Journal of Animal and Veterinary Advances 8 (2): 362-367, 2009
ISSN: 1680-5593

Effects of GA_3 and Kinetin Pre-Sowing Treatments on Seedling Emergence and Seedling Growth in Wheat under Saline Conditions

Zekeriya Akman
Department of Field Crops, Faculty of Agriculture, Suleyman Demirel University, 32600 Isparta, Turkey

Abstract: This study was conducted to evaluate the effect of NaCl salinity and Gibberellic Acid (GA_3) and Kinetin (KIN) pre-sowing treatments on the germination and emergence of 3 winter-wheat cultivars. Thus, the seeds soaked with GA_3 and kinetin were germinated using 0, 94, 164 and 240 mM NaCl solutions in pod soil. The percentage seedling emergence of wheat genotypes decreased and delayed with salinity stress conditions. The pre-treatments of hormones, especially GA_3, not only hastened seedling emergence but also overcame the germination-delaying effect of the high concentrations of NaCl in all investigated wheat genotypes except for initial germination of Kisla domestic variety. Exogenous kinetin treatment was not as effective as GA_3. Seedling growth parameters under salt-stress conditions decreased with increased salt stress. Under salinity conditions, GA_3 was significantly stimulated root length and shoot height in Kisla domestic variety and B-22 line, respectively.

Key words: Genotypes, plant growth regulators, salt stress, seedling emergence, wheat

INTRODUCTION

Salinity is one of the major ecological problems to food production because it limits crop yield and restrict use of land previously uncultivated. It is estimated that over 800 million hectares of land in the world is affected by salinity (Shannon, 1997). The salts accumulating in the soil profile are mainly chlorides, sulfates, bicarbonates and borates and the most dominating salt in the soil profile is NaCl (Richards, 1954; Geschenke, 1984). Salinity has negative effects on seed germination and on the emergence of seedlings because of limited water uptake (Reynolds, 1975; Braun and Khan, 1976) and toxic effect (Redmann, 1974). Salinity cause significant reductions in the rate and final percentage of germination, which in turn may lead to uneven stand establishment and reduced crop yields (Mer *et al.*, 2000). Therefore, reducing the spread of salinization and increasing the salt tolerance of crops are issues of global importance. Salt tolerance refers to the ability of plants to maintain the growth under saline conditions. Foolad and Lin (1997) reported that differences in salt tolerance exist, not only among different species, but also within certain species. Wheat is regarded as moderately tolerant to salt (Mass and Hoffmann, 1977) but this salt sensitivity may also differ according to genotypes (Ashraf *et al.*, 1997; Singh *et al.*, 1997; Bagci *et al.*, 2007).

The salt tolerance at the early growth stage differs from that developed during the late growth stages (Ashraf *et al.*, 1997). In many crops, the seed germination and early seedling growth are the most sensitive stages to environmental stresses such as salinity (Sivritepe *et al.*, 2003; Yildirim and Guvenç, 2006). Mass and Poss (1989) reported that wheat was the most sensitive during the vegetative and early reproductive stages to salinity stress. Morever, there is increasing evidence showing that better germination and seedling growth have a great effect on final yield as shown in wheat (Bagci *et al.*, 2000). Therefore, information regarding growth stage response to salinity is important in adopting suitable genetic and management strategies for saline soils.

One of the most effective ways to overcome salinity problems is also the use of Plant Growth Regulators (PGRs) (Jamil and Rha, 2007). Plant growth regulators have been found to play a central role in the integration of the plant responses expressed by plants to stress conditions (Amzallag *et al.*, 1990). Gibberellic acid and kinetin have been reported to increase germination percentage and seedling growth and overcome the preventive effects of the salt stress on germination (Kabar and Baltepe, 1987). Kabar (1990) stated that the seeds whose germination was normally inhibited by high levels of salts, may be grown in medium or soils with rather high saline content if they are pre-treated with a suitable growth regulator.

The objectives of the present study were to evaluate the effects of NaCl salinity, gibberellic acid and kinetin pre-sowing treatments on the emergence of 3 winter wheat cultivars and to determine their potential for salt tolerance and their response to hormone treatments during germination and seedling growth.

MATERIALS AND METHODS

Seed material and PGRs pre-treatments: Caryopses of Altin-98 commercial wheat variety (*Triticum aestivum* L.), B-22 wheat line (*Triticum aestivum* L.) and Kisla domestic variety (*Triticum durum* L.) were used as material and obtained from Field Crops Research Institute, Eskisehir, Turkey; Agricultural Faculty, Gaziosmanpasa University, Turkey and Kisla village, Isparta, Turkey, respectively. The seeds were surface sterilized with 1.0% sodium hypochlorite. Before sowing, they were soaked in distilled water (control, C) and of aqueous solutions of growth regulators in predetermined concentrations (0.5 mM Kinetin (KIN), 2.0 mM Gibberellic Acid (GA_3)) for 3 h at room temperature. Thereafter, the solutions were decanted off and the seeds were vacuum dried for 1 h (Braun and Khan, 1976).

Seed emergence and plant growth: Loamy soil was collected from the soil surface (0-15 cm), air-dried, ground, passed through a 5 mm mesh screen and thoroughly mixed. The soil consisted of 23.1 clay, 33.9 silt, 43 sand and 1.34% (organic matter). The pH of the soil was 8.1 and its electrical conductivity ranged from 0.2-0.4 mmhos cm^{-1}. Four lots of soil, of 100 kg each, were spread separately over thick plastic sheets. These separated soils were moistened with treatment solutions of 0 (no NaCl), 94 mM, 164 mM and 240 mM concentration of NaCL in deionized water to achieve a 13% moisture content. The electrical conductivities of these salt solutions were determined with a Jenway Model 470 conductivity meter (Jenway Limited) as 8, 16 and 24 dS m^{-1}, respectively. Each salinity treatment was consistent of 36 polyethylene bags containing 2 kg of the soil mixtures described above and ten seeds were sown in each bag at a depth of about 8-12 mm. For each salinity levels (0 (no NaCl), 94, 164 and 240 mM concentration of NaCL), there were four replicates for each pre-sowing hormones treatment (control, KIN and GA_3) and genotype. After sowing, bags containing soil were randomized on a platform in the greenhouse. Immediately soils were watered and thereafter watering was carried out on alternate days. Emergence of seedlings was recorded every day. Plants were harvested 30 days after sowing and their fresh weights determined. The seedlings were counted and cleaned and their morphological characteristics were recorded. Shoot height and root length were also determined in centimetres. Dry weights of seedlings were determined 24 h after plants were maintained in an oven at 80°C.

Statistical analysis: The experiment arranged in a completely randomised design with 4 replications. The 1st factor had 3 seed pre-treatments (control, KIN and GA_3) and the 2nd one had 4 salt levels (0, 94, 164 and 240 mM NaCl) and 3rd factor had 3 wheat cultivars. Data were analysed with Analysis of Variance (ANOVA) using the GLM procedure of SAS for emergence percentage, shoot height, root length and plant fresh and dry matter. Percentage data were transformed using arcsin prior to statistical analysis. Differences between the means were compared using Duncan's multiple range test ($p<0.05$).

RESULTS AND DISCUSSION

Emergence: The emergence rates of Altin-98 cv. seedlings were approximately 96, 93, 92 and 73% in control treatment at 0, 93, 164 and 240 mM of NaCL levels, respectively (Fig. 1). The emergence rate and level in this genotype was slightly affected by 94 mM NaCL solution. At this salt level, there was an initial delay in the seedling emergence rate, but by day 7, it recovered and was similar to that in the 0 salinity level. However, the highest concentration of NaCL treatments retarted and decreased seedling emergence as compared to the control treatment. In the NaCL with deionized water seed treatment, Altin-98 commercial variety exhibited more of the delay in seedling emergence in 240 mM salt concentration than in any other soil salinity level. The seeds that survived GA_3 soaking treatment substantially stimulated the seed germination in control and salt treatments. GA_3 and KIN decreased inhibitive effect on the initial seedling emergence of the salinity stress to a great extends at the concentrations of 93 mM and 164 and 240 mM NaCl (The delay of emergence decreased with GA_3 seed pre-treatment at all salinity levels in day after day). KIN slightly enhanced the seedling emergence at increasing concentrations of NaCl compared with the GA_3. The rate and total level of emergence of B-22 seedlings decreased in 240 mM NaCL (Fig. 2). On the other hand, 93 and 164 mM NaCl slightly decreased and delayed the initial emergence of wheat seedlings. By day 4, they recovered and were similar to that in o salinity level. GA_3 increased germination and stimulated seed germination at all NaCl concentrations on the rate of all days and recovered seedling emergence. However, GA_3 slightly stimulated emergence in 240 mM

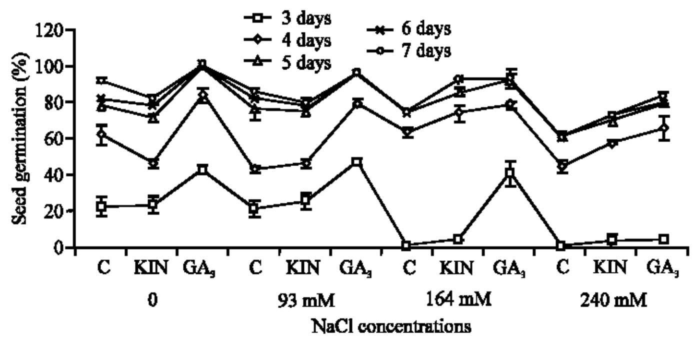

Fig. 1: The effects of PGR and different salt concentrations (mM) on the emergence of Altin-98 seedlings

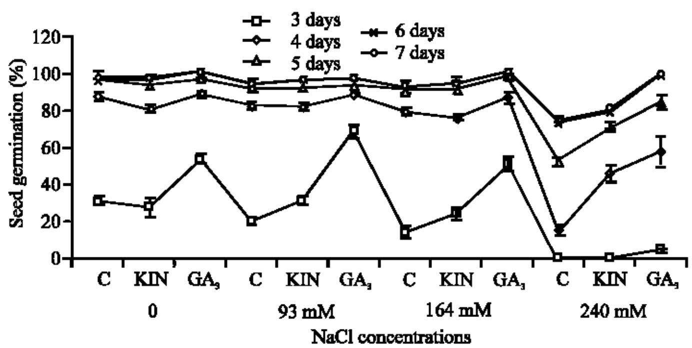

Fig. 2: The effects of PGR and different salt concentrations (mM) on the emergence of B-22 seedlings

salt concentration in 3rd day in comparison with the control and KIN treatments. KIN positively affected the seed germination percentage for all 0, 93 and 164 mM salt solutions except for the 240 mM NaCl concentration on 3rd day of B-22 line. Salt treatments affected the emergence of Kisla domestic variety seedlings more adversely than other genotypes studied (Fig. 3). The most adverse effect of salt stress on the initial seedling emergence on the 3rd day was observed. Then for this genotype of seedling emergence was stimulated by GA_3 in no salt treatment, but this stimulative effect of GA_3 was not observed for the other salinity treatments in the day 3rd (Fig. 1). For all NaCl, concentrations, there were both a delay in the rate and overall level of emergence as compared to no salt and hormone treatment. PGRs pre-treatments, especially GA_3, increased rapidity and final rate of seedling emergence in Kisla domestic variety in comparison with the control in increasing soil salinity levels after the 3rd day.

Increase in NaCl significantly affected the emergence of seedlings and growth of three wheat genotypes studied. The detrimental effect of salt on seedling emergence can be attributed largely to the lower osmotic potential of the soil solution due to the increased concentration of salt in the root environment (Croser *et al*., 2001). Salinity can also affect germination by facilitating the uptake of toxic ions, which can cause changes to certain enzymatic or hormonal activities of the seed. These physico-chemical effects upon the seed seem to result in a slower and/or lower rate of germination (Shannon and Grieve, 1999). The result of our study are in good agreement with those reported by Mer *et al*. (2000) and Yildirim and Güvenç (2006). A reduction and delay in initially seedling emergence in Kisla may be due to the inability of the seed to overcome external osmotic potential and take up water for embryo expansion (Al-Niemi *et al*., 1992). The differences in germination among the wheat genotypes in this study can be explained with genotypic variation. The percentage seedling emergence of wheat genotypes was decreased and delayed considerably by salinity stress conditions. The pre-treatments of hormones not only hastened seedling emergence but also overcame the germination-delaying effect of the high concentrations of NaCl. This finding demonstrates that GA_3 and KIN blocks the inhibitive effect of the salt stress on germination in addition to its stimulative effect on the synthesis of the enzymes creating the mobilization of the reserve material in endosperm (Kabar ve Baltepe, 1987). Kabar (1990) determined the stimulative effect of gibberellic acid on percentage germination and seedling growth in *Hordeum vulgare*, *Triticum aestivum* and *Avena sativa*. Exogenous

Table 1: Effect of salinity and hormone treatments on root length and shoot height (cm seedling $^{-1}$) of wheat cultivars

Cultivars	Salt (EC mmhos cm^{-1})	Root length			Shoot height		
		C	KIN	GA_3	C	KIN	GA_3
	0	14.37b-g*	16.67a-d	15.27a-f	31.50a-f	35.13a-e	41.73ab
Kisla	8	10.17d-h	10.80d-h	19.23ab	29.43b-f	30.13b-f	42.67a
	16	10.57d-h	17.67a-c	21.20a	28.83c-f	31.40a-f	33.80a-f
	24	8.63f-h	14.37b-g	16.07a-e	24.33ef	26.70def	33.30a-f
	0	10.83c-h	14.90c-h	12.40c-h	33.80a-f	33.50a-f	34.20a-e
B-22	8	10.37d-h	11.20c-h	11.00c-h	31.37a-f	35.10a-e	34.17a-e
	16	10.47d-h	11.83c-h	12.90b-h	31.50a-f	33.67a-f	35.47a-e
	24	8.73f-h	10.17d-h	11.67c-h	21.73f	38.47a-d	39.07a-c
	0	10.67d-h	12.47c-h	10.67d-h	25.10ef	32.33a-f	29.93b-f
Altin-98	8	9.07f-h	10.73d-h	10.37d-h	23.47ef	28.63c-f	29.67b-f
	16	7.93gh	10.17d-h	9.37e-h	26.13d-f	24.83e-f	25.73e-f
	24	7.30h	9.80d-h	11.07c-h	25.73e-f	27.27c-f	25.87e-f

*Figures in a column sharing a similar letter are not significantly different from each other at 5% level of probability according to Duncan's Multiple Range Test

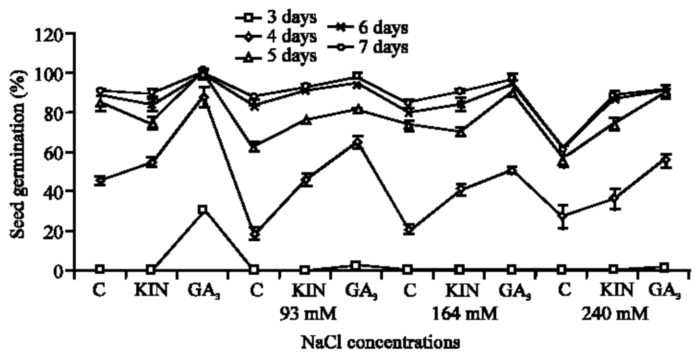

Fig. 3: The effects of PGR and different salt concentrations (mM) on the emergence of Kisla seedlings

kinetin treatments were not so successful as GA_3 in the present study. Gibberellins play a primary role in germination. Cytokinins are secondary agents and are necessary to remove the inhibitive effect of stress (Khan and Downing, 1968). Kabar (1990) indicated that GA_3 application for monocot seeds, at least in the *Gramineae* and kinetin treatment for dicots may be more useful.

Plant growth: The effect of salinity and hormone pre-treatments on shoot height, root length of the three wheat genotypes is presented in Table 1. All these parameters decreased with increased salt stress in the absence of hormone. GA_3 increased significantly increased the root length of Kisla domestic variety in studied salt levels (93, 164 and 240 mM NaCl) in comparison with Altin-98 and B-22 genotypes (Table 1). The highest root length was obtained with Kisla from the GA_3 treatment at 164 mM NaCl level. Similarly, GA_3 had a significant effect on shoot height of Kisla and the greatest shoot height was obtained from this variety at 93 mM NaCl. There was also a significant increase in shoot height in GA_3 and KIN treatments at 240 mM NaCl levels compared to control treatment in B-22 wheat line. The highest fresh weight was obtained from Kisla domestic variety (8.80 g plant^{-1}) at 93 mM of NaCl in GA_3 treatment, while the lowest was Altin-98 genotype with 2.90 g plant-1 at the highest NaCl concentration in KIN treatment. The lowest dry matter yield was determined in Altin-98 variety at the highest concentration of NaCl but the highest dry matter yield was obtained from this variety in GA_3 treatment at 0 salinity level (1.38 g plant^{-1}) (Table 2).

The root and shoot length are the most important parameters for salt stress because roots are in direct contact with soil and absorb water from soil and shoots supply it to the rest of the plant. For this reason, root and shoot length provides an important clue to the response of plants to salt stress (Jamil and Rha, 2004). With increasing salt concentration in the absence of hormones, there was a decrease in root length and shoot height in all wheat genotypes. But this decrease was not significant except in Kisla domestic variety. Salinity has been reported to reduce the height of young plants of *Triticum aestivum*, *Cicer arienitum Brassica juncea* (Mer *et al*., 2000) as well as *Picea mariana*, *Picea glauca*, *Pinus banksiana* (Croser *et al*., 2001). It was observed that seed GA_3 pre-treatment in Kisla domestic variety improved root length compared with the other genotypes in all of the

Table 2: Effect of salinity and hormone treatments on fresh weight and dry matter weight (g plant $^{-1}$) of wheat cultivars

Cultivars	Salt (EC mmhos cm^{-1})	Fresh weight			Dry matter weight		
		C	KIN	GA_3	C	KIN	GA_3
	0	6.43a-j*	6.93a-h	7.36a-f	0.77a-e	0.820a-e	1.38a
Kisla	8	5.66a-k	6.38a-k	8.80a	0.74a-e	0.72b-e	1.32ab
	16	4.77c-k	6.27a-k	7.33a-f	0.60c-e	0.92a-e	0.92a-e
	24	4.35d-k	6.28a-k	5.61a-k	0.51c-e	0.82a-e	0.62c-e
	0	6.98a-g	7.24a-f	7.37a-f	1.05a-d	0.64c-e	0.88a-e
B-22	8	6.75a-j	7.42a-e	8.28ab	0.92a-e	0.76a-e	1.05a-d
	16	6.52a-j	7.42a-e	7.85a-d	1.06a-d	0.97a-d	1.08a-c
	24	6.40a-k	5.34a-k	8.10a-c	0.89a-e	0.83a-e	0.92a-e
	0	5.03b-k	3.43h-k	3.93e-k	0.77a-e	0.53c-e	0.55c-e
Altin-98	8	3.89f-k	3.87f-k	4.78c-k	0.48c-e	0.58c-e	0.62c-e
	16	3.38i-k	2.93jk	2.96jk	0.43c-e	0.45c-e	0.44c-e
	24	3.63g-k	2.90k	4.30e-k	0.31e	0.32e	0.43de

*Figures in a column sharing a similar letter are not significantly different from each other at 5% level of probability according to Duncan's Multiple Range Test

NaCl concentrations. The same effect of GA_3 on shoot height was also observed in Kisla at 93 mM NaCl solution. Kabar (1990) reported that shoot was more salt-sensitive than radicle and that the shoots of the *Gramineae* responded better to GA_3 treatment. There were no significant differences among the salt levels and hormone treatments for fresh weight and dry matter accumulation in all wheat genotypes (Table 2). It was reported that increase in the salt concentration of growth medium caused a significant reduction in mean fresh weights of barley (Kabar and Baltepe, 1987) and sugar beet (Jamil and Rha, 2004). Bagci *et al*. (2007) reported that dry matter production of 16 bread wheat genotypes was negatively affected by high NaCl concentrations, but this effect of salt concentrations changed depending on the genotype.

The results indicate the presence of genetic variation within wheat cultivars for emergence and seedling growth parameters under salt-stress conditions. In tested salinity levels, there was an initial delay in the seedling emergence rate and the highest concentration of NaCL retarted and decreased seedling emergence in control seed pre-treatments. Salinity stress more adversely affected the initial seedling emergence in Kisla domestic variety and seed pre-treatments was not effective in 3rd day. Generally, exogenously applied PGRs was found to overcome inhibition of seedling emergence induced by salt stress in three wheat genotypes.GA_3 not only increased germination percentage of the seeds under high temperature but also shortened the time required for germination except for initial seedling emergence of Kisla. Kinetin was not as successful as GA_3. GA_3 stimulated root length and shoot height of Kisla domestic variety and B-22 line, respectively. Major effect of seed PGRs treatments on seedling growth was due to faster emergence, thus giving seedlings a longer time to develop.

CONCLUSION

GA_3 pre-sowing seed treatments within studied PGRs should be useful applications for the increase in salt tolerance for saline soils in investigated wheat genotypes.

REFERENCES

Amzallag, G.N., H.R. Lener and A. Poljakoff-Mayber, 1990. Exogenous ABA as a modulator of the response of sorghum to high salinity. J. Exp. Bot., 41 (5): 1529-1534. http://jxb.oxfordjournals.org/cgi/content/abstract/41/12/1529.

Al-niemi, T.S., W.F. Campbell and M.D. Rumbaugh, 1992. Response of alfalfa cultivars to salinity during germination and post germination growth. Crop Sci., 32: 979-980.

Ashraf, M., K. Aasiya and A. Khanum, 1997. Relationship between ion accumulation and growth in 2 spring wheat lines differing in salt tolerance at different growth stages. J. Agron. Crop Sci., 178: 39-51.

Bagci, A.S., H. Ekiz and A. Yilmaz, 2007. Salt tolerance of 16 wheat genotypes during seedling growth. Turk. J. Agric. For., 31: 363-372. http://journals.tubitak.gov.tr/agriculture/issues/tar-07-31-6/tar-31-6-2-0708-28.pdf.

Braun, J.W. and A.A. Khan, 1976. Alleviation of salinity and high temperature stress by plant growth regulators permeated into lettuce seeds via acetone. J. Am. Soc. Hort. Sci., 101: 716-721.

Croser, C., S. Renault, J. Franklin and J. Zwiazek, 2001. The effect of salinity on the emergence and seedling growth of *Picea mariana*, *Picea glauca* and *Pinus banksiana*. Environ. Pollut., 115: 9-16. DOI: 10.1016/S0269-7491(01)00097-5. http://www.sciencedirect.com/science?_ob=ArticleURL&_udi=B6VB5-43TP7HN-2&_user=746176&_rdoc=1&_fmt=&_orig=search&_sort=d&view=c&_version=1&_urlVersion=0&_userid=746176&md5=52ec1e9d21c4473782e960eb32e4cdd8.

Foolad, M.R. and G.Y. Lin, 1997. Genetic potential for salt tolerance during germination in *Lycopersicon* species. HortScience, 32: 296-300.

Geschenke, W.D., 1984. K^+-Na^+ Exchange at Cellular Membranes, Intracellular, Compartmentation of Cations and Salt Tolerance. Salinity Tolerance in Plants, Strategies for Crop Improvement. In: Staples, R.C. and G.H. Teonniessen (Eds.). A. Wiley Interscience Publication, John Wiley and Sons, Toronto, Singapore, pp: 37-66.

Jamil, M. and E.S. Rha, 2007. Gibberellic Acid (GA_3) enhance seed water uptake, germination and early seedling growth in sugar beet under salt stress. Pak. J. Biol. Sci., 10 (4): 654-658. http://www.ansijournals.com/pjbs/2007/654-658.pdf.

Jamil, M. and E.S. Rha, 2004. The effect of salinity (NaCL) on the germination and seedling of sugar beet (*Beta vulgaris* L.) and cabbage (*Brassica oleracea* L.). Korean J. Plant Res., 7: 226-232.

Kabar, K., 1990. Comparison of kinetin and gibberellic acid effects on seed germination under saline conditions Phyton (Horn, Austria), 30 (2): 291-298.

Kabar, K. and S. Baltepe, 1987. Alleviation of salinity stress on germination of barley seeds by plant growth regulators. Turk. J. Biol. (Genetics, Microbiologie, Moleculer Biology, Cytology), 11 (3): 108-117.

Khan, A.A. and R.D. Downing, 1968. Cytokinin reversal of dormin Cabscisin II) inhibition of growth and α-amylase synthesis in barley seed. Physiol. Plant., 21: 1301.

Mass, E.V. and G.F. Hoffiman, 1977. Crop salt tolerance-current assessment. J. Irrig. Drinage Div. ASCE 103, pp: 115-134.

Mass, E.V. and J.A. Poss, 1989. Salt sensitivity of cowpea at various growth stages. Irri. Sci., 10: 313-320.

Mer, R.K., P.K. Prajith, D.H. Pandya and A.N. Pandey, 2000. Effect of salts on germination of seeds and growth of young plants of *Hordeum vulgare*, *Triticum aestivum*, *Cicer arietinum* and *Brassica Juncea*. J. Agron. Crop Sci., 185: 209-217. DOI: 10.1111/j.1439-037X.2000.00423. http://www3.interscience.wiley.com/cgi-bin/fulltext/120716651/PDFSTART.

Redmann, R.E., 1974. Osmotic and specific ion effects on the germination of alfalfa. Can. J.Bot., 52: 803-808.

Reynolds, T., 1975. Characterization of osmotic restraints on lettuce fruit germination. Ann. Bot., 39: 791-796.

Richards, L.A., 1954. Origin and nature of saline and alkali soils. In: Diagnosis and Improvement of Saline and Alkali Soils. Agricultural Handbook No. 60, USDA, Washington D.C., USA, pp: 1-6.

Shannon, M.C., 1997. Adaptation of plants to salinity. Adv. Agron., 60: 75-120.

Shannon, M.C. and C.M. Grieve, 1999. Tolerance of vegetables to salinity. Scientia Hort., 78: 5-38.

Singh, K.N., C. Ravish and R. Chatrath, 1997. Combining ability studies in bread wheat (*Triticum aestivum* L. Em Thell) under salt stress environments. Indian J. Gen. Plant Breed., 57: 127-132.

Sivritepe, N., H.O. Sivritepe and A. Eris, 2003. The effects of NaCL priming on salt tolerance in melon seedlings grown under saline conditions. Scientia Hort., 97: 229-237. DOI: 10.1016/S0304-4238(02)00198-X. http://www.sciencedirect.com/science?_ob=ArticleURL&_udi=B6TC3-47CXDG1-8&_user=746176&_rdoc=1&_fmt=&_orig=search&_sort=d&view=c&_version=1&_urlVersion=0&_userid=746176&md5=4adc3af7668da3b24729d88d8e85e00f.

Yildirim, E. and I. Güvenç, 2006. Salt tolerance of pepper cultivars during germination and seedling growth. Turk. J. Agric. For., 30: 347-353. http://journals.tubitak.gov.tr/agriculture/issues/tar-06-30-5/tar-30-5-5-0606-6.pdf.