Trials for Alleviating the Adverse Effects of Soil Salinity on Seed Germination, Growth and Nutritional Status of Wheat Seedlings (*Triticum Aestivum* L.)

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Abstract: This study was conducted during 2011 and 2012 seasons to examine the effects of seeds soaking in salicylic acid (SA) at 0.5 mM on seed emergence %, growth, plant pigments K^+/ Na^+ as well as percentages and uptakes of N, P, K, Mg and Ca in wheat cv. Giza 164 grown under different soil salinity levels (40 & 80 & 160 mM NaCl). Salinity caused by NaCl at 40 to 160 mM measurably reduced emergence %, all growth characters, plant pigments, K^+/ Na^+ as well as percentages and uptake of N, P, K, Mg and Ca in relative to non- salinization. In contrast, the investigated parameters were considerably stimulated with soaking the seeds with SA under either non-salinity and/ or salinity conditions comparing with using saline soil alone. Soaking the seeds in SA solution was very effective in counteracting the adverse effects of soil salinity on wheat seedlings. Soaking seeds of wheat cv. Giza 164 on salicylic acid solution at 0.5 mM was beneficial to improve growth and nutritional status under soil salinity conditions.

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1. Introduction

Salinity is one of the main problems that negatively affect soil fertility and limit plant production. Soil salinity affects osmatic stress, decreasing water availability, ionic stress and changes in the cellular ionic balance. The effect of salinity was determined at seedling stage of wheat range from reduction in germination %, fresh and dry weights of shoots and roots to the uptake of various nutrients **(Afzal *et al*., 2005).** Salt stress increases the formation of reactive oxygen species causing deterioration of membrane functions **(Erdal *et al*., 2010).** Plants have evolved enzymatic and nonenzymatic defense mechanisms in order to reduce oxidative damages by detoxifying free radicals. The enzymatic defense system includes peroxidase and catalase. These enzymes detoxify H_2O_2 (an active oxygen species) to H_2O **(Borsani *et al*., 2011).**

Salicylic acid (SA) is recognized as an endogenous signal molecule mainly involved environmental stress tolerance in plants **(Dela- Rosa and Maiti, 1995).** It is synthesized by many plants and is accumulated in the plant tissues under the impact of unfavourable abiotic factors, contributing to the increase of plant resistance to salinization **(Ding *et al*., 2002 and Kang and Saltveit, 2002).** The role of SA in defense mechanisms has been known for several years **(Chinnusamy and Zhu, 2003; Shim *et al*., 2003 and Gunes *et al*., 2007)**. It is a key endogenous signal involved in plant deference response to environmental stressors such as salinity **(Raskin *et al*., 1990)**. SA included increase in the resistance of wheat seedlings to salinity **(Shakirova and Bezrukova, 1997).** Thus, the detrimental effects of high salts on the early growth of wheat seedlings may be alleviated by treating seeds with the proper concentration of a suitable hormone **(Darra *et al*., 1973).**

Previous studies showed that SA is beneficial on counteracting the adverse effects of soil and water salinity on germination of seeds, growth and nutritional status of agronomical and horticultural crops **(Pancheva *et al*., 1996; Ashraf *et al*., 1997; Gutirrez-Coronado *et al*., 1998; Lee *et al*., 2001; Hamada and Al- Hakimi, 2001; Tari *et al*., 2002; Shakirova *et al*., 2003; Khan *et al*., 2003; Singh and Usha, 2003; Khodary, 2004; Guzelordu, 2005; Basra *et al*., 2005; El- Tayeb 2005; Gunes *et al*., 2005 Szepesi *et al*., 2005; Arfan *et al*., 2007; Kaydan *et al*., 2007 and Erdal *et al*., 2011).**

The target of this study was examining the effect of soaking the seeds of wheat cv. Giza 164 on SA solution on germination of seeds, growth and nutritional status of the plants grown under saline soil conditions. Also, the merit was testing the tolerance of such wheat cv. to soil salinity and the role of SA in alleviating the adverse effects of salinity on germination growth and nutritional status.

2.Material And Methods

This pot trial was conducted in greenhouse located at Sohag district in Sohag Governorate during 2011 and 2012 seasons (2nd week of Nov.). Wheat cv. Giza 164 was selected for carrying out this experiment. Seeds were surface sterilized then rinsed with sterilized water and air dried.

This experiment included the following eight treatments:-

1. Control (unsalinized soil).
2. Soil salinity at 40 mM NaCl with out SA.
3. Soil salinity at 80 mM NaCl with out SA.
4. Soil salinity at 160 mM NaCl with out SA.
5. Soaking seeds in SA at 0.5 mM.
6. Soil salinity at 40 mM + SA at 0.5 mM.
7. Soil salinity at 80 mM + SA at 0.5 mM.
8. Soil salinity at 160 mM + SA at 0.5 mM.

Each treatment was replicated three times, ten plants per each replicate. Salicylic acid at 0.5 mM (**Shakirova *et al*., 2003)** (90 mg SA L^{-1} water) was dissolved in distilled water and the pH was adjusted at 6.5 with NaOH. The seeds were soaked with SA for 12 h in the dark at 22 ̊C. Each plastic pot (14 × 17 cm) was filled- up with 2 kg air- dried silty clay soil and kept inside the glass greenhouse under natural light

Soil Description:-

The soil samples were air dried and sieved (< 2 mm stainless steel mesh) before determination of chemical properties. They were dried powdered and mixed thoroughly. The texture of the soil is silty clay (organic matter 24 %, total salt 0.033 %, pH 7.99, total N 0.6 % available P 14.2 mg P Kg^{-1} dry soil and exchangeable K 400 mg K kg^{-1} dry soil) according to **Wilde *et al*. (1985**).

Soil salinity was caused by NaCl to adjust to 40, 80 and 160 mM NaCl concentrations (i.e. 1.46, 2.92 and 5.84 g NaCl/ L^{-1} soil, respectively.

Soil was allowed to equilibrate the greenhouse for one week before sowing the seeds. All pots were fertilized with 80, 60 and 50 mg kg N, P and K/ pot, respectively. Presoaked with SA seeds ten seeds were sown in each pot.

The seedlings were irrigated with non-salinized tap water at field capacity every 2 – 3 days. Unsoaked with SA seeds were used and some seeds were soaked with SA only and sown in non-salinized. The design of this investigation was complete randomized. After 30 days of sowing emergence % was calculated. The seedlings were harvested and separated into shoots and roots for measuring averages fresh and dry weight of shoots and roots (mg/ plant). Whole plant dry weight was estimated by summation of dry shoots and roots.

Plant pigments namely chlorophylls a & b and total carotenoids (mg/ g^{-1} F.W.) were determined from fully expanded leaves according to **Arnon (1948)**. Total chlorophylls as mg/ g^{-1} F.W. was calculated by summation of chlorophyll a plus chlorophyll b. Both K and Na in the dried samples of whole plant were determined by a flame photometer **(A.O.A.C., 1995),** then K^+/ Na^+ was calculated. Also, percentages of N, P, K, Mg and Ca were determined in the dried whole plant (according **A.O.A.C., 1995**). Uptake of each nutrients/ plant was recorded by multiplying percentage of each clement by dry of whole plant (mg in the whole per plant).

Statistical analysis was done according to **Mead *et al*. (1993)** using revised L.S.D at 5 % for made all comparisons among different treatment means.

3.Results and Discussion

1-Emergence %:

It is clear from the data in Table (1) that soil salinity at 40 to 160 mM with or without soaking the seeds in salicylic acid (SA) at 0.5 mM significantly reduced emergence % in relative to the unsalinized conditions or soaking the seeds in SA alone. Soaking the seeds in SA significantly increased emergence % compared to unsoaking. Soaking the seeds in SA and planting in salinity soil (40 to 160 mM) significantly reduced the adverse effects of salinity on emergence % rather than unsoaking plus planting the seeds in salinity soil significant differences on such character were noticed among all SA and soil salinity treatments. The reduction on emergence % significantly was associated with increasing salinity concentrations. The maximum values were recorded when the seeds were soaked in SA at 0.5 mM and grown under non- salinized soil (88.9 and 90 % during both seasons, respectively). The lowest values (31.2 and 33.3 % in both seasons) were recorded when the seeds were soaked in SA followed by planting them under soil salinity that reached 160 mM NaCl. These results were true during both seasons.

The adverse effects of salinity on the activity of enzymes that were responsible for nutrition of embryo and increasing ABA as well as the biosynthesis of IAA and cytokinins could explain the present results. The beneficial effect of SA on enhancing emergence % could be explained on the light of its action on encouraging the biosynthesis of IAA and cytokinins and different enzymes **(Raskin *et al*., 1990).**

These results are in agreement with those obtained by **Erdal *et al*., (2011)**. The results of **Kaydan *et al*., (2007)** supported the benefits of SA on enhancing emergence %.

2-Growth characters:

It is obvious from the data in Table (1) that planting wheat seedlings in salinity soil at 40 to 160 mM NaCl significantly reduced all growth characters namely fresh and dry weights of shoots and roots as well as whole plant dry weight in relative to unsalinization condition with or without soaking in SA solution. The depression in these characters significantly was in proportional to the increase in salinity levels. Soaking the seeds in SA under unsalinity conditions significantly stimulated all growth rather than unsoaking under the same conditions. Using Si in salinized soil significantly controlled the great damage on growth caused by soil salinity compared with unsoaking under salinity conditions. The maximum growth values were recorded on the seedlings that their seeds were soaked in SA and grown under non- salinized soil. Salinity conditions with using SA gave the lowest values. These results were true during both seasons.

The action of SA on increasing the resistance of plants to different stresses as well as enhancing the uptake of nutrients especially Mg **(Raskin *et al*., 1990)** could explain the plant.

These results are in agreement with those obtained by **Erdal *et al*. (2011)**. The results of **Kaydan *et al*., (2007)** supported the benefits of SA on enhancing emergence %.

3-Plant pigments and K^+/ Na^+ in the leaves:

It is evident from the data in Table (2) that the three plant pigments (chlorophylls a & b and total carotenoids), total chlorophylls and K^+/ Na^+ significantly tended to reduce on plants grown under soil salinity conditions (40 to 160 mM) in relative to the check treatment (unsalinized soil). The depression was significantly in proportional to the increase in salinity soil levels from 40 to 160 mM. Soaking the seeds in SA under non- salinized soil was superior than unsoaking under the same conditions. Results further reveal that soaking the seeds in SA and planting under soil salinity was significantly preferable than unsoaking + planting under salinity conditions. This confirmed the beneficial effect of soaking the seeds presowing in SA solutions to avoid the harmful effects of salinity on plant pigments and K^+/ Na^+. The maximum values were recorded on seedlings that subjected to seed soaking in SA and sowing in unsalinized soil. Sowing the unsoaked seeds in SA in soil salinized with NaCl at 160 gave the lowest values. Similar trend was noticed during both seasons.

The adverse effects of salinity on plant pigments and K^+/ Na^+ might be attributed to its negative action on destroying plastids as well as inhibiting plant metabolism and plant uptake of Mg **(Nijjar, 1985).** The reduction on Na^+ and the promotion on Na^+ could result in reducing K^+/ Na^+.

The results of **Singh and Usha (2003); Khodary (2004) and Khan *et al*., (2009)** emphasized the beneficial effect of using SA on counteracting the adverse effects of soil salinity on plant pigments.

4-Concentrations and uptake of N, P, K, Mg and Ca by plant:

It is worth to mention from the data in Tables (3 & 4) that sowing wheat seeds in salinized soil at 40 to 160 mM without soaking in SA significantly reduced concentrations and uptake of N, P, K, Mg and Ca in relative to growing the seeds in unsalinized soil with or without soaking the seeds in SA solution. The reduction was significantly associated with increasing salt concentrations from 40 to 160 mM. Soaking the seeds presowing in SA significantly was responsible for alleviating the adverse effects of salinity on concentration and uptake of these nutrients when the seeds were sowing in salinized soil. The highest concentration and uptake of these nutrients were observed owing to soaking the wheat seeds in SA solution followed by sowing in unsalinized soil. Unsoaking the seeds in SA followed by sowing them in salinized soil at 160 mM NaCl gave the lowest values. These results were true during both seasons.

The inferior effects of salinity on concentration and uptake of nutrients might be attributed to its negative effect on enhancing soil osmotic pressure, nutrient imbalance, impaired internal distribution and shoot transport of minerals **(Gunes *et al*., 2007).**

The action of SA on increasing the resistance of plants to different stresses as well as enhancing the uptake of nutrients especially Mg **(Raskin *et al*., 1990)** could explain the plant.

These results are in harmony with those obtained by **Borsani *et al*. (2001); Khan *et al*. (2003); Khodary (2004) and Erdal *et al*. (2011).**

As a conclusion, it is suggested to soak the seeds of wheat cv. Giza- 164 in SA solution at 0.5 mM as a potential growth regulator for improving plant growth and nutritional status under soil salinity conditions.

Table (1): Effect of soaking the seeds of wheat cv. Giza- 164 plants in salicylic acid on emergence % and some growth characters under soil salinity conditions during 2011 and 2012 seasons.

Salinity & salicylic acid (SA) treatments	Emergence %		Shoot fresh weight (mg/ seedling^{-1})		Shoot dry weight (mg/ seedling^{-1})		Root fresh weight (mg/ seedling^{-1})		Root dry weight (mg/ seedling^{-1})		Whole plant dry weight (mg/ seedling^{-1})	
	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012
Control (unsalinized soil)	85.6	87.1	218.3	225.0	36.0	38.3	65.4	68.3	11.1	11.6	47.1	49.9
Soil salinity at 40 mM NaCl	71.0	72.8	191.0	196.0	31.5	33.0	57.0	59.0	9.7	9.4	41.2	42.4
Soil salinity at 80 mM NaCl	62.3	64.0	150.0	157.5	24.0	25.1	45.0	47.5	7.7	7.7	31.7	32.8
Soil salinity at 160 mM NaCl	31.2	33.3	100.5	106.8	16.7	17.0	30.9	32.1	5.3	5.0	22.0	22.0
SA at 0.5 mM	88.9	90.0	222.3	230.5	36.3	39.3	66.7	69.3	11.9	11.0	48.2	50.5
Soil salinity at 40 mM + SA	76.0	78.2	201.0	208.0	32.2	36.0	60.0	61.3	10.0	9.8	42.2	46.0
Soil salinity at 80 mM + SA	71.0	73.0	171.0	180.0	28.1	31.0	51.3	54.0	8.0	8.2	36.1	39.0
Soil salinity at 160 mM + SA	42.3	46.3	120.0	128.3	19.3	20.6	36.0	37.7	5.1	5.9	24.4	25.7
Revised L.S.D at 5 %	**1.9**	**2.1**	**2.4**	**2.3**	**1.2**	**1.4**	**1.2**	**1.1**	**0.9**	**1.**	**1.1**	**1.3**

Table (2): Effect of soaking the seeds of wheat cv. Giza- 164 plants in salicylic acid on some plant pigments and K^+/Na^+ in the leaves under soil salinity conditions during 2011 and 2012 seasons.

Salinity & salicylic acid (SA) treatments	Chl. a (mg/ g^{-1} F.W.)		Chl. b (mg/ g^{-1} F.W.)		Total chlorophylls (mg/ g^{-1} F.W.)		Total carotenoids (mg/ g^{-1} F.W.)		Leaf k^+/Na^+	
	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012
Control (unsalinized soil)	11.1	13.1	5.6	6.6	16.7	19.7	2.8	3.3	1.88	1.94
Soil salinity at 40 mM NaCl	9.1	9.6	4.6	4.8	13.7	14.4	2.3	2.4	0.88	0.99
Soil salinity at 80 mM NaCl	7.2	7.3	3.6	3.7	10.8	11.0	1.8	1.9	0.71	0.78
Soil salinity at 160 mM NaCl	5.1	5.5	2.6	2.8	7.7	8.3	1.3	1.4	0.57	0.56
SA at 0.5 mM	12.1	14.1	6.0	7.1	18.1	21.2	3.0	3.6	1.91	1.97
Soil salinity at 40 mM + SA	10.1	10.9	5.0	5.5	15.1	16.4	2.5	2.8	1.51	1.60
Soil salinity at 80 mM + SA	8.5	8.9	4.2	4.5	12.7	21.6	2.1	2.3	1.07	1.13
Soil salinity at 160 mM + SA	6.6	6.5	3.3	3.3	9.9	9.8	1.7	1.7	0.99	1.05
Revised L.S.D at 5 %	**0.6**	**0.7**	**0.5**	**0.6**	**0.8**	**1.0**	**0.3**	**0.4**	**0.07**	**0.08**

Table (3): Effect of soaking the seeds of wheat cv. Giza- 164 plants in salicylic acid on percentages of N, P, K, Mg and Ca in the whole plant under soil salinity conditions during 2011 and 2012 seasons.

Salinity & salicylic acid (SA) treatments	N % in the whole plant		P % in the whole plant		K % in the whole plant		Mg % in the whole plant		Ca % in the whole plant	
	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012
Control (unsalinized soil)	1.51	1.60	0.22	0.22	1.38	1.40	0.60	0.64	2.71	2.77
Soil salinity at 40 mM NaCl	1.41	1.50	0.15	0.15	1.18	1.19	0.40	0.44	2.00	2.06
Soil salinity at 80 mM NaCl	1.35	1.39	0.12	0.13	1.15	1.17	0.36	0.40	1.89	1.95
Soil salinity at 160 mM NaCl	1.30	1.22	0.09	0.11	1.10	1.11	0.33	0.35	1.60	1.67
SA at 0.5 mM	1.71	1.82	0.25	0.25	1.44	1.47	0.65	0.68	2.84	2.91
Soil salinity at 40 mM + SA	1.47	1.57	0.18	0.18	1.34	1.35	0.44	0.50	2.41	2.48
Soil salinity at 80 mM + SA	1.41	1.45	0.15	0.15	1.28	1.29	0.40	0.45	2.02	2.11
Soil salinity at 160 mM + SA	1.35	1.35	0.12	0.13	1.22	1.22	0.35	0.40	1.85	1.82
Revised L.S.D at 5 %	**0.04**	**0.05**	**0.03**	**0.04**	**0.03**	**0.04**	**0.03**	**0.03**	**0.05**	**0.04**

Table (4): Effect of soaking the seeds of wheat cv. Giza- 164 plants in salicylic acid on the uptake of N, P, K, Mg and Ca by plant under soil salinity conditions during 2011 and 2012 seasons.

Salinity & salicylic acid (SA) treatments	N uptake/ plant (mg)		P uptake/ plant (mg)		K uptake/ plant (mg)		Mg uptake/ plant (mg)		Ca uptake/ plant (mg)	
	2011	2012	2011	2012	2011	2012	2011	2012	2011	2012
Control (unsalinized soil)	0.71	0.80	0.10	0.11	0.65	0.70	0.28	0.32	1.28	1.38
Soil salinity at 40 mM NaCl	0.58	0.64	0.06	0.06	0.49	0.50	0.16	0.19	0.82	0.87
Soil salinity at 80 mM NaCl	0.43	0.46	0.04	0.04	0.36	0.38	0.11	0.13	0.60	0.64
Soil salinity at 160 mM NaCl	0.29	0.27	0.02	0.02	0.24	0.25	0.07	0.08	0.35	0.37
SA at 0.5 mM	0.82	0.92	0.12	0.13	0.69	0.74	0.31	0.34	1.37	1.47
Soil salinity at 40 mM + SA	0.62	0.72	0.08	0.08	0.56	0.62	0.19	0.23	1.01	1.14
Soil salinity at 80 mM + SA	0.51	0.57	0.05	0.06	0.46	0.50	0.14	0.18	0.73	0.82
Soil salinity at 160 mM + SA	0.33	0.35	0.03	0.03	0.30	0.31	0.09	0.10	0.45	0.47
Revised L.S.D at 5 %	**0.09**	**0.10**	**0.02**	**0.02**	**0.04**	**0.05**	**0.02**	**0.03**	**0.10**	**0.09**

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