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Responce of snap bean plants *(phaseolus vulgare)* to mycorrhizal fungi under different levels of phosphate

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ABSTRACT

The effect of arbuscular mycorrhizal (AM) fungus and soil phosphorus levels (P) sole or combined application were studied on growth parameters, total chlorophyll, chemical concentrations, yield and quality as well as mycorrhizal dependency (MD) of snap bean grown at El-Baramoun Farm, Mansoura Horticulture Research Station, Dakahlia Governorate, Egypt, during two successive seasons of 2014 and 2015. A factorial design 2X3 experiment was designed and conducted to characterize the relationships between three soil phosphorus levels (0, 50% and 100% of recommended P fertilization) and two vesicular-arbuscular mycorrhizal (VAM) fungi treatments which were without mycorrhizae (NAMF) and with mycorrhizae (AMF). Mycorrhizal inoculation significantly increased all studied parameters compared with nonmycorrhizal plants. Addition of soluble phosphate improved all growth parameters, photosynthetic pigment, mineral composition (N, P and K), yield (yield/plant and early and total yield/feddan) and pod quality expressed as TSS, protein and fiber percentages.The interaction results between mycorrhizal bean and P amendment had higher growth records i.e., plant height, dry weight and leaf area, but no significant differences between 50% and 100%P were observed in both seasons. IN addition, 50%P was more superior in increasing total chlorophyll, chemical concentrations and yield component in AMF bean plants, except P concentration in the first season. In addition, pod characteristics was enhanced with increasing P level combined with AMF inoculation and the highest records of pod weight, length and diameter were obtained with inoculation with AMF and 50% of the recommended dose of P fertilizer. TSS and protein percentages of pods were significantly increased, whereas, fiber% was significantly decreased in mycorrhizal bean compared with nonmycorhizal one at the same levels of P. Mycorrhizal inoculation was more superior in improving pod quality of plants supplemented with 50%P.In general, growth, photosynthetic pigments, nutrition, yield and pod quality of snap bean plants showed a high degree of dependency on the mycorrhizal fungus in nonfertilized soil and 50%P when compared with the soil fertilized with100% P.The economic feasibility of snap bean cultivation shows that the highest net return and benefit-cost ratio (13353 LE fed^{-1} and 2.39, respectively) were obtained with mycorrhizal bean amended with half dose of phosphorus recommendation compared with other treatments under the condition of this study.This study confirmed that Bronco variety showed better vigorous growth of plants with higher pod yield and its quality in response to bio-fertilizer application(arbuscular vesicular mycorrhizal fungi) with the two levels of P (50,100%) and 50%P was more superior under high pH soil conditions of Nile Delta soils, where P availability is low.

Keywords: *Phaseolus vulgaris,* Micorrhizal dependency, P, growth, fruit quality, yield, net return

INTRODUCTION

Legumes have the potential to support global protein production by partially replacing meat and dairy products in the human diet. Common bean (*Phaseolus vulgaris, L.)* is of important crop in the world and, in some countries, which is the primary source of proteins and carbohydrates in human diets (Broughton *et al*.,, 2003) Besids, leguminous crops can be self-sufficient for all or part of their nitrogen requirements, when their roots are nodulated with effective nitrogen fixing strains of rhizobia.Thereby, it plays a role in improving soil fertility via N^2 fixation (Ellafi *et al*., 2011). In Egypt, biomass production, yield and nutritional quality of snap bean pod are limited by many factors including the fertility of the soil where it is planted thus, it is necessary to carry out agronomic practices such as application of fertilizer and biofertilizer. Phosphorus presents mainly as a structural component of the DNA, RNA, phospholipids and ATP, which is of immediate use in all processes that require energy with the cells. P can also be used to modify the activity of various enzymes by phosphorylation and is used for cell signaling. Although it is critical for plant growth and makes up about 0.2% of dry weight, it is the most sensitive nutrient to soil pH (Malakooti, 2000). The external P requirement of plants is the amount of available P in the soil required to obtain maximum growth. Arpana *et. al., (*2002) reported that a great proportion of phosphorus in chemical fertilizer becomes unavailable to the plants after its application in the soil. They referred this to formation of strong bonds between phosphorous with Ca^{+2} and Mg^{+2} in alkaline pH, likewise the Delta and Nile Valley soils of Egypt, and the same bonds with Fe and Al in acidic soils. In addition, the mobility of this element is very slow in the soil and cannot respond to rapid uptake by plants. This causes the creation and development of phosphorus depleted zones near the contact area of roots and soil in rhizosphere. Thus, the plants need an assisting system beyond the depletion zones and help to absorb the P from wider area by developing an extended network around root system (Salehrastin, 1999).Therefore, large amounts of phosphate fertilizers are used to improve growth and yield of crops. However, increasing costs of these fertilizers and environmental concerns related to their use have led to the development of alternative strategies. The most common of these strategies worldwide is AM symbiosis, since the use of beneficial soil microorganisms could reduce the amount of fertilizer input by increasing the efficiency of nutrient availability and other plant growth promoting activities. Biofertilizers hold a promise to balance many drawbacks of the conventional chemical based technology and could recuperate healthy farming practices and organic farming. Increased growth and yield of plants in the presence of AM fungi (AMF) has been attributed mainly to the enhanced uptake of P

(Smith and Jakobsen, 2004; Smith *et. al*., 2011 Abdel-Fattah and Asrar, 2012 Soha and Rabie**,** 2014; Hussain *et.al*.,2015). In addition, AM fungi produce glomalin, which is considered stably gluing hyphae to soil and lead to the stability of aggregates (Rillig *et al*., 2002).

The plant results higher response with inoculation of mycorrhiza because of more strong relationship with mycorrhiza to fulfill their phosphorus and other nutrients requirement and shared some photosynthates with fungus to maintain the symbiosis. In other word, improving plant acquisition of phosphate by AMF depends on the particular plant-fungus combination, symbiotic phosphate uptake may partially participate or dominate over all phosphate acquisition (Smith *et al*., 2003). Based on plant ability to grow with or without mycorrhizae at different levels of nutrients, plants can be separated into two major groups: nonmycotrophic and mycotrophic. Mycotrophic plants are also classified according to their degree of dependence on the mycorrhizae from obligatory to facultative (Ortas and Akpinar, 2006).Plants differ in P-uptake efficiency, which depends mainly on the morphology of root system (Foshe *et al*., 1991), the cation: anion uptake ratio (Bekele *et al*.,1983) and other physiological properties such as root exudations (Dinkelaker *et al*., 1989). Root morphology is also directly related to the mycorrhizal dependency of plants (Plenchette and Moral,1996) and consequently, mycorrhizae increase the P-uptake efficiency according to their mycorrhizal dependency. In addition to highly reduced mobility of phosphate in the soil, rapid phosphate uptake into the root leads to development of a phosphate depletion zone, causing a new pool of soluble phosphate (Abdel-Fattah 1997). Whereas, in nonmycorrhizal roots, the phosphate depletion zone greatly exceeds the root hair cylinder.This indicates that phosphate is not directly available to the plant. In fact, the external hyphae of AMF can absorb phosphate beyond the depletion zones around the root hairs and transport it to the root tissues (Smith and Gianinazzi-Pearson 1988, Abdel-Fattah 1997, Wu *et.al*., 2010). Phosphate fertilizers are routinely used in intensive agricultural practices to maintain soil P fertility level. Thus, phosphate fertilizers make a significant contribution to current global food production and security. However, residual P from repeated applications of high rates of mineral P to soil increases soil P saturation, which in turn inhibits AMF development (Smith *et al*., 2011*)*.

MATERIALS AND METHODS

Tow field experiments were conducted at El-Baramoun Farm, Mansoura Horticulture Research Station during two successive seasons of 2014 and 2015 to study the response of snap bean plants (*Phasolus vulgaris*) cv. Bronco to vesicular-arbuscular mycorrhiza (VAM) fungi under different levels of phosphorus. Soil samples to 25 cm depth were taken before planting to determine the physical and chemical analyses according to the method of Piper (1950) (Table1). Bean seeds were sown on 7th September in both seasons in 5cm apart on one side of ridge. The experimental unit area was 7.35m^2 and it contains three ridges with 3.5m in length and 70cm in width.

Table (1):Some physical and chemical characteristics of experimental soil in 2014 and 2015 seasons.

Soil characteristics	2014	2015
Physical analysis		
Coarse sand %	1.90	1.92
Fine sand %	17.88	17.68
Silt %	28.17	29.22
Clay %	50.02	51.45
Texture class	Clayey	Clayey
Chemical analysis		
Available N (ppm)	44.3	45.5
Available P (ppm)	14.7	16.0
Available K (ppm)	312.0	295.0
Total soluble solids%	0.19	0.18
pH*	7.9	8.1
EC** (dS/m)	1.2	1.1
Field capacity	34	35
Organic matter %	2.02	1.83
CaCO3 %	2.83	2.66

*** 1 : 2.5 (Soil : Water) water suspens**
**** 1 : 5 (Soil : Water) water extract.**

Experimental design:

The experiment with 2 × 3 factorial design in a complete randomized block design with three replicates was used in both growing seasons. The experimental treatments consisted of two AMF treatments *;* inoculated (AMF) and noninoculated (NAMF) plants both grown either with added phosphate at 15.5, 31 kg P_2O_5 /fed or without P. Phosphate was thoroughly mixed with the soil during planting. All cultural practices were performed as recommended.

Three different levels of P i.e., 0%, 50% and 100% of the recommended dose (200kg super phosphate calcium/ feddan)) were thoroughly mixed with the soil before planting as calcium superphosphate [$Ca(H_2PO_4)_2$(15.5% P_2O_5)].

AMF inoculum preparation:

The AMF inoculum, consisting spores, soil, hyphae, and infected root fragments of Sudan grass plants from a stock culture of mixture of *Glomus mosseae* (Nicol. & Gerd.) Gerd. & Trappe, *Glomus intraradices* (Schenck & Smith), *Glomus clarum* (Nicol. & Schenck), *Gigaspora gigantean* (Nicol. & Gerd.) Gerd. & Trappe, and *Gigaspora margarita* (Becker & Hall), was provided from Plant Pathology Ins., Agric. Res. Center, Cairo, Egypt. The AMF inoculums consisting of 20 g of rhizosphere soil (approx. 950 spores) and 0.5 g of infected root fragments. Mycorrhizal inoculation was done by planting the seed over a thin layer of the mycorrhizal inoculum (3cm-depth) at the time of sowing before sunrise to avoid the inhibition effect of direct light on mycorrhizal spores. The NAMF-treated plants were supplied with filtered washings of an equal amount of the mycorrhizal soil inoculum to provide the same associated microorganisms other than mycorrhizal propagules.

Data recorded:

1- Plant growth:

Three plants from each treatment were randomly taken at 55 days after sowing and the following data were recorded: plant height, leaf area per plant and dry weight of whole plant.

2- Photosynthetic Pigments and mineral analysis of leaves:

At 55 days from snap bean sowing, total chlorophyll in leaves was determined as described by Wettstein (1957). Also N% in leaves was determined in the dry matter using the micro keldahl apparatus according to Cotteni *et al*., (1982). Leaves P% was determined calorimetrically according to Sandell (1950) and potassium was determined according to Horneck and Hanson (1998).

3- Total green pod yield and number of pods/plant:

Number of pods\plant (random mean of 10 plants from each plot) was detrmined. Total yield and early yield (the sum of the first two pickings) as kg/plot were recorded, then calculated as ton/fed.

4- Green pod characteristics and quality:

Twenty pods were taken randomly from each replicate to determine average pod diameter, average pod length and average pod weight. Representative samples from green pods from each experimental plot were taken randomly to determine TSS by a hand refract meter. Total nitrogen content was determined in the dry weight of pods using the micro keldahl apparatus according to Cotteni *et al*., (1982). A factor of 6.25 was used for conversion for total nitrogen to protein in pods. Fibers content according to the method described in A.O.A.C.(1990).

5-Mycorhizal Dependency (MD):-

Mycorrhizal dependency (MD) or AMF growth responses (AMR) was defined as the degree to which a plant species is dependent on the mycorrhizal condition to produce its maximum growth at a given level of soil fertility (Shibata and Yano, 2003) .This definition is most pronounced for P requirement and calculated for every studied parameter using the formula of Menge *et al*., (1978) and modified by Son and Smith (1988) as follows equation:

$$\text{MD }\% = (\text{DMm} - \text{DMnm})/\text{DMnm} \times 100$$

where DMm represents the dry mass of mycorrhizal plants and DMnm the dry mass of nonmycorrhizal plants.

6- Economic Performance:

Economic performance of snap bean plants *i.e*., gross return, treatment cost, total variable cost, net return and benefit-cost ratio were calculated based on market prices as average of the two seasons. The benefit-cost ratio was determined according to Boardman *et al*., (2001) by dividing the net return (LE/ fed) on total variable cost (LE/ fed).

Correlation analysis and Statistical analysis:

Correlation between green pod yield and either dry weight of plant, leaf chlorophyll content or leaf P% were also analyzed and data were statistically subjected to analysis of variance (ANOVA) and the means were compared using the Least Significant Difference test (L.S.D.) at 5% level according to CoState (Version 6.303, CoHort, USA, 1998-2004).

RESULTS AND DISCUSSION

1-The mean performance of growth characters:

AMF inoculation improved growth parameters (plant height, dry weight and leaf area) in both seasons of this study (Table 2). However, addition of P to the soil significantly increased all growth parameters, particularly at 100%P. No significant differences between 50% and 100%P in plant height and dry weight were observed in the second season. Concerning with the interaction between AMF inoculation and P levels, data in the same table reveal that addition of P stimulate growth parameter of both AMF and NAMF bean plants. No significant differences between 50% and 100%P in growth parameters of AMF plants in both seasons were observed, except dry weight in the second season and leaf area in both seasons where100%P treatment was more announced in AM bean compared with 50%P treatment

Table (2): Effect of phosphate fertilization levels and mycorrhizal inoculation on growth parameters of snap bean during 2014 and 2015 seasons.

Treatments		Plant height (cm)			Dry wt./plant (gm)			Leaf area/plant (m^2)		
		1st S	2nd S	mean	1st S	2nd S	mean	1st S	2nd S	mean
AMF status										
AMF status	NAMF	36.83	39.56	38.195	12.90	15.04	13.970	0.196	0.199	0.198
	AMF	42.17	42.99	42.580	15.21	17.47	16.340	0.219	0.226	0.223
LSD 5%		0.56	0.57	0.565	0.45	0.71	0.580	0.004	0.003	0.004
P levels										
	P0	35.92	39.08	37.500	10.21	12.18	11.195	0.148	0.184	0.166
P levels	P1	41.26	42.07	41.665	14.86	17.98	16.420	0.219	0.202	0.211
	P2	43.33	42.67	43.000	17.09	18.63	17.860	0.256	0.251	0.254
LSD 5%		0.68	0.70	0.690	0.55	0.87	0.710	0.004	0.004	0.004
Interaction										
	P0	33.23	36.83	35.030	9.59	11.10	10.345	0.140	0.157	0.149
NAMF	P1	38.67	39.83	39.250	12.39	16.37	14.38	0.169	0.171	0.170
	P2	38.60	42.00	40.300	16.72	17.67	17.195	0.238	0.231	0.235
	P0	38.60	41.33	39.965	10.83	13.27	12.050	0.185	0.212	0.199
AMF	P1	43.84	44.30	44.070	17.33	20.90	19.115	0.230	0.235	0.233
	P2	44.07	43.33	43.700	17.47	18.30	17.885	0.273	0.270	0.272
LSD 5%		0.97	0.99	0.980	0.78	1.23	1.005	0.006	0.006	0.006

AMF: arbuscular mycorrhizal fungi, NAMF: nonmycorrhizal fungi; P0: without P; P1 : 50% P ; P2: 100% P.

The increases in growth parameters (plant height, dry weight and leaf area) of mycorrhizal plants were directly proportional to the respective level of the mycorrhizal colonization (Abdel-Fattah *et. al.,* 2014). The same pattern of response to the mycorrhizal infection in low P soils is entirely consistent with previous studies (Smith *et al*. 2011, Abdel-Fattah *et al*., 2014). However, addition of soluble P to soil can certainly alter characteristics of root colonization (particularly reducing arbuscule development) and markedly decrease AM fungal biomass per plant biomass (Smith *et al*.,2011) and appressorium (P entry points) formation (Balzergue *et al*., (2011) and consequently reduced the mycorrhizal benefits (Smith and Gianinazzi-Pearson, 1988).

2- Photosynthetic pigments and nutrient contents:

Generally, the presence of AMF significantly increased total chlorophyll and mineral composition (N, P and K) (Table 3). However, these parameters increased significantly with P addition to the soil, especially with 100%P. Concerning the interaction between AMF inoculation and P levels, data in the same table show that chemical composition (N, P and K) was improved in AMF bean plants compared with those of NAMF plants, particularly in 50% and 100%P treatments. IN addition, 50%P was more superior in increasing total chlorophyll content and chemical concentrations in AMF bean plants, except P concentration in the first season.

Table (3): Effect of phosphate fertilization levels and mycorrhizal inoculation on photosynthetic pigments and chemical composition of snap bean during 2014 and 2015 seasons.

Treatments		Total chl. (mg/g fw)			N%			P%			K%		
		1st S	2nd S	mean	1st S	2nd S	mean	1st S	2nd S	mean	1st S	2nd S	mean
AMF status													
AMF	NAMF	9.10	8.96	9.030	3.49	3.62	3.555	0.301	0.306	0.304	2.62	2.89	2.755
status	AMF	9.57	9.37	9.470	3.95	4.25	4.100	0.360	0.396	0.378	2.98	3.32	3.150
LSD 5%		0.025	0.053	0.039	0.04	0.02	0.030	0.009	0.016	0.0125	0.02	0.02	0.020
P levels													
	P0	8.95	8.86	8.905	3.40	3.54	3.470	0.272	0.283	0.278	2.71	2.92	2.815
P levels	P1	9.38	9.21	9.295	3.80	3.97	3.885	0.330	0.354	0.342	2.76	3.17	2.965
	P2	9.69	9.33	9.510	3.96	4.31	4.135	0.390	0.415	0.403	2.92	3.23	3.075
LSD 5%		0.030	0.065	0.048	0.05	0.02	0.035	0.011	0.020	0.016	0.03	0.03	0.030
Interaction													
	P0	8.64	8.72	8.68	3.05	3.10	3.075	0.227	0.240	0.234	2.46	2.61	2.535
NAMF	P1	8.94	8.93	8.935	3.53	3.52	3.525	0.290	0.330	0.310	2.56	2.96	2.760
	P2	9.73	9.24	9.485	3.89	4.25	4.070	0.387	0.407	0.397	2.83	3.10	2.965
	P0	9.25	9	9.125	3.76	3.97	3.865	0.317	0.327	0.322	2.96	3.22	3.090
AMF	P1	9.81	9.73	9.77	4.07	4.42	4.245	0.370	0.435	0.403	2.95	3.38	3.165
	P2	9.65	9.39	9.52	4.03	4.36	4.195	0.393	0.423	0.408	3.01	3.36	3.185
LSD 5%		0.043	0.092	0.068	0.07	0.03	0.050	0.016	0.028	0.022	0.04	0.04	0.040

AMF: arbuscular mycorrhizal fungi; NAMF: nonmycorrhizal fungi; P0: without P; P1: 50% P; P2: 100% P

Increased photosynthetic pigments in AM bean leaves is expected as a reflection to increase vegetative growth, especially leaf area (Table 2), mineral content that related to chlorophyll metabolism and presence of large number of chloroplast bundle sheath in the leaves (Krishna and Bagyaraj, 1984). The mycorrhizal plants had higher contents of N, P and K in shoots than those in the nonmycorrhizal plants. Such increases in nutrient concentrations in response to the mycorrhizal effects were highly associated with the level of the mycorrhizal infection (Abdel-Fattah, 1997). Moreover, AMF structure may play very important role in minerals uptake, since it can extend their external hyphae from root surfaces to areas of soil beyond the depletion zone, thereby exploring a greater volume of the soil than is accessible to the unaided root. The external hyphae of some AMF may spread 10–12 cm from the root surface. Assuming a radial distribution of hyphae around roots, it has been estimated that the volume of soil explored by the mycorrhizal root exceeds that explored by the unaided root by as much as 100 times (Sieverding, E. 1991). Also, AM fungal hyphae are 2.5–5 times smaller in diameter than plant roots and therefore have a greater surface area per unit volume. This surface area makes the fungi much more efficient than roots in the uptake of P (Bolan, 1991).

Moreover, the smaller diameter of AMF hyphae allows them to explore micropores in the soil that are not accessible to roots.AM fungi may have biochemical and physiological capabilities for increasing the supply of available P or other immobile nutrients. These mechanisms may involve acidification of the rhizosphere (Bago and Azcon-Aguilar, 1997), increases in root `

As mentioned, the most prominent effect of AMF is to improve P nutrition of the host plant in soils with low P levels due to the large surface area of their hyphae and their high affinity P uptake mechanisms. To substantiate this concept of plant growth promotion by AMF, several studies have shown that AM fungi contribute up to 90% of plant P demand (Van der Heijden *et al*., 2006). For instance, the P depletion zone around a non-mycorrhizal roots extends to only 1-2 mm, nearly the length of a root hair whereas extra radical hyphae of AMF extends 8 cm or more beyond the root making the P in this greater volume of soil available to the host plant.

3- Number of pods/plant, total green pod yield, and yield correlation analysis:

Data in Table 4 indicate that AMF inoculation significantly increased bean yield expressed as number of green pods per plant, early yield of green pods (ton/fed) and total yield of green pods per feddan. P supplement to the soil also enhancing these characters and 100%P was more superior in this respect. Regarding to the interaction between AMF inoculation and P levels, data in the same table show that all yield parameters were improved in AMF bean plants compared with those of NAMF plants under the same levels of P in both seasons. IN addition 50%P was more superior in increasing all yield parameters in AMF bean plants, except number of pods/plant where the difference between AFM beans at 50% and100%P did not reach to the significance at 5% level in both seasons.

It could be concluded that, these results may be attributed to the effect of AM fungi particularly in P1 soil on promoting vegetative growth characters (Table 2) and photosynthetic pigments and chemical constituents (Table3). In other meaning it can be concluded that, the final result of all physiological processes including vegetative growth, total chlorophyll and nutrient uptake will be reflecting on yield and it is obviously clear from the figures (1, 2 & 3),which showed highest positive correlation between some vegetative characters *i.e.* dry weight, chlorophyll content and phosphorus concentration from one side in snap bean plant and the total pod yield to the other side. This results are in agreement with Abd El-Dayem; *et. al*.,(2015) and Tabassum *et al*.,(2013) who revealed that maximum increase in the number of bean green pods existed in case of AM inoculation. However a significant decrease was obtained in total yield of mycorrhizal bean amended with high P level compared with the lowest level and this may be due to low level of mycorrhizal infection and efficiency under high soluble P level (Abdel-Fatteh *et al*., 2014). Also, Weber,1992 stated that the application of AMF in soils has shown a tremendous improvement in growth and yields of diverse legumes. For instance, inoculation with AMF improved growth of chickpea (*Cicer arietinum, L.*) and doubled P uptake at low and intermediate levels of P. In common bean, tomato and pepper, El-Melegy (2001) revealed that the inoculation with AMF plus 50% P gave higher value of early yield/fed than AFM bean amended with 100% P and the enhancing total yield in AMF bean may be due to increasing the portion of flower producing fruits and total number of fruits/plant .

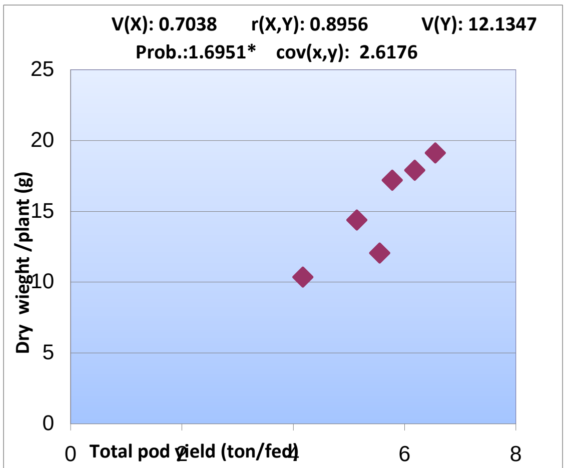
Table (4): Effect of phosphate fertilization levels and mycorrhizal inoculation on yield components of snap bean during 2014 and 2015 seasons.

Treatments		No. of pod /plant			Early yield of green pods(ton/fed)			Total green pod yield(ton/fed)		
		1st S	2nd S	mean	1st S	2nd S	mean	1st S	2nd S	mean
AMF status										
AMF status	NAMF	18	18	18	2.52	2.39	2.455	4.80	4.63	4.715
	AMF	20	19	19.5	3.14	3.06	3.100	6.26	6.15	6.205
LSD	5%	0.495	0.655	0.575	0.06	0.04	0.050	0.05	0.12	0.085
P levels										
P levels	P0	17	17	17	2.43	2.27	2.350	4.97	4.91	4.940
	P1	19	19	19	2.77	2.82	2.795	5.49	5.43	5.460
	P2	20	20	20	3.28	3.09	3.185	6.13	5.83	5.980
LSD	5%	0.606	0.802	0.704	0.07	0.05	0.060	0.06	0.14	0.100
Interaction										
NAMF	P0	17	16	16.5	1.30	1.31	1.305	4.67	4.83	4.750
	P1	19	18	18.5	1.71	1.70	1.705	5.12	5.17	5.145
	P2	20	20	20	1.85	1.77	1.810	6.06	5.50	5.780
AMF	P0	18	17	17.5	1.53	1.63	1.580	5.56	5.59	5.557
	P1	21	20	20.5	2.26	2.31	2.285	6.51	6.60	6.555
	P2	20	21	20.5	2.00	1.96	1.980	6.20	6.17	6.185
LSD	5%	0.858	1.135	0.997	0.10	0.06	0.081	0.08	0.20	0.140

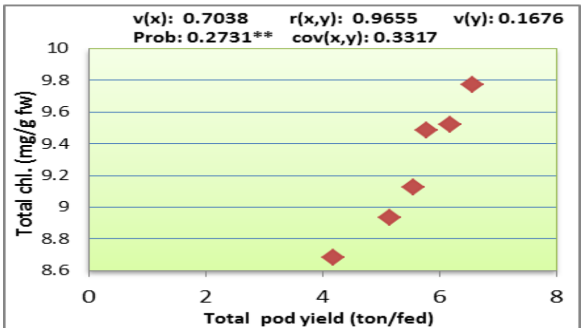
AMF: arbuscular mycorrhizal fungi; NAMF : nonmycorrhizal fungi; P0: without P; P1: 50% P; P2: 100% P; fed: feddan ($4200m^2$).

4- Pod characteristics:

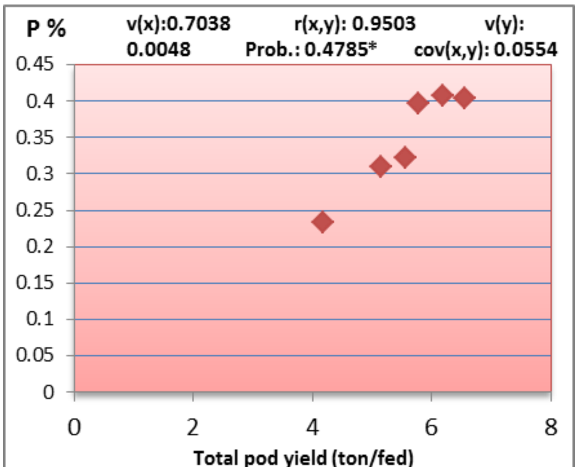
Concerning with pod characteristics, data in Table 5 show that pod characters i.e., weight, length and diameter of the inoculated bean were higher than that of noninoculated one. Also, with increasing level of P soil up to100% of the recommended dose, all pod characters were significantly increased. However, data of the interaction between AMF and P levels clear that mycorrhizal bean had the higher weight, length and diameter of pods compared with the NAMF bean under the same level of P soil. AMF bean supplemented with 50% P almost gave the higher records, except pod length and pod diameter in the first season, where 100% P was more announced in this respect. Similar findings have been reported by Tabassum *et al*.,(2013) concerning with pod length and Abd El-Dayem; *et. al*., (2015) concerning with pod length and diameter



Fig(1):Correlation between green pod yield and dry wieght of plant.



Fig(2): Correlation between green pod yield and total chlorophyll of leaves



Fig(3):Correlation between pod yield and leaf P%

5-Pod quality:

Table 6 indicates that, the presence of AMF significantly increased TSS and protein percentages and decreased fiber% in bean pods compared with those of NAMF plants. On the other hand, increasing P in the soil significantly increased the percentages of TSS and protein and decreased fiber percentage and P2 was more superior in this respect. Generally, pod quality was enhanced in AMF bean compared with NAMF bean amended with the same P level. No significant difference in protein percentage of pod between mycorrhized bean supplemented with 100% and 50%P was observed. However, the highest TSS and protein percentages and the lowest fiber % were obtained with AM bean in 50%P soil, except for TSS% in the first season which was more superior with AMF bean in100% P soil.

Table (5): **Effect of phosphate fertilization levels and mycorrhizal inoculation on snap bean green pod characteristics during 2014 and 2015 seasons.**

Treatments		Pod weight (g)			Pod length (cm)			Pod diameter (mm)		
		1st S	2nd S	mean	1st S	2nd S	mean	1st S	2nd S	mean
AMF status										
AMF status	NAMF	4.12	4.14	4.13	12.32	12.85	12.585	8.12	8.07	8.095
	AMF	4.23	4.25	4.24	12.75	12.34	12.545	8.30	8.25	8.275
LSD5%		0.018	0.012	0.015	0.085	0.030	0.058	0.026	0.077	0.052
P levels										
	P0	4.06	4.11	4.09	12.01	12.05	12.030	8.08	8.03	8.055
P levels	P1	4.18	4.20	4.19	12.67	12.74	12.705	8.23	8.22	8.225
	P2	4.28	4.38	4.33	12.94	12.98	12.960	8.31	8.25	8.28
LSD5%		0.022	0.015	0.019	0.104	0.037	0.071	0.031	0.090	0.061
Interaction										
	P0	4.02	4.05	4.035	11.73	11.70	11.715	8.02	7.93	7.975
NAMF	P1	4.08	4.11	4.10	12.43	12.35	12.390	8.13	8.13	8.13
	P2	4.27	4.28	4.28	12.80	12.96	12.880	8.21	8.17	8.19
	P0	4.10	4.17	4.14	12.28	12.40	12.340	8.41	8.13	8.27
AMF	P1	4.28	4.30	4.29	12.90	13.13	13.015	8.33	8.30	8.315
	P2	4.29	4.29	4.29	13.08	13.00	13.040	8.41	8.33	8.37
LSD5%		0.031	0.021	0.026	0.147	0.052	0.100	0.044	0.133	0.089

AMF: arbuscular mycorrhizal fungi; NAMF : nonmycorrhizal fungi ; P0: without P; P1: 50% P; P2: 100% P.

Table (6): **Effect of phosphate fertilization levels and mycorrhizal inoculation on green pod quality of snap bean during 2014 and 2015 seasons.**

Treatments		TSS%			Protein%			Fibers%		
		1st S	2nd S	mean	1st S	2nd S	mean	1st S	2nd S	mean
AMF status										
AMF	NAMF	5.91	6.71	6.310	4.460	4.528	4.494	14.31	12.74	13.525
inoculation	AMF	6.54	7.39	6.965	5.172	5.353	5.263	10.66	9.42	10.040
LSD5%		0.03	0.03	0.030	0.244	0.045	0.145	0.02	0.01	0.015
P levels										
	P0	5.72	6.56	6.140	3.498	3.642	3.566	14.16	13.51	13.835
P levels	P1	6.31	7.00	6.655	5.123	5.197	5.160	12.22	10.92	11.570
	P2	6.64	7.60	7.120	5.830	5.983	5.682	11.06	8.83	9.945
LSD5%		0.03	0.03	0.030	0.299	0.055	0.177	0.03	0.01	0.020
Interaction										
	P0	5.33	6.21	5.770	3.220	3.253	3.237	16.31	15.42	15.865
NAMF	P1	5.91	6.33	6.120	4.337	4.360	4.349	14.50	13.51	14.005
	P2	6.48	7.58	7.030	5.830	5.870	5.850	12.11	9.30	10.705
	P0	6.11	6.9	6.505	3.777	4.030	3.904	12.01	11.60	11.805
AMF	P1	6.71	7.66	7.185	5.910	6.033	5.933	9.96	8.33	9.145
	P2	6.80	7.62	7.210	5.832	6.000	5.916	10.00	8.35	9.175
LSD5%		0.04	0.05	0.045	0.423	0.077	0.250	0.04	0.02	0.03

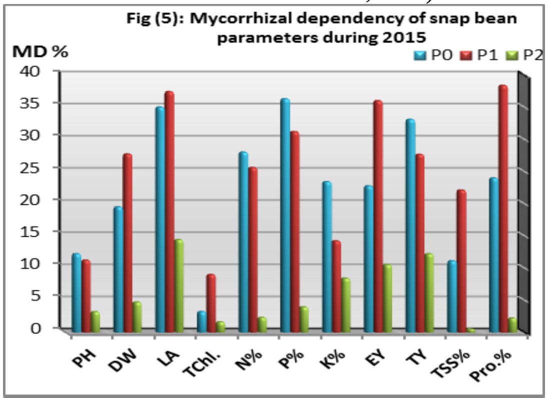
AMF: arbuscular mycorrhizal fungi; NAM : nonmycorrhizal fungi; P0: without P; P1: 50% P ; P2: 100% P.

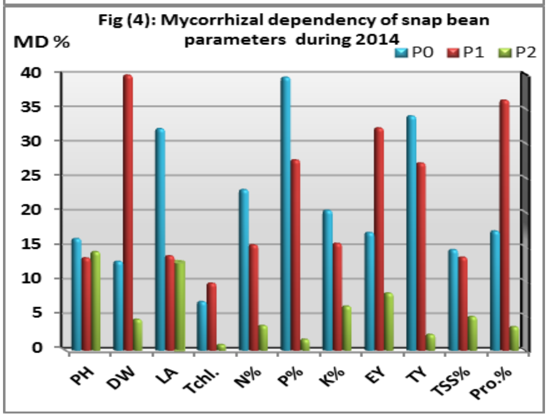
In this investigation, protein and TSS percentages may be increased because AMF inoculation supports photosynthesis process, by increasing photosynthetic pigment contents and mineral composition, particularly N% (Table 3). These results are in accordance with the results of Abd El-Dayem; *et. al*., (2015) and Hussain (2015). Therefore, integrated application of P with AMF can be highly recommended in common bean planting for improving quality of pods. Moin *et al.,* (2014) reported that there was a significant enhancement in seed protein content in common bean due to the application of adequate P and N biofertilizers (mixture of *Rhizobium spp* and *Glomus intraradices*).

6-Mycorrhizal Dependency:

Data in Table 7 and Fig.4 and 5 show that the enhancement in many parameters i.e., dry weight, chlorophyll A and B, early yield/fed, and protein% due to the AMF inoculation (MD) were more pronounced in the plants grown in 50%P, however, plant height, N% and K%, P% and total yield per feddan were stimulated in response to mycorrhizal inoculation in plants grown in P0 soil in both seasons of study. Meanwhile, leaf area and TSS% were differed in its response to mycorrhizal inoculation between P0 and P1 in both seasons. In all parameters under study, the lowest mycorrhizal dependency (MD) was obtained with plants grown in 100% P soil. Improving bean plant growth depends, significantly, on arbuscular mycorrhizal fungi which indicate that AM colonization can stimulate nutrient uptake which reflected on pod yield and quality.

Addition of soluble phosphate, in high rates, to soil significantly reduced the mycorrhizal growth response in associated snap bean plants as compared with the nonfertilized soil.This result supports the previous findings which indicated that adding P to soil generally reduced AMF development and consequently the mycorrhizal benefits (Smith and Gianinazzi-Pearson1988 and Abdel-Fattah *et al*., 2014).





MD: mycorrhizal dependency; P0: without P; P1: 50% P; P2: 100% P; PH: plant height; DW: dry weight; LA: leaf area; Tchl.: total chlorophyll; EY: early yield; TY: total yield; Pro.%: protein%.

Table (7): Mycorrhizal dependency (MD) of snap bean parameters during 2014 and 2015 seasons.

Parameters	P_0	P_1	P_2	LSD 5%
2014				
Plant height	16.20	13.4	14.3	1.3
Dry weight	12.9	39.9	4.5	0.36
Leaf area	32.14	13.7	12.8	1.4
Total chlorophyll	7.06	9.73	0.83	0.7
N%	23.3	15.3	3.6	0.2
P%	39.6	27.6	1.6	0.6
K%	20.3	15.5	6.4	0.6
Early Yield/fed	17.07	32.24	8.3	1.5
Total yield/fed	39.4	27.15	2.3	2.6
TSS%	14.6	13.5	4.9	1.4
Protein%	17.3	36.27	3.43	0.6
2015				
Plant height	12.2	11.2	3.2	0.7
Dry weight	19.5	27.7	4.7	1.2
Leaf area	35.0	37.4	14.4	0.7
Total chlorophyll	3.21	8.96	1.62	0.5
N%	28.0	25.6	2.3	0.4
P%	36.3	31.18	3.93	1.3
K%	23.4	14.2	8.4	1.2
Early Yield/fed	22.75	36.0	10.5	2.1
Total yield/fed	36.65	27.66	12.2	3.2
TSS%	11.1	22.1	0.5	0.3
Protein%	23.98	38.37	2.21	0.6

P0: without P; P1: 50% P; P2: 100% P; fed: feddan= 4200m²

According to the carbohydrate hypothesis, high concentrations of N and P promote root growth and hence protein synthesis in the plant, thereby decreasing the amount of available carbohydrates in the roots which is a pre-requisite for mycorrhizal formation, and as aresult reducing symbiotic association. Again,the level of phosphorus in the plant has been shown to influence the establishment of mycorrhizae with high levels inhibiting the density of AM fungi spores in soil and colonization by mycorrhizae (Menge *et al*., 1978).Very high and very low phosphorus levels could reduce mycorrhizal colonization (Koide, 1991). In other words, to increase mycorrhizal benefits, it is important to avoid excessive application of phosphorus fertilizers. Moreover, Ortas and Akpinar (2006) postulated that although plant growth was strongly affected by the P and Zn supply, and mycorrhizal inoculation increased P and Zn uptake, this was more strongly dependent on the P supply than Zn supply. Results obtained support the hypothesis that snap bean is mycorrhizal dependent, nevertheless with increasing P and Zn, the dependency is reduced.

Economic feasibility:

The economic feasibility of snap bean cultivation as affected by mycorrhizal treatment and application of different levels of phosphate are presented in Table 8. Mycorrhizal bean gained the highest records i.e., net return and benefit cost compared with non micorrhizal one. In addition, these results show that the highest net return and benefit-cost ratio (13353 LE fed^{-1} and 2.39, respectively) were obtained with mycorrhizal bean amended with half dose of phosphorus recommendation, followed by that amended with the recommended dose in comparison with the other treatments. Therefore, this treatment considered economical for snap bean production under the conditions of the present study.

Table (8): Economic feasibility of snap bean cultivation as affected by phosphate fertilization levels and mycorrhizal inoculation

Treat.		Total Yield (ton fed^{-1}) (1)	Gross return (£E fed^{-1}) (2)	Treat Cost (£Efed^{-1}) (3)	Total variable cost (£E fed^{-1}) (4)	Net return (£E fed^{-1}) (5)	Benefit cost ratio (6)	Order
	P0	4.75	16625	_	٨٧٩٠	٧٨٣٥	1.89	٦
NAMF	P1	5.145	18007	٣٠٠	٩٠٩٠	٨٩١٧	1.98	٥
	P2	5.780	20230	٥٥٠	٩٣٤٠	١٠٨٩٠	2.05	٣
	P0	5.557	19450	٥٠٠	٩٢٩٠	١٠١٦٠	2.09	٤
AMF	P1	6.555	22943	٨٠٠	٩٥٩٠	١٣٣٥٣	2.39	١
	P2	6.185	21647	١٠٥٠	٩٨٤٠	١١٨٠٧	2.20	٢

(1)Snap bean yield as average of two seasons, (2) Gross return as yield (ton fed^{-1}) x3500 £E ton^{-1}, (3) Treatment cost was calculated according to the following prices: Super phosphate calcium=50£E / 50 kg, Mycorrhizal inoculation = 500£E fed^{-1}, (4) Total variable cost (£E fed^{-1}): including Treatment cost plus land leasehold, seeds, labors and other agricultural practices, which equal nearly 8790 £E fed^{-1}. (5) = (2)-(4). (6)= (2)/ (4).

The results obtained here concluded that mycorrhizae could reduce the excessive amount of chemical fertilizers used in conventional agriculture practice which with a long time have adverse toxic effects on plant, environment and consequently human health. Again, mycorrhizal inoculation can be a suitable way (biofertilizer agents) to improve growth and yield of snap bean plants, particularly in poor soils and farmers should be aware of the beneficial effects of mycorrhizae as biofertilizer agents. Practical applications of these fungi are now possible, but these should bear in mind the factors affecting mycorrhizal development and function such as formulation, type of applications, viability of spores… etc. Further studies should be carried out in this field.

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استجابة نباتات الفاصوليا لفطريات الميكورهيزا تحت مستويات مختلفة من الفوسفور

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تمت دراسة تأثير التلقيح بفطر الميكورهيزا ومستويات من الامداد الأرضى بالفوسفور على صفات النمو الخضرى ،محتوى الأوراق من الكلوروفيل والعناصر المعدنيه ،محصول و جودةقرون الفاصوليا فضلا عن نسبة استجابة العائل للفطر وذلك بالمزرعه البحثيه بالبرامون التابعه لمحطة بحوث البساتين بالمنصوره خلال الموسمين الصيفيين ٢٠١٤ و٢٠١٥ و صممت لذلك تجربة عامليه ٢ X٣ تمثل التفاعل بين ثلاثة مستويات من الفوسفور(صفر، ٥٠% ، ١٠٠% من المعدل الموصى به للفدان) و معاملتين من فطر الميكورهيزا (بدون التلقيح بالميكورهيزا و التلقيح بالميكورهيزا). أظهرت النتائج أن الفاصوليا الملقحة بالميكورهيزا أعطت أفضل النتائج على مستوى جميع الصفات المدروسه مقارنة بالنباتات الغير معامله بالفطر. كما أوضحت النتائج أيضا أنه باضافة الصوره الذائبه من الفوسفور تتحسن جميع صفات النبات بداية بصفات النمو الخضرى ومرورا بمحتوى الأوراق من الكلوروفيل والعناصر المعدنيه وصولا الى المحصول من حيث الكم والجوده. فيما يخص معاملات التفاعل أعطت معاملات التفاعل بين التلقيح بالميكروهيزا وإضافة الفوسفور أعلى القيم للنمو الخضرى (إرتفاع النبات، المساحه الورقيه و الوزن الجاف للنبات) بينما لم تظهر فروقا معنويه فى صفات النمو الخضرى للنباتات الملقحه بالميكورهيزا بين المعامله ب٥٠% و ١٠٠% من المعدل الموصى به من الفوسفور. كما أوضحت النتائج أن النباتات الملقحه بالميكورهيزا ومسمده ب ٥٠% قد تفوقت فى محتوى الأوراق من الكلوروفيل ، النيتروجين ، الفوسفور والبوتاسيوم فضلا عن المحصول ومكوناته مقارنة بباقى المعاملات فيما عدا محتوي الأوراق من الفوسفورحيث كانت أعلى القيم مع الجرعه الكامله من السماد الفوسفاتى فى الموسم الأول. كما تحسنت صفات القرن من حيث الوزن و القطر والطول بزيادة مستوى الفوسفور اللأرضى وبالتلقيح بالفطركل على حده وكانت معاملة التفاعل بين الفطر و٥٠% P_2O_5 افضل المعاملات تأثيرا على صفات القرن فيما عدا طول وقطر القرن فى الموسم الأول حيث كانت المعامله بالفطر مع ١٠٠% ه P_2O الأكثر تأثيرا. أدى التلقيح بفطر الميكورهيزا الى زيادة معنويه فى نسبتى المواد الصلبه الكليه والبروتين وانخفاض نسبة الألياف بالقرون مقارنة بالنباتات الغير ملقحه عند نفس المستوى من الفوسفور وكانت أعلى النتائج المتحصل عليهافى صفات جودة القرون تلك الناتجه من المعامله بالفطر مع ٥٠% P_2O_5. فيما يختص بنسبة استجابة النبات للفطرأظهرت النتائج عامة أن جميع الصفات المدروسه (النمو، محتوى الأوراق من الكلوروفيل والمعادن ، المحصول وجودة القرون) كانت أعلى فى النباتات التى لم تسمد بالفوسفوروالمسمده بنصف المعدل الموصى به من الفوسفورللفدان مقارنة بتلك المسمده بالمعدل الموصى به. يتضح أيضا من دراسة الجدوى ان العائد الاقتصادى الناتج من االمعامله بالميكورهيزا مع نصف الجرعه الموصى بها من الفوسفور هى الأعلى من حيث صافى الربح ونسبة المنافع الى التكاليف (١٣٣٥٣ و 2.39 على التوالى) بالمقارنة مع المعاملات الأخرى وذلك تحت ظروف هذه الدراسه.تؤكد هذه الدراسه على امكانية الحصول على أقوى نمو خضرى مع أعلى محصول وجوده لقرون الفاصوليا صنف برونكو استجابة للتسميد الحيوى بفطر الميكورهيزا الداخليه مع التسميد الكيماوى بمعدل ١٠٠% و٥٠% من الفوسفور كما تؤكد الدراسه على أن التسميد بمعدل النصف كان الأكثر تفوقا فى ظروف ارتفاع pH التربه فى أراضى الدلتا ،حيث ينخفض تيسر الفوسفور.