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Effect of AMF application on growth, productivity and susceptibility to Verticillium wilt of olives grown under desert conditions

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Abstract The effect of arbuscular mycorrhizal fungi (AMF) on olive (*Olea europaea*) growth and development was followed for 4 years after transplanting in irrigated commercial orchards under arid conditions. Sites I and II were irrigated with saline water (EC=4.5 dS/m). In site I, the soil was infested with *Verticillium dahliae* and olive varieties 'Picual' (*Verticillium* susceptible) and 'Barnea' (relatively *Verticillium* tolerant) were tested. In site II, the soil was virgin soil (previously non-cultivated soil) and olive varieties 'Souri' and 'Barnea' were tested. Plants for all sites were inoculated in the nursery with *Glomus intraradices* alone or in a mixture with *G. mosseae*. Relative to non-inoculated trees, AMF colonization enhanced vegetative growth, expressed as tree height and trunk circumference, at all sites. At first commercial harvest, AMF-treated trees had higher fruit and oil yields than non-mycorrhitic controls. Under saline water irrigation, differences between inoculated and non-inoculated treatments were reduced in the slow-growing 'Souri' but remained apparent in the modern fast-growing 'Barnea'. AMF colonization did not appear to improve tolerance of either 'Picual' or 'Barnea' to *V. dahliae*, and both were more susceptible than the non-inoculated controls. Thus inoculation of olive plants with AMF improves transplant growth and adaptation in arid areas during the first 3 years of growth and until the first commercial harvesting season.

Keywords Arbuscular mycorrhizal symbiosis · Olives · Verticillium wilt

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1 Introduction

Arbuscular mycorrhizal (AM) symbiosis plays a major role in ecosystems, facilitating nutrient cycling by providing plants with essential nutrients. The AM fungi (AMF) are members of the fungal phylum *Glomeromycota* (Schüssler et al. 2001) and form symbiotic associations with most terrestrial vascular flowering plants (Smith and Read 1997). In addition to increasing nutrient uptake, AMF improve plant rooting and establishment, enhance vegetative growth, and accelerate budding and flowering (Smith and Read 1997). Moreover, the plant-AMF symbiosis has been shown to promote the plant's ability to withstand numerous biotic and abiotic stresses (reviewed by Purin and Rillig 2008; Koltai and Kapulnik 2010); AMF promote plant resistance to stress, and enhance productivity and sustainability of crops, especially under prolonged stress conditions.

Olive (*Olea europaea*) trees have been reported to be a responsive host to AMF (Citernesi et al. 1998; Marìn-Zamora et al. 2002; Calvente et al. 2004; Porras-Piedra et al. 2005). Mycorrhizal inoculation of propagated olive plantlets has been shown to have a positive influence on plantlet development in the nursery, resulting in greater mineral uptake and a significantly shorter period of nursery-stage plantlet development (Martin et al. 2006; Soriano-

Martín et al. 2006; Dag et al. 2009; Porras-Soriano et al. 2010). Moreover, AMF colonization enhanced the acclimatization of young trees transplanted to the field (Estaún et al. 2003). These contributions to plant development are especially important for the establishment of olive orchards in arid regions.

Olive trees exhibit great adaptability to adverse soil conditions and are traditionally grown under rain-feeding. Nevertheless, in Israel, like many Mediterranean countries, most of the newly planted olive orchards are grown under irrigation, which allows extending the cultivation area to desert regions characterized by high temperatures, marginal soils with low fertility and deficit irrigation, all of which affect plant survival and productivity. Moreover, in many of these areas, farmers are using saline water as an alternative water source for cultivation (Dag et al. 2008). Thus, AMF symbiosis can serve as a simple biological solution to support plant development under such conditions in olive orchards. The effects of AM colonization on olive tree growth under saline conditions have been recently reported (Porras-Soriano et al. 2009). AMF-colonized olive trees in a controlled environment were more tolerant to the salinity, as expressed by significantly higher growth and nutrient uptake relative to non-inoculated trees. This general effect on plant nutrition was suggested to also be a key element, among others, in the resistance of mycorrhitic plants to pathogens (Davis et al. 1979). However, Porras-Soriano et al. (2006) demonstrated that under nursery conditions, although AMF application results in better olive plantlet growth performance, it does not improve resistance of olives plants to attack by *Verticillium dahliae*. Moreover, not much information is available on the effect of AMF colonization on olive productivity in the field and the ability of mycorrhitic olive trees to tolerate soilborne pathogens.

Verticillium wilt (VW) on olive, caused by *V. dahliae*, is widely distributed in all olive-growing regions of the world, and is one of the major limiting factors in olive oil production (Hartmann et al. 1971; Blanco-López et al. 1984; Levin et al. 2003a, b; Tsror (Lahkim) 2010). Economic damage caused by the disease has increased over the last 20 years because of changing conditions for olive growing, especially irrigation (use of fresh or saline water), high-density planting and intercropping with *V. dahliae*-susceptible hosts such as potato or cotton (Tjamos 1993; Blanco-López and Jiménes-Días 1995; Levin et al. 2003a, b, 2007; Tsror (Lahkim) 2010). The disease can be spread by use of latently *V. dahliae*-infected seedlings, by contaminated water, and by bad sanitation (Tsror (Lahkim) 2010). Since control of this disease is both difficult to achieve and expensive (Tjamos et al. 1991), alternative practical treatments are being explored.

In the present study, we examined the effects of AMF inoculation on olive growth, development and fruit and oil yields, after transplanting to orchards irrigated with saline water. Furthermore, this study evaluated the effect of AMF inoculation on development of VW in trees grown in naturally *V. dahlia*—infected soil during the first fruiting season (i.e., 4 years after transplanting to the field).

2 Materials and methods

2.1 General conditions and inoculation

The AMF used, *G. intraradices* [Schenck & Smith] and *G. mosseae*, were prepared and applied as previously described by Dag et al. (2009). The inoculum consisted of spores, hyphae and infected roots in vermiculite as a carrier, at a rate of 150 infection units/ml (a local isolate produced by Agrogold Ltd, Moshav Shadmot Dvora Israel). The inoculum was applied to the growth substrate at 10% (v/v), placed at the bottom of the pot up to one third of its volume so that the transplanted rooted cutting was in direct contact with the inoculum. The rest of the pot's volume (two third) was filled with growth substrate that did not contained AMF inoculum. After transplanting into pots, the seedlings were fertilized and irrigated in the nursery according to normal commercial practices as described previously (Dag et al. 2009). In all cases except the 'non-inoculated+fertilizer' treatment, no fertilizers were applied during the first month after transplanting, to ensure colonization of the AMF. According to a previous study (Dag et al. 2006), under the above-described inoculation conditions and treatments, 6 months following inoculation, control plants are not colonized whereas AM-treated plants are well-colonized with mycorrhizal fungi. Subsequent to inoculation fertilizers including N and K (40 mg/L each), P (12 mg/L) and micronutrients were applied routinely via the irrigation system. After another period of 6–12 months in the nursery, the plants were planted in the field.

2.2 Experiment I (site I: Revivim)

To evaluate AMF's potential contribution to olive tree resistance to *V. dahliae*, susceptible and tolerant cultivars, the Spanish 'Picual' and the local Israeli cv. Barnea (Lavee et al. 1986; Levin et al. 2003a, b), respectively, were tested. The experiment was carried out in a field previously cropped with potatoes, where symptoms of *V. dahliae* had appeared in an adjacent olive orchard that was established earlier on the site. The site is located in an arid Mediterranean zone with average winter rainfall of 100 mm and summers characterized by high temperatures, low relative humidity and high radiation levels (Dag et al. 2008), and the soil is sandy loam. Irrigation water (saline water, EC=4.5–5.0 dS/m) and

fertilizers were applied via an automated drip-irrigation system (Netafim, Ltd. Israel).

Seedlings were inoculated in the nursery with either *G. intraradices* alone (Gi) or a mixture of *G. intraradices* and *G. mosseae* (Gi+Gm) (at a rate of 1:1 infection units prior to application) and were compared to a non-inoculated control. On 11 May 2006, 1 year after AMF inoculation, trees were planted at 3×7 m spacing. Each cultivar in each treatment in each plot (replicate) was evaluated with four adjacent trees. Trees were assigned to five blocks (row), where trees belonging to the same treatment were planted adjacent to each other. This planting design was chosen to reduce cross-contamination of the non-inoculated control trees with AMF from neighboring pre-inoculated trees. Growth rates in this experiment and others throughout this study were determined by measuring tree height and trunk circumference 9 and 7 times during the experiment, respectively. Plants were grown under the conditions and agronomical practices of commercially irrigated olives in the area. Upon reaching the appropriate level of ripeness, all fruit from individual trees were harvested and weighed. A sample of 100 g was brought to the lab to determine oil content by chemical extraction.

2.3 Experiment II (site: Be'er Hail)

One year after AMF inoculation, on 11 May 2006, the olive seedlings were transplanted to a virgin field, i.e. one which had not been previously cultivated. This site is located also in an arid Mediterranean zone with an average winter rainfall of 100 mm and summers characterized by high temperatures, low humidity and high radiation levels (Dag et al. 2008). The soil at this site is local loess, with low organic matter and nutrient contents and lacking structure (Dag et al. 2008). Prior to planting, the soil was irrigated with large amounts of water to leach salts. The trees were grown under the conditions and practices recommended for commercial irrigated olive cultivation in the region. Trees were planted at 3×7 m spacing in four blocks (rows) with five plants per replicate. Irrigation and fertilization were applied via an automated drip system. Irrigated water had high EC values, ranging from 4.5 to 5.0 dS/m. The tested cultivars—the traditional 'Souri' and the relatively new locally selected cv. Barnea—were chosen to represent the cultivars used in approximately 50% of the recently newly planted olive orchards in Israel. Seedlings were inoculated in the nursery with either Gi or Gi+Gm and were compared to a non-inoculated control that was fertilized after transplanting (non-inoc.+F) and to a non-inoculated control that was not fertilized in the month after transplanting (non-inoc.-F). Growth rates in this experiment were determined by measuring tree height and trunk circumference 9 and 8 times during the experiment, respectively.

2.4 Verticillium wilt evaluation in the field

Disease evaluation was carried out by visual observation. Disease incidence represents the number of trees with symptoms out of the total number of trees, for each plot. Symptoms were evaluated twice a year during the study on a scale of 0–5: 0 = healthy tree; 1 = up to 25% of the tree with symptoms, including chlorotic leaves and dead stems; 2 = up to 50% of the tree affected, including symptoms on lateral branches; 3 = up to 75% of the tree affected, including most branches; 4 = up 95% of the tree affected; 5 = dead tree. Diseased branches were removed in summer and winter. After each pruning, the trees were classified as symptomless.

Severity index (SI, on a scale of 2–10) takes into consideration the number of diseased trees at each symptom level and was calculated as follows: SI = [(number of trees in level 1 × 2) + (number of trees in level 2 × 4) + (number of trees in level 3 × 6) + (number of trees in level 4 × 8)) + (number of trees in level 5 × 10)]/ total diseased trees (modified from Tsror (Lahkim) et al. 1998; Levin et al. 2003a, b).

2.5 Isolation of *V. dahliae* from trees

Samples of 20 diseased or symptomless stems, 5–8 mm thick, from each of the trees from each plot were collected during the study. Four 4-cm long segments from each stem were surface-sterilized with 0.3% HClO for 7 min and rinsed with sterile water. Three pieces (2–5 mm long) from each were transferred to sorbose agar medium (SA) (0.2% w/v sorbose, 1.5% w/v agar, 100 ppm streptomycin), incubated at 25°C in the dark, and examined after 2 weeks for the presence of *V. dahliae.*

2.6 Statistical analysis

Data were analyzed using JMP 5.0 software (SAS Institute Inc., Cary, NC). Effects of the treatments on growth parameters were examined by one-way ANOVA. Differences between treatments of the same cultivar were determined using Tukey-Kramer honestly significant difference test (at $P \leq 0.05$).

3 Results

3.1 Effect of AMF inoculation on olive tree growth in the field

The effect of AMF on plant development during the first 4 years of growth was followed in two sites, Revivim (site I) and Be'er Hail (site II). In site I, the general vegetative

growth performance of 'Barnea' was higher than that of 'Picual', although both varieties responded positively to the AMF inoculation by showing enhanced tree growth relative to the non-inoculated control. Further, there were no significant differences between plants inoculated with a single strain of AMF and plants inoculated with the mixture of two strains (Gi and Gi+Gm, respectively). Differences in plant height between inoculated and non-inoculated plants were similar for 'Picual' and 'Barnea' for the first 945 days after transplanting (Fig. 1a, b). At this stage, average plant height in 'Picual' for Gi− and Gi+Gm-inoculated trees was 274 cm and 292 cm, respectively, whereas in the non-inoculated controls, plant height was 210 cm. Average plant height in 'Barnea' for Gi− and Gi+Gm-inoculated trees was 372 cm and 387 cm, respectively, vs. 329 cm in the non-inoculated controls. Nevertheless, at the end of the fourth year, 1,383 days after transplanting, values between treatments did not differ significantly in 'Barnea', mainly due to the fact that this cultivar was trimmed to meet optimal canopy height. In 'Picual' on the other hand, these differences prevailed, albeit to a lesser extent.

Similar, but more pronounced trends were observed for trunk circumference (Fig. 1c, d). At the end of the first 920 days of growth, average circumference for the Gi− and Gi+Gm-treated trees was 241 mm and 245 mm, respectively, for 'Picual', and 279 mm and 279 mm, respectively, for 'Barnea', both significantly larger ($P<0.05$) than the circumference of the non-inoculated trees of both varieties (145 mm for 'Picual' and 230 mm for 'Barnea'). At the end of the fourth year, in both varieties, trunk circumference of AMF-treated trees was still larger than that of non-inoculated trees, but values between treatments no longer differed significantly (Fig. 1c, d). It can be concluded that until about 2.5 years after transplanting, AMF treatments have a strong effect on plant growth. Effects then gradually decline, although 4 years after transplanting, AMF-inoculated plants were still larger than the non-inoculated ones.

In site II, both cultivars exhibited enhanced vegetative growth in terms of tree height and trunk circumference due to AMF inoculation, throughout the experiment (Fig. 2a, b). In general, 'Barnea' grows more vigorously than 'Souri',

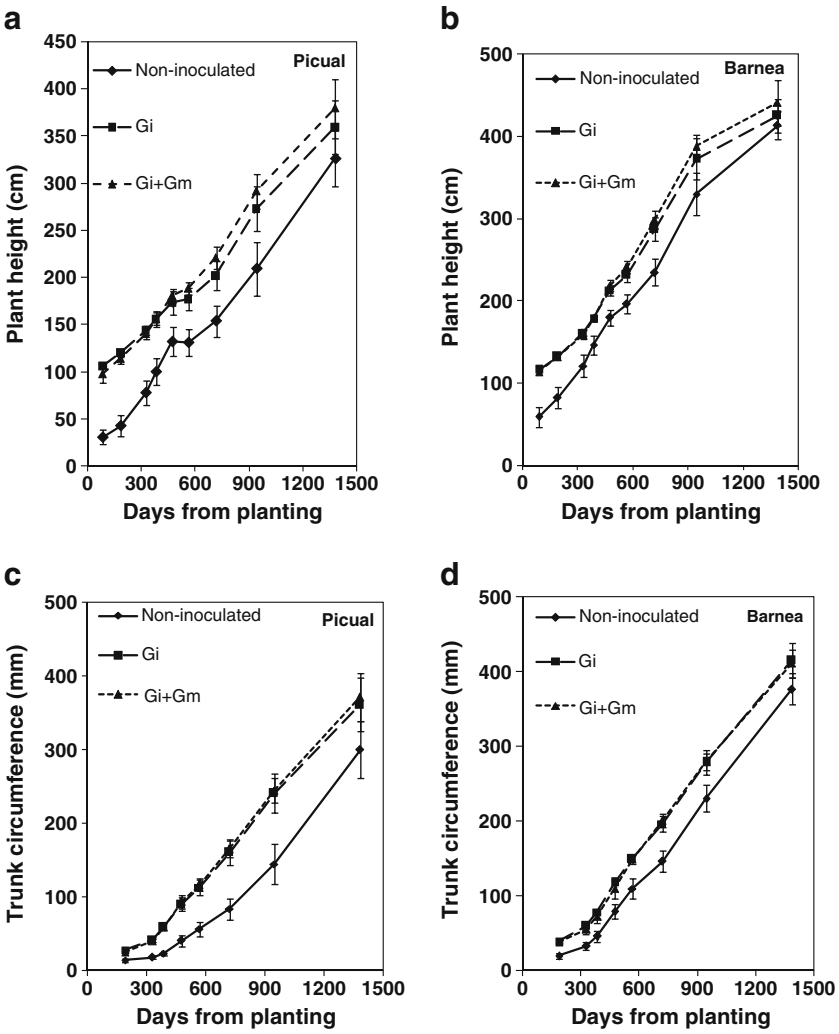

Fig. 1 Effect of mycorrhizal inoculation with *G. intraradices* (Gi) or a mixture of *G. intraradices* and *G. mosseae* (Gi+Gm) on **a** Picual and **b** Barnea growth (height) and **c** Picual and **d** Barnea growth (trunk circumference), Exp. I (Site I), Revivim

which is a slow-growing variety (Dag et al. 2009); accordingly, plant height of the non-inoculated, non-fertilized trees were 299 cm and 118 cm, respectively, 945 days after transplanting in the field (Fig. 2a, b). For cv. Souri, differences in accumulated height between plants inoculated with a single strain of AMF (Gi) and those inoculated with the mixed strains (Gi+Gm), relative to controls, were smaller than for 'Barnea' (Fig. 2a). Moreover, 'Souri' trees gained more from the AMF symbiosis when inoculated with a mixture of strains (Gi+Gm) than with the individual strain (Gi) (Fig. 2a), while this differential effect was not observed with 'Barnea' under the same growing conditions (Fig. 2b). Notably, 'Barnea' trees, and especially the AMF-treated trees, reached the recommended canopy height sooner and had to be trimmed back to the optimal size of the orchard (Fig. 2b). Trunk circumference was also altered by application of AMF throughout the entire growth period relative to the non-inoculated controls (Fig. 2c, d). Nevertheless, while 'Barnea' responded positively to the Gi-only inoculant, 'Souri' responded positively to the inocula containing both AM fungi (Gi+Gm; Fig. 2c, d), relative to their respective non-inoculated control treatments. These results suggest that the ability to respond noticeably to AMF inoculation depends not only on the olive genotype but also on the identity of the inoculated AM fungi.

In the non-inoculated controls, absence of fertilization for 1 month after transplanting the rooted cuttings into pots significantly reduced tree growth in 'Barnea' (Fig. 2b, d) relative to its fertilized counterparts (representing common commercial practice in olive nurseries). A similar, albeit less pronounced trend was observed for 'Souri' (Fig. 2a, b).

3.2 Effect of AMF inoculation on olive tree yield

Fruit and oil yields were evaluated at the first commercial harvest (i.e., during the fourth growing season from transplanting) in sites I. Relative to the non-inoculated trees, inoculation of 'Barnea' with AMF enhanced fruit yield by 2.8- and 6-fold in the Gi− and Gi+Gm-inoculated plants, respectively, compared to the non-inoculated control. How-

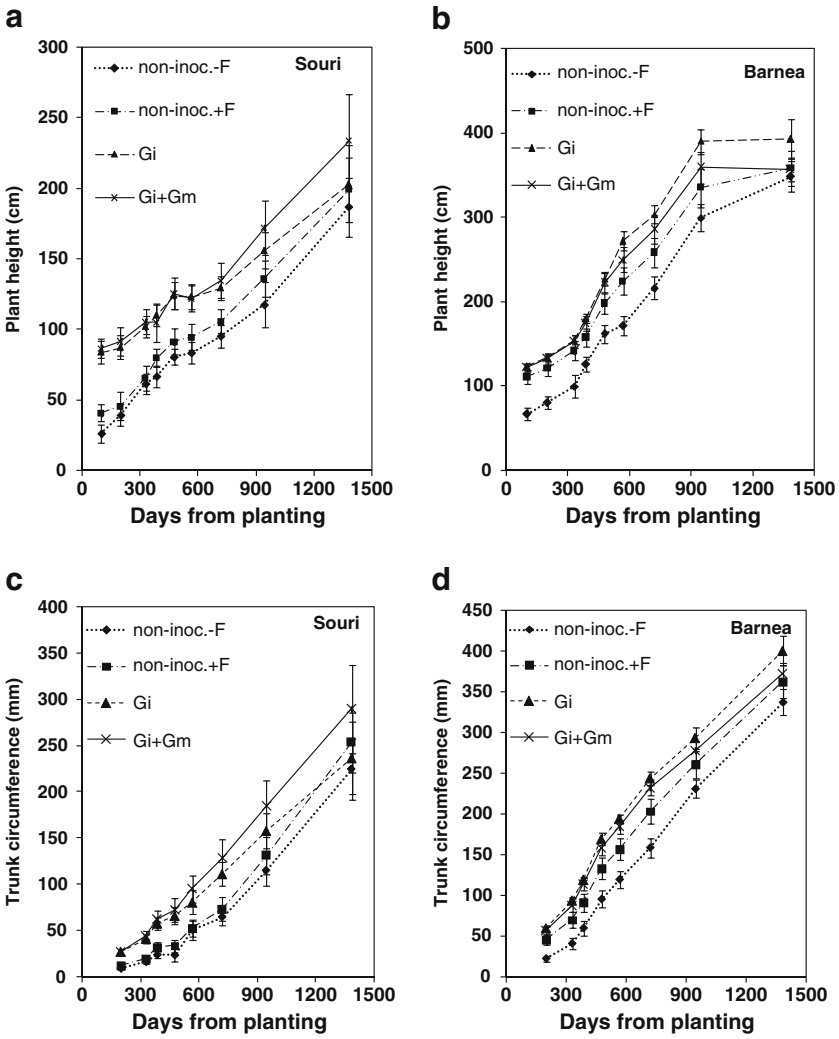

Fig. 2 Effect of mycorrhizal inoculation with *G. intraradices* (Gi) or a mixture of *G. intraradices* and *G. mosseae* (Gi+Gm) in comparison to non-inoculated controls with fertilization in the month after transplanting (non-inoc.+F) or without (non-inoc.-F) on **a** Souri and **b** Barnea growth (height) and **c** Souri and **d** Barnea growth (trunk circumference), Exp. II (Site II), Be'er Hail

ever, only results for the Gi+Gm inoculation treatment were statistically different from the non-inoculated treatment (Table 1). Similar trends were observed for the yield weight of 'Picual' at the same site (Table 1). Accordingly, oil yields in site I followed the same trends as fruit yield (Table 1) where Gi+Gm-inoculated plants, of both cultivars, gave higher oil production significantly relative to the non-inoculated respected controls .

3.3 Effect of AMF inoculation on Verticillum wilt development on olive trees

The first symptoms of Verticillium wilt on both cvs. 'Barnea' and 'Picual' were observed 20 months after planting. Disease incidence had increased, up to 53 and 40% in 'Barnea' and 'Picual', respectively (Fig. 3a, c). In both cultivars, disease incidence in the mycorrhiza-treated plants was higher (but not statistically different) than in the non-treated ones, especially in the susceptible cv. Picual (Fig. 3a, c). Nevertheless, at the end of the fourth year, the differences in disease incidence among treatments were smaller. In addition, no differences between trees inoculated with a single strain of AMF and trees inoculated with the mixture of two strains (Gi and Gi+Gm, respectively) were found.

During the first 2 years, disease indices values (which consider the number of symptomatic trees and the severity levels of the symptoms) were higher in 'Barnea' than in 'Picual' (Table 2), but after pruning (815 days after planting), disease incidences were either similar in both cultivars or higher in 'Picual'. In general, in both cultivars, disease incidences in the controls with no mychorrizae inoculum, were lower than in treatments inoculated with the combination of Gi+Gm, or Gi alone. In 'Picual', 970 days after planting, disease index was significantly higher in the plants treated with the combination of Gi+Gm, compared with the non-treated ones (Table 2).

Fungal colonization was detected in all treatments, in both cultivars (Fig. 3b, d). Colonization levels were very low during the first 2 years, but increased 815 days after planting. They were lowest in non-inoculated 'Picual' (Fig. 3b). Not surprisingly, colonization in 'Barnea', 4 years after planting, was lower than that in 'Picual,' regardless of the treatment.

4 Discussion

Vegetative growth parameters, represented by tree height and trunk circumference, were enhanced by AMF application in commercial orchards yet, differences were not always statistically significant. AMF also significantly increased fruit and oil productivity at first harvest. In most cases, differences between inoculated and non-inoculated plants appeared to be consistent throughout the period starting with transplanting of the young trees in the orchard and ending with the last measurement, taken some 1,400 days later. These AMF-induced differences in the field can be attributed to the different growth rates at the nursery stage. Nevertheless, we expected that all tree roots, including controls, would eventually be colonized with naturally occurring AMF populations, thus diminishing the nursery advantage with time. However, the benefit of AMF introduction at the early plant stages prevailed for a long time (in most cases at list one of the inoculation treatments for about 4 years after transplanting), and in one example, was further expressed by higher commercial fruit yield and, consequently, oil yield, as compared to the non-inoculated control plants (Table 1). These long-term influences of AMF application were greater than those found previously under more moderate conditions (Estaún et al. 2003), probably due to the desert conditions: soils contain low natural AMF populations (Dag et al. 2009), which probably cannot outcompete the inoculated/introduced AMF strains.

When the locally isolated *G. intaradices* strain was the only colonizing fungus in the inocula, it significantly enhanced the growth response of the locally bred olive cv. Barnea, but did not necessarily enhance growth in the other olive cultivars. 'Barnea' is characterized by its high growth rate and yield productivity relative to other cultivars (Dag et al. 2008 and also Figs. 1 and 2 of the present work). In fact, in terms of root colonization under salt stress conditions, *G. intaradices* is superior to *G. mosseae* on olive roots (Porras-

Table 1 Effect of AMF application on olive yield in the first harvesting season, i.e. the fourth year of growth after transplanting

Location	Cultivar	Treatment	First harvest	
			Fruit yield (kg/tree)	Oil yield (kg/tree)
Site I	Barnea	Control	1.90a	0.39a
		Gi	5.50ab	0.85ab
		Gi+Gm	11.39b	2.14b
	Picual	Control	0.02a	0a
		Gi	4.08ab	0.59ab
		Gi+Gm	8.01b	0.93b

Values are means of four replicates, each consisting of three trees for each species. Values followed by the same letter within cultivars do not differ significantly at $P>0.05$

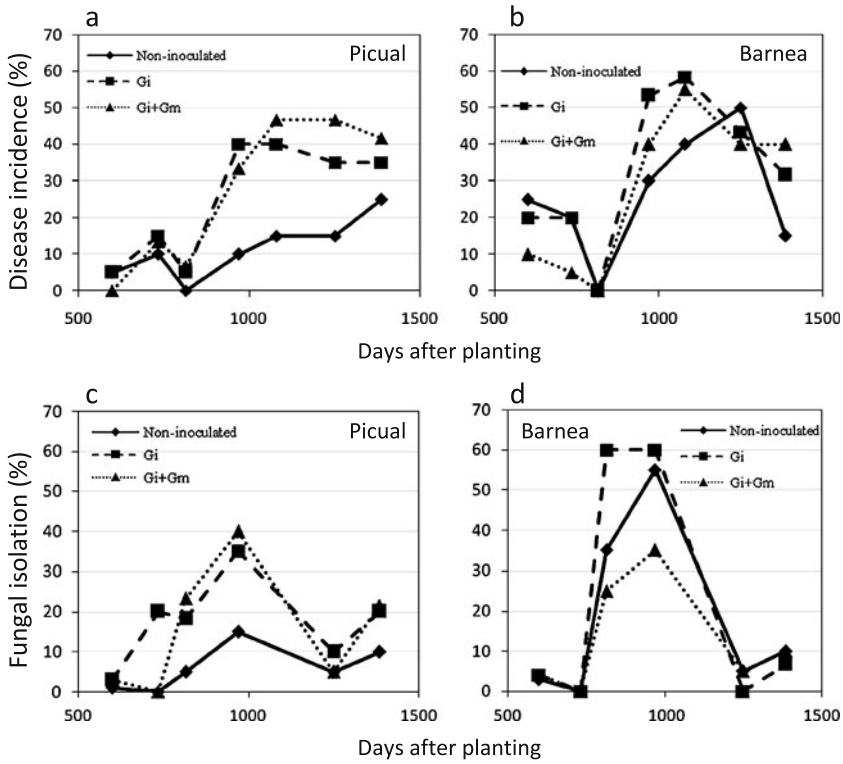

Fig. 3 Effect of mycorrhizal inoculation with *G. intraradices* (Gi) or a mixture of *G. intraradices* and *G. mosseae* (Gi+Gm) on incidence of *V. dahliae* in **a** Picual and **b** Barnea, and fungal isolation from **c** Picual and **d** Barnea, Exp. I, Revivim. Disease parameters were subjected to one-way analysis of variance (ANOVA), and separated by Student multiple range test (P=0.05). No statistical differences between the treatments were found

Soriano et al. 2009) and to (*G. mosseae*) co-inoculated at a 1:1 ratio with *G. intraradices* on *Medicago sativa* roots (Alkan et al. 2006). Accordingly, 'Souri' trees, which develop more slowly, preferentially grow better when inoculated with the inoculant that contains the lower proportion of this local *G. intraradices* strain (Fig. 2c), indicating that use of AMF that are native to the target agrosystem does not necessarily imply their high effectiveness as inoculants on other varieties, especially under abiotic stresses, such as salinity. These results indicate (i) the specific compatibility relationships that exist among symbionts, where AM symbiotic efficiency attributed to olive plants is dependent on plant cultivar and AMF species and (ii) that AMF-host relationships (e.g. fungal effectiveness) might be suitable under particular environmental conditions, but not under others. These conclusions are also supported by earlier observations (Caravaca et al. 2003a, b), and emphasize the importance of appropriate selection processes aimed at choosing the most effective AMF inoculants for the target plant species or even variety, as advised by Enkhtuya et al. (2000). However, since in each of the sites we have tested different tree cultivars, conclusions about AMF-host compatibility may need further validation.

Application of AMF to olive trees enhanced the probability of both susceptible and tolerant cultivars being infected by *V. dahliae* which was already present in the soil, due to previous potato cropping. Several studies have shown that AMF colonization in host roots alters disease severity in the host. While most studies have shown a beneficial effect of AMF (Dehne and Schonbeck 1975; Schönbeck and Dehne 1977; Stewart and Pfleger 1977; Davis and Menge 1981; Graham and Menge 1982; Dugassa et al. 1996), some have shown an increase in pathogenic attack (Davis et al. 1979; Baath and Hayman 1983). The interaction of AMF with *V. dahliae* in olive trees under field

Table 2 Effect of AMF application on Verticillium wilt severity index in olive

Days after planting		600	734	815	970	1,080	1,250	1,386
Cultivar	Treatment							
Picual	Control	0.4 a	0.0 a	0.0 a	0.7 b	0.9 a	0.5 a	1.5 a
Picual	Gi	0.4 a	0.6 a	0.2 a	2.3 ab	2.5 a	0.7 a	0.6 a
Picual	Gi+Gm	0.0 a	0.5 a	0.1 a	3.0 a	2.4 a	1.4 a	3.5 a
Barnea	Control	1.5 A	1.7A	0.0	1.1 A	1.4 A	0.9 A	0.6 A
Barnea	Gi	1.6 A	1.7A	0.0	2.1 A	2.0 A	0.7 A	0.8 A
Barnea	Gi+Gm	0.6 A	0.4 A	0.0	1.9 A	2.3 A	1.0 A	1.3 A

Disease parameters were subjected to one-way analysis of variance (ANOVA), and separated by Student multiple range test (P=0.05). Different letters in rows indicate significant differences among treatments, separately for each cultivar

conditions has not been much studied. However, under nursery growth conditions, Porras-Soriano et al. (2006) demonstrated that inoculation of olive (cv. Cornicabra) plantlets with different AMF (i.e. *G. intraradices, G. mosseae* or *G. claroideum*) did not appear to improve tolerance to Verticillium wilt in the nursery when the plantlets were artificially inoculated with a conidial suspension of *V. dahliae*. In the present work, we took this one step further by planting susceptible and tolerant olive cultivars that were pre-inoculated with AM inoculants in a naturally *V. dahliae*-infested soil. Symptoms on both cultivars spread slowly in the adult trees which eventually, upon reaching the first harvest season 4 years after transplanting, collapsed. In general, the mycorrhitic trees were larger than the non-inoculated controls, and the increased rate of disease severity relative to the non-inoculated trees might be attributed to one or more of the following: (i) a larger population of *V. dahliae* in mycorrhizal plants due to their improved nutritional status, (ii) an increase in the number of potential infection sites for the pathogen, due to the larger penetration sites on the root surface ruptured by AMF colonization, or (iii) enhanced upward movement of *V. dahliae* conidia in the xylem vessels of mycorrhitic plants, where the greater amount of tissue (root and shoot) results in greater transpiration.

In conclusion, AMF inoculation is a powerful tool for enhancing the growth rate of olive seedlings. The AMF-conferred advantage, mainly observed at the nursery stage, was sustained in the field after planting. Under arid Mediterranean conditions, where soils are naturally poorly populated with AMF, pre-inoculation with AMF appears to be even more beneficial than for more typical, less severe Mediterranean conditions. However, in *V. dahliae*-infested soil, the risk for disease development might be enhanced, especially in susceptible cultivars.

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