

## **RESPONSE OF WHEAT PLANTS TO EM (EFFECTIVE MICROORGANISMS) APPLICATION AND/OR CYANOBACTERIA INOCULATION UNDER SANDY SOIL CONDITION**

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### **ABSTRACT**

During the successive seasons (2008-09 and 2009 -10) wheat field experiments were carried out at EL- Ismailia Agriculture Research Station Agric. Res. Center (ARC) to study the effect of cyanobacteria inoculation and/or effective microorganisms and mineral nitrogen fertilizer on wheat plants (Sakha 69). In this work, cyanobacteria inoculation and the effective microorganisms (EM) were applied to wheat each alone or both in combination under different levels of nitrogen, i.e.,  $\frac{1}{4}$ ,  $\frac{1}{2}$  and full N recommended dose). Results revealed that cyanobacteria inoculation combined with EM application exhibited an economical view that it can save about 50 % was noticed when cyanobacteria combined with EM which recorded a grain yield not significantly different from that obtained by the full recommended nitrogen dose in wheat cultivation. Cyanobacteria inoculation to wheat crop along with EM have also enhanced the NPK- uptake by wheat plant, the soil microbial community, wheat grains technology, dehydrogenase activity and  $\text{CO}_2$  evolution as index of soil fertility.

### **INTRODUCTION**

Wheat is the most important cereal crop in Egypt and is the staple food of the people and thus occupies a central position in forming agricultural policies and dominates all crops in production attention in an attempt to meet the gap arises between the consumption and production. However, in attempting to develop productive, profitable and sustainable agriculture systems, several agriculturists turn to farming methods, which are based on biotechnologies. Two of the several approaches to achieve this goal are using the nitrogen fixing cyanobacteria and the effective microorganism (EM) in order to improve soil fertility and productivity. The use of nitrogen fixing cyanobacteria ensures entirely or partially the mineral nitrogen, while EM is expected to enhance the availability of soil nutrients and humus formation and to control certain plant diseases and pathogens (Myint, 1999). There is a great deal of interest in creating novel association between agronomically important plants, particularly cereals such wheat and  $\text{N}_2$ -fixing microorganisms including cyanobacteria (Spiller *et al.*, 1993). The heterocystous cyanobacterium *Nostoc* sp. is usual among characterized cyanobacteria in its ability to form tight association with wheat roots and penetrate both roots epidermis and cortical intracellular space (Gantar *et al.*, 1991). The nitrogen fixed by *Nostoc* sp. in association with wheat is taken up by the plant and supports its growth, improving grain yields and grain quality (Gantar *et al.*, 1995).

A recent introduction namely “Effective Micro-organisms (EM) technology” is reported to increase soil fertility through enhanced release of the plant nutrients and is getting popularity among the farming community due to low capital cost. Majority of the farmers still have concerns about the significance of this material over the chemical fertilizers and organic manures. Effectiv-emicroorganisms have paid much attention lately in the entire world.

Higa and Parr (1994) reported that the use of EM will supply the following benefits for plants, i.e., grow high quality and seedlings., quickly establishes a beneficial micro-ecology that promotes extremely stable growing conditions and safeguards against common stress-related problems, such as over watering, helps maintain plant health with antioxidants (vitamins and trace minerals), enzymes, and organic acids, reduce problems associated with soils drying out and being re-hydrated due to improved texture and structure. They added that EM contains more than 70 beneficial organisms, more importantly lactic acid bacteria, photosynthetic bacteria (*Rhodopseudomonas palustris*) and yeast. Surprisingly, use of EM helps in augmenting the photosynthesis by about 30 % in all the crops. Further, it controls viruses and fungal damage to crops by inoculating lactic acid bacteria and actinomycetes.

Therefore, the aim of this work is to evaluate the effect of Effective-microorganisms solution and cyanobacteria inoculation each applied alone or both in combination on wheat cultivated in sandy soil in terms of wheat yield and its components, NPK uptake for both wheat grains and straw, soil available NPK and soil biological activity after wheat harvesting.

## MATERIALS AND METHODS

Wheat field experiments were carried out at EL- Ismailia Agriculture Research Station, ARC, during two winter successive seasons of 2008/2009 and 2009/2010 to study the effect of both cyanobacteria inoculation and effective microorganisms (EM) application each alone or both in combination on wheat (*Triticum aestivum* cv. Sakha 69) growth under different mineral nitrogen fertilizer levels as urea of full recommended dose (FRD) (80 kg N ), 50 % (FRD) (40 kg N ) and 25 % (FRD) (20 kg N ). The physio-chemical analyses (Page *et al.*, 1982) of the experimental soil are as shown in Table (1).

**Table (1): Some chemical and physical analyses of the investigated soil**

pH (1:2.5)	EC dS/m	Soluble cations (meq/L)				Soluble anions (meq/L)			
		Ca <sup>++</sup>	Mg <sup>++</sup>	Na <sup>+</sup>	K <sup>+</sup>	CO <sub>3</sub> <sup>-</sup>	HCO <sub>3</sub> <sup>-</sup>	Cl <sup>-</sup>	SO <sub>4</sub> <sup>=</sup>
8.14	1.4	4.56	2.60	3.07	0.36	--	6.60	2.83	1.16
<b>Coarse sand %</b>	<b>Fine sand %</b>	<b>Silt %</b>	<b>Clay %</b>	<b>CaCO<sub>3</sub> %</b>	<b>Textural class</b>				
76.18	15.17	2.35	6.30	1.5	Sandy				
<b>Available N (ppm)</b>		<b>Available P (ppm)</b>			<b>Available K (ppm)</b>				
20		4			49				

The experimental field was prepared by ploughing and puddling, and then divided into 36 plots (3m X 3.5 m each) to represent 12 treatments in three replicates arranged in statistical split plot design. Nitrogen fertilizer represents the main plot in three treatments, while cyanobacteria inoculation and/or effective microorganisms (EM), their combination and the control treatment without inoculation or (EM) application to represent the sub plot. Uniform application of phosphate at the rate of 200 kg fed<sup>-1</sup> as superphosphate (15.5 % P<sub>2</sub>O<sub>5</sub>) and potassium in the form of potassium sulfate (48% K<sub>2</sub>O) at the rate of 50 kg fed<sup>-1</sup> were done as basal to each plot. Nitrogen as urea was applied in three split equal doses according to the treatment. Cyanobacteria inoculation at the rate of 10 kg dried *Nostoc* sp. was executed 7 days after sowing of wheat seeds, while (EM) was foliar sprayed two weeks after sowing at the rate of 40 L fed<sup>-1</sup> in three split doses monthly starting from two weeks after sowing. Irrigation was carried out every three days using the sprinkler system.

At harvest for both seasons, wheat plants were cut just above the soil surface to determine the wheat yield, its components and NPK uptake by wheat grains and straw (Chapman and Pratt, 1961). The remained soil was sampled and subjected to evaluate the available NPK (Page *et al.*, 1982), as well as to determine the counts of bacteria (Allen, 1959), *Actinomyces* (Williams and Davis, 1965), total fungi (Martin, 1950), *Azotobacter* & *Azospirillum* (Cochran, 1950) and cyanobacteria (Allen and Stanier, 1968) CO<sub>2</sub> evolution (Pramer and Schmidt, 1964) and dehydrogenase (Casida *et al.*, 1964) activity as a soil fertility index.

**Statistical analysis:**

All obtained results in both seasons were statistically analyzed as mean values for both seasons which compared for the least significant difference (L. S. D.) as described by Gomez and Gomez (1984).

**Cyanobacteria Inoculum:**

The cyanobacteria inoculums containing the cyanobacterium *Nostoc* sp., was taken from Soils, Water & Environ. Res. Inst., ARC., Giza, Egypt. The isolated *Nostoc* sp. was used to prepare the dried soil based cyanobacteria inoculum as described by Venkataraman (1972).

**What is the effective microorganisms:**

The effective microorganisms (commonly termed EM) is an organic biofertilizer containing a mixture of Lactica acid bacteria (*Lactobacillus plantarum* and *Lactobacillus casei*), photosynthetic bacteria (*Rhodospseudomonas palustris* and *Radobacter sphaeroides*), yeasts (*Saccharomyces cerevisia* and *Candida utilis*), ray fungi (*Streptomyces albus* and *Streptomyces griseus*) and fungi (*Aspergillus oryzae*). These microorganisms were blended in molasses or sugar medium, maintained at low pH under ambient conditions (kato *et al.*, 1999) and used.

## RESULTS AND DISCUSSION

### Wheat yield components:

Data in Table (2) indicated the effect of cyanobacteria inoculation and/or EM application and N fertilization levels on wheat yield components. All the treatments increased significantly the wheat grain yield over the control treatments. The highest grain yield ( $1295.34 \text{ kg fed}^{-1}$ ) attained by cyanobacteria plus EM treatments combined with full dose of mineral N ( $80 \text{ kg N fed}^{-1}$ ) followed by  $1274.77 \text{ kg fed}^{-1}$  for cyanobacteria + EM combined with  $\frac{1}{2}$  full mineral nitrogen ( $40 \text{ kg N fed}^{-1}$ ) dose treatment. However, there was no significant difference between these two treatments. The application of full nitrogen dose gave significantly the highest mean wheat grain yield ( $1043.91 \text{ kg fed}^{-1}$ ) compared with the other two levels of nitrogen ( $\frac{1}{4}$  and  $\frac{1}{2}$  N dose).

Same behavior exhibited by grain yield was observed for straw yield indicating the highest straw yield ( $3.31 \text{ tons fed}^{-1}$ ) for the treatment with cyanobacteria plus EM under full nitrogen dose and followed by ( $2.97 \text{ tons fed}^{-1}$ ) for cyanobacteria plus EM under  $\frac{1}{2}$  N level treatment without significant difference between each others.

Towards nitrogen application also full dose recorded the highest significant mean straw yield ( $2.68 \text{ tons fed}^{-1}$ ) due to full N dose application compared with the other two nitrogen levels.

1000-grain weight showed an indefinite trend in response to the tested treatments. However, this notice depends on the number of panicles plant<sup>-1</sup>, which correlated drastically with the grain yield.

These results are in agreement with those described by Abd-Alla *et al.* (1994) and Mussa *et al.* (2003) who attributed the increase in wheat yield components resulted from the cyanobacteria inoculated treatments to the substantial increases in  $\text{N}_2$  fixation in soil due to nitrogenase activity of the cyanobacteria succeeded to create tight association with the roots of wheat plants. They also added that cyanobacteria inoculation led to soil structure improvement, which being reflected on soil fertility and consequently on cultivated crop. Also, in the present study, inoculation with *Nostoc* sp. Inoculation increased significantly wheat yield and its components especially when combined with EM plus  $\frac{1}{2}$  N dose. In this concern, Ragab *et al.* (2010) explained the increase in faba bean yield due to inoculation with *Azotobacter* plus EM may be attributed to that the role of *Rhizobium leguminosarum* that increased the growth and  $\text{N}_2$ -fixation by faba bean plants. While the high response of bean seed yield to EM application can be explained on the basis that EM increases germination, stimulates the photosynthetic process and enhances the enzymes activities. Microorganisms of EM and N-fixers have a beneficial role in speedy emergence of seedlings, leaf photosynthesis, diseases and herbs resistance which consequently, produces healthy growth and wealthy yield. Also, they reported that the use of EM enhanced yield of bean due to greater rates of photosynthesis and dry matter accumulation. Significant increase was found in all wheat yield parameter with application of EM was applied along with  $\frac{1}{2}$  recommended nitrogen dose producing (2831

kg grains ha<sup>-1</sup>) very close to full-recommended nitrogen dose (3017 kg grains ha<sup>-1</sup>) (Hussain *et al.*, 1999). These observations are in parallel with the results in this study.

**Table (2): Effect of cyanobacteria inoculation, effective micro-organisms (EM) application and N-fertilization on wheat yield components (Data are a mean of two seasons)**

N-fertilization	Treatments				Means
	Control	Cyanobacteria	EM	Cyanobacteria +EM	
	<b>Grain yield kg fed<sup>-1</sup></b>				
1/4 N	574.28	780.96	845.49	962.25	790.75
1/2 N	736.09	869.57	866.46	1274.77	936.72
full dose N	872.10	993.48	1014.70	1295.34	1043.91
Means	727.49	881.34	908.88	1177.45	
L.S.Dat 5% N: Treatment: Interaction:	96.77 72.26 144.00				
	<b>Straw yield ton fed<sup>-1</sup></b>				
1/4 N	2.61	2.33	2.59	2.28	2.45
1/2 N	2.41	3.03	2.42	2.97	2.71
full dose N	2.69	2.49	2.21	3.31	2.68
Means	2.57	2.62	2.41	2.85	
L.S.Dat 5% N: Treatment: Interaction:	0.31 0.27 0.53				
	<b>1000 grains weight (g)</b>				
1/4 N	45.87	53.30	48.77	49.00	49.24
1/2 N	46.97	53.83	54.50	48.30	50.90
full dose N	42.63	49.97	40.10	50.90	45.90
Means	45.16	52.37	47.79	49.40	
L.S.Dat 5% N: Treatment: Interaction:	2.26 3.92 4.44				

**NPK uptake by wheat grains:**

Results in Table (3) revealed that inoculation with cyanobacteria and EM application under full nitrogen dose gave the highest N uptake amount (22.03 kg N fed<sup>-1</sup>) with no significant difference with that recorded by cyanobacteria and EM treatment under ½ N dose (21.67 kg N fed<sup>-1</sup>). Due to nitrogen application alone, there was no significant difference in N uptake values obtained by either ½ or full N dose treatments. Their respective mean N uptake values were 15.71 and 16.87 kg N fed<sup>-1</sup>.

Phosphorus uptake and K uptake indicated the same trend in response to the tested treatments as shown in N uptake. In case of P uptake the highest value of 2.17 kg P fed<sup>-1</sup> was not significantly differ from 2.07 kg P

fed<sup>-1</sup> for cyanobacteria plus EM plus ½ N dose and cyanobacteria plus EM plus full N dose, treatments, respectively. While with K uptake, the highest value of 6.63 kg K fed<sup>-1</sup> was not significantly differed from that of 6.35 kg K fed<sup>-1</sup> for cyanobacteria plus EM + ½ N dose and cyanobacteria + EM + Full dose treatments, respectively.

**Table (3): Effect of cyanobacteria inoculation, effective micro-organisms (EM) application and N-fertilization on NPK uptake by wheat grains (Data are a mean of two seasons)**

N-fertilization	Treatments				Means
	Control	Cyanobacteria	EM	Cyanobacteria +EM	
	<b>N-uptake kg fed<sup>-1</sup></b>				
1/4 N	8.04	10.15	15.20	16.36	12.44
1/2 N	12.51	14.78	13.86	21.67	15.71
full dose N	18.31	11.92	15.22	22.03	16.87
Means	12.95	12.28	14.76	20.02	
L.S.Dat 5%					
N:					1.45
Treatment:					1.20
Interaction:					2.29
	<b>P-uptake kg fed<sup>-1</sup></b>				
1/4 N	0.75	1.17	1.18	1.35	1.11
1/2 N	1.18	1.74	1.39	2.17	1.62
full dose N	1.35	1.49	1.93	2.07	1.71
Means	1.09	1.47	1.50	1.86	
L.S.Dat 5%					
N:					0.16
Treatment:					0.13
Interaction:					0.24
	<b>K-uptake kg fed<sup>-1</sup></b>				
1/4 N	2.99	3.75	4.22	4.62	3.90
1/2 N	3.98	4.87	3.99	6.63	4.87
full dose N	4.45	5.17	5.38	6.35	5.34
Means	3.81	4.60	4.53	5.87	
L.S.Dat 5%					
N:					0.52
Treatment:					0.38
Interaction:					0.76

For nitrogen application means, the priority in NPK uptake by wheat grains was for the use of full N dose. Their respective mean values, were 16.87 kg N fed<sup>-1</sup> (N uptake), 1.71 kg P fed<sup>-1</sup> (p-uptake) and 5.34 kg K fed<sup>-1</sup> (K-uptake).

**NPK uptake by wheat straw**

Data in Table (4) indicated that N uptake value recorded by the cyanobacteria + EM + ½ N dose (14.83 kg N fed<sup>-1</sup>) was not significantly differed from that recorded by Cyanobacteria + EM + full N dose treatment (16.55 kg N fed<sup>-1</sup>). In contrast, P uptake value of 4.15 kg P fed<sup>-1</sup> (cyanobacteria + EM + ½ N dose) was significantly higher than that recorded by cyanobacteria + EM + Full N dose treatment (2.98 kg P fed<sup>-1</sup>).

Same observations were noticed by K –uptake, that the K- uptake value recorded by the treatment of cyanobacteria + EM + Full N dose (22.18

kg K fed<sup>-1</sup>) was significantly higher than that recorded by cyanobacteria + EM + ½ N dose treatment (16.91 kg k fed<sup>-1</sup>).

**Table (4): Effect of cyanobacteria inoculation, effective micro-organisms (EM) application and N-fertilization on NPK uptake by wheat straw (Data are a mean of two seasons)**

N-fertilization	Treatments				Means
	Control	Cyanobacteria	EM	Cyanobacteria +EM	
	<b>N-uptake kg fed<sup>-1</sup></b>				
1/4 N	13.05	27.94	10.37	11.95	15.83
1/2 N	24.18	30.27	14.54	14.83	20.96
full dose N	16.16	14.94	13.38	16.55	15.26
Means	17.80	24.38	12.76	14.44	
L.S.Dat 5% N: Treatment: Interaction:	2.65 2.15 4.14				
	<b>P-uptake kg fed<sup>-1</sup></b>				
1/4 N	1.55	3.51	2.34	1.36	2.19
1/2 N	4.10	4.52	2.91	4.15	3.92
full dose N	2.24	1.24	4.01	2.98	2.62
Means	2.63	3.09	3.09	2.83	
L.S.Dat 5% N: Treatment: Interaction:	0.59 0.43 0.87				
	<b>K-uptake kg fed<sup>-1</sup></b>				
1/4 N	16.18	12.82	14.65	17.55	15.30
1/2 N	25.34	19.37	12.60	16.91	18.56
full dose N	17.77	19.67	15.84	22.18	18.87
Means	19.76	17.29	14.36	18.88	
L.S.Dat 5% N: Treatment: Interaction:	3.15 2.32 4.66				

Increasing the nutrient uptake by wheat grain and straw in response to the use of both EM and cyanobacteria as biofertilizer separately was confirmed by those of Abd EL- Rasoul *et al.* (2004) and Mussa *et al.* (2003) who indicated that spraying both EM and nitrogen fixing biofertilizers individually had significantly increased N P K uptake by grains and straw over the control treatments. The use of cyanobacteria along with EM plus full-N dose had achieved grains and straw yields, NPK uptake by grains and straw which were very close to and not significantly different from those achieved by same treatments under the influence of ½ N dose. This trend are in parallel to what revealed by EL- Mancy *et al.* (1997) who found that combination between biofertilizers with reduced amount of the mineral nitrogen can lead to saving chemical-N fertilizer (about 50 %) and improving NPK uptake by rice grains and straw. Inoculation with the nitrogen fixing

*Azospirillum* to wheat as biofertilizer combined with ½ recommended N dose increased significantly grains and straw yields and NPK- uptake by grains and straw compared to the control without inoculation (EL- Kasas, 2002).

**Soil available NPK:**

In respect to available NPK amounts remained in soil after wheat harvesting, results in Table (5) indicated that available-N significantly increased in response to increasing nitrogen fertilizer doses over ½ N dose with priority to ½ N-dose treatment, which recorded 126.25 ppm available N.

**Table (5): Effect of cyanobacteria inoculation, effective micro-organisms (EM) application and N-fertilization on soil available nitrogen, phosphorus and potassium after wheat harvesting (Data are a mean of two seasons)**

N-fertilization	Treatments				Means
	Control	Cyanobacteria	EM	Cyanobacteria +EM	
	<b>Available nitrogen (ppm)</b>				
1/4 N	90	100	80	100	92.50
1/2 N	120	125	120	140	126.25
full dose N	80	90	100	110	95.00
Means	96.66	105	103.33	116.60	
L.S.Dat 5% N: Treatment: Interaction:	2.65 2.14 4.14				
	<b>Available phosphorus (ppm)</b>				
1/4 N	7.40	9.00	11.40	5.90	8.43
1/2 N	4.16	8.30	6.90	7.07	6.61
full dose N	6.34	6.50	7.60	7.50	6.99
Means	5.97	7.93	8.63	6.82	
L.S.Dat 5% N: Treatment: Interaction:	1.40 1.16 2.50				
	<b>Available potassium(ppm)</b>				
1/4 N	103	110	86	58	89.25
1/2 N	70	79	81	76	<b>76.50</b>
full dose N	100	79	118	70	91.75
Means	91	89.33	95	68	
L.S.Dat 5% N: Treatment: Interaction:	3.54 3.09 6.93				

Apart from nitrogen doses, cyanobacteria inoculation combined with EM application had achieved the highest available-N amounts (116.60 ppm). This high available-N amount was significantly differed from that recorded by either cyanobacteria (105.00 ppm-N) or EM (103.33 ppm-N) treatments each applied alone.

Both cyanobacteria inoculation and/or EM application each alone or both in combination were affected with the different fertilizer-N dose. The highest available-N amount (140.00 ppm) was noticed in the treatment

received cyanobacteria +EM +  $\frac{1}{2}$  N dose. However, this high amount was significantly exceeded all other interacted treatments.

Available phosphorus amounts decreased significantly in response to nitrogen fertilizer doses, since they recorded less amounts of 6.61 and 6.99 ppm than that of 8.43 ppm for  $\frac{1}{2}$ , full and  $\frac{1}{4}$  N-dose treatments, respectively.

Due to cyanobacteria inoculation and EM each alone or both in combination, results revealed that the highest significant available phosphorus amount of 8.63 ppm for EM treatment alone followed by 7.93 and 6.82 ppm for cyanobacteria and cyanobacteria + EM treatments, respectively. The highest significant available-P amount of 11.40 ppm was observed in samples treated with EM combined with  $\frac{1}{4}$  N-dose compared to the other interaction treatments except for cyanobacteria combined with  $\frac{1}{4}$  N-dose (9.00 ppm-P) treatment.

Available potassium amounts had fluctuated between decrease in response to  $\frac{1}{2}$  and full nitrogen dose compared to  $\frac{1}{4}$  N-dose application. Nevertheless, the highest available-K amount of 91.75 ppm (full-N dose) was significantly higher than that recorded by  $\frac{1}{2}$  N dose (76.50 ppm-K) but did not than that of 89.25 ppm-K for  $\frac{1}{4}$  N-dose treatment.

On the other hand, EM applied alone had achieved the available-K amount (95.00 ppm) being significantly higher than those of cyanobacteria (89.33ppm-K) and cyanobacteria + EM (68.00 ppm-K) treatments.

Nitrogen, cyanobacteria and EM interactions resulted in the highest significant available-K amount of 118.00 ppm more than those recorded by the other interaction relations.

The use of cyanobacteria and EM enhanced the chemical properties of the wheat post harvest remained soil. Mandal *et al.* (1999) emphasized that inoculation with cyanobacteria (SBI) might help to regenerate quickly and improve soil structure. Albeit, cyanobacteria are known to excrete extracellularly a number of compounds like polysaccharides, peptides, lipids...etc. during their growth in soil particles, and hold / glue them together in the form of micro-aggregates being a reason to improve the nutrient availability in soil.

#### **Soil Micro-organisms Counts:**

Total fungi count in soil after wheat harvesting (Tables 6 & 7) exhibited no significance in response to nitrogen fertilizer doses. However, the highest total fungi count ( $32.90 \times 10^2$  cfu g<sup>-1</sup> soil) obtained by  $\frac{1}{2}$  N-dose treatment.

**Table (6): Effect of cyanobacteria inoculation, effective micro-organisms (EM) application and N-fertilization on total fungi, actinomycetes and total bacterial counts after wheat harvesting (Data are a mean of two seasons)**

N-fertilization	Treatments				Means
	Control	Cyanobacteria	EM	Cyanobacteria +EM	
	<b>Total fungi x10<sup>2</sup></b>				
1/4 N	7.20	29.10	53.70	38.90	<b>32.23</b>
1/2 N	22.60	36.00	15.70	57.30	32.90
full dose N	15.60	25.50	33.20	12.80	<b>21.77</b>
<b>Means</b>	<b>15.13</b>	<b>30.20</b>	<b>34.20</b>	<b>36.33</b>	
L.S.Dat 5%					
N:	NS				
Treatment:	6.49				
Interaction:	14.19				
	<b>Actinomycetes x10<sup>3</sup></b>				
1/4 N	1.00	38.20	58.10	31.80	<b>32.275</b>
1/2 N	18.70	46.70	18.60	84.10	<b>42.025</b>
full dose N	8.30	31.60	38.80	7.10	<b>21.450</b>
<b>Means</b>	<b>9.30</b>	<b>38.83</b>	<b>38.50</b>	<b>71.00</b>	
L.S.Dat 5%					
N:	5.22				
Treatment:	9.01				
Interaction:	14.14				
	<b>Total bacteria x10<sup>6</sup></b>				
1/4 N	26.10	38.70	273.20	45.10	106.42
1/2 N	38.20	77.60	32.00	131.40	<b>69.80</b>
full dose N	28.50	34.70	47.80	17.70	<b>32.18</b>
<b>Means</b>	<b>30.93</b>	<b>50.30</b>	<b>117.66</b>	<b>64.73</b>	
L.S.Dat 5%					
N:	25.26				
Treatment:	15.72				
Interaction:	24.20				

Apart from nitrogen doses, cyanobacteria + EM treatment gave the highest insignificant mean of total fungi count of 36.33 x 10<sup>2</sup> cfu g<sup>-1</sup> soil compared to those recorded by the other treatments.

Interaction effect due to the treatments under the influence of N-doses resulted in higher significant total fungi count (57.30 x 10<sup>2</sup> cfu g<sup>-1</sup>soil) compared to the other interaction influences except for those recorded by cyanobacteria + EM + ¼ N-dose (38.90 x 10<sup>2</sup> cfu g<sup>-1</sup> soil).and EM + ¼ -N dose (53.70 x 10<sup>2</sup> cfu g<sup>-1</sup> soil).

Actinomycetes count indicated significant response due to N-dose application, when the highest count (42.03 x 10<sup>3</sup>) cfu g<sup>-1</sup> soil) recorded by the use of ½ N- dose compared with the other applied N-doses.

Cyanobacteria treatment + EM had recorded the highest significant actinomycetes count (71.00 x 10<sup>3</sup> cfu g<sup>-1</sup>soil) compared with those recorded by either cyanobacteria or EM each alone and/or control.

On the other hand, when cyanobacteria combined with EM under the effect of ½ N-dose gave the highest actinomycetes count of (84.10 x 10<sup>3</sup> cfu

g<sup>-1</sup>soil) compared to the other treatments under the influence of N-doses application.

Due to the total bacterial count, the highest significant values were recorded by the ¼ N-dose, EM treatment, and cyanobacteria combined with EM under the effect of ½ N-dose application. The corresponding total bacterial count were 106.42, 117.66 and 131.40 x 10<sup>6</sup> cfu g<sup>-1</sup> soil.

Table (7) revealed that *Azotobacter* gave its highest count (9.10 x 10<sup>5</sup> cfu g<sup>-1</sup> soil) in response to EM + ¼ N dose treatment. This high count was significantly higher than those given by all the other tested treatments.

**Table (7): Effect of cyanobacteria inoculation, effective micro-organisms (EM application) and N-fertilization on N<sub>2</sub>-fixers (*Azotobacter*, *Azospirillum* and total cyanobacteria) counts in soil after wheat harvesting (Data are a mean of two seasons)**

N-fertilization	Treatments				
	Control	Cyanobacteria	EM	Cyanobacteria +EM	Means
<b><i>Azotobacter</i> x10<sup>5</sup></b>					
1/4 N	5.60	4.40	9.10	2.50	5.40
1/2 N	2.00	5.40	2.90	6.30	<b>4.15</b>
full dose N	5.40	2.30	3.10	7.00	<b>4.45</b>
Means	4.33	4.03	5.03	5.27	
L.S.Dat 5%					
N:	0.22				
Treatment:	0.23				
Interaction:	0.40				
<b><i>Azospirillum</i> x 10<sup>5</sup></b>					
1/4 N	5.20	1.20	6.30	1.70	<b>3.60</b>
1/2 N	1.10	4.80	2.20	6.00	<b>3.53</b>
full dose N	4.60	1.60	2.20	7.90	4.08
Means	<b>3.63</b>	<b>2.53</b>	<b>3.57</b>	<b>5.20</b>	
L.S.Dat 5%					
N:	0.227				
Treatment:	0.156				
Interaction:	0.354				
<b>N<sub>2</sub>-fixing cyanobacteria x 10<sup>3</sup></b>					
					<b>Means</b>
1/4 N	1.70	11.70	2.00	27.70	<b>10.775</b>
1/2 N	6.00	4.00	19.00	43.30	18.075
full dose N	0.63	3.30	1.00	1.00	<b>5.93</b>
Means	<b>2.77</b>	<b>6.33</b>	<b>7.33</b>	<b>24.00</b>	
L.S.Dat 5%					
N:	7.19				
Treatment:	6.49				
Interaction:	9.34				

*Azospirillum* had recorded its favorite count number of 7.90 x 10<sup>5</sup> cfu g<sup>-1</sup> soil with the use of cyanobacteria + EM + full- N dose treatment which, was significantly differed from those attained by the other tested treatments.

Due to the number of the nitrogen fixing cyanobacteria, it was obvious that increasing the nitrogen levels to full- N dose drastically suppressed the presence of cyanobacteria in soil. However, the treatment of cyanobacteria + EM + ½ N dose had achieved the highest significant cyanobacteria count ( $43.30 \times 10^3$  cfu  $g^{-1}$  soil) in soil compared to the other treatments.

**CO<sub>2</sub> evolution:**

Carbon dioxide evolution by soil (Table 8) calculated after wheat harvesting showed its highest significant amount of 391.33, 394.30 and 470.00 mg CO<sub>2</sub> 100 g<sup>-1</sup> soil due to the application of EM treatment alone, ½ N-dose alone and cyanobacteria combined with EM under the influence of ¼ N-dose, respectively in comparison to their related treatment without nitrogen.

**Table (8): Effect of cyanobacteria inoculation, effective micro-organisms (EM) application and N-fertilization on CO<sub>2</sub>-evolution and dehydrogenase activity in soil (Data are a mean of two seasons)**

N-fertilization	Treatments				
	Control	Cyanobacteria	EM	Cyanobacteria +EM	Means
	<b>CO<sub>2</sub>- evolution (mg CO<sub>2</sub> / 100g soil /day)</b>				
1/4 N	199	368	470	372	352.2
1/2 N	370	431	315	461	394.3
full dose N	257	358	389	249	313.3
<b>Means</b>	280.33	358.66	391.33	360.66	
<b>L.S.Dat 5%</b>					
<b>N:</b>	2.75				
<b>Treatment:</b>	2.94				
<b>Interaction:</b>	5.17				
	<b>Dehydrogenase activity (µg TPF / 100g soil /day)</b>				
1/4 N	9.20	15.0	47.30	18.9	22.60
1/2 N	13.80	9.30	18.30	16.2	14.40
full dose N	26.10	30.8	14.80	19.9	22.90
<b>Means</b>	16.36	18.36	26.80	18.33	
<b>L.S.Dat 5%</b>					
<b>N:</b>	2.69				
<b>Treatment:</b>	2.60				
<b>Interaction:</b>	4.70				

**Dehydrogenase activity (DHA):**

Dehydrogenase activity in soil remained after wheat harvesting (Table 9) expressed its highest significant value of 47.30 mg TPF 100 g<sup>-1</sup> soil/day due to EM combined with ¼ N-dose compared to other studied treatments. Due to the effect of nitrogen levels applied alone, both ¼ N and full N doses treatments gave DHA mean values of 22.60 and 22.90 mg TPF 100 g<sup>-1</sup> soil/ day, which were not significantly differed from each others, while they were significantly higher than that of 14.40 mg TPF 100 g<sup>-1</sup> soil/ day for ½ N-dose. As for the treatments received EM and/or cyanobacteria either each applied alone or both in combination, EM treatments alone gave the

highest significant DHA value of 26.80 mg TPF 100 g<sup>-1</sup> soil/ day compared to the other treatments including the control one.

Acea *et al.* (2003) showed that inoculation with different cyanobacteria strains induced great microbial proliferation as well as high increases in soil organic matter and available nutrients that improved the cultivated plant status. EL- Kasas (2002) reported that Inoculation with the nitrogen fixing *Azospirillum* to wheat increased the soil *Azospirilla* and other microbial population including fungi, actinomycetes and *Azotobacter* , and consequently increased both the dehydrogenase activity and CO<sub>2</sub> evolution, which are considered as index for biological activity and soil fertility (Ghazal, 1980). Due to The use of EM, Frighetto *et al.* (1999) confirmed that the use of EM has improved enhancement in soil biological and chemical properties which ensure not only organic and biological sources of the essential nutrients supply but also show some positive interaction with chemical fertilizers through increasing their efficiency and thereby reducing the environmental hazards.

### **CONCLUSION**

Generally, It could be concluded that the use of cyanobacteria inoculation technology combined with EM along with one half of the recommended nitrogen dose in wheat production (especially in the areas with new reclaimed soil such as in this study) can enhance grains and straw yields, grains quality, NPK-uptake by grains and straw, and improve the soil nutrient status of the soil remained after wheat harvesting. However, this study needs to be confirmed through its execution in different locations in Egypt, with special concern with effective microorganisms application.

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### **استجابة نباتات القمح للتلقيح بالسيانوبكتريا و الميكروبات الفعالة تحت ظروف التربة الرملية**

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معهد بحوث الأراضى والمياه والبيئة – مركز البحوث الزراعية – الجيزة -مصر**

لقد أجريت تجربة حقلية بمحطة البحوث الزراعية بالاسماعيلية والتابعة لمركز البحوث الزراعية وذلك فى موسمين شتويين متتاليين (2009/1008 – 2010/2009) لتقييم امكانية استخدام كل من السيانوبكتريا ومحلول الميكروبات الفعالة كسماد حيوى يوفر جزء من السماد النيتروجينى اللازم لعملية انتاج القمح وتأثير ذلك على محصول القمح من الحبوب والقش، المحتوى النيتروجينى والفوسفورى والبوتاسيومى لكل من الحبوب والقش، الصفات التكنولوجية للحبوب، النيتروجين المتاح بالتربة بعد حصاد القمح ، المحتوى الميكروبي بالتربة بعد حصاد القمح.

في هذه التجربة تم تلقيح القمح بالسيانوبكتريا جافة بمعدل 15 كجم / للفدان وكذلك رش محلول الميكروبات الفعالة بمعدل 40 لتر للفدان بحيث تم استخدام أى منهما منفردا أو مجتمعين تحت تأثير مستويات مختلفة من النيتروجين هي المستوى الموصى به و 1/4 المستوى الموصى به و 1/2 المستوى الموصى به. وكانت أهم النتائج المتحصل عليها كما يلي:

أولا: مكونات محصول القمح (الحبوب والقش):

- 1- تحقق أعلى محصول للحبوب (34 و 1295 كجم/ فدان) عند استخدام السيانوبكتريا + الميكروبات الفعالة + المستوى النيتروجيني الكامل الا أن هذا المحصول كان غير مختلف معنوياً مع ذلك المتحصل عليه عند استخدام نفس المعاملة + 1/2 المستوى النيتروجيني الكامل (77 و 1274 كجم/ فدان).
- 2- أعطى استخدام المستوى النيتروجيني الكامل منفردا أعلى محصول للحبوب بالمقارنة مع استخدام مستويات النيتروجين الأخرى منفردة.

3- أظهر محصول القش نفس الاتجاه المتحقق مع محصول الحبوب.

4- لم يكن هناك اتجاه محدد لتأثير المعاملات تحت الدراسة على وزن ال 1000 حبة.

ثانياً: لمحتوى العناصر (نيتروجين - فوسفور - بوتاسيوم) لكل من الحبوب والقش:

- 1- تحقق أعلى محتوى نيتروجيني للحبوب (3 و 22 كجم نيتروجين/ فدان) عند استخدام السيانوبكتريا + الميكروبات الفعالة + المستوى النيتروجيني الكامل الا أن هذا المحتوى النيتروجيني كان غير مختلف معنوياً مع ذلك المتحصل عليه عند استخدام نفس المعاملة + 1/2 المستوى النيتروجيني الكامل (67 و 21 كجم نيتروجين / فدان).

2- أظهر محتوى الحبوب من الفوسفور والبوتاسيوم نفس الاتجاه المتحقق مع المحتوى النيتروجيني.

3- أظهر محتوى القش من النيتروجين نفس الاتجاه المتحقق مع المحتوى النيتروجيني للحبوب.

- 4- كان محتوى القش من الفوسفور عند استخدام السيانوبكتريا و الميكروبات الفعالة + 1/2 المستوى النيتروجيني الكامل (15 و 4 فوسفور كجم/ فدان) أعلى معنوياً من ذلك المتحصل عليه عند استخدام نفس المعاملة + المستوى النيتروجيني الكامل (98 و 2 كجم فوسفور / فدان) وعلى العكس من ذلك كان محتوى القش من البوتاسيوم أعلى معنوياً لمعاملة السيانوبكتريا + الميكروبات الفعالة + المستوى النيتروجيني الكامل من معاملة السيانوبكتريا + الميكروبات الفعالة + 1/2 المستوى النيتروجيني الكامل.

ثالثاً: العناصر المتاحة بالتربة بعد حصاد القمح:

- 1- أدى التلقيح بالسيانوبكتريا أو الرش بمحلول الميكروبات الفعالة الى زيادة كل من النيتروجين والفوسفور المتاح بالتربة بعد حصاد القمح بالمقارنة مع المعاملات الأخرى. ومن ناحية أخرى فقد تآرجح تركيز البوتاسيوم المتاح بالتربة بعد حصاد القمح بين الزيادة والنقصان نتيجة لأثر المعاملات تحت الدراسة.

رابعاً: أعداد الميكروبات بالتربة بعد حصاد القمح:

- 1- تأثر عدد الميكروبات بالتربة بعد حصاد القمح نتيجة المعاملات المختلفة حيث أدى التلقيح بالسيانوبكتريا أو الرش بمحلول الميكروبات الفعالة الى زيادة غير معنوية في أعدادها كما في أعداد الفطريات أو زيادة معنوية في أعدادها كما في أعداد الأكتينوميسيتات.

خامساً: تركيز غاز ثاني أكسيد الكربون ونشاط انزيم الديهيدروجينيز بالتربة بعد حصاد القمح:

- 1- حققت المعاملة السيانوبكتريا + الميكروبات الفعالة + 1/4 المستوى النيتروجيني الكامل أعلى تركيز لغاز ثاني أكسيد الكربون بالتربة بالمقارنة مع المعاملات الأخرى.
- 2- حققت المعاملة الميكروبات الفعالة + 1/4 المستوى النيتروجيني الكامل أعلى نشاط لانزيم الديهيدروجينيز بالتربة بالمقارنة مع المعاملات الأخرى.

قام بتحكيم البحث

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