

## **RESPONSE OF PEA (*Pisum sativum* L.) PLANT GROWN IN SAND CULTURE UNDER SALINITY STRESS TO FOLIAR APPLICATION BY KINETIN AND GIBBERELIC ACID.**

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### **ABSTRACT**

Pot experiment was carried out at Faculty of Agriculture, El Bostan, Alexandria University in order to estimate the response of pea plant (*Pisum sativum* L., variety Master B) grown in sand culture under salinity stress to foliar application by kinetin or gibberillic Acid (GA<sub>3</sub>).

Seeds of pea were sown in plastic pot of 20 cm diameter and 30 cm depth containing 10 kg prewashed sand. The water of irrigation consisted from base nutrient solution of Hoagland containing 0, 25, 50 and 100 mM NaCl. After three weeks from sowing, the plants were foliar sprayed by 0, 25 and 50 mg. kinetin l<sup>-1</sup> or by 0, 50 and 100mg. GA<sub>3</sub>l<sup>-1</sup>. Samples of plants were collected after 38 and 88 days from sowing.

The obtained results showed significant decrease of the growth characters of 38 days old plant with increasing salinity. At 100 mM NaCl treatment, the relative decrease of shoot F.W. was 59.0% and that of root was 71.7%. There were also significant decreases of Chl b, and carotenoids and significant increase of proline contents with increasing salinity.

There were no significant stimulating effects due to foliar application by kinetin on all growth characters, Chl a, Chl b and proline contents. However, foliar application by GA<sub>3</sub> improved growth characters, at each salinity level, of 38 days old plants. However, there were significant reductions in grains F.W. of 88 days old plant with foliar application of GA<sub>3</sub> at each level of salinity treatment.

**Keywords :** Pea plant, Relative growth rate (RGR), Kinetin, Gibberellic acid (GA<sub>3</sub>), Salinity stress.

### **INTRODUCTION**

Salinity is an a abiotic stress adversely affecting plant growth. High salinity can limit plant productivity and leads to dwarfism and inhibit root growth. It can reduce the ability of plant root to take up water and this quickly cause reduction in plant growth rate. In salt-sensitive plants, shoot and to less extent root growth is permanently reduced within hours of salt stress and this effect does not appear to depend on Na<sup>+</sup> concentrations in the growing tissues, but rather is a response to the osmolarity of the external solution (Munns, 2002, and Tester and Davenport, 2003),.

Osmotic adjustment in plant subjected to salt stress can occur by accumulation of high concentrations of either inorganic ions or low molecular organic solutes. The compatible osmolytes generally found in higher plants are low molecular weight sugars, organic acids, amides, amino acids and soluble proteins (Ashraf and Harris,2004).

Pea (*Pisum sativum* L.), a winter season food legume is grown in Egypt and other countries of the Meditteream region and, therefore, is adversely affected by salinity. Fedina and Tsonev (1997) found that salt stress leads to reduction in the growth of shoot and root of pea plant exposed to salinization

with 30 mM NaCl for 3 and 6 days. They also found that the chlorophyll a chlorophyll b and carotenoids contents were declined but free proline content was increased at 30 mM NaCl treatment. Several studies reported that salt-stressed pea plant contains high concentrations of proline (Olmos and Hellin, 1996; Lutts *et al.* 1999; Tester and Davenport, 2003). and Kumar *et al.*, 2003. It has been also reported that proline accumulation in plants leaves has been the consequence of salinity (Ahmadi *et al.*, 2009).

Recently, plant growth regulators have been applied in order to overcome the deleterious effects of salt stress. Kinetin is one of the cytokinins known to improve the growth of plants grown under salinity (Gadallah, 1999). Also, foliar application of gibberellic acid (GA<sub>3</sub>) overcomes the effect of salt stress and improve growth parameters, yield and yield components of salt-stressed plant (Akbari *et al.*, 2008).

The objectives of this study, therefore, were to assess the effect of salinity on the growth characters of pea plant and to estimate the response of this plant to foliar application by kinetin or gibberellic acid.

## **MATERIALS AND METHODS**

### **Experimental Layout :**

Pot experiment was carried out at the greenhouse, Faculty of Agriculture at El-Bostan, Alexandria University to investigate the effects of foliar application of kinetin or gibberellic acid on the growth characters of pea (*Pisum sativum* L., variety Master B) plant grown in sand culture under salt stress.

Modified Hoagland and Arnon nutrient solution was used as the base solution (Hewitt, 1966). The concentrations of macro- and secondary- nutrients in this base solution are : 112.62, 14.52, 31.00, 197.23, 72.35, 23.90 and 32.01 mg<sup>-1</sup> for N-NO<sub>3</sub>, N-NH<sub>4</sub>, P, K, Ca, Mg and S, respectively, and those of micronutrients are : 0.25, 0.25, 0.01, 0.025, 0.30 and 0.025 mg.l<sup>-1</sup> for B, Mn, Cu, Zn, Fe and Mo, respectively. The used water of irrigation consisted of both the base nutrient solution containing 0, 25, 50 and 100 mM NaCl.

Split plot layout, in randomized completely block design, with six replicates was used; three replicates were collected after 38 days from sowing and the other three were collected after 88 days. The main plot treatments were four salt levels (0, 25, 50 and 100 mM NaCl) and the subplot treatments were three kinetin levels (0, 25, 50 and 100 mg.l<sup>-1</sup>) or three gibberellic acid levels (0.50 and 100 mg.l<sup>-1</sup>).

Seeds of pea were surface sterilized by soaking in H<sub>2</sub>O<sub>2</sub> (10%) for 10 min, then washed thoroughly by tap water then by distilled water (Hewitt, 1961). Ten seeds were sown in plastic pot of 20 cm diameter and 30 cm depth containing 10 kg pre-washed sand (Hewitt, 1961). Each pot was irrigated daily by tap water and after 11 days the plants were thinned to four seedlings per pot. Each pot was then irrigated daily by one liter irrigation solution. After three weeks from sowing, the plants were foliar sprayed two times, a week interval, by 100 ml per pot solution of kinetin or gibberellic acid

and the control treatment was foliar sprayed with 100ml distilled water per pot.

**Plant Sampling :**

Samples of plant were collected after 38 and 88 days from planting, washed with tap water then by distilled water. The fresh weight (F.W) of plant organs of the first collection (shoot and root) and those of the second collection (shoot, root, pods and grains) were measured. Proportion of the fresh leaves were preserved for photosynthetic pigments and proline analysis. The plant organs were then oven-dried at 65°C for 48 hrs, and the oven-dried weights were measured, then finely ground using stainless steel mill and kept for analysis.

**Plant Analysis :**

**Photosynthetic pigments :** Half gram fresh leaves was cut to small pieces, extracted with 110ml N,N-Dimethylformamide. The optical density was measured at wave length of 662 nm for chlorophyll a (Chl a), of 644 nm for chlorophyll b (Chl b), and of 440 nm for carotenoids using spectrophotometer (Jenway 6305 UV/Vis spectrophotometer). The following equations were used :  $\text{Chl a} = 9.784E_{662} - 0.99E_{644} = \text{mg l}^{-1}$ ,  $\text{Chl b} = 21.426 E_{644} - 4.65 E_{662} = \text{mg l}^{-1}$ ,  $\text{carotenoids} = 4.695E_{440} - 0.268 (\text{Chl a} + \text{Chl b}) = \text{mg l}^{-1}$ , where E is the optical density at the wave length indicated (Moran and Porath, 1980).

**Proline :** The concentration of proline was measured in fully expanded fresh leaves according to the method described by Bates *et al.* (1973).

**Total sugars :** The concentration of total sugars was determined in the grinding oven- dried grains according to the method described by Dubois *et al* (1956).

**Statistical Analysis**

The data obtained were statistically analyzed for the least significant difference using the SAS statistical analysis software (SAS Inst. 1985).

## RESULTS AND DISCUSSION

Under the growth conditions of this study, pea plants grew smaller and flowering was more prolific under salinity conditions (data not shown). The following paragraphs outline the data obtained under salinity treatments and growth regulators foliar application.

**Effect of Salinity- Kinetin Interaction**

**Pea plant after 38 days from sowing**

**Growth characters :** Table 1 showed significant decrease of shoot F.W. at 100 mM NaCl irrigation treatment and that of root F.W. at 50 mM NaCl irrigation treatment. This indicates that pea plant root is more sensitive to salinity than shoot. However, on the D.W. basis, significant decrease was found for shoot and root at 50 mM NaCl.

The values of the relative decrease of shoot F.W. were 2.4, 21.5 and 59.0%, and those of root F.W. were 3.3, 36.3 and 71.7% at 25, 50 and 100 mM NaCl treatments, respectively. On the D.W. basis, these values were

11.6, 39.1 and 62.1% for shoot, and were 18.0, 44.0 and 66.0% for root, at 25, 50 and 100 mM NaCl treatments, respectively. Thus, the relative reduction in shoot growth was less than of the root. This means that pea plant root is more sensitive to salinity than shoot.

**Table 1: The mean value of growth characters of pea plant after 38 days from sowing as influenced by NaCl and kinetin concentrations interactions.**

Treatments		Shoot (g.plant <sup>-1</sup> )		Root (g.plant <sup>-1</sup> )		Sh/Rt ratio		Shoot height (cm)
NaCl (mM)	Kinetin (mg.l <sup>-1</sup> )	F.W.	D.W.	F.W.	D.W.	F.W.	D.W.	
0	0	6.97a	0.69a	10.69 a	0.50 a	0.65 de	1.39 a	20.7 ab
	25	6.80a	0.66ab	10.69 a	0.48 ab	0.64 de	1.37 a	21.0 a
	50	5.59a	0.58ab	8.75 ab	0.40 ac	0.66 de	1.48 a	20.3 ab
25	0	6.80a	0.61ac	10.43 ab	0.41 ac	0.65 de	1.48 a	19.9 ac
	25	5.90a	0.50bd	9.48 ab	0.39 bc	0.62 e	1.28 a	19.4 ac
	50	6.49a	0.59ad	9.59 ac	0.43 ac	0.68 de	1.36 a	19.9 ac
50	0	5.47a	0.42 d	6.81 cd	0.28 d	0.81 be	1.52 a	17.7 bc
	25	5.61a	0.49 bd	7.54 bd	0.33 cd	0.76 ce	1.53 a	20.5 ab
	50	5.27a	0.44 cd	6.28 d	0.29 d	0.84 bd	1.55 a	17.0 cd
100	0	2.86b	0.24 e	3.03 e	0.17 e	0.93 ac	1.54 a	13.4 e
	25	2.48b	0.22 e	2.62 e	0.13 e	0.99 ab	1.76 a	13.9 e
	50	2.83b	0.26 e	2.72 e	0.16 e	1.04 a	1.74 a	14.7 de
LSD <sub>0.05</sub>		1.65	0.16	2.78	0.10	0.18	0.51	2.8

The data obtained showed that the amounts of moisture content in shoot decreased from 6.28 g. plant<sup>-1</sup>, at 00 mM NaCl (the control plant) to 2.62 g. plant<sup>-1</sup>, at 100 mM NaCl. This represents a relative reduction of 58.3% in the moisture control of shoot of plant grown under 100 mM NaCl. These amounts decreased in roots from 10.19 g. plant<sup>-1</sup> (the control plant) to 2.86 g. plant<sup>-1</sup> (at 100 mM NaCl). This represents a relative reduction of 71.9% in the moisture content of root of plant grown under 100 mM NaCl. These data point out to high reduction in the amount of water taken up by plant when grown under high salinity. Since the moisture control at 100 mM NaCl in shoot (2.62 g.plant<sup>-1</sup>) and root (2.80 g.plant<sup>-1</sup>) are very close this points out that high salinity reduces the ability of plant root to take up water from the saline medium and the rate of water transport from root to shoot is very low. Munns and Passioura (1984) found that the dehydration of shoot and root with increasing NaCl concentration was associated with significant decrease in both the fresh and dry weights of plant. It has been also reported that the mechanisms controlling the growth response to high salt concentration are not specific to salinity. The reductions in the rate of leaf and root growth are probably due to factors associated with water stress rather than a salt specific effect (Munns, 2002).

Table 1 showed marked increase in the values of Sh/Rt ratio, on both F.W. and D.W. basis, with increasing NaCl concentration treatments. The highest Sh/Rt ratio was at 100 mM NaCl. This is due to high reduction in the growth rate of root relative to that of shoot. Also, the observed higher values of Sh/Rt ratio on D.W. basis than on F.W. basis indicate higher moisture content in root than in shoot, at each NaCl treatment.

Table 1 showed also significant decrease in plant height with increasing the concentration of NaCl in the water of irrigation, especially at 50 and 100mM NaCl. The relative reductions in plant height were 3.9, 14.5 and 35.5% at 25,50 and 100 mM NaCl, respectively.

**Photosynthetic pigments :** Table 2 showed significant decrease in the concentration of Chl a at 50 mM NaCl, while that of Chl b was not significant.

The concentration of carotenoids significantly decreased only at 50 mM NaCl treatment. The relative reductions in the concentration of Chl a were 5.0, 15.5 and 17.8% and those of Chl b were 0.5, 13.7 and 12.4% at 25 , 50 and 100 mM NaCl treatments, respectively. This points out to high sensitivity of the photosynthetic system to high salinity. Wang *et al.* (2007) found that chlorophyll content in leaves of wheat seedlings was decreased by 27.5 and 51.6% under salinity of 150 and 300 mM NaCl, respectively. They reported that the growth of wheat seedlings was decreased by salinity and the relative reductions of plant biomass were 36.1 and 46.5% at 150 and 300 mM NaCl, respectively.

**Table 2: The mean value of the concentrations of photosynthetic pigments (mg.100<sup>-1</sup> F.W) in leaves of pea plant after38 days from sowing and of proline (μ M.g<sup>-1</sup> F.W) as influenced by NaCl and kinetin concentrations interactions.**

Treatments		Chl a	Chl b	Carotenoids	Proline
NaCl (mM)	Kinetin (mg.l <sup>-1</sup> )				
0	0	73.52 ab	37.98 ab	30.78 ab	1.26 b
	25	75.58 a	40.36 a	31.90 a	0.85 b
	50	75.20 a	40.26 a	31.72 a	0.96 b
25	0	69.83 ac	37.78 ab	28.06 ac	0.86 b
	25	64.28 be	33.71 bc	27.45 ac	0.79 b
	50	68.36 ad	36.52 ac	28.11 ac	0.75 b
50	0	62.15 cf	32.77 bd	25.77 bc	0.75 b
	25	52.58 f	27.26 d	23.99 c	0.82 b
	50	57.78 ef	30.65 cd	25.69 bc	0.97 b
100	0	60.47 cf	33.29 bd	28.71 ac	5.37 a
	25	59.22 df	32.77 bd	28.31 ac	4.72 a
	50	66.21 ac	36.15 ac	31.10 a	5.57 a
LSD <sub>0.05</sub>		8.94	5.68	4.83	2.24

**Proline :** Table 2 showed significant increase in the concentration of praline in leaves of pea plant only at 100 mM NaCl concentration treatment. However, there were marked reductions in proline contents at 25 and 50 mM NaCl treatments with relative decrease of 31.7 and 40.5%, respectively. However, the relative increase in proline content at 100 mM NaCl was 325.2%. Wang *et al.* (2007) found that the increase of proline in leaves of wheat seedlings was not remarkable at 150 mM NaCl, but was significant at 300 mM NaCl with value of relative increase of 183.0%. Several studies showed that Salt. stressed pea plants contained high concentration of proline in plant leaves (Olmos and Hellin, 1996; Fedina and Tsonev, 1997; Lutt. *et al.*, 1999 and Tester and Davenport, 2003). It has been also reported that proline accumulation in plants leaves is the consequence of salinity (Ahmadi

*et al.*, 2009). It has been proposed that praline accumulation can serve as an adaptive mechanism to salt stress in higher plants (Kumar *et al.*, 2003).

**Salinity kinetin interaction** : Table1 showed no significant effect of foliar application of kinetin on the growth characters of pea plant at each level of salinity treatment. However, Gadallah (1999) found that foliar application of kinetin had ameliorated the deleterious effect of salinity on the growth of shoot of wheat plant. It is also clear that foliar application of kinetin has no significant effects on the concentrations of Chl a, Chl b, carotenoids and proline in leaves of pea plant treated with different concentration of NaCl.

**Pea plant after 88 days from sowing**

**Growth characters** : Table 3 showed significant decrease in the fresh weight of shoot, root, pods and grains with NaCl concentration of 50 mM. It is also clear that the growth of pea plant totally inhibited with irrigation water containing 100 mM NaCl. The relative reductions in shoot F.W. were 04, 41.8 and 100%, and those of root F.W. were 15.8, 41.4 and 100% at 25, 50 and 100 mM NaCl treatments, respectively. These values for pods F.W. were 3.7, 46.0 and 100%, and those for grains F.W. were 33.2, 62.8 and 100%, respectively. The total inhibition of plant growth under 100 mM NaCl treatment could be due to the excessive accumulation of salt in plant which can eventually rise to toxic levels that plant cannot sustain growth. Similar trend was found with respect to grains D.W. in relation to salinity since the relative reduction of grains D.W. was 69.9% at 50 mM NaCl. It can be concluded that salt inhibits plant growth for two reasons; first : the presence of salt reduces the ability of the plant to take up water and leads to slower growth; second: excessive amount of salt entering the transpiration stream will eventually injure cells in the transpiring leaves and this may further reduce plant growth (Munns *et al.*, 2006).

**Table 3 :The mean value of growth characters and total sugars of pea plant after 88 days from sowing as influenced by NaCl and kinetin concentrations interactions.**

Treatments		Fresh weight (g.plant <sup>-1</sup> )				Grains D.W (g.plant <sup>-1</sup> )	Total sugars (mg.g <sup>-1</sup> D.W.)
Nacl (mM)	Kinetin mg.l <sup>-1</sup>	shoot	Root	Pods	Grains		
0	0	46.37 a	31.58 ac	43.07 a	19.36 a	4.10 a	92.92 ac
	25	42.08 a	35.17 a	34.43 ab	11.84 b	2.38 b	107.50 ac
	50	40.95 a	32.53 ab	28.87 ab	10.13 b	2.15 b	79.21 bc
25	0	46.20 a	26.58 ad	41.49 a	12.94 ab	2.54 b	107.67 ac
	25	38.08 ab	22.39 bd	34.41 ab	12.95 ab	2.54 b	137.25 bc
	50	40.06 a	25.88 ad	29.58 ab	11.04 b	2.11 d	104.71 ac
50	0	26.98 bc	18.51 d	23.26 a	7.21 bc	1.48 be	141.84 a
	25	22.17 c	16.62 d	22.13 b	7.92 b	1.65 b	55.00
	50	27.91 bc	21.53 cd	29.74 ab	11.80	2.63 b	16.69 d
100	0	-	-	-	-	-	-
	25	2.41 d	0.95 e	2.25 c	0.54 c	0.11 c	14.45 d
	50	1.70 d	0.86 e	2.17 c	0.52 c	0.13 c	14.64 d
LSD <sub>0.05</sub>		11.47	10.21	14.17	6.87	1.38	60.32

**Relative growth rate (RGR):** The RGR of pea plant, generally, decreased with increasing salinity. The relative growth rates of fresh shoot were 0.79,

0.79 and 0.43 g.g<sup>-1</sup> day and those of fresh root were 0.42, 0.33 and 0.23g.g<sup>-1</sup> day at 0.25 and 50 mM NaCl treatments, respectively. The lower RGR of root than shoot points to higher sensitivity of pea root to salinity than shoot.

**Total sugars** : Table 3 showed marked and not significant increase in the concentration of total sugar in grains with increasing salinity. These levels were 92.92 , 107.67 and 141.84 mg g<sup>-1</sup> D.W. at 0 , 25 and 50 mM NaCl treatments, respectively. The accumulation of soluble carbohydrates in plant has been widely reported as a response to salinity (Ashraf and Harris, 2004).

**Salinity- kinetin interaction:** Foliar application of kinetin showed no significant effect on the growth characters of pea plant and also on total sugar content in grains (Table 1). This is evident by studying the values of RGR of fresh shoot at 00 mM NaCl with 0 , 25 and 50 mg. kinetin l<sup>-1</sup> which were 0.79, 0.71 and 0.71 g.g<sup>-1</sup> day. These values at 25 mM NaCl were 0.79, 0.64 and 0.61g.g<sup>-1</sup> day and at 50 mM NaCl were 0.43, 0.33 and 0.45 g.g<sup>-1</sup> day, respectively. This reveals that foliar application of kinetin did not stimulate the growth of plant shoot at any salinity level. In the case of fresh root, values of RGR at 00 mM NaCl with 0, 25 , 50 mg kinetin l<sup>-1</sup> were 0.42, 0.49 and 0.48 g.g<sup>-1</sup> day, at 25 mM NaCl were 0.32, 0.26 and 0.32 g.g<sup>-1</sup> day and at 50 mM NaCl were 0.23, 0.18 and 0.31 g.g<sup>-1</sup> day, respectively. These data indicate that foliar application of kinetin did not stimulate the growth of root at any salinity level. However, as the plant growth totally inhibited at 100 mM NaCl treatment, foliar application of kinetin at level of 25 mg l<sup>-1</sup> enhanced plant growth slightly. Relative to the control plant (00 mMNaCl and 00 g l<sup>-1</sup> kinetin) and at 100 mM NaCl and foliar application of 25 mg kinetin l<sup>-1</sup>, the values of relative reductions were 94.3% for shoot F.W; 97.3% for root F.W; 93.4% for pods F.W. ; 95.4% for grain F.W.; 95.4% for grains D.W. and 86.6% for total sugars relative to 100 mM NaCl and 00 mg kinetin<sup>-1</sup>, treatment. Foliar application of 50 mg kinetin l<sup>-1</sup> was relatively less effective in stimulating plant growth as 25 mg kinetin l<sup>-1</sup> and, at 100 mM NaCl treatment.

#### **Effect of Salinity- Gibberellic Acid Interaction**

##### **Pea plant at 38 days from sowing**

**Growth characters** : Table 4 showed significant decrease of shoot F.W. at 100 mM NaCl and that of shoot D.W. at 50 mM NaCl. Also, there were significant decreases in root F.W. and D.W. at 50 mM NaCl treatment. The values of relative decrease of shoot F.W. were 2.4, 21.5 and 59.0% and those of root F.W. were 2.4, 36.3 and 71.7% at 25 , 50 and 100 mM NaCl, respectively. These values on D.W. basis were 11.6, 39.1 and 65.2% for shoot, and 18.0, 44.0 and 66.0% for root at 25 , 50 and 100 mM NaCl, respectively. This indicates higher reduction in the growth of root of pea plant than of shoot at each salinity level.

Table 4 also showed that the moisture contents in shoot were 6.28, 6.19, 5.05 and 2.62 g. plant<sup>-1</sup> and those in root were 10.19, 10.42, 6.53 and 2.86 g. plant<sup>-1</sup> at 0, 25, 50 and 100 mM NaCl, respectively. Thus, there were marked decrease in moisture contents in plant organs with increasing salinity which were higher in root than in shoot at each NaCl treatment. This moisture reduction was associated with reduction in plant growth.

**Table 4 :The mean value of growth characters of pea plant after 38 days from sowing as influenced by the interaction between NaCl and gibberellic acid concentrations.**

Treatments		Shoot (g plant <sup>-1</sup> )		Root (g plant <sup>-1</sup> )		Sh /Rt Ratio		Shoot height (cm)
NaCl (mM)	Gib (mg.l <sup>-1</sup> )	F.W.	D.W.	F.W.	D.W.	F.W.	D.W.	
0	0	6.97 bc	0.69 bc	10.69 a	0.50 a	0.65 e	1.39 d	20.74 de
	50	8.36 ac	0.75 ac	9.53 a	0.46 ab	0.88 cd	1.68 cd	39.71 a
	100	8.93 a	0.87 a	9.31 a	0.40 b	0.96 bd	2.19 ac	41.75 a
25	0	6.80 cc	0.61 cd	10.43 a	0.41 ab	0.65 e	1.48 d	19.85 de
	50	8.72 ab	0.74 ac	9.60 a	0.40 b	0.92 cd	1.85 bd	38.71 ab
	100	9.47 a	0.84 ab	9.39 a	0.37 b	1.01 bc	2.26 ab	39.08 ab
50	0	5.47 c	0.42 c	6.81 b	0.28 c	0.81 d	1.52 d	17.68 e
	50	8.20 ad	0.69 bc	7.40 b	0.27 c	1.11 b	2.52 a	35.08 b
	100	6.46 dc	0.49 dc	5.90 b	0.21 cd	1.10 b	2.36 ab	31.04 c
100	0	2.86 f	0.24 f	3.03 c	0.17 d	0.93 cd	1.54 d	13.42 f
	50	3.38 f	0.19 f	2.50 c	0.08 e	1.36 a	2.43 a	22.33 d
	100	3.42 f	0.17 f	2.59 c	0.07 e	1.33 a	2.39 ab	22.47 d
LSD <sub>0.05</sub>		1.60	0.14	1.83	0.09	0.16	0.52	4.00

The increased Sh/Rt ratio, on F.W. and D.W. basis, with increasing salinity (Table 4) points out that the growth of root was decreased by salinity more than shoot. The higher value of Sh/Rt ratio on D.W. basis than on F.W. basis is due to the higher moisture content in root than in shoot at each salinity level. It can be attributed, the higher sensitivity of root to salinity than shoot, to the higher dehydration (water stress) of root than shoot.

Table 4 showed also significant decrease of shoot height with increasing salinity and at and 100 mM NaCl treatment, the relative reduction of shoot height was 35.3%.

**Photosynthetic pigments** : Table 5 showed marked decrease in Chl a, Chl b and carotenoids contents in leaves of pea plant with increasing salinity.

**Proline** : Table 5 showed significant increase in the concentration of proline at 100 mM NaCl treatment with a relative increase of 326.2%. However, there were no significant variations in proline contents at 0, 25, and 50 mM NaCl treatments. It has been reported that high significant proline concentration in plant leaves is, generally, the consequence of salinity (Olmos and Hellin, 1996; Fedina and Tsonev, 1997; Wang *et al.*, 2007 and Ahmadi *et al.*, 2009).

**Salinity-gibberellic acid interaction** : Table 4 showed significant increase of shoot F.W. and D.W. with foliar application by GA<sub>3</sub>. At the control plant (00 mM NaCl) the relative increases, with 50 and 100 mg GA<sub>3</sub> l<sup>-1</sup>, were 19.9 and 28.1% for shoot F.W. and were 8.7 and 20.7% for shoot, D.W. respectively. At 25 mM NaCl treatment., these values were 22.0 and 39.3% for shoot F.W. and were 21.3 and 37.7% for shoot D.W., respectively. At 50 mM NaCl, these values were 49.9 and 18.1% for shoot F.W. and were 64.3 and 16.3% for shoot D.W., respectively. At 100 mM NaCl, these values were 18.2 and 19.6% for shoot F.W. and were- 20.8 % and -29.2 % for shoot D.W., respectively. On the other hand, there were no significant stimulating effects on the root F.W. and D.W. at all levels of salinity as a result of foliar application with GA<sub>3</sub> (Table 4). These data indicate that root growth had no ability to response to GA<sub>3</sub> foliar application at all levels of NaCl salinity.

**Table 5 :The mean value of photosynthetic pigments (mg 100 g<sup>-1</sup> F.W) and proline (µ M g<sup>-1</sup> F.W) in leaves of pea plant after 38 days from sowing as influenced by the interaction between NaCl and gibberillic acid uncentrations.**

Treatments		Chl a	Chl b	Carotenoids	Proline
NaCl (m M) (mM)	Gib. (Mg l <sup>-1</sup> )				
0	0	73.52 ac	37.98 ad	30.78 ac	1.26 b
	50	77.63 ab	43.02 a	34.90 a	0.66 b
	100	81.08 a	42.77 a	35.79 a	0.64 b
25	0	69.83 ac	37.78 ad	28.06 bc	0.86 b
	50	69.41 ac	38.54 ac	31.67 ac	0.67 b
	100	63.70 bd	32.31 cd	31.55 ac	0.65 b
50	0	62.15 bd	32.77 cd	25.77 c	0.75 b
	50	53.22 d	30.03 cd	26.77 bc	0.77 b
	100	52.86 d	29.64 d	27.66 bc	0.86 b
100	0	60.47 cd	33.29 bd	28.71 bc	5.37 a
	50	63.30 bd	35.58 ad	31.41 ac	5.54 a
	100	66.99 ad	41.37 ab	32.75 ab	4.56 a
LSD <sub>0.05</sub>		14.42	7.62	5.53	1.62

Table 4 showed increasing Sh/Rt ratio on both F.W. and D.W. basis with increasing salinity, and within each salinity treatment with increasing dose of GA<sub>3</sub> foliar application. This indicates the effectiveness of GA<sub>3</sub> foliar application for stimulating shoot growth as compared with root growth.

Foliar application of GA<sub>3</sub> significantly increased shoot height relative to the GA<sub>3</sub>- untreated plant. At 00 mM NaCl, the relative increases, with 50 and 100 mg GA<sub>3</sub> l<sup>-1</sup> foliar application, were 91.5 and 101.3%, respectively. At 25 mM NaCl, there values were 95.0 and 96.9%; at 50 mM NaCl, these values were 98.4 and 75.6%; and at 100 mM NaCl, these values were 66.4 and 67.4%, respectively. These date indicate the effective role of the stimulating action of GA<sub>3</sub> for improving shoot height whether under normal non-saline condition or under salinity stress. It is also clear that foliar application by 50 mg GA<sub>3</sub> l<sup>-1</sup> has very close similar effect as by foliar application with 100 mg GA<sub>3</sub> l<sup>-1</sup>.

Foliar application of GA<sub>3</sub> of the control plant (00 mM NaCl) markedly increased Chl a, Chl b and carotenoids. However, this effect was not pronounced at 25, 50 and 100 mM NaCl treatments (Table 5). This indicates the in-effectiveness of GA<sub>3</sub> under salinity stress on photosynthetic pigments contents in plant leaves.

It is clear from Table 5 that there were no significant effects on the concentration of proline in leaves of pea plant as a result of foliar application by 50 and 100 mg GA<sub>3</sub> l<sup>-1</sup>, at each level of NaCl treatment.

**Pea plant at 88 days from sowing**

**Growth characters :** Table 6 showed significant decrease in shoot and root F.W. at 50 mM NaCl treatment in addition total inhibition of plant growth at 100 mM NaCl treatment. The relative reduction of shoot F.W. at 50 mM NaCl was 41.8% and that of root F.W. was 41.4%. This indicates that both shoot and root growth had similarly and significantly decreased at 50 mM NaCl

treatment. There was also significantly decrease in pods F.W. at 50 mM NaCl with relative decrease of 46.0%.

**Table 6 :The mean value of pea plant growth characters (g. plant<sup>-1</sup>) and total sugars ( M g.g<sup>-1</sup> D.W) of plant after 88 days from sowing as influenced by the interaction between NaCl and gibberillic acid concentrations.**

Treatments		Shoot F.W	Root F.W	Pods F.W	Grains F.W	Grains D.W	Total sugars
NaCl(m M)	Gib.(mg l <sup>-1</sup> )						
0	0	46.37 a	31.58 a	43.07 a	19.36 a	4.10 a	92.92 a
	50	37.03 ab	22.73 ac	30.16 bc	9.88 bc	2.08 bc	125.30 a
	100	43.14 a	21.28 bc	30.74 ac	10.08 bc	2.30 bc	112.21 a
25	0	46.20 a	26.58 ab	41.49 ab	12.94 b	2.54 b	107.67 a
	50	39.77 ab	23.37 ac	33.78 ac	10.72 bc	2.13 bc	88.17 a
	100	35.19 ac	21.65 bc	30.19 bc	8.04 cd	1.75 bd	110.17 a
50	0	26.98 bc	18.51 bd	23.26 cd	7.21 cd	1.48 cd	141.84 a
	50	28.59 bc	15.21 cd	13.56 d	3.91 d	0.87 d	106.92 a
	100	20.90 c	9.53 d	13.00 d	3.47 d	0.74 d	124.63 a
100	0	-	-	-	-	-	-
	50	-	-	-	-	-	-
	100	-	-	-	-	-	-
LSD <sub>0.05</sub>		14.46	9.20	12.42	4.79	1.04	74.60

There were also significant reductions in the grains F.W. at 25 and 50 mM NaCl which represented relative decreases of 33.2 and 62.8%, respectively. Similar significant reductions in the grains D.W. were found at 25 and 50 mM NaCl and represented values of relative reduction of 38.1 and 63.9%, respectively. It is also clear from Table 6 that the moisture contents in the grains decreased with increasing salinity and were 12.26, 10.40 and 5.73 g. plant<sup>-1</sup> at 0, 25 and 50 mM NaCl, respectively. This indicates that the dehydration of pea grain is increased when plant is grown under salinity stress.

**Total sugars** : Table 6 showed no significant variations in the amounts of total sugars in the grains with increasing salinity. However, marked increase in total sugar can be observed at 50 mM NaCl treatment. It is reported that accumulation of soluble carbohydrates in plant occurs as a response to salinity (Ashraf and Harris, 2004).

**Salinity-gibberillic acid interaction** : Foliar application of GA<sub>3</sub>, at levels of 50 and 100 mg l<sup>-1</sup>, had no significant effect on the shoot and root F.W. at all salinity treatments . However, at 00 mM NaCl (the control plant), foliar application by 100 mg GA<sub>3</sub> l<sup>-1</sup> decreased significantly root F.W. Also , foliar application by 50 and 100 mg GA<sub>3</sub> l<sup>-1</sup> decreased significantly pods F.W. at 00 mM NaCl (the control plant).

Table 6 showed that foliar application of GA<sub>3</sub> at levels of 50 and 100 mg l<sup>-1</sup> and at the control plant (00 mM NaCl) significantly decreased grains F.W. This is also found at 25 mM NaCl with 100 mg GA<sub>3</sub> l<sup>-1</sup>. Also, there were significant decreases of grains D.W. at 00 mM NaCl (the control plant) with 50 and 100 mg GA<sub>3</sub> l<sup>-1</sup> and at 25 mM NaCl treatment with 100 mg GA<sub>3</sub> l<sup>-1</sup>. At the control plant ( 00 mM NaCl), the relative decreases of grains F.W. and D.W, with 50 mg GA<sub>3</sub> l<sup>-1</sup> foliar application, were 49.0 and 49.3%, respectively,

and with 100 mg GA<sub>3</sub>l<sup>-1</sup> foliar application, were 47.9 and 43.9%, respectively. These data indicate that foliar application of GA<sub>3</sub> did not significantly stimulate the growth of 88 days old plant. However, another study reported that GA<sub>3</sub> foliar application usually overcomes the effect of salt stress and improve the growth parameters; yield and yield components of salt-stressed plant (Akbari *et al.*, 2008).

**Relative growth rate (RGR) :** The relative growth rates at 0, 25 and 50 mM NaCl treatments (without GA<sub>3</sub> treatments) for shoot F.W. were 0.79, 0.79 and 0.43 g.g<sup>-1</sup> day, and for root F.W. were 0.42, 0.32 and 0.23 g.g<sup>-1</sup> day, respectively. This clearly shows that root growth was more inhibited by salinity than shoot at each salinity level. The data also showed that 50 mM NaCl treatment decreased markedly the RGR of shoot and root as compared with those of 00 mM and 25 mM NaCl.

The values of the RGR at 0, 25 and 50 mM NaCl with foliar application by 50 mg GA<sub>3</sub>l<sup>-1</sup> were 0.57, 0.62 and 0.41 g.g<sup>-1</sup> day for shoot F.W. and were 0.26, 0.28 and 0.16 g.g<sup>-1</sup> day for root F.W., respectively. These values, with 100 mg GA<sub>3</sub>l<sup>-1</sup>, were 0.68, 0.51 and 0.29 g.g<sup>-1</sup> day for shoot F.W. were 0.24, 0.25 and 0.08 g.g<sup>-1</sup> day for root D.W. These data indicate low response of root growth to GA<sub>3</sub> foliar application and relatively high response of shoot growth to GA<sub>3</sub> foliar application.

### CONCLUSION

The obtained results showed that increasing NaCl concentration in water of irrigation more than 25 mM (about more than 4 dSm<sup>-1</sup>) after germination decreased significantly the growth and yield of pea plant grown on sand culture. Increasing the concentration of NaCl to 100 mM decreased the growth of 38 days old plant and inhibited totally the growth of 88 days old plant.

Foliar application of kinetin at 25 and 50 mg l<sup>-1</sup> had no significant effects to overcome salt stress on plant growth. On the other hand, foliar application by GA<sub>3</sub> at 50 and 100 mg l<sup>-1</sup> improved growth characters of 38 days old plant and did not affect the growth and yield of 88 days old plant.

### REFERENCES

- Ahmadi, A., Y.Emam and M. Pessaraki (2009) Response of various cultivars of wheat and maize to salinity stress. *J. Food Agric. Environ.* 7:123-128
- Akbari, N., M.Barani and H.Ahmadi (2008) Effect of gibberellie acid (GA<sub>3</sub>) on agronomic traits of green gram (*Vigna radiata* L. Wilezek) irrigated with different levels of saline water. *World Applied Sci. J.* 5: 199-203
- Ashraf, M. and P.J.C. Harris (2004) Potential biochemical indicators of salinity tolerance in plants. *Plant Sci.* 166: 3-16
- Bates, L.S., R.P.Waldern and I.D. Teare (1973) Rapid determination of free proline for water – stress studies. *Plant and Soil* 39: 205-205.

- Dubois, M.; K.A. Gilles; J.K.Hamilton; P.A. Roers, and F. Smith (1956) Coloremtric method for determination of sugar and related substances. *Anal. Chem.* 28: 350-356.
- 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: 735-740
- Gadallah, M.A.A.(1999) Effect of kinetin on growth, grain yield and some mineral elements in wheat plants grown under excess salinity and oxygen deficiency. *Plant Growth Regul.* 27 : 63-74
- Hewitt, E.G.(1966)*Sand and Water Culture Methods Used in The Study of Plant Nutrition.* Technical Communication No. 22. 2<sup>nd</sup> Ed. Commonwealth Agricultural Bureaux, Fornham Royal, England.
- Kumar, S.G.,A.M.Reddy and C.Sudhakar (2003). NaCl effects on proline metabolism in two high yielding genotypes of mulberry (*Morus alba* L.) with contrasting salt tolerance. *Plant Sci.* 165:1245-1251.
- Lutt, S., V.Majerus and J.M. Kinct (1999). NaCl effects on proline metabolism in rice (*Oryza sativa* L)seedlings. *Physiol. Plant* : 105:450-458
- Moran, R. and D. Porath (1980) Chlorophyll determination in intact tissues using N,N-dimethylformamide. *Plant Physiol.*65: 478-479.
- Munns, R. and J.B.Passioura (1984) Effect of prolonged exposure to NaCl on the osmotic pressure of leaf xylem sap from intact, transpiring barely plants. *Aust. J. Plant Physiol.* 11: 497-507.
- Munns, R.(2002)*Comparative physiology of salt and water stress.* *Plant, Cell and Environ.* 25: 239-250
- Munns, R., R.A. James, and A. Lauchli (2006) Approches to increasing the salt tolerance of wheat and other cereals. *J.Exp.Bot.* 57: 1025-1043
- Olmos, E. and E.Hellin (1996) Mechanism of salt tolerance in a cell line of *Pisum sativum* : biochemical and physiological aspects. *Plant Sci.* 120: 34-45
- SAS Inst. (1985): *SAS User's Guide, Statistics, Version 5 Ed.* SAS Inst. Inc., Cary, Nc.
- Tester, M. and R.Davenport (2003) Na<sup>+</sup> tolerance and Na<sup>+</sup> transport in higher plants. *Annals of Botany* 91: 503-527.
- Wang, Z-Q., Y-Ze Yuan, J-Q.Ou, Q-H. Lin, and C-F. Zhang (2007) Glutamine synthetase and glutamate dehydrogenase contribute differentially to proline accumulation in leaves of wheat (*Triticum aestivum* L.) seedlings exposed to different salinity. *J. Plant Physiol.* 164: 695-701.

استجابة نبات البسلة النامي في مزرعة رملية تحت إجهاد ملحي للإضافة الورقية  
بالكينيتين وحامض الجبريلليك  
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أجريت تجربة أصص في كلية الزراعة بالبستان - جامعة الإسكندرية لى تقييم استجابة  
نبات البسلة النامي في مزرعة رملية تحت إجهاد ملحي للإضافة الورقية بمحلول الكينيتين او حامض  
الجبريلليك .

زرعت البذور في اصص تحتوى 10 كيلو جرامات رمل سبق غسله بالماء واستخدم في  
الرى ماء مكون من محلول مغذى يحتوى على صفر ، 25 ، 50 ، 100 مليمول كلوريد صوديوم  
واستخدم في الإضافات الورقية محلول الكينيتين بتركيزات صفر ، 25 ، 50 ملجرام/لتر او محلول  
حامض الجبريلليك بتركيزات صفر ، 50 ، 100 ملجرام/لتر وجمعت عينات النبات بعد 38 وبعد  
88 يوم من الزراعة لاجراء التحاليل المختلفة .

أوضحت نتائج الدراسة حدوث نقص معنوى للنمو الخضرى ولمحصول الحبوب مع زيادة  
تركيز كلوريد الصوديوم في ماء الرى . كذلك حدث نقص معنوى في كمية كل من كلوروفيل أ  
وكلوروفيل ب والكاروتين ولكن حدثت زيادة معنوية في كمية البرولين في اوراق النبات مع زيادة  
الملوحة . ولم تؤثر معنويا الإضافة الورقية بالكينيتين على صفات النمو الخضرى او على الصبغات  
في الأوراق للنبات عند عمر 38 يوم بينما حدث تحفيزا صغيرا نسبياً للنمو للنبات عند عمر 88 يوم  
المعامل بمحلول يحتوى على 100مليمول كلوريد صوديوم . وقد أدت الإضافة الورقية بحامض  
الجبريلليك الى تحسين النمو الخضرى للنبات عند عمر 38 يوم بينما لم يحدث تحسين للنمو في حالة  
النبات عند عمر 88 يوم حيث أدت زيادة الملوحة الى نقص كل من النمو و محصول الحبوب .

قام بتحكيم البحث

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