nature microbiology

Article

https://doi.org/10.1038/s41564-023-01520-w

Soil microbiome indicators can predict crop growth response to large-scale inoculation with arbuscular mycorrhizal fungi

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Supplementary Results and Discussion

Most important soil parameters for MGR prediction (extended)

The 15 soil parameters in the full model could explain about 26 % of the variations in MGR (**Fig. 4 A**). These comprised cMIC, Nmin and magnesium. Total nitrogen content was negatively correlated with MGR (**Supplementary Table 2**), while SAF22 was positively associated with MGR in the model. A higher content of easily leachable magnesium (H2O extraction) was also associated with a higher MGR, while this was reversed for more metal-bound and thus less bioavailable magnesium (EDTA extraction). This could be due to the fact that magnesium plays an important role in stimulating root colonisation by AMF (Gryndler, Vejsadová, and Vančura 1992) when bioavailable.

Most important soil OTUs for MGR prediction (extended)

Analogous to the soil parameters we reduced the number of candidate sOTUs for model input, again using different approaches (**Fig. 3 B**). An indicator species analysis revealed 11 and 20 sOTUs characteristic of fields with high and low MGR, respectively (**Supplementary Table 7**). Differential abundance analysis between fields with high and low MGR resulted in 5 and 14 sOTUs, respectively (**Supplementary Table 8**). Furthermore, a random forest analysis was performed on the continuous MGR values (**Supplementary Table 9**). Only the top 30 predictors were retained since this number of sOTUs corresponds to the identified number of sOTUs in the other two analyses.

Comparing all three methods (**Supplementary Table 10**), only two fungal sOTUs were identified as important by all three methods (sOTU18: *Trichosporon* sp. (high MGR) and sOTU182: *Dendryphion europaeum* (low MGR)), which was in contrast to the relatively large overlap of soil parameters selected by different approaches. In addition, seven sOTUs were selected by two methods and 49 sOTUs by one method only. For reasons of stricter selectivity and due to the small overlap between the random forest and the other two analyses, sOTUs identified solely by random forest were not retained. The combined results of the indicator species and DESeq2 analyses yielded a total of 44 sOTUs.

While fields with high MGR were characterized by higher pathogen load, the main indicators of low MGR included the genera *Phaeohelotium*, *Phaeosphaeria*, *Powellomyces* and three sOTUs without database match. Not much is known about these genera in the literature, but they could be indicators of generally healthy soils, as suggested by the positive correlation of the most important predictor OTU58 (no database match) with soil fertility and microbial respiration (**Supplementary Fig. 13**).

Model with all fungal sOTUs classified as pathogens not able to predict MGR

Since soil pathogenic fungi were the strongest predictors in the model, we investigated whether the relative abundance of all soil pathogenic fungi would perform better in predicting MGR. Therefore, we evaluated fungal guilds of all sOTUs using FUNGuild and summed the abundance of sOTUs that were classified as pathogens (Supplementary Table 13). However, their abundance was neither significantly correlated with MGR (R=0.029, p=0.83; Supplementary Fig. 20) nor performed well in a linear regression model (R= 0.0036, p= 0.6701). On the one hand, this could be because in most cases there are no exact matches in the sequence databases and because even within a species, traits are often highly diverse. Therefore, functional annotations are often unreliable if not conducted on the strain level. On the other hand, it may indeed be that only a few pathogens AMF strains are likely specialised in their ability to protect against specific pathogens.

Root microbiome data confirms findings of MGR prediction model

Of note, we identified *Trichosporon*, the most important soil fungal OTU in the predictive model, also in the data of the root fungal communities (rOTU20 shares 100% sequence similarity with sOTU18 over its entire amplicon length). Although the *Trichosporon* rOTU20 was not significantly (p=0.263) different in abundance between control or inoculated roots, it showed a trend of a decrease (log2FC=-0.583) similar to the differentially abundant pathogens in roots of fields with high MGR (**Supplementary Table 15**).

Inoculum application

The inoculum was manually mixed with field soils. This application mode is highly suitable for scientific experiments, but it is not feasible for large scale agricultural production. In order to achieve a better cost-benefit ratio, the dosage and method of application of the AMF inoculum must also be optimised, ideally in cooperation with farmers and specialised biotech companies.

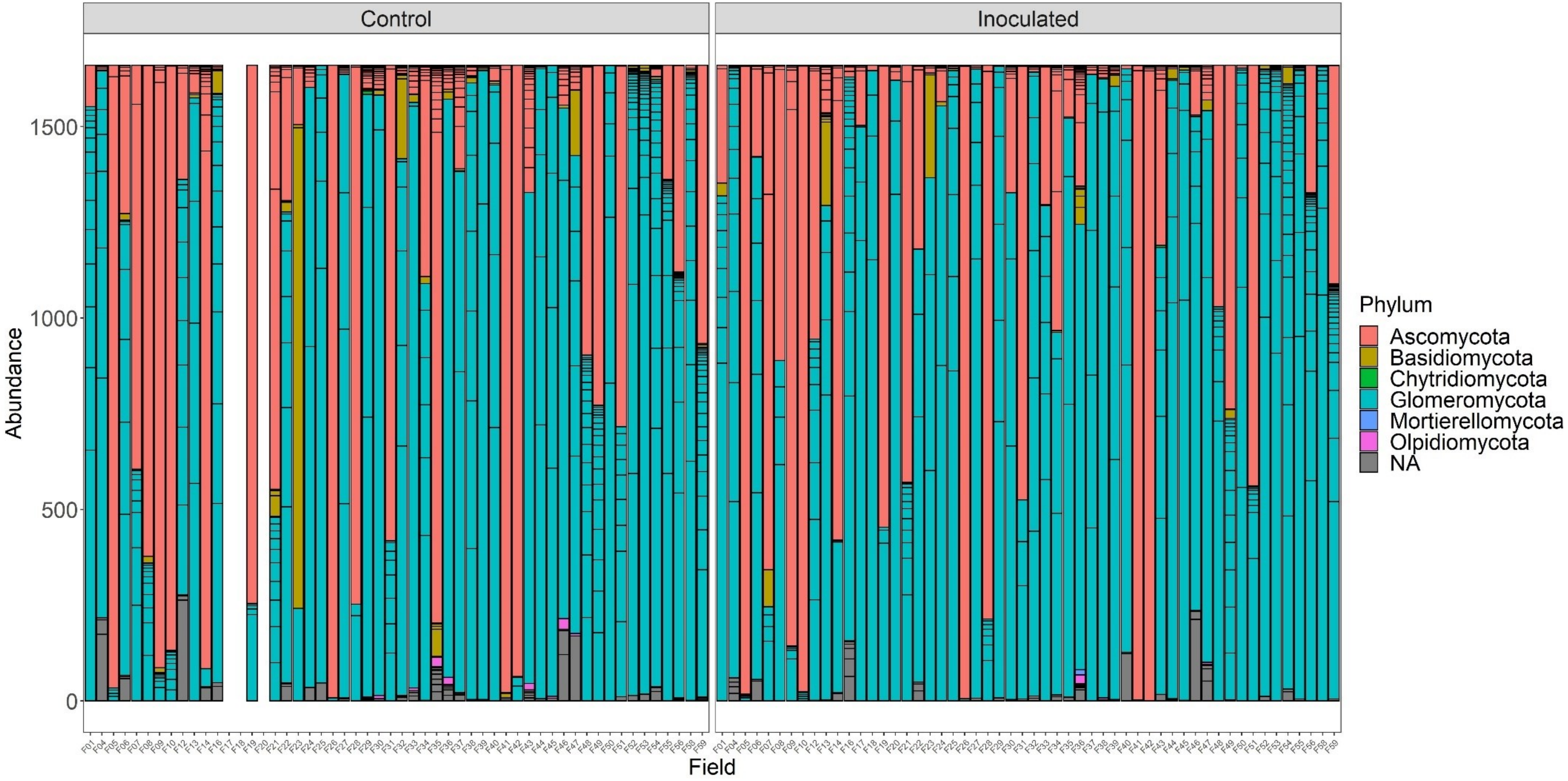
References

Bender, S. Franz, Klaus Schlaeppi, Alain Held, and Marcel G.A. Van der Heijden. 2019. “Establishment Success and Crop Growth Effects of an Arbuscular Mycorrhizal Fungus Inoculated into Swiss Corn Fields.” *Agriculture, Ecosystems & Environment* 273 (March): 13–24. https://doi.org/10.1016/J.AGEE.2018.12.003.

Gryndler, M., H. Vejsadová, and V. Vančura. 1992. “The Effect of Magnesium Ions on the Vesicular—Arbuscular Mycorrhizal Infection of Maize Roots.” *New Phytologist* 122 (3): 455–60. https://doi.org/10.1111/J.1469-8137.1992.TB00073.X.

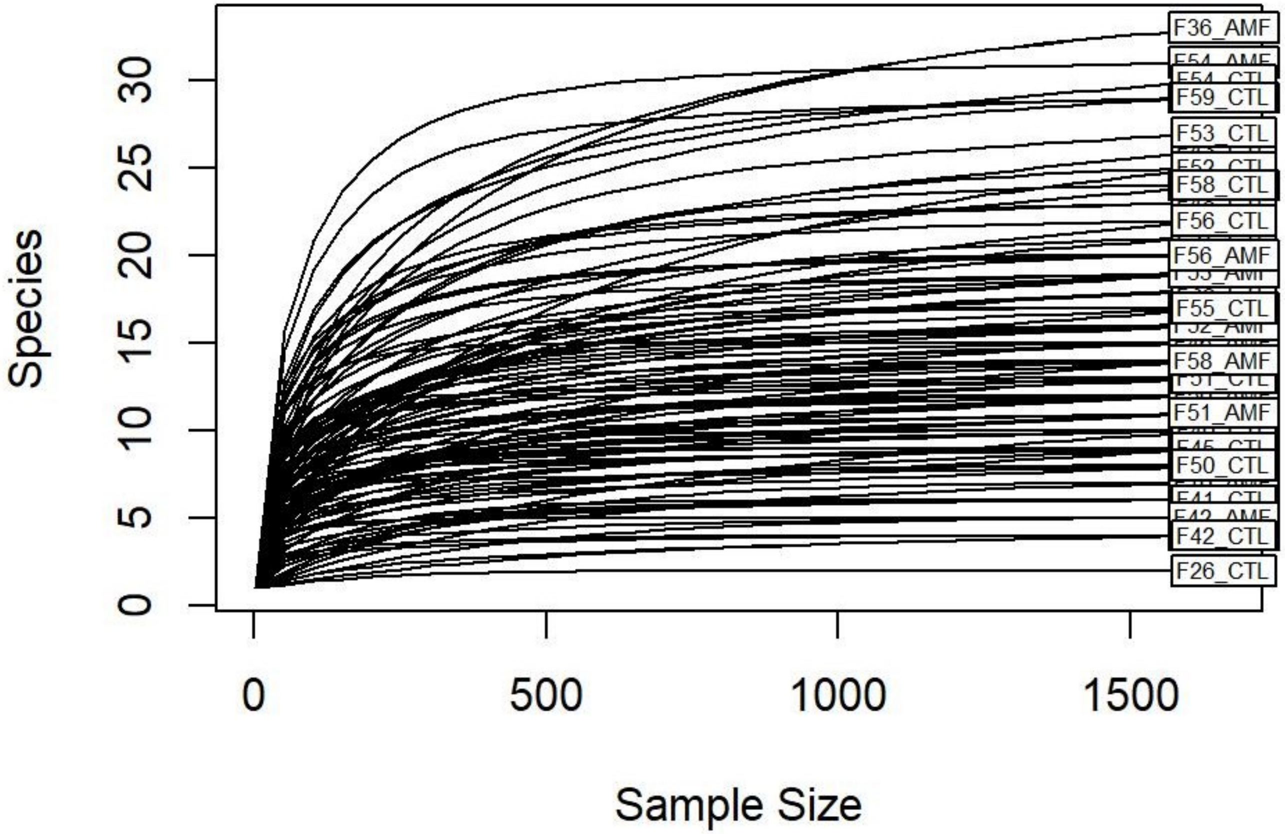
Supplementary Fig. 1: Relative abundance of the root microbiome at the phylum level.

Control samples are shown on the left and corresponding samples inoculated with the AMF SAF22 on the right. Sequencing of the root microbiome with AMF-specific primers led not not only to the amplification of the main target phylum Glomeromycota, but also Ascomycota and Basidiomycota. The plot is based on the rarefied OTU table. White bars represent missing samples that could not be sequenced due to the concentrations of the PCR products being too low.



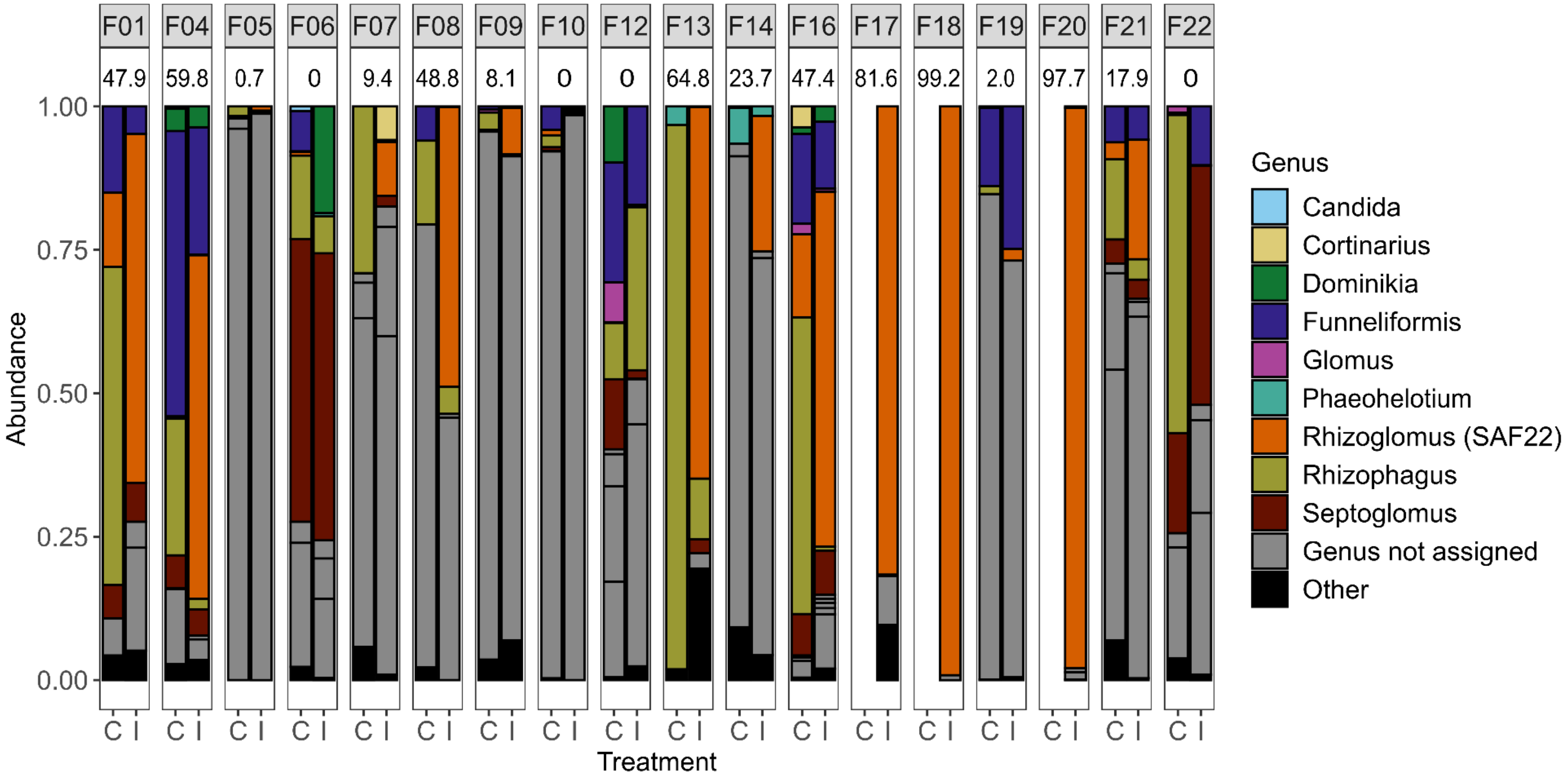
Supplementary Fig. 2: Rarefaction curves of subsampled root OTU table.

Plateauing curves show that sufficient sequencing depth was retained after rarefying to an even sequence number of 1660 reads per sample.



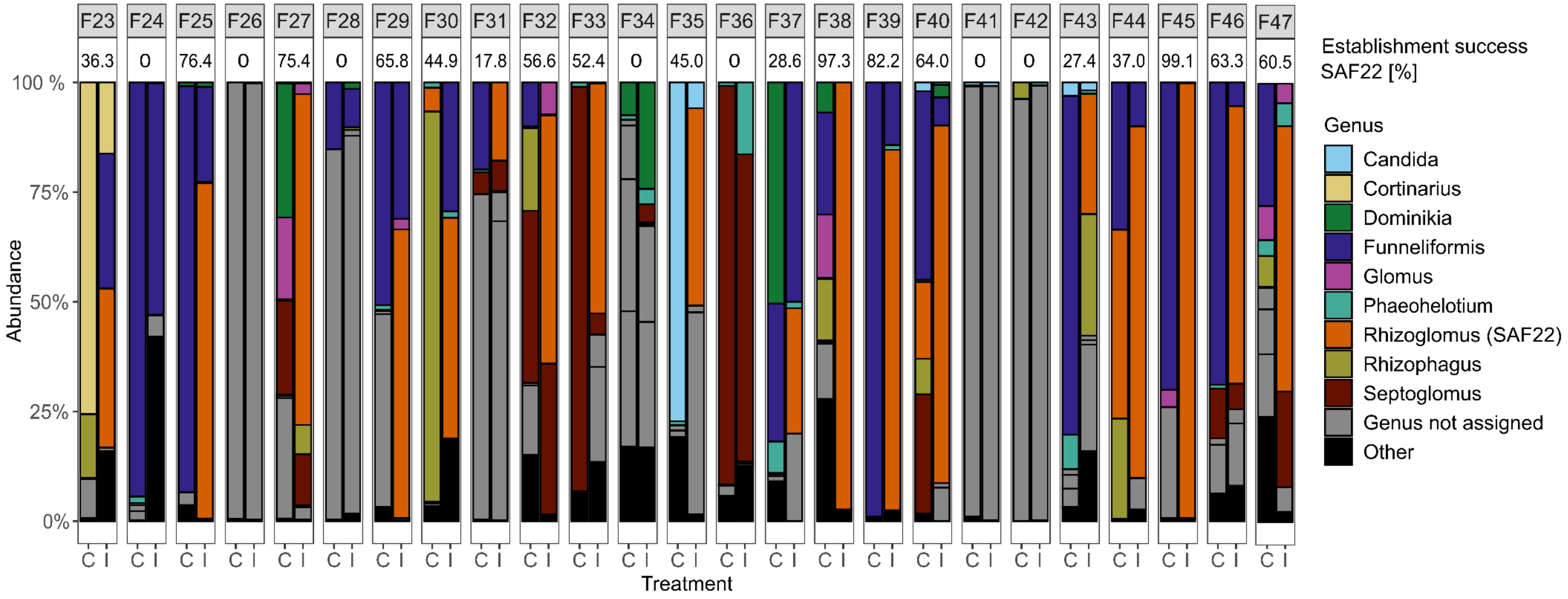
Supplementary Fig. 3 A (2018): Root community composition at genus level.

The plot shows the relative abundance of the root microbiome for control (C) and inoculated (I) samples for each field. The 15 most abundant genera were selected for better visualisation. Within these genera, OTUs without genus assignment are summarised in grey (genus not assigned). Less abundant genera are displayed in black. The establishment success of the inoculated *Rhizoglomus irregulare* SAF22 is shown below the field IDs and represents the difference between inoculated and control samples. White bars represent missing samples that could not be sequenced due to the concentrations of the PCR products being too low.



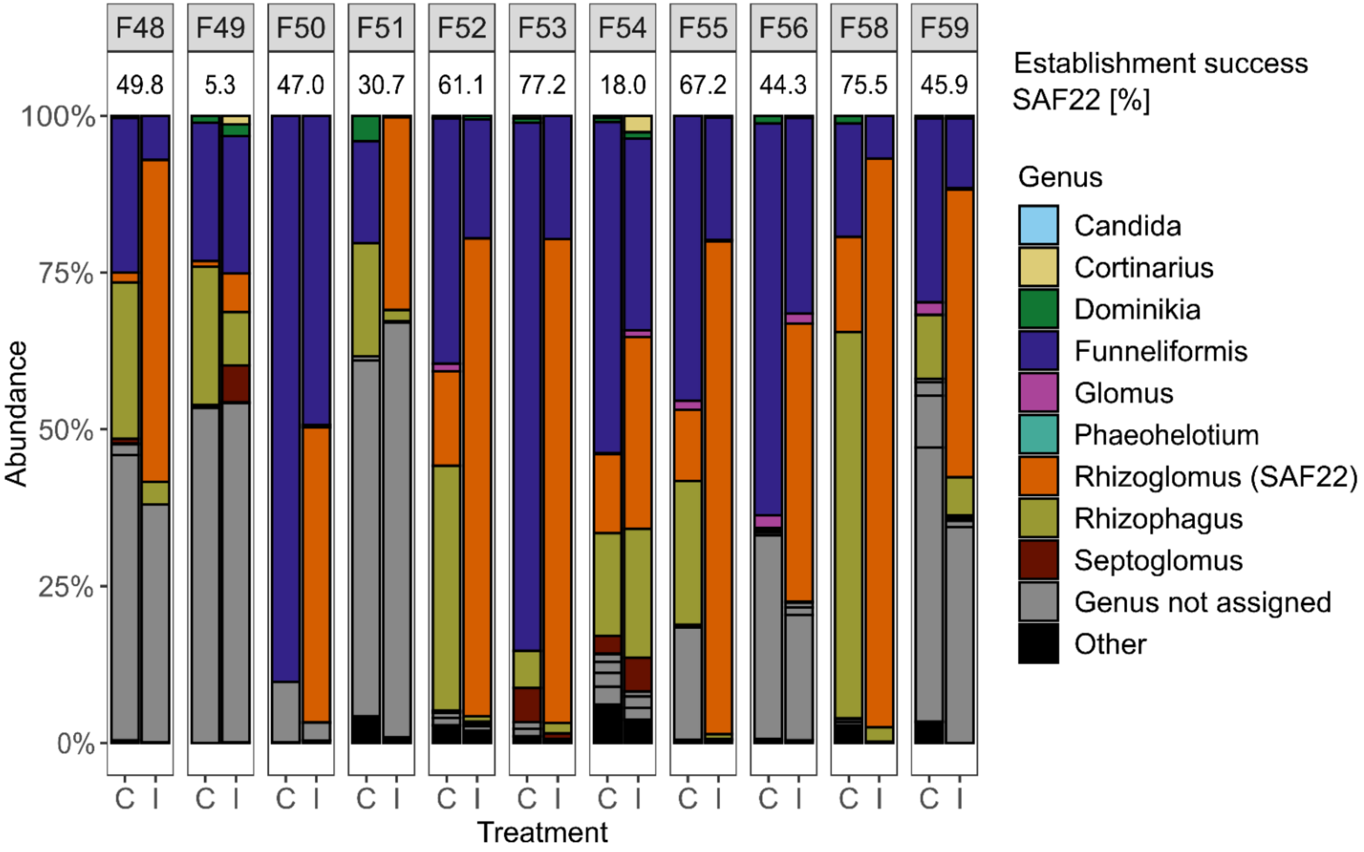
Supplementary Fig. 3 B (2019): Root community composition at genus level.

The plot shows the relative abundance of the root microbiome for control (C) and inoculated (I) samples for each field. The 15 most abundant genera were selected for better visualisation. Within these genera, OTUs without genus assignment are summarised in grey (genus not assigned). Less abundant genera are displayed in black. The establishment success of the inoculated *Rhizoglomus irregulare* SAF22 is shown below the field IDs and represents the difference between inoculated and control samples. White bars represent missing samples that could not be sequenced due to the concentrations of the PCR products being too low.



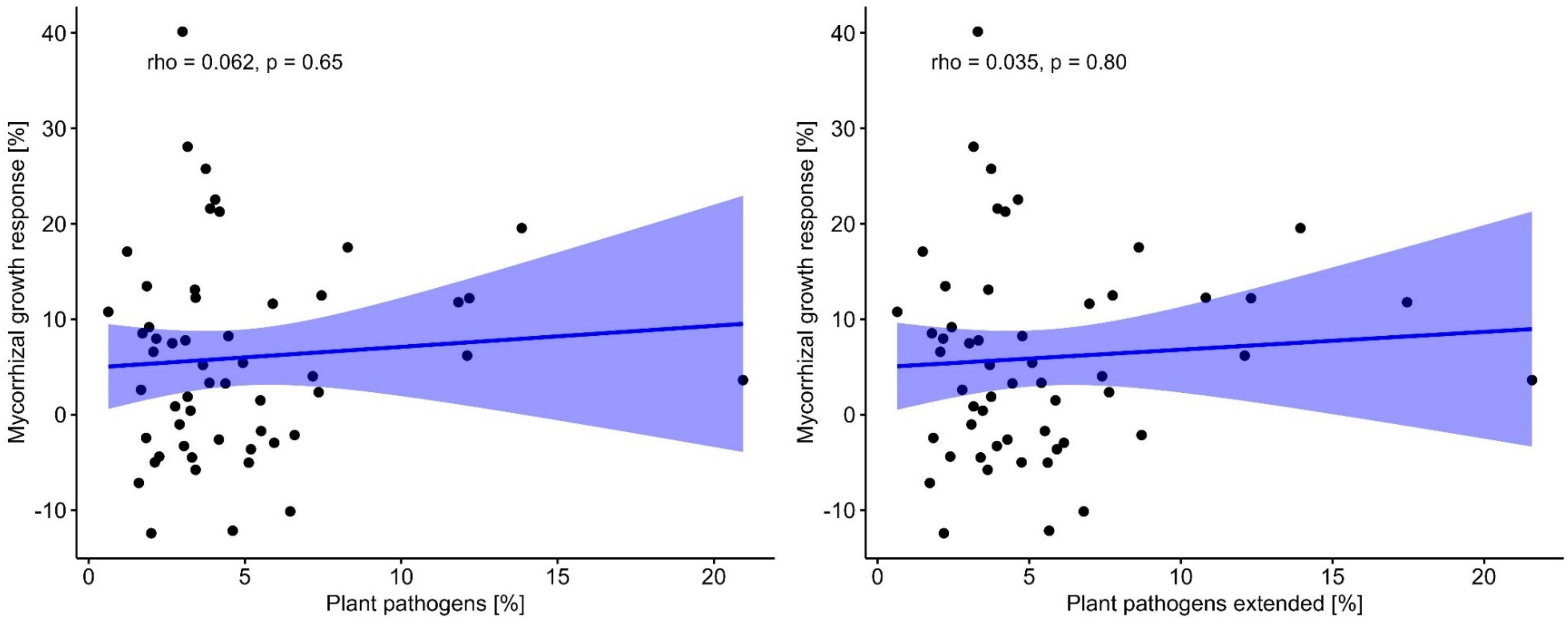
Supplementary Fig. 3 C (2020): Root community composition at genus level.

The plot shows the relative abundance of the root microbiome for control (C) and inoculated (I) samples for each field. The 15 most abundant genera were selected for better visualisation. Within these genera, OTUs without genus assignment are summarised in grey (genus not assigned). Less abundant genera are displayed in black. The establishment success of the inoculated *Rhizoglomus irregulare* SAF22 is shown below the field IDs and represents the difference between inoculated and control samples. White bars represent missing samples that could not be sequenced due to the concentrations of the PCR products being too low.



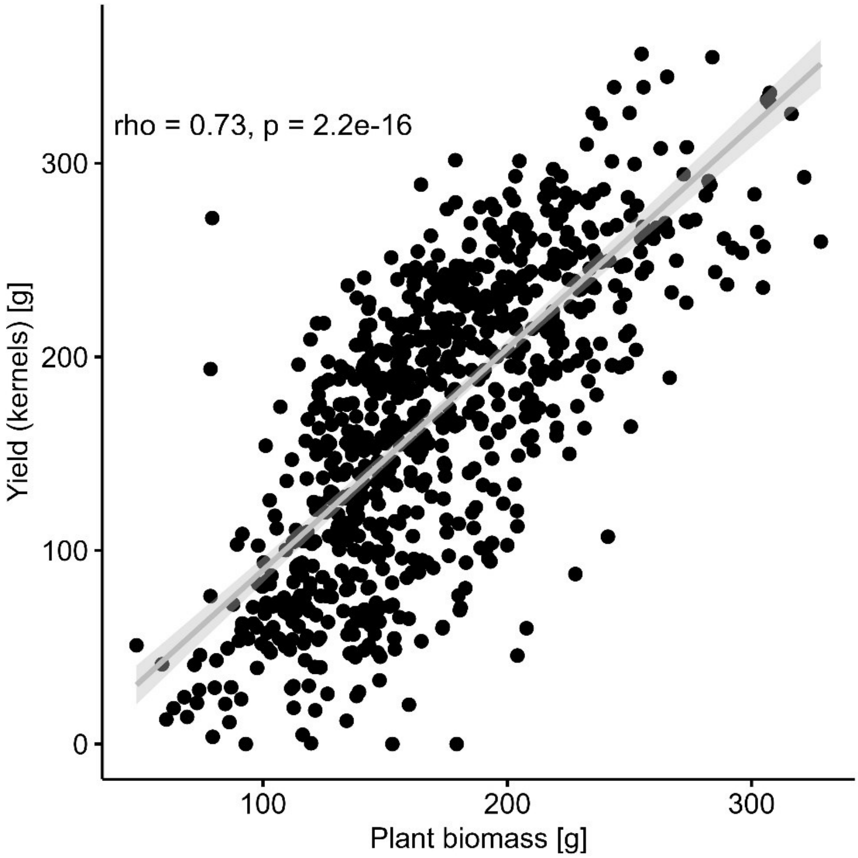
Supplementary Fig. 4: Relationship between all pathogen sOTUs and MGR.

Fungal guilds of all sOTUs were assessed and the abundance of pathogenic OTUs were summed up. The scatter plot on the left refers only to the guild "plant pathogen", while the plot on the right summarises all guilds with the keyword "plant pathogen". No significant correlations were found. On the one hand, this could be because in most cases there are no exact matches in the sequence databases and because even within a species, traits are often highly diverse. Therefore, functional annotations are often unreliable if not conducted on the strain level. On the other hand, it may indeed be that only a few pathogens are important in the studied context of one particular AMF strain and maize as host plant. AMF strains are likely specialised in their ability to protect against specific pathogens. The correlation coefficients (Spearman rank) and the significant values (p) are displayed in the plots. The blue shaded area represents the 95% confidence interval.



Supplementary Fig. 5: Correlation of plant biomass and yield data.

The grey shaded area represents the 95% confidence interval. The correlation coefficient (Spearman rank, rho) and the significant value (p) is displayed in the plot.



Supplementary Table 1: Overview of inoculated fields including corresponding mycorrhizal growth response and soil parameters.

See separate spreadsheet “Supplementary Tables”.

Supplementary Table 2: Pairwise correlations of soil parameters and MGR.

Table show Spearman rank correlations coefficients and associated p-values in increasing order. No significant ($p<0.05$) correlations were found. Magnesium, Corg, humus and Ntot were weakly correlated with MGR.

	r	p
magnesium_H2O_lbu	0.260	0.057
Corg_agro	-0.258	0.060
humus_agro	-0.252	0.066
Ntot_agro	-0.262	0.089
Ctot_agro	-0.242	0.118
clay_lbu	-0.214	0.120
silt_lbu	-0.198	0.150
clay_agro	-0.182	0.187
WHC_agro	-0.172	0.215
sand_agro	0.156	0.259
Nmin_agro	0.154	0.265
ammonium_H2O_lbu	-0.173	0.268
calcium_agro	-0.152	0.271
respiration_lbu	-0.153	0.273
nitrate_agro	0.148	0.284
slan_lbu	-0.147	0.292
CEC_agro	-0.142	0.307
calcium_EDTA_lbu	-0.154	0.325
iron_H2O_lbu	0.153	0.327
Ptot_agro	-0.135	0.332
respiration_agro	-0.131	0.343
iron_EDTA_lbu	0.131	0.345
sodium_agro	0.122	0.379
fertility_lbu	-0.123	0.390
cMIC_agro	-0.118	0.394
nitrate_H2O_lbu	0.129	0.411
potassium_EDTA_lbu	-0.112	0.419
magnesium_CC_lbu	0.108	0.439
nMIC_agro	-0.096	0.491
ph_lbu	0.093	0.502
ammonium_agro	0.075	0.591
P_olsen_lbu	-0.082	0.605
hydrogen_agro	-0.070	0.613
sodium_H2O_lbu	0.078	0.621
magnesium_agro	0.055	0.694
phosphorus_CO2_lbu	-0.051	0.717
copper_EDTA_lbu	0.045	0.746
boron_EDTA_lbu	-0.041	0.771
silt_agro	-0.037	0.791
magnesium_EDTA_lbu	-0.036	0.794
potassium_agro	-0.027	0.845

manganese_EDTA_lbu	-0.027	0.847
boron_H2O_lbu	-0.027	0.865
phosphorus_EDTA_lbu	-0.020	0.888
phosphorus_H2O_lbu	-0.013	0.925
potassium_CO2_lbu	0.013	0.926
vast_lbu	-0.012	0.932
zinc_EDTA_lbu	-0.008	0.959
humus_lbu	-0.006	0.964
calcium_H2O_lbu	0.006	0.967
BS_agro	-0.005	0.972
potassium_H2O_lbu	0.004	0.978

Supplementary Table 3: Overview of co-correlated parameters.

The 22 parameters on the left were kept for further model selection, whereas parameters on the right were filtered out since they were strongly co-correlated (r>0.8 or r<-0.8).

Selected parameters	Co-correlated parameters and their respective correlation coefficients
Ptot_agro	
potassium_agro	potassium_CO2_lbu (0.88), potassium_H2O_lbu (0.91), potassium_EDTA_lbu (0.93)
pH_lbu	BS_agro (0.88), hydrogen_agro (-0.86)
Corg_agro	CEC_agro (0.80), WHC_agro (0.80), humus_agro (1.0)
humus_lbu	
sodium_agro	
magnesium_H20_lbu	magnesium_agro (0.85), magnesium_CC_lbu (0.83)
magnesium_EDTA_lbu	
calcium_agro	calcium_H2O_lbu (0.83), CEC_agro (0.87), respiration_agro (0.82)
phosphorus_H2O_lbu	phosphorus_CO2_lbu (0.93), phosphorus_EDTA_lbu (0.86)
manganese_EDTA_lbu	
boron_EDTA_lbu	
copper_EDTA_lbu	
iron_EDTA_lbu	
cMIC_agro	nMIC_agro (0.91), respiration_agro (0.83)
Nmin_agro	nitrate_agro (0.99)
ammonium_agro	
sand_agro	
silt_agro	
clay_agro	
silt_lbu	
clay_lbu	

Supplementary Table 4: Random forest analysis of soil parameters.

The analysis was conducted on the reduced pool of 22 parameters (see Supplementary Table 3). Parameters are sorted by descending importance (IncNodePurity).

Soil parameter	IncNodePurity
sand_agro	488
silt_agro	467
magnesium_H2O_lbu	420
magnesium_EDTA_lbu	348
Ptot_agro	324
cMIC_agro	313
Corg_agro	311
iron_EDTA_lbu	306
manganese_EDTA_lbu	303
Nmin_agro	256
clay_agro	213
boron_EDTA_lbu	197
copper_EDTA_lbu	192
phosphorus_H2O_lbu	186
ph_lbu	184
potassium_agro	181
humus_lbu	179
ammonium_agro	154
calcium_agro	147
sodium_agro	140
silt_lbu	103
clay_lbu	90

Supplementary Table 5: Soil parameters selected for final model input.

Comparison of the most important predictors of MGR identified by the glmulti, stepAIC and random forest analyses. The selection by the respective methods is shown with a grey background. The *glmulti* analysis was limited to a maximum number of ten predictors. Similarly, for the random forest analysis, only the top ten predictors are shown.

Soil parameter	**Parameter name in main text**	**glmulti (top 10)**	**stepAIC**	**Random forest (top 10)**
magnesium_EDTA_lbu	magnesium(EDTA)	▨	▨	▨
magnesium_H2O_lbu	magnesium(H2O)	▨	▨	▨
manganese_EDTA_lbu	Manganese	▨	▨	▨
Nmin_agro	Nmin	▨	▨	▨
iron_EDTA_lbu	Iron	▨	▨	▨
cMIC_agro	cMIC	▨	▨	▨
phosphorus_H2O_lbu	phosphorus(H2O)	▨	▨	
ammonium_agro	Ammonium	▨	▨	
Corg_agro	Corg	▨		▨
sand_agro	Sand			▨
silt_agro	Silt			▨
Ptot_agro	Ptot			▨
boron_EDTA_lbu	Boron		▨	
clay_lbu	Clay	▨		
pH_lbu	pH		▨	

Supplementary Table 6: Soil OTU table.

Table shows merged OTU abundances for the four replicates of each samples and rarefied to an even sampling depth (i.e., 4272 reads). The fungal community was characterized by a high relative abundance of Ascomycota, followed by Mortierellomycota and Basidiomycota. The relative abundance of Glomeromycota was very low and ranged from 0 to 1.8%. See separate spreadsheet “Supplementary Tables”.

Supplementary Table 7: Results of the indicator species analysis of fields with low and high MGR.

Displayed are soil OTUs that were associated with low or high MGR (Spearman rank correlation, p<0.1).

OTU	MGR	statistic	p-value	Taxonomy
OTU100	High	0.327	0.086	Ascomycota; Pezizomycetes; Pezizales; Pyronemataceae; Byssonectria; Byssonectria_fusispora
OTU16	High	0.341	0.058	Ascomycota; Sordariomycetes; Hypocreales; Nectriaceae; Gibberella; Gibberella_intricans
OTU177	High	0.249	0.047	Ascomycota; Sordariomycetes; Sordariales; Chaetomiaceae; Humicola; Humicola_nigrescens
OTU18	High	0.305	0.089	Basidiomycota; Tremellomycetes;
OTU243	High	0.349	0.096	NA
OTU274	High	0.255	0.037	Ascomycota; Saccharomycetes; Saccharomycetales; Phaffomycetaceae; Cyberlindnera; Cyberlindnera_misumaiensis
OTU38	High	0.333	0.084	Basidiomycota; Tremellomycetes; Cystofilobasidiales; Mrakiaceae; Mrakia; Mrakia_frigida
OTU388	High	0.328	0.095	Ascomycota; Sordariomycetes; Hypocreales; Stachybotryaceae; Striaticonidium; Striaticonidium_humicola
OTU49	High	0.365	0.081	Olpidiomycota; Olpidiomycetes; Olpidiales; Olpidiaceae; Olpidium; Olpidium_brassicae
OTU561	High	0.358	0.093	NA
OTU106	Low	0.277	0.080	NA
OTU142	Low	0.374	0.048	Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium; NA
OTU15	Low	0.311	0.097	Ascomycota
OTU179	Low	0.358	0.069	Ascomycota; Leotiomycetes; Helotiales
OTU182	Low	0.329	0.099	Ascomycota; Dothideomycetes; Pleosporales; Torulaceae; Dendryphion; Dendryphion_europaeum
OTU217	Low	0.523	0.008	NA
OTU227	Low	0.335	0.092	Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium
OTU273	Low	0.353	0.080	Ascomycota; Eurotiomycetes; Chaetothyriales; Herpotrichiellaceae
OTU366	Low	0.448	0.039	NA
OTU392	Low	0.388	0.094	Chytridiomycota; Spizellomycetes; Spizellomycetales; Powellomycetaceae; Powellomyces
OTU451	Low	0.347	0.100	Ascomycota; Sordariomycetes; Hypocreales; Hypocreales_fam_Incertae_sedis; Acremonium; Acremonium_tubakii
OTU544	Low	0.313	0.099	Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium
OTU58	Low	0.348	0.092	NA

OTU589	Low	0.376	0.039	NA
OTU608	Low	0.408	0.04	Ascomycota; Sordariomycetes; Sordariales; Chaetomiaceae; Dichotomopilus; Dichotomopilus_erectus
OTU68	Low	0.407	0.038	Ascomycota; Dothideomycetes; Pleosporales; Phaeosphaeriaceae
OTU69	Low	0.378	0.048	Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium
OTU72	Low	0.434	0.030	Basidiomycota; Agaricomycetes; Agaricales; Psathyrellaceae; Psathyrella; Psathyrella_sacchariolens
OTU884	Low	0.339	0.084	Ascomycota; Eurotiomycetes; Onygenales

Supplementary Table 8: Results of the DESeq2 analysis of soil OTUs.

Table shows soil OTUs with differential abundance between low and high MGR fields (Wald test, adjusted for multiple comparison, p<0.1). A negative log2FC corresponds to a higher relative abundance in high MGR fields, whereas a positive log2FC corresponds to a higher relative abundance in low MGR fields.

OTU ID	baseMean	log2FC	lfcSE	stat	pvalue	padj	Taxonomy
OTU156	5.551	-2.740	0.738	-3.712	0.0001	0.083	Chytridiomycota, Rhizophlyctidomycetes, Rhizophlyctidales, Rhizophlyctidaceae
OTU18	56.013	-2.597	0.762	-3.410	0.001	0.083	Basidiomycota, Tremellomycetes
OTU251	3.269	-2.402	0.722	-3.326	0.0001	0.048	Chytridiomycota, Cladochytriomycetes, Cladochytriales, Cladochytriaceae, Cladochytrium
OTU100	4.154	-2.252	0.665	-3.388	0.001	0.083	Ascomycota, Pezizomycetes, Pezizales, Pyronemataceae, Byssonectria_fusispora
OTU253	2.614	-2.107	0.719	-2.930	0.003	0.095	Ascomycota, Eurotiomycetes, Onygenales, Onygenales_fam_Incertae_sedis, Chrysosporium_lobatum
OTU242	2.185	1.716	0.588	2.917	0.004	0.095	Ascomycota, Sordariomycetes, Hypocreales, Hypocreales_fam_Incertae_sedis, Sarocladium_strictum
OTU273	2.400	1.772	0.574	3.087	0.002	0.083	Ascomycota, Eurotiomycetes, Chaetothyriales, Herpotrichiellaceae
OTU281	2.364	1.865	0.612	3.046	0.002	0.083	Ascomycota, Sordariomycetes, Hypocreales, Bionectriaceae, Clonostachys_phyllophila
OTU74	3.581	1.926	0.647	2.976	0.003	0.090	NA
OTU252	3.116	1.939	0.639	3.033	0.002	0.083	Basidiomycota, Agaricomycetes, Trechisporales, Hydnodontaceae
OTU631	2.471	1.948	0.617	3.154	0.002	0.083	NA
OTU134	2.579	2.026	0.669	3.026	0.002	0.083	Ascomycota, Leotiomycetes, Helotiales, Helotiaceae
OTU268	2.901	2.122	0.679	3.126	0.002	0.083	Ascomycota, Eurotiomycetes, Onygenales
OTU375	2.794	2.170	0.657	3.302	0.001	0.083	NA
OTU80	9.025	2.224	0.718	3.100	0.002	0.083	Basidiomycota, Tremellomycetes, Cystofilobasidiales, Cystofilobasidiales_fam_Incertae_sedis, Mrakiella_aquatica
OTU75	3.510	2.252	0.709	3.175	0.001	0.083	NA

Supplementary Table 9: Random forest analysis of soil OTUs.

The most important soil OTUs (IncNodePurity > 30) for the prediction of MGR are displayed in descending order.

OTU ID	IncNodePurity
OTU151	359
OTU18	285
OTU26	202
OTU15	177
OTU1325	122
OTU724	109
OTU181	91
OTU52	89
OTU529	82
OTU38	75
OTU9	75
OTU194	67
OTU147	65
OTU182	64
OTU24	57
OTU39	53
OTU322	46
OTU728	41
OTU31	40
OTU422	40
OTU318	39
OTU3	38
OTU30	36
OTU58	35
OTU32	35
OTU33	34
OTU25	34
OTU16	32
OTU34	31
OTU211	31
OTU110	31
OTU40	31
OTU273	31
OTU41	30
OTU155	30

Supplementary Table 10: Soil fungal OTUs selected for final model input.

Comparison of the 44 soil OTUs selected (grey background) by the indicator species (31 sOTUs), DESeq2 (19 sOTUs) and random forest analyses (continuous ranking of sOTUs). Within the top 30 sOTUS identified by random forest, only 5 sOTUs were overlapping with the indicator species and DESeq2 analyses. The combined results of the indicator species and DESeq2 analyses (44 sOTUs, no further sOTUs of the random forest analysis where added in order to not inflate the final pool of sOTUs) were subjected to a further selection step with glmulti. The resulting final 13 sOTUs are highlighted in bold. In addition to the automated taxonomic assignments, these sequences were searched against the NCBI BLAST database to obtain a more refined taxonomic identification.

MGR	OTU ID	Indicator species	DESeq2	Random forest	Taxonomy	NCBI BLAST (% identity)
High	**sOTU16**	▨			Ascomycota; Sordariomycetes; Hypocreales; Nectriaceae; Gibberella; Gibberella_intricans	Fusarium equiseti (100%)
	sOTU18	▨	▨	▨	Basidiomycota; Tremellomycetes	Trichosporon sp. (100%)
	sOTU38	▨		▨	Basidiomycota; Tremellomycetes; Cystofilobasidiales; Mrakiaceae; Mrakia; Mrakia_frigida	
	sOTU49	▨			Olpidiomycota; Olpidiomycetes; Olpidiales; Olpidiaceae; Olpidium; Olpidium_brassicae	Olpidium brassicae (99.83%)
	sOTU93	▨			Ascomycota; Sordariomycetes; Glomerellales; Plectosphaerellaceae; Plectosphaerella	
	sOTU100	▨	▨		Ascomycota; Pezizomycetes; Pezizales; Pyronemataceae; Byssonectria; Byssonectria_fusispora	
	sOTU156		▨		Chytridiomycota, Rhizophlyctidomycetes, Rhizophlyctidales, Rhizophlyctidaceae	
	sOTU177	▨			Ascomycota; Sordariomycetes; Sordariales; Chaetomiaceae; Humicola; Humicola_nigrescens	*Chaetomium* sp. (98.83%)
	sOTU243	▨			NA	
	sOTU251		▨		Chytridiomycota; Cladochytriomycetes; Cladochytriales; Cladochytriaceae; Cladochytrium	No blast hit
	sOTU253		▨		Ascomycota; Eurotiomycetes; Onygenales; Onygenales_fam_Incertae_sedis; Chrysosporium; Chrysosporium_lobatum	
	sOTU274	▨			Ascomycota; Saccharomycetes; Saccharomycetales; Phaffomycetaceae; Cyberlindnera; Cyberlindnera_misumaiensis	
	sOTU388	▨			Ascomycota; Sordariomycetes; Hypocreales; Stachybotryaceae; Striaticonidium; Striaticonidium_humicola	Striaticonidium sp./ Myrothecium sp. (99.64%)

	sOTU561	▨			NA	No blast hit
Low	OTU15	▨		▨	Ascomycota	
	OTU58	▨		▨	NA	No blast hit
	OTU68	▨			Ascomycota; Dothideomycetes; Pleosporales; Phaeosphaeriaceae	Phaeosphaeria sp./Dokmaia sp. (100%)
	OTU69	▨			Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium	
	OTU72	▨			Basidiomycota; Agaricomycetes; Agaricales; Psathyrellaceae; Psathyrella; Psathyrella_sacchariolens	
	OTU106	▨			NA	
	OTU134		▨		Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium	
	OTU142	▨			Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium	No blast hit
	OTU179	▨	▨		Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium	
	OTU182	▨	▨	▨	Ascomycota; Dothideomycetes; Pleosporales; Torulaceae; Dendryphion; Dendryphion_europaeum	
	OTU217	▨			NA	
	OTU227	▨			Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium	No blast hit
	OTU243		▨		Ascomycota, Sordariomycetes, Hypocreales, Hypocreales_fam_Incertae_sedis, Sarocladium_strictum	
	OTU252		▨		Basidiomycota; Agaricomycetes; Trechisporales; Hydnodontaceae; Trechispora	
	OTU268		▨		Ascomycota, Eurotiomycetes, Onygenales	
	OTU273	▨	▨		Ascomycota; Eurotiomycetes; Chaetothyriales; Herpotrichiellaceae	
	OTU281		▨		Ascomycota, Sordariomycetes, Hypocreales, Bionectriaceae, Clonostachys_phyllophila	
	OTU284		▨		Basidiomycota; Agaricomycetes; Boletales; Boletaceae; Indoporus; Indoporus shoreae	
	OTU366	▨			NA	
	OTU375		▨		NA	
	OTU392	▨			Chytridiomycota; Spizellomycetes; Spizellomycetales; Powellomycetaceae; Powellomyces	No blast hit
	OTU451	▨			Ascomycota; Sordariomycetes; Hypocreales; Hypocreales_fam_Incertae_sedis; Acremonium; Acremonium_tubakii	
	OTU544	▨			Ascomycota; Leotiomycetes; Helotiales; Helotiaceae; Phaeohelotium	

OTU589	■		NA
OTU608	■		Ascomycota; Sordariomycetes; Sordariales; Chaetomiaceae; Dichotomopilus; Dichotomopilus_erectus
OTU631	■	■	NA
OTU74		■	NA
OTU75		■	NA
OTU80		■	Basidiomycota, Tremellomycetes, Cystofilobasidiales, Cystofilobasidiales_fam_Incertae_sedis, Mrakiella_aquatica
OTU884	■		Ascomycota; Eurotiomycetes; Onygenales

Supplementary Table 11: Root OTU table.

Table shows OTU abundances rarefied to an even sampling depth (i.e., 1660 reads). See separate spreadsheet “Supplementary Tables”.

Supplementary Table 12: AMF establishment success.

On the left, the summed relative abundance of all root OTUs corresponding to SAF22 (rOTU2, rOTU4, rOTU9, rOTU16, rOTU74, rOTU84, rOTU165) is shown for the control (CTL) and inoculated (AMF) samples, as well as their difference (Δ). On the right, the averaged (eight replicates) total root colonisation of the control and inoculated samples is shown, as well as their difference (Δ).

Field	**Relative abundance of SAF22 rOTUS**			**Total root colonisation**		
	CTL [%]	AMF [%]	Δ (AMF - CTL) [%]	CTL_mean [%]	AMF_mean [%]	Δ (AMF-CTL) [%]
F01	13.0	60.8	47.9	66.8	81.9	15.1
F04	0.1	59.9	59.8	70.9	83.3	12.4
F05	0.0	0.7	0.7	76.3	86.0	9.7
F06	0.7	0.0	0.0	61.9	56.1	-5.7
F07	0.0	9.4	9.4	56.6	70.5	13.9
F08	0.0	48.8	48.8	45.1	72.6	27.5
F09	0.0	8.1	8.1	61.9	70.6	8.7
F10	0.9	0.4	0.0	57.5	54.0	-3.5
F12	0.0	0.0	0.0	81.6	92.3	10.6
F13	0.0	64.8	64.8	38.1	68.9	30.8
F14	0.0	23.7	23.7	81.9	75.3	-6.6
F16	14.5	61.9	47.4	82.4	80.3	-2.1
F17	0.0	81.6	81.6	62.1	87.3	25.1
F18	0.0	99.2	99.2	27.5	96.8	69.3
F19	0.0	2.0	2.0	45.9	59.9	14.0
F20	0.0	97.7	97.7	21.1	51.0	29.9
F21	3.0	20.8	17.9	74.4	75.4	1.1
F22	0.0	0.0	0.0	53.3	56.0	2.8
F23	0.0	36.3	36.3	59.8	80.1	20.4
F24	0.0	0.0	0.0	24.1	43.1	19.0
F25	0.0	76.4	76.4	41.0	87.0	46.0
F26	0.0	0.0	0.0	44.6	40.1	-4.4
F27	0.0	75.4	75.4	73.4	91.4	18.0
F28	0.0	0.0	0.0	51.8	66.1	14.4
F29	0.0	65.8	65.8	52.0	80.6	28.6
F30	5.4	50.3	44.9	68.6	80.0	11.4
F31	0.0	17.8	17.8	64.1	90.8	26.6
F32	0.0	56.6	56.6	56.8	83.5	26.8
F33	0.0	52.4	52.4	67.1	77.9	10.8
F34	0.0	0.0	0.0	70.0	64.6	-5.4
F35	0.0	45.0	45.0	55.1	80.5	25.4
F36	0.0	0.0	0.0	55.5	53.9	-1.6
F37	0.0	28.6	28.6	47.3	73.9	26.6
F38	0.0	97.3	97.3	82.9	96.6	13.8

F39	0.0	82.2	82.2	56.3	79.6	23.3
F40	17.5	81.5	64.0	68.5	88.8	20.3
F41	0.0	0.0	0.0	62.6	67.0	4.4
F42	0.0	0.0	0.0	40.9	50.3	9.4
F43	0.0	27.4	27.4	34.6	55.5	20.9
F44	43.1	80.1	37.0	63.4	90.6	27.2
F45	0.0	99.1	99.1	74.4	89.3	14.8
F46	0.0	63.3	63.3	68.6	87.6	18.9
F47	0.0	60.5	60.5	54.3	57.8	3.5
F48	1.6	51.3	49.8	39.6	70.3	30.7
F49	0.9	6.2	5.3	76.4	69.3	-7.1
F50	0.0	47.0	47.0	74.8	89.4	14.7
F51	0.0	30.7	30.7	21.0	57.0	36.0
F52	15.1	76.1	61.1	84.5	92.5	8.0
F53	0.0	77.2	77.2	59.8	89.5	29.8
F54	12.7	30.6	18.0	95.2	87.0	-8.2
F55	11.4	78.6	67.2	78.2	96.0	17.8
F56	0.0	44.3	44.3	73.8	72.9	-0.9
F58	15.2	90.7	75.5	44.6	88.3	43.6
F59	0.0	45.9	45.9	78.0	79.5	1.5

Supplementary Table 13: Fungal guilds of soil OTUs.

See separate spreadsheet “Supplementary Tables”.

Supplementary Table 14: Results of the DESeq2 analysis of root OTUs in low MGR fields.

Table shows root OTUs with a significant (Wald test, adjusted for multiple comparison, $p < 0.05$) differential abundance between control and inoculated samples. A negative log2FC corresponds to a higher relative abundance in control samples, whereas a positive log2FC corresponds to a higher relative abundance in inoculated samples. In addition, the values for rOTU20, which likely matches the important soil predictor sOTU18, are highlighted in grey.

OTU ID	baseMean	log2FC	lfcSE	Stat	Pvalue	Padj	Taxonomy
OTU8	4.727	-2.168	0.369	-5.883	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU5	7.582	-1.999	0.363	-5.505	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis sp.
OTU10	3.780	-1.484	0.344	-4.312	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus sp.
OTU13	2.202	-1.348	0.307	-4.394	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU19	2.329	-1.284	0.316	-4.066	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU21	1.567	-0.973	0.316	-3.075	0.002	0.013	Glomeromycetes
OTU3	18.010	-0.970	0.392	-2.479	0.013	0.059	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis mosseae
OTU38	1.568	-0.895	0.283	-3.161	0.002	0.011	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU49	1.459	-0.811	0.300	-2.705	0.007	0.036	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus sp.
OTU28	1.451	-0.755	0.285	-2.649	0.008	0.039	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis sp.
OTU34	1.599	-0.674	0.294	-2.291	0.022	0.090	Glomeromycetes, Glomerales, Glomeraceae, Glomus, Glomus sp.
OTU98	1.342	-0.656	0.294	-2.231	0.026	0.090	Sordariomycetes, Glomerellales, Plectosphaerellaceae, Verticillium
OTU43	1.411	-0.607	0.282	-2.153	0.031	0.099	Tremellomycetes
OTU25	1.313	-0.603	0.270	-2.235	0.025	0.090	Paraglomeromycetes, Paraglomerales, Paraglomeraceae, Paraglomus
OTU20	1.197	0.109	0.263	0.415	0.678	0.837	100% similarity with full length of soil OTU18 (Trichosporon sp.)
OTU50	1.342	0.628	0.292	2.153	0.031	0.099	Glomeromycetes, Glomerales, Glomeraceae, Dominikia, Dominikia iranica
OTU29	1.518	0.660	0.291	2.266	0.023	0.090	Glomeromycetes, Glomerales, Glomeraceae, Septoglomus, Septoglomus viscosum

OTU4	9.592	1.098	0.383	2.869	0.004	0.024	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU16	2.558	1.925	0.287	6.696	0.0001	00.001	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU9	4.254	2.764	0.332	8.317	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU2	25.142	5.302	0.345	15.363	0.0001	0.0001	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22

Supplementary Table 15: Results of the DESeq2 analysis of root OTUs in high MGR fields.

Table shows root OTUs with a significant (Wald test, adjusted for multiple comparison, p <0.05) differential abundance between control and inoculated samples. A negative log2FC corresponds to a higher relative abundance in control samples, whereas a positive log2FC corresponds to a higher relative abundance in inoculated samples. In addition, the values for OTU20, which likely matches the important soil predictor OTU18, are highlighted in grey.

OTU ID	baseMean	log2FC	lfcSE	Stat	pvalue	Padj	Taxonomy
OTU19	17.890	-3.163	0.465	-6.804	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU26	8.096	-3.052	0.415	-7.355	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus sp.
OTU13	10.887	-2.801	0.432	-6.488	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU38	5.362	-2.729	0.404	-6.759	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU8	37.470	-2.561	0.528	-4.855	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU5	64.335	-2.455	0.509	-4.825	0	0	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis sp.
OTU10	27.822	-2.022	0.506	-4.000	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus sp.
OTU17	4.008	-1.829	0.390	-4.687	0	0	Glomeromycetes, Glomerales, Claroideoglomeraceae
OTU24	3.506	-1.617	0.385	-4.195	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus silesianum
OTU51	2.380	-1.590	0.357	-4.451	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU28	2.476	-1.543	0.343	-4.505	0	0	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis sp.
OTU63	2.206	-1.405	0.329	-4.275	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU3	172.515	-1.197	0.515	-2.323	0.020	0.062	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis mosseae
OTU34	3.946	-1.176	0.387	-3.037	0.002	0.011	Glomeromycetes, Glomerales, Glomeraceae, Glomus, Glomus sp.
OTU11	11.000	-1.088	0.494	-2.204	0.027	0.073	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis caledonium
OTU83	2.452	-1.072	0.326	-3.288	0.001	0.005	Leotiomycetes, Helotiales, Helotiaceae, Tetracladium, Tetracladium sp.
OTU25	2.593	-1.017	0.361	-2.818	0.005	0.019	Paraglomeromycetes, Paraglomerales, Paraglomeraceae, Paraglomus
OTU104	1.677	-0.926	0.302	-3.067	0.002	0.01	Glomeromycetes, Glomerales, Glomeraceae, Funneliformis, Funneliformis sp.
OTU58	2.257	-0.919	0.310	-2.960	0.003	0.013	Dothideomycetes, Capnodiales, Cladosporiaceae, Cladosporium, Cladosporium spp.
OTU132	1.541	-0.847	0.291	-2.909	0.004	0.015	Tremellomycetes, Tremellales, Bulleribasidiaceae, Vishniacozyma, Vishniacozyma sp.
OTU98	1.721	-0.764	0.304	-2.514	0.012	0.039	Sordariomycetes, Glomerellales, Plectosphaerellaceae, Verticillium
OTU68	1.769	-0.760	0.292	-2.607	0.009	0.032	Dothideomycetes, Pleosporales, Cucurbitariaceae, Pyrenochaeta
OTU69	2.529	-0.726	0.322	-2.255	0.024	0.069	Glomeromycetes, Glomerales, Glomeraceae, Rhizophagus, Rhizophagus irregularis
OTU111	1.510	-0.721	0.287	-2.512	0.012	0.039	Leotiomycetes, Helotiales, Helotiales fam Incertae sedis, Mycochaetophora, Mycochaetophora gentianae

OTU81	2.681	-0.717	0.336	-2.134	0.033	0.082	Olpidiomycetes, Olpidiales, Olpidiaceae, Olpidium, Olpidium brassicae
OTU145	1.419	-0.656	0.272	-2.411	0.016	0.050	Leotiomycetes, Helotiales, Helotiaceae, Phaeohelotium
OTU133	1.392	-0.649	0.298	-2.178	0.029	0.076	NA
OTU150	1.328	-0.628	0.282	-2.225	0.026	0.073	Agaricomycetes, Agaricales, Psathyrellaceae, Coprinopsis, Coprinopsis vermiculifer
OTU144	1.419	-0.572	0.279	-2.053	0.040	0.095	Glomeromycetes, Glomerales, Claroideoglomeraceae, Claroideoglomus
OTU116	1.290	-0.554	0.275	-2.015	0.044	0.099	NA
OTU20	2.401	-0.640	0.338	-1.894	0.058	0.129	100% similarity with full length of soil OTU18 (Trichosporon sp.)
OTU134	1.399	0.591	0.286	2.066	0.039	0.095	NA
OTU50	1.547	0.622	0.306	2.029	0.042	0.098	Glomeromycetes, Glomerales, Glomeraceae, Dominikia, Dominikia iranica
OTU56	1.438	0.656	0.297	2.210	0.027	0.073	Glomeromycetes, Glomerales, Glomeraceae, Septoglomus, Septoglomus constrictum
OTU96	1.659	0.703	0.307	2.289	0.022	0.066	Sordariomycetes
OTU171	1.447	0.800	0.305	2.623	0.009	0.032	NA
OTU47	2.314	1.07	0.334	3.206	0.001	0.007	Glomeromycetes, Glomerales, Claroideoglomeraceae
OTU84	1.738	1.188	0.322	3.694	0	0.001	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU74	1.829	1.291	0.333	3.877	0	0.001	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU4	84.543	1.480	0.530	2.792	0.005	0.020	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU16	15.497	4.579	0.389	11.771	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU9	30.702	5.074	0.442	11.482	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22
OTU2	241.62	7.024	0.429	16.371	0	0	Glomeromycetes, Glomerales, Glomeraceae, Rhizoglomus, Rhizoglomus irregulare SAF22

Supplementary Table 16: Number of AMF OTUs (Phylum Glomeromycota) in the root samples. Table shows the number of OTUs with the taxonomic assignment "Glomeromycota" for the control and inoculated samples, as well as their difference (delta). A positive number represents an increase in AMF OTUs as response to inoculation with SAF22 (44% of the fields) and a negative number a decrease in AMF OTUs (42% of the fields). The mean change in biodiversity across all fields is 0.46.

Field	**OTU richness CTL**	**OTU richness AMF**	**delta (AMF-CTL)**
F01	14	9	-5
F04	8	13	5
F05	4	5	1
F06	8	7	-1
F07	9	4	-5
F08	11	4	-7
F09	7	5	-2
F10	10	5	-5
F12	10	10	0
F13	5	6	1
F14	1	1	0
F16	15	19	4
F19	5	3	-2
F21	11	11	0
F22	8	3	-5
F23	1	3	2
F24	2	2	0
F25	7	8	1
F26	1	1	0
F27	4	6	2
F28	2	7	5
F29	3	7	4
F30	3	3	0
F31	8	3	-5
F32	6	7	1
F33	2	7	5
F34	6	3	-3
F35	0	3	3
F36	1	1	0
F37	3	5	2
F38	8	2	-6
F39	2	5	3
F40	6	7	1
F42	3	0	-3
F43	1	6	5
F44	7	5	-2
F45	6	5	-1
F46	2	5	3
F47	6	3	-3
F48	20	11	-9
F49	18	15	-3

F50	8	11	3
F51	9	10	1
F52	22	13	-9
F53	23	18	-5
F54	18	27	9
F55	16	13	-3
F56	16	19	3
F58	20	14	-6
F59	23	26	3