

ECO-MORPHIC VARIATION FOR SALT TOLERANCE IN SOME GRASSES FROM CHOLISTAN DESERT, PAKISTAN

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Abstract

A study was conducted to evaluate the effect of salt stress on some ecologically different populations of three grasses viz., *Cymbopogon jwarancusa* (Jones) Schult, *Lasiurus scindicus* Henr., and *Ochthochloa compressa* (Forssk.) Hilu] from the Cholistan desert. The populations of all three grasses from the highly salt-affected habitats were relatively more salt tolerant. Of the three grasses, *Ochthochloa compressa* was the most tolerant as it was least affected due to salt stress in terms of different growth attributes measured in the present study. *Cymbopogon jwarancusa* was moderately salt tolerant, whereas *Lasiurus scindicus* was salt sensitive.

Introduction

Salt stress is one of the major stress factors which considerably reduce growth of most plants (Rausch *et al.*, 1996; Ashraf, 2004). Yields are steadily affected at low salt concentrations (Maggio *et al.*, 2001) and decline to zero as salt concentration increases. Under natural conditions, most plants are exposed to high salinity (Allen *et al.*, 1994) and they show a wide spectrum of salinity tolerance (Robinson *et al.*, 1997). Among them halophytes are known to be best adapted for saline environments (Winicov & Bastola, 1997).

The salinity tolerance of plants can be best evaluated in terms of growth or survival as it depicts many physiological mechanisms occurring within the plants (Niknam & McComb, 2000). Morpho-agronomic characters reflect the combined genetic and environmental impacts on plants, and parameters like survival under unfavourable conditions, plant height, leaf area, injury to salt stress, relative growth rate, and relative growth reduction are considered as selection criteria for salt tolerance (Ashraf & Harris, 2004).

So the ability of the plants to continue to grow would indicate a high level of salt tolerance (Winicov, 1998). Salt tolerant plants can minimize the detrimental effects of high salinities by producing a series of anatomical, morphological and physiological adaptations (Poljakoff- Mayber, 1988), such as an extensive root system (Hameed & Ashraf, 2008), reduction in growth in terms of leaf area (Monteverdi *et al.*, 2008) and salt secreting glands, and hairs on the leaf surface (Marcum *et al.*, 1998, Naz *et al.*, 2009).

The plants inhabiting desert environments face extreme adverse conditions that are extremely detrimental to plant growth and development (Arshad *et al.*, 2008). Highly saline patches in the hot and arid Cholistan Desert is an important characteristic of this desert, and naturally adapted plants species to such stressful conditions can provide excellent material for investigating the adaptation mechanisms in relation to their growth and survival to counteract high salinities (Ashraf, 2003).

The present studies were focused on the morphological adaptations of three grass species from saline and arid environments of the Cholistan Desert under salt stress.

Cymbopogon jwarancusa was a tussock forming perennial grass with moderate to high tolerance against salt stress (Ashraf *et al.*, 2006). *Lasiurus scindicus* is a highly nutritive grass and has been found to be a moderately tolerant to salt stress (Yadava *et al.*, 2004; Ashraf *et al.*, 2006). *Ochthochloa compressa* is a mat-forming rhizomatous perennial grass with relatively high tolerance against salt stress (Naz *et al.*, 2009).

Materials and Methods

An experiment to study some of morphological features of three potential salt tolerant grasses inhabiting saline patches of the Cholistan desert was carried out at the Botanical Garden, Department of Botany, University of Agriculture, Faisalabad during the year 2007-9.

Three salt tolerant forage grasses *Cymbopogon jwarancusa*, *Lasiurus scindicus* and *Ochthochloa compressa* were collected from three different salt affected habitats in the Cholistan Desert. Derawar Fort (DF) was the least salt-affected habitat (coordinates 29° 24' 31.95" N, 71° 27' 32.83" E, pH 8.35; ECe 15.21 dS m⁻¹, SAR 2049.27, Na⁺ 3236.56 mg L⁻¹, Cl⁻ 1493.11 mg L⁻¹). This area was considered as a control for comparing the response of the grasses to salt stress because all three selected grasses in the present study are endemic to the Cholistan area. Bailahwala Dahar (BD) is moderately salt-affected site (coordinates 29° 38' 18.17" N, 70° 93' 23.38" E, pH 8.42; ECe 26.7 dS m⁻¹, SAR 2419.37; Na⁺ 4392.35 mg L⁻¹, Cl⁻ 2456.52 mg L⁻¹). Ladam Sir (LS) was highly salt-affected site (coordinates 30° 53' 26.47" N, 72° 64' 25.08" E, pH 8.38; ECe 49.18 dS m⁻¹, SAR 2795.57, Na⁺ 5139.30 mg L⁻¹, Cl⁻ 2637.73 mg L⁻¹).

Soil samples of the rhizosphere of the grass species from each habitat were taken at 15 to 24 cm depth, for the determination of physico-chemical characteristics and 200g of dried soil were used for the preparation of saturation paste. Soil ECe and pH were determined from soil extract using a pH / electrical conductivity meter (WTW series InoLab pH/Cond 720). Soil Na⁺ was determined with a flame photometer (Model Sherwood 410, USA) and Cl⁻ with a chloride meter (Model 926, Sherwood Scientific Ltd., Cambridge). Sodium adsorption ratio (SAR) was calculated as described in the USDA Handbook 60 US Salinity Laboratory Staff (Anon., 1954).

Populations of all three grasses from the three habitats were grown in a normal non-saline soil (loamy-clay) for a period of one year in the Botanic Garden research area, University of Agriculture, Faisalabad. The plants were irrigated regularly when required until their establishment in the Faisalabad environment. Ramets, each with two mature tillers of uniform size, were detached from each plant and grown in half-strength Hoagland's nutrient solution (Hoagland & Arnon, 1950) for 8 weeks in hydroponics. The system was aerated with the help of air pumps about 12 h daily. Ten plants of each grass were used from each population and four replications planted on thermopore sheets. The experiment was organized in a 3-factor CRD (completely randomized design) with three study sites (DF, BD, and LS), three grasses (*Cymbopogon jwarancusa*, *Lasiurus scindicus* and *Ochthochloa compressa*), and four salinity levels (0, 100, 200 and 300 mM of NaCl) in solution culture. Plants were carefully removed from the hydroponics after 60 days of the start of salt treatment for examining morpho-agronomic characteristics.

Results

Shoot length of all populations generally decreased with increase in salt level of the growth medium. The effect of salt stress was more prominent in the populations from

Derawar Fort (DF) of all three grasses. However, *Lasiurus scindicus* was the most adversely affected among grasses in relation to shoot length (Fig. 1). In *Ochthochloa compressa*, shoot length was promoted by 100 mM NaCl level in the populations from Derawar Fort (DF) and Bailahwala Dahar (BD), but at higher salt levels this parameter was decreased consistently. The population of this grass from Ladam Sir (LS) was not affected up to 200 mM NaCl, but the highest salt level (300 mM NaCl) caused a slight reduction in shoot length.

All the three grasses showed quite variable response in terms of root length with increasing salinity levels of the growth medium. This character was adversely affected in all the populations of *L. scindicus*, but root length in the population from LS was least affected due to increase in salt level. *Cymbopogon jwarancusa* showed almost similar trend as was recorded in *L. scindicus*, but the LS population showed increase in root length with increase in salt level of the growth medium. In *Ochthochloa compressa* populations from DF and BD, root length was little affected by increasing salt levels. The LS population, in contrast, showed a significant increase in its root length with increase in salt level of the rooting medium.

The response of all the three grasses was quite similar in terms of number of leaves per plant under salt stress as it generally decreased with increase in external salt level (Fig. 1). A progressive decrease was recorded in the number of leaves per plant in the DF populations of *L. scindicus* and *O. compressa*, while this character showed a slight increase in BD and LS populations of these grasses at 100 mM NaCl only. In *C. jwarancusa*, on the other hand, number of leaves was promoted by 100 mM NaCl level in all three populations (Fig. 1), but the LS population showed stability at higher salt levels.

Total leaf area per plant was adversely affected by increasing salt levels in the DF and BD populations of *C. jwarancusa* and *L. scindicus*. However, the LS populations of both these grasses were relatively less affected due to salt stress as compared to the other populations. In *O. compressa*, leaf area in DF population was constantly decreased with increasing salt levels, whereas in BD and LS populations this characteristic showed increase at 100 mM NaCl and thereafter a slight decrease at higher salt levels (Fig. 1).

Fresh weight of shoot was one of the most adversely affected characters by the increasing salt level (Fig. 2). All the populations of *L. scindicus* and *C. jwarancusa* showed a substantial reduction in shoot fresh weight with increase in salt levels compared to control. However, shoot fresh weight in all the populations of *O. compressa* was relatively stable at 100 mM NaCl, but thereafter it gradually decreased with a further increase in external salt level.

Shoot dry weight was adversely affected in the DF populations of all three grasses with increase in salt level (Fig. 2). In *L. scindicus*, this character was increased at 100 mM NaCl and then decreased gradually at higher salt levels. In LS population of *C. jwarancusa* shoot dry weight was increased up to 200 mM NaCl and then it decreased slightly at the highest salt level. In *O. compressa*, this character was promoted by 100 mM NaCl level and then slightly decreased at the higher levels.

Fresh weight of root showed quite a variable response in all three grasses under salt stress. In *C. jwarancusa*, root fresh weight decreased gradually in all three populations with increase in salt level of the external growth medium. In *L. scindicus*, root fresh weight was decreased in the DF population with increase in salt level. In contrast, this parameter in the BD and LS populations was increased at 100 mM NaCl level. In *O. compressa*, the populations from DF and BD showed decrease in root fresh weight, but the LS population showed a consistent increase in root weight with increase in salt level (Fig. 2).

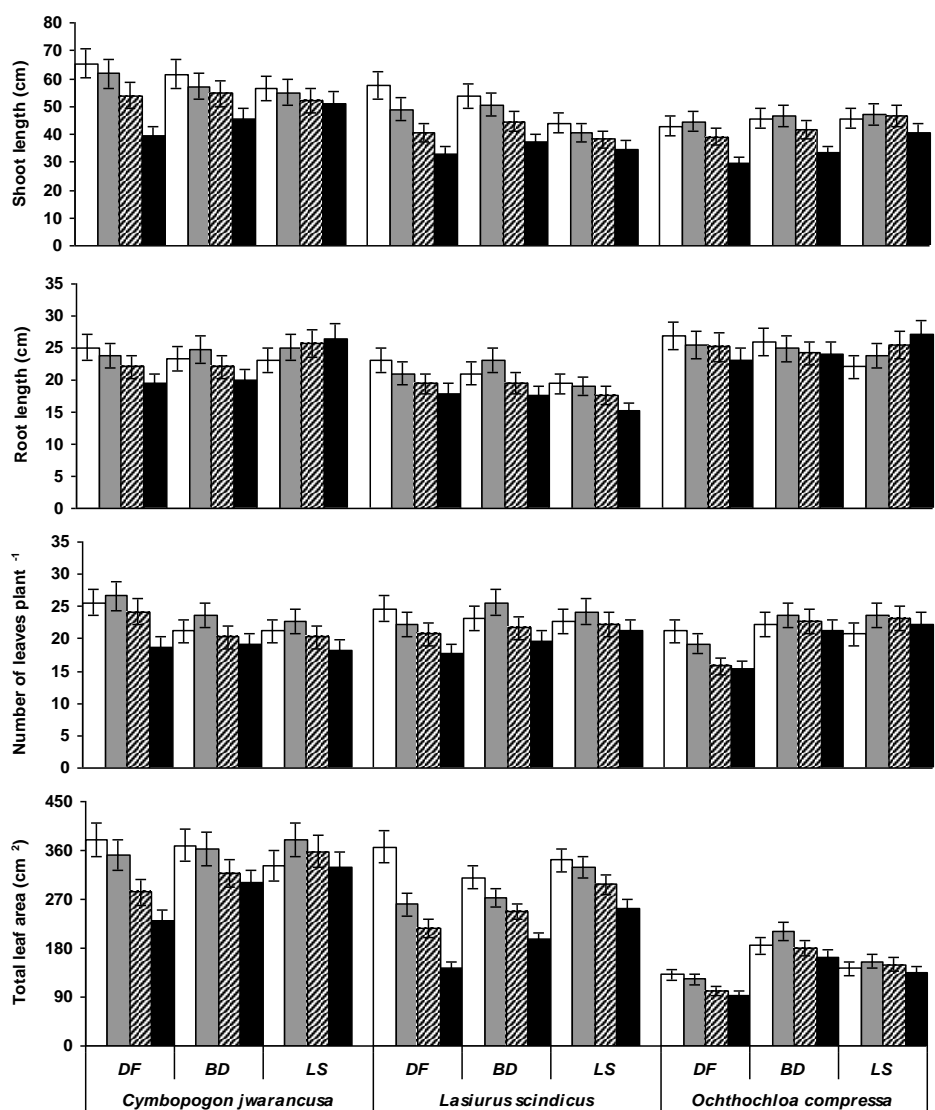


Fig. 1. Growth parameters in some salt tolerant grasses from the Cholistan desert under salt stress (□ 0 mM, ■ 100 mM, ▨ 200 mM, and ■ 300 mM NaCl).

The pattern of reduction in root dry weights of all the grasses was very much similar to that recorded for root fresh weight (Fig. 2). The populations of *C. jwarancusa* and *L. scindicus* from DF were the most affected due to salt stress, in which a consistent decrease in root dry weight was observed with increase in external salt level. Root dry weight in the populations of *O. compressa*, from all the ecotypes was least affected by the increasing salt levels, but the LS population showed increased root dry weight with increase in salt level of the external growth medium.

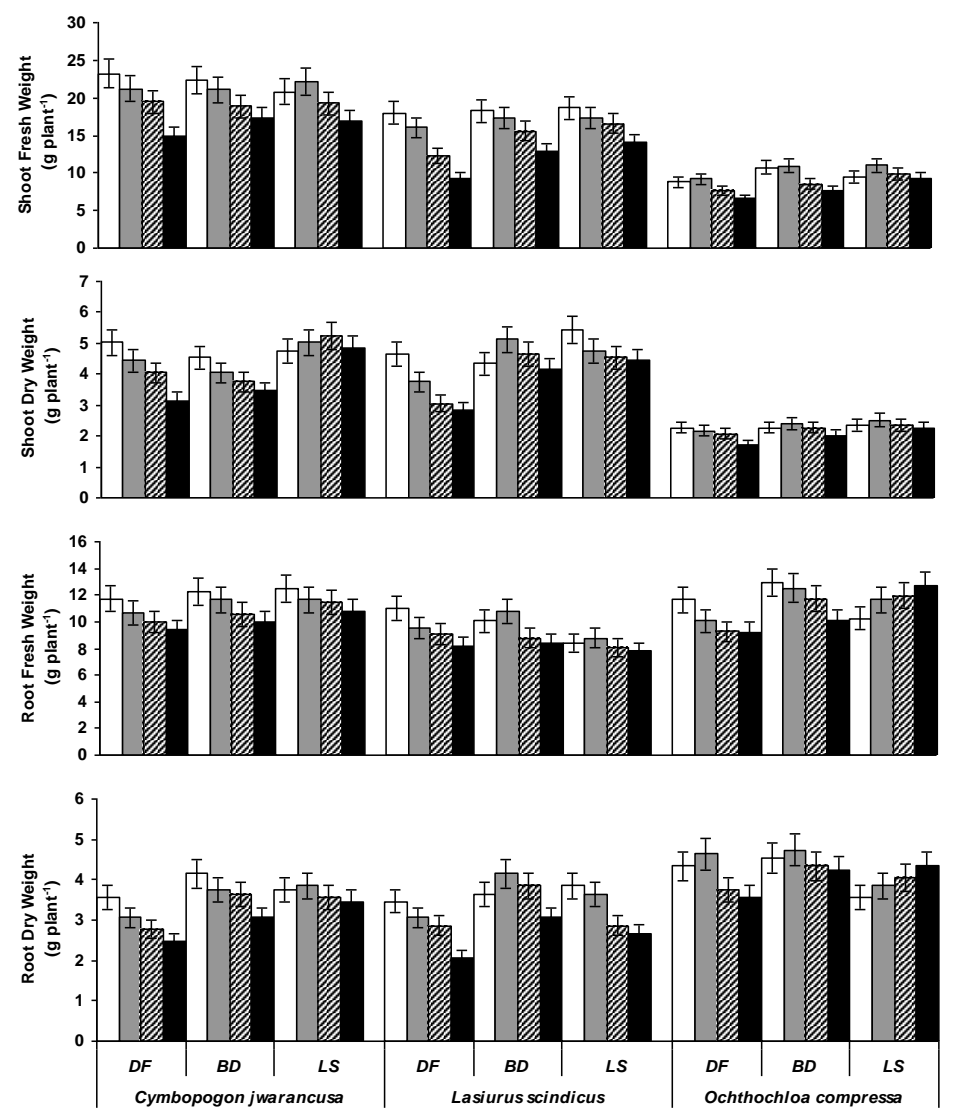


Fig. 2. Root and shoot fresh and dry weights in some salt tolerant grasses from the Cholistan desert under salt stress (□ 0 mM, ■ 100 mM, ▨ 200 mM, and ■ 300 mM NaCl).

Discussion

Plants growing in desert regions have to counteract a number of environmental hazards such as high temperature, soil salinity and water stress due to low precipitation which make the desert region a very challenging environment for plants (Khan & Weber, 2006; Weber, 2009). Such natural harsh climatic conditions in predominantly arid regions evolve a diverse group of salt tolerant plants that have multiple mechanisms of salt tolerance (Breckle, 2004).

Cholistan is the hottest desert in Pakistan characterized by high sand dunes and saline inter-dunal flats at patches, which supports very specific vegetation. The inhabiting species are facing multiple stresses simultaneously, and these natural populations must have developed diverse mechanisms for their growth and survival under unfavourable conditions. The adapted species of the Cholistan desert can be very useful to study a variety of mechanisms in relation to agro-morphology, physiology and structural modifications in detail.

Shoot growth is of prime importance among a number of growth parameters in appraising the salt tolerance of plants. It may vary with species or cultivars, or even within a population. There is a lot of material available on salt-induced growth reduction, even in halophytic species, e.g., Qian *et al.*, (2001) in Kentucky bluegrass, Gulzar *et al.* (2003) in coastal salt marsh grass *Urochondra setulosa* and Alshammmary *et al.*, (2004) in Kentucky bluegrass, tall fescue, alkaligrass, and saltgrass. The main reason of growth reduction could be due to decrease in turgor developed by water potential imbalance (Bohnert *et al.*, 1995). Natural populations have ability to limit the assimilation rate, and ultimately enhance water use efficiency, so such reduction in growth may be an important adaptation in salt tolerant populations to cope with harsh climatic conditions (Monteverdi *et al.*, 2008).

The populations from Ladam Sir (LS) of all three grasses were relatively less affected by elevating salt levels in the growth medium. However, *Ochthochloa compressa* performed little better in relation to shoot length, as 100 mM NaCl level in the DF and BD populations and 100 and 200 mM NaCl in the LS population stimulated their growth. Growth stimulation with salt was also reported by Hameed *et al.*, (2008) in salt tolerant grasses *Cynodon dactylon* and *Sporobolus arabicus*.

Leaf area is also one of the important growth attributes contributing to overall plant growth under normal or saline conditions (Monteverdi *et al.*, 2008). The reduction in leaf area is the principal strategy that makes it possible to attenuate the effects of the reduced availability of water under saline stress (Alem *et al.*, 2002). The populations of all three grasses from Derawar Fort (DF) were adversely affected by increasing salt levels of the growth medium. However, the populations of *Lasiurus scindicus* from all three habitats were most adversely affected due to salt stress as compared to other grasses used in the experiment. Leaf area of *O. compressa* was least affected due to salt stress as was reported by Greipsson & Davy (1996) in more tolerant genotypes of *Leymus arenarius*.

Generally, salt stress can considerably affect root length in grasses (Ashraf *et al.*, 2002), but this may vary with plant species or genotype. Similar reduction in root length was reported by Alshammmary *et al.*, (2004) in salt tolerant grasses like Kentucky bluegrass, tall fescue, alkaligrass, and saltgrass. Furthermore, Jaradat *et al.*, (2004) related salt tolerance with increased root number and length under salinity stress. Similar findings have earlier been reported by Marcum (1999) in some salt tolerant grasses. The populations of *O. compressa* and *C. jwarancusa* from LS increased their root length with increase in salt level of the medium, and therefore, can be rated as more salt tolerant in view of the above cited literature.

The populations of all three grasses showed reduction in their shoot and root fresh and dry weights under salt stress. However, the population of *O. compressa* was relatively less affected in relation to fresh and dry biomass production than that recorded in the other grasses.

Ochthochloa compressa from BD and LS and *C. jwarancusa* from LS showed increased shoot fresh and root biomass under salt stress and therefore rated as more tolerant to salt stress than the others. Salt-stimulated growths in monocotyledons are rare (Bell & O'Leary, 2003). However, there are some reports on the optimal growth of salt tolerant or halophytic species at moderate salinities e.g., in *Puccinellia peisonis* (Stelzer

& Laüchli, 1977), *Leymus arenarius* (Greipsson & Davy, 1996), *Sporobolus virginicus* (Bell & O'Leary, 2003), and *Cynodon dactylon* (Hameed and Ashraf, 2008). Increased root fresh and dry weight under salt stress in *O. compressa* from LS and this was again a confirmation of highly salt tolerant species as was reported by Gulzar *et al.*, (2003) in *Aeluropus lagopoides*.

In conclusion, all the ecotypes of all three grasses from highly saline habitat Ladam Sir (LS) were relatively less affected by salt stress in terms of various growth parameters studied than their counterparts from moderately saline Bailahwala Dahar (BD) and low saline Derawar Fort (DF). Of the three grasses, *Ochthochloa compressa* was superior in salt tolerance as it was least affected by high salt levels of the growth medium, and even the growth of the population from the highly saline LS was stimulated at 100-200 mM NaCl in terms of root and shoot length and fresh and dry biomass production.

Acknowledgement

This work is a part of Ph.D. thesis of Miss Nargis Naz, Regd. No. 2003-ag-468.

References

- Alem, C., M. Labhilili, K. Brahmi, M. Jlibene, N. Nasrallah and A.F. Maltouf. 2002. Hydrous and photosynthetic adaptations of common and durum wheat to saline stress. *Comptes Rendus Biologies*, 325: 1097-1109.
- Allen, J.A., J.L. Chambers and M. Stine. 1994. Prospects for increasing salt tolerance of forest trees: a review. *Tree Physiol.*, 14: 843-853.
- Alshammmary, S.F., Y.L. Qian and S.J. Wallner. 2004. Growth response of four turfgrass species to salinity. *Agric. Water Manag.*, 66: 97-111.
- Anonymous. 1954. USDA Laboratory Staff. Diagnosis and Improvement of Saline and Alkali Soils. US Government Printing Office, Washington, DC.
- Arshad, M., Anwar-ul-Hussan, M.Y. Ashraf, S. Noureen and M. Moazzam. 2008. Edaphic factors and distribution of vegetation in the Cholistan Desert, Pakistan. *Pak. J. Bot.*, 40: 1923-1931.
- Ashraf, M. 2003. Relationships between leaf gas exchange characteristics and growth of differently adapted populations of Blue panicgrass (*Panicum antidotale* Retz.) under salinity or waterlogging. *Plant Sci.*, 165: 69-75.
- Ashraf, M. 2004. Some important physiological selection criteria for salt tolerance in plants. *Flora*, 199: 361-376.
- Ashraf, M. and H. Ahmad. 1995. Response of three arid zone grasses to salt and waterlogging. *Arid Soil Res. Rehabil.*, 9: 137-154.
- Ashraf, M. and N. Yasmin. 1997. Responses of some arid zone grasses to brackish water. *Tropenlandwirt*, 98: 3-12.
- Ashraf, M. and P.J.C. Harris. 2004. Potential biochemical indicators of salinity tolerance in plants. *Plant Sci.*, 166: 3-16.
- Ashraf, M., M. Hameed, M. Arshad, Y. Ashraf and K. Akhtar. 2006. Salt tolerance of some potential forage grasses from Cholistan Desert of Pakistan. In: *Ecophysiology of High Salinity Tolerant Plants*. (Eds.): M.A. Khan and D.J. Weber. Springer, Netherlands.
- Ashraf, M., Y.K. Akhtar, G. Sarwar and M. Ashraf. 2002. Evaluation of arid and semi-arid ecotypes of guar (*Cyamopsis tetragonoloba* L.) for salinity (NaCl) tolerance. *J. Arid Environ.*, 52: 473-482.
- Bell, H.L. and J.W. O'Leary. 2003. Effects of salinity on growth and cation accumulation of *Sporobolus virginicus* (Poaceae). *Am. J. Bot.*, 90: 1416-1424.
- Bohnert, H.J., D.E. Nelson and R.G. Jensen. 1995. Adaptations to environmental stresses. *Plant Cell*, 7: 1099-1111.
- Breckle, S.W. 2004. Flora, Vegetation und Ökologie der alpin-nivalen Stufe des Hindukusch (Afghanistan). In: *Proceed. 2nd Symposium A.F.W. Schimper-Foundation: Results of worldwide ecological studies*. (Eds.): S.W. Breckle, B. Schweizer & A. Fangmeier. Stuttgart-Hohenheim, p. 97-117.

- Greipsson, S. and A.J. Davy. 1996. Sand accretion and salinity as constraints on the establishment of *Leymus arenarius* for land reclamation in Iceland. *Ann. Bot.*, 78: 611-618.
- Gulzar, S., M.A. Khan and I.A. Ungar. 2003. Effects of salinity on growth, ionic content, and plant-water status of *Aeluropus lagopoides*. *Comm. Soil Sci. Plant Anal.*, 34: 1657-1668.
- Hameed, M. and M. Ashraf. 2008. Physiological and biochemical adaptations of *Cynodon dactylon* (L.) Pers., from the Salt Range (Pakistan) to salinity stress. *Flora*, 203: 683-694.
- Hameed, M., N. Naz, M.S.A. Ahmad, Islam-ud-Din and A. Riaz. 2008. Morphological adaptations of some grasses from the salt range, Pakistan. *Pak. J. Bot.*, 40: 1571-1578.
- Hoagland, D.R. and D.I. Arnon. 1950. The water culture method for growing plants without soil. In: *Circular No. 347, Univ Calif Agric Exp Stn, Berkeley, CA*, pp. 1-39.
- Jaradat, A.A., M. Shahid and A. Al-Maskri. 2004. Genetic diversity in the batini barley landrace from Oman. II. Response to salinity stress. *Crop Sci.*, 44: 997-1007.
- Khan, M.A. and D.J. Weber. 2006. *Ecophysiology of High Salinity Tolerant Plants*. Springer, Netherlands.
- Maggio, A., P.M. Hasegawa, R.A. Bressan, M.F. Consiglio and R.J. Joly. 2001. Unraveling the functional relationship between root anatomy and stress tolerance. *Aust. J. Plant Physiol.*, 28: 999-1004.
- Marcum, K.B. 1999. Salinity tolerance mechanisms of grasses in the subfamily Chloridoideae. *Crop Sci.*, 39: 1153-1160.
- Marcum, K.B., S.J. Anderson and M.C. Engelke. 1998. Salt gland ion secretion: A salinity tolerance mechanism among five zoysiagrass species. *Crop Sci.*, 38: 806-810.
- Monteverdi, C.M., M. Lauteri and R. Valentini. 2008. Biodiversity of plant species and adaptation to drought and salt conditions. Selection of species for sustainable reforestation activity to combat desertification. In: *Biosaline Agriculture and High Salinity Tolerance*. (Eds.): C. Abdelly, M. Öztürk, M. Ashraf and C. Grignon. Birkhäuser Verlag, Switzerland, pp. 197-206.
- Naz, N., M. Hameed, A. Wahid, M. Arshad and M.S.A. Ahmad. 2009. Patterns of ion excretion and survival in two stoloniferous arid zone grasses. *Physiol. Plant.*, 135: 185-195.
- Niknam, S.R. and J. McComb. 2000. Salt tolerance screening of selected Australian woody species-a review. *Forest Ecol. Manag.*, 139: 1-19.
- Poljakoff-Mayber, A. 1988. Ecological-physiological studies on the responses of higher plants to salinity and drought. *Sci. Rev. Arid Zone Res.*, 6: 163-183.
- Qian, Y.L., S.J. Wilhelm and K.B. Marcum. 2001. Comparative responses of two Kentucky bluegrass cultivars to salinity stress. *Crop Sci.*, 41: 1895-1900.
- Rausch, T., M. Kirsch, R. Low, A. Lehr, R. Viereck and A. Zhigang. 1996. Salt stress responses of higher plants: the role of proton pumps and Na^+/H^+ -antiporters. *J. Plant Physiol.*, 148: 425-433.
- Robinson, D., D.C. Gordon and W. Powell. 1997. Mapping physiological traits in barley. *New Phytol.*, 137: 149-157.
- Sabir, P. and M. Ashraf. 2007. Screening of local accessions of *Panicum maliaceum* L., for salt tolerance at the seedling stage using biomass production and ion accumulation as selection criteria. *Pak. J. Bot.*, 39: 1655-1661.
- Stelzer, R. and A. Lauchli. 1977. Salt- and flooding tolerance of *Puccinellia peisonis*: I. The effect of NaCl- and KCl-salinity on growth at varied oxygen supply to the root. *Z. Pflanzenphysiol.*, 83: 35-42.
- Weber, D.J. 2009. Adaptive mechanisms of halophytes in desert regions. In: *Salinity and water stress*. (Eds.): M. Ashraf, M. Ozturk and H.R. Athar. Springer-Verlag, pp: 179-186.
- Winicov, I. 1998. New molecular approaches to improving salt tolerance in crop plants. *Ann. Bot.*, 82: 703-710.
- Winicov, I. and D.R. Bastola. 1997. Salt tolerance in crop plants: new approaches through tissue culture and gene regulation. *Acta Physiol. Plant.*, 19: 435-449.
- Yadava, N.D., N.P. Singh, M.L. Soni and R.K. Beniwal. 2004. Response of sewan (*Lasiurus syndicus*) to saline water irrigation and fertilizer application for fodder production in arid western Rajasthan. *Curr. Agri.*, 28: 27-31.