

## RESPONSE OF ORNAMENTAL GRASSES CULTIVATED UNDER SALINITY STRESS

Monika Henschke✉

Poznań University of Life Sciences

### ABSTRACT

Particularly adverse growing conditions are found in urban green areas, where apart from salinity stress plants are exposed to drought stress. For this reason the aim of this study was to determine the effects of negative action and enhanced resistance to substrate salinity caused by increasing doses of NaCl in cultivation of ornamental grasses *Koeleria glauca* (Spreng.) DC., *Sesleria caerulea* (L.) Ard. and *Sorghastrum nutans* (L.) Nash. Recorded results will provide a contribution to a preliminary classification of analysed grass species in terms of their tolerance to substrate salinity. *Sesleria caerulea* and *S. nutans* may be considered to be halophytes, since they tolerate substrate salinity caused by a dose of 30 g NaCl·dm<sup>-3</sup>, losing max. 50% fresh matter of the aboveground parts. At this salinity level neither RWC nor total N content changed in leaves of these grasses in comparison to those of plants not watered with saline solution. It was shown that contents of K<sup>+</sup> and Ca<sup>2+</sup> in leaves of *S. nutans* increased under the influence of salinity. *Koeleria glauca* may be considered a salt-tolerant glycophyte, since a 50% loss of fresh matter of the aboveground parts in this grass was observed at salinity caused by the dose of 10 g NaCl·dm<sup>-3</sup>, while it also accumulated much more Na<sup>+</sup> and Cl<sup>-</sup> in leaves than the above-mentioned species.

**Key words:** fresh weight, halophytes, ion homeostasis, nitrogen, relative water content, specific leaf area

### ABBREVIATIONS

FWAP – fresh weight of aboveground parts of plants,  
 FWUP – fresh weight of underground parts of plants,  
 DWAP – dry weight of aboveground parts of plants,  
 DWUP – dry weight of underground parts of plants,  
 RWC – relative water content in the leaves,  
 SLA – and specific leaf area.

### INTRODUCTION

Soil pollution caused by salinity is becoming increasingly serious. In urban green areas it is connected with the seasonal application of sodium chloride to de-ice roads [Greszta and Gruszka 2000]. Such a method of road maintenance is used mainly in Europe, as well as Canada and the USA [Howard and

Maier 2007, Cunningham et al. 2008]. A narrow belt of soil exposed to salinity, extending over many miles of roads and motorways, is actually a huge area. Sodium and chlorine ions are cyclically accumulated in roadside areas. They may be leached to deeper soil horizons; however, even heavy rainfall

✉ mohen@up.poznan.pl

may not leach them out completely. They may also be retained in soil during water evaporation [Wrochna et al. 2006]. The effect of salinity on plants is visible at the level of both individual cells and the entire organism. First of all osmotic stress and disrupted ion homeostasis disturb plant growth and development and lead to cell structure damage [Wrochna et al. 2010]. They reduce plant mass, mainly in aboveground parts, due to withering of older leaves accumulating harmful ions, they cause succulence by reducing the number of leaves, thickening of both epidermal tissues and whole leaves as well as accumulation of water [Wignarajah et al. 1975]. Mechanisms of resistance consist mainly in the regulation of ion transport by compartmentisation of  $\text{Na}^+$  and its redistribution to various organs, e.g. through an increase in  $\text{Ca}^{2+}$  content, activating genes triggering  $\text{Na}^+$  removal from cells, or an increase in the content of  $\text{K}^+$ , a nutrient playing the main role in cell osmoregulation [Parvaiz and Satyawati 2008, Türkoglu et al. 2011]. The cellular mechanisms are especially important in the case of glycophytes, in which physiological and biochemical processes contribute to their adaptation to salt stress [Sudhakar et al. 2001]. Leaf contents of primary macronutrients, i.e. nitrogen and phosphorus, may also indicate plant response to salinity. Nitrogen is an essential components of amino acids (proline, alanine, glutamine, asparagine) and proteins (osmotin) released during salt stress, while phosphorus participates e.g. in the maintenance of cell membrane structural integrity.

Plants tolerating these growing conditions are needed for urban green areas. Tolerant to salinity, perennial plants are a valuable group, since they produce the aboveground parts only seasonally. During winter, when they would be at risk of drought and salinity, their aboveground parts wither. As a result they can tolerate stress factors better than trees and shrubs, e.g. their aboveground parts are not exposed to a direct contact with saline. An attractive group within perennials comprises ornamental grasses [Dana 2002], which by nature are well-adapted to unfavorable growth conditions.

The aim of this study was to determine the effects of the negative action and enhancement of resistance responses to substrate salinity caused by increasing

NaCl doses in cultivation of ornamental grasses: *Koeleria glauca* (Spreng.) DC., *Sesleria caerulea* (L.) Ard. and *Sorghastrum nutans* (L.) Nash.

## MATERIAL AND METHODS

### Plant materials and salinity treatments

An experiment was conducted at the Marcelin Experimental Station in a greenhouse of the Poznań University of Life Sciences in the years 2010, 2011 and 2012. Young plants of *Koeleria glauca* (Spreng.) DC., *Sesleria caerulea* (L.) Ard. and *Sorghastrum nutans* (L.) Nash. were propagated in boxes of 60 specimens and prepared for further analyses. Rhizome cuttings were collected on 4 May and grown to 21 June. Plants were transplanted on that day to pots of 0.750 dm<sup>3</sup>. Each pot contained the same mass of the substrate and was placed on a bowl to prevent an uncontrolled outflow of saline. Plants were grown on the substrate of highmoor peat with mineral sand at a 20:1 ratio (v:v). Highmoor peat produced by Klasmann with pH 3.91 was limed on the basis of the neutralization curve to pH 6.40 using  $\text{CaCO}_3$  at a dose of 7.0 g·dm<sup>3</sup> peat. The ready-to-use substrate mixtures were supplemented with 2 g·dm<sup>3</sup> of a fertilizer Peters Professional PL Special (20:20:20). Young plants grown for 6 weeks under these conditions were next watered with salt solutions. At this stage *K. glauca* developed 4 blades, 12 leaves, the mean length of the flag leaf was 13.4 cm, *S. caerulea* developed 2–3 blades, 5–6 leaves, with the mean flag leaf length of 28.0 cm, while *S. nutans* developed 4 blades, 4 leaves, with the mean flag leaf length of 37.6 cm. One day after watering plants with a solution of salt the analysis of the medium showed that it contained (mg·dm<sup>-3</sup>): N- $\text{NO}_3$  – 83.3, P – 25.4, K – 75.0, Ca – 250.7, Mg – 22.7 and Cl – 12.5 at pH in  $\text{H}_2\text{O}$  6.1 and EC  $\text{mS}\cdot\text{cm}^{-1}$  2.0. Plants were subjected to salt stress induced by a range of NaCl content (g·dm<sup>-3</sup>): 0 (control), 5, 10, 15, 30 and electrical conductivity (EC) of soil saturation extract was respectively ( $\text{mS}\cdot\text{cm}^{-1}$ ): 2.0, 2.5, 3.5, 4.0, 5.7. In each variant 100 ml of saline solution were poured into the pot, while in the control it was 100 ml of distilled water. Prior to watering plants with salt solutions,

they were watered to keep 50–55% moisture by weight of the substrate. Watering salt solutions was done once. While growing, plants were watered with water to 55–60% moisture by weight of the substrate. Growing in saline lasted 8 weeks.

Plant measurements comprised growth parameters and analyses of element contents in leaves. Plants were measured and analysed after 8 weeks of cultivation under saline conditions.

### Experimental design

**Growth parameters.** Both fresh weight of aboveground parts (FWAP) and underground parts (FWUP) of plants and dry weight of aboveground parts (DWAP) and underground parts (DWUP) of plants were evaluated. Relative water content (RWC) in the leaves [Castillo 1996] and specific leaf area (SLA) were also determined [Garnier et al. 2001].

Analyses of element contents in leaves: matured and healthy leaves were sampled from all plants. Samples were dried at a temperature of 50°C, then they were homogenised and after mineralisation the content of nitrogen (total N) was determined according to Kjeldahl, phosphorus (total P) by colorimetry with ammonium molybdate, while contents of K<sup>+</sup>, Ca<sup>2+</sup> and Na<sup>+</sup> – by photometry, Mg by atomic absorption spectroscopy (AAS) and Cl<sup>-</sup> by the titration method [Nowosielski 1974]. Content ratios of K/Na, Mg/Na and Ca/Na also was evaluated.

**Statistical analysis.** Results were analysed in terms of cultivar and salinity levels, as well as fixed factors. Values of the growth parameters from three years (2010, 2011, 2012) were averaged. In each of the salinity treatments there were three replicates of five plants (each cultivated in a separate pot) and of the combinations of element contents in leaves there were three replications (years: 2010, 2011 and 2012). The Newman-Keuls test was employed to analyse differences between measured parameters. A graphical presentation of the Newman-Keuls test results is provided in the present study.

## RESULTS AND DISCUSSION

Reduction of growth mainly in the aboveground parts of plants and less frequently also the under-

ground parts is the primary symptom of the negative effect of substrate salinity, resulting from disturbed water homeostasis. The adverse effect of substrate salinity in this experiment was expressed mainly in the reduced plant mass (tab. 1). A greater resistance to salinity was found for *S. caerulea* and *S. nutans*, since substrate salinity had a negative effect only on the aboveground parts of the former species and only on the underground part of the latter species. The lowest saline dose (5 g NaCl·dm<sup>-3</sup>) did not influence FWAP in *S. caerulea*, while the greatest dose (30 g NaCl·dm<sup>-3</sup>) had a more adverse effect than a dose 50% lower (15 g NaCl·dm<sup>-3</sup>). *Koeleria glauca* and *S. nutans* responded even to the lowest salt concentration in the substrate. In the former species reduced growth was observed both in the aboveground and underground parts, whereas in the latter species it was only in the underground parts. It needs to be stressed that *S. nutans* is a C4 grass [Fay et al. 2003] with a very well developed root system, as indicated by the results presented in Table 1, where the mass of the aboveground parts is comparable to that of the underground parts, which could have contributed to the pronounced effect of the greater sensitivity of the underground parts of plants to salinity. The negative effect of salinity on dry matter of shoots and roots in *Phragmites australis* (a C3 grass, a halophyte) and *Spartina alterniflora* (a C4 grass, a halophyte) was also shown by Vasquez et al. [2006], while Nabati et al. [2013] showed it for the accumulation of dry matter in *Sorghum bicolor* (a C4 grass). However, reduction of growth only in the case of shoots under the influence of nutrient solution salinity in hydroponic culture (340 and 510 mM) was also observed by Wu et al. [1998] in *Spartina patens* (a C4 grass, a halophyte) and by Warren and Brockelman (1989) in *Distichlis spicata*, a halophytic C4 grass.

Glenn [1987] classified grasses and sedges which survived culture at a salinity of 540 mol·m<sup>-3</sup> as halophytes. In this experiment halophytes may be defined as the species, which tolerate substrate salinity caused by a dose of 30 g NaCl·dm<sup>-3</sup> (520 mol·m<sup>-3</sup>), losing under such conditions max. 50% fresh matter of the aboveground parts. In this experiment they are *S. caerulea* and *S. nutans* (tab. 1). According to Glenn [1987], species, which survived salinity at

**Table 1.** The effect of salinity stress on growth parameters of *Koeleria glauca*, *Sesleria caerulea* and *Sorghastrum nutans*

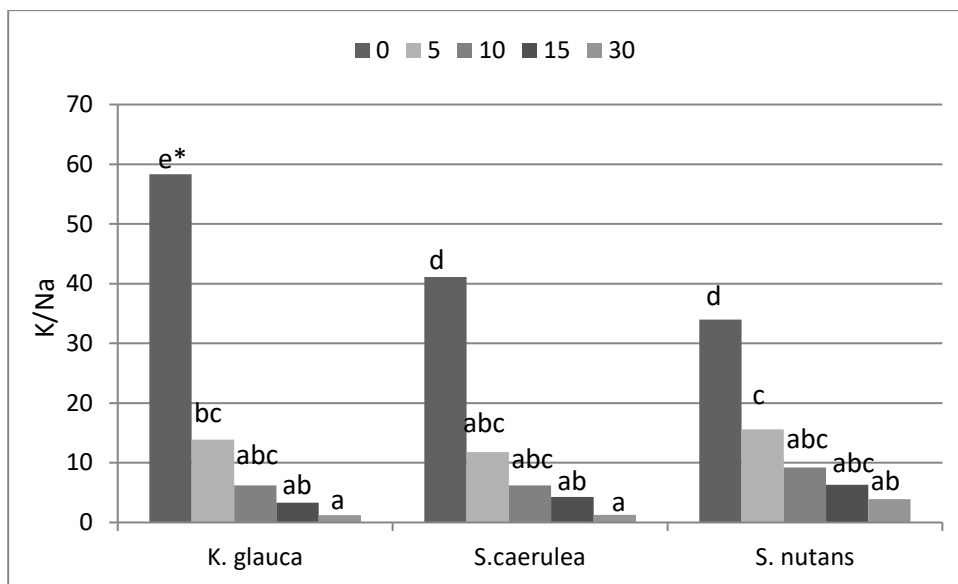
	Species	Salt concentration (g NaCl·dm <sup>-3</sup> )					Mean
		0	5	10	15	30	
Fresh weight of aboveground part FWAP (g·plant <sup>-1</sup> )	<i>Koeleria glauca</i>	12.20 j*	9.42 i	7.63 h	5.01 cd	3.61 ab	7.57 C
	<i>Sesleria caerulea</i>	7.17 gh	6.12 fg	5.54 df	5.91 ef	3.98 abc	5.74 B
	<i>Sorghastrum nutans</i>	6.44 fg	6.26 fg	4.73 cd	4.39 bc	3.17 a	5.00 A
	mean	8.61 E	7.27 D	5.97 C	5.10 B	3.59 A	
Dry weight of aboveground part DWAP (g·plant <sup>-1</sup> )	<i>Koeleria glauca</i>	5.98 h	5.14 fg	5.78 gh	4.75 ef	4.56 ef	5.24 C
	<i>Sesleria caerulea</i>	5.57 gh	4.00 de	3.63 cd	3.99 de	2.93 bc	4.03 B
	<i>Sorghastrum nutans</i>	3.57 cd	3.21 bcd	2.70 b	2.03 a	1.65 a	2.63 A
	mean	5.04 D	4.12 C	4.04 C	3.59 B	3.05 A	
Fresh weight of underground part FWUP (g·plant <sup>-1</sup> )	<i>Koeleria glauca</i>	3.51 bc	2.20 abc	2.45 abc	2.10 ab	1.44 a	2.34 A
	<i>Sesleria caerulea</i>	3.72 c	3.06 bc	2.56 abc	2.51 abc	2.56 abc	2.88 B
	<i>Sorghastrum nutans</i>	10.24 g	9.29 g	7.76 f	6.64 e	5.21 d	7.79 C
	mean	5.83 E	4.85 D	4.26 C	3.69 B	3.07 A	
Dry weight of underground part DWUP (g·plant <sup>-1</sup> )	<i>Koeleria glauca</i>	1.27 c	0.87 b	0.74 b	0.70 b	0.47 a	0.81 A
	<i>Sesleria caerulea</i>	0.99 b	0.84 b	0.79 b	0.97 b	0.72 b	0.86 A
	<i>Sorghastrum nutans</i>	3.46 g	3.22 f	3.13 f	2.39 e	1.82 d	2.91 B
	mean	1.91 D	1.64 C	1.56 C	1.35 B	1.00 A	
RWC	<i>Koeleria glauca</i>	0.86 bcd	0.93 fg	0.87 bcde	0.82 b	0.84 bc	0.86 A
	<i>Sesleria caerulea</i>	0.83 b	0.91 efg	0.88 cdef	0.73 a	0.94 fg	0.86 A
	<i>Sorghastrum nutans</i>	0.95 g	0.75 a	0.93 fg	0.90 defg	0.90 defg	0.89 B
	mean	0.88 B	0.87 B	0.89 B	0.82 A	0.89 B	
SLA	<i>Koeleria glauca</i>	5.22 ef	4.23 c	3.03 b	5.00 def	3.16 b	4.13 AB
	<i>Sesleria caerulea</i>	4.67 cde	4.45 cd	3.50 b	4.65 cde	4.21 c	4.30 B
	<i>Sorghastrum nutans</i>	5.22 ef	5.58 f	4.27 c	2.43 a	2.42 a	3.98 A
	mean	5.04 E	4.75 D	3.60 B	4.03 C	3.26 A	

\* Average marked with the same letters are not significantly different at  $\alpha = 0.05$

**Table 2.** Effect of salinity stress on elements contents in leaves of *Koeleria glauca*, *Sesleria caerulea* and *Sorghastrum nutans* (g·kg<sup>-1</sup>DM)

	Species	Salt concentration (g NaCl·dm <sup>-3</sup> )					Mean
		0	5	10	15	30	
N total	<i>Koeleria glauca</i>	31.40 cde*	30.13 bcd	29.07 bc	29.07 bc	26.87 b	29.31 B
	<i>Sesleria caerulea</i>	34.90 efg	36.30 fg	37.70 g	36.33 fg	33.33 def	35.71 C
	<i>Sorghastrum nutans</i>	11.00 a	11.40 a	12.00 a	12.00 a	15.00 a	12.28 A
	mean	25.77 A	25.94 A	26.26 A	25.80 A	25.07 A	
P total	<i>Koeleria glauca</i>	13.20 c	13.97 c	12.27 c	12.40 c	9.90 b	12.34 B
	<i>Sesleria caerulea</i>	18.67 d	21.13 d	20.07 d	20.13 d	18.67 d	19.73 C
	<i>Sorghastrum nutans</i>	5.33 a	6.47 a	6.47 a	5.67 a	4.47 a	5.68 A
	mean	12.40 B	13.86 B	12.93 B	12.73 B	11.01 A	
K <sup>+</sup>	<i>Koeleria glauca</i>	50.70 f	49.20 f	40.03 de	39.97 de	36.37 d	43.25 C
	<i>Sesleria caerulea</i>	28.37 c	25.47 bc	23.73 abc	22.00 ab	22.03 ab	24.32 A
	<i>Sorghastrum nutans</i>	19.03 a	23.83 abc	25.77 bc	28.33 c	42.17 e	27.83 B
	mean	32.70 B	32.83 B	29.84 A	30.10 A	33.52 B	
Ca <sup>2+</sup>	<i>Koeleria glauca</i>	14.83 abc	19.63 c	20.33 c	24.70 d	27.87 d	21.47 C
	<i>Sesleria caerulea</i>	11.93 a	13.33 a	12.73 a	13.97 ab	16.90 abc	13.77 A
	<i>Sorghastrum nutans</i>	12.73 a	15.20 abc	16.20 abc	16.33 abc	19.33 bc	15.96 B
	mean	13.17 A	16.06 B	16.42 B	18.33 B	21.37 C	
Mg <sup>2+</sup>	<i>Koeleria glauca</i>	3.63 a	4.13 a	3.73 a	3.47 a	3.33 a	3.66 A
	<i>Sesleria caerulea</i>	3.70 a	3.87 a	3.80 a	3.77 a	3.43 a	3.71 A
	<i>Sorghastrum nutans</i>	4.20 a	3.90 a	4.00 a	3.97 a	3.93 a	4.00 B
	mean	3.84 A	3.97 A	3.84 A	3.73 A	3.56 A	
Na <sup>+</sup>	<i>Koeleria glauca</i>	0.87 a	3.54 a	6.49 a	12.19 b	29.28 d	11.01 B
	<i>Sesleria caerulea</i>	0.69 a	2.15 a	3.82 a	5.16 a	16.84 c	5.73 A
	<i>Sorghastrum nutans</i>	0.56 a	1.53 a	2.80 a	4.47 a	10.74 b	4.02 A
	mean	1.61 A	2.41 A	4.37 A	7.27 B	18.95 C	
Cl <sup>-</sup>	<i>Koeleria glauca</i>	7.60 b	12.07 c	15.07 d	23.63 e	32.27 g	18.13 B
	<i>Sesleria caerulea</i>	4.23 a	8.10 b	11.50 c	10.63 c	30.93 g	13.08 A
	<i>Sorghastrum nutans</i>	5.33 a	7.40 b	10.33 c	11.17 c	29.23 f	12.69 A
	mean	5.72 A	9.19 B	12.30 C	15.14 D	30.81 E	

\* Explanation as in Table 1



**Fig. 1.** Effect of salinity stress (0, 5, 10, 15 and 30 g NaCl·dm<sup>-3</sup>) on K/Na ratios in leaves of *Koeleria glauca*, *Sesleria caerulea* and *Sorghastrum nutans*. \* Explanation as in Table 1

180 mol·m<sup>-3</sup>, are salt-tolerant glycophytes. This group would also include *K. glauca* from this experiment, which at a dose of 10 g NaCl·dm<sup>-3</sup> (170 mol·m<sup>-3</sup>) lost max. 50% of fresh aboveground mass. If the content of salts in the substrate exceeds a threshold value, the capacity of plants to absorb water from the substrate is limited. Damage to the root system additionally enhances this process, leads to osmotic stress and slow down growth processes [Xiong and Zhu 2002, Munns et al. 2006]. An important aspect in this respect is to determine the degree of plant sensitivity to salinity. Apart from the mass of plants, it is also advisable to determine relative water content in leaves (RWC). Sometimes response to salinity is connected with the hydration of cell sap, which contributes to the dilution of salts contained in cells and limits the adverse effect of Na<sup>+</sup> and Cl<sup>-</sup>. In this experiment the analysis of RWC showed such a response in *K. glauca* and *S. caerulea*, but only at the lowest concentration of salts (tab. 1). It is also significant that greater salt concentrations did not reduce RWC in any of the analysed species. Other studies also indicate that under salinity conditions water content in shoots of *Phragmites australis*

and *Spartina alterniflora* remained at a level similar to that in the control plants; however, the greatest salt concentration (0.5 M NaCl) caused a decrease in water content in leaves of these grasses [Vasquez et al. 2006]. A similar effect was observed in the culture of rice seedlings on Petri dishes, in which RWC was greater at a lower salinity, while at the maximum concentration (250 mM NaCl) it decreased to 54% [Farooq and Azam 2006]. However, it needs to be stressed here that seedlings grown under laboratory conditions are more sensitive to salinity than plants grown in a peat substrate.

Succulence as a result of a stressor is manifested in the thickening of leaves, epidermal cells and the thickness of the cuticle increasing at the expense of the number of stomata. Analysis of SLA in this study showed no thickening of leaves in the analysed grass species as a result of salinity (tab. 1). In contrast, substrate salinity reduced SLA in *K. glauca* and *S. nutans*, which indicates a negative effect of the stress factor. Only *S. caerulea* showed no negative effect of salinity on SLA. Also Longstreth and Strain [1977] found no changes in the area or dry matter of leaves in *Spartina alterniflora*

under the influence of strong salinity, while specific leaf weight (SLW) increased.

Under the influence of salinity caused by NaCl grasses may accumulate  $\text{Na}^+$  and  $\text{Cl}^-$  in their shoots and roots, e.g. *Zea mays*, *Festuca arundinacea*, *Phalaris arundinacea* [Maeda and Nakazawa 2008]. Potential of plants connected with defence against the adverse effect of  $\text{Na}^+$  and  $\text{Cl}^-$  is species or even variety specific, as evidenced by *Spartina alterniflora*, which tolerates salinity better than *Phragmites australis* due to its ability to use  $\text{Na}^+$  for osmotic adjustment in the shoots [Vasquez et al. 2006]. Plants may accumulate lesser amounts of these ions thanks to the barriers in their uptake from soil or their accumulation in the root system. Roots exhibit a considerable tolerance to salinity, they may control the concentration of  $\text{Na}^+$  and  $\text{Cl}^-$  ions, as well as limit xylem transport and accumulation in shoots [Munns et al. 2006]. In this experiment *K. glauca* accumulated in its leaves much greater amounts of  $\text{Na}^+$  and  $\text{Cl}^-$  than other grasses, which had a negative effect on its growth and development (tab. 2). It was not equipped with any effective barriers protecting it against accumulation of  $\text{Cl}^-$ , since with each dose of saline its content in leaves was significantly greater. However, despite the accumulation of  $\text{Cl}^-$  in the aboveground parts already at the lowest salt dose FWUP had comparable total N and  $\text{K}^+$  contents at the first three salt concentrations (5, 10, 15 g  $\text{NaCl}\cdot\text{dm}^{-3}$ ) to those in plants not watered with saline solution (tabs 1, 2). In turn, *S. caerulea* accumulated more  $\text{Na}^+$  than *S. nutans* grown in the substrate with the highest salt dose (tab. 2). Both grasses had similar  $\text{Cl}^-$  contents; in both species accumulation of this component was detected already at the lowest applied salt dose and an increase in the dose from 10 to 15 g  $\text{NaCl}\cdot\text{dm}^{-3}$  had no effect on its content in the aboveground parts of plants. However, accumulation of  $\text{Cl}^-$  already at the lowest salt dose in *S. caerulea* had no effect on root mass (tabs 1, 2). Also an increased  $\text{Na}^+$  content following the application of the greatest salt dose had no significant effect on root mass. In the case of *S. nutans*, despite the accumulation of  $\text{Cl}^-$  in the aboveground parts of plants already at the lowest salt dose, the mass of aboveground parts or FWUP were not reduced. It is possible that *S. nutans* is equipped with

adaptations to transfer  $\text{Na}^+$  from leaves to roots, which as it has already been shown, were damaged under the influence of salinity, while the aboveground parts were intact. Accumulation of  $\text{Na}^+$  in aboveground parts and in roots under the influence of salinity is frequently observed in various plant species, including halophytes, e.g. *Spartina patens* [Wu et al. 1998] and *Distichlis spicata* [Warren and Brozckelman 1989]. Moreover, halophytes are capable of inactivating  $\text{Na}^+$  by transporting them to the vacuole, it requires  $\text{Na}^+/\text{H}^+$  antiports in the tonoplast,  $\text{H}^+$  ATPases and PPlases in order to provide proton motive force. In the case of tolerant glycophytes NaCl activates antiports in the tonoplast [Glenn et al. 1999]. However, in the case of *S. nutans* used in this experiment, which accumulated less  $\text{Na}^+$  in leaves than *K. glauca* and under the influence of salinity suffered greater damage to its roots rather than the aboveground parts, it may be assumed that it is the root system that is responsible for the regulation of  $\text{Na}^+$  ions.

Substrate salinity most frequently limits leaf contents of total N,  $\text{K}^+$  and  $\text{Mg}^{2+}$  as well as reduces the K:Na ratios. The decrease in contents of primary macronutrients, i.e. total N, total P and  $\text{K}^+$ , in leaves of *K. glauca* is a definite symptom of the negative effect of salinity (tab. 2). A lower  $\text{K}^+$  content in leaves was also recorded in *Spartina pectinata* grown in saline nutrient solution in comparison to culture in the medium, which did not contain NaCl [Wu et al. 1998]. *Sesleria caerulea* and *S. nutans* in this experiment had similar total N contents in aboveground parts, irrespective of substrate salinity (tab. 2). This indicates the potential of these grasses to accumulate nitrogen from the substrate despite accumulation of  $\text{Na}^+$  and  $\text{Cl}^-$  in cells and despite its sparse availability in soils. According to Cheong and Yun [2007], in order for the plant to be able to absorb  $\text{K}^+$ ,  $\text{Ca}^{2+}$  and  $\text{NO}_3^-$ , the influx of chlorine and sodium to cells is inhibited by various defence responses, e.g.  $\text{Na}^+$  is excluded from cells or it is compartmentised. In turn, due to the marked increase in  $\text{Na}^+$  content in leaves, the ratios of this element with  $\text{K}^+$  under the influence of salinity were markedly reduced (fig. 1). It is a result frequently reported in publications on the subject [Wrochna et al. 2006]. However, it needs to

be stressed that *S. caerulea* and *S. nutans* when not watered with saline had much lower K:Na ratios than *K. glauca*. It is in halophytes rather than glycophytes that we observe a lower K:Na content [Glenn 1987]. In this experiment it was also shown that the greatest difference in the K:Na ratio between plants not watered with saline and those watered with the lowest dose (5 g NaCl·dm<sup>-3</sup>) was recorded in *K. glauca*.

Moreover, elevated contents of certain nutrients were also shown in leaves of grasses under the influence of salinity (tab. 2). The content of Ca<sup>2+</sup> in *K. glauca* under the influence of applied doses of 15 and 30 g NaCl·dm<sup>-3</sup> was significantly greater, while in *S. nutans* it was under the influence of the greatest dose in comparison to the control plants not watered with saline. In leaves of *S. nutans* we also showed a marked accumulation of K<sup>+</sup> with an increase in applied salinity doses. *Sesleria caerulea* did not have an increased content of these cations. An increase in Ca<sup>2+</sup> contents may inhibit the influx of Na<sup>+</sup> and Cl<sup>-</sup> to cells. An elevated Ca<sup>2+</sup> content indicates signal transduction, which activates genes responsible for the Na<sup>+</sup>/H<sup>+</sup> membrane antiport, regulating cytoplasm pH, cell turgor and sodium level [Shi et al. 2002, Xiong and Zhu 2002]. An increased content of Ca<sup>2+</sup>, this time in the underground parts, was shown also in other studies, in which *Spartina pectinata* was grown in saline-containing nutrient solution [Wu et al. 1998]. An increase in K<sup>+</sup> contents in *S. nutans* in this experiment indicates efficient ion metabolism. A similar effect of an increase in K<sup>+</sup> content in rice tissues (with a simultaneous elevation of RWC) under the influence of low NaCl concentrations was also shown by Farooq and Azam [2006]. Adequate nitrogen and potassium fertilisation leads to an increase in tolerance of halophytic plants to the production of biomass and to an increase in K<sup>+</sup> content in leaves of plants grown under salinity conditions [Noaman 2004]. An adequate mineral supply to plants grown in this experiment made it possible to detect such a dependence in *S. nutans*.

## CONCLUSIONS

Results of this study provide a contribution to the preliminary classification of grass species in terms of

their tolerance to substrate salinity. Halophytes may be defined as species, which tolerate substrate salinity caused by a dose of 30 g NaCl·dm<sup>-3</sup>, since under such conditions they lose max. 50% fresh weight of aboveground parts. They include *S. caerulea* and *S. nutans*. Moreover, RWC did not decrease and total N did not change in comparison to plants not watered with saline solution. *Sorghastrum nutans* had contents of K<sup>+</sup> and Ca<sup>2+</sup> significantly greater than in plants not watered with saline. The K:Na ratio was much lower in both halophytic species in culture with no salinity factor applied. *Koeleria glauca* may be considered to be a glycophytic, because accumulated much greater amounts of Na<sup>+</sup> and Cl<sup>-</sup> in leaves than the previously described species and the loss of 50% fresh mass of the aboveground parts in this grass was reported at salinity conditions caused by the application of 10 g NaCl·dm<sup>-3</sup>. At this level of salinity RWC and contents of total N, total P as well as Ca<sup>2+</sup> were comparable, while K<sup>+</sup> content was lower than in plants not watered with saline.

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