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Original Article

Effects of arbuscular mycorrhizal fungus *Glomus mosseae* and phosphorus application on plant growth rate, essential oil content and composition of coriander (*Coriander sativum* L.)

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Summary. This study evaluated the interactive effect of inoculation of *Glomus mosseae*, an arbuscular mycorrhizal fungus (AMF), and exogenous phosphorus (P) on growth, gas exchange, different nutrients, oil contents and composition in fruits of *Coriander sativum* L. Two contrasting concentrations of KH_2PO_4 (F0–without P, F1–100 mg kg^{-1}) were applied. Soil P supply significantly promoted all growth parameters, P and N concentrations and most photosynthetic parameters of both mycorrhizal and non-mycorrhizal plants. It also partially inhibitions soil acid (ACP) and alkaline phosphatase (ALP). The mycorrhizal inoculation significantly increased growth responses, plant nutrients (P and N in shoot and root tissues), ACP, ALP, total soluble proteins in root tissues, fruit yield and essential oil contents compared to nonmycorrhizal plants. Those stimulations were related to the level of mycorrhizal colonization in the root tissues for each treatment. Mycorrhizal plants showed higher net photosynthetic rate, stomatal conductance and transpiration rate than nonmycorrhizal plants, especially in soil without added phosphorus. This suggests that an addition of soluble P to soil generally reduces the percentage of mycorrhizal colonization levels in coriander root tissues and consequently the mycorrhizal benefits. The results showed that coriander plants were highly dependent on AMF in non-fertilized soil when compared to fertilized soil with phosphorus. Essential oil content in plants inoculated was significantly higher than other treatments. Furthermore, oil composition improved with AMF inoculation. Linalool that enhance the essence quality, increased in inoculated plant compared to non-inoculated plants particularly in P deficient soil. Inoculation of arbuscular mycorrhizal fungi into coriander plants is a feasible alternative to increase growth, nutrition, essential oil production and reduce the use of P fertilizers required to obtain economic production of coriander under phosphorus-deficient soil condition.

Key words: Acid and alkaline phosphatases, arbuscular mycorrhiza, nutrition, coriander, gas-exchange, essential oils.

Introduction

Phosphorus (P) is one of the major essential macronutrients for biological growth and development. Plants have the ability to uptake immobile nutrients, such as phosphorus via forming a symbiosis relationship with mycorrhizal fungi via plant root. The arbuscular mycorrhizal (AM) fungi are found in a wide range of soils, they are playing a positive role to the plants in all the soils where they are found in especially in P deficient soils as their hypha have the ability to uptake P and transfer it to the plants (1, 2). Mycorrhizal fungi forms a mutual symbiotic relationship with plant, from this relationship the mycorrhizal fungi take the carbon, and the plant takes nutrients, as the fungal hypha is spread out in the soil, mycorrhizal vesicles work as connection points between the external hyphae and the plants (3). The spreading of mycorrhizal hypha in the soil raises the active absorbance area of the plants, reaching areas that the roots cannot reach alone, which finally leads

to supporting the plants with nutrient like P (which in the normal non-mycorrhizal cases are out of plant reach) (4). This mycorrhizal mediated absorbed nutrient is then transferred to plants via plant roots, thereby decreasing the ratio between the distances of P diffusion to the absorbing surface (5). The concentration on accumulated secondary compounds in the shoots and leaves of mycorrhizal plants has been raised in the recent years. Mycorrhization of coriander plants has resulted in improved essential oil content attributable to an improved P status of the plant (6). (7) also observed that inoculation with AM fungi led to an increase (89%) in the essential oil and menthol contents of *Mentha arvensis* plants. (8) reported that *Glomus mosseae* directly increases the essential oil content in shoots of different species of Apiaceae plants. (9) observed that the increase in essential oil concentration in mycorrhizal oregano (*Origanum vulgare* L) plants is not due to an improved P nutrition, but directly depends on association with the AM fungus.

Soil acid phosphatase (ACP) and alkaline phosphatase (ALP) are of particular importance in the enzyme system participated in P absorption, assimilation and metabolism. The ability of mycorrhiza to accumulate and store polyphosphate in their hypha have been reported and studied by many authors like (10). The arbuscules are mycorrhizal cells found in the cortex of roots of arbuscular mycorrhizal plants these arbuscules have phosphatase which might be important in the transfer of P from the hypha to the plant tissues. AS reported by (11), there is high content of phosphatase in the root area of mycorrhizal plants this phosphatase may be a product of the mycorrhiza itself or of the symbiont plant root. Mycorrhizal plants uptake P from the same soil that no-mycorrhizal ones are planted in, so it may be predicted that mycorrhizal phosphatase has a role in utilizing organic P (12). The mycorrhizal phosphatase is predicted to have a role in the hydrolysis of phytate, this was the case in (13) experiment on adding calcium phytate to soybeans, as the content of available P was higher in the root area (rhizosphere) of mycorrhizal plants compared to that of nonmycorrhizal ones.

Coriander is an annual plant from the Apiaceae family with a 90-120 days growth period. In many countries it is grow as a spring plant and in some others as a winter plant. Traditionally the coriander plants (fruits and the green herb) were used for two main purposes, which are medicinal and culinary uses. The green coriander her is used in making sauce and as a flavor for curries and soups , the fruits are considered carminative diuretic, tonic, stomachic, antibilious, refrigerant and aphrodisiac (14). Linalool is one of the main contents of coriander oils extracted from fruits (15) this essential oil is antibacterial (16) and antioxidant (17). AM fungi have emerged as potential biofertilisers, a cheap, environmentally friendly alternative to expensive chemical fertilisers (18). There is a key role played by AM fungi in improving growth and some metabolic processes of plants, which raised the productivity of these plants raising their economical profit in the agricultural sectors. Despite what is stated in the review of this paper till now about the useful effect of mycorrhiza on plants, the available information about the mycorrhizal effects on aromatic and medicinal plants is very little (8, 19, 20, 21 and 22).

Therefore, the aim of this work was to study the effects of soil P fertilization in association with AM fungi, *G. mosseae* (Nicol. & Gerd.), on growth, yield, acid and alkaline phosphatases, gas exchange parameters, nutrient content, and essential oil concentration and quality of coriander plants with an aim to reduce the application of chemical fertilizer for sustainable system.

Materials and Methods

Experimental design

Pots filled with soil (see below) were placed under greenhouse conditions where light intensity was 225 μmol m^{-2} s^{-1} with a light period of about 16 hours, the temperature daytime and night time were 27/20°C and the relative humidity was from 70 to 80 percent. The experiment was arranged as a two factorial randomized complete block design with the following factors: (a) inoculation with [*G. mosseae* (AMF) and non-inoculated (non-arbuscular mycorrhizal plants (Non-AMF))], (b) KH_2PO_4 supply (F0–without P and F1–100 mg kg^{-1} P).The replication number for all of the treatments was 10 replications, so there were four treatment and ten replications which make a forty pots (one plant in every pot).

Mycorrhizal fungal inoculum preparation

The mycorrhizal propagules were prepared through trap culturing mycorrhizal fungus (containing *Glomus mosseae*) on the roots of sudangrass (*Sorghum halepense* L.) as the inoculum taken with this way contained soil and infected root parts, the plants of sudangrass were planted under water stress conditions in the Experimental Station of Plant Production Department, College of Food and Agriculture Sciences, King Saud University. Twenty pots (50 % of the experiment) were inoculated with fifteen grams of the inoculum, the other half received the same amount of spore washing water without spores (referred as non-mycorrhizal plants).

Soil characteristics

The surface soil (0-20 cm) was collected from Dirab Experimental Agricultural Research Station, Riyadh region, Saudi Arabia. The soil was air dried and ground to pass through a 2-mm sieve. Some physical and chemical properties of the planting media were: sandy loam texture, silt 18%, clay 5.6%, sand 70.4%, organic matter 0.43%, $CaCO_3$ 0.9%, pH (1:2 H_2O) 7.4; an available nitrogen (25.0 mg kg^{-1}); an available phosphorus (7.02 mg kg^{-1}); a potassium (52 mg kg^{-1}); a magnesium (85 mg kg^{-1}) and EC (0.35 dS m^{-1}). Before loaded to pots, the soil was air-dried, passed through 2-mm sieve, and autoclaved at 120°C for 2 h in order to kill other fungi spores. Phosphorus was added as an aqueous solution of KH_2PO_4, sequentially, at the concentration previously mentioned. After thorough mixing, the soil was allowed to stabilize for 30 days before seedling planting, and three samples of each treatment were taken for chemical analysis.

Plant growth conditions

Coriander seeds (*Coriandrum sativum*) were surface-sterilized with 70% alcohol for 15 min and placed on sterilized moist filter paper for germination in darkness at 28°C. Homogeneous seedlings were transplanted into 3x3x3.5 inch plastic pots filled with moist-autoclaved soil (vermiculite) and grown under the previously greenhouse described conditions. Two-litre plastic pots were filled with sterilized soil at the bottom (non-rhizosphere soil), and a layer of 500 mesh nylon net (pore size of 30 μm) with the sterilized soil of 2 mm thickness was placed above the bottom layer (rhizosphere soil), then a 500 mesh nylon bag (15 cm in diameter, 8 cm in depth) filled with soils with or without AMF inoculum was put on the top. For the inoculated group (IM), 100 g of prepared inocula were thoroughly mixed with the soil, while the same amount of sterilized inocula were mixed in the non-inoculated group (NM). The interspace between nylon bag and the pot was filled with sterilized soil. The same amount were added to the Non-AMF plants as filtered washings in order to provide the same associated microorganisms other than mycorrhizal propagules. Plants were watered to maintain soil moisture at 60%–70% of the water-holding capacity by adding sterilized tap water during the experimental period, and it was fertilized biweekly with Hoagland's solution without P at a rate of 50 ml per pot.

Measurement of plant growth

The experiment was terminated when all plants possessed ripe fruits. Plants of coriander were harvested 9 weeks after seed sowing and inoculation. Shoots and root dry weights, plant height and number of branches were recorded to evaluate treatment response. The ratio between roots and shoots as dry weights for each treatment was determined. The Li-3000A area meter (LiCor Biosciences, Nebraska, USA) was used to determine the leaf area. Arbuscular mycorrhizal growth responses (AMR) was determined by calculating the growth percentage of plants that has been grown under AMF conditions, and calculated by (23) modification of (24) formula: AMR% = [(dry weight inoculated/dry weight non-inoculated) × 100]. Similarly, AMR was calculated for all data among mycorrhizal and non-mycorrhizal treatments in each estimated parameter.

Phosphorus and nitrogen determinations

Shoot (SDW) and root dry weights (RDW) of seedlings were determined after harvesting. Samples were oven-dried at 68°C for 48 h. Dried samples were ground, digested in a nitro-perchloric solution as described by (25). Phosphorus content in the digested samples was determined according to (25) with UV

spectrophotometer at 700 nm. Nitrogen was determined using the macro-Kjeldahl digestion method (26).

Gas exchange measurements

Two months after inoculation (85 d of growth) gas exchange measurements [net photosynthesis (A), stomatal conductance (g_s) and transpiration rate (E)] were performed. However, before measurements of gas exchange parameters, the plantlets were acclimated for 20 min on light produced by a 1,000W metal halide lamp (photosynthetic photon flux was 1,000 μmol m^{-2} s^{-1} at plant height), which was filtered through a 5-cm non-circulating water bath enclosed in a Plexiglas box. Measurements were performed between 8:00 and 11:00 on each leaf of the first pair of fully expanded leaves from the shoot apex using a LI–6200 Portable Photosynthesis System (LI-COR Inc., Lincoln, NE, USA). Each leaf was a single replication, and there were 10 replications per treatment (n = 10), which represented four plants per treatment.

Determination of root colonization

Collected fresh root samples were rinsed three times with tap water. In order to visualize the AM mycorrhizal colonization the roots were cleared using 10% KOH and the staining was made by the Trypan blue (0.1%) pigment in lactoglycerol (27). To measure the percentage of colonized roots was measured using 100 × magnification according to (28) method. The compound microscope (400 × magnifications) was used to examine root segments taken from the mycorrhizal treated plants. Four to nine 1-cm segments were mounted on glass slides and rated using a scale from 0 to 4 in order to evaluate the frequency of mycorrhizal colonization (F %), presence and intensity of infection (M %) and rate of arbuscular development (A %) of the stained roots. These parameters were estimated by the method of (29).

Determination of phosphatase activity

The rhizosphere and non-rhizosphere soils were separately and thoroughly mixed after roots were removed at harvest and 100g fresh soil was taken from each part. Soil samples were freeze-dried for the assay of phosphatase activities, which were determined by measuring the *p*-nitrophenol (mg PNP g^{-1} h^{-1}) released by phosphatase activity using *p*-nitrophenyl phosphate disodium (PNPP) as substrates. The soil was incubated with buffered (pH 6.5 for ACP, pH 11 for ALP) sodium *p*-nitrophenyl phosphate solution and toluene at 37°C for 24 h, which was modified according mainly to the method proposed by (30).

Total soluble protein determinations

Total soluble protein in root extract was determined by (31) method. The determination of protein content was made using standard curve with bovine serum albumin (Sigma-Aldrich Chemie, Steinheim, Germany).

Essential oil extraction and estimation

Essential oils were determined in non-mycorrhizal in mycorrhizal plants inoculated with the AMF. Samples for essential oil isolation were taken at the end of the experiment. Mature coriander fruits were air-dried at room temperature for 20 d and then stored at room temperature and ambient humidity for 6 months. The hydro-distillation method with a modified Clevenger trap was used to extract the essential oil of coriander. Coriander mature fruits (100g) were crushed and homogenized sung an electric grinder with distilled water. The pulverized mass was loaded in to the still for distillation. The condenser cooled the hot vapours received from the still. The jet of vapours consisting of steam and essential oil were cooled in the condenser tubes and condensate flowed out into the receiver. The oil was drawn off and the volume of oil was noted and percentage of essential oil content measured in volume/100 g fresh weight basis.

The essential oil was analysed with gas chromatography/mass spectrophotometer (GC/MS) for chemical analysis of oil compounds. GC/MS analyses were performed on a Varian 3400 in the following analytical conditions: helium as carrier gas; 1 ml of sample injected at 250°C with a column flow of 1.2 ml/min in a DB-1 column (60 m×0.25 mm, 0.25 μm film thicknesses). Components were identified according to databases and quantified by comparison with certified standards for eight oils. Authentic reference substances, α-Pinene,

β-Pinene, *p*-Cymene, *δ*-Linalool, Geraniol, *t*-Anethol, *β*-Elemene, and *β*-Caryophyllene (Sigma-Aldrich, India), were used to detect normal reference times.

Statistical analysis

Two-factor analyses of variance (ANOVA) were used to investigate whether there was a significant difference among various experiments used. Means were separated by Duncan's multiple range tests. *P* values $P \leq 0.05$ were considered significant. All of the measurements were achieve four times for each treatment, and the mean and standard error (SE) was mentioned.

Results

Growth characters and plant productivity

As shown in Table 1 addition of P improved growth parameters in plants such as height, shoot and root DM, branches number, and LA of both AMF and Non-AMF coriander plants. Growth was higher in AM plants compared to nonmycorrhizal ones regardless of P level. However, the stimulations in growth criteria due to mycorrhizal colonization (AMR) were more pronounced in plants grown in P-deficient soil and are precisely proportional to the level of mycorrhizal colonization in root tissues. Root-to-shoot dry mass production ratio of mycorrhizal plants was significantly lower than non-mycorrhizal coriander plants grown in P-deficient soil (Table 1). In general, no significant differences in root-to-shoot dry mass yield ratio were noticed between mycorrhizal and non-mycorrhizal coriander plants which planting in soil amended with phosphorus.

P and N contents

Generally, nutrient aggregation in either shoots or root tissues of coriander was significantly affected by its utilization to soil and by AM inoculation. P and N contents in shoots and roots of mycorrhizal plants were much higher than those of nonmycorrhizal plants particularly in P-deficient soil (Table 3). Concentrations of P and N in mycorrhizal plants were higher than in nonmycorrhizal plants particularly in P-deficient soil. On the other hand, such increases in nutrient contents in response to the mycorrhizal effects (AMR) were highly associated with the intensity of mycorrhizal colonization for each treatment.

Changes in gas exchange parameters

Values of *A*, *E* and g_s, in leaves of mycorrhizal coriander plants were significantly greater than those in nonmycorrhizal plants grown either in P fortified or unfortified soil (Table 2). The effect was more re-

Table 1. Effect of phosphate fertilizer and mycorrhizal inoculation on the growth response of *C. sativum* plants grown in sterilized soil.

Treatments		(Dry mass)					
P (KH_2PO_4) (mg kg^{-1})	**AMF status**	**Shoot (g $plant^{-1}$)**	**Root**	**Root/shoot ratio (%)**	**Plant height (cm $plant^{-1}$)**	**Branches number ($plant^{-1}$)**	**Leaf area (mm^2 $plant^{-1}$)**
F0	Non-AMF	4.25 ± 0.53^{C}	1.37 ± 0.16^{B}	0.462 ± 0.02^{A}	80.3 ± 3.41^{C}	6.4 ± 0.37^{B}	248 ± 7.04^{C}
Without P	AMF	6.67 ± 0.64^{B}	2.36 ± 0.35^{A}	0.386 ± 0.03^{B}	96.4 ± 4.90^{B}	8.5 ± 0.15^{A}	365 ± 8.12^{B}
	AMR (%)	84.2 ± 2.35	62.0 ± 2.14	–	32.4 ± 0.68	45.7 ± 1.60	45.7 ± 1.83
F1	Non-AMF	5.81 ± 0.56^{B}	1.94 ± 0.36^{B}	0.365 ± 0.01^{Bc}	97.4 ± 4.70^{B}	7.4 ± 0.56^{B}	379 ± 7.34^{B}
100	AMF	7.35 ± 0.62^{A}	2.43 ± 0.17^{A}	0.421 ± 0.01^{B}	106.3 ± 5.95^{A}	9.4 ± 0.45^{A}	451 ± 8.23^{A}
	AMR (%)	39.5 ± 1.93	38.7 ± 1.76	–	13.5 ± 0.65	28.7 ± 1.64	17.2 ± 0.57
LSD (0.05)		0.640	0.304	0.038	6.340	0.630	15.43

Values in each column (except AMR) followed by the same letter(s) are not significantly different at $p \leq 0.05$ (Duncan's multiple range test). Each value represents the mean of five replicates ± SE. AMF – arbuscular mycorrhizal fungi, Non-AMF – nonmycorrhizal fungi, AMR – arbuscular mycorrhizal response.

Table 2. Net photosynthetic rate (*A*), transpiration rate (*E*) and stomatal conductance (g_s) in leaves of mycorrhizal (AMF) and non-mycorrhizal (Non-AMF) *C. sativum* plants grown in sterilized soil with or without phosphorus fertilizer.

Treatments		Gas exchange parameters		
P (KH_2PO_4)	**AMF status (mg kg^{-1})**	**A (µmol m^{-2} s^{-1})**	**E (mmol m^{-2} s^{-1})**	**g_s (mol m^{-2} s^{-1})**
F0	Non-AMF	06.48 ± 0.51^{C}	4.063 ± 0.41^{C}	0.127 ± 0.023^{B}
Without P	AMF	09.53 ± 0.39^{A}	6.239 ± 0.56^{A}	0.186 ± 0.03^{A}
	AMR (%)	31.7 ± 2.05	42.1 ± 1.76	53.7 ± 2.36
F1	Non-AMF	07.54 ± 0.71^{B}	4.652 ± 0.47^{B}	0.129 ± 0.021^{B}
100	AMF	10.54 ± 0. 64^{A}	6.132 ± 0.62^{A}	0.179 ± 0.036^{A}
	AMR (%)	21.6 ± 1.43	31.7 ± 1.86	43.7 ± 1.870
LSD (0.05)		0.930	0.204	0.051

Values in each column (except AMR) followed by the same letter(s) are not significantly different at p ≤ 0.05 (Duncan's multiple range test). Each value represents the mean of five replicates ± SE. AMR – arbuscular mycorrhizal response.

Table 3. Concentrations (%) of phosphorus (P) and nitrogen (N) in shoots and roots of mycorrhizal (AMF) and nonmycorrhizal (Non-AMF) *C. sativum* plants grown in sterilized soil with or without phosphorus fertilizer.

Treatments		P %			N %		
P (KH_2PO_4) (mg kg^{-1})	**AMF status**	**Shoot**	**Root**	**Total**	**Shoot**	**Root**	**Total**
F0	Non-AMF	0.16 ± 0.005^{C}	0.15 ± 0.005^{C}	0.31 ± 0.015^{D}	2.76 ± 0.26^{C}	1.76 ± 0.104^{C}	4.52 ± 0.07^{C}
Without P	AMF	0.29 ± 0.007^{A}	0.24 ± 0.006^{B}	0.53 ± 0.022^{B}	3.26 ± 0.23^{A}	2.29 ± 0.173^{A}	5.55 ± 0.32^{Ab}
	AMR (%)	75.3 ± 2.43	72.3 ± 1.00	72.0 ± 1.87	11.9 ± 0..97	26.5 ± 0.75	17.6 ± 0.17
F1	Non-AMF	0.22 ± 0.005^{B}	0.23 ± 0.006^{B}	0.45 ± 0.025^{C}	2.87 ± 0.24^{B}	2.24 ± 0.226^{B}	5.11 ± 0.22^{B}
100	AMF	0.34 ± 0.006^{A}	0.28 ± 0.007^{A}	0.62 ± 0.037^{A}	3.19 ± 0.29^{A}	2.39 ± 0.213^{A}	5.58 ± 0.21^{A}
	AMR (%)	43.6 ± 1.74	29.5 ± 0.75	38.4 ± 1.06	9.7 ± 0.46	12.7 ± 0.28	11.5 ± 0.26
LSD (0.05)		0.026	0.021	0.040	0.260	0.121	0.560

Values in each column (except AMR) followed by the same letter(s) are not significantly different at p ≤ 0.05 (Duncan's multiple range test). Each value represents the mean of five replicates ± SE. AMR – arbuscular mycorrhizal response.

marked in P-deficient soil. Such stimulations in gas-exchange parameters were related to the degree of mycorrhizal colonization for each treatment.

Mycorrhizal colonization levels

Roots of inoculated plants were extensively colonized by *G. mosseae* irrespective of P, whereas roots of uninoculated plants remained uncolonized. Intensity of infection (M) and arbuscular frequency (A) were significantly reduced in coriander tissues of root with addition of P to the soil (Table 4). But, no significant differences were noticed in the rate of arbuscular frequency (F) between AMF plants which planting in soil either amended or un-amended with P. No mycorrhizal colonization was realized in the noninoculated coriander plants.

Differences in acid and alkaline phosphatase activities

Values of soil acid phosphatase (ACP) were significantly higher than those of alkaline phosphatase (ALP) in all treatments (Table 5). This suggests a major role played by ACP in soil nonmycorrhizal inoculation treatments. Furthermore, ACP and ALP activities in both rhizosphere and non-rhizosphere decreased with increasing P treatment. The ACP activities in the rhizosphere were significantly higher than that

Table 4. Frequency of mycorrhizal colonization (F %), intensity of mycorrhizal colonization (M %), and arbuscular frequency (A %) in the root tissues of mycorrhizal (AMF) and nonmycorrhizal (Non-AMF) *C. sativum* plants grown in sterilized soil with or without phosphorus fertilizer.

Treatments		Mycorrhizal colonization Levels		
P (KH_2PO_4) (mg kg^{-1})	**AMF status**	**F (%)**	**M (%)**	**A (%)**
F0	Non-AMF	0.0	0.0	0.0
Without P	AMF	93.4 ± 7.6^{A}	69.7 ± 6.8^{A}	49.2 ± 4.7^{A}
F1	Non-AMF	0.0	0.0	0.0
100	AMF	76.0 ± 8.3A	52.0 ± 6.2B	37.0 ± 4.0B
LSD (0.05)		9.43	7.13	5.52

Values in each column followed by the same letter(s) are not significantly different at p ≤ 0.05 (Duncan's multiple range test). Each value represents the mean of five replicates ± SE.

Table 5. Soluble acid and alkaline phosphatase activities, quantitative changes in protein (μg g^{-1} root fresh mass), fruit yield, essential oil content of mycorrhizal (AMF) and nonmycorrhizal (Non-AMF) *C. sativum* plants grown in sterilized soil with or without phosphorus fertilizer.

Treatments		Phosphatase activities (mU ml^{-1})			Root protein (μg g^{-1})	Fruit yield (g $plant^{-1}$)	Essential oil (%)
P (KH_2PO_4) (mg kg^{-1})	**AMF status**	**Acid**	**Alkaline**	**Total**			
F0	Non-AMF	763 ± 11.6^{B}	232 ± 1.08^{B}	995 ± 0.82^{B}	531 ± 5.92^{C}	6.1 ± 0.41^{C}	1.21 ± 0.38^{B}
Without P	AMF	843 ± 10.2^{A}	271 ± 1.09^{A}	1114 ± 0.79^{A}	685 ± 8.07^{A}	7.2 ± 0.54^{A}	1.59 ± 0.14^{A}
	AMR (%)	8.05 ± 0.76	19.3 ± 0.16	11.2 ± 0.09	51.2 ± 1.06	42.3 ± 1.78	52.0 ± 2.06
F1	Non-AMF	743 ± 8.45^{C}	187 ± 1.02^{C}	930 ± 0.52^{C}	485 ± 6.92^{C}	15.3 ± 0.47^{B}	2.29 ± 0.17^{B}
100	AMF	762 ± 8.14^{B}	214 ± 1.04^{C}	976 ± 0.76^{B}	573 ± 2.83^{B}	17.4 ± 0.60^{A}	2.65 ± 0.35^{A}
	AMR (%)	5.12 ± 0.36	5.45 ± 0.05	4.56 ± 0.05	35.8 ± 0.79	31.6 ± 1.85	53.0 ± 2.07
LSD (0.05)		23.52	15.01	31.54	26.43	0.204	0.306

Values in each column (except AMR) followed by the same letter(s) are not significantly different at p ≤ 0.05 (Duncan's multiple range test). Each value represents the mean of five replicates ± SE. AMR – arbuscular mycorrhizal response.

in the non-rhizosphere of the control; nevertheless, no significant differences were found in rhizosphere and non-rhizosphere soils with P treatment. However, ALP activities in the rhizosphere were significantly higher than that in the non-rhizosphere in all P treatments. When inoculated with AMF, ACP and ALP activities in the rhizosphere were significantly greater than that in the non-rhizosphere. The ACP activities in either rhizosphere or non-rhizosphere soils increased and reached the maximum under100 mg kg^{-1} P treatment. In contrast, ALP activities simply decreased with P treatment (Table 5).

Total soluble protein content

Total soluble protein of the mycorrhizal and nonmycorrhizal root extracts of coriander plants were greatly decreased with the addition of P to the soil (Table 5). However, colonization of coriander plants with AMF significantly increased total soluble protein in root extracts than in non-AMF plants disregarding of P treatments. This beneficial effect was more obvious in plants grown in P-deficient soil. On the other hand, no significant differences in total soluble protein contents in root extracts were observed between nonmycorrhizal coriander plants grown either in un-amended or amended soil with P (Table 5).

Fruit yield and essential oils

AM fungal inoculation significantly increased the fruit yield and content of essential oils, as compared to non-inoculated plants when no phosphorus were added (control soil) (Table 5). However, no significant differences in essential oil content were observed between the nonmycorrhizal plants grown in either P0 or P100 soil.

Essential oil extraction and estimation

Chemical characterization of essential oil by GC/MS revealed that the oil quality improved on mycorrhization with significant increase in linalool concentration. The proportion of linalool increased significantly in *G. mosseae* inoculated plants over either control or non-inoculated plants regardless the P level. Linalool was the most abundant composition of the essential oils of *C. sativum*, followed by Geraniol, *t*-Anethol, α-Pinene, *p*-Cymene, Caryophyllene, β-Elemene, α- and β-Pinene were present in lesser amounts (Table 6).

Discussion

The data presented in Table (1) indicate that the AM inoculation significantly increased growth parameters (viz. dry mass, plant height, number of branches per plant and leaf area) of coriander plants grown in soil either with or without P comparing to nonmycorrhizal plants. Such increases resulted from mycorrhizal colonization and were directly proportional to the respective level of mycorrhizal colonization. The rate of growth in response to the mycorrhizal colonization was more pronounced in P-deficient soils. (32) indicated that the AM contributes to the growth of plant *via* assimilation of immobile soil P. This is probably due to the widening the absorption surface area provided by extended fungal hyphae. It is known that mycorrhizal roots have not only a frequently greater P inflow rates, but also to have a pathway of the phosphate uptake with significantly higher affinity for phosphate than non-mycorrhizal roots. Furthermore the hyphae AM have a lower threshold for uptake of P than that of non-colonized plant that would permit mycorrhizal plants to increase P concentrations (33). Indeed, it is well established that plant responses to AM colonization are usually positive when P is limiting (34). Our results support previous findings which indicated that applying phosphorus to soil generally reduces AMF development and consequently the mycorrhizal benefits (35, 36, 37 and 38). There is no significant differences in root:shoot ratios were found between the mycorrhizal and the nonmycorrhizal soybean plants grown in P-fertilized soil. The expand root system was maybe due to a higher P uptake. The shoot growth was enhanced by inoculation with AM as well as root growth (39). One of the main effect of AM colonization on coriander growth was a significant increase in leaf area (LA) of the autotrophs. The increase in LA indicates that łeaves of AM plants were thinner than those of their non-AM counterparts. The considerably higher rates of photosynthesis noticed in leaves of AM plants. In the current study suggests that the rate of photosynthesis, and hence the amount of C fixed per unit investment in photosynthetic machinery, was much greater in AM plants. This was also observed, in the latter stages of the experiment when the variation in the rate of photosynthesis between AM and Non-AM plants was decreased (40).

Table 6. Effects of mycorrhizal (AMF) and nonmycorrhizal (NAMF) on proportion (%) of various constituents of essential oil extracts from *C. sativum* grown in sterilized soil with or without phosphorus fertilizer.

Treatments		Constituent							
P (KH_2PO_4) (mg kg^{-1})	**AMF status**	**α-Pinene**	**β-Pinene**	**p-Cymene**	**δ-Linalool**	**Geraniol**	**t-Anethol**	**β-Elemene**	**β-Caryophyllene**
F0	Non-AMF	0.23±0.001c	0.32±0.02c	1.12±0.43b	24.21±0.50c	1.26±0.013c	0.01±0.21c	1.01±0.012b	1.13±0.04b
Without P	AMF	0.40±0.005c	0.48±0.02c	1.33±0.99b	56.55±0.60b	3.93±0.07b	8.18±0.26a	1.26±0.005a	1.32±0.06c
F1	Non-AMF	0.63±0.01b	0.62±0.00b	2.60±0.06b	44.42±0.17c	1.57±0.015c	0.02±0.25c	1.04±0.015b	1.89±0.05b
100	AMF	1.19±0.02a	0.71±0.005a	5.06±0.06a	61.72±0.52a	19.99±0.60a	9.76±0.29b	1.41±16ab	2.17±0.14a

Values within a row followed by the same letter(s) are not significantly different at $p \leq 0.05$ (Duncan's multiple range test). Each value represents the mean of three replicates ± SE. AMF – arbuscular mycorrhizal fungi, Non-AMF – nonmycorrhizal fungi.

Phosphorus and N concentrations in shoots and roots of mycorrhizal and nonmycorrhizal coriander plants increased significantly with phosphate addition to soil. However, mycorrhizal plants had higher contents of P in both roots and shoots than nonmycorrhizal plants, particularly in the unamended soil. Our results indicated that the plants inoculated with mycorrhizal enhanced plant phosphorus and nitrogen N uptake, mainly under low P conditions, which led to increase both shoot and root biomass. The AM effect on nutrient uptake may be due to the well-known ability of the external mycelium to extend the soil volume that the plants are able to explore for P (or N) uptake (12). (41) and (42) found that colonization of plant roots by AMF greatly increase the plant uptake of P and N. The most prominent contribution of these fungi was increased uptake of nutrient by extraradical mycorrhizal hyphae. Many tested fungal isolates increased P and N uptake of the plants by absorbing phosphate, ammonium, and nitrate from soil. The contribution of AM fungi to plant P uptake, however, was in general much greater than the contribution of plant N uptake.

Mycorrhizal inoculation of plants increased the photosynthetic rate and stomatal conductance as compared to the non-inoculated plants grown in soil either amended or unamended with phosphorus. Such increases were related to the degree of mycorrhizal colonization. It is clear that the chlorophyll content increased in the mycorrhizal treated plants there is a simultaneous increase in *A* that can be due to more absorption of mineral and increase in gaseous exchange by enhanced conductivity of leaf stomata. Without P, the value of leaf chlorophyll was lower as well as *A* and *gs*. Conversely, (43) stated that the application phosphorus built up a stress in the plants and declines the normal physiological function, while the AM has been shown to effectively enhance the stomatal conductance, photosynthetic rate and water use efficiency of their host through serious stress conditions. It seems likely, in the presence of lower tissue phosphorus concentrations in non-AM compared to AM plants P was lower in non-AM plants, that the encouragement of photosynthetic rate was attributable to a non-nutritional impact of fungal colonization upon *C assimilation*. The importance of the cytosolic pool of inorganic P as a main determinant of the photosynthesis rate, but as it is complex to determine this pool with precision we have attempted, by exogenous phosphorus application, simply to obviously that phosphorus does not limit the photosynthesis rate in either AM or non-AM plants (44). The increase in photosynthesis noticed in the present study was similar to that observed by Johnson (45) and (46) in *Citrus aurantium* mycorrhized with *Glomus intraradices*, *G. mosseae* and *G. max*, in comparison with NM plants of similar N and P status. Also, they attributed the increase in *A* to increased sink strength caused by the presence of the mycobiont. In recent times, (47) observed that even at very low values of colonization by *G. mosseae*, enhancement of the greatest rate of photosynthesis of the youngest, fully expanded leaf of *Hordeum vulgare*, cultivated at a low P level supply, was noticed relative to that seen in non-AM plants of similar foliar P status. Furthermore, in the current study the mycorrhizal coriander plants produced more shoot and root dry weight than nonmycorrhizal plants (Table 1). The capability of AMF to increase leaf area and root intensity is harmonic with earlier studies (48, 49 and 50).

Reductions in AMF colonization levels in root tissues of coriander plants with increasing the amount of P to soils are consistent with previous field and greenhouse growth-room studies (51, 52 and 38). It was also observed in this study the beneficial effects of AMF on growth and biomass of coriander plants, more pronounced in the P-deficient soil treatment (Table 1). The effects are probable attributed to the enhancement of P and N nutrition (52), enhanced of gas exchange parameters and the increase of root length intensity (53).

Soil ACP and ALP activities were considerably higher in mycorrhizal than in nonmycorrhizal root extracts of the coriander plants grown either in soil with or without P. The present suggest that an AM fungus symbiosis contributed to the increase in the phosphatases activities. This might be a result of a direct contribution from the external mycelium and an indirect effect of improved host plant P status (54). So, AMF contributes to plant P nutrition without a considerable contribution from rhizospheric microorganisms. (55), (13) and (56) stated that the higher activities of phosphatases in the rhizosphere of plants inoculated with *G. mosseae* was contributed to by both AM and plant roots. The close relation between myc-

orrhizal growth responses and the intensity of mycorrhizal colonization support the hypothesis that phosphatases are involved in some way in the assimilation of P by AMF (57).

AM inoculated coriander plants had higher contents of protein in root extracts than nonmycorrhizal plants grown either in P amended or unamended soils. The observed increase in protein contents in response to mycorrhizal colonization was highly pronounced in the P-deficient soil treatment. These data are in agreement with the results of other findings (23, 52 and 58). In connection, total soluble protein of the mycorrhizal and nonmycorrhizal root extracts of coriander plants were generally reduced with increasing P in the soil. This result is in conformity with the obtained by (59) who stated that increased soil P inhibited AMP development and then decreased the beneficial effects of AMF symbiosis.

The pattern of difference in shoot-P concentration and concentration of essential oil in fresh fruits, plants with greater concentration of shoot-P possessed higher essential oil in their fruits. Fruits of coriander plants inoculated with *G. mosseae* contained more essential oil than those of non- inoculated and control plants. We have assiociate increase in essential oil concentration in AM plants of coriander to increased P-nutrition (6). AM fungi increase plant growth and essential oil production because mycorrhization allows the root system to exploit a greater volume of soil by (a) extending the root zone; (b) reaching smaller soil pores not accessible by root hairs; (c) acquiring organic phosphate through production of extracellular acid phosphatases (60). The quality enhancement of essential oil in coriander fruits on AMF inoculated plants may be due to increased nutrient uptake (especially P). Moreover, the enhanced yield in total essential oils in *G. mosseae*-treated coriander plants may be associated to the greater number of peltate glands, the structures responsible for oil production (61). These results are in agreement with the most previous studies (19, 20, 21, 7 and 61). However, knowledge about the roles of AM fungi on the production of essential oils is scanty, and only a less reports in a limited number of species have been published and the exact mechanism involved and the significance of the altered content of essential oil requires more investigations. Therefore, the present study promoters the utilization of AM fungi for improving the production and quality yield of essential oil in coriander fruits, thus making its planting further remunerative and eco-friendly.

Conclusions

Arbuscular mycorrhizal inoculation significantly improved growth of coriander plants grown in a sandy loam soil not only through increasing N and P contents, but also *via* stimulating gas exchange parameters and some metabolic contents of coriander plants particularly in P deficient soil. These benefits generally decreased when P was added to soil, suggesting that P fertilizer reduced AMF function and effects. Phosphorus application and AM fungus inoculation substantially improved P and N uptake by the plant. Furthermore, the quality enhancement of essential oil in coriander fruits on AM inoculation may be due to enhanced nutrient uptake (especially P). AMF inoculation increases the productivity and reduces the fertilizer application required to obtain economic production of coriander crop.

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