

## **EFFECT OF COMPOST, (AM) MYCORRHIZAE AND N<sub>2</sub>-FIXING BACTERIA ON GROWTH AND YIELD OF WHEAT**

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### **ABSTRACT**

A field experiment was carried out in sandy soil at El-Ismailia Agricultural Research Station, El-Ismailia Governorate, Agricultural Research Center (ARC) to study the effect of treating the soil with different types of compost (plant residues with organic manure and plant residues with cellulose decomposers), (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on the growth and yield of wheat plants. Plant dry weight recorded a significant increase when the soil was treated with compost provided with cellulose decomposers and biofertilizers. The highest number of tillers obtained with compost B amended with organic manure and biofertilizers.

The maximum infection percentage was due to compost A provided with organic manure. The optimum nitrogenase enzyme activity obtained when the soil was treated with compost B plus cellulose decomposers and biofertilization. High significant nitrogen percentage was shown when the soil was amended with compost D + organic manure + biofertilizers. Inoculation with N<sub>2</sub>-fixers + (AM) mycorrhizae led to the increase of phosphorus % and this was clearly obtained with compost "B" plus cellulose decomposers. Inoculation with mixed diazotrophs + (AM)-mycorrhizae increased potassium % and this was obtained with compost D plus organic manure at the period of 45 days from sowing.

Results showed significant increases in yield parameters, i.e., 1000-grain weight, grain yield and straw yield. The addition of compost B amended with organic manure + N<sub>2</sub>-fixers + (AM) mycorrhizal gave the superior yield. Generally, it was remarkable that the essential role of organic matter represented in compost, N<sub>2</sub>-fixers and (AM) mycorrhizae in enhancing the growth and yield of wheat.

**Keywords:** Wheat, *Azospirillum lipoferum*, *Azotobacter chroococcum*, *Bacillus polymyxa*, *Klebsiella pneumoniae*, AM-mycorrhizae.

### **INTRODUCTION**

Wheat is considered the main economic crop in the world. The necessity of using compost from different sources of plant residues as an organic matter is a paramount importance to wheat productivity. Composting is a microbiological, non-polluting and safe method disposal through converting organic matter (wastes) into resources that provides nutrients to crop and enhance the tilth, fertility and productivity of soil. Nutrients became more available to plants while pathogens destroyed (Crecchio *et al.*, 2004). Compost made from bio-solids contains macro and micronutrients essential for plant growth. In addition to humic substances which promote and improve nutrients availability (Wong *et al.*, 1999). The addition of organic matter stimulated total microbial growth particularly in the presence of biofertilizers. These treatments show corresponding improvements in microbial counts of *Azotobacter*, phosphate dissolving bacteria and *Azospirilla* (Estafanous, 2003).

Addition of compost to soil increased the incidence of bacteria in plant rhizosphere area, therefore, the number of bacteria and fungi increased when organic manure compost was added to soil (Mohammed *et al.*, 2007 and Steven and Armelle, 2008). Application of organic materials namely garbage compost, maize stalks and soybean straw with half dose of N-fertilizer to either clay or sandy soil led to high increase in the dry matter of wheat plants inoculated with *Azospirillum* or *Bacillus*. The application of farmyard manure compost and cellulose decomposers and their mixture with addition of NPK increased significantly stem length, number of leaves, and dry weight of pepper compared to the control (Khalif *et al.*, 2000 and Blaise *et al.*, 2005).

Application of biofertilizers is important economically to reduce the cost of fertilizers and ecologically to reduce pollution of the environment. Biological nitrogen fixation plays a vital role in the global nitrogen cycle. Various nitrogen-fixing microorganisms are found in different natural ecosystems. Nitrogen fixing bacteria have been found to occur in the rhizosphere and on and in roots of various plants. So, inoculation of wheat and barely plants with *Azospirillum* spp. and *Bacillus* was similar to those caused by addition of gibberellic acid in growth (Kucey, 1988).

Many investigators showed that strains of *Azospirillum lipoferum*, *Azotobacter chroococcum* and *Bacillus polymyxa* increased the yield of wheat and maize plants if they inoculated as a mixture (Ishac *et al.*, 1991; Mitkess *et al.*, 1996; Amara and Dahdoh, 1997 and Pandey *et al.*, 1998). Application of inorganic nitrogen and inoculation with *Bacillus polymyxa* increased the dry weight and N-yield of wheat plants (Moharram *et al.*, 1997).

(AM)-mycorrhizae can produce dramatic benefits to plant growing in P-deficient soils. (AM)-mycorrhizae increase the growth and yield of most plants by improving P-uptake, tolerance to drought and salinity and resistance to pathogens (Jacobson, 1997). Plants may support vesicular arbuscular mycorrhizae and asymbiotic nitrogen fixing bacteria. The importance of interaction between mycorrhizae and asymbiotic nitrogen fixing bacteria has been reported by Barea *et al.* (1983). They showed that the interaction improved plant growth particularly in the presence of organic matter.

Therefore, the main objective of this study is to confirm the vital role of bio-fertilizers and compost due to the growth and yield of wheat plant grown in sandy soil.

## **MATERIALS AND METHODS**

- **Field experiment:** A field experiment was carried out in sandy soil at El-Ismailia Agricultural Research Station, El-Ismailia Governorate, Agricultural Research Center (ARC) to study the effect of treating the soil with different types of compost (plant residues with organic manure and plant residues with cellulose decomposers), (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on the growth and yield of wheat plants. The analyses of soil and compost used were summarized in Table (1).

Experimental area was divided into equal plots of 2 x 2m. Plots were arranged in split plot design in three replicates for each treatment. Where the

main plots were various compost sources according to the source of plant residues, i.e., the compost includes the straw of:

Compost A: Rice, wheat, faba bean and maize.

Compost B: Rice, wheat, clover and maize.

Compost C: Rice, faba bean, clover and maize.

Compost D: Rice, wheat, clover, faba bean and maize.

Compost was added to the soil at a rate of 10 m<sup>3</sup> /feddan and mixed with the soil before sowing. These residues were treated with either, organic manure or cellulose decomposers as a sub main plot. Each plot was divided into eleven rows. The grains are spread in each row at a depth of 4 to 5 cm apart, then covered with soil.

- **Grains:** Wheat grains (*Triticum sativum*'s) cultivar Giza 164 were surface sterilized with Clorox solution (0.05%), watered several times then coated with N<sub>2</sub>-fixers inocula.

**Table (1): Some physico-chemical properties of the soil sample (sandy loam soil) and compost used**

Soil analysis		Compost analysis				
Mechanical (%)		Compost type	A	B	C	D
		Properties				
Sand	70.70	pH	7.20	7.15	7.19	7.40
Silt	2.30	EC (dS/m)	13.33	13.99	13.35	13.70
Clay	27.00	Bulck density g/cm <sup>3</sup>	0.320	0.350	0.340	0.360
<b>Chemical</b>		Total nitrogen %	1.20	1.15	1.30	1.30
Organic carbon %	0.143	Organic carbon%	29.50	28.90	28.00	35.00
Total nitrogen %	0.020	Organic matter %	51.00	50.0	48.27	60.34
Total P (ppm)	0.020	C/N ratio	22.91	25.31	21.53	26.92
Available P (ppm)	0.025	Total phosphorus%	0.21	0.24	0.21	0.28
WHC (%)	25.00					
pH	7.75					
EC (dS/m)	1.150					
CaCO <sub>3</sub> %	0.600					
<b>Anions and cations (meq/l)</b>						
Carbonate	Trace					
Bicarbonate	1.13					
Chloride	0.20					
Sulphate	0.84					
Calcium	0.64					
Magnesium	0.11					
Sodium	0.29					
Potassium	1.13					

- **N<sub>2</sub>-fixers:** Mixed cultures of the following diazotrophs and arbuscular mycorrhizal spores (AM) were used in combination as biofertilizers: *Azotobacter chroococcum* was grown on modified Ashby's medium (Abd El-Malek and Ishac, 1968). *Azospirillum lipoferum* was grown on N-deficient semisolid malate medium (Dobereiner *et al.*, 1976). *Bacillus polymyxa* was grown on nitrogen deficient medium of Hino and Wilson (1958). *Pseudomonas* spp. were grown on *Pseudomonas* medium (Haahtela *et al.*, 1983) and *Klebsiella* spp. were grown on N-deficient medium for *Klebsiella*

(Yoch and Pengra, 1966). The growth media were inoculated and incubated at 30°C for the proper time for each microorganism. The density was evaluated to standardize the inocula to 10<sup>6</sup> cells /ml. Mixed cultures of bacterial species in equal ratio were prepared and applied to wheat grains after being mixed with a suitable amount of Arabic gum solution as adhesive agent.

- **Mycorrhizal spores;** Mixed spores of (AM)-mycorrhizal genera: via *Glomus* spp., *Gigaspora* spp., and *Acaulospora* spp., were prepared after propagation and mixed with sand as a carrier (40-50 spore/gram inoculum), and then added to the soil before sowing at the rate of 100g inoculum /line (Daft and Hogarth, 1983).

- **Chemical fertilizers:** Phosphorus as super phosphate (P<sub>2</sub>O<sub>5</sub> 15.5%) and potassium (K<sub>2</sub>SO<sub>4</sub> 48%) were added to the soil before sowing at rates of 200 and 50kg/fed., respectively. Urea (46%) as a nitrogen fertilizer was added as a half dose (60 kg N/fed) for all treatments except control (1) which received full dose of NPK and control (2) plain soil. Urea was added as 6 split equal doses.

- **Treatments and periods:** Six inoculation treatments were included:

1. Grains inoculated with N<sub>2</sub>-fixers grown in soil supplemented with compost having organic manure (T<sub>1</sub>).
2. Un-inoculated grains grown in soil supplemented with compost having organic manure (T<sub>2</sub>).
3. Grains inoculated with N<sub>2</sub>-fixers grown in soil supplemented with compost having cellulose decomposers (T<sub>3</sub>).
4. Un-inoculated grains grown in soil supplemented with compost having cellulose decomposers (T<sub>4</sub>).
5. Full dose of NPK (Control 1).
6. Plain soil (Control 2).

Plant samples were collected after 45, 75 and 120 days from sowing. Mean of 5 plants was taken for the determination of plant height (cm/plant) and dry weight (g/plant). A mean number of shoot tillers was counted at 120 days. Plant shoots were dried, ground and stored for chemical determinations. After harvesting, straw weight (ton / fed), 1000-grain weight (g) and the grain yield (ard. /fed.) were determined.

- **Estimation of root infection rate of (AM)-mycorrhizae:** Root samples were washed several times with tap water. Each sample was cut to small pieces (1.0 cm long) and covered with 10% KOH in bottles and heated in a water bath (80–90°C) for 10–40 min. (Kormanik *et al.*, 1980), this method was modified by using the autoclave at pressure 1 atm. and temperature 121°C for about 20 minutes depending on the age and size of the roots. The roots were washed with tap water and acidified with 1% HCl. The dilute acid was then poured off. The trypan blue (0.05%) stain in lactic acid added to cover the root and then heated in water bath at (80 - 90°C) for 10-15 min. according to the root size (Phillips and Hayamn, 1970).

- **Estimation of nitrogenase enzyme activity:** The nitrogenase activity was determined by using the acetylene reduction technique as  $\mu\text{mole C}_2\text{H}_4 \text{ g}^{-1} \text{ root dry weight h}^{-1}$  according to Somasegaran (1985).

- **Total phosphorus, nitrogen and potassium in plant:** Total phosphorus, nitrogen and potassium in wheat plants were determined as described by Jackson (1973).

## RESULTS AND DISCUSSION

### 1. Effect of different compost types, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on some growth parameters of wheat plant:

#### a) plant height (cm plant<sup>-1</sup>):

Data in (Table 2) revealed that, the height of wheat plants generally increased by inoculation with mixed culture of N<sub>2</sub>-fixers and (AM)-mycorrhizae in presence of organic matter represented in different compost heaps with half dose of mineral nitrogen.

Increase of plant height in comparison with control 1 and control 2 was due to the positive effects of inoculation, treatments and compost type. At the period of 45 days the optimum lengths were obtained with compost C amended with cellulose decomposers and biofertilization combined with the mixed culture of diazotrophs and (AM)-mycorrhizae. It was 46cm plant<sup>-1</sup> and increased to 1.91 and 2.30 fold higher than those in the control (1 and 2), respectively.

Concerning the interaction between the biofertilization, treatments either with organic manure or cellulose decomposers and/or compost type, results showed significant differences at the same period (45 days). Regarding to 75 days period, the optimum lengths were also obtained with compost C amended with cellulose decomposers and N<sub>2</sub>-fixers biofertilization plus (AM)-mycorrhizae. The corresponding values were 99.33cm plant<sup>-1</sup> and increased to 1.716 and 2.33 fold higher than those in the control (1 and 2), respectively.

On the other hand, at 120 days period, the highest wheat lengths was obtained with compost A amended with organic manure and biofertilization combined with the mixed culture of N<sub>2</sub>-fixers plus (AM)-mycorrhizae, where it recorded 105.70cm plant<sup>-1</sup> over the other treatments. It represented 1.24 and 1.76 fold higher than those in the control (1 and 2), respectively.

In general, the positive effect of biofertilization with diazotrophs and (AM)-mycorrhizae combined with half recommended dose of chemical nitrogen enhanced the growth parameters involving the wheat plant heights particularly in the presence of organic matter regardless amendment of compost with organic manure or cellulose decomposers.

Organic matter present in compost is important because it stimulated N<sub>2</sub>-fixers densities and plant growth, though, the significant positive effect of compost on vegetative growth characters may be due to the improvement in soil physical and biological properties and also the chemical characteristics resulting in more release of available nutrients to be absorbed by plant roots. This would affect the physiological process such as photosynthesis activity and in turn the utilization of carbohydrates in addition to water use efficiency by different plants. Consequently when the compost treated with either organic manure or cellulose decomposers associated with biofertilizers inoculation led to positive response in all growth parameters; i.e. plant height

was markedly increased and recorded higher values than control. These results were in the same harmony with those obtained and discussed by Abd El-Moez *et al.* (1999) and El-Gahdban *et al.* (2002).

**Table (2): Effect of different compost heaps amended with cellulose decomposers or biofertilizers on the lengths and dry weight of wheat plants at different agricultural periods**

Compost	Parameter Treatments	Plant length (cm)			Dry weight (g plant <sup>-1</sup> )		
		45 days	75 days	120 days	45 days	75 days	120 days
C	C <sub>1</sub>	24.10	57.90	85.00	0.53	4.40	29.10
	C <sub>2</sub>	19.96	42.33	60.00	0.46	3.28	11.96
A	T <sub>1</sub>	41.97	91.70	105.70	0.66	9.42	26.00
	T <sub>2</sub>	34.40	90.70	103.00	0.82	7.90	20.27
	T <sub>3</sub>	39.80	89.80	99.66	2.30	8.20	34.90
	T <sub>4</sub>	40.30	86.00	95.70	0.94	5.33	17.83
B	T <sub>1</sub>	42.90	83.70	98.00	0.66	7.51	21.32
	T <sub>2</sub>	34.90	79.00	91.70	0.69	8.35	14.06
	T <sub>3</sub>	45.40	92.33	103.33	1.06	8.20	13.84
	T <sub>4</sub>	38.30	83.70	89.00	1.29	7.38	20.46
C	T <sub>1</sub>	38.13	90.70	96.33	0.84	7.36	27.44
	T <sub>2</sub>	37.50	83.00	94.00	0.86	9.93	10.68
	T <sub>3</sub>	46.00	99.33	101.70	0.79	10.20	18.76
	T <sub>4</sub>	40.00	80.00	83.00	0.89	7.08	9.10
D	T <sub>1</sub>	41.00	93.33	99.00	0.93	6.35	21.80
	T <sub>2</sub>	36.60	81.70	93.70	0.69	6.27	8.90
	T <sub>3</sub>	40.43	90.00	98.33	0.94	6.90	18.90
	T <sub>4</sub>	34.70	77.00	84.00	0.64	5.60	17.70
<b>LSD 0.05:</b>							
Inoculation		<b>1.228</b>	n.s	<b>1.014</b>	<b>0.050</b>	<b>0.442</b>	<b>0.063</b>
Treatment		<b>0.877</b>	n.s	n.s	<b>0.091</b>	n.s	<b>0.089</b>
Compost		n.s	n.s	<b>1.427</b>	<b>0.076</b>	<b>0.838</b>	<b>0.162</b>
<b>Interaction:</b>							
Inoculation x Treatment		n.s	n.s	n.s	<b>0.142</b>	n.s	<b>0.152</b>
Inoculation x Compost		<b>1.3</b>	n.s	<b>1.020</b>	<b>0.128</b>	<b>1.310</b>	<b>0.225</b>
Treatment x Compost		<b>0.90</b>	n.s	n.s	<b>0.168</b>	<b>0.84</b>	<b>0.255</b>
Inoculation x Treatm x Compost		<b>2.00</b>	n.s	<b>1.450</b>	<b>0.227</b>	<b>1.311</b>	<b>0.341</b>

\* C<sub>1</sub>= Control 1 (Full dose of NPK only).

C<sub>2</sub>= Control 2 (non inoculation, non organic matter and NPK fertilization (plain soil)).

T<sub>1</sub> = Biofertilization with [N<sub>2</sub>-fixers + (AM)-mycorrhizae] + Compost treated with organic manure.

T<sub>2</sub> = Without biofertilization + Compost treated with organic manure.

T<sub>3</sub> = Biofertilization + Compost treated with cellulose decomposers.

T<sub>4</sub> = Without biofertilization + Compost treated with cellulose decomposers.

- Biofertilization: mixed culture of diazotrophs (*Azospirillum* spp., *Bacillus* spp., *Azotobacter* spp., *Pseudomonase* spp. And *Klebsiella* spp.) and (AM)-mycorrhizal spores.

#### b) Wheat dry weight:

Results in Table (2) show the main effect of compost type and biofertilizer on wheat dry weight (shoot and root dry weight). The superior dry weight was (2.30g plant) with compost "A" provided with cellulose decomposers, N<sub>2</sub>-fixers and (AM)-mycorrhizae plus half dose of mineral nitrogen at 45 days from planting. This high dry weight was significantly increased to 4.34 and 5.0 fold higher than those in the control (1 and 2), respectively.

After 75 days of sowing the highest dry weight was recorded by compost "C" provided with cellulose decomposers in the presence of biofertilization combined with the mixture of diazotrophs and (AM)-mycorrhizal fungi plus half dose of chemical nitrogen. It gave 10.20g plant<sup>-1</sup> and increased to 2.31 and 3.10 fold more those of control (1 and 2), respectively. On the other hand, after 120 days of sowing, the optimum dry weight yield was obtained with compost "A" provided with cellulose decomposers in the presence of biofertilization plus half dose of chemical nitrogen, it represented 34.90g plant<sup>-1</sup> and increased to 1.19 and 2.92 fold higher than control (1 and 2), respectively. Data showed better response of wheat to compost treated with cellulose decomposers and also showed the positive effect of inoculation with N<sub>2</sub>-fixers on the dry weight of wheat plants in all periods.

Inoculation with composite of diazotrophs and (AM)-mycorrhizae in presence of compost as an organic matter combined with half recommended dose of mineral nitrogen led to an increase of total dry weight of wheat plants. The addition of compost increased the N<sub>2</sub>-fixers densities, which in turn stimulated the infection of (AM)-mycorrhizal fungi in wheat roots that stimulated plant growth and consequently total dry weight yield through the enhancing of microbial and fungal activities in root region. This result agrees with the findings of Massoud (1999), Sabry *et al.* (2000) and El-Gahdban *et al.* (2002).

### **c) Number of tillers plant<sup>-1</sup> of wheat:**

Results in Table (2) showed that the inoculation of wheat plants with N<sub>2</sub>-fixers in the presence of (AM)-mycorrhizal fungi and compost as an organic matter increased the number of tillers as compared with control (1 and 2). The highest number of tillers plant<sup>-1</sup> obtained by compost "B" amended with organic manure and biofertilizers, where it recorded 5.45 tillers plant<sup>-1</sup>. It increased to 3.6 and 5.45 fold higher than those in the control (1 and 2), respectively.

Increasing the tillering at the period of 75 days, in particular, was due to the positive interaction between bacteria and fungi, which can improve plant nutrition, this in fact reflected on plant height, total dry weight and number of tillers particularly in the presence of compost as organic matter, these results are in harmony with those obtained by Abdalla *et al.* (1992), Massoud (1999) and Harridy *et al.* (2001).

### **2. Effect of compost type, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on infection percentage (%) of (AM)-mycorrhizae on the roots of the wheat plants:**

Results in Table (3) revealed that after 45 days of wheat sowing, the percentage of AM root infection was high with compost "A" amended with organic manure, cellulose decomposers and biofertilizers. The corresponding percentage was 84, the same trend was also obtained with compost "C" amended with cellulose decomposers and biofertilizers. The percentage increased to 1.58 and 3.82 fold higher than those in the control (1 and 2), respectively.

At the period of 75 days of sowing the percentage of (AM)-mycorrhizae slightly increased with the application of different compost types. Whereas, a

marked decrease in the percentage of (AM)-mycorrhizae was observed at 120 days from wheat sowing, for all treatments.

Inoculation with (AM) fungi may increase the rate of infection and hence mycorrhizal symbiosis influenced both soil and plant development. The highest infection was recorded with compost treated with either cellulose decomposers or organic manure in presence of N<sub>2</sub>-fixers as biofertilizers. Miller (1994) reported that organic matter represented in compost may contain substrates, which stimulate the growth of fungi and provide growth substances for these fungi. In addition, Thomas *et al.* (1999) stated that the rhizosphere of plant in natural involves more complex interactions than those between a plant and a mycorrhizal fungus. They also added that there are increased indications that bacteria are considered to be the most abundant in soils in root-mycorrhizal fungus interactions in the presence of organic matter. However, mycorrhizae act a helper for bacteria that enhance the extention of fungal colonization. This supports the evidence that N<sub>2</sub>-fixing bacteria in presence of compost enhanced mycorrhizal root colonization due to the role of these bacteria in promoting plant growth and nutrient uptake.

**Table (3): Effect of different compost heaps amended with organic manure, cellulose decomposers or biofertilizers on wheat (AM)-mycorrhizae percent infection and nitrogenase activity in the rhizosphere at different agricultural periods**

Compost	Parameter Treatments	Mycorrhizae infection (%)			Nitrogenase activity (µmole C <sub>2</sub> H <sub>4</sub> g <sup>-1</sup> root dry weight h <sup>-1</sup> )		
		45 days	75 days	120 days	45 days	75 days	120 days
C <sub>1</sub>		53.0	56.7	32.30	2.050	90.50	88.30
	C <sub>2</sub>	22.0	25.7	18.00	0.960	39.20	22.40
A	T <sub>1</sub>	84.0	84.0	65.00	2.36	126.63	40.20
	T <sub>2</sub>	78.3	73.0	61.00	1.56	440.30	150.03
	T <sub>3</sub>	82.3	81.7	73.30	2.65	546.50	204.6
	T <sub>4</sub>	69.3	69.0	55.70	2.46	3.15.30	85.43
B	T <sub>1</sub>	75.0	72.7	60.33	2.75	94.28	30.50
	T <sub>2</sub>	67.0	67.3	56.33	2.75	434.70	133.50
	T <sub>3</sub>	83.7	83.3	72.00	3.68	604.30	219.00
	T <sub>4</sub>	68.7	68.7	51.00	2.19	203.06	71.10
C	T <sub>1</sub>	77.0	82.3	61.00	2.647	135.06	33.50
	T <sub>2</sub>	71.0	77.7	54.00	3.227	267.00	80.80
	T <sub>3</sub>	84.0	84.0	68.00	1.072	235.00	85.60
	T <sub>4</sub>	70.7	68.0	48.00	2.248	318.80	94.60
D	T <sub>1</sub>	84.3	82.7	64.00	2.224	272.60	141.30
	T <sub>2</sub>	80.0	72.7	52.70	1.776	244.00	82.10
	T <sub>3</sub>	84.0	87.7	62.3	2.300	301.00	83.20
	T <sub>4</sub>	68.7	75.7	59.66	2.927	170.00	56.00
<b>LSD 0.05:</b>							
Inoculation		<b>2.585</b>	<b>2.528</b>	<b>3.338</b>	<b>0.37</b>	<b>0.678</b>	<b>1.73</b>
Treatment		<b>2.150</b>	<b>1.700</b>	<b>1.270</b>	<b>ns</b>	<b>1.261</b>	<b>1.61</b>
Compost		<b>1.965</b>	<b>1.800</b>	<b>1.180</b>	<b>n.s</b>	<b>2.160</b>	<b>2.29</b>
<b>Interaction:</b>							
Inoculation x Treatment		<b>n.s</b>	<b>n.s</b>	<b>4.603</b>	<b>0.38</b>	<b>1.938</b>	<b>3.340</b>
Inoculation x Compost		<b>4.365</b>	<b>4.33</b>	<b>4.520</b>	<b>0.38</b>	<b>2.880</b>	<b>4.011</b>
Treatment x Compost		<b>4.121</b>	<b>3.40</b>	<b>2.410</b>	<b>n.s</b>	<b>3.490</b>	<b>3.902</b>
Inoculation x Treatm x Compost		<b>6.700</b>	<b>6.23</b>	<b>5.790</b>	<b>0.39</b>	<b>4.055</b>	<b>4.642</b>



The formation of vesicles and arbuscules at the period between 45 – 75 days old in all treatments referred to three essential stages of the vesicular arbuscular mycorrhizae, where fungal life cycle, could be followed in different parts of the wheat roots. Stage one was characterized by predominance of arbuscules, this stage was observed in young parts of secondary roots. Stage two was characterized by a predominance of vesicles, which, observed in older parts of the root branches and the main roots. Stage three, spores formation was predominant. The fungus produced massive amounts of spores, enclosing the vascular cylinder of root parts older than 5 weeks (Verma and Arya, 1998).

**3. Effect of different compost types, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on nitrogenase activity (N-ase) in the rhizosphere of wheat plants:**

Results in Table (3) revealed that, the highest (N-ase) activity was recorded in wheat rhizosphere plants treated with compost "B" provided with cellulose decomposers and biofertilizers after 45 days of sowing where N-ase value was 3.68  $\mu\text{mol C}_2\text{H}_4 \text{ g}^{-1} \text{ dry root h}^{-1}$ . It increased to 1.80 and 3.83 fold higher than those the control (1 and 2), respectively. After 75 days period a sharp increase in nitrogenase activity was recorded the best results was obtained with compost "A" and compost "B" provided with cellulose decomposers and biofertilizers. At these treatments, the enzyme activity increased to 6.04 and 6.7 times higher than those in the control (1 and 2), respectively. After 120 days of sowing, the enzyme activity markedly increased compared to that it after 75 days. However, the results were still higher than after 45 days from sowing.

Obviously, the response of wheat rhizosphere plants to compost treated with either cellulose decomposers or organic manure and biofertilizers especially after 75 days led to an increase of nitrogenase enzyme activity. This is because the favorable effects of the combination between compost and biofertilizers. This increase in the production of some growth regulators, auxins and vital enzymes involving nitrogenase. Where, the nitrogenase efficiency increases with increasing the efficiency of N<sub>2</sub>-fixing bacteria. The present results are in agreement with those obtained and discussed by Pandey *et al.* (1998), Massoud *et al.* (1999) and El-Gahdban *et al.* (2002). The decrease in nitrogenase activity in control "1" may be due to the increase of mineral nitrogen that inhibited N<sub>2</sub>-fixers to fix atmospheric nitrogen. These results were previously shown by Kefologranni and Aggelis (2002) who reported that nitrogenase enzyme activity was strongly inhibited by mineral nitrogen.

**4. Effect of different compost types, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on nitrogen percentage of wheat plant:**

Data in Table (4) revealed that at 45 days period, the application of compost "B" provided with organic manure and biofertilizers gave the highest wheat nitrogen percentage where it was 4.09. Compost "B" recorded significant increases in nitrogen percentage as compared with control (1 and 2) where it increased to 1.18 and 2.40 fold, respectively. After 75 days of planting, the highest nitrogen percentage was recorded with compost "D" provided with organic manure and biofertilizers, this was followed by compost

"B" and "C" they represented approximately two times higher than that in the control (1). On the other hand, at the period of 120 days from sowing, the nitrogen percentage was decreased to approximately two times lower than at 75 days. The optimum percentage of nitrogen in the plant was obtained at 75 days.

The positive response of wheat plants to compost application and inoculation with mixed culture of diazotrophs and AM mycorrhizal fungi was accompanied by significant increases in shoots dry weight as well as nitrogen content. The combination of N<sub>2</sub>-fixers composite and (AM)-mycorrhizal fungi in the presence of compost led to the increase of nitrogen percentage on wheat plants. Application of a mixed biofertilizers with compost showed a superior affect on soil nitrogen availability, these results may be due to the positive effect of organic materials on the biological activity. These results are in agreement with those obtained by El-Gahdben *et al.* (2002), Eghball *et al.* (2003) and Ginting *et al.* (2003).

**5. Effect of different compost types, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on percentage of wheat potassium content:**

Data in Table (4) showed that at the period of 45 days old the highest potassium content (%) in plant was obtained with compost "D" provided with organic manure and biofertilizers. The respective potassium percentage was 5.66 followed by compost "C" and compost "A" where they both amended with the same amendments as compost "D". Their corresponding potassium percent were 5.53 and 5.51, respectively.

On the contrary, after the periods of 75 and 120 days of planting it was a sharp decrease in potassium content in plant was occurred. This may be due to consumption of K in the early age of the plant. The increase of total potassium content in shoot might be due to the positive response of wheat to compost as an organic matter where organic matter amendments can improve soil properties and crop quality as well as stimulating soil microbial populations and soil biological activity (Brady and Weil, 1999). Addition of compost increased total organic matter, macronutrient and micronutrient. However, the addition of compost to soil and the response of wheat to inoculation with diazotrophs plus (AM) mycorrhizae led to a significant increase in plant growth compared to control including increasing phosphorus, nitrogen and potassium contents. The results obtained are in the same harmony with those given by Wong *et al.* (1999) and Lee *et al.* (2004). These results are also in agreement with those obtained by El-Mohandes (1999) and Biswas *et al.* (2000).

**6. Effect of different compost types, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on percentage of wheat phosphorous content:**

Results presented in Table (4) showed that the highest phosphorous percentage in plant was recorded after 45 days of planting with compost "A" amended with cellulose decomposers and biofertilizers which gave 0.070, where it increased to 2.3 and 3.5 fold higher than that at control (1 and 2), respectively. With respect to 75 days old, the highest phosphorous percent was recorded with compost "B" treated with cellulose decomposers and biofertilizers where it exhibited 3.03%. It increased to 1.90 and 4.32 fold, higher than those in the control (1 and 2), respectively. There were significant

differences between the interaction of compost, treatments and biofertilizers. After 120 days from sowing the same trend was remarked where the optimum "P" percentage was also obtained with compost "B" amended with cellulose decomposers and biofertilizers. It was obvious that the phosphorous contents were higher at 75 days old than those at the period of 45 and 120 days, respectively. On the other hand, the interaction between compost, and biofertilization indicated positive significant differences.

**Table (4): Effect of different compost heaps amended with organic manure, cellulose decomposers or biofertilizers on percentage of total N, P and K of wheat plants at different agricultural periods**

Compost	Parameter	Total N (%)			Total K (%)			Total P (%)		
		45 days	75 days	120 days	45 days	75 days	120 days	45 days	75 days	120 days
C	Treatments									
	C <sub>1</sub>	3.45	3.73	2.88	4.30	1.25	1.07	0.030	1.60	1.40
	C <sub>2</sub>	1.76	2.63	0.69	3.30	1.20	1.01	0.020	0.70	0.11
A	T <sub>1</sub>	3.73	5.43	2.61	5.51	1.34	0.86	0.060	1.50	1.55
	T <sub>2</sub>	3.66	4.17	1.99	5.23	1.27	0.84	0.050	1.33	1.36
	T <sub>3</sub>	3.45	4.43	2.81	5.37	1.18	0.82	0.070	1.27	0.91
	T <sub>4</sub>	3.14	4.23	2.86	4.93	1.35	0.85	0.040	1.78	1.67
B	T <sub>1</sub>	4.09	6.27	2.55	5.45	1.37	0.95	0.050	2.23	1.74
	T <sub>2</sub>	3.60	5.80	2.42	5.40	1.51	0.92	0.060	1.57	1.36
	T <sub>3</sub>	3.23	5.50	2.43	5.45	1.84	0.97	0.060	3.03	1.84
	T <sub>4</sub>	3.61	5.50	2.38	5.44	1.22	0.84	0.040	1.47	1.40
C	T <sub>1</sub>	3.73	6.20	2.63	5.53	1.44	0.83	0.060	1.97	1.52
	T <sub>2</sub>	3.68	4.81	2.39	5.03	1.43	0.85	0.050	1.43	0.92
	T <sub>3</sub>	3.46	5.46	2.36	5.40	1.49	0.89	0.050	1.37	1.03
	T <sub>4</sub>	3.08	4.50	2.63	4.95	1.38	0.82	0.040	1.93	1.46
D	T <sub>1</sub>	3.44	6.30	2.59	5.66	1.60	0.97	0.063	1.52	1.33
	T <sub>2</sub>	3.44	5.73	2.49	5.30	1.50	0.87	0.057	1.60	1.23
	T <sub>3</sub>	3.47	5.57	2.37	5.41	1.45	0.89	0.063	1.30	1.07
	T <sub>4</sub>	3.16	5.20	2.14	4.57	1.29	0.85	0.043	1.67	1.14
<b>LSD 0.05:</b>										
Inoculation		<b>0.191</b>	<b>0.159</b>	n.s	<b>0.0953</b>	<b>0.088</b>	<b>0.007</b>	<b>0.0032</b>	<b>0.086</b>	n.s
Treatment		<b>0.191</b>	<b>0.159</b>	<b>0.060</b>	<b>0.0953</b>	n.s	<b>0.007</b>	<b>0.0032</b>	<b>0.086</b>	n.s
Compost		<b>0.269</b>	<b>0.226</b>	<b>0.085</b>	<b>0.1347</b>	n.s	<b>0.009</b>	<b>0.0045</b>	<b>0.121</b>	<b>0.139</b>
<b>Interaction:</b>										
Inoc. x Trea,		n.s	<b>0.318</b>	<b>0.060</b>	<b>0.190</b>	<b>0.130</b>	<b>0.014</b>	<b>0.008</b>	<b>1.80</b>	<b>0.180</b>
Inoc. x Compost		n.s	<b>0.385</b>	<b>0.085</b>	<b>0.180</b>	n.s	<b>0.016</b>	<b>0.008</b>	<b>0.210</b>	n.s
Treat.t x Compost		<b>0.420</b>	<b>0.385</b>	<b>0.140</b>	<b>0.210</b>	n.s	<b>0.016</b>	n.s	<b>0.206</b>	n.s
Inocu.x Treatm x Compost		<b>0.550</b>	<b>0.535</b>	<b>0.141</b>	n.s	<b>0.210</b>	<b>0.023</b>	n.s	<b>0.292</b>	<b>0.319</b>

Regarding the main effect of compost, (AM)-mycorrhizae and N<sub>2</sub>-fixers on plant phosphorus content, the data revealed that the addition of compost to soil can result in increased soil concentrations of nutrients and organic matter. Those results are in agreement with those obtained by Eghball *et al.* (2003). Phosphorus concentration increased in shoots especially at the period of 75 days old, and this result is in agreement with Mahammed *et al.* (1998) who reported that (AM) and N<sub>2</sub>-fixers inoculation with organic matter increased grain yield more than 25% only when mineral phosphorus was not added to the soil. However, the beneficial effect of (AM)

on nutrient uptake may account for the increase in plant growth and grain yield.

**7. Effect of different compost types, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on yield parameters:**

**a) Weight of 1000-grains:**

Results in Table (5) indicated that compost "B" treated with organic manure + biofertilizers gave the best weight of 1000-grains where it recorded 71.40g followed by compost "A" amended with the same amendments as compost "B" where the 1000-grains weight was 62.23g. Other treatments as well as control "1" and control "2" gave low weight of 1000-grains. The wheat grain yield significantly increased with compost "A" and "B" and exhibited 1.18 and 1.35 times higher than in the control (1), respectively. It was also obvious that there were significant differences among biofertilization treatments and compost types.

**b) Grain yield (ard. Fed<sup>-1</sup>):**

Data in Table (5) indicated that the highest grain yield of wheat obtained with compost "B" provided with organic manure plus biofertilizers where it exhibited 16.94ard feddan<sup>-1</sup>. The same compost when treated with cellulose decomposers plus biofertilizers gave 16.1 ard. Feddan<sup>-1</sup>.

**Table (5): Effect of different compost heaps amended with organic manure, cellulose decomposers or biofertilizers on number of tillers (plant<sup>-1</sup>), 1000-grain weight (g), grain yield (ard. fed<sup>-1</sup>) and straw yield (ton fed<sup>-1</sup>)**

Compost	Parameter	1000-grain weight (g)	Grain yield (ard. fed <sup>-1</sup> )	Straw yield (ton fed <sup>-1</sup> )
	Treatments			
C	C <sub>1</sub>	52.60	12.20	3.45
	C <sub>2</sub>	25.80	2.52	2.15
A	T <sub>1</sub>	62.23	8.36	4.80
	T <sub>2</sub>	55.33	6.75	4.76
	T <sub>3</sub>	41.17	8.20	5.60
	T <sub>4</sub>	47.47	6.78	3.73
B	T <sub>1</sub>	71.40	16.94	6.79
	T <sub>2</sub>	43.47	6.95	4.65
	T <sub>3</sub>	47.03	16.10	5.95
	T <sub>4</sub>	50.60	5.00	5.60
C	T <sub>1</sub>	43.23	14.94	5.19
	T <sub>2</sub>	37.60	5.08	4.50
	T <sub>3</sub>	37.57	13.15	3.50
	T <sub>4</sub>	47.33	6.85	3.72
D	T <sub>1</sub>	44.40	15.26	5.34
	T <sub>2</sub>	44.70	6.09	4.65
	T <sub>3</sub>	32.00	13.12	5.49
	T <sub>4</sub>	30.60	6.33	4.91
<b>LSD 0.05:</b>				
Inoculation		<b>0.446</b>	<b>0.057</b>	<b>0.009</b>
Treatment		<b>0.446</b>	<b>0.057</b>	<b>0.009</b>
Compost		<b>0.631</b>	<b>0.018</b>	<b>0.012</b>
<b>Interaction:</b>				
Inoc. x Trea,		<b>0.892</b>	<b>0.114</b>	<b>0.018</b>
Inoc. x Compost		<b>1.070</b>	<b>0.071</b>	<b>0.021</b>
Treat.t x Compost		<b>1.070</b>	<b>0.071</b>	<b>0.021</b>
Inocu. x Treatm x Compost		<b>1.711</b>	<b>0.134</b>	<b>0.030</b>

On the other hand, control "1" (full dose of NPK) recorded 12.20 and feddan<sup>-1</sup> more than some of the other treatments. Compost "B" provided with organic manure increased to 1.38 and 6.72 fold higher than those in the control (1 and 2), respectively. It was also obvious that there were significant differences among the interaction of compost, treatment and fertilization.

**c) Straw yield (ton Fed<sup>-1</sup>):**

Table (5) shows the effect of using compost type, (AM)-mycorrhizae and N<sub>2</sub>-fixing bacteria on straw yield (ton fed<sup>-1</sup>) of field grown wheat as compared to control (1) (recommended dose of NPK) and control (2) (plain soil without any additions). It is obvious that compost "B" amended with organic manure and biofertilizers recorded the optimum yield of straw higher than all other treatments where its value recorded 6.79 ton feddan<sup>-1</sup>.

It significantly increased to 1.97 and 3.16 fold higher than in the control (1 and 2), respectively. Data also showed that there were significant differences between the interaction of inoculation with biofertilization, compost type and treatments.

These results are in accordance with those obtained by Mekail (1998) and also, Lee *et al.* (2004) who reported that there are significant increases in yields (grain, straw and biological yields) of some crop such as lettuce and wheat due to organic materials addition as compared to control. They also reported that these significant increases in wheat yield could be attributed to the rapid decomposition of these organic materials due to narrow C/N ratio. Also, Khamis and Metwally (1998) reported that incorporation of organic materials enriched with microbial decomposers, (AM) mycorrhizae fungi and nitrogen fixers improved their decomposition in soil and increased grain and straw yield of wheat.

The results of this study are also in occurrence with those obtained by Badran (2002) who found that the highest components (grain, straw and biological) of wheat and barley plants under sandy soil conditions were obtained by combining organic materials such as compost and 50% N. This author also demonstrated that the addition of 50% N gave yield better than 100% N (complete fertilizer dose). Addition of organic materials, biofertilizers and N application were also found to be necessary to obtain maximum wheat yield (Blaise *et al.*, 2005).

**8. Effect of different compost types, AM (mycorrhizae) and N<sub>2</sub>-fixing bacteria on some chemical properties of sandy soil after harvesting:**

Data in Table (6) showed that the application of any compost type caused a slight decrease in pH values compared to un-amended control. With respect to EC results showed an increase in EC due to application of compost with 1/2 dose of mineral nitrogen. Concerning the organic carbon and organic matter data presented in Table (6) show an increase of these parameters due to the application of any compost types.

**Table (6): Physico-chemical properties of the soil samples after wheat harvesting**

Properties	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	C <sub>1</sub>	C <sub>2</sub>
<b>Mechanical (%):</b>						
Sand	70.20	69.30	68.75	70.10	70.00	70.17
Silt	2.10	2.00	1.95	2.00	2.10	2.10
Clay	27.70	28.50	31.25	27.9	27.90	27.73
<b>Chemical:</b>						
Organic carbon (%)	0.55	1.01	1.05	0.95	0.52	0.15
Total nitrogen (%)	0.11	0.10	0.09	0.09	0.11	0.03
Total phosphorous (ppm)	0.09	0.08	0.075	0.05	0.07	0.01
C/N ratio	5:1	10.1:1	11.4:1	10.05:1	4.7:1	5:1
Water holding capacity (%)	25.90	28.00	28.31	27.90	25.00	25.00
pH	7.84	7.62	7.82	7.65	7.71	7.76
EC <sub>e</sub>	0.24	0.34	0.34	0.30	0.26	1.35
CaCO <sub>3</sub> (%)	0.60	0.63	0.65	0.60	0.60	0.60
<b>Anions and cations (meq / L):</b>						
Carbonate	traces	traces	traces	Traces	Traces	Traces
Bicarbonate	1.14	1.43	1.71	1.00	1.45	0.09
Chloride	0.20	0.40	0.40	0.30	0.20	0.20
Sulphate	0.12	0.22	0.09	0.08	0.03	0.06
Calcium	0.64	0.85	0.64	0.60	0.64	0.64
Magnesium	0.30	0.09	0.49	0.35	0.49	0.11
Sodium	0.31	0.55	0.50	0.41	0.33	0.28
potassium	0.29	0.56	0.54	0.30	0.20	0.10

With respect to total nitrogen, data show increases in total nitrogen and also increasing the available phosphorus and potassium. Application of compost can improve physical and chemical properties of soil after harvesting and consequently increase soil fertility and quality for several years as reported by Ginting *et al.* (2003). They stated that increased levels of N, P, K, organic carbon and organic matter in soil can increase crop yield beyond the application years. The same trend was also recorded by Eghball *et al.* (2003) who found that the increased plant-available P-level in soil following N-based manure or compost application combined with (AM) mycorrhizae can contribute to crop P-uptake for up to 10 years without any additions. The present results are also in agreement with those obtained by Ginting *et al.* (2003) and Eghball *et al.* (2003). The slight decrease in soil pH by application of organic materials may be due to the organic acids produced from the decomposition of these organic materials and similar trend was obtained by Mikhaeel *et al.* (1997).

The increase in EC in sandy soil could be attributed to the release of soluble salts of composted materials during their decomposition in soil. These findings agree with those obtained by Abdel-All (2001) who reported that the increasing in EC could be attributed to the dissolving action of organic acids on the native salts in soils. The increases in the organic carbon and organic matter of the compost-treated soil could be due to the release of organic compounds during the mineralization of organic materials. As assumed by Mekail (1998). Moreover, the application of composted plant residues tended to improve the C/N ratio of sandy soil. In this concern, Abdel-All (2001)

reported that the application of organic materials to sandy soils produced narrow C/N ratio.

Finally, the increase in N, P and K contents may be due to the decomposition processes with the help of AM (mycorrhizae) which release their nutrients. Also, the increase in the W.H.C. would enhance the solubility and consequently the availability of nutrients as well as the retention of K by organic colloid against leaching. These results are in agreement with Mekail and Zarouny (1998) who documented that the addition of different kinds of organic materials to sandy, calcareous and clay soils increased both soil moisture retention and the availability of phosphorus and potassium.

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## تأثير الكمبوست وفطريات الميكوريزا ومثبتات النيتروجين الحوى على نمو ومحصول القمح

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مركز البحوث الزراعية - معهد بحوث الأراضى والمياه والبيئة - قسم بحوث الميكروبيولوجيا الزراعية

أجريت تجربة حقلية فى أرض رملية بمحطة بحوث الإسماعيلية، والتابعة لمركز البحوث الزراعية؛ لدراسة تأثير إضافة : الكمبوست ( المعامل بمحلات السليولوز أو السماد البلدى ) ، وفطريات الميكوريزا ، وخليط من مثبتات النيتروجين الجوى ، على نمو ومحصول القمح ، وأظهرت النتائج المتحصل عليها أن : أحسن كثافة عددية للميكروبات المثبتة لنيتروجين الهواء الجوى وجدت عندما عوملت التربة باللقاح الحيوى ( مثبتات النيتروجين + فطريات الميكوريزا ) + الكمبوست المزود بمحلات السليولوز بعد 54 ، 57 يوماً من الزراعة ، كذا وجد زيادة فى أطوال النباتات عند معاملة التربة باللقاح الحيوى + الكمبوست المزود بالسماد البلدى ، وكانت الزيادة معنوية فى الوزن الجاف للنباتات عندما عوملت التربة بالسماد الحيوى + الكمبوست المزود بمحلات السليولوز ، أكبر زيادة فى التفرعات النامية من الجذور وجدت عندما عوملت التربة باللقاح الحيوى + الكمبوست B والمزود بالسماد البلدى .

سجلت أعلى إصابة للجذور بفطريات الميكوريزا عند معاملة التربة بالكمبوست A المزود بالسماد البلدى، وكان أعلى نشاط لإنزيم النيتروجينيز عند المعاملة باللقاح الحيوى + الكمبوست B المزود بمحلات السليولوز ، وكانت هناك زيادة معنوية للنسبة المئوية للنيتروجين فى النباتات عند معاملة التربة باللقاح الحيوى + الكمبوست D المزود بالسماد البلدى ، التلقيح باللقاح الحيوى أدى إلى زيادة النسبة المئوية للفوسفور فى النباتات خاصة عند إضافة الكمبوست B والمزود بمحلات السليولوز ، وكذا زيادة النسبة المئوية للبتواسيوم عند استخد الكمبوست D المزود بالسماد البلدى بعد 54 يوماً من الزراعة .

أظهرت النتائج كذلك زيادة معنوية فى التقديرات المحصولية من خلال وزن الـ 0001 حبة ، ووزن محصول الحبوب ، ووزن القش ، حيث سجل أعلى محصول للحبوب عند المعاملة باللقاح الحيوى + الكمبوست B المزود بالسماد البلدى . بصفة عامة الجدير بالملاحظة هو الدول الضرورى للمادة العضوية ( الممتلئة بالكمبوست ) ومثبتات النيتروجين وفطريات الميكوريزا فى زيادة نمو ومحصول القمح .