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Screening of turfgrass species and cultivars for NaCl tolerance

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Summary Information regarding the relative levels of salt tolerance between cultivars of Kentucky bluegrass (*Poa pratensis* L.) is lacking. The objectives of this study were to 1) develop a simple, quick and sensitive method of screening turfgrass species for NaCl tolerance and 2) to compare the relative salt tolerance of five cultivars of Kentucky bluegrass ('Ram I', 'Adelphi', 'Baron', 'Bensun', and 'Nassau') to other known salt tolerant turfgrass species such as alkalaigrass (*Puccinellia distans* (L.) Parl. cv. Fults) and two cultivars of red fescue (*Festuca rubra* L. 'Dawson', and 'Checker').

Alkalaigrass and both cultivars of red fescue retained a high level of salt tolerance compared to the Kentucky bluegrass cultivars. Significant variability in salt tolerance was apparent among the Kentucky bluegrass cultivars with 'Adelphi' and 'Ram I' exhibiting the best overall tolerance.

Introduction

The relative levels of salt tolerance among most of the widely used turfgrass species and cultivars have not been adequately characterized. Those species retaining some level of salt tolerance could provide a more acceptable turf in areas where only low quality irrigation water is available or saline soil conditions exist.

Several turf-type grasses are known to retain high levels of salt tolerance. Among these, alkalaigrass (*Puccinellia distans* (L.) Parl. cv. 'Fults') is known to be the most tolerant[2,5]. Harivandi *et al.*[2] have compared the salt tolerance between alkalaigrass and 4 other cool season turfgrass species. Results had shown Kentucky bluegrass (*Poa pratensis* L.) to be the least tolerant, slender creeping red fescue (*Festuca rubra* trichophylla (L.) Gaud. cv. Dawson) to be moderate, and alkalaigrass to possess a high level of salt tolerance. Further investigation had shown that tolerance and the capacity to accumulate Na^+ and Cl^- ions in alkalaigrass increased with plant age[3]. Alkalaigrass, however, is not a widely used turfgrass species. The red fescues are widely used in turf culture and have been categorized for their inherent salt tolerance[4,7]. The most tolerant group are the slender creeping red fescues. The strong creeping red fescues (*Festuca rubra* L. rubra) were categorized as moderately tolerant, and the Chewings red fescues (*Festuca rubra* L. commutata) are the least tolerant group. These variations in salt tolerance among red fescues have been well established[1].

In contrast, the salt tolerance levels between the most utilized of cool season turfgrasses, the Kentucky bluegrasses, have received very little attention. Kinbacher *et al.*[5] have reported variations among K. bluegrass cultivars for their tolerance toward 0.8% (w/v) $CaCl_2$ in irrigation water. Results had shown 'Ram I', Nugget', and 'Baron' as the most tolerant and 'Adelphi' being the least tolerant.

The objectives of this investigation were to 1) develop a sensitive and relatively quick method for screening cultivars of K. bluegrass for their tolerance to various levels of NaCl and 2) compare the salt tolerance levels of K. bluegrass to known salt tolerant turfgrass species; 'Fults' alkalaigrass, and 'Dawson' red fescue.

Materials and methods

Salt tolerance was screened between five cultivars of K. bluegrass ('Ram I', 'Adelphi', 'Baron', 'Nassau', and 'Bensun'), two cultivars of red fescue ('Dawson', and 'Checker'), and 'Fults' alkalaigrass.

Seeds from each species except the K. bluegrass were dehusked by agitation in 50.0% H_2SO_4 for 15 min followed by a 5 min rinse in distilled water. The Kentucky bluegrass cultivars were manually dehusked. The dehusked seeds were then surface sterilized with a 0.5% sodium hypochlorite solution for 10 min and then rinsed 3 times in sterile deionized water. Surface sterilizing was shown to have no detrimental effects upon seed viability. Seeds were then placed upon a modified inorganic nutrient agar medium containing the appropriate salt (NaCl) treatments.

The basal growth medium used in this study consisted of $\frac{1}{2}$ strength Murashige and Skoog (MS)[6] salt mixture. Salt (NaCl) treatments were prepared by dividing and supplementing the media with either 0.0, 17, 43, 85 or 170 m*M* NaCl (equivalent to 0.1, 0.25, 0.5 and 1.0% NaCl on a w/v basis). Agar at 8 g/l was then added to solidify the media. All media were pH adjusted to 5.8 and then autoclaved for 20 min at 121°C and 1.0 kg/cm^2 pressure. All media were then dispensed at 10 ml volumes into previously autoclaved vials. The vials were capped and slanted and media was allowed to settle for 24 hours.

Surface-sterilized seed from each cultivar tested were aseptically placed upon appropriate media. Five seeds per vial and 10 vials per salt treatment were prepared for each cultivar tested. All treatments were then incubated at 21°C ± 2.0 under 1.63 $W m^{-2}$ of fluorescent lighting for 30 days at a 12 hour light/12 hour dark diurnal cycle.

The % germination of each cultivar for all treatments was determined during the experimental period. After the 30 day incubation period, individual plants for all treatments were extracted and data was recorded for mean ($\bar{x}$) leaf number, leaf length, root number and root length.

Results

The effects of increasing media salinity on % germination were similar for all cultivars (Table 1). The percent germination remained the same after 8 days of incubation for the Kentucky bluegrasses and 6 days for the red fescues and alkalaigrass. Total % germination, compared to control treatments, declined for all cultivars with the salt-tolerant red fescues and alkalaigrass being least affected. Within the

Table 1. Effect of NaCl on percent germination between cultivars of red fescue, Kentucky bluegrass and alkalaigrass

Percent germination					
	NaCl concentration (m*M*)				
Cultivar	17	43	85	170	Mean ($\bar{x}$) germination, %*
Checker (red fescue)	87	100	100	80	92
Fults (Alkalaigrass)	88	89	80	80	84
Dawson (red fescue)	86	86	71	70	78
Kentucky bluegrass					
Ram I	81	94	90	38	75
Adelphi	100	73	52	40	67
Nassau	79	93	74	14	65
Bensun	100	75	44	0	54
Baron	71	57	0	0	32

* Percent germination compared to control treatments representing 100% germination.

Table 2. Effects of NaCl salinity on leaf initiation between alkalaigrass and cultivars of red fescue and Kentucky bluegrass

Mean ($\bar{x}$) seedling leaf number						
	NaCl concentration (m*M*)					Mean % variation*
Cultivars	0.0	17	43	85	170	from control
Dawson (red fescue)	2.7a**	2.7a	2.9a	2.4a	2.3a	– 4.7
Checker (red fescue)	2.6ab	2.3ab	2.9a	2.1ab	2.3a	– 7.7
Fults (alkalaigrass)	2.1bcd	2.1abc	2.0b	2.0bc	1.8b	– 6.0
Kentucky bluegrass						
Adelphi	2.0bcd	1.8bc	2.1b	1.8bc	1.8b	– 6.3
Ram I	2.0bcd	2.1abc	1.7bc	1.7c	1.3c	– 15.0
Nassau	2.2abc	1.3c	1.5cd	1.0d	1.0d	– 45.5
Bensun	1.5d	1.8bc	1.3cd	1.0d	N.G.	– 31.7
Baron	1.8cd	2.0abc	1.2d	N.G.	N.G.	– 55.6

* % variation represents the mean of all NaCl treatments compared to control (0.0 NaCl).
** Mean followed by the same letter within columns are not significantly different at the $p = 0.05$ level.
N.G. = No germination.

K. bluegrasses, 'Ram I' and 'Adelphi' yielded the highest germination percentages.

Roots of developing seedlings grew into the semi-solid media and were easily extracted without damage when collecting whole plants for measurement.

Leaf initiation and leaf length

Cultivar variation within control treatments (0.0 NaCl) for both leaf initiation and leaf length was minimal (Tables 2 and 3). Increasing

Table 3. Effects of NaCl salinity on mean ($\bar{x}$) leaf growth between alkalaigrass and cultivars of red fescue and Kentucky bluegrass

Mean ($\bar{x}$) seedling leaf length (cm)

Cultivars	NaCl concentration, (m*M*) 0.0	17	43	85	170	Mean % variation* from control
Dawson (red fescue)	4.3ab**	4.8a	4.6a	4.0ab	3.4a	– 2.4
Checker (red fescue)	5.0a	4.0ab	4.4a	5.0a	3.3a	– 16.5
Fults (alkalaigrass)	4.4ab	3.8abc	3.4ab	3.2bc	3.2a	– 22.8
Kentucky bluegrass						
Adelphi	3.7ab	4.6a	3.4ab	2.1c	1.8b	– 19.6
Ram I	4.0ab	3.3abc	3.3ab	3.2bc	1.5d	– 30.0
Nassau	3.5b	2.4c	1.8c	1.2d	1.1b	– 53.6
Bensun	3.1b	3.6abc	2.5bc	1.5d	N.G.	– 38.8
Baron	3.6ab	2.8bc	1.6c	N.G.	N.G.	– 69.5

* % variation represents the mean of all NaCl treatments compared to control (0.0 NaCl).
** Means followed by the same letter within columns are not significantly different at the $p = 0.05$ level.
N.G. = No germination.

Table 4. Effects of NaCl salinity on root initiation between alkalaigrass and cultivars of red fescue and Kentucky bluegrass

Mean ($\bar{x}$) seedling root number

Cultivars	NaCl concentration,(m*M*) 0.0	17	43	85	170	Mean % variation* from control
Dawson (red fescue)	1.2b**	1.5ab	2.0ab	2.3a	1.1b	+ 10.4
Checker (red fescue)	2.2a	1.8a	2.2a	1.8b	1.4a	– 18.2
Fults (alkalaigrass)	1.2b	1.1c	1.1c	1.3bc	1.0b	– 6.3
Kentucky bluegrass						
Adelphi	1.3b	1.3bc	1.2c	1.0c	1.0b	– 13.5
Ram I	1.1b	1.1c	1.1c	1.0c	1.0b	– 4.5
Nassau	1.1b	1.0c	1.0c	1.0c	1.0b	– 9.1
Bensun	1.1b	1.1c	1.0c	1.0c	N.G.	– 29.5
Baron	1.4b	1.2bc	1.3bc	N.G.	N.G.	– 55.4

* % variation represents the mean of all NaCl treatments compared to control (0.0 NaCl).
** Means followed by the same letter within columns are not significantly different at the $p = 0.05$ level.
N.G = No germination.

NaCl levels reduced the extent of leaf production for all cultivars (Table 2). The percent reduction in leaf initiation as compared to control treatments was, in general, greatest for the K. bluegrass compared to the salt-tolerant red fescues and alkalaigrass with differences arising at the 43 m*M* NaCl concentration.

Results for mean ($\bar{x}$) leaf length closely reflected leaf initiation data (Table 3). In both cases, ranking cultivars from least to most affected by NaCl is as follows: 'Dawson' $<$ 'Checker' $\leqslant$ Fults $\leqslant$

Table 5. Effects of NaCl salinity on mean ($\bar{x}$) root length between alkalaigrass and cultivars of red fescue and Kentucky bluegrass

Mean ($\bar{x}$) seedling root length (cm)

	NaCl concentration (m*M*)					Mean % variation*
Cultivars	0.0	17	43	85	170	from control
Dawson (red fescue)	2.7ab**	2.6a	2.9a	2.3a	2.2a	– 7.5
Checker (red fescue)	2.1ab	2.2a	2.1b	2.8a	1.7b	+ 10.0
Fults (alkalaigrass)	2.5ab	2.3a	2.1a	2.3a	2.5a	– 8.0
Kentucky bluegrass						
Adelphi	2.3ab	2.4a	2.2b	1.6b	1.2c	– 19.6
Ram I	2.7a	2.3a	2.5ab	2.2a	1.2c	– 24.1
Nassau	2.5ab	2.0a	1.4c	1.3b	1.1c	– 42.0
Bensun	1.9b	1.8a	1.5c	1.2b	N.G.	– 40.8
Baron	2.5ab	2.0a	2.2c	N.G.	N.G.	– 58.0

* % variation represents the mean of all NaCl treatments compared to control (0.0 NaCl).
** Means followed by the same letter within columns are not significantly different at the $p = 0.05$ level.
N.G. = No germination.

'Adelphi' < 'Ram I' < Nassau ⩽ Bensun ⩽ Baron. As expected, the salt-tolerant red fescues and alkalaigrass were less affected than the K. bluegrass for each parameter. More importantly, these results show a consistent level of variability between the K. bluegrasses with 'Adelphi' and 'RamI' retaining the best overall NaCl tolerance.

Root initiation and root length

Root initiation increased for 'Dawson' red fescue between 17 and 85 m*M* NaCl concentrations (Table 4). Averaging data over all NaCl treatments for each cultivar than determining mean ($\bar{x}$) percent variations from controls had resulted in an overall increase in root production for 'Dawson'. All other cultivars had shown a reduction in root numbers with little variation occurring between 'Fults' and the K. bluegrasses 'Nassau', 'Ram I' and 'Adelphi'. Although 'Checker' retained the highest level of root initiation over most NaCl concentrations, reduction from control was higher than all other cultivars except 'Bensun' and 'Baron'.

An overall positive response of mean ($\bar{x}$) root length to NaCl treatments was observed for 'Checker' red fescue indicating that root extension is less salt sensitive than root initiation in this cultivar (Table 5). All other cultivars had shown a reduction in root length with differences becoming apparent at the 43 m*M* NaCl concentration. As expected, the salt-tolerant red fescues and alkalaigrass were less affected by NaCl than the K. bluegrasses. Variation among the

K. bluegrasses for root length was consistent with previous observations indicating 'Adelphi' and 'Ram I' as being more NaCl-tolerant than 'Nassau', 'Bensun' or 'Baron'.

Discussion

Ranking of species and cultivars for overall NaCl tolerance was highly consistent between germination, leaf initiation, leaf length, root initiation and root length measurements. The trend throughout this report ranks the salt-tolerant red fescues and alkalaigrass above the Kentucky bluegrass cultivars for NaCl tolerance. Harivandi *et al.*[2] and Skirde[8] report similar findings. Among the Kentucky bluegrasses, 'Adelphi' and 'Ram I' were shown to be consistently more NaCl tolerant than other cultivars tested. 'Ram I' has previously been shown to retain a relatively high level of tolerance towards $CaCl_2$ salinity[5]. However, the lack of information regarding salt tolerance among Kentucky bluegrass cultivars makes comparisons in methodology difficult.

Alkalaigrass ('Fults') and 'Dawson' red fescue were included in this study as "standards" since their high level of salt-tolerance is well documented[2,3,5]. Throughout this study 'Dawson', as expected, exhibited a very high level of NaCl-tolerance, even higher than 'Fults'. Alkalaigrass has previously been shown to be more salt-tolerant than 'Dawson'[2] and is also known to increase in tolerance and accumulate more Na and Cl- ions in shoots with increasing plant age. These results clearly indicate an important problem associated with short screening studies (as reported in this paper) where only seedling response is measured. Characterization of inherent salt-tolerance can only be accomplished with long-term field studies under a variety of different conditions. However, the ranking of species and cultivars within this study remained relatively constant between a number of different measurements and consistently reflected general differences in NaCl-tolerance already known to exist. Therefore, the results of this study may provide a useful basis for screening large numbers of genotypes or existing turfgrass cultivars for further salt tolerance studies.

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