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Integration of Arbuscular Mycorrhizal Fungi to Grape Vine (*Vitis vinifera* L.) in Nursery Stage

M. Eftekhari[1], M. Alizadeh[1]*, K. Mashayekhi[1], H. Asghari[2], B. Kamkar[1]

[1*]*Department of Horticulture, Pardis Faculty of Agriculture, Gorgan University of Agricultural Sciences and Natural Resources (GUASNR), P.O. Box 386, Golestan, Gorgan, I.R. Iran.*
[2]*Shahrood University of Technology, Semnan, Shahroud, I.R., Iran.*

Abstract: The Arbuscular Mycorrhizal (AM) association is being considered as the commonest Mycorrhizal type involved in grape community. Low population density of these useful fungi in vineyard soil suggests the need for manual inoculation of grapevine plantlets at the nursery stage. The influence of three commercial Arbuscular Mycorrhizal fungi strains (*Glomus intraradious, G. mosseae, G. fasciculatus* and a mixture of them) on growth and biochemical status of four grapevine varieties (Shahroodi, Asgari, Keshmeshi and Khalili) was investigated under greenhouse conditions. Rooted plantlets derived from hardwood cuttings were transplanted in pots containing leaf mold and sand (1:1) followed by inoculation with different fungal inoculums. Various physiological and biochemical parameters were measured at 30 days intervals. The percentage of root colonization was found to be slightly different amongst inoculated vines but it was found to be significantly different with non-inoculated, control plants. Most growth related parameters (vine length, shoot length and leaf area) were enhanced following Mycorrhization but root length and number of leaves were not significantly affected by any fungal intervention. Treated plants typically showed more obvious modifications in their biochemical status. The chlorophyll content (especially "b" and total), total root and shoot phenols were raised in treated plants. The chlorophyll "a" and total soluble sugars were not statistically different in inoculated and control plants. The overall results of the present study suggest that AM fungi can be manually applied, as an easy and economical approach during nursery production, to boost the physiological and biochemical status of the treated plants and production of high quality healthy plantlets.

Keywords: Arbuscular mycorrhizal fungi, *Vitis vinifera* L., Growth, Biochemical analysis.

1. Introduction

The roots of most plant species show symbiosis with a kind of soil microorganisms. Approximately 70% of all plant families contain species that develop specialized Endomycorrhizae called vesicular-arbuscular mycorrhizae (VAM) or just arbuscular mycorrhizae (AM) on their roots (1). This kind of symbiosis has been known to increase plant growth in a very wide variety of plant species including several crops and trees (2). The effects of AM fungi on the growth and development of horticultural plants have been well documented (3, 4 & 5). It has been known for over a century that grapevines (*Vitis* spp.) form symbiotic associations in their roots with such micro-organisms (6). Mycorrhizal colonization of grafted grapevines was studied during early establishment of an experimental rootstock vineyard to determine rootstock variability forming a functional association (7). There are also some reports on the role of AM fungi as an aid to hardening in micropropagated grape plantlets to reduce transplantation shock and alleviation of stresses in weaning stage, the process which is commonly known as bio-hardening (5 & 8). According to Aguin *et al*., (9), population of AM fungi in the field may be low or rare (in fumigated soils), suggesting the need for AM inoculation of grapevine plants at the nursery stage. Hence, addition of AM fungi inoculums to rooting

*Corresponding author:
E-mail: alizadehpub@gmail.com; Tel: +98-1714423303.

substrate could be an effective strategy for the nursery production of Mycorrhizal plants. Differential growth of Mycorrhizal field-inoculated grapevine root-stocks in replant soils have been also recently studied (10). Owing to extension activities held by private and governmental institutes, integration of AM fungi to horticulture and particularly vineyard management is recently getting popular in this area. Hence, the present investigation was designed to examine the influence of three AM fungal species on growth and other morpho-physiological parameters of grape hardwood cuttings during nursery production.

2. Materials and Methods

2.1 Plant materials

Hard-wood stem cuttings of four table grape (*Vitis vinifera* L.) varieties namely, Shahroodi, Asgari, Keshmeshi and Khalili were collected from a well maintained vineyard at the Shahrood Agricultural Research Center, Semnan Province (latitude 35º 34´ N, longitude 53º 23´ E, altitude 1130m), by mid-March. The cuttings were further dissected and pruned to at least four buds (about 30cm long) and the same were inserted in a pre-soaked sawdust medium to induce rooting without any hormonal treatment. These were raised in a glass-house under normal day length (12 hours) and an average temperature of 25ºC. Root emergence was observed in all varieties within three weeks following insertion.

2.2 Inoculum preparation and application

Three AM fungi species namely, *Glomus mosseae, G. fasciculatum* and *G. intraradices* were used. Mycorrhizal inocula were procured from a commercial laboratory (Turan Biotech Co., Shahroud Iran). These consisted of soil, spores (spore density of 150/100 g dry soil), mycelium, and infected/colonized host root fragments. The rooted grape cuttings were transplanted in plastic pots (three per pot) containing natural decomposed forest leaf mold mixed with fine sand (1:1 v/v). For mycorrhizal inoculation, each pot was inoculated with 100 g soil based inoculums (1:50) from above mentioned strains, just distributed beneath the rhizosphere (root zone area) to facilitate root colonization. An additional treatment also was used as mixed species (combination of all three strains). The transplanted plants were irrigated about 80% of field capacity and kept under glass-house conditions for further growth and evaluation. The non-inoculated pots were filled with the same potting mixture (without inoculum) and were used as control.

2.3 Assessment of root colonization

Root colonization percentages were measured 60 days after inoculation (DAI) through the modified method proposed by Phillips and Hayman (11). Fresh root segments were stained with 0.01% Trypan blue in lactic acid. The stained roots were distributed in a glass petri-dish in which a grid with 0.5 × 0.5 inch squares was affixed to the base (Fig. 1). Total number of intersects between lines and roots (R1) and total number of intersects where the root was mycorrhized (R2) were recorded using an inverted microscope equipped with a digital camera. Percentage of AM infections were calculated using the following formula proposed by Nicolson (12):

$$\text{Percent root colonization} = (R2/R1) \times 100$$

2.4 Growth parameters and measurement of biochemical status

Morphological parameters; viz., vine length (VL), root length (RL), number of shoots (SN) and leaves (LN) and total leaf area were recorded at 30, 60 and 90 DAI.

Biochemical analyses were made 90 DAI. The leaf chlorophyll contents (a, b and total) were assessed following method suggested by (13). Fully matured leaf samples were cut and dipped in dimethyl sulphoxide (DMSO) and incubated at 70ºC for 4 h. The absorbance of the solution was then read against blank (solvent) at 645, 663 and 480 nm is using a spectrophotometer. Total phenol contents present in the leaf (LTP) and root (RTP) samples were assayed using slightly modified method proposed by Malik and Singh (14). Immature leaves/root tips (10cm in length) were collected. Foliar and root samples were dried in an oven (40ºC for 72 h) and approximately 500mg dry matter of each sample was extracted with 80% Methanol by means of a shaker (120 RPM for 24h) followed by filtering through filter paper. The supernatant was collected and evaporated to dryness. Residues were dissolved in distilled water. Folin-Ciocalteu reagent and Na_2CO_3 solution (20% w/v) were added, mixed thoroughly and placed in a hot water bath exactly for 1 min. Then it was cooled down and the absorbance was read at 650nm. Estimation of total soluble sugars (TSS) was carried out according to the method described by (15). 100 mg of fresh leaf samples were hydrolyzed by HCl in boiling water bath for 3 h and then it was neutralized with sodium carbonate and centrifuged. Thereafter, anthrone reagent was added and heated for 8 min in a boiling water bath. Then it was cooled down immediately and finally, the absorbance was measured at 630 nm.

2.5 Statistical analysis

The experiment was carried out as a complete randomized block design with factorial arrangement including four replications. The average values obtained from three plants per each pot were used for analysis. Data were analyzed by analysis of variance using the GLM procedure in SAS software (16) and mean values were compared using the Least Significant Difference (LSD) test ($P < 0.05$).

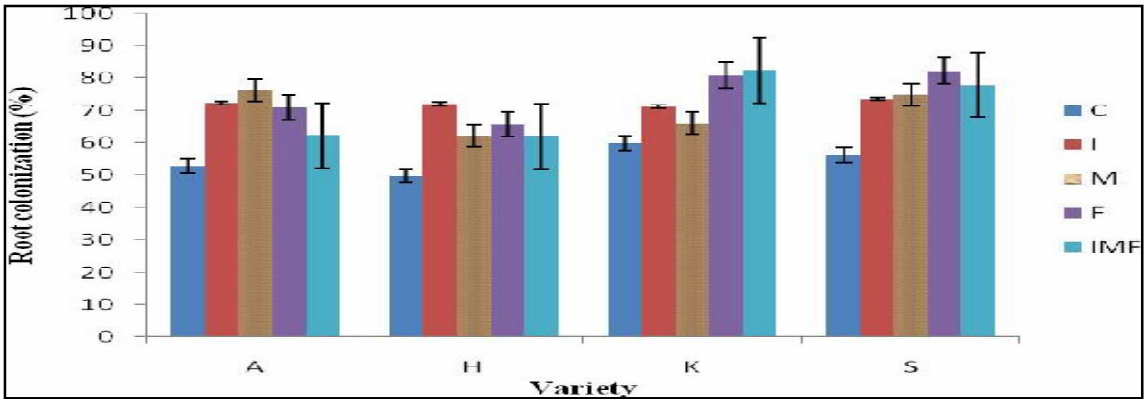

Fig. 1. Percent of AMF root colonization in four grape varieties, 90 DAI. Columns with the same letter(s) are not significantly different. A = Asgari, H = Khalili, K = Keshmeshi, S = Shahroodi, C = control, I = *G. intraradices*, M = *Glomus mosseae*, F = *G. fasciculatum*, IMF = Mixed Strains. The data are the means ± standard errors of the means (n=48).

3. Results

3.1 Root colonization

Results of root fragment staining and microscopy observations (Fig. 2) revealed that the highest root colonization occurred in Keshmeshi grape inoculated with mixed AM strains (82.6%) followed by 'Shahroodi' inoculated with *G. fasciculatum* (82.4%). The least percentage of root colonization was recorded in non-inoculated 'Khalili' (49.9%). The percentage of root colonization was found to be slightly different amongst inoculated vines but it was found to be significantly different with non-inoculated control plants.

3.2 Growth and Morpho-physiological parameters

According to ANOVA (Table 1), significant differences observed among the three different stages of sampling (30, 60 and 90 DAI) with respect to various growth parameters. Furthermore, interactions of AM inoculum and plant variety were also statistically different for the same parameters. Mean values of recorded different characters are shown in Table 2. 'Keshmeshi' plantlets inoculated with *G. mosseae* attained minimum height (10.6cm) that was not significantly different with control (12.2cm) as well. However, 'Shahroodi' plantlets inoculated with the same AM strain revealed the highest length (32.3cm). Total leaf area in treating plants was either different among AM strains (Khalili and Keshmeshi varieties) or it was not influenced by symbiosis (Shahroodi and Asgari varieties). Some treatments increased number of shoots (Table 2). Changes in number of leaves were found to be more relevant in the case of 'Asgari' and 'Keshmeshi'. Though the higher number of leaves and longer roots were produced in cuttings inoculated with AMF strains as compared to control but, overall it can be perceived that these traits were not significantly affected by any fungal interventions.

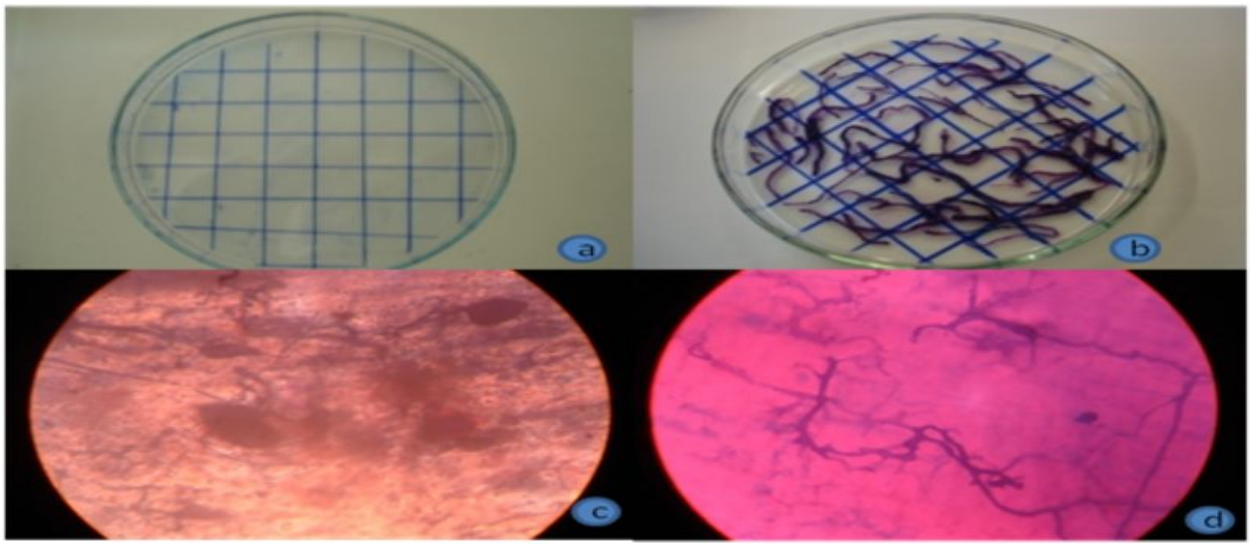

Fig. 2. Squared petri-dish utilized for measurement of root colonization (a), stained root segments distributed in petri-dish (b), root colonization occurred in Shahroodi variety plants growing on leaf-mould (c) and inoculated with *G. mosseae* (b) 90 days after inoculation.

Table 1. ANOVA table (Pr > F values) for the effects of variety, AM inoculum, and their interaction on AM root colonization, physiological and biochemical parameters.

Treatment	PRC*	Cha	Chb	TCh	LTP	RTP	TS	LN	SN	TLA	VL	RL
Inoculum (AM)	<.0001	0.9135	0.0028	0.0030	0.0407	0.0352	0.0023	0.0571	0.0054	0.6080	0.1584	0.0287
Variety (V)	0.0010	0.1034	0.4880	0.3412	0.4172	0.0137	0.0003	<.0001	<.0001	<.0001	<.0001	<.0001
AM × V	<.0001	0.4840	0.0061	0.0123	0.0424	0.0383	0.0368	0.0048	<.0001	<.0001	<.0001	0.0422
Stage	-	-	-	-	-	-	-	<.0001	<.0001	<.0001	<.0001	<.0001
Block	0.1407	0.0001	0.1227	0.0214	0.9822	0.5030	<.0001	0.0857	0.6625	0.0864	0.9956	0.8917

(*PRC-Percent root colonization, SN-shoot number, TLA-total leaf area, LN-leaf number, VL-vine length, RL- root length, TS-total sugars, Cha-chlorophyll a, Ch b-chlorophyll b, TCh-total chlorophyll, LTP- leaf total phenol, RTP-root total phenol).

Table 2. Morphological parameters of grape cuttings following Inoculation with different AMF strains.

Variety	Inoculum	SN	TLA (cm^2)	LN	VL (cm)	RL (cm)
Asgari	*G. mosseae*	1.11 ± (0.1) bcd	560.2 ± (55.7) a	14.7 ± (2.2) abc	20.3 ± (3.0) cdef	12.4 ± (2.6) ab
	G. intraradious	1.78 ± (0.2) a	460.0 ± (30.3) abcd	17.2 ± (2.1) a	16.6 ± (2.4) def	13.1 ± (2.6) ab
	G. fasciculatus	1.67 ± (0.2) a	469.7 ± (44.3) abcd	17.0 ± (2.2) ab	19.2 ± (2.7) cdef	15.5 ± (3.0) ab
	Mixed AMF	1.42 ± (0.1) ab	526.2 ± (59.6) a	15.0 ± (2.3) abc	19.1 ± (3.2) cdef	13.3 ± (3.0) ab
	Control	1.33 ± (0.2) abc	488.8 ± (76.4) abc	14.9 ± (2.7) abc	14.0 ± (2.7) ef	10.6 ± (2.7) abc
Khalili	*G. mosseae*	1.30 ± (0.2) abc	189.7 ± (31.0) g	10.5 ± (2.0) bcd	16.0 ± (2.1) def	12.1 ± (1.9) abc
	G. intraradious	1.61 ± (0.1) ab	242.7 ± (23.8) fg	14.7 ± (1.6) abc	22.3 ± (2.3) bcde	16.1 ± (2.2) a
	G. fasciculatus	1.47 ± (0.1) ab	316.0 ± (28.9) defg	14.7 ± (1.8) abc	24.9 ± (2.8) abcd	14.8 ± (1.8) ab
	Mixed AMF	1.58 ± (0.2) ab	323.7 ± (41.4) efgh	15.8 ± (2.3) ab	22.6 ± (2.6) bcde	13.6 ± (1.7) ab
	Control	1.61 ± (0.2) ab	275.6 ± (46.4) efg	13.6 ± (2.2) abcd	19.7 ± (2.9) cdef	11.9 ± (1.7) abc
Keshmeshi	*G. mosseae*	0.86 ± (0.2) cd	213.5 ± (42.5) fg	7.94 ± (1.6) d	10.6 ± (2.4) f	8.25 ± (2.2) bc
	G. intraradious	1.08 ± (0.1) bcd	397.7 ± (51.0) bcde	11.3 ± (1.8) abcd	16.9 ± (3.4) def	9.26 ± (2.3) abc
	G. fasciculatus	1.36 ± (3.0) abc	359.9 ± (53.6) cdef	12.2 ± (0.2) abcd	18.2 ± (3.0) cdef	10.2 ± (2.4) abc
	Mixed AMF	0.86 ± (0.1) cd	296.8 ± (43.2) efg	9.03 ± (1.3) cd	15.0 ± (2.9) ef	9.62 ± (2.2) abc
	Control	0.67 ± (0.1) d	335.9 ± (76.6) defg	8.06 ± (1.9) d	12.2 ± (3.6) f	5.20 ± (2.0) c
Shahroodi	*G. mosseae*	1.50 ± (0.1) ab	565.7 ± (36.2) a	13.9 ± (1.6) abcd	32.3 ± (3.5) a	16.6 ± (2.1) a
	G. intraradious	1.47 ± (0.1) ab	579.9 ± (41.3) a	13.8 ± (1.5) abcd	31.4 ± (3.3) ab	14.1 ± (1.3) ab
	G. fasciculatus	1.30 ± (0.1) abc	465.0 ± (37.9) abcd	10.8 ± (1.3) abcd	25.7 ± (3.1) abcd	13.0 ± (1.8) ab
	Mixed AMF	1.30 ± (0.1) abc	498.8 ± (47.2) abc	12.3 ± (1.5) abcd	27.6 ± (3.1) abc	12.7 ± (1.6) ab
	Control	1.11 ± (0.06) bcd	589.2 ± (46.8) a	11.8 ± (1.3) abcd	29.9 ± (3.4) ab	14.3 ± (1.8) ab

[SN: shoot number; TLA: total leaf area; LN: leaf number; VL: vine length; RL: root length. The data are the mean values ± standard errors (n=48)].

Table 3. Biochemical changes of grape cuttings following inoculation with different AMF strains.

Variety	Inoculum	Ch a (mg/g FW)	Ch b (mg/g FW)	TCh (mg/g FW)	TSS (%)	LTP (mg/100g DW)	RTP (mg/100g DW)
Asgari	*G. mosseae*	158.7 ± (6.3) a	37.6 ± (3.3) a	16.2 ± (1.2) ab	2.42 ± (0.1) abcd	29.2 ± (2.7) b	30.8 ± (5.2) abc
	G. intraradious	154.7 ± (19.7) a	30.0 ± (1.0) abcd	13.2 ± (0.6) bcd	3.60 ± (0.6) a	54.8 ± (10.3) ab	25.8 ± (7.9) abc
	G. fasciculatus	154.0 ± (1.1) a	34.4 ± (2.2) abc	15.0 ± (0.8) abc	3.00 ± (0.2) abcd	42.2 ± (7.2) ab	30.4 ± (3.8) abc
	Mixed AMF	193.7 ± (15.5) a	28.5 ± (1.2) bcd	13.5 ± (0.3) abcd	1.90 ± (0.07) d	53.4 ± (20.4) ab	34.5 ± (9.6) ab
	Control	145.2 ± (9.3) a	25.5 ± (1.7) cd	11.7 ± (0.6) cd	2.62 ± (0.2) abcd	32.5 ± (8.5) b	20.3 ± (4.9) abc
Khalili	*G. mosseae*	196.5 ± (13.3) a	33.5 ± (3.0) abc	15.5 ± (1.2) ab	2.62 ± (0.3) abcd	29.9 ± (5.4) b	17.7 ± (6.3) bc
	G. intraradious	186.7 ± (7.4) a	34.1± (2.4) abc	15.2 ± (0.9) abc	3.32 ± (0.4) abc	55.1 ± (23.7)ab	18.9 ± (4.5) bc
	G. fasciculatus	172.0 ± (7.6) a	30.3 ± (0.6) abcd	13.7 ± (0.2) abcd	2.62 ± (0.4) abcd	22.8 ± (1.1) b	32.4 ± (1.2) abc
	Mixed AMF	155.0 ± (27.8) a	38.8 ± (3.2) a	17.0 ± (1.2) a	2.27 ± (0.2) bcd	71.3 ± (20.3) a	28.2 ± (2.9) abc
	Control	155.0 ± (12.3) a	28.0 ± (4.0) bcd	12.8 ± (1.7) bcd	2.17 ± (0.3) cd	24.8 ± (3.5) b	29.2 ± (6.2) abc
Keshmeshi	*G. mosseae*	175.0 ± (6.4) a	36.2 ± (2.4) ab	16.0 ± (0.8) ab	2.77 ± (0.2) abcd	23.7 ± (3.9) b	15.5 ± (0.7) c
	G. intraradious	169.2 ± (13.4) a	33.4 ± (1.7) abc	14.7 ± (0.5) abcd	2.87 ± (0.3) abcd	22.8 ± (0.7) b	20.6 ± (3.5) abc
	G. fasciculatus	196.7 ± (32.3) a	35.2 ± (1.1) ab	15.7 ± (0.2) ab	2.52 ± (0.5) abcd	38.8 ± (6.9) ab	16.7 ± (2.6) c
	Mixed AMF	180.7 ± (22.8) a	27.7 ± (1.3) bcd	13.0 ± (0.8) bcd	2.70 ± (0.1) abcd	30.0 ± (3.2) b	24.2 ± (0.9) abc
	Control	195.0 ± (24.1) a	23.2 ± (5.7) d	11.3 ± (1.8) d	3.32 ± (0.4) abc	47.9 ± (10.3) ab	16.4 ± (2.5) c
Shahroodi	*G. mosseae*	170.0 ± (10.2) a	34.5 ± (2.5) abc	15.2 ± (1.03) abc	3.52 ± (0.4) b	29.1 ± (1.7) b	20.6 ± (1.9) abc
	G. intraradious	174.0 ± (9.5) a	31.2 ± (1.4) abcd	14.2 ± (0.5) abcd	3.70 ± (0.5) a	39.1 ± (5.4) ab	19.0 ± (4.8) bc
	G. fasciculatus	183.7 ± (15.9) a	34.1 ± (1.8) abc	15.2 ± (0.8) abc	2.92 ± (0.3) abcd	44.6 ± (9.1) ab	36.2 ± (7.4) a
	Mixed AMF	164.0 ± (9.4) a	30.2 ± (3.2) abcd	13.7 ± (1.3) abcd	3.52 ± (0.7) ab	41.8 ± (2.9) ab	29.0 ± (5.9) abc
	Control	176.7± (16.2) a	35.2 ± (3.6) ab	15.7 ± (1.7) ab	3.07 ± (0.2) abcd	25.2 ± (3.7) b	32.2 ± (3.5) abc

[Cha: chlorophyll a; Ch b: chlorophyll b; TCh: total chlorophylls; TSS: total soluble sugars; LTP: leaf total phenols; RTP: root total phenols. The data are the mean values ± standard errors (n=48)].

4. Discussion

The AMF association being the commonest mycorrhizal type involved in agricultural systems (3) and the variability of AM species in their ability to improve the growth of different plant species has been largely demonstrated (17). It is necessary to determine the best mycorrhiza corresponding special plant varieties. It has been recognized that the creation of a permanent relationship between host and fungus is in result of the identification and approval of molecular signals by both symbionts, which consequence in genome expression of both organisms and it can be understood that the percentage root colonization is under control of plant genotype (3, 18). It might be for the same reason that, in the present study, different grape genotypes colonized with a varying degree following inoculation with selected mycorrhizal strains. Such variations in root colonization among genotypes of a species have been already confirmed in some grapevine rootstocks (5, 8) and some other plants namely, wheat (19), corn (20) and citrus (21). In the majority of these studies, the sterile media and/or fumigated soils were used but we have used a natural, non-fumigated mixture (commonly used medium for grape propagation in Iranian nurseries) and as a result some amount of colonization was also observed in non-inoculated, control plantlets that was actually due to the presence of native fungal strains.

Since, in the present study, clonally propagated plant materials (i.e. Hardwood cuttings) were used, the uniform growth pattern might be expected, hence, any morphological amelioration could be attributed to the fungal interference. However, irrespective of any fungal intervention, the overall measured growth parameters suggested that 'Shahroodi' was the most vigorous variety followed by 'Khalili', 'Asgari' and 'Keshmeshi'. It is obvious that in our morphological data (Table 2), integration of AM fungi to a nursery bed simply enhanced the growth parameters. However, this improvement was considerably different with regard to the type of fungal strain used, for example, all inoculated plantlets generally gained higher length than control, but given a particular strain, different vine lengths were observed in four inoculated grape varieties. However, irrespective of the statistical aspects, an elevated trend could be observed for vine length as well as other morphological characters following microbial treatments. The inoculation resulted in higher growth rate of the mycorrhizal plants but the degree of enhancement was limited to host-fungi interaction.

Although, our results obtained for number of leaves per vine is in incongruity with the findings of (22) on pepper plants that AM-inoculated plants showed the lowest number of leaves, the same is in agreement with the results obtained on Chrysanthemum (23), Guava plantlets (24), *Salix repens* (25) and olive (26). Furthermore, it was found that *Glomus fasciculatum* had most effect on 'Keshmeshi' grapevine growth which was corresponded by the works of Bheemareddy and Lakshman (19) on some *Glomus* species. Considering morphological characters (Table 2), mycorrhizal inoculation did not increase significantly the growth rate of 'Shahroodi' plants, but according to Schiavo *et al.*, (27), AMF inoculation increased the height of *Acacia* sp. and *Sesbania* sp. as compared to control under glass-house conditions.

Different AMF strains varied in their efficacy to increase the synthesis of different biochemical, thereby improving the plant growth. These differences may depend on the genetically controlled physiological characters of the fungal strains (18).

In this study, increased total chlorophyll and chlorophyll 'b' in plants of 'Asgari' and 'Keshmeshi' grapevines is similar to the findings of Bavaresco and Fogher (28) on the effect of *G. mosseae*. However, in case of 'Khalili' grape, the mixed AM strain treatment lead to the highest biosynthesis of chlorophylls. The positive effect of AM symbiosis on chlorophyll content was also reported in Maize (29), Sesbania (30), Lotus (31) and Zucchini (32). Furthermore, Estrada-Luna (24) indicated that leaf chlorophyll may vary according to light conditions (or other factors such as the mineral status of the plants, in which N, Mg, Cu and Fe have important roles). Increased chlorophyll content after AM inoculation has also been reported by Mathur and Vyas (33, 34); Krishna *et al.*, (5, 8). Who suggested that the high chlorophyll content in mycorrhizal plants may be due to the higher concentrations of Mg, Fe and Cu in foliar tissues thereby influencing chlorophyll synthesis. Mycorrhizae regulate not only uptake but also the relative abundance of available and transportable nutrients in the tissue concentration of essential micronutrients like Cu and Zn. Siderophores are formed by mycorrhizal fungi that enable the fungus to take up Fe from solutions in low amounts (17).

Phenols are important components of plant defense mechanism against the diseases. Phenolic compounds occur naturally in plant system and owing to their antimicrobial properties inhibit fungal spore germination and toxin production by pathogens (35). In the present study, inoculated grape cuttings accumulated higher phenolic compounds in their root as well as foliage. The mixed AM inoculum was found the most efficient one in enhancing leaf phenolics in 'Khalili'. Earlier, Tang *et al.*, (36) reported that a significant increase in the level of phenolic compounds in the bark of AM-inoculated poplar plants. The increased level of total phenols suggests higher resistance of inoculated plants against diseases, which led to increased plant survival under nursery or glass-house as well as field conditions (36). Furthermore, organic grown tomatoes had increased total phenolic contents in their fruits as a result of the AMF treatment (37), Devi and Reddy (38); Kapoor (39) showed that

AM inoculation induced quantitative and qualitative changes in phenolics of groundnut and tomato, respectively. In addition, inoculated *in vitro* grown grapevine plantlets had the higher phenols during their hardening period (5, 8).

In conclusion, taking overall account of the results obtained in the present study, it can be stated that AM fungi can be manually applied in the nursery, where moderate amounts of colonization are often naturally achieved so that following transplanting to the vineyard they could colonize and enhance plant growth and production. Future research works must be undertaken on the effects of these fungi on the performance of such developed cuttings under field conditions and upon fruiting.

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