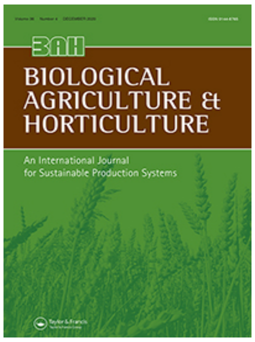

Taylor & Francis
Taylor & Francis Group

Biological Agriculture & Horticulture

An International Journal for Sustainable Production Systems

ISSN: (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/tbah20

Orchard management practices affect arbuscular mycorrhizal fungal root colonisation of almond

Christos Vasilikiotis, Meng Li, Jennifer E Schmidt, Anna Azimi, Joshua Garcia, Astrid Volder, Bruce Lampinen & Amélie CM Gaudin

To cite this article: Christos Vasilikiotis, Meng Li, Jennifer E Schmidt, Anna Azimi, Joshua Garcia, Astrid Volder, Bruce Lampinen & Amélie CM Gaudin (2020) Orchard management practices affect arbuscular mycorrhizal fungal root colonisation of almond, Biological Agriculture & Horticulture, 36:4, 230-248, DOI: 10.1080/01448765.2020.1802777

To link to this article: https://doi.org/10.1080/01448765.2020.1802777

BIOLOGICAL AGRICULTURE & HORTICULTURE
2020, VOL. 36, NO. 4, 230–248
https://doi.org/10.1080/01448765.2020.1802777

Orchard management practices affect arbuscular mycorrhizal fungal root colonisation of almond

Christos Vasilikiotis[a], Meng Li[b], Jennifer E Schmidt[b], Anna Azimi[b], Joshua Garcia[c], Astrid Volder[b], Bruce Lampinen[b] and Amélie CM Gaudin[b]

[a]Department of Agro-Environmental Systems Management, Perrotis College, 57001, Thermi, Thessaloniki, Greece; [b]Department of Plant Sciences, University of California, Davis, CA USA; [c]School of Integrative Plant Science, Cornell University, Ithaca, NY, USA

ABSTRACT

Arbuscular mycorrhizal fungi (AMF) are mutualistic fungi that play important roles in plant nutrition and soil ecosystem functions. While AMF are known to benefit diverse host plants under a range of conditions, little is known about their presence in commercial almond orchards and how frequently used management practices regulate AMF root colonisation. A large-scale survey of almond orchards in the Central Valley of California was conducted to determine the extent of mycorrhizal associations with roots and the impact of orchard management practices and soil properties on AMF root colonisation rates. The roots in all orchards were colonised, with an overall average rate of 64.4%. Organically managed orchards had higher AMF root colonisation rates (73.2%) as compared with conventionally managed orchards (62.1%), primarily due to the presence of soil vegetative cover rather than organic matter inputs. Choice of rootstock and fumigation had only marginal effects while inoculation at planting increased AMF root colonisation of young trees by 27% compared to non-inoculated control. These results highlighted the ubiquitous presence of AMF in commercial almond orchards and significant interacting influences of common management practices on AMF root colonisation under field conditions. Further research into the functional implications of mycorrhizal associations in these orchards will help guide the development of management practices that increase AMF abundance and root colonisation to improve the sustainability of this rapidly expanding industry.

ARTICLE HISTORY
Received 8 August 2019
Accepted 26 July 2020

KEYWORDS
Arbuscular mycorrhizal fungi; cover crop; fumigation; organic agriculture; *Prunus dulcis*

Introduction

Almond (*Prunus dulcis* (Mill.)) growers are increasingly recognising that while intensive water, synthetic fertiliser and pesticide inputs have allowed significant increases in yield over the last decades, such practices often deplete natural resources and may impair the ability of agroecosystems to sustain production into the future. Almond production systems are rapidly expanding across California, USA (CDFA 2018) and common management practices such as year-long bare understories, agrochemical use and low organic matter inputs (Bentley 2010; Lopus et al. 2010) are degrading soil ecosystems and the services they provide (Kroodsma and Field 2006; Schellenberg et al. 2012; Decock et al. 2017). There is an increasing need to develop management strategies that improve soil ecological function and biodiversity to maintain the environmental, social and

CONTACT Christos Vasilikiotis cvasil@afs.edu.gr Department of Agro-Environmental Systems Management, Perrotis College, 57001, Thermi, Thessaloniki, Greece
Supplemental data for this article can be accessed here.

economic performance of almond production systems. In particular, better exploitation of beneficial plant–microbe interactions such as the symbiosis between almond and arbuscular mycorrhizal fungi (AMF) could offer opportunities to enhance productivity while promoting natural soil ecological processes (Thirkell et al. 2017).

Arbuscular mycorrhizal fungi (AMF) are common and widely distributed obligate symbionts that form mutualistic associations with the roots of 80% of terrestrial plant species (Treseder and Cross 2006; Smith and Read 2008). Their wide-ranging benefits to the host plants include increased nutrient uptake (Jeffries et al. 2003), enhanced abiotic stress tolerance (Augé 2001; Schützendübel et al. 2002; Evelin et al. 2009), and improved defence against pathogens, particularly soil-borne fungal diseases (Harrier and Watson 2004; Whipps 2004; Gosling et al. 2006; Sikes et al. 2009; Krishna et al. 2010) and nematodes (Veresoglou and Rillig 2012). Mycorrhizal colonisation of almond trees has been reported to enhance plant growth and uptake of soil nutrients, especially phosphorus (Roldan-Fajardo et al. 1982). Extensive hyphal networks developed by AMF can help overcome P depletion zones that develop around plant roots due to the limited mobility of P in the soil. Few studies have reported on almonds and AMF, but AMF have been shown to be capable of colonising the rootstocks of other *Prunus* species onto which almond trees are commonly grafted (Roldan-Fajardo et al. 1982; Calvet et al. 2001, 2004). Mycorrhizal colonisation has been shown to improve tree growth, nutrient acquisition, and suppression of root-knot nematodes in grafted orchards as well (Calvet et al. 2004;. Wu et al. 2011b). In addition, AMF have been shown to improve soil structure by producing extraradical hyphae and glomalin that promoted soil aggregation (Smith and Read 2008; Wu et al. 2008; Evelin et al. 2009) and therefore enhanced critical soil ecological functions such as water and nutrient-holding capacity.

Although AMF are known to colonise almond and other *Prunus* species, very little is known about which management practices, from conventional to organic, improve colonisation and how this symbiosis can improve productivity and sustainability of commercial no-till systems. Like all mutualistic interactions, the AMF-almond mutualism is context-dependent, and the outcome of the association can be influenced by multiple management decisions. Common non-biological soil management practices such as pre-plant fumigation, herbicide application, and inorganic fertiliser inputs have been shown to decrease the abundance and diversity of AMF in soils (Kabir et al. 1997; Avio et al. 2013). The mechanisms of action of these agrochemicals may vary; for example, herbicide application was found to directly reduce fungal spore activity, while inorganic fertilisation decreased root colonisation and the abundance of active hyphae (Kabir et al. 1997). In contrast, less-intensive management practices such as addition of organic amendments, cover cropping, and reduced inorganic fertiliser inputs increased AM fungal activity by allowing more extensive hyphal network development and increased production of soil glomalin (Sattelmacher et al. 1991; Ryan et al. 1994; Mäder et al. 2000a; Kabir 2005; Gottshall et al. 2017). Location and orchard management (organic versus integrated) may also affect the diversity of native AMF communities, for example species richness was shown to be significantly higher in organically managed apple orchards than in integrated ones (Turrini et al. 2017a). Host plant species and variety as well as mycorrhizal fungus genotype also affected the outcome of the interactions: root colonisation of *Prunus* rootstock cultivars inoculated with AM fungi varied from 7.6% to 69.6% depending on the rootstock and mycorrhizal species (Calvet et al. 2004; Wu et al. 2011a). Despite strong evidence of the effects of individual management decisions on AMF, few studies have measured root colonisation under field conditions in semi-arid irrigated systems where multiple management practices were applied simultaneously. This would be a critical first step for harnessing the benefits of AMF for productivity and environmental quality.

This knowledge gap was addressed by conducting a survey of AMF root colonisation of almonds under varying management practices in commercial orchards and research trials across California. The influence of popular almond orchard management practices was examined, including fertility management, cover crops, organic amendments, choice of rootstock, pre-plant fumigation, and AMF inoculation, as well as soil chemical properties on AMF root colonisation. By elucidating how common management practices, including some biological approaches, affect the AMF-almond symbiosis, the results of this survey will inform the design of management strategies that capitalise on AMF to increase productivity and improve soil health in almond orchards.

Materials and methods

Experimental sites and soil sampling

Fifteen orchards, ranging from 1.5 to 20 years old, were selected across the Central Valley of California, USA (Figure 1). These sites represented a large management gradient and included both commercial farms and research trials (Table 1). Three orchards were managed organically; site 1 has been certified by CCOF (Santa Cruz, California, USA) since its establishment in 2006; site 6 has been managed following organic practices since 2006, but was not certified, while site 7 has been

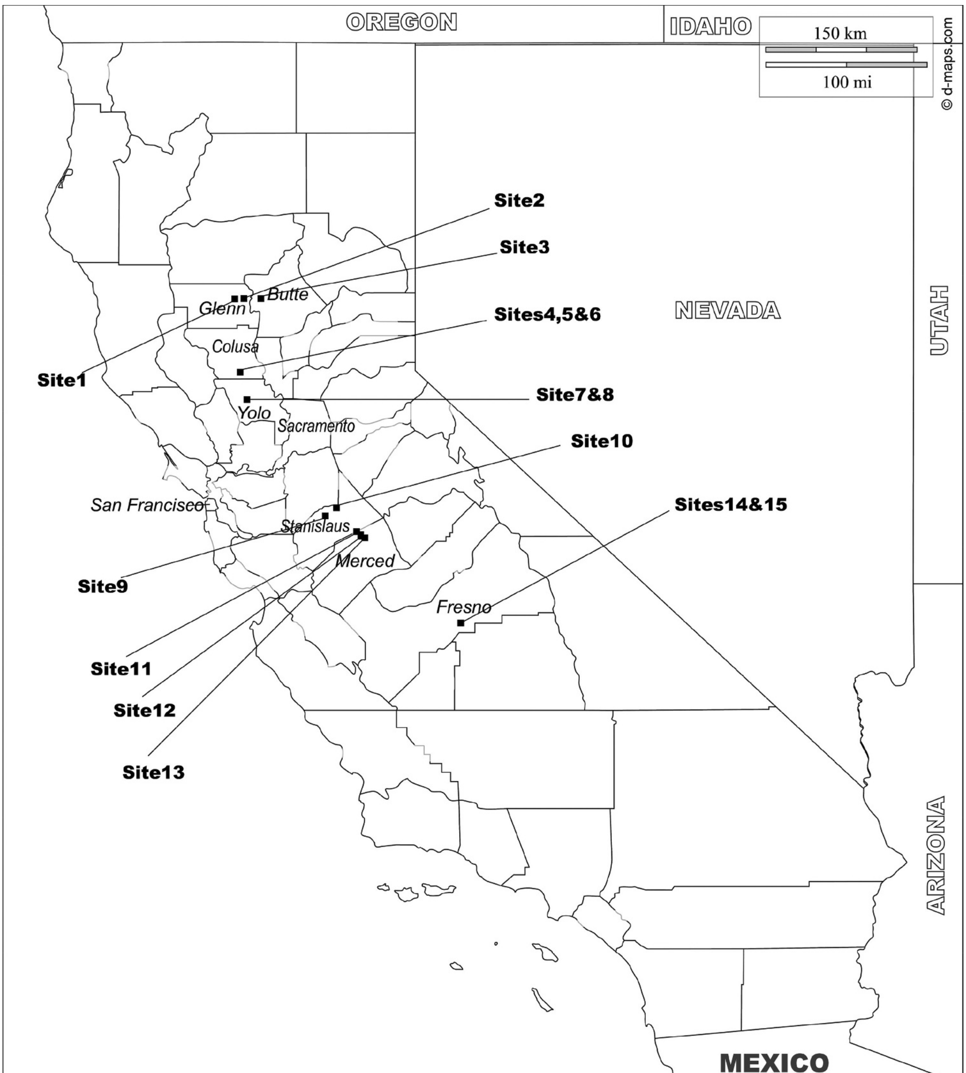

Figure 1. Map of study area and locations of surveyed almond orchards. Site numbers correspond to Table 1. County lines are shown and counties containing research sites are labelled.

Table 1. Management and treatments applied at the commercial and the research orchards surveyed.

Site	Site information	Management system	Rootstock	Year trees established	Management	Precipitation (mm)[b]	Mean AMF root colonisation rates (% ± SE)
Organic/conventional paired sites							**Av. 67.92**
Site 1	Commercial farm	Organic	Nemaguard	2006	Planted cover crop, grazing, compost, no fumigation	533	76.42 ± 2.67
Site 2		Conventional	Nemaguard	1996	Bare soil, compost, fumigation	672	65.74 ± 4.12
Site 5	Research station	Conventional	Lovell	2006	Bare soil, no compost, no fumigation	380	47.65 ± 4.05
Site 6		Organic	Lovell	2006	Resident vegetation, compost, no fumigation	380	69.34 ± 4.13
Site 7	Commercial farm	Organic	Lovell	2008	Planted cover crop, grazing, compost, no fumigation	533	73.70 ± 3.07
Site 8		Conventional	Lovell	2001	Bare soil, no compost, fumigation	533	74.65 ± 3.74
Research trials							**Av. 65.02**
Site 9	Organic amendment trials	Conventional	Nemaguard	2011	Bare soil, ± compost, no fumigation	317	73.88 ± 1.82
Site 10		Conventional	Nemaguard	2014	Bare soil, ± green waste compost, ± manure compost	355	34.21 ± 3.67
Site 4	Rootstock trial	Conventional	Atlas, Lovell, Nemaguard, Nickels	1997	Bare soil, no compost, fumigation	380	72.54 ± 2.31
Site 13	Rootstock x Fumigation trial	Conventional	Nemaguard, Hansen 536, Empyrean-1	2014	Bare soil, no compost, ± fumigation (chloropicrin 35% and 1,3-dichloropropene 65% (Telone C-35))	381	69.05 ± 3.62
Site 12	Fumigation trial	Conventional	Nemaguard	2011	Bare soil, no compost, ± fumigation (Telone C-35, Methyl bromide)	312	76.60 ± 2.12
Site 14		Conventional	Nemaguard	2015[a]	Bare soil, no compost, ± fumigation (ASD, Telone C-35)	317	65.24 ± 2.15
Site 15	Replant trial	Conventional	Nemaguard	2009	Bare soil, no compost, spot fumigation Telone C35 (0.5 m diameter only around tree sites)	317	63.62 ± 4.93
Others							**Av. 51.73**
Site 3	Commercial Farm	Conventional	Marianna	1991	Bare soil, no compost, no fumigation	584	39.98 ± 5.40
Site 11		Conventional	Nemaguard	2015[a]	Bare soil, compost, ± inoculation at planting	330	63.48 ± 3.82

Notes: [a] Non-bearing orchards; [b] 10-year annual precipitation average managed organically since 2010 and certified by Yolo Certified Organic Agriculture (Woodland, California, USA).

At sites 1, 6 and 7 a vegetative soil cover was maintained, while the remaining sites kept a vegetation-free orchard floor. Compost was added as soil amendment at sites 1, 2, 6, 7, 9 and 11, while site 10 comprised of a trial comparing the use of green waste compost with manure as organic soil amendments.

Pre-plant chemical fumigation with dichloropropene (Telone C-35®) had been used at sites 12, 13, 14 and 15 and methyl bromide at site 12 (Table 1). Anaerobic soil disinfestation (ASD), a non-chemical alternative to prevent Prunus replant disease that involved covering and saturating the soil with water for 6 weeks after the incorporation of wood chips to induce anaerobic soil conditions (Browne et al. 2017), had also been tested at site 14 (Table 1). A pre-plant whole orchard grinding and recycling trial took place at site 15 and more information about the management of the orchard has been described by Jahanzad et al. (2020).

The rootstocks used are shown in Table 1. At most sites, the Nonpareil almond variety had been grafted to the rootstock, except for at site 03 (Mission/Padre), site 11 (Independence) and site 14 (P16.013). At all sites, ten trees per treatment and replicate were randomly selected along a transect and three intact soil cores per tree were taken and composited. Fine root and soil samples were collected from 0 to 30 cm depth, 100 cm away from the tree within the drip or microsprinkler irrigation zones to capture the tree root zone, from May through June 2016. Two of the sites (Site 7 and 8) were also sampled in September 2015. Soil samples containing roots were stored at 4°C prior to processing and analysis.

AMF root colonisation

Roots were separated by sieving the soil through a 1 mm sieve and stored in 50% ethanol. Only roots of almond trees were present at the time of sampling and very limited resident vegetation cover existed. Roots were cut into 2 cm segments, cleared with KOH and stained using vinegar and Schaeffer black ink (Vierheilig et al. 1998). AMF root colonisation was assessed using the gridline intersection method (Giovannetti et al. 1980) under a dissecting microscope with 50 to 100X magnification. Arbuscules, vesicles, hyphae (connected to arbuscules/vesicles or non-connected) were observed on 100 intersects per sample. Percent root length colonised was determined as the proportion of root intersects containing any AM structure and connected hyphae (McGonigle et al. 1990).

Soil analysis

Composited soil samples from each orchard were analysed at the University of California, Davis Analytical Laboratory. Total nitrogen (N) and carbon (C) were determined via combustion analysis in accordance with the AOAC Official Method 972.43, Microchemical Determination of Carbon, Hydrogen, and Nitrogen, Automated Method 1997 (AOAC 1997). Soil nitrate was measured via flow injection analyser (Knepel 2003). Extractable phosphorus (P) was determined according to Olsen and Sommers (Olsen and Sommers 1982). Soil exchangeable potassium (K), calcium (Ca), magnesium (Mg), and sodium (Na) were measured using an ICP-AES (Thomas 1982). Organic matter content was determined via the loss-on-ignition method (Nelson and Sommers 1996). Soil pH was measured on a saturated paste extract.

Statistical analyses

Analysis of variance (ANOVA) were performed to determine the impact of treatments on AMF root colonisation rates at each site using JMP Pro v.12. Treatment effects and tree age were analysed separately as fixed effects at each paired location with repetitions as random factors when applicable. All sites were included in the analysis of the effects of ground cover. For paired analysis of organic and conventional sites (Table 1), the difference between means at paired sites was compared to the variability within each treatment group. Results of replicated trials were also analysed separately. For all analysis, residuals were checked for normality and heterogeneity and data were arcsine transformed prior to analysis as necessary to meet the assumptions of ANOVA. Post hoc mean comparisons were conducted using Tukey's HSD test ($p \le 0.05$). Changes in soil chemical properties as a function of orchard floor management strategies (i.e., bare ground, cover crop, and resident vegetation) were visualised with non-metric multidimensional scaling (NMDS) using the *metaMDS* function of the vegan package (Oksanen et al. 2017). Correlations between soil chemical properties and AMF root colonisation data were calculated using linear regression and Pearson correlation coefficients. Principal component analysis (PCA) was conducted to investigate and visualise the relationships between AMF root colonisation rate and soil physical and chemical properties across all surveyed almond orchards. PCA was performed using the *prcomp* function of the statistical package in R 3.4.1 (R Core Team 2018).

Results

Roots were colonised by mycorrhizal fungi at all sites surveyed, but mean AMF root colonisation rates differed greatly across locations and treatments. AMF root colonisation rates ranged from 34% on the rootstock in a conventionally managed orchard with a sandy loam soil and 355 mm precipitation to 76.6% in a conventional orchard with a silt loam and 312 mm precipitation, on the similar Nemaguard rootstock (Table 1). Fertility management, vegetative cover, inoculation with commercial AMF products, and soil P and N significantly impacted the extent of AMF root colonisation.

Soil vegetative cover promoted stable root colonisation by AMF

Orchard management system was found to be a major factor regulating mycorrhizal associations in almond (Table 1). In general, AMF root colonisation was 11.1% higher in organically managed orchards than in conventionally managed orchards (Figure 2a, $p = 0.0012$). A long-term replicated trial comparing organic to conventional management over a 10-year period showed significantly higher AMF root colonisation in the organically managed trees (69.3%) compared to the conventionally managed trees (47.7%) (sites 5 and 6, Table 1). Management system and sampling date had interactive effects such that AMF root colonisation was stable across consecutive seasons in an organically managed orchard (~72%, site 07), but AMF root colonisation was reduced in the conventional paired orchards in late summer (55% late summer, 72% early spring, site 08) (Figure 2b). The two orchards had similar rootstocks, soil types and precipitations. Tree age was different, but no significant correlations were found between year of orchard establishment and AMF root colonisation rates across this study ($R^2 = 0.01$, $p = 0.49$).

The organic management encompassed a wide array of practices. All organic orchards surveyed had winter soil vegetative cover composed of either planted cover crops or resident vegetation, in contrast to the chemically-controlled bare soil found in most of the conventional orchards. Presence of soil vegetative cover was found to be the most significant factor in enhancing mycorrhizal associations. When comparing impacts of cover crops or resident vegetation vs bare ground across all orchards surveyed (Table1), orchards with winter soil cover had a significantly higher AMF root colonisation rate (75.1%) compared with vegetation-free orchards (60.1%) (Figure 3a). Presence of soil vegetative cover significantly influenced soil physical and chemical properties (Supplemental Table 1), such that soils with winter soil cover tended to have similar soil physicochemical properties across the sites surveyed (Figure 3b).

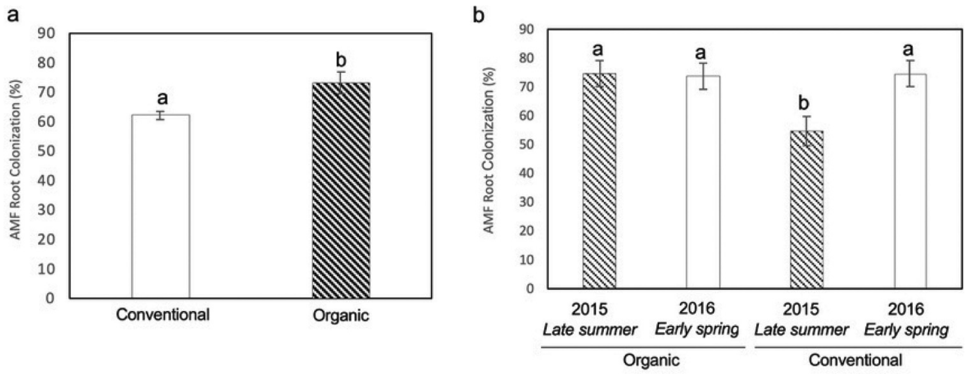

Figure 2. Impact of organic management on AMF root colonisation rates. Means and standard errors are shown to compare the impacts of (a) orchard management system ($n = 30$, 3 paired orchards with 10 samples) and (b) different sampling season ($n = 10$, site 5 and 6 long term trial). Bars with the same letter do not differ according to Tukey's test ($p = 0.05$).

Type of organic input influenced AMF root colonisation

The location had a significant effect on differences in AMF root colonisation rates comparing the two sites where the use of compost was tested (sites 10 and 9) ($p < 0.001$). At site 10, the annual addition of green waste compost for 2 years resulted in significantly higher AMF root colonisation rates than the addition of composted manure, but neither form of organic amendment significantly increased AMF root colonisation compared to non-amended trees (Figure 4a, $p = 0.0315$). Despite a higher basal level of AMF root colonisation, similar results were observed in the other compost trial at site 9 (Figure 4b). Similarly, the one-time large organic matter input in the form of pre-plant whole orchard grinding and recycling did not enhance mycorrhizal associations 10 years after incorporation compared with the control where the trees had been burnt (Figure 4c).

Chemical fumigation and anaerobic soil disinfestation did not inhibit AMF root colonisation

Pre-plant chemical fumigation with common fumigants such as dichloropropene (Telone C-35®) or methyl bromide was not found to inhibit mycorrhizal associations as soon as 1 year after establishment at two replicated fumigation trials (sites 12 and 14) on Nemaguard rootstocks (Table 1, Figure 5a). Anaerobic soil disinfestation (ASD) at site 14 tended to reduce AMF root colonisation rates, although the effect was not significant (Figure 5a). Interestingly, however, the impacts of fumigation

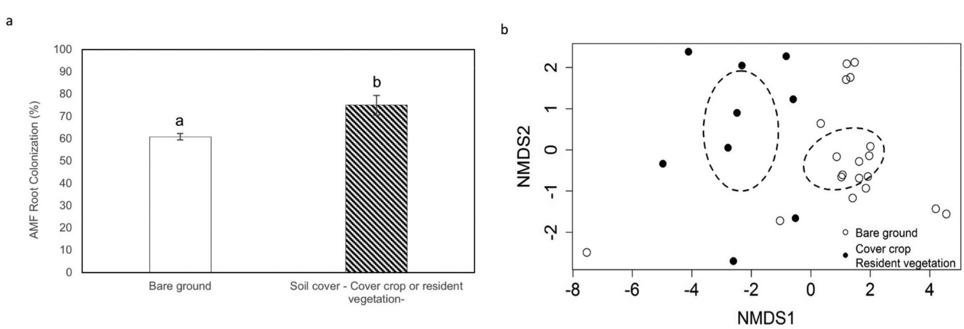

Figure 3. Relationship between soil cover, soil properties, and AMF root colonisation rate. (a) AMF root colonisation rates across all bare orchards and orchards with soil cover surveyed ($n = 15$, 15 orchards with 10 samples per treatment). Means and standard errors are shown. Bars with the same letter do not differ according to Tukey's test ($p = 0.05$). (b) Nonmetric multidimensional scaling (NMDS) ordination of soil properties under different floor management practices. Soil properties are shown in Supplemental Table 1.

on AMF root colonisation were found to be rootstock-dependent. Fumigation had a positive impact on AMF root colonisation of Hansen 536 rootstocks (+28%) while no differences were observed in other rootstocks (Figure 5b).

Inoculation at planting increased AMF root colonisation rates in newly planted orchards

AMF inoculation after fumigation is a common practice when planting new conventional orchards, but AMF root colonisation rates are rarely evaluated using a systematic and replicated approach. It was observed that inoculation of saplings with a commercial AMF preparation (site 11) resulted in a statistically significant increase of 15% in AMF root colonisation rates within the first year of planting, with 71% AMF root colonisation of inoculated trees as compared to 56% AMF root colonisation of uninoculated trees by endogenous AMF (Figure 5c, $p = 0.0455$).

Rootstocks only had a marginal effect on AMF root colonisation rates

Almond trees grown as part of two conventionally managed rootstock trials established in 1997 (site 4) and 2014 (site 13) were surveyed. The location had a significant effect on differences in AMF root colonisation rates between rootstocks ($p < 0.001$). At site 4, almond trees on Atlas rootstock tended to have a lower AMF root colonisation rate (62%), but differences with other rootstocks were not significant. At site 13 where the trees had been most recently established, trees on Nemaguard rootstocks had the lowest AMF root colonisation rates (57.3%), which was significantly different to that of the highly colonised Empyrean-1 rootstock (80.5%) (Table 2).

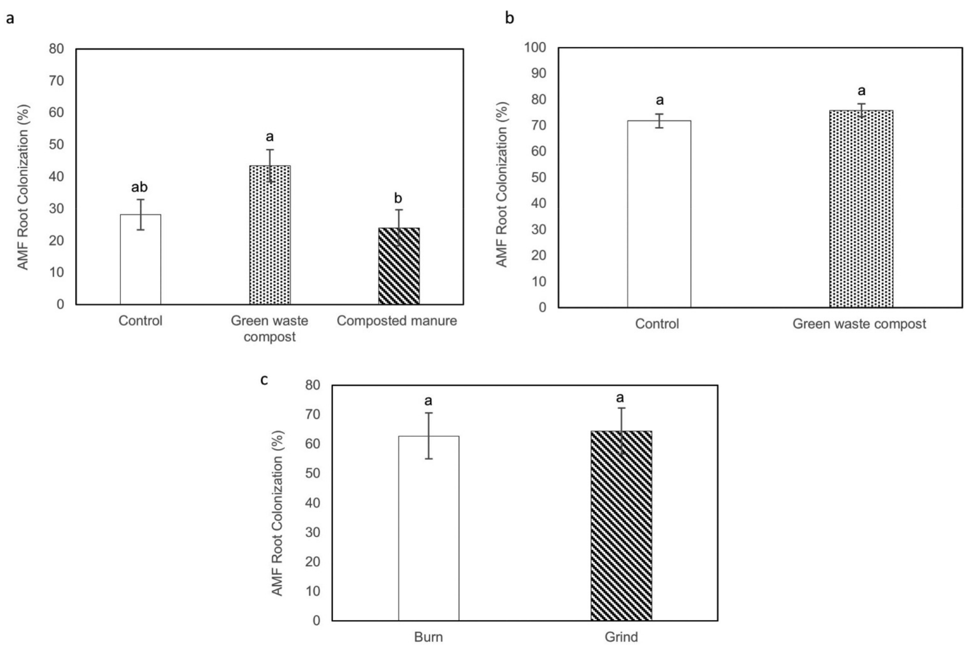

Figure 4. Impact of organic inputs on AMF root colonisation rates. (a-b) Compost trials established in (a) 2014 (site 10), and (b), 2011 (site 9). (c) Long-term impact of whole orchard grinding and recycling vs. burned control prior to almond replanting (site 15). Means and standard errors are shown. Letters indicate significant differences and bars with the same letter do not differ ($p = 0.05$).

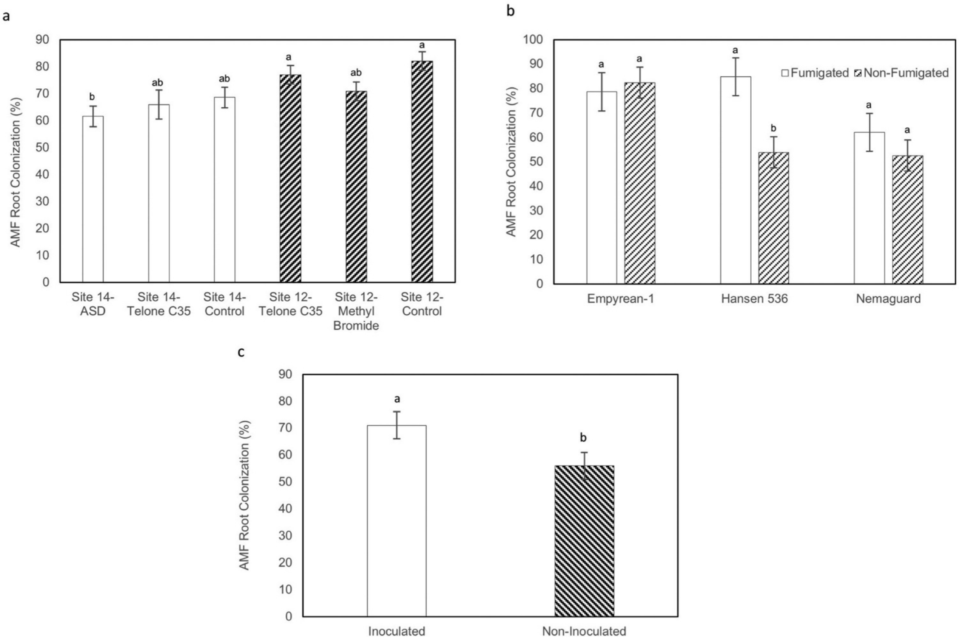

Figure 5. Impact of pre-plant fumigation and inoculation on AMF root colonisation. Means and standard errors are shown for (a) two trials testing chemical fumigants and ASD (white bars indicate site 14 and grey bars indicate site 12; see Table 1 for more details); (b) interaction between rootstocks and chemical fumigation; (c) AMF root colonisation post-inoculation compared to non-inoculated control (Site 11). Letters indicate significant differences and bars with the same letter do not differ (p = 0.05).

High soil phosphorus and nitrate concentrations inhibited AMF root colonisation

AMF root colonisation rate was significantly negatively correlated with soil phosphorus and nitrate concentrations based on Pearson's correlation coefficient ($r = 0.69$, $p < 0.0001$ and $r = 0.47$, $p < 0.05$, respectively) (Table 3). No significant correlation was detected with organic matter levels (Table 3). The first and second principle components (PCs) explained 46.7% (PC1) and 18.9% (PC2) of the variation among samples (Figure 6). PC1 was primarily affected by soil OM, and soil Ca, and PC2 was positively associated with soil nitrate-N and K, but negatively associated with soil Na. Principal Component analysis confirmed that P and nitrate-N were closely related and had a negative impact on AMF root colonisation (Figure 6). This inverse relationship between soil P and AMF root colonisation was especially evident at site 10, which had the lowest AMF root colonisation rate

Table 2. Impact of rootstock on mean AMF root colonisation rates at two research trials.

Site	Rootstock	Mean AMF root colonisation rate (%)
Site 04	Nemaguard	78.76 ± 3.97 a
	Lovell	73.32 ± 3.97 a
	Nickels	71.82 ± 3.97 a
	Atlas	62.07 ± 5.13 a
Site 13	Empyrean-1	80.51 ± 5.02 a
	Hansen 536	69.33 ± 5.02 ab
	Nemaguard	57.32 ± 5.02 b

Notes: Means ± standard errors are shown (n = 10). Means, within each site, followed by the same letter were not significantly different at p = 0.05. See Table 1 for details of the research sites sampled.

Table 3. Linear correlations between AMF root colonisation and soil properties.

Parameter	R^2	*p*-value
N (Total)	−0.2178	0.2656
C (Total)	−0.2737	0.1587
NO_3-N	−0.4723	0.0112 *
Olsen-P	−0.6897	< 0.0001 *
X-K mg kg^{-1}	−0.2211	0.2583
X-Na mg kg^{-1}	0.2317	0.2355
X-Ca	−0.2163	0.2688
X-Mg	−0.1739	0.3763
OM (LOI)	−0.277	0.1535
pH	0.0111	0.9552

Note: * indicate significant at p = 0.05.

(34%, Table 1) and the highest soil P concentration (106 mg kg^{-1} Supplemental Table 1). However, at site 9, which also had high soil P concentration and high nitrate-N, AMF root colonisation was not affected and AMF root colonisation rates were found to be high in the green waste compost treatment (76%; Figure 4b and Supplemental Table 1). OM was also found to be high in these orchards. Overall, AMF root colonisation rates were less affected by soil texture (sand, silt, and clay contents).

Discussion

This study was the first systematic survey of AMF root colonisation in almonds as impacted by common conventional and biological management practices. The results confirmed the ubiquitous presence of AMF in almond orchards and high average AMF root colonisation (64%) across almond rootstocks. It was shown that management practices such as presence of a vegetative cover during the winter (cover crop or resident vegetation) mostly implemented in organic orchards strongly influenced AMF root colonisation, likely through its impact on carbon flow from multiple plant hosts and soil properties (Figure 3, Supplemental Table 1). Other management decisions such as organic matter inputs, choice of rootstocks and fumigation had marginal and inconsistent effects on AMF root colonisation (Figures 4 and 5; Table 2).

Overall, the adoption of organic management practices promoted AMF root colonisation. The three organic sites had high AMF root colonisation rates, ranging from a minimum of 69.3% to a high of 76.4%. AMF root colonisation rates at sites with conventional management were more variable, ranging from 34.2to 76.6%, but averageing at 62.1%. At the three paired sites, AMF root colonisation was higher in the organic orchards at two of the paired sites with identical microclimate, but not at the other (Table 1). However, the site where organic management did not increase AMF root colonisation already had a very high AMF root colonisation rate in the conventional treatment (74.7 %, Table 1). Although the variability in management practices was high between the organic orchards and studies, the results concurred with several studies across a wide range of crops and regions that report generally higher AMF root colonisation rates for organically managed fields compared to conventionally managed ones (Sattelmacher et al. 1991; Ryan et al. 1994; Mäder et al. 2000b, 2002; Bedini et al. 2013; Van Geel et al. 2016).

AMF root colonisation might have been higher in organic systems for a number of reasons. Firstly, the winter vegetative cover that was present in all organic orchards surveyed likely enhanced AMF root colonisation through positive effects on C inputs and soil physical and chemical properties (Gómez et al. 2004, 2009) (Figure 3). Cover crops can act as hosts to AMF during winter, helping maintain populations that can colonise and benefit subsequent plantings (Kabir 2005) and serving as sources of AMF propagules for successive crops (Kabir and Koide 2002; Karasawa and Takebe 2012; Lehman et al. 2012). The presence of continuous vegetative cover may have accounted

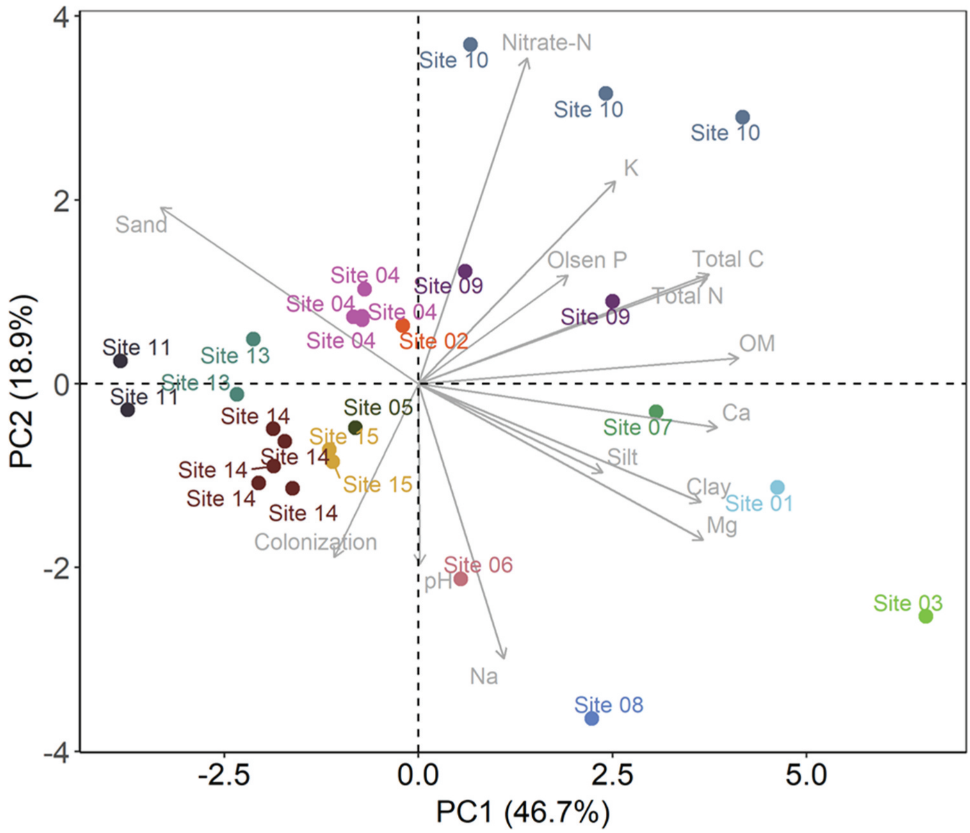

Figure 6. Principal component analysis (PCA) biplot of soil physicochemical properties and AMF root colonisation rates across surveyed almond orchards. Different colours represent different sampling sites. Percentages in parentheses indicate the variance explained by each axis.

for the consistently high AMF root colonisation across seasons in the organically managed orchards, in contrast to the seasonal dynamic observed in the conventionally managed orchard (Figure 2b). Cover crops can also increase the abundance and diversity of rhizosphere microbial populations that have synergistic effects with AMF (Fitter and Garbaye 1994; Moreno et al. 2009). The greater concentration of roots in cover-cropped orchards would increase the availability of low molecular weight exudates and carbon compounds from root biodegradation, which can represent a significant source of carbon and nitrogen for soil microorganisms (França et al. 2007). As such, beneficial effects of plant covers have been reported on soil biological activity, including mycorrhizal inoculum potential and soil respiration in orchard systems (Moreno et al. 2009; Turrini et al. 2017b). Cover crops may also mitigate the potential impacts of poor soil physical properties on soil microbial populations by regulating soil temperatures, mitigating anoxia in wet winters by improving drainage and removing water via transpiration or decreasing compaction (Folorunso et al. 1992; Kahimba et al. 2008).

Secondly, the lower AMF root colonisation in conventionally managed systems may have been due to the use of fumigants, fungicides, and herbicides that have been shown to inhibit AMF or other soil organisms (Kabir et al. 1997; Avio et al. 2013; Gaupp-Berghausen et al. 2015). Although not directly tested in this study, herbicides used in the conventional orchard may have decreased AMF root colonisation potential by reducing arbuscule formation and AMF spore viability (Druille et al. 2013a, 2013b, 2016). Repeated applications of glyphosate have been shown to decrease AMF root colonisation (Ronco et al. 2008; Druille et al. 2013a, 2013b), likely by directly affecting AMF structures and spores or damaging other soil microorganisms essential to AMF development (Garbaye 1994). Similarly, the chemical simazine has been shown to reduce the population of mycorrhizae propagules (Granger et al. 1995). Agrochemicals may also impact AMF indirectly by reducing populations of other soil organisms, such as earthworms, that interact synergistically with AMF populations and their beneficial effects on plant hosts (Zaller et al. 2014; Cao et al. 2015; He et al. 2018). An increasing number of studies have shown a detrimental effect of herbicides or fungicides on earthworm activity, development, reproduction and lifespan (Correia and Moreira 2010; Gaupp-Berghausen et al. 2015; Stellin et al. 2018). Interestingly, in this study, fumigation with various biocides prior to planting of conventional orchards was found to have no impact on AMF root colonisation in orchards of different ages (Figure 5). It is possible that the removal of antagonistic microorganisms through fumigation may level the playing field and allow for rapid root recolonisation by mycorrhizae. The mechanisms behind the effect of fungicides on AMF root colonisation are not well understood, as fungicide applications have been shown to result in decreased, unaffected, or increased AMF root colonisation rates, especially at reduced application rates (Gosling et al. 2006). The effect of fungicides on AMF has also been shown to be species-dependent, and both positive and negative effects on mycorrhizal population parameters have been reported. The fungicide benomyl has also been shown to reduce AMF mycelial growth and mycelial morphology, interconnectedness and viability (de Novais et al. 2019). On a peach almond rootstock, *G. mossae* was found to be affected negatively by metalaxyl and propamocarb, while *G. intraradices* was unaffected (Fontanet et al. 1998). Fosetyl-Al markedly inhibited root growth and AMF root colonisation in another study (Sukarno et al. 1998). In other studies, the fungicides captan, benomyl, and PCNB were found to decrease percent AMF root colonisation and AM metabolic activity for *G. etunicatum, G. mosseae* and *G. rosea* in pea plants, while effects on spore abundance varied among AM species (Willis et al. 2013). Plant-driven rhizosphere processes likely contribute to regulation of AMF root colonisation, which is not always correlated with spore abundance in the soil (Gosling et al. 2010).

For almond, the results of this study showed that rootstocks only had a marginal effect on AMF root colonisation rates (Table 2). Previous greenhouse studies evaluating AMF root colonisation of 18 *Prunus* sp. rootstocks by various fungal species showed that rootstocks can differ widely in their ability to associate with specific mycorrhizal isolates (Calvet et al. 2004). Although the mechanisms are unclear, the outcome of the AMF-rootstock symbiosis has been shown to vary in a genotype-dependent matter (Calvet et al. 2004; Wu et al. 2011a). While rootstocks may not have affected AMF root colonisation across all conditions, the observation that choice of rootstock affected the response to fumigation (Figure 5b), highlights the importance of rootstock-AMF-management interactions. Host plant genotype is known to be a primary factor shaping AMF community composition (Turrini et al. 2018) and other studies in *Prunus* sp. have confirmed that AMF isolates have different root colonisation abilities (Wu et al. 2011b). Given the previously described variation in responses to agrochemical inputs by diverse AMF taxa, host rootstocks that increase or decrease the abundance of sensitive taxa may have significantly different root colonisation rates under long-term management involving fumigation or pesticides. Differences in endogenous AMF populations across sites may also explain some of the variations observed.

Interestingly, the addition of organic matter common in organic systems in the form of composted manure and green waste compost did not significantly impact AMF root colonisation, and soil organic matter levels were not correlated with AMF root colonisation rates (Figures 4 and 6; Table 3). Continuous addition of P-rich organic amendments such as manures may have increased plant-available soil P, known to be a primary factor regulating AMF establishment, thus negating potential positive impacts of organic carbon and N source on AMF (Jordan 2000; Gosling et al. 2006). Such trends were observed across the trial where high soil phosphorus and nitrate concentrations were negatively correlated with AMF root colonisation (Table 3, Figure 6). Trees grown in P- and N-rich soils would likely preferentially allocate resources to shoots and leaves rather than roots, potentially reducing belowground biomass and investment in the AMF symbiosis

(Johnson et al. 2013). High fertilisation rates can also reduce spore density and the development of external AMF mycelia (Gryndler et al. 2006), decreasing the potential benefits of association with AMF. The dominant forms of soil P and their rate of release, combined with the availability of other nutrients such as N, might therefore be a critical factor influencing AMF root colonisation dynamics (Thirkell et al. 2016). High soil P may partly explain the non-significant effects of the addition of organic matter on AMF root colonisation at these sites (Johnson et al. 2015). Another factor affecting AMF root colonisation dynamics might be the quality of composted organic amendments added. In a long-term study comparing organic and conventional farming systems, the quality of the manure played a significant role in AMF root colonisation (Oehl et al. 2004). Organically managed fields fertilised with aerobically composted farmyard manure had a higher AMF diversity and plant infection potential compared to conventionally managed fields with anaerobically rotted farmyard manure and mineral fertilisers (Oehl et al. 2004). The quality of compost might be related to the humic acid content. Addition of humus-rich composts was found to significantly increase AMF root colonisation and lettuce crop growth (Solaiman et al. 2019).

Apart from the effect of organic management on AMF root colonisation, it was found that inoculation with a commercial AMF isolate increased root colonisation of newly planted orchards to more than 70% (Figure 5c). Inoculation with mycorrhizal population blends at planting is a frequent practice to counteract the potential short-term negative impacts of fumigation and deep soil tillage on beneficial soil microbes. Young peach-almond hybrid and peach rootstocks, both used as almond rootstocks, have been shown to respond well to mycorrhizal inoculation and inoculation can increase AMF root colonisation in agricultural soils, which may have low indigenous AMF diversity and abundance (Calvet et al. 2004; Verbruggen et al. 2013). Inoculation may be less successful in mature trees if the inoculum potential is already high and rhizosphere communities are well-established. Inoculation of seedlings allows the AM fungi to become established in the root system early, gaining an advantage over other soil-borne species (Smith and Read 2008). However, the differences observed in this study might only have had importance for the initial development of the trees and their roots, since older fumigated orchards often achieved comparable AMF root colonisation rates.

The functional significance of increased AMF root colonisation for tree growth and yield at varying levels of input and potential benefits to agricultural sustainability remains to be established in commercial orchards. A recent meta-analysis found that greater root colonisation by AMF was correlated with higher plant biomass and soil P uptake, especially in non-N fixing woody plants (Treseder 2013). Young olive trees inoculated with commercial or indigenous AMF also exhibited significantly higher P concentrations in leaves, stems and roots compared with non-inoculated controls (Briccoli Bati et al. 2014). AMF root colonisation rates above 75% increased AM-induced stomatal conductance by 46% on average (Augé et al. 2015), suggesting the potential for greater benefits of enhanced AMF root colonisation above current levels observed in this survey in the water-limited irrigated Central Valley of California. These results indicated that AMF could likely improve plant nutrition and water relations in conventionally managed commercial almond orchards.

Furthermore, increasing evidence that AMF can mobilise nitrogen from organic sources in the soil and transfer it to its associated host plant (Hodge et al. 2001; Hodge and Fitter 2010; Veresoglou et al. 2012) suggests that mycorrhizal associations could be especially important for organic almond production, where N is primarily obtained from composted amendments and other organic matter. Nonetheless, given research showing that mycorrhizae benefit crops most in lower-fertility soils (Roldan-Fajardo et al. 1982), it remains unclear how intensive production agriculture and interacting management practices might alter the magnitude or direction of the functional effects. Long-term compost applications, which can result in high soil P and N concentrations, was shown to reduce AMF root colonisation of maize roots (Liu et al. 2019). The effect of added compost depended on the growth stage of the crop, with the effect being most prominent at the 13-leaf collar stage of maize. While there is some evidence of a negative effect of compost additions on

AMF root colonisation, other studies have shown a positive effect. A review paper found that in 51% of the published studies, addition of compost had a positive effect on AMF root colonisation, while only in 8% of the published studies there was a negative effect (Cavagnaro 2015). The rate of compost application might also be important, with higher rates resulting in a decrease in the percent AMF root colonisation of roots (Cavagnaro 2014). Cavagnaro (2014) also showed that even though the percent AMF root colonisation was decreased, increased compost application rates were accompanied with increased rates of root growth, indicating that AMF colonisation per total root biomass might increase.

Beyond the direct impacts on plant nutrition, AMF play important roles in agroecosystems such as increasing resource use efficiency, improving the physical and biological aspects of soil health, and suppressing pathogens. Given the increasing scarcity of P and the negative environmental impacts associated with overuse of phosphorous fertilisers, mycorrhiza can increase agricultural sustainability by maximising P uptake from the soil and fertiliser use efficiency (Gilbert 2009). Even if soils where almonds are grown are not currently limited in P, mycorrhizal associations will make a greater contribution to plant nutrition as the availability of phosphorous fertilisers declines. AMF can further improve resource use efficiency by reducing nutrient leaching, as they increase the extent of nutrient interception zones (Cavagnaro et al. 2015; Bowles et al. 2018). Mycorrhizal fungi are also considered to be a keystone functional group, contributing to the maintenance of the abundant, diverse soil microbial communities that are critical to sustainable agricultural management. Hyphae and glomalin produced by AMF also improve soil aggregate formation and stability, organic matter formation, and water-holding capacity, all of which are important for agroecosystem functioning and plant productivity (Veresoglou et al. 2012). Finally, AMF can suppress soil-borne pathogens and soil nematodes (Hol and Cook 2005; Hu et al. 2010; Krishna et al. 2010; Okada and Matsubara 2012). In soils that have low mycorrhizal abundance or diversity, as may be the case in intensively managed almond orchards, ecology-based management practices that increase AMF root colonisation could help improve resource use efficiency and decrease the environmental footprint of this rapidly expanding industry.

Conclusions

Use of cover crops or resident vegetation and soil P concentrations were the most important factors for high AMF root colonisation in almond orchards. However, further studies are required to identify which combination of agricultural management practices would best support the beneficial association between almonds and AMF in Mediterranean climates. An improved mechanistic understanding of the relationship between mycorrhizal community composition and functions could similarly be used to inform management decisions that differentially affect AMF taxa. This study highlighted the interactive impact of management decisions on AMF root colonisation of almond roots. This provides the agricultural community with preliminary data to guide the adoption of soil health-building management practices and input strategies to optimise synergies between soil microbes and plant functions, especially under acute stress or chronic suboptimal input levels. Soil fertilisation is one of the most important agricultural management practices affecting AMF abundance and more studies are needed in irrigated agricultural land to elucidate the interactive effects of fertigation, water management, and salinity on soil biological potential. Understanding how management decisions impact soil microbial communities and plant-microbe interactions is critical to maintaining biodiverse, resilient agricultural systems in an increasingly challenging production environment.

Acknowledgments

The authors would like to thank UCCE farm advisors, researchers and growers for participating in this study, assist with sampling and for their valuable comments on the manuscript.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This work was supported by a grant from the Almond Board of California (#15-STEWCROP-5) to A.G, BL and A.V and USDA-NIFA, Agricultural Experiment Station Project #CA-D-PLS-2332-H to A.G.Almond Board of California [#15-STEWCROP-5];National Institute of Food and Agriculture [#CA-D-PLS-2332-H];

References

AOAC. 1997. Official method 972.43, microchemical determination of Carbon, Hydrogen, and Nitrogen, automated method 1997. In: Off Methods Anal AOAC Int. 17th ed. Arlington (VA): AOAC International; p. 5–6.

Augé RM. 2001. Water relations, drought and vesicular-arbuscular mycorrhizal symbiosis. Mycorrhiza. 11(1):3–42. doi:10.1007/s005720100097.

Augé RM, Toler HD, Saxton AM. 2015. Arbuscular mycorrhizal symbiosis alters stomatal conductance of host plants more under drought than under amply watered conditions: a meta-analysis. Mycorrhiza. 25(1):13–24. doi:10.1007/s00572-014-0585-4.

Avio L, Castaldini M, Fabiani A, Bedini S, Sbrana C, Turrini A, Giovannetti M. 2013. Impact of nitrogen fertilization and soil tillage on arbuscular mycorrhizal fungal communities in a Mediterranean agroecosystem. Soil Biol Biochem. 67:285–294. doi:10.1016/j.soilbio.2013.09.005.

Bedini S, Avio L, Sbrana C, Turrini A, Migliorini P, Vazzana C, Giovannetti M. 2013. Mycorrhizal activity and diversity in a long-term organic Mediterranean agroecosystem. Biol Fertil Soils. 49(7):781–790. doi:10.1007/s00374-012-0770-6.

Bentley GR. 2010. Environmental pollutants and fertility. In: Bentley Gillian R, Mascie-Taylor CGN, editors. Infertil Mod World. Cambridge: Cambridge University Press; p. 85–152.

Bowles TM, Jackson LE, Cavagnaro TR. 2018. Mycorrhizal fungi enhance plant nutrient acquisition and modulate nitrogen loss with variable water regimes. Glob Chang Biol. 24(1):e171–e182. doi:10.1111/gcb.13884.

Briccoli Bati C, Santilli E, Lombardo L. 2014. Effect of arbuscular mycorrhizal fungi on growth and on micronutrient and macronutrient uptake and allocation in olive plantlets growing under high total Mn levels. Mycorrhiza. 25 (2):97–108. doi:10.1007/s00572-014-0589-0.

Browne G, Ott N, Poret-Peterson A, Gouran H, Lampinen B. 2017. Efficacy of anaerobic soil disinfestation for control of prunus replant disease. Plant Dis. 102(1):209–219. doi:10.1094/PDIS-09-16-1392-RE.

Calvet C, Estaún V, Camprubí A, Hernández-Dorrego A, Pinochet J, Moreno MA. 2004. Aptitude for mycorrhizal root colonization in Prunus rootstocks. Sci Hortic (Amsterdam). 100(1–4):39–49. doi:10.1016/j.scienta.2003.08.001.

Calvet C, Pinochet J, Hernández-Dorrego A, Estaún V, Camprubí A. 2001. Field microplot performance of the peach-almond hybrid GF-677 after inoculation with arbuscular mycorrhizal fungi in a replant soil infested with root-knot nematodes. Mycorrhiza. 10(6):295–300. doi:10.1007/PL00009998.

Cao J, Huang Y, Wang C. 2015. Rhizosphere interactions between earthworms (Eisenia fetida) and arbuscular mycorrhizal fungus (Funneliformis mosseae) promote utilization efficiency of phytate phosphorus in maize. Appl Soil Ecol. 94:30–39. doi:10.1016/j.apsoil.2015.05.001.

Cavagnaro TR. 2014. Impacts of compost application on the formation and functioning of arbuscular mycorrhizas. Soil Biol Biochem. 78:38–44. doi:10.1016/j.soilbio.2014.07.007.

Cavagnaro TR. 2015. Biologically Regulated Nutrient Supply Systems. Compost and Arbuscular Mycorrhizas-A Review. Adv Agron. 129:293–321.

Cavagnaro TR, Bender SF, Asghari HR, van der Heijden MGA. 2015. The role of arbuscular mycorrhizas in reducing soil nutrient loss. Trends Plant Sci. 20(5):283–290. doi:10.1016/j.tplants.2015.03.004.

CDFA. 2018. California agricultural statistics review 2017-2018. Sacramento (CA, USA):California Department of Food and Agriculture.

Correia FV, Moreira JC. 2010. Effects of glyphosate and 2,4-D on earthworms (Eisenia foetida) in laboratory tests. Bull Environ Contam Toxicol. 85(3):264–268. doi:10.1007/s00128-010-0089-7.

de Novais CB, Avio L, Giovannetti M, de Faria SM, Siqueira JO, Sbrana C. 2019. Interconnectedness, length and viability of arbuscular mycorrhizal mycelium as affected by selected herbicides and fungicides. Appl Soil Ecol. 143:144–152. doi:10.1016/j.apsoil.2019.06.013.

Decock C, Garland G, Suddick EC, Six J. 2017. Season and location–specific nitrous oxide emissions in an almond orchard in California. Nutr Cycl Agroecosystems. 107(2):139–155. doi:10.1007/s10705-017-9824-3.

Druille M, Cabello MN, Omacini M, Golluscio RA. 2013a. Glyphosate reduces spore viability and root colonization of arbuscular mycorrhizal fungi. Appl Soil Ecol. 64:99–103. doi:10.1016/j.apsoil.2012.10.007.

Druille M, García-Parisi PA, Golluscio RA, Cavagnaro FP, Omacini M. 2016. Repeated annual glyphosate applications may impair beneficial soil microorganisms in temperate grassland. Agric Ecosyst Environ. 230:184–190. doi:10.1016/j.agee.2016.06.011.

Druille M, Omacini M, Golluscio RA, Cabello MN. 2013b. Arbuscular mycorrhizal fungi are directly and indirectly affected by glyphosate application. Appl Soil Ecol. 72:143–149. doi:10.1016/j.apsoil.2013.06.011.

Evelin H, Kapoor R, Giri B. 2009. Arbuscular mycorrhizal fungi in alleviation of salt stress: a review. Ann Bot. 104 (7):1263–1280. doi:10.1093/aob/mcp251.

Fitter AH, Garbaye J. 1994. Interactions between mycorrhizal fungi and other soil organisms. Plant Soil. 159 (1):123–132. doi:10.1007/BF00000101.

Folorunso OA, Rolston DE, Prichard T, Loui DT. 1992. Soil surface strength and infiltration rate as affected by winter cover crops. Soil Technol. 5(3):189–197. doi:10.1016/0933-3630(92)90021-R.

Fontanet X, Estaún V, Camprubí A, Calvet C. 1998. Fungicides added to potting substrate affect mycorrhizal symbiosis between a peach-almond rootstock and Glomus sp. HortScience. 33(7):1217–1219. doi:10.21273/HORTSCI.33.7.1217.

França SC, Gornes-da-Costa SM, Silveira APD. 2007. Microbial activity and arbuscular mycorrhizal fungal diversity in conventional and organic citrus orchards. Biol Agric Hortic. 25(2):91–102. doi:10.1080/01448765.2007.9755039.

Garbaye J. 1994. Tansley review No.76 Helper bacteria: a new dimension to the mycorrhizal symbiosis. New Phytol. 128(2):197–210. doi:10.1111/j.1469-8137.1994.tb04003.x.

Gaupp-Berghausen M, Hofer M, Rewald B, Zaller JG. 2015. Glyphosate-based herbicides reduce the activity and reproduction of earthworms and lead to increased soil nutrient concentrations. Sci Rep. 5(1):1–9. doi:10.1038/srep12886.

Gilbert N. 2009. Environment: the disappearing nutrient. Nature. 461(7265):716–718. doi:10.1038/461716a.

Giovannetti M, Mosse B, Giovannetti M, Giovanetti M, Mosse B, Giovannetti M. 1980. An evalution of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. New Phytol. 84(3):489–500. doi:10.1111/j.1469-8137.1980.tb04556.x.

Gómez JA, Romero P, Giráldez JV, Fereres E. 2004. Experimental assessment of runoff and soil erosion in an olive grove on a Vertic soil in southern Spain as affected by soil management. Soil Use Manag. 20(4):426–431. doi:10.1079/SUM2004275.

Gómez JA, Sobrinho TA, Giráldez JV, Fereres E. 2009. Soil management effects on runoff, erosion and soil properties in an olive grove of Southern Spain. Soil Tillage Res. 102(1):5–13. doi:10.1016/j.still.2008.05.005.

Gosling P, Hodge A, Goodlass G, Bending GD. 2006. Arbuscular mycorrhizal fungi and organic farming. Agric Ecosyst Environ. 113(1–4):17–35. doi:10.1016/j.agee.2005.09.009.

Gosling P, Ozaki A, Jones J, Turner M, Rayns F, Bending GD. 2010. Organic management of tilled agricultural soils results in a rapid increase in colonisation potential and spore populations of arbuscular mycorrhizal fungi. Agric Ecosyst Environ. 139(1–2):273–279. doi:10.1016/j.agee.2010.08.013.

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. doi:10.1016/j.agee.2017.03.011.

Granger RL, Khanizadeh S, Meheriuk M, Berard LS, Dalpe Y. 1995. Effects of simazine on the mycorrhizal population in soil beneath an apple tree canopy. Fruit Var J. 49:90–93.

Gryndler M, Larsen J, Hršelová H, Řezáčová V, Gryndlerová H, Kubát J. 2006. Organic and mineral fertilization, respectively, increase and decrease the development of external mycelium of arbuscular mycorrhizal fungi in a long-term field experiment. Mycorrhiza. 16(3):159–166. doi:10.1007/s00572-005-0027-4.

Harrier LA, Watson CA. 2004. The potential role of arbuscular mycorrhizal(AM) fungi in the bioprotection of plants against soil-borne pathogens in organic and/or other sustainable farming systems. Pest Manag Sci. 60(2):149–157. doi:10.1002/ps.820.

He X, Chen Y, Liu S, Gunina A, Wang X, Chen W, Shao Y, Shi L, Yao Q, Li J, et al. 2018. Cooperation of earthworm and arbuscular mycorrhizae enhanced plant N uptake by balancing absorption and supply of ammonia. Soil Biol Biochem. 116:351–359. doi:10.1016/j.soilbio.2017.10.038.

Hodge A, Campbell CD, Fitter AH. 2001. An arbuscular mycorrhizal fungus accelerates decomposition and acquires nitrogen directly from organic material. Nature. 413(6853):297–299. doi:10.1038/35095041.

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(31):13754–13759. doi:10.1073/pnas.1005874107.
Hol WHG, Cook R. 2005. An overview of arbuscular mycorrhizal fungi-nematode interactions. Basic Appl Ecol. 6 (6):489–503. doi:10.1016/j.baae.2005.04.001.
Hu JL, Lin XG, Wang JH, Shen WS, Wu S, Peng SP, Mao TT. 2010. Arbuscular mycorrhizal fungal inoculation enhances suppression of cucumber fusarium wilt in greenhouse soils. Pedosphere. 20(5):586–593. doi:10.1016/S1002-0160(10)60048-3.
Jahanzad E, Holtz BA, Zuber CA, Doll D, Brewer KM, Hogan S, Gaudin ACM, Ma Y. 2020. Orchard recycling improves climate change adaptation and mitigation potential of almond production systems. PLoS One. 15 (3):1–22. doi:10.1371/journal.pone.0229588.
Jeffries P, Gianinazzi S, Perotto S, Turnau K, Barea J-M. 2003. The contribution of arbuscular mycorrhizal fungi in sustainable maintenance of plant health and soil fertility. Biol Fertil Soils. 37(1):1–16. doi:10.1007/s00374-002-0546-5.
Johnson NC, Angelard C, Sanders IR, Kiers ET, Klironomos J. 2013. Predicting community and ecosystem outcomes of mycorrhizal responses to global change. Ecol Lett. 16:140–153. doi:10.1111/ele.12085.
Johnson NC, Wilson GWT, Wilson JA, Miller RM, Bowker MA. 2015. Mycorrhizal phenotypes and the Law of the Minimum. New Phytol. 205(4):1473–1484. doi:10.1111/nph.13172.
Jordan ZH. 2000. Arbuscular-mycorrhizal fungi: potential roles in weed management. Weed Res. 40(5):397–410. doi:10.1046/j.1365-3180.2000.00207.x.
Kabir Z. 2005. Tillage or no-tillage: impact on mycorrhizae. Can J Plant Sci. 85(1):23–29. doi:10.4141/P03-160.
Kabir Z, Koide RT. 2002. Effect of autumn and winter mycorrhizal cover crops on soil properties, nutrient uptake and yield of sweet corn in Pennsylvania, USA. Plant Soil. 238(2):205–215. doi:10.1023/A:1014408723664.
Kabir Z, O'halloran IP, Fyles JW, Hamel C. 1997. Seasonal changes of arbuscular mycorrhizal fungi affected by tillage practices and fertilization: hyphal density and mycorrhizal root colonization. Plant Soil. 192(2):285–293. doi:10.1023/A:1004205828485.
Kahimba FC, Sri Ranjan R, Froese J, Entz M, Nason R. 2008. Cover crop effects on infiltration, soil temperature, and soil moisture distribution in the canadian prairies. Appl Eng Agric. 24(3):321–333. doi:10.13031/2013.24502.
Karasawa T, Takebe M. 2012. Temporal or spatial arrangements of cover crops to promote arbuscular mycorrhizal colonization and P uptake of upland crops grown after nonmycorrhizal crops. Plant Soil. 353(1–2):355–366. doi:10.1007/s11104-011-1036-z.
Knepel K. 2003. Determination of Nitrate in 2 M KCl soil extracts by flow injection analysis. QuikChem method 12-107-04-1-B. Loveland (CO): Lachat Instruments.
Krishna H, Das B, Attri BL, Grover M, Ahmed N. 2010. Suppression of Botryosphaeria canker of apple by arbuscular mycorrhizal fungi. Crop Prot. 29(9):1049–1054. doi:10.1016/j.cropro.2010.05.005.
Kroodsma DA, Field CB. 2006. Carbon sequestration in California agriculture, 1980-2000. Ecol Appl. 16 (5):1975–1985. doi:10.1890/1051-0761(2006)016[1975:CSICA]2.0.CO;2.
Lehman RM, Taheri WI, Osborne SL, Buyer JS, Douds DD. 2012. Fall cover cropping can increase arbuscular mycorrhizae in soils supporting intensive agricultural production. Appl Soil Ecol. 61:300–304. doi:10.1016/j.apsoil.2011.11.008.
Liu W, Zhang Y, Jiang S, Murray PJ, Liao L, Li X, Zhang J. 2019. Spatiotemporal differences in the arbuscular mycorrhizal fungi communities in soil and roots in response to long-term organic compost inputs in an intensive agricultural cropping system on the North China Plain. J Soils Sediments. 19(5):2520–2533. doi:10.1007/s11368-019-02244-3.
Lopus SE, Santibáñez MP, Beede RH, Duncan RA, Edstrom J, Niederholzer FJA, Trexler CJ, Brown PH. 2010. Survey examines the adoption of perceived best management practices for almond nutrition. Calif Agric. 64 (3):1689–1699. doi:10.3733/ca.v064n03p149.
Mäder P, Edenhofer S, Boller T, Wiemken A, Niggli U. 2000a. Arbuscular mycorrhizae in a long-term field trial comparing low-input (organic, biological) and high-input (conventional) farming systems in a crop rotation. Biol Fertil Soils. 31(2):150–156. doi:10.1007/s003740050638.
Mäder P, Fliessbach A, Dubois D, Gunst L, Fried P, Niggli U. 2002. Soil fertility and biodiversity in organic farming. Science. 296(5573):1694–1697. doi:10.1126/science.1071148.
Mäder P, Vierheilig H, Boller T, Frey B, Christie P, Wiemken A. 2000b. Transport of 15N from a soil compartment separated by a polytetrafluoroethylene membrane to plant roots via the hyphae of arbuscular mycorrhizal fungi. New Phytol. 146(1):155–161. doi:10.1046/j.1469-8137.2000.00615.x.
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(3):495–501. doi:10.1111/j.1469-8137.1990.tb00476.x.
Moreno B, Garcia-Rodriguez S, Cañizares R, Castro J, Benítez E. 2009. Rainfed olive farming in south-eastern Spain: long-term effect of soil management on biological indicators of soil quality. Agric Ecosyst Environ. 131(3–-4):333–339. doi:10.1016/j.agee.2009.02.011.

Nelson DW, Sommers LE. 1996. Total Carbon, Organic Carbon, and Organic Matter. In: Sparks DL, editor. Methods soil anal part 3 chem methods SSSA B Ser no 5. 3rd ed. Madison (WI.): Soil Science Society of America and American Society of Agronomy; p. 1001–1006.

Oehl F, Sieverding E, Mäder P, Dubois D, Ineichen K, Boller T, Wiemken A. 2004. Impact of long-term conventional and organic farming on the diversity of arbuscular mycorrhizal fungi. Oecologia. 138(4):574–583. doi:10.1007/s00442-003-1458-2.

Okada T, Matsubara Y. 2012. Influence of arbuscular mycorrhizal fungi and sodium chloride on fusarium root rot and antioxidative abilities in asparagus plants. J Japanese Soc Hortic Sci. 81(3):257–262. doi:10.2503/jjshs1.81.257.

Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, Mcglinn D, Minchin PR, O 'Hara RB, Simpson GL, Solymos P, et al 2017. vegan: community ecology package. R Packag version 24-3. cited 2019 Jul 22. Available from: https://cran.r-project.org/web/packages/vegan/index.html

Olsen SR, Sommers LE. 1982. Phosphorus. In: Page AL, Miller RH, Keeney DR, editors. Methods soil anal part 2 chem microbiol prop. 2nd ed. Madison (WI): Am. Soc. Agron; p. 403–430.

R Core Team. 2018. R: a language and environment for statistical computing. R Found Stat Comput Vienna Austria. cited 2019 Jul 22. Available from: http://www.r-project.org/

Roldan-Fajardo BE, Barea JM, Ocampo JA, Azcon-Aguilar C. 1982. The effect of season on VA mycorrhiza of the almond tree and of phosphate fertilization and species of endophyte on its mycorrhizal dependency. Plant Soil. 68 (3):361–367. doi:10.1007/BF02197941.

Ronco MG, Ruscitti MF, Arango MC, Beltrano J. 2008. Glyphosate and mycorrhization induce changes in plant growth and in root morphology and architecture in pepper plants (Capsicum annuum L.). J Hortic Sci Biotechnol. 83(4):497–505. doi:10.1080/14620316.2008.11512413.

Ryan MH, Chilvers GA, Dumaresq DC. 1994. Colonisation of wheat by VA-mycorrhizal fungi was found to be higher on a farm managed in an organic manner than on a conventional neighbour. Plant Soil. 160(1):33–40. doi:10.1007/BF00150343.

Sattelmacher B, Reinhard S, Pomikalko A. 1991. Differences In mycorrhizal colonization of rye (Secale-Cereale L) grown in conventional or organic (biological-dynamic) farming system. J Agron Crop Sci Fur Acker Und Pflanzenbau. 167(5):350–355. doi:10.1111/j.1439-037X.1991.tb00967.x.

Schellenberg DL, Alsina MM, Muhammad S, Stockert CM, Wolff MW, Sanden BL, Brown PH, Smart DR. 2012. Yield-scaled global warming potential from N2O emissions and CH4 oxidation for almond (Prunus dulcis) irrigated with nitrogen fertilizers on arid land. Agric Ecosyst Environ. 155:7–15. doi:10.1016/j.agee.2012.03.008.

Schützendübel A, Nikolova P, Rudolf C, Polle A. 2002. Cadmium and H2O2-induced oxidative stress in Populus x canescens roots. Plant Physiol Biochem. 40(6–8):577–584. doi:10.1016/S0981-9428(02)01411-0.

Sikes BA, Cottenie K, Klironomos JN. 2009. Plant and fungal identity determines pathogen protection of plant roots by arbuscular mycorrhizas. J Ecol. 97(6):1274–1280. doi:10.1111/j.1365-2745.2009.01557.x.

Smith SE, Read DJ. 2008. Mycorrhizal symbiosis. 3rd ed. New York (NY): Academic Press.

Solaiman ZM, Yang H, Archdeacon D, Tippett O, Tibi M, Whiteley AS. 2019. Humus-rich compost increases lettuce growth, nutrient uptake, mycorrhizal colonisation, and soil fertility. Pedosphere. 29(2):170–179. doi:10.1016/S1002-0160(19)60794-0.

Stellin F, Gavinelli F, Stevanato P, Concheri G, Squartini A, Paoletti MG. 2018. Effects of different concentrations of glyphosate (Roundup 360®) on earthworms (Octodrilus complanatus, Lumbricus terrestris and aporrectodea caliginosa) in vineyards in the North-East of Italy. Appl Soil Ecol. 123. doi:110.1016/j.apsoil.2017.07.028.

Sukarno N, Smith FA, Scott ES, Jones GP, Smith SE. 1998. The effect of fungicides on vesicular-arbuscular mycorrhizal symbiosis. III. The influence of VA mycorrhiza on phytotoxic effects following application of fosetyl-Al and phosphonate. New Phytol. 139(2):321–330. doi:10.1046/j.1469-8137.1998.00204.x.

Thirkell TJ, Cameron DD, Hodge A. 2016. Resolving the 'nitrogen paradox' of arbuscular mycorrhizas: fertilization with organic matter brings considerable benefits for plant nutrition and growth. Plant, Cell Environ. 39 (8):1683–1690. doi:10.1111/pce.12667.

Thirkell TJ, Charters MD, Elliott AJ, Sait SM, Field KJ. 2017. Are mycorrhizal fungi our sustainable saviours? Considerations for achieving food security. J Ecol. 105(4):921–929. doi:10.1111/1365-2745.12788.

Thomas GW. 1982. Exchangeable cations. In: Page AL, editor. Methods soil anal part 2 chem microbiol prop agron monogr 92. Madison (WI.): ASA, SSSA; p. 159–165.

Treseder KK. 2013. The extent of mycorrhizal colonization of roots and its influence on plant growth and phosphorus content. Plant Soil. 371(1–2):1–13. doi:10.1007/s11104-013-1681-5.

Treseder KK, Cross A. 2006. Global distributions of arbuscular mycorrhizal fungi. Ecosystems. 9(2):305–316. doi:10.1007/s10021-005-0110-x.

Turrini A, Agnolucci M, Palla M, Tomé E, Tagliavini M, Scandellari F, Giovannetti M. 2017a. Species diversity and community composition of native arbuscular mycorrhizal fungi in apple roots are affected by site and orchard management. Appl Soil Ecol. 116:42–54. doi:10.1016/j.apsoil.2017.03.016.

Turrini A, Caruso G, Avio L, Gennai C, Palla M, Agnolucci M, Tomei PE, Giovannetti M, Gucci R. 2017b. Protective green cover enhances soil respiration and native mycorrhizal potential compared with soil tillage in a high-density olive orchard in a long term study. Appl Soil Ecol. 116:70–78. doi:10.1016/j.apsoil.2017.04.001.

Turrini A, Bedini A, Loor MB, Santini G, Sbrana C, Giovannetti M, Avio L. 2018. Local diversity of native arbuscular mycorrhizal symbionts differentially affects growth and nutrition of three crop plant species. Biol Fertil Soils. 54:203–217. doi:10.1007/s00374-017-1254-5.

Van Geel M, De Beenhouwer M, Ceulemans T, Caes K, Ceustermans A, Bylemans D, Gomand A, Lievens B, Honnay O. 2016. Application of slow-release phosphorus fertilizers increases arbuscular mycorrhizal fungal diversity in the roots of apple trees. Plant Soil. 402(1–2):291–301. doi:10.1007/s11104-015-2777-x.

Verbruggen E, van der Heijden MGA, Rillig MC, Kiers ET. 2013. Mycorrhizal fungal establishment in agricultural soils: factors determining inoculation success. New Phytol. 197(4):1104–1109. doi:10.1111/j.1469-8137.2012.04348.x.

Veresoglou SD, Chen B, Rillig MC. 2012. Arbuscular mycorrhiza and soil nitrogen cycling. Soil Biol Biochem. 46:53–62. doi:10.1016/j.soilbio.2011.11.018.

Veresoglou SD, Rillig MC. 2012. Suppression of fungal and nematode plant pathogens through arbuscular mycorrhizal fungi. Biol Lett. 8(2):214–217. doi:10.1098/rsbl.2011.0874.

Vierheilig H, Coughlan AP, Wyss URS, De RC. 1998. Ink and vinegar, a simple staining technique for arbuscular-mycorrhizal fungi. Appl Environ Microbiol. 64(12):5004–5007. doi:10.1128/AEM.64.12.5004-5007.1998.

Whipps JM. 2004. Prospects and limitations for mycorrhizas in biocontrol of root pathogens. Can J Bot. 82 (8):1198–1227. doi:10.1139/b04-082.

Willis A, Rodrigues BF, Harris PJC. 2013. The ecology of arbuscular mycorrhizal fungi. CRC Crit Rev Plant Sci. 32 (1):1–20. doi:10.1080/07352689.2012.683375.

Wu QS, Li GH, Zou YN. 2011a. Improvement of root system architecture in peach (Prunus persica) seedlings by arbuscular mycorrhizal fungi, related to allocation of glucose/sucrose to root. Not Bot Horti Agrobo Cluj-Napoca. 39(2):232–236. doi:10.15835/nbha3926232.

Wu QS, Li GH, Zou YN. 2011b. Roles of arbuscular mycorrhizal fungi on growth and nutrient acquisition of peach (Prunus persica L. Batsch) seedlings. J Anim Plant Sci. 21:746–750.

Wu QS, Xia RX, Zou YN. 2008. Improved soil structure and citrus growth after inoculation with three arbuscular mycorrhizal fungi under drought stress. Eur J Soil Biol. 44(1):122–128. doi:10.1016/j.ejsobi.2007.10.001.

Zaller JG, Heigl F, Ruess L, Grabmaier A. 2014. Glyphosate herbicide affects belowground interactions between earthworms and symbiotic mycorrhizal fungi in a model ecosystem. Sci Rep. 4:1–8.