http://www.pjbs.org

PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 11 (10): 1390-1393, 2008
ISSN 1028-8880

Growth and Some Physiological Parameters of Four Sugar Beet (*Beta vulgaris* L.) Cultivars as Affected by Salinity

[1,2]R.A. Khavari-Nejad, [1]F. Najafi and [3]S. Khavari-Nejad
[1]Department of Biology, Tarbiat Moallem University, P.O. Box 15815-3587, Tehran, Iran
[2]Department of Biology, Faculty of Science, Islamic Azad University,
Science and Research Unit, Tehran, Iran
[3]Institute of Biochemistry and Biophysics, University of Tehran, Tehran 13145, Iran

Abstract: The comparative responses of certain biochemical and physiological characteristics to salinity were studied in 4 cultivars of sugar beet (*Beta vulgaris* L.) plants. Eight weeks old plants were treated with NaCl at 0, 25 and 50 mM in nutrient solutions. Plants were grown under controlled environment and harvested after 3 weeks for measurements of biochemical and physiological parameters. Results showed that in 25 mM NaCl for cultivars of ET5 and C3-3, soluble sugars in leaves, photosynthetic rate and growth parameters were significantly increased as compared to those of other cultivars. In 50 mM NaCl photosynthetic rate and soluble sugars were significantly increased only in ET5 cultivar as compared with those of others. Results indicated that in 25 mM NaCl, ET5 cultivar showed high growth responses and tolerated to 50 mM NaCl.

Key words: *Beta vulgaris*, salinity, gas exchange, saccharides, growth, sugar beet

INTRODUCTION

Salinity is an important limiting factor for agricultural production by causing two distinct types of stress in plants: hydric stress, caused by the greater difficulty of water absorption and ionic stress, associated to the sodium ion effect on the diverse cellular functions, decreased nutrient absorption, enzyme activities, photosynthesis and metabolism (Zhu, 2001). Salt stress causes inhibition of growth and development, reduction in photosynthesis, respiration and protein synthesis and disturbs nucleic acid metabolism (Levine *et al*., 1990).

Changes in these parameters depend on the severity and duration of stress (Lakshmi *et al*., 1996) and on plant species (Dubey, 1994).

Under salt stress, plants have evolved complex mechanisms allowing for adaptation to osmotic and ionic stress caused by high salinity. These mechanisms include osmotic adjustment by accumulation of compatible solutes such as glycinebetaine, prolines 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; Binzel *et al*., 1988).

The adaptation to salinity stress is accompanied by alternations in the levels of numerous metabolites, proteins and mRNA (Serrano, 1996). Various genes, expression of which is activated in response to salt stress, have been identified (Kawasaki *et al*., 2001). Some of these genes encode for protective proteins such as osmotin (Zhu *et al*., 1995), Late Embryogenesis Abundant (LEA) proteins (Espelund *et al*., 1992) and ion transporters (Blumwald, 2000) others code for enzymes that participate in metabolic processes specifically triggered by salinity stress (Gong *et al*., 2001).

The aim of the present study is to assess four sugar beet varieties for their salt tolerance and to give more information on the significance of soluble sugar accumulation under salt stress, also growth and gas exchanges were studied.

MATERIALS AND METHODS

Sugar beet (*Beta vulgaris* L.) seeds were prepared from Agricultural Research Center, Tehran, Iran in June. Sugar beet seeds were sterilized in 5% (w/v) sodium hypochlorite (15 min) and washed five times with sterile distilled water. Seeds germinated in pots containing sand in a growth chamber under a 24°C temperature and at a relative humidity of 70%. Germinated seeds were translated to pots in growth chamber with 17 h light periods and 300 μmol quanta m^{-2} sec^{-1} light intensity, day/night temperatures of 25/18°C and irrigated with Hoagland`s solution. After 10 days, the seedlings were

Corresponding Author: Farzaneh Najafi, Department of Biology, Tarbiat Moallem University, P.O. Box 15815-3587, Tehran, Iran
Tel: +982188848940 Fax: +982188848940

transplanted in the saline nutrient solutions containing 0, 25 and 50 mM sodium chloride, with pH 6.5 and fresh nutrient solution replaced the old one every week. The plants were grown under controlled environment (17 h light periods, 300 μmol quanta m^{-2} sec^{-1} light intensity, day/night temperatures of 25/18°C) in a greenhouse. After 21 days of experimental duration, for each physiological analysis from each treatment, four plants were harvested. Photosynthetic rate (P_n), respiration rate and CO_2 compensation concentration (Γ) were determined from intact plants, employing an infrared gas (CO_2) analyzer (IRGA) as described by Khavari-Nejad (1980, 1986). Growth analyses were carried out using the equations of Watson (1952) and Evans and Hughes (1962). Saccharides (soluble sugars and starch) were measured using phenol sulfuric acid method according to Hellubust and Craigie (1978).

The research was conducted using completely randomized design with four replications. Data were analyzed statistically using SAS software.

RESULTS AND DISCUSSION

In 25 and 50 mM NaCl, P_n was significantly increased in ET5, but for other cultivars changes were not significant. For the four cultivars, NaCl caused reduction of respiration rate, however, with increasing NaCl, Γ only in ET5 was significantly decreased and in other cultivars changes were not significant (Table 1).

For the two sugar beet cultivars, ET5 and C3-3, 25 mM NaCl caused a significant increase for all growth parameters involved (Table 2). However, in 50 mM NaCl, changes were observed not to be significant as compared to those of controls. Reduction in growth parameters were observed at 25 and 50 mM NaCl for cultivars of 41RT and 19669-T. In 25 mM NaCl treatment, LWCA in ET5, C3-3 and 41RT were significantly increased in comparison with other treatments (Table 2), however, in 50 mM NaCl LWCA only in ET5 was increased.

Soluble sugar also was affected by NaCl treatment, with a greater increase as the NaCl concentration was increased. It was the highest in cultivars ET5 and C3-3. And in cultivars 41RT and 19669-T changes of soluble sugar were not significant as compared with control. For insoluble sugar, it appeared that it was the less affected parameter in comparison with others in all cultivars (Table 3).

Sugar beet is a glycophytic member of the Chenopodiaceae. It is sensitive to elevated salinity at the germination and early seedling phase of development (Durrant *et al.*, 1974; Ghoulam and Fares, 2001). Established plants showed a high osmotic adjustment (Katerji *et al.*, 1997) and accumulation of glycinebetaine, proline and inorganic ions under salt stress (Hanson and Wyse, 1982; Heuer and Plaut, 1989; Gzik, 1996).

In the present study, the presence of 25 mM NaCl in the nutrient solution, increased growth generally in ET5 cultivar. However, in other cultivars, with increasing NaCl, growth either did not significantly changes or decreased. McCue and Hanson (1992) reported that leaf expansion in beet cultivar Great Western D-2 declined steadily as NaCl concentration was raised. Also, the growth of habituated sugar beet Altissima callus was inhibited by

Table 1: Effects of NaCl on photosynthetic rate (P_n), respiration rate and CO_2 compensation concentration (Γ)

Cultivar	NaCl (mM)	Photosynthetic rate (μmol dm^{-2} sec^{-1})	Respiration rate (μmol dm^{-2} sec^{-1})	CO_2 compensation concentration (μl L^{-1})
ET5	0	21.99±1.86bc	33.06±0.92a	114.00±10.2abc
	25	28.90±0.41a	29.26±3.80ab	84.09±2.90e
	50	26.09±0.52ab	29.21±3.60ab	91.77±0.95de
C3-3	0	19.00±0.89dc	29.40±2.18ab	98.17±5.60cde
	25	18.75±1.50dc	18.69±0.73d	115.10±2.40ab
	50	14.80±0.88d	17.30±1.34de	106.50±2.61abcd
41RT	0	27.40±3.17ab	21.86±1.17dc	99.60±4.15bcde
	25	27.67±2.24ab	11.63±1.45e	93.33±3.27de
	50	28.30±2.38ab	11.94±0.79e	91.80±6.79de
19669-T	0	27.20±1.80ab	25.44±3.20bc	116.10±6.70ab
	25	32.16±3.20a	15.89±1.56de	121.50±6.40a
	50	31.61±2.75a	16.21±0.87de	112.20±2.56abc

Means (±SE) of four replications, Numbers followed by the same letter(s) are not significantly different (p>0.05)

Table 2: Effects of NaCl on growth parameters

Cultivar	NaCl (mM)	RGR (g kg^{-1} day^{-1})	RLGR (cm^2 m^{-2} day^{-1})	NAR (gm^{-2} day^{-1})	SLA (m^2 kg^{-1})	LWCA (gm^{-2})
ET5	0	63.22±3.50cd	1007.90±37.0ab	3.89±0.20ab	47.88±2.67a	318.89±10.8de
	25	75.36±3.20ab	1115.20±37.0a	4.30±0.22a	46.20±1.90a	351.38±7.40bcd
	50	63.12±3.20cd	972.92±46.0bc	3.37±0.15bcd	46.35±1.90a	382.98±17.9ab
C3-3	0	59.60±2.12cd	872.40±42.0cde	3.16±0.17cde	46.40±2.08a	352.40±11.3bcd
	25	61.20±1.97cd	907.11±28.1bcd	3.74±0.08abc	44.19±0.83ab	395.62±12.6a
	50	61.68±1.41cd	906.40±37.2bcd	3.55±0.05bcd	41.74±1.75ab	332.30±8.95cde
41RT	0	68.16±3.10bc	737.55±20.9fg	2.99±0.24de	45.04±1.88a	305.62±12.5e
	25	55.38±2.10d	603.80±20.3h	2.55±0.14e	42.89±2.33ab	365.20±15.7abc
	50	62.48±1.81cd	675.98±19.3gh	3.11±0.11de	37.97±1.03b	337.35±14.2cde
19669-T	0	76.20±1.76a	870.59±40.5cde	3.83±0.25ab	44.03±3.74ab	334.35±4.80cde
	25	66.90±4.40c	827.06±68.4def	3.20±0.32cd	44.40±1.22ab	316.25±7.66de
	50	61.30±2.14cd	752.90±56.8efg	3.05±0.13de	40.90±1.16ab	321.33±9.25de

Means (±SE) of four replications, Numbers followed by the same letter(s) are not significantly different (p>0.05)

Table 3: Effects of NaCl on saccharides (soluble sugar and insoluble sugar) concentration

Cultivar	NaCl (mM)	Soluble sugar (mg g^{-1})	Insoluble sugar (mg g^{-1})
ET5	0	8.57±1.15ef	4.90±0.56d
	25	14.29±0.72abc	6.56±0.85dc
	50	16.18±0.68a	8.42±0.68bc
C3-3	0	7.47±0.32f	15.00±1.43a
	25	12.80±2.20bc	4.83±0.89d
	50	15.70±0.68ab	16.00±0.83a
41RT	0	9.54±0.59def	6.80±1.00dc
	25	12.48±0.95cd	4.53±0.5d
	50	8.52±0.31ef	8.28±0.69bc
19669-T	0	11.28±0.65cde	5.36±0.86d
	25	12.64±0.38c	10.84±0.59b
	50	12.45±0.92cd	13.84±1.17a

Means (±SE) of four replications, Numbers followed by the same letter(s) are not significantly different (p>0.05)

NaCl concentrations higher than 30 mM (Hagege *et al*., 1990). Similar results were reported for other species such as *Atriplex prostrate*, where leaf area, dry mass of leaves and roots were significantly reduced by increasing salinity but the number of nodes was not affected by salt treatment (Wang *et al*., 1997).

As shown, the sugar beet cultivars 41RT and 19669-T showed the greatest reductions of growth parameters under salt stress (Table 2). Thus, they could be judged as the less tolerant and ET5 and C3-3 as the more tolerant, cultivars.

Salt treatment induced a reduction in LWCA in 19669-T (Table 2). The decrease in LWCA indicated a less turgor that resulted in limited water availability for cell extension process. LWCA in 25 and 50 mM NaCl in ET5 cultivars, was significantly enhanced.

Under salt stress, the tested cultivars accumulated more soluble sugar in leaves of tolerant cultivars (ET5 and C3-3) than that of sensitive cultivars (41RT and 19669-T). This accumulation of soluble sugars could play an important role in osmotic adjustment, in stressed sugar beet plants.

With increasing NaCl in the root growing medium, photosynthetic rate was significantly increased in ET5 cultivar in comparison with other cultivars. Also in 25 and 50 mM NaCl in ET5, respiration rate and CO_2 compensation concentration were significantly increased. However, changes in the rates of gas exchanges in other cultivars were not significant.

Results obtained in the present research revealed a more accumulation of soluble sugars in the ET5 cultivars treated with NaCl than other cultivars. Accordingly, it may be concluded that high soluble sugars play an important role in turgor maintenance.

Also, with increasing NaCl in solution LWCA was significantly enhanced in ET5 cultivars. As described earlier (Tester and Davenport, 2003) an ability to grow in saline conditions has been attributed to an ability to close stomata. In fact both glycophytes and halophytes tend to show reduced stomatal conductance in high NaCl conditions (Ball, 1988; Robinson *et al*., 1997; James *et al*., 2002).

It is concluded that *Beta vulgaris* cv. ET5 plants are much more tolerant to 50 mM NaCl than other cultivars of *Beta vulgaris.*

CONCLUSION

Although use of ions for osmotic adjustment may be energetically more favorable than biosynthesis of organic osmolyte under osmotic stresses, many plants accumulate organic osmolytes to tolerate osmotic stresses. These osmolytes include proline, betaine, polyols, sugar alcohols and soluble sugars (Chinnusamy *et al*., 2005). In present study has shown sugar beet plants (cv. ET5) have an ability to change the osmotic potential under saline condition and soluble sugars play a main role in the regulation of osmotic potential, also, their Leaf Water Content Area (LWCA) is enhanced in comparison with other cultivars.

ACKNOWLEDGMENTS

This research was funded by a grant from the Tarbiat Moallem University and the seeds were kindly provided from Sugar Beet Seed Institute.

REFERENCES

Ball, M.C., 1988. Salinity tolerance in the mangroves, *Aegiceras corniculatum* and *Avicennia marina.* 1. Water use in relation to growth, carbon partitioning and salt balance. Aust. J. Plant Physiol., 15 (3): 447-464.

Binzel, M.L., F.D. Hess, R.A. Bressan and P.M. Hasegawa, 1988. Intracellular compartmentation of ions in salt adapted tobacco cells. Plant Physiol., 86 (2): 607-614.

Blumwald, E., 2000. Sodium transport and salt tolerance in plants. Curr. Opin. Cell Biol., 12 (4): 431-434.

Bohnert, H.J., H. Su and B. Shen, 1999. Molecular Mechanisms of Salinity Tolerance. In: Molecular Responses to Cold, Drought, Heat and Salt Stress in Higher Plants, Shinozaki, K. and K. Yamaguchi-Shinozaki (Eds.). University of Arizona, Arizona, pp: 29-60.

Chinnusamy, V., A. Jagendorf and J.K. Zhu, 2005. Understanding and improving salt tolerance in plants. Crop Sci., 45 (2): 437-448.

Dubey, R.S., 1994. Protein Synthesis by Plants under Stressful Conditions. In: Handbook of Plant and Crop Stress, Pessaraki, M. (Ed.). Marcel Dekker, New York, pp: 277-299.

Durrant, M.J., A.P. Draycott and P.A. Payne, 1974. Some effects of sodium chloride on germination and seedling growth of sugar beets. Ann. Bot., 38 (5): 1045-1051.

Espelund, M., S. Saeboe-Larssen, D.W. Hughes, G.A. Galav, F. Larsen and K.S. Jakobsen, 1992. Late embryogenesis-abundant genes encoding proteins with different numbers of hydrophilic repeats are regulated differentially by abscisic acid and osmotic stress. Plant J., 2 (2): 241-252.

Evans, G.C. and A.P. Hughes, 1962. Plant growth and the aerial environment. III on the computation of unit leaf rate. New Phytol., 61 (3): 322-327.

Ghoulam, C. and K. Fares, 2001. Effect of salinity on seed germination and early seedling growth of sugar beet (*Beta vulgaris* L.). Seed Sci. Technol., 29 (2): 357-364.

Gong, Z., H. Koiwa, M.A. Cushman, A. Ray, D. Bufford, S. Kore-Eda, T.K. Matsumoto, J. Zhu, J.C. Cushman, R.A. Bressan and P.M. Hasegawa, 2001. Genes that are uniquely stress regulated in Salt Overly Sensitive (SOS) mutants. Plant Physiol., 126 (1): 363-375.

Gzik, A., 1996. Accumulation of proline and pattern of alpha-amino acids in sugar beet plants in response to osmotic, water and salt stress. Environ. Exp. Bot., 36 (1): 29-38.

Hagege, D., C. Kevers, F. Le Dily, T. Gaspar and J. Boucaud, 1990. NaCl dependent growth rate of normal and habituated sugar beet calli, ethylene production and peroxidase activity. C.R. Acad. Sci. Paris, 310 (Ser. 3): 259-264.

Hajibagheri, M.A., D.M.R. Harvey and T.J. Flowers, 1987. Quantitative ion distribution within root cells of salt-sensitive and salt-tolerant maize varieties. New Phytol., 105 (3): 367-379.

Hanson, A.D. and R. Wyse, 1982. Biosynthesis, translocation and accumulation of betaine in sugar beet and its progenitors in relation to salinity. Plant Physiol., 70 (4): 1191-1198.

Hellubust, J.A. and J.S. Craigie, 1978. Handbook of Physiological and Biochemical Methods. Cambridge University Press, Cambridge.

Heuer, B. and Z. Plaut, 1989. Photosynthesis and osmotic adjustment of two sugar beet cultivar grown under saline conditions. J. Exp. Bot., 40 (4): 437-440.

James, R.A., A.R. Rivelli, R. Munns and S. Von Caemmerer, 2002. Factors affecting CO_2 assimilation, leaf injury and growth in salt-stressed durum wheat. Functional Plant Biol., 29 (12): 1393-1403.

Katerji, N., J.W. Van Hoorn, A. Hamdy, M. Mastrorilli and E. Mou Karzel, 1997. Osmotic adjustment of sugar beets in response to soil salinity and its influence on stomatal conductance, growth and yield. Agric. Water Manage., 34 (1): 57-69.

Kawasaki, S., C. Borchert, M. Deyholos, H. Wang, S. Brazille, K. Kawai, D. Galbraith and H.J. Bohnert, 2001. Gene expression profiles during the initial phase of salt stress in rice. Plant Cell, 13 (4): 889-905.

Khavari Nejad, R.A., 1980. Growth of tomato plants in different oxygen concentrations. Photosynthetica, 14 (3): 326-336.

Khavari Nejad, R.A., 1986. Carbon dioxide enrichment preconditioning effects on chlorophylls contents and photosynthetic efficiency in tomato plants. Photosynthetica, 20 (3): 315-317.

Lakshmi, A., S. Ramanjulu, K. Veeranjaneyulu and C. Sudhakar, 1996. Effect of NaCl on photosynthesis parameters in two cultivars of mulberry. Photosynthetica, 32 (2): 285-289.

Levine, R.L, D. Garland, C. Oliver, A. Amici, I. Clement, A.G. Lenz, B.W. Ahn, S. Shaltiel and E.R. Stadtman, 1990. Determination of carbonyl content of oxidatively modified proteins. Methods Enzymol., 1186 (4): 464-478.

McCue, K.F. and A.D. Hanson, 1992. Effects of soil salinity on the expression of betaine aldehyde dehydrogenase in leaves: Investigation of hydraulic, ionic and biochemical signals. Aust. J. Plant Physiol., 19 (4): 555-564.

Robinson, M.F., A.A. Very, D. Sanders and T.A. Mansfield, 1997. How can stomata contribute to salt tolerance? Ann. Bot., 80 (4): 387-393.

Serrano, R., 1996. Salt tolerance in plants and microorganisms: Toxicity targets and defense responses. Int. Rev. Cytol., 165 (1): 1-52.

Tester, M. and R. Davenport, 2003. Na^+ tolerance and Na^+ transport in higher plants. Ann. Bot., 91 (5): 503-527.

Wang, L.W., A.W. Showalter and I.A. Ungar, 1997. Effect of salinity on growth, ion content and cell wall chemistry in *Atriplex prostrata* (Chenopodiaceae). Am. J. Bot., 84 (9): 1247-1255.

Watson, D.J., 1952. The physiological basis of variation in yield. Adv. Agron., 4 (1): 101-145.

Zhu, B., T.H. Chen and P.H. Li, 1995. Expression of three osmotin- like protein genes in response to osmotic stress and fungal infection in potato. Plant Mol. Biol., 28 (1): 17-26.

Zhu, J.K., 2001. Plant salt tolerance. Trends Plant Sci., 6 (2): 66-71.