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Effect of Inoculation With *Bradyrhizobium sp.*, Va-mycorhiza and Mineral Fertilizers on Seed Production and Quality of Pea

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THIS investigation was carried out at the experimental farm of Shandaweel, Agri. Res. Station, Sohag Governorate, Egypt, during 2012/2013 and 2013/2014 seasons, to study the effect of using *Bradyrrhizobium sp* and vesicular arbuscular mycorrhiza as an effective alternative for the chimical fertilizer, on the productivity of pea (*Pisum sativum L.*) seeds, Master B cultivar. The experimental designed as complete randomized block, with four replications.

Observations were made on days to 50% flowering, number of branches/ plant, stem length, number of pods/ plant, number of seeds/pod, 100 seeds weight, seed yield and seeds germination percentage. All the parameters were significantly influenced by inoculation with *Bradyrrhizobium sp* and VA-mycorrhiza compared to control in both seasons. The highest values of number of branches/ plant, 100-seeds weight, seed yield (kg/fed.) and seed germination% were obtained under the treatments of *Bradyrrhizobium sp* and VA-mycorrhiza plus 15, 30 kg NP/fed. While, the lowest values of these traits were obtained under control treatment, in both studied seasons.

Keywords: Pea- *Bradyrrhizobium sp*- VA-mycorrhiza.

Pea (*Pisum sativum L.*) is one of the most important favorable vegetable crops grown in Egypt, for both local consumption and exportation. Symbiosis is a biological phenomenon involving dynamic changes in the genome metabolism and signaling network Kawaguchi and Minamisawa (2010). Most of the legumes possess two types of microbial symbionts namely mycorrhizal fungi and nitrogen fixing bacteria thereby establishing triple association, capable of supplying N and P contents to the plants Silveira and Cardoso (2004). Both the mycorrhizal fungi and Rhizobium act as biofertilizer and have the unique ability to convert nutritionally important elements into available form through biological process (Hegde *et al.,* 1999 and Vessey, 2003). Therefore, it becomes essential to use the un-traditional fertilizers as a substitute or a supplement for chemical fertilizers. Hence, the symbiotic relation between higher plants and mycorrhizae, particularly vesicular-arbusular mycorrhizae (VAM) fungi represents one of the most striking biological phenomena. Moreover, the endomycorrhizal fungi produce a highly branched hyphal structure within the plant cell. This infection creates an absorptive structure with a very high surface area of transfer for nutrients between the plant and the fungus. Mycorrhizal fungi hyphae secrete

acid and alkaline phosphatases (APA and ALP) into the rhizosphere. It was established that APA activity increases in roots growing under P stress (Woolhouse, 1975). Therefore, the regulation of these enzymes is critical to a plant's survival in soils with limited P resources (Duff *et al.,* 1991). There is extensive evidence for a decrease in the number of arbuscules under high external P (Bethlenfalvay *et al*., 1990 and Smith & Smith 1996).

Phosphorus has a key role in the energy metabolism of all plant cells, and particularly in nitrogen fixation (Dilworth, 1974). It was established that nodulating legumes require more P than legumes growing on mineral nitrogen (Al-Niemi *et al.* 1997). Garden pea has relatively high requirement for phosphorus (Slinkard and Drew, 1988). Yield and its components can be enhanced by phosphorus fertilizer in soil testing low in phosphorus, Slinkard & Drew (1988) and Murat *et al*. (2009). In most soils, in spits of the considerable addition of phosphorus fertilizers, available phosphorus for plants is usually low since it converting to unavailable form by its reaction with the soil constituents. Soil in Egypt needs high amount of mineral phosphorus fertilization to fulfill requirements of plants. However, the increase in the rate of applied phosphorus fertilizer may be increase production costs and environmental pollution, Mahmoud and Abdel-Hafez (1982). In soybean (Bethlenfalvay *et al.,* 1990) and pea (Xavier and Germida, 2003) were reported the AM fungi associated with legumes are an essential link for effective phosphorus nutrition, leading to enhanced nitrogen fixation that in turn promotes root and mycorrhizal growth. Synergistic effect of dual colonization of roots with AM fungi and Rhizobium on growth, nutrient uptake and nitrogen fixation. The effectiveness of the tripartite symbiosis AM fungi, Rhizobium and plant, depends on the competition of the three symbionts for carbon (Jakobsen and Rosendahl, 1990). Roots with AM fungi receive about 4–20% more photosynthates than comparable non-mycorrhizal roots (Smith and Read, 1997). Jakobsen and Rosendahl (1990) estimated that AM fungi could use up to 20% of the total fixed $^{14}CO_2$ in young plants. Concomitant development of mature functional nodules and effective AM infection depended on microsymbiont species and strains (Saxena *et al*., 1997). The synchronization between the two symbiotic systems needs an optimal P level in the nutrient medium to stimulate the nodulation and nitrogen fixation and not to slow down the formation of effective mycorrhizal associates. Nitrogenous chemical fertilizers are commonly added to soil to produce high yield of vegetable crop. The chemical fertilizers containing ammonium salts, anhydrous ammonia or urea are generally nitrified rapidly. Consequently, many plants consume and assimilate much amounts of the formed nitrate and store it in large quantities (Hammad and Abdel-Ati, 1998). Nitrate ion is a well known environmental pollutant because of its potential role in infants methemoglobinemia associated with the consumption of nitrate- rich water or vegetables (Alexander, 1977).

The present investigation was designed as an attempt to get benefit from the important role of visicular arbuscular mycrrhizae (VAM), *Bradyrhizobium* and chemical fertilizer with phosphorus and nitrogen.

Material and Methods

The present study was carried out during the winter seesons of 2012/ 2013 and 2013/2014 at the experimental farm at Shandaweel Agriculture Research Station, Sohag Governorate, Egypt, to investigate the efficiency of incoculation with *Bradyrizobium sp.* and VA-mycorrhiza as a bio-fertilizer and an effective alternative for nitrogen and phosphorus chemical fertilizer on productivity of pea (*Pisum sativum L.*) seeds, Master B cultivar.

Bradyrizobium sp. was obtained from the microbiology Dept., Soil and Water Research Institute, Agric. Res. Center, Giza, Egypt. Seeds of pea were inoculated at the rate of 10 g Inoculat. / 200 g Seeds. Un-inoculated seeds were soaked in tap water for some time (about two hour before planting). Gumarabic solution was used an adhesive material at the rate of 3 ml/100 g Seeds.

VA–Mycorrhizal inoculants: Two species of endomycorrhizal fungi were used in this study (*Glomus fasiculatum* and *Glomus mosseas*). They were obtained frome the Microbiology Dept., Fac. Agric. Minia University, Egypt. For preparing VA-mycorrhizal inoculants, fried pots of 30 cm in diameter were filled by sterilized clay loam soil.

The soil of each plot was inoculated with the two species of endomycorrhizal fungi. Ten onion seed linges were transplanted in each plot as a host plant. At the end of the used pots was mixed together then VAM spores were counted as described Msondu and Giller (1994). The spore count was found to be 93-100 spores/g soil. This soil contained mixture of VAM spores, mycelia and chopped roots. The prepared VAM inocoulants were added at rate of 4 kg of soil per plot and drilled in the ridges just before the first irrigation. Each ridge received equal quantity frimVA- mycorrhizal inoculonts (1 kg/ ridge).

Nitrogen and phosphorus Rates

Four rates of ammonium sulphate (20.5% N) 30, 22.5, 15 and 7.5 N kg/fed. Plus four rates of calcium super phosphate (15% P_2O_5) 60, 45, 30 and 15 P kg./fed. The four rates present 1/4, 1/2, 3/4 and whole recommended dose according to Ministry of Agriculture. These doses were applied after 21 days from sewing date.

The experimental design was complete randomized block, with four replications. Each replicate contained one of the following treatments:

- Control (seed without inoculation and fertilization).
- Inoculation of seeds with *bradyrrhizobium sp*.
- Inoculcation with (VAM) fungi.

- Inoculation with *Bradyrrhizobium sp* and (VAM) fungi.
- The recommended nitrogen and phosphorus fertilization (30,60 NP kg.fed).
- The mixture of *Bradyrrhizobium sp* plus (VAM) fungi and 22.5,45 NP kg/fed.
- The mixture of *Bradyrrhizobium sp* plus (VAM) fungi and 15,30 NP kg/fed.
- The mixture of *Bradyrrhizobium sp* plus (VAM) fungi and 7.5, 15 NP kg/fed.
- Inoculation with *Bradyrrhizobium sp* and 22.5, 45 NP kg/fed.
- Inoculation with *Bradyrrhizobium sp* and15, 30 NP kg/fed.
- Inoculation with *Bradyrrhizobium sp* and 7.5, 15 NP kg/fed.
- Inoculation with (VAM) fungi and 22.5, 45 NP kg/fed.
- Inoculation with (VAM) fungi and 15, 30 NP kg/fed.
- Inoculation with (VAM) fungi and 7.5, 15 NP kg/fed.

Each experimental unit was 10.5 m^2 consisting 4 ridges at 75 cm apart and 3.5 m length, sowing was done in 3rd and 5th of October in the first and second seasons, respectively. Two seeds were sown per hill on the sides of ridge at 10 cm apart for Master B cultivar. Standard agricultural practices known for commerecial garden pea production other than the applied treatments were followed. Dry pods were harvested in two times. The physical and chemical characteristics of the experimental site were as shown in Table 1.

TABLE 1. Soil characterization for the experimental location.

Seasons	Texture	$CaCo_3$ %	Soil pH	Organic matter (O.M.%)	Available nutrients in soil (ppm)		
					N	P	K
2012-2013	Sandy loam	7.50	6.9	0.6	13	18	12
2013-2014	Sandy loam	8.70	7.3	0.9	25	14.7	31

Ten plants were randomly chosen from each plot to determine the following characteristics:

Vegetative growth characteristics

- Days to flowering: recorded as the number of days from sowing to 50% flowering.
- Number of the branches/plant.
- Stem height (cm), from cotyledon node to the top of the main stem.

Seed yield and quality

Harvesting of dry pods was carried out three times from four ridges per plot before dropping seeds and the following data were recorded:

- Number of dry pods/ plant.
- Number of dry seeds per pod
- 100 dry seed weight (g)
- Dry seeds yield (kg/fed.): total dry seeds yield per fed. (kg).
- Seed germination % : was performed in petri dishes, containing cotton moist with water in the thermo incubator at 24^{0}C for one week.

$$\text{Germination \%} = \frac{\text{Number of germination seeds}}{\text{Total number of sown seeds}} \times 100$$

- Root length (cm), final count was observed on 12 day then, 10 normal seedlings were selected randomly to measure the root length. The root length was measured from the tip of primary root to base of the hypocotyledon and the mean root length was expressed in centimeters.
- Shoot length (cm), the same ten normal seedlings which selected randomly for measurement of root length was used to determine the shoot length. The shoot length was measured from the base of primary leaf to the base of hypocotyledon and the mean shoot length was expressed in centimeters, Anon (1996).

Chemical composition of dry seeds

Random samples of seeds were taken after harvesting and dried at 70 $^{\circ}$C. The dried samples were ground and used to determine N and P for the first and second experiments.

Statistical analysis

Analysis of variance for randomized complete block design was done according to Gomez and Gomez (1984). Significance among means was tested by using LSD method (least significant deference) at 5% level.

Results and Discussions

Vegetative growth characteristics

Days to flowering

Data presented in Table 2 show the effect of the inoculation with *Bradyrhizobium sp.* and VA-Mycorhiza fungi and application of NP-chemical fertilizers on days to 50% flowering. The lowest values of days to flowering (44.33 and 40.00) were obtained with application of 15 & 30 NP kg/fed. to the plants which received dual inoculation (*Bradyrhizobium sp.* + VAM) in the first and second seasons, respectively. These results are in agreement with those found by Abd El-Ati *et al.* (2000), while the highest values (50.00 and 47.33) were obtained under control treatment in the first and second seasons, respectively.

Number of branches / plant

Data presented in Table 2 show the response of pea plants to application of NP-chemical fertilizers and the inoculation with *Bradyrhizobium sp* and VA-Mycorhiza fungi. Data indicated that application of NP-chemical fertilizers, inoculation with *Bradyrhizobium sp* and inoculation with VA-Mycorhiza fungi treatments alone or in different combinations overcome the control in respect to improving the number of branches/plant. Such effect was cleared in both seasons. The highest growth measurements (1.80 and 1.93) were obtained with application of 15 & 30 NP kg/fed. to the plants which received dual inocula (*Bradyrhizobium sp.* + VAM), in the first and second seasons, respectively. Similar results were obtained by Bagyaraj *et al.* (1979) and Nurat *et al.* (2009).

On the other hand, the lowest values were (1.20 and 1.27), in both seasons, respectively.

TABLE 2. Effect of inoculation with *Bradyrhizobium sp* VA-mycorhiza and mineral fertilizer on plant length (cm), number of days to flowering and number of branches/plant for pea at 2012/2013 and 2013/2014 seasons.

Treatments	Days to flowering		Number of branches/ plant		Stem height (cm)	
	2012/ 2013 season	2013 /2014 season	2012/ 2013 season	2013/ 2014 season	2012 / 2013 season	2013 / 2014 season
Un-inoculated	50.00	50.33	1.20	1.27	37.33	37.00
***Bradyrhizobium sp.* (B.)**	47.00	46.67	1.53	1.57	34.00	42.86
Mycorrhiza (M.)	47.33	45.33	1.53	1.60	42.87	43.80
(B.) + (M.)	45.33	45.00	1.57	1.63	43.70	42.00
30,60 NP kg / fed.	45.67	47.33	1.67	1.86	47.33	48.70
(B.)+(M.)+22.5, 45 NP kg/fed.	45.00	44.33	1.73	1.87	47.90	49.70
(B.) + (M.) + 15, 30 NP kg/fed	44.33	44.00	1.80	1.93	47.47	49.90
(B.) + (M.) + 7.5, 15 NP kg/fed	46.33	45.66	1.70	1.85	46.00	45.27
(B.) + 22.5, 45 NP kg/fed.	45.33	46.33	1.57	1.57	47.53	43.20
(B.) + 15, 30 NP kg/fed	46.33	45.00	1.53	1.70	43.73	49.03
(B.) + 7.5, 15 NP kg/fed	46.00	46.65	1.57	1.57	42.87	43.00
(M.) + 22.5, 45 NP kg/fed	45.33	46.00	1.50	1.67	46.03	43.00
(M.) + 15,30 NP kg/fed	45.67	45.33	1.67	1.78	44.73	43.60
(M.) 7.5,15 NP kg/fed	46.67	47.33	1.43	1.63	42.90	42.73
LSD at 0.05	**3.43**	**3.01**	**0.28**	**0.29**	**5.53**	**4.85**

Stem height (cm)

Data presented in Table 2 show the response of pea plants to application of NP-chemical fertilizers and the inoculation with *Bradyrhizobium sp.* and VA-Mycorhiza fungi on stem height (cm). It is cleared that application of 22.5& 45 NP kg/fed. to the plants which received dual inocula (*Bradyrhizobium sp.* +

VAM) gave the highest value of stem height (cm) in the first season. Whereas, in the second season, the highest values were recorded for plant which received dual inocula plus15&30 NP kg/fed. On the other hand, the lowest value of plant height (cm) was obtained by un-inoculated, in the two seasons. These results were in agreement with those obtained by Soddi *et al*. (1994). These results might be due to that applying bio-fertilizers increased microorganisms in the soil, which convert the ability of mobilizing the unavailable form of nutrients elements to available form, Ishac (1989).

Seed yield and quality

Number of dry pods/plant and number of seeds /pod

Data in Table 3 clear the effect of inoculation of pea plants with *Bradyrhizobium sp.* and VA-mycorhiza fungi plus application of NP-chemical fertilizers. Application of different doses of nitrogen and phosphorus to each individual inoculation or to the dual inoculation significantly increased number of dry pods/plant and number of seeds/pod as compared to control. Furthermore, application of dual inoculation plus 15 & 30 NP kg/fed. appeared the highest values of these traits and it surpassed the recommended dose of nitrogen and phosphorus *i.e.,* 30 & 60 NP kg/fed. in both seasons. These results are in harmony with those obtained by Abd El-Ati *et al.,* (2000), Yadegari *et al.* (2008) and Giri & Joshi (2010). These results may be due to the positive effect of dual inocula, *i.e., Bradyrhizobium sp*. plus VA-mycorrhiza in improving nutritional status and number of seeds per pod. Moreover, Douds and Naghashi (2000) reported that mycorrhizal fungi extracted strong chemical agents into the soil, therby, made elements like phosphorus, iron, zinc, copper and boron available.

100-dry seeds weight (g)

Data presented in Table 3 show the effect of inoculation of plants with *Bradyrhizobium sp.* and VA-mycorihza and supplying with different doses of nitrogen and phosphorus. Supplying of different doses of nitrogen and phosphor to dual inoculated significantly increased average of in both seasons. The highest values of 100-dry seeds weight (17.08 and 18.25g.) were recorded for plant which received dual inocula plus 15 & 30 NP kg/fed. These results are similar to those obtained by Abbot and Robson (1984) who reported that total mycorrhizal root surface was increased 30 time more than non mycorrhizal roots, Smith and Reed (1997) who reported that several micronutrients as Fe, Zn, Mn ,and Cu transferred through the VA- mycorrhizal haphae, and Fatima *et al.* (2010).

Dry seed yield (kg/fed.)

Results illustrated in Table 3 clearly show the effect of inoculation of pea seeds with *Bradyrhizobium sp.* and VA-mycorhiza fungi plus application of NP-chemical fertilizers on dry seed yield (kg/fed.). It could be concluded that increasing the ratio of N & P-chemical fertilizer up to 15 & 30 kg/fed. with the inoculation of pea seeds with *Bradyrhizobium sp.* and VA-mycorhiza fungi improved the productively of pea plants and produced the highest dry seed yield (770 and 780 kg.), in the first and second seasons, respectively. However, the

lowest values (250 and 280 kg.) were obtained without inoculation and fertilization (control). The effect of inoculation on dry seeds yield of pea plants was reported by many authors such as Nadia *et al.*, (2008) and Salih *et al.*, (2015). These result my be due to positive effect of VA-mycorhiza inocula in improving growth and yield components previously mentioned. Ross and Harper (1970) recorded 29% yield increased in soybean grown in fumigated field plots, due to double inoculation with endomycorrhiza plus *Bradyrhizobium* over single inoculation with only Rhizobium.

TABLE 3. Effect of icoculation with *Bradyrhizobium sp*. VA-mycorhiza and mineral fertilizer on number of dry pods/plant, number of seeds/pod 100-seed weight (g) and dry seeds yield (kg/fed.) for pea at 2012/2013 and 2013/2014 seasons.

Treatments	Number of dry pods/ plant		Number of dry seeds per pod		100 dry seeds weight		Dry seeds yield (kg/fed.)	
	2012/ 2013 season	2013/ 2014 season	2012 /2013 season	2013/ 2014 season	2012/ 2013 season	2013/ 2014 season	2012/ 2013 season	2013/ 2014 season
Un-inoculated	7.43	6.53	6.73	6.10	12.78	12.38	250	289
***Bradyrhizobium sp.* (B.)**	10.85	10.10	8.27	8.47	14.35	15.23	450	460
Mycorrhiza (M.)	10.00	10.40	8.83	8.37	15.16	14.83	441	456
(B.) + (M.)	11.20	12.10	8.55	9.03	14.50	15.53	560	609
30,60 NP kg / fed.	12.15	12.83	8.60	9.30	15.50	16.13	710	720
(B.)+(M.)+22.5,45 NP kg/fed.	13.50	12.90	9.70	9.63	16.13	18.13	730	750
(B.) + (M.) + 15,30 NP kg/fed	14.27	14.83	9.97	9.67	17.08	18.25	770	780
(B.) + (M.) + 7.5,15 NP kg/fed	10.55	11.9	8.90	9.34	15.23	16.03	631	640
(B.) + 22.5,45 NP kg/fed.	11.58	13.43	9.47	9.57	15.83	16.45	685	690
(B.) + 15,30 NP kg/fed	13.23	11.77	8.67	8.90	15.09	15.63	624	640
(B.) + 7.5,15 NP kg/fed	10.40	10.33	8.52	8.87	14.84	15.08	550	572
(M.) + 22.5,45 NP kg/fed	10.43	12.38	9.07	9.63	15.13	16.18	640	670
(M.) + 15,30 NP kg/fed	11.53	12.10	9.57	9.10	15.42	16.02	610	640
(M.) 7.5,15 NP kg/fed	10.36	10.90	8.37	8.33	14.50	14.91	550	564
LSD at 0.05	**2.51**	**3.21**	**1.35**	**1.46**	**1.14**	**1.91**	**23.23**	**32.45**

Seed germination %, root length and shoot length (cm)

Data in Table 4 indicated that inoculation of pea plants with VA-mycorrhiza fungi separately or combined with *Bradyrhizobium sp.* in the presence or absence of different doses of nitrogen and phosphorus chemical fertilizers dramatically increased the percentage of germination, root length and shoot length (cm) as compared to control treatment. These results held well in the two experimental seasons. The highest values of percentage of germination, root

length and shoot length were recorded for plants which received dual inocula plus 15 & 30 NP kg/fed. These results are in accordance with those reported by El-Shaikh and Mohmmed, (2009) who worked on okra plants and found that inoculation with VA-mycorrhiza fungi significantly increased the percentage of germination, Iraj *et al.,* (2009) who reported that inoculation soybean seeds with *B. japonicum* and *G. mosseae* increased primary stem length, and Lenin *et al.,* (2010) who reported that maximum increase in root length and shoot length was observed in AM fungi treated seedlings when compared to non-mycorrhizal seedlings (controlled).

TABLE 4. Effect of inoculation with *Bradyrhizobium sp*. VA-mycorhiza and mineral fertilizer on shoot length, seed germination% and root length for pea at 2012/2013 and 2013/2014 seasons.

Treatments	Seed germination%		Root length		Shoot length	
	2012/ 2013 season	2013/ 2014 season	2012/ 2013 season	2013 /2014 season	2012/ 2013 season	2013/ 2014 season
Un-inoculated	79.00	69.80	5.82	5.25	5.67	5.98
***Bradyrhizobium sp.* (B.)**	88.43	78.00	6.99	6.30	7.21	6.88
Mycorrhiza (M.)	87.30	74.50	6.85	6.23	7.24	6.85
(B.) + (M.)	91.00	78.37	7.23	6.58	7.32	6.70
30,60 NP kg / fed.	91.80	84.23	7.23	6.70	7.38	7.11
(B.)+(M.)+22.5, 45 NP kg/fed.	92.00	89.60	7.77	7.03	7.49	6.75
(B.) + (M.) + 15, 30 NP kg/fed	92.90	90.60	7.79	7.37	7.88	7.72
(B.) + (M.) + 7.5,15 NP kg/fed	90.13	83.90	7.11	6.45	7.30	7.00
(B.) + 22.5, 45 NP kg/fed.	89.10	80.00	7.47	6.33	7.25	7.11
(B.) + 15, 30 NP kg/fed	92.00	88.30	7.52	6.91	7.38	6.90
(B.) + 7.5, 15 NP kg/fed	88.80	79.10	7.10	6.33	7.24	6.88
(M.) + 22.5, 45 NP kg/fed	90.20	81.67	7.11	6.40	7.38	7.09
(M.) + 15, 30 NP kg/fed	92.00	88.00	7.33	6.45	7.28	6.90
(M.) 7.5, 15 NP kg/fed	88.00	78.70	7.09	6.30	7.23	6.67
LSD at 0.05	**5.95**	**4.51**	**0.60**	**0.61**	**0.46**	**0.53**

Chemical composition of dry seeds

Data in Table 5 indicated that the inoculation of pea with VA-mycorrhiza fungi separately or combined with *Bradyrhizobium sp.* in the presence or absence of different doses of chemical fertilizer dramatically increased the percentage of protein (N %) and phosphorus (P %) as compared to the un-inoculated ones.

It is clear from the obtained data that inoculation of pea plants with *Bradyrhizobium sp.* significantly increased the percentage of protein in dry seeds as compared to uninoculated ones in both seasons. The increase in the protein percentage in dry seeds pea inoculated with *Bradyrhizobium sp*. might be attributed to the promoting effect of Inoculation of legumes with BradyRhizobium increase the nodulation of legumes causing more nitrogen fixation and making it available for the plants and therefore, it is used as an alternative for urea to minimize the cost of produce Karim *et al.* (2001). Moreover, it also increases rhizospheric microflora viz. acid producers and phosphate solubilizers causing more available phosphorus Lipman and Conybeare, 1936. Rhizobium inoculation has been shown to improve the nitrogen and phosphorus contents of legumes Abd-Alla and Omar (2001). These results are in harmony with those obtained by Hegazy *et al*. (1990).

Results in Table 5 showed that inoculation with VA-mycorrhiza significantly increased percentage of protein and phosphorus in dry seeds as compared to un-inoculated plants. One of the most significant effects of mycorrhizal inoculation on the host plant is the increase in phosphorus uptake Bai *et al.* (2007) due to enhanced capacity to absorb more phosphorus from the soil which is otherwise unavailable to the plants. AM fungi supported nitrogen fixation by providing legumes with phosphorus and other immobile nutrients which are essential for nitrogen fixation Clark and Zeto, 2000.

Regarding to the effect of dual incula (i.e, *Bradyrhizobium sp.* Pluse VA-mycorrhiza) on the pre mentioned traits, data showed that dual incula significantly increased percentage of protein and phosphorus in dry seeds as compared to uninoculated plants. These results held well in both seasons. Similar finding were reported by Rahman *et al.* (2010).

It is clear from data in Table 5 that application of nitrogen and phosphorus chemical fertilizers to plants which inoculated with both *Bradyrhizobium sp*. and VA-mycorrhiza increased the previously mentioned characters. The differences were statistically approved in the two seasons. Data also reveal that the highest values of protein and phosphorus were recorded for plants which received dual inocula plus 15 & 30 NP kg/fed. Many reports are available on the increased nitrogen and phosphorus contents of legumes due to dual inoculation of AM fungi and Rhizobium which are supported by the findings of our present study (Soliman *et al*., 2012, Chakrabarty *et al.*, 2007 and Talaat & Abdallah, 2008).

TABLE 5. Effect of inoculation with *Bradyrhizobium sp* VA-mycorhiza and mineral fertilizer on percentage of protein and phosphorus in dry seeds in 2012/2013 and 2013/2014 seasons.

Treatments	Protein % in dry seeds		Phosphorus% in dry seeds	
	2012/2013 Season	2013/2014 season	2012/2013 season	2013/2014 season
Un-inoculated	18.15	16.00	0.45	0.39
Bradyrhizobium sp. (B.)	20.12	19.35	0.58	0.50
Mycorrhiza (M.)	20.55	19.36	0.67	0.55
(B.) + (M.).	20.23	18.64	0.63	0.52
30, 60 NP kg / fed.	20.36	18.93	0.62	0.54
(B.) + (M.) + 22.5, 45 NP kg./fed.	20.44	19.32	0.66	0.55
(B.) + (M.) + 15, 30 NP kg./fed	22.36	21.31	0,70	0.63
(B.) + (M.) + 7.5, 15 NP kg./fed	18.72	18.65	0.56	0.57
(B.) + 22.5, 45 NP kg./fed.	21.31	18.00	0.58	0.47
(B.) + 15, 30 NP kg./fed	20.03	18.74	0.57	0.45
(B.) + 7.5, 15 NP kg./fed	19.88	18.64	0.47	0.46
(M.) + 22.5, 45 NP kg./fed	19.94	20.25	0.56	0.49
(M.) + 15, 30 NP kg./fed	19.54	18.74	0.52	0.45
(M.) 7.5, 15 NP kg./fed	19.74	18.50	0.56	0.49
LSD at 0.05	**1.37**	**1.99**	**0.069**	**0.052**

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تـأثير التلقـيح بـالبرادى رايزبيـوم والميكروهيـزا والتسـميد المعـدنى على انتاج وجودة تقاوي البسلة

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اجري هذا البحث فى مزرعة محطة البحوث الزراعية بشندويل – مركز البحوث الزراعيـــة – محافظـــة ســـوهاج – مصـــر، خـــلال موســـمى (٢٠١٣/٢٠١٢) و(٢٠١٤/٢٠١٣م) . وذلك لدراسة تـاثير التلقيح بـالبرادى رايزبيوم والميكروهيزا كبـدائل للتسـميد الكيميـائى ،علـى انتاجيـة تقـاوي البسـلة (صـنف ماسـتر بـي). كـان التصميم المستخدم فى التجربة هو القطاعات كاملة العشوائية فى اربعة مكررات.

تـم اخـذ القـراءات علـى صـفات عـدد الايـام حتـى ٥٠٪ تزهيـر، عـدد الافـرع بالنبات، طول الساق ، عدد القرون بالنبات ، عدد البذور بالقرن ، وزن ١٠٠بذرة ، محصول البذور للفدان والنسبة المئوية للانبـات. وقد اظهرت النتـائج ان جميع هذه الصفات قد تـاثرت معنوياً بعمليـة التلقيح بـالبرادى رايزبيوم والميكروهيزا مقارنـة بالمعاملة الكونترول (بـدون تلقيح اوتسميد) ، فـى كـلا الموسـمين. وقد تـم الحصـول على اعلى القيم من عدد الافرع بالنبات، وزن ١٠٠بذرة ، محصـول البـذور للفدان والنسـبة المئويـة للانبـات مـن خـلال معاملـة التلقـيح بـالبرادى رايزبيـوم + التلقـيح بالميكروهيزا + (١٥نيتروجين + ٣٠ فوسفور للفدان) ، بينما تم الحصول على اقل القيم من معاملة الكونترول ، وذلك فى كلا الموسمين.