COMMUNICATIONS IN SOIL SCIENCE AND PLANT ANALYSIS
Vol. 34, Nos. 11 & 12, pp. 1657–1668, 2003

Effects of Salinity on Growth, Ionic Content, and Plant–Water Status of *Aeluropus lagopoides*

Salman Gulzar,[1,2] M. Ajmal Khan,[1] and Irwin A. Ungar[3,*]

[1]Department of Botany, University of Karachi, Karachi, Pakistan
[2]Department of Botany, Government Superior Science College, Shah Faisal Colony, Karachi, Pakistan
[3]Department of Environmental and Plant Biology, Ohio University, Athens, Ohio, USA

ABSTRACT

The effect of sodium chloride (NaCl) in the external medium upon the growth, plant–water status, and ionic content of *Aeluropus lagopoides* was studied. Plants grown in non-saline and 200 mM NaCl had the greatest fresh and dry weights. Increasing salinity (400 to 1000 mol m^{-3} NaCl) caused a decrease in fresh and dry weights of plants. The water potential and osmotic potential of shoots became more negative with an increase in salinity, which was associated with an accumulation of Na^+ and Cl^- in leaves. Shoot Ca^{2+}, Mg^{2+}, and K^+ concentration remained constant in salinity treatments, while Na^+ increased in salt treatments and reached greater than 4 mol m^{-3} g^{-1} dry weight.

*Correspondence: Irwin A. Ungar, Department of Environmental and Plant Biology, Ohio University, Athens, OH 45701-2979, USA; Fax: 740-593-1130; E-mail: ungar@ohio.edu.

DOI: 10.1081/CSS-120021303 0010-3624 (Print); 1532-2416 (Online)

Key Words: *Aeluropus lagopoides*; Growth; Halophyte; Ionic content; Salt tolerance; Water relations.

INTRODUCTION

Salinization of soils is a major environmental, agricultural and community problem throughout the Indus Basin of Pakistan. Canal irrigation by flooding raises the water table, which eventually leads to either waterlogging or high soil salinity. In addition, salinity builds up rapidly in coastal areas, which are not regularly inundated with seawater. Salt tolerance of various grass species has been reported,[1–5] and, in addition, some information is also available on the effect of salinity on local grasses.[5–11]

Mahmood et al.[11] found that grasses collected from saline areas of Faisalabad, Pakistan, vary in their salt tolerance. *Sporobolus arabicus* was more tolerant to salinity than other grasses *Cynodon dactylon*, *Polypogon monspeliensis*, and *Desmostachya bipinnata*. *Sporobolus arabicus* could survive in up to 30 $dS\,m^{-1}$ specific conductance (309 mM NaCl) and this response was attributed to a greater accumulation of Na^+ in plant tissues than for other species.[11] Khan et al.[5] found that fresh and dry weights of roots and shoots of *Halopyrum mucronatum* were greatest for plants grown at 90 mM NaCl, but growth was inhibited at higher salinities and all plants died at 360 mM NaCl. Osmotic adjustment occurred under increased salinity and was with an increase in the sodium and chloride concentrations of shoots, decreased shoot potassium concentration, and decreased shoot succulence.[4,12] The accumulation of high salt concentrations in organs of grasses usually serves to achieve osmotic balance under saline conditions by adjusting the plants water potential to more negative levels than that of the growth medium.[5,13–15]

Aeluropus lagopoides (Linn.) Trin. Ex Thw. (Poaceae) is a salt-secreting rhizomatous perennial grass distributed from Northern Africa (Morocco to Somalia), Sicily and Cyprus, through the Middle East to Central Asia, Pakistan and India.[16] It is capable of vegetative reproduction through rhizome growth after monsoon rains and also can produce numerous flowers and seeds from April to October.[17] It is often found in association with *Cressa cretica* in the inland communities and with *Cyperus arenarius*, *Cressa cretica*, and *Halopyrum mucronatum* in the coastal communities located at the backwaters of the Manora creek, Pakistan. *Aeluropus lagopoides* is used as a forage plant in parts of India throughout the year.[18] They found little variation in salt content of plant parts even with a three-fold increase in soil salinity in summer possibly due to salt secretion through leaf glands. The low salt content of *A. lagopoides* shoots is a favorable characteristic for its use as fodder. Gulzar and Khan[17] reported that seeds of *A. lagopoides* showed 30% germination at 500 mM NaCl under optimal temperature conditions. This indicates that this species is highly salt tolerant during germination. *Aeluropus lagopoides* is a potential fodder crop and it could also be used as a coastal dune stabilizer. There is little information available on the salt tolerance of this species. This study was conducted to determine the level of salt tolerance in this species. Growth parameters, plant-water status and ionic responses of *A. lagopoides* to different levels of salinity were investigated.

MATERIALS AND METHODS

Seeds of *A. lagopoides* were collected during the winter of 2000 from an arid inland saline area located at the University of Karachi campus, Pakistan. Seeds were separated from the inflorescence and stored at 4°C and growth studies were started immediately in an open air greenhouse located at the University of Karachi. Seeds were surface sterilized using the clorox (0.85% sodium hypochlorite). Seeds were germinated in 10 cm × 8 cm plastic pots filled three fourths with sandy soil and seedlings were irrigated with half-strength Hoagland and Arnon solution No. 2[19] for two weeks until they reached 5 cm in height. They were then thinned to five plants in each pot and a half-strength Hoagland and Arnon solution no. 2 nutrient solution was used to supply the macronutrients and micronutrients were added.[19] Pots were placed in trays and then sub-irrigated, and the water level was adjusted daily to correct for evaporation. Salt solutions were completely replaced once a week to avoid build-up of salinity in pots. Six salinity treatments were employed (0, 200, 400, 600, 800, and 1000 mM NaCl) after a preliminary test of salinity tolerance. In order to avoid salt shock, salinity levels were raised gradually at daily intervals. Plants were grown under saline conditions for 6 weeks after maximum salinity was achieved.

Plants were harvested after six weeks and measurements were made of fresh and dry weight of shoot and root, length of shoot and root, and number of leaves and tillers. Plants were oven-dried at 80°C for 48 h before dry weight was determined. Water potential was measured by the psychrometric technique on punched leaf disks from randomly chosen leaves in a C-52 chamber with a HR-33 dew point microvoltmeter (Wagtech). Sap was expressed from leaves onto Whatman No. 1 filter paper disks for measuring leaf osmotic potential with the dew point microvoltmeter. Leaf turgor pressure was estimated by calculating the difference between the leaf osmotic and water potentials. Leaf stomatal conductance was determined with a Delta-T AP-4 porometer. Chloride ion was measured with a Beckman specific ion electrode. Cation content of roots and shoots was analyzed using a Perkin–Elmer model 360 atomic absorption spectrophotometer. The Na^{+} and K^{+} content of shoots and roots were determined by flame emission spectrophotometry and Ca^{2+} and Mg^{2+} levels by atomic absorption spectrophotometry.

A completely randomized ANOVA analysis was used to test for significant differences among mean values for growth, plant-water relations and ion concentrations. A Bonferroni test was carried out to determine whether or not significant differences occurred between individual treatment means.[20]

RESULTS

Fresh weight of shoots ($F = 5.73$, $P < 0.012$) and roots ($F = 6.72$, $P < 0.007$) decreased progressively with an increase in salinity (Fig. 1).

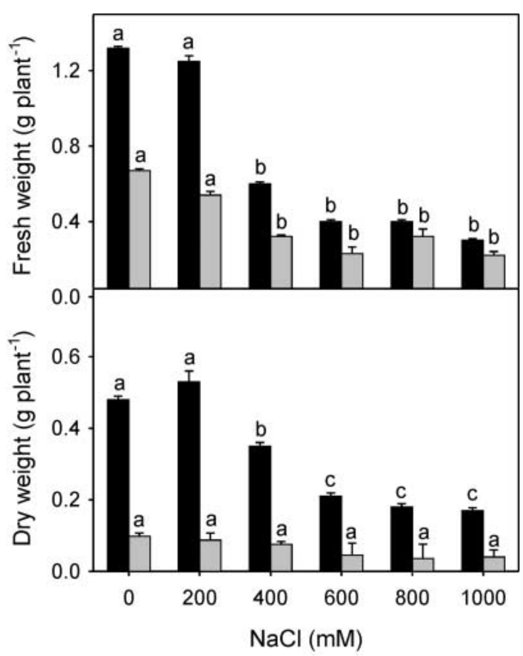

Figure 1. Effects of NaCl (0, 200, 400, 600, 800, 1000 mM) on mean ± S.E. fresh and dry weight of shoots (black bar) and roots (gray bar) of *Aeluropus lagopoides*. Different letters above bars represent significant ($P < 0.05$) differences between salinity treatments.

Root fresh weight was much lower than that of shoot at low salinity, however, at higher salinities differences were small (Fig. 1). Dry weight of shoots ($F = 9.679$, $P < 0.002$) increased at 200 mM NaCl, but declined with further increases in salinity (Fig. 1), root dry weight ($F = 6.274$, $P < 0.009$), however, decreased with an increase in salinity but the difference was not statistically significant. Plants survived after 60 days at 1000 mol m^{-3} NaCl treatment but growth was limited (Fig. 1). Shoot and root lengths were greatest in the non-saline control (Fig. 2). Shoot ($F = 25.7$, $P < 0.0001$) and root ($F = 91.2$, $P < 0.0001$) length decreased significantly up to 400 mM NaCl, but further increases in salinity were not more inhibitory (Fig. 2). Leaf number per plant was reduced significantly ($F = 27.3$, $P < 0.0001$) from 25 leaves in the control to 5 leaves under high salinity (Fig. 3). Number of tillers per plant also decreased significantly ($F = 28.68$, $P < 0.0001$) from eight in the non-saline control to only one in 1000 mM NaCl (Fig. 3).

Water potential ($F = 52.97$, $P < 0.0001$) and osmotic potential ($F = 107.31$, $P < 0.0001$) was lowered with an increase in salinity, with the greatest decrease occurring in 800 mM NaCl (Fig. 4). Pressure potential ($F = 0.889$, $P < 0.505$) did not change significantly with an increase in

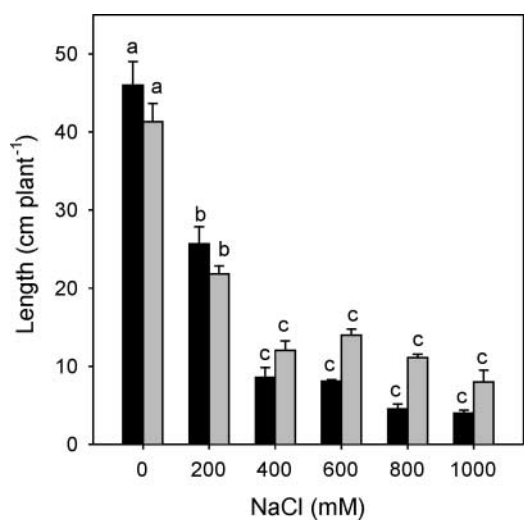

Figure 2. Effects of NaCl (0, 200, 400, 600, 800, 1000 mM) on mean ± S.E. shoot length (black bar) and root length (gray bar) of *Aeluropus lagopoides*. Different letters above bars represent significant ($P < 0.05$) differences between salinity treatments.

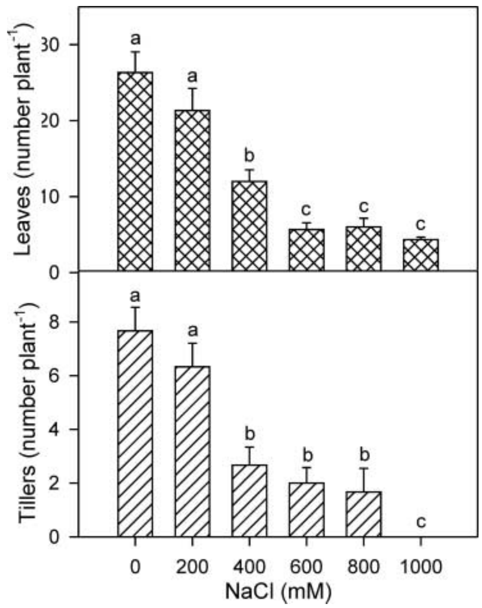

Figure 3. Effects of NaCl (0, 200, 400, 600, 800, 1000 mM) on mean ± S.E. number of leaves and tillers of *Aeluropus lagopoides*. Different letters above bars represent significant ($P < 0.05$) differences between salinity treatments.

salinity (Fig. 4). Stomatal conductance ($F = 50.713$, $P < 0.0001$) decreased significantly with an increase in salinity (Fig. 4).

A two way ANOVA showed a significant ($P < 0.05$) affect of plant organ (except for Na^+) and salinity (except for Ca^{2+}) in determining element concentrations and their interactions were significant for all elements except Ca^{2+}. Element levels in the roots were higher than those for shoots (Fig. 5). Sodium and Cl^- concentrations increased with an increase in salinity, whereas, concentrations of K^+, Mg^{2+}, and Ca^{2+} remained unchanged with an increase in salinity (Fig. 5). Ash content did not change significantly with an increase in salinity and ranged from a mean of 8 to 14% of the total dry weight of roots and shoots (Fig. 5).

DISCUSSION

Results of the present study indicate that *A. lagopoides* is a salt tolerant species. Little inhibition was recorded in media containing 200 mM NaCl, but

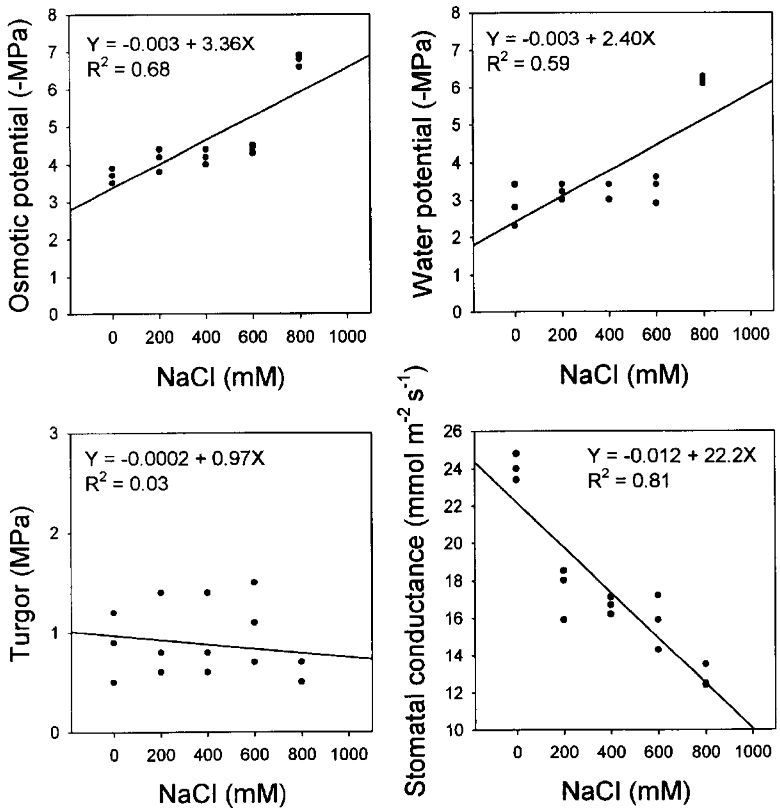

Figure 4. Effects of NaCl (0, 200, 400, 600, 800, 1000 mM) on shoot water potential, osmotic potential, pressure potential, and stomatal conductance of *Aeluropus lagopoides.*

400 mM NaCl was inhibitory to the growth. Plants survived in 600 mM NaCl with low mortality and reduced growth, but a further increase in salinity caused an increase in mortality, reaching about 50% in 45 days. Root dry weight declined at higher salinities, but the difference was not statistically significant. Other growth parameters like number of tillers, number of leaves, and shoot and root length showed a similar pattern of decline with an increase in salinity.

Grasses are quite variable in their tolerance to salinity.[5,9,10,21] Khan et al.[5] showed that biomass production of *Halopyrum mucronatum* was promoted at low salinity (90 mM NaCl) but decreased with an increase in salinity.

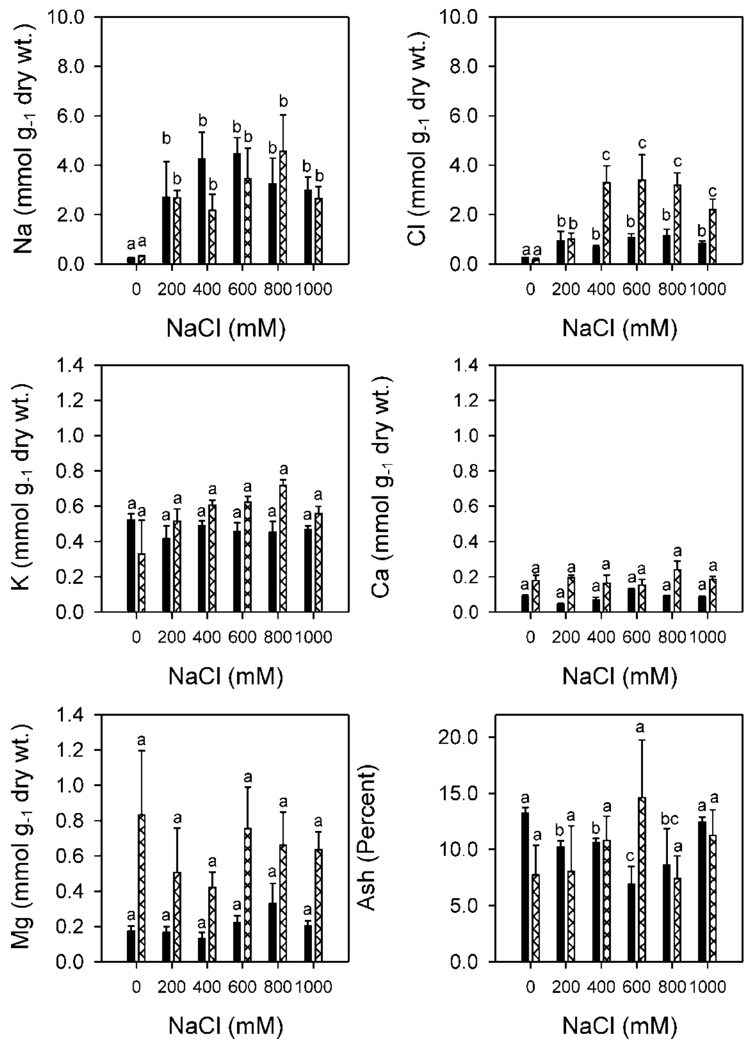

Figure 5. Effects of NaCl (0, 200, 400, 600, 800, 1000 mM) on mean ± S.E. ion and ash content of shoots (black bar) and roots (gray bar) of *Aeluropus lagopoides*. Different letters above bars represent significant ($P < 0.05$) differences between salinity treatments.

Bodla et al.[10] showed that *A. lagopoides* could survive in up to 110 dS m^{-1} NaCl (1500 mM NaCl). Mauchamp and Mésleard[22] reported that growth of *Phragmites australis* plants decreased 50% in comparison to control at 0.75% NaCl (125 mM) and plants died at 2% NaCl (342 mM). Other graminoid species like *Paspalum distichum* L.,[10] *Cyperus rotundus* L.,[7] *Cynodon dactylon*,[8] *Cenchrus pennisetiformis* Hochst. and Steud; *Panicum turgidum* Forssk.[9] are reported to survive at moderate salinities.

Aeluropus lagopoides always maintained a more negative water potential than the medium, and it decreased with an increase in salinity concentration. Khan et al.[5] also reported that *Halopyrum mucronatum* adjusted osmotically, maintaining a more negative osmotic potential than that of the medium. Antlfinger and Dunn[23] found that species growing in higher soil salinities had a more negative xylem pressure potential than plants growing in less saline areas. Xylem pressure potential for graminoids ranged from −2.2 to −2.9 MPa. Our data showed a water potential of −3.0 MPa for *A. lagopoides* plants growing under high salinity.

Aeluropus lagopoides accumulated a large amount of Na^+ and Cl^- ions in shoots and roots and lower amount of K^+, Mg^{2+}, and Ca^{2+}. Generally, ion accumulation was greater in roots than in shoots. A large percentage of Cl^- was sequestered in the roots and did not reach the shoot. Khan et al.[5] reported that Na^+ and Cl^- concentrations increased with an increase in salinity, while Ca^{2+}, Mg^{2+}, and K^+ decreased. Keiffer and Ungar[24] reported that *Hordeum jubatum* stored a large amount of Na^+ and Cl^- in its organs compared with other cations. Flowers et al.[25] determined that most monocotyledonous halophytes accumulated K^+ and excluded Na^+ from the shoot. At all salinities in this experiment with *A. lagopides*, plants selectively accumulated Cl^- and Mg^{2+} in roots. Warwick and Halloran[26] reported that Cl^- concentration in the sheath was lower than that of Na^+, even though there were equal amounts of Na^+ and Cl^- in the external solution and that *Diplachne fusca* also had the capacity to exclude greater proportion of Cl^- than Na^+ from shoot tissue. Bhatti and Wieneke[27] and Huang and Van Steveninck[28] found higher levels of Cl^- in the epidermal and mesophyll cell of sheath tissue compared with the mesophyll of the leaf blade in salt-stressed *Hordeum vulgare* seedlings. Exclusion of high concentration of Cl^- from shoots may be a mechanism to prevent this ion from interfering with photosynthetic and other metabolic processes in leaves.

Shoots of *A. lagopoides* maintained relatively constant K^+ concentration with increasing salinity, resulting in high K^+ selectivity ratios. Similar responses for shoot K^+ at high salinity have been observed in other halophytic grasses.[4,12] This contrasts with the large drop in shoot K^+ with increasing salinity frequently observed in dicotyledenous halophytes.[29] The maintenance of fairly constant K^+ concentration with increasing salinity may be interpreted as the requirement for a minimum cytoplasmic K^+ level, possibly associated with the K^+ requirement of protein synthesis.[30]

In summary, our results showed that *A. lagopoides* is a highly salt tolerant halophytic grass, which accumulates relatively low amounts of inorganic ions (10% of dry weight) to achieve a water potential gradient between soil and plants so that water uptake is not disrupted. Plants accumulated a greater amount of Cl^- in roots than shoots but Na^+ is accumulated equally in both roots and shoots. *Aeluropus lagopoides* may be used as a turf grass in the areas where fresh water is not available or to stabilize sand dunes both in the coastal and inland saline deserts of Pakistan. Joshi and Bhoite[18] indicated that *A. lagopoides* could also be used in increasing forage production in salt affected wastelands because of its high protein content and high salinity tolerance.

ACKNOWLEDGMENTS

M. Ajmal Khan thanks the University of Karachi for providing a research grant. This research was supported in part by National Science Foundation research grant INT-9730882

REFERENCES

1. Russell, J.S. Comparative salt tolerance of some tropical and temperate legumes and tropical grasses. Aust. J. Exp. Agric. Anim. Husb. **1976**, *16*, 103–109.
2. Shannon, M.C. Testing salt tolerance variability among tall wheatgrass lines. Agron. J. **1978**, *70*, 719–722.
3. Venables, V.; Wilkins, D.A. Salt tolerance in pasture grasses. New Phytol. **1978**, *80*, 613–622.
4. Marcum, K.B.; Murdoch, C.L. Growth responses, ion relations, and osmotic adaptations of eleven C_4 turfgrasses to salinity. Agron. J. **1990**, *82*, 892–896.
5. Khan, M.A.; Ungar, I.A.; Showalter, A.M. The effect of salinity on growth, ion content, and osmotic relations in *Halopyrum mucronatum* (L.) Stapf. J. Plant Nutr. **1999**, *22*, 191–204.
6. Ashraf, M.; McNeilly, T.; Bradshaw, A.D. The potential for evolution of salt (NaCl) tolerance in seven grass species. New Phytol. **1986**, *103*, 299–309.

7. Shamsi, S.R.A.; Ahmad, B. Studies on salt tolerance of purple nutsedge (*Cyperus rotundus*). Indian J. Exp. Biol. **1986**, *24*, 499–504.
8. Kumar, A. Forage yield of grasses as affected by the degree of soil sodicity and soil amelioration caused by their growth. In *Proceedings of Indo-Pak workshop on soil salinity and water management*; Kumar, A., Ed.; PARC: Islamabad, Pakistan, 1990; 435–444.
9. Ashraf, M.; Naqvi, M.I. Responses of three arid zone grass species to varying Na/Ca ratios in saline sand culture. New Phytol. **1991**, *119*, 285–290.
10. Bodla, M.A.; Chaudhry, M.R.; Shamsi, S.R.A.; Baig, M.S. Salt tolerance in some dominant grasses of Punjab. In *Biology of Salt Tolerant Plants*; Khan, M.A., Ungar, I.A., Eds.; University of Karachi: Karachi, Pakistan, 1995; 190–198.
11. Mahmood, K.; Malik, K.A.; Lodhi, M.A.K.; Sheikh, K.H. Seed germination and salinity tolerance in plant species growing on saline wastelands. Biol. Plant. **1996**, *38*, 309–315.
12. Marcum, K.B.; Murdoch, C.L. Salinity tolerance mechanisms of six C_4 turfgrasses. J. Am. Soc. Hortic. Sci. **1994**, *119*, 779–784.
13. Peacock, C.H.; Deudeck, A.E. Physiological and growth response of seashore *Paspalum* to salinity. Hortic. Sci. **1985**, *20*, 111–112.
14. Ungar, I.A. *Ecophysiology of Vascular Halophytes*; CRC Press: Boca Raton, FL, 1991.
15. Dudeck, A.E.; Peacock, C.H.; Wildmon, J.C. Physiological and growth responses of St. Angustinegrass cultivars to salinity. Hortic. Sci. **1993**, *28*, 46–48.
16. Cope, T.A. Poaceae. In *Flora of Pakistan*; Nasir, E., Ali, S.I., Eds.; University of Karachi: Karachi, Pakistan, 1982.
17. Gulzar, S.; Khan, M.A. Seed germination of a halophytic grass *Aeluropus lagopoides*. Ann. Bot. **2001**, *87*, 319–324.
18. Joshi, A.J.; Bhoite, A.S. Fluctuation of mineral ions in saline soils and halophytic grass *Aeluropus lagopoides* L. Ann. Arid Zone **1988**, *27*, 191–196.
19. Moore, R.H. *Laboratory Guide for Elementary Plant Physiology*; Burgess Publishing Company: Minneapolis, MN, 1960.
20. SPSS. *SPSS 9.0 for Windows Update;* SPSS, Inc.: Chicago, IL, 1999.
21. Hester, M.W.; Mendelssohn, I.A.; McKee, K.L. Intraspecific variation in salt tolerance and morphology in the coastal grass *Spartina patens* (Poaceae). Am. J. Bot. **1996**, *83*, 1521–1527.
22. Mauchamp, A.; Mésleard, F. Salt tolerance in *Phragmites australis* populations from coastal Mediterranean marshes. Aquatic Bot. **2001**, *70*, 39–52.

23. Antlfinger, A.; Dunn, E.L. Water use and salt balance in three salt marsh succulents. Am. J. Bot. **1983**, *70*, 561–570.
24. Keiffer, C.W.; Ungar, I.A. The effect of extended exposure to hypersaline conditions on the germination of five inland halophyte species. Am. J. Bot. **1997**, *84*, 104–111.
25. Flowers, T.J.; Hajibagheri, M.A.; Clipson, N.J.W. Halophytes. Quart. Rev. Biol. **1986**, *61*, 313–337.
26. Warwick, N.W.M.; Halloran, G.M. Variation in salinity tolerance and ion uptake in accessions of brown beetle grass (*Diplachne fusca* (L.) Beauv.). New Phytol. **1991**, *119*, 161–168.
27. Bhatti, A.S.; Wieneke, J. Time sequence studies on uptake and distribution of Na^+ and Cl^- in *Diplachne fusca* (Kallar grass) grown in NaCl. J. Plant Nutr. **1984**, *7*, 1511–1526.
28. Huang, C.X.; Van Steveninck, R.F.M. Maintenance of low Cl^- concentrations in mesophyll cells of leaf blades of barley seedlings exposed to salt stress. Plant Physiol. **1989**, *90*, 1440–1443.
29. Albert, R. Salt regulation in halophytes. Oecologia **1975**, *21*, 57–71.
30. Wyn Jones, R.G.; Storey, R.; Leigh, R.A.; Ahmed, N.; Pollard, A. A hypothesis on cytoplasmic osmoregulation. In *Regulation of Cell Membrane Activities in Plants*; Marre, E., Ciferri, O., Eds.; North Holland: Amsterdam, 1977.