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THE POTENTIAL IMPROVEMENT OF *Zea mays* YIELD AND ITS CONTENT OF PHOSPHORUS AND NITROGEN AS A RESULT OF DIFFERENT INOCULATION METHODS WITH PHOSPHATE-DISSOLVING BACTERIA

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ABSTRACT

The interaction between phosphate-dissolving bacteria and *Zea mays* plants was investigated. Results showed that: Highly variation in the ability of phosphate-solubilizing bacteria in bringing down the pH of the media and consequently varied in their ability in releasing soluble phosphorus. The highest level of soluble-P (421.9 ppm) was released by isolate No. 15. Greatest amount of maize dry weight and grain yield was obtained with the inoculation at 10 and 20 days after planting. The inoculation at 10 and 20 days with cell suspension was more active than its application as cell suspension to the grains one hour before sowing. P and N content in maize plants and their grain were increased significantly with the treatments of grain slurry plus supplementary inoculation of cell suspension at 10 or at 10 and 20 days after planting and those inoculated with cell suspension at 10 and 20 days from planting. There are a positive correlation between inoculation with phosphate dissolvers and N-content in the plant during the two seasons. Results also showed maximum counts of phosphate dissolving bacteria in rhizosphere soil of maize after 60 days from sowing in all treatment, which gradually increased up to 60 days, thereafter decreased. The inoculation of maize plants with phosphate dissolving bacteria increased the amount of soluble phosphorus in the rhizosphere soil of maize compared to the non-inoculated soil. The increase of phosphorus content was significant with the treatment of seed slurry plus supplementary cell suspension at 10 and 20 days after planting. Therefore, the application of rock phosphate and inoculation with efficient phosphate-dissolving bacteria produced more amount of soluble phosphorus in the soil , which plays an important role in metabolic pathways in living organisms and thus increased the yield of plant.

Keywords: *Zea mays*, phosphate-solubilizing bacteria, inoculation, rock phosphate.

INTRODUCTION

Maize is one of the major grain crops worldwide including as well as in Egypt. Any effort to increase maize yield to face the continual increasing of consumption is highly appreciated. It is well known that phosphorus is considered as one of the limiting factors to achieve the high yield of crop, which it is a core of the metabolic pathways of the living organisms. With the steadily increasing phosphorus and nitrogen fertilizers prices and the pollution problems, efforts to decrease chemical fertilizers by using biofertilizers might reduce financial costs as well as chemical pollution. Also,

there is a wide interest in the use of compination of mineral– and bio-fertilizer as substituent and cheap source for chemical fertilizers (Hernandez *et al.*, 1995; Hauka *et al.*, 1996; Farag, 1998; Kotb, 1998 Hauka, 2000 and Hammouda *et al.*, 2001).

The population of phosphate-solubilizing bacteria was higher in some soil herizones. The population mostly conistituted more than 10% of the total bacteria (Gupta *et al.*, 1986 and Krishanaraj & Gowda, 1990). Phosphate-solubilizing bacteria, *Azotobacter* sp and vesicular- arbuscular mycorrhiza (VAM) increased grain yield by 31, 37 and 6%, respectively, in comparison with non biofertilized control. Very high grain yield (25%) was recorded with application of such inoculants compared to those of NPK alone (Mishra *et al.*, 1995). Also, application of biofertilizers to soil improved the microbial counts of *Azotobacter*, *Azospirillium* and phosphate-dissolving bacteria in rhizosphere of plants (El-Sersawy *et al.*, 1997). Thus, there have been many successful attempts to improve maize yield and its grains supplemented by using mixtures of nitrogen-fixing bacteria and mycorrhiza (Hao *et al.*, 1991 and Hauka, 2000). Maize plants growth and nutrient content were increased by dual inoculation with phosphate-dissolving bacteria and VA mycorrhiza in the presence of rock phosphate (Heggo & Barakah, 1983 and Hauka, 2000). Furthermore, the beneficial influence of phosphate-solubolizing bacteria on survival of nitrogen–fixing bacteria in the rhizosphere has been observed. Other results have been recorded after inoculation of sorghum with *Azospirillium* and phosphate-dissolving bacteria (Kundu & Gaur, 1980; Alagawadi & Gaur, 1988 and Hauka, 2000).

Therefore, this study was focused to evaluate the effect of different inoculation methods of phosphate-solubilizing bacteria on stalks and grain yield of *Zea mays* plants and its containing of phosphorus and nitrogen amount grown in soil amended with rock phosphate.

MATERIALS AND METHODS

Microorganisms:

Twenty bacterial isolates of *Bacillus megaterium* were selected from rhizosphere soil of maize which showed positive reaction on plates of Bunt and Rovira medium (1955). To evaluate the efficiency of each isolate in increasing the availability of precipitated phosphorus, Erlenmeyer flasks containing Bunt and Rovira medium (1955) modified by Taha *et al.* (1969) were prepared. Each flask containing 90 ml of the medium was supplemented with 0.25 g of tricalcium phosphate. The pH was adjusted to 7.4 to ensure minimal soluble phosphate concentration in the medium. Each bacterial isolate was grown separately for 10 days at 30°C.

Changes of pH and amounts of soluble phosphorus were measured after 10 days. The soluble phosphorus was determined according to methods recorded by Jackson (1973). The most efficient isolate was selected to be used as phosphate dissolving bacterial inoculum.

Preparation of inocula:

The selected phosphate dissolving bacteria was grown in nutrient broth medium (Oxoid Manual, 1965) for five days (giving 10^9 cells/ml), and either impregnated into a sterile finely ground composted cotton and husk as a carrier to give 10^8 cells / g, or applied directly as a cell suspension containing 10^8 cells/ml. The following inoculation treatments were tested: grain inoculation with cell suspension one hour before sowing (SI); inoculation the soil beside the plants with 10 ml/plant with cell suspension (CS) at 10 or at 10 + 20 days after planting (DAP); grain slurry inoculation with composted cotton seed husk inoculum (SS) one hour before sowing; seed slurry followed by supplementary inoculation with 10 ml/Plant of the cell suspension beside the plants at 10 (SS + CS 10) or at 10 + 20 (SS + CS 10 and 20 DAP), and non-inoculated seeds as control.

Field experiments:

The present work was carried out under field conditions in the Experimental Farm of the Faculty of Agriculture, Minia University during 2002 and 2003 seasons.

The soil type was clay loam, with pH 7.85, coarse sand, 2.4%, fine sand 23.5%, silt 31.2%, clay 42.5%, organic matter 1.45%, total N 0.14%, $CaCO_3$ 2.25% and available P 13.35 ppm.

A randomized complete block design with four replicates was used. Each plot area was 3 x 3 m. grains of maize (*Zea mays* L.) variety of Twc 310 were planted with 50 and 25 cm between and within rows, respectively. Five seeds were planted and later thinned .to two plants per hole.

Recommended dose of nitrogen fertilizer (120 Kg/fed) as reported by Ministry of Agriculture in Egypt was applied in three equal doses before first, second and third irrigation in the form of urea (46% N).

Rock phosphate (28.0% P_2O_5) was added at a rate of 100 Kg/fed one week before sowing. All agricultural practices of growing maize were conducted as usual.

At harvest the three inner rows were selected for determining dry matter of straw (Kg/fed), grain yield (ardab/fed), and P and N content in stalks and grain (Kg/fed).

Total N and total P were determined according to the methods recorded by Jackson (1973),

Statistical analysis:

All collected data were subjected to analyses of variance according to the procedure outlined by Steel and Torrie (1980).

Enumerating phosphate-dissolving bacteria in rhizosphere soil of maize:

Samples of rhizosphere soil of maize were collected by mechanical removal of the tightly adhering soil after shaking the roots together. Serial dilution of rhizosphere soil were made under sterile conditions. Bunt and Rovira medium (1955) modified by Taha *et al.* (1969) was used. Phosphate-dissolving bacteria were readily detected by clear zones around the colonies

after incubation at 30°C for 48 h. The counts were recorded at intervals of 15 days up to 90 days after sowing.The amounts of soluble P (ppm) in the rhizosphere soil were also measured.

RESULTS AND DISCUSSION

Results presented in Table (1) showed the reduction in pH values and the increase of the soluble phosphorus in the media during the growth of the tested phosphate-dissolving bacterial isolates (*Bacillus megaterium)* in liquid medium containing insoluble phosphate. It is known that the ability of bacteria to dissolve the precipitated form of phosphorus, $Ca_3(PO_4)_2$, depends on its ability in producing inorganic acids or organic ones and CO_2 (Laune *et al.*,1981 and Rajan *et al.*,1991). As shown in Table (1) the tested bacterial isolates varied in their ability in bringing down the pH of the media and consequently varied in their ability in releasing soluble phosphorus. The highest amount-P (421.9 ppm) was released by bacterial isolate No. 15, which was able to reduce the pH of the medium to 3.85 within 10 days. Accordingly, this isolate (No. 15) was selected to be used as phosphate dissolving inoculum for further experimentation.

Table (1): The efficiency of phosphate-dissolving isolates in solubilizing the precipitated phosphorus in liqid culture media after incubation at 30°C for 10 days.

Isolate No.	Soluble P (ppm)	PH
1	115.8	5.10
2	265.3	4.70
3	108.0	5.60
4	39.4	7.10
5	135.8	4.85
6	279.3	4.60
7	37.4	7.10
8	48.6	6.90
9	315.3	4.20
10	170.2	5.17
11	130.0	5.62
12	196.3	5.31
13	415.6	4.15
14	161.4	5.60
15	421.9	3.85
16	312.7	5.10
17	326.4	4.61
18	281.7	4.85
19	95.3	6.10
20	119.5	5.71

The effect of inoculation methods with *Bacillus megaterium* as phosphate-disscivers inoculant on dry weight (Kg/fed) and grain yield (ardab/fed) of maize is presented in Table (2). The data clearly show that, the split addition with phosphate dissolver inoculum recorded the greatest values of dry weight and grain yield of maize at the two seasons. The slurry plus supplemental suspension inoculation at 10 and 20 days after planting was slightly better (but not significant) than the slurry plus cell suspension inoculation at 10 days after planting. However, the two treatments showed significant values of dry matter and grain yield as compared to other treatments. On the other hand, application of phosphate dissolvers inoculum as cell suspension to the plants at 10 and 20 days after planting more efficient active than its application as cell suspention to the seeds one hour before sowing. However, the non-inoculated treatments recorded the lowest values of dry matter and grain yield in both the two seasons, indicating the beneficial effect of phosphate dissolvers inoculation (Satter and Guar, 1989; Saber and Kabesh, 1990; Datta *et al.*, 1992; Heggo & Barakah, 1993; Singh *et al.*, 1995 and Dubey *et al.*, 1997).

Table (2): Effect of different inoculation treatment by phosphate dissolvers on dry matter of straw (KG/fed) and grain yield (ardab/fed) of maize.

Inoculation treatments	2002 season		2003 season	
	Stalks	Grain yield	Stalks	Grain yield
SI	2150[a]	17.8[a]	2190[a]	18.2[a]
CS 10 DAP	2180[a]	18.1[a]	2210[a]	18.6[a]
CS 10 and 20 DAP	2211[a]	18.6[a]	2230[a]	18.9[a]
SS	2155[a]	18.3[a]	2180[a]	18.3[a]
SS + CS 10 DAP	2261[b]	18.9[b]	2280[b]	19.2[b]
SS + CS 10 and 20 DAP	2290[b]	19.6[b]	2320[b]	20.3[b]
Uninoculated (Control)	2138[a]	17.6[a]	2150[a]	17.8[a]

Values within a column followed by the same letter are not significant at the 0.05 level.cell suspension one hour before sowing (SI); inoculation the soil beside the plants with 10 ml/plant with cell suspension (CS) at 10 or at 10 + 20 days after planting (DAP); seed slurry inoculation with composted cotton seed husk inoculum (SS) one hour before sowing; seed slurry followed by supplementary inoculation with 10 ml/Plant of the cell suspension beside the plants at 10 (SS + CS 10) or at 10 + 20 (SS + CS 10 and 20 DAP), and non-inoculated seeds as control.

Data in Table (3) show that, the inoculated plants contained much quantities of P and N as compared to non-inoculated one during 2002 and 2003 seasons. The increase in P and N-content was significant with the treatments of seed slurry plus supplementary inoculation of cell suspension at 10 or at 10 and 20 days after planting and those inoculated with cell suspension at 10 and 20 days from planting. While the increase in P and N-content resulted from the other treatments was not significant.

The data also indicated that there is a positive correlation between inoculation with phosphate dissolvers and N-content in the plants during the two seasons. Similar findings were also reported by Saber and Kabesh (1990), Datta *et al.* (1992) and Heggo and Barakah (1993).

Table (3): Effect of different inoculation methods by phosphate dissolvers on P and N content in straw and grain (Kg/fed) of maize plants.

Inoculation treatments	2002 season				2003 season			
	P		N		P		N	
	Straw	Grain	Straw	Grain	Straw	Grain	Straw	Grain
SI	5.7[a]	9.7[a]	41.2[a]	72.3[a]	5.9[a]	10.1[a]	41.3[a]	72.5[a]
CS 10 DAP	5.9[a]	9.8[a]	41.6[a]	72.4[a]	6.7[b]	10.2[a]	41.5[a]	72.5[a]
CS 10 and 20 DAP	6.2[b]	10.6[b]	41.7[a]	72.6[a]	6.5[b]	10.8[b]	42.0[a]	72.7[a]
SS	5.8[a]	9.8[a]	41.3[a]	72.3[a]	6.1[a]	10.3[a]	41.5[a]	72.6[b]
SS + CS 10 DAP	6.3[b]	11.3[b]	42.2[b]	73.1[b]	6.7[b]	11.5[b]	43.3[b]	73.2[b]
SS + CS 10 and 20 DAP	7.5[b]	11.7[b]	43.7[b]	73.6[b]	7.9[b]	12.1[b]	41.2[a]	73.5[b]
Uninoculated (Control)	5.4[a]	9.2[a]	40.4[a]	71.5[a]	5.6[a]	9.8[a]	41.2[a]	72.4[a]

cell suspension one hour before sowing (SI); inoculation the soil beside the plants with 10 ml/plant with cell suspension (CS) at 10 or at 10 + 20 days after planting (DAP); seed slurry inoculation with composted cotton seed husk inoculum (SS) one hour before sowing; seed slurry followed by supplementary inoculation with 10 ml/Plant of the cell suspension beside the plants at 10 (SS + CS 10) or at 10 + 20 (SS + CS 10 and 20 DAP), and non-inoculated seeds as control.

Results presented in Table (4) reveal that, the Nos. of phosphate dissolving bacteria in rhizosphere soil of maize increased gradually, reached their maxima after 60 days from sowing in all the inoculation treatments under investigation during the two seasons (2002 and 2003), and thereafter decreased slightly. The data also showed that the highest counts of phosphate-dissolving bacteria were recorded in the rhizosphere soil of seed slurry maize, which received supplemental cell suspension inoculum at 10 and 20 days after planting in both 2002 and 2003 seasons. The low numbers of phosphate-dissolving bacteria were recorded in the rhizosphere of non inoculated soil. Zaghloul *et al.* (1996) also found that, inoculation of wheat seeds with phosphate solubilizing bacteria gave the highest counts of phosphate dissolvers in the soil. However, the phosphate dissolving bacteria generally occur in abundance in the rhizosphere and non-rhizosphere area of different crops. They vary in the numbers according to the type of soil, soil pH and the type of cultivated crop (Martinez *et al.*, 1990 and Rokade and Patri (1992). In addition, Gupta *et al.* (1986) reported that, organic carbon content of the soil markedly influenced the numbers of phosphate dissolving bacteria. Grass and forest lands had larger populations than cultivated soils. Data presented in Table (5) show the amount of soluble phosphorus in the rhizosphere soil of maize during different growth phases as affected by varying the methods of inoculation with phosphate dissolving bacteria. The results indicate that inoculation of maize with phosphate dissolving bacteria increased the amount of soluble phosphorus in the rhizosphere soil of maize as compared to the non inoculated treatments.The increase was significant with the treatments of seed slurry plus supplementary cell suspension at 10 and 20 days after planting, while other inoculation treatments recorded non-significant increases. The maximum amounts of soluble phosphorus were reduced after 60 days from sowing in all treatments during the two seasons (2002 and 2003). The results also show positive correlation among the

amount of soluble P (Table 5) and the number of phosphate-dissolving bacteria (Table 4). However, the phosphate dissolving bacteria vary in their phosphate solubilizing ability (Alexander, 1977). In addition, the amounts of solubilizing phosphate were related to the amount of that form of phosphate present in the soil. The efficiency of bacterial strains in phosphate solubilization varied greatly with the form of inorganic phosphate (Martinez *et al.*, 1990).

Table (4): Log numbers of phosphate-dissolving bacteria (per gram) in rhizosphere soil of maize amended with rock phosphate during different growth phase as affected by variation of inoculation methods with phosphate dissolving bacteria.

Inoculation treatment	Season 2002					Season 2003				
	Days after inoculation									
	15	30	45	60	90	15	30	45	60	90
1	5.42	6.31	6.85	7.35	7.15	5.60	6.45	7.10	7.63	7.26
2	5.61	6.35	6.93	7.41	7.25	5.91	6.63	7.21	7.81	7.41
3	6.32	6.71	7.10	7.65	7.43	6.50	6.91	7.45	7.93	7.50
4	5.45	6.51	6.93	7.40	7.20	5.50	6.50	7.15	7.73	7.36
5	6.51	6.92	7.64	7.75	7.51	6.61	7.41	7.95	8.10	7.65
6	7.10	7.50	7.93	8.21	7.75	7.35	7.95	8.15	8.63	7.90
7	5.31	6.10	6.75	7.15	6.80	5.10	6.13	7.21	7.60	7.10

1=cell suspension one hour before sowing (SI);2= inoculation the soil beside the plants with 10 ml/plant with cell suspension (CS) at 10 or 3=at 10 + 20 days after planting (DAP); 4=seed slurry inoculation with composted cotton seed husk inoculum (SS) one hour before sowing; 5=seed slurry followed by supplementary inoculation with 10 ml/Plant of the cell suspension beside the plants at 10 (SS + CS 10) or 6= at 10 + 20 (SS + CS 10 and 20 DAP), and 7= non-inoculated grains as control.

Table (5): Amounts of soluble phosphorus in the rhizosphere soil of maize during different growth phase as affected by varying the methods of inoculation with phosphate dissolving bacteria.

Inoculation treatment	Season 2002					Season 2003				
	Days after inoculation									
	15	30	45	60	90	15	30	45	60	90
1	13.35	13.51^{a}	13.85^{a}	14.50^{a}	14.60^{a}	14.25	14.62^{a}	14.91^{a}	16.21^{a}	15.61^{a}
2	13.35	13.68^{a}	13.91^{a}	14.60^{a}	14.71^{a}	14.25	14.75^{a}	15.16^{a}	16.36^{a}	15.73^{a}
3	13.35	13.51^{a}	14.21^{a}	14.95^{b}	15.21^{b}	14.25	15.10^{b}	15.52^{b}	16.49^{a}	16.11^{b}
4	13.35	13.63^{a}	13.90^{a}	14.81^{a}	14.70^{a}	14.25	14.75^{a}	14.95^{a}	16.25^{a}	15.68^{a}
5	13.35	13.81^{a}	14.61^{b}	15.90^{b}	15.60^{b}	14.25	15.61^{b}	15.93^{b}	17.31^{b}	16.53^{b}
6	13.35	14.22^{b}	14.85^{b}	14.15^{b}	16.10^{b}	14.25	15.86^{b}	16.45^{b}	17.82^{b}	17.15^{b}
7 control	13.35	13.38^{a}	13.61^{a}	14.10^{a}	13.81^{a}	14.25	14.51^{a}	14.76^{a}	15.82^{a}	15.31^{a}

* Values within a column followed by the same letter are not significant at the 0.05 level.1=cell suspension one hour before sowing (SI); 2= inoculation the soil beside the plants with 10 ml/plant with cell suspension (CS) at 10 or 3= at 10 + 20 days after planting (DAP); 4=grain slurry inoculation with composted cotton seed husk inoculum (SS) one hour before sowing; 5=grainslurry followed by supplementary inoculation with 10 ml/Plant of the cell suspension beside the plants at 10 (SS + CS 10) or 6= at 10 + 20 (SS + CS 10 and 20 DAP), and 7= non-inoculated grains as control.

Generally, the present study indicated that, addition of rock phosphate and inoculation with phosphate dissolving bacteria increased dry matter, grain yield, N and P-uptake by maize as compared to those received rock phosphate alone without inoculation control. Inoculation also gave highest counts of phosphate dissolving bacteria in the rhizosphere zone of maize and enhanced the amounts of soluble phosphate in the rhizosphere soil of maize. Inoculation of maize seed (slurry) followed by supplementary inoculation of the cell suspension beside the plants at 10 and 20 days after planting recorded the best results as compared to other inoculation treatments. In addition, the laborator studies of soil-rock phosphate reactions have shown that the poor effectiveness of the rock phosphate is primarily due to limited dissolution of this fertilizer ((Gilkes and Bolland, 1992).

The present study show that application of rock phosphate and inoculation with efficient phosphate dissolving bacteria seed slurry plus supplemental suspension inoculation produced more amount of soluble phosphorus in the soil as compared to rock phosphate alone without inoculation, which leads to the conclusion that, the post emergence cell suspension inoculation with phosphate dissolving bacteria may be necessary particularly when rock phosphate use as phosphorus fertilizer.

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التحسين الحيوى لمحصول الأذرة ومحتواها من الفوسفور والنيتروجين كنتيجة لطرق التلقيح المختلفة باليكتيريا المذيبة للفوسفات

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[1و4] قسم البيئة والزراعة الحيوية - كلية الزراعة - جامعة الأزهر - القاهرة - مصر .

[2] قسم الميكروبيولوجيا الزراعية - كلية الزراعة - جامعة المنيا - المنيا - مصر .

[3] قسم الميكروبيولوجيا - معهد الأراضى والمياه والبيئة - مركز البحوث الزراعية - الجيزة - مصر .

فى الأونة الأخيرة إنتشرت أمراض العصر من سرطان وفشل كلوى وغيرها ومن ثم إتجهت الأنظار نحو الإستبدال ولو جزئيا للأسمدة الكيماوية ببدائلها من الأسمدة الحيوية أو ما يعرف بالمخصبات الحيوية ولذلك إستهدفت الدراسة فى هذا البحث إلى إستخدام طرق تلقيح مختلفة للبكتيريات المذيبة للفوسفور كمخصبات حيوية لنبات الأذرة ودراسة تأثيرها على كمية الحطب ومحصول حبوب الأذرة ومحتواها من عنصر الفوسفور والنيتروجين الحيويين لأى كائن حى ، وقد بينت الدراسة النتائج التالية :

١- تباينت قدرة العزلات المذيبة للفوسفات موضع الدراسة فى قدرتها على خفض pH بيئة النمو ومن ثم إختلفت قدرتها فى إذابة عنصر الفوسفور المرتبط ، وقد بينت العزلة رقم ١٥ قدرة عالية فى إذابة الفوسفور حيث بلغ معدل الإذابة ٤٢١,٩ جزء فى المليون .

٢- تم الحصول على أعلى كمية من المادة الجافة لحطب الأذرة وكذلك محصول الحبوب وذلك بتلقيح النباتات بالبكتيريات المذيبة للفوسفات بعد ١٠ ، ٢٠ يوم من الزراعة .

٣- تباينت طرق التلقيح فى معدل إذابة الفوسفور حيث تبين أن التلقيح بمعلق الخلايا المذيبة للفوسفور بعد ١٠ ، ٢٠ يوم من الزراعة أفضل من تلقيح البذور بعد ساعة من النقع .

٤- محتوى القش والحبوب من الفوسفور والنيتروجين قد زاد زيادة معنوية بتلقيح البذور بعد ١٠، ٢٠ يوم من نمو نبات الأذرة وكذلك للحبوب الملقحة بمعلق الخلايا بعد ١٠ ، ٢٠ يوم من الزراعة .

٥- تواجدت علاقة إيجابية بين التلقيح بمذيبات الفوسفور ومحتوى النبات من النيتروجين .

٦- تم الحصول على أعلى الأعداد من البكتيريا المذيبة للفوسفور فى منطقة ريزوسفير الأذرة بعد ٦٠ يوما من النقع فى كل المعاملات حيث زادت أعداد هذه البكتيريات زيادة تدريجية حتى ٦٠ يوم ثم تناقصت فيما بعد .

٧- أدى التلقيح لنبات الأذرة باليكتيريات المذيبة للفوسفات إلى زيادة كمية الفوسفور الذائب فى منطقة ريزوسفير نبات الأذرة مقارنة بالأرض الغير ملقحة .

٨- كانت الزيادة فى محتوى الفوسفور معنوية فى معاملات البذور الملقحة بمعلق الخلايا المذيبة للفوسفور بعد ١٠ ، ٢٠ يوم من الزراعة .

ولذلك تبين هذه الدراسة أن إستخدام صخر الفوسفات واليكتيريا الفعالة فى إذابة الفوسفور يؤدى إلى إنتاج كمية عالية من الفوسفور الذائب فى التربة والذى يلعب دورا هاما فى العمليات الحيوية داخل أى كائن حى ومن ثم يزيد المحصول الناتج .