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Effect of proline, soluble carbohydrates and water potential on resistance to salinity of three *Salsola* species (*S.rigida, S. dendroides, S.richteri)*

A. Teimouri[a*], M. Jafari[b], H. Azarnivand[c]

[a] *Senior Expert in Desert Management, Faculty of Natural Resources, University of Tehran, Karaj, Iran*
[b] *Professor, Faculty of Natural Resources, University of Tehran, Karaj, Iran*
[c] *Associate Professor, Faculty of Natural Resources, University of Tehran, Karaj, Iran*

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Abstract

Various resistances to salinity are due to plants genetically variations, and selection on the basis of one factor will not be a suitable criterion in measurement of resistance to salinity, therefore in choosing resistant varieties, it is necessary to notice to a set of resistant indexes to salinity, creating changes in physiology, anatomy and morphology of plant species. This research was conducted using a factorial experiment based on CRD design with 4 replication at research institute of forests and rangelands. Experimental treatments included a combination of three species (*S.rigida, S.dendroides* and *S.richteri*) along with 7 salinity levels of 0, 100, 200, 300, 400, 500, and 600 mM concentrations. The purpose of this research was to investigate metabolites, accumulation in terms of compatibility or incompatibility value for salinity stress in three species. The results of physiology features measurement demonstrated that *S.dendroides* water potential is more than two others. Comparing of the measured characteristics, averages demonstrated that *S.rigida* and *S.dendroides* have reposed in one statistical group in terms of proline concentration and significant different have with *S.richteri*. Also, increasing salinity levels leads to the highest proline concentration in *S.richteri* in salinity treatment of 400 mM. In all three species, it is seen that salinity increase reduces plants soluble carbohydrates concentration rate and it reveals that salinity have a negative effect on these materials.

Keywords: salinity stress; Praline; Soluble carbohydrates; Water potential; sodium chloride

1. Introduction

Soil salinity is one of the great concerns in arid and semi-arid regions in the world. According to the studies %7 of the world’s lands is saline and %3 is high saline, because of low precipitation, high evaporation and Irrigation by saline waters, soil salinity is getting increased (Alavipanah, 1992). According to of FAO reports, more than %40 of Iran agriculture lands is against secondary salinity (Pessarakli, 1993).

Nowadays biological method is used to contrast and solve soil salinity problem. But significant success will be found when plant reserve resources having desired genetically changes are available. Because various resistances to salinity are due to plants genetically variations and selection on the basis of one factor will not be a suitable criterion in measurement of resistance to salinity (Jones and Qualset, 1984), therefore in choosing resistant varieties, it is necessary to notice to a set of resistant indexes to salinity, creating changes in physiology, anatomy and morphology of plant species. By using the selection and plant breeding method of saline resistance plants, we would be able to increase production level, in arid and semi-arid regions such as Iran. So, in order to regeneration of arid and semi-arid ranges, planting of salt-tolerant species is

* Corresponding author. Tel.: +98 261 2249313;
Fax: +98 21 2249313.
E-mail address: Teimouri_a@yahoo.com

needed, to tolerate unfavorable environment. One of these species, are halophytes that have more tolerable potential against the worse condition and are dispread in saline lands and deserts (Hasheminia *et al*., 1997). Halophytes are able to grow and complete life cycle in saline soils. The main use of halophytes is in animal feeds, otherwise, they are important in protection of sensitive regions' soil and water, from erosion and evaporation, respectively (Tamartash, 2001).

Lutts (1996) in studying the effects of salinity stress on the aggregation of proline in 5 varieties of rice: sensitive, semisuffering and suffering, concluded that salinity causes considerable rise in root proline content of sensitive varieties to salt, but there was no rise in root proline content of suffering varieties. After one week stress in every amount of NaCl, proline aggregated in the stem of sensitive varieties to salt but in suffering types, it aggregated just in the highest concentration of salt (40-50mM). After 2 week of tension, proline levels have been increased in all varieties but it was lesser in suffering types to salt. Regarding to quantity, exact action of proline is inconsiderable in osmotic regulation. Even if we consider that proline aggregates in cytoplasm is stead of vacuole, with this supposal that cytoplasm occupy %10 of cell volume, portion of proline in osmotic potential is variable between %1.5-%4.8 (Lutts *et al*., 1996). Khodary (1992) reported that proline and carbohydrate content increase in wheat and sorghum plant lets by increase of NaCl while Amino acids' content decreases (Khodary, 1992). Tombesi (1986) studied the effect of drought tension on the photosynthesis, stomata's status and carbohydrates' amount in olive plants. They observed that leaf dissolved saccharin amount increases by decreasing the water of soil but leaf amylum amount decreases in a liner manner (Tombesi *et al*., 1986).

Krishchenko (1984) studied on 27 species of 8 chenopodiaceae family genus as *salsola* regarding to food value in Libya. Results proved that in most of the species, protein and saccharin content decreases by growth of plant and wood content (cellulose) increase. In most of them there is few amount of Ether and great amounts of insoluble or little soluble proteins. It was observed that there is Alkaloid in 5 species and glycoside in 14 species and saponin in 15 species. During plant growth, Amino acid amount increased considerably (Krishchenko *et al*., 1984).

Peterson (1992) studied the effect of water potential amount on the function and quality of Alfalfa and declared that stem water potential decrease from -1.3 Mpa to -3.8 Mpa, reduces the function until %33 and increases leaf proportion to stem from %1.7 to %5.1 (Peterson *et al*., 1992).

Liu (1993) reported a meaningful cohesion, between endurance to salinity and RWC (Relative Water Cells Percentage) amount in some of forage grasses (Liu *et al*., 1993). The purpose of this research was to investigate metabolites accumulation in terms of compatibility or incompatibility value for salinity stress in three species.

2. Materials and methods

This study was carried out to effect of salinity stress due to sodium chloride on physiological characters three *salsola* species. This research was conducted using a factorial experiment based on CRD design with 4 replication at research institute of forests and rangelands. Experimental treatments included a combination of three species (*S.rigida, S.dendroides* and *S.richteri*) along with 7 salinity levels of 0, 100, 200, 300, 400, 500, and 600 mM concentrations (Valizade and Moghaddam, 1996). Experiment was performed as green-house cultivation and in plastic flowerpots with 15 ^{cm} mouthpiece diagonal and 20 ^{cm} height. Quartz was used to create homogeneous conditions of soil bed for plants. The action of quartz disinfection was done by oven in temperature 72$^{°c}$ for 8 hours. All flowerpots and grids were completely disinfected before begging cultivation of plants' seeds. Then 15 seeds were put inside flowerpots. It's remarkable that in plant depth of seeds was considered about 1 ^{cm}. After germination the seeds, irrigation of plantlets was done for 7 days with common water and after 7 ^{the} day, they were irrigation with Hoagland solution. After shrubs' confirmation, the act of thinning was done, 30 days after implanting the seeds. So that 3-4 shrubs was remained in each flowerpot. Different salinity cares was applied about 60 days later until the effects of salinity on the plant was clearly shown. Proline was estimated by the method described by Bates *et al.* (1973) and soluble carbohydrates were estimated by the method described by Irrigoyen *et al.* (1992). Plant water status was evaluated by shoot xylem pressure potential measured with a pressure bomb. Experimental traits were proline, soluble carbohydrates and water potential, that normality tested by Minitab software, then data statistically analyzed by SAS and MSTATC software's. Finally results were showed in tables and diagrams, mode by applying Excel program (Farkhah, 2001).

3. Results

Analysis of variance (ANOVA) indicated that plant species significantly influenced the proline content (P<0.01), and salinity level had significant influence on proline content at P<0.001 (Table 1). Interaction effect of salinity* species was significant on proline content. Comparing of the measured characteristics, averages demonstrated that *S.rigida* and *S.dendroides* have reposed in one statistical group in terms of proline concentration and significant different have with *S.richteri* (Table 2). Also, increasing salinity levels leads to the highest proline concentration in *S.richteri* in salinity treatment of 400 mM (Fig. 1).

Results of analysis of variance indicated that plant specie significantly influenced the soluble carbohydrates content (P<0.01), and salinity level had significant influence on soluble carbohydrates content at P<0.001 (Table1). Interaction effect of salinity* species was significant on soluble carbohydrates content at P<0.001 (Table 1). Comparison of means by measurement characters of plant indicate that *S.rigida* and *S.richteri* have reposed in one statistical group in terms of amount soluble carbohydrates concentration and different significant with *S.dendroides* (Table 2). In all three species, it is seen that salinity increase reduces plants soluble carbohydrates concentration (Fig. 2).

Results of analysis of variance showed that salinity have had significant different negative influence on water potential of plant (Table 1). Salinity cause decrease of water potential, among influence was observed different species, significant at P<0.001. But there was no significant different in plant species *salinity. Figure (3) showed that increasing the salinity, decreased water potential of plants and became more negative with an increase in salinity. Among water potential *S.dendroides* had the lowest water potential.

Table 1. Results of ANOVA characteristics by water potential, proline and soluble carbohydrates treatments in three salsola species

		MS(Mean Square)		
S.O.V (Sources Of Variance)	df (Degree of Freedom)	Water Potential	Proline	Soluble Carbohydrate
Plant	2	269.6793**	0.00335989**	3.5751**
Salinity	6	627.0510**	0.04127815**	41.03288**
Plant*Salinity	12	185.1987ns	0.00364146**	3.46072**
Error Experimental	65	4.9925	0.000018	0.00004996
Variation Coefficient Percentage	-	18.97	1.32	0.2219

*, ** and ns in order significant different at level 1% and 5% and non significant

Table 2. Comparison of means measurement attributes in three salsola species

Character	Water Potential (-bar)	Proline (μmol/gFw)	Soluble Carbohydrate (mg/gdw)
Species			
S.rigida	12.61c	0.275 b	0.121 b
S. dendroides	16.71a	0.292 b	0.159 a
S.richteri	15.13 b	0.385 a	0.114 b

Values in each column having the same letter are not significantly different.
Comparison of means was classified basis Duncan multiple-range test at level of probability 5%.

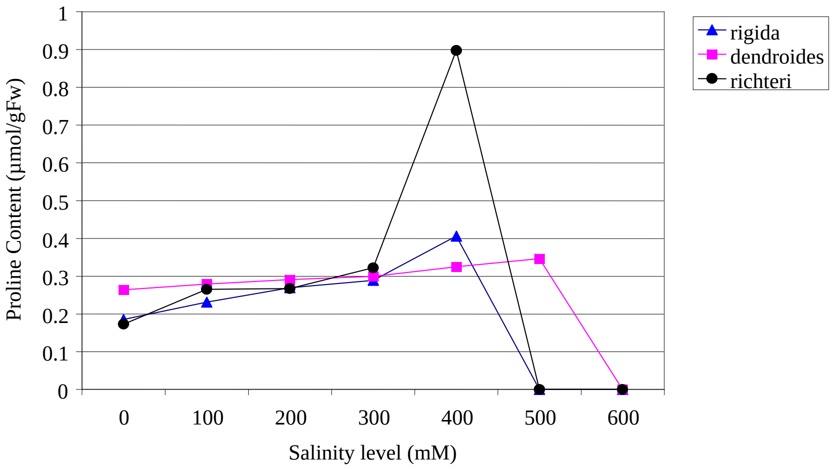

Fig.1. Effects of NaCl on proline content changes in three *Salsola* species

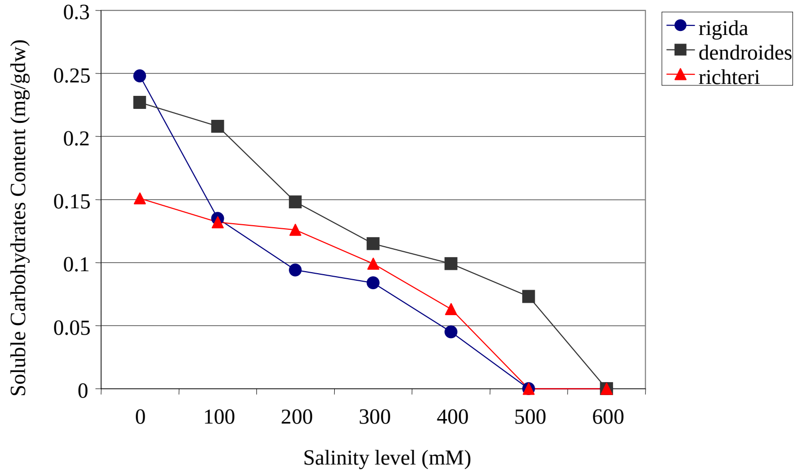

Fig.2. Effects of NaCl on soluble carbohydrates content changes in three *Salsola* species

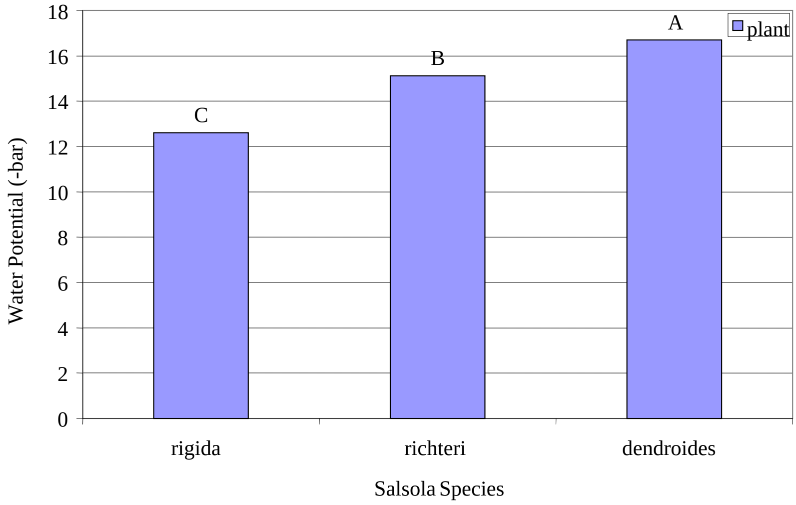

Fig.3. Comparison of means water potential in three *Salsola* species

4. Discussion and conclusions

Soil salinity puts the plant under effect of physiologic drought tension because of increasing the dissolved osmotic potential of soil. This means that even when plant is in a condition with enough moisture, it absorbes water easily and this is caused by increase of dissolved osmotic potential of soil (more negative of osmotic potential) and decrease of root hydraulic ability in water transferring. Decrease of steam accumulation in under room of stomata cells is one of the causes in perspiration decrease that is performed because of water potential decrease (Dianatnagad and Behfar, 1987). Water control in salinity conditions is considered as a part of insistence process, because water and salts' content altogether specify the turgor pressure amount (Glenn *et al.*, 1997). In every 3 plants of under experiment plants, osmotic pressure increased by increasing the entrance of ions. Increase of Na^+ and Cl^- after that in plants: *S. dendroides* and *S. richteri* can have an important rule in osmotic regulation, certainly it's likely that other metabolites are also used in order for osmotic regulatory which aren't mentioned in this research. Measured characters' averages' comparison cleared that 2 species of *S. dendroides* and *S.richteri* are put in a statistic group regarding to their proline concentration amount and there is a meaningful different from *S.richteri*. According to diagram (1), it's obvious that a considerable addition happens in proline concentration amount in *S.richteri* specie during salinity care 400 mm and the result can show that plant *S.richteri* probably uses proline in order for opposing salinity tension. Repartitioning to diagram (1), it's observable that changes of proline concentration amount in plant *S.rigida* have an almost same procedure in different salinity cares and the highest proline concentration amount is in salinity care 400 mM that has a meaningful difference from other salinity cares. Morgan in 1984 after studying the effects of salinity tension in excellent plants, reported that in plants which were under salinity tension, naturally adjusting metabolites are gathered for turgor preservation and fixity of enzymes' active form (Morgan, 1984). Chaparzade in 1996 during studying the effects of salinity tension and Ca^{2+} an alfalfa plant showed that salinity causes proline increase in root and aerial organs and protein and amylum of aerial units were increased by salinity increase, but inversely dissolved carbohydrates amount was decreased(Chaparzade, 1996). Other metabolites which were attended in this study are dissolved carbohydrates that in all 3 under experiment plants its concentration amount was decreased by salinity increase. According to diagram2, changes rate of carbohydrate in *S.dendroides* is less than 2 other species, this plant probably uses carbohydrate as an osmotic regulator element. Decrease of dissolved carbohydrates may be caused by power decrease of transferring through phloem vasculum or increase of saccharin usage in aerial organs in order to exit Na^+ from cell (Irrigoyen *et al*, 1992). Farbudnia in 1995 through applying water tension noticed that dissolved carbohydrates amount in root decreases because of root activity decrease, carbohydrates metabolism decrease and transferring decrease in libber (Farbudnia, 1995). Plant water potential almost follows water absorption rate by root and its perspiration rate by leaf (Levitt, 1980). Results of plant water potential measurement show that plant water potential has negative thought salinity tension. Outcome results of this research show that plant water potential decreases meaningfully by increase of salinity tension. Plant water potential measurement showed that a meaningful different was observed in possibility level of %1 among different forage species which were under experiment, remarking this point that it can be used as a norm for recognizing the suffering species to salinity. Among under experiment species, plant *S.dendroides* has the lowest water potential. Plants usually cause more concentration incline resulting water absorption from soil near root by negative of water potential. Leaf water potential decrease is an appropriate reaction for championing with salinity tension. Outcome results of research agree with levitt research (Levitt, 1980).

Recomendation

Regarding to salinity importance in Iran and also existence of different forage species in our country and lack of their physiologic features' recognition, it's offered that drought and salinity tension effects should be studied by NaCl, CaC_{l2} and Na_2SO4 salts combination and compared in 2 steps: pullulating and germination growth on morphological, physiological and anatomical features by measuring the important norms in salinity insistence specification of considered species.

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