

Supporting Information

Data S1. Root colonization data used for genome-wide association mapping. All metrics reflect percent of root length colonized with arbuscular mycorrhizal fungi. Mean Colonization Rate (LS Mean) is the least squares mean of root colonization for each line (genotype) when corrected for spatial gradients in the greenhouse. Maximum Colonization is the highest colonization rate of the four biological replicates of each line. Range of Colonization represents the difference in colonization rate between the biological replicate with the highest colonization rate and the lowest colonization rate for each line. The Inclusion in GWAS column indicates the lines that were included in genome-wide association analysis, after 27 lines were excluded due to high residual heterozygosity.

Data S2. Above-ground dry biomass and nutrient concentrations in dried plant tissues, measured using Inductively-Coupled Plasma Mass Spectrometry (ICP-MS) for the Core 12 lines. 'Sample Number' refers to the unique identifier given to each sample. An asterisk next to the sample number indicates that the sample had to be pooled with other replicates due to low biomass. All pooled individuals are listed with that sample. 'Line Number' refers to the genotype number within the sunflower association mapping panel. 'USDA Ames Number' refers to the accession number with the USDA North Central Regional Plant Introduction Station in Ames, Iowa. 'Derived from USDA or INRA Accession' refers to the USDA or INRA accession from which the genotype is derived via single-seed descent (see Mandel et al., 2011). 'Treatment' refers to whether the individual was inoculated by AM fungi (I, inoculated) or not (C, control). 'Bench' refers to the bench, equivalent to a replicate block, in which each individual was located. 'Above-ground dried biomass' is the dry shoot mass of each individual (grams). The nutrients analyzed were aluminum (Al), boron (B), calcium (Ca), copper (Cu), iron (Fe), magnesium (Mg), manganese (Mn), molybdenum (Mo), phosphorus (P), potassium (K), sodium (Na), sulfur (S), and zinc (Zn), presented as indicated in either ppm or % of dry mass.

Data S3. List of all genes within significant and suggestive regions. Colocate.block is the identifier for the linkage block containing each gene. Start and End indicate the position of the gene on each chromosome within the HA412-HO genome assembly v.2 (Temme et al. 2020). Locus_tag is the unique identifier for the gene within the HA412-HO genome assembly v.2. Product is the expected product as annotated by Todesco et al. 2020. Ontology_term lists all gene ontology (GO) terms associated with that gene. Chromosome indicates the linkage group on which each gene can be found.

Figure S1. Mycorrhizal colonization and biomass of wild and cultivated sunflower in low, medium, and high phosphorus conditions (see Methods S1). The low phosphorus condition was used for this study. The two cultivated sunflowers had lower colonization rates across all nutrient levels compared to the wild sunflower (two-way ANOVA, $p = 0.002$). The dried biomass produced by cultivated sunflowers was not dependent on mycorrhizal treatment but was dependent on phosphorus treatment, unlike in wild sunflower.

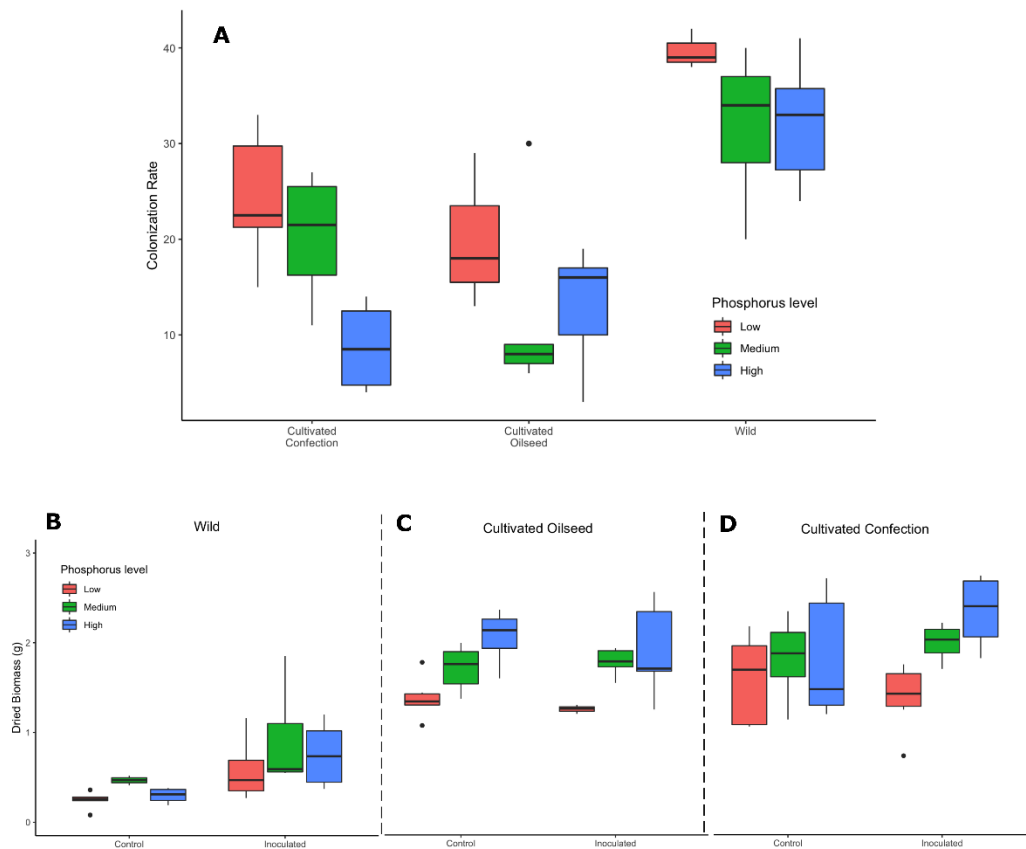


Figure S2. Microscopic images of *Helianthus annuus* roots stained with trypan blue after inoculation with *Rhizophagus intraradices*. A) Roots colonized with arbuscular mycorrhizal fungi had arum-type colonization with intercellular hyphae (H) and vesicles (V). B) Roots from uninoculated controls lacked all fungal structures. Scale bar is 100 μm .

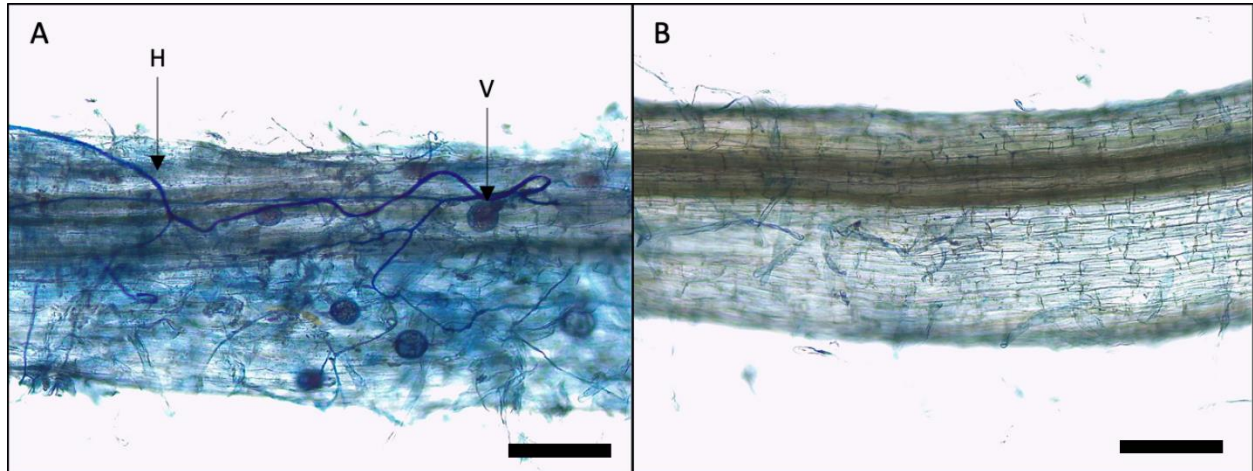


Figure S3. Validation of the method used to score AM colonization in roots against the McGonigle method (McGonigle et al. 1990). Scoring using both methods was completed by a single observer.

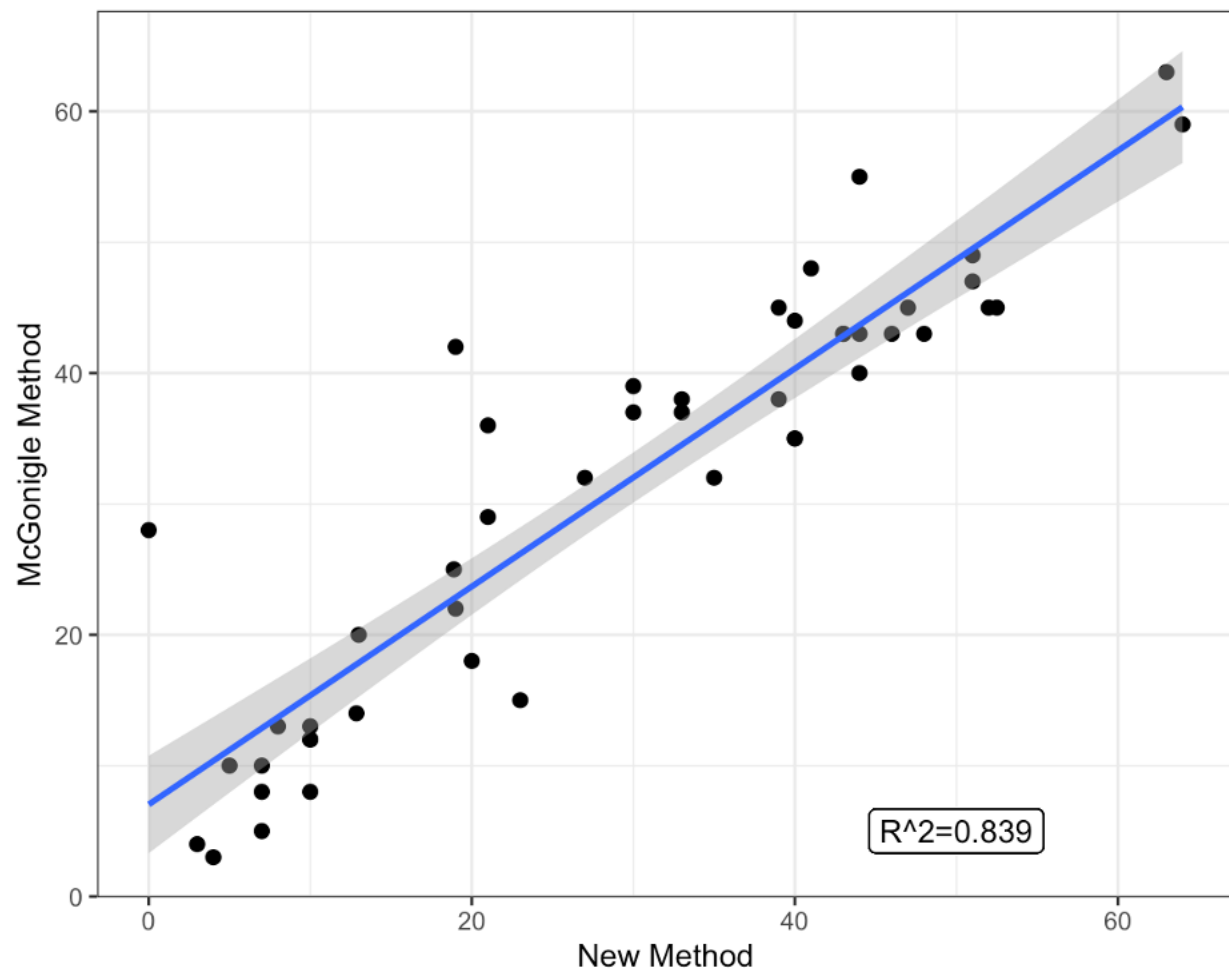


Figure S4. Mean colonization rate, mycorrhizal growth response (MGR), and shoot nutrient response of phosphorus and potassium as measured in the core 12 lines. A) MGR and colonization rate are not correlated. B) Shoot P and shoot K response are correlated with an R^2 value of 0.898. C) MGR and shoot P response are correlated with an R^2 value of 0.666. D) MGR and shoot K response are correlated with an R^2 value of 0.774. The shaded area represents the 95% confidence interval of the regression.

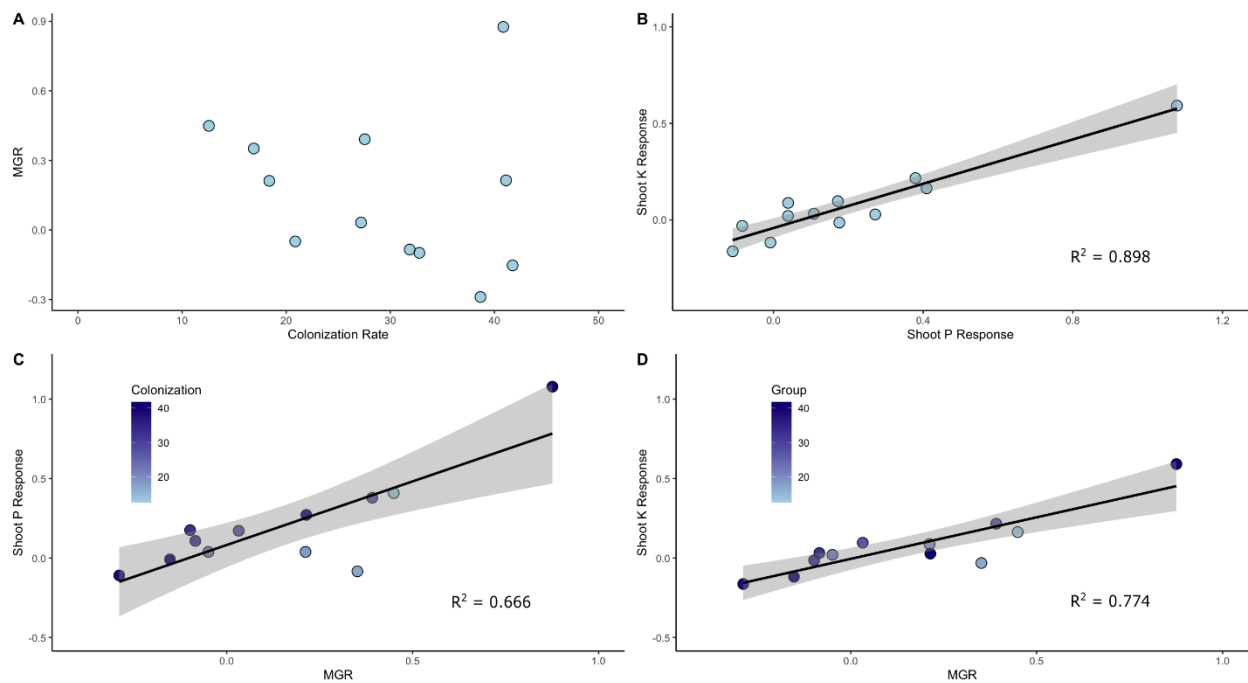
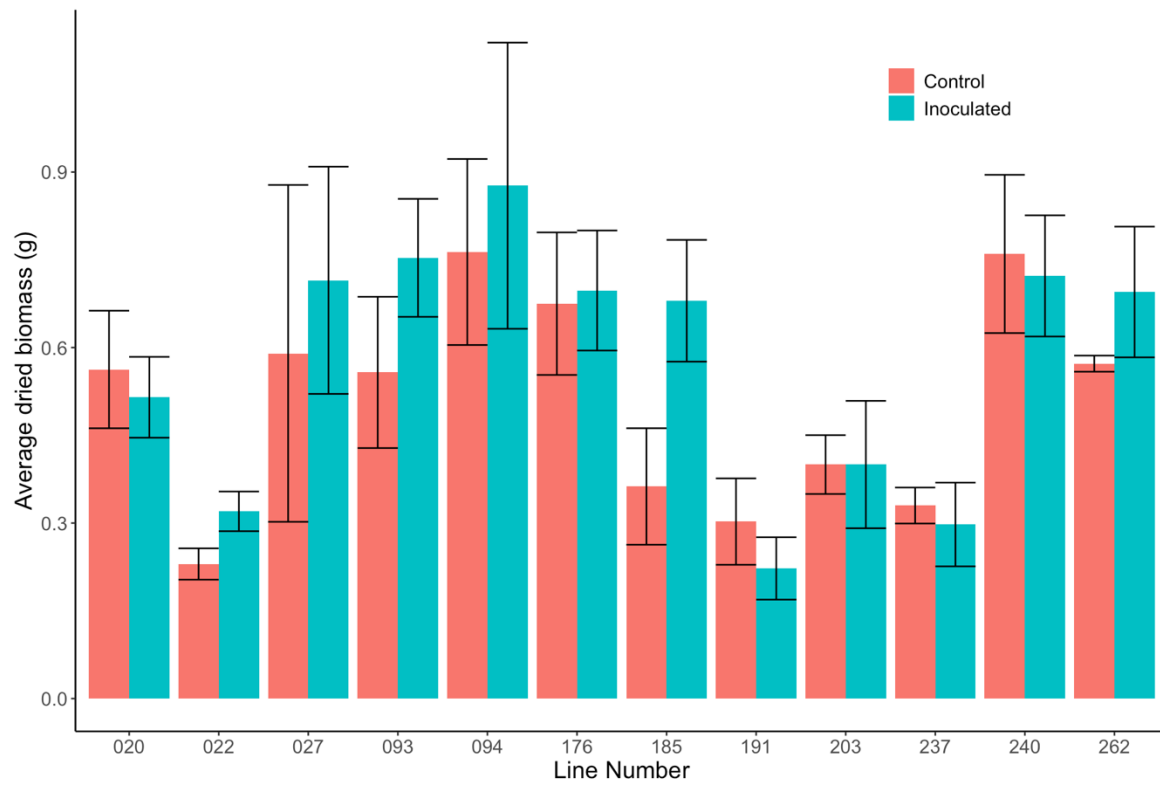


Figure S5. Mean aboveground dry biomass for the Core 12 lines under control and inoculated conditions (n= approx. 4 per line-by-treatment combination, see Data S2). Error bars are standard errors of the mean.



Methods S1

During the summer of 2017, we performed a pilot study to assess the effects of soil phosphorus on *Rhizophagus intraradices* colonization rate in both cultivated and wild sunflower. We used two cultivated sunflower hybrid cultivars, the Hornet high oleic oilseed hybrid, and the Jaguar DMR confection hybrid (Nuseed, Breckenridge, Minnesota, USA) and a wild sunflower accession (PI 673305, USDA GRIN). Seeds were surface sterilized and planted in sterile sand in 5 L tree pots (Model CP512, Stuewe and Sons, Tangent, Oregon). Mycorrhizal pots were inoculated with 10 mL of 10% *Rhizophagus intraradices* UT126, while control pots received 10 mL of sterilized sand. Plants were fertilized with either low (0.1 mM KH_2PO_4), medium (0.5 mM KH_2PO_4), or high (1 mM KH_2PO_4) phosphorus Hoagland solutions twice a week until they reached the V8 growing stage. Aboveground biomass was harvested and dried at 60°C in a forced-air drying oven until reaching a constant mass. The roots from each plant were stained using the methods of Phillips & Hayman (1970) and scored using the gridline intersect method (Giovanetti & Mosse, 1980).

References

- Giovanetti M, M. 1980.** An evaluation of techniques for measuring vesicular arbuscular mycorrhizal infection in roots. *New Phytologist* **84**:489-500.
- Phillips JM, Hayman DS. 1970.** Improved procedure for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Transactions British Mycological Society* **55**:158-161.