

EFFECT OF MINERAL AND BIO-FERTILIZATION ON NPK AVAILABILITY, UPTAKE, AND MAIZE YIELD.

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ABSTRACT

Two field trials were conducted during the two successive seasons 2007-2008 on maize plants at the experimental farm of Sakha Agric. Res. Station, Kafr El-Sheikh Governorate, Egypt. The aim of this study was to investigate the influence of maize grain treated with *Azotobacter chroococcum* and *Bacillus megatherium* and their combinations with NPK under three N-levels; 80, 100, and 120 kg N/fad for N1, N2, and N3 to increase yield, NPK uptake by maize plants and the availability of NPK in the soil. The experiments were conducted in split plot design, with three replicates.

The observed results can be summarized as follows:

- The yield and components of maize, NPK uptake and the availability of NPK in the studied soil were affected significantly by biofertilizer treatments and N-levels.
- The maximum values of available N were obtained by the application of *Azotobacter chroococcum* and *Bacillus megatherium* with NPK under N3 treatment. The maximum values of available P were recorded by the application of *Bacillus megatherium* with NPK under N3 treatment.
- Combination of *A.chroococcum*, *B.megatherium*, and NPK fertilizers under N3 significantly increased grain yield (20.9 and 17.9%), straw yield (16.8 and 20.6%), 100-grain weight (9.9 and 13.3%), ear weight (21.0 and 18.0) and N, P, and K uptake by maize grain [(35.1 and 31.3%), (21.2 and 26.9%) and (18.1 and 31.1%)] over the control for N, P, and K, respectively, in 2007 and 2008 seasons. While shelling percentage did not significantly affected by different treatments. The application of these results should help in reducing environmental pollution.

Keywords: maize (*Zea mays* L), biofertilizer, *Azotobacter chroococcum*, *Bacillus megatherium*, mineral fertilization.

INTRODUCTION

Maize (*Zea mays* L) among the crops is an important in arid and semiarid regions, because of increasing demand for food and livestock feed. In Egypt, the annually cultivated area with maize is about 1.5- 2.0 million faddan. Thus a great attention should be paid to raise its productivity per unite urea. For optimum plant growth, nutrients must be available in sufficient and balanced quantities. Nitrogen and phosphorus are essential nutrient for plant growth and development in maize (Wua, *et al.*, 2005). Large quantities of chemical fertilizers are used to replenish soil N and P resulting in high costs and several environmental contaminations (Dai *et al.*, 2004). Thus, increased attention is now being paid to develop and integrate plant nutrients system that maintains or enhances soil productivity through balanced use of all sources of nutrients, including chemical fertilizers and biofertilizers. Biofertilizers are products containing living cells of different types of microorganisms that have an ability to mobilize nutrients from insoluble form through biological processes and these groups of microorganisms may either

fix atmospheric nitrogen or solubilizing the insoluble phosphorous making them available for crops (Prabhakar and Singh., 2008). The contribution of non-symbiotic nitrogen fixing microorganisms to the supply of fixed nitrogen in agricultural soils and natural ecosystems is well recognized. Microorganisms including *Azotobacter* as non-symbiotic nitrogen fixers as well as *Bacillus megatherium* a phosphate – solubilizing bacteria (PSB) are continuously being isolated from various ecosystems and their performance in the laboratory and field conditions are assessed. Many experiments in greenhouses and in field conditions have shown that several crops respond positively to microbial inoculation. Enhancement and maintenance of soil fertility through microorganisms will be an important issue in future agriculture. Hence, several beneficial microorganisms can effectively be used as a chemical fertilizer alternatively to minimize the application of inorganic fertilizers. *Azotobacter chroococcum* was used previously in increasing plant parameters (Ahmad *et al.*, 2004 and Yasmin *et al.*, 2007). Yazdani *et al.*, 2009 reported that inoculation with rhizobacteria can be efficiently used to improve growth and grain yield of corn, reduce fertilizer costs and reduce leaching of NO_3^- to ground water as well as reducing emission of the green house gas N_2O . They concluded too that the application of N_2 -fixing and P-solubilizing bacteria could reduce P application by 50 % without any significant reduction of maize grain yield. However, this treatment could not compensate 50% of N application. Chandrasekar *et al.*, 2005 found that both morphology and yield parameters produce a better results during the combination of biofertilizers and chemical fertilizers than using either method alone.

The aim of the present study was to evaluate the effect of maize grain treatment with *Bacillus megatherium* as Phosphorus Solubilizing Bacteria and *Azotobacter chroococcum* as non-symbiotic nitrogen fixer bacteria in presence of NPK soil fertilization under three N levels on yield and components, NPK uptake by maize plants and the availability of NPK in the soil.

MATERIALS AND METHODS

Series of laboratory and filed experiments were carried out at Faculty of Agriculture, Kafr El-Sheikh University and Sakha Agric. Res. Station, to study the relationship between the laboratory data and crop production.

a-Laboratory experiments:

• Microorganisms isolation and Identification:

Azotobacter chroococcum as non-symbiotic nitrogen fixer bacteria was isolated and identified in previous study (Omar and Belal 2007). Phosphate – solubilizing bacteria (PSB) was isolated in this study from rhizosphere of maize plants according to Yasmin *et al.*, (2007). Identification of grown isolated colonies based on morphological, biochemical and culturing characteristics were identified according to Parry *et al.*, (1983).

• **Effect of pH and temperature on *Bacillus megatherium* growth:**

100ml Pikovskaia's liquid medium were used to determine the effect of temperature and pH on growth of *Bacillus megatherium*. This medium was inoculated by 1ml (10^8 cfu/ml) of culture of *Bacillus megatherium* isolate. The experiments were carried out at pH 6, 7 and 8 and the culture was incubated at 30°C and 150 rpm for 3 days. To determine the optimum temperature Pikovskaia's broth medium at pH7, cultures were incubated at 20, 30 and 40°C and 150 rpm for 3 days. The growth was determined as intracellular protein content ($\mu\text{g/ml}$) for bacterial isolates after 3days according to Lowry et al., (1951), where the bacterial cells were digested as described by Belal (2003).

• **Application of grain treatments with *Azotobacter chroococcum* and *Bacillus megatherium***

Azotobacter chroococcum and *Bacillus megatherium* were applied at the time of planting as grain treatment. Grains were immersed in each bacterial suspension (10^8 cfu/ml) for 30min. and then air dried. Grains were then sown in the soil.

• **Effect of different treatments on total microbial count in maize rhizosphere plants**

The total microbial count in rhizosphere of untreated and treated soils with *Azotobacter chroococcum* and *Bacillus megatherium* strains in combination of different chemical fertilizers was counted three times after 30, 60 and 90days during the growing season by using dilution series on standard-plate count agar.

b- Field experiments:

Two field experiments were conducted at the experimental farm of Sakha Agric. Res. Station during the two successive seasons of 2007 and 2008 using maize grain (*Zea mays* L) Giza 352. The experiments were conducted in split-plot design with three replicates. The main plots were to N-treatment ;80, 100, and 120 (the recommended dose) Kg N/ fad for N1, N2, and N3. These treatments were 66, 83, and 100% of the recommended doses, respectively as urea 46%N), the sub-plots were to bio fertilizers treatments (grain bacterial inoculated). The treatments were, without inoculation (T1), inoculated with *Bacillus megatherium* (T2), inoculated with *azotobacter chroococcum* (T3), and inoculated with *Bacillus megatherium* and *Azotobacter chroococcum* (T4). All experiment plots were treated with 30 Kg P_2O_5 / fad as superphosphates 15% P_2O_5 at sowing and 24 Kg K_2O / fad as potassium sulphate (48% K_2O) before 2nd irrigation. The maize plants were harvested at 20th 2007 and 25th 2008 of Sep, grain and straw yield were determined after maturity and weighed at 15% moisture content. Grain and straw samples were taken and dried in an oven at 70 C for 48 hours. Dry sample was digested by wet digesting (Jackson, 1967). N, P, and K were determined in the digested plant materials.

Soil surface samples (0- 15 cm) were collected from the treated plots after maize harvesting. The collected soil samples were air-dried and prepared for chemical analysis. Available nitrogen was extracted by K-sulphate and determined using the microkjeldahl method according to

(Jackson, 1967). Available phosphorus was extracted by using NaHCO_3 according to Olsen, (1954) and then determined spectrophotometrically according to Jackson, (1967). Available potassium was determined by using flame photometer in ammonium acetate extract, according to Jackson, (1967). The data were subjected to statistical analysis according to Snedocor and Cochran, (1980). The soil characteristics of experiment location are presented in Table 1.

Table (1): Some chemicals and physical properties of the soil surface layer (0-30 cm) before planting

Characteristics	Values
pH (1:2.5 soil : water suspension)	7.52
EC, dSm^{-1}	3.20
OM%	1.92
Available nutrients, mg/ Kg soil:	
N	22
P	5.8
K	415
Particle size distribution%:	
Clay %	52.00
Silt %	23.9
Sand %	24.1
Texture class	Clay

RESULTS AND DISCUSSION

Isolation and identification of the organisms

An evaluation of different rhizosphere maize plants were used to isolate the phosphate – solubilizing bacteria (PSB) in Pikovskaia's medium (Yasmin *et al.*, 2007). In the present work they were collected from different locations in Kafr El-Sheikh Governorate, Egypt, resulting in isolation of one bacterial isolate and tentatively was identified as *Bacillus megatherium* on the basis of morphological and physiological behavior as described in Parry *et al.*, (1983).

Effect of pH and temperature on growth of *Bacillus megatherium*.

A phosphate – solubilizing bacteria (PSB) depends on the prevailing environmental conditions such as pH value and temperature in the soil and biological components, including all root-colonizing plant-beneficial bacteria and fungi.

Optimum pH value:

The influence of pH on growth of *Bacillus megatherium* (EB2) is shown in Fig (1). Generally, pH 7 was the optimal pH for *Bacillus megatherium* growth. The maximum of intracellular protein content ($\mu\text{g/ml}$) was recorded at pH7. Most of the bacterial strains are known to prefer the neutral pH. The measured soil pH and in water samples had no obvious effect on *Bacillus megatherium* in the present work. This bacterial strain can grow at range from pH 6-8.

Therefore, it can be deduced from the results that the pH is considered an important environmental factor in the rhizosphere which affect on *Bacillus megatherium* efficiency.

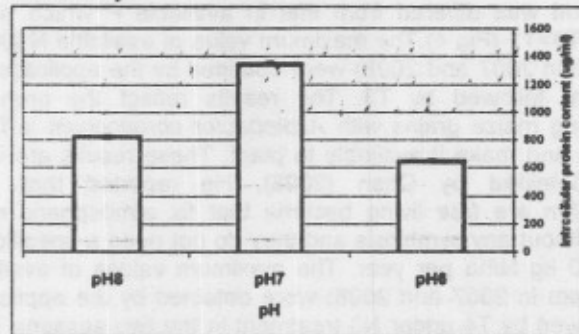


Fig.(1). Effect of pH value on growth of *Bacillus megatherium*.

Optimum temperature:

The effect of different temperature degrees on growth of *Bacillus megatherium* are shown in Fig 2. A temperature 30°C appears to be the optimum for growth of *Bacillus megatherium*. The prices of nitrogen and phosphorus fertilizers have nearly doubled during the last 3-4 years. This has necessitated to search for cheaper source of nitrogen and phosphorus to meet the needs of crops. The species of *Azotobacter* are known to fix naturally atmospheric nitrogen in the rhizosphere on an average of 30% contributing towards the nitrogen availability for crop plants. *Azotobacter chroococcum* was isolated used previously as a biofertilizer for improvement and increasing mango crop (Ahmad *et al.*, 2004). These results are in agreement with our previous findings on both optimal growth conditions (Omar and Belal 2007 and Belal *et al.*, 2008)

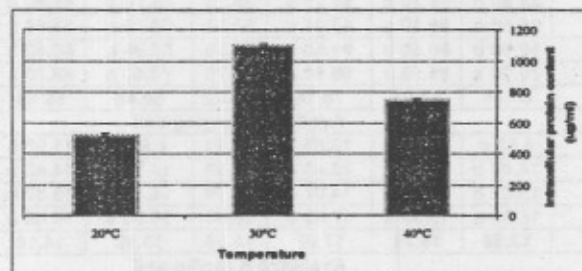


Fig.(2). Effect of temperature on growth *Bacillus megatherium*.

Soil available of N, P, and K:

Data presented in Table (2) show that the application of biofertilizer treatments under N-levels significantly increased the availability of P and N in the soil compared with the control (NPK).

T1, T2, T3, and T4 gave the highest values of available N and P under N3 in the two seasons. Fig (3) show that available N in the soil as affected by biofertilizer treatments were in this order: T4>T3>T2>T1 in the two seasons. This trend was differed from that in available P which were in the order: T2>T4>T3>T1 (Fig 4). The maximum value of available N (98.8 and 112.7 mg N/kg soil in 2007 and 2008) were obtained by the application of T4 under N3 treatment followed by T3. The results reflect the pronounced effect of inoculating maize grains with *Azotobacter chroococcum* in fixing atmospheric nitrogen and make it available to plant. These results are in agreement with those obtained by Chen (2008). He reported that *Azotobacter* and *Azospirillum* are free living bacteria that fix atmospheric nitrogen in cereal crops without any symbiosis and they do not need a specific host plant, it can fix 15-20 kg N/ha per year. The maximum values of available P (23.4 and 24.67 ppm in 2007 and 2008) were obtained by the application of T2 under N3 followed by T4 under N3 treatment in the two seasons. These results are in agreement with those obtained by Chen (2008), and Yazdani *et al.*, (2009). They reported that phosphobacterins can make insoluble phosphorus available to the plant. The solubilization effect is generally due to the production of organic acid that lower the soil pH and bring about the dissolution of bond forms of phosphate.

Available K had no significant effect with biofertilizer treatments as well as N-level in the first season, while in the second season the values of available K was affected significantly with biofertilizers and N-level. The maximum values of available K (563.4 and 563.2mg/kg soil in 2007 and 2008 seasons) were obtained by applying T4 under N3.

Table (2) Effect of N-levels and biofertilizers treatments on available of N, P, and K in the soil

Treatments	2007				2008			
	N1	N2	N3	Mean	N1	N2	N3	Mean
Available N mg/kg soil								
T1	29.80 d	56.70 d	59.28 d	48.59	35.70 d	49.80 d	56.50 d	47.33
T2	38.50 c	59.30 c	63.07 c	53.62	39.50 c	58.90 c	68.60 c	55.67
T3	62.90 b	80.50 b	91.50 b	78.30	58.90 b	68.60 b	78.40 b	66.97
T4	70.20 a	91.28 a	98.80 a	86.76	72.50 a	88.20 a	112.70 a	91.13
Mean	50.35	71.95	78.16	66.82	50.40	66.38	79.05	65.28
Available P mg/kg soil								
T1	9.60 c	12.13 b	12.83 c	11.52	8.80 c	11.90 b	12.50 c	11.07
T2	14.40 b	15.16 a	23.43 a	17.66	15.73 b	14.40 b	24.67 a	18.27
T3	13.87 b	13.40 b	14.07 c	13.78	14.40 b	13.87 b	14.87 b	14.71
T4	16.39 a	16.33 a	17.93 b	16.89	15.80 a	16.46 a	20.07 b	17.44
Mean	13.56	14.26	17.07	14.96	13.68	14.16	18.27	15.38
Available K mg/kg soil								
T1	497.53 a	417.10 a	466.87 b	460.50	447.00 b	405.60 b	461.00 c	437.87
T2	510.67 a	419.67 a	541.73 ab	490.69	517.20 a	430.87 b	489.90 b	479.32
T3	499.40 a	415.47 a	490.13 ab	468.33	461.00 b	405.60 b	451.00 c	439.20
T4	438.40 a	472.47 a	563.40 a	491.42	447.00 b	443.00 a	563.20 a	484.40
Mean	486.50	431.18	515.53	477.74	468.05	421.27	491.28	460.20

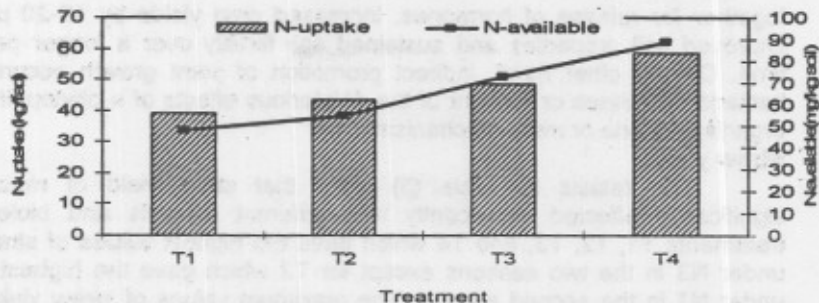


Fig. 3. Effect of biofertilizers treatments on N-uptake of maize grain (kg/fad) and available N in the soil (mg/kg soil) (average of the two seasons).

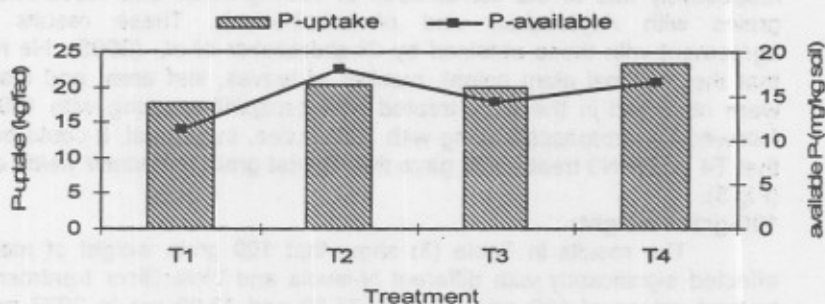


Fig. 4. Effect of biofertilizer treatments on P-uptake of maize grain (kg/fad) and available P in the soil (mg/kg soil) (average of the two seasons).

Yield and yield components:

Grain yield:

The results in Table (3) show that the grain yield of maize was affected significantly by different N-levels and biofertilizers treatments. T1, T2, T3, and T4 treatments gave the highest values of grain yield under N3 (the recommended dose) in the first season. On the other hand, in the second season T2 and T3 treatments gave the highest values under N1 (66% of the recommended dose). Meanwhile, T1 and T4 treatments gave the highest values under N3 exhibit the same trend as in the previous season. The maximum values of grain yield; 4443 and 4429 kg/fad in 2007 and 2008 seasons, respectively were obtained by the application of T4 treatments under N3. The grain yields were positively increased by about 20.9 and 17.9% in 2007 and 2008, respectively due to the combination of biofertilizers and 120 kg N/fad. The obtained data by Prabhakar and Singh (2008) and Anjum *et al.*, (2007), supported these results. They reported that, the response of field crops to inoculating with *Azotobacter* and *phosphobacteria*

together for release of hormones, increased crop yields by 10-20 percent, improved soil properties and sustained soil fertility over a longer period of time. On the other hand, indirect promotion of plant growth occurs when bacteria decreases or prevent of the deleterious effects of a phytopathogenic organism by one or more mechanisms.

Straw yield:

The results in Table (3) show that straw yield of maize was significantly affected significantly with different N-levels and biofertilizers treatments T1, T2, T3, and T4 which gave the highest values of straw yield under N3 in the two seasons except for T2 which gave the highest values under N1 in the second season. The maximum values of straw yield; 6604 and 6470 kg/fad in 2007 and 2008 seasons were obtained by the application of T4 under N3 treatment in the two seasons. The straw yields were increased by about 16.76 and 20.6% over the control (T1) in 2007 and 2008, respectively due to the combination of 120 kg N/fad and inoculating maize grains with *Azotobacter* and *phosphobacteria*. These results are in agreement with those obtained by Chandrasekar *et al.*, (2005). He revealed that the maximal plant height, number of leaves, leaf area, and leaf length were observed in the plots treated with *azospirillum* along with 100% urea followed by *Azotobacter* along with 100% urea. In general, it could be stated that T4 under N3 treatments gave the highest grain and straw yields of maize (Fig 5).

100-grain weight:

The results in Table (3) show that 100 grain weight of maize was affected significantly with different N-levels and biofertilizer treatments. The highest values of 100 grain weight; 37.00 and 33.09 gm in 2007 and 2008 were obtained by the application of T2 under N2 treatment in the first season and T4 under N1 treatment in the second season, respectively. The 100-grain weight was increased by about 9.9 and 13.3% over the control (T1) in 2007 and 2008 seasons, respectively due to the application of biofertilizer treatments with NPK under N3.

Ear weight:

The results in Table (3) show that the ear weight of maize was significantly affected with different N-levels and biofertilizer treatments, the highest values of ear weight; 222.17 and 221.17 gm in 2007 and 2008, respectively were obtained by the application of T4 under N3 treatment in the two seasons. Ear weights were positively increased by about 21.0 and 18.0% due to the combination of biofertilizer treatments and NPK. These results are supported by Yazdani *et al.*, (2009). They found that using of phosphate solubilization and fixed nitrogen microorganisms in addition to conventional fertilizer applications (NPK) could improve ear weight, and grain number per row and ultimately increased grain yield.

Shelling percentage:

The results in Table (3) show that the shelling percentage did not significantly affected by N-levels and biofertilizer treatments. T4 under N3 treatments showed the highest value of shelling percentage; 85.5%.

Table (3) Effect of N-levels and biofertilizers treatments on grain and straw yields, 100-grain weight, ear weight, and shelling percentage in the two seasons

Treatments	2007				2008			
	N1	N2	N3	Mean	N1	N2	N3	Mean
Grain yield, kg/fad								
T1	2927 d	3555 c	3665 b	3382.33	3155 b	3480 b	3887 b	3507.56
T2	3217 c	3621 bc	3758 b	3532.00	3918 a	3793 b	3669 b	3793.33
T3	3622 b	3778 b	3860 b	3753.33	3834 a	3636 b	3704 b	3724.89
T4	4000 a	4385 a	4443 a	4276	4132 a	4258 a	4429 a	4273.00
Mean	3441.5	3834.75	3931.5	3735.92	3759.92	3791.92	3922.25	3824.69
Straw yield, kg/fad								
T1	4390 c	5350 c	5497 c	5497 c	4624 c	5200 c	5200 c	5014.67
T2	4852 b	5020 d	5621 bc	5621 bc	5500 b	5100 c	5400 c	5333.38
T3	4758 b	5667 b	5790 b	5790 b	5375 b	5818 b	5866 b	5686.33
T4	5940 a	6470 a	6604 a	6604 a	6120 a	6360 a	6470 a	6316.67
Mean	4985.00	5626	5626.75	5878.00	5404.75	5624.50	5734.00	5587.75
100-grain weight (g)								
T1	28.76 b	30.19 b	35.20 a	31.38	27.36 c	26.03 c	30.19 b	27.86
T2	30.11 b	37.00 a	35.14 a	34.08	29.55 c	31.41 a	28.55 b	29.94
T3	31.23 ab	35.75 a	35.87 a	34.28	30.59 b	31.49 a	32.06 a	31.38
T4	33.68 a	36.14 a	34.61 a	34.81	33.09 a	30.83 b	32.48 a	32.13
Mean	30.95	34.77	35.21	33.64	30.14	29.94	30.82	30.82
Ear weight(g)								
T1	146.33 d	177.77 b	183.27 b	169.12	157.8 c	174.00 c	194.37 b	175.39
T2	160.83 c	181.03 bc	187.90 b	179.59	195.90 ab	189.00 b	181.17 b	188.69
T3	181.10 b	188.90 b	193.00 b	187.67	191.70 b	198.50 b	184.50 b	191.57
T4	200.00 a	219.27 a	222.17 a	213.81	206.6 a	212.90 a	221.17 a	213.56
Mean	172.07	191.74	196.58	186.10	188.00	193.6	195.20	192.30
Shelling percentage								
T1	80.85 a	81.90 ab	84.53 a	82.23	80.22 a	82.30 a	85.23 a	82.58
T2	81.01 a	84.50 a	83.20 a	82.91	81.25 a	85.30 a	82.40 a	82.98
T3	82.50 a	80.80 b	84.50 a	82.60	81.24 a	79.98 a	83.22 a	81.48
T4	83.50 a	84.50 a	85.50 a	84.50	82.50 a	84.62 a	85.50 a	84.21
Mean	81.82	82.93	84.40	83.50	81.30 a	83.05	84.09	82.81

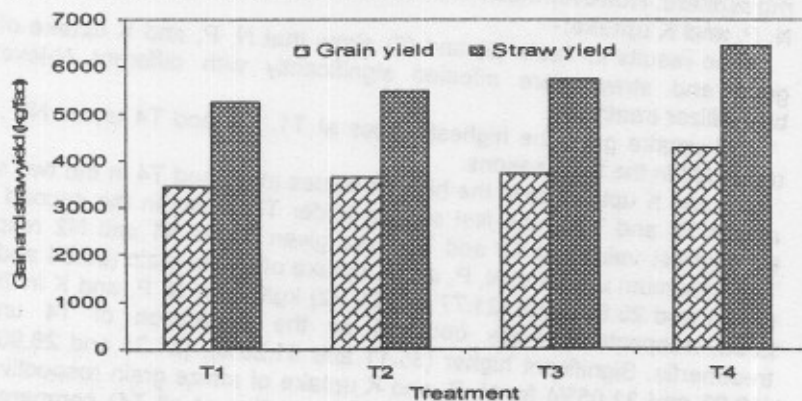


Fig. 5 Effect of biofertilizers treatments on grain and straw yield of maize (average of two seasons).

Table (4) Relative increase (Δ%) of maize grains due to bio-fertilizer inoculation

Treatment	2007			2008		
	N1	N2	N3	N1	N2	N3
T2	9.0%	2%	2%	20%	8%	-
T3	19%	6%	5%	18%	4%	-
T4	27%	19%	18%	24%	18%	12.2%

For example Δ increase of maize grain as a result of *Bacillus megatherium* (T2, under N1 treatment) can be calculated as following:

$$(\Delta\%) = \frac{\text{Grain yield (T2 under N1)} - \text{Grain yield (T1 under N1)}}{\text{Grain yield (T2 under N1)}} \times 100 = 9.0\%$$

Relative effect of biofertilizer in maize grain

Data in Table (4) show that by the increasing of N-levels fertilizer, the activity of soil bacteria decreased, consequently biofertilizer sharing percent on increasing the grain yield reduced. For example in (2007) the relative increase % of maize grain due to *Bacillus megatherium* inoculation under N1 was 9.0% then decreased to 2.0% under N3, the corresponding values for *Azotobacter chroococcum* were 19.0% under N1 and 5.0% under N3. Also these values for the combination of *Bacillus megatherium* and *Azotobacter chroococcum* were 27.0% under N1 and 18.0% under N3. These results were supported by Chen (2008). He reported that the application of chemical fertilizers can result in negative effects such as destruction of microorganisms and friendly insects.

In the comparison with T4 treatments at N1, N2, and N3, we can see that at N3 (the recommended dose) the maize grain yield increased only by 8.3 and 2.6% over that at N2 (66% of the recommended dose) and N1 (83% of the recommended dose), respectively (mean of two seasons). This saved 33 and 17% of nitrogen fertilizer, which consequently decreases environment pollution, health hazard, and fertilizer costs and benefits microflora in the rhizosphere. However much research is still needed.

N, P, and K uptake:-

The results in Table (5) and (6) show that N, P, and K uptake of maize grain and straw were affected significantly with different N-levels and biofertilizer treatment.

N uptake gave the highest values at T1, T2, and T4 under N3, and T3 under N2 in the two seasons.

P and K uptake gave the highest values at T1 and T4 in the two seasons and at T2 and T3 in the first season under T3. While in the second season the highest values at T2 and T3 were given under N1 and N2 respectively. The maximum values of N, P, and K uptake of maize grain (66.64 and 60.68), (22.21 and 25.58), and (21.77 and 22.92) kg/fad for N, P, and K in 2007 and 2008, respectively were obtained by the application of T4 under N3 treatments. Significant higher (35.11 and 31.25%), (21.24 and 26.95%), and (18.07 and 31.05%) for N, P, and K uptake of maize grain respectively were recorded by the application of T4 (mean value of all T4) compared to the control in 2007 and 2008 seasons. These increments can be explained as follows, *Azotobacter chroococcum* and *bacillus megatherium* can fix the

atmospheric free nitrogen or increase the availability of phosphorus in the soil, respectively and hence make the nitrogen and phosphorus in easier form for maize plant in rhizosphere. These results were supported by the data obtained by Ghulam *et al.*, (2007) and Davison (1988). They reported that biofertilizers producing a compound to the plant that is synthesized by bacterium or facilitating the uptake of nutrient from environment.

Table (5) Effect of N- level and biofertilizers on N, P, and K uptake by maize grain

Treatments	2007				2008			
	N1	N2	N3	Mean	N1	N2	N3	Mean
N-uptake, kg/fad								
T1	34.39 d	39.10 d	44.34 d	39.28	38.91 d	40.96 c	45.87 bc	39.24
T2	38.60 c	45.99 c	47.73 c	44.11	42.12 c	43.24 c	43.29 c	42.88
T3	47.44 b	51.76 b	50.57 b	49.92	46.31 b	48.43 b	47.04 b	47.26
T4	54.80 a	60.16 a	66.64 a	60.53	56.37 a	54.20 a	60.68 a	57.08
Mean	43.81	49.85	58.32	48.46	43.93	46.71	49.22	46.62
P-uptake, kg/fad								
T1	14.64 d	16.35 c	19.42 b	16.80	15.62 c	18.44 c	21.38 bc	18.48
T2	17.69 a	19.91 a	19.92 b	19.17	23.31 a	21.24 b	20.18 c	21.57
T3	19.56 b	17.38 b	19.20 b	18.74	19.28 b	21.93 b	21.85 b	21.02
T4	21.60 a	20.17 a	22.21 a	21.33	24.79 a	25.55 a	25.58 a	25.30
Mean	18.37	18.45	20.21	19.01	20.75	21.79	22.25	21.60
K-uptake, kg/fad								
T1	12.88 c	13.51 d	17.96 b	14.78	11.51 d	15.31 c	18.27 b	15.03
T2	14.77 b	14.85 c	15.03 c	14.88	20.37 b	18.09 b	17.24 bc	18.57
T3	16.66 a	16.25 b	17.75 b	16.89	16.49 c	20.25 a	16.30 c	17.68
T4	14.82 b	17.54 a	21.77 a	18.04	22.31 a	19.16 ab	22.92 a	21.80
Mean	14.78	15.54	18.13	16.15	17.67	18.20	18.93	18.27

Table (6) Effect of N-levels and biofertilizers treatments on N, P, and K uptake by maize straw the two seasons.

Treatments	2007				2008			
	N1	N2	N3	Mean	N1	N2	N3	Mean
N-uptake, kg/fad								
T1	17.21 c	26.75 c	31.88 c	25.28	18.12 c	29.23 c	29.12 d	25.49
T2	14.56 d	25.10 d	32.60 c	24.09	19.74 b	28.56 c	30.78 c	26.36
T3	18.56 b	34.24 b	39.37 b	30.64	21.07 b	34.45 b	35.20 b	30.24
T4	29.11 a	38.24 a	45.57 a	37.63	30.23 a	37.39 a	44.00 a	37.21
Mean	19.86	31.02	37.36	29.41	22.29	32.41	34.77	29.82
P-uptake, kg/fad								
T1	16.32 a	12.19 d	18.96 a	15.83	10.68 d	11.37 d	13.00 c	11.68
T2	11.40 b	14.20 c	15.45 b	13.68	12.98 c	13.87 b	13.76 b	13.53
T3	10.56 c	15.69 b	13.03 c	13.09	14.24 b	12.90 c	13.20 bc	13.45
T4	16.15 a	18.76 a	15.43 b	16.78	18.36 a	19.47 a	14.56 a	17.46
Mean	13.61	15.21	15.72	14.85	14.06	14.40	13.63	14.03
K-uptake, kg/fad								
T1	101.62 b	107.27 c	123.68 b	110.86	103.81 a	127.29 a	109.46 b	116.85
T2	100.92 b	106.58 c	115.79 c	107.76	138.08ab	107.46 b	109.89 b	118.48
T3	102.44 b	127.34 b	122.43 b	117.37	137.06 b	134.78 a	132.54 a	134.79
T4	128.48 a	150.95 a	142.00 a	140.48	144.13 a	134.01 a	110.99 b	129.71
Mean	108.37	123.03	125.96	119.12	130.77	128.38	115.72	124.96

Fig (3 and 4) show that N and P uptake by maize grain were in line with available N and P in soil.

This trend in nutrients uptake by maize grain among the treatments was the same for nutrients uptake of maize straw (Table 6). The observed reduction in N uptake by maize straw could be explained by Chandrasekar (2005), who concluded that the developing grains utilize nitrogen from the vegetative parts for the synthesis of storage and non storage grain proteins. As a consequence, the nitrogen content of vegetative parts decreased after the formation of spikes.

Effect of different treatments on total microbial count in maize rhizosphere plants

The total microbial flora in the rhizosphere of soil treated with *Azotobacter chroococcum* and *Bacillus megatherium* increased gradually as the age of plant increased compared with non-treated soil (Table 7). Similar results were obtained by Badr El-Din and Sahab (1986) and El-Nady and Belal (2005). The total rhizosphere microorganisms were increased by inoculation with *Azotobacter chroococcum* and *Bacillus megatherium*. This can be explained on the fact that *Azotobacter chroococcum* and *Bacillus megatherium* can fix the atmospheric free nitrogen or increase availability of phosphorus in the soil, respectively and hence make the nitrogen and phosphorus in easier form for the maize plant and other microorganisms in the rhizosphere. Also, *Azotobacter chroococcum* produce growth promoting substances such as indole acetic acid which encourage microbial population in the rhizosphere (El-Mahrouk and Belal 2007). Grain inoculation with the used bacterial strains leads to increasing the total microbial counts and this may be due to the population of the both kind of bacteria which increased in the plant rhizosphere.

Table (7): Total microbial flora in the rhizosphere of maize plants inoculated and non-inoculated with *Azotobacter chroococcum* and *Bacillus megatherium* during different growth periods.

Treatments	Total microbial counts (cfu/gm dry soil)		
	After 30 days	After 60 days	After 90 days
N1 + Az + B	1 X 10 ⁸	3 X 10 ⁸	6 X 10 ¹⁰
N1 + Az	8 X 10 ⁵	3 X 10 ⁷	6 X 10 ⁹
N1 + B	9 X 10 ⁴	3 X 10 ⁶	6 X 10 ⁸
N1	1 X 10 ³	3 X 10 ⁴	6 X 10 ⁵
N2 + Az + B	3 X 10 ⁸	1 X 10 ⁸	6 X 10 ⁹
N2 + Az	2 X 10 ⁵	3 X 10 ⁶	7 X 10 ⁷
N2 + B	5 X 10 ⁴	1 X 10 ⁶	2 X 10 ⁷
N2	1 X 10 ³	3 X 10 ⁴	1 X 10 ⁵
N3 + Az + B	5 X 10 ⁸	8 X 10 ⁷	6 X 10 ⁹
N3 + Az	2 X 10 ⁵	1 X 10 ⁷	2 X 10 ⁸
N3 + B	6 X 10 ⁴	7 X 10 ⁶	5 X 10 ⁷
N3	1 X 10 ³	3 X 10 ⁴	6 X 10 ⁵

N1 = 80 kg N/fad as urea (66% of the recommended dose)

N2 = 100 kg N/fad as urea (83% of the recommended dose)

N3 = 120 kg N/fad as urea (100% of the recommended dose)

Az = *Azotobacter chroococcum* B = *Bacillus megatherium*

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تأثير التسميد المعدني والحيوي على صلاحية وامتصاص النيتروجين و الفوسفور والپوتاسيوم ومحصول الذرة
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أجريت تجربتان حقليتين في المزرعة البحثية بسخا محافظة كفر الشيخ خلال الموسمين المتعاقبين ٢٠٠٧ و ٢٠٠٨.

الهدف من البحث دراسة تأثير تلقيح بذور الذرة (جيزة ٣٥٢) ببكتريا *Azotobacter chroococcum* (المثبتة للنيتروجين) و *Bacillus magetherium* (المثبتة للفوسفور) أو كلاهما معا مع وجود التسميد المعدني (نيتروجين وفوسفور وبوتاسيوم) تحت ثلاث مستويات من التسميد النيتروجيني (٨٠ و ١٠٠ و ١٢٠ كجم نيتروجين / فدان وهي الجرعة الموصى بها) على تحمسين الذرة ومكوناته وامتصاص الحبوب والقش من النيتروجين والفوسفور والبوتاسيوم ومحتوى الأرض الميسر منها وقد أقيمت التجريبتين في تصمم قطع منشقة مرة واحدة مع ثلاث مكررات ويمكن تلخيص النتائج كما يلي :-

- محصول الذرة ومكوناته وامتصاص حبوب وقش الذرة من عناصر النيتروجين و الفوسفور و البوتاسيوم وكذلك محتوى التربة الميسر من هذه العناصر تأثرت معنويا بالتسميد الحيوي ومستويات التسميد النيتروجيني .

- أعلى قيمة للنيتروجين الميسر كانت عند التسميد الحيوي بكل من البكتريا المثبتة للنيتروجين والمثبتة للفوسفور وفي وجود التسميد المعدني تحت مستوى النيتروجين N3 بينما أعلى قيمة الفوسفور الميسر كانت عند التسميد الحيوي بالبكتريا المثبتة للفوسفور فقط مع وجود التسميد المعدني تحت مستوى النيتروجين الثالث.

- التسميد المعدني مع البكتريا المثبتة للنيتروجين والمثبتة للفوسفور تحت مستوى N3 أدى إلى زيادة معنوية في كل من محصول الحبوب (٢٠٠٩ و ١٧,٩) ومحصول القش (١٦,٨ و ٢٠,٦) ووزن المائة حبة (٩,٩ و ١٣,٣) ووزن الكوز (٢١,٠ و ١٨,٠) في موسمي ٢٠٠٧ و ٢٠٠٨ على التوالي.

- امتصاص عناصر النيتروجين والفوسفور والبوتاسيوم لحبوب الذرة (٣٥,١ و ٢١,٣) و (٢١,٢ و ٢٦,٩) و (١٨,١ و ٣١,١) على التوالي عن مثيلاتها في معاملة المقارنة في ٢٠٠٧ و ٢٠٠٨ على التوالي بينما نسبة التفريط لم تزداد معنويا باختلاف المعاملات.

و بتطبيق هذه النتائج يمكن أن تساعد على تقليل التلوث البيئي