RESPONSE OF SOYBEAN TO PHOSPHORUS FERTILIZER UNDER THE EFFECT OF SOME PLANT GROWTH PROMOTING RHIZOBACTERIA

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ABSTRACT

Two-field experiments were conducted at Nubaria Agricultural Research Station, ARC. in summer seasons of 2006 and 2007; consecutively to study the response of soybean cultivar Giza 111, inoculated with Bradyrhizobium japonicum, to different rates of phosphorus fertilizer in the presence of Bacillus megatherium, Bacillus polymyxa or a mixture of both to give a highlight on the role of these microorganisms in increasing enzymatic activity in soil after 75 days of sowing and in increasing soybean yield and N, P and K seeds contents under the calcareous soil conditions in Nubaria. With respect to nitrogenase activity of nodules and rhizosphere and nodules weight, data show that the inoculation increased significantly nitrogenase activity (N2ase) of nodules in both seasons, while it gave a significant increase in nodules weight of the 2nd season. N₂- ase of rhizosphere soil increased significantly in the 1st season as a result of inoculation. Regarding the effect of inoculation and phosphorus fertilizer on dehydrogenase activity (DHA) of rhizosphere soil, results indicated that inoculation with both bacteria led to a significant increase in DHA only in the 2nd season. Results showed also that inoculation of soybean seeds with a mixture of both bacteria plus 30 kg P₂O₅/fed gave the highest significant grain yield, 100-grain weight, protein yield and N, P, and K contents of soybean seeds in both tested seasons over uninoculated treatments. Consequently, results revealed that inoculation of soybean seeds with B. polymyxa combined with 30 kg P2O5/fed produced significantly the highest oil vield as compared to uninoculated treatment.

It can be concluded that the improvement of soybean yield and its quality could be achieved by the application of 30 kg P_2O_5 /fed together with inoculation of seeds with (*Bradyrhizobium japonicum* and a mixture of *B. megatherium*, *B. polymyxa*) under calcareous soil conditions.

Keywords: Soybean, phosphate dissolving bacteria, *B. megatherium, B. polymyxa,* grain yield, N, P and K, inoculation,

INTRODUCTION

Soybean [*Glycine max* (L) Merr.] is an important nutritional and industrial crop and its seeds contain approximately 40% protein of high nutritional value approaching animal protein. Due to the effect of research work and extension, soybean acreage has developed and yield has been increased in Egypt that became at the top level of the world.

Phosphorus (P) is an essential macronutrient, being required by plants in relatively large quantities (~ 0.2 to 0.8%) (Mills and Jones 1996). Providing adequate P to plants is difficult, especially in calcareous soil due to the presence of significant quantities of free excess calcium or magnesium carbonate that causes phosphorus to precipitate in the form of calcium phosphate. For this reason phosphorus is applied to plants particularly legumes such as soybean in high rates.

Regarding the effect of phosphorus on plant, Russel (1973) found that phosphorus enhanced legumes root growth. Attia *et al.* (1990) indicated that application of phosphorus up to 30 kg P_2O_5 /fed. led to a significant increase in soybean yield and its components. Osman *et al.* (2000) reported that soybean plant height, number of pods/plant, dry weight of plant, seed yield, phosphorus and zinc content in leaves were significantly increased by increasing phosphorus fertilizer up to 60 kg/fed. Also, Knany *et al.* (2004) stated that phosphorus can enhanced nodulation and N₂-fixation.

Field trials showed that simultaneous inoculation of non-symbiotic N₂fixing bacteria and naturally rhizobia-colonizing legumes resulted in substantial increases in nitrogenase activity, greater number of nodules and eventually yield increase (Yahalom *et al.*, 1990 and Del Gallo and Fabbri, 1991). The naturally occurring soil bacteria that capable of simulating plant growth named as plant-growth-promoting-rhizobacteria (PGPR) such as *Bacillus* spp. (Kloepper and Schorth, 1981).

Bacillus polymyxa is considered as N₂-fixer; P-dissolving bacteria; phytohormones producer and antibiotic biosynthetic. While, Bacillus megatherium is mainly P-dissolving bacteria; might also be phytohormones producers. Another effect includes an increase in mobilization of insoluble nutrients followed by enhancement of uptake by the plants (Lifshutz et al., 1987). Co-inoculation of rhizobium with PGPR has been shown to increase nodulation, N₂-fixation and yield in a number of legumes, including soybean, under normal growth conditions (Poloneko et al., 1987). Many investigators studied the effect of phosphate dissolving bacteria together with chemical P fertilizer and the results were positive in a step to reduce phosphorus fertilizer being added to legumes such as soybean, for example, El-Sayed (1999) reported that inoculation of lentil seeds with phosphate dissolving organisms increased lentil yield. Ahmed et al. (2002) reported that biofertilization (Rhizobium and phosphate dissolving organisms in addition to chemical fertilization (60 kg N + 30 kg P2O5/fed) increased soybean seed yield and its contents of nutrients as well as oil and protein. Similar results were obtained upon co-inoculation with Bacillus and Rhizobium on faba bean and chickpea (Hassanein et al., 2006). Also, Radwan et al. (2007) found under a field experiment, which was conducted at Malawi station, that the dual inoculation with Rhizobium leguminosarum with Bacillus polymyxa under 50%P led to significant increases in seed and straw yields and seed crude protein per cent compared to control, while under 75% p, gave the highest increases in plant growth, N&P contents and nodulation of faba bean.

The objective of the current study is to investigate the response of soybean to different rates of phosphorus fertilizer in the presence of phosphate dissolving bacteria viz. *Bacillus megatherium* and *Bacillus polymyxa* in calcareous soil and to what extent this technique can increase soybean yield and improve its quality.

MATERIALS AND METHODS

Two field experiments were carried out at Nubaria Agricultural Research Station farm, Agric. Res. Center in summer seasons of 2006 and 2007, consecutively to illustrate the response of soybean [*Glycine max* (L) Merr] cultivar Giza 111, to different rates of phosphorus fertilizer in the presence of bacterial inoculation with *Bacillus megatherium* and *Bacillus polymyxa* and a mixture of them and to highlight the role of these microorganisms in increasing soybean yield under calcareous soil conditions in Nubaria. Representative soil samples from the experimental sites were taken to determine some soil physical and chemical properties according to Chapman and Pratt (1961). The obtained results are shown in Table (1).

The experiment in each season examined 16 treatments as defined below:

(1) Zero addition P₂O₅/fed. (Control)

- (2) 15 kg P_2O_5 /fed
- (3) 30 kg P₂O₅/fed
- (4) $45 \text{ kg P}_2\text{O}_5/\text{fed}$
- (5) Zero addition P_2O_5 /fed + coating with *Bacillus megatherium* (A).
- (6) 15 kg P_2O_5 /fed + coating with (A).
- (7) $30 \text{ kg P}_2O_5/\text{fed} + \text{coating with (A)}$.
- (8) 45 kg P_2O_5 /fed + coating with (A)
- (9) Zero addition P_2O_5 /fed + coating with *Bacillus polymyxa* (B).
- (10) 15 kg P_2O_5 /fed + coating with (B).
- (11) $30 \text{ kg P}_2O_5/\text{fed} + \text{coating with (B)}.$
- (12) 45 kg P_2O_5 /fed + coating with (B).
- (13) Zero addition P_2O_5 + coating with (A+B)
- (14) 15 kg P_2O_5 /fed + coating with (A+B).
- (15) 30 kg P_2O_5 /fed + coating with (A+B).
- (16) 45 kg P_2O_5 /fed + coating with (A+B).

Inocula used:

Bradyrhizobium japonicum, Bacillus megatherium and Bacillus polymyxa, were kindly provided by Biofertilizer Production Unit, Soils, Water &Environment Research Institute, Agric. Res. Center (ARC)Giza, Egypt.

Preparation of bacteria inocula: *Rhizobium* strain was grown on yeast extract mannitol broth medium (Vincent, 1970) incubated at 30°C for three days until early log phase (5X 10^9 cfu / mL culture). Vermiculite supplemented with 10% Irish beat as a carrier was packed in polyethylene bags (200 g carrier per bag), then sealed and sterilized by gamma irradiation (5.0 X 10^6 rads). *Rhizobium* culture was injected into the carrier to satisfy 60% of the maximal water holding capacity. *Bacillus* spp. were grown on nutrient broth (Difico Manual, 1984) incubated for 24 hr at 30°C to obtain population of 5 X 10^8 cfu / mL culture.

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coperintental sites.								
Properties	2006	2007						
Percentages of calcium carbonate and particle sizes								
CaCO₃	29.78	29.65						
Clay	28.20	30.75						
Silt	20.10	20.10						
Sand	51.70	49.15						
Texture class	Sand c	lay loam						
Soil chemical pro	operties							
pH (1:2.5)	8.18	8.16						
О.М	0.56	0.80						
EC dS/m (soil paste extract)	3.10	3.10						
Soluble cations (meq/L)								
Ca ²⁺	9.50	10.11						
Mg ²⁺	6.10	6.00						
Na ⁺	12.20	13.10						
K⁺	3.80	2.90						
Soluble anions (meq/L)							
CO ₃ ²⁻	0.00	0.00						
HCO3-	10.10	9.00						
Cl-	14.45	15.30						
SO4 ²⁻	7.05	7.81						
Available N,P, K								
N (ppm)	17	19						
P (ppm)	6.75	6.25						
K (ppm)	136.00	123.00						

Table (1): Physical and chemical properties of soil samples of the experimental sites.

Mineral fertilizers:

Phosphorus fertilizer in the form of ordinary superphosphate (15.5 % P_2O_5) at a rate following the required treatments was incorporated with soil while preparing seed bed. Potassium fertilizer was added in the form of potassium sulfate (48% K_2O) at the rate of 24 kg K_2O /fed equally to all experimental plots before the first irrigation, similarly, Nitrogen fertilizer was added as ammonium sulfate 20.5% N at the rate of 60 kg N/fed. into two equal split doses to all experimental plots after 30 and 45 days from sowing.

The above mentioned treatments were arranged in a complete randomized design with three replicates and soybean seeds, cultivar Giza 111, kindly supplied by Field Crops Research Institute, ARC, Giza, were wetted by Arabic gum solution (16%) as adhesive agent, then inoculated with *Bradyrhizobium japonicum* at a rate of 400 g inoculum/40 kg seeds. The inoculum was added and thoroughly mixed with seeds and finally, seeds were inoculated with *B. megatherium* and/or *B. polymyxa* at a rate of 10⁶ cfu /

seed, which grown in summer of 2006 and 2007 by in plots of 3 x3.5 m (1/400 Faddan) each. In both seasons, wheat was prior to soybean in the two experimental sites, which were under furrow irrigation.

After 75 days of sowing, nitrogenase enzyme (N₂-ase) activity of nodules and rhizosphere was assayed by the acetylene reduction assay (Hardy *et al.*, 1973), also nodules were dried then weighed. Dehydrogenase (DHA) enzyme activity of rhizosphere plants was also determined by the method described by Thalmann (1967).

At harvest, seed yield was estimated in ton/fed and seed index was calculated as weight of 100-seeds. Seed nutrient contents of N, P and K were determined in line with Jackson (1973). Moreover, oil and protein contents in seeds and their yields were evaluated as per AOAC (1980).

Statistical analysis was carried out as a randomized complete design (Snedecor and Cochran, 1980) using LSD test to compare means of treatments in investigation.

RESULTS AND DISCUSSION

I-Enzyme activities

Nitrogenase activity in soybean root nodules in μ mole C₂H₄ /g dry nodules / h, nodules weight, nitrogenase enzyme in soybean rhizosphere in µ mole C₂H₄ /g dry nodules / day and dehydrogenase enzyme in rhizosphere zone of roots in µg TPF/g dry soil /day after 75 days of soybean sowing are presented in Table (2). With respect to nitrogenase and nodules weight, results indicate that inoculation in general led to increase N2-ase activity in both tested seasons. In details, in 1st season, the highest value of nitrogenase activity was due to the treatment received 45 kg P2O5 and inoculated with *B. polymyxa*, while this trend was achieved by the treatment received 45 kg P₂O₅ and inoculated with *B. polymyxa* and *B. megatherium* at 2nd season. Similar results were obtained by Rai (1983) on chickpea. The reached results at uninoculated treatments were acceptable even they were lower than the other inoculated treatments, this behavior could be due to the positive role of native bacteria mainly rhizobia and other PGPR inhabiting soil among several decades ago (Kloepper and Schorth 1981). Poloneko et al. 1987 reported that co-inoculation with rhizobium and PGPR increased nodulation, N₂-fixation and yield in a number of legumes, including soybean, under normal growth conditions. Also, inoculation increased significantly nodules weight in the 2nd season. This result is in harmony with those obtained by Yahlom et al. 1990 who stated that these increases attributed to synergistic effect upon co-inoculation include auxins production, which have a vital role either in nodules formation or nodules activity where auxins are important for maintaining functional role nodules; effective nodules have a higher IAA content than ineffective nodules. Also, Abo El-Soud et al. (2003) and Mekhemar et al. (2005) found that inoculation with rhizobium alone or combined with helper bacteria increased dry weight of shoots, N-content, nodulation and yield of faba bean, which could be attributed to nitrogen fixation that consequently increased plant growth.

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Table (2	2): E	ffect	of	phosp	horous	fertilizer	and	pho	osphate	e d	issolving
	k	oacteri	a o	n nitro	ogenase	(N ₂ -ase)	activi	ty, r	nodules	5 W	eight and
	C	dehydr	oge	enase	activity	(DHA)	after	75	days	of	soybean
	S	sowing	j at	2006 a	and 200	7 summe	r seas	sons	5		

Treatment	N ₂ -ase (µ mole C ₂ H₄/g dry nodules /h)		Nodules weight (g)		N ₂ -ase (C ₂ Hz rhizosphe	u mole /g ere/day)	DHA (µg TPF/g rhizosphere soil / day)	
	2006	2007	2006	2007	2006	2007	2006	2007
Zero P ₂ O ₅ fed ⁻¹	14.17255	18.9432	0.0597	0.0730	31.942	7.94	36.809	103.03
15 kg P₂O₅fed⁻¹	15.98058	131.0969	0.0895	0.0332	39.301	8.73	53.472	91.79
30 kg P₂O₅fed⁻¹	25.12425	36.91279	0.0570	0.0500	43.447	8.54	48.828	103.00
45 kg P₂O₅fed⁻¹	47.47574	93.76736	0.1567	0.0460	4.693	1.18	45.231	81.62
Zero P₂O₅fed⁻¹ + A	85.36302	693.7875	0.0360	0.0650	56.266	15.43	58.817	122.99
15 kg P₂O₅fed⁻¹+ A	103.8283	411.0768	0.0245	0.0515	58.254	11.43	60.671	105.37
30 kg P₂O₅fed⁻¹+ A	94.95374	398.7878	0.0228	0.0430	110.835	17.81	64.889	137.84
45 kg P₂O₅fed⁻¹+ A	49.91112	568.4404	0.1758	0.0255	37.448	16.80	59.318	143.94
Zero P₂O₅fed⁻¹ + B	90.78277	361.7289	0.0420	0.0120	56.328	7.99	63.067	105.73
15 P₂O₅fed ⁻¹ + B	49.97405	712.1132	0.1079	0.0180	35.932	8.55	57.439	114.20
30 P ₂ O₅fed ⁻¹ + B	53.98022	584.2486	0.0461	0.0342	89.387	9.80	57.757	148.03
45 P ₂ O ₅ fed ⁻¹ + B	134.4815	498.0503	0.0729	0.0370	76.652	6.48	60.909	104.64
Zero P ₂ O ₅ fed ⁻¹ + (A+B)	116.4238	1315.411	0.0476	0.0088	91.4191	20.80	63.027	144.95
15 kg P₂O₅fed⁻¹ + (A+B)	102.0403	1765.343	0.0412	0.0910	63.1101	16.05	61.723	91.25
30 kg P₂O₅fed⁻¹ + (A+B)	98.7209	1279.039	0.0904	0.0285	85.2417	7.48	60.366	155.48
45 kg P₂O₅fed⁻¹ + (A+B)	79.0273	1117.402	0.1063	0.0200	162.312	18.85	52.237	113.74
LSD at 0.05	49.71	674.2494	N.S.	0.0288	62.62	N.S.	N.S.	36.61
A· B. megatheriu	m			B B no	lvmvya			

polymy

Srinivasan et al.(1996) stated that Bacillus isolates combined with Rhizobium inoculation increased nitrogenase activity and nodule protein content of Phaseolus vulgaris. In this connection, Rashad and Ragab (2003) added that faba bean plants inoculated with PGPR showed significant increases in nodules numbers and dry weight, this could be due to the stimulation effect of tested bacteria, which encouraged growth of bacteria in rhizosphere including native rhizobia, and root hairs of plants (Antoun et al., 1998).

II. Yield and its related components:

Data in Table (3) illustrate that in seasons, 2006 and 2007; soybean seeds inoculated with a mixture of both Bacillus spp. and received 30 kg P2O5 fed⁻¹ statistically achieved the highest grain yield, one 100-grain weight and protein yield over uninoculated seeds. The enhancement effect of phosphorus fertilizer on soybean plants may be due to the development of plant nucleoproteins, transfer of metabolite compounds and the efficiency of its root system leading to more absorption of water and nutrients, which led to increase the rate of physiological processes and gave better yields. The

present results are in agreement with Attia *et al.* (1990), Seif El-Nasr Osman *et al.* (2000) and Badran (2003). El-Sayed (1999) and Ahmed *et al.* (2002) who studied the effect of phosphate solubilizing microorganisms and phosphorus fertilizer on soybean and lentil plants, they found that this treatment led to higher nutrient concentrations in seeds and an increment in seed yield.

Regarding the effect of PGPRs on yield, Lata-Saxena and Tilak (2002) stated that phosphate solubilizing microorganisms offer a biological rescue system capable of solubilizing the insoluble inorganic P of soil and let it available to the plants. Hussein *et al.* (1997) and Saleh *et al.* (2000) reported that P fertilizers combined with rhizobial inoculation led to better nodulation and higher increases in yield of soybean and faba bean in clay loam soil. This result is in harmony with those obtained by Abo EI-Soud *et al.* (2003) and Mekhemar *et al.* (2005) who found that rhizobium alone or combined with helper bacteria increased dry weight of shoots, N-content, nodulation and yield, which could be attributed to nitrogen fixation that consequently increased plant growth.

Results also showed that in both seasons soybean seeds inoculation with *B. megatherium* supplied with 30 kg P_2O_5 fed⁻¹ produced significantly the highest oil yield as compared with uninoculated treatments. This increment in oil yield could by related to the increase in oil percentage and grain yield of soybean. Similar results were achieved by Ahmed *et al.*(2002).

Treatment	Grain yield (Ton/fed)		100-grain weight(g)		Protein yield (kg/fed)		Oil yield (kg/fed)	
	2006	2007	2006	2007	2006	2007	2006	2007
Zero P₂O₅fed ⁻¹	0.641	0.684	17.94	17.90	171.73	188.10	109.03	131.67
15 kg P₂O₅fed⁻¹	0.741	0.744	19.78	18.78	209.23	192.79	121.16	143.08
30 kg P₂O₅fed⁻¹	0.786	0.693	18.95	19.67	228.24	217.45	127.04	120.14
45 kg P₂O₅fed⁻¹	0.770	0.757	19.52	18.63	216.78	244.45	113.83	148.25
Zero P ₂ O ₅ fed ⁻¹ + A	0.751	0.751	18.04	18.98	220.16	232.59	131.49	137.20
15 kg P₂O₅fed⁻¹+ A	0.814	0.687	18.07	18.64	209.52	225.42	127.81	132.25
30 kg P₂O₅fed⁻¹+ A	0.794	0.719	20.21	19.93	198.50	238.28	118.31	127.68
45 kg P₂O₅fed⁻¹+ A	0.826	0.801	20.16	18.35	252.70	250.21	106.32	142.12
Zero P ₂ O ₅ fed ⁻¹ + B	0.668	0.688	19.87	18.68	202.49	198.52	107.55	127.28
15 P ₂ O ₅ fed ⁻¹ + B	0.706	0.723	19.92	19.61	218.82	225.84	122.55	139.11
30 P ₂ O ₅ fed ⁻¹ + B	0.828	0.809	20.57	19.23	228.36	264.64	186.23	190.12
45 P ₂ O ₅ fed ⁻¹ + B	0.847	0.819	19.69	20.13	262.04	274.36	138.16	139.23
Zero P ₂ O ₅ fed ⁻¹ + (A+B)	0.737	0.700	19.58	19.62	222.88	221.07	132.91	126.00
15 kg P₂O₅fed⁻¹ + (A+B)	0.864	0.734	20.41	19.22	244.78	239.30	149.09	124.72
30 kg P₂O₅fed⁻¹ + (A+B)	0.965	0.924	21.21	22.23	390.02	370.18	108.08	152.46
45 kg P₂O₅fed⁻¹ + (A+B)	0.790	0.823	19.29	19.78	217.25	279.31	136.74	143.97
LSD at 0.05	0.081	0.076	1.37	1.21	50.68	40.13	28.19	17.76

Table (3): Effect of phosphorous fertilizer and phosphate dissolving bacteria on yield, 100-grain weight, protein and oil yield in soybean in 2006 and 2007 summer seasons

A: B. megatherium

B: B. polymyxa

III. N, P and K uptake of soybean seeds:

Data in Table (4) indicate that inoculation of soybean seeds with both microbial strains mixture combined with 30 kg P_2O_5 / fed achieved significantly the highest N, P and k content in seeds in both seasons as compared with uninoculated soybean seeds. However, higher nutrient concentrations in seeds and the increment in seed yield due to the effect of phosphorus fertilizer and phosphate dissolving bacterial strains are in harmony with those reported by El-Sayed (1999) and Ahmed *et al.* (2002). Lata- Saxena and Tilak (2002) stated that supplying combined nitrogen by nitrogen fixing bacteria had affected the development and function of roots by improving mineral (NO³, PO³ and K⁺) and water uptake. Also, Ragab and Rashad (2003) found that rhizobacteria stimulate plant growth by producing growth regulators such as vitamins and indole acetic acid, enhance plant nutrients and water uptake.

Table (4):	Effect of phosphorous fertilizer and phosphate dissolving
	bacteria on N, P and K content in soybean seeds in 2006 and
	2007 summer seasons

	1	N		C	K						
Treatment		Kg fed ⁻¹									
	2006	2007	2006	2007	2006	2007					
Zero P₂O₅fed⁻¹	27.48	30.10	4.56	4.90	8.31	8.92					
15 kg P₂O₅fed⁻¹	33.47	30.85	5.19	4.49	8.75	10.39					
30 kg P₂O₅fed⁻¹	36.52	34.79	5.90	4.80	11.62	9.36					
45 kg P₂O₅fed⁻¹	34.68	39.11	5.78	5.30	14.17	11.36					
Zero P₂O₅fed⁻¹ + A	35.28	37.22	4.73	4.73	11.92	11.89					
15 kg P₂O₅fed⁻¹+ A	33.53	36.07	5.92	4.81	9.34	11.77					
30 kg P₂O₅fed⁻¹+ A	31.76	38.13	6.04	5.03	9.93	11.60					
45 kg P₂O₅fed⁻¹+ A	40.43	40.03	5.20	5.05	12.31	13.16					
Zero P₂O₅fed⁻¹ + B	32.40	31.76	4.21	4.33	9.62	9.91					
15 P ₂ O₅fed ⁻¹ + B	35.01	36.13	5.06	4.55	10.25	11.27					
30 P₂O₅fed⁻¹ + B	36.54	42.34	4.43	5.37	13.82	12.16					
45 P₂O₅fed⁻¹ + B	41.93	43.90	4.24	5.73	11.48	13.57					
Zero P₂O₅fed⁻¹ +	35.66	35.37	3.69	4.06	12.98	12.30					
15 kg P₂O₅fed ⁻¹ +	39.16	38.29	5.44	4.87	11.63	10.74					
30 kg P₂O₅fed⁻¹ +	62.40	59.23	8.49	7.39	14.76	14.51					
45 kg P₂O₅fed ⁻¹ +	34.76	44.69	4.98	5.22	11.77	11.39					
LSD at 0.05	8.11	6.42	0.69	0.43	2.84	1.78					
A: B. megatherium	•	Ē	B: B. polyn	iyxa	•	•					

Finally, as concluded in Tables (3 & 4) the significant effect of inoculation with both tested bacteria combined with 30 kg P_2O_5 / fed resulted in more absorption of N, P and K elements by soybean plant roots and consequently the rate of physiological processes were increased and reflected to soybean seed and protein yields and 100-seed weight. While, the

significant increase in oil yield was attributed to the inoculation only with *B. polymyxa* with the same rate of P-fertilizer.

Conclusion

In conclusion, the inoculation of soybean with *Bradyrhizobium japonicum* combined with a mixture of *B. megatherium* and *B. polymyxa* along with 30 kg P_2O_5 fed⁻¹may be useful in improving soybean seed and quality in calcareous soils. However, more studies should be done to establish this idea.

REFERENCES

- Abo El-Soud, A. A.; A. A. Ragab; G. A. A. Mekhemar and F. T. Mekhaeel (2003): response of faba bean to inoculation with N₂-fixers and phosphate dissolving bacteria as influenced by different sources of phosphorus. Egypt J. Appl. Sci.,18: 73-90.
- Ahmed, Kh. A; Madiha M. Badran and Samia, H. Ashmawy (2002): Response of soybean to chemical and bio-fertilization. Egypt. J. Appl. Sci., 17 (6), 207-218.
- A. O. A. C. (1980): "Official Methods of Analysis". The Association of Official Agriculture Chemists 13th ed Washington DC., USA.
- Antoun, H.; C. J. Beauchamp; N. Goussard; R. hbat and R. Lalande (1998): Potential of *Rhizobium* and *Bradyrhizobium* species as plant growth promoting rhizobacteria on non-legumes; Effect on rdishes (*Raphanus stivus* L.). Plant and Soil. 204: 57-67.
- Attia, A.N; M.H. Ghonema and A.M. Salama (1990): Effect of timing of nitrogen application, phosphorus, potassium and zinc fertilization on growth and yield of soybean. Proc. 4th Conf. Agron., Cairo. 11: 135-148.
- Badran, Madiha M.(2003): Effect of nitrogenous and phosphatic fertilization on some economical characters of soybean Crawford cultivar under calcareous soil conditions. Egypt. J. Agric. Res., 81 (2): 433-439.
- Chappman, S. R. and P. F. Pratt (1961): Methods of Analysis for Soils, Plants and Water. University of California, USA. p 317.
- Del Gallo, M. and P. Fabbri (1991): Effect of soil organic matter on chickpea inoculated with Azospirillum brasilense and Rhizobium leguminosarum bv. Ciceri. Plant and Soil. 137:171-175.
- Difico Manual (1984): Dehydrated culture midi and Reagents for Microbiology. 10th Edition. Difico Laboratories, Detroit, Michigan, USA.
- El-Sayed, S. A. M. (1999): influence of Rhizobium and phosphate solubilizing bacteria on nutrient uptake and yield of lentil in the New valley. Egypt J. Soil Sci., 39 (2):175.
- Hardy, R. W. F.; R. C. Burns and R. D. Holsten (1973): Application of the acetylene-ethylene reduction assay for measurement of nitrogen fixation. Soil Biol. Biochem., 5: 47-81.
- Hassanein, A. M., A. M. El-Garhy and G. A. A. Mekhemar (2006): Symbiotic nitrogen fixation process in faba bean and chickpea as affected by

biological and chemical control of root-rot. J. Agric. Sci., Mansoura Univ., Egypt. 31 (2): 963-980.

- Hussein, A. H. A.; S. A. Saleh; M. A. El Deeb and W. Kadry (1997): Effect of Rhizobial inoculation, phosphorus and potassium fertilization on growth, nodulation and yield of faba bean cultivated in the newly reclaimed soils of middle Egypt. Bull. Fac. Agric. (Cairo Univ., Egypt).48:201-214.
- Jackson, M. L. (1973): Soil Chemical Analysis. Prentice-Hall of India, New Delhi.,India.
- Knany, R. E.; A. M. Masoud and Y. B. El-Waraky (2004): Comparative study between biofertilization and sulphur on availability of added phosphorus to Faba bean plants under high p^H soil conditions. J. Agric. Sci., Mansoura Univ., 29: 4801-4809.
- Kloepper, J. W. and M. N. Schroth (1981): Plant growth promoting rhizobacteria and plant growth under genobiotic conditions. Phytopathology. 71:642-644.
- Lata-Saxena, A. K. and Tilak, K. V. (2002): Biofertilizers to Augment Soil Fertility and Crop production. In: Soil Fertility and Crop Production. (Ed. Krishna, K. R.), Science Publishers, USA, pp. 279 – 312.
- Lifshutz, R.; J. W. Kloepper and M. Kozolowski (1987): Growth promotion of canola (rapeseed) seedlings by a strain of *Pseudomonas putida* under genobiotics conditions. Can. J. Microbiol., 33: 390-395.
- Mekhemar, G. A. A.; M. Shaaban; A. A. Ragab and A. M. M Biomy (2005): Response of faba bean to inoculation with *Rhizobium leguminosarum* bv. Viceae and plant growth promoting rhizobacteria undes newly reclamed soils. Egypt J. Appl. Sci.,20: 126-144.
- Mills, H. A. and J. B. Jones. (1996): Plant Analysis Handbook II. MicroMacro Publishing, Athens, GA.
- Osman, A.S; Y.M.Y. Abido and S.M.M. Allam (2000): Response of soybean to phosphorus and zinc fertilization under irrigation regime. Annalls of Agric. Sci. Ain Shams Univ., Cairo. 45 (1): 229-238.
- Poloneko D. R.; F. M. Scher; J. W. Kloepper; C. A. Singleton; M. Laliberte and I. Zaleska. (1987): Effect of root colonizing bacteria on nodulation of soybean roots by *Bradyrhizobium Japonicum*. Can. J, Microbiol., 33:498-503.
- Radwan, T. E. E; B. A. Hassona; Ilham I. El-Khatib and B. A. A. Kandil (2007): Impact of nitrogen fixers and phosphate dissolving bacteria on faba bean under the influence of different amounts of phosphorus. Egypt. J. Microbiol., (16) :177-184.
- Ragab, A. A. and M. H. Rashad (2003): Enhancement of nutrients uptake and metabolism efficiency of sorghum (*sorghum bicolor* L) plants by biofertilizer under water stress. Zagazig J. Agric. Res., 30: 147-166.
- Rashad, M. H. and. A. A.Ragab (2003): the influence of some plant growth promoting rhizobacteria on the growth and yield of sorghum *bicolor* L.) plnts under drought stress. Plant nutrition-Food security and Sustainability of Agro-ecosystems, 664-665.

- Rai, R. (1983): Efficacy of associative N₂-fixation by streptomycin-resistant mutants of *Azospirillum brasilense* with genotypes of chickpea *Rhizobium* strains. J. Agric. Sci., 100:75-80.
- Russel, E. W. (1973): Soil conditions and plant growth. Language Book Society and Longman. London. 30 - 37.
- Saleh, S. A.; M. A. El-Deeb and A. A. Ragab (2000): Response of faba bean (*Vicia Faba* L.) to Rhizobium inoculation as affected by nitrogen and phosphorus fertilization. Bull. Fac. Agric., Cairo Univ., Egypt. 51: 17-30.
- Snedecor, G. W. and W. G. Cochran (1980): Statistical Methods 7th ed Iowa State Univ. press Iowa, USA.
- Srinivasan, M.; D. J. Peterson and F. B. Holl (1996): Influence of IAA producing *Bacillus* isolates on nodulation of *Phaseolus vulgaris* by *Rhizobium itli*. Cand. J. Microbiol., 42: 1006-1014.
- Thalmann, A. (1967): Uber die microbiello aktiviatat undihr benziehung zu frucht- bartkeits merkmalen einiger acherboden unter besonderer beruksi chtigung der Dehydrogenase aktiviatat (TTC. Reduktion). Biss, Gieben. pH. D. Thesis, W. Germany.
- Vincent, J. M. (1970) : A manual for practical study of root-nodule bacteria. IBP Handbook, No. 15, Blackwell Sci., Pub., Ltd., Oxford and Edinburgh, UK.
- Yahalom, E.; Y. Okka and A. Dovart (1990): Possible mode of action of Azospirillum brasilense strain CD on the root morphology and nodule formation in burr medic (*Medicago polumorpha*). Cand. J. Microbiol., 36:10-14.

إستجابة فول الصويا للسماد الفوسفاتي تحت تأثير بعض البكتريا المشجعة لنمو النبات

محمد السيد مرسي ، مدحت يماني أبوزيد و حمدي و هبه أحمد و هبه معهد بحوث الأراضي والمياه والبيئة ــمركز البحوث الزراعية- الجيزة ــ مصر

أقيمت تجربتان حقليتان في محطة البحوث الزراعية بالنوبارية التابعة لمركز البحوث الزراعية في الموسمين الصيفيين 2006 ، 2007 على التوالي لدراسة إستجابة فول الصويا صنف جيزة 111- الملقح ببكتيريا برادي ريزوبيوم جابونيكوم- لمعدلات مختلفة من السماد الفوسفاتي في وجود بكتريا باسيلاس ميجاثيريوم أو باسيلاس بوليميكسا ومخلوط منهما وابراز دور هذه الكائنات الدقيقة في زيادة النشاط الإنزيمى لميكروبات التربة بعد 75 يوم من الزراعة وزيادة محصول فول الصويا كذلك محتوى البذور من عنصر النيتروجين والفوسفور والبوتاسيوم تحت ظروف الأرض وزيادة محصول فول الصويا كذلك محتوى البذور من عنصر النيتروجين والفوسفور والبوتاسيوم تحت ظروف الأرض ووزن العقد الجذرية أظهرت البياتات أن التلقيح البكتيري زاد نشاط انزيم النيتر وجينيز معنويا في العقد الجذرية في ووزن العقد الجذرية في العقد الجذرية ومنطقة نمو جذور النبات (الريزوسفير) ووزن العقد الجذرية أظهرت البيانات أن التلقيح البكتيري زاد نشاط انزيم النيتر وجينيز معنويا في العقد الجذرية في ووزن العقد الجذرية أظهرت النيان التلقيح البكتيري زاد نشاط انزيم النيتري وجينيز معنويا في العقد الجزية في الريزوسفير معنويا في الموسم الأول نتيجة للتلقيح وفيما يتعلق بتأثير التلقيح البكتيري والسماد الفوسفاتي على نشاط انزيم الريزوسفير معنويا في الموسم الأول نتيجة للتلقيح وفي التقيح البكتيري والسماد الفوسفاتي على نشاط انزيم البريزيوسينيز في منطقة الريزوسفير أشارت النتلقيح البكتيري بكلا من نوعى البكتيريا قد أدى إلى زيادة نشاط البريزيم الديهيدر وجينيز معنويا في الموسم الثاني فقط والناني م النيترة وجينيز الزيم الديهيدر وجينيز معنويا في الموسم الثاني فقط وأظهرت النتلقيح البكتيري والسماد الفوسفاتي على نشاط انزيم البكتيريتين + 30 كم فوح أدري أسرت النتاقيح النتانيوي في والماد الغروبين ومحتوى الإزيم الديهيدر وجينيز معنويا في الموسفور والبوتاسيوم في كلا الموسمين مقارنة بالمعاملات البروتين ومحتوى البذوم من عناصر النيتروجين والفوسفور والبوتاسيوم في كلا الموسمين مقارنة بالمعاملات الغير ما محتوى أشارت التذائج أن تلقيح بذور فول الصويا ببكتيريا باسيلاس بوليميكسا مع 30 كجم فوح أول فدان قد أنتج أعلى محصول مونو ما رزيم مقارنة بالمعاملة الغير ماقصة.

والُخلاصة أنه من الممكن تُحسين فول الصويا كما ونوعا بإضافة 30 كجم فو2 ا5/ فدان مع التلقيح البكتيري للتقاوي بمخلوط من بكتيريا برادي ريزوبيوم جابونيكوم وبكتيريا باسيلاس ميجا ثيريوم وبكتيريا باسيلاس بوليميكسا تحت ظروف الأراضي الجيرية.