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# Poor Quality of Commercial Arbuscular Mycorrhizal Inoculants Used for Agriculture and Home Gardening

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## ABSTRACT

There is an urgent need to develop microbial inoculants that can consistently improve crop performance as part of efforts to implement sustainable agricultural practices and reduce the environmental impact of intensive farming. One of the best known examples of beneficial soil microbes that can promote plant growth and ecosystem performance are arbuscular mycorrhizal fungi (AMF). AMF-based inoculants are increasingly being marketed to enhance key ecosystem functions such as soil nutrient uptake, soil structure, carbon storage and ecosystem health. Despite this potential, the efficacy of commercial AMF products is still poorly documented and highly variable. In this study, we evaluated 16 commercially available AMF inoculants (nine marketed for agricultural use and seven for home gardening) and, for comparison, seven AMF inoculants for research propose, all tested under controlled greenhouse conditions. Our findings revealed that only three commercial AMF products led to root colonisation, and only one promote plant growth. One-third of the agricultural inoculants colonised plant roots, whereas none of the seven commercial home gardening products successfully established a symbiosis with plant roots. In contrast, products intended for research purposes consistently induced AMF colonisation and often resulted in a positive growth response, likely due to higher propagule density. Together with three recent studies analysing worldwide AMF products, our study revealed that 85% of the 64 commercial arbuscular mycorrhizal inoculants tested are of poor quality and did not colonise plant roots. Thus, standardised quality control across the industries is necessary to ensure product effectiveness and promote widespread acceptance by farmers, as well as successfully spreading the use of mycorrhizal inoculants as a viable tool for enhancing sustainable agricultural and gardening practices.

## 1 | Introduction

Although intensive agricultural practices help to meet the growing global demand for food, they come at a significant environmental cost. The extensive use of agrochemicals disrupts soil microbial communities (Edlinger et al. 2022) and lead to greenhouse gas emissions, surface water eutrophication, and loss of biodiversity (Foley et al. 2005; Allan et al. 2015). Furthermore, these practices rely on finite resources, such as phosphorus fertilisers, which raises concerns about the long-term sustainability

of current agricultural practices. To address these challenges, there is an urgent need to develop sustainable agricultural practices that minimise environmental impact while maintaining crop yield and quality (Campbell et al. 2017). One promising approach is to harness ecosystem functions by promoting the ecosystem services provided by beneficial soil organisms and microorganisms (Bender et al. 2016).

Soil microbial communities play a crucial role in maintaining soil fertility, driving nutrient cycling, and ensuring plant health

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(Trivedi et al. 2020). The abundance and functional diversity of the soil microbiome are strongly influenced by agricultural management practices, including crop rotation, reduced tillage, and organic amendments, which selectively promote beneficial microbial groups (Wittwer et al. 2021). In light of the recent ban on many plant protection products and the need to reduce fertiliser use, targeted microbial inoculation has emerged as a sustainable way of introducing or enhancing specific microbial functions in agroecosystems (French et al. 2021).

Of the various beneficial soil microbes, arbuscular mycorrhizal fungi (AMF) are of particular interest due to their ability to form mutualistic symbioses with most agricultural crops (van Der Heijden et al. 2015). AMF are known to improve both plant hydro-mineral nutrition and resilience to biotic and abiotic stresses (Wipf et al. 2019). However, land-use intensification is correlated with a decline in AMF diversity and abundance (Oehl et al. 2003; Verbruggen et al. 2010), largely driven by practices such as high fertilisation rates, intensive tillage, and crop rotations involving non-host species (Jansa et al. 2006; Säle et al. 2015). Experimental evidence suggests that reduced AMF diversity can impair key soil ecosystem services, including nutrient cycling, soil structure and plant stress tolerance and resilience (Wagg et al. 2014). To mitigate these negative effects, AMF inoculation has been proposed as a strategy to support plant nutrition and growth in intensively managed or degraded soils (Köhl et al. 2016). While AMF inoculation has shown promising results in controlled environments, its efficacy under field conditions remains inconsistent and challenging (Faye et al. 2013; Tarbell and Koske 2007; Lutz et al. 2023). This is primarily due to the limited understanding of the factors that influence inoculation success in the field, such as soil characteristics or plant genotypes (Verbruggen et al. 2013; Boussageon et al. 2023), as well as the lack of quality control (Salomon et al. 2022a). Also, the inoculation success showed negative relationship with soil health and plant productivity (Rog et al. 2025).

The global market for arbuscular mycorrhizal inoculants is estimated to have reached \$995 million by 2025, with an estimated compound annual growth rate of 14.8% (Koziol et al. 2025). Despite their potential, the global commercialisation of a wide range of microbial inoculations has lagged behind expectations. This is partly due to diverse regulatory definitions between countries and the absence of mandatory quality control criteria (Salomon et al. 2022a; Koziol et al. 2025).

AMF inoculants includes spores and mycorrhized root fragments as propagules and these are embedded in carrier materials (e.g., zeolite, peat, alginate beads) to ensure protection (Gianinazzi and Vosátka 2004). The first critical step in inoculum production is selecting an appropriate fungal strain. Commercial AMF products often use species from the *Glomus* genus, particularly *Rhizophagus intraradices*, due to its adaptability in various environments (Savary et al. 2018). Factors such as soil type, abiotic stress conditions, and host plant compatibility must be considered when selecting a strain to maximise benefits and establishment (Verbruggen et al. 2013). However, the limited ecological knowledge of many commercial strains often restricts their adaptability to various soil conditions and often it is not reported whether the inoculated AMF established after application. Beyond strain selection, the method of AMF production is equally

important. Commercial AMF propagation relies on techniques such as greenhouse cultivation and in vitro systems (Dalgé and Monreal 2004). Greenhouse-based production involves culturing AMF on substrates such as vermiculite, sand, or expanded clay. These methods are cost-effective and scalable (IJdo et al. 2011). Other methods include nutrient film techniques, aeroponic culture, and in vitro root organ cultures in fermenters (Declerck et al. 1996). The formulation of AMF inoculants is also critical for determining the success of inoculation, which is linked to plant colonisation and physiological benefits (Basiru et al. 2020). Success of AMF inoculants is directly influenced by critical factors such as propagule viability (e.g., whether propagules are actually alive and have the ability to establish a symbiotic relationship with host plant roots), propagule concentration (which ensures sufficient fungal units for colonisation), and the type of carrier material (which provides a suitable environment for propagule survival and delivery). Poor quality control of the products combined with all these factors explains the poor outcomes of inoculants in the market. For example, a study testing 25 mycorrhizal inoculants under greenhouse and field conditions found that 80% of the products either failed to colonise plants or failed to increase biomass (Salomon et al. 2022b). Another study assessed 23 mycorrhizal inoculants and revealed limited viability of AM fungi, pathogen contamination, and negative microbial effect on crop growth (Koziol et al. 2024). These unpredictable results affect consumer confidence and hinder widespread adoption (Salomon et al. 2022a). Thus, given the growing interest in AMF inoculants, there is a need for an independent, thorough evaluation of the reliability and efficacy of commercially available products. So far few studies compared many different inoculum (e.g., Salomon et al. 2022a; Koziol et al. 2024). Moreover, these earlier studies testing the quality of inoculants, did not assess whether the inoculum quality varies among product categories (e.g., inoculants for agricultural use, for home gardening or produced for research purposes might differ in quality). It is important to test this as these inocula can differ in inoculum traits (e.g., carrier material or spore density) and for sales and consumer trust.

In the present study, we evaluated the efficacy of 16 commercially available mycorrhizal inoculants (nine marketed for agricultural use and seven for home gardening) and, in parallel, seven mycorrhizal products formulated for research purposes. Specifically, we compared three product categories: inoculations for agricultural use, home gardening and scientific research, to assess their ability to promote plant growth and colonising plant roots, using molecular and microscopic tools. This focus has not been discussed yet, while previous studies primarily focused on agricultural products, highlighting variable efficacy and quality issues, the home gardening segment has so far been overlooked, despite its rapid expansion and importance to consumers. By including this category alongside agricultural and research formulations, our study provides a more comprehensive picture of the current market landscape in different usage contexts.

## 2 | Materials and Methods

The study evaluated a total of 23 commercial arbuscular mycorrhizal fungi (AMF) inoculants available on the European market (16 products), as well as seven products intended for research purposes. All products were commercially sourced and

deidentified for this study, with labels ranging from B to X. Product A corresponded to a non-inoculated negative control. The products were classified into three categories according to their intended use: agricultural, home gardening and research.

## 2.1 | AMF Inocula Selection

To be included in this study, inoculants had to contain at least one AMF isolate and be labelled for use in commercial agriculture or home gardening, or for research purposes. Sixteen different inoculants were purchased from the European market using online search engines and knowledge of commercial producers. Of these, 12 were labelled to also contain additional plant growth-promoting microbes, and four also contained ectomycorrhizal propagules. None of the products had exceeded their expiry date, if provided. Most inoculants used ground clay or other inert materials as carrier substances.

Concerning research products, five inocula from companies that produce research inoculum were collected via online searches and knowledge of commercial producers. The sixth and seventh research inocula, labelled S and X, correspond to the *Rhizophagus irregularis* SAF22 inoculum produced in our laboratory for research purposes and used as a positive control. The two conditions S and X, correspond to two different inoculum concentrations, at 5% and 1% of the pot volume respectively. In brief, the inoculum for treatment S and X was subcultured on *Plantago lanceolata* (Europe) in a closed pot system, with substrate containing AMF propagules used as inoculum. Previous studies have demonstrated the effectiveness of this isolate, as evidenced by the high level of mycorrhizal root colonisation observed following inoculation (Lutz et al. 2023).

## 2.2 | Greenhouse Experimentation

Onion seeds (*Allium cepa* cv. Crockett, supplied by Bejo Zaden B.V.) were selected for the experiment. The seeds were surface-sterilised in a solution of 2.5% KCIO for 10 min, then rinsed several times with sterile deionised water and finally soaked in sterile deionised water overnight. The seeds were pre-germinated on autoclaved sand (121°C for 30 min) at 25°C for 24 h, after which they were grown in the dark at room temperature for 72 h.

Three plantlets were then transplanted into each 1.5 litre pot, which was filled with an autoclaved mixture (1:1, v/v) of quartz sand (diameter < 1 mm) and agricultural soil collected from Agroscope Reckenholz (47°42'69" N, 8°51'43" E; see Supporting Information S1: Table 1 for soil parameters). Before transplantation, inocula were applied to the pots according to the manufacturer's recommendations (Table 2). The plants were then grown in a greenhouse at the University of Zürich (47°39'47" N, 8°55'02" E) under a 16-h light/8-h dark cycle with temperatures set at 24°C (day) and 18°C (night). The plants were watered to field capacity using deionised water and fertilised weekly with 20 ml of modified Hoagland solution containing 0.125 mM phosphorus (as  $\text{NH}_4\text{H}_2\text{PO}_4$ ).

The experiment followed a randomised complete block design (RCBD) with eight replicates per treatment, resulting in a total

of 192 experimental pots. At the end of the experiment, the shoots and roots were harvested separately.

## 2.3 | Extraction of Product Propagules and Nutrient Analysis

To verify product quality, the arbuscular mycorrhizal fungi (AMF) propagules (mycorrhized roots and/or AMF spores) potentially present in the commercial inoculants were extracted using the wet sieving method, followed by sucrose density centrifugation (Oehl et al. 2005). In brief, before wet sieving, the solid inoculants were suspended in sterilised distilled water and treated in an ultrasonic bath at 30 kHz for 120 s (Bandelin Sonorex; Boyno et al. 2023). The inoculants were then poured through sieves with mesh sizes of 1000, 500, 125 and 32  $\mu\text{m}$  and rinsed with distilled water. Potential root fragments on the 500  $\mu\text{m}$  mesh were collected and gathered with remnants from the 125  $\mu\text{m}$  and 32  $\mu\text{m}$  meshes. The sucrose density centrifugation method was then used to separate the propagules from contaminants that would interfere with counting. The propagules were then counted under a binocular microscope at 16x magnification. Root and/or spore concentrations were determined relative to the weight or volume of the inoculum sample used. The nutrient content of the products was analysed for nitrogen, phosphorus and potassium (Agroscope, Reckenholz, Zürich).

## 2.4 | Mycorrhizal Root Colonisation Measurement by Microscopy

For each plant, a subsample of fresh roots was used to determine the degree of arbuscular mycorrhizal fungus (AMF) colonisation. The root fragments were cut to a length of 1 cm, stained with 0.005% trypan blue solution (w/v in a mixture of lactic acid, glycerol and water at a ratio of 1:1:1, w/w/w) at 60°C for 10 min in a 15 mL tube in a water bath, then distained for 24 h in a solution of 1% HCl in glycerol (w/w) (Phillips and Hayman 1970). Root colonisation (frequency of hyphae, arbuscules and vesicles) was estimated using the grid-line intersection method (Giovannetti and Mosse 1980). No AMF structures were observed in non inoculated (NM) plant roots.

## 2.5 | DNA Extraction, and Quantification of Root Mycorrhizal Abundance by qPCR

For each plant, a subsample of approximately 100 mg of fresh roots was snap-frozen and stored at -80°C. Genomic DNA was extracted from each root sample using the NucleoSpin 96 Plant II kit (Macherey-Nagel GmbH) according to the manufacturer's instructions. The quality and concentration of the extracted DNA were assessed using a NanoDrop ND-1000 spectrophotometer (NanoDrop Technologies).

The abundance of the nuclear small ribosomal subunit (SSU) of AMF sequences was determined by qPCR absolute quantification for each sample. Amplifications were performed using HOT FIREPol® EvaGreen® qPCR Mix Plus and the Bio-Rad CFX Maestro 2.2 thermocycler. The primer pair AMG1F (Hewins et al. 2015) and

AM1 (Helgason et al. 1998) was used to target AMF SSU sequences. These primers are specific to the phylum Glomeromycota and have been validated as suitable for detecting a broad diversity of AMF taxa (Bodenhausen et al. 2021). Quantitative RT-PCR was performed in a 384-well plate with the following PCR cycling conditions: 95°C for 12 min; 40 cycles of 95°C for 15 s, 62°C for 30 s and 72°C for 20 s. Primer specificity was further verified by melting curve analysis at the end of each qPCR run (65°C–95°C, 0.5°C increments), ensuring amplification of a single product.

Standard curves were generated using tenfold serial dilutions ( $1.39 \times 10^{-3}$  –  $1.39 \times 10^{-9}$  ng DNA  $\mu\text{L}^{-1}$ ) of a 230 bp PCR amplicon derived from AMF genomic DNA previously isolated from field soil. Each qPCR plate included a complete standard series and no-template controls (NTCs) to monitor potential contamination. Amplification efficiency (mean = 1.953) and coefficient of determination ( $R^2 > 0.98$ ) confirmed reliable and consistent quantification.

Each biological sample was analysed in triplicate qPCR reactions, and mean values were used for further analyses. Raw fluorescence data were processed with LinRegPCR v2016.0 to determine the threshold cycle ( $C_t$ ) and reaction efficiencies. Copy numbers per ng of DNA were calculated from the standard curve and normalised across samples.

## 2.6 | Statistical Analysis

All statistical analyses were performed using R version 4.2.0 (R Core Team, 2022). The `ggplot` function from the `ggplot2` package (version 3.3.3) was used for data visualisation. Multiple plots were arranged and displayed using the `patchwork` package (version 1.3.0). All analysis of variance (ANOVA) tests were assessed for normality using the Shapiro-Wilk normality test from the `stats` package (version 4.0.3). When the normality assumption was not met, data were normalised using the `bestNormalize` package (version 1.8.2), which identified the square root transformation ( $\sqrt{x}$ ) as the most appropriate normalisation method. Homogeneity of variance was checked using the Levene test from the `car` package (version 3.0-10). When the data structure permitted, a more complex model was employed. Specifically, a linear mixed effects model (LMM) was fitted using the `lme4` package (version 1.1-35.5) with restricted maximum likelihood (REML) to estimate variance components and account for fixed and random effects. Post hoc pairwise comparisons using estimated marginal means (EMM) were performed using the `emmeans` package (version 1.5.1). For all statistical tests, a significance level of  $p < 0.05$  was considered significant. Relation between mycorrhizal colonisation rate (%) and abundance of mycorrhizal copy number measured by qPCR was assessed based on Pearson's correlation test.

## 3 | Results

### 3.1 | Growth Response to Mycorrhizal Product Inoculation

Of the nine agricultural products tested, five (D, G, I, K and Q) produced a significant positive growth response. In the

home gardening category, two products (E and H) significantly enhanced plant growth, while one product (B) resulted in a significant negative growth response. Of the products intended for research use, four out of seven promoted a significant increase in plant growth following inoculation (Figure 1A).

### 3.2 | Estimation of AMF Root Colonisation With Microscopy and Molecular Tools

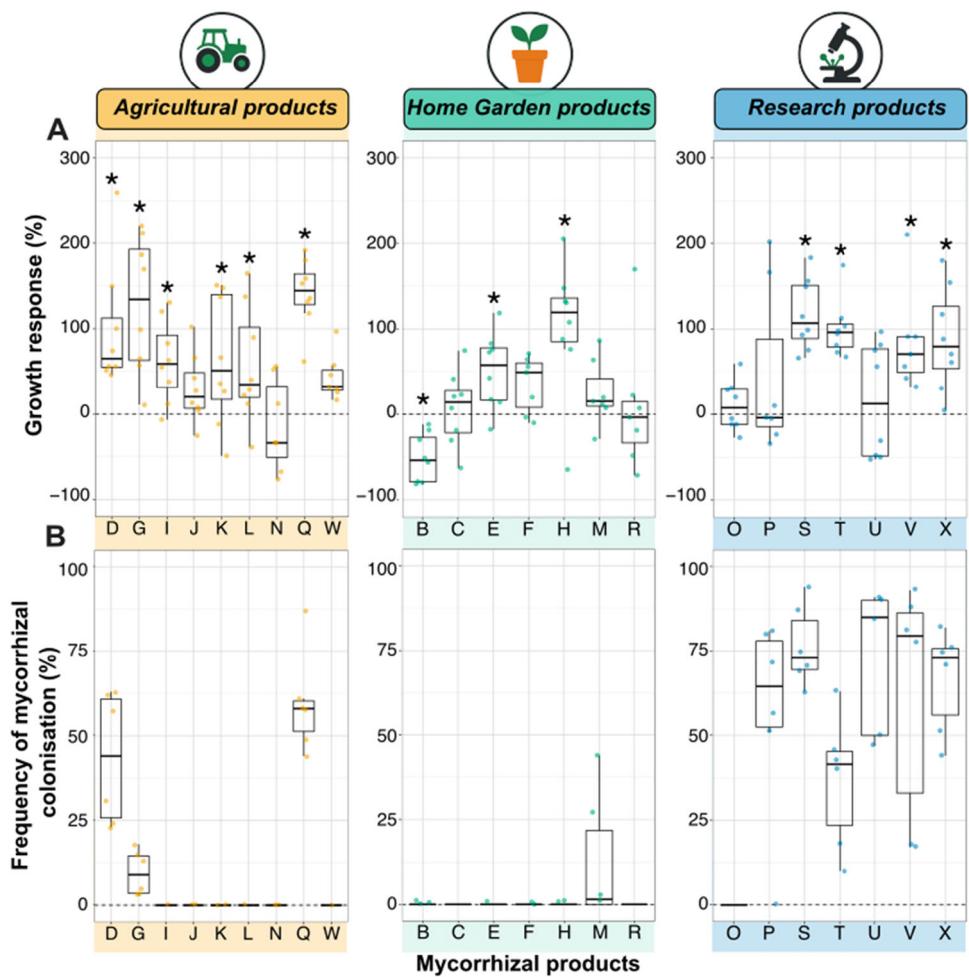
Microscopic assessment of root colonisation revealed that, out of nine agricultural products, only three (D, G and Q) effectively established colonisation by AMF, with colonisation rates of 45%, 10% and 60% respectively (Figure 1B). In contrast, none of the home gardening products resulted in observable AMF colonisation. Of the inoculants intended for research use, all but one product induced root colonisation, with most exhibiting levels exceeding 40%. Colonisation rates measured via quantitative PCR (qPCR) showed a significant correlation ( $R = 0.59$ ,  $p < 2.10^{-16}$ ) with those obtained through microscopic evaluation in onion (Supporting Information S1: Figure 1), in line with an earlier report showing that it is plant species dependent (Corona Ramírez et al. 2023).

### 3.3 | Assessing Propagules Concentration in the Mycorrhizal Products

The assessment of propagules (root fragments and spores) in agricultural products revealed an average concentration of 112.3 root fragments and 178.4 spores per gram or millilitre of product. Specifically, products D, G, K, L, N and Q contained moderate to high numbers of propagules, whereas products I, J and W showed only a low concentration. Home gardening products had the lowest overall propagule content, with an average of only 0.7 root fragments and 2.7 spores per g or mL; most of these products either lacked propagules entirely or contained extremely small amounts. Some research products contained exceptionally high numbers of propagules (particularly O and P), which is consistent with their formulation for experimental purposes (Table 1). For all agricultural and home gardening products, the number of propagules communicated by the manufacturer, if any, was consistently higher than the number measured (Table 1).

### 3.4 | Assessing Nutrient Contents in the Products

The products differed in their physicochemical compositions (Supporting Information S1: Tables 2 and 3). Carbon content ranged from less than 1 g/kg DM in products F to over 531 g/kg DM in product W, with products B, L and W exhibiting the highest concentrations. This suggests that the carrier for the formulation of products B, L and W is made of organic matter. The highest total nitrogen content was found in products B (33.32 g/kg DM) and R (34.35 g/kg DM). Phosphorus content ranged from 0.07 to 20.52 g/kg DM, while potassium content varied from 0.4 to 66.82 g/kg DM (Supporting Information S1: Tables 3).



**FIGURE 1** | Mycorrhizal growth response (A) and frequency of AMF root colonisation assessed by microscopy (B) across the 23 mycorrhizal inoculant products tested. The products are grouped into three categories: Agricultural products, Home Garden products, and Research products. Asterisks indicate statistically significant differences between each inoculated treatment and the NM control (condition A), based on post hoc comparisons using the emmeans test. Not significant:  $p \geq 0.05$ ;  $*p < 0.05$ .

## 4 | Discussion

The use of microbial inoculants in agriculture dates back more than a century, and their adoption has increased significantly in recent years (Santos et al. 2019). The development of sustainable practices has driven a growing market for commercial mycorrhizal products; however, their efficacy remains highly variable mainly because poor inoculum quality (Salomon et al. 2022b; Hart et al. 2018; Koziol et al. 2024). Although inoculation with mycorrhizal fungi has the potential to enhance plant yield and product quality (Bender et al. 2016), the effectiveness of commercial formulations depends on various factors, such as formulation type and propagule viability, application strategy, as well as biotic and abiotic factors in the field, such as soil type, plant variety, and soil microbiome (Lutz et al. 2023; Berruti et al. 2016). In this study, we systematically evaluated 23 mycorrhizal products grouped into three categories: Agricultural products, Home Garden products, and Research products. Unlike previous assessments, which have primarily focused on large-scale agricultural formulations, our work is the first to provide a systematic evaluation of 'Home Garden' products, a rapidly growing yet poorly regulated segment of the market. We found that most of the commercial products (Agricultural and Home

Garden products) do not lead to both growth responses and AMF root colonisation, highlighting important limitations in the current European market landscape.

### 4.1 | Only Three out of 16 Commercial Products Induced Positive MGR and Lead to Root Colonisation With Arbuscular Mycorrhizal Fungi

Assessment of propagules in commercial products (agricultural and home gardening categories) revealed that the majority of products did not meet the propagule numbers reported by manufacturers. This finding is consistent with previous reports indicating that some commercial mycorrhizal products contain significantly fewer viable propagules than claimed (Tarbell and Koske 2007; Salomon et al. 2022b; Koziol et al. 2024). Among the 16 commercially available AMF products tested in this study (agricultural and home gardening categories), only three induced root colonisation by AMF and positive growth response. Similar patterns have been documented in the United States (Tarbell and Koske 2007; Koziol et al. 2024), Africa (Faye et al. 2013), as well as in Europe and Australia (Salomon et al. 2022b). Despite the absence of AMF colonisation, five out

of 16 commercial products still induced a significant growth response. In some cases, it has been suggested that the promotion of growth by certain products may be due to nutrient enrichment (e.g., N, P, K) rather than a true microbial effect (Salomon et al. 2022b; Eric Wiseman et al. 2009). However, in our study, the impact of high nutrient levels on plant growth could not be attributed to any products except product B, which had a negative effect on plant performance, likely due to nutrient toxicity or osmotic stress.

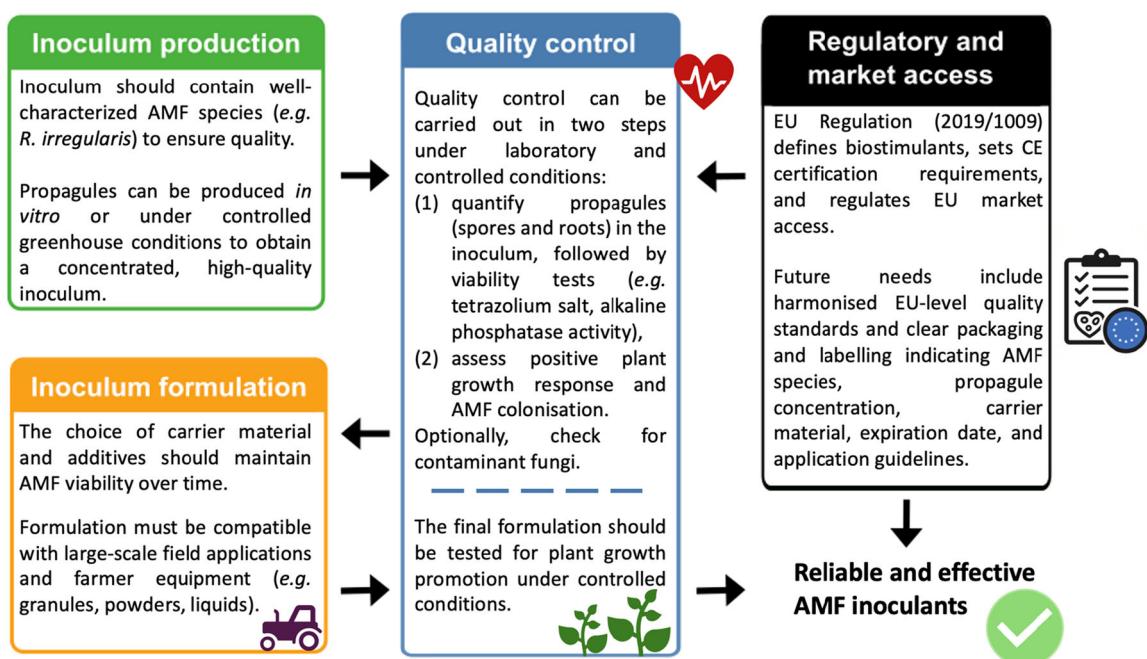
In other products, the observed growth promotion may be linked to the inclusion of additional beneficial microorganisms such as *Trichoderma* spp. or plant growth-promoting rhizobacteria (PGPR), both of which are well-documented for their capacity to improve plant growth independently of AMF (Complant et al. 2025). For instance, the positive effects observed with products I and H could be attributed to the presence of *Trichoderma*, while the growth stimulation observed with product L may be explained by the presence of PGPR (e.g., *Bacillus amyloliquefaciens*). Synergies between AMF, PGPR, and fungi such as *Trichoderma* have been reported to enhance plant performance (Noceto et al. 2021). Although these interactions are intensively studied in the laboratory, they remain poorly represented in commercial formulations (Complant et al. 2025). Nevertheless, synthetic microbial communities (SynComs) are emerging as a promising approach for developing effective inoculants in sustainable agriculture (Delgado-Baquerizo et al. 2025). SynComs are designed to enhance the resilience of crops against biotic and abiotic stresses by mimicking natural microbiome structures and functions. In

comparison to traditional single-isolate inoculants, SynComs offer a range of benefits over a wider spectrum of stresses and soil conditions (Delgado-Baquerizo et al. 2025).

## 4.2 | Research Products Contain High Quality Inocula

For research products, all treatments with positive growth response exhibited fungal colonisation with AMF, although colonisation did not consistently lead to positive MGR in all treatments (Figure 1). Research products were typically formulated with a single AMF species, *R. irregularis* (Basiru et al. 2020), a model organism that is widely used in mycorrhizal studies due to its sequenced genome and broad ecological adaptability (Savary et al. 2018). In addition, the mass production of *R. irregularis* remains a cost-effective solution in both greenhouse and in vitro systems, due to the significant yield of propagules (Rosikiewicz et al. 2017).

Overall, the research products exhibited higher propagule concentrations than the commercial products as observed earlier (Tarbell and Koske 2007; Koziol et al. 2024). Notably, differences were observed even between research and commercial formulations produced by the same manufacturer. For instance, product M (a home-garden commercial formulation by one manufacturer) contained significantly fewer propagules than product T (a research formulation produced by the same manufacturer), failed to establish root colonisation, and did not enhance plant growth. In contrast, Product T



**FIGURE 2 |** Practical framework for evaluating the quality and reliability of commercial AMF products. The process involves four key components: (1) Inoculum production, typically via *in vitro* culture, greenhouse, or open-pot systems to obtain high propagule density and viability; (2) Inoculum formulation, which maintains AMF viability and is adapted to field application to ensure plant benefits; (3) Quality control, including propagule quantification, viability assays, greenhouse validation, and pathogen screening; and (4) Regulatory and market access, guided by EU Regulation 2019/1009, which defines standards for biostimulants, CE certification, and market access. Reliable and effective AMF inoculants require harmonised quality standards and third-party.

led to successful AMF colonisation and a measurable positive effect on plant performance. For another manufacturer, Product Q (the commercial version of the research product P) achieved colonisation rates above 50% (like the research product) despite having substantially lower propagule densities (spore counts were reduced by 50%, and root fragment numbers by 800%).

Although it has been suggested that colonisation may require application rates five to 10 times higher than recommended (Tarbell and Koske 2007), these findings suggest that differences in formulation quality, not necessarily propagule number, account for disparities in product performance. Enhancing inoculum quality (e.g., through increased viability and extended shelf life) may be a more effective and practical strategy from both agronomic and commercial perspectives for improving product efficacy than simply increasing application rates (Koziol et al. 2025; Basiru et al. 2020).

#### 4.3 | Call for Systematic Quality Control

To improve the reliability and effectiveness of microbial inoculants, it is essential that inoculum producers adopt a standardised and systematic quality control framework (Salomon et al. 2022a; Koziol et al. 2025). For instance, the manufacturer of agricultural product I communicated about 10% of 'propagule enrichment', while the manufacturer of product M communicated about '5000 pieces of AMF', which is not scientifically meaningful. Homogenised counting system is therefore necessary to compare the number of propagules (root fragments and spores) in different inoculum products. Moreover, the number of propagules should be communicated on every packagings.

Propagules counting should be combined with viability tests (Figure 2). Several staining techniques are available to evaluate the viability of AMF (e.g., tetrazolium salt staining; [Pepe

**TABLE 1** | Number of AMF propagules claimed by the manufacturers and number of propagules counted under the microscope after extraction.

Products	Number of propagules claimed by the manufacturers per g or mL of product Propagules	Number of propagules counted per g or mL of product	
		Root fragments	Spores
<b>Agricultural products</b>			
D	900	71	0
G	250	2	91
I	10%	1	1
J	1'975'042	0	5
K	2'000 (spores)	2	514
L	7'500	257	865
N	1'528	650	102
Q	145	28	24
W	178	0	4
<b>Home Garden</b>			
B	NA	0	0
C	245 (spores)	0	1
E	220	3	0
F	NA	1	1
H	NA	1	14
M	5'000 (pieces)	2	9
R	NA	0	0
<b>Research</b>			
O	2000 (spores)	0	2196
P	NA	24,712	1288
S	NA	1912	34
T	NA	47	39
U	NA	47	16
V	NA	NA	0
X	NA	1912	34

*Note:* The numbers are expressed as the number of propagules per gram or millilitre of product and are separated into root fragments and spores where possible. Products are grouped by usage: Agricultural products, Home Garden products and Research products. For product V, the material was pure root fragments, so counting the root fragments was not possible.

**TABLE 2** | Amount of each product applied per pot following manufacturer's recommendations.

Product	Customer	Formulation	Field application	Application/pot
B	Home garden	Powder	2 g/10 cm Seed furrow	2.6 g/pot (2.6 g/13 cm seed furrow)
C	Home garden	Powder	Mix 5 g product with planting soil	5 g/pot (Mixed into substrate)
D	Agricultural	Granules	10 g/L medium	10 g/pot (Mixed into substrate)
E	Home garden	Powder	2 g/L volume	3 g/pot (Mixed with substrate)
F	Home garden	Substrate	1–2 Teaspoon/plant	2 Teaspoons/pot
G	Agricultural	Powder	Layer 3 cm below seed 150 g/m <sup>2</sup>	2 g/pot (3 cm below seed)
H	Home garden	Powder	Apply into seed furrow	2 teaspoons directly into seed furrow
I	Agricultural	Granules	1–3 kg/1000 L substrate	3 g/pot (Mixed with substrate)
J	Agricultural	Liquid	90 g/150 L water for 30 m <sup>2</sup>	Dissolving 0.6 g/L water, 6.5 ml/pot
K	Agricultural	Powder	500 g/m <sup>3</sup>	0.5 g/pot (Mixed into substrate)
L	Agricultural	Water soluble powder	1 kg/1000 L/ha	1 g dissolved/L water, 2 ml into seed furrow/pot
M	Home garden	Granules	10 mL/L soil volume	15 mL/pot (Mixed into substrate)
N	Agricultural	Liquid	4–6 g/0.41 ha (spray into seed furrow)	2 ml/20 mL water, 2 ml into seed furrow/pot
O	Research	Liquid	No information	200 spores/pot
P	Research	Root fragments	No information	800 mg mixed with 8 g Sand
Q	Agricultural	Powder	5% of substrate volume	25 mL/pot (Mixed into substrate)
R	Home garden	Granules	100–150 g/m <sup>2</sup>	2 g/pot
S	Research	Powder	5% of substrate volume & 0.1% root fragments	50 mL/pot (Mixed into substrate)
T	Research	Granules	10 mL/L soil volume	15 mL/pot (Mixed into substrate)
U	Research	Liquid	2400 spores/mL	600 spores/pot
V	Research	Roots	No information	2 g/pot
W	Agricultural	Granules	40 g/100 metre seed furrow	0.15 g/pot
X	Research	Powder & roots	1% of substrate volume & 0.1% root fragments	15 mL/pot (Mixed into substrate)

et al. 2018]) and to identify metabolically active propagules (e.g., alkaline phosphatase activity; [Pepe et al. 2018]). In addition to laboratory tests, greenhouse experimentation under controlled conditions using sterilised substrate remains essential to assess real effective root colonisation potential and plant growth responses (Figure 2).

Producers should clearly declare the AMF species and strains present in their inoculum (Faye et al. 2013). Moreover, a recent study identified the presence of fungal plant pathogens in certain commercial AMF products (Koziol et al. 2024), indicating the need for enhanced monitoring of production systems for plant pathogens (Figure 2).

Quality control measures should be implemented before and following product formulation to ensure that the manufacturing process does not affect the effectiveness of the active ingredients. Developing a formulation that (i) maintains AMF viability, (ii) is easily implemented, and (iii) is compatible with farm

machinery presents a significant challenge (Figure 2). Recent work suggests that exposing inoculum to low temperatures may help increase AMF shelf life by inducing a state of partial dormancy, thereby reducing metabolic activity and slowing propagule deterioration (Liu et al. 2022). Such strategies, alongside optimised carriers and protective additives, could help preserve viability during storage and transport. Nonetheless, overcoming these technical constraints is necessary to ensure the consistency, efficacy, and broader adoption of mycorrhizal inoculants in agricultural systems (Salomon et al. 2022a).

#### 4.4 | Call for Common Regulatory Framework

The global agricultural inoculants market, encompassing products like rhizobia and Trichoderma species, was valued at \$1.1 billion in 2022 and is anticipated to reach \$1.7 billion by 2027 (Santos et al. 2019). In comparison, the market for

**TABLE 3** | Success rates of commercial arbuscular mycorrhizal inoculants in inducing effective root colonisation.

Study	Origin of the inoculum tested	Number of AMF inoculants tested	Number of AMF inoculants leading to root colonisation	Success rate of effective root colonisation
This study	E.U.	16	3	18.8%
Faye et al. (2013)	NA	12	3	25.0%
Salomon et al. (2022a)	E.U. & Australia	20	6	30.0%
Koziol et al. (2024)	USA	16	4	25.0%
		52	13	25.0%

*Note:* This table reports the number of products tested, the number that achieved colonisation and the corresponding success rate, as well as the results from this study and previous studies. AMF inoculants leading to root colonisation were defined as being significantly different to the non-inoculated control and/or inducing  $\geq 10\%$  colonisation in sterilised substrate.

mycorrhizal inoculants alone is projected to reach \$1.1 billion by 2027 (Santos et al. 2019). However, at present, more than 80% of commercial products fail to facilitate even minimal root colonisation, resulting in an estimated \$850 million wasted annually on ineffective products (Koziol et al. 2025). The inconsistent efficacy of these products has limited their adoption, often restricting their use to ‘idealistic’ farmers, while many conventional farmers remain hesitant (Salomon et al. 2022a).

Regulatory frameworks are also vital to ensure product quality and efficacy. Currently, the management of quality control is primarily the responsibility of the producers themselves, with no uniform regulatory standards in place at a European level (von Alten et al. 2002). Collaboration among scientists, regulatory bodies, and businesses is required to address challenges such as product standardisation and regulatory compliance (Ghorui et al. 2024). Addressing these issues will help ensure the reliability and efficacy of AMF products, encouraging their adoption in sustainable agricultural practices (Figure 2).

In this regard, a major advancement for commercial products was the implementation of Regulation (EU) 2019/1009 (European Union) in July 2022. This regulation defines biostimulants by function, distinguishing them from Plant Protection Products, and enables manufacturers to obtain an EU-type certificate with the CE mark, granting access to the EU market (Ghorui et al. 2024). Such certification should provide reassurance to farmers that AMF inoculants meet quality standards and provide agronomic and environmental benefits. The ultimate goal of this process is to enhance the quality of commercial AMF products, for broader adoption in the agro-systems, to improve plant yield and food quality for consumers.

## 5 | Conclusion

Despite growing interest in microbial inoculants, AMF products remain inconsistent and opaque in terms of their formulation and labelling (Salomon et al. 2022a; Koziol et al. 2024; Koziol et al. 2025). Across our study, in which we tested 16 commercial inoculants, and previous investigations by Faye et al. (2013), Salomon et al. (2022a) and Koziol et al. (2024), a total of 64 products were assessed. However, only around 25% of the inoculants actually demonstrated the ability to successfully

colonise plant roots (Table 3). This low success rate highlights the persistent lack of quality control and independent validation that continues to undermine user confidence in AMF inoculants (Salomon et al. 2022a).

Developing cost-effective formulations that ensure both viability and field efficacy poses a significant challenge to the AMF industry. To fully harness the potential of these symbionts, producers must improve production efficiency and adapt formulations to diverse agricultural systems. Inspiration could be taken from advances in the formulation of other microbial inoculants, such as *Rhizobium* or *Trichoderma*, or even from pharmaceutical delivery systems that prioritise precision and stability.

Even when viable inoculants are available, inoculation success under field conditions remains highly variable. While recent studies have begun to identify the key factors influencing AMF establishment and performance (e.g., soil type, host compatibility, and native microbial competition) (Lutz et al. 2023; Rog et al. 2025), many have relied on unrealistically high inoculum doses that do not reflect commercial practice. There is therefore an urgent need for applied research using efficient commercial formulations under realistic farming conditions, to understand the factors influencing inoculation success (Salomon and Watts-Williams 2025).

## Author Contributions

R.B. and M.H. conceived the experiments. R.B., M.E., and J.F. conducted the experiments, measured and analysed the results. M.H. secured the funding. R.B. wrote the manuscript and all co-authors reviewed the manuscript.

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## Ethics Statement

The authors have nothing to report.

## Conflicts of Interest

The authors declare no conflicts of interest.

## Data Availability Statement

Data are either provided in the manuscript within the main document, or as supporting information, or can be requested from the corresponding author.

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## Supporting Information

Additional supporting information can be found online in the Supporting Information section.

**Figure S1:** Abundance of AMF sequence in plant root assessed by qPCR across the 23 mycorrhizal inoculant products tested. The products are grouped into three categories: Agricultural products, Home Garden products, and Research products. **Figure S2:** Relationship between mycorrhizal colonization rate (%) and abundance of mycorrhizal copie number measured by qPCR. Each point represents an individual sample. **Table S1:** Soil analysis for the soil used for the experimentation. **Table S2:** Composition analysis of the 23 products tested. **Table S3:** Nutrient analysis of nitrogen, phosphorus and potassium for the 23 products tested.