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RESEARCH ARTICLE

ROLE OF SALT STRESS ON SEED GERMINATION AND GROWTH OF SUGAR BEET CULTIVARS

Mahdi zare, Mahdi Ghaemi and Khodadad Mostafavi

[1]Department of Agriculture, Firoozabad Branch, Islamic Azad University, Firoozabad, Iran
[2] Departments of Agronomy, Faculty of Agriculture, Mashhad Branch, Islamic Azad University, Mashhad, Iran
[3] Departments of Agronomy and Plant Breeding, Karaj Branch, Islamic Azad University, Karaj, Iran

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ABSTRACT

In this experiment, five sugar beet genotypes inclusive F-20505, H30938, H30939, SBSI-6 and SBSI-9 were evaluated in five levels of salinity treatment (distilled water as control, 4, 8, 12 and 16 dS/m) by using different NaCl concentrations. The results of this study reveal that various concentrations of NaCl had a significant effect on the all measured traits. The differences between the means (Genotypes and salinity stress levels) were compared by Duncan multiple range test. It observed that, in all of genotypes there was a decrease in germination percentage due to salinity stress increment and maximum germination percentage was delayed. Among the sugar beet genotypes, H30939 had the highest germination percentage. In addition, it was clearly determined that there were no statistical differences between measured genotypes at high salinity levels (16 dS/m) for all investigated traits. Cluster analysis was done using the data for all measured traits at the mid highest salt level (12 dS/m), because this salt level was found very effective in discriminating the genotypes. Results of cluster analysis (Ward's minimum variance method) showed that genotypes H30939 was found to be tolerant, while SBSI-6 and SBSI-9 sensitive to salt.

INTRODUCTION

The two major environmental factors that currently reduce plant productivity are drought and salinity (Serrano et al. 1999). Salinity becomes a concern when an "excessive" amount or concentration of soluble salts occurs in the soil, either naturally or as a result of mismanaged irrigation water. Worldwide, salt-affected soils are most abundant in arid regions, and in irrigated lands the formation of salt-affected soils is the most important process of chemical soil degradation. Salinity in soil or water is one of the major stresses and especially in arid and semi arid regions, can severely limit crop production (Shannon. 1998). Iran is mostly located in arid and semi arid region of the globe (Golbashy *et al.* 2010).

Seed germination is usually the most critical stage in seedling establishment, determining successful crop production (Bhattacharjee, 2008). Crop establishment depend on an interaction between seedbed environment and seed quality (Khajeh-Hosseini *et al.*, 2003). Factors adversely affecting seed germination may include sensitivity to salt tolerance (Ozdener and Kutbay, 2008). sugar beet crop in semi-arid conditions needs to be irrigated. Emergence of sugar beet (Beta vulgaris L.) seedlings is a major factor limiting satisfactory stand establishment. The variability in emergence is caused in part by differences in germination and seedling vigor among seed cultivar, or between cultivars (Habib, 2010). Studies on abiotic stress tolerance in sugar beet have been undertaken for the identification of physiological and environmental factors. Germination percentage of sugar beet measured under standard conditions correlated with the seedling establishment in the field under stress conditions (Durrant and Gummerson 1990).

Nevertheless, breeders assume that there is considerable variability in abiotic stress tolerance in sugar beet germplasm. Hanson and Wyse (1982) evaluated sugar beet, fodder beet and Beta maritime under increasing salinity conditions, and concluded that salinization increased betaine levels of roots and shoots two- to three-fold. The plants that grow in saline soils have diverse ionic compositions and a range in concentrations of dissolved salts (Volkmar *et al.* 1998). Small differences in the concentration of NaCl did not change number of germinated seeds but greatly affected water uptake and seedling growth (Durrant *et al.* 1974).

Salinity impairs seed germination, reduces nodule formation, retards plant development and reduces crop yield (Greenway *et al.* 1980). To solve this problem, more tolerant sugar beet varieties must be selected and recommended for the saline areas. Accurate selection requires an understanding of the mechanisms involved in salt tolerance in this species (Ghoulam *et al.* 2002).

Under salt stress, plants have evolved complex mechanisms allowing for adaptation to osmotic and ionic stress caused by high salinity. These mechanisms includes osmotic adjustment by accumulation of compatible solutes such as glycinebetaine,

* Corresponding author: +91
E-mail address: mostafavi@kiau.ac.ir

proline and polyols (Bohnert *et al.*, 1999) and lowering the toxic concentration of ions in the cytoplasm by restriction of Na+ influx or its sequestration into the vacuole and/or its extrusion (Hajibagheri *et al.*, 1987)

The present study was therefore, conducted with the objectives to determine the response of sugar beet genotype to salinity stress at germination and seedling stages under controlled conditions. Moreover, NaCl was used for salinity stress induction in sugar beet.

MATERIALS AND METHODS

In order to study the effects of salinity stress on germination and early seedling growth in sugar beet genotypes, an experiment was conducted in factorial form, using a completely randomized design with three replications. In this experiment, five sugar beet genotypes inclusive F-20505, H30938, H30939, SBSI-6 and SBSI-9 were evaluated in five levels of salinity treatment (distilled water as control, 4, 8, 12 and 16 dS/m) by using different NaCl concentrations. This experiment was carried out at Biotechnology Laboratory, Islamic Azad University- Karaj Branch, Iran.

The seeds were sterilized by soaking in a 5% solution of hypochlorite sodium for 5 min. After the treatment, the seeds were washed several times with distilled water. 25 seeds were put in each Petridish (with 9cm diameter) on filter paper moistened with respective treatment in 3 replications. The petridishes were covered to prevent the loss of moisture by evaporation. The petridishes were put into an incubator for 12 days at 25 centigrade degrees temperature and 65% relative humidity. Every 24 hours after soaking, germination percentage and other traits were recorded daily. After 12 days of incubation, shoot length, root length, seed vigor and root to shoot ratio of germinated seeds was measured. Seeds were considered germinated when the emergent radical reached 2 mm length. Rate of germination, germination percentage and seed vigor were calculated using the following formulas (Mostafavi 2011):

Formula 1: GP = SNG/SN0 × 100%

where GC is germination percentage, SNG is the number of germinated seeds, and SN0 is the number of experimental seeds with viability (Close and Wilson 2002; Danthu et al. 2003).

Formula 2: GR=∑N/∑(n×g)

Where: GR: Germination race; n: number of germinated seed on gth day and g: Number of total germinated seeds

Formula 3: Seed Vigor = [seedling length (cm) × germination percentage]

Analysis of variance was performed using standard techniques and differences between the means were compared through Duncan multiple range test ($P < 0.05$) using SAS release 9.1 (SAS, 2002) software package. All investigated traits were subjected to hierarchical cluster analysis using procedure Ward's minimum variance method as a clustering algorithm using StatGraphics Plus (Ver 2.1) software. Ward's minimum method is a hierarchical clustering procedure in which similarity used to join clusters is calculated as the sum of squares between the two clusters summed over all variables (Hair et al., 1998). It minimizes them within cluster sums of squares across all partitions.

RESULT

Analysis of variance showed that, there were significantly differences between genotypes for all investigated traits except germination rate. There was no significantly difference between salinity stress levels for germination rate, mean germination time and root to shoot ratio traits.

Also analysis of variance showed that, interaction effects was significant for all investigated characters except germination rate, mean germination time, seedling dry weight and root to shoot ratio. The results of this study reveal that various concentrations of NaCl had a significant effect on the all measured traits (Table1). The control showed clear genetically differences among the genotypes regards germination percentage, and such differences were statistically significant. Germination percentage of all sugar beet genotypes was adversely affected due to the application of different levels (0, 4, 8, 12 and 16 dS/m) of NaCl.

Table 1 Analysis of variance of measured traits of sugar beet genotypes under salinity stress

S.O.V	df	Germination Percentage	Germination Rate	mean germination time	Root Length	Shoot Length
Genotype	4	35.35**	1.79 ns	5.67**	27.07**	25.78**
Stress	4	21.05**	1.64 ns	1.11ns	15.54**	18.63**
Genotype*Stress	16	3.40**	1.53 ns	1.23ns	2.77**	2.63**
Error	50	245.54	1.59	5.64	6.49	11.22

Table 1 continued-

S.O.V	df	Seedling Fresh Weight	Seedling Dry Weight	Seedling Length	Root/Shoot Length
Genotype	4	20.41**	16.35**	29.88**	3.84**
Stress	4	15.41**	5.83**	19.76**	1.28ns
Genotype*Stress	16	1.97*	1.15 ns	2.73**	1.27ns
Error	50	0.00	0.00	29.88	0.13

*, **, ns : significant at 5%, 1% level and not significant, respectively

The differences between the means (Genotypes and salinity stress levels) were compared by Duncan multiple range test and are shown in Table 2. It observed that, in all of genotypes there was a decrease in germination percentage due to salinity stress increment and maximum germination percentage was

delayed. While in this experiment different genotypes had different response to the salinity stress.

Among the sugar beet genotypes, H30939 had the highest germination percentage and germination rate of 67.46% and 17.4 respectively.

Some studies referred that stress can contribute to improve germination rate and seedling emergence in different plant species by increasing the expression of aquaporins (Gao et al., 1999), enhancement of ATPase activity, RNA and acid phosphathase synthesis (Fu et al., 1988), also by increase of

Table 2 Mean comparison of main effects using Duncan multiple range test (at 5% probability level).

	Germination Percentage	Germination Rate	mean germination time	Root Length	Shoot Length
Genotype					
F-20505	47.46b	0.13b	7.74a	8.66a	10.60b
H30938	52.80b	0.21a	4.46bc	6.52b	11.25b
H30939	67.46a	0.17ab	6.05ab	8.64a	13.80a
SBSI-6	11.20c	0.14ab	4.05c	1.65c	3.41c
SBSI-9	17.33c	0.17ab	5.29c	2.16c	5.08c
Salinity stress					
0	62.13a	0.19a	5.23a	8.57a	11.99a
4	48.80b	0.18a	4.60a	6.72ab	10.61a
8	41.06bc	0.14a	5.60a	5.74b	9.80a
12	31.46c	0.17a	5.85a	5.07b	9.35a
16	12.80d	0.12a	6.31a	1.53c	2.40b

	Seedling Fresh Weight	Seedling Dry Weight	Seedling Length	Root/Shoot Length
Genotype				
F-20505	0.09a	0.004a	19.27ab	0.75a
H30938	0.10a	0.003a	17.77b	0.52abc
H30939	0.09a	0.003a	22.44a	0.60ab
SBSI-6	0.02b	0.0008b	5.06c	0.28c
SBSI-9	0.03b	0.001b	7.24c	0.35bc
Salinity stress				
0	0.09a	0.002a	20.56a	0.68a
4	0.09ab	0.002a	17.33ab	0.52a
8	0.07ab	0.003a	15.54b	0.49a
12	0.06b	0.003a	14.42b	0.40a
16	0.02c	0.001b	3.93c	0.43a

Values in a column bearing different superscript are significantly different at 0.05 level.

However, maximum reduction in germination percentage was observed at the highest level i.e., 16 dS/m of NaCl (date not showen). At the highest salt level (16 dS/m), H30939 produced maximum germination percentage and germination rate of all genotypes and they were considered as relatively tolerant.

Results of means comparison using Duncan multiple range test showed that germination percentage and germination rate decreased by increasing in osmotic potential and maximum germination rate and percentage, root length, shoot length, seedling fresh weight, seedling length and root to shoot ratio were obtained at 0 dS/m level (control treatment).

amylases, proteases or lipases activity (Ashraf and Foolad, 2005).

Imposition of varying levels of NaCl significantly reduced all measured traits of all 5 investigated genotypes. Root length is one of the most important characters for salinity stress because roots are in contact with soil and absorb water from soil. For this reason, root length provides an important clue to the response of plants to salinity stress. A marked reduction in root length, shoot length and seedling length of all genotypes of sugar beet was observed due to salt stress. Among the genotypes, the longest root length was commonly determined

Table 3 Supplementary analysis of interaction effects

Salinity Level	Germination Percentage	Germination Rate	mean germination time	Root Length	Shoot Length
0	16.97**	0.54ns	0.57 ns	11.41**	2.96*
4	16.58**	1.47 ns	1.74 ns	11.09**	11.73**
8	9.31**	1.66 ns	1.91 ns	3.87**	8.26**
12	5.23**	2.62*	1.38 ns	10.69**	11.45**
16	0.84ns	1.60 ns	4.99 **	1.08 ns	1.90 ns

Table 3 continued-

Salinity Level	Seedling Fresh Weight	Seedling Dry Weight	Seedling Length	Root/Shoot Length
0	2.57*	2.02 ns	5.99**	2.26 ns
4	11.96**	5.68**	13.25**	1.73 ns
8	6.04**	6.29**	6.93**	0.96 ns
12	6.29**	5.03**	12.87**	1.80 ns
16	1.45 ns	1.92 ns	1.76 ns	2.19 ns

*, **, ns : significant at 5%, 1% level and not significant, respectively

in genotypes F20505 and H30939 while SBSI-9 and SBSI-6 gave the shortest root length. Generally, increasing salinity levels decreased root length, and F-20505 genotype exhibited the greater performance in respect of root length.

Result of this study showed that, shoot length diminished with increasing salinity levels in all genotypes (Table 2). The highest and the lowest seedling length were observed in H30939 and SBSI-6 genotypes, respectively (Table 2). The most effective level in reducing these attributes was 16 dS/m of NaCl (table 3). Best level of NaCl concentration in root length, shoot length, seedling length and seed vigor was 4 dS/m. Seeding dry weight increased with increase in osmotic potential until 8 dS/m but decreased in 12 dS/m (Fig. 1).

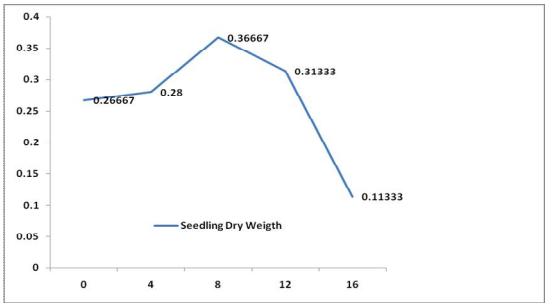

Fig.1 Seedling dry weight of sugar beet genotypes under different salinity stress

A significant inter-genotype variation was observed under salt stress. Of all genotypes, F-20505 and H30939 produced highest mean germination time, root length, seedling dry weight, seedling length and root to shoot ratio at all salt regimes, but lowest seed vigority was recorded in SBSI-6 while the remaining genotypes were moderate in this attribute. Variation in the set of genotypes about germination percentage, mean germination time, root length, shoot length and seedling fresh weigth, was not possible to discern at lower external salt levels, however, genotypes differed significantly at the two higher salt levels i.e. 0 and 16 dS/m of NaCl (Table 3).

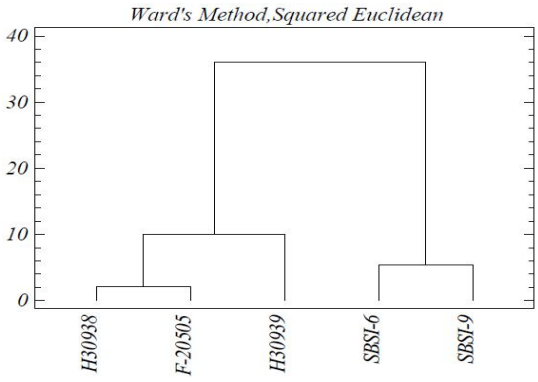

Fig. 2 Cluster analysis of sugar beet genotypes under 12 dS/m level of salinity stress using Ward's minimum variance method

In addition, it was clearly determined that there were no statistical differences between measured genotypes at high salinity levels (16 dS/m) for all investigated traits except mean germination time (Table 3). Cluster analysis was done using the data for all measured traits at the mid highest salt level (12 dS/m), because this salt level was found very effective in discriminating the genotypes. Results of cluster analysis (Ward's minimum variance method) showed that genotypes H30939 was found to be tolerant, while SBSI-6 and SBSI-9 sensitive to salt (Fig. 2).

Ajmal Khan and Weber (2006) found that, resistance to stress at germination stage and primary growth of seedling is independent from next growth stages and evaluation of stress tolerance need more experiment at next growth stages.

DISCUSSION

Screening of available germplasm of a crop is a feasible means of identifying salt tolerant genotypes or genotypes which could maintain a comparatively reasonable yield on salt affected soils (Ashraf & McNeilly, 1987). For the latter crops, it is advisable to assess degree of salt tolerance at each growth stage. In the present study, H30939 genotype was found to be tolerant, while SBSI-6 and SBSI-9 sensitive to salt. Ranking of the genotypes was done using the data for all measured traits at the mid highest salt level (12 dS/m), because this salt level was found very effective in discriminating the genotypes. These results can be related to some earlier studies in which genotypes identified as salt tolerant at the earlier growth stages showed tolerance when tested at the later growth stages.
Although a considerable magnitude of variation for salt tolerance was observed in a set of 5 available genotypes of sugar beet while screening them at germination stages, but a further study needs to be carried out to assess whether the genotypes marked as salt tolerant at the initial growth stages, maintain their degree of salt tolerance when tested as adult.

CONCLUSION

In the present study, salt stress adversely affected the germination percentage, germination rate, shoot length, root length, seedling length, root to shoot length ratio and mean germination time of all 5 genotypes of sugar beet and a significant variation in salt tolerance was observed among all the sugar beet.
Many researchers have been reported similar results (Demir, and Aril, 2003; Mauromicale and Licandro, 2002). Obviously, acceptable growth of plants in arid and semiarid lands which are under exposure of salinity stress is related to ability of seeds for best germination under unfavourable conditions, so necessity of evaluation of salinity resistance genotypes is important at primary growth stage. To find the best tolerant genotype to such conditions, taking all traits into account in this study, we found that H30939 is the most resistant and SBSI-6 and SBSI-9 are the most sensitive genotypes respectively.

References

Ajmal Khan M and Weber DJ .2006. Ecophysiology of high salinity Tolerant plants. Springer the Netherlands, PP 11-30

Ashraf M and Foolad M.R .2005. Pre-sowing seed treatment-a shotgun approach to improve germination growth and crop yield under saline and none-saline conditions, Advan. Agron., 88, 223-271.

Ashraf M and McNeilly T. 1988. Variability in salt tolerance of nine spring wheat cultivars. J. Agron. Crop Sci., 160: 14-21.

Bhattacharjee S .2008. Triadimefon pretreatment protects newly assembled membrane system and causes up-regulation of stress proteins in salinity stressed Amaranthus lividus L. during early germination. J. Environ. Biol., 29, 805-810.

Bohnert HJ, Su H, Shen B .1999. Molecular mechanisms of salinity tolerance. In: Shinozaki, K., Yamaguchi-Shinozaki, K. (Eds.), Molecular Responses to Cold, Drought, Heat and Salt Stress in Higher Plants. University of Arizona, Arizona, pp. 29–60

Close DC, Wilson SJ .2002. Provenance effects on pre-germination treatments for Eucalyptus regnans and E delegatensis seed. Forest Ecol Manag 170:299–305.

Danthu P, Ndongo M, Diaou M, Thiam O, Sarr A, Dedhiou B, Ould Mohamed Vall A. 2003. Impact of bush fire on germination of some West African acacias. Forest Ecol Manag 173:110

Demir M and Aril I .2003. Effects of different soil salinity levels on germination and seedling growth of safflower Turkish J Agric 27: 221-227

Durrant MJ and Gummerson RJ .1990. Factors associated with germination of sugar beet seed in the standard test and establishment in the field. Seed Sci. Technol. 18,561-575.

Durrant MJ, Draycott AP, Payne PA .1974. Some effects of sodium chloride on germination and seedling growth of sugarbeet. Ann. Bot. 38, 1045—1051.

Fu JR, Lu XH, Chen RZ, Zhang BZ, Liu ZS, Li ZS, Cai DY .1988. Osmo conditioning of peanut *Arachis hypogaea* (L.) seeds with PEG to improve vigour and some biochemical activities, Seed Sci Technol 16 197-212

Gao YP, Young L, Bonham-smith P, Gusta LV .1999. Characterization and expression of plasma and tonoplast membrane aquaporins in primed seed of Brassica napus during germination under stress conditions, Plant Mol Biol 40 635-444

Ghoulam C, Foursy A, Fares K .2002. Effects of salt stress on growth, inorganic ions and proline accumulation in relation to osmotic adjustment in five sugar beet cultivars. Environmental and Experimental Botany. 47 : 39–50.

Golbashy M, Khavari Khorasani S, Ebrahimi M, choucan R .2010. Study of response of corn hybrids to limited irrigation. 11th Iranian Crop Science Congress Tehran, 24-26 July 2010. University of Shahid Beheshti. P. 218 (In Persian).

Greenway H and Munns R .1980. Mechanisms of salt tolerance in non-halophytes, Annu. Rev. Plant Physiol. 31: 149–190.

Habib M .2010. Sugarbeet (*Beta vulgaris* L.)_seed pre-treatment with water and HCl to improve germination. African Journal of Biotechnology .9(9): 1338-1342.

Hair FF, Anderson RE, Tatham RL, Black WC .1998. Multivariate data analysis, 5th Ed. Prentice-Hall Inc, Upple Saddle River, NJ, USA pp. 239-325.

Hajibagheri MA, Harvey DMR, Flowers TJ .1987. Quantitative ion distribution within root cells of salt-sensitive and salt-tolerant maize varieties. New Phytol. 105, 367–379.

Hanson AD and Wyse R .1982. Biosynthesis, translocation, and accumulation of betaine in sugar beet and its progenitors in relation to salinity. Plant Physiol. 70, 1191—1198.

Khaje-hosseini M, Powell AA, Bingham IJ .2003. The interaction between salinity stress and seed vigour during germination of soybean seeds, Seed Sci. Technol., 31, 715–725.

Mauromicale G, Licandro P .2002. Salinity and temperature effects on germination, emergence and seedling growth of globe artichoke Agronomic 22: 443-450

Mostafavi K, Sadeghi Geive H, Dadresan M, Zarabi M .2011. Effects of drought stress on germination indices of corn hybrids (Zea mays L.). International Journal of AgriScience. 1(2):10-18.

Ozdener Y and Guray Kutbay H .2008. Effect of salinity and temperature on the germination of Spergularia marina seeds and ameliorating effect of ascorbic and salicylic acids. J. Environ. Biol., 29, 959-964

SAS .2002. SAS/STAT® user's guide, SAS Institute, Cary, NC, USA. 9.1.

Serrano R, Macia FC, Moreno V .1999. Genetic engineering of salt and drought tolerance with yeast regulatory genes, Sci. Hortic.78: 261-269.

Shannon MC .1998. Adaptation of plant to salinity, Adv. Agron. 60: 75 119.

Volkmar KM, Hu Y, Steppuhn H .1998. Physiological responses of plants to salinity: a review, Can. J. Plant Sci. 78: 19-27.

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