



Soil tillage drives multifunctionality in the onset of a diversified cropping system

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ABSTRACT

Intensified agricultural production with cereal-dominated crop rotations and high fertiliser and pesticide inputs, often combined with inversion tillage, degrades cropping system multifunctionality. Cropping system diversification via integration of service crops, increasing the functional diversity of crops in the rotation as well as reduced tillage has been suggested to enhance agroecosystem multifunctionality. However, evidence is predominantly based on assessment of single practices and single agroecosystem functions, leaving their combined effects on cropping system multifunctionality poorly understood. Here, we assess cropping system multifunctionality in a Swedish cropping system diversification trial based on 15 indicators of soil physical health, nutrient capture and cycling, natural weed and pest regulation, and macronutrient output collected during the first three years under contrasting diversification strategies (crop rotation diversification, service crop use, and tillage reduction). Using a combined index of diversification practices, we found that high diversification was associated with compacted soils, more soil detritivores and fewer crop pests, likely mainly driven by the implementation of tillage reduction. The combined index masked many practice-related effects: tillage reduction was most influential for cropping system multifunctionality, followed by increased crop rotation diversity, whereas the inclusion of service crops did not affect any indicators. Several indicators were interrelated and affected by the same diversification practice, highlighting the complex dependencies in agroecosystems. Despite multiple benefits, we also identified diversification-related trade-offs: tillage reduction benefited pest regulation but increased soil compaction and hampered weed regulation, while increased crop rotation diversity reduced nitrogen inputs but also lowered weed regulation and metabolisable energy output. Our results emphasise that cropping system diversification is a promising pathway to sustainable agriculture yielding various benefits for cropping system multifunctionality when trade-offs are managed.

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1. Introduction

Cropping systems in temperate regions often rely heavily on mineral fertilisers and pesticides, enabling cereal-dominated crop rotations (Popp et al., 2013). Although such systems are designed to maximise short-term economic sustainability, their ability to ensure long-term ecological, social and economic sustainability is questionable. For example, nitrogen losses from agricultural land substantially pollute waterbodies and the atmosphere and contribute to climate change (Schulte-Uebbing et al., 2022). Intensive agricultural management further drives soil degradation and threatens soil fertility (European Environment Agency, 2024). Observed negative impacts on biodiversity, related functioning, the environment and human health have led to restrictions or bans of many pesticides, e.g., neonicotinoid insecticides (Sánchez-Bayo, 2014; Rundlöf et al., 2015; Frank, 2024), and few new pesticides are entering the market (Duke, 2012). Cereal agriculture is dependent on the non-selective herbicide glyphosate (Wiese and Steinmann, 2020), which faces potential regulatory bans (Finger et al., 2023) and increasing problems with rapidly evolving herbicide resistance (Baek et al., 2021). Therefore, it is imperative to develop glyphosate-free cropping systems that function without increasing the use of other environmentally harmful herbicides (Neve et al., 2024).

Cropping system diversification through increased crop and plant diversity and conservation of soil and water resources (Rasmussen et al., 2024) is increasingly demonstrated as a promising strategy for substituting agrochemical inputs with enhanced ecosystem functioning (Tamburini et al., 2020; Bommarco, 2024). Common in-field diversification practices include service cropping with cover- and intercrops (Wittwer et al., 2017; Boetzel et al., 2023), extending and diversifying crop rotations (Shah et al., 2021), and reduced soil disturbance via tillage reduction (Roger-Estrade et al., 2010; Rosa-Schleich et al., 2024). Many of these diversification practices can benefit biodiversity and related functions such as water retention or climate regulation while maintaining crop yields (Tamburini et al., 2020).

Most of the evidence for the benefits of diversification is, however, derived from studies of single practices targeting single agroecosystem functions, e.g., pest or weed regulation, nutrient cycling, or soil organic carbon accumulation (Tamburini et al., 2020), and there is an emerging interest in exploring multifunctional outcomes of diversification practices (e.g. Wittwer et al. 2021). Our current understanding of multifunctional outcomes of diversification practices is limited. The drivers of ecosystem functions are interconnected (Jiao et al., 2022), and single organisms can contribute to several functions (Dunn et al., 2020; Boetzel and Knapp, 2024). Farming practices targeting specific outcomes change the biophysical environment in crop fields and are likely to affect multiple organisms and functions, likely generating both synergies and trade-offs in ecosystem functioning. For instance, reducing herbicide use can benefit aboveground biodiversity but may necessitate intensified tillage to manage weeds, which negatively affects soil biodiversity and depletes soil organic carbon (Betancur-Corredor et al., 2022). Similarly, cover or service crops often promote soil biodiversity, weed suppression and subsequent crop yields, but can reduce yields of the main crops or increase nitrous oxide emissions depending on local soil and climatic conditions, crop management and service crop species and timing (Abdalla et al., 2019; Peng et al., 2024; Yousefi et al., 2024).

Multifunctionality approaches allow a simultaneous examination of multiple ecosystem functions and their interactions, facilitating the identification of synergies and trade-offs (Byrnes et al., 2014; Manning et al., 2018). Studies on multifunctionality commonly rely on averaged composite indices, either as responses or as predictors (see e.g. Wittwer et al. 2021), potentially camouflaging opposing effects of practices or treatments and leading to a loss in management-relevant information (Byrnes et al., 2014). Identifying key practices with a disproportionate impact is essential, particularly where possibilities to implement multiple diversification practices are limited. For example, crop rotation diversity contributed less to the multifunctionality of European cereal

rotations than the total crop cover duration of the rotation (Garland et al., 2021). Conversely, focussing solely on the effects of singular practices obscures potential combined effects on cropping system functionality, including synergies and trade-offs, that might not be apparent when analysed separately (see e.g. He et al. 2023). Contrasting single and combined effects of diversification practices is thus necessary for the holistic and nuanced understanding of the dependencies of cropping system functionality needed to redesign sustainable cropping systems.

Here, we explore the single and combined effects of three common diversification practices, increased crop rotation diversity, the inclusion of service crops, and tillage reduction, on 15 indicators of cropping system multifunctionality related to soil physical health, nutrient capture and cycling, natural weed regulation, natural pest regulation and macronutrient output in a glyphosate-free system. We hypothesised that the inclusion of diversification practices, alone or in combination, increases functionality, but that trade-offs between single functions would occur due to soil compaction resulting from reduced tillage depth harming indicators related to soil biodiversity. In a field experiment, we assessed (i) how ecosystem functions are interrelated and (ii) how increasing diversification and single management practices of increased crop rotation diversity, inclusion of service crops and shallow soil tillage affected indicators for ecosystem multifunctionality.

2. Materials and methods

2.1. Field experiment, crop and soil management

All data was collected from the “EcoServ” long-term field experiment in Uppsala, Sweden (59°50'19.1"N, 17°47'15.8"E), established in 2020 on a silty-clay soil (45% clay, 50% silt, 5% sand) in a humid continental climate (Köppen climate zone Dfb; Beck et al. 2018). Prior to the start of the experiment, the field was characterised by a region-typical cereal-based crop rotation with regular inversion tillage to a depth of approximately 25 cm. The experiment aims to investigate agroecosystem functionality of glyphosate-free crop production systems in relation to (i) crop rotation diversity, (ii) tillage reduction and (iii) the inclusion of service crops.

The experiment comprises a randomised and fully phased design with 28 independent plots (24 m × 36 m), each divided into two sub-plots (12 m × 36 m). Crop rotation (simple or diversified) and soil tillage (shallow or inversion) treatments are implemented at the plot level, whereas service crops (absent or present) are implemented at the sub-plot level in a split-plot design (Fig. 1). The homogeneous soil conditions at the experimental site did not necessitate the establishment of blocks in the experiment.

The two implemented crop rotations vary in crop functional diversity: (i) a simple two-year cereal rotation of alternating winter wheat (*Triticum aestivum*; 4 plots) and spring barley (*Hordeum vulgare*; 4 plots) and (ii) a diversified five-year rotation of spring barley (4 plots), winter wheat (8 plots as the crop occurs twice in the rotation), spring oilseed rape (*Brassica napus*; 4 plots), winter wheat and field beans (*Vicia faba*; 4 plots; Fig. S1). Each crop in each rotation is replicated in four plots in each year. Crop residues remain in the field throughout all rotations and crops. For this study, we analysed the first three years of the experiment resembling different three-year crop rotations with varying realised crop rotation diversity (Fig. S1).

Half of the plots in each rotation are managed with annual inversion tillage to a depth of about 25 cm, with no restrictions on the selection of tillage tools for seedbed preparation and crop residue management while the other half is managed with reduced, shallow tillage. Here, shallow, non-inversion tillage is considered as diversified soil tillage practice compared to conventional inversion tillage, as shallow tillage reduces soil disturbance. Plots with shallow tillage experienced a maximum soil disturbance depth of 3–5 cm (CrossCutter Disc, Väderstad AB, Sweden). While no till would have caused even less soil disturbance,

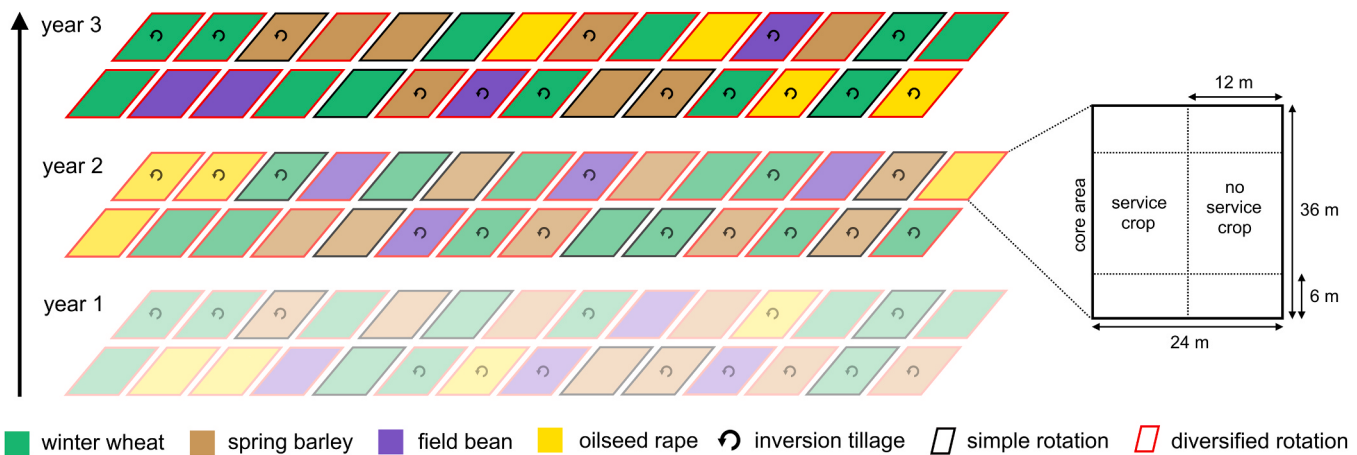


Fig. 1. Schematic of the field experiment with 28 plots subdivided in 56 subplots and the crops grown across the three first seasons which were included in this study. Different edge colours indicate the two crop rotations; different colour fills indicate different crops; arrows indicate plots that received inversion tillage.

shallow tillage had to be implemented for weed control purposes, representing a minimum-tillage system without use of non-selective herbicides. Crops were seeded in both soil tillage treatments with a standard single disc seed drill (Rapid, Väderstad AB, Sweden).

At the start of the experiment, one of the subplots within each plot was randomly assigned the inclusion of service crops, either sown as post-harvest cover crops or undersown with the main crop. The service crop species varied according to the specific position within the crop rotation and the ecosystem services they are anticipated to deliver for the subsequent main crop (for the actual species, see Fig. S1). The primary anticipated ecosystem services include weed suppression between and during cropping seasons, nitrogen provisioning from leguminous species, prevention of nutrient leaching, and soil erosion mitigation between cropping seasons.

Glyphosate or any alternative non-selective herbicides are not used in any of the plots irrespective of their management. Pesticide-based weed, disease and crop pest management follows an integrated pest and weed management approach and is adapted to pest and weed pressure in the respective subplots.

2.2. Data collection

We assessed 15 indicators for cropping system multifunctionality, linked to soil physical health (soil air capacity, soil penetration resistance, plant-available water; Doran et al. 1996), nutrient capture and cycling (soil N content, N inputs, soil detritivore density), natural weed regulation (weed seed bank density, weed seed bank evenness, weed seed predation), natural pest regulation (crop pest emergence, crop pest predation, below-ground natural enemies, natural enemy emergence) and macronutrient output (metabolisable energy output, crude protein output; Table 1 & Fig. 2). The indicators were assessed across all plots and subplots, covering all crop types. All measurements were taken in the core area of each subplot (Fig. 1) and at a distance of at least 1 m to the plot edges to mitigate edge effects. For a detailed description of the methodology and data collection, see the Supplementary Note in the Supporting Information.

2.3. Statistical analysis

For all analyses, we accumulated all measured data on the subplot level, resulting in one value per subplot and 56 data points in total. In a first step, we tested for correlations between the 15 indicators (see Fig. 2) of cropping system multifunctionality that could indicate general synergies or trade-offs with Pearson correlations. Then, we tested whether the indicators were affected by a combined index of cropping system diversification (Fig. 3). Lastly, we tested how they were affected

by the interplay of individual diversification practices (Fig. 4).

As the crop rotation diversity at the time of sampling differed between plots with same long-term crop rotation due to different crop types grown in preceding years, we calculated the realised crop rotation diversity at the plot level. Crop rotation diversity across all three years was calculated for each plot as the Shannon diversity of different main crops grown across all three years using the time the respective crop was present in the plot as ‘abundance’ (based on sowing and harvesting dates). In this calculation, we only included days within the vegetative period, which has a defined start at the first day within a period of six days with average daily temperature above 5°C in the calendar year, and a defined end at the last day before a period of six days with average daily temperature below 5°C (after 01 July; The Swedish Meteorological and Hydrological Institute 2025).

To assess how progressive cropping system diversification affected the multifunctionality indicators, we calculated a combined index of cropping system diversification and extensification (henceforth: diversification index) similar to the land-use intensity index proposed by Herzog et al. (2006):

$$\mu_{div} = (\text{soil tillage} + \text{service crops} + \text{crop rotation diversity}) / 3.$$

where shallow soil tillage and the presence of service crops were coded as 1, conventional soil tillage and the absence of service crops as 0, and the crop rotation diversity was standardised between 0 and 1 by dividing the measured value in each subplot (see 2.1) by the maximum value measured across all subplots. While shallow tillage is still more invasive than zero tillage, the latter usually requires increased herbicide inputs (Chantre and Cardona, 2014) which potentially affects many aspects of cropping system multifunctionality and we thus would not consider zero tillage systems as more diversified. In the resulting dimensionless diversification index μ_{div} , 1 indicates a maximally diversified cropping system with service crops and shallow tillage, 0 a minimally diversified system without service crops and with inversion tillage. This index assumed equal weight of all diversification practices. We fitted separate generalised linear mixed effects models (package ‘glmmTMB’, version 1.1.10; Brooks et al. 2017) for each of the 15 indicators of cropping system multifunctionality to determine the effects of cropping system diversification. Models included the fixed effect ‘diversification index’ (continuous) and a random effect of ‘plot’ to account for the nested subplot design. For all responses that were not assessed across the whole 3-year rotation (all indicators except nitrogen inputs, metabolisable energy and crude protein outputs), the crop (factor, 4 levels) grown during the assessment was also included.

Lastly, we fitted generalised linear mixed effects models for each of the 15 indicators of cropping system multifunctionality as described above to determine the effects of individual management decisions. Instead of ‘diversification index’, initial models included the fixed

Table 1

Indicators of cropping system multifunctionality with brief description, assessment timing and references.

indicator	methodology	assessment timing	references
soil physical health			
soil air capacity	undisturbed soil cores	during the growing season in the third year of the rotation	Reynolds et al. (2002)
soil penetration resistance	hand penetrometer	during the growing season in the third year of the rotation	Bengough and Mullins (1990)
plant-available water	undisturbed soil cores	during the growing season in the third year of the rotation	Reynolds et al. (2002); Seybold and Harms (2012)
nutrient capture and cycling			
soil N content	soil cores	spring of the third year of the rotation	Wehrmann and Scharpf (1979)
N inputs	accumulated inputs	annually, across the whole rotation	Cassman et al. (2002)
soil detritivore density	soil cores & Tullgren extraction	autumn of the third year of the rotation	Trägårdh (1933), Maran et al. (2020)
natural weed regulation			
weed seed bank density	soil cores & seed germination	during the growing season in the third year of the rotation	Cardina et al. (2002)
weed seed bank evenness	soil cores & seed germination	during the growing season in the third year of the rotation	Adeux et al. (2019)
weed seed predation	sentinel prey cards with seeds	during the growing season in the third year of the rotation	Daouti et al. (2022)
natural pest regulation			
crop pest emergence	emergence traps	during the growing season in the third year of the rotation	Sunderland et al. (1995)
crop pest predation	sentinel prey cards with aphids	during the growing season in the third year of the rotation	Boetzel et al. (2019)
below-ground natural enemies	soil cores & Tullgren extraction	autumn of the third year of the rotation	Trägårdh (1933), Maran et al. (2020)
natural enemy emergence	emergence traps	during the growing season in the third year of the rotation	Sunderland et al. (1995)
macronutrient output			
metabolisable energy output		annually, across the whole rotation	Vico et al. (2026)
crude protein output		annually, across the whole rotation	Vico et al. (2026)

effects ‘service crops’ (factor, 2 levels), ‘crop rotation diversity’ (continuous; for calculation see 2.1), ‘soil tillage’ (factor, 2 levels) and all two-way interactions between these three predictors. The models were subsequently reduced using likelihood ratio testing (but all single fixed effects and random effects were retained in all models as they are related to our research questions or necessary to account for properties of the study design; see Table S1).

All models for positive, non-integer data used a gamma residual distribution with log-link, models for proportional data, i.e. weed seed

and pest predation rates, used a beta regression with logit link (0 or 1 boundary values were absent in the data). We checked all models for under- and overdispersion, and suitability of chosen residual distributions using the DHARMA package (version: 0.4.7; Hartig 2025) and detected no violation of the model assumptions. Model outputs were obtained using type III sums of squares Wald chi-square tests with the command ‘Anova’ (library ‘car’, version 3.1–3; Fox and Weisberg 2019) and R^2 values with the command ‘performance’ (library ‘performance’, version 0.12.4; Lüdtke et al. 2021).

3. Results

3.1. Indicators of cropping system multifunctionality are interconnected

We found 16 positive and twelve negative correlations between indicators of cropping system multifunctionality (out of 105 possible correlations). Positive correlations were detected between soil air capacity, soil mineral N content and crop pest emergence; soil penetration resistance and soil decomposer density, weed seed bank density and below-ground natural enemy density; N inputs and soil mineral N content, weed seed bank evenness and metabolisable energy output; soil decomposer density and below-ground natural enemy density and natural enemy emergence; weed seed bank density and below-ground natural enemy density and weed seed predation; weed seed bank evenness and natural enemy emergence and metabolisable energy output; natural enemy emergence and metabolisable energy output; crude protein output and metabolisable energy output (Fig. 2). We found negative correlations between air capacity and penetration resistance, plant-available water and natural enemy emergence; penetration resistance and weed seed bank evenness and crude protein output; soil decomposer density and crude protein output; weed seed bank density and weed seed bank evenness, natural enemy emergence and metabolisable energy output; weed seed bank evenness and weed seed predation and below-ground natural enemy density; below-ground natural enemy density and crop pest emergence (Fig. 2).

3.2. Effects of diversification on cropping system multifunctionality

Four of the 15 tested indicators were related with the combined diversification index. An increasing diversification index was negatively associated with air capacity and positively related with penetration resistance, i.e. soils were more compact with increasing diversification index (Fig. 3 A, B, C; Table S2). The density of soil detritivores was positively related with the diversification index, while the density of emerging crop pests was negatively related (Fig. 3 A, D, E; Table S2). In addition, weed seed bank evenness tended to decrease with increasing diversification index ($p = 0.067$; Fig. 3 A; Table S2).

3.3. Diversification practices affect cropping system multifunctionality

Assessing the effects of single management practices, we found soil tillage to be most influential for indicators of cropping system multifunctionality, followed by crop rotation diversity, while the presence of service crops rarely affected indicators. Air capacity and penetration resistance were on average 46.7% lower and 54.9% higher, respectively, under shallow tillage compared to inversion tillage (Fig. 4; Table S3). Soil N content and soil detritivore density were on average 2.5% and 15.4% higher under shallow tillage compared to inversion tillage (Fig. 4; Table S3). The density of below-ground natural enemies was 17.3% higher and crop pest emergence was 47.0% lower under shallow tillage compared to inversion tillage (Fig. 4; Table S3). Weed seed bank density was 5.9% higher and weed seed bank evenness 45.0% lower under shallow tillage compared to inversion tillage (Fig. 4; Table S3).

The effects of soil tillage on soil penetration resistance were modulated by the presence of service crops and by crop rotation diversity. When service crops were present, soil penetration resistance was 41.5%

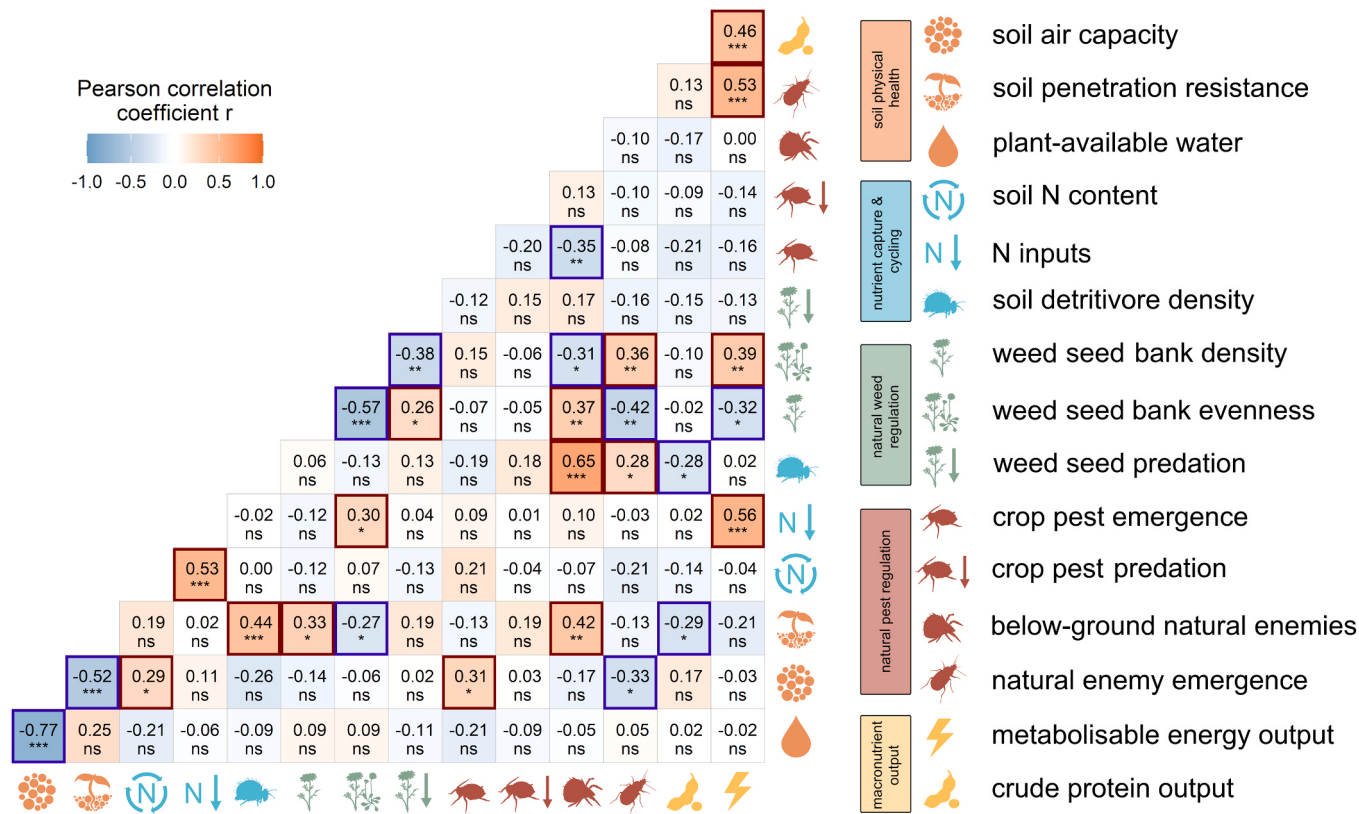


Fig. 2. Pearson correlations between the 15 indicators for cropping system multifunctionality to uncover synergies and trade-offs. Icon colours relate to the five categories of cropping system functionality. Bold panel borders indicate statistically significant correlations ($p < 0.05$).

higher under shallow tillage while it was 64.9% higher when service crops were absent (Table S3). Soil penetration resistance was positively related to crop rotation diversity only in plots with inversion tillage (Table S3).

Crop rotation diversity was negatively related to soil N content and N input, with decreases of 2.6% and 6.2%, respectively, for each 0.1 increase in crop rotation diversity (Fig. 4; Table S3). For N input, the effect of crop rotation diversity was modulated by the presence of service crops with a statistically significant decrease when service crops were present in the rotation while this relationship only tended to be significant ($p = 0.071$) when service crops were absent (Table S3). Increasing crop rotation diversity reduced natural weed regulation, increasing weed seed bank density by 10.7% and decreasing weed seed bank evenness by 2.9% for each 0.1 increase in crop rotation diversity (Fig. 4; Table S3). Lastly, metabolisable energy output was negatively related to crop rotation diversity, decreasing by 3.6% for each 0.1 increase in crop rotation diversity (Fig. 4; Table S3).

4. Discussion

Our assessment of single and combined effects of service crops, diversified crop rotations, and shallow tillage on agroecosystem multifunctionality revealed both synergies and trade-offs among indicators of agroecosystem functioning. Cropping system diversification improved some, but not all biodiversity-related indicators and individual practices simultaneously affected several indicators, emphasising potential off-target effects by changes in management. Using a combined index of diversification provided little additional information and masked practice-specific effects. Through our holistic and multifaceted approach, differentiating between diversification *per se* and individual diversification practices on 15 indicators of multifunctionality, we identified several benefits and some trade-offs from diversification, showing overall that diversification is a promising pathway to achieve

multifunctional cropping systems when trade-offs are managed.

4.1. Limited effects of the combined diversification index

Using a combined diversification index we found relatively few effects of cropping system diversification compared with similar research (e.g., He et al. 2023, Tamburini et al. 2020). Increasing cropping system diversification was associated with decreased soil air capacity, and increased soil penetration resistance and soil detritivores density, likely driven by reduced soil disturbance under shallow tillage at higher diversification index values (Betancur-Corredor et al., 2022). In addition, crop pest emergence decreased with increasing cropping system diversification, indicating enhanced pest regulation. This effect may reflect higher densities of below-ground natural enemies under shallow tillage that was, however, only visible when diversification practices were analysed separately and not detected in analyses of the composite diversification index.

Overall, the combined diversification index revealed fewer effects of cropping system diversification on cropping system multifunctionality than analyses of individual practices, and no synergies emerged. This could be due to the equal weighting of diversification practices in the composite index that allows practices with disproportionate effects to mask the effects of other practices or even overshadow opposing effects of other practices. For example, soil mineral nitrogen content in spring increased under shallow tillage and decreased with increasing crop rotation diversity, resulting in no net effect of the diversification index. Notably, indicators explained by the diversification index, such as soil detritivore density, were similarly predicted when analysing individual diversification practices separately. To refine the diversification index, for example through weighing individual practices according to their importance on indicators, further assessments of individual practices and the suitability of indicators remain crucial. Conclusions drawn from combined indices, therefore, lack the necessary nuance to inform

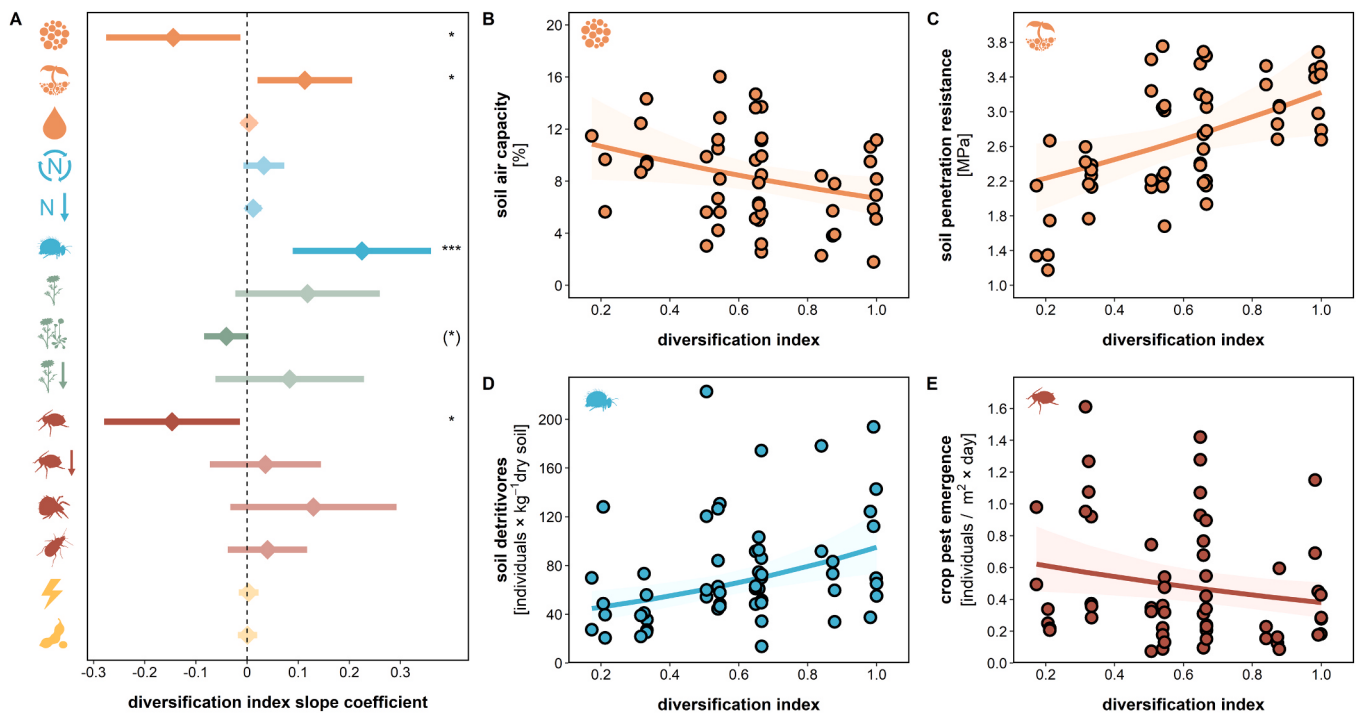


Fig. 3. (A) Slope coefficients for the diversification index for the 15 indicators of cropping system multifunctionality (see Fig. 2 for figurine legend). Standardised slope coefficients are scaled to magnitudes of the standard deviation of the diversification index (z scaling) and can be interpreted as changes in the response on the log scale per 1 standard deviation of the diversification index (SD = 0.247). Slope coefficients that do not overlap with the dashed line represent statistically significant effects ($p < 0.05$). A coefficient estimate to the right of the dashed line indicates a positive relationship, a coefficient estimate to the left of the dashed line a negative relationship with the indicator. (B-E) Statistically significant relationships ($p < 0.05$). Solid lines represent estimated marginal mean predictions with 95% confidence interval (shaded area), points represent data from the individual subplots (see Fig. 1). Icon colours relate to the five categories of functionality (see Fig. 2). For model specifications, see text. For statistics, see Table S2. (*) indicates $p < 0.1$; * indicates $p < 0.05$; ** indicates $p < 0.01$; *** indicates $p < 0.001$.

decision making on the implementation of specific management strategies.

4.2. Synergies and trade-offs of diversification practices

Among the diversification practices, soil tillage reduction had the strongest effects on ecosystem functions, affecting eight indicators of multifunctionality, followed by increased crop rotation diversity affecting five, while the inclusion of service crops affected none. Some responses to shallow tillage and increased crop rotation diversity were consistent, but others diverged in direction. Importantly, no single practice resulted in desirable outcomes for all affected indicators, emphasising the need to identify combinations of diversification practices that enhance overall benefits while minimising trade-offs.

A switch from inversion to shallow tillage decreased soil air capacity and increased soil penetration resistance, as expected due to the lack of mechanical soil loosening (Or et al., 2021). It further increased soil mineral nitrogen content, possibly due to inversion tillage moving nitrogen-rich crop and weed residues into deeper soil layers, or due to reduced nitrogen use by preceding crops under shallow tillage (Brennan et al., 2014). Shallow tillage increased the density but reduced the evenness of weed seed banks, driven by the dominance of two species, *Tripleurospermum inodorum* and *Stellaria media* (data not shown). Both species prefer high N availability and are adapted to regular soil disturbances (Tyler et al., 2021; Hickman et al., 2025), potentially indicating a legacy effect of the recent shift to shallow tillage in the experiment. Given that weed management is a major challenge under reduced herbicide use and tillage intensity (Armengot et al., 2015), the observed weed assemblage shift should be weighed against concurrent benefits for other indicators. In our study, we make no comparisons with no-till systems, as most no-till systems rely on herbicides for weed management and are therefore unlikely to be widely adopted under the

scenario of a glyphosate ban. Shallow tillage increased densities of soil detritivores and below-ground natural enemies, and reduced crop pest emergence. Reducing tillage intensity can benefit below-ground mesofauna (Betancur-Corredor et al., 2022), and we show that positive effects persist also under multiple diversification strategies. As soil detritivores are a key prey for soil-dwelling and overwintering natural enemies in soil food webs (Riggi and Bommarco, 2019; Heinen et al., 2023), their increase may foster crop pest regulation in the soil (Jowett et al., 2021). Detected negative correlations between crop pest emergence and natural enemy emergence support this. By quantifying crop pests emergence, our approach links crop field management more directly to pest pressures than many previous observational assessments (Rowen et al., 2020).

Compared with soil tillage, increased crop rotation diversity affected fewer indicators of cropping system multifunctionality, though it increased weed seed bank density, and reduced soil mineral nitrogen content, nitrogen input, weed seed bank evenness, and metabolisable energy output. Its more limited effects compared with soil tillage could be a result of including only annual crops with very similar biomass accumulation in the rotations. Increasing vegetation permanence via the inclusion of perennials in the rotation is likely to affect a greater number of indicators (Garland et al., 2021; Heinen et al., 2023; El Khosht et al., 2025). The undesired outcomes for weed seed banks were unexpected, as diversifying crop rotations often reduce weed density regardless of tillage regime (Weisberger et al. 2019, but see Benaragama et al. 2016). This could, however, be explained by the inclusion of crops where weeds are more difficult to control than in cereals (legumes and spring oilseed rape) and by the short duration of the experiment. Repeated measures throughout the crop rotation will be an important next step to validate the value of increased crop rotation diversity for indicators of multifunctionality across the full rotation. The reduced metabolisable energy output under increased crop rotation diversity partly contrasts previous

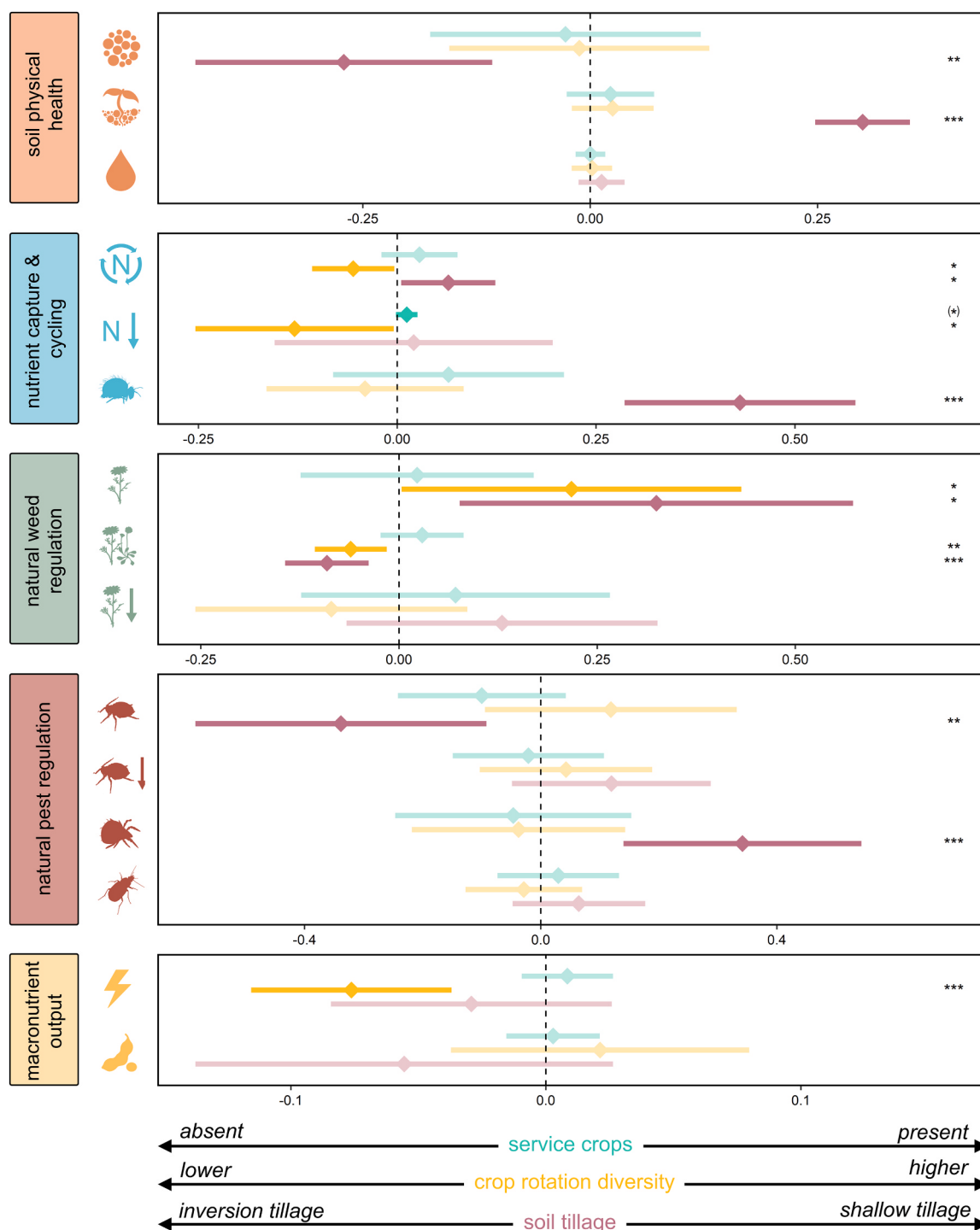


Fig. 4. Model coefficients for service crop presence, crop rotation diversity and soil tillage for the 15 indicators of cropping system multifunctionality (see Fig. 2 for figurine legend). Coefficient estimates that do not overlap with the dashed line represent statistically significant effects ($p < 0.05$). For the categorical predictors service crops and soil tillage, the direction indicates the level where the indicator is higher, for the continuous predictor crop rotation diversity, a coefficient estimate to the right of the dashed line indicates a positive relationship, a coefficient estimate to the left of the dashed line a negative relationship with the indicator. Crop rotation diversity was scaled to magnitudes of its standard deviation (z scaling), thus coefficients can be interpreted as changes in the response on the log scale per 1 standard deviation of crop rotation diversity ($SD = 0.215$). For model specifications, see text and Table S3. (*) indicates $p < 0.1$; * indicates $p < 0.05$; ** indicates $p < 0.01$; *** indicates $p < 0.001$. Colours of the icons relate to the five categories of functionality (see text), for an explanation of the icons, see Fig. 2.

findings of increased cereal yields in diversified crop rotations (Smith et al., 2023) and is caused by the lower caloric-content per unit of yield in non-cereal crops. Diversifying crop rotations is expected to benefit soil mineral nitrogen as non-cereal crops generally deplete stocks less than cereals (Engström and Lindén, 2009). We, however, found the opposite which could have arisen from an overestimation of cereal

nitrogen demand when calculating inputs and from an underestimation of nitrogen-mineralisation. Potential benefits and drawbacks require further testing and should, if confirmed, be carefully considered and communicated to stakeholders when scaling up diversified agriculture.

Service crops did not affect any of the indicators of multifunctionality, despite previously reported benefits for nitrogen retention

and weed suppression (Finney and Kaye, 2017; Finney et al., 2017). Soil tillage can offset benefits of service crops compared with no-till systems (Daryanto et al., 2018), and it is possible that shallow tillage affected service crop functionality. A more likely explanation, however, is that the cold climate and short daylength at the high latitude location (59°) of our experiment limited species selection, establishment and growth and thus lowered effects (Murrell et al., 2017; Heller et al., 2024).

4.3. Dependencies among indicators not linked to diversification practices

Several indicators of cropping system multifunctionality were interrelated without obvious relationships to diversification practices. Natural enemy emergence, though unaffected by individual practices, was negatively correlated with weed seed bank density and positively with weed seed bank evenness. As weed seed bank density and evenness had contrasting responses to shallow soil tillage and were hence negatively correlated, they had contrasting associations with other indicators. Weed seed predation was positively correlated with below-ground natural enemy density and weed seed bank density (and thus negatively with weed seed bank evenness). These relationships indicate enhanced natural weed regulation prior or during natural enemy overwintering at higher natural enemy densities (Daouti et al., 2022). In addition, metabolisable energy output was positively correlated with the density of emerging natural enemies, with cereal-heavy rotations including winter wheat plots having the highest calorie output. Likewise natural enemy emergence was highest in winter wheat plots, that likely provided undisturbed overwintering habitat for natural enemies. Crude protein output was negatively correlated with both soil penetration resistance and soil detritivore densities, likely due to their strong positive correlation, which might indicate soil compaction under shallow tillage affecting root development and nutrient uptake. These dependencies among cropping system functions suggest that diversification practices can render additive, synergistic, or contrasting effects among functions.

4.4. Implications for cropping system diversification

Cropping system diversification has potential to enhance agroecosystem functioning and sustainability but emerging trade-offs need to be managed. We tested the effects of cropping system diversification as well as those of individual diversification practices on 15 indicators of multifunctionality, representing a holistic assessment of agroecosystem functioning. While cropping system diversification affected two thirds of the indicators, none of the biodiversity-related indicators, including important ecosystem services such as natural weed or pest regulation, were affected, highlighting the necessity to refine cropping system diversification. Interdependencies among some indicators of cropping system functioning imply that diversification practices can have additive, synergistic, or contrasting effects. Identifying these directions and effect sizes is key to minimise trade-offs and designing sustainable cropping systems requires compromises. While our analysis demonstrates that a combined index captures overall trends, the nuance on contrasting practice-specific effects vanishes. With effects of different diversification practices, in parts, varying in magnitude and directionality, potentially important consequences for agronomic outcomes are masked. Overall, soil tillage reduction affected cropping system multifunctionality more than increased crop rotation diversity or the inclusion of service crops and responses to soil tillage can outweigh benefits achieved by diversifying other aspects of the cropping system. Given the short duration of the experiment, some effects may have been underestimated: For example, the density of problematic perennial weeds can increase over time under reduced tillage (Armengot et al., 2015), as do yield benefits from diversified crop rotations (Smith et al., 2023). Long-term and replicated studies across different climates and soils are therefore essential to better resolve the magnitude and consistency of the effects of diversification measures on agroecosystem functioning.

CRedit authorship contribution statement

Alexander Menegat: Writing – review & editing, Validation, Supervision, Project administration, Methodology, Investigation, Funding acquisition, Data curation, Conceptualization. **Riccardo Bommarco:** Writing – review & editing, Validation, Supervision, Project administration, Methodology, Funding acquisition, Conceptualization. **Valeria Carini:** Writing – review & editing, Validation, Methodology, Conceptualization. **Cassandra Vogel:** Writing – review & editing, Writing – original draft, Validation, Investigation. **Thomas Keller:** Writing – review & editing, Validation, Funding acquisition. **Göran Bergkvist:** Writing – review & editing, Validation, Funding acquisition. **Ola Lundin:** Writing – review & editing, Validation, Supervision, Methodology, Investigation, Funding acquisition, Conceptualization. **Fabian A. Boetzel:** Writing – review & editing, Writing – original draft, Visualization, Validation, Methodology, Investigation, Formal analysis, Data curation, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agee.2026.110657.

Data availability

Data are available from Zenodo at <https://doi.org/10.5281/zenodo.21508585>

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