the total variation of MIG V in Temperate and Mediterranean grasslands, respectively. This relationship is explained by the leaf structure of the grass swards. While smaller grass leaves cause a random pixel distribution, broad-leaved forbs create more clustered patterns and therefore lower MIG Vs which is in accordance with Bonin *et al.* (2014). The plant diversity measured by Shannon index showed a negative correlation with MIG V in Temperate and with MIG H in the Mediterranean grasslands (Table 1). Concerning the MIG H, it was significantly affected by height (df=1; $F=21.2$; $P<0.001$) in Temperate and by Shannon diversity (df=1; $F=5.5$; $P<0.05$) in Mediterranean grasslands.

Table 1. Pearson correlation (r) between MIG and composition and structure of grasslands.[1]

Dataset	MIG	% Grasses	Height	Shannon
Temperate	Value	0.364***	0.099	-0.268*
	Hue	0.085	0.474***	-0.021
Mediterranean	Value	0.400***	-0.107	-0.101
	Hue	0.248*	0.032	-0.408***

[1] $P<0.001$***; $P<0.01$**; $P<0.05$*.

The relationship between MIG and plant diversity might be modulated by the percentage of grasses due to a negative correlation between grass dominance and diversity (r=-0.655 in Temperate and r=-0.210 in Mediterranean grasslands). For Mediterranean grasslands, the negative relationship between diversity and MIG H (r=-0.408), confirmed by a significant effect of the Shannon index in the linear-mixed-effects model ($F=5.5$; $P<0.05$), could be promoted by the flowering stage of the Mediterranean grasslands. Proulx *et al.* (2014) also found decreasing MIG values of grey-tone pictures with increasing plant species richness. This method is not suitable to account for total floristic richness and patrimonial species since species with low cover are not represented by pixel patterns. Indices accounting for abundance correlate better with MIG than plant richness. Further research is needed to test this method with larger and more variable datasets to assess its feasibility and statistical power.

Conclusions

The relationship between grass cover and MIG V points to the potential feasibility as a rapid and simple index for the prediction of the functional group composition in pictures of swards consisting of grass and also likely one other functional group (dicots).

Acknowledgements

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References

Bonin, L., Proulx, R. and Rheault, G. (2014) A digital photography protocol for the rapid assessment of herbaceous communities in riparian buffers. *Riparian Ecology and Conservation* 2(1), 35-44.

Jerrentrup, J.S., Wrage-Mönnig, N., Röver, K.U. and Isselstein, J. (2014). Grazing intensity affects insect diversity via sward structure and heterogeneity in a long-term experiment. *Journal of Applied Ecology* 51(4), 968-977.

Proulx, R. and Parrott, L. (2008) Measures of structural complexity in digital images for monitoring the ecological signature of an old-growth forest ecosystem. *Ecological Indicators* 8(3), 270-284.

Proulx, R., Roca, I.T., Cuadra, F.S., Seiferling, I. and Wirth, C. (2014) A novel photographic approach for monitoring the structural heterogeneity and diversity of grassland ecosystems. *Journal of Plant Ecology* 7(6), 518-525.

Effects of soil type and competition on *Bituminaria bituminosa var. albomarginata* cv. LANZA® biomass production: preliminary results

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Abstract

The novel perennial legume *Bituminaria bituminosa* var. *albomarginata* is a promising forage for Mediterranean climates. It has shown outstanding drought resistance and good forage quality. Recently, an improved variety named LANZA® has been developed as a result of a collaborative breeding programme led by Australian and Spanish researchers. Further research is needed to study its adaptation to the varied environmental conditions of the Mediterranean regions. Previous research suggests that soil water saturation could limit the initial development of *B. bituminosa* var. *albomargina*. Slow initial development might compromise its successful establishment in natural grasslands, where grass species might outcompete *B. bituminosa*. This study aims to investigate the effect on the development of LANZA® in two contrasting soil textures (sandy and clay), the competition with *Lolium rigidum* and the interaction between both factors in a pot experiment with six replicates per treatment. Plants were grown in 6-litre pots for five months maintaining high soil water content. The results showed significant reduction in dry mass production of LANZA® due to *L. rigidium* competition and lower dry mass of thin rootsin clay soils.

Keywords: soil texture, *Lolium rigidum*, development, adaptation

Introduction

The use of perennial legumes in Mediterranean extensive livestock systems is recognized as one of the most promising measures to improve the sustainability of farms. Research efforts have been focused on the search for species that could meet the requirement of Mediterranean livestock systems (Rognli *et al.*, 2021). One of the most promising species is *Bituminaria bituminosa*, especially var. *albomarginata* from the Canary Islands. Previous research has demonstrated that this species is productive under very low rainfall (<200 mm) and it is suitable for feeding livestock (Mendez *et al.*, 1991; Oldham *et al.*, 2015). An improved variety named LANZA® has been developed as a result of a collaborative breeding programme lead by Australian and Spanish researchers (Real *et al.,* 2014).

There is still research to be done in order to clarify the potential adaptation of this species to the wide range of environmental conditions of Mediterranean environments. Previous research suggested that soil water saturation could be unfavourable for *B. bituminosa* var. *albomargina* (Fernández-Habas *et al.*, 2021). Also, the slow initial development of this plant might compromise successful establishment in temporary or permanent grasslands, where annual grass species might outcompete *B. bituminosa*. This study aimed to investigate the effect on cv. LANZA® in two contrasting categories of soil texture (sandy and clay), the competition with *Lolium rigidum* and the interaction between both factors.

Materials and methods

A complete block design with 6-litre pots was established in a shade house. The experiment consisted of six replicates per treatment and the treatments were: Soil type (sandy soil, denoted by S, or clay soil denoted by C) and Competition (pure culture of Lanza® and pure culture of *L. rigidum*, or Mixed culture of LANZA® and *L. rigidum*). In pure cultivation of LANZA®, one plant with three folioles was planted per pot. In pure cultivation of *L. rigidum*, two plants of *L. rigidum* with three leaves were planted per pot. In mixed cultivation one plant of LANZA® and two of *L. rigidum* were planted per pot. Sandy soil texture was: 73% sand, 13% silt and 14% clay. Clay soil texture was: 29% sand, 25% silt and 46% clay. Soil pH, cation exchange capacity and organic matter content was: 7.10, 11 (meq 100 g^{-1}) and 1.97% for sandy soil and 8.12 (1 2.5^{-1}), 31 (meq 100 g^{-1}) and 3.21% for clay soil. Both soils were mixed at 1/5 (v:v) with soil collected from natural populations of *B. bituminosa* to promote rhizobium inoculation (soil data correspond to the soil already mixed). Soil water content was maintained at a high level during the experiment (between 90-100% of soil water content at dripping point) and was controlled by weighing the pots and manually irrigating to the desired soil water content. The experiment was conducted from March to July 2021. At the end of the experiment, plants were cut to ground level. In pure cultivation, roots were also separated from soil and divided in thin (<2 mm) and thick roots (2> mm). Biomass was oven-dried at 60 °C for 72 h and then weighed. Firstly, the differences in dry mass production of pure culture (LANZA® or *L. rigidum* only) and mixed culture (summed production of LANZA® and *L. rigidum*) and by soil type were tested by two-way ANOVA. Then, we tested the shoot dry mass production of each species when cultivated without competition (Pure) and with competition (Mixed) by two-way ANOVAs independently for each species. Finally, thick and thin roots were compared between soil types in pure cultivation for each species by one-way ANOVA since separation by species in mixed cultivation was not possible. When differences were significant (P<0.05), post-hoc Tukey's test at the 0.05 level was carried out to separate homogeneous groups.

Results and discussion

Differences in dry mass production of pure and mixed cultures by soil type were not significant (Table 1). The total dry mass production of pure LANZA® culture was significantly higher than the total dry mass production of the mixture of LANZA® and *L. rigidum* and *L. rigidum* pure culture alone, whose dry mass production was significantly lower than the production of the mixture. These differences remain when shoot and root dry mass were compared separately (Table 1).

When the shoot dry mass production of each species in mixture culture was compared to their dry mass production in pure culture, LANZA® showed a significant reduction of 86% (Figure 1). Conversely, *L. rigidum* showed a significant increase of 43% of shoot dry mass production in mixed culture. A slower development of LANZA® in clay soils was observed during the initial stages, although the shoot dry mass at the end of the experiment showed no significant differences by soil type in either pure or mixed culture (df=1; F=3.97; P=0.06) (Figure 1).

Table 1. Pure and mixed dry mass of LANZA® and *Lolium rigidum* in sandy (S) and clay (C) soil types

Culture	Soil type	Total dry mass (g)	Shoot dry mass(g)	Root dry mass(g)
LANZA®	S	48.1±3.8 a	32.0±2.1 a	16.1±2.0 a
	C	38.1±5.1 a	26.3±3.3 a	11.8±1.8 a
LANZA® and *L. rigidum*[1]	S	26.1±5.0 b	17.0±2.6 b	9.1±2.6 b
	C	26.9±4.3 b	19.7±2.4 b	7.2±2.0 b
L. rigidum	S	11.4±0.8 c	9.4±0.5 c	2.0 ±0.3 c
	C	12.9±1.5 c	10.7±1.0 c	2.2±0.5 c

[1] Summed dry mass of LANZA® and *L. rigidum*.

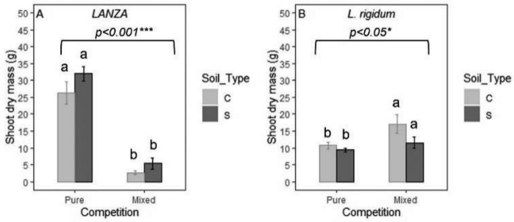

Figure 1. Effect of soil type and competition on shoot dry mass of LANZA® and *Lolium rigidum; P*-values indicate the main effect of competition. Level of significance: $P<0.001$***; $P<0.01$**; $P<0.05$*. *Pure* refers to cultivation without competition and *Mixed* refers to cultivation with competition.

LANZA® showed no significant difference between both soil types in thick roots dry mass. However, the dry mass of thin roots of LANZA® was 44% significantly lower when cultivated in clay soils while the soil type did not affect the dry mass of thin roots of *L. rigidum*. *L. rigidum* did not produce thick roots (2> mm).

These results suggest that LANZA® in pure cultivation could be more productive than when mixed with annual grasses. The establishment and production of LANZA® can be compromised when competing with annual grasses. This might be a major limitation for this species to be introduced into Mediterranean permanent grasslands where annual grasses are abundant (Olea and San Miguel, 2006). It also highlights the importance of avoiding competition in the initial phases of development to achieve a proper establishment in temporary grasslands.

Although shoot dry mass and total root dry mass production were unaffected by soil type, the lower dry mass of thin roots of LANZA® in clay soils indicates that sandy soils could be more suitable for its development. Given the possible effects of the culture in pots on the natural development of the plants, further field-scale trials are needed to verify these results.

Conclusions

Further research is needed to investigate measures aimed at reducing early competition of grasses and forbs to successfully establish LANZA®. The higher dry mass of thin roots of LANZA® in sandy soils shows a better adaptation of this species to poor sandy soils.

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References

Fernández-Habas, J., Hidalgo-Fernández, M. T., Leal-Murillo, ... and Fernández-Rebollo, P. (2021) Effects of two water regimes on morphological traits, nutritive value and physiology of three *Bituminaria bituminosa* varieties from the Canary Islands. *Journal of Agronomy and Crop Science*. https://doi.org/10.1111/jac.12485

Oldham, C.M., Wood, D., Milton, J., Real, D., Vercoe, P. and Van Burgel, A.J. (2015) An animal house study on utilisation of fresh tedera (*Bituminaria bituminosa* var. *albomarginata* and *crassiuscula*) by Merino wethers. *Animal Production Science* 55, 617-624.

Méndez, P., Fernández, M. and Santos, A. (1991) Variedades de '*Bituminaria bituminosa*' (L.) Stirton (Leguminosae) en el archipiélago canario. *Pastos: Revista de La Sociedad Española Para El Estudio de Los Pastos* 2021, 157-166.

Real D., Oldham C.M., Nelson M.N., Croser J., Castello M., Verbyla A., Pradhan A., Van Burgel A., Méndez P., Correal E., Teakle N.L., Revell C.K. and Ewing M.A. (2014) Evaluation and breeding of tedera for Mediterranean climates in southern Australia. *Crop and Pasture Science* 65,1114-1131.

Rognli, O.A., Pecetti, L., Kovi, M.R. and Annicchiarico, P. (2021) Grass and legume breeding matching the future needs of European grassland farming. *Grass and Forage Science* 76, 175-185.

Olea, L. and San Miguel-Ayanz, A. (2006) The Spanish dehesa. A traditional Mediterranean silvopastoral system linking production and nature conservation. *Grassland Science in Europe* 11, 3-13.

Flora biodiversity in silvopastoral systems under *Pinus radiata* D. Don in Galicia (NW Spain)

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Abstract

The main objective of the EU biodiversity strategy for 2030 is to protect nature and reverse the degradation of ecosystems. In this context, agroforestry plays an important role in conserving and even enhancing biodiversity from farms to landscape level. However, in silvopastoral systems, the flora biodiversity in the understorey depends, among other factors, on the tree canopy cover and the climatic conditions. This study aimed to evaluate the flora biodiversity in the understorey of silvopastoral and forest systems established under *Pinus radiata* D. Don with different tree canopy covers in the interior and coastal area of Galicia (NW Spain). In September 2020, a total of 48 plots were selected in the interior and coastal areas of Galicia. In each area, 12 plots with silvopasture and 12 forest plots were selected, both with different tree canopy covers (0, 50 and 100%), to evaluate the flora biodiversity in the understorey. The results show that silvopasture increases the flora biodiversity in the understorey.

Keywords: agroforestry, grazing, understorey, shade

Introduction

Biodiversity has become a key international environmental and production issue in the last decades (UN, 1992). Recently, action on biodiversity has been reinforced with initiatives such as the EU biodiversity strategy for 2030 (EU, 2020). This biodiversity strategy highlights that the uptake of agroforestry support measures under rural development should be increased as agroforestry practices have great potential to provide multiple benefits for biodiversity, people and climate (EU, 2020). Silvopastoral systems (SS), in which woody vegetation is combined with forage and animal production on the same land, can promote biodiversity through the creation of micro-sites within the plantation (shaded and unshaded areas) and the reduction of habitat fragmentation (Mosquera-Losada *et al.*, 2009). However, once SS are established, the flora biodiversity in the understorey can be modified by the tree growth and the climatic conditions of the area. Therefore, one of the best ways for conserving biodiversity in the SS is through the knowledge of the effect of the tree canopy cover on the flora biodiversity in the understorey. This study aimed to evaluate the flora biodiversity in the understorey of SS established under *Pinus radiata* D. Don with different tree canopy covers (0, 50 and 100%) in the interior and coastal areas of Galicia (NW Spain).

Materials and methods

The experiment was carried out in Galicia (NW Spain), within the Atlantic biogeographic region of Europe. Galicia is characterized by high rainfall levels, with well over 1000 mm a year across almost the entire region and a thermal oscillation between the interior and coastal area (range of oscillation: 1-4 °C).

In September 2020, a total of 48 plots with *Pinus radiata* D. Don (833 trees ha^{-1}) and different tree canopy covers (0, 50 and 100%) were selected in the interior and coastal areas of Galicia. In each area, 12 plots had a silvopasture land use and 12 plots had a forest land use (2 areas × 2 land uses × 3 tree canopy covers × 4 replicates). In each plot, the botanical composition of the understorey was estimated at three random points by visual identification of the percentage of species present on a known surface (4 m^2). Annual abundance diagrams were completed in which the percentage of senescent material was not taken into account (Magurran, 1988). Data were analysed using ANOVA with the statistical software package SAS (2001).

Results and discussion

In both areas of Galicia, the number of species was lower in the high tree canopy cover than in the other tree canopy covers (0%: 5.45^{a}, 50%: 5.45^{a}, 100%: 4.33^{b}; different superscript letters indicate significant differences between tree canopy covers) ($P<0.001$). The negative effect of the tree canopy cover development on the number of species could be explained by the interception of light and water by the canopy cover of the trees (Mosquera-Losada *et al.*, 2009). Therefore, in silvopastoral systems established under conifers, trees should be pruned, cleaned or thinned to maintain an adequate number of species in the understorey over time. Moreover, the number of species in the understorey was not significantly modified by the grazing and the climatic conditions of the areas included in this study ($P>0.05$). However, it seems that in both areas, the number of species tended to be higher in the silvopasture plots than in the forest plots. This trend could be due to the presence of animals, as they can enhance biodiversity through the creation of microsites caused by trampling, faeces deposition and plant species selection during grazing (Buttler *et al.*, 2009).

Finally, Figure 1 shows that in both studied areas, silvopasture implied a higher proportion of herbaceous species in the understorey, such as *Agrostis capillaris* L. and *Agrostis curtisii* Kerguélen, compared to the forest plots where scrub species comprised the main understorey vegetation, including *Ulex europaeus* L., *Erica mackaiana* Bab. or *Rubus* spp. This result is very relevant in this area because Galicia is one of the most fire-prone regions in Europe and reduction of the shrub proportion in the understorey might reduce the fire risk, as shrubs are more inflammable than herbaceous vegetation (Silva-Pando *et al.*, 2002). In this context, it is important to be aware that a lower fire risk implies a lower risk of biodiversity losses because plants in the burned areas become more prone to extinction. Moreover, forest fires can be considered as a significant source of carbon emissions to the atmosphere, contributing to global warming which could lead to biodiversity changes.

Conclusions

Agroforestry practices as silvopasture can provide solutions to meet the European biodiversity strategy for 2030, not only increasing biodiversity when an adequate tree canopy cover is maintained but also decreasing biodiversity losses due to the establishment of herbaceous species that decrease the forest fire risk compared to the shrubs.

Acknowledgements

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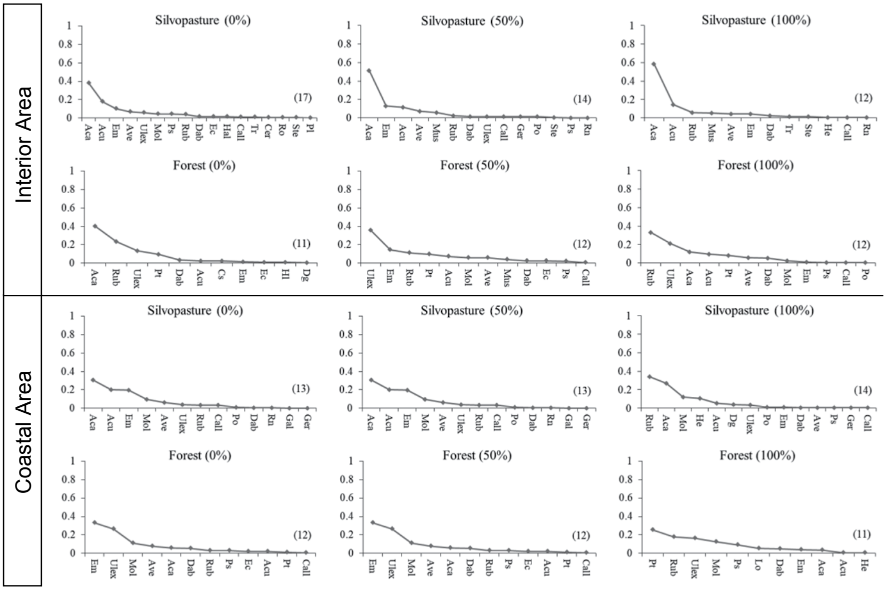

Figure 1. Species abundance diagrams for the silvopasture and forest plots established under different *Pinus radiata* D. Don canopy covers (0, 50 and 100%) in the interior and coastal area of Galicia (NW Spain). Aca: *Agrostis capillaris* L., Acu: *Agrostis curtisii* Kerguélen, Ave: *Avenula marginata* (Lowe) J. Holub, Call: *Calluna vulgaris* L., Cer: *Cerastium glomeratum* Thuill, Cs: *Cytisus scoparius* L., Dab: *Daboecia cantabrica* (Huds.) K. Koch, Dg: *Dactylis glomerata* L., Ec: *Erica cinerea* L., Em: *Erica mackaiana* Bab., Gal: *Galactites tomentosus* Moench, Ger: *Geranium dissectum* L., Hal: *Halimium lasianthum* subsp. *alyssoides* (Lam.) Greuter, He: *Hedera* sp, Hl: *Holcus lanatus* L., Lo: *Lonicera periclymenum* L., Mus: Moss, Mol: *Molinia caerulea* (L.) Moench, Pl: *Plantago lanceolata* L., Ps: *Pseudarrhenatherum longifolium* (Thore) Rouy, Pt: *Pteridium aquilinum* (L.) Kuhn, Po: *Potentilla* sp, Rn: *Ranunculus repens* L., Ro: *Rumex obtusifolius* L., Rub: *Rubus* spp., Ste: *Stellaria media* L. (Vill), Tr: *Trifolium repens* L., Ulex: *Ulex europaeus* L. Figures between brackets at the bottom of each chart indicate the number of species.

References

Buttler, A., Kohler, F. and Gillet, F. (2009) The Swiss mountain wooded pastures: patterns and processes. In: Rigueiro-Rodriguez, A., McAdam, J. and Mosquera-Losada, M.R. (eds.) *Agroforestry in Europe*, Springer, Dordrecht, the Netherlands, pp. 377-396.

EU (2020) *Communication from the Commission to the European Parliament, the Council, the European Economic and Social Committee and the Committee of the Regions. EU Biodiversity Strategy for 2030. Bringing nature back into our lives*. Available at: https://ec.europa.eu/info/sites/default/files/communication-annex-eu-biodiversity-strategy-2030_en.pdf

Magurran, A.E. (1988) *Ecological diversity and its Measurement*. Princeton University Press, London, UK.

Mosquera-Losada, M.R., Rodríguez-Barreira, S., López-Díaz, M.L., Fernández-Núñez, E. and Rigueiro-Rodríguez, A. (2009) Biodiversity and silvopastoral system use change in very acid soils. *Agriculture, Ecosystems and Environment* 131, 315-324.

SAS (2001) SAS/Stat User's Guide: Statistics. SAS Institute Inc., Cary, NC, USA.

Silva-Pando, F.J., González-Hernández, M.P. and Rozados-Lorenzo, M.J. (2002) Pasture production in a silvopastoral system in relation with microclimate variables in the Atlantic coast of Spain. *Agroforestry Systems* 56, 203-211.

UN (1992) *The Rio declaration on environment and development*. Available at: https://www.un.org/en/development/desa/population/migration/generalassembly/docs/globalcompact/A_CONF.151_26_Vol.I_Declaration.pdf

Long-term mineral fertilizer application strongly influences soil microbial community structure but not diversity

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Abstract

Mineral fertilizer application is a common management practice in grassland production. However, its potential long-term influence on soil microbial communities requires further study. In 1972, a split-plot experiment was established in the Jura region of Switzerland containing a non-fertilized control and a fertilized treatment of 150, 80 and 240 kg ha^{-1} yr^{-1} mineral N, P and K, respectively. Soil samples for microbial community analysis were taken in the summer of 2018. There was a highly significant effect of mineral fertilizer application on both soil fungal and bacterial community structure (all P<0.001), with unique fungal and bacterial indicator OTU (operational taxonomic unit) being associated to each treatment. Contrastingly, alpha diversity measures (OTU richness and inverse Simpson index) were not affected (P>0.05). These results demonstrated that long-term mineral fertilizer application had a strong influence on soil microbial community structure, promoting different microbial taxa, but did not affect the overall alpha diversity of these communities.

Keywords: permanent grassland, mineral fertilizer, soil fungi, soil bacteria

Introduction

Grassland management intensity strongly influences soil microbial communities, as has recently been demonstrated by comparing intensively and extensively managed grasslands in multiple European countries (Fox *et al.,* 2021). Grassland management, however, encompasses numerous individual practices administered throughout the growing season, such as mineral and/or organic fertilizer application, utilizations (cuttings and grazing events), reseeding and herbicide application. Thus, further study is required to elucidate the influence that such individual practices have on soil microbial communities. This is a critical knowledge gap, as the soil microbiome is fundamental in a number of agroecosystem processes, such as soil nutrient cycling, organic matter decomposition and plant productivity (Bertola *et al.*, 2021). In this study, we take one individual aspect of grassland management, mineral fertilizer application, and examine its long-term influence on: (1) soil fungal and bacterial community structure; and (2) soil fungal and bacterial alpha diversity measures.

Materials and methods

In 1972, a long-term experiment was established in the Jura region of Switzerland (930 m, 242' 090/617' 120). Included was a non-fertilized control (Con) and a fertilized treatment which received 150, 80 and 240 kg ha^{-1} yr^{-1} of mineral N, P and K, respectively (NPK, both n=6, 2-cut and 3-cut variants pooled together). Soil samples were taken in July 2018 (46 years after commencement), with eight cores being taken per plot, using a soil auger (ø 2.5 cm) to a depth of 20 cm. Each sample was homogenized and soil DNA extracted. The fungal internal transcribed spacer region (ITS2) and the bacterial 16S rRNA gene were PCR amplified as community markers and an amplicon-based Illumina Miseq sequence analysis conducted (Frey *et al.* 2016; Tedersoo *et al.* 2014). Operational taxonomic units (OTU) were clustered at 97% identity, and OTU richness and the inverse Simpson alpha diversity measures were calculated using the 'summary.single' command in MOTHUR (version 1.36.1). Taxonomic assignment was done using a database extracted from NCBI for fungal sequences and the SILVA database (version 132) for bacterial sequences. The effect of treatment on both fungal and bacterial community structure was tested with PERMANOVA using the 'adonis2' function (vegan package), and on the alpha diversity measures using analysis of variance. Associations of OTU to each treatment was determined by correlation-based indicator species analysis ('multiplatt' function), with an indicator OTU being defined as having an IndVal = 1.0 and a *P*-value ≤0.05. Analysis was done using R statistical software, version 4.0.2.

Results and discussion

There was a highly significant effect of mineral fertilizer application on both soil fungal and bacterial community structure (both $P<0.001$, Figure 1, panels A and B, respectively). The effect size was bigger for soil fungi than for soil bacteria, as was seen in the R^2 of the model (0.449 compared to 0.301, respectively) and the community centroid distance (0.570 compared to 0.255, respectively). Such a pronounced effect is in line with recently reported differences in soil fungal and bacterial community structures between extensively and intensively managed grasslands in European grasslands (Fox *et al.,* 2021).

Additionally, different indicator microbial taxa were associated to both Con and NPK, supporting the notion that contrasting nutrient inputs in grasslands select for specific microbial groups (Leff *et al.* 2015). A higher number of indicator fungal OTU were associated to Con (51) than to NPK (27), while the same number of indicator bacterial OTU were associated to both treatments (7). Among the indicator fungal OTU associated to Con, were those assigned to the genera *Clavaria* and *Leohumicola,* which have previously been shown to be indicator fungi for extensively managed grasslands in Europe (Fox *et al.,* 2021, McHugh *et al.,* 2001).

In contrast to what was observed for community structure, there was no significant difference ($P>0.05$) between Con and NPK in terms of fungal OTU richness (avg. 654 and 653 OTU, respectively), bacterial OTU richness (avg. 1,828 and 1,973 OTU, respectively), fungal inverse Simpson index or bacterial inverse Simpson index (Figure 2). The lack of a treatment effect on fungal alpha diversity is somewhat surprising, as fungal OTU richness has been reported as being lower in intensively managed grasslands, compared to extensively managed grasslands, in many European regions (Fox *et al.,* 2021). Therefore, this reported decline in fungal OTU richness may not solely be due to mineral fertilizer application, but rather other aspects of grassland management and/or their interactive effects. These results indicate that while mineral fertilization significantly affects the abundance of individual microbial taxa, it does not influence microbial community diversity.

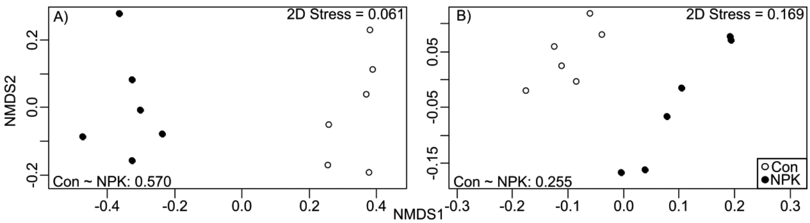

Figure 1. A non-metric multidimensional scaling plot (NMDS) displaying the difference in both fungal (panel A) and bacterial (panel B) community structure between the non-fertilized control (Con, open symbols) and the fertilized NPK treatment (closed symbols). Also displayed in each panel is the community centroid distance between treatments (Euclidian distance).

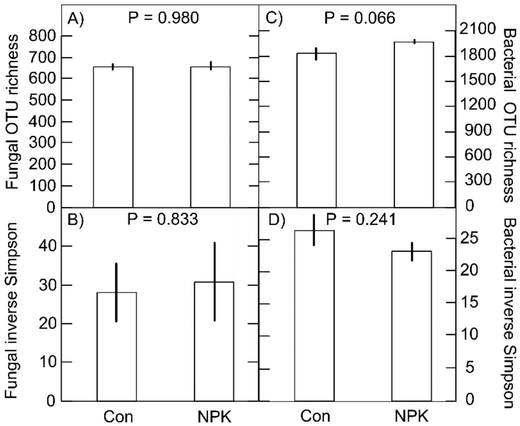

Figure 2. Effect of mineral fertilizer application (± 1 standard error) on (A) fungal OTU richness, (B) fungal inverse Simpson index, (C) bacterial OTU richness and (D) bacterial inverse Simpson index.

Conclusions

Our study demonstrates the strong, highly significant effect that mineral fertilizer application has on soil microbial community structure, suggesting that it is a major driver behind the recently reported differences in soil microbial community structure between grasslands of highly contrasting management types in Europe. The lack of effect mainly on fungal alpha diversity measures, however, would indicate that mineral fertilizer application alone may not explain the lower levels of fungal diversity previously reported in intensively managed European grasslands.

Acknowledgements

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References

Bertola M., Ferrarini A. and Visioli G. (2021) Improvement of soil microbial diversity through sustainable agricultural practices and its evaluation by -omics approaches. *Microorganisms* 9, 1400.

Fox A., Widmer F., Barreiro A., Jongen M., Musyoki M., Vieira Â. ... and Lüscher A. (2021) Small-scale agricultural grassland management can affect soil fungal community structure as much as continental scale geographic patterns. *FEMS Microbiology Ecology* 97, fiab148.

Frey B., Rime T., Phillips M., Stierli B., Hajdas I., Widmer F. and Hartmann M. (2016) Microbial diversity in European alpine permafrost and active layers. *FEMS Microbiology Ecology* 92(3), fiw018.

Leff J.W., Jones S.E., Prober S.M., Barberán A., Borer E.T., Firn J.L. ... and Fierer N. (2015) Consistent responses of soil microbial communities to elevated nutrient inputs in grasslands across the globe. *Proceedings of the National Academy of Sciences of the USA* 112, 10967-10972.

McHugh R., Mitchel D., Wright M. and Anderson R. (2001) The fungi of Irish grasslands and their value for nature conservation. *Biology and Environment Proceedings of the Royal Irish Academy* 101B, 225-242.

Tedersoo L., Bahram M., Põlme S., Kõljalg U., Yorou N.S., Wijesundera R., ... and Abarenkov K. (2014) Fungal biogeography. Global diversity and geography of soil fungi. *Science* 346(6213), 1256688.

Differences in soil fungal community structure driven by grassland management not sampling period

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Abstract

This study investigated if differences in soil fungal community structure occur between intensively and extensively managed permanent grasslands, and whether they persist across management events and changes in weather conditions within the growing season. Soil samples were taken from both grassland management intensities six times through 2017 (May – October) in the Zurich region, Switzerland. For fungal community structure (measured using Illumina Miseq next generation sequencing), the influence of management ($\sqrt{CV}=0.227$, $P\leq0.001$) was much greater than that of sampling event ($\sqrt{CV}=0.079$, $P\leq0.001$). Importantly, no interaction occurred between management and sampling event ($P>0.05$), and consequently, pairwise significant differences in fungal community structure between intensive and extensive grasslands persisted across each of the sampling events (all at least $P\leq0.05$). These results highlight the temporal stability of pairwise differences in soil fungal structures between contrasting grassland management intensities through the whole growing season, despite changing meteorological conditions and multiple management events.

Keywords: permanent grassland, management intensity, soil fungi, sampling events

Introduction

The soil microbiome is fundamental in a number of agroecosystem processes such as soil nutrient cycling, organic matter decomposition and plant productivity (Bertola *et al.*, 2021). Grassland management intensity is a major driver of grassland soil fungal community structure, with this influence having been observed both at the continental scale and in multiple European regions, as was recently reported in the BIOINVENT project (Barreiro *et al.,* 2022; Fox *et al.,* 2021). Soil microorganisms can, however, also exhibit temporal variation, though the extent to which this shapes the structure of the soil microbial community in agricultural systems remains poorly understood (Dunn *et al.*, 2021). It is an important knowledge gap, as the pertinence of incorporating temporal aspects into soil microbiome studies is being increasingly highlighted in the literature (Geisen, 2021). This is particularly the case with managed grasslands, as not only do climatic conditions and plant phenological stage change as the growing season progresses, but they also undergo numerous management events over this time period (i.e. fertilization, cuttings). The principal aims of this study were to test: (1) if important differences in soil fungal community structure occur between intensively and extensively managed grasslands; and (2) whether these persist throughout the entire growing season.

Materials and methods

Two sharply contrasting grassland management systems were sampled (five fields of each) around the Zurich region of Switzerland. These were: (1) a highly intensively managed permanent grassland (INT, high fertilization with early, frequent utilizations); and (2) an extensively managed permanent grassland (EXT, no fertilization with infrequent utilizations). Soil samples were taken six times throughout the 2017 growing season (May – October), along which management events occurred (i.e. cuttings, fertilizer applications), as well as changing weather conditions. At each sampling, sixteen soil cores were taken along a transect in the field centre, using a soil auger (ø 2.5 cm) to a depth of 20 cm. Soil DNA was extracted from each sample, with the fungal internal transcribed spacer region (ITS2) being PCR amplified and an amplicon-based Illumina Miseq sequence analysis conducted to generate operational taxonomic units (OTU, 97% sequence identity). A pairwise Bray-Curtis dissimilarity matrix was then constructed, which served as the response in a repeated measures PERMANOVA to determine the influence of grassland management intensity (factor 'Manage'), sampling event (factor 'Time') and the Manage×Time interaction on fungal community structure (PRIMER-7 statistical software package, version 7, Plymouth UK). The variance explained by each factor in the model was reported as the square-root of the component of variation (√CV). A constrained ordination, maximizing the differences within 'Manage' and 'Time' was conducted in R, using the vegan package for figure construction (R core team, 2021).

Results and discussion

The factor 'Manage' had a strong determinant effect on fungal community structure, as indicated by the square-root of the component of variation of the model (√CV=0.227, $P \leq 0.001$, Table 1) and evident from Figure 1A. The management effect was far stronger than the 'Time' effect (√CV=0.079, $P \leq 0.001$). A stronger effect of land-use on soil microbial community structure, compared to sampling event, has been demonstrated previously (e.g. Gschwend *et al.*, 2021), but such studies have used very different land-use types (i.e. grasslands vs forest). Here, we even demonstrate this effect within the same land-use type of permanent grassland, just with contrasting management regimes.

Importantly, there was no significant 'Manage'×'Time' interaction ($P > 0.05$, Table 1). Consequently, the clear, highly significant differences in fungal community structure between INT and EXT persisted throughout the six sampling events over/across the growing season (all at least $P \leq 0.05$, analysis not shown). The lesser influence of sampling event is also apparent from Figure 1B. Here, despite using a constrained ordination maximizing the differences between the six sampling events, there is still a clear separation between INT and EXT. Furthermore, little variation in fungal community structure across the six sampling events is evident in each sampling replicate, with the points of the six sampling times

Table 1. Results of a repeated measure PERMANOVA model of the effect of the two grassland management intensities (Manage), sampling event (Time), and the Manage×Time interaction on fungal community structure.[1]

Factor	df	MS	Pseudo F	√CV
Manage	1	2.149	3.385***	0.227
Rep(Manage)	8	0.637		0.308
Time	5	0.139	1.783***	0.079
Manage×Time	5	0.083	1.057^{ns}	0.030
Residual	39	0.078		0.280

[1] Rep(Manage) denotes the term for the replicate sites over which Manage is tested. Shown are the degrees of freedom (df), the mean sum of squares (MS), the Pseudo-F value and the square-root of the component of variation (√CV). Significance: '***' $P \leq 0.001$, 'ns' $P > 0.05$.

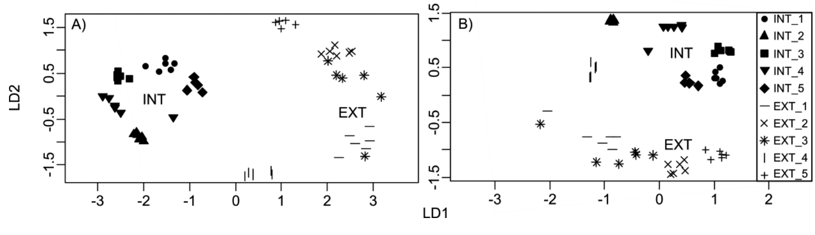

Figure 1. Constrained ordination (based on canonical analysis of principal coordinates) of fungal community structure (A) maximizing the differences between the five intensively (INT, filled symbols) and extensively (EXT, line symbols) managed grassland sites across the six sampling events and (B) maximizing the differences between each of the six sampling events across the two management intensities.

clustering tightly together (Figure 1B). Recently, the BIOINVENT project demonstrated the strong influence of grassland management (i.e. intensive vs extensive) on soil fungal community structure in multiple European regions (Fox *et al.,* 2021), though this study only utilized a single sampling event at the peak plant productivity of the growing season. The results presented here both confirm and strengthen this finding, and demonstrate that such management-induced differences are not merely present at peak plant productivity, but persist regardless of plant phenological stage or individual management events.

Conclusions

Our study highlights that sharp differences in fungal community structure are induced by grassland management. These differences are temporally stable throughout the entire growing season. These results strengthen the findings reported in the BIOINVENT project, which also demonstrated the strong influence of grassland management on soil fungal community structure in multiple European regions, though it utilized only one sampling event.

Acknowledgements

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References

Barreiro A., Fox A., Jongen M., Melo J., Musyoki M., Vieira Â. ... and Dimitrova Mårtensson L-M. (2022) Soil bacteria respond to regional edapho-climatic conditions while soil fungi respond to management intensity in grasslands along a European transect. *Applied Soil Ecology* 170, 104264

Bertola M., Ferrarini A. and Visioli G. (2021) Improvement of soil microbial diversity through sustainable agricultural practices and its evaluation by -omics approaches: a perspective for the environment, food quality and human safety. *Microorganisms* 9, 1400.

Dunn L., Lang C., Marilleau N., Terrat S., Biju-Duval L., Lelièvre M. ... and Prevost-Boure NC. (2021) Soil microbial communities in the face of changing farming practices: A case study in an agricultural landscape in France. *PLoS ONE* 16, e0252216.

Fox A., Widmer F., Barreiro A., Jongen M., Musyoki M., Vieira Â. ... and Lüscher A. (2021) Small scale agricultural grassland management can affect soil fungal community structure as much as continental scale geographic patterns. *FEMS Microbiology Ecology*, 97, fiab148.

Geisen S. (2021) The future of (soil) microbiome studies: current limitations, integration, and perspectives. *mSystems* 6(4), e00613-21.

Gschwend F., Hartmann M., Hug A-S., Enkerli J., Gubler A., Frey B. ... and Widmer F. (2021) Long- term stability of soil bacterial and fungal community structures revealed in their abundant and rare fractions. *Molecular Ecology* 30, 4305-4320.

Performance of *Trifolium repens* and *T. pratense* under marginal growing conditions

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Abstract

The current UK clover varietal testing programme only conducts trials under very favourable growing conditions. This study tested commercial and experimental varieties when grown under typical marginal conditions in an upland area on nutrient poor soils. A total of 10 different varieties of *Trifolium repens* (AberLasting, AberDai, AberAce, Aran, Alice, AberSwan plus 4 experimental lines/varieties) and 8 different varieties of *Trifolium pratense* (AberClaret, AberChianti, Merviot plus 5 different experimental lines/varieties) were planted. Four replicate 6×1.2 m plots of each variety were prepared in a randomized block design. Sowing rates and establishment were in accordance with the United Kingdom National List Trials, and the plots were subsequently managed using a similar cutting regime. Samples were taken towards the end of the first three growing seasons and botanical separations carried out to determine percentage biomass contribution of the sown varieties on a dry matter basis. All species showed a substantial decline from the first to third year, from an average of 55.7 to 7.9% for *T. repens* varieties, and 57.4 to 6.8% for *T. pratense* varieties. The results highlight the need to develop specific varieties adapted to low fertility conditions.

Keywords: clover, varieties, persistency, less favoured areas

Introduction

Plant breeding has made major advances in the decades since large tracts of the UK uplands were last improved. The introduction of new forage legumes would not only radically improve livestock production yields and efficiencies in marginal areas (Fraser *et al.*, 2004); it could also enhance ecosystem service provision beyond primary production. However, legume (and grass) varietal testing in the UK is carried out only on sheltered, fertile sites. This study explored the relative performance of different varieties of *Trifolium repens* (white clover) and *Trifolium pratense* (red clover) when grown under typical marginal conditions in an upland area on nutrient poor soils. We tested recommended commercial varieties of each species, plus new lines/varieties currently under development.

Materials and methods

A total of 10 different varieties of white clover (AberLasting, AberDai*, AberAce*, Aran*, Alice*, AberSwan* plus 4 experimental lines/varieties) and 8 different varieties of red clover (AberClaret*, AberChianti*, Merviot* and 5 different experimental lines/varieties) were tested. Varieties marked with an asterisk are on the current (2020/2021) Recommended Grass and Clover Lists for England and Wales (AHDB, 2021). Four replicate 6×1.2 m plots of each variety were prepared in a randomized block design on brown podzolic soil. The exposed site was located 250 m above sea level. The area receives a mean annual rainfall of approximately 1,850 mm and has average minimum and maximum air temperatures of 5.2 and 11.9 °C respectively. Sowing rates and management were in accordance with the United Kingdom National List Trials: Trial Procedures for Official Examination of Value for Cultivation and Use (APHA, 2018a,b). White clover varieties were sown with a companion ryegrass (*Lolium perenne*, cv AberMagic) (APHA, 2019a) while red clover varieties were sown as a monoculture (APHA, 2019b). Samples of herbage within two 1×0.14 m strips per plot were cut at the end of each of the first three growing seasons, and botanical separations carried out to determine percentage biomass contribution of the sown varieties on a dry matter basis. Following an angular transformation of the data statistical analysis was carried out via a two-way ANOVA with a treatment structure of Variety × Year.

Results and discussion

Year had a highly significant effect on the proportion of white clover within the sward by the end of the growing season ($P<0.001$), with the untransformed means 55.7, 14.2 and 7.9% for Year 1, Year 2 and Year 3 respectively (Figure 1). Between-variety differences were noted but were less pronounced ($P<0.05$). There were no Year × Variety interaction effects.

A similar pattern was recorded for the red clover varieties (Figure 2). Year again had a pronounced effect on percentage contribution to the overall sward biomass ($P<0.001$), with the percentage recorded in Year 1 markedly higher than those recorded in subsequent years. Untransformed means were 57.4, 8.3 and 6.8% for Year 1, Year 2 and Year 3 respectively. Variety also had a highly significant effect on percentage inclusion within the sward, with AberClaret and the older variety Merviot having higher proportions overall than experimental lines 2 and 4. There were no Year × Variety interaction effects.

The results highlight the challenges faced by forage legumes when sown in marginal conditions. Although many of the varieties tested are on the current Recommended Lists for white and red clover for use in Wales (AHDB, 2021), persistency was poor. Around 80% of Wales carried the EU designation of Less Favoured Area due to challenging growing conditions typified by the experimental site. During the current study the plots were cut to simulate grazing. Were such pastures to carry livestock the situation would likely be exacerbated by selective grazing.

Conclusions

The results indicate that the performance of white clover and red clover varieties as reported in the Recommended Lists for England and Wales (AHDB, 2021) cannot be assumed to be representative of what would be achieved in more marginal growing conditions. To capitalize on the considerable potential for legumes to deliver productivity and environmental benefits for upland areas varieties also need to be developed for, and tested in, low fertility conditions.

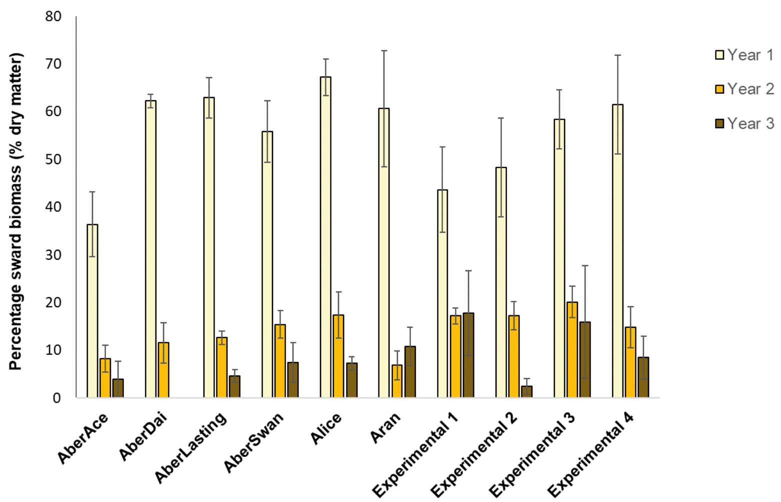

Figure 1. Proportion of biomass within plots accounted for by the white clover variety originally sown.

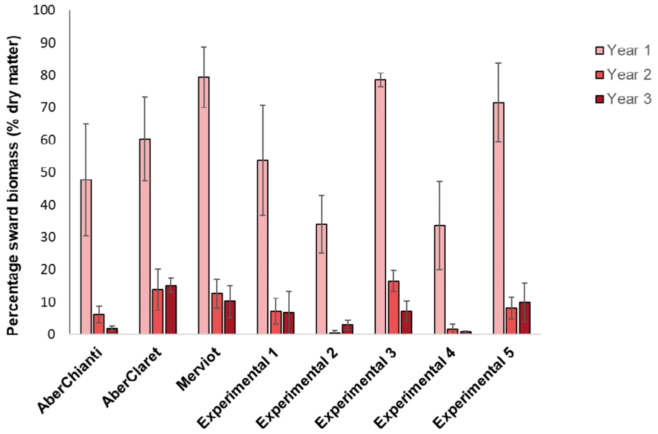

Figure 2. Proportion of biomass within plots accounted for by the red clover variety originally sown.

Acknowledgements

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References

AHDB (2021) *Recommended grass and clover lists for England and Wales*. Agriculture and Horticulture Development Board, Stoneleigh, UK.

APHA (2018) United Kingdom National List Trials: Trial procedures for official examination of value for cultivation and use (VCU) Harvest 2018 – White clover; Animal & Plant Health Agency, Surrey, UK.

APHA (2018) United Kingdom National List Trials: Trial procedures for official examination of value for cultivation and use (VCU) Harvest 2018 – Red clover and lucerne; Animal & Plant Health Agency, Surrey, UK.

Fraser M.D., Speijers M.H.M., Theobald V.J., Fychan R. and Jones R. (2004) Production performance and meat quality of grazing lambs finished on red clover, lucerne or perennial ryegrass swards. *Grass and Forage Science* 59, 345-356.

Effects of establishment method on forage yield and composition

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Abstract

An experiment tested the hypothesis that direct drilling (DD) with a diverse grass mixture will increase forage yield compared to a perennial ryegrass (*Lolium perenne*) (PRG)/red clover (*Trifolium pratense*) (RC) mix (PR) and a similar yield to the same forage mixture established by ploughing, when established with the use of herbicide. Two forage mixtures: (1) PR; and (2) PRG, RC cocksfoot (*Dactylis glomerata*) and timothy (*Phleum pratense*) (PR+TC) were evaluated. Replicate plots of each forage treatment were sown either by: (1) DD without herbicide; (2) DD after herbicide; or (3) after ploughing and cultivating after herbicide. Effects of establishment method (EM) and sown species (S) were evaluated by harvesting silage cuts over 3 years. A diverse grass mixture increased sown species DM yield but not total forage yield compared to the PR mixture. DD plots established without herbicide had a lower DM yield than DD with herbicide and a lower sown species yield compared to other treatments. There was no effect of DD or ploughing on total DM yield or sown species yield when plots were established with herbicide.

Keywords: tillage, mixed swards, cultivation, direct drill, plough, no-till

Introduction

Grassland renovations are needed to maintain forage productivity and quality but establishment can be costly in both monetary and environmental terms when ploughing and related operations are adopted. The establishment of grass reseeds by direct drilling or surface sowing offers an alternative, with reduced costs involved as well as environmental and soil health benefits (Crotty *et al.*, 2016). However, direct drill (DD) establishment success rates can be variable and are highly dependent on soil conditions during establishment. Timothy is more cold tolerant (Gudleifsson *et al.*, 1986; Turner *et al.*, 2012) and cocksfoot is more drought tolerant (Turner *et al.*, 2012) compared to ryegrass. A sward containing these diverse grasses may establish more quickly to mitigate these establishment method (EM) risks when new swards are established by direct drill. An experiment was conducted to compare two forage treatments and three establishment methods. The aim of the experiment was to test the hypothesis that DD with a diverse grass mixture will increase forage dry matter (DM) yield compared to perennial ryegrass-red clover (*Lolium perenne* – *Trifolium pratense*) mix and a similar DM yield to a diverse grass mixture established by ploughing, when established with the use of herbicide to test the hypothesis that increasing sward diversity will improve establishment success, as indicated by a higher sown species DM yield.

Materials and methods

Two sward types were evaluated (1) PR, a sward comprising late heading tetraploid ryegrass (*Lolium perenne* cv AberGain (PRG)) and red clover (*Trifolium pratense* cv AberClaret (RC)) sown at the rate of 26 and 7.5 kg ha^{-1} respectively and (2) PR+TC, a diverse sward comprising PRG, RC, cocksfoot (*Dactylis glomerata* cv Donata) and timothy (*Phleum pratense* cv Presto) sown at the rate of 11, 7.5, 7.5 and 7.5 kg ha^{-1} respectively. Each sward type was sown either (1) by direct drilling without glyphosate application (DD); (2) by direct drilling post-glyphosate application (Gly+DD); or (3) after ploughing and cultivating post-glyphosate application (Gly+Plough). Four replicate 5×2 m plots of each treatment combination were established in a randomized block design at Gogerddan, Aberystwyth University, Wales (52.430, -4.022) on an area of permanent pasture situated on a silty loam soil of the Rheidol series with a pH of 6.2, P and K of 20 and 166 mg l^{-1}. Areas allocated to Gly+DD and Gly+Plough were treated with glyphosate (360 g l^{-1}) herbicide (Gallup XL, Barclay Ltd, Mulhuddart, Dublin, Ireland) at the rate of 6 l ha^{-1} on 20 May 2019. Areas allocated to Gly+Plough were ploughed to a depth of 150 mm and power-harrowed on 23 May. All plots were sown on 24 May 2019 using an Aitchison Grassfarmer direct drill (Reese Group Ltd, Palmerston North, New Zealand), but with the drill raised when sowing the Gly+Plough treatment. Phosphate and potash were applied to maintain indices throughout the study but no N was applied. Plant density was enumerated in 8 quadrats (0.25×0.36 m) per plot 8 weeks after sowing. Plots were harvested on 3 occasions in the establishment year and 4 occasions, at approximately 6-week intervals, in each of the two subsequent years to simulate a silage-cutting regime. Total forage dry matter (DM) yield was determined by mechanically harvesting a strip, 3.5×1.5 m, within each plot using a Haldrup 1500 plot harvester (J. Haldrup a/s, Løgstør, Denmark) with a cutting height of 6 cm. Botanical composition was determined by manual separation of a 100 g subsample of fresh forage into sown and unsown species each oven dried at 100 °C for 48 h. Treatment effects were examined by analysis of variance of the 2×3 factorial design.

Results and discussion

Density of each of the sown grass species 8 weeks post-sowing was higher after ploughing than direct drilling ($P<0.05$) (Table 1), but red clover density was unaffected by either treatment ($P=0.408$ and 0.811 for EM and S respectively, overall mean 159 plants m^{-2} (data not shown)). Unsown grass coverage 8 weeks post sowing differed between establishment methods ($P<0.001$) with 66% coverage in DD plots reduced ($P<0.05$) to 7% with herbicide. Using the more diverse seed mixture increased sown species yield over the three years, relative to the PR mixture ($P=0.034$) but the effect on total yield was not significant ($P=0.111$). Yield of sown species was also affected by establishment method ($P<0.001$) with higher offtakes from plots treated with herbicide than from untreated plots. Total yields were higher from plots established by Gly+DD than from DD plots with no herbicide ($P<0.05$); yields following Gly+Plough were intermediate. Red clover was the dominant forage in all treatments, contributing over 70% of DM yield over the three years but with an interaction between treatments ($P=0.006$). There was no effect ($P>0.05$) of species sown on the percentage of red clover when plots were established by DD. For PR the proportion of red clover was not affected by EM, but with PR+TC the proportion of red clover decreased when herbicide was used ($P<0.05$). Ryegrass contributed less than 2% of yield on DD plots irrespective of sowing mixture but this contribution increased ($P<0.05$) when glyphosate was used and more so when PR rather than PR+TC was sown. Cocksfoot and timothy contributions to yield were also lower in plots established without herbicide ($P<0.05$), likely due to competition from unsown species in the existing sward. The yield from timothy was particularly poor, even with herbicide application, possibly due to its inability to compete under the prevailing environmental conditions during this experiment.

Conclusions

A diverse grass mixture increased the yield of sown species, but not total forage yield, compared to the PR treatment, when herbicide treatment was used prior to establishment. DD without prior treatment with herbicide was the least successful option for establishment and forage production.

Acknowledgements

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Table 1. Species establishment and forage yield and sown species contribution over 3 years.[1]

	Species sown (S)	Establishment method (EM)			Mean	Effect	SEM	Prob
		DD	Gly+DD	Gly+Plough				
Plant density 8 weeks post sowing (n m^{-2} unless stated otherwise)								
PRG	PR	89^{a}	146^{a}	$^{A}262^{b}$	166	EM	11.5	<0.001
	PR+TC	71	81	$^{B}122$	91	S	9.4	<0.001
	Mean	80	113	192		EM × S	16.3	0.007
Cocksfoot	PR+TC	$5.7^{a'}$	$56.7^{b'}$	$91.2^{c'}$	-	EM	$0.39^{\$}$	<0.001
Timothy	PR+TC	$8.3^{a'}$	$21.5^{a'}$	$73.6^{b'}$	-	EM	$0.74^{\$}$	0.004
Unsown grass (% cover)	PR	65.4	7.2	6.3	22.1	EM	$1.08^{¥}$	<0.001
	PR+TC	66.2	6.6	7.5	22.1	S	$0.88^{¥}$	0.760
	Mean	$65.8^{b'}$	$6.9^{a'}$	$6.9^{a'}$		EM × S	$1.53^{¥}$	0.806
Forage yield								
Total (Mg DM ha^{-1})	PR	36.52	37.86	36.59	36.95	EM	0.401	0.018
	PR+TC	36.71	38.91	37.57	37.74	S	0.327	0.111
	Mean	$36.62^{a'}$	$38.38^{b'}$	$37.03^{ab'}$		EM × S	0.567	0.674
Sown species (Mg DM ha^{-1})	PR	32.20	37.16	35.72	35.03	EM	0.471	<0.001
	PR+TC	33.77	38.27	36.86	36.30	S	0.384	0.034
	Mean	$32.99^{a'}$	$37.72^{b'}$	$36.29^{b'}$		EM × S	0.666	0.927
PRG (% in total)	PR	1.4^{a}	$^{B}12.8^{b}$	$^{B}18.5^{c}$	10.9	EM	0.72	<0.001
	PR+TC	1.0^{a}	$^{A}7.8^{b}$	$^{A}8.9^{b}$	5.9	S	0.59	<0.001
	Mean	1.2	10.3	13.7		EM × S	1.02	0.002
Red clover (% in total)	PR	86.7	$^{B}85.2$	$^{B}79.4$	83.8	EM	1.14	<0.001
	PR+TC	86.7^{b}	$^{A}73.0^{a}$	$^{A}71.6^{a}$	77.1	S	0.93	<0.001
	Mean	86.7	79.1	75.5		EM × S	1.61	0.006
Cocksfoot (% in total)	PR+TC	$3.9^{a'}$	$16.4^{b'}$	$16.0^{b'}$	-	EM	1.24	<0.001
Timothy (% in total)	PR+TC	$0.3^{a'}$	$1.2^{b'}$	$1.6^{b'}$	-	EM	0.17	0.005

[1] Means within columns ($^{a',b',c'}$) differ based on Sidak test, $P<0.05$; Means within rows/columns ($^{a,b,c/A,B}$)differ based on Bonferroni adjusted test, $P<0.05$; Standard error of the mean (SEM) applies to means on square root and angular scale ($^{\$,\ ¥}$), respectively.

References

Crotty F.V., Fychan R., Sanderson R., Rhymes J.R., Bourdin F., Scullion J. and Marley C.L. (2016) Understanding the legacy effect of previous forage crop and tillage management on soil biology, after conversion to an arable crop rotation. *Soil Biology and Biochemistry* 103, 241-252.

Gudleifsson B.E., Andrews C.J. and Bjornson H. (1986) Cold hardiness and ice tolerance of pasture grasses grown and tested in controlled environments. *Canadian Journal of Plant Science* 66(3), 601-608.

Turner L.R., Holloway Phillips M.M., Rawnsley R.P., Donaghy D.J. and Pembleton K.G. (2012) The morphological and physiological responses of perennial ryegrass (*Lolium perenne* L.), cocksfoot (*Dactylis glomerata* L.) and tall fescue (*Festuca arundinacea* Schreb.; syn. *Schedonorus phoenix* Scop.) to variable water availability. *Grass and Forage Science* 67, 507-518.

Measures to control yellow rattle in extensive grassland

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Abstract

Due to farm abandonment and changes in land use, the proportion of extensively used grasslands is steadily decreasing. When adequately managed, these areas have significantly higher floristic biodiversity than intensively used grasslands. Because of the moderate use, various flowering herbs can persist in these stands. However, in addition to the desired flowering herbs, undesirable stand partners can also establish and cause lasting damage. One of these undesirable herbs is the yellow rattle (*Rhinanthus minor*). We conducted a field trial at AREC Raumberg-Gumpenstein on a long-term experimental plot to address this problem. On a two-cut grassland that had been unfertilized for ten years, we investigated the effects of moderate fertilization and a single early first cut on species group ratio and, in particular, on the fraction of yellow rattle. In the year following the early first cut, we observed a significant decrease of yellow rattle compared to the usual cutting time.

Keywords: extensive grassland, *Rhinanthus minor*, weed control

Introduction

Compared to intensively used grasslands, semi-natural, extensively used grasslands have much higher plant biodiversity (Pötsch *et al.*, 2005) and provide a habitat for many animal species. Unfortunately, due to farm abandonment and changes in land use, the proportion of these ecologically valuable grasslands is steadily decreasing. In Austria they have decreased by about 50% in the last 60 years. On the one hand, high-yielding sites have been intensified, while low-yielding sites have been abandoned or selectively afforested. This is associated with biodiversity loss, which poses increasing existential problems for bees, wild bees, and other flower-pollinating insects (Seibold *et al.*, 2019). Therefore, great attention must be paid to their maintenance to preserve the remaining areas.

On the one hand, biodiversity in these areas should be promoted. On the other hand, the effort for conservation must be kept as low as possible to allow sustainable management practices. The extensive two-cut use with a late first cut allows the flowering herbs to seed naturally, but this type of maintenance also provides ideal conditions for some undesirable stand partners. One of these species is the yellow rattle (*Rhinanthus minor*). This species resides as a hemiparasitic plant mainly on legumes and grasses and can affect the entire plant community (Mudrák and Lepš, 2010). While a small proportion of this species is valuable for biodiversity, its increased presence can reduce forage yield and projective cover of valuable flowering forbs. In literature, an early first cut is mentioned as a possible control measure, but there are few accurate data available (Westbury, 2004). For this reason, we conducted a field trial at AREC Raumberg-Gumpenstein in 2020. We investigated the effect of a single early first cut on species group ratio and, in particular, on the proportion of yellow rattle in unfertilized and moderately fertilized grassland.

Materials and methods

We established the field experiment at AREC Raumberg-Gumpenstein, Austria (47°29N, 14°06E; elevation 710 m a.s.l.) in May 2020 on a long-term experimental plot. The plant stand on the experimental plots was a species-rich *Arrhenatherion* meadow, established in 2009 and it received no fertilization until autumn 2018 (Schaumberger *et al.*, 2021). In October 2018, we started different fertilization treatments. This paper considers only the unfertilized control and one other treatment, fertilized with 45 kg ha^{-1} N (compost fertilizer). To investigate the effects of the early first cut, the initial plant population of the fertilized and the unfertilized plots (14.5×3 m) was surveyed in June 2020, each in twelvefold repetition. For this purpose, we estimated the projective cover of species groups (grasses, legumes, herbs) and the projective fraction of yellow rattle. After the surveys, we split the plots and we mowed one-half (7.25×3 m) on June 19 before yellow rattle seed maturity. We mowed the second half three weeks later on July 8 (regular cutting date). We made the second cut on September 15 for all treatments. To investigate the effectiveness of early cutting, we assessed the same parameters again in June 2021. We performed the analysis with the statistical program R (product version). Due to lack of normal distribution, we performed a Kruskal-Wallis test and a Shapiro rank-sum test. The significance of the differences was calculated with a significance level of $P<0.05$.

Results and discussion

The studies from the initial situation showed that compost-fertilized plots had a higher proportion of projective cover for grasses and legumes compared to the non-fertilized plots. The proportion of projective cover for herbs was almost the same in both variants and there were no differences in the fraction of yellow rattle in 2020 (Table 1).

In 2021, the effects of the early first cut in the previous year were evident. In both fertilization treatments, the proportion of yellow rattle decreased significantly due to the earlier cutting time, whereas it increased significantly under regular management (Table 2). In all cases, the increase in yellow rattle was associated with a substantial increase in the proportion of herbs. In the compost-fertilized treatment, the proportion of legumes increased regardless of the cutting time, whereas, in the unfertilized treatment only an earlier cut led to an increase in legumes. The early first cut also showed an increase in grasses, although this was only significant in the compost-fertilized variant.

Concerning the promotion of biodiversity, the early cutting time in the unfertilized variant is particularly interesting. On the one hand, the fraction of yellow rattle was significantly reduced, and on the other hand, the general proportion of herbs and legumes increased strongly. The increasing inflorescences of different flowering species also better support the different pollinator groups.

Table 1. Comparison of the mean projective cover of species groups and *Rhinanthus minor* [%] within certain years.[1]

	2020				2021			
	Grasses	Herbs	Legumes	*R. minor*	Grasses	Herbs	Legumes	*R. minor*
Unfertilized	18.50^{b}	29.33^{a}	15.50^{b}	7.76^{a}	14.92^{c}	57.75^{c}	15.67^{c}	16.83^{b}
Compost fertilized	22.67^{a}	30.33^{a}	20.33^{a}	8.94^{a}	25.25^{b}	45.58^{b}	27.50^{b}	15.17^{b}
Unfertilized; early first cut in 2020					21.67^{b}	45.00^{b}	26.67^{ab}	2.46^{a}
Compost fertilized; early first cut in 2020					37.92^{a}	28.33^{a}	33.33^{a}	1.88^{a}

[1] Index a-c stands for the differences between treatments within a particular year.

Table 2. Comparison of the mean projective cover of species groups and *Rhinanthus minor* [%] within certain fertilization treatments.[1]

	Unfertilized				Compost fertilized			
	Grasses	Herbs	Legumes	*R. minor*	Grasses	Herbs	Legumes	*R. minor*
2020	18.50^{ab}	29.33^{a}	15.50^{a}	7.76^{a}	22.67^{a}	30.33^{a}	20.33^{a}	8.94^{a}
2021 (early first cut in 2020)	21.67^{a}	45.00^{b}	26.67^{b}	2.46^{b}	37.92^{b}	28.33^{a}	33.33^{b}	1.88^{b}
2021	14.92^{b}	57.75^{c}	15.67^{a}	16.83^{c}	25.25^{a}	45.58^{b}	27.50^{c}	15.17^{c}

[1] Index a-c stands for the differences between treatments within a particular fertilization level.

The results clearly show the significant impact that simple management changes can provide in the short term. However, the trial will be further monitored to understand the sustainability of this one-time measure.

Conclusions

The research showed a significant reduction of yellow rattle due to an early first cut in the previous year, both in the unfertilized and in the fertilized treatment. An early first cut thus offers a good control strategy, but the sustainable effect needs to be further investigated.

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References

Mudrák O. and Lepš J. (2010) Interactions of the hemiparasitic species *Rhinanthus minor* with its host plant community at two nutrient levels. *Folia Geobotanica* 45, 407-424. https://doi.org/10.1007/s12224-010-9078-1

Pötsch E.M., Blaschka A. and Resch R. (2005) Impact of different management systems and location parameters on floristic diversity of mountainous grassland. *Grassland Science in Europe* 10, 315-318.

Schaumberger S., Blaschka A., Krautzer B., Graiss W., Klingler A. and Pötsch E.M. (2021) Successful transfer of species-rich grassland by means of green hay or threshing material: Does the method matter in the long term? *Applied Vegetation Science* 24(3), e12606. https://doi.org/10.1111/avsc.12606

Seibold S., Gossner M. M., Simons N. K., Blüthgen N., Müller J., Ambarli, D. and Weisser W. W. (2019) Arthropod decline in grasslands and forests is associated with landscape-level drivers. *Nature* 574(7780), 671-674. https://doi.org/10.1038/s41586-019-1684-3

Westbury D.B. (2004) *Rhinanthus minor* L. *Journal of Ecology* 92, 927. https://doi.org/10.1111/j.0022-0477.2004.00929.x

Grass-clover leys for a sustainable N yield: *Trifolium pratense* cultivar × mixture effects

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Abstract

Grass-clover leys allow sustainable intensive forage and livestock production but large pedoclimatic differences make mixture × cultivar trials necessary to identify site-specific optimal combinations. We addressed experimentally the questions: (1) whether cultivars of *Trifolium pratense* must be evaluated in a specific seed mixture, or whether their biomass and N yield and symbiotic N_2 fixation can be ranked in any mixture; and (2) whether mixtures perform differently depending on the choice of *T. pratense* cultivar. The trial was set up with three cultivars (Milvus, Semperina, Spurt) and three regionally recommended grass-clover mixtures (IR, KG, WW), differing, among others, in *T. pratense* abundance. At the third cut after autumn seeding, *T. pratense* dominated the dry matter and N yield in mixture- and cultivar-specific manners. Irrespective of mixture and *T. pratense* cultivar, 56.7±1.1% of total N was acquired via symbiotic N_2 fixation, owing to compensatory dynamics in sward structure and degree of reliance on N_2 fixation. Cultivars Spurt and Milvus boosted dry matter and N yield, with cv. Spurt accumulating most N, and cv. Semperina tending to acquire proportionally most N via symbiosis. Our findings suggest that mixture × cultivar evaluation trials may not always be necessary as long as the mixtures are of similar composition and demonstrate N yield stability, despite different structure.

Keywords: agronomic performance, cultivar, leys, symbiotic N_2 fixation, red clover

Introduction

Multispecies temporary grassland for intensive forage production regenerates soil fertility, thereby contributing to globally sustainable food production through reliance on local feed stuff (Jan *et al.*, 1992). In climatic regions with cold winters and thus relatively short growing seasons, but sufficient precipitation, *Trifolium pratense* is the forage legume of choice to increase forage quality and late-season yield. However, despite big strides in breeding and designing grass-clover mixtures (Grieder *et al.*, 2019), it is still unclear: (1) whether the relative performance of cultivars of *T. pratense* must be tested in specific target mixtures or if the ranking of cultivar performance in one standard mixture is also meaningful for other mixtures; and (2) whether cultivars of *T. pratense* exert consistent effects on parameters of agronomic interest across different mixtures. We addressed these questions in respect to the agronomic traits of dry matter (DM) and N yield, and the sustainability trait of symbiotic N_2-fixation, using a two-factorial seed mixture and cultivar evaluation trial.

Materials and methods

Three cultivars of *T. pratense* (Milvus, Semperina, Spurt), which had been found to perform similarly in a monospecific cultivar trial at the same site, were alternatively used in three grass-clover seed mixtures for leys [KG and WW, recommended for South Tyrol, and IR, recommended for Austria (Krautzer *et al.*, 2020)], differing primarily in seed weight proportion of several species and showing an approximate two-fold reduction in the abundance of *T. pratense* from 27.3% in KG to 13.3% in IR and to 5.0% in WW. The other legume components in the mixtures were *Trifolium repens*: IR, 8.8%; KG, 4.6%; WW, 8.0%, and *Trifolium hybridum*: KG, 4.5%; missing in the other seed mixtures. The trial was setup on 29.08.2019 in Teodone/Dietenheim (South Tyrol, NE Italy; 46° 48' 8.064' N, 11° 57' 23.908' E; 891 m a.s.l., 8.4 °C mean annual temperature, 733.5 mm mean annual precipitation) in a randomized complete block design with three replicates and a plot size of 1.2×7.2 m. The total soil N content was 1.17 g kg^{-1} prior to application of 40 kg N ha^{-1} and 53 kg ha^{-1} K in mineral form in spring and of 15 m^3 ha^{-1} digested cow slurry after the first and second of four annual harvests. The yield of *T. pratense*, *T. repens*, and of the remaining fraction (mostly grass) of the third harvest on 12 August 2020 was assessed by means of manual species separation from a 0.25 m^2 sampling area per plot. The N concentrations ([N]) and $\delta^{15}N$ signatures of the shoots were determined by isotope ratio mass spectrometry. Dry matter (DM) yield was assessed at plot level, as was the [N], measured according to Dumas. The proportion of N derived from the atmosphere (pNdfa) by symbiotic N_2 fixation was calculated at plot level, using ^{15}N natural abundances and the formula $(\delta^{15}N_{Grasses} - \delta^{15}N_{Clover\ species}) / (\delta^{15}N_{Grasses} - B)$, where $\delta^{15}N$ stands for the $^{15}N/^{14}N$ isotope ratio in per mil, following Unkovich *et al.* (2008). The B-values used were the means for shoots of the *Trifolium* spp. determined of Carlsson *et al.* (2006). N yields, Ndfa, and N derived from soil (Ndfs) were calculated as the products of the DM yields and [N], pNdfa, and (1-pNdfa), respectively. The data was analysed by ANOVA accounting for seed mixture, *T. pratense* cultivar, their interaction, and block, all treated as fixed factors, followed by LSD test. N yield and pNdfa of *T. repens* were square root-transformed to meet the ANOVA assumptions. The significance level was set at P=0.05.

Results and discussion

The DM and N yields of the swards were similarly affected by seed mixture and *T. pratense* cultivar, but the effect of seed mixture was about four-fold stronger than that of cultivar for the yield of *T. pratense* and even ten-fold stronger for that of *T. repens* (Figure 1) due to the differences in the abundance of the individual clover species. For none of the investigated parameters was there an interaction found between seed mixture and *T. pratense* cultivar. The DM yield of the seed mixtures ranked: KG > WW with IR not significantly differing from each of them, and the DM and N yields of the *T. pratense* cultivars: Spurt > Semperina with Milvus not significantly differing from each of them. Ndfa contributed on average 56.7±1.1% (n=27, standard error of the mean) to the N yield of the swards, irrespective of seed mixture, *T. pratense* cultivar, and mixture × cultivar combination. Cultivar Semperina tended to acquire most N via symbiosis, but this positive trait traded off against a low DM yield in mixture. The *T. repens* and grass fraction of the seed mixtures largely compensated *T. pratense* abundance and cultivar effects on pNdfa. Compensatory dynamics between mixture components hence explain the stable and high N yields of the evaluated grass-clover ley mixtures as known from other studies (Finn *et al.*, 2013; Hejduk *et al.*, 2010). However, the agronomic relevance of these findings from just one harvest in just the first year of three cultivation years needs to be verified with the data of the entire three cultivation years and will afterwards have to be confirmed in multi-site trials under different fertilization regimes.

Conclusions

Absence of statistical interactions between the factors seed mixture and *T. pratense* cultivar for all agronomically relevant parameters of the third harvest in the first cultivation year suggest that testing cultivars in all possible target mixtures might not always be necessary. Our findings show, however, that testing the cultivars in mixed swards is necessary to identify the best performing *T. pratense* cultivar.

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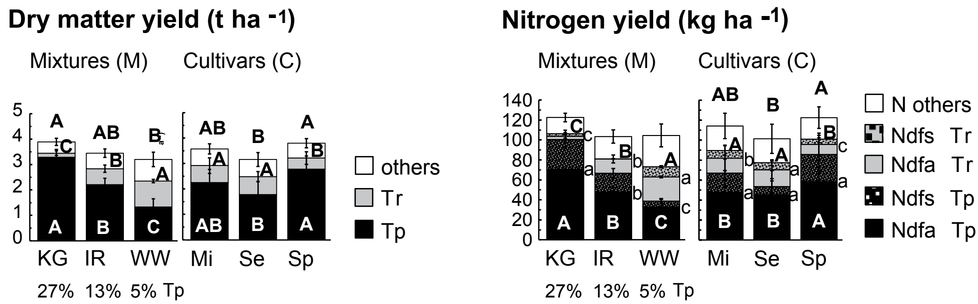

ANOVA:	M(F2,16):	C(F2,16):	M x C(F4,16):
Sward:	4.768*	4.095*	0.488 ns
Tr:	40.720***	4.791*	1.285 ns
Tp:	28.847***	7.568**	0.231 ns

Note: Block(F2,16) was never significant.
*, P ≤ 0.05; **, P ≤ 0.01; ***, P ≤ 0.001

ANOVA:	M(F2,16):	C(F2,16):	M x C(F4,16):
N sward:	3.461 ns	4.762*	1.664 ns
N Tr:	37.550***	4.031*	1.101 ns
Ndfa Tr:	60.912***	12.029***	1.200 ns
N Tp:	29.238***	8.051**	0.169 ns
Ndfa Tp:	11.060***	0.598 ns	0.703 ns

Note: Block(F2,16) was never significant.
*, P ≤ 0.05; **, P ≤ 0.01; ***, P ≤ 0.001

Figure 1. Differences in dry matter and nitrogen (N) yield of three grass-clover seed mixtures (KG, IR, WW), differing in seed weight percentage and cultivar identity [Milvus (Mi), Semperina (Se), Spurt (Sp)] of *Trifolium pratense* (Tp). Means, standard errors, and associated statistical results of ANOVA and multiple comparisons by LSD are shown for the entire sward and the *T. pratense* and *T. repens* (Tr) components (n=9) in capital letters and for the amounts of N derived from the atmosphere (Ndfa) and soil and fertilizer (Ndfs) by the two *Trifolium* species in small letters.

References

Carlsson G., Palmborg C., and Huss-Danell K. (2006) Discrimination against ^{15}N in three N_2-fixing *Trifolium* species as influenced by *Rhizobium* strain and plant age. *Acta Agriculturae Scandinavica, Section B – Soil & Plant Science* 56, 31-38.

Finn J.A., Kirwan L., Connolly J., Sebastià M.T., Helgadottir A., Baadshaug O.H., ... and Lüscher A. (2013) Ecosystem function enhanced by combining four functional types of plant species in intensively managed grassland mixtures: a 3-year continental-scale field experiment. *Journal of Applied Ecology* 50, 365-375.

Hejduk S. and Knot P. (2010) Effect of provenance and ploidity of red clover varieties on productivity, persistence, and growth pattern in mixture with grasses. *Plant Soil and Environment* 56, 111-119.

Jan P., Repar N., Nemecek T. and Dux D. (2019) Production intensity in dairy farming and its relationship with farm environmental performance: Empirical evidence from the Swiss alpine area. *Livestock Science* 224, 10-19.

Grieder C., Tanner P. and Schubiger F. (2019) Selection progress in a commercial forage breeding programme. *Grassland Science in Europe* 24, 385-387.

Krautzer B., Egger H., Frank P., Frühwirth P., Graiss W., Greisberger M. ... and Starz W. (2020). *Handbuch für ÖAG Qualitätssaatgutmischungen - Dauergrünland und Feldfutterbau - Mischungssaisonen 2020/21/22*. HBLFA Raumberg-Gumpenstein, Irdning, Austria, 40 pp.

Unkovich M., Herridge D., Peoples M., Cadisch G., Boddey B., Giller K., Alves B. and Chalk P. (2008) *Measuring plant-associated nitrogen fixation in agricultural systems*. ACIAR Monograph, Canberra, Australia. 258 pp.

A multicriteria method to evaluate the resilience of grass-based dairy farms to climate change in Brittany

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Abstract

Grass-based dairy systems have proven their interest for sustainability, but their resilience to climate change has been less studied. As crop diversification is usually seen as good practice for more resilient farming systems, we want to study if the sole use of grasslands as forage resources is less resilient than the use of grass and maize. To do so, we designed a three-step method for assessing the resilience of dairy farms to climate change. First, we established correlations between farm performance indicators and climatic variables during the last 5 years, to estimate their vulnerability and their adaptations to these external drivers. Then, we compared the different farming systems through a multi-criteria approach of resilience co-designed with the farmers. Our results show that climatic variables have a low impact on the performances of studied farms. However, grass-based systems experience more inter-annual variations in milk production than grass maize systems, while their economic performances are more robust. This work provides a useful tool for resilience assessment of dairy farms.

Keywords: resilience, multi-criteria assessment, dairy farm, forage system

Introduction

The latest IPCC report predicts a future increase in the frequency and intensity of extreme heat and heavy precipitation in France, which will affect agricultural systems. Brittany is the main dairy region in France. A wide diversity of forage systems is observed in the region, with a majority of dairy farms having more than 40% maize silage in their forage area, but also a significant proportion of farms based mostly on grassland.

These grass-based dairy farms have shown their performance in terms of sustainability (Alard *et al.*, 2002). However, they face questions regarding their capacity to cope with climate change. In particular, their dependence on grasslands raises the question of their greater vulnerability compared to more diversified farms, with crop diversification identified as a major strategy for increasing resilience to climate change (Bowles *et al.*, 2020).

A collaborative study between a farmers' association (CEDAPA) and researchers was set up to work on this issue. The present work details the construction of a method for assessing the resilience of dairy farms to climate change and the first results obtained. More detailed results can be found in the project report (Geffroy, 2021).

Materials and methods

Characterization of the vulnerability of farms to hazards

This step is inspired by the work of Martin *et al.* (2017). Its objective is to define the vulnerability of farms to climatic events, considering that a farm less vulnerable to hazards is more resilient. We chose five vulnerability variables that we could collect over the previous five years, between 2015 and 2019.

The variables had to be archived and common to all farmers, so we chose accounting data: economic efficiency (gross margin / product), labour efficiency (gross operating surplus / annual work unit), available income (gross operating surplus - annuity - inventory changes), milk gross margin per ha forage, and animal productivity (sold milk / number of dairy cows). Four climatic exposure variables were also selected based on Martin *et al.* (2017) and expert knowledge: spring precipitation (from April to June), summer precipitations (July-September), summer + spring precipitation and number of days with heat stress (above 25 °C). Then, we used a mixed linear model to analyse the effect of climatic exposure variables on vulnerability variables, with year as a random effect.

Analysis of vulnerability through a set of resilience indicators

Around eighty indicators linked to farm resilience were collected through a literature review. They were then presented to a group of farmers, advisers and INRAE scientists in order to collectively select a smaller panel with the most relevant indicators covering the different dimensions of resilience. Each participant had to select the twenty indicators that seemed most relevant to evaluate the various dimensions of farm resilience, with the possibility of proposing new ones. After a collective discussion, the indicators that received more votes were selected. Then, we attempted to explain farm vulnerability with this set of resilience indicators.

Data collection

To test our method, 29 dairy farmers were interviewed. All farmers are part of the CEDAPA farmers' association and located in Brittany, mostly in Côtes-d'Armor. The choice of farms was made randomly according to the interest and availability of the farmers during the survey phase which took place from 18 May to 26 June 2021. During the survey, we collected all needed information to calculate the indicators, as well as technical data for farm description and information relative to the implementation of adaptations to climate change. The climatic data used is modelled daily data from the French meteorological model SAFRAN for the five studied years. We selected the 8×8 km grid cell in which each farm was located.

Results and discussion

An analysis of climatic exposure variables showed that the five years were significantly different, with an unusually dry summer in 2016 (88 mm), a variable distribution of rainfall between spring and summer and less heat stress in 2015. However, although these variations affected crop yields, only one vulnerability variable, the labour efficiency, was significantly correlated with a climatic variable: it increased when spring and summer rainfall increased (P=0.025). This is not unexpected, as important spring and summer rainfall increase forage production and can reduce the need to purchase external feed. However, what is surprising is the lack of correlation with other economic variables. This reveals that the 29 surveyed farms were not very economically vulnerable to the chosen climatic variables for years 2015-2019. The survey showed us that a large majority of farmers have already implemented adaptation strategies, for instance lowering animal density per hectare, diversifying plant species in grasslands, making security forage stocks or grouping calvings in spring. This might partly explain their low vulnerability to climatic hazards.

The analysis of vulnerability variables with our set of resilience indicators showed that milk production per cow was much more variable from year to year in farms with more grass than in farms with more maize in the ration (Figure 1; P=0.03). Those grass-based farms can therefore be defined as more vulnerable to hazards. However, we also observed that these farms have a better average revenue and a lower variation of economic efficiency (gross margin / product; P=0.09).

Grass has greater fluctuations in yield than silage maize (Devun *et al.*, 2013), which can explain the greater variation in milk production of grass-based farms. Moreover, many of these farms seek to be self-sufficient by reducing external inputs. Therefore, a variation in milk production might not be endured by

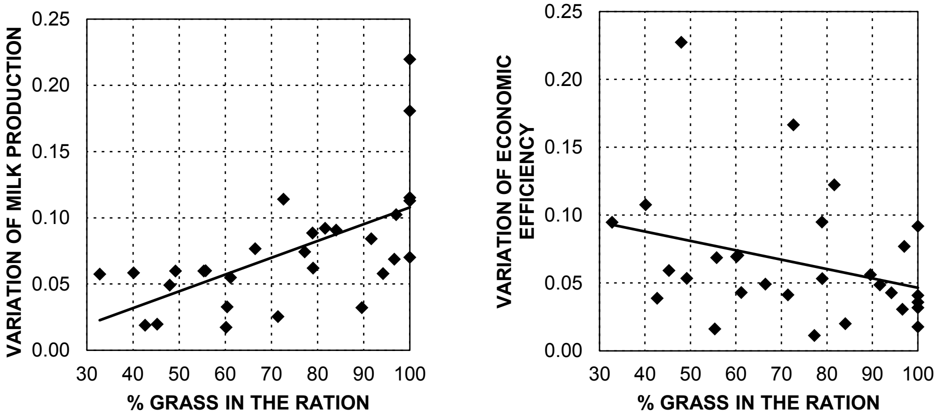

Figure 1. Interannual variation coefficients of milk production and economic efficiency compared to percentage of grass in the ration of dairy cows (yearly data, 2015-2019).

farmers but rather part of a strategy of autonomy, while other farmers might in this case purchase external feed. These characteristics could explain why we observed a lower variation of economic efficiency in these grass-based systems, as other authors do (Michaud *et al.*, 2021).

Conclusions

We developed a method to evaluate the resilience of dairy farms. This work showed that dairy farmers in Brittany are currently resilient to climatic hazards, possibly due to their observed adaptation to climate change. Studied grass-based dairy systems seem to accept more variability in milk production than grass-maize systems, but have a better economic resilience. Our method seems an interesting tool to analyse the resilience of dairy farms.

References

Alard V., Béranger C. and Journet M. (2002) A la recherche d'une agriculture durable: étude de systèmes herbagers économes en Bretagne. Editions Quae, Paris, France. 346p.

Bowles T.M., Mooshammer M., Socolar Y., Calderón F., Cavigelli M.A., Culman S.W. ... and Grandy A.S. (2020) Long-term evidence shows that crop-rotation diversification increases agricultural resilience to adverse growing conditions in North America. *One Earth* 2(3), 284-293.

Devun J., Moreau J.-C., Lherm M. and Mosnier C. (2013). Variabilité interannuelle des productions fourragères de 2000 à 2011. Analyse par région à partir d'observations en fermes. *Fourrages* 215, 221-230.

Geffroy K. (2021) Study of the resilience of grass-based dairy systems in the Côtes-d'Armor to face climate change. Masters thesis report, VetAgroSup, Lempdes, France. 116p.

Martin G., Magne M.-A. and Cristobal M.S. (2017) An integrated method to analyze farm vulnerability to climatic and economic variability according to farm configurations and farmers' adaptations. *Frontiers in Plant Science* 8, 1483.

Michaud A., Plantureux S., Baumont R. and Delaby L. (2021) Les prairies, une richesse et un support d'innovation pour des élevages de ruminants plus durables et acceptables. *INRAE Productions Animales* 33(3), 153-172. https://doi.org/10.20870/productions-animales.2020.33.3.4543

Drought and plant diversity effects on the agronomic multifunctionality of intensively managed grassland

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Abstract

Multi-species grassland mixtures offer an opportunity to increase the sustainability of production from intensively managed European grasslands. However, a larger panel of ecosystem functions are now expected to be delivered from grasslands, with increasing emphasis on environmental issues. Maintaining performance under extreme weather events such as summer drought is now of great importance, due to climate change. In grassland communities subjected to a two-month experimental drought in 2018 and 2019, we investigate the effects of mixing species and functional groups with the aim of improving grassland forage production, forage quality and other sustainability metrics. From a pool of six plant species and three functional groups (grass, legume and herb), we sowed communities with 1, 2, 4, 5 and 6 species in a field experiment in Wexford, Ireland. Plots were managed intensively with regular harvests and fertilizer application during two years of a grassland phase, followed by one year of crop to simulate a 3-year grassland-crop rotation. We modelled and compared the effect of plant diversity on multiple functions: dry matter yield, nitrogen yield, forage digestibility, yield stability, weed suppression and legacy effect on a follow-on crop. Results showed a higher performance in multiple functions (multifunctionality) from grassland mixtures exposed to drought stress, compared to monocultures. We conclude that plant diversity is a reliable agronomic tool to enhance temperate grassland sustainability.

Keywords: multifunctionality, intensive, diversity, interaction, drought, nitrogen

Introduction

Agricultural intensification can cause decreases in many ecosystem functions. In addition, extreme weather events induced by climate change are increasing threats to agricultural systems, and may cause further declines in ecosystem functions. Plant diversity in grasslands can be a practical way to enhance individual functions under intensive management, such as productivity (Grange *et al.*, 2021) or weed suppression. Suter *et al.* (2021) showed that interactions between legume and non-legume enhanced grassland multifunctionality (MF). Multi-species grasslands can also improve resistance and resilience to drought (Haughey *et al.*, 2018). We aim to assess the effect of plant diversity on grassland agronomic multifunctionality across a rotation when stressed by drought. Under seasonal drought, we investigate the effect of functional group (FG) diversity and interactions on different functions, and identify trade-offs between functions or FG composition.

Materials and methods

A grassland experiment manipulating plant diversity was established in 2017. Commercial cultivars of six perennial competitive forage species from three functional groups (grass: *Lolium perenne* and *Phleum pratense*, legume: *Trifolium pratense* and *Trifolium repens*, and herb: *Cichorium intybus* and *Plantago lanceolata*) were combined in a simplex design to form 19 communities of 1 to 3 FGs. All 39 plots were managed intensively (150 kg N ha^{-1} yr^{-1} of fertilizer and seven cuts yr^{-1}) and subdivided in two halves with a water supply treatment randomly allocated to each half: either rainfed control or drought, where rainfall was excluded for two months. Here, we present results only from the drought treatment.

We measured several ecosystem functions in each plot. Average annual dry matter yield (DMY) was measured in 2018 and 2019. Standard deviation of the harvested biomass across harvests was used to indicate yield stability and harvested material was analysed to estimate forage digestibility. Weed suppression was calculated as one minus the proportion of weed biomass. In 2020, the plots were sprayed with herbicide to establish a *Lolium multiflorum* model crop. Keeping the same field layout and managing all plots the same in 2020 (40 kg N ha^{-1}, 4 cuts, no experimental drought), we measured the net effect of the preceding grassland communities on the yield of a follow-on crop, the legacy effect. Total nitrogen yield (NY) was measured across 2018, 2019 and 2020 after measuring nitrogen content in harvested biomass from each plot. Data analysis was performed on data that were standardized to scale values from zero to one, with one representing the maximum performance (averaged across the top three observations). A multivariate Diversity-Interactions model was fitted to the above-named functions as responses, with species identities and interactions being the explanatory variables (see Dooley *et al.* (2015) for more detail on the method). This approach enabled simultaneous assessment of the link between grassland diversity and multiple ecosystem functions, taking the covariance among functions into account.

Results and discussion

There was a significant reduction in ecosystem function due to experimental drought (unpublished data) but plant diversity was strongly associated with increased multifunctionality within grassland communities subjected to drought (Figure 1).

Across most functions, there were strong dissimilarities across different FGs. Digestibility was stable, and attained high levels across all FG combinations. In contrast, other functions were driven by functional group composition. The grass FG monocultures favoured higher yield and weed suppression, with low legacy effect and low NY. The legume FG monocultures were not able to suppress weeds, but had both a high legacy effect and NY. Finally, the herb FG monocultures showed high yield stability, and had mixed performances in other functions. These divergent outcomes highlight trade-offs between functions when grasslands are sown with monocultures. As an example, the legacy effect and weed suppression were negatively correlated.

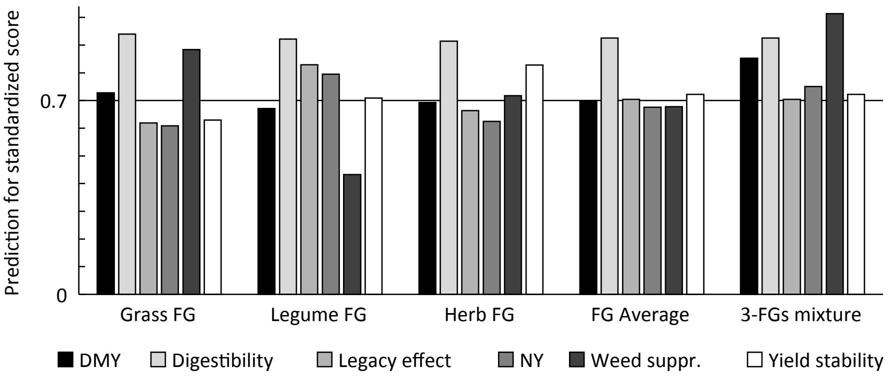

Figure 1. Modelled predictions for standardized performance (scaled from 0 to 1) for the sub-plots subjected to experimental drought. Values are presented for each monoculture of the grass, legume and herb functional groups (FG). Each FG displayed is the average of the multifunctionality values for each of the two component species monocultures. The last two groups display the average of each of the FGs (= the average of all 6 monocultures), and the 3-FG mixture (= the equi-proportional 6-species mixture). DMY = dry matter yield of harvested forage; NY = total nitrogen yield over three years; weed suppr. = weed suppression.

Comparing the FG average with the equi-proportional 3-FG mixture (Figure 1) shows the effect of significant FG interactions, i.e. the synergistic benefit of mixing the species together. These interactions strongly enhanced DMY and weed suppression and increased NY. For DMY and weed suppression, there was a significant transgressive over-performance, i.e. a higher performance from the mixture than any of the component FGs. The other functions were not affected by FG interactions; thus, their performance in mixture was a linear combination of the species' performance in monoculture. The absence of between-FG interaction effects, however, did not mean an absence of effects of plant diversity; for example, the 3-FG equi-proportional mixture performed as predicted from the average of all FGs in monoculture, thus mitigating the occurrence of extreme values for these functions. The combined effects of diversity and interactions resulted in the equi-proportional 3-FG mixture achieving a score of >0.7 for all functions. None of the three FGs (nor their component species as monocultures) achieved such a threshold for all six functions (Figure 1).

Conclusions

Grassland communities were exposed to two-month experimental droughts in two successive years before establishing a crop in a rotation. Within the communities subjected to drought, plant diversity was associated with an overall improvement in multiple ecosystem functions. Trade-offs between functions occurred in monocultures; in mixtures, interactions among FGs moderated the occurrence of trade-offs between functions.

Acknowledgements

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References

Dooley A., Isbell F., Kirwan L., Connolly J., Finn J.A. and Brophy C. (2015) Testing the effects of diversity on ecosystem multifunctionality using a multivariate model. *Ecology Letters* 18, 1242-1251.

Grange G., Finn J.A. and Brophy C. (2021) Plant diversity enhanced yield and mitigated drought impacts in intensively managed grassland communities. *Journal of Applied Ecology* 58, 1864-1875.

Haughey E., Suter M., Hofer D., Hoekstra N.J., McElwain J.C., Luscher A. and Finn J.A. (2018) Higher species richness enhances yield stability in intensively managed grasslands with experimental disturbance. *Scientific Reports* 8, 15047.

Suter M., Huguenin-Elie O. and Luscher A. (2021) Multispecies for multifunctions: combining four complementary species enhances multifunctionality of sown grassland. *Scientific Reports* 11, 3835.

Dry matter production of multispecies swards at three nitrogen application rates under dairy grazing

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Abstract

Irish dairy farmers have become increasingly interested in the use of multispecies (MS) swards for intensive grazing systems. A grazed plot experiment was established to investigate the dry matter (DM) yield of multispecies swards containing three plant functional groups: grass, legume and herb, under three levels of nitrogen (N) fertilizer application (N100, N150 and N200) compared to zero N (N0). Ten sward types were established which included a perennial ryegrass (PRG, *Lolium perenne* L.) monoculture and sward mixtures of the following species: PRG, white clover (WC, *Trifolium repens* L.), red clover (*T. Pratense* L.), chicory (*Chicorium intybus* L.) and ribwort plantain (PL, *Plantago lanceolate* L.), with monoculture PRG N200 as the control. Plots were grazed by lactating dairy cows on eight occasions in year one and nine occasions in year two. Swards in the N200 treatment produced the highest level of DM with an average of 1,337±131 kg DM ha^{-1} (LS means ± standard error) more than N0 swards ($P<0.001$). The sward mixture of PRG, WC and PL produced the highest level of DM; 2,916±198 kg DM ha^{-1} more than the PRG monoculture sward on average across all levels of N fertilizer. These results indicate MS swards containing both legumes and herbs can contribute increased DM production, particularly where PL and WC are included.

Keywords: chicory, plantain, clover, grazing, dry matter production

Introduction

The majority of Irish dairy farms are grazing focused, as this is the most efficient method of harvesting herbage from pasture. In order to meet animal nutritional requirements farmers must use highly digestible pasture species that will provide sufficient dry matter (DM) throughout the grazing season. For the past number of decades perennial ryegrass (PRG; *Lolium perenne* L.) has dominated swards as it provides up to 15 t DM ha^{-1} $year^{-1}$ of highly digestible forage over a minimum ten-year period, allowing farmers to stock farms profitably at a low cost (O'Donovan *et al.*, 2011). Like many other pasture species, PRG is dependent on nitrogen (N) for growth and N is often supplied via chemical fertilizers. In recent years the use of white clover (WC; *Trifolium repens* L.) in PRG swards has increased as it can fix atmospheric N into plant-available forms and has been shown to improve animal performance (McClearn *et al.*, 2020). Recently, interest in the use of forage herbs in grazing swards has increased. Herbs such as ribwort plantain (*Plantago lanceolate* L.) and chicory (*Chicorium intybus* L.) are deeper rooting than PRG or WC and can provide resilience to drought and increased sward resource-use efficiency through complementarity (Grace *et al.*, 2018). Few data are available on the performance of herbs within multispecies (MS) swards under intensive dairy grazing as practised in Ireland. The objective of this study was to investigate the DM yield potential of MS swards, which include PRG, WC, red clover (RC; *Trifolium pratense* L.) and forage herbs, under dairy grazing at three different rates of N fertilizer.

Materials and methods

A grazed plot trial was sown in June 2019 at Teagasc Moorepark (52° 16'N; 8° 26'W). The soil type is a free draining acid brown earth soil of sandy loam texture. The experiment was conducted over two full grazing seasons, March-November, in 2020 and 2021.The trial included ten sward mixtures (Table 1), each of which were managed under four different rates of fertilizer N (kg ha^{-1} year^{-1}) – N0, N100, N150 and N200. The five species represented three plant functional groups. Species used were: PRG (cv. AberGain), WC (cv. Buddy), RC (cv. Amos), PL (cv. Tonic) and CH (cv. Puna). The PRG N200 treatment was the control. Plots were arranged in a randomized block design over three replicates. Nitrogen fertilizer was applied proportionally to each treatment throughout the year in the form of protected urea. Phosphorus, potassium and lime were supplied throughout the grazing season via chemical fertilizer as per annual soil test requirement.

Plots were grazed by 40 lactating dairy cows on 8 occasions in 2020 and 9 occasions in 2021, and grazed over 24-36 hours to a target residual height of 4 cm. Plots were measured and grazed when the control treatment was estimated to have reached a pre-grazing herbage mass of 1,200-1,400 kg DM ha^{-1} (>4 cm). Plot yield was measured immediately prior to each grazing by cutting a subsection of each plot using an Etesia mechanical harvester (Etesia Hydro 124D; Etesia UK Ltd.). Cut herbage was weighed and 0.1 kg dried at 90 °C for 16 hours to determine DM content. Data analysis was conducted using SAS 9.4; a linear mixed model was used to estimate DM production differences where N rate, sward mixture, year and associated interactions were included as fixed effects, and replicate was included as a random effect.

Results and discussion

There was a large difference in the yield of all sward types between evaluation years (P<0.001); swards produced an average of 3,537±92 kg DM ha^{-1} more DM in year 2 compared to year 1 (all data refer to LS means ± standard error unless otherwise stated). Sward mixture and N application rate were both associated with annual DM yield (P<0.001) across both evaluation years. The N200 treatment produced the highest level of DM in all swards except the PRG, WC and PL treatment where the N150 produced the highest DM of 12,022±295 kg DM ha^{-1}; N0 produced the lowest level of DM in all swards with an average of 9,467±111 kg DM ha^{-1}. The PRG, WC and PL sward treatment produced the highest level of DM on average across all N application rates (Figure 1) while the PRG monoculture produced lowest amount of DM. Swards including WC produced more than all others (P<0.001), except the PRG and PL treatment, on average across all N treatments. The inclusion of WC in swards is likely to have contributed to higher levels of N being available in the soil for use by all species; an increase in sward yield due to increasing levels of N via either high N fertilizer rates or inclusion of clover is to be expected (Ledgard *et al.*, 2009). The DM yield advantage from including herbs with PRG and WC agrees with the findings of Grace *et al.* (2018) where swards including grass, legumes and herbs produced more than a PRG and WC sward at similar N fertilizer rates. The DM production advantage of MS swards in the current study appears to rely on the inclusion of WC and PL in the sward mixture.

Table 1. Sward species mixture treatments.

Treatment	Sward mixture
1	perennial ryegrass
2	perennial ryegrass and white clover
3	perennial ryegrass and red clover
4	perennial ryegrass, white clover and red clover
5	perennial ryegrass and chicory
6	perennial ryegrass and plantain
7	perennial ryegrass, chicory and plantain
8	perennial ryegrass, white clover and plantain
9	perennial ryegrass, white clover, plantain and chicory
10	perennial ryegrass, white clover, red clover, plantain and chicory

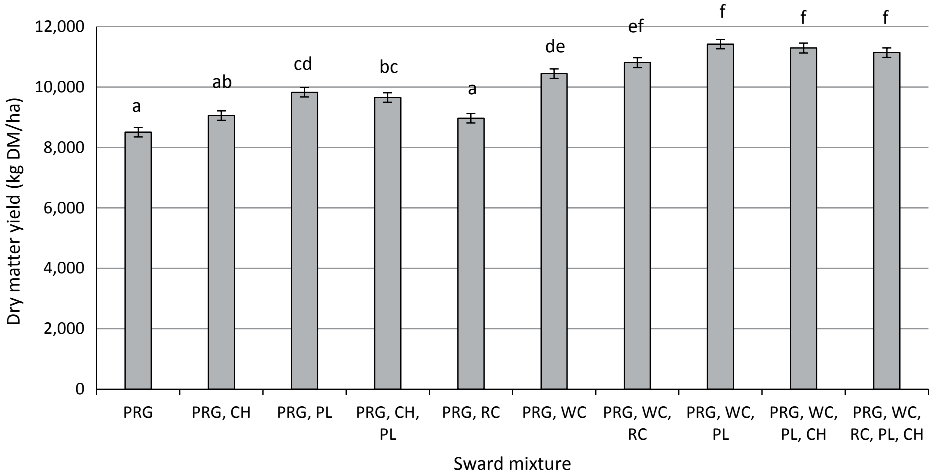

Figure 1. Mean dry matter production for each sward across all N treatments over two production years (2020 and 2021); error bars represent standard error. Swards with differing superscript letters differ significantly (P<0.05) in dry matter production.

Conclusions

Over the two evaluation years, including forage herbs and clover with PRG in sward mixtures was associated with increased DM production; inclusion of PL and WC in particular appears to contribute to increased DM production. Increasing N rate was also associated with increased DM production. Year had a large effect on sward DM production; further work is required to assess sward species content changes and DM production persistency over time.

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References

Grace C., Boland T.M., Sheridan H., Lott S., Brennan E., Fritch R. and Lynch M.B. (2018) The effect of increasing pasture species on herbage production, chemical composition and utilization under intensive sheep grazing. *Grass and Forage Science* 73, 852-864.

Ledgard S., Schils R., Eriksen J. and Luo J. (2009) Environmental impacts of grazed clover/grass pastures. *Irish Journal of Agricultural and Food Research* 48, 209-226.

McClearn B., Gilliland T., Guy C., Dineen M., Coughlan F. and McCarthy B. (2020) The effect of perennial ryegrass ploidy and white clover inclusion on milk production of dairy cows. *Animal Production Science* 60, 143-147.

O'Donovan M., Lewis E. and O'Kiely P. (2011) Requirements of future grass-based ruminant production systems in Ireland. *Irish Journal of Agricultural and Food Research* 50, 1-21.

Strengthening the resilience of grasslands against the unpalatable C4 grass *Setaria pumila*

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Abstract

With increasing frequency of dry and hot events, ryegrass-dominated grasslands become more vulnerable to infestation by the drought and heat resistant, but unpalatable C4 grass *Setaria pumila*. Grassland management must therefore be readjusted to strengthen resilience against weather hazards and resulting weed infestation. We investigated strategies to strengthen high quality forage grasses, as well as overseeding grass species more resistant to drought than ryegrass. The study involved a factorial experiment testing mowing height, time interval between harvests and overseeding, and an on-farm survey on the effects of farmers' management practices on *S. pumila* abundance in 31 grasslands. Increasing mowing height from 3 to 8 cm or skipping one or two harvests during the summer months reduced *S. pumila* abundance during the years of strong infestation. These management options were also clearly beneficial for the abundance of forage grasses in the sward. A negative relationship between *S. pumila* abundance and mowing height was also observed in the on-farm survey. Overseeding cocksfoot significantly increased its abundance only by the fourth experimental year. We conclude that careful management focusing on the competitive ability of forage grasses is an important driver of grassland resilience against infestation by *S. pumila*.

Keywords: yellow foxtail, cutting height, defoliation frequency, oversowing

Introduction

Setaria pumila (Poir.) Roem. & Schult. is an annual C4 grass native to Europe. In Switzerland it mainly occurs in the lowland, under full sun on moderately dry, nutrient-rich soils (Landolt *et al.*, 2010). It is a common arable weed (Dekker, 2003) and is on the rise in grassland areas and spreading into higher altitudes. It germinates on patches of bare soil starting mid-spring and develops its biomass during the hot summer months. Its biomass is unpalatable to livestock. In grasslands, *S. pumila* has been shown to benefit from dry and warm conditions (Orlandi *et al.*, 2015), and thus from climate change. This grass is currently a serious challenge for forage production in some Swiss regions. After cutting, it is able to replace the lost flower stems and produce mature seeds within a very short time. Seed propagation can therefore not be prevented by intensive mowing. The aim of this study was to test three management options to reduce the abundance of *S. pumila* in intensively used meadows, and to evaluate the potential importance of management practices encountered on-farm.

Materials and methods

A factorial field experiment was run from 2017 to 2021 in Buochs (lat/lon 46.97, 8.39; 450 m a.s.l.) to test: (1) mowing height, either low (3 cm) or high (8 cm); (2) intervals between harvests; and (3) overseeding, yes or no. The intervals between harvests were: (1) repeated harvesting at early heading stage of the ryegrass with six harvests per year, thereafter abbreviated 0Sk; (2) third growth cycle left standing until the date of the fourth harvest of 0Sk, i.e. skipping one harvest in July (1Sk); and (3) four harvests per year on the dates of the first, third, fifth and sixth harvests of 0Sk, i.e. two harvests skipped, one in June and one in August (2Sk). The four replicates of the treatments were randomized in complete blocks, with overseeding being nested in the other treatments. High mowing height, 1Sk and 2Sk were chosen to try increasing the competitive ability of forage grasses against *S. pumila*, especially during the summer months. The occurring forage grasses were *Lolium multiflorum, Lolium perenne, Dactylis glomerata* and *Poa pratensis* (respectively about 35, 10, 10 and 10% of the biomass in the initial sward). *Poa trivialis* – not a forage grass – was also abundant (35%) in the initial sward. Overseeding was performed yearly with a mixture of *D. glomerata*, *L. perenne*, *P. pratensis* and *Festuca rubra*. The evolution of the botanical composition was appraised by six visual assessments from spring to autumn. We here use the annual mean for the relative abundance of forage grasses and the mean of the August-September assessments for *S. pumila* as a proxy for its yearly peak of biomass. In addition, the abundance of *S. pumila* and the management of 31 grasslands was surveyed during 2017 to 2019. All grasslands were located in the same valley and visited during August-September, but not all during the same week. Because the relative biomass of *S. pumila* evolves quickly during the summer months, its abundance was assessed using a point method (presence/absence at each point) for the survey. We here use the means over years from the survey data. The factorial experiment was analysed by repeated measures ANOVA, the survey by GLMM using Statistica™ 13.5.0.

Results and discussion

In the experiment, the relative abundance of *S. pumila* was similarly high during the first three years (grand mean 20-25% *S. pumila* at the end of summer). Increasing mowing height from 3 to 8 cm decreased *S. pumila* abundance in all harvest interval treatments ($P<0.001$, Figure 1A). Harvest interval also influenced *S. pumila* abundance ($P<0.001$), and the interaction height × interval was significant. The reduction in *S. pumila* abundance achieved with High-1Sk could not be significantly outperformed by skipping a second harvest (Low-2Sk and High-2Sk). In 2020 and 2021, *S. pumila* abundance dropped to only 7 and 3%, respectively, on average over the whole experiment. This was most probably due to the moist weather conditions of these two years. At these low abundances, differences among treatments were not significant.

Mowing height and interval also influenced the competitive ability of the forage grasses as indicated by the significant differences in the trajectory of their abundance over the years (Figure 1B, interaction treatment × year: $P<0.001$). The Low-0Sk treatment was clearly detrimental to the forage grasses. On the contrary, these species considerably increased their relative abundance in all treatments with higher mowing height. Increasing mowing height might be preferred over skipping harvests because this first option does not reduce forage digestibility, while the second does (data not shown). Overseeding significantly increased the relative abundance of *D. glomerata*, but only starting 2020 (data not shown). Thus, it might contribute to an increased resilience against *S. pumila* infestation but only after several years.

Figure 1. (A) Effect of the interval between harvests (0Sk, 1Sk, 2Sk) and of the mowing height (Low/High) on the relative abundance of *Setaria pumila* for the years 2017 to 2019. The box plots show the median, the quartiles and the non-outlier range (n=8). Letters above the x-axis indicate significant differences (Tukey HSD test, $P<0.05$). (B) Evolution of the relative abundance of forage grasses from 2017 to 2021 in the different harvest mowing height × interval treatments. The plots show the means of 4 replicates × 2 overseeding treatments (n=8) and the whiskers the 0.95 confidence intervals.

Among the management parameters recorded during the survey (Table 1), only the mowing height appeared to have a significant effect on *S. pumila* abundance (*P*=0.017). For the survey, this parameter was categorized in the four classes <5, 5-6, 6-7 and >7 cm. Higher *S. pumila* abundance was not associated with higher numbers of fertilizer applications (range = 1 to 7 applications). No effect of overseeding or self-reseeding (one growth cycle harvested after seed maturity of the targeted forage grass species to allow it to shed seeds), summarized as 'Seed input' in Table 1, could be observed with this dataset. The results show that *S. pumila* infestation can affect both permanent and temporary grasslands. From an agricultural extension perspective, the survey was also very useful for discussing careful grassland management with farmers.

Table 1. Generalized linear mixed model summary with *Setaria pumila* abundance in 31 grasslands surveyed on-farm as dependent variable of grassland management factors. Seed input: overseeding and/or self-reseeding. Temporary/permanent grasslands as categorical variable.

Parameters	Estimate	Std. error	Significance
Intercept	0.619	0.202	**
Mowing height	-0.213	0.083	*
Number of fertilizer applications	0.038	0.033	ns
Seed input	0.129	0.137	ns
Temporary/permanent	-0.009	0.070	ns

Conclusions

Caring for the forage grasses with an adapted mowing height and time interval between harvests increases the competitive ability of the good quality grasses and decreases *S. pumila* abundance during years of high *S. pumila* occurrence. Such management options are a mid-term investment and are becoming increasingly important with increasing frequency of adverse weather events.

Acknowledgements

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References

Dekker J. (2003) The foxtail (*Setaria*) species-group. *Weed Science* 51, 641-656.

Landolt E., Bäumler B., Erhardt A., Hegg O., Klötzli F., Lämmler W., … and Wohlgemuth T. (2010) *Flora indicativa. Ecological indicators values and biological attributes of the flora of Switzerland and the Alps*, 2nd ed., Haupt Verlag, Bern, Switzerland, 376 pp.

Orlandi S., Nucera E., Mosimann E., D'Adda G., Garzoli D., Bertosssa M., … and Lombardi G. (2015) Drivers of *Setaria pumila* (Poir.) Roem. et Schult growth and impact on forage quality in lowland Switzerland meadows. *Grass and Forage Science* 72, 154-162.

Operability of agroecological practices: the case of parasite dilution in sheep/cattle mixed-grazing

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Abstract

Sheep liveweight gain (LWG) can be higher under sheep/cattle mixed-grazing than under monospecific grazing. We conducted an experiment to assess the relative role of parasite dilution and complementarity of feeding niches to improved sheep LWG, under mixed-grazing. We compared LWG of ewe lambs grazing alone or with heifers, at two contrasting cattle/sheep ratios (~50/50% and 80/20% in livestock units). We assessed parasitism through strongyle eggs per gram of faeces (EPG) and foraging mechanisms through faecal nitrogen content (N). We observed that: (1) N did not significantly differ across treatments (~2%); (2) EPG was ~50% lower in mixed compared to monospecific treatments (545 and 716 vs 1278, $P<0.001$); and (3) LWG were higher in mixed compared to monospecific treatments (~40 g per day higher, $P<0.001$). Neither EPG nor LWG significantly differed between the two cattle/sheep ratios. We conclude that: (1) improved sheep LWG was mainly due to parasite dilution; and (2) benefitting from this mechanism can be obtained on a broad range of sheep/cattle ratios. As no fine-tuning of the cattle/sheep ratio is required to benefit from mixed-grazing, we consider this practice as operable, i.e. likely to be 'easily' put into operation, from a biological viewpoint.

Keywords: mixed-grazing, agroecology, strongyle, diet selection, diet quality

Introduction

Extending agroecology to livestock farming systems calls for a diversification of feed resources and animal species (Dumont *et al.*, 2013). Mixed-grazing with cattle and sheep is one example of this diversification and it has received significant attention over the last decades (Fraser *et al.*, 2013; Jerrentrup *et al.*, 2020; Mahieu *et al.*, 1997; Marley *et al.*, 2006; Mosnier *et al.*, 2021; Nolan and Connolly, 1989). d'Alexis *et al.* (2014) have reviewed that sheep liveweight gain (LWG) is usually higher under sheep/cattle mixed-grazing than under sheep monospecific grazing. This could result from parasite dilution and/or from complementary use of forage niches between species, but the relative contribution of the two mechanisms remains unclear. In addition, the way sheep/cattle ratio modulates the effects of mixed-grazing is poorly understood. We therefore conducted an experiment to study these two questions.

Materials and methods

We conducted our experiment at the INRAE/Herbipôle experimental facility of Laqueuille, in the uplands of central France (*Massif Central*), in 2019 and 2020. Experimental plots were continuously grazed from May to October by *Romane* ewe lambs of 5-9 months and Holstein heifers of 17-20 months at the beginning of the grazing season (new animals being used each year). We compared three treatments where animals were grazed at the same stocking rate (0.82 LU ha^{-1}): a monospecific sheep grazing treatment and two mixed-grazing treatments, with two replicates each year. The mixed treatments comprised a balanced cattle/sheep group (Mixed-: 55/45% for cattle/sheep in livestock units) and a group markedly skewed towards cattle (Mixed+: 78/22%). We did not add a cattle monospecific treatment, as cattle have been shown to not benefit from mixed-grazing (d'Alexis *et al.*, 2014).

Sheep were not treated against strongyle infection before or during the experiment, and we tested their initial infection level by counting strongyle eggs per gram of faeces (EPG), before they entered experimental plots in May. Animals were balanced for weight and EPG across replicates and treatments. Sheep were then weighed monthly to calculate their LWG, and we monitored their level of strongyle infection through EPG on five 'core' ewe lambs per treatment and replicate. Diet selection by the same five animals was assessed through faecal nitrogen content (N). All heifers that were less numerous were monitored for LWG, EPG and faecal N.

Results and discussion

Overall LWG of sheep + cattle per hectare was significantly higher in mixed compared to monospecific grazing treatments (P<0.001; Table 1). Sheep LWG was higher (~40 g per day higher; P<0.001) in mixed compared to monospecific treatments, while cattle LWG was stable across the two cattle/sheep ratio treatments. Sheep EPG was ~50% lower in mixed compared to monospecific treatments (P<0.001), whereas faecal N content was stable (P=0.427; Table 1). The fact that sheep LWG and EPG differed across treatment, while faecal N content was stable, indicates that the higher sheep growth under mixed-grazing results from parasite dilution, rather than from an improvement in sheep foraging.

Animals in the two mixed-grazing treatments also did not significantly differ in terms of LWG (P=0.960), EPG (P=0.184) and faecal N content (P=0.393). This suggests that the benefits of mixed-grazing would be the same over a wide range of cattle/sheep ratios. As no fine-tuning of cattle/sheep ratio is required to benefit from mixed-grazing, implementing mixed grazing does not seem to be overly complex for farmers. Therefore, based on the fact that mixed-grazing relies on a biological mechanism (parasite dilution), and the definition of the adjective operable ('likely to be 'easily' put into use, operation or practice'), we consider mixed-grazing a biologically operable practice (Joly *et al.*, 2021).

Table 1. Experimental settings and results of animal measurements.

Group type	Mono-specific	Mixed –	Mixed +
Experiment settings			
% sheep in group in livestock units (LU)	100%	45%	22%
Ewe lambs 5-9 months (heads)	20	10	5
Adult ewe + 12 months (heads)	5	2	1
Heifer 17-20 months (heads)	0	2	3
Total livestock number[1] (LU)	2.15	2.2	2.3
Paddock area (ha)	2.63	2.69	2.81
Stocking rate (LU ha^{-1})	0.82	0.82	0.82
Initial liveweight (sheep + cattle) (kg ha^{-1})	437	517	554
Experiment results[3]			
Final liveweight[2] (sheep + cattle) (kg ha^{-1})***	458^a	617^b	650^b
Overall LWG per ha (sheep + cattle) (g day^{-1} ha^{-1})***	124^a	604^b	582^b
Ewe lamb LWG (g day^{-1})***	2.36^a	41.79^b	44.40^b
Ewe lamb strongyle infection (eggs (g faeces)$^{-1}$) ***	1278^a	716^b	545^b
Ewe lamb faecal N content (% faeces mass) (ns)	1.98	1.94	2.01
Cattle LWG (g/day) (ns)		498	440
Cattle strongyle infection (eggs per gram of faeces) (ns)		24	40
Cattle faecal N content (% faeces mass) (ns)		1.92	1.85

[1] Ewe lamb: 0.07 LU; Adult ewe: 0.15 LU; Heifer: 0.6 LU

[2] Includes a virtual ewe lamb to compensate for sudden death of an individual in the Monospecific replicate in 2019.

[3] Significance: * P<0.05, ** P<0.01, *** P<0.001, ns P≥0.05. Columns sharing the same superscripts are not significantly different at P=0.05.

Conclusions

Sheep/cattle mixed-grazing is one application of agroecology principles relevant to livestock farming systems. It benefits sheep LWG and under our experimental conditions (continuous grazing at a moderate stocking rate), parasite dilution was the main mechanism involved. We also observed that similar LWG were obtained under two contrasted cattle/sheep ratios, which suggests that defining a pertinent sheep/cattle ratio is not overly complex. Our results thus illustrate the biological operability of mixed-grazing.

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References

d'Alexis S., Sauvant D. and Boval M. (2014) Mixed grazing systems of sheep and cattle to improve liveweight gain: a quantitative review. *Journal of Agricultural Science* 152, 655-666. https://doi.org/10.1017/S0021859613000622

Dumont B., Fortun-Lamothe L., Jouven M., Thomas M. and Tichit M. (2013) Prospects from agroecology and industrial ecology for animal production in the 21st century. *Animal* 7, 1028-1043. https://doi.org/10.1017/S1751731112002418

Fraser M.D., Vale J.E. and Dhanoa M.S. (2013) Alternative upland grazing systems: Impacts on livestock performance and sward characteristics. *Agriculture, Ecosystems & Environment* 175, 8-20. https://doi.org/10.1016/j.agee.2013.05.002

Jerrentrup J.S., Komainda M., Seither M., Cuchillo-Hilario M., Wrage-Mönnig N. and Isselstein J. (2020) Diverse swards and mixed-grazing of cattle and sheep for improved productivity. *Frontiers in Sustainable Food Systems* 3, 125. https://doi.org/10.3389/fsufs.2019.00125

Joly F., Benoit M., Martin R. and Dumont B. (2021) Biological operability, a new concept based on ergonomics to assess the pertinence of ecosystem services optimization practices. *Ecosystem Services* 50, 101320. https://doi.org/10.1016/j.ecoser.2021.101320

Mahieu M., Aumont G., Michaux Y., Alexandre G., Archimède H., Boval M. and Thériez M. (1997) L'association d'ovins et de bovins sur prairies irriguées en Martinique. *INRA Productions Animales* 10, 55-65.

Marley C.L., Fraser M.D., Davies D.A., Rees M.E., Vale J.E. and Forbes A.B. (2006) The effect of mixed or sequential grazing of cattle and sheep on the faecal egg counts and growth rates of weaned lambs when treated with anthelmintics. *Veterinary Parasitology* 142, 134-141. https://doi.org/10.1016/j.vetpar.2006.06.030

Mosnier C., Benoit M., Minviel J.J., Veysset P. (2021) Does mixing livestock farming enterprises improve farm and product sustainability? *International Journal of Agricultural Sustainability*. https://doi.org/10.1080/14735903.2021.1932150

Nolan T., Connolly J. (1989) Mixed v. mono-grazing by steers and sheep. *Animal Production* 48, 519-533. https://doi.org/10.1017/S0003356100004049

Joint effects of biocontrol herbivory and plant competition greatly reduce the growth of *Rumex obtusifolius*

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Abstract

Augmentative biological control offers a potentially effective, but largely unexplored, opportunity to control native weeds with native phytophagous insects. Herbivore effects on the target weed may further be enhanced by interspecific competition with other plant species. We assessed the impact of root-boring larvae of the Sesiid moth *Pyropteron chrysidiforme* on the target weed *Rumex obtusifolius* for two groups of initially small and large plants, with or without competition from a *Lolium perenne* sward. In a field experiment, 106 *Rumex* roots were planted into plots with either pure *L. perenne* or bare soil, and *R. obtusifolius* plant performance was measured after one year. Overall, competition from the grass sward strongly reduced aboveground biomass and root mass of *R. obtusifolius*. Herbivory alone had little impact on *R. obtusifolius* growth. However, in combination with grass competition, herbivory negatively affected above- and belowground biomass of *R. obtusifolius* plants, but only when growing from initially smaller roots (agent × competition interaction: $P<0.05$ for each). Our results indicate that joint effects between augmentative biological control and plant competition can reduce the growth of a major grassland weed.

Keywords: augmentative biocontrol, *Pyropteron chrysidiforme*, weed control

Introduction

Rumex obtusifolius L. (broad-leaved dock) is one of the most problematic weeds in intensively managed permanent grasslands in Europe (Grossrieder and Keary, 2004) and is considered a major hindrance for conversion to organic farming in Switzerland. There is thus a need to develop effective non-chemical control measures against *R. obtusifolius*. The native European clearwing moth *Pyropteron chrysidiforme* (Esper) (Lepidoptera; Sesiidae) has been proposed as a candidate for augmentative biological control of *R. obtusifolius* (Grossrieder and Keary, 2004) as the root-boring larvae can promote degradation of the plant's storage organ (Scott and Sagliocco, 1991). However, in a previous study the impact of *P. chrysidiforme* was insufficient to significantly reduce the performance of established plants of *R. obtusifolius* in permanent grasslands (Hahn *et al.*, 2016). The effects of herbivory can possibly be enhanced by interspecific plant competition (Sheppard, 1996), and a potential competitor of *R. obtusifolius* is *Lolium perenne* L. (perennial ryegrass) (Keary and Hatcher, 2004; Niggli *et al.*, 1993). In this study, we assessed the impacts of herbivory by *P. chrysidiforme* and competition with *L. perenne* on small and large plants of *R. obtusifolius*.

Materials and methods

A field experiment was set up in June 2019 near Zürich, Switzerland. Field-collected, small and large roots of *R. obtusifolius* (106 roots in total) were planted into established, pure swards of *L. perenne* and plots with bare soil (16 plots in total: dimension 1.8×5 m; 3-4 roots of each size class per plot). The average mass of transplanted small and large roots was 2.9 g (standard error ±0.20 g) and 57.5 (±5.19) g, respectively. One half of the roots from each size class was inoculated with eggs of the biological control agent *P. chrysidiforme*, the other half served as the control with no application (split-plot design). Aboveground biomass of *Rumex* plants was harvested three times in autumn 2019 and twice in spring 2020, dried to constant weight and summed over harvests to obtain the cumulative aboveground biomass.

Roots were excavated in May 2020, washed free of soil and weighed. Data were analysed with generalized linear mixed-effects models (GLMMs) using a log link function. Explanatory factors were competition from *L. perenne* (2 levels), application of *P. chrysidiforme* (2 levels), and initial root mass of *R. obtusifolius* (2 levels), including all interactions. The split-plot structure was accounted for by a random intercept for plot (analyses done with software R, version 4.1.1, R Core Team, 2021).

Results and discussion

The aboveground biomass of all *R. obtusifolius* plants was significantly reduced by competition from *L. perenne* (main effect: χ^2=66.5, P<0.001; Figure 1); yet, plants from initially small roots were more suppressed by competition than plants grown from large roots (competition × init. root mass interaction: χ^2=23.8, P<0.001). While there was no effect of *P. chrysidiforme* application when *R. obtusifolius* plants grew without competition, *P. chrysidiforme* significantly reduced the aboveground biomass of initially small roots under competition from the *L. perenne* sward (z=3.9, P<0.001; compare contrasts Figure 1).

Similar to aboveground biomass, final root mass of all *R. obtusifolius* plants was negatively affected by competition from *L. perenne* (main effect: χ^2=39.3, P<0.001; Figure 2), and the competition effect was stronger for initially small roots (competition × init. root mass interaction: χ^2=20.6, P<0.001). Also, final root mass of initially small roots was significantly impacted by the application of *P. chrysidiforme* only under competition from the *L. perenne* sward (z=4.8, P<0.001; compare contrasts Figure 2B).

Our findings provide evidence that interspecific competition and herbivory cause interactive impacts on the growth of *R. obtusifolius* and that these effects were plant size-dependent. Competition from a grass sward has been shown to reduce resource availability for neighbour plants, thereby affecting their growth

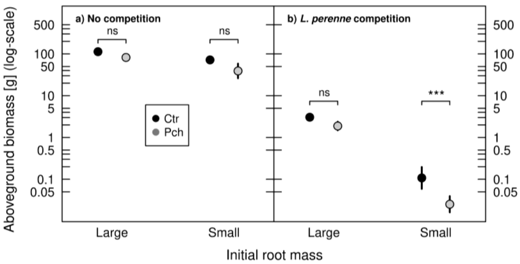

Figure 1. Aboveground biomass of *Rumex obtusifolius* plants grown under no competition (A) and competition from a *L. perenne* sward (B) depending on the initial root mass and *Pyropteron* treatments (no application [Ctr], *P. chrysidiforme* [Pch]). Displayed are means ± standard error. Non-visible standard errors are due to small values. The statistical inference is based on a GLMM. *** P<0.001, ns = not significant.

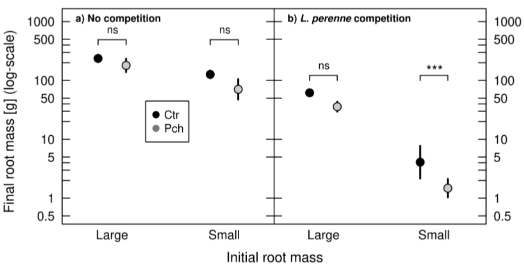

Figure 2. Final root mass of *Rumex obtusifolius* plants grown under no competition (A) and competition from a *L. perenne* sward (B) depending on the initial root mass and *Pyropteron* treatments (no application [Ctr], *P. chrysidiforme* [Pch]). Displayed are means ± standard error. Non-visible standard errors are due to small values. The statistical inference is based on a GLMM. *** P<0.001, ns = not significant.

(Jeangros and Nösberger, 1990). Yet, Niggli *et al.* (1993) have demonstrated a high regrowth potential of *R. obtusifolius* after cutting, even when grown in competition with *L. perenne* and other grass swards.

The high competitive ability of *R. obtusifolius* was attributed to its efficient use of nitrogen and its carbohydrate reserves (Niggli *et al.*, 1993). This finding may explain why in our study small *Rumex* roots had a lower potential to resist competition, as their reserves are small. The herbivory effect, although generally weaker than the competition effect, further suppressed initially small, but not the larger roots when subjected to competition from *L. perenne*.

Conclusions

Combining augmentative biological control and plant competition can reduce the growth of *R. obtusifolius* below its single effects, yet only for small plants. Such combined effects should more often be explored in integrated weed management.

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References

Grossrieder M. and Keary I.P. (2004) The potential for the biological control of *Rumex obtusifolius* and *Rumex crispus* using insects in organic farming, with particular reference to Switzerland. *Biocontrol News and Information* 25, 65N-79N.

Hahn M.A., Schaffner U., Häfliger P. and Lüscher A. (2016) Establishment and early impact of the native biological control candidate *Pyropteron chrysidiforme* on the native weed *Rumex obtusifolius* in Europe. *Biocontrol* 61, 221-232.

Jeangros B. and Nösberger J. (1990) Effects of an established sward of *Lolium perenne* L. on the growth and development of *Rumex obtusifolius* L. seedlings. *Grass and Forage Science* 45, 1-7.

Keary I.P. and Hatcher P.E. (2004) Combining competition from *Lolium perenne* and an insect-fungus combination to control *Rumex obtusifolius* seedlings. *Weed Research* 44, 33-41.

Niggli U., Nösberger J. and Lehmann J. (1993) Effects of nitrogen-fertilization and cutting frequency on the competitive ability and the regrowth capacity of *Rumex-obtusifolius* L in several grass swards. *Weed Research* 33, 131-137.

R Core Team (2021) R: A language and environment for statistical computing. Vienna, Austria https://www.r-project.org.

Scott J.K. and Sagliocco J.L. (1991) Host-specificity of a root borer, *Bembecia-chrysidiformis* [*Lep*, *Sesiidae*], a potential control agent for *Rumex* spp [*Polygonaceae*] in Australia. *Entomophaga* 36, 235-244.

Sheppard A.W. (1996) The interaction between natural enemies and interspecific plant competition in the control of invasive pasture weeds. In: *Proceedings of the IX International symposium on Biological Control of Weeds*. Stellenbosch, University of Cape Town, South Africa, pp. l-26.

Spatial variation in vegetation height as an indicator of aboveground carbon stocks in grazed grasslands

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Abstract

Spatial heterogeneity in plant and soil properties is known to influence ecosystem functions, but the linkages between spatial variation and ecosystem services in grasslands are unclear. Here we examine within-field variation in sward structure (vegetation height) and test whether indices of spatial heterogeneity can be used as a simple indicator of aboveground production services or carbon stocks over time in upland grasslands. Two upland pastures with continuous cattle grazing (high versus low stocking rate and N inputs) were studied over a six-year period using a spatially explicit sampling scheme, as part of a long-term field management trial in central France. We found that grassland height showed significant spatial and temporal variation during the study period in both grazing management treatments. Contrary to expectations, stocking rate did not affect the degree of within-field variation in vegetation height recorded during the study. Mean annual biomass in both grazed paddocks showed a positive relationship with variation in end-of-season vegetation height, but was unrelated to vegetation properties at the start of the growing season. Simple metrics of height variation appear to be a useful proxy of grassland standing biomass, and hence carbon stocks, at broader spatial and temporal scales, and could improve the prediction of field-scale function in a changing environment.

Keywords: biomass production, vegetation height, upland grassland, management

Introduction

Within-field variability and spatial patterns in plant and soil properties reflect the interplay between geology, topography and biotic processes, both past and present. Studies at small spatial scales suggest that spatial heterogeneity plays a key role for ecosystem functions (i.e. biomass production, biogeochemical cycling, nutrient losses), with significant implications for the provision of ecosystem services in managed systems (Bloor and Pottier, 2014). This is of particular relevance for grazed grasslands where large herbivores promote within-field variation in plant biomass and soil nutrients due to their grazing activities (non-uniform activities of defoliation, animal returns). Indeed, grazed paddocks encompass a range of grazed/ungrazed patches of differing sward height, and areas affected by animal returns and trampling. Impacts of grazing herbivores on sward structure may vary depending on animal species and stocking rate, fertilizer inputs and vegetation composition, which modify patterns of defoliation and subsequent plant growth. However, the influence of spatial heterogeneity on field-scale processes is poorly understood, and the description of within-field variation in vegetation and soil resources in relation to grazing management remains limited (Bloor *et al.*, 2020). Improved understanding of the linkages between within-field variation and field-scale ecosystem functions is required to establish whether the enhancement (or reduction) of spatial heterogeneity should be considered in the elaboration of management strategies (Dronova, 2017), and could have significant implications for biodiversity and sustainable agriculture. In order to address this issue, we examined within-field variation in vegetation structure (height of green and senescent biomass) and tested whether indices of spatial variation could be used as a simple indicator of production services over time in upland grasslands.

Materials and methods

Measurements were carried out over a six-year period (2016-2021) at the long-term French research platform (SOERE-ACBB, ICOS: Laqueuille; 45°64″ N, 02°73″ E; 1,100 m a.s.l.). The site is characterized by Andosol soil, with mean annual temperature of 8 °C and annual rainfall of 1000 mm. The field trial was established in 2002, and consists of two paddocks managed with continuous cattle grazing from May to October; the 'Intensive' management treatment is a 5.4 ha field with moderate stocking rate (1.1 LSU ha^{-1} y^{-1}) and N inputs (200 kg ha^{-1} y^{-1}), whereas the 'Extensive' treatment is a 3.4 ha field with low stocking rate (0.5 LSU ha^{-1} y^{-1}) and no N inputs. Both paddocks are dominated by grasses; species such as *Dactylis glomerata*, *Poa pratensis* and *Agrostis capillaris* are common across treatments, but *Trifolium repens* is present only in the 'Intensive' treatment, and forb species are more abundant in the 'Extensive' treatment (Klumpp *et al.*, 2011). Field-scale standing biomass during the growing season is assessed every year by harvesting aboveground samples at five dates at monthly intervals across each paddock. Potential productivity is assessed by grazing exclusion cages, and sampled at the same intervals as standing biomass. At each cut, biomass is sampled at a height of 5 cm above soil surface (70×70 cm quadrats), oven-dried (60 °C for 48 h) and weighed. Within-field variation was examined from 2016 onwards using a 30×20 m grid in the centre of each field (117 points per grid, regular 2.5 m distances). Permanent corner markers were installed in May 2016, and geographic coordinates of each sample point were established (precision 5 cm; Trimble R8 GPS Systems, Trimble Navigation Limited, USA) in order to repeatedly measure the same points at each subsequent sampling campaign. Maximum heights of both green and senescent shoots were determined using a sward stick at each grid point at the end of the grazing season each year (October – November depending on weather conditions and snow). This sampling period was chosen in order to capture the maximum effect of grazing on vegetation structure; effects of grazing on vegetation structure are known to become progressively more marked over the course of the plant growing-season (Rossignol *et al.*, 2011). Standard deviation (SD) and coefficients of variation (CV, standard deviation/ mean × 100) were used to assess the absolute and relative variation, respectively, of plant height within each treatment. Relationships between field-level biomass productivity and within-field variation were assessed using GLM models.

Results and discussion

During the study, mean annual productivity was greater in the 'Intensive' treatment than the 'Extensive' grazing treatment (mean of 7.2 and 3.3 Mg ha^{-1} for Intensive and Extensive treatments respectively; $F_{1,11}$=54.5, P<0.001; Figure 1A). This result is consistent with previous observations at this site (Klumpp *et al.*, 2011), and reflects higher N inputs in the Intensive treatment and increased nutrient cycling under high stocking rates (Bardgett and Wardle, 2003). In contrast, standing biomass ranged from 0.2 to 1.3 Mg ha^{-1} and showed no significant difference between grazing treatments (P>0.05, Figure 1B). We found there was a high degree of within-site variation for height measured for both green and senescent shoots irrespective of grazing treatment, and both absolute variation (SD) and relative variation (CV) showed interannual variation. Absolute variation in green vegetation height at the end of the growing season (green height SD) showed a positive relationship with mean annual field-scale standing biomass in both grazing treatments (R^2=0.65, P<0.001 across treatments), and showed some discrimination between treatments (Figure 1C). Green height SD was more closely linked to mean standing biomass than was mean vegetation height across years (mean green height range: 3.3-11.8 cm; R^2=0.46, P<0.001 across treatments). Neither mean green vegetation height nor any metrics of variation in green height were related to field-level productivity. Moreover, no significant relationship was found between metrics of variation in senescent height and either field-level productivity or standing biomass (P>0.05). Our results suggest that within-field variation in end-of-season green vegetation height may be an integrated indicator of biomass state during the year (i.e. quantity of standing biomass available for ingestion over time), with implications for the estimation of aboveground carbon stocks and carbon input into the soil. In contrast, these simple metrics of within-field variation do not appear to provide useful proxies of biomass fluxes, and hence production services, in the grazed grassland systems studied here.

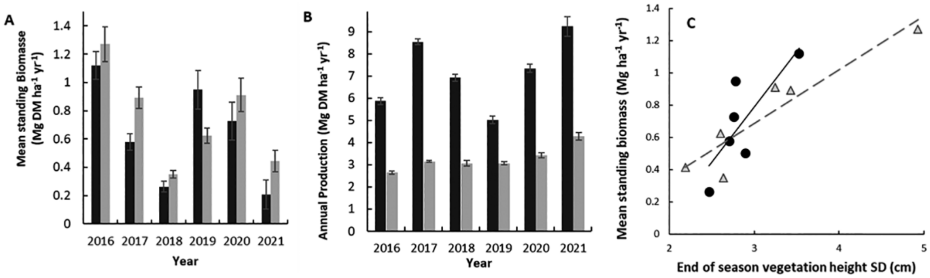

Figure 1. Interannual variation in field-scale annual biomass production (A) and mean standing biomass (B); data are means ± standard error. Graph C shows relationship between within-field variation in green vegetation height (standard deviation) and estimations of mean annual field-scale standing biomass during the study. Grazing treatments are given by: Intensive, black-filled bars/circles; Extensive, grey-filled bars/triangles.

Conclusions

Metrics of within-field variation in vegetation height at the end of the growing season were more closely linked to mean annual standing biomass than grassland productivity, and showed some evidence of treatment-induced changes in ecosystem function. These results suggest that simple measurements of grassland heterogeneity could be useful for model validations and the prediction of field-scale carbon stocks in a changing environment.

References

Bardgett R.D. and Wardle D.A. (2003) Herbivore-mediated linkages between aboveground and belowground communities. *Ecology* 84, 2258-2268.

Bloor J.M.G. and Pottier J. (2014) Grazing and spatial heterogeneity: Implications for grassland structure and function. In: Mariotte, P. and Kardol, P. (eds.) *Grassland Biodiversity and Conservation in a Changing World*; Nova Science Publishers, Inc., Hauppage, NY, USA.

Bloor,J.M.G., Tardif A. and Pottier J. (2020) Spatial heterogeneity of vegetation structure, plant N pools and soil N content in relation to grassland management. *Agronomy* 10, 716. https://doi.org/10.3390/agronomy10050716

Dronova I. (2017) Environmental heterogeneity as a bridge between ecosystem service and visual quality objectives in management, planning and design. *Landscape and Urban Planning* 163, 90-106.

Klumpp K., Tallec T., Guix N. and Soussana J-F. (2011) Long-term impacts of agricultural practices and climatic variability on carbon storage in a permanent pasture. *Global Change Biology* 17, 3534-3545.

Rossignol N., Chadoeuf J., Carrère P. and Dumont B. (2011) A hierarchical model for analysing the stability of vegetation patterns created by grazing in temperate pastures. *Applied Vegetation Science* 14, 189-199.

Adaption of selected grasses towards micro-environmental conditions under long-term extensive grazing

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Abstract

Under long-term extensive grazing, patches of different heights evolve as a consequence of preference for young and leafy vegetation (short <10 cm height) and avoidance of areas where the vegetation is mature and stemmy (tall >10 cm height). Among these patches the same plant species may survive site-by-site despite the divergent growing conditions of frequent defoliation against infrequent defoliation. In the present study, we investigated the question of whether the grass species show adaption to long-term environmental conditions that remain intact when plants are transferred to uniform conditions. Focus was given to the influence on resource partitioning above- and belowground in a transfer approach with the grasses *Dactylis glomerata* and *Festuca rubra* obtained from replicated paddocks of two ongoing long-term grazing experiments ('Forbioben' in Central Germany and the 'Oldřichov Grazing Experiment' located in the Jizerské hory in Czech Republic). While the patch-origin had an effect on the aboveground biomass and root-to-shoot ratio for the grasses from the Oldřichov experiment, no such significant effect was found at the Forbioben experiment.

Keywords: low-input grassland, patch grazing, root production

Introduction

The regularly recurring defoliation of frequently grazed short patches in extensive grassland creates a microhabitat that differs in light and nutrient availability from neighbouring tall patches that are hardly defoliated, so that patch-specific plant species can survive (Dumont *et al.*, 2012). While focus is usually given to the contrast in vegetation composition between patches, several plant species endure in either patch type, of which *Dactylis glomerata* and *Festuca rubra* are frequently found despite the contrasting environmental conditions between patches, pointing at large intra-specific adaption. The selective export of nutrients from short patches over many years and the random distribution of these nutrients via dung and urine, leads to a relative shift of plant available nutrients in the soil towards tall patches (Tonn *et al.*, 2019). Plants surviving in short patches are, consequently, adapted to less N availability and regular defoliation while those inhabiting tall patches endure under higher soil N levels without defoliation. The rationale underlying this study was, consequently, that the productivity of either species under low or high defoliation intensity depends on the patch of origin because of, so far, unrecognized adaption. We hypothesized that plants originating from short patches would be more productive under frequent than under infrequent defoliation intensity, while the opposite would be true for plants from tall patches because conditions in the patches had selected for genetically different subpopulations. We further expected plants from short patches to allocate a greater biomass proportion belowground than plants from tall patches.

Materials and methods

To test the hypothesis, one grazing treatment each on the research platform 'Forbioben' in the Solling Uplands, Germany (51°46'50' N; 9°41'55' E) and the 'Oldřichov Grazing Experiment' in the Jizerské hory in the Czech Republic (50°50'34' N; 15°05'36' E) was sampled in replicated paddocks in spring 2019. The Oldřichov experiment consists of two field replicates (for details see Pavlu *et al.*, 2021) while three field replicates are available in the Forbioben experiment (Tonn *et al.*, 2019). Single plants of *F. rubra* and *D. glomerata* were collected from pairs of visually identified tall and short patches. Patches within a pair were selected from areas as close to each other as possible. From each patch within a field replicate, two plants (fully developed tussocks) of each species were collected (two on tall and two on short patches) in Forbioben and three base plants of each species and patch within field replicate at the Oldřichov site. In total two and three pairs were sampled per paddock in Forbioben and Oldřichov, respectively resulting in 24 plants in total per site (n = six plants per species × patch origin). After excavation, plants for experimentation were established by transplanting of single tillers from the plant population approximately 14 d after the removal off the field. For this, roots and aboveground biomass were cut at a level of 4 to 6 cm, planted in compost soil-filled multi-pot plates and watered regularly. Four well-grown tillers of each plant were selected after a 14-d growth period and each tiller was then transplanted into black plastic pots (11 cm diameter), containing a mixture of sand and compost soil (9:1-ratio) and each tiller was assigned to one of four experimental treatments. Experimental treatments consisted of nitrogen (N) fertilization (referring to equivalent masses of 0 vs 240 kg N ha^{-1}) and defoliation frequency (frequent vs undefoliated) to imitate environmental conditions common to tall and short patches. Pots were arranged in a full randomized design (Göttingen) and a randomized complete block design (Oldřichov) and placed outdoors under natural climatic conditions with n = six replicates of each origin × plant species × nutrient level × defoliation treatment combination for an experimental duration of 8 weeks. The N fertilization was applied weekly in equal doses as ammonium nitrate dissolved in water. Unfertilized plants received equal amounts of water at each dressing and pots were generally watered regularly. Frequently defoliated plants were cut manually in fortnightly intervals at 4 cm plant height while undefoliated plants were harvested once after 8 weeks. Accumulated standing belowground biomass was assessed after 8 weeks by washing roots accumulated within pots free from soil in an automatic elutriation system and manual sorting in floatation. All biomass samples were dried in a forced air oven until constant weight to obtain accumulated above- and belowground biomass (AGB, BGB) and to calculate the root to shoot ratio (R/S ratio). Statistical analyses were performed with the R software (4.0.1, R Core Team, 2020) using linear-mixed effects modelling for each experiment separately. Accumulated AGB, BGB and R/S ratios were analysed with the following main and interactive effects of patch origin, plant species and treatment (combination of N × defoliation) as fixed effects. The plot served as a random effect for the Forbioben experiment and the block for the Oldřichov experiment. Tukey-HSD tests were followed posthoc to analyse differences between means for significant influencing variables.

Results and discussion

Patch origin had no effect in the Forbioben plant population where, however, the main effects of species (AGB: $P<0.001$; BGB: $P<0.001$, R/S ratio: $P<0.01$) and treatment (AGB: $P<0.001$; BGB: $P<0.001$, R/S ratio: $P<0.001$) were significant (Table 1). *Dactylis glomerata* was more productive than *F. rubra* above- and belowground and, in general, N fertilization under infrequent defoliation clearly increased AGB. Within defoliation system, N fertilizer reduced BGB and the treatment defoliated once at the end promoted BGB. In the absence of N fertilizer, consequently, greater proportions of biomass were allocated to roots and, generally, less under frequent defoliation. Similar to Forbioben, *D. glomerata* at Oldřichov was more productive than *F. rubra* (Table 1) although the differences between species depended on the treatment as given by the significant species × treatment interaction (AGB: $P<0.001$; BGB: $P<0.05$, R/S ratio: $P<0.001$) (Table 1). The treatment defoliated at the end and N fertilizer increased both AGB and

BGB with a greater root allocation in frequently defoliated and unfertilized treatments (Table 1). Patch origin had a significant effect on AGB (P<0.01) and R/S ratio (P<0.05). Plants from short patches were significantly more productive aboveground with a difference of 29% across treatments but allocated a smaller proportion to the roots (R/S ratio tall vs short). Short patches can be expected to have fewer resources available to invest in root production as the constant defoliation requires continuous regrowth several times per year (Guitian and Bardgett, 2000).

Table 1. Model estimates ± standard error of the above- and belowground biomass (AGB, BGB, g DM pot^{-1}) and the root-to-shoot ratio (R/S ratio) accumulated over the 8-week study period in relation to the main effects of species and treatment (Forbioben) or their interaction (Oldřichov).[1]

Site	Treatment		AGB	BGB	R/S ratio
Forbioben	freqN0		0.22±0.02 a	0.23±0.05 b	1.0±0.3 d
	freqN240		0.29±0.07 a	0.07±0.02 a	0.2±0.04 b
	undefN0		1.4±0.2 b	0.59±0.1 c	0.39±0.04 c
	undefN240		4.1±0.7 c	0.29±0.04 b	0.07±0.01 a
	D. glomerata		1.1±0.1 b	0.4±0.05 b	0.34±0.03 b
	F. rubra		0.6±0.07 a	0.1±0.02 a	0.23±0.03 a
Oldřichov	*D. glomerata*	freqN0	0.03±0.01 aB	1.1±0.1 aB	31.9±5.0 cB
		freqN240	0.12±0.02 bB	0.97±0.1 aB	8.7±1.9 bA
		undefN0	0.62±0.1 cA	4.0±0.3 bB	6.4±0.8 bB
		undefN240	3.3±0.6 dB	2.9±0.5 bB	0.8±0.1 aA
	F. rubra	freqN0	0.02±0.004 aA	0.18±0.04 aA	9.6±1.6 cA
		freqN240	0.009±0.002 aA	0.06±0.02 aA	6.3±2.2 bcA
		undefN0	0.38±0.1 bA	1.1±0.1 cA	2.5±0.3 bA
		undefN240	0.55±0.1 bA	0.4±0.1 bA	0.9±0.1 aA

[1] Lowercase letters denote significant differences between treatments or species (Forbioben) or between treatments within species (Oldřichov). Capital letters indicate significant differences between species within treatment (P<0.05) (Oldřichov). Freq/undef refer to the defoliation frequency; N0 and N240 represent the N fertilizer doses applied in total.

Conclusions

Grasses under frequent defoliation were less productive but only those originating from short patches in the Oldřichov were more productive across treatments which partly confirms our hypothesis. Further research is needed to determine whether or not the different response of patch origin between experimental sites was caused by the four-year longer duration of the Oldřichov experiment or by plant functional traits.

References

Dumont B., Rossignol N., Coucogaray G., Carrère P., Chadoeuf J., Fleurance G., Bonis A., Farruggia A., Gaucherand S., Ginane C., Louault F., Marion B., Mesléard F. and Yavercovski N. (2012) When does grazing generate stable vegetation patterns in temperate pastures? *Agriculture, Ecosystems and Environment* 153, 50-56.

Guitian R. and Bardgett R.D. (2000) Plant and soil microbial responses to defoliation in temperate semi-natural grassland. *Plant Soil* 220, 271-277.

Pavlů K., Kassahun T., Pavlů V.V., Pavlů L., Blažek P. and Homolka P. (2021) The effects of first defoliation and previous management intensity on forage quality of a semi-natural species-rich grassland. *Plos One* 16(3): e0248804

Tonn B., Densing E.M., Gabler J. and Isselstein J. (2019) Grazing-induced patchiness, not grazing intensity, drives plant diversity in European low-input pastures. *Journal of Applied Ecology* 56, 1624-1636.

Biomass, soil profile and C concentration of timothy (*Phleum pratense*) and tall fescue (*Lolium arundinaceum*) roots

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Abstract

Perennial grasses are considered to have an abundant and deep root system and therefore high potential to sequester carbon (C) into the soil. However, empirical evidence of increased C inputs is scarce for intensively managed short-term grasslands. The aim of this study was to increase knowledge of root systems of timothy and tall fescue in terms of potential for C sequestration. A field experiment was established on mineral soil in central Finland in 2019. It was managed intensively and harvested for silage three times per season, cut at silage growth stage. Root samples were taken after each harvest in 2020. Each profile was divided into four layers (0-2, 2-10, 10-20, 20-40 cm). Biomass, C and N concentration and chemical quality of C (based on WEAN-fractions) were analysed from each profile. Root biomass of tall fescue was greater (means 5,160, 5,960, 6,360 kg dry matter (DM) ha^{-1}) than that of timothy (means 3,990, 4,360, 4,620 kg DM ha^{-1}) in all three harvests (standard error of mean: 289, 194 and 182, respectively). The difference in root biomass between species increased during the summer. Root profile, C-content and C fractions are discussed. Results indicate that the C input to the soil by tall fescue is superior to that from timothy.

Keywords: root, carbon, grass, *Festuca arundinacea*, minirhizotron

Introduction

The greatest potential to increase soil carbon (C) storage in arable land has been found to be in perennial crops with abundant below ground biomass (BM) production (Rasse *et al.*, 2005; Kätterer *et al.*, 2011). In Nordic grasses the formation of root biomass and its chemical composition are poorly understood (Palosuo *et al.*, 2016). The main aim of this study was to increase our knowledge of C input by Nordic perennial grass species used for forage production by investigating the quantity and the chemical composition (in terms of C, N and WEAN fractions) of root BM.

Material and methods

The field experiment was established in 2019 at the Natural Resources Institute Finland (Luke) at Maaninka (63°09'N, 27°20'E). The experiment was sown on an agricultural field with sandy loam soil (1.7% of organic matter in the 0-20 cm soil layer). The experiment was carried out as a randomized complete block design with four replicates. The treatment was species: timothy (*Phleum pratense* L. cv 'Nuutti'; T) and tall fescue (*Lolium arundinaceum* (Schreb.) Darbysh; TF). Plots were harvested three times per year and fertilized according to the Finnish recommendations. Total N (NH_4^+ + NO_3^-) fertilization was 100, 90 and 50 kg N ha^{-1} for cuts 1, 2 and 3. Root biomass was assessed by soil coring to a 40 cm depth at each cut. Roots were separated from the soil with a hydropneumatic elutriation system (Smucker *et al.*, 1982) and by hand picking. Separated roots were dried at 50 °C until constant weight and analysed for total dry matter (DM), N and C concentration by dry combustion (Leco® CHN 900 or TruMac® CN analyser). To estimate carbon stability, WEAN-fraction (water, ethanol and acid soluble and insoluble fractions) of C were analysed using a method described in Heikkinen *et al.* (2021). Statistical analyses were calculated using ANOVA (SAS 9.4, Mixed-procedure). First, cuts were calculated separately, treatment was considered as a fixed and replicate as a random effect. Secondly, the cut was considered as a repeated effect and treatment, cut and treatment x cut interaction were fixed factors and replicate and replicate x cut interaction were considered as a random effect.

Results and discussion

Root BM in the 0-40 cm soil profile of TF increased from the 1st cut (5,160 kg DM ha^{-1}, standard error of the mean (SEM) 289) to the 2nd (5,961 kg DM ha^{-1}, SEM 194, $P<0.10$) and 3rd cuts (6,357 kg DM ha^{-1}, SEM 183, $P<0.05$; Figure 1). The increase was observed to be significant in the profile of 2-10 cm ($P<0.05$) and showed a tendency in 20-40 cm ($P<0.10$) from 1st cut to the 2nd and 3rd cut (Figure 1, Table 1). The total root BM of T was approximately the same in each cut ($P>0.05$). TF had approx. 1,500 kg DM ha^{-1} more ($P<0.05$) total root BM in each cut than T (Figure 1, Table 1). The difference increased from 1st cut to the 2nd and 3rd cuts. Root BM differed between species in the 2nd and 3rd ($P<0.05$) cut and was not significant in 1st cut ($P<0.10$). In 1st cut the higher root BM of TF compared to T was detected in the 0-2 cm ($P<0.05$) and 2-10 ($P<0.10$) profiles. In the 2nd and 3rd cuts differences were detected in all profiles except 0-2 cm in the third cut. Results indicated that TF develops root BM through the growing season, especially in soil profile below 2 cm and up to 40 cm depth, the deepest profile measured. In contrast T seems to develop roots near the soil surface. In estimating the total root BM production and C input to the soil during the growing season the turnover rate (TR) of roots should be determined. TR (consisting of root exudates and dead roots) has been estimated to be 65-100% of root BM (Kätterer *et al.*, 2011) and to vary across species (Kagiva *et al.*, 2019). The TR of T and TF is not known. TR and root profile up 80 cm depth will be analysed later using the minirhizotron images collected from the study site.

The concentration of root C of T and TF was 41 mg g^{-1} (SEM 0.6), N concentration was 1.1 mg g^{-1} (SEM 0.03) and C/N ratio 37.6 (SEM 1.21) with no difference between species. N concentration varied between cuts ($P<0.05$), highest in the 2nd cut and lowest in the 3rd, which was reflected to the C/N ratio (Table 1). Lower N concentration can be explained by the higher proportion of dead roots or lower N availability. Root C content was higher than the 33.3% reported for Italian ryegrass in Heikkinen *et al.* (2021).

C fractions (WEAN) of root BM are important for modelling C stability and changes in soil C content (Heikkinen *et al.*, 2021). In general there was higher proportion of E and N in TF than in T ($P<0.05$) (Figure 1). The lower proportion of W was measured on T ($P<0.10$) in the 1st cut but on TF ($P<0.05$) in the 2nd cut. In the 3rd cut the proportion of W was significantly higher than in 1st or 2nd cut. There were small differences in the proportion of WEAN-fractions compared to the results for Italian ryegrass reported Heikkinen *et al.* (2021).

Conclusions

In general, total root BM of TF in each measured soil profile was higher for TF than T, and TF seemed to develop roots through the growing season. Root BM of T increased only in the top layer. Due to varying proportion of root C-fractions the decomposition rate of roots can be different between T and TF. Turnover rate of root DM will be included in final results which will improve the estimation of C input to the soil. The results are important for estimating the C input and changes in soil C stock under Nordic short-term grassland.

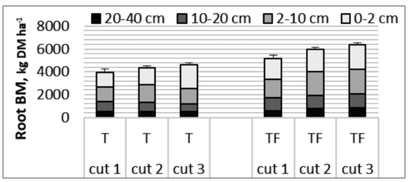

Figure 1. Root dry matter biomass (BM, kg DM ha^{-1}) of timothy (T) and tall fescue (TF) in profiles 0-2 cm, 2-10 cm, 10-20 cm and 20-40 cm in cut 1, 2 and 3. Error bars represent standard error of means of total root BM.

Table 1. Effect of species (tr; T: timothy, TF: tall fescue) on root biomass (RM) in soil profiles 0-2 cm (RM_1), 2-10 cm (RM_2), 10-20 cm (RM_3) and 20-40 cm (RM_4) and carbon (C) and nitrogen (N) concentration (mg/g) of root biomass and the proportion (%) of water (W), ethanol (E) and acid (A) soluble carbon and insoluble ($_i$N) carbon of root C.[1]

	First cut				Second cut				Third cut				*P*-value		
	T	TF	*P*-value	SEM	T	TF	*P*-value	SEM	T	TF	*P*-value	SEM	tr	Cut	tr×cut
RM_1	1,294	1,801	*	120	1,497	1,948	*	156	2,076	2,126	ns	123	**	*	ns
RM_2	1,323	1,642	*o*	121	1,558	2,090	**	87.2	1,359	2,143	*	114	***	o	ns
RM_3	881	1,145	ns	101	812	1,119	*	56.6	696	1,261	***	26.9	***	ns	ns
RM_4	490	572	ns	52.4	497	805	*	62.1	487	827	**	32.8	**	o	o
W	4.2	5.1	o	0.22	4.5	5.9	*	0.29	10.0	10.0	ns	0.32	**	***	o
E	1.6	2.5	**	0.09	1.4	2.2	***	0.09	1.8	2.3	ns	0.24	***	**	**
A	74.9	70.8	**	0.45	73.0	68.3	**	0.34	68.4	65.1	**	0.52	***	***	**
$_i$N	19.3	21.7	*	0.55	21.1	23.6	*	0.48	19.7	22.5	*	0.43	***	*	ns
C	42.1	40.6	ns	0.51	41.4	40.2	ns	0.63	40.6	40.6	ns	0.52	ns	ns	ns
N	1.1	1.1	ns	0.03	1.2	1.2	ns	0.03	1.0	1.0	ns	0.02	ns	*	ns
C/N	39.1	36.2	ns	1.54	36.0	34.8	ns	1.21	39.1	40.7	ns	0.72	ns	*	ns

[1] Significance: ***$P<0.001$, **$P<0.01$, *$P<0.05$, o $P<0.10$, ns $P\geq0.10$; SEM = standard error of mean; tr = treatment.

References

Heikkinen J., Ketoja E., Seppänen L., Luostarinen S., Fritze H., Pennanen T., Peltoniemi K., Velmala S., Hanajik P. and Regina K. (2021) Chemical composition controls the decomposition of organic amendments and influences the microbial community structure in agricultural soils. *Carbon Management* 12(4), 359-376.

Kagiya N., Reinsch T., Taube F., Salminen J-P., Kluβ C., Hasler M. and Malisch C. (2019) Turnover rates of roots vary considerably across temperate forage species. *Soil Biology and Biochemistry* 139, 107614.

Kätterer T, Bolinder M., Andrén O., Kirchmann H. and Menichetti L. (2011) Roots contribute more to refractory soil organic matter than above-ground crop residues, as revealed by a long-term field experiment. *Agriculture, Ecosystems & Environment* 14, 184-192.

Palosuo T., Heikkinen J. and Regina K. (2016) Method for estimating soil carbon stock changes in Finnish mineral cropland and grassland soils. *Carbon Management* 6(5-6), 207-220.

Rasse D.P., Rumpel C. and Dignac M-F. (2005) Is soil carbon mostly root carbon? Mechanisms for a specific stabilization. *Plant and Soil* 269, 341-356.

Assessment of grassland sensitivity to drought in the Massif Central region using remote sensing

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Abstract

Drought is a natural phenomenon that is expected to increase in frequency and duration with climate change, leading to more intense disturbance of ecosystems like grasslands. Moreover, sensitivity of grasslands to drought is expected to differ across landscapes. To assess and explain such variation in sensitivity, knowledge drawn from several grasslands and local parameters must be considered. Our study focused on 143 permanent grasslands in the Massif Central Region of Metropolitan France. With the use of satellite remote sensing, we quantified the relationship between vegetation index anomalies and a modified version of the Standardized Precipitation-Evapotranspiration Index (mSPEI) to acquire grassland sensitivity values. These anomalies provide estimates of the historical long-term fluctuations of grassland vegetation reflectance to climatic water balance between 1985 and 2019. A model selection procedure was used to determine whether the derived sensitivities can be attributed to explanatory variables such as vegetation diversity, pedoclimatic conditions, or management practices. Then a variance partitioning of the included explanatory variables was performed. Our results highlight that soil available water capacity, time of first use, and plant functional diversity all had key influences on the sensitivity of grasslands to drought within those parcels selected in the region.

Keywords: drought, mSPEI, grassland response, sensitivity, remote sensing, vegetation index

Introduction

The Intergovernmental Panel for Climate Change has projected an overall increasing trend in the global temperature due to climate change, together with more frequent and longer extreme climatic events (IPCC, 2021). Among these events, droughts have large-scale impacts on ecosystems like grasslands. However, the grassland responses or sensitivities to drought are expected to vary across landscapes depending on grassland local properties. A better understanding of grassland sensitivity to drought, over landscapes outside of controlled experiments, may help promote agricultural practices supporting grassland stability. To this end, remote-sensing technologies offer new opportunities for fine-resolution monitoring of grasslands (Reinermann *et al.*, 2020). This study aims to assess the variability of grassland sensitivity to drought over the Massif Central using remotely sensed vegetation dynamics, and highlights key drivers of sensitivity, including pedoclimate, biodiversity, and agricultural management.

Materials and methods

We analysed the sensitivity to drought of 394 plots from 143 permanent grassland parcels distributed over the Massif Central (AOP field surveys, 2008-2019) using satellite-based remote sensing. To do so, we first mapped the severity of drought events from 1985 to 2019, with a modified version of the Standardized Precipitation-Evapotranspiration Index (mSPEI; Beguerıa *et al.*, 2014) computed from the daily weather data provided by the *Système d'Analyse Fournissant des Renseignements Atmosphériques à la Neige* (SAFRAN) meteorological data of France. Then, we derived standardized vegetation index (VI) anomalies over the same period. These anomalies are values of the departure of VI from their long-term daily mean. The VI time-series were calculated from Landsat images from 1985 to 2019. Finally, we quantified grassland sensitivity to drought, during the growing season (March to November), as the regression slope between the standardized VI anomalies and mSPEI (Ji and Peters, 2003). From 25 computed VIs and based on a Principal Component Analysis (PCA), we selected the Normalized Multi-band Drought Index (NMDI), which was developed for monitoring soil and vegetation moisture (Wang and Qu 2007). To better understand the drivers of grassland sensitivity to drought, we used a statistical model selection procedure with drought sensitivity as the response variable. The explanatory variables, derived from the field surveys and the Copernicus Land Monitoring services' high-resolution digital elevation model, pertain to three categories: (1) management practices; (2) vegetation diversity, denoted by the taxonomic and functional indices plus the community weighted mean of traits related to growth, phenology, and reproduction; and (3) pedoclimatic conditions, depicted by soil physical and chemical properties (including soil available water capacity or AWC), terrain wetness index and aspect. Lastly, we performed a variance partitioning of the model explanatory variables to quantify their relative influences.

Results and discussion

In our study we found a large variation in satellite-sensed drought sensitivities across the Massif Central grasslands (Figure 1) with 35.62% coefficient of variation. The model selection procedure led to a final sensitivity model with seven explanatory variables and an R-squared of 0.52 (Table 1). According to the variance partitioning (Figure 2), pedoclimatic factors explained the largest part (35%) of variation. From these factors, AWC had a strong negative effect to drought sensitivity. As expected, higher soil water retention capacity mitigates meteorological drought. In contrast, a south-facing slope promoted sensitivity most likely due to higher solar radiation exposure (local underestimation of mSPEI) compared to north-facing slopes. Management factors explained 23% of the total variation. From the model, delayed first uses resulted in higher drought sensitivity. Such factors have been understudied so far, hindering a clear understanding of effect on grassland drought sensitivity. Grasslands that were preferentially grazed (type of use) showed higher drought sensitivity than mown grasslands. This is consistent with the field experiment by Deléglise *et al.* (2015), where drought conditions in grazing plots led to lower annual biomass than in mowing plots. The mean number of uses per year also increased sensitivity to drought. When grasslands are frequently used throughout the growing season, stored carbohydrates, which are necessary for plant regrowth, may become limited during drought events (Fulkerson and Donaghy, 2001). Our model also underlined the role of plant functional diversity (9%), which is mostly shared

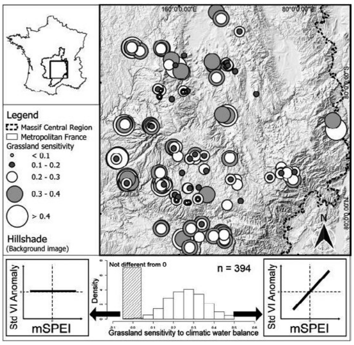

Figure 1. Spatial (top) and statistical (bottom) distributions of sensitivities to drought in the Massif central. Sensitivity is the slope of the linear relationship of the standardized (std) VI anomaly and mSPEI.

Table 1. Final sensitivity model (R-squared is 0. 5242) and variance partitioning results.

Category	Explanatory variable	Beta coefficient	t value	Pr(>|t|)
Pedoclimate	AWC (topsoil)	-0.5396292	-8.603	2.27e-14
	South-facing slope (aspect)	0.2355297	3.656	0.000372
Management	Time of first use (as GDD)	0.5163540	7.081	8.23e-11
	Type of use (grazing or mowing)	0.2936155	3.812	0.000213
	Mean number of uses	0.1295399	1.853	0.066173
Diversity	FDis: plant growth form	-0.1600510	-2.379	0.018819
	CWM: plant height	-0.1416316	-1.978	0.050051

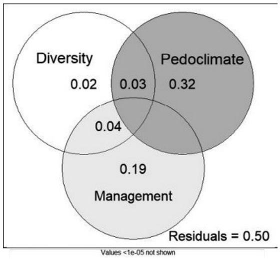

Figure 2. Variance partitioning of the main variable categories (R package: vegan).

with the pedoclimatic and management factors. We found that more diverse plant growth strategies promoted lesser drought sensitivity, as already shown in experimental studies (Weisser, *et al.*, 2017). In addition, grassland plots with taller plants, or higher CWM height, exhibited lower drought sensitivity, as confirmed by Nunes *et al.* (2017).

Conclusions

Using remote sensing, we assessed drought sensitivities on a wide range of grasslands across the Massif Central region. These sensitivities were highly variable, but the majority of the variation could be explained by pedoclimatic, diversity and management factors. AWC had the largest influence. We also underlined the role of diversity shared with pedoclimate and management on grassland drought sensitivity, which is in line with previous grassland-drought field experiments. We further assessed the relative importance of these drivers in real agricultural systems.

References

Beguería S., Vicente-Serrano S.M., Reig F. and Latorre B. (2014) Standardized precipitation evapotranspiration index (SPEI) revisited: Parameter fitting, evapotranspiration models, tools, datasets and drought monitoring. *International Journal of Climatology* 34, 3001-3023. https://doi.org/10.1002/joc.3887

Deléglise C., Meisser M., Mosimann E., Spiegelberger T., Signarbieux C., Jeangros B. and Buttler A. (2015) Drought-induced shifts in plants traits, yields and nutritive value under realistic grazing and mowing managements in a mountain grassland. *Agriculture, Ecosystems and Environ*ment 213, 94-104. https://doi.org/10.1016/j.agee.2015.07.020

Fulkerson W. and Donaghy D. (2001) Plant-soluble carbohydrate reserves and senescence - Key criteria for developing an effective grazing management system for ryegrass-based pastures: A review. *Animal Production Science* 41, 261-275. https://doi.org/10.1071/EA00062

IPCC (2021) *Climate Change 2021: The Physical Science Basis*. Contribution of Working Group I to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change [Masson-Delmotte V., P. Zhai, A. Pirani, S.L. Connors, C. Péan, S. Berger, ... and B. Zhou (eds.)]. Cambridge University Press, Cambridge, UK.

Ji L. and Peters A. (2003) Assessing vegetation response to drought in the northern Great Plains using vegetation and drought indices. *Remote Sensing of Environment* 87, 85-98. https://doi.org/10.1016/S0034-4257(03)00174-3

Nunes A., Köbel M., Pinho P., Matos P., Bello F., Correia O. and Branquinho C. (2017) Which plant traits respond to aridity? A critical step to assess functional diversity in Mediterranean drylands. *Agricultural and Forest Meteorology* 239, 176-184. https://doi.org/10.1016/j.agrformet.2017.03.007.

AOP (nd) Pôle Fromager AOP Massif Central. Prairies Naturelles du Massif Central: Plus de 10 ans de co-construction de R&D! Aurillac, France. Available at: http://pole-fromager-aop-mc.org/prairies-du-massif-central/

Reinermann S., Asam S. and Kuenzer C. (2020) Remote sensing of grassland production and management – a review. *Remote Sensing* 12(12), 1949. https://doi.org/10.3390/rs12121949

Wang L. and Qu J.J. (2007) NMDI: A normalized multi-band drought index for monitoring soil and vegetation moisture with satellite remote sensing. *Geophysical Research Letters* 34(20), L20405. https://doi.org/10.1029/2007GL031021

Weisser W., Roscher C., Meyer S., Ebeling A., Luo G., Allan E., Beßler H., ... and Eisenhauer N. (2017) Biodiversity effects on ecosystem functioning in a 15-year grassland experiment: Patterns, mechanisms, and open questions. *Basic and Applied Ecology* 23, 1-73. https://doi.org/10.1016/j.baae.2017.06.002

Dynamics of grassland vegetation in two sheep-grazed agrivoltaic systems in plain and upland areas

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Abstract

Agrivoltaic systems emerged to deal with the dual challenges of ensuring renewable energy production and agricultural production at the same site; however, their ability to deliver grassland ecosystem services is questioned. During one year, we studied direct effects of various shade conditions induced by solar panels on abiotic factors (light, soil water and temperature) and vegetation (growth height, greenness: NDVI, quantity of forage) at one plain and one upland sheep-grazed site. Under exclosure of grazing, three treatments per site were set up: control (without solar-panel influence), inter-rows (variable influence) and panel (full influence). The results showed significant modifications of plant microclimate under solar panels. Soil temperature was cooler in spring and summer, and the soil moisture response differed at each site. Unexpectedly, vegetation growth under the solar panels was taller in spring and summer than that in the control, and biomass was larger during summer drought, but the latter declined during spring of the following year. The results emphasised that, forage quantity and canopy greenness (NDVI) could be much wider in sheep-grazed agrivoltaic systems than in open grasslands

Keywords: photovoltaic panel, grassland, growth, biomass, microclimate

Introduction

The 2018 multiannual energy plan of the French government set a target for renewable energy production by photovoltaics of 35-45 GW by 2028. To meet this target and avoid land-use conflict, agrivoltaic systems could combine agricultural activity and photovoltaic systems. However, in view of the few studies available, especially in France and in pastoral areas, the ability of agrivoltaic systems to deliver ecosystem services is questioned. The main objective of this study was to describe seasonal (spring and summer) grassland dynamics at a plain and an upland site.

Materials and methods

Two sheep-grazed agrivoltaic systems in France were monitored from summer 2020 to spring 2021. The plain site of Braize (Br) (Allier – 46.68°N, 2.64°E) has been exploited since October 2018 on a sandy soil. The upland site of Marmanhac (Ma) (Cantal – 45.02°N, 2.45°E) has been exploited since January 2014 on a silty-sandy andosol. At these sites, south-oriented solar panels are set in the ground. The solar panel tables are 3.5 m wide, with 4 m spacing between rows, at the Br site, and 2.9 m wide, with 1.85 m spacing, at the Ma site. Three treatments were set up: 'Panels' (P, under the solar panels), 'Inter-row' (I, between two rows of panels), and 'Control' (C, without panel influence). In an area of exclosure of grazing, and for each treatment, three transects were set up (three probes each) to measure soil moisture and soil temperature at a depth of 20 cm (SMT100, STEP System GmbH, Germany). Light availability was measured using PAR sensors in P and C treatments (JYP1000, SDEC, France). Daily microclimatic variables were averaged by period: summer 2020 (16 July – 13 August (Br) and 24 July – 25 August (Ma)) and spring 2021 (3 May – 1 June (Br) and 7 May – 3 June (Ma)). Each month, on each side of the probes (i.e. in 54 quadrats (0.50×0.50 m^2)), daily vegetation growth height (cm d^{-1}), NDVI [0-1] (GreenSeeker, Trimble Ag, USA) and percentage of bare soil and moss were measured, as well as biomass regrowth (g m^{-2}), taken at a height of 5 cm (oven dried for 48 h at 60 °C). The quadrats on either side of a probe were averaged (n=9 per treatment). Treatment effects were tested for the variables measured during each period. When assumptions of normality and homogeneity of variances were not met, Kruskall-Wallis tests were performed, followed by Dunn's post-hoc test. When nonparametric tests were similar to the corresponding single-factor ANOVA, linear mixed models were used, followed by Tukey's post-hoc test. All statistical analyses were performed in R software (v 4.1.2).

Results and discussion

For both sites and periods, the presence of solar panels strongly decreased the radiation availability for plants (92-94% lower) and soil temperature (Br: -6.6 to -3.5 °C lower; Ma: -3.8 to -3.1 °C lower), compared to C (Table 1). However, the soil moisture response was less clear. In summer, soil moisture measured at Ma was higher in P than in C (+84%, Table 1) but not at Br. In spring of the following year this effect disappeared at both sites, because C had wetter soil than P (+ 43%, Table 1). These microclimatic results are explained mainly by effects of direct shade in P and confirm results obtained from other agrivoltaic systems (Armstrong *et al.*, 2016; Adeh *et al.*, 2018). Soil moisture dynamics were also related to infiltration of rain through the panels and to the soil water holding capacity at each site. Indeed, a soil with more organic matter (i.e. the Andosol at Ma) retains more water in summer under panels (Adeh *et al.*, 2018), which is not the case with a sandy soil, like that at Br.

Microclimatic conditions in I were intermediate to those in the other treatments when there was a significant difference between P and C treatments (Table 1). During summer in P, daily growth height (Br = +3,780%; Ma = +885%), NDVI (Br = +61%; Ma = +110%) and biomass production (Br = +396%; Ma = +366%) were much higher than those in C (Table 1). Solar panels behave as a 'parasol' by limiting desiccation and the stopping of growth, as emphasised in other studies (e.g. Adeh *et al.*, 2018). During the spring of the following year, the effect on growth height was still observed but to a lesser extent than in summer (Br = +50%, Ma = +73%, Table 1). Unlike in the summer, NDVI did not differ between treatments at Br, but was 16% higher in C than in P at Ma. These inverse effects on

Table 1. Means ± standard errors of vegetation monitoring at the two sites for two periods (summer 2020: 16 July – 13 August (Br) and 24 July – 25 August (Ma); spring 2021: 3 May – 1 June (Br) and 7 May – 3 June (Ma).[1]

Variables	**Tr**	**Summer 2020**		**Spring 2021**	
		Braize	**Marmanhac**	**Braize**	**Marmanhac**
Soil moisture (%)	C	6.32±0.50 a	15.40±0.94 b	17.51±0.99 a	35.83±0.94 a
	I	5.08±0.32 a	23.47±1.9 a	14.58±0.79 b	34.84±2 a
	P	7.38±0.43 a	28.36±2.08 a	12.28±1.12 c	33.42±2.14 a
Soil temperature (°C)	C	27.11±0.07 a	21.60±0.12 a	15.59±0.06 a	13.12±0.010 a
	I	24.91±0.61 ab	19.30±0.35 b	15.0±0.32 a	11.73±0.19 b
	P	20.50±0.15 b	17.81±0.12 c	12.05±0.07 b	9.99±0.05 c
Daily growth height (cm d^{-1})	C	0.005±0.002 b	0.020±0.005 b	0.402±0.036 b	0.346±0.020 b
	I	0.010±0.006 b	0.134±0.025 a	0.397±0.028 b	0.427±0.040 ab
	P	0.194±0.016 a	0.197±0.011 a	0.602±0.029 a	0.598±0.059 a
NDVI (0-1)	C	0.28±0.02 b	0.30±0.01 b	0.51±0.02 ab	0.75±0.02 a
	I	0.32±0.02 b	0.64±0.05 a	0.47±0.04 b	0.79±0.01 a
	P	0.45±0.02 a	0.63±0.03 a	0.55±0.02 a	0.65±0.04 b
Biomass regrowth (g m^{-2})	C	2.81±0.39 b	8.36±1.04 b	73.39±6.67 a	72.25±6.59 a
	I	3.13±0.77 b	35.81±5.16 a	51.31±5.59 b	60.40±4.26 a
	P	13.95±1.16 a	38.95±11.63 a	38.92±2.05 b	33.46±5.18 b

[1] For each variable, period and site, different letters indicate a significant (P<0.05) difference among treatments (Tr) (Control (C), Inter-panel rows (I), Underneath panels (P)).

NDVI can be explained by the bare soil area, which counterbalanced the daily growth in P, resulting in less biomass regrowth (Br: -89%; Ma: -116%), as highlighted by Armstrong *et al.* (2016). Our results in spring can also be explained by the shade produced by the panels, which reduces biomass production of temperate grassland species, especially at 10% of full light availability (Semchenko *et al.*, 2012). In addition, biomass response is related to plant density as well as to plant growth. The larger proportion of bare soil observed in spring in P could have been caused by several factors, such as a 'splash' effect during rainfall by runoff from structures (Armstrong *et al.,* 2016) or the past behaviour of sheep, which lie down under panels (Maia *et al.*, 2020). Soil colonization could also be limited by the ecological valence of heliophilic species in I and C and seed mortality under shade (Semchenko *et al.*, 2012). However, the positive effect of shade on growth height observed in both periods can be explained by plant acclimation induced by phototropism, as described by many authors (e.g. Jones (2014)). In summer at Br, the response of vegetation in I was similar to that of vegetation in C, whereas at Ma its response was closer to that of vegetation in P. This can be explained by the inter-rows being twice as wide at Br than at Ma. In spring of the following year, the response of I seemed similar to that of C at both sites, except for biomass at Br, which may have been due to moss cover.

Conclusions

Our study confirms that under summer drought conditions, solar panels act as 'parasol' for the vegetation, ensuring cooler and even wetter soil conditions (depending on soil texture) and thus promoting higher growth and biomass production at both plain and upland sites. These strong effects are buffered in spring but still maintained for growth height. The larger proportion of bare soil under solar panels counterbalances the higher growth height, which explains the decrease in biomass. The response of inter-row vegetation varied between sites, potentially in relation to the density of the infrastructure. More studies are necessary over several seasons and years to consider grazing effects on vegetation dynamics and ecosystem services provided by grasslands.

References

Adeh E.H., Selker J.S. and Higgins C.W. (2018) Remarkable agrivoltaic influence on soil moisture, micrometeorology and water-use efficiency. *PloS One* 13, e0203256.

Armstrong A., Ostle N.J. and Whitaker J. (2016) Solar park microclimate and vegetation management effects on grassland carbon cycling. *Environmental Research Letters* 11, 074016.

Jones H.G. (2014) *Plants and Microclimate. A Quantitative Approach to Environmental Plant Physiology*. Cambridge University Press.

Maia A.S.C., de Andrade Culhari E., Fonsêca V.D.F.C., Milan H.F.M. and Gebremedhin K.G. (2020) Photovoltaic panels as shading resources for livestock. *Journal of Cleaner Production* 258, 120551.

Semchenko M., Lepik M., Götzenberger L. and Zobel K. (2012) Positive effect of shade on plant growth: amelioration of stress or active regulation of growth rate? *Journal of Ecology* 100, 459-466.

Production and replacement costs of permanent grasslands compete with those of sown grasslands

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Abstract

Farmers' endorsement is crucial to conserve biodiversity in permanent grasslands, but the lack of visibility on their economic value is a major obstacle. We studied the production costs (i.e. the cost to produce one Mg of dry matter) and the replacement costs (i.e. the cost to replace grassland fodder with a mixture of wheat, soya and cereal straw) of 59 permanent and two sown grasslands from the Vosges Mountains (eastern France). We measured profitability as the difference between replacement costs and production costs. Our results highlighted a strong variability between grasslands, with lower production costs associated with grazing and higher production costs associated with low productivity. Permanent grasslands with low productivity were also associated to low profitability, but our calculation did not take into account their high conservation status which could deliver public subsidies. However, 75% of the mainly cut permanent grasslands were more profitable than the cut sown grassland, and 75% of the exclusively grazed grasslands were more profitable than the grazed sown grassland. Overall, permanent grasslands can be more profitable than sown grasslands while protecting biodiversity. We argue for the maintenance of agri-environment schemes that preserve grasslands of high ecological interest.

Keywords: economy, profitability, husbandry, ecosystem services

Introduction

European permanent grasslands are the main source of fodder, but they are often seen as poorly productive and thus, of poor economic interest. Production costs, replacement costs and profitability of permanent grasslands are mainly affected by management. Grazing is generally perceived as less expensive than cutting, because it requires less material and fuel. Fertilization improves yield and nutritive value, but increases production costs, especially when mineral fertilizer is used instead of manure. Several studies highlighted a positive correlation between plant biodiversity and profitability. These studies were often run on sown temporary grasslands (Schaub *et al.*, 2020a) or experimental sown permanent grasslands (Schaub *et al.*, 2020b), which limits the generalization of their results to spontaneous permanent grasslands (Tonn *et al.*, 2021). In this study, we calculated production costs, replacement costs and profitability of spontaneous permanent grasslands managed by farmers. We hypothesize that permanent grasslands can be more profitable than sown grasslands, due to their lower production costs.

Materials and methods

We studied 58 commercial permanent grasslands from the Vosges Mountains (eastern France). Grasslands were either cut, grazed, or cut and grazed, and N-fertilization varied from 0 to 259 kg ha^{-1}. Elevation, climate and soil properties also strongly differed between grasslands. We sampled fodder during the first use (at production peak) to analyse nutritive value in 2018 and 2019. We interviewed farmers to obtain information about management and to calculate mean annual yield production. Two representative sown grasslands were also studied: one only cut and one only grazed.

Production costs (€ Mg^{-1}) included costs for machinery use, working force, fuel and fertilizers (Bayeur *et al.*, 2013; Table 1). Replacement costs (€ Mg^{-1}) were purchase prices to substitute grassland fodder with soya, cereal grains, and cereal straw (Chambre d'Agriculture des Deux-Sèvres, 2018; Table 1). Profitability (€ Mg^{-1}) was the difference between replacement costs and production costs.

Results and discussion

Our results showed a high diversity of costs between grasslands: production costs ranged from 9.6 to 298.8 € Mg^{-1}, replacement costs from 105.6 to 221.6 € Mg^{-1} and profitability from -143.2 to 212 € Mg^{-1} (Figure 1). This diversity is mainly related to the mode of use and to fodder yields.

Grazed grasslands had lowest production costs, resulting in higher profitability than cut grasslands. Of the cut grasslands, 34 (81%) had positive profitability. The eight unprofitable cut grasslands produced low yield, mainly due to high altitude or sandy soil. However, high altitude and dry grasslands are known to be particularly important for biodiversity conservation (Napoleone *et al.*, 2021).

Unlike Schaub *et al.* (2020b), we did not observe a clear relationship between specific richness and profitability. However, it is now important to extend the scope of future research to the relationship between conservation status and profitability. For example, despite their important conservation role, the specific richness of high altitude and dry grasslands do not correlate with their conservation status (Napoleone *et al.*, 2021).

Among the cut permanent grasslands, 30 (75%) were more profitable than the sown cut grasslands. Among the grazed permanent grasslands, 12 (75%) were more profitable than the sown grazed grasslands. This result shows the high economic potential of permanent grasslands compared to sown temporary grasslands, and is critical to counter the destruction and abandonment of European permanent grasslands (Young *et al.*, 2005). Moreover, our calculation did not take into account the conservation status of permanent grassland which could provide public subsidies for biodiversity conservation and/or carbon sequestration. Also, the price of mineral fertilizer recently increased, which might increase costs of sown grasslands. This increase should weakly affect permanent grasslands: only 4 (7%) of the studied permanent grasslands received mineral fertilizer. Thus, permanent grasslands could be more resilient to global economy evolutions.

Table 1. Data used for production costs and replacement costs calculations, from Bayeur *et al.* (2013) and Chambre d'Agriculture des Deux-Sèvres (2018).

Management	**€ ha^{-1}**	**Mineral fertilizer**	**€ kg^{-1}**
Hay gathering	173	N	1
Wrapped bales gathering	208	P	1.1
Silage gathering	249	K	0.7
Grazing	49	S	0.2
Solid manure spreading	32	Replacement costs	€ Mg^{-1}
Liquid manure spreading	32	Soya	395
Mineral fertilizer spreading	8	Cereal grains	182
Harrowing	14	Cereal straw	70

Figure 1. Production costs, replacement costs and profitability of permanent grasslands (boxplots) and representative sown grasslands (white dots). Permanent grassland are mainly cut (n=42) or only grazed (n=16).

However, an integral switch from permanent grasslands to sown grasslands would induce massive adjustment at the farm level, due to changes in grassland yields quantity, quality and seasonality. Partial budget analysis or cost-benefice analysis would be appropriate to study the long-term economic consequences at the farm scale.

Conclusions

Most permanent grasslands were more profitable than sown grasslands while also protecting biodiversity. We argue for the maintenance of agri-environment schemes that preserve grasslands of high ecological interest, which often are less profitable.

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References

Bayeur C., Kleiber F., L'Hospitalier M., Loridat F. and Plantureux S. (2013) Typologie des prairies permanentes Vosges du Nord et Vosges Mosellanes : guide technique 2013.

Chambre d'Agriculture des Deux-Sèvres (2018) Prix des fourrages et aliments. Available at: https://deux-sevres.chambre-agriculture.fr/fileadmin/user_upload/Nouvelle-Aquitaine/103_Inst-Deux-Sevres/Documents/Technique_innovation/Productions_animales/ValeurAlim2018.xlsm

Napoleone F., Giarrizzo E. and Burrascano S. (2021) Habitat conservation state and plant diversity respond to different drivers in semi-natural grasslands. *Journal of Vegetation Science* 32 (4), e13055.

Schaub S., Buchmann N., Lüscher A. and Finger R. (2020a) Economic benefits from plant species diversity in intensively managed grasslands. *Ecological Economics* 168, 106488.

Schaub S., Finger R., Leiber F., Probst S., Kreuzer M., Weigelt A., Buchmann N. and Scherer-Lorenzen M. (2020b) Plant diversity effects on forage quality, yield and revenues of semi-natural grasslands. *Nature Communication* 11, 768.

Tonn B., Komainda M. and Isselstein J. (2021) Results from a biodiversity experiment fail to represent economic performance of semi-natural grasslands. *Nature Communication* 12, 2125.

Young J., Watt A., Nowicki P., Alard D., Clitherow J., Henle K., Johnson R., Laczko E., McCracken D., Matouch S., Niemela J. and Richards C. (2005) Towards sustainable land use: identifying and managing the conflicts between human activities and biodiversity conservation in Europe. *Biodiversity and Conservation* 14, 1641-1661.

Rethinking grasslands in 3D: feeding preferences of dairy cows between temperate fodder trees

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Abstract

Planting fodder trees in grasslands increases vegetation diversity, reduces grassland vulnerability to climate change and provides additional fodder resource during periods of drought. However, the palatability of temperate fodder trees remains poorly studied. During 10 mornings in July 2021, we allowed 12 dairy cows to feed freely in a 4-year-old chicory-based pasture planted with 168 pollarded trees from 4 species (common ash, white mulberry, Lutèce elm, Italian alder). Every 4 minutes, the number of cows browsing each individual tree was recorded (i.e. 550 scans per tree). A generalized linear mixed model (GLMM) was used to analyse the cows' feeding preferences among the four tree species. Results indicate a strong preference for Lutèce elm (280 of 470 feeding observations, i.e. 60% of browsing behaviour), and low preferences for common ash and Italian alder (respectively 7 and 6%). This study shows that fodder trees may represent a feeding resource complementary to herbage in summer. Further investigations are needed to confirm and understand this preference pattern, as well as to quantify the part of the diet fodder tree would represent.

Keywords: agroforestry, animal behaviour, browsing, pollard

Introduction

In many European regions, trees have been removed from agricultural ecosystems and this has adversely affected ecosystem services related to carbon sequestration, biodiversity conservation, soil enrichment and air and water quality (Jose, 2009). The use of woody species as animal fodder appears to offer an incentive to replant trees in agricultural systems. Tree browsing is already a common practice in Mediterranean and tropical areas, where herbage production is constrained during drought seasons (Vandermeulen *et al.*, 2018a). Recent *in vitro* studies have highlighted the good nutritional value of the leaves of certain tree species during summer, which were as much or even more nutritious than herbaceous forages such as ryegrass and cocksfoot under European temperate conditions (Mahieu *et al.*, 2021). However, these studies also highlighted that the contents of condensed tannins in tree leaves could exceed those in sainfoin, possibly decreasing their palatability and so feed intakes (Patra, 2009). Despite the growing interest for fodder trees in temperate areas, little is known about cattle preferences for different tree species. We therefore studied cow feeding preferences between four tree species.

Materials and methods

The study was conducted on a tree-planted plot within the OasYs system experiment (Novak *et al.*, 2016) located at the INRAE facility in Lusignan, western France (46°25'19.0'N, 0°07'18.1'E). We studied the feeding behaviour of 12 lactating dairy cows, from 2 to 7 years old and in their 1st to 4th lactation (101±17.1 days in milk). The cows were the product of a three-way crossbreeding between Hostein, Jersey and Scandinavian Red. The plot was a 4-year old chicory-based pasture of 2 ha, planted with 168 pollarded trees from 4 species (https://doi.org/10.15454/SRBXQ9): common ash (*Fraxinus excelsior*), white mulberry (*Morus alba*), Lutèce elm (*Ulmus* 'Nanguen') and Italian alder (*Alnus cordata*). Trees were planted in 2014 in four rows (20 m inter-row spacing), with a tree density of 84 trees ha^{-1}. Trees were pollarded at 50 or 80 cm above ground level in 2019. In July 2021, trees were classified into 6 foliar biomass classes, based on an expert's visual scanning. Before the browsing experiment, the grassland was grazed with no access to trees in order to reduce herbage availability. Tree browsing occurred on the mornings of 12 to 21 July 2021, from 8:30 am to 12:30 pm. Four observers recorded cows' feeding preferences using a scan sampling focused on individual trees. Every 4 minutes, the number of cows that were feeding on each pollard was recorded (i.e. 550 scans per pollard, or 92,400 scans in total).

A generalized linear mixed model (GLMM) was used to investigate whether cows exhibited feeding preferences among the four tree species. The model was controlled by observer identity and tree biomass class, to avoid biases. We computed estimated marginal means to highlight food preferences among trees species. Statistical analysis were performed with R software (v 4.1.2), and the packages 'lme4' (v 1.1-27.1) and 'emmeans' (1.7.0).

Results and discussion

Cows were observed feeding on trees on 472 occurrences over the 10 mornings. Cows significantly preferred Lutèce elm (280 feeding occurrences, 60%), followed by white mulberry (128 occurrences, 27%), then common ash (31 occurrences, 7%) and finally Italian alder (28 occurrences, 6%) ($P \leq 0.003$, Figure 1, Table 1).

Our results confirm that cattle exhibit feeding preferences between tree species. Common ash was one of the least preferred browsed species, despite its high nutritional value (Mahieu *et al.*, 2021) and potential high voluntary intake, as observed in an *in vivo* study with sheep fed indoors *ad libitum* (Bernard *et al.*, 2020). These inconsistencies may be due to the choice situation that included other tree species potentially preferred by cows, to differences in chemical composition between directly browsed ash trees and cut then browsed trees, or to the lack of experience of cows relative to tree browsing. Differences in ash palatability between sheep and cows may also be involved as supported by the very low ash preference by dairy heifers observed by Vandermeulen *et al.* (2018b).

Lutèce elm was the most preferred species, although its nutritional value was lower than that of common ash, white mulberry and Italian alder in a previous *in vitro* study (Mahieu *et al.*, 2021). The assessment of our tree leaves chemical composition will help to better understand its role in cows' feeding preferences, relative to the one of sensory characteristics such as taste, odour or resistance to fracture. We noticed that Lutèce elm leaves were easier to detach from the branches and had a rougher texture, which may have facilitated their access by the cows, leading to a greater attractiveness.

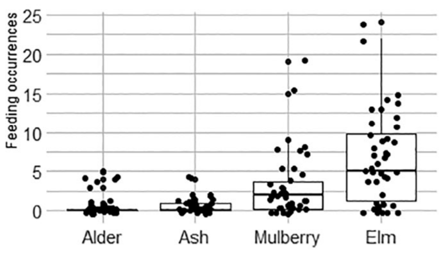

Figure 1. Number of cows observed feeding on trees (i.e. feeding occurrences). Each black dot represents one of the 168 studied trees.

Table 1. Feeding preferences between preferred tree species (first column) and less eaten tree species (first line). *P*-values are those of estimated marginal means.

	Mulberry	Ash	Alder
Elm	$P<0.001$	$P<0.001$	$P<0.001$
Mulberry		$P<0.001$	$P<0.001$
Ash			$P=0.003$

Conclusions

This study is one of the first offering lactating dairy cows the choice between four pollarded temperate tree species directly browsable at pasture. Cows expressed strong feeding preferences which were inconsistent with what is known about species nutritional values from literature. Further studies are needed to confirm and understand this preference pattern, as well as to quantify the part of the dairy cow diet that tree fodder would represent.

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References

Bernard M., Ginane C., Deiss V., Emile J.C. and Novak S. (2020) Voluntary intake and *in vivo* digestibility of leaves from two tree species: common ash (*Fraxinus excelsior*) and white mulberry (*Morus alba*). *Fourrages* 242, 55-59.

Favreau-Peigné A., Baumont R. and Ginane C. (2013) Food sensory characteristics: their unconsidered roles in the feeding behaviour of domestic ruminants. *Animal* 7, 806-813.

Jose S. (2009) Agroforestry for ecosystem services and environmental benefits: an overview. *Agroforestry Systems* 76, 1-10.

Mahieu S., Novak S., Barre P., Delagarde R., Niderkorn V., Gastal F. and Emile J.-C. (2021) Diversity in the chemical composition and digestibility of leaves from fifty woody species in temperate areas. *Agroforestry Systems* 95, 1295-1308.

Novak S., Liagre F. and Emile, J.C. (2016). Integrating agroforestry into an innovative mixed crop-dairy system. In: *Celebrating 20 Years of Agroforestry Research in Europe*. Montpellier, France, 4 pp.

Patra A.K. (2009) A meta-analysis on effects of supplementing low-quality roughages with foliages from browses and tree fodders on intake and growth in sheep. *Livestock Science* 121, 239-249.

Vandermeulen S., Ramírez-Restrepo C.A., Beckers Y., Claessens H. and Bindelle J. (2018a) Agroforestry for ruminants: a review of trees and shrubs as fodder in silvopastoral temperate and tropical production systems. *Animal Production Science* 58, 767-777.

Vandermeulen S., Ramírez-Restrepo C.A., Marche C., Decruyenaere V., Beckers Y. and Bindelle J. (2018b) Behaviour and browse species selectivity of heifers grazing in a temperate silvopastoral system. *Agroforestry Systems* 92, 705-716.

Taxonomic and functional biodiversity positively influence agronomic characteristics of permanent grassland

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Abstract

European permanent grasslands are the main source of livestock fodder and the main hotspot of botanical diversity, but the trade-offs between fodder production and botanical diversity conservation remain debated. This study aims to identify what grassland features influence fodder characteristics and to estimate the direction of correlation between biodiversity and fodder characteristics. We focused on a diverse sample of 58 permanent grasslands from the Vosges Mountains (eastern France). For each grassland, we estimated the quantity and quality of the fodder using 10 fodder characteristics, and extracted 26 grassland features related to management, environment, and taxonomic and functional diversity. We used random forest algorithms to investigate what grassland features best predicted fodder characteristics. Our results showed that fodder characteristics could be well estimated using only 14 grassland features (R^2>0.4) pertaining to management, soil, climate, taxonomic and functional diversity. Diversity was negatively correlated to three fodder characteristics, but positively correlated to six. We conclude that biodiversity is a key predictor of grassland fodder characteristics, and enhances most of them. We argue that conservation of permanent grassland biodiversity and agricultural production can both benefit from synergies.

Keywords: yield, nutritive value, antioxidant, management, environment, ecology

Introduction

Permanent grasslands are the main source of fodder in Europe, and may host a high botanical diversity (Wilson *et al.*, 2012). It is generally considered that there is opposition between fodder production and botanical diversity. This is because intensification of agricultural practices increases yields, but greatly reduces biodiversity (Gaujour *et al.*, 2012). In mature permanent grasslands, studies concluded there are hump-shaped relationships between diversity and biomass production: potential yields are maximized at intermediate levels of biodiversity (Guo, 2007). Effects of diversity on nutritive value differ among studied characteristics, but remain weak or insignificant in sown experiments (Schaub *et al.*, 2020). Here, we aimed to study the effect of taxonomic and functional diversity in permanent grasslands managed by farmers, under environmental and management gradients. We hypothesize that poorly studied nutritive values could be positively related to biodiversity, especially mineral content, antioxidant activity and flexibility of management.

Materials and methods

We studied 58 permanent grasslands from the Vosges Mountains (North-Eastern France). Environmental conditions strongly differed among grasslands: elevation varies from 184 to 1,222 m a.s.l and soil pH from 4.2 to 8.0. Grasslands are cut, grazed, or cut and grazed, and N-fertilization varies from 0 to 259 kg ha^{-1} (mineral and organic fertilization, and animal deposition). In 2018 and 2019, we realized botanical relevés and vegetation samples in six 0.5 m^2 per grassland. Vegetation samples were used to calculate yield (then normalized at 1,100 degree day – base 0 °C from 1st February), pastoral value and flexibility of management, and to measure neutral detergent fibre, acid detergent fibre, acid detergent lignin, crude protein, mineral content, potential milk production and antioxidant activity (i.e. 10 agronomic characteristics). From botanical relevés, we calculated four taxonomic (species richness, Shannon exponential, inverse Simpson, taxonomic evenness) and four functional (functional richness, functional evenness, functional diversity, Rao's Q diversity) features, as well as Ellenberg indices for fertility and humidity. We extracted 15 more features from farmers' interviews, soil analysis and topographic model, to inform about management and environment.

We used spatial random forest algorithms to investigate what grassland features best predicted agronomic characteristics (Benito, 2021). For each agronomic characteristic, we then selected the few features allowing the best prediction accuracy (R^2). We assume that characteristics were predicted well if their best R^2 was higher than 0.4. Finally, we check for direction of the correlation between agronomic characteristics and their selected features. This statistical approach did not aim to highlight whether there was a relation between grassland characteristics and features, but to highlight the best features for characteristic predictions.

Results and discussion

We could predict all agronomic characteristics correctly: all R^2 are higher than 0.4. Among the 26 grassland features related to management, environment, taxonomic and functional diversity, only 14 were selected for the best prediction of agronomic characteristics. Six out of ten characteristics required biodiversity features to be best predicted. Six correlations between biodiversity and characteristics were positive, and three were negative (Table 1). Ellenberg fertility index was the most important feature, useful for the prediction of eight out of ten characteristics (Pittarello *et al.*, 2020).

The relation between yield and biodiversity were hump-shaped, confirming the conclusion of Guo (2007). However, the results from our large scale study differed from those of Schaub *et al.* (2020) obtained in one experimental station: biodiversity had a mostly positive effect on nutritive value. Similarly to Brun *et al.* (2019), our results highlighted that the relation between biodiversity and agronomic characteristics depends on considering taxonomic or functional diversity, but our study brings new horizons about the relation between biodiversity and nutritive value.

Table 1. Selected features for the prediction of each agronomic characteristics, prediction quality (R^2) and direction of the correlation between features and characteristics (positive, negative or unselected).[1,2]

Agronomic characteristics	R^2	Management					Environment					Biodiversity			
		Ellenberg fertility	LUI	Number of cut	Degree day	Mode of use	Ellenberg humidity	Elevation	Soil sand	Soil pH	Soil depth	Taxonomic richness	Shannon exponential	Functional richness	Rao's Q diversity
Yield (1,100 d.g)	0.68	+		+								-	+		
Pastoral value	0.68	+								+			+	-	
NDF	0.56	+			+	+		-		+					
ADF	0.50	+			+	+	-	-		+					+
ADL	0.45	+	-	-	+	+	-	+							+
CP	0.56	+			-		+								
Mineral content	0.55	+			-		+			+		+	+		
Milk potential	0.45		+		-			+							
Flexibility	0.58	-								-			-		
Antioxidant activity	0.41								-		-				

[1] Unselected features are not shown. 'Mode of use' is the proportion of cut on grassland number of use.

[2] NDF = neutral detergent fibre; ADF = acid detergent fibre; ADL = acid detergent lignin; CP = crude protein.

As hypothesized, mineral content and flexibility of management were influenced by taxonomic diversity, but they were weakly influenced by functional diversity. However, antioxidant activity was weakly sensitive to diversity features, but could be related to water stress (Sairam and Srivastava, 2001).

Conclusions

Only three out of ten agronomic characteristics were negatively related to one biodiversity feature. These promising results highlight that biodiversity conservation and agricultural production can both benefit from synergies. More research is needed to better understand the role of botanical diversity on production and conservation, especially in the face of climate change.

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References

Benito B.M. (2021) SpatialRF: easy spatial regression with random forest.

Brun P., Zimmermann N.E., Graham C.H., Lavergne S., Pellissier L., Münkemüller T. and Thuiller W. (2019) The productivity-biodiversity relationship varies across diversity dimensions. *Nature Communication* 10, 5691.

Gaujour E., Amiaud B., Mignolet C. and Plantureux S. (2012) Factors and processes affecting plant biodiversity in permanent grasslands. A review. *Agronomy for Sustainable Development* 32, 133-160.

Guo Q. (2007) The diversity–biomass–productivity relationships in grassland management and restoration. *Basic and Applied Ecology* 8, 199-208.

Pittarello M., Lonati M., Ravetto Enri S. and Lombardi G. (2020) Environmental factors and management intensity affect in different ways plant diversity and pastoral value of alpine pastures. *Ecological Indicators* 115, 106429.

Sairam R.K. and Srivastava G.C. (2001) Water stress tolerance of wheat (*Triticum aestivum* L.): Variations in hydrogen peroxide accumulation and antioxidant activity in tolerant and susceptible genotypes. *Journal of Agronomy and Crop Science* 186, 63-70.

Schaub S., Finger R., Leiber F., Probst S., Kreuzer M., Weigelt A., Buchmann N. and Scherer-Lorenzen M. (2020) Plant diversity effects on forage quality, yield and revenues of semi-natural grasslands. *Nature Communication* 11, 768.

Wilson J.B., Peet R.K., Dengler J. and Pärtel M. (2012) Plant species richness: the world records. *Journal of Vegetation Science* 23, 796-802.

Involvement of fructans in the protection of leaf meristems of grassland species during drought

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Abstract

Due to climate change, grasslands are subjected to more frequent and severe drought periods. Better knowledge of the drought resistance mechanisms of plant species should allow the adaptation of agricultural practices to contribute to the resilience of grasslands in the face of climate change. Fructans are the main plant non-structural carbohydrates in temperate grasses. In addition to their role as carbon reserve, they also contribute to resistance to abiotic stresses, but the underlying mechanisms are poorly understood. The objectives of this study were to evaluate the potential of fructans to protect membranes during drought in leaf meristems of perennial ryegrass (*Lolium perenne*), a major species of temperate grasslands. After a six-day lag, water content and membrane stability at the base of the leaves decreased in the water-stressed plant while fructan content was maintained, indicating that their metabolism was preserved despite the dehydration. After 12 days of drought, cell membrane stability decreased further and fructans were released from the vacuole to the apoplast. This release could help protect the plasmalemma of meristematic cells. However, this protection must be impaired by sucrose accumulation when the water stress is prolonged.

Keywords: *Lolium perenne*, drought, fructans, membrane protection, apoplastic fluid

Introduction

With the increasing frequency of drought episodes due to climate change, water stress is an important factor to consider in the development of new cultivars and agricultural practices in temperate areas. In many grassland species, fructans represent a dynamic carbon pool which not only constitutes a carbon reserve but also contributes to the resistance to abiotic stresses, by stabilizing cell membranes (Hincha *et al.*, 2007). We aim to study the involvement of fructans in the protection of leaf meristems during drought in *L. perenne*, a major species of temperate grasslands. We assumed that under drought, (1) the water-soluble carbohydrates (WSC) in the leaf meristems changes in terms of quality (degrees of polymerization) and quantity (content) and that these changes affect membrane stability as observed in *Dactylis glomerata* and *Holcus lanatus* (Volaire *et al.,* 2020), (2) fructans migrate from the vacuole to the apoplast, as has been shown in response to freezing (Livingston and Henson, 1998), and that this migration allows membrane protection.

Materials and methods

Seeds (*L. perenne* var. 'Delika') were germinated on a fine wire mesh in contact with pure water for seven days. The seedlings were then transferred to pots (11×11×20 cm) containing a mixture (50:50) of sand and perlite. The plants (25 per pot) were grown in a controlled environment with a day/night temperature of 20/18 °C, 60% relative humidity, and a 16h photoperiod of 400 μmol m^{-2} s^{-1} of photosynthetic active radiation. Each pot was irrigated 4 times per day with 60 ml of a nutrient solution containing 1mM NH_4NO_3 (see Ould-Ahmed *et al.,* 2014 for the other nutrients). After 40 days of growth the plants were cut at 5 cm above the ground; the well-watered plants continued to be irrigated while the irrigation was stopped for the water-stressed plants. The 0-3 cm of the base of leaves (stubble) containing the leaf meristems as well as the leaf sheaths of mature leaves were harvested after 0, 6, 9, 12, 15 and 18 days. Water content, cell membrane stability, water-soluble carbohydrate content and analysis by high performance anion exchange chromatography coupled with pulsed amperometric detector (HPAEC-PAD) were carried out as described in Volaire *et al.* (2020). The measurement of cell membrane stability (CMS) was adapted from Charrier and Améglio (2011) and the method for obtaining the apoplastic fluid from O'Leary *et al.* (2014).

Results and discussion

After a latency of six days, the water content and the membrane stability at the base of the leaves decreased in the water-stressed plant (Figure 1A,C). The decrease was particularly rapid and pronounced for membrane stability, indicating that the variety 'Delika' is sensitive to drought. Under both conditions the decrease in fructan content during the first six days, and the increase during the following six days, correspond to the well-known U-shape curve due to mobilization and replenishment of fructan reserves after defoliation (Morvan-Bertrand *et al.,* 1999). In addition, the increase of fructan content after day 6 in water-stressed plants indicates that the metabolism of fructans was preserved despite the decrease in water content. On day 12, the fructan content started to be slightly lower in water-stressed plants but the HPAEC-PAD chromatograms obtained with the water-soluble extracts (Figure 2A,B) show that the distribution of polymers (DP 8 to 40) was not altered. This indicates that, contrary to our first hypothesis, the water stress did not induce polymerization or depolymerization of fructans during the first 12 days. However, as assumed in our second hypothesis, analysis of fructans in apoplastic fluid (Figure 2C,D) show a large increase of fructan content in water-stressed compared to well-watered plants. This indicates that fructans migrated from the vacuole in response to drought. As the membrane stability decreased sharply during this period (Figure 1B), we suggest that fructans stored in the vacuoles of the older leaf sheaths were released into the apoplast after rupture of the membranes and that this release could help protect the plasmalemma of meristematic cells. After day 12, the fructan content began to decrease in water-stressed plants in parallel with the increase of sucrose content revealing that severe drought affected fructan synthesis from sucrose. This strong increase in sucrose content could explain the decrease of membrane stability, as previously suggested by Volaire *et al.* (2020).

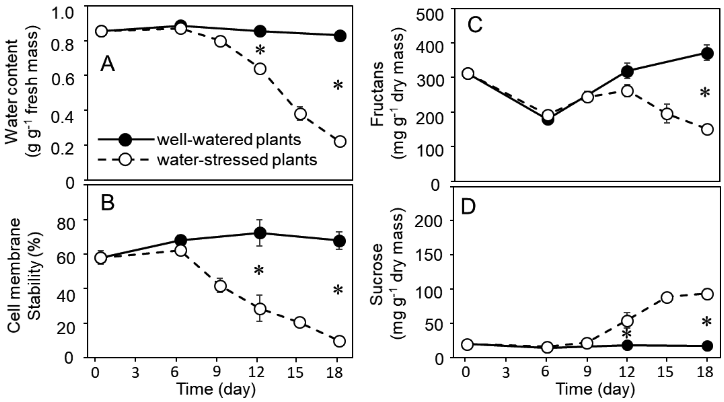

Figure 1. Water content (A), cell membrane stability (B), fructan (C) and sucrose (D) contents in the base of the leaves after stopping irrigation (water-stressed plants, white circles) and in well-watered plants (black circles). Values are mean ± standard error with n=4; * indicates P<0.05 after the t-test comparing control and drought.

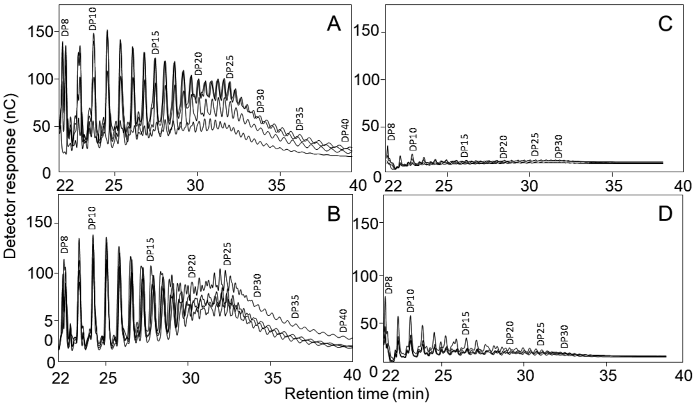

Figure 2. HPAEC-PAD chromatograms of fructans with degrees of polymerization (DP) greater than 7 in water-soluble extracts (A, B) and in apoplastic fluids (C, D) of the base of the leaves from plants sampled 12 days after stopping irrigation (water-stressed plants, B, D) or from well-watered plants (A, C). In each panel, the four chromatograms correspond to biological replicates (n=4).

Conclusions

Under drought, the fructans released from the vacuoles to the apoplast due to the rupture of membranes could help protect the leaf meristems by interaction with the plasmalemma. To assess the relationship between migration of fructans, membrane stability and drought resistance, this approach will be used with other *L. perenne* varieties or grass species and with plants treated with biostimulants for improving drought resistance. In addition, to deepen the understanding of this cell protection mechanism, we aim to visualize the migration of fructans by immunolocalization using developing anti-fructan antibodies.

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References

Charrier G. and Améglio T. (2011) The timing of leaf fall affects cold acclimation by interactions with air temperature through water and carbohydrate contents. *Environmental and Experimental Botany* 72, 351-357.

Hincha D.K., Livingston D.P., Premakumar R., Zuther E., Obel N., Cacela C. and Heyer A.G. (2007) Fructans from oat and rye : Composition and effects on membrane stability during drying. *Biochimica et Biophysica Acta* 1768, 1611-1619.

Livingston D.P. and Henson C.A. (1998) Apoplastic sugars, fructans, fructan exohydrolase, and invertase in winter oat: responses to second-phase cold hardening. *Plant Physiology* 116, 403-408.

Morvan-Bertrand A., Boucaud J. and Prud'homme M.-P. (1999). Influence of initial levels of carbohydrates, fructans, nitrogen, and soluble proteins on regrowth of *Lolium perenne* L. cv. Bravo following defoliation. *Journal of Experimental Botany* 341, 1817-1826.

O'Leary B.M., Rico A., McCraw S., Fones H.N. and Preston G.M. (2014) The infiltration-centrifugation technique for extraction of apoplastic fluid from plant leaves using *Phaseolus vulgaris* as an example. *Journal of Visualized Experiments* 94, 1-8.

Ould-Ahmed M., Decau M., Morvan-Bertrand A., Prud'homme M.-P., Lafrenière C. and Drouin P. (2014) Plant maturity and nitrogen fertilization affected fructan metabolism in harvestable tissues of timothy (*Phleum pratense* L.). *Journal of Plant Physiology* 171, 1479-1490.

Volaire F., Morvan-Bertrand A., Prud'homme M.-P., Benot M.-L., Augusti A., Zwicke M., Roy J., Landais D. and Picon-Cochard C. (2020) The resilience of perennial grasses under two climate scenarios is correlated with carbohydrate metabolism in meristems. *Journal of Experimental Botany* 71, 370-385.

Effect of intensive management on grassland mixtures

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Abstract

Pasture demand in spring and autumn on sheep farms heightens attention to the regrowth capacity in seed mixtures and their response to variable climatic growing conditions. To investigate this we established field trials at eleven locations in Norway ranging from 59 °N to 68°N, and localized on the margin of growing perennial ryegrass (*Lolium perenne*) to assess dry matter yield (DMY) in seven different grassland mixtures and one pure species. Mixtures differed in species quantity and proportion. In addition to one or two cuts in summer, trials were also cut in spring and autumn to simulate grazing. Mean DMY in three ley years was 9.86 t ha^{-1} yr^{-1} with only small differences between seed mixtures. Pure orchard grass (*Dactylis glomerata*) and a five-species mixture without timothy (*Phleum pratense*) yielded higher than mixtures with timothy when winter conditions were optimal, otherwise it was lower. Mixtures without timothy yielded less in spring and more in autumn compared to mixtures with timothy. Multispecies mixtures with a low proportion of timothy (30%) may be a strategy to adapt to changing climate as different species will dominate at different climate conditions. Demand for pasture herbage in spring or autumn should also be considered when choosing seed mixtures.

Keywords: seed mixtures, grass yield, simulated grazing, forage quality

Introduction

The Norwegian growing season has become longer due to climate change. This requires seed mixtures that contain species with good regrowth throughout the season, or seed mixtures that complement each other over years as much as possible, i.e. when timothy regresses other species will take over. Timothy has been the major grass species in the Nordic countries due to its superior persistence (Larsen and Marum, 2006; Østrem *et al.*, 2013). In most Norwegian-marketed seed mixtures timothy has been the dominating species although it does not tolerate intense grazing or many cuts (Steinshamn *et al.*, 2016). Therefore, species with increased regrowth capacity should be increased in seed mixtures used for both grazing and harvesting. The aim of the study was to investigate the appropriateness of different seed mixtures for grazing and harvesting in terms of yield and duration when they are used in climatically different areas and if a seed mixture with low proportion of timothy can produce at a satisfactory yield level.

Materials and methods

Eleven field trials were established in 2013 or 2014 in coastal and inland climates of Norway at about 59-68°N, of which eight were considered as southern and three as northern trials, according to their latitude or altitude above sea level. The number of harvests varied between the trials according to geographical location and the length of the growing season. There was at least one cut in spring and one in autumn to simulate grazing, in addition to one or two cuts for silage production during summer. At harvesting, plant samples were taken to determine dry matter by drying for two days at 60 °C. Yield was registered for three ley years. Seed mixtures of timothy (30%), meadow fescue (*Festuca pratensis*), smooth meadow grass (*Poa pratensis*), red clover (*Trifolium pratense*) and white clover (*Trifolium repens*) were tested in which the last 10% were either two different cultivars of perennial ryegrass (Figgjo or Trygve), two different cultivars of Festulolium (Hykor or Lofa) or smooth meadow grass (seed mixtures 1-5). Additionally pure orchard grass (*Dactylis glomerata*) (6), a mixture without timothy (7) and a mixture with smooth brome grass (*Bromus Inervis*) (8) were tested (Table 1).

Table 1. Content of different species (weight %) in the different tested seed mixtures 1-8.

Species	Scientific name	Cultivar	1	2	3	4	5	6	7	8
Timothy	*Phleum pratense*	Grindstad/ Nordeng	30	30	30	30	30			25
Meadow fescue	*Festuca pratensis*	Fure	20	20	20	20	20		20	
Smooth meadow-grass	*Poa pratensis*	Knut	20	20	20	20	30		20	15
Red clover	*Trifolium pratense*	Lea	10	10	10	10	10			10
White clover	*Trifolium repens*	Hebe	10	10	10	10	10			
Perennial ryegrass	*Lolium perenne*	Figgjo[1]/Trygve[2]	10[1]	10[2]					20[1]	
Festulolium	*Festulolium* sp.	Hykor[1]/Lofa[2]			10[1]	10[2]			20[2]	
Orchard grass	*Dactylis glomerata*	Frisk/Laban						100	20	
Smooth brome	*Bromus inermis*	Leif								50

Statistics

Yields were tested with trials, mixtures, and ley year as fixed variables to distinguish significant effects. R studio 4.1 with 'Tidyverse' package was used for statistical analysis of total dry matter yield per year. Distribution of yield throughout the season were carried out in all trials that had yield registration in both spring, summer and autumn.

Results

The average total dry matter yield (DMY) for all 11 trials over three years was 9.86 t ha^{-1} yr^{-1}. DMY varied from 10.1 t ha^{-1} yr^{-1} (mixture 6, 100% orchard grass) to 9.64 t ha^{-1} yr^{-1} (mixture 8, smooth brome mixture), and pure orchard grass gave a significantly higher yield than the smooth brome mixture (Figure 1). In the second ley year DMY was significantly higher (10.4 t ha^{-1} yr^{-1}) than in first and third year. DMY of all field trials as mean for three ley years ranged from 10.1-9.64 t ha^{-1} yr^{-1}.

There were significant interaction effects between trials and seed mixtures for all southern trials due to the large yield differences between locations. There were also significant interactions between trials and years, and treatments and years. Southern and northern trials analysed separately did not give significant differences between the different seed mixtures. Both southern and northern trials were statistically different within their geographical group.

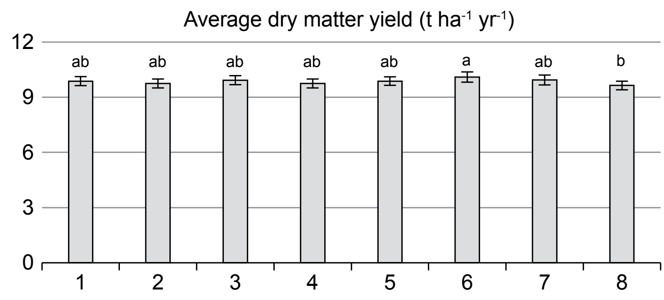

Figure 1. The average total dry matter yield per year for 8 mixtures over all trials over three years (n=99). Different letters represent significant difference between mixtures based on Tukey's test.

Discussion

The study showed that all the tested seed mixtures, except the bromegrass mixture, are appropriate mixtures for an intensive grazing and harvesting regime in terms of yield and duration when they are used in climatically different areas.

The distribution in crops throughout the season varied for the different seed mixtures. The timothy-based mixtures did best in the spring, and 100% orchard grass and the mixture without timothy did best in the autumn. With four annual harvests, the lower regrowth capacity in timothy was demonstrated along with sufficiently overwintering ability in the species and cultivars included in addition to yield potential. At sheep farms a seed mixture with low proportion of timothy will satisfy the need of good growth in spring and autumn.

As for timothy, the regrowth capacity of bromegrass is rather restricted and is of low value in an intensive management system as studied.

The results are interesting in terms of the rather small differences between seed mixtures in the different field trials and in showing considerable yield difference between sites. This means that a multispecies seed mixture may facilitate a satisfactory DMY when grown in very different environments both regarding length of growing season and overwintering conditions. Winter damage can happen even in an area where it is not expected. We saw this in the winter after the 2012 establishment year, where in some field trials perennial ryegrass did not survive and mixtures with 10% Hykor and 30% smooth meadow-grass gave the largest and second largest yield, while the mixture without timothy yielded the second lowest. For the trials that were established in 2013, 100% orchard grass gave the largest yields, closely followed by the mixture without timothy, while many of the timothy-based mixtures yielded lower. In general, there were small differences between the mixtures in total annual yield over years. The species that have performed best under different conditions have taken over in the mixtures. It may therefore be appropriate to use versatile mixtures so that the more winter-hardy species can take over in the event of winter damage. The benefits of using versatile mixtures in a Nordic climate is also found in other studies (Sturludottir *et al.*, 2013).

References

Larsen A., and Marum P. (2006) Breeding goals and possibilities in future timothy breeding. In: Sveinsson T. (ed.) *Timothy productivity and forage quality possibilities and limitations*. Report from NJF Seminar 384, Akureyri, Iceland. Rit LbhÍ 10, pp. 31-39.

Steinshamn, H., Nesheim, L. and Bakken A.K. (2016) Grassland production in Norway. In: *Grassland Science in Europe* 21, 4-8.

Sturludottir E., Brophy C., Belanger G., Gustavsson A-M., Jørgensen M., Lunnan T. and Helgadottir A. (2013) Benefits of mixing grasses and legumes for herbage yield and nutritive value in Northern Europe and Canada. *Grass and Forage Science* 69, 229-240.

Østrem L., Volden B. and Larsen A. (2013) Morphology, dry matter yield and phenological characters at different maturity stages of ×*Festulolium* compared with other grass species. *Acta Agriculture Scandinavica, Section B, Soil and Plant Science* 63, 531-542.

Impact of irrigation, cutting and fertilization on the phenology of Sahelian ranges

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Abstract

The main source of feed of Sahelian livestock is the annual herbaceous layer of Sahelian rangeland. The annual grass has a quick growth during the rainy season. Growth stops when the grass is flowering because of the photoperiod. The study of phenology is a key factor for the understanding of herbaceous growth. The objective of our study was to evaluate the impact of management on phenology. On ungrazed rangeland in northern Senegal, we tested 22 different irrigation, cutting and fertilization treatments repeated four times. Treatments included water regime (rainfall with and without irrigation), cutting (height and period of cutting), and fertilization (cattle manure). Each water regime was composed of treatments coupled or not with cutting or fertilization. We monitored the phenology of each treatment every ten days. The phenology was noted in three categories: vegetative stage, reproductive stage, senescence stage. At the beginning of the rainy season, before cutting, irrigation has effect on vegetative stage. During the season this effect disappeared. Only the effect of cutting is expressed until the end of the season on the phenological stage. This study confirms previous results that showed phenology is mostly influenced by cutting in the Sahel region.

Keywords: northern Senegal, ungrazed, herbaceous, cattle manure, rainfall

Introduction

The herbaceous layer plays an important role in livestock production in the Sahel. It is the principal source of feed for livestock in the pastoral system, which is one of the main economic resources in the Sahel (Akpo *et al.*, 2003; Diop, 2007). This vegetation is strongly dependent on rainfall (annual variations, quantity, and distribution of rainfall) and influenced by the management practices adopted by the population. The study of phenology is a key factor in understanding herbaceous growth. However, information about the impact of irrigation, cutting and fertilization on the phenology of Sahelian rangelands is lacking. The objective of this study was to evaluate the impact of management practices and the water regime on the phenology of the herbaceous layer of Sahelien rangelands.

Material and methods

The trial was conducted on an ungrazed site at the Centre de Recherches Zootechniques (CRZ) of Dahra in northern Senegal. Eighty-eight plots of 1 m^2 were delimited and 22 different irrigation, cutting and fertilization treatments were tested. The total cumulative rainfall received by the plots was 379.2 mm. The treatment description is presented in Table 1. We evaluated the relative coverage of plants at the vegetative stage (%V), reproductive stage (%F), and senescence (%S) by plot. The non-parametric test of Kruskal Wallis and the Dunn.test function were used to compare the phenological stage between the treatments using the version 4.1.2. of R software.

Table 1. Lists of treatments carried out.

Treatments	Quantity of water (mm/m^2)	Start date of irrigation (2021)	Duration (months)	Cutting height	Cutting period	Fertilizer ($kg\ ha^{-1}$)
2						
1	120	mid-July	1			
3	120	mid-August	1			
4	100	mid-July	2			
5	100	mid-August	2			
6				low to the ground	27/07/21	
7				low to the ground	04/09/21	
11				5 cm from the ground	27/07/21	
12				5 cm from the ground	04/09/21	
10	100	mid-July	2	low to the ground	27/07/21	
8	100	mid-July	2	low to the ground	04/09/21	
9	100	mid-July	2	5 cm from the ground	27/07/21	
13	100	mid-July	2	5 cm from the ground	04/09/21	
14	100	mid-August	2	low to the ground	27/07/21	
15	100	mid-August	2	low to the ground	04/09/21	
16	100	mid-August	2	5 cm from the ground	27/07/21	
17	100	mid-August	2	5 cm from the ground	04/09/21	
18						1000
19						2,000
20	100	mid-July	2			1000
21	100	mid-July	2			2,000
22	100	mid-August	2			1000

Results and discussion

On 27 July 2021, the %V of irrigated treatment 8 (64±11%), was significantly lower than treatments 6 (92±5%), 16 (86±10%), 17 (94±3%), 15 (85±17%), which only received rain water. Irrigated plot 10 (78±13%) had a significantly lower %V than plot 17 (94±3%) that received only rain water. Fertilized plots 19 (86±18%) and 20 (89±9%) had a higher %V than plot 8 (64±11%). Cutting effect was observed on treatments 6 to 17 (Table 1). On 2 September, the cutting plot 7 had a significantly different %V and %F than the control 2 (respectively %V: 45±30% vs 98±2% and %F: 55±30% vs 3±2%). Early cut %V and %F was significantly lower in treatments 10, 11, and 16 (respectively %F: 98±1%, 97±1%, 98±1%, and %V: 3±1%, 4±1%, 2±1%), than late cut treatments 12 and 13 (respectively %V: 71±23%, 56±23% and %F: 29±23%, 44±23%). On 12 October, irrigated at mid-July and cut 5 cm above ground plot 13 had a lower %V than plot 7 not irrigated and cut low to the ground (%V: 1±1% vs 6±4%). Irrigated at mid-July and cut at 5 cm above ground Plot 14 (%S 6±7%) had a lower %S than plots 11 and 9 both irrigated at mid-July and cut low to the ground (respectively %S: 47±25%, 47±27%). Treatment 11 (%F 53±25%) had a significantly lower %F than 14 (%F 94±6%).

These results show that at the beginning of the season irrigation induces a reduction of the %V of the irrigated treatments. Indeed, the irrigated plots started to flower faster than the others. The sensitivity of the herbaceous layer to the variation of water resources has been noted in the literature (Richard *et al.*, 2019), particularly at the beginning of the season (July) by Diawara *et al.*, (2020) in Burkina Faso. Also, fertilization associated or not with irrigation leads to an increase of more than 20% of the %V. During the

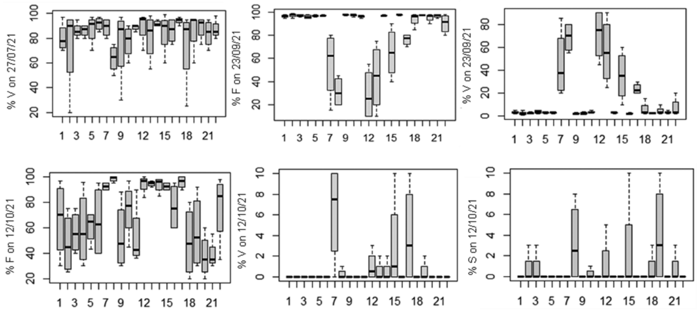

Figure 1. Average percentage of the vegetative stage (%V), flowering stage (%F), and/or senescence (%S) stage at different herbaceous vegetation monitoring dates at CRZ Dahra; x-axis refers to the treatments in Table 1.

rainy season the effect of irrigation and fertilization disappeared and the cutting period, in particular, had a significant effect on the %V and %F. At this period, rainfall become frequent and enough to satisfy the water needs of all plants. Plants in the cut plots resuming their cycle while those in the non-cut treatments continue theirs. At the end of the season, in addition to the effect of the cutting period on the %V and %F, an effect of the cutting height, coupled or not with irrigation, appeared on vegetative, flowering and senescence. The influence of cutting height and timing on grass regrowth, and thus to resume their cycle, was noted by Klein *et al.* (2013).

Conclusions

This study shows that the effect of fertilization, irrigation and cutting on phenology differs according to phenological stage and rainfall. At the start of the rainy season, before cutting, irrigation has a negative effect on leaf growth. During the season, when the rains become regular and satisfy the plant requirements this effect disappears. Only the effect of cutting is expressed until the end of the season. This study shows that at the beginning of the season the phenology can be influenced by water resources and quantity of soil organic matter. However, when the rainfall becomes enough to satisfy the needs of the plants this effect disappears.

References

Akpo L.E., Banoin M. and Grouzis M. (2003) Effet de l'arbre sur la production et la qualite fourrageres de la vegetation herbacee: bilan pastoral en milieu sahelien. *Revue de Médecine Vétérinaire* 154(10), 619-628.

Diawara M.O., Hiernaux P., Sissoko S., Mougin E., Ba A., Soumaguel N. and Diakité H.S. (2020) Sensibilité de la production herbacée aux aléas de la distribution des pluies au Sahel (Agoufou, Mali): une approche par modélisation. *International Journal of Biological and Chemical Sciences* 14, 1341-1353.

Diop A.T. (2007) Dynamique écologique et évolution des pratiques dans la zone sylvopastorale du Sénégal: perspectives pour un développement durable. *Faculté des Sciences Economiques et de Gestion (FASEG)* 296.

Klein H.-D., Rippstein G., Huguenin J., Toutain B. and Guerin H. (2013) *Les cultures fourragères*. éditions Quae, Paris, France.

Richard D., Alary V., Corniaux C., Duteurtre G. and Lhoste P. (2019) *Dynamique des élevages pastoraux et agropastoraux en Afrique intertropicale*. éditions Quae, Paris, France.

Variability of multispecies grasslands production in a diversified agroecological dairy system

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Abstract

The objective of the study was to evaluate over 2017-2020 the grassland production and the profile of the cattle diet in a mixed crop-dairy system named OasYs. Conducted without irrigation and with limited inputs, it was designed with an agroecological approach to permit farmers to live from their dairy system in a context of climate change, while preserving environment and contributing to animal welfare. The forage system is based on multispecies grasslands and on the grazing of several forage resources. Grasslands are diversified in terms of composition (mixtures of different species and cultivars), management (from 100% grazed to 100% cut) and age (1 to 5 years). Their yields ranged from 2.2 to 12.9 t dry matter ha^{-1} year^{-1} and were greater (1) for cut versus grazed and cut grasslands; and (2) for grasslands containing chicory. Over 2017-2020, herbage (grazed and conserved) represented 28 to 100% of the monthly diet of the dairy herd. Other forages were grazed fodder beet or other grazed crops and silages of sorghum, maize or of cereal-legumes mixtures. This diversification allowed the system nearly reaching self-sufficiency in forages and in protein.

Keywords: mixed crop-dairy system, temporary grasslands, agroecology, low-input, OasYs

Introduction

Grassland-based dairy systems present several assets both environmentally and economically (Delaby *et al.*, 2020). Their capability to provide enough forage to feed a dairy herd in the context of climate change may yet be challenged, especially if droughts increase in frequency and intensity. Achieving self-sufficiency in forage with grass seems all the more difficult in dairy systems aiming to limit the use of external inputs and irrigation. In this context, the diversification of composition, management and age of grasslands, coupled with the diversification of complementary forage resources seems promising to attain self-sufficiency in forages. This diversification of forage resources and a new livestock breeding strategy are tested in the climate-smart agroecological dairy cattle system 'OasYs' (Novak *et al.*, 2018). We present here the grazed and/or cut grasslands production per year at the farm level, and the profile of the dairy cattle diet at a monthly scale over the 2017-2020 period.

Materials and methods

The OasYs dairy cattle system has been carried out since June 2013 in a plain area already affected by summer droughts (mean total summer rainfall of 134 mm over 2017-2020), located south of the French leading dairy regions (Lusignan, Nouvelle Aquitaine, France) in an INRAE facility. The forage system (91.5 ha) aims to produce the fodder necessary to feed the dairy cattle herd (72 milking cows, and replacement heifers) without irrigation and with limited use of mineral nitrogen (N) fertilizer and pesticides. Forage resources are diversified in terms of species, cultivars, age and management, and 2/3 of the area is accessible for grazing (Novak *et al.*, 2018). Five- and four-year multispecies and legume-rich grasslands (52 ha, 18 plots on average) represent the heart of the forage system, complemented by annual crops (mainly maize, grain sorghum, cereal-legumes mixtures, fodder beet) that are cut or grazed. The grassland included tall fescue, cocksfoot, perennial and annual ryegrass, lucerne, chicory, plantain, sainfoin, and various perennial and annual clovers. Only 27 of the 71 plot.year modalities in grasslands were fertilized with organic or mineral N, half of them receiving less than 50 kg N ha^{-1} year^{-1}. The livestock breeding strategy is in coherence with the forage system, with two calving periods in spring and autumn, lactation lengths of 16 months and three-way crossing of dairy breeds (Holstein, Scandinavian Red, and Jersey) (Novak *et al.*, 2018). The amounts of herbage grazed by the dairy herd were evaluated at paddock level for each rotation over 2017-2020 by the Herbvalo method (Delagarde *et al.*, 2018). The amounts of the non-herbage forage crops (e.g. fodder beet) grazed were estimated by linking weekly measurements of the crop yields with the daily grazed area and herd size. Intake of conserved forages fed in the barn was measured daily.

Results and discussion

The growing season (March to November, mean temperature of 14.9 °C) was characterized by a high variability of its total amount of rainfall, ranging from 384 mm in 2020 to 778 mm in 2019. Grazed and/or cut grassland yields ranged from 2.2 to 12.9 t dry matter (DM) ha^{-1} $year^{-1}$ over the 2017-2020 period (Table 1), and averaged 6.6, 6.3 and 7.9 t DM ha^{-1} $year^{-1}$ respectively for grazed, grazed and cut, and cut grasslands. They were statistically higher for: (1) cut versus grazed-and-cut grasslands; and (2) grasslands containing chicory (7.9 compared to 6.0 t DM ha^{-1}). Forage chicory had already been shown to produce large quantity of high quality feed, especially in warm and drought conditions (Li and Kemp, 2005). The great intra-annual variability of yields may be due to quality of seedling establishment, fertilization, age of the grassland, its composition and soil nutrient status. These annual yields are in the same order of magnitude compared to local references for temporary grasslands, which averaged 7.2 t DM ha^{-1} $year^{-1}$ during this period and were generally more highly fertilized (Agreste, 2020). The large use of legumes in sown grasslands certainly explains these results.

Over the 2017-2020 period, herbage (grazed and conserved) averaged 70% of the annual diet of the dairy cattle herd and ranged from 28 to 100% at a monthly time scale (Figure 1). Grazed herbage was the main forage in the diet during April, May and June. Other grazed forage resources were mainly fodder beet, grazed from summer to winter, and various intercrops including either grass intercrops or crop residues and re-growths of cereal-legumes mixtures and grain sorghum after their silage. Annual crops such as mixtures of rape and turnip, and grain sorghum associated to lablab, were also grazed. Conserved forage proportion ranged from 0 (in May) to 95% (in January) in the monthly diet with an annual average value of 49%. They were mainly composed of herbage (on average 6% of hay and 26% of silage over the year), followed by silages of sorghum (7%), maize (6%) or cereal-legumes mixtures (5%). Concentrates proportion ranged from 0 to 10%, representing only 4% of the average annual diet, on a DM basis, and 47 g per litre of milk. The annual average of fat and protein corrected milk production ranged from 6,468 to 6,978 litres per cow.

Table 1. Grazed and/or cut grassland yields per year at the system level (t DM ha^{-1}).[1]

	Grazed grasslands		Grazed and cut grasslands		Cut grasslands	
	Mean	Min-max	Mean	Min-max	Mean	Min-max
2017	6.8	4.2-8.4	5.8	2.6-7.4	7.7	5.6-11.6
2018	6.6	2.2-12.9	6.8	3.3-9.2	8.6	6.0-12.9
2019	7.3	4.0-10.8	6.7	5.9-7.8	8.7	5.7-11.7
2020	5.8	2.7-11.5	5.8	5.0-7.3	6.9	5.8-7.6
mean	6.6[ab]		6.3[b]		7.9[a]	
SD	2.6		1.5		2.1	
n	26		22		23	

[1] Letters indicate significant differences (P<0.05) according to pairwise t-tests. SD = standard deviation.

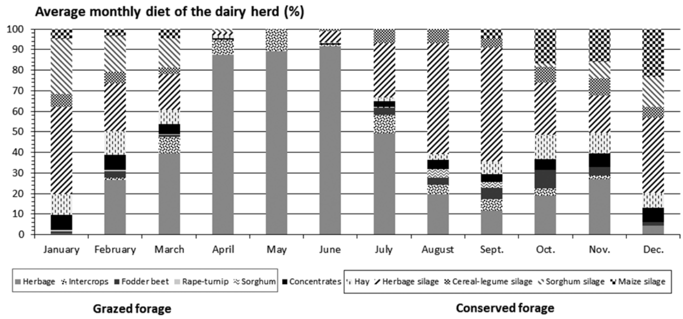

Figure 1. Profile of the dairy cattle diet at a monthly scale over the 2017-2020 period.

Conclusions

The multispecies and legume-rich temporary grasslands allowed good forage production levels whatever the year and the management (grazing or cutting), over the 2017-2020 period, despite the low N fertilization. Chicory was useful to extend grazing season, especially during dry periods. Thanks to the diversification of grazed or conserved forage resources, including mainly multispecies grasslands but also grazed annual crops or intercrops, and to a large use of legumes, the low-input dairy cattle system OasYs nearly reached self-sufficiency in forages and protein.

Acknowledgements

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References

Agreste (2020) Conjonctures Prairies. DRAAF Nouvelle-Aquitaine. Available at: https://draaf.nouvelle-aquitaine.agriculture.gouv.fr/Rendements-annuels-des-prairies

Delaby L., Finn J.A., Grange G. and Horan B. (2020) Pasture-based dairy systems in temperate lowlands: challenges and opportunities for the future. *Frontiers in Sustainable Food Systems* 4, 543587.

Delagarde R., Robic,Y., Leurent-Colette S. and Delaby L. (2018) HerbValo – a method for calculating annual pasture utilisation by dairy cows at paddock level. *Grassland Science in Europe* 23, 968-970.

Li G.D. and Kemp P.D. (2005) Forage chicory (*Cichorium intybus* L.): A review of its agronomy and animal production. *Advances in Agronomy* 88, a187-222.

Novak S., Chargelègue F. and Emile J.C. (2018) An agroecological dairy cattle system to graze all year round. *Grassland Science in Europe* 23, 473-475.

Evaluating some winter forage legumes under Mediterranean rain-fed conditions

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Abstract

Maximizing quantity and quality of home-grown forages is a timeless goal for ruminant production farming systems. Climatic conditions in semi-arid Mediterranean areas set a limit to this goal. With the aim to evaluate a range of forage legumes a trial was set up at a site in Western Peloponnese, where forage legumes were sown in a randomized design with three replicates. Forage species sown were: *Trifolium alexandrinum* (cv Mario), *Trifolium dasyurum* (cv Sothis), *Trifolium pratense* (cv Zoja), *Trifolium incarnatum* (cv Alberobello), *Trifolium michelianum* (cv Vista), *Trifolium subterraneum* (cv Dalkeith, Campeda), *Hedusarum coronarium* (cv Sulla), *Biserulla pelecinus* (Cashbah), *Ornithopus sativus* (Margurita), *Ornithopus compressus* (Santorini) *Medicago polymorpha* (cv Scimitar). Quantity and quality characteristics (ash, crude protein, crude fibre as well as NDF and ADF contents) of the forage produced were recorded. It was observed that *T. incarnatum* (cv Alberobello) and *T. michelianum* (cv Vista) were the most productive in forage DM, but *M. polymorpha* (cv Scimitar) was competitive against weeds. Forage nutritional characteristics were similar between species. It was concluded that productivity and competitiveness of a forage species (or cultivar) is a crucial factor for the production of rain-fed forage material of adequate quality under Mediterranean conditions.

Keywords: forage legumes, rain-fed crops, Mediterranean conditions, nutritional quality

Introduction

Livestock production in Mediterranean areas is based on dairying with small ruminants and it contributes greatly to the income of rural populations, while providing local societies with a range of ecosystem services, including prime food production, support of biodiversity, protection of water and soil resources, recreational values, carbon sequestration (Varela and Robles-Cruz, 2016). Despite their reliance on the extensive rangelands for feeding, due to the Mediterranean climate, these animals are also fed cereal straw and alfalfa hay cropped locally, as well as grain (Porqueddu *et al.*, 2017). However, although alfalfa provides good quality forage, it grows under irrigation in the area and this adds to cost and environmental burdens. Leguminous forages present significant potential to ensure agronomic sustainability and mitigate the negative environmental effects of intensification through diversification of the systems (Porqueddu *et al.*, 2017). Although knowledge of the cultivation and utilization of leguminous forages has increased greatly in temperate regions and new species and advanced varieties have been produced (Rochon *et al.*, 2004), information is largely lacking on growing forage legumes efficiently in Mediterranean conditions, particularly on practical subjects and on their integration into farming systems (Ates *et al.*, 2013). A comparative study was undertaken to test, under the pedoclimatic conditions of south-western Greece (Region of Ilia), conventional and new forage legume germplasm of Mediterranean origin. Alfalfa is the major forage legume crop in the area, while rain-fed crops are represented only by berseem clover (*T. alexandrinum*) though it is rarely found in the area. Therefore all other winter growing species were new ones and were compared with berseem clover.

Materials and methods

A comparative test of sown forage legumes was carried out the Region of Ilia in western Peloponesse (plot site: N 37°40'34'', E 21°23'12'', altitude 5 m a.s.l.). The study had a randomized design, with three replicates, and included 12 legume forage species and varieties thereof. The plant species tested (cultivar in parenthesis) were: *Trifolium alexandrinum* (cv Mario), *Trifolium pratense* (cv Zoja), *Trifolium incarnatum* (cv Alberobello), *Trifolium michelianum* (cv Vista), *Trifolium subterraneum* (cv Dalkeith, Campeda), *Trifolium dasyurum* (cv Sothis), *Hedusarum coronarium* (cv Sulla), *Biserulla pelecinus* (Cashbah), *Ornithopus sativus* (Margurita), *Ornithopus compressus* (Santorini) and *Medicago polymorpha* (cv Scimitar). Plots were 2.25 m^2 in area (1.5×1.5 m), separated by corridors of 0.5 m, and were sown on 30/11/2020, with a density of about 250 seeds m^{-2}, after fine surface soil preparation. No fertilization, herbicides or irrigation was applied on this trial. Forage material was harvested in June 2021 from an area of 0.5 m^2 at the centre of each plot. Cut herbage was separated into sown and spontaneous vegetation and dried at 60 °C for 48 hours. Forage quality was assessed on the sown dried material after it was ground in a hammer mill to pass a 6 mm sieve and then finely ground on a laboratory mill through a 1 mm screen. Analyses were conducted for moisture (AOAC method 930.15), crude protein (CP) by the Kjeldahl method (AOAC method 984.13), ash content (Ash) by ashing overnight at 550 °C (AOAC method 942.05), crude fibre (CF), neutral detergent fibre (NDF) and acid detergent fibre (ADF) on an ANKOM 220 (AOAC 978.10, AOAC 2002:04 and AOAC 973.18, respectively). Data were subjected to a one-way Analysis of Variance (ANOVA) and Least Significant Difference (LSD) at 0.05 level was applied to define significantly different means among the species. The Statgraphics Centurion 16 statistical package was used for this task.

Results and discussion

The soil on the site was characterized as a light type (LS), with low organic matter (1.98% OM) and low Total N (0.25%), light alkaline (pH: 8.1) and medium in Ca content (15.2% $CaCO_3$). Climatic conditions in the area are mild with a yearly mean air temperature of 19. °C, an absolute low of 0.5 °C and an absolute max of 42.7 °C. Precipitation is 832 mm $year^{-1}$ (values are 2 years averages) though very erratic within a year. In fact during the study months temperatures ranged from above zero up to 34 °C, following a normal pattern for the area, but precipitation was abundant (>250 mm $month^{-1}$) during December and January, to drop at 50 mm in February and March, while April and May received less than 10 mm each thus creating severe water deficit.

Herbage DM produced by each of the 12 legume forages varied widely between species. *Trifolium incarnatum* (cv Alberobello), *Trifolium michelianum* (cv Vista) were the most productive among species, while *Ornithopus sativus* (Margurita), *Ornithopus compressus* (Santorini) were the least productive (Table 1). Herbage nutritional characteristics also varied widely between species. CP content was higher for *Trifolium michelianum* (cv Vista) and *Trifolium subterraneum* (cv Campeda) while *Trifolium dasyurum* (Sothis) had the lowest (Table 1). An opposite pattern was observed for CF, NDF and ADF contents. The different legume forages matured at different times, while at sampling time not all species crops were at full maturation, therefore such differences are naturally expected.

Conclusions

Several of the legume forages tested produced forage appropriate for hay of good quality, suitable for the feeding of dairy small ruminants, although at a wide range of productivities. It is advisable to educate farmers cultivating new species in an effort to shift their crops to those that are more productive and to confront the adverse effects of climate change through the use of fast growing species that are more tolerant of high temperatures.

Table 1. Legume forage DM production and the crude chemical composition (Ash, crude protein, crude fibre and the fibre fractions (NDF, ADF), for the species tested (g 100 g^{-1} of DM).

Species and varieties	Herbage ($g DM m^{-2}$)	Ash (% DM)	CF (% DM)	NDF (% DM)	ADF (% DM)	CP (% DM)
Biserulla pelecinus (Cashbah)	60.9^{cd}	8.4^{ab}	34.3^{ab}	50.2^{ab}	36.8^{abc}	9.9^{ab}
Medicago polymorpha (Scimitar)	356.3^{c}	10.1^{bcd}	38.2^{cd}	53.7^{bc}	44.1^{cd}	11.6^{cd}
Hedysarum coronarium (Sulla)	52.8^{a}	10.3^{cd}	35.4^{abc}	46.8^{ab}	33.8^{ab}	11.8^{cd}
Ornithopus compressus (Santorini)	0.0^{a}					
Ornithopus sativus (Margurita)	20.9^{ab}	12.5^{cde}	33.2^{ab}	38.3^{a}	27.6^{a}	13.1^{cde}
Trifolium alexandrinum (Mario)	382.1^{c}	11.0^{cde}	34.4^{ab}	45.6^{ab}	32.8^{ab}	12.5^{cd}
Trifolium dasyurum (Sothis)	147.7^{ab}	7.4^{a}	39.2^{d}	61.8^{c}	47.7^{d}	8.9^{a}
Trifolium incarnatum (Alberobello)	572.9^{d}	9.8^{bc}	36.0^{bc}	54.8^{ab}	44.9^{bcd}	11.3^{bc}
Trifolium michelianum (Vista)	451.8^{cd}	13.0^{e}	34.7^{ab}	48.0^{ab}	36.5^{abc}	14.5^{e}
Trifolium subterraneum (Campeda)	271.4^{bc}	12.5^{de}	34.9^{ab}	42.8^{a}	30.1^{a}	14.3^{de}
Trifolium subterraneum (Dalkeith)	148.6^{ab}	10.7^{cd}	32.4^{a}	50.1^{ab}	33.2^{ab}	12.2^{c}
Trifolium pratense (Zoja)	81.5^{ab}	9.9^{bcd}	33.9^{ab}	41.6^{a}	30.6^{a}	11.5^{cd}
Standard error	21.0	0.21	0.36	1.19	0.92	0.21
P-value	0.0001	0.0001	0.009	0.027	0.003	0.0002

References

Ates S., Feindel D., El Moneim A. and Ryan J. (2013) Annual forage legumes in dryland agricultural systems of West Asia and North Africa Regions: research achievements and future perspective. *Grass and Forage Science* 69, 17-31.

Porqueddu C., Melis R.A.M., Franca A., Sanna F., Hadjigeorgiou I. and Casasus I. (2017) The role of grasslands in the less favoured areas of Mediterranean Europe. *Grassland Science in Europe* 22, 3-22.

Rochon J.J., Doyle C.J., Greef J.M., Hopkins A., Molle G., Sitzia M., Scholefield D. and Smith C.J. (2004) Grazing legumes in Europe: a review of their status, management, benefits, research needs and future prospects. *Grass and Forage Science* 59, 197-214.

Varela E. and Robles-Cruz A.B. (2016) Ecosystem services and socio-economic benefits of Mediterranean grasslands. *Options Mediterraneennes, Serie A*: *Mediterranean Seminars* 114, 13-27.

Overyielding in multi-species swards under simulated grazing management

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Abstract

Species from different functional groups when grown together can produce higher biomass yield compared to the respective monoculture yields of component species, a phenomenon known as overyielding. This paper reports on yield effects from a multi-species sward (MSS) plot study in Northern Ireland and reviews likely explanations of overyielding. Two contrasting seed mixtures were sown in replicated plot trials in 2019, along with their component species as monocultures. *NoGrass* contained chicory, plantain, white clover and red clover. *GrassMix* contained tall fescue, cocksfoot, timothy, late heading perennial ryegrass and white clover. *NoGrass* produced a higher dry matter yield (P<0.05) in 2021 than all of its components, with the exception of the red clover component (12.2 t dry matter (DM) ha^{-1}; red clover 11.2 t DM ha^{-1}). *GrassMix* also exhibited overyielding for dry matter production compared with its component varieties. Further research is needed to explain this overyielding phenomenon in MSS.

Keywords: multi-species, yield, overyielding, seed mixtures

Introduction

Multi-species swards (MSS) could make an important contribution to sustainable livestock farming systems. Various studies have shown that mixtures of species with differing characteristics can produce greater yield than either the weighted average of the respective monocultures of the component species (Finn *et al.*, 2013; Sanderson *et al.*, 2004) or the best-performing monoculture. Explanation of such overyielding could be partly due to the respective use of resources, above and below ground characteristics of the component species as well as the effects of symbiotic nitrogen fixation (Suter *et al.*, 2015) in mixtures of functional groups of species such as grasses, legumes and herbs. The aim of this study was to investigate overyielding in MSS plot trials in Northern Ireland.

Materials and methods

The study, which lasted for 28 weeks in each of 2 consecutive years at AFBI Loughgall (54°27′N, 6°04′W), was composed of 3 replicates of 16 treatments comprising 8 treatments of clover and herb monocultures and 1 clover/herb mixture (*NoGrass*= chicory, plantain, white and red clover) and 8 treatments of grass monocultures and 1 multiple grass species mixture (*GrassMix*= tall fescue, cocksfoot, timothy, late heading perennial ryegrass and white clover). Two fertilizer regimes were applied: *NoGrass* and clover and herb monocultures received an early March application only (83 kg ha^{-1} N; 88 kg ha^{-1} P_2O_5; 71 kg ha^{-1} K_2O); *GrassMix* and grass monocultures received the equivalent March application and a June application (total of 155 kg ha^{-1} N; 88 kg ha^{-1} P_2O_5; 110 kg ha^{-1} K_2O) to reflect typical industry practice for grass-based swards. P_2O_5 and K_2O fertilizer was applied to maintain soil P and K at target Index 2, as per RB209 soil nutrient index values (AHDB, 2021). Plots were managed under a simulated grazing regime, with 8 sampling dates for herbage yield during the growing season, taken from early April to late October each year with harvest dates determined by sward height (target of 15 cm above ground level). Herbage was defoliated to 75 mm stubble height using a plot harvester (Haldrup, F55) and regrowth intervals ranged from 18 to 32 days. Fresh weight yields were recorded in 2020 and 2021 (not presented), with dry matter yields calculated in 2021 only. Analysis of Variance was applied to assess the effects of variety and mixture on yield using Genstat (VSNi, 2017). Means were separated by Fisher's least significant difference (LSD) test at $P<0.05$.

Results and discussion

Grange *et al.* (2021) has described the effect by which mixtures can produce higher yield than any of their constituent components as transgressive overyielding. In 2021, *NoGrass* produced higher dry matter (DM) yield ($P<0.05$) than all of its components (Figure 1), with the exception of red clover variety A (*NoGrass* 12.2 t DM ha^{-1}; red clover A 11.2 t DM ha^{-1}); Chicory had the lowest yield in 2021 (6.2 t DM ha^{-1}). *GrassMix* also exhibited overyielding for DM yield in 2021 (Figure 2), yielding 11.0 t DM ha^{-1} compared to an average of 8.7 t DM ha^{-1} for the component varieties which is in keeping with the study of Vojtech *et al.* (2008), which showed that mixtures of grass species with differing foliar architecture can improve overall light interception and biomass production compared with monocultures. Overall, these results demonstrate that transgressive overyielding can occur when contrasting species are grown together in herb/legume and in grass/legume combinations. That *NoGrass* was the highest yielding treatment with only two functional groups present could suggest that combining species with contrasting traits may be more important than the number of functional groups present. Asynchrony in shoot growth of component species has been considered to be more important for yield than mixing species with contrasting foliar architecture (Husse, 2016). By contrast, Lorenz *et al.* (2020) found that increasing species diversity did not increase yield, as some species were replaced by others with similar functional traits. It is also noteworthy that *NoGrass* out-yielded *GrassMix* and all of the grass monocultures despite the slightly higher fertilizer N applied (83 and 155 kg ha^{-1} N respectively), which is in keeping with Grange *et al.* (2021), for whom higher fertilizer N perennial ryegrass monocultures (300 kg ha^{-1} N) yielded less than the most species-diverse mixture.

Conclusions

In general, legume-herb and grass-legume species combinations yielded more than their components grown as monocultures. It would appear that functional group composition could be more important than the number of species present for overyielding to occur. The factors controlling overyielding are not fully understood but asynchrony of growth patterns may be more important than contrasting foliar architecture, which will require further research to establish. Further research is also required to verify the productivity of diverse mixtures particularly under both conservation and grazing conditions, and at varying fertilizer rates, to more fully understand their yielding characteristics, along with yield stability and herbage quality in order to capture the full potential of MSS for sustainable livestock farming systems.

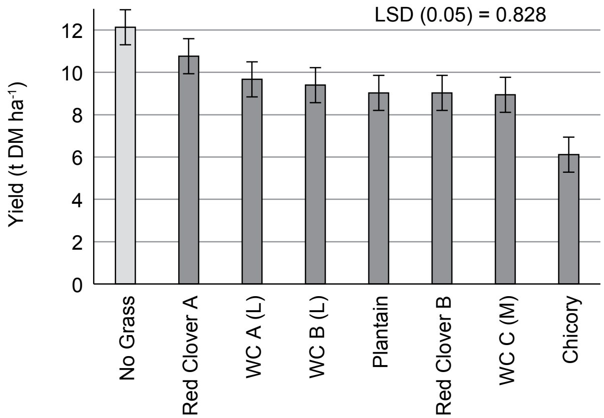

Figure 1. Dry matter yield (t DM ha^{-1}) of *NoGrass* and component varieties. WC = white clover; L = large leaved; M = medium leaved; PRG = perennial ryegrass; Int = intermediate maturity; Late = late maturity; LSD = least significant difference.

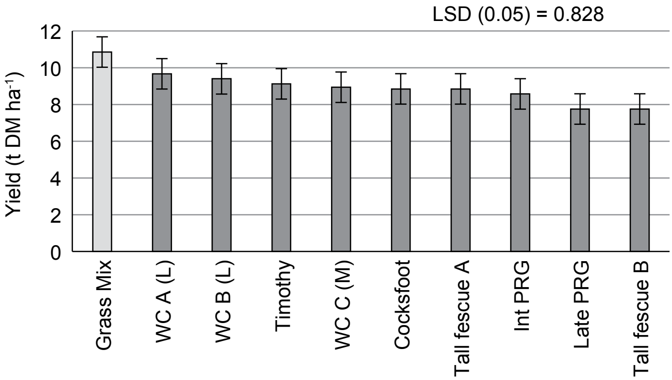

Figure 2. Dry matter yield (t DM ha^{-1}) of *GrassMix* and component varieties. WC = white clover; L = large leaved; M = medium leaved; PRG = perennial ryegrass; Int = intermediate maturity; Late = late maturity; LSD = least significant difference.

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References

AHDB (Agriculture and Horticulture Development Board) (2021) Nutrient Management Guide (RB209) Section 3 grass and forage crops. Available at: https://ahdb.org.uk/knowledge-library/rb209-section-3-grass-and-forage-crops

Finn J.A., Kirwan L. ... and Connolly J. (2013) Ecosystem function enhanced by combining four functional types of plant species in intensively managed grassland mixtures: a 3-year continental-scale field experiment. *Journal of Applied Ecology* 50, 365-375.

Grange G., Finn J.A. and Brophy C. (2021) Plant diversity enhanced yield and mitigated drought impacts in intensively managed grassland communities. *Journal of Applied Ecology* 58(9), 1864-1875.

Husse S. (2016) Larger yields of mixtures than monocultures of cultivated grassland species match with asynchrony in shoot growth among species but not with increased light interception. *Field Crops Research* 194, 1872-6852.

Lorenz H., Reinsch T., Kluβ C., Taube F. and Loges R. (2020) Does the admixture of forage herbs affect the yield performance, yield stability and forage quality of a grass clover ley? *Sustainability* 12(14), 5842. https://doi.org/10.3390/su12145842

Sanderson M.A., Skinner R.H., Barker D.J., Edwards G.R., Tracy B.F. and Wedin D.A. (2004) Plant species diversity and management of temperate forage and grazing land ecosystems. *Crop Science* 44, 1132-1144.

Suter M., Connolly J., Finn J.A., Loges R., Kirwan L., Sebastià M.T. and Lüscher A. (2015) Nitrogen yield advantage from grass–legume mixtures is robust over a wide range of legume proportions and environmental conditions. *Global Change Biology* 21, 2424-2438.

Vojtech E., Loreau M., Yachi S., Spehn E.M. and Hector A. (2008) Light partitioning in experimental grass communities. *Oikos* 117, 1351-1361.

VSN International (2017) Genstat for Windows 19th Edition. VSN International, Hemel Hempstead, UK.

Comparison of milk production of Holstein cows grazing perennial ryegrass or multispecies swards

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Abstract

Grazing swards for dairy herds in Ireland are predominantly perennial ryegrass mixtures (*Lolium perenne* L). Recently, there has been increasing interest in diversifying pasture composition for commercial dairy systems. This study compared the milk production of two groups (n=20) of Holstein cows grazing either a ryegrass-dominated sward (LP) or a diverse sward (DS), containing *Lolium perenne, Phleum pratense, Trifolium repens, Trifolium pratense, Chichorium intybus*, and *Plantago lanceolate*. The LP and DS swards were established on separate farmlets in autumn 2019, and the experiment was carried out across the full grazing season (Feb to Nov) of 2020. Grazing was managed on a rotational basis, with target pre-grazing herbage dry matter (DM) (kg DM ha^{-1}), post-grazing residual height (cm), and herbage allowance (kg DM cow^{-1}), used to make daily decisions. Farm cover (herbage DM kg ha^{-1}) and grazing rotation interval dictated weekly sward management decisions. Milk data were analysed using week of lactation as a repeated measure. No significant differences (P>0.1) in milk volume (kg^{-1}), fat or protein content (g kg^{-1}) were detected across lactation. Post grazing residuals were similar for the treatments. Further work is required to elucidate annual herbage yields and species persistency within the swards.

Keywords: diverse swards, milk production, grazing dairy cattle

Introduction

In Ireland, the predominant feeding system for dairy cattle involves grazing ryegrass-dominant sward across a long grazing season (Feb to Nov), with moderate supplementary concentrate and conserved silages fed during lactation. This system has typically involved use of in excess of 200 kg ha^{-1} of chemical N applied annually (McCarthy *et al.*, 2010). Given national and EU policy objectives to reduce greenhouse gas (GHG) emissions from agricultural sources however, strategies to reduce chemical N input while optimizing stocking rate and animal performance are required. While systems involving ryegrass-white clover (*Trifolium repens*) swards offer clear benefits (Egan *et al.*, 2018), the potential for additional functionality of diverse swards involving grass-legume-herb mixes has received comparatively less focus, particularly for intensive dairy grazing systems. Grange *et al.* (2021), in a plot cutting experiment, showed that the combination of herb, grass and legume functional groups delivered highest yields, both under rain-fed control and experimental drought conditions. In a two-year grazing experiment in France, an increase of botanical complexity from one to five species (two grasses, two clovers and chicory) resulted in positive effects on animal performance (Roca-Fernández *et al.*, 2016). Our objective therefore was to compare the annual milk production performance of dairy cattle managed on a grazing system based on diverse swards, relative to a ryegrass-based grazing system.

Materials and methods

The study was conducted at Teagasc Johnstown Castle Co. Wexford, Ireland (52.29' N, 6.49' W; 53 m a.s.l.). Soil type at this site is a free draining acid brown earth of loam to clay loam texture. Mean annual rainfall during the experimental period was 1,150 mm (minimum 17 mm per month and 325 mm cumulative rainfall during peak growing season May to Aug inclusive); mean annual soil temperature (10 cm) was 10.6 °C (exceeding 5.5 °C from Mar to Nov inclusive). In autumn 2019, two farmlets of were configured in 8 paddock subdivisions and treatment swards established on each; LP containing 95%

(by seed weight) of a perennial ryegrass (*Lolium perenne* L.) diploid-tetraploid mix (2:1) with 5% white clover; and DS, containing 60% of the same perennial ryegrass mix, 12% timothy (*Phleum pratense*), 16% white clover, 4% red clover (*Trifolium pratense)*, 4% chicory (*Chicorium intybus,* and 4% plantain (*Plantago lanceolate).* Swards were established by a standard plough-till-sow method, and were grazed at least once post-emergence before winter closing.

In spring 2020, 40 spring-calving Holstein cows were blocked according to parity, calving date (median 19^{th} Feb) and genetic merit for milk yield, and assigned to either the LP or DS sward treatments, resulting in a farmlet stocking rate (SR) of 2.60 cows ha^{-1}. Grazing was managed as a standard rotational system, with cows receiving a target daily herbage allowance to achieve a target post grazing sward height of 4.0 cm in the initial spring rotation, and 4.5 cm in subsequent rotations. A rising plate meter was used to estimate pre-grazing herbage mass and post grazing residuals. Surplus herbage accumulations were removed as silage from the farmlets. A cereal-pulp based concentrate was offered in the milking parlour to offset daily feed deficits to a maximum 5 kg DM d^{-1}, with silage offered to meet larger daily deficits. The LP treatment received 220 kg ha^{-1} of chemical N fertilizer annually while the DS treatment received 96 kg ha^{-1} annually, this differential arose due to four N fertilizer applications of 31 kg ha^{-1} being omitted from DS during May to late August. Milk yield was recorded daily, while milk composition was recorded weekly. Pre- and post-grazing herbage masses were recorded for each grazing, with herbage growth and mean herbage cover ha^{-1} measured weekly for each farmlet. Milk and herbage mass data were analysed as repeated measures using the MIXED procedure in SAS.

Results and discussion

Annual lactation milk yield was similar for the LP and DS treatments, in terms of milk volume and compositional quality (Table 1). The pattern of milk solids production across lactation was also similar (Figure 1). The DS treatment received a marginally greater daily concentrate supplementation rate during early spring due to lower initial herbage availability on the farmlet; however, this was not statistically significant on a cumulative annual basis.

Pre-grazing herbage mass did not differ significantly between LP and DS across the growing season; this was a function of within-farmlet rotation length management as well as weekly growth rate. Mean post-grazing residual was similar for the sward treatments within experimental week or across lactation. This indicates that the grazing efficiency (proportion of grown herbage that is utilized by the animal) of grass-legume-herb mixed swards compares well with ryegrass swards when managed under an intensive grazing decision rules. Nonetheless, the persistency of the individual species within the sward under such management conditions remains to be fully evaluated, and work is ongoing in this regard.

Table 1. Milk yield and sward production outcomes for ryegrass and diverse sward systems.[1]

	LP	DS	Sig
Annual milk kg cow^{-1}	6,694	6,712	NS
Milk fat content g/kg^{-1}	43.0	42.5	NS
Milk protein content g/kg^{-1}	36.2	36.0	NS
Milk solids (fat+ protein) yield kg	530	527	NS
Annual herbage t DM ha^{-1}	15.0	13.9	0.12
Pre-grazing sward mass kg DM ha^{-1}	1,538	1,506	NS
Post grazing residual height cm	4.62	4.18	NS
Concentrate fed kg DM cow^{-1}	790	847	0.15

[1] LP = 95% perennial ryegrass; DS = diverse sward containing grass, legumes and herbs

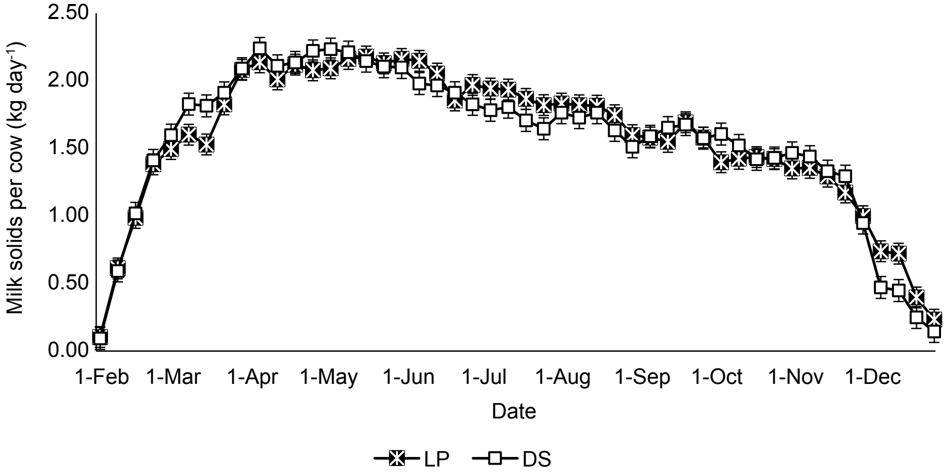

Figure 1. Daily milk solids (fat plus protein) yield for cows grazing ryegrass (LP) or diverse species (DS) swards across lactation.

Annual herbage production from the DS sward was 7% lower compared to the LP sward, albeit for 124 kg ha^{-1} less chemical N applied. In the context of the systems compared, this was manifest as a reduction in conserved forage production rather than a reduction in herbage utilized by grazing. The DS sward did not provide sufficient conserved forage from within the farmlet to meet annual requirements, whereas the LP farmlet exceeded its annual forage requirement. This may have implications for determination of optimal stocking rate for DS systems, though multi-annual results are required before definitive conclusions can be drawn.

Conclusions

Holstein cows grazing grass-legume-herb swards, produced similar milk yield of similar compositional quality to cows grazing perennial ryegrass-dominated swards. The efficiency of herbage utilization did not differ between the swards. The diverse sward system produced less total annual herbage yield; however chemical N input was also reduced. Further work is warranted to clarify grazing intensity effects on species persistency within the diverse sward, and also to compare swards' performance across a series of N fertilizer application rates.

References

Egan M., Galvin N. and Hennessy D. (2018) Incorporating white clover (*Trifolium repens* L.) into perennial ryegrass (*Lolium perenne* L.) swards receiving varying levels of nitrogen fertiliser: Effects on milk and herbage production. *Journal of Dairy Science* 101, 3412-3427.

McCarthy B., Delaby L., Pierce K.M., Journot F. and Horan B. (2010) Meta-analysis of the impact of stocking rate on the productivity of pasture-based milk production systems. *Anima*l 5, 784-794.

Roca-Fernández I., Peyraud J.L., Delaby L. and Delagarde R. (2016) Pasture intake and milk production of dairy cows rotationally grazing on multi-species swards. *Animal* 10, 1448-1456.

Diversity mitigates overwintering damage due to prolonged snow cover during ley establishment

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Abstract

A prolonged snow cover is an unpredictable event in mountain regions of the Alps. This can have detrimental effects on newly established leys, resulting in damage to sown species. To explore the effect of diversity in mitigating such damage during establishment, we used the data set from a field experiment set up in the mountain environment of South Tyrol (NE Italy). Six forage species belonging to three different functional groups were combined in different proportions according to a simplex design. Following extensive damage due to a prolonged persistence of the snow cover during the first winter, strong losses of *Plantago lanceolata* and, to a lesser extent, of *Trifolium repens* and *Trifolium pratense* occurred. The yield proportion of non-sown species was used to explore the effects of diversity of sown species on weed establishment and growth. Increasing diversity was found to reduce the yield proportion of weeds, as shown by species-specific convex functions and further decreases caused by most pairwise interactions between sown species. No significant change of the yield proportion of non-sown species was observed depending on the growth cycles.

Keywords: overwintering damages, seed mixtures, leys, diversity effects, mitigation

Introduction

Winters in mountain regions of the Alps can be severe and long, and snow cover may last for several weeks to months. This unpredictable event can promote the establishment of pathogens and have detrimental effects on newly established leys, resulting in damage and losses of forage species, thus adversely affecting forage production and forage quality in the following years. It is known that diversity positively affects yield production of leys (Nyfeler *et al.,* 2009), but it is also relevant to explore whether diversity also affects robustness and resilience overwintering damage.

Materials and methods

To explore the effect of diversity in mitigating the consequences of overwintering damage, we used the data from a one-year field experiment in the mountain environment of South Tyrol (NE Italy, 11°57'25"E, 46°14'8"N, 892 m a.s.l.) set up on 9 September 2020 following winter rye. Six forage species belonging to three different functional groups, grasses, legumes and forbs (*Lolium perenne* = Lp, *Dactylis glomerata* = Dg, *Trifolium repens* = Tr, *Trifolium pratense* = Tp, *Plantago lanceolata* = Pl, *Cichorium intybus* = Ci), were combined in different proportions according to a simplex design (Kirwan *et al.*, 2009), including three monospecific plots of each species, each possible combination of two equally represented species (50% of each species), of three species (33% of each species) excluding the mixtures with more than one species belonging to the same functional group, of four species (25% of each species) excluding the mixtures with only one species belonging to the same functional group and three replicates of the centroid (17% of each species). Proportions were determined based on the locally adopted seed rates of these species for the establishment of monospecific swards (25 kg ha^{-1} for Lp, 20 kg ha^{-1} for Dg, Tr and Tp, and 10 kg ha^{-1} for Ci and Pl). Treatments were randomly assigned to plots of 7×3 m. After an unusually prolonged persistence of the snow cover (until 6 March 2021), extensive fungal infection with *Typhula incarnata* on *T. repens* and *T. pratense* was visually assessed on 23 March, (41.7 and 17.5% incidence on average in monospecific plots respectively), as well as apparent lack of vital signs of almost the whole aboveground plant parts of *P. lanceolata* (later, at the time of the first harvest, the yield proportion of monospecific plots of *P. lanceolata* was 26% on average). The experiment was fertilized with 30 kg ha^{-1} mineral nitrogen in spring and with 11.8 m^3 ha^{-1} of digested slurry after the first cut. Three harvests (on 1 June, 12 July and 24 August) were performed. Prior to each cut, the yield proportion of non-sown species (NSYP) was visually assessed in each plot. We used NSYP as dependent variable to evaluate the effects of diversity on weed establishment and growth and thus on mitigating the consequences of overwintering damage through competition against weeds. Data analysis was performed by means of mixed models accounting for the growth cycle (treated as a factor with repeated measurements with the plots as a subject), the species identity, and exploring the interactions patterns according to Kirwan *et al.* (2009). The data were arcsine-transformed to meet the ANOVA assumptions. The model selection was based on the Akaike's Information Criterion (AIC) using Maximum Likelihood as estimation method.

Results and discussion

Among all tested diversity-interaction patterns, the best model fit (AIC = 832.018) was achieved by the model accounting for the identity effects of the sown species and including also all pairwise interactions (Table 1).

Pl, Dg, and Tr exhibited the highest coefficients (55.98, 30.52 and 26.34), indicating a stronger increase of NSYP than for the other species. Indeed, Pl and Tr were the two species apparently most affected by the overwintering damage. For Dg, a possible explanation for this fact might be due to its tussock growth form, leading to a less dense turf if sown at high proportion; this in turn allowed weed establishment from

Table 1. Parameter estimates for modelling NSYP accounting for the growth cycle, the identity effects of the sown species and all pairwise interactions.[1]

Parameter	Estimate	Std. Error	*t*-value	*P*-value	95% Confidence interval	
					Lower bound	Upper bound
Growth cycle	0.12	0.518	0.23	0.820	-0.92	1.16
Lp	14.14	1.499	9.43	<0.001	11.14	17.13
Ci	16.32	1.499	10.88	<0.001	13.32	19.31
Tp	19.62	1.499	13.08	<0.001	16.62	22.61
Tr	26.34	1.499	17.57	<0.001	23.35	29.34
Dg	30.52	1.499	20.36	<0.001	27.53	33.52
Pl	55.98	1.499	37.33	<0.001	52.98	58.97
Tr×Pl	-107.91	8.563	-12.60	<0.001	-125.13	-90.68
Tp×Pl	-75.77	8.563	-8.85	<0.001	-93.00	-58.54
Dg×Pl	-57.26	8.563	-6.69	<0.001	-74.48	-40.03
Dg×Tp	-53.60	8.563	-6.26	<0.001	-70.83	-36.38
Dg×Tr	-52.36	8.563	-6.11	<0.001	-69.59	-35.13
Lp×Pl	-44.04	8.563	-5.14	<0.001	-61.26	-26.81
Lp×Tr	-43.98	8.563	-5.14	<0.001	-61.21	-26.76
Pl×Ci	-40.45	9.387	-4.31	<0.001	-59.34	-21.57
Lp×Ci	-33.76	8.563	-3.94	<0.001	-50.99	-16.54
Lp×Dg	-30.84	9.387	-3.29	0.002	-49.72	-11.96
Dg×Ci	-25.88	8.563	-3.02	0.004	-43.11	-8.66
Tp×Ci	-17.26	8.563	-2.02	0.050	-34.49	-0.03
Lp×Tp	-7.70	8.563	-0.90	0.373	-24.92	9.53
Tr×Tp	5.43	9.387	0.58	0.566	-13.45	24.32
Tr×Ci	10.31	8.563	1.20	0.235	-6.92	27.53

[1] Analysis was done with arcsine-transformed data.

the soil seed bank. However, significant positive coefficients were observed for all species identity effects. When back-transformed on the original scale, they all correspond to convex quadratic functions (e.g. for Pl: NSYP = -1.6177 + 22.872 x + 49.286 x^2, where x is the sown proportion of Pl; for Tr: NSYP = -0.0925 + 1.2823 x + 18.611 x^2, where x is the sown proportion of Tr; for Tp: NSYP = -0.029 + 0.4011 x + 10.936 x^2, where x is the sown proportion of Tp). Such functions indicate a positive diversity effect reducing NSYP, especially at a low to intermediate sown proportion. Moreover, most of the pairwise interactions (all but Lp×Tp, Tr×Tp and Tr×Ci) showed a further relevant effect in reducing NSYP, as shown by the negative coefficients, which were particularly high for most of the interactions involving Pl and Dg.

No further fit improvement could be achieved by more parsimonious models accounting (besides growth cycle and species identity) for evenness, additive species-specific contributions to interactions or functional groups effects (Kirwan *et al.*, 2009), possibly because of the species-specific damages having altered the planned proportions between species and between functional groups.

The growth cycle had no significant effect in any of the models explored. This suggests that the mitigation of overwintering damage in form of competition against weeds had already taken place during the first growth cycle and no further improvement occurred thereafter.

Conclusions

The results provide evidence that diversity of sown species effectively allows some mitigation of the consequences of overwintering damage on ley establishment.

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References

Kirwan L., Connolly J., Finn J.A., Brophy C., Lüscher A., Nyfeler D. and Sebastià M-T. (2009) Diversity–interaction modeling: estimating contributions of species identities and interactions to ecosystem function. *Ecology* 90, 2032-2038.

Nyfeler D., Huguenin-Elie O., Suter M., Frossard E., Connolly J. and Lüscher A. (2009) Strong mixture effects among four species in fertilized agricultural grassland led to persistent and consistent transgressive overyielding. *Journal of Applied Ecology* 46, 683-691.

Effect of climate change on forage production at plot/farm level – a case study in Vosges (France)

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Abstract

In 2020, the territory of Deodatie (south-west of the Vosges massif) launched an assessment of the vulnerability of livestock systems to climate change. Twelve farms representative of the diversity of pedoclimatic conditions and forage systems (proportion of grassland in forage areas, level of intensification of production) formed the focus for this study. The forage production of the 315 plots (grasslands, maize) on these 12 farms were simulated over the historical period 1990-2020, and over the period 2021-2100. The simulations were carried out using the STICS model developed by INRAE, and considering the different types of grasslands of the Vosges massif characterized by the recent agroecological typology work 2017-2020. The results of these simulations show that the effects of climate change differ not only between grassland and maize, but also between different types of permanent grassland. Positive or negative impacts of climate change was simulated, either on annual forage yield and its variability. Increasing future forage resilience on farms therefore requires the maintenance of a diversity of forage resources, including a diversity of permanent grassland types.

Keywords: grassland, forage, climate change, typology

Introduction

Adaptation to climate change has become a key issue for the sustainability of forage systems in Europe. Droughts and heat waves are becoming more frequent, posing a threat to fodder production. On the other hand, the warming of the temperature in cold areas, and the associated increase in the atmospheric CO_2 content, are likely to stimulate production. A recent worldwide synthesis of 256 studies on grasslands concluded that, according to warming scenarios, an increase in yield was simulated by models either in 50% of cases (+2 °C in 2100) or only in 10% of cases (+4 °C in 2100) (Li *et al.*, 2018). Effects have also been reported on the functional diversity of grasslands (Cantarel *et al.*, 2013). It is, however, important to assess the consequences at both the plot and farm level. The objective of this work was to assess the consequences of climate change considering two climatic scenarios and the type of forage resource. The production of all the plots of 12 farms in the Vosges mountains (North Eastern France) was simulated over the period 1990-2100.

Materials and methods

Twelve farms were chosen to represent the forage diversity of the Deodatie region of the Vosges mountains (350-1000 m a.s.l.). These livestock holdings (dairy and beef cattle) mainly use permanent grasslands (78% UAA), but also temporary grasslands (12%) and maize silage (10%). Forage production of 315 plots was simulated with the STICS™ model, developed for crops (Brisson *et al.*, 2003), and recently adapted to grasslands (Graux *et al.*, 2020; Ruget *et al.*, 2008). For the parameterization of the model, we used the French national soil database developed by INRAE, and the climatic information of Météo-France. Two IPCC climate scenarios were selected (Representative Concentration Pathway 2.6 and 8.5) as extreme situations. The climatic data, available at a scale of 8×8 kms, were corrected for each plot considering its altitude, slope and orientation. Based on the typology of permanent grasslands in the Vosges massif (Mesbahi *et al.*, 2018; Plantureux *et al.*, 2021), 12 different types of permanent grassland were parameterized in the model, changing the grass growth and development parameters.

These simulations make it possible to calculate the daily production of each plot between 1990 and 2100, and therefore the annual dry matter yields. Interannual variability in production was assessed by comparing the coefficient of variation (standard deviation/mean) for the years 2070-2100 compared to 1990-2020. These productions were then aggregated at the farm level, considering the respective areas.

Results and discussion

In general, the annual yields of forage plots evolve differently over time depending on the type of production (grassland or maize), the type of permanent grassland, and the climate scenario (Figure 1).

In climate scenario 2.6 (dotted lines), the evolution of yields by 2100 shows either a decrease (5 types of grassland, e.g. type CP2) or remains static or shows a slight increase (7 types, e.g. type CF 5). The yield of temporary grassland and maize increases by 10%. The interannual variability of yields changes little, but it is greater for maize (CV 16%) than for temporary grassland (10%) and permanent grassland (8%).

In climate scenario 8.5 (solid lines), grassland yields increase by 10 to 20% for 7 types of permanent grassland (e.g. type CF5) and for temporary grasslands, and remain static or decreases for 5 types of permanent grassland and maize (-5%). Interannual variability evolves in a comparable trend to scenario 2.6.

At farm level, these simulations resulted in an overall maintenance of the quantities of fodder produced, but with an increase in interannual variability in climate scenario 8.5. Climate scenario 8.5 generally allows for an increase in fodder production, but some farms experience a reduction in their total forage production.

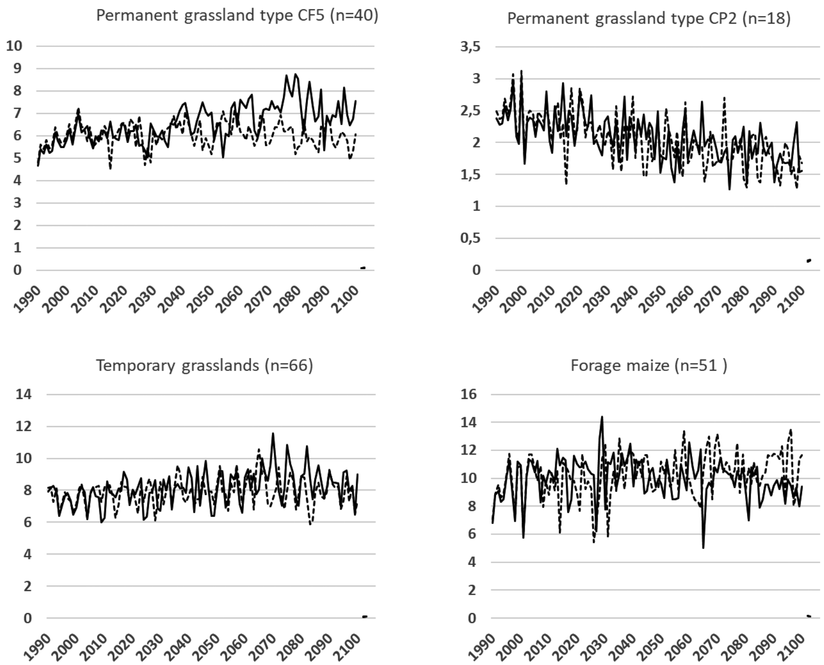

Figure 1. Simulation of annual dry matter forage production for 4 types of forage types, and for 2 IPCC scenarios (RCP2.6 dashed lines – RCP8.5 solid lines).

Any simulation model is based on assumptions, including the values chosen for the input parameters. We do not know the level of accuracy of climate change forecasts by 2100, which leads to uncertainty in the outputs of the models. In terms of fodder production, there is also the question of the ease of harvesting grasslands. Indeed, if warmer springs and autumns are expected, they will also be rainier, sometimes making it difficult or impossible to access the plots.

The results of maintaining or even increasing yields in 2100 can be explained by: (1) the fact that the Vosges mountains are experiencing a cold climate where warming will be favourable to plant growth; (2) compensation for the effects of heat waves and summer droughts by the more favourable springs and autumns, and higher CO_2 concentrations. In scenario 8.5, maize with a summer production cycle does not benefit from this compensation.

Conclusions

The main result of this simulation work is to show that the effects of climate change differ not only between grassland and maize, but also between different types of permanent grassland. No type of forage resource is evolving in the same direction, and future forage resilience of farms therefore requires the maintenance of a diversity of forage resources, including a diversity of permanent grassland types. Additional studies are underway to test not only technical adaptation scenarios (intensification, extensification, new forage crops), but also the economic consequences of these production forecasts.

References

Brisson N., Gary C., Justes E., Roche R., Mary B., ... and Sinoquet H. (2003) An overview of the crop model stics. *European Journal of Agronomy* 18, 309-332. https://doi.org/10.1016/S1161-0301(02)00110-7

Cantarel A.A.M., Bloor J.M.G. and Soussana J.-F. (2013) Four years of simulated climate change reduces above-ground productivity and alters functional diversity in a grassland ecosystem. *Journal of Vegetation Science* 24, 113-126. https://doi.org/10.1111/j.1654-1103.2012.01452.x

Graux A.-I., Resmond R., Casellas E., Delaby L., Faverdin P. ... and Peyraud J.-L. (2020) High-resolution assessment of French grassland dry matter and nitrogen yields. *European Journal of Agronomy* 112, 125952. https://doi.org/10.1016/j.eja.2019.125952

Li W., Li X., Zhao Y., Zheng S. and Bai Y. (2018) Ecosystem structure, functioning and stability under climate change and grazing in grasslands: current status and future prospects. *Current Opinion in Environmental Sustainability* 33, 124-135. https://doi.org/10.1016/j.cosust.2018.05.008

Mesbahi G., Bayeur C. and Plantureux S. (2018) Prediction of grassland values by phytosociological and agronomical approach. *Grassland Science in Europe* 23, 571-573.

Plantureux S., Mesbahi G. and Bayeur C. (2021) *Les prairies permanentes du massif des Vosges*, PNR Vosges du Nord. ed.

Ruget F., Godfroy M. and Plantureux S. (2008) Improved estimation of mountain grassland production by using local input data in a crop model. *Grassland Science in Europe* 13, 305-308.

Increased mineral soil N availability contributes to post-drought yield outperformance of *Lolium perenne*

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Abstract

Reoccurring drought events severely restrict forage production. However, intensively managed grasslands have recently been reported to recover quickly after drought stress and to even outperform control yields. Despite several studies showing increased mineral N availability after drought, the contributions of the two N sources: (1) fertilizer-derived N accumulated during drought; and (2) increased availability of soil-derived mineral N, remain unclear. Thus, we examined the effect of a 2-month experimental summer drought and two fertilization levels during drought (non-fertilized and fertilized) on the recovery of *Lolium perenne* swards after rewetting. Even for non-fertilized swards, dry matter yield (DMY) and plant-available N of drought and rewetted (DRW) plots exceeded controls. Fertilization during drought increased the effects of DRW on DMY and on plant available N. Consequently, our study shows that formerly drought stressed swards surpass control yields by profiting from higher N availability, not only deriving N from accumulated fertilizer N but also from increased availability of soil-derived N due to changed soil processes.

Keywords: grassland, drought, yield outperformance, ryegrass, N availability, resilience

Introduction

Drought events during the past years have severely restricted forage production and the frequency of drought events is predicted to keep increasing (Emadodin *et al.*, 2021; Gharun *et al.*, 2020). However, temperate grasslands have recently been reported to show outstanding resilience after experimental drought and rewetting (DRW) and to be even more productive after DRW than non-drought stressed control plots (Hahn *et al.*, 2021; Hofer *et al.*, 2016). Hofer *et al.* (2017) reported higher plant foliar N concentrations after DRW of *L. perenne*, suggesting higher availability of mineral N after rewetting. But their experiment was not able to quantify the contributions of: (1) fertilizer N accumulated during drought; and (2) of the increased availability of N from mineralization of soil organic matter (hereafter soil-derived N). Thus, this study aims to disentangle the contributions of fertilizer derived N and soil-derived N on post-drought yield outperformance of perennial ryegrass (*L. perenne*).

Materials and methods

To test whether yield outperformance after DRW was driven by fertilizer-derived N accumulation during drought and/or by increased availability of soil-derived N, a field experiment was set up in the vicinity of Zurich on an intensively managed ryegrass (*L. perenne*, cv 'Allodia') ley. To simulate summer drought, rain-out shelters were set up for two months on half of the plots. After shelter removal, all plots were re-watered including the controls. Plots were harvested and dry matter yield (DMY) was determined six weeks after shelter removal and rewetting to assess yield resilience. To distinguish fertilizer from soil nutrient effects on DMY, half of the plots were fertilized during drought with 47.5 kg ha^{-1} ammonium nitrate (27% N), and the other half was not fertilized.

For measuring plant available nitrogen in the soil, we used PRS (Plant Root Simulator) ion-exchange membranes (Western AG, Saskatoon, Canada) which mimic soil nutrient sorption by plant roots (Qian and Schoenau, 1994). To avoid confounding effects from potential competition of PRS with plants for nutrient uptake, PRS were installed in plant exclusion cylinders (Huang and Schoenau, 2011). After 4 weeks of incubation they were removed, washed and sent back to Western AG for measuring plant available N concentrations.

Statistical analysis of DMY and plant available soil N were performed using ANOVA with a model considering the interaction of drought treatment (control versus drought) and fertilization (without versus with). All statistical analyses were performed using R (version 4.0.2, 2020).

Results and discussion

Six weeks after rewetting we observed significantly higher DMY in formerly drought stressed plots, compared to control plots, for both plots without and with fertilization (DRW, $P<0.001$, Figure 1). Plots with fertilization showed higher DMY compared to plots without fertilization after DRW (fertilization, $P<0.001$) and fertilization increased the effect of DRW on DMY (drought stress × fertilization, $P<0.01$).

DRW resulted in higher plant-available soil N during the first recovery regrowth compared to control soil (DRW, $P<0.01$) in both, plots without and with fertilization during drought (Figure 2). Also, plots with fertilization during drought showed higher plant-available soil N (fertilization, $P<0.1$). The interaction of DRW and fertilization on available soil N was not statistically significant but shows a tendency towards a higher DRW effect in plots with fertilization compared to plots without fertilization.

Greater DRW effects of fertilized plots, compared to non-fertilized, for DMY, and a similar trend in plant-available N, suggest that yield overcompensation in plots with fertilization under drought occurred partially due to fertilizer N that had accumulated during drought stress. Remarkably, DRW resulted in higher yield and plant-available N not only in plots with fertilization during drought, but also in plots that were not fertilized during drought. Measuring plant-available soil N under plant exclusion allowed us to measure DRW effects of soil processes on the amount of soil-derived N without the effects of N uptake by the plants. Therefore, higher DMY and plant-available N after DRW in plots without fertilization during drought indicate that changed processes in the soil such as higher N-mineralization rates due to higher microbial activity (Borken and Matzner, 2009; Gordon *et al.*, 2008) increased the availability of soil derived N and led to the observed yield outperformance of DRW swards.

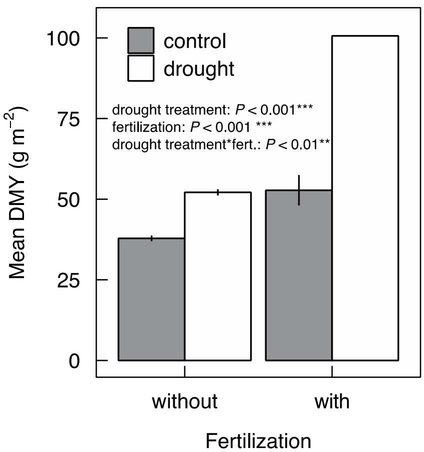

Figure 1. Dry matter yield (DMY) six weeks after release of drought for control and formerly drought-stressed plots with or without fertilization during drought (mean ± standard error, n=4).

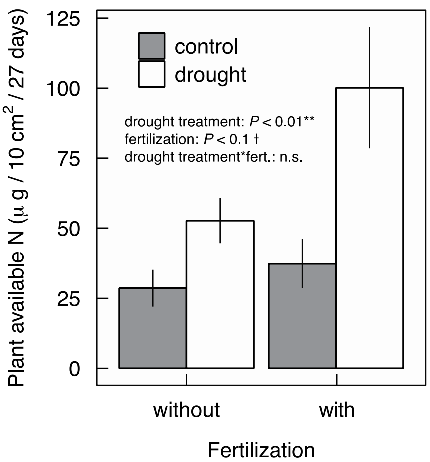

Figure 2. Plant-available N in the soil six weeks after release of drought, for control and formerly drought-stressed plots with or without fertilization during drought (mean ± standard error, n=4).

Conclusions

Formerly drought-stressed *L. perenne* swards were highly resilient and outperformed non-drought stressed controls not only in dry matter yield but also in showing higher plant-available total N in fertilized and non-fertilized swards. Thus, our study shows that the increased availability of total mineral N for plant growth after drought stress and rewetting derive from both the fertilizer N accumulated during drought where fertilizer was applied, and from increased availability of soil-derived mineral N.

References

Borken W. and Matzner E. (2009) Reappraisal of drying and wetting effects on C and N mineralization and fluxes in soils. *Global Change Biology* 15, 808-824.

Emadodin I., Corral D.E.F., Reinsch T., Kluß C. and Taube F. (2021) Climate change effects on temperate grassland and its implication for forage production: A case study from northern Germany. *Agriculture* 11, 232. https://doi.org/10.3390/agriculture11030232.

Gharun M., Hörtnagl L., Paul-Limoges E., Ghiasi S., Feigenwinter I., Burri S., Marquardt K., Etzold S., Zweifel R., Eugster W. and Buchmann N. (2020) Physiological response of Swiss ecosystems to 2018 drought across plant types and elevation. *Philosophical Transactions Royal Soc B* 375, 20190521. https://doi.org/10.1098/rstb.2019.0521.

Gordon H., Haygarth P.M. and Bardgett R.D. (2008) Drying and rewetting effects on soil microbial community composition and nutrient leaching. *Soil Biology and Biochemistry* 40, 302-311. https://doi.org/10.1016/j.soilbio.2007.08.008.

Hahn C., Lüscher A., Ernst-Hasler S., Suter M. and Kahmen, A. (2021) Timing of drought in the growing season and strong legacy effects determine the annual productivity of temperate grasses in a changing climate. *Biogeosciences* 18, 585-604. https://doi.org/10.5194/bg-18-585-2021.

Hofer D., Suter M., Buchmann N. and Lüscher A. (2017) Nitrogen status of functionally different forage species explains resistance to severe drought and post-drought overcompensation. *Agriculture, Ecosystems & Environment* 236, 312-322. https://doi.org/10.1016/j.agee.2016.11.022.

Hofer D., Suter M., Haughey E., Finn J.A., Hoekstra N.J., Buchmann N. and Lüscher A. (2016) Yield of temperate forage grassland species is either largely resistant or resilient to experimental summer drought. *Journal of Applied Ecology* 53, 1023-1034. https://doi.org/10.1111/1365-2664.12694.

Huang W.Z. and Schoenau J.J. (2011) Seasonal and spatial variations in soil nitrogen and phosphorus supply rates in a boreal aspen forest. *Canadian Journal of Soil Science* 77, 597-612. https://doi.org/10.4141/s97-002.

Qian P. and Schoenau J.J. (1994) Assessing nitrogen mineralization from soil organic matter using anion exchange membranes. *Fertiliser Research* 40, 143-148. https://doi.org/10.1007/bf00750099.

R Core Team (2020) R: A Language and Environment for Statistical Computing. Vienna, Austria.

Impact of drought stress and climate change on yield and forage quality of grassland

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Abstract

Climate change puts many grassland areas and their associated livestock farms under increasing pressure. Increased frequency and intensity of droughts will also adversely affect those climatic regions currently still favoured. To assess the impact of climate change on the overall grassland ecosystem, we compare the ambient climatic situation with future climate scenarios on permanent grassland. Therefore, we use a multifactor, multilevel approach on 54 plots with three cuts per year and standardized fertilization *(ClimGrass)*. It was established in 2014 and combines elevated temperature, CO_2 enrichment, and rainout shelter for drought stress simulations. The results of two drought experiments show a significant decrease in water availability during drought periods. We observe a reduction in yield under drought conditions for all treatments. The simulation shows a more severe drought effect under future climate conditions, resulting in a yield reduction of about 20% compared to the current climatic conditions. Research findings of this experiment serve as a basis for adaptation strategies for resilient grassland management.

Keywords: grassland, drought, climate change

Introduction

With grassland management adapted to site-specific environments, efficient land use is feasible in productive regions and Areas Facing Natural or other Specific Constraints (ANCs). However, the water requirement of grassland is comparatively high, so the annual precipitation in grassland marginal areas needs to be at least 700 to 800 mm (Forstner *et al.,* 2021). Climate change affects future weather, e.g. through a rise in temperature and more uneven rainfall, the risk of severe droughts increases, especially in already dry regions. Due to the comparatively poor water use efficiency of grassland, traditional grassland regions with sufficient rainfall in the past will be more vulnerable in the future (IPCC, 2014). The results of the field experiment *ClimGrass* at AREC Raumberg-Gumpenstein (47°29'38.2' N 14°06'03.0' E, 695 m a.s.l.; 8.5 °C, 1,077 mm; deep cambisol), where the impact of climate change on the grassland ecosystem is studied, show significant effects of climate change on plant composition, yield, phenological development and soil water balance (Pötsch *et al.*, 2019). In this paper, we present selected results of two drought experiments conducted on *ClimGrass* where we show how severe drought events affect yield and forage quality in comparison of current and future climate conditions.

Materials and methods

The sward on all *ClimGrass* plots consisted of about 85% grasses, 10% herbs, and 5% legumes. Dominant grass, legume, and herb species were *Dactylis glomerata, Trisetum flavescens, Arrhenatherum elatius, Festuca pratensis, Trifolium repens and Taraxacum officinale.* On 54 plots, each 16 m^2, of a free-air CO_2 enrichment (FACE) experiment the impact of the current climate is compared to a simulation based on the factors temperature and CO_2 concentration. In the middle of each plot, there is a fumigation ring on a height-adjustable frame, which supplies ambient air enriched with CO_2 at the canopy height and infrared radiators, which heat the irradiated surface. The CO_2 concentration measured on several reference plots (C0) is increased by +300 mg l^{-1} (C2), the reference temperature (T0) by +3.0 °C (T2) (treatments C0T0 and C2T2). While the temperature is applied all year when the snow cover is below a thickness of 10 cm, CO_2 is only added during the day within the growing season. On 12 plots equipped with dynamic rainout shelters controlled by rain sensors, water stress can be generated in combination with current and future climate (C0T0R and C2T2R). In the experimental years 2017 and 2020, we closed the rainout shelter for the entire second growth of the three-cut system. A subset of 15 plots was selected to evaluate drought impact with four replicates for the treatments C0T0, C0T0R, and C2T2R and three replicates for C2T2. We analysed the influence of the drought, the year, and their interaction using the glimmix procedure in SAS (Version 9.4, SAS Institute Inc.). A generalized linear mixed model was used to take care of the correlation of the longitudinal data.

Results and discussion

Drought stress under future climate conditions leads to significantly lower yields on the studied permanent grassland stands. Figure 1 shows the development of the climatic water balance (precipitation – evapotranspiration according to Penman-Monteith) for current climate and drought simulation from the start of the growing season to the harvest of the second growth with the corresponding yields of the different treatments for the selected year 2017.

While the dry matter yield in Figure 2 demonstrates the impact of drought, there are minor differences between the treatments regarding forage quality (crude protein). However, our experiments show a significantly lower lignin content (acid detergent lignin; ADL) in the drought-stressed treatments, which correspond to studies by Sanaullah *et al.* (2014). Lower Leaf Area Index values (measured with AccuPAR LP-80) in the drought-stressed treatments correspond to lower yields and, concerning the lower ADL values, indicate more stocky grassland stands with a reduced stem fraction.

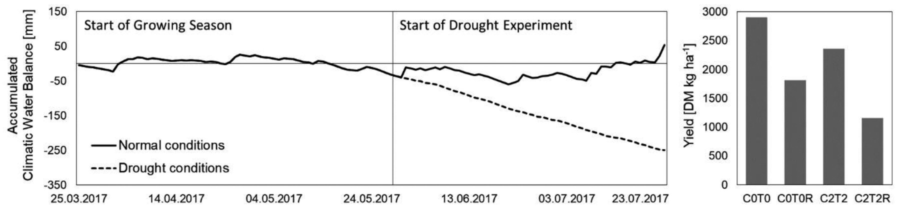

Figure 1. Accumulated climatic water balance for 2017 with corresponding yields of current (C0T0, C0T0R) and future climate (C2T2, C2T2R).

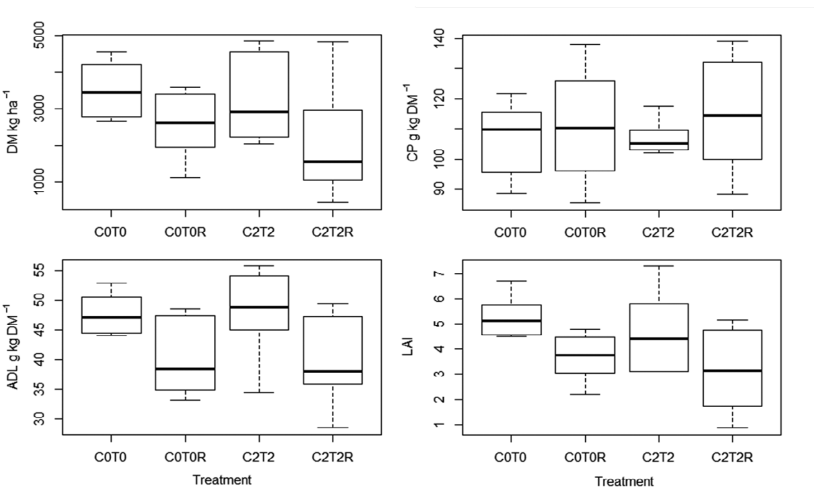

Figure 2. Dry matter yield, crude protein content, ADL content, and leaf area index (LAI) under current (C0T0, C0T0R) and future (C2T2, C2T2R) climate in comparison of ambient conditions and drought stress.

The simulated drought events in 2017 and 2020 are embedded in the long-term experiment that has been showing the effects of increased temperature and CO_2 concentration on permanent grassland since 2014. The results in this paper focus only on the direct effects of both drought events. However, we observed a general change of the sward in climate simulation treatments with a decrease of grasses in favour of herbs. This led to a reduction in aboveground biomass not only in the drought years but also in all other trial years, which is in the line of findings of Cantarel *et al.* (2013).

Conclusions

The increase in drought risk that comes with climate change leads to strong grassland yield fluctuations with significant regional differences. While forage quality remains stable even under dry conditions and digestibility may even be positively influenced due to the reduced lignin content, the decrease in yield results in a shortage of forage supply. Hence, timely adaptation strategies to climate change are crucial for preserving grassland management in particularly vulnerable regions. The adaptation of the species composition towards better drought tolerance as well as the use of a wider range of species are first steps to provide sustainable grassland yields under future conditions.

References

Cantarel A.A.M., Bloor J.M.G. and Soussana J.-F. (2013) Four years of simulated climate change reduces above-ground productivity and alters functional diversity in a grassland ecosystem. *Journal of Vegetation Science* 24 (1), 113-126.

Forstner, V., Groh J., Vremec M., Herndl M., Vereecken H., Gerke H.H., Birk S. and Pütz T. (2021) Response of water fluxes and biomass production to climate change in permanent grassland soil ecosystems. *Hydrology and Earth System Sciences* 25(12): 6087-6106.

IPCC (2014) Synthesis Report. Contribution of Working Groups I, II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. IPCC, Geneva, Switzerland, 151 pp.

Pötsch E.M., Herndl M., Schaumberger A., Schweiger M., Resch R. and Adelwöhrer M. (2019) Auswirkung zukünftiger Klimabedingungen auf Ertrag und Futterqualität im Grünland. 21. Alpenländisches Expertenforum zum Thema 'Klimawandel im Alpenraum – Auswirkungen auf das Ökosystem Grünland und dessen Bewirtschaftung', Gumpenstein, HBLFA Raumberg-Gumpenstein, 26.-27. März 2019, pp. 49-54.

Sanaullah M., Chabbi A., Girardin C., Durand J.-L., Poirier M. and Rumpel C. (2014) Effects of drought and elevated temperature on biochemical composition of forage plants and their impact on carbon storage in grassland soil. *Plant and Soil* 374(1-2), 767-778.

Responses of perennial ryegrass cultivars and their mixtures to white and red clovers as companion species in swards

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Abstract

The yield potential of swards depends on local climate, soil properties, grass and legume species and their varieties, number of species and varieties, and management options. The aim of this study is to understand possible interactions between perennial ryegrass cultivars and cultivars in mixtures, and growth prospectives in mixtures with clovers. The effects on yield of three tetraploid perennial ryegrass cultivars, 'Elena DS', 'Raminta', 'Verseka,' and these three in a cultivars-mixture, differed in a two-year experiment. The most productive, as a two-year average, was 'Elena' at 8 t ha^{-1} dry matter yield (DMY). The cultivation of legumes in mixtures with perennial ryegrass was compared with the effect of mineral N fertilizers in perennial ryegrass monoculture swards. Comparisons between the most productive single-species sward of perennial ryegrass with N_{150} and a mixture with white and red clover with N_0, revealed the DMY was higher for the mixture by 6%. Only trace amounts of white clover were detected in a mixture with perennial ryegrass and cultivars-mixture in the first year of sward use, and therefore the DMY was not increased significantly by these legumes. In summary, the results of the study suggest that a perennial ryegrass-clovers mixture could be used without additional N fertilizers because no significant differences were found compared with fertilized monoculture sward.

Keywords: legumes, nitrogen, monocultures, multi-species, yield-stability

Introduction

Grass-legume mixtures are usually higher yielding than unfertilized pure grass swards. The quantity and identity of species in a mixture are important for the mixture's productivity (Komainda and Isselstein, 2020; Wiering *et al.*, 2021). Cultivar-specific functional traits such as phenology or growth form affect the competitive ability and yield in mixtures (Prerostova *et al.*, 2021). Cultivars and cultivar-mixtures with different functional traits affect productivity and forage quality in mixtures. Little is known about functional traits of different cultivars and cultivar-mixtures. This research reinforces the knowledge about perennial ryegrass cultivars and cultivar-mixtures' ability to improve and maintain sward productivity and stability.

Materials and methods

An experiment was conducted at the Lithuanian Research Centre for Agriculture and Forestry. In spring 2018, three perennial ryegrass cultivars 'Elena DS', 'Raminta', 'Verseka' and a mixture of cultivars were sown in single-species swards and fertilized at 150 N kg ha^{-1} per year. Perennial ryegrass 'Elena DS' was selected in mixtures with white clover and also with white and red clover, and in cultivars-mixture with no additional fertilization. Each treatment was sown in a fully randomized design with four replicates. Pure seed rates of perennial ryegrass, white and red clover were 18, 10 and 15 kg ha^{-1}. In 2019, the first year of sward use, there were 4 cuttings, and in 2020 there were 5 cuttings with 35 days longer vegetation period than 2019. All cuts were taken depending on the predominant plant species – before perennial ryegrass flowering stage. The swards of the seed mixture treatments consisted of 40% legumes and 60% grasses. The soil in the experimental site was loamy Endocalcaric Epigleyic Cambisol (WRB, 2014); soil P and K availability were high, according to Lithuanian evaluation methods. During this experiment, dry matter yield (DMY) was measured at every cut (harvest) and individual plant species were separated for the evaluation of the botanical composition of the swards. For evaluation of yield, the above-ground biomass including unsown species in a subplot of ≥15 m^2 was cut to a height of 5 cm at each harvest. The samples were weighed to determine the fresh matter yield and samples then oven dried at 105 °C and dried samples were weighed to determine the dry matter weight and DMY.

Other samples were taken and dried at 65 °C for chemical analysis, and ground to pass through a 2 mm sieve. To analyse the effects of the treatment, an analysis of variance (ANOVA) was conducted. Significant differences between the experimental treatments were determined using Duncan's multiple range test at the 5% probability level (P<0.05).

Results and discussion

Comparisons of the differences in sward productivity between the single-species and multi-species swards revealed significant differences in DMY in both the first and the second year of the sward use (P<0.05). During the two years of the experiment, different dry matter yields were obtained depending on the botanical composition of the swards. In the first year of sward use there was no significant differences between productivity of single ryegrass cultivars and the ryegrass-cultivars mixture, similar to the findings of Tubritt *et al.* (2021). In the first year of sward use there were significantly higher DMY from single-species swards with different cultivars, and the cultivars-mixture of perennial ryegrass sward fertilized with N at an annual N_{150} rate, compared with mixtures that contained white clover. The perennial ryegrass cultivar 'Elena DS' and white clover mixture showed 24% lower yield, and the cultivars-mixture with white clover a 28% lower yield, compared with average yield of monocultures. Meanwhile, the yield of mixture of perennial ryegrass 'Elena DS' with white and red clover did not differ significantly from fertilized single-species swards.

Table 1. The effect of grass mixtures on the productivity, crude protein content, and botanical composition of each treatment in each year of the 2-year experiment.[1]

Treatments			Botanical composition, kg ha^{-1}						
	Total annual DMY, kg ha^{-1}	Crude protein % in DMY	Perennial ryegrass 'Elena DS'	Perennial ryegrass 'Raminta'	Perennial ryegrass 'Verseka'	Cultivars-mixture E+R+V	White clover	Red clover	Forbs
2019									
1 (N_{150})	7,320^{d}	11.8^{a}	7,253						68
2 (N_{150})	6,940^{bcd}	12.8^{ab}		6,628					312
3 (N_{150})	7,213^{cd}	12.1^{a}			7,150				63
4 (N_{150})	6,917^{bcd}	12.4^{a}				6,862			56
5 (N_0)	5,392^{a}	13.4^{abcd}	4,664				648		80
6 (N_0)	5,139^{a}	13.4^{abcd}				4,511	476		152
7 (N_0)	6,695^{bcd}	14.5^{bcd}	3,967				123	2,427	178
8 (N_0)	6,088^{ab}	14.7^{d}				3,940	66	1,783	300
2020									
1 (N_{150})	8,699^{bcd}	10.8^{a}	8,603						96
2 (N_{150})	6,999^{a}	11.0^{a}		6,935					64
3 (N_{150})	7,492^{ab}	11.4^{a}			7,327				166
4 (N_{150})	7,981^{ab}	10.9^{a}				7,890			92
5 (N_0)	7,423^{ab}	15.0^{b}	4,732				2,617		74
6 (N_0)	7,271^{ab}	15.2^{bcd}				3,934	3,251		86
7 (N_0)	10,308^{d}	16.2^{d}	3,681				732	5,802	92
8 (N_0)	8,243^{ab}	15.9^{bcd}				3,150	309	4,723	61

[1] Different combinations of letters indicate significantly different means in different years (P<0.05, Duncan's test).

The perennial ryegrass cultivar 'Elena DS' here gave higher yield than a three cultivar-mixture grown together with legumes; the yield with white clover, and white and red clover, was higher by 6 and 9% in the first year, and 2 and 20% in the second year of sward use. These results are consistent with results reported in the literature (Lowry *et al.*, 2020). In the second year of sward use the most productive sward was the perennial ryegrass 'Elena DS' mixture with white and red clover: this was 1,610 kg ha^{-1} higher yielding than the most productive monoculture sward which was fertilized. No significant differences were found between other treatments. These two swards were the only treatments that differed significantly in dry matter yield compared with other treatments.

Further analysis showed that, in both experimental years, herbage from the grass-legume mixtures contained significantly more crude protein than that of the other four single-species swards (significant at $P=0.05$). Assessing the average dry matter yield of the two experimental years, the mixture of perennial ryegrass 'Elena DS' with white clover plus red clover was the most productive and had a 6% higher dry matter yield than the most productive fertilized single-species sward of perennial ryegrass 'Elena DS'.

Conclusions

This study has shown that reduction of mineral-N fertilizer use on swards is possible by selecting suitable legumes that, in association with perennial ryegrass cultivars and their mixtures, can help maintain good sward productivity and improve forage quality. Increasing the proportion of legume plants in the sward will contribute to the nitrogen supply of perennial ryegrass and reduce the need for N-fertilization.

References

Komainda M. and Isselstein, J. (2020) Effects of functional traits of perennial ryegrass cultivars on forage quality in mixtures and pure stands. *Journal of Agricultural Science* 158(3), 173-184.

Lowry C.J., Bosworth S.C., Goslee S.C., Kersbergen R.J., Pollnac F.W., Skinner R.H., Warren N.D. and Smith R.G. (2020) Effects of expanding functional trait diversity on productivity and stability in cultivar mixtures of perennial ryegrass. *Agriculture, Ecosystems and Environment* 287, 106691.

Prerostova S., Zupkova B., Petrik I., Simura J., Nasinec I., Kopecky D., Knirsch V., Gaudinova A., Novak O. and Vankova R. (2021) Hormonal responses associated with acclimation to freezing stress in *Lolium perenne*. *Environmental and Experimental Botany* 182, 104295.

Tubritt T., Delaby L., Gilliland T.J. and O'Donovan M. (2021) The agronomic performance of perennial ryegrass varieties evaluated as monocultures and mixtures of varying complexity under animal grazing. *European Journal of Agronomy* 124, 126226.

Wiering N.P., Ehlke N.J., Catalano D., Martinson K. and Sheaffer C.C. (2021) Forage potential of winter-hardy perennial ryegrass populations in monoculture and binary alfalfa mixture. *Agronomy Journal* 113: 5183-5195.

WRB (2014) *World reference base for soil resources*. World Soil Resources Reports No. 106. FAO, pp. 187-189.

Multispecies grass-legume swards productivity and reducing nitrogen fertilization

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Abstract

Multispecies grass-legume swards are an important component in agroecosystems. Fertilization of swards has been identified as the major driver for increase of productivity, but also has negative effects on the environment. In our experiment, four species of grasses (Poaceae) and four legume (Fabaceae) plants that are suitable for local growth conditions were used. Single-species swards and grass-legume mixtures were grown in the experimental plots with different plant species and different numbers of plants. Two fertilization rates N_0 and N_{150} kg ha^{-1} yr^{-1} were used. A two-year experiment showed an interaction between different species composition and the use of mineral N fertilizers. The most productive sward was in the high-diversity sward with six sown plant species, the two-year average dry matter yield was 10% higher than the single-species sward with ×*Festulolium* grass, which was fertilized at N_{150} rate. Fertilizers significantly increased the yield of monoculture swards and the yield of other grasses in mixtures but reduced the yield of legumes in the mixtures with reduced amount of crude protein. This study shows that mineral N fertilizers are not always necessary in the swards of different species compositions. Results of this study make a major environmental contribution linked to an economic and ecological approach.

Keywords: biodiversity, environment, grassland, multi-species

Introduction

In recent decades, the issue of environmental pollution has become more relevant, encouraging deepening of scientific discussions, and searching for new solutions to this problem (Suter *et al.*, 2015). There is a growing need to study the inter-species compatibility and productivity of legume and grasses in the agroecosystems due to the possibility to reduce the use of mineral nitrogen fertilizers. Excessive concentrations of nitrogen fertilizers threaten soil fertility, surface and groundwater quality and human health, and contribute to increasing greenhouse gas emissions into the atmosphere (Barneze *et al.*, 2020). Therefore, measures to reduce the negative externalities arising from the production and use of nitrogen fertilizers are increasingly sought. The swards of various species composition in this study used grass-legume mixtures, the effects of which were compared with the use of mineral nitrogen fertilizers.

Materials and methods

The sward productivity measurements were realized in 2019 and 2020, at the Lithuanian Research Centre for Agriculture and Forestry (Akademija 55°22′59.7″ N; 23°51′42.1″ E). This experiment aimed to compare the performance of different types of grass-legume mixtures, with and without N-fertilizers, in a two-year experiment. The experimental design included eight plant species of perennial ryegrass, ×*Festulolium*, meadow fescue, timothy, white clover, red clover, lucerne and sainfoin. Seeding rates were 18, 18, 20, 12, 10, 15, 15 and 80 kg pure live seeds ha^{-1}, respectively. Ten grass-legume mixtures consisted of three, four, six and eight plant species and two monoculture swards were sown. The legume/grass ratio in the sown mixtures was 40:60 (Table 1). The experiment was arranged in a randomized design with four replicates per treatment.

The year of sowing was 2018, prior to which NPK fertilizers were applied at a rate of N-P-K 5-20.5-36 kg ha^{-1}. No pesticides or herbicides were used, only three cleaning cuts on the fields. In 2019 and 2020,

Table 1. The types of swards used in the experiment; the numbers in the columns indicate the targeted proportion of the species at sowing.

Treatment	Number of grass sp.	Number of legume sp.	Perennial ryegrass (G1)	×*Festulolium* (G2)	Meadow fescue (G3)	Timothy (G4)	White clover (L1)	Red clover (L2)	Lucerne (L3)	Sainfoin (L4)
1	1	0	100							
2	1	0		100						
3	1	2	60				20	20		
4	1	2		60			20	20		
5	2	2	30	30			20	20		
6	4	2	15	15	15	15	20	20		
7	2	2	30	30			20		20	
8	4	2	15	15	15	15	20		20	
9	2	2	30	30			20			20
10	4	2	15	15	15	15	20			20
11	2	4	30	30			10	10	10	10
12	4	4	15	15	15	15	10	10	10	10

60 kg fertilizer N ha^{-1} was applied in spring, and 45 kg N ha^{-1} after 1st and 2nd harvests. In the two-years of the experiment there were 4 and 5 harvests, in the 1st and 2nd years, respectively. Grass and legume above-ground biomass were harvested and separated, with dry matter content determined by oven drying at 105 °C for 24 h. Subsamples were dried at 60 °C for 24 h, then ground and analysed for crude protein content of forages. The soil of the experimental site was loamy Endocalcaric Epigleyic Cambisol (WRB, 2014). The soil characteristics of the arable layer were neutral pH 6.9, relatively high soil organic carbon 1.76%, with a high content of plant available potassium (K) 144 mg $kg^{-1}$and phosphorus (P) 98 mg kg^{-1}. Two-way factorial ANOVA was conducted, for the interaction of N fertilizers and manipulation diversity in mixtures. Significant differences between the experimental treatments were determined using post hoc exploratory analysis at the 5% probability level ($P<0.05$).

Results and discussion

A two-factor analysis of variance (ANOVA) showed that the effects of different swards composition (grass-legume mixtures) and meeting nutritional needs with N-mineral fertilizers N_{150} and no N-fertilization N_0 (fertilization) on the variation of dry matter yield were significant. From this output we can see that both amount of fertilizers and grass-legume mixtures explain a significant amount of variation in average sward yield (*P*-values <0.005), and the interaction between these terms was also significant (Table 2).

Over two years, comparing unfertilized and fertilized, the most productive treatment was the unfertilized mixture of lucerne, white clover, perennial ryegrass, ×*Festulolium*, meadow fescue and timothy with 9,758 kg ha^{-1}. This was 10% higher yield than the most productive fertilized sward of ×*Festulolium*. Several reports (Barneze *et al.*, 2020; Abalos *et al.*, 2021) also found significant considerable differences between grass-legume mixtures and possibility to reduce the need for N fertilizer in multi-species swards. Our study revealed that the total productivity of the sward is significantly affected by the species of legumes in the mixture more than by the actual number of species sown. Comparing the unfertilized treatments with two and four legume species and the same set of grass species, our results showed that white and

Table 2. Results of two-factor analysis of variance of the dry matter yield.[1]

Year	2019			2020		
Source of variation	df	F	*P*-value	df	F	*P*-value
Factor A (grass/legume mixtures)	11	7.270	0.000	11	7.539	0.000
Factor B (fertilization)	1	9.682	0.002	1	9.758	0.002
Interaction A × B	11	5.207	0.000	11	4.941	0.000

[1] df = degrees of freedom, F = the test statistic from the F-test, *P*-value = the critical value of the Fisher test at a significance level 0.05, effect is significant under $P>95\%$.

red clover in the mixture sustain 8,027 kg ha^{-1} yield productivity, white clover and lucerne 9,309 kg ha^{-1}, white clover and sainfoin 6,027 kg ha^{-1} and all four legumes 8,929 kg ha^{-1}. The yield of the mixture with three supplementary legume species was 13% higher than the average yield (7,787 kg ha^{-1}) of the mixtures with one supplementary legume species and the same grasses species. However, the mixture with the three supplementary legume species was not better than the best mixture with one supplementary species (the one with lucerne), but its yield was higher than what could have been expected from the average of the corresponding mixtures. In summary, the results in Figure 1 show that mineral nitrogen fertilizer increased grass biomass in mixtures but reduced legumes by more than 50%. If we now turn to forage quality, significant differences were observed between monoculture swards with lower crude protein in the forage and higher forage quality of multi-species swards (data not presented here). Previous studies (Moloney *et al.*, 2021) also have noted the importance of plant diversity for forage quality.

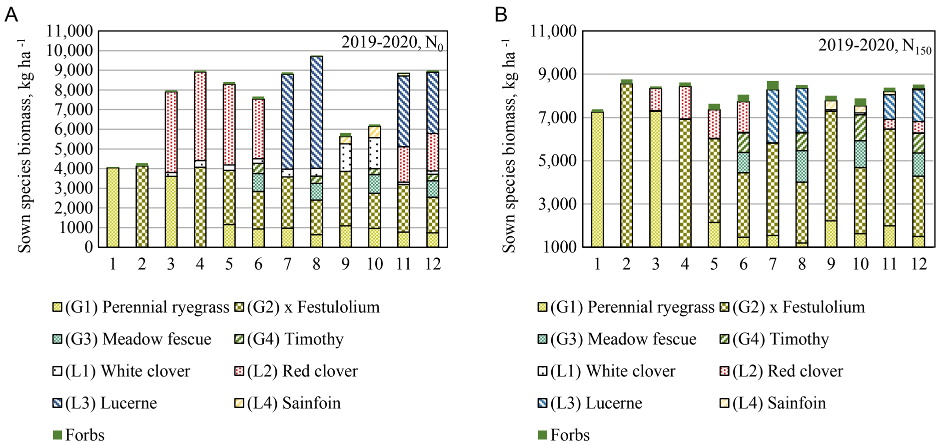

Figure 1. Two-years average botanical composition of the swards; unfertilized N_0 (A), fertilized at N_{150} rate (B).

Conclusions

In the first year of sward use, mineral nitrogen fertilizers significantly increased the productivity of monocultures and mixtures with two legumes – white clover and sainfoin. However, in the most productive mixture with white clover and lucerne, Dry matter yield was significantly reduced (by 13%) using the N_{150} rate. In the second year of sward use, no significant differences were found between mixtures using the N_{150} rate. Also, a two-year analysis showed that the most productive sward was without mineral N fertilizers and with the highest forage quality.

References

Abalos D., De Deyn G.B., Philippot L., Oram N.J., Oudová B., Pantelis I., Clark C., Fiorini A., Bru D., Mariscal-Sancho I. and Groenigen J.W. (2021) Manipulating plant community composition to steer efficient N-cycling in intensively managed grasslands. *Journal of Applied Ecology* 58, 167-180.

Barneze A.S., Whitaker J., McNamara N.P. and Ostle N.J. (2020) Legumes increase grassland productivity with no effect on nitrous oxide emissions. *Plant and Soil* 446, 163-177.

Moloney T., Sheridan H., Grant J., O'Riordan E.G. and O'Kiely P. (2021) Conservation efficiency and nutritive value of silages made from grass-red clover and multi-species swards compared with grass monocultures. *Irish Journal of Agricultural and Food Research* 59(1), 150-166.

Suter M., Connolly J., Finn J.A., Loges R., Kirwan L., Sebastià M.T. and Lüscher A. (2015) Nitrogen yield advantage from grass–legume mixtures is robust over a wide range of legume proportions and environmental conditions. *Global Change Biology* 21, 2424-2438.

WRB (2014) *World reference base for soil resources.* World Soil Resources Reports No. 106. FAO, pp. 187-189.

Enhancing native species seed supply to improve the resilience of Mediterranean pastures

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Abstract

Mediterranean pastures exhibit a great deal of biodiversity with therophytes species as a major component. In Sardinia, where sheep are the most widespread grazing animal, self-sowing annual grasses and legumes show an excellent adaptation. Due to their characteristics, they can tolerate grazing and disseminate a large number of seeds, thereby ensuring the persistence of the pasture. However, this ability to spread the seeds makes it difficult to use traditional combine threshers. Some attempts to directly harvest burr medic legumes (*Medicago polimorfa* L.) with these threshers have been made with promising results. This study aims to verify the best harvest conditions by comparing 2 sowing dates: 4 November 2020 (D1) and 20 January 2020 (D2) as well as 2 different seed rates: 15 kg·ha^{-1} (R1) and 25 kg·ha^{-1} (R2). The potential legume production was 1,844 and 1,356 kg·ha^{-1}, for D1 and D2, respectively; the harvested yield was 413 and 387 kg·ha^{-1} for D1 and D2, respectively. The average harvesting efficiency was 23 and 33% for D1 and D2, respectively. Seed rates did not seem to influence legume production and harvest efficiency.

Keywords: annual legumes, burr medic, seed production, pasture biodiversity

Introduction

Mediterranean pastures exhibit a great deal of biodiversity with therophytes species as a major component. In Sardinia, where sheep are the most widespread grazing animal, self-sowing annual grasses and legumes show an excellent adaptation (Porqueddu *et al.*, 2001; Fara *et al.*, 1997). Due to their characteristics, they can tolerate grazing and disseminate a large number of seeds, thereby ensuring the persistence of the pasture. However, this ability to spread the seeds makes it difficult to use traditional combine threshers. Nevertheless, the use of a native species seed supply obtained from local seed production is one of the key factors in order to preserve pastures' biodiversity and improve the quantity and quality of livestock products over time. For annual medics the difficulty of harvesting with an Australian vacuum harvester is the major constraint for developing a local seed production chain (Chatterton and Chatterton, 1991; Lelievre *et al.*, 1996; Pau 2008; Sulas *et al.*, 2001). The use of a traditional combine harvester would make it possible to set up a local production chain. Some attempts to directly harvest burr medic legumes (*Medicago polimorfa* L.) with combine threshers have been made with promising results (Spanu and Peddis, 2021). This study aims to verify the best harvest conditions by comparing two sowing dates and two different seed rates.

Materials and methods

The trial was carried out at Ussana in Southern Sardinia (39°24' N, 9°05' E, 150 m a.s.l.- WGS84). Two different treatments in a randomized block design with 3 replicates were applied: (1) sowing date, 4 November 2020 (D1) and 20 January 2020 (D2); and (2) seed rate: 15 kg·ha^{-1} (R1) and 25 kg·ha^{-1} (R2).

The plots sown in November were mown once on 24 March 2021, in order to reduce vegetative growth, while the plots sown in January were never mown.

The legume harvest was carried out using a Hege 140 plot combine harvester on 8 June 2021. The legumes not harvested by the combine harvester were collected by hand in two test areas of 0.1 m^2 per plot, in order to determine the harvest efficiency. Legumes and seed weights as well as germinability were subsequently carried out in the laboratory.

Data were analysed with Multi-way ANOVA procedure of Rcommand package for R.

Results and discussion

Concerning harvested legume yield and seed yield, sowing date was not statistically different (413 and 387 kg·ha^{-1} for D1 and D2 and 97 and 107 kg·ha^{-1} for D1 and D2, respectively) (Table 1).

Concerning legume and seed potential production, there was a statistically significant difference at $P \leq 0.05$ between D1 (sown in November) and D2 (sown in January) (1,844 and 1,356 kg·ha^{-1} for D1 and D2, respectively and 663 and 410 kg·ha^{-1} for D1 and D2, respectively). Therefore, the harvesting efficiency of legumes with the combine harvester is 23% for D1 and 33% for D2 (Table 1). At the harvest, the plants sown in January showed a smaller development than those sown in November. Hence, the greater biomass observed during early sowing (i.e. November) hindered the activity of the combine harvest machine thereby resulting in a lower harvest efficiency.

Concerning sowing rate, no statistically significant difference was found at $P \leq 0.05$ (Table 1). Concerning harvested legume weight, a statistically significant difference at $P \leq 0.01$ for 1000 legume weight was found (51.98 and 43.59 for D1 and D2, respectively). Conversely, no statistically significant differences were found for either 1000 legume weight at two different seed rates or for 1000 seed weight both at two different sowing dates and seed rates (Table 2). The different yield observed for 1000 legume weight between D1 and D2 might be due to the greater availability of space and light during ripening for legumes.

Table 1. Harvested yield and potential production of legumes and seeds, kg ha^{-1}.[1]

	Harvested yield		Potential production		Legume harvest efficiency
Seed date	**Legumes**	**Seeds**	**Legumes**	**Seeds**	**%**
November (D1)	413	97	1,844	663	23
January (D2)	387	107	1,356	410	33
Significance ($P \leq$)	n.s.	n.s.	0.05	0.05	0.05
Seed rate					
15 kg ha^{-1} (R1)	397	101	1,574	512	28
25 kg ha^{-1} (R2)	403	103	1,627	561	28
Significance ($P \leq$)	n.s.	n.s.	n.s.	n.s.	n.s.

[1] n.s. = not significant.

Table 2. Harvested legume and seed weight and germinability.[1]

	1000 legumes	1000 seeds	Germinability
Seed date	**g**	**g**	**%**
November (D1)	51.98	3.02	79
January (D2)	43.59	2.99	78
Significance ($P \leq$)	0.01	n.s.	n.s.
Seed rate			
15 kg ha^{-1} (R1)	49.53	3.00	81
25 kg ha^{-1} (R2)	45.97	3.01	79
Significance ($P \leq$)	n.s.	n.s.	n.s.
Overall mean	47.75	3.01	79

[1] n.s. = not significant.

Conclusions

These data suggest that harvesting legumes with a combine harvester is possible with appropriate techniques. Sowing carried out in January and a reduced seed rate of 15 kg ha^{-1} can be a good solution in order to establish a specialized seed production of burr medic and can represent a valid alternative for cereal growers in rotation with durum wheat. However, further trials to maximize the legume production and harvest efficiency are needed.

According to the authors, the development of a seed production chain of local genotypes such as burr medic and other annual medics will depend on the possibility of setting up a field harvesting method proving to as simple and economic as possible.

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References

Chatterton B. and Chatterton L. (1991) Medic seed production in Tunisia. *Agricultural Systems* 37, 77-85.

Fara G., Franca A., Porqueddu C., Caredda S. and Roggero P.P. (1997) Mediche e trifogli annuali autoriseminanti per usi foraggeri e non convenzionali: 1. Adattamento e persistenza. *Rivista di Agronomia* 31, 4, 1009-1018.

Lelievre F., Porqueddu C., Genier G. and Sulas L. (1996) Seed production and harvest of annual burr medic (Medicago polymorpha L.): first results in south of France and Italy (Sardinia). In: *Proceedings of the 3rd International Herbage Seed Conference*. Halle (Saale), Germany, pp. 314-317.

Pau A. (2008) *Tecniche di raccolta di seme di Medicago polymorpha L. cv Anglona*. Degree thesis University of Sassari A.A., Italy, 2007-2008.

Porqueddu C., Ledda L. and Roggero P.P. (2000) Role of forage legumes and constraints for forage legume production in Mediterranean Europe. *Cahiers Options Méditerranéennes* 45, 453-460.

Sulas L., Fara G. and Re G.A. (2001) La raccolta del seme in mediche annuali e trifoglio sotterraneo. *Sementi Elette* 6, 49-58.

Spanu E. and Peddis A. (2021). Legumi di medica polimorfa, innovazioni nella raccolta. *Informatore Agrario* 1/2021, 40-42.

Productivity and forage quality of Alaska brome and smooth brome pure stands and mixtures

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Abstract

The objective of this study was to compare productivity and forage quality of two *Bromus* species in pure stands and in mixtures with two RC cultivars in the growth conditions of Estonia. Alaska brome (AB) cultivar (cv.) Hakari and smooth brome (SB) cv. Lehis were tested in pure stands and in mixtures with red clover (RC) cv. Varte and cv. Ilte during 2017-2021. Pure stands of AB and SB received 200 kg ha^{-1} N per year in three applications. A three-cut harvest system was used. Over the four years, average dry matter yield (DMY) of SB was not significantly higher than that of AB. Based on the four-year average, the mixture of AB with early type RC cv. Varte had a significantly higher DMY than the mixture with late type RC cv. Ilte. Mixtures of SB with either cultivar of RC produced equal DMYs. AB and SB in mixtures as well as in pure stands differed in nutritive value, but generally all produced forage of good feeding value.

Keywords: Alaska brome, smooth brome, red clover, yield, herbage quality

Introduction

During the last twenty years there has been a tendency for increased average annual temperatures compared with the previous period which dates back to 1922. This increase is one reason to search for new forage species in Estonia. Cultivating more drought-tolerant species is one possibility to manage for dry periods and climate change in general. Alaska brome grass (*Bromus sitchensis* Trin. in Bong.) (AB) and smooth brome grass (*Bromus inermis* Leysser) (SB) are promising species in this respect. AB originates from the Pacific coast of North America and has been investigated and cultivated recently in Estonia (Tamm *et al.*, 2018). According to the description, AB is considered a drought resistant and productive species as well as being fast in establishment and regrowth (https://kingsagriseeds.com/wp-content/uploads/2018/02/Hakari.pdf). SB has been cultivated for a long period in Estonia. It is characterized as cold resistant and is well-adapted to dry conditions. SB is a long-lived grass: 5-7 years on average, but can live as long as 10 years or more (Dinkel and Czapla, 2012). AB and SB are good companion species with legumes such as alfalfa (*Medicago sativa* L.) or red clover (Roberts and Kallenbach, 2000). Red clover (*Trifolium pratense* L.) (RC) is one of the most important forage legumes in Estonia because of its high nutritive value and yield. The objective of this study was to compare productivity and forage quality of two *Bromus* species in pure stands and in mixtures with two RC cultivars in the growth conditions of Estonia.

Materials and methods

The trial was carried out during the years 2017-2021 at the Estonian Crop Research Institute in Jõgeva, located in north-eastern Europe (58°45'N, 26°24'E; average annual temperature 5.3 °C, and precipitation 670 mm). The soil of the experimental field was classified as Calcaric Cambic Phaeozem (Loamic), clay loam (40-50% of clay) (IUSS, 2015). The characteristics of the soil horizon were as follows: pH_{KCl} 5.8 (ISO 10390), P 191, K 220 and Ca 1501 mg kg^{-1} (determined by Mehlich III). The seeds were sown on 20 July 2017. The weather conditions were favourable for germination and further growth: total amount of precipitation from August until the end of October was higher in 2017 (313 mm) than the long term average (LTA; 225 mm) and the temperature was similar to LTA. The trial was arranged in a randomized complete block design with four replicates. Cultivars of AB Hakari, SB Lehis and RC Varte (early maturing) and Ilte (late maturing) were included in the trial. Seeding rates were 30 kg ha^{-1} for AB and 38 kg ha^{-1} for SB. In the mixtures with RC the seeding rates were 20 kg ha^{-1} of grasses and 12 kg ha^{-1} of RC. Pure stands of AB and SB received 200 kg ha^{-1} N in three applications (80-60-60 kg ha^{-1}, in spring, after the first and second cut, respectively). The sward was harvested three times a year: first cut at the stage of early heading of SB, second cut 45-50 days later and last cut at the end of growing season (end of September). Crude protein content (CP) and neutral detergent fibre (NDF) were determined. Weather conditions varied significantly during experimental years. First year after sowing was hot and dry (Table 1), whereas 2019 and 2020 were favourable for plant growth. The growth period in 2021 was extremely hot and dry compared to LTA, particularly during the regrowth periods in June and July. Annual dry matter yields (DMY) were calculated as the sum of the DMYs at each cutting. Nutritive value data were averaged over the four years. Statistical analyses were carried out by statistical package Agrobase 20™. Analysis of variance (ANOVA) was performed to test the DMY differences within experimental years. Two-way ANOVA was used to determine the effect of crops, year and their interaction on DMY and nutritive value. Significant differences were calculated using the post-hoc Fisher's LSD test.

Results and discussion

Herbage DMY was significantly (P<0.001) affected by crop species, year and their interaction. AB and SB had a high yield potential in pure stand when a high level of nitrogen fertilizer was applied. As an average of 2018-2021, pure stand of SB out-yielded pure stand of AB, but not significantly (by 3.6%) (Table 2). Plant growth was affected by weather conditions (Table 1). In the spring, when soil is wet and water is not limited for growth, the regrowth and total seasonal DMY is strongly affected by the amount of precipitation. In 2020 the amount of precipitation was abundant during the entire vegetation period, and AB produced significantly more DMY (by 30%) than SB. However, SB was 30% more productive than AB in the following year. We suppose that drought and high daily temperatures in 2021 during the regrowth period (June–July) affected the formation of DMY more in AB than in SB. Dinkel and Czapla (2012) reported also that SB is resistant to drought and temperature extremes. Among mixtures, the average DMYs (2018-2021) ranged from 6.72 to 8.43 Mg ha^{-1}. RC is a short-lived species that significantly increases DMYs in the first three years and then declines in the fourth year. We used RC cultivars with different development rhythm: RC Varte matured 14-16 days earlier than RC Ilte. Based on the four-year average, mixture of AB with RC Varte had a significantly higher DMY than mixture with RC Ilte. Mixtures of SB with either cultivar of RC showed no significant differences between DMYs.

The mixtures and pure grass stands differed significantly (P<0.001) in their nutritive value (Table 3). Based on four-year averages, AB and SB in the mixtures with RC cultivars generally produced forage of high nutritive value (recommended NDF concentration ≤550 and CP>140 g kg^{-1} DM) (Tamm, 2017) in all harvest times (Table 3). Our observations showed that SB matured earlier than AB, and therefore needs earlier harvesting of first cut to ensure feed with high protein content.

Table 1. Weather data of the vegetation periods in 2018-2021 at Jõgeva.

	Average air temperature, °C					Sum of precipitation, mm				
	May	June	July	August	September	May	June	July	August	September
2018	14.5	15.0	20.3	17.9	13.6	16.6	23.0	15.2	75.6	72.4
2019	10.6	17.8	15.9	15.7	11.1	50.0	54.4	33.9	49.1	67.1
2020	9.1	17.9	15.7	16.1	13.4	27.6	135.9	100.5	82.7	74.0
2021	10.4	19.0	21.0	15.3	9.7	87.7	11.0	0	104.5	54.8
1991-2020[1]	5.1	10.9	15.0	17.5	16.0	33.5	45.2	84	64.6	89.3

[1] Long term average.

Table 2. Dry matter yield (Mg ha^{-1})of Alaska brome (AB) and smooth brome (SB) in the mixtures with red clover (RC) cultivars and in pure stands.[1]

	2018	2019	2020	2021	Average 2018-2021
SB + RC Varte	8.71^{b}	9.92^{a}	7.76^{d}	5.69^{c}	8.02^{c}
AB + RC Varte	7.98^{b}	10.31^{a}	9.61^{b}	5.84^{c}	8.43^{b}
SB + RC Ilte	7.17^{c}	9.56^{a}	9.09^{bc}	5.27^{c}	7.77^{c}
AB + RC Ilte	6.01^{d}	8.08^{b}	8.69^{c}	4.11^{d}	6.72^{d}
SB	9.59^{a}	9.81^{a}	9.88^{b}	11.21^{a}	10.12^{a}
AB	9.39^{ab}	8.04^{b}	13.00^{a}	8.62^{b}	9.76^{a}
Average	8.14^{B}	9.29^{A}	9.67^{A}	6.79^{C}	Crop: P<0.001;
	P<0.001	P<0.001	P<0.001	P<0.001	Crop × Year: P<0.001

[1] Mean values without common lowercase letters (a,b,c,d) within columns and mean values without common uppercase letters (A,B,C,D) in rows are statistically significantly different (P<0.05, Fisher LSD test).

Table 3. Average crude protein (CP) and neutral detergent fibre (NDF) contents (g kg^{-1} DM) in Alaska brome (AB) and smooth brome (SB) in pure stands and in mixtures with red clover (RC) cultivars averaged over 2018-2021.[1]

	Cut 1		Cut 2		Cut 3	
	CP	NDF	CP	NDF	CP	NDF
SB + RC Varte	137.6^{c}	423.9^{c}	174.2^{b}	381.4^{cd}	185.0^{b}	391.5^{c}
AB + RC Varte	146.7^{ab}	400.9^{de}	165.4^{c}	398.4^{c}	185.7^{b}	396.2^{c}
SB + RC Ilte	142.5^{bc}	405.5^{d}	181.0^{a}	356.9^{d}	191.6^{a}	361.2^{d}
AB + RC Ilte	149.5^{a}	389.4^{e}	161.0^{cd}	392.1^{c}	186.6^{b}	386.2^{c}
SB	116.5^{d}	559.7^{a}	158.3^{d}	504.2^{b}	145.5^{c}	505.5^{b}
AB	138.5^{c}	524.7^{b}	135.9^{e}	552.5^{a}	146.2^{c}	529.4^{a}
Average	138.6	450.7	162.6	430.9	173.4	428.3

[1] Crop: P<0.001; Year: P<0.001; Crop × Year: P<0.001. Mean values with different lowercase letters (a,b,c,d) within columns are statistically significantly different (P<0.05, Fisher LSD test).

Conclusions

The average DMY of SB over 4 years was not significantly higher than that of AB. Mixture of AB with early type RC Varte had a significantly higher DMY than mixture with late RC Ilte. DMYs of the mixture including SB with either early or late RC did not differ significantly. Mixtures containing AB and SB as well as pure stands differ in nutritive value, but all produced forage of high nutritive value. Due to earlier maturation, SB needs earlier harvesting than AB in order to ensure good feed quality. To achieve excellent feed quality, SB would require even earlier RC companion than cultivar Varte.

References

Dinkel C.L. and Czapla P.K. (2012) Alaska Forage Manual. Available at: http://plants.alaska.gov/pdf/forage_manual.pdf.

https://www.kingsagriseeds.com/wp-content/uploads/2018/02/Hakari.pdf.

IUSS (2015) World reference base for soil resources 2014, update 2015. Reports. 106. FAO, Rome, Italy.

Roberts C.A. and Kallenbach R.L. (2000) *Smooth bromegrass*. MU Ext. Publ. G4672.

Tamm U. (2017) *Parema toiteväärtusega rohusööt.* Eesti Taimekasvatuse Instituut, Saku, Estonia, 57 pp.

Tamm U., Meripõld H., Tamm S. and Edesi L. (2018) The nutritive value of Alaska brome and tall fescue forage using different growing technologies. *Grassland Science in Europe* 23, 363-365.

What is a suitable management for *Typha latifolia* control in wet meadow?

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Abstract

Typha latifolia causes serious problems in wet meadows by overgrowing and suppressing other native plants. To find suitable management for *T. latifolia* control, a long-term experiment was established in the Malá Strana nature reserve (Czechia) in 2005. The experiment consists of five treatments (unmanaged control; cutting once a year in June without biomass removal and with biomass removal; cutting twice per year in June and August without biomass removal and with biomass removal) with three replications. Percentage cover of *T. latifolia* and other vascular plant species was visually estimated (2005-2017) and *T. latifolia* traits (number of tillers per each plot, height in transect, dry matter biomass yield (DMBY) and litter mass) were measured during the years 2005-2018 at the end of June. Cutting twice per year significantly decreased cover, number of tillers, plant/tiller height in transect (cm), DBMY (t ha^{-1}) and litter (t ha^{-1}) of *T. latifolia*. Biomass removal significantly decreased number of tillers, plant/tiller height in transect (cm), DMBY (t ha^{-1}) and litter (t ha^{-1}) of *T. latifolia.* Cutting twice per year was found to be the most appropriate management to reduce *T. latifolia* plants and for maintaining desirable plant species composition of wet meadow. Further, reduction of *T. latifolia* plants was achieved by biomass removal.

Keywords: biomass removal, cutting, plant species composition, richness, traits, grassland

Introduction

Typha latifolia belongs to the group of aquatic macrophytes that have caused dozens of weed problems throughout the world (Ball, 1990). *T. latifolia* tolerates a broad range of climatic conditions and soils that remain wet or saturated by water over a major part of growing season (Grace and Harisson, 1986). Cutting is considered as one of the most appropriate management options for *Typha* spp. control. Harvesting of *T. latifolia* decreases the amount of nutrients and non-structural carbohydrates transferred from the shoots to belowground organs, which are necessary for initial growth of new shoots in the following spring (Maddison *et al.*, 2009). Although many previous studies have addressed the effect of regular cutting management on *T. latifolia* growing in deep water in different waterlogged areas, none have considered the long-term effect of cutting on this species, growing in the wet meadows with shallow water supply. To address this knowledge gap, a long-term experiment in Malá Strana nature reserve in the wet meadow alliance *Calthion* with *T. latifolia* dominance was established.

Materials and methods

The experiment was established in 2005 in the Jizerské hory Mts. (Czechia, 50°45'52'N, 15°12'19'E; 722 m elevation). The area of each plot is 16 m^2 (4×4 m). The experiment is arranged in three completely randomized blocks with five treatments: unmanaged control (U), one cut per year in June without biomass removal (1O), one cut per year in June with biomass removal (1R), two cuts per year in June and August without biomass removal (2O), two cuts per year in June and August with biomass removal (2R). The cutting height is approximately 3 cm above the soil surface. The number of *T. latifolia* tillers was recorded from each experimental plot (16 m^2) through the study period (2009-2018) and height of all presented *T. latifolia* tillers was measured along a 30 cm wide transect diagonally in each experimental plot through the study period 2008-2017. *T. latifolia* herbage and litter were subsampled in the years 2008-2017 from the harvested herbage. The percentage cover of *T. latifolia* and other vascular plant species was estimated visually in each experimental plot every year 2005-2011, 2013 and 2017. General linear models (GLMs) were used to test effect of intensity, biomass removal and intensity×biomass removal interaction on all *T. latifolia* traits, after excluding U treatment from the model. Effect of replications was used as random factor. Then principal component analysis (PCA) was used to detect trends in vegetation development over the period of experiment duration (2005-2017). Treatments were used as covariables.

Results and discussion

Two cuts significantly decreased cover, number of tillers, plant/tiller height in transect (cm), DBMY (t ha^{-1}) and litter (t ha^{-1}) of *T. latifolia* (Figure 1, Table 1 and 2). Cutting two or three times per vegetation season was suggested by Apfelbaum (1985) and by Sale and Wetzel (1983) as the optimal management for successful *T. latifolia* control. Similar results were achieved in our experiment and revealed that cutting twice per year led to a significant reduction of *T. latifolia* plants in comparison with the control treatment. Biomass removal significantly decreased number of tillers, plant/tiller height in transect (cm), DMBY (t ha^{-1}) and litter (t ha^{-1}) of *T. latifolia.* Cutting intensity × biomass removal interaction significantly decreased plant/tiller height of *T. latifolia* in transect (cm). Results of PCA analysis showed temporal development of vegetation in all treatments over the duration of the experiment (Figure 2). Temporal vegetation development in the control treatment (U) did not show marked changes between all sampling years compared with that of the cutting treatments (1O, 1R, 2O, 2R), which resulted in different plant species composition.

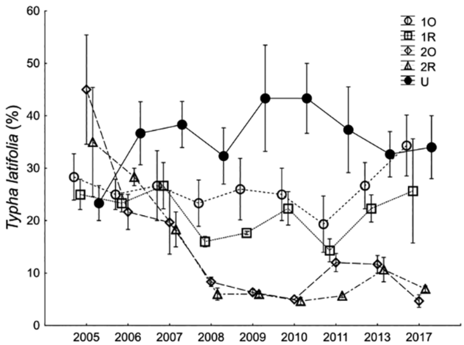

Figure 1. Cover of dominant plant species *Typha latifolia*. Treatment abbreviations are explained in Materials and methods section.

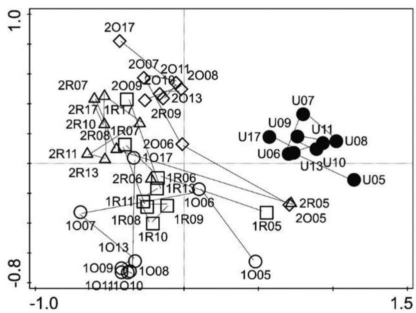

Figure 2. PCA analysis. Results of PCA (2005-2017). Treatment abbreviations are explained in Materials and methods section. Last two numbers after the treatment abbreviations are abbreviations for individual years throughout the study period.

Table 1. GLM analysis.[1]

	Tested variable	Cutting intensity	Biomass removal	Cutting intensity × Biomass removal
Cover of *Typha latifolia* (%)	*F*-ratio	108.9	6.3	2.6
	P-value	<0.001	0.015	0.115
Number of *T. latifolia* tillers	*F*-ratio	89.7	8.16	1.45
	P-value	<0.001	0.005	0.231
Height of *T. latifolia* in transect (cm)	*F*-ratio	421.9	42.3	13.2
	P-value	<0.001	<0.001	<0.001
T. latifolia dry matter biomass yield (t ha^{-1})	*F*-ratio	56.79	7.38	0.33
	P-value	<0.001	0.007	0.565
T. latifolia litter (t ha^{-1})	*F*-ratio	144.8	11.8	5.2
	P-value	<0.001	0.001	0.023

[1] *F* ratio = F statistics for the test of particular analysis, *P*-value = obtained probability value. Significant results of *p*-values (after table-wise Benjamini-Hochberg's FDR correction) are in background shading. Df = Degree of freedom was 1 for Intensity, Biomass removal and Intensity×Biomass removal interaction in all analysis performed.

Table 2. GLM analysis.[1]

	Cutting intensity		Biomass removal	
	1 cut	2 cut	Biomass removal	No biomass removal
Cover of *Typha latifolia* (%)	22.75±1.46	7.33±0.52	13.19±1.45	16.89±1.87
Number of *T. latifolia* tillers	153.75±9.30	72.68±3.61	98.37±5.73	128.07±10.72
Height of *T. latifolia* in transect (cm)	88.78±0.69	80.68±1.23	84.67±0.94	87.42±0.82
T. latifolia dry matter biomass yield (t ha^{-1})	0.63±0.04	0.33±0.04	0.41±0.03	0.54±0.04
T. latifolia litter (t ha^{-1})	0.09±0.008	0.01±0.002	0.04±0.005	0.06±0.007

[1] Numbers represent average of three replications ± standard error of the mean. Df = Degree of freedom was 1 for Cutting intensity and Biomass removal.

Conclusions

Cutting twice per year seems to be a suitable management for maintaining desirable plant species composition and for the control of *T. latifolia*. Further reduction of *T. latifolia* can be supported by biomass removal. The results of this study could be used by nature protection authorities for the suggestion of sustainable managements of wetland meadows with *T. latifolia* dominance as a background for the preparation of the management schemes in the nature reserves.

Acknowledgements

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References

Apfelbaum, S.I. (1985) Cattail (*Typha* spp.) Management. *Natural Areas Journal* 5(3), 9-17.

Ball, J.P. (1990) Influence of subsequent flooding depth on cattail control by burning and mowing. *Journal of Aquatic Plant Management* 28, 32-36.

Grace, J.B. and Harrison, J.S. (1986) The biology of Canadian weeds. 73. *Typha latifolia* L., *Typha angustifolia* L. and *Typha x glauca* Godr. *Canadian Journal of Plant Science* 66, 361-379.

Maddison, M., Mauring, T., Remm, K., Lesta, M. and Mander, Ü. (2009) Dynamics of *Typha latifolia* L. populations in treatment wetlands in Estonia. *Ecological Engineering* 35, 258-264.

Sale, P.J.M. and Wetzel, R.G. (1983) Growth and metabolism of *Typha* species in relation to cutting treatments. *Aquatic Botany* 15, 321-334.

Comparison of vegetation growth in a chicory based pasture and a multi-species based grassland

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Abstract

With climate change, summer droughts tend to exacerbate. Such conditions are disadvantageous for vegetation growth and, therefore, for animal grazing. Thus, forage plants resistant to drought may be a way to ensure summer feeding for animals. Chicory (*Cichorium intybus* L.) has deep roots reduce its sensitivity to drought. Experimentation on the use of chicory as feed for equine diets was conducted in 2020, and heights and biomass growth of a chicory based pasture were evaluated in comparison with control pastures of multi-species based permanent grassland. In early July, when grazing began, control grasslands showed better growth. After two dry and hot months, however, this growth decreased while chicory growth was steady. Computing linear mixed models, the interaction between type of grassland and the grazing period appeared significant ($P<0.05$) on height growth. During a drought period better growth of chicory was observed (0.36 ± 0.14 cm d^{-1} compared with 0.08 ± 0.07 cm d^{-1} for control). Differences were highlighted only with regard to height and not biomass, which might be due to the higher water content of chicory. These results tend to confirm the efficiency of chicory as a species resistant to summer drought and its acceptability by horses.

Keywords: chicory, drought, equine grazing, plant growth

Introduction

Crops from temperate areas have a maximal growth when outdoor temperatures are between 15 and 23 °C (Perera *et al.*, 2019) and growth is inhibited above 35 °C (Langworthy *et al.*, 2015). Water shortage and high temperatures limit vegetation development, making summer a challenging season for animal grazing (Hatfield and Prueger, 2015; Lemaire, 1987). To overcome this issue, plants adapted to drought can be used to provide food when growth of currently used meadow species is limited and/or inhibited. Chicory (*Cichorium intybus* L.) has deep roots and better access to soil water than most grass species (Langworthy *et al.*, 2015; Perera *et al.*, 2019). In addition, the nutritional values of chicory are suitable for animal feeding (Delagarde *et al.*, 2014). Thus, the aims of this study were to evaluate: (1) the efficiency of chicory as a forage to provide horses with enough food during summer; and (2) chicory resistance to horse grazing.

Materials and methods

Experiments took place in Chamberet (France), in 2020. From 24 June to 8 October, 28 two-year-old horses were included in the experiment. Five were conducted on chicory-sown pasture (C) and 23 on a control grassland (Ctl). The chicory pastures were sown with only chicory seeds (var. Choice) in April after ploughing and manure input. The control grasslands were permanent pasture multi-species swards based on 60% grass (mainly ryegrass, fescue and cocksfoot), 25% legume (mainly clover and alfalfa) and 15% other plants. In early April, 60 U of nitrogen was applied to these pastures. Animals were managed under rotational grazing and they went through several grazing cycles on each plot (7 cycles for C group; 4 cycles for Ctl group). The ground cover of C pastures was evaluated at 100 points along two diagonals in each plot, before and after grazing, using a Herbostick. At each point, what was found at the tip of the Herbostick was identified (chicory, legume, grass, other type of plants, faeces or bare soil) and the height recorded. Both configurations had the same number of animals per hectare (C=16.27 animal ha^{-1}; Ctl=16.28 animal ha^{-1}).

Measures of vegetation height and biomass were performed for the two types of pasture each time horses went in or out a plot. Depending the size of the plot, grass height was evaluated on 30 to 45 points using a plate meter (Jenquip©). Biomass sampling consisted of one sample taken on a surface of 10×5 m. Grass height was measured with a Herbostick at 5 points inside the delimited surface before and after grass cutting. From the known values of height and weight of the harvested grass, density (D) was calculated (D=weight of grass/height of grass). Grass samples were dried at 60 °C during 72 hours to estimate dry matter (DM). These data allowed the evaluation of height growth (HG) and biomass growth (BG) with:

HG = (height before $grazing_n$ – height after $grazing_{n-1}$) / number of days between grazings;

BG = (height before $grazing_n$ – height after $grazing_{n-1}$) × D / number of days between grazings;

with n = number of the grazing cycle.

Effects of chicory grazing on horses were monitored by weekly observations on their general attitude, hair state, body condition score (French scoring, scale from 1 to 5), swollen belly (presence or absence) and the texture of faeces.

Statistical treatments were realised using R software computing linear mixed models (LMM) for HG and BG. In each case, fixed effects were group, period (from 1 to 6 set as a continuous variable, with 1: 'early July'; 2: 'end of July'; 3: 'early August'; 4: 'end of August; 5: 'early September'; 6: 'end of September') and the interaction between groups and period, while plots were set as random effect. When the interaction between period and group was significant Wilcoxon tests were applied for each period.

Results and discussion

Chicory grazing had no adverse effects on horses' health. The C group showed only more slack faeces than the Ctl group. More detailed information was published previously regarding these data (Valleix *et al.*, 2021).

Although only chicory was sown in C pastures, some other species grew. Pasture swards were mainly chicory (40-60%) but grasses (5-15%), legumes (up to 3%) and other plants (23-30%) were observed. Faeces (0-6%) and bare soil (9-23%) were recorded. These values are from assessments made throughout the grazing season before and after grazings.

The DM for C- and Ctl-based pastures were not different at the beginning of the experiment and the time spent by horses on each pasture type was equivalent along the grazing season (~3 days per grazing cycle). Intercycle times, however, were reduced for the C group compared to the Ctl group (13 and 23 days on average, respectively), which illustrates the higher growth of C and its usefulness as animal forage.

Growths of Ctl grassland were the highest early July, both for HG (0.41±0.09 cm d^{-1}) and BG (33.70±13.86 kg of DM ha^{-1} d^{-1}). Then, HG and BG decreased until early October (HG = 0.05±0.02 cm d^{-1} and BG = 3.22±1.64 kg of DM ha^{-1} d^{-1}).The growth of the C fluctuated more than Ctl but remained between 0.11±0.04 cm d^{-1} and 0.36±0.14 cm d^{-1} for HG, and 8.0±3.13 kg of DM ha^{-1} d^{-1} and 33.32±21.43 kg of DM ha^{-1} d^{-1} for BG. Results obtained from LMM on HG and BG are summarized in Table 1. The linear mixed model highlights a significant effect ($P<0.05$) of the interaction between the type of grassland and the grazing period on HG.

Table 1. Results of LMM for HG and BG with group, period and the group:period interaction as fixed effects and the plot as random effect.

	HG			BG		
	Amplitude	Std. error	*P*-value	Amplitude	Std. error	*P*-value
Group	0.12	0.06	0.22	12.19	10.13	0.26
Period	$-4.5.10^{-3}$	0.01	0.76	-0.65	1.60	0.69
Group:Period	-0.06	0.02	<0.05	-4.51	2.63	0.09

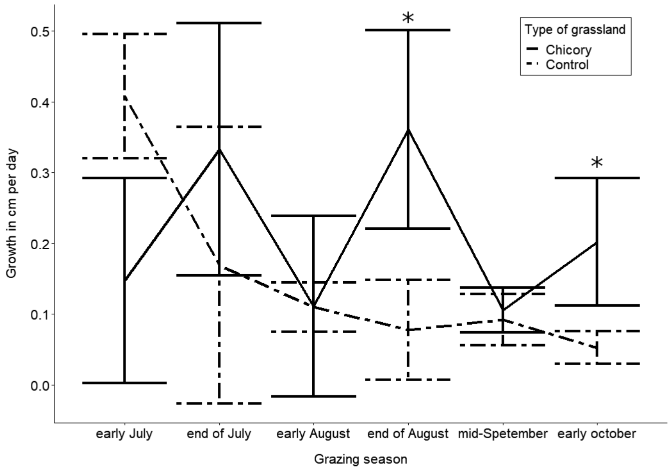

Figure 1. Mean height growth of the vegetation per period during the grazing season depending of the type of grasslands, with standard deviation. Significance ($P<0.05$) of the difference between chicory and control pastures is indicated by *.

Evolution of HG for both C and Ctl pastures are represented in Figure 1. In early July, HG tends to be higher for Ctl ($P=0.07$; C=0.15±0.14 cm d^{-1}; Ctl=0.41±0.09 cm d^{-1}) while HG for C is significantly ($P<0.05$) higher at the end of August (C=0.36±0.14 cm d^{-1}; Ctl=0.08±0.07 cm d^{-1}) and early October (C=0.20±0.09 cm d^{-1}; Ctl=0.05±0.02 cm d^{-1}).

From July to August rainfall was low (15.6 mm between 1 July 2020 and 12 August 2020). Average temperatures were high (20.67±3.43 °C with a maximum of 37.40 °C). This period can is considered as a summer drought. Thus, the growth of chicory is higher than the control sward after the drought period. Even though no significant differences were identified for BG regarding group, period or the interaction, it might be mentioned that the average BG of chicory was 17.82±13.34 kg of DM ha^{-1} d^{-1} throughout the season; this is in accordance with previously published data that estimated production of between 5 and 22.8 kg DM ha^{-1} d^{-1} for chicory (Powell *et al.*, 2007). Differences between C and Ctl were observed only regarding height and not in terms of DM. High content of water of the chicory can explain this observation (Perera *et al.*, 2019) as this plant has less dry matter for the same amount of fresh matter as grasses or clovers (Delagarde *et al.*, 2014).

Conclusions

Results from this study showed the resistance of chicory to drought and its capacity for continuing growth despite intensive equine grazing. Besides the confirmation that horses can be kept on chicory-based pastures, this experiment indicates that growth of chicory swards is higher than from multi-species based grassland during and after a period of drought.

References

Delagarde R., Roca Fernandez A.I. and PeyraudJ.L. (2014) Prairies multispéclfiques avec ou sans chicorée : densité du couvert mesurée à l'herbomètre et composition chimique. *Fourrages* 218, 177-180.

Hatfield,J.L. and Prueger J.H. (2015) Temperature extremes: effect on plant growth and development. *Weather and Climate Extremes* 10, 4-10. https://doi.org/10.1016/j.wace.2015.08.001.

Langworthy A., Pembleton K., Rawnsley R., Harrison M., Lane P., Henry D. and Corkrey R. (2015) Chicory (*Cichorium intybus* L.) can beat the heat during summer drought in southeast Australian dairying regions. *Building Productive, Diverse and Sustainable Landscapes*, 4 pp. Proc 17th ASA Conference.

Lemaire G (1987) Physiologie de la croissance de l'herbe: applications au pâturage. *Fourrages* 112, 352-344.

Perera R.S., Cullen B.R. and Eckard R.J. (2019) Growth and physiological responses of temperate pasture species to consecutive heat and drought stresses. *Plants* 8. https://doi.org/10.3390/plants8070227.

Powell A.M., Kemp P.D., Jaya I.D. and Osborne M.A. (2007) Establishment, growth and development of plantain and chicory under grazing. *Proceedings of the New Zealand Grassland Association* 69, 41-45. https://doi.org/10.33584/jnzg.2007.69.2684

Valleix M., Brossier M. and Wimel L. (2021) The chicory: a potential crop against summer drought for horses. *72nd Annual Meeting, Davos, Switzerland, p.* 312.

Assessing the effect of grassland type on invertebrates

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Abstract

Biodiversity is under pressure worldwide. In the Netherlands, decreasing numbers of meadow birds, like the black-tailed godwit and the northern lapwing, are of concern to society. One of the possible reasons for this is reduced available of food (invertebrates: insects and worms). Even though the birds are easily spotted, this is not the case for their feed. Therefore, a protocol was developed with methods to easily monitor the occurrence of invertebrates on a large scale at commercial dairy farms. In the protocol, focus is on different layers of grasslands: above soil (flying insects), on soil (walking insects) and in soil (worms). Invertebrates are monitored using sticky traps, pitfall traps and 20×20×20 cm soil samples, respectively. Data can be stored on-site using a monitoring app, developed as part of the protocol. The protocol was tested throughout 2021 at the experimental farm 'Aeres Farms' in the Netherlands. Results showed no clear effect of grassland with different botanical composition on number of invertebrates. The number of invertebrates was highest in late spring and early summer.

Keywords: biodiversity, invertebrates, pitfall traps, soil samples, sticky traps

Introduction

Biodiversity is the variety of life in a particular habitat. It contains all living organisms, including plant and animals. The distribution of plant and animal life and the variety of landscapes are the product of complex interactions between natural processes and human activities. As a result of human activities, biodiversity is under pressure. This is of concern to society since biodiversity is fundamental for many ecosystem services provided to society (Cardinale *et al.*, 2012). Many of these ecosystem services are related to grasslands and valued by stakeholder groups (Van den Pol-van Dasselaar *et al.*, 2014). In the Netherlands, society is especially concerned about the decreasing numbers of meadow birds, like the black-tailed godwit (*Limosa limosa*) and the northern lapwing (*Vanellus vanellus*) (SOVON, 2020). Possible reasons for this decrease are increasing land use intensity, decreasing diversity in grasslands, less available feed (invertebrates: insects and worms), predation, and the continuing urbanisation. This paper focuses on occurrence of invertebrates in grasslands as indicator of biodiversity and as indicator for available feed for meadow birds. Even though the birds are spotted easily, this is not the case for their feed. The abundance of terrestrial insects has been reported to be decreasing worldwide (Van Klink *et al.*, 2020), but the number of studies on invertebrates in grasslands is still limited. The aim of this study was therefore to explore the options for easy methods for studying the occurrence of invertebrates in practice on grasslands on commercial dairy farms at a large scale, for example by students.

Materials and methods

In the autumn of 2020, a protocol was developed to easily monitor the occurrence of invertebrates on grasslands at commercial dairy farms. The focus was on monitoring different layers within the grasslands, i.e. above the soil (flying insects), on the soil (walking insects), and in the soil (worms). The first version of the protocol was tested by students of Aeres University of Applied Sciences in autumn 2020. Results and experiences from this test led to further refinements of the protocol. In the first half year of 2021, the final protocol was used by another group of students to monitor five paddocks of the experimental farm of Aeres in the Netherlands ('Aeres Farms'). These paddocks were all organic and were regularly grazed. They were sown with different grass mixtures, i.e. a paddock with a mixture of *Lolium perenne* (perennial ryegrass) and *Trifolium pratense* (red clover), two paddocks with a mixture of *L. perenne* + *Trifolium repens* (white clover), a paddock with *L. perenne* and seven herbs, and a mixture of *Festuca arundinacea* (tall fescue) and seven herbs. The seven herbs were (1) *Trifolium pratense*; (2) *Trifolium repens*; (3) *Onobrychis viciifolia* (common sainfoin); (4) *Carum carvi* (caraway); (5) *Cichorium intybus* (common chicory); (6) *Lotus corniculatus* (birdsfoot trefoil); and (7) *Plantago lanceolata* (ribwort plantain).

Results and discussion

Methods for monitoring invertebrates on commercial dairy farms should not only be easy to use, but also robust since they should provide comparable results under different circumstances and when used by different persons. Therefore, the protocol was based on counting the number of insects and worms. In addition, these insects and worms may be identified using determination guides. However, this requires some additional expertise and was therefore not part of this protocol. The final standardized protocol was as follows:

- Flying insects are monitored using three yellow sticky traps of 10×25 cm per field. They are collected 48 hours after they have been placed. Analysis includes: trap photography, counting of number of insects <4 mm, 4-10 mm and >10 mm.
- Walking insects are monitored using three pitfall traps per field with a top diameter of 10 cm that are collected 48 hours after they have been placed. Analysis includes: counting of numbers of insects <4 mm, 4-10 mm and >10 mm.
- Worms are monitored using three 20×20×20 cm soil samples per field. Analysis includes counting of number of worms in the soil samples.

Uniform sampling was facilitated by providing detailed instruction cards with picture images that were sealed to ensure that they were easy to use in the field. It was further facilitated by a monitoring app that was developed as part of the protocol. Data could thus be stored on-site via mobile phones. The app provides further guidance of the protocol and ensures that all necessary data are directly stored. The final protocol, supported by the instruction cards and the monitoring app, was assessed as satisfactory by the users and used in spring 2021 at the five paddocks of the experimental farm 'Aeres Farms'. Results for sticky traps, pitfall traps and soil samples are shown in Figure 1. An analysis of variance showed significant main effects of time of measurement (five moments in the period March to May) and of grassland paddock on number of invertebrates. There were, however, also significant interactions between time of measurement and grassland paddock on the number of invertebrates (P<0.001, P=0.016 and P=0.144, for sticky traps, pitfall traps and worms, respectively). Therefore, we could not determine a clear effect of grassland type on number of invertebrates. The number of invertebrates was highest in late spring and early summer. Especially the number in spring is important since this is when the meadow birds are present and the young birds are hatched. Insects found on sticky traps and in pitfall traps were divided into different size categories. For the pitfall traps, the pattern in time of different size categories did not differ from the overall pattern of total number of insects (not shown). The results for the sticky traps show that the higher number of insects at late spring and early summer was mainly due to insects <4 mm (Figure 2). It has to be noted that sticky traps are not suitable for all insect groups, and this may have influenced the results. Furthermore, measurements need to be prolonged over a longer period of time to study temporal variation in number of invertebrates present.

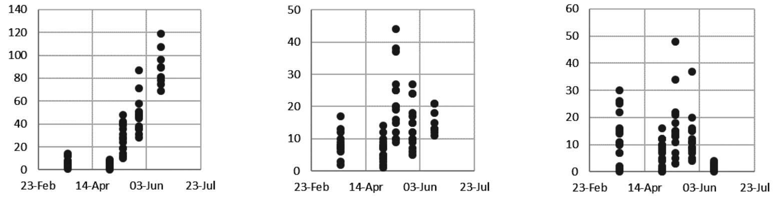

Figure 1. Number of insects per sticky trap (left), insects per pit fall (middle) and worms per soil sample (right) according to the protocol at 'Aeres Farms' in 2021.

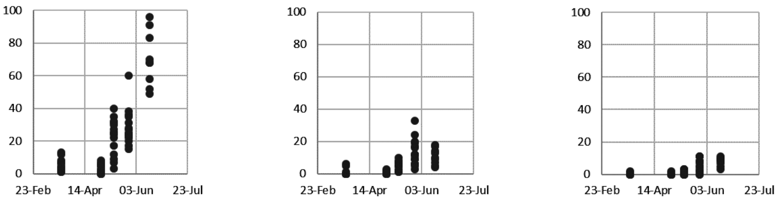

Figure 2. Number of insects per sticky trap in the size category <4 mm (left), 4-10 mm (middle) and >10 mm (right) according to the protocol at 'Aeres Farms' in 2021.

Conclusions

This study developed a protocol and a monitoring app for monitoring easily the occurrence of invertebrates at grasslands of commercial dairy farms. Results of research with this protocol within one farm showed no clear effect of grassland with different botanical composition on number of invertebrates. Measurements need to be prolonged to study temporal variation.

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References

Cardinale B.J., Duffy J.E., Gonzalez A., Hooper D.U., Perrings C., Venail P., Narwani A., Mace G.M., Tilman D., Wardle D. A., Kinzig A.P., Daily G.C., Loreau M. and Grace J.B. (2012) Biodiversity loss and its impact on humanity. *Nature* 486 (7401), 59-a67.

SOVON (2020) *Vogelbalans*. SOVON, Nijmegen, the Netherlands, 22 pp.

Van den Pol-van Dasselaar A., Goliński P., Hennessy D., Huyghe C., Parente G. and Peyraud J.L. (2014) Évaluation des fonctions des prairies par les acteurs européen. *Fourrages* 218, 141-146.

Van Klink R., Bowler D.E., Gongalsky K.B., Swengel A.B., Gentile A. and Chase J.M. (2020) Meta-analysis reveals declines in terrestrial but increases in freshwater insect abundances. *Science* 368, 417-420.

Productivity and management of herb-rich mowed grasslands in Flanders: a practice-oriented field trial

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Abstract

Interest in herb-rich productive grasslands is growing in Belgium, but farmers and advisers lack practical knowledge. A plot-scale field trial was started in autumn 2020 where crop yield and evolution in sward composition of grass-clover+herbs (plantain or plantain+chicory) mixtures at different N fertilization levels were compared to perennial ryegrass and grass-clover under intensive mowing conditions. Results from first production year 2021 revealed that grass-clover+plantain+chicory at 75 kg N ha^{-1} can achieve the same crop yields as perennial ryegrass at 375 kg N ha^{-1} and grass-clover at 125 kg N ha^{-1}. Increasing the N fertilization had little effect on yields of the grass-clover+ plantain+chicory mixtures; however, it did influence the sward composition. The clover content decreased as N fertilization increased. The herb content was the highest in the 75 kg N ha^{-1} fertilization, compared to zero N and 375 kg N ha^{-1} fertilization. In the mixtures with the lowest N fertilization, the grass share in the 4^{th} and 5^{th} cut was nearly gone. Observations in the coming year should reveal whether the grass component has returned next spring and if the plantain and chicory can be maintained in the sward. Replacing perennial ryegrass in the mixtures by tall fescue had no effects in the first year. Important to note is that the relative differences in this field trial could be influenced by the growing season of 2021, which had more precipitation than normal, had no periods of drought/heat stress, and resulted in higher grassland yields than normal.

Keywords: grass-clover, plantain, chicory, botanical composition, crop yield, N fertilization

Introduction

In Belgium, perennial ryegrass is preferred because of its high digestibility and high production under Belgian temperate maritime climate conditions. Recent droughts, restrictions on N fertilizer use, and incentives to stimulate use of leguminous plants and herb-rich mixtures are leading many farmers to explore different management approaches for at least some of their grasslands. Farmers and advisers have little practical knowledge about management of herb-rich grasslands, however. ILVO started a field trial in 2020 as a demonstration of productive herb-rich grassland mixtures and with the aim of gaining more practical knowledge about fertilization and botanical evolution. We compared perennial ryegrass and grass-clover, with a grass-clover + herbs mixtures at several N fertilization levels. One of the mixtures contains tall fescue instead of perennial ryegrass. Every mixture with clover contained both white and red clover. The selected herbs were plantain and chicory. The objectives of this study were to determine: (1) if tall fescue is a better grass component than perennial ryegrass in a herb-rich mixture; (2) if crop yields of herb-rich grassland are comparable to perennial ryegrass and grass-clover; and (3) the optimal N fertilization level of herb-rich grassland.

Materials and methods

The field trial was carried out in Merelbeke (Belgium) on an arable field (sandy loam; pH_{KCl}=6.3; 0.82% C) with flax as the preceding crop. The field trial was sown on 15 September 2020. The design was a randomized complete block design with 8 treatments in 4 blocks (Table 1). Field plots were 6×2.5 m. There were 5 cuttings in 2021. The N fertilization was dependent of the treatment and divided over the cuts (75 kg N ha^{-1}: 50-25-0-0-0, 125 kg N ha^{-1}: 75-50-0-0-0, 375 kg N ha^{-1}: 125-125-75-50-0). The K fertilization was 190 kg K ha^{-1}, divided over the cuts (95-63-32-0-0), and was the same for every plot.

Table 1. Treatments of the field trial.

Treatment	Mixture	N fertilization
PR_375N	perennial ryegrass 30 kg ha^{-1}	375 kg N ha^{-1}
PR+CL_125N	perennial ryegrass 30 kg ha^{-1} + white clover 3 kg ha^{-1} + red clover 8 kg ha^{-1}	125 kg N ha^{-1}
PR+CL+P_75N	perennial ryegrass 30 kg ha^{-1} + white clover 3 kg ha^{-1} + red clover 4 kg ha^{-1} + plantain 1.5 kg ha^{-1}	75 kg N ha^{-1}
TF+CL+P+C_75N	tall fescue 35 kg ha^{-1} + white clover 3 kg ha^{-1} + red clover 4 kg ha^{-1} + plantain 1.5 kg ha^{-1} + chicory 1.5 kg ha^{-1}	75 kg N ha^{-1}
PR+CL+P+C_0N	perennial ryegrass 30 kg ha^{-1} + white clover 3 kg ha^{-1} + red clover 4 kg ha^{-1} + plantain 1.5 kg ha^{-1} + chicory 1.5 kg ha^{-1}	0 kg N ha^{-1}
PR+CL+P+C_75N		75 kg N ha^{-1}
PR+CL+P+C_125N		125 kg N ha^{-1}
PR+CL+P+C_375N		375 kg N ha^{-1}

Net field plots of 6×1.5 m were harvested with a Haldrup forage harvester. Dry matter (DM) crop yields of each replicate were calculated after drying the subsample in a forced-draught oven at 70 °C for 72 hours. To determine the botanical composition, a grab subsample in two blocks of treatments PR+CL_125N, PR+CL+P+C_0N, PR+CL+P+C_75N, PR+CL+P+C_125N and PR+CL+P+C_375N were separated into grass, clover, herb and weed components.

Results and discussion

Previous field trials showed that tall fescue seemed to be a better partner for clover in mixture. It germinates and develops more slowly than perennial ryegrass in the weeks after sowing and the first year of production, thus allowing the clover and herbs a better start. Although this was confirmed visually in autumn 2020 and winter 2020-2021, no significant differences in sward composition and DM crop yield (results not shown) were observed between PR+CL+P+C_75N and TF+CL+P+C_75N in the first production year.

Grass-clover with added plantain, or plantain and chicory, can achieve the same yields as perennial ryegrass and grass-clover, even at lower N fertilization levels (Table 2). As expected, the perennial ryegrass is more productive in the first two cuts. In comparison with perennial ryegrass, the grass-clover+herbs treatment follows the same fluctuations as grass-clover in crop yield, and maintains the crop yield in the last two cuts. The herbs percentage in the first two cuts is below 15%. Although it increases over the growing season like clover, the share of clover is always bigger in the sward. The percentage grass still achieves >10% in the last two cuts of grass-clover, although there is hardly any grass harvested (<3%) in the grass-clover+herbs mixtures. Observations of the second production year will reveal if the grass component will be productive again and if the herbs maintain the same share in the sward.

Table 2. Comparison between perennial ryegrass, grass-clover and grass-clover+herbs in the first production year.[1]

Treatment		Cut 1	Cut 2	Cut 3	Cut 4	Cut 5	Total
PR_375N	DM yield (kg ha^{-1})	3,779a	8,050a	4,559a	2,504b	347c	19,239
PR+CL_125N	DM yield (kg ha^{-1})	3,709a	6,744b	4,663a	3,378a	1,107b	19,600
	Grass/clover%	64/28	75/25	34/66	13/87	11/89	
PR+CL+P_75N	DM yield (kg ha^{-1})	3,322b	6,098b	4,462a	3,534a	1,170ab	18,586
PR+CL+P+C_75N	DM yield (kg ha^{-1})	3,341b	4,859b	5,347a	3,553a	1,332a	19,117
	Grass/clover/herbs%	54/28/12	40/46/13	12/50/38	1/73/26	3/60/37	

[1] Mean DM yield (n=4) and botanical composition are shown per cut. Statistical significant differences in crop yield between treatments (Tukey $P<0.05$) are indicated with different letters.

Table 3 shows the effect of N fertilization on grass-clover+herbs. Fertilization with 75 or 125 kg N ha^{-1} had a significantly positive effect on crop yield, while a further increase in N fertilization had no additional effect. Even the difference in crop yield between 75 and 125 kg N ha^{-1} was quite modest. Increasing the N fertilization from zero to 75 kg N ha^{-1} had a positive effect on the share of herbs in the sward in every cut. Further increase up to 375 kg N ha^{-1} lead to a decrease in percentage of herbs. Increasing the N fertilization had a negative effect on the share of clover in all cuts. It seems that 75 kg N ha^{-1} is a good N fertilization level for a grass-clover with added plantain and chicory to achieve high crop yields and maintain and a large share in clover and herbs in the sward. It is important to note that the entire growing season of 2021 was quite wet, resulting in above-average grass yields. The absence of seasonal drought/heat stress on the swards could have influenced our results.

Table 3. Comparison of grass-clover+herbs at several N fertilization levels in the first production year.[1]

Treatment		Cut 1	Cut 2	Cut 3	Cut 4	Cut 5	Total
PR+CL+P+C_0N	DM yield (kg ha^{-1})	2,197c	5,654b	4,620b	3,594	1,249	17,313c
	Grass/clover/herbs%	35/54/7	35/61/3	13/65/22	3/77/20	4/72/24	
PR+CL+P+C_75N	DM yield (kg ha^{-1})	3,341b	6,045ab	4,846ab	3,553	1,332	19,117b
	Grass/clover/herbs%	52/31/13	57/36/4	15/47/38	1/73/26	3/60/37	
PR+CL+P+C_125N	DM yield (kg ha^{-1})	4,304a	6,547a	4,911ab	3,398	1,382	20,542a
PR+CL+P+C_375N	DM yield (kg ha^{-1})	4,421a	6,477a	5,160a	3,684	1,365	21,106a
	Grass/clover/herbs%	70/21/4	82/16/2	43/44/13	37/51/12	17/60/23	

[1] Mean DM yield (n=4) and botanical composition per cut of the sward. Statistical significant differences in crop yield between treatments (Tukey $P<0.05$) are indicated with different letters.

Conclusions

We conclude from the first production year after autumn installation that a sown sward of grass-clover+herbs can reach the same crop yields as grass-clover and perennial ryegrass. N fertilization of 75 kg N ha^{-1} seems to compromise the maximum yield without reducing the share of clover and herbs in the sward during the first year. We observed no positive effects of replacing perennial ryegrass by tall fescue in the grass-clover+herb mixtures. Further observations of this field trial will reveal whether the share of grass will again increase in the coming spring and whether plantain and chicory will maintain their share in the sward.

Supplemental irrigation – a measure to sustain yield in mixed grass-legume ley systems during drought periods

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Abstract

Climate change is altering the conditions for agriculture, resulting in more frequent shortfalls in forage production. In two field experiments carried out in 2021 within established first-year grass-legume leys on the Swedish islands of Öland and Gotland, this study compared four different irrigation treatments in randomised blocks: an unirrigated control, entire-season irrigation (SI), irrigation up to first cut (D1) and irrigation up to second cut (D2). Field measurements showed clear increases in dry matter (DM) yield with irrigation during periods of rainfall deficit. Stored soil water was sufficient to give similar yield levels in all treatments at cut 1. A clear water deficit was seen in the unirrigated control and also the short irrigation treatment D1, with insufficient water to support a second cut. Similar responses to irrigation were observed at the third cut. By cuts 2 and 3, all irrigated treatments had significantly higher yield than the unirrigated control ($P<0.001$). Entire-season irrigation (SI) gave on average 30 and 66% higher DM yield than in the unirrigated control at the Öland and Gotland sites, respectively. No or very weak effects of irrigation on botanical composition were observed.

Keywords: deficit irrigation, first-year ley, dry matter yield, botanical composition

Introduction

Ley, a water-intensive crop, is grown on 37% of arable land in Sweden. In recent years, precipitation deficits during the growing season have led to shortages of forage in Sweden. Future ley production system must be adapted to cope with unpredicted water shortages, since vulnerability and production losses will otherwise occur more frequently in the years ahead. The main objective of this study was to assess the positive effects of irrigation on ley production in terms of higher forage yield and impact on botanical composition.

Materials and methods

The work was conducted in the 2021 growing season, the first year of 3-year field trials comparing (1) drought-tolerant species (50% cocksfoot (*Dactylis glomerata* L.) and 50% lucerne (*Medicago sativa* L.)) and (2) less drought-tolerant species (50% timothy (*Phleum pratense* L.), 30% meadow fescue (*Festuca pratensis* Huds.), and 20% red clover (*Trifolium pratense* L.)). The trials were located at two sites under conventional management at Torslunda on the Swedish islands of Öland (56°63'51"N 16°51'04"E) and under organic management in Lövsta on Gotland (57°54'17"N 18°44'56"E). The experiments comprised four irrigation treatments that were replicated in four blocks (randomized block design) with a total of 16 experimental plots. The irrigation treatments, represented different strategies for supplemental irrigation in terms of irrigation period length. The treatments were: no irrigation (control); supplemental irrigation with no water stress, where irrigation amount was regulated to meet crop water demand and carried out during the entire season when soil moisture level approached 55% plant-available water (treatment SI); deficit irrigation as in SI, but only to the first cut (treatment D1); and deficit irrigation as in SI, but only to the second cut (treatment D2). A soil water balance was calculated at each site on a daily basis to determine the need for irrigation in advance, and also to assess soil moisture changes and water stress during the whole cropping season. Dry matter (DM) yield was measured for each plot at harvest and ley botanical composition was assessed at plot level. Analysis of variance (ANOVA-Main effect) was used to determine if irrigation treatments had a significant effect on yield. The Tukey test was used to identify significant differences between treatments.

Results and discussion

On Öland, precipitation was 30% higher than normal (30-year period 1960-1991) during the 2021 growing season, while on Gotland total precipitation was close to normal (Table 1). This led to the precipitation deficit, and thus irrigation demand, being twice as high in the plots on Gotland as in those on Öland. The precipitation deficit on Öland was covered by the full irrigation in treatment SI (140 mm), while the precipitation deficit on Gotland was covered to 60% by the full irrigation in treatment SI (160 mm).

At cut 1, there were no significant differences in DM yield between unirrigated and irrigated plots on Öland (Table 2). On Gotland, however, there were significant differences in yield at cut 1 in the trial with more drought-resistant species (Table 2). At cut 2, the two treatments irrigated after cut 1 (SI, D2) gave significantly higher yield than the unirrigated control at both experimental sites. At cut 3, the full irrigated treatment (SI) had significantly higher yield than the unirrigated treatments at both sites. On Öland, S1 and D2 gave 25 and 14%, respectively, higher total yield than the unirrigated treatment with drought-tolerant species and 36 and 29% respectively, higher total yield than the unirrigated treatment with less drought-tolerant species. The corresponding yield increase in treatments SI and D2 on Gotland was 63 and 58%, respectively, with drought-tolerant species and 70% and 56%, respectively, with less drought-tolerant species. The increments in kg per mm applied water (water productivity) on Öland varied between treatments, with values ranging from -29 to 40 kg per mm, with the lower value in D1 with drought-tolerant species. On Gotland water productivity showed less variation between the irrigation treatments and ranged from 28 to 43 kg per mm. In contrast to the site on Öland, water productivity was highest in D1 with the drought-tolerant species on Gotland, most likely due to the high precipitation deficit in May (Table 1). Since, in general, water productivity increases when less water is applied, a weighting against yield losses should be assessed to determine the best irrigation strategy for a particular farm (Lindenmayer *et al.*, 2010). No significant differences in species composition were found among the irrigation treatments at either site.

Table 1. Climate and irrigation data (mm) on Öland and Gotland during the growing season 2021 with precipitation (P), average precipitation during years 1961-1990 (P 1961-1990), potential evapotranspiration (ET_0), precipitation deficit (P_{def}) and irrigation amount (Irr) in treatments supplying entire-season irrigation (SI) and irrigation to cut 1 (D1) or to cut 2 (D2), in trials with drought-tolerant and less tolerant species on Öland (conventional) and on Gotland (organic).

Sites	Amount (mm)	April	May	June	July	August	September	Total
Öland	P	8	58	10	48	131	70	325
	P, 1961-1990	28	34	36	57	49	46	250
	ET_0	54	69	118	105	72	44	461
	P_{def}	46	11	107	57	-60	-26	136
	Irr SI	0	20	60	40	20	0	140
	Irr D1	0	20	0	0	0	0	20
	Irr D2	0	20	60	0	0	0	80
Gotland	P	9	34	16	87	96	21	263
	P, 1961-1990	28	30	29	49	49	62	247
	ET_0	54	87	146	131	73	44	535
	P_{def}	45	53	130	44	-23	23	272
	Irr SI	0	25	80	55	0	0	160
	Irr D1	0	25	0	0	0	0	25
	Irr D2	0	25	80	25	0	0	130

Table 2. Ley dry matter (DM) yield in cuts 1-3 and total yield (kg DM ha^{-1}) in the unirrigated control, and in treatments supplying entire-season irrigation (SI) and irrigation to cut 1 (D1) or cut 2 (D2), in trials with drought-tolerant and less tolerant species on Öland (conventional) and on Gotland (organic).[1]

Sites	Drought-tolerant	Cut 1	Cut 2	Cut 3	Total	Relative total yield
Öland	Control	4,669	2,185^{a}	3,050^{a}	9,904^{a}	100
Öland	SI	4,527	3,638^{b}	4,248^{b}	12,414^{b}	125
Öland	D1	4,415	1,927^{a}	2,988^{a}	9,330^{a}	94
Öland	D2	4,670	3,595^{b}	3,034^{a}	11,299^{ab}	114
Gotland	Control	4,578^{a}	1,575^{a}	2,253^{a}	8,407^{a}	100
Gotland	SI	5,208^{b}	5,124^{b}	3,399^{b}	13,731^{b}	163
Gotland	D1	4,915^{ab}	1,948^{a}	2,617^{ab}	9,480^{a}	113
Gotland	D2	5,186^{b}	5,171^{b}	2,959^{ab}	13,317^{b}	158
	Less drought-tolerant	**Cut 1**	**Cut 2**	**Cut 3**	**Total**	**Relative total yield**
Öland	Control	5,156	1,990^{a}	1,809^{a}	8,955	100
Öland	SI	5,382	3,780^{b}	3,029^{b}	12,191	136
Öland	D1	5,168	2,387^{ab}	2,200^{ab}	9,755	109
Öland	D2	5,712	3,654^{b}	2,211^{ab}	11,578	129
Gotland	Control	4,556	1,411^{a}	1,572^{a}	7,539^{a}	100
Gotland	SI	4,816	5,297^{b}	2,727^{b}	12,841^{b}	170
Gotland	D1	5,062	1,752^{a}	1,421^{a}	8,235^{a}	109
Gotland	D2	4,452	5,274^{b}	2,053^{c}	11,788^{b}	156

[1] Different superscript letters (a-c) indicate significant differences between treatments.

Conclusions

Our findings highlight the value of supplemental irrigation as a measure to sustain yields under periods with rain deficit. In the first year of a three-year grass-legume ley, a clear yield increase was observed with irrigation before cuts 2 and 3 under both conventional and organic management. The positive effects on yield were directly linked to duration of irrigation during the growing season under conditions of rain deficit. The effects of supplemental irrigation were positive with both drought-tolerant and less drought-tolerant species mixtures. No or very weak effects of irrigation on ley botanical composition were observed.

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References

Lindenmayer R.B., Hansen N.C., Brummer, J. and Pritchett J.G. (2010) Deficit irrigation of alfalfa for water-savings in the Great Plains and Intermountain West: A review and analysis of the literature. *Agronomy Journal* 103, 45-50.

Theme 4.
Looking for synergy between animal production, grasslands and crops

Reconnecting cropping and livestock operations to enhance circularity and avoid ecological collapse

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Abstract

Agriculture has undergone dramatic changes over the past century. Many would argue that the changes have been unquestionably positive with huge gains in productivity, reduced labour requirements, and alleviation of food insecurity for most people. However, the adoption of increasingly specialized and separated crop and livestock enterprises has also had widespread negative consequences on biodiversity simplification, degradation of groundwater and surface waters with agrochemical pollutants, poor soil health with monoculture crop production, large greenhouse gas emissions from both specialized cropping systems relying on external inputs and concentrated animal feeding operations that accumulate wastes, and general lack of ecological integrity among components of these specialized systems. Integrated systems offer opportunities to reconnect the synergies available when mixed crop-livestock systems rely on organic-based nutrient cycling dynamics, ecologically based weed, insect and disease controls, and system-level sharing of resources in a circular-based agroecosystem. We provide a few examples of how annual and perennial forages can be an integral component of integrated crop-livestock systems, including grazing of cover crops, pasture-crop rotations, and among-farm integration. To be truly sustainable, the ecological integrity of agriculture requires different types of forages utilized across a diverse landscape.

Keywords: ecological integrity, environmental quality, forages, grazing, integrated crop-livestock systems, perennial pasture

Introduction

Modern, industrialized agricultural systems have become highly specialized to focus on economies of scale by maximizing the efficiency of a particular product with as little human labour input as possible. This strategy has been deployed in industrialized countries for both crop and livestock production systems, and the process of this specialization was initiated in the mid-20th century, primarily because of cheap fossil fuel reserves, abundant and low-cost inorganic fertilizer manufacturing, rapid development of mechanization, plant and animal genetic improvements, and scientific understanding of the processes needed to maximize productivity. As a result of these production-focused improvements and a shift towards specialization of either crop production only or livestock production only, agricultural demographics changed to fewer farmer operators over time (Figure 1), but with greater agricultural product output per farm. The average size of a farm in the US has more than tripled since the beginning of the 20th century, and this same magnitude of change has occurred in France since 1950 (Figure 1). In France, the number of farms with mixed crop and livestock operations declined from 55,800 in 2000 to 36,600 in 2016 (INSEE, 2020) while the number of specialized crop farmers increased from 49,500 to 61,500 in the same period (INSEE, 2020).

Distribution of farm size based on land area in the US has shifted in the past half century. In 1964, 26% of farms were very small (<20 ha), 69% of farms were medium-sized (20-400 ha), and 5% of farms were large (>400 ha) (USDA-NASS, 2020). In 2017, proportions of farms were 42, 49, and 9% as small-, medium-, and large-sized. Therefore, although the proportion of large farms increased during this time,

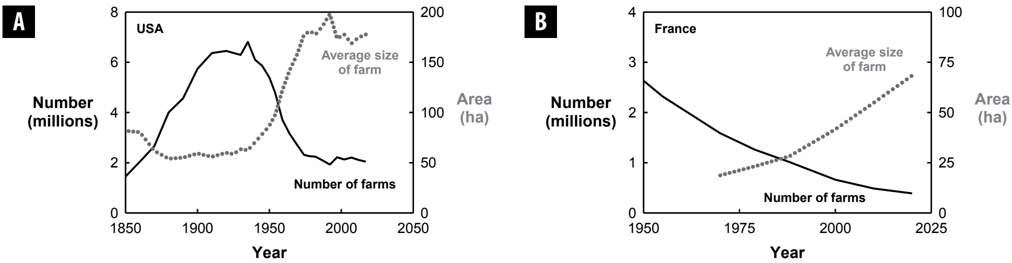

Figure 1. Number of farms (solid line) and average size of farm (dotted line) during a 200-year period in the United States (A) and during a 75-year period in France (B). Data from USDA-NASS (2020) and Agreste (2022).

the proportion of medium-sized farms was mainly replaced with small-sized farms rather than the shift to larger farms. A similar shift in farm size distribution has occurred in France (Figure 2). The proportion of small-sized farms has steadily declined from ~90% of all farms in 1892 to <50% of all farms in 2010, while the proportions of medium- and large-sized farms has steadily increased, particularly since 1980.

Livestock inventories on the average farm in the US have generally risen dramatically over the past century. Similar change in all livestock inventories has occurred in France. In the US, average inventories of dairy have increased from 13 head per farm in 1964 to 175 head in 2017. Average swine inventories have increased from 37 head per farm in 1964 to 1,089 head in 2017. Average layer chicken inventory per farm has increased from 162 in 1964 to 1,584 in 2017. The beef production system in the US has three distinct phases from birth to slaughter, i.e. cow-calf operations spread throughout the country, stocker grazing of weaned calves throughout the southern and eastern US, and feedlot finishing in the Great Plains region (Franzluebbers *et al.*, 2011). Therefore, portions of the beef production system are geographically distributed and small scale, but the finishing phase is typically highly concentrated and specialized. A similar geographical movement occurs in Europe wherein calves produced in France are shipped to Italy to be fattened.

Distribution of livestock by inventory size in the US illustrates the trend toward larger operations, but also the baseline condition for smaller operations to potentially reclaim a more diversified and integrated agricultural system. For example, the average inventory of layer hens throughout the US has been relatively constant at 359.5 million in 1920 and 368.2 million in 2017 (USDA-NASS, 2020). The number of farms with small layer flocks far dominates the farm landscape, and yet the total inventory of layers is dominated by a relatively small proportion of the farms (Figure 3).

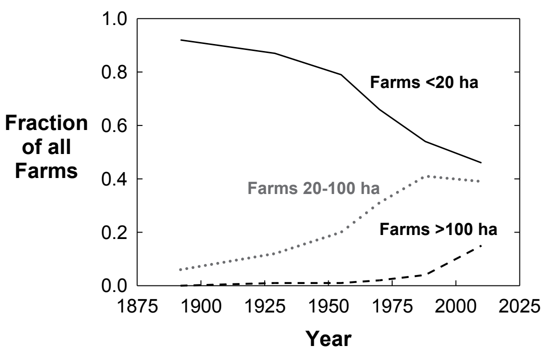

Figure 2. Fraction of farms in size categories over time in France. Data from Agreste (2022).

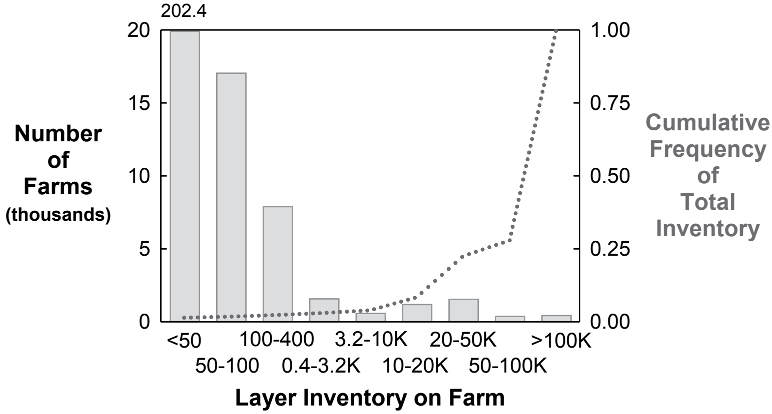

Figure 3. Number of farms having layer hens (bars) and cumulative frequency of inventory (dotted line) as affected by inventory size classification in 2017 in the United States. Data from USDA-NASS (2020).

In the US, most farmers have expertise on smaller land areas of croplands and smaller herd sizes of livestock. However, the agricultural enterprise as a whole is dominated by a small minority of large, specialized farmers, who appear to have large political influence. This may be one reason why government policies continue to favour industrial modes of production without sufficient attention to agroecological approaches. Policies may be easier and more efficient when dealing with fewer farms, especially when ignoring the indirect costs of this specialization on human health, environmental pollution and ecological integrity. When faced with public recognition and demand for pollution control from confined animal feeding operations (CAFOs), large industrial farms often react with even greater specialization and techno-engineering solutions. For example, animal waste flushed from CAFOs is often held in a settling pond and water volume reduced by spraying onto a nearby field. The fate of settled solids is less clear, and not necessarily widely addressed due to an issue that has not yet emerged from relatively new lagoons that have not needed to be removed of solids to date. Likewise in France, technology-centric solutions are becoming dominant in public debates on agriculture, for example with genetic engineering of plants and animals, robotics, etc. Low-tech solutions based on collective intelligence and harmony with nature are not equally promoted, despite sufficient knowledge of the benefits of agroecology on production and protection of the environment. The livestock unit efficiency and economic outcome of high-technology, specialized systems are considered high, especially when costly infrastructure is supported by low-cost loans and can be amortized over years. Government incentives for co-locating large processing facilities nearby are often pitched as economic incentives to hire labour and deliver benefits to local communities. The public generally doesn't know the working conditions of such operations and consumers appreciate low-cost food supplies. However, large industrial agricultural systems cause enormous impacts on the environment and the health of rural communities. In Europe, agri-food industries may lobby against policies of which they disapprove; for example, to improve animal welfare or environmental integrity (Swinnen *et al.*, 2021), and then may even threaten to relocate their operations elsewhere. When forced, large agricultural industries can clean up nutrient losses by storing more of the nutrients in engineered lagoons. However, this may cause greater greenhouse gas emissions, and engineered solutions may result in gas digesters or lagoon covers to minimize losses. Natural disasters occur, and these engineered operations are prone to failure under stressful conditions. The threat of enormous nutrient losses following hurricanes, tornadoes, and floods is real. Odour emission from CAFOs can drive rural communities into decline. Animal welfare in CAFOs is a topic that is debated, as manufactured housing offers feeding and environmental comforts that open spaces do not, and yet natural movements and exploration on the landscape are hindered. Farmer well-being is another subject of importance in this shift from integrated to specialized agricultural systems, as farming has had some of the highest suicide rates in Europe. French farmer suicide rate is 20% greater than that of the general population, and 30% greater for dairy cattle farmers (Santé Publique France, 2018). Ecologically focused non-government organizations are gaining popularity and the livestock sector is under increasing pressure to change. The industrial model can be seen as at the brink of disaster with the looming threats of environmental, pandemic, and geopolitical disturbances. This is certainly not the idyllic view of agriculture that many people might have considered. There must be another way...

Recognizing the need for change

A circular economy that is sustainable for all sectors recognizes that each component has intrinsic value and that final products cannot be the only outcome of importance. The process of the economy must be valued at every step. Agroecology, as one element of a circular economy, can be extended to the entire agri-food system beyond the farm gate into the rural landscape and community (Francis *et al.*, 2003). A circular agroecological system has many components that can be defined as serving one or more functions, and the extrinsic values of those components should be considered in addition to the intrinsic value of end-products of the agricultural production system. As an analogy, a centrifuge containing several differently sized critical components linked to a central axis will rotate effectively at low speed, but when turned to high speed will develop a wobble that can lead to dysfunction and breaking of the machine. In a similar manner, industrialized agriculture without a balanced load of components tied to the axis will spin out of control and lead to disaster. A balanced, agroecologically-based system will have balanced inputs and outputs that can lead to smooth operation for long periods of time. In a more complex manner, those components that may be neglected in an industrial agroecosystem and lead to disaster of the whole machine can be considered necessary for an agroecological system to function smoothly and over a long period of time. In agroecologically based systems, emphasis is put on slowly changing variables (e.g. soil organic matter content, farmer's knowledge) that shape the ability of the system to respond to perturbations over both the short and long term, whereas industrial-based agriculture has mainly a short-term focus on variables of immediate interest (e.g. plant-available N). We explore some of these functions of agroecological systems through the integration of livestock with crops in the following sections. Placing value on all components of the system allows the system to function efficiently to meet production, environmental, and social goals over time.

It is argued that forages and the products derived from grasslands and grazing lands when integrated with other components of agricultural production contribute to a healthy, circular production system that can lead to a sustainable food supply to support thriving human populations while contributing to the vital ecosystem services that are needed at full capacity to withstand the threats of climate change. These ecosystem services derived from an agroecological approach with integration of crops and livestock will reduce contributions to greenhouse gas emissions, water quality impacts, soil loss, and suppression of biodiversity (Martin *et al.*, 2020). Therefore, integrated agricultural systems that rely on natural processes of nutrient cycling, abundant capture of sunlight to drive ecosystem processes, soil biological activity to promote soil health and filter water, and biotic interactions with the environment to control pests and diseases are fundamental to a better way of agricultural production for a sustainable future.

Points along a circular agricultural system with integration of crops and livestock

Nature has an inherent capacity to return to rhythms of natural cycles. When land is denuded, a diversity of plants is recruited to heal the scars. When inorganic nutrients are depleted in the soil, plant uptake and decomposition create an organic cycle to limit further losses. Ecological integrity is returned to systems with limited disturbance. The relatively recent move in human history towards crop and livestock specialization and monoculture cultivation should likely have never been considered sustainable, but intensive use of fertilizers, pesticides and antibiotics allowed this model to overcome its ecological deficiencies. Today, interest is growing for more diversified agricultural operations that rely on the restorative nature of forages and ley systems on ecosystem services.

Historically, crop cultivation relied on livestock production for soil fertility with animal manures and rotation with perennial pastures, i.e. achieving near balance between production and respiration (Billen *et al.*, 2010). In fact, evidence exists from botanical macro-remains that cereals, wetland and woodland pastures, and fallow fields occurred together from 2920 to 2620 BCE in the Netherlands (Kooistra, 2008). In the Mediterranean area of southern France, evidence from the 5th to 12th century CE suggests farming was mixed, combining cereals, oilseeds, pulses, vines and fruits with wetlands and meadows (Ruas, 2005). Ferdière (2021) identified the first cases of specialization (wine production in the Narbonne area) in Roman Gaul, although mixed farming would have largely been the rule at that time, together with the practice of crop rotation and green manuring of pastures. The advent of widespread global trade of food and feed commodities in the industrial era has altered nutrient balances for many countries throughout the world (Galloway *et al.*, 2007).

How should we utilize forages and grasslands in a new era of high-productivity farming systems? We do not envision going back to historical low productivity systems in this dawning of a new era of integrated crop-livestock systems. We have at our disposal many modern agricultural management techniques, such as conservation tillage, high N-fixing legumes, and availability of additional organic amendments, that can keep productivity at high levels, but importantly that can be used to minimize environmental degradation and hopefully improve ecological conditions of agricultural landscapes. An important outcome of this integrated approach will be to have vibrant rural communities that can be sustained with the diversity of farming practices needed to balance productivity and environmental quality.

A few examples of contemporary success of agroecological approaches are briefly summarized here to illustrate the potential for greater profit with organic premiums and greater yield due to rotational benefits. In the Palouse region of the northwestern US, growing organic grains following alfalfa (*Medicago sativa*) led to greater return over variable costs (\$615 ha^{-1} yr^{-1}) than a typical conventional rotation (\$477 ha^{-1} yr^{-1}), despite quinoa (*Chenopodium quinoa*) as an alternative crop performed poorly (Wiema *et al.*, 2020). The authors observed that advances in organic weed control and development of regionally adapted quinoa varieties could help reduce the risk to farmers attempting cropping system diversification. Incorporating a leguminous forage crop, such as alfalfa, in the rotation was crucial for the economic and agronomic performance of these dryland organic cropping systems. In the Midwest US, more diverse cropping systems that included forages in the rotation [corn (*Zea mays*)-soybean (*Glycine max*)-small grain-alfalfa) had equal or greater harvested yields and profit than in more simplified rotations, despite reductions of agrochemical inputs (Davis *et al.*, 2012). Weeds were effectively suppressed and ecotoxicity of the more diverse systems was orders of magnitude lower than in the conventional system. In the southeastern US on land that was previously in permanent pasture, summer grain and stover production were greater under no-till than under conventional-till, but winter grain and stover production were unaffected by tillage system (Franzluebbers and Stuedemann, 2007). In addition, both winter and summer cover crops were more productive under no-till than under conventional-till. Both rye and pearl millet cover crops provided an abundant and high-quality diet for either yearling calves or cow-calf pairs for 26-77 days in the early spring. Calf gain on cover crops was 250±97 kg ha^{-1} under no-till, which was an average of 21% greater than under conventional-till. Net return over variable costs was greater with grazing of cover crops, averaging –\$63 ha^{-1} when not grazed and \$302 ha^{-1} when grazed.

Each particular ecoregion has a set of defining characteristics from which appropriate agricultural operations will be possible (Bell *et al.*, 2014; Moraes *et al.*, 2014; Peyraud *et al.*, 2014; Sulc and Franzluebbers, 2014). Not all farming practices will be possible in all environments. Not all management styles will be acceptable in all types of farming communities. Therefore, matching the genetics of plant and animal species within a particular environmental setting might require unique management styles based on surrounding economic conditions, as well as cultural heritage of rural communities. These factors fit well within the G × E × M × S (genetics × environment × management × socio-economic) research platform (https://agroinformatics.org/). In some regions, integrated crop-livestock systems might deploy cattle grazing winter cover crops to consume high-quality forages at a time of low forage availability from perennial pastures, and keep crop fields productive and with sufficient surface residue using no-till to foster improved soil health and efficient nutrient cycling (Sulc and Franzluebbers, 2014). In other regions, perennial pastures may be needed to provide livestock forage, and rotated periodically with crops to capture the improved soil health benefit of soil organic matter accumulation during the pasture phase along with the nutrient carryover to the crop phase, as well as to reduce weed and pest issues in cropping phases (Martin *et al.*, 2020). Other regions may require agreements between specialized crop and livestock farmers so that land application of animal manures can avoid environmental contamination of sensitive portions of a watershed (Ryschawy *et al.*, 2017). Examples of these approaches are described in the following section focusing on key features that can alter outcomes of productivity, profitability, environmental quality, and/or ecological interactions.

Grazing of cover crops

The soil health movement in the US has created interest (Wood and Bowman, 2021) and increasing implementation of cover cropping (Dunn *et al.*, 2016), particularly in the eastern US (Bastos-Martins *et al.*, 2021). Along with this interest, several literature reviews have shown the positive impacts of cover crops on soil properties and processes (Farmaha *et al.*, 2022; Poeplau and Don, 2015; Schipanski *et al.*, 2014). More recently, a few multiple-year research studies have been initiated and can now provide some insights into the possible negative and positive impacts expected with grazing of cover crops on system productivity, soil responses, and environmental quality. Several different types of winter cover crops can provide timely forage with high nutritive value for grazing by ruminant livestock. Good average daily gain of 0.8 to 1.1 kg d^{-1} for growing cattle (Franzluebbers and Stuedemann, 2007) can be expected from these winter forages that may be grazed starting in fall if planted following an early harvest of summer cash crop, and lightly over the winter and again intensely in the spring. The periods of transition between cash crops and cover crops and vice versa can lead to significant N mineralization and susceptibility of that inorganic N to volatile or leaching loss, but this effect can be minimized with timely planting operations. The stage of plant development at the time of grazing can have an impact on animal performance, as nutritive value for most species will decrease with increasing maturity when entering the reproductive stage. An oft-voiced concern by cropland farmers is the potential compaction of livestock grazing on a field with winter cover crops. Intensive cattle trampling with significant damage of the sward and reduction in soil animal populations can occur (Cluzeau *et al.*, 1992), and can cause significant pugging of wet-prone soil (Pietola *et al.*, 2005), but the majority of evidence from upland soils indicates that soil bulk density is increased only a small amount by repeated cattle trampling on well-vegetated land (Blanco-Canqui *et al.*, 2020). Kelly *et al.* (2021) also found that greater bulk density with cattle grazing of summer cover crops in no-till wheat (*Triticum aestivum*) cropping systems in the Great Plains of the US was below a critical threshold. In a study with several years of cash cropping with grazing of cover crops following no-till termination of a perennial pasture, the impact of moderate animal traffic was mitigated by the robust soil surface organic matter present (Franzluebbers and Stuedemann, 2008).

Grazing of cover crops can have mixed effects on subsequent crop yield. Sometimes a negative impact of cover crop grazing occurs on subsequent yield, perhaps due to exposure of soil to enhanced water evaporation (Franzluebbers and Stuedemann, 2007). As well, greater subsequent crop growth has been attributed to enhanced fertility status with greater soil organic matter (George *et al.*, 2013). However, most research findings have found little difference in subsequent crop growth during relatively short-term evaluations, while a variety of ecosystem services can be simultaneously increased (Blanco-Canqui *et al.*, 2015). A balance must be achieved between the benefit of reducing nitrate-N leaching with cover cropping and the negative impact on subsequent cash crop growth from reduction in soil water and inorganic N removal (Meyer *et al.*, 2022).

Pasture-crop rotations

Long rotation sequences that included multiple years of perennial pasture in rotation with grain and fibre crops were common practice in many temperate agricultural regions prior to the development of fossil-fuel powered technologies post World War II (Aref and Wander, 1997). Modern pasture-crop rotations are still important for some dairy production systems, such as in the Midwest and Northeast US and in Northwest Europe. Organic farmers are also more prone to use pasture-crop rotation sequences to quickly rebuild soil organic matter during the pasture phase that is lost during the tilled crop phase (Mayer *et al.*, 2015), as well as to control perennial weeds that are more difficult to kill with tillage alone. Some pasture phases may be short-term with 1-2-year green manure crops, such as with grass-clover mixtures that are either cut for hay or grown simply for biomass accumulation with significant N fixation from the legume component (Blanco-Canqui *et al.*, 2017). In Europe, ryegrass (*Lolium multiflorum*)-red clover (*Trifolium pratense*) are often used for short-term hay harvest, while ryegrass-white clover (*Trifolium repens*) are used in longer term grazed pastures followed by spring cereals or maize on beef and dairy farms. The benefits of pasture-crop rotations are numerous to both pasture and crop production phases, as well as more importantly to improvements in soil organic matter, fertility and environmental quality. Greater diversity of plant species with these long rotations can have ecological benefits far beyond the immediate farm gate, such as to avian and transient wildlife populations, biological insect control and watershed runoff quality improvement (Duru *et al.*, 2014; Havet *et al.*, 2014; Moraine *et al.*, 2017; Peyraud *et al.*, 2014).

The production, environmental, and ecological implications of pasture-crop rotations in different environments have been summarized elsewhere (Franzluebbers *et al.*, 2014; Martin *et al.*, 2020). The intrinsic value of pasture-crop rotations to a more sustainable, ecologically based approach to agriculture cannot be overstated. The enormous benefits imparted on soil organic matter, soil health, ecosystem stability, water quality protection, greenhouse gas mitigation and biodiversity enhancement contribute significantly to sustained production performance. This was evident in mature agricultural systems prior to the development of specialized farming approaches post-World War II. Although often considered ecologically important for productive use of land unsuitable for cropping, perennial grasslands serving as forage for grazing livestock may potentially have an equal value for restoring inherent soil fertility when rotated with croplands on prime cropland (Franzluebbers and Gastal, 2019). Modern agricultural techniques of improved crop and forage genetics, integrated pest management, and conservation tillage management will likely make future pasture-crop rotations far more productive and ecologically resilient than historical pasture-crop rotations. The flexibility of having perennial grassland in a crop rotation allows for organic amendments to be used effectively, as well as to wean agricultural enterprises from intensive pesticide usage.

Among-farm integration

The use of integrated crop-livestock systems has diminished, and the trend towards specialization continues. Reverting to integrated systems may not be readily feasible, as the infrastructure investments (specific machinery, fencing, layout of buildings) may be too expensive, human knowledge and technical skills required may have been lost in the previous transition to specialization, or the essential veterinary and slaughterhouse support services may not be present in some regions. Against these challenges, integrated crop-livestock systems might rather be organized beyond the farm level (Martin *et al.*, 2016; Moraine *et al.*, 2014). Various types of collaboration among farms are possible according to the level of spatial, temporal, and organizational coordination. This may be from exchange of basic materials (straw, grain, hay, manure) among farms to coordination of land-use collaborations, livestock movements, and sharing of equipment and workforce among more ambitious groups of farmers.

In addition to the ecological and agronomic benefits, reconnecting crops, pastures, and livestock allows fuel savings with livestock substituting machinery interventions for mowing and manure spreading (Ryschawy *et al.*, 2021). Cooperative regional markets might be developed to spread risks (Asai *et al.*, 2018). Weather disasters that render crops too damaged for regular sale could be contracted with livestock farmers for sacrificial crop grazing. Cooperative markets might also be a way to reduce income variability due to volatility of the conventional market. Cover crop grazing supports the joint production of food and feed on the same parcel of land and can be used as a marketing strategy to increase income per unit area (Ryschawy *et al.*, 2021). Finally, reconnection of crops and livestock offers the opportunity to spare unsuitable land from cultivation of crops, and therefore, to use resources more efficiently.

Reconnecting crops, pastures, and livestock among farms may also stimulate knowledge exchange among farmers and their adaptive capacity to respond to problems (Stark *et al.*, 2021). The most ambitious cases of reconnection can even include sharing labour of employees, task delegation, or shared tasks between farmers (e.g. making silage), as well as shared equipment to increase productivity (Andersson *et al.*, 2005). This could help reduce peak workload issues on some farms. Multiple socio-technical constraints remain to attain more integrated systems, such as lack of knowledge of the benefits, lack of governance models, costs of logistics, and lack of suitable equipment (Asai *et al.*, 2018). These constraints increase the costs for information gathering, collective decision-making, and implementation of circular systems.

Conclusions

Integrated crop-livestock systems have been explored and continue to gain interest in the scientific community, as we know that synergies among these agricultural components are necessary for developing sustainable production systems. It takes work in the field and on paper to develop systems that are profitable and protective of the environment. Agricultural systems are meant to be complicated to develop resilience of systems within a farm and region. Simplification cannot be our fate! However, relatively easy steps are possible to begin to reconnect cropping and livestock operations, such as routine application of moderate quantities of animal manure to cropland, dedicated planting of cover crops and stocking with grazing livestock, developing a diversity of ecologically relevant rotations with perennial forages and cash crops, and within watershed trading of grains to feed local livestock and distribution of animal manures to avoid environmental contamination. These steps can help develop resilience to climate change, mitigate against greenhouse gas emissions, build soil health, avoid environmental degradation, and foster biodiversity.

Annual and perennial forages are viewed as a necessary component of a circular, healthy, and sustainable food production system. Pasture-based livestock contribute to animal welfare and to the positive image of herbivores in farming systems. Well-managed grasslands support biodiversity enrichment, make landscapes more hydrologically functional, assist in regulating biogeochemical cycles, reduce the need for pesticides, store significant quantities of carbon in soil, and resist soil erosion. A diversity of approaches for integration of forages into modern cropping systems will be needed to adjust to the wide array of environmental, ecological, and social conditions in different parts of the world.

References

Agreste (2022) Chiffres et analyses. Accessed at: https://agreste.agriculture.gouv.fr/

Andersson H., Larsén K., Lagerkvist C.-J., Andersson C., Blad F., Samuelsson J. and Skargren P. (2005) Farm cooperation to improve sustainability. *Ambio* 34, 383-387.

Aref S. and Wander M.M. (1997) Long-term trends of corn yield and soil organic matter in different crop sequences and soil fertility treatments on the Morrow Plots. *Advances in Agronomy* 62, 153-197.

Asai M., Moraine M., Ryschawy J., de Wit J., Hoshide A.K. and Martin G. (2018) Critical factors for crop-livestock integration beyond the farm level: A cross-analysis of worldwide case studies. *Land Use Policy* 73, 184-194.

Bastos-Martins L., Rejesus R.M., Reberg-Horton C. and Myers R.L. (2021) Understanding the market for cover crop seeds in the United States: Background and potential policy directions. *Journal of Soil and Water Conservation* 76, 83A-88A.

Bell L.W., Moore A.D. and Kirkegaard J.A. (2014) Evolution in crop–livestock integration systems that improve farm productivity and environmental performance in Australia. *European Journal of Agronomy* 57, 10-20.

Billen G., Beusen A., Bouwman L. and Garnier J. (2010) Anthropogenic nitrogen autotrophy and heterotrophy of the world's watersheds: Past, present, and future trends. *Global Biogeochemical Cycles* 24, https://doi.org/10.1029/2009GB003702.

Blanco-Canqui H., Drewnoski M.E., MacDonald J.C., Redfearn D.D., Parsons J., Lesoing G.W. and Williams T. (2020) Does cover crop grazing damage soils and reduce crop yields? *Agrosystems, Geosciences & Environment* 3, e20102.

Blanco-Canqui H., Francis C.A. and Galusha T.D. (2017) Does organic farming accumulate carbon in deeper soil profiles in the long term? *Geoderma* 288, 213-221.

Blanco-Canqui H., Shaver T.M., Lindquist J.L., Shapiro C.A., Elmore R.W., Francis C.A. and Hergert G.W. (2015) Cover crops and ecosystem services: Insights from studies in temperate soils. *Agronomy Journal* 107, 2449-2474.

Cluzeau D., Binet F., Vertes F., Simon J.C., Riviere J.M. and Trehen P. (1992) Effects of intensive cattle trampling on soil-plant-earthworms system in two grassland types. *Soil Biology and Biochemistry* 24, 1661-1665.

Davis A.S., Hill J.D., Chase C.A., Johanns A.M. and Liebman M. (2012) Increasing cropping system diversity balances productivity, profitability and environmental health. *PLoS ONE* 7, e47149.

Dunn M., Ulrich-Schad J.D., Prokopy L.S., Myers R.L., Watts C.R. and Scanlon K. (2016) Perceptions and use of cover crops among early adopters: Findings from a national survey. *Journal of Soil and Water Conservation* 71, 29-40.

Duru M., Jouany C., Le Roux X., Navas M.L. and Cruz P. (2014) From a conceptual framework to an operational approach for managing grassland functional diversity to obtain targeted ecosystem services: Case studies from French mountains. *Renewable Agriculture and Food Systems* 29, 239-254.

Farmaha B.S., Sekaran U. and Franzluebbers A.J. (2022) Cover cropping and conservation tillage improve soil health in the southeastern United States. *Agronomy Journal* https://doi.org/10.1002/agj2.20865

Ferdière, A. (2021) Agriculture in Roman Gaul. In: Hollander D. and Howe T. (eds.) *A companion to ancient agriculture*, 1st ed. John Wiley & Sons, Inc., pp. 447-477.

Francis C., Lieblein G., Gliessman S., Breland T.A., Creamer N., Harwood R., Salomonsson L., *et al.* (2003) Agroecology: The ecology of food systems. *Journal of Sustainable Agriculture* 22, 99-118.

Franzluebbers A.J. and Gastal F. (2019) Building agricultural resilience with conservation pasture-crop rotations. In: Lemaire G., Carvalho P.C.F., Kronberg S. and Recous S. (eds.) *Agroecosystem diversity: reconciling contemporary agriculture and environmental quality*. Elsevier, Academic Press, pp. 109-121.

Franzluebbers A., Steiner J., Karlen D., Griffin T., Singer J. and Tanaka D. (2011) Rainfed farming systems in the USA. In: Tow P., Cooper I., Partridge I. and Birch C. (eds.) *Rainfed farming systems*. Springer, Dordrecht, the Netherlands, pp. 511-560.

Franzluebbers A.J. and Stuedemann J.A. (2007) Crop and cattle responses to tillage systems for integrated crop–livestock production in the Southern Piedmont, USA. *Renewable Agriculture and Food Systems* 22, 163-180.

Franzluebbers A.J. and Stuedemann J.A. (2008) Soil physical responses to cattle grazing cover crops under conventional and no tillage in the Southern Piedmont USA. *Soil and Tillage Research* 100, 141-153.

Galloway J.N., Burke M., Bradford G.E., Naylor R., Falcon W., Chapagain A.K., *et al.* (2007) International trade in meat: The tip of the pork chop. *Ambio* 36, 622- 629.

George S., Wright D.L. and Marois J.J. (2013) Impact of grazing on soil properties and cotton yield in an integrated crop–livestock system. *Soil and Tillage Research* 132, 47-55.

Havet A., Coquil X., Fiorelli J.L., Gibon A., Martel G., Roche B., Ryschawy J., Schaller N. and Dedieu B. (2014) Review of livestock farmer adaptations to increase forages in crop rotations in western France. *Agriculture, Ecosystems & Environment* 190, 120-127.

Kelly C., Schipanski M.E., Tucker A., Trujillo W., Holman J.D., Obour A.K., Johnson S.K., Brummer J.E., Haag L. and Fonte S.J. (2021) Dryland cover crop soil health benefits are maintained with grazing in the U.S. High and Central Plains. *Agriculture, Ecosystems and Environment* 313, 107358.

Kooistra L.I. (2008) Vegetation history and agriculture in the cover-sand area west of Breda (province of Noord-Brabant, The Netherlands). *Vegetation History and Archaeobotany* 17, 113-125.

Martin G., Moraine M., Ryschawy J., Magne M.-A., Asai M., Sarthou J.-P., Duru M. and Therond O. (2016) Crop-livestock integration beyond the farm level: A review. *Agronomy for Sustainable Development* 36, 53.

Martin G., Durand J.L., Duru M., Gastal F., Julier B., Litrico I., Louarn G., *et al.* (2020) Role of ley pastures in tomorrow's cropping systems. A review. *Agronomy for Sustainable Development* 40, 17.

Mayer J., Gunst L., Mäder P., Samson M.-F., Carcea M., Narducci V., Thomsen I.K. and Dubois D. (2015) Productivity, quality and sustainability of winter wheat under long-term conventional and organic management in Switzerland. *European Journal of Agronomy* 65, 27-39.

Meyer N., Bergez J.-E., Justes E. and Constantin J. (2022) Influence of cover crop on water and nitrogen balances and cash crop yield in a temperate climate: A modelling approach using the STICS soil-crop model. *European Journal of Agronomy* 132, 126416.

Moraes A., Carvalho P.C.F., Anghinoni I., Lustosa S.B.C., Andrade-Costa S.E.V.G. and Kunrath T.R. (2014) Integrated crop–livestock systems in the Brazilian subtropics. *European Journal of Agronomy* 57, 4-9.

Moraine M., Duru M., Nicholas P., Leterme P. and Therond O. (2014) Farming system design for innovative crop-livestock integration in Europe. *Animal* 8, 1204-1217.

Moraine M., Duru M. and Therond O. (2017) A social-ecological framework for analyzing and designing integrated crop–livestock systems from farm to territory levels. *Renewable Agriculture and Food Systems* 32, 43-56.

Peyraud J.-L., Taboada M. and Delaby L. (2014) Integrated crop and livestock systems in Western Europe and South America: A review. *European Journal of Agronomy* 57, 31-42.

Pietola L., Horn R. and Yli-Halla M. (2005) Effects of trampling by cattle on the hydraulic and mechanical properties of soil. *Soil and Tillage Research* 82, 99-108.

Poeplau C. and Don A. (2015) Carbon sequestration in agricultural soils via cultivation of cover crops – A meta-analysis. *Agriculture, Ecosystems and Environment* 200, 33-41.

Ryschawy J., Martin G., Moraine M., Duru M. and Therond O. (2017) Designing crop–livestock integration at different levels: Toward new agroecological models? *Nutrient Cycling in Agroecosystems* 108, 5-20.

Ryschawy J., Tiffany S., Gaudin A., Niles M.T. and Garrett R. (2021) Moving niche agroecological initiatives to the mainstream: A case-study of sheep-vineyard integration in California. *Land Use Policy* 109, 105680.

Ruas M.P. (2005) Aspects of early medieval farming from sites in Mediterranean France. *Vegetation History and Archaeobotany* 14, 400-415.

Santé Publique France (2018) Le suicide des agriculteurs Available at: https://www.santepubliquefrance.fr/les-actualites/2018/le-suicide-des-agriculteurs

Schipanski M.E., Barbercheck M., Douglas M.R., Finney D.M., Haider K., *et al.* (2014) A framework for evaluating ecosystem services by cover crops in agroecosystems. *Agricultural Systems* 125, 12-22.

Stark F., Ryschawy J., Grillot M., Mettauer R., Martin G., Cassagnes A., Bertin Y. and Moraine M. (2021) Biotechnical and sociotechnical conditions required to engage an agroecological transition: A case-study of organic sheep crop integration in South-western France. *Landscape 2021 Congress, Diversity for Sustainable and Resilient Agriculture*, Online, 20-22 September 2021.

Sulc R.M. and Franzluebbers A.J. (2014) Exploring integrated crop-livestock systems in different ecoregions of the United States. *European Journal of Agronomy* 57, 21-30.

Swinnen J., Olper A. and Vandevelde S. (2021) From unfair prices to unfair trading practices: Political economy, value chains and 21st century agri-food policy. *Agricultural Economics* 52, 771-788.

USDA-NASS (National Agricultural Statistics Service) (2020) *Census of Agriculture*. United States Department of Agriculture, Washington, DC, USA. Available at: https://www.nass.usda.gov/AgCensus/

Wiema R.A., Carpenter-Boggs L.A., Crowder D.W., Murphy K.M. and Reganold J.P. (2020) Agronomic and economic performance of organic forage, quinoa, and grain crop rotations in the Palouse region of the Pacific Northwest, USA. *Agricultural Systems* 177, 102709.

Wood S.A. and Bowman M. (2021) Large-scale farmer-led experiment demonstrates positive impact of cover crops on multiple soil health indicators. *Nature Food* 2, 97-103.

Herbage production and nutritive value of timothy fertilized according to the YARA crop nutrition programme

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Abstract

Estonian and Finnish timothy (*Phleum pratense* L.) cultivars were studied for dry matter yield (DMY) and nutritive value (NV) differences in a field trial during 2019-2021. The studied experimental factors were: cultivars, four-day harvest delays (after the first and second cuts) and supplemental fertilization pursuant to the YARA crop nutrition programme (YARA) that entails application of N 189, P 10, K 108, Mg 19, S 27 kg, + B 315, Zn 45 and Se 14 g ha^{-1} $year^{-1}$. This was compared with limited plant nutrition (N 120 and K 50 kg ha^{-1}). Cultivars affected DMY and thirteen herbage NV characteristics. Harvest delays led to significant increase in herbage DMY and Ca content, twelve characteristics became degraded and P contents remained unchanged. YARA increased the DMY but eleven NV characteristics of timothy deteriorated due to higher content of fibre and a dilution effect of valuable nutritional factors in the larger amount of biomass (by 46% compared with NK-fertilization). Herbage P concentration was the only characteristic that remained stable across the management regimes. Significant deterioration of NV took place in timothy in just four hot days. Instead of visual assessment, determination of cutting time based upon the summation of effective air temperatures could provide a proper criterion for optimal harvest timing.

Keywords: herbage yield, cultivars, harvest delay, macro elements

Introduction

Timothy (*Phleum pratense* L.) is a widely cultivated forage grass in northern latitudes of the temperate climate zone. It has superior tolerance to adverse wintering conditions and fungal pathogens. Its limited cultivation is caused by slow regrowth and rapidly deteriorating nutritive value (NV) after optimal harvest time. The species has intensive growth during long days in summer, which speeds up the plants' development. Timothy forms culms in its regrowth, which decreases leaf-to-culm ratio and leads to poor forage digestibility. In addition, cultivars' maturity, environment and fertilization can affect forage NV. Besides continued efforts to enhance biomass yields through breeding, in timothy the improvement of NV attracts more attention.

Since the registration of cv. Tika in 1992 and Tia in 1993, timothy has not been bred further in Estonia. Among Finnish cultivars, registered in 2002-2019, Dorothy and Rhonia are selected for enhanced herbage production, while Tuure and Uula maintain their superior NV for a longer period, enabling more flexible harvest management. Genetic variability for digestibility decline with maturity was reported in timothy by McElroy and Christie (1986). We verified the expression of reported virtues of Finnish cultivars in Estonian conditions and measured the effect of enhanced and versatile fertilization on herbage production and NV.

Materials and methods

Cattle slurry (20 m^3 ha^{-1}) was incorporated into the soil of the experimental field directly before tillage. Two Estonian (Tika and Tia) and four Finnish timothy cultivars (Dorothy, Rhonia, Tuure and Uula) were seeded on 7 June 2018. In the harvest years 2019-2021 two fertilization regimes were implemented: (1) Traditional – N 69 kg ha^{-1} as NH_4NO_3 and K 50 kg ha^{-1} as KCl applied between 10 and 20 April, plus N 52 kg ha^{-1} as NH_4NO_3 between 8 and 26 June for the second crops; and (2) the YARA crop nutrition programme (further YARA) composed of three applications of Yara Mila (YM) fertilizers with trace elements – N 90, P 10, K 56 and S 14 kg ha^{-1} as YM 20-5-15 applied between 18 and 27 March followed by two applications of YM 22-0-14 equivalent to N 66, K 35, Mg 13, S 9 kg ha^{-1} between 8 and 29 June, and N 33, K 17, Mg 6, S 5 kg ha^{-1} between 30 July and 31 August. Foliar fertilizer Yara Vita Gramitrel (contains N, Mg, Mn, Zn, Cu) was sprayed (1 l ha^{-1}) two weeks before harvesting the first crops of YARA treatment on 1 June 2020 and 25 May 2021.

Half of the plots were harvested at full heading between 4 and 15 June, and the second harvests between 24 July and 19 August. The first and second delayed harvests (DH) of the remaining half of the plots were scheduled four days later. The third cuts were harvested between 2 September and 7 October. Thus, the following total annual nutrient rates were applied in 2019-2021: Traditional – N 120 and K 50 kg ha^{-1}, YARA – N 189, P 10, K 108, Mg 19, S 27 kg ha^{-1} + B 315, Zn 45 and Se 14 g ha^{-1} $year^{-1}$.

Grass yield per plot was weighed and 1 kg of herbage was sampled. Nutritive value characteristics (NVCs) were measured in the laboratory of plant biochemistry of the Estonian University of Life Sciences. Statistical package Agrobase 20™ was used to perform a three-factorial ANOVA (cultivar, harvest time, fertilization and their interaction) and calculation of statistical significance of the differences. Year effects was not taken into account.

Results and discussion

As a sum for the years 2019-2021 the mean dry matter yield (DMY) of the cultivars totalled 23.7 Mg ha^{-1} or mean of 7.9 Mg ha^{-1} $year^{-1}$. Direct effects of cultivars, DH and YARA were all significant for DMY and for thirteen NVCs (Table 1). Cultivar had no effect on herbage Mg, DH on P, and YARA on herbage P and ash content. DH led to significant increase in herbage DMY and Ca content, but to deterioration of twelve NVCs. YARA significantly increased DMY, but reduced values of eleven NVCs relative to traditional fertilization. There was no interaction between the cultivars and fertilization regimes to the herbage DMY and NVCs. Co-effects between the cultivars and DH were determined for herbage Ca and Mg contents. Joint effects of the three experimental factors were established for eight NVCs.

There were 27, 20 and 39 days in Jõgeva in 2019-2021, respectively when the daily maximums of air temperatures exceeded 25 °C. Balasko and Smith (1971) state that 20 °C temperature is considered optimum for obtaining maximum yields in temperate grasses like timothy. Drought is a major limiting factor of crop growth and productivity among various abiotic stresses (Marvin and Jerry, 2008). This explains the reduction of mean DMYs especially on the third harvest year. In a favourable year 2020 it was 9.5, but in the dry and hot 2021 it was only 4.8 Mg ha^{-1}.

Lasting summer heat periods speeded up the plants' senescence, causing a significant deterioration of herbage NV. Increased growth temperature increases the content of all fibre fractions. In timothy, the daily rate of forage digestibility decline has been reported to be 0.06 percentage units for each degree increase in temperature (Thorvaldsson, 1992). Thorvaldsson and Andersson (1986) suggested a decline in timothy digestibility of 2 to 7 g kg^{-1} DM day^{-1}. The average respective rate during the DH in our experiment was 9 g kg^{-1} DM day^{-1}. Table 1 quantifies the deterioration rate of nearly all NVCs, caused by DH and enhanced fertilization.

From the perspective of dairy cattle feeding, ADF, ME, K and RFV of the harvested herbage was consistent with the criteria set for forage of superior grade (NRC 2001; Tamm 2017). NDF and digestibility of dry matter (DDM) comply with the criteria for acceptable quality.

RFV index >106 indicated that the DDM and DMI levels of timothy surpassed those of alfalfa at full-bloom, which is 100. Potassium contents were double the recommended values, yet remained below the allowed dietary maximum. Low content of crude protein and starch + sugars are distinctive to excessively mature grass, while low P and Mg contents to modest soil fertility.

Table 1. Total dry matter yield (DMY, Mg ha^{-1}) and means for fourteen nutritive value characteristics (g kg^{-1} DM) averaged over six timothy cultivars (cv) and years 2019-2021.[1,2]

Characteristic	Range among cultivars	Harvest		Fertilization		Significance	
		early	late	traditional	YARA	cv×DH	cv×DH×YARA
DMY	22.2-26.1***	22.8	24.6***	19.2	28.1***	ns	ns
NDF	550-571***	551	572***	551	573***	ns	*
ADF	315-329***	316	328***	313	332***	ns	*
Crude protein	79.8-90.7***	89.6	84.8***	88.9	85.5**	ns	ns
Ash	75.3-79.5***	79.6	75.4***	77.0	77.9ns	ns	ns
Crude fat	24.9-27.1***	26.9	25.4***	26.6	25.7***	ns	*
Starch+sugars	238-261***	253	242***	257	238***	ns	ns
P	1.99-2.16***	2.11	2.09ns	2.09	2.12ns	ns	ns
K	20.37-21.58***	21.52	20.83***	20.56	21.86***	ns	ns
Ca	5.03-5.38***	5.07	5.34***	5.29	5.13***	**	*
Mg	0.97-1.02ns	1.02	0.97***	1.09	0.90***	*	ns
DMI	2.14-2.24***	2.22	2.15***	2.23	2.14***	ns	*
DDM	633-644***	643	633***	645	631***	ns	*
RFV	106-113***	112	106***	113	106***	ns	*
ME	9.91-10.12***	10.10	9.92***	10.14	9.88***	ns	*

[1] Significance of direct and joint effects induced by three experimental factors. ns = not significant; * P<0.05; ** P<0.01; *** P<0.001. Cultivar × YARA interactions were absent (not presented).

[2] NDF = neutral detergent fibre; ADF = acid detergent fibre; DH = delayed harvest; YARA = fertilization; DMI = dry matter intake, % of body weight; DDM = digestibility of dry matter; RFV = relative feed value, points; ME = metabolizable energy, MJ kg^{-1} DM.

Conclusions

Postponing the timothy harvests by four days after full heading of the spring growth plus after reaching harvest maturity in the second cut increased the herbage DMY by 8%. The YARA treatment increased the DMY by 46% compared with limited NK-fertilization. In the case of enriched fertilization, the NV of timothy deteriorated due to dilution effect of valuable nutritional factors and increase of structural component (fibres) in more abundant biomass.

References

Balasko J.A. and Smith D. (1971) Influence of temperature and nitrogen fertilization on the growth and composition of switchgrass (*Panicum virgatum* L.) and timothy (*Phleum pratense* L.) at anthesis. *Agronomy Journal* 63, 853-857.

Marvin H. and Jerry C. (2008) *Agronomy Facts 24 Timothy,* University Park, PA: Penn State College of Agricultural Sciences research and extension programs.

McElroy A.R. and Christie B.R. (1986) Variation in digestibility decline with advance in maturity among timothy (*Phleum pratense* L.) genotypes. *Canadian Journal of Plant Science* 66, 323-328.

National Research Council (2001) *Nutrient Requirements of Dairy Cattle: Seventh Revised Edition.* The National Academies Press, Washington, DC, USA. https://doi.org/10.17226/9825

Tamm U. (2017) *Parema toiteväärtusega rohusööt*. ETKI, Saku, Estonia, 57 pp. (in Estonian).

Thorvaldsson G. (1992) The effects of temperature on digestibility of timothy (*Phleum pratense* L.), tested in growth chambers. *Grass and Forage Science* 47, 306-308.

Thorvaldsson G. and Andersson S. (1986) Variations in timothy dry matter yield and nutritional value as affected by harvest date, nitrogen fertilization, year and location in northern Sweden. *Acta Agriculturae Scandinavica* 36, 367-385.

Does liming grasslands increase biomass production without causing negative impacts on net greenhouse gas emissions?

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Abstract

This global review of grassland liming research assesses the impacts of liming on soil pH, biomass production and net greenhouse gas (GHG) exchanges (N_2O, CH_4 and CO_2). All studies showed that liming either reduced or had no effects on the emissions of N_2O and CH_4. Though the liming of grasslands can increase net CO_2 emissions, the impact on net GHG emissions is small due to the higher 100-year global warming potential (GWP) of N_2O and CH_4 than CO_2. Moderate liming of grassland significantly increases soil pH, grass productivity and species richness, and reduced fertilizer requirement, which justifies its wider adoption.

Keywords: grassland, lime, N_2O, CO_2, CH_4, SOC

Introduction

In grasslands, lime is applied to the soil surface and either left on the surface or incorporated into soil especially at sward renewal (Mosier *et al.*, 1998). Previous studies on agricultural (grasslands, croplands and forests) liming found that application of lime optimized plant growth by adjusting soil pH and mitigating N_2O emissions, but the impact on soil organic carbon (SOC) was inconsistent (e.g. Goulding, 2016). Moreover, the addition of lime increased CH_4 oxidiser activity and, thereby, reduced the total greenhouse gas (GHG) emissions (Kunhikrishnan *et al.*, 2016). Unlike liming of cropland, liming of grassland is often neglected, especially when the overall profit of grassland is low (Goulding, 2016). Due to scarcity of field data, it is still unknown how lime exactly influences grass productivity and nutrient use efficiency in different soil pH, botanical and agro-climatic conditions. This review aims to use the available literature globally to assess the impacts of liming grasslands on soil pH, biomass production and net GHG emissions.

Materials and methods

To cover all peer-reviewed publications (1980-2021) on the impacts of liming on soil pH, grassland biomass production and net GHG emissions (i.e. nitrous oxide (N_2O), methane (CH_4) and net CO_2 emissions), we carried out a comprehensive search on the Web of Science database. We used the keywords: grassland, lime, N_2O, CO_2, CH_4 and SOC. We only included studies carried out in the field and had a control treatment. From 12,468 papers, 55 papers (31 papers on soil pH and grass production and 24 papers on SOC and GHG emissions) with data from 72 sites were found suitable for this review. Our quantitative analysis was confined to data on soil pH and grass biomass production; papers on SOC (15 studies) and GHG emissions (N_2O (4 studies); CH_4 (2 studies) and CO_2 (5 studies)) were reviewed and summarized. All types of lime materials were converted to calcium carbonate equivalent (CCE), which is the neutralizing value of a liming material compared to pure calcium carbonate. We considered net CO_2 emissions as changes in SOC stored in the soil and net GHG emissions as the sum of net CO_2 emissions and GHG (N_2O, CH_4) emissions. Datasets referred to moist cool (MC) or moist warm (MW) climates (zonation by Smith *et al.* (2008)). We explored, analysed, and visualised the data with R version 4.0.3.

Results and discussion

Impacts of liming on soil pH and biomass production

A paired test with random effects showed that liming significantly increased soil pH (P<0.001; n=49). This effect held true also within climate zones MC (P<0.001; n=36) and MW (P<0.001; n=13), and within classes monoculture (P<0.001; n=19) and multi-species (excluding perennial ryegrass or white clover) grassland (P<0.001; n=30) (Table 1). Similar impacts of liming were reported in previous studies (e.g. Zurovec *et al.*, 2021).

A paired test with random effects showed that liming had a statistically significant positive effect on grass biomass production compared to the control treatments (P<0.001; n=69). This effect was also found within each climatic zone MC (P<0.01; n=50) and MW (P<0.001; n=19), and within monoculture (P<0.001; n=34) and multi-species grassland (P<0.01; n=35) (Table 1). Liming reduces the N fertilizer requirement to attain certain biomass production. Biomass increment due to liming was negatively correlated with initial soil pH (R^2=0.29, P=0.05, n=13), and with lime dose (R^2=0.27; P=0.1; n=11). Excess liming can decrease grass productivity due to reduced nutrient availability (e.g. phosphorus and minor nutrients) in alkaline conditions (Higgins *et al.*, 2012). Therefore, to get the maximum benefit of liming grassland, acid soils should be regularly limed but at a low rate depending on soil type and initial soil pH. The maximum recommended lime rate for grasslands in England and Wales is 7.5 t ha^{-1} for each application (AHDB, 2021).

Impacts of liming on net greenhouse gas emissions

Due to the scarcity of published data on GHG emissions from liming grasslands, this part was only reviewed and summarized. Available studies show that liming either decreased or had no significant effect on N_2O and CH_4 emissions. Increasing soil pH by liming can improve the capacity of denitrifiers to reduce N_2O to N_2 and thereby reduce N_2O emissions. Zurovec *et al.* (2021) found a decrease in soil N_2O emissions and yield-scaled N_2O emissions due to liming. Moreover, reduced N fertilizer requirement for a given yield significantly mitigates N_2O emissions from fertilized grasslands.

The Park Grass experiment (Stiehl-Braun *et al.*, 2011) showed that the interaction of soil pH with N fertilization was important for CH_4 consumption. Here, liming for more than 100 years did not restore the CH_4 oxidising capacity of the soil that had received NH_4-N fertilizer, whereas in the soil that had received NO_3-N fertilizer it was restored (Silvertown *et al.*, 2006). The authors argued that NH_4-N

Table 1. Statistical analysis of the effects of liming (t ha^{-1}) on soil pH and grass dry biomass production (t ha^{-1}) under different climatic zones (MC = moist, cool; MW = moist, warm) and number of species.

	Soil pH	Avg applied N fertilizer (kg ha^{-1})	Control (Mean±SD)	Limed (Mean±SD)	n	t-value	*P*-value
Soil pH	All data	189	4.56±1.36	5.43±0.52	49	4.69	<0.001
	MC	180	4.73±1.20	5.38±0.47	36	3.19	<0.01
	MW	231	4.09±1.68	5.57±0.62	13	4.01	<0.001
	Monoculture grass	258	3.72±1.88	5.36±0.66	19	4.24	<0.001
	Multi-species grass	134	5.09±0.30	5.48±0.40	30	11.27	<0.001
Dry biomass	All data	189	4.31±2.82	4.74±2.98	69	4.10	<0.001
	MC	180	3.91±2.21	4.21±2.17	50	11.52	<0.01
	MW	231	5.37±3.89	6.12±4.23	19	4.38	<0.001
	Monoculture grass	258	3.93±2.49	4.54±3.07	34	3.31	<0.001
	Multi-species grass	134	4.68±3.10	4.93±2.91	35	2.65	<0.01

[1] n = number of studies; SD = standard deviation; Avg = average.

fertilization had caused a shift in microbial population or resulted in persistent NH_4^+ inhibition of CH_4 oxidation to CO_2. Ammonium sulphate (an acidifier) seemed to cause an increase in CH_4 emissions at low soil pH when no lime was applied. Soil pH strongly influences CH_4 consumption through several pathways, which are still not fully understood (Stiehl-Braun *et al.*, 2011).

Our analysis showed that liming grasslands resulted in higher net CO_2 emissions because of increased CO_2 emissions and decreased SOC. This net effect can be due to: (1) greater organic matter (OM) inputs from increased growth; (2) increased OM mineralization due to more favourable soil pH for OM mineralization and OM turnover (Marcelo *et al.*, 2012); (3) direct CO_2 emission from applied lime (Raza *et al.*, 2021); and (4) enhanced aggregation of clay minerals and aggregate stability due to Ca^{2+}, thereby protecting SOC (Haynes and Naidu, 1998). However, as the GWP of CO_2 is low compared to that of N_2O and CH_4 (*c*. 273 and *c*. 28 times that of CO_2 over a 100-year period for N_2O and CH_4 respectively; IPCC, 2021), increased CO_2 efflux from liming of grassland will have negligible effect on the net GHG emissions. Overall, the increase in net CO_2 emissions due to liming will be compensated by the saving in GHG emissions due to the reduction in N_2O and CH_4 emissions.

Conclusions

According to our review, liming grasslands raises soil pH and enhances grass biomass production in acidic soils. It decreases or has no effect on N_2O and CH_4 emissions but increases net CO_2 emissions. Given the higher GWP of N_2O and CH_4, the result of liming will be a negligible effect on net GHG emissions. Therefore, it makes sense to lime productive, acidic grasslands to increase nutrient-use efficiency within livestock grazing systems.

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References

Higgins S., Morrison S. and Watson C.J. (2012) Effect of annual applications of pelletized dolomitic lime on soil chemical properties and grass productivity. *Soil Use and Management* 28, 62-69.

IPCC (2021) In: Climate Change 2021: The Physical Science Basis. Contribution of Working Group I to the Sixth Assessment Report of the IPCC [Masson-Delmotte V.P. Zhai *et al.* eds.]. Cambridge CUP.

Kunhikrishnan A. and Thangarajan R *et al.* (2016) Functional relationships of soil acidification liming and greenhouse gas flux. *Advances in Agronomy* 139, 1-71.

Marcelo A.V., Corá J.E. and La Scala-Junior N. (2012) Influence of liming on residual soil respiration and chemical properties in a tropical no-tillage system. *Revista Brasileira de Ciência do Solo* 36 (1), 45-50.

Mosier A.R., Delgado J.A. and Keller M. (1998) Methane and nitrous oxide fluxes in an acid oxisol in western Puerto Rico: effects of tillage liming and fertilization. *Soil Biology & Biochemistry* 30, 2087-2098.

Raza S., Zamanian K., Ullah S., Kuzyakov Y., Virto I. and Zhou J. (2021) Inorganic carbon losses by soil acidification jeopardize global efforts on carbon sequestration and climate change mitigation *Journal of Cleaner Production 315*, 128036.

Silvertown J., Poulton P., Johnston E. *et al.* (2006) The park grass experiment 1856-2006: its contribution to ecology. *Journal of Ecology* 94, 801-814.

Smith P., Martino D. and Cai Z. (2008) Greenhouse gas mitigation in agriculture. *Philosophical Transaction of the Royal Society B: Biological Sciences* 363, 789-813.

Stiehl-Braun P.A., Powlson D.S., Poulton P.R. and Niklaus P.A. (2011) Effects of N fertilizers and liming on the micro-scale distribution of soil methane assimilation in the long-term Park Grass experiment at Rothamsted. *Soil Biology and Biochemistry* 43 (5), 1034-1041.

Zurovec O., Wall D.P. *et al.* (2021) Increasing soil pH reduces fertilizer derived N_2O emissions in intensively managed temperate grassland. *Agriculture Ecosystems and Environment* 311, 107319.

Grazing farms differentiation through the expression of microARNs and AI algorithm

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Abstract

Milk production based on grazing is being promoted over cattle housed indoors, because of the advantages regarding animal welfare, milk quality and the environment. Cows' milk is rich in miRNAs, molecules that regulate gene expression in eukaryotes. Their profiles may vary depending on environmental factors such as farm management and feeding. We hypothesize that miRNA can be used as a certification tool for dairy farms whose milk production is based on grazing. The objective is to apply an artificial intelligence algorithm to the results of miRNA expression in milk to evaluate the possibility of designing a fast and cheap traceability tool that can differentiate the milk produced in a grazing-based system from milk produced in indoor systems. Cells and fat fractions were isolated from seventy-three milk tank samples from 'No-Grazing' (n=47) vs 'Grazing' (n=26) farms. MiRNA expression was analysed in the cells and the fat fractions of the milk samples. Following miRNAs expression analysis, decision trees were built for their expression results using the C4.5 machine learning algorithm. The algorithm was not able to correctly classify each sample in its group, nor was it able to identify relevant miRNAs. We assume that the enormous internal variability (diets, botanical composition of the pastures, and grazing duration, etc.) in commercial grazing farms could be the cause of the difficulty in machine learning of how to classify milk from grazing farms.

Keywords: dairy cow, grazing, biomarkers, microRNA

Introduction

Collective awareness about the consumption of sustainable products implies that grazing is being promoted over cattle housed indoors, so mechanisms are required for the authentication of the origin of milk. MicroRNA (miRNA) are molecules that regulate gene expression in eukaryotes (He and Hannon, 2004). Their profiles vary depending on environmental factors, such as farm management and feeding (Li *et al.*, 2015; Muroya *et al.*, 2015, 2016; Wang *et al.*, 2016). Based on these results, miRNA could be used as biomarkers of milk's origin. The main objective of this work is to apply an artificial intelligence (IA) algorithm to the results of miRNA expression in milk, in order to evaluate the possibility of designing a fast and cheap traceability tool that can differentiate the milk produced in a grazing-based system from the milk produced in indoor systems.

Materials and methods

A representative set of farms (n=73) of Asturian milk production systems was sampled from the tank. In every milk sample, cells and fat fractions were separated (Li *et al.*, 2016), and the total RNA was extracted from 146 samples and then used for cDNA synthesis. The expression, in cells, of bta-miR-181a, bta-miR-197, bta-miR-2284y, bta-miR-2285e, bta-miR-342, bta-miR-3432a, bta-miR-574, bta-miR-28, bta-miR-345-3p (Abou el qassim, 2017), bta-miR-148, bta-miR-155, bta-miR-21-5p, bta-miR-451-5p (Li *et al.*, 2015; Muroya *et al.*, 2015, 2016; Wang *et al.*, 2016) and the expression in fat of bta-miR-215, bta-miR-369-5p, bta-miR-6520, bta-miR-7863, bta-miR-99a-3p, bta-miR-532, bta-miR-27b, bta-miR-151-3p (Abou el qassim, 2017), bta-miR-148, bta-miR-451-5p relative to the levels of determined stable internal miRNAs, were determined by the quantitative real-time PCR (RT-qPCR). The samples were grouped according to grazing practice: 'Non-Grazing' farms (n=47) and 'Grazing farms' (n=26). Decision trees were built for miRNAs expression results using the C4.5 machine learning algorithm (Quinlan, 1993). The algorithm allows, on the one hand, to classify each sample in a 'Grazing' or 'Non-Grazing' category then to calculate its ability to learn to classify the samples in their categories, and on the other hand to identify the most informative miRNAs.

Results and discussion

In a leave-one-out experiment, the algorithm C4.5 correctly classified 41 samples of 'Non-Grazing' group (n=47); however, only 8 'Grazing' samples were correctly assigned in their group (n=26), as shown in the confusion matrix (Table 1). The number of hits is about 67.12%. Nevertheless, the result is not sufficient since the majority class accounts for 64.38% of the 73 examples.

The best decision tree obtained (Figure 1) points out bta-miR-215, bta-miR-6520 and bta-miR-99a from fat fraction and bta-miR-2284y, bta-miR-28 and bta-miR-148 from cell fraction, as relevant attributes. The up regulated level of bta-miR-215 was related in a previous study (Abou el qassim *et al.*, 2021) to maize silage consumed, and in general grazing farms do not usually include corn silage in the diet. The decision tree shows that the algorithm selected first miRNA from fat and then from cells. In our previous study about maize silage as a class, the algorithm did not even select miRNA from cells, which might be expected since the cells fraction is more heterogeneous, and it may also not reflect the true metabolic state of mammary gland cells because they are usually dead cells (Krappmann *et al.*, 2012).

Table 1. Confusion matrix of the samples assignment the in the studied classes, by the algorithm C4.5.

	Classified as grazing	Classified as non-grazing
Grazing samples	8	18
Non-grazing samples	6	41

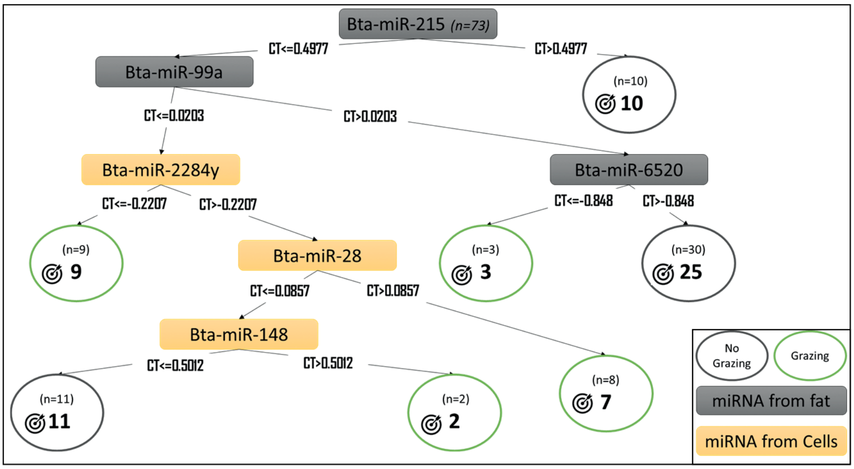

Figure 1. Decision tree with the relevant miRNAs, cut-off thresholds, and the successful assignments. Ct: RT-qPCR cycle threshold reflecting the relative expression of miRNAs.

Conclusions

Learning to classify the grazing farms seems complicated, due to the enormous internal variability of diets, the botanical composition variability of pastures, and the grazing duration, etc. However, when considering other criteria with less variability, such as the case of presence/absence of maize in the diet (data not shown, Abou el qassim *et al.*, 2021), the algorithm learns more.

In the near future we want to explore the study of the botanical composition of pasture in grazing farms and its variability, and the effect of exercise on the differential expression of milk miRNA.

References

Abou el qassim L., Alonso J., Royo L.J. and Díez J (2021) Determinación del origen de la leche cruda de vaca mediante la expresión de microARN y algoritmos de IA. *AIDA XIX Jornadas sobre Producción Animal*, p. 87.

Abou el qassim L. (2017) *Variations of microRNA profiles in raw cow's milk according to the feeding system.* (Master's Thesis) International Centre for Advanced Mediterranean Agronomic Studies, Zaragoza, Spain, 129 pp.

He L. and Hannon G.J. (2004) MicroRNAs: small RNAs with a big role in gene regulation. *Nature Reviews Genetics* 5, 522-531.

Krappmann K., Weikard R. and Kühn C. (2012) Evaluation of a replacement method for mammary gland biopsies by comparing gene expression in udder tissue and mammary epithelial cells isolated from milk. *Research in Veterinary Science* 93, 970-974.

Li R., Beaudoin F., Ammah A.A., Bissonnette N., Benchaar C., Zhao X., Lei C. and Ibeagha-Awemu E.M. (2015) Deep sequencing shows microRNA involvement in bovine mammary gland adaptation to diets supplemented with linseed oil or safflower oil. *BMC Genomics* 16, 1-16.

Li R., Dudemaine P.L., Zhao X., Lei C. and Ibeagha-Awemu E.M. (2016) Comparative analysis of the miRNome of bovine milk fat, whey and cells. *PloS One* 11, e0154129.

Muroya S., Ogasawara H., and Hojito M. (2015) Grazing affects exosomal circulating microRNAs in cattle. *PLoS One* 10, e0136475.

Muroya S., Shibata M., Hayashi M., Oe M. and Ojima K. (2016) Differences in circulating microRNAs between grazing and grain-fed wagyu cattle are associated with altered expression of intramuscular microRNA, the potential target PTEN, and lipogenic genes. *PLoS One* 11, e0162496.

Quinlan J.R. (1993). *C4.5: programs for machine learning*. Morgan Kaufmann Publishers Inc., San Francisco, CA, USA.

Valenti B., Martin B., Andueza D., Leroux C., Labonne C., Lahalle F., Larroque H., Brunschwig P., Lecomte C., Brochard M. and Ferlay A. (2013) Infrared spectroscopic methods for the discrimination of cows' milk according to the feeding system, cow breed and altitude of the dairy farm. *International Dairy Journal* 32, 26-32.

Wang D., Liang G., Wang B., Sun H., Liu J., and Guan L.L. (2016) Systematic microRNAome profiling reveals the roles of microRNAs in milk protein metabolism and quality: insights on low-quality forage utilization. *Scientific Reports* 6, 21194.

Effectiveness of measures on dairy farms to improve nitrogen balance and nitrogen use efficiency

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Abstract

The aim of this study is to evaluate the effects of measures taken on dairy farms to reduce nitrogen (N) losses within the framework of a national initiative for improving agricultural N use efficiency (NUE). Farm gate N balances were assessed to determine the N surplus and the NUE of 11 dairy farms (6 specialized with >65% grassland of total agricultural land and 5 mixed with >40% arable land of total agricultural land). The farmgate N balances were assessed in 2019 and 2020 and compared to the baseline (2015 to 2017) in order to evaluate the effects of the measures taken on the farms. Individual measures were taken on each farm to reduce the N surplus and to increase the NUE. On all farms N input via concentrates was reduced and manure composition was analysed to improve fertilization. Additional measures consisted of the optimization of animal management, manure application and crop rotation. The mean N surplus was reduced from 146 to 106 kg N ha^{-1} and NUE increased from 44 to 53% on average in the years in which measures were taken. These results show an improvement regarding N surplus when measures were taken and indicate a general potential for improvement in N use on dairy farms.

Keywords: nitrogen, dairy, farmgate balance, grassland, measures

Introduction

The farmgate N balance is considered a useful tool to identify the sustainability of nutrient use and the potential for N losses into the environment (Oenema *et al.*, 2003). Nitrogen inputs from concentrates and mineral fertilizer have been identified as important N inputs (apart from biological N_2 fixation and atmospheric N deposition) (Akert *et al.*, 2020). Different technical measures have been proposed to farmers to reduce N surplus and to improve N use efficiency (NUE) at the field and farm level of arable and livestock farms (Hutchings *et al.,* 2020). The aim of this study is to analyse the effectiveness of measures introduced to reduce N losses and improve the NUE on Swiss dairy farms.

Materials and methods

Farmgate N balances were calculated for 11 dairy farms based on data obtained from the national nutrient budgeting tool 'Suisse-Bilanz' and from farm records. Six dairy farms were specialized grassland farms (SF) with >65% grassland of total agricultural land and five farms were mixed arable farms (MF) with >40% arable land of total agricultural land (Table 1). Nitrogen inputs such as animals, fertilizers, manure, feed, seed and plant material, biological N_2 fixation and atmospheric N deposition were accounted for. Biological N_2 fixation by legumes was calculated assuming legume shares of 0.1, 0.15 and 0.2 of the annual dry matter yield for extensively managed meadows, pastures, and intensively managed leys respectively, with an average N input of 30 g N kg^{-1} of dry matter legume yield according to Boller *et al.* (2003). Atmospheric N deposition was calculated based on the amount of deposited N, as published by the Federal Office for the Environment for the corresponding region of each farm. Animals, milk, eggs, manure, plant products and forage were considered as farm N outputs. Nitrogen surplus per hectare of farmland for each farm was calculated annually as N inputs minus N outputs. Nitrogen use efficiency was referred to as the relation of N outputs to N inputs per hectare of farmland. For each farm, individual measures were defined to reduce N surplus and increase NUE in 2019 and 2020 compared to the baseline of 2015-2017. Measures taken on all farms consisted of reducing or abandoning the N supply from off-farm feed and analysing chemical manure composition before application on the fields. Measures taken on individual farms included further planning of mineral fertilizer and manure management on the farmland, optimizing crop rotation, using nitrification or urease inhibitors for manure, optimizing livestock husbandry, or management and low emission manure application techniques. Statistical analysis was performed on the mean for the baseline and period with measures implemented. The baseline was compared to the period with measures on each farm applying a mixed model with period as fixed factor and the farm as random factor accounting for repeated measurements. The farm type (SF or MF) was excluded from the model as the factor was not statistically significant. When model assumptions for analysis of variance were not met (no normal distribution of residuals), the analysis of variance was performed on the square root transformed parameter. Comparisons between means for the two periods were performed with Tukey's method at a significance level of $P<0.05$.

Results and discussion

The farm N surplus was reduced by 28% in 2019/20 compared to the baseline (Table 2). According to the ideal pathways of Quemada *et al.* (2020) to improve the N use, the farms in this study have improved by extensification and sustainable intensification. Total N inputs were reduced in the period with measures taken to reduce N surplus on the farms. Significant reduction of mineral fertilizer input and roughage input were made compared to the baseline. Interestingly, N outputs did not show a significant reduction compared with the level of the baseline, even though N inputs were reduced. The large range of N inputs indicate potential for optimization on individual farms. When measures were applied on the farms, NUE was increased, on average for all farms, from 43.5 to 53.3%, and the minimum NUE increased from 31.0 to 37.7%. Even though NUE on arable farms have been found to exceed NUE on dairy farms (Quemada *et al.*, 2020), no difference was found on the farms investigated. This was probably due to the similar N output via milk (56.1 and 65.1 kg N ha^{-1} a^{-1} for SF and MF in 2019/20 respectively, data not shown) and the relatively small difference of arable production on MF (50 vs 14% open arable land of total farmland of MF and SF respectively).

Conclusions

With the application of specific measures, the N surplus could be reduced and the NUE increased compared to the baseline on the individual farms. Reducing N input via mineral fertilizer seems to be an effective way for reducing N surplus and increasing NUE on dairy farms. Short-term effects such as reduced N output due to substantially reduced crop yields could not be observed. To account for the interannual variance three years of baseline have been included and two years of measures taken. Nevertheless, additional years with measures taken will be included to account for short-term effects and also for long-term effects such as mineral N stock changes in the soil.

Table 1. Farm characteristics of all farms and grouped according to farm type for the specialized grassland farms (SF) and the mixed arable farms (MF) presented as mean of the baseline (2015 to 2017) and the years 2019 and 2020 with individual measures taken.

		n	Farmland area (ha)	Arable crops (ha)	Total livestock units (LU)	Cattle (LU)	Stocking rate (no of LU ha^{-1})	Concentrate (kg cow^{-1} a^{-1})	Milk yield[1] (kg cow^{-1} a^{-1})
Baseline	all farms	11	42.9	11.3	66.2	58.6	1.60	1,244	7,937
	SF	6	44.4	5.1	71.2	61.9	1.64	1,153	7,623
	MF	5	41.0	19.2	59.8	54.5	1.55	1,359	8,497
2019/20	all farms	11	43.8	11.7	69.1	60.0	1.65	1,099	8,022
	SF	6	46.3	5.0	73.9	61.2	1.64	920	7,892
	MF	5	40.9	19.7	63.4	58.5	1.66	1,314	8,238

[1] All farms n=8; SF n=5; MF n=3.

Table 2. Annual nitrogen (N) inputs and outputs (arithmetic mean, minimum and maximum in kg N ha^{-1} a^{-1}, share of total in %), N surplus and N use efficiency of 11 dairy farms calculated for the baseline years and for 2019/20 where measures on farms were taken to reduce N surplus and increase N use efficiency.

	Baseline (n=11) 2015/16/17			2019/20 (n=11)			2019/20 vs Baseline	*P*-value
	mean	min	max	mean	min	max		
Total farm N inputs	258	133.9	450.0	229	96.6	361.5	-29.0	*
Mineral fertilizer	47.1	5.4	113.7	30.8	5.4	104.8	-16.3	**
Animal manure	17.2	0	54.9	19.6	0	62.2	2.4	NS
Livestock	15.7	2	47.2	15.8	1.8	55.6	0.1	NS
Plant material	0.5	0	1.3	0.5	0	1.3	0.0	NS
Feed pigs/chicken	15.3	0	110.8	12.7	0	101.5	-2.6	NS
Feed rearing	1.5	0	5.0	1.4	0	4.5	-0.1	NS
Roughage	24.5	4.2	65.8	21.2	0	67.3	-3.3	*
Concentrates dairy cows	78.7	5.3	236.6	67.1	0	158.8	-11.6	NS
Biological N fixation	32.7	21.6	58.2	34.8	19.2	63.4	2.1	NS
Atmospheric deposition	25.0	19.2	29.9	25.2	19.3	29.8	0.2	NS
Total farm N outputs	111.7	45.8	172.9	123.5	48	210.7	11.8	NS
Milk protein	55.5	24.7	115.1	59.3	28.4	137.4	3.8	NS
Livestock	27.7	8.6	91.6	27.4	7.7	88.3	-0.3	NS
Egg protein	0	0	0.2	0	0	0.4	0.0	NS
Plant product	16.4	0	56.6	16.5	0	74.9	0.1	NS
Roughage	5.7	0	26.3	6.9	0	35.1	1.2	NS
Animal manure	6.8	0	23.6	13.3	0	40.7	6.5	NS
Farm N balance	146.3	84.5	302.5	105.5	43.7	208.3	-40.8	*
Farm N use efficiency	43.5	31.0	63.4	53.3	37.7	72.5	9.8	***

[1] *** $P<0.001$; ** $P<0.01$; * $P<0.05$; NS = not significant.

[2] Parameter was square root or log10 transformed for statistical analysis.

References

Akert F.S., Dorn K., Frey H., Hofstetter P., Berard J., Kreuzer M. and Reidy B. (2020) Farm-gate nutrient balances of grassland-based milk production systems with full- or part-time grazing and fresh herbage indoor feeding at variable concentrate levels. *Nutrient Cycling in Agroecosystems* 117(3), 383-400.

Boller B., Lüscher A. and Zanetti S. (2003) Schätzung der biologischen Stickstoff-Fixierung in Klee-Gras-Beständen. *Schriftenreihe der FAL* 45, 47-54.

Hutchings N.J., Sørensen P., Cordovil C.M., Leip A. and Amon B. (2020) Measures to increase the nitrogen use efficiency of European agricultural production. *Global Food Security* 26, 100381.

Oenema O., Kros H. and De Vries W. (2003) Approaches and uncertainties in nutrient budgets: Implications for nutrient management and environmental policies. *European Journal of Agronomy* 20(1-2), 3-16.

Quemada M., Lassaletta L., Jensen L.S., Godinot O., Brentrup F., Buckley C., Foray S., Hvid S.K., Oenema J., Richards K.G. and Oenema O. (2020) Exploring nitrogen indicators of farm performance among farm types across several European case studies. *Agricultural Systems* 177, 102689.

Drone-based multispectral imagery is effective for determining forage availability in arid savannas

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Abstract

Dry savannahs are highly sensitive to climate change and are under intense anthropogenic pressure. Therefore, methods for assessing their status should be easy and repeatable. Monitoring through satellite data and field measurements are limited in accurately assessing the spatiotemporal dynamics of ecosystems. Fortunately, emerging technologies like drones and associated miniaturized sensors allow to transcend these limitations. Yet, extensive calibration with field data in arid savannah systems is still limited. We fill this gap by correlating drone and field estimated forage biomass in an arid savannah. Our results revealed significant relationships ($P<0.001$) between dry biomass and vegetation indices (NDVI, OSAVI, GDVI, TNDVI), with OSAVI adapted for arid systems showing the best fit ($F=178.1$, $DF=34$, $R^2=0.84$). Integrating UAV-based prediction models of ecosystem parameters that are highly responsive to rangeland condition such as forage biomass in monitoring could greatly assist in climate-adapted management. This will prevent further land degradation and associated threats to biodiversity and livelihoods.

Keywords: arid savannah, forage provision, ground-truthing, multispectral sensor

Introduction

Rangeland condition in drylands continues to deteriorate due to overgrazing compounded by limited moisture availability and changing climatic conditions (Millennium Ecosystem Assessment, 2005). This requires that monitoring methods are easy to apply and provide reliable and repeatable results at various spatial and temporal scales to improve management and answer ecological questions. However, the usual methods applied at global and local levels through satellite data and field observations, respectively, provide only limited information for detecting degradation in its early stages (Al-Bukhari *et al.*, 2018).

Satellite-based indicators of degradation, such as vegetation indices and land cover maps, provide automated and repeatable data at large scales. However, arid rangelands present unique challenges due to irregular growing seasons, complex mosaics of woody and herbaceous plants, high soil background reflectance, and large spatial heterogeneity (Wu, 2014). On the other hand, although field-based observations provide fine-scale information, they are labour-intensive, intrusive, require field specialists and extrapolations are based on a limited set of samples (Al-bukhari *et al.*, 2018; Theau *et al.*, 2021).

Recently, unmanned aerial vehicles (UAVs) with associated sensors have become popular amongst the ecological research community that is increasingly integrating them for monitoring vegetation and other ecosystem components (Assmann *et al.*, 2018; Gillan *et al.*, 2020; Laliberte *et al.*, 2010). Yet, their application is still limited in dynamic and heterogeneous ecosystems like savannahs. Thus, extensive calibration with field data is necessary if they are to be integrated for long-term monitoring (Gillan *et al.*, 2020). Our study aims to validate drone-based assessment of rangeland condition in a Namibian arid savanna. Here, the most responsive indicator of rangeland condition, forage biomass, is observed directly in the field and correlated to drone-based vegetation indices (VIs) adapted for dryland systems.

Materials and methods

We conducted the research in a semi-arid savannah in central Namibia, a typical representative of arid rangelands used for cattle production. A MicaSense RedEdge MX sensor with 5 spectral bands mounted on a DJI Matrice 200 v2 drone was used to acquire multispectral imagery over four transects. The imagery was acquired between February and March 2021 using the Pix4DCapture flight planning application. Radiometrically calibrated reflectance maps were generated in Pix4DMapper Pro (Pix4D, Switzerland, V3.3). These were further analysed in ENVI to produce four VIs to estimate forage provision. We calculated the commonly used Normalized Difference Vegetation Index (NDVI), as well as three other VIs developed specifically for arid systems (Baghi and Oldeland, 2019; Wu, 2014), namely the Optimized Soil Adjusted Vegetation Index (OSAVI), the Generalized Difference Vegetation Index (GDVI), and the Transformed Normalized Difference Vegetation Index (TNDVI). After the drone flights we harvested forage biomass from nine plots along each of the transects. The samples were oven dried at 65 °C for 48 hours to obtain the dry weight. To ensure that forage biomass estimated by the two methods was directly comparable, aerial targets that are visible in the imagery were placed in the corners of the plots. We determined the correlation coefficients between the field and drone estimates of forage biomass in R v3.4.3 (RStudio Team, 2021) and compared the four VIs.

Results and discussion

Vegetation characteristics such as above-ground biomass can objectively and flexibly be collected by sensors mounted on drones as inputs for regular monitoring of rangelands (Gillan *et al.*, 2020; Laliberte *et al.*, 2010). Developing robust drone-based models requires field data to calibrate them, which is still limited in dynamic systems. Our validation results in an arid savanna yielded significant positive relationships (P<0.001, n=36) between the VIs and dry forage biomass, with OSAVI being the best fit (F=178.1, df=36, R^2=0.84) (Figure 1). The latter illustrates the significance of testing different VIs before developing prediction models to provide optimal results (Baghi and Oldeland 2019; Theau *et al.* 2021). For example, the commonly used NDVI which is often used as a default for biomass estimation,

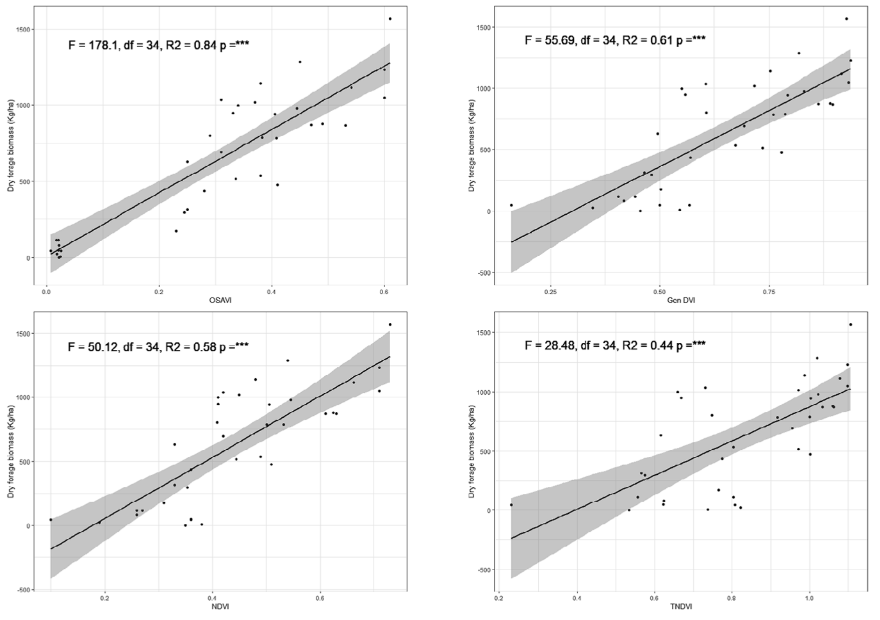

Figure 1. Linear regressions between field observed dry forage biomass (kg ha^{-1}) and four drone-based vegetation indices (n=36). The line shows the model fit, while the grey denotes 95% confidence bands and the points represent samples.

even in drylands despite the well documented limitations associated with it (Baghi and Oldeland, 2019; Wu, 2014), showed a weaker relationship than those indices adapted for arid systems. Here OSAVI is shown to be sensitive to estimating forage availability in areas with very low biomass, which is important for identifying overgrazed areas.

The results confirm the applicability of UAV technology to evaluate primary production even in a highly heterogenous system, which agrees with studies conducted in other ecosystems (Laliberte *et al.*, 2010; Theau *et al.*, 2021).

Conclusions

We used field data to test and compare how four UAV-based VIs perform in estimating forage availability in an arid savannah. We found significant agreements, with OSAVI being the best fit. Our results corroborate the applicability of this efficient and flexible technology to assess dynamic ecosystems such as arid savannas. Integrated as a monitoring tool it could aid climate-adapted rangeland management which is pertinent in a changing world.

References

Al-Bukhari A., Hallet S., and Brewer T. (2018) A review of potential methods for monitoring rangeland degradation in Libya. *Pastoralism: Research, Policy and Practice* 8(13), 1-14.

Assmann J.J., Kerby J.T., Cunliffe A.M. and Myers-Smith I.H. (2018) Vegetation monitoring using multispectral sensors – best practices learned from high altitudes. *Journal of Unmanned Aerial Vehicle Systems* 7, 54-75.

Baghi G.N. and Oldeland J. (2019) Do soil-adjusted or standard vegetation indices better predict above ground biomass of semi-arid, saline rangelands in North-East Iran? *International Journal of Remote Sensing* 40(22), 8223-8235.

Gillan J.K., Karl J.W. and Van Leeuwen, W.J.D. (2020) Integrating drone imagery with existing rangeland monitoring programs. *Environmental Monitoring and Assessment* 192(5), 269.

Laliberte A.S., Herrick J.E., Rango A. and Winters C. (2010) Acquisition, orthorectification, and object-based classification of unmanned aerial vehicle (UAV) imagery for rangeland monitoring. *Photogrammetric Engineering and Remote Sensing* 76(6), 661-672.

Millennium Ecosystem Assessment (2005) *Ecosystems and human well-being*. Island Press, Washington, DC, USA.

Pix4D: A unique photogrammetry software suite for drone mapping. Available at: https://www.pix4d.com.

RStudio Team (2021). RStudio: Integrated Development Environment for R. RStudio, PBC, Boston, MA, USA.

Theau J., Lauzier-Hudon E., Aube L. and Devillers N. (2021) Estimation of forage biomass and vegetation cover in grasslands using UAV imagery. *PLoS ONE* 16(1), e0245784

Wu W. (2014) The generalized difference vegetation index (GDVI) for dryland characterization. *Remote Sensing* 6(2), 1211-1233.

Test of a sensor to estimate grazing and ruminating time in dairy cow behaviour at pasture

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Abstract

Use of automated tools on farms is increasing worldwide and there are diverse applications available including optimization of grazing through monitoring rumination and ingestion times. The objective of this study was to evaluate the accuracy of the sensor developed by Medria to estimate grazing (G) and rumination (Ru) on pasture. This sensor includes a tri-axial accelerometer and provides data every 5 minutes. The trial lasted 12 days on 12 grazing cows in the CTA (Belgium), representing 380 h of observations. The data registered by the Medria device (M) were compared with visual observation (VO). The datasets were compared using Fleiss-κ estimating the concordance of 5-min observations and linear regression analysis was used to estimate the Pearson correlation coefficients (rp) and relative prediction error (RPE). Moderate for G (κ=0.502) and poor agreement for Ru (κ=0.175) were observed. Linear relationship between VO and M was highlighted with rp: 0.793; P<0.001 for G and rp=0.32; P<0.05 for Ru. The relative error prediction was 0.16 and 0.44 for G and Ru respectively. With regards to these results, reliable data about grazing are provided on a daily basis. The reliability of rumination data was poor.

Keywords: smart farming, sensors, rumination, grazing, accelerometer

Introduction

Increasing size of farms and lack of manpower have contributed to the development of smart farming which allows automation of several tasks and facilitates animal management by providing information on, e.g. the behaviour of the animals, and this can even generate alerts if the recorded behaviour does not correspond to that registered on previous days. Evaluation of grazing and rumination times of grazing dairy cows is of key interest for managing grazing. Lack of information on intake of grazed grass has been highlighted as a reason for stopping grazing (Lessire *et al.*, 2019). Sensors using different technologies are commercialized: nosebands recording electrical resistance, microphones recording ingestion and rumination and bi- or tri-axial accelerometers (Ambriz-Vilchis *et al.*, 2015; Delagarde and Lamberton, 2015; Pereira *et al.*, 2018; Werner *et al.*, 2018). The objective of this study is to estimate the accuracy of a tri-axial accelerometer commercialized by the French company Medria (Saint-Lo, France) for estimation of grazing and rumination times in grazing cows. This accelerometer is mounted on a collar that determines the major activity on a time interval of 5 minutes. Eight activities are discriminated. The data provided by the sensors are compared with visual observations that are considered as reference standard.

Materials and methods

The study was carried out at the Centre of Agronomic Technologies (50.507°N; 5.31°E) at Strée (Modave) in Belgium. The trial was conducted from 14 to 21 June and from 6 to 13 July 2021, for a total of 380 h of observations. Twelve Prim'Holstein cows (milk yield – MY: 25.3±6.1 kg; lactation number – LN: 2.9±1.9, including 4 primiparous; days in milk – DIM: 173±39 days) selected out of 2 groups grazing permanent grassland paddocks. The paddocks of 1.96 and 2.02 ha respectively were managed by strip grazing, granting access to fresh grass. A complementation of 4 kg concentrate (crude protein 16%; 870 VEM) was supplied at barn.

Sensors were mounted on collars adjusted on the neck of the cows. Four cows were observed per day during 8 hours divided into 4 observation periods (6:10-8:10, 9:00-12:00, 13:00-15:00 and 16:00-17:00). The most predominant behaviour on a 5 min period was noted. The Medria device continuously records behaviours, but only indicates a 'single majority activity' over a 5 min interval. Visual observations were collected over the entire trial period by the same trained operator. A trial period involving several observers validated the operator's recognition of the behaviour according to the definitions of Medria: grazing is 'a low head position, close to the ground and necessarily efficient frontal and slightly lateral movements with straight segments' and 'the animal may ingest a little with its head up'. Thus the behaviour recorded as grazing includes searching, prehension and mastication. For rumination, the Medria device defines it as 'a metronome movement, from a high position of the head'. The reference method is based on the definition: 'with circular movements of the head and jaw, the cow continuously regurgitates and swallows a bolus'. Every behaviour lasting more than 30 s was recorded; we then choose to select the most predominant activity, i.e. the one with the longest cumulative duration over this period. Other activities such as heat-related behaviour, resting, ruminating, eating or over-activity are also reported by Medria, so that 8 behaviours were discriminated, i.e. standing (S), lying (L), lying rumination (LR), standing rumination (SR), ingestion at barn (BI) or on pastures (G), over-activity (OA – which compares current activity to that recorded on previous days to indicate the likelihood of heat) and other (O). A total of 380 h of observation was reached. In this study we will only discuss the results obtained for G and Ru summing the data of LR and SR.

Statistical analysis using SAS software and R includes the comparison of each collected data (visual observations – VO vs sensor observations – M) on a 5 min period using the proc freq procedure to determine the freq of agreement between both methods, then the concordance coefficient of Fleiss (κ) was calculated using R (package irr). In a second step, duration of each behaviour was calculated considering it lasted during 5 min so that the duration over the daily observation time (8 h) was estimated for each of them for VO and M. Linear regression (proc reg and proc corr; SAS) was used to assess the agreement between the two methods. The mean root prediction error and relative prediction error were calculated following the method described by Delagarde *et al.* (2015). Only results regarding G and rumination (Ru) are shown.

Results

Figure 1 shows the frequencies (%) of the different behaviours recorded by M. The M agreed with VO for G in 64.0%, 30.4% for LR and 12.1% for SR. In 28.4% of the behaviours identified by VO as LR, M indicated L while SR was frequently confused with LR (36%). Thus, we decided to merge LR and

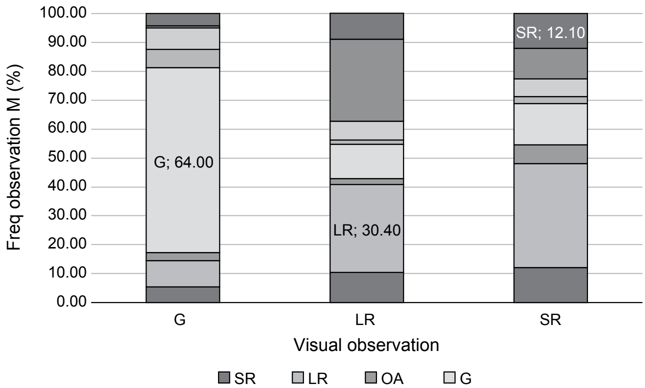

Figure 1. Comparison of the occurrence of the different behaviours recorded by M with the behaviour G, LR and SR identified by VO. Abbreviations: G = grazing. LR and SR = lying and standing rumination.

SR in Ru. The κ-coefficient estimated at 0.502 for G and 0.175 for Ru indicated moderate and poor agreement for G and Ru, respectively.

The total duration of G and Ru over a daily 8 h observation period were 192±71 min (range: 70-395 min), 112±40 min (range: 35-205 min) for VO vs 168±62 min (range: 55-335 min), 141±52 (range: 40-270 min) for M. The duration of G and Ru estimated by M were linearly correlated with VO. The Pearson coefficient was 0.793; P<0.001 for G and 0.32; P<0.05 for Ru. The results are summarized in Table 1.

Table 1. Results of the linear regression linking M and VO observations.[1]

	n obs	mean VO (min)	mean Medria (min)	intercept	slope	SE	R^2	MSPE	RPE
Grazing	47	191.7	167.3	35.11	0.69	0.08	0.63	29.9	0.16
Rumination	47	112.3	140.6	87.75	0.47	0.18	0.13	49.5	0.44

[1] VO = visual observation; SE = standard error of the slope; MSPE = mean square prediction error; RPE = relative prediction error.

Discussion and conclusion

The advantages of Medria sensors are numerous: they are cheap, robust, with long battery life, and they provide other valuable information, e.g. about the probability of heat. Alerts about unusual behaviour duration are sent by different media, so that they are appreciated by the farmers. In consideration with the acceptable value of κ-coefficient and the strength of the correlation coefficient between VO and M observations, grazing time could be considered as accurately measured by the Medria sensors. Lower duration values than observed are measured but the RPE remained low (12%). Whether based on the κ-coeficient or the RPE estimate, rumination behaviour was not accurately discriminated and duration time provided by Medria sensors was not reliable. We conclude from the conditions of our study that data provided by Medria sensor could be used to assess the G behaviour but not rumination.

References

Ambriz-Vilchis V., Jessop N.S., Fawcett R.H., Shaw D.J. and Macrae A.I. (2015) Comparison of rumination activity measured using rumination collars against direct visual observations and analysis of video recordings of dairy cows in commercial farm environments. *Journal of Dairy Science* 98(3), 1750-1758. https://doi.org/10.3168/jds.2014-8565

Delagarde R. and Lamberton P. (2015) Daily grazing time of dairy cows is recorded accurately using the Lifecorder Plus device. *Applied Animal Behaviour Science* 165, 25-32. https://doi.org/10.1016/j.applanim.2015.01.014

Lessire F., Jacquet S., Veselko D., Piraux E. and Dufrasne I. (2019) Evolution of grazing practices in Belgian dairy farms : results of two surveys. *Sustainability* 11(3397), 1-15.

Pereira G.M., Heins B.J. and Endres M.I. (2018) Technical note: Validation of an ear-tag accelerometer sensor to determine rumination, eating, and activity behaviors of grazing dairy cattle. *Journal of Dairy Science* 101(3), 2492-2495. https://doi.org/10.3168/jds.2016-12534

Werner J., Leso L., Umstatter C., Niederhauser J., Kennedy E., Geoghegan A., Shalloo L., Schick M. and O'Brien, B. (2018) Evaluation of the RumiWatchSystem for measuring grazing behaviour of cows. *Journal of Neuroscience Methods* 300, 138-146. https://doi.org/10.1016/j.jneumeth.2017.08.022

Multispecies swards improve animal growth and performance at slaughter in a dairy calf to beef production system

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Abstract

Pasture-based ruminant production systems are faced with numerous challenges, not least the requirement to maintain output while minimizing environmental impact. Previous studies have demonstrated the potential of multispecies swards (MSS) to enhance animal performance, from reduced fertilizer nitrogen (N) inputs in sheep production systems. The aim of this study was to assess the effect of sward type on animal growth and performance at slaughter in a dairy calf to beef system (2.5 LU ha^{-1}). Three sward types were investigated in a farmlet experiment: (1) perennial ryegrass (*Lolium perenne*) (PRG; 205 kg N ha^{-1} yr^{-1}); (2) PRG and white clover (*Trifolium repens*) (PRGWC; 90 kg N ha^{-1} yr^{-1}); (3) MSS (90 kg N ha^{-1} yr^{-1}) consisting of PRG, timothy (*Phleum pratense*), white and red (*Trifolium pratense*) clover, chicory (*Cichorium intybus*) and plantain (*Plantago lanceolata*). Animals offered MSS and PRGWC had higher average daily gain during their first grazing season, their first winter indoors and their second grazing season than animals offered PRG (P<0.05). Carcass characteristics were unaffected (P>0.05) by pasture type, but animals offered PRGWC or MSS reached slaughter three weeks earlier than those offered PRG. Multispecies swards support improved animal performance in a dairy calf to beef system.

Keywords: multispecies swards, dairy-beef, legumes, forage herbs, nitrogen

Introduction

The EU 'Farm to Fork Strategy' targets a reduction in fertilizer nitrogen use of 20% by 2030 with concomitant reductions in nutrient losses and reversals in biodiversity decline (European Commission, 2020). This places a particular focus on pasture-based agricultural systems within Ireland (Rath and Peel, 2005). Temperature pasture-based production systems are frequently reliant on *Lolium perenne* L. (perennial ryegrass) monocultures which require high inputs of nitrogen (N) to achieve high DM yields (Grace *et al.,* 2019a). Recent findings from our group show that productivity of pasture-based sheep production can be improved, while also reducing fertilizer nitrogen input, when animals are offered swards consisting of several species compared to a perennial ryegrass monoculture (Grace *et al.*, 2019b).

Furthermore, Ireland has witnessed an increase of dairy cow numbers of 50% since 2010 (CSO, 2020), largely due to milk quota removal in 2015, with a concurrent increase in the number beef animals originating in the dairy herd. This increase in dairy-origin animals from within the beef herd has the potential to reduce the carbon footprint of beef production (Murphy *et al.*, 2015) although performance of such animals when offered multispecies swards is unknown. The objective of the current study was to assess the impact of offering a multispecies sward to male Hereford cattle from the dairy herd on animal performance, with the hypothesis that cattle offered multispecies swards will have improved performance compared to cattle offered a perennial ryegrass monoculture.

Materials and methods

The study was undertaken on the University College Dublin, Lyons Farm Long-Term Grazing Platform (53°29′N, 6°53′W). The site is divided into three 8 ha farmlets, of three sward types. These are: a *L.*

perenne monoculture receiving 205 kg N ha^{-1} yr^{-1} (PRG), a *L. perenne* and *Trifolium repens* sward receiving 90 kg N ha^{-1} yr^{-1} (PRGWC), and a six-species sward (*L. perenne, Phleum pratense* L., *T. repens, Trifolium pratense* L. (red clover), *C. intybus and P. lanceolata*) receiving 90 kg N ha^{-1} yr^{-1} (MSS). The PRGWC sward comprised 77% *L. perenne*, 13% *T. repens* and 10% unsown species, while MSS comprised 38% *L. perenne*, 4% *Phleum pratense,* 3% *T. repens,* 20% *Trifolium pratense*, 10% *Plantago lanceolata,* 20% *Cichorium intybus* and 2% unsown species. Each farmlet was stocked at 2.5 LU ha^{-1} comprising 20 Hereford cross weanlings (<1 years) followed by 20 Hereford cross yearling steers (1>2 years) from the dairy herd. Each 8 ha farmlet is subdivided into 8×1 ha grazing divisions. Each farmlet was operated as a self-contained unit, with silage production within the 8 ha farmlet, used to provide winter feed for those cattle during the winter housing periods.

The data presented represent the performance of two separate groups per sward type (i.e. Hereford cross weanling steers (June 2020-March 2021) and Hereford cross yearling steers (March 2020-November 2020). Swards were grazed to a target post-grazing residual height of 4 cm above ground level for the PRG and PRGWC farmlets, and 6 cm for the MSS farmlet, in a leader-follower system (yearlings following weanlings). Pre-grazing herbage mass above grazing residual was calculated before stock entered each paddock (target pre-grazing herbage mass was 1,500 kg DM ha^{-1}). Animals live weight was recorded monthly, and animals were selected for slaughter once they reached a liveweight of 620 kg on a treatment basis. Animals in their first year of grazing entered the system at approximately 15 weeks of age in late June 2020 and remained at pasture until housed in early November 2020. These animals were then housed indoors and offered a diet of *ad libitum* silage plus 1.25 kg of concentrate (composition: flaked barley, 500.0 g^{-1} kg^{-1}; flaked oats, 247.5 g^{-1} kg^{-1}; flaked beans, 150.0 g^{-1} kg^{-1}; molasses, 70.0 g^{-1} kg^{-1}, and minerals and vitamins, 32.5 g^{-1} kg^{-1}) until turnout in March 2021. Hereford cross yearling steers commenced grazing in mid-March 2020 and remained at pasture until housing in mid-October 2020. These animals were then adjusted to a diet of silage plus concentrates (as described above) offered at a 50:50 forage to concentrate ratio on a DM basis.

Results and discussion

The effect of sward type on animal performance is presented in Table 1. During the grazing season and during their first winter, housed indoors, animals from the PRGWC and MSS farmlets achieved higher (P<0.05) daily growth rates and subsequently a higher turnout weight than animals from the PRG farmlet. Pettigrew *et al.* (2017) reported improvements in animal performance of Friesian bulls grazing a herb-clover mix which did not contain grass, linked to enhanced nutritive value of the herb-clover mix (Handcock *et al.*, 2015) driving enhanced intakes. However in the current study the MSS also contained approximately 40% grass on a DM basis.

Hereford cross yearling steers from the PRGWC and MSS farmlets achieved higher (P<0.01) daily growth rates at pasture than animals from the PRG farmlet, resulting in higher housing liveweight for PRGWC and MSS (P<0.05). As all animals were selected on a treatment basis to meet the predefined carcass specification there were no differences in the carcass traits measured; however, animals from the PRG group required on average an additional 19 days to reach the desired carcass specification. These findings align with the responses reported in sheep production systems which also reported enhanced animal performance and reduced days to slaughter when animals were offered multispecies swards compared to perennial ryegrass only (Grace *et al.,* 2019a).

Findings from the current study indicate the potential to enhance animal performance in a dairy-calf to beef production system, resulting in a reduced lifetime to slaughter, without compromising carcass traits, with concurrent reductions in the requirement for fertilizer N by altering the botanical composition of the grazed and conserved swards.

Table 1. The impact of sward type on animal performance.[1]

	PRG	PRGWC	MSS	SEM	*P*-value
ADG of weanlings at pasture (kg.d^{-1})	0.70^a	0.85^b	0.82^b	0.02	$P<0.05$
Liveweight of weanlings at housing (kg)	215	242	227	0.73	$P>0.05$
ADG of weanlings indoors (kg.d^{-1})	0.70^a	0.86^b	0.91^b	0.01	$P<0.05$
Liveweight of weanlings at turnout (kg)	317^a	372^b	360^b	5.89	$P<0.05$
ADG of yearlings at pasture (kg.d^{-1})	0.70^a	0.80^b	0.82^b	0.02	$P<0.01$
Liveweight of yearlings at housing (kg)	509^a	534^b	541^b	6.45	$P<0.05$
ADG of finishers indoors (kg.d^{-1})	1.65^a	1.65^a	1.47^b	0.08	$P<0.05$
Carcass weight (kg)	316	312	313	3.95	$P>0.05$
Kill out percentage (%)	51	51	51	0.29	$P>0.05$

[1] Within rows, means with differing superscript letters differ significantly ($P<0.05$).

Conclusions

Accelerating days to slaughter and reductions in the quantity of fertilizer nitrogen required in pasture-based beef production have the potential to enhance the sustainability of pasture-based livestock production systems.

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References

Central Statistics Office (2020) Census of Agriculture 2020. https://www.cso.ie/en/releasesandpublications/ep/p-coa/censusofagriculture2020-preliminaryresults/kf/.

European Commission (2020) *Farm to Fork Strategy – For a Fair, Healthy and Environmentally-Friendly Food System.* Communication from the Commission to The European Parliament, The Council, The European Economic and Social Committee and The Committee of the Regions. Available at: https://ec.europa.eu/info/sites/default/files/communication-annex-farm-fork-green-deal_en.pdf.

Grace C., Boland T.M., Sheridan H., Brennan E., Fritch R. and Lynch M.B. (2019a) The effect of grazing versus cutting on dry matter production of multispecies and perennial ryegrass only swards. *Grass and Forage Science* 74, 437-449.

Grace C., Lynch M.B., Sheridan H., Lott S., Fritch R. and Boland T.M. (2019b) Grazing multispecies swards improves ewe and lamb performance. *Animal 13*, 1721-1729.

Handcock R.C., Hickson R.E., and Back P.J. (2015) The use of herb mix and lucerne to increase growth rates of dairy heifers. *Proceedings of the New Zealand Society of Animal Production* 75, 132-135.

Murphy B., Crosson P., Kelly A.K. and Prendiville R. (2015) An economic and greenhouse gas emissions evaluation of pasture-based dairy calf-to-beef production systems. *Agricultural Systems* 154, 124-132.

Pettigrew E.J., Morris S.T., Back P.J., Kenyon P.R., Berry J., Donald A.J., Lane A.L. and Hickson R.E. (2017) Growth of weaned Friesian bull calves on a herb sward or with concentrate supplementation during late summer and early autumn. *New Zealand Journal of Agricultural Research* 60 (1), 70-79.

Rath M. and Peel S. (2005) Grassland in Ireland and the UK. In: McGilloway D.A. (ed.) *Grassland: A Global Resource*, Proceedings XX International Grassland Congress, Dublin 2005, pp. 13-27.

Efficiency of cows' diets in Galician dairy farms under the feed-food competition perspective

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Abstract

The objective of the present work is to compare dairy cows' feeding systems typically used in the Galician dairy region (NW Spain), in terms of the conversion efficiency ratio of feed into human-edible animal product. A sample of 316 Galician dairy farms was interviewed in 2014 and grouped according to the predominant feeding system used. On average of all systems, 78% of the daily dry matter consumed by a lactating cow comes from non-human consumable food resources. This value reaches 85% in systems based on grass (fresh or silage) and drops to 66% in the most intensive systems, based on maize silage. An average of 0.67 kg of protein potentially usable by humans is consumed by cows to produce 1.0 kg of animal protein, with a range between 0.88 kg kg^{-1} in the most intensive systems and approximately 0.50 kg kg^{-1} in grass-based systems. The results illustrate the valuable contribution of dairy production systems to the global food supply and the variability between different production systems, quantifying the lower food competition in the feed/food dilemma in the most extensive systems.

Keywords: feed efficiency, dairy production, feeding systems

Introduction

The dairy sector is not indifferent to the ongoing debate about competition between animals and humans for available food resources. The demand for food of animal origin by the population shows an increasing trend, particularly in developing countries (Enahoro *et al.*, 2018). The use in animal diets of consumable resources by humans, the competition for the use of land to produce food for humans and animals and the relatively low efficiency of animals to convert feed into human consumable products (Mottet *et al.*, 2017) are questions of growing interest. In the case of ruminants, their ability to digest fibre allows them to obtain energy and protein of high biological value for human consumption from forages and by-products of the food industry, which places them in a vital ecological niche as nutrient vectors for humans from plant cellulosic substrates (Van Soest, 1994). To date, there is no information on the competition between animal/human food uses of the different feeding systems used in dairy farms in Galicia. The objective of this work is to compare, under this perspective, the most common dairy cows' diets in Galician farms considering the efficiency of transformation of dry matter and protein in the animal diet into human-edible products.

Materials and methods

For the elaboration of this work, the data of a study (Flores *et al.*, 2017) carried out at the CIAM have been used, in which information was obtained about the farm characteristics, milk production, land uses and predominant feeding systems of a stratified random sample of 316 Galician dairy farms interviewed in 2014. Based on the proportion of the predominant forage in the total dry matter (DM), four typical diets were considered (MS = maize silage, GS-MS = grass and maize silages, GS = grass silage and FG = fresh grass).

For the classification of the ingredients of the diets used in the farms and the elaboration of the food efficiency indices, the criterion of Mottet *et al.* (2017) was used. According to these authors, cereal grains and protein crops are considered as potentially consumable products by humans, while fresh and silage grasses and legumes, oilseed cakes, milling by-products, beet pulp, straws, reeds and molasses were considered as only consumable by livestock. In the case of maize silage, it was estimated that the proportion of grain (potentially consumable by humans) represented, on average, 40% of the total plant harvested. Dry matter intake (DMI) of lactating cows was estimated according NRC (2001) and this value was increased proportionally according to the replacement heifers and dry cows present in the farm.

The conversion efficiency of feed into animal product was measured by the following indices: (1) ratio between the amount of DM consumed by cows per unit of crude protein (CP) in milk (EFC1 = kg total DMI kg^{-1} CP-milk; EFC2 = kg consumable by humans DM kg^{-1} CP-milk); and (2) ratio between CP consumption by cows and CP production in milk (PEFC1 = kg total CP intake kg^{-1} CP-milk; PEFC2 = kg consumable by humans CP kg^{-1} CP-milk). An analysis of variance was carried out considering the feeding system the group to which each farm belonged as a fixed factor, using the procedure GLM from SAS (SAS Institute, 2009).

Results and discussion

Table 1 shows the average composition of the diet of the different groups, the herd size and the milk production per cow. Silage diets, compared to fresh grass, corresponded with a more intensive system, with higher milk and higher use of concentrate per cow, showing a gradient of intensification related to the importance of maize silage in the ration.

Considering the total farms sampled, the average DMI of the daily diet of dairy cows that does not compete with human food was 780 g kg^{-1} consumed, varying from 661 g kg^{-1} in MS farms to 852 g kg^{-1} in FG farms (Table 2). Total milk CP produced was 0.78 kg cow^{-1} day^{-1} on average, ranging between 1.03 kg for MS and 0.67 kg for FG farms. Efficiency ratios showed that, on average, 33.9 kg of DM are needed to produce 1.0 kg of CP-milk (EFC1), of which only 6.7 kg DM would be consumable by humans (EFC2). These ratios varied significantly between groups, with EFC1 being higher for the grass-based farms and the EFC2 for farms feeding silage-maize based diets. An average amount of 4.7 kg of total protein in feed was needed to produce 1.0 kg of milk protein (PEFC1), of which only 0.67 kg of CP was potentially consumable by humans (PEFC2). Farm type affected significantly these ratios, with values of 3.7 and 4.8 kg of total feed protein kg^{-1} milk protein in MS and FG farms and of 0.88 and 0.50 kg of potentially human-edible feed protein kg^{-1} milk protein in MS and GS farms, respectively.

Table 1. Diet composition, herd size and milk production in each group.[1]

	MS	GS-MS	GS	FG	*P*-value
Diet composition (% DM)					
Fresh grass	0.6 ^{c}	5.6 ^{b}	7.8 ^{b}	39.1 ^{a}	***
Grass silage	17.1 ^{d}	28.4 ^{b}	45.3 ^{a}	22.5 ^{c}	***
Maize silage	39.2 ^{a}	23.5 ^{b}	0.7 ^{d}	7.1 ^{c}	***
Dry forages	4.3 ^{c}	7.0 ^{c}	13.9 ^{b}	5.8 ^{c}	***
Concentrate	38.8 ^{a}	35.4 ^{ab}	32.3 ^{b}	25.5 ^{c}	***
Herd size					
Dairy cows $farm^{-1}$	63.8 ^{a}	42.0 ^{b}	23.8 ^{c}	22.0 ^{c}	***
No. lactations cow^{-1}	3.3 ^{c}	3.9 ^{b}	4.8 ^{a}	4.8 ^{a}	***
Milk production					
Total lactation (kg cow^{-1})	9,491 ^{a}	8,134 ^{b}	7,123 ^{bc}	6,327 ^{c}	***

[1] MS = maize silage; GS-MS = grass and maize silage; GS = grass silage; FG = fresh grass; *** P<0.001.

Table 2. Efficiency ratios by group.[1]

	MS	GS-MS	GS	FG	*P*
Total DM consumed (kg cow^{-1} d^{-1})					
DMI (kg cow $^{-1}$ d^{-1})	27.8 ^{a}	25.0 ^{b}	22.8 ^{c}	22.4 ^{c}	***
DM potentially consumed by humans (kg cow^{-1} d^{-1})					
From maize silage	4.4 ^{a}	2.4 ^{b}	0.0 ^{d}	0.6 ^{c}	***
From concentrates	5.0 ^{a}	4.1 ^{b}	3.5 ^{b}	2.6 ^{c}	***
Total human-edible DM consumed	9.4 ^{a}	6.5 ^{b}	3.5 ^{d}	3.3 ^{d}	***
CP intake (kg cow^{-1} d^{-1})					
Total CP	4.18 ^{a}	3.82 ^{b}	3.59 ^{b}	3.60 ^{b}	***
Human-edible CP	1.01 ^{a}	0.71 ^{b}	0.42 ^{d}	0.38 ^{d}	***
CP output (kg cow^{-1} d^{-1})					
CP-milk	1.14 ^{a}	0.98 ^{b}	0.84 ^{c}	0.74 ^{c}	***
Efficiency ratios					
EFC1 (kg total DMI kg^{-1} CP-milk)	28.1 ^{b}	29.8 ^{ab}	34.7 ^{a}	35.1 ^{a}	***
EFC2 (kg human-edible DMI kg^{-1} CP-milk)	9.3 ^{a}	7.6 ^{b}	4.9 ^{c}	5.1 ^{c}	***
PEFC1 (kg total CP intake kg^{-1} CP-milk)	3.7 ^{c}	3.9 ^{bc}	4.3 ^{a}	4.8 ^{a}	***
PEFC2 (kg human-edible CP kg^{-1} CP-milk)	0.88 ^{a}	0.73^{b}	0.50 ^{c}	0.51 ^{c}	***

[1] MS = maize silage; GS-MS = grass and maize silage; GS = grass silage; FG = fresh grass; *** $P<0.001$.

Information on comparable ratios for dairy farms is scarce, apart from the cited work of Mottet *et al.* (2017) to which the conversion rates of the food consumed by different species of animals into animal protein are referred. For ruminants, these authors indicate values of 133 kg DM kg^{-1} CP for EFC1, 5.9 kg DM kg^{-1} CP for EFC2, 2.0 kg kg^{-1} CP for PEFC1 and 0.6 kg kg^{-1} CP for PEFC2. In the same work, for monogastric species (pigs and poultry) the authors cite values of EFC1: 30 kg MS kg^{-1} CP, EFC2: 15.8 kg MS kg^{-1} CP, PEFC1: 14.0 kg kg^{-1} CP and PEFC2: 2.0 kg kg^{-1} CP.

Conclusions

The results illustrate about the valuable contribution of dairy production systems to the global food protein supply, with an average net return of 1.0 kg of protein of high biological value for each 0.67 kg of feed protein potentially consumable by humans. The grass-based systems showed a higher efficiency, with an output of high biological value protein in milk that doubled the amount of human-edible protein consumed in the feed.

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References

Enahoro D., Lannerstad M., Pfeifer C. and Dominguez-Salas P. (2018) Contributions of livestock-derived foods to nutrient supply under changing demand in low- and middle-income countries. *Global Food Security* 19, 1-10.

Flores G., Martínez-Fernández A., Doltra J., García A. and Eguinoa P. (2017) *Encuesta sobre estructura y sistemas de alimentación de las explotaciones lecheras de Galicia, Cornisa Cantábrica y Navarra*. INTIA, Pamplona, Spain, 52 pp.

Mottet A., de Haan C., Falcucci A., Tempio G., Opio C. and Gerber P. (2017) Livestock: On our plates or eating at our table? A new analysis of the feed/food debate. *Global Food Security* 14, 1-8.

National Research Council (2001) *Nutrient Requirements of Dairy Cattle: Seventh Revised Edition, 2001*. The National Academies Press, Washington, DC, USA, 405 pp.

SAS Institute (2009) SAS/Stat User's Guide, v.9.2, SAS Institute Inc., Cary, NC, USA.

Van Soest P.J. (1994) *Nutritional Ecology of the Ruminant*. 2nd ed. Cornell Univ. Press, Ithaca, NY, USA.

Sainfoin grazing by dairy goats to manage gastro-intestinal parasitism and improve milk performance

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Abstract

Optimal use of grazing pastures represents a solution for improving goat production by ensuring protein self-sufficiency and sustainability. However, in dairy goats grazing systems, infections with gastro-intestinal nematodes (GINs) are a major threat for goats' health and welfare. The usual control method for GINs has relied on chemical anthelminthics (AHs). However, resistance to AHs is now a worldwide issue. Sainfoin (*Onobrychis viciifolia*) is a forage legume containing tannins, which represents a solution to limit GIN infections and the development of AH resistance. A 23-day trial was performed with 2 homogeneous groups of 30 dairy goats grazing either sainfoin or multi-species pastures without sainfoin. Objectives were to evaluate the effects of sainfoin grazing on milk performance and on GIN infections. Compared to the multi-species pasture, grazing sainfoin led to greater milk production (+12%, $P<0.05$) and milk urea concentration, due to greater pasture crude protein concentration (+4%; $P<0.05$). Mean faecal egg count of GINs (FEC) in the Sainfoin group decreased between the beginning and the end of the trial (-66%; $P<0.001$) although the final FEC did not differ between the 2 groups ($P=0.072$). This experiment confirms that sainfoin is an interesting forage for grazing by goats. Its impact on parasitism under natural conditions must be confirmed.

Keywords: goat, grazing, sainfoin, parasitism, milk performance

Introduction

France is the largest producer of goat's milk in Europe. Since 2000, French dairy goat farms have turned towards intensive farming, largely through increasing purchased inputs, particularly feeds. To improve sustainability of dairy goat farms, a larger use of grazing, in particular of legume-based pastures, might be a solution (Lüscher *et al.*, 2014). The development of grazing, however, increases the risk of gastro-intestinal nematode (GIN) infestation, particularly in goat systems (Hoste *et al.*, 2012). One lever to limit the negative impacts of parasitism is to use plants with bioactive secondary metabolites (BSM), such as sainfoin (*Onobrychis viciifoliae*) which contains tannins (Hoste and Niderkorn, 2019). The aim of this study was to evaluate, under conditions of natural infestation, the impacts of sainfoin grazing on milk performance and as a solution to limit GIN infestation level.

Materials and methods

This trial was carried out according to a continuous design during 23 days in spring 2021 (12 May to 03 June), at the INRAE experimental farm FERLus-Patuchev (Lusignan, New Aquitaine; 46.43°N, 0.12°E). Two treatments were compared: grazing pure sainfoin (namely S) and grazing a multi-species pasture without BSM plants (namely C = control). Two balanced groups of 30 goats (5 primiparous and 25 multiparous) were created based on their individual characteristics measured during a reference week (26 to 30 April): lactation number (3.0±1.7 lactations), stage of lactation (55±16 days in milk), milk production (3.5±0.7 kg d^{-1}), milk fat concentration (34.1±5.6 g kg^{-1}), milk protein concentration (32.8±3.2 g kg^{-1}), body weight (57.2±9.1 kg) and GIN eggs per gram of faeces (321±444 EPG). Each group was then assigned to one of the two treatments for the duration of the trial.

Goats were bred and fed in a goat shed and had access to pasture 10.5 h d^{-1} over two sessions, 6.5 h between AM and PM milkings (from 09:30 to 16:00) and 4 h after the PM milking (from 17:00 to 21:00). Goats were milked twice daily at 08:15 and 16:30. Each goat received 65 g dry matter (DM) d^{-1} of a commercial protein-rich concentrate (300 g crude protein kg^{-1} DM) at milking times, 138 g DM d^{-1} of a mixture of whole grains (triticale 68%; peas 19%; faba bean 13%) and 135 g DM d^{-1} of a commercial energy-rich concentrate twice daily through an automatic feeder at 08:45 and 16:45 (i.e. 675 g DM d^{-1}). Daily pasture allowance was 2.5 kg dry matter (DM) d^{-1} measured at 4.8 cm above ground level and goats received no forage supplement. These grazing conditions were considered as non-limiting for herbage intake (Charpentier *et al.*, 2019a,b). The two groups of goats grazed under a strip-grazing system, with the front fence moved after every morning milking and the back fence moved twice weekly. The area allocated daily to each treatment was calculated from a daily estimate of pre-grazing pasture mass by multiplying daily pre-grazing sward height (from a plate meter) by sward bulk density (from cut strips). The pastures used were sown in spring 2020. The control pasture was a mixture of perennial ryegrass, tall fescue, timothy, red clover, white clover and inoculated lucerne, and sainfoin pasture was based on pure sainfoin (cv Perly, 147 kg ha^{-1} of unshelled seeds). For chemical composition, 13 daily samples were collected from days 8 to 22 from handfuls of pasture, cut with scissors (>5 cm), randomly selected in the offered area each day.

After one week of adaptation (week 1), individual milk production and composition were measured during 4 days every week. Each goat was weighed on two consecutive days at the beginning and at the end of the trial. Goats were also weighed 3 days after the end of the trial to avoid a possible difference in digestive content between the two groups. Faeces were individually sampled during the reference week, at the beginning (day 0) and at the end (day 20) of the trial, to measure faecal egg count of GIN (FEC) by using the McMaster method.

Milk production, milk composition and body weight were analysed according to the Mixed procedure of SAS Institute (2013). The model included the effects of treatment (n=2), week (n=3; reference, experimental week 2, experimental week 3) and their interaction. The FEC variable was analysed after square root transformation in order to normalize the variances, and was modelled using the Mixed procedure of SAS (2013). The model included the fixed effects of treatment (n=2), the collected day (n=3; reference, day 0; day 21) and their interaction. Repeated measures data of all models were analysed using a Variance Components structure. Goats are introduced as random effect. The FEC results are presented with no transformation. The significance level used was $P \leq 0.05$ for all models used.

Results and discussion

Pasture crude protein concentration was greater by 35 g kg^{-1} DM on S than on C (191 vs 156 g kg^{-1} DM). The NDF concentration was not different between sward types (410 g kg^{-1} DM) and ash concentration was lower on S than on C (79 vs 92 g kg^{-1} DM). Compared to the multi-species sward, sainfoin grazing was associated with a higher milk production by 14% in experimental week 3 ($P<0.05$) (Table 1). Milk urea concentration was greater in experimental week 1 in S than in C (+12%; $P<0.05$), due to greater pasture crude protein concentration. This content increased for the two groups between week 2 and 3 and was no longer different at the 5% threshold in week 3. No difference between the groups was demonstrated for other milk components and body weight. All goats lost 1.4 kg on average during the trial, which is typical for this stage of lactation.

The FEC in the S group decreased between day 0 and day 20 of the trial (-66%; $P<0.001$), unlike the C group (Table 1). At day 20, the FEC was not different between the groups, although a statistical trend was noticed ($P=0.072$). Pure sainfoin, when grazed in spring, does seem to be an interesting forage in terms of its effect in limiting GIN infestation level. However, it would be interesting to study sainfoin effects in other periods of the year because concentration of tannins is variable (Theodoridou *et al.*, 2011).

Table 1. Effects of sward type and week on milk production and milk urea concentration, and of collection day (0 vs 20) on faecal egg count of GIN of goats.[1]

		Reference	Week 2	Week 3
3.5% fat corrected milk yield (kg d^{-1})	Control	3.48^b	3.49^b	3.16^a
	Sainfoin	3.47^a	3.75^b	3.59^a
	P-value (S vs C)	0.930	0.181	0.016
Milk urea concentration (mg l^{-1})	Control	180^a	351^b	403^c
	Sainfoin	169^a	394^b	431^c
	P-value (S vs C)	0.212	0.003	0.065
		Reference	**Day 0**	**Day 20**
Faecal egg count of GIN (eggs g^{-1})	Control	318	430	315
	Sainfoin	323^a	463^b	158^a
	P-value (S vs C)	0.546	0.468	0.072

[1] In the same row (time effect), adjusted means with different superscript letters are significantly different ($P<0.05$).

Conclusions

Sainfoin is a very palatable forage for dairy goats and in this study milk production was maintained despite the plant's stage of evolution. It appears to be a valuable legume in goat diets; however, we have observed on farms that this plant is not very perennial (2-3 years). The impact on GINs under natural conditions remains to be explored further in terms of using sainfoin as an alternative solution to anthelmintics.

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References

Charpentier A., Caillat H., Gastal F. and Delagarde R. (2019a) Intake, milk yield and grazing behaviour of strip-grazing Alpine dairy goats in response to daily pasture allowance. *Animal* 13, 2492-2500.

Charpentier A., Caillat H., Gastal F. and Delagarde R. (2019b) Intake, milk production and grazing behaviour responses of strip-grazing dairy goats to daily access time to pasture and to dehydrated lucerne supplementation. *Livestock Science* 229, 90-97.

Hoste H. and Niderkorn V. (2019) Le sainfoin et la chicorée: deux modèles de plantes bioactives pour répondre aux défis agroécologiques en élevage de ruminants. *Fourrages* 238, 319-328.

Hoste H., Manolaraki F., Arroyo-Lopez C., Torres Acosta J.F.J. and Sotiraki S. (2012) Spécificités des risques parasitaires des chèvres au pâturage: conséquences sur les modes de gestion. *Fourrages* 212, 319-328.

Lüscher A., Mueller-Harvey I., Soussana J.F., Rees R.M. and Peyraud J.L. (2014) Potential of legume-based grassland–livestock systems in Europe: a review. *Grass and Forage Science* 69, 206-228.

SAS Institute Inc. (2013) *Base SAS 9.4®. SAS User's Guide: Statistics*. SAS Institute Inc., Cary, NC, USA

Theodoridou K., Aufrere J., Andueza D., Le Morvan A., Picard F. and Stringano E. (2011) Effect of plant development during first and second growth cycle on chemical composition, condensed tannins and nutritive value of three sainfoin (*Onobrychis viciifolia*) varieties and lucerne. *Grass Forage Science* 66, 402-414.

Dairy goats grazing plantain: milk performance and consequences on gastro-intestinal parasitism

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Abstract

Dairy goats are very sensitive to gastrointestinal nematodes (GIN) under grazing systems. Plantain (*Plantago lanceolata* L.), because of its expected medicinal properties, could help to manage GIN parasitism. Two homogeneous groups of 25 goats were created to evaluate the impact of grazing plantain on milk production and resilience to GIN. During 23 days, with 8 h of daily access to pasture, a pure plantain (PLA) pasture was compared to a multi-species pasture without plantain (MSS). Goats were supplemented with 856 g of concentrate and 200 g hay daily. Pasture CP concentration was 102 and 170 g kg^{-1} DM in PLA and MSS, respectively, due to the late stage of growth of the plantain. Live weight, GIN egg excretion level, milk protein content, and somatic cells count were not affected by the sward type. Milk production was lower (-16%, $P<0.001$) for PLA than for MSS, as was milk urea concentration (-16%, $P<0.001$). The plantain effect on the resilience of goats to parasitism was not been demonstrated. Grazing pure plantain during 23 days in spring did not show any specific interest. Plantain should, however, be tested in a mixed sward and at an earlier stage of growth when its quality would be higher.

Keywords: goat, grazing, plantain, parasitism, milk performance

Introduction

France is the highest producer of goats' milk in the EU. French goat farms have a low feed self-sufficiency, about 61% (Brocard *et al.*, 2016). Ruminant production systems based on grazing provide solutions to economic, environmental and societal issues (Michaud *et al.*, 2020). The development of grazing, however, increases the risk of GIN, particularly in goat systems (Hoste *et al.*, 2012). One lever to limit the negative impacts of parasitism is to use plants with bioactive secondary metabolites (BSM) such as plantain. Bioactive secondary metabolites are molecules that are not involved in basic plant metabolism but provide defence functions against predators or pathogens (Hoste and Niderkorn, 2019). Plantain contains aucubin, and positive effects of aucubin on animal resilience and its anti-parasite effect have been demonstrated in ewes and lambs (Judson *et al.*, 2009). The aim of this study was to evaluate milk performance and consequences on parasitism of grazing plantain in dairy goats.

Materials and methods

This trial was carried out according to a continuous design during 23 days in spring 2021 (12 May to 03 June), at the INRAE experimental farm FERLus-Patuchev (Lusignan – New Aquitaine- 46.43°N, 0.12°E). Two treatments were compared: grazing pure plantain (namely PLA) and grazing a multi-species pasture without plantain (namely MSS). Two balanced groups of 25 goats (11 primiparous and 14 multiparous) were created according to their individual characteristics measured from 27 April to 30 April: lactation number (2.3±1.6 lactations), stage of lactation (218±11.7 days in milk), milk production (2.1±0.6 kg d^{-1}), milk fat concentration (34.9±6.6 g kg^{-1}), milk protein concentration (38.9±4.3 g kg^{-1}), body weight (56.0±10.8 kg) and GIN eggs per gram of faeces (55.0±64.8 EPG). The difference between groups did not exceed 2% for any of these variables, except for lactation number (13%). Each group was assigned to one of the two treatments for the duration of the experiment.

Goats were bred and fed in the goat shed and had access to pasture from 09:00 to 17:00 every day, with a daily pasture allowance of 2.1 kg dry matter (DM) d^{-1} measured at 5 cm above ground level. These grazing conditions were considered as non-limiting for herbage intake (Charpentier *et al.*, 2019a,b). The two groups of goats grazed under a strip-grazing system, with the front fence moved daily after morning milking and the back fence moved twice weekly. The area allocated daily to each treatment was calculated from a daily estimate of pre-grazing pasture mass by multiplying daily pre-grazing sward height (from a plate meter) by sward bulk density (from cut strips). The pastures used were sown in spring 2019. The MSS pasture was a mixture of perennial ryegrass, tall fescue, timothy, red clover, white clover and inoculated lucerne, and the PLA pasture was based on plantain (cv Ceres Tonic, 12 kg ha^{-1}) and white clover (1 kg ha^{-1}). For chemical composition, 13 daily samples were collected from days 8 to 22 from handfuls of pasture, cut with scissors (>5 cm), randomly selected in the offered area each day. Each goat received 65 g DM d^{-1} of a commercial concentrate, fed at milking time and 365 g DM d^{-1} of mixture of whole grains twice daily through an automatic feeder at 08:30 and 17:30 (i.e. 730 g DM d^{-1}). After PM milking, each group received a supplementation of hay (182 g DM $goat^{-1}$ d^{-1}). The mixture of whole grains consisted of the following ingredients: barley 65%; oat 21%; sunflower seed 10%; vetch 2%; and peas 2%.

Goats were milked once a day. After one week of adaptation, individual milk production and composition were measured during 4 days every experimental week. Each goat was weighed on two consecutive days at the beginning and at the end of the trial. Goats were also weighed 3 days after the end of the trial to avoid a possible difference in digestive content between the two groups. Individual faeces were collected during the reference week, at day 0 and day 20 of the trial to measure faecal egg count of GIN (FEC) by using the McMaster method.

Milk production, milk composition and body weight for which a covariate from the reference period was available were analysed according to the Generalised Linear Model of SAS Institute (2013). The models include the effects of treatment (n=2), parity (n=2) and the interactions between these effects. Parasitology data were analysed with the non-parametric Wilcoxon-Mann-Whitney test to compare the mean infestation levels between groups and their evolution during the trial. The significance level used was $P \leq 0.05$ for all models used.

Results and discussion

Pasture crude protein concentration was greater by 68 g kg^{-1} DM on MSS than PLA, and NDF concentration was lower by 94 g kg^{-1} DM (389 vs 483 g kg^{-1} DM). Ash concentration of plantain was lower than of MSS (92 vs 100 g kg^{-1} DM), contrary to what the literature indicated (Novak *et al.*, 2020).

There were no effects of parity or of the parity × sward type interaction. Body weight, FEC level, milk protein concentration, and somatic cells count were not affected by the sward type. Milk production was lower (-16%, $P<0.001$) for PLA than for MSS, as was milk urea concentration (-16%, $P<0.001$) (Table 1). Milk fat concentration was significantly greater in PLA than in MSS (+5.1 g kg^{-1}, $P<0.001$). These results can be explained by the difference in quality of the pastures grazed, their palatability and probably their herbage intake.

Goats were, however, at the end of lactation (249 days in milk), and should have gained weight, especially primiparous goats, highlighting a lack of energy in the diet. The offered plantain had a clear lower nutritive value than the mixed sward, even if goats consumed few plantain stems, and mainly the leaves and the flowers.

Table 1. Adjusted means of milk production, milk composition according to the sward type.

Variable	Pasture type		*P*-value
	Plantain	Multi-species sward	
Milk production (kg d^{-1})	1.86	2.21	<0.001
Milk fat concentration (g kg^{-1})	39.1	34.0	<0.001
Milk protein concentration (g kg^{-1})	36.2	36.9	0.0822
Milk urea concentration (mg l^{-1})	239	284	<0.001

The FEC was low (0 to 350 EPG), with no significant difference between sward type, which means difficult conditions to evaluate any potential effect of PLA on parasitism.

Plantain does not seem to be an interesting forage for goats when grazed as a pure sward over several weeks. However, it would be interesting to study plantain effects in a context of greater initial infestation of goats, at an earlier stage of the plantain growth or at the beginning of lactation. It should also be studied in mixed swards, but this would make it more difficult to evaluate its impact on goats' performance and health.

Conclusions

Although it had no impact on the weight of the goats and allowed a decrease in the milk urea concentration, grazing pure plantain during 23 days in spring did not show any interest with regard to the loss of milk production. Moreover, its effectiveness in the management of gastro-intestinal parasitism in low-infested grazing dairy goats has not been proven.

Acknowledgements

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References

Brocard V., Jost J., Rouillé B., Caillaud D., Caillat H. and Bossis N. (2016) Feeding self-sufficiency levels in dairy cow and goat farms in Western France: current situation and ways of improvement. *Grassland Science in Europe* 21, 53-55.

Charpentier A., Caillat H., Gastal F. and Delagarde R. (2019a) Intake, milk yield and grazing behaviour of strip-grazing Alpine dairy goats in response to daily pasture allowance. *Animal* 13, 2492-2500.

Charpentier A., Caillat, H., Gastal, F. and Delagarde, R. (2019b) Intake, milk production and grazing behaviour responses of strip-grazing dairy goats to daily access time to pasture and to dehydrated lucerne supplementation. *Livestock Science* 229, 90-97.

Hoste H., Manolaraki F., Arroyo-Lopez C., Torres Acosta J.F.J. and Sotiraki S. (2012) Spécificités des risques parasitaires des chèvres au pâturage: conséquences sur les modes de gestion. *Fourrages* 212, 319-328.

Hoste H. and Niderkorn V. (2019) Le sainfoin et la chicorée: deux modèles de plantes bioactives pour répondre aux défis agroécologiques en élevage de ruminants. *Fourrages* 238, 319-328.

Judson H., McAnulty R.W. and Sedcole J.R. (2009) Evaluation of 'Ceres Tonic' plantain (*Plantago lanceolata*) as a lactation feed for twin-bearing ewes. *Proceedings of the New Zealand Grassland Association* 71, 201-205.

Michaud A., Plantureux S., Baumont R. and Delaby L. (2020) Les prairies, une richesse et un support d'innovation pour des élevages de ruminants plus durables et acceptables. *INRAE Productions Animales* 33(3), 153-172.

Novak S., Barre P., Delagarde R., Mahieu S., Niderkorn V. and Emile J. C. (2020) Composition chimique et digestibilité *in vitro* des feuilles d'arbre, d'arbuste et de liane des milieux tempérés en été. *Fourrages* 242, 35-47.

SAS Institute Inc. (2013) Base SAS 9.4®. SAS User's Guide: Statistics. SAS Institute Inc., Cary, NC, USA.

The effect of incorporating white clover into sheep grazed swards on lamb and sward performance

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Abstract

Ireland's competitive advantage in sheepmeat production is based on the efficient production and utilization of pasture. Challenges facing the agricultural sector are based on maintaining or improving current levels of production to maintain an economically viable sector but with an enhanced focus on environmental sustainability and a reduced dependence on chemical nitrogen use. The aim of this study was to assess the influence of incorporating white clover into sheep grazed swards on lamb performance and pasture production. A farm systems experiment was established in 2018 and ran for three years. Three pasture treatments were investigated: (1) perennial ryegrass (PRG) only, receiving 145 kg N ha^{-1} yr^{-1} (GO); (2) PRG plus white clover, receiving 145 kg N ha^{-1} yr^{-1} (GCHN); and (3) PRG plus white clover receiving 90 kg N ha^{-1} yr^{-1} (GCLN). Within these systems, detailed sward measurements and animal performance were recorded. Results show that inclusion of white clover in the sward, relative to perennial ryegrass alone, resulted in lambs reaching slaughter weight 9 days earlier on average. In terms of sward dry matter production there was no difference between any of the three pasture treatments, resulting in a positive environmental and economic result for the GCLN treatment.

Keywords: grassland, grazing, sheep, clover, nitrogen

Introduction

Ireland's competitive advantage in sheepmeat production is based on the efficient production and utilization of pasture. Perennial ryegrass is the most dominant forage grown in Ireland (DAFM, 2020). It can produce high dry matter yields, especially in spring and autumn, reducing the seasonality of production. It can however be difficult to maintain sward quality at certain times of the year especially during the plants reproductive phase. It also requires relatively high levels of chemical nitrogen application to maximize its growth potential. Challenges facing the agricultural sector are based on maintaining or improving current levels of production to maintain an economically viable sector but with an enhanced focus on environmental sustainability and a reduced dependence on chemical nitrogen use (European Commission, 2020). The incorporation of white clover into pasture-based production systems can reduce the need for chemical nitrogen application, and increases the nitrogen-use efficiency of the farm system. The aim of this study was to assess the influence of incorporating white clover into sheep grazed swards on lamb performance and pasture production.

Materials and methods

This study was undertaken at the Sheep Research Demonstration Farm, Teagasc, Animal and Grassland Research Centre, Mellows Campus, Athenry, Co. Galway, Ireland (54°80'N; 7°25'W) from March 2018 for 3 production years (2018-2020). Three pasture treatments were investigated in the study: (1) perennial ryegrass (PRG) only, receiving 145 kg N ha^{-1} yr^{-1} (GO); (2) PRG plus white clover, receiving 145 kg N ha^{-1} yr^{-1} (GCHN); and (3) PRG plus white clover receiving 90 kg N ha^{-1} yr^{-1} (GCLN). Within these systems detailed sward measurements and animal performance were recorded. The experiment was a completely randomized design. Animals were blocked for ewe age, live weight, body condition score, litter size and breed and then randomly allocated to pasture treatment. Ewe breed was predominately Belclare and Suffolk crossbred ewes mated to Charollais rams. There were 55 ewes rearing approx. 1.6

lambs ewe^{-1} in each treatment group at a stocking rate of 11 ewes ha^{-1}. Animals remained on the same farmlet treatment for the duration of the experiment unless they were culled or died.

Post-lambing, ewes and lambs were turned out to pasture and grazed in a rotational grazing system. Target pre-grazing sward heights were 7-9 cm (1,200-1,500 kg dry matter (DM) ha^{-1}) across all treatments for the duration of the experiment. Target post-grazing sward height was 3.5 cm for the first rotation and 4.5 cm for all subsequent pre-weaning rotations. Lambs were weaned on average at 14 weeks of age, with a leader follower grazing system in place thereafter. Lambs were removed from the paddocks at a target post-grazing height of 6 cm, with ewes immediately introduced to graze to a target post-grazing height of 4.5 cm. Herbage availability (herbage mass; kg DM ha^{-1}) was estimated weekly using the rising plate meter method and recorded on Pasturebase (a grass budgeting management tool; Pasturebase Ireland). Prior to each grazing event a quadrat (0.25 m^2) was cut to height of 3.5 cm above ground level using Bosch Isio shears, (Bosch Power Tools, GmbH, Ulm, Germany). The entire sample was collected, weighed and separated into grass and white clover fractions, dried overnight for 16 h at 90 °C to determine DM yield.

Lambs were weighed at birth, 6, 10 and 14 weeks of age (weaning), and at 2-week intervals from weaning to slaughter. All lambs received an anthelmintic treatment at 6 weeks of age to combat *Nematodirus* infection. Beginning from 6 weeks of age, one pooled faecal sample per group was collected fortnightly and faecal egg counts were determined using the FECPAK technique. Subsequent anthelmintic treatments were administered when faecal egg counts exceeded 500 eggs per gram. Average daily gain (ADG) and days to slaughter (DTS) were calculated accordingly. Lifetime ADG = drafting live weight minus birth weight divided by DTS was calculated as the difference from birth date to slaughter date. Lambs were drafted for slaughter at live weights of 42-46 kg over the months June-October respectively to produce a target carcass weight of 20 kg. Lamb performance was analysed using mixed model in SAS 9.4 with sward type, year, litter size, sex and dam parity included as fixed effects and dam included as a random effect.

Results and discussion

Results show pasture treatment (P<0.01) had a significant effect on lamb lifetime ADG and days to slaughter (Table 1). Lambs in the perennial grass plus white clover treatments had a higher growth rate and lower days to slaughter compared to the perennial ryegrass-only pasture treatment. Pasture treatment had no significant effect on carcass grade, fat score or dressing proportion.

There was no significant difference in sward DM production, averaging 12.7 tons DM ha^{-1} as shown in Figure 1. Average sward white clover content across the grazing season was 12.3 and 14.3% for the GCHN and GCLN treatments, respectively.

Table 1. Effect of pasture treatment on lamb average daily gain (ADG in g day^{-1}), days to slaughter and slaughter characteristics.[1]

	GO	GCHN	GCLN	SEM	*P*-value
ADG lifetime (g day^{-1})	211^a	224^b	219^b	4.0	<0.01
Days to slaughter	205^a	194^b	198^b	5.23	<0.01
Carcass conformation	2.72	2.66	2.66	0.07	NS
Fat score	2.97	2.98	2.9	0.08	NS
Dressing proportion	0.46	0.46	0.46	0.004	NS

[1] Pasture treatments are: GO = PRG only +145 kg N ha^{-1} yr^{-1}; GCHN = PRG plus white clover +145 kg N ha^{-1} yr^{-1}; GCLN = PRG plus white clover + 90 kg N ha^{-1} yr^{-1}.

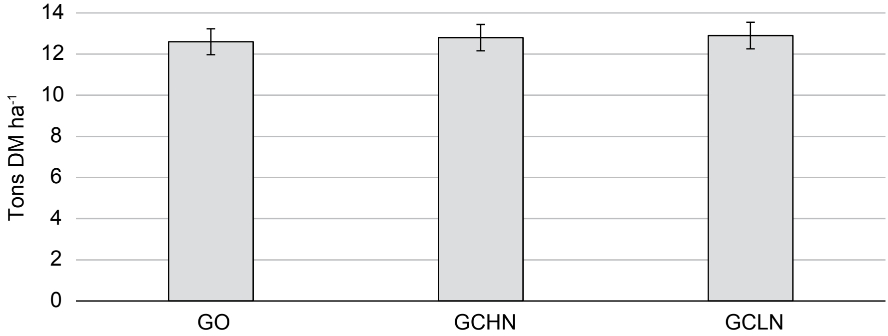

Figure 1. Effect of pasture treatment on sward dry matter yield (t DM ha^{-1}).

Conclusions

Results show that the inclusion of white clover in the sward, compared with swards of perennial ryegrass alone, resulted in lambs reaching slaughter weight earlier. In terms of sward DM production there was no effect of the pasture treatment, thus resulting in a positive environmental and economic result for the GCLN treatment.

References

DAFM (2020) *Grass and White Clover Varieties, Irish Reccommended List 2020*. Department of Agriculture, Food and the Marine, Dublin, Ireland.

European Commission (2020) *Farm to Fork Strategy*.

Dry matter intake and weight gain of grazing heifers on tall fescue and perennial ryegrass

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Abstract

Climate change results worldwide in more extreme weather conditions like longer dry periods during the growing season. Increased frequency of these periods jeopardizes the production of high quality forage grass. Tall fescue (*Festuca arundinacea* Schreb.) is less sensitive to summer drought than perennial ryegrass (*Lolium perenne* L.), however, grazed tall fescue is supposed to lead to less voluntary intake and a lower digestibility, inhibiting the adoption of this species in North-West European dairy production. There is, however, a paucity of results, particularly for the new varieties of tall fescue bred for improved digestibility. We hypothesized that the liveweight gain of grazing dairy heifers is comparable when grazing on recently bred varieties of tall fescue and perennial ryegrass. To investigate this, pastures with either perennial ryegrass or tall fescue were grazed by two groups of heifers in a cross-over design. Surprisingly, heifers grazing tall fescue showed a significantly greater liveweight gain, whereas dry matter intake was not different. Although more research over different years and conditions is needed, this finding underpins the value of tall fescue for grazing.

Keywords: dairy cow, grazing, ryegrass, tall fescue, liveweight gain, dry matter intake

Introduction

In North-West Europe more periods of summer drought are expected due to climate change (IPCC, 2018). Increased frequency of these periods jeopardizes the production of high quality forage grass. Ryegrasses (*Lolium* sp.) dominate the grassland (Haquin, 2012) but they are relatively sensitive to drought stress (Frame, 1992). Tall fescue (*Festuca arundinacea* Schreb.) is more drought-tolerant (Graiss *et al.*, 2011), resists cold temperatures and flooding (Gilbert and Chamblee, 1965) and adapts to different kinds of soil conditions (Burns and Chamblee, 1979). With the same amount of fertilizer tall fescue has a similar crude protein content and a 20-30% higher yield compared to ryegrass (Cougnon *et al.*, 2013). Despite these advantages of tall fescue, the lower voluntary intake and digestibility of tall fescue compared to ryegrass (Luten and Remmelink, 1984) inhibit the adoption of this species in North-West European dairy production. In the last decades several new varieties with higher digestibility have been developed but as yet there are few results of animal trials with these new varieties. In this research we hypothesized that liveweight gain and dry matter intake of grazing heifers is comparable when grazing on newly bred varieties of tall fescue and perennial ryegrass.

Materials and methods

The trial was sown on 17 October 2020 on a sandy loam soil in Melle, Belgium. The previous crop was potatoes. The land was divided into four paddocks of 36×156 m of which two were sown with perennial ryegrass (LP) and two with tall fescue (FA). Within each LP paddock one half was sown with the diploid cv. Barmazing (Lp2), and the other half with tetraploid cv. Melforce (LP4). Within each FA paddock one half was sown with cv. Paolo (FA1), and the other half with cv. Apalona (FA2). Mineral fertilization was applied at the beginning of March 2021 with 94.5 kg N ha^{-1} and 120 kg K ha^{-1}. At the end of March Primstar (2.5 g l^{-1} Florasulam + 100 g l^{-1} fluroxypyr) was used for weed control at 1.5 litre ha^{-1}. After the first cut on 1 June

2021 all paddocks were fertilized with 64.5 kg N ha^{-1} and 80 kg K ha^{-1}. A cross-over design with 2 periods of 4 weeks started with 8 grazing heifers per group on 24 June. Because of low grass growth and availability, caused by cold weather and an open turf, we limited the number of animals per group to 6 after one week for the remaining of the trial. On 20 July (start of the second period), each group changed to another paddock and different grass species. On 1 July a second cut had been made on these 2 paddocks. On 2 August the animals switched back to the paddocks of the first treatment period but remained on the same grass species. The trial ended on 19 August. To measure liveweight gain of the grazing animals, each animal was weighed on two consecutive days at the beginning and end of each treatment period. The weighing was performed in a nearby shed with a built-in scale. In addition to liveweight gain we also estimated dry matter intake (DMI). We determined dry matter yield (DMY) on 10.5 m^2 plots (4 per strip, 8 per treatment) at the beginning and end of each treatment using a Haldrup© (Mod. GR, No. 325). Regrowth on the pasture during the treatment was measured with grazing cages (4 per strip, 8 per treatment, 9 m^2 per cage).

Data analysis

Heifer live weight gains were analysed using a linear mixed model with treatment and period as fixed factor and animal as random effect. All statistical analyses were performed using the statistical software program R (R Core Team).

Results and discussion

Data from one outlier (cfr. precautionary principle) and one animal that became sick at the end of the trial were excluded from the analysis, resulting in 10 observations per treatment. Heifers grazing on Fa1+Fa2 had a higher liveweight gain (0.9±0.1 kg $heifer^{-1}$ day^{-1}) compared to heifers grazing on Lp2+Lp4 (0.6±0.1 kg cow^{-1} day^{-1}) ($P<0.05$). Weight gain was clearly lower in the second period, although dry matter intake was higher (Table 1). Dry matter intake over periods and treatments varied between 6.2 and 7.4 kg DM $heifer^{-1}$ day^{-1}. When comparing DMI with ADG, we assume underestimation of the DMI in the first period. Average daily growth corrected for the period effect was lower for heifers grazing on Lp2+Lp4 in comparison with Fa1+Fa2 (Figure 1). For both periods the between-animal variation was lower in animals grazing on Lp2+Lp4 than on Fa1+Fa2 (Figure 1 and 2). For three animals (grazing in different groups) the corrected growth was lower on Fa1+Fa2 than for Lp2+Lp4. (Figure 3).

Table 1. Average estimated dry matter intake (DMI in kg $heifer^{-1}$ day^{-1}) and average daily weight gain (ADG in kg $heifer^{-1}$ day^{-1}) (LSMeans ± standard error of the mean).

Group	Period	Treatment	DMI	ADG
1	1	Lp2+Lp4	6.2	1.0±0.1
2	1	Fa1+Fa2	6.4	1.1±0.1
1	2	Fa1+Fa2	6.8	0.6±0.1
2	2	Lp2+Lp4	7.4	0.2±0.1

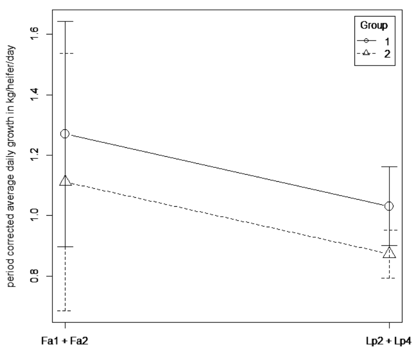

Figure 1. Period corrected average daily growth (kg $heifer^{-1}$ day^{-1}) of grazing heifers on ryegrass (LP2+Lp4) and tall fescue (Fa1+Fa2).

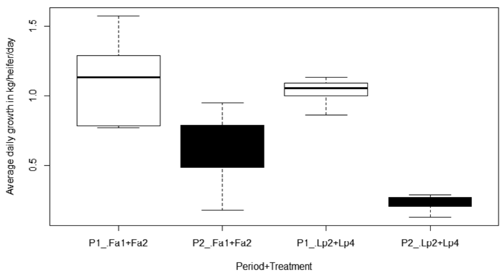

Figure 2. Boxplot of liveweight gain (kg heifer^{-1} day^{-1}) of grazing heifers on ryegrass (Lp2+Lp4) and tall fescue (Fa1+Fa2).

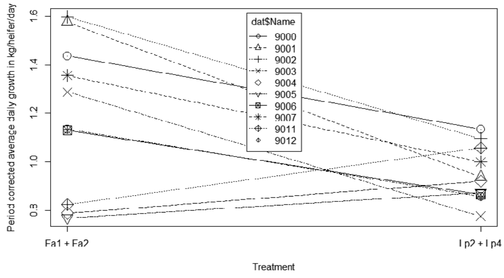

Figure 3. Period corrected average daily growth (kg heifer^{-1} day^{-1}) for all animals grazing on ryegrass (Lp2+Lp4) and tall fescue (Fa1+Fa2).

Conclusions

This is the first trial of a series that will be done on these adjacent paddocks with the same sensitivity conditions. Despite the low number of observations in this trial and a short period, we found a significantly better liveweight gain on tall fescue than on perennial ryegrass. Although some observations on DMI were probably underestimated, we found a similar DMI for both treatments. This underpins the good intake under grazing of these new varieties of tall fescue and is promising for further implementation of tall fescue on farms in North-West Europe. Because there were no drought events during this experimental period we cannot draw any conclusions regarding resilience, but based on previous trials it is expected that tall fescue will perform better, and keep animal productivity on a better level. Further research questions are:

- Can we replicate these results in different seasons and different years?
- How can we fit tall fescue into common rations of ruminants without losing animal productivity while also considering the economic and ecological aspects?
- Is there a difference in CH_4 production, so that we do not aggravate climate mitigation issue?

References

Burns J.C. and Chamblee D.S. (1979) Adaptation. In: Buckner R.C. and Bush L.P. (eds.) *Agronomy monograph no. 20: Tall Fescue:* 9-30. American Society of Agronomy, Madison, USA.

Cougnon M., Baert J., Van Waes C. and Reheul D. (2013) Performance and quality of tall fescue (*Festuca arundinacea* Schreb.) and perennial ryegrass (*Lolium perenne* L.) and mixtures of both species grown with or without white clover (*Trifolium repens* L.) under cutting management. *Grass and Forage Science* 69(4), 666-677.

Easton H.S., Lee C.K. and Fitzgerald R.D. (1994)Tall Fescue in Australia and New Zealand. *New Zealand Journal of Agricultural Research* 32, 1-6.

Frame J. (1992) *Improved grassland management*. Farming Press, Ipswich, UK.

Gilbert W.B. and Chamblee D.S. (1965) Effect of submersion in water on tall fescue, orchardgrass and ladino clover. *Agronomy Journal* 57, 502-504.

Graiss W., Krautzer B. and Potsch E.M., 2011. Suitability of alternative grass species for grassland management in Austria under changing climatic conditions. *Grassland Science in Europe* 16, 440-442.

Haquin F. (2012) Semences fourragères, tableaux comparatifs. *Semences et Progrès* 154, 12-83.

IPCC (2018) *Climate change (2018) Working group II: Impacts, Adaptation and Vulnerability*. Cambridge, UK.

Luten W. and Remmelink G.J. (1984) *Intake of perennial ryegrass, tall fescue and Italian ryegrass by dairy cattle*. Proefstation voor Rundveehouderij, Lelystad, the Netherlands.

R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available at: https://www.R-project.org/

At grazing, the nutritive value of grass offered to the dairy cow is like a 'natural' total mixed ration

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Abstract

At grazing, the nutritive value of grass offered is, together with intake, the key component of feed supply, which is an important driver of animal performance. A large database of 1052 samples of grass offered at grazing to dairy cows has been created to describe the chemical composition and nutritive value and to examine some factors of variation along the year. The samples were collected during multi-years experiments (1995-2019) at the INRAE Le Pin experimental farm (Normandy). The database contains chemical composition including dry matter (DM), organic matter (OM), crude protein (CP), fibre contents (CF, NDF-ADF) and pepsin-cellulase digestibility. The INRA 2018 equations have been used to calculate the OM digestibility (OMd), the fill unit for dairy cows (UEL), the net energy for milk (UFL) and protein digestible in the intestine (PDI) values. On average, the OM, CP and CF, NDF and ADF contents are 895±23.7, 180±32.6, 225±25.5, 513±26.3 and 257±23.9 g kg^{-1} DM, respectively. The variation of grass composition and feed value is partly explained by season and pre-grazing pasture characteristics. With a UFL and PDI average value of 0.95±0.08 and 99±8.1 g kg^{-1} DM, well managed grazed grass has similar composition to a total mixed ration and is self-sufficient in feeding dairy cows.

Keywords: grazing, grass quality, chemical composition, nutritive value

Introduction

Grass grazed is the natural and cheapest ration for ruminants. Even for dairy cows, grazed grass can be the only feed in the ration. In this situation, the chemical composition of offered grass has a large influence on feed supplies. In combination with the intake level, the nutritive value of grass will significantly influence animal performance. Better knowledge of grass quality offered to dairy cows and the main factors influencing variation can help to improve our understanding of animal performance, and to define the ideal forage supplement or the concentrate composition according to the milk potential of the dairy cow. In the context of grazing dairy cows, the regular grass sampling realized before grazing on the INRAE Le Pin experimental farm during 25 years provides a unique opportunity to describe the chemical composition and nutritive value of grass offered and to examine some factors of variation during the year.

Materials and methods

Between 1995 and 2019, three pluri-annual experiments (Dall Orsoletta *et al.*, 2019; Delaby *et al.*, 2003, 2009) were conducted at the INRAE experimental farm of Le Pin-au-Haras in Normandy (48.44 N 0.09 E). These experiments are characterised by grass-based dairy systems and a long grazing season between mid-March and mid-November. The grazing area, placed on drained clay-loam soils rich in organic matter (6 to 8%) is composed of permanent and old sown pastures, with varying quantities of white clover. In sown pastures, perennial ryegrass (PRG) is the largely dominant grass species, and in permanent pastures, also with other grasses (expressed in frequency) like rough meadow grass (*Poa trivialis* 20%), fine bent (*Agrostis stolonifera* 15%), Yorkshire fog (*Holcus lanatus* 15%), foxtail (*Alopecurus pratensis* 10%) and timothy (*Phleum pratense* 10%) and white clover (*Trifolium repens* 15%). The annual level of mineral N applied ranges from medium to high (120 to 280 kg ha^{-1}), applied in 4 to 5 applications. The annual stocking rate on the grazing platform varies between experiments and years (1.75 to 3.00 cows ha^{-1}).

Due to poor grass growth at certain times, higher stocking rates required higher supplement-use levels of grass or maize silage to extend the grazing season. The simplified rotational grazing system described by Hoden *et al.* (1991) and the management rules associated were applied. The main characteristics of this grazing system is the long residence time in large paddocks, varying from 8 to 12 days according to the pre- and post-grazing height (PreGH – PostGH) and the milk yield evolution during the 10-day paddock grazing process.

The day before each grazing event, a standard protocol (Delaby and Peyraud, 2003) is applied on every paddock including 2 to 4 strips per ha cut at 5 cm with a motorscythe, the heights pre- and post-cutting measured with an electronic plate meter and a handful of grass taken, bulked per paddock for drying in a ventilated oven (60 °C – 48 h), ground (screen 0.8 mm) and sent to the laboratory (Labocea, Ploufragan, France) for chemical analysis. On each grass sample, the ash, crude protein (CP – N × 6.25 – method Dumas) and crude fibre (CF) content and the pepsin-cellulase digestibility (dCS – Aufrère *et al.*, 2007) have been analysed. The neutral and acid detergent fibre (NDF – ADF) content as well as the organic matter digestibility (OMd) have been calculated according to dedicated equations published by INRA (2018). Finally, the net energy value for milk (expressed in UFL with 1 UFL = 1,760 kcal), the protein digestible in intestine (PDI in g kg^{-1} DM), the rumen protein balance (RPB in g kg^{-1} DM) and the fill unit for lactation (UEL kg^{-1} DM with 1 UEL = 140 g DM intake kg^{-1} metabolic BW) of the grass samples have been calculated as proposed by INRA (2018).

The available database contains 1052 lines, divided by season into 261, 470 and 321 samples for spring (15/3 to 31/5), summer (1/6 to 31/8) and autumn (1/09 to 30/11), respectively. The data have been analysed with the SAS GLM procedure (2013) including the effects of year, the experimental treatment within year and season.

Results and discussion

On average, with a PreGH of 11.2 (±2.4 cm) and a biomass of 1,890 (±712) kg DM ha^{-1}, the grass offered to the dairy cows had an excellent nutritive value, with on average 73.8%; 0.95; 99 g and 30 g for OMd, UFL, PDI and RPB values, respectively. These values are consistent with the reference values published in the feed value tables of INRA (2018). The influence of the season on the main characteristics of the grass offered is described in Table 1, which also specifies the 5 and 95 percentiles for each characteristic. The season effect is always highly significant, sometimes with little biological relevance due to the rarely limiting nutritive values to the feeding of dairy cows.

The summer season is characterized by the lowest CP content (164 g kg^{-1} DM) and the highest fibre content (CF, NDF or ADF), whereas the differences between spring and autumn are less important. This summer effect is often observed and is a consequence of the high temperatures combined with less rain and moisture availability resulting in an acceleration of the ageing process. Consequently, the PDI and RPB values decrease (-6 and -26 g kg^{-1} DM) in summer (96 and 16 g kg^{-1} DM) compared to spring and autumn (102 and 42 g kg^{-1} DM). The highest OM digestibility is observed in spring and is 5 percentage points higher than in summer and autumn. This has a direct effect on the UFL value (1.02 kg^{-1} DM in spring) which declines in summer and autumn, with 0.94 and 0.91 UFL kg^{-1} DM respectively. The UEL values change little between seasons and stay on average below 1.00 UEL kg^{-1} DM, thereby supporting excellent voluntary intake. Among the many advantages associated with legumes (Lüscher *et al.*, 2014), their summer production and high nutritive value would limit the decline in summer nutritive value observed on pasture.

Globally, the energy/fill and protein/energy ratios classically used to evaluate dairy cow rations are respectively higher than 0.95 UFL/UEL and 100 g PDI/UFL. This confirms that, when following recommended grazing management rules (age of regrowth, pre and postGH – Delaby and Horan, 2017), and the given conditions, grazed grass is one of the only forages which is sufficient in energy and protein to feed dairy cows. Unsurprisingly, with 14 to 16 kg of DM daily ingested, the nutrient supply in energy or protein is sufficient to produce between 22 and 25 kg of milk at herd level from a grass-only diet (Delaby *et al.*, 2003).

Table 1. Main characteristics of grass nutritive value according to the season (least square means, 5% and 95% centiles between brackets – g kg^{-1} DM).

Season	Spring	Summer	Autumn	*P*
DM (% fresh matter)	19.7 [13.7-25.4]	23.9 [15.4-35.4]	21.3 [13.7-35.7]	<0.0001
OM	891 [841-921]	906 [889-921]	883 [821-916]	<0.0001
CP	192 [135-244]	164 [123-210]	195 [143-244]	<0.0001
CF	211 [168-252]	241 [212-271]	215 [178-247]	<0.0001
NDF	497 [454-536]	529 [500-564]	502 [464-536]	<0.0001
ADF	246 [204-283]	271 [241-300]	248 [211-279]	<0.0001
OMd (%)	77.5 [70.4-83.5]	72.7[65.8-78.9]	72.1 [63.8-79.7]	<0.0001
UFL (kg^{-1} DM)	1.02 [0.88-1.13]	0.94 [0.83-1.05]	0.91 [0.77-1.05]	<0.0001
PDI	102 [89-114]	96 [84-108]	102 [89-116]	<0.0001
RPB	40 [-7-85]	16 [-18-54]	44[5-85]	<0.0001
UEL (kg^{-1} DM)	0.95 [0.91-1.01]	0.98 [0.94-1.03]	0.97 [0.92-1.04]	<0.0001

Conclusions

As stated in the title, the nutritive value of grass offered to grazing dairy cows is able to be like a 'natural' Total Mixed Ration (TMR). The main challenge is to manage grazing to produce highly digestible and ingestible grass and to obtain high levels of individual grass intake along the grazing season.

References

Aufrère J., Baumont R., Delaby L., Peccatte J.R., Andrieu J., Andrieu J.P. and Dulphy J.P. (2007) Prévision de la digestibilité des fourrages par la méthode pepsine-cellulase. Le point sur les équations proposées. *INRA Productions Animales* 20(2), 129-136.

Dall-Orsoletta A.C., Leurent-Colette S., Launay F., Ribeiro-Filho H.M.N. and Delaby L (2019) A quantitative description of the effect of breed, first calving age and feeding strategy on dairy systems enteric methane emission. *Livestock Science* 224, 87-95.

Delaby L. and Peyraud J.L. (2003) The effect of two contrasting grazing management and level of concentrate supplementation on the performance of grazing dairy cows. *Animal Research* 52, 437-460.

Delaby L. and Horan B. (2017) Improved efficiency in temperate grass based dairy systems. 54th Annual Meeting Brazilian Society Animal Science, Foz do Iguaçu, Brazil.

Delaby L., Peyraud J.L. and Delagarde R. (2003) Faut-il complémenter les vaches laitières au pâturage ? *INRA Productions Animales* 16(3), 183-195.

Delaby L., Faverdin P., Michel G., Disenhaus C. and Peyraud J.L. (2009) Effect of different feeding strategies on lactation performance of Holstein and Normande dairy cows. *Animal* 3(6), 891-905.

Hoden A., Peyraud J.L., Muller A., Delaby L. and Faverdin P. (1991) Simplified rotational grazing management of dairy cows: effects of rates of stocking and concentrate. *Journal of Agricultural Science* 116, 417-428.

INRA (2018) *INRA feeding system for ruminants*. Wageningen Academic Publishers, Wageningen, the Netherlands, 640pp.

Lüscher A., Mueller-Harvey I., Soussana J.F., Rees R.M. and Peyraud J.L. (2014) Potential of legume-based grassland–livestock systems in Europe: a review. *Grass and Forage Science* 69, 206-228.

SAS Institute Inc. (2013). *Base SAS 9.4®. SAS User's Guide: Statistics*. SAS Institute Inc., Cary, NC, USA.

Milk production and grazing behaviour responses of dairy cows to partial mixed ration supplementation

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Abstract

Supplementing grazing dairy cows with a partial mixed ration (PMR) is widely used on farm. The dose-response of grazing cows to PMR is not well known. The aim of this study was to compare the effects of three supplementation levels (0, 4 and 8 kg dry matter (DM) d^{-1} of PMR (85:15 maize silage/soyabean meal ratio, DM basis) on milk production and behaviour of cows grazing on perennial ryegrass pastures in spring. Pasture allowance was specific to each treatment to achieve the same post-grazing sward height between treatments. Twenty-four mid-lactation Holstein cows were used in a complete and balanced 3×3 Latin square design with three 21-day periods. Cows ate less PMR than offered (15-25% refusals). Herbage N concentration was low (119 g kg^{-1} DM). Milk production increased linearly by 0.52 kg kg^{-1} DM of PMR intake. Milk fat concentration was reduced whereas milk protein concentration was not affected by increasing the supplementation level. Daily grazing time decreased linearly by 9 min kg^{-1} DM of PMR intake, suggesting a low substitution rate between pasture and PMR, and explaining the great milk production response to PMR. The reduced grazing activity was mainly noticeable after the evening milking and the PMR distribution.

Keywords: dairy cow, grazing, supplementation, maize silage, behaviour

Introduction

In many grazing systems of Western Europe, dairy cows are often supplemented with maize silage or with a partial mixed ration (PMR) based on maize silage and oilseed meal. The economic interests of such a supplementation partly depend on cow biological responses, particularly in spring with good-quality pasture and few pasture herbage shortages. Contrary to the well-known dose-response of cows to concentrate supplementation level, only a few studies have investigated the dose-response to PMR supplementation in grazing dairy cows (Miguel *et al.*, 2014; Moran and Croke, 1993; Stockdale, 1994). The milk production response to PMR is known to vary in a large range, depending on allowance and quality of the pasture and of the supplement. The objective of this study was to determine the effects of increasing PMR supplementation level on the milk production and feeding behaviour responses of dairy cows grazing perennial ryegrass swards.

Materials and methods

The experiment took place at the INRAE farm of Méjusseaume (Le Rheu, France) from April to June, 2021. Three levels of PMR supplementation (0, 4 and 8 kg DM d^{-1}, namely S0, S4 and S8, respectively) were compared on 24 multiparous Holstein dairy cows according to a 3×3 Latin square design, with three 21-day periods. The PMR was composed of 85% of maize silage and 15% of soyabean meal, on a DM basis. The PMR chemical composition was 125 g of CP and 429 g of NDF kg^{-1} DM. Mean pre-experimental characteristics of the cows were: 626 kg of body weight, 104 days in milk and 37 kg d^{-1} of milk production.

Cows strip-grazed adjacent paddocks, with one paddock per treatment, and one new strip given once daily in the morning. Cows in S0 (control) received a fixed and medium herbage allowance of 20 kg DM d^{-1} at 4 cm above ground level. Daily offered areas in S4 and S8 were frequently adjusted to achieve similar post-grazing sward height than in S0. Herbage allowances were thus lower in S4 and S8 than in S0 due to the substitution rate and lower herbage intake. Cows grazed day and night and had access to pasture 19 h d^{-1}. Fresh drinking water was always available at grazing.

Pre and post-grazing sward heights were measured daily using an electronic plate meter (30×30 cm, 4.5 kg m^{-2}). The pre-grazing herbage mass per treatment was estimated daily from pre-grazing sward height and sward bulk density. Sward bulk density was determined 4 times per period by cutting strips with a motor scythe, allowing also chemical composition of offered herbage from oven-dried subsamples (60 °C for 48 h) to be determined.

Cows had individually access to PMR 1 h after each milking, knowing that cows stopped eating PMR long before this delay. The remaining PMR was thus considered as refusals and weighed. Milk production was measured per cow at each milking. Milk fat and protein concentrations were determined on days 18 to 21, and milk urea concentration was determined on days 19 and 21. Grazing activity pattern, meal size and daily grazing time were recorded individually from days 14 to 21 thanks to the Lifecorder Plus (Suzuken, Japan) device, based on a mono-axial accelerometer (Delagarde and Lamberton, 2015). Cow data were averaged per period and then analysed by ANOVA taking into account of the cow, period and treatment effects. Two orthogonal contrasts were used to determine the linear and quadratic effects of the PMR supplementation level.

Results and discussion

Pre-grazing herbage mass (3.8 t DM ha^{-1}), sward height (15.8 cm), and herbage crude protein (CP) (119 g kg^{-1} DM) and eutral detergent fibre (NDF) (469 g kg^{-1} DM) concentrations did not differ between treatments. The pasture herbage comprised 80% grasses, 12% clover and 8% of other species. As expected, post-grazing sward height averaged 6.5 cm and did not differ between treatments, thanks to the reduction of herbage allowance through offered area in herds receiving the supplement (allowance of 20.7, 16.5 and 14.1 kg DM d^{-1} for S0, S4 and S8, respectively).

The PMR intake averaged 3.5 and 6.1 kg DM d^{-1} in S4 and S8, respectively, due to partial refusals. The milk production increased linearly (P<0.001) from 22.2 to 25.3 kg d^{-1} with increasing PMR supplementation level, i.e. an increase of 0.52 kg of milk d^{-1} per kg DM of PMR eaten (Table 1). Milk fat concentration decreased linearly by 0.2 g kg^{-1} per each kg DM d^{-1} of PMR eaten (P<0.05), perhaps because soyabean meal in PMR allowed greater ruminal fermentations (low N herbage). Milk protein concentration was unaffected by treatment. Milk urea concentration was low and increased with increasing PMR supplementation level. The total grazing time averaged 445 min d^{-1} and decreased linearly by 9.3 min d^{-1} per each kg DM of PMR eaten (P<0.001). This reduction was mainly due to that of the night grazing time (6.7 min d^{-1} per kg DM of PMR eaten, P<0.001). The mean grazing bout duration was lower in S8 than in S0 and S4 (P<0.05). The daily number of grazing bouts was unaffected by supplementation level.

This study was characterized by a low herbage N concentration, resulting in low milk urea concentration of unsupplemented cows. It can be hypothesized that the positive milk production response to PMR originates from an increase in total DM and energy supplies, but perhaps also to the greater diet CP concentration as shown by the increase in milk urea concentration with increasing PMR supplementation level. Moreover, from the known great milk production response of dairy cows to protein-rich concentrate while grazing on low-N pastures (+1.4 to +1.9 kg of milk kg^{-1} DM eaten; Delagarde *et al.*, 1999), it can be estimated that 30-50% of the milk response to PMR in this study originates from the supply of soyabean meal *per se*. Reduction of grazing time with increasing PMR supplementation level was slightly lower than previously reported in the literature (9 vs 20 to 30 min kg^{-1} DM of supplement). This suggests a low substitution rate between herbage and PMR in this study, which is in agreement with the positive milk production response to PMR supplementation.

Table 1. Dose-response to PMR supplementation level on grazing dairy cows.[1]

Variable	Treatments			RSD	Treatment effect (P<)	Dose-response (P<)	
	S0	S4	S8			Linear	Quadratic
Milk production, kg d^{-1}	22.2^c	24.2^b	25.3^a	1.53	0.001	0.001	0.193
Milk fat concentration, g kg^{-1}	37.7^a	37.2^{ab}	36.5^b	1.70	0.055	0.017	0.792
Milk protein concentration, g kg^{-1}	29.2	29.4	29.5	0.67	0.188	0.068	0.862
Milk urea concentration, mg l^{-1}	120	140	155	19.2	0.001	0.001	0.606
Total grazing time, min d^{-1}	471^a	451^b	414^c	25.8	0.001	0.001	0.209
Day grazing time, min	251^b	264^a	255^{ab}	15.2	0.018	0.378	0.008
Night grazing time, min	220^a	187^b	158^c	18.4	0.001	0.001	0.578
Number of grazing bouts, bouts d^{-1}	6.2	5.8	6.2	0.70	0.113	0.795	0.038
Mean grazing bout duration, min	82^a	83^a	72^b	11.2	0.002	0.004	0.022

[1] S0, S4, S8: 0, 4 and 8 kg DM d^{-1} of PMR, respectively; RSD: Residual standard deviation; Means within rows having different superscript letters differ significantly.

Conclusions

At similar post-grazing sward height, supplementation with 4 and 8 kg DM d^{-1} of PMR based on maize silage and soyabean meal linearly increased the milk production of dairy cows grazing perennial ryegrass during spring, by 0.52 kg of milk kg^{-1} DM of supplement eaten. Milk fat concentration was reduced whereas milk protein concentration was not affected by increasing PMR supplementation level. Daily grazing time decreased with increasing supplementation level, mainly due to a reduction of grazing time after the evening milking, following the supplement distribution.

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References

Delagarde R. and Lamberton P. (2015) Daily grazing time of dairy cows is recorded accurately using the Lifecorder Plus device. *Applied Animal Behaviour Science* 165, 25-32.

Delagarde R., Peyraud J.L. and Delaby L. (1999) Influence of carbohydrate or protein supplementation on intake, behaviour and digestion in dairy cows strip-grazing low-nitrogen fertilized perennial ryegrass. *Annales de Zootechnie* 48, 81-96.

Miguel M.F., Ribeiro Filho H.M.N., de Andrade, A.E., Moraes Genro T. C. and Delagarde R. (2014) Pasture intake and milk production of dairy cows grazing annual ryegrass with or without corn silage supplementation. *Animal Production Science* 54, 1810-1816.

Moran J.B. and Croke D.E. (1993) Maize silage for the pasture-fed dairy cow. 5. A comparison with wheat while grazing low quality perennial pastures in the summer. *Australian Journal of Experimental Agriculture* 33, 541-549.

Stockdale C.R. (1994) Persian clover and maize silage. I. Silage as a supplement for lactating dairy cows offered herbage of different quality. *Australian Journal of Agricultural Research* 45, 1751-1765.

Effect of pre-grazing herbage mass and post-grazing sward height on steer grazing behaviour

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Abstract

Grazing behaviour determines herbage dry matter intake (DMI) and resulting animal performance, and is influenced by sward structure. The objective of this study was to determine the effects of pre-grazing herbage mass (PGHM: 1,500 or 2,500 kg DM ha^{-1}) and post-grazing sward height (PGSH: 4 or 6 cm) on steer grazing behaviour. Charolais steers (n=96, ca. 17 months of age, 515±34.6 kg) rotationally grazed *Lolium perenne*-dominant swards and were fitted with a RumiWatch noseband sensor (Itin & Hoch GmbH) for four consecutive days. Pasture was allocated such that each grazing treatment group would graze their designated PGHM to their PGSH over a 48 h allocation. Bite mass, bite rate, intake rate and associated DMI were unaffected by PGHM. Compared to PGSH-4, PGSH-6 had a greater bite mass (P=0.08), bite rate (P<0.01), intake rate (P<0.001) and DMI (P<0.001). There were PGHM × PGSH interactions (P<0.05) for rumination time, mastications, mastication rate, boli and boli per rumination bout, whereby there was no difference between 2500-6 and 1500-6, but 2500-4 was greater than 1500-4. In conclusion, a higher PGSH increased animal DMI, whereas PGHM had no impact on DMI.

Keywords: beef, defoliation, dry matter intake, ruminating behaviour, RumiWatch, sward structure

Introduction

Current grazing guidelines in temperate systems recommend grazing a pre-grazing herbage mass (PGHM) of ca. 1,500 kg DM ha^{-1} (>4 cm) to a post-grazing sward height (PGSH) of ca. 4 cm to optimize herbage production, sward nutritive value and stocking rate (Maher *et al.*, 2017). Data suggests that increasing PGSH can change animal grazing behaviour and increase dry matter intake (DMI) (Chacon and Stobbs, 1976; Doyle *et al.*, 2021) and this requires further investigation. However, there is a paucity of published research information investigating the effect of these recommended grazing guidelines, and the interactive effect of PGHM × PGSH on grazing and rumination behaviour (Amaral *et al.*, 2013), especially for beef cattle. Therefore, the objective was to determine the effect of PGHM and PGSH on steer grazing and ruminating behaviour.

Materials and methods

Suckler-bred Charolais steers (n=96) were blocked on live-weight and randomly assigned to one of twelve grazing groups. Each group was randomly assigned to a two (PGHM >4 cm: 1,500 or 2,500 kg dry matter (DM) ha^{-1}) × two (PGSH: 4 or 6 cm compressed height) factorial arrangement of treatments, with three replicate groups of 8 steers per treatment. Steers rotationally grazed *Lolium perenne*-dominant swards in their replicate grazing groups for 222 days and pastures were not mechanically topped during the grazing season. Grazing behaviour was recorded for each animal (515±34.6 kg) over four consecutive days between 12 and 30 August (day 144 to 162 of the grazing season) using the RumiWatch noseband sensor (Itin & Hoch GmbH, Liestal, Switzerland) (Norbu *et al.*, 2021). The grazing behaviour data were converted into 1 h summaries with the RumiWatch converter V.0.7.3.36 (Itin & Hoch GmbH) (Norbu *et al.*, 2021). Dry matter intake was estimated using the herbage disappearance method as described by Doyle *et al.* (2021). Pre-and post-grazing herbage mass were estimated with a rising platemeter (1000 heights ha^{-1}). During this four-day measurement period, pasture was allocated to each grazing treatment group on a 48 h basis such that each grazing group would graze their designated PGHM to their PGSH, twice. Average 48 h area allocation per group was 0.052, 0.081, 0.025 and 0.040 ha for 1500-4, 1500-6, 2500-4 and 2500-6, respectively. Pre-grazing stem mass was calculated as described by Doyle *et al.* (2021). Grazing behaviour data for each of the two consecutive 24 h measurement periods were statistically analysed using the MIXED procedure of SAS with grazing group as the experimental unit. The model contained fixed effects for PGHM, PGSH and their interactions within each day (first and second 24 h period). Differences between means were tested for significance using the PDIFF statement and adjusted by Tukey, as appropriate.

Results and discussion

There was no PGHM × PGSH interaction or effect of PGHM on DMI, bite mass and intake rate. Compared to PGSH-6, PGSH-4 had a lower DMI (P<0.001; 5.3 vs 7.2 kg DM day^{-1}). Despite the similar DMI, bite rate was greater (P<0.05) and grazing bout duration was longer (P<0.05) for PGHM-2500 than PGHM-1500 during the first 24 h of the 48 h allocation (Table 1). Furthermore, 2500-6 had a greater (P<0.05) number of grazing bites than 1500-4, 1500-6 and 2500-4 (Table 1). However, in the second 24 h, eating time was shorter (P<0.05) and there was a tendency for bite rate to be lower (P=0.10) for PGHM-2500 than PGHM-1500. Furthermore, the number of grazing bites was lower for 2500-4, 2500-6 and 1500-4 than 1500-6. Differences in grazing behaviours in the second 24 h may be attributed to a greater stem mass (P<0.001; 656 vs 320 kg DM ha^{-1}) in the lower grazing horizons, likely associated with the longer regrowth interval for PGHM-2500 compared to PGHM-1500, as steers try to avoid stem and select leaves (Amaral *et al.*, 2013).

Table 1. Effect of pre-grazing herbage mass (1,500 or 2,500 kg dry matter (DM) ha^{-1}) and post-grazing sward height (4 or 6 cm) on grazing behaviour during the first and second 24 h and ruminating behaviour over the total 48 h allocation.[1]

PGHM	1,500		2,500		SEM	Significance		
PGSH	4	6	4	6		PGHM	PGSH	PGHM×PGSH
First 24 hours								
Eating time (min d^{-1})[2]	691^{a}	623^{a}	611^{a}	675^{a}	19.7	NS	NS	*
Grazing bouts (n d^{-1})	8.3	8.3	6.3	6.5	0.49	**	NS	NS
Grazing bout duration (min bout^{-1})	89.8	80.0	100.3	108.6	6.21	*	NS	NS
Grazing bites (n d^{-1})	27,900^{b}	27,652^{b}	28,225^{a,b}	33,144^{a}	1,151.2	*	0.08	*
Bite rate (bites min^{-1})[3]	47.1	52.9	54.6	57.4	2.52	*	NS	NS
Second 24 hours								
Eating time (min d^{-1})[2]	492	542	446	466	22.4	*	NS	NS
Grazing bouts (n d^{-1})	8.1	7.2	7.1	7.4	0.41	NS	NS	NS
Grazing bout duration (min bout^{-1})	68.4	80.5	71.2	71.5	6.05	NS	NS	NS
Grazing bites (n d^{-1})	13,971^{b}	22,621^{a}	12,814^{b}	16,009^{b}	1,012.9	**	***	*
Bite rate (bites min^{-1})[3]	34.9	50.0	34.6	42.3	2.14	0.1	***	NS
Ruminating behaviour over 48-hours								
Ruminating time (min d^{-1})	314^{b}	415^{a}	437^{a}	458^{a}	13.7	***	**	*
Ruminating bouts (n d^{-1})	12.5	13.1	13.7	13.6	0.31	*	NS	NS
Ruminating bout duration (min bout^{-1})	27	33	33	36	1.1	**	**	NS
Ruminating mastications (n d^{-1})	18,665^{b}	27,136^{a}	27,417^{a}	30,240^{a}	971.9	***	***	*
Ruminating mastication rate (chews min^{-1})	60^{c}	65^{a}	63^{b}	66^{a}	0.6	**	***	*
Ruminating boli (n d^{-1})	347^{b}	460^{a}	499^{a}	517^{a}	14.8	***	**	*

[1] SEM = standard error of the mean for PGHM × PGSH; Means within a row with different superscript letters differ (P<0.05); NS = non-significant, * P<0.05, ** P<0.01, *** P<0.001.

[2] Eating time includes eat up + eat down time on the RumiWatch system..

[3] Bite rate is calculated as (number of grazing bites/eat down time).

The differences in DMI between PGSH-4 and PGSH-6 can be largely attributed to a lower bite mass (P=0.08; 0.26 vs 0.29 g DM) and intake rate (P<0.001; 11.3 vs 14.9 g min^{-1}) observed for PGSH-4. Both of these parameters decrease linearly with sward depletion height due to a lower bite depth (Chacon and Stobbs, 1976). Furthermore, the lower herbage DMI for PGSH-4 compared to PGSH-6 was more evident during the second 24 h of the 48 h allocation (Table 1) due to the lower bite rate for PGSH-4 (P<0.001) and lower number of grazing bites for 1500-4, 2500-4 and 2500-6 than 1500-6, implying that steers had little desire to select out small quantities of herbage (Chacon and Stobbs, 1976). Under the circumstances of this rotational stocking experiment, steer grazing behaviour at the end (PGSH) rather than the beginning (PGHM) of the graze-down process had a larger impact on steer DMI.

Over the 48 h allocation, there were PGHM × PGSH interactions (P<0.05) for rumination time, mastications (chews), mastication rate, boli and boli per rumination bout, whereby there was no difference between 2500-6 and 1500-6, but 2500-4 was greater than 1500-4 (Table 1).

Conclusions

Under the conditions of this study, DMI did not differ between PGHM treatments, but was lower for PGSH-4 than PGSH-6, due to a lower bite mass, intake rate and bite rate.

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References

Amaral M.F., Mezzalira J.C., Bremm C., Da Trindade J.K., Gibb M.J., Suñe R.W.M. and Carvalho, P.C.F. (2013) Sward structure management for a maximum short-term intake rate in annual ryegrass. *Grass and Forage Science* 68, 271-277.

Chacon E. and Stobbs T.H. (1976) Influence of progressive defoliation of a grass sward on the eating behaviour of cattle. *Australian Journal of Agricultural Research* 27, 709-727.

Doyle P.R., McGee M., Moloney A.P., Kelly A.K. and O'Riordan E.G. (2021) Effect of post-grazing sward height, sire genotype and indoor finishing diet on steer intake, growth and production in grass-based suckler weanling-to-beef systems. *Animals* 11, 2623.

Maher J. (2017) Lessons learned from the beef finalists of the Teagasc Grass10 Farmer of the Year Competition. In: O'Riordan E.G. Kelly P. and Ferguson L. (eds.) *Planning for Healthy Profits.* Teagasc, National Beef Conference 2017. Teagasc, Tullamore, Ireland, pp. 15-20.

Norbu N., Alvarez-Hess P., Leury B., Wright M., Douglas M., Moate P., Williams S., Marett L., Garner J. and Wales W. (2021) Assessment of RumiWatch noseband sensors for the quantification of ingestive behaviors of dairy cows at grazing or fed in stalls. *Animal Feed Science and Technology* 280, 115076.

How spatial temporal grazing management affects productive results of grassland-based cattle systems in Uruguay?

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Abstract

Cow-calf grazing farm systems are the main economic and social activity on Campos grassland. However, lack of grazing intensity management (overgrazing) has limited farm production, income and sustainability for decades. The aim of this study was to define farm grazing models and analyse the relationship with evolution of systems state variables (forage height, FH; body condition score, BCS; and kg of weaned calf per breeding cow), biological processes (cow forage intake and metabolic hormones), and beef meat production per hectare. A case study of three farms was carried out during two years. A grazing paddock usage pattern based on FH, cow BCS and physiological status was defined as a spatial-temporal grazing manager farm, and produced 213±4 kg of calf per breeding cow and 132±2 kg of beef meat per hectare, while Traditional Non Grazing Manager farms produced 117±11 and 83±7 kg respectively. Higher levels of cow energy intake (+47%, $P<0.05$) and cows serum insulin-like growth factor (IGF-1) (+79%, $P<0.05$) associated with higher levels of FH evolution (+90%, 6.2±1.5 cm) contributed to explaining outcome differences. Farm agroecological intensification included improvement of levels of seasonal FH, but spatial-temporal management was key to couple energy intake, efficiency and beef production of grassland systems.

Keywords: Campos grassland, grazing management, cow-calf system

Introduction

Beef grazing farms on Campos grassland (Allen *et al.*, 2011) are the main economic and social agricultural activity in Río de la Plata. However, decades of low reproductive, productive, cow energy intake and body condition score (BCS) results have been mainly explained by lack of grazing management (overgrazing). Farm systems (FS) with applied spatial temporal grazing management have improved meat production per ha by 30% compared to non-manager (Paparamborda, 2017). A co-innovation approach study (Dogliotti *et al.*, 2014) demonstrated that FS redesign based on spatial temporal management improved beef meat production by 20% per hectare without affecting the environment (Ruggia *et al.*, 2021). However, the levels of state variables and biological processes associated with different grazing management at grassland cow-calf systems have not been reported. The aim of this study was to quantify and explain the relationship between FS grazing and cow-calf practices management and the resulting levels of forage height (FH), forage allowance (FA), cow liveweight (LW), BCS, forage dry matter intake (DMI), metabolic hormones response (IGF-1), and the production of calves (in kg) per breeding cow and beef meat per surface during two consecutive years on Campos grassland in Uruguay. Our hypothesis was that spatial temporal grazing management, with the adoption of main cow-calf practices to control cow energy intake and balance during a trajectory in time, is associated with higher seasonal levels of FH, FA, cow BCS, DMI and anabolic hormones response, kg production of calves per breeding cow and beef meat per surface.

Materials and methods

A case study of three cow-calf FS (FS1, FS2, FS3) was carried out during two breeding seasons (BS) during 2017-18 (BS1) and 2018-19 (BS2) in Rocha, Uruguay. Campos grassland (CG) was the main forage resource for all farms, with 18% of pasture improvement (PI) used for heifers in FS3 and 4% in FS1 and FS2. A Spatial Temporal grazing management scheme was constructed per FS according to monthly paddocks usage pattern for the breeding cows. Also, a cow-calf adoption practices index was calculated (IPC scale 1 to 100) based on strategic (e.g. breeding and weaning season control), decision aid (e.g. pregnancy diagnosis, cow BCS score) and tactics (e.g. temporary weaning and/or improvement of energy intake 'flushing') practices (Paparamborda, 2017). Two different FS management models were defined: (1) a spatial-temporal grazing manager (STM) associated to FS3, who assigned different land units (up to 4 paddocks) to different breeding cow groups according to cow BCS, physiological status and FH and season. FS3 also had an IPC of 85 form 100 points; and (2) a traditional non manager model (TNM) associated to FS1 and FS2, who made few to no decisions on animal assignment to land units during the study, and also resulted in low to moderate level of cow-calf practices adoption with an IPC of 35 from 100 points. The FH (Do Carmo *et al.*, 2020) and FA (Sollenberger *et al.*, 2005) was estimated for each paddock/s assigned to breeding cows during the study. Cows BCS (Vizcarra *et al.*, 1986), LW were measured, cow DMI was estimated (CSIRO, 1990) for spring, summer, autumn and winter during both BS and cow serum IGF-1 in same periods during BS1. Calves LW was registered in November and at weaning during both BS. Pregnancy rate (PR), kilograms of weaned calf per breeding cow and beef meat per hectare was calculated for each FS and agriculture year. Relationship between FS, and FH and FA evolution was analysed using general linear models considering date as a replica. Effect of FS, date and their interactions on BCS, LW, and serum concentrations of IGF-1was analysed via mixed models with date as the repeated-measure. All data were analysed using procedures of the SAS Systems program (SAS Institute Inc., Cary, NC, USA).

Results and discussion

The FS1 (TNM) resulted in CG FH seasonal average of 3.4±1.7, 3.3±1.4, 3.5±1.8, 3.5±1.7 cm and BCS 4.1±0.5, 3.9±0.4, 4.2±0.6, 4.5±0.5 for spring, summer, autumn and beginning of winter respectively. FS2 (TNM) FH levels were 3.7±1.6, 3.4±1.3, 5.1±3.0 and 2.0±0.7 cm and cow BCS 3.8±0.6, 4.3±0.5, 4.2±0.5, 3.8±0.6 units for spring, summer, autumn and beginning of winter respectively. FS3 (STM) resulted in CG FH levels of 6.1±2.0, 7.0±1.3, 6.2±2.1 and 4.9±0.7 cm and cow BCS of 4.8±0.5, 4.7±0.5, 5.0±0.4, 5.4±0.5 units for spring, summer, autumn and winter respectively. Complementary, two-year average results for each FS are presented in Table 1. FS3 PR was 100% for both BS, produced 213 kg of weaned calf per breeding cow and 132 kg of beef meat per hectare, 90 and 58% more kg of weaned calf per breeding cow and beef meat per hectare, respectively, compared to FS1 and FS2. These differences could be explained in part by the resulting levels of biological processes: overall 47% higher DMI and 79% (72 $\mu g\ ml^{-1}$) higher IGF-1 ($P<0.05$) at calving in FS3, higher levels of energy intake and efficiency to produce calves (Claramunt *et al.* 2018). Also, despite a reduction of 33% of FH and FA during BS2, reproductive and productive results remained at similar levels in FS3, which could probably be explained by a process known as metabolic memory.

This case study allowed to associate FS grazing management schemes with the seasonal measurable levels of forage and animal state variables and processes results (energy intake and efficiency in utilization) on real farms, and to contribute to understanding part of the associations within the great complexity of heterogeneous FS.

Table 1. Farm systems forage height and allowance, cow body condition score and live weight (2017-2019).[1]

	FH[3] (cms)	FA[4] (kgs DM kg LW^{-1})	Cows BSC[5] (1-8)	Cows LW[6] (kg)
BS1[2]				
FS1	3.4±1.2a	3.0±0.5a	4.0±0.5a	404±42a
FS2	3.3±1.0a	2.6±0.5a	3.9±0.5a	403±44a
FS3	7.5±0.6b	6.1±0.1b	4.9±0.5b	512±39b
BS2[2]				
FS1	3.2±0.2a	2.6±0.2a	4.2±0.5a	404±35a
FS2	5.0±2.8b	4.8±3.5b	4.2±0.5a	440±37a
FS3	5.0±1.1b	4.4±0.8b	4.9±0.5b	539±33b

[1] Different letters within columns when statistically different (P<0.05).

[2] Breeding season: period from calving to weaning at cow-calf systems. BS1: 2017-2018 and BS2: 2018-2019.

[3] Forage height as the average of 100 measurements per paddock assigned to breeding cows (Do Carmo *et al.* 2020).

[4] Forage allowance as the quotient between kg of dry matter (DM) and kg of animal live weight (LW).

[5] Cow body condition score, indicator of body fat reserve in a scale from 1-8 (Vizcarra *et al.* 1986).

[6] Cow unfastened live weight in kg.

Conclusions

Farm spatial temporal grazing management based on cows BCS and FH data to assign animals to land units during two years, was associated to improvement of seasonal levels of FH (+90%), cow BCS (+1 unit) and energy intake (+47%) and efficiency (IGF-1 +79%), that resulted in 213 kilograms of weaned calf per breeding cow (+91%) and 132 kg of beef meat per hectare (+58%). This productive level can contribute to achieve a more sustainable and resilient grazing cow-calf system at Campos Grassland ecosystem.

References

Allen V.G., Batello C., Berretta E.J., Hodgson J., Kothmann M., Li X, McIvor J., Milne J., Morris C., Peeters A. and Sanderson M. (2011) An international terminology for grazing lands and grazing animals. *Grass and Forage Science* 66, 2-28.

Claramunt M., Fernández-Foren A. and Soca P. (2018) Effect of herbage allowance on productive and reproductive responses of primiparous beef cows grazing on Campos grassland. *Animal Production Science* 58(9), 1615-1624.

Do Carmo M., Cardozo G., Mecatti F., Soca P. and Hirata M. (2020) Number of samples for accurate visual estimation of mean herbage mass in Campos grasslands. *Agronomy Journal* 112 (4), 2734-2740.

Dogliotti S., García M.C., Peluffo S., Dieste J.P., Pedemonte A.J., Bacigalupe G.F., Scarlato M., Alliaume F., Alvarez J., Chiappe M. and Rossing W.A.H. (2014) Co-innovation of family farm systems: A systems approach to sustainable agriculture. *Agricultural Systems* 126, 76-86.

Paparamborda, I. (2017) *¿Qué nos dicen las prácticas de gestión del pastoreo en los predios ganaderos familiares sobre su funcionamiento y resultado productivo*? Tesis de Maestría, Facultad de Agronomía, Universidad de la República, Montevideo, Uruguay.

Ruggia A., Dogliotti S., Aguerre V., Albicette M.M., Albin A., Blumetto O. and Rossing W.A.H. (2021) The application of ecologically intensive principles to the systemic redesign of livestock farms on native grasslands: A case of co-innovation in Rocha, Uruguay. *Agricultural Systems*, *191*, 103148.

Sollenberger L., Moore J., Allen V. and Pedreira C.G. (2005) Reporting forage allowance in grazing experiments. *Crop Science* 45(3), 896-900.

Vizcarra J., Ibañez W. and Orcasberro R. (1986) Repetibilidad y reproductibilidad de dos escalas para estimar la condición corporal de vacas Hereford. *Investigaciones Agronomicas* 86-87, 45-47.

Milk solids and fatty acid composition during transition from summer to winter diets in relation to grazing

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Abstract

Changes in milk solids and fatty acid (FA) composition in milk from 90 farms in the Netherlands were investigated over a two-year period at 17 occasions. Relationships between FA composition and fat and protein content were analysed in view of diets and season. Tank milk samples were taken and the milk FA profiles were measured with gas chromatography. Samples were categorized into four groups: spring, summer, autumn and winter, to investigate relationships between (classes of) FA and milk fat content within and among the seasons. Data were recorded on diet composition and hours cows spent at pasture prior to each sampling. From June to September, most cows had access to pastures and from November to March, all cows were housed and only conserved forage was available. Transition periods (spring and autumn) were intermediate. The results showed a negative relationship between poly-unsaturated fatty acids (PUFA) and milk fat content across the four seasons; comparable relationships were found for conjugated linoleic acids (CLA) and n-3 FA versus milk fat. Within the summer period, variability in FA contents was larger than during winter. Milk protein and fat contents as well as fatty acid profiles were significantly altered when cows changed from grazing fresh forage at pasture to conserved forages in autumn and vice versa in spring. Cows at pasture generally had increased levels of PUFA, CLA and n-3 FA compared with cows fed conserved forage. Although access to grass does not ensure grazed herbage intake, as this depends on other feed provided to the animals and to herbage allowance at pasture, this study showed that grazing can be an important tool in meeting the needs of the value chain and consumers.

Keywords: milk fat, grazing, cow diet, farmer's milk price

Introduction

For a farmer, it is of interest how milk composition can be influenced by feeding, as the payment is partly based on milk solids. In the Netherlands, farmers producing so-called 'pasture-derived milk' should allow their cows to graze for at least 120 days per year for at least 6 h per day; they receive a premium price from dairy companies. Kelly *et al.* (1998) and Elgersma *et al.* (2004) showed that milk composition changes when cows switch from a silage-based diet to a fresh grass-based diet and back. Because such changes in the diet of cows can occur very rapidly, milk composition can change markedly even on a weekly basis as was shown for Dutch milk (Heck *et al.*, 2009). Various Dutch studies with raw milk were carried out with a limited number of samples and/or relatively few farms (Capuano *et al.*, 2014), and/or over a limited period of time (Liu *et al.*, 2020). The objectives of this study were therefore: (1) to compare cow milk composition and fatty acid (FA) profile within a large group of farms during various years and seasons in relation to feeding; and (2) to investigate relationships between poly-unsaturated fatty acids (PUFA) and milk fat content across the seasons.

Materials and methods

Tank milk samples were collected from 90 different farms in the North of the Netherlands, on 17 dates over a two-year period. The sampling scheme covered the summer period when most cows would have access to pasture (June – September), the autumn transition period to winter feeding (October), the winter period when all cows are housed and no fresh grass but only conserved forage is available (November – March), the spring transition period to summer (April), the second summer period (May – September) and the second autumn transition period to winter feeding (October). Data on hours that cows spent at pasture and diet composition during the days prior to each milk sampling were collected by means of interviews with farmers and questionnaires. Milk samples for milk solids composition and fatty acid analysis were collected when full milk tanks were emptied and analysed as described in Heck *et al.* (2009). Here, milk fat and protein contents and proportions of PUFA, n-3 FA and conjugated linoleic acids (CLA) (C18:2c9, t11) in milk fat are reported. To determine the presence of seasonal variation in a milk component, the month of sampling was tested as a fixed effect in a general linear model using Genstat. Samples were also categorized into four groups: spring (1 April sampling date), summer (8 dates in May-August), autumn (6 dates in September-October) and winter (2 dates in November-January) to investigate relations between PUFA and milk fat content within and among the seasons.

Results and discussion

Out of 90 farms, 78 had cows still grazing on 20 October of year 2 due to favourable grass growth conditions and relatively dry autumn weather, as opposed to 53 farms in year 1. Milk fat and protein concentrations, averaged across all farms, fluctuated from 4.05 to 4.65 g 100 g^{-1} milk for fat and 3.34 to 3.65 g 100 g^{-1} milk for protein during the measurement period (Figure 1). Protein content was lower in spring and summer (April – September) than during winter. In both years, fat content was lower (P<0.01) between May and August (range: 4.05-4.15 g 100 g^{-1} fat) than during the transition and indoor periods, i.e. 20 October to 20 April (range: 4.38-4.65 g 100 g^{-1} fat). During the measurement period, contents of PUFA, CLA and n-3 FA in milk fat ranged on average from 2.3 to 3.1, 0.4 to 1.0, and 0.6 to 0.9 g 100 g^{-1} FA, respectively. In summer, contents of PUFA and CLA were higher (P<0.01) than in winter.

There was a negative relationship between PUFA and milk fat content across the four seasons (Figure 2); comparable relationships were found for CLA and n-3 FA versus milk fat (not shown). Within the summer cluster, variability in FA contents was larger than in winter. The four points with PUFA >3 g 100 g^{-1} FA originated from year 1 when 81, 83, 85 and 85 farms practised grazing, and the four points with PUFA <3 g 100 g^{-1} FA from year 2 when 74, 79, 82 and 82 farms practised grazing at the time of sampling. The four autumn samples with fat content <4.35 g 100 g^{-1} are from samplings in September until 2 October when 83, 81, 78 and 69 farms practised grazing; the two autumn points with fat content >4.35 g 100 g^{-1} and a lower PUFA content were taken later in October when 78 and 53 farms practised

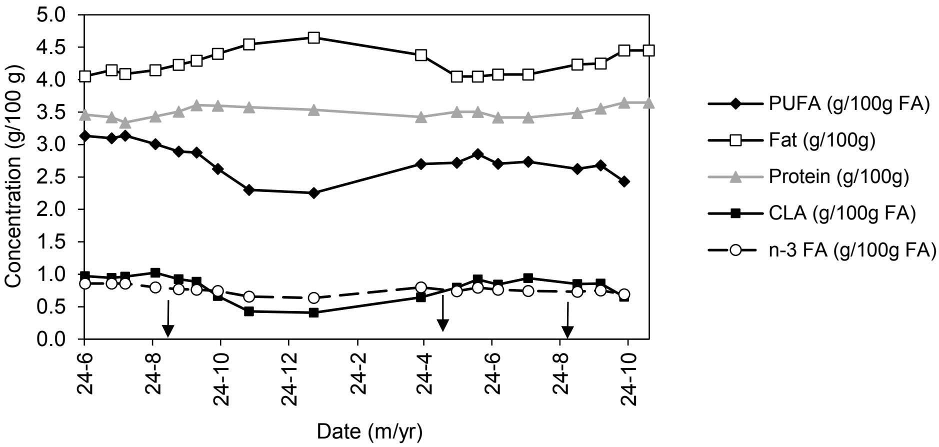

Figure 1. Contents of milk fat and protein (g 100 g^{-1} milk) and of PUFA, CLA and n-3 fatty acids (g 100 g^{-1} FA) in tank milk of 90 farms in the Netherlands, collected 17 times during 17 months in 2008 and 2009; average values are shown. Arrows at the X-axes indicate the end, the start, and the end of the summer grazing period.

grazing at the time of sampling. Pasture-fed cows generally had increased levels of PUFA, CLA and n-3 FA compared with cows fed conserved forage, in line with Elgersma (2015). In 2005, Dutch milk contained on average 3.48 g 100 g^{-1} fat and 4.38 g 100 g^{-1} protein (Heck *et al.*, 2009). This corresponds well with our data, indicating that farms were representative. In our study, the n-3 FA contents (0.6 to 0.9 g 100 g^{-1} FA) were above the Dutch December value (0.43 g 100 g^{-1} FA) and exceeded the annual mean (0.50 g 100 g^{-1} FA reported by Heck *et al.* (2009)). This may indicate that in this region during the sampling years, the farmers practised more grazing than at a national level in 2005 and would have met the requirements for pasture-derived milk premium. Although access to grass does not ensure grazed herbage intake, as this depends on other feed provided to the animals and to herbage allowance at pasture, this study showed that grazing can be an important tool in meeting the needs of the value chain and consumers.

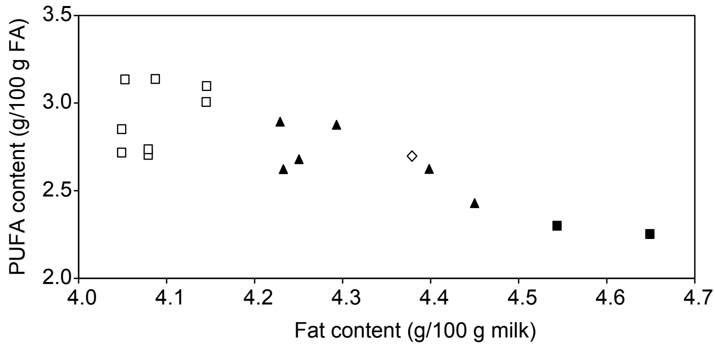

Figure 2. Relation between contents of PUFA and milk fat sampled at 17 dates in milk of 90 farms in the Netherlands, average values are shown during summer (□), autumn (▲), spring (◊) and winter (■).

References

Capuano E., van der Veer G., Boerrigter-Eenling R., Elgersma A., Rademaker J., Sterian A. and van Ruth S.M. (2014) Verification of fresh grass feeding, pasture grazing and organic farming by cows farm milk fatty acid profile. *Food Chemistry* 164, 234-241.

Elgersma A. (2015) Grazing increases the unsaturated fatty acid concentration of milk from grass-fed cows: A review of the contributing factors, challenges and future perspectives. *European Journal of Lipid Science and Technology* 117, 13451369.

Elgersma A., Ellen G., van der Horst H., Muuse B.G., Boer H. and Tamminga S. (2004) Quick changes in milk fat composition after transition from fresh grass to a silage diet and effects on consumer health benefits. *Animal Feed Science and Technology* 117, 13-27.

Heck J.M.L., van Valenberg H.J.F., Dijkstra J. and van Hooijdonk A.C.M. (2009) Seasonal variation in the Dutch raw bovine milk composition. *Journal of Dairy Science* 92, 4745-4755.

Kelly M.L., Kolver E.S., Bauman D.E., van Amburgh M.E. and Muller L.D. (1998) Effect of intake of pasture on concentrations of conjugated linoleic acid in milk of lactating cows. *Journal of Dairy Science* 81, 1630-1636.

Liu N., Pustjens A.M., Erasmus S.W., Yang Y., Hettinga K. and van Ruth S.M. (2020) Dairy farming system markers: The correlation of forage and milk fatty acid profiles from organic, pasture and conventional systems in the Netherlands. *Food Chemistry* 314, 126153.

Nitrogen flows in dairy cows fed various proportions of low-N fresh grass and maize silage

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Abstract

Dairy farms must reduce their environmental impacts and nitrogen (N) losses to ensure sustainable and efficient production. Combining fresh grass and conserved forages is common in dairy cow diets. However, the consequences of these associations, in different proportions, on N-use efficiency and flows at the animal level are poorly understood. This study aimed at quantifying the effects of four maize silage proportions in a fresh grass-based diet (0:100, 17:83, 34:66, 51:49 of maize silage:fresh grass ratio, dry matter (DM) basis), without any concentrate supply, on cow N excretion and N-use efficiency. The experiment was conducted with 7 lactating Holstein cows in a Latin square design. Intake, milk, urine and faeces amounts and their N concentration were measured individually. The crude protein (CP) concentrations of the 4 diets ranged between 107 and 86 g kg^{-1} DM, due to the very low grass CP concentration. DM intake and milk production decreased with increasing maize proportion in the diet, as well as faecal and urinary N excretions. Including 50% of maize silage in a low-N grass diet improved N-use efficiency. These results should help to improve predictive equations of N losses for feeding strategies combining fresh grass and maize silage.

Keywords: dairy cow, fresh grass, maize silage, N-use efficiency, N flows

Introduction

Dairy systems must enhance their feed self-sufficiency, while reducing their environmental impacts to ensure sustainable production. Fresh grass is one of the solutions, being a low-cost on-farm feed, with an interesting feeding value, and providing environmental services. But its composition and availability vary throughout the year, leading farmers to frequently associate it with the use of conserved forages like maize silage. The effect of maize silage inclusion in a fresh grass-based diet on N excretion and efficiency is poorly understood. It is recognized that reducing dietary N concentration and intake has beneficial impacts on N excretion and efficiency (Castillo *et al.,* 2000; Spanghero and Kowalski, 2021). Moreover, combinations of forages might change N use by the cow through digestive interactions (Valk, 1994). This experiment aimed to quantify the effects of increasing the level of substitution of fresh grass by maize silage on N excretion and efficiency, without any concentrate supply.

Materials and methods

This experiment was conducted at the INRAE experimental dairy farm of Le Rheu (France), from April to June 2021. Four maize silage proportions in a fresh grass-based diet (0:100, 17:83, 34:66, 51:49 of maize silage:fresh grass ratio, DM basis) were compared with 7 mid-lactation Holstein cows, during 3 periods of 3 weeks, in an incomplete Latin square design. Each period was composed of 15 adaptation days and 6 measurement days. Cows were housed in tie stalls and were milked twice daily. On average, cows were at 166±39.8 days in milk, produced 22.2±4.78 kg of milk d^{-1} and weighed on average 601±83.1 kg just prior to the experiment. During the experiment, cows were fed *ad libitum*, with more than 10% of refusals in each treatment, without any concentrate supply. The fresh grass was perennial ryegrass (*Lolium perenne* L.), cut once daily, and available from 08:00 to 18:00 h. Maize silage was distributed at 18:00 and was available all night, as was fresh grass for the 0:100 diet. The proportions of grass and maize in the ingested diet were controlled carefully and daily, while maintaining at least one forage *ad libitum*. Feeds offered and refused were weighed and dried daily to determine the individual dry matter intake (DMI).

Milk production and composition were recorded daily. Urine and faeces were totally and separately collected during 5 consecutive days at the end of each period. Faecal samples per cow were dried daily. Nitrogen concentration of offered and refused forages, milk, urine and faeces were determined with the Dumas method. Nitrogen-use efficiency was calculated by dividing milk N (g d^{-1}) by N intake (g d^{-1}). Nitrogen balance default was calculated from N intake less milk N, faecal N and urinary N (g d^{-1}). Data were analysed using ANOVA considering the treatment, period and cow effects. The linear and quadratic effects of the proportion of maize silage in the diet were determined by orthogonal contrasts.

Results and discussion

As expected, the maize silage proportion in the ingested diet followed a regular interval between diets. Total DMI and digestible organic matter (OM) intake decreased linearly by 0.67 and 0.57 kg d^{-1}, respectively, for each increase of 10 percentage points of maize silage in the diet, while fresh grass DMI decreased with a quadratic effect (Table 1). There was no significant difference in diet DM concentration between treatments. The diet CP concentration and *in vivo* OM digestibility decreased respectively by 4.41 g kg^{-1} DM and 10.4 g kg^{-1} for each increase of 10 percentage points of maize silage in the diet. The milk production was reduced by 0.72 kg d^{-1} for each increase of 10 percentage points of maize silage. Milk fat concentration was unaffected by diets, while milk true protein concentration was greater for the 0:100 diet than for diets including maize. The N intake decreased by 20 g d^{-1} for each increase of 10 percentage points of maize silage in the diet. The milk N was greater (+20 g d^{-1}) for the 0:100 diet than for diets including maize silage. Faecal and urinary N decreased linearly by 5.5 and 5.0 g d^{-1}, respectively, for each increase of 10 percentage points of maize silage in the diet. The N-use efficiency was significantly higher (+0.06) for the 51:49 diet compared to the 3 other treatments. Finally, the N balance default decreased by 4.8 g d^{-1} for each increase of 10 percentage points of maize silage in the diet.

Table 1. Intake, milk production, N flows and efficiency responses to the maize silage proportion in a fresh grass-based diet on dairy cows.[1]

Variable	Treatments				RSD	Response (P<)	
	0:100	17:83	34:66	51:49		Linear	Quadratic
DMI, kg d^{-1}	15.7^a	14.1^b	13.4^{bc}	12.4^c	0.92	0.002	0.475
Fresh grass DMI, kg d^{-1}	15.7^a	11.6^b	8.9^c	6.2^d	0.58	0.000	0.047
Digestible OM intake, kg d^{-1}	11.0^a	9.7^{ab}	9.0^b	8.2^b	0.95	0.004	0.652
Diet DM concentration, g kg^{-1}	228	245	256	263	26.6	0.077	0.716
Diet CP concentration, g kg^{-1} DM	107^a	99^b	92^c	86^c	4.10	0.000	0.595
Diet OM digestibility, g kg^{-1}	749^a	737^{ab}	715^{ab}	699^b	26.9	0.022	0.870
Milk production, kg d^{-1}	16.4^a	14.8^{ab}	13.0^b	13.2^b	1.30	0.006	0.202
Milk fat concentration, g kg^{-1}	41.9	39.9	42.0	43.2	2.05	0.207	0.162
Milk protein concentration, g kg^{-1}	32.4^a	30.3^b	30.3^b	30.8^b	0.72	0.018	0.011
N intake, g d^{-1}	267^a	223^b	197^{bc}	168^c	18.4	0.000	0.430
Milk N, g d^{-1}	89.9^a	76.0^b	67.1^b	67.6^b	6.12	0.002	0.050
Faecal N, g d^{-1}	104^a	90^b	85^{bc}	76^c	6.74	0.001	0.543
Urinary N, g d^{-1}	66.1^a	59.9^a	51.1^b	42.1^c	4.62	0.000	0.550
N use efficiency, g g^{-1}	0.33^a	0.34^a	0.34^a	0.40^b	0.03	0.022	0.136
N balance default, g d^{-1}	7.7^a	-2.7^{ab}	-6.4^{ab}	-17.2^b	10.8	0.013	0.968

[1] DM = dry matter; DMI = dry matter intake; CP = crude protein; OM = organic matter; RSD = residual standard deviation; Treatments expressed as maize-silage:fresh-grass ratios; Means within rows having different superscript letters differ significantly at the 0.05 confidence level.

This experiment was conducted with very low-CP diets for lactating dairy cows, due to both an unusually low CP concentration of fresh grass and the inclusion of high proportions of maize silage without any N-rich concentrate supply. The large reduction of DMI with increasing maize silage proportion in the diet most likely resulted from the low diet CP concentration, known to reduce voluntary intake (Rico-Gómez and Faverdin, 2001). The drop in DMI is certainly the main cause of the reduction of faecal N, as reported in the literature (Castillo *et al.*, 2000). The mean faecal N:DMI ratio of 6.4 g kg^{-1} DMI is rather low compared to previous studies (7.2 g kg^{-1} DMI; Peyraud and Delaby, 2006). The reduction of urinary N was largely caused by the N intake reduction, in accordance with the literature (Castillo *et al.,* 2000). The use of the Spanghero and Kowalski (2021) predictive equation led to a mean overestimation of faecal N of 43 g d^{-1} compared to our measurements. On the contrary, this equation conducted to a mean underestimation of urinary N by 45 g d^{-1}, even leading to a negative urinary N for the diet with the lowest N intake (51:49). It is noteworthy that our experiment investigated very deficient-N diets, rarely studied in dairy cows. Most of the current predictive equations are probably inaccurate for these extreme diets.

The milk N reduction with increasing maize silage proportion in the diet was lower than the N intake reduction, leading to a greater N-use efficiency for the 51:49 diet. Ruminants are indeed able to recycle urea in the rumen to save nitrogen and compensate, at least partially, the N deficit (Peyraud and Delaby, 2006). It may also be hypothesized that cows fed with the lowest N diets mobilized N from their body reserves and from the involution of the digestive tract caused by the DMI reduction (INRA, 2018). This would explain the negative N balance default for the maize-silage rich diets.

Conclusions

Increasing the proportion of maize silage in unusually low-N grass diets induced very N-deficient diets, and large reductions of DMI, milk production, faecal and urinary N excretions. Regardless of the maize silage proportion, the milk N did not change for the diets combining fresh grass and maize silage, leading to increased N-use efficiency for the 51:49 diet. These results should help to improve future predictive equations of N losses for feeding strategies combining fresh grass and maize silage, particularly with low N concentrations.

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References

Castillo A.R., Kebread E., Beever D.E. and France J. (2000) A review of efficiency of nitrogen utilisation in lactating dairy cows and its relationship with environmental pollution. *Journal of Animal and Feed Sciences* 9, 1-32.

INRA (2018) *INRA feeding system for ruminants*. Wageningen Academic Publishers, Wageningen, the Netherlands, 640 pp.

Peyraud J.L. and Delaby L. (2006) Grassland management with emphasis on nitrogen flows. In: Elgersma A., Dijkstra J. and Tamminga S. (eds.) *Fresh herbage for dairy cattle: The key to a sustainable food chain*, Springer, the Netherlands, 103-123.

Rico-Gómez M. and Faverdin P. (2001) La nutrition protéique modifie l'ingestion des vaches laitières: analyse bibliographique. *Rencontres Recherche Ruminants* 8, 285-288.

Spanghero M. and Kowalski Z.M. (2021) Updating analysis of nitrogen balance experiments in dairy cows. *Journal of Dairy Science* 104, 7725-7737.

Valk H. (1994) Effects of partial replacement of herbage by maize silage on N-utilization and milk production of dairy cows. *Livestock Production Science* 40, 241-520.

Milk production from grass-white clover systems over two full lactations

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Abstract

A farm systems experiment was undertaken at Teagasc, Moorepark, Ireland over two full lactations (2019 and 2020). The objective was to compare milk yield and quality production from grass-only swards receiving 250 kg N ha^{-1} (GR250) and grass-white clover swards receiving 150 kg N ha^{-1} (CL150) or 100 kg N ha^{-1} (CL100). All treatments were stocked at 2.74 cows ha^{-1}. Target pre-grazing herbage mass was 1,300-1,600 kg dry matter (DM) ha^{-1} and target post grazing height was 4 cm for all treatments. Average annual herbage production was similar for all treatments, though pre-grazing herbage mass was significantly (P<0.05) lowest with CL100 and post-grazing sward heights significantly greatest (P<0.05) with GR250, resulting in similar herbage removed from all swards. Unexpectedly, this resulted in no significant differences in milk yield or quality between cows grazing swards with and without clover. This did, however, indicate that milk outputs can be maintained using fertilizer N inputs of 100-150 kg N ha^{-1} less when white clover is actively present.

Keywords: clover, milk production, stocking rate, nitrogen

Introduction

The inclusion of white clover in swards of perennial ryegrass can contribute to the sustainability of intensive agriculture by reducing the requirement for chemical N fertilizer through the process of biological N fixation. It has been shown that reducing chemical N fertilizer application rates to 150 kg N ha^{-1} on grass-white clover swards can maintain or improve herbage dry matter (DM) production compared to grass-only swards receiving 250 kg N ha^{-1} (Egan *et al.*, 2018). In Ireland, the standard model for intensive pasture dairy systems is based on perennial ryegrass swards with high levels of chemical N fertilizer supplementation. Globally, policy makers and consumers are becoming more environmentally conscious and white clover offers opportunities to improve sustainability. A recent meta-analysis of the literature on white clover concluded that for cows grazing perennial ryegrass-white clover swards, mean daily milk and milk solids (MS) yields were improved by 1.4 and 0.12 kg, respectively, compared to grass-only swards (Dineen *et al.*, 2018). This improved performance can be attributed to the superior nutritional feed quality and higher voluntary herbage DM intake (DMI) associated with white clover (Egan *et al.*, 2018). In a life-cycle assessment, it was shown that grass-white clover swards can reduce the environmental impact of spring-calving, pasture-based intensive dairy systems through improved animal performance and reduced total emissions (Herron *et al.*, 2021). The current study sought to quantify these potential benefits within an intensive dairy system over two lactations by comparing milk production from grass-only and grass-white clover systems.

Materials and methods

A full lactation farm-systems experiment was undertaken at Teagasc, Moorepark, Ireland from February to November 2019 and 2020. The experiment had three treatments: grass-only swards at 250 kg N ha^{-1} (GR250) and grass-clover swards at 100 or 150 kg N ha^{-1} (CL100 and CL150 respectively). The GR250 swards were a 50:50 perennial ryegrass (*Lolium perenne* L.) mixture of cv. AstonEnergy (tetraploid) and cv. Tyrella (diploid) sown at 27 kg ha^{-1}. The grass-clover swards contained the same perennial ryegrass mixture (27 kg ha^{-1}) plus a 50:50 blend of medium leaf size Chieftain and Crusader white clovers

(*Trifolium repens* L.) sown at 5 kg ha^{-1}. In February each year, 54 spring calving dairy cows (Fresian and Fresian × Jersey) were selected and balanced on mean calving date (10 February), lactation number (2.8), parity (2.23), pre-experimental milk yield, and pre-experimental MS yield, gathered during the two weeks prior to commencement of the experiment and randomly allocated to one of three treatment groups (n=18). All treatments were stocked at 2.74 cows ha^{-1} in a closed farm system with cows staying in their treatment groups for the entire lactation. Average concentrate supplementation was 535 kg cow^{-1} fed throughout the entire lactation. The cows received a daily herbage allowance of 17 kg DM cow^{-1} above 4 cm and an individual concentrate allocation of 1 kg per cow per day. In spring and autumn, the quantity of concentrate fed to the treatment cows increased to 3 kg. Swards were rotationally grazed. Target pre-grazing herbage mass was 1,300-1,600 kg DM ha^{-1} and this was measured twice weekly by harvesting two strips from each paddock to be grazed next, using an Etesia mower (Etesia UK Ltd., Warick, UK). Herbage production was categorized as herbage or silage production. Cumulative herbage production was recorded and calculated using the online tool PastureBase. The target post-grazing height was 4 cm. Sward clover content was measured prior to each grazing, twice weekly using the method described by Egan *et al.* (2018). Milk yield was recorded daily (Dairymaster, Causeway, Co. Kerry, Ireland) and milk composition (fat and protein concentrations) was measured twice weekly using MilkoScan 203 (Foss Electric, Hillerød, Denmark). Data were analysed in SAS using Proc Mixed with terms for treatment, time (week or rotation) and associated interactions. Fixed terms were treatment and week or rotation, and random terms were cow and paddock.

Results and discussion

The average annual clover content for both CL100 and CL150 was 20%, with peak clover content occurring in September each year. The average total annual pasture production was 12.9 t DM ha^{-1} on CL100, 13.8 t DM ha^{-1} on CL150 and 14.4 t DM ha^{-1} on GR250. The similarity in herbage production from all three sward treatments indicates that biologically fixed N from the clover was largely compensating for the additional 100-150 kg N ha^{-1} applied to the grass-only sward. Pre-grazing herbage mass was significantly (P<0.05) lower on the CL100 treatment compared to CL150 and GR250 (Table 1). Post-grazing sward heights was significantly greater (P<0.05) on the GR250 treatment compared to the CL100 and CL150 treatments. The height of the grazing horizon removed from CL100 and CL150 averaged 2.84 cm, while GR250 averaged 3.14 cm, which were not significantly different. This observation conflicts with previous studies including Egan *et al.* (2018), where significantly greater DMI has been reported with grass-white clover diets compared to grass-only diets. Notably, all three swards supported the same mean average daily milk yield and milk solids yield, and were also similar in their fat and protein fractions (Table 1). Likewise, the similar cumulative milk yield and milk solids yield and quality from cows grazing the different sward types was also unexpected. The absence of a DMI difference between the grass-only and grass-clover swards was unexpected, as others such as Steinshamn (2010) have reported white clover grazing preferences and faster rumen passage rates resulting in increased DMI compared to grazing pure grass. According to Frame and Laidlaw (1998) and Rochon *et al.* (2004), to gain the full potential benefit from white clover requires a content level of over 30% to be maintained within the sward from year to year. Therefore, as the clover content was lower in the current study, this could explain why the numerically larger responses from the swards containing clover were not large enough to be statistically significant.

Table 1. Comparison of milk and herbage production on grass-only and grass-clover swards.[1]

	GR250	CL150	CL100	SE	*P*-value
Milk yield (kg cow^{-1} day^{-1})	20.9	21.2	21.7	0.5243	ns
Milk fat (g kg^{-1})	49.71	50.94	50.31	0.4848	ns
Milk protein (g kg^{-1})	37.10	37.74	38.03	0.2749	ns
Milk solids yield (kg cow^{-1} day^{-1})	1.77	1.84	1.87	0.3672	ns
Cumulative milk yield (kg cow)	5,873	6,104	6,133	181.27	ns
Cumulative milk solid yield (kg cow^{-1})	508	524	534	14.869	ns
Pre-grazing herbage mass (kg DM ha^{-1})	1,521^a	1,469^a	1,396^b	32.621	<0.05
Post-grazing sward height (cm)	4.10^a	4.01^b	4.01^b	0.027	<0.05

[1] GR250 = grass only at 250 kg N ha^{-1}; CL150 and CL100 = grass-clover at 150 and 100 kg N H ha^{-1}, respectively.

Conclusions

The similar cumulative milk volume and solids yield maintained on all three treatments in this study has demonstrated an opportunity to limit N fertilizer application rates to around 100 - 150 kg N ha^{-1} on grazed grass-clover swards, stocked at 2.74 cows ha^{-1}. This potential opportunity to reduce N applications without significant loss of milk productivity in a full farm system, at clover contents averaging of around 20%, is important. This is particularly so, as the Irish Government target for reducing GHG emissions by 2030 is 51% compared to 2018 levels (www.gov.ie/en/press-release/16421-climate-action-plan-2021-securing-our-future/), with a significant proportion of this linked to N fertilizer use (Environmental Protection Agency, 2020, https://www.epa.gov/nutrient-policy-data).

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References

Dineen M., Delaby L., Gilliland T. and McCarthy B. (2018) Meta-analysis of the effect of white clover inclusion in perennial ryegrass swards on milk production. *Journal of Dairy Science* 101, 1804-1816.

Egan M., Galvin N. and Hennessy D. (2018) Incorporating white clover (*Trifolium repens* L.) into perennial ryegrass (*Lolium perenne* L.) swards receiving varying levels of nitrogen fertilizer: Effects on milk and herbage production. *Journal of Dairy Science* 101, 3412-3427.

Enriquez-Hidalgo D., Gilliland T.J. and Hennessy D. (2016) Herbage and nitrogen yields, fixation and transfer by white clover to companion grasses in grazed swards under different rates of nitrogen fertilization. *Grass and Forage Science* 71, 559-574.

Herron J., Hennessy D., Curran T., Moloney A. and O'Brien D. (2021) The simulated environmental impact of incorporating white clover into pasture-based dairy production systems. *Journal of Dairy Science* 104, 7902-7918.

Laidlaw A.S., Withers J.A and Toal L.G. (1995) The effect of surface height of swards continuously stocked with cattle on herbage production and clover content over four years. *Grass Forage Science* 50, 48-54.

Rochon J.J., Doyle C.J., Greef J.M., Hopkins A., Molle G., Sitzia M., Scholefield D. and Smith C.J. (2004) Grazing legumes in Europe: A review of their status, management, benefits, research needs and future prospects. *Grass and Forage Science* 59, 197-214.

Steinshamn H. (2010) Effect of forage legumes on feed intake, milk production and milk quality – a review. *Animal Science Papers and Reports* 28, 195-206.

Milk production and quality from grass-only, PMR and TMR feeding systems

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Abstract

Where access to additional land is limited, increasing production forces farmers to either increase herbage production from the same land area, or feed higher levels of concentrate at grass, or consider alternative diets, e.g. partial mixed ration (PMR) or total mixed ration (TMR). The objective of this study was to determine the extent to which these dietary alternatives support higher milk production than a predominately grass-based system over a full lactation cycle. The three treatments were, TMR (grass/maize silage, concentrate), PMR (grass/maize silage, concentrate, grazed grass) and grazed grass plus concentrate (G). TMR had the highest cumulative milk yield (MY) and milk solids yield (MSY) (8,047 and 656 kg cow^{-1}, respectively), with the PMR intermediate (7,709 and 617 kg cow^{-1}, respectively) and G lowest (6,045 and 515 kg cow^{-1}, respectively). The TMR and PMR diets resulted in significantly greater (P<0.001) daily milk and milk solids yield compared to G cows but no differences in fat or protein contents.

Keywords: grass, total mixed ration, partial mixed ration, milk production, diet

Introduction

In temperate regions, low input dairy farming systems are characterized by a long grazing season and a predominantly pasture-based diet is the lowest cost feed system for milk production. However, there are many challenges associated with grass utilization and quality which include cow type, cow and sward interaction and factors affecting dry matter (DM) intake (DMI) (Hennessy *et al.*, 2020). Limitations in grass based dairy production systems such as access to additional land, lower feed efficiency due to limited energy intake and the removal of the EU milk quota in 2015 has forced some farmers to consider supplementation in the form of either partial mixed ration (PMR) or total mixed ration (TMR) (O'Neill *et al.*, 2012). There are many advantages associated with PMR and TMR, such as a fully nutritionally balanced diet with fewer digestive upsets, fewer incidents of issues, such as milk fat depression, higher DMI and greater milk production potential (Schingoethe, 2017). Fluctuations in grass quality and supply influenced by factors such as stage of growth, sward nutrition and water content which can reduce control of feed quality resulting in more variable milk production results from a grass-only diet (Hennessy *et al.*, 2020). Pasture derived dairy products are seen by consumers as more natural and more environmentally conscious because animals are allowed to express normal behaviours (Legrand *et al.*, 2009). In contrast, consumer perception of TMR derived milk products is not as favourable. Therefore, the study objective was to examine these dietary options and determine to what extent milk yield and quality could be raised above a grass-based diet.

Materials and methods

A full lactation farm systems experiment was conducted at Teagasc, Moorepark, Fermoy, Co. Cork, Ireland from February to November 2020. A total of 54 spring calving dairy cows were randomly allocated to one of three treatment groups according to breed (Friesian or Jersey × Friesian), calving date (19/02/2020), lactation number (2.78), parity (2.31), and pre-experimental milk yield (MY) and milk solid yield (MSY). Three treatments were imposed: TMR, PMR, and grass-only (G). PMR and G swards received 250 kg N ha^{-1}. In the TMR treatment, cows were cows housed indoors fulltime and fed 9 kg concentrate (100 g kg^{-1} of DM beet pulp, 280 g kg^{-1} of DM 48% crude protein soybean meal, 120 g kg^{-1} of DM rolled barley, 180 g kg^{-1} of DM maize, 100 g kg^{-1} of DM maize distillers, 100 g kg^{-1} of DM rapeseed meal, 80 g kg^{-1} of DM soya hulls, 10 g kg^{-1} of DM fat and 25 g kg^{-1} of DM plus post-calver maize minerals), 9 kg maize silage and 4.5 kg grass silage on a DM per cow basis, using an electronic controlled Roughage Intake Control system feed bins (Hokofarm Group B.V., Marknesse, the Netherlands). The cows were fed ad-libitum to achieve approximately 10% refusal levels. In the PMR treatment cows were housed indoors at night and fed the same ration as the TMR cows, at half the rate, using the Roughage Intake Control system and grazed grass during the day. The PMR diet consisted of 4.5 kg concentrate, 4.5 kg maize silage, 2.25 kg grass silage and 9 kg grazed grass. TMR was fed at 08.30 h each day using Keenan diet feeder (Keenan, Borris, Carlow, Ireland). The G and PMR swards were predominantly perennial ryegrass (*Lolium perenne* L.). The G treatment was stocked at 2.74 cows ha^{-1} and daily herbage allowance was 17 kg DM cow^{-1}. Target pre-grazing herbage mass was 1,300-1,500 kg DM ha^{-1} and target post-grazing height was 4-4.5 cm. All cows received the 1 kg concentrate in the parlour per day. In spring and autumn the quantity of concentrate fed to the G cows increased to 3 kg. Total concentrate fed to the G cows was 570 kg cow^{-1}. Target pre-grazing herbage mass was 1000-1,200 kg DM ha^{-1} and the target post grazing height was 4-4.5 cm for the PMR treatment. Milk yield was recorded daily (Dairymaster, Causeway, Co. Kerry, Ireland) and milk composition (fat and protein concentrations) was measured weekly using MilkoScan 203 (Foss Electric, Hilerod, Denmark). Data were analysed in SAS using Proc Mixed. Fixed terms were treatment and week and, the random terms were cow.

Results and discussion

Mean daily milk yield and milk solids yield were significantly ($P<0.01$ and $P<0.01$, respectively) greater on the TMR and PMR treatments than the G treatment (Table 1). This was likely a reflection of the higher and more consistent quality of the nutritionally balanced TMR and PMR diets compared to pasture based diets (Bargo *et al.*, 2002). One of the main constraints on milk production of cows consuming high quality pasture is intake of nutrients (Kolver and Muller, 1998). The higher milk volume and solids of those on the higher energy diets was therefore consistent with previous studies.

It was notable that body condition score was highest for cows on the TMR, intermediate for those on the PMR and lowest for those on the G pasture (3.22, 3.14 and 3.08, respectively, $P<0.05$). The differences in body condition score, which can be attributed to a number of factors, was most probably due to the lower energy intake and higher energy maintenance requirements of the G cows, due to walking and grazing activity (Bargo *et al.*, 2002). The G diet provided a significantly lower ($P<0.001$) cumulative MY and cumulative MSY compared with PMR and TMR (Table 1) and this was largely expressed consistently throughout the lactation (Figure 1). There was no significant treatment effect on fat and protein concentration. The less-expected responses were the absence of changes in the fat and

Table 1. Comparison of three dairy cow diets for daily and cumulative milk production and quality.[1]

	GR250	TMR	PMR	SE	*P*-value
Milk yield (kg cow^{-1} day^{-1})	21.5^a	26.9^b	26.2^b	1.1283	<0.01
Fat content (g kg^{-1})	49.45	49.58	47.83	0.6394	ns
Protein content (g kg^{-1})	36.66	37.31	36.07	0.4104	ns
Lactose content (g kg^{-1})	46.67^a	47.50^b	46.48^a	0.1518	<0.001
Milk solids yield (kg cow^{-1} day^{-1})	1.82^a	2.29^b	2.13^b	0.0669	<0.01
Cumulative milk yield (kg cow^{-1})	$6,045^a$	$8,047^b$	$7,709^b$	314.457	<0.001
Cumulative milk solids yield (kg cow^{-1})	515^a	656^b	617^b	24.147	<0.001

[1] ns = not significant.

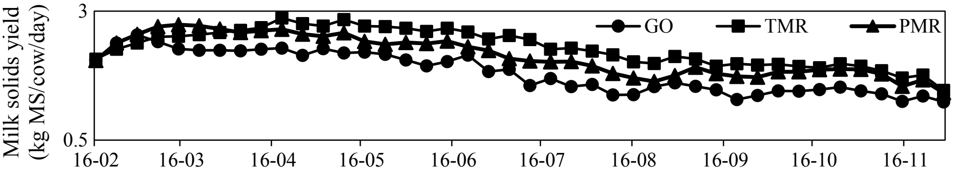

Figure 1. Comparison of three dairy cow diets for average daily milk solids yield, February to November.

protein fractions, despite the large differences between the diets. This suggests that the G diet with a 1 kg supplementation plus the loss in body condition score was largely sufficient to meet the full dietary needs of these cows, when producing these milk volumes. Milk yield was more persistent later into lactation on PMR and TMR diets compared to G, likely due to a more consistent feed supply compared to the pasture based diets.

Conclusions

Feed system (TMR, PMR or G) had a significant effect on milk production. Milk production was significantly greater on the TMR and PMR diets compared to the G diet. Milk yield was more persistent later into lactation on PMR and TMR diets compared to G, likely due to a more consistent feed supply compared to pasture based diets. Offering a more consistent feed quality diet, such as that in the ration offered to the TMR and the PMR at night, resulted in higher cumulative and daily milk yield and milk solids yield. Further examination of DMI and grass and feed quality are required to determine the full effect of diet on milk production.

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References

Bargo F., Muller L.D., Delahoy J.E. and Cassidy T.W. (2002) Performance of high producing dairy cows with three different feeding systems combining pasture and total mixed rations. *Journal of Dairy Science* 85(11), 2948-2963.

Hennessy D., Delaby L., van den Pol-van Dasselaar A. and Shalloo L. (2020) Increasing grazing in dairy cow milk production systems in Europe. *Sustainability* 12(6), 2443.

Kolver E.S. and Muller L.D. (1998) Performance and nutrient intake of high producing Holstein cows consuming pasture or a total mixed ration. *Journal of Dairy Science* 81(5), 1403-1411.

Legrand A.L., Von Keyserlingk M.A.G. and Weary D.M. (2009) Preference and usage of pasture versus free-stall housing by lactating dairy cattle. *Journal of Dairy Science* 92(8), 3651-3658.

O'Neill B.F., Deighton M.H., O'Loughlin B.M., Galvin N., O'Donovan M. and Lewis E. (2012) The effects of supplementing grazing dairy cows with partial mixed ration on enteric methane emissions and milk production during mid to late lactation. *Journal of Dairy Science* 95(11), 6582-6590.

Schingoethe D.J. (2017) A 100-year review: total mixed ration feeding of dairy cows. *Journal of Dairy Science* 100(12), 10143-10150.

Co-grazing horses and cattle requires appropriate management to provide its expected benefits

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Abstract

Co-grazing different herbivores at pasture is assumed to increase vegetation use because of the complementarity of their feeding choices, and to reduce parasitism as the result of a dilution effect. Here, we compare the effects of mixed horse-cattle grazing and monospecific horse grazing (1.4 lifestock units ha^{-1}) on animal foraging behaviour, sward characteristics and horse parasitism in a mesophile grassland (central France). In both treatments, animals alternatively grazed two subplots, each for 15 to 21 days. All horses selected short (≤4 cm) and intermediate (5-8 cm) high-quality patches and avoided reproductive and dead herbage areas contaminated by their faeces. Cattle, which are more constrained by sward surface height, selected intermediate and tall (≥9 cm) vegetative swards. They used short vegetative patches proportionally to their availability as the alternate stocking management enabled short patches to regrow before animals entered the subplots again. Cattle avoided reproductive and dead herbage areas which limited their ability to remove parasitic larvae from the environment. Co-grazing horses and cattle did not reduce sward structural heterogeneity and thus did not enhance herbage quality. We conclude that understanding and optimizing ecological processes in mixed grazing systems is required so that these systems can provide their expected benefits.

Keywords: agroecology, mixed grazing, diet selection, herbage quality, parasite dilution

Introduction

Enhancing diversity within animal production systems is a key principle of agroecology to improve livestock sustainability. While several studies have reported the ability of mixed grazing between horses and cattle to preserve biodiversity in semi-natural habitats, references are lacking in more productive grasslands which support saddle horse systems. Here, we compared co-grazing of saddle horses and beef cattle with horses grazing alone at the same stocking density in a mesophile grassland of central France. We hypothesized that the strong selection of short high quality patches by horses would constrain cattle that are more limited by sward surface height to switch onto taller vegetation and thus to consume grass close to horse latrine areas. Conversely, cattle are assumed to use dicotyledons more than do horses, as they are better able to detoxify their secondary compounds (Ménard *et al.*, 2002). Co-grazing by cattle and horses is thus assumed to homogenize sward structure and enhance herbage quality, and to remove parasites from the environment, both processes beneficial for horse nutrition and health.

Materials and methods

The study was carried out at the French Horse and Riding Institute (IFCE) experimental farm in Chamberet (440 m a.s.l) over three grazing seasons (April to October 2015-2017). The horses were drenched each year with pyrantel before pasture turn-out. Comparison of treatments (mixed grazing by two 2-yr old saddle horses and three heifers vs grazing by four horses; 1.4 lifestock unit (LU) ha^{-1} with 1 LU = 600 kg liveweight) was replicated in three blocks. In both treatments, animals alternatively grazed two subplots A and B of 1.35 ha each for 15 to 21 days. Sward surface height (SSH) was measured with a stick each time the animals entered and were turned out from a subplot (200 sampling points). Dietary choices were observed by scan sampling at 10 min intervals in May, July and September, with one observation-day (from dawn to dusk) per subplot A and per season. Bites were recorded according to sward height type (vegetative short, VS, ≤4 cm; vegetative intermediate, VI, 5-8 cm; vegetative tall, VT, ≥9 cm; reproductive and dead herbage) and dominant botanical family. We also recorded the presence of horse dung within one metre around each bite location, as most of small strongyle larvae move less than 1 m from dung (Fleurance *et al.*, 2007). Diet selection was quantified with Jacobs' indices: $S_i = (c_i - a_i) / (c_i + a_i - 2c_i a_i)$ where c_i and a_i are proportions of component i in the diet and in the subplot A, respectively. Data for individuals within each species were aggregated per day and were then related to this bite type abundance in subplot A (400 sample points). S_i varies from -1 (never used) to +1 (exclusively used), with negative and positive values indicating avoidance and selection, respectively. We estimated overall biomass and herbage quality (CP, NDF) close to dietary choice measurements in May, July and September: six samples were cut at ground level in each sward height type and overall biomass and herbage quality were estimated from the sward height types' proportions in the subplot. Horse parasite burden was estimated from monthly individual faecal egg counts (FECs). Animals were weighed on two successive days at the start and end of each grazing season. Finally, we recorded agonistic behaviours between horses and heifers using focal observations during daylight three weeks per month. Jacobs' indexes in horses and sward characteristics were analysed using the Anova procedure of SAS for repeated measurements including the effects of grazing management, date, grazing management × date, block, year and block × year. Jacobs' indexes in horses and cattle grazing mixed plots were analysed for the effects of species, date, species × date and year. Jacobs' indexes were compared to zero using Student's t-test. Individual FECs and horse daily liveweight gains were analysed using a mixed model with individual as a random effect and grazing management, date (FECs), grazing management × date (FECs), block, year and block × year as fixed effects.

Results and discussion

Only seven agonistic interactions were reported in horses towards heifers. Horses, whether they grazed alone or with cattle, exhibited typical patterns of diet selection (Ménard *et al.*, 2002). They selected VS and VI patches and preferred bites dominated by grasses (Figure 1 for mixed plots). They used VT swards and legumes in proportion to their availability and rejected forbs, reproductive swards and dead herbage. Cattle selected VT swards and used VS patches proportionally to their abundance (Figure 1). This result contrasts with previous studies in which cattle were excluded from the short patches (1-4 cm) created by horses (Cornelissen and Vulink, 2015; Ménard *et al.*, 2002). Here, the strong use of the VS patches by cattle probably resulted from the alternate stocking management, which let short swards regrow to an average of 3.9 cm before animals entered the subplots again. An additional explanation can be found in the high selectivity of Limousin cattle (D'Hour *et al.*, 1995). No differences in selection between the two species were found for the other bite types (Figure 1). Mean SSH (14 cm) and herbage biomass (198 g m^{-2}) were comparable in both treatments. A consequence of the similarity between horses and cattle choices is that the SSH coefficient of variation, an indicator of sward heterogeneity, did not differ between treatments (on average 55%). Cattle, by avoiding the reproductive and dead herbage areas, as did horses, did not improve herbage quality (mean crude protein: 115 g kg DM^{-1}, neutral detergent fibre: 599 g kg dry $matter^{-1}$). Moreover, we could not find evidence any reduction of parasite egg excretion in horses grazing with cattle (difference of 11 eggs g^{-1}). 40% of the horse faeces were recorded in the reproductive and dead herbage areas. By selecting vegetative regrowth, the cattle avoided grazing close to horse dung (S_i=-0.17±0.06, P=0.006) and thus ingested few parasitic larvae. While Forteau *et al.* (2020) have reported that co-grazing with cattle could reduce strongyle infection in young horses, we suggest that it would require an appropriate management of herds and plots. Consistently, average horse liveweight gains were similar in both treatments (378 g d^{-1}).

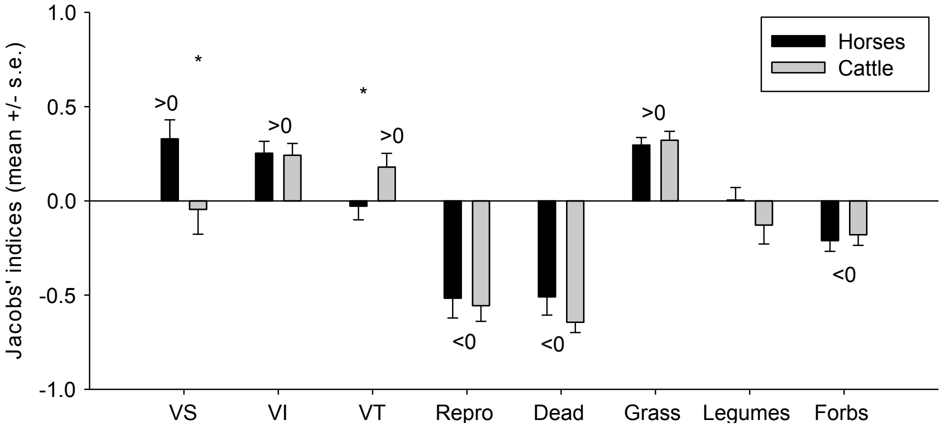

Figure 1. Diet selection by horses and cattle in mixed plots according to sward height type and dominant botanical family. Species effect: *$P<0.05$; >0 and <0 is for significant selection for or against this bite type.

Conclusions

We conclude that rather than considering mixed grazing as a turn-key solution, its management needs to be adapted to support the complementarity of horses and cattle dietary choices and thus provide the expected benefits of multi-species grazing.

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References

Cornelissen P. and Vulink, J.T. (2015) Density-dependent diet selection and body condition of cattle and horses in heterogeneous landscapes. *Applied Animal Behaviour Science* 163, 28-38.

D'Hour P., Petit M. and Garel J.P. (1995) Components of grazing behaviour of 3 breeds of heifers. *Annales de Zootechnie* 44, 270.

Fleurance G., Duncan P., Fritz H., Cabaret J., Cortet J. and Gordon I.J. (2007) Selection of feeding sites by horses at pasture: testing the anti-parasite theory. *Applied Animal Behaviour Science* 108, 288-301.

Forteau L., Dumont B., Sallé G., Bigot G. and Fleurance G. (2020) Horses grazing with cattle have reduced strongyle egg count due to the dilution effect and increased reliance on macrocyclic lactones in mixed farms. *Animal* 14, 1076-1082.

Ménard C., Duncan P., Fleurance G., Georges J.Y. and Lila M. (2002) Comparative foraging and nutrition of horses and cattle in European wetlands. *Journal of Applied Ecology* 39, 120-133.

In pasture-based dairy systems, breeding and feeding strategies affect GHG emissions and nitrogen losses

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Abstract

Which pasture-based dairy system is the most beneficial from an environmental impacts perspective? The results of the long-term experiment 'Which cow for which system?' (WCWS) undertaken at the INRAE experimental farm of Le Pin-au-Haras in Normandy were used to address this question. One hundred and sixty dairy systems were designed from the WCWS trial based on six factors monitored in the experiment, including feeding strategy (low (LFS) with only grass or high (HFS) with less grass, maize silage and concentrates) and breed (Holstein or Normande). Their common objective was to produce yearly 420,000 litres of milk. The CAP'2ER tool was used to determine the greenhouse gas (GHG) emission and the nitrogen losses of each system. The average GHG gross emission was 1.18±0.08 kg eqCO_2 l^{-1} of milk and the average nitrogen balance was 146±10 kg N ha^{-1}. The 'LFS' systems and the 'Holstein breed' systems had lower gross GHG emissions. The LFS had a higher nitrogen surplus (+7 kg N ha^{-1}) but the nitrogen leaching risk was significantly lower than HFS (-27 kg N ha^{-1}). This comparative analysis highlights the need for environmental trade-offs in the face of the diversity of impacts assessed.

Keywords: dairy cow, grazing systems, environmental impact, milk carbon footprint

Introduction

Pasture-based dairy systems are often highlighted from an environmental point of view. In grassland, nitrogen losses are limited and less dependent on imported protein or nitrogen resources. These systems also compensate a part of their greenhouse gas (GHG) emissions by helping to maintain the carbon stock in soils. However, the intensification of production in these pasture-based systems can lead to significant environmental impacts, as for example in New Zealand (Richard *et al.*, 2017). In this context, all the data from the 'Which cow for which system' (WCWS) experiment conducted between 2006 and 2015 on the INRAE experimental farm of Le Pin au Haras were mobilized to study the environmental responses (GHG emissions, nitrogen fluxes) of different grass-based systems at the farm level in a Normandy context.

Materials and methods

From the model and animal performance studied in the WCWS experiment over the 10 years of trial (Dall-Orsoletta *et al.*, 2018), one hundred and sixty dairy systems were designed. The factors introduced in the definition of the systems were breed (Holstein or Normande), age at first calving (24 or 36 months), genetic type (potential favourable to milk volume or to milk contents), feeding system (low feeding strategy – LFS – with grass grazed and conserved only or high feeding strategy – HFS – with less grass grazed, maize silage and concentrates), strategy of selling heifers in excess of renewal requirement (15 days of age or ready to calve) and replacement rate (from 20 to 40% in steps of 5%). The combination of these six factors composed of two to five models resulted in the construction of the dairy systems. Their common objective was to produce just over 420,000 litres of milk, representing a sales quantity of 400,000 kg of milk per year. The herd demography (cows and heifers) of each system was based on this production target, the breeding performance and was established on real data from the WCWS experiment, including the observed production level according to parity and the applied renewal rates. The useful agricultural area of each system was determined from the quantities of forage required per feeding period, the average biomass yields observed on farm and the typical rotations practised on the farm or in Normandy.

The environmental assessment of each dairy system was carried out using the CAP'2ER tool (2018) developed by the Institut de l'Elevage and focuses on all environmental impacts (GHG emissions, milk carbon footprint, nitrogen balance, nitrate leaching, etc.) in order to analyse the risk of pollution transfer between the different compartments.

The effect of the different factors studied on the environmental performance of the systems was evaluated by analysis of covariance according to a model integrating the breed, the feeding strategy, the age at first calving, the sales models of heifers in excess and the replacement rate added as a covariate.

Results and discussion

Table 1 presents synthetic data of the main characteristics of the studied dairy systems. The average agricultural area (AA) used for dairying by the farms in these systems is 68.9 ha, ranging from 37.5 to 116.0 ha. It is entirely occupied by permanent grassland in all configurations of the low feeding strategy. Between 45 and 104 cows are needed to meet the 420,000 litres milk target, reflecting very different levels of production between systems, both at the animal level (from 4,050 to 9,350 litres cow^{-1} year^{-1}) and at the forage area level (3,630 to 12,380 litres ha^{-1} year^{-1}). The surplus of the nitrogen balance was on average 146 kg N ha^{-1} (and ranged from 122 to 163 kg N ha^{-1}). These values are mainly a consequence of the N mineral fertilization applied and seem relatively high compared to the references of grassland systems or maize-grass systems found in western France (Foray *et al.*, 2017), but comparable to the values shown in Irish dairy systems (Buckley *et al.*, 2016).

In all the simulations carried out, the gross GHG emissions are on average 1.18±0.08 kg eqCO_2 l^{-1} of milk and the net carbon footprint is 0.91±0.12 kg eqCO_2 l^{-1}.

Table 2 summarizes the main environmental performances associated with the factors studied.

The system with the lowest GHG emissions per litre of milk (1.03 kg eqCO_2 l^{-1}) and the smallest milk carbon footprint is the Holstein system, low feeding strategy, with an age at first calving of 24 months, a 'volume' genetic orientation, a sale of heifers at 15 days and a replacement rate equal to 20%. However, the nitrogen balance is 160 kg N ha^{-1}, far from the 122 kg N ha^{-1} of the Holstein system with a high feeding strategy, an age at first calving of 36 months, a 'volume' orientation, and a renewal rate of 40%. Conversely, its GHG gross emissions reach 1.15 kg eqCO_2 l^{-1} milk.

Table 1. Average characteristics and environmental performance of the 160 systems designed from the WCWS experiment.

Indicators	Mean	Standard deviation	Min	Max
Agricultural area (ha) (AA)	68.9	19.4	37.5	116
Forage area (ha) (FA)	67.2	20.8	34	116
Permanent grasslands (ha)	52.6	33.2	6	116
No. of milking cows	69	15	45	104
Stocking rate (livestock unit ha^{-1} FA)	1.59	0.2	1.2	2.2
Milk production / forage area (l ha^{-1} yr^{-1})	6,901	2,170	3,628	12,379
Nitrogen balance / agricultural area (kg N ha-^{1} AA)	146	10	122	163
Nitrate leaching potential (kg N ha^{-1} AA)	57	15	32	93
GHG gross emission (kg eqCO_2 l^{-1} corrected milk)	1.18	0.08	1.03	1.34
Milk carbon footprint (kg eqCO_2 l^{-1} corrected milk)	0.91	0.12	0.71	1.11

Table 2. Effects of the main characterization factors of dairy systems on overall environmental performance.

Factors	Gross GHG emission (kg eqCO_2 l^{-1} corr. milk)		Milk carbon footprint (kg eqCO_2 l^{-1} corr. milk)		Nitrogen balance (kg N ha^{-1} AA)	
Breed	**Holstein**	**Normande**	**Holstein**	**Normande**	**Holstein**	**Normande**
	1.12	1.25	0.86	0.96	143	149
	P<0.001		P<0.001		P=0.002	
Feeding system	**LFS**	**HFS**	**LFS**	**HFS**	**LFS**	**HFS**
	1.21	1.15	0.81	1.01	149	143
	P<0.001		P<0.001		P<0.001	
Age at 1st calving	**24 months**	**36 months**	**24 months**	**36 months**	**24 months**	**36 months**
	1.16	1.20	0.92	0.90	154	138
	P=0.001		NS		P<0.001	

In this study, systems built around the Normande breed are penalized by the lower individual milk production, which requires more dairy cows and heifers for the same volume of milk delivered, insufficiently compensated by the associated additional meat product. Breeding systems with a 36 months age at first calving produce more milk per cow, but these cows emit more enteric methane. The herd sizes in these systems with a 36 months calving age are higher than with a 24 months calving age, leading to higher gross GHG emissions. Overall, at the system level, grass-based feeding strategies, without use of concentrates, are penalized by herd demographics but contributes to carbon storage through the increased presence of permanent grassland and partly compensate for gross GHG emissions.

Conclusions

It remains difficult to define the most relevant system from an environmental point of view by integrating all the indicators used. Indeed, low GHG emissions are not systematically synonymous with a low carbon footprint, nor with limited nitrogen balance or losses.

This study confirms the interest in a global and integrated approach to the different factors involved in the functioning of dairy systems in the evaluation of their environmental performance. Complementary and multidisciplinary approaches need to be devised to better define trade-off situations, depending on local or more global issues.

References

Buckley C., Wall D. P., Moran B., O'Neill S. and Murphy P.N.C. (2016) Farm gate level nitrogen balance and use efficiency changes post implementation of the EU Nitrates Directive. *Nutrient Cycling in Agroecosystems* 104, 1-13.

Dall-Orsoletta A.C., Leurent-Colette S., Foray S., Launay F. and Delaby L. (2018) Influence de la race, de la stratégie d'alimentation et de l'âge au 1er vêlage sur les émissions de méthane entérique intégrées à l'échelle du système laitier. *Rencontres autour des recherches sur les ruminants (3R), Dec 2018, Paris, France,* pp. 488-491.

Faverdin P. and Van Milgen J. (2019) Intégrer les changements d'échelle pour améliorer l'efficience des productions animales et réduire les rejets. *INRAE Productions Animales* 32(2), 305-322.

Foray S., Billy C., Manneville V., Dollé J.-B., Le Gall A., Vertes F. and Godinot O. (2017) Gestion de l'azote dans les systèmes d'élevage herbivores: évaluation et amélioration de l'efficience de l'azote, réduction des transferts vers les milieux aquatiques. *Idele, collection résultats*.

Richard M., Carlier M., Chotteau P. and Foray S. (2017) La filière laitière en Nouvelle-Zélande rattrapée par les exigences de durabilité. *Idele, Dossiers Economie de l'Elevage* 484.

Grassland Production Index, the future foundation of grassland insurance in France?

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Abstract

An index-based insurance solution was developed to estimate and monitor near real-time grassland production using the indicator Grassland Production Index. It is based on the sum of a biophysical parameter (i.e. the daily values of the fraction of vegetation cover (fCover)) derived from medium spatial resolution time series images enriched with meteorological data. The grassland production index computes the grassland biomass production over a determined geographical area through the grass growing season and is compared to a past reference available using time series images since 2003. To evaluate the index, *in situ* monitoring of grassland production on nine experimental farms in various pedoclimatic zones was carried out over 3 to 4 years depending on the farm. The monitoring was based on rising plate meter measurements associated with sward density for the grazed paddocks and total yield measurements at harvest for the mown paddocks. In total, over 630 paddocks were measured to determine their production. For each site, annual grassland production monitored *in situ* was compared to the index. There was a good correlation (R^2=0.7) between the measured data and the index considering the variability of the phenomenon studied, the operator effect and the difficulty of establishing a reference data set. The prospect of accurate estimation of grassland biomass in real time by satellite remote sensing is strengthened.

Keywords: grassland, remote sensing, rising plate meter, yield, insurance

Introduction

One way of managing the risk of yield loss in grassland production due to a climatic event is through an insurance contract. There are many obstacles to easily assess grass production: heterogeneity within grazing paddocks, multiple harvests across the year, forage mostly consumed on-farm. The continuous monitoring of grasslands by satellite observations is one way to overcome these difficulties (Le Poivre, 2020).

Since 2012, an index-based insurance is being developed to estimate and monitor grassland production in France (Roumiguié *et al.*, 2015). By comparing the cumulative data of a year with a reference, currently the Olympic average of the previous five years, it is possible to calculate a loss rate. To validate the veracity of this index, several scientific studies have been carried out with different protocols (Roumiguié *et al.*, 2016). This time, it was compared with data on grass height and thus the production of grazed pasture. The objective of this study is to validate the values of the Grassland Production Index (GPI) by comparing them to the annual variations of the production derived from in situ measurements collected on wide range of growth conditions.

Materials and methods

The grassland production index is based on images with a spatial resolution of 250 and 500 m from the satellite Terra. Optical images are taken at a daily frequency. The productions of grassland were estimated through the empirical model proposed by Roumiguié *et al.* (2015), by deriving the GPI from medium resolution satellite images and climatic data. The approach was based on the sum of a biophysical variable (i.e. the daily values of the fraction of vegetation cover (F_{COVER})) observed throughout the grass growing season, and taking into account efficiency-reducing factor related to temperature and water stress. The time series of Fcover were obtained using the Overland image processing software developed by Airbus Defence and Space (Poilvé, 2010), which considered reflectance acquired by MODIS as input variable of an inversion scheme of a radiative transfer model. The size of a pixel is 600×600 m^2, in order to have information only on grassland areas, they make it match with two land-cover maps databases (i.e. the Corinne land cover and the French 'Registre Parcellaire Graphique') and apply a disaggregation step to obtain a grassland-specific Fcover.

In situ measurements were made with a manual rising plate meter (Jenquip, New Zealand) or an automatic one (True North Technologies, Ireland), depending on the farm.

The study was carried out on 9 experimental farms spread over the French territory in various pedoclimatic contexts (oceanic, continental, and mountainous) and with different types of production (dairy cattle, beef cattle, sheep, dairy goats).

On each farm, grass height measurements were carried out from the start of grass growth between February and April, depending on the farm, until November at the latest, with a pause in the summer during prolonged droughts. Measurements were taken at a weekly frequency with a minimum of 30 measurements per hectare. Depending on the farm, the measurements of the increase in grass height between two dates was used to calculate biomass using a density table. This density was adjusted for each farm following the protocol described by Defrance *et al.* (2004).

The study was conducted between 2016 and 2018 for the nine farms, and six farms had a fourth year of measurement in 2019. In total, more than 630 paddocks were measured, generating more than 14,000 grass growth data points. The sum of the weekly grass growth was used to calculate average annual production for each farm. The grassland production index was computed on the same period as grass height measurements occurred, giving a single value per farm per year.

For insurance purposes, the variation in production between years was compared with a reference. The reference was the average annual grass production on each farm for the years studied.

Results and discussion

Between farms and years, the median annual production was 6.1 t dry matter (DM) ha^{-1}, from *in situ* measurements. The year 2019 had the lowest production, with a median value of 5.1 t DM ha^{-1}, against 7.5, 6.3, and 5.6 t DM ha^{-1} for 2016, 2017 and 2018. The lowest herbage production measured on a single farm was 3.2 t DM ha^{-1} in 2019, and the highest herbage production was 11.6 t DM ha^{-1} in 2017. The variation in grassland production assessed on the nine farms over three years of measurements shows a very strong correlation between the two methods, giving an R^2 of 0.81. When the correlation is studied on the six farms where four years of measurements were carried out, the R^2 is slightly reduced to 0.7 (Figure 1). Roumigué *et al.* (2015) found R^2 of between 0.71 and 0.9 on a different dataset.

The four years had very different grass growth dynamics. Between farms, annual grass production varied between -38% and +55% compared to their three- or four-year average production. The grassland production index predicted these variations without bias towards one extreme.

In this context, the use of a grassland production index based on medium resolution satellite imagery is considered to be consistent. However, this medium spatial resolution and its aggregation to a larger scale

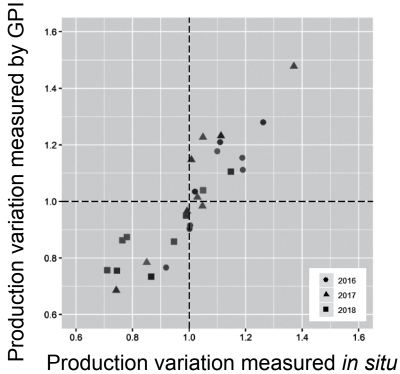

Figure 1. Annual production variation comparison for three years between GPI and grass measurements.

may hide undetected variations within the area. The effect of the farmer's practices is therefore erased by the GPI, whereas it is highlighted by the rising plate meter. Depending on the farm, the number of grid cells to monitor all the plots varied between one and four. A grid cell corresponds to the geographical area where historically the index follows the same variations, it has an average area of over 3,000 ha.

The use of a rising plate meter allows farmers to estimate the production of grasslands over a large area relatively quickly, but the method also has limitations to establish a reliable herbage production reference (Matthieu and Fiorreli, 1985).

Conclusions

The correlation of the results is considered satisfactory in view of the natural variability of the phenomenon studied. Therefore, the grassland production index could be safely used for insurance, although the difficulty of establishing a reference dataset remains the main limitation of this type of study. The prospect of an accurate monitoring of grassland biomass in real time by satellite remote sensing is strengthened and opens the door to an automatic and homogeneous evaluation system at national level for grazing management decision making tools.

References

Defrance P., Delaby L. and Seuret J.M. (2004) Mieux connaître la densité de l'herbe pour calculer la croissance, la biomasse d'une parcelle et le stock d'herbe disponible d'une exploitation.*Rencontres Recherches Ruminants* 11, 291-294.

Lepoivre B. (2020) L'assurance des prairies. *Fourrages* 244, 87-92.

Matthieu A. and Fiorelli J.L. (1985) Utilisation d'un herbomètre pour l'interprétation du déroulement du pâturage. *Fourrages* 101, 1-29.

Poilvé H. (2010) *Towards an operational GMES land monitoring core service - BIOPAR Methods Compendium – MERIS FR Biophysical*, Geoland2: Immenstaad, Germany, 45 pp.

Roumiguié A., Jacquin A., Sigel G., Poilvé H., Lepoivre B. and Hagolle O. (2015) Development of an index-based insurance product: validation of a forage production index derived from medium spatial resolution fCover time series. *GIScience & Remote Sensing* 52:1, 94-113.

Roumiguié A., Sigel G., Poilvé H., Bouchard B., Vrieling A. and Jacquin A. (2016) Insuring forage through satellites: testing alternative indices against grassland production estimates for France.*International Journal of Remote Sensing* 38(7), 1912-1939.

Viability of *Trifolium* seed following *in sacco* degradation

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Abstract

A substantial proportion of the improved permanent pastures found across the UK were last reseeded many decades ago. Over time the grasses and legumes originally planted have been replaced by unsown species, leading to a decline in pasture and stock performance. Sowing forage legumes into such swards can lead to substantial improvements in both nutrient supply and nutrient use efficiency. An alternative approach to mechanical seeding could be to feed clover seeds to stock. Using the dacron bag technique, we assessed the viability of seeds of varieties of white clover (*Trifolium repens*; n=8) and red clover (*Trifolium pratense*; n=8) following degradation in the rumen. Most seeds germinated within the rumen, regardless of clover species or variety. For red clover, only seeds from the variety Sangria continued to grow, and the proportion of these relative to those incubated was low (0.22). Almost all the white clover varieties had seeds grow on, but the proportions relative to those that germinated were variable and generally low. The highest proportions were recorded for the varieties AberAce (0.60), AberDai (0.46) and Reisling (0.46). The between-variety variation in seedling performance suggests that improved seedling resilience could be selected for as part of breeding programmes targeting more sustainable, multi-functional grasslands.

Keywords: clover, reseeding, digestion, sward improvement

Introduction

From the 1950s to the 1970s there was a concerted policy push across the UK incentivising farmers in less favoured areas to replace native grassland with more productive sown pastures. While some swards have been reseeded again since, the majority of these pastures have not. Over time the grasses and legumes originally planted have been replaced by unsown grasses and other species, leading to a long-term decline in both pasture and stock performance (Yu *et al.*, 2010). Many of the species and varieties of grass used in the seed mixtures sown were heavily reliant upon substantial quantities of inorganic fertilizers being added to remain competitive, and thus these detrimental changes in sward performance and nutritional value have been accelerated by reductions in fertilizer application rates in response to economic and environmental pressures (Yu *et al.*, 2010). Complete replacement of permanent pastures is economically costly and risks soil and carbon loss. Over-seeding by direct drilling or broadcast sowing is more commonly undertaken, but related costs can still be high and establishment rates can be variable, especially on low-nutrient status soils.

An alternative approach to introducing legumes that had localised popularity decades ago was to feed clover seeds to sheep. Having passed through the digestive tract the seeds would then be deposited within the animals' faeces. The faeces were expected to not only supply nutrients to the seeds as they germinated, but also offer some protection from grazing as the seedlings established. However, very few studies have been published in the scientific literature relating to the degradability of seeds in the gastrointestinal tract, and those that have relate to species of plant and growing conditions that are not commonly found in the UK or similar temperate areas (Cocks, 1988; Ghassali *et al.*, 1998; Grande *et al.*, 2016; Thomason *et al.*, 1990). They do, however, report that there can be considerable variation in recovery rates for different varieties of the same legume species. This project tested the viability of seeds of different varieties of white clover (*Trifolium repens*) and red clover (*Trifolium pratense*) following *in sacco* ruminal degradation.

Materials and methods

Different species/varieties of *Trifolium* were tested for survival following incubation in the rumen using the Dacron bag technique. Different varieties each of white and red clover were tested. The varieties assessed were: AberAce (*T. repens*), AberDai (*T. repens*), AberLasting (*T. repens*), AberSwan *(T. repens*), Alice (*T. repens*), Aran (*T. repens*), Coolfin (*T. repens*), Iona (*T. repens*), Klondike (*T. repens*), Reisling (*T. repens*), AberChianti (*T. pratense*), AberClaret (*T. pratense*), Amos (*T. pratense*), Atlantis (*T. pratense*), Magellan (*T. pratense*), Merviot (*T. pratense*), and Sangria (*T. pratense*). The experiment used 5 rumen-fistulated cows. Replicate (n=5) samples of 10 g of each variety of clover seed were weighed out and placed in Dacron bags with a pore size of 40 μm. All 17 varieties were then incubated in each animal (giving a total of 85 bags for the experiment). Following 24 h of incubation the bags were removed. Each bag was soaked in water for 5 min, then rinsed under a running tap for around 1 min. The bags were dried for 30 min at room temperature, before 10 seeds were selected at random for germination testing. The germination tests were carried out using trays with 25 wells arranged in a 5×5 design. One plate was used for each variety. A total of 10 seeds were taken from each Dacron bag and plated out in 5 wells (2 seeds per well). Each row of 5 wells corresponded to one cow. In total 50 seeds per variety were plated out. Approximately 1 ml of tap water was then added. Once prepared, the plates were covered to reduce evaporation and placed in a location where they received around 10 h of light per day at a temperature of approximately 20 °C. Following standard clover testing protocols, the number of germinated seeds after 3 days was recorded. This process was then repeated after 7 and 10 days. The number of seeds showing evidence of further growth (i.e. lengthening of the radicle and emergence of the plumule) was also recorded at this time. Following an angular transformation of the data, statistical comparisons were made using one-way ANOVA (Genstat 18; VSN International Ltd) with species as treatment.

Results and discussion

When the dacron bags were removed after 24 h it was found that a high proportion of seeds of varieties of both white and red clover had already started to germinate within the rumen, and in most cases there was little difference in germination proportion after 10 d compared to 3 d post removal and plating out (Table 1). It was observed that variability in germination, particularly of white clover seeds, appeared to be linked to seed colouring: white seeds readily germinated; yellow, orange and brown were slower; and dark brown seeds appeared impermeable (N. Gordon, personal observation). This requires further testing.

As well as noting germination rates for the seeds that had been incubated in the rumen, the percentage of seeds showing evidence of continued growth was recorded after 10 d. Although most of the red clover seeds had germinated, only seeds from the variety Sangria continued to grow. In contrast, significantly more of the white clover seeds continued to grow (P=0.004); notably AberAce and AberDai and Riesling. While it is likely that the process of being handled, washed and plated will have had an impact on seed viability to some extent, it seems there are substantial between-variety differences in response to exposure to the rumen environment.

Conclusions

There are species and varietal differences in the response of clover seeds when exposed to *in sacco* ruminal degradation. Based on these results there is little expectation that feeding white or red clover seeds would currently be a viable alternative to mechanised seed introduction at the current time. However, the between-variety variation in seedling performance suggests that improved seedling resilience could be selected for as part of breeding programmes targeting more sustainable, multi-functional grasslands.

Table 1. Germination rates (%) of different varieties of white and red clover following incubation within the rumen for 24 h, plus percentage showing continued growth after 10 d.

	Percentage of seeds germinated			Percentage of seeds growing
	After 3 days	After 7 days	After 10 days	
Trifolium pratense				
AberChianti	82	85	85	0
AberClaret	89	91	91	0
Amos	93	94	94	0
Atlantis	92	92	92	0
Magellan	95	95	95	0
Merviot	77	80	80	0
Sangria	83	83	83	22
Trifolium repens				
AberAce	98	98	98	60
AberDai	96	98	98	46
AberLasting	90	91	91	10
AberSwan	92	94	94	18
Alice	90	90	90	0
Aran	96	98	98	6
Coolfin	66	66	66	14
Iona	76	76	76	30
Klondike	91	91	91	10
Reisling	100	100	100	46

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References

Cocks P.S. (1988) Seed production and seed survival under grazing of annual medics (*Medicago* spp.) in North Syria. *Journal of Agricultural Science* 110, 455-463.

Ghassali F., Osman A.E. and Cocks P.S. (1998) Rehabilitation of degraded grasslands in north Syria: The use of Awassi sheep to disperse the seeds of annual pasture legumes. *Experimental Agriculture* 34(4), 391-405.

Grande D., Mancilla-Leyton J.M., Vicente A.M. and Delgado-Pertinez M. (2016) Can goats disperse seeds of herbaceous pasture plants in Mediterranean grasslands? *Small Ruminant Research* 143, 67-74.

Thomson E.F., Rihawi S., Cocks P.S., Osman A.E. and Russi L. (1990) Recovery and germination rates of seeds of Mediterranean medics and clovers offered to sheep at a single meal or continuously. *Journal of Agricultural Science* 114, 295-299.

Yu Y.W., Fraser M.D. and Evans J.G. (2010) Long-term effects on sward composition and animal performance of reducing fertilizer inputs to upland permanent pasture. *Grass and Forage Science* 66, 138-151.

Crop nitrogen balance in dairy feeding systems in the north-west of Spain

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Abstract

Crop nutrient balance – the difference between the input of nutrients to cropland and the amount removed by crops – is an indicator of agricultural sustainability. The aim of this study was to determine and compare crop nitrogen (N) balance and efficiency at farm level, according to the different feeding systems used on dairy farms in Galicia (NW Spain): grazing (G), grass silage (GS), maize and grass silage (MGS) and maize silage (MS). Intensification of the systems increases in the order G, GS, MGS and MS, with increased stocking rate, milk production (with no differences between MGS and MS systems) and concentrate intake per cow (differences between MS and G systems). The inputs indicated that more nitrogen was supplied by slurry as the system intensified. Supplies of organic N accounted for, on average, 60% of the inputs, and supplies of mineral N accounted for 28% of the inputs. N input, N output, N balance and NUE did not differ between feeding systems. Better nitrogen use is required to improve N balance and NUE, i.e. slurry application with minimal volatilization of ammonia and applied when required by the crop, which would significantly reduce the amounts of mineral fertilizers required.

Keywords: nitrogen surplus, inputs, outputs, organic fertilizers, slurry

Introduction

Calculation of crop N-balance and crop N-use efficiency takes into account N input via fertilizers, soil supplies and N fixation and N output via uptake by crops (De Klein *et al.*, 2016). Crop nutrient balance is an important indicator of agricultural sustainability, because surplus nutrients can pollute soil, water and air (Leip *et al.*, 2015) and indicate economic inefficiency. However, most nitrogen balance studies are based on field experiments, which do not represent the conditions in real farm systems in relation to crop rotation, fertilization and yield (Chmelíková *et al.*, 2021).

The aim of this study was to determine and compare crop N-balance and crop nitrogen-use efficiency (NUE) at farm level according to the different feeding systems used on dairy farms in Galicia (Botana *et al.*, 2018).

Material and methods

A total of fourteen dairy farms in Galicia were selected to represent the four types of feeding systems used in the region: grazing (G n=3); grass silage (GS n=3); maize and grass silage (MGS n=4); and maize silage (MS n=4).

The farms were surveyed to determine stocking rate, milk production, feed concentrates, agricultural area, forage crop distribution (grazing grassland, silage grassland, grazing plus silage grassland, maize in rotation with winter crops, maize and other), percentage of legumes in grasslands, organic and mineral fertilization (dose and management) and average yield for each crop. For each farm, the nutrient balance was calculated per hectare of each forage crop. The N inputs were determined from the doses of mineral and organic fertilizers applied (excreta when grazing, solid manure and slurry) and the respective N contents. Symbiotic N fixation by legumes was also calculated (Bossuet *et al.*, 2006). The N output included removal of N by crops determined from crop yield and N content (Piñeiro and Pérez, 1992;

García *et al.*, 2012). Ammonia volatilization was determined according to organic fertilizer management, weather condition and application method (García *et al.*, 2018). Finally, the N balance was established for an average hectare on the farm, according to the area occupied by each crop. NUE was defined as N output in relation to N input. Data were analysed by one-way ANOVA.

Results and discussion

Intensification of the feeding systems increased in the order G, GS, MGS and MS, with increases in the stocking rate (no differences between G and the GS and MGS systems), milk production (no differences between MGS and MS systems) and intake of concentrates per cow (differences between MS and G systems) (Table 1).

Regarding the N balance (Table 1), as the systems intensified, more N input was supplied by slurry, with differences between MS and the G and GS systems and between MGS and GS systems, N input of excreta was higher in the G than in the other systems and N input of mineral fertilizers was lower in the MGS. The organic N supplied accounted for, on average, 60% of the inputs, and the mineral N accounted for 28%. N input, N output, N balance and NUE did not differ among the systems. A huge individual variability was found both between and within the feeding systems, indicating the large margin for improvement on many of the dairy farms. The N balance and NUE values are within the range reported in other European studies (Aarts *et al.*, 2000; Chmelíková *et al.*, 2021; Oenema *et al.*, 2012), except for two farms in the MS and two farms in the MGS system, in which N balance was higher, with 226, 256, 216 and 404 kg N ha^{-1}, and NUE was lower, with 44, 49, 44 and 34%, respectively.

The key change that will increase NUE and reduce surplus N is improving nitrogen use by recycling within the system, mainly slurry (De Klein *et al.*, 2016), minimizing volatilization of ammonia, which accounts for an average of 48% of the organic N applied, and applying slurry when required by the crop, which would enable a significant reduction in the amount of mineral fertilizers required.

Table 1. Characteristics of dairy feeding systems in Galicia, including crop N balance and crop NUE.[1]

	G	GS	MGS	MS	Mean	SG[2]
No. cows	56±17[6]	62±7	83±33	210±187	109	NS
Agricultural area (ha)	40.3±13.4	49.4±14.6	50±21.3	96.5±88.9	61.1	NS
Livestock unit ha^{-1}	1.7±0.1 bc	1.6±0.2 c	2.2±0.5 b	2.8±0.3 a	2.2	***
L cow^{-1} $year^{-1}$	5,985±1,207c	8,307±1,011b	10,309±1,486a	10,985±941a	9,146	**
Concentrates (kg cow^{-1} $year^{-1}$)	1,599±369b	2,860±1,214ab	3,020±457ab	3,964±1,353a	2,951	*
N input (kg N ha^{-1})	349±54	325±34	378±96	440±120	378	NS
Slurry	85±29 c	147±88 bc	243±108 ab	290±86 a	202	*
Manure	0±0	2±3	1±3	7±8	3	NS
Excreta	67±18 a	18±19 b	10±15 b	0±0 b	21	***
Mineral fertilizers	117±13 a	111±22 a	71±5 b	127±19 a	106	**
Symbiotic fixation	80±24	45±61	52±25	16±30	47	NS
N ouput (kg N ha^{-1})						
Crop removal	231±11	204±19	237±40	206±33	220	NS
Crop N balance (kg N ha^{-1})	118±43	121±17	141±117	234±117	158	NS
Crop NUE[7] (%)	67±7	63±2	67±24	49±12	61	NS
Ammonia volatilization (kg N ha^{-1})	78±19	80±34	107±56	152±63	108	NS

[1] Values are Mean value ± standard deviation; G = grazing; GS = grass silage; MGS = maize and grass silage; MS = maize silage; NUE = nitrogen use efficiency.

[2] SG = significance: *** $P<0.001$, ** $P<0.01$, * $P<0.05$ and NS = non-significant;;

Ammonia volatilization can be minimized by slurry application methods such as injection at crop establishment, rather than the prevalent splash plate application, and incorporation in the following hours (survey data) and by application to pastures and grasslands using methods such as trailing hose and trailing shoe rather than splash plate (survey data).

Conclusions

Crop N-balance and crop NUE balance did not differ among the different feeding systems used on dairy farms in Galicia. More N input was supplied by slurry as the systems intensified. The huge individual variability among and within the feeding systems indicates a large margin for improvement on many of the dairy farms. Improvement of N balance and NUE requires better N use, by applying slurry (main N input) using methods that reduce volatilization of ammonia and when required by the crop, which would significantly reduce the need for mineral fertilizers.

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References

Aarts H.F.M., Habekotte B. and van Keulen H. (2000) Nitrogen management in the De Marke dairy farming system. *Nutrient Cycling in Agroecosystems* 56, 231-240.

Bossuet I., Chambaur H., Le Gall A. and Raison C. (2006) Study of the distribution of N surplus in experimental dairy farms of the Atlantic Area. In: Jarvis S. and Plimlin A. (eds.) *Proceedings of the final Seminar of Green Dairy Project: Interreg Altantica Area III B N°100.* Institut de l'Élevage, Rennes, France, pp. 67-96.

Botana A., Resch-Zafra C., Pereira Crespo S., Veiga M., González L., Dagnac T., Lorenzana R., Fernández-Lorenzo B. and Flores-Calvete G. (2018) Contrasting diets and milk composition on Galician dairy farms. *Grassland Science in Europe* 23, 730-732.

Chmelíková L., Schmid H., Anke S. and Hülsbergen K.J. (2021) Nitrogen-use efficiency of organic and conventional arable and dairy farming systems in Germany. *Nutrient Cycling in Agroecosystems* 119, 337-354.

De Klein C., Monoghan R.M., Alfaro M., Gourley C., Oenema O. and Powell J.M. (2016) Realistic nitrogen use efficiency goals in dairy production systems: a review and case study examples. In: *Solutions to improve nitrogen use efficiency for the world.* Proceedings of the 2016 INI Conference, Melbourne, Australia.

García M.I., Báez D., Castro J. and Louro A. (2012) Ajuste de las recomendaciones de fertilización en el maíz forrajero para las condiciones gallegas. *Afriga* 97, 130-136.

García M.I., Báez D., Gilsanz. C. and García V. (2018) Mejora de la eficiencia de la fertilización nitrogenada en los cultivos forrajeros. *Vaca Pinta* 2, 116-126.

Leip A., Billen G., Garnier J., Grizzetti B., Lassaletta L., Reis S., Simpson D, Sutton M.A., De Vries W., Weiss F. and Westhoek H. (2015) Impacts of European livestock production: Nitrogen, sulphur, phosphorus and greenhouse gas emissions, land-use, water eutrophication and biodiversity. *Environmental Research Letters* 10, 115004.

Oenema J., van Ittersum M. and van Keulen H. (2012) Improving nitrogen management on grassland on commercial pilot dairy farms in the Netherlands. *Agriculture, Ecosystems & Environment* 162, 116-126.

Piñeiro J. and Pérez M. (1992) Especies pratenses y modo de aprovechamiento I. Efecto sobre el contenido en proteína bruta, fósforo y potasio. XXXIII Reunión Científica de la SEEP, Pamplona, España, pp. 255-260.

Economic and environmental performance of French dairy farms through the scope of three farm economic strategies

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Abstract

Three main farm strategies are often observed among dairy farmers as a way to achieve high economic efficiency: *costs reduction*, *productivity*, and *value addition*. This paper aims to explore the link between economic and environmental performances through the scope of these three main farm strategies. The three groups have been put together using a hierarchical clustering analysis on a sample of 650 French dairy farms from 2009 to 2017. The economic and environmental data have allowed the comparison of performances across the three studied farm strategies. Findings show that the *value addition* strategy performs significantly better than the others, both in terms of economics and environmental issues. Also, the *cost reduction* strategy performs significantly better than the *production* strategy on the two aspects. It appears that the presence of grassland in the fodder system is key in the characterization of the three strategies and on the influence on both performances.

Keywords: dairy farming, farm strategies, fodder system, LCA, economic performance

Introduction

When analysing the environmental impacts of diverse French dairy farms, there is a consensus in the literature to analyse results according to fodder systems and production conditions (grass vs maize, plain vs mountain) (Dollé *et al.*, 2013a, 2013b). This paper takes a different prospective by proposing a methodology to identify economic strategies in which a dairy farm positions itself relative to others. In fact, during the past decade economic difficulties such as the milk price crisis, new CAP agreements and rising farm input prices have led French dairy farms to choose different strategies in order to be economically viable (Fagon *et al.*, 2017). This paper develops a methodology to reconstruct the three strategies of *costs reduction* (CR), *productivity* (P), and *value addition* (VA) and compares their practices and environmental performances.

Data and method

Technical and economic data are from the INOSYS network for the years 2009 to 2017, which are stored in the software DIAPASON® (Reuillon *et al.*, 2012). Environmental data for each farm are calculated through a life cycle inventory performed by using available data of the technical and economic dataset and based on the CAP'2ER® life cycle assessment (LCA) model (Moreau *et al.*, 2016). A number of missing life cycle inventory items were replaced by hypothetical standards in line with CAP'2ER® database. The classification of farms into the strategies needs to be done relatively to pairs. Thereby, a factorial analysis of mixed data (FAMD) is performed for each of the 9 years, using 6 structural and economic variables marked with an asterisk in Table 1. Based on this FAMD, a hierarchical cluster analysis differentiates three classes of dairy farms so that the inter-class variability is maximized and the intra-class variability is minimized through the R package FactoMineR. Finally, the analysis of the statistical significance of the differences between the three strategies' environmental performances (Table 2) is performed through a two-factor ANOVA test (year and strategy) combined with a Tukey test for every pair of samples.

Table 1. Description of the three farm strategies. Mean of the 9 annual means 2009-17 (Mean of standard error).

		Value addition	Cost reduction	Productivity
	Average number of farms per year	**55**	**85**	**82**
Structure	Forage and grassland area (ha)	84 (41)	78 (36)	66 (27)
	Work units (WU)	1.9 (0.8)	1.8 (0.7)	2.0 (0.8)
	Dairy livestock units (LU)	92 (41)	98 (37)	107 (42)
	Work productivity (l WU^{-1}) *	160,000 (62k)	250,000 (83k)	347,000 (114k)
	Land productivity (l ha^{-1} of forage area) *	4,100 (1,300)	6,100 (2,000)	9,800 (3,200)
Economic	Price of sold milk € 1000 l^{-1} *	478 (119)	339 (23)	333(17)
	engaged in official quality and origin signs *	96.7%	3.6%	5.1%
	Husbandry, veterinary, bedding costs € LU^{-1} *	186 (65)	178 (45)	255 (61)
	Cost of the fodder system € LU^{-1} *	1,040 (297)	995 (191)	1,331(223)
	Dairy activity level: production cost € 1000 l^{-1}	508 (134)	368 (68)	369 (58)
Practices	Livestock density (dairy LU ha^{-1} of forage area)	1.1 (0.3)	1.4 (0.4)	1.7 (0.5)
	Grassland/forage area (%)	95% (8%)	78% (16%)	62% (15%)
	Volume of milk per dairy cow (l)	5,687 (1,075)	7,007 (1,069)	8,450 (844)
	Concentrates for dairy cow (g l^{-1})	196 (77)	209 (73)	234 (63)
	Autonomy in protein (%)	79% (13%)	69% (12%)	55% (10%)
	Mineral fertilisation kg N ha^{-1} dairy area	12 (18)	52 (30)	79 (38)
	Time at grazing (milking herd) (days $year^{-1}$)	175 (48)	142 (55)	118 (61)

Table 2. Environmental performances.[1]

	VA	CR	P
GHG emissions (kg CO_2-eq l^{-1} FPCM)	0.98^a	0.98^a	1.00^c
GHG emissions – SOCS by permanent grassland (net kg CO_2-eq l^{-1} FPCM)	0.64^a	0.80^b	0.92^c
N balance – kg N ha^{-1}	60^a	95^b	125^c
Potential nitrogen loss to the water kg N ha^{-1}	24^a	34^b	42^c
Potential nitrogen loss to the air kg N ha^{-1}	19^a	48^b	77^c
Direct and indirect energy use MJ l^{-1} FPCM	2.6^a	2.5^a	2.9^c

[1] Different letters in a row indicate a significant difference α=0.05.

Results and discussion

First, the clustering seems to be robust since the three observed groups are consistent through time and 82% of the farms stay in the same group across the 7 years. Farms that opted for the VA strategy are smaller both in terms of land, production and labour, and achieve economic efficiency through high milk prices. CR farms are between the two other groups in terms of size, and differentiate themselves with very low operational expenses per means of production. P strategy relies on returns to scale, by maintaining a high production of milk per hectare and worker despite higher costs.

Different economic strategies imply different practices. VA farms opt for, on average, a low livestock density. The labels they are engaged in (51% of the farms are in organic farming, 46% in a protected designation of origin label, and 9% transform their own milk) might restrict the fodder system so that they have a higher share of grasslands and a low production of milk per cow, and thus a higher protein autonomy. This management is made possible thanks to the price premium. P farms show a higher density of livestock, and higher level of milk production per cow, which stems from a diet based on maize and concentrates and limited grazing. CR farms are in between the two groups in these aspects. They do not opt for a more extensive management such as the VA group because the lower price of milk they would receive cannot compensate for the reduction of milk production and higher costs.

As regards environmental results, the VA and the CR groups emit on average less greenhouse gases (GHG) per litre of fat and protein-corrected milk (FPCM) than the P group, but the difference between the VA and CR groups is not statically significant. The three strategies significantly differ in terms of the net carbon footprint per litre of milk, which considers soil organic carbon sequestration (SOCS) by permanent grasslands (570 kg C ha^{-1} y^{-1}). Similarly, nitrogen balances and potential losses to the air and water significantly differ across the groups. As for energy consumption, the P farms consume more fossil fuel to produce the same quantity of milk than their pairs, since the LCA accounts for the indirect use of energy through the purchase of farm inputs.

The figures obtained are in line with the literature on French dairy farms: grass-based dairy systems (represented here by VA and CR farms) show lower risk of nitrogen losses (Dollé *et al.*, 2013a) and have a lower net carbon footprint when taking SOCS into account (Dollé *et al.*, 2013b). However, results in this paper suggest that maize-based dairy systems do not emit less GHG per litre of milk if they do not adopt a CR strategy that limits their use of inputs. Productivity per animal and hectare should not be a goal *per se* in order to improve the economic and environmental performances of a dairy farm because it is not compatible with an optimal valorization of grass and grazing (Delagarde, 2009), which are virtuous for both performances (Peyraud *et al.*, 2014).

Conclusions

This paper proposes a robust methodology to classify French dairy farms into three strategies. Results suggest that grass-based systems achieve the best environmental performance, and go through a VA strategy when it is possible to valorise product through a label, or through a CR strategy when not. The fodder system turns out to be central between economic and environmental performance in dairy production, with optimal grassland management being key to achieve both.

References

Delagarde R. (2009) Outils et indicateurs pour calculer et concilier ingestion des vaches laitières et valorisation de l'herbe au pâturage. *Fourrages* 198, 175-190.

Dollé J.B., Delaby L., Plantureux S., Moreau S., Amiaud B., *et al.* (2013a) Impact environnemental des systèmes bovins laitiers français. *INRA Productions Animales* 26(2), 207-220.

Dollé J.B., Faverdin P., Agabriel J., Sauvant D. and Klumpp K. (2013b) Contribution de l'élevage bovin aux émissions de GES et au stockage de carbone selon les systèmes de production. *Fourrages* 215, 181-191.

Fagon J., Caillaud D., Seegers J. and Dockes A.C. (2017) Les éleveurs bovins lait face aux crises et aux aléas. Regards sur la résilience des exploitations du réseau INOSYS. *Collection Théma*. Idele.

Moreau S., Brocas C. and Dollé J.B. (2016) CAP'2ER®, the environmental footprint calculator and decision making for ruminant production systems. *Book of abstracts of the 10th International Conference on Life Cycle Assessment of Food, 19-21 October 2016*, Dublin, Ireland, pp. 1223-1225.

Peyraud J.L., Delaby L., Delagarde R. and Pavie J. (2014) Les atouts sociétaux et agricoles de la prairie. *Fourrages* 218, 115-124

Reuillon J.L., Fagon J., Charroin T. and Monique L. (2012) Coût de production en élevage bovin lait. Manuel de référence de la méthode proposée par l'Institut de l'Elevage. *Collection Résultats.* Idele. 46p.

Legacy effects in a grassland-crop rotation enhanced by legume content

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Abstract

Crop-grassland rotations are common in European crop-livestock systems, and we investigated how best to maximize legacy effects from pasture leys within a rotation. In a three-year grassland–crop rotation experiment, we manipulated plant diversity and experimental drought in a grassland phase (with 150 kg ha^{-1} N fertilizer, plus a 300 kg ha^{-1} N perennial ryegrass monoculture comparison), sprayed off the grassland, and sowed a follow-on crop of Italian ryegrass (IRG) to measure legacy effects of the preceding grassland ley as yield of IRG. Legume species produced the highest legacy effect (>6 t ha^{-1} of IRG); the lowest (4.5 t ha^{-1}) was from perennial ryegrass monoculture, and even lower (4.2 t ha^{-1}) with higher fertilizer rate. Chicory delivered the highest non-legume legacy effect. There was no synergistic effect of mixing species on the legacy effect. Drought generally reduced the legacy effect by 0.4 t ha^{-1}. Multi-species grassland communities with high legume content at lower fertilizer input delivered higher full rotation yield than a rotation based on a highly fertilized grass monoculture. We conclude that higher legume proportion in diverse swards is crucial for higher legacy effects on crop yield, but plant diversity underpins the productivity and sustainability of grassland-crop system.

Keywords: legacy effect, rotation, multi-species, legume, drought

Introduction

In mixed farming systems, grassland and crops are often alternated in rotations. We know that grassland leys can be beneficial to a follow-on crop through legacy effect (Crotty *et al.*, 2016), but the effect of grassland management (e.g. plant diversity, fertilizer use) on the subsequent crop is poorly understood (but see Fox *et al.*, 2020). In previous work, multi-species mixtures of grass, legume and herb species enhanced grassland yield and mitigated drought effects (Grange *et al.*, 2021). We investigate whether this benefit of plant diversity extends to the follow-on crop in a rotation. Our hypothesis is that diverse swards deliver higher legacy effects than monocultures, as a residual effect of synergistic interactions between grassland species during the ley phase. In addition, we expect to have a neutral effect of a two-month summer drought on the legacy effect, because grasslands often show a rapid recovery of yield after disturbance (Haughey *et al.*, 2018).

Materials and methods

A 3-year rotation experiment was established in 2017 at Johnstown Castle, Wexford (south-east of Ireland). In the first two years, a grassland ley phase was cultivated intensively. Plant diversity was manipulated using a six species pool from grass, legume and herb functional groups (FG), to create communities of 1, 2, 4, 5 and 6-species following a simplex design. Aboveground biomass was harvested seven times a year and annual fertilizer rate was 150 kg N ha^{-1} yr^{-1} (150N). A *Lolium perenne* monoculture receiving double nitrogen fertilizer (300N) was included as a low-diversity, high-input comparison. A total of 39 plots were subdivided with two treatments randomly assigned to each half: rainfed control or a two-month simulated drought to reach extreme level of stress (soil Water potential below -1.5 MPa for more than a month; see Grange *et al.* (2021) for more details).

The grassland plots were terminated in 2020, and an Italian ryegrass (*Lolium multiflorum*) model crop was established, while keeping the same plot layout. All plots (including plots that were fertilized at a higher rate in the grassland phase) were fertilized with 40N, harvested on four occasions throughout the year and no drought treatment was applied. Thus, all plots were managed uniformly, so the differences in crop yield translate into differences in grassland legacy effect.

Data were analysed using a Diversity-Interactions model, adapted from Kirwan *et al.* (2009) to predict crop dry matter yield (DMY). Plant diversity, drought and fertilizer were used as explanatory variables, with diversity being defined by species identity and interaction effects, weighed by species proportions. More information about the methods and results can be found in (Grange *et al.*, unpublished data).

Results and discussion

After a model selection process, we found no evidence of interspecific interaction effects among grassland species on the legacy effect, and therefore no overyielding in the follow-on crop (Table 1). Thus, the effects of different species combinations of grassland leys on yield of Italian ryegrass (the legacy effect) was estimated from linear combinations of the species' identity effects (e.g. the estimated legacy effect for the equi-proportional 6-species mixture was the average of the identity effects of the six species).

We speculate that this effect is the outcome of opposing processes occurring in mixtures. On one side, diverse swards produced more biomass in the grassland phase (Grange *et al.*, 2021), and thus may have delivered higher organic matter to the follow-on crop through a greater biomass of soil organic matter (roots and dead material). On the other side, complementarity in resource acquisition from mixtures may be the reason for higher export of nutrients from the system. These antagonistic effects could result in the neutral interactions. A similar study by Fox *et al.* (2020) with ley phase fertilized with 200 kg N ha^{-1} yr^{-1}, found a strong synergistic interaction between grassland legume and non-legume species in the delivery of legacy effects. Our divergent results suggest that we currently have too few sites to generalize about residual interaction effects between grassland species on the follow-on crop performance.

Among the different species studied, the legumes delivered the highest legacy effect (Table 1). For example, a former grassland of *Trifolium repens* resulted in Italian ryegrass DMY of 6.5 t ha^{-1}. The 300N *L. perenne* had the lowest legacy effect with 4.2 t ha^{-1} of Italian ryegrass DMY. The 300N comparison delivered the lowest legacy effect (Table 1); we suspect that the extra fertilizer applied in grassland phase was either exported as forage, or lost through leachates or N_2O emissions (Guo *et al.*, 2010). The 6-species

Table 1. Model estimates of the dry matter yield (DMY) of *L. multiflorum* for the growing season (four harvests) of the follow-on crop, regressed on the species sown proportions (labelled by species name) in the preceding grassland ley communities.[1]

Model estimates	DMY (t ha^{-1})
Lolium perenne	4.5±0.20
Phleum pratense	5.1±0.19
Trifolium pratense	6.1±0.20
Trifolium repens	6.5±0.20
Cichorium intybus	5.5±0.20
Plantago lanceolata	4.8±0.20
300N *L. perenne*	4.2±0.20
Drought	-0.4±0.09
6-species mixture	5.4±0.08

[1] Shown here are the separate effects of previous grassland plant diversity, fertilizer level and drought treatment. Drought had a significantly negative constant effect, and affected the yield of each community with the same magnitude.

grassland mixture legacy effect was significantly higher than those of the 150N or 300N *L. perenne* monoculture. This comparative benefit was stimulated by the proportion of legume in the mixture. Herbs had similar impacts on the legacy effect as grasses (Table 1). We conclude that diversity through legume inclusion in grassland swards is a promising tool to enhance follow-on crop productivity and reduce reliance on nitrogen fertilizer.

The effect induced (on the follow-on crop) by the former drought treatment (in the grassland phase) significantly reduced Italian ryegrass DMY. Interestingly, all communities were affected similarly (Table 1) and there was no evidence of interactions between, for example, species identity effects and drought. A consequence of such a drought effect on the grassland legacy effect is that although rapid recovery after drought can occur (Haughey *et al.*, 2018), persistent effects of the disturbance not captured within the growing season it occurred may also arise. This suggests the need for longer-term assessment of the effect of extreme weather events on farming systems.

Conclusions

This experiment showed the importance of considering the design of grassland leys as part of a rotational system. Plant diversity enhanced yield of the grassland ley, and was not associated with lower follow-on crop performance. In contrast, increased fertilizer use did not enhance the legacy effect on the follow-on crop (no significant difference between *L. perenne* and 300N *L. perenne* in Table 1). This suggests that the extra nitrogen applied was lost from the system. Legume species proportion was the main driver of the follow-on crop yield, and thus should be favoured in grassland leys to increase legacy effects for follow-on crops.

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References

Crotty F.V., Fychan R., Sanderson R., Rhymes J.R., Bourdin F., Scullion J. and Marley C.L. (2016) Understanding the legacy effect of previous forage crop and tillage management on soil biology, after conversion to an arable crop rotation. *Soil Biology and Biochemistry* 103, 241-252.

Fox A., Suter M., Widmer F. and Lüscher A. (2020) Positive legacy effect of previous legume proportion in a ley on the performance of a following crop of Lolium multiflorum. *Plant and Soil* 447, 497-506.

Grange G., Finn J.A. and Brophy C. (2021) Plant diversity enhanced yield and mitigated drought impacts in intensively managed grassland communities. *Journal of Applied Ecology* 58, 1864-1875.

Guo R., Nendel C., Rahn C., Jiang C. and Chen Q. (2010) Tracking nitrogen losses in a greenhouse crop rotation experiment in North China using the EU-Rotate_N simulation model. *Environmental Pollution* 158, 2218-2229.

Haughey E., Suter M., Hofer D., Hoekstra N.J., McElwain J.C., Luscher A. and Finn J.A. (2018) Higher species richness enhances yield stability in intensively managed grasslands with experimental disturbance. *Science Reports* 8, 15047.

Kirwan L., Connolly J., Finn J.A., Brophy C., Luscher A., Nyfeler D. and Sebastia M.T. (2009) Diversity-interaction modeling: estimating contributions of species identities and interactions to ecosystem function. *Ecology* 90, 2032-2038.

Faba bean silage as a substitute for grass silage in dairy cow diets

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Abstract

Faba bean (*Vicia faba*) is an interesting alternative feed that can be used both as whole crop silage and concentrate to provide energy and protein to ruminants. However, faba bean protein is low in methionine (Met). The aim of this study was to investigate how the partial replacement of grass silage by faba bean silage in faba bean seed-supplemented dairy cow feeding affects the performance, and whether a rumen protected Met could improve the utilization of faba bean protein in milk production. Two dairy cow experiments were conducted. In the first study, the rumen protected Met increased energy corrected milk and protein yields when faba bean was included into the diet both as forage and as concentrate. However, Met supplementation was ineffective in diets with grass silage as the only forage. In the latter experiment, the inclusion of faba bean silage increased feed intake, energy corrected milk, and protein yield compared to pure grass silage and decreased ruminal methane production relative to dry matter intake by 10%.

Keywords: faba bean, methionine, methane, dairy cow

Introduction

Domestic legumes with N-fixing ability are of particular interest in European animal production owing to the rapidly rising prices of inorganic N fertilizers and the low self-sufficiency in protein feeds. Faba bean (*Vicia faba*) is an interesting alternative for dairy cow feeding both as whole crop silage and as concentrate, due to the high biomass potential of the crop and the high protein and starch content of the seed. However, faba bean protein is known for its high rumen degradability and low Met content that may limit the production responses (Puhakka *et al.,* 2016; Halmemies-Beauchet-Filleau *et al.,* 2018). The aim of this study was to investigate how the partial replacement of grass silage by faba bean silage in faba bean seed-supplemented dairy cow feeding affects feed intake, milk yield and ruminal methane emission. In addition, it was investigated whether a rumen-protected Met supplement could improve the utilization of faba bean protein in milk production. We hypothesized that: (1) milk production response of faba bean protein can be improved by rumen-protected Met supplement irrespective of basal forage species; and (2) replacing grass silage with faba bean silage decreases ruminal methane emissions due to faba beans' inherently higher starch and lower fibre content.

Materials and methods

Two dairy cow experiments were conducted at the University of Helsinki during autumn 2019. The first study was designed as a replicated 4×4 Latin Square with 21-d periods and 2×2 factorial arrangement of treatments. The first factor was total mixed ration (TMR) of forage species and the second factor was rumen-protected Met supplement. Experimental animals were kept in tie stalls and were 4 primiparous (averaging 31 days in milk) and 4 multiparous (averaging 181 days in milk) Nordic Red dairy cows. Forage was either pure grass silage (*Phleum pratense* and *Schedonorus pratensis*, D-value 664 g kg^{-1} dry matter (DM)) or a silage mixture where two thirds of grass silage DM was replaced by faba bean silage (D-value 593 g kg^{-1} DM). Concentrate (400 g $kg^{-1}$of TMR DM) contained barley, oats and faba beans (311 g kg^{-1} of concentrate DM) as protein source. Total mixed rations were offered *ad libitum.* Rumen-protected Met (Smartamine M, Kemin Europa, Herentals, Belgium; 20 g d^{-1} absorbed in the small intestine) was offered mixed with 1 kg d^{-1} molassed sugar beet pulp during milkings.

In the second experiment, all Nordic Red cows of the free stall barn (n=34-38 depending on calvings and drying-offs) were fed a TMR based on grass silage for 4 weeks. The forage was then switched to a silage mixture where two-thirds of grass silage DM was replaced by faba bean silage for another 4 weeks after which all cows returned to the original grass silage based diet for 4 weeks. The silages were the same as in the first experiment. The experimental TMRs were offered *ad libitum.* They contained (350 g kg^{-1} DM) concentrate that was composed of oats, barley, pea (50 g kg^{-1} of concentrate DM) and faba beans (200 g kg^{-1} of concentrate DM) as the main protein source. The cows visited freely milking robot (Lely Astronaut A3, Lely International, Maassluis, the Netherlands) equipped with Greenfeed system (C-Lock Inc, Rapid City, USA) that measures ruminal methane emissions during the visits. Standard concentrate composed mainly of barley, wheat, molassed sugarbeet pulp and rapeseed meal was distributed to the cows at the milking robot according to their milk yield (4.6-9.0 kg d^{-1}). Only the cows (n=15; averaging 60 days in milk) that had 10 or more methane recordings from the last week of each experimental period were accepted to methane production data. Furthermore, eight multiparous cows averaging 36 days in milk were sampled more intensively for milk and faeces.

In both experiments, chemical analysis were conducted as described by Puhakka *et al.* (2016). The first data were statistically analysed by ANOVA with a model that included the fixed effects of treatment, square and period within a square, and a random effect of a cow within a square (SAS 9.4.). The orthogonal contrasts were used to compare the effects of (1) forage species, (2) the use of rumen-protected Met, and (3) their interaction. The latter data were analysed by ANOVA with a model that included the fixed effect of treatment and the random effect of a cow (SAS 9.4.) for (1) linear and (2) quadratic response.

Results and discussion

In the first experiment, replacing two thirds of grass silage DM with faba bean silage increased DM intake (P<0.01; Table 1) in line with previous legume silage studies (Steinshamn *et al.,* 2010; Lamminen *et al.,* 2015). Rumen protected Met decreased milk yield when the forage was purely grass silage, but had no effect on milk yield, when faba bean silage was dominating the forage (P=0.04 for interaction). Irrespective of forage species, Met decreased milk lactose concentration (P<0.01), but increased milk protein (P<0.01) and fat concentrations (P=0.06). Increases in milk protein (Halmemies-Beauchet-Filleau *et al.,* 2020) or milk fat concentration (Varvikko *et al.,* 1999) are typical responses to increased Met supply. Energy corrected milk yield was increased with Met supplement on diets based on a mixture of faba bean and grass silage, but decreased when based on grass silage only (P<0.03 for interaction). It seems that mammary gland Met supply was limiting milk synthesis, when the dairy cow diet contained a large proportion of faba bean. This is supported by lower plasma Met concentrations on diets with faba bean as major feed constituent compared to grass silage diets (P<0.02).

The second experiment confirmed the good palatability and high milk production potential of the faba bean silage. Indeed, the inclusion of faba bean silage into forage increased (P<0.03) DM intake (26.7 vs 30.2 kg d^{-1}) and as a consequence energy corrected milk yield (42.2 vs 46.0 kg d^{-1}) and protein yield (1.40 vs 1.59 kg d^{-1}). Despite lower D-value of faba bean silage relative to grass silage, the whole tract digestibility of neutral detergent fibre was higher (P<0.01) with diets containing faba bean silage. Forage species did not affect the total ruminal methane production, which averaged 488 g d^{-1}. However, methane production relative to feed intake decreased (P=0.04) with the inclusion of faba bean silage into forage, from 20.0 g to 18.0 g methane kg^{-1} DM eaten.

Table 1. The effect of forage species and rumen protected methionine on dairy cow performance (first experiment)

	Experimental diets					Significance		
	Grass	Grass + Met	Faba bean	Faba bean + Met	SEM	Grass vs faba bean	Met	Interaction
Dry matter intake, kg d^{-1}	21.2	21.3	23.0	22.3	0.52	<0.01	0.36	0.29
Plasma Met, µmol l^{-1}	22.2	52.2	19.8	42.3	2.54	0.02	<0.01	0.12
Yield, kg d^{-1}								
Milk	26.0	24.2	25.2	25.3	1.20	0.77	0.04	0.04
Energy corrected milk	28.3	26.8	27.4	29.0	1.02	0.34	0.99	0.03
Yield, g d^{-1}								
Lactose	1,139	1,045	1,110	1,092	57.6	0.59	<0.01	0.04
Fat	1,216	1,152	1,166	1,277	52.4	0.43	0.63	0.08
Protein	953	915	931	969	26.7	0.30	0.99	0.03
Concentration, g kg^{-1}								
Lactose	43.9	43.1	43.9	43.3	0.47	0.58	<0.01	0.78
Fat	46.8	47.8	46.6	50.5	1.75	0.33	0.06	0.25
Protein	36.7	37.9	37.2	38.9	1.18	0.17	<0.01	0.62
Urea, mg dl^{-1}	26.2	28.1	33.8	33.1	1.58	<0.01	0.41	0.09

Conclusions

Faba bean silage increased DM intake in both experiments when mixed together with grass silage. In the latter experiment, this increase was large enough to improve energy corrected milk yield. In addition, ruminal methane production relative to DM intake decreased by 10% with the inclusion of faba bean silage into forage. When the dairy cow diet contained faba bean, both as protein feed and as a major part of the forage, rumen-protected Met increased energy corrected milk yield and protein yield. However, in diets based on grass silage only, rumen-protected Met supplementation was ineffective.

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References

Halmemies-Beauchet-Filleau A., Rinne M., Lamminen M., Mapato C., Ampapon T., Wanapat M. and Vanhatalo A. (2018) Alternative and novel feeds for ruminants: nutritive value, product quality and environmental aspects. *Animal* 12(s2), 295-309.

Halmemies-Beauchet-Filleau A., Jaakkola S., Kokkonen T. and Vanhatalo A. (2020) Type of protein supplementation on dairy cow performance on grass silage-based diet. *Grassland Science in Europe* 25, 264-266.

Lamminen M., Kokkonen T., Halmemies-Beauchet-Filleau A., Termonen T., Vanhatalo A. and Jaakkola S. (2015) Partial replacement of grass silage with faba bean whole-crop silage in the diet of dairy cows. *Grassland Science in Europe* 20, 446-448.

Puhakka L., Jaakkola S., Simpura I., Kokkonen T. and Vanhatalo A. (2016) Effects of replacing rapeseed meal with fava bean at 2 concentrate crude protein levels on feed intake, nutrient digestion, and milk production in cows fed grass silage-based diets. *Journal of Dairy Science* 99, 7993-8006.

Steinshamn H. (2010) Effect of forage legumes on feed intake, milk production and milk quality – a review. *Animal Science Papers and Reports* 28, 195-206.

Varvikko T., Vanhatalo A., Jalava T. and Huhtanen P. (1999) Lactation and metabolic responses to graded abomasal doses of methionine and lysine in cows fed grass silage diets. *Journal of Dairy Science* 82, 2659-2673.

Spatial distribution of virtually and physically fenced cattle in relation to forage availability

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Abstract

Grazing causes disturbance to the grass sward which is used as an indicator for management decisions based on herbage disappearance. Cattle grazing on a pasture move primarily to identify feeding stations that fulfil their daily dietary requirements. However, possible stress caused by new virtual fencing technology could also affect daily movement. This study used virtually or physically fenced Fleckvieh heifers to evaluate movement behaviour and effects on the grass sward on rotationally grazed permanent grassland. Pre- and post-grazing images from an unmanned-aerial-vehicle (UAV) were used to create digital orthophotos of the grass canopy to assess the influence of grazing livestock on the vegetation in relation to local positions as determined from GPS-signals. The GPS-positions were logged (one-minute-intervals) by virtual-fencing-collars (Nofence, Batnfjordsøra, Norway) and rasterized in a 2.5×2.5 m grid. It was found that the calculated Red-Green-Blue Vegetation Index obtained through UAV imagery and its change during grazing is correlated to spatial cattle positions. Consequently, the index has potential to be used as a proxy of grass sward disturbance. There was no indication of an impact of the fencing system on the daily movement of cattle.

Keywords: cattle grazing behaviour, remote sensing, cattle motion behaviour, Fleckvieh

Introduction

Grazing is a worthwhile form of grassland management due to its low costs (Isselstein *et al.*, 2005) and potential biodiversity benefits (Tälle *et al.*, 2016). Reconciling agronomic and ecological goals requires control of access of grazing livestock to the pasture at a high spatial and temporal resolution. In practice this is achieved by fencing. Virtual fencing has the potential to fulfil these aims better than physical fencing (Stevens *et al.*, 2021) as virtual fences are flexibly adaptable to a wide range of needs. Using GPS logged collar-data to localise grazing animals helps to identify frequently used areas on pastures or avoided ones, and this information can be combined with unmanned-aerial-vehicles (UAV) based geoinformation. We tested whether information obtained from virtual fencing collars and UAVs can be used to evaluate disturbance to the grass sward in relation to spatial behaviour of cattle on rotationally grazed pastures of varying forage availability. Furthermore, we investigated whether the fencing system influences the movement behaviour of the cattle.

Materials and methods

The present study was part of a larger grazing trial conducted on permanent grassland at the experimental farm of the University of Goettingen in Relliehausen, Solling Uplands, Lower Saxony, Germany, from July to September 2021. The trial was approved by the animal welfare service of LAVES (Lower Saxony State Office for Consumer Protection and Food Safety – ref. Number: 33.19-42502-04-20 / 3388).

Treatments consisted of two forage availabilities (adequate (A) vs surplus (S)) and two fencing systems (virtually (VF) vs physically (PF)). Each forage availability (determined beforehand) × fencing system combination was assigned to one 2-ha pasture area and grazed with eight Fleckvieh heifers per group, blocked according to age and liveweight (12-15 months, 308-463 kg initial weight). Paddocks were stocked for 3-4 days with rotation lengths of 15 days each resulting in four paddocks grazed in total per treatment-group (two rotations). The heifers were equipped with Nofence collars (Nofence®, AS, Batnfjordsøra Norway) which recorded minute-wise GPS-positions and with IceTag accelerometers (Icerobotics Ltd, Edinburgh, Scotland) to measure lying time. In the present study, data within the second rotation and from the second and the fourth paddock (26-30 August and 2-6 September) were analysed. Pre- and post-grazing images from an UAV (DJI Phantom 4) were used to create orthomosaics with the structure-from-motion software Agisoft Metashape. The Red-Green-Blue Vegetation Index (RGBVI) and the difference of the RGBVI between pre- and post-grazing were calculated. All spatial measurements refer to 2.5×2.5 m grid-cells arranged within paddocks. Spatial distribution of cattle refers to the sum of seconds spent active (lying time was excluded) within each grid cell as derived from GPS positions and expressed as the Camargo's index of evenness, which is a measure of spatial distribution (Payne *et al.*, 2005). Values near zero indicate a patchy distribution and values near one a more even distribution. An increasing value should mean higher movement behaviour of the cattle. Lying time and walking distance were used to check the validity of the Camargo's index and to evaluate possible differences between the treatments. Statistical analyses were carried out with the software R (version 4.1). Linear-mixed effects models were applied to determine lying time and walking distances, stocking within paddock with the fixed effects of forage availability (two levels), fencing system (two levels) and day within paddock (three levels from day two to four). The individual animal served as a random effect. The first and the last day within paddock were excluded to avoid bias. For the Camargo's index, paddock (two levels) was used as a random effect instead of individual animal, since it refers to the sum of duration within grid cells across animals. Model reduction was performed from the global model using the MuMIn package and the most parsimonious model with the smallest AICc was chosen as final model.

Results and discussion

The Camargo's index of evenness was affected by the main effect of day within paddock (F=8.1, P=0.0027). On the first measurement day (day two) within paddock the heifers' distribution was more patchy 0.24±0.05 than on day three 0.43±0.03 and four 0.45±0.02 (estimated mean ± standard error (SE)). The RGBVI change within grid cell was correlated to the accumulated active time (s) per grid cell (Figure 1) and showed lower values at adequate (A) than surplus (S) (not shown). On the first day, the heifers obviously invested less time searching for feed items than on the last day, and therefore the walking distances (estimated mean ± SE) were the lowest (first day: A: 3,217±98.3; S: 3,338±103.7 m, last day: A: 3,733±99.6; S: 3,373±103.3 m). On the first day they only needed a few feeding stations to fulfil their nutritional requirements irrespective of forage treatment and fencing system. Yet, there was a significant interaction of forage availability × day within paddock for lying time (F=12.3, P<0.0001) and walking distance (F=17.8, P<0.0001). The heifers in S lay the longest (776±19.5 min), the heifers in A lay the shortest time (612±13.4 min) and walked the longest distance (3,733±99.6 m) (estimated mean ± SE) on the last measured day of stocking (day four). It can be assumed that the animals in A had to walk longer distances to find the appropriate feed (see also Hamidi *et al.,* 2021) and had thus shorter lying time on day four. The fencing system had no significant effect on any of the target variables.

Conclusions

Forage availability seems to be the relevant determinant of cattle movement, as shown by the livestock GPS data and the related RGBVI changes. The GPS data from the VF collars in combination with the UAV data offer a future perspective for an improved grazing management that better allocates the grazing livestock to the pasture resources. Digital technologies and virtual fencing in particular can significantly improve the implementation of grazing management decisions.

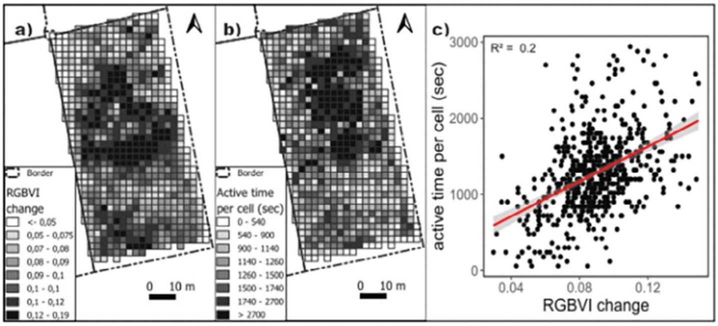

Figure 1. Example of the (A) RGBVI changes, (B) summed active time (s) of all heifers of the group per grid cell and (C) the correlation of both with each other for the treatment physical fenced heifers (forage availability: adequate) during the last stocking (02/09/2021 – 06/09/2021).

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References

Hamidi D., Komainda M., Tonn B., Harbers J., Grinnell N.A. and Isselstein J. (2021) The effect of grazing intensity and sward heterogeneity on the movement behaviour of suckler cows on semi-natural grassland. *Frontiers in Veterinary Science* 8, 639096.

Isselstein J., Jeangros B. and Pavlů V. (2005). Agronomic aspects of biodiversity targeted management of temperate grasslands in Europe – a review. *Agronomy Research* 3, 139-151.

Payne L.X., Schindler D., Parrish J.K. and Temple S.A. (2005) Quantifying spatial pattern with Eveness Indices. *Ecological Applications* 15 (2), 507-520.

Stevens D.R., Thompson B.R., Johnson P., Welten B., Meenken E. and Bryant J. (2021) Integrating digital technologies to aid grassland productivity and sustainability. *Frontiers in Sustainable Food Systems* 5, 602350.

Tälle M., Deák B., Poschlod P., Valkó O., Westerberg L. and Milberg P. (2016) Grazing vs. Mowing: A Meta-Analysis of Biodiversity Benefits for Grassland Management *Agriculture, Ecosystems & Environment* 222, 200-212.

Effects of electrical impulses on cattle grazing behaviour: virtual vs physical fencing

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Abstract

Pastures fenced with electric wire fences are common practice for grazing and these physical fences emitting electrical impulses are largely accepted. However, the upcoming technology of virtual fencing (Nofence, Batnfjordsøra, Norway) raises animal welfare concerns because electrical impulses are emitted by a collar. In this study, we compared groups of growing heifers (Fleckvieh) grazing virtually (VF) or physically fenced (PF) pastures, respectively, to investigate the time needed to recommence grazing after having received an electrical impulse. The two fencing system groups VF vs PF, with four heifers each, were grazed in adjacent paddocks (1000 m^2 each) for 12 days in three successive periods. Reactions to electrical impulses from the VF-collar (n=156) in the VF-group or from the physical-fence in both groups (n=93) were retrieved from observational data and internal-data-loggers in the VF-group. A generalized-least-squares model showed significantly (P=0.015) shorter time until restart of grazing after having received an electrical impulse by collar compared to an impulse by the wire fence (22.0±2.6 vs 33.6±4.2 s. (estimated means ± standard error, respectively). As cattle returned to grazing faster after a collar-impulse, the electric impulses by the VF-collars seem to be less disruptive to the animals than the PF-impulses.

Keywords: Fleckvieh beef heifers, smart farming technology, grassland management

Introduction

The development of virtual fences opens up many perspectives to simplify and improve common grazing systems. In this respect, reduced labour input for fencing or the protection of environmentally sensitive areas are some well-recognized features (Campbell *et al.,* 2019). Undisturbed animal welfare is a basic prerequisite for the implementation of new technologies, such as virtual fences, where animals are allocated an area of pasture between invisible borders. Within virtual fences, electrical impulses are emitted through the collar if animals cross the pre-defined virtual border irrespective of acoustic warning signals. The application of electric collar impulses, however, raises animal welfare concerns. Grazing is the main behaviour of cattle on pasture (Kilgour, 2012) and the time spent grazing is used as an important indicator for the animals' welfare on pasture. The objective of this study is, therefore, to assess the time elapse between an electric collar impulse and restart of grazing, and to compare this against time elapse after an electric impulse from the physical fence.

Materials and methods

The study was conducted from August to September 2020 at the experimental farm of the University of Goettingen in Relliehausen, Solling Uplands, Lower Saxony, Germany (51°46'55.9'N, 9°42'11.9'E; 250 m above sea-level) and was split into three subsequent periods of 12 days each, serving as study periods by means of replication (17-28 August, 31 August–11 September, 14-25 September). We examined the ability of Fleckvieh heifers to learn the virtual fencing (VF) system with Nofence collars (Nofence®, AS, Batnfjordsøra Norway) (pulse energy 0.2 Joule, duration=1.0 s) and compared their behaviour to a physically fenced (PF) group. Each group consisted of four Fleckvieh heifers per period (n=24 cattle in total, 14-16 months, 320-451 kg initial weight). The two groups grazed in adjacent paddocks (fenced with standard electric wire fence) on permanent grassland. The electric fence device was commercially available (® Siepmann, Herdecke, Germany) with a pulse energy of up to 4.1 joules (ex-device) at contact, varying according to electric wire conductivity and distance to the device itself. One GPS-coordinated VF-line separated the pasture of the VF-group into an exclusion and an inclusion zone. The pasture of the PF-group was divided by a PF-line (Figure 1A). One observer per group continuously recorded the behaviour of each of the four heifers per group during 4-h pasture access durations per day (change of observer to avoid bias per period). Data were retrieved from these observations (electric impulse from the physical fence) and the Nofence collar report (electric impulse from the VF-collar). In total, n=156 electric impulses from the VF-collars and n=93 electric impulses from the physical fence were recorded across periods and fencing system treatments. We used a generalized least squares model which regressed the time elapse between an electric impulse and recommencement of grazing on the effect of impulse type (VF vs PF). The severity of the impulse was consequently measured as duration of interruption of usual behaviour. Data were log-transformed before analysis in order to improve the normality of residuals.

Results and discussion

As far as we can tell, there is no knowledge yet on: (1) how the reactions of animals to electric impulses from collar compared to electric wire fences differ; and (2) how intensely the behaviour is affected after having received an electric impulse. We have approached these questions by comparison of the time needed after an electric impulse from the Nofence collar against an electric impulse of the physical fence until returning to grazing. The type of impulse received significantly influenced the time until grazing (P=0.015). After having received an electric impulse from the VF-collar, the time (estimated means ± standard error) until grazing was significantly shorter (22.0±2.6 s) than after an electric impulse from the physical fence (33.6±4.2 s) (Figure 1B). Thus, our data do not support the presumption that animal welfare is compromised in the VF system. An obvious advantage of the VF system is the always constant (in duration and strength) pulse energy that is emitted at the neck. The pulse energy of the physical fence likely varied in intensity in relation to the contact duration, fence wire conductivity and distance to the device, all of which determine the local charge of the fence at the contact point. Furthermore, the same physical stressor produces different effects, depending on whether its occurrence is predictable or not (Weiss, 1970). The electric impulse of the Nofence collar is likely predictable for the animal (after having learned the system) as it is always announced by an acoustic signal. If cattle can learn to avoid a suitable level of electrical stimulus it is likely not harmful for them (Lee *et al.*, 2008). We assume from our preliminary results that the electric impulse of the physical fence imposes more stress on the grazing animal than the electric impulse of the collar, as it caused a longer time elapse until grazing. To what extent

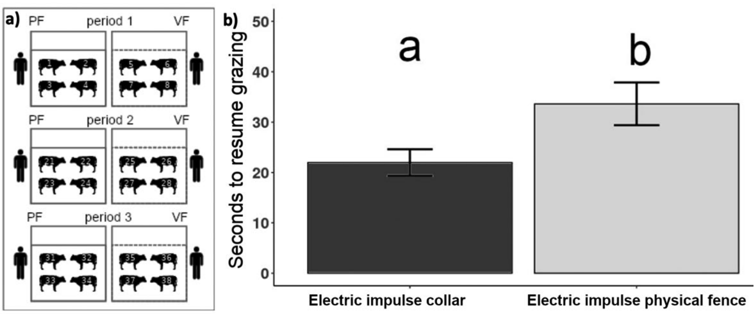

Figure 1. (A) Experimental setup with physically (PF) and virtually fenced (VF) Fleckvieh heifers; (B) Estimated means ± standard error of time (s) to resume grazing after having received an electric impulse from the Nofence® collar (only VF-group) or from the physical fence (both groups).

receiving electric impulses from the VF collar might be an improvement for animal welfare compared to the common PF electric impulses needs to be further evaluated in future.

Conclusions

After electric impulses emitted from the Nofence collar, cattle had a faster return to grazing than after contact with the physical fence. The combination of acoustic signalling followed by electric impulses when virtual barriers are crossed seems to make electric impulses more predictable for the cattle and reactions to the aversive stimulus smaller which indicated an improvement in animal welfare. It remains to be seen whether these preliminary results can be strengthened by further research in this area.

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References

Campbell D.L.M., Lea M.L., Keshavarzi H. and Lee C. (2019) Virtual fencing is comparable to electric tape fencing for cattle behaviour and welfare. *Frontiers in Veterinary Science* 6, 445.

Kilgour R.J. (2012) In pursuit of 'Normal': A review of the behaviour of cattle at pasture. *Applied Animal Behaviour Science* 138, 1-2.

Lee C., Fisher A.D., Reed M.T. and Henshall J.M. (2008) The effect of low energy electric shock on cortisol, β-endorphin, heart rate and behaviour of cattle. *Applied Animal Behaviour Science* 113, 1-3.

Weiss J.M. (1970) Somatic effects of predictable and unpredictable shock *Psychosomatic Medicine* 32 (4), 397-408.

Effect of concentrate supplement level and type on milk fat production in grazing dairy cows

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Abstract

The objective of this experiment was to investigate the effect of concentrate supplement level and type on milk fat production in early to mid-lactation, grazing dairy cows. Eighty Holstein Friesian dairy cows were blocked based on pre-experimental milk production and parity, and randomly assigned to 1 of 5 dietary treatments. The 5 dietary treatments were: a pasture (P) only control; P supplemented with 2 kg of dry matter (DM) cow^{-1} day^{-1} of an industry standard concentrate; P supplemented with 4 kg of DM cow^{-1} day^{-1} of an industry standard concentrate; P supplemented with 4 kg of DM cow^{-1} day^{-1} of a concentrate containing sodium hydroxide treated straw; and P supplemented with 4 kg of DM cow^{-1} day^{-1} of a concentrate containing rumen-protected fat. The experiment consisted of an initial 2-wk covariate period, 1-wk of diet acclimatisation and a 12-wk period of data collection. Overall, concentrate supplement level or type had no significant effect on milk fat concentration; however, there was a significant effect on milk fat yield. Results suggest that concentrate supplement level and type can influence milk fat yield but do not influence milk fat concentration in grazing dairy cows.

Keywords: dairy cow, milk fat, supplement, grazing

Introduction

Milk fat contributes substantially to the economic value of milk as it can be processed into a range of food ingredients such as butter, cheese, cream and whole milk powder. Milk fat is considered the most variable milk component with many nutritional and non-nutritional factors affecting the milk fat production of dairy cows. This provides opportunity for producers to increase the economic sustainability of their system. In an analysis of Irish Holstein-Friesian dairy herds, the greatest prevalence of a reduction in milk fat concentration occurred in the months of April or May, with 9.1% of herds recording a milk fat concentration <3.3% in May of 2014 (Carty *et al.*, 2017). Furthermore, the highest prevalence of a reduction in milk fat concentration occurred in April and May for both spring and autumn calving dairy cows, suggesting that time of year was more important than days in milk (Carty *et al.*, 2017). The association of a reduction in milk fat concentration with time of year could be related to environmental factors (e.g. day length) or nutritional factors (e.g. diet composition) that are associated with this risk period, which warrants further investigation. Therefore, the objectives of this experiment were to test the effect of: (1) increasing concentrate supplement level; and (2) concentrate supplement type on the milk fat production of grazing dairy cows during this high-risk period.

Materials and methods

The experiment was conducted at the Dairygold Research Farm (Teagasc Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland; 52°09'N; 8°16'W) between April and July 2021. Eighty Holstein Friesian dairy cows (55±15 days in milk and 483±50 kg of body weight) were blocked based on pre- experimental milk production and parity, and randomly assigned to 1 of 5 dietary treatments (n=16). The experiment consisted of an initial 2-wk covariate period, 1-wk of diet acclimatisation and a 12-wk period of data collection. The 5 dietary treatments were: a pasture (P) only control (P0); P supplemented with 2 kg of DM cow^{-1} day^{-1} of an industry standard concentrate (P2); P supplemented with 4 kg of DM cow^{-1} day^{-1} of an industry standard concentrate (P4); P supplemented with 4 kg of DM cow^{-1} day^{-1} of a concentrate containing sodium hydroxide treated straw (P4S); and P supplemented with 4 kg of DM cow^{-1} day^{-1} of a concentrate containing rumen-protected fat (P4F). The major ingredients included in the industry standard concentrates (P2 and P4) were on average: 200 g kg^{-1} maize meal, 150 g kg^{-1} barley, 150 g kg^{-1} maize gluten, 150 g kg^{-1} soya hulls, 90 g kg^{-1} beet pulp, 70 g kg^{-1} rapeseed meal, 70 g kg^{-1} molasses, 50 g kg^{-1} maize distillers and 30 g kg^{-1} soya hulls. The concentrates P4S and P4F were similar to the industry standard concentrates; however, either 100 or 50 g kg^{-1} of barley was replaced with sodium hydroxide treated straw or a rumen-protected fat ingredient, respectively. The rumen-protected fat ingredient was a Ca-salt (9% Ca and 84% fat) with the fat component comprising 58% palmitic acid, 28% oleic acid and 5% stearic acid. All cows grazed together as a single group and had *ad libitum* access to fresh water. Cows were allocated either a 24-h or 36-h residence time within each paddock or until a targeted post-grazing residual compressed sward height of 4 to 4.5 cm was achieved. Weekly milk production was determined from individual daily milk yield (kg), which was recorded using electronic milk meters (Dairymaster, Causeway, Co. Kerry, Ireland). Milk fat and protein concentrations were determined weekly from successive p.m. and a.m. milk samples using a Milkoscan FT6000 (Foss Electric). Weekly milk solids (kg fat + protein) were then calculated. Once-weekly body weight (BW) was recorded using an electronic scale and Winweigh software package (Tru-test Limited). Body conditioning score (BCS) was recorded weekly using a 1 to 5 scale (where 1 = emaciated and 5 = extremely fat) with 0.25 increments. All data were analysed in a repeated measures model using the MIXED procedure of SAS version 9.4.The model included fixed effects of treatment, week, their interactions and parity. Cow within treatment was included as the random effect with a covariate adjustment applied for each cow. The repeated measures analysis were based on week. Polynomial contrasts were included for evaluation of linear and quadratic responses to concentrate supplement level. Preplanned contrasts were included to evaluate the effect of standard concentrate vs altered concentrate ingredients [P4 vs (P4S + P4F) / 2] and concentrate ingredient type (P4S vs P4F). Significance was considered at $P \leq 0.05$, trends at $0.05 < P \leq 0.10$.

Results and discussion

Level of concentrate supplement linearly increased milk fat yield (Table 1; $P<0.01$); however, while there was a tendency, there was no significant effect on milk fat concentration ($P=0.08$). This is contrary to our first hypothesis that increasing the level of concentrate supplement would reduce milk fat concentration. In the literature, the effect of level of concentrate supplement on milk fat concentration is equivocal, as some studies observed no significant effect (McEvoy *et al.*, 2008) whereas others observed linear, negative responses (Delaby *et al.*, 2001). A number of factors could be responsible for this such as cow genetics, pasture chemical composition or concentrate ingredients (Wales *et al.*, 2009). In the current experiment, altering the concentrate ingredients [P4 vs (P4S + P4F) / 2] had no significant effect on milk fat concentration ($P=0.45$) or milk fat yield ($P=0.60$). When investigating the specific type of ingredient (P4S vs P4F), there was a significant effect on milk fat yield ($P=0.02$); however, there was no significant effect on milk fat concentration ($P=0.80$). As a result, the data also do not support our second hypothesis, that altering the concentrate type would increase milk fat concentration when compared with a standard concentrate. Interestingly, level of concentrate supplement increased milk protein concentration and milk protein yield (Table 1). In a review by Bargo *et al.* (2003), the authors reported an increase in milk protein concentration with increased levels of concentrate supplement, whereas McEvoy *et al.* (2008) found no difference. Altering the concentrate ingredient in the current experiment had no significant effect on milk protein concentration ($P=0.24$) or milk protein yield ($P=0.40$). However, cows fed P4F had reduced milk protein concentration ($P=0.04$) and tended to increase milk protein yield ($P=0.08$) when compared with cows fed P4S. There was no effect of concentrate supplement level or type on BW or BCS.

Table 1. Effect of concentrate supplement level and type on milk production, milk composition, BW and BCS in early-lactation grazing dairy cows.[1]

	Diet[2]						P-value[3]			
Items	**P0**	**P2**	**P4**	**P4S**	**P4F**	**SEM**	**Lin**	**Quad**	**Ingred**	**Type**
Milk yield, kg d^{-1}	22.2	24.0	25.4	25.3	26.8	0.45	<0.01	0.07	0.21	0.02
Fat, %	4.44	4.35	4.32	4.23	4.26	0.08	0.08	0.66	0.45	0.80
Protein, %	3.62	3.66	3.73	3.74	3.65	0.03	0.01	0.42	0.24	0.04
Fat, kg d^{-1}	0.98	1.05	1.09	1.07	1.14	0.02	<0.01	0.33	0.60	0.02
Protein, kg d^{-1}	0.80	0.88	0.95	0.94	0.98	0.02	<0.01	0.02	0.40	0.08
Milk solids, kg d^{-1}	1.78	1.92	2.03	2.01	2.11	0.03	<0.01	0.09	0.47	0.03
BW, kg	506	506	511	519	517	5.34	0.14	0.24	0.30	0.76
BCS	3.00	2.99	3.02	3.02	3.03	0.02	0.45	0.30	0.92	0.84

[1] BW = body weight; BCS = body condition score; SEM = standard error of the mean.

[2] P0 = pasture-only control; P2 = pasture + 2 kg DM concentrate supplement; P4 = pasture + 4 kg DM concentrate supplement; P4S = pasture + 4 kg DM concentrate supplement containing sodium hydroxide treated straw; P4F = pasture + 4 kg DM concentrate supplement containing rumen-protected fat.

[3] Lin = linear effect of concentrate supplement level; Quad = quadratic effect of concentrate supplement level; Ingred = P4 vs (P4S + P4F) / 2; Type = P4S vs P4F.

Conclusions

In this experiment, during the high-risk period for reduced milk fat concentration, concentrate supplement level and type had no effect on milk fat concentration. Further investigation is required to determine the factors responsible for reduced milk fat concentration during April and May in Irish grazing dairy cows.

Acknowledgements

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References

Bargo F., Muller L.D., Kolver E. S., and Delahoy J.E. (2003) Invited review: Production and digestion of supplemented dairy cows on pasture. *Journal of Dairy Science* 86, 1-42.

Carty C., Fahey A., Sheehy M., Taylor S., Lean I., McAloon C., O'Grady L., and Mulligan F. (2017) The prevalence, temporal and spatial trends in bulk tank equivalent milk fat depression in Irish milk recorded herds. *Irish Veterinary Journal* 70, 14.

Delaby L., Peyraud J.L., and Delagarde R. (2001) Effect of the level of concentrate supplementation, herbage allowance and milk yield at turnout on the performance of dairy cows in mid lactation at grazing. *Animal Science* 53, 171-181.

McEvoy M., Kennedy E., Murphy J., Boland T., Delaby L. and O'Donovan M. (2008) The effect of herbage allowance concentrate supplementation on milk production and dry matter intake of spring-calving dairy cows in early lactation. *Journal of Dairy Science* 91, 1258-1269.

Wales W.J., Kolver E.S., Egan A.R., and Roche J.R. (2009). Effects of strain of Holstein-Friesian and concentrate supplementation on the fatty acid composition of milk fat of dairy cows grazing pasture in early lactation. *Journal of Dairy Science* 92, 247-255.

Using white clover to reduce nitrogen fertilizer application – results from an eight-year study

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Abstract

White clover is the most commonly sown legume species in temperate grassland. It grows well in association with grass and is tolerant of grazing. There are several benefits associated with the use of white clover in grass-based milk production systems including nitrogen (N) fixation resulting in reduced requirement for fertilizer N, herbage production and quality, increased milk production and increased N-use efficiency. Eight years (2013-2020) of research at Teagasc, Moorepark, Ireland comparing the standard grass-only grazing system receiving 250 kg fertilizer N ha^{-1} with a grass-white clover system receiving 150 kg N ha^{-1} have been completed. Both systems were stocked at 2.74 cows ha^{-1}, and concentrate feeding was the same for both treatments (438 kg cow^{-1}). Herbage production was similar on the two sward types (13.5 t DM ha^{-1}), despite the 100 kg ha^{-1} reduction in N fertilizer used on the grass-clover swards. Average sward clover content was 22%. Milk yield was similar on the two treatments (6,200 kg cow^{-1} $year^{-1}$). Milk solids yield were greater ($P<0.05$) on the grass-clover system (510 kg cow^{-1} $year^{-1}$) compared to grass-only (490 kg cow^{-1} $year^{-1}$).

Keywords: white clover, milk production, herbage production, grazing

Introduction

White clover (*Trifolium repens* L.) is the most commonly sown legume species in grassland in Ireland and many temperate regions. It grows well in association with grass and is tolerant of grazing. Enriquez-Hidalgo *et al.* (2016) reported that, even with chemical nitrogen (N) application rates >150 kg N ha^{-1}, white clover can fix over N making it available for plant growth in intensively grazed swards (8-10 grazings per year; post grazing sward height ~4 cm). Milk production systems in Ireland are reliant on productive perennial ryegrass (*Lolium perenne* L.) swards receiving high inputs of chemical N fertilizer (up to 250 kg N ha^{-1} annually). Incorporating white clover in grassland swards offers a real opportunity to reduce chemical N fertilizer use. Other benefits associated with the use of white clover in grass-based milk production systems including herbage production and quality (e.g. Andrews *et al.*, 2007; Enriquez-Hidalgo *et al.*, 2018), increased milk production (e.g. Andrews *et al.*, 2007) and increased N-use efficiency (Hennessy *et al.*, 2020). The objective of this study was to compare herbage and milk production on from a grass-only grazing system receiving 250 kg fertilizer N ha^{-1} with that from a grass-white clover system receiving 150 kg N ha^{-1}.

Materials and methods

A full lactation farm systems experiment was undertaken over 8 years (2013 to 2020) at Teagasc, Moorepark, Fermoy, Co. Cork, Ireland. The study had two treatments: grass only swards receiving 250 kg N ha^{-1} (Grass250) and grass-white clover swards receiving 150 kg N ha^{-1} (Clover150). In February each year, 34-40 (depending on the year) spring calving dairy cows (Fresian and Fresian×Jersey) were selected and balanced on mean calving date, lactation number and pre-experimental milk yield milk solids yield and randomly allocated to one of the two treatments. Each treatment was stocked at 2.74 cows ha^{-1} in a closed farm system with cows staying in their treatment groups for the entire lactation each year. Farmlet size varied depending on number of cows available each year. Average concentrate supplementation was the same in each treatment and was 438 kg cow^{-1} fed throughout the lactation. Swards were rotationally grazed 8 to 10 times per year. Grass growth was recorded weekly in PastureBase Ireland (Hanrahan *et* *al.*, 2018). Sward clover content was measured prior to each grazing using the method described by Egan *et al.* (2017). Milk yield was recorded daily (Dairymaster, Causeway, Co. Kerry, Ireland) and milk composition (fat and protein concentrations) was measured weekly using MilkoScan 203 (Foss Electric, Hilerod, Denmark). Data were analysed in SAS using Proc Mixed with terms for treatment, year and associated interactions.

Results and discussion

There was no significant effect of treatment on total annual herbage production (13,431 kg dry matter (DM) ha^{-1}) or total silage conserved (2,703 kg DM ha^{-1}) despite the reduction in chemical N fertilizer applied to Clover150 (Table 1). Average annual sward clover content was 22%; greater than the 20% required to see an animal production benefit of including white clover in the sward (Andrews *et al.*, 2007). Sward clover content peaked in September at average of 35% (Figure 1).

Total annual milk production per cow was similar (6,189 kg cow^{-1}) (Table 1). Milk solids production per cow were significantly (P<0.05) greater for Clover150 (510 kg cow^{-1}) compared to Grass250 (490 kg cow^{-1}) (Table 1). Milk fat and protein contents were similar for both treatments (Table 1).

This study shows the potential to reduce chemical N fertilizer input by up to 40% (100 kg N ha^{-1}) without negatively impacting herbage or milk production in intensive pasture-based systems. This is particularly important as due increasing requirements on farmers to reduce chemical N fertilizer use to meet targets in reducing greenhouse gas emissions.

Table 1. Annual total herbage production, silage yield and milk and milk solids production for cows grazing grass-only swards receiving 250 kg N ha^{-1} and grass-white clover receiving 150 kg N ha^{-1}.

	Grass 250 kg N ha^{-1}	Grass-white clover 150 kg N ha^{-1}	SEM	*P*-value[1]
Total annual DM yield (kg DM ha^{-1})	13,467	13,396	397.5	ns
Annual silage yield (kg DM ha^{-1})	2,716	2,690	309.7	ns
Annual milk yield (kg cow^{-1})	6,068	6,311	253.2	0.0599
Annual milk solids yield (kg cow^{-1})	490	510	6.2	<0.05
Annual milk fat (%)	4.43	4.36	0.219	ns
Annual milk protein (%)	3.66	3.67	0.021	ns

[1] ns = not significant.

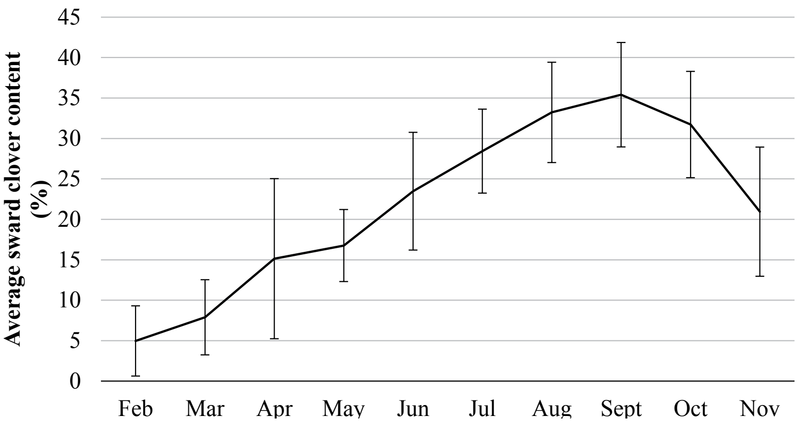

Figure 1. Average sward clover content from February to November on grass-white clover swards receiving 150 kg N ha^{-1} from 2013 to 2020.

Conclusions

This study shows that incorporating white clover into grass swards in an intensive grazing system allows chemical N fertilizer application to be reduced from 250 to 150 kg N ha^{-1} without negatively impacting annual herbage production. Cows grazing grass-white clover also had significantly greater milk solids yield compared to cows grazing grass-only.

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References

Andrews M., Scholefield D., Abberton M.T., McKenzie B.A., Hodge S. and Raven J.A. (2007) Use of white clover as an alternative to nitrogen fertiliser for dairy pastures in nitrate vulnerable zones in the UK: Productivity, environmental impact and economic considerations. *Annals of Applied Biology* 151, 11-23.

Egan M, Lynch M.B. and Hennessy D. (2017) Including white clover in nitrogen fertilized perennial ryegrass swards: effects on dry matter intake and milk production of spring calving dairy cows. *Journal of Agricultural Science* 155, 657-668.

Enriquez-Hidalgo D., Gilliland T. and Hennessy D. (2016) Herbage and nitrogen yields, fixation and transfer by white clover to companion grasses in grazed swards under different rates of nitrogen fertilization. *Grass and Forage Science* 71, 559-574.

Enriquez-Hidalgo D., Gilliland T.J., Egan M. and Hennessy D. (2018) Production and quality benefits of white clover inclusion into ryegrass swards at different nitrogen fertilizer rates. *Journal of Agricultural Science* 156, 378-386.

Hanrahan L., Geoghegan A., O'Donovan M., Griffith V., Ruelle E., Wallace,M. and Shalloo L. (2017) PastureBase Ireland: A grassland decision support system and national database. *Computers and Electronics in Agriculture* 136, 193-201.

Hennessy D., McAuliffe S., Egan M. and Ruelle E. (2019) What is the effect of incorporating white clover (*Trifolium repens* L.) into grazed grassland on the farm-gate nitrogen balance? *Grassland Science in Europe* 24, 143-145.

Comparing three methods to quantify fresh grass intake in grazing trials

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Abstract

For the assessment of fresh grass intake in grazing experiments various methods are available, which differ in reliability, but also in cost price. The alkane method is reliable but relatively expensive and labour intensive. In an ongoing grazing experiment, grass intake was estimated based on a (derived) calculation of: (1) energy coverage (Dutch VEM system); (2) mowing strips before and after grazing; and (3) cow sensor data. These methods were applied in: (1) a 24 h (strip) grazing system; and in (2) a system with limited (strip) grazing on two different grass lengths (short: 7-8 cm and long 15-17 cm). In this article we report a comparison of the three methods for the estimation of grass intake. Grass intake based on mowing showed overall the highest coefficient of variation (36.5%, compared with about 19% for the other two methods) and the highest estimated intake when there was a relatively large pasture residue. Estimation of grass intake based on data recorded at individual cow level gives more constant and less variable results.

Keywords: estimating fresh grass intake, comparing methods, energy coverage, sensor data, mowing strips

Introduction

In practice, the individual grass intake of dairy cows in the pasture is often unknown, and in research it is also difficult to estimate grass intake. Grass intake can be easily measured in stables, for example by using Roughage Intake Control (RIC) bins. The most reliable method for estimating fresh grass intake by grazing animals is to use markers, for example alkanes (Smit *et al.,* 2005). However, this method is expensive, labour-intensive and can cause discomfort to the animals due to frequent faecal sampling. For this reason alternatives are used, such as mowing grass strips or determining grass intake based on energy coverage. A new technique is based on a model which uses data of cow sensors combined with individual cow data (Schils *et al.*, 2019). The aim of this study was to compare the variability of three methods for estimating fresh grass intake, based on daily calculated fresh grass intake.

Materials and methods

To compare three methods to estimate grass intake, data collected during an ongoing grazing trial were used. This research was conducted at the research innovation centre Dairy Campus (Leeuwarden, the Netherlands). Grass intake was calculated for three treatment groups of 16 dairy cows each: unlimited one-day strip grazing (U) with an allowance of 20 kg DM per cow; limited grazing on short grass (8 cm, S); and limited grazing on long grass (15-17 cm, L), S and L both having an allowance of 9 kg dry matter (DM) per cow. Limited grazing consisted of 8 hours of grazing during the day and supplementary feeding with grass silage at night (16 hours) in an approximately 50:50 ratio. All three groups were supplemented with 5 kg of (the same) concentrates. Every morning a new grazing strip was offered. All cows were equipped with the Nedap Smarttag Neck sensor (Nedap, Groenlo, the Netherlands) to record time spent eating, ruminating, resting or active. The amount of supplementary feeding (grass silage) was recorded using RIC bins. The trial was conducted during three periods (spring, summer and autumn) of 14 days each. Three methods to estimate fresh grass intake were compared: energy coverage (Dutch VEM system), mowing strips and sensor data.

Mowing strips: the grass uptake in the pasture at herd level was determined using the difference between the pre- and post-grazing mass to calculate the total grass intake of the entire herd (as an average per cow at group level). The pre- and post herbage mass were expressed in kg DM per cow per day. To this end, a daily yield determination was done before and after grazing, by randomly mowing 5 strips of approximately 5 m, cut to 4 cm stubble height, then weighing and determining the dry matter content (oven-dried at 103 °C).

Energy coverage: using the Dutch Energy (VEM) system as described by Van Es (1975) and CVB (2016), the total energy requirement for lactating dairy cattle was calculated as well as the energy content of the diet. The energy unit VEM stands for Milk Feed Unit and is an energy parameter that indicates the net energy content of a feed material for lactating cows. The energy requirement of dairy cows was based on individual animal characteristics: body weight, days in lactation, parity, calving date and (fat and protein corrected) milk production. The complete model is described by CVB (2016). Using the VEM coverage, the fresh grass uptake from the paddock was estimated according to the formula:

$$Fresh\ grass\ intake\ (kg\ DM) = \frac{VEM\ requirement - VEM\ intake\ (grass\ silage + concentrates)}{VEMcontent\ of\ fresh\ grass}$$

The extent to which the total VEM requirement was fulfilled by grass silage was calculated (only for the cows in S and L) and also that from concentrates (all cows, 5 kg of the same product) of which the intake was measured directly. The remaining VEM requirement was assumed to be accounted for by fresh grass. By dividing this difference by the VEM value of the grass, the amount of fresh grass intake was estimated.

Sensor data: The cows were equipped with a Nedap Smarttag Neck sensor. In the years 2015 to 2017, within the Amazing Grazing project (Timmer *et al.*, 2016; Schils *et al.*, 2019), a model was developed to estimate grass uptake based on sensor data combined with individual cow data. The model included six predictors: eating time, number of steps, milk production, lactation number, supplemented silage and lactation stage, and was calibrated based on appropriate reference data collected with the alkane method. At group level this model achieved a RMSEP of 0.6 kg DM cow^{-1} (Schils *et al.,* 2019). This model was used to calculate fresh grass intake per individual cow.

For all three methods data were averaged per group per day and compared using ANOVA with method, treatment groups and period as treatment factors.

Results and discussion

Overall, the difference in grass uptake calculated according to the three methods was significantly different ($P<0.001$). The figures are shown in Table 1. Based on mowing strips, the average calculated intake was the highest, but the variation was also high due to large differences in the post-grazing mass. Because of this, especially in treatment L, the estimates of grass intake based on mowing strips were much higher than for the two other methods. The sensor data were calibrated in a limited grazing system; nevertheless, the differences between the methods were not larger in treatment U (unlimited grazing) than in the other treatments. To make a robust estimation of fresh grass intake we suggest that methods based on individual cow data are more robust than methods that collect data at herd level.

Conclusions

Determining grass intake based on mowing strips only gives an estimate at the herd level and this method resulted in the largest variation, compared with the other two methods applied in this experiment, especially with a large pasture residue. Estimation of grass intake based on data recorded at individual cow level gives a more constant and less variable outcome.

Table 1. Means, standard deviation (SD) and coefficient of variation (CV) of grass intake estimated with three methods by limited (L, S) and unlimited (U) grazing.

Treatment	**S**			**L**			**U**			**Mean**	
Method	**Mean**	**SD**	**CV**	**Mean**	**SD**	**CV**	**Mean**	**SD**	**CV**	**Intake**	**CV**
Energy cover	7.04	1.75	24.9	7.34	1.25	17.0	14.57	1.98	13.6	9.7	18.5
Mowing strips	6.03	2.7	44.8	10.39	3.17	30.5	13.46	4.57	34.0	10.0	36.4
Sensor	5.79	1.21	20.9	5.76	1.23	21.4	14.8	2.25	15.2	8.8	19.2

References

CVB (2019) *CVB Feed Table 2019*. Central Bureau of Livestock Feeding, Product Board Animal Feed, The Hague, the Netherlands

Schils R., Philipsen B., Hoekstra N., Holshof G., Zom R., Hoving I., Reenen K, Stienezen M., Klootwijk C., Van der Werf J., Sebek L., Van Eekeren N., Van Dixhoorn I. and Pol-van Dasselaar A. (2019) Amazing grazing: A public and private partnership to stimulate grazing practices in intensive dairy systems. *Sustainability* 11(20).

Smit H.J., Taweel H.Z., Tas B.M, Tamminga S. and Elgersma A. (2005) Comparison of techniques for estimating herbage intake of grazing dairy cows. *Journal of Dairy Science* 88, 1827-1836.

Timmer B., Zom R.L.G., Holshof G., Spithoven M. and Van Reenen C.G. (2016) The application of behaviour sensors and sward measurement to support grazing management. *Grassland Science in Europe* 21, 80-82.

Van Es A.J.H. (1975) Feed evaluation for dairy cows. *Livestock Production Science 2*(2), 95-107.

Vegetation indices obtained by UAV-mounted sensors to determine pasture biomass in a simulated grazing system

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Abstract

Different vegetation indices obtained with unmanned aerial vehicles (UAVs) and multispectral cameras have shown strong correlations with pasture biomass. However, complex photogrammetry processing remains a barrier to the utilization of this technology on-farm. Here, we describe the use of a UAV mounted with multispectral camera and radiometric calibration sensor to capture images required to generate a number of vegetation indices. UAV flights were conducted eight times throughout the 2021 grazing season, capturing data from 24 perennial ryegrass plots, which were managed to represent a range of biomass covers and regrowth stages evident within intensive rotational grazing systems. Following each UAV flight, platemeter measures of biomass, and total biomass yields (plot fresh weight and oven-dried DM%, 1 to 4 t DM ha^{-1}) were obtained. With the use of real-time kinematic (RTK) positioning, repeatable flights provided highly accurate location data, and rapid in-field generation of vegetation index values was achieved through the Pix4DFields software application. Non-linear regression analysis of the relationship between two vegetation indices (NDVI, GNDVI) and total biomass yield recorded across 6 harvests indicated a significant correlation (P<0.001) between grass cover (kg DM ha^{-1}) and both NDVI and GNDVI values, accounting for 68 and 69% of the variance, respectively.

Keywords: biomass, grass, NDVI, GNDVI, UAV

Introduction

There is significant interest in the utilization of remote sensing technology, including image capture by unmanned aerial vehicles (UAVs) to estimate pasture biomass (kg DM ha^{-1}) in agriculture. On temperate grassland farms, including in Northern Ireland, improved sustainability and profitability are driven by maximizing both pasture utilization and grazing efficiency. Achieving these targets relies on the effective allocation of pasture to grazing livestock, but in Northern Ireland only an estimated 13.5% of dairy farmers regularly make any assessment of pasture biomass cover at present (McConnell *et al.*, 2020). The use of remote sensing technology offers the potential to reduce the labour requirement associated with biomass assessment using more traditional methods (e.g. cut & weigh, or rising platemeter) and therefore encourage greater uptake of measurement activity. In order to do this, remote biomass assessment needs to be rapid, repeatable and straightforward to be undertaken on-farm by either farmers or contractors.

Previous studies have shown the potential for vegetation indices, calculated by measuring the difference between the absorbance and reflectance of specific bands of solar radiation, to correlate with pasture biomass covers in grazing systems (Poley and McDermid, 2020). The Normalized Difference Vegetation Index (NDVI) is calculated by measuring the difference between near-infrared (which vegetation strongly reflects) and red light (which vegetation absorbs) using the following equation: NDVI = (NIR – Red) / (NIR + Red). Generated values may range from -1 to +1, with higher values associated with healthy vegetation because of the high absorbance of red light and reflectance of near-infrared light by chlorophyll. The Green Normalized Difference Vegetation Index (GNDVI) is generated in the same way but uses visible green light instead of visible red and near infrared. Both NDVI and GNDVI have previously been used in pasture biomass estimation (Poley and McDermid, 2020). In this study we describe an approach for multispectral image capture using real-time kinematic (RTK) positioning to ensure repeatability, and the rapid generation of both NDVI and GNDVI index values for plots of swards of varying pre-grazing biomass covers, compared to total biomass harvest values from those plots as part of a simulated-grazing plot experiment.

Materials and methods

A total of 24 trial plots measuring 1.5 m x 5 m were established in March 2021 on a perennial ryegrass pasture at the Agri-Food and Bioscience Institute (AFBI) research farm, Hillsborough, Northern Ireland (54°27'N; 06°04'W). Plots were established in 6 replicate blocks of 4. Initially, all plots were cut to a standard residual of approximately 1,200 kg dry matter (DM) ha^{-1} using an Agria mower with the cutting bar height at 4 cm. Each following week for 3 weeks one plot in each of the 4 replicate blocks was cut using the same equipment so that four weeks after the initial trim, each replicate block of plots had a plot with 7, 14, 21 and 28 days of grass regrowth. This was designed to be representative of covers within a rotational grazing system. Four weeks from the initial trim, the plot area was overflown with a DJI P4M drone (DJI, China) equipped with RTK positioning, an in-built spectral sunlight sensor and a multispectral camera. The camera pitch was 90°, and flights were conducted at a 25 m height. The RTK positioning data were obtained through a network-RTK link using the Ordnance Survey of Northern Ireland RTK network. Immediately after plot images had been collected, eight platemeter measures were taken of the biomass cover on each plot using a Jenquip EC10 platemeter before each plot was cut using the Agria mower and fresh biomass yields recorded. A subsample of the fresh biomass was collected from each individual plot and oven-dried at 60 °C for 48 h to determine the dry matter (DM) content of the pasture biomass, and subsequently calculate the DM yield at cutting. Following plot cutting, a further 8 platemeter measures were taken of each plot to estimate the post-cutting cover. This process was then repeated for 6 consecutive monthly harvests from April-September 2021 at 28-day intervals, with the exception of the August harvest which was delayed by 7 days (14, 21, 28 and 35 days regrowth) due to adverse weather conditions preventing a UAV flight for image capture.

The multispectral images captured during each flight were processed and orthomosaics generated using the Pix4DFields software package (Pix4D, Switzerland). Radiometric calibration was automatically performed using in-flight data captured by the spectral sunlight sensor. Polygons were drawn over each plot area (excluding the border area), NDVI and GNDVI indices generated using Pix4DFields and the average NDVI and GNDVI values for each individual plot exported as a CSV file. These values were compared to the total biomass yields (corrected for the platemeter estimated post-cut residual) recorded at each harvest. The relationship between DM_Yield_kg_ha and Mean_NDVI was modelled via a non-linear regression analysis in Genstat (VSN International, UK) using an exponential curve ($y=a+br^x$). DM_Yield_kg_ha was fitted as the response variable while Mean_NDVI was fitted as the explanatory variable. This was repeated looking at the relationship between DM_Yield_kg_ha and Mean_GNDVI.

Results and discussion

The harvested plot yields in this experiment ranged from 1,072 to 3,696 kg DM ha^{-1}, representative of biomass covers found in rotational grazing systems. The exponential regression analysis identified a highly significant relationship between both NDVI and GNDVI and biomass yields ($P<0.001$) (Figure 1). Using NDVI, 68% (standard error (SE) 366) of the variation in plot biomass yields could be explained, and this increased to 69% (SE 360) with GNDVI. This is comparable to biomass estimates made using a rising platemeter (~74% variation accounted for) (Huson *et al.*, 2020). The exponential nature of this relationship indicates that at higher biomass covers this approach will be less able to accurately detect variations in biomass, and saturation in the NDVI and GNDVI would prevent the use of this approach in higher biomass covers (without the addition of other measures (Poley and McDermid, 2020)).

The data shown in Figure 1 indicate that increased variability and decreased accuracy would be encountered with covers above ~2,000 kg DM ha^{-1}, although the relationship between each VI and higher covers was still evident across the range of biomass values reported in this study (up to 3,969 kg DM ha^{-1}). Nonetheless, with pre-grazing targets typically <3,500 kg DM ha^{-1} and the clear benefits of adopting some form of pasture measurement and recording on both pasture productivity and farm profitability (McConnell *et al.*, 2020) this method shows promise for utility in grazing systems as a repeatable and straightforward approach to obtaining rapid pasture biomass estimates with limited labour requirements. Further validation of the generated equations (Figure 1) is required in future.

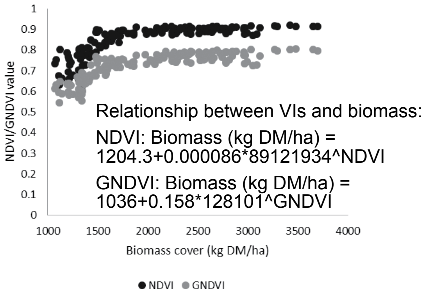

Figure 1. Vegetation index values (NDVI and GNDVI) against total plot biomass yields.

Conclusions

In this study both NDVI, and to a slightly higher degree GNDVI, showed a strong positive correlation to pasture biomass covers when determined using an exponential regression equation. These results are promising for the utility of this remote sensing approach to measure biomass in grazing systems, although its utility is likely to be limited at covers >2,000 kg DM ha^{-1}, and not applicable to covers above typical pre-grazing levels of approximately 3,000-3,500 kg DM ha^{-1}. Whilst the equipment and software utilized likely remain cost-prohibitive to farmers at the moment, in the near future if costs reduce (and in consideration of the economic value of optimizing grassland management through regular pasture measurements) using a similar protocol may be viable as a tool for routine pasture measurements on-farm.

Acknowledgements

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References

Huson K.M., Scoley G., Barr S. and McConnell D.A. (2020) Evaluation of remote-sensing and labour-saving technologies to measure pasture biomass. *Grassland Science in Europe* 25, 605.

McConnell D.A., Huson K.M., Gordon A. and Lively F.O. (2020) Identifying barriers to improving grassland utilization on dairy farms. *Grassland Science in Europe* 25, 713.

Poley L.G. and McDermid G.J. (2020) A systematic review of the factors influencing the estimation of vegetation aboveground biomass using unmanned aerial systems. *Remote Sensing* 12, 1052.

Investigation of UAV-LiDAR penetration depth in meadows for monitoring forage mass

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Abstract

Non-destructive monitoring of sward traits is of interest for grassland management. Remote sensing methods using sensors mounted on Unmanned Aerial Vehicles (UAVs) can provide timely and detailed information. Photogrammetric analysis of UAV-based image data can measure sward height which is used to estimate forage mass. In this contribution, we investigate the potential of 3D point clouds obtained with UAV-LiDAR in comparison with the established Structure from Motion and Multiview Stereopsis (SfM/MVS) analysis workflow based on UAV-derived image data to determine sward height. We (1) focused on penetration depth in meadows of UAV-LiDAR; (2) compared the results to SfM/MVS-derived sward height; and (3) evaluated the results of the UAV approaches to RPM measurements.

Keywords: UAV, LiDAR, SfM, biomass, grass, forage mass, sward height

Introduction

Non-destructive estimation of sward height (SH) is beneficial for grassland management (Bareth, 2021). Sward height is known to provide a robust estimation of forage mass (Evans and Jones, 1958). Measurement of compressed SH using rising plate meters (RPMs) is an established method to provide sward production rates (Sanderson *et al.*, 2001). Bareth and Schellberg (2018) propose replacing RPM measurements with a remote sensing analysis workflow using UAV-derived images. By using Structure from Motion (SfM) and Multiview Stereopsis (MVS), multi-temporal Digital Surface Models (DSMs) can be created to provide SH. Bareth and Schellberg (2018) reported excellent performance of UAV-derived SH compared to manual RPM measurements (R^2=0.86) for six growth periods across three years. Similar results are reported by Viljanen *et al.* (2018), Grüner *et al.* (2019) and Theau *et al.* (2021). However, SFM/MVS data processing is computing-intensive, and several analysis steps are required. An improvement might be the direct measurement of SH using an active sensor, a UAV-mounted laserscanner (LiDAR). UAV-LiDARs are widely used in forestry to provide canopy height (Sankey *et al.*, 2017) and are lately applied for crops and grasslands (Bates *et al.*, 2021; Hütt *et al.*, 2021; Maesano *et al.*, 2020). The overall objective of this contribution is to investigate the performance of a UAV-LiDAR for forage mass estimation. Therefore, we (1) focused on penetration depth in meadows of UAV-LiDAR; (2) compared the results to SfM/MVS-derived sward height; and (3) evaluated the results of the UAV approaches with RPM measurements.

Materials and methods

We conducted our study on a conventionally managed meadow field in the Wesermarsch, northern Germany (53°24'05.3'N, 8°16'36.3'E). The field is in the immediate neighbourhood of the Jade Bight, very close to the dike. The area has a temperate humid climate (Cfb), mean annual temperature 10.0 °C and a mean annual precipitation of 833 mm (www.de.climate-data.org). The soils in the area are drained mires, and meadows and pastures are the typical land use for this dairy farm region. The investigated meadow is managed for three to four cuts for silage production annually. For data acquisition, we used a self-developed Real Time Kinematic (RTK-) RPM for precise georeferencing of the measurements of compressed SH. For UAV data acquisition, two different RTK systems were operated: (1) we conducted two UAV campaigns with a DJI Phantom 4 RTK (1' sensor, 20 MP) for RGB data acquisition on 14

May and 1 June 2021; (2) A UAV-LiDAR was flown on one date, 14 May, capturing 3D point clouds with a Riegl miniVUX-1UAV laserscanner mounted on a DJI Matrice 600 Pro. Agisoft Metashape and Esri ArcGIS pro were used to analyse the RGB image data. In Figure 1, the different systems are shown. As a result, we produced one Digital Surface Model (DSM) for each date while the DSM for 1 June represents the ground model after the grass was cut. The difference between the two DSMs represents absolute sward height in cm (P4RTK-SH). The UAV-LiDAR data was analysed with Lastools retrieving sward height directly from one flight on 14 May (LiDAR-SH). Further regression analysis was performed in R software.

Results and discussion

As mentioned before, numerous studies have shown that SfM/MVS analysis provides precise canopy data to derive SH data. Therefore, we compare via regression analysis the P4RTK-SH against the LiDAR-SH. The results are shown in Figure 2. The R^2 between the two SH-datasets is 0.62 but shows significantly higher SHs for the P4RTK-SH data than for the LiDAR-SH data over the complete data range. It appears that the UAV-LiDAR used was not able of penetrating the complete grass canopy to derive ground points for precise sward height estimation.

The potential of the two different methods to estimate forage mass based on deriving spatial SH data is presented in Figure 3. For this analysis, we plotted the results of the two methods against the manual RTK-RPM measurements. In Figure 3A, the results of LiDAR-SH are shown, resulting in a moderate R^2 of 0.58. After the investigation of the penetration depth, this moderate performance was expected. In Figure 3B, the performance of the P4RTK-SH is shown. As expected and documented by several studies (Bareth and Schellberg, 2018; Viljanen *et al.*, 2018), the established SfM/MVS analysis workflow for

Figure 1. For data acquisition (A) a self developed RTK-RPM, (B) a DJI Phantom 4 RTK, and (C) a Riegl miniVUX-1UAV mounted on a DJI Matrice 600 pro (UAV-LiDAR) are used.

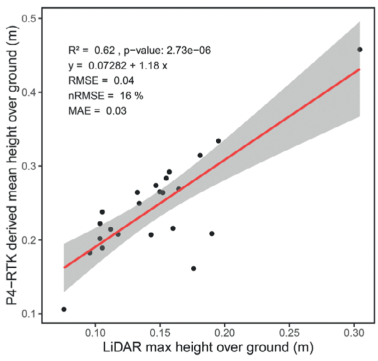

Figure 2. Investigating UAV-LiDAR penetration depths with SfM/MVS analyses.

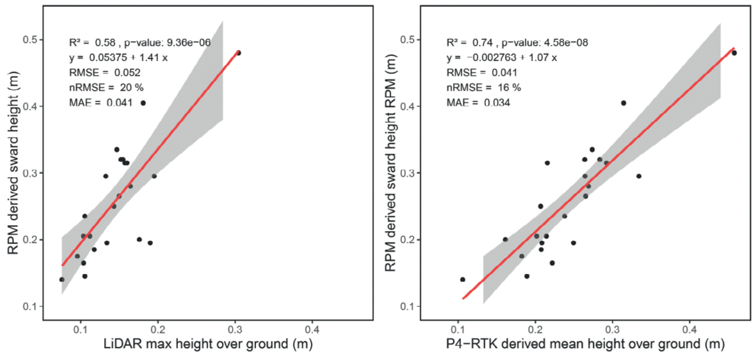

Figure 3. Comparison of single date UAV LiDAR data and optical, SfM/MVS-based multitemporal analysis of Phantom4 RTK data with rising plate meter measurements.

UAV-derived RGB data results in a R^2 of 0.74 having a trendline inclination of approx. 1. In this study, the UAV-LiDAR does not perform as well as established SfM-MVS analysis workflow to derive SH data for estimating forage mass. However, we investigated UAV-LiDAR using only one date to derive absolute SH, while for the P4RTK-SH we used two dates, before and after the cut.

Conclusions

We conclude that penetration depth of UAV-LiDAR is limited in grassland canopies, and one acquisition date is insufficient to derive absolute SH in cm. Therefore, we propose investigating UAV-LiDAR using two dates, before and after cut, to improve the method. Finally, we have to extend our analysis on a larger data set for multiple growths in a year and multiple years.

Acknowledgements

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References

Bareth G. (2021) Towards an informed grassland farming – sensors, platforms and algorithms. *Grassland Science in Europe* 26, 11-20.

Bareth G. and Schellberg J. (2018) Replacing manual rising plate meter measurements with low-cost UAV-derived sward height data in grasslands for spatial monitoring. *Journal of Photogrammetry, Remote Sensing and Geoinformation Science* 86, 157-168.

Bates J.S., Montzka C., Schmidt M. and Jonard F. (2021). Estimating canopy density parameters time-series for winter wheat using UAS Mounted LiDAR. *Remote Sensing* 13(4), 710.

Evans R.A. and Jones M.B. (1958) Plant height times ground cover versus clipped samples for estimating forage production. *Agronomy Journal* 50 (9), 504-506.

Grüner E., Wachendorf M. and Astor T. (2020) The potential of UAV-borne spectral and textural information for predicting aboveground biomass and N fixation in legume-grass mixtures. *PLoS ONE* 15(6), e0234703.

Hütt C., Bolten A., Hohlmann B., Komainda M., Lussem U., Isselstein J. and Bareth G. (2021) First results of applying UAV laser scanning to a cattle grazing experiment. *Grassland Science in Europe* 26, 135-137.

Maesano M., Khoury S., Nakhle F., Firrincieli A., Gay A., Tauro F. and Harfouche A. (2020). UAV-based LiDAR for high-throughput determination of plant height and above-ground biomass of the bioenergy grass *Arundo donax*. *Remote Sensing* 12(20), 3464.

Sanderson M.A., Rotz C.A., Fultz S.W. and Rayburn E.B. (2001) Estimating forage mass with a commercial capacitance meter, rising plate meter, and pasture ruler. *Agronomy Journal* 93(6), 1281-1286.

Sankey T., Donager J., McVay J. and Sankey J.B. (2017) UAV lidar and hyperspectral fusion for forest monitoring in the southwestern USA. *Remote Sensing of Environment* 195, 30-43.

Théau J., Lauzier-Hudon É., Aubé L. and Devillers N. (2021) Estimation of forage biomass and vegetation cover in grasslands using UAV imagery. *PLoS ONE* 16(1), e0245784.

Milk production potential of regrowth grass silages in northern latitudes

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Abstract

The objective of this study was to examine if short regrowth time has positive effects on grass nutritive value, dry matter (DM) intake (DMI) and milk production of dairy cows. The experiment was conducted using 39 dairy cows in an incomplete cross-over design with two periods. The experiment included first (H1), second (H2), and third-cut (H3) grass silages. The diets were fed as a total mixed ration, where the average forage to concentrate ratio was 640:360 on a DM basis. The energy values of the silages were 11.3, 11.5, and 11.2 MJ metabolizable energy (ME) kg^{-1} DM for H1, H2, and H3 respectively. Milk production was highest with H2. The energy-corrected milk (ECM) yields were 32.4, 34.1, and 32.3 kg d^{-1} for H1, H2, and H3 respectively ($P<0.05$). Dry matter intake was highest with H1 and lowest with H3 ($P<0.01$). The milk production and feed efficiency (MJ ME kg^{-1} ECM) of H1 was smaller than in the other harvests. The milk production of H2 was higher than that for typical regrowth grass. These results demonstrate that grass ensiled in a short regrowth time can positively affect the silage digestibility and milk production of dairy cows.

Keywords: feed intake, harvesting strategy, regrowth grass silage

Introduction

A three-cut harvesting strategy for silage has increased in the northern part of Finland during the last decade. This strategy is recommended if the main aim is to maximize digestible grass yield over the whole growing season (Hyrkäs *et al.*, 2015). However, the milk production of regrowth silages has been lower than primary growth, even when feed values have been similar between harvests (e.g. Kuoppala *et al.*, 2008). One solution to improve the milk production potential of regrowth silages is to reduce the regrowth time between harvests. With this strategy, the third harvesting period starts to grow relatively early, and the late autumn growing period is avoided. This has beneficial effects on silage quality (Sairanen *et al.*, 2021). The aim of this experiment was to study the effects of an early harvested first regrowth strategy on the dry matter (DM) intake (DMI) and milk production of dairy cows.

Materials and methods

Experimental silages were produced during the growing season of 2020 at the experimental farm of Natural Resources Institute (Luke) in Maaninka, Finland. The feeding trial included three harvest times: primary growth (H1), first regrowth (H2), and second regrowth (H3) grass (Table 1 and 2). All the silages were based on a mixture of timothy (*Phleum pratense* L.) and meadow fescue (*Festuca pratensis* Huds.). H1 and H3 were stored in bunker silos, and H2 in round bales. The pre-wilting time of silages was approximately one day.

The feeding trial was conducted using 39 free-stall housed mid- and late-lactating Holstein and Nordic red dairy (16 primiparous and 23 multiparous) cows. The average pre-experimental milk yield (MY) was 30 (standard deviation (SD) 8.4) kg, and days in milk (DIM) 185 (SD 72.7). Diets were fed as a total mixed ration (TMR) with an average forage to concentrate ratio 640:360 on a DM basis. The experimental diets and their chemical composition are presented in Table 2. The cows received 1.5 kg commercial concentrate from automated feeders. The cows were divided into seven blocks according to their parity, pre-experimental MY and DIM. The experiment was conducted as an incomplete cross-over

Table 1. Dates of harvests, growing days, and descriptive weather statistics during the 2020 growing season for the first (H1), second (H2), and third harvest (H3) of grass silage.

	H1	H2	H3
Date of harvest	16 June	16 July	20 August
Effective temperature sum, °C d [1,2]	271	357	404
Growing days [2]	30	30	35
Precipitation, mm [2]	6	126	49

[1] Accumulated temperature over 5 °C since onset of the growing season.

[2] Accumulated precipitation since onset of the growing season for H1, and since the previous cut for H2 and H3.

Table 2. Chemical composition of the first (H1), second (H2), and third (H3) harvest of grass silages and in the experimental diets of the first (D1), second (D2), and third (D3) harvest of grass silages.

	H1	H2	H3	D1	D2	D3
Dry matter (DM), g kg^{-1}	423	296	288	500	405	362
Crude protein	174	213	178	174	199	176
Neutral detergent fibre	513	434	476	392	340	369
Ammonium N, g kg^{-1} N	27	41	35	-	-	-
Metabolizable energy, MJ kg^{-1} DM	11.26	11.53	11.16	11.68	11.85	11.62
SDMI index[1]	117	111	103	-	-	-

[1] SDMI index = the relative intake potential of silage dry matter (Huhtanen *et al.,* 2007).

design, with two periods and three dietary treatments. The data were analysed using a SAS MIXED procedure, including period, harvesting time, and block as fixed variables and animal as a random variable.

Results and discussion

The weather conditions were good during harvesting, which can be seen in the reasonably high DM content and good fermentation quality of silages (Table 2). Only ammonia and volatile fatty acids proportions in H2 were slightly high, which is a typical phenomenon with baled silages. However, the relative intake potential of silage dry matter (SDMI index, Huhtanen *et al.*, 2007) was high in H2, indicating the good quality of silage. The SDMI index was highest in H1 and lowest in H3. A two-week drought period after the first harvest delayed the start of grass growth, which maintained the early maturity stage and consequently D-value of H2. The energy and crude protein (CP) content was highest, and the neutral detergent fibre content was lowest, with H2.

The highest DMI was observed with H1 and the lowest with H3 (Table 3), which is in line with previous studies (Pang *et al.*, 2019; Sairanen *et al.*, 2021). The ME content of silage has been reported to predict the expected DMI most accurately within the harvest time (Pang *et al.*, 2021), but the effect of harvest time itself can overrule this, as seen in this study. The ME content of H2 was clearly highest, and the fermentation quality did not explain the differences in the DMI. The effect of harvest time is categorical, and there is no clear explanation for this (Pang *et al.*, 2019; Sairanen *et al.*, 2021). Ensiled herbage in H2 lacked dead material and did not show evidence of plant diseases, so the texture of the grass did not affect the silage palatability. One explanation for the limited intake of H2 may be the high silage CP content. CP is converted to ammonia in the rumen, limiting metabolic feed regulation.

The MY of H1 was smaller than expected according to ME intake (Table 3), especially compared to H2 and H3. In general, the ME use-efficiencies in milk production were lower than the reference value of

Table 3. Milk performance and feed intake of experimental diets of the first (H1), second (H2), and third (H3) harvest of grass silages.[1,2]

		H1	H2	H3	SEM	*P*-value
Production, kg	Milk	28.5^{a}	29.8^{b}	28.5^{a}	0.48	<0.05
	ECM	32.4^{a}	34.1^{b}	32.3^{a}	0.67	<0.05
Milk urea, mg dl^{-1}		23.4^{a}	29.6^{b}	21.8^{c}	0.62	<0.01
Intake, kg DM	Silage	15.0^{a}	14.4^{b}	14.3^{b}	0.24	<0.005
	Concentrate	9.0^{a}	9.1^{a}	8.6^{b}	0.13	<0.001
	Total	24.0^{a}	23.5^{a}	22.9^{b}	0.36	<0.01
	ME MJ d^{-1}	259^{a}	253^{b}	246^{c}	3.60	<0.05
Efficiency of milk production	MJ ME kg^{-1} ECM	6.00^{a}	5.56^{b}	5.70^{b}	0.13	<0.05

[1] DM = dry matter; ECM = energy-corrected milk; ME = metabolizable energy; SEM = standard error of the mean.

[2] Means in a row within each experiment without a common superscript letter differ significantly according to the Tukey test.

5.15 MJ ME kg^{-1} ECM in the feed tables (Luke, 2021). This can partly be explained by the lactation stage of the cows. Pang *et al.* (2019) stated that the energy value of high digestibility silages is overestimated. Our findings support this hypothesis. H2 had a very high CP content, which was reflected in high milk urea content. An excess amount of ammonia in the rumen has to be excreted via urine, and this requires extra energy.

It has been stated that energy in regrowth silages is directed more into MY at the expense of body tissues, which causes an apparent increase in feed efficiency (Pang *et al.*, 2019; Sairanen *et al.*, 2021). This can be a problem for certain groups of animals, especially early lactation cows, which easily suffer ketosis.

Conclusions

In this study, the milk production from first regrowth silage was higher than with typical regrowth silage. These results demonstrate that grass ensiled in a short regrowth time can have positive effects on the silage digestibility and milk production of dairy cows.

References

Huhtanen, P., Rinne, M. and Nousiainen, J. (2007) Evaluation of the factors affecting silage intake of dairy cows: a revision of the relative silage dry-matter intake index. *Animal* 1, 758-1770.

Hyrkäs, M., Sairanen, A., Kykkänen, S., Virkajärvi, P. and Isolahti, M. (2015) Different harvesting strategies and cultivar mixtures for grass silage production in Finland. *Grassland Science in Europe* 20, 239-241.

Kuoppala, K., Rinne, M., Nousiainen, J. and Huhtanen, P. (2008) The effect of cutting time of grass silage in primary growth and regrowth and the interactions between silage quality and concentrate level on milk production of dairy cows. *Livestock Science* 116, 171-182.

Luke (2021) Feed Tables and Nutrient Requirements. Natural Resources Institute Finland, Jokioinen, Finland. Available at: www.luke.fi/feedtables.

Pang, D., Krizsan, S., Sairanen, A., Nousiainen, J. and Huhtanen, P. (2019) Modelling feed intake and milk yield responses to different grass ley harvesting strategies. *Grass and Forage Science* 74, 509-524.

Pang, D., Yan, T. and Krizsan, S. (2021) Effect of strategy for harvesting regrowth grass silage on performance in dairy cows. *Journal of Dairy Science* 104, 367-380.

Sairanen, A., Kajava, S., Palmio, A. and Rinne, M. (2021) A comparison of grass silages harvested at first, second and third cut on feed intake and milk production of dairy cows. *Agricultural and Food Science* 30, 74-84.

Drivers of N dynamics after ploughing-up of different grassland systems for maize

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Abstract

The break-up of 3-5 year-old grassland in grass-arable rotations can lead to increased C and N mineralization and result in large NO_3-N leaching losses over winter after harvest of the follow-up crop. Among the main drivers for the N dynamics following break-up are previous and current N fertilization, time of break-up, temperature and amount of rainfall. In a field experiment on a sandy soil in north-west Germany, we investigated how N yields and NO_3-N leaching of silage maize as a follow-up crop (no N applied) were affected by different N fertilizer management (synthetic and organic; 0-360 kg N ha^{-1}) over three years of grassland. Periods of drought occurred in the year previous to break-up and during the cultivation of maize. We tested how N input and N balances of the grass phase and hot-water-soluble C and N, as an indicator of the mineralization potential, were related to NO_3-N leaching after maize. Dry matter and N yield of maize did not differ among former N management regimes. Nitrate-N leaching amounted to 70-110 kg N ha^{-1}. Larger N leaching was related to previous N input with slurry >240 kg N ha^{-1} and higher hotwater-soluble C and N.

Keywords: N leaching, hot water-soluble N, climate change, drought

Introduction

In compliance with EU funding legislation, grass swards on fields with arable status are usually ploughed-up every five years for cultivation of an arable crop over an interval of 1-3 years. Under permanent grassland the content of C and N will usually accumulate in the topsoil, and break-up for arable use will then result in long-term supply of N from mineralization processes (Buchen *et al.*, 2017). However, even in grass-arable systems, break-up of grassland will usually lead to enhanced mineralization of N, at least in the first year. Depending on the choice of follow-up crop, amount of N fertilizer, and weather conditions (rain and temperature as drivers of mineralization) N surpluses might occur and larger residual N in autumn can result in increased N leaching over the following winter (Buchen *et al.*, 2017). At present, there is insufficient information on N efficiency and N leaching in these extended grass-arable systems.

Material and methods

We used data from a grass-based field experiment with five different treatments of N fertilizer. The experimental site was located in northwest Germany on a coarse-textured soil (sand to loamy sand). The area has a maritime climate, with a long-term average temperature of 8.7 °C and an average annual rainfall of 786 mm. The sward was established in 2013 and was based on a ryegrass (*Lolium perenne* L.) dominated mixture with no clover. All treatments were ploughed-up for maize five years after establishment of the grass sward and after three experimental years.

The experiment started in 2016 and consisted of a one-factorial design replicated in four blocks, where we compared different combinations of N input by cattle slurry and synthetic N fertilizer (Table 1). Plots were 12×4.5 m in size. Cattle slurry had been applied by sliding shoe to all treatments, apart from the Control, with 120 kg N ha^{-1} in spring. For treatments >120 kg N ha^{-1} N input, cattle slurry and CAN (calcium-ammonium-nitrate) were applied in doses of 60 kg N ha^{-1} after the first and second cutting. In spring 2019, the grass sward was ploughed-up for cultivation of silage maize (FAO 240, 8 plants m^{-2}).

The maize crop did not receive any N from fertilizer, but a mineral supply with P and K (30 kg P ha^{-1} and 150 kg K ha^{-1}). Maize harvest took place on 24 Sept. and yield was determined from sub-plots with an plot harvester (Haldrup); biomass samples were taken for subsequent N analysis. In each plot, six suction cups at 75 cm depth allowed the regular sampling of leached water. NO_3-N was analysed photometrically with a flow-injection analyser. Leaching, as determined by a water-balance model, occurred from late October 2019 to mid-March 2020 and amounted to 281 mm. Hotwater-soluble N was analysed from air-dry soil (0-10 cm) that was sampled before break-up of grassland in spring 2019. Analysis of variance analysis was carried out; values for NO_3-N leaching were log-transformed to achieve normality in data.

Results and discussion

A prolonged summer drought during the last year of the grass phase in 2018 was followed by another dry period during the cultivation of maize in summer 2019. Consequently, maize dry matter yields (11.0-12.7 t ha^{-1}) and N yields (148-173 kg ha^{-1}) were below the values from an experiment on a similar site of 13.5-16.5 t ha^{-1} and 160-220 kg ha^{-1}, respectively (Kayser *et al.*, 2011). Despite the omission of N fertilization, NO_3-N leaching after harvest of maize amounted to 70-110 kg ha^{-1}. The N balances show that 190-250 kg N ha^{-1} was provided by mineralization of soil N for N yield and NO_3-N leaching (Table 1). Under similar conditions, Kayser *et al.* (2008) found NO_3-N leaching losses in maize following break-up of permanent grassland of 118 kg ha^{-1} (no N to maize) and 216 kg ha^{-1} (synthetic N fertilizer 160 kg N ha^{-1}).

The differences among the former N treatments (Table 1) during the grass phase for DM yield and N yield of the following maize were small and not significant. Leaching losses of NO_3-N were 20-30 kg ha^{-1} higher for N input >240 kg ha^{-1} during the grass phase but this could not be confirmed by ANOVA. However, a post-hoc contrast analysis showed significant differences ($P<0.05$) in NO_3-N leaching for the following combinations: SLM360 > Control; SLR240 > Control; and SLM360/SLR240 > Control/SLR120.

The effects of the former N fertilization regime during the grass-phase on N dynamics during cultivation of maize were strongly altered by: (1) the consequences of the drought period in 2018 and the drought phase in summer 2019 affecting maize growth; and (2) mineralization of N from grassland break-up. During the two years a sequence and combination of massive disruptions affected the N dynamics in the plant-soil system: in the last year of the grass phase (2018), yield depression and limited N uptake because of drought, sward turning brown, rewetting in autumn with N mineralization and high NO_3-N leaching over winter (20-220 kg ha^{-1}) followed by ploughing up of the grass sward in the following spring

Table 1. Input of N with cattle slurry and synthetic N (CAN) NO_3-N leaching (N leach) and hot water-soluble N (HWS-N) during the grass phase and DM yield, N yield, NO_3-N leaching, and N balance for the follow-up crop maize.[1,2]

	(2018/2019) Grass				(2019/2020) Maize				
Treatment	Slurry-N	CAN-N	N leach[3]	HWS-N	N Input	DM yield	N yield	N leach[3]	N balance
	kg ha^{-1}	kg ha^{-1}	kg ha^{-1}	g kg^{-1}	kg ha^{-1}	t ha^{-1}	kg ha^{-1}	kg ha^{-1}	kg ha^{-1}
Control	0	0	22^{a}	0.079^{a}	0	11.8	160	73	-203
SLR120	120	0	28^{a}	0.084^{a}	0	11.6	150	70	-190
SLR240	240	0	87^{b}	0.094^{b}	0	11.0	148	106	-224
SLM240	120	120	146^{bc}	0.084^{a}	0	11.7	145	81	-196
SLM360	240	120	221^{c}	0.098^{b}	0	12.7	173	110	-253
P-value			<0.001	<0.001		0.60	0.32	0.11	

[1] N balance = N deposition (30 kg N ha^{-1}) – (Nyield + N leaching);

[2] Means of 4 replications; ANOVA, means with different letters differ at $P<0.05$.

[3] ANOVA and comparison of means based on log-transformed values (natural logarithm); back-transformed.

(2019), cultivation of maize and another drought period leading again to smaller yields and N offtake. The amount of water leached in the following winter was slightly above that of the long-term average which also added to the increased level of NO_3-N leaching losses.

Hot water soluble N (and C) is an indicator of the labile, easily mineralizable organic N pool in soil and is closely related to microbial biomass. It appears to be a valuable indicator of estimates of management induced changes in soil organic matter composition (Sparling *et al.*, 1998). In our experiment, NO_3-N leaching increased with increasing content of hot water-soluble N; and hot water-soluble N content was higher in treatments with slurry input (as compared to the Control) and especially at application rates of slurry >240 kg N ha^{-1}. Hot water-soluble N expressed as a percentage of total N (TN) was related to the amount and form of N input and was also related to NO_3-N leaching. After only three years of differing N input there seems to have been an effect on soil microbial processes with a direct impact on NO_3-N leaching after grassland break-up and cultivation of maize (Figure 1).

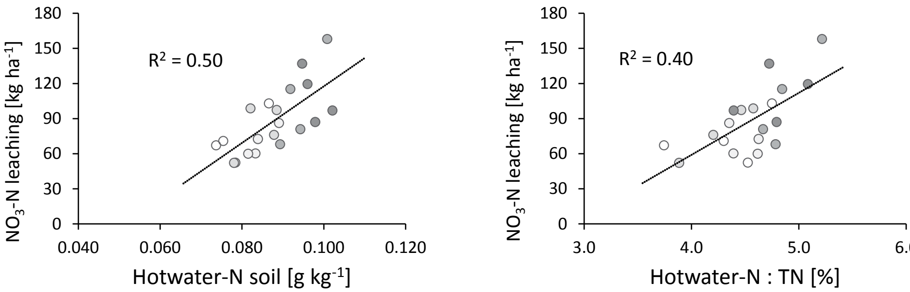

Figure 1. Left: Relationship between hot water-soluble-N (g kg^{-1}) and NO_3-N leaching (kg ha^{-1}) and right: relationship between hot water-soluble-N:TN ratio (%; percentage of HWS-N of TN), and NO_3-N leaching (kg ha^{-1}). Hot water-soluble-N determined before grassland break-up and NO_3-N leaching of winter period after maize harvest. White circles, control, grey to darker circles, increasing N input 120-360 kg N ha^{-1}, see Table 1.

Conclusions

Our results confirm the complexity of N dynamics related to grassland break-up and the strong impact of weather and management on N mineralization and N efficiency. This can only be partly controlled by management and calls for an adapted N fertilization after break-up. The findings also suggest that if climate change induced drought periods occur more frequently this would affect N efficiency in grass-arable systems, especially on lighter soils.

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References

Buchen C., Well R., Helfrich M., Fuß R. .... and Flessa H. (2017) Soil mineral N dynamics and N_2O emissions following grassland renewal. *Agriculture, Ecosystems & Environment* 246, 325-342.

Kayser M., Seidel K., Müller J. and Isselstein J. (2008) The effect of succeeding crop and level of N fertilization on N leaching after break-up of grassland. *European Journal of Agronomy* 29, 200-207.

Kayser M., Benke M. and Isselstein J. (2011) Little fertilizer response but high N loss risk of maize growing on a productive organic-sandy soil. *Agronomy for Sustainable Development* 31 (4), 709-718.

Sparling G., Vojvodic-Vukovic M. and Shannon P.W. (1998) Hot water soluble C as a simple measure of labile soil organic matter: the relationship with microbial biomass C. *Soil Biology and Biochemistry* 30, 1469-1472.

Dry matter intake and enteric methane emissions from two contrasting silage qualities fed over the prepartum period

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Abstract

The quality of silage fed during the dry cow period can dictate the level of feed intake, which is strongly correlated with enteric methane (CH_4) emissions. Therefore, the objective of the experiment was to evaluate the impact of silage quality on dry matter intake (DMI) and CH_4 emissions of non-lactating dairy cows. Thirty dairy cows were randomly selected and assigned to one of two (high quality (HS) and low quality, (LS)) treatments (n=15). The experiment was conducted over a six-week period. Daily CH_4 emissions were assessed using the Greenfeed emission-monitoring unit and individual cow feed intake was assessed using the Hokofarm RIC feed stations. Bodyweight and body condition score were assessed at three time points over the experimental period. Dry matter intake ($P<0.05$) was greater in the HS treatment compared to the LS treatment (HS 12.8, LS 8.9, kg DM cow^{-1}, respectively). This resulted in higher daily CH_4 in cows offered the HS silage ($P<0.05$). However the cows on the HS treatment had a lower ($P<0.05$) CH_4 yield expressed as g kg^{-1} dry matter intake. Silage quality is a key factor that affects the level of dry matter intake and CH_4 emissions by cows over the dry cow period.

Keywords: dry cow period, silage quality, methane emissions, dry matter intake

Introduction

The quality of forages fed over the dry cow period is reported to be imperative to the level of feed intake, metabolic status and lactation performance of dairy herds (Richards *et al.*, 2020). However, the impact of nutrition on the level of methane (CH_4) emitted during this period is often over looked. Enteric CH_4 accounts for 64% of Ireland's agricultural greenhouse gas emissions (Lanigan *et al.*, 2019). Ireland has now committed to a target to reduce agricultural greenhouse gas emissions by 22-30% by 2030 (DECC, 2021). Improving forage quality is a key mitigation strategy for enteric CH_4 emissions in lactating dairy cows (Hristov *et al.*, 2013, Eugène *et al.*, 2021). However, there is limited research evaluating the impact of forage quality on CH_4 emissions during the dry cow period. Dry cow diets in pasture-based dairy systems typically comprise grass silage, which is harvested and ensiled during periods of excess herbage production and fed during the winter dry period (Dillon *et al.*, 1995). Therefore the quality of the silage fed during this period is a key factor impacting dry matter intake (DMI), bodyweight, body condition score (Butler *et al.*, 2011) and enteric CH_4 emissions of dairy cows. The objective of the current experiment was to compare the impact of silage of high and low quality on DMI and CH_4 emissions of dry cows.

Materials and methods

A six-week experiment (21 Dec – 31 Jan) was established at the Teagasc, Grassland Research and Innovation centre, Moorepark, Fermoy, Co. Cork. Thirty pregnant non-lactating dairy cows were randomized and balanced for breed, parity, lactation, expected calving date, economic breeding index (EBI), bodyweight, body condition score (BCS) and allocated to one of two treatments (n=15): High quality silage (HS) and Low quality silage (LS). The HS treatment received high quality grass-clover silage throughout the experiment. The LS treatment received low quality second-cut pit silage, harvested in July 2020. Silage samples were collected twice weekly and analysed using near infra-red spectrometry for chemical composition. Bodyweight and BCS was assessed at the start, week 4, and at the end of the experiment. Daily individual cow DMI was determined using Hokofarm feed stations. Daily CH_4 emissions was monitored using the C-Lock GreenFeed emissions monitoring (GEM) system. Animals had free access to a GEM unit with a known amount of concentrates (0.68±0.2, kg dry matter (DM) cow^{-1} day^{-1}) dispensed. Daily visits (3±1, n day^{-1}) and visit duration (241±29, s), were above the recommended average (Arthur *et al.*, 2017). Data were analysed using a mixed model computed through SAS 9.4 (SAS Insistitue Inc, Cary, USA). The mixed model of a dependent variable incorporated fixed effects of week, treatment, treatment by week, with week the repeated effect measured and animal the random factor.

Results and discussion

Chemical composition of both treatments silage is reported in Table 1. The HS treatment silage had greater dry matter digestibility resulting in cows consuming a significant (P<0.05) 3.85 kg cow^{-1} of additional DMI compared with the LS treatment (Table 2). This resulted in the HS treatment emitting significantly (P<0.05) more CH_4 (g day^{-1}) (Table 2). The HS treatment emitted higher CH_4 emissions mainly due to improved forage digestibility elevating animal DMI; however, this in turn reduced CH_4 yield (g kg^{-1} DMI). Treatment did not significantly impact bodyweight or body condition score over the experimental period (618 kg and 3.35, respectively). Dry matter intake has previously been reported to be strongly correlated (R^2=0.86) with CH_4 emissions (Hristov *et al.*, 2013); this was evident in the current study with a positive correlation (R^2=0.51). Van Gasetelen *et al* (2019) and Hristov *et al* (2013) both reported the relationship of improving forage quality with increasing DMI, and in turn reducing CH_4 yield on a per kg of DMI basis. The current experiment findings support those of Van Gasetelen *et al* (2019); however, the LS treatment had lower daily CH_4 emissions due to reduced forage quality suppressing animal DMI, consuming below their energy requirement (Richards *et al.*, 2020).

Table 1. Forage analysis of grass silage fed to the HS and LS treatment with the level of significance of treatment.

Item	HS	LS	SEM	Significance of treatment
Dry matter (%)	37.4	21.8	1.09	0.001
Crude protein (g kg^{-1} DM)	111.4	84.9	2.67	0.001
Dry matter digestibility (g kg^{-1} DM)	706.8	568.7	6.56	0.001

Table 2. The effect of feeding high quality (HS) and low quality (LS) silage on bodyweight, BCS, DMI and CH_4 emissions with the level of significance of treatment, week and their interaction.

Item	HS	LS	SEM	Level of significance		
				Treatment	Week	Treatment × Week
DMI (kg DM cow^{-1})	12.8	8.93	0.60	0.001	0.001	0.001
CH_4 (g day^{-1})	253.7	213.16	7.12	0.001	0.001	0.001
CH_4 (g kg^{-1} DMI)	21.2	25.79	1.22	0.008	0.001	0.001
Bodyweight (kg)	630	606	13.7	0.195	-	-
BCS (1-5)	3.36	3.35	0.064	0.897	-	-

Conclusions

Silage quality is a key factor dictating animal DMI, which is key driver of CH_4 emissions. Feeding a lower quality silage over the dry-cow period suppressed animal DMI, which in turn reduced daily CH_4 emissions; however it increased CH_4 yield (g kg^{-1} DMI).

References

Arthur P.F., Barchia I.M., Weber C., Bird-Gardiner T., Donoghue K.A., Herd R.M. and Hegarty R.S. (2017) Optimizing test procedures for estimating daily methane and carbon dioxide emissions in cattle using short-term breath measures. *Journal of Animal Science* 95, 645-656.

Butler M., Patton J., Murphy J.J. and Mulligan F.J. (2011) Evaluation of a high-fibre total mixed ration as a dry cow feeding strategy for spring-calving Holstein Friesian dairy cows. *Livestock Science* 136, 85-92.

DECC (2021) Climate Action Plan 2021. Department of the Environment, Climate and Communications, Dublin, Ireland.

Dillon P., Crosse S., Stakelum G. and Flynn F. (1995) The effect of calving date and stocking rate on the performance of spring-calving dairy cows. *Grass and Forage Science,* 50, 286-299.

Eugène M., Klumpp K. and Sauvant D. (2021) Methane mitigating options with forages fed to ruminants. *Grass and Forage Science* 76, 196-204.

Hristov A.N., Oh J., Firkins J.L., Dijkstra J., Kebreab E., Waghorn G., Makkar H.P.S., Adesogan A.T., Yang W., Lee C., Gerber P.J., Henderson B. and Tricarico J.M. (2013) Special Topics-Mitigation of methane and nitrous oxide emissions from animal operations: I. A review of enteric methane mitigation options. *Journal of Animal Science* 91, 5045-69.

Lanigan G., Donnellan T., Hanrahan K., Carsten P., Shalloo L., Krol D., Forrestal P.J., Farrelly N., O'Brien D. and Ryan M. (2019) An analysis of abatement potential of Greenhouse Gas emissions in Irish agriculture 2021-2030. Teagasc, Ireland.

Richards B.F., Janovick N.A., Moyes K.M., Beever D.E. and Drackley J.K. (2020) Comparison of prepartum low-energy or high-energy diets with a 2-diet far-off and close-up strategy for multiparous and primiparous cows. *Journal of Dairy Science* 103, 9067-9080.

Van Gastelen S., Dijkstra J. and Bannink A. (2019) Are dietary strategies to mitigate enteric methane emission equally effective across dairy cattle, beef cattle, and sheep? *Journal of Dairy Science* 102, 6109-6130.

The impact of feeding lactic acid bacteria inoculated silage on milk production in late lactation

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Abstract

In pasture-based dairy systems late lactation is a period of reduced animal performance and grass supply resulting is grass silage being commonly fed during this period. Silage quality is a key factor dictating the level of milk production when fed to lactating dairy cows. The objective of this experiment was to evaluate the impact of feeding grass silage inoculated with *Lactobacillus plantarum* (LP58) and *Lactococcus lactis* (SL242), on late lactation milk production. In autumn 2020, 30 late-lactation dairy cows were randomly assigned to two treatments (n=15) LAB and CONT for a seven-week period. The LAB treatment were fed lactic acid bacteria-inoculated grass silage, whereas the CONT treatment received untreated grass silage. Milk yield was recorded daily with milk composition measured weekly. The LAB treatment silage had a lower DM concentration and tended to have higher protein concentrations ($P<0.1$). There was no effect of treatment on milk production. Week had a significant effect on all milk production variables ($P<0.001$). Milk yield was similar between treatments; however, fat and protein corrected milk (FPCM) yield was higher in the LAB treatment. There was a significant treatment by week interaction on milk fat concentration ($P<0.05$) and milk solid production ($P<0.05$). Inoculating grass silage with *L. plantarum* and *L. lactis* did not significantly improve milk production; however, changes in milk composition were clearly evident.

Keywords: lactic acid bacteria, silage quality, milk production

Introduction

Late lactation is a period of reduced animal performance due to day in milk, and also reductions in grass supply, in spring-calving pasture-based dairy systems (McKay *et al.*., 2019). During periods of reduced grass supply, grass silage is commonly fed to lactating dairy cows. Grass silage accounts for 18% of an Irish cow's diet annually (O'Brien *et al.*., 2018). Thus, silage quality is imperative to improving and maintaining milk production of lactating dairy cows. Previous research evaluating high grass silage diets reported reductions in animal performance when compared to diets abundant in grazed grass (Claffey *et al.*., 2018). When silage is fed, it has been previously observed that management strategies (cutting date, sward type, and fertilizer regime) can improve silage quality, hence improving milk production (Pang *et al.*., 2021). Another such strategy in improving silage quality is the use of silage inoculants, which have been widely reported to improve the quality of grass silage (Carvalho *et al.*., 2021) and increase animal performance (Oliveira *et al.*., 2017). However, the results have varied according to forage type and bacterial strain used. Combining bacterial strains in silage inoculants has the main aim of achieving symbiotic benefits from both bacterial strains (Muck *et al.*., 2018). Therefore the objective of the current study examined the impact of feeding grass silage inoculated with *Lactobacillus plantarum* and *Lactococcus lactis* on milk production in late lactation.

Materials and methods

In autumn 2020, a seven week experiment (18 Oct - 6 Dec) was conducted using thirty spring-calving dairy cows selected from the Moorepark dairy herd and assigned to one of two treatments (n=15). The lactic acid bacteria treatment (LAB) was fed lactic acid bacteria inoculated grass silage with the control treatment (CONT) receiving untreated grass silage. Animals were randomized according to calving date, parity, breed, lactation number, economic breeding index, days in milk, two week pre experimental; milk yield, milk protein, milk fat, milk solids, bodyweight and body condition score. For the duration of the experiment, animals were housed in a purpose built cubicle shed. Grass silage was harvested in July 2020 at a pre-cutting herbage mass of 4,000 kg DM ha^{-1}. The LAB treatment silage was inoculated with *Lactobacillus plantarum* (LP58) and *Lactococcus lactis* (SL242) via direct application onto the cut grass swath to achieve an application rate of 0.12 kg additive per 1000 kg DM of grass. The CONT silage was made from the same area and was harvested without the additional application of the silage additive. All silage was baled and ensiled for 98 days prior to commencement of feeding. Treatments were fed once daily using a Keenan mechfibre 350 diet feeder. Fresh silage samples were taken twice weekly for chemical composition analysis. At milking, each treatment received four kg (FW) of a 20% crude protein concentrate. Cows were milked twice daily with individual milk yields measured daily and milk composition determined once weekly from one successive evening and morning milking using mid-infrared spectroscopy analysis. Individual dry matter intake (DMI) was estimated on week 4, using the n-alkane techniques as described by Mayes *et al.* (1986), and modified by Dillon and Stakelum (1989). All data were analysed using a PROC mixed model in SAS 9.4 (SAS Insistitue Inc, Cary, USA), with dependent variables analysed for each week with the model contained terms for treatment, week and their associated interactions, week was the repeated measure with animal the random factor.

Results and discussion

Silage chemical composition is reported in Table 1. Lactic acid bacterial-inoculated grass silage had lower DM content than untreated grass silage, with no other difference evident (Table 1). Week had a significant effect ($P<0.05$) for all observed milk production variables. Treatment had no effect on either milk yield or milk composition (Table 2). Ellis *et al.* (2016) reported similar findings on animal production using similar bacterial strains and forages, mainly due to lack of improvement in forage digestibility and animal DMI. Increases in milk yield and milk fat production particularly after week 4 in the LAB treatment resulted in significant treatment by week interaction ($P<0.05$) for fat and protein corrected milk yield (FPCM) and daily milk solids. There was no effect of treatment on silage DMI (12.56 kg cow^{-1}), which accounts for the lack of improvement in animal performance compared to CONT treatment. Enhancing silage quality is critical for increasing animal silage DMI, which increases net energy intake and thus milk production (Pang *et al.*, 2021). Silage quality was comparable between the two treatments, which explains the LAB treatment's lack of response in milk yield; however, weekly milk fat alterations, particularly in week 4 and 5 resulted in significant treatment by week interaction for milk composition in LAB treatment (Table 2).

Table 1. Chemical composition of both lactic acid bacteria treated silage (LAB) and untreated control silage (CONT) and the level of significance for treatment.

	LAB	CONT	SEM	Level of significance
Item				Treatment
Dry matter (%)	26.40	26.63	0.056	0.01
Organic matter digestibility (g kg^{-1} DM)	675.94	677.84	2.596	0.614
Crude protein (g kg^{-1} DM)	125.53	120.04	1.954	0.067
NDF (g kg^{-1} DM)	422.99	426.53	4.611	0.596
UFL (kg DM)	0.64	0.64	0.014	0.709

Table 2. Milk production, milk composition and silage DMI of both lactic acid bacteria treatment (LAB) and control treatment (CONT) with the level of significance for treatment, week and treatment by week interaction.

				Level of significance		
Item	LAB	CONT	S.E	Treatment	Week	Treatment × Week
Milk yield (kg day^{-1})	13.75	13.02	0.771	0.458	0.001	0.273
FPCM yield[4] (kg day^{-1})	16.54	15.03	0.859	0.183	0.001	0.013
Milk fat (kg day^{-1})	0.73	0.65	0.045	0.135	0.001	0.006
Milk protein (kg day^{-1})	0.56	0.55	0.031	0.813	0.001	0.341
Milk solids (kg day^{-1})	1.29	1.19	0.068	0.266	0.001	0.016
Silage DMI (kg cow^{-1})	12.84	12.32	0.339	0.205	-	-

Conclusions

The inoculation of grass silage with *L. plantarum* and *L. lactis* did not improve silage quality, DMI and milk production when fed to late lactation dairy cows. However, milk composition increases were observed during the course of the experiment, most notably in milk fat.

References

Carvalho B.F., Sales G.F.C., Schwan R.F. and Ávila C.L.S. (2021) Criteria for lactic acid bacteria screening to enhance silage quality. *Journal of Applied Microbiology* 130, 341-355.

Claffey A., Boland T., Delaby L. and Kennedy E. (2018) Supplementing with grass silage or restricting pasture: strategies to overcoming short term feed deficits in the diet of early lactation dairy cows in early spring. 10th International Symposium on the Nutrition of Herbivores (ISNH), 2018. Cambridge University Press, Cambridge, UK.

Dillon P. and Stakelum G. (1989) Herbage and dosed alkanes as a grass measurement technique for dairy cows. *Irish Journal of Agricultural Research* 28, 104.

Ellis J.L., Hindrichsen I.K., Klop G., Kinley R.D., Milora N., Bannink A. and Dijkstra J. (2016). Effects of lactic acid bacteria silage inoculation on methane emission and productivity of Holstein Friesian dairy cattle. *Journal of Dairy Science* 99, 7159-7174.

IDF (2015) Bulletin of the International Dairy Federation 479/2015. *International dairy federation a common carbon footprint approach for dairy.* International Dairy Federation, Brussels, Belgium.

Mayes R.W., Lamb C.S. and Colgrove P.M. (1986) The use of dosed and herbage n-alkanes as markers for the determination of herbage intake. *Journal of Agricultural Science* 107, 161-170.

McKay Z.C., Lynch M.B., Mulligan F.J., Rajauria G., Miller C. and Pierce K.M. (2019) The effect of concentrate supplementation type on milk production, dry matter intake, rumen fermentation, and nitrogen excretion in late-lactation, spring-calving grazing dairy cows. *Journal of Dairy Science* 102, 5042-5053.

Muck R.E., Nadeau E.M.G., Mcallister T.A., Contreras-Govea F.E., Santos M.C. and Kung L. (2018) Silage review: Recent advances and future uses of silage additives. *Journal of Dairy Science* 101, 3980-4000.

O'Brien D., Moran B. and Shalloo L. (2018) A national methodology to quantify the diet of grazing dairy cows. *Journal of Dairy Science* 101, 8595-8604.

Oliveira A.S., Weinberg Z.G., Ogunade I.M., Cervantes A.A.P., Arriola K.G. ... and Adesogan A.T. (2017) Meta-analysis of effects of inoculation with homofermentative and facultative heterofermentative lactic acid bacteria on silage fermentation, aerobic stability, and the performance of dairy cows. *Journal of Dairy Science* 100, 4587-4603.

Pang D., Yan T. and Krizsan S. (2021) Effect of strategy for harvesting regrowth grass silage on performance in dairy cows. *Journal of Dairy Science* 104, 367-380.

Grass availability and silage supplementation impact on enteric methane emissions in early lactation

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Abstract

Spring grass availability and growth can vary from year to year and, as such, grass deficits can result in supplementation to meet the intake requirements of lactating dairy cows. The objective of this experiment evaluated the impact of silage supplementation in early lactation; high low grass (HLG) vs low high grass (LHG), on methane emissions (CH_4). Forty spring calving dairy cows were randomly assigned to two treatments (Diet) in a crossover design. For the first six weeks (P1), HLG allocated a high daily herbage allowance (DHA) with no silage supplementation, with the LHG allocated a low DHA with 3 kg dry matter cow^{-1} of grass silage fed daily. Treatments were then crossed over at week 6 for the remaining 6 weeks (P2). This was followed by a 10-week carryover period (P3) with both groups managed similarly. Daily CH_4 was measured using greenfeed emissions monitoring units. Diet had a significant (P<0.05) effect on daily CH_4 emissions, particularly in periods of silage supplementation. Daily CH_4 emissions (g day^{-1}) and CH_4 intensity (g kg^{-1} milk solids) were impacted (P<0.05) by period due to lower CH_4 emissions in P1 compared to P2. Increased levels of silage in the diet impact both daily CH_4 emissions and CH_4 intensity.

Keywords: methane emissions, daily herbage allowance, silage

Introduction

Ruminant livestock enteric methane (CH_4) emissions account for 64% of Ireland's agricultural greenhouse gas emissions (Lanigan *et al.*, 2019). Ireland has now committed to reducing agricultural greenhouse gas emissions by 22-30% by 2030 (DECC, 2021). As a result, it is imperative to correctly quantify and validate the amount of CH_4 emitted by ruminant livestock systems. In pasture-based dairy systems, such as those seen in Ireland, the primary focus is on a spring calving pattern that maximizes the utilization of grazed grass with the goal of increasing farm profitability (Hanrahan *et al.*, 2018). Dairy cows consume 77% of their annual diet from grazed grass (O'Brien *et al.*, 2018); however, early lactation can be a period of reduced grass supply, with increased levels of supplementation (Claffey *et al.*, 2018, Kennedy *et al.*, 2007). At pasture, modern CH_4 emission measurement technologies (Waghorn *et al.*, 2016) have permitted a better knowledge of the factors that influence the level of CH_4 emitted during this period. Daily herbage allowance (DHA) is a key factor dictating the level of animal performance in early lactation (Claffey *et al.*, 2019). However, intense grazing is viewed as a potential strategy for mitigating CH_4 emissions (Gerber *et al.*, 2013), with DHA and supplementation key factors impacting CH_4 emissions in pasture based dairy systems (O'Neill *et al.*, 2012). The objective of the current experiment evaluated the impact of DHA and grass silage supplementation on gross CH_4 emissions (g day^{-1}) and CH_4 intensity (g kg^{-1} milk solids) in early lactation.

Materials and methods

This study was conducted at the Animal and Grassland Research and Innovation Centre, Teagasc Moorepark, Ireland. A twelve-week cross-over design experiment was established followed by a 10-week carry over period in early lactation. Over period 1 (P1), one week after calving, cows were allocated to one of two treatments (Diet) (n=20); high low grass (HLG), low high grass (LHG). All animals were randomized and balanced for breed, parity, lactation number, calving date, economic breeding index, milk production, bodyweight and body condition score. In the first 6 weeks of the experiment, the HLG received a high DHA (9.7 kg dry matter (DM) cow^{-1}) with the LHG receiving a low DHA (7.3 kg DM cow^{-1}) with 3 kg DM of grass silage fed daily. Over the second 6 weeks (P2) treatment diets were crossed over with the HLG now receiving a low DHA (11.6 kg DM cow^{-1}) with 3 kg DM of grass silage fed daily while the LHG received a high DHA (15.2 kg DM cow^{-1}) with no silage supplementation. Cows were allocated the same level of concentrates (average 2.26 kg cow^{-1} day^{-1}) throughout the experiment. During difficult grazing conditions due to adverse weather, particularly in P1, animals were housed and fed grass silage; however, a 3 kg DM cow^{-1} differential in silage intake was maintained between treatments. After P2 both groups were managed similarly for the following 10 weeks to monitor carryover (P3), cows were allocated a similar DHA and no silage supplemented. Cows were milked twice daily with milk yields measured daily and milk composition determined weekly from one successive morning and evening milk sample. The C-Lock Greenfeed Emissions Monitoring (GEM) system was used to monitor daily CH_4 emissions. Cows of each treatment had free access to a GEM unit daily. Daily visits to each GEM unit was similar between treatments (HLG 2.0±0.6 n day^{-1}, LHG 2.1±0.6 n day^{-1}) with visit duration above the recommended average (207.2±17.1 s). Weekly milk data and fortnightly CH_4 data was averaged within each period. Statistical analysis was carried out using a PROC mixed model in SAS 9.4 (SAS Insistitue Inc, Cary, USA), with CH_4 emissions and CH_4 intensities analysed for each period with the model contained terms for diet, period and their associated interactions, period was the repeated measured and animal the random factor.

Results and discussion

Milk solid production, daily CH_4 emissions and CH_4 intensity over the experiment are presented in Table 1 and Figure 1. Silage in the diet over P1 and P2 had a significant effect ($P<0.05$) on daily CH_4 emissions. Cows receiving a lower DHA supplemented with grass silage emitted a greater level of CH_4 in P1 for the LHG and over P2 for the HLG (Figure 1). Daily CH_4 emissions and CH_4 intensity were lower in P2 compared to P1, resulting in a significant effect of period ($P<0.05$). During P1, poor grazing conditions led to higher levels of silage supplemented with reduced DHA, but as grazing conditions improved in week 5, DHA increased and CH_4 emissions decreased in both groups (Figure 1). Periods with the greatest level of silage supplementation resulted in significant diet differences but also a diet by period interaction ($P<0.05$) for CH_4 intensity particularly in P2 where the HLG had higher CH_4 intensity. O'Neill *et al.* (2012), reported similar findings, with animals on a partial mixed ration and a low DHA emitting the greatest levels of CH_4 compared to the high grass allowance treatment. This became evident over P3 as CH_4 emissions was similar between both groups. Bielak *et al.* (2016) reported a significant increase in CH_4 emissions over time post-partum, although the opposite trend was evident in the current experiment with CH_4 emissions lower ($P<0.05$) in P3 compared to P1.

Table 1. Milk solid production and daily CH_4 emissions and CH_4 intensity within each period, with the level of significance of diet, period and their interaction.

	P1		P2		P3		Level of significance			
Item	HLG	LHG	HLG	LHG	HLG	LHG	SEM	Diet	Period	Diet × Period
Milk solids (kg day^{-1})	1.98	1.92	2.08	2.13	1.75	1.79	0.032	0.155	0.003	0.843
CH_4 (g day^{-1})	321	344	323	315	314	324	4.0	0.005	0.017	0.164
CH_4 (g kg^{-1} milk solids)	165	176	163	154	188	187	2.1	0.719	0.001	0.017

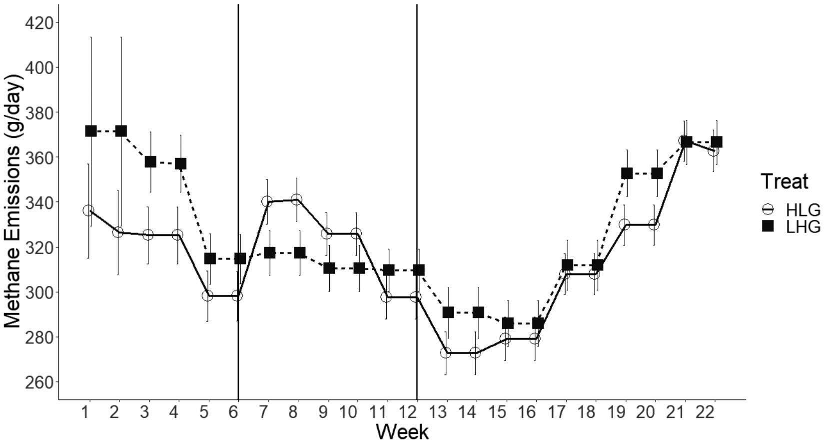

Figure 1. Daily CH_4 emissions (g day^{-1}) between treatments over the course of the experiment.

Conclusions

Silage in the diet of cows in early lactation can have a significant impact on CH_4 emissions. Higher levels of grass silage in the diet resulted in greater levels of emitted CH_4 (g day^{-1}) and CH_4 intensity (g kg^{-1} milk solids) clearly during periods with highest level of silage supplementation.

References

Claffey A., Boland T., Delaby L. and Kennedy E. (2018) Supplementing with grass silage or restricting pasture: strategies to overcoming short term feed deficits in the diet of early lactation dairy cows in early spring. 10th International Symposium on the Nutrition of Herbivores (ISNH), Cambridge University Press, Cambridge, UK.

Claffey A., Delaby L., Lewis E., Boland T.M. and Kennedy E. (2019) Pasture allowance, duration, and stage of lactation – Effects on early and total lactation animal performance. *Journal of Dairy Science,* 102, 8986-8998.

DECC 2021. Climate Action Plan 2021. Department of the Environment, Climate and Communications, Dublin, Ireland.

Gerber P.J., Henderson B. and Makkar H.P. (2013) *Mitigation of greenhouse gas emissions in livestock production: a review of technical options for non-CO_2 emissions*, Food and Agriculture Organization of the United Nations (FAO).

Hanrahan L., McHugh N., Hennessy T., Moran B., Kearney R., Wallace M. and Shalloo L. (2018) Factors associated with profitability in pasture-based systems of milk production. *Journal of Dairy Science* 101, 5474-5485.

Kennedy E., O'Donovan M., O'Mara F.P., Murphy J.P. and Delaby L. (2007) The effect of early-lactation feeding strategy on the lactation performance of spring-calving dairy cows. *Journal of Dairy Science* 90, 3060-3070.

Lanigan G., Donnellan T., Hanrahan K., Carsten P., Shalloo L., Krol D., Forrestal P.J., Farrelly N., O'Brien D. and Ryan M. (2019) An analysis of abatement potential of Greenhouse Gas emissions in Irish agriculture 2021-2030. Teagasc, Ireland.

O'Brien D., Moran B. and Shalloo L. (2018) A national methodology to quantify the diet of grazing dairy cows. *Journal of Dairy Science* 101, 8595-8604.

O'Neill B.F., Deighton M.H., O'Loughlin B.M., Galvin N., O'Donovan M. and Lewis E. (2012) The effects of supplementing grazing dairy cows with partial mixed ration on enteric methane emissions and milk production during mid to late lactation. *Journal of Dairy Science* 95, 6582-6590.

Waghorn G.C., Jonker A. and Macdonald K.A. 2016. Measuring methane from grazing dairy cows using GreenFeed. *Animal Production Science* 56, 252.

The impact of dairy cow genotype on methane emissions within a grazing dairy system

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Abstract

The objective of this study was to evaluate the impact of genetic selection using the economic breeding index (EBI) on methane (CH_4) emissions within a grass based dairy system. Two genetic groups (GG) of Holstein-Friesian dairy cows were assembled: (1) a high EBI group (Elite; n=22); and (2) a National Average EBI group (NatAv; n=23). Methane emissions and milk production were measured over a 34-week period (early March to mid October). The results showed no difference (P>0.05) in milk yield between both GG. The Elite had greater milk fat % and protein % (P<0.001) compared to the NatAv, which resulted in the Elite producing greater quantities of milk solids (fat + protein kg; P<0.001). Daily CH_4 emissions did not differ between GG (P>0.05), but CH4 emissions in g kg^{-1} milk solids was lower for Elite than NatAv (P<0.05). The results of this study demonstrate that selection using high EBI genetics can improve milk solids output without increasing CH_4 emissions and also reduce CH_4 emitted per kg of milk solids within grass-based dairy systems.

Keywords: methane, dairy cows, genetics

Introduction

Increasing levels of scrutiny relating to the environmental impact of the dairy industry has led to growing levels of attention in developing approaches for improving sustainability. Breeding should be considered a viable strategy to improve sustainability as breeding is cumulative and permanent meaning genetic progress will be compounded with successive generations. The economic breeding index (EBI) was developed to breed cows suited to Irish grass-based systems of milk production (Veerkamp *et al.*, 2002). Research using the life cycle assessment methodology has demonstrated that the EBI is delivering improved environmental credentials through lowering greenhouse gas (GHG) emissions per unit of milk solids output (Lahart *et al.*, 2021). These calculations are similar to the IPCC methodology, which calculates methane (CH_4) emissions based on energy requirements for milk production, maintenance, pregnancy and body weight change (IPCC, 2019). To date, no research has been completed across the grazing season with measured CH_4 emissions between animals that are divergent for EBI. The objective of the current study was to investigate the impact of genetic selection using EBI on CH_4 emissions.

Materials and methods

This study was undertaken at the Dairygold Research Farm (Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland). Two genetic groups (GG) of Holstein-Friesian dairy cows were assembled: (1) a high EBI group (Elite; n=22), representative of the top 5% of dairy cows for the Economic Breeding Index (EBI; mean EBI of €233, standard deviation (SD) = 20.9); and (2) a National Average group (NatAv; n=23; mean EBI of €133, SD=26.5), representative of the average dairy cow for the EBI. The mean calving date of the cows used in the study was the 14th (SD=12.9) of February. The animals were split into two equally sized groups, balanced for GG, parity and calving date, which grazed separately on two individual farmlets. Both groups were managed identically, with target pre- and post-grazing residual sward heights of 9 and 4.5 cm, respectively. Each grazing group had access to a GreenFeed (C-Lock; Rapid City, SD, USA) for the purpose of CH_4 measurement.

Animals were trained to use the GreenFeed units over a 4-week period prior to the beginning of the experiment. Animals were enticed to visit the GreenFeed by offering a small quantity of concentrate at each visit. The experimental period lasted from early March to mid-October (34 weeks). Concentrate (2 kg cow^{-1} day^{-1}) was offered to the cows (1 kg from the GreenFeed and 1 kg from the milking parlour) throughout the study. Methane was recorded on a daily basis using the GreenFeed units. All cows were milked twice daily and samples to measure milk fat and protein % were collected on a weekly basis. Milk production and CH_4 variables were analysed using the MIXED procedure of SAS 9.4 (SAS Institute Inc., Cary NC, USA). Terms for GG, parity, week, grazing group and calving date centred within genotype were included in the model.

Results and discussion

Mean (SD) for pre- and post-grazing residuals throughout the experiment were 9.6 (0.07) cm and 4.5 (0.09) cm. The number of daily visits to the GreenFeed averaged 2.5.

The effect of GG on milk yield, milk constituents, milk solids (MS), CH_4 emissions and CH_4 kg^{-1} MS is presented in Table 1. There was no significant difference in milk yield between the Elite and NatAv (P>0.05). The Elite had greater milk fat and protein % (P<0.001), which resulted in an 8% greater (P<0.001) MS yield compared to the NatAv. There was no significant difference in CH_4 emissions between the Elite and NatAv (P>0.05). Similar to Lahart *et al.* (2021), the increased MS yield of the Elite resulted in dilution of their CH_4 emissions on a per unit of output basis resulting in 6% less CH_4 emitted per kg of MS (Figure 1).

Table 1. The effect of genetic group of Holstein-Friesian (Elite = high EBI; NatAv = national average EBI) on milk yield, milk constituents, milk solids yield (kg fat + protein), methane emissions and methane per kg of milk solids.[1]

Trait	Elite	NatAv	SE	*P*-value
Milk yield (kg d^{-1})	22.3	22.2	0.45	0.901
Milk fat %	5.00	4.50	0.091	<0.001
Milk protein %	3.84	3.67	0.033	<0.001
Milk solids (kg d^{-1})	1.94	1.79	0.028	<0.001
Methane (g d^{-1})	301	297	6.5	0.618
Methane / milk solids (g kg^{-1})	163	174	3.9	0.031

[1] SE = standard error.

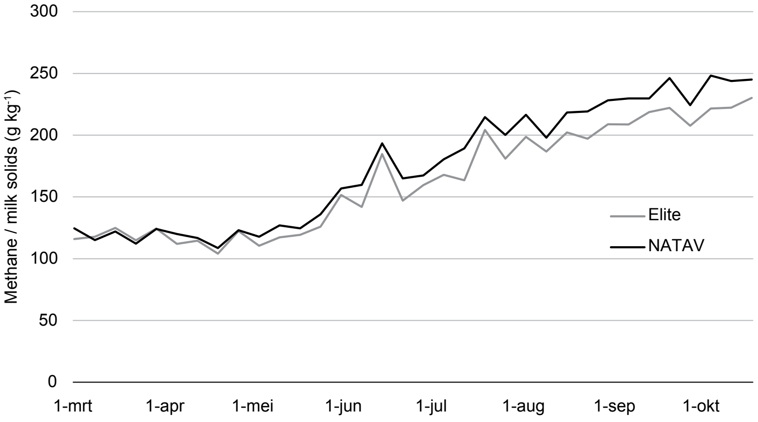

Figure 1. The impact of genotype (Elite = high EBI; NatAv = national average EBI) on daily methane per unit of milk solids across the experimental period.

Conclusions

Within this study, selection using the EBI delivered increased milk solids production without any increase in CH_4 emissions, which is crucial in relation to national GHG inventories. The increased productivity of high EBI cows leads to a dilution of their CH_4 emissions and a reduction in CH_4 emitted per unit of MS output. Further work is required to elucidate the mechanisms responsible for the increased efficiency of the Elite cows. The inclusion of these effects in a carbon footprint model would increase the benefit associated with the Elite animals.

Acknowledgements

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References

IPCC (Intergovernmental Panel on Climate Change) (2019) Refinement to the 2006 IPCC guideline for National Greenhouse Gas Inventories. Volume 4 Agriculture, Forestry and Other Land Use.

Lahart B., Shalloo L., Herron J., O'Brien D., Fitzgerald R., Boland T.M. and Buckley F. (2021) Greenhouse gas emissions and nitrogen efficiency of dairy cows of divergent economic breeding index under seasonal pasture-based management. *Journal of Dairy Science* 104, 8039-8049.

Veerkamp R.F., Dillon P., Kelly E., Cromie A.R. and Groen A.F. (2002) Dairy cattle breeding objectives combining yield, survival and calving interval for pasture-based systems in Ireland under different milk quota scenarios. *Livestock Production Science* 76, 137-151.

Effects of day or night grazing schedule on milk production and methane emissions at high latitudes

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Abstract

The aim of this study was to compare enteric methane (CH_4) emissions and milk yield from dairy cows under a day or night grazing management at high latitudes in summer time. Twenty-six Swedish red dairy cows were allocated to one of two treatments: one represented 10 h day-time pasture access and the other 12 h night-time pasture access. Each treatment received the same total mixed ration indoors and were offered strip grazing with *ad libitum* herbage allowance. Cows were adapted to their treatments during the first 4 weeks and the last week, from 26 June, was the recording period. By using a system of concentrate feeders with an integrated gas measurement (GreenFeed®), individual CH_4 emissions were recorded both on pasture and inside the barn. Milk yield, body weight and feed intake were individually recorded daily. Using a mixed model, milk yield and CH_4 emissions were analysed with cow as random effect, treatment (DAY and NIGHT) as fixed effect and body weight as covariate. No significant differences were found on milk yield (26.3 and 25.1 kg cow^{-1} d^{-1}) or CH_4 emissions (375 and 368 g cow^{-1} d^{-1}) for DAY and NIGHT, respectively. We conclude that a day or night grazing management has no effect on milk yield or CH_4 emission under the conditions evaluated.

Keywords: dairy, GreenFeed, Nordic, management, GHG, pasture

Introduction

In the Nordic countries, most of the information regarding methane (CH_4) emissions from dairy production is based on indoor recordings and therefore further research from grazing dairy cows should be undertaken. The main driver for enteric CH_4 emissions from dairy cows is dry matter intake (DMI; Ramin and Huhtanen, 2013). Estimation of DMI at pasture is complex and influenced by numerous factors, such as herbage characteristics, animal behaviour, grazing system, feeding strategy and environment (Gregorini *et al.*, 2006). Previous studies have shown that dairy cows have a diurnal grazing behaviour with two main grazing bouts observed; one in the morning and one in the evening (Linnane *et al.*, 2001). The specificity of high latitude lands, like the Scandinavian countries, is that the days have a very long photoperiod in summer, up to 24 h of daylight. Therefore, it is of interest to investigate the effects of grazing period on greenhouse gas emissions and milk yield (MY) in this specific environment and geographical region. The main objectives of this experiment were to compare DMI, MY and enteric CH_4 emissions from dairy cows under a day-time or night-time grazing at high latitudes in northern Sweden.

Materials and methods

A five weeks grazing trial was conducted from 1st of June to 2nd of July 2021 at the SLU experimental farm of Röbäcksdalen, Umeå, Sweden (63.81°N, 20.23°E). Twenty-six dairy cows (Swedish Red) were allocated to two treatment groups: 10 h access to pasture during day-time (DAY) or 12 h during night-time (NIGHT). The DAY group had pasture access from 07:00 to 17:00 h and the NIGHT group, from 18:00 to 06:00 h each group offered a new grass strip daily. While not at the pasture, the animals were kept in the same free stall barn equipped with a 2×8 herringbone-milking parlour. Rotations between pasture and barn occurred at 06:00 and 17:00 h each day after milking. The experimental cows were allocated to a treatment, 11 in the DAY and 15 in the NIGHT, by balancing for days in milk, MY and parity and resulted on average (±standard deviation), body weight (BW) of 633±24.7 and 597±18.4 kg, days in milk of 168±31.7 and 206±32.4, parity of 1.5±0.25 and 1.5±0.19, MY of 27±2.2 and 28±1.5 kg, respectively.

Each treatment received the same *ad libitum* total mixed ration (TMR) indoors and cows were offered strip grazing with *ad libitum* herbage allowance (18 kg DM cow^{-1} d^{-1}). The TMR was composed, on DM basis, of 430 g kg^{-1} silage, 557 g kg^{-1} concentrate, 13 g kg^{-1} minerals. Additional concentrates were also distributed through the GreenFeed® (GF) and concentrate feeder, accounting for 78 g kg^{-1} DM. With grazing, this resulted in a total diet ratio of 60% forage and 40% concentrate in the two treatments (Table 1). During the first four weeks, all animals were adapted to their indoor diets, grazing treatments, trained to visit the GF and the last week was used as the recording period (26 June to 2 July).

For measuring the cows' CH_4 emissions, two GF units with integrated gas measurement equipment were used: one mobile unit on the pastures and one unit with a fixed location in the barn. The mobile GF was moved twice daily to the new grass strip offered. Registrations of MY and BW were automatically conducted daily using automatic recorders. The indoor voluntary intake was recorded on individual basis using automated feed bunks. Daily herbage dry matter intake (HDMI) was estimated at group level using a raising plate meter to measure grass height before and after grazing and divided by number of grazing animals. Grass samples were collected and analysed for nutritional content.

The effects of grazing treatments on MY, indoor DMI, and CH_4 emissions were analysed using the mixed model applied on SAS 9.4 release software (2013) with cow as random effect, treatment (DAY and NIGHT) as fixed effect and body weight as covariate. T-tests were used to identify differences between HDMI and visits to the GF between the treatment groups.

Results and discussion

Access time to pasture did not seem to be a limiting factor (P=0.53) as daily estimated herbage DMI were 4.3 (±0.3) and 4.6 (±0.4) kg for AM and PM, respectively (Table 2). The cows in the AM group had shorter access period to pasture (2 hours less) and achieved a similar estimated feed intake as cows in the PM group. The diurnal grazing behaviour observed by Linnane *et al.* (2001) in Ireland split between morning and late afternoon bouts cannot be transferred to Scandinavian summer conditions with 24-h light. Behavioural observations conducted from the experiment may show the animals' grazing behaviour.

Table 1. Feed nutrient content during the recording week.

Characteristics	DM (g kg^{-1})	NDF (g kg DM^{-1})	CP (g kg DM^{-1})	iNDF (g kg DM^{-1})	WSC (g kg DM^{-1})
Pasture	260	411	186	68.3	161
Total mixed ration					
Silage	305	513	148	177	15
Concentrate	880	225	180	-	-
Protein concentrate	890	270	350	-	-
Minerals	-	-	-	-	-
Additional concentrate	890	270	350	-	-

Table 2. Methane emissions, milk yield, dry matter intake indoor, body weight, visit GreenFeeds and estimated herbage dry matter intake of cows offered pasture during DAY or NIGHT.

Variable (unit)	DAY		NIGHT		Significance
	Mean	SE Mean	Mean	SE Mean	
Methane emissions (g d^{-1})	375	16.1	368	13.9	0.75
Milk yield (kg d^{-1})	26.3	1.93	25.1	1.65	0.64
Indoor DMI (kg d^{-1})	15.3	0.73	14.6	0.63	0.49
Body weight (kg)	645	6.5	602	5.8	0.25
Visits to GreenFeed (n)	3.6	0.11	3.1	0.11	<0.05
Estimated HDMI (kg d^{-1})	4.3	0.30	4.6	0.44	0.53

The indoor DMI did not differ significantly (P=0.49) between treatments (Table 2) leaving a total daily DMI of recorded TMR, supplementary feed DMI and HDMI, to about 19 kg DM cow^{-1} in both groups. The treatments had no significant effect on milk production (26.3 and 25.1 kg cow^{-1} d^{-1}; P=0.64) or CH_4 emissions (375 and 368 g cow^{-1} d^{-1}; P=0.75). The non-significance of estimated total DMI between the two groups is in line with the CH_4 data showing no effect on CH_4 and MY (Table 2). Ramin and Huhtanen (2013) showed that DMI is the main driver for CH_4 emissions in their meta-analysis based on indoor feeding. The result published by Waghorn *et al.* (2013) highlighted a positive correlation between DMI and CH_4 emission on a full-time grazing trial. Our experiment tested partial grazing and we found that CH_4 emissions were the same between treatments with similar DMI.

Cows in the DAY group had more visits to the GF units (3.6 visits) that cows in the NIGHT group (3.1 visits; P<0.05). Unit visits were twice as high for the indoor unit (2.2 visits d^{-1}) compared to the outdoor unit (1.1 visits d^{-1}). The DAY group stayed indoors 2 h longer, which may explain the differences in visits, but we still noticed less interest to the outdoor unit in both groups.

Conclusions

This experiment demonstrated it was possible to record CH_4 emissions with two GreenFeed units linked, one outdoor and one indoor. The dry matter intake, milk yield and methane emissions were not significantly affected by the two treatments.

References

Gregorini P., Tamminga S. and Gunter S.A. (2006) Review: behavior and daily grazing patterns of cattle. *Professional Animal Science* 22, 201-209.

Hernández-Ortega M., Martínez-Fernández A., Soldado A., González A., Arriaga-Jordán C.M., Argamentería A. ... and Vicente F. (2014) Effect of total mixed ration composition and daily grazing pattern on milk production, composition, and fatty acids profile of dairy cows. *Journal of Dairy Research* 81, 471-478.

Linnane M.I., Brereton A.J. and Giller P.S. (2001) Seasonal changes in circadian grazing patterns of Kerry cows (Bos Taurus) in semi-feral conditions in Killarney National Park, Co. Kerry, Ireland. *Animal Behaviour Science* 71, 277-292.

Ramin, M., and Huhtanen P. (2013) Development of equations for predicting methane emissions from ruminants. *Journal of Dairy Science* 96, 2476-2493.

SAS Institute Inc. (2013). *Base SAS 9.4®. SAS User's Guide: Statistics.* SAS Institute Inc., Cary, NC, USA.

Waghorn G.C., Jonker A. and Macdonald K.A. (2016) Measuring methane from grazing dairy cows using GreenFeed. *Animal Production Science* 56, 252-257.

Sheep grazing semi-natural pastures on islands in northern Norway

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Abstract

The Norwegian sheep industry is based on utilization of 'free' rangeland pasture resources. Use of mountain pastures is dominating, with about two million sheep grazing these pastures during summer. Regional challenges related to e.g. loss of sheep to large carnivores, make the Norwegian coast, especially the islands, attractive for sheep farming. Use of islands for summer pasture is an alternative, but knowledge about required management is defined by decades-long experience by farmers with little scientific knowledge. We examined average daily gain from week 21 to week 37 of 230 lambs distributed on three islands (Sandvær, Sjonøya and Buøya) for 3 years (2012, 2013 and 2014). Lambs were born during May and were 1 to 4 weeks of age at release. Pasture quality and stocking rate differed between islands. At Sandvær, 92% of the island was characterized as high nutritional value while this was the case for only 15% at Sjonøya and Buøya. We found an average daily lamb growth rate of 0.320 kg d^{-1}. Lambs on Sandvær had a higher daily gain (P<0.05) than those on Sjonøya and Buøya. We conclude that there is a potential to utilize islands for sheep grazing during summer.

Keywords: daily growth rate, lamb, stocking rate, management

Introduction

The Norwegian sheep industry is based on utilization of spatially diverse rangeland pasture resources as reflected in different management systems and local adaptations. Mating takes place indoors during November/December with lambing in April/May. Ewes and lambs are released to pastures soon after lambing and ewes are nursing their lambs until gathering in September. Lambs big enough (minimum 23 kg carcass weight) are then slaughtered. Only 3% of Norway is used for crop production, but more than 50% of land area has potential value as livestock pasture. Approximately 2 million sheep are, each summer, released onto extensive pastures for grazing (SSB, 2019). Rangeland pastures in mountainous areas are the dominant grazing area for the sheep but regional challenges due to high mortality to large carnivores have increased the interest in utilizing pastures on islands and islets along the coast.

Nordland county, stretching from 65°N to 69°N has a coastline estimated at about 27,000 km of which 21,000 km are island coastlines. The Nordland coast is scattered with some 18,000 islands of all sizes. Most of these islands are flat (rising to 40 to 50 m above sea level) and natural fresh water supply can be limited during summer. The climate is dominated by mild winters and wet summers. Vegetation types, their proportion and distribution and thus pasture value varies substantially between islands (Rekdal, 2001). Among the benefits of using mountain pastures are the diverse vegetation, the young phenological stages of plants, high in nitrogen and low in fibre resulting from the snow line retreating upwards. On the islands, phenological stage is species-specific and depending on water. Thus, management of stocking rate customized to available pasture resources is necessary to ensure animals' performance and welfare. However, stocking rates are defined by the farmers who have little scientific knowledge about sheep performance on such pastures. The aim of the study was to describe lamb daily weight gain on three islands with varying pasture quality and stocking rate.

Materials and methods

Three islands situated in Lurøy and Rødøy municipalities, Nordland, Norway were used in the study. Sandvær (66°20′35 N, 12°43′55 E) covers 39 ha and rises to 20 meters above sea level (m.a.s.l.), Sjonøya (66°21′51 N, 12°52′42 E) covers approximately 208 ha (40 m.a.s.l) while Buøya (66°37′31 N, 12°56′35 E) covers 36 ha (40 m.a.s.l). The total livestock unit (LU) at Sandvær, Sjonøya and Buøya were 1.3, 9.2 (including a flock of Old Norwegian Sheep) and 2.7, respectively, on average over three years (2012, 2013 and 2014). Vegetation mapping of the islands identified a total of 19 different vegetation types, both natural and semi-cultivated (Lind *et al.*, 2020). We classified the vegetation types into four main classes based on value for sheep grazing: 'Not Suitable' (no grazing value or inaccessible), 'low', 'medium, or 'high'. Vegetation types of high nutritional value contain species such as common bent (*Agrostis capillaris*), sweet vernal grass (*Anthoxanthum odoratum*), Kentucky bluegrass (*Poa pratensis*) and red fescue (*Festuca rubra*). Wavy hairgrass (*Deschampsia flexuosa*), blueberry (*Vaccinium myrtillus*) and sweet vernal grass (*Anthoxanthum odoratum*) are found in medium nutritional value classes while the low nutritional value class is dominated by crowberry (*Empetrum nigrum*), heather (*Calluna vulgaris*) and purple moor-grass (*Molinia caerulea*).

Two commercial farmers randomly selected adult ewes (>2 years) with twin lambs from their flocks of the breed Norwegian White sheep. The animals had access to all vegetation types within each island from June to August during each of the three years. No supplementary feeding was offered during the grazing period. The animals were weighed before, twice during, and after the grazing period, and average daily weight gain was calculated. After the animals were gathered, they returned to commercial production of the farmers. Data on a total of 230 twin lambs were analysed using general mixed linear model in Proc Mixed of SAS statistical software (SAS Institute Inc., 2015). Lamb age at release (average 16 days) and ewe weight (average 74 kg) were regression variables. Lamb sex (male or female), ewe age (average 3 years), islands and years were considered class variables.

Results and discussion

Lambs' average daily gain on the island pastures was 0.320 kg d^{-1} (standard deviation (SD)=0.067 kg d^{-1}), and they spend on average 89 days on the islands (SD=13 days). The results show that age of ewe, weight of ewe and sex of lamb, significantly (P<0.05) affected lamb growth. Lamb age at release had no effect on growth rate on the islands (P=0.66). The daily weight gain is similar to the average weight gain of Norwegian White Sheep lambs at 0.290 kg d^{-1} reported for the country (Animalia, 2019).

As much as 92% of the area of Sandvær is characterized as high nutritional value which here includes the vegetation types of low herb meadow, high forb meadow, moist meadow and pasture (former cultivated areas) (Rekdal, 2001). The pasture alone covering 12 ha could sustain 3.6 licestock units (LU) while only 1.3 LU grazed the island every summer. Forage quality is maintained when grazed at an optimum stocking rate but when it is too low non-grazed areas will degrade. The result at Sandvær was meadowsweet (*Filipendula ulmaria*) dominating the pastures, a plant with little grazing value for sheep (French, 2017).

At Sjonøya, about 80% of the area is characterized as low nutritional value with the island dominated by coastal heath (31%) and damp heath (41%). Most of the remaining area is classified as medium to high nutritional value (low herb meadow, meadow birch forest and pasture). The stocking rate was estimated to 9.2 LU and the capacity to 12 LU thus close to a maximum. However, lamb daily gain on Sjonøya was significantly lower than that of both Sandvær and Buøya. Sjonøya consists of 4 smaller islands connected only at low tide. Most of the cultivated pasture type is located on one of them and sheep could be temporarily stranded on an island with mostly low nutritional value vegetation types.

At Buøya six vegetation types were present and the island is dominated by low nutritional value classes (86%). On this island, high nutritional value is only found on patches of pasture (14%). According to Rekdal (2001) the island could carry around 2.5 LU and we calculated 2.7 LU at the island. Lambs' growth rate was significantly lower on this island compared to Sandvær and could be explained by the animals being forced to graze in medium and low nutritional value vegetations types in addition to the high nutritional vegetation types.

The investigated islands all had a high degree of plant species diversity. Over a three-month period, the nutritional value-change would be species-specific and influenced by general phenological development as well as the within-year impact of grazing. A dynamic management plan when using island pastures is important. Ideally, the stocking rate should be higher in the spring when high quality forage is in abundance. During the summer, the lambs' need for high-quality forage increase while at the same time the pasture quality declines. The pasture quality can to some extend be maintained if the stocking rate is adjusted during the grazing season. To release and collect the animals at the right time are therefore critical for an optimum production. In the present case, some animals could have been gathered earlier when they were big enough for slaughter, while other lambs needed additional fattening after they were collected.

Conclusions

The average daily weight gains of lambs raised on the three investigated islands are similar to the weight gain of the breed at a national level. A dynamic and adaptive management strategy for release and collection of the animals to the islands, following the phenological development of pastures, is important. There are potential benefits for increasing the use of island pastures, not least to avoid predator-livestock conflicts.

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References

Animalia (2019) Årsrapport 2018 [Annual report 2018] www.animalia.no/globalassets/sauekontrollen---dokumenter/arsmelding-sau-2018.pdf.

French K.E. (2017) Species composition determines forage quality and medicinal value of high diversity grasslands in lowland England. *Agriculture, Ecosystems and Environment* 241, 193-204

Lind V., Holand Ø., Haugen F-A. and Steinheim G. (2020) Lamb performance on island pastures in Northern Norway. *Frontiers in Veterinary Science* 7, 402.

Rekdal Y. (2001) Husdyrbeite i fjellet, vegetasjonstypar og beiteverdi. [Livestock grazing in the mountains. Vegetation types and forage value]. *NIJOS-rapport* 7/01. Norsk institutt for jord- og skogkartlegging, Ås, Norway.

SAS Institute Inc. (2015) *Base SAS 9.4 Procedures Guide. 5th ed*. SAS Institute Inc., Cary, NC, USA.

SSB (2019). Holdings, agricultural area, and livestock. Statistics Norway. https://www.ssb.no/en/statbank/table/12660/tableViewLayout1/

Biochar as feed additive to sheep did not affect feed intake, growth rate and enteric methane production

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Abstract

Biochar is pyrolyzed biomass and a good candidate for increasing carbon storage in soils as a method for mitigating atmospheric carbon. There is an increasing interest of biochar as a feed additive in animal systems to improve animal performance and reduce their greenhouse gas emissions. The aim of the current study was to investigate the effect of including biochar in feed to sheep on daily dry matter (DM) intake, lambs' daily growth rate and their enteric methane (CH_4) emissions. Two experiments were conducted using the Norwegian White Sheep breed. The animals were fed grass silage fed *ad libitum* and a formulated concentrate with inclusion of biochar at 0 (control) or 1.2 g kg^{-1} biochar of DM. In Experiment 1, daily feed intake was measured on adult ewes. In Experiment 2, daily feed intake, weight gain and enteric CH_4 emissions were measured on lambs. All animals were stalled individually and CH_4 production was measured using open circuit respiration chambers. We found that inclusion of biochar had no effect on dry matter intake (P=0.389), daily growth rate (P=0.358) or enteric CH_4 emissions (P=0.578). We conclude that more *in vivo* studies with different sources of biochar should be performed.

Keywords: charcoal, GHG emission, *in vivo*, Norway

Introduction

The primary methods by which agriculture is expected to meet emission reduction targets are through reduced greenhouse gas (GHG) emissions and increased carbon storage. Biochar has been proposed internationally as a tool to mitigate GHG emissions and increase carbon storage (Lehmann, 2007). There is documented evidence over several thousand years of the use of biochar as a dietary supplement for both humans and animals for the treatment of digestive complaints (O'Toole *et al.*, 2016). In the last decade, there has been increasing interest in the use of biochar as a feed additive in animal nutrition. A meta-analysis by Schmidt *et al.* (2019) showed that livestock fed biochar had increased mean weight gain of 9.9%, compared to a control diet. Kammann *et al.* (2017) reviewed the use of biochar to reduce enteric GHG emissions and conclude that no groups have repeated the results from Leng *et al.* (2012). Newer studies by Terry *et al.* (2019) confirm that biochar supplemented to Angus × Hereford heifers had no mitigating effect on enteric methane emissions. Mirheidari *et al.* (2020) studied the productivity effects of biochar fed to ram lambs, showing a positive effect on average daily weight gain when added at 1-2% inclusion in dry matter (DM). They did not measure CH_4 emissions, leaving sheep production systems a relatively understudied livestock system in relation to this topic. The aim of the current study was to investigate the effect of including biochar in feed to ewes and lambs on daily DM intake (DMI), lamb daily growth rate and enteric CH_4 emissions.

Materials and methods

Two feeding experiments with Norwegian White Sheep were conducted in an uninsulated barn at NIBIO Tjøtta in Nordland county, Norway (65°49'22 N, 12°25'37 E). To measure daily feed intake (Experiment 1), 20 sheep aged 2 to 6 years (average body weight ± standard deviation of 88.0±12.4 kg) were individually fed grass silage *ad libitum* and 0.4 kg concentrate daily for 20 days. The animals were allocated to two concentrate diets with different inclusion of biochar in g kg^{-1} DM (0 (control) or 4.6) with

10 animals per diet. Grass silage offered per 24 hour and leftovers were recorded to calculate daily feed intake. All feeds were analysed for nutritional composition (DM, ash, neutral detergent fibre (NDF), crude protein (CP), energy) (Table 1). Biochar was produced from a mixture of spruce (*Picea abies*) and pine (*Pinus sylvestris*) wood chips in a Pyreg 500 continuous slow pyrolysis reactor operated by NovoCarbo Gmbh (DE). Pyrolysis temperatures ranged between 500-600 °C.

To measure feed intake, growth rate and enteric CH_4 emissions (Experiment 2), 24 ewe lambs aged 5 months (average bodyweight ± standard deviation: 40.3±5.9 kg) were individually fed grass silage *ad libitum* and 0.4 kg concentrate daily for 35 days. Animals were allocated to the same two concentrate diets as mentioned above with 12 animals per diet. Animals were weighed regularly to calculate individual growth rate. Methane emissions were measured using open circuit respiration chambers (Pinares and Waghorn, 2014). Enteric CH_4 was sampled via plastic tubes into a seven port Servopro Multiexact 4100 Analyzer (Servomex Group Inc, Woburn, MA, USA) on a rotational basis, taking one gas sample reading after 3 min per port, measuring ambient air and each chamber every 21 min. After 20 days of adaptation to their respective diets, the animals were kept in the chambers for 72 h during which they were fed and managed as in the barn. Animal's feed intake was measured daily while in the respiration chambers. Water was available *ad libitum*.

To determine feed intake, daily growth and CH_4 production between diets, data were analysed using ANOVA procedure (2016 Minitab Inc) considering diet as a fixed effect and animals as random effects. When a significant effect of diet was found, *post hoc* comparisons of means were made using Tukey Kramer test. Differences were considered significant at $P<0.05$.

Results and discussion

The 1.2 g kg DM^{-1} inclusion rate of biochar in the concentrate made up 0.72 g kg DM^{-1} in total DMI in Experiment 1. Biochar consists mainly of carbon and thus it could be discussed if inclusion of biochar in the diet of sheep would have an impact on the daily feed intake. In our experiments, DMI ranged from 1.84 kg d^{-1} (control) to 2.02 kg d^{-1} (Experiment 1) and 1.32 to 1.39 g d^{-1} (Experiment 2). None of the results were significantly different between groups. This corresponds with Mirheidari *et al.* (2020) who found no effect on DMI when biochar was added to sheep diet at 2% inclusion rate. Further, the authors found an improved daily weight gain in the sheep fed a biochar diet compared to the control diet. We found a similar trend where lambs' growth rate was numerically higher (NS) for those animals receiving the biochar diet (Table 2). There are limited numbers of *in vivo* experiments reporting the use of biochar as a mitigating tool for CH_4 production from ruminants. The first proposal for using biochar as a dietary CH_4 mitigation tool came from Leng *et al.* (2012). Biochar produced from rice husks fed to 'Yellow' cattle (0.6 g kg DM^{-1}) reduced CH_4 production by 22%. However, the results should be interpreted with caution as they used spot samples for CH_4 measurements. Using open circuit respiration chambers, we did not find any mitigating effect of biochar on enteric CH_4 production (Table 2).

Table 1. Average chemical composition (g kg^{-1} DM) of diet ingredients.[1]

	Experiment 1 (grass silage)	Experiment 2 (grass silage)	Concentrate
DM, g kg^{-1} fresh	460	379	871
Ash	54.0	61.4	58.7
Crude protein	125	117	148
NDF	552	527	137
DOM	713	693	n.a.
FU[2]	0.85	0.81	n.a.

[1]NDF = neutral detergent fibre; DOM = *in vitro* digestible organic matter.

[2] FU Scandinavian Feed Unit: 1 FU = 7.89 MJ net energy.

Table 2. Dry matter intake (g d^{-1} $animal^{-1}$), average growth rate (g d^{-1}), enteric methane production (g CH_4 d^{-1}, g CH_4 kg DMI^{-1}).

	Control	Biochar	SEM[1]	*P*-value
Experiment 1 sheep (n=20)				
Dry matter intake	1.84	2.02	0.36	0.550
Experiment 2 lambs (n=24)				
Dry matter intake	1.32	1.39	0.04	0.389
Growth rate	133.5	155.8	25.0	0.358
CH_4 g d^{-1}	24.8	24.1	1.04	0.729
CH_4 g kg^{-1} DMI	0.94	1.0	0.04	0.386

[1] SEM = standard error of the mean.

Conclusions

We conclude that biochar up to 1.2 g kg DMI^{-1} had no negative impact on total DMI. Biochar did not increase daily growth rate significantly and had no mitigating effect on enteric methane production from Norwegian sheep. Further *in vivo* studies should be performed.

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References

Kammann C., Ippoplito J., Hagemann N., Borchard N., Cayuela M.L., Estavillo J.M. ... Wrage-Mönnig, N. (2017) Biochar as a tool to reduce the agricultural greenhouse-gas burden – knowns, unknowns and future research needs. *Journal of Environmental Engineering and Landscape management* 25, 114-139.

Lehmann J. (2007) A handful of carbon. *Nature* 447, 143.

Leng R.A., Preston T.R. and Inthapanya S. (2012) Biochar reduces enteric methane and improves growth and feed conversion in local 'Yellow' cattle fed cassava root chips and fresh cassava foliage. *Livestock Research for Rural Development* 24, 199.

Mirheidari A., Torbatinejad N.M., Shakeri P. and Mokhtarpour A. (2020) Effects of biochar produced from different biomass sources on digestibility, ruminal fermentation, microbial protein synthesis and growth performance of male lambs. *Small Ruminant Research* 183, 106042.

O'Toole A., Andersson D., Gerlach A., Glaser B., Kammann C., Kern J., ... Stenström, J. (2016) Current and future applications for biochar. In: Shackley S., Ruysschaert G., Zwart K. and Glaser B. (eds.) *Biochar in European soils and agriculture: science and practice*. Routledge, London, UK.

Pinares C. and Waghorn G. (2014) *Technical manual on respiration chambers designs*. Ministry of Agriculture and Forestry, New Zealand.

Schmidt H., Hagemann N., Draper K. and Kammann C. (2019) The use of biochar in animal feeding. *PeerJ* 7, e7373.

Terry A., Ribeiro G.O., Gruninger R.J., Chaves A.V., Beauchemin K.A., Okine E. and McAllister T.A. (2019) A pine enhanced biochar does not decrease enteric CH_4 emissions but alters the rumen microbiota. *Frontiers of Veterinary Science* 6, 308.

Assessing the frequency of pasture allocation and distance walked on Irish dairy farms

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Abstract

The objective of this study was to examine the current status of grazing infrastructure on Irish dairy farms, pasture allocation frequency (PAF), and distance travelled on farm roadways by dairy cows in a range of herd sizes. It was hypothesized that as Irish dairy herd size has grown since quota abolition in 2015, paddock size may have not adapted in line with herd expansion. A cohort of 138 dairy farms (3,760 grazing paddocks) were selected and PAF were calculated using Pasturebase Ireland database. Paddock sizes were normalized relative to herd size at each farm. Herds of 150 cows or greater spent more time in smaller paddocks, 43.2% of paddocks surveyed were only suitable for 12-h allocations compared to 16.6% of paddocks in herds less than 150 cows. This can have a negative impact on animal performance, particularly in primiparous animals. Herd size also impacted distance walked per year ($P<0.05$). On farms with herds ≥150 cows a greater proportion of time was spent walking. Factors such as farm layout, orientation relative to the milking parlour position on the farm, and the extent of natural boundaries also impacted on distance travelled.

Keywords: pasture allocation frequency, paddock size, farm roadways, walking

Introduction

The Irish dairy industry has experienced significant change since the abolition of milk quotas in 2015. With the average herd size increasing from 64 cows in 2010 to 82 cows in 2020 (National farm survey, Teagasc). Pasture allowance can directly affect milk production and pasture utilization (Curran *et al.*, 2010). If pasture allocation has not adapted from 12 to 24 or 36 hour allocations through enlarged paddock sizes to accommodate larger herds, pasture allocation frequency (PAF) will be increased, which can cause a decrease in animal performance, particularly for primiparous animals (Pollock *et al.*, 2020). The objective of this study was to examine whether paddock size have adapted to increased herd sizes, while quantifying distance animals are walking to access pasture on commercial dairy farms.

Materials and methods

The current study generated data from commercial pasture-based dairy farms across Ireland, using Pasturebase Ireland (PBI) database, to determine PAF in relation to herd size and the annual distance travelled on farm roadways during the 2020 grazing season. Parameters were specified such that data could be collected from a cohort of 138 farms encompassing a wide range of herd sizes adapted from Kelly *et al.* (2020) and soil types. Farms involved were split into two categories: H1 (<150 cows) or H2 (≥150 cows) based on the median herd size of 150 cows. Farm maps were analysed for paddock location and access routes to the milking parlour. Additional data collected from PBI included paddock sizes, number of paddocks, number of grazings per paddock and the total number of days at grass. Across all farms, the pre-grazing herbage mass (PGHM) was assumed to equal 1,400 kg dry matter (DM) ha^{-1} and dry matter intake (DMI) was estimated to be 17 kg DM cow^{-1} (Wims *et al.*, 2014). Paddocks were normalized relative to the herd size of the farm in question for their suitability for pasture allocations of 12, 24, 36, or 48 h assuming a PGHM of 1,400 kg DM ha^{-1}. The distance in meters was measured from each paddock to the milking parlour. This was combined with PBI grazing data to predict distance walked yearly (DWY) per farm as well as distance walked daily (DWD) across the grazing season for 2020 based on individual paddock grazing data. Data were statistically analysed using Rstudio through Rx64 4.0.2, using an independent t-test to assess paddock size normalized to herd size and distance walked relative to herd size.

Results and discussion

There was a significant difference in the proportion of paddocks only suitable for 12 h allocations ($P<0.05$) increasing with herd size, rising from 16.6% for herds in H1 to 43.2% for herds in H2. The number of paddocks on the farm rose substantially as herd size increased, 24±0.04 to 30.5±0.03 *($P<0.05$)* for H1 and H2 herds respectively. There was a significant difference ($P<0.05$) in the grazings achieved per farm between both groups, with H1 achieving 7.2±0.02 grazings per year compared to 7.8±0.03 for H2. As herd size has increased post quota (National farm survey, Teagasc) paddock sizes have not adapted, particularly in larger herds (>150 cows), resulting in an increase in 12 h paddocks, consequential increasing PAF per animal (Pollock *et al.*, 2020). Werner *et al.* (2019) reported a reduction in the post-grazing sward height when daily herbage allocation was reduced, which can significantly reduce growth rates of the plant (Ganche *et al.*, 2015). Where PAF is increased above once per day, as is the case in this study through 12 h allocations, this potentially presents a competitive advantage to multiparous over primiparous animals due to a higher social rank in the herd (Phillips and Rind, 2002) where resources are restricted. Pollock *et al.* (2020) saw a reduction in milk production in primiparous animals while grazed in 12 h allocations as opposed to 24 or 36 h allocations, which may be due to reduced DMI through increased PAF.

There was a positive linear correlation between herd size and DWY (R^2=0.4612) ($P<0.001$, Figure 1). There is also a large dispersion across the datum points, due to a range of factors including farm layout, natural boundaries and the milking parlour position on the farm in relation to the grazing platform.

The H2 herds walked a significantly further ($P<0.05$) DWY than those in H1, 636.5±22.87 km and 440.6±19.17 km, respectively. The DWD was also significant ($P<0.05$) with H1 walking 1,682.3±96.42 m compared to H2 2,327.1±79.29 m. This is similar to that reported by Neave *et al.* (2021) where DWD increased with increasing herd size; however, they did not report a reduction in milk yield but it did result in reduced ruminating time. The DWD is below that assessed by D'Hour *et al.* (1994) where milk yield was not affected until the distance walked exceeded 6.4 km per day; however, in their study only 9 animals participated whereas in the current study the smallest herd present contained 47 animals and the largest had 750 animals. Long walking distances, track width and poor track maintenance can be a risk factor for traumatic lameness in pastured cows (Chesterton, 2015). Furthermore, Buijs *et al.* (2019) noticed a difference in walking speed under different track surfaces.

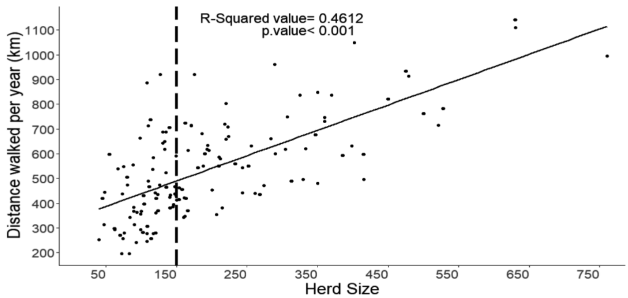

Figure 1. The correlation between the distances walked on farm roadways throughout the grazing season of 2020 and the herd size of each farm involved. There is a positive linear correlation.

Conclusions

Paddock sizes have not adapted to meet the demand of increased herd sizes particularly in H2, which can reduce milk production in primiparous animals due to competition for pasture. Reducing PAF will reduce inter-cow competition and improve milk production in these animals. As herd size has increased, so too has the DWD, and further investigation is required to discover if farm roadways have adapted to cater for larger herds, or if there has been a similar trend to that observed in PAF. A greater DWD may increase the presence of lameness in the herd if farm roadways have not been adapted, in relation to roadway width or adequate maintenance. Labour demand may also increase with increased DWD to access pasture.

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References

Buijs S., Scoley G. and McConnell D. (2019) Artificial grass as an alternative laneway surface for dairy cows walking to pasture. *Animals* 9, 891.

Chesterton R.N. (2015) The lame game- Chile vs New Zealand – can we both be winners? 18th International Symposium and 10th Conference on Lameness in Ruminants, Valdivia, Chile, pp. 26-29.

Curran J., Delaby L., Kennedy E., Murphy J.P., Boland T.M. and O'Donovan M. (2010) Sward characteristics, grass dry matter intake and milk production performance are affected by pre-grazing herbage mass and pasture allowance. *Livestock Science* 127, 144-154.

D'Hour P., Hauwuy A., Coulon J. and Garel J. (1994) Walking and dairy cattle performance. *Annales de zootechnie* 43, 369-378.

Ganche E., O'Donovan M., Delaby L., Boland T.M. and Kennedy E. (2015) Does post-grazing sward height influence sward characteristics, seasonal herbage dry-matter production and herbage quality? *Grass and Forage Science* 70, 130-143.

Kelly P., Shalloo L., Wallace M. and Dillon P. (2020) The Irish dairy industry – Recent history and strategy, current state and future challenges. *International Journal of Dairy Technology* 73, 309-323.

Neave H.W., Edwards J.P., Thoday H., Saunders K., Zobel G. and Webster J.R. (2021) Do walking distance and time away from the paddock influence daily behaviour patterns and milk yield of grazing dairy cows? *Animals* 11, 2903.

Phillips C.J.C. and Rind M.I. (2002) The effects of social dominance on the production and behavior of grazing dairy cows offered forage supplements. *Journal of Dairy Science* 85, 51-59.

Pollock J.G., Gordon A.W., Huson K.M. and McConnell D.A. (2020) The effect of frequency of fresh pasture allocation on pasture utilisation and the performance of high yielding dairy cows. *Animals* 10, 2176.

Werner J., Umstatter C., Kennedy E., Grant J., Leso L., Geoghegan A., Shalloo L., Schick M. and O'Brien B. (2019) Identification of possible cow grazing behaviour indicators for restricted grass availability in a pasture-based spring calving dairy system. *Livestock Science* 220, 74-82.

Wims C.M., Delaby L., Boland T.M. and O'Donovan M. (2014) Effect of pre-grazing herbage mass on dairy cow performance, grass dry matter production and output from perennial ryegrass (*Lolium perenne* L.) pastures. *Animal* 8, 141-151.

The different services provided by grasslands in livestock-crop reconnection

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Abstract

Grasslands can play an important role in livestock-crop reconnection. In France, INRAE and ACTA together with partners involved in livestock, crop, fruit and vegetable production sectors have joined forces to analyse the research and R&D initiatives already underway in these recoupling avenues both upstream and downstream of the sectors. A database of 10 European, 35 national projects, and 43 local initiatives has been built. In this database we inventoried 12 R&D projects and 18 field initiatives focused on the utilization of grasslands in livestock-crop reconnection. The main services provided by grasslands that are quoted are feed autonomy, feed cost reduction and system resilience for the farming system, and the reduction of pesticide use and nitrogen fertilization, soil fertility improvement and carbon storage for the environment. A significant number of projects and initiatives involve monogastric animals. In particular, we have identified: (1) the use of grasslands in agroforestry and free-range poultry to promote animal welfare and the emergence of a quality label specification; and (2) the use of alfalfa leaf meal to provide local protein feed to pigs.

Keywords: livestock-crop reconnection, crop diversification, protein autonomy, animal welfare, agroecological transition

Introduction

Grasslands are a resource enabling breeders to achieve forage and protein autonomy at a lower cost. The issues surrounding agriculture are now posed in new terms: reduction in inputs, greenhouse gas emissions and phytosanitary products, improvement of the feed autonomy of livestock farms, relocation of production to supply cities, etc. The development of agricultural systems that close biogeochemical cycles is a promising way to meet these challenges (Dourmad *et al.*, 2019; Peyraud *et al.*, 2019). Initiatives aimed at reconnecting animal and plant production at different scales are emerging but currently remain dispersed, which does not allow their evaluation or dissemination. The objective of this study is to better identify the possibilities of reconnection, the levers and obstacles to their success and to have, in the long term, methods to evaluate the performance of grassroots projects mobilizing crop/livestock complementarity. In this article, we will focus on the roles played by grasslands in these livestock-crop reconnection processes.

Materials and methods

At the French scale, an inventory of projects (from 2013 to present day) studying the complementarity between crops and livestock was made. This inventory allowed us to identify 88 projects of interest (43 local initiatives in France, 35 Research and Development (R&D) national projects supported by French funding agencies, and 10 projects funded by the European Union. The database inventorying these R&D projects and fields initiatives includes the following items:

- Crops: arboriculture, viticulture, market gardening, field crops/covers, etc.
- Animal species: poultry (geese, chickens, ducks), sheep, cattle, horses, pigs, rabbits, etc.
- Forms of coupling: mixed farming, exchanges between breeders and farmers (animal feed, effluents), creation of new sectors, methanization, etc.

The database summarizes factual data (dates, acronyms, regions, themes, partners, etc.), the aim of the project, the research question, the scale, the barriers encountered in the execution of the project and the instruments implemented to overcome these barriers. To collect this information, we have retrieved the project documents and deliverables produced. From this database we extracted R&D projects and field initiatives that focus on the use of grasslands. By reading the deliverables and other documents produced by these initiatives, we identified the different services provided by the grasslands in the crop-livestock reconnection processes. We crossed our analysis with the study of Michaud *et al.* (2020) who made a list of the different services provided by grasslands to the environment, animals, farmers and citizens and consumers. To illustrate the outcomes of the database we describe two examples of livestock-crop reconnection involving grasslands and monogastric animals.

Results and discussion

We identified 12 R&D projects and 18 field initiatives that mention the interest of grasslands in crop-livestock reconnection processes. Out of the 12 R&D projects, eight are specific to ruminant farming (six focus on sheep) and four concern both ruminant and monogastric farming. Out of the 18 field initiatives, 11 are specific to ruminant farming (six focus on cattle and three on sheep), four are specific to monogastric farming (three focus on pigs and one on poultry) and three concern both ruminant and monogastric farming. It is noteworthy that four out of 12 R&D projects and seven out of 18 field initiatives involve monogastric animals that are not the common users of grasslands.

Sixteen services provided by grasslands were identified and distributed into four main categories: services provided to the environment, to the farming system, to livestock, and to consumers and citizens. For each of these 16 services, we counted the number of R&D projects and field initiatives that quoted them (Table 1). Both for R&D projects and field initiatives, the most quoted services are those provided to the farming system, in particular feed autonomy, feed cost reduction and system resilience. For the R&D projects, services provided to the environment are also highly quoted, especially regarding the reduction of pesticide use and nitrogen fertilization, soil fertility improvement and carbon storage. For the field initiatives, the quoted services are relatively balanced between the four major categories.

Table 1. Percentage of the R&D projects and field initiatives that quote the different services provided by grasslands.

Categories of services	Services	% of R&D projects quoting this service	% of field initiatives quoting this service
Services to the environment	Reduction of pesticides	75	44
	Reduction of N fertilization	67	50
	Reduction of erosion / soil cover	33	28
	Carbon storage / soil fertility improvement	75	22
	Reduction of nutrient loss/water quality	58	33
	Maintain and increase of biodiversity	33	17
	Recycling of effluents	33	28
Services to the farming system	Improvement of feed autonomy	92	83
	Reduction of feed costs	92	83
	System resilience through diversity	92	78
	Social link between farmers	42	17
Services to the livestock	Better balance of the ration for the animals	17	61
	Animal welfare	25	17
Services provided to consumers and citizens	Response to societal expectations regarding livestock practices	25	39
	Quality of the landscape	8	22
	Quality of animal products	8	28

Among the field initiatives, we have identified two emerging topics that involve monogastric farming and grasslands: the use of grasslands in agroforestry, free-range poultry to promote animal welfare, and the production of alfalfa leaf meal to provide local protein feed to pigs.

For the first topic, the following initiatives were analysed: (1) an association of 75 producers of fattened palmipeds in the Périgord region who are experimenting with agroforestry and free-range poultry; (2) an agricultural extension group in the Haute-Vienne Department (87) that aims to promote the emergence of agroforestry systems for the poultry, cattle and sheep sectors. The benefits that are quoted by these two field initiatives concern the animals (animal welfare), the environment (reduction of erosion, recycling of effluents and increase of biodiversity) and the citizens and consumers (Response to societal expectations, quality of animal products and of the landscape). These systems, developed by a few pioneers, have proven beneficial, and it would be interesting to obtain more technical references in order to be able to extend these systems to a larger scale.

For the second topic we analysed an experiment conducted in Brittany (Calvar *et al.*, 2020) which tested the performance of sixty pigs fed with a feed containing 5% alfalfa leaf meal. The experiment showed that pigs consume alfalfa, but their growth is significantly lower and the feed conversion ratio degraded, compared to a conventional feed where the protein supply is based solely on oilcake. The poorer results can be explained by the lesser knowledge of the nutritional values of this product. Few complete data are currently available and this is a strong limitation of the study. In addition, the formulation should probably have been revised to correct the imbalances (energy and amino acids) of this feed.

Conclusions

Grasslands produced a wide diversity of services in the crop-livestock reconnection process for ruminant as well as for monogastric farming. Services provided to the farming system and to the environment are the most investigated ones. In many cases more scientific and technical references are needed regarding the role of grasslands in livestock-crop reconnection.

References

Calvar C., Duputel M. and Renaudeau D. (2020) Valorisation de la luzerne par les porcs en engraissement. *Essai Chambres d'agriculture de Bretagne.*

Dourmad J.-Y., Guilbaud T., Tichit M. and Bonaudo T. (2019) Les Productions Animales Dans La Bioéconomie. *INRAE Productions Animales* 32(2), 205-220.

Michaud A., Plantureux S., Baumont R. and Delaby L. (2020) Grasslands, a source of richness and support for innovation for more sustainable and acceptable ruminant livestock farming. *INRAE Productions Animales* 33 (3), 153-172.

Peyraud J.-L., Aubin J., Barbier M., Baumont R., Berri C., Bidanel J.-P., Citti C., Cotinot C., Ducrot C., Dupraz P., Faverdin P., Friggens N., Houot S., Nozières-Petit M.-O., Rogel-Gaillard C. and Santé-Lhoutellier V. (2019) Quelle science pour les élevages de demain ? Une réflexion prospective conduite à l'INRA. *INRAE Productions Animales* 32(2), 323-338.

Agroforestry reduces nitrogen surplus of organic poultry and pig production

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Abstract

Agroforestry promotes animal welfare, but very little data are available for nitrogen (N) flows in these production systems. A field experiment was conducted in 2018 in Denmark on sandy soils with grass-clover, willow and outdoor organic poultry with hens fed either a standard organic feed or soybean cake partly replaced by protein from local grass-clover ('green'), at either low (6 m^2 hen^{-1}) or high stocking density (4 m^2 hen^{-1}). Nitrogen flows were quantified in 2018-2019 in the paddocks by direct measurements, including soil solution sampling, and empirical estimates. The surface balances – difference between feed and meat – ranged 628-651 and 835-864 kg N ha^{-1} for the low and the high density, respectively, the lower end in each range being for green protein feed treatments and not significantly different from the higher end (control feed). The soil balances further deducted environmental flows and were 12-15% lower than the surface balances, indicating environmental pressure in the paddocks, although with 40% lower leaching in the willow than the grass-clover zone. Another experiment in 2015 also on sandy soil with grass-clover in Denmark involved organic outdoor pigs with or without access to poplar trees, and a control without trees, with respective surface balance of 436, 397 and 468 kg N ha^{-1} $year^{-1}$, and soil balance 60-70% lower than the surface values due to large leaching that was nevertheless significantly lower in the tree, compared to the grass-clover zone. Both studies collectively show sound prospects to reduce N surplus from organic outdoor animal production on sandy soils by agroforestry, with great offset by the trees of N leaching from manure deposition and grass-clover residues mineralization.

Keywords: clover, grass, hen, leaching, mass balance, pig, poplar, willow

Introduction

Agroforestry holds potential to increase biodiversity and reduce surface- and groundwater pollution, as trees use soil nitrogen (N), e.g. Manevski *et al.* (2019). Integrated in cropping systems, trees and grasses are able to reduce N leaching and tighten the soil N balance, i.e. the difference in input and output flows to the soil, due to efficient N uptake (e.g. Manevski *et al.*, 2018; Pugesgaard *et al.*, 2015). For agroforestry in Europe, very few studies have directly addressed their soil N balances and environmental implications. The objective of the study was to quantify soil N balances of outdoor poultry and pig systems with agroforestry.

Materials and methods

Field trials in certified organic settings were conducted on sandy soils in Denmark. The poultry trial (April to November 2018) involved willow clones 'Inger' (*Salix triandra* × *Salix viminalis*) and 'Tordis' (*Salix schwerinii* × *S. viminalis*; 50% each, 1000 stems ha^{-1}) established in 2014 (first harvested in February 2017) and occupied about 20% of paddocks with grass-clover (*Lolium perenne – Trifolium repens*; sown 2014). Each paddock (20×34.5 m length×width) contained a double-hut in its upper-central area (10 m width) with an effective frontal 'grass zone' and three rows of willow on each side with 4 m inter-row space. The experiment was a 2×2 factorial design with three replicates and dietary composition as first factor, with hens fed either standard 'control' diet or with the soybean cake partly replaced by 'green protein' produced from grass-clover. The diets were balanced in protein and energy according the hen's requirement, with crude protein concentration of 19-20%. The second factor was stocking density being either high (4 m^2 hen^{-1}) or low (6 m^2 hen^{-1}).

The pig experiment (April 2015 to April 2016) involved poplar clones OP42 (*Populus maximowiczii* (Henry) × *Ppoulus trichocarpa* (Torr. et Gray), 1000 stems ha^{-1}) established in 2011 (not harvested) occupying 20% of paddocks (10×33 m) either non-fenced (access to trees) or fenced (no access to trees), with control paddocks (12×27 m) without trees, and each with 7 replicates and holding a lactating sow with its piglets (Manevski *et al.*, 2019).

For each paddock in the two experiments, soil N balance (kg N ha^{-1}) was estimated for the hydrological year (April to April) according to OECD (2001):

$$\text{Soil N balance} = [N_{feed} - N_{animal}] + N_{inflow}\ (\text{straw, fix, atm}) - N_{outflow}\ (\text{leach, ammonia, nitrous oxide})$$

where *feed* is measured mean feed N (protein/6.25), *animal* is N of 28.8, 18.1 or 25 g kg^{-1} for, respectively, hen, egg or piglets and calculated according to the measured number and weight of animals. The difference between *feed* and *animal* is the surface N balance. Further, *straw* is 5 kg N ha^{-1} provided to huts, *fix* of 30 kg ha^{-1} is biological N fixation by grass-clover (decreased from typically higher values due to disturbance such as grazing, trampling, roaming), *atm* is 15 kg N ha^{-1} atmospheric deposition, *leach* is N (nitrate) leaching from the root zone (1 m) calculated by a percolation-weighted concentration method, where percolation was simulated by the process-based Daisy model (Manevski *et al.*, 2019). Emissions of *ammonia* and *nitrous oxide* were estimated using emission factors of, respectively, 0.13 (of feed) and 0.001 (of manure deposition) for grazing, and 0.5 and 0.01 for crop residues and N loss by surface runoff was disregarded due to flat terrain (<2% slope).

Results and discussion

The surface N balances for the poultry systems were large, i.e. 627±7 to 651±5 kg N ha^{-1} at low- and 834±18 to 894±8 kg N ha^{-1} $year^{-1}$ at high stocking density. The lower end in each range was systematically for the green protein treatment and not significantly different from the 'control' higher end. This result shows that low stocking density tightens the surface N balance and it is feasible to replace the soy portion in the organic feed with local protein. The N efficiency of the organic egg production was 29-30% (Table 1), close to the 32% reported for conventional egg production in Denmark (Groenestein *et al.*, 2019). The soil N balances were, on average, 12-15% lower than the surface balances, i.e. 524±13 to 572±9 and 748±22 to 754±15 kg N ha^{-1} for low and high stocking density, respectively. Thus, reducing the stocking density of the hens of 33% reduced the soil N surplus of about 26%. Manure deposition was not recorded, yet hens moved freely in the paddocks and N leaching in the willow zone was, on average, 40% lower compared to the grass zone (Figure 1A).

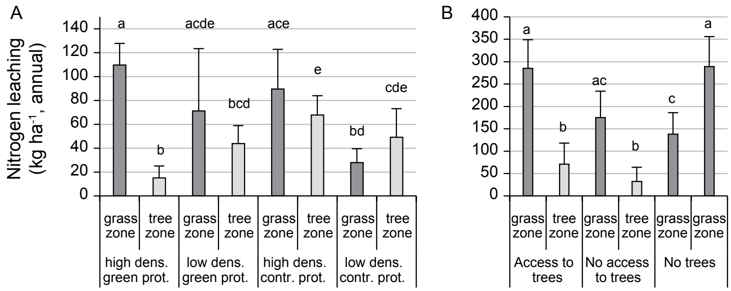

Figure 1. Nitrogen leaching in paddocks with hens (A; tree is willow) and pigs (B; tree is poplar) under outdoor organic settings in Denmark. Means (standard deviation, error bars) with different letters are significantly different ($P \leq 0.05$). Note plots different y-axis.

Table 1. Annual nitrogen balances (kg N ha^{-1}) for organic poultry (2018-2019; tree is willow) and pig (2015-2016; tree is poplar) experiments with agroforestry in Denmark. Nitrogen (N) efficiency is the ratio of animal/product to imported feed.

		Experiment with growing hens				Experiment with lactating sows		
		High dens. green prot.	High dens. contr. prot.	Low dens. green prot.	Low dens. contr. prot.	Access to trees	No access to trees	No trees (control)
Inflows	Imported feed	1,113	1,146	827	847	576	564	600
	Straw	5	5	5	5	5	5	5
	Atm. deposition	15	15	15	15	15	15	15
	Clover fixation	30	30	30	30	30	30	30
	Total input	1,163	1,196	877	897	627	615	651
	Animal/product	329	332	250	246	191	219	183
	Surface N balance	835	864	628	651	436	397	468
	N efficiency (%)	30	29	30	29	30	36	28
Outflows	Ammonia							
	Grazing	0	0	0	0	75	73	78
	Crop residues	15	15	15	15	15	15	15
	Nitrous oxide							
	Grazing	0	0	0	0	8	7	9
	Crop residues	2	2	2	2	2	2	2
	Nitrogen oxides	10	10	10	10	12	12	13
	Dinitrogen							
	Manure	5	5	5	5	24	22	26
	Crop residues	5	5	5	5	6	6	6
	Nitrogen leaching	50	73	66	42	175	101	206
	Soil N balance	748	754	524	572	118	157	113

The N efficiency for the pig system was similar to the poultry (28-36%), also with large surface N balances of 397-468 kg N ha^{-1} (Table 1). However, leaching was considerable in these systems (100-200 kg N ha^{-1}), which left 113-157 kg N ha^{-1} unaccounted for in the soil (Manevski *et al.*, 2018). The poplar trees offset N leaching manifold (Figure 1B).

Conclusions

These are among the first N-balance data for agroforestry systems with poultry and pig production in Europe. The results show the prominent role of willow and poplar trees to offset N leaching from paddocks on sandy soils, thus reducing the surplus of the soil N balance.

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References

Groenestein C.M., Hutchings N.J, ... and Webb J. (2020) Comparison of ammonia emissions related to nitrogen use efficiency of livestock production in Europe. *Journal of Cleaner Production* 211, 1162-1170.

Manevski K., Jakobsen M., ... and Jørgensen U. (2019) Effect of poplar trees on nitrogen and water balance in outdoor pig production – A case study in Denmark. *Science of the Total Environment* 646, 1448-1458.

Manevski K., Lærke P.E., Olesen J.E. and Jørgensen U. (2018) Nitrogen balances of innovative cropping systems for feedstock production to future biorefineries. *Science of the Total Environment* 633, 372-390.

OECD (2001) *Environmental indicators for agriculture*. OECD, Paris, France.

Pugesgaard S., Schelde K., Larsen S.U., Lærke P.E. and Jørgensen U. (2015) Comparing annual and perennial crops – influence on nitrate leaching and energy balance. *Global Change Biology Bioenergy* 7, 1136-1149.

Forage quality predicted by hyperspectral reflection measurements across climate zones

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Abstract

Forage quality is an important ecosystem service of grasslands, with its quantification being still laborious and costly. Remote sensing technologies, such as hyperspectral measurements, enable its fast and non-destructive estimation. However, such methods are still limited in transferability to other locations or climatic conditions. This study aims to predict forage quality from hyperspectral canopy reflections of grasslands across three climate zones. We took hyperspectral measurements with a field spectrometer from grassland canopies in temperate, subtropical, and tropical grasslands, and analysed corresponding biomass samples for metabolizable energy. Machine learning methods were used to establish prediction models for single climate regions and across climate regions. First results indicate good performance in both local and broad trans-climatic predictions. Best model accuracies resulted in using the 1st derivation of the full hyperspectral signature (nRMSE=10.10, R^2=70.30). For further analysis, we expect even higher accuracies using deep learning models. We concluded that the models based on hyperspectral measurements offer great potential to assess or even map the forage quality of grasslands across climate zones.

Keywords: hyperspectral measurements; forage quality; remote sensing; trans-climatic modelling

Introduction

The world's grassland ecosystems provide multiple ecosystem services, with forage provision – both forage quality and quantity – being among others the most important ones. However, it is still difficult to quantify forage provision, especially its component forage quality. Its analysis is expensive and time-consuming to measure, as it usually requires the analysis of biomass samples in the laboratory. Here, remote sensing technologies, such as hyperspectral measurements, gain increasing importance, since they allow for fast and non-destructive estimations (Ferner *et al.*, 2015). Hyperspectral modelling approaches have been used to estimate, e.g. metabolizable energy content for West African savanna grasslands (Ferner *et al.*, 2018, 2021), nutritive value for South African savanna grasslands (Singh *et al.*, 2017), and temperate ryegrass canopies (Smith *et al.*, 2020). Yet, forage quality predictions are still limited in transferability to other locations or climatic conditions. We aim to fill this gap by predicting metabolizable energy content from remotely sensed hyperspectral canopy reflection of grassland communities covering grasslands in three different climate zones located on the African and the European continent. This approach allows the set-up of regional but also trans-climatic calibration models.

Material and methods

Our study areas covered grasslands in temperate, subtropical, and tropical climates. Sites were located in: (1) subtropical to tropical grasslands in West Africa's Sudanian savannas (Ferner *et al.*, 2015; Guuroh *et al.*, 2018); (2) subtropical grasslands in Namibia's semi-arid thorn bush savannas; and (3) temperate Central European meadows and pastures, mostly within the three Biodiversity Exploratories in north-eastern, central, and south-western Germany (Fischer *et al.*, 2010). We took 344 hyperspectral measurements with the aid of full-range field spectrometers (123 from Germany, 101 from West Africa, and 120 from Namibia). Samples of aboveground biomass were collected from all measured areas, and dried samples were analysed for metabolizable energy content (ME) as a proxy for forage quality, following the procedure of Menke and Steingass (1988). The spectral signatures of the hyperspectral measurements were smoothened and corrected regarding atmospheric dynamic artifacts. For each spectrum, we determined the 1st and 2nd derivation, several vegetation indices (VIs), as well as absorption features from the whole spectrum. Both derivations and original reflection spectra were categorized in 'Spectra', 'Spectra 1st derivation', 'Spectra 2nd derivation', and 'Features/Indices' as potential predictors for forage quality. Machine learning algorithms, including partial least squares regression (PLSR) and random forest regression, were used to establish prediction models for the single climate zones and across the three zones.

Results and discussion

First results with PLSR modelling using available ME data from the temperate sites (n=75) and (sub-) tropical sites (n=101) showed promising model qualities after leave-one-out cross-validation. Best accuracies for ME prediction were achieved by models using the 1st derivation, followed by models based on absorption features and vegetation indices (Figure 1).

Here, normalized root means square errors (nRMSE) of around ten and a coefficient of determination (R^2) of >70% could be achieved, which is superior to previous prediction accuracies of usually well below 70% (e.g. Ferner *et al.*, 2015; Smith *et al.*, 2020). We only found a slight underestimation of high forage quality values, which may be a hint for overfitting. This artifact could probably be reduced after adding more data for the temperate region with higher ME values. Interestingly, the accuracies of the trans-climatic models were within the range of, or even higher than the local models, both for the 1st derivation and based on absorption features and indices of the spectra. The similar accuracy despite a higher variation of the data may be explained by the higher number of samples (n=176) of the trans-climatic model compared to the local ones (n=75, n=101). This effect of similar or higher accuracy while adding more climatic conditions is a promising result. For our upcoming analyses, adding data from the subtropical Namibian savanna and using deep learning methods, we expect prediction models of similar quality in both the local and the trans-climatic models.

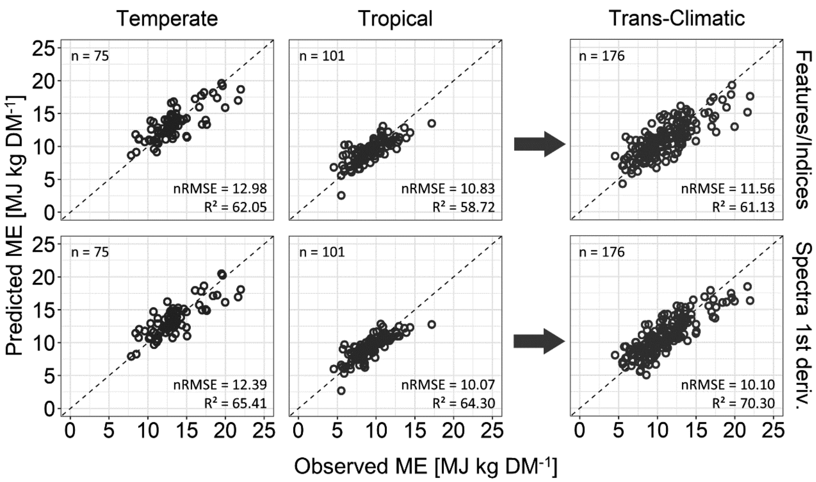

Figure 1. PLSR model validation of regional and trans-climatic prediction models of forage quality (measured as metabolizable energy content, ME) based on a combination of absorption features and vegetation indices, and 1st derivation of the reflectance spectra. Model accuracy was quantified with the normalized root mean square error (nRMSE) and the coefficient of determination (R^2).

Conclusions

Hyperspectral forage quality models showed good performance in both local and broad trans-climatic applications. Trans-climatic models will be improved by adding more data from Namibia and applying deep-learning methods. Hyperspectral models thus offer great potential for farmers and ecologists, to assess or even map the forage quality of grassland areas worldwide.

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References

Ferner J., Linstädter A., Rogass C., Südekum K.-H. and Schmidtlein S. (2021) Towards forage resource monitoring in subtropical Savanna grasslands: going multispectral or hyperspectral? *European Journal of Remote Sensing* 54(1), 364-384.

Ferner J., Linstädter A., Südekum K.-H. and Schmidtlein S. (2015) Spectral indicators of forage quality in West Africa's tropical savannas. *International Journal of Applied Earth Observation and Geoinformation* 41(September), 99-106.

Ferner J., Schmidtlein S., Guuroh R.T., Lopatin J. and Linstädter A. (2018) Disentangling effects of climate and land-use change on West African drylands' forage supply. *Global Environmental Change* 53, 24-38.

Fischer M., Bossdorf O., Gockel S., Hänsel F., Hemp A., Hessenmöller D. *et al.* (2010) Implementing large-scale and long-term functional biodiversity research: The Biodiversity Exploratories. *Basic and Applied Ecology* 11(6), 473-485.

Guuroh R.T., Ruppert J.C., Ferner J., Čanak K., Schmidtlein S. and Linstädter A. (2018) Drivers of forage provision and erosion control in West African savannas – A macroecological perspective. *Agriculture, Ecosystems & Environment* 251, 257-267.

Menke K.H. and Steingass H. (1988) Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. *Animal Research and Development* 28, 7-55.

Singh L., Mutanga O., Mafongoya P. and Peerbhay K. (2017) Remote sensing of key grassland nutrients using hyperspectral techniques in KwaZulu-Natal, South Africa. *Journal of Applied Remote Sensing* 11(3), 36005.

Smith C., Karunaratne S., Badenhorst P., Cogan N., Spangenberg G. and Smith K. (2020) Machine learning algorithms to predict forage nutritive value of in situ perennial ryegrass plants using hyperspectral canopy reflectance data. *Remote Sensing* 12(6), 928.

Bacterial and botanical diversity of the pasture influence the raw milk cheese sensory properties

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Abstract

The interactions among plant and bacterial ecosystems of pastures, and livestock and their products, are intrinsic to quality schemes for protected designation of origin, but still lack in-depth comprehension. To study the transfer of bacteria from pastures to cheese, a highly biodiverse permanent grassland and an adjacent old temporary grassland were grazed by 2×3 balanced groups of 4 dairy cows each. A total of 18 Cantal-type cheeses were produced from raw milk (3 replicates per group) and ripened during 9 weeks. Bacterial community profiles (16S rDNA metabarcoding) differed significantly in the simulated herbage bites selected by dairy cows and to a lesser extent in raw milk and cheese depending on the pasture type. Sixty-seven bacterial sequence variants were shared between simulated bites, milk, and cheese core and rind. The most abundant sequence in cheese core and rind (assigned to *Lactococcus lactis*) was found also in simulated bites and in milk. Other sequences with above 8% abundance in cheese rind (assigned to *Brevibacterium aurantiacum* and *Brachybacterium* sp.) were also shared with simulated bites. The less firm texture of the cheeses from the highly biodiverse pasture could be attributable to their higher fat in dry matter content, whereas their stronger dry fruits odour and flavour could be partly explained by their specific bacterial community profile.

Keywords: biodiversity, 16S rDNA metabarcoding, raw milk cheese, flavour

Introduction

The botanical diversity of pastures has long been associated with the sensory characteristics of raw milk cheeses (Martin *et al.*, 2005). Nevertheless, evidence of the direct influence of the diversity of plant secondary compounds on the development of the cheese odour and flavour is often hypothesized, but was experimentally disproved (Tornambé *et al.*, 2008). The diversity of bacterial species in the phyllosphere in association with the botanical diversity of grasslands may also be a driver of the development of the raw milk cheese sensory quality (Frétin *et al.*, 2018). We hypothesize that the botanical diversity of the pastures shapes the microbiota along a continuum from the aboveground surface of plants to raw milk and eventually to raw milk cheese. A controlled replicated experiment with standardized cheesemaking procedures was carried out in order to characterize the bacterial communities from the herbage selected by dairy cows (i.e. simulated bites) to the ripened cheeses.

Materials and methods

The experiment was conducted at the INRAE Herbipôle experimental farm (https://doi.org/10.15454/1.5572318050509348E12). Twenty-four Holstein and Montbéliarde cows were randomly allocated to 6 groups of 4 cows, each balanced by breed, lactation number, milk yield, as well as milk fat and protein yields. Three groups were assigned to a highly biodiverse permanent grassland (HD; Shannon diversity index *H'* 312) and the 3 others to a low diversified adjacent old-temporary grassland plot (LD; *H'* 219). The cows were exclusively pasture-fed and had free access to NaCl and water. Three weeks of samplings followed 2 weeks of adaptation to the respective pasture. On one day per sampling week, samples of simulated bites of each group were collected by following the cows on pasture between morning and evening milking and collecting herbage samples with scissors by mimicking their selection for plant species and plant parts. Microbial contaminations were minimized by wearing gloves, and disinfecting scissors between different groups. The morning bulk milk of each group was collected once per sampling week, sampled for analysis and simultaneously processed to Cantal-type cheeses in an experimental cheesemaking facility (INRAE, UMR Fromage, Aurillac). After 9 weeks of ripening, the 18 cheeses were sampled for chemical and sensory analyses according to Manzocchi *et al.* (2021). Bacterial communities in samples of simulated bites, milk, cheese rind, and cheese core were characterized by 16S rDNA metabarcoding according to Frétin *et al.* (2018). The amplicon sequence variants (ASVs) in each sample were identified and the *H'* indexes were calculated with the rANOMALY package in R (Theil and Rifa, 2021). The data were analysed with a linear mixed model (mixed procedure in SAS, version 9.4) including pasture type as fixed effect and group (all data), panellist, and session (only for sensory data) as random effects.

Results and discussion

Cheeses derived from the botanically highly diverse pasture were tendentially more yellow and less firm, and had a higher fat in dry-matter content, as well as more intense 'dry fruits' odour and flavour (Table 1). No differences were observed in the spreadability index (*cis*-9 C18:1-to-C16:0 ratio) of the cheeses between the pasture types. A higher total number of bacterial ASVs and a higher bacterial *H'* index were found in the bites selected on the botanically highly diverse pasture than in those selected on the less diverse pasture (Table 2). The total number of ASVs and the *H'* index of milk, cheese core, and cheese rind did not differ significantly between pasture types. Simulated bites, milk, as well as cheese core and rind derived from the two pasture types had 67 common ASVs, 15 of which were found in all analysed compartments regardless of the botanical diversity of the pasture (Figure 1). Among the latter, an ASV assigned to *Lactococcus lactis* with a very high relative abundance in the cheese core (93.9%) and cheese rind (49.4%) was found with lower abundances also in milk (5.9%) and in simulated bites (0.2%). Two ASVs assigned to *Brevibacterium aurantiacum* and *Brachybacterium sp*., both notably involved in cheese ripening, were very abundant in the cheese rind (17.3%) and were identified with lower abundances also in all other compartments, which might indicate a possible transfer of these bacteria from the pastures to raw milk and cheese.

Conclusions

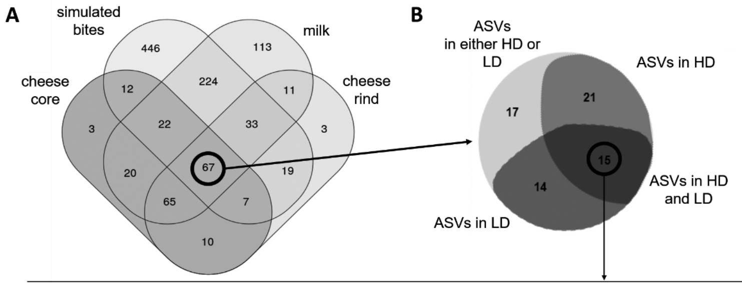
In conclusion, the less firm texture of the cheeses from the highly biodiverse pasture may be attributable to their higher fat in dry matter content, their more intense 'dry fruits' odour and flavour could be partly

Table 1. Effect of the botanical diversity of the pasture on the cheese composition, proteolysis, colour of the curd, fatty acid (FA) composition, and sensory properties.

	Botanical diversity		SEM	*P*-value
	High (HD)	Low (LD)		
Fat in dry matter, g 100 g^{-1}	51.4	47.5	0.44	<0.001
Water-soluble N / total N, g 100 g^{-1}	34.0	36.4	4.13	0.705
Phosphotungstic acid soluble N / water-soluble N, g 100 g^{-1}	30.3	32.1	0.72	0.074
Phosphotungstic acid soluble N / total N, g 100 g^{-1}	10.3	11.7	1.52	0.543
Brightness of the curd, L* index	74.5	70.9	1.02	0.068
Yellowness of the curd, b* index	24.1	21.4	0.72	0.053
Spreadability index (*cis*-9 C18:1-to-C16:0 ratio)	0.95	0.92	0.05	0.666
Firmness, sensory score 0-10	5.7	6.4	0.27	0.092
Global flavour, sensory score 0-10	6.0	5.6	0.13	0.128
Dry fruits odour, sensory score 0-10	3.2	2.6	0.13	0.038
Dry fruits flavour, sensory score 0-10	3.1	2.5	0.11	0.004

Table 2. Effect of the botanical diversity of the pasture on the number of ASVs and the Shannon diversity index (*H'*) in simulated bites, milk, cheese core, and cheese rind.

	Number of ASVs				Shannon diversity index (*H'*)			
	High (HD)	Low (LD)	SEM	*P*-value	High (HD)	Low (LD)	SEM	*P*-value
Simulated bites	295.8	242.3	14.18	0.016	4.21	3.70	0.102	0.002
Milk	144.6	113.6	15.41	0.163	2.35	2.74	0.348	0.462
Cheese core	44.1	35.6	3.87	0.141	0.40	0.34	0.042	0.325
Cheese rind	44.6	37.5	2.66	0.083	1.57	1.59	0.060	0.816



C

ASV name	Relative abundance in bacterial community (%): Simulated bites	Milk	Cheese core	Cheese rind	Species
2535950da4f5ce2e05509946880e1912	0.20	5.94	93.03	49.40	*Lactococcus lactis*
4b7d82b0d94caafd265a16ec66ded57e	0.005	0.004	0.008	17.31	*Brachybacterium sp.*
17dfc19d75554282393e010d9c051027	0.018	0.0009	0.0061	8.20	*Brevibacterium aurantiacum*

Figure 1. Number of ASVs shared between bacterial compartments derived from the two pasture types (A), distribution of common ASVs according to the pasture type (B), and relative abundance in the bacterial communities of the main ASVs that were common to the two pasture types (C).

explained by their specific bacterial community profile. Eventually, the botanical diversity of pastures may contribute to the shaping of the bacterial communities of milk and cheese through the transfer of bacteria from the grassland's surface to raw milk. Other microbial communities (i.e. fungi) and ecosystems, such as the soil, the rumen, as well as the teat-skin surface, will be investigated to unravel the potential pathways of microbial transfer.

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References

Frétin M., Martin B., Rifa E., Verdier-Metz I., Pomiès D., Ferlay A. ... and Delbès C. (2018) Bacterial community assembly from cow teat skin to ripened cheeses is influenced by grazing system. *Scientific Reports* 8, 200.

Manzocchi E., Martin B., Bord C., Verdier-Metz I., Bouchon M., De Marchi M. ... and Coppa M. (2021) Feeding cows with hay, silage, or fresh herbage on pasture or indoor affects sensory properties and chemical composition of milk and cheese. *Journal of Dairy Science* 104, 5285-5302.

Martin B., Verdier-Metz I., Buchin S., Hurtaud C. and Coulon J.-B. (2005) How do the nature of forages and pasture diversity influence the sensory quality of dairy livestock products? *Animal Science* 81, 205-212.

Theil S. and Rifa E. (2021) rANOMALY: Amplicon workflow for microbial community analysis. *F1000 Research* 10, 7.

Tornambé G., Cornu A., Verdier-Metz I., Pradel P., Kondjoyan N., Figueredo G. ... and Martin B. (2008). Addition of pasture plant essential oil in milk: influence on chemical and sensory properties of milk and cheese. *Journal of Dairy Science* 91, 58-69.

The effect of the addition of a companion forage to a perennial ryegrass sward on lamb performance

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Abstract

Ireland's competitive advantage in sheep meat production is based on the efficient and cost effective production and utilization of pasture. The objective of this study was to assess the influence of binary mixtures of perennial ryegrass (*Lolium perenne* L.) plus a companion forage on lamb performance at pasture. Five sward mixtures were investigated, namely: (1) perennial ryegrass (*Lolium perenne* L.; PRG); (2) PRG and white clover (*Trifolium repens* L.:PRG+WC); (3) PRG and red clover (*Trifolium pratense* L.;PRG+RC); (4) PRG and plantain (*Plantago lanceolate* L.;PRG+Plan); and (5) PRG and chicory (*Chicorium intybus* L.;PRG+Chic). Five farmlets with one sward treatment assigned to each were established, which 23 ewes plus their lambs grazed from March to December for the years 2018 to 2021. Farmlets were stocked at 11.5 ewes ha^{-1}. Lambs were weaned on average at 15 weeks of age. Post-weaning a leader-follower grazing system was implemented with lambs grazing ahead of the ewes. Animal performance was monitored fortnightly with lambs drafted at liveweights of 43-46 kg targeting a 20 kg carcass. The number of days to reach slaughter was calculated as the number of days from birth to slaughter. Results show that lambs grazing swards containing a companion forage had reduced days to slaughter of between 16.0 days (PRG+Plan) (P<0.001) and 28.3 days (PRG+RC) (P<0.001) relative to those grazing PRG-only swards.

Keywords: clover, herb, perennial ryegrass, lamb performance

Introduction

Perennial ryegrass is the most commonly sown grass variety in Ireland accounting for 95% of forage grass seed sales (DAFM, 2021). Perennial ryegrass monocultures have the potential to produce high nutritive value dry matter yields, which can support high levels of animal performance; however, they require high levels of inorganic nitrogen inputs in order to achieve this (Moloney *et al.*, 2020). The need to reduce dependence on inorganic nitrogen application is an issue of increasing importance as a result of N price volatility and availability issues and the EU 'Farm to Fork strategy' which aims to reduce fertilizer usage by 20% by 2030 (EU, 2020). In recent years numerous studies have been carried out on more botanically diverse swards which have shown the potential for increased sward production and animal performance and reductions in nitrogen requirements in comparison to perennial ryegrass monocultures. In a study by Grace *et al.* (2019) ewe and lamb performance at pasture was improved for those grazing a multispecies sward, which yielded similar levels of dry matter herbage production at 90 kg N ha^{-1} to that of a perennial ryegrass sward receiving 163 kg N ha^{-1}. The objective of this study was to investigate the effect of binary mixtures of perennial ryegrass and a companion forage on lamb performance in an intensive pasture-based sheep production system.

Materials and methods

This study was undertaken at the Sheep Research Demonstration Farm, Teagasc, Animal and Grassland Research Centre, Mellows Campus, Athenry, Co. Galway, Ireland (54°80'N; 7°25'W) from January 2018 for 4 production years (2018-2021). A complete randomized block design was used to determine the effect of sward type on ewe and lamb performance and output in an intensive grazing system. The sward types under investigation were: (1) perennial ryegrass (*Lolium perenne* L.; PRG); (2) PRG and white clover (*Trifolium repens* L.:PRG+WC); (3) PRG and red clover (*Trifolium pratense* L.;PRG+RC); (4) PRG and plantain (*Plantago lanceolata* L.;PRG+Plan); and (5) PRG and chicory (*Cichorium intybus* L.;PRG+Chic). Experimental farmlets were established in 2017 at seeding rates of 10 kg ha^{-1} of perennial ryegrass, with the addition of 2.5 kg ha^{-1} white clover, 3.25 kg ha^{-1} red clover, 1.5 kg ha^{-1} plantain, and 1.5 kg ha^{-1} chicory in each of the respective mixtures. The farmlets were stocked at 11.5 ewes ha^{-1} and managed in a 5-paddock rotational grazing system. The study consisted of 115 Texel × Belclare ewes (n=23 per sward type) which were blocked for age, breed and parity and balanced for body weight, body condition score and litter size and mated to Texel rams, with a mean lambing date of March 7^{th}. Ewes and their lambs were randomly assigned to one of the five sward types. All lambs were tagged, weighed, and matched to their dams and all male lambs were castrated within 24 hours of birth. Post-lambing, ewes and lambs were turned out to pasture within their allocated sward type. Lambs were weaned on average at 15 weeks of age, with a leader-follower grazing system in place thereafter. Target pre-grazing sward heights were 7-9 cm (1,200-1,500 kg dry matter (DM) ha^{-1}) across all treatments for the duration of the experiment. Target post-grazing sward height was 4.0 cm for the first rotation and 4.5 cm for all subsequent pre-weaning rotations. Post-weaning lambs were removed from the paddocks at a target post-grazing height of 6 cm, with ewes immediately introduced to graze to a target post-grazing height of 4.5 cm. Inorganic fertilizer application rates were applied at a rate of 130 kg N ha^{-1} yr^{-1} in the form of protected urea. Lambs were drafted at live weights of 42-46 kg over the months June-October respectively to produce a target carcass weight of 20 kg. Lambs that were not drafted by October in each treatment were housed indoors and finished on *ad libitum* grass silage and concentrates when grass supply dropped below 50 kg DM ewe^{-1} ha^{-1} or when lamb growth rate dropped below 100 g day^{-1}. Lambs were weighed at birth, 6, 10 and 15 weeks of age (weaning), and at 2-week intervals from weaning to slaughter. All lambs received an anthelmintic treatment at 6 weeks of age to combat *Nematodirus* infection. Beginning from 6 weeks of age, 1 group faecal sample per group was collected fortnightly and faecal egg counts were determined using the FECPAK technique. Subsequent anthelmintic treatments were administered when faecal egg counts exceeded 500 eggs per gram. Average daily gain (ADG) and days to slaughter (DTS) were calculated accordingly. Carcass conformation was scored using the EUROP grid system (E=excellent and P=poor) and external fat score was scored using a one to five scoring system in order of increasing fatness (1=low fat cover; 5=high fat cover). Dressing proportion was calculated for each lamb as cold carcass weight divided by the pre-slaughter live weight. Lifetime ADG = drafting live weight minus birth weight divided by number of days required to reach slaughter (DTS), which was lamb age at slaughter. Lamb performance was analysed using mixed model in SAS 9.4 with sward type, year, litter size, sex and dam parity included as fixed effects and dam included as a random effect. Deviations in age at weaning from the group mean were also included as a fixed effect for pre-weaning ADG and weaning weight.

Results and discussion

In this study lamb performance was separated into two production stages, pre-weaning (from birth until weaning) and post-weaning (from weaning until slaughter date). Results, as per Table 1, show that sward type had a significant effect on lamb weaning weight and pre-weaning ADG. Lambs grazing PRG+RC had a significantly higher weaning weight and pre-weaning ADG than all other sward types ($P<0.05$) with the exception of PRG+Chic.

Post-weaning ADG was significantly affected by sward type ($P<0.001$). Lambs grazing PRG+Chic had the highest post-weaning ADG of 176 g day^{-1}. This was significantly higher than PRG+WC, PRG+RC and PRG+Plan ($P<0.05$), which were again significantly higher than PRG at 133 g day^{-1} ($P<0.001$). Lambs grazing PRG+Chic had a significantly greater lifetime ADG than lambs on all other sward types with the exception of PRG+WC ($P<0.05$).

Table 1. Lamb performance 2018-2021.[1,2]

	PRG	PRG + WC	PRG + RC	PRG + Plan	PRG + Chic	SEM	*P*-value
ADG birth – weaning (g day^{-1})	237^{a}	238^{a}	251^{b}	236^{a}	243^{ab}	1.3	<0.05
Weaning weight (kg)	29.5^{a}	29.5^{a}	31.1^{b}	29.7^{a}	30.2^{ab}	0.55	<0.05
ADG weaning – slaughter (g day^{-1})	133^{a}	155^{b}	162^{b}	158^{b}	176^{c}	5.4	<0.001
ADG lifetime (g day^{-1})	184^{a}	205^{bc}	213^{b}	200^{c}	214^{b}	5.2	<0.001
Days to slaughter	228.3^{a}	209.4^{bd}	200.0^{c}	212.4^{b}	200.5^{cd}	4.56	<0.001
Carcass conformation	2.41	2.50	2.55	2.49	2.51	0.074	NS
Fat score	3.04	3.05	3.00	2.93	2.88	0.083	NS
Dressing proportion	0.46	0.46	0.45	0.45	0.45	0.004	NS

[1] Carcass conformation was scored using the EUROP grid system (E=excellent and P=poor), and expressed where E=1, U=2, R=3, O=4, P=5, external fat score was scored using a one to five scoring system in order of increasing fatness (1=low fat cover; 5=high fat cover), dressing proportion was calculated for each lamb as cold carcass weight divided by the pre-slaughter live weight.

[2] Values in a row with different superscript letters are significantly different ($P<0.05$); NS = not significant.

Average days to slaughter for lambs grazing PRG was 228.3 days, which was reduced by 18.9, 28.3, 16.0 and 27.8 days for lambs grazing PRG+WC, PRG+RC, PRG+Plan and PRG+Chic respectively ($P<0.001$). Furthermore, this led to reduced rates of concentrate supplementation required where average concentrates consumed per lamb drafted was reduced by 6.1, 11.3, 8.2 and 10.6 kg concentrates per lamb drafted for lambs grazing PRG+WC, PRG+RC, PRG+Plan and PRG+Chic respectively in comparison with lambs grazing PRG, receiving 14.2 kg concentrates per lamb drafted ($P<0.001$). Sward type had no significant effect on carcass grade, fat score or dressing proportion.

Conclusions

Binary sward mixtures containing perennial ryegrass and a companion forage (white clover, red clover, plantain or chicory) have the potential to support increased lamb performance above that of a perennial ryegrass monoculture in terms of pre-weaning, post-weaning and lifetime ADG and subsequent days to slaughter.

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References

DAFM (2021) *Grass and White Clover Varieties, Irish Recommended List 2021*. Department of Agriculture Food and the Marine.

EU (2020) European Commission Farm to Fork Strategy 2020. European Union.

Grace C., Lynch M.B., Sheridan H., Lott S., Fritch R. and Boland T.M. (2019) Grazing multispecies swards improves ewe and lamb performance. *Animal* 13, 1721-1729.

Moloney T., Sheridan H., Grant J., O'Riordan E. and O'Kiely P. (2020) Yield of binary-and multi-species swards relative to single-species swards in intensive silage systems. *Irish Journal of Agricultural and Food Research* 59, 12-26.

Climate impacts due to albedo change of grassland through grazing and mowing practices in various pedoclimatic situations

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Abstract

Ruminant farming impacts climate change (CC) because of land use and greenhouse gas emissions. Grasslands management also affects the climate by changing land surface albedo (α). Adapted grassland management, in order to increase α, could be a lever for CC mitigation as well as soil carbon storage. This paper examined how mowing and grazing practices influence albedo, using one year of continuous α measurements on seven French grasslands. Daily field measurements were used to analyse α changes following management practices and environmental conditions. Grazing decreased albedo by 4% during 15 days on average. At one of the plots, α decreased by 14% after mowing and 7% after grazing, during 31 and 23 days, respectively. From a perspective of CC mitigation, our results suggest that grassland management should be adapted to preserve and enhance surface albedo.

Keywords: albedo, grassland, ruminant, climate change, mitigation

Introduction

Ruminant farming contributes to climate change through greenhouse gas (GHG) emissions that can be partially compensated by soil carbon sequestration. Furthermore, as land surface properties regulate the exchange of energy and water between the land and the atmosphere, their biogeophysical properties influence climate from local to global scales. For instance, grassland management changes land surface reflectivity (albedo, α), emissivity and evapotranspiration. The surface α, represents the fraction of incident solar radiation reflected from the ground. It controls the amount of energy available at the surface and in the Earth system. Increasing α leads to a reduction in net shortwave radiation at the surface and at the top of atmosphere, with the potential to cool local and global mean temperatures. Increasing α on grasslands could reduce local warming and compensate part of their greenhouse gas emissions. If the α effect has been well studied for forests (e.g. Bonan *et al.*, 2008) and crops (e.g. Carrer *et al.*, 2018), few studies have been conducted on grasslands (e.g. Yang *et al.*, 2019). But, in not considering the seasonal variability of α, due to crops or grassland growth and management, as done in the previous IPCC reports, leads to strong over or underestimations of the albedo effects (Bright *et al.*, 2015). Results presented here come from the Albedo-prairies project, which aims to: (1) characterize the α dynamics resulting from soil-climate variability in mowed or grazed grasslands; and (2) to quantify the magnitude of the mitigation effect compared to carbon storage. This comparison is a methodological challenge that will be studied later in this project. In this paper, focus on the α change resulting from grassland management.

Materials and methods

Since July 2020, surface α has been continuously measured at seven French experimental farms with contrasting grassland management and pedoclimates (Table 1). Temperature, hygrometry, and soil moisture are also continuously monitored to analyse the surface α dynamics. Mowing, grazing dates and soil wetting events are collected. Albedo change (Δ_α) following precipitation or management events were calculated as the difference in α between a reference status (α_{Ref}, i.e. before the event) and the mean α (α_{mean}) measured during the whole period (p) following the event and until α returns to ±2.5% of the α_{Ref} value or, if mowing or grazing occurred before the end of the expected recovery in α_{mean} till the α_{Ref} value (Figure 1A). Therefore, the mean α change (Δ_α) relative to α_{Ref} was calculated according to the following equation:

$$\Delta_\alpha(\%) = -\left(1-\left(\frac{\left(\sum\frac{\alpha mean}{p}\right)}{\alpha_{Ref}}\right)\right)$$

Table 1. Characteristics of each experimental sites.

Experimental farm	Trévarez	Derval	Rheu[1]	Thorigné	Mourier	Jalogny	Pradel
Grassland type	Temporary	Temporary	Temporary	Temporary	Permanent	Permanent	Permanent
Type of livestock	Dairy cow	Dairy cow	Dairy cow	Beef	Sheep	Beef	Goats
Climate	Oceanic	Oceanic	Altered oceanic	Altered oceanic	Altered oceanic	Semi continental	Meso Mediterranean
Precipitation (mm)	1219	726	855	724	930	856	842

[1] IE PL, INRAE, 2021. Dairy nutrition and physiology, https://doi.org/10.15454/yk9q-pf68.

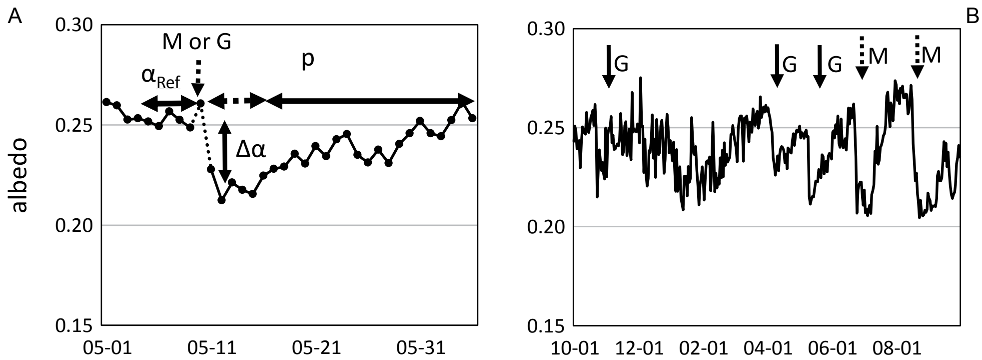

Figure 1. On the left (A), schematic representation of the method for estimating α change and its duration following an event (mowing, grazing, rain). On the right (B), α dynamic over 1 year (Oct. 2020 – Oct.2021) at the Trévarez farm.

Results and discussion

The measurements taken in 2020-21 recorded 37 grazing and 3 mowing events, as well as 30 soil wetting events following a period without rain. Albedo dynamics differed among sites and were caused by differences in management practices, rapid changes in vegetation and fraction of exposed soil (Figure 1B). On average, grazing decreased albedo by 4% (α=-0.008) for 15±9 days, with the lowest decrease observed at the Jalogny farm (-1.5% ≈ α=-0.003, for 14 days) associated with the lowest instantaneous daily stocking rate (3.3 LU day^{-1}). The biggest decrease in α was observed at Trévarez (-7% ≈α=-0.017), with a short grazing time (3.8 days) but among the highest stocking rates (43.1 livestock units (LU) day^{-1}) (Table 2). Those grazing effects are significant, as Carrer *et al.* (2018) simulated an increase in surface albedo equal to 0.002, following the introduction of cover-crops for 3 months, over 4.5% of the arable lands in Europe (EU-28), that led to a cooling effect equivalent to 2.92 $MtCO_2$-eq.$year^{-1}$. If our results show a poor linear relationship between the mean albedo decrease and the daily stocking rate expressed in LU ha^{-1} day^{-1} (R^2=0.14), considering the cumulative decline in α improved this relationship (R^2=0.24). Results are significantly better with R^2=0.39 when considering the cumulative decline of α and the daily stocking rate weighted by the % of grass consumed in the animal ration during the grazing period (called 'grazing pressure' in Table 2, expressed in LU-eq ha^{-1} day^{-1}). However, this is a rough estimate of the amount of grass taken from the plot. Only three mowing events occurred, two of which concern the Trévarez farm. On average, α decreased by 14% (against 7% for grazing) for 31 days (23 for grazing) (Figure 1B). Those results suggest that α could be affected more by mowing than by grazing.

Table 2. Grazing and rainfall effects on surface albedo at the seven recorded grassland sites.

Experimental farm	Trévarez	Derval	Rheu[1]	Thorigné	Mourier	Jalogny	Pradel
Mean α effect ± sd	0.241±0.03	0.236±0.05	0.215±0.01	0.223±0.07	0.241±0.06	0.216±0.04	0.225±0.06
Grazing (n) / duration (d)	(5) / 3.8	(9) / 5.9	(6) /6.7	(5) /6	(5) / 4.4	(4) / 15	(4) / 4.8
Stocking rate LU ha^{-1} day^{-1}	43.1	31.9	36.6	8.3	23.1	3.3	3.6
Grazing pressure[2]	27.2	12.5	20	8.3	22.3	3.3	2.5
Mean α decrease	-7.00%	-3.10%	-2.10%	-5.30%	-6.40%	-1.50%	-1.40%
α cumulative decrease	-176%	-58%	-26%	-97%	-121%	-26%	-5%
$α_{mean}$ decrease duration (d)	23.2	13.4	10.8	17.2	17.4	14.5	6
Soil wetting event (n)	No period	-5	-7	-3	-4	Regularly under	-11
α average decrease	available	-0.70%	-6.40%	-18.20%	-9.50%	water	-5.90%
α cumulative decrease		-8%	-116%	-134%	-155%		-280%
$α_{mean}$ decrease duration (d)		6.6	7.6	11.3	10.5		13.9

[1] IE PL, INRAE, 2021. Dairy nutrition and physiology, https://doi.org/10.15454/yk9q-pf68.

[2] Grazing pressure = [stocking rate] × [%grass in the ration]. Example: 2.5 = [3.6 LU ha^{-1} day^{-1}] × [70% grass in the ration].

When rain occurred following a dry period (see soil wetting effect in Table 2), α decreased on average by 7%, for 10.5 days (±8). This effect was stronger when the soil was not covered by vegetation, particularly in 2020 after a drought. This is because bare soil α is lower than vegetation albedo at our sites. Therefore, degraded pastures leaving the soil visible could cause a 'warming effect', probably reinforced by a decrease in evapotranspiration and an increase in sensible heat fluxes (Bright *et al.*, 2017). At this stage, further investigation through acquisition of additional data is needed to evaluate how differences in grassland management affect the albedo dynamics, and whether mowing or grazing intensity may have effects on the albedo-induced radiative forcing and on climate in general.

Conclusions

Grazing and mowing reduce surface albedo, which suggests that adapting the intensity of those practices may contribute to climate cooling through the albedo effects on the radiative forcing and other biogeophysical effects (e.g. decrease in sensible heat fluxes). Also, we show that rain events cause a stronger decrease in surface albedo when vegetation is not fully covering the soil. Longer observations in contrasting pedoclimatic conditions and for contrasting management regimes are required to propose adaptations of the current grassland management regimes to enhance climate mitigation based on the albedo effects. Also, comparisons of the albedo effects (converted in eq-CO_2) with the carbon sequestration potential and the GHG emission of different farming systems will be needed to identify the practices that are most efficient for climate mitigation.

References

Bonan G.B. (2008) Forests and climate change: forcings, feedbacks, and the climate benefits of forests. *Science* 320, 1444-1449.

Bright R., Zhao K., Jackson R.B. and Cherubini F. (2015) Quantifying surface albedo and other direct biogeophysical climate forcings of forestry activities. *Global Change Biology* 21(9), 3246-3266.

Bright R., Davi, E., O'Halloran T., Pongratz J., Zhao K. and Cescatti A. (2017) Local temperature response to land cover and management change driven by non-radiative processes. *Nature Climate Change* 7(4), 296-302.

Carrer D., Pique G., Ferlicoq M., Ceamanos X. and Ceschia E. (2018) What is the potential of cropland albedo management in the fight against global warming? A case study based on the use of cover crops. *Environmental Research Letters* 13, 4.

Yang Y. and Wang K. (2019) The effect of different land use patterns on the microclimate and ecosystem services in the agro-pastoral ecotone of Northern China. *Ecological Indicators* 106, 105552.

Evaluating GHG emissions and profitability of innovative grassland-based farming systems on a Dutch peat meadow

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Abstract

The majority of Dutch peatlands are drained and used for dairy farming. However, lowering the water table alters the hydrology of the ecosystem resulting in increased greenhouse gas (GHG) emissions, due to the oxidation of peat. Innovative grassland-based farming systems are needed to enable more sustainable production on peat for the future. We evaluated three scenarios for the Rondehoep polder in the Netherlands aimed at tackling this challenge. The scenarios can largely reduce GHG emissions (by 22-65%). For the scenarios to be viable for farmers, alternative business models are needed. Through further iteration these scenarios can provide a way forward for grassland-based farming systems on peat soils that can help achieve climate targets.

Keywords: peatlands, dairy, soil subsidence, greenhouse gas emissions

Introduction

The majority of Dutch peatlands are drained and used for dairy farming. With lower water tables, the bearing capacity of the soil can better withstand the impact of machinery and animals (De Jong *et al.*, 2021). However, lowering the water table alters the hydrology of the ecosystem resulting in increased greenhouse gas (GHG) emissions, due to the oxidation of peat, and loss of wetland biodiversity (Tanneberger *et al.*, 2020). The Dutch Climate Agreement has promised to reduce the emissions from peatlands in the Netherlands by 1 Mton by 2030 (Rijksoverheid, 2019). This study evaluated the GHG emission reduction potential and profitability of three management scenarios on Dutch peat meadow the Rondehoep.

Materials and methods

This study took place from autumn 2020 to autumn 2021 as part of the TiFN Regenerative Farming project. The Rondehoep polder (52.268° N, 4.900° E) was used as a case study (Figure 1A). The agricultural area of the polder is 1,060 ha and managed with attention to the meadow bird population. In the centre of the polder is a 160 ha meadow bird reserve. There are 18 dairy farmers on the polder with predominantly Holstein-Friesian cows. The water table depth (WTD) is maintained around -40 cm during the summer to raise the carrying capacity of the land for grazing and machinery. The WTD of the polder is controlled with inlet and outlet pumps.

Calculations for GHG emissions associated with three scenarios (Table 1) were made using the approach of Couwenberg *et al.* (2011). These calculations were based on average WTD. An emission reduction target for the polder was calculated based on the national emission reduction target for peatlands and the total peatland area of the Netherlands under agricultural use (Born *et al.*, 2016; Rijksoverheid, 2019). Profit calculations were made on polder level and were based on income from milk production, income from subsidies, and costs related to fertilizer use and concentrates. Milk production was estimated by accounting for the production of different cow breeds, the related grass production per WTD, and the stocking density (LSU) of the polder.

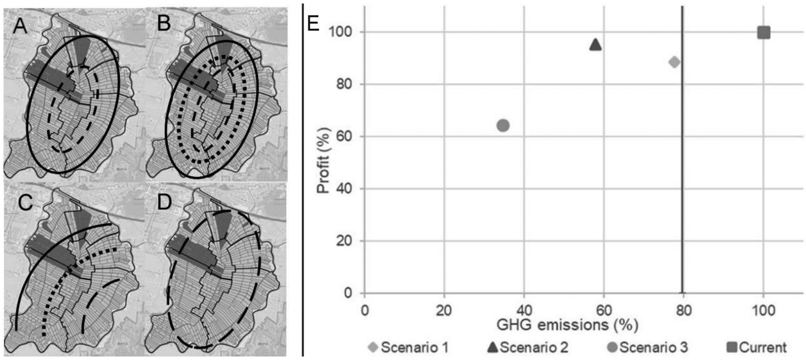

Figure 1. (A) Current situation. (B) Scenario 1. (C) Scenario 2. (D) Scenario 3. The dashed line indicates low intensity farming, the dotted line medium intensity, and the solid line high intensity (Table 1). (E) The performance of the scenarios on profitability and greenhouse gas emissions (in CO_2-eq) is provided relative to the current situation. The line signifies the emission reduction target for peatlands set by the Dutch Climate Agreement (Rijksoverheid, 2019).

Table 1. Specifications for the scenarios used in the evaluation. Zones are specified by the line type depicted in Figure 1.

	Current (A)		Scenario 1 (B)			Scenario 2 (C)			Scenario 3 (D)
Zone	Solid	Dashed	Solid	Dotted	Dashed	Solid	Dotted	Dashed	Dashed
WTD (cm)	-50	-30	-40	-40	-20	-40	-30	-20	-20
Pasture type	perennial ryegrass	permanent species-rich	perennial ryegrass	temporary species-rich	permanent species-rich	perennial ryegrass	temporary species-rich	permanent species-rich	permanent species-rich
Grazing system(s)	rotational grazing, (rotational) continuous grazing		rotational grazing, rotational continuous grazing			rotational grazing, rotational continuous grazing	rotational continuous grazing	(rotational) continuous grazing	(rotational) continuous grazing
Cow breed(s)	Holstein-Friesian, Holstein-Friesian crossbreed		Holstein-Friesian, Montbeliarde, Fleckvieh, Maas-Rijn-IJssel			Holstein-Friesian	Jersey	Blaarkop	Blaarkop, Jersey
Livestock intensity (LSU ha^{-1})	2		1.9		1.2	1.9	1.6	1.2	1.2
Sources of additional income	subsidies species-rich grasslands		subsidies species-rich grasslands			milk of higher quality, subsidies species-rich grasslands			milk of higher quality, subsidies species-rich grasslands

Results and discussion

Figure 1 shows the three scenarios that were developed and their performance relative to the current situation. Table 1 provides the specifications for each scenario. All proposed scenarios meet the emission reduction target for peatlands set by the Dutch Climate Agreement. This gives perspective to interested parties on what can be done on a regional level to meet national climate targets. At this emission reduction target, there is still quite a lot of GHG emissions (80% of current levels) and the peat will continue to oxidize. For long-term sustainability it is important to stop the degradation of peat. This will not happen in any of the scenarios proposed. Other options must be explored, such as integrating wet crop production at an even higher WTD.

GHG emissions and profit were the focus of the evaluation in this research, but there are many other important objectives for future-proofing of production on peat, such as improved biodiversity and nutrient cycling. It is expected that the scenarios perform better than the current situation on a multitude of ecosystem services, but further work is needed on the evaluation of the scenarios to consider their performance on these services.

Due to a lack of validation data for grasslands on peat, there is a high level of uncertainty in the calculations. In reality, the behaviour of peat is hard to predict (Abdalla *et al.*, 2016; Couwenburg, 2011). Raising the water table greatly lowers CO_2 emissions but it raises CH_4 emissions (Abdalla *et al.*, 2016). New systems must be tested to get data to validate predictions. This can provide farmers, policy makers and carbon credit investors with more certainty about the emission reduction that is actually realized.

In all scenarios milk production is reduced, indicating a need for financial support for farmers and further experimentation with alternative business models. The model calculations accounted for additional sources of income that are available today but other sources may be possible in the future such as carbon credits and payment for additional ecosystem services. Societal costs related to farming on this polder, such as costs related to water management and infrastructure, were not accounted for in the calculations. It is expected that in the scenarios these costs will be reduced.

Further research is needed into how to maintain a higher WTD as this is difficult to manage and if done wrong can lead to even higher GHG emissions.

Conclusions

The evaluated scenarios have the potential to greatly reduce GHG emissions (by 22-65%) from the polder; however, profits are reduced in all scenarios (by 5-36%). To be viable, alternative business models are needed to ensure a liveable income for farmers and that farmers are compensated for their efforts.

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References

Abdalla M., Hastings A., Truu J., Espenberg M., Mander Ü. and Smith,P. (2016) Emissions of methane from northern peatlands: a review of management impacts and implications for future management options. *Ecology and Evolution* 6, 7080-7102.

Bos A.P., Koerkamp P.G., Gosselink J.M.J. and Bokma S. (2009) Reflexive interactive design and its application in a project on sustainable dairy husbandry systems. *Outlook on Agriculture* 38, 137-145.

Couwenberg, J., Thiele, A., Tanneberger, F., Augustin, J., Bärisch, S., Dubovik, D., ... and Joosten, H. (2011) Assessing greenhouse gas emissions from peatlands using vegetation as a proxy. *Hydrobiologia* 674, 67-89.

Jong, M. de, Hal, O. van, Pijlman, J., Eekeren, N. van, and Junginger, M. (2021) Paludiculture as paludifuture on Dutch peatlands: An environmental and economic analysis of Typha cultivation and insulation production. *Science of the Total Environment* 792, 1-12.

Rijksoverheid. (2019) Klimaatakkoord. Den Haag.

Tanneberger, F., Schröder, C., Hohlbein, M., Lenschow, U., Permien, T., Wichmann, S., and Wichtmann, W. (2020) Climate change mitigation through land use on rewetted peatlands – cross-sectoral spatial planning for paludiculture in Northeast Germany. *Wetlands* 40, 2309-2320.

Van den Born G.J., Kragt F., Henkens D., Rijken B., van Bemmel B., and van der Sluis S. (2016) Dalende bodems, stijgende kosten: mogelijke maatregelen tegen veenbodemdaling in het landelijk en stedelijk gebied: beleidsstudie. Planbureau voor de Leefomgeving, Den Haag, the Netherlands.

The effect of sward type and fertilizer rate on milk production of spring calving, grazing dairy cows

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Abstract

Grazed grass is considered the cheapest feed available for dairy cows in temperate regions. The objective of this study was to quantify the effect of sward type (perennial ryegrass (*Lolium perenne L.*; PRG)) sown with and without white clover (*Trifolium repens L.*; WC) and nitrogen (N) fertilizer rate (150 and 250 kg N ha^{-1}) on milk production of grazing dairy cows. 120 cows were randomly divided to each of the four grazing treatments (PRG-only receiving either 150 or 250 kg N ha^{-1} and PRG-WC receiving either 150 or 250 kg N ha^{-1}) as they calved, and swards were rotationally grazed at stocking rate of 2.75 cows ha^{-1}. There was a significant effect of sward type on milk production (P>0.001). Over the three-year study, cows grazing the PRG-WC treatments had greater milk yield (+222 kg) and milk solids (kg fat + protein) yield (+27 kg) than cows grazing the PRG-only treatments. Nitrogen fertilizer rate did not affect milk production but did affect herbage production. This significant increase in milk production suggests the inclusion of white clover in grazing systems can be used effectively to increase milk production and reduce nitrogen use.

Keywords: perennial ryegrass, white clover, milk production

Introduction

Grazed grass is the cheapest feed source of nutrients available for dairy cows and dairy farmers must maximize the use of this high quality feed where possible (Finneran *et al.,* 2012). To reduce cost inputs and environmental impacts of inorganic nitrogen (N) use (Hoekstra *et al.*, 2020) and to increase farm gate N-use efficiency (NUE - (Chapman *et al.*, 2020)), there is renewed interest in the incorporation of legumes, and white clover (*Trifolium repens L.*; WC) in particular, in perennial ryegrass (*Lolium perenne* L.; PRG) pasture-based production systems (Lüscher *et al.*, 2014). An increase in milk production from cows grazing PRG-WC swards has also been observed and can be attributed to an overall increase in herbage dry matter (DM) intake from PRG-WC swards and to the high nutritional value of WC (Ribeiro Filho *et al.,* 2005). The objective of this study was to determine the effect of WC inclusion in PRG swards and N fertilizer rate on milk production of spring-calving grazing dairy cows. The hypothesis of the experiment was that milk production of spring-calving dairy cows would reduce when inorganic N fertilizer was lowered but could be replaced by the inclusion of WC in the swards.

Materials and methods

The experiment was undertaken at Clonakilty Agricultural College, Clonakilty, Co. Cork, Ireland from February 2019 to November 2021. The experiment was a 2×2 factorial design; two sward types (PRG-only and PRG-white clover) at two fertilizer rates (150 and 250 kg N ha^{-1}). This resulted in four separate grazing treatments; a PRG-only sward receiving 150 kg N ha^{-1} (GO-150), a PRG-only sward receiving 250 kg N ha^{-1} (GO-250), a PRG-white clover sward receiving 150 kg N ha^{-1} (GC-150) and PRG-white clover sward receiving 250 kg N ha^{-1} (GC-250). Twenty blocks of four paddocks were created and balanced for location, topography and soil type. Each treatment was randomly assigned in each block and a separate farmlet of 20 paddocks was created for each treatment. There were 30 cows per treatment and each treatment was stocked at 2.75 cows ha^{-1}, with four breeds used and balanced for amongst each treatment: Holstein-Friesian (HF), Jersey x HF, Norwegian Red × (Jersey × HF) and HF × (Norwegian Red × (Jersey × HF)). Within breed, cows were assigned to treatment based on calving date, parity, pre-experimental milk yield (mean day 7.8 milk yield post-calving) and economic breeding index. Cows had a mean calving date of 8th of February, 284 DIM and were on a silage only diet over the winter dry period (December and January). Cows remained in their treatments for the entire grazing season in each year and were re-randomized and assigned to treatments each experimental year. Treatments were rotationally grazed from early-February to mid-November each year and target post-grazing sward height was 4 cm. Nitrogen fertilizer applications were similar for all treatments in late-January, mid-March and April. Thereafter, the 150 kg N ha^{-1} treatments received 40% of the 250 kg N ha^{-1} treatment rate for each subsequent rotation and received 50% for the final rotation. Each farmlet was walked weekly to monitor average farm cover (Hanrahan *et al.,* 2017) and when surpluses arose they were removed in the form of baled silage. If a feed deficit occurred across all treatments then all treatments were supplemented with concentrate; on average, 598 kg fresh weight concentrate was fed per year per cow across all treatments. If a feed deficit occurred in an individual treatment then cows were supplemented with conserved forage produced from within that treatment. Pre-grazing herbage yield in each paddock was determined twice weekly by harvesting 2 strips using an Etesia mower (Etesia UK Ltd., Warwick, UK) in the area to be grazed next. Pre- and post-grazing heights were measured daily using a rising plate meter (Jenquip, Fielding, New Zealand). Sward WC content was measured before grazing in each paddock in each rotation by cutting 15 random grab samples to 4 cm with a Gardena hand shears, separating the sample into PRG and WC fractions and drying at 90 °C for 16 h. Milk yield was recorded daily and milk composition weekly by taking milk samples from a consecutive evening and morning milking. Data from 120 cows over three years (360 variables) were available for analysis. Grazing characteristics (Table 1) were analysed using Proc MIXED in SAS (SAS 9.4). Terms included in the model were year, block, rotation, WC treatment, fertilizer rate treatment, and their interactions. Due to the Covid-19 pandemic in 2020, full grazing season data were not collected so only results from 2019 and 2021 are presented. Milk data were also analysed using Proc MIXED in SAS (SAS 9.4). Terms included in the model were year, WC treatment, fertilizer rate treatment, parity, breed and their interactions.

Results and discussion

The effect of sward type and N fertilizer rate on grazing characteristics is presented in Table 1. There was an interaction between sward type and N fertilizer rate on pre-grazing yield as the GO-150 treatment had a lower pre-grazing yield than GC-150 whereas there was no difference in pre-grazing yield between GO-250 and GC-250. There was also an interaction between sward type and N fertilizer rate for pre-grazing sward height. Post-grazing sward height was lower for the 150 kg N ha^{-1} treatments compared to the 250 kg N ha^{-1} treatments (4.09 vs 4.16 cm, P=0.004). Herbage allowance was not affected by sward type or N fertilizer rate. White clover inclusion had a significant (P<0.001) positive effect (Table 2) on milk production, whereas N fertilizer rate had no effect. Over the three years, PRG-only swards produced,

Table 1. Effect of sward type (ST) and nitrogen fertilizer rate (FR; mean of entire grazing season of 2019 and 2021 only) on grazing characteristics.[1]

	GO-150	GO-250	GC-150	GC-250	SE	ST	FR	ST × FR
Pre-grazing yield (kg DM ha^{-1})	1,652	1,830	1,762	1,827	64.8	0.038	<0.001	0.027
Pre-grazing height (cm)	8.91	9.55	9.22	9.58	0.065	0.009	<0.001	0.038
Post-grazing height (cm)	4.11	4.21	4.07	4.16	0.031	0.116	0.004	0.884
Herbage allowance (kg DM cow^{-1})	16.7	17.1	17.2	17.3	0.01	0.266	0.456	0.733
Clover content (%)	-	-	18.1	15.4	1.03	-	0.086	-

[1] GO-150 = PRG-only sward receiving 150 kg N ha^{-1}, GO-250 = PRG-only sward receiving 250 kg N ha^{-1}, GC-150 = PRG-WC sward receiving 150 kg N ha^{-1}, GC-250 = PRG-WC sward receiving 250 kg N ha^{-1}.

on average, 5,549 kg milk and 468 kg milk solids (kg fat + protein) cow^{-1} $year^{-1}$. In comparison, PRG-WC swards produced 5,859 kg milk and 499 kg milk solids cow^{-1} $year^{-1}$. The difference in milk solids production between the PRG-only and PRG-WC treatments occurred due to the higher milk yield produced rather than a difference in fat or protein content, which is similar to previous results reported (Egan *et al.*, 2018). However, N fertilizer rate had no significant effect on milk production or composition over the three years.

Table 2. Effect of sward type (ST) and fertilizer rate (FR) inclusion on full lactation milk production.[1]

	GO-150	GO-250	GC-150	GC-250	SE	ST	FR	ST × FR
Milk yield (kg cow^{-1})	5,503	5,596	5,909	5,809	60.7	<0.001	0.295	0.526
Fat content (g kg^{-1})	47.8	47.9	47.6	47.4	0.04	0.478	0.938	0.727
Protein content (g kg^{-1})	38.2	38.5	38.7	38.2	0.02	0.519	0.587	<0.05
Lactose content (g kg^{-1})	46.4	46.3	46.3	46.4	0.01	0.787	0.839	0.448
Milk solids yield (kg cow^{-1})	463	472	499	499	4.8	<0.001	0.309	0.352

[1] GO-150 = PRG-only sward receiving 150 kg N ha^{-1}, GO-250 = PRG-only sward receiving 250 kg N ha^{-1}, GC-150 = PRG-WC sward receiving 150 kg N ha^{-1}, GC-250 = PRG-WC sward receiving 250 kg N ha^{-1}.

Conclusions

The inclusion of WC in PRG swards had a significant positive affect on milk yield and solids production. Reducing N fertilizer rate from 250 to 150 kg N ha^{-1} did not affect milk production.

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References

Chapman D., Pinxterhuis I., Ledgard S. and Parsons T. (2020) White clover or nitrogen fertilizer for dairying under nitrate leaching limits? *Animal Production Science* 60, 78-83.

Egan M., Galvin N. and Hennessy D. (2018) Incorporating white clover (Trifolium repens L.) into perennial ryegrass (*Lolium perenne* L.) swards receiving varying levels of nitrogen fertilizer: Effects on milk and herbage production. *Journal of Dairy Science* 101, 3412-3427.

Finneran E., Crosson P., O'Kiely P., Shalloo L., Forristal D. and Wallace M. (2012) Stochastic simulation of the cost of home-produced feeds for ruminant livestock systems. *Journal of Agricultural Science* 150, 123-139.

Hanrahan L., Geoghegan A., O'Donovan M., Griffith V., Ruelle E., Wallace M. and Shalloo L. (2017) PastureBase Ireland: A grassland decision support system and national database. *Computers and Electronics in Agriculture* 136, 193-201.

Hoekstra N.J., Schulte R.P.O., Forrestal P.J., Hennessy D., Krol D. J., Lanigan G.J., Müller C., Shalloo L., Wall D.P. and Richards K.G. (2020) Scenarios to limit environmental nitrogen losses from dairy expansion. *Science of the Total Environment* 707, 134606.

Lüscher A., Mueller-Harvey I., Soussana J.F., Rees R.M. and Peyraud J.L. (2014) Potential of legume-based grassland-livestock systems in Europe: a review. *Grass and Forage Science* 69, 206-228.

Ribeiro Filho H., Delagarde R. and Peyraud J. (2005) Herbage intake and milk yield of dairy cows grazing perennial ryegrass swards or white clover/perennial ryegrass swards at low-and medium-herbage allowances. *Animal Feed Science and Technology* 119, 13-27.

Can a urease inhibitor improve the efficacy of N use under Irish grazing conditions?

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Abstract

This study sought to determine whether urease inhibited urea could be a more efficient nitrogen (N) fertilizer than other forms of applied N, under Irish conditions when rotationally grazed. A 3×2 factorial plot arrangement was used to compare calcium ammonium nitrate (CAN), urea and urea + N-(n-butyl) thiophosphoric triamide (urea + NBPT) at 150 and 250 kg ha^{-1} yr^{-1}. Zero N plots were also included in the study. The study was conducted at four sites, each with 4 replicates, giving three years of data collection at two sites and two years at the other two sites. Plots were grazed in March, early April and thereafter on an approximate three-week cycle when the control treatment (CAN-250) reached a pre-grazing herbage yield of 1,500 kg of dry matter ha^{-1}. Prior to grazing, plots were sampled for pre-grazing herbage yield and fertilizer was applied after each grazing event. All four sites gave similar responses with significant differences observed between applied N rates for pre-grazing herbage yield and total herbage production but not between fertilizer types. Above 150 kg N ha^{-1}, all fertilizers had a similar N response to the lower fertilizer rate.

Keywords: CAN, urea, NBPT-urea, grazing, ryegrass, herbage production

Introduction

Inorganic nitrogen (N) fertilizer is a major contributor to ammonia (NH_3) from urea-based fertilizers and to greenhouse gas (GHG) emissions through nitrous oxide (N_2O) losses from calcium ammonium nitrate (CAN) fertilizers (Krol *et al.*, 2020). The agricultural industry in Ireland accounts for 37% of total GHG emissions, of which 10.6% comes from N fertilizers (Environmental Protection Agency (EPA), 2020). Urease inhibitors (e.g. N-(n-butyl) thiophosphoric triamide, NBPT) reduce NH_3 volatilization from urea by inhibiting the enzyme urease which catalyses urea hydrolysis (Forrestal *et al.*, 2016). Forrestal *et al.* (2017) reported that there was no difference in herbage production between CAN, urea and urea + NBPT under a cutting regime, despite finding that urea + NBPT reduced NH_3 losses compared to urea by 79% and N_2O emissions by 71% compared to CAN. Although Forrestal *et al.* (2017) illustrated the efficacy of urea + NBPT under a cutting regime, there was slow uptake of the new technology at an industry level in Ireland (Department of the Environment, Climate and Communications (DECC), 2021). Therefore, the current study investigated the same fertilizer types (at 150 and 250 kg N ha^{-1}yr^{-1}) at four sites across Ireland, with the aim of determining what responses occur when perennial ryegrass (*Lolium perenne* L.) swards are rotationally grazed. This would entail more fertilizer application splits across the year and a greater number of defoliation events, which may encourage greater growth rates (Peters *et al.*, 2021). The two fertilizer rates were employed to strengthen industry relevance of the experiment, as a lower N fertilizer rate is desired due to environmental limitations while also observing if any differences in nitrogen response at different N rates occur (Finneran *et al.*, 2012). The hypothesis of the experiment was that urea and urea + NBPT would give similar responses in terms of pre-grazing herbage yield and herbage production to CAN under rotational grazing conditions in Ireland at different sites.

Materials and methods

The experiment was undertaken at four sites: Teagasc Moorepark, Cork (52.16° N, 8.24° W), Clonakilty Agricultural College, Cork (51°63 N, 08°85 E), Ballyhaise Agricultural College, Cavan (54°015' N, 07°031' W) and Mellows Campus, Athenry, Galway (54°80' N; 7°25' W). The design was a 3×2 factorial configuration, comparing CAN, urea and urea + NBPT at two rates (150 kg ha^{-1} and 250 kg ha^{-1}) with 4 replicates. The study was conducted from 2019 in Moorepark and Clonakilty, with Ballyhaise and Athenry added in 2020, to 2021, with 0 N plots added in all sites in 2020 giving 28 plots (8×6 m) at each site. Three sites were grazed with lactating dairy cows, whereas sheep were used in Athenry. At each site, the first grazing occurred in March six weeks after first N application and thereafter when the CAN-250 had a pre-grazing herbage yield of 1,500 kg of dry matter (DM) ha^{-1} with a target of ten rounds of grazing. Subsequent fertilizer applications were applied after each grazing event. The number of cows or sheep allocated was based on the available herbage, with the aim of having a post-grazing sward height of 4 cm. Pre-grazing herbage yield was measured prior to grazing by harvesting one strip (5×1.2 m) from each plot to a height of 4 cm using an Etesia mower (Etesia UK Ltd., Warwick, UK) at Moorepark, Clonakilty and Athenry, and from a quadrat (0.5×0.5 m) in Ballyhaise, with a 100 g subsample dried at 60 °C for 48 h to determine DM. A folding pasture plate meter with a steel plate (Jenquip, Fielding, New Zealand) was used to measure ten compressed sward heights before and after harvesting on each cut strip and also for the pre- and post-grazing sward height, on each plot. Sward density (kg of DM cm^{-1}) was calculated using the following equation: pre-grazing herbage yield / (pre-cut height – post-cut height). Sward density was then used to calculate herbage removed: kg of DM ha^{-1} removed = (pre-grazing sward height – post-grazing sward height) × sward density. Analysis was undertaken using PROC MIXED in SAS (SAS 9.4). Terms included in the model were site-year, fertilizer type, fertilizer rate, rotation and their subsequent interactions. Individual plot was the experimental unit. Site-year was included in the model as all sites were not included every year within the dataset. Tukey's test was used to determine differences between treatment means.

Results and discussion

Highly similar responses were recorded at all four sites and years, and so all data was combined for presentation. Table 1 shows that within N rate, the three fertilizer types had similar pre-grazing herbage yield. A significant difference between N fertilizer rates (P<0.001) was observed, the mean difference being 242 kg DM ha^{-1}. In terms of overall herbage production, fertilizer type had an effect (P=0.004) as CAN and urea + NBPT had greater herbage production than urea (+424 kg DM ha^{-1}). Urea + NBPT and CAN were not significantly different in terms of herbage production. Similarly, herbage production was significantly different (P<0.001) between N fertilizer rates, as the 250 N ha^{-1} rate delivered a mean of +21.6 kg DM ha^{-1} for each additional 1 kg N ha^{-1} applied.

All fertilizers gave a similar DM response for the first 150 kg N ha^{-1} applied (CAN, 21.6; urea + NBPT, 22.5; urea, 19.4 kg DM ha^{-1}) as the 0 N plots yielded 9,113 kg DM ha^{-1}. This DM response continued with the use of the additional 100 kg N ha^{-1}, applied (CAN, 22.3; urea + NBPT, 21.2; urea, 21.3 kg DM ha^{-1}). No significant yield differences occurred between the three fertilizer types at any individual rotation and no clear trends were found that could indicate any underlying effects (Figure 1).

Table 1. Effect of nitrogen fertilizer type and rate on herbage production.[1]

Fertilizer N rate	250 kg N ha^{-1}			150 kg N ha^{-1}			SE	N type	N rate
Fertilizer N type	CAN	Urea + NBPT	Urea	CAN	Urea + NBPT	Urea			
Pre-grazing yield (kg DM ha^{-1})	1,605	1,606	1,552	1,364	1,353	1,320	25.5	0.091	<0.001
Herbage production (kg DM ha^{-1})	14,592	14,601	14,151	12,364	12,484	12,023	148.1	0.004	<0.001

[1]All data are means of four sites over 2-3 years (total = 10 site-years). S.E. = standard error.

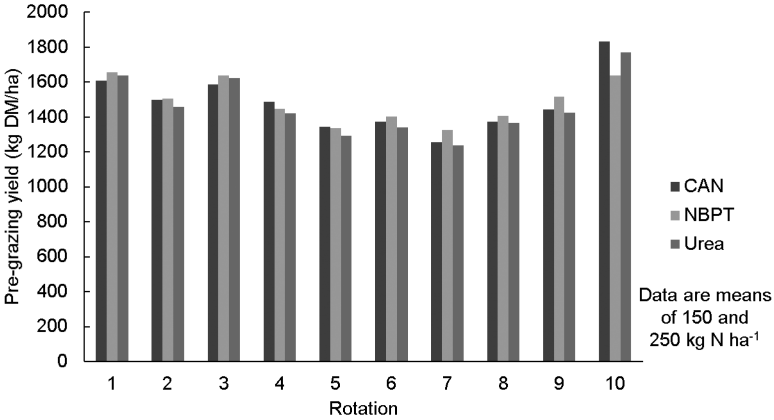

Figure 1. Seasonal responses in pre-grazing herbage yield to nitrogen fertilizer type.

Conclusions

There was an overall benefit from using protected versus unprotected urea. This could imply that more of the N is available for the plant to use. Similar herbage production was observed for CAN and urea + NBPT under grazing conditions, at all sites and providing further evidence of its efficacy, similar to Forrestal *et al.* (2017). However, the hypothesis has to be rejected as herbage production was lower for urea, although biologically these differences are small. Given these findings from multiple sites and years, farmers and industry should have confidence to use urea + NBPT without impacting herbage production.

References

Department of the Environment, Climate and Communications (2021) Climate Action Plan 2021. Available at: https://assets.gov.ie/78961/6cfd0527-085e-4420-9529-215aa303644b.pdf

Finneran E., Crosson P., O'Kiely P., Shalloo L., Forristal P.D. and Wallace M. (2012) Economic modelling of an integrated grazed and conserved perennial ryegrass forage production system. *Grass and Forage Science* 67(2), 162-176.

Forrestal P.J., Harty M., Carolan R. *et al.* (2016) Ammonia emissions from urea, stabilized urea and calcium ammonium nitrate: insights into loss abatement in temperate grassland. *Soil Use and Management* 32, 92-100.

Forrestal P.J., Harty M., Carolan R. *et al.* (2017) Can the agronomic performance of urea equal calcium ammonium nitrate across nitrogen rates in temperate grassland? *Soil Use and Management* 33, 243-251.

Krol D.J., Forrestal P.J., Wall D., Lanigan G.J., Sanz-Gomez J. and Richards K.G. (2020) Nitrogen fertilizers with urease inhibitors reduce nitrous oxide and ammonia losses, while retaining yield in temperate grassland. *Science of the Total Environment 725*, 138329.

Peters T., Taube F., Kluβ C., Reinsch T., Loges R. and Fenger F. (2021) How does nitrogen application rate affect plant functional traits and crop growth rate of perennial ryegrass-dominated permanent pastures? *Agronomy 11*, 2499.

Long-term P fertilization experiment on grass – effects on plant and soil

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Abstract

The effects of P fertilization on grass yield, grass P concentration (GPC) and soil P status (ammonium acetate extraction; P_{AAC}) were monitored in long-term field experiments. The experiments were carried out in sandy loam soils on intensively managed ley during 2003–2020 on two sites in Central Finland. The study included P0 plots, and P fertilization (average 16 kg P ha^{-1} y^{-1}), based on soil test recommendations. Dry matter yield and GPC were measured on each cut. P_{AAC} was analysed initially and after each growing season. Despite the decline of the average P_{AAC} (16-20 mg l^{-1} → 8-13 mg l^{-1}) between 2003 and 2020, no consistent yield response to P fertilization was observed even when the soil P status decreased to a level where yield response has occurred previously. The average GPC of the P treatment in the last ley rotation was lower than in the first rotation at one site, but not at the other site. Yield responses to P fertilization were rare, but the decrease of GPC indicates a gradual decrease of P supply. P fertilization of ley can be lowered, but the impact on soil P status has to be monitored. Availability of slowly soluble P reserves in soil needs further research.

Keywords: grass, phosphorus, silage, yield, soil P

Introduction

Grass production is the most important factor behind sustainable cattle farming. Appropriate use of phosphorus (P) plays a large role in improving the profitability and reducing the environmental impacts of farming. In the last 25 years, P fertilization has been reduced and the concentration of easily soluble P has declined in many regions in Finland, being currently 11 mg P_{AAC} l^{-1} in cattle production areas (Ylivainio *et al.*, 2014). At the same time, concern about depletion of available soil P and the sufficiency of P content in silage for animal health has increased. Recent results of P fertilization experiments on ley have shown no constant yield response (Kykkänen *et al.*, 2018), even when the soil P status has decreased to the level where a response was expected based on earlier experiments (Valkama *et al.*, 2009). The aim of this study is to compare the long-term effects of recommended and P0 fertilization on grass yield and soil P status and to identify needs for future studies.

Material and methods

The experiment with four replicates and seven P fertilization treatments as a randomized complete block design was established in 2003 on Site 1 (Maaninka, 63°08' N, 27°19' E, sandy loam) and Site 2 (Ruukki, 64°44'N, 25°15'E, sandy loam) in Finland. This paper includes two treatments: a mineral P application according to the Agri-environmental Scheme limits (PF) and a control with no added P (P0) covering four ley rotations from year 2003 to 2020. At four- or five-year intervals, the ley was established with whole crop barley (*Hordeum vulgare* L.; years 2003, 2007, 2012 and 2017) as a cover crop for a mixture of timothy (*Phleum pratense* L.) and meadow fescue (*Festuca pratensis* Huds.). The PF applied in the beginning of each growing season averaged at 16 (8-28) kg P ha^{-1}. Other nutrients were provided as recommended for both treatments. Soil P status ($P_{AAC,}$ mg l^{-1}) in the 0-20 cm layer was measured at the beginning of the study and at the end of each growing season by ammonium acetate extraction, pH 4.65 (Vuorinen and Mäkitie, 1955). The leys were harvested two or three times annually, except the year of establishment. Dry matter yield (DMY; kg dry matter (DM) ha^{-1}) was measured and the grass

P concentration (GPC; g kg^{-1} DM) was determined. The cumulative DMY (Mg DM ha^{-1}), P yield (kg ha^{-1}) and P balance (kg ha^{-1}) from 2003-2020 were calculated, including the whole crop years (cut height 6 cm). The GPC is presented as the average of the first grass cut of a year for each ley rotation. Statistical analyses were calculated using ANOVA (SAS 9.4., *Mixed*-procedure).

Results and discussion

The annual average DMY of grass at Site 1 for P0 was 9,000 and for PF 9,110 kg ha^{-1} while at Site 2 yields were P0 9,590 kg ha^{-1} and PF 9,680 kg ha^{-1}. There was no significant difference between the cumulative DMY of P0 and PF (Figure 1A). At Site 1, the DMY of PF was significantly higher (10-18%) only in the whole crop (barley) years 2007, 2012 and 2017. At Site 2, grass yield of PF was 5% higher in 2009 and 7% in 2018, but no difference in yield of barley was observed in the whole crop years. In contrast to Valkama *et al.* (2009), a stronger yield response of cereals compared to grasses was not consistently detected.

A GPC≤1.0 g DM kg^{-1} indicates a severe P shortage for grass. In this study, the critical P concentration was not reached, indicated by the absence of difference in DMY between P0 and PF. In the P0 treatment, the average GPC of the first cut decreased from the 1st rotation (2004-2006) to the last one (2018-2020) in both sites: From 3.1 to 2.1 g kg^{-1} in Site 1 (P<0.001) and from 2.7 to 2.4 g kg^{-1} in Site 2 (P=0.001). In the PF treatment, the decrease was observed only at Site 1 (from 3.3 to 2.3 g kg^{-1}, P<0.001), indicating at the beginning of the experiment, higher capacity of the soil to provide P for grass requirements at Site 1 than at Site 2. At Site 2, GPC of PF was 2.8 g kg^{-1} in both first and last rotations. A difference in GPC between the treatments is likely caused by P fertilization and decrease of soil P_{AAC}. As in an earlier experiment (Kykkänen *et al.*, 2018), the GPC was higher when P fertilization was used, but the effect varied by site. Soil properties (structure, organic matter content and the size and composition of slowly released P pool, etc.) may affect GPC. It is important to clarify the role of soil organic matter content and nitrogen supply on GPC and yield when determining the critical P concentration of grass (Belanger *et al.*, 2017).

Due to decreased GPC, the cumulative P yield of P0 was significantly lower compared to PF (Figure 1B). The average P yield of grass per year was 21 and 25 kg ha^{-1} in P0 and 23 and 27 kg ha^{-1} in PF at Site 1 and Site 2, respectively. The cumulative P balance of P0 treatment was highly negative at both sites (Figure 1C). The P fertilization increased the balance of PF, but it was still negative at both sites. Highly negative P balances of P0 treatment indicate that perennial grasses can utilise soil P reserves more efficiently that was expected based on previous results (Belanger *et al.*, 2017; Valkama *et al.*, 2009).

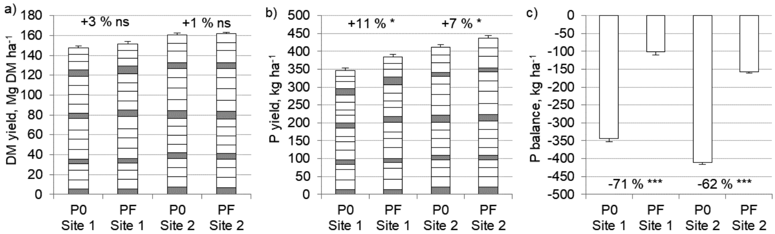

Figure 1. (A) Cumulative dry matter yield (Mg DM ha^{-1}), (B) P yield (kg ha^{-1}) and (C) P balance (kg ha^{-1}) of P0 and PF treatments at Site 1 (Maaninka) and Site 2 (Ruukki) in 2003-2020. Cumulative P fertilization 2003-2020 was 0 kg P for P0 and 285 kg P for PF. Establishment years (whole crop barley; 2003, 2007, 2012 and 2017) are highlighted in grey. Percentages above the bars are differences between P0 and PF. Statistical significances: *** P<0.001, * P<0.05, ns = non-significant. Error bars show the standard error of means (SEM).

The negative P balances caused a decrease of soil P_{AAC} at both sites and both treatments. In the spring 2003, P_{AAC} was 19.5 and 14.8 mg l^{-1} at Site 1 and 2, respectively. At Site 1, P_{AAC} declined to 9.3 in P0 and to 13.0 mg l^{-1} in PF in 2020. At Site 2, the decline was from 14.8 to 8.4 in P0 and to 10.4 mg l^{-1} in PF in 2020. In both sites, the P_{AAC} of P0 declined below 10 mg l^{-1}, where the yield response has occurred in earlier studies (Valkama *et al.*, 2009). However, P_{AAC} indicates the P intensity in soil solution rather than the amount of available P reserves (Valkama *et al.* 2016). According Belanger *et al.* (2017), the lack of response to P fertilization in low P concentrations may indicate a need to revise the interpretation of soil P test method for forage grasses.

Conclusions

According to the results of this study, some lowering of the P fertilization recommendations for perennial grasses appears to be possible without incurring yield losses, even down to a soil P_{AAC} of 8 mg l^{-1}, which is currently considered suboptimal for ley. However, the availability of slowly soluble soil P reserves to grasses requires further study. In the future, revision of the soil P test classification in Finland may also be necessary.

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References

Belanger G., Ziadia N., Lajeunesse J., Jouany C., Virkajarvi P., Sinaj S. and Nyiraneza J. (2017) Shoot growth and phosphorus-nitrogen relationship of grassland swards in response to mineral phosphorus fertilization. *Field Crops Rese*arch 204, 31-41.

Kykkänen S., Hartikainen M., Hyrkäs M., VirkajärviP., Toivakka M. and Kauppila R. (2018) Forage grasses effectively use soil P-pools in low-PAAC soils in Finland. *Grassland Science in Europe* 23, 271-273.

Valkama E., Uusitalo R., Ylivainio K., Virkajärvi P. and Turtola E. (2009) Phosphorus fertilization: A meta-analysis of 80 years of research in Finland. *Agriculture, Ecosystems & Environment* 130, 3-4.

Valkama E., Virkajärvi P., Uusitalo R., Ylivainio K. and Turtola E. (2016) Meta-analysis of grass ley response to phosphorus fertilization in Finland. *Grass and Forage Science* 71, 36-53.

Vuorinen J. and Mäkitie O. (1955). The method of soil testing in use in Finland. *Agrogeological Publications* 63, 1-44.

Ylivainio K., Sarvi M., Lemola R., Uusitalo R. and Turtola E. (2014) Regional P stocks in soil and in animal manure as compared to P requirement of plants in Finland: Baltic Forum for Innovative Technologies for Sustainable Manure Management. WP4 Standardisation of manure types with focus on phosphorus. MTT report 124.

Deployment of models to predict compressed sward height at a large scale: results and feedback

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Abstract

There is currently high interest in integrating data linked to remote sensing and methods from the machine-learning domain to develop tools to support pasture management. In this context, over the past two years, we have published models predicting the available compressed sward height (CSH) in pastures using Sentinel-1, Sentinel-2, and meteorological data. These scalable models could provide the basis of a decision support system (DSS) available for Walloon farmers. A platform performing the CSH prediction was developed and this paper aims to provide some insights in its prediction capabilities and tackle the challenge of using data acquired at different moments in time. Predictions were made from the beginning of January until the end of October 2021 using our most promising published models. After data cleaning, the coefficient of variation of CSH predictions, calculated for each studied date (n=35) and parcel (n=192,862), ranged from 0 to 986. This extreme variation suggests some prediction imperfections. Before the integration of the platform in a DSS, the main task to solve is the issue of missing or non-operational S1 or S2 data. Indeed, even if a gap filling method was applied, only 62% of potentially exploitable dates were usable.

Keywords: machine learning; decision support system; dairy cows; grazing management; pasture

Introduction

Recently published papers (Shalloo *et al.,* 2018, 2021) underline the 'work in progress' nature of the integration of data linked to remote sensing and methods from the machine learning (ML) domain in the ecosystem of tools available for managing pastures. Our team developed ML models predicting the compressed sward height (CSH) available on pastures from satellite and meteorological data (Nickmilder *et al.*, 2021). To bridge the gap between these research models and their potential use by farmers, a platform performing the prediction was implemented to be the data provider for a decision support system (DSS). This paper aims to provide some insights in the prediction capabilities of this platform and tackle the challenge of using data acquired at different moments in the season.

Material and methods

The models implemented in the prediction platform were the three best performing ones (i.e. a cubist, a neural network perceptron (nnet) and a random forest (rf)) based on a previous work done by Nickmilder *et al.* (2021) with a RMSE of CSH estimated from an independent validation around 20 mm. Those models use meteorological, Sentinel-1 (S1), and Sentinel-2 (S2) data to predict CSH. The workflow of the prediction platform is the following. First, the platform acquires daily the newly available data and launches the pre-processing when needed. Then, it performs a spatial standardization, realizes the prediction process, and finally makes some post-processing if needed. The data treatment was similar to Nickmilder *et al.* (2021). The S1, S2 and meteorological data were acquired in a form that covered the whole area of Wallonia (the southern part of Belgium) and thus all its 194,657 parcels of agricultural area with pastures and was collected from mid-January 2021 to the end of October 2021. The meteorological data came from the Agromet platform (CRA-W, 2021) and were aggregated on a daily basis. The S1 data were acquired from the European Space Agency. The S2 data were acquired from the Theia platform (Theia, 2021) under the form of level-2A products. All these datasets were resampled on a raster grid with 10 m resolution. Each parcel was thus constituted of pixels, and each time there was S1 and/or S2 data in the pixel it was considered as a record. Some filtering on S2 tiles was made: removal of a tile with too many missing values/ saturated pixels or too much cloud cover (75% threshold each time).

To deal with missing acquisition, a gap filling method was applied. For every day of the year, a check was made to confirm the availability of S2 data. If it was confirmed, the date was considered as usable (UD). All the pixels were filled with available data at this UD and the incomplete pixels were filled with data acquired the day before, and so on until 4 days before the UD. Thus, a dataset for one UD is in fact a composite dataset gathering S1 and S2 data up to 4 days before the actual UD. To assess the relevance of the CSH predictions and the reliability of the prediction platform scaling local models to a greater scale (i.e. entire Walloon Region), we have studied: the occurrence of concurring data acquisition, the raw values, the presence of outliers, and the descriptive statistics for each date and parcel.

Results and discussion

Theoretically, the S2 satellites have a revisit frequency over Wallonia of 3 to 5 days. Considering the worst-case scenario of 5 days, we should have at least 58 dates (i.e. 290/5 days) usable for the period studied. However, even with the application of a gap filling methodology, only 35 dates (62%) had enough S2 data of sufficient quality to be further processed and 25 of these dates covered the grazing season (April – October). Without the gap filling application, only 17 dates would have been considered (29%). On these 35 dates, 99% (192,866) of the parcels were represented at least once and the total number of records was 201,875,534. Unfortunately, there was a huge number of non-usable S2 data. This might be due to a combination of edgy position of pixels relatively to the satellite orbits, poor weather and out of range/missing input values. These values cannot deliver reliable information. Therefore, a post-processing filter was applied after the prediction step to remove all the non-finite values. This decreased the predicted set to 92,782,075 records. The data distribution of those predictions per model is summarized in Table 1. Some values (less than 2%) were out of the range of expected CSH values (i.e. [0 mm; 250 mm]). After deletion, the dataset was composed of 92,757,937 records. Given that a parcel is composed of several pixels, therefore, the estimation of the coefficient of variation of CSH is a measure of its CSH heterogeneity. The cubist CSH predictions gave the highest variability, the nnet and rf models were more stable although the trends of higher CSH were asynchronous as shown in Figure 1. This meant that the models used extracted different part of the information in the dataset and a combination of this information must be accounted in the future. The observed null values were due to the presence of only one pixel (Table 1). To visualize the evolution of CSH throughout the year, the average CSH was calculated for each date (Figure 1). As expected, due to the response of plants to increasing temperatures, we observed a slight increase during the spring (April – June) and then a decrease. However, the sparse data acquisition due to the poor weather conditions during the summer blurred the trends. Another point underlined in Figure 1 is the sensitivity of the models to cloudiness: for example, the 11th February was cloudy with very thin clouds that were not detected as such, and thus decreasing the quality of the predictions for this specific date.

Conclusions

From a technological point of view, the platform is operational and now usable to predict on a daily-routine basis the CSH in Wallonia. However, given the low proportion of exactly concurring data, we had to implement a time lag tolerance in the platform for its future use in a DSS. This means that for

Table 1. Data and coefficient of variation distribution of CSH predictions (in mm) obtained by the three tested models on the 192,866 parcels, delivering 92,782,075 records with some extreme associated predictions on the 35 dates of acquisition.

	Raw CSH predictions (n=92,782,075)			Coefficient of variation per parcel and date (n=92,757,937)		
	Cubist	Nnet	Rf	Cubist	Nnet	Rf
Minimum	5.39	45.26	25.89	0.00	0.00	0.00
1%	31.58	45.26	37.25	1.42	0.00	1.22
1st quartile	47.83	56.23	53.00	6.50	0.00	5.65
Median	56.68	56.23	60.09	10.21	9.19	9.09
Mean	62.41	64.74	64.39	12.03	11.40	10.46
3rd quartile	70.832	62.08	70.92	15.26	17.78	13.85
99%	145.86	130.64	130.36	41.89	42.18	30.91
Maximum	331.88	130.64	203.15	906.95	68.65	74.62

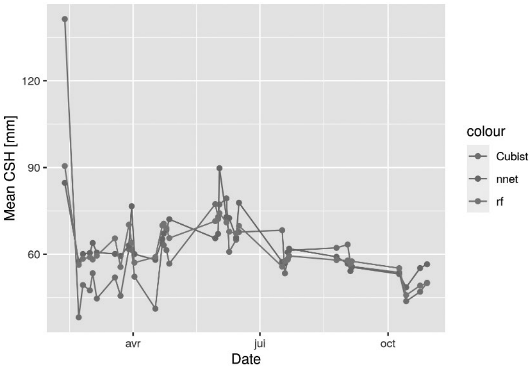

Figure 1. Representation of the mean of the parcel's mean height over each acquisition date.

each S2 acquisition date, predicted datasets were completed with data going back up to four days. This methodology managed to decrease the impact of non-concurring data in a context of predicting with all the datasets but there is still work to do. Indeed, only 62% of dates were exploitable. Hence, the models still need to be improved given the occurrence of those quite extreme values.

Acknowledgements

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References

CRA-W (2021) Agromet.be. Available at: https://agromet.be/

Nickmilder C., Tedde A., Dufrasne I., Lessire F., Tychon B., Curnel Y., Bindelle J. and Soyeurt H. (2021) Development of machine learning models to predict compressed sward height in Walloon pastures based on Sentinel-1, Sentinel-2 and meteorological data using multiple data transformations. *Remote Sensing* 13, 408. https://doi.org/10.3390/rs13030408

Shalloo L., O'Donovan M., Leso L., Werner J., Ruelle E., Geoghegan A., Delaby L. and O'Leary N. (2018) Review: Grass-based dairy systems, data and precision technologies. *Animal* 12 (s2): S262-S271. https://doi.org/10.1017/S175173111800246X

Shalloo L., Byrne T., Leso L. *et al.* (2021) A review of precision technologies in pasture-based dairying systems. *Irish Journal of Agricultural and Food Research*. https://doi.org/10.15212/ijafr-2020-0119

Theia (2021) Available at: https://www.theia-land.fr/

Nitrogen use efficiency and carbon footprint of an agroecological dairy system based on diversified resources

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Abstract

A mixed crop-dairy system named OasYs, based on temporary grasslands and on the grazing of diversified forage resources, was implemented in 2013 (Lusignan, France) in a plain area affected by summer droughts. Conducted without irrigation and with limited inputs, this agroecological system aims to permit farmers to live from their dairy system in a context of climate change, while preserving environment and contributing to animal welfare. We present the nitrogen (N) balance and N-use efficiency, and the carbon (C) footprint of this system assessed by the CAP'2ER tool over 2017-2020. OasYs showed small average N surpluses (47 kg N ha^{-1} UAA) and good N-use efficiency (49%). Greenhouse gases (GHG) gross emissions expressed in kg CO_2-equivalent per litre of corrected milk decreased from 2017 (0.90) to 2020 (0.78). They were lower compared to other grassland-based systems from lowland areas. These results can be explained by the sharply reduced use of N fertilizer and concentrates counterbalanced by a large use of legumes and a new breeding strategy. The net C footprint of milk is, however, higher than the average of grassland-based systems from lowland areas, because of lower soil C storage resulting from the ploughing of temporary grasslands.

Keywords: mixed crop-dairy system, agroecology, grazing, legumes, OasYs

Introduction

In order to face the current challenges of dairy farming in north-west Europe, a mixed crop-dairy system was designed to permit farmers to live from their dairy system in a context of climate change, while preserving the environment and contributing to animal welfare.

This cattle system named OasYs (Novak *et al.*, 2018) is based on the principles of agroeocology (Dumont *et al.*, 2014) and it relies on the grazing of diversified forage resources, comprising mainly multispecies temporary grasslands but also annual crops. The originality of this system is to test several innovations both on the forage system (e.g. grazing of annual crops and fodder trees, large use of legumes) and on the livestock system (e.g. three-way crossbreeding, extension of lactation length), and to integrate them into a consistent way at the farm level. The whole farming system was redesigned to address together the challenges of adaptation and mitigation to climate change while allowing for the delivering of other ecosystem services. We hypothesize that a greater diversity of the dairy system's components and of their functions will both improve the resilience of the overall system against climatic hazards, and permit high production levels and environmental performance. This paper presents the N balance, greenhouse gas (GHG) emissions and net C footprint of this innovative system, assessed by the CAP'2ER tool over the 2017-2020 period.

Materials and methods

The OasYs dairy cattle system has been in place experimentally since June 2013 in a plain area traditionally affected by summer droughts located south of Brittany (Lusignan, Nouvelle Aquitaine, France), in an INRAE facility. The forage system (91.5 ha) produces the fodder necessary to feed the dairy cattle herd (72 milking cows, and replacement heifers) without irrigation and with limited use of mineral N fertilizer (<50 kg N ha^{-1} $year^{-1}$ on grassland) and of pesticides (<10% of regional references). Forage resources are diversified in terms of species, cultivars, age and management, and priority is given to their grazing

(Novak *et al.*, 2018). Five- and four-year multispecies and legume-rich grasslands (52 ha) represent the heart of the forage system, complemented by annual crops (maize, grain sorghum, cereal-legume mixtures). Calving periods are centred on grassland peak productions (spring and autumn). The extension of lactation length to 16 months and the three-way crossing of dairy breeds (Holstein, Scandinavian Red, and Jersey) are used to improve reproduction performance and cow lifetime (Novak *et al.*, 2018). The CAP'2ER tool, based on Life Cycle Assessment following the IPCC methodology (https://cap2er.fr/Cap2er/), was used to determine GHG gross emissions, the C footprint of the milk, the N balance, and associated indicators over the 2017-2020 period. Results were compared to national references, as regional references were not available for similar systems (i.e. lowland systems with less than 30% maize).

Results and discussion

The N balance showed small surpluses (48 kg N ha^{-1} on average) and varied between 27 and 69 kg N ha^{-1} mainly according to the legume N fixation amounts, which represented 52 to 72% of total N inputs (Table 1). N-use efficiency at the farm scale varied between 37 and 62% and was also closely linked to the amount of legumes present each year on cropland. These results are quite good compared with other dairy cattle systems from lowland areas of France, based either on grassland or on grassland and maize (Table 1) or to other literature data (Hutchings *et al.*, 2020). These results can be explained by the smaller amounts of mineral N fertilizer and concentrates used in OasYs, resulting in high protein feed self-sufficiency (94% on average).

GHG gross emissions expressed in kg CO_2 equivalent (CO_2e) per litre of fat and protein corrected milk (FPCM) decreased from 2017 (0.90) to 2020 (0.78). They were lower compared with other dairy cattle systems from French lowland areas (Table 2) or those of many other countries (Mazzetto *et al.*, 2021). As for N surplus, these results could be explained by the sharply reduced use of mineral N fertilizer and concentrates counterbalanced by a large use of legumes, and certainly also by the new livestock breeding strategy. When the soil C storage is taken into account, the net C footprint of milk (0.78 kg CO_2e litre^{-1} FPCM on average) is higher than the average of French grassland-based systems from lowland areas (0.67) but it remains lower than other systems using more maize. OasYs is indeed based on temporary grasslands that store less C in soils than permanent ones, and the C stored in the above-ground biomass of trees is not yet taken into account by CAP'2ER.

Table 1. N balance and N-use efficiency of OasYs compared with other French dairy systems.

	OasYs				Inosys data on 2009-2013 (Foray *et al.*, 2017)	
	2017	2018	2019	2020	Grassland-based lowland systems	Grassland and maize lowland systems
Forage (kg N ha^{-1})	0	0	4	0	10	6
Concentrates (kg N ha^{-1})	9	3	7	9	37	43
Mineral N fertilizer (kg ha^{-1})	25	15	15	6	45	65
Symbiotically-fixed N (kg N ha^{-1})	47	73	74	47	18	19
Total N inputs (kg N ha^{-1})	91	101	110	72	120	145
Total N outputs (kg N ha^{-1})	52	44	41	45	40	49
N surplus (kg N ha^{-1})	39	57	69	27	80	96
N-use efficiency (%)	57	44	37	62	33	34

Table 2. C footprint of OasYs compared with other French dairy systems.[1]

	OasYs				CAP'2ER data on 2013-2019 (Goumand & Castellan, 2021)	
	2017	2018	2019	2020	Grassland-based lowland systems	Grassland and maize lowland systems
Proportion of grasslands in UAA	67%	64%	64%	65%	80%	58%
FPCM (litres cow^{-1})	6,624	6,468	6,915	6,978	5,507	7,187
Concentrates (g $litre^{-1}$)	42	28	54	63	130	161
GHG gross emissions (kg CO_2e $litre^{-1}$ FPCM)	0.9	0.83	0.83	0.78	0.99	0.96
Soil C storage (kg CO_2e $litre^{-1}$ FPCM)	0.05	0.06	0.06	0.05	0.32	0.15
GHG net emissions (kg CO_2e $litre^{-1}$ FPCM)	0.85	0.78	0.77	0.73	0.67	0.82

[1] UAA = utilized agricultural area; FPCM = fat and protein corrected milk; GHG = greenhouse gases.

Conclusions

The innovative agroecological dairy system OasYs, based on diversified and legumes-rich forages resources and a new livestock breeding strategy, showed good results either on the N balance or on the GHG emissions per litre of FPCM. Its net C footprint could be further improved by extending the duration of grasslands, e.g. through relay cropping. It was certainly overestimated by the CAP'2ER tool, which currently does not take into account the above-ground C stored by agroforestry trees. Redesigning dairy systems by integrating coherently the forage and livestock systems through an agroecological approach seems to be a promising strategy to reduce their environmental burden.

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References

Dumont B., Gonzalez-Garcia E., Thomas M., Fortun-Lamothe L., Ducrot C., Dourmad J.Y. and Tichit M. (2014) Forty research issues for the redesign of animal production systems in the 21st century. *Animal* 8, 1382-1393.

Foray S., Billy C., Manneville V., Dollé J.B., Le Gall A., Vertes F. and Godinot O. (2017) Gestion de l'azote dans les systèmes d'élevage herbivores. Available at: https://idele.fr/en/detail-article/gestion-de-lazote-dans-les-systemes-delevage-herbivores.

Goumand E. and Castellan E. (2021) Fiches références CAP2'ER. Available at: https://idele.fr/en/detail-dossier/fiches-references-cap2err-edition-2021.

Hutchings N. J., Sørensen P., Cordovil C.M.D.S., Leip A. and Amon B. (2020) Measures to increase the nitrogen use efficiency of European agricultural production. *Global Food Security* 26, 100381.

Mazzetto A., Falconer S. and Ledgard S. (2021) Mapping the carbon footprint of milk for dairy cows. Report for DairyNZ. Available at: https://ucanr.edu/sites/programteams/files/347129.pdf.

Novak S., Chargelègue F. and Emile J.C. (2018) An agroecological dairy cattle system to graze all year round. *Grassland Science in Europe* 23, 473-475.

A comparison of once-a-day compared to twice-a-day milking in late lactation

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Abstract

In seasonal calving pasture-based systems of production, grass growth slows in the autumn/winter months, coinciding with late lactation. Altering milking frequency from twice-a-day (TAD) to once-a-day (OAD) can help reduce demand for pasture while also providing a better work-life balance for the farmer. The objective of this experiment was to examine the effect of milking OAD for either eleven or seven weeks from the end of lactation compared to milking TAD for the entire lactation. Results showed an immediate reduction of 19% in milk production and up to 12% reduction in kg fat + kg protein (milk solids yield) with OAD milking compared to TAD. Milking cows OAD increased somatic cell score (SCS) immediately, on average SCS was 12% higher when cows were milked OAD compared to TAD in late lactation. This suggests OAD milking for the last number of weeks of lactation is a viable option but somatic cell count needs to be low before switching to OAD milking.

Keywords: once-a-day milking, dairy cows, late lactation, milk production

Introduction

Seasonal calving pasture-based systems of milk production offer advantages, such as the ability to match herd feed demand with grass supply. Compact calving is a cornerstone of grass-based systems but it can provide challenges at peak labour periods, such as at calving, increasing demands on calf-care, as well as cow care and feeding and other farm duties (slurry and fertilizer spreading) (Deming *et al.*, 2018).

While twice-a-day (TAD) milking is accepted as the standard milking frequency, more dairy farmers are using once-a-day (OAD) milking at different periods in the lactation, particularly at times such as late lactation, where there is an opportunity to reduce workload and improve work-life balance. There are, however, potential drawbacks of OAD milking: these include a reduction in milk production (Davis *et al.*, 1999; McNamara *et al.*, 2008; Patton *et al.*, 2006) and increased somatic cell count (SCC) (Clark *et al.*, 2006). The objective of this experiment was to examine the effect of milking OAD for either eleven or seven weeks from the end of lactation compared to milking TAD for the entire lactation.

Materials and methods

The study was carried out from September 25 to December 11, 2020. Prior to the start of the experiment, 51 dairy cows (12 primiparous and 39 multiparous) were balanced based on calving date, milk yield, yield of fat plus protein (milk solids yield; MSY), bodyweight (BW) and body condition score (BCS) during the two-weeks preceding the experiment. Cows were assigned to one of three milking-frequency treatments in a randomized block design. The three treatments were: (1) TAD milking for the entire lactation (TAD_F); (2) TAD milking until 11-weeks from the end of lactation, cows were milked OAD for the last 11-weeks of lactation (OAD11); (3) TAD milking until 7-weeks from the end of lactation, for the last 7-weeks of lactation cows were milked OAD (OAD7). Milking times were 0700 and 1600 when cows were milked TAD; OAD milking was at 07:00. Fresh pasture was offered on a 24-hour basis after morning milking. Pre- and post-grazing sward heights were measured daily using a rising plate meter. Herbage mass (HM; >4 cm) was measured twice weekly. Cows grazed in two separate herds; cows milked TAD were in one herd and cows milked OAD in another. As cows changed from TAD to OAD milking they also changed grazing herds. Herds grazed adjacent to each other to ensure similar HM and grass quality was offered. Milk yield was recorded daily; milk composition and somatic cell count (SCC) was measured weekly. The SCC data were log transformed to create somatic cell score (SCS). Data were analysed using covariate analysis and mixed models in SAS v9.4. Terms for parity, treatment and week of experiment were included. Pre-experimental values and days in milk were used as covariates in the model.

Results and discussion

Average pre-grazing yield was 1,515 kg dry matter (DM) ha^{-1} while pre-grazing heights were similar between treatments (10.0 cm). Post-grazing height of the herds milked OAD and TAD were 4.6 and 4.4 cm, respectively (P<0.001). Concentrate input was 2 kg DM cow^{-1} day^{-1}. Eleven weeks from the end of lactation OAD milking was implemented for the OAD11 herd. It caused an immediate reduction in production. Average milk yield during the first four weeks of OAD milking for the OAD11 herd was 19% lower than cows milked TAD (16.3 kg cow^{-1} day^{-1}; Table 1) while MSY was reduced by 12% compared to TAD cows (1.47 kg cow^{-1} day^{-1}). When the OAD7 cows were transitioned to OAD milking seven weeks from the end of lactation they also had a 19% reduction in milk production and a 10% reduction in MSY compared to the TAD_F herd (14.5 and 1.4 kg cow^{-1} day^{-1}, respectively). Stelwagen and Knight (1997) reported that the relative production loss is significantly less in late lactation compared with early lactation, as milk volumes are considerably lower in late lactation. Kennedy *et al.* (2021) reported a 21 and 20% reduction in milk and MSY, respectively when milking OAD during the first four weeks of early lactation compared to TAD milking. During weeks five to eight of OAD milking the magnitude of the reduction increased as the OAD11 cows were producing 26% less milk and 20% less MSY than the TAD_F cows (14.5 and 1.36 kg cow^{-1} day^{-1}, respectively) and the OAD6 produced 31 and 20% less than the TAD_F, respectively (12.8 and 1.2 kg cow^{-1} day^{-1}, respectively).

When cumulative milk production for the final 11-weeks of lactation was considered all treatments were significantly different to each other (P<0.001; 978, 824 and 1,124 kg cow^{-1} for OAD7, OAD11 and TAD_F, respectively). Cumulative MSY for the last 11-weeks was similar for OAD7 and TAD_F (102 kg cow^{-1}), but OAD11 cows produced significantly less (P<0.01; 79 kg cow^{-1}) than all other treatments.

Milking cows OAD increased SCS, the increase was immediate once changed to OAD (Figure 1). On average, SCS during the last 11 weeks of lactation was different (P<0.001) between the three treatments, OAD11 had the highest SCS (2.15), TAD_F had the lowest (1.99) and OAD6 was intermediate (1.84). On average SCS was 12% higher when cows were milked OAD compared to TAD in late lactation.

Table 1. Effect of milking cows once-a-day from either seven or eleven weeks.

	OAD7	OAD11	TAD_F	Standard error	Signifance
Milk yield					
Weeks 1 to 4 (kg day^{-1})	16.5^a	13.2^b	16.2^a	0.50	0.001
Weeks 5 to 8 (kg day^{-1})	11.8^a	10.7^a	14.5^b	0.75	0.001
Weeks 9 to 11 (kg day^{-1})	8.9^a	8.1^a	12.8^b	0.69	0.001
kg fat + kg protein (MSY)					
Weeks 1 to 4 (kg day^{-1})	1.5^a	1.3^b	1.5^a	0.04	0.010
Weeks 5 to 8 (kg day^{-1})	1.2^a	1.1^a	1.4^b	0.07	0.010
Weeks 9 to 11 (kg day^{-1})	1.0^a	0.8^a	1.2^b	0.09	0.001

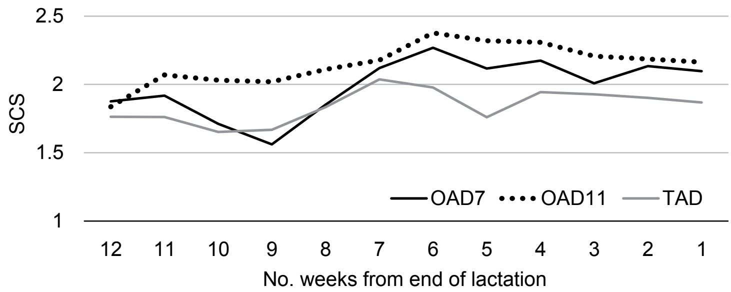

Figure 1. Effect of milking frequency in late lactation on SCS.

Conclusions

The results of this study show that OAD milking in late lactation reduces milk production, milking OAD for seven compared to 11-weeks at the end of lactation will reduce milk production losses. Milking OAD increases SCS, hence it is advisable to have a low SCC before commencing OAD milking.

References

Clark D.A., Phyn C.V.C., Tong M.J., Collis S.J. and Dalley D.E. (2006) A systems comparison of once- versus twice-daily milking of pastured dairy cows. *Journal of Dairy Science* 89(5), 1854-1862.

Davis S.R., Farr V.C. and Stelwagen K. (1999) Regulation of yield loss and milk composition during once-daily milking: a review. *Livestock Production Science* 59(1), 77-94.

Deming J., Gleeson D., O'Dwyer T., Kinsella J. and O'Brien B. (2018) Measuring labor input on pasture-based dairy farms using a smartphone. *Journal of Dairy Science* 101(10), 9527-9543.

Kennedy E., Murphy J.P., Delaby L. and O'Donovan M. (2021) Effects of short-term once-a-day milking in early lactation on dairy cow performance when managed in a seasonal-calving pasture based system. *Livestock Science* 251, 104648.

McNamara S., Murphy J.J., O'Mara F.P., Rath M. and Mee J.F. (2008) Effect of milking frequency in early lactation on energy metabolism, milk production and reproductive performance of dairy cows. *Livestock Science* 117(1), 70-78.

Patton J., Kenny D.A., Mee J.F., O'Mara F.P., Wathes D.C., Cook M. and Murphy J.J. (2006) Effect of milking frequency and diet on milk production, energy balance, and reproduction in dairy cows. *Journal of Dairy Science* 89(5), 1478-1487.

Stelwagen K. and Knight C.H. (1997) Effect of unilateral once or twice daily milking of cows on milk yield and udder characteristics in early and late lactation. *Journal of Dairy Research* 64(4), 487-494.

Performance of two rising plate meters in assisting grazing management in semi-natural grassland

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Abstract

Rising plate meters (RPM) are well established tools for pasture management to assess compressed sward height (CSH). Management of extensively grazed semi-natural grasslands for conservation purposes can be defined by target CSH. New and technically advanced RPMs like the Grasshopper (GH) need evaluation of CSH measurement performance in comparison with established models (Castle-RPM), as management depends on target CSHs defined by the latter. The GH and Castle-RPM were compared in the long-term grazing experiment 'Forbioben' with three stocking rates (medium, lenient, very lenient) in a paired sampling approach over two years. The CSH obtained by the GH was predicted by the CSH obtained from the Castle-RPM using regression analysis. The CSH from GH underestimated the CSH from the Castle-RPM and showed a horizontally asymptotic behaviour with increasing sward height – this can be explained by technological differences between RPMs and constraints of the GH in tall swards. The underestimation by GH was affected by stocking rate. The differences in measurement showed that the GH is currently not suitable for extensive pastures with higher swards under lenient grazing. Additionally, target sward heights need to be redefined when changing to different measurement technologies in long-running management schemes.

Keywords: extensive grazing, rising plate meter, compressed sward height

Introduction

Grazing management in extensive low-input grassland aims to combine agronomic and conservation purposes. This can be achieved by adapting the stocking rate according to pre-defined target sward heights. The sward heights are measured regularly using rising plate meters (RPMs) which allows the monitoring of grazing pressure. The RPM technique provided by Castle (1976) is a simple and established instrument but technologically outdated. More recent models like the Grasshopper (North Shore Technologies, Shannon, Ireland) record measurements automatically and provide higher accuracy in compressed sward height (CSH) readings (McSweeney *et al.,* 2019). Adaptation of technological advancements can be challenging in long-term experiments or grazing systems set at target sward heights as the management targets need to be accomplished irrespective of differences in technology. For instance, the weight:area ratio of the plate of the Grasshopper (GH)-RPM is 0.49 g m^{-2} compared to 0.28 g m^{-2} for the Castle-RPM, which will affect the CSH and, consequently, the management decision. Particularly in extensively managed pastures, swards can be mature and thus stemmy and CSH could therefore be misleading. The deviation of the RPMs from each other therefore probably depends on the long-term grazing intensity which determines the extent of heterogeneity (Adler *et al.,* 2001). Consequently, target sward heights in long-running management schemes need to be redefined when changing to different RPM technologies.

Material and methods

The experiment was conducted in the long-term cattle grazing experiment 'Forbioben' located in the Solling Uplands in Germany during 2019 and 2020. The design of the experiment is a one factorial randomized block design with three replications. It compares three different cattle stocking intensities defined by target CSH as the experimental factor: Moderate (M; target sward height of 6 cm), lenient

(L; 12 cm) and very lenient stocking (VL; 18 cm) on nine 1-ha paddocks (Tonn *et al.,* 2019). Sward height was measured approximately bi-weekly with the Castle-RPM during the grazing season taking fifty measurements per paddock. To assess the deviation of the GH-RPM from the Castle-RPM the sward measurements were conducted spatially as close as possible in a paired procedure on thirteen occasions resulting in 5829 records. The maximum measuring height of the GH-RPM is 22.3 cm due to technical restrictions, whereas the maximum measuring height of the Castle-RPM is technically not limited. Therefore, a model assuming an asymptotic behaviour to predict the CSH of the GH-RPM from Castle-RPM measurements was generated with the software R 4.1.2. To account for this technical prerequisite and to achieve valid linearity, a non-linear mixed effects model was used. The dependent variable CSH of the GH-RPM was predicted by the CSH of the Castle-RPM with the equation $Y = a + (b - a)e^{-ecx}$ with a representing the maximum attainable value for Y (CSH of GH-RPM), b the initial value of Y and c is proportional to the relative increase of Y with increasing X (CSH of the Castle-RPM) representing the logarithmic rate constant. Additionally, a fixed effect of the categorical factor stocking rate treatment was included, which allowed variation of the parameter c. The parameters a and b were constant as those are determined by the construction of the RPM. A random structure was included to allow the variation of c for the effects of the block design and the temporal nesting of the sampling. The model was fitted with the package 'nlme' (Pinheiro *et al.,* 2021). The differences of the predicted values from target CSH for the grazing treatments M, L and VL were tested with a one-sample Wilcoxon test (α=0.05).

Results and discussion

The maximum 1% of CSHs measured with the Castle-RPM were 17.0, 23.5 and 27.0 cm for the stocking rate treatments M, L and VL. Corresponding values for measurements conducted with the GH-RPM were 15.5, 20.4 and 21.2 cm, respectively. Including the fixed term for grazing treatment for parameter c affected and improved the model significantly as indicated by the marginal Wald test and AICc (P<0.0001, $AICc\ \Delta$ = -33.56, $df\ \Delta$ = 2). The coefficient of the parameters a and b were 29.68 and -0.22. For parameter c the coefficients were -3.38, -3.29 and -3.25, for the stocking intensity treatments M, L and VL, respectively, with a RMSE of 2.23 cm. Model predictions at the target CSH for the grazing intensities M, L and VL were significantly different from the target CSH defined with the Castle-RPM. Additionally, the differences between the RPMs for the three grazing intensities were not equal: predicted mean CSH at the targets CSHs for M, L and VL were 5.7, 10.4 and 14.2 cm, respectively. Therefore, the deviation from the target CSHs determined for the Castle-RPM were -0.3 cm for M, -1.6 cm for L and, -3.8 cm for VL (all P<0.0001) (Figure 1). The slope <1 and the decreasing slope with increasing CSH of the Castle-RPM indicate that the CSH obtained by the GH-RPM underestimates the CSH measured with the Castle-RPM with increasing magnitude in taller swards, i.e. with more extensive stocking (Figure 1). The underestimation can be explained by a heavier specific disc weight of the GH-RPM and a greater compaction compared with the Castle-RPM (Bransby *et al.*, 1977). The increasing magnitude of underestimation by the GH-RPM can be explained by the technical limitation of measuring sward heights >22.3 cm. Additionally, the measurement accuracy of the GH-RPM is only validated up to a CSH of 17.8 cm (McSweeney *et al.,* 2019). Furthermore, high CSH values in extensive pastures arise from tall patches, which deviate from short ones in botanical composition, phenology (Correll *et al.*, 2003) and resistance against a pressure plate. The differences in parameter c for the different stocking intensity treatments indicate an effect of sward heterogeneity with different proportions of tall and short patches (Tonn *et al.,* 2019).

Conclusions

For the monitoring of extensive grazing operations, the Grasshopper rising plate meter in its current state is not sufficient. Measurements of tall swards, as common in low-input extensive pastures are not possible due to technical restrictions in taller grass swards. If different RPMs are used for long-term monitoring based on target sward heights these need to be adjusted.

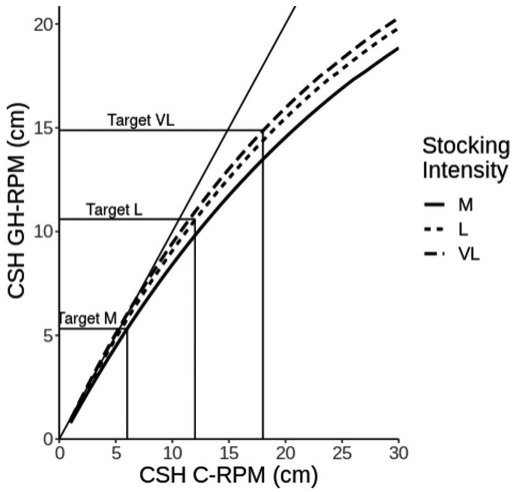

Figure 1. Non-linear relationship between the CSH measurements of the Castle- and GH-RPM for the stocking intensity treatments M, L and VL. Target CSHs of the respective stocking treatments are marked correspondingly for the respective RPM tool.

References

Adler P., Raff D. and Lauenroth W. (2001) The effect of grazing on the spatial heterogeneity of vegetation. *Oecologia* 128(4), 465-479.

Bransby D.I., Matches A.G. and Krause G.F. (1977) Disk meter for rapid estimation of herbage yield in grazing trials. *Agronomy Journal* 69, 393-396.

Castle M.E. (1976) A simple disc instrument for estimating herbage yield. *Grass and Forage Science* 31, 37-40.

Correll O., Isselstein J. and Pavlu V. (2003) Studying spatial and temporal dynamics of sward structure at low stocking densities: the use of an extended rising-plate-meter method. *Grass and Forage Science* 58, 450-454.

McSweeney D., Coughlan N.E., Cuthbert R.N., Halton P. and Ivanov S. (2019) Micro-sonic sensor technology enables enhanced grass height measurement by a rising plate meter. *Information Processing in Agriculture* 6, 279-284.

Pinheiro J., Bates D., DebRoy S., Sarkar D. and R Core Team. (2021) nlme: Linear and Nonlinear Mixed Effects Models. Available at: https://CRAN.R-project.org/package=nlme

Tonn B., Raab C. and Isselstein J. (2019) Sward patterns created by patch grazing are stable over more than a decade. *Grass and Forage Science* 74, 104-114.

Fresh and conserved herbage in cows' diet improves milk fatty acids and antioxidants profile

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Abstract

With the aim of studying the content of fatty acids (FA) and liposoluble antioxidants (AOX: vitamins A and E, xanthophylls and carotenoids) in the different dairy milk production systems in Galicia (NW Spain), 70 representative farms were visited quarterly during the years 2014 to 2016. Systems range from grazing, including organic and conventional, to animal-confined farms which base the cows' diet on conserved forages (grass and maize) and concentrates. A total of 317 bulk-tank milk samples of known dietary provenance were analysed for FA and AO by chromatography methods. It was confirmed that the feeding system modifies the FA and AOX concentrations in milk, showing a more favourable profile for systems based on fresh and conserved herbage, particularly on the organic farms.

Keywords: grazing, feeding systems, fatty acids, antioxidants

Introduction

Almost half of the total cow's milk produced in Spain comes from the Galician (humid-temperate, Atlantic NW Spain) region, where 6,500 holdings produce 2.9 million tonnes of milk per year. Most Galician dairy farms follow an intensive system of production, based on all-year confined cows and total mixed rations (TMR) composed of maize and home-made grass silages and purchased concentrates (Flores *et al.*, 2017). It is estimated that one out of ten litres of milk produced in this region come from grazing systems and that only one out of ten litres of grazing milk come from organic farms. The increasing cost of production of milk is putting at risk the predominant dairy system, and now more farmers are considering a strategic return to grazing as a way to assure the farm profitability. In addition, there is growing interest among consumers about the differential presence of bioactive compounds in milk from grazing cows, making this product more attractive as a part of so-called healthy diets (e.g. Dewhurst *et al.*, 2006). The objective of the present work is to compare the presence of both fatty acids (FA) and liposoluble antioxidants (AOX) in the different Galician dairy systems.

Materials and methods

A sample of 70 commercial farms was chosen as representative of the most common feeding systems in the Atlantic humid-temperate dairy production area of Galicia. Farms were grouped according the existence of cows' grazing in organic (GO) or conventional (GC) systems, or tied systems with TMR feeding based on herbage silage (HS), grass and maize silage (HMS) or maize silage (MS). During the years 2014 to 2016 each farm was visited quarterly each year in order to know the diet composition fed to dairy cows and to take samples of feed ingredients and bulk-tank milk. Milk samples were immediately frozen (-20 °C) until posterior chromatographic analysis (FID-GC for FA and HPLC for AOX), following the routines established by the official inter-professional laboratory of milk analysis of the region (Laboratorio Interprofesional Galego de Análise do Leite, LIGAL). A total of 317 valid observations were available, of which FA and AOX composition, as well as diet composition, was known. An ANOVA analysis was performed on FA and AOX milk content using the group (fixed effect) as the class variable. Data were analysed using SAS package (SAS Institute, 2009).

Results

The main results obtained in the study are summarized in Table 1. Grazing farms were of smaller size and with less productive cows, and used less concentrate per litre of milk. Average grazing time was higher in organic grazing compared with conventional grazing farms. There was a positive relationship between the proportion of maize silage in the daily cows' diet and the average milk yield per cow, herd size and the efficiency of conversion (not shown in tables) of dry matter (DM) into milk. Organic farms showed a higher proportion of fresh grass in the average daily DM ration compared with conventional grazing farms. The proportion forage:concentrate was approximately 85:15 and 75:25 in GO and GS farms and approximately 62:38 in the silage-based farms. This is evidence that even for the more intensive Galician farms, home-grown forage makes a substantial contribution to the cows' ration.

The milk FA profile is affected by the feed system, confirming the results reported by other authors (e.g. Larsen *et al.*, 2012). Milk from grazing farms showed, on average, a healthier FA profile compared with all-silage farms, with higher contents of alpha-linolenic and *c9t11* conjugated linoleic acid (CLA) and a lower of omega-6:omega-3 FA ratio. Within grazing farms, the milk produced on organic farms showed a significantly better profile compared with conventional grazing farms. It is not clear if the higher proportion of fresh grass in the diet of organic farms is the main reason of these differences, since the variability observed in the botanical composition of pastures on organic farms can play an important role in explaining these differences.

There were marked differences between the average concentrations of milk AOX among farm types. Whilst the differences in retinol were comparatively reduced, the concentration of alpha-tocopherol in milk was positively related to the importance of herbage (fresh or ensiled) in cows' diet, whilst the minority isomer gamma-tocopherol followed an inverse trend. In a similar fashion, the highest concentrations of xanthophylls and carotenes were found in the milk of organic farms, followed by conventional grazing farms and all-silage grass farms. These results agree with the results of other authors that report the existence of a relationship between grazing and the increased concentration of both polyunsaturated FA in milk and liposoluble antioxidants (e.g. Bloksma *et al.*, 2008) with potential benefits for human health.

Conclusions

A higher presence of herbage (fresh or conserved as silage) in the dairy cows' ration produces a higher milk-fat content of bioactive FA, tocopherols and carotenoids compared with that from maize-silage based farms. Milk from grazing farms shows a better profile than milk from all-silage-herbage farms. Milk from organic farms shows a healthier composition, from a human diet perspective, than that of the other dairy cow feeding systems analysed.

Acknowledgements

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Table 1. Milk production, diet composition, milk fatty acid profile and milk concentration in liposoluble antioxidants by farm type.[1,2]

	GO	GC	HS	HMS	MS	*P*-value
n	44	67	84	59	63	
Milk yield[3] (kg cow^{-1} d^{-1})	19.9[e]	25.3[d]	27.2[c]	30.9[c]	33.1[a]	***
Herd size (dairy cows' number)	36.5[c]	37.0[c]	38.0[c]	56.3[b]	91.0[a]	***
Grazing time (hours d^{-1})	8.0[a]	6.7[b]	0.8[c]	0.0[c]	0.0[c]	***
Concentrate use (g kg^{-1} milk)	146[c]	215[b]	308[a]	292[a]	282[a]	***
Diet composition (g kg^{-1} total DM)						
Fresh grass	394[a]	343[a]	95[b]	0[c]	0[c]	***
Maize silage	48[d]	100[c]	65[d]	323[b]	466[a]	***
Herbage silage	266[b]	225[b]	398[a]	268[b]	104[c]	***
Dry forages	145[a]	76[b]	65[b]	26[c]	41[bc]	***
Concentrates	148[c]	255[b]	377[a]	381[a]	383[a]	***
FA profile (g kg^{-1} total FA)						
Trans-vaccenic (C18:1*t11*)	24[a]	18[b]	14[c]	9.7[d]	11.3[d]	***
Linoleic (C18:2*c9c12* n6)	15.8[d]	17.2[c]	20.1[b]	19.6[b]	21.8[a]	***
Alpha-linolenic (C18:3 *c9c12c15* n3)	8.7[a]	6.2[b]	5.2[c]	3.6[e]	4.3[d]	***
Conjugated Linoleic Acid (CLA) *c9t11*	11.8[a]	10.3[b]	8.9[c]	6.5[d]	7.7[cd]	***
Omega-6 total	18.6[c]	19.7[c]	22.2[b]	21.8[b]	24.1[a]	***
Omega-3 total	11.0[a]	8.2[b]	7.3[c]	5.9[d]	6.8[c]	***
FA ratios						
Omega6: Omega3	1.79[d]	2.62[c]	3.34[b]	3.90[a]	3.70[a]	***
(*t11:t10*) C18:1	11.4[a]	6.87[b]	4.70[c]	3.09[d]	1.91[d]	***
Vitamins (µg g^{-1} fat)						
Vit. A (retinol)	10.4[c]	12.9[a]	13.4[a]	12.1[ab]	11.2[bc]	***
Vit. E (α-tocoferol)	27.6[a]	24.1[b]	22.3[b]	19.0[c]	16.3[d]	***
Vit. E (γ-tocoferol)	0.63[d]	0.92[c]	1.18[b]	1.21[b]	1.69[a]	***
Xanthophylls (µg g^{-1} fat)						
Lutein	0.47[a]	0.32[b]	0.22[c]	0.18[cd]	0.15[d]	***
Zeaxanthin	0.05[a]	0.04[b]	0.02[c]	0.02[c]	0.02[d]	***
β-Criptoxanthin	0.06[a]	0.04[b]	0.03[c]	0.03[c]	0.03[d]	*
Carotenes (µg g^{-1} fat)						
(*All-t-β*) carotene	5.90[a]	5.10[b]	4.28[c]	3.27[d]	2.48[e]	***
(*9+ 13 cis β)* carotene	0.19[a]	0.14[b]	0.10[c]	0.07[d]	0.06[d]	***

[1] GO = organic grazing; GC = conventional grazing; HS = herbage silage; HMS = herbage and maize silages; MS = maize silage.

[2] Values in the same line under different superscript are significantly (P<0.05) different. * P<0.05; ** P<0.01; *** P<0.001.

[3] 40 g kg^{-1} fat corrected milk

References

Bloksma J., Adriaansen-Tennekes R., Machteld H., Lucy P.L., Vijver D.V., Baars T. and Wit D.J. (2008) Comparison of organic and conventional raw milk quality in the Netherlands. *Biological Agriculture and Horticulture* 26, 69-83.

Dewhurst R.J., Shingfield K.J., Lee M.A. and Scollan N.D. (2006) Increasing the concentrations of beneficial polyunsaturated fatty acids in milk produced by dairy cows in high-forage systems. *Animal Feed Science and Technology* 131, 168-206.

Flores G., Martínez-Fernández A., Doltra J., García A. and Eguinoa P. (2017) *Encuesta sobre estructura y sistemas de alimentación de las explotaciones lecheras de Galicia, Cornisa Cantábrica y Navarra*. Informe del Proyecto INIA-RTA2012-00065-C05. Available at: http://ciam.gal/pdf/informeinia.pdf.

Larsen M., Fretté X., Kristensen T., Eriksen J., Søegaard K. and Nielsen J. (2012) Fatty acid, tocopherol and carotenoid content in herbage and milk affected by sward composition and season of grazing. *Journal of the Science of Food and Agriculture* 92(14), 2891-2898.

SAS Institute (2009) *SAS/Stat User's Guide, v.9.2*. SAS Institute Inc., Cary, NC, USA.

Combining remote sensing data and the BASGRA model to predict grass yield at high latitudes

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Abstract

The impact of weather, soil and management on yield and nutritive value of grassland can be evaluated using process-based simulation models. These models may be calibrated using data on biomass, leaf area and other characteristics acquired from drones, hand-held devices, and satellites. The objective of this study was to compare the prediction accuracy of the BASGRA model calibrated with grassland data from Northern Norway obtained in 2016 and 2017. The data were acquired either from: (1) ground registrations; or (2) a hand-held spectrometer and satellites. Data on crude protein and fibre content from NIRS analyses were used in both calibrations. Daily air temperature, precipitation, relative air humidity, wind speed and solar radiation that were input to the BASGRA simulations were taken from The Norwegian Meteorological Institute and The Agrometeorology Norway network. Information about soil texture, cutting regime and N fertilization was obtained from farmers and advisers. The differences between simulated and observed biomass, and crude protein and fibre content were similar after the two calibrations. Observed crude protein and fibre content were simulated with a higher accuracy than biomass for both types of calibration data.

Keywords: nutritive value, prediction, processed-based model, Sentinel, satellite, UAV

Introduction

Grass forage is the main feed component for meat and dairy production at high latitudes. The impact of weather, soil, and management on grasslands can be evaluated using process-based simulation models (Kipling *et al.*, 2016). Data on biomass, leaf area and other grassland characteristics acquired from remote devices such as drones, hand-held devices and satellites are increasingly used to calibrate such models (Kasampalis *et al.*, 2018). The BASGRA model, which simulates forage grass as a function of weather, soil and management (Höglind *et al.*, 2020) has been used extensively to simulate grassland yield (Korhonen *et al.*, 2018) and nutritive value (Persson *et al.*, 2019) at high latitudes. Using remote sensing data for calibration of the BASGRA model could potentially improve its simulation accuracy for grasslands whose characteristics are difficult to assess by ground registrations. The objective of this study was to compare the prediction accuracy of the yield and nutritive value of grasslands in northern Norway using the BASGRA model calibrated against different combinations of ground registrations and remote sensing data.

Materials and methods

Data on grassland yields from 27 grassland fields in Northern Norway acquired from a hand-held FieldSpec3 spectrometer, the Sentinel 2 satellite or ground registrations were used to calibrate the BASGRA model. Ground registrations were carried out either by cutting, drying and weighing the above-ground biomass in 12-15 quadrats of 0.25 m^2 per field, or by counting silage bales per field and weighing three randomly selected bales per field. The simulation of the Sentinel-2 data was done by means of binning the 2151 spectral points acquired with the FieldSpec3 spectrometer based on the spectral range for each Sentinel-2 band (Ancin-Murguzur *et al.*, 2019). In addition, time series parameters were calculated from Sentinel-2 vegetation index time series from start to peak growing season. Both FieldSpec and Sentinel-2 data sets were then used to develop a partial least squares regression model for estimating yield. Two calibrations of the BASGRA model were carried out against: (1) the ground registration data; and (2) the remote sensing biomass data. In both calibrations, crude protein and fibre (neutral detergent fibre, NDF) content data from NIRS analyses were used in addition. The calibration procedures followed Bayesian techniques (Van Oijen *et al.*, 2005) according to a protocol based on previous calibrations of the BASGRA model (Höglind *et al.*, 2020; Persson *et al.*, 2019) using a chain length of 350,000 iterations. Minimum and maximum air temperature, precipitation, relative humidity, wind speed and global radiation input data were obtained from weather stations within the networks of The Norwegian Meteorological Institute and The Agrometeorology Norway network. Soil characteristics and cutting frequency data were obtained from farmers or advisory service organizations. Nitrogen fertilizer data were obtained from either the same sources or, in cases where field-specific data were missing, assumed to represent normal regional practices. Ground observations and data from the NIRS analyses from five randomly selected fields were excluded from the calibration and used for model validation. The prediction accuracy of above-ground dry matter, crude protein and NDF was evaluated using the root mean squared error (RMSE), normalized by dividing it by the mean of the observations, and the relative mean bias error rMBE, as a value of the relative under- or overpredictions (Table 1).

Results and discussion

The differences between simulated and observed biomass, and crude protein and fibre content were similar after the two calibrations. The observed nutritive value components were simulated with a higher accuracy than the observed above ground dry matter after both calibrations. This is not very surprising since the former varied less than the latter (data not shown) across fields from different locations and botanical composition. The biomass simulation accuracy was lower than simulations based on calibrations against ground data of monoculture timothy grass trials (Korhonen *et al.*, 2018; Persson *et al.*, 2019), whereas the crude protein and NDF simulations were in the same range as these. Further comparisons of calibrations using remote sensing data and ground registrations could provide more information about how these registration methods could be used to improve the simulation accuracy and usefulness of the BASGRA model. Grassland fields in Northern Norway are often botanically heterogeneous and this heterogeneity also varies considerably between fields. The comparisons with previous model evaluations indicate that model calibrations based on botanical composition could be one way to generate biomass predictions with higher accuracy. The higher prediction accuracy of the nutritive value components is not very surprising since the former varied less than the latter (data not shown) across fields from different locations and botanical composition.

Table 1. Root mean squared error (RMSE), normalized RMSE, relative mean bias error (rMBE) and Willmott's index of agreement (d-index) of the above ground biomass, crude protein (CP) and Neutral Detergent Fibre (NDF) content in the validation dataset.

	Number of observations	Mean of observation	Mean of simulation	RMSE	Normalized RMSE	rMBE	d-index
Ground data calibration							
Dry matter (g m^{-2})	5	607	414	343	0.57	-0.32	0.18
CP (g g^{-1} DM)		0.128	0.120	0.021	0.16	-0.055	0.91
NDF (g g^{-1} DM)		0.545	0.635	0.12	0.22	0.16	0.46
Remote sensing data calibration							
Dry matter (g m^{-2})	5	607	413	338	0.56	-0.32	0.31
CP (g g^{-1} DM)		0.128	0.124	0.031	0.24	-0.010	0.83
NDF (g g^{-1} DM)		0.545	0.610	0.10	0.19	0.14	0.47

Conclusions

The differences between simulated and observed biomass, and crude protein and fibre content were similar after the two calibrations. The simulation accuracy was higher for the nutritive value components than for the above ground dry matter after both calibrations.

Acknowledgements

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References

Ancin-Murguzur F.J., Taff, G., Davids, C., Tømmervik, H., Mølmann, J. and Jørgensen, M (2019) Yield estimates by a two-step approach using hyperspectral methods in grasslands at high latitudes. *Remote Sensing* 11, 400.

Höglind M., Cameron D., Persson T., Huang X. and van Oijen M (2020) BASGRA_N: A model for grassland productivity, quality and greenhouse gas balance. *Ecological Modelling* 417, 108925.

Kasampalis D.A., Alexandridis T.K., Deva C., Challinor A., Moshou D. and Zalidis G. (2018) Contribution of remote sensing on crop models: A Review. *Journal of Imaging* 4, 52.

Kipling R.P., Virkajärvi P., Breitsameter L., Curnel Y., De Swaef T., Gustavsson A.-M., Hennart S., Höglind M., Järvenranta K., Minet J., Nendel C., Persson T., Picon-Cochar C., Rolinski, S., Sandars D.L., Scollan N.D., Sebek L., Seddaiu G., Topp C.F.E., Twardy S., Van Middelkoop J., Wu L. and Bellocchi G (2016) Key challenges and priorities for modelling European grasslands under climate change. *Science of the Total Environment* 566-567, 851-864.

Korhonen P., Palosuo T., Persson T., Höglind M., Jégo G., Van Oijen M., Gustavsson A.-M., Bélanger G. and Virkajärvi P (2018) Modelling grass yields in northern climates – a comparison of three growth models for timothy. *Field Crops Research* 224, 37-47.

Persson T., Höglind M., Van Oijen M., Korhonen P., Palosuo T., Jégo G., Virkajärvi P., Bélanger G. and Gustavsson A.M (2019) Simulation of timothy nutritive value: A comparison of three process-based models. *Field Crops Research* 231, 81-92.

Van Oijen M., Rougier J. and Smith R (2005) Bayesian calibration of process-based forest models: bridging the gap between models and data. *Tree Physiology* 25, 915-927.

Annual course of dietary cation-anion difference (DCAD) on drained fen grassland

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Abstract

Dietary Cation Anion Difference (DCAD) is used for optimization of dairy cattle diets with respect to their health and performance. Particularly on our fen sites, grass silage DCAD varies between years and over the different cuts from +400 to -300. We conducted three K fertilization RCB trials, A with 3, B with 4, and C with 5 cuts per year, adjacently positioned on a drained fen grassland site in 2017-2019. In all years, the annual draining period started in spring causing decreasing ground-water tables (GWT) until the start of autumn. However, in 2017, heavy rainfall flooded the site at end of June, followed by a high GWT during summer. Based on the results of the 0 K treatment, the course of DCAD in the grass was described. During all growing periods, the DCAD decreased with decreasing GWT rapidly from spring to the start of summer. When GWT increased again (in 2018 and 2019 at start of autumn and in 2017 with a flooding event in June) the DCAD also increased. At correlation coefficients of 0.66, 0.63 and 0.83 for the trials A, B and C respectively, the DCAD values corresponded very well with the mean GWT during a three-week period before each cut.

Keywords: dairy cow, ground water table

Introduction

The concept of managing the dietary cation-anion difference (DCAD) for improved health and performance of dairy cows has been well-established for more than 30 years. A lower DCAD in the diet fed during the prepartum period improves calcium status and decreases risk of hypocalcaemia during the immediate postpartum period. Increasing the DCAD of diets fed during lactation increased milk yield and dry matter intake (Overton, 2020). Compared to other feed, DCAD in grass silages of our region varied to a larger extent. DCAD in grass silages of the first cut were higher than in the later cuts (Boss and Pickert, 2021). There was a particular variation in grass silages produced on fen soils. In the different grass silages over a year on a milking cow farm, we found DCAD ranging between -338 and +447 (Pickert *et al.,* 2021). The objective of this study was to describe the annual course of DCAD on fen grassland in northeast Germany, depending on the hydrological boundary conditions. For the experiment we selected a typical drained fen grassland site in Brandenburg, Germany (Paulinenaue, 52°68'N, 12°72'E; 28.5-29.5m a.s.l.; mean annual temperature 9.2 °C, mean annual precipitation 534 mm).

Materials and methods

Grass samples were taken from three K fertilizer trials A, B and C, designed as RCB experiments with four replications and conducted from 2017 to 2019. The mixed sward was composed of *Festuca arundinacea, Festuca pratensis, Festuca rubra, Lolium perenne, Phleum pratense, Poa pratensis, Agrostis capillaris* and *Trifolium repens*, but dominated by *F. arundinacea*. The three field trials were adjacently positioned on the site. In order to vary the dates of sampling over the season, Experiment A was harvested with five, B with four and C with three cuts per year. In this study, only the results of the 0 K plots were evaluated. Weather data were taken from ZALF Paulinenaue Research Station. GWT was measured weekly in a measuring tube on the experimental site. For calculation of DCAD, the content of K, Na and S were analysed according to DIN EN ISO 11885 and Cl according to DIN 38405-D1-2. DCAD was calculated on the basis of the Na, K, S and Cl contents in the grass dry matter (DM) (g kg^{-1} DM) with DCAD = (Na × 43.5 + K × 25.6) – (Cl- × 28.2 + S × 62.4) and given as meq kg^{-1} DM.

Results and discussion

In all three trials, the higher the GWT the higher the DCAD in the cut grass. The correlation coefficients to DCAD were higher when referred to the GWT over a period of three weeks before harvest (Table 1).

On the fen grassland, the ground-water was retained in the site over the winter period resulting in a GWT of between 30 and 0 cm distance from the surface. In early spring, the grassland site was drained and GWT was lowered from 40 to 80 cm. From May onwards, the discharge stopped and the water was retained in the site again. During this period the resulting GWT depended on the actual rainfall during summer.

Figure 1 exemplarily describes the groundwater situation during the experimental period in a wet summer (2017, Figure 1A) and a dry summer (2018, Figure 1B) as well as the course of DCAD depending on the summer rainfall. GWT started at a high level in spring and decreased over summer. When GWT increased, as in summer 2017, DCAD values also increased. In all years the course of DCAD followed the course of GWT. In spring, at the beginning of the harvesting period with the 1^{st} and 2^{nd} cut, the relationship of DCAD and GWT was stricter than during the summer months.

Conclusions

In our experiments, GWT seemed to be very useful to indicate higher or lower DCAD of different cuts on fen grassland. Depending on GWT, grass of the fields concerned could be sampled and analysed for DCAD and selected for feeding to certain animal groups or herds depending on the DCAD value. Further measurements are necessary in order to validate the findings of the trials at the Paulinenaue grassland on other sites. A further question arises: is it really just the GWT, or does the GWT in conjunction with the geogenic situation influence the DCAD? It is conceivable that the stratigraphic conditions are influenced by prehistoric salt zones. This question should also be clarified in further investigations.

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Table 1. Correlation coefficients of DCAD and GWT in the 3 cutting rate trials 2017-2019.

Trial No.	Cutting rate	GWT 1 week before harvest	GWT 1-2 weeks before harvest	GWT 1-3 weeks before harvest
A	5	0.55	0.60	0.66
B	4	0.60	0.62	0.63
C	3	0.82	0.83	0.83
[A – C		0.64	0.67	0.70]

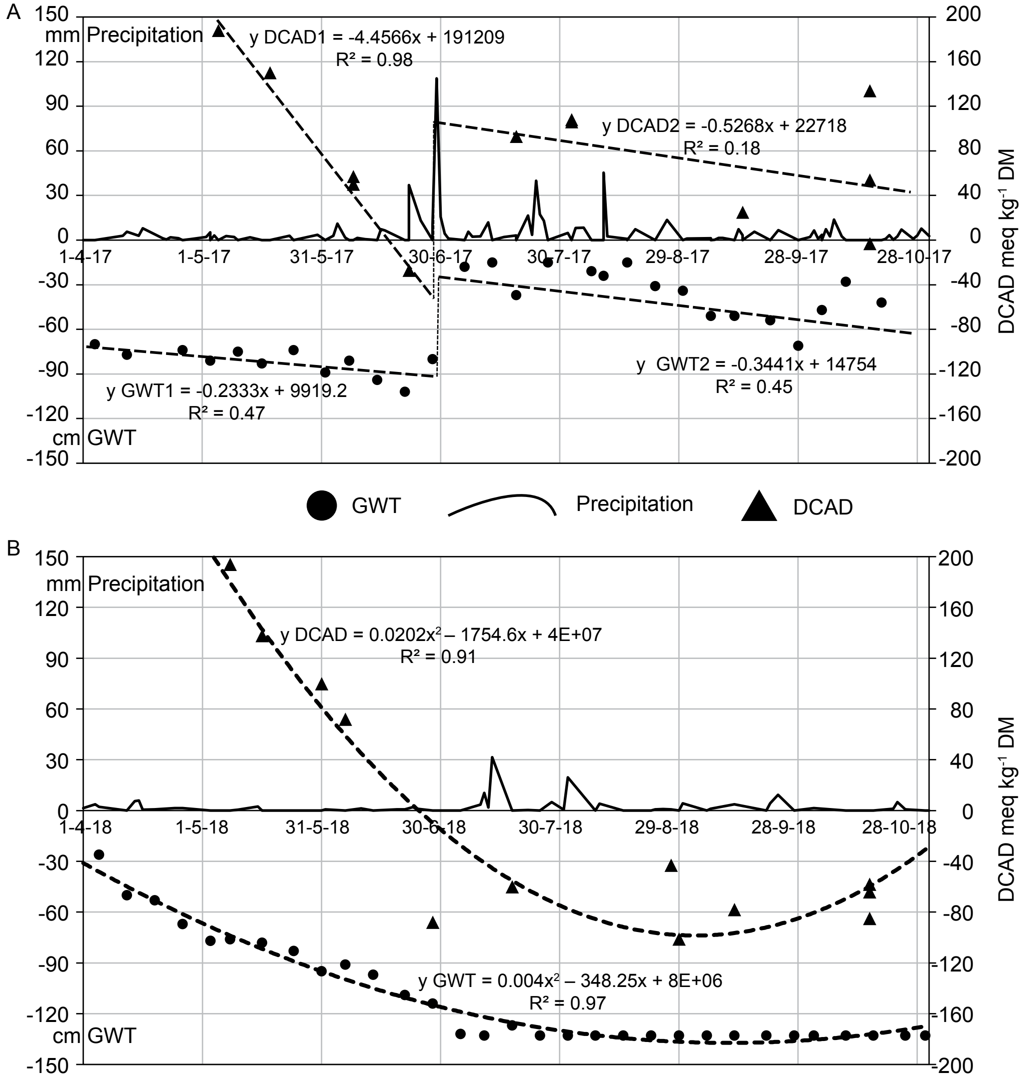

Figure 1. Course of precipitation, groundwater level GWT (week 1-3 before harvest) and dietary cation anion difference (DCAD) in different grass cuts on fen soil (Paulinenaue, Germany) in (A) 2017 with a wet summer and (B) 2018 with a dry summer.

References

Boss B. and Pickert J. (2021) Sind Säuren und Basen im Gleichgewicht? (Are acids and bases in balance?) *Bauernzeitung Berlin* 44, 42-44.

Overton T.R. (2020) Update on DCAD for dry and lactating cows. Proceedings Cornell Nutrition Conference. Cornell University. 82nd Meeting. October 19-22, 2020, pp. 186-191,

Pickert J., Eulenstein F., Merz C. and Behrendt A. (2021) Wasserhaushalt bestimmt Säure-Basen-Verhältnis (Water management influences acid-base balance). *Bauernzeitung Berlin* 44, 42-44.

Effects of two approaches for outdoor access on the welfare of lactating Nordic red cows

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Abstract

To determine the differences between two approaches of providing access to grasslands on the welfare of dairy cows, we enrolled cows into three treatments: partial outdoor access to pasture: (1) with sufficient forage for grazing (pasture); (2) without grazeable forage (outdoor paddock); or (3) indoor confinement. Cows were moved to outdoor treatments for 6 h d^{-1}. A total of twenty-seven lactating primi- and multiparous Nordic red cows were housed at the University of Helsinki's Viikki research farm. Treatments were implemented in a replicated 3×3 Latin Square design with three-week periods. Dry matter intake from feed bins was higher in indoor than overall mean of outdoor treatments, and significantly higher in pasture than paddock treatment. Milk yield was higher in paddock than pasture treatment with no significant difference between indoor and overall mean of outdoor treatments. Provision of a supplemental total mixed ration may further improve the welfare of these cows by supporting greater milk production without altering overall behavioural patterns when outdoors.

Keywords: dairy cow, welfare, grazing, outdoor paddock, pasture

Introduction

Use of grasslands offers a possibility to improve the welfare of dairy cattle but successful grazing may be difficult to implement in modern large scale dairy herds. With a combination of natural living, biological function, and affective state defining welfare (Fraser *et al.*, 1997), the effect of outdoor access on welfare can be positive (Hernandez-Mendo *et al.*, 2007; Washburn *et al.*, 2002) or negative (Lean *et al.*, 2008). Providing grasslands to dairy cows allows grazing and a softer surface relative to indoor housing. However, the provision of a total mixed ration (TMR) indoors resulted in cows preferring to remain indoors instead of utilizing grassland access (Charlton *et al.*, 2011). TMR sustained higher milk yield when compared to pasture-based diets (McAuliffe *et al.*, 2016). Despite this apparent preference for TMR, grazing can be included as one of the natural behaviours described in five freedoms and prevention of natural behaviours can lead to frustration which in turn decreases animal welfare (Rutter, 2010). Approach to pasture access has conflicting consequences, which indicates the need to understand what cows value when outdoors. Our objective is to determine the differences between grazing and outdoor access on the welfare of dairy cows in automatic milking system (AMS) conditions. Our hypothesis is that cows will gain benefits from outdoor access regardless of grazing.

Materials and methods

The study was conducted at the University of Helsinki Viikki research farm in Finland during summer 2021. Twenty-seven lactating primi- (n=5, mean 220 days in milk (DIM) at the beginning of the experiment) and multiparous (n=22, mean 182 DIM) Nordic red cows were used in replicated 3×3 Latin squares with 21-d periods. The treatments were: (1) outdoor access with grazeable forages (6 h d^{-1}; pasture); (2) outdoor access with no grazeable forages (6 h d^{-1}; outdoor paddock); and (3) indoor confinement (control). When indoors, all cows had *ad libitum* access to TMR via an automatic feeding system (Insentec, Marknesse, the Netherlands) with 22 feed bins (stocking density = 200%) and milked by automatic milking system (AMS; Astronaut A3, Lely Industries NV, the Netherlands). The same

TMR was offered *ad libitum* from a mobile feed cage, moved every two days to prevent concentration of manure, with 12 feeding spaces (stocking density = 75%) in the outdoor paddock. Cows were offered supplemental concentrate from the AMS at a fixed rate throughout the experiment. Adjacent grasslands (4.5 hectares; originally sown with timothy grass, 50%; fescue-ryegrass hybrid, 15%; ryegrass, 15%; tall fescue, 10%; and meadow fescue, 10%; neutral detergent fibre range was 54.7-60.0% of dry matter) were divided into seven segments, one of them (0.4 ha) was used as the outdoor paddock after mowing and the others were used for the grazing treatment.

Milk production was measured at each milking and the data from the last week of each period was used. Using direct observation (ten-minute scan sampling), behaviour of the cows was assessed outdoors during the last week of every period for three days. Feeding time outdoors was calculated as the combination of eating TMR and attempted grazing or solely grazing, depending on the treatment. Dry matter intake (DMI) indoors was recorded automatically during each visit to a feeding bin.

One cow was removed from the data due to drying off before the end of the experiment. The data were analysed by mixed model that included fixed effects of treatment, square, and period within a square and random effect of cow within a square (SAS 9.4.). The orthogonal contrasts were used to determine the effects of: (1) outdoor treatments (pasture and paddock) vs indoor confinement; and (2) pasture vs paddock.

Results and discussion

Pasture cows tended to have longer feeding time outdoors (Table 1, P=0.08). While this difference in time may not be meaningful, the outdoor paddock cows accomplished their feeding time with a combination of TMR consumption (15%) and attempting to graze (12%). This suggests that grazing is a highly motivated behaviour. The opportunity to express natural behaviours is seen as an important way to increase animal welfare through natural living (Fraser *et al.*, 1997).

The DMI from feed bins indoors was greater in control than outdoor treatments (Table 1, P<0.001) and in pasture than paddock treatment (P<0.001). The apparent demand for TMR may come from the energy expenditure of high milk yield of modern dairy cows. In previous studies, cows have preferred to stay indoors if TMR is available only indoors (Charlton *et al.*, 2011). This follows current results of cows eating more TMR indoors if they have no access to TMR when outdoors. The provision of TMR outdoors alongside grazing allows cows to satisfy these competing behavioural motivations of consuming TMR and actively grazing.

Milk yield was not different between outdoor and indoor treatments (Table 1, P=0.812), but the milk yield was greater in paddock than pasture treatment (P=0.009). TMR promoted greater milk yield in comparison to pasture-based diets (McAuliffe *et al.*, 2016). This was evident in our study as cows on the paddock had *ad libitum* access to TMR indoors and outdoors, whereas the cows on pasture had no access to TMR outdoors.

Table 1. Effects of outdoor access on milk yield, DMI and eating behaviour.

	Treatment			SEM[1]	Significance	
	Control	Pasture	Paddock		Outdoor vs control	Pasture vs paddock
Eating behaviour, proportion of time outdoors	-	0.30	0.27[2]	0.01	-	0.08
Intake from bins, kg dry matter d^{-1}	18.8	16.6	12.8	1.01	<0.001	<0.001
Milk yield, kg d^{-1}	31.7	30.9	32.6	0.71	0.812	0.009

[1] SEM = standard error of the mean.

[2] Combination of eating TMR (15%) and attempted grazing (12%).

Conclusions

Opportunity to graze may be an important behaviour for dairy cows as evident by the substantial proportion of time outdoors dedicated to this behaviour even with the limited access to grass in presence of TMR. The provision of supplemental TMR may further improve the welfare of these cows by providing choice in diet and supporting greater biological function (milk production) without altering overall behavioural patterns when outdoors.

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References

Charlton G.L., Rutter S.M., East M. and Sinclair L.A. (2011) Preference of dairy cows: Indoor cubicle housing with access to a total mixed ration vs. access to pasture. *Applied Animal Behaviour Science* 130, 1-9.

Fraser D., Weary D.M., Pajor E.A. and Milligan B.N. (1997) A scientific conception of animal welfare that reflects ethical concerns. *Animal Welfare* 6, 174-186.

Hernandez-Mendo O.M, von Keyserlingk M.A.G., Viera D.M. and Weary D.M. (2007) Effects of pasture on lameness in dairy cows. *Journal of Dairy Science* 90, 1209-1214.

Lean I.J., Westwood C.T. and Playford M.C. (2008) Livestock disease threats associated with intensification of pastoral dairy farming. *New Zealand Veterinary Journal* 56, 261-269.

McAuliffe S., Gilliland T.J. and Hennessy D. (2016) Comparison of pasture-based feeding systems and a total mixed ration feeding system on dairy cow milk production. *Grassland Science in Europe* 23, 289-291.

Rutter S.M. (2010) Grazing preferences in sheep and cattle: implications for production, the environment and animal welfare. *Canadian Journal of Animal Science* 90, 285-293.

Washburn S.P., White S.L., Green Jr. J.T. and Benson G.A. (2002) Reproduction, mastitis, and body condition of seasonally calved Holstein and Jersey cows in confinement or pasture systems. *Journal of Dairy Science* 85, 105-111.

Grazing by red deer counteracts atmospheric nutrient deposition in semi-natural open habitats

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Abstract

Semi-natural habitats are vulnerable to eutrophication, which can result from atmospheric deposition. Maintaining habitat-specific nutrient conditions despite atmospheric inputs is a challenge for conservation. Grazing wild red deer (*Cervus elaphus*) is suggested as an alternative management measure for open habitats, but effects of red deer on nutrient dynamics have not yet been evaluated. To quantify import and export of nitrogen (N) and phosphorus (P) by red deer, we collected data on vegetation productivity, forage removal, dung quantity, and nutrient concentrations from permanently marked plots in two habitat types (European dry heaths, lowland hay meadows) on a military training area in Germany. The annual nutrient export of N and P by red deer grazing was notably higher than the nutrient import through excreta in both habitats. Net nutrient removal averaged 13.9 and 29.5 kg N ha^{-1} yr^{-1} and 1.1 and 3.3 kg P ha^{-1} yr^{-1} in heathlands and grasslands, respectively, thus exceeding estimated values for local atmospheric deposition. In consequence, red deer grazing can mitigate the effects of atmospheric nutrient deposition in semi-natural open habitats.

Keywords: atmospheric nutrient deposition, *Cervus elaphus*, Natura 2000, nitrogen, phosphorus, rewilding, wildlife grazing

Introduction

Intensification of agricultural practices and abandonment leading to succession into forest are the most important threats to semi-natural open habitats, but impacts of atmospheric emissions and air pollution have been underestimated (EEA, 2020). Substantial atmospheric deposition caused by anthropogenic activities is widespread in Europe, threatening the biodiversity of semi-natural habitats characterized by low nutrient availability. Not only atmospheric nitrogen (N) deposition (Clark *et al.*, 2017; Wilkins *et al.*, 2016) but also phosphorus (P) deposition can have detrimental ecological effects (Camarero and Catalan, 2012; Tipping *et al.*, 2014).

Herbivores take up nutrients while foraging and transport them to different locations, with P being exclusively excreted in dung and N in dung as well as in urine (Haynes and Williams, 1993). Extensive livestock grazing is an established conservation measure for open habitats and there is evidence that livestock grazing can compensate for current levels of atmospheric nutrient deposition (Uytvanck *et al.*, 2010). An alternative conservation measure, particularly suitable for large or inaccessible target areas, is grazing by wild ungulates, e.g. red deer (*Cervus elaphus*) (Riesch *et al.*, 2019), but we do not yet know how red deer affect nutrient dynamics in open habitats. Therefore, we quantified nutrient import and export by wild red deer in heathlands and grasslands hypothesizing that red deer counteract atmospheric nutrient deposition because nutrient import through excreta is low and export through grazing is high (H1). Second, we supposed that higher forage removal in grasslands results in higher nutrient export and a larger difference between nutrient export and import than in heathlands (H2).

Materials and methods

We assessed the nutrient fluxes through red deer in the Grafenwöhr military training area (GTA) in Bavaria, Germany. The area (230 km^2 in total; Natura 2000 Site DE6336301) is inhabited by a large population of wild red deer. Due to a targeted wildlife management regime by the Federal Forests Administration (Bundesforst), the red deer use the open habitats (ca. 30% of the total area (Raab *et al.*, 2019)) for foraging all year round (Richter *et al.,* 2020). In heathlands and grasslands (Natura 2000 habitat types 4,030 European dry heaths and 6,510 lowland hay meadows), we selected four sampling sites of ca. 0.5 ha (heathlands) or 1 ha (grasslands). We compiled data on vegetation productivity and quality, forage removal by red deer (Riesch *et al.*, 2019) and dung quantity (Wichelhaus, 2020) from two plots (15×15 m^2) per site over one year (sampling in April, May, June, August, October 2015, and April 2016).

We quantified the export of nutrients through grazing based on the forage removal by red deer assessed by movable exclusion cages (McNaughton *et al.*, 1996) combined with rising-plate meter measurements and calibration cuts. Hand-pluck samples were collected to determine forage nutrient concentrations. To quantify the import of nutrients, we assessed the red deer dung mass accumulated per sampling date using the faecal accumulation rate method (Mayle *et al.*, 1999). We determined N concentrations in dried plant and dung samples by Dumas combustion and analysed P concentrations after digestion with HNO_3 by ICP-OES.

We obtained the quantity of exported and imported N or P per plot for each sampling period by multiplying the forage removal and the dung quantity with the mean over nutrient concentrations at the start and end sampling date of a sampling period. To estimate total N import through both faeces and urine, we employed an equation for the ratio of urinary to faecal N excretion related to plant N concentration (Hobbs, 1996). To assess the annual rates of nutrient export and import, we summed up the mean values per sampling period and habitat and used Gaussian error propagation to obtain the associated uncertainty in R version 4.0.3.

Results and discussion

We found between 0 and 112 dung pellet groups per plot and sampling date. The annually imported dung dry mass was 155±15 kg ha^{-1} in heathlands and 97±3 kg ha^{-1} in grasslands. In line with our hypothesis (H1), the annual nutrient export by red deer exceeded the import in both habitat types (Table 1). Even when accounting for urinary N, the difference between import and export of N was negative. As hypothesized (H2), the difference between annual nutrient import and export was more pronounced in grasslands than in heathlands, which has consequences for habitat conservation in view of atmospheric nutrient deposition.

The atmospheric N deposition of 9-11 kg ha^{-1} yr^{-1} in GTA (Umweltbundesamt, 2021) could impair the studied plant communities (Wilkins *et al.*, 2016). The considerable net N export by red deer in grasslands, however, should be able to prevent detrimental N input despite current atmospheric deposition levels. In heathlands, which are more susceptible to eutrophication, at an atmospheric deposition of 11 kg N ha^{-1} yr^{-1}, a net input of ca. 6 kg N ha^{-1} yr^{-1} could be possible despite red deer grazing. Neither grassland nor

Table 1. Annual fluxes of nitrogen (N) and phosphorus (P) in kg ha^{-1} through excretion and grazing of wild red deer and resulting difference (import-export) in heathlands and grasslands.[1]

	Faecal N import	Total N import	N export	ΔN	P import	P export	ΔP
Heathlands	3.0 [2.3, 3.7]	4.9 [3.8, 6.0]	18.8 [9.9, 27.7]	-13.9 [-22.9, -4.9]	0.4 [0.2, 0.6]	1.5 [0.8, 2.2]	-1.1 [-1.8, -0.3]
Grasslands	2.6 [2.4, 2.8]	4.9 [4.5, 5.4]	34.4 [22.7, 46.1]	-29.5 [-41.2, -17.8]	0.73 [0.7, 0.8]	4.0 [2.5, 5.5]	-3.3 [-4.8, -1.8]

[1] The numbers between brackets give the lower and upper limit of the 95% confidence interval.

heathland plant communities, however, showed evidence of nutrient enrichment (Riesch *et al.*, 2020), arguing against severe nutrient input in the last decades.

Assuming the estimated average European atmospheric P deposition of 0.30 kg P ha^{-1} yr^{-1} (Tipping *et al.*, 2014), red deer would cause a net P depletion in GTA, especially in grasslands. This could help explain why red deer grazing benefits plant diversity in this habitat type (Riesch *et al.*, 2020), since increasing plant-available P often leads to a decrease in grassland diversity (Ceulemans *et al.*, 2014).

Conclusions

The favourable effects of red deer on nutrient dynamics affirm red deer grazing as an appropriate management option for open habitats. If red deer are abundant and feel safe foraging in open habitats, they can assist habitat conservation, which might be advantageous in large target areas where implementing conventional management is not possible.

References

Camarero L. and Catalan J. (2012) Atmospheric phosphorus deposition may cause lakes to revert from phosphorus limitation back to nitrogen limitation. *Nature Communications* 3, 1118.

Ceulemans T., Stevens C.J., Duchateau L., Jacquemyn H., Gowing D.J.G., Merckx R., Wallace H. ... and Honnay O. (2014) Soil phosphorus constrains biodiversity across European grasslands. *Global Change Biology* 20, 3814-3822.

Clark C.M., Bell M.D., Boyd J.W., Compton J.E., Davidson E.A., Davis C., Fenn M.E., Geiser L., Jones L. and Blett T.F. (2017) Nitrogen-induced terrestrial eutrophication, cascading effects and impacts on ecosystem services. *Ecosphere* 8, e01877.

EEA (2020) *State of nature in the EU – Results from reporting under the nature directives 2013-2018. European Environment Agency.* Publications Office of the European Union, Luxembourg, Luxembourg.

Haynes R.J. and Williams P.H. (1993) Nutrient cycling and soil fertility in the grazed pasture ecosystem. *Advances in Agronomy* 49, 119-199.

Hobbs N.T. (1996) Modification of ecosystems by ungulates. *Journal of Wildlife Management* 60, 695-713.

Mayle B.A., Peace A.J. and Gill R.M.A. (1999) *How many deer? A field guide to estimating deer population size.* Forestry Commission, Edinburgh, UK.

McNaughton S.J., Milchunas D.G. and Frank D.A. (1996) How can net primary productivity be measured in grazing ecosystems? *Ecology* 77, 974-977.

Raab C., Tonn B., Meißner M., Balkenhol N. and Isselstein J. (2019) Multi-temporal RapidEye Tasselled Cap data for land cover classification. *European Journal of Remote Sensing* 52, 653-666.

Richter L., Balkenhol N., Raab C., Reinecke H., Meißner M., Herzog S., Isselstein J. and Signer J. (2020) So close and yet so different. The importance of considering temporal dynamics to understand habitat selection. *Basic and Applied Ecology* 43, 99-109.

Riesch F., Tonn B., Meißner M., Balkenhol N. and Isselstein J. (2019) Grazing by wild red deer: management options for the conservation of semi-natural open habitats. *Journal of Applied Ecology* 56, 1311-1321.

Riesch F., Tonn B., Stroh H.G., Meißner M., Balkenhol N. and Isselstein J. (2020) Grazing by wild red deer maintains characteristic vegetation of semi-natural open habitats, evidence from a 3-year exclusion experiment. *Applied Vegetation Science* 23, 522-538.

Tipping E., Benham S., Boyle J.F., Crow P., Davies J., Fischer U., Guyatt H., Helliwell R., Jackson-Blake L., Lawlor A.J., Monteith D.T., Rowe E.C. and Toberman H. (2014) Atmospheric deposition of phosphorus to land and freshwater. *Environmental Science, Processes and Impacts* 16, 1608-1617.

Umweltbundesamt (2021) UBA.gdi | Hintergrundbelastungsdaten Stickstoff – Bezugszeitraum, Dreijahresmittelwert der Jahre 2013-2015.

Uytvanck J.V., Milotic T. and Hoffmann M. (2010) Nitrogen depletion and redistribution by free-ranging cattle in the restoration process of mosaic landscapes. The role of foraging strategy and habitat proportion. *Restoration Ecology* 18, 205-216.

Wichelhaus A. (2020) Endozoochorer Transport von Pflanzendiasporen durch den Rothirsch (*Cervus elaphus*). PhD dissertation, University of Kassel, Kassel, Germany.

Wilkins K., Aherne J. and Bleasdale A. (2016) Vegetation community change points suggest that critical loads of nutrient nitrogen may be too high. *Atmospheric Environment* 146, 324-331.

Adaptation of fresh lactating dairy cows to grazing

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Abstract

An experiment was carried out to study the productive performance and grazing behavioural adaptation of multiparous and primiparous dairy cows exposed to different feeding strategies during the first 21 days postpartum. Two treatments were compared: T21 (n=15) where cows were fed in a compost barn a total mixed ration (TMR) diet *ad libitum* during the first 21 days in milk (DIM) and T0 (n=15) where cows started grazing the day after calving and were fed a TMR (13.6±1.7 kg dry matter cow^{-1} day^{-1}) after pm milking. At day 22 the T21 cows were moved to T0 treatment till 60 DIM. During the first 21 DIM milk production was higher for T21 than T0 (30.5 vs 27.7 kg cow^{-1} day^{-1}; P<0.01) and for multiparous than primiparous cows (+7.2 kg). Actual grazing time was higher for T21 than T0 during 22 to 60 days postpartum (281 vs 257 min^{-1} day^{-1}; P<0.01) without difference in rumination time (164.4±4.1 min^{-1} day^{-1}). Changes in feeding management during the first 21 DIM had an impact on production and behavioural adaptation of multiparous dairy cows during grazing.

Keywords: fresh dairy cow, adaptation to grazing, feeding strategy

Introduction

Intensification of dairy production systems in Uruguay has involved the implementation of strategies that incorporate confinement with total mixed ration (TMR) either in dry lots or in low-cost barns. Despite the positive potential impact of these strategies on cows productivity, there is also an increased concern on animal welfare (Chilibroste, 2021). Grazing systems are perceived to offer greater freedom for natural behaviour when compared to confinement systems (Arnott *et al.*, 2016). The main objective of this research is to study the productive performance and behavioural adaptation of dairy cows after a period of differential feeding management during the first 21 days postpartum.

Materials and methods

The experiment was performed according to the protocol approved by the Animal Experimentation Committee (CHEA) of the Universidad de la República (UdelaR, Uruguay). It was carried out at the Research Station 'Dr. Mario A. Cassinoni' of the School of Agronomy (Paysandú, Uruguay). After calving, 30 Holstein dairy cows blocked by parity, body weight, body condition score and due calving date, were randomly distributed between two treatments: T21 (n=15; 5 primiparous and 10 multiparous): cows were fed a TMR diet ad-libitum (25.9±1.5 kg dry matter (DM) cow^{-1} day^{-1}) during the first 21 days in milk (DIM) in a compost barn and T0 (n=15: 5 primiparous y 10 multiparous): cows started grazing the day after calving. The cows accessed the grazing paddocks between am and pm milking (8 hours) and were supplemented (13.6±1.7 kg DM cow^{-1} day^{-1}) after pm milking. At day 22 the T21 cows were moved to T0 treatment till 60 DIM. All cows were equipped with Boumatic devices fixed on a collar and placed around the cow neck. The Boumatic collars are being successfully validated for our production systems (Fast, unpublished results, 2021).

The TMR diet offered to the T21 cows was composed, on a DM basis (g kg^{-1}), of maize silage (366), moha hay (62), corn grain (220), soybean meal (97), canola meal (73.2), sunflower meal (52), soybean hulls (97), vitamin and minerals (29). The feeding of grazing treatments consisted of direct grazing and supplementation with TMR (13 kg DM cow^{-1} day^{-1}). Both treatments grazed a mixture of grasses and legumes in a 7-d rotational system with a mean herbage allowance above the ground level of 24±4.5 kg DM cow^{-1} day^{-1}, and 2,142±638 and 1,156±361 of herbage mass pre and post grazing (kg DM ha^{-1}), respectively.

Data were analysed as a complete randomized block design using the SAS System program (SAS® University Edition, SAS Institute Inc., Cary, NC, USA). The model includes effect of treatments (n=2), parity (n=2), grazing week (n=6) and the interactions between effects. The chemical composition of TMR for T0 and T21 and pasture were 341, 353 and 461 g kg^{-1} for neutral detergent fibre, 138, 131 and 189 g kg^{-1} for crude protein and 572, 560 and 228 g kg^{-1} for DM, respectively.

Results and discussion

During the first 3 weeks of differential feeding management, milk production of T21 cows was higher than T0 cows (30.5 vs 27.7 kg cow^{-1} day^{-1}, P=0.02). The average milk yield produced during the first 21 DIM was significantly higher for the multiparous than the primiparous cows (+7.2 kg; P<0.001). After the differential management, T21 cows maintained numerically higher values of milk production, with a residual effect in the next 3 weeks (+3.8 kg; P=0.08) for multiparous (Figure 1) but a residual effect for primiparous cows was not detected (P>0.1). The differences found between T21 and T0 were in line with national antecedents (Fajardo *et al.*, 2015) that compared grazing cows + TMR vs full TMR with dairy cows during the first 60 DIM.

The daily access time to the grazing paddocks was on average 569±56 min per day and the mean grazing time (GT) was 9% higher in T21 than T0 (281 vs 257 min^{-1} day^{-1}, P<0.01). Rumination time during the grazing session was not different between treatments (164.4±4.1 min^{-1} day^{-1}). These differences might be related to the higher milk production (and surely higher DMI) of T21 cows during the first 21 DIM as well as changes in grazing strategies and ultimately selectivity (Chilibroste *et al.*, 2015; Menegazzi *et al.,* 2021). A similar pattern was observed for parity where multiparous cows spent more time grazing (8%) than primiparous cows (279 vs 259 min^{-1} day^{-1}, P<0.01). This reflects the difficult of primiparous cows to adapt to grazing in early lactation as previously described by Chilibroste *et al.* (2012).

Conclusions

Changes in feeding strategy during the first 21 DIM positively impacted on productivity and grazing ingestive behaviour of dairy cows. The impact was different according to parity, with multiparous cows exhibiting a higher direct and residual effect on milk production than primiparous cows. Further research is required for a more comprehensive understanding of the adaptation of dairy cows to grazing during the first weeks in milk.

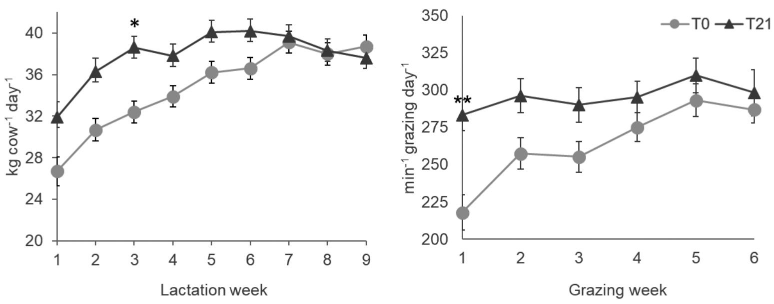

Figure 1. Milk production and grazing time of multiparous according treatments (*; P<0.05; *** P<0.01).

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References

Arnott G., Ferris C.P. and O'Connell N.E. (2016) Review: welfare of dairy cows in continuously housed and pasture-based production systems. *Animal* 11(02), 261-273.

Chilibroste P. (2021) A major challenge for the Uruguayan dairy industry: sustainable growth. *Agrociencia Uruguay* 25(2): e970. Available at: http://agrocienciauruguay.uy/ojs/index.php/agrociencia/article/view/970.

Chilibroste P., Gibb M.J., Soca P. and Mattiauda D.A. (2015) Behavioural adaptation of grazing dairy cows to changes in feeding management: do they follow a predictable pattern? *Animal Production Science* 55, 328-338.

Chilibroste P., Mattiauda D.A., Bentancur O., Soca P. and Meikle A. (2012) Effect of herbage allowance on grazing behavior and productive performance of early lactation primiparous Holstein cows. *Animal Feed Science and Technology* 173, 201-209.

Fajardo M., Mattiauda D.A., Motta G., Genro T.C., Meikle A., Carriquiry M. and Chilibroste P. (2015) Use of mixed rations with different access time to pastureland on productive responses of early lactation Holstein cows. *Livestock Science* 181, 51-57.

Menegazzi G., Giles P.Y., Oborsky M., Fast O., Mattiauda D.A., Genro T.C.M. and Chilibroste P. (2021) Effect of post grazing sward height on ingestive behavior, dry matter intake, and milk production of Holstein dairy cows. *Frontiers in Animal Science* 2, 742685.

Impact of soil type and fertilizer level on forage self sufficiency of Irish dairy farm

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Abstract

For pasture-based dairy production systems, identifying the appropriate yearly stocking rate (SR; cows ha^{-1}) based on the farm grass growth is the key strategic decision driving the overall farm business. This paper simulates the effects of varying stocking rates (2 to 3 cow ha^{-1}, 0.25 unit changes), fertilizer nitrogen (N) application rates (0 to 300 kg N ha^{-1} 50 kg ha^{-1} unit changes), soil type, (heavy and a free draining soil) through the PBHDM (Pasture-Based Herd Dynamic Milk) model in conjunction with the Moorepark St Gilles (MoSt) grass growth model. The model outputs show an average N response of 18.2 kg dry matter (DM) kg^{-1} N applied (range of 8.3-29.2 kg DM kg^{-1} N). Soil type did not affect grass growth or N response. More of the grass on the free draining soil was consumed as grazed grass (average +644 kg DM cow^{-1} and +1,611 kg DM ha^{-1} at 250 kg N ha^{-1} and SR 2.5 cow ha^{-1}) due to increased grazing days, while cows on the heavy soils consumed more grass silage. Systems stocked at 2.5 cows ha^{-1} and applying 250 kg N ha^{-1} were self-sufficient for forage production, while a decrease of 50 kg of N fertilizer required a reduction of stocking rate of 0.20 cow ha^{-1} to maintain forage self-sufficiency.

Keywords: N response, stocking rate, farm modelling, self-sufficiency, Irish dairy system

Introduction

Irish dairy farm profitability is driven by increased grass productivity and improved efficiency of the conversion of grazed pasture to animal products. The removal of the European Union milk quota regime has allowed the Irish dairy industry to increase cow numbers facilitated by increased land area associated with dairying (Kelly *et al.*, 2020). The Department of Agriculture, Food and the Marine 'Food Wise 2025' strategy set as its guiding principle that environmental protection and economic competitiveness would be equally considered and complementary; one was not to be achieved at the expense of the other. While the Irish dairy industry has expanded since the removal of milk quotas, the European Green Deal announced by the European Commission in December 2019 has also identified the requirement to reduce dependency on fertilizer in food production systems. The objective of this paper is to investigate the influence of different N application rates and its interaction with SR and soil type on the self-sufficiency of Irish dairy farms using a modelling approach.

Materials and methods

This paper simulates the effects of varying yearly stocking rates (2 to 3 cow ha^{-1}, 0.25 unit changes), fertilizer application rates (0 to 300 kg N ha^{-1}, 50 kg ha^{-1} unit changes), soil type (heavy and a free draining soil) and weather conditions (16 years) through the PBHDM (Pasture-Based Herd Dynamic Milk) model (Ruelle *et al.*, 2016) in conjunction with the Moorepark St Gilles (MoSt) grass growth model (Ruelle *et al.*, 2018). The models works with a daily time step, predicting daily grass growth (paddock level), animal intake (both at grazing and indoor), milk yield, body condition score and bodyweight change. Farm size was fixed at 40 ha of grassland and included 40 paddocks. Concentrate supplementation was fixed for all simulation and was fed at a level of 600 kg dry matter (DM) cow^{-1} year^{-1}. Grass silage fed, purchase and sale was the changing variable. Sixteen years of daily weather data from the Moorepark research farm (2003-2018, 52°09'52.3'N 8°15'36.6'W) were used in the simulation. A total of 70 scenarios were analysed: (1) 5 stocking rates – 2.00, 2.25, 2.50, 2.75 and 3.00 cow ha^{-1}; (2) 7 N chemical fertilizer rates: 0 to 300 kg N ha^{-1} increasing by steps of 50 kg ha^{-1}; and (3) 2 soil types: a free draining soil (FDS: 6% organic matter (OM), 60% sand and 15% clay, soil depth of 100 cm) and a heavy soil (HS: 8% OM, 28% sand and 36% clay, soil depth of 100 cm). For each scenario, simulations were completed for 16 years on a continuous cycle, meaning that an event occurring in Year 1 had consequences in Year 2 and so on. Main outputs simulated were grass growth, dairy cow grass DM intake, silage fed, and farm forage self-sufficiency.

Results and discussion

The average grass growth across scenarios and years was close to 12,500 kg DM ha^{-1}, with the minimum grass growth of 5,000 kg DM ha^{-1} (SR 2, 0 fertilizer, FDS, 2018) and the maximum grass growth of 17,400 kg DM ha^{-1} (SR 3,300 fertilizer, HS, 2011).

The N response was calculated based on the previous level of N fertilizer (increasing in steps of 50 kg N ha^{-1}; Table 1). The average N response due to increasing N fertilizer application was 18.2 kg DM ha^{-1} with a minimum of 8.3 kg DM ha^{-1} (SR of 3 cows ha^{-1} and increasing fertilizer from 250 to 300 kg N ha^{-1}, FDS in 2018) and a maximum of 29.2 kg N ha^{-1} (SR of 2.25 cows ha^{-1} and increasing fertilizer from 0 to 50 kg N ha^{-1}, FDS, 2014). The N response decreased with increasing N fertilizer rate with an average response across all the simulations of 22.5 kg DM kg^{-1} N applied between the level 0 and 50 kg N ha^{-1}, and a response of 14.5 kg DM kg^{-1} N applied as rate increases from 250 to 300 kg N ha^{-1}. Previous studies have shown similar results; Enriquez-Hidalgo *et al.* (2016) found a response ranging from 8.0 to 28.3 kg DM kg^{-1} N applied at application rate of 120 kg N ha^{-1} compared to 0 and Hennessy *et al.* (2008) had responses varying between 15 and 33.1 kg DM kg^{-1} N applied, compared to 0.

Across all simulations, the average intake per cow annually was 3,063 kg DM grazed grass, 1,635 kg DM grass silage and 600 kg DM of concentrate fixed. At the SR 2.5 cows ha^{-1}, the increase in grass intake per cow per kg of N applied was of 4.7 and 1.8 kg DM kg^{-1} N for the FDS and HS, respectively. These results show that an increase in N fertilizer application on a FDS leads to an increase in grass intake per cow, while on HS the extra fertilizer produced a small increase of grass intake per cow and a bigger increased silage stock. Grass intake per cow declined as SR increased, while grass intake per ha increased.

Table 1. Mean yearly grass growth, grazing indicators and dairy cow DM intake at different N fertilizer rates on different soil types at SR 2.5 cow ha^{-1}.

	Free draining soil					Heavy soil				
Fert level	Full Gdays	Grass growth	N marginal response	Grass intake	Total silage	No SupDay	Grass growth	N response	Grass intake	Total silage
kg N ha^{-1}	Days[1]	kg DM ha^{-1}	kg DM kg^{-1} N	kg DM cow^{-1}	kg DM cow^{-1}	Days[1]	kg DM ha^{-1}	kg DM kg^{-1} N	kg DM cow^{-1}	kg DM cow^{-1}
0	119	8,921		3,002	1,729	167	10,249		2,759	1,922
50	150	9,989	21.4	3,147	1,576	181	11,378	22.6	2,862	1,825
100	168	11,026	20.7	3,260	1,467	188	12,407	20.6	2,899	1,796
150	183	12,003	19.5	3,362	1,387	191	13,338	18.6	2,932	1,764
200	202	12,861	17.2	3,462	1,287	194	14,159	16.4	2,952	1,745
250	211	13,669	16.2	3,527	1,227	198	14,933	15.5	2,964	1,731
300	219	14,403	14.7	3,567	1,190	198	15,645	14.2	2,974	1,721

[1] Full Gdays: number of days with grass only (Concentrate could be fed but no silage).

With the introduction of the Farm to Fork Strategy (2021) there will be a reduction in the quantities of chemical N fertilizer applications on farms. In this study when fertilizer N application was reduced, the number of SR categories that were forage self-sufficient declined (Figure 1). A linear regression has been conducted on all the data to find the relationship between N applied, SR and farm self-sufficiency. Other parameters included in the regression were the soil type, location and year. The results showed that a decrease in N fertilizer of 50 kg ha^{-1} $year^{-1}$ will require a decrease of SR in 0.20 cow ha^{-1} to maintain self-sufficiency (R^2=0.91) if no other action is taken. Long term strategies exist that could be used to maintain grass production with reduced N application, e.g. achieving and maintaining optimum soil fertility (P,K applications) and considerable levels of white clover inclusion.

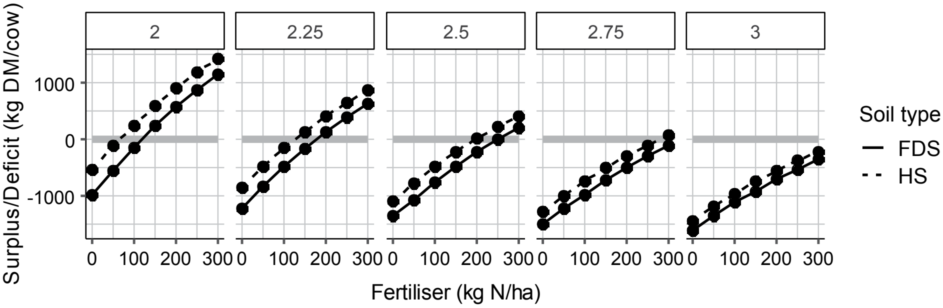

Figure 1. On farm surplus or deficit depending on soil type, stocking rate and fertilizer level. (FDS = free draining soil, HS = heavy soil).

Conclusions

This study shows that irrespective of soil type, an average reduction of 50 kg in chemical N fertilizer application per ha would lead to a reduction in SR of 0.20 cow ha^{-1} if no additional measures are taken on farm to maintain/increase herbage production. The variation in grass growth, grass intake per cow and N response across years, seasons and soil types highlights the necessity for farmers to be reactive to be able to adapt each year to the dynamic conditions imposed by both weather and soil conditions.

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References

Enriquez-Hidalgo D., Gilliland T.J., Egan M. and Hennessy D. (2018) Production and quality benefits of white clover inclusion into ryegrass swards at different nitrogen fertilizer rates. *Journal of Agricultural Science* 156, 378-386.

Hennessy D., O'Donovan M., French P. and Laidlaw A. (2008) Manipulation of herbage production by altering the pattern of applying nitrogen fertilizer. *Grass and Forage Science* 63, 152-166.

Kelly P., Shalloo L., Wallace M. and Dillon P. (2020) The Irish dairy industry–Recent history and strategy, current state and future challenges. *International Journal of Dairy Technology* 73, 309-323.

Ruelle E., Delaby L., Wallace M. and Shalloo L. (2016) Development and evaluation of the herd dynamic milk model with focus on the individual cow component. *Animal* 10(12), 1986-1997.

Ruelle E., Hennessy D. and Delaby L. (2018) Development of the Moorepark St Gilles grass growth model (MoSt GG): a predictive model for grass growth for pasture based systems. *European Journal of Agronomy* 99, 80-91.

A review of beef and sheep grazing management suitable for hill and upland environments

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Abstract

The hills and uplands contain an array of sward types, with different grazing and conservation requirements. Grazing has a direct impact on plant community structure and composition, which subsequently affects the whole ecosystem. Thus, there is a need to identify optimal grazing management that will ensure the sustainability of these environments for a range of ecosystem services including biodiversity and food production. This paper reviews the scientific literature on beef and sheep grazing strategies for the hills and uplands, with a focus on animal health, performance and the environment, particularly biodiversity.

Keywords: animal performance, health, environment, vegetation, soil, biodiversity

Introduction

The hills and uplands are renowned for their harsh conditions, difficult terrain, shorter growing season and low quality vegetation. In NI, almost 90% of farms in the hills and uplands consist of cattle and sheep (DAERA, 2019) and therefore these livestock are the predominant method of managing these environments (Fraser *et al.*, 2014). The hills and uplands contain a range of sward types, habitats and biodiversity, which deliver of a range of ecosystem services. As such, their sustainable management is important to ensure their ability to support the livestock industry in future years, and for the protection and enhancement of this natural environment (Fraser *et al.*, 2014). The aim of this paper is to review the scientific literature on beef and sheep grazing management in the hills and uplands, assessing the impacts on animal health and performance as well as the environment and biodiversity, while also identifying key knowledge gaps.

Beef and sheep grazing management for the hills and uplands

Mono grazing consists of grazing just one livestock species and is often implemented using sheep, due to their ability to graze the hills and uplands for most of the year. In the literature, high and low stocking rates (SR) were reported at 1.50 and 0.66 ewes ha^{-1}, respectively (Critchley *et al.*, 2008). Stocking rate of the Irish national hill flock is estimated at 0.7 ewes ha^{-1} (Walsh *et al.*, 2016). Often SR are reduced during the winter months and ewes removed from the uplands for mating and lambing (Walsh *et al.*, 2016). Mono-grazing of cattle is less well documented in the literature and often only occurs during the summer. A 'biodiversity-friendly rotation' is an alternative to a set-stocked mono grazing strategy. Here, the grazing area is divided into several rotational plots, with one plot having an extended grazing interval over the main summer flowering period. This was assessed using both cattle and sheep mono grazing systems (Enri *et al.*, 2017).

Mixed grazing involves grazing both cattle and sheep on the same pasture and utilizes the different grazing behaviours of both ruminant species (Rook *et al.*, 2004). In mixed grazing, cattle are often only grazed during the summer months. Examples of SR from the literature are 0.66 ewes ha^{-1} plus 0.75 cows ha^{-1} (Critchley *et al.*, 2008) and 0.5 ewes ha^{-1} plus 0.3 cattle ha^{-1} (Holland *et al.*, 2008). The ratio of cattle to sheep is often determined by the type and quantity of vegetation available, the terrain and the ground conditions. Out-wintering of cattle on the hill is also practised; however, there may be a need for supplementary feeding. In the Burren Uplands in Co. Clare a substantial 73.5% of the area was used for wintering grazing cattle at a mean SR of 0.25 suckler cows ha^{-1} (Dunford and Feehan, 2001). Agri-environmental (AE) schemes aim to conserve and restore biodiversity while avoiding the negative impacts of intensification or abandonment. Some AE schemes dictate the SR or grazing duration. However, this has received criticism, as an SR could be appropriate for one area, yet ecologically damaging for another (Critchley *et al.*, 2008). Instead, habitat-specific SR, that are flexible and tailored are needed (Walsh *et al.*, 2016).

Impacts on animal health and performance

In a mono-grazing system on degraded upland wet heath, lower SR have been reported to result in improved ewe and lamb live weight (LW) (Critchley *et al.*, 2008). Walsh *et al.* (2016) reported that mono-grazing sheep on unimproved hill has the potential to sustain a long-term productive sheep enterprise. Mixed grazing can increase livestock productivity and output per hectare compared to mono grazing, primarily due to the greater LW gain (LWG) of calves relative to lambs (Fraser *et al.*, 2014). However, Wright *et al.* (2006) reported contrasting results, with gains in lamb LWG occurring at the expense of steer LWG, suggesting that cattle are more susceptible to poor grazing conditions and may be disadvantaged where SR is too high. The impacts on performance from out-wintering cattle do not appear to be documented in the literature. Likewise the effects of winter grazing sheep are limited in the literature despite the fact that management of pasture and ewes during this time will have large impacts on spring vegetation quality and height and lambing performance (Dwyer *et al.*, 2016). Furthermore, the literature is particularly limited on the impacts on animal health. A survey of extensive hill sheep flocks in GB reported that sheep scab was considered the greatest health concern in terms of impact on welfare and productivity, followed by lameness, ectoparasites, abortion, feeding/minerals and endoparasites (Morgan-Davies *et al.*, 2006). Furthermore, some diseases can be fatal for wildlife (Sargison and Edwards, 2009). Although current research rarely considers health impacts, it is clear that problems exist within these environments, and can have wider implications for wildlife. Thus, future research is required to address this knowledge gap.

Impacts on the environment

Mono-grazing of sheep has been associated with overgrazing and selective grazing, leading to a reduction in fauna and flora diversity (Enri *et al.*, 2017). Reducing SR can assist in the recovery of vegetation, provided the component species are still present (Critchley *et al.*, 2008); otherwise it simply leads to greater herbage height (Holland *et al.*, 2008). Overgrazing can also lead to the formation of sheep scars in the landscape, causing exposure of bare soil and subsequent erosion (Evans, 1997). A biodiversity-friendly rotational mono-grazing strategy assists in increasing both the abundance and diversity of flower-visiting insect species in comparison to a continuously grazed plot (Enri *et al.*, 2017).

Sheep are selective grazers, grazing high quality plant parts (Rook *et al.*, 2004), whereas cattle patch-graze, consume woody material and invasive hill grasses (Critchley *et al.*, 2008). In addition, cattle and sheep tend to graze at different locations and heights, with sheep accessing higher and steeper areas of the hill (Holland *et al.*, 2008). Furthermore, the different distribution of excreta from cattle and sheep is beneficial, as the concentrating of nutrients at excreta patches may alter the competitive advantage between plant species (Rook *et al.*, 2004). Mixed grazing, however, does not always fulfil the desired objective of increasing sward diversity as was the case for Holland *et al.* (2008). As with any grazing strategy negative impacts could be specific to one species of vegetation due to animals selecting for large quantities of live shoots (Grant *et al.*, 1987). Thus, grazing management may need to be altered at the first signs of damage to vegetation (Critchley *et al.*, 2008). Trampling by cattle can be beneficial by opening regeneration niches for species colonisation (Rook *et al.*, 2004). However, excessive trampling can negatively affect vegetation and the physical properties of soil, leading to reduced infiltration rates, increased runoff and erosion (Nguyen *et al.*, 1998). Supplementary feeding of out-wintered cattle can upset upland ecology by introducing additional nutrients to pastures (Dunford and Feehan, 2001). However, positive effects can also be realised such as facilitating early seedling growth (Evans *et al.*, 2006). The long-term sustainability of grazing management needs to be assessed, as both negative (Critchley *et al.*, 2008) and positive (Walsh *et al.*, 2016) environmental impacts have been documented in the literature.

Conclusions

This review has highlighted a number of knowledge gaps, and the need for tailored management in the hills and uplands. Thus, a comprehensive research program is required not only to gain a better understanding of the long term grazing management suitable for this dynamic environment but also to determine how best these systems need to be monitored to assess their impacts.

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References

Critchley C.N.R., Adamson H.F., Mclean B.M.L. and Davies O.D. (2008) Vegetation dynamics and livestock performance in system-scale studies of sheep and cattle grazing on degraded upland wet heath. *Agriculture Ecosystems & Environment* 128, 59-67.

DAERA (2019) Statistical Review of Northern Ireland Agriculture 2019.

Dunford B. and Feehan J. (2001) Agricultural practices and natural heritage: a case study of the Burren Uplands, Co. Clare. Tearmann. *Irish Journal of Agri-environmental Research* 1, 19-34.

Dwyer C.M., Conington J., Corbiere F., Holmoy I.H., Muri K., Nowak R., Rooke J., Vipond J. and Gautier J.M. (2016) Invited review: Improving neonatal survival in small ruminants: science into practice. *Animal* 10, 449-459.

Enri S.R., Probo M., Farruggia A., Lanore L., Blanchetete A. and Dumont B. (2017) A biodiversity-friendly rotational grazing system enhancing flower-visiting insect assemblages while maintaining animal and grassland productivity. *Agriculture, Ecosystems and Environment* 241, 1-10.

Evans D.M., Redpath S.M., Elston D.A., Evans S.A., Mitchell R.J. and Dennis P. (2006) To graze or not to graze? Sheep, voles, forestry and nature conservation in the British uplands. *Journal of Applied Ecology* 43, 499-505.

Evans R. (1997) Soil erosion in the UK initiated by grazing animals – A need for a national survey. *Applied Geography* 17, 127-141.

Fraser M.D., Moorby J.M., Vale J.E. and Evans D.M. (2014) Mixed grazing systems benefit both upland biodiversity and livestock production. *Plos One,* 9, 8.

Grant S.A., Torvell L., Smith H.K., Suckling D.E., Forbes, T.D.A. and Hodgson J. (1987) Comparative studies of diet selection by sheep and cattle: blanket bog and heather moor. *Journal of Ecology* 75, 947-960.

Holland J., Waterhouse A., Robertson D. and Pollock M. (2008) Effect of different grazing management systems on the herbage mass and pasture height of a *Nardus stricta* grassland in western Scotland, United Kingdom. *Grass and Forage Science* 63, 48-59.

Morgan-Davies C., Waterhouse A., Milne C.E. and Stott A.W. (2006) Farmers' opinions on welfare, health and production practices in extensive hill sheep flocks in Great Britain. *Livestock Science* 104, 268-277.

Nguyen M.L., Sheath G.W., Smith C.M. and Cooper A.B. (1998) Impact of cattle treading on hill land 2. Soil physical properties and contaminant runoff. *New Zealand Journal of Agricultural Research* 41, 279-290.

Rook A.J., Dumont B., Isselstein J., Osoro K., Wallisdevries M.F., Parente G. and Mills J. (2004) Matching type of livestock to desired biodiversity outcomes in pastures – a review. *Biological Conservation* 119, 137-150.

Sargison N. and Edwards G. (2009) Tick infestations in sheep in the UK. *In Practice* 31, 58-65.

Walsh M., Hanrahan J. P., O'Malley L. and Moles R. (2016) Trends, over 14 years, in the ground cover on an unimproved western hill grazed by sheep, and associated trends in animal performance. *Irish Journal of Agricultural and Food Research,* 55, 47-62.

Wright I.A., Jones J.R., Davies D.A., Davidson G.C. and Vale J.E. (2006) The effect of sward surface height on the response to mixed grazing by cattle and sheep. *Animal Science* 62, 271-276.

Composition of excreta generated by dairy cattle on farms in NW Spain with different feeding systems

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Abstract

The study investigated the effects of season and feeding system on the chemical composition and fibre content of the excreta generated by lactating cows. To this end, 19 farms in Galicia (NW Spain) were selected for study and classified into different feeding systems according to the main type of fodder provided: pasture grass (PG), grass silage (GS), corn silage (CS) and combined grass/corn silage (GCS). Season only affected the carbon content, with higher values in spring than in autumn. Regarding the type of fodder provided, the excreta generated in the CS system yielded the highest amount of dry matter, which was significantly higher than in the PG and GCS systems. By contrast, the CS system yielded the lowest total N, which differed significantly from that in the GS system, and also the lowest organic matter, carbon, phosphorus and magnesium contents, which differed significantly from those in the GCS system. The GS and GCS systems yielded the highest acid detergent fibre (ADF) values.

Keywords: faecal excretion, dairy cow, manure, excreta, fertilization

Introduction

Optimization of nutrient flows in livestock farms requires appropriate management of manure. Animal manure is a nutrient resource containing most of the essential elements required for plant growth. Conservation and recycling of nutrients by using manure as fertilizer is important to prevent generation of surplus nutrients that pollute soil, atmosphere and water. The type and composition of the diet are among the main factors that affect the composition of the excreta (Weiss *et al.*., 2009). Lack of knowledge about the relationship between the chemical composition of food and of the excreta in lactating cows was addressed in Galicia, the main dairy producing region in Spain, with the aim of improving nutrient management and recycling.

Materials and methods

Nineteen dairy cow farms were selected in order to represent the different feeding systems in Galicia (Botana *et al.*, 2019) according to the main type of fodder supplied: pasture grass (PS, n=5), grass silage (GS, n=4), grass and corn silage (GCS, n=5) and corn silage (CS, n=5). Between April 2018 and April 2019, the farms were surveyed to determine milk yield (4% fat corrected milk, ECM) and the type of feed provided. Feed and excreta were sampled in spring and autumn in all feeding systems and in PS also in summer and winter. The nutritional value of the feed was determined in the laboratory by determining organic matter (OM), crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF). Excreta were analysed to determine the dry matter (DM), OM, total nitrogen (N), total carbon (C), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sodium (Na) contents, NDF, ADF and the cellulose, lignin and hemicellulose fractions of NDF and ADF. The N:P and C:N ratios were calculated. Data were analysed by ANOVA with two fixed factors (feeding system and season) using Rstudio (2016).

Results and discussion

The observed difference in the composition of the diets between seasons in the PS system, with a higher proportion of fresh grass in spring and summer (49.2%) than in autumn and winter (31.5%), and a lower proportion of grass silage in spring and summer (18.5%) than in autumn and winter (35.4%), was not reflected in differences in the excreta composition (data not shown). Overall analysis of the four systems showed that the season (spring vs autumn) only affected the C content of the excreta, which was higher in spring (44.7% DM basis) than in autumn (41.5% DM basis, $P<0.05$).

The diets of lactating cows (Table 1) influenced DM, OM, C, N, P and Mg in the excreta (Table 2). The CS system yielded the highest DM (14.7% FW basis, $P<0.001$) and the lowest values of OM, C, N and P (75.8 and 40.4% DM basis, 27.9 and 6.0 g kg^{-1} DM, respectively, $P<0.05$). The GCS system yielded the highest OM and C values (84.9 and 45.2% DM basis, respectively, $P<0.05$), with differences only relative to CS. The amount of N excreted was highest in GS, significantly higher than in CS, but not different from the values in PS and CGS. Although intake of N was greater in GCS and CS (622.2 and 646.4 g cow^{-1} and day^{-1}, $P<0.001$), according to Santiago *et al.* (2020) N-use efficiency of the diet in relation to milk production is higher for CS, which may explain the lower excretion of N in this system. In addition, the higher excretion of N in GS may be related to the low digestibility of the protein in silage, along with a high input of energy via consumption of concentrates (11.31 kg cow^{-1} and day^{-1}), which may increase the amount of protein in the excreta (Broderick *et al.*, 2003).

P excretion was highest in GCS (7.7 g kg^{-1} DM) and differed significantly from that in CS (6.0 g kg^{-1} DM, $P<0.05$). According to Arriaga *et al.* (2009), improved N-use efficiency at animal level can optimize the efficient use of P, reducing the amount excreted. The Mg contents were highest in GS and GCS.

Although the PS diet had the highest ADF (Table 1), the highest ADF values in excreta were yielded by the PS and GCS systems (Table 2). The cellulose content was slightly higher in GCS, and the lignin content was higher in PS. The higher ash content in CS was related to the presence of silica derived from the sand used as bedding in this system.

Table 1. Characteristics of the diets of lactating cows according to the feeding system.[1,2]

	PS	GS	GCS	CS	*P*-value
Number of samples	10	7	10	10	
Intake, kg cow^{-1} day^{-1}:					
Total dry matter intake, kg	18.00 c	22.68 b	24.56 a,b	25.80 a	***
Pasture grass, kg DM	6.51 a	0.21 b	0.00 b	0.00 b	***
Grass silage, kg DM	5.34 b,c	10.86 a	5.89 b	2.18 c	***
Corn silage, kg DM	1.04 c	0.00 c	8.20 b	11.26 a	***
Dry forage, kg DM	1.06	0.29	0.46	0.92	NS
Concentrates, kg DM	4.05 b	11.31 a	10.00 a	11.45 a	***
Diet composition:					
Forage, % DMI	77.82 a	49.95 c	59.09 b	55.58 b,c	***
Concentrates, % DMI	22.18 c	50.05 a	40.91 b	44.42 a,b	***
OM, g kg^{-1} DM	904.36	892.36	930.04	920.30	NS
CP, g kg^{-1} DM	162.88	146.11	159.12	155.96	NS
NDF, g kg^{-1} DM	442.40 a	338.20 b	336.91b	351.11b	***
ADF, g kg^{-1} DM	257.68 a	205.56 b	204.51 b	190.58 b	***
OMD, g kg^{-1} OM	657.05 b	704.20 a,b	697.62 a,b	719.44 a	**

[1] PS = pasture grass; GS = grass silage; GCS = grass and corn silage; CS = corn silage.

[2] Means within the same row with different letters differ among treatments (Scheffé's test). NS = not significant ($P>0.05$); * $P<0.05$; ** $P<0.01$; *** $P<0.001$.

Table 2. Chemical composition and fibre content of the excreta of lactating cows according to the feeding system and the season in Galicia.

	PS	GS	GCS	CS	Spring	Autumn	F	S	*P*-value
No. of samples	15	9	17	16	37	20			
pH	7.8	7.6	7.5	7.6	7.6	7.7	NS	NS	NS
DM, g $100g^{-1}$ FW	11.8 c	13.5 a,b	12.9 b,c	14.7 a	13.1	13.4	***	NS	***
OM, g $100g^{-1}$ DM	81.7 a,b	80.1 a,b	84.9 a	75.8 b	81.3	79.7	**	NS	*
C, g $100g^{-1}$ DM	44.7 a,b	44.2 a,b	45.2 a	40.4 b	44.7 a	41.5 b	*	*	*
N, g kg^{-1} DM	30.4 a,b	35.5 a	31.5 a,b	27.9 b	30.8	30.9	*	NS	*
P, g kg^{-1} DM	6.2 a,b	7.6 a,b	7.7 a	6.0 b	6.6	7.2	*	NS	*
K, g kg^{-1} DM	14.5	14.1	12.0	10.1	12.4	12.5	NS	NS	NS
Ca, g kg^{-1} DM	23.4	35.0	26.0	17.3	24.0	24.8	NS	NS	NS
Mg, g kg^{-1} DM	5.5 b	8.5 a	8.0 a	5.4 b	6.5	7.1	***	NS	*
Na, g kg^{-1} DM	4.0	4.9	4.8	4.4	4.6	4.4	NS	NS	NS
C: N	15.1	13.1	15.0	14.6	14.9	14.0	NS	NS	NS
N: P	5.1	4.8	4.4	4.9	5.0	4.4	NS	NS	NS
NDF, g $100g^{-1}$ DM	52.4	46.8	54.6	50.3	52.3	50.2	NS	NS	NS
ADF, g $100g^{-1}$ DM	32.6 a	28.1 b	32.4 a	28.7 b	31.1	30.1	**	NS	*
HC, g $100g^{-1}$ DM	19.9	19.0	22.3	21.7	21.3	20.3	NS	NS	*
CE, g $100g^{-1}$ DM	19.4 a,b	14.9 b	21.1 a	19.4 a,b	19.5	18.6	**	NS	*
LIG, g $100g^{-1}$ DM	13.21 a	13.19 a,b	11.3 a,b	9.4 b	11.6	11.5	***	NS	**
Ash, g $100g^{-1}$ DM	5.8 b	5.1 b	2.7 b	14.0 a	6.6	7.9	***	NS	*

[1] PS = pasture grass; GS = grass silage; GCS = grass and corn silage; CS = corn silage; F = feeding system; S = season; FW = fresh weight; HC = hemicellulose; CE = cellulose; LIG = lignin.

[2] Means within the same row with different letters differ among treatments (Duncan's test). NS = not significant ($P>0.05$); * $P<0.05$; ** $P<0.01$; *** $P<0.001$.

Conclusions

The feeding system strongly influences the chemical composition of the excreta (DM, OM, N, P, Mg) and the type of fibre. This is expected to condition the subsequent processes of OM transformation and N losses during pit storage and also the slurry composition.

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References

Arriaga H., Pinto M., Calsamiglia S. and Merino P. (2009) Nutritional and management strategies on nitrogen and phosphorus use efficiency of lactating dairy cattle on commercial farms: An environmental perspective. *Journal of Dairy Science* 92(1), 204-215.

Botana A. (2019) Estudio de los sistemas de alimentación en explotaciones de vacuno de Galicia: efecto de la composición de la dieta sobre el perfil de ácidos grasos y antioxidantes liposolubles de la leche. Doctoral thesis, University of Santiago de Compostela, Spain.

Broderick G.A. (2003) Effects of varying dietary protein and energy levels on the production of lactating dairy cows. *Journal of Dairy Science* 86, 1370-1381.

Santiago C., García M.I. and Báez D. (2020) Nitrogen use efficiency in dairy cows from different diets in north-western Spain. *Grassland Science in Europe* 25, 348-350.

Weiss W.P., Willet L.B., St-Pierre N.R., Borger D.C., Mckelvey T.R. and Wyatt D.J. (2009) Varying forage type, metabolizable protein concentration, and carbohydrate source affects manure excretion, manure ammonia, and nitrogen metabolism of dairy cows. *Journal of Dairy Science* 92, 5607-5619.

Comparison of a feeding variant of the current and future grassland-based milk production programme

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Abstract

The aim of the Grassland-Based Milk and Meat (GMM) programme, part of the direct payment ordinance in Switzerland, is to maintain ruminant production based on herbage and the reduced use of concentrates. Variants of the current and future GMM programme were compared in our study. During the winter, the forage of 64 dairy cows consisted of hay. From the end of March onwards, a continuous switch to full grazing was made. During the first 90 days in milk, cows in the 'current GMM' treatment (ACT) received 2 kg of energy and 1 kg of protein concentrate (as fed). In the 'future GMM' treatment (FUT), the cows received 3 kg of energy concentrate exclusively. The results of the first six official milk recordings are presented here. Cows in the FUT treatment produced less energy-corrected milk (ECM) compared to those in the ACT group, and their milk urea content was lower. The lactose content was minimally higher in the FUT treatment group than in the ACT group. The milk fat and protein contents, as well as the somatic cell counts, were not influenced by the treatment type. With a herbage-based ration, the protein-reduced concentrate supplementation of the cows leads to a lower milk yield with a reduced milk urea content.

Keywords: grassland, feeding systems, dairy cow, herbage, protein

Introduction

Feed-food competition and the massive import of protein rich feeds like soya bean meal, of which at least 1/3 is used as ruminant feed, are largely disapproved of by society and authorities, especially as the medium-term environmental goals of Swiss agriculture with regard to nitrogen losses and emissions will probably not be achieved.

On the 1st January 2014 the GMM programme was introduced in Switzerland as a part of the direct payment ordinance. The aim of the GMM programme is to maintain ruminant production geared to local conditions that are based on fresh and preserved herbage and a reduced use of concentrates. Participation in the GMM programme is voluntary and farmers receive contributions upon their participation in the programme. In this context, the reorientation of the future GMM and administrative simplifications of the programme were discussed among stakeholders and the Federal Office of Agriculture. Subsequently, the suitability of the future variants proposed by the Federal Office of Agriculture was assessed (Schori, 2020). One proposed variant of the future GMM programme would only allow the purchase of concentrates for ruminants with a maximum crude protein (CP) content of 12% per kg dry matter (DM). Consequently, the main part of the protein that is needed to cover the protein requirements of ruminants should come from the feed produced on the farm. In this study, variants of the current and future GMM were compared in a herbage-based feeding system throughout the standard lactation period of 305 d of dairy cows.

Animals, materials and methods

The experiment was carried out on the organic farm, Ferme-Ecole de Sorens, located in Sorens, Switzerland. During the winter, the forage of the Holstein and Swiss Fleckvieh cows consisted of hay *ad libitum* (5.3±0.10 (standard deviation [sd]) MJ net energy for lactation [NEL] and 118±4.8 (standard deviation, SD) g CP per kg DM, n=4). From the end of March onwards, a continuous switch to full grazing was made (fresh herbage: 6.1±0.39 (SD) MJ NEL and 158±31.7 (SD) g CP per kg DM, n=6). The average calving date of the experimental dairy cows was 25 February 2021 (±35 d (SD)). During the first 90 days in milk, the cows in the ACT treatment received 2 kg of an energy (7.7±0.17 (SD) MJ NEL and 136±2.7 (SD) g CP per kg DM, n=3) and 1 kg of a protein concentrate (8.2±0.02 (SD) MJ NEL and 412±2.9 (SD) g CP per kg DM, n=3). In the FUT treatment, the cows received 3 kg of the same energy concentrate exclusively. In total, 64 cows, of which 40% were primiparous, were paired in relation to their breed, lactation number, and calving date. Every 14-days, during the official milk recording, the individual milk yield was recorded. At the same time, milk samples from two consecutive milkings were taken from each cow. The fat, protein, lactose, and urea contents as well as the somatic cell counts of these milk samples were analysed. The energy-corrected milk (ECM) was calculated based on the fat, protein, and lactose content of the milk according to Münger *et al.* (2021). A mixed linear model (R Core Team, 2021) was used for the evaluation, with the treatment type and number of recordings as well as their interaction forming the fixed factors. The cow pairs were set as a random factor.

Results and discussion

The preliminary results of the first six official milk recordings are presented in Table 1. The Holstein and Swiss Fleckvieh dairy cows in the FUT treatment produced less milk and ECM compared to those in the ACT group. With a CP-to-NEL ratio of 22.3 and 25.7 g MJ^{-1} the hay and the fresh herbage, respectively, should contain sufficient CP. Nevertheless, with the effect of an additional 257 g CP on the milk yield, the finding of approximately 1.7 kg more milk per cow and day during the first 12 weeks of lactation, is rather surprising. Law *et al.* (2009) found with an increase of the CP content of the ration from 114 to 144 g kg^{-1} DM and 144 to 173 g kg^{-1} DM an increase in milk yield during the first 150 days of lactation of 6.4 and 3.6 kg per cow and day, respectively. In our experiment, the difference between the ACT vs FUT treatments was approximately 15 g CP kg^{-1} DM feed. Consequently, our results seem to be consistent with the milk yield difference obtained by Law *et al.* (2009) at a feed protein level between 144 to 173 g CP kg^{-1} DM. Interestingly, the difference in the milk protein yield between the two feeding treatments in our experiment agrees quite well with the estimated value according to Huthanen and Hristov (2009) for a northern European data set (47 vs 42 g d^{-1}). In contrast to our results, Zang *et al.* (2021) obtained a significant effect of the protein content of the ration, on the milk yield of dairy cows in the first three weeks in milk, but not during the first 13 weeks. Moreover, the lactose content was minimally higher in the FUT treatment group than in the ACT treatment group. In general, lactose content is relatively constant and related to energy balance (Reist *et al.*, 2002), udder health, and metabolic disorders (Costa *et al.*, 2019). Milk fat and protein content were not influenced in our study by the treatments (FUT vs ACT). With the increase of 144 to 173 g CP kg^{-1} DM, Law *et al.* (2009) also detected no differences in the content of the milk from dairy cows at the beginning of lactation. As an indication of udder health, the somatic cell count was used in our study. No differences were observed according the somatic cell counts between the FUT vs ACT treatments. Not surprisingly, the milk urea content was lower in the FUT treatment group compared to the ACT treatment group. An advantage of the future GMM variant would be that less protein-rich feedstuffs, such as soya bean meal, rapeseed meal, or grain legumes, are used in the feeding of dairy cows. Lower urea levels in the milk of the dairy cows may indicate lower N emissions and/or losses from the milk production system (Powell *et al.*, 2011), which would be another benefit for society. A disadvantage for milk producers would be the reduced milk yield and, consequently, reduced revenue. At the magnitude of our experiment, such revenue losses cannot be compensated by savings in concentrate costs.

Table 1. Results (least squares means) of the first six official milk recordings.[1]

	Actual treatment (ACT)	Future treatment (FUT)	Standard error	*P*-values
Milk yield (kg d^{-1})	29.6	27.9	0.76	<0.001
ECM yield (kg d^{-1})	29.1	27.4	0.78	<0.001
Milk fat (g kg^{-1})	40.7	40.3	0.55	0.51
Milk protein (g kg^{-1})	30.6	30.8	0.29	0.54
Lactose (g kg^{-1})	48.1	48.4	0.20	0.005
Milk urea (mg dl^{-1})	19.7	15.9	0.58	<0.001
SCC (log 10 ml^{-1})	4.58	4.59	0.050	0.94

[1] ECM = energy-corrected milk; SCC = somatic cell counts.

Conclusions

Even with herbage rations of an average quality regarding nutritive values, the use of a cereal mixture as the sole concentrate supplementation for dairy cows leads to lower milk yields with similar milk contents and reduced milk urea contents compared to a partial protein concentrate supplementation. The future GMM variant has benefits for society as well as disadvantages for milk producers. Overall, more in-depth investigations are needed to study the interaction between forage CP content and reduced protein concentrate supplementation on milk production, nitrogen losses, animal-welfare and fertility.

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References

Costa A., Egger-Danner C., Mészáros G., Fuerst C., Penasa M., Sölkner J. and Fuerst-Waltl B. (2019) Genetic associations of lactose and its ratios to other milk solids with health traits in Austrian Fleckvieh cows. *Journal of Dairy Science* 102, 4238-4248.

Huthanen P. and Hristov A.N. (2009) A meta-analysis of the effects of dietary protein concentration and degradability on milk protein yield and milk N efficiency in dairy cows. *Journal of Dairy Science* 92, 3222-3232.

Law R.A., Young F.J., Patterson D.C., Kilpatrick D.J., Wylie R.G. and Mayne C.S. (2009) Effect of dietary protein content on animal production and blood metabolites of dairy cows during lactation. *Journal of Dairy Science* 92, 1001-1012.

Münger A., Schori F., and Schlegel P. (2021) Apports alimentaires recommandés pour les ruminants (Livre vert) [Feeding recommandations for ruminants (green book)]. Chapitre 7, Ed. Agroscope, Posieux, Switzerland. Available at: https://www.agroscope.ch/livre-vert.

Powell J.M., Wattiaux M.A. and Broderick G.A. (2011) Short communication: Evaluation of milk urea nitrogen as a management tool to reduce ammonia emissions from dairy farms. *Journal of Dairy Science* 94, 4690-4694.

R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available at: https://www.R-project.org/.

Reist M., Erdin D., von Euw D., Tschuemperlin K., Leuenberger H., Chillard Y., Hammon M., Morel C., Philipona C., Zbinden Y., Kuenzi N. and Blum J.W. (2002) Estimation of energy balance at the individual and herd level using blood and milk traits in high-yielding dairy cows. *Journal of Dairy Science* 85, 3314-3327.

Schori F. (2020) Begrenzung der Proteinzufuhr in der Rindviehfütterung [Limiting the protein supply in cattle Feeding]. *Agroscope Science* 96, 1-96.

Zang Y., Ji P., Morrison S.Y., Koba Y., Grant R.J. and Dann H.M. (2021) Reducing metabolizable protein supply: Effects on milk production, blood metabolites, and health in early-lactation dairy cows. *Journal of Dairy Science* 104, 12443-12458.

Isotopic signatures of topsoil and slurry on dairy farms with differing management and soils

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Abstract

Analysis of isotopic signatures of nitrogen (^{15}N) is a widely used method for identifying and tracking pathways of N losses in agriculture. It is not yet clear how isotopic signatures in soil, plant and manures are related to N balances at the farm level. In the 'Waterbuddies' project, the N fluxes and N balances of 25 dairy farms in Northwest Germany were investigated. We examined, at the farm level, the isotopic signature of manures (slurry) and at the field level the isotopic signatures (^{15}N) of 51 grassland fields (topsoil, aboveground biomass) in three different soil landscapes (moraine land/sandy, peatland/organic, and marshland/clay). We also calculated gross farm gate balances (StoffBilV) and compiled information on farm management. Results show that $\delta^{15}N$ values of topsoil and slurry were significantly higher on farms on marshland than on farms in other soil landscapes. The $\delta^{15}N$ value of the aboveground biomass did not differ among soil landscapes. The isotopic signatures ($\delta^{15}N$ values) of farm slurry and topsoil were only weakly related to the gross N farm gate balances. We found that N transformation processes are influenced by the soil landscape, but still a gap remains between field N dynamics and N balances at the farm level.

Keywords: stable isotopes, natural ^{15}N abundance, dairy farms, farm gate balance

Introduction

Nitrogen (N) is an important driver of agricultural crop production. However, only 47% of the reactive nitrogen added globally to cropland is converted into harvested products (Lassaletta *et al.*, 2014). Thus, there is a need for identifying and tracking pathways of N flows in agricultural practice as a part of food production. Nitrogen occurs naturally as two stable isotopes (^{14}N and ^{15}N) and their natural abundance in plants and soil is regarded as an efficient method for tracking N dynamics and N pathways without disturbing the system (Högberg, 1997; Robinson, 2001). As part of the research project 'Waterbuddies', 25 dairy farms were investigated with regard to their nitrogen flows, for a more efficient nutrient management. We used measurements of $\delta^{15}N$ as a diagnosis tool for providing information on N pathways. The aim was to identify the main factors that influence N flows and N surpluses on the farms and on selected fields. We examined the relationships between isotopic signatures of topsoil, aboveground biomass, slurry, and gross N farm gate balances. We hypothesized that farms with high farm gate balances also have higher $\delta^{15}N$ values in their manures (slurry) and in the topsoil of their fields.

Material and methods

The study area is located in the Wesermarsch, northern Lower Saxony, Germany. The main business of the 25 farms is dairy farming which includes grazing (Table 1). The farms operate on three different soil landscapes: moraine land, peatland, and marshland, which are predominantly characterized by sandy, organic and clay soils, respectively. On a total of 51 grassland sites (mainly permanent grasslands) hand-plucked samples of the aboveground biomass and samples of topsoil (0-10 cm) were taken in November 2019 and analysed for the natural abundance of ^{15}N isotopes ($\delta^{15}N$); at the farm level, slurry was analysed isotopically (^{15}N). The gross N farm gate balance was calculated according to StoffBilV (Federal Ministry of Food and Agriculture, 2017) for 2019: all amounts of N inputs to the farm (e.g. concentrated feed, mineral and organic fertilizers, purchase of animals) and N outputs (e.g. farm manure, animal and plant products – especially milk – and animals) were recorded.

We applied a one-way analysis of variance (ANOVA) to assess the effects of the soil landscapes on $\delta^{15}N$ values in topsoil, plant material and farm manure. This was followed by a Tukey post hoc test (α=0.05) to compare means. To assess the relationship between gross N balances and $\delta^{15}N$ in topsoil, aboveground biomass, and slurry, Pearson correlation analyses were conducted with gross N balance as the independent variable.

Results and discussion

The $\delta^{15}N$ values in the topsoil differed significantly among all soil landscapes ($P<0.001$), with the highest $\delta^{15}N$ values in marshland and the lowest $\delta^{15}N$ values in the peatland soils. The $\delta^{15}N$ values of aboveground biomass did not differ significantly among the soil landscapes ($P=0.237$). The $\delta^{15}N$ values in slurry were highest for farms in the marshland ($P<0.05$; Figure 1). The reason for this is not yet clear. The higher $\delta^{15}N$ values in the topsoil of marshland farms might partly be explained by higher $\delta^{15}N$ values in slurry of marshland farms. The lower $\delta^{15}N$ values in peatland topsoil might be related to smaller amounts of slurry application as there is an enrichment in ^{15}N in organic fertilizers of animal origin compared to synthetic fertilizers (Dittert *et al.*, 1998). Trafficability on peat soils is limited and could lead to smaller amounts of slurry being used on these fields and substituted by synthetic N. Often farms on peat land have access to moraine or marsh fields and might use more slurry there.

The isotopic signatures of topsoil, aboveground biomass and slurry were only weakly related to the gross N farm gate balance (Figure 2). The N surplus of the gross farm gate balances explained only 18, 23 and 27% of the variation in ^{15}N signatures of topsoil, aboveground biomass, and slurry, respectively.

Table 1. Some selected farm data (2019): means with min-max in brackets.

	Moraine land	Peatland	Marshland
Farm size [ha]	182 (50-396)	145 (102-182)	136 (62-290)
Grassland proportion [%]	60 (40-77)	88 (75-99)	88 (78-100)
N farm gate balance [kg ha^{-1}]	156 (68-212)	131 (62-196)	161 (110-273)
Purchase syn. fertilizer N [kg ha^{-1}]	115 (53-156)	96 (0-202)	118 (71-163)

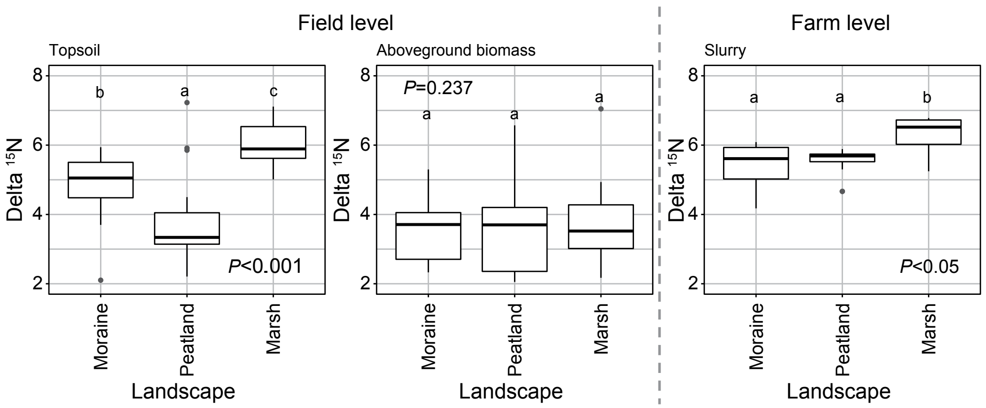

Figure 1. Means of $\delta^{15}N$ values of topsoil, aboveground biomass, and slurry for different regions. Same letters indicate no significant difference ($P<0.05$).

This finding supports results of Wrage *et al.* (2011). The relatively weak explanation of ^{15}N signatures by gross N farm gate balances does not fully confirm our hypothesis and indicates that there are more influencing factors among the varying farm conditions apart from the assignment to a soil landscape (e.g. use and management of single fields; distance farm – field).

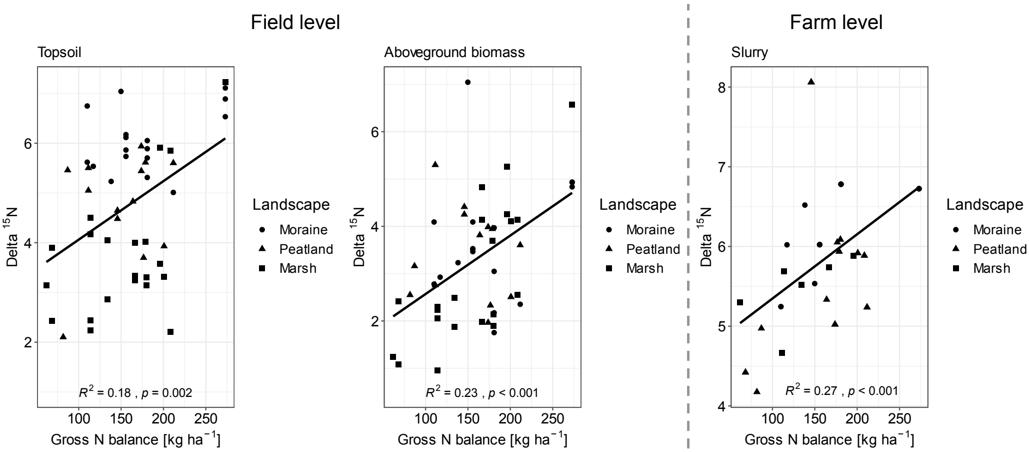

Figure 2. Relationship between gross N farm gate balance (kg ha^{-1}) and $\delta^{15}N$ values of topsoil, aboveground biomass, and slurry. Coefficients of determination (R^2) and *P*-values indicate results of Pearson correlation analysis.

Conclusions

Marshland farms and fields showed higher $\delta^{15}N$ values in slurry and topsoil compared to other soil landscapes. Under the widely varying farming conditions in practice, it remains difficult to establish a direct relationship between gross N farm gate balance and isotopic signatures.

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References

Dittert K., Goerges T. and Sattelmacher B. (1998) Nitrogen turnover in soil after application of animal manure and slurry as studied by the stable isotope ^{15}N: A review. *Zeitschrift für Pflanzenernährung und Bodenkunde* 161, 453-463.

Federal Ministry of Food and Agriculture (2017) Verordnung über die Anwendung von Düngemitteln, Bodenhilfsstoffen, Kultursubstraten und Pflanzenhilfsmitteln nach den Grundsätzen der guten fachlichen Praxis beim Düngen (Düngeverordnung – DüV).

Högberg P. (1997) ^{15}N natural abundance in soil-plant systems. *New Phytologist* 137, 179-203.

Lassaletta L., Billen G., Grizzetti B., Anglade J. and Garnier J. (2014) 50 year trends in nitrogen use efficiency of world cropping systems: the relationship between yield and nitrogen input to cropland. *Environmental Research Letters* 9, 105011

Robinson D. (2001) $\delta^{15}N$ as an integrator of the nitrogen cycle. *Trends in Ecology and Evolution* 16, 153-162.

Wrage N., Küchenmeister F. and Isselstein, J. (2011) Isotopic composition of soil, vegetation or cattle hair no suitable indicator of nitrogen balances in permanent pasture. *Nutrient Cycling in Agroecosystems* 90, 189-199.

Milk production of dairy cows fed grass-clover silage pulp

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Abstract

Silage pulp (SP) is the solid fraction extracted from silage in biorefinery that can be used as forage source for ruminants. There is a lack of information regarding the complete replacement of dietary silage for SP on performance of dairy cows. The purpose of this study was to evaluate the complete substitution of grass-clover silage for SP on the milk yield of dairy cows. A grass-clover mixture was harvested, wilted, and ensiled in bunker silos. The silage was screw pressed in a biorefinery for solid and liquid separation. 72 lactating cows were used in a completely randomized block design, receiving either the silage- or the SP-based diet. The SP-based diet had lower concentrations of water-soluble carbohydrates and crude protein but greater fibre concentration compared to the silage-based diet. Milk yield and energy corrected milk were greater for cows receiving a silage-based diet compared to SP-based diet but first after 10 and 5 weeks of feeding, respectively. Milk composition, body condition score and body weight were not affected by diets. The complete substitution of silage for SP reduced the milk production of dairy cows over time.

Keywords: biorefinery, fibre, forage press cake ruminant, screw press

Introduction

Fibrous pulp is a by-product and its direct comparison with the original forage for animal feeding is meaningless if the diets are not adjusted based on the differences between the forages. The mechanical pressing removes moisture and soluble nutrients, increasing fibre content and decreasing the nutritional value of the pulp compared to the original forage (Savonen *et al.*, 2020). Those differences have to be considered when formulating diets, especially when feeding high-producing dairy cows, because dietary forage neutral detergent fibre (NDF) content is the primary factor limiting intake and performance when high-producing dairy cows are fed forage-based diets (Allen, 2000). However, to our knowledge, no study has evaluated the complete substitution of silage for silage pulp (SP) on the performance of dairy cows. We hypothesized that the mechanical process of pressing the silage would increase fibre digestibility of SP. The greater fibre digestibility could compensate for the increased fibre content of the SP-diet, resulting in similar milk yield (MY) compared to dairy cows fed the silage-based diet. The aim of this study was to evaluate the complete substitution of grass-clover silage for SP on performance of dairy cows in an organic production system.

Materials and methods

The experiment was conducted at the organic dairy farm Sötåsen Agricultural High School, Töreboda, Sweden (N 58° 41', E 14° 8',), during 17 weeks, from 23 November 2020 to 14 April 2021. Grass-clover leys were mown and wilted to a dry-matter (DM) content of 300 g kg^{-1} before being chopped and ensiled in bunker silos. Silage was either used as the silage treatment or pressed through the screw press at 1.5 Mg h^{-1} to produce the SP treatment.

Seventy-two lactating cows (28 primiparous and 44 multiparous) of Holstein (49), Swedish Red (11), Jersey (8), and mixed breeds (Swedish Red × Ayrshire cattle) (4) were used in a completely randomized block design. Cows were blocked based on their breed, lactation number, days in milk (DIM) and energy-corrected milk (ECM) yield and randomly assigned to one of the two treatments (silage- or SP-based diet) within block (n=36). To minimize the difference in intake regulation by filling effect, the experimental diets were formulated aiming to have similar forage NDF concentrations between treatments. However, due to the regulations for organic production (KRAV, 2021) that limit the minimum forage inclusion, the forage NDF concentration of the SP-based diet was still greater than the silage-based diet, even with greater inclusion of silage compared to SP. The silage- and SP-based diets were composed (% of DM) of forage (62.2 vs 52.5); mixed cereals (17.0 vs 16.9); faba bean (5.7 vs 14.7); pellets (14.9 vs 15.2); mineral and vitamin mix (0.2 vs 0.6). The chemical composition of the silage- and SP-based diets were 533 and 671 g DM kg^{-1}; 336 and 376 g NDF; 284 and 310 g forage NDF; 164 and 170 g CP; 81.3 and 58.1 g water soluble carbohydrates per kg DM, respectively.

Forages and concentrates were fed to the cows separately. Silage and SP were mixed with the mineral/vitamin mix in separate total mixed ration mixers and delivered to each group twice a day by an automatic feeding wagon. Concentrate ingredients were individually fed in automatic feed stations placed in each pen. Milk yield was registered twice a day when cows were milked in a fishbone milking parlour. Milk samples were collected at milking in the afternoon and the following morning every other week throughout the experiment. The milk was analysed for fat, protein and lactose, and ECM was calculated according to Sjaunja *et al.* (1991). Body weight (BW) and body condition score (BCS) were recorded (Edmonson *et al.*, 1989) once a month.

MY, ECM, milk components, BW and BCS were analysed using the MIXED procedure of SAS (version 9.4, SAS Institute Inc., Cary, NC) with time as repeated measures. The model included treatment, time, and treatment-by-time interaction as fixed effects, and block and cow within block as random effects. The statistical model was: $Y_{ijkl} = \mu + F_i + T_j + FT_{ij} + B_k + C_l (B_k) + e_{ijkl}$ where Y_{ijkl} is the observed response, μ is the overall mean, F_i is the fixed effect of forage (i = 1 to 2), T_j is the fixed effect of time (j = 1 to 17 for MY; j = 1 to 8 for ECM and milk components; j = 1 to 4 for BCS and BW), FT_{ij} is the interaction between forage and time, B_k is the random effect of block, $C_l (B_k)$ is the random effect of cow within block, and e_{ijkl} is the error term. Means were determined using the least square means statement and treatment means were compared using the PDIFF option with Tukey adjustment. Statistical significance was considered at $P \leq 0.05$ and tendency to significance at $0.05 < P \leq 0.10$.

Results and discussion

When compared to the silage-based diet, the SP-based diet was drier and with greater NDF and forage NDF concentrations, which might have affected DMI and consequently milk production of dairy cows. In the current study, the dietary forage NDF concentration was 9.1% higher in the SP-based diet (310 g kg^{-1} DM) than the silage-based diet (284 g kg^{-1} DM), while MY decreased by 8% and ECM yield by 12% for cows receiving the SP-based diet compared to cows receiving the silage-based diet (Table 1 and Figure 1). In a recent meta-regression study, Allen *et al.* (2019) evaluated the effect of dietary filling factors on lactating cows and observed that dietary forage NDF concentration was negatively correlated with MY. According to Allen (2000), the dietary forage NDF concentration is the main factor limiting intake when intake is regulated by fill capacity of the rumen due to its slow passage rate.

Milk composition between the treatments was similar throughout the study; however, as MY was generally greater for cows fed the silage-based diet, yields of milk components were greater than when cows received the SP-based diet (Table 1). BW and BCS were not affected by the treatments, suggesting that MY was supported by diets only and body reserves were preserved. We hypothesized that the mechanical pressing during silage juice extraction would increase NDF digestibility of the SP, which could compensate for the greater NDF content. However, there was no difference in NDF digestibility between silage and SP (729 vs 721 g kg^{-1} NDF).

Table 1. Performance of cows fed silage- or silage pulp (SP)-based diets (n=36).

Item	Treatment		SEM	*P*-value		
	Silage	SP		Treatment	Time	Treatment × Time
MY, kg d^{-1}	34.0	31.3	1.63	0.165	0.030	0.028
ECM, kg d^{-1}	37.0	32.5	1.72	0.013	0.012	0.093
Composition, %						
Protein	3.44	3.52	0.06	0.240	0.362	0.597
Fat	4.38	4.56	0.14	0.233	0.005	0.598
Lactose	4.90	4.92	0.04	0.754	<0.001	0.232
Yield, kg d^{-1}						
Protein	1.20	1.05	0.05	0.011	0.182	0.398
Fat	1.53	1.36	0.07	0.035	0.001	0.306
Lactose	1.72	1.53	0.10	0.053	0.608	0.206
BCS	2.68	2.73	0.04	0.379	0.195	0.216
BW, kg	679	679	19.1	0.336	0.027	0.506

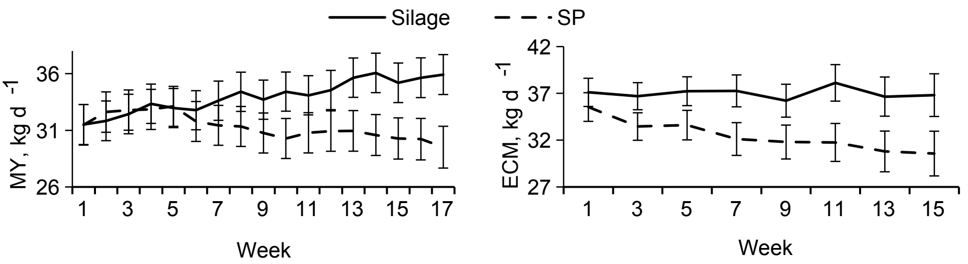

Figure 1. Milk yield (MY) and ECM of cows fed silage- or silage pulp (SP) diets. Error bars indicate SEM.

Conclusions

The mechanical pressing process did not increase fibre digestibility in SP; thus, the complete substitution of silage for SP reduced the milk production of dairy cows over time.

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References

Allen M.S. (2000) Effects of diet on short-term regulation of feed intake by lactating dairy cattle. *Journal of Dairy Science* 83(7), 1598-1624.

Allen M.S., Sousa D.O. and VandeHaar M.J. (2019) Equation to predict feed intake response by lactating cows to factors related to the filling effect of rations. *Journal of Dairy Science* 102(9), 7961-7969.

Edmonson A.J., Lean I.J., Weaver L.D., Farver T. and Webster G. (1989) A body condition scoring chart for Holstein dairy-cows. *Journal of Dairy Science* 72(1), 68-78.

KRAV (2021) *Standards for KRAV-certified Production – 2021 edition*, 324.

Savonen O., Franco M., Stefanski T., Mantysaari P., Kuoppala K. and Rinne M. (2020) Grass silage pulp as a dietary component for high-yielding dairy cows. *Animal* 14(7), 1472-1480.

Sjaunja L.O., Baevre L., Junkkarinen L., Pedersen J. and Setala J. (1991) A Nordic proposal for an energy corrected milk (ECM) formula. *Performance Recording of Animals: State of the Art*, 1990, 50, 156-192.

Ruminal *in vitro* vs *in vivo* digestion using different silage, barley and inoculum types

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Abstract

Accurate quantitative information on digestibility and methane production of dairy cow diets is essential to improve the animal efficiency and to reduce the negative environmental impact of the dairy sector. *In vitro* rumen fermentation methods are fast, cost-effective, and allow the study of a large number of samples simultaneously, but their accuracy has been criticized. The objectives of this experiment were to evaluate the effects of grass silages differing in fermentation characteristics and dried vs crimped and ensiled barley grains on digestibility and methane production potential *in vitro* using two types of inoculums, and to directly validate the *in vitro* against the *in vivo* results. There was no significant effect of diets on *in vitro* organic matter degradability (771±12.2 mg g^{-1}), gas (235.4±10.71 ml g^{-1}), and methane production (20.3±1.39 mg g^{-1}) in line with the *in vivo* data. The organic matter *in vitro* degradability was significantly higher compared with apparent *in vivo* total tract digestibility (771 vs 724 mg g^{-1}) and the methane production was significantly lower (20.3 vs 30.2 mg g^{-1}). The results suggest that the tested *in vitro* method can be used for screening the samples for degradability and their potential methane production.

Keywords: *Hordeum vulgare*, high-moisture grain, methane, digestibility, silage additive

Introduction

Accurate quantitative information on digestibility and methane production of dairy cow diets is essential to improve the animal efficiency and to reduce the negative environmental impact of the dairy sector. Experiments with animals give the most reliable data but they are expensive and time consuming. Thus, the importance of *in vitro* methods is constantly increasing. To confirm the biological and scientific value of the *in vitro* data, the *in vitro* method should be validated against *in vivo* results.

Additives are commonly used to facilitate good preservation of ensiled grains and forages. The most common additives are based on selected lactic acid bacteria (LAB) inoculants, but in some areas organic acids are also commonly used. The additives based on formic acid effectively restrict silage fermentation, while LAB is used to accelerate and direct the lactic acid fermentation in the silage. These two types of additives induce specific changes in silage composition, with relatively high water-soluble carbohydrate concentration and low lactic acid and volatile fatty acid concentrations in silages based on formic acid additive, with the opposite being the case for LAB silages (McDonald *et al.*, 1991; Muck *et al.*, 2018). It has been shown that the differences in the silage composition affect rumen fermentation and milk composition of dairy cows (Jaakkola *et al.*, 2006; Shingfield *et al.*, 2002).

The diets of donor animals may also affect the degradability and methane production *in vitro,* since the major factor affecting the microbial population in the rumen and the microbial activities of inoculum are the nutrient intake and the diet composition (Mould *et al.*, 2005a,b). The main objective of this study was to evaluate the effects of grass silages differing in fermentation characteristics and dried vs crimped ensiled barley grain on degradability and methane production potential *in vitro*. The second objective was to perform direct comparison of the *in vitro* results with results of an *in vivo* study conducted simultaneously. In addition, the effects of the diet of inoculum donor animals on *in vitro* degradation were tested.

Materials and methods

The grass was ensiled using either a formic and propionic acid-based silage additive (FAPA) or a homofermentative LAB inoculant. Barley grain was used as dried or crimped and ensiled. The detailed description and the results of the *in vivo* study will be reported elsewhere. In brief, the feeds were given in a 2×2 factorial arrangement to 4 Nordic Red dairy cows. Four experimental diets were fed as total mixed rations comprising 0.50 grass silage, 0.275 concentrate, and 0.225 barley grain on dry matter (DM) basis as follows: (1) AD (FAPA grass silage and dry barley); (2) AE (FAPA grass silage and crimped ensiled barley); (3) ID (LAB inoculated grass silage and dry barley); and (4) IE (LAB inoculated grass silage and crimped ensiled barley). The *in vivo* apparent total tract digestibility was measured by total faeces and urine collection, and methane emission was measured in calorimetric chambers.

In the *in vitro* study the same diets were used, and the effect of the diet of the inoculum donor animals on degradability, gas and methane production *in vitro* were tested, resulting in a study design with two grass silages, two barley grains, and two types of inoculums (type one inoculum: four inoculums from cows on the same diet as tested (SA), type two inoculum: standard inoculum (ST)). The rumen liquid for inoculum was collected using stomach tubing. For the standard inoculum, rumen liquid was collected from three cows at different stages of lactation receiving a standard diet based on grass silage and concentrate (50:50). In addition, inoculums from the donor cows fed the experimental diets in the *in vivo* study were collected.

The rumen fluid was filtered through two layers of cheese cloth and mixed with buffer (60:40) to create inoculums. The *in vitro* incubations were carried out in 250 ml glass flasks under anaerobic conditions at 39 °C in a water bath equipped with constant shaking. There were four *in vitro* runs with four flasks with the same treatment within each run. Two of the flasks were used for total gas production measurements using the Ankom modules, and the gas production method was described in detail by Rinne *et al.* (2016). The remaining two flasks were used for measurements of methane concentrations. A modification of the gas production method enabled samples of gas to be taken for methane measurements. In brief, to measure the methane concentration, the Ankom module was replaced with a rubber plug with a silicone hose, and the gas produced during incubation was collected into air-tight gas collection bags. In the flask for total gas measurement, 0.5 g of DM of sample was incubated in 60 ml of buffer and 20 ml of inoculum for 72 h. To increase the weight of the residue for degradability measurements after fermentation, 1 g of DM samples with 120 ml of buffer and 40 ml of inoculum were incubated for 48 h in the flasks for methane measurements. To measure the degradability, the residues from flasks for methane measurements after 48 h were quantitatively transferred to small Ankom fibre analysing bags, dried, and analysed. The data was analysed using a MIXED procedure of SAS with inoculum and diet as fixed effects and run as random effect.

Results and discussion

There were no significant differences between inoculum types on OM residue after fermentation, gas, or methane production from inoculum only (blank). Also, there were no significant effects of diet and inoculum type (except the SA inoculum from ID diet that had lower OM degradability) on OM degradability, gas, or methane production *in vitro* (Table 1) which is consistent with the *in vivo* results indicating no effects of diet on apparent total tract diet digestibility and methane production. The comparison of *in vivo* and *in vitro* trials showed that the OM digestibility was lower and methane production was higher in the *in vivo* experiment (Table 2). The correlation coefficient R^2 for OM digestibility was 0.330 and for methane production 0.312. The lower OM digestibility *in vivo* is at least partially explained by the endogenous and metabolic OM (apparent digestibility) that is not produced *in vitro*.

Table 1. Effect of inoculum and diet type on degradability of organic matter, gas and methane production *in vitro*.[1]

									SEM	*P*-value		
Diet (D)	AD	AD	AE	AE	ID	ID	IE	IE		D	I	D×I
Inoculant (I)	ST	SA	ST	SA	ST	SA	ST	SA				
DOM, mg g^{-1}	778	765	774	773	775	755	773	777	5.3	0.34	0.05	0.13
Gas prod., ml g^{-1} OM	237	237	244	239	230	229	238	231	6.0	0.21	0.38	0.87
CH_4, mg g^{-1} DOM	20.1	20.8	20.5	20.2	20.6	20.2	20.3	19.6	0.74	0.82	0.73	0.68

[1] DOM = degraded organic matter; OM = organic matter; AD = organic acid-treated grass silage and dry barley; AE = organic acid-treated grass silage and crimped ensiled barley; ID = lactic acid bacteria treated grass silage and dry barley; IE = lactic acid bacteria treated grass silage and crimped ensiled barley; ST = inoculum from donor animals on standard diet; SA = inoculum from donor animal on the same diet as tested; SEM = standard error of the mean.

Table 2. Direct comparison of *in vivo* and *in vitro* organic matter digestibility and methane production across diets.1

	Method			SEM	
	In vivo	*In vitro*, ST	*In vitro*, SA		*P*-value
DOM, mg g^{-1}	724	775	767	3.3	<0.001
CH_4, mg g^{-1} DOM	30.2	20.3	20.4	0.33	<0.001

[1] ST = inoculum from donor animals on standard diet; SA = inoculum from donor animal on the same diet as tested; DOM = digested organic matter; SEM = standard error of the mean.

Conclusions

Performing direct validation of *in vitro* results against *in vivo* data is essential to validate the *in vitro* systems. There was no significant effect of diets on *in vitro* and *in vivo* OM digestibility and methane production data, indicating that the *in vitro* system can be used to screen the samples for degradability and methane production potential. However, the lack of difference between the dietary treatments in OM degradability and methane production in the current data set did not allow to evaluate if the *in vitro* system can rank the feeds similarly as the *in vivo* data, and to test the effect of the inoculum type on the *in vitro* fermentation.

References

Jaakkola S., Kaunisto V. and Huhtanen P. (2006) Volatile fatty acids proportions and microbial protein synthesis in the rumen of cattle receiving grass silage ensiled with different rates of formic acid. *Grass and Forage Science* 61, 282-292. https://doi.org/10.1111/j.1365-2494.2006.00532.x.

McDonald P., Henderson A.R. and Heron S.J.E. (1991) *The Biochemistry of Silage*, 2nd ed. Chalcombe Publications, Marlow, UK, pp. 340.

Mould F.L., Kliem K.E., Morgan R. and Mauricio R.M. (2005a) In vitro microbial inoculum: a review of its function and properties. *Animal Feed Science and Technology* 123-124, 31-50.

Mould F.L., Morgan R., Kliem K.E. and Krystallidou E. (2005b) A review and simplification of the *in vitro* incubation medium. *Animal Feed Science and Technology* 123, 155-172.

Muck R.E., Nadeau E.M.G., McAllister T.A., Contreras-Govea F.E., Santos M.C. and Kung L. Jr. (2018) Silage review: Recent advances and future uses of silage additives. *Journal of Dairy Science* 101, 3980-4000. https://doi.org/10.3168/jds.2017-13839

Rinne M., Kautto O., Kuoppala K., Ahvenjärvi S., Willför S., Kitunen V., Ilvesniemi H. and Sormunen-Cristian R. (2016) Digestion of wood-based hemicellulose extracts as screened by *in vitro* gas production method and verified *in vivo* using sheep. *Agricultural and Food Science* 25, 13-21.

Shingfield K.J., Jaakkola S. and Huhtanen P. (2002) Effect of forage conservation method, concentrate level and propylene glycol on diet digestibility, rumen fermentation, blood metabolite concentrations and nutrient utilisation of dairy cows. *Animal Feed Science and Technology* 97, 1-21. https://doi.org/10.1016/S0377-8401(02)00006-8

Yield response of grass and grass-clover leys in crop rotations to phosphorus fertilization

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Abstract

Grass or grass-clover leys are important parts of many crop rotations, providing forage for livestock and exerting positive effects on soil structure and fertility. One of the most important nutrients for optimum ley and especially clover growth is phosphorus (P). Based on eight Swiss and Danish long-term field experiments, we examined the response of grass and grass-clover yields to up to 26 years of varying P fertilization rates, the development of soil test P (STP) values and potential effects of clay content, organic carbon content and pH of the soil, average temperature and precipitation. While different fertilization rates often resulted in significantly different STP values, yields mostly showed significant differences between unfertilized and fertilized treatments, but not between treatments with different fertilizer amounts. Increasing soil clay contents increased relative yields in relation to Olsen-P values.

Keywords: soil test phosphorus, P fertilizer, long-term field experiments, grass, clover

Introduction

Phosphorus fertilizer recommendations often have the long-term aim to either build up soil P content to ensure a certain soil test P (STP) value required for achieving optimal yields, or in the case of high STP levels, to reduce these by fertilizing with less P than exported with the crop (Jordan-Meille *et al.*, 2012). In addition to STP values and fertilizer rates, crop type, soil properties and climatic factors may influence crop yield response (Hirte *et al.*, 2021). Corresponding previous studies on effects on grass and clover yields were mostly conducted on permanent grassland, while studies concerning arable land have mainly focused on other crops. We hypothesize that the influences listed above also affect the yield of grass and grass-clover leys in crop rotations. Therefore, the objectives of this paper are to examine the effects of varied long-term P fertilization on STP and yield and to clarify if these effects depend on general soil characteristics and climate.

Material and methods

We used data on STP and crop yield response to different rates of P fertilization from four Danish (Aarhus University: Borris, Højer, Ødum and Rønhave; Rubæk and Sibbesen, 2000) and four Swiss (Agroscope: Ellighausen, Oensingen, Rümlang-Altwi, Zurich-Reckenholz; Hirte *et al.*, 2021) long-term field experiments including grass or grass-clover leys in their crop rotations. Other crops in the rotations were various cereals, beets, legumes, rapeseed and potatoes. STP values given as H_2O-CO_2-P (extraction with CO_2-saturated water; Dirks and Scheffer, 1930) were converted to Olsen-P (extraction with 0.5 M sodium bicarbonate; Olsen *et al.,* 1954): Olsen-P = 15.78 × H_2O-CO_2-P (mg kg^{-1} soil; Neyroud and Lischer, 2003).

The different fertilizer rates (three + unfertilized treatment for the Danish, five + unfertilized treatment for the Swiss experiments) were aligned by classifying according to average relative fertilizer amounts (ARF = fertilizer (kg P ha^{-1}) / export (kg P ha^{-1})). This resulted in 4 classes: without (ARF = 0), low

(ARF = 0.32-0.64), balanced (ARF = 0.91-0.96) and excessive (ARF = 1.28-1.60; only present in the Swiss experiments). The different fertilizer treatments were compared for all years combined as well as in year-groups (years 1-8, 11-18 and 21-26 of the experiments; not all experiments represented in all groups) to portray the development of differences over time.

All analyses were conducted using R (R Core Team, 2021). Due to a lack of normal distribution and pairing of samples in experiment-year combinations, the Friedman and Nemenyi tests were used for comparisons (significance level of 0.05). A Mitscherlich type model based on Olsen-P was fitted to predict relative yield (RY = $Yield_{without, low or excessive}$ / $Yield_{balanced}$). Considered covariates were relative fertilizer amount, clay content (%), organic carbon content (%) and pH ($CaCl_2$) as well as the long-term mean temperature (°C) and mean annual precipitation (mm) of the experimental sites. To avoid overfitting, covariates were initially tested individually. Afterwards covariates with significant effects were tested in groups of three or less.

Results and discussion

Comparisons of Olsen-P values and yields between the fertilization classes 'without', 'low' and 'balanced' are shown in Figure 1A and 1B, respectively. As expected, mean Olsen-P values increased with increasing fertilization. Differences were significant except between class 'low' and the other classes in the groups of experimental years 11-18 and 21-26. Yields significantly differed only between the class 'without' and the fertilized classes, while 'low' and 'balanced' did not show significant differences in any of the groups of experimental years. In years 11-18, the significance of differences between fertilization classes matches between Olsen-P and yield. Separate analyses of the Swiss data including the class 'excessive' did not show any significant yield differences between 'excessive' and 'balanced'. The same is true for comparisons between 'excessive' and 'low' for all groups except all years combined. This demonstrates that relative fertilization rates that are far below amounts export by the crop (in this case 0.32-0.64) usually exert no disadvantage of yield response. Similar results were found by Valkama *et al.* (2009), where even the lowest rates of P fertilization led to maximum yield response for grass mixtures.

We expected that Olsen-P and yield differences between treatments increase with the number of experimental years; however, this was not confirmed. Yields in the group of 21-26 experimental years were even similar among treatments. A possible explanation is that the experiments with data for those years had relatively high Olsen-P values in the class 'without' or relatively low values in the class 'balanced'. These values might be based on experiment-specific Olsen-P development combined with fluctuation of extractable soil P among years, created by weather conditions. Effects of varying crops or duration of grass and clover cultivation (one to three consecutive years) have not been examined, but cannot be ruled out.

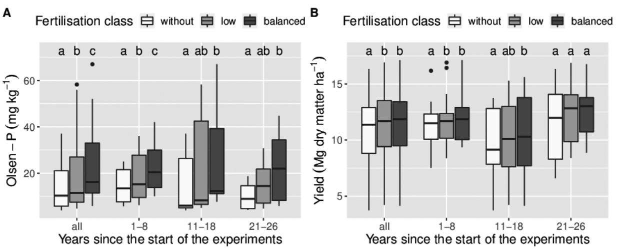

Figure 1. Olsen-P (A) and yield (B) of the different fertilization classes grouped by years since the start of the experiment. All: n=33 per class; 1-8: n=15 per class; 11-18: n=10 per class; 21-26: n=8 per class. Different letters indicate significant differences (P<0.05) between fertilization classes in the same group of experimental years (Nemenyi test).

For modelling of RY, the inclusion of clay content (%) alone as a covariate was the best option (RY = 100 × (1 – exp ((0.04 – 0.02 × clay) × Olsen-P)); Residual standard error: 6.8%). Here, low clay contents led to higher Olsen-P values necessary for near 100% of RY, in accordance with, e.g. the findings by Morel *et al.* (2000) for maize. The inclusion of the other considered covariates did not improve the model.

Conclusions

Even though many years of different fertilizer application rates led to significant differences in Olsen-P, the yield response varied between unfertilized and fertilized treatments only, with few exceptions. This lack of differences in yield response between low, balanced and often even excessive fertilizer rates implies that low fertilizer rates are sufficient for optimal yields. Relative yields in relation to Olsen-P values increased with increasing soil clay contents.

Acknowledgements

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References

Dirks B. and Scheffer H. (1930) Der Kohlensäure-Bikarbonatauszug und der Wasserauszug als Grundlage zur Ermittlung der Phosphorsäurebedürftigkeit der Böden. *Landwirtschaftliche Jahrbücher* 71, 73-99 (in German).

Hirte J., Richner W., Orth B., Liebisch F. and Flisch R. (2021) Yield response to soil test phosphorus in Switzerland: Pedoclimatic drivers of critical concentrations for optimal crop yields using multilevel modelling. *Science of the Total Environment* 755, 143453.

Jordan-Meille L., Rubæk G.H., Ehlert P.A.I., Genot V., Hofman G., Goulding K., Recknagel J., Provolo G. and Barraclough P. (2012) An overview of fertilizer-P recommendations in Europe: soil testing, calibration and fertilizer recommendations. *Soil Use and Management* 28, 419-435.

Morel C., Tunney H., Plénet D. and Pellerin S. (2000) Transfer of phosphate ions between soil and solution: Perspectives in soil testing. *Journal of Environmental Quality* 29, 50-59.

Neyroud J.-A. and Lischer P. (2003) Do different methods used to estimate soil phosphorus availability across Europe give comparable results? *Journal of Plant Nutrition and Soil Science* 166, 422-431.

Olsen R., Cole C.V., Watanabe F.S. and Dean L.A. (1954) *Estimation of available phosphorus in soils by extraction with sodium bicarbonate*. USDA Circular No. 939 USDQ, Government printing office, Washington, DC, USA.

R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing. Vienna. Austria. Available at: https://www.R-project.org/.

Rubæk G.H. and Sibbesen E. (2000) *Long term phosphorus fertilization – Effects on crop yield and soil phosphorus status*. Danish institute of Agricultural Sciences, Tjele, Denmark, 43 pp.

Valkama E., Uusitalo R., Ylivainio K., Virkajärvi P. and Turtola E. (2009) Phosphorus fertilization: A meta-analysis of 80 years of research in Finland. *Agriculture, Ecosystems & Environment* 130, 75-85.

Satellite-based estimation of herbage mass: comparison with destructive measurements and UAV model's estimation

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Abstract

Regular estimation of herbage mass (HM) is a prerequisite for efficient pasture management. In addition to classical estimation using rising plate meters, remote-sensing methods using unmanned aerial vehicles (UAV) or satellites are available. Pasture.io has developed a model that estimates HM based on daily satellite data, herbage growth models and herbage-ingested input data recorded by farmers combined with artificial intelligence. This study compared the accuracy of Pasture.io HM estimations with UAV estimations and destructive measurements. Pastures from three Swiss farms were assessed regularly in May, June and July 2021. It was found that Pasture.io estimates HM with an error value RMSE 342 kg dry matter (DM) ha^{-1} while the UAV model's estimation showed a higher RMSE of 447 kg DM ha^{-1}. The results suggest that even in small pasture structures (mean paddock size: 1.2 ha), it is possible to estimate HM with reasonable accuracy based on satellite data and artificial intelligence.

Keywords: grassland, artificial intelligence, pasture, spectroscopy, remote sensing, pasture-based agriculture

Introduction

Grasslands comprise a large part of the Swiss territory. Although some farmers use tools, such as rising plate meters (RPM) to estimate herbage mass (HM) for pasture management, there is still considerable potential for optimization. New technologies have brought new estimation methods, including aerial photos taken by unmanned aerial vehicles (UAV) (Sutter *et al.* 2021) or satellites. The Pasture.io platform already offers comprehensive support for pasture management worldwide, including daily satellite overflights of pasture areas and estimations of herbage growth and HM on pastures. These estimations are based on artificial intelligence. The platform therefore promises the next level of automation in pasture management. Our study aimed to test this tool under practical conditions on Swiss dairy farms and compare it to RPMs and UAVs.

Materials and methods

Four paddocks on three different dairy farms were studied in May, June and July 2021. The pastures were measured weekly with an RPM and Pasture.io estimated the HM using artificial intelligence. The input data for the Pasture.io model are satellite data, weather data and the amount of herbage grazed. Herbage growth curves from previous years and similar sites were provided to enable Pasture.io to estimate HM. The Pasture.io model corrects its HM estimations based on the amount of herbage ingested by the grazing animals. Therefore, each day the farmers recorded which paddock was grazed on the Pasture.io platform. Herbage intake was estimated by the platform based on the number of animals, their milk yield, and the supplementary feeding in the barn.

In addition to RPM measurements and Pasture.io estimations, the areas were flown over in the same period with a UAV, and the HM estimated using a random forest model as described in Sutter *et al.* (2021).

The two HM estimation methods were compared with field measurements. Five experimental plots per paddock were cut with a lawnmower to height of 5 cm over an area of at least 1 m^2 and the mown herbage was dried at 105 °C for 48 hours to calculate the dry mass per hectare. This was done at a random time during regrowth. The mean value of these five measurements was defined as the paddock's dry matter yield (DMY). A total of 54 DMY measurements was available for the study. Pasture.io HM estimations were also available for these areas. Due to the prevailing weather conditions, not all paddocks could be surveyed with the UAV before field measurements were taken. There were therefore only 27 UAV HM estimations.

Results and discussion

The DMY measurements obtained from the experimental plots ranged from 322 to 2,225 kg dry matter (DM) ha^{-1}. The root-mean-square error (RMSE) for the Pasture.io estimations was 342 kg DM ha^{-1}, corresponding to a normalised root-mean-square error (NRMSE) of 39% (Figure 1A). A similar approach by Askari *et al.* (2019) based on the Sentinel-2 satellite data resulted in a RMSE of 600 kg DM ha^{-1} or NRMSE of 32%, thus achieving comparable values to our Pasture.io results. However, unlike Pasture.io, Askari *et al.* (2019) did not use artificial intelligence and additional input data. Our study was limited to the three months mentioned. However, since the Pasture.io model works with input data and artificial intelligence, it improves with increasing input data. In order to capture this development, investigations over several years would be necessary.

Estimation by UAV resulted in an RMSE of 447 kg DM ha^{-1} (NRMSE = 48%; Figure 1B). The deviation of HM estimation by this method was thus substantially higher, as observed in previous trials (Sutter *et al.*, 2021). One possible explanation for the poorer performance of the model could be related to the botanical composition of the pastures. Most of the pastures consisted of semi-natural multi-species grassland, whereas the model was trained on pastures with fewer species. It is also noticeable that both the estimations by Pasture.io and with the UAV model show the greatest deviations from the measurements on 4 July (Figure 1, squares). It is a great challenge to define five representative locations within a pasture area of >1 ha for field measurements. Unlike field measurements, the two models always estimate HM based on data from the entire pasture area. It is therefore possible that the field measurements were not

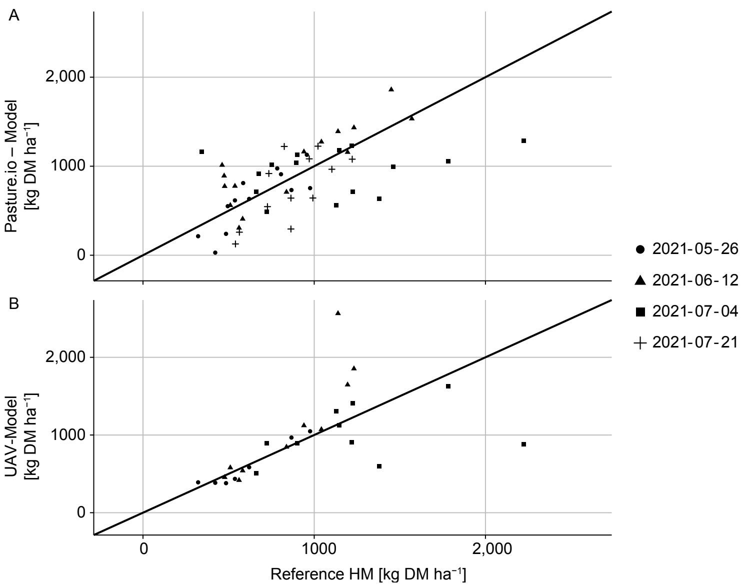

Figure 1. Comparison of the two herbage mass (HM) estimation methods with values measured in the field by cutting and weighing in May, June and July 2021 on pastures. The available HM was measured with a post-grazing height of 5 cm. (A) shows the estimation using the Pasture.io platform (n=54) and (B) shows the estimation using the UAV model (n=27).

representative enough for the whole area. Heterogeneity increases during exclusive grazing of areas within the pasture, which would support the hypothesis of a lack of representativeness in the field measurements.

Pasture.io was also compared to RPM measurements and both methods were found to estimate HM with similar RMSE (data not shown).

Conclusions

The average paddock size in the study was 1.2±0.46 ha. It thus seems possible to estimate HM adequately using satellite data, even on small farms as in Switzerland. However, further improvements of the Pasture.io model should be investigated within studies covering a more extended period.

References

Askari M.S., McCarthy T., Magee A. and Murphy D.J. (2019) Evaluation of grass quality under different soil management scenarios using remote sensing techniques. *Remote Sensing* 11(15), 1-23.

Sutter M., Aebischer P. and Reidy B. (2021) Estimating grassland biomass using multispectral UAV imagery, DTM and a random forest algorithm. *Grassland Science in Europe* 26, 71-73.

Combination of cattle slurry and mineral N fertilizer for efficient grass production in Finland

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Abstract

Fertilization of forage grasses using cattle slurry and mineral N fertilizers is a common nitrogen (N) management strategy on Finnish dairy farms. However, studies concerning yield response to N fertilization and N balances are mainly based on mineral N only. N fertilization responses on mineral soils were studied using three slurry application strategies (no slurry, slurry for the second harvest, and slurry for the first and the second harvest). Each of these strategies included five N fertilization levels as subplots from 0 to 450 kg N ha^{-1} $year^{-1}$. Two experiments were conducted during 2019-2021 with pure timothy (Site 1) and a timothy-meadow fescue-mixture (Site 2). Due to dry periods during each of the three growing seasons, the highest dry matter (DM) yields were moderate (10 Mg DM ha^{-1} $year^{-1}$). The yield gap between mineral N and slurry + mineral-N combination was small in these conditions. The use of slurry increased the soluble N and the total N balances.

Keywords: cattle slurry, grass, nitrogen, nutrient balance, residual effect, silage

Introduction

Due to the long winter season, Finnish milk and beef production is based on high-quality grass silage. Yield response of forage grasses to mineral N fertilization has increased during the past few decades in Finland (Termonen *et al.*, 2020). However, a more typical method on cattle farms, the combination of cattle slurry and mineral fertilizers is less studied. The aim of this study was to compare the effects of mineral and organic N fertilization on yield production, yield CP concentration, N yield and N balances of forage grasses cultivated for silage.

Materials and methods

The experiment was carried out as a split plot design with four replicates at two study sites in Finland: in North Savo (Site 1, 63°09'N, 27°20'E, 2019-2021, loam, soil organic matter (SOM) 3.4%) and in North Ostrobothnia (Site 2, 64°41' N, 25°9' E, 2020-2021, sandy loam, SOM 17.5%). Study sites were established in 2018 on fields cultivated for several years without organic fertilizers. The grass at Site 1 was timothy (*Phleum pratense* L., cv. 'Nuutti') and at Site 2 a mixture of timothy-meadow fescue (cv. 'Nuutti'; *Festuca pratensis* Huds, cv. 'Valtteri'). Fertilization treatments are described in Figure 1. Slurry was spread to a depth of 5-7 cm using a plot-sized disc slurry injector. The C:N ratio of slurry was approximately 16. The total N level of 'slurry + slurry' exceeded the allowed N rate from organic sources in the Nitrates Directive in Finland (170 kg N ha^{-1} y^{-1}). P and K fertilizer applications were increased along with the N rate to at least at the level of Finnish recommendations. Dry matter yields (kg DM ha^{-1}) were measured, and crude protein (CP) concentrations (g kg^{-1} DM) were determined by NIRS (Valio Ltd) for each harvest. Statistical analysis was calculated using GLIMMIX-procedure of SAS 9.4, managing sol-N fertilization as a continuous variable. The main plot, sol-N fertilization and their interaction were fixed variables while replicate × main plot -interaction was a random variable. Depending on variable, sol-N fertilization^2 and sol-N fertilization^3 were added to the model. Pairwise comparisons were calculated in sol-N levels 250 and 450 kg ha^{-1}. Sites and years were analysed separately. The experiment was funded mainly by the European Agricultural Fund for Rural Development ('Production Resilience from Grass – VarmaNurmi'-project).

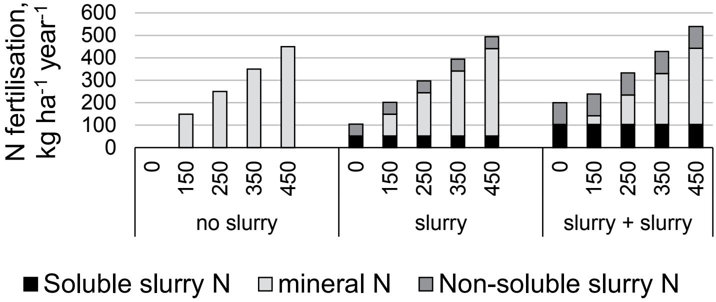

Figure 1. N fertilization provided under the different treatments.

Results and discussion

The DM yields at the sol-N level 450 kg ha^{-1} (Figure 2) were much lower than yields reported by Termonen *et al.* (2020) for a similar sol-N rate. In 2019 at Site 1, 'no slurry' produced significantly higher DM yield than both slurry strategies at sol-N level 250 kg ha^{-1} (the maximum allowed rate in the Nitrates Directive in Finland). This is probably due to lower nutrient use efficiency of slurry during dry growing season, which may have reduced organic N mineralization, increased NH_3 volatilization and/or damage caused to plants by slurry injection. However, a short study period without history of organic fertilizers at study sites can underestimate the fertilizer value of slurry (Schröder, 2005). In 2020 and 2021 at Site 1, no differences between main plots were observed even when the growing seasons had hot and dry periods, which indicates mineralization of organic N reserves from previous year's slurry application and soil. At Site 2 higher SOM content, which indicates higher soil released N, led to lower yield response to N fertilization than at Site 1 (Figure 2). At Site 2 the DM yield of 'no slurry' was significantly higher compared to the 'slurry + slurry' in 2020 and 2021 at the sol-N level 250 kg ha^{-1} but no difference was observed at sol-N level 450 kg ha^{-1}, where the rate of mineral-N was more than 300 kg ha^{-1} of total sol-N. No other differences in the DM yields were detected at these two sol-N levels.

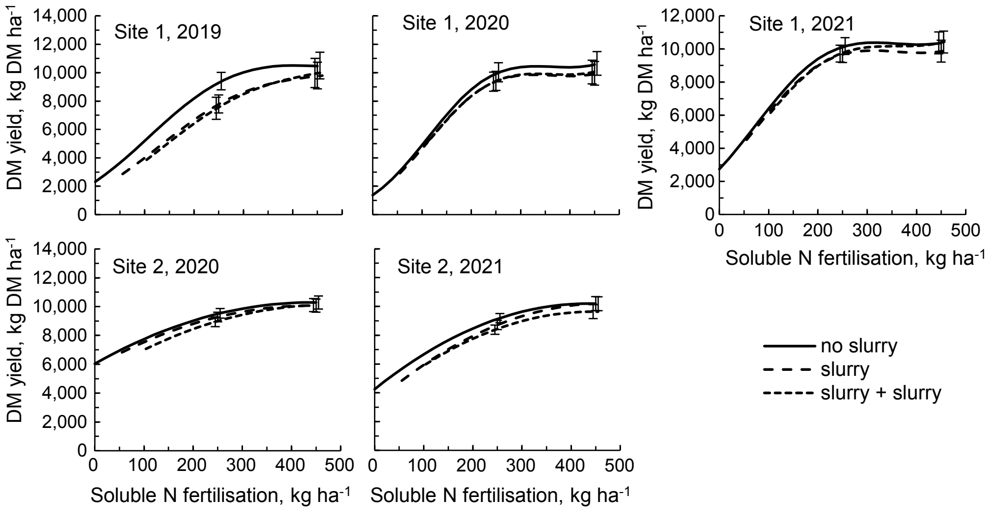

Figure 2. Total yields (kg DM ha^{-1} y^{-1}) in 2019, 2020 and 2021 at Site 1 and in 2020 and 2021 at Site 2. Slurry = slurry for the second harvest. Slurry + slurry = slurry for the first and the second harvest. Soluble N = NH_4^+-N + NO_3^--N. Error bars are 95% confidence intervals for soluble N fertilization levels 250 and 450 kg ha^{-1} y^{-1}.

N fertilization increased CP content and N yield (Table 1). Slurry application significantly increased CP content only at Site 1 in year 2021 when the 'slurry' had higher CP content (143 g kg^{-1} DM) than other strategies (136-138 g kg^{-1} DM) at sol-N level 450 kg ha^{-1}. At Site 2, 'no slurry' produced higher CP content compared to both slurry strategies in 2020 at sol-N 250 kg ha^{-1} and 'slurry + slurry' at sol-N 450 kg ha^{-1}. At Site 1 differences in N yield were compatible with the DM yields but at Site 2 there were some dissimilarities. 'No slurry' produced the lowest sol-N balance especially in 2019 at Site 1 and in 2020 at Site 2. Tot-N balance was always the highest at 'slurry + slurry' strategy and the smallest at 'no slurry' strategy. However, the high total N surplus does not necessarily lead to increased N leaching when using within the growing season (Salo and Turtola, 2006).

Table 1. Ranges of crude protein (CP) content, N yield, sol-N (NH_4^+-N + NO_3-N) and tot-N balances at Site 1 2019-2021 and Site 2 2020-2021 at sol-N levels 250 kg and 450 kg.

		250 kg sol-N			450 kg sol-N		
		no slurry	slurry	slurry + slurry	no slurry	slurry	slurry + slurry
CP	g kg^{-1} DM	130-153	129-143	129-141	160-185	161-182	158-179
N yield	kg ha^{-1}	190-242	158-222	156-215	272-318	252-310	252-289
Sol-N balance	kg ha^{-1}	19-72	44-100	58-103	57-147	49-158	57-164
Tot-N balance	kg ha^{-1}	19-72	97-136	152-183	57-147	99-195	165-266

Conclusions

The dry periods kept the maximum yields moderate. The yield gap between mineral N and slurry + mineral-N combination was small and narrowed after the first study year, which indicates that slurry can be an effective part of N fertilization on high-yielding forage grasses. The amount of slurry (one or two applications per growing season) had only a minor effect on the DM yield. However, the use of slurry increased the sol-N and the total N balances compared to mineral N use, which can cause an environmental risk under high application rates. Assessment of the N leaching risk would have required measurements of leaching.

References

Salo T. and Turtola E. (2006) Nitrogen balance as an indicator of nitrogen leaching in Finland. *Agriculture, Ecosystems and Environment* 113, 98-107.

Schröder J. (2005) Revisiting the agronomic benefits of manure: a correct assessment and exploitation of its fertilizer value spares the environment. *Bioresource Technology* 96, 253-261.

Termonen M., Korhonen P., Kykkänen S., Kärkönen A., Toivakka M., Kauppila R. and Virkajärvi P. (2020) Effects of nitrogen application rate on productivity, nutritive value and winter tolerance of timothy and meadow fescue cultivars. *Grass and Forage Science* 75, 111-126.

Technical, economic and environmental performances of two contrasting dairy systems

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Abstract

A trial was undertaken at Trevarez experimental farm (Brittany, France) from 2013 to 2017 to assess the technical, economic and environmental consequences of two contrasting dairy production systems. The two systems implemented were representative of those found in westerm France: one based on maize silage (S1; 0.15 ha of grazable area per cow), one based predominantly on grazing (S2; 0.4 ha of grazable area per cow). Both systems were run simultaneously after final allocation of fields and dairy cows. Each system involved 60 cows and 60 hectares. On average, the dairy cows from system S1 produced 8,162 kg of milk per year with 118 g of concentrate per kg milk, of which 7,551 litres (l) were sold with a feeding cost of €79 per 1000 l sold. On average, the dairy cows from system S2 produced 7,608 kg of milk per year with 91 g concentrate per kg, of which 7,167 l were sold with a feeding cost of €58 per 1000 l sold. The Farm Gross Surplus of the S2 system was on average €28 per 1000 l higher than from system S1. The surplus of the N balance of S2 was lower by 23 kg per ha compared to S1, the net carbon footprint being close but lower for S2. These results indicate that if well managed, both systems may be both productive and environment friendly.

Keywords: dairy cows, production systems, carbon footprint, sustainability

Introduction

Since the end of milk quotas, farmers are looking for options to reach the triple performance leading to sustainability: the economic (optimization of feeding cost), social (quality of life, workload) and environmental (nitrogen discharge, greenhouse gas (GHG) emissions) performances. To evaluate solutions, an experiment was conducted in Trevarez farm (CRA of Brittany, Idele) from 2013 to 2017 to assess the impact of two dairy systems representative of typical local dairy farms, based on the forage pillars of the region: maize silage and grazed grass.

Materials and methods

This trial was run with the sustainable dairy farms prototyping method (Coquil *et al.*, 2011): systems studied in parallel with annual improvements. This global assessment method does not allow to perform specific statistical analyses. The technical results obtained were used for economic simulations. In this article, only the last 5 years of each system (2013-2017) are presented, although the crop rotations were started in 2010. Trevarez experimental farm is located in Brittany (oceanic climate). The dairy herd is composed of 120 Holstein dairy cows with an average genetic level for the region. The first system (S1) was based on a limited grazable area (0.15 ha per cow), a high level of maize silage supplementation, and a medium level of concentrate allowing the expression of the genetic merit while mastering the feeding cost. The second system (S2) used a large part of pasture (0.40 ha per cow) supported by maize silage supplementation during the winter. Both systems were run in parallel after final allocation of cows (same genetic merit in each system) and surfaces (same soil potential in each system). Each system had approximately 60 hectares and 60 cows (Table 1). The heifers of the two systems were raised together but reintroduced to their mother's systems after calving. Both herds were managed as compact split-calving systems (2×3 months: March-May and September-November) with a target of 12 months calving interval. In both systems, cows' diets were based on maize silage during the winter, with protein concentrate supplementation to reach a nitrogen balance of 95 g PDI/UFL (protein digestible in the small intestine/Unité Fourragère Lait (forage unit for lactation)), plus 4 kg of production concentrate per cow per day during 120 first days of lactation.

Throughout the trial, all technical data about animals, soils, diets and feedstuffs were recorded similarly (cf. Brocard *et al.*, 2020). The analysis of environmental impacts was undertaken for each system with the CAP'2ER® tool (Idele), integrating water quality, air quality, global warming and energy consumption. The gross carbon footprint considers all GHG emissions linked to the milk production, whether direct or indirect. The net carbon footprint is the difference between the gross carbon footprint and an estimate of the carbon storage associated with grassland. Finally, the economic analysis was based on the data recorded, with two simulations, either with the same number of cows or with the same amount of milk sold. Data directly used from Trevarez were the sales, the milk discarded, and all variable costs. The fixed costs were evaluated with mechanization of crops done by contractor. For buildings and equipment, we simulated new investments with depreciation and loan durations of 20 years for the buildings and the milking parlour, and 9 years for equipment.

Results and discussion

On average over 5 years, the S1 system was based on 59.8 ha of agricultural area (AA): 5.4 ha of cereals and 54.5 ha of forage area (FA), of which 46% was maize silage (Table 1). The S2 system consisted of 64.9 ha of AA: 4.2 ha of cereals and 60.6 ha of FA, of which 28% was maize silage. The share of maize monoculture reached 67% in S1 versus 12% in S2 and may explain a lower maize yield of 1.7 t dry matter

Table 1. Technical performances, 2013-2017.

System	S1	S2
Agricultural area AA (ha)	59.8	64.9
Grazeable area per cow	0.15	0.40
Forage area (ha)	54.4	60.6
Cereals (ha)	5.4	4.2
% Maize silage in forage area	46	28
N# of dairy cows	59	64
N# of heifers LU	26	29
Livestock unit ha^{-1} of forage	1.55	1.52
Grass yield DM ha^{-1}	6.0	6.7
Maize silage yield DM ha^{-1}	11.8	13.5
% Maize in monoculture	67	12
ha ploughed per year % AA	52	41%
Maize silage cow^{-1} $year^{-1}$ (t DM)	4.4	2.9
Pasture cow^{-1} $year^{-1}$ (t DM)	1.2	2.5
Concentrate cow^{-1} $year^{-1}$ (kg)	965	690
of which soya equivalent (kg)	611	310
N# days without N concentrate y^{-1}	35	135
N# days without maize $year^{-1}$	0	70
% primiparous	34	32
% cull cows	26.1	27.9
Milk produced cow^{-1} $year^{-1}$ (kg)	8,162	7,608
Milk sold cow^{-1} $year^{-1}$ (l)	7,551	7,167
Fat content (g kg^{-1})	41.1	40.3
True protein content (g kg^{-1})	31.7	31.0

(DM) per ha in S1. The share of total area ploughed each year was 11% lower in S2. The high proportion of maize silage in the S1 diet required 611 kg of soybean equivalent concentrate cow^{-1} year^{-1} vs 310 kg in the S2 diet. On average, S1 cows spent only 35 days per year with no protein concentrate compared with 135 days in S2 where the maize silo could be closed 70 days per year thanks to a larger share of grazed grass (0 day in S1). The milk production was on average 554 kg yr^{-1} higher for S1 cows than for S2 cows, with more protein (+0.6 g kg^{-1}) and fat (+0.8 g kg^{-1}). When retrieving the milk discarded (after treatment for mastitis, or given to calves), the difference in milk delivered is reduced to +384 l per cow per year in favour of S1. The S1 used 118 g of concentrate per litre of milk sold compared with 91 g for S2. No differences in live weight, body condition score or reproduction performance were observed between systems. In both groups, the proportion of cows culled empty was close to 33%. Finally, there were slightly more cases of mastitis and lameness in S1, with a higher total number of disease incidents per treatment (+22 cases per 100 lactations).

In terms of environmental impacts, the N inputs per ha are higher in S1 due to a greater use of concentrate purchased (+30 kg N ha^{-1}). Thus, its surplus balance is higher by 23 kg ha^{-1} generating a potential leaching of 51 kg N ha^{-1} vs only 15 kg in S2. In both systems, the N balance is close to or lower than the reference data (Foray *et al.*, 2019). The N efficiency reaches 40% in S2, higher than the existing references for this type of dairy system (+4%). For GHG emissions (Table 2), the gross carbon footprints are very close, and lower than the CarbonDairy references. Thanks to a higher share of grass and more hedges, the C storage is higher in S2 with an average net C footprint of 0.81 kg eq CO_2 l^{-1} milk, (-0.05 below S1). For both systems, the enteric emissions of CH_4 represent the main share of the gross emissions.

In terms of economics, both systems produce the same volume of milk despite 5 extra cows in S2. However, S2 delivered a higher proportion of milk (+17,000 l per year). The feeding cost was also lower by €28 1000 l^{-1} sold in S2. Both systems are close to the regional economic references (Inosys). The margin over feeding cost (Milk price – feeding cost, MOFC) in S2 is €19 higher for 1000 l sold than in S1. The veterinary and reproduction costs per cow were similar in both systems, and close to the references.

Table 2. Environmental and economic impacts.

System	**S1**	**S2**
C footprint (g eq CO_2 l^{-1} corrected milk)[1]		
Gross C footprint	0.96	0.94
Net carbon footprint	0.86	0.81
Economic results[2], 2014-2017 (€ 1000 l^{-1})		
Milk produced (l year^{-1})	468,040	470,461
Milk sold (l year^{-1})	447,898	465,058
Milk price	327.0	326.3
Feeding cost dairy cows	79	58
MOFC	249	268
Total income	439	439
Variable costs	153	128
Fixed costs	104	100
GOS before labour	183	211
Income	86	110

[1] CarbonDairy Bretagne 2017 (Idele, 2018).

[2] Inosys Bretagne 2014-2017.

The lower level of variable costs in S2 directly impacts the Gross Operating Surplus (GOS) before labour and the income, leading to a total of +€ 48,000 over 5 years in favour of S2 for a farm selling 400,000 l of milk per year (24 × 5 × 400). With the same number of 60 cows, the difference in income would reach +€ 11,250 $year^{-1}$ in favour of S2.

Conclusions

The two systems implemented (predominantly maize or grass) were well above the average Breton dairy farm in terms of economic and environmental impacts. In the situation studied, the grazing-based system was the most economically efficient. It seems judicious to fully exploit this route of production through fodder and grazed grass provided there is accessibility for grazing. Otherwise, a 0.15 ha system can be optimized but will keep weaknesses: the share of monocultures (impacting soils, yields and use of phytosanitary products), the high cost of imported nitrogen inputs, and the lower economic robustness when the ratio of feed cost to milk price increases.

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References

Brocard V., Tranvoiz E., Dupre S., Foray S., Le Coeur P., Raison M., Trou G. and Follet D. (2020) Performances techniques, économiques et environnementales de deux systèmes laitiers contrastés. *Rencontres Recherches Ruminants* 25, 554-558.

Coquil X., Fiorelli, J.-L. *et al.*, 2011. Conception de systèmes de polyculture élevage laitiers en agriculture biologique: Synthèse de la démarche pas à pas centrée sur le dispositif expérimental INRA ASTER-Mirecourt. *Rencontres Recherches Ruminants* 18, 57-60.

CRA Bretagne (2017) Repères techniques et économiques en élevage laitier. *Réseau d'élevage* INOSYS. 8p.

Foray S. and Manneville V. (2019) *A la reconquête de la qualité de l'eau en France*. Idele, France, 39 p

Idele (2018) *Emissions de gaz à effet de serre et contributions positives en élevages laitiers*. Available at: https://www.idele.fr.

Creating and utilizing a DNA reference library for faecal DNA metabarcoding to determine diet composition of herbivores

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Abstract

Grazing animals are selective feeders, making continuous choices as to where and what to eat. Differences in nutritional value of plant species consumed can directly affect performance, reproduction, survival and overall population dynamics. Faecal DNA (*f*DNA) metabarcoding provides a non-invasive approach to determine the diet composition in herbivores more accurately. While metabarcoding approaches are increasingly popular, questions remain about the accuracy to which diet components can be identified down to species classification. Through the use of an automated reference sequence package (MetaCurator) a study-specific plant DNA reference library was curated to test, *in silico,* three plant barcoding markers *trn*L, *rbc*L, ITS2 to determine which exhibits the greatest sensitivity for taxonomic classification for use with *f*DNA metabarcoding. Initial results support using complementary markers, *rbc*L and ITS2, to give the greatest taxonomic breadth and, greatest taxonomic resolution of 86, 93 and 95% at the species, genus and family level. This *in silico* study advocates the use of these two primers for further dietary studies testing *f*DNA metabarcoding to determine the diet composition of herbivores.

Keywords: herbivory, faecal DNA, metabarcoding, high-throughput sequencing

Introduction

Plant-herbivore interactions are fundamental in shaping ecosystem function and population community dynamics; however, the diets of semi-wild animals have been persistently difficult to quantify (Garnick *et al.*, 2018). While past studies have used techniques such as visual grazing observation and microhistology they can be time consuming and limited on providing taxonomic precision regarding the botanical composition of a diet (Garnick *et al.*, 2018). Over the last decade the number of DNA-based dietary studies has rapidly risen as the development of high-throughput next generation sequencing platforms continues to improve the speed and accuracy of genetic analysis, especially when applied to relatively complex mixtures of substrates such as faecal samples (Alberdi *et al.*, 2019; Sousa *et al.*, 2019). Two critical factors influencing the outcome of DNA metabarcoding are: (1) the choice of appropriate marker region(s), these need to be short yet highly variable between different species in order to detect degraded DNA recovered form faecal samples, and (2) the availability of suitable DNA barcode reference libraries, if sequencing results are to achieve a high taxonomic resolution (Deagle and Tollit, 2007; Moorhouse-Gann *et al.*, 2018). Curating study-specific reference libraries has, to date, been a major challenge due to the possibility of errors or incomplete barcodes in online published DNA sequence depositories or the associated reliability of taxonomic assignment (Valentini *et al.*, 2009). This project reports the approaches and methodologies used to generate a study-specific reference library in preparation for experimental study exploring the impact of physiological status, health status and seasonal variation in the availability of vegetation resources on diet composition by free-ranging wild Soay sheep on the island of St Kilda, and describes in detail how this reference library was created using the MetaCurator software package (Richardson *et al.*, 2020). Specific goals within this development work were: (1) to evaluate *in silico* the extent to which the use of an exhaustive curated St Kilda reference database would increase species-level taxonomic assignment via faecal (*f*)DNA metabarcoding, and (2) to identify which universal primer or primer combination would provide the best PCR amplification, when testing three plant barcoding markers *trn*L, *rbc*L, ITS2.

Materials and methods

A master list was compiled of all known vegetation to be found on St Kilda with additional known test plant species used in zero-grazing trials with sheep at Pwllpeiran Upland Research Centre, Aberystwyth University. The final vegetation master list to be used as a reference library comprised 203 species across 59 families. Relevant scripts published in the study by Richardson (*et al*., 2020) included the search terms for each plant marker in question (*rbc*L, *trn*L, and ITS2) to be inputted to the NCBI nucleotide database GenBank, whereby a full range of every sequence available for each representative primer was obtained (Table 1).

MetaCurator was used to automate extracting sequence data for the specific plant primers. For each primer the full sequence list was filtered to contain information relating to species on the St Kilda vegetation reference list. Appropriate accession numbers, a unique identifier number for a species' specific sequence record were obtained. Accumulated accession numbers were then ran through an R package 'Taxonomizr' to convert accession numbers into taxonomy information before running through MetaCurator. The output data from MetaCurator then provided sequences that were trimmed down for each primer based upon the known species list at 100% clustering identity, therefore providing information based on just the sequence region that would be amplified with the primer in question rather than whole gene length.

Results and discussion

Utilizing MetaCurator to construct a plant reference database it was possible to determine which species sequences are available with each plant marker, whether there were any species missing, and if yes, whether this was because they were clustering with taxonomically similar species or because there were no corresponding sequence data currently available. All markers had 80-95% coverage of the 59 families and 121 genera associated with the St Kilda vegetation master list. After filtering the curated reference library to show species with sequence data available across all three markers, it resulted in a total of 195 species across 56 families, accounting for 95% of species coverage from the original St Kilda vegetation list. For the purpose of this abstract the most abundant four families are presented. Table 2 indicates what % of species within a family can be identified to the species level at the 100% clustering threshold, and therefore have their own unique sequence reference available.

From this *in silico* study *trn*L appears to have the least coverage for identifying vegetation down to both the species and genus level compared to the other two markers. While *rbc*L appears to have the greatest taxonomic coverage for species in the Poaceae family, ITS2 complements this with greater taxonomic coverage for species resolution in the families Asteracea and Fabaceae. There appears to be discrepancies amongst all three markers, suggesting that taxonomic assignment could be improved using complementary multiple markers approach such as *rbc*L and ITS2 to aid resolution for determining herbivore diets via *f*DNA metabarcoding.

Table 1. Sequence database available from GenBank for each representative marker between start search dates 01/01/1800 – 29/03/2021 summarized for both total entries and % coverage of different taxa against the targeted St Kilda vegetation reference master list. (n=) represents the total number of families, genera and species within the final St Kilda vegetation reference master list.

Study System	DNA plant marker	All species total database	St Kilda master list		
			Family (n=59)	Genus (n=121)	Species (n=203)
Plant	*rbc*L	251,435	95%	93%	86%
	*trn*L	310,998	95%	80%	76%
	ITS2	432,865	95%	82%	82%

Table 2. Summary of which primer is predicted to be the most effective at distinguishing the most species per family.[1]

Family	Total no. genus	Total no. species	% of unique species sequences available		
			RBCL	TRNL	ITS2
Poaceae	15	27	63	48	48
Asteraceae	13	17	41	24	76
Cyperaceae	5	17	76	41	76
Fabaceae	4	6	50	67	83

[1] Cell colour indicates the level of species resolution from dark grey indicating a high species level resolution, decreasing through to light grey at a poor species level resolution. The number within each cell represent the % of species within a family that can be identified down to species level at 100% clustering threshold.

Conclusions

Curating a comprehensive reference sequence library to improve the accuracy of taxonomic resolution via *f*DNA metabarcoding supports new avenues to investigate unanswered questions associated with nutritional ecology. This information will increase our understanding of plant-herbivore interactions, specifically addressing diet choice, immunity, gut bacteria, parasite burden, and how these further influence population dynamics under natural conditions. Outputs will have relevance to both wild and domesticated herbivore populations and, the knowledge gained will improve further decision making and the development of predictive models used for ecosystem conservation and livestock management. This information will too support optimal grazing regimes for improving animal nutrition via grazing management, and avoiding over- exploitation and depletion of habitat resources.

References

Alberdi A., Aizpurua O., Bohmann K., Gopalakrishnan S., Lynggaard C., Nielsen M. and Gilbert M.T.P. (2019) Promises and pitfalls of using high-throughput sequencing for diet analysis. *Molecular Ecology Resources,* 19(2), 327-348.

Deagle B.E. and Tollit D.J. (2007) Quantitative analysis of prey DNA in pinniped faeces: Potential to estimate diet composition? *Conservation Genetics*, 8(3), 743-747.

Garnick S., Barboza P.S. and Walker J.W. (2018) Assessment of animal-based methods used for estimating and monitoring rangeland herbivore diet composition. *Rangeland Ecology and Management* 71(4), 449-457.

Moorhouse-Gann R.J., Dunn J.C., De Vere N., Goder M., Cole N., Hipperson H. and Symondson W.O (2018) New universal ITS2 primers for high-resolution herbivory analyses using DNA metabarcoding in both tropical and temperate zones. *Scientific Reports* 8(1), 8542.

Richardson R.T., Sponsler D.B., McMinn-Sauder H. and JohnsonR.M. (2020) MetaCurator: A hidden Markov model-based toolkit for extracting and curating sequences from taxonomically-informative genetic markers. *Methods in Ecology and Evolution* 11(1), 181-186.

Scasta J.D., Jorns T., Derner J.D., Lake S., Augustine D.J., Windh J.L. and Smith T.L. (2019) Validation of DNA metabarcoding of fecal samples using cattle fed known rations. *Animal Feed Science and Technology* 255, 114219.

Sousa L.L., Silva S.M. and Xavier R. (2019) DNA metabarcoding in diet studies: Unveiling ecological aspects in aquatic and terrestrial ecosystems. *Environmental DNA* 1(3), 199-214.

Valentini A., Pompanon F. and Taberlet P. (2009) DNA barcoding for ecologists. *Trends in Ecology and Evolution* 24(2), 110-117.

Pasture type effect on fatty acids and fat-soluble antioxidants profile in grazing cows' dairy milk

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Abstract

In an experiment conducted in the Atlantic zone of Galicia (NW Spain) during the spring period (April-June), two groups of Holstein cows were allocated to grazing one of two low nitrogen (N) input pasture types: perennial ryegrass receiving 100 kg N ha^{-1}, vs red clover, 0 kg N ha^{-1}). Grazing took place during the day and the cows were stalled during the night, having access to a low-concentrate (ca. 100 g cow^{-1}) total mixed ration based on maize silage. The effect of the treatments on milk production and milk fatty acids and fat-soluble antioxidants profiles was analysed. The average daily production achieved was higher (+14.6%) in cows grazing the red clover pasture (30.6 kg cow^{-1}) compared to cows grazing perennial ryegrass (26.7 kg cow^{-1}). Milk from cows grazing red clover swards showed a less-saturated, higher-polyunsaturated profile and lower omega6:omega3 ratio compared with milk from cows grazing perennial ryegrass, whilst no major differences were detected between treatments in the vitamins A and E and carotenoids content of milk.

Keywords: grazing, dairy production, fatty acids, antioxidants

Introduction

There is a general agreement on the higher nutritional quality associated with milk produced in grazing systems. For example, Stergiadis *et al.* (2015) reported higher concentrations of omega-3 fatty acids (FA), lutein and zeaxanthin in milk from cows in grazing vs confined systems based on maize silage and concentrate diets, indicating that a switch to pasture-based dairy products would increase the intake of milk's beneficial compounds and reduce consumption of less-desirable saturated FA. The use of legume species in pastures can offer additional advantages compared with the use of grass-based swards, based both on an enhanced milk yield and FA profile, and reduced input of nitrogen (N) fertilizer on the farm (Dewhurst *et al.*, 2009). The objective of this paper was to compare the effect of grazing perennial ryegrass or red clover swards on milk production and milk fatty acids, vitamins and carotenoids profile.

Materials and methods

The grazing experiment took place from mid-April to mid-July at the Centro de Investigacións Agrarias de Mabegondo (CIAM) research station farm (Galicia, NW Spain, 43°15′N, 8°18′W, 100 m above sea level) on a silt loam soil. Twenty Holstein-Friesian cows were randomly distributed into two equal groups of ten cows and assigned to one of two pastures: a perennial ryegrass (*Lolium perenne* L.) sward (PR, fertilized with 100 kg N ha^{-1}) and a red clover (*Trifolium pratense* L.) sward (RC, no N fertilization). Cows rotationally grazed the paddocks between the morning and the afternoon milking (9:00 am to 19:00 pm), after which the animals were fed in the barn a total mixed ration composed of 5.0 kg of maize silage, 0.5 kg of grass hay and 2.5 kg of a commercial concentrate on a dry-matter (DM) basis per cow and day. The concentrate had a vitamin supplementation of 30,000 UI of vitamin A and 60 mg kg^{-1} of vitamin E (alpha-tocopherol). Individual milk yield was recorded daily at the milking parlour and milk samples were taken per animal in the morning and evening milkings of 3 consecutive days in the weeks 3, 6, 9 and 12 of the experiment. Samples were immediately stored at 4 °C and transported to the official regional interprofessional milk laboratory (LIGAL) where they were subjected to routine FTMIR analysis (milk composition) or immediately frozen (-20 °C) until posterior chromatographic analysis (FID-GC for FA and HPLC for vitamins A and E and carotenoids) following the routines established by the LIGAL. A two factors (sward type and period) repeated measures analysis was performed using Proc GLM of SAS (SAS Institute, 2009) where the period was the within subjects (repeated) variable.

Results and discussion

Milk production and milk protein production was significantly higher for the cows in the RC treatment (Table 1) compared to PR, while milk urea content in PR was very low compared with RC milk samples (82 vs 273 mg urea l^{-1} milk). These differences were attributed to the extremely low crude protein content of the perennial ryegrass compared with the red clover, in contrast with the higher values observed in the perennial ryegrass pasture for the content in water-soluble carbohydrates and the *in vitro* organic matter digestibility. The milk FA profile was affected by the pasture type showing the RC milk a significantly less-saturated, high-polyunsaturated profile. The significantly higher values for the alpha-linolenic FA content in milk of RC compared with PR together with the lower omega-6:omega-3 ratio in the former treatment contribute to confirm the results of previous studies indicating that the inclusion of legume pastures in the dairy cows' diet can improve the milk profile from the perspective of human health (Dewhurst *et al.*, 2009).

Table 1. Pasture nutritive value, milk productivity and milk fatty acids profile.[1,2]

	PR	RC	*P*-value
n	**120**	**117**	
Pasture nutritive value			
Crude protein (g kg^{-1} DM)	74	187	***
Water soluble carbohydrates (g kg^{-1} DM)	311	96	***
In vitro organic matter digestibility (g kg^{-1})	815	705	***
Milk productivity (kg cow^{-1} d^{-1})			
Milk[3]	26.7	30.8	*
Milk fat	0.99	1.13	NS
Milk protein	0.75	0.91	**
Main FA groups (g kg^{-1} total FA)			
Saturated	696	659	*
Monounsaturated	267	283	NS
Polyunsaturated	37	57	**
Omega-6 total FA	21	30	**
Omega-3 total FA	9	17	**
Individual FA (g kg^{-1} total FA)			
C16:0	351	301	**
Oleic acid (C18:1n9*c*)	207	216	NS
Linoleic acid (C18:2n6*c*)	16	23	*
Alpha-linolenic (C18:3 n3)	8	16	**
Omega-6:Omega-3	2.3	1.8	**

[1] PR = perennial ryegrass; RC = red clover

[2] NS = non-significant (P>0.05); * P<0.05; ** P<0.01; *** P<0.001.

[3] Milk corrected at 35 g kg^{-1} fat and 32 g kg^{-1} protein.

The effect of treatments on the concentration of vitamins A and E and carotenoids is shown in Table 2. Vitamins A (retinol) and E (alpha-tocopherol) concentrations in milk varied from 10.66 and 20.00 mg kg^{-1} fat in PR, and 12.73 and 17.44 mg kg^{-1} fat in RC. with no significant differences between treatments. Similarly, the concentration in milk of the main carotenoids, lutein and *all-trans+9 cis* β-carotene, in PR and RC was not significantly different between pasture types. Only for the minority xanthophylls, zeaxanthin and β-cryptoxanthin (higher values in PR and RC, respectively), and for 13-*cis*-β-carotene (higher value in RC) the differences reached statistical significance. The results observed are in line with other studies which investigated the content of vitamins and carotenoids in dairy milk. For example, Mogensen *et al.* (2012) found the highest concentrations of retinol, α-tocopherol and β-carotene in farms using grass-red clover silage in their study in five organic dairy herds in Denmark. It is known that concentrations of vitamins A and E and carotenoids in milk are dependent on the amounts consumed by the cow (e.g. Weiss *et al.*, 1990) which indicates that, in this regard, PR and RC pasture types exhibited a similar behaviour in our study.

Table 2. Concentration of vitamins A and E, xanthophylls and carotene in milk.[1,2]

	PR	RC	*P*-value
n	120	117	
Vitamins (mg kg^{-1} fat)			
Vit. A (retinol)	10.66	12.73	NS
Vit. E (α-tocopherol)	20.00	17.44	NS
Vit. E (γ-tocopherol)	0.38	0.37	NS
Xanthophylls (μg l^{-1} milk)			
Lutein	0.34	0.33	NS
Zeaxanthin	0.05	0.04	**
β-cryptoxanthin	0.06	0.06	**
Carotene (μg l^{-1} milk)			
(All-t-β+ 9-c-β) carotene	3.25	3.70	NS
13-*cis*-β-carotene	0.16	0.18	**

[1] PR = perennial ryegrass; RC = red clover

[2] NS = non-significant (P>0.05); ** P<0.01.

Conclusions

No major differences between milk from grass or red clovers swards were found, in terms of milk fat-soluble antioxidants content. Milk from cows grazing red clover swards showed a less-saturated and higher-polyunsaturated FA profile.

Acknowledgements

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References

Dewhurst R.J., Delaby L., Moloney A., Boland T. and Lewis E. (2009) Nutritive value of forage legumes used for grazing and silage. *Irish Journal of Agricultural and Food Research* 48, 167-187.

Mogensen L., Kristensen T., Søegaard K., Jensen S.K. and Sehested J. (2012) Alfa-tocopherol and beta-carotene in roughages and milk in organic dairy herds. *Livestock Science* 145(1-3), 44-54.

Stergiadis S., Leifert C., Seal C., Eyre M., Larsen M., Slots T., Nielsen J.H. and Butler G. (2015) A 2-year study on milk quality from three pasture-based dairy systems of contrasting production intensities in Wales. *Journal of Agricultural Science* 153(4), 708-731.

Weiss W.P., Hogan J.S., Smith K.L. and Hoblet K.H. (1990) Relationships among selenium, vitamin E and mamary-gland health in commercial dairy herds. *Journal of Dairy Science* 73(2), 381-390.

Biochar decreases the ammonia emissions of cattle slurry

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Abstract

Dairy cattle use only 25-35% of their dietary nitrogen for the synthesis of milk protein, and the remainder is excreted in faeces and urine. A large proportion of the cattle slurry is applied as organic fertilizer for grassland production. Slurry application to crop fields recycles animal wastes and is a valuable source of nutrients, and it can improve the physical and chemical edaphic properties of the soil. However, slurry can be a source of environmental pollution because a high proportion of the nitrogen is released as ammonia, and the non-ammonia nitrogen would be converted into ammonia-N in short term. Biochar, a charcoal produced by pyrolysis of biomass, is being investigated as a means of carbon sequestration and it may be useful to mitigate climate change. We studied the capacity of biochar to bind ammonia, and increase the non-ammonia nitrogen of slurry to be used as organic fertilizer. Results show that the addition of 2% biochar to cattle slurry reduces ammonia emissions by up to 16%, increasing the nitrogen content per m^3 after treatment, as well as that of dry matter and ash. Therefore, slurry with biochar can have a higher fertilizer value than untreated slurry.

Keywords: dairy cow, slurry, ammonia, biochar

Introduction

Manure and slurry may result in emissions of potent greenhouse gases. Furthermore, nitrogen losses via ammonia volatilization during slurry storage may reduce their fertilizer value as a soil amendment. Therefore, it is important to reduce gaseous emissions during composting. Ammonium removal from slurry using membrane reactors, anaerobic oxidation, denitrification in lagoons, and by adsorption methods with activated carbons, ion exchange resins and zeolites have all received reasonable research attention (Değermenci *et al.*, 2012; Huang *et al.*, 2010). Biochar, a charcoal produced by pyrolysis of biomass in the absence of oxygen, is being investigated as a means of carbon sequestration. It has also been reported that the application of biochar in pig slurry adsorbs up to 60% of the N-NH_3 emitted (Kizito *et al.*, 2015). Studies have been carried out on the addition of biochar to cattle slurry together with the use of acidifiers and molasses (Schmidt, 2012). It was further suggested that the application of biochar in animal bedding or as an additive for slurry storage could reduce manure-related methane emissions (Kammann *et al.*, 2017). The main objective of this paper was to study the capacity of biochar to bind ammonia, and increase the non-ammonia nitrogen of slurry to be used as organic fertilizer.

Materials and methods

A trial to evaluate the effectiveness of biochar to ammonia sequestration from cattle slurry was conducted using an *in vitro* system based on the one described by Hassouna *et al.* (2016). This method is based on the affinity between ammonia and an acid solution. The ammonia in the air emitted from slurry over a known period and partially sampled at a known airflow was trapped in 1N boric acid in an impinger. This method is able to trap nearly 100% of the ammonia in the air sampled.

The treatments evaluated were slurry with 2% (w/w) biochar, as recommended by Schmidt (2012), and untreated slurry as control. The trial was carried out with 2 kg of slurry per fermenter bottle with three incubation batches. Both treatments were evaluated in duplicate in all incubation runs and extended for 15 days. The slurry used came from the SERIDA experimental farm. The trial was carried out in an aerobic stability chamber with dry and still atmosphere at an ambient temperature of 21±2 °C. The dry matter, ash and nitrogen contents of the slurry were analysed at the beginning and at the end of each batch. The concentration of $N\text{-}NH_4^+$ in the acid solution was analysed by Kjeldahl method. Then, the ammonia concentration was calculated in the air taking account of the mass of the acid solution in the impinger and the volume of air that has passed through the impinger.

The results were analysed by an analysis of variance applying R software (R Core Team, 2021) taking account of the treatment (untreated or biochar treated slurry) and incubation batch as fixed effects. Significance was set at $P<0.05$.

Results and discussion

The slurry used was very diluted, with 3.6% dry matter and 1.1% nitrogen, and 25% of the nitrogen was in the form of ammoniacal nitrogen.

Figure 1 shows the cumulative ammonia emission per litre of slurry over the test period. The total ammonia emission was significantly lower with the biochar-treated slurry ($P<0.001$). The biochar reduces ammonia emissions by 4% after 72 hours, increasing the retention to 10.75% after 5 days of incubation and stabilizing at 16% after the ninth day of incubation. The results were in line with Sarkhot *et al.* (2014), who concluded that biochar addition adsorbed up to 18% of $N\text{-}NH_4^+$ from dairy manure effluent. Likewise, Chowdhury *et al.* (2014) reported that biochar addition reduces ammonia losses from composting hen manure.

At the end of the trial, the slurry composition between treatments was not statistically different (Table 1). However, the slurry treated with 2% biochar would apply up to 18% more nitrogen to the soil than

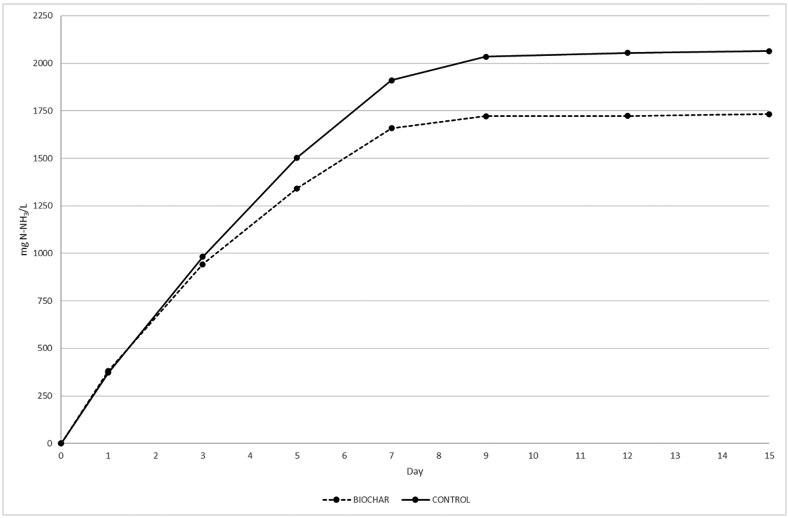

Figure 1. Ammonia emission evolution, in mg l^{-1} of slurry, according to treatment with or without biochar addition.

Table 1. Slurry composition (kg/m^3) before the start of the study (Pre-treated) and after incubation with 2% biochar (Biochar) or without biochar addition (Control).

	Pre-treated	Biochar	Control	Standard error	*P*-value
Dry matter	35.38	51.29	38.81	11.870	0.310
Ash	8.37	11.06	9.86	3.908	0.713
Nitrogen	0.82	1.05	0.89	0.252	0.555

untreated slurry. This could be a consequence of the lower evaporation of ammonia nitrogen from the treated slurry.

The high variability in the results prevents significant differences from being achieved. The results are, however, in line with several other studies (Sarkhot *et al.*, 2014; Wang *et al.*, 2012). The uptake of ammonia nitrogen to biochar-treated slurry is more efficient in terms of recycling organic nitrogen as a fertilizer (Krounbi *et al.*, 2021). In addition, dairy manure with biochar can provide sufficient P and K requirements for corn and forage crops (Piash *et al.*., 2021). The application of biochar-treated dairy slurry as a supplemental fertilizer is a beneficial strategy to reduce nutrient contamination and supply nutrients to forage crops.

Conclusions

Based on the results obtained, it can be concluded that the addition of 2% biochar to cattle slurry reduces ammonia emissions to the environment by up to 16%. Biochar has potential as ammonia binder of cattle slurry and, therefore, of being a mitigation agent for environmentally detrimental nitrogen losses.

References

Chowdhury M.A., de Neergaard A. and Jensen L.S. (2014) Potential of aeration flow rate and bio-char addition to reduce greenhouse gas and ammonia emissions during manure composting. *Chemosphere* 97, 16-25.

Değermenci N., Ata O.N. and Yildız E. (2012) Ammonia removal by air stripping in a semi-batch jet loop reactor. *Journal of Industrial and Engineering Chemistry* 18, 399-404.

Hassouna M., Eglin T., Cellier P., Colomb V., Cohan J.P., Decuq C. and Fauvel Y. (2016) *Measuring emissions from livestock farming: greenhouse gases, ammonia and nitrogen oxides.* INRA-ADEME, 2016.

Kammann C., Ippolito J., Hagemann N., Borchard M., Cayuela J., Estavillo T., Fuertes-Mendizabal T., Jeffery S., Kern J., Novak J., Rasse D., Saarnio S., Schmidt H., Spokas K. and Wrage-Mönnig N. (2017) Biochar as a tool to reduce the agricultural greenhouse-gas burden-knowns, unknowns and future research needs. *Journal of Environmental Engineering and Landscape Management* 25, 114-139.

Kizito S., Wu S., Kirui W.K., Lei M., Lu Q., Bah H. and Dong R. (2015) Evaluation of slow pyrolyzed wood and rice husks biochar for adsorption of ammonium nitrogen from piggery manure anaerobic digestate slurry. *Science of the Total Environment* 505, 102-112.

Krounbi L., Enders A., Gaunt J., Ball M. and Lehmann J. (2021) Plant uptake of nitrogen adsorbed to biochars made from dairy manure. *Scientific Reports* 11, 15001.

Piash M.I., Iwabuchi K., Itoh T. and Uemura K. (2021) Release of essential plant nutrients from manure-and wood-based biochars. *Geoderma* 397, 115100.

R Core Team (2021) *R: A language and environment for statistical computing.* R Foundation for Statistical Computing, Vienna, Austria.

Sarkhot D.V., Ghezzehei T.A. and Berhe A.A. (2014) Effectiveness of biochar for sorption of ammonium and phosphate from dairy effluent. *Journal of Environmental Quality* 42, 1545-1554.

Schmidt H.P. (2012) Treating liquid manure with biochar. *Ithaka Journal* 1, 273-276.

Wang T., Arbestain M.C., Hedley M. and Bishop P. (2012) Chemical and bioassay characterisation of nitrogen availability in biochar produced from dairy manure and biosolids. *Organic Geochemistry* 51, 45-54.

Impact of autumn closing date and spring defoliation date on herbage production and clover content

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Abstract

The objective of this study was to investigate the impact of opening farm cover (OFC) and spring defoliation date on herbage production and sward clover content. A split plot design was used with four repetitions, giving 48 plots (1.2×5 m). Four spring OFCs were established by closing swards on 25 Sept (Very High, VH), 9 Oct (High, H), 30 Oct (Medium, M) and 20 Nov (Low, L), and three spring defoliation dates early (15 Feb), normal (9 March) and late (29 March). Herbage mass and clover content were determined at each harvest. Herbage mass at first defoliation was greatest in the VH OFC and lowest on the L OFC, at the early spring defoliation (2,079 and 490 kg dry matter ha^{-1}, respectively). Clover content was highest on medium OFC that were defoliated on the normal grazing date and lowest in plots with a VH OFC that were also defoliated on the normal grazing date (28.8 and 17.4%, respectively). Opening with a high OFC in spring increased the overall yield; however, there is a negative impact on sward clover content particularly with the normal and late defoliation dates and, for this reason, high covers should be defoliated as early as possible in spring.

Keywords: autumn closing, defoliation date, herbage mass, grazed herbage, clover content

Introduction

Autumn closing date of pastures has a major impact on spring herbage availability. Each day delay in autumn closing date reduces spring grass availability by up to 16 kg dry matter (DM) ha^{-1} day^{-1} (Looney *et al.,* 2021; Lawrence *et al.*, 2017). There is a reduction in the nutritive quality of early closed swards compared to later closed swards, but there is still an increase in milk production in spring due to higher levels of grass available to cows to meet their increasing energy demands (Claffey *et al.*, 2019). The date at which a sward is grazed in spring can influence herbage yield and quality in subsequent rotations. Reductions in herbage mass can still be seen in summer as a result of late autumn closing dates (Hennessy *et al.*, 2006). Early grazing in spring increases tillering (Roche *et al*, 1996) and reduces the build-up of dead material caused by leaf senescence at the base of the sward (Carton *et al.*, 1989). Herbage mass has a negative impact on sward clover content (Davies *et al.*, 1990). The objective of the current study was to investigate the effect of herbage mass and defoliation date in spring on herbage production and sward clover content.

Materials and methods

The experiment was carried out at the Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Ireland from February 2021 to October 2021. The plot experiment was a 4×3 factorial design with four repetitions giving a total of 48 plots (1.2×5 m). Four autumn closing dates (CD) were used to achieve four levels of herbage mass in spring: 25 September (very high, VH), 9 October (high, H), 30 October (medium, M) and 20 November (low, L); with three spring defoliation dates (DD); Early (15 February), normal (9 March) and late (29 March). Sward clover content was determined for the experimental site before assigning treatments. Treatments were randomly assigned to plots across the four repetitions to ensure an accurate representation of the effect of treatment. Plot heights were recorded using a rising plate meter every three weeks from 20 November to determine over-winter growth rates. All plots were defoliated at an average pre-grazing herbage mass of 1,500 kg dry matter (DM) ha^{-1} for the second and subsequent defoliations, with a total of eight defoliations. Sward clover content was also determined at each DD in spring and on four more occasions during the experimental period. Plots were mechanically defoliated (Etesia Hydro 124D; Etesia UK Ltd.) to determine herbage mass (>3.5 cm above ground) for each plot. Cut herbage was weighed and 0.1 kg dried at 90 °C for 16 hours to determine DM content. Data were analysed using PROC MIXED in SAS 9.4 (SAS Institute Inc., Cary, NC, USA, 2002) CD, DD and associated interactions were included as fixed effects; repetition was included as a random effect.

Results and discussion

Opening farm cover had a significant effect (P<0.0001) on herbage mass in spring. The pre-grazing herbage mass for the four opening farm cover (OFC) were: 1,672, 1,356, 977 and 744 kg DM ha^{-1} for VH, H, M and L, respectively. This is similar to the findings of Looney *et al.* (2021) who noted that early closed swards had higher herbage masses in the subsequent spring compared to later closed swards. Spring defoliation date also had a significant effect (P<0.0001) on herbage DM yield for the first cut. Late defoliation date had the highest DM yield (1,524 kg DM ha^{-1}), normal defoliation date was intermediate (1,276 kg DM ha^{-1}), and early defoliation was the lowest (761 kg DM ha^{-1}). There was no significant effect of the interaction of CD and DD on spring yield. Opening farm cover had a significant effect (P<0.001) on sward clover content from cut 3 (24 May) to cut 7 (1 September). The VH had the lowest clover content (20.1%) while M had the highest clover content (26.4%). This is similar to the findings of Phelan *et al.* (2013) who reported that higher herbage mass reduced light penetration to the base of the sward, which resulted in reduced growth of clover.

Closing date in autumn had a significant effect (P<0.01) on over-winter growth rates (OW-GR). The VH treatment closed on 25 September had a loss of -2.58 kg DM day^{-1} over winter and the L treatment closed on 20 November had the highest OW-GR at 1.25 kg DM day^{-1}. This is similar to the findings of Looney *et al.* (2021) who reported that swards reached a ceiling yield at 2,000 kg DM ha^{-1} and lost up to 18 kg DM ha^{-1} day^{-1} thereafter due to increased senescence. The higher pre-grazing herbage mass on VH was achieved through the growth that occurred from closing until the ceiling yield was reached. Opening farm

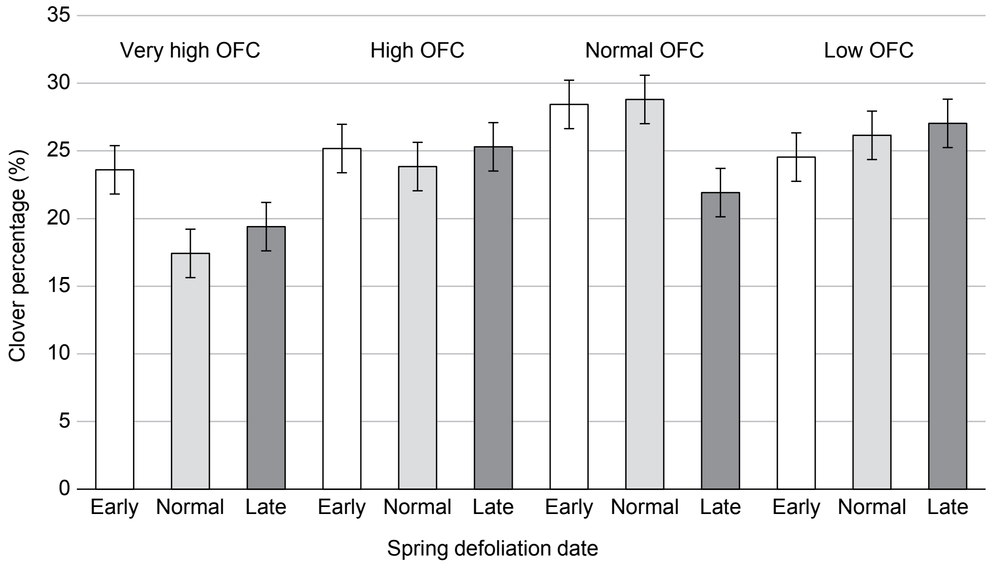

Figure 1. Mean sward clover content of plots from cut 3 (24 May) to cut 7 (1 September) with early (15 February), normal (9 March) and late (29 March) spring defoliation dates and very high (VH), high (H), medium (M) and low (L) opening farm covers (OFC). Data shown are means ± SE.

cover had a significant effect (P<0.05) on cumulative DM yield for the experimental period (February 2021 – October 2021). The VH plots grew the most during the experimental period (10,614 kg DM ha^{-1}) and L plots grew the least (9,921 kg DM ha^{-1}). Spring defoliation date also had a significant effect (P<0.05) on cumulative DM yield. Early defoliated plots grew the most (10,757 kg DM ha^{-1}), normal spring defoliation date was intermediate (10,350 kg DM ha^{-1}) and late spring defoliation plots yielded the least (10,037 kg DM ha^{-1}).

Conclusions

Autumn closing date has a significant impact on spring herbage availability, and subsequent cumulative DM yield. However, VH herbage masses in spring have a negative impact on sward clover content for the remainder of the year. Early defoliation in spring of VH herbage masses can reduce the negative effects on sward clover content, but will not recover to the same level as lower OFC in spring (M and L).

References

Carton O.T., Brereton A.J., O'Keeffe W.F. and Keane G.P. (1989) Effect of turnout date and grazing severity in a rotationally grazed reproductive sward: 1. Dry Matter production. *Irish Journal of Agricultural Research* 28(2), 153-163. http://www.jstor.org/stable/25556243

Claffey A., Delaby L., Galvin N., Boland T.M. and Egan M. (2019) The effect of spring grass availability and grazing rotation length on the production and quality of herbage and milk in early spring. *Journal of Agricultural Science* 157(5), 434-448.

Davies A. and Evans M.E. (1990) Effects of spring defoliation and fertilizer nitrogen on the growth of white clover in ryegrass-clover swards. *Grass and Forage Science* 45, 345-356.

Hennessy D., O'Donovan M., French P. and Laidlaw A.S. (2006) Effects of date of autumn closing and timing of winter grazing on herbage production in winter and spring. *Grass and Forage Science* 61, 363-374. https://doi.org/10.1111/j.1365-2494.2006.00543.x

Lawrence D.C., O'Donovan M. and Kennedy E. (2017) Effects of autumn and spring defoliation management on the dry-matter yield and herbage quality of perennial ryegrass swards throughout the year. *Grass and Forage Science* 72, 38-49.

Looney C., Hennessy D., Wingler A. *et al.* (2021) An examination of the effect of autumn closing date on over-winter herbage production and spring yield. *Irish Journal of Agricultural and Food Research* 60, 65-77. https://doi.org/10.15212/ijafr-2020-0129

Looney C., Wingler A. and Egan M. (2021) The impact of herbage mass on perennial ryegrass swards in autumn on autumn and over winter production and characteristics. *Agronomy* 11(6), 1140.

Phelan P., Casey I.A. and Humphreys J. (2014) The effects of simulated summer-to-winter grazing management on herbage production in a grass–clover sward. *Grass and Forage Science* 69, 251-265.

Roche J.R., Dillon P., Crosse S. and Rath M. (1996) The effect of closing date of pasture in autumn and turnout date in spring on sward characteristics, dry matter yield and milk production of spring-calving dairy cows. *Irish Journal of Agricultural and Food Research* 35(2), 127-140.

The effect of spring grass availability on milk production in early lactation

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Abstract

Spring grass availability can have a significant impact on early lactation milk production in spring calving herds. The objective of this study was to investigate the effect of opening farm cover (OFC) and silage supplementation on early lactation milk production. A high and low OFC were established for two treatment groups: 1,080 kg dry matter (DM) ha^{-1} (HG) and 800 kg DM ha^{-1} (LG) (>3.5 cm) in spring. Cows on the LG treatment were offered a lower daily herbage allowance (DHA) (8.07 kg DM cow^{-1} day^{-1} during weeks 1-5 (Period 1) and 11.56 kg DM cow^{-1} day^{-1} during weeks 6-12 (Period 2)) and supplemented with 3 kg DM silage. Cows on the HG treatment were offered a higher DHA (10.70 kg DM cow^{-1} day^{-1} in Period 1 and 15.11 kg DM cow^{-1} day^{-1} in Period 2) with no silage supplementation. Milk yields were recorded daily and milk composition weekly. Cows on the HG treatment had a greater ($P<0.01$) milk protein concentration in Period 2 compared to the LG treatment (+1.8 g kg^{-1}). Silage supplementation on the LG treatment resulted in a lower total dry matter intake (DMI) compared to the HG treatment ($P<0.001$) in Period 2 (16.4 and 17.3 kg DM cow^{-1} day^{-1}, respectively). Including silage in the diet during early lactation results in a lower DMI and milk protein concentration. DMI can be increased with a higher OFC.

Keywords: spring grassland management, milk production, dry matter intake, supplementation

Introduction

Spring grass is the most important feed source in pasture-based milk production systems in temperate regions (Claffey *et al.*, 2019). However, early spring is a period of reduced grass growth and cows require supplementation. The diet of dairy cows in early lactation has a significant impact on milk production (Claffey *et al.*, 2019). Previous studies have reported that cows offered grazed grass compared to grass silage had a higher dry matter intake (DMI), which in turn resulted in an increase in milk production (Kennedy *et al.*, 2006). Increasing daily herbage allowance (DHA) can have a positive effect on animal performance in early lactation (Kennedy *et al.*, 2007). DMI of dairy cows is mainly influenced by the intake capacity and milk production of the cow (McEvoy *et al.*, 2009). Increasing spring herbage (measured as opening farm cover, OFC) can allow a greater DHA, which can result in an increase in milk production (Claffey *et al.*, 2019); however, the inclusion of silage in the diet is often required during early lactation and this can reduce grass DMI compared to cows offered grass only (Kennedy *et al.*, 2006).

Materials and methods

The experiment was carried out at the Teagasc Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Ireland from February 2021 to June 2021. Two closing strategies were established in autumn 2020 to create a high (1,080 kg dry matter (DM) ha^{-1}) and low (800 kg DM ha^{-1}) OFC in spring 2021. A total of 80 spring-calving Holstein Friesian and Jersey × Holstein dairy cows (19 primiparous and 61 multiparous) were blocked by calving date (12/02/2021), parity, breed, pre-experimental milk production, bodyweight and body condition score. Cows were randomly assigned to one of two treatments as they calved: High grass (HG) and low grass (LG). Cows on the HG treatment were offered a higher DHA, while cows on the LG treatment were given a lower DHA and supplemented with 3 kg of silage. The HG group required some silage supplementation during period 1 due to inclement weather;

however, the 3 kg difference of silage supplementation was maintained as HG received 2 kg per day and LG were allocated 5 kg per day. Both groups were offered the same amount of concentrate daily (average 2.2 kg cow^{-1} day^{-1}). Cows were assigned to either treatment as they calved and grazing began on 1 February 2021 until 6 June 2021 (18 weeks). The experiment was split into 3 periods: Period 1 (1 February – 13 March), Period 2 (14 March – 17 April) and period 3 (18 April – 6 June - carryover). All cows were managed the same during the carryover period, receiving the same DHA and no silage supplementation. Milk yields were recorded daily and milk composition weekly. Bodyweight was measured weekly. Pre-grazing herbage mass was recorded for each paddock and pre- and post-grazing sward heights were measured daily using a rising plate meter. DMI was measured using the N-alkane technique. Data were analysed using PROC MIXED in SAS 9.4 (SAS Institute Inc., Cary, NC, USA, 2002) treatment, week, period and associated interactions were included as fixed effects, week was included as a random effect and animal the subject.

Results and discussion

Grazed grass had a greater crude protein concertation (224 and 121 g kg^{-1} DM, respectively) and lower NDF (389 and 478 g kg^{-1} DM, respectively) compared to the grass silage, similar to Claffey *et al.* (2019). Treatment had a significant effect ($P<0.05$) on pre-grazing herbage mass in period 1 (HG=1,241 kg DM ha^{-1} and LG=1,003 kg DM ha^{-1}) due to the high and low OFCs that were established for the HG and LG treatment, respectively. Treatment had a significant effect ($P<0.001$) on DHA as HG were offered 2.3 kg DM cow^{-1} more than LG in period 1 and 3.55 kg DM cow^{-1} more in period 2. Total DMI was significantly higher ($P<0.05$) for HG in period 2, HG=17.34 kg DM cow^{-1} and LG=16.48 kg DM cow^{-1}; similar to Kennedy *et al.* (2011) who reported that offering grass silage during early lactation lowered DMI in cows.

Treatment had no significant effect ($P>0.05$) on milk yield in period 1 or 2 (HG=23.68 kg cow^{-1} and LG=23.26 kg cow^{-1}) or milk solids (HG=2.06 kg cow^{-1} and LG=2.05 kg cow^{-1}). There was, however, a significant effect ($P<0.05$) of treatment on milk protein concentration in period 2 (HG=35.1 g kg^{-1} and LG=33.3 g kg^{-1}). Previous studies have reported a similar reduction in milk protein concentration with the inclusion of grass silage in the diet due to lower energy intake and lower nitrogen content of silage compared to grass (Kennedy *et al.*, 2011). The LG treatment had a greater fat concentration (53.6 g kg^{-1}) compared to the HG treatment (49.9 g kg^{-1}) due to a higher fibre content in the diet with the inclusion of silage (Kennedy *et al.*, 2006). Cows on the HG treatment had significantly higher bodyweight from weeks 14 to 17 (average 481 kg and 508 kg for the LG and HG treatment, respectively). Claffey *et al.* (2019) also noted that cows grazing a high OFC, and subsequent greater total DHA, had a greater bodyweight than cows grazing a lower DHA.

Table 1. Effect of the high grass (HG) and low grass (LG) treatment on pre-grazing herbage mass, post-grazing sward height, grass and silage intake and total daily intake during the experimental period (1 Feb to 17 April).[1]

	Period 1		Period 2		SE	*P*-value		
	HG	LG	HG	LG		Trt	Period	Trt×Per
Pre-grazing herbage mass (DM ha^{-1})	1241	1003	1872	1879	89.07	NS	0.001	0.05
Post-grazing height (cm)	4.34	4.30	4.32	4.27	0.07	NS	NS	NS
Daily herbage allowance	10.60	8.30	15.11	11.57	0.36	0.001	0.001	NS
Silage (kg cow^{-1} day^{-1})	2.32	4.95	0.20	2.87	0.10	0.001	0.001	NS
Total intake (kg DM cow^{-1} day^{-1})	13.5	13.1	18.1	16.8	0.41	NS	0.001	0.05

[1] SE = standard error; NS = non-significant.

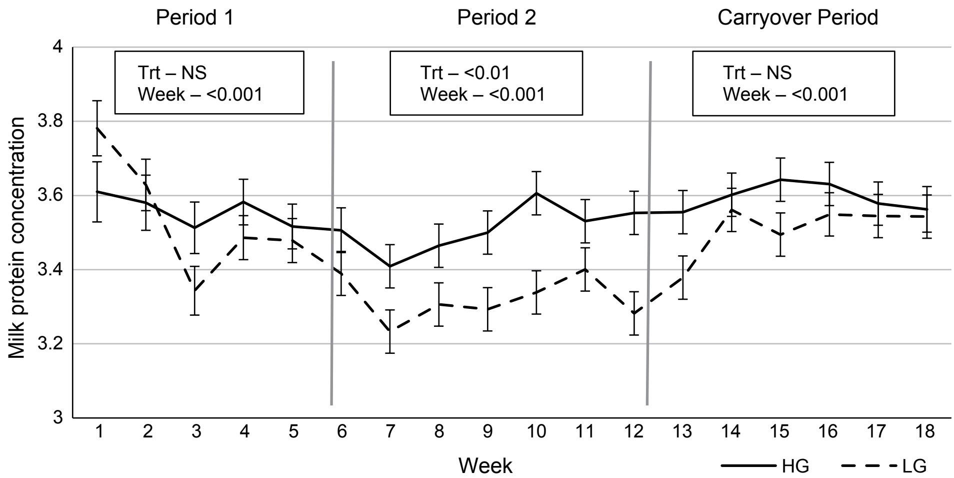

Figure 1. Milk protein % for the high grass (HG) and low grass (LG) treatments during period 1 (weeks 1-6), period 2 (weeks 6-12) and the carryover period 3 (weeks 12-18).

Conclusions

Supplementing grass silage in early lactation of pasture-fed dairy cows has a negative impact on DMI and milk protein concentration; however, milk fat concentration increased. A higher OFC will allow for increased DHA during the second half of the first rotation (Period 2) which will increase DMI. It is more beneficial to offer silage supplementation to cows during the first 6 weeks of lactation, as silage has less of a negative impact on milk protein concentration and DMI in weeks 1-6 (Period 1) than in weeks 6-12 (Period 2) of lactation.

References

Claffey A., Delaby L., Galvin N., Boland T.M. and Egan M. (2019) The effect of spring grass availability and grazing rotation length on the production and quality of herbage and milk in early spring. *Journal of Agricultural Science* 157, 434-448. https://doi.org/10.1017/S0021859619000613

Dillon P., Crosse S., Stakelum G. and Flynn F. (1995) The effect of calving date and stocking rate on the performance of spring-calving dairy cows. *Grass and Forage Science* 50, 286-299.

Kennedy E., O'Donovan M., Murphy J.P., O'Mara F.P. and Delaby L. (2006) The effect of initial spring grazing date and subsequent stocking rate on the grazing management, grass dry matter intake and milk production of dairy cows in summer. *Grass and Forage Science* 61, 375-384.

Kennedy E., O'Donovan M., O'Mara F.P., Murphy J.P. and Delaby L. (2007) The effect of early-lactation feeding strategy on the lactation performance of spring-calving dairy cows. *Journal of Dairy Science* 90, 3060-3070.

Kennedy E., Curran J., Mayes B., McEvoy M., Murphy J.P. and O'Donovan M. (2011) Restricting dairy cow access time to pasture in early lactation: the effects on milk production, grazing behaviour and dry matter intake. *Animal* 5, 1805-1813.

McEvoy M., Delaby L., Kennedy E., Boland T.M. and O'Donovan M. (2009) Early lactation dairy cows: Development of equations to predict intake and milk performance at grazing. *Livestock Science* 122, 214-221.

Effect of sward type on *in-vivo* dry matter intake, digestibility and methane output in sheep

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Abstract

Companion forages are gaining popularity alongside perennial ryegrass (*Lolium perenne* L.; PRG) in temperate regions due to improvements in animal performance. The aim of this study was to assess the effect of sward type on dry matter intake (DMI), diet digestibility and methane (CH_4) output in sheep. A 5×5 Latin square design experiment was undertaken to investigate five dietary treatments: PRG only or PRG plus white clover (*Trifolium repens* L.; WC), red clover (*Trifolium pratense* L.; RC), chicory (*Cichorium intybus* L.; Chic) or plantain (*Plantago lanceolata* L.; Plan) at a ratio of 75% PRG and 25% of the respective companion forage species on a dry matter (DM) basis. Twenty wether sheep (c.14 months and weighing 60.5 kg) were housed in metabolism crates across five periods. Individual DMI and faeces data were recorded daily. Methane (CH_4) measurements were collected using portable accumulation chambers (PAC). The DMI ranged from 1.55±0.038 (PRG) to 1.76±0.038 (PRG + Chic) kg DMI day^{-1}. Both PRG + RC and PRG + Plan had a lower digestibility than all other diets (793±0.003 digestibility (DMD), g kg^{-1} DM) while PRG + Chic had the highest DMD (804±0.003 DMD, g kg^{-1} DM). Methane yield was highest for animals offered PRG only (16.2±0.62 g CH_4 kg^{-1} DMI) and lowest for those consuming PRG + WC (11.8±0.61 g CH_4 kg^{-1} DMI). Results collected suggest that consumption of PRG plus a companion forage increased DMI while reducing CH_4 yield, possibly linked to diet quality as CH_4 production is directly correlated to the dietary constituents consumed by the animal.

Keywords: dry matter intake, digestibility, alternative forages

Introduction

Dry matter intake (DMI) and digestibility are key drivers of animal production from grazed forage diets (Hurley *et al.,* 2021). Perennial ryegrass is the most widely used forage in ruminant production systems across temperate regions due to its high digestibility and grazing tolerance. However, successful growth relies on high chemical fertilizer inputs making the sward less environmentally and economically viable as a feedstuff. Including companion forages such as red clover and white clover in the grazed sward gives the potential to increase animal performance from a lower chemical nitrogen input. Furthermore, studies have shown that lambs grazing multispecies swards have reduced parasite loads, thus requiring less need for anthelmintic treatment than those animals grazing perennial ryegrass monocultures (Grace *et al.,* 2019). While research has shown that legumes and forages, such as white clover, red clover, chicory and plantain benefit sheep systems, the physiological reasons behind these improvements in performance are yet to be fully investigated. Complementarities between grass and legumes are well known from an agronomic point of view but their interactions at animal level are relatively understudied (Niderkorn and Baumont, 2009). The objective of this experiment was to determine the effect of sward type on DMI, digestibility and enteric methane (CH_4) emissions in sheep.

Materials and methods

The study was performed at Teagasc, Athenry, Co. Galway, Ireland (N53.288024, W8.778380). An *in-vivo* 5×5 Latin square design experiment with five dietary treatments and five feeding periods was conducted using twenty Belclare wether sheep (ca. 14 months of age) with an average starting and final body weight of 60.5 kg and 67.2 kg, respectively. The five herbage treatments investigated were PRG only or PRG plus white clover (*Trifolium repens* L.; WC), red clover (*Trifolium pratense* L.; RC), chicory (*Cichorium intybus* L.; Chic) or plantain (*Plantago lanceolata* L.; Plan). Diets were formulated at 75% PRG and 25% of the respective companion forage species on a dry matter (DM) basis. *In-vivo* DMI was measured as described by Baumont *et al.*, (2004). Animals had *ad libitum* access to water. There were four sheep per dietary treatment per period. Herbage was harvested daily at 08:30 h using a push cutter bar mower. Approximately 50% of the herbage treatment was offered in the morning after cutting (09:00 h). The remaining dietary mix was refrigerated at 4 °C until it was offered at 16:00 h. The quantity of grass and forage mix was adjusted daily for DM prior to the evening feed. Herbage quantity offered to and refused by each sheep was recorded daily to calculate DMI. Individual animal daily faecal samples were weighed and frozen at -20 °C. Samples were defrosted and weighed prior to drying at 60 °C for 48 h or until dry in a Binder FED 720 (Binder GmbH, Tuttlingen, Germany) drying oven. Methane measurements were obtained on the final day of each measurement phase (n=5) using portable accumulation chambers (PAC) as described by Jonker *et al.* (2018). Animals were removed from feed and weighed a minimum of 1 hr prior to entering the PAC. Methane (ppm), oxygen (%) and carbon dioxide (%) measurements were obtained using individual chambers over a 50-min measurement period at three specific time points (0, 25 and 50 min after entry) using the RKI Eagle 2 monitor (Weatherall Equipment and Instruments Ltd, UK). Data were assessed for normality using PROC UNIVARIATE (SAS Institute Inc., Cary, NC, USA). Data were analysed using a linear mixed model, PROC MIXED, where herbage treatment, period and their interaction were included as fixed effects. Animal was included as the repeated and random effect.

Results and discussion

There was a significant effect of treatment diet on DMI (P<0.0001) whereby animals offered PRG plus a companion forage had higher DM intakes than animals offered PRG only (Table 1). The highest DMI was recorded for animals offered PRG + Chic, while animals offered PRG only had the lowest DMI (P<0.0001). Sheep are selective grazers with preference for feeds which can be ingested more quickly and which are highly digestible providing satiety and energy (Baumont *et al.,* 2000). This aligns with the higher observed intakes for diets with companion forage inclusion in this study. A similar digestibility (DMD) was recorded for PRG only, PRG + WC and PRG + Chic. White clover and chicory are known to have faster passage rates with quicker clearance from the rumen in comparison to PRG (Niderkorn and Baumont, 2009; Niderkorn *et al.,* 2017) allowing for increased intake despite the similar DMD. Both PRG + RC and PRG + Plan had lower digestibility than all other diets (P<0.05). While DMI increased with the inclusion of a companion forage, CH_4 yield was reduced in all scenarios (P<0.0001). The PRG-only diet ranked highest whilst PRG + WC ranked the lowest for CH_4 yield (Table 1). Similar results were seen by Hammond *et al.* (2011) where increased intakes reduced CH_4 yield.

Table 1. The effect of dietary treatment on dry matter intake (DMI), digestibility (DMD) and methane yield.[1]

Trait	PRG	PRG + WC	PRG + RC	PRG + Chic	PRG + Plan	SEM	*P* value
DMI (kg day^{-1})	1.55^a	1.72^c	1.74^c	1.76^c	1.63^b	0.038	0.0001
DMD (g kg^{-1} DM)	800^a	801^a	793^b	804^a	793^b	0.003	0.0222
Methane yield (g CH_4 kg^{-1} DMI)	16.16^a	11.76^c	13.00^b	13.82^b	14.31^b	0.623	0.0001

[1] SEM = standard error of the mean. Within row means with different superscripts are significantly different.

Conclusions

Results indicate that sheep DMI is positively influenced by the inclusion of companion forages thus partially explaining their role in improving animal performance. There were small differences seen when DMD was investigated, which is unsurprising given the high proportion of PRG in each diet. The reduction in CH_4 yield among all animals offered a companion forage treatment requires further investigation, albeit very positive in the development of potential CH_4 emissions mitigation strategies for ruminants.

Acknowledgements

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References

Baumont R., Prache S., Meuret M. and Morand-Fehr P. (2000) How forage characteristics influence behaviour and intake in small ruminants: a review. *Livestock Production Science* 64, 15-28.

Grace C., Lynch M., Sheridan H., Lott S., Fritch R. and Boland T. (2019) Grazing multispecies swards improves ewe and lamb performance. *Animal* 13, 1721-1729.

Hammond K., Hoskin S., Burke J., Waghorn G., Koolaard J. and Muetzel S. (2011) Effects of feeding fresh white clover (Trifolium repens) or perennial ryegrass (Lolium perenne) on enteric methane emissions from sheep. *Animal Feed Science and Technology* 166, 398-404.

Hurley M., Lewis E., Beecher M., Garry B., Fleming C., Boland T. and Hennessy D. (2021) Dry matter intake and *in vivo* digestibility of grass-only and grass-white clover in individually housed sheep in spring, summer and autumn. *Animals* 11, 306.

Jonker A., Hickey S.M., Rowe S.J., Janssen P.H., Shackell G.H., Elmes S., Bain W.E., Wing J., Greer G.J. and Bryson B. (2018) Genetic parameters of methane emissions determined using portable accumulation chambers in lambs and ewes grazing pasture and genetic correlations with emissions determined in respiration chambers. *Journal of Animal Science* 96, 3031-3042.

Niderkorn V. and Baumont R. (2009) Associative effects between forages on feed intake and digestion in ruminants. *Animal* 3, 951-960.

Niderkorn V., Martin C., Le Morvan A., Rochette Y., Awad M. and Baumont R. (2017) Associative effects between fresh perennial ryegrass and white clover on dynamics of intake and digestion in sheep. *Grass and Forage Science* 72, 691-699.

Productivity and nitrogen flows for grass systems targeting future biorefineries: a 5-year study in Denmark

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Abstract

High and stable supplies of biomass with large nitrogen (N) contents and low reactive flows are all essential parts of the supply chain to future biorefineries for feed protein. In a five-year study on sandy soil in Denmark, rotation of annual crops optimized for maximum biomass production and perennial grasses were compared with traditional systems of continuous maize or triticale, and a cereal crop rotation. The fertilized grasses and the optimized rotation significantly increased biomass N content and reduced leaching compared to the traditional systems. While providing guidelines for the design of innovative cropping systems to future biorefineries, these results will be linked with recently established field study within the Grass Tools project aiming to, among others, assess the usefulness of optical remote sensing at field and aerial scale to diagnose plant N status and biomass production based on a combination of model- and data-driven (machine learning) approaches.

Keywords: biomass, grass, nitrogen content, nitrate leaching, rotation

Introduction

Perennial herbaceous plants from the *Poaceae* family (grasses), in pure stands or in mixtures with species from *Fabaceae* (e.g. clovers), have recently been in research focus as potential candidates for biorefining of protein feed due to their ability to grow locally across Europe, utilize external resources efficiently (e.g. fertilization) and lengthily (e.g. radiation), ultimately providing large amounts of biomass with high protein (i.e. nitrogen, N) content, compared to annual cereals (Solati *et al.*, 2018). Moreover, short-term studies reveal reduced N-leaching and nitrous oxide emissions (e.g. Baral *et al.*, 2019; Manevski *et al.*, 2018), as well as increasing soil carbon and N contents (Chen *et al.*, unpublished data), whereas longer-term effects are yet to be reported. Such knowledge is essential to optimize the systems for duration and supply of biomass to biorefinery. The objective of this study was, therefore, to quantify biomass production and N leaching, based on medium time scale (5 years), from different cropping systems functionally divided on annual and perennial and optimized for biomass supply to biorefinery for feed protein. The outcome is linked with the recently started Grass Tools project that aims to provide robust knowledge to the economic sector in Denmark (https://projects.au.dk/grasstools/) on ensuring reductions of greenhouse gas emission and N leaching from grassland production.

Materials and methods

Field experiments started in 2013 in Denmark on sandy loam soil with two functional cropping systems tested for large biomass production, an optimized rotation with annual crops and perennial plants, both compared to 'traditional' cropping systems. The optimized rotation was based on four years and involved maize (*Zea mays* L.), sugar beet (*Beta vulgaris* L.), hemp (*Cannabis sativa* L.) and winter triticale (*Triticosecale*) as 'major' crops, as well as winter rye (*Secale cereale* L.) or grass-clover (*Festuca rubra* L. – *Trifolium repens* L.) as 'second' crops between the major crops. Perennial grasses were highly fertilised festulolium (× *Festulolium*, naturally occurring hybrid between *Festuca* and *Lolium*), low fertilised miscanthus (*Miscanthus* × *giganteus*) and unfertilized grass-legume mixture (industrial blend ForageMax 45 from DLF, Denmark). The traditional systems were common to many regions across Northwest Europe, i.e. continuous monocultures of maize, triticale, and a cereal rotation of spring barley

(*Hordeum vulgare* L.), winter barley and oilseed rape (*Brassica napus*). The systems were organized in a randomized incomplete-block design with four replicates (Manevski *et al.*, 2018). Biomass sampling was conducted regularly at each harvest and dry matter, N contents and N leaching were determined by standard methods.

To evaluate differences between means, linear mixed-effects models were built and fitted to annual data of biomass, N content and leaching with the *lmer* function in the 'lme4' package for R (R Core Team, 2013):

$$N_{ikn} = \mu + C_i + A_k + C_i \times A_k + P_n + e_{ikn}$$

where N is the dependent variable (either biomass, N content or leaching), μ is the overall mean, C_i is the effect of cropping system, A_k is the effect of year, Pn is the random effect of main plot, e is the residual variation.

Results and discussion

Year and cropping system, and their interaction, had significant influence on biomass, its N content and leaching (P<0.01; Table 1). Annual differences in biomass ranged from 21 to 32% between the mean value of optimized rotations (rotation 2), the perennial crops and the traditional system. On average, the biomass value did not differ between the optimized rotation, the fertilized perennial grasses and the continuous maize, which was the most competitive traditional system (Figure 1). In contrast, grass-legume mixture, triticale and cereal rotation had significantly lower biomass yield than other systems.

Festulolium had the highest biomass N and miscanthus the lowest; both were significantly different from each other and most of the other systems (Figure 2). Compared to the traditional system, optimized rotation and perennial grasses had higher N accumulation except for miscanthus. However, from the

Table 1. Significance levels of biomass, biomass nitrogen and nitrate leaching under different year and cropping systems.[1]

Factors	Biomass (Mg ha^{-1})	Biomass nitrogen (Kg N ha^{-1})	Nitrogen leaching (Kg N ha^{-1})
Year (Y)	**	**	**
Cropping systems (CS)	**	**	**
Y×CS	**	**	**

[1] Significant difference at ** P<0.01.

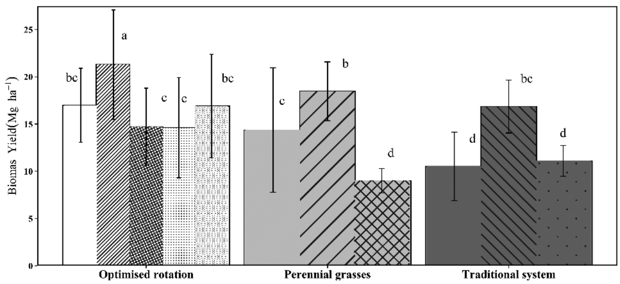

Figure 1. Mean (2013-2017; n=4) annual harvested biomass across different cropping systems. Different letters on the top bar indicate significant difference (P<0.05). The bars are indicated:optimized rotation1, optimized rotation2, optimized rotation3, optimized rotation4, optimized rotation mean, *M.* × *giganteus*, *Festulolium*, grass-legume DLF, triticale, maize, cereal rotation, from left to right, respectively.

leaching perspective, the traditional systems of maize and triticale had significantly higher values than the other systems. Miscanthus and grass-legume mixture had low leaching. These findings will be linked with remote sensing data obtained on recently established field by optical sensors (Yara N-Tester and multispectral camera mounted on unmanned aerial vehicle) to build non-parametric model for plant diagnosing N status at <10 kg ha^{-1} precision (Peng *et al.*, 2021).

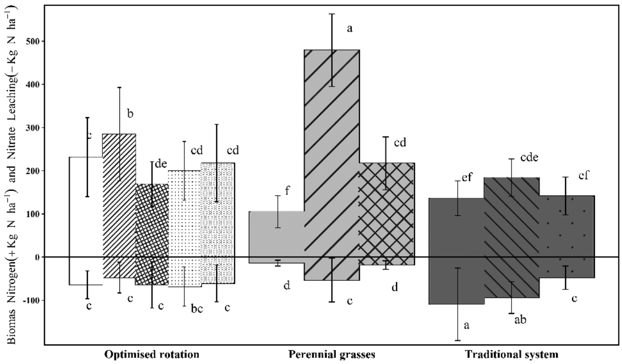

Figure 2. Mean (n=4; 2013-2017) annual biomass nitrogen and leaching across different cropping systems. Different letters on the top bar indicate significant difference (P<0.05). The bars are indicated:optimized rotation1, optimized rotation2, optimized rotation3, optimized rotation4, optimized rotation mean, *M.* × *giganteus*, *Festulolium*, grass-legume DLF, triticale, maize, cereal rotation, from left to right, respectively.

Conclusions

Despite year and system effects, on average over 5 years there was significantly lower amounts of leaching from perennial systems compared to all others.

Acknowledgements

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References

Baral K.R., Lærke P.E., and Petersen S.O. (2019) Nitrous oxide emissions from cropping systems producing biomass for future biorefineries. *Agriculture, Ecosystems & Environment* 283, 106576.

Manevski K., Laerke P.E., Olesen J.E. and Jorgensen U. (2018) Nitrogen balances of innovative cropping systems for feedstock production to future biorefineries. *Science of the Total Environment* 633, 372-390.

Peng J., Manevski K. ... and Andersen M.N. (2021) Random forest regression results in accurate assessment of potato nitrogen status based on multispectral data and the critical concentration approach. *Field Crops Research* 268, 108158.

Solati Z., Manevski K. ... and Lærke P.E. (2018) Crude protein yield and theoretical extractable true protein of potential biorefinery feedstocks. *Industrial Crops and Products* 115, 214-226.

Theme 5.
Initiatives for the transfer and co-construction of innovations on and for grasslands

EIP-AGRI: EU initiatives for the transfer and co-creation of innovations on and for grassland

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Abstract

The EIP-AGRI is dedicated to foster competitive and sustainable farming and forestry that 'achieves more and better from less' and therefore ensuring a steady supply of food, feed and biomaterials. The aim of this paper is to provide insight into the consideration of grasslands in the EIP-AGRI. It discusses focus groups (FG), operational groups (OG), multi-actor projects (MAP) and thematic networks (TN) with a link to grasslands. Grassland is one of the many agricultural topics covered by these EIP-AGRI key building blocks. In FG, grassland topics are well represented (they were addressed in 46.5% of all FG) in contrast to OG, where they are the subject of a marginal number of OG (7%). The OG cover a wide range of grassland related topics. The topics addressed by OG are less focused on environmental issues than, e.g. recommendations coming from the FG. Under the EIP initiatives, several grassland MAP/TN were implemented as H2020 research projects, where farmers are at the centre of practice-based innovation.

Keywords: European Union, operational group, focus group, multi-actor projects, thematic networks

Introduction

The European Innovation Partnership for Agricultural productivity and Sustainability (EIP-AGRI) was launched in 2012 in the context of the Innovation Union (EC, 2013) and is one of five EIPs. It contributes to the European Union's strategy 'Europe 2020' for smart, sustainable and inclusive growth. It is dedicated to foster competitive and sustainable farming and forestry that 'achieves more and better from less' and therefore in ensuring a steady supply of food, feed and biomaterials, developing its work in harmony with the essential natural resources on which farming depends (https://ec.europa.eu/eip/agriculture/en/about). This strategy sets the strengthening of research and innovation as one of its five main objectives and supports a new interactive approach to innovation (Wielinga, 2014). According to Faure *et al.* (2019), brokering functions and new services are key in this process to support actors to innovate by facilitating interactions for the co-production of knowledge, co-design of technologies, and identification of new institutional arrangements.

Having an idea is one thing; turning it into an innovation action is another. Different types of available funding sources can help get an agricultural innovation project started, such as the European Rural Development policy or the EU's research and innovation programme Horizon 2020. The EIP-AGRI contributes to integrating different funding streams so that they contribute together to the same goal and avoid unnecessary duplication of results. The EIP-AGRI adheres to the 'interactive innovation model' which brings together specific actors (e.g. farmers, advisers, researchers, businesses, NGOs and others) in agriculture and forestry to work together in multi-actor projects to find a solution for a specific issue or developing a concrete opportunity. Together these actors form an EU-wide EIP network (https://ec.europa.eu/eip/agriculture/en/node). Within this network, Operational Groups (OG), Multi Actors Projects (MAP) and Thematic Networks (TN) are all key building blocks. Focus Groups (FG) play a role in inspiring OG ideas and directions of MAP and TN. From January 2015, the European Rural Networks' Assembly was successfully launched as the main governance body of the ENRD and EIP-

AGRI Networks. The Assembly provides the strategic framework for the activities of the network support units and forms a platform to share stakeholders' priorities and concerns, covering all aspects of the Rural Development Policy 2014-2020. The aim of this paper is to provide insight into the consideration of grasslands in the EIP-AGRI.

Description and objectives of the four instruments

EIP-AGRI focus group

An EIP-AGRI Focus Group is a temporary group of 20 selected experts focusing on a specific subject formulated by the EU Directorate-General for Agriculture and Rural Development (DG AGRI) based on input from stakeholders (Focus Groups, 2016). The FG discusses and documents best practices and research results, explores practical innovative solutions to the problems or opportunities in the field that were listed, and draws on experience derived from related useful projects. The FG promotes the sharing and exchange of knowledge and experience among experts involved (researchers, farmers, advisers, etc.). Participants of an EIP-AGRI Focus Group are selected in a transparent manner from the pool of applications according to their competences based on documented expertise to support the work of the EIP-AGRI Focus Group. With the main emphasis on the expertise, the EIP-AGRI Focus Group is also composed with the intention of geographical balance and an adequate proportion as regards the fields of expertise and professional activity.

The FG results may have implications for dissemination and possible further directions for research that may help to solve practical problems in the agricultural or forestry sector and in rural areas. These may be related to production, processing, consumption, transport or other issues. The tangible output is focused on practical knowledge and where to get that knowledge as well as ideas for new OG. An EIP-AGRI FG is moderated by DG AGRI and several (usually three) experts of the EIP-AGRI Support Facility. An additional external expert can be invited on ad hoc basis, subject to the authorization of DG AGRI.

Objectives of each EIP-AGRI FG are:
1. To take stock of the state of play of practice in the field of the EIP-AGRI FG activity, listing problems and opportunities.
2. To take stock of the state of play of research in this field, summarizing possible solutions to the problems listed.
3. To identify needs from practice and propose directions for further research.
4. To propose priorities for innovative actions by suggesting ideas for practical OG or other project formats to test solutions and opportunities, including ways to disseminate the practical knowledge gathered.

EIP-AGRI operational group

OG are innovative projects tackling a certain (practical) problem or opportunity which may lead to an innovation (Operational Groups, 2016). Innovation in the agricultural and forestry sectors can be described in general terms as the introduction of something new (or renewed, a novel change) which turns into an economic, social or environmental benefit for rural practice. The OG approach makes best use of different types of knowledge (practical, scientific, technical, organizational, etc.) in an interactive way. Therefore, each OG is composed of those key actors that are in the best position to realize the project's goals, to share implementation experiences and to disseminate the outcomes broadly (such as farmers, advisers, researchers, businesses, NGOs, etc.). OG and Innovation Support Services are funded by the national or regional Rural Development Programmes (RDP). The EU member states or regions decide on the precise conditions to support innovation projects through their RDP.

OG can use support to develop new products, practices, processes and technologies in the agriculture, food and forestry sectors. Further possible areas of action include joint work processes, short supply chains, joint climate change actions and collective environmental projects. RDP support can cover the funding of the OG project but can also help to set up OG. Innovation brokers can help to develop a rough new idea into an innovation group ready to start a project (ISS, 2014). Bringing the right partners together and clear agreements on the concrete work plan and cooperation arrangements is key for the future success of OG projects.

Currently there are over 2200 OG set up in the EU, some of them related to grassland. A first evaluation of the OG was done in 2018 (Knotter *et al.*, 2019). However, no quantification and specific analysis for grassland-related OG has been done so far, to our knowledge.

H2020 projects: multi-actor projects and thematic networks

While OG are funded under the RDP, MAP and TN are supported by the Horizon 2020 Programme. Apart from the more classical research projects under Horizon 2020, the call includes several opportunities to support multinational interactive innovation projects in agriculture and forestry through TN and through MAP. The multi-actor approach should lead to innovative solutions that are more likely to be applied in the field, because those who need the solutions will be involved right from the start: from defining the questions, to planning, to implementing research work, to experiments and right up until possible demonstrations and dissemination. Openness to involve relevant groups operating in the EIP context (such as OG) has been strongly recommended in the work programme of Horizon 2020. The TN aim to:

1. Collect existing scientific knowledge and best practices on the chosen theme, and facilitate their use.
2. Develop easily understandable material for end-users like farmer, foresters, advisers, such as info sheets in a common format and audio-visual material. The material should be long-term available and easily accessible to end-users.

Analysis methods to evaluate the innovation effort in relation to grassland

Analysis of the Focus Groups (FG)

A first screening of FG and OG led to a list of grassland-related topics, potentially suitable to include most grassland-related aspects addressed by FG, OG and research projects. These aspects are: grassland farming profitability, grazing management, biodiversity and/or nature conservation, forage quality, smart grassland management, climate change mitigation, enhancement of functional diversity, quality and marketing of grassland-based animal products, meadows management, permanent grassland, fertilization and nutrient cycling, temporary grassland/leys, other, weed control, grass refinery, mechanization and agroforestry. For the FG the final reports of 43 FG were analysed according to these grassland-related topics based on their state of play in December 2021 (EIP-AGRI, 2021). The following classification of FG by topics was undertaken: (1) grassland-related FG (8-15 grassland-related topics addressed in the final FG report); (2) grassland-marginally related FG (2-7 topics); and (3) grassland-not related FG (0 topics).

Analysis of the EIP-AGRI database of operational groups

The EIP-AGRI maintains an extensive, freely accessible database (https://ec.europa.eu/eip/agriculture/en/find-connect/projects), which aims to provide basic information about running and completed OG, in order to foster the cooperation and ensure cross-fertilization between projects sharing common aims at a European level. To select and analyse the grassland-related OG only, a search of the database (download on 29 January 2022) was done by using some potentially grassland-related keywords ('grassland', 'grazed', 'ensiling', 'fodder', 'grazing', 'pasture', 'meadows', 'forage', 'silage', 'ruminants'). Following this first screening, the descriptions of the remaining OG were evaluated according to the description of the aims and of the undertaken actions to achieve the stated objectives. All cases were retained that were either explicitly related to grassland or that included actions having a direct relationship to grassland. For instance, OG dealing only with animal product-quality were dropped unless grassland-related actions were also described. In total, 164 grassland-related cases were retained for further analyses and each case was assigned to all grassland-related topics addressed (as defined for the FG analysis). Other descriptors already included in the EIP-AGRI database (starting year, location, lead partner category) were used for the analysis as well. Moreover, each case, depending on the location of the OG, was assigned to one of the biogeographical regions of Europe according to the European Environment Agency (2017). Austria and Slovenia were assigned to the Alpine region, Belgium, Ireland, Netherlands and United Kingdom to the Atlantic region, Finland, Lithuania and Sweden to the Boreal region, Poland to the Continental region, Portugal and Spain to the Mediterranean region, and Hungary to the Pannonian region. For France, Germany and Italy, because of their biogeographical heterogeneity, the single OG were assigned to the respective biogeographical region based on their location. In order to analyse the relationship between biogeographical regions, topics and lead partner categories, a principal component analysis (PCA) was performed expressing the levels of the three factors as binary variables.

Online survey of operational groups

An anonymous online survey was conducted from 28 January to 10 February 2022 to gain a more detailed insight into aspects of the grassland-related OG not covered by the EIP-AGRI database. The coordinator of each OG was invited via e-mail to take part in a survey, implemented in Microsoft Forms, and to provide information about the topics addressed (according to the same list used for the analysis of FG and the EIP-AGRI database), the animal categories addressed (cattle, sheep, other animal species and no animals species; goats were merged with other animal species because of the too-low number of respondents), the kind of animal products addressed (milk and dairy products, meat, other animal products and no animal products), the stakeholder categories involved in the OG (developer of innovation, farmer, researcher, extension service/adviser, facilitator agent/innovation broker, veterinarian, local administration, policy maker, industry (supply and processing), marketing organization, retail, non-governmental organization (NGO), consumers' organization, farmers union, professional school for agriculture, student/pupil, journalist, other), the occurrence of international cooperation and, for completed OG only, the request to self-assess on a 1-10 scale (whereas 10 is the best result) the achievement of the stated aims and the achievement of the targeted impact on the local agricultural practice.

Analysis of multi-actor projects and thematic networks

The CORDIS database (www.cordis.europe.eu) was searched for H2020 MAP and TN with a relation to grasslands using the key words: 'grassland', 'grazed', 'ensiling', 'fodder', 'grazing', 'pasture', 'meadows', 'forage', 'silage', 'ruminants'. Further criteria for inclusion in a list of relevant MAP/TN was an EU contribution >500,000 euro. Following this first screening, the descriptions of the remaining MAP and TN were evaluated in a similar way as the OG. All cases were retained which were either explicitly related to grassland or contained actions having a direct relationship to grassland.

Consideration of grasslands in the EIP-Agri: focus groups and grassland

The analysis following the classification criteria resulted in nine grassland-related FG (20.9%), 11 marginally grassland-related FG (25.6%) and 23 non-grassland-related FG (53.5%). In the grassland-related FG the operational objectives for the research and MAP/TN that help to solve practical problems in grassland management were defined (Table 1).

Table 1. EIP focus groups with relations to grassland and their main results.

Topic of focus group	Main question	Main issues/operational goals for research that help to solve practical problems in grassland management
Profitability of permanent grassland (2014-2015)	How to manage permanent grassland in a way that combines profitability, carbon sequestration and biodiversity?	• Defining grassland typology in relation to biodiversity and productivity • Achieving grassland production and quality to match livestock needs • Benchmarking grassland dry matter production and its utilization at regional and national levels • Increasing grassland functionality by diversifying sward plant composition • Increasing resource efficiency to improve profitability and sustainability • Differentiating grass-based products for higher market value • Evaluate environmental impacts of grassland-based systems using Life Cycle Thinking
High Nature Value – Farming profitability (2014-2016)	How to make HNV farming more profitable without losing the HNV characteristics?	• Better access to semi-natural land for grazing (quantity and quality) • Making more efficient use of semi-natural fodder resources • Complementary use of HNV and semi-intensive land • Developing better technical and management solutions for HNV farming, e.g. HNV grassland • Increasing selling price of HNV products, e.g. grass-based products, and improving access to markets
Grazing for carbon (2017-2018)	How to increase the soil carbon content from grazing systems?	• Improving the understanding of strategies promoting better soil C management in grazed grasslands • Establishing monitoring schemes for C storage • Developing incentives to promote the adoption of good and appropriate grazing systems • Reaching equilibrium of soil organic content through optimal grazing management in different soil conditions • Providing guidelines for good grazing management/education/knowledge dissemination
Livestock emissions – Reducing emissions from cattle farming (2016-2017)	How to reduce cattle livestock emissions in a cost-effective way for farmers?	• Using grazing as a management tool to reduce ammonia emissions • Improving management practices and breeding/adopting new species and cultivars for obtaining the quantity and quality of feed to animals and also, in some regions and systems, enhance soil carbon storage • Combining controlled rotational grazing with precision management of grassland and monitoring of animal parameters • Monitoring nutrient composition and feed intake for grazing cattle • Development and testing of decision tools to improve N-efficiency
Water and agriculture (2015-2016)	What farm level adaptation strategies exist or can be developed to deal with water scarcity?	• Increasing water productivity by improvement in pasture and grazing management and feeding, or in animal health, and therefore an increase in the system's output • Improving water holding capacity and water infiltration by increasing soil organic matter: conservation agriculture and maintaining soil surface covered with residues, mulching, cover cops or green manure, and crop rotation using leys • Increasing irrigation efficiency monitored by remote sensing and calibrated and evaluated for local conditions • Increasing farm resilience under water scarcity by natural water retention measures: grasslands, buffer strips, agroforestry
Agroforestry: introducing woody vegetation into specialised crop and livestock systems (2016-2017)	How to develop agroforestry as a sustainable farming system which can boost agricultural productivity and profitability?	• Developing agroforestry system in farms with plantations of high value trees related with meadow or grazed orchards • Improving agroforestry system in high nature value farms related with mountain pastoralism • Differentiating grass-based products from agroforestry systems for higher market value • Managing silvopastoral farming for shaping landscape structure to prevent wind and water erosion and enhance water balance

Table 1. Continued.

Robust and resilient dairy production systems (2016-2017)	How to create good conditions for dairy cattle husbandry in different production systems?	• Farm management strategies to increase the robustness and resilience of dairy farming systems • Assessing feed quality will ensure that daily rations are prepared based on the real chemical composition of on-farm feeds • Choosing the right type or breed of cow for the right system • Introducing dry matter, energy and protein self-sufficiency evaluation on-farm as indicator of farm robustness and resilience • Better standardization and comprehensiveness of life cycle calculation including the valuation of ecosystem services • Implementing measures to increase farm robustness and resilience and responding to general or local consumer requirements
Mixed farming systems: livestock/cash crops (2015-2016)	How to develop livestock / cash crop interactions and promote their benefits as a sustainable alternative to farm or territorial specialization?	• Using more efficiently crops and grasslands to feed animals and fertilizing their fields with manure from the animals • Recoupling nitrogen and carbon cycle through legumes/grasslands in arable rotations • Enhance regional integration of mixed farming systems by the diversification of crops and grasslands produced on-farm • Including woody vegetation, conservation agriculture and permanent grasslands to improve existing mixed farming systems and their impacts on landscape mosaic
Sustainable beef production systems (2020-2021)	How can grass-based beef production systems, based on agro-ecology principles, remain sustainable?	• Applying of novel holistic assessment methods and tools as important innovations for assessing the real value of products deriving from grass-based beef systems • Improving grassland resource management and plant diversity to produce beef with low resource input, while providing high output in terms of ecosystem services and public goods • Developing the grazing management and stocking density for grass-based beef systems depend on local environment • Using the new decision support tools to improving herd and grazing management, soil health and feed quality • Raising awareness on sustainable beef production by promoting its evidence-based benefits on human health, landscapes, biodiversity, rural communities and keeping European traditions alive • Setting up methods and techniques for differentiating grass-based beef from beef from other systems, including certification/labelling

The goals of Table 1 relate to different types of grasslands, different intensity of their use, profitability in terms of the production of herbivores, as well as environmental issues. Climate change mitigation, grazing management and biodiversity and/or nature conservation were the most frequently addressed topics (70 to 80%), followed by permanent grassland, forage quality, enhancement of functional diversity, agroforestry and fertilization and nutrient cycling with frequencies of at least 50 and up to 65% (Table 2). Climate change mitigation, agroforestry, biodiversity and/or nature conservation, forage quality, enhancement of functional diversity and fertilization and nutrient cycling were much more frequently addressed in FG than in OG (more than 25% more frequently, according to the results of the OG survey).

Grassland-related topics were used marginally in 11 other FG (Table 3). They indicate that grasslands, with regard to their economic and environmental aspects, are important for the analysis and search for innovative solutions in other agricultural areas. It also shows the diversity of services that can be expected from grassland.

The proportion of grassland within the European agriculture area and its food system is substantial (Huyghe *et al.*, 2014; Krause *et al.*, 2018). When looking at the presence of grassland within the 43 FG, we might say that with 20 FG addressing grassland it is well represented.

Table 2. Number and percentage of focus groups and operational groups according to the EIP-AGRI database and to the operational groups online survey addressing topics from the defined list.

Topic	FG (n)	FG (%)	EIP-AGRI database (n)	EIP-AGRI database (%)	OG Survey (n)	OG Survey (%)
Grassland farming profitability	7	35.0	46	28.0	9	20.0
Grazing management	14	70.0	44	26.8	25	55.6
Biodiversity and/or nature conservation	14	70.0	40	24.4	16	35.6
Forage quality	12	60.0	37	22.6	13	28.9
Smart grassland management	4	20.0	33	20.1	7	15.6
Climate change mitigation	16	80.0	26	15.9	11	24.4
Enhancement of functional diversity	11	55.0	22	13.4	11	24.4
Quality and marketing of grassland-based animal products	6	30.0	21	12.8	6	13.3
Meadows management	9	45.0	20	12.2	12	26.7
Permanent grassland	13	65.0	19	11.6	25	55.6
Fertilization and nutrient cycling	10	50.0	19	11.6	10	22.2
Temporary grassland/leys	7	35.0	16	9.8	10	22.2
Other	0	0.0	16	9.8	9	20.0
Weed control	5	25.0	7	4.3	2	4.4
Grass refinery	2	10.0	7	4.3	4	8.9
Mechanization	5	25.0	6	3.7	6	13.3
Agroforestry	11	55.0	5	3.0	2	4.4

Table 3. EIP focus groups with grassland mentioned in their results.

Topic of focus group	Issue related to grassland
Organic farming – Optimising arable yields (2013-2014)	Importance of new crops combinations including leys, legumes, mixed farming, agroforestry
Protein crops (2013-2014)	Leaf protein from alfalfa and forage grasses as a source of high protein content products
Soil organic matter content in Mediterranean regions (2014-2015)	Improving the soil organic matter content of soils by using grass/legume species in time (crop rotation, use of cover crops) or space (intercropping or agroforestry systems, e.g. grassed orchards or vineyards)
Optimising profitability of crop production through Ecological Focus Areas (2014-2015)	Using of grassy or flower strips for enhance landscape features and contribute to the profitability of arable crop production
Carbon storage in arable farming (2017-2018)	Perennial grass leys for providing soil carbon through the grass roots and through the promotion of the soil organisms that are boosted by this
Enhancing production and use of renewable energy on the farm (2017-2018)	Using grass biomass for biogas production and biomass from agroforestry systems, which constitute potential income or cost reduction for farmers, if used as fuels to produce heat or electricity
New feed for pigs and poultry (2018-2019)	Implementation of green biomass (grass/clover) and protein extract made from grass/clover as a new feed option
Bee health and sustainable beekeeping (2019-2020)	Attracting pollinators thanks to multi-functional buffer zones surrounding fields composed of various herbs and grasses
Protecting agricultural soils from contamination (2019-2020)	Managed grazing that builds organic matter in the soil as a practice of regenerative agriculture
Wildlife and agricultural production (2020-2021)	Grassland as forage alternatives to wildlife and a practical strategy to reduce and avoid damages in arable crops
Climate-smart (sub)tropical food crops in the EU (2020-2021)	Maintaining grasslands or including grassland into crop rotations in order to store more carbon in soils, apart from agroforestry, as a practice for conservation agriculture in (sub)tropical zone

Consideration of grasslands in the EIP-Agri: operational groups and grassland

Analysis of the EIP-AGRI database on operational groups

The percentage of grassland-related OG between 2015 and 2020 was found to range between 3.8% and 9.1% of the total number of OG starting in the respective year, with a peak achieved in 2018 with 44 OG, corresponding to 9.1% of the total number of OG starting that year (Table 4). Apparently, both in terms of number of OG and of their percentage, there was an increase of the grassland-related OG until 2018 followed by a decrease to values around 6%.

In the EIP-Agri database, about half of the OG were located within the Atlantic region (78/164 OG), followed by the Continental region with about 20% of the cases (32 OG) (Table 4). The Alpine region, despite its relatively small area, was well represented with about 14% of cases (17 OG). The Mediterranean (23 OG), Boreal (12 OG) and Pannonian (2 OG) regions seem to be underrepresented, compared to their geographical area, whilst the opposite is apparently true for the Atlantic region, although this is the most favourable area for grassland farming from a climatic and (in most cases) topographic point of view.

Within the EIP network, the OG are multi-actor approaches. According to Kelly (2020), they are better for complex innovations; they address real issues, deliver clearer messages, more consistently, with less risk of contradiction and loss of relevance to targeted users.

The OG were most frequently led by advisers (34.1%) or researchers (31.7), whereas farmers, NGO representatives or small or medium enterprises led them with low frequencies (7.9%, 6.7% and 4.3%, respectively) (Table 5).

Concerning the OG topics, the most frequently addressed form of grassland utilization was grazing management, which was part of the OG concept and activities in slightly more than one third of the OG, whilst meadows management was addressed at about half of this frequency (Table 2). The most frequently addressed topics besides grazing management were grassland farming profitability, biodiversity and nature conservation, forage quality and climate change mitigation, with frequencies ranging between 28.0 and 15.9%. It is interesting that these topics represent a mixture of basic issues pivotal for a rational and profitable grassland management (and, in turn, livestock farming) and other emerging topics more related to environmental aspects. The frequency of all other topics other than the ones mentioned above was lower than 15% and particularly low values were observed for weed control, grass refinery,

Table 4. Number and percentage of grassland-related operational groups (OG) 2014-2019.[1]

Starting year	All OG (n)	Grassland-related OG (n)	Grassland-related OG (%)	Grassland-related OG included in the online survey (n)[2]	Grassland-related OG included in the online survey (%)
2014	1	0	0.0	0 (0)	
2015	79	3	3.8	1 (1)	33.3
2016	232	20	8.6	4 (4)	20.0
2017	409	30	7.3	8 (5)	26.7
2018	482	44	9.1	9 (5)	20.5
2019	530	32	6.0	9 (3)	28.1
2020	367	23	6.3	8 (0)	34.8

[1] Because of incomplete data from 2021 onwards, only data until 2020 are shown.

[2] In brackets the number of already concluded OG.

Table 5. Lead partner category according to the EIP-AGRI database and stakeholder categories acting within the operational groups according to the online survey (reclassified, where possible, according to the stakeholder categories used in the EIP-AGRI database).

Partner category	Lead partner EIP-AGRI database (n)	Lead partner EIP-AGRI database (%)	Stakeholder categories OG survey (n)	Stakeholder categories OG survey (%)
Adviser	56	34.1	32	71.1
Researcher	52	31.7	36	80.0
Other	25	15.2	22	48.9
Farmer	13	7.9	44	97.8
Representative of an NGO	11	6.7	16	35.6
Small and medium enterprises	7	4.3		
Industry			19	42.2
Civil servant			6	13.3

mechanization and agroforestry. Similar, relatively low frequencies were also observed for permanent grassland and temporary grassland/leys.

While for the FG, grassland topics were well represented, for the OG this was just a marginal 5%. We did not analyse why this representation was so low. It could be due to the selection of allowed/desired topics in the calls for OG by the RDP in the different member states, or to other agricultural sectors being more into multi-actor approach projects with producers. Another possible explanation could be that grassland farmers, advisers and researchers are less interested in participating in OG. This situation may be influenced by the poorly developed innovation brokering system related to grassland in many European countries (Goliński *et al.*, 2018), which means that the topic of grassland is less represented in OG than in other sectors of agriculture. This fact deserves further elucidation and, as grassland plays an important role within European circular and sustainable food systems, more R&D with involvement of farmers seems advisable. Concerning the analysis of the OG topics addressed in the EIP-AGRI database, it is worth remembering that some of them might not have been mentioned in the short description of the OG, as they were obvious to the authors of the abstract, and therefore escaped from being included in the analysis. Moreover, discrepancies between the topic frequencies of FG and OG might be due to the fact that the FG calls often required some set of topics to be mandatorily addressed and being usually related to the EU policies like the European Green Deal. The same applies to the OG calls within the frame of the RDP of some member states.

The first two components of a PCA accounting for the biogeographical region, the lead partner category and the addressed topics according to the short description of the OG in the EIP-AGRI database could explain the variability of the observations only to a small extent (23.8%) (Figure 1). Along the first component, advisors as lead partner were related mainly to the Atlantic region, whilst researchers in this role were mainly related to the Alpine and Mediterranean regions and to the topics of quality and marketing of grassland-based animal products. This may be related to the requirement for sound arguments to justify higher product prices needed under unfavourable production environments to sustain farm profitability. Along the second component, biodiversity and/or nature protection was found to be related to climate change mitigation, profitability of grassland farming, grazing management, permanent grassland and NGO as lead partner category, as opposed to smart grassland farming, underlining how grazing is regarded as part of a viable strategy to meet high environmental standards in combination with economic sustainability of farms. Finally, a quite close correlation was found between the Continental biogeographical region and the forage quality topic, both with moderately high loadings on Component 1 and Component 2.

The OG are addressing most of the time several topics at once. The PCA clearly shows the logic combination of the different regions. Combinations as profitability & biodiversity, alpine & quality and marketing of animal products, permanent grassland & grazing are obvious. Grassland is a source of many ecosystem services, both provisioning as well as environmental and social (Huyghe *et al.*, 2014), and the PCA provides evidence that OG take different categories of ecosystem services into account at the same time.

Based on our grassland-related OG analysis, we see the topics mixture of basic issues for the rational and profitable grassland management (and, consequently, livestock farming) on the one hand, and other emerging topics more related to environmental aspects on the other hand. It is probably related to the new awareness and/or pressure of public opinion or politics and is in line with the principles of the European Green Deal (Guyomard *et al.*, 2020). In this way, we can conclude that grassland is a friendly sector of agriculture that meets the expectations and objectives of the agricultural policy in the new EU financial perspective.

Online survey

The OG database only presents the objectives of the project. The project results are not uploaded and also background information is lacking. In our opinion, structured feedback requested to the OG participants and aiming at gaining specific knowledge about strengths and weaknesses of the process and its outcome would be beneficial to designing the strategy for future actions.

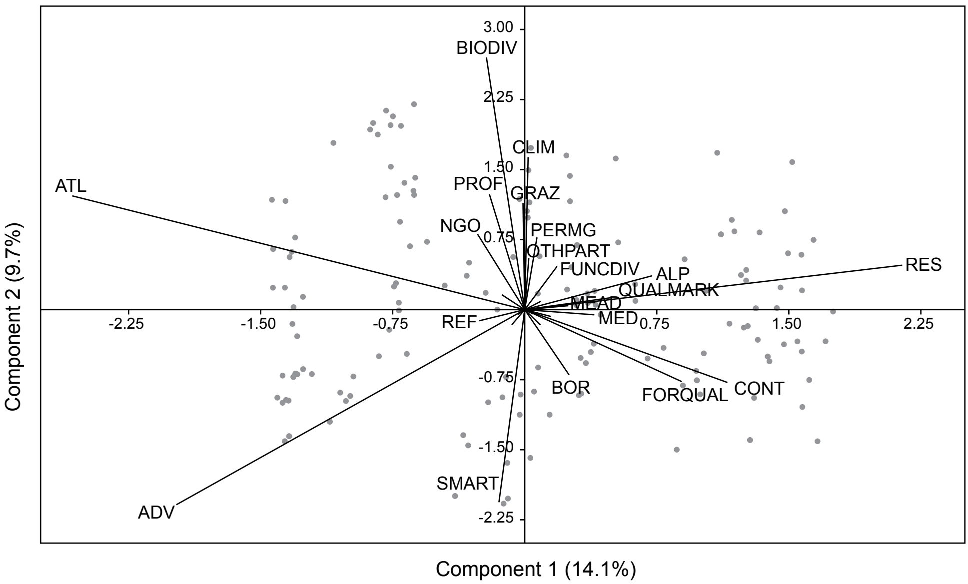

Figure 1. PCA biplot of the first two components, accounting for the region, the lead partner category and the addressed topics according to the short description of the OG in the EIP-AGRI database. The results are shown on eigenvalue scale. The labels of short vectors at axes intersection are omitted for better readability. Biogeographic region: ALP = Alpine, ATL = Atlantic, BOR = Boreal, CONT = Continental, MED = Mediterranean; Topic: BIODIV = biodiversity and/or nature protection, CLIM = climate change mitigation, FORQUAL = forage quality, FUNCDIV = functional diversity, GRAZ = grazing management, MEAD = meadows management, PERMG = permanent grassland, PROF = profitability of grassland farming, QUALMARK = quality and marketing of animal products, REF = grassland refinery; Lead partner category: ADV = advisers, OTHPART = other partner, NGO = representative of an NGO, RES = researcher.

The online survey allowed us to gain a more detailed insight. There was a total of 49 respondents to the online survey, corresponding to 29.9% of the total number of grassland-related OG. An acceptable proportion of respondents took part in the survey for each start year (ranging within relatively similar proportions of between 20.0 and 34.8%, without any consistent temporal trend), suggesting that the results of the survey might be representative for the grassland-related OG screened from the EIP-AGRI database (Table 2). However, we decided not to attempt an interpretation of the evaluations (mean scores: 8.3±1.2 standard deviation (SD) for the achievement of the stated aims and 7.1±1.9 SD for the achievement of the targeted impact on the local agricultural practice), as they were just 11.0 and 10.4%, respectively (n=18 and n=17) of the total number of grassland-related OG and, most of all, because a high proportion of OG after 2017 was still running at the time of the survey, resulting in their missing evaluation and leading to a temporal bias in the data set.

Cattle was the animal category most frequently addressed (in about three-quarters of cases), followed by sheep, and other animal species, with similar frequencies (15.6 and 17.8%, respectively). The percentage of grassland-related OG not addressing any animal species was low (11.1%). This was in line with expectations, because there is an obvious link between forage production and its utilization by ruminants.

There was a relatively high number of stakeholder categories per OG (4.9±1.9 SD). Farmers were represented (either as farmers themselves or as farmers unions) in each OG, as their involvement in the OG was indeed required by OG definition (Table 6). It is striking, nevertheless, that they seldom acted as lead partner (Table 5). Most OG are coordinated by researchers (mainly in the Alpine and Mediterranean biogeographical regions) or by advisers (mainly in the Atlantic region). Researchers and advisers were the second and third most represented category (80.0 and 62.2%, respectively, but veterinarians with 15.6% also could be ascribed to the category of advisers). It is notable that developers of the innovation and industry were frequently involved in the OG (53.3 and 40.0%). NGOs and educational institutions (also including students/pupils) relatively often took part in the OG, with frequencies ranging between 28.9% for the agricultural schools and 15.6% for students/pupils. All in all, the data suggest that a good connection of scientific and practice-oriented institutions, covering the production chain up to industry, was achieved. It has to be taken into account that higher values for all stakeholder categories are observed

Table 6. Stakeholder categories acting within the Operational Groups according to the online survey.

Stakeholder category	Survey (n)	Survey (%)
Farmer	44	97.8
Researcher	36	80.0
Extension service/adviser	28	62.2
Developer of innovation	24	53.3
Industry (supply and processing)	18	40.0
Professional school for agriculture	13	28.9
Non-governmental organization (NGO)	9	20.0
Other unknown categories	8	17.8
Facilitator agent/innovation broker	7	15.6
Veterinarian	7	15.6
Student/pupil	7	15.6
Farmers union	6	13.3
Local administration	5	11.1
Other known categories[1]	9	20.0

[1] The category 'others' includes 'policy maker', 'marketing organization', 'consumers' organisation', 'journalist' and 'retail', all with two observations each, except the latter with just one observation.

in the OG survey in comparison to those extracted from the EIP-AGRI database due to the fact that in the OG survey not only the lead partner is listed, but all stakeholder categories taking part in the OG.

An encouraging figure was found also for transnational cooperation, which was implemented in slightly more than a quarter of cases (28.9%). OG also involved, on average, 1.2±0.4 SD other countries besides that of the lead partner.

Consideration of grasslands in the EIP-Agri: MAP, TN and grasslands

From 2016 onwards, Horizon 2020 (H2020) introduced MAP and TN following the interactive model promoted by the EIP-AGRI. This puts farmers at the centre of practice-based innovation in research projects. New and existing scientific knowledge was used to produce implementable solutions for farmers that were shared across a broad network. The H2020 programme supported research related to grassland via several TN and MAP:

- EuroDairy (TN, 2016-2019) was a Europe-wide thematic network supporting a sustainable future for EU dairy farmers. It mobilized 120 innovating Pilot Farmers to increase the economic, social and environmental sustainability of dairy farming. It focused on four key issues: socio-economic resilience, resource efficiency, animal care and biodiversity (Keatinge and Korevaar, 2017).
- HNV-Link (TN, 2016-2019; High Nature Value Farming: Learning, Innovation and Knowledge) was dedicated to developing and sharing innovations that support high nature value farming systems and communities by simultaneously improving their socio-economic viability and environmental efficiency. Poux *et al.* (2018) concluded that the success of HNV innovation brokering was depending on the available means (financial allocation and thus human means for accompanying the local dynamics).
- SheepNet (TN, 2016-2019; Sharing Expertise and Experience towards sheep Productivity through NETworking) was designed to stimulate knowledge exchange between research and stakeholders to widely disseminate best practices and innovations with the objective of increasing ewe productivity. Nutrition/grassland management was identified as the most important challenge for reaching this aim (Keady *et al.*, 2018).
- Inno4Grass (TN, 2017-2019; Shared Innovation Space for Sustainable Productivity of Grasslands in Europe) aimed to bridge the gap between practice and science communities to ensure the implementation of innovative systems on productive grasslands. Grassland-related knowledge was made available for local conditions by a methodology to collect farmers' innovative ideas and to stimulate collaboration among various stakeholders (farmers' groups, extension services, education and research) (Krause *et al.*, 2018).
- The main topic of AFINET (TN, 2017-2019; Agroforestry Innovation Networks) was the promotion of agroforestry to foster climate change. AFINET followed a multi-actor approach linked to nine Regional Innovation Networks (Villada *et al.*, 2018).
- EuroSheep (TN, 2020-2023; European Network for interactive and innovative knowledge exchange on animal health and nutrition between the sheep industry actors and stakeholders) is about dairy and meat sheep production with the objective to exchange existing knowledge between farmers and stakeholders at all stages of the supply chain.
- iSAGE (MAP, 2016-2020; Innovation for Sustainable Sheep and Goat Production in Europe) was set up to improve the sustainability and innovative capacity of the sheep and goat sector in Europe. It was shown that all farm types in all countries are facing challenges regarding their overall sustainability (Paraskevopoulou *et al.*, 2020).
- SUPER-G (MAP, 2018-2023; Developing SUstainable PERmanent Grassland systems and policies) co-develops permanent grassland systems that will be effective in optimizing productivity, whilst supporting biodiversity and delivering a number of other ecosystem services. It applies a multi-actor approach, working with farmers; land owners/managers and their advisers; third sector and

civil society groups; non-governmental organizations (NGOs) and researchers, policy and business (Newell-Price *et al.*, 2022; in these proceedings)

- The GO-GRASS (MAP, 2019-2023: Grass-based circular business models for rural agri-food value chains) project aims to unlock the overlooked potential of grassland across Europe and to create new business opportunities in rural areas by developing a set of small-scale bio-based solutions to produce protein concentrates, biochar, animal bedding and paper and carton products from grass and green fodder. Orozco *et al.* (2021) conclude that capacity building and alignment efforts need to be strengthened and coordinated at local and higher levels to enable the replication and scale-up of these novel grass-based businesses in Europe and beyond.

Conclusions

Grassland is one of the many agricultural topics covered by the EIP-AGRI key building blocks. In FG, grassland topics are well represented. This is in contrast to OG, where grassland topics only have a marginal number. The discrepancies between the topic frequencies of FG and OG might be due to the fact that the FG calls often required some set of topics to be mandatorily addressed and that they are usually related to the EU policies like the European Green Deal. This is also true for the OG calls within the frame of the Rural Development Programs of some member states. The topics addressed in the FG, OG, MAP and TN show that grassland is an environmentally friendly sector of agriculture that meets the expectations and objectives of the agricultural policy in the new EU financial perspective.

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References

EC (2013) *Innovation Union, A pocket guide on a Europe 2020 initiative*. Publications Office of the European Union, Luxembourg, Luxembourg, 16 pp. Available at: https://ec.europa.eu/eip/agriculture/sites/default/files/innovation-pocket-book_en.pdf

EIP-AGRI (2021) EIP-AGRI Focus Groups state of play December 2021, 4 pp. Available at: https://ec.europa.eu/eip/agriculture/sites/default/files/focus_groups_state_of_play_december_2021_final.pdf

European Environment Agency (2017) *Biogeographical regions in Europe*. Available at: https://www.eea.europa.eu/data-and-maps/figures/biogeographical-regions-in-europe-2.

Focus Groups (2016) *Focus Groups: Sharing knowledge to inspire action.* EIP-AGRI Service Point, 4 pp. Available at: https://ec.europa.eu/eip/agriculture/sites/default/files/eip-agri_brochure_focus_groups_2016_en_web.pdf

Faure G., Knierim A., Koutsouris A., Ndah H., Audouin S., Zarokosta E. and Heanue K. (2019) How to strengthen innovation support services in agriculture with regard to multi-stakeholder approaches. *Journal of Innovation Economics and Management* 28, 145-169.

Goliński P., van den Pol-van Dasselaar A., Golińska B., Paszkowski A., Nilsdotter-Linde N., O'Donovan M., Melis R.A.M., Czerwińska A., Delaite B., Bauer A., Florian C., Baste F., Fradin J., Gauder P., de Kort H. and Krause A. (2018) Analysis of innovation brokering systems related to grasslands across Europe. *Grassland Science in Europe* 23, 983-985.

Guyomard H., Bureau J.-C. *et al.* (2020) *Research for AGRI Committee – The Green Deal and the CAP: policy implications to adapt farming, practices and to preserve the EU's natural resources*. European Parliament, Policy Department for Structural and Cohesion Policies, Brussels, Belgium, 162 pp.

Huyghe C., De Vliegher A. and Goliński P. (2014) European grasslands overview: temperate region. *Grassland Science in Europe* 19, 29-40.

ISS (2014) *Innovation Support Service.* EIP-AGRI Service Point, 12 pp. Available at: https://ec.europa.eu/eip/agriculture/sites/default/files/eip-agri_brochure_innovation_support_services_2014_en_web.pdf

Keady T.W.J., Gautier J.M., Morgan-Davies C., Carta A., Gavojdian D., Ocak S., Corbière F., Ruiz R. and Beltrán de Heredia I. (2018) What are the main challenges to improve ewe productivity in Ireland and Europe? *Grassland Science in Europe* 23, 986-989.

Keatinge R. and Korevaar H. (2017) EuroDairy – a European thematic network for dairy farming. *Grassland Science in Europe* 22, 170-172.

Kelly T. (2020) Knowledge exchange approaches for better decision-making and innovation processes. *Grassland Science in Europe* 25, 697-703.

Knotter S., Kretz D. and Zeqo K. (2019) *Operational Groups Assessment 2018*. IDEA Consult nv, Brussels, Belgium.

Krause A., Becker T., Feindt, P.H., Huyghe C., O'Donovan M., Peeters A. and Van den Pol-van Dasselaar A. (2018) Towards sustainable European grassland farming with Inno4Grass: an infrastructure for innovation and knowledge sharing. *Grassland Science in Europe* 23, 925-936.

Newell-Price J.P., Bufe C., Frewer L., Hejduk S., Hunter E., Klopčič, Lively F., Lombardi G., Mulvenna C., Rankin J., Ravetto Enri S., Schils R.L.M., Smith K., ten Berge H., Tindale S., Tonn B. and Williams J.R. (2022) An overview of European permanent grasslands: SUPER-*G* proposals to improve their sustainability and multifunctionality. In: *Grassland at the heart of circular and sustainable food systems* (eds. L. Delaby, R. Baumont, V. Brocard, S. Lemauviel-Lavenant, S. Plantureux, F. Vertes and J.L. Peyraud). *Grassland Science in Europe* 27, 781-790.

Operational Groups (2016) *Operational Groups: Turning your idea into innovation. Update 2016.* EIP-AGRI Service Point, 12 pp. Available at: https://ec.europa.eu/eip/agriculture/sites/default/files/eip-agri_brochure_operational_groups_update_2016_2016_en_web.pdf

Orozco R., Mosquera-Losada M.R., Rodriguez J., Adamseged M.E. and Grundmann P. (2021) Supportive business environments to develop grass bioeconomy in Europe. *Sustainability* 13, 12629. https://doi.org/10.3390/su132212629

Paraskevopoulou C., Theodoridis A., Johnson M., Ragkos A., Arguile L., Smith L., Vlachos D. and Arsenos G. (2020) Sustainability assessment of goat and sheep farms: a comparison between European countries. *Sustainability* 12(8), 3099.

Poux X., Bernard-Mongin C., Dumitras D., Ferraz de Oliveira M.I., Gouriveau F., Goussios D., Herzon I., Jitea M., Kazakova Y., Lerin F., Ljung M., Lomba A., Mihai V., Moran J., Pinto-Correia T. and Vlahos G. (2018) What is the relevant approach for engaging territorial actors in a HNV innovation project? A strategic perspective – findings from HNV-Link H2020 Thematic Network. SISA International Workshop (6-8/11/2018, Riga).

Villada A., Verdonckt P., Ferreiro-Domínguez N., Rodríguez-Rigueiro F.J., Arias-Martínez D., Rois-Díaz M., den Herder M., Paris P., Pisanelli A., Reubens B., Nelissen V., Paulo J.A., Palma J.H.N., Vityi A., Szigeti N., Borek R., Galczynska M., Balaguer F., Smith J., Westaway S., Rigueiro-Rodríguez A.and Mosquera-Losada M.R. (2018) AFINET: agroforestry innovation thematic network. *4th European Agroforestry Conference Agroforestry as Sustainable Land Use,* pp. 355-359.

Wielinga E. (2014) Concepts for co-creating innovations in the EIP. In: Aenis T., Knierim A., Riecher M.C., Ridder R., Schobert H. and Fischer H. (eds.) *11th European IFSA symposium, farming systems facing global challenges: capacities and strategies, proceedings.* Berlin, Germany, 1-4 April 2014, pp. 824-834.

An overview of European permanent grasslands: SUPER-*G* proposals to improve their sustainability and multifunctionality

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Abstract

Permanent grasslands (PG) occupy around 34% of the utilized agricultural area across the EU-27 and provide a suite of important ecosystem services (ES), including supporting biodiversity, regulating climate, mitigating risks of erosion risk and downstream flooding, and providing clean water, food and fibre. However, the degree to which these ES are delivered varies significantly between countries, regions and farms, reflecting differences in production systems and the specific livestock types, stocking rates and management practices employed. Given the large areas involved, the general lack of soil disturbance and the important role of ruminant livestock, changes to PG management and the farming systems in which PG are integrated have the potential to make significant contributions to meeting current biodiversity and climate change challenges. Understanding the potential of PG to deliver different ES requires quantification of the extent, state and functioning of PG in Europe; an assessment of existing and new data on the land use; testing and adoption of beneficial management practices through co-innovation with farmers; understanding of social, behavioural and economic barriers to uptake; and an appreciation of citizens' priorities and preferences. This paper provides an overview of how the Horizon 2020 SUPER-*G* project is contributing to these challenges.

Keywords: ecosystem services, land use, land management, co-innovation

Introduction

Permanent grassland (PG) is defined, for administrative purposes, as 'any land dominated by grasses or herbaceous forage that can be grazed/mown and has not been included in the crop rotation of a holding for five years or more' (Eurostat, 2019). According to Eurostat data for 2013, PG covers almost 50 million hectares across the EU-27 and accounts for 34% of the total Utilised Agricultural Area (UAA) (Eurostat 2021), although there are large differences between countries in terms of proportion of UAA, spatial fragmentation and distribution. This results in contrasting priorities in terms of the specific roles played by PG in different countries and regions. The four countries with the greatest area of PG are France (8.2 million ha), Spain (8.0 million ha), Germany (4.6 million ha) and Romania (4.4 million ha); together they make up 48% of the total permanent grassland area in the EU. Permanent grassland represents 95% of UAA in Montenegro; 81% in Northern Ireland; and around 60% in Slovenia and Switzerland.

Across Europe there has been a trend for a reduction in PG area (Peyraud *et al.*, 2014). Across the EU-6 (Belgium, Denmark, Germany, France, Italy and Luxembourg), between 1970 and 2010, permanent grassland losses were around 30%, i.e. a loss of c.7 million ha (Eurostat, 2017). More recently, from 2005 to 2013, across the different biogeographical regions (BGR) European countries have reported both increases and reductions in PG area. For example, within Hungary (Pannonian biogeographic region, BGR) the proportion of PG area has increased by 50% (or 234,000 ha) whereas Sweden (Boreal BGR) has seen a 13% (or 66,000 ha) reduction.

PG support social infrastructure and high levels of biodiversity that in turn can enhance ecosystem function and value to society (Cardinale *et al.*, 2012). PG are also the basis for many highly valued landscapes and offer recreational potential in many regions (e.g. for hiking, dog walking, skiing and hunting). The ability of farmers and land owners/managers to maintain and manage grasslands for ES delivery depends on local conditions (including soil type, slope, groundwater level and prevailing weather conditions), farm type (e.g. dairy, beef, horses, pigs, sheep and goat), the profitability of the farming business, regulations and any financial support or incentive provided by rural development programmes. The degree to which PG provide a balance of goods and services therefore varies significantly between countries, regions and farms depending on the production systems and the specific livestock types, stocking rates and management practices employed. Frequency of cutting, grazing intervals and the type and rate of fertilizers used can all affect ecosystem function. However, while national and European datasets and findings from a farm network survey carried out by the authors in the framework of the SUPER-*G* project (Mulvenna *et al.*, 2021) provide useful information, there is generally limited information about such PG management practices across Europe.

The preservation of PG and how it is managed is of particular importance to meeting two of the major challenges of the 21st century; namely, biodiversity loss and climate change (IPBES, 2019; IPCC, 2022), including protecting vital soil carbon stocks, reducing greenhouse gas (GHG) emissions and improving adaptation and resilience. Understanding how PG management affects ecosystem function and how ES delivery on PG compares to other land uses within a 10-20 year timeframe will be critical to meeting these challenges. However, this is only the first step in achieving sustainable PG systems that contribute towards achieving FAO Sustainable Development Goals (FAO, 2021). There is a need to understand social, economic and behavioural barriers to the adoption of sustainable PG management practices and how these may vary in the different countries and regions of Europe. Equally, agricultural and environmental policies and financial support to farmers and land managers should take account of citizen needs, priorities and preferences for both policy implementation and ES delivery.

The objectives of the SUPER-*G* project are therefore to achieve: (1) better understanding of the importance and functioning of European PG; (2) benchmarking of PG performance across Europe; (3) co-development of integrated approaches for profitable and sustainable PG management; and (4) co-development of tools and policy mechanisms, which are inclusive of stakeholder and citizen priorities, to support the maintenance and sustainable management of PG. In terms of the ES, based on the Common International Classification of Ecosystem Services CICES, V4.3, 2013 (Haines-Young and Potschin-Young, 2018) delivered by PG, the project focuses on food production (meat, milk, dairy products, honey), wool and biomass (wood, cork, bioenergy, fibre, bedding material); biodiversity (including maintenance of ecosystem functions); climate regulation (through carbon sequestration and regulation of GHG emissions); water quality (for drinking and non-drinking purposes; and to maintain favourable living conditions for terrestrial and aquatic biota); mediation of water flows (for supply and discharge; and flood protection/mitigation); erosion control (vegetation cover to protect/stabilize terrestrial ecosystems); and landscape and recreation. This paper provides an overview of project outputs to date and expected project impacts and outcomes. The next section outlines: (1) the development of a new PG typology and new classification of grassland-based farming systems, both of which can be represented spatially; and (2) the results of an integrated systematic review investigating how PG management affects ES delivery, and how PG compares to other land uses in terms of ES delivery. Following sections focus on PG management challenges and the co-development of management options and innovative technologies; outlines a review of socio-economic facilitators of, and barriers to, adoption of sustainable PG systems; and describes the development of tools for aiding decision-making by grassland farmers and policy makers.

Developing sustainable systems

In Europe, beyond the 'administrative' definition of PG used in CAP regulations, there is a wide diversity of PG types and associated production systems (Peeters *et al.*, 2014). EIP-AGRI (2016) identified the need for a standard PG typology, that is, one that is easily applied to all regions of Europe, and easy to grasp for all types of stakeholders (farmers, policy makers, the public and scientists). Such a typology would provide a harmonized basis for evaluating the potential of European grasslands to improve productivity (where appropriate) and ES delivery. Understanding the diversity of PG within associated farming systems will help to understand how PG management may be improved for the farming business and/or for wider societal benefits, and to spot potential trade-offs between private and public goals. A standard PG typology can also aid the inventory and mapping of PG, provide a framework for data collection, and improve communication about PG across Europe, including knowledge transfer.

Peeters *et al.* (2014) proposed a classification of grassland types in which PG are classified based on their management intensity into 'semi-natural' PG, 'improved' PG and PG 'no longer used for production', with semi-natural PG further subdivided into pastures and traditional hay meadows. Building on this, Tonn *et al.* (2020) proposed a two-level PG typology that consists of eight first-level and 18 subordinate second-level classes (Figure 1), based on the presence of woody plants, renewal interval, management intensity and productivity potential (using the Agroclimatic Indicators provided by the Copernicus Climate Change Service for the period of 2011-2021). It is applicable both at field and regional scales, is cross-referenced with existing classification schemes such as the EUNIS and Natura 2000 habitats classes (EUNIS, 2019), and includes a number of cross-cutting attributes that affect management and ecosystem delivery: presence of acidic, organic, stony or wet (poorly drained or high groundwater level) soils, presence of steep slopes, irrigation practice and climatic limitations by short vegetation period or by summer drought.

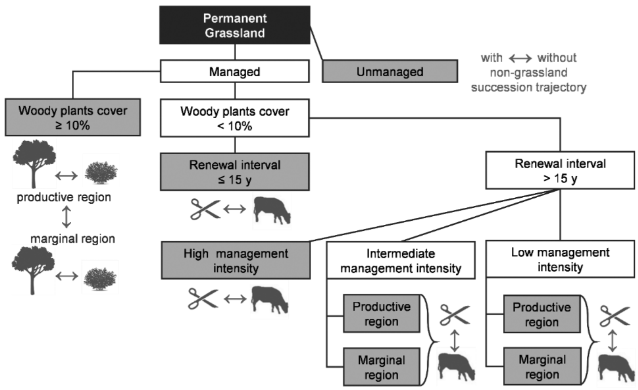

Figure 1. Overview of the proposed permanent grassland (PG) typology. First-level classes of the typology in grey boxes, subordinate second-level classes indicated by text and symbols (tree- vs shrub-dominated; predominantly cut vs predominantly grazed) outside boxes.

Management intensity is characterized by utilization intensity (based on cutting frequency and/or stocking rate of grazing animals) and fertilization intensity (total amount of manufactured fertilizer and organic manure nitrogen inputs), with interval between grassland renewal events (destruction of sward followed by resowing) used as an additional classification criterion. Thresholds for these management parameters were first set using expert knowledge within the SUPER-*G* consortium and validated and modified based on a stakeholder survey of 127 respondents from 27 European countries. The final typology will be made available as an atlas including an online classification tool for practitioners that links to PG type portraits and management options.

To improve understanding of how PG is typically incorporated within farms across Europe, Lombardi and Ravetto Enri (2021) developed a five-level, grassland-based farming system classification that considers the relationships between each farming system and the PG within it (Table 1). The classification was applied to the 2017 FADN database, which included 41,926 farms with PG, located in 1,063 NUTS3 regions. Each farm was assigned to one class in the first three levels of the classification and the FADN spatial information used to produce a series of distribution maps, which revealed significant variation across Europe in the importance of PG within farming systems (Figure 2), and the dominance of different livestock species and stocking rates. The FADN data mapping indicated that farms dominated by beef cattle are generally associated with moderate stocking rates (0.5-1 livestock units (LU) ha^{-1}) and a significant proportion of PG on farm (50-70%). Dairy farms are associated with higher stocking rates (>2 LU ha^{-1}) and a lower proportion of PG (<10%). The mapping of farm types indicated a significant variation in the importance of PG within grazing livestock systems and in stocking rates, a useful indicator of intensity. However, there are numerous management practice variables that can influence PG productivity and ES delivery.

Schils *et al.* (2022) performed a systematic literature review on the multifunctionality of PG in Europe, examining the effects of land use and management on 19 grassland ecosystem service indicators. They found that land use change from PG to cropland and temporary grassland decreased multifunctionality, while a lower PG management intensity was associated with benefits for biodiversity, climate regulation and water quality, but impacted the provision of high-quality animal feed. For nitrogen (N) input, there were significantly unfavourable effects on biodiversity, water quality and climate regulation (for GHG emissions but not carbon sequestration). For cutting and grazing frequency, there were overall negative effects on biodiversity and N losses to water. Grass renewal had significant favourable effects on forage

Table 1. Overview of farm system typology based on five levels and the categories per level.

1st level	2nd level	3rd level	4th level	5th level
Livestock species[1]	Stocking rate[2] on total UAA	PG share on total UAA	Exploitation regime	PG forage value[3] at farm scale
1. Beef cattle	1. <0.5 LU ha^{-1}	1. <10%	1. Predominantly grazing[4]	1. Low
2. Milking cows	2. 0.5-1 LU ha^{-1}	2. 10-30%	2. Predominantly cutting[5]	2. Intermediate
3. Mixed bovines	3. 1-2 LU ha^{-1}	3. 30-50%	3. Grazing & cutting	3. High
4. Sheep & goats	4. >2 LU ha^{-1}	4. 50-70%	4. Non-feeding or not relevant	4. Very high
5. Mixed ruminants		5. >70%		
6. Mixed & others				
7. None				

[1] Corresponding to >75% of livestock units (LU) on the farm.

[2] The stocking rate is calculated for every farm as the ratio between ruminant LU (i.e. bovines, sheep, and goats) and total UAA.

[3] Lombardi and Ravetto Enri (2021) propose that the typical quantity and quality of the forage from each PG type on farm can be synthesized as a single value to characterise overall PG forage value at farm scale.

[4] >75% PG area grazed.

[5] >75% PG area cut.

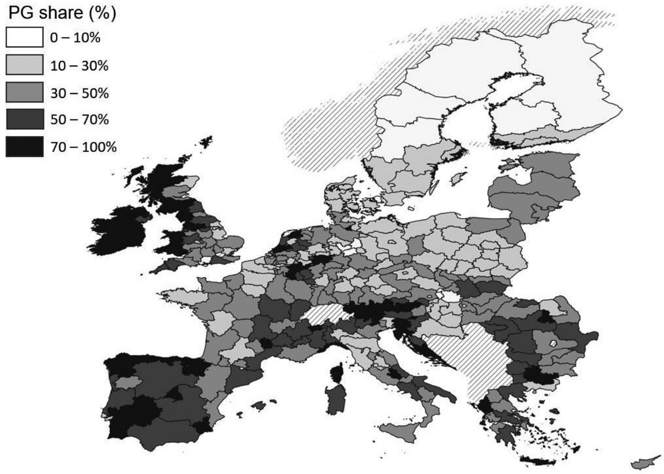

Figure 2. Map of the 3rd level of the classification (PG share on UAA). The average PG share per NUTS3×BGR is represented.

yield, but significantly increased nitrous oxide emissions and N losses to water. These three management interventions (N input, defoliation frequency and grazing renewal) were used as a proxy for intensity, as stocking rate was rarely included in the research papers that were reviewed. Schils *et al.* (2022) concluded that increasing support for lower intensity grassland management could help protect PG and secure the provision of multiple ecosystem services (ES), including vital carbon stocks and threatened species.

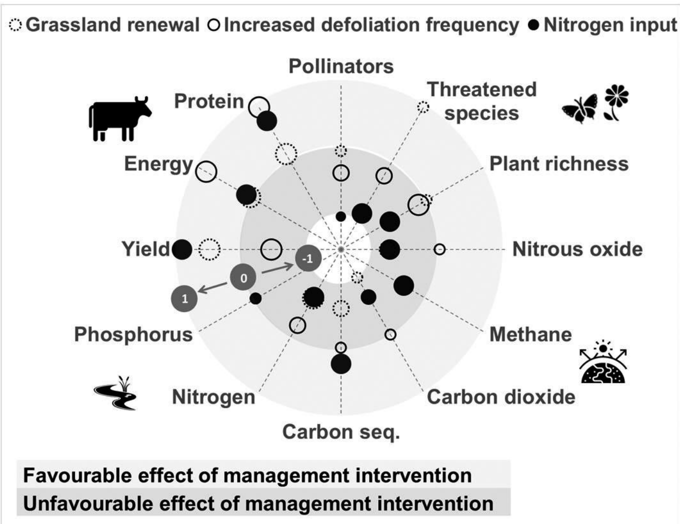

Figure 3. Effects, in terms of direction of change (favourable/unfavourable), of intensification management interventions on indicators for ecosystem services (ES). The boundary of lighter and darker shaded zones represents a mean score of 0 (no overall effect). The lighter shaded outer zone represents a favourable score (moving outwards, the mean score increases from 0 to 1). The darker shaded inner zone represents an unfavourable score (moving inwards, the mean score decreased from 0 to -1). Dot size indicates number of underlying cases (small: <5 cases, medium: 5-9 cases. Large: >9 cases).

Benchmarking and testing

We may have a good understanding of how PG management influences ES delivery within specific contexts. However, to improve our understanding of trade-offs and synergies associated with PG management it is important to work with farmers and other stakeholders to gather new data and experience on the practical implementation of management options and innovative technologies. Co-innovation workshops and a PG management survey covering 23 farm networks across six European BGR indicated that farmers face similar challenges in managing PG although priorities can vary significantly between BGR (Mulvenna *et al.*, 2021). Common challenges included limits to grass production potential due to stony soils, sloping land, sward composition or compaction; deciding when to reseed and which seed mix to select; adapting to more extreme weather patterns (drought tolerance and improved drainage); improving grazing management; weed control; economics (cost/benefit of management interventions); and improving the utilization and nutritional quality of grass. The length of tenancies; lack of incentive to retain PG; the importance of livestock breeds; and conflicts between productivity and ES such as biodiversity were also noted.

Mulvenna *et al.* (2021) found that the management practices considered most important for improving PG performance varied according to the BGR that the networks were in (n=352 farms, $X^2 = 29.39$, df = 10, $P = 0.001$). For example, the importance of 'drainage' declined in the order Atlantic > Boreal = Continental > Mediterranean > Alpine > Pannonian. There were also clear contrasts in the perceived importance of 'soil nutrient status' as a factor affecting PG performance, with 71% of Atlantic farmers thinking it was 'very important', compared with 28% of Alpine and 19% of Boreal. The other management practices surveyed were addressing soil compaction, selecting suitable sward species, measuring grass growth and measuring grass quality. There are also clear differences between BGR in the social, economic and practical constraints associated with the implementation of specific management options (Mack *et al.*, 2021).

PG management options, innovations and technologies were discussed during the co-innovation workshops and trials and experiments proposed. This has led to multiple on-going trials, demonstrations and experiments investigating the effect of overseeding with legumes and multi-species swards (e.g. Nölke *et al.*, 2021); precision grassland management (e.g. virtual fencing and remote and proximal sensing for yield and quality estimation - Fernández-Habas *et al.*, 2021); nutrient management (e.g. variable rate fertilizer application and improving the use of livestock manures); agri-environment innovation (e.g. grazing management and other management strategies for productivity, biodiversity and other public goods); and other practices such as conserving in-field grass/deferred grazing.

There are therefore numerous management options and innovative technologies available to farmers but there are cost and technical understanding constraints to their use as well as some confusion around how to manage PG for climate regulation and biodiversity, for example. Farmers need greater clarity on how to manage PG within sustainable production systems. This can be provided through guidance, technical support, advice and policies that incentivise retention and optimal management of PG, including for public benefits.

Securing performance

To support farmers in sustainable PG management and to achieve a healthy balance of land uses and land management practices it is important to understand the socio-economic facilitators of, and barriers to, adoption of sustainable PG systems; citizens priorities and preferences in the way they interact with land and its management; and the types of policies and policy options that can secure sustainable land use and management in the future.

In a review of stakeholder (including citizens and farmers/land managers) attitudes to grasslands and to ES delivery, Tindale *et al.* (2019) found that citizens were more likely to describe valuing cultural services including food, tourism, cultural heritage and landscape over supporting or regulating services. Farmers were more likely to give importance to provisioning and regulating services, such as food production, erosion control and water regulation. In relation to facilitators and barriers for decision-making, important considerations included the costs of implementing management options and their impact on farm income and profit. Policy characteristics were also an important focus, often related to agri-environment schemes and subsidies. An analysis of FADN data from 2008-2017 showed that farmer reliance on Rural Development Programme (RDP) and direct payments varies across countries and farm types (Elliott *et al.*, 2019). For example, in the UK it is estimated that payments represented 55% of farm incomes in 2014 (Institute for Government, 2019). An Italian study of High Nature Value (HNV) farms similarly found that subsidies comprised more than 40% of the Net Value Added (Trisorio and Borlizzi, 2011). Nevertheless, despite farm business reliance on subsidies, these payments are often seen to be problematic in relation to bureaucracy and uncertainty issues (Tindale *et al.*, 2019). Other important considerations for PG management included biophysical factors, relating to the influence of landscape, climate and biodiversity. Important enablers of decision-making were social (peer to peer) learning, communication and knowledge sharing, with the role of farm advisors and extension services being identified as important in this process. Social norms were also an important influence, particularly around family values, historical and cultural influence, and opinion and action of other farmers.

Tindale *et al.* (unpublished data) report on research focused on understanding of citizens' socio-cultural valuation of grassland landscapes, ES provision and management across Europe. Three focus groups, involving residents of rural areas, urban areas, and young adults from rural areas (aged 18-26) (n=104), were conducted in each of Spain, Sweden, UK, Switzerland and the Czech Republic between 2020 and 2021. In general, participants perceived grassland landscapes positively, associating them with positive emotions, advantageous environmental characteristics, outdoor leisure activity, and cultural identity. Prioritisation of ES from grassland varied between countries, influenced by grassland system diversity and complex socio-cultural and socio-economic differences. However, citizens across different countries shared farming ideals relating to farming for biodiversity.

In a review of policies promoting sustainable PG management, Hunter *et al.* (2020) found that previous polices relevant for grassland management have not fully considered the demand for ES and that if more holistic landscape policies are to be implemented across Europe, there needs to be a more coherent and transparent process that allows citizens' interests to be reflected in policy making. Securing the performance of PG is therefore a complex challenge that requires improved understanding of multiple physical, social, economic and political processes.

Aiding decision-making

Farmers can benefit from the use of decision-support tools (DSTs) to improve the efficiency of farm management. Policy makers also use DSTs to understand how changes in land use and land management can impact on social, economic and environmental outcomes. Sagoo *et al.* (2020) provided an overview of 127 paper-based, spreadsheet and software DSTs from 16 countries used by farmers, advisers, policy makers and others to support PG management in Europe. Most DSTs (115) were targeted at grass production and all of the countries that identified DSTs included DSTs in the grass production ES. In contrast, far fewer of the DSTs address other ES and, in many cases, countries had no DSTs addressing one or more of the other ES. For example, the review included 21 DSTs from the Netherlands, and of these, 21 targeted 'grass production', two targeted 'water quality' and one targeted 'carbon storage and GHG'. There were no DSTs addressing 'biodiversity and pollination', 'flood and erosion control' or 'landscape and recreation'. This highlights an opportunity to expand the remit of existing DSTs to consider other ES.

This observation is supported by the views of farmers and advisers in co-innovation workshops and surveys (Mulvenna *et al.*, 2021) that the benefits of PG management need more promotion. A tool that could demonstrate how grazing livestock farms deliver public good across a range of ES would be well received and could be a useful communication tool.

Finally, a series of workshops and surveys was carried out in Portugal, Italy, Switzerland, Hungary, the UK and Brussels to discuss the main PG policy issues at national and European level; the existing datasets that could be used within a policy DST; and the key research and policy questions that policy makers need to investigate (Lively and Rankin, 2020). To achieve maximum policy impact it was concluded that a tool would need to encompass whole farming systems and consider the synergies and trade-offs (socio-economic impacts) of various interventions, e.g. consider the impact of various changes in land use and PG management practice on farm income and the delivery of ES. One possibility is to combine spatial elements of the PG typology (Figure 1) and the farming system classification (Table 1; Figure 2) to enable a consistent, joined-up approach across regions. The next step in developing a SUPER-*G* policy tool will be to investigate the other datasets, such as FADN, that can be most practically applied to form the baseline farm and land use data and complement the land use and management models developed by the EC Joint Research Centre (JRC).

Conclusions

There is significant variation across Europe in the importance of PG within farming systems, the dominance of different livestock species, exploitation regime (in terms of whether grass is predominantly grazed or cut), management intensity (in terms of fertilizer use, frequency of cutting/grazing and stocking rates) and the range of ecosystem services (ES) provided. There is an urgent need to assess the sustainability of grassland-based farming systems, and to recognise and value the ES they deliver. Such systems must be socially, environmentally and economically viable in the long term and must clearly address urgent biodiversity and climate change mitigation and adaptation challenges. To help achieve this goal, the right policies must be put in place to support farmer livelihoods and farming systems that provide net positive environmental services for society. Increasing support for lower intensity grassland management could help protect PG, secure the provision of multiple ES, sustain rural communities and reflect citizens' interests. For example, longer-term farmer support payments could be linked to agri-environment indicators of multifunctionality. SUPER-*G* will develop a set of policy options for PG that take account of farmer concerns and citizen's needs, identifying the key changes required around land use, PG protection and PG management, and mapping out the actions needed for necessary changes in policy and practice to be realized (i.e. following the logic behind the theory of change).

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References

Cardinale B., Duffy J., Gonzalez A., Hooper D., Perrings C., Venail P. ... and Naeem S. (2012) Biodiversity loss and its impact on humanity. *Nature* 486, 59-67. https://doi.org/10.1038/nature11148

EIP-AGRI (2016) Profitability of permanent grassland. EIP-AGRI Focus Group final report. 12 April 2016. 44pp.

Eurostat (2017) Permanent grassland: number of farms and areas by agricultural size of farm (UAA) and size of permanent grassland area. Online data code EF_POGRASS. Available at: https://ec.europa.eu/eurostat/databrowser/view/EF_POGRASS/default/table?lang=en

Eurostat (2021) The share of the utilised agricultural area (UAA) occupied by the main agricultural land uses (arable land, permanent grassland and land under permanent crops); based on Farm Structure Survey data. Online data code TAI05. Available at: https://ec.europa.eu/eurostat/databrowser/view/AEI_EF_LU/default/table?lang=en

Elliott J., Nicholson F., Newell Price P., Tindale S., Hunter E., Sánchez ZamoraP. ... and Frewer L. (2019) Review of economic drivers of PG management options to deliver specific ecosystem services. EC project no. 774124, SUPER-*G* deliverable report 4.1b, 46 pp.

EUNIS (2021) EUNIS Habitat Classification. Available at: https://www.eea.europa.eu/data-and-maps/data/eunis-habitat-classification-1

FAO (2021) Tracking progress on food and agriculture-related SDG indicators 2021: A report on the indicators under FAO custodianship. FAO, Rome, Italy. https://doi.org/10.4060/cb6872en

Fernández-Habas J., García Moreno A.M., Hidalgo-Fernández T., Leal-Murillo J.R., Abellanas Oar B., Gómez-Giráldez P.J., ... and Fernández-Rebollo P. (2021) Investigating the potential of Sentinel-2 configuration to predict the quality of Mediterranean permanent grasslands in open woodlands. *Science of the Total Environment* 791, 148101.

Haines-Young R. and Potschin-Young M. (2018) Revision of the common international classification for ecosystem services (CICES V5.1): a policy brief. *One Ecosystem* 3: e27108. https://doi.org/10.3897/oneeco.3.e27108

Hunter E., Johansson C., Nybom J., Gallardo Cobos R., Sánchez Zamora P., Caño Vergara B., ... and Frewer L. (2020) The effectiveness of policies promoting sustainable permanent grasslands across five European countries (representing five biogeographic regions): Mapping, understanding, and key stakeholder perceptions. EC project no. 774124, SUPER-*G* deliverable report 4.1c, 360 pp. Available at: https://pub.epsilon.slu.se/24342/

Institute for Government (2019) Common Agricultural Policy. Available at: https://www.instituteforgovernment.org.uk/explainers/common-agricultural-policy

IPBES (2019) Summary for policymakers of the global assessment report on biodiversity and ecosystem services of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services. Plenary of the IPBES Seventh session, Paris, 29 April – 4 May, 2019. 44 pp.

IPCC (2022) *Climate Change 2022. Impacts, Adaptation and Vulnerability. Technical Summary*. IPCC WGII Sixth Assessment Report. 96 pp.

Lively F. and Rankin J. (2020) Set of requirements for a policy DST to achieve maximum policy impact. EC project no. 774124, SUPER-*G* deliverable report 5.3 (5.2b), 15 pp.

Lombardi G. and Ravetto Enri S. (2021) Report and maps with main farming systems on PG in Europe. EC project no. 774124, SUPER-*G* deliverable report 2.3, 31 pp.

Mack G., El Benni N., Tindale S., Hunter E., Newell Price J.P. and Frewer L. (2021) Effects of innovative management options on perennial grassland in the mountain area of Switzerland. *Grassland Science in Europe* 26, 159-162.

Mulvenna C., Smith K., Lively F., Rankin J. and Newell Price J.P. (2021) Overview of data, key gaps, and trends in PG management in the different biogeographic regions. EC project no. 774124, SUPER-*G* deliverable report 3.2, 141 pp.

Nölke I., Komainda M., Tonn B., Feuerstein U. and Isselstein J. (2021) Including chicory and selecting white clover varieties as strategies to improve temporal stability of forage yield and quality in white-clover-based temporary grassland. *European Journal of Agronomy* 130, 126362.

Peeters A., Beaufoy G., Canals R.M., De Vliegher A., Huyghe C., Isselstein J., ... and Wilkins R.J. (2014) Grassland term definitions and classifications adapted to the diversity of European grassland-based systems. *Grassland Science in Europe* 19, 743-750.

Peyraud J.L., van den Pol-van Dasselaar A., Collins R.P., Huguenin-Elie O., Dillon P. and Peeters A. (2014) Multi-species swards and multi scale strategies for multifunctional grassland-base ruminant production systems: An overview of the FP7-MultiSward project. *Grassland Science in Europe* 19, 695-715.

Schils R.L.M., Bufe C., Rhymer C.M., Francksen R.M., Klaus V.H., Abdalla M., ... and Newell Price J.P. (2022) Permanent grasslands in Europe: Land use change and intensification decrease their multifunctionality. *Agriculture, Ecosystems and Environment* 330, 107891.

Sagoo L., Dowers J., Lively F. and RankinJ. (2020) Review of existing tools available for use on permanent grassland. EC project no. 774124, SUPER-*G* deliverable report 5.1, 277 pp.

Tindale S., Frewer L., Hunter E., Sánchez Zamora P., Gallardo R., Caño Vergara B., ... and Miškolci S. (2019) Review of facilitators of, and barriers to, adoption and choice of PG management options to deliver specific ecosystem services. EC project no. 774124, SUPER-*G* deliverable report 4.1a, 71 pp.

Tonn B., Bausson C., Ten Berge H., Buchmann N., Bufe C., Eggers S., ... and Newell Price J.P. (2020) A management-based typology for European permanent grasslands. *Grassland Science in Europe* 25, 412-414.

Trisorio A. and Borlizzi A. (2011) Assessing the impact of rural policy on biodiversity: high nature value farming in Italy. In: 122nd EAAE Seminar 'Evidence-Based Agricultural and Rural Policy Making: Methodological and Empirical Challenges of Policy Evaluation'. Ancona: National Institute of Agricultural Economics, pp.1-9.

Utilizing unsupervised learning to improve sward content prediction and herbage mass estimation

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Abstract

Sward species composition estimation is a tedious one. Herbage must be collected in the field, manually separated into components, dried, and weighed to estimate species composition. Deep learning approaches using neural networks have been used in previous work to propose faster and more cost-efficient alternatives to this process by estimating the biomass information from a picture of an area of pasture alone. Deep learning approaches have, however, struggled to generalize to distant geographical locations and necessitated further data collection to retrain and perform optimally in different climates. In this work, we enhance the deep learning solution by reducing the need for ground-truthed (GT) images when training the neural network. We demonstrate how unsupervised contrastive learning can be used in the sward composition prediction problem and compare with the state-of-the-art on the publicly available GrassClover dataset collected in Denmark as well as a more recent dataset from Ireland where we tackle herbage mass and height estimation.

Keywords: biomass prediction, herbage mass prediction, unsupervised learning, clover

Introduction

Developing tools to help estimate biomass yield can improve decision making at the farm level. By fixing nitrogen (N) from the air into soils, white clover (*Trifolium repens*) has been shown (Nyfeler *et al.*, 2009) to be an important element of pasture sward management. White clover is an important motivator in the cow's diet, increasing dry matter (DM) intake and milk production (Egan *et al.*, 2018). A regular estimation of the expected herbage mass and biomass composition of the sward would provide a stepping-stone towards targeted N fertilization, reducing the cost for the farmer and the environmental impacts of fertilizer (Ju *et al.*, 2004). Estimation tools from canopy view images using neural networks have shown to be accurate but highly sensitive to different herbages with different visual characteristics (Narayanan *et al.*, 2020). Reducing the amount of GT data required to train these models is essential to quickly adapt to different climates and geographic locations. Skovsen *et al.* (2019) proposed using synthetically generated images to train a segmentation network paired with a linear regression model for composition detection and Albert *et al.* (2021) automatically labelled un-GT images to augment the training dataset. In this paper, we use un-GT images to train an unsupervised model using contrastive learning (Phuc *et al.*, 2020) which provides stronger initial weights to a neural network model performing composition detection. We show that this allows us to reduce biomass prediction error rates using un-GT images when compared to state-of-the-art algorithms. We compare with the state-of-the-art on the publicly available GrassClover dataset as well as an Irish dataset where we also look at biomass weight and herbage height estimation as well as predicting on images captured using phones.

Materials and methods

We aim to evaluate whether unsupervised learning can be used on un-GT images to bridge the gap between a partially GT dataset and a fully GT one. To do so, we use two image datasets to address biomass prediction and herbage weight estimation from canopy view images. The GrassClover image dataset gathered in Denmark in 2018 (Skovsen *et al.*, 2019) is composed of 152 fully GT biomass images (100 for training, 52 for validation) with the biomass composition comprising grass, weed, red clover and white clover. More images are available for test submission in an online challenge (https://competitions.codalab.org/competitions/21122), and 31,000 un-GT images are also provided. The second dataset was gathered in 2020 in Ireland (Hennessy *et al.*, 2021) using a Canon camera and is composed of 528 GT images with the biomass composition comprising grass, weed and clover. This dataset additionally provides ground-truth for the herbage mass (kg DM ha^{-1}) and herbage height post-cutting (cm). Additional views taken from a phone are also provided for evaluation purposes. We consider 52 images for training, 104 for validation and 372 for testing. A further 594 un-GT images are provided. For the unsupervised algorithm, we use the i-Mix algorithm (Lee *et al.*, 2021). This algorithm aims to compare two data augmented views of the same image against different images from the same mini batch, promoting the learning of visual features useful to differentiate between images. After completing the unsupervised learning phase, we use the parameters (weights) learned to initialize the neural network before starting the supervised training phase. In the supervised training phase, we train the network to predict the species composition in each dataset by minimizing a root mean square error (RMSE) objective over the training data. The network then outputs percentage values for weed, clover and grass and two values between 0 and 1 for the normalized herbage weight and height (Irish data only). All instructions to reproduce our results are available at https://git.io/JMrY1.

Results and discussion

We compare with state-of-the-art algorithms proposing low supervision alternatives to solve the biomass estimation problem. In Table 1, we report the RMSE of our approach and of Skovsen *et al.* (2019) (generating synthetic images), Narayanan *et al.* (2020) (using strong data augmentation) and Albert *et al.* (2021) (automatically labelling the unlabelled set) on the GrassClover dataset. For the Irish clover dataset (Hennessy *et al.*, 2021), we predict the herbage mass and multiply it by the predicted biomass percentage to evaluate the mass per species. We evaluate the Herbage RMSE (HRMSE) with regards to the ground-truth. We additionally report the Herbage Relative Error $HRE = (pred/gt)$, as in O'Donovan *et al.* (2002) where *gt* is the ground-truth value and *pred* is the predicted value by the network. Finally, HE is height prediction error (RMSE) in cm (Table 2).

Table 1. Biomass prediction results on the GrassClover dataset.

	Grass	Any clover	White clover	Red clover	Weeds	Avg.
Skovsen *et al.*	9.05	9.91	9.51	6.68	6.50	8.33
Narayanan *et al.*	8.64	8.73	8.16	10.11	6.95	8.52
Albert *et al.*	8.78	8.35	7.72	7.35	7.17	7.87
Ours	8.02	7.31	7.74	8.22	6.61	7.54

Table 2. Biomass prediction results on the Irish clover dataset.[1]

	HRMSE					HRE	RMSE				HE
	Total	Grass	Clover	Weeds	Avg.		Grass	Clover	Weeds	Avg.	
Albert *et al.*	230.10	220.84	34.86	27.13	94.28	1.14	4.81	4.75	3.42	4.33	2.15
Ours	229.12	218.02	37.65	29.21	94.96	1.09	4.58	4.22	3.44	4.08	2.03
Albert *et al.*	226.59	215.85	36.28	27.00	93.04	1.31	5.44	5.08	3.70	4.74	1.80
Ours	236.87	221.22	27.18	34.61	94.34	1.03	4.03	4.11	4.72	4.28	2.02

[1] The top two rows indicate test results on the Canon images while the bottom two rows report results on held out phone images.

Conclusions

By initializing the neural network on unsupervised images, we reduce the prediction error when training with a limited amount of GT images. This opens the possibility of quickly adapting an herbage mass and composition algorithm to an unseen environment using a limited quantity of labour-intensive GT images paired with a large quantity of un-GT data. Our results are comparable to Albert *et al.* (2021) without the need to generate synthetic images.

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References

Albert P., Saadeldin M., Narayanan B., Mac Namee B., Hennessy D., O'Connor A., O'Connor N. and McGuinness K. (2021) Semi-supervised dry herbage mass estimation using automatic data and synthetic images. In *Proceedings of the IEEE/CVF International Conference on Computer Vision,* pp. 1284-1293.

Egan M., Galvin N. and Hennessy D. (2018) Incorporating white clover (*Trifolium repens* L.) into perennial ryegrass (*Lolium perenne* L.) swards receiving varying levels of nitrogen fertiliser: effects on milk and herbage production. *Journal of Dairy Science* 101(4), 3412-3427.

Hennessy D. Saad M., MacNamee B., O'Connor N.E., McGuinness K., Albert P., Narayanan B. and O'Connor A. (2021) Using image analysis and machine learning to estimate sward clover content. *Grassland Science in Europe* 26, 132-134.

Ju X., Liu X., Zhang F. and Roelcke M. (2004) Nitrogen fertilization, soil nitrate accumulation, and policy recommendations in several agricultural regions of China. *Journal of the Human Environment* 33(6), 300-305.

Lee K., Zhu Y., Sohn K., Li C.L., Shin J. and Lee H. (2020) I-mix: A domain-agnostic strategy for contrastive representation learning. In *International Conference for Learning Representations.*

Narayanan B., Saadeldin M., Albert P., McGuinness K. and MacNamee B. (2020) Extracting pasture phenotype and biomass percentages using weakly supervised multi-target deep learning on a small dataset. In *Irish Machine Vision and Image Processing Conference,* pp. 21-28.

Nyfeler D., Huguenin-Elie O., Suter M., Frossard E., Connolly J. and Lüscher A. (2009) Strong mixture effects among four species in fertilized agricultural grassland led to persistent and consistent transgressive overyielding. *Journal of Applied Ecology* 46(3), 683-691.

O'Donovan M., Dillon P., Rath M. and Stakelum G. (2002) A comparison of four methods of herbage mass estimation. *Irish Journal of Agricultural and Food Research*, pp. 17-27.

Phuc L.-K.H., Healy G. and Smeaton A.F. (2020) Contrastive representation learning: A framework and review. *IEEE Access* 8, 193907-193934.

Skovsen S., Dyrmann M., Mortensen A.K., Laursen M.S., Gislum R., Eriksen J., ... and Jorgensen R.N. (2019) The grassclover image dataset for semantic and hierarchical species understanding in agriculture. In *Proceedings of the IEEE/CVF Conference on Computer Vision and Pattern Recognition Workshops*.

Near infrared reflectance spectroscopy analysis of multi species swards in Northern Ireland

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Abstract

Multi species swards (MSS) grown in a temperate climate, have the potential to produce high energy, high protein herbage and have a positive effect on nitrogen use within grassland management systems. This study investigated the potential of using near infrared reflectance spectroscopy (NIRS) to determine a more efficient method of MSS analysis compared to the proximate analytical techniques. The trial included plots of specific mixtures and monocultures of species, such as perennial ryegrass, timothy, plantain, chicory, red clover and white clover. MSS trial calibration ranges were: crude protein (CP) (144.89±36.77 g kg^{-1} dry matter (DM); acid detergent fibre (ADF) (227.6±41.7 g kg^{-1} DM); ash (92.6±15.0 g kg^{-1} DM); water soluble carbohydrate (WSC) (189.2±72.5 g kg^{-1} DM), neutral detergent fibre (NDF) (430.9±52.0 g kg^{-1} DM) and digestibility value (761.6±64.35 g kg^{-1}). NIRS correlation coefficients (RSQ) for ash (0.964), ADF (0.751), CP (0.981), D value (0.993), NDF (0.739) and WSC (0.985) were created using WinISI software. This NIRS technique will be an efficient way of evaluating MSS within agricultural systems.

Keywords: herbage, infrared, proximate, spectroscopy, temperate

Introduction

At present an efficient and precise analytical tool for determining the feed quality of multi species swards (MSS) is not widely available, and this has been identified as a key research gap (Patterson, personal communication, 2021). Successful near infrared reflectance spectroscopy (NIRS) analysis requires forage material to be homogeneous, as in fresh grass silage analysis (Park, 1998); to determine the nutrient values of monocultures of ryegrass (*Lolium perenne* L) (Burns, 2013); or to evaluate germplasm within a ryegrass breeding programme (Archer, 2014). MSS are too heterogeneous to analyse fresh, so a dried, milled method was adopted (Norman, 2019). The dried, milled MSS samples were NIRS scanned and the spectra graphs created for each sample. The spectra had the format Log (1/Reflectance) verse wavelength 400 to 2,498 nm, with resolution of 2 nm. This is directly related to the intensity of organic chemical groups which, in turn, are used to determine the chemical composition of feed nutrient fractions (Wang *et al.*, 2010). Chemometrics (Brereton, 2003) was applied to differentiate spectral data, and reference chemistry analysis was applied to 33% of the trial set. The sub-sampled spectra were then used to create the MSS NIRS calibrations. The reference chemical analysis of crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre (NDF), water soluble carbohydrates (WSC), ash and dry organic matter digestibility (DOMD) was determined using *in vitro* methods (AOAC, 1990). The NIRS calibrations developed from this research have potential to be used for determining the nutrient feed value of MSS as a complementary feed in ruminant diets.

Materials and methods

The trial was located at AFBI, Loughgall in County Armagh, Northern Ireland. Monocultures included two red clover, three white clover, two herb and five grass varieties plus two mixtures in the trial (Table 1). The layout was a lattice design with three replicates per sward type in a randomized format, with plots measuring 1.5×4 m. Harvested samples (n=177) were taken from eight simulated grazing harvests when average plot height reached 150 mm. Herbage was harvested using a Haldrup plot harvester, set at 75 mm mowing height above ground level. Samples were oven dried at 85 °C for 16 h, then milled using a Tecator cyclotec mill through a 1 mm sieve, prior to scanning using NIRS. Samples for analysis were taken on 27 April, 31 July and 1 Oct 2020 from an eight-cut sequence, to reflect seasonal variation. The dried, milled MSS herbage samples were scanned using a NIRS spectrometer, model 6500 FOSS-NIR System (Silver Spring, MD) for the spectral range 400 to 2,498 nm. The NIRS spectral data were recorded (log 1/R) at a resolution of 2 nm. Chemometric analysis (WinISI software package version 1.5) applied modified partial least squares regression analysis (MPLS). The feed quality parameters ash, CP, ADF, NDF, WSC and DOMD *in vitro* were developed and the best ranked were selected (by cross-validation) according to a lower standard error of cross validation (SECV) and a higher determination coefficient of cross validation (RSQ). Laboratory Proximate Analysis (AOAC, 1990) was as follows: Ash – furnace; 550 °C, nitrogen by DUMAS, ADF/NDF sequential fibre analysis, DOMD by Pepsin/Cellulase Digestion – *in vitro*, and WSC – anthrone method.

Results and discussion

Due to the heterogeneous nature of fresh MSS herbage and the need to develop an accurate analytical technique, the harvested material had to be dried, milled (1 mm sieve) and scanned using the NIRS technique. The NIRS calibration required reference chemistry analysis for ash, CP, ADF, NDF, WSC and DOMD *in vitro* (Table 1). Chemical analysis was determined for 33% of the trial with 59 samples chosen using the Global H value (WinISI™) also known as Mahalanobis value (Silveira, 2003) and selecting 5-6 trial samples from each trial type. Table 2 shows the range of chemical analysis for the trial types in the MSS, which was used to create the NIRS calibrations. Table 3 shows the NIRS calibration statistics for MSS. The slope values are very significant, showing linearity between reference chemistry and NIRS predictions. The RSQ values are significant for ash, CP, DOMD, and WSC, with ADF (0.751) and NDF (0.739) less significant.

Table 1. Mean chemical analysis values (g kg^{-1} DM) for MSS reference samples.[1]

Constituent	CY	RC	WC	C T	Mix A	Mix B	PN	RTTC	PRG	TF	Tim
ADF	266.3	278.4	222.4	231.4	203.1	214.9	271.5	248.7	218.6	227.2	227.8
Ash	123.0	102.4	96.7	104.5	87.9	91.3	121.4	102.2	86.8	93.6	77.9
CP	162.5	208.1	226	154.7	134.6	133.6	141.5	153.1	116.0	146.5	141.1
DOMD	706.9	716.9	741.8	735.2	795.7	785.7	700.0	724.1	803.7	733.2	769.3
NDF	362.4	429.3	388.3	469.5	434.1	432.2	335.5	409.1	447.3	472.7	506.4
WSC	141.2	101.0	101.2	163.8	239.8	236.0	130.0	167.4	262.1	176.0	189.0

[1] CY = chicory; RC = red clover; WC = white clover; CT = cocksfoot; Mix A = PRG/WC; Mix B = PRG/WC/CY/PN; PN = plantain; RTTC = PRG/TF/Tim/WC; PRG = perennial ryegrass; TF = tall fescue; Tim = timothy.

Table 2. Chemical analysis statistics for the MSS reference samples (g kg^{-1} DM).

Constituent	n	Min	Max	Mean	SD
ADF	58	173.62	333.62	221.55	38.75
Ash	59	67.69	143.61	94.51	17.48
CP	59	88.25	295.69	154.64	48.53
DOMD	59	632.55	886.80	761.62	64.40
NDF	58	311.43	631.15	432.87	58.88
WSC	59	63.49	310.62	188.68	70.90

Table 3. NIRS calibration statistics from using the MSS reference samples (g kg^{-1} DM).[1]

Constituent	Slope	BIAS	SEP	RSQ	SEP(C)	Mean	SD	SECV
ADF	1.020	2.497	19.318	0.751	19.332	227.62	41.72	27.08
Ash	1.058	0.270	4.129	0.946	4.153	92.59	14.97	5.25
CP	1.025	0.315	6.764	0.981	6.811	144.89	36.77	5.70
DOMD	1.000	0.001	0.485	0.993	0.489	761.62	64.35	9.11
NDF	1.069	4.233	30.252	0.739	30.231	430.88	52.00	23.35
SC	0.988	-1.113	8.593	0.985	8.589	189.23	72.52	8.78

[1] Slope = linear regression; RSQ = R squared; SEP = standard error of prediction; SEP(C) = standard error of prediction corrected; SECV = standard error of cross validation; BIAS = mean difference between predicted and observed.

Conclusions

Chemical analysis was carried out on 59 reference samples, which were strategically selected using multi variant analysis of NIRS spectral data from the MSS plot trial. NIRS calibrations generally require many hundreds of samples, so use of an algorithm to select reference material removed the outlier material and allowed calibration to be robust. This technique for NIRS calibration fast tracked the calibration process to create robust NIRS calibrations for MSS. NIRS analysis of MSS material will have potential application with nutritionists and farmers.

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References

Association of Official Analytical Chemists (1990) Official methods of analysis. 16th ed. Vol 1. Gaithersburg, MD, USA.

Archer J.E. (2014) The assessment of Lolium perenne L F2 clones for Drechslera siccans disease resistance and susceptibility using rapid analytical techniques. PhD Thesis, Queens University Belfast Library, Belfast, UK.

Brereton R. (2003) Principal component analysis: The method. In: Brereton R.G. (ed.) *Chemometrics. Data analysis for the laboratory and chemical plant*. John Wiley and Sons, Ltd, Chichester, UK, pp. 191-223.

Burns G.A., Gilliland T.J., Grogan D., Watson S., and O'Kiely P. (2013) Assessment of herbage yield and quality traits of perennial ryegrasses from a national variety evaluation scheme. *Journal of Agricultural Science* 151, 331-346.

Norman H.C., Hulm E., Humphries A.W., Hughes S.J. and Vercoe P.E. (2019) Broad, multi-species NIRS calibrations to predict the nutritional value of over 100 forage species within the Australian feedbase. *Applied Spectroscopy* 60(11), 1111-1122.

Park R.S., Agnew R.E., Gordon F.J. and Steen R.W.J. (1998) The use of near infrared reflectance spectroscopy (NIRS) on undried samples of grass silage to predict chemical composition and digestibility parameters. *Animal Feed Science and Technology* 72, 155-167.

Silveira L., Sathaiah S., Zangaro R.A., Pacheco M.T.T., Chavantes M.C. and Pasqualucci C.A. (2003) Near-infrared raman spectroscopy of human coronary arteries: Histopathological classification based on mahalanobis distance. *Journal of Clinical Laser Medicine & Surgery* 21(4), 203-208.

Wang L., Meng Q.X., Ren L.P. and Yang J.S. (2010) Near Infrared Reflectance Spectroscopy (NIRS) and its application in the determination for the quality of animal feed and products. *Spectroscopy and Spectral Analysis* 30(6), 1482-1487.

WinISITM (2003) Windows Infra Soft International software version 1.5, Agent, Foss, UK.

Using participatory research approaches to favour grazing practices of large herds of dairy cows

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Abstract

The 'LIT OUESTEREL' association is an open innovation device that relies on living lab principles. Its ambition is to reconcile livestock and society by addressing societal issues linked to animal welfare and the use of antimicrobials. In that framework, we develop in Normandy a specific project aimed at maintaining grazing practices for large herds of dairy cattle. This project relies on a six-year experiment called Tripl'XL implemented at the INRAE experimental farm of Le Pin-au-Haras. The objective of this experiment is to assess the behaviour, health, and welfare of dairy cows on pasture, by developing animal welfare indicators of grazing cows. One objective is to assess whether increasing pasture practices increases animal welfare. This experiment is also used to implement a participatory research approach involving local stakeholders, from farmers to consumers. The objective here is to co-construct innovations that can be tested within the experiment. It is also to assess the advantages and disadvantages of innovations (multi-criteria analysis) to define the conditions of adoption and diffusion. This communication briefly introduces the association and the experiment. It describes the methodology of the participatory research approach and provides a first appraisal of the latter after more than one year of implementation.

Keywords: living lab, participatory research, co-design, animal welfare, grazing cows

Introduction

To complete its ambition of reconciling livestock and society by addressing societal demands linked to livestock conditions, notably for animal welfare and the use of antimicrobials, the LIT OUESTEREL imagines and implements co-constructed projects of different types (research, development, innovation, education, etc.). It relies on 'living lab' principles and thus involves different types of stakeholders in the co-design of projects. Within that framework, a research and experiment project called Tripl'XL and implemented in Normandy (Le Pin-au-Haras) aims at maintaining and if possible, increasing the grazing of large herds of dairy cows. In addition to environmental benefits, grazing is likely to be a source of increasing animal welfare that will be assessed thanks to the development of welfare indicators at grazing. It will also be used to support a participatory research approach. We successively present the LIT OUESTEREL, the Trip'XL experiment, expected results and perspectives.

The LIT OUESTEREL: a living lab focused on animal welfare and health

The living labs concept appeared at the end of the 1990s in the domain of information and communication technologies, and is based on the principle of co-construction where economic and non-economic actors collaborate from the beginning of the co-construction process to imagine, develop, and implement methods, tools, technologies, products, services ... (hereafter solutions). Living labs are participatory devices that rely on different methodologies depending on the project type and phase; for example, surveys, focus groups, creative workshops, development of prototypes ... (Janin *et al.*, 2013). This new way of co-producing solutions is at odds with the linear or sequential model of innovation (Cooper, 1990). Although living labs differ in function of themes and territories, they share three common characteristics by establishing an ecosystem composed of both private and public actors interested, directly or indirectly, by the thematic, by using various open innovation working methodologies that involve all users, rely on iterative creation processes and test solutions in real environments, and by establishing a shared mind-set based on mutual trust.

The LIT OUESTEREL living lab aims at increasing livestock welfare, reducing the use of antibiotics in livestock while guaranteeing animal health, and improving income, work and living conditions of actors (farmers, carriers, slaughterers). These three objectives are not immediately compatible. They generate trade-offs that can be addressed by co-construction approaches. The living lab brings together today more than 50 stakeholders grouped into colleges (research/education, development, agri-food industries, retailers, numerical start-ups, veterinarians, animal welfare NGOs, etc.). It covers the three species of poultry, pigs and cattle, and is located in the west part of France (administrative regions of Bretagne, Normandy and Pays de la Loire) with three infra-regional pilot territories where most co-design projects are developed.

The Tripl'XL experiment

The Tripl'XL experiment has been developed at the INRAE experimental unit of Le Pin-au-Haras in Normandy. Its main objective is to analyse conditions that will allow the maintenance and, if possible, increase of grazing practices for large herds of dairy cows in a context where these practices are decreasing because of economic, structural, and cultural reasons (CNIEL, 2019). This is despite grazing practices presenting several advantages from different points of view, including low input and low feed cost systems, low environmental impacts, and higher levels of animal welfare (freedom of movement, expression of grazing and social behaviour). Both farmers and citizens appreciate grazing.

The experiment will last six years. It will provide a multi-criteria analysis of a large herd of 150 grazing dairy cows that will be assessed from different points of view (zootechnical, economic, environmental, animal health and welfare, etc.). It will examine how different parameters may influence this set of performances (breeds, genotypes, feeding strategies, land plot management, weather conditions, etc.). A specific objective is to develop animal welfare indicators that are currently lacking for grazing livestock. Indicators will rely on both automated data obtained from various sensors and direct measurements on animals. The project aims at measuring animal welfare at grazing and linking the latter to grazing practices and parameters. In addition, the experiment is the concrete support of a participatory approach involving all interested stakeholders (direct and indirect users). In so doing, we hope to better inform direct and indirect users on the advantages and disadvantages of grazing, and to co-design complementary or alternative solutions whose impact could be analysed and discussed thanks to the experiment, and to constitute a first circle of stakeholders, notably of farmers, to ensure the diffusion of good grazing practices. This will be achieved by on-site meetings with professional and non-professional actors, creation of participatory workshops, and the use of different diffusion supports (videos, social medias, etc.). The Tripl'XL project does not address the question of antibiotics; that will be the focus of complementary projects.

Expected results and perspectives

It is too early to present the first results of the experiment. Only data for the first year (2021) of the experiment are available. In addition, animal welfare data have been collected for a complete grazing season. They will be analysed in the coming months. This will likely lead to a revision of the animal welfare protocol to improve both its relevance and operational capability. This revision will consider the fact that the ultimate objective is to implement this protocol in commercial farms in a context where they would be able to argue of improved animal welfare performance to justify higher product prices and/or targeted direct aids.

During this first year, we were impacted by the Covid crisis. We have, however, been able to organize several conferences, seminars, open days, working meetings, expression contests, etc. For instance, the experimental unit of Le Pin-au-Haras opened its doors during four days in October 2021 within the framework of the Science Festival, with visits and workshops (farmers, scholars, citizens). Events have covered a wider thematic and geographical spectrum than the grazing of cows and the Tripl'XL experiment with, for instance, works on food relocation in the three pilot territories. One lesson that emerges is that co-constructing with all stakeholders on technical issues is difficult because of knowledge gaps between professional and non-professional actors. This led us to adapt our participatory methodology based on an 'Y' approach with some events reserved to professionals (left branch of the Y), others reserved to non-professionals (right branch), and finally common events (low branch). In parallel, we have identified and characterized innovations issued from research, development, and innovation actors (top-down innovations) and from farmers (bottom-up innovations). This 'innovation hunt' work will be continued in two complementary directions, first by analysing conditions of genericity, diffusion, and transferability to other contexts of solutions developed in each environment, second by feeding codesign meetings with examples of solutions that have proven themselves in a given environment.

Conclusions

Despite an unfavourable sanitary context and a project starting less than two years ago, a lesson that already emerges is that the living lab approach offers opportunities to promote dialogue between the livestock production sector and society, dialogue must include all interested parties because it is only by co-designing solutions that it will be possible to propose a shared future for livestock.

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References

Janin C., Pecqueur B. and Besson R. (2013) Rapport de recherche PACTE. Les living labs: Définition, enjeux, comparaisons et premiers retours d'expérience.

Copper R.G. (1990) Stage-gate systems: a new tool for managing new products. *Business Horizon* 33(3), 44-54.

CNIEL (2019) Observatoire des élevages laitiers. Le pâturage des vaches laitières françaises. Etat des lieux de la pratique pour l'ensemble des territoires français.

CarSolEl, a user-friendly tool to predict carbon stocks evolution in grassland-based farms

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Abstract

Maintaining or increasing soil organic carbon (SOC) stocks is crucial to meet the 2050 carbon neutrality target. The contribution of crop and grassland to SOC stocks was shown to be variable in France according to the soil, climate, and management situations. To assist farmers assessing how their land use and management modifies SOC stocks, a user-friendly interactive tool was developed. This tool is based on random decision forests built on fine gridded model simulations of SOC changes over France, carried out within the framework of the '4p1000' INRAE study. The tool estimates a 30-year SOC evolution (0-30cm layer) for three land use types: permanent grasslands, and crop rotations with and without temporary grassland. The tool inputs are 26 easily available information sources related to soil, climate, and length of the rotation (including the share of grassland and cover crops), fertilization and irrigation practices and grass management. The tool is a good approximation of complex process-based crop and grassland models. It is therefore a promising tool for estimating *in situ* SOC evolution of livestock farms.

Keywords: soil carbon sequestration, grasslands, cropping systems, climate mitigation option

Introduction

About 20,000 French livestock farmers have already assessed and examined their milk or meat carbon foot print with the CAP'2ER® Tool (https://cap2er.fr/Cap2er/), sometime as part of the implementation of the CarbonAgri method (https://www.france-carbon-agri.fr/methodologie-carbon-agri/), which grants 'Low Carbon Label' certification. Unfortunately, until now, CAP'2ER® only accounts for changes in land use and subsequent evolution of the soil organic carbon (SOC), while leaving out the effects of agricultural practices and local soil and climate context on SOC of the reviewed farms. The CarSolEl project aimed to develop a user-friendly tool, which fills this gap. Specifications and development of this tool are presented, with preliminary results.

Materials and methods

STICS (Brisson *et al.*, 2003) and PaSim (Ma *et al.*, 2015) are process-based soil crop models dedicated to the daily simulation of water, nitrogen, and carbon cycles in, respectively, arable land and permanent grasslands under temperate climate. Regarding carbon cycle, both models consider main soil carbon inputs: plant litter and crop residues, animal restitution, organic fertilization, roots. The simulated organic carbon can join humified carbon pools or be degraded by mineralization delivering CO_2 back to atmosphere. Nevertheless, using these complex models in commercial farms is unaffordable due to their high number of inputs and the technicity level they required. In Pellerin *et al.* (2019), more than 500,000 simulations carried out with these two models, as part of the INRAE '4p1000' study, have provided 30-year average values for the SOC of French soils in the 0-30 cm layer. We used these SOC simulations and their soil, climate, and management inputs to develop a user-friendly prediction tool, we called 'CarSolEl tool'. An analysis was conducted on the '4p1000' dataset to determine which variables most influenced the SOC. Three random forests were built, one per land use, using the most influential variables and 80% of the '4p1000' dataset (randomly drawn). The remaining 20% of the '4p1000' dataset were used to assess the prediction error committed by each forest, based on the dataset set aside. Developed in R, the CarSolEl tool was further supplied with a shiny user interface and tested with farmers and advisers on 81 fields of 14 French typical farm situations, to evaluate its acceptability and relevance. During the CarSolEl project, the ability of the STICS and PaSim models to simulate SOC dynamics in grassland soils was also evaluated and their calibration improved with data from large long-term experimental setups. This version of the process-based crop models could not yet benefit from the improvement of the tool presented here.

Results and discussion

The CarSolEl tool provides an estimate of the 30 years SOC change in the first 30 cm of soil from only 26 input parameters available in most farms (Table 1). This change corresponds to the average gain or loss obtained when maintaining agricultural practices for 30 years. The tool can be used for permanent grasslands (PG), crop rotations with temporary grassland (CTG) and crop rotations without temporary grassland (C). CarSolEl can be run at field or farm scale when aiming to analyses different cropping systems or grassland management types. The tool's prediction of SOC is close to that of the STICS and PaSim models, with a mean error of 30 kg C ha^{-1} yr^{-1} for crops, 36 for grass ley and 56 in permanent pasture. Correlation coefficient is very high (i.e. over 0.99 whatever the land use).

Table 1. Description of the CarSolEl tool inputs.

Climate	Type of climate (Joly *et al* 2010, https://journals.openedition.org/cybergeo/23155)
	Minimum daily temperature (30-year median) (°C)
	Maximum daily temperature (30-year median) (°C)
	Annual rainfall (30-year average) (mm)
Soil (0-30 cm layer)	Initial SOC (t C ha^{-1} yr^{-1})
	Pebbles in the soil mass (%)
	pH and $CaCO_3$
	Limestone (%)
	Sand (%)
	Soil texture
	Total depth (cm)
Crop rotation	Duration of the rotation (yr)
	Types of crops (% each)
	Average harvested yield on crops over rotation time TDM ha^{-1} yr^{-1}
	Share of grassland in the rotation
	Share of cover crops in the rotation (%)
Fertilization and irrigation management	Nitrogen supplied by mineral fertilizers (kg N ha^{-1} yr^{-1})
	Nitrogen provided by organic fertilizers (kg N ha^{-1} yr^{-1})
	Type of organic fertilizer
	Indicator of whether irrigation is taking place
	Indicator of whether the grass area is mowed or grazed
Grassland management	Number of mowing events (events yr^{-1})
	Average grass harvested yield TDM ha^{-1} yr^{-1}
	Number of grazing sequences (max. 10 days each)
	Number of grazing days (days LSU ha^{-1} yr^{-1})

At farm scale, the CarSolEl tool was found very useful to point out which field management is not sufficient to maintain carbon stocks and to discuss agricultural practices to be implemented with the farmer in order to preserve or increase the SOC of his cultivated areas. Table 2 shows an example for an intensive dairy farm (average stocking density of 1.7 LSU ha^{-1}, 48% silage maize in the fodder area) located in the French Brittany region (1,040 mm yr^{-1}) on soil with high initial SOC value.

The CarSolEl tool simulates the 30-year average SOC evolution in farms for present cropping systems and management, and for alternative scenarios, with acceptable uncertainties. The tests confirmed that its required inputs are easily accessible. However, in farms with grazing, the information of the number or grazing days is difficult to fulfil, linked to allotment and grazing management, and requires discussions with farmers. Moreover, some production situations cannot be handled by the tool, as they are not present in the '4p1000' dataset in terms of land use (e.g. hemp, crop-legume mixtures), soil type (e.g. hydromorphic/peaty soils, erosion) or agricultural practices. In grassland-based farms in particular, the diversity of grassland type and management (e.g. dynamic rotational grazing, winter grazing, and forage supplementation) is not fully considered. Finally, it seems essential to properly set up the initial SOC of each field, as its value strongly influences the estimate of SOC change and reflects the cultivation and fertilization history of the field.

Table 2. CarSolEl estimate of the expected average 30-year SOC change with the actual cropland and management in an intensive French dairy farm.

Grassland type	Fertilizer inputs		Grass management		Soil organic carbon	
	Mineral (kg N ha^{-1})	Manure (kg N ha^{-1})	Grazing days (LSU d ha^{-1} yr^{-1})	Number of harvests	Initial value (t C ha^{-1})	Evolution (kg C ha^{-1} yr^{-1})
C[1]	65	101	-	-	85	-234
CTG[2]	42	0	644	0	88	-65
	42	70	280	1	88	+120
PG	50	70	384	1	93	+ 151

[1] Maize silage/wheat rotation.

[2] Maize silage/6 years grass-clover mixture rotation, only grazed or grazed and cut with manure application.

Conclusion and perspectives

The '4P1000' dataset has been updated by rerunning the simulations with the improved version of the STICS and PaSim models and will soon result in an update of the CarSoEl tool. The latter is therefore planned to be available in 2022, as a 'standing alone' tool and an API version. It will allows improving the estimation of SOC evolution in the CAP'2ER® tool which is widely used for the environmental assessment of livestock farms. Improved estimated of the SOC evolution will significantly increase the relevance of advice on carbon sequestration as part of the climate action plan.

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References

Brisson N., Gary C., Justes E., Roche R., Mary B. *et al.* (2003) An overview of the crop model STICS. *European Journal of Agronomy* 18, 309-332.

Ma S., Lardy R., Graux A-I., Touhami H.B., Klumpp K. *et al.* (2015) Regional-scale analysis of carbon and water cycles on managed grassland systems. *Environmental Modelling and Software* 72, 10.1016.

Pellerin S. *et al.* (2019) Stocker du carbone dans les sols français, Quel potentiel au regard de l'objectif 4 pour 1000 et à quel coût ? Synthèse du rapport d'étude, INRA, France, 114 p.

AEOLE – a collaborative initiative which benefits both farmers and biodiversity

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Abstract

The AEOLE project was designed as part of the collaborative platform 'Cluster Herbe' to design supporting tools to support farmers in their transition to livestock systems that value grassland resources and their multifunctionality (trade-off between ecosystem services). To describe the grassland diversity at regional scale, a typology of 60 agroecological types of grasslands was constructed and integrated into a multifunctional diagnostic tool that allows farmers to optimize their grass-based system. A serious game was created to raise farmers' awareness of the potential of semi-natural grasslands in relation to climate change adaptation.

Keywords: grasslands, multifunctionality, grass-based livestock farming

Introduction

Massif central (MC) is an important livestock farming territory of 85,000 km^2 in the middle of France, a highland region mostly covered by permanent grasslands. Livestock – dairy and suckler cow, suckler sheep – is the main agricultural production system, with a part of the production under official signs identifying quality and origin (SIQO). With a wide range of ecological conditions and an important diversity of agricultural practices, MC hosts a huge diversity of grasslands, mostly semi-natural. They greatly rely on farmers' practices, and are a source of specific and intrinsic qualities of dairy and meat products (Farruggia *et al.,* 2008).

In order to promote these territorial resources, the collaborative network 'Cluster Herbe' involves partners from research, development, extension services, agriculture and food industries. It contributes to co-elaborate R&D projects and multi-scales innovations, from biotechnical tools to organizational approaches, in favour of grass-fed animal productions. Since 2016 more than 50 projects have led to improve knowledge on grassland ecology, climate change mitigation and adaptation, and to design innovative economic schemes (see https://www.sidam-massifcentral.fr/cluster-herbe/).

The AEOLE project describes the diversity of grasslands at regional scale and integrates it into a multifunctional diagnostic tool to help farmers to optimize their grass-based systems. A serious game was developed to raise farmers' awareness about the potential of semi-natural grasslands, for example to focus on the importance to get more resilient grassland in a context of climate change.

Materials and methods

AEOLE results from a 12-year collaboration between research and more than 15 stakeholders who work towards the recognition of grassland in MC as a response to agroecological transition and climate change adaptation, at the regional scale. Data used came from two main sources:

- A network of 143 plots throughout the Massif central area, monitored during 2 consecutive years at 3 key springtime periods (early, middle and end of first cycle of vegetation growth), ending up to 729 agronomical samples, 400 botanical surveys, 138 soil analyses and 143 practices surveys.

- A consortium of 80 experts, including researchers, technicians, extension officers, specialized in agronomy, animal science, ecology, botany, soil science and nutrition. After two plenary sessions and two to five meetings, depending on the focus group, the consortium produced academic and empiric knowledge aggregated into indicators of agroecological potentials (proxy of ecosystem services) or quality indices (from nutritional or organoleptic point of view).

Results and discussion

All the products and results of the AEOLE program are available online for free on the following website: https://www.sidam-massifcentral.fr/developpement/aeole/.

A typology book to organize and to describe the diversity of grassland

This output is based on a cross-analysis between botanical, agronomical and ecological variables measured on the reference network. Sixty types of grasslands were identified (Galliot *et al.*, 2020) and organized in an identification key, depending on 4 simple parameters: altitude, practice (mowing or grazing), soil humidity and soil fertility. Each grassland type is described through a two-page booklet (Figure 1) which summarizes agricultural practices, botanical composition, agronomical values and services (yield, forage quality, flexibility), environmental values and services (species richness, carbon storage, pollinator supply), and product qualities (antioxidant, fatty acid). The typology is to be used by extension services to farmers, and by scientists for research work.

A diagnostic tool to support grass-based production

The multifunctional diagnostic tool (DIAM) measures and evaluates the coherence between the diversity of grasslands, agricultural practices and production, environmental services and intrinsic qualities of products. This diagnostic provides indicators that helps agricultural advisors to work with farmers on their different parcel types, bringing grasslands in the centre of the farming system (Figure 2). DIAM comes with a map plug-in to visualize the parcels of land regarding different properties (indicators of services). DIAM can also be used at a territory scale, e.g. aromatic compounds for PDO cheese. Since 2018 about 150 DIAM have been realised in the Massif central.

A serious game to learn the potentials and benefits of grasslands

The serious game 'AEOLE-le-jeu' aims to raise stakeholders' awareness about the benefits of grassland diversity for sustainable agricultural production in the context of global change (Carrère *et al.*, 2019). This collaborative game illustrates the consequences of individual decisions on grassland services, both at a farm and a territory scale: every player run their farm to reach individual goals while keeping the balance between agricultural (economic), environmental and societal services in order to sustainably develop the territory according to other players (Figure 3).

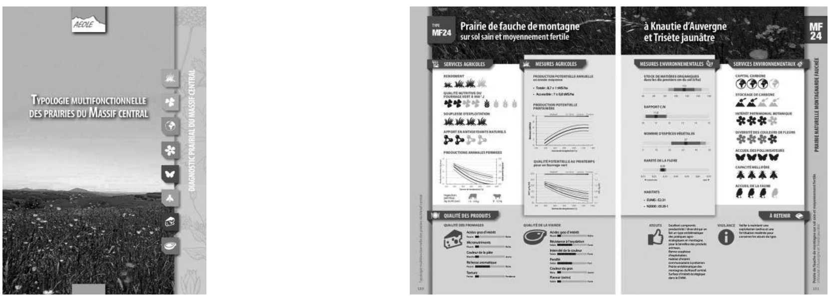

Figure 1. Main view of the typology cover and details of the agroecological potential and ecosystem services for a mesophile mowed upland grassland (source: Galliot *et al.*, 2020).

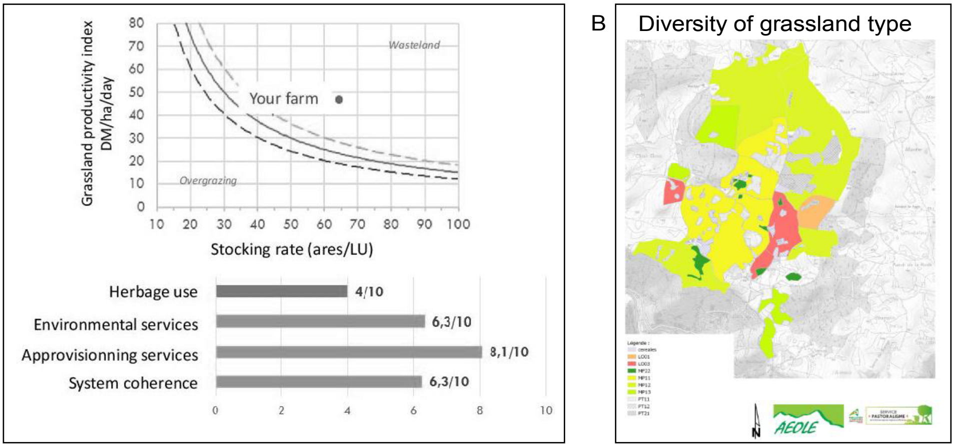

Figure 2. DIAM outputs give a global view of the system potentiality and coherence (A), associated to a map of the farm plots diversity (B) (Sources: CDA63).

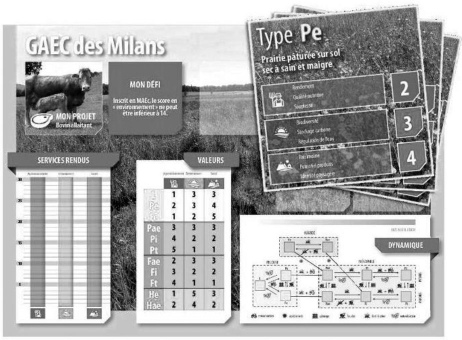

Figure 3. Main phase game of the serious game 'Aeole-le-jeu' and a farm description sheet used by a player.

Conclusions

After 12 years of co-development, AEOLE has fostered a shared vision between stakeholders on grassland diversity and multifunctionality. Knowledge produced by this R&D partnership has been summarized in a toolbox which contributes (1) to raise awareness among the real values of semi-natural grasslands, and (2) to optimize the use of grasslands on farms towards a balance between economic, environmental and social issues. Our results show that biodiversity preservation and agricultural production are compatible at the scale of a farm (Le Henaff, 2018). In the context of agroecological transition and climate change, agricultural advice must enhance a better understanding of ecosystem functioning, with the help of a diversity of tools and agroecological approaches, in order to support sustainable grass-based production methods and to optimize multifunctionality (trade-off between ecosystem services) in livestock systems. Training sessions and reuse of AEOLE tools in other projects will contribute to spread and integrate this knowledge into usual approaches. Animal productions in MC have several arguments to stand out from other classical productions. One of the challenges addressed to the Cluster Herbe network is to convert the multiperformance of farms, based on grassland diversity and resulting in high quality products, into added value for the benefits of the whole territory.

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References

Carrère P., Galliot J.N., Perera S. and Le Henaff P.M. (2019) Déclaration d'invention et de résultats valorisable concernant la conception du serious game «AEOLE-le-jeu» – enregistrement DI-RV-20-0040.

Farruggia A., Martin B, Baumont R., Prache S., Doreau M., Hoste H. and Durand D. (2008) Quels intérêts de la diversité floristique des prairies permanentes pour les ruminants et les produits animaux? *INRA Production Animales* 21(2), 181-200.

Galliot J.N., Hulin S., Le Henaff P.M., Farruggia A., Seytre L., Perera S., Dupic G., Faure P. and Carrère P. (2020) *Typologie multifonctionnelle des prairies du Massif central*. SIDAM-AEOLE, 284 p.

Le Henaff P.M. (2018) Les enjeux de conservation de la trame herbacée, *Revue d'Auvergne*, 628-629, 421-426.

Ten years of mobile milking at experimental farm Trévarez in France

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Abstract

Trévarez experimental farm is located in western France (oceanic climate and good grass growth). Like many farms, it has a fragmented land with little grazeable area around the cowshed. In 2012 the choice was made to design a mobile milking robot and to move it twice a year from the winter barn to a block of 23 ha of grass located 4.5 km away. From April to October, 50 cows in organic production stay on a 100% grazing diet. By end of 2021 eighteen transfers have been performed. After 10 years of use, we conclude the mobile robot is robust and transfers can easily be done by farmers. The cows remained on average for 5 months with 100% grazing on the summer site. Production is 18 kg of milk d^{-1} and the milking frequency is 1.6 milkings cow^{-1} d^{-1}. During this period the feeding cost is reduced by 78% compared to wintertime. Enlarging the milking platform remains the most profitable option if possible.

Keywords: dairy cow, robotic milking, grazing, production costs

Introduction

Robotic milking is now a common solution to reduce labour in Europe. In France, the number of farms with an AMS (automatic milking system) doubled between 2010 and 2016 and is now reaching some 15% of the dairy farms. However, after the purchase of an AMS, farmers usually reduce grazing or even suppress it (AUTOGRASSMILK, 2012). Though grazing leads to lower feeding costs (Dillon *et al.*, 2005), and is necessary to reach the higher levels of protein self-sufficiency that are the basis of the feeding system in organic dairy farms, farmers often miss technical advice to efficiently combine AMS and grazing. Therefore, an experiment was led at the Trevarez experimental farm to test practical solutions of grazing management and develop 'users' guidelines' for AMS farmers.

Materials and methods

Trevarez experimental farm is located in western France and has an oceanic climate and a potential grass growth of 10 t DM per ha per year. As with many dairy farms, it has fragmented land with little grazeable area around the cowshed. In 2012, an AMS was purchased to reduce working time, and in 2013 the farm was converted to organic. To maximize the use of grazed grass, the AMS was put on a trailer to move it from the winter barn to a block of 23 ha of pastures located 4.5 km away (Brocard *et al.,* 2012; 2016). During the winter, the trailer with the robot is located inside the shed and the trailer with the tank remains outside. Cows can graze around the shed during early spring and autumn. From April to October, the trailers, the drafting gate and the cows are all moved to the summer site; the 50 dairy cows stay on a 100% grazed grass diet with or without concentrates. By the end of 2021 eighteen transfers had been performed.

Results and discussion

Grass valorisation and animal performances

Since 2014, cows have been 100% grazing for 157 days per year on average, allowing a grazed grass intake of 2.9 t DM cow^{-1} yr^{-1} (Table 1). The milk production averaged 18 kg cow^{-1} d^{-1} and the milking frequency was 1.6 milkings cow^{-1} d^{-1} with little variability between the years (1.5 to 1.8, Table 1). These performances are consistent with previous experiments in the same grazing conditions (AUTOGRASSMILK, 2012).

Table 1. Main results of the 100% grazing period.

2014 to 2021	Mean	Min	Max
No. of days 100% grazing in season	157	141	181
Grazed grass intake (t DM yr^{-1} cow^{-1})	2.9	2.4	3.4
Number of milking cows	48	45	52
Milk per cow (kg d^{-1})	17.7	17.1	18.6
Milk per AMS (kg d^{-1})	764	671	914
Concentrate (kg cow^{-1} d^{-1})	0.5	0.09	0.9
Milking frequency (d^{-1})	1.6	1.5	1.8
Av. lactation stage (months)	5.9	5.3	6.8
Cows in first lactation (%)	39	30	45

The average number of cows and the milk production showed greater fluctuations due to the grass growth: empty cows remained longer in the herd in case of high grass growth, positively impacting the production per AMS.

Different management solutions tested

Grazing management

To increase cow traffic without fetching cows, different systems were tested: a two-way (AB, 2 paddocks per 24 h) versus a three-way (ABC, 3 paddocks per 24 h) grazing system were compared in 2014 and 2016 (Brocard *et al.*, 2017). Different access times to paddock and milking permission were also tested in order to find the best management system to combine animal performance, minimum labour and low feeding costs. Allocating 3 paddocks per 24 h improved the cow traffic but was more difficult to manage and more time consuming.

Facing grass deficit

On the summer site, maintaining a 100% grazed grass-based diet was not possible during dry summers. Different solutions were tested to face this challenge: (1) A return to the winter site. In 2015, the cows and the mobile AMS returned to the winter site from 1 July to 18 August to allow grass regrowth. During this period cows grazed around the shed and received haylage and maize silage in the cowshed. They easily adapted to their new location and diet. This option is well adapted to a long period of grass deficit since the transfer takes time and organization. (2) Crossing the road to graze a less-accessible paddock. This option was tested for 4 summers since 2017, during short periods of 1-2 weeks/year, representing 69 days in total. The cows grazed this paddock during the night and remained on the normal grazing platform during the day. This solution is appropriate when the period of grass deficit is short. The milking frequency decreased as the AMS was not accessible to cows during the night, but milk production often increased thanks to the good grass quality. (3) Buffer feed in a paddock. In 2020 and 2021, 10 kg DM cow^{-1} d^{-1} of haylage were delivered in racks in the day paddock close to the AMS. This solution meant that cows were only grazing the pasture during the night. This strategy did not impact on either the milk production, the milking frequency or the traffic around the AMS. This solution is also appropriate in cases of short-term grass deficit, with no other optional paddock available. However, the feeding cost increased because each kg of DM of haylage costs 6 times more than 1 kg DM of grazed grass.

Removing concentrate

From 2014 to 2020, the cows received a minimum of 0.5 kg of cereals per milking as a motivation to come to the AMS. However, concentrate is expensive and not necessary during the grazing period. To check the influence of concentrate allocation, its use was stopped in May 2020, one month after arriving on the summer site. Neither the traffic, the milking times, nor the milk production and milk sales were affected. No concentrate at all was delivered during the whole grazing season 2021, with the same conclusions, showing that the cows are mostly motivated by the fresh grass available after being milked at the AMS. Concentrates saved during grazing season can be used in winter, or suppressed to reduce the feeding costs.

Estimation of the feeding costs, investment and yearly running costs

To face the problem of having fragmented land area and to keep grazed grass as the only forage during 5-6 months per year, we designed a mobile robot and created a platform in an empty area, with no pre-existing infrastructure (Cloet *et al.*, 2017). The challenge was to compensate these investments by reducing feeding costs during the grazing period. On average, the feeding cost during this period is limited to 22 € per 1000 litres, i.e. reduced by 78% compared to wintertime; moreover, the daily working time is reduced by 2 compared to winter. In terms of investments (Table 2), the total costs for the robot itself, its equipment and the costs related to its mobility (trailers, infrastructure of summer site) reached 279,000 € (depreciation over 10 years). The total running costs including maintenance (contract, spare parts, repairs, reagents, hygiene and washing products), fluids (water, energy ...), insurance, and networks (phone, internet) averaged 8,100 € per year. The total yearly cost for investment and running reaches 36,000 € (43 € per 1,000 l). When adding the labour cost (3 hours per day × 2 SMIC per hour; based on French minimum wage rates of 10.15 € per hour in 2020), the total operating + investment cost reaches 51,560 € per year. This high running cost is hard to compensate even with a low feeding cost based on grazed grass. But for this experimental farm and its fragmented land area, it was the only way to combine an AMS with an organic feeding system.

Table 2. Investment and running costs € per year (depreciation over 10 years).

Investment	Robot	13,300
	Equipment	2,400
	Mobility	12,200
	Total	27,900
Running cost	Maintenance	6,200
	Fluids	1,300
	Insurance, networks	600
	Total	8,100
Total without labour		36,000
Labour		15,560

Conclusions

This long-term experiment shows that it is possible to design and implement a mobile AMS and to combine it with a 100% grazed grass system, with limited or no resort to concentrate. The grass management with two paddocks per 24 h system improves the cow traffic with limited human interventions. This way, the grass valorisation reached is far over the references for AMS farms. Though the total cost including investments and running costs remains high, this solution was probably the best one on an experimental site with a fragmented land area. Enlarging the milking platform remains the most profitable option if possible (exchange of land with neighbours, road underpasses ...).

References

AUTOGRASSMILK (2012) Innovative and sustainable systems combining automatic milking and precision grazing, Technical report, Animal and Grassland Research and Innovation Centre, Teagasc, Moorepark, Ireland.

Brocard V. and Le Coeur P. (2012) Mobile milking program in Trévarez: challenging land fragmentation. Innovations in Grazing, Proceedings of the 2nd meeting EGF Working Group Grazing. WUR report 644, Wageningen, the Netherlands.

Brocard V., Dufrasne I., Lessire F. and François J. (2016) Challenging land fragmentation thanks to a mobile milking robot: statement of two cases of implementation. *Proceedings of the 66th EAAP congress* (21), p. 330.

Brocard V., Déprés C. and Cloet E. (2017) Organic pasture-based automatic milking systems: comparison of two systems of pasture allocation. *Grassland Science in Europe* 22, 121.

Cloet E., Brocard V., Lessire F. and Guiocheau S. (2017) Maximizing grazing with a mobile milking robot. *Proceedings of the 8th European Conference on Precision Livestock Farming.*

Dillon P., Roche J., Shalloo L. and Horan B. (2005) Optimising financial return from grazing in temperate pastures. In: Murphy J.J. (ed.) *Utilisation of grazed grass in temperate animal systems,* Proceedings XXth International Grassland Congress, SatelliteWorkshop, Cork, Ireland, July 2005, pp 131-147.

How do farmers define the health of their grassland?

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Abstract

Faced with environmental crises, concepts such as One Health are being developed in the scientific world to consider the interdependence between human health, animal health and ecosystem health. Grassland health is integrated in these global approaches to health. For farmers, what does grassland health mean? The aim of this study was to characterize grassland health (definition and indicators), according to farmers. Sixteen dairy cow, sheep, and goat agroecological farms were surveyed in 2021 in the Massif Central (France). This work highlighted two different ways of defining the health of a grassland: (1) a state as such –characterized by a specific aspect reflecting its health (8 farmers); (2) an area capable of regenerating (7 farmers). Finally, one farmer was unable to define this notion. Farmers use different indicators to characterize grassland health: floral diversity, yield, animal, and soil observation. These indicators – based on academic knowledge, deviated, or constructed by farmers can be indicators of means (vegetation stage for cutting) or results (animal health, yield). The links between indicators used by farmers and the definition given are not very clear, as the indicators are very diverse. Some indicators are different from those in the literature but need to be integrated to put grassland health into action.

Keywords: grassland health, agroecology, indicators, interviews, farmers' knowledge

Introduction

While health issues have traditionally focused on human health, the current ecological and health crises attest to the interdependencies between human health, the environment, and the health of other living beings (Vieweger and Döring, 2015). This awareness has contributed to the development of different interdisciplinary health approaches such as One Health or Eco Health (Lerner and Berg, 2017). Eco Health and One Health are both holistic approaches which sit at the complex interface between human, animal, and environmental health interactions. Agriculture and livestock farming are at the heart of these issues since they are impacted by climatic hazards, but agricultural practices also impact the environment, animal health and welfare and human health (zoonoses, contribution to antibiotic resistance). Plant, soil, animal, and farmer health are integrated within these concepts but grassland health – defined as a comprehensive, multi-scale measure of system vigour, organization, and resilience, delimited to the grassland (Costanza, 2012) – is less studied and documented. The aim of this study was to characterize grassland health and whether this concept is used by the farmers, and how it is defined and quantified.

Materials and methods

A study was conducted on 16 dairy sheep, cow, and goat farms in 2021 in the Massif Central (France). Farms were chosen among volunteers, according to the following criteria: farmers claiming to be agroecologists, diversity of structure (size, animals), and diversity of practices. We conducted one interview on each farm. These interviews were governed by the methodology proposed by Kauffmann (1996) the pivot of which is the absence of hierarchy between the interviewer and the farmer, which enables the creation of proximity and encourages the farmer to open up. The interviews started with questioning on the definition of grassland health, according to the farmers, and then on the signs and indicators used by the farmers to characterize this health. The interview took place in different grasslands to illustrate what the farmers said. The interviewer follows up by validating what the farmer has said, asking questions about understanding (why, how, what), without trying to confirm or invalidate what the farmer has said. The analysis of the transcripts of the interviews first allowed us to type the farmers according to their definition of grassland health. The indicators mentioned by the farmers to assess the health of the grasslands or to make decisions were then analysed. For this purpose, the indicators were divided into two categories: means or results (Payraudeau and Van der Werf, 2005). The means indicators correspond to the practices put in place, with a health objective. Result indicators report on the effects of actions on health. A typology by expertise, based on the grouping of results and achievement indicators, was carried out. The result indicators are the first step in determining elements for the groups' composition. The mean indicators and the definitions given by the farmers were added, in a second step.

Results and discussion

The diversity of farms sought during the sampling is well present and is representative of the diversity in the study area in terms of structure size (LU from 15 to 80, UAA from 20 to 190 ha) and production. Cattle systems occupy an important part (11 farms) but goat farms (2 farms), sheep farms (1 farm) or farms with several flocks (2 farms) are also represented. Due to their location and altitude (850m, median), these farms are essentially grazing livestock systems as shown by the median grazing period of 8 months (from 6 month to 12 months).

First, this work highlighted two different ways of defining the health of a grassland. For four farmers grassland health is a state – characterized by a specific aspect reflecting its health – described by visual indicators. So, the health of the grassland is '*its state: the way it grows, the colour, the species present. A healthy grassland is varied and diverse*'. Six farmers defined grassland health as the grassland capacity to renew and adapt itself. A healthy grassland is thus '*a grassland that lives well, functions well, does not degrade. It adapts and does not need me to grow and renew*'. The last six farmers firstly understood what grassland health is but were not able to give a definition. When asked again, four of them used visual indicators to describe grassland health, one of them associated grassland health to grassland resilience and the last one was still unable to give a definition. The scientific definition of grassland health (Costanza, 2012) is in line with the definition given by 7 farmers out of 16, the second group. Then, a focus on the results indicators was carried out. The results indicators used to assess grassland health belong to various categories according to the farmers: they can concern the grass (yield/density/colour ...), the diversity of plants present (bio-indicator plant, number of species ...), resilience, but they can also be taken at the level of the soil, the herd, or the wildlife. The indicators used are based on academic knowledge, such as vegetation species – which is always used – and diversity or soil moisture (Costanza *et al.*, 2012). But some farmers also used other indicators, such as animal behaviour or milk production to assess the health of their grassland. One farmer explains that he knows if a grassland is healthy thanks to his cows: '*when I put my cows in a grassland, I see how they react*'. From the indicators used by the farmers, we constructed four groups (Table 1). All groups used grass indicators (group 1) but one of them also used soil indicators (group 2), resilience indicators (group 3) and the last group used resilience and herd indicators (group 4). Adding the definition used within these groups shows that farmers can have the same definition of grassland health but use different results indicators to assess it (Table 1). The same result indicator can also be used by all the farmers, whatever the groups (Table 1).

Farmers can use the same results indicators to assess grassland health but put in place a large diversity of practices, as in the two groups where all the farmers used the soil to assess grassland health but while some of them use a harrow, others do not intervene (Table 1). Some practices are also implemented by some farmers regardless of the group (e.g. harrowing). There is therefore no link between the definition of grassland health, results and mean indicators.

Table 1. Grassland health indicators and definition according to the farmers.

	Results indicators	Means indicators	Grassland health definition
Group 1 (n=3)	Grass: vegetal species, density, height of grass, yield, grass colour	harrowing (n=3), over-seeding (n=2), organic fertilization (n=2)	state (n=3)
Group 2 (n=4)	Grass: vegetal species, density, yield Soil: compaction, moisture, fauna, structure	diversity of practices (no tillage vs harrowing, late cut, hedges), do not overgraze (n=3)	state (n=2), resilience (n=2)
Group 3 (n=4)	Grass: vegetal species and diversity, density, grass colour Resilience: re-seeding, growing without inputs	fertilization (organic, not every year) (n= 3), grazing (n=1), moon (n=2), hedge (n=1), late cut (n=1), harrowing (n=1)	state (n=3), resilience (n=1)
Group 4 (n=5)	Grass: vegetal species and diversity, grass density Resilience: re-seeding, growing without inputs Herd: behaviour, health, milk	harrowing (n=4), late cute (n=5), organic fertilization (n=5), moon (n= 4), hedge (n=1), diversity of grazing	resilience (n=4), don't know (n=1)

Conclusions

To improve overall ecosystem and farm health, grassland health is important. This work shows that it is a concept that speaks to farmers and that they are implementing practices to improve grassland health. The results indicators and practices used by farmers are very diverse and may differ from those in the literature. Indeed, farmers develop their own indicators that need to be integrated to implement grassland health.

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References

Costanza R. (2012) Ecosystem health and ecological engineering. *Ecological Engineering* 45, 24-29. https://doi.org/10.1016/j.ecoleng.2012.03.023

Kaufmann J. (1996) *L'entretien compréhensif.* Nathan, Paris, France.

Lerner H. and Berg C. (2017) A comparison of three holistic approaches to health: one health, ecohealth, and planetary health. *Frontiers in Veterinary Science* 4, 163. https://doi.org/10.3389/fvets.2017.00163

Payraudeau S. and Van der Werf H.M.G. (2005) Environmental impact assessment for a farming region: a review of methods. *Agriculture, Ecosystems & Environment* 107(1), 1-19. https://doi.org/10.1016/j.agee.2004.12.012

Vieweger A. and Döring T.F. (2015) Assessing health in agriculture – towards a common research framework for soils, plants, animals, humans, and ecosystems. *Journal of the Science of Food and Agriculture* 95(3), 438-446. https://doi.org/10.1002/jsfa.6708

Adapting a Northern Ireland grass growth model to produce 14-day regional forecasts across the UK

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Abstract

GrassCheckGB is an on-farm grass growth and quality monitoring network established in 2019 involving 50 commercial livestock farms spread across Great Britain. The project is an extension of the long-running GrassCheck grass-growth monitoring project in Northern Ireland (NI), in which 7- and 14-day grass-growth rate forecasts are generated through the grazing season using the GrazeGro model described by Barrett *et al.* (2005). Using farm-specific weather and grass growth data from the GrassCheckGB farm network, the existing NI grass growth model has been adapted to provide regional grass growth predictions across four regions of Great Britain (Scotland, North England, Wales and South England). Following the adaptations described in this paper, the accuracy of grass growth predictions from the four regional GB grass growth models across a whole-season (March-October) was found to have a Root Mean Square Error of just 10.09-19.88 kg dry matter ha^{-1} day^{-1}, demonstrating its potential utilization as a decision-support tool for GB farmers.

Keywords: grass, grazing, grass growth forecast

Introduction

Grassland is a major component of agriculture in the United Kingdom (UK), making up 71% of the farmed land area (DEFRA 2018). The UK ruminant livestock sector is underpinned by the ability of UK farms to produce substantial quantities of grass forage each year, as a sustainable and cost-effective feed source. In Northern Ireland GrassCheck (est. 1999) has provided short-term 7- and 14-day grass growth forecasts as part of weekly grass growth bulletins published throughout the grazing season since the mid-2000s. These grass growth forecasts are produced using the GrazeGro grass growth model (Barrett *et al.*, 2005), and are valued by NI farmers as a tool to inform short-term grassland management decisions. This is particularly important in times of atypical weather patterns. GrassCheckGB was established in 2019 to bring together a network of 50 farms spread across Great Britain (GB) to promote the value of grass and good grassland management to farmers in GB, and to monitor both grass growth and quality throughout the grazing season (Huson *et al.*, 2020). An aim of this initiative was also to adapt the GrazeGro grass forecasting model to be used in 4 regions across GB: Scotland, Wales, Northern England and Southern England. In order to facilitate this, weekly grass growth data provided by farms within the project were used as a comparison on which to assess the performance of the GrazeGro model. To improve the suitability of the model for the GB regions various aspects of the model were adjusted. This paper describes those adaptations and the resulting accuracy of model predicted weekly grass growth rates compared to the average of on-farm measurements recorded for each region.

Materials and methods

In total 50 farms were enlisted into the GrassCheckGB project, all operating rotational grazing systems on predominantly perennial ryegrass pastures. Farms participating in the GrassCheckGB project were equipped with a Vantage Pro2 automatic weather station (Davis Instruments, California, USA) with additional soil moisture and temperature sensors planted at a 10-15 cm depth, and a solar energy (PAR) sensor. Farmers were also supplied with a platemeter (Jenquip EC10 or EC20), and walked their grazing platform weekly to record grass covers. All grass measurement and grazing event data were entered weekly into an AgriNet account provided to each farm, and average weekly grass growth rates calculated on a regional basis, with average calculations weighted by production system to account for variations in the number of Dairy vs Beef and Sheep farms in each region. Weather data for each farm were reported at 30-minute intervals from each station and collated to produce daily weather records for each farm, which were then averaged across all farms in each of the four regions as a regional average. These data were used as the input data for the adapted GrazeGro model, which requires daily average temperature, PAR and rainfall data in order to generate predicted grass growth rates.

The core features of the GrazeGro model remain unchanged since its original publication (Barrett *et al.*, 2005). However, currently in NI the calculation of grass growth restriction in conditions of moisture stress has been updated to account not only for soil moisture deficit but also for the effect of soil saturation, with a maximum restriction of 0.19 applied to grass growth during estimated soil saturation based on experimental findings by Laidlaw (2009). Nitrogen fertilizer application remains as a dynamic feature of the model. The standard protocol for N application accounted for in the model totals 270 kg N ha^{-1} yr^{-1}, with applications every 3 weeks from March to mid-September. However, in times of atypical weather conditions it may be appropriate to adapt this figure in response to on-farm conditions and management. In 2021 the total N rate applied was reduced to 210 kg N ha^{-1} yr^{-1}, with spring applications drastically reduced when temperatures were initially too cold for significant N uptake, and then when soil conditions were too dry. Further, the constant value in the original model for the initiation of reproductive growth was set at day 90. This has been reduced to day 45 in order to permit greater early spring growth in the model when favourable weather conditions are recorded. To assess model performance, the season average root mean square error was calculated of the actual-predicted weekly grass growth rates.

Results and discussion

The calculated RMSE for each regional model using weather data from 2019-2021, when compared to average on-farm grass growth rates, is shown in Table 1.

Recorded grass growth curves and model estimations of grass growth rates for each region in 2021 are shown in Figure 1. Given the wide variation in farms contributing to GrassCheckGB data and the high number of variable that can influence grass growth rates, the performance of this model proves acceptable. For use as a decision support tool when using weather forecast data, figures from the model are suggested to be indicative of likely trends in grass growth rates, and not to provide exact figures. Nonetheless, these indications will be useful to farmers for making short-term grassland management decisions such as deciding whether to cut paddocks for silage or retain the standing sward as extra grazing depending on high or low grass growth rate predictions.

Table 1. Root mean square error values for the whole-season (March to October) accuracy of the weekly grass growth model predictions compared to regional average figures recorded from GrassCheckGB farms.

Year / Region	North England	South England	Wales	Scotland
2019	13.90	19.00	15.42	13.97
2020	17.94	17.01	19.88	18.93
2021	10.09	16.56	13.70	13.53

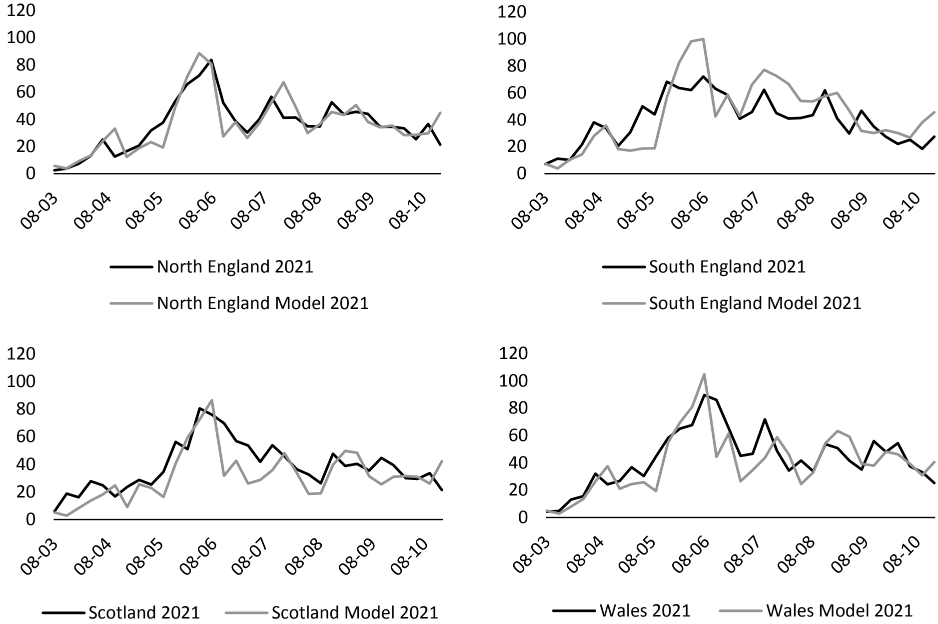

Figure 1. Comparisons between regional average weekly grass growth data recorded on GrassCheckGB farms in 2021, and predicted weekly grass growth figures from the adapted GrazeGro grass growth model using on-farm weather data. Growth rates in kg DM ha^{-1} d^{-1}.

Conclusions

Following minor adaptations the GrazeGro model has proved its suitability to be utilized for making short-term grass growth rate forecasts for four defined regions of GB, with typical accuracy of 10.09-19.88 kg DM ha^{-1} d^{-1} (as RMSE) across the past 3 years (2019-2021) compared to on-farm grass growth data. The performance of this model is similar to other predictive tools in use for grass growth in temperate regions, and the predictive trends in expected grass growth rates can provide a valuable decision support tool for GB farmers.

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References

Barrett P.D., Laidlaw A.S. and Mayne C.S. (2005) GrazeGro: A European herbage growth model to predict pasture production in perennial ryegrass swards for decision support. *European Journal Agronomy* 23, 37-56.

DEFRA (2018) Agriculture in the United Kingdom. EFRA, London, UK.

Huson K.M., Lively F.O., Aubry A., Takahashi T., Gordon A. and McConnell D.A. (2020) GrassCheck: monitoring grass growth and maximising grass utilisation on UK farms: meeting the future demands for grassland production. *Grassland Science in Europe* 25, 716.

Laidlaw A.S. (2009) The effect of soil moisture content on leaf extension rate and yield of perennial ryegrass. *Irish Journal of Agricultural and Food Research* 48, 1-20.

Combination of grassland surveys and knowledge transfer in the SatGrass project

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Abstract

In 'SatGrass', a comprehensive research project, we relate satellite and meteorological data to extensive field surveys on meadows with different utilization intensities across Austria to implement an area-wide yield and quality estimation. We established a survey network including research institutes, the Chamber of Agriculture, agricultural educational institutions, and agricultural associations to cover Austria's various grassland regions. Along with all stakeholders, we collect data on 170 farms bi-weekly. To meet our high quality standards, all participants, including farmers, teachers, consultants, and scientific staff, join a webinar once at the beginning and once during the growing season. The focus lies on the detailed survey procedure, containing the estimation of botanical parameters, the measurement of crop height, and biomass sampling. We use a smartphone app that guides the entire survey and ensures standardized data collection and transmission from the field to the server. After the growing season, we centrally collect the forage samples for chemical analysis. We use the data for calibration and validation of grassland models and prepare them for the participating farmers. In a further meeting, we discuss the data with the farmers, show possibilities of interpretation, and derive management decisions.

Keywords: grassland, survey, yield, forage quality, remote sensing

Introduction

Grassland is the predominant land use type in Austria's topographically and climatically disadvantaged regions (Buchgraber *et al.*, 2011). It covers 1.34 million hectares in Austria and is highly diverse in terms of productivity and sward composition due to different site conditions and various management intensities (BMLRT, 2021). For nationwide yield and forage quality modelling, it is essential to take all these spatial differences sufficiently into account. In SatGrass, we planned an exhaustive field campaign that covers all relevant grassland regions in Austria. The project's main objective is to develop applications that work from the field level to a regional level.

Establishment of a survey network

Yield and quality models based on statistical or machine-learning approaches require comprehensive data for training and validation. The data should cover the most common climatic regions and the very different management intensities. Therefore, we published a call for participation in the Austrian Association for Grassland and Livestock Farming newspaper with 4,500 subscribers. In addition, the Chamber of Agriculture has referred some potential farms to us. We selected the most suitable farms in terms of site and management conditions and reachability. Two employees of AREC Raumberg-Gumpenstein carry out the sampling on these 50 farms. In addition, the Austrian machinery ring (an Austrian farmers association) carries out field surveys on approximately 100 farms. This cooperation significantly expands the reach of the project.

Further cooperation partners are agricultural education institutions in Austria. At these 11 sites, teachers and pupils are carrying out field surveys. Figure 1 shows the distribution of the SatGrass-sites over Austria.

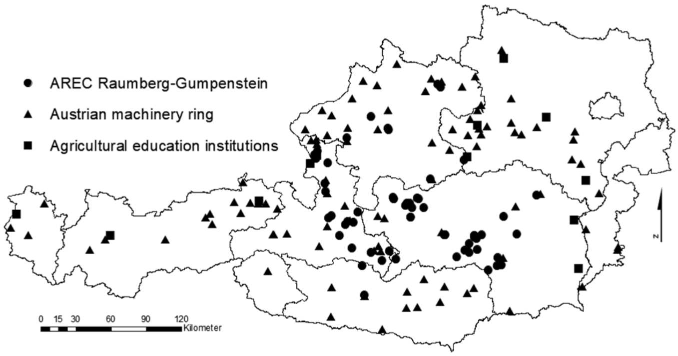

Figure 1. SatGrass-sites in Austria grouped by main partners.

Data collection

All participants had to announce the grassland fields where the surveys take place at the beginning of the project. We defined a homogenous sub-area within each grassland field where the surveys take place using a productivity index based on Sentinel-2 LAI time series (Essl *et al.*, 2021). We ensure that less, medium, and highly productive areas are present in a balanced way. We harvest three to five times at each growth depending on the period between the cuts to monitor precisely the grass growth dynamics. Therefore, the last survey of each growth should be as close as possible to the actual cut.

We developed a smartphone app that guides the entire grassland survey step by step. The app saves the entered data via mobile connection directly on a server. This standardized approach increases the comparability, improves the quality of the data, and minimizes paperwork. In the beginning, the executing person has to enter the farm-ID, the number of the actual growth, the date of the survey, and the respective repetition. A complete survey consists of three repetitions. The repetitions are located within 10 m of each other. In the next step, the harvest frame, of dimensions 1×1 m, has to be positioned on a representative area within the defined sub-area. We pay particular attention to crop height, vegetation cover, and species group distribution. Once the frame is positioned, the app prompts to take a picture from nadir. Simultaneously, the app logs the position using the internal GPS unit from the smartphone. Next, the app guides through the botanical surveys.

We estimate vegetation cover according to Peratoner and Pötsch (2019) and species groups proportion following Klapp (1930). Additionally, all partners have to note the most common species. After the botanical surveys, the partner from the machinery ring and the agricultural educational institutions measure the sward height within the harvest ring in threefold repetition. The employees from AREC Raumberg-Gumpenstein are equipped with a AccuPAR LP-80 Ceptometer (Decagon Devices Inc., Pullman, WA, USA) and measure sward height and also the leaf area index. We harvest the above-ground biomass with a grass clipper to a cutting height of 5 cm, to determine dry matter yield and to provided samples for analysis of chemical composition. Only plants whose basal stems are within the frame are cut. After determining the fresh matter weights of each repetition, we take a composite sample of 1000 g fresh matter. The partners dry the composite sample in a well-ventilated room and store it on their farms until the end of the vegetation period and then bring it to the laboratory for dry-matter determination and further chemical analysis.

We only use robust laboratory data (dry matter, crude protein content) for the model calibration. The botanical estimates ensure a better understanding of the data, but we do not include them in the models. Since model calibration is highly dependent on data quality, we calibrate the models with data of the highest quality level (collected by AREC). Data from the other partners provide the basis for model validation. Besides the vegetation surveys, our partners note the harvest dates. We use this data for the development of a satellite-based cut detection model.

Knowledge transfer

At the beginning of each vegetation period, all partners participate in a workshop. We draw the partners' attention there to the most critical steps in the survey process. Through frequent surveys, farmers, teachers, and pupils become better acquainted with their essential grassland resources. This promotes grassland management on the farms. After the analysis of the samples in our lab, we prepare the data for each partner and highlight the most important results. In a further workshop, we help our partner understand the data and give management suggestions tailored explicitly for their farm.

Conclusions

A nationwide application of satellite data requires comprehensive accessibility of yield and forage quality data. Planning and implementing a complex campaign that fulfills all requirements is challenging but indispensable for reliable and robust model results with high prediction accuracy. In addition, the active exchange with farmers, teachers, and pupils promotes knowledge transfer from science into practice.

Acknowledgements

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References

BMLRT (2021) *Grüner Bericht 2021 – Die Situation der österreichischen Land- und Forstwirtschaft*. Bundesministerium für Landwirtschaft, Regionen und Tourismus, Wien, Austria, 288.

Buchgraber K., Schaumberger A. and Pötsch E.M. (2011) Grassland Farming in Austria – status quo and future prospective. *Grassland Science in Europe* 16, 13-24.

Essl L., Atzberger C., Sandén T., Spiegel H., Blasch J. and Vuolo F. (2021): Multidisziplinäre Untersuchungen zur nachhaltigen Stickstoffdüngung unter Berücksichtigung der Möglichkeiten der satellitengestützten Präzisionslandwirtschaft. Die Bodenkultur: *Journal of Land Management, Food and Environment* 72(1), 45-56.

Klapp E. (1930) Zum Ausbau der Graslandbestandsaufnahme zu wissenschaftlichen Zwecken. *Pflanzenbau* 6, 197-210.

HappyGrass, a unique set of applications to manage grazing and meadows from sowing to harvest

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Abstract

HappyGrass is a unique set of smartphone applications to manage pasture and meadows from sowing to harvest entirely dedicated to the technical management of meadows and grazing. This new app, unique in France, brings together 14 decision support tools within 3 modules: (1) agronomic management of meadows (sowing, harvesting, fertilization, identification of flora, mowing periods, etc.); (2) grazing management with the inclusion of a grazing calendar and the management of grass growth for a weekly advice on the grazing circuit; and (3) a cartographic module that allows users to organize their plot to optimize grazing and animal circulation. HappyGrass is built for ruminant livestock breeders and their technicians, and includes current knowledge and references. It aims to make information more accessible to breeders when they are on site at their meadows. Equipped with GPS, Bluetooth, cartographic elements and meteorological data in a continuous flow, HappyGrass also offers interactivity by alerting the user to the approach of key grassland management phases (e.g. heat waves that can generate heat stress on animals). The proposed interventions show the extent of the functionalities, and how references and tools have been aggregated together in a coherent manner. At a time of adaptation to climate change and the search for fodder and protein autonomy where advice is increasingly difficult to obtain, HappyGrass offers comprehensive tools for optimal grassland management to ruminant livestock breeders and their technicians.

Keywords: ruminant livestock breeders, grassland management, smartphone application, decision tool

Introduction

HappyGrass is a set of smartphone applications entirely dedicated to the technical management of meadows and grazing. This assistant, unique in France, brings together 14 decision-support tools within 3 modules, (1) agronomic management of meadows (sowing, harvesting, fertilization, identification of flora, management of mowing periods, etc.), (2) the management of the pasture with the realization of a digital grazing calendar and the management of the growth of the grass for a weekly advice on the grazing circuit and finally (3) a cartographic module which allows users to draw and organize their plot to optimize its pasture and the circulation of animals. Aimed at ruminant livestock breeders and their technicians, HappyGrass mobilizes broad knowledge and numerous references and aims at making them more accessible to breeders when they wonder in front of their meadows. Equipped with technologies, GPS, Bluetooth, cartographic elements, and a supply of meteorological data in constant flow, HappyGrass also offers interactivity with its user by alerting him to the approach of key grassland management phases or periods, such as heat waves that can generate heat stress on animals.

A two-step construction

HappyGrass is the result of the merger of two tools initially designed and developed separately within two separate consortia. HappyGrass brings together solutions proposed in 2018 by the 'GrassMan' tool (Hardy, 2018; Ripoche, 2018) more oriented towards the agronomic management of meadows, and those of the 'PâturNet' tool which concerned grazing monitoring. The complementarity of the offers and the desire to offer the most complete service possible to its users led to the merger of the two teams in

2019. HappyGrass is therefore a set of decision support and registration tools unique to the market and exclusively centred on the meadows and their management from sowing to harvesting without forgetting the pasture and upstream, the mapping of the plot. HappyGrass is made up of seven founding members: the Institut de l'Elevage (Idele), the seed companies Cérience and Mas Seeds, as well as four breeding consultancy organizations (Eva Jura, CEL25-90, Cantal Conseil Elevage, Gén'IA test). The construction of its solutions also mobilized the expertise of INRAe and relied on collaboration with the engineering school SupAgroMontpellier.

The ambition of HappyGrass is the optimization of meadows. HappyGrass aims to put as much knowledge as possible within the reach of users to facilitate decision-making at the appropriate times, carry out diagnostics to clarify technical behaviour (choice of species, type, and dates of harvest, etc.); to simplify the approach to the meadow and its management; to make the meadow more attractive by providing it with new piloting technologies such as those that may exist in other productions, and to promote collaborative exchanges between breeders. The ambition of HappyGrass is also to produce more fodder on the meadows to improve the food and protein autonomy of ruminant farms and help to limit ration costs and production costs; improve the quality of fodder produced, whether grazed or harvested, to gain in feed value, palatability and ingestibility by animals; to make better use of the available grass and to limit losses in fields which have a non-negligible economic cost, and finally to strengthen the income of breeders.

Tools and services for the whole season

HappyGrass is available in three main modules: 'Prairie', 'Pâturage' and 'Parcelles'.

The 'Prairie' section offers eight very user-friendly tools. The idea is to provide the user with an answer to their question with just a few clicks when they are facing their meadow and must decide. The tools mobilize different repositories that allow offline use of certain tools. In addition, HappyGrass has the user's GPS position at the scale of a plot and has access to local weather data, modeled by triangulation. The eight tools of the 'Prairie' module are: Anticipate, Alert, Compose, Identify, Fight, Fertilize, Mowing and Qualify.

The 'Anticipate' tool allows the calculation of the grass area needs of a herd to ensure its grass feed on the full growth of spring.

With 'Alert' the objective is to anticipate decision-making, by mobilizing local weather forecasts. Alerts on the grassland stages according to the accumulated temperatures in base 0 from 1 January or 1 February are sent to the breeder to alert him on the actions to be taken. First nitrogen supply, turn out, harvests at optimum stages according to the types of harvests. This module also alerts the user to the level of heat stress experienced by the animals. It is calculated locally by measuring the temperature humidity index using the forecast weather data flow over 7 days. The alert is sent 24 hours before the expected stress exposure.

'Compose' offers the most suitable sowing compositions for the plot, its pedoclimatic context and the expectations of the breeder. It mobilizes more than 16,000 combinations of meadow cover adapted to the diversity of situations encountered. This tool also integrates two other services for 'Choosing your forage species' or 'Choosing your intercrop plants for services'.

The 'Identify' module is a support tool for the recognition and diagnosis of grassland flora. 'Fight' offers solutions to users for the management of invasive species in meadows. As for the 'Fertilize' module, it allows the users to calculate a nitrogen balance at the plot scale.

The last two tools proposed relate to harvests: 'Mowing' offers a visualization of the possible mowing windows according to the types of crops targeted, thanks to the meteorological data available in HappyGrass. Finally, the 'Qualify' tool makes it possible to approach the quality of the crops (hay, silage, and wraps) by questioning and observations in the absence of forage analysis.

With the 'Pâturage' Module, HappyGrass offers the first digital grazing calendar on the market (Ripoche, 2021a,b). Users record from a smartphone or the web platform (happygrass.fr) all the events during a grazing season: entry and exit of animals on the different plots and all interventions such as fertilization, mowing, etc. The ergonomics are designed in relation to the screen of a smartphone which allows direct entry of all information. All the elements recorded allow a complete analysis of fodder production, the level of productivity and valuation of the plots. This information should lead the user to optimize his grazing management and help him in his technical choices. If the breeder measures grass heights with a plate meter, he can then integrate these values into HappyGrass App and obtain a visualization of his grazing circuit with the calculation of the days in advance available. Grass height measurements made by the breeder are also instantly compared to local historical references and other users in their area.

The 'Parcelles' module is a mapping tool created in 2021 to design a plot best suited to grazing. This module allows you to draw the plots, with different types of fences, to place the entrances to the plots, but also the water points and to draw the access paths. The design that can be done on different base maps offers an ergonomic view and user-friendly use with the originality of proposing a costing of the investment for the development as the plot is built. The breeder can thus test different plot organizations and compare the investment costs.

For a wide range of users

The target users of HappyGrass are primarily ruminant breeders, but also technicians called upon to provide advice on meadows. The ease of use of some tools also makes HappyGrass suitable for teaching and learning in the field, smartphone in hand. HappyGrass is also intended for companies or organizations with specific specifications for their products including conditions of use of meadows.

Conclusions

The first and one of a kind, HappyGrass is a bundle of smartphone apps that offers users a very comprehensive set of features that can accompany them throughout a forage season. Each application is an autonomous decision support tool that can be mobilized separately. This tool is accessible in the whole of France through a network of authorized distributors. It is offered for sale under a 'discovery' formula with limited access to a few features, or in its complete 'expert' formula.

References:

Hardy D. (2018) Grassman: un bouquet d'applis pour gérer la prairie. *Pâtre* 656 août-septembre, 31-32.

Ripoche F. (2018) Numérique: gérer ses prairies avec GrassMan. *Biofil* 118 juillet/août / Grassman une application dédiée à la prairie. Republished in *Lait* 326, juillet-août.

Ripoche F. (2021a) HappyGrass: Tirer le meilleur parti de l'herbe. *Biofil* 36 juillet/août, 38-39.

Ripoche F. (2021b) HappyGrass: témoignages d'utilisateurs – Le numérique pour optimiser ses choix. *Biofil* 136 juillet/août, 40-42.

Pre-estimation of silage density via an application by using data available on farm

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Abstract

Aerobic instability of silages is still a widely reported problem. The main reasons are the interaction of easily available carbohydrates and yeast, as well as oxygen due to insufficient compaction and sealing. Silage additives can help to improve stability, but mistakes in compaction can seldom be overcome. Optimum compaction is needed to reduce air ingress and yeast activity in open bunkers. Estimating the silage density during the filling of a bunker silo is still complicated at farm level. An on-farm tool is needed to verify the technical process during bunker filling. Therefore, to estimate silage density during or before filling, a software application (APP) was developed based on comprehensive data sets and formulae derived from them. The main input data are crop dry matter (DM), rate of delivery, compaction weight, layer thickness and several bunker data. By changing the inputs, the bunker filling technology can be optimized before the filling starts. Playing with the APP may help the farmer to understand the interaction of the different impact variables. Research data are transferred into an easy applicable tool. The tool helps to improve the silage density and leads therefore to reduced silage DM-losses, CO_2 emissions and thus improved sustainability.

Keywords: aerobic stability, silage density, compaction, filling bunker, calculation

Introduction

High feed value of silages, as regards their nutrition and economics, is only achieved when the ensiled material keeps its quality until it is fed to the animals (Ross *et al.*, 2008). Aerobically unstable silages are often a problem on dairy farms (Kaiser and Piltz, 2002). The interaction of easily available carbohydrates, oxygen and yeasts are the main reason why silages heat up. In addition to the loss of energy and nutrients, the activity of moulds and their associated toxins are potentially harmful for livestock. The loss of dry matter (DM), nutritional value and hygienic quality always results in economic losses (Robinson *et al.*, 2005).

Low porosity within the silage can only be achieved by strong compaction of the substrate. A high compaction minimizes the ingress of oxygen during the feed-out. During the filling of the bunker, assessment of the compaction reached, and therefore comparison with recommended values, is difficult to achieve under farm conditions. The development of a software application APP to support estimations of silage density should help the farmer to plan or adjust the compaction parameters of bunker filling to reach the desired compaction density.

Materials and methods

An APP, the so-called compaction calculator using Microsoft Excel as the calculation tool, was developed for an assessment of compaction during bunker filling (available at: http://silagecalc.addcon.com). The underlying calculations are taken from the publication of Holmes and Muck (1999). They found the most important variables for predicting silage density were the crop DM content, the number of compacting machines, the weight of the machines, the delivery rate of forage, the layer thickness, the filling height and the time of compaction.

Holmes and Muck (1999) analysed numerous field data and the data modelling resulted in the subsequent formula:

$$Est\ DMD\ (lbs\ DM\ ft^{-3}) = (8.5 + PF \times 0.0155) \times (0.818 + 0.0136 \times D) \quad (1)$$

The expected dry matter density (*Est. DMD*) is calculated using the compaction factor (*PF*) multiplied by the average height of the bunker (*D*).

The average height of the bunker was determined from the arithmetic means of the height at the side and of that in the middle of the bunker. The compaction factor *PF* includes the proportional average weight of compaction machines *W*:

$$PF = (W/L) \times \sqrt{(N \times DM{:}C)} \quad (2)$$

The 'layer thickness' (*L*) is the thickness of the material layer after it is brought to the bunker and spread, but not compacted. The compacting machine equivalent *N* includes a time factor of the use of the compaction machines. In the APP the *N* is equated to one because continual use of the compacting machines is assumed. The dry matter (*DM*) complies with the compacted plant material. The delivery rate (*C*) is the tonnage of delivered plant material per hour. For the APP the American units were converted to SI units.

The estimation of the technological bunker filling data, like time needed or the frequency of passes, are based on our own calculations. The width of the bunker multiplied by the used 'filling length' represents the available area that can be filled. The number of necessary passes over the whole bunker is calculated using the tyre width and bunker area to be filled currently. The necessary speed of the compaction machine is calculated from the area to be filled and the track and tyre width. The time of compaction or the needed speed of the machines is in addition; this depends on the input at line 'technological/private breaks', which means breaks required for refuelling, waiting for transport units, staff rest breaks, etc. The parameters mainly available and used at farm level are the dry matter content, the delivery rate, the weight of compacting machines, the layer thickness and the geometry of the bunker.

The output of the calculation is the forthcoming compaction density under the given circumstances. The calculated density will be compared to recommended compaction densities from Honig (1987) and DLG (2012). The quality of the given compaction technique can be rated and modified if the calculated compaction is lower than the recommended value.

Results and discussion

The number and weight of the compaction machines are decisive for the success of the compaction (Muck and Holmes, 2000). This parameter has the second highest coefficient of correlation (0.262) of the factors affecting the silage dry matter density and a level of significance of $P<0.05$ (Table 1). But the layer thickness has the strongest effect on the silage dry matter density in the bunker, with a correlation coefficient of 0.279 (Table 1).

When the layer thickness is doubled the necessary speed for the passes has to be halved. A doubled length of passage time is the result, but nevertheless the potential for silage compaction decreases by more than 20%. A reduction of the compaction weight by half also results in a reduced compaction density by >20%. If the delivery rate is increased from 75 t h^{-1} to 100 t h^{-1} the effect is not strong and the estimated bulk density drops only by 5%. The DM of the delivered substrate has the third largest effect, described by a correlation coefficient of 0.209 (Table 1). But the crop DM can no longer be controlled at the harvest.

Table 1. Correlations between different compacting factors and the silage dry matter density (modified from Muck and Holmes, 2000).

Compacting factor	Correlation coefficient[1]
Layer thickness	-0.279*
Averaged weight of compaction machines	0.262*
Dry matter content	0.209*

[1] Significance * $P<0.05$.

Via the APP the farmer can enter the necessary data that have significant impacts on silage density. Moreover, farmers can recognize the interactions of impact factors. Before starting the bunker filling process, a check of the planned technology can be made. For example, when the calculated filling area, which is determined using the trailer volume, the bulk density, and the layer thickness, is bigger than the available filling area, the output 'necessary layers' is greater than one. Thus, the delivered material needs to be introduced into the bunker in multiple steps. The delivery rate of the crop material must be adapted to the geometry of the bunker and the compaction performance. Most important, the compaction performance must be the determining factor of the harvesting/bunker filling process, and not *vice versa*.

Conclusions

The compaction calculator is easy to use under practical conditions and gives an outlook on the reachable silage density based on farm-available data during the bunker filling process. In addition to the expected silage DM density, other practically relevant data like the number of layers depending on the delivery rate or the number of passes is calculated. The comparison to target values can be done immediately. The aim of the compaction calculator is to improve the too-low dry matter densities that are often found in farm bunkers. Playing with the APP may help the farmer to understand the interaction of the different impact variables and to adapt them till the optimum silage density is reached.

References

DLG (2012) Silagelagerung. In: *Praxishandbuch Futter- und Substratkonservierung*, DLG-Verlag, pp. 97-128

Holmes B.J. and Muck R.E. (1999) Factors affecting bunker silo densities. USDA-ARS, U.S. Dairy Forage Research Center, April 12, 1999.

Honig H. (1987) Influence of forage types and consolidation on gas exchange and losses in silo. In: *8th Silage Conference*, Hurley, UK.

Kaiser E.G. and Piltz J.W. (2002) Silage production from tropical forages in Australia. In: The XIIIth International Silage Conference, 11-13 Sept. 2002, Auchincruive, Scotland, UK, pp. 48-61

Muck R.E. and Holmes B.J. (2000) Factors affecting bunker silo densities. *Applied Engineering in Agriculture* 16 (6), 613-619.

Robinson P.H., Swanepol N., Heguy J. and Meyer D. (2015) 'Shrink' in corn silage piles. What are the real losses? In: *Proc. 2015 Western Dairy Management Conference*, Reno, NV. USA, 3-5 March 2015.

Ross F., Maack C. and Büscher W. (2008) Einfluss von Trockenmasse-Gehalt, Lagerungsdichte und Häcksellänge auf das Porenvolumen im Siliergut. *Landtechnik* 63 (6), 344-345.

Drone RGB imaging for phenotyping of red clover stand density in field experiments

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Abstract

Agriculture in Nordic countries relies heavily on grasslands. To maintain satisfactory productivity levels it is necessary to develop and select grass and clover cultivars which are resistant to winter kill, due to the harsh winter conditions at such latitudes. Assessments of stand density are typically performed using field operator grading, which is time-consuming and prone to subjective bias. Drones are being used increasingly for agricultural monitoring and could offer a time-efficient and objective solution for evaluating the stand density in cultivar selection. We used drone-acquired RGB images of red clover experiments in Sweden to evaluate the spring stand density of individual plots, which is a proxy for winter survival. In total, 255 plots were used to build up the regression models, where the response variable was the stand density as estimated by field operators. Cover and line percentages were used as explanatory variables. Resulting models show root mean squared errors between 11.2 and 9.1%, the best performances being obtained with line percentages. These results need to be confirmed before a practical system can be developed; nevertheless, results suggest that there is good potential for relatively cheap drones to be used for stand density assessment for red clover.

Keywords: winter survival, stand density, phenotyping, red clover, drone, RGB

Introduction

Grasslands are a corner stone for agriculture in the Nordic countries, where the meat and dairy sectors are major actors of the agricultural industry. Due to the harsh winter conditions at such latitudes (i.e. >55°N), the selection of winter-resistant cultivars of grass and clover plays a major role in the overall performance of the sector. Winter resistance of candidate cultivars is typically evaluated from field assessments in autumn and spring, relying on tedious individual assessments of numerous plots. Drones could provide a relevant solution to reduce both the time (and thus, cost) for plot assessments and the risk of bias between sites and operators. Several studies underlined the interest in drones for monitoring plot experiments. For example, Grüner *et al.* (2019) used a drone equipped with a simple RGB camera to estimate the yield of forage experimental plots in Germany. They showed that reasonable accuracy could be achieved (R^2 ranging between 0.59 and 0.81 for all treatments). Jin *et al.* (2017) estimated the stand density of winter wheat plots at emergence stages in France using a drone equipped with a high resolution RGB camera, obtaining a relative error of 14.3%.

The main aim of this study was to evaluate how drones mounted with RGB cameras could be used to evaluate the stand density of red clover cultivars in selection trials. Two methods were used, the first one based on a surface ratio, the second one based on a line ratio. Compared to the surface ratio approach, the line-wise ratio is closer to the actual way that field estimations are performed, but might require more data processing time, reducing its interest for an industrial use. Both approaches were compared in terms of accuracy.

Materials and methods

We used red clover spring stand density data acquired in 2019 from two sites in Sweden: Lännäs (63.16°N, 17.65°E) and Svalöv (55.91°N, 13.11°E). The topography of both sites was flat, and the combined field experiments resulted in 255 plots with a total of 49 different accessions, each plot containing 9 (Svalöv) or 10 (Lännäs) sowing lines. Individual plots were assessed for stand density on 2019-04-08 (Svalöv) and 2019-05-21 (Lännäs). Each site had its own field operator to perform the assessments. The plot stand density was evaluated by averaging the stand density values of individual lines.

Drone flights were performed on the same day as visual assessments using a Phantom 4 Pro with an RGB camera (DJI, Shenzhen, China). Predefined flight plans were used for each site with the software Pix4D capture (Pix4D, Prilly, Switzerland), resulting in 0.52 cm pixel resolution. Acquired images were processed using Pix4D mapper (Pix4D, Prilly, Switzerland) to obtain reflectance maps from which green leaf index (GLI; Louhaichi *et al.*, 2001) images were computed.

GLI images were further processed using QGIS 3.14. The polygons corresponding to the edges of plots were manually delineated (Figure 1A), GLI images clipped for each plot and masked using a site-adjusted threshold (Figure 1B) and further converted into binary images. The ratio of vegetative to the total number of pixels for each plot was defined as the cover percentage (*CP*).

Within plots, lines were manually created (Figure1C) to compute individual line percentages (*LP*) as the ratio between vegetation pixels to the total number of pixels for each line, and further averaged plot-wise. Linear models were adjusted between field observations and drone derived *CP* and *LP*. Resulting regression models were evaluated using R^2 and RMSE metrics.

Results and discussion

Obtained results are presented in Figure 2 and Table 1. Clear differences appear between sites, with higher R^2 but lower RMSEs for Lännäs compared to Svalöv. It appears that the *LP* approach shows slightly better performances compared to *CP* for both sites (Table 1). Interestingly, *LP* tend to always show superior values to the *CP* values (Figure 2C), which is probably related to the empty space between rows. This is obviously a flaw for the *CP* approach, as the canopy closure will vary greatly depending on the date of assessment.

Both *CP* and *LP* tend to underestimate the stand density (Figure 2A,B). This trend needs to be further assessed with independent datasets. Regardless, with RMSEs close to 10%, this preliminary study suggests that there is a good potential for drone-based RGB imagery to evaluate the stand density of red clover experimental plots.

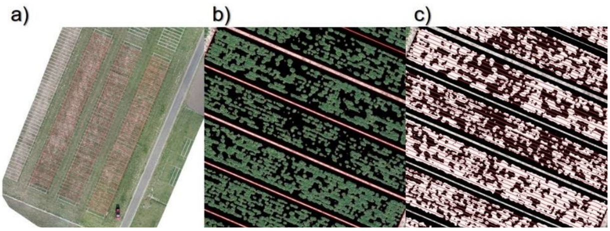

Figure 1. (A) Orthomosaic of the experiment in Svalöv. (B) A more detailed view over a group of plots with the masked image (background pixels are represented in black). (C) The same view, showing within plots lines and the binary image (vegetation pixels are shown in white).

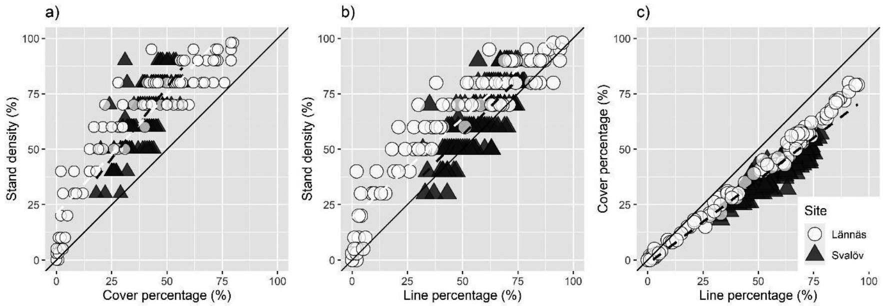

Figure 2. (A) Stand density vs cover percentage for both sites. (B) Stand density vs line percentage for both sites. (C) Cover percentage vs line percentage for both sites. The full black line shows the 1:1 line, dashed lines show individual regression lines.

Table 1. Metrics of the regression models.

Site	Method	R^2	RMSE
Svalöv	Cover percentage	0.57	9.5
Svalöv	Line percentage	0.61	9.1
Lännäs	Cover percentage	0.84	11.2
Lännäs	Line percentage	0.87	10.2

Conclusions

Drone-acquired RGB imagery has good potential to evaluate the stand density of red clover experimental plots. More work is required to confirm these results, but a line-wise approach might be preferable compared to a cover-wise approach as it is less dependent on the measurement date and, by extension, on the regrowth of red clover.

Acknowledgements

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References

Grüner E., Astor T. and Wachendorf M. (2019) Biomass prediction of heterogeneous temperate grasslands using an SfM approach based on UAV imaging. *Agronomy* 9(2), 54. https://doi.org/10.3390/agronomy9020054

Jin X., Liu S., Baret F., Hemerlé M. and Comar A. (2017) Estimates of plant density of wheat crops at emergence from very low altitude UAV imagery. *Remote Sensing of Environment* 198, 105-114. https://doi.org/10.1016/j.rse.2017.06.007

Louhaichi M., Borman M.M., Johnson D.E. (2001) Spatially located platform and aerial photography for documentation of grazing impacts on wheat. *Geocarto International* 16, 65-70. https://doi.org/10.1080/10106040108542184

Sustainable management model for the preservation of valuable open mountain areas: the Open2preserve project

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Abstract

In south-western Europe, the abandonment of traditional practices in mountains is causing a homogenization of the landscape and an increased risk of extreme wildfires, endangering ecosystems of high ecological value. The Interreg Sudoe Open2preserve project (2018-2021) aimed to develop a sustainable management model based on the principles of the pyric herbivory (combination of prescribed fires and targeted grazing) to preserve a mosaic of open landscapes in abandoned and high fire-prone areas. Thirteen partners from Spain, France and Portugal established eight pilot experiments in protected areas of contrasting environments in the SUDOE region (Atlantic mountains, Pyrenees, and Mediterranean areas). In these pilot experiments, we implemented a common management and monitoring design based on an initial removal of biomass and a multi-year targeted grazing with autochthonous horses and sheep. We monitored the intensity and severity of the burnings, the livestock welfare, the vegetation dynamics, and the soil function for two years, and tested drone and GPS technological tools. This multidisciplinary and international project gathered the main bases to implement pyric herbivory practices on mountain grasslands valuing prescribed burnings and targeted grazing, which promote the preservation of biodiversity and resilient ecosystems, while ensuring the environmental, social and economic sustainability of these practices.

Keywords: knowledge transfer, pyric herbivory, mountain grasslands, native livestock breeds, technological tools

Introduction

In south-western Europe, the abandonment of traditional management in mountain grasslands and the decrease of livestock grazing is causing the expansion of native woody plants, the homogenization and loss of diversity of the landscape and the accumulations of biofuels, increasing the risk of extreme wildfire events (Múgica *et al.*, 2021) and endangering ecosystems of high ecological value. Appropriate management of pyric herbivory, the spatial and temporal interaction of fire and grazing (Fuhlendorf *et al.*, 2009), should be restored in this region to preserve the ecosystem services associated with these pastoral systems. Despite its interest, the traditional knowledge of these practices has been lost in many areas (Fernández-Giménez and Fillat, 2012), and new research is needed to establish the guidelines for an adequate management adapted to the new global change scenarios. This is the framework in which the Interreg-Sudoe project Open2preserve ('Sustainable management model for the preservation of open mountain areas with high environmental value', https://open2preserve.eu/en/) arose, aiming to develop a sustainable management model based on the principles of the pyric herbivory, combining prescribed fires and targeted grazing to preserve a mosaic of open plant communities in abandoned mountain regions of SW Europe.

Materials and methods

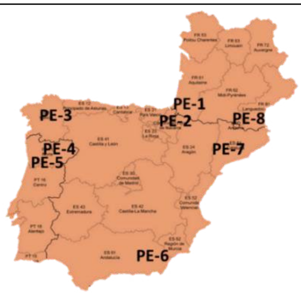
The project Open2preserve started in March 2018 and finished in December 2021. Project partners from Spain, France and Portugal established in their territories eight pilot experiments (PE) in representative, valuable mountain areas of temperate and Mediterranean environments. The eight study regions shared a similar socioeconomic situation derived from the abandonment of extensive ranging and traditional use of the mountain resources, but they differed in their climatic, geological and topographic conditions and developed a different type of dominant vegetation. We implemented in all PE a common management protocol, which was based on an initial removal of lignified biomass by prescribed burning (except in a PE where mechanical clearing was carried out due to climatic constraints), and a subsequent targeted grazing with animals of autochthonous breeds (equine and ovine mainly) during the following two years (Table 1). First, we analysed the state-of-art of pyric herbivory in south-Europe and then we compiled the lessons learned during the implementation of the PE, information that is presented in the results section. For two years we monitored the implemented practices (burning and grazing) and studied their effects on the environment (soil and vegetation) while ensuring conditions of animal welfare (body condition and weight) in the pilot experiments. In addition, the project tested new technological tools for livestock monitoring (GPS) and for surveys of fuel and burns (by drone flights). The vegetation dynamics was monitored by field surveys (floristic inventories, biomass measurements) and by drone images to determine shrub regrowth and floristic diversity. Soil physical, chemical and biological properties were also measured in six of the PE to determine the burning and grazing effects in the short and the mid-term.

Results: state-of-art in south-Europe and lessons learned from the project

The legislation of each region, the meteorology and the people-training capacity are the factors that determine the potential of a given region to execute prescribed burnings with environmental and preventive purposes in SW Europe. In the early eighties, the first countries that developed fire-use policies and burning by specialized teams were Portugal and France, and Spain progressed in the same direction afterwards (Canals, 2019). Nowadays, the legislation on fire is inconsistent among regions, and different situations occur, from the prohibition to the promotion of technical burns. When permitted, a strict calendar for the implementation of burns must be respected, constraining them to late autumn and early spring (the cold and rainy period of the year). Regulations also indicate which personnel are allowed to do the burns, after requesting permission from the authorities, and establish an accurate burning action protocol to avoid fire outbreaks. In all cases, burns must be applied under specific meteorological

Table 1. Location of the eight pilot experiments of the project and the applied treatments.

PE	Location	Initial treatment	Livestock specie/Breed
PE-1	Aquitania	Burning	Sheep/Basco Béarnaise
PE-2	Navarra	Burning	Horses/Burguete
PE-3	Galicia	Burning	Horses/Caballo Gallego
PE-4	Trás-os-Montes	Mechanical clearing	Sheep/Churra Galega Bragançana
PE-5	Vila Real	Burning	Horses/Garrano
PE-6	Andalucía	Burning	Sheep/Segureño
PE-7	Cataluña	Burning	Sheep/Ripollesa
PE-8	Easter Pyrenees	Burning	Autochthonous sheep and cattle



conditions that enable the control of flames: low environmental temperature, low wind speed and high soil humidity are the most important. These environmental conditions ensure burns of low intensity. Despite high flame temperatures (e.g. >700 °C) being reached at certain moments, and at ground surface (e.g. >300 °C), changes in topsoil temperature are hardly observed, indicating that these burnings are not at all comparable to wildfire events (high temperatures maintained over time, e.g. >800 °C).

The project also highlights the importance of operating in small (but well-selected) areas through a multi-year grazing plan to increase the efficiency and to obtain the desired objectives of consolidating mosaic landscapes. The animals selected for this environmental grazing must be robust (native breeds) and respond to a particular physiological state of low energy needs, to ensure their physical well-being and their tolerance to stressful situations. Two main different grazing management strategies, which depended on the domestic herbivore, arose as particularly successful in this project (Table 1): medium-sized (less than 700 animals) sheep flocks guided by a shepherd which grazed in a large area that included the pilot plot (of around 2-6 ha) according to a previously established itinerary; and small herds of equines (around 5 mares) that rotated between different fenced plots (of around 2-3 ha $plot^{-1}$) during selected periods of the year considering the animals' needs, the biomass offer of grasses and the shrub resprout. In addition to a shepherd or fences, implementing a targeted grazing implies the establishment of mobile water points and food baits to supplement animal's diet and to increase the pressure of the herbivore in the avoided areas.

Conclusions

The practice of the pyric herbivory is a useful tool to be applied in particular areas of the territory, i.e. critical points for wildfire extinction and wild-urban interfaces. This multidisciplinary project established the basis of a suitable use of the pyric herbivory in SW Europe for the purpose of maintaining the biodiversity of abandoned mountain grasslands while generating resilient landscapes facing the current risks of the global change.

Acknowledgements

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References

Canals, R.M. (2019) Landscape in motion: revisiting the role of key disturbances for the preservation of mountain ecosystems. *Cuadernos de Investigación Geográfica* 45, 515-531.

Fernández-Giménez, M.E. and Fillat, F. (2012) Pyrenean pastoralists' ecological knowledge: Documentation and application to natural resource management and adaptation. *Human Ecology* 40, 287-300. https://doi.org/10.1007/s10745-012-9463-x

Fuhlendorf, S.D., Engle, D.M., Kerby, J. and Hamilton, R. (2009) Pyric herbivory: Rewilding landscapes through the recoupling of fire and grazing. *Conservation Biology* 23, 588-598.

Múgica, L., Canals, R.M., San Emeterio, L. and Peralta, J. (2021) Decoupling of traditional burnings and grazing regimes alters plant diversity and dominant species competition in high-mountain grasslands. *Science of the Total Environment* 790, 147917. https://doi.org/10.1016/j.scitotenv.2021.147917

PastureBase Ireland - the adoption of grassland knowledge on Irish grassland farms

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Abstract

Grass enables low-cost systems of milk and meat production and promotes a sustainable, green, and high-quality image of ruminant production across the world. Through a combination of climate and soil type, Ireland possesses the ability to grow large quantities of high-quality grass and convert it into high quality grass-based milk and meat products. In 2013, PastureBase Ireland (PBI), the national grassland database was developed. The objective of this development was to increase the level of pasture measurement on farms and increase/improve grazing management practice. In recent years there has been a large uptake of this grassland technology. The number of grassland farmers completing 20 or more grass cover measurements more than doubled, increasing from 708 in 2016 to 1,739 in 2020. The mean DM production on dairy farms has ranged from 11.6 t dry matter (DM) ha^{-1} to 14.3 t DM ha^{-1} across the years; however, there has always been a large range (21.7-5.8) in DM production between farms. The continued adoption of best grazing management practices at farm level will be critical in confronting the ongoing environmental challenges at farm level.

Keywords: pasture management, grazing management, PastureBase, grassland database

Introduction

PastureBase Ireland (PBI) (Hanrahan *et al.*, 2017) is an internet-based grassland management programme for all Irish grassland farmers. In operation since 2013, it offers farmers 'grassland decision support' and stores a vast quantity of grassland data from dairy, beef and sheep farmers in a central national database used for research. In total there are over 7,000 farms registered on PBI. This includes research, commercial and student accounts. In 2021, dairy farms account for approximately 600,000 cows, >30% of the national dairy herd been managed through PBI. During the first three months of 2021, there were 3,821 individual commercial farms that recorded >10 covers. Since the introduction of the PBI Grass App, there has been a steady increase in the number of commercial farms recording fertilizer data. Approximately 50% of all grass covers are now uploaded to the database from the PBI app.

Dataset and engagement

The dataset in this paper is composed of 983 dairy and drystock farms who completed at least 30 farm covers in PBI in 2020. By choosing farms with high level of farm cover measurement, the level of grass production is quantified better. The number of grass covers completed by farmers using PBI has steadily increased from 14 to 20 in the period 2017 to 2021. Generally, as the numbers of measurements increase in the system, the accuracy and dependability of the farm data also increases.

The mean DM production on dairy farms has ranged from 11.6 t DM ha^{-1} to 14.3 t DM ha^{-1} across the years, and there has always been a large range (21.7-5.8) in DM production between farms (Figure 1). The number of grazing events per paddock is a grazing management efficiency measure derived from PBI. This measure shows how many actual grazing or silage harvest took place on paddocks in the growing season. The number of grazing events varied from 7.5 to 8.6 on dairy farms and varied from 5.0 to 6.3 on drystock farms (Figure 2).

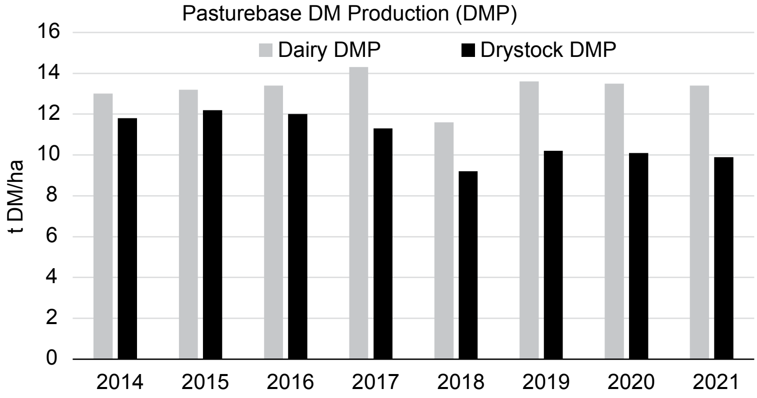

Figure 1. Total grass DM production (t DM ha^{-1}) on dairy and drystock farms recorded on PastureBase Ireland with 30 or greater (dairy) and 20 or greater (drystock) grass covers measurements from 2014-2021.

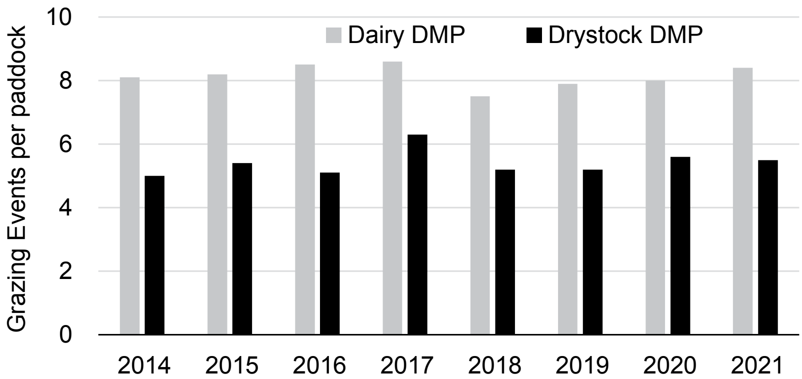

Figure 2. Total number of grazing events in dairy and drystock farms with 30 or greater (dairy) and 20 or greater grass covers measurements from 2014-2021.

Figure 3 shows the association between grazing/silage events with total DM production, to which the R^2 is 0.47. This relationship clearly shows the influence of increasing grazing events across the farm. For every extra grazing the pre-grazing yield increases by 1,293 kg DM ha^{-1}. Increasing the number of grazing events per paddock was clearly established as an avenue of increasing grazing utilization across the farm.

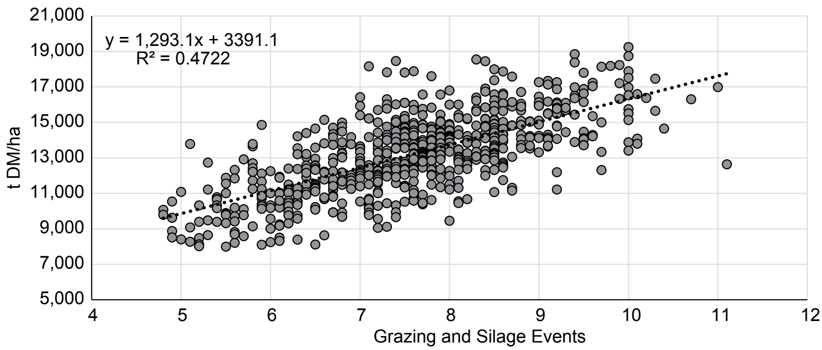

Figure 3. The association between grazing and silage events with total DM production across farms in 2019.

Conclusions

An important influence of grass production on farms is to increase the grazing events on individual farm paddocks. In the past 8 years there has been increased uptake of PBI grassland technology in Ireland, and this is further enhanced with the development of the MoSt grass growth model (Ruelle *et al.*, 2018). Given the positive effect of grazing appropriate pre-gazing yield on the reduction of greenhouse gas emissions (Wims *et al.*, 2010), the continued uptake of effective grazing technology will continue in Ireland.

References

Hanrahan L., Geoghegan A., O'Donovan M., Griffith V., Ruelle E., Wallace M. and Shalloo L. (2017) PastureBase Ireland: A grassland decision support system and national database. *Computers and Electronics in Agriculture* 136, 193-201.

Ruelle E., Hennessy D. and Delaby L. (2018) Development of the Moorepark St Gilles grass growth model (MoSt GG model): a predictive model for grass growth for pasture based systems. *European Journal of Agronomy* 99, 80-91.

Wims C.M., Deighton M.H. Lewis E., O'Loughlin B., Delaby L., Boland T. and O'Donovan M. (2010) Effect of pregrazing herbage mass on methane production, dry matter intake and milk production of grazing dairy cows during the mid season period. *Journal of Dairy Science* 93, 4976-4985.

Monitoring warming of silage with IoT-based tool may help to predict silage quality

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Abstract

Making silage is a well-known and well-researched process with known quality parameters. However, it is still possible and not very rare for unwanted aerobic fermentation to continue in parts of the bunker, which appears as increased temperature, and eventually lowered feeding value of the silage. For this field study we used a practical tool made for remote monitoring of silage temperature. Temperature was monitored with needle-like rods having sensors inside. By taking samples close to the sensor points our objective was to research if the rise of temperature could be used to predict silage quality. A total of 15 rods in three lines and five rows were installed in a bunker silo to measure temperature. Height of silage bunker was 250 cm. Our results show warming happens especially in the top layer of the silage (-50 cm). The results show that continuous warming that lasts for over 20 days results in silage deterioration which is shown first as lowered sugar content and later also lower eating index.

Keywords: feed management, silage temperature monitoring, silage heat modelling, silage quality

Introduction

The production of high-quality silage by the farmer is essential for the economy of dairy feeding. Lower quality leads to increased purchase of supplementary fodder which increases transportation both locally and globally (Green *et al.*, 2009). In order to preserve the nutritional quality of the silage, essential conditions need to be met during the storage process. Respiration is the primary cause of silage quality loss, and this depends on the supply of oxygen (O_2), heat, and water (McDonald *et al.*, 2002). When making silage it is important to pack and seal the stack to prevent build-up of aerobic micro-organisms and ensure subsequent stability of silage. It is not rare for unwanted aerobic fermentation to continue after sealing, even for several weeks. The change from desirable to unwanted fermentation process is difficult to detect without opening the storage. In this project we focused on creating a practical, IoT-based tool for remote and continuous monitoring of the temperature in the silage.

Materials and methods

This study was carried out at a private dairy farm in the Jokioinen municipality in south-western Finland in 2020. The study was a part of EIP-Agri project Good for Cattle (2019-2022). The study started on 15 June and ended on 16 November. Rods of 3 m, each having three temperature sensors inside, were inserted into the silage. The hole in the plastic was sealed to prevent airflow into the silage during storage. Each rod was connected wirelessly to a nearby router that sent the data hourly to the server of the Quanturi company, from where it was downloaded for further analyses. Samples of the silage for quality analyses were taken with a drill. The samples were analysed by the Valio feed laboratory in Seinäjoki, Finland. Statistical tests were run using JMP software. The temperature rod that we used was developed for this project by Quanturi and is now commercially available.

Results and discussion

Our results on long-term temperature monitoring from June 2020 to November 2020 (154 days) show that warming takes place especially in the surface part of the silage (Figure 1). Fermentation uses up sugars (Figure 2) and when extended it also results in further deterioration of the silage, shown as lowered eating index (Figure 3). Our results are in accordance with Huhtanen *et al.* (2007) and Kung (2011). Our earlier results show that in bigger silos (W 10 m) heat remains for a longer time, whereas smaller silos (W 6 m) cool faster. Fodder sample drilling was a challenging task and it may explain why some dispersion of quality parameters and clear outliers were found in the statistical analyses.

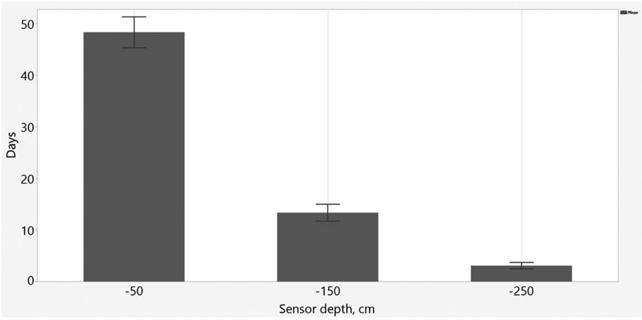

Figure 1. Number of days of incremental daily temperatures in bunker silo measured at the surface (-50 cm), in the middle (-150 cm) and at the bottom (-250 cm) sections of silage. Each mean (± standard error) represents 11 measuring points.

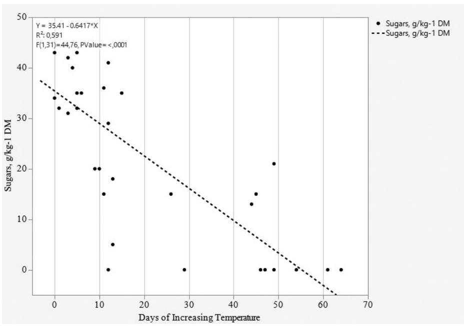

Figure 2. Relationship between sugar content (g kg^{-1} dry matter(DM)) and number of days of incremental daily temperatures in grass silage.

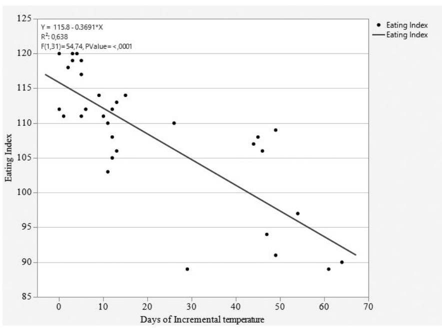

Figure 3. Relationship between D-value (g kg dry $matter^{-1}$) at the end of storage time (154 d) and days of incremental temperature during storage.

Conclusions

Because silage has such an important role in the economics of milk production, early characterization of its quality is decisive. The IoT-based tool used in this field study for temperature monitoring was durable and useful for our research. Because the study used only one bunker and one crop of silage, further research is needed, e.g. to determine practical guidelines for acceptable temperature development of silage during storage.

Acknowledgements

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References

Green O., Nadimi S.E., Blanes-Vidal V., Jörgensen R.N., Drejer Storm I.M.L. and Sörensen C.G. (2009) Monitoring and modelling temperature variations inside silage stack using novel sensor network. *Computers and Electronic in Agriculture* 69, 149-157.

Huhtanen P., Rinne M. and Nousiainen J. (2007) Evaluation of the factors affecting silage intake of dairy cows: a revision of the relative silage dry-matter intake index. *Animal* 1(5), 758-770.

Kung L. (2011) Silage temperatures: how hot is too hot. Available at: https://cdn.canr.udel.edu/wp-content/uploads/2014/02/HowHotisTooHot-2011.pdf

McDonald P., Edwards R.A., Greenhalgh J.F.D. and Morgan C.A. (2002) *Animal Nutrition* 6th edn. Pearson, Harlow, UK, pp. 521-522.

Farmer led innovation in the use of multi-species swards on Northern Ireland farms

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Abstract

There is evidence to suggest that increasing the diversity of plant species (multi-species swards (MSS)) can counteract some of the challenges faced by Northern Ireland (NI) ruminant livestock farmers. There are many suggested benefits from incorporating a mix of grass, legume and herb species into grazing platforms, such as their deep rooting properties, improved soil health and reduction in the requirement for manufactured nitrogen fertilizer input. However, there is a considerable lack of information surrounding the management of MSS on farms in NI. AgriSearch and AFBI have been working with eight NI commercial farmers trialling MSS on their farms through the SUPER-G and EcoSward projects and a European Innovation Partnership (EIP) Operational Group. With support from AgriSearch, AFBI and Queen's University the farms involved have trialled a range of seed mixtures and establishment options. Initial results have shown the MSS are significantly more drought resilient and can produce comparable dry matter yields to conventional perennial ryegrass dominant (PRG) swards with lower fertilizer inputs. Furthermore, animals grazing MSS required less use of anthelmintics than those on grass swards. In addition to the knowledge provided by researchers, the peer-to-peer learning and support has been beneficial, particularly regarding establishment techniques and initial management of MSS.

Keywords: multi-species swards, herbal lays, peer learning

Introduction

Farmers in Northern Ireland (NI) are increasingly facing financial, production and environmental challenges. Low profitability in beef and sheep production enterprises in particular is a real threat to the viability of the NI livestock sector. Finding a suitable balance between maintaining profitable and sustainable livestock performance from grassland and improving farm ecosystem service provision is critical to sustaining farm businesses and the wider industry for the future.

Multi-species swards (also referred to as species-rich, herbal leys or diverse grasslands; MSS) are communities comprised of grass, legume and herb species. There is a growing body of evidence to suggest that increasing the diversity of plant species in grassland can meet many of the aforementioned challenges, delivering a wide range of ecosystem services, reducing management costs and positively influencing sustainable livestock production.

Since grassland is the predominant crop in Northern Ireland, incorporation of MSS presents a significant opportunity for the livestock sector. Success will, however, be dependent on uptake and whilst the potential benefits of MSS are numerous there is a distinct lack of information on their establishment and initial sward management for NI commercial beef and sheep farmers.

Materials and methods

The first project (undertaken as part of the SUPER-G project) involved the establishment of MSS on seven dairy, beef and sheep farms across NI containing perennial ryegrass, white clover, chicory and plantain alongside a control mix containing the same perennial ryegrass and white clover but without the plantain and chicory. The swards were established during autumn 2019 and spring 2020. The farmers provided sward management information, including pre-and post-grazing sward covers and organic and artificial fertilizer application rates. In addition, botanical assessments, herbage quality and mineral samples were taken three times per year (spring, summer and autumn).

Having been encouraged by their initial experiences, the beef and sheep farmers involved in the SUPER-G project, with assistance from AgriSearch, applied for funding for a European Innovation Partnership operational group to further investigate the feasibility of MSS for beef and sheep production. The first stage of the project investigated the most appropriate MSS mixtures to use on their individual farms. They were assisted in this by a literature review undertaken by AFBI as part of the project (Lowe *et al.*, 2021). Scientists from AFBI and Queen's University are also members of the EIP operational group.

The operational group sought to examine specifically the challenges in establishing and successfully managing MSS and, crucially, communicating both the benefits and challenges to other farmers. A series of 'meet the farmer' videos were recorded to introduce the farmers involved in the project, explain their interest in MSS and outline initial experiences in establishing the swards. Animal performance will be measured on-farm in the 2022 grazing season. This will compare the performance of ewe and lambs, finishing lambs or growing cattle on multi-species swards compared with either perennial ryegrass or perennial ryegrass with white clover swards.

A farm walk was held in September 2021 with each of the farmers communicating their experiences of establishing and managing multi-species swards.

Results and discussion

The farmers involved used one of three establishment methods which included full ploughing and cultivation, minimal cultivation, and surface seeding (with and without burning off the existing sward). In situations where complete cultivation was used, farmers who combined this with stale seed-bed techniques found reduced broad-leaved weed growth in the newly established sward.

Many of the farms involved are located in the eastern parts of County Down, which in recent years have experienced regular late spring/early summer droughts which further narrows the opportunities for reseeding. Autumn reseeds have lowered weed burden but from a practical perspective has resulted in the fields being out of production for a longer period of time.

The farmers involved quickly realized that a change of mindset was needed to manage these swards, in that they take longer to establish than a conventional reseed and require different early management techniques. These include using less artificial N fertilizer, along with higher entry and residual sward heights. In addition, when grazing MSS, the farmers aimed to increase the grazing rotation length to 3-4 weeks, to allow swards to recover and in an attempt to improve herb persistency.

While it is still relatively early in the project, the farmers involved have observed that the MSS perform considerably better than perennial ryegrass monocultures especially during drought events. Initial findings from the 2021 grazing season show that the MSS sown as part of the SUPER-G/EcoSward trial had a 7.4% higher dry matter yield than the control mixture of perennial ryegrass and white clover, while using 11.2% less nitrogen.

Initial herbage mineral analysis from the SUPER-G/EcoSward farm sites also show that the MSS have considerably higher mineral content (44% higher for boron, 11% higher for copper, 32% higher for calcium and 15% higher for phosphorus) which merits further investigation.

All the farmers involved in the EIP group are intending to establish additional MSS in the future. There has also been a great deal of interest in this topic in NI. Over 100 farmers and advisers attended a farm walk on this topic held in September 2021. Of those who attended, 94% rated the presentations as 'very good' or 'excellent', with many of the farmers who attended expressing an interest in establishing their own MSS in the future.

One major challenge is that there are very few farm advisers that currently have knowledge of managing MSS. The project is seeking to engage with local farm advisers to help address this issue.

Conclusions

While further institute-based research on MSS is very much needed, the experiences gained by sowing a selection of MSS sward types across a range of farms using a variety of establishment methods has been most beneficial. For example, the research scientists involved in the study have found the farmers' experience invaluable in drawing up management protocols for institute-based research trials. Ultimately, farmers learn best through peer learning alongside support from research scientists. The operational group has created an effective mutual support network, has highlighted areas for further research and has inspired many other farmers in NI to establish their own MSS.

Acknowledgements

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References

Lowe D.E., Perkins L., Rutherford N. and Lively F.O. (2021) A review of multi-species swards for beef and sheep. (Published by the Multi-Species Swards for Beef & Sheep EIP Group). Available at: https://www.agrisearch.org/add-file-to-publications/a-review-of-multi-species-swards-for-beef-and-sheep/viewdocument/490

Grass growth prediction in Ireland to improve grazing management practice

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Abstract

In pasture-based systems, farmers need to make daily management decisions to ensure that their livestock have enough feed and that it is of high quality, both during the grazing season and during the housing period. Being able to predict grass growth for the following week at farm level helps farmers to better anticipate variations in grass growth. The Moorepark St Gilles Grass Growth (MoSt GG) model works at the paddock and farm level. The model is currently used on 78 farms across Ireland. Each Tuesday, grass growth predictions for the following week are communicated to the farmers involved in the project as well as the grassland industry. The predictions are published in a weekly newsletter and other media sources. While only 78 farms are currently involved in predictions, the model will soon be incorporated into PastureBase Ireland (PBI) to predict specific farm growth for all PBI users.

Keywords: grass growth, prediction, decision support, model

Introduction

Each additional kilogram of grass dry matter (DM) utilized on farm increases farm profitability (Hanrahan *et al.*, 2017). In Ireland, an increase of 1 tonne of grass dry matter (DM) ha^{-1} eaten will increase profit by €105 ha^{-1} on beef farms and €181 ha^{-1} on dairy farms. PastureBase Ireland (PBI; Hanrahan *et al.*, 2017) is the Irish grassland management decision tool for farmers. It helps farmers to manage the grass on their farms, identify surpluses or deficits in the supply of grass and take appropriate action. At present, farmers can only make decisions within PBI based on historical information. The Moorepark St-Gilles Grass Growth (MoSt GG; Ruelle *et al.*, 2018) is currently used to predict farm grass growth for the forthcoming week with a view to incorporating it into PBI.

Presentation of the predictions

The MoSt GG model (Ruelle *et al.*, 2018) was used to predict grass growth on farm. The project started in 2018 after a low grass-growing year due to a cold spring and a drought in the summer. Initially, grass growth was predicted on 30 farms. Since then, this number has increased to 78, predominantly commercial farms. Most of the data required to undertake the prediction are collected in PBI (Hanrahan *et al.*, 2017). For each farm, the data necessary to run the model present in PBI are: paddocks and their area, grazing and cutting dates, number of grazing animals and their supplementation, and nitrogen fertilization (chemical and organic) rate and date of application. Other data required include the soil type for each paddock (which is determined using the Irish Soil Information System, https://www.teagasc.ie/environment/soil/irish-soil-information-system/), and historical and forecast weather data (provided by Met Éireann, www.met.ie). The 78 farms chosen for this programme are managed by farmers who regularly (minimum once a week during the main grazing season) enter all the information required to run the model. They are evenly distributed across the country, representing a variability of locations and soil types (Figure 1).

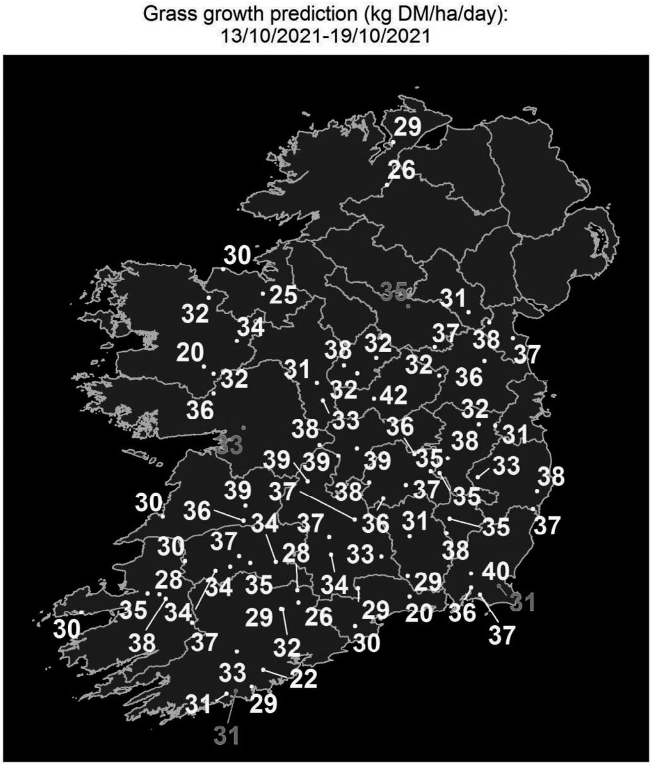

Figure 1. Grass growth predictions (kg DM ha^{-1} day^{-1}) for the week of September 13, 2021. Each dot corresponds to the exact location of the simulated farm. White corresponds to commercial farms, grey to experimental farms.

The data are sent weekly to the involved farmers in the form of a map (Figure 1), but also by e-mail to Teagasc advisors with other information such as predicted rainfall and soil temperatures to help them in their advice to farmers. Grass growth predictions are also available to the public each week through the Grass10 newsletter on the PBI website (www.pbi.ie). The predictions are also sent to the Department of Agriculture, Food and the Marine, Coops, agribusiness agencies, and agrimedia. Since August 2020, the grass growth predictions are also presented each Sunday on Ireland's national television channel, RTÉ 1, as part of the 'Weekly Meterological Farming Forecast'.

Feedback and evaluation

Farmers' feedback is very positive. A survey conducted in 2019 found that farmers rated the accuracy of the model 3.7/5.0 and its usefulness 4.0/5.0. More importantly, 70% of farmers said they had adapted their management based on predictions. The model is regularly evaluated against the values recorded in PBI Ireland measured data. Using forecasted weather the RMSE, at country level was of 6.9 kg DM ha^{-1} day^{-1} in 2019 and 6.0 kg DM ha^{-1} day^{-1} in 2020. While this is an evaluation of the prediction, this is not an evaluation of the model as the weather data used are forecasted data. When using historical weather data instead of forecasts for the same period, the RMSE was reduced to 6.0 kg DM ha^{-1} day^{-1} in 2019, but increased to 7.1 kg DM ha^{-1} day^{-1} in 2020. This highlights one of the weaknesses of the system, because what is called historical weather is in fact modelled weather. We have noticed that in those data rainfall is often overestimated (10-20%), especially during small spring droughts (as shown, for example, by the overestimation in spring 2020).

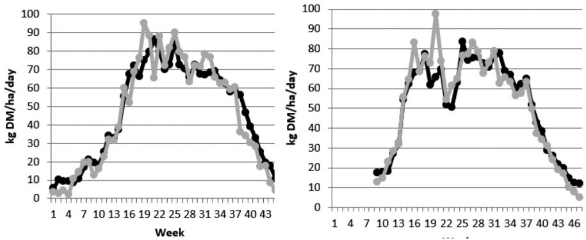

Figure 2. Comparison between the data sent to farmers (light grey) and the data recorded into PBI (black) in 2019 (A) and 2020 (B); average of all farms in the project (35 in 2019 and 58 in 2020).

At farm level, the RMSE values were similar for both years, ranging from 10.6 to 24.9 kg DM ha^{-1} day^{-1} with an average of 16.5 kg DM ha^{-1} day^{-1}. It was noted that, in most cases, as farmers measured their grass more frequently, the model provides more accurate predictions. Incorrect historical weather information and inaccurate measurements taken by farmers were also factors contributing to explain the high RMSE values in some farms.

Conclusions

The MoSt GG model is used weekly to inform farmers and the industry about future grass growth. The accuracy of the model is satisfactory and allows the farmers to anticipate and make informed management decisions. At present, only 78 farms have grass growth forecasts, but the model will soon be incorporated into PBI, allowing any farmer who enters the required information weekly to obtain accurate predictions for his/her farm.

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References

Hanrahan L., Geoghegan A., O'Donovan M., Griffith V., Ruelle E., Wallace M. and Shalloo L. (2017) PastureBase Ireland: A grassland decision support system and national database. *Computers and Electronics in Agriculture* 136, 193-201.

Ruelle E., Hennessy D. and Delaby L. (2018) Development of the Moorepark St Gilles grass growth model (MoSt GG model): a predictive model for grass growth for pasture based systems. *European Journal of Agronomy* 99, 80-91.

Communicating knowledge on grassland management using videos and the internet

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Abstract

Digital information is of growing importance to farmers and land managers, as a survey of stakeholders in 2020 revealed. However, too often the practical implications of scientific research are not communicated to non-scientists. To close the gap between science and practice, we produced various educational videos and web contents about pasture management and maintenance in a participatory process. Each 10 min video features a farmer and an expert, each of whom presents for about half of the time. Implementing peer and expert teaching facilitates knowledge transfer to the heterogeneous agricultural community. We established a standardized procedure. Before creating videos and web content, we gathered all available knowledge and elaborated take-home messages. These were discussed in a group of experts (scientists and farm advisers) and together with farmers to elaborate a coherent and understandable scenario. Special emphasis was placed on adapting pasture management and mechanical intervention rather than the use of herbicides. The standardized participatory procedure to produce educational videos and web contents has proven to be cost and time-efficient. We are convinced that communicating scientific knowledge by means of digital channels will improve the adoption in agricultural practice.

Keywords: pasture management, educational video, knowledge transfer, communication, website, YouTube

Introduction

Digital information is of increasing interest to farmers and land managers (Chivers *et al.* 2021; Hoffet and Mettler, 2021). Scientists do a lot of research on providing advice and solutions for farmers. However, these results are too often not accessible to non-scientists. We decided to close the gap between science and practice by presenting the information in an easily accessible medium. Therefore, we produced several educational videos and web contents about pasture management and maintenance.

A severe gap in knowledge transfer was identified for weed control on alpine summer pastures (Hoffet and Mettler, 2021). These grasslands, covering a third of the agriculturally used land in Switzerland (Lauber *et al.*, 2013) provide important ecological and cultural values and are important for the Swiss identity. However, these areas are under pressure from problematic plants that are reducing the forage quality and ecological value of the pastures. Therefore, direct payments to alpine summer farms require careful management of pastures in order to maintain biodiversity, openness and beauty (Swiss Ordinance 910.13).

Due to its seasonal nature, work on alpine pastures is frequently carried out by external staff without formal training (Calabrese *et al.*, 2014). Staff sometimes change on an annual basis, impeding knowledge accumulation and transfer. Unfortunately, advisory documents concerning pasture management and weed control are scattered in different formats and from different organizations. Therefore, we set up a project to synthetize existing knowledge and to make this information easily accessible through a new website and educational videos.

Materials and methods

Selection of topics

The activities were launched on the topic of problematic plants on alpine pastures. Species were prioritized and selected for a first series of videos. The videos of around ten minutes duration always featured a farmer and an expert, presenting for about half of the time. Combining both the peer and expert teaching, facilitates knowledge transfer to the heterogeneous community of alpine staff (Franz *et al.*, 2010). Special emphasis was placed on adapting pasture management and mechanical intervention rather than the use of herbicides.

Video production

Videos were prepared in a standardized participatory procedure involving scientists, farm advisers and farmers (Table 1): (1) Gathering the available knowledge by literature reviews and interviews was time-consuming since information was often scattered in scientific literature and in practical experience. It was, however, used for the video production and the web contents. (2) The video production was planned by elaborating a screenplay, which listed messages and contents to be presented, together with ideas for possible image settings. (3) The video was usually filmed during one day. A second day was occasionally required for additional scenes, especially if several seasonal aspects of the plant needed to be captured. Filming was carried out using a single-lens reflex camera with external microphone and complemented by a drone for overview scenes. (4) The video was edited and (5) the draft version was revised by the group of authors. (6) The final version was edited. (7) Finally, speeches were transcribed. Because no automated speech-to-text translation was available for Swiss German, this needed to be done by hand. The video was subtitled in German, French, Italian and English, and released on YouTube.

Background information

Alongside the video, detailed information was prepared for release on the new website. As for the videos, a standardized format was established. The primary sections were (A) occurrence and distribution of the problematic plant, (B) situation analysis, (C) regulation measures, (D) adjustment of management, (E) mechanical intervention and (F) chemical regulation. After consultation within the editorial team and with additional experts and practitioners, the information was published on the web.

Results and discussion

The website www.patura-alpina.ch (Figure 1) was released in summer 2019 and is freely available in two languages (German and French). In 2021, it contained information about six plant species groups, commonly perceived problematic on alpine pastures: (1) alpine dockweed (*Rumex alpinus*), (2) white helleborum (*Veratrum album*), (3) rush (*Juncus effusus* and *Juncus inflexus*), (4) alpine ragwort (*Senecio*

Table 1. The seven key steps to a successful video and the amount of time approximately needed per video by the editorial team (ET) and the filming team (FT).

	Action	Who	Time (days)
1	Summary of available knowledge (scientific literature and farmers experiences)	ET	3-5
2	Elaborate key messages in form of a video script, planning of video	ET/FT	1-1.5
3	Filming	FT	1-2
4	Raw video editing	FT	1-1.5
5	Consultation round	ET	1
6	Final video editing	FT	0.5
7	Transcript and translation of subtitles and release on YouTube	FT	1-2
Total per video			8.5-13.5

alpinum) (5) bracken (*Pteridium aquilinum* and *Dryopteris filix-mas*) and (6) dwarf shrubs (*Juniperus communis* and *Rhododendron hirsutum*). Further videos and website contents on thistles (*Cirsium palustre* and *Cirsium arvense*) and green alder (*Alnus viridis*) are scheduled for release in early 2022. The website is continually updated and complemented with testimonies of farmers. Information is also available in the smartphone app of Agridea, the Swiss centre for agricultural advisory and extension services. Since the website was released, the videos have been viewed nearly 15,000 times in total (Table 2). Each video was viewed 3.3 times per day. Direct feedback received from farmers and farm advisers was generally positive.

Figure 1. Websites like www.patura-alpina.ch make information easy to access.

Table 2. Date of release and number of views of the videos on 1.12.2021.

Topic	Date of release	Days	Views	Views day^{-1}
Rumex alpinus	10.04.2019	966	3,201	3.3
Veratrum album	09.04.2019	967	3,863	4.0
Senecio alpinus	26.10.2019	767	1,232	1.6
Juncus effusus / J. inflexus	25.12.2019	707	1,978	2.8
Pteridium aquilinum and *Dryopteris filix-mas*	02.11.2019	760	2,631	3.5
Dwarf shrubs	06.02.2021	298	1,848	6.2
Total			14,753	

Conclusions

Communicating to farmers by means of video and web contents is a contemporary means of knowledge transfer. The standardized participatory procedure involving scientists, extension services and farmers, to produce educational videos and web content may inspire similar approaches on other topics in grassland science.

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References

Calabrese C., Mann S. and Dumondel M. (2014) Alpine farming in Switzerland: discerning a lifestyle-driven labor supply. *Review of Social Economy* 72(2), 137-156.

Chivers C.A., Bliss K., de Boon A., Lishman L., Schillings J., Smith R. and Rose D.C. (2021) Videos and podcasts for delivering agricultural extension: achieving credibility, relevance, legitimacy and accessibility. *Journal of Agricultural Education and Extension*. https://doi.org/10.1080/1389224X.2021.1997771

Franz N.K., Piercy F., Donaldson J. and Richard R. (2010) How farmers learn: Implications for agricultural educations. *Journal of Rural Social Sciences* 25(1), 37.

Hoffet F. and Mettler D. (2021) Umfrage Wissenstransfer in der Alpwirtschaft. AGRIDEA available at: https://docplayer.org/207777855-Umfrage-wissenstransfer-in-der-alpwirtschaft-agridea-und-sav-franziska-hoffet-daniel-mettler-agridea.html

Lauber S., Herzog F., Seidl I., Böni R., Bürgi M., Gmür P., ... and Wunderli R. (2013) Zukunft der Schweizer Alpwirtschaft. Fakten, Analysen und Denkanstösse aus dem Forschungsprogramm AlpFUTUR. Eidgenössische Forschungsanstalt für Wald, Schnee und Landschaft WSL, Birmensdorf, Switzerland.

Developing sustainable management of Alpine and pre-Alpine grasslands – from research to practice

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Abstract

The SUSALPS project (www.susalps.de/en) aims at improving our knowledge on how climate change and agricultural management affect ecosystem functions of Alpine and pre-Alpine grasslands. The experimental sites in southern Germany combine a space-for-time approach with an intensive and an extensive fertilizer and cutting regime to quantify the effects of climate change (i.e. increased temperature, changed precipitation) and management on biomass production, plant biodiversity, soil C and N turnover and fluxes, soil microbiology, and water fluxes. Linking biogeochemical-modelling approaches with remote sensing data allows the assessment and optimization of grassland ecosystem services at regional scale. Farmers' decisions on grassland management, as subject to different policies, are modelled with an agent-based socioeconomic model. Together with the biogeochemical model LandscapeDNDC, a bio-economic model is developed and tested in cooperation with local farmers. The bio-economic model is the backbone of a user-friendly decision support system (DSS) for improved and sustainable grassland management (i.e. to improve or adapt nutrient management and cutting regimes). The presentation will synthesize major project results related to vegetation and soil functions, introduce the DSS and discuss possibilities for knowledge transfer between scientists, stakeholders, and farmers while developing the DSS.

Keywords: climate change, grassland management, bio-economic model, agricultural decision support

Introduction

Forage production is an important agricultural usage of Alpine and pre-Alpine grassland soils. They provide significant economic value through supporting milk and meat production (Soussana and Lüscher, 2007). Furthermore, grassland sustains other regulating, supporting and cultural ecosystem services (ESS) (Le Clec'h *et al.*, 2019). In comparison with other ecosystems, grassland soil functions and related ESS are highly vulnerable not only to climate change but also to management intensification (Felipe-Lucia *et al.*, 2020; Lamarque *et al.*, 2014). Still little is known about which grassland management strategy maximizes the supply of multiple ESS (Neyret *et al.*, 2020). Knowledge about synergies and trade-offs between grassland ESS, e.g. productivity vs biodiversity and soil C stocks (Felipe-Lucia *et al.*, 2020) is of great importance for adapting and optimizing grassland management. The SUSALPS project addresses these challenges for pre-Alpine and Alpine grassland systems. It combines natural and socio-economic sciences from plot to regional scale, combining field, laboratory and modelling approaches. The results feed into a decision support system, which is developed in close cooperation with farmers and other stakeholders to provide a tool to adapt agricultural management towards improved environmental and economic sustainability.

Materials and methods

The core study sites of the SUSALPS project are located in Alpine/pre-Alpine grassland areas in southern Germany. They cover an elevation gradient from 1,260 m a.s.l. to 600 m a.s.l. in an Alpine/pre-Alpine environment, and a further site in Bayreuth (BT, northern Bavaria, Germany) at 350 m a.s.l. In 2016, intact plant-soil mesocosms were taken from the sites Esterberg (EB, 1,260 m a.s.l.), Graswang (GW, 860 m a.s.l.) and Fendt (FE, 600 m a.s.l.) and translocated downslope, i.e. (a) from EB to GW, FE and BT, (b) from GW to FE and BT, (c) from FE to BT) (Berauer *et al.*, 2019; Schlingmann *et al.*, 2020). This set-up mimicked projected climate change conditions (Smiatek *et al.*, 2009) and exposed the translocated mesocosms to higher temperatures and reduced summer precipitation. The effects on above and below ground biomass, plant species composition, microbiome, soil and plant C and N were analysed during each vegetation period after the translocation in 2016. The SUSALPS experiments were backed by data of the TERENO pre-Alpine infrastructure (Kiese *et al.*, 2018). The lysimeter network also applies a space-for-time approach and covers an elevation range from 860 to 600 m a.s.l. (GW to FE). It was installed in 2011 and provides data on above ground biomass, hydrometeorology and C and N fluxes.

At regional scale, biogeochemical processes were simulated with the LandscapeDNDC model, and supported by remote sensing data, e.g. cutting frequencies or biomass estimates. Furthermore, an agent-based socio-economic model was adopted to assess the effects of management decisions on economic benefits, but also on the quality of various ecosystem services. Both models are currently coupled to a bio-economic model framework. A decision support system (DSS) is informed by this model framework. The DSS was designed to support farmers to optimize their nutrient management and, more generally, to increase their resilience to climate change. The DSS is developed in close cooperation with potential users. Test and feedback loops with farmers help to make the DSS a user-friendly and targeted product. First, the DSS was tested with farmers involved in the SUSALPS and TERENO projects. Their feedback was implemented in the DSS. The DDS was then tested by additional farmers and agricultural consultants. Again, comments from this workshop have been taken into account. In autumn 2022 a further workshop for a larger user community is planned.

Results and discussion

Results from the translocation experiment indicated that grassland soil N (and C) stocks are decreasing and that the decline in soil organic N (SON) and SOC, which jeopardizes soil health, is accelerated by both management intensity and warming. Warming also persistently reduced the plant species richness. The extent to which climate change is promoting plant N uptake and exports, which are the key drivers for SON mining, was affected by the effects of (1) growing season temperature and (2) precipitation. Increased temperature stimulates SON mineralization and subsequently plant nutrient uptake, while summer droughts in addition to plant water stress, lead to reduced plant N and P availability. However, effects were less pronounced for species-rich and extensively managed grasslands, which showed lower N exports and reduced soil N mining, but also more stable productivity under climate warming and summer drought compared to intensively managed grasslands. Scenario simulations with the biogeochemical LandscapeDNDC model support the experimental evidence that sites fertilized with solid farmyard manure showed significantly higher SON and SOC stocks than sites fertilized with slurry. Based on these results, the DSS can be applied with site-specific conditions (e.g. management, soil, and climate). An example of a DSS results in the user interface is given in Figure 1.

Figure 1. Example of DSS results: 'traffic light system' to indicate management effects on the quality of harvest and biogeochemical indicators (A) and simulated dry matter harvest (B).

Conclusions

The project outcomes indicate that high biomass production and biodiversity conservation/C sequestration can hardly be achieved for a given grassland plot at the same time. These conflicting goals may thus only be achieved by organizing grassland management at farm and regional scales. Therefore, in addition to synthesis of experimental outcomes across climate and land use gradients, biogeochemical and agent-based socio-economic modelling as well as remote sensing approaches must be particularly dedicated to providing management recommendations at farm and regional scale.

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References

Berauer B.J., Wilfahrt P.A., Arfin-Khan M.A.S., Eibes P., von Heßberg A., Ingrisch J., … and Jentsch A. (2019) Low resistance of montane and alpine grasslands to abrupt changes in temperature and precipitation regimes *Arctic, Antarctic and Alpine Research* 51, 215-231. https://doi.org/10.1080/15230430.2019.1618116

Felipe-Lucia M.R., Soliveres S., Penone C., Fischer M., Ammer C., Boch S., … and Fiore-Donno A.M. (2020) Land-use intensity alters networks between biodiversity, ecosystem functions, and services. *Proceedings of the National Academy of Sciences* 117, 28140-28149.

Kiese R., Fersch B., Baessler C., Brosy C., Butterbach-Bahl K., Chwala C., … and Schmid H.P. (2018) The TERENO pre-Alpine observatory: integrating meteorological, hydrological, and biogeochemical measurements and modeling. *Vadose Zone Journal* 17, 180060.

Lamarque P., Lavorel S., Mouchet M. and Quetier F. (2014) Plant trait-based models identify direct and indirect effects of climate change on bundles of grassland ecosystem services. *Proceedings of the National Academy of Sciences* of the USA 111, 13751-13756. https://doi.org/10.1073/pnas.1216051111

Le Clec'h S., Finger R., Buchmann N., Gosal A.S., Hörtnagl L., Huguenin-Elie O., … and Huber R., 2019. Assessment of spatial variability of multiple ecosystem services in grasslands of different intensities. *Journal of. Environmental Management* 251, 109372. https://doi.org/10.1016/j.jenvman.2019.109372

Neyret M., Fischer M., Allan E., Hölzel N., Klaus V.H., Kleinebecker T., … and Manning P. (2020) Landscape management for grassland multifunctionality. *bioRxiv* 2020.07.17.208199.

Schlingmann M., Tobler U., Berauer B., Garcia-Franco N., Wilfahrt P., Wiesmeier M., …and Dannenmann M (2020) Intensive slurry management and climate change promote nitrogen mining from organic matter-rich montane grassland soils. *Plant and Soil* 456, 81-98. https://10.1007/s11104-020-04697-9

Smiatek G., Kunstmann H., Knoche R. and Marx A. (2009) Precipitation and temperature statistics in high-resolution regional climate models: Evaluation for the European Alps. *Journal Geophysical Research* 114, D19, 107.

Soussana J.F. and Lüscher A. (2007) Temperate grasslands and global atmospheric change: a review. *Grass and Forage Science* 62, 127-134.

Pasture evaluation program improves horse health and grassland management

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Abstract

The University of Kentucky Horse Pasture Evaluation Program monitors pasture botanical composition to determine the management practices that should be implemented on Central Kentucky horse farms. Of particular concern are pastures which contain significant populations of tall fescue *Lolium arundinaceum* (Schreb.) Darbysh; [*Schedonorus phoenix* (Scop.) Holub] because the majority of the plants are infected with a toxic fungal endophyte (*Epichloë coenophialum*). This endophyte produces a series of ergot alkaloids, with ergovaline in the highest concentration. Ergovaline causes a range of toxicities in late term pregnant mares including prolonged gestation, thickened placenta, dystocia, agalactia, and foal and mare mortality. The University of Kentucky Horse Pasture Evaluation Program utilizes ergovaline and endophyte testing and as well as pasture species composition to calculate an estimate of ergovaline in the total diet in broodmare pastures. This data is used to help farm managers determine low risk pastures for mares and target their renovation efforts to higher risk pastures.

Keywords: horses, tall fescue, ergovaline, ergot alkaloids, pregnant mares, ergovaline in total diet

Introduction

Tall fescue, *Lolium arundinaceum* (Schreb.) Darbysh; [*Schedonorus phoenix* (Scop.) Holub] a cool-season, perennial bunch type grass, dominates over 15 million ha in the southeastern US (Ball *et al.*, 2007) including approximately 2 million ha in the state of Kentucky. Most plants are infected with a common toxic endophyte, *Epichloë coenophialum*. The endophyte and plant form a symbiotic relationship, with the plant providing nutrients and a hospitable environment for the endophyte, and *E. coenophialum* increasing plant pest, drought, and grazing tolerance (Gwinn and Gavin, 1992; Arachevaleta *et al.*, 1989). However, mares grazing toxic tall fescue can exhibit prolonged gestation, dystocia, and high foal or mare mortality (Putnam *et al.*, 1991), thickened or retained placentas (Monroe *et al.*, 1988), and agalactia, limited or no milk production (Kosanke *et al.*, 1987; Thompson *et al.*, 1986).

The University of Kentucky Horse Pasture Evaluation Program (https://forages.ca.uky.edu/pasture_eval) is a fee-based service offered by University of Kentucky Forage Extension to collect on-farm pasture data and make management recommendations. This program combines species composition data with ergovaline quantification to calculate 'ergovaline in total diet', and this information is used to make pasture-specific recommendations.

Materials and methods

Pastures are sampled at 10 to 20 locations (depending on size) between April and November by trained individuals using the occupancy method (Payne *et al.*, 2021; Vogel and Masters, 2001). Categories include tall fescue, Kentucky bluegrass (*Poa pratensis* L.), orchardgrass (*Dactylis glomerata* L.), white clover (*Trifolium repens* L.), broadleaf weeds, nimblewill (*Muhlenbergia schreberi* J.F. Gmelin), and bare soil (which includes warm season annual grasses). For ergovaline analysis, plant material is harvested 7-10 cm above the ground from 10 to 20 random locations in the pasture to obtain a minimum total fresh sample weight of at least 300 g. Samples are analysed using ultra-High Performance Liquid Chromatography with fluorescence detection to quantify ergovaline, and its isomer, ergovalinine, as total ergovaline concentration, at the University of Kentucky Veterinary Diagnostic Laboratory (http://vdl.uky.edu/).

The full method is described in Lea *et al.* (2014). Additionally, twenty tall fescue tillers are collected throughout each pasture, and analysed for the presence of *E. coenophialum* using the Agrinostics tiller test kit (Watkinsville, GA) following the procedure described by Vincelli *et al.* (2017) at the University of Kentucky Regulatory Services (https://www.rs.uky.edu/home/).

From these data collected on pastures, ergovaline in total diet is calculated by multiplying ergovaline concentration by the percentage of tall fescue found from all grazable forage species: tall fescue, KY bluegrass, orchardgrass and white clover.

$$\text{Ergovaline in total diet } (\mu g\ kg^{-1}) = \frac{\%\text{Tall Fescue}}{\%\text{Tall Fescue} + \%\text{Bluegrass} + \%\text{Orchardgrass} + \%\text{White Clover}} \times \text{ergovaline}$$

Results and discussion

Because horses are known to eat randomly throughout the field, ergovaline or endophyte analysis does not provide the full understanding of ergovaline intake and therefore complicates evaluating pastures for risk. By combining this information with species composition, ergovaline in total diet can be determined and compare the risk of toxicosis from one pasture to the next.

Table 1 shows the botanical composition of pastures on two central Kentucky Horse farms in 2019, as well as the endophyte, ergovaline, and ergovaline in total diet. For this program, 200 $\mu g\ kg^{-1}$ ergovaline in total diet is the assumed threshold level for late term pregnant mares (Lea *et al.*, 2014; Lea and Smith, 2021). To simplify, Kentucky bluegrass, orchardgrass and white clover are combined as 'desirable species', and broadleaf weeds, nimblewill, warm season annual grasses and bare soil are combined as 'undesirable species'.

Pastures 1 and 3 contain similar levels of endophyte (80-85%) and ergovaline (~ 550 $\mu g\ kg^{-1}$). However, Pasture 1 is much lower in ergovaline in total diet because it contains less tall fescue and more desirable species as a percentage of total botanical composition. Pasture 1 would be considered a low risk for late term pregnant mares, while Pasture 3 would not be. Paddock N has a similar endophyte percentage to other pastures, but much higher ergovaline concentration, well over the threshold level of 200 $\mu g\ kg^{-1}$. Testing just endophyte percentage may falsely suggest that this pasture posed no more of a risk than others. Pasture 10 had ergovaline levels above the suggested threshold of 200 $\mu g\ kg^{-1}$, but because tall fescue is present in such a small proportion compared to desirable species, it is diluted below this threshold in the total diet and would be a low-risk grazing area for mares. Finally, Paddock IP2 is dominated by endophyte infected tall fescue, but ergovaline is below the detectable level of the method. Because this was sampled in late spring when levels should be high, this suggests this pasture has been seeded with a novel endophyte tall fescue, which contains an endophyte that does not produce ergovaline and therefore is safe for pregnant mares to graze.

Table 1. Contains botanical composition data as well as endophyte and ergovaline analysis and calculated Ergovaline in total diet from two thoroughbred breeding farms in Central Kentucky.

Pasture	Tall Fescue (%)	Desirable Species[1] (%)	Undesirable Species[2] (%)	Endophyte (%)	Ergovaline ($\mu g\ kg^{-1}$)	Ergovaline in total diet ($\mu g\ kg^{-1}$)
Pasture 1	23	50	28	80	552	178
Pasture 3	46	32	17	85	547	321
Paddock N	59	24	14	79	977	699
Pasture 10	7	48	38	67	289	39
Paddock IP2	67	12	20	80	<100	<100

[1] Sum of KY Bluegrass, Orchardgrass, and White Clover.

[2] Sum of broadleaf weeds, nimblewill, warm season annual grasses and bare soil.

Conclusions

Endophyte and ergovaline testing are key elements of understanding and reducing on-farm risk of tall fescue toxicosis in horses. Pasture composition measurements are an additional indicator of toxicosis risk and should always be considered before making management decisions. The University of Kentucky Horse Pasture Evaluation Program has found combining these data gives the best overall understanding of the health and risks of individual pastures, allowing managers to place mares in low-risk pastures and focus renovation efforts on higher risk pastures. Since 2005, the University of Kentucky Horse Pasture Evaluation Program has completed 281 evaluations on over 170 farms in central Kentucky, representing over 67,000 total pasture acres. Recent improvements including modified sampling methods and digital data collection have streamlined the evaluation process, providing more consistent information with a rapid turnaround.

References

Arachevaleta M., Bacon C.W., Hoveland C.S. and Radcliffe D.E. (1989) Effect of the tall fescue endophyte on plant response to environmental stress. *Agronomy Journal* 81, 83-90.

Ball D.M., Hoveland C.S. and Lacefield G.D. (2007) Cool Season Grasses. In: Ball D.M., Hoveland C.S. and Lacefield G.D. (eds.) *Southern Forages,* International Plant Nutrition Institute, Norcross, GA, USA, pp. 40-49.

Gwinn K.D. and Gavin A.M. (1992) Relationship between endophyte infestation level of tall fescue seed lots and *Rhizoctonia zeae* seedling disease. *American Phytopathological Society* 76, 911-914.

Kosanke J.L., Loch W.E., Worthy K. and Ellersieck M.R. (1987) Effect of toxic tall fescue on plasma prolactin and progesterone in pregnant pony mares. In: Proceedings of the Tenth Equine Nutrition and Physiology Symposium. Fort Collins, CO, USA 11-13 June 1987, pp. 663-668.

Lea K. and Smith S.R. (2021) On-farm monitoring of ergovaline and tall fescue composition for horse pasture management. *Toxins* 13, 683.

Lea K., Smith L., Gaskill C., Coleman R. and Smith S.R. (2014) Ergovaline stability in tall fescue based on sample handling and storage methods. *Frontiers in Chemistry* 2014: 76. https://doi.org/10.3389/fchem.2014.00076

Monroe J.L., Cross D.L., Hudson L.W., Hendricks D.M., Kennedy S.W. and Bridges W.C. (1988) Effect of selenium and endophyte-contaminated fescue on performance and reproduction in mares. *Equine Veterinary Science* 8, 148-153.

Payne K., Smith S.R. and Goff B.M. (2021) Enhanced efficiency nitrogen formulation on stockpiled tall fescue production. *Agronomy Journal* 113(2), 1596-1606, https://doi.org/10.1002/agj2.20553

Putnam M.R., Bransby D.I., Schumacher J., Boosinger T.R., Bush L., Shelby R.A., Vaughan J.T., Ball D. and Brendemuehl J.P. (1991) Effects of the fungal endophyte *Acremonium coenphialum* in fescue on pregnant mares and foal viability. *America Journal of Veterinary Research* 52, 2071-2074.

Thompson F.N., Caudle A.B., Kemppainen R.J., Nett T.M., Brown J. and Williams D.J. (1986) Thyroidal and prolactin secretion in agalactic mares. *Journal of Theriogenology* 25, 575-580.

Vincelli R., Smith S.R. and Tillery T. (2017) Sampling for the tall fescue endophyte in pasture or hay stands. University of Kentucky Extension Publication PPA-30. Available at: http://www2.ca.uky.edu/agcomm/pubs/PPA/PPA30/PPA30.pdf.

Vogel K.P. and Masters R.A. (2001) Frequency Grid-A simple tool for measuring grassland establishment. *Journal of Range Management* 54, 653-655.

Exemplary on-farm research of region-, period- and sward-specific grassland yield prediction using geoprocessing methods

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Abstract

Many farmers suffer from inaccurate and imprecise yield prediction of grasslands. Lack of information on site-specific yield and quality of harvested material prevents optimal and sustainable use across the entire value chain. For this, new and practical methods need to be developed in cooperation with farmers on their fields in order to provide them with easily accessible yield information for their grasslands. One measurement system is the rising plate meter Grasshopper®, which calculates the available herbage dry matter based on the measured compressed sward height. The calculation depends on different parameters, like the growing period or sward structure. Accordingly, prediction equations adapted to local conditions are necessary for a precise yield calculation. By on-farm research on four exemplary farms in the Black Forest, it was possible to develop precise equations for different grasslands and growing conditions in this special region of Southwest Germany. These equations are integrated into an application that enables the farmers to adapt the yield prediction to their local conditions and to obtain a precise yield map output. Thus, through the cooperation of science, farmers and companies, a tool was developed that facilitates grassland management for farmers.

Keywords: yield prediction, on-farm research, Rising Plate Meter, geoprocessing, yield map, Kriging interpolation

Introduction

The entire field of agriculture is becoming more and more digitalized through the use of sensors (Bogue, 2017). In the domain of grassland management, lack of information on yield prediction is an issue for many farmers. Digital technologies can provide a remedy and the rising plate meter (RPM) is one system whose suitability for this purpose has been studied for many years (Rayburn and Rayburn, 1998). It allows users to make precise measurements of the compressed sward height (CSH) at multiple positions field locations (McSweeney *et al.*, 2019). A yield prediction can be calculated from the values of the CSH using a linear relationship (Dillard *et al.*, 2016). The accuracy of this prediction depends on how well it is adapted to the grassland and environmental conditions (Cárdenas *et al.*, 2020). Therefore, numerous different prediction equations have been published for various regions and sward types (Hart *et al.*, 2020; O'Brien *et al.*, 2019). Three factors have been shown to substantially influence prediction: the adjustment to the region (topographical and climatic characteristics), the measurement date during the vegetation period, and the sward type, which increases the prediction accuracy the most (Cho *et al.*, 2019; Rayburn, 2020). For this reason, the objective of this study is to investigate how farmers can be supported with an easily accessible RPM-based yield prediction that is specific to the growing conditions of their grasslands.

Materials and methods

The four test sites of the study are located in the Black Forest region near to Titisee-Neustadt, South-West of Germany and managed by four different organic farmers. According to the method described by Klapp and Stählin (in Voigtländer and Voss, 1979) the sward composition was determined and three different sward types were classified following Nussbaum *et al.* (2004). To represent the entire test site, four test parts were spread over the area. The CSH of a 1 m^2 plot of every test part was measured with the RPM Grasshopper® once a week. After the height measurement the plot was cut by hand at 70 mm, and the height of the cut was checked with the RPM. The fresh weight of cut herbage was then determined, and samples oven-dried at 60 °C for more than 48 h and weighed to determine dry matter (DM) content and thus DM yield for every sample. During the vegetation periods 2020 and 2021, a total of 311 measurements were recorded at weekly intervals by this method of on-farm research. This total dataset was divided into nine sub-datasets based on the criteria of: (1) sward type (*grass-rich* with a grass proportion of >70%; *balanced* with grasses 50 to 70%; or *clover- and herb-rich* with a proportion of grasses of <50%) and (2) the usual growing periods of this region (*1st period* from April until the beginning of June; *2nd period* from beginning of June until end of July; *3rd period* from end of July until mid-September). By means of regression analysis, nine specific equations could be derived to predict the grassland yield in the Black Forest region.

The data measured with the RPM are processed semi-automatically with the geoprocessing tools of the ArcGIS ModelBuilder. For this purpose, the equations are stored in a database, the suitable prediction equation is applied to the measurement data and the Kriging interpolation method is used to develop a yield map for the entire field.

Results and discussion

The first step of the geoprocessing delivers as a result a yield prediction value for the measurement points. For this purpose, the suitable equation is selected from the database to calculate the prediction from the measured CSH. The decisive selection criteria are the region, which is determined from the GPS position, and the farmer's input on the growing period and sward type. A yield map is then created as an easily interpretable result by applying the Kriging interpolation. Two examples of such a specific yield map are shown in Figure 1. In both maps, the dotted lines, shown in blue or red, represent the measurement points of the RPM taken along a W-pattern.

Considering a balanced sward type in each case, the left map represents the yield prediction for the first growth and the right map the prediction of the second growth. Thus, a different equation is used for the prediction of the left and right yield map respectively.

Comparison of the maps shows that the yield level differs greatly between the first and second growth. However, it is apparent that the areas with relatively high and relatively low yields coincide in both maps. Accordingly, areas with different yield levels can be identified, providing the basis for site-specific management.

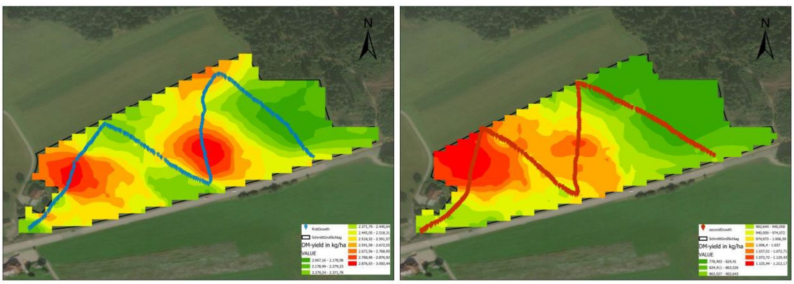

Figure 1. Calculated yield maps of one test site in the Black Forest region for different growing conditions (A: first growing period, 10 June 2021; B: second growing period, 16 July 2021).

Conclusions

On-farm research can be used to develop region- and sward-specific prediction equations for predicting DM yield from measurements of compressed sward height. These equations can be stored in a database which is linked to semi-automated geoprocessing. Only by using this geoprocessing it is possible to apply the equation, which is adjusted to the growing conditions of each specific farm, and this leads to an increase in the accuracy of yield prediction compared to the default prediction equation for the rising plate meter. The results are provided to the farmer in an easily accessible form as a yield map. These advantages improve the acceptance of the sensor technology and enable wider application by farmers. The application can be continuously supplemented, disseminated and adapted. As a next step, equations described in literature for other regions will be integrated into the database. This means that additional grass height measurements for these regions are not necessary. In this way, the geoprocessing contributes to the transfer of sensor technologies into practical farming situations.

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References

Bogue R. (2017) Sensors key to advances in precision agriculture. *Sensor Review* 37, 1-6.

Cárdenas J., Balocchi O. and Calvache I. (2020) Calibration of the rising plate meter for mixed pastures of ryegrass (*Lolium perenne* L.) and kikuyo (*Cenchrus clandestinus*). *Chilean Journal of Agricultural and Animal Sciences* 36, 216-223.

Cho W., Wade Brorsen B., Biermacher J. and Rogers J.K. (2019) Rising plate meter calibrations for forage mass of wheat and rye. *Agricultural and Environmental Letters* 4, 1-4.

Dillard S.L., Hafla A.N. Rubano M.D., Stout R.C., Brito A.F. and Soder K.J. (2016) Evaluation of a rising plate meter for use in multispecies swards. *Agricultural & Environmental Letters* 1, 1-4.

Hart L., Werner J., Velasco E., Perdana-Decker S., Weber J., Dickhöfer U. and Umstätter C. (2020) Reliable biomass estimates of multispecies grassland using rising plate meter. *Grassland Science in Europe* 25, 641-643.

McSweeney D., Coughlan N.E., Cuthbert R.N., Halton P. and Ivanov S. (2019) Micro-sonic sensor technology enables enhanced grass height measurement by rising plate meter. *Information Processing in Agriculture* 6, 279-284.

Nussbaum H., Weißbach F., Elsäßer M., Schenkel H., Staudacher W., von Borstel U., ... and Rieder J.B. (2004) *Grobfutterbewertung*. Deutsche Landwirtschafts-Gesellschaft e.V. (ed.), Frankfurt, Germany, p. 4.

O'Brien B., Murphy D., Askari M.S., Burke R., Magee A., Umstätter C. ... and McCarthy T. (2019) Modelling precision grass measurements for a web-based decision platform to aid grassland management. *Precision Livestock Farming* 9, 858-863.

Rayburn E. (2020) Plate meter calibrations for forage mass follow a continuum of sward basal density. *Crop, Forage and Turfgrass Management* 6, 1-11.

Rayburn E. and Rayburn S. (1998) A standardized plate meter for estimating pasture mass in on-farm research trials. *Agronomy Journal* 90, 238-241.

Voigtländer G. and Voss N. (1979) *Methoden der Grünlanduntersuchung und -bewertung*. E. Ulmer, Stuttgart, Germany, 207 pp.

The role of grasslands in PATHWAYS

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Abstract

With the aim of reducing environmental impacts while addressing societal demands for safe, nutritious, and affordable meat and dairy products, PATHWAYS is about identifying and increasing sustainable practices along the supply and production chains of the European livestock sector. Coordinated by the Swedish University of Agricultural Sciences and comprising 28 partners from 12 countries, this 5-year (2021-2026) €9 million Horizon 2020 project contributes to the EU Farm-to-Fork Strategy, which is at the heart of the EU Green Deal. Grasslands have an important role in PATHWAYS, since grasslands play a key role in transitions to improved sustainability in ruminant production systems. PATHWAYS will, among others, define and assess interactions between livestock production, the environment and land-use by incorporating technical and economic indicators into a framework to assess ecosystem services. This framework will enable a quantitative assessment of ecosystem services that grasslands provide. The indicators will be established in a co-innovative process by stakeholder consultation and measured by collecting data in grassland related practice hubs of farmers. Development pathways will be identified that meet multifaceted societal demands, both currently and in the future.

Keywords: livestock sector, pathways, stakeholder consultation, sustainability

Introduction

Worldwide demand for animal products is predicted to double over the next decades due to population growth and increasing economic prosperity (Godfray *et al.*, 2018). This may lead to further intensification of production which in turn may put pressure on available resources like land or water, and lead to higher greenhouse gas emissions and other environmental impacts (Van Zanten *et al.*, 2018; Willett *et al.*, 2019). Furthermore, there is an increasing concern about the negative impacts of intensive production on animal welfare in livestock farming (Godfray *et al.*, 2018). At the same time, livestock farming plays a vital role in food and nutrition security by providing nutrient-rich food, whilst contributing to efficient agriculture and the vitality of rural territories (Mehrabi *et al.*, 2020). Also, livestock systems can recycle biomass and help to close nutrient cycles at farm and territorial levels. The lack of a holistic sustainability assessment approach makes it difficult to measure livestock's contribution to society, hampering evidence-based debates about trade-offs and leading policymakers to focus on 'highly tangible, but essentially weak, leverage points' (Abson *et al.*, 2017; Scown *et al.*, 2019). PATHWAYS aims to reduce environmental impacts while addressing societal demands for safe, nutritious, and affordable meat and dairy products. The aim of this paper is to provide insight into the role of grasslands in PATHWAYS.

Materials and methods

PATHWAYS is coordinated by the Swedish University of Agricultural Sciences (SLU) and comprises 28 partners from 12 countries (Sweden, Denmark, United Kingdom, the Netherlands, Belgium, Germany, Poland, Switzerland, Romania, Italy, France, and Spain). It started in September 2021 and will end in August 2026. This €9 million Horizon 2020 project contributes to the EU Farm-to-Fork Strategy, which is at the heart of the EU Green Deal. PATHWAYS focuses on dairy, beef, pork, and poultry.

Within PATHWAYS, we will address the challenges of the livestock sector through the development of a multi-dimensional assessment and a holistic scenario evaluation to improve the overall sustainability of terrestrial livestock production systems in Europe. At the heart of PATHWAYS is a reflective learning approach through stakeholder involvement at multiple levels, ensuring actors within livestock value chains buy into the project outcomes through ownership of the process and the results. We will ensure this strong multi-actor approach via a European multi-actor platform (international group of supply chain actors), national practice hubs (national groups of farmers) and a community of practice (forum involving several hundred international and national stakeholders from different value chains).

PATHWAYS will define and assess interactions between livestock production, the environment and land-use by incorporating technical and economic indicators into a framework to assess ecosystem services. The importance of the different ecosystem services will be established in a co-innovative process by stakeholder consultation. The indicators will be measured by collecting data in practice hubs of farmers supporting holistic evaluation. An LCA farm system model (Schader *et al.*, 2014) will be updated by integrating critical farm-system flows as well as recently updated IPCC methodologies. We will use the project outcomes to identify development pathways that meet multifaceted societal demands, both currently and in the future (Figure 1).

The role of grasslands in PATHWAYS

In ruminant production systems, grasslands play a key role in transitions to improved sustainability (Huyghe *et al.*, 2014). Worldwide, grasslands provide many ecosystem services, of which carbon sequestration and forage production are mentioned most frequently (Zaho *et al.*, 2020). Previous research in Europe showed that the different functions of grasslands are highly recognized and appreciated by all relevant stakeholder groups (Van den Pol-van Dasselaar *et al.*, 2014). The large European grassland area appears to be essential for economy, environment, and people. There are, however, complex interactions between livestock production, the environment and land-use in different socio-ecological contexts, which obstruct progress. The PATHWAYS framework to assess ecosystem services will, among others, enable a quantitative assessment of the value of ecosystem services that grasslands provide. Technical and economic indicators for grasslands will be measured by collecting data in practice hubs of farmers. Practice hubs that focus on grasslands used by livestock are in Denmark, France, Italy, Romania, Spain, Sweden, and the United Kingdom. Some of these practice hubs, both for dairy systems and beef systems, aim for 100% pasture-fed. In addition, specific attention will be paid to harmonisation of methods for

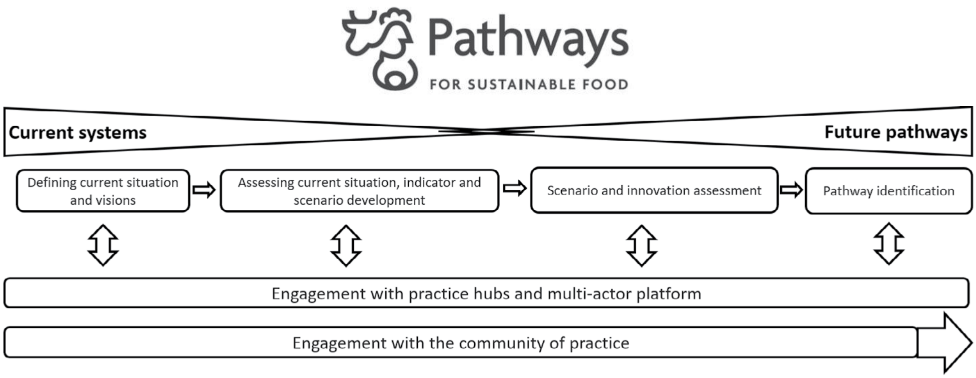

Figure 1. Project overview. PATHWAYS will bring together researchers and industry to develop visions and holistic scenario evaluations based on new indicators and methods. This will inform the selection of pathways for sustainable development of livestock systems and foster the development of future innovations in policy, business and research through engagement with a community of practice.

soil C sequestration estimates in Life Cycle Analyses through a systematic review and expert consultation process (Goglio *et al.*, 2015). The evaluation of practice hubs will include the potential for C sequestration in grass systems where a change has been made from lower to higher productivity and the converse (i.e. soil C loss if reducing productivity and not increasing root biomass and aboveground residue).

Conclusions

The sections of PATHWAYS that will focus on production systems of ruminants like dairy, beef, sheep, and goats will provide ample opportunities in the years 2021-2026 for the transfer and co-construction of innovations in grasslands for improved sustainability, thus emphasising the key role of grasslands in these systems.

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References

Abson D.J., Fischer J., Leventon, J., Newig J., Schomerus T. Vilsmaier U., von Wehrden H., Abernethy P., Ives C.D., Jager N.W. and Lang D.J. (2017) Leverage points for sustainability transformation. *Ambio* 46, 30-39.

Godfray H.C.J., Aveyard P., Garnett T., Hall J.W., Key T.J., Lorimer J., Pierrehumbert R.T., Scarborough P., Springmann M. and Jebb S.A. (2018) Meat consumption, health, and the environment. *Science* 361(6399). https://doi.org/10.1126/science.aam5324

Goglio P., Smith W.N., Grant B.B., Desjardins R.L., McConkey B.G., Campbell C.A. and Nemecek T. (2015) Accounting for soil carbon changes in agricultural life cycle assessment (LCA): a review. *Journal of Cleaner Production* 104, 23-39.

Huyghe C., De Vliegher A., van Gils B. and Peeters A. (2014) *Grasslands and Herbivore Production in Europe and Effects of Common Policies*. Quae Editor, Versailles, France, 320 pp.

Mehrabi Z., Gill M., van Wijk M., Herrero M. and Ramankutty N. (2020) Livestock policy for sustainable development. *Nature Food* 1, 160-165.

Scown M.W., Winkler K.J. and Nicholas K.A. (2019) Aligning research with policy and practice for sustainable agricultural land systems in Europe. *Proceedings of the National Academy of Sciences* 116(11), 4911-4916.

Schader C. Jud K., Meier M.S., Kuhn T., Oehn B. and Gattinger A (2014) Quantification of the effectiveness of greenhouse gas mitigation measures in Swiss organic milk production using a life cycle assessment approach. *Journal of Cleaner Production* 73, 227-235.

Van den Pol-van Dasselaar A., Goliński P., Hennessy D., Huyghe C., Parente G. and Peyraud J.L. (2014) Évaluation des fonctions des prairies par les acteurs européens. *Fourrages* 218, 141-146.

Van Zanten H.H.E., Herrero M., Van Hal O., Röös E., Muller A., Garnett T., Gerber P.J., Schader C. and De Boer I.J.M. (2018) Defining a land boundary for sustainable livestock consumption. *Global Change Biology* 24(9), 4185-4194.

Willett W., Rockström J., Loken B., Springmann M., Lang T., Vermeulen S., Garnett T., Tilman D., DeClerck, and Murray C.J.L. (2019) Food in the Anthropocene: the EAT Lancet Commission on healthy diets from sustainable food systems. *The Lancet* 393(10170), 447-492.

Zhao Y., Liu Z. and Wu J. (2020) Grassland ecosystem services: a systematic review of research advances and future directions. *Landscape Ecology* 35, 793-814.

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