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PHYSIOLOGICAL RESPONSE OF PEA PLANTS TO SALT STRESS TREATMENTS IN RELATION TO BIOFERTILIZER (HALEX 2) OR MINERAL FERTILIZER (ZINC)

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ABSTRACT: Two pot experiments were conducted during the two winter seasons of 2002/2003 and 2003/2004 , to investigate the effect of salt stress treatments, the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) at different levels and their interaction on plant growth characters, leaf water relations, chemical components, yield and its attributes as well as seed quality of pea plants. The obtained results indicated that plant growth characters were differently affected by salt stress conditions. Under low salinity treatment (60 mM NaCl) there was a significant reduction in some growth characters, i.e., plant height, number of leaves, fresh and dry weights of stems and leaves as well as leaf area. On the other hand, this low saline concentration significantly increased leaflet length, leaflet width, as well as root fresh and dry weights. Application of higher salt concentration (120 mM NaCl) led to more significant decrease in all studied growth characters. The inoculation with biofertilizer (Halex 2) or the addition of supplemental mineral fertilizer (zinc) at different levels significantly increased all studied growth characters. Better results were recorded by inoculation with Halex 2. Under salt stress conditions, the inoculation with Halex 2 or the use of different zinc concentrations not only mitigated the inhibitory effect of saline stress treatments on plant growth characters but also increased these characters significantly compared with there owing controls. Increasing salinity level in root media decreased relative water content % and degree of sclerophylly% but increased leaf water deficit %. Under NaCl salinity stress, the inoculation with Halex 2 or the addition of zinc

overcome the inhibitory effect of salinity hazard on leaf water relations and improved water status of pea leaves. A highly significant reduction in the concentration of photosynthetic pigments (chl. a, chl. b, chl. a+b and carotenoids), total soluble sugars, total carbohydrates and total protein was observed in response to salinity stress treatments. The highest decrement in these chemical parameters was more pronounced at the highest NaCl level (120 mM). The inoculation with Halex 2 or the application of zinc significantly increased these chemical components. The interaction between saline stress treatments and the application of Halex 2 or zinc limited the inhibitory effect of saline stress and significantly improved photosynthetic pigments, total soluble sugars, total carbohydrates and total protein concentrations. Proline concentration was significantly increased in salinized pea leaves compared with the control. The inoculation with Halex 2 or the usage of zinc markedly depressed proline accumulation in pea leaves, particularly under salt stress conditions. With rising salinity level in root media, concentrations of N, P, K^+, Ca^{2+} and Zn in pea leaves as well as K^+/Na^+ and Ca^{2+}/Na^+ ratio were progressively decreased comparing to the control and the highest decrease was obvious at 120 mM NaCl. Under salt stress conditions, the inoculation with biofertilizer or the application of mineral fertilizer alleviated the inhibitory effect of salinity treatments and increased N, P, K^+, Ca^{2+} and Zn as well as K^+/Na^+ and Ca^{2+}/Na^+ ratio compared with untreated plants. On the other hand, the accumulation of Na^+ in pea leaves was significantly increased as the salinity increased. The application of Halex 2 or zinc especially under salinity stress treatments led to a significant decrement in Na^+ accumulation in pea leaves. Pea yield and its components represented by number of pods/plant, weight of pods/plant, pod length, pod width, number of seeds/pod, weight of seeds/pod and weight of 100 seeds (seed index) as well as seed quality expressed as total carbohydrates and total protein were significantly decreased in response to salt stress treatments. The inoculation with Halex 2 or the addition of zinc increased pea yield and seed quality.

Key words: Salinity, pea plants, biofertilizer, zinc, stress conditions.

INTRODUCTION

Pea (*Pisum sativum* L.) is a very popular vegetable crop and considered one of the most important legume crops in Egypt for local consumption and exportation. This crop is widely used as a source of protein in human diets due to its high content of protein, ascorbic acid, carbohydrates, balanced amino acids composition and good digestibility. In general, this crop gives high yield and ensures high profits, especially when cultivated for green pods. Therefore, it occupies a prominent position among other legumes in the Egyptian agriculture.

Salinity has a considerable effect on world agriculture, results in reducing productivity of agricultural plants. Soil salinity is an increasing threat for agriculture and is a major factor in decreasing plant production. Due to over irrigation, salinity, which hampers metabolic activities in plant cells, is increasing in many areas around the world (Sanan-Mishra *et al.*, 2005).

Salt affected soils represent a stress condition for crop plants that is of increasing importance in agriculture. Of the world's cultivated lands, about 23% are saline. This situation tends to aggravate year by year, particularly in the arid and semiarid regions, where it is necessary to use low quality water for irrigation (Rengel, 1992). The reduction in plant growth by salinity depends on the solute concentration in the soil solution and on the plant species (Greenway and Munns, 1980).

Growth suppression may be a non-specific effect of salts, depending more on the total concentration of soluble salts than on specific ions (Adams, 1991). Although salinity may reduce growth, it can induce water stress as it increases the osmotic pressure in the soil solution. Moreover, high salinity may also result in too high internal ion concentration (ion toxic) and thus causes growth reduction (Greenway and Munn, 1980).

Water stress induced by salinity may influence plant growth by adverse effects on dry matter partitioning, cell extension, cell division, leaf photosynthesis and/or transpiration (Greenway and Munns, 1980).

Plant growth and production on saline areas, is often limited by a lack of nitrogen. The danger of increasing the soil salinity and the pollution of agro-ecosystem as well as deterioration of soil fertility is likely to further limit the application of N to saline soil

(Mohammad *et al.*, 1989). Therefore the importance of biological fixation to nitrogen has increased.

In legumes salt stress significantly limits productivity because of the adverse effects on growth of plants, root nodule bacteria and the nitrogen fixation capacity (Bekki *et al.*, 1987). In *Vicia faba*, salinity of the culture medium decreased total plant N content as well as the nitrogen fixation by affecting both the appearance of new root nodules and the efficiency of association (Cordovilla *et al.*, 1994).

It has been suggested that *Azospirillum* inoculation could enhance plant growth and yield by promoting mineral and water uptake in colonized roots (Kapulnik *et al.*, 1985). Many *Azospirillum* species produce several plant hormones such as indole acetic acid, isobutyric acid and cytokinins in liquid culture (Omay *et al.*, 1993), and it is firmly stated that hormonal effects are the main mechanism by which *Azospirillum* enhances plant growth. Furthermore, phytohormones such as IAA, gibberellic acid and kinetin known to be involved in the regulation of plant response to salinity stress, these hormones were found to be reversed the adverse effect of stress conditions (Walker and Dumbroff, 1981).

Micro-nutrients play an important role in plant metabolism. The role of zinc as a micro-element has been reported by many researchers (Etman, 1992).

Zinc is known to be required for a variety of metabolic processes in plants such as photosynthetic reactions, nucleic acids metabolism, proteins and carbohydrates biosynthesis and starch metabolism (Marschner, 1986). Zinc is necessary for the synthesis of tryptophan and hence indirectly for the synthesis of auxin. The activities of a number of respiratory enzymes, the accumulation of quinines and catechol aggregates, respiratory impairment, changes in the levels of proteins and amino acids have been reported to follow restrictions in the zinc supply (Klein *et al.*, 1962). It is well known, that zinc positively affects cell division and expansion. Moreover, zinc was found to ameliorate plant growth under saline soils (Shukla and Mukhi, 1985).

The positive effects of zinc on the growth and yield of legume plants have been observed by many investigators such as Singh *et al.* (1992) and Etman (1992).

The aim of this study is to evaluate the effect of salt stress treatments, the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction on growth characters, leaf water relations, chemical composition, yield and its characteristics as well as seed quality of pea plants, in an attempt to alleviate the inhibitory effect of salt stress on studied parameters.

MATERIALS AND METHODS

Two pot experiments were performed at the Experimental Farm, Faculty of Agriculture, Minufiya University, Shibin El-Kom, during the two successive winter seasons of 2002/2003 and 2003/2004 in order to study the responses of vegetative growth characters, leaf water relations, chemical parameters as well as yield and seed quality of pea (*Pisum sativum* L.) plants to salinity stress treatments and to investigate the effect of biofertilizer (Halex 2) or mineral fertilizer (zinc) at different levels in alleviating salinity stress.

The soil of these experiments was clay loam, the texture and certain properties of the experimental soil were analyzed according to Page *et al.* (1982) and their data are presented in Table 1.

Table 1.Physical and chemical properties of the experimental soil in the Experimental Farm, Faculty of Agriculture, Minufiya University, Shibin El- Kom, Egypt

Particle Size	Properties
Sand %	35%
Silt %	18.3%
Clay %	46.7%
Soil texture	Clay
EC (dsm^{-1})	1.03
pH	7.76
Soluble cations (meq/l)	
Ca^{2+}	3.25
Mg^{2+}	3.15
Na^{+}	4.34
K^{+}	1.51
Soluble anions (meq/l)	
HCO^{-}_{3}	2.48
Cl^{-}	5.60

Seeds of pea cultivar Little Marvel were obtained from the Agricultural Research Center, Cairo, Egypt. Five pea seeds were sown on October 31st and 28th in the first and the second seasons, respectively, in plastic pots, 30 cm inner diameter and 30 cm depth filled with 7 kg air dried soil and fertilized with 1.6 g P_2O_5 in form of calcium superphosphate (15.5% P_2O_5). Soil moisture was kept at 65% of the total water holding capacity of the soil during the growth period by irrigation with tap water whenever. Three weeks after sowing, the seedlings were thinned to three uniformed plants per pot. Pots were fertilized with N at 1.36 g/pot and K at o.8 g/pot in form of ammonium nitrate (33.5% N) and potassium sulphate (48% K_2O), respectively, during the experimental period. Other agricultural practices were done as commonly recommended in growing pea.

Four weeks after planting pots were irrigated with saline water at 60 mM NaCl, 120 mM NaCl and tap water as a control.

Halex 2, a biofertilizer containing a mixture of growth promoting N-fixing bacteria of genera *Azospirillum*, *Azotobacter* and *Klebsiella*, which was kindly supplied by Prof. Dr. M. G. Hassouna, Biofertilizer Unit, Plant Pathology Dept., Alex. Univ., was used in this study and was added in irrigation water at a rate of 1 g/pot after 6 weeks from sowing.

Zinc sulphate ($ZnSO_4$; H_2O = 36% Zn) was used as a source of zinc at rates of 20 mg/kg soil (Zn 1) and 40 mg/kg soil (Zn 2) and was added in irrigated water after 6 weeks from sowing.

The experimental design was split-plot in randomized complete blocks, with five replications. Each experiment included 12 treatments, which were all possible combinations of three salinity stress conditions 0 (control), 60 and 120 mM NaCl as well as four fertilizer treatments Control, Biofertilizer (Bio), Zn at 20 and 40 mg/kg soil. The salinity treatments were considered as the main plots and the fertilizer treatments were considered as the sub-plots and were randomly assigned within each main plot.

RECORDED DATA

Vegetative growth characters

After 60 days from sowing, a sample of four plants from each plot was taken randomly. In each plant sample, plant height (cm), number of leaves/plant, leaflet length (cm), leaflet width (cm), fresh and dry weights of roots, stems and leaves, (dried at 70°C for 72 hours) g/plant and leaf area cm^2/plant using the dry weight method described by Aase (1978) were recorded.

Leaf water relations

Relative water content (RWC%), leaf water deficit (LWD%) and the degree of sclerophyll (DScl%) were estimated according to Kalapos (1994).

Chemical analysis

The same samples were used for determination of the following chemical characters:

a- Photosynthetic pigments were estimated in fresh leaves as described by Wettestein

(1957), then calculated as mg/g dry weight.

b- Total soluble sugars and total carbohydrates of dry leaves were determined colorimetrically using the phenol sulfuric acid method of Dubois *et al.* (1956).

c- Free proline in fresh leaf samples was estimated using the method described by Bates *et al.* (1973).

d- Total nitrogen concentration in dry leaves was determined using micro-kjeldahl method according to Ling (1963).

e- Phosphorous in dry leaves was determined colorimetrically following the hydroquinone method as described by Snell and Snell (1954).

f- Potassium and calcium were estimated in dried leaves using Flame Photometer according to Allen (1974).

g- Sodium and zinc were measured in dry leaves using the atomic absorption spectrophotometer according to Cottenie *et al.* (1982).

h- Total protein concentration was calculated by multiplication total nitrogen by 6.25.

Total Yield and its Characteristics

Green pods yield

Marketable green pods were picked, 80 days after planting and the following data were recorded:

- Number of pods/plant.
- Length of pod (cm).
- Width of pod (cm).

Seed yield

Pea plants were left until full plant maturity (drying stage) then the following data were determined:

-Number of seeds per pod.

-Weight of seeds per pod (g).

-Weight of 100seeds (seed index, g).

Seed quality

Protein and total carbohydrates concentrations in dry seeds were estimated, as above mentioned, for dry leaves.

The collected data of both seasons were statistically analyzed using Costat Software program (1985). Treatment means were compared based on the revised L. S. D. test at 0.05, level (Snedecor and Cochran, 1981).

RESULTS AND DISCUSSION

Growth Characters

Data recorded in Table 2 show that treating pea plants with different soil salinity levels (60 and 120 mM NaCl) significantly decreased plant height and number of leaves compared with untreated plants. At 120 mM NaCl the reduction reached to 25.83% and 28.86% for plant height and 26.14% and 35.23% for number of

Table 2. Vegetative growth characters of pea plants as affected by salinity stress treatments and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction, in 2002/2003 and 2003/2004 seasons after 60 days from sowing.

Treatments		Plant height (cm)		Number of leaves / plant		Leaf length (cm)		Leaf width (cm)		Root fresh weight (g)		Stem fresh weight (g)	
NaCl Level mM	Fertilizer rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		26.21^{A}	27.86^{A}	11.17^{A}	11.58^{A}	2.91^{B}	2.03^{B}	2.08^{B}	2.03^{B}	0.45^{B}	0.43^{B}	1.14^{A}	1.44^{A}
60		23.13^{B}	23.82^{B}	9.83^{B}	9.33^{B}	3.34^{A}	2.43^{A}	2.50^{A}	2.43^{A}	0.54^{A}	0.57^{A}	0.98^{B}	1.02^{B}
120		19.44^{C}	19.82^{C}	8.25^{C}	7.50^{C}	2.44^{C}	1.67^{C}	1.87^{C}	1.67^{C}	0.36^{C}	0.32^{C}	0.84^{C}	0.74^{C}
	Control	17.87^{D}	18.59^{C}	8.11^{D}	8.11^{C}	2.40^{C}	1.53^{C}	1.87^{C}	1.53^{C}	0.34^{D}	0.33^{C}	0.72^{C}	0.86^{C}
	Bio	28.09^{A}	29.80^{A}	11.66^{A}	11.44^{A}	3.49^{A}	2.46^{A}	2.47^{A}	2.46^{A}	0.57^{A}	0.57^{A}	1.41^{A}	1.31^{A}
	Zn 1	24.58^{B}	25.18^{B}	10.22^{B}	9.78^{B}	3.01^{B}	2.17^{AB}	2.19^{B}	2.17^{AB}	0.49^{B}	0.46^{AB}	1.02^{B}	1.11^{AB}
	Zn 2	21.18^{C}	21.76^{BC}	9.0^{C}	8.55^{C}	2.69^{BC}	2.01^{B}	2.08^{C}	2.01^{B}	0.41^{C}	0.41^{BC}	0.79^{C}	0.97^{B}
0	Control	20.50^{cd}	22.70^{bcde}	9.33^{cd}	10.67^{b}	2.40^{def}	2.30^{fg}	1.93^{de}	1.43^{ef}	0.35^{def}	0.33^{de}	0.80^{de}	1.24^{abcd}
	Bio	32.10^{a}	36.73^{a}	13.33^{a}	13.33^{a}	3.60^{ab}	3.37^{ab}	2.30^{bcd}	2.47^{ab}	0.45^{bc}	0.58^{abc}	1.73^{a}	1.73^{a}
	Zn 1	27.70^{ab}	28.27^{bc}	11.33^{b}	11.33^{b}	3.03^{bcd}	2.93^{cd}	2.10^{cde}	2.23^{abc}	0.47^{cd}	0.41^{bcde}	1.17^{bc}	1.46^{ab}
	Zn 2	24.53^{bc}	23.73^{bcd}	10.67^{bc}	11.0^{b}	2.60^{cde}	2.47^{ef}	2.0^{cde}	2.0^{bcde}	0.44^{cde}	0.39^{cde}	0.85^{de}	1.32^{abc}
60	Control	17.60^{d}	18.30^{de}	8.33^{d}	8.0^{c}	3.0^{bcd}	2.97^{cd}	2.30^{bcd}	1.97^{bcde}	0.39^{def}	0.41^{bcde}	0.73 e	0.72^{de}
	Bio	28.50^{ab}	29.20^{ab}	11.33^{b}	11.33^{b}	3.73^{a}	3.70^{a}	2.77^{a}	2.80^{a}	0.68^{a}	0.74^{a}	1.31 b	1.32^{abc}
	Zn 1	24.97^{bc}	25.60^{bcd}	10.33^{bc}	9.67^{bc}	3.43^{ab}	3.20^{bc}	2.50^{ab}	2.50^{ab}	0.62^{ab}	0.61^{ab}	1.05 cd	1.07^{bcde}
	Zn 2	21.47^{cd}	22.17^{bcde}	9.33^{cd}	8.33^{c}	3.20^{abc}	2.80^{de}	2.40^{abc}	2.43^{ab}	0.48^{cd}	0.53^{abcd}	0.83 de	0.95^{bcde}
120	Control	15.50^{d}	14.77^{e}	6.67^{e}	5.67^{d}	1.80^{f}	1.93^{g}	1.37^{f}	1.20^{f}	0.27^{f}	0.25^{e}	0.63 e	0.61^{e}
	Bio	23.67^{bc}	23.47^{bcde}	10.33^{bc}	9.67^{bc}	3.13^{abc}	2.73^{de}	2.30^{bcd}	2.10^{bcd}	0.48^{cd}	0.38^{cde}	1.20 bc	0.88^{cde}
	Zn 1	21.07^{cd}	21.67^{bcde}	9.0^{cd}	8.33^{c}	2.57^{cde}	2.33^{f}	1.97^{cde}	1.77^{cde}	0.38^{def}	0.36^{de}	0.84 de	0.82^{cde}
	Zn 2	17.53^{d}	19.37^{cde}	7.0^{e}	6.33^{d}	2.27^{ef}	2.17^{fg}	1.83^{e}	1.60^{def}	0.32^{ef}	0.30^{e}	0.70 e	0.64^{e}

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

leaves, in the first and second seasons, respectively, compared with the control plants. These results are in agreement with those obtained by Cachorro *et al.* (1994) who reported that plant shoot growth of *Phaseolus vulgaris* was inhibited after exposure to an external NaCl. In this concern, Mansour *et al.* (1996) found that shoot length and number of leaves of the salinized faba bean plants were significantly reduced in comparison to their controls. Additionally, Yadav and Yadav (1998) indicated that plant height of pea plants decreased with increasing salinity.

Inoculation with Halex 2 showed the highest significant increase in plant height and number of leaves than supplemental zinc levels Table 2. The highest increment was 57.19% and 60.30% for plant height and 43.77% and 41.06% for number of leaves of Halex 2 tested plants, in the first and second seasons, respectively, compared with untreated plants.

Under saline stress conditions the inoculation of pea plants with Halex 2 or the application of zinc resulted in an increase in plant height and number of leaves compared with the control plants Table 2.

Results listed in Tables 2 and 3 demonstrate that under the low salinity level (60 mM NaCl) there was a significant increase in leaflet length, leaflet width, root fresh and dry weights in both seasons compared to the nonsalinized plants. Meanwhile, increasing soil salinity level up to 120 mM NaCl had adversal effect on these characters and caused significant decrease in these parameters comparing with untreated plants.

The inoculation of pea plants with biofertilizer or the usage of zinc at different levels markedly increased the above mentioned characteristics compared with the control plants. The maximum increment in these mean values was observed by using Halex 2 followed by the lower zinc concentration (Zn1).

The data in Tables 2 and 3 reveal that under salts stress conditions the usage of Halex 2 or zinc at different concentrations alleviated the inhibitory effect of salinity and increased the leaflet width, leaflet length as well as root fresh and dry weights. The second season showed the same trend. Similar results were obtained by Bhatti and Sarwar (1977) who found that application of zinc increased the dry weight of corn roots at all salt treatments.

Table 3. Vegetative growth characters of pea plants as affected by salinity stress treatments and biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction, in 2002/2003 and 2003/2004 seasons after 60 days from sowing.

Treatments		Leaves fresh weight (g)		Root dry weight (g)		Stem dry weight (g)		Leaves dry weight (g)		Leaf area (cm^2)	
NaCI Level mM	Fertilizer Rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		3.51 A	3.68 A	0.053 B	0.041 B	0.27 A	0.31 A	0.84 A	0.93 A	255.94 A	270.80 A
60		2.80 B	2.82 B	0.102 A	0.066 A	0.20 B	0.19 B	0.58 B	0.56 B	168.70 B	157.15 B
120		2.16 C	2.08 C	0.035 C	0.027 C	0.13 C	0.11 C	0.35 C	0.39 C	94.45 C	97.48 C
	Control	1.71 C	2.15 C	0.050 B	0.034 C	0.13 C	0.13 C	0.38 C	0.38 C	128.66 D	124.29 D
	Bio	3.99 A	3.67 A	0.080 A	0.060 A	0.30 A	0.27 A	0.86 A	0.90 A	216.06 A	217.67 A
	Zn 1	2.94 B	2.99 B	0.065 B	0.046 B	0.20 B	0.21 B	0.62 B	0.66 B	196.65 B	197.82 B
	Zn 2	2.66 B	2.63 BC	0.056 B	0.037 BC	0.17 BC	0.19 B	0.50 C	0.56 B	150.75 C	160.79 C
0	Control	2.27 cde	2.60 cdef	0.043 e	0.033 cde	0.17 bcd	0.21 cd	0.49 def	0.59 cde	192.79 d	217.16 d
	Bio	4.87 a	4.92 a	0.063 cde	0.055 bc	0.45 a	0.39 a	1.29 a	1.37 a	313.17 a	319.01 a
	Zn 1	3.49 bc	3.81 b	0.057 cde	0.041 cde	0.25 b	0.34 ab	0.90 b	0.96 b	311.18 a	312.96 b
	Zn 2	3.42 bcd	3.39 bcd	0.049 de	0.036 cde	0.22 bc	0.31 ab	0.67 bcd	0.80 bcd	206.61 c	234.05 c
60	Control	1.61 e	2.32 bcde	0.077 bcd	0.047 cd	0.13 cd	0.11 def	0.36 ef	0.32 ef	93.35 h	81.85 j
	Bio	3.96 ab	3.43 bc	0.142 a	0.092 a	0.26 b	0.28 bc	0.84 bc	0.83 bc	228.20 b	215.23 d
	Zn 1	3.09 bcd	2.91 bcde	0.102 b	0.07 b	0.22 bc	0.19 cde	0.59 cde	0.57 cdef	187.0 d	171.15 e
	Zn 2	2.52 cde	2.61 cdef	0.086 bc	0.053 bc	0.20 bcd	0.17 def	0.51 def	0.52 cdef	166.26 e	160.36 f
120	Control	1.24 e	1.54 f	0.031 e	0.21 e	0.09 d	0.077 f	0.28 f	0.22 f	99.84 g	73.83 k
	Bio	3.15 bcd	2.67 cdef	0.042 e	0.034 cde	0.20 bcd	0.14 def	0.44 def	0.51 cdef	106.82 f	118.76 g
	Zn 1	2.23 cde	2.24 def	0.036 e	0.027 de	0.14 cd	0.11 def	0.37 ef	0.46 def	91.78 h	109.35 h
	Zn 2	2.03 de	1.88 ef	0.032 e	0.023 e	0.10 d	0.10 ef	0.32 ef	0.37 ef	79.37 i	87.95 i

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

Data recorded in Tables 2 and 3 point out that fresh and dry weight of pea stems and leaves as well as leaf area were significantly reduced under NaCl stress. Similar results were observed by Mansour (1991) working on soybean, Ismail (1996) working on *Phaseolus vulgaris*, Garg *et al.* (1997) and Sibole *et al.* (1998) working on bean who reported that salinity induced a reduction in plant height, plant fresh and dry weight and leaf growth. Furthermore, Marcelis and Van Hooijdonk (1999) indicated that under salinity stress conditions the reduction in dry weight growth of the radish plants could largely be attributed to the reduced leaf area. The decrease in shoot dry matter under high salinity could be attributed to interference in the absorption of plant nutrients and physiological water stress created by high salt concentration due to increased osmotic pressure of soil solution (Shukla and Mukhi,1985).

As seen in Tables 2 and 3 results show clearly significant increases in the above mentioned parameters compared with untreated plants. Data also reveal that the inoculation with Halex 2 caused highly significant increase in the fresh and dry weights of pea stems and leaves as well as leaf area followed by lower zinc concentration (Zn1) and higher zinc level (Zn2). In this connection, Singh *et al.* (1992) mentioned that zinc application resulted in more vigorous vegetative growth than in control pea plants.

Concerning the interaction between salinity and the treatments with Halex 2 or zinc at different concentrations, data in Tables 2 and 3 show that under saline conditions the usage of Halex 2 or zinc supply significantly stimulated fresh and dry weights of stems and leaves as well as leaf area of pea plants compared with untreated plants. These results are in agreement with those obtained by Hamdia and El-Komy (1998) who showed that under saline condition *Azospirillum* inoculation increased fresh and dry weights of maize plants. Furthermore, El Sherif *et al.* (1990 a) and Alpaslan *et al.* (1999) reported that under saline conditions the addition of zinc increased the fresh and dry weight of tomato plants. This increase in shoot dry matter with zinc under salt affected soil conditions was perhaps, related to increased concentration of Ca^{2+}, K^{+} and Zn and decreased concentration of Na^{+}.

It could be concluded that saline stress treatments negatively affected the vegetative growth characters of pea plants and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) ameliorate this inhibitory effect.

The reduction in most growth characters may be related to direction toxicity due to the accumulation of high ion concentration in plant tissue (Sibole *et al.*, 1998), or to the osmotic stress due to the lowering of the external water potential (Greenway and Munns, 1980). The inhibitory effect of NaCl salinity on the growth of seedling has been attributed to decreasing water uptake thus reducing the activity of certain enzymes by decreasing the rate of transcription and/or translation (Dodd and Donovan, 1999). Moreover, decreasing growth parameters, hormonal imbalance due to increasing ABA and reducing cytokinins, GA_3 and IAA (Roy *et al.*, 1995), or toxic effects caused by sodium and chloride ions (Dodd and Donovan, 1999) may decrease growth of pea plants. Application of biofertilizer under salinity conditions may result in an increase in root depth, which increase the nutrients uptake as shown in Tables 1 and 2 and previously reported by Turner and Backman (1991) and promotion growth substances (mainly IAA, GA_3 and cytokinin) produced by the microorganisms (Omay *et al.*, 1993) may enhance the growth of tested pea plants. The positive effect of zinc on plant growth characters in relations to salinity stress was observed also by El Sherif *et al.* (1990 a) who reported that zinc application enhanced plant growth characters under saline soil conditions.

Leaf Water Relations

Relative water content % (rwc %) and leaf water deficit % (lwd %)

Data illustrated in Fig. 1 show that with increasing salinity level in root media RWC% was sharply decreased, meanwhile, LWD% was increased compared with unsalinized plants. The maximum reduction in RWC% and the highest increment in LWD% was obvious by the usage of 120 mM NaCl. In this connection, El-Kady *et al.* (1983) reported that the inhibitory effects of salinity on plant behavior might be due to unavailability of water to the plant as a result of increasing osmotic pressure and toxicity of salt. The increase in leaf osmotic pressure as salinity levels increased could be attributed to, either the accumulation of organic substances or to more absorption of mineral salts. The obtained results are in agreement with those obtained by Ruilian and Gang (1997) and Fedina and Tsonev (1997) working on pea who mentioned that relative water content % was significantly reduced under salt stress treatments. Furthermore, Hernandez *et al.* (1999) reported that leaf water relations in pea plants decreased progressively with increasing NaCl stress.

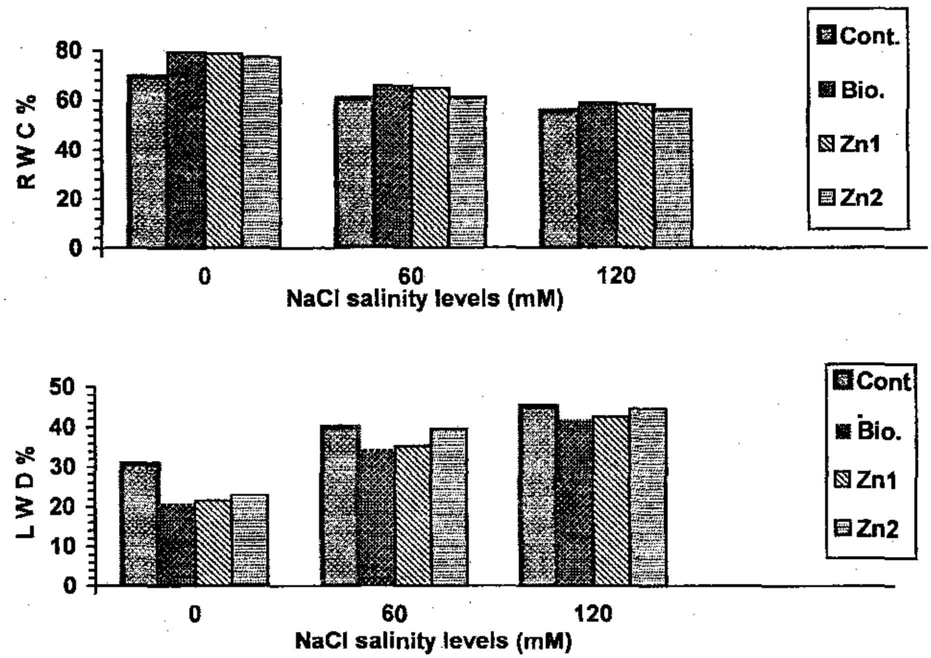

Fig. 1. RWC% and LWD% of pea plants as affected by NaCl treatments and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction in 2002/2003 season.

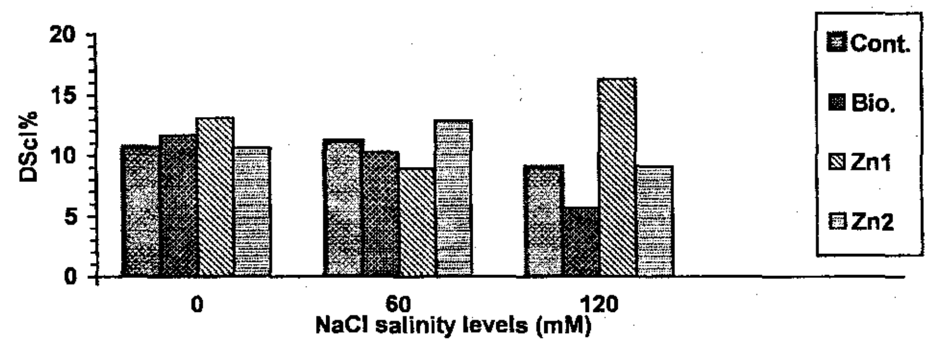

Fig. 2. DScl% of pea plants as affected by NaCl treatments and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction in 2002/2003 season

Results in Fig. 1 reveal that the addition of biofertilizer (Halex 2) or mineral fertilizer (zinc) increased RWC% and decreased LWD% compared with the control. The inoculation with Halex 2 led to the highest increase in RWC% and the maximum reduction in LWD% followed by Zn1 and Zn2, respectively. In this concern, El-Ghinbihi and Abd El-Fattah (2001) showed that inoculation of taro plants with Halex 2 significantly increased relative water content%.

The obtained data Fig. 1 indicate that under saline stress conditions the application of biofertilizer (Halex 2) or mineral fertilizer (Zn1 and Zn2) overcome the inhibitory effect of salinity hazard and improved water status in pea leaves as compared with untreated plants. Similar results were obtained by Noel *et al.* (1996) who found that the application of biofertilizer under saline condition alleviated the depressive effect of salinity by improving leaf water potential and water uptake. The second season showed the same trend.

Degree of sclerophylly % (DScl %)

Results illustrated in Fig. 2 demonstrate that there was a reduction in DScl% of pea leaves as a results of increasing salinity level compared with unsalinized plants. The highest reduction in DScl % was observed under high salinity rate (120 mM NaCl).

According to the effect of biofertilizer (Halex 2) or mineral fertilizer (Zn1 and Zn2) on DScl%, data in Fig 2 indicate that the inoculation with Halex 2 generally decreased this paramcters compared with uninoculated plants, meanwhile, the usage of zinc increased DScl % mean value compared with untreated plants. The highest increment was recorded by Zn1.

Data in Fig. 2 show that under salinity stress treatments the application of mineral fertilizer (zinc) alleviated the inhibitory effect of salinity stress conditions on DScl% and increase its mean value. Similar results were obtained in the second season.

Chemical Composition

Photosynthetic pigments

Results presented in Table 4 indicate that under saline conditions the concentration of chl. a, chl. b, chl. a+b and carotenoids was significantly decreased compared with unsalinized plants. The application of 120 mM NaCl severely affected photosynthetic pigments and caused the highest significant reduction in the photosynthetic pigments. This decrease was 34.14% and 33.16% for chl. a, 72% and 75.41% for chl. b, 46.24% and 46.75% for chl. a+b as well as 60.83% and 61.72% for

Table 4. Photosynthetic pigments, total soluble sugars, total carbohydrates and protein concentrations of pea leaves as affected by salinity stress treatments and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction (as mg/g d. wt.), in 2002/2003 and 2003/2004 seasons after 60 days from sowing..

Treatments		Chlorophyll a		Chlorophyll b		Chlorophyll a+b		Carotenoids		Total soluble sugars		Total carbohydrates		Protein concentration	
NaCl Level mM	Fertilizer Rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		4.13 ^{A}	3.86 ^{A}	2.0 ^{A}	1.83 ^{A}	6.12 ^{A}	5.69 ^{A}	2.17 ^{A}	2.09 ^{A}	38.11 ^{A}	40.71 ^{A}	145.93 ^{A}	150.0 ^{A}	183.16 ^{A}	175.74 ^{A}
60		3.25 ^{B}	3.10 ^{B}	1.68 ^{B}	1.57 ^{B}	4.93 ^{B}	4.67 ^{B}	1.66 ^{B}	1.60 ^{B}	25.33 ^{B}	26.89 ^{B}	121.51 ^{B}	126.10 ^{B}	148.35 ^{B}	141.78 ^{B}
120		2.72 ^{C}	2.58 ^{C}	0.72 ^{C}	0.55 ^{C}	3.44 ^{C}	3.13 ^{C}	0.85 ^{C}	0.80 ^{C}	20.0 ^{C}	22.05 ^{C}	100.68 ^{C}	106.27 ^{C}	126.42 ^{C}	118.61 ^{C}
	Control	2.06 ^{D}	1.95 ^{D}	0.91 ^{D}	0.83 ^{D}	2.97 ^{D}	2.78 ^{D}	0.86 ^{D}	0.83 ^{D}	15.06 ^{D}	16.67 ^{D}	79.58 ^{D}	84.48 ^{D}	128.83 ^{D}	122.23 ^{D}
	Bio	2.98 ^{C}	2.85 ^{C}	1.29 ^{C}	1.09 ^{C}	4.27 ^{C}	3.94 ^{C}	1.0 ^{C}	0.94 ^{C}	22.33 ^{C}	24.04 ^{C}	106.61 ^{C}	112.20 ^{C}	187.56 ^{A}	181.58 ^{A}
	Zn 1	3.91 ^{B}	3.67 ^{B}	1.65 ^{B}	1.51 ^{B}	5.56 ^{B}	5.18 ^{B}	1.84 ^{B}	1.76 ^{B}	30.92 ^{B}	33.45 ^{B}	143.83 ^{B}	147.80 ^{B}	156.54 ^{B}	148.08 ^{B}
	Zn 2	4.51 ^{A}	4.25 ^{A}	2.01 ^{A}	1.84 ^{A}	6.52 ^{A}	6.09 ^{A}	2.53 ^{A}	2.45 ^{A}	42.93 ^{A}	45.37 ^{A}	160.81 ^{A}	165.33 ^{A}	137.63 ^{C}	129.61 ^{C}
0	Control	2.67 ^{fg}	2.56 ^{gh}	1.27 ^{f}	1.14 ^{e}	3.94 ^{fg}	3.70 ^{h}	1.22 ^{g}	1.18 ^{f}	19.59 ^{i}	21.32 ^{h}	95.63 ^{i}	99.84 ^{i}	154.88 ^{f}	149.63 ^{d}
	Bio	3.43 ^{def}	3.21 ^{def}	1.67 ^{d}	1.43 ^{d}	5.10 ^{de}	4.64 ^{e}	1.46 ^{e}	1.39 ^{e}	27.33 ^{e}	29.77 ^{d}	132.77 ^{f}	138.20 ^{f}	225.94 ^{a}	221.44 ^{a}
	Zn 1	4.77 ^{b}	4.37 ^{b}	2.28 ^{b}	2.17 ^{b}	7.05 ^{b}	6.54 ^{b}	2.27 ^{c}	2.19 ^{c}	43.55 ^{b}	46.55 ^{b}	167.50 ^{b}	171.42 ^{b}	185.69 ^{b}	174.88 ^{b}
	Zn 2	5.63 ^{a}	5.28 ^{a}	2.76 ^{a}	2.59 ^{a}	8.39 ^{a}	7.87 ^{a}	3.73 ^{a}	3.59 ^{a}	61.95 ^{a}	65.21 ^{a}	187.81 ^{a}	190.52 ^{a}	166.13 ^{d}	157.0 ^{c}
60	Control	2.17 ^{g}	2.10 ^{h}	1.02 ^{g}	0.97 ^{f}	3.19 ^{g}	3.07 k	3.07 ^{j}	1.0 ^{g}	14.27 k	15.75 ^{j}	78.13 k	83.74 k	130.31 ^{i}	122.63 ^{g}
	Bio	2.70 ^{fg}	2.57 ^{gh}	1.51 ^{e}	1.39 ^{d}	4.21 ^{efg}	3.96 ^{g}	1.07 ^{i}	1.03 ^{g}	20.74 ^{h}	22.23 ^{g}	101.95 ^{h}	107.20 ^{h}	179.56 ^{c}	174.0 ^{b}
	Zn 1	3.90 ^{cd}	3.72 ^{cd}	1.91 ^{c}	1.78 ^{c}	5.81 ^{cd}	5.50 ^{d}	2.09 ^{d}	1.98 ^{d}	26.95 ^{f}	28.44 ^{e}	146.56 ^{d}	150.35 ^{d}	150.63 ^{g}	143.31 ^{e}
	Zn 2	4.21 ^{bc}	4.02 ^{bc}	2.29 ^{b}	2.13 ^{b}	6.50 ^{bc}	6.15 ^{c}	2.45 ^{b}	2.39 ^{b}	39.37 ^{c}	41.12 ^{c}	159.38 ^{c}	163.11 ^{c}	132.88 ^{h}	127.19 ^{f}
120	Control	1.33 ^{h}	1.19 ^{i}	0.45 ^{i}	0.38 ^{i}	1.78 ^{h}	1.57 ^{l}	0.34 ^{l}	0.30 ^{i}	11.31 ^{l}	12.94 k	64.97 ^{l}	69.87 ^{l}	101.31 ^{k}	94.44 ^{i}
	Bio	2.82 ^{fg}	2.76 ^{fg}	0.69 ^{j}	0.45 ^{i}	3.51 ^{g}	3.21 ^{j}	0.45 k	0.41 ^{h}	18.93 ^{j}	20.11 ^{i}	85.10 ^{j}	91.21 ^{j}	157.19 ^{e}	149.31 ^{d}
	Zn 1	3.05 ^{ef}	2.92 ^{efg}	0.76 ^{h}	0.57 ^{h}	3.81 ^{fg}	3.49 ^{i}	1.17 ^{h}	1.12 ^{f}	22.27 ^{g}	25.37 ^{f}	117.43 ^{g}	121.63 ^{g}	133.31 ^{h}	126.06 ^{f}
	Zn 2	3.68 ^{cde}	3.44 ^{de}	0.98 ^{g}	0.81 ^{g}	4.66 ^{ef}	4.25 ^{f}	1.40 ^{f}	1.36 ^{e}	27.48 ^{d}	29.77 ^{d}	135.25 ^{e}	142.36 ^{e}	113.88 ^{j}	104.63 ^{h}

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

carotenoids in the first and second seasons, respectively, compared with unsalinized plants. Furthermore, the obtained data reveal that under the high salinity level (120 mM NaCl) the reduction was greater in chl. b followed by carotenoids. This reduction in photosynthetic pigments which observed in pea plants may be attributed to the depressive effect of salinity on the absorption of ions such as iron which involved in chloroplast biosynthesis (Maria, 1992) and /or due to increased ABA concentration resulting in promoting chlorophyll breakdown (Hall and McWha, 1981). In this connection, Ismail (1996) on *Phaseolus vulgaris*, Garg *et al.* (1997) and Sibole *et al.* (1998) on bean and Hernandez *et al.* (1999) on pea reported that NaCl salinity decreased chlorophyll concentration.

As shown in Table 4 data demonstrate that the higher level of zinc (Zn2) caused the highest significant increase in chl. a, chl. b, total chl. (chl. a+b) and carotenoids followed with Halex 2. This increase was 118.93% and 117.95% for chl. a, 154.43% and 327.91% for chl. b, 129.58% and 155.88% for total chlorophyll as well as 194.19% and 195.18% for carotenoids in the first and second seasons, respectively, compared with untreated plants.

Data in the same Table reveal that under salinity stress treatments, the inoculation of pea plants with Halex 2 or the application of zinc nutrition at different levels (Zn1 and Zn2) significantly enhanced photosynthetic pigments compared with the control plants. In this concern, the usage of zinc at the higher level (Zn2) led to the maximum mean values of chl. a, chl. b, chl. a+b and carotenoids compared with control plants. The obtained results are in accordance with those recorded by Hamida and El-Koumy (1998) who mentioned that the inhibitory effect of NaCl on chlorophyll and carotenoid in maize leaves was completely eliminated by *Azospirillum* treatment.

Total soluble sugars and total carbohydrates concentrations

Results in Table 4 point out that increasing salinity levels up to 120 mM NaCl significantly decreased total soluble sugars and total carbohydrates. The highest decreases (47.52% and 45.84% for total soluble sugars as well as 31.0% and 29.15% for total carbohydrates) were observed under the higher level of salinity (120 mM NaCl). Similar results were observed by Gendy and Hammad (1993) who showed that reducing and total sugars in soybean plants were decreased by increasing salinity levels. Furthermore, Ismail (1996) found that increasing NaCl salinity decreased carbohydrate metabolism in *phaseolus vulgaris* plants. The reduction in total carbohydrates and total soluble sugars may be attributed to the adverse effect of salinity on carbohydrate metabolism through photosynthetic activity (Hammad, 1992).

Data presented in Table 4 indicate that the inoculation of pea plants with Halex 2 or the usage of zinc at different concentrations had a pronounced effect on the concentration of total soluble sugars and total carbohydrates in pea leaves as compared to the control. Treatment with the higher zinc level (Zn2) led to the highest increase in this respect followed by the treatment with the low zinc concentration (Zn1) and the inoculation with Halex 2 compared with untreated plants, in both seasons.

Presented data Table 4 show that under salinity stress conditions treatments with Halex 2 or zinc significantly enhanced the concentration of total soluble sugars and total carbohydrates compared with untreated plants. Furthermore, the higher zinc level (Zn2) gave highly significant increase in these parameters compared with the other treatments and the control, in both seasons. In this concern, Hamida and El-Koumy (1998) found that treatment with *Azospirillum* induced a marked increase in soluble saccharide concentration in shoots of maize plants under salinity levels.

Total protein concentration

Results listed in Table 4 reveal that total protein concentration in pea leaves was significantly depressed after salinity stress treatments. Increasing NaCl concentration significantly reduced total protein concentration. The highest reduction (30.98% and 32.51% in the first and second season, respectively, compared with the control plants) was detached by using 120 mM NaCl. These results are in harmony with those recorded by Fedina and Tsonev (1997) working on pea and Garg *et al*. (1997) working on bean who observed that salt stress decreased protein concentration.

Data presented in the same Table state that the inoculation of pea plants with Halex 2 caused the maximum significant increment in protein concentration. This increment was 45.59% and 48.56% in the first and second seasons, respectively, compared with uninoculated plants, followed by

Zn1 and Zn2. Thus biofertilizer might play an important role in the protein biosynthesis either by direct nitrogen supply (through fixation of nitrogen) or indirectly by enhancing the uptake of soil nitrogen and or by enhancing the photosynthetic apparatus (Hamdia and El-Komy, 1998).

The interaction between saline stress treatments and the inoculation with Halex 2 or the application of different levels of zinc nutrition had a pronounced effect on the concentration of total protein in pea leaves. Under salinized conditions, the inoculation of pea plants with biofertilizer gave the highest significant mean value of total protein concentration followed by Zn1 and Zn2. The second season showed patterns similar to the first one. In this concern, Hamida and El-Koumy (1998) indicated that *Azospirillum* increased protein concentration in maize plants irrespective of the salinity level.

Proline concentration

Data presented in Table 5 show that increasing salinity stress treatments significantly increased the accumulation of proline in pea leaves. The highest increase was observed by using 120 mM NaCl and reached to 51.07% and 50.94% in the first and second seasons, respectively, compared to untreated plants. Similar results were reported by Fedina and Tsonev (1997) on pea and Garg *et al.* (1997) on bean who mentioned that salinity stress increased proline concentration. The accumulation of proline in pea leaves under salinity stress conditions may be related to the decrease in leaf osmotic potential and/or the stimulation of its biosynthesis, inhibition of its oxidation and inhibition of protein synthesis (Stewart and Hanson, 1980). Proline is a substance inducing osmotic adjustment, it is a source of energy, carbon and nitrogen for the recovering tissues and protects several enzymes against the inactivating effect of heat (Stewart and Lee, 1974).

Results in the same Table indicate that the use of biofertilizer (Halex 2) or mineral fertilizer (zinc) significantly decreased proline accumulation in pea leaves. In this concern, Halex 2 led to the highest reduction in proline concentration followed by the lower zinc concentration (Zn1).

Data in Table 5 demonstrate that under saline stress conditions the inoculation with Halex 2 or the application of zinc significantly decreased proline accumulation in pea leaves compares with the control plants, in both seasons. In this connection, Del Zoppo *et al.*

Table 5. Proline (ug/g d. wt.) and mineral concentrations (mg/g d. wt.) of pea leaves as affected by salt stress treatments and the application of biofertilizer (Halex 2)or mineral fertilizer (zinc) and their interaction, in 2002/2003 and 2003/2004 seasons after 60 days from sowing.

Treatments		Proline		N		P		K^+		Ca^{2+}	
NaCl Level mM	Fertilizer Rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		372.28 C	362.95 C	29.31 A	28.12 A	3.32 A	3.84 A	30.84 A	31.64 A	25.66 A	24.41 A
60		494.06 B	485.59 B	23.74 B	22.69 B	2.82 B	3.50 B	24.05 B	25.35 B	21.54 B	20.58 B
120		562.39 A	547.83 A	20.23 C	18.98 C	2.31 C	2.87 C	20.91 C	22.14 C	17.97 C	16.83 C
	Control	559.23 A	549.15 A	20.61 D	19.56 D	2.16 D	2.59 D	22.43 D	23.61 D	17.50 D	16.47 D
	Bio	403.07 D	395.35 D	30.01 A	29.05 A	2.47 C	3.04 C	25.02 C	26.13 C	20.43 C	19.35 C
	Zn 1	449.26 C	435.49 C	25.05 B	23.69 B	2.89 B	3.53 B	25.43 B	26.46 B	23.34 B	22.11 B
	Zn 2	493.41 B	481.84 B	22.02 C	20.74 C	3.73 A	4.45 A	28.20 A	29.31 A	25.62 A	24.49 A
0	Control	410.32 f	402.13 h	24.78 e	23.94 d	2.51 g	2.91 i	27.52 d	28.19 d	21.90 e	20.62 e
	Bio	326 98 i	321.98 k	36.15 a	35.43 a	2.96 e	3.32 f	31.99 b	32.87 b	23.86 d	22.47 d
	Zn 1	368.65 h	356.36 j	29.71 b	27.98 b	3.65 c	4.15 c	29.52 c	30.33 c	26.91 b	25.73 b
	Zn 2	383.18 g	371.33 i	26.58 d	25.12 c	4.15 a	4.98 a	34.33 a	35.17 a	29.96 a	28.81 a
60	Control	572.45 b	563.94 b	20.85 g	19.62 g	2.16 j	2.89 i	21.66 i	23.12 h	17.16 h	16.27 h
	Bio	411.22 f	405.83 g	28.73 c	27.84 b	2.46 h	3.03 g	22.93 h	24.16 g	20.10 f	19.32 f
	Zn 1	469.19 e	457.98 f	24.10 f	22.93 e	2.81 f	3.47 e	25.22 f	26.32 e	23.72 d	22.48 d
	Zn 2	523.36 c	514.62 d	21.26 g	20.35 f	3.85 b	4.62 b	26.40 e	27.81 d	25.16 c	24.23 c
120	Control	694.91 a	681.37 a	16.21 i	15.11 i	1.80 l	1.98 k	18.12 l	19.52 j	13.44 i	12.51 i
	Bio	471.0 e	458.25 f	25.15 e	23.89 d	2.0 k	2.76 j	20.13 k	21.37 i	17.32 h	16.27 h
	Zn 1	509.95 d	492.12 e	21.33 g	20.17 f	2.22 i	2.97 h	21.54 j	22.73 h	19.39 g	18.11 g
	Zn 2	573.69 b	559.56 c	18.22 h	16.74 h	3.2 d	3.75 d	23.86 g	24.94 f	21.74 e	20.42 e

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

(1999) reported that under saline conditions application of biofertilizer, may increase plant resistance to salinity by accumulation of soluble compounds leading to osmotic adjustment.

Mineral concentration

Nitrogen concentration (N)

Results in Table 5 point out that treating pea plants with different salinity levels (60 and 120 mM NaCl) progressively decreased N concentration in pea leaves compared with untreated plants. The usage of 120 mM NaCl led to highly significant reduction in N concentration compared with the other treatment and the control. This reduction reached to 30.80% and 32.50 in the first and second seasons, respectively, compared with unsalinized plants. Similar findings were previously observed by Ismail (1996) who found that N concentration decreased significantly under salt stress treatments.

Data presented in Table 5 indicate that N concentration was increased as a result of the inoculation with Halex 2 or the application of zinc. The inoculation with boifertilizer gave the higher significant increase in N concentration followed by the Zn1. This increase reached to 45.61% and 48.52% in the first and second seasons, respectively, compared with untreated plants.

The interaction between salinity levels and biofertilizer or mineral fertilizer (zinc) treatments stated that these treatments significantly enhanced N concentration in pea leaves and completely eliminated the inhibitory effect of saline stress conditions on N concentration, in both seasons Table 5.

Phosphorous concentration (P)

Data in Table 5 show that with increasing salinity level, there was a significant decrease in P concentration in pea leaves and the maximum reduction was recorded at 120 mM NaCl. This decrease reached to 30.42% and 25.26% in the first and second seasons, respectively, compared with the control plants. These results are in agreement with those obtained by Dahiya and Singh (1976) who reported that the concentration of P in pea crop was greatly decreased with increasing salinity. Moreover, Ismail (1996) mentioned that NaCl salinity decreased P concentration in *Phaseolus vulgars* plants.

Application of biofertilizer (Halex 2) or different mineral fertilizer (zinc) levels caused a significant increase in P mean value in pea leaves. The usage of higher zinc concentration (Zn2) led to the maximum increase in this respect. This increase was 72.69%

and 71.81% in the first and second seasons, respectively, compared with untreated plants.

It can be noticed from Table 5 that under salinized conditions, the application of biofertilizer or supplemental zinc alleviated the inhibitory effect of salinity on P concentration and increased P mean value significantly. The second season showed the same trend.

Potassium concentration (K^+)

Results listed in Table 5 demonstrate that K^+ concentration in pea leaves was significantly decreased with raising salinity level in root media. The maximum reduction was recorded by 120 mM NaCl, which reached to 32.20% and 30.03% as compared with unsalinized plants. These results are in harmony with those observed by Shukla and Mukhi (1985) working on maize who showed that increasing salinity levels decreased shoot K^+ due to antagonistic effect of Ca^{2+}, Mg and Na^+ on K^+. Moreover, Ismail (1996) found that increasing NaCl salinity decreased K^+ concentration in *Phaseolus vulgars* plants. Relatively high concentration of Na^+ and Cl^- in the soil or plant depressed the uptake of other ions such as potassium, calcium, magnesium, nitrate or phosphate (Turhan and Eris, 2004). The reduction in K^+ concentration under salinity stress conditions may be due to the disturbance in the osmotic pressure of soil solution which was responsible for lowering the movement of water and solvents from roots to vegetative portions (Al-Qubaie, 2002), and/or due to the reducing effect of salinity on the activity of xylem tissues which in turn reduces the absorption and translocation of nutrients towards vegetative organs (Miller *et al.*, 1990).

Data in Table 5 indicate that inoculation of pea plants with Halex 2 or the addition of supplemental zinc at different levels significantly increased K^+ accumulation in pea leaves compared with untreated plants, especially under salt stress conditions. In this concern, the maximum increase was observed by the higher zinc level (Zn2) followed by the lower zinc concentration (Zn1). In this concern, Shukla and Mukhi (1985) mentioned that zinc application counteracted the adverse effects of salinity on K^+ absorption and shoot K^+ was invariably more after Zn application.

Calcium Concentration (Ca^{2+})

Analysis of variance show that generally increasing NaCl salinity level up to 120 mM NaCl caused a significant decrease in pea leaf Ca^{2+} concentration comparing with their controls Table 5. Under extremely salinity condition (120 mM NaCl) Ca^{2+} concentration recorded the highest reduction. This decrease reached to 29.97% and 31.05% in the first and second seasons, respectively, comparing with unsalinized plants. Similar results were observed by Shukla and Mukhi (1985) who reported that high salinity levels decreased the concentration of Ca^{2+} in maize shoots. Furthermore, Cachorro *et al.* (1994) found that Ca^{2+} concentration in *Phaseolus vulgaris* salinized seedlings was lower than in nonsalinized seedlings and the transport of Ca^{2+} to the shoots was substantially affected by salinity. NaCl inhibits Ca^{2+} transport from root to shoot by interfering with the release of Ca^{2+} into the root xylem, possible via an effect on the active loading of Ca^{2+} into xylem vessels (Lynch and Lauchli, 1985).

Regarding the effect of Halex 2 inoculation or zinc application on the accumulation of Ca^{2+} in pea leaves, data in Table 5 reveal that the application of these treatments significantly increased Ca^{2+} concentration in pea leaves. In this concern, zinc at high level (Zn2) caused the greatest significant increase in Ca^{2+} concentration (46.4% and 48.69%) followed by low zinc level (Zn1) and Halex 2.

Results shown in Table 5 indicate that increasing the concentration of zinc or using Halex 2 at the root media of pea plants grown under salinity conditions alleviated the negative effect of salinity on Ca^{2+} concentration, furthermore, increased Ca^{2+} concentration in pea leaves significantly compared with untreated plants. In this respect, El Sherif *et al.* (1990 a) mentioned that the interaction between salinity and zinc application resulted in a marked increase of Ca^{2+} concentration.

Sodium concentration (Na^{+})

Results recorded in Table 6 demonstrate that with increasing salinity levels, there was an increase in Na^{+} accumulation in pea leaves. The highest increase in Na^{+} concentration was achieved by 120 mM NaCl and reached to 122.45% as well as 132.71% in the first and second seasons, respectively, compared with untreated plants. Similarly, Gendy and Hammad (1993) observed that increasing salinity levels increased Na^{+} percentage in soybean plants. Moreover, Sibole *et al.* (1998) recorded that salt treated bean plants showed an increase in leaf Na^{+} concentration.

Concerning the effect of the inoculation with biofertilizer or the addition of mineral zinc nutrition on Na^{+} concentration in pea leaves, data in Table 6 indicate that these

Table 6. Mineral concentration, Ka^{+}/Na^{+} and Ca^{2+}/Na^{+} ratio of pea leaves as affected by salt stress treatments and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction, in 2002/2003 and 2003/2004 seasons after 60 days from sowing.

Treatments		Na^{+} concentration (mg/g dry weight)		Zn concentration (ug/g dry weight)		K^{+}/Na^{+} ratio		Ca^{2+}/Na^{+} ratio	
NaCl Level mM	Fertilizer Rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		4.32 C	3.76 C	119.50 A	120.49 A	7.74 A	9.65 A	6.51 A	7.59 A
60		7.27 B	6.14 B	98.02 B	99.14 B	3.52 B	4.35 B	3.19 B	3.58 B
120		9.61 A	8.75 A	78.05 C	79.28 C	2.34 C	2.78 C	2.04 C	2.16 C
	Control	9.32 A	8.43 A	67.06 D	68.03 D	2.75 D	3.18 D	2.16 D	2.25 D
	Bio	7.61 B	6.85 B	71.45 C	72.68 C	4.08 C	4.70 C	3.24 C	3.40 C
	Zn 1	6.24 C	5.31 C	91.97 B	93.21 B	4.79 B	6.13 B	4.39 B	5.16 B
	Zn 2	5.09 D	4.26 D	163.62 A	164.63 A	6.51 A	8.37 A	5.86 A	6.96 A
0	Control	6.21 h	5.87 e	78.55 g	79.32 g	4.43 cd	4.80 e	3.53 e	3.51 f
	Bio	4.26 j	3.92 g	82.53 f	83.74 f	7.51 b	8.39 c	5.60 c	5.73 c
	Zn 1	3.68 k	2.84 h	114.31 d	115.59 d	8.02 b	10.68 b	7.31 b	9.06 b
	Zn 2	3.12 l	2.39 h	202.62 a	203.32 a	11.0 a	14.72 a	9.60 a	12.05 a
60	Control	9.02 c	7.48 c	65.51 j	66.44 j	2.40 g	3.09 i	1.90 h	2.18 i
	Bio	8.18 e	6.89 d	70.59 i	71.82 i	2.80 fg	3.51 h	2.46 g	2.80 g
	Zn 1	6.72 g	5.64 e	85.32 e	86.52 e	3.75 de	4.67 f	3.53 e	3.99 e
	Zn 2	5.16 i	4.53 f	170.67 b	171.77 b	5.12 c	6.14 d	4.88 d	5.35 d
120	Control	12.72 a	11.93 a	57.13 l	58.33 l	1.42 h	1.64 l	1.06 i	1.05 k
	Bio	10.38 b	9.74 b	61.24 k	62.47 k	1.94 gh	2.19 k	1.67 h	1.67 j
	Zn 1	8.33 d	7.45 e	76.28 h	77.53 h	2.59 fg	3.05 j	2.33 g	2.43 h
	Zn 2	6.99 f	5.87 e	117.56 c	118.79 c	3.41 ef	4.25 g	3.11 f	3.48 f

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

treatments decreased Na^{+} accumulation in pea leaves compared with untreated plants. Furthermore, the high zinc level (Zn2) caused the highest significant reduction in Na^{+} concentration (45.39% and 49.47% in the first and second seasons, respectively, comparing with uninoculated plants) followed by the low zinc concentration (Zn1).

Under saline stress conditions the inoculation of pea plants with Halex 2 or the addition of different zinc levels matigated salinity hazard and significantly decreased Na^{+} accumulation in pea leaves compared with their controls. The decrease in Na^+ concentration after zinc application under saline conditions could be explained from the Zn-Na^+ antagonism (Shukla and Mukhi (1985). Moreover, Alpaslan *et al.* (1999) pointed out that Na^+ concentration in tomato plants decreased with increasing the rate of zinc application in saline soils.

Zinc concentration (Zn)

Results presented in Table 6 point out that zinc concentration was significantly decreased with raising salinity level in root media. The highest significant reduction in Zn concentration was observed by using high salinity level (120 mM NaCl), in both seasons. Similar results were observed by Barrow and Ellis (1986) who reported that zinc concentration decreased by high salt concentration.

Data in the same Table indicate that the addition of supplemental zinc in higher concentration (Zn2) led to the maximum significant increment in zinc concentration (143.99% and 142%) followed by the lower zinc concentration (Zn1) and Halex 2.

As seen in Table 6 under salt stress conditions the application of Halex 2 or zinc significantly enhanced zinc accumulation in pea leaves and alleviated the inhibitory effect of salinity stress on zinc concentration in pea leaves, in both seasons. In this concern, Shukla and Mukhi (1985) observed that shoot zinc was increased with zinc application under salinity treatments. Additionally, El Sherif *et al.* (1990 b) and Alpaslan *et al.* (1999) found that the addition of different zinc rates increased zinc concentration in tomato plants in the presence of soil salinity.

K^+/Na^+ and Ca^{2+}/Na^+ Ratios

Data recorded in Table 6 demonstrate that with the increase in NaCl salinity levels, the ratio of K^+/Na^+ and Ca^{2+}/Na^+ was gradually decreased. Under salt stress conditions, this ratio was sharply depressed and the highest reduction was recorded by using 120 mM NaCl. This decrease was 69.77% and 71.19% for K^+/Na^+ ratio as well as 68.66% and 71.54% for Ca^{2+}/Na^+ ratio in the first and second seasons, respectively, compared to owing controls. In this concern, Shukla and Mukhi (1985) mentioned that salinity treatments decreased Ca^{2+}/Na^+ and K^+/Na^+ ratio in maize plants. The reduction in K^+/Na^+ ratio under salt stress conditions may be attributed to the decrease in K^+ uptake and the increase in Na^+ uptake by NaCl treatment resulted in a lower K^+/Na^+ ratio (Hoo-Kim Yeong *et al.* 1999).

Results in Table 6 show that treatments with Halex 2 or zinc at different levels increased K^+/Na^+ and Ca^{2+}/Na^+ ratio significantly. Increasing the concentration of zinc (Zn2) caused highly significant increment of K^+/Na^+ and Ca^{2+}/Na^+ ratio. The inoculation of pea plants with Halex 2 showed lower K^+/Na^+ and Ca^{2+}/Na^+ ratio than zinc supplemental. Similar results were observed in the second season. Presented data indicate that zinc treatments increased Ca^{2+}/Na^+ ratio, which may have a significant effect on some essential enzymatic activities (Rengel, 1992). It is well known that a high Ca^{2+}/Na^+ ratio protects, at least in part, the selective permeability of membranes against Na^+ injury, moreover, K^+/Na^+ ratio is considered to be important in the response of plants to salinity (Lauchli and Schubert, 1989).

The application of biofertilizer or mineral fertilizer (zinc) at the root media of pea plants grown under salinity stress significantly increased K^+/Na^+ and Ca^{2+}/Na^+ ratio compared with the control plants Table 6. The increase in Ca^{2+}/Na^+ and K^+/Na^+ ratio with zinc application was due to increased absorption of Ca^{2+}, K^+ and decreased absorption of Na^+. This effect of zinc on Ca^{2+} and K^+ was also perhaps partly responsible for improved plant growth with zinc under salt affected soil conditions (Shukla and Mukhi, 1985).

It could be noticed that increasing NaCl salinity had an inhibitory effect on minerals accumulation in pea leaves. On the other hand, the inoculation with biofertilizer or the addition of mineral fertilizer (zinc) significantly improved mineral status and alleviated the negative effect of salinity stress in pea leaves. In this connection, Caballero-Mellado *et al.* (1992) reported that biofertilizer inoculated plants showed better uptake of mineral ions from the soil or applied fertilizer than uninoculated plants. These bacteria increase nutrient availability through altering the root surface characteristics involved in nutrient uptake (Lin *et al.*,1983). Moreover, biofertilizer increases the ion concentration of plant shoots as well as tissues water content, biofertilizer might increase the efficiency of water utilization under stress conditions. Thus, biofertilizer resulted in pronounced increase in some organic and/or inorganic osmotically active components of the plant cell sap, which in turn results in a concomitant decrease in the osmotic potential as a possible mechanism of adaptation to salinity (Hamida and El-Koumy, 1998). Furthermore, El Sherif *et al.* (1990 b) concluded that the application of zinc might increase mineral concentrations in tomato plants under saline soil conditions.

Yield and its Components

Data recorded in Tables 7 and 8 show clearly that pea yield and its components expressed as number of pods/plant, weight of pods/plant, pod length, pod width, number of seeds/pod, weight of seeds/pod and weight of 100 seeds (seed index) were significantly decreased under salt stress treatments. The highest reduction in pod number/plant (51.02% and 50.16%), weight of pods/plant (44.53% and 33.44%), pod length (10.70% and 23.11%), pod width (24.69% and 22.92%), number of seeds/pod (25.04% and 37.28%), weight of seeds/pod (63.25% and 58.84%) and seed index (25.06% and 26.96%) was obvious under 120 mM NaCl in the first and second seasons, respectively, compared to their controls. In this concern, Dahiya and Singh (1976) showed that the dry matter yield of pea crop decreased significantly with increased in salinity levels. Moreover, Garg *et al.* (1997) reported that salinity stress reduced bean seed yield. Yadav and Yadav (1998) found that number of pods/plant and yield/plant of pea plants decreased with increasing salinity. This reduction in pea yield and its components may be due to the osmotic effect of soil solution and the specific action of ions, which caused decreasing in water and nutrient uptake, furthermore, absorption of toxic NaCl from saline media and their accumulation up to a toxic level, which affect the metabolic activity of plant tissues (El-Kouny, 2002).

With respect to the application of biofertilizer or zinc at different levels, results in Tables 7 and 8 indicate that the usage of these substances exhibited a high increase in pea yield and its components. The highest significant increment was achieved by inoculation with Halex 2 and reached to 137.08% and 138.89% for number of pods/plant, 80.42 and 62.53% for weight of pods/plant, 33.53% and 29.63% for pod length, 50.60% and 44.07% for pod width, 70.27% and 64.27 for number of seeds/pod, 219.63% and 165.29% for seed weight/pod as well as 63.22% and 53.95% for seed index in the first and second seasons, respectively, compared with untreated plants. In addition, the application of low zinc concentration (Zn1) significantly increased pea yield and its characteristics. Similar results were observed by Singh *et al.* (1992) who reported that application of zinc resulted in

Table 7. Yield and its characteristic of pea plants as affected by salt stress treatments and the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction, in 2002/2003 and 2003/2004 seasons.

Treatments		Number of pods per plant		Weight of pods per plant (g)		Pod length (cm)		Pod width (cm)	
NaCl Level mM	Fertilizer Rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		3.92^{A}	3.17^{A}	6.76^{A}	6.46^{A}	8.97^{A}	8.22^{A}	2.43^{A}	2.40^{A}
60		3.08^{B}	2.33^{B}	5.12^{B}	5.48^{B}	8.01^{B}	7.33^{B}	2.11^{B}	2.17^{B}
120		1.92^{C}	1.58^{C}	3.75^{C}	4.30^{C}	6.86^{C}	6.32^{C}	1.83^{C}	1.85^{C}
	Control	1.78^{D}	1.44^{C}	3.78^{C}	4.35^{C}	6.80^{C}	6.48^{C}	1.68^{D}	1.77^{D}
	Bio	4.22^{A}	3.44^{A}	6.82^{A}	7.07^{A}	9.08^{A}	8.40^{A}	2.53^{A}	2.55^{A}
	Zn 1	3.33^{B}	2.67^{B}	5.39^{B}	5.63^{B}	8.29^{AB}	7.49^{B}	2.26^{B}	2.25^{B}
	Zn 2	2.56^{C}	1.89^{C}	4.84^{B}	4.61^{C}	7.61^{BC}	6.79^{C}	1.98^{C}	1.99^{C}
0	Control	2.33^{def}	3.0^{def}	5.15^{cd}	5.20^{cdef}	7.97^{abcd}	7.77^{bc}	2.05^{bc}	1.97^{de}
	Bio	5.33^{a}	4.67^{a}	8.05^{a}	8.36^{a}	9.88^{a}	9.23^{a}	2.85^{a}	2.80^{a}
	Zn 1	4.33^{ab}	3.67^{b}	7.25^{a}	6.45^{bc}	9.23^{ab}	8.40^{ab}	2.64^{a}	2.60^{ab}
	Zn 2	3.67^{bc}	2.33^{cde}	6.63^{abc}	5.81^{bcde}	8.80^{abc}	7.47^{bcde}	2.18^{b}	2.23^{cd}
60	Control	2.0^{efg}	1.33^{ef}	3.80^{def}	4.55^{defg}	7.0^{cde}	6.33^{efg}	1.70^{d}	1.86^{e}
	Bio	4.33^{ab}	3.33^{bc}	6.97^{ab}	6.97^{ab}	9.23^{ab}	8.43^{ab}	2.60^{a}	2.43^{bc}
	Zn 1	3.33^{bcd}	2.67^{bcd}	5.10^{cd}	6.13^{bcd}	8.30^{abcd}	7.65^{bcd}	2.15^{b}	2.23^{cd}
	Zn 2	2.67^{cde}	2.0^{def}	4.60^{de}	4.28^{efg}	7.50^{bcd}	6.90^{cdef}	2.0^{bcd}	2.16^{cde}
120	Control	1.0^{g}	1.0^{f}	2.40^{f}	3.31^{g}	5.43^{e}	5.33^{g}	1.30^{e}	1.47^{f}
	Bio	2.0^{cde}	2.33^{cdf}	5.48^{bcd}	5.87^{bcde}	8.13^{abcd}	7.53^{bcde}	2.13^{b}	2.43^{bc}
	Zn 1	2.33^{def}	1.67^{def}	3.81^{def}	4.3^{efg}	7.33^{bcde}	6.42^{defg}	2.0^{bcd}	1.93^{de}
	Zn 2	1.33^{fg}	1.33^{ef}	3.30^{ef}	3.73^{fg}	6.53^{de}	6.0^{fg}	1.77^{cd}	1.57^{f}

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

more pods/plant and higher pod and seed yields in pea plants compared with the controls.

Concerning the effect of salinity stress conditions and application of Halex 2 or zinc at different concentrations, the present results indicate that the usage of Halex 2 or zinc not only mitigated the adverse effect of salinity stress but also resulted in significant improvement in pea yield and its components, in both seasons.

Seed Quality

Data recorded in Table 8 indicate that seed quality expressed as total carbohydrates and total protein was affected by salt stress,

Table 8. Seed yield and its quality of pea plants as affected by salt stress treatments and boifertilizer (Halex 2) or mineral fertilizer (zinc) and their interaction in 2002/2003 and 2003/2004 seasons.

Treatments		Number of seeds/pod		Weight of seeds/ pod (g)		Seed index (weight of 100 seeds) (g)		Total carbohydrates (mg/g dry weight)		Total protein (mg/g dry weight)	
NaCl Level mM	Fertilizer Rate	First season	Second season	First season	Second season	First season	Second season	First season	Second season	First season	Second season
0		6.67 A	6.92 A	3.02 A	2.94 A	13.17 A	10.72 A	308.19 A	297.18 A	245.57 A	236.28 A
60		5.83 B	5.67 B	2.06 B	2.04 B	11.43 B	9.34 B	292.61 B	277.12 B	216.66 B	204.83 B
120		5.0 C	4.34 C	1.11 C	1.21 C	9.87 C	7.83 C	270.44 C	257.36 C	183.83 C	173.02 C
	Control	4.44 C	4.33 C	1.07 C	1.21 C	8.89 D	7.60 C	253.54 D	241.78 D	176.19 D	168.04 D
	Bio	7.56 A	7.11 A	3.42 A	3.21 A	14.51 A	11.70 A	338.92 A	321.96 A	251.67 A	240.50 A
	Zn 1	6.0 B	6.0 B	2.13 B	2.38 B	12.22 B	9.41 B	302.90 B	293.0 B	226.69 B	213.29 B
	Zn 2	5.33 BC	5.11 BC	1.64 BC	1.46 C	10.34 C	8.47 BC	266.29 C	252.14 C	206.86 C	197.0 C
0	Control	5.33 cde	5.67 bcd	1.49 de	1.63 ef	10.56 def	9.13 bcd	277.81 g	263.42 g	207.69 g	199.94 f
	Bio	8.33 a	8.67 a	4.71 a	4.43 a	16.20 a	14.45 a	354.38 a	342.63 a	277.63 a	264.88 a
	Zn 1	6.67 abc	7.0 ab	3.27 abc	3.39 b	13.52 b	10.11 bc	315.25 d	307.92 c	258.31 b	250.81 b
	Zn 2	6.33 bc	6.33 bc	2.60 bcd	2.31 cde	12.41 bcd	9.20 bcd	285.31 f	274.73 e	238.63 d	229.50 d
60	Control	4.33 de	4.33 cde	1.03 de	1.32 fg	9.11 efg	7.65 cde	256.25 i	243.55 i	176.06 k	168.38 i
	Bio	7.67 ab	7.0 ab	3.83 ab	3.08 bc	14.33 ab	11.44 b	337.13 b	312.49 b	255.88 c	244.5 c
	Zn 1	6.0 bcd	6.0 bc	1.92 cde	2.51 cd	12.14 bcd	9.30 bcd	308.13 e	301.11 d	230.19 e	213.31 e
	Zn 2	5.33 cde	5.33 bcd	1.46 de	1.24 fg	10.13 def	8.95 bcd	268.94 h	251.33 h	204.50 h	193.13 g
120	Control	3.67 e	3.0 e	0.68 e	0.67 g	7.0 g	6.02 e	226.56 k	218.37 k	144.81 l	135.81 j
	Bio	6.67 abc	5.67 bcd	1.71 cde	2.12 de	13.0 bc	9.21 bcd	325.25 c	310.75 b	221.50 f	212.13 e
	Zn 1	5.33 cde	5.0 bcde	1.20 de	1.23 fg	11.0 cde	8.81 cd	285.31 f	269.97 f	191.56 i	175.75 h
	Zn 2	4.33 de	3.67 de	0.85 e	0.82 fg	8.49 fg	7.26 de	244.63 j	230.35 j	177.44 j	168.38 i

Values marked with same alphabetical letter(s), within a comparable group of means, do not significantly differ using revised L.S.D. test at 0.05 level.

where the concentrations of these components were significantly decreased under salt stress treatments. The highest reduction in total carbohydrates concentration (12.25% and13.40%) or total protein concentration (25.14% and 26.77%) were obvious at 120 mM NaCl in the first and second seasons, respectively, comparing with unsalinized plants.

Analysis of variance show that application of Halex 2 or zinc at different levels significantly improved pea seed quality Table 8, moreover, the obtained results revealed that the maximum increment in total carbohydrates and total protein concentrations was observed after inoculation with Halex 2 followed by low zinc level (Zn1).

Regarding the interaction between salt stress treatments and the application of biofertilizer or mineral fertilizer, data in Table 8 mention that the use of Halex 2 or zinc at different levels mitigated the inhibitory effects of salinity stress conditions and significantly increased total carbohydrates and total protein concentrations, in both seasons.

It could be concluded that salt stress treatments significantly decreased most plant growth characters, RWC%, DScl%, chemical components, yield and its characteristics as well as seed quality in pea plants compared with untreated plants. On the other hand, low salt concentration (60 mM NaCl) significantly increased leaflet length, leaflet width as well as root fresh and dry weights. Moreover, salinity stress treatments increased LWD% and proline concentration. The application of biofertlizer (Halex 2) or different concentrations of mineral fertilizer (zinc) significantly improved growth parameters, leaf water relations, chemical composition, yield and its attributes as well as seed quality compared with the control in both seasons. The interaction between saline stress conditions and biofertilizer or supplemental zinc treatments revealed that under salt stress conditions the inoculation with biofertilizer or applying zinc mitigated the inhibitory effect of salt stress and induced significant increment in all characters under study. Therefore, it can be recommended that the application of biofertilizer (Halex 2) or mineral fertilizer (zinc) can overcome the inhibitory effects of NaCl stress on growth characters, water relations, chemical components, yield and seed quality of pea plants.

REFERENCES

Aase, I. K. 1978. Relationship between leaf area and dry matter in winter wheat. Agron. J., 70: 663-665.

Adams, P. 1991. Effects of increasing the salinity of the nutrient solution with major nutrients or sodium chloride on the yield quality and composition of tomato grown in rock wool. J. Hortic. Sci., 66: 201-207.

Allen, E. S. 1974. Chemical Analysis of Ecological Materials. Bluekuell Scientific Publication. Gany Mead. Oxford, pp. 563.

Alpaslan, M.; A. Inal; A. Gunes; Y. Cikili and H. Ozcan. 1999. Effect of zinc treatment on the alleviation of sodium and chloride injury in tomato (*Lycopersicum esculentum* L. Mill. cv. Lale) grown under salinity. Turkish J. Botany, 23 (1): 1-6.

Al-Qubaie, A. I. 2002. Effect of saline irrigation water on growth and chemical constituents of Neem (*Azadirachta indica* L.). Annals of Agric. Sci., Moshtohor, 40 (1): 397-412.

Barrow, N. J. and A. S. Ellis. 1986. Testing a mechanistic model. V. The points of zero salt affect for phosphate retention for zinc retention and for acid/alkali titration of a soil. J. Soil Sci., 37: 303.

Bates, L. S.; K. P. Waldren and I. D. Teare. 1973. Rapid determination of free proline for water-stress studies. Plant and Soil, 39: 205-207.

Bekki, A.; J. C. Trinchant and J. Rigaud. 1987. Nitrogen fixation (C_2H_2 reduction) by Medicago nodules and bacteriods under sodium chloride stress. Physiol. Plant., 71: 61-67.

Bhatti, A. S. and G. Sarwar. 1977. Response of corn to micronutrients (Zn and Cu) on a saline soil. 1- Growth and ionic relationships. Plant and Soil, 48, 719-724.

Caballero-Mellado, G.; M. G. Carcano Mantiel and M. A. Mascaarua-Esparza. 1992. Field inoculation of wheat (*Triticum aestivum*) with *Azospirillum brasilense* under temperate climate. Symbiosis 13: 243-253. [C. F. Biol. Plant., 40 (1):109-120].

Cachorro, P.; A. Ortiz and A. Cerda. 1994. Implications of calcium nutrition on the response of *Phaseolus vulgaris* L. to salinity. Plant and Soil, 159 (2): 205-212.

Cordovilla, M. P.; F. Ligero and C. Lauch. 1994. The effect of salinity on nitrogen fixation and assimilation in *Vicia faba*. J. exp. Bot., 45: 1483-1488.

Costat Software. 1985. User's Manual-Version 3, CoHort. Tusson. Arizona, U. S. A.

Cottenie, A.; M. Verloo; L. Kikens; G. Velghe and R. Camerlynck. 1982. Analytical problems and methods in chemical plant and soil analysis. Hand book Ed. A. Cottenie, Gent, Blegium.

Dahiya, S. S. and M. Singh. 1976. Effect of salinity, alkalinity and iron application on the availability of iron, manganese, phosphorus and sodium in pea (*Pisum sativum* L.) crop. Plant and Soil, 44: 697-702.

Del Zoppo, M.; L. Galleschi; A. Onnis and A. Pardossi. 1999. Effect of salinity on water relations, sodium accumulation, chlorophyll content and proteolytic enzymes in a wild wheat. Bio. Plant.,42(1):97-104.

Dodd, G. L. and L. A. Donovan. 1999. Water potential and ionic effects on germination and seedling growth of two cold desert shrubs. Amer. J. Botany, 86 (8): 1146-1153.

Dubois, M.; K. A. Gilles; J. K. Hamilton; P. A. Robers and F. Smith. 1956. Colorimetric method for determination of sugars and related substances. Anals. Chem., 28: 350-356.

El-Ghinbihi, Fatma H. and M. A. Abd El-Fattah. 2001. Mitigation of the adverse effects of water stress imposed at different growth stages of taro (*Colocasia esculenta* L. Schott) plants using several treatments: II- The influence of biofertilizer (Halex 2) or yeast extract application on growth, chemical composition and yield of taro plants grown under water stress conditions. Minufyia J. Agric. Res., 26 (4): 1005-1034.

El-Kady, M. M., M. A. Mansour and A. A. El-Sheweikh. 1983. A comparative study on two Mexican wheat varieties and the local variety Giza 155 grown under different levels of salinity. Minufyia J. Agric. Res., 6: 1-15.

El-Kouny, H. M. 2002. Effect of organic fertilizers (compost) at different rates of application under salinity stress condition on soil and tomato plant. Minufiya J. Agric. Res., 27 (2): 355-368.

El-Sherif, A. F.; S. M. Shata and R. A. Youssef. 1990a. Response of tomato seedling to zinc application under different salinity levels I- Dry matter, Ca^{2+}, Mg, K^+ and Na^+ content. Egypt. J. Hort., 17 (2): 131-142.

El-Sherif, A. F.; S. M. Shata and R.A. Youssef. 1990b. Response of tomato seedling to zinc application under different salinity levels II- Micronutrients content. Egypt. J. Hort., 17 (2): 143-150.

Etman, A. 1992. Growth and productivity of faba bean as affected by foliar spraying with some micronutrients. Alex. J. Agric. Res., 37 (1): 161-177.

Fedina, I. S. and T. D. Tsonev. 1997. Effect of pretreatment with methyl jasmonate on the response of *Pisum sativum* to salt stress. J. Plant Physiol., 151 (6): 735-740.

Garg, B. K.; S. Kathju; S. P. Vyas and A. N. Lahiri. 1997. Sensitivity of clusterbean to salt stress at various growth stages. Indian J. Plant Physiol., 2 (1): 49-53.

Gendy, A. A. and A. A. Hammad. 1993. The response of soybean plants to different levels of salinity. Zagazig J. Agric. Res., 20 (6): 1751-1768.

Greenway,H. and R. Munns. 1980. Mechanism of salt tolerance in nonhalophytes. Ann. Rev. Plant Physiol., 31: 149-190.

Hall, H. K. and J. A. McWha. 1981. Effect of abscisic acid on growth of wheat *Triticum aestivum*. Ann. Bot., 47:427-433.

Hamdia, M. A. and H. M. El-Komy. 1998. Effect of salinity, gibberellic acid and *Azospirillum* inoculation on growth and nitrogen uptake of *zea mays*. Biol. Plant., 40 (1): 109-120.

Hammad, A. A. 1992. Physioanatomical studies on the effect of salinity on soybean plants. M. Sc. Thesis, Agric. Botany. Fac. Agric., Minufiya Univ., Egypt.

Hernandez, J. A.; A. Campillo; A. Jimenez; J. J. Alarcon and F. Sevilla. 1999. Response of antioxidants systems and leaf water relations to NaCl stress in pea plants. New Phytol., 141 (2): 241-251.

Hoo-Kim Yeong; Shim-Lesung; K. Kobayashi; K. Usmi; K. Y. Hoo and I. S. Shim. 1999. Relationship between Na^+ content or K^+/Na^+ ratio in shoots and salt tolerance in several gramineous plants. J. Weed Sci. Technol., 44 (4): 293-299.

Ismail, A. M. 1996. Interaction of NaCl salinization and KNO_3 fertilization on physiology of bean plants. Acta Agronomica Hungarica, 44 (3): 221-227.

Kalapos, T. 1994. Leaf water potential, leaf water deficit relationship for ten species of a semiarid grassland community. Plant and Soil, 160: 105-112.

Kapulnik, Y; R. Gafny and Y. Okon. 1985. Effect of *Azospirillum* spp. inoculation on root development and NO_3^- uptake in wheat (*Triticum* *aestivum* cv. Miriam) in hydroponic system. Can. J. Bot., 63: 627-631.

Klein, R. M.; E. M. Caputo and B. A. Witterholt. 1962. The role of zinc in the growth of plant tissue cultures. American J. Botany, 49 (4): 323-327.

Lauchli, A. and S. Schubert. 1989. The role of calcium in the regulation of membrane and cellular growth processes under salt stress. In Environmental Stress in Plants. J. H. Cherry (Ed.) pp.131-138, Springer Verlag, Berlin.

Lin, W.; Y. Okon and R. W. F. Hardy. 1983. Enhanced mineral uptake by *Zea mays* and *Sorghum bicolor* roots inoculated with *Azospirillum brasilense*. Appl. Environ. Microbiol., 45: 1175-1179.

Ling, E. R. 1963. Determination of total nitrogen by semimicro-kjeldahl method. Dairy Chem., 11: 23-84.

Lynch, J. and A. Lauchli. 1985. Salt stress disturbs the calcium nutrition of barley (*Hordeum vulgare* L.). New Phytol., 99: 345-354.

Mansour, S. H. 1991. Genetical studies on soybean (*Glycine max* L. Merel). Ph. D. Thesis in Genetics, Genetics Department, Fac. of Agric.,Alexandria Univ.

Mansour, S. H.; M. E. Sorial; M. B. Habeeb and M. S. Soliman. 1996. Salt tolerance in faba bean, natural variability and its relation to some physiological traits. Minufiya J. Agric. Res., 21 (6): 1403-1422.

Marcelis, L. F. M. and J. Van Hooijdonk. 1999. Effect of salinity on growth, water use and nutrient use in radish (*Raphanus sativus* L.). Plant and Soil, 215: 57-64.

Maria, A. M. 1992. Effect of growth regulators on salt tolerance in wheat plants. Minufiya J. Agric. Res., 17 (3): 1067-1081.

Marschner, H. 1986. Mineral Nutrition in Higher Plants. Academic Press. Harcout Brace Jovanovish, Publishers, pp. 674.

Miller, R. W.; R. L. Donahue and J. N. Miller. 1990. Soils "An introduction to soil and plant growth" 5th Prentice Hall international Inc. Englewood Cliffs, New Jersey, pp.308-339.

Mohammad, M.; W. F. Campbell and M. D. Rumbaugh. 1989. Variation in salt tolerance of alfalfa. Arid Soil Rehabil., 3: 11-30. [C. F. Biol. Plant., 40 (1): 109-120, 1997/98].

Noel, T.C.;C. Sheng; C.K. Yost; R.P. Pharis and M. E. Hynes. 1996. *Rhizobium leguminosarum* as plant growth-promoting rhizobacterium direct growth promotion of canola and lettuce. Can. J. Microbiol., 42 (3): 279-283.

Omay, S. H.; W. A. Schmidt and P. Martin. 1993. Indole acetic acid production by the rhizosphere bacterium *Azospirillum brasilense* cd. under in vitro conditions. Can. J. Microbiol., 39: 187-192.

Page, A. L.; R. H. Miller and D. R. Keeney. 1982. Methods of Soil Analysis. Part 1, 2 American Society of Agronomy, Madison, W. I.

Rengel, Z. 1992. The role of calcium in salt toxicity. Plant Cell Environment, 15: 625-632.

Roy, D.; N. Basu; R. Marik and S. K. Banerjee. 1995. Changes in levels of endogenous gibberelline and abscisic acid in salt-sensitive germinating rice by NaCl-salinity. Plant Physiol., 8 Biochem., New Delhi, 22 (2): 200-202.

Ruilian, Z. and W. Gang. 1997. Water stress induced changes in protection enzyme activities and effects of proline enhancement on drought resistance in peas. Acta Prataculturae Sinica, 6 (4): 39-43. [C. F. Hort. Abst., 1998, 68 (12): 10545].

Sanan-Mishra, N.; XH. Phan; SK. Sopory and N. Tuteja. 2005. Pea helicase 45 over expression in tobacco confers high salinity tolerance without affecting yield. PNAS, 102 (2): 509-514.

Shukla, U. C. and A. K. Mukhi. 1985. Ameliorative of zinc on maize growth (*zea mays* L.) under salt affected soil conditions. Plant and Soil, 87: 423-432.

Sibole, J. V.; E. Montero; C. Cabot; C. Poschenriedes and J. Barcelo. 1998. Role of sodium in the ABA-mediated long-term growth response of bean to salt stress. Physiol. Plant., 104 (3): 299-305.

Singh, A.; B. B. Singh and C. S. Patel. 1992. Response of vegetable pea (*Pisum sativum*) to zinc, boron and molybdenum in an acid Alfisol of Meghalaya. Indian J. Agronomy, 37 (3): 615-616.

Snedecor, G.W. and W.G. Cochran 1981. Statistical Methods. 7th Ed. Iowa State Univ. Press Amer., Iowa, U. S. A.

Snell, R. and G. Snell. 1954. Colorimetric method of analysis. D. Van Nostrad Comp., New York.

Stewart, C. R. and J. A. Lee. 1974. The role of proline accumulation in halophytes. Planta, 120: 279-289.

Stewart, C. R. and A. D. Hanson. 1980. Proline accumulation as a metabolic response to water stress. In: N. C. Turner and P. J. Kramer (Eds.), Adaptation of Plant to Water and High Temperature Stress. John Wiley, New York, pp. 173-189.

Turhan, E. and A. Eris. 2004. Effect of sodium chloride applications and different growth media on ionic composition in strawberry plant. J. Plant Nutrition, 27 (9): 1653-1665.

Turner, J. T. and P. A. Backman. 1991. Factors relating yield increase after seed treatment with *Bacillus subtillis*. Plant Disease, 75: 347-353.

Walker, A. M. and B.E. Dumbroff. 1981. Effect of salt stress on abscisic and cytokinin levels in tomato. Z. Pflanzenphysiol., 11: 461-470. [C. F. Biol. Plant., 40 (1): 109-120].

Wettestein, D.V.1957. Chlorophyll - Letal und Submikroskopische Formel-wechsel der Plastiden. Exptl. Cell Res., 12: 427-433.

Yadav, S. S. and A. C. Yadav. 1998. Effect of chloride and sulphate salinity and phosphate fertilization on growth and yield of pea (*Pisum sativum*). Crop Research 16 (3): 415-417.

الاستجابة الفسيولوجية لنباتات البسلة لمعاملات الاجهاد الملحى وعلاقتها باستخدام السماد الحيوى (هالكس ٢) أوالسماد المعدنى (الزنك)

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اجريت تجربتى أصص بمزرعة التجارب بكلية الزراعة بشبين الكوم جامعة المنوفية خلال الموسمين الزراعيين ٢٠٠٣/٢٠٠٢ و ٢٠٠٤/٢٠٠٣ وذلك لدراسة التأثيرات الفسيولوجية لمعاملات الاجهاد الملحى وكذلك دراسة تأثير استخدام كل من السماد الحيوى (هالكس ٢) أو السماد المعدنى (الزنك) بتركيزات (صفر أو ٢٠ أو ٤٠ ملليجرام كبريتات زنك/ كيلو جرام تربة) وايضا دراسة تأثير التفاعل بين هذه المعاملات المختلفة على صفات النمو والعلاقات المائية للورقة والتركيب الكيماوى للأوراق والمحصول وكذلك جودة البذور لنباتات البسلة صنف لتل مارفيل.

وقد عرضت نباتات البسلة لمعاملات الأجهاد الملحى (الملوحة) باستخدام كلوريد الصوديوم بتركيزات (صفر أو ٦٠ أو ١٢٠ ملليمول) وأشارت النتائج إلى مايلى:-

أدى تعرض نباتات البسلة للتركيز الملحى المنخفض (٦٠ ملليمول كلوريد صوديوم) إلى حدوث نقصا معنويا في بعض صفات النمو الخضرى المتمثلة في أرتفاع النبات وعدد الأوراق والوزن الغض والجاف للسوق والأوراق وكذلك مساحة الأوراق بينما أدى هذا التركيز الملحى المنخفض إلى حدوث زيادة معنوية في طول الورقة وعرضها وكذلك الوزن الغض والجاف للجذور. أوضحت النتائج أن استخدام التركيز الملحى العالى (١٢٠ ملليمول كلوريد الصوديوم) أدى إلى حدوث أعلى نقص معنوى في جميع صفات النمو الخضرى تحت الدراسة.

أثر التلقيح بالسماد الحيوى (هالكس ٢) أو استخدام السماد المعدنى (الزنك) تأثيرا ايجابيا على صفات النمو الخضرى لنباتات البسلة حيث أدى إلى حدوث زيادة معنوية في صفات النمو الخضرى وكذلك أدى إلى التغلب على الأثر الضار للملوحة. وقد أوضحت الدراسة أن المعاملة بالسماد الحيوى هى أكثر المعاملات كفاءة حيث أعطت أعلى زيادة معنوية في صفات النمو الخضرى.

أوضحت الدراسة أن تعرض نباتات البسلة لمعاملات الاجهاد الملحى أدى إلى حدوث نقص في النسبة المئوية للمحتوى النسبي للماء في الأوراق وأيضا النسبة المئوية لدرجة الاسكلوروفيلى بينما أدى إلى حدوث زيادة في النسبة المئوية لنقص الماء في الأوراق.

أدى التلقيح بالسماد الحيوى (هالكس ٢) إلى حدوث زيادة واضحة في النسبة المئوية للمحتوى النسبي للماء بينما أدى إلى نقص النسبة المئوية لنقص الماء ودرجة الاسكلوروفيلى في الأوراق. وقد أدى استخدام السماد المعدنى (الزنك) إلى زيادة النسبة المئوية للمحتوى النسبي للماء والنسبة المئوية لدرجة الاسكلوروفيلى في أوراق البسلة بينما أدى إلى حدوث نقص في النسبة المئوية لنقص الماء في الأوراق. كما أظهرت النتائج أنه تحت تأثير معاملات الاجهاد الملحى أدى استخدام كل من السماد الحيوى والسماد المعدنى إلى التغلب على التأثير الضار للملوحة على العلاقات المائية لأوراق البسلة.

أظهرت نتائج التحليل الكيماوى لأوراق البسلة حدوث نقص معنوى في تركيز كل من صبغات البناء الضوئى (كلوروفيل أ وكلوروفيل ب وكلوروفيل أ+ب والكاروتينيدات) والسكريات الذائبة الكلية والكربوهيدرات الكلية وايضا البروتينات الكلية وذلك نتيجة لتعرض النباتات لمعاملات الاجهاد الملحى. وقد لوحظ أن أعلى نقص معنوى في هذه المكونات الكيماوية كان واضحا عند استخدام التركيز الملحى العالى (١٢٠ ملليمول كلوريد صوديوم).

أدى استخدام السماد الحيوى أو السماد المعدنى (الزنك) إلى زيادة تركيز كل من صبغات البناء الضوئى والسكريات الذائبة الكلية والكربوهيدرات الكلية وكذلك البروتينات الكلية في أوراق البسلة زيادة معنوية كما أدى إلى التغلب على التأثير الضار لمعاملات الاجهاد الملحى.

أوضحت الدراسة أن معاملات الاجهاد الملحى أدت إلى زيادة معنوية فى تراكم البرولين في أوراق نباتات البسلة مقارنة بالنباتات الغير معاملة.

أشارت النتائج إلى أن التلقيح بالسماد الحيوى (هالكس ٢) أو استخدام السماد المعدنى (الزنك) أدى إلى حدوث نقصا معنويا في تركيز البرولين في أوراق البسلة خاصة تحت ظروف الاجهاد الملحى.

أظهرت معاملات الملوحة نقصا معنويا في تركيزكل من النتروجين والفوسفور والبوتاسيوم والكالسيوم والزنك في أوراق نباتات البسلة وكذلك نسبة البوتاسيوم/الصوديوم وايضا نسبة الكالسيوم/الصوديوم. وقد أدى استخدام التركيز الملحى العالى (١٢٠ ملليمول كلوريد صوديوم) إلى حدوث أعلى نقص معنوى في تركيز هذه العناصر بينما أدت معاملات الاجهاد الملحى إلى زيادة تراكم الصوديوم في أوراق البسلة زيادة معنوية وذلك مقارنة بالنباتات الغير معاملة.

أثر التلقيح بالسماد الحيوى (هالكس ٢) أو استخدام السماد المعدنى (الزنك) تأثيرا ايجابيا على تركيز كل من النتروجين والفوسفور والبوتاسيوم والكالسيوم والزنك وكذلك نسبة البوتاسيوم/الصوديوم ونسبة الكالسيوم/الصوديوم حيث أدى إلى زيادة تراكم هذه العناصر في أوراق البسلة معنويا كما أدى إلى التغلب على التأثيرات الضارة لمعاملات الملوحة. كما أوضحت النتائج أن استخدام السماد الحيوى أو الزنك خصوصا تحت تأثير معاملات الاجهاد الملحى أدى إلى حدوث نقص معنوى في تراكم الصوديوم في أوراق البسلة.

أشارت النتائج إلى حدوث انخفاض معنوى في محصول البسلة المتمثل في عدد القرون/النبات ووزن القرون/النبات وطول القرن وعرض القرن وعدد البذور/القرن ووزن البذور/القرن ووزن ١٠٠ بذرة وكذلك صفات الجودة في البذور الممثلة في تركيز الكربوهيدرات الكلية والبروتين الكلى وذلك نتيجة لتعرضها لمعاملات الاجهاد الملحى وخصوصا التركيز الملحى العالى (١٢٠ ملليمول كلوريد صوديوم).

بينت النتائج أن التلقيح بالسماد الحيوى أو استخدام الزنك أدى إلى حدوث زيادة معنوية في محصول البسلة وكذلك تحسين صفات الجودة للبذور كما أدى ايضا إلى تقليل الآثار الضارة نتيجة للتعرض الملوحة.

ومن نتائج هذه الدراسة يمكن التوصية باستخدام السماد الحيوى (هالكس ٢) أو استخدام السماد المعدنى (الزنك) كمحاولة لتقليل الآثار السلبية لمعاملات الاجهاد الملحى على النمو الخضرى والعلاقات المائية للورقة والتركيب الكيماوى للأوراق والمحصول وجودة البذور لنباتات البسلة النامية في ظروف الاجهاد الملحى (الملوحة).