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Impact of Biofertilizers Application on Improving Wheat (*Triticum aestivum* L.) Resistance to Salinity

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Abstract: A wire house experiments were conducted during the two successive seasons; 2005-2006 and 2006-2007, to investigate the effectiveness of some biofertilizers' treatments i.e., cerealien, phosphorien, cerealien and their combination on wheat (*Triticum aestivum* L., var. Seets) under different levels of salinity (0, 3000, 6000, 9000 ppm) for improving salinity tolerance. The obtained data confirmed the absolute superiority of the cerealien + phosphorien in inducing the highest degree of the physiological tolerance to salinity which enables the stressed plants of the Seets cultivar to be adapted and keep better performance against all applied levels of salinity. This performance was reflected by the increase in growth, dry matter accumulation, yield as well as chemical constituents. All chemicals constituents including N, P, K^+, sugars, proline and were increased as compared to their control treatments in the cultivar Seets.

Key words: Wheat, Salinity, Biofertilizers, NPK, Sugars, Proline.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the important cereal crops as the main food stable for the Egyptian public. Improving the productivity of this crop is a main task due to its short supply which mandated importing about 50% of the needed wheat grains from outside the country. There is insufficient fresh water to develop all potential arable land. So, the use of saline water agriculture is a subject of vital importance for arid and semi arid zones to meet increasing food demand[43]. Use of soil microorganisms which can either fix atmospheric nitrogen or solublize phosphate or plant growth through synthesis of growth promoting substances or by enhancing the decomposition of plant residues to release vital nutrients and increase humic content of soils, will be environmentally begin approach for nutrient management and ecosystem function[46]. Kaci *et al.*,[20] reported that, these microorganisms are known to deliver a number of benefits including plant nutrition and tolerance to adverse soil conditions.

The aim of this study is to investigate the response of wheat plant to nitrogen fixing bacteria and phosphate dissolving bacterial fertilizers under different levels of salinity. Moreover, investigating the effect of these biofertilizers combinations as a biological technique for reducing the harmful effect of salinity and find out the level of salinity that plant can be tolerate and their effects on the growth and yield. In addition, the use of biofertilizers may have additional benefits such as nitrogen fixation, mobilizing phosphate and micronutrients through the production of organic acids and lowering soil pH[37]. Besides, microorganisms such as *Pseudomonus*, *Azotobacter*, *Azospirillium* and mycorrhizae can secrete growth promoting factor, i.e., gibberellins, cytokinins like substance and auxins[18]. The strain of nitrogen fixing bacteria (*Azotobacter* spp.) under the commercial name cerealien and phosphate dissolving bacteria (*Bacillus megaterium*) under the commercial name phosphorien used under different levels of salinity. The previously mentioned treatments were evaluated for their effects on wheat plant growth, yield and chemical constituents. Usually biofertilizers contain one or more of following; symbiotic and/or non-symbiotic N-fixing bacteria or phosphorus dissolving bacteria such as *Bacillus megatherium*.

MATERIALS AND METHODS

Pot experiments were conducted during 2005 - 2006 and 2006 - 2007 seasons in the wirehouse of Plant Physiology Section, Faculty of Agriculture, Cairo University, Giza. The experiments include cultivar of wheat (*Triticum aestivum* L.) seets. Seeds were planted on November 15, 2005 and 2006 in a mixture of one clay: two sand soil. Plastic pots of 30 cm in diameter and 30 cm in depth were used in both experiments. Fertilization (mineral fertilizer) was carried out according to the recommended dose by the Ministry of Agriculture; the mineral fertilizers consisted of 2.2g of calcium superphosphate (15.5% P_2O_5), 1.1g potassium sulphate (48% K_2O) and 2.0g ammonium sulphate (20.5%N) in

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each pot before planting and added 2.0 g ammonium sulphate 30 days after planting. Four levels of salinity (0, 3000, 6000 and 9000 ppm) by using a mixture of sodium chloride, calcium chloride and magnesium sulphate at the ratio 2: 2: 1 respectively were used as the main plot. Each subplot has the treatments non-treated control, cerealien, phosphorien and cerealien + phosphorien. Both cerealien and phosphorien are produced by biofertilizers unit, General Organization of Agriculture Equalizaton Fund, Agriculture Research Centre, Ministry of Agriculture, Giza, Egypt. Wheat grains were planted in 5 groups/pot and each group consists of two grains (seeds). Grains before sowing were treated with the biofertilizers cerealien, phosphorien and cerealien + phosphorien in addition to the non-treated control treatment. Coating of wheat grains was conducted as recommended by the Ministry of Agriculture, Giza, Egypt. Emerging plants were thinned to 4 plants/pot after two weeks. Two vegetative samples were taken at 45 and 105 days after sowing (DAS). Another sample was harvested at 140 DAS for yield. In the first two samples, the plants were divided into shoots and roots and the following measurements were recorded: shoot height (cm), root length (cm), number of tillers/plant, number of leaves, dry weight of shoots and roots (g).

In the third sample, yield and yield components including number of spikes/plant and weight of 1000 grain were estimated at harvest; 140)DAS(. In order to obtain dry weight, plant materials were chopped into small pieces and weighed then were kept in an electric fan oven at 70 ^{0}C for 48 hr. after that dry materials were ground into fine powder using an electric Wily Mill grinder, mixed thoroughly and packed in air tight glass containers and were kept for chemicals analysis.

Determinations of total nitrogen, phosphorus and potassium were carried out on the ground dry material. The samples were digested in a mixture of sulfuric acid, salicylic acid hydrogen peroxide according to Linder[26]. Nitrogen was determined by using the modified “Micro Kjeldahl” apparatus of Parnas and Wagner as described by Pregl[34]. Phosphorus was determined spectrophotometerically by using stannous chloride method according to[1]. Potassium was determined using the flame photometer (ELE) UK. Proline concentration was determined as follow according to Bates *et al.*[8].

Reducing, non-reducing and total sugars in the different plant parts were determined by phosphomolybdic acid method according to[1].

Data of growth characters and yield components were statistically analyzed by using three factorial completely randomized design and the mean values were compared using the least significant difference test [New L.S.D.] at five % level[15]. All data are shown as a combined analysis for both seasons.

RESULTS AND DISCUSSION

Growth: All discussed data are the mean values of the two seasons. The mean values of shoot height of var. Seets are presented in Table (1). All biofertilizer treatments gave significantly higher in the shoot heights, as compared to the non-treated treatment “control” in both samples. Moreover, the combined phosphorien + cerealien treatment showed better treatment than the other two individual treatments in both samples. Generally, the results reveal that, biofertilization caused a significant increase in shoot height. These results are in agreement with those reported by Favilli *et al.*[13] and Abd Alla *et al.*[2]. Data in Table (1) show similar trend to that obtained with the shoot height. This indicates that the biofertilizer treatments enhanced the root length (cm), in the two successive samples.

These results indicated that phosphorien + cerealien treatments gave good performance and gave the highest values compared with the control in both samples. The increase of root length may be attributed to the increase in cell growth enhancement by plant growth hormone after inoculation with biofertilizers treatments. Sheteawi and Tawfik[40] recorded that addition biofertilizers mitigated the harmful effect of water stress. Also, Ramazan *et al.*[35] noticed that plant growth promoting bacteria inoculation increased plant height by 2.2 % - 24.6 % in wheat.

Rodriguez *et al.*[36] showed that salt exposed plant exhibited a reduction in shoot and root growth and biomass related to control plants. When coincident with high salinity, exposure to biofertilizers resulted in a reversal of shoot related responses to salt stress. NaCl reduced shoot length by 54 % and root length by 62 %.

As previously mentioned with the shoot height, an appreciable alleviation of the detrimental effects of salinity occurred due to the applied biofertilizer treatments. The obtained data in Table 1 concerning the mean values of root length as affected by the different treatments under all salinity levels clearly revealed that the increase of root length with phosphorien + cerealien was highly significant.

Data concerning number of tillers per plant of both cultivars as affected by biofertilization under different levels of salinity are presented in Table (2). The results indicate that the number of tillers per plant increased as plants grew older. Also, number of tillers increased with all biofertilizer treatments under different salinity levels.

The obtained data in Table (2) clearly revealed that the response of the produced number of tillers by the stressed wheat plants to the various salinity levels was similar to that previously described for their shoot height and root length as many workers suggested that the harmful effect of salts on plant growth and plant height may be attributed to high osmotic pressure of soil solution

Table 1: Mean values of shoot height and root length)cm(as affected by different biofertilizers treatments under salinity conditions (seasons of 2005-2006 & 2006-2007).

Shoot height

	Salinity level (ppm)									
	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Plant age (Days)	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Biofertilizers										
Control	56.22	54.12	51.11	45.85	51.83	69.23	66.23	60.06	52.11	61.91
Cerealien	61.27	58.14	53.12	50.21	55.66	72.55	68.33	61.45	55.45	64.44
Phosphorine	57.89	58.32	51.14	48.42	53.94	71.11	69.02	62.12	54.68	64.23
Cerealien+Phosphorine	64.61	65.32	61.02	55.62	61.64	85.23	80.89	76.33	68.24	77.67
Mean	59.99	58.97	54.18	50.03		74.53	71.12	64.99	57.62	
L.S.D value at 0.05										
Salinity (S)	0.543									
Biofertilizer (B)	0.543									
S x B	1.18									

Root length

	Salinity level (ppm)									
	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Plant age (Days)	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Biofertilizers										
Control	23.25	20.52	18.58	17.01	19.84	33.52	29.21	25.22	20.75	21.25
Cerealien	24.95	21.85	19.57	17.44	20.92	37.5	31.52	26.75	22.42	29.54
Phosphorine	25.53	22.21	20.46	18.54	21.69	36.86	30.44	25.49	21.72	28.63
Cerealien+Phosphorine	32.44	30.87	27.61	22.94	28.47	42.04	38.51	32.56	28.11	35.31
Mean	26.54	23.86	21.55	18.98		37.48	25.57	27.51		
L.S.D value at 0.05										
Salinity (S)	0.537									
Biofertilizer (B)	0.537									
S x B	0.812									

Table 2: Mean values of number of tillers and number of leaves as affected by different biofertilizers treatments under salinity conditions (seasons of 2005-2006 & 2006-2007).

Number of tillers

	Salinity level (ppm)									
	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Plant age (Days)	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Biofertilizers										
Control	3.00	2.60	2.00	1.30	2.23	3.85	3.30	2.70	2.00	2.96
Cerealien	61.27	58.14	53.12	50.21	55.66	72.55	68.33	61.45	55.45	64.44
Phosphorine	4.20	3.50	2.85	2.00	3.14	4.65	4.15	3.50	3.00	3.83
Cerealien+Phosphorine	5.70	5.30	5.15	5.00	5.37	6.50	6.00	5.30	5.15	5.74
Mean	4.30	3.67	3.25	2.65		4.93	4.36	3.70	3.28	
L.S.D value at 0.05										
Salinity (S)	0.3058									
Biofertilizer (B)	0.3058									
S x B	N.S.									

Table 2: Continued

Number of Leaves

	Salinity level (ppm)									
	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Plant age (Days)	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Biofertilizers										
Control	22.50	19.50	16.50	10.15	17.16	28.92	25.16	21.28	15.60	22.74
Cerealien	31.50	26.53	21.46	15.66	23.78	35.23	31.45	27.36	23.44	29.37
Phosphorine	32.53	24.75	22.24	14.23	23.43	36.18	28.81	25.84	22.85	28.42
Cerealien+Phosphorine	42.75	39.75	34.33	25.63	35.62	50.23	46.80	41.28	38.52	44.20
Mean	32.32	27.63	23.63	16.41		37.64	33.15	28.94	25.10	
L.S.D value at 0.05										
Salinity (S)	0.2078									
Biofertilizer (B)	0.2076									
S x B	N.S.									

which restricts the absorption of water by plant roots and/or to the toxic effects of certain ions present in soil solution[21,6,33]. The salt causes a slower rate or shorter duration of expansion of cells and this led to compromised the size of the leaves[45]. The overall effect of salinity on plants is the eventual shrinkage of leaf size, which leads to death of the leaf and finally the plant. Sultana *et al.*[41] reported that salinity can affedt germination, metabolism, the size of plants, branching, leaf size and overall plant anatomy. Salt also affects photosynthetic components such as enzyme and chlorophyll. The percentage germination of wheat grains as well as the growth criteria (plumule length) was significantly affected by salinity level (0, 50 and 100 mM NaCl). Similar results were reported by Barraclough and Kyte[7]. Also, Essa[12] noted that at salinity levels of 805 ds/m, a significant reduction in plant height was recorded in three tested cultivars of soybean. Similar results were also reported by Salem *et al.*[38] with faba bean cultivars. Creus *et al.*[9] observed that *Azosprillum brasilense* improved water status in wheat seedlings under salt and osmotic stresses, which could explain a better shoot growth and a faster elongation in inoculated wheat plants than in control.

In this connection, Hartmann[17] found that *Azosprillum brasilense* is highly tolerant to salt in pure cultures. Also, Alvarez *et al.*[9] observed that stimulating effect of *Azosprillum brasilense* inoculation on coleoptile growth speed has been observed in *Buck Ombu* seedlings, exposed to moderate water stress for 48 h and 90 h. Mashhoor *et al.*[28] found that application of a mixture of the tolerant *Azotobacter chroococcum* strains + EM1-inoculant resulted in the capability to withstand soil stress and gave higher growth of wheat plants, compared with the use of EM1-inoculant singly as a biofertilizers.

Yield: The means of yield components (number of spikes/plant and weight of 1000 grains) for cultivar Seets as affected by biofertilizer treatments under different salinity levels are presented in Tables 3. As for the biofertilizer treatments Table (3) cleared that biofertilizer inoculation generally increased Weight of 1000 grains. It is clear that inoculation of biofertilizers under different levels of salinity induced an increase in number of spikes/plant. The beneficial effect of biofertilizer on the yield of wheat plant grown under saline condition was the highly significant increase in the grain yield. This explains the increasing of tillering capacity of the inoculated plants with biofertilizers. Increasing salinity level in the irrigation water decreased growth and yield components of wheat plants. These results agree with those reported by Kumar *et al.*[24], Hank *et al.*[16], Mass and Crieve[29] and Frankline *et al.*[14].

Moreover, Munns and Rawsan[31] found that salinity decreased of spikelet primordial formation and final spikelet numbers at spike emergence were reduced. Also, Zeng *et al.*[47] reported that, highly significant linear responses were found between salinity and rice grain weight per plant, grain weight per panicle, spiklet number per panicle and tiller number per plant. Tiller number per plant and spikelet number per panicle contributed for the most variation in grain weight per plant under salinity. Reduction in seedling survival, tiller number per plant and spikelet number per panicle were the major causes of yield loss under salinity. Khater *et al.*[22] reported that salinity significantly reduced the values of spike length and grain yield of wheat plant. Also, Tawfik[42] noticed that water stress reduces plant growth and yield. However water stress that exists at the reproductive stage affects grain yield of mungbean more than its occurrence at other stage[44].

Table 3: Mean values of number of spikes/plant and weight of 1000 kernel as affected by different biofertilizers treatments under different salinity levels (seasons of 2005-2006 & 2006-2007).

	Salinity level (ppm)									
	1st Sample (45 D.A.S.) number of spikes/plant					2nd Sample (105 D.A.S.) weight of 1000 kernel				
Plant age (Days) Biofertilizer	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	4.51	3.30	2.17	0.82	2.70	18.85	16.46	12.18	6.17	13.41
Cerealien	6.27	4.61	4.24	2.55	4.42	25.49	20.19	18.24	11.06	18.75
Phosphorine	6.20	4.70	3.52	2.74	4.29	24.50	21.05	17.50	11.52	18.64
Cerealien+Phosphorine	8.10	7.15	5.67	4.11	6.26	29.23	25.21	22.14	18.65	23.81
Mean	6.29	4.92	3.85	2.60		2.56	3.13	3.97	4.72	
L.S.D value at 0.05 Salinity (S)	0.2643	0.4921								
Biofertilizer (B)	0.2111	0.4921								
S x B		1.125								

Table 4: Effect of biofertilizers treatments on nitrogen, phosphorus and potassium concentration (mg/g dw) in the shoot of wheat plants under saline condition during seasons 2005-2006 & 2006-2007.

	Salinity level (ppm) Nitrogen									
	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Plant age (Days)	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	21.23	19.31	16.52	14.03	17.77	29.45	25.50	21.42	17.56	23.48
Cerealien	28.65	25.19	19.22	16.24	22.33	32.56	29.43	25.45	21.96	27.35
Phosphorine	27.80	24.52	17.30	14.95	21.14	31.92	28.78	24.63	19.85	26.29
Cerealien+Phosphorine	30.11	27.43	23.15	19.68	25.12	35.11	32.52	29.88	26.55	31.02
Mean	26.95	24.11	19.05	16.23		32.26	29.05	25.34	21.48	
	Phosporus									
	Salinity level (ppm)					Salinity level (ppm)				
	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	2.53	2.31	2.23	1.50	2.14	3.75	3.21	3.01	2.50	3.12
Cerealien	2.90	2.60	2.42	2.13	2.51	5.23	4.87	4.33	3.25	4.42
Phosphorine	3.09	2.80	2.44	2.20	2.63	5.34	5.08	4.76	3.62	4.70
Cerealien+Phosphorine	3.62	3.20	3.08	2.51	3.10	5.96	5.52	5.11	4.28	5.22
Mean	3.04	2.73	2.54	2.08		5.07	4.67	4.30	3.41	
	Potassium									
	Salinity level (ppm)		Salinity level (ppm)							
	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	22.30	19.90	18.40	14.70	18.83	28.75	25.32	22.56	17.80	23.61
Cerealien	27.24	24.11	19.87	12.45	20.92	33.68	31.42	24.50	19.23	27.21
Phosphorine	27.41	25.23	19.69	12.43	21.19	34.05	31.85	25.11	19.45	27.62
Cerealien+Phosphorine	31.23	28.69	23.54	15.92	24.85	37.56	34.20	27.41	22.51	30.42
Mean	27.05	24.48	20.37	13.87		33.51	30.69	24.89	19.75	

Table 5: Effect of biofertilizers treatments on reducing, non-reducing and total sugars concentration (mg glucose/g dw) in the shoot of wheat plants under saline condition during 2005-2006 & 2006-2007 seasons.

	Salinity level (ppm) Reducing Sugars									
	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Plant age (Days)	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	20.51	24.34	27.54	30.77	25.79	29.56	35.23	43.52	48.75	39.27
Cerealien	28.62	32.42	35.42	40.32	34.20	40.13	44.34	47.23	52.78	46.12
Phosphorine	27.86	31.53	36.08	41.12	34.15	39.78	43.12	46.45	53.21	45.64
Cerealien+Phosphorine	30.21	34.51	38.45	42.73	36.48	44.50	46.07	50.20	56.23	49.25
Mean	26.80	30.70	34.37	38.74		38.49	42.19	46.85	52.74	
	Non-reducing sugars									
	Salinity level (ppm)					Salinity level (ppm)				
	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	22.86	27.64	30.41	34.82	28.93	31.45	38.56	48.23	55.13	43.34
Cerealien	31.66	36.23	42.51	45.62	39.01	44.72	53.46	58.25	69.65	56.52
Phosphorine	32.50	37.05	41.64	44.53	38.93	43.03	51.42	60.21	70.17	57.22
Cerealien+Phosphorine	34.56	39.25	45.61	49.62	42.26	48.23	56.24	62.34	74.22	60.26
Mean	30.40	35.04	42.29	43.65		41.86	49.92	57.26	67.29	
	Total Sugars									
	Salinity level (ppm)					Salinity level (ppm)				
	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	43.37	51.98	57.95	65.59	54.72	61.01	73.79	91.75	103.88	82.61
Cerealien	60.28	68.65	77.93	85.94	73.08	84.85	97.80	105.48	122.43	102.64
Phosphorine	60.36	68.58	77.72	85.65	73.08	82.81	94.54	106.66	123.38	102.86
Cerealien+Phosphorine	64.77	73.76	84.06	92.35	78.74	92.73	102.31	112.54	130.45	109.51
Mean	57.20	65.74	74.42	82.38		80.35	92.11	104.11	120.03	

El-Kased *et al.*[11] observed that application of biological fertilizer increased the yield by 183 % over the control. The grain yield is a function of number of tillers / m^2 and 1000 grain weight, while grain yield correspond fairly well with the number of tiller / m^2.

Application of biofertilizers such as (Azofert, phosfert, Bioplin and Vitormone) increased the average grain yield of wheat plant,[30,10]. Also, Sharma *et al.*[39] reported that application of biofertilizers)*Azotobacter* and phosphate solubilizing bacteria(increased yield of wheat.

Chemical Analysis: The obtained results showed the concentrations of nitrogen, phosphorus and potassium were adversely affected by increasing salinity level (Table 4). Hence, such concentrations tended to decreased gradually by increasing salinity level to reach their lowest values in the plants irrigated with 6000 ppm. By contrast, the stressed plant organs treated either with cerealien, phosphorien and cerealien + phosphorien showed much higher values of nutrients concentrations. Generally the concentrations of these elements were higher in the second sample as compared to the first sample. Biofertilizer inoculation generally increased the concentration of NPK as compared to control. However dual inoculation with cerealien and phosphorien insignificantly surpass the single one. Tawfik *et al.*[43] noticed that the effect of salinity on potassium and phosphorus content could be attributed to the difficulty of its uptake bue to competition with the high concentration of the sodium in the root medium.

Mahajan and Tuteja[27] said that potassium is one of the essential elements and is required by the plant in large quantaties. Potassium is required for maintaining the osmotic balance.

Regarding sugar concentration the obtained data clearly reveal that increasing salinity level caused a gradual increase in the concentrations of reducing, non-reducing and total sugars in the shoots to reach its maximum at the highest level of salinity i.e. 6000 ppm, among all samples (Table 5). On the other hand, response

Table 6: Effect of biofertilizers treatments on free proline concentration (mg/g fw) in the shoot of wheat plants under saline conditions during 2005-2006 & 2006-2007 seasons.

	Salinity level (ppm)									
Plant age (Days)	1st Sample (45 D.A.S.)					2nd Sample (105 D.A.S.)				
Biofertilizer	0	3000	6000	9000	Mean	0	3000	6000	9000	Mean
Control	1.62	1.80	1.97	2.13	1.88	2.09	2.77	3.15	3.42	2.86
Cerealien	2.05	2.71	3.26	3.90	2.98	2.57	3.18	4.12	5.08	3.74
Phosphorine	2.00	2.65	3.20	3.84	2.92	2.77	3.15	4.15	5.12	3.80
Cerealien+Phosphorine	2.21	3.02	3.42	4.18	3.21	2.79	3.42	4.44	5.27	3.98
Mean	1.97	2.55	2.96	3.51		2.56	3.13	3.97	4.72	

of sugar concentrations in shoots as affected by each treatment regardless the salinity level, clearly reveal that, the absolute superiority was confirmed for the cerealien + phosphorien treatment which recorded the highest considerable increments in the all cases over the respective values of the control. Ingram and Bartels[19] stated that under water stress soluble sugars can function in two ways which are difficult to separate as osmotic agent and as osmoprotectors. As an osmotic agent, the increased sugar induced by water stress was significantly correlated to osmotic adjustment and turgor maintainance. As osmoprotectors, sugar stabilizes protein and membranes.

Increasing salinity level caused gradual increase in proline concentration in the stressed shoots and the highest values was found at the high level of salinity as compared with the control treatment (Table 6). On the other hand, response of proline concentration in the treated organs with cerealien + phosphorien recorded the highest mean values when compared with other treatments. Kishore *et al*.[23] concluded that praline is known to occur widely in higher plants and normally accumulates in large quantities in response to environmental stress. Ashraf and Foolad[5] noticed that there is a positive relationship between praline accumulation and plant stress tolerance. Tawfik *et al*.[43] showed that raising irrigation salinity levels significantly increase the content of carbohydrate and praline. Similar results were obtained by Ashour *et al*.[4]. In this respect, Murphy *et al*.[32] suggested that both praline and soluble carbohydrates act as compatible solutes under high salinity levels. Kusaka *et al*.[25] added that, the observed increase in the osmotic potential might be due to the accumulation of inorganic solutes, several organic components such as sucrose, glucose and amino acids including praline.

Conclusion: It could be concluded that biofertilizers cerealien and phosphorien stimulated plant growth and yield and induced salinity tolerance by enhancing the accumulation of certain metabolites i.e. sugars and praline. Which are considered to be a sort of plant adaptation to stress. Accordingly, the present work was designed to investigate the effectiveness of biofertilizers applications which are reducing soil salinity while it increased the availability of N, P and K for improving the wheat cultivar, at different levels of salinity; 0, 3000, 6000 and 9000 ppm. It is of a great importance to mention here that the obtained data clearly revealed that, each cerealien and phosphorien gave the highest effect on the improvement of the growth parameter. The used of biofertilizers as mixture gave superiority in inducing the highest degree of adaptation to the applied levels of salinity was cerealien + phosphorien treatment, which resulted in the highly significant increases in all studied growth parameters; dry matter accumulation and yield components. In addition, the accumulation of considerable quantities of sugars, proline and required nutrients in the stressed wheat plant was also studied. It is clear that inoculation improves all the tolerance feature of wheat plants and increase plant adaptation to saline irrigation.

REFERENCES

1. A.O.A.C., 1975. Official methods of analysis of the Association of Official Analysis Chemists, 12th. ed., published by A.O.A.C., Washington. C.200044.
2. Abd Alla, M.H., A.L.E. Mahmoud and A.A. Issa, 1994. Cyanobacterial biofertilizer improved growth of wheat. Phyton Horn., 34(1): 11-18.
3. Alvarez, M.I., R.J. Sueldo and C.A. Barssi, 1996. Effect of *Azospirillum* on coleptile growth in wheat seedlings under water stress. Cer. Res. Commun., 24: 101-107.
4. Ashour, N.I., K.H. Batanouny, A.T. Thalooth, K.M. Zaid and M.M. Tawfik, 2004. Response of *Medicago sativa* and some halophytic forage plants to irrigation with diluted seawater, 1: Effect on some physiological aspects. Bull. NRC. 29: 361-377.
5. Ashraf, M. and M.R. Foolad, 2006. Roles of glycinebetaine and praline in improving plant abiotic stress resistance. Environmental and Experimental Botany. Available on line at www.Sciencedirect.com.

6. Balba, A.M., 1984. Salinity and crop production. Reuse of Drainage water.proceedings of the Reuse Workshop, Held in Cairo, Egypt.
7. Barraclough, P.B. and A.M. Kyte, 2001. Effect of water stress on chlorophyll meter readings in winter wheat. Plant Nutr., 23: 722-723.
8. Bates, L.S., R.P. Waldern and L.D. Teare, 1973. Rapid determination of free proline for water-stress studies. Plant and soil, 39: 205-207.
9. Creus, C.M., R.J. Sueldo and C.A. Barassi, 1997. Shoot growth and water status in *Azospirillum* inoculated wheat seedlings grown under osmotic and salt stresses. Plant Physiology and Biochemistry Paris, 35(12): 939-944.
10. Das, A.K., M.K. Bera and M. Mohiuddin, 2001. Effect of different yield attributes on the productivity of wheat as influenced by growth regulators and biofertilizers. Environ. Ecol., 19(10: 145-148.
11. El-kased, F.A., R.N. Kamh and B.F. Abd El Ghany, 1999. Wheat response to bio and mineral nitrogen fertigated in newly reclaimed sandy soil. Desert Institute Bulletin, Egypt. 1996, publ., 46(2): 373-386.
12. Essa, T.A., 2002. Effect of salinity stress on growth and nutrient composition of three soybean (*Glycine max* L. Merril) cultivars. J. Agronomy and Crop Sci., 188: 86-93.
13. Favilli, F., S. Taruntoli and W. Balloni, 1988. Azolla biomass as a biofertiliszer for wheat, sunflowers and maize. Annali di Microbiologia ed Enzimologica. 38(1): 109-121.
14. Franklin, W.T., J.S. Olsen and P.N. Soltanpour, 1991. Effects of excessive magnesium in irrigation water on wheat and corn growth. Communications in Soil Science and Plant Analysis, 22(1-2): 49-61.
15. Gomez, K.A. and A.A. Gomez, 1984. Statistical procedures for Agricultural Research. 2nd Ed. International Rice Res. Inst. J. Wiley and Sons, New York, U.Y.
16. Hank, R.J., L.M. Dundley, R.L. Cartee, W.R. Mace, E. Pomela, R.L. Kidman and J.M. Wraith, 1989. Use of saline waste water from electric power plants for irrigation. Report part 1. Soils irrigation water and crop yield studies. Research Report Utah Agricultural Experiment Statia, 128: 1-60.
17. Hartmann, A., 1988. Osmoregulatory properties of *Azospirillum* Spp. In *Azospirillum* IV: Genetics, Physiology, Ecology, Kling muller W., ed., Springer-Verlag, Berlin, 122-130.
18. Hartmann, A., A. Singh and W. Klingmuller, 1983. Isolation and characterization of *Azospirillum* mutants excreting high amount of indole acetic acid. Can. J. Microbiol., 29: 916-923.
19. Ingram, J. and D. Bartels, 1996. The molecular basis of dehydration tolerance in plants. Ann. Rev. Plant Physiol. Plant Mol. Biol., 47: 377-403.
20. Kaci, Y., A. Heyraud, B. Mohamed and T. Heulin, 2005. Isolation and identification of an EPS-producing Rhizobium strain from arid soil (Algeria): characterization of its EPS and the effect of inoculation on wheat rhizosphere soil structure.Research in Microbiology, 156: 522-531. Khadr, A.A., N. Nowalgy and M.M. Hussein, 1980. Differential tolerance of wheat, barley, broad bean and pea seedlings to various levels of salinity, Bull. N. R. C. Egypt., 5: 283-289.
21. Khadr, A.A., N. Nowalgy and M.M. Hussein, 1980. Differential tolerance of wheat, barley, broad bean and pea seedlings to various levels of salinity, Bull. N. R. C. Egypt., 5: 283-289.
22. Khater, E.A., T.S.A. Aal and F.R. Farrag, 2002. Potential effects of low quality water for irrigation in El Fayoum Governorate, Egypt. Egyptian J. Soil Sci., 42(1): 161-183.
23. Kishore, P.B.K., S. Sangam, R.N. Amrutha, P.S. Laxmi, K.B. Naidu, K.R.S.S. Bao, S. Rao, K.J. Reddy, P. Theriappan and N. Sreenivasula, 2005. Regulation of prolinebiosynthesis, degradation, uptake and transport in higher plants: its implications in plant growth and abiotic stress tolerance. Curr. Sci., 88: 424-438.
24. Kumar, V., D. Kumar and C.P.S. Chauhan, 1987. Effect of saline water varying Cl: SO_4 ratios and phosphorus levels on the growth and yield of bread-wheat and soil properties. Indian Journal of Agricultural Research, 17(2): 209-212.
25. Kusaka, M., M. Ohta and T. Fujimura, 2005. Contribution of inorganic components to osmotic adjustment and leaf folding for drought tolerance in *Pearl millet.* Physiol. Plant., 125: 474-489.
26. Linder, R.C., 1944. Rapid analytical methods for some of the more common inorganic conistituents of plant tissues. Plant Physiol., 19(1): 76-79.
27. Mahajan, S. and N. Tuteja,, 2005. Cold, salinity and drought stresses. Archieves of Biochemistry and Biophysics, 444: 139-158.
28. Mashhoor, W.A., M.A. El Borollosy, H.H. Abdel Azeem, S.A. Nasr and S.M. Selim, 2002. Biofertilization of wheat plants exposed to environmental stress conditions. Arab Univ. J. Agric. Sci., 10(2): 543-565.
29. Mass, E.V. and C.M. Crieve, 1990. Spike and leaf development in salt stressed wheat. Crop Sci., 30: 1309-1313.
30. Mohiuddin, M., A.K. Das and D.C. Ghosh, 2000. Growth and productivity of wheat as influenced by integrated use of chemical fertilizer, biofertilizer and growth regulator. Indian J. Plant Physiology, 5(4): 334-338.

31. Munns, R. and H.M. Rawson, 1999. Effect of salinity on salt accumulation and reproductive development in the apical meristem of wheat and barley. Australian Journal of Plant Physiology, 26(5): 459-464.
32. Murphy, L.R., S.T. Kinsey and M.J. Durako, 2003. Physiological effects of short term salinity changes on *Ruppia maritima*. Aquatic Bot., 75: 293-309.
33. Nour El Din, N.A., A.M. Hegazi, M.A. Abd El Gawad and M.O.M. Salem, 1984. Effect of gibberellic acid application on some growth characters of wheat under wadi sunder conditions. Egypt. J. Agron., 9(1-2): 17-28.
34. Pregl, F., 1945. Quantitative-organic Micro-analysis. 4th ed. J. and A., Churchill. Led. London.
35. Ramazan, C., E. Mustafa, E. Ummugulsum and F.D. Mesude, 2007. The influence of plant growth promoting rhizobacteria on growth and enzyme activites in wheat and spinach plants. J. Plant Nutrition and Soil Science, 170: 288-295.
36. Rodriguez, A.A., A.M. Stella, M.M. Storni, G. Zulpa and M.C. Zaccaro, 2006. Effects of cyanobacterial extracellular products and gibberellic acid on salinity tolerance in *Oryza sativa* L. Available on line at http://www.salinesystems.org/content/2/1/7.
37. Saber, Y.M., 1993. Salt tolerance of wheat cultivars. J. Genetics and Breeding, 49(1): 55-60.
38. Salem, S.M., O.M. El Shihy, A.M. Ghallab and H.M. Youssef Ibrahim, 2002. Studies on the correlation between resistance against *Orobanche* and tolerance to salinity in some *Vicia faba* cultivars. J. Agric. Mansoura Univ., 27(7): 4643-4669.
39. Sharma, A.K., A.K. Kelkar, O.R. Misra, S.S. Kushwaha and A.M. Rajput, 2000. Response of wheat (*Triticum durum*) to integrated nutrient management under irrigated conditions. Research on Crops, 1(1): 105-107.
40. Sheteawi, S.A. and K.M. Tawfik, 2007. Interaction effect of some biofertilizers and irrigation water regime on mung bean (*Vigna radiate*) growth and yield. J. App. Sci. Res., 3(3): 251-262.
41. Sultana, N., T. Ikeda and R. Itoh, 1999. Effect of NaCl salinity on photosynthesis and dry matter accumulation in developing rice grain. Environ. Exp. Bot., 42: 211-220.
42. Tawfik, K.M., 2008. Effect of water stress in addition to potassiomag application on mungbean. Australian J. Basic and App. Sci., 2(1): 42-52.
43. Tawfik, K.P., Amany A. Bahr and A.K.M. Salem, 2006. Response of kaller grass (*Leptochloa fuscal* L.) to biofertilizer inoculation under different levels of sea water irrigation. J. App. Sci. Res., 2(12): 1203-1211.
44. Thomas, Roberston, M.J., S. Fukai and M.B. Peoples, 2004. The effect of timing and serveity of water deficition growth, development, yield accumulation and nitrogen fixationof mungbean. Field Crop Research, 86: 67-50.
45. Volkmar, K.M., Hu Y. and H. Stppuhn, 1998. Physiological responses of plants to salinity: A review. Can. J. Plant Sci., 78: 19-27.
46. Wu, S.C., Z.H. Cao, Z.G. Li, K.C. Chcung and M.H. Wong, 2005. Effects of biofertilizer containing N- fixer, P and K solubilizers trial. Geoderma, 125: 155-166.
47. Zeng Linghe, M.C. Shannon and L.H. Zeng, 2000. Salinity effects on seedling growth and yield components of rice. Crop Science, 40(4): 996-1003.