



Soil cropping selects for nutrient efficient but more costly indigenous mycorrhizal fungal communities

Paula A. Buil^{1,2} · Jan Jansa³ · Martin Rozmáš³ · Michala Kotianová³ · Petra Bukovská³ · Gabriel Grilli⁴ · Nicolás Marro^{1,4} · Martina Janoušková¹

Received: 17 October 2024 / Revised: 13 February 2025 / Accepted: 19 February 2025 / Published online: 4 March 2025
© The Author(s) 2025

Abstract

Conventional agriculture has been suggested to promote less mutualistic arbuscular mycorrhizal fungi (AMF). The main aim of this study was to test this assumption by a detailed functional analysis of the plant mycorrhizal benefits and costs. A cross-inoculation experiment was established with *Plantago lanceolata* as a host plant and inocula of AMF sourced from four pairs of conventionally managed arable fields and neighbouring grasslands. Mycorrhizal effects were determined for a range of plant parameters including fluxes of isotopically labelled phosphorus (P), nitrogen (N) and carbon (C), and related to root colonization and composition of the different AMF communities.

The association of *P. lanceolata* with arable-field inocula was less beneficial in terms of plant growth promotion and it also led to more pronounced P accumulation in plant biomass, as compared to grassland inocula. Furthermore, arable-field AMF increased ¹⁵N depletion in soil and ¹⁵N transfer to shoots, and induced higher ¹³C drain to soil. These differential functional parameters were related to consistent compositional differences between arable-soil and grassland AMF communities in the roots. Differential effects of the AMF inocula on N and C partitioning in the soil–plant system suggest faster foraging for nutrients by arable-soil AMF and higher demand for C, which are characteristics associated with ruderal AMF. This implies that arable-soil AMF may be less beneficial in conditions of plant growth limitation by C than the grassland AMF.

Keywords Arable soil · Functional traits · Grassland · Mycorrhizal benefits · Nitrogen · Phosphorus

Introduction

Arbuscular mycorrhiza (AM) is primarily a nutritional symbiosis of plants with obligately biotrophic arbuscular mycorrhizal fungi (AMF), where fungi forage for low-available nutrients in soil and provide them to their host plants in exchange for photosynthates. Thus, the plant net benefits from the symbiosis can be simplified as the sum of gross

benefits in terms of increased nutrient uptake, and carbon (C) costs in terms of the share of photosynthates provided to the fungal partner (Smith and Read 2008). The classical trade balance model states that the functioning of AM is mostly determined by the availability of plant C and the two most important macronutrients, namely nitrogen (N) and phosphorus (P) (Johnson 2010). While plant benefits from the fungal-mediated uptake of P are indisputable especially at low P availability in soil, the fungal contribution to plant N uptake depends not only on N availability in soil, but also on its forms, P availability and the N demand of the fungal symbionts (Corrêa et al. 2015; Hodge and Storer 2015; Johnson 2010; Püschel et al. 2016). Typically, a strongly mutualistic AM, i.e. beneficial for both symbionts, is predicted for conditions of low P availability and high N availability in soil, in conjunction with high availability of plant C to the fungal partners (Johnson 2010; Johnson et al. 2015).

In natural conditions, AM is usually formed by AMF communities, in which different taxa compete for root space and plant-derived C (Bennett and Bever 2009; Pearson

✉ Paula A. Buil
paula.buil@ibot.cas.cz

¹ Institute of Botany, Czech Academy of Sciences, Zámek 1, 252 43 Průhonice, Czech Republic

² Department of Botany, Faculty of Science, Charles University, Benátská 2, 128 01 Prague 2, Czech Republic

³ Institute of Microbiology, Czech Academy of Sciences, Vídeňská 1083, 142 00 Prague, Czech Republic

⁴ Instituto Multidisciplinario de Biología Vegetal (IMBIV), CONICET, FCEfYN, Universidad Nacional de Córdoba, 495, 5000 Córdoba, CC, Argentina

et al. 1993; Werner and Kiers 2015), while their extraradical mycelia proliferate into soil. Abundant evidence shows that species/genotypes of AMF differ in a range of morphological and developmental traits such as mycelial growth rates, extent and density of extraradical mycelia, their responsiveness to soil patchiness, and/or dynamics and intensity of spore production (e.g. Hart and Reader 2002a; Koch et al. 2017; Lendenmann et al. 2011; Mensah et al. 2015; Munkvold et al. 2004; Oehl et al. 2009; Powell et al. 2009; Säle et al. 2021; Sun et al. 2024; Marrassini et al. 2024). Based on this knowledge, a life-history differentiation of AMF has been proposed following Grime's C-S-R framework (Grime 1977), which suggests traits indicative for competitors, stress-tolerators and ruderals, and predicts trait selection by specific environmental conditions (Chagnon et al. 2013). For example, high growth rates, early and abundant sporulation are proposed as traits of ruderal species, which are favoured by high level of soil disturbance. However, while morphological traits can be relatively easily measured and display consistent taxonomic patterns (Hart and Reader 2002b; Koch et al. 2017; Pellegrino et al. 2024; Powell et al. 2009), it has proven difficult to relate specific morphological traits to functional traits such as the rate of P transfer to host plants or requirements for plant-derived C (Hart and Reader 2002a; Koch et al. 2017; Mensah et al. 2015). Consequently, functional aspects of the proposed life-history differentiation of AMF remain rather hypothetical (Chagnon et al. 2013).

A typical habitat imposing a strong selection pressure on AMF is conventional agriculture, where soils are regularly disrupted by tillage, enriched with readily available nutrients, planted with monocultures of fast-growing crops and disbalanced by pesticide application (Edlinger et al. 2022; Fritze et al. 2024). These selection pressures, considered unfavourable to functional mycorrhiza, have been suggested to promote dominance of genotypes of AMF, which invest more in competition and reproduction than in provisioning of nutrients to the host plant (Johnson and Gibson 2021; Porter and Sachs 2020; Verbruggen et al. 2010). Indeed, practicing conventional agriculture often suppresses or eliminates disturbance-sensitive species of AMF (Jansa et al. 2002; Lekberg et al. 2008; Ryan and Graham 2018), promotes fast-growing AMF which produce ample spores (Chagnon et al. 2021; Oehl et al. 2009) or reduces species associated with high ratios of N:P (Verbruggen et al. 2015). However, the consequences of these taxonomic and trait shifts for plant benefits from the mycorrhizal association require further investigation (Chagnon et al. 2021).

Indigenous AMF from arable soils improve crop nutrition and growth in suitable soil conditions (e.g. Buil et al. 2023; Chatterjee and Margenot 2023; Edlinger et al. 2022; Liu et al. 2002; Martinez and Johnson 2010;), but it is also indisputable that AM functions differently in conventionally

managed agricultural soils as compared to less intensive management types (Cofré et al. 2020; Edlinger et al. 2022; Kahiluoto et al. 2009; Peng et al. 2024; Williams et al. 2017). This can be partly attributed to the less favourable soil conditions and lower infectivity of AMF in intensively managed arable soils (Buil et al. 2023; Köhl et al. 2016; Lekberg and Koide 2005), but also to the above-outlined selection for specific taxa and functional traits by intensive agricultural management. Considering the latter, the functional differences of AMF communities from differently managed soils should also persist (at least transiently) upon changing their environmental conditions. Yet, testing this requires direct functional comparison of the different AMF communities upon identical soil–plant conditions.

Indeed, some of such direct comparisons previously revealed lower net benefits of plants associated with indigenous AMF communities from high-input agricultural soils as compared to less intensive management types (Eason et al. 1999; Gottshall et al. 2017; Johnson 1993; Peng et al. 2024; Scullion et al. 1998). Others, however, reported more fine-scale functional differentiation (Andrino et al. 2021; García de Leon et al. 2020; Köhl et al. 2014; Marro et al. 2020; Verbruggen et al. 2012). For example, AMF from less intensive management types provided more P to their host plants, but their extensive mycelia were also suggested to impose higher C costs (Köhl et al. 2014; Verbruggen et al. 2012). Unfortunately, previous studies only rarely interlink the observed differences in plant benefits with differences in the community composition of AMF or provide more detailed insight into the functioning of the symbiosis as per the trade balance model (Johnson 2010). Usually, only plant growth is focused as the net symbiotic benefit of the plant, while the gross benefits in terms of P or N uptake, or even the C costs of the symbiosis remain partly or entirely unknown. Understanding their contribution to the (lower) net benefits of mycorrhizas formed with AMF communities from arable soils may improve the predictability of the lower benefits.

The main aim of our study was therefore to quantify plant symbiotic benefits and costs of mycorrhizas initiated by AMF inocula sourced from conventionally managed arable fields and from neighbouring minimally disturbed grasslands and link them to the community composition of the AMF. In accordance with the assumption that intensive agricultural management selects against mycorrhizal mutualism (Johnson and Gibson 2021; Porter and Sachs 2020; Verbruggen et al. 2010), we expected arable-field AMF inocula to differ from grassland inocula in providing (1) lower net benefits to plants as approximated by plant biomass, which would be related to (2) lower supply of P from the soil to the host plant. Furthermore, we expected the functional differences in the mycorrhizas to be associated with (3) differences in AMF community composition, indicative for higher

proportion of ruderal taxa within the arable-field communities. A cross-inoculation experiment was designed to evaluate the responses of *Plantago lanceolata* to AMF inocula from four pairs of arable soils and grasslands, accounting for the effect of source soils.

Materials and methods

Soils and inocula

The soils and AMF inocula were collected at four sites in northern Bohemia, Czech Republic (Drahobuz, Encovany, Malíč and Svařenice; henceforth Dra, Enc, Mal and Sva, respectively)—each site including a pair of an arable field and a neighbouring seminatural grassland (Online resource 1: Table S1). The arable fields were long-term conventionally managed including annual tillage and fertilization with mineral fertilizers, and all four were planted with winter wheat in the year of sampling. The grasslands were exposed to low level of disturbance: unfertilized, maintained by irregular mowing or grazing, with typical vegetation of the Bromion community characterized by high species richness (Ellenberg 1988). The area is a well explored system (Dostálek et al. 2022; Florianová et al. 2022; Knappová et al. 2017) and more details on the plant species composition of these grasslands are given by Chýlová and Münzbergová (2008). The soils from the four fields and four grasslands were collected at a transect of at least four spots within each site, at least 2 m distant from each other. They were air dried, sieved through a 1-cm sieve and sterilized by gamma irradiation (> 25 kGy by a ^{60}Co source, Bioster, Veverská Bítýška, Czech Republic). A portion of the non-sterile soils from the above sampling campaign was used to analyse physico-chemical soil properties, and presented in Table S1. The inocula (i.e., living soil samples) were collected at a transect parallel to the field/grassland margin, always at a distance of 20 m from the border. Six subsamples of ca. 200 mL (collected to the depth of 20 cm) were collected from each of the eight field sites into plastic bags, each for one replicate pot. Care was taken to cause minimal disturbance to these soil samples and to maintain integrity of soil lumps (1–3 cm). Samples were stored for a maximum of two weeks in a fridge at 4 °C.

Design and establishment of the experiment

Six treatments were established with material from each site, i.e. pair of grassland and arable field: Plants were grown in previously sterilized grassland or arable soil from the site, and in each of them, either left non-inoculated with AMF, inoculated with grassland AMF or inoculated with arable-field AMF from the same site. Considering

all four sites, the experiment combined in a $3 \times 2 \times 4$ factorial design the factors Inoculation treatment (grassland AMF, arable-field AMF, or no inoculation with AMF), Soil provenance (grassland or arable-field soil) and Site (four levels). Thus, the experiment had 24 treatments in total, each of them replicated six times. For an overview on the experimental treatments see Fig. S1.

The sterilised soils from each site were mixed with autoclaved sand 1:1 (v:v) and about 1200 mL of this substrate was filled into each pot (2 L volume, $10 \times 10 \times 20$ cm, $1 \times w \times h$) over a layer of coarse (autoclaved) gravel. In order to equalize soil nutritional conditions in all inoculation treatments within each site, the addition of inoculum (unsterile soil) was compensated by addition of the same amount of sterilized soil in the other inoculation treatments. Thus, the inoculation treatments were established as follows: 1) Pots inoculated with grassland AMF inoculum received 200 mL of unsterile grassland soil and 200 mL of sterilized arable-field soil from the same site; 2) Pots inoculated with field AMF inoculum received 200 mL of unsterile arable-field soil and 200 mL of sterilized grassland soil from the same site. 3) Non-inoculated controls received 200 mL of sterilized grassland soil and 200 mL of sterilized arable-field soil from the same site. Both added soils were carefully mixed to prevent disturbance of soil aggregates/clods, placed as a layer over the sterilized soil-sand mixture and covered by 300 mL of the same soil-sand mixture as in the bottom. At the establishment of the experiment, each pot also received 20 mL of microbial wash from non-sterile soil (corresponding to the soil used to generate the soil-sand mixture) to re-introduce the indigenous soil bacterial communities. To prepare the microbial wash, 500 g of unsterile soil was mixed with 2 L of distilled water, shaken for 2 h, washed through a 30- μm sieve and twice through a 5- μm filter.

Plantago lanceolata L. was chosen as a host plant for this experiment, because it colonizes both grasslands and disturbed ruderal sites such as field margins, thus representing a plant, which naturally grows at the boundary of both habitats (Chytrý et al. 2021). Seeds of *P. lanceolata* (*Planta naturalis*, Czech Republic) were pre-germinated on moist filter paper and after five days, six seedlings were transferred into each pot. After three weeks, the seedlings were thinned to four plants per pot by removing the weakest seedlings and maintaining regular spacing, where necessary. The pots were kept in a greenhouse and watered daily to maintain approx. 70% of water holding capacity based on the weights of randomly selected pots. The photoperiod was 14 h (natural light + metal halide lamps extending the photoperiod). The temperature and light conditions were monitored every 15 min from August 13th to 25th September 2020. The mean temperature and daylight intensity were 26.16 °C and 9065 Lux respectively.

Tracing C, P and N transfers using isotopic labelling

A root-free compartment was inserted into each pot at the establishment of the experiment. It was prepared of a horizontally placed PVC tube (3.5 diameter, 3 cm length) covered on both sides with a 40 µm nylon mesh (Uhelon 130 T, Silk & Progress, Beněec, Czech Republic), as previously described more in detail by Bukovská et al. (2018). Each such compartment was filled with 44 g of a sterilized 1:1 soil-sand mixture (v:v), identical in all treatments, where the soil was a mixture of equal portions of each of the eight soils used above. The substrate was amended with a mixture of the microbial washes described above. The root-free compartment had a hole at the top side and a pipette tip fixed in it to facilitate injection of isotopically labelled nutrients one week before the harvest.

At days 68 through 72 after planting, one replicate pot from each treatment (i.e., 24 pots per day) were labelled with $^{13}\text{CO}_2$ under a Plexiglass canopy as described previously (Slavíková et al. 2017), using 5 g of $\text{NaH } ^{13}\text{CO}_3$ (^{13}C enrichment > 99%, Cambridge isotope laboratories, Tewksbury MA, USA) for each labelling run. The pots were incubated in the enriched $^{13}\text{CO}_2$ atmosphere for 1 h. CO_2 concentration (measured by LI-820 gas monitor, LICOR, Lincoln NE, USA) inside the canopy reached approximately 950 ppm following $^{13}\text{CO}_2$ release (please note the sensitivity of this detector is different for $^{12}\text{CO}_2$ and $^{13}\text{CO}_2$ so these numbers remain only approximate), whereas it dropped below 400 ppm within 1 h of labelling. Thereafter, the canopy was removed, and P and N isotopes were injected into the root-free compartment of the previously ^{13}C labelled pot, ^{33}P as a carrier-free orthophosphate (Hartmann Analytics via Lacomel, Kralupy and Vltavou, Czechia) and ^{15}N as $^{15}\text{NH}_4\text{Cl}$, > 99% ^{15}N enrichment (Cambridge isotope laboratories). Each of the labelled pots received 2 mg of ^{15}N and 250 kBq of ^{33}P , mixed with 500 µL water and injected into the root-free compartment with a needle through the fixed pipette tip, subsequently washed by 1 mL of ultrapure water. During the 5 labelling campaigns extending over 5 days, five replicate pots per each treatment combination were isotopically labelled, whereas the sixth replicate served as unlabelled isotopic control.

Harvest and data collection

The plants were harvested after 75–79 days of growth, i.e. exactly seven days after the isotopic labelling of each respective pot. The isotopic control (unlabelled) pots were harvested at 79 days of growth. Shoots and roots were separated, and the roots were washed with tap water to remove adhering soil. After determining the fresh weight of the root systems, a weighed random subsample was cut into 1-cm segments and preserved in 50% ethanol for the determination

of root colonization. Substrate from the root-free compartment as well as a subsample of homogenized substrate from the pot (subsequently termed root zone soil) were dried at 65 °C to measure organic C and total N concentrations and isotopic composition.

Shoot and root dry weights were recorded after drying in an oven at 65 °C to constant weight and then milled using the Retsch MM200 device (Retsch GmbH, Haan, Germany). The dry weights of roots were corrected to total dry weights by adding extrapolated dry weights of the removed subsample. A subsample of 0.1 g was incinerated at 550 °C in a muffle furnace for 12 h and extracted by hot concentrated HNO_3 to determine the P concentration, employing the malachite green method (Ohno and Zibilske 1991). Radioactivity of ^{33}P in the same acid extracts (2 mL aliquots) was measured by β -scintillation counting (within decay energy window 2–300 keV), using PerkinElmer AB scintillation cocktail, combined with the samples in a ratio 5:1 (v:v, cocktail:extract). Background radioactivity in the acid extracts was estimated using analytical blanks (i.e., samples with no plant biomass input). All scintillation counting was carried out using TriCarb 2900TR Liquid Scintillation Analyzer (PerkinElmer, Shelton CT, USA), using automatic decay correction for the isotope activity date. The N concentration and ^{15}N enrichment in biomass and substrate samples was determined using a Flash EA 2000 elemental analyser (Thermo Fisher Scientific, Waltham, MA, USA). The N and P contents of shoots or roots were calculated by multiplying the concentration by the dry weight.

The ^{15}N partitioning in the system was calculated as fractions of the added ^{15}N appearing in shoots, roots, root zone soil, and the root-free compartment, considering the isotopic controls as the background. For ^{33}P , only the shoot and root fractions were measured and calculated (including the radioactive decay correction), as the soil fraction can be assumed to sum up to the added ^{33}P due to the low mobility of P in soil. Excess of ^{13}C in the system (i.e. the sum of ^{13}C originating from the labelling pulse in shoots, roots and soil samples) was calculated as described previously (Slavíková et al. 2017), considering the isotopic controls.

Subsequently, partitioning was calculated as fractions of the ^{13}C excess in shoots, roots and soil, the last being the sum of ^{13}C excess in root zone soil and the root-free compartment.

The root subsamples for the determination of root colonization were soaked in 10% KOH and stained with 0.05% of trypan blue in lactoglycerol (Koske and Gemma 1989). Frequency of root colonization was determined microscopically for each of the four main intraradical structures of AMF (hyphae, arbuscules, vesicles and coils) using the magnified intersection method (McGonigle et al. 1990); 100 intersections per sample within 30 root segments were recorded at 100× magnification (Olympus BX60).

Communities of AMF in the root samples were characterized as described previously (Bukovská et al. 2021). Briefly, DNA was extracted from a subsample of milled dried roots (10 mg of powder) using the Plant DNeasy kit (Qiagen) according to manufacturer's instructions. Partial SSU of rDNA was amplified using WANDA (Dumbrell et al. 2011) and AML2 (Lee et al. 2008) primers. The AMF-specific AML2 primer is reasonably specific and inclusive for AMF (van Geel et al. 2014). On the other hand, it may miss rare genera as compared to some primers targeting the LSU of rDNA (Řezáčová et al. 2016). The amplicon pools were sequenced and data bioinformatically processed as in Dudáš et al. (2022). Representative sequences of operational taxonomic units (97% similarity) were blasted against EUKARYOME database (Eukaryome.org, version 1.9.2, accessed December 18, 2024; Tedersoo et al. 2024), see Table S2. In order to facilitate interpretation of the encountered patterns, OTUs were pooled to families according to the taxonomy described by Hyde et al. (2024) including the separation of *Glomeraceae* into four families proposed by da Silva et al. (2024). Sequence data were deposited in the Sequence Read Archive (NCBI) under accession number PRJNA935258.

AMF abundance in soil was evaluated using SYBR-green based quantitative PCR with primers NS31-AML2 as described previously (Bukovská et al. 2021), following DNA extraction from soil samples using DNeasy Power-Soil kit (Qiagen, Venlo, the Netherlands). The results were expressed as copy numbers of rDNA per mg of soil. From the measured values, mean value of the respective non-mycorrhizal control was subtracted (gross average for root zone soil: 1716 rDNA copies per mg, for the root-free compartment: 653 rDNA copies per mg). Occasionally, when the measured value in the mycorrhizal pot was lower than the average of the respective non-mycorrhizal treatment, the value for the mycorrhizal pot was set at zero.

Statistical analysis

To show the relationship of plant performance with the experimental factors and their interactions, redundancy analyses (RDA) were computed with 11 basic plant parameters as response variables (shoot and root dry weights, N and P concentrations in shoots and roots, N and P contents of shoots and roots, N:P ratio in shoots). The experimental factors (Inoculation treatment, Soil provenance, Site, the first one including 3 levels, i.e., living inoculum from arable field, living inoculum from grassland, and non-mycorrhizal control) were entered as explanatory variables alone or together with their two-way interactions. Partial RDA was used to determine the effect of each experimental factor and interaction.

To directly compare how grassland AMF and arable-field AMF influenced the plant parameters, responses

to mycorrhization were calculated using the equation $R = \log(M/NM)$, where M is the individual value of a parameter in a replicate mycorrhizal plant and NM is the mean value of the parameter in the corresponding non-mycorrhizal control treatment. The responses to mycorrhization were calculated for the 11 basic plant parameters as well as for the isotopic fractions. Differences in responses to inocula of the two provenances (grassland vs. arable field) were tested by generalized linear mixed model with Site as random factor. Hence, the models included Inoculum provenance, Soil provenance and their interactions as fixed factors, while Site and Harvest day (day 75 to 79 after planting) were included as random factors (package glmmTMB in R) (Brooks et al. 2017). The assumptions of ANOVA on normality and homogeneity of variance were tested using Shapiro–Wilk test and Levene's test, respectively. When the model did not meet the homogeneity of variance, the source of heterogeneity from the fixed factors was corrected by including it in a new model using generalised least squares method (GLS, package nlme in R) (Pinheiro et al. 2021).

RDA and partial RDA were also used to evaluate the effects of the experimental factors and their interactions on the composition of the communities of AMF. To determine which families were associated with a specific Inoculum provenance, indicator species analysis (Dufrene and Legendre 1997) was applied as implemented by the function `indval()` in the R package `labdsv` (Roberts 2023). Effect of Inoculum provenance on community richness (i.e. number of families per sample) and Shannon diversity (Shannon 1948) was tested by a linear model as described above. The same procedure was applied to test the effect of Inoculum provenance on the root colonization parameters, which were arcsine-transformed prior to the analyses, and on rDNA copy numbers in the substrate, which were logarithmically transformed. Pearson correlation was employed to test relationships between variables.

RDA and partial RDA were carried out in Canoco 5 (Ter Braak and Šmilauer 2012), while all the other analyses were performed in R (R Core Team et al. 2020).

Results

Plant performance in the experimental system

Plant performance was significantly affected by all three experimental factors according to RDA, whereby the proportions of variation explained by Site and Inoculation treatment were comparable (22.4% and 23.4%, respectively), and one order of magnitude higher than that by Soil provenance (2.2%). Considering the two-way interactions, Site × Inoculation treatment explained the highest proportion of variation (25.7%), followed by the Site × Soil

provenance interaction (6.7%), whereas the interaction Soil provenance $\times$ Inoculation treatment had no significant effect (Table S4). The share of 46.9% of variation remained unexplained by the three factors and the most important Site $\times$ Inoculation interaction (Fig. 1).

The effect of Inoculation treatment was mainly based on differences between non-mycorrhizal and mycorrhizal plants, while the interaction Site $\times$ Inoculation treatment reflected differences between non-mycorrhizal plants from the Enc site on one hand and those from the other sites on the other hand (Fig. 1; Online resource 3: Table S3). Across all sites, non-mycorrhizal status of plants was related to smaller shoot and root dry weights, which was most pronounced for non-mycorrhizal plants growing in the soils from the Enc site. Non-mycorrhizal status was also related to higher N:P ratios in shoots with highest values found in plants grown in Enc soils. Mycorrhizal plants, in contrast, were more homogenous among the sites (Fig. 1, Table S3).

Differences between grassland and arable-field AMF communities

The variation in the composition of the root-colonizing communities of AMF was mainly explained by Inoculum provenance (10.7%) and Site (5.8%), while Soil provenance had no significant effect according to RDA (Table S5). From the two-way interactions, only Site $\times$ Inoculum provenance explained a significant proportion of variation (10.2%), the interaction consisted in more differentiated grassland communities among the sites as compared to the arable-field communities (Fig. 2). On the other hand, 77% of variation remained unexplained by the three factors and the most important Site $\times$ Inoculation interaction (Fig. 2). Out of the three most abundant families (*Sclerocystaceae*, *Dominikiaceae*, *Septoglomeraceae*), *Dominikiaceae* was indicative family for grasslands and *Sclerocystaceae* for arable-fields (Table 1, Fig. S2). The communities established from grassland AMF inocula had further four indicative families with low relative abundances (< 3% in the whole data

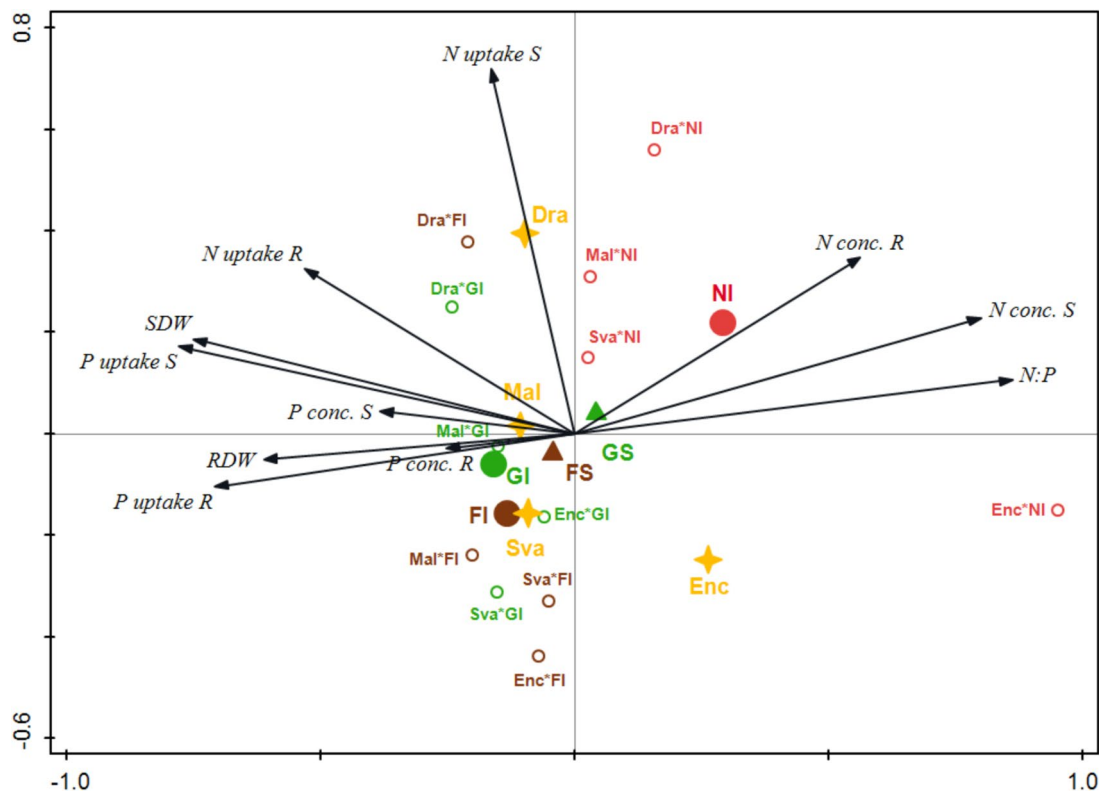


Fig. 1 Relationship between plant performance and the experimental factors including the two-way interaction of Site $\times$ Inoculation treatment according to a redundancy analyses (RDA). The model (pseudo- $F=14.4$, $p<0.002$) accounted for 53.1% of variation in the data (38.7% by the first axis, 8.1% by the second axis). The experimental factors were: Site (Dra, Mal, Sva, Enc—yellow crosses), Soil provenance (FS pots filled with soil from arable field, GS pots filled with soil from grassland—triangles) and Inoculation treatment (NI non-

inoculated, GI inoculated with grassland inocula, FI inoculated with arable-field inocula—circles). The parameters used to characterize plant performance were: shoot and root dry weights (SDW, RDW), N and P concentrations (N conc., P conc.) in shoots (S) and roots (R), N and P contents (N uptake, P uptake) in S and R, the N:P ratio in shoots (N:P). Small empty circles indicate the interactive effects of Site $\times$ Inoculation treatment

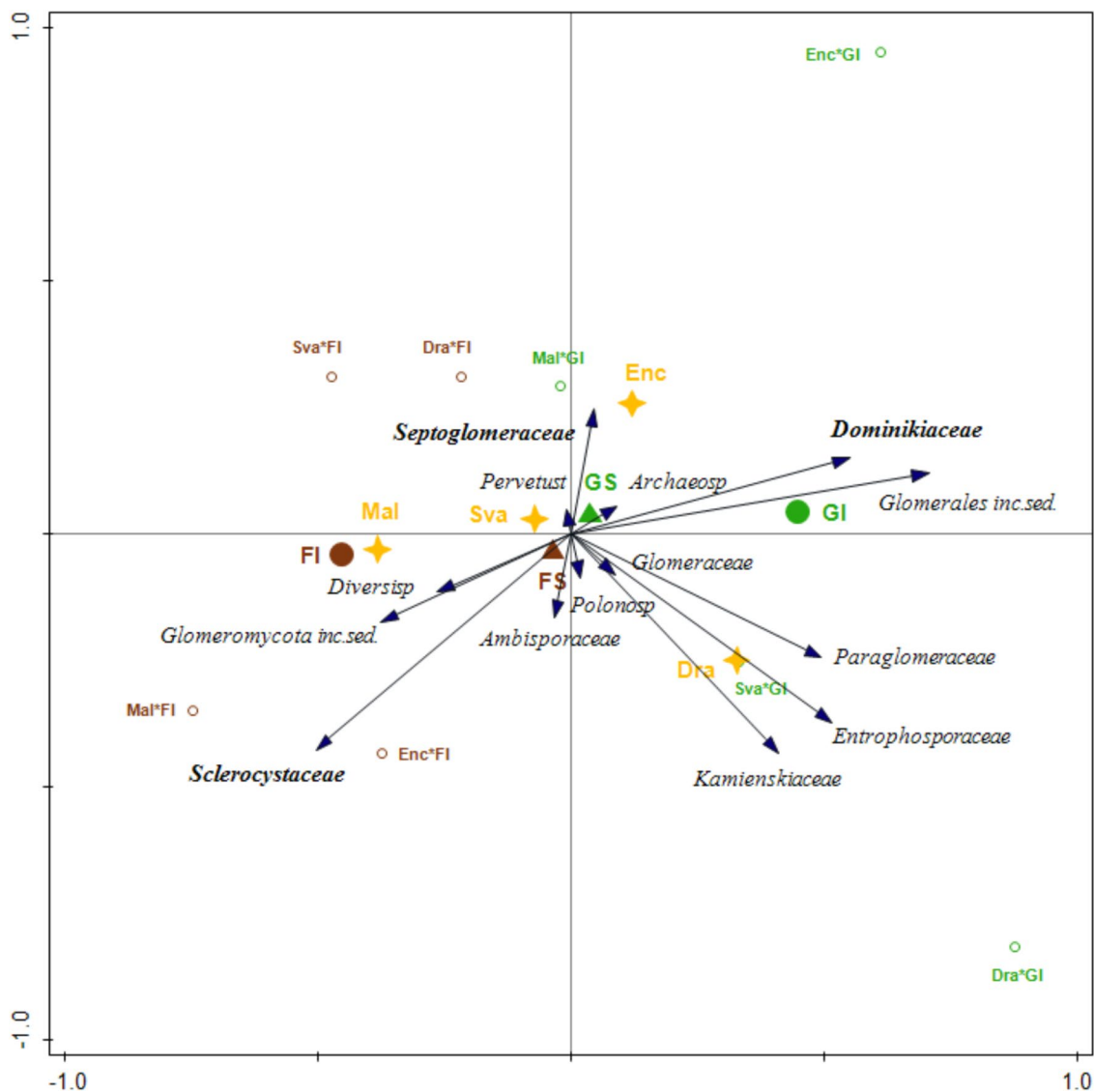


Fig. 2 Relationship between the relative abundances of arbuscular mycorrhizal fungal genera and the experimental factors according to a redundancy analyses (RDA) including the two-way interaction of Site $\times$ Inoculum provenance. The model (pseudo- $F=4.5$, $p=0.002$) accounted for 23.0% of variation in the data (14.1% by the first axis, 5.4% by the second axis). The experimental factors were: Site (Dra, Mal, Sva, Enc—yellow crosses), Soil provenance (FS pots filled with

soil from arable field, GS pots filled with soil from grassland—triangles) and Inoculum provenance (GI inoculated with grassland inocula, FI inoculated with arable-field inocula—circles). Small empty circles indicate the interactive effects of Site $\times$ Inoculum treatment. The three genera with highest relative abundances (> 20%) are marked in bold. Names of families Archaeosporaceae, Diversisporaceae, Polonosporaceae and Pervetustaceae are abbreviated

set), including the group of OTUs not assigned to any family within Glomerales (Glomerales inc. sed.). Only one low-abundance indicative genus was identified for the field AMF-communities, together with the OTUs that could not be assigned to any family within Glomeromycota (Glomeromycota inc. sed.) (Table 1). Consistently, the taxon richness of the communities was significantly higher for the pots added with grassland inocula than for arable-field inocula due to higher number of low-abundance genera (Table 2).

Root colonization by AMF was overall higher in soils from the Enc site than from the other sites (Table S3).

Across all four sites, the frequencies of hyphae, arbuscules and coils were significantly affected by Soil provenance, with higher values recorded in grassland soils than in field soils. Inoculum provenance only significantly affected the frequency of coils, which was higher in roots colonized by grassland AMF (Table 2). No root colonization by AMF was detected in the roots of the non-inoculated plants.

AMF absolute abundances in the root zone soil and the root-free compartment were also overall the highest in soils from the Enc site (Table S3) and they were significantly correlated ($r=0.306$, $t=3.12$, $p=0.002$).

Table 1 Relative abundances of families of arbuscular mycorrhizal fungi in communities initiated with arable-field inocula or grassland inocula

Family	Relative abundance in samples (%)		Indicator value (p-value)
	Field inoculum	Grassland inoculum	
<i>Sclerocystaceae</i>	57.38 (4.64)	22.81 (3.63)	0.640 (<0.001) ^F
<i>Dominikiaceae</i>	26.72 (4.19)	46.20 (4.20)	0.594 (<0.001) ^G
<i>Septoglomeraceae</i>	14.42 (3.57)	21.63 (3.56)	
<i>Kamienskiaceae</i>	0.77 (0.30)	3.47 (1.05)	0.548 (<0.001) ^G
<i>Entrophosporaceae</i>	0.09 (0.04)	1.41 (0.54)	0.571 (<0.001) ^G
<i>Paraglomeraceae</i>	0.01 (0.01)	0.27 (0.11)	0.428 (<0.001) ^G
<i>Diversisporaceae</i>	0.11 (0.05)	0	0.271 (<0.001) ^F
<i>Glomeraceae</i>	0.02 (0.01)	0.06 (0.03)	
<i>Pervetustaceae</i>	0	0.04 (0.04)	
<i>Archaeosporaceae</i>	0.02 (0.01)	0.02 (0.01)	
<i>Ambisporaceae</i>	0.01 (0.01)	<0.01 (<0.01)	
<i>Polonosporaceae</i>	<0.01 (<0.01)	<0.01 (<0.01)	
Glomerales inc. sed	0.36 (0.15)	4.08 (0.58)	0.740 (<0.001) ^G
Glomeromycota inc. sed	0.08 (0.03)	0.01 (<0.01)	0.346 (0.002) ^F

Values in the first two columns are means (SE) pooled across the other two experimental treatments (Site, Soil provenance). ^G indicative for grassland community, ^F indicative for arable-field community

Table 2 Root colonization of the experimental plants by hyphae (Hy%), arbuscules (Ar%), vesicles (Ve%) and coils (Co%) as well as the number of families detected per sample (Richness) and Shannon index (Shannon), effects of the experimental factors Soil provenance (Soil), Inoculum provenance (Inoculum), and their interaction

Soil	Inoculum	Hy (%)	Ar (%)	Ve (%)	Co (%)	Richness	Shannon
Field	Field	39.5 (3.5)	32.4 (3.2)	12.1 (1.7)	17.6 (2.6)	5.04 (0.30)	0.60 (0.07)
Field	Grassland	39.9 (3.7)	31.5 (2.8)	6.9 (1.6)	26.2 (3.0)	6.21 (0.26)	0.98 (0.06)
Grassland	Field	54.5 (4.7)	46.1 (4.7)	17.1 (3.5)	28.5 (2.5)	5.37 (0.32)	0.62 (0.06)
Grassland	Grassland	47.1 (4.0)	37.1 (3.3)	7.2 (1.5)	36.3 (3.3)	6.29 (0.23)	0.82 (0.07)
	df	X^2 (p-value)					
Soil (A)	1	9.947 (0.002)	5.843 (0.016)	1.686 (0.194)	14.025 (<0.001)	n.i	n.i
Inoculum (B)	1	0.008 (0.930)	0.030 (0.862)	2.715 (0.099)	8.561 (0.003)	14.293 (>0.001)	20.213 (>0.001)
A x B	1	1.349 (0.245)	1.298 (0.255)	1.168 (0.280)	n.i	n.i	n.i

Values in the upper part of the table are means (SE) across all sites. The effects of the experimental factors (X^2 =Chi-square value) are given according to linear models fitted using generalised linear mixed (GLMM) method or generalised least squares (GLS) method to correct for heterogeneity of variances, which included Site and Harvest day as random variables. Significant effects of the factors are highlighted in bold, n.i. factors not included into the final model

Furthermore, the AMF abundances in substrate were correlated with the frequencies of hyphae in roots ($r=0.247/0.284$, $t=2.476/2.875$, $p=0.015/0.005$, for the root zone soil/root-free compartment, respectively). The abundances tended to be higher in grassland soils than in field soils, particularly for grassland AMF, but the only significant effect was the interactive effect of Soil provenance and Inoculum provenance in the root-free compartment (Table S6).

Differential responses of plants to grassland and arable-field AMF inocula

Consistently with the RDA results on plant performance (Fig. 1), the most pronounced effects of mycorrhization were increased biomass and P uptake on one hand, and decreased N shoot concentration on the other hand (Fig. 3a). The size of mycorrhizal effects significantly differed between the grassland and arable-field inocula for

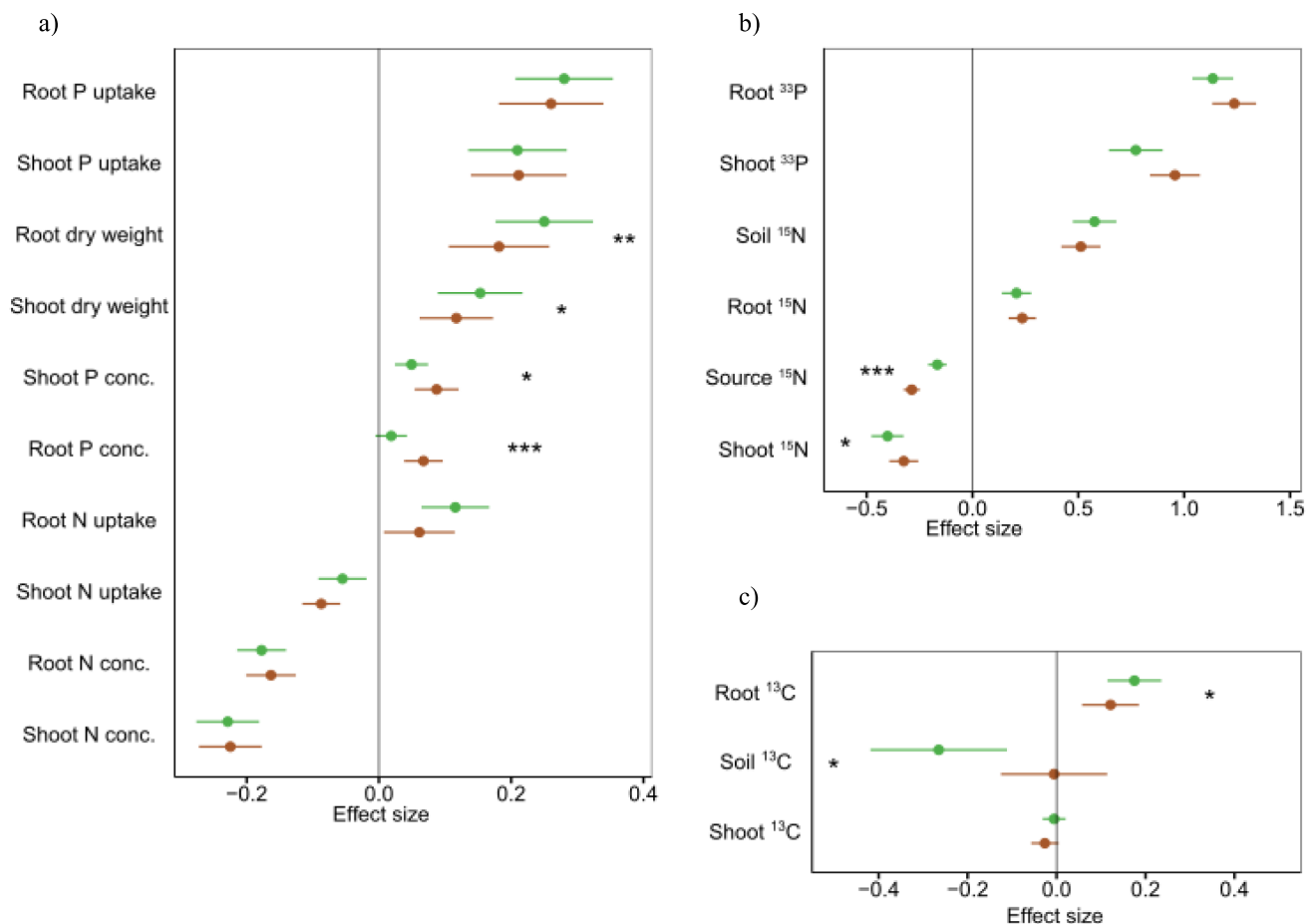


Fig. 3 Effects of inoculation with arbuscular mycorrhizal fungi from grasslands (green symbols) and arable fields (brown symbols) on plant growth, nitrogen (N) and phosphorus (P) concentrations (conc.) and contents (uptake) in plant biomass (a), partitioning of ^{15}N and ^{33}P provided in a root-free compartment (Source) (b), partitioning of photosynthetically fixed ^{13}C in the soil–plant system (c). The points

are means, horizontal lines 95% confidence intervals as calculated across the experimental factors Site and Soil provenance. The vertical lines (0) mark no effects, asterisks show significant difference between the two types inocula as determined by linear models given in Table 3. * $0.05 > p \geq 0.01$, ** $0.01 > p \geq 0.001$, *** $p < 0.001$

growth and P concentrations, while Inoculum provenance did not significantly affect the plant responses in P contents, N concentrations or contents (Table 3).

The positive mycorrhizal growth responses were significantly higher with grassland inocula than with arable-field inocula (Table 3, Fig. 3a). Considering variability among the four sites characterized by differential performance of non-mycorrhizal plants (see Fig. 1), mycorrhizal growth response was clearly related to the shoot growth and the N:P ratio of non-mycorrhizal plants (Fig. S3).

The increases of P concentrations in shoots and roots were significantly higher with arable-field inocula than with grassland inocula, whereas no relationship was found of the difference with soil conditions, as approximated by the growth or N:P ratio of non-mycorrhizal plants (analyses not shown).

P, N and C partitioning

Mycorrhization had a large positive effect on plant ^{33}P uptake from the root-free compartment (Fig. 3b) owing to negligible uptake of ^{33}P by non-mycorrhizal plants (Table S3). Nevertheless, grassland and field AMF did not significantly differ in their effects, while mycorrhizal effects on ^{33}P uptake into roots were significantly higher in field soil than in grassland soil (Table 3). The mycorrhizal effects on plant ^{33}P uptake were not overall correlated with AMF abundance in root zone soil or the root-free compartment (analyses not shown).

Mycorrhizal effects were more complex in the case of ^{15}N : Mycorrhization generally increased ^{15}N uptake into roots but decreased ^{15}N uptake (transfer) into shoots. At the same time, mycorrhization decreased the ^{15}N fraction

Table 3 Effects of the experimental factors Soil provenance (Soil) and Inoculum provenance (Inoculum) on the responses to mycorrhization in plant growth, nitrogen (N) and phosphorus (P) concentrations and contents in shoot and roots, partitioning of ^{15}N and ^{33}P provided in a root-free compartment (source), partitioning of photosynthetically fixed ^{13}C in the soil–plant system

Response to mycorrhization	Soil (A)		Inoculum (B)		A × B	
	X ²	p-value	X ²	p-value	X ²	p-value
Shoot dry weight	6.295	0.012	7.220	0.007	n.i	
Root dry weight	1.380	0.240	5.494	0.019	n.i	
Shoot N concentration	0.102	0.750	0.046	0.830	n.i	
Shoot P concentration	0.189	0.664	5.166	0.023	n.i	
Root N concentration	0.535	0.465	0.187	0.665	n.i	
Root P concentration	6.884	0.009	12.358	< 0.001	n.i	
Shoot N content	0.936	0.333	0.068	0.795	2.282	0.131
Shoot P content	5.027	0.025	0.015	0.904	n.i	
Root N content	0.089	0.766	0.425	0.515	2.082	0.149
Root P content	0.036	0.849	0.612	0.434	n.i	
^{15}N in shoots	19.514	< 0.001	3.887	0.049	n.i	
^{15}N in roots	0.286	0.593	0.304	0.581	0.145	0.703
^{15}N in soil	0.310	0.578	0.118	0.731	0.231	0.631
^{15}N in source			18.376	< 0.001	n.i	
^{33}P in shoots	2.039	0.153	0.986	0.321	1.082	0.298
^{33}P in roots	15.320	< 0.001	2.551	0.110	n.i	
^{13}C in shoots	6.182	0.013	3.498	0.061	n.i	
^{13}C in roots	0.014	0.903	3.945	0.047	n.i	
^{13}C in soil	0.266	0.606	6.226	0.013	n.i	

The effects of the experimental factors (X^2 = Chi-square value) are given according to linear models fitted using generalised linear mixed (GLMM) method or generalised least squares (GLS) method to correct for heterogeneity of variances, which included Site and Harvest day as random variables. Significant effects of the factors are highlighted in bold, n.i. factors not included into the final model

that remained in the root-free compartment and increased that in the root zone soil (Fig. 3b, Table S3). Consequently, larger ^{15}N fractions were accumulated in root zone soil and

roots of mycorrhizal plants, while non-mycorrhizal plants had larger ^{15}N fractions in shoots and the root-free compartments (Fig. 4).

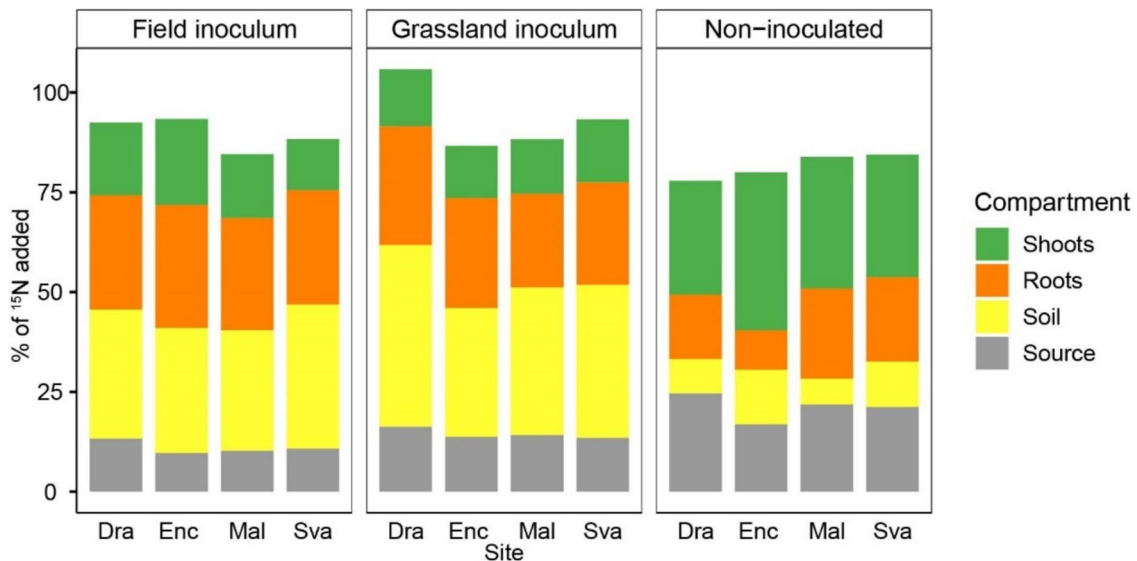


Fig. 4 Partitioning of ^{15}N into shoots, roots, the root zone soil (Soil) and the root-free compartment (Source). The columns are means per combinations of Site and Inoculation treatment, i.e. the data were pooled across the Soil provenance factor

These general mycorrhizal effects were partly modulated by Inoculum provenance: The ^{15}N decrease in the root-free compartment was significantly higher with arable-field inocula than with grassland inocula, meaning that field AMF faster depleted ^{15}N from the source than grassland AMF (Fig. 3b, Table 3). The decrease was, counterintuitively, overall negatively correlated with AMF absolute abundance in the root-free compartment ($r=0.271$, $t=2.482$, $p=0.015$), and this relationship was due to differences in AMF abundance between grassland and arable-soil inocula (Inoculum provenance $F=17.100$, $p<0.001$, AMF abundance $F=0.711$, $p=0.401$ in a model with both predictors of the ^{15}N decrease).

No significant effect of Inoculum provenance was found for the mycorrhizal effects on ^{15}N partitioning into root zone soil and roots and the effects were unrelated to AMF abundance in the corresponding compartments (root zone soil or root, analyses not shown). On the other hand, the decrease of the ^{15}N fraction in shoots upon mycorrhization was significantly higher with grassland inocula than with arable-field inocula. Additionally, this decrease was significantly higher in grassland soil than in field soil (Table 3, Table S3).

Mycorrhization had no effect on ^{13}C partitioning into shoots (Fig. 3c), but it clearly enhanced ^{13}C partitioning into roots, whereby the effect was significantly higher with grassland AMF than with arable-field AMF (Fig. 3c, Table 3). The proportion of ^{13}C transferred from plants into soil was highly variable depending on site, with one order of magnitude higher values for the soils from Enc site as compared to Dra site (Table S3). It was significantly decreased by mycorrhization with grassland AMF, but not with arable-soil AMF across all sites (Fig. 3c, Table 3). There was a significant correlation of the ^{13}C released to soil by mycorrhizal plants and absolute AMF abundance in soil ($r=0.36$, $t=3.412$, $p=0.001$).

Discussion

AMF from arable fields and grasslands similarly affected the outcome of the symbiosis along the mutualism-parasitism continuum (Johnson 2010) at the gradient of conditions represented by grassland and field soils from the four different sites. Thus, the principal responses of the host plants to mycorrhization were similar regardless of the Inoculum provenance. However, the size of some responses differed between the grassland and arable-field inocula across all sites, which enables to infer differential functioning of the mycorrhizas and partly relate them to the characteristics of the AMF communities.

Functioning of mycorrhizas sourced from differently managed soils

In concordance with our first hypothesis, positive mycorrhizal growth response, as a proxy for plant net benefits from mycorrhiza, was lower with arable-soil inocula than with grassland inocula. This is in line with several previous studies, where plants either produced less biomass with AMF from intensively managed agricultural soils or the provenance of inoculum played no role (Eason et al. 1999; García de Leon et al. 2020; Gottshall et al. 2017; Johnson 1993; Marro et al. 2020; Ryan and Graham 2018; Scullion et al. 1998). Our finding, on a gradient of mycorrhizal growth responses ranging from highly positive to slightly negative, corroborates this pattern as robust. On the other hand, mycorrhizal effects on P uptake of the experimental plants did not depend on Inoculum provenance, thus contradicting our second hypothesis and demonstrating that the differential effects on plant growth can be independent of P supply. In contrast to the study of Köhl et al. (2014), higher P uptake with AMF inocula from less disturbed conditions was not even recorded in soils from Enc site, where non-mycorrhizal plants were strongly P-limited (N:P ratio in shoots >30 , Table S3; Koerselman and Meuleman 1996). Köhl et al. (2014) related higher P uptake with AMF from no-till plots to higher density of extraradical mycelia. However, our experiment did not find any overall differences in the soil abundance of grassland and arable-soil AMF or relationship between plant P uptake and abundance of AMF in soil. This might be due to different methodical approach employed to quantification of AMF in soil by Köhl et al. (2014) and in our study—microscopically determined hyphal length by the former, quantification of rDNA copy number in our case. Hyphal measurement quantifies absorptive structures only, but does not distinguish between living and dead hyphae. On the other hand, quantification of rDNA copy numbers is more accurate and less laborious, but does not distinguish between abundance of hyphae and spores.

In parallel with the smaller growth response induced, arable-soil inocula increased P concentration in shoots more than grassland inocula, which is consistent with a recent study of Marrassini et al. (2024): They reported AMF isolates from arable soils to less promote plant growth but to induce higher concentrations of various nutrients in plant biomass, as compared to isolates from a less disturbed grassland. Increased nutrient contents along with decreased biomass may indicate luxurious consumption, meaning, in our experiment, that the P taken up could not be fully utilized for higher biomass production (Chapin 1980). This could be due to limitation by some other nutrient, being N the most probable candidate, as it has been identified as a significant determinant of mycorrhizal growth responses (Hoeksema et al. 2010). Furthermore, according to the trade balance

model (Johnson 2010), the growth of mycorrhizal plants could be restricted by C limitation, i.e. by higher C drain to arable-field AMF than to grassland AMF in our experiment.

The shoot N:P ratio of mycorrhizal plants, which ranged between 5.9 and 10.6, independently of the N:P status of non-mycorrhizal plants (Table S3), indeed indicates that the growth of all mycorrhizal plants was limited by N (Koerselman and Meuleman 1996). The N limitation can be attributed not only to improved P uptake, but also to decreased N supply into plant shoots upon mycorrhization because the N concentration and content in the shoots of mycorrhizal plants was lower than in non-mycorrhizal plants (Fig. 3b). While N was taken up by the fungal hyphae and also translocated into plant roots, as evidenced by the increased ^{15}N partitioning into roots of mycorrhizal plants, translocation into plant shoots was clearly restricted in comparison with non-mycorrhizal plants (Fig. 3b). This strongly suggests that N was immobilized within the hyphae of AMF, consistently with previous reports (Boussageon et al. 2022; Hodge and Fitter 2010; Ingraffia et al. 2020; Püschel et al. 2016; Treseder and Allen 2002). As summarized by Correa et al. (2015), AMF may provide plants only with N, which is in excess, because they act as a strong N sink. The increased ^{15}N content in mycorrhizal roots, attributable to ^{15}N immobilization in the intraradical AMF structures, is supportive to this assumption. Unique evidence, however, is provided by the increased ^{15}N contents in the root zone soil of mycorrhizal plants: if this soil ^{15}N fraction had not been immobilised (in the fungal mycelia), the N-deficient mycorrhizal plant would have taken it up and transferred into shoots (which was the case in non-mycorrhizal plants).

Mycorrhizal effects on N uptake did not indicate more pronounced N limitation of mycorrhizal plants with arable-soil inocula than with grassland inocula. In contrast, the differential effects of the inocula on ^{15}N partitioning revealed higher rates of ^{15}N transferred into shoots in conjunction with more pronounced depletion of ^{15}N from the source by arable-soil AMF than by grassland AMF. The ^{15}N partitioning, determined relatively shortly after application of ^{15}N also reflects the speed of the N flow. Taxa of AMF may differ in how fast they transfer nutrients from a localized source to their host plants, either due to differences in foraging speed (Šmilauer et al. 2020; Thonar et al. 2011) and/or due to transient nutrient immobilization in their mycelia (Boddington and Dodd 1999; Thonar et al. 2011). Thus, it seems that AMF from arable fields were faster at transferring N from soil to plant, but equally efficient N suppliers in the long-term.

C limitation of plants associated with arable-soil inocula, as a second factor potentially responsible for the lower net benefits, is indicated by the significantly higher ^{13}C fraction transferred into the root zone soil of plants associated with arable-soil inocula as compared to grassland inocula. This

principal difference is remarkable as it was consistent across highly variable proportions of ^{13}C transferred. Lower ^{13}C drain into soil with grassland AMF than without mycorrhiza is in accordance with well-documented decrease of rhizodeposition upon mycorrhization, particularly in P-deficient conditions (Marschner et al. 1997; Nazeri et al. 2014; Ryan et al. 2012). Comparable ^{13}C drain with arable-soil AMF as without mycorrhiza could be either due to poor alleviation of P stress by the AMF (which is contradicted by the P biomass data in our experiment) or due to higher C flow into the extraradical structures of arable-soil AMF as compared to grassland AMF, which counterbalanced the decreased root exudation. Distinguishing between the contribution of AMF and roots to the C release into soil would require a more detailed investigation with an experimental set up better separating roots from AMF hyphae.

Interestingly, growth limitation of mycorrhizal plants by high C costs has been previously proposed for communities of AMF from less intensive land-use types than conventional agricultural systems because of larger extraradical mycelia (Köhl et al. 2014; Verbruggen et al. 2012). Our results indeed confirmed higher ^{13}C partitioning into roots and soil at higher fungal abundance, in accordance with previous reports (Lendenmann et al. 2011; Lerat et al. 2003), even though the relationship was relatively weak and only evident across all soils.

Functional differentiation due to AMF or other microbes?

An inherent problem associated with experiments including different AMF inoculation treatments and non-inoculated controls is the impossibility to create equal communities of other microorganisms than AMF in the different treatments (Gryndler et al. 2018). This also means that the re-introduced microbial communities (via inoculum or microbial filtrate) are not identical with the original background. In that, interpretation of effects of AMF or their communities is always loaded with uncertainty as to which extent they can be attributed to the AMF themselves or to other soil microorganisms, or whether the established microbial conditions of the soil are representative. It is known that other soil microorganisms than AMF also may affect nutrient uptake or root exudation by plants (Canarini et al. 2019; Philippot et al. 2013; Sasse et al. 2018), even interactively with mycorrhiza (Zhang et al. 2022).

In our experiment, we have reintroduced to each previously sterilized soil its own microbiota by the application of a microbial wash, and the inoculation treatments were created by addition of non-sterile soil. Consequently, some treatments received combination of both types of microorganisms (e.g. arable-field inoculum in grassland soil), while other received only one type (e.g. non-mycorrhizal

plants in grassland soil). If other soil microbiota than AMF were the dominant drivers of the observed effects, we would expect interactive effects of the factors Inoculum provenance and Soil provenance, which were overall non-significant (Table 3). The analyses of Gryndler et al. (2018) also suggest that soil microbial communities (other than AMF) added into soils by different approaches create compositionally different, but functionally similar communities possibly due to functional redundancy. Still, we cannot exclude that specific differential effects of arable-field and grassland inocula, e.g. ^{13}C release to soil, were partly due to differences in other microbes than AMF. This cannot be excluded even by significant correlations of the mycorrhizal effects with fungal abundance, as bacteria also colonize surfaces of fungal hyphae (Emmett et al. 2021; Faghihinia et al. 2023; Wang et al. 2024).

Different communities of AMF in arable-soils and grasslands

To summarize the differential plant responses to the two types of inocula, arable-soil inocula 1) conferred lower net benefits to plants, 2) were faster in foraging and supplying N, but not more efficient in the total supply of N or P, and 3) induced higher C drain into soil as compared to grassland inocula. Are these differences in line with the assumption that arable soils selected for more ruderal AMF (Verbruggen et al. 2010)? The trait-based framework assumes that ruderal AMF have high growth rates and fast hyphal turnover (Chagnon et al. 2013), which would enable faster nutrient foraging due to faster access to nutrient patches. Also, Šmilauer et al. (2021) described increasing foraging speed of AMF communities in the order field-established < greenhouse-cultivated < greenhouse-cultivated and fertilized, consistently with the assumption that disturbance and higher nutrient inputs/availabilities select for AMF with higher foraging speed. In the same line, readily available mineral fertilizers, detrimental for disturbance sensitive AMF with high demands of plant C for foraging nutrients via extensive extraradical mycelia (Chagnon et al. 2021; Konvalinková et al. 2017), can select for faster spore production rather than for soil mycelium for survival over time (Verbruggen et al. 2015).

The functional differentiation of the indigenous mycorrhizas is interesting having in mind that communities of AMF that develop during experimental greenhouse cultivation usually contain only a subset of the species from the field site (Chung et al. 2019; Šmilauer et al. 2021; Sýkorová et al. 2007), particularly if they originate from less disturbed habitats and are cultivated only for short periods of time (Oehl et al. 2009). Consequently, the functioning of mycorrhizas with our experimental communities is based on a subset of the most disturbance-tolerant members and fastest

root colonizers from the original communities (although we exercised all possible care to minimize disturbance to the AMF upon sampling and pot establishment). In that, it is probable that the differentiation is underestimated since the original grassland communities probably contained higher proportions of taxa of other life-history strategies (such as competitors or stress-tolerators) with potentially different functional traits than the arable-field communities (Chagnon et al. 2013; Verbruggen et al. 2015).

On the other hand, it is remarkable to encounter functional differentiation within the subset of relatively disturbance tolerant fast root-colonizers. As root colonization level was not significantly different between grassland and arable-soil inocula, the functional differentiation was not related to this factor, previously reported as important for mycorrhizal benefits (Blažková et al. 2021; Lekberg and Koide 2005). While we cannot rule out that it was partly due to other microbiota associated with the inocula (as outlined above), it corresponded to a compositional differentiation of the AMF communities, which is in accordance with our third hypothesis. The difference mainly consisted in contrasting relative abundances of two dominant families, *Sclerocystaceae* (more than twice as high in plants inoculated with arable-soil AMF than with grassland AMF) and *Dominikiaceae* spp. (the opposite pattern). *Sclerocystaceae* family was dominated by OTUs classified as genus *Rhizoglossus* (more than 98% of reads, see Table S2). *Rhizoglossus intraradices* or *R. irregulare*, two prominent representatives of *Rhizoglossus* genus, are classified as generalist species (Oehl et al. 2010; Sýkorová et al. 2007), abundant in arable soils (Jansa et al. 2002, 2014; Manoharan et al. 2017; Pellegrino et al. 2012; Verbruggen et al. 2010) and tolerant to tillage (Jansa et al. 2003; Köhl et al. 2014). Less is known about the autecology of species within the relatively recently described (Błaszowski et al. 2015a, b) genus *Dominikia*, which dominated *Dominikiaceae* family in our root samples (more than 97% of reads, Table S2). Their morphology and trap cultivation suggest that they are fast root colonizers and persistent AMF owing to intraradical sporulation, while their extraradical structures may be more susceptible to adverse conditions (Błaszowski et al. 2018). They can be relatively abundant across different agricultural managements (Säle et al. 2015), but were also reported sensitive to tillage, fertilization and drought stress (Baltruschat et al. 2019; Müller et al. 2019).

This suggests that communities from the two land management types differentiate in the relative abundance of fast root colonizers, which differ in sensitivity to disturbance: The more disturbance-tolerant *Rhizoglossus* spp. dominate in the regularly tilled fields, the more sensitive *Dominikia* spp. in the minimally disturbed grassland soils. High C cost of mycorrhiza with a fast colonizer such as *R. irregulare* does not necessarily decrease the plant net benefit from mycorrhiza, particularly if high relative abundance of this AMF

enables early mycorrhiza establishment and improvement of the host plant's P nutrition (Blažková et al. 2021; Buil et al. 2023). However, our experiment suggests that early establishment of mycorrhiza can also be provided by other fast root colonizers (*Dominikia* spp.) at lower costs within the less disturbed communities. Three *Dominikia* species (*D. duoreactiva*, *D. difficilevidera* and *D. distichia*) have been identified as forming intraradical coils in the roots of *P. lanceolata* (Błaszowski et al. 2015a, b), similar to the structures observed in our model plants. This finding aligns with the prevalence of *Dominikiaceae* family and the higher percentages of coils encountered with the grassland inocula compared to the arable-field inocula. As arbuscules and coils may have different functions within arbuscular mycorrhiza (Perotto and Balestrini 2024), it would be valuable to investigate whether differences in the structure of root colonization might have measurable effects on plant performance for a deeper understanding of the functioning of native AMF communities.

Conclusions

Our functional analysis suggests that the selection pressures of conventional agricultural management promote mycorrhizas, which are equally efficient in providing P and N, but have higher requirements for plant-derived C. Even though we cannot fully exclude contribution of other microbiota than AMF, our cross-inoculation approach suggests that the effect can be primarily attributed to AMF. This implies that mutualistic functioning of mycorrhizas by arable-soil AMF could be more sensitive to factors that may decrease C availability to plants such as limited efficiency of photosynthesis or enhanced root exudation. While the functional differences between the two types of communities were relatively small in our experiment with one ruderal host plant and stable environmental conditions, they may become more pronounced in different contexts. For example, slower-growing competitive or stress-tolerant plant species can be more susceptible to higher C demand by mycorrhizas, as indicated by the preferential association of plants and AMF of corresponding traits in natural conditions (Davison et al. 2020; López-García et al. 2017). The functionally shifted communities of AMF may then influence plant competition, given that differential functioning of mycorrhizas with different fungal partners changes competitive interactions between plant species (van der Heijden et al. 1998; Wagg et al. 2011). Overall, our findings suggest that future research should focus on how AMF communities shaped by conventional agricultural management interact with plant species exhibiting different life-history strategies. Specifically, research should examine the influence of specific AMF communities on plant establishment and interspecific competition, which

may importantly influence secondary plant succession on arable soils.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s00374-025-01900-w>.

Acknowledgements The work was financially supported by Czech Science Foundation, project No. GA23-05453S; Ministry of Education, Youth and Sports of the Czech Republic (project CZ.02.01.01/00/22_008/0004597 Talking microbes—understanding microbial interactions within One Health framework); and the long-term research development programs of Czech Academy of Sciences RVO 67985939 and RVO 61388971. The authors are grateful to Veronika Chroustová for her excellent technical assistance and to Zuzana Münzbergová for statistical consultation.

Author contributions MJ and JJ designed the study. Material preparation and data collection were performed by MR, MK, PAB and PB, data were analysed by PAB, JJ, PB, GG and MJ. The first draft of the manuscript was prepared by PAB and MJ, while JJ, PAB, NM and MJ finalized the manuscript.

Funding Open access publishing supported by the institutions participating in the CzechELib Transformative Agreement. The work was financially supported by Czech Science Foundation, project No. GA23-05453S, by the Ministry of Education, Youth and Sports of the Czech Republic project Talking microbes—understanding microbial interactions within One Health framework (CZ.02.01.01/00/22_008/0004597), and the long-term research development programs of Czech Academy of Sciences RVO 67985939 and RVO 61388971.

Data availability Sequence data were deposited in the Sequence Read Archive (NCBI) under accession number PRJNA935258. Part of the experimental data is provided as an Online Resource. The primary data will be made publicly available in the PlutoF repository (<https://plutof.ut.ee/>) upon request.

Declarations

Competing interests The authors have no relevant financial or non-financial interests to disclose.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

- Andrino A, Guggenberger G, Sauheitl L, Burkart S, Boy J (2021) Carbon investment into mobilization of mineral and organic phosphorus by arbuscular mycorrhiza. *Biol Fertil Soils* 57:47–64. <https://doi.org/10.1007/s00374-020-01505-5>

- Baltruschat H, Santos VM, da Silva DKA, Schellenberg I, Deubel A, Sieverding E, Oehl F (2019) Unexpectedly high diversity of arbuscular mycorrhizal fungi in fertile Chernozem croplands in Central Europe. *CATENA* 182:104135. <https://doi.org/10.1016/j.catena.2019.104135>
- Bennett AE, Bever JD (2009) Trade-offs between arbuscular mycorrhizal fungal competitive ability and host growth promotion in *Plantago lanceolata*. *Oecologia* 160:807–816. <https://doi.org/10.1007/s00442-009-1345-6>
- Blaszowski J, Chwat G, Goralska A, Ryszcza P, Kowacs GM (2015a) Two new genera, *Dominikia* and *Kamienskia*, and *D. disticha* sp. nov. in Glomeromycota. *Nov Hedwigia* 100:225–238. https://doi.org/10.1127/nova_hedwigia/2014/0216
- Blaszowski J, Chwat G, Symanczik S, Góralska A (2015b) *Dominikia duoreactiva* sp. nov. and *Dominikia difficilevidera* sp. nov., two new species in the Glomeromycota. *Botany* 93:389–396. <https://doi.org/10.1139/cjb-2015-0016>
- Blaszowski J, Ryszcza P, Kozłowska A (2018) *Dominikia litorea*, a new species in the Glomeromycotina, and biogeographic distribution of *Dominikia*. *Phytotaxa* 338:241–254. <https://doi.org/10.11646/phytotaxa.338.3.2>
- Blažková A, Jansa J, Püschel D, Vosátka M, Janoušková M (2021) Is mycorrhiza functioning influenced by the quantitative composition of the mycorrhizal fungal community? *Soil Biol Biochem* 157:108249. <https://doi.org/10.1016/j.soilbio.2021.108249>
- Boddington CL, Dodd JC (1999) Evidence that differences in phosphate metabolism in mycorrhizas formed by species of *Glomus* and *Gigaspora* might be related to their life-cycle strategies. *New Phytol* 142:531–538. <https://doi.org/10.1046/j.1469-8137.1999.00422.x>
- Boussageon R, Marro N, Janoušková M, Brulé D, Wipf D, Courty PE (2022) The fine-tuning of mycorrhizal pathway in *Sorghum* depends on both nitrogen-phosphorus availability and the identity of the fungal partner. *Plant Cell Environ* 45:3354–3366. <https://doi.org/10.1111/pce.14426>
- Ter Braak CJF, Šmilauer P (2012) Canoco reference manual and user's guide: software for ordination, version 5.0. Microcomputer Power, Ithaca, USA.
- Brooks ME, Kristensen K, Benthem KJV, Magnusson A, Berg CW, Nielsen A, Skaug HJ, Mächler M, Bolker BM (2017) glmmTMB Balances Speed and Flexibility Among Packages for Zero-inflated Generalized Linear Mixed Modeling. *R J* 9:378. <https://doi.org/10.32614/RJ-2017-066>
- Buil PA, Jansa J, Blažková A, Holubík O, Duffková R, Rozmoš M, Püschel D, Kotianová D, Janoušková M (2023) Infectivity and symbiotic efficiency of native arbuscular mycorrhizal fungi from high-input arable soils. *Plant Soil* 482:627–645. <https://doi.org/10.1007/s11104-022-05715-8>
- Bukovská P, Bonkowski M, Konvalinková T, Beskid O, Hujšlová M, Püschel D, Řezáčová V, Gutiérrez-Núñez MS, Gryndler M, Jansa J (2018) Utilization of organic nitrogen by arbuscular mycorrhizal fungi—is there a specific role for protists and ammonia oxidizers? *Mycorrhiza* 28:269–283. <https://doi.org/10.1007/s00572-018-0851-y>
- Bukovská P, Rozmoš M, Kotianová M, Gančarčíková K, Dudáš M, Hršelová H, Jansa J (2021) Arbuscular mycorrhiza mediates efficient recycling from soil to plants of nitrogen bound in chitin. *Front Microbiol* 12:1–16. <https://doi.org/10.3389/fmicb.2021.574060>
- Canarini A, Kaiser C, Merchant A, Richter A, Wanek W (2019) Root Exudation of Primary Metabolites: Mechanisms and Their Roles in Plant Responses to Environmental Stimuli. *Front Plant Sci* 10:157. <https://doi.org/10.3389/fpls.2019.00157>
- Chagnon PL, Bradley RL, Maherali H, Klironomos JN (2013) A trait-based framework to understand life history of mycorrhizal fungi. *Trends Plant Sci* 18:484–491. <https://doi.org/10.1016/j.tplants.2013.05.001>
- Chagnon PL, Bradley RL, Lafond J, Paré MC, Penaud V (2021) Trait-based and phylogenetic filtering of arbuscular mycorrhizal fungal communities under long-term agricultural practices. *Plant Soil* 471:273–287. <https://doi.org/10.1007/S11104-021-05155-W>
- Chapin FS (1980) The mineral nutrition of wild plants. *Annu Rev Ecol Syst* 11:233–260. <https://doi.org/10.1146/annurev.es.11.110180.001313>
- Chatterjee N, Margenot AJ (2023) Crop growth is increased by arbuscular mycorrhizae for both phosphate rock and soluble phosphorus fertilizers, but fertilizer solubility primarily determines crop growth. *Biol Fertil Soils* 59:843–862. <https://doi.org/10.1007/s00374-023-01751-3>
- Chung YA, Jumpponen A, Rudgers JA (2019) Divergence in diversity and composition of root-associated fungi between greenhouse and field studies in a semiarid grassland. *Microb Ecol* 78:122–135. <https://doi.org/10.1007/s00248-018-1277-y>
- Chýlová T, Münzbergová Z (2008) Past land-use co-determines the present distribution of dry grassland plant species. *Preslia* 80:183–198
- Chytrý M, Danihelka J, Kaplan Z, Wild J, Holubová D, Novotný P, Řezníčková M, Rohn M, Dřevojan P, Grulich V, Klimešová J, Lepš J, Lososová Z, Pergl J, Sádlo J, Šmarda P, Štěpánková P, Tichý L, Axmanová I, Bartušková A, Blažek P, Chrtek J Jr, Fischer FM, Guo WY, Herben T, Janovský Z, Konečná M, Kühn I, Moravcová L, Petřík P, Pierce S, Prach K, Prokešová H, Štech M, Těšitel J, Těšitelová T, Večeřa M, Zelený D, Pyšek P (2021) Pladias Database of the Czech Flora and Vegetation. *Preslia* 93:1–87. <https://doi.org/10.23855/preslia.2021.001>
- Cofré N, Becerra AG, Marro N, Domínguez L, Urcelay C (2020) Soybean growth and foliar phosphorus concentration mediated by arbuscular mycorrhizal fungi from soils under different no-till cropping systems. *Rhizosphere* 16:100254. <https://doi.org/10.1016/j.rhisph.2020.100254>
- Corrêa A, Cruz C, Ferrol N (2015) Nitrogen and carbon/nitrogen dynamics in arbuscular mycorrhiza: the great unknown. *Mycorrhiza* 25:499–515. <https://doi.org/10.1007/S00572-015-0627-6>
- da Silva GA, de Assis DMA, Sieverding E, Oehl F (2024) Four new families of arbuscular mycorrhizal fungi within the order Glomerales. *Taxonomy* 4:761–779. <https://doi.org/10.3390/taxonomy4040041>
- Davison J, García de León D, Zobel M, Moora M, Bueno CG, Barceló M, Gerz M (2020) Plant functional groups associate with distinct arbuscular mycorrhizal fungal communities. *New Phytol* 226:1117–1128. <https://doi.org/10.1111/nph.16423>
- Dostálek T, Knappová J, Münzbergová Z (2022) The role of plant-soil feedback in long-term species coexistence cannot be predicted from its effects on plant performance. *Ann Bot* 130:535–546. <https://doi.org/10.1093/aob/mcac080>
- Dudáš M, Pjevac P, Kotianová M, Gančarčíková K, Rozmoš M, Hršelová H, Bukovská P, Jansa J (2022) Arbuscular mycorrhiza and nitrification: Disentangling processes and players by using synthetic nitrification inhibitors. *Appl Environ Microbiol* 88:e01369–e1422. <https://doi.org/10.1128/aem.01369-22>
- Dufrene M, Legendre P (1997) Species Assemblages and Indicator Species: The Need for a Flexible Asymmetrical Approach. *Ecol Monogr* 67:345–366. [https://doi.org/10.1890/0012-9615\(1997\)067\[0345:SAIST\]2.0.CO;2](https://doi.org/10.1890/0012-9615(1997)067[0345:SAIST]2.0.CO;2)
- Dumbrell AJ, Ashton PD, Aziz N, Feng G, Nelson M, Dytham C, Fitter AH, Helgason T (2011) Distinct seasonal assemblages of arbuscular mycorrhizal fungi revealed by massively parallel pyrosequencing. *New Phytol* 190:794–804. <https://doi.org/10.1111/j.1469-8137.2010.03636.x>
- Eason WR, Scullion J, Scott EP (1999) Soil parameters and plant responses associated with arbuscular mycorrhizas from

- contrasting grassland management regimes. *Agric Ecosyst Environ* 73:245–255. [https://doi.org/10.1016/S0167-8809\(99\)00054-7](https://doi.org/10.1016/S0167-8809(99)00054-7)
- Edlinger A, Garland G, Hartman K, Banerjee S, Degrune F, García-Palacios P, Hallin S, Valzano-Held A, Herzog C, Jansa J, Kost E, Maestre FT, Pescador DS, Philippot L, Rillig MC, Romdhane S, Saghai A, Spor A, Frossard E, van der Heijden MGA (2022) Agricultural management and pesticide use reduce the functioning of beneficial plant symbionts. *Nat Ecol Evol* 6:1145–1154. <https://doi.org/10.1038/s41559-022-01799-8>
- Ellenberg H (1988) *Vegetation Ecology of Central Europe*, 4th edn. Cambridge University Press, Cambridge, UK
- Emmett BD, Lévesque-Tremblay V, Harrison MJ (2021) Conserved and reproducible bacterial communities associate with extraradical hyphae of arbuscular mycorrhizal fungi. *ISME J* 15:2276–2288. <https://doi.org/10.1038/s41396-021-00920-2>
- Faghihinia M, Jansa J, Halverson LJ, Staddon PL (2023) Hyphosphere microbiome of arbuscular mycorrhizal fungi: a realm of unknowns. *Biol Fertil Soils* 59:17–34. <https://doi.org/10.1007/s00374-022-01683-4>
- Florianová A, Knappová J, Münzbergová Z (2022) Soil origin and duration of a garden experiment affect competitive outcomes between dominant grassland species. *J Veg Sci* 33:e13160. <https://doi.org/10.1111/jvs.13160>
- Fritze H, Tuomivirta T, Orrù L, Canfora L, Cuartero J, Ros M, Pascual JA, Zornosa R, Egea-Cortines M, Lång K, Kaseva J, Peltoniemi K (2024) Effect of no-till followed by crop diversification on the soil microbiome in a boreal short cereal rotation. *Biol Fertil Soils* 60:357–374. <https://doi.org/10.1007/s00374-024-01797-x>
- García de Leon D, Vahter T, Zobel M, Koppel M, Edesi L, Davison J, Al-Quraishy S, Hozzein WN, Moora M, Oja J, Vasar M, Öpik M (2020) Different wheat cultivars exhibit variable responses to inoculation with arbuscular mycorrhizal fungi from organic and conventional farms. *PLoS ONE* 15:1–17. <https://doi.org/10.1371/journal.pone.0233878>
- Gottshall CB, Cooper M, Emery SM (2017) Activity, diversity and function of arbuscular mycorrhizae vary with changes in agricultural management intensity. *Agric Ecosyst Environ* 241:142–149. <https://doi.org/10.1016/j.agee.2017.03.011>
- Grime JP (1977) Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. *Am Nat* 111:1169–1194. <https://doi.org/10.1086/283244>
- Gryndler M, Šmilauer P, Püschel D, Bukovská P, Hřelová H, Hujšlová M, Gryndlerová H, Beskid O, Konvalinková T, Jansa J (2018) Appropriate nonmycorrhizal controls in arbuscular mycorrhiza research: a microbiome perspective. *Mycorrhiza* 28:435–450. <https://doi.org/10.1007/s00572-018-0844-x>
- Hart MM, Reader RJ (2002a) Host plant benefit from association with arbuscular mycorrhizal fungi: Variation due to differences in size of mycelium. *Biol Fertil Soils* 36:357–366. <https://doi.org/10.1007/s00374-002-0539-4>
- Hart MM, Reader RJ (2002b) Taxonomic basis for variation in the colonization strategy of arbuscular mycorrhizal fungi. *New Phytol* 153:335–344. <https://doi.org/10.1046/j.0028-646X.2001.00312.x>
- Hodge A, Fitter AH (2010) Substantial nitrogen acquisition by arbuscular mycorrhizal fungi from organic material has implications for N cycling. *Proc Natl Acad Sci U S A* 107:13754–13759. <https://doi.org/10.1073/PNAS.1005874107>
- Hodge A, Storer K (2015) Arbuscular mycorrhiza and nitrogen: Implications for individual plants through to ecosystems. *Plant Soil* 386:1–19. <https://doi.org/10.1007/s11104-014-2162-1>
- Hoeksema JD, Chaudhary VB, Gehring CA, Johnson NC, Karst J, Koide RT, Pringle A, Zabinski C, Bever JD, Moore JC, Wilson GWT, Klironomos JN, Umbanhowar J (2010) A meta-analysis of context-dependency in plant response to inoculation with mycorrhizal fungi. *Ecol Lett* 13:394–407. <https://doi.org/10.1111/j.1461-0248.2009.01430.x>
- Hyde KD, Noorabadi MT, Thiagaraja V et al (2024) The 2024 Outline of Fungi and fungus-like taxa. *Mycosphere* 15:5146–6239. <https://doi.org/10.5943/mycosphere/15/1/25>
- Ingraffia R, Amato G, Sosa-Hernández MA, Frenda AS, Rillig MC, Giambalvo D (2020) Nitrogen type and availability drive mycorrhizal effects on wheat performance, nitrogen uptake and recovery, and production sustainability. *Front Plant Sci* 11:760. <https://doi.org/10.3389/fpls.2020.00760>
- Jansa J, Mozafar A, Anken T, Ruh R, Sanders IR, Frossard E (2002) Diversity and structure of AMF communities as affected by tillage in a temperate soil. *Mycorrhiza* 12:225–234. <https://doi.org/10.1007/s00572-002-0163-z>
- Jansa J, Mozafar A, Kuhn G, Anken T, Ruh R, Sanders IR, Frossard E (2003) Soil tillage affects the community structure of mycorrhizal fungi in maize roots. *Ecol Appl* 13:1164–1176. [https://doi.org/10.1890/1051-0761\(2003\)13\[1164:STATCS\]2.0.CO;2](https://doi.org/10.1890/1051-0761(2003)13[1164:STATCS]2.0.CO;2)
- Jansa J, Erb A, Oberholzer HR, Šmilauer P, Egli S (2014) Soil and geography are more important determinants of indigenous arbuscular mycorrhizal communities than management practices in Swiss agricultural soils. *Mol Ecol* 23:2118–2135. <https://doi.org/10.1111/mec.12706>
- Johnson NC (1993) Can Fertilization of Soil Select Less Mutualistic Mycorrhizae? *Ecol Appl* 3:749–757. <https://doi.org/10.2307/1942106>
- Johnson NC (2010) Resource stoichiometry elucidates the structure and function of arbuscular mycorrhizas across scales. *New Phytol* 185:631–647. <https://doi.org/10.1111/j.1469-8137.2009.03110.x>
- Johnson NC, Gibson KS (2021) Understanding multilevel selection may facilitate management of arbuscular mycorrhizae in sustainable agroecosystems. *Front Plant Sci* 11:627345. <https://doi.org/10.3389/fpls.2020.627345>
- Johnson NC, Wilson GWT, Wilson JA, Miller RM, Bowker MA (2015) Mycorrhizal phenotypes and the Law of the Minimum. *New Phytol* 205:1473–1484. <https://doi.org/10.1111/nph.13172>
- Kahiluoto H, Ketoja E, Vestberg M (2009) Contribution of arbuscular mycorrhiza to soil quality in contrasting cropping systems. *Agric Ecosyst Environ* 134:36–45. <https://doi.org/10.1016/j.agee.2009.05.016>
- Knappová J, Hemrová L, Knapp M, Münzbergová Z (2017) Establishment limitation may be more important than species dispersal: insights from dry grasslands and old-fields. *J Veg Sci* 28:34–42. <https://doi.org/10.1111/jvs.12462>
- Koch AM, Antunes PM, Maherali H, Hart MM, Klironomos JN (2017) Evolutionary asymmetry in the arbuscular mycorrhizal symbiosis: conservatism in fungal morphology does not predict host plant growth. *New Phytol* 214:1330–1337. <https://doi.org/10.1111/NPH.14465>
- Koerselman W, Meuleman AFM (1996) The vegetation N: P ratio: A new tool to detect the nature of nutrient limitation. *J Appl Ecol* 33:1441–1450. <https://doi.org/10.2307/2404783>
- Köhl L, Oehl F, van der Heijden MGA (2014) Agricultural practices indirectly influence plant productivity and ecosystem services through effects on soil biota. *Ecol Appl* 24:1842–1853. <https://doi.org/10.1890/13-1821.1>
- Köhl L, Lukaszewicz CE, van der Heijden MGA (2016) Establishment and effectiveness of inoculated arbuscular mycorrhizal fungi in agricultural soils. *Plant Cell Environ* 39:136–146. <https://doi.org/10.1111/pce.12600>
- Konvalinková T, Püschel D, Řezáčová V, Gryndlerová H, Jansa J (2017) Carbon flow from plant to arbuscular mycorrhizal fungi is reduced under phosphorus fertilization. *Plant Soil* 419:319–333. <https://doi.org/10.1007/s11104-017-3350-6>

- Koske RE, Gemma JN (1989) A modified procedure for staining roots to detect VA mycorrhizas. *Mycol Res* 92:486–505. [https://doi.org/10.1016/S0953-7562\(89\)80195-9](https://doi.org/10.1016/S0953-7562(89)80195-9)
- Lee J, Lee S, Young JPW (2008) Improved PCR primers for the detection and identification of arbuscular mycorrhizal fungi. *FEMS Microbiol Ecol* 65:339–349. <https://doi.org/10.1111/j.1574-6941.2008.00531.x>
- Lekberg Y, Koide RT (2005) Is plant performance limited by abundance of arbuscular mycorrhizal fungi? A meta-analysis of studies published between 1988 and 2003. *New Phytol* 168:189–204. <https://doi.org/10.1111/j.1469-8137.2005.01490.x>
- Lekberg Y, Koide RT, Twomlow SJ (2008) Effect of agricultural management practices on arbuscular mycorrhizal fungal abundance in low-input cropping systems of southern Africa: A case study from Zimbabwe. *Biol Fertil Soils* 44:917–923. <https://doi.org/10.1007/s00374-008-0274-6>
- Lendenmann M, Thonar C, Barnard RL, Salmon Y, Werner RA, Frossard E, Jansa J (2011) Symbiont identity matters: Carbon and phosphorus fluxes between *Medicago truncatula* and different arbuscular mycorrhizal fungi. *Mycorrhiza* 21:689–702. <https://doi.org/10.1007/s00572-011-0371-5>
- Lerat S, Lapointe L, Gutjahr S, Piché Y, Vierheilig H (2003) Carbon partitioning in a split-root system of arbuscular mycorrhizal plants is fungal and plant species dependent. *New Phytol* 157:589–595. <https://doi.org/10.1046/j.1469-8137.2003.00691.x>
- Liu A, Hamel C, Elmi A, Costa C, Ma B, Smith DL (2002) Concentrations of K, Ca and Mg in maize colonized by arbuscular mycorrhizal fungi under field conditions. *Can J Soil Sci* 82:272–278. <https://doi.org/10.4141/S01-022>
- López-García Á, Varela-Cervero S, Vasar M, Öpik M, Barea JM, Azcón-Aguilar C (2017) Plant traits determine the phylogenetic structure of arbuscular mycorrhizal fungal communities. *Mol Ecol* 26:6948–6959. <https://doi.org/10.1111/mec.14403>
- Manoharan L, Rosenstock NP, Williams A, Hedlund K (2017) Agricultural management practices influence AMF diversity and community composition with cascading effects on plant productivity. *Appl Soil Ecol* 115:53–59. <https://doi.org/10.1016/j.apsoil.2017.03.012>
- Marrassini V, Ercoli L, Kuramae EE, Kowalchuk GA, Pellegrino E (2024) Arbuscular mycorrhizal fungi originating from soils with a fertility gradient highlight a strong interspecies functional variability. *Appl Soil Ecol* 197:105344
- Marro N, Cofré N, Grilli G, Alvarez C, Labuckas D, Maestri D, Urcey C (2020) Soybean yield, protein content and oil quality in response to interaction of arbuscular mycorrhizal fungi and native microbial populations from mono- and rotation-cropped soils. *Appl Soil Ecol* 152:103575. <https://doi.org/10.1016/j.apsoil.2020.103575>
- Marschner P, Crowley DE, Higashi RM (1997) Root exudation and physiological status of a root-colonizing fluorescent pseudomonad in mycorrhizal and non-mycorrhizal pepper (*Capsicum annuum* L.). *Plant Soil* 189:11–20. <https://doi.org/10.1023/A:1004266907442>
- Martinez TN, Johnson NC (2010) Agricultural management influences propagule densities and functioning of arbuscular mycorrhizas in low- and high-input agroecosystems in arid environments. *Appl Soil Ecol* 46:300–306. <https://doi.org/10.1016/j.apsoil.2010.07.001>
- McGonigle TP, Miller MH, Evans DG, Fairchild GL, Swan JA (1990) A new method which gives an objective measure of colonization of roots by vesicular arbuscular mycorrhizal fungi. *New Phytol* 115:495–501. <https://doi.org/10.1111/j.1469-8137.1990.tb00476.x>
- Mensah JA, Koch AM, Antunes PM, Kiers ET, Hart M, Bücking H (2015) High functional diversity within species of arbuscular mycorrhizal fungi is associated with differences in phosphate and nitrogen uptake and fungal phosphate metabolism. *Mycorrhiza* 25:533–546. <https://doi.org/10.1007/S00572-015-0631-X>
- Müller A, Eltigani A, George E (2019) The abundance of arbuscular mycorrhizal fungal species in symbiosis with okra plants is affected by induced drought conditions in a calcareous substrate. *Rhizosphere* 10:100150. <https://doi.org/10.1016/j.rhisph.2019.100150>
- Munkvold L, Kjølner R, Vestberg M, Rosendahl S, Jakobsen I (2004) High functional diversity within species of arbuscular mycorrhizal fungi. *New Phytol* 164:357–364. <https://doi.org/10.1111/j.1469-8137.2004.01169.x>
- Nazeri NK, Lambers H, Tibbett M, Ryan MH (2014) Moderating mycorrhizas: arbuscular mycorrhizas modify rhizosphere chemistry and maintain plant phosphorus status within narrow boundaries. *Plant Cell Environ* 37:911–921. <https://doi.org/10.1111/pce.12207>
- Oehl F, Sieverding E, Ineichen K, Mäder P, Wiemken A, Boller T (2009) Distinct sporulation dynamics of arbuscular mycorrhizal fungal communities from different agroecosystems in long-term microcosms. *Agric Ecosyst Environ* 134:257–268. <https://doi.org/10.1016/j.agee.2009.07.008>
- Oehl F, Laczo E, Bogenrieder A, Stahr K, Bösch R, van der Heijden MGA, Sieverding E (2010) Soil type and land use intensity determine the composition of arbuscular mycorrhizal fungal communities. *Soil Biol Biochem* 42:724–738. <https://doi.org/10.1016/j.soilbio.2010.01.006>
- Ohno T, Zibilske LM (1991) Determination of low concentrations of phosphorus in soil extracts using malachite green. *Soil Sci Soc Am J* 55:892–895. <https://doi.org/10.2136/sssaj1991.03615995005500030046x>
- Pearson JN, Abbott LK, Jasper DA (1993) Mediation of competition between two colonizing VA mycorrhizal fungi by the host plant. *New Phytol* 123:93–98. <https://doi.org/10.1111/j.1469-8137.1993.tb04534.x>
- Pellegrino E, Turrini A, Gamper HA, Cafa G, Bonari E, Young JPW, Giovannetti M (2012) Establishment, persistence and effectiveness of arbuscular mycorrhizal fungal inoculants in the field revealed using molecular genetic tracing and measurement of yield components. *New Phytol* 194:810–822. <https://doi.org/10.1111/j.1469-8137.2012.04090.x>
- Pellegrino E, Arcidiacono M, Francini A, Ercoli L (2024) Arbuscular mycorrhizal fungi with contrasting life-history strategies differently affect health-promoting compounds in field-grown tomato by changing arbuscule occurrence and mycorrhizal assemblages in roots. *Biol Fertil Soils* 60:115–136. <https://doi.org/10.1007/s00374-023-01778-6>
- Peng Z, Johnson NC, Jansa J, Han J, Fang Z, Zhang Y, Jiang S, Xi H, Mao L, Pan J, Zhang Q, Feng H, Fan T, Zhang J, Liu Y (2024) Mycorrhizal effects on crop yield and soil ecosystem functions in a long-term tillage and fertilization experiment. *New Phytol* 242:1798–1813. <https://doi.org/10.1111/nph.19493>
- Perotto S, Balestrini R (2024) At the core of the endomycorrhizal symbioses: intracellular fungal structures in orchid and arbuscular mycorrhiza. *New Phytol* 242:1408–1416. <https://doi.org/10.1111/NPH.19338>
- Philippot L, Raaijmakers JM, Lemanceau P, van der Putten WH (2013) Going back to the roots: the microbial ecology of the rhizosphere. *Nat Rev Microbiol* 11:789–799. <https://doi.org/10.1038/nrmicro3109>
- Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core Team (2021) nlme: Linear and Nonlinear Mixed Effects Models. <https://CRAN.R-project.org/package=nlme>
- Porter SS, Sachs JL (2020) Agriculture and the disruption of plant-microbial symbiosis. *Trends Ecol Evol* 35:426–439. <https://doi.org/10.1016/j.tree.2020.01.006>

- Powell JR, Parrent JL, Hart MM, Klironomos JN, Rillig MC, Maherali H (2009) Phylogenetic trait conservatism and the evolution of functional trade-offs in arbuscular mycorrhizal fungi. *Proc R Soc B Biol Sci* 276:4237–4245. <https://doi.org/10.1098/rspb.2009.1015>
- Püschel D, Janoušková M, Hujšlová M, Slavíková R, Gryndlerová H, Jansa J (2016) Plant–fungus competition for nitrogen erases mycorrhizal growth benefits of *Andropogon gerardii* under limited nitrogen supply. *Ecol Evol* 6:4332–4346. <https://doi.org/10.1002/ece3.2207>
- R Core Team, R Development Core Team, R Core Team (2020) R: A language and environment for statistical computing. In: R Found. Stat. Comput. Vienna, Austria. <https://www.r-project.org/>
- Řezáčová V, Gryndler M, Bukovská P, Šmilauer P, Jansa J (2016) Molecular community analysis of arbuscular mycorrhizal fungi - Contributions of PCR primer and host plant selectivity to the detected community profiles. *Pedobiologia* 59:179–187. <https://doi.org/10.1016/j.pedobi.2016.04.002>
- Roberts DW (2023) labdsv: Ordination and Multivariate Analysis for Ecology. R package version 2.1–0, <https://CRAN.R-project.org/package=labdsv>
- Ryan MH, Graham JH (2018) Little evidence that farmers should consider abundance or diversity of arbuscular mycorrhizal fungi when managing crops. *New Phytol* 220:1092–1107. <https://doi.org/10.1111/nph.15308>
- Ryan MH, Tibbett M, Edmonds-Tibbett T, Suriyagoda LDB, Lambers H, Cawthray GR, Pang J (2012) Carbon trading for phosphorus gain: the balance between rhizosphere carboxylates and arbuscular mycorrhizal symbiosis in plant phosphorus acquisition. *Plant Cell Environ* 35:2170–2180. <https://doi.org/10.1111/j.1365-3040.2012.02547.x>
- Säle V, Aguilera P, Laczko E, Mäder P, Berner A, Zihlmann U, van der Heijden MGA, Oehl F (2015) Impact of conservation tillage and organic farming on the diversity of arbuscular mycorrhizal fungi. *Soil Biol Biochem* 84:38–52. <https://doi.org/10.1016/j.soilbio.2015.02.005>
- Säle V, Palenzuela J, Azcón-Aguilar C, Sánchez-Castro I, Alves da Silva G, Seitz B, Sieverding E, van der Heijden MGA, Oehl F (2021) Ancient lineages of arbuscular mycorrhizal fungi provide little plant benefit. *Mycorrhiza* 31:559–576. <https://doi.org/10.1007/s00572-021-01042-5>
- Sasse J, Martinoia E, Norrhen T (2018) Feed Your Friends: Do Plant Exudates Shape the Root Microbiome? *Trends Plant Sci* 23:25–41. <https://doi.org/10.1016/j.tplants.2017.09.003>
- Scullion J, Eason WR, Scott EP (1998) The effectivity of arbuscular mycorrhizal fungi from high input conventional and organic grassland and grass-arable rotations. *Plant Soil* 204:243–254. <https://doi.org/10.1023/A:1004319325290>
- Shannon CE (1948) A mathematical theory of communication. *Bell Syst Tech J* 27:379–423. <https://doi.org/10.1002/j.1538-7305.1948.tb01338.x>
- Slavíková R, Püschel D, Janoušková M, Hujšlová M, Konvalinková T, Gryndlerová H, Gryndler M, Weiser M, Jansa J (2017) Monitoring CO₂ emissions to gain a dynamic view of carbon allocation to arbuscular mycorrhizal fungi. *Mycorrhiza* 27:35–51. <https://doi.org/10.1007/s00572-016-0731-2>
- Šmilauer P, Šmilauerová M, Kotlínek M, Košnar J (2020) Foraging speed and precision of arbuscular mycorrhizal fungi under field conditions: An experimental approach. *Mol Ecol* 29:1574–1587. <https://doi.org/10.1111/MEC.15425>
- Šmilauer P, Šmilauerová M, Kotlínek M, Košnar J (2021) Arbuscular mycorrhizal fungal communities of forbs and C3 grasses respond differently to cultivation and elevated nutrients. *Mycorrhiza* 31:455–470. <https://doi.org/10.1007/s00572-021-01036-3>
- Smith SE, Read DJ (2008) *Mycorrhizal Symbioses*, 3rd edn. Academic Press, Cambridge, UK
- Sun D, Rozmoš M, Kokkoris V, Kotianová M, Hršelová H, Bukovská P, Faghihinia M, Jansa J (2024) Unraveling the diversity of hyphal explorative traits among *Rhizophagus irregularis* genotypes. *Mycorrhiza* 34:303–316. <https://doi.org/10.1007/s00572-024-01154-8>
- Sýkorová Z, Ineichen K, Wiemken A, Redecker D (2007) The cultivation bias: different communities of arbuscular mycorrhizal fungi detected in roots from the field, from bait plants transplanted to the field, and from a greenhouse trap experiment. *Mycorrhiza* 18:1–14. <https://doi.org/10.1007/s00572-007-0147-0>
- Tedersoo L, Moghaddam MSH, Mikryukov V, Hakimzadeh A, Bahram M, Nilsson RH, Yatsiuk I, Geisen S, Schwelm A, Piwosz K, Prous M, Sildever S, Chmolewska D, Rueckert S, Škaloud P, Laas P, Tines M, Jung JH, Choi JH, Alkahtani S, Anslan S (2024) EUKARYOME: the rRNA gene reference database for identification of all eukaryotes. Database 2024:baae043. <https://doi.org/10.1093/database/baae043>
- Thonar C, Schnepf A, Frossard E, Roose T, Jansa J (2011) Traits related to differences in function among three arbuscular mycorrhizal fungi. *Plant Soil* 339:231–245. <https://doi.org/10.1007/s11104-010-0571-3>
- Treseder KK, Allen MF (2002) Direct nitrogen and phosphorus limitation of arbuscular mycorrhizal fungi: a model and field test. *New Phytol* 155:507–515. <https://doi.org/10.1046/j.1469-8137.2002.00470.x>
- van der Heijden MGA, Boller T, Wiemken A, Sanders IR (1998) Different Arbuscular Mycorrhizal Fungal Species Are Potential Determinants of Plant Community Structure. *Ecology* 79:2082. <https://doi.org/10.2307/176711>
- van Geel M, Busschaert P, Honnay O, Lievens B (2014) Evaluation of six primer pairs targeting the nuclear rRNA operon for characterization of arbuscular mycorrhizal fungal (AMF) communities using 454 pyrosequencing. *J Microbial Methods* 106:93–100. <https://doi.org/10.1016/j.mimet.2014.08.006>
- Verbruggen E, Kiers ET (2010) Evolutionary ecology of mycorrhizal functional diversity in agricultural systems. *Evol Appl* 3:547–560. <https://doi.org/10.1111/j.1752-4571.2010.00145.x>
- Verbruggen E, Rölting WFM, Gamper HA, Kowalchuk GA, Verhoef HA, van der Heijden MGA (2010) Positive effects of organic farming on below-ground mutualists: Large-scale comparison of mycorrhizal fungal communities in agricultural soils. *New Phytol* 186:968–979. <https://doi.org/10.1111/j.1469-8137.2010.03230.x>
- Verbruggen E, Kiers ET, Bakelaar PNC, Rölting WFM, van der Heijden MGA (2012) Provision of contrasting ecosystem services by soil communities from different agricultural fields. *Plant Soil* 350:43–55. <https://doi.org/10.1007/s11104-011-0828-5>
- Verbruggen E, Xiang D, Chen B, Xu T, Rillig MC (2015) Mycorrhizal fungi associated with high soil N: P ratios are more likely to be lost upon conversion from grasslands to arable agriculture. *Soil Biol Biochem* 86:1–4. <https://doi.org/10.1016/j.soilbio.2015.03.008>
- Wagg C, Jansa J, Stadler M, Schmid B, van der Heijden MGA (2011) Mycorrhizal fungal identity and diversity relaxes plant–plant competition. *Ecology* 92:1303–1313. <https://doi.org/10.1890/10-1915.1>
- Wang L, George TS, Feng G (2024) Concepts and consequences of the hyphosphere core microbiome for arbuscular mycorrhizal fungal fitness and function. *New Phytol* 242:1529–1533. <https://doi.org/10.1111/nph.19396>
- Werner GDA, Kiers ET (2015) Order of arrival structures arbuscular mycorrhizal colonization of plants. *New Phytol* 205:1515–1524. <https://doi.org/10.1111/nph.13092>

- Williams A, Manoharan L, Rosenstock NP, Olsson PA, Hedlund H (2017) Long-term agricultural fertilization alters arbuscular mycorrhizal fungal community composition and barley (*Hordeum vulgare*) mycorrhizal carbon and phosphorus exchange. New Phytol 213:874–885. <https://doi.org/10.1111/nph.14196>
- Zhang L, Zhou J, George TS, Limpens E, Feng G (2022) Arbuscular mycorrhizal fungi conducting the hyphosphere bacterial

orchestra. Trends Plant Sci 27:402–411. <https://doi.org/10.1016/j.tplants.2021.10.008>

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.