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Variation in Germination and Ion Uptake in Barley Genotypes under Salinity Conditions

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Abstract: In this paper twelve barley (*Hordeum vulgare* L.) genotypes were screened for salt tolerance during seed germination in the Crop Production Department, Faculty of Agriculture, Jordan University of Science and Technology, Irbid, Jordan. To evaluate salt tolerance during germination, 30 seeds of each genotype were placed on towel paper in 9 cm Petri dishes containing 20 mL distilled water or 1:1 M ratio NaCl and $CaCl_2$ solutions at various concentrations [(control), 100, 200, 300 mM] to give electrical conductivities (EC) of 0.05 (control), 10.6, 19.0 and 27.0 dS m^{-1}, respectively. Our data indicated that salinity level × genotype interaction effects ($p<0.05$) were observed for seed germination percentage, seed viability and ion uptake. Seed germination decreased significantly by increasing salinity level. Germination was significantly diminished at the highest level of salt (300 mM) with significant variation among genotypes and the genotype ACSAD1430 had higher germination percentage than other tested genotypes. Results presented in this article also indicated that the increasing seed pretreatment duration by hyper-saline medium significantly reduced seed recovery when transferred to distilled water. Also our data indicated that the increasing seed pretreatment duration by hyper-saline medium significantly reduced seed recovery when transferred to distilled water. The Na concentration of seeds after imbibitions significantly increased with increasing salinity with a considerable variation among genotypes. K concentration also affected by salinity. Generally, increasing salt stress significantly decreased K concentration in barley seeds after one day of imbibitions. The present study indicated that salt stress must be removed from soil surface for successful seedling establishment.

Key words: Barley · salinity · genotype · Jordan · germination

INTRODUCTION

Soil salinity is a major factor limiting plant productivity, affecting about 95 million hectares worldwide [1]. The UNEP (United Nations Environment Program) estimates that 20% of the agricultural land and 50% of the cropland in the world is salt-stressed [2]. Salinity imposes serious environmental problems that affect grassland cover and the availability of animal feed in arid and semi-arid regions [3]. Greenway and Munns [4] reported that some crops are moderately tolerant of saline conditions; many crops are negatively affected by even low levels of salt. Salt stress unfavorably affected plant growth and productivity during all developmental stages [5]. For example Epstein *et al*., [5] reported that salinity decreases seed germination, retards plant development and reduces crop yield. Seed germination is defined as the emergence of the radical through the seed coat [6]. Othman [7] reported that seed germination can be initiated by water imbibitions and any shortage in water supply will let seed under stress. Shokohifard *et al*., [8] reported that salt stress negatively affected seed germination; either osmotically through reduced water absorption or ionically through the accumulation of Na and Cl causing an imbalance in nutrient uptake and toxicity effect. Further, Younis *et al*., [9] reported that low moisture content under salt stress caused cessation of metabolism or inhibition of certain steps in metabolic sequences of germination. Conversely, salt stress increased the intake of toxic ions which may altered certain enzymatic or hormonal activities of the seeds during germination [10]. The barley is an example of a salt

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tolerant species [11], with genotypes that can germinate in seawater (i.e., about 47.0 dS m^{-1}) [12]. Barley is the most widely grown cereal crop in Jordan and other West Asian countries. The barley-based farming system exists in wide areas along the dry margins (200-300 mm rainfall per year) of cultivation in Syria, Jordan and Iraq [13, 14]. It is grown mainly as feed for livestock. Barley is considered highly salt tolerant of the agriculturally important cereals and has been grown successfully in fields that irrigation has rendered unsuitable for other crops [15]. Generally, barley grown near soil surface where the salts accumulated and at this point of soil, the concentration of salt change over time by continuous evapotranspiaration gradually rising salt levels or rainfall leaching salts from the soil surface supplying water to seeds [16, 17]. Variation in salt levels may restricted seed germination and in some cases resulting in the death of seeds [18, 19]. Germination of barley genotypes under different levels of salt stress and the ability of barley seeds to germinate after an extended period of exposure to salinity is unclear. Thus, the objectives of this study were to screen twelve barley genotypes for salt tolerance during germination, to study the ability of seed to germinate after exposure to different duration times of salt stress and to determine the ionic differences under salt stress.

MATERIALS AND METHODS

Three distinct experiments were conducted in the Crop Production Department, Faculty of Agriculture, Jordan University of Science and Technology, Irbid, Jordan.

Plant material: Twelve barley genotypes (ACSAD176, WI-2291/ ER/Apm, SLB-6, Roho/A.Abiad, ACSAD1430, Esp//808/harmel, Aths/lignee68, JLB70-01, RUM, Mari/Aths/Attiki, ACSAD1212, ER/APM/3/) were used. Seeds of these genotypes were provided by the National Centre for Agricultural and Technology Transfer (NCARTT), Amman, Jordan.

Experiment 1: To evaluate salt tolerance during germination, 30 seeds of each genotype were placed on towel paper in 9 cm Petri dishes containing 20 mL distilled water or 1:1 M ratio NaCl and $CaCl_2$ solutions at various concentrations [(control), 100, 200, 300 mM] to give electrical conductivities (EC) of 0.05 (control), 10.6, 19.0 and 27.0 dS m^{-1}, respectively. Seeds were incubated in the dark at 22±1°C in completely randomized design arrangement with four replicates. Germination counts were made daily. Each petri dish was marked to indicate the solution level that must be maintained over the experimental period. Distilled water was added to each Petri dish as needed to maintain salt concentrations near target levels. Seeds were considered germinated when the radicle was at least 5 mm long. The experiment ended after about 10 days of incubation when no further seeds germinated for three successive days. Germination percentage was calculated using the equation:

Final germination percentage =
number of germinated seeds/ total number of seeds planted × 100

Experiment 2: In this experiment, the effects of salt duration pretreatment on seed viability were evaluated 30 seeds of each genotypes (replicated 4 times) were incubated with 1:1 M of NaCl and $CaCl_2$ solution (EC = 85 dS m^{-1}) for 1, 3, 5 and 7 days at 22°C in the dark. Seeds of each treatment and replicate were rinsed with distilled water then, transferred to Petri dishes containing distilled water. Seeds were counting daily and final germination percentages were determined as mentioned in experiment 1.

Experiment 3: In this experiment, ionic differences were evaluated. Seeds of all barley genotypes were incubated as mentioned in experiment 1. Seeds were soaked in distilled water or in salt solution in 50 mL glass containers. Three replicates of 30 seeds in 25 mL of solution were used for each soaking in solutions. After 24 h of incubation, seeds were removed from the container, dried to a constant dry weight at 80°C, ground to pass a 0.5 mm sieve in laboratory mill. The ground materials were mixed thoroughly and samples of 1.0 g were ashed for 5 h at 550°C in a muffle furnace, then the ash was dissolved in 2 M HC1 for determination of K and Na [20]. K and Na concentrations determined using flame photometer (Ion3).

Statistical analysis: Data were statistically analyzed using analyses of variance (ANOVA) using the MSTATC program (Michigan State Univ., East Lansing, MI and USA). Probabilities of significance among treatments and interaction and LSDs ($p \leq 0.05$) were used to compare means within and among treatments.

RESULTS AND DISCUSSION

Effects of salt stress on seed germination: Analysis of variance revealed that significant differences among the barley genotypes for germination percentage. Salinity level × genotype interaction effects ($p<0.05$) were

Table 1: Germination percentage of barley genotypes as influenced by salinity levels after 10 days of incubation

	Salt level (mM)			
Genotypes	0	100	200	300
Roho/A.Abiad	84.5 ¶	57.9	33.3	3.7
ACSAD1212	82.3	65.0	51.7	26.7
Esp//808/harmel	85.8	64.0	34.2	16.7
SLB-6	84.3	47.4	50.8	25.8
Mari/Aths/Attiki	86.0	44.0	36.7	25.0
Aths/lignee686	81.8	42.5	60.8	26.7
RUM	85.5	64.2	23.3	25.0
ACSAD1430	84.3	86.3	68.3	41.7
ACSAD176	83.3	63.3	50.0	27.5
WI-2291/ER/Apm	83.0	57.9	51.8	24.9
JLB70-01	84.0	84.5	47.5	26.7
ER/APM/3/	84.3	65.8	52.9	25.8
LSD (0.05)		8.4		

Table 2: Germination percentage of barley genotypes as influenced by high level of salinity after 1, 3, 5 and 7 days of incubation

	Duration			
Genotypes	1 day	3 day	5 day	1 week
Roho/A.Abiad	62.3 ¶	21.3	4.25	2.25
ACSAD1212	48.0	50.8	74.50	16.30
Esp//808/harmel	70.3	52.3	35.00	6.50
SLB-6	41.0	30.0	18.00	13.50
Mari/Aths/Attiki	59.5	66.5	34.80	16.50
Aths/lignee686	68.3	61.0	42.00	26.50
RUM	61.0	56.5	30.30	8.50
ACSAD1430	89.8	85.0	60.30	5.50
ACSAD176	76.5	58.3	33.80	23.50
WI-2291/ER/Apm	54.3	42.8	18.30	6.50
JLB70-01	64.5	62.3	49.50	15.80
ER/APM/3/	60.0	41.5	17.50	5.50
LSD (0.05)		2.7		

Table 3: Sodium (Na) concentration (mg g^{-1}) of barley seeds soaked in distilled water and salt solutions

	Salt level (mM)			
Genotypes	0	100	200	300
Roho/A.Abiad	0.9 ¶	1.0	2.3	3.1
ACSAD1212	1.2	2.2	3.4	5.5
Esp//808/harmel	0.9	2.3	3.5	4.4
SLB-6	1.0	2.3	4.4	4.5
Mari/Aths/Attiki	1.0	3.5	3.4	3.8
Aths/lignee686	1.1	2.3	3.5	5.6
RUM	1.1	2.3	3.4	4.6
ACSAD1430	1.0	2.2	3.3	5.4
ACSAD176	1.0	1.5	4.5	6.1
WI-2291/ER/Apm	1.0	2.8	3.6	4.6
JLB70-01	1.2	4.4	4.1	4.3
ER/APM/3/	1.0	2.3	3.8	4.8
LSD (0.05)		0.50		

observed for seed germination (Table1). Interactions show that differences between genotypes depended on the salinity level. Germination percentage of barley genotypes was strongly affected by all salinity levels. Increased salt concentration caused a decrease in final germination percentage. Germination was greatly reduced at the highest level of salt (300 mM). Considerable variation among genotypes in response to salinity was observed for germination percentage. At 100 mM the genotypes ACSAD1430 and JLB70-01 had the highest germination percentage while at 200 and 300 mM salt the genotype ACSAD1430 had higher germination percentage than other tested genotypes. These results were in agreement with Basalah [21] who found that high levels of soil salinity can significantly inhibit seed germination. Further, Waisel [22] found that increasing salinity concentrations in germination often cause osmotic and/or specific toxicity which may reduce or retard germination percentage. Salt induced inhibition of seed germination could be attributed to osmotic stress or to specific ion toxicity [23].

Effect of salt duration pretreatment on seed viability: For this experiment salinity level × genotype interaction effects ($p<0.05$) were also observed for seed germination (Table 2). Interactions show that differences between genotypes depended on the salinity level. This variation possibly due to genetic variability among genotypes [24, 25]. During this experiment, seed pretreatment with salt solution for 1, 3, 5 and 7 days had adverse effect on germination percentage since no seed germination was observed. Xue *et al*., [26] reported that salt stress may affected seed germination through its toxicity effect by increasing Na and Cl concentration or by effects on the concentration and uptake rates of mineral nutrients. Recovery of germination was observed when transferred to distilled water. Increasing duration time of seed pretreatment by salt significantly reduced the germination percentage (Table 2). Based on the order of decreasing salt tolerance for seed germination after salt pretreatment (with their means overall duration pretreatment), the genotype ACSAD1430 had higher germination percentage than other genotypes while Roho/A.Abiad had the lowest germination percentage (Table 2). These results are in agreement with those reported by several researchers [9, 18, 27, 28].

Effects of salt stress on ion uptake: Analysis of variance revealed that significant differences among the barley genotypes for ion uptake. Our results also indicated that ion uptake was strongly affected by all salinity treatments. For example; seed Na concentration significantly increased with increasing salinity level for all studied genotypes (Table 3). At 100 mM, the genotype JLB70-01 had higher Na concentration than other genotypes. At 200 mM, the genotypes ACSAD176 and JLB70-01

Table 4: Potassium (K) concentration (mg g^{-1}) of barley seeds soaked in distilled water and salt solutions

	Salt level (mM)			
Genotypes	0	100	200	300
Roho/A.Abiad	5.1 ¶	4.2	3.3	3.3
ACSAD1212	6.6	3.8	4.0	4.1
Esp//808/harmel	5.9	3.5	4.1	2.7
SLB-6	6.8	4.0	4.9	3.9
Mari/Aths/Attiki	6.1	5.1	4.2	3.6
Aths/lignee686	5.5	4.9	3.4	4.2
RUM	4.7	3.5	4.4	3.6
ACSAD1430	8.0	3.3	5.5	3.1
ACSAD176	5.0	4.2	4.0	2.6
WI-2291/ER/Apm	5.0	3.3	3.4	3.5
JLB70-01	5.5	4.1	3.8	3.9
ER/APM/3/	5.9	4.3	3.8	3.1
LSD (0.05)		0.76		

had the highest Na concentration. On the other hand, at 300 mM the genotype ACSAD176 had higher concentration of Na than other genotypes.

Potassium concentration also significantly affected by salt stress (Table 4). Generally, K concentration decreased by increasing salinity (Table 4). Bhivare and Nimbaker [29] found that the reduction of K content and the increased of Na content in plant could be attributed to the effect of competition between Na and K ions on the absorptive sites of the plant. The reduction in K concentration causes a growth reduction by decrease the capacity of plants for osmotic adjustment and turgor maintenance or by the negative effects on metabolic functions as protein synthesis [4].

At 100 mM the genotype Mari/Aths/Attiki had higher K concentration than other genotypes while at 200 mM the genotype ACSAD1430 had the highest K concentration. In saline environment, high concentration of Na and Cl are usually the most injurious and predominant salts [30]. High level of Na caused a direct damage in plant cell membranes [31]. Haq *et al*., [32] found that Na concentration increased significantly with an increase in salinity from 1.2 to 15 dS m^{-1} and this increase was 13.3 fold as compared to Na in plants grown under nonsaline conditions. Meanwhile Na concentration increased in barley seeds soaked in salt solution, seed germination decreased either by (a) increasing salt level or by (b) increasing duration time. This result support the hypothesis that Na increment inside plants had a toxic effects on seed germination, mainly by affecting the plant water relations or through displacement of Ca by Na from critical cell wall binding sites which could disrupt cell wall synthesis and hence inhibit plant growth [26, 33, 34].

In conclusion our result indicated the results of this study demonstrate that salt tolerance during germination exists within barley genotypes which represent a genetic material for development of salt tolerance of barley.

REFERENCES

1. Szabolcs, I., 1994. Soils and salinization. In: M. Pessarakli (Ed.), Handbook of plant and crop stress. Marcel Dekker, New York, pp: 3-11.
2. Flowers, T.J. and A.R. Yeo, 1995. Breeding for salinity resistance in crop plants. Aust. J. Plant Physiol., 22: 875-884.
3. El-Kharbotly, A., O. Mahgoub, A. Al-Subhi and A. Al-Halhali, 2003. Indigenous grass species with potential for maintaining rangeland and livestock feeding in Oman. Agric. Ecosyst. Environ., 95: 623-627.
4. Green Way, H. and R. Munns, 1998. Mechanisms of salt tolerance in non halophytes. Annual Rev. Plant Phys., 81: 149-190.
5. Epstein, E., J.D. Norlyn, D.W. Rush, R.W. Kinsbury, D.B. Kelly, G.A. Gunningham and A.F. Wrona, 1980. Saline culture of crops: A genetic approach. Sci., 210: 399-404.
6. Copeland, O.L. and M.B. Mcdonald, 1995. Seed science and technology. Third Edn., Chapman and Hall. New York.
7. Othman, Y., 2005. Evaluation of barley cultivars grown in Jordan for salt tolerance. Thesis, Jordan University of Science and Technology, Jordan.
8. Shokohifard, G., K.H. Sakagam. and S. Matsumoto, 1989. Effect of amending materials on growth of radish plant in salinized soil. J. Plant Nutr., 12: 1195-1294.
9. Younis, S.A., H.A. Shahatha, P.G. Hagop and F.I. Al-Rawi., 1991. Effect of salinity on the viability of rice seeds. Plant Growth, Drought and Salinity in the Arab Region, pp: 235-244.
10. Smith, P.T. and B.G. Comb, 1991. Physiological and enzymatic activity of pepper seeds (*Capsicum annuum*) during priming. Physiol. Plant., 82: 71-78.
11. Maas, E.V., 1986. Salt tolerance of plants. Appl. Agric. Res., 1: 12-26.
12. Mano, Y. and K. Takeda, 1995. Varietals variation and effects of some major genes on salt tolerance in barley seedlings. Bull. Res. Inst. Bioresour. Okayama Univ., 3: 71-81.
13. Al-Jamali, A.F., M.A. Turk and A.M. Tawaha, 2002. Effect of Ethephon spraying at three developmental stages of barley planted in arid and semi-arid Mediterranean locations. J. Agron. Crop Sci., 188: 254-259.
14. Tawaha, A.M., M.A. Turk and G.A. Maghaereh, 2002. Response of Barley to herbicide versus mechanical weed control under semi-arid conditions. J. Agron. Crop Sci., 188: 106-112.

15. Hopkins, W.G., 1995. Introduction to plant physiology, Wiley, London.
16. Al-Karaki, G.N., 1997. Barley response to salt stress at varied levels of phosphorus. J. Plant Nutr., 20: 1635-1643.
17. Tobe, K., X. Li and K Omasa, 2000. Seed germination and radicle growth of a halophyte, *Kalidium capsicum* (*Chenopodiaceae*). Ann. Bot., 85: 391-396.
18. Ramoliya, P.J. and A.N. Pandey, 2003. Effect of salinization of soil on emergence, growth and survival of seedlings of *Cordia rothii*. Forest Ecol. Manag., 176: 185-194.
19. Radic, S., M. Prolic, M. Pavlica and B. Pevalek-Kozlina, 2005. Cytogenetic effects of osmotic stress on the root meristem cells of *Centaurea ragusina* L. Eviron. Exper. Bot., In press.
20. Ryan, J., G. Estefan and A. Rashid, 2001. Soil and analysis laboratory manual. ICARDA., pp: 172.
21. Basalah, M.O., 1991. Effect of salinity on seed germination and growth of sequash (*Cucubita pepo* L.). Arab Gulf J. Sci. Res., 9: 87-97.
22. Waisel, Y., 1972. Biology of halophytes. Academic Press, New York and London.
23. Huang, J. and R.E. Redmann, 1995. Salt tolerance of *Hordeum* and *Brassica* species during germination and early seedling growth. Can. J. Plant Sci., 75: 815-819.
24. Al-Karaki, G.N., 2000. Germination of tomato cultivars as influenced by salinity. Crop Res., 19: 225-229.
25. Soltani, A., S. Galeshi, E. Zeinali and N. Latifi, 2002. Germination, seed reserve utilization and seedling growth of chickpea as affected by salinity and seed size. Seed Sci. Technol., 30: 51-60.
26. Xue Z., D. Zhi, G. Xue, H. Zhang, Y. Zhao and G. Xia, 2004. Enhanced salt tolerance of transgenic wheat (*Triticum aestivum* L.) expressing a vacuolar Na^+/H^+ antiporter gene with improved grain yields in saline soils in the fioed an a reduced level of leaf Na^+. Plant Sci., 167: 849-859.
27. Khan, M.A., I.A. Ungar and B. Gul, 1998. Action of compatible osmotica and growth regulators in alleviating the effect of salinity on germination of dimorphic seeds of *Arthorocnemum indicum* L. J. Plant Sci., 159: 313-317.
28. Al-Karaki, G.N., 2001. Germination, sodium and potassium concentration of barley seeds as influenced by salinity. J. Plant Nutr., 24: 511-522.
29. Bhivare, V.N. and J.D. Nimbalkar, 1984. Salt stress effect on growth and nutrition of French beans. Plant Soil, 80: 91-98.
30. Yahya, A., 1998. Salinity effects on growth and uptake and distribution of sodium and some essential mineral nutrient in sesame. J. Plant Nutr., 21: 1439-1459.
31. Leopold, A.C. and R.P. Willing, 1984. Evidence for toxicity effects of salt on membranes. In: Staples, R.C., G.H. Toenniessen (Eds.), Salinity tolerance in plants: strategies for crop improvement. New York, pp: 67-76.
32. Haq, T., K. Mahmood, A. Shahzad and J. Akhtar, 2003. Tolerance potential of wheat cv. lu-26s to high salinity and water logging interaction. J. Agric. Biol., 5: 162-165.
33. Zidan, I., H. Azaizeh and P.M. Neumann, 1991. Does salinity reduce growth of maize root epidermal cells by inhibiting their capacity for cell wall acidification?. Plant Physiol., 93: 7-11.
34. Martinez-Ballesta, M.C., V. Martinez and M. Carvajal, 2004. Osmotic adjustment, water relations and gas exchange in pepper plants grwn under NaCl or KCl. Environ. Exp. Bot., 52: 161-174.