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# Growth Responses of Tall Fescue (*Festuca arundinacea* Schreb.) to Salinity Stress

Y. Gao and D. Li
(Department of Plant Sciences, North Dakota State University, Fargo, ND, USA)

## Summary

Tall fescue is moderately tolerant to salinity with great potential for use as turfgrass under saline conditions. The objective of this study was to investigate the growth habit and nutrient uptake of tall fescue under salinity stress. Two tall fescue cultivars, 'Tar Heel II' and 'Wolfpack' were seeded in plastic tubes filled with washed silica sand. Salinity treatment was applied by adding NaCl and $CaCl_2$ (1:1 w/w) to the Hoagland solution in 16 g $L^{-1}$. Salinity resulted in an average tiller number reduction of 50 % compared to the control, with 'Wolfpack' showed 5 % more reduction than 'Tar Heel II'. Root/shoot ratio was not affected by salinity treatment ($P > 0.05$). The tiller appearance rate (TAR) (0.4 week$^{-1}$) and leaf appearance rate (LAR) (0.35 week$^{-1}$) of tall fescue plants under salinity stress were significantly lower than the TAR and LAR of control, 1.90 week$^{-1}$ and 1.0 week$^{-1}$, respectively. It appeared that TAR was more affected than LAR by the stress. Salt treatment also reduced the uptake of $K^+$ and $Mg^{2+}$, but increased the uptake of $Na^+$ and $Ca^{2+}$ in the shoots of tall fescue. Salt treatment did not increase $Ca^{2+}$ in the roots of tall fescue. In conclusion, maintaining $K^+$, $Ca^{2+}$ and $Mg^{2+}$ ionic balance in the shoots is important to improve the stress tolerance in tall fescue as indicated by a higher TAR.

**Key words.** carbonate – chloride – osmotic – production – sodium – sulfate

## Introduction

Tall fescue has increasingly been used as turfgrass since the 1950s resulting from breeding efforts to develop turf-type grass (BUCKNER et al. 1979). Physiological studies since the 1980s revealed many aspects of stress tolerance in tall fescue including heat, drought, shade, salinity, and wear (BEARD 1981). Tall fescue has moderate tolerance to soil salinity (LUNT et al. 1961). With improved genetic traits, the species has become the turfgrass of choice for medium or low maintenance uses (WATKINS et al. 2011) especially for the transition zone in the USA where extreme temperatures and drought predominate (FUNK and CLARKE 1989).

Salinity stress can be imposed on turfgrass when soil or water has elevated salt levels. Recycled water is increasingly used for irrigation to conserve potable water in many municipalities. Recycled waters, such as reclaimed municipal effluent and brackish drainage water typically have elevated salt levels (MIYAMOTO and CHACON 2006). There is a renewed interest in understanding how tall fescue growth and tissue mineral nutrient uptake are affected by salinity stress and their influence on turf quality. Salinity reduces turfgrass quality by decreasing shoot density (DEAN et al. 1996). The density of tall fescue is affected by tiller and leaf number. Tiller development depends largely on environmental conditions and cultural practices. Tall fescue tillering rate is limited by and negatively correlated to leaf appearance (TEMPLETON et al. 1961). Tillering rate is not affected by water soluble carbohydrates in the leaves, roots, or stem base tissues (ZARROUGH et al. 1984). However, shoot biomass is closely related to the tiller carbohydrate reserve and photosynthesis capacity of leaves (SMITH 1973). Usually three leaves per tiller are active at any time (ALBURQUERQUE 1967) and individual leaves are photosynthetically active for about 6 weeks during spring and summer (JEWISS and WOLEDGE 1967).

The detrimental effects of salinity on turfgrass can be attributed to toxicity of excessive $Na^+$, $Cl^-$, $SO_4^{2-}$, $CO_3^{2-}$, $HCO_3^-$, and $BO_3^-$, as well as nutrient imbalance and deficiency (BOWMAN et al. 2006). High levels of salinity disrupt ion homeostasis in plants by inhibiting the uptake of essential nutrients like K, Ca, N, while increasing the accumulation of Na and Cl (MARSCHNER 1995; ZHU 2001; LACERDA et al. 2003). As a consequence, cell division and elongation decline or stop completely (HASEGAWA et al. 2000). Salinity reduces the uptake and partitioning of N to leaves and increases the retention of N in the roots of tall fescue (BOWMAN et al. 2006).

Tall fescue nutrient uptake and growth rate are affected by salinity stress (LUNT et al. 1961; BOWMAN et al. 2006).

Nutrient uptake then affects biomass, tiller density and leaf number (BEARD 1985; PITMAN 1999). GROSS (1973) ranked tall fescue as a high Mg user among the cool-season grasses. The reported critical levels for Ca and Mg in grass are about 0.1 and 0.06 %, respectively (DEWIT et al. 1963; WILKINSON and LOWREY 1973). There is a lack of information regarding the mechanisms of salinity effects on tall fescue growth and nutrient uptake to guide turfgrass managers in managing tall fescue under saline conditions. The objective of this study was to investigate growth habit and nutrient uptake of two tall fescue cultivars with different salinity tolerance grown under saline conditions.

## Materials and Methods

The experiment was conducted in a greenhouse at North Dakota State University in 2009 and repeated in 2010. Two tall fescue cultivars, 'Tar Heel II' (salt tolerant) and 'Wolfpack'(salt sensitive) (WIPFF and ROSE-FRICKER 2003), were seeded in plastic tubes measuring 4 cm in diameter and 20 cm in depth filled with silica sand. The plants were maintained in a greenhouse with an average temperature of 25/15 °C (day/night), 14-h photoperiod, and an average photosynthetically active radiation of 400 µmol $m^{-2}$ $s^{-1}$. The sand growth medium was maintained at field capacity by watering twice a day until the seeds germinated, and then the seedlings were watered once a day. The seedlings were fertilized by watering with half strength Hoagland solution (HOTHEM et al. 2003) in 10 ml per tube twice a week. At the 2-leaf stage, the plants were thinned to one plant in each tube.

At the 4-leaf stage, 20 mL of full strength Hoagland solution was applied to each tube every other day. The salt treatment was applied by adding 16 g $L^{-1}$ of NaCl and $CaCl_2$ in equal amounts to the full strength Hoagland solution to reach an EC about 25 dS $m^{-1}$ (WIPFF and ROSE-FRICKER 2003). Plants that received Hoagland solution only were included as a control. The experimental design was a randomized complete block with three replicates.

On the day prior to the initiation of treatments and weekly thereafter, tiller number and leaf number on the primary shoot of the seedlings were recorded. A tiller was defined as one shoot with at least one fully developed leaf. A leaf with ligule and auricles exposed was considered as fully developed. Leaf appearance rate (LAR) (number of leaves $plant^{-1}$ $d^{-1}$) and tiller appearance rate (TAR) (number of new tiller $plant^{-1}$ $d^{-1}$) were calculated based on the leaf and tiller number data.

At the end of four weeks after treatment (WAT), plant height was measured from 10 plants in each treatment unit by measuring the distance from the growth medium surface to the tip of the youngest fully developed leaf on the primary shoot. At the end of each experiment, shoot biomass was harvested by clipping at the growth medium surface. Roots were harvested by washing the sand off on a 2-mm sieve with tap water and rinsed with de-ion water. The root and shoot dry mass were recorded after drying at 68 °C for 48 h. The dry tissues were ground to pass 0.178 mm for tissue nutrient analysis. The K, Ca, Mg, Na, and Fe of the tissues were analyzed using an AA7000w flame atomic absorption spectrophotometer (Beijing East & West Electronic Company, Beijing, China) following dry ashing in a muffle furnace at 490 °C for 8 h and digestion with 5 M aqua regia (GREWELING 1976; JONES et al. 1991).

The data were subjected to analysis of variance (ANOVA) using a mixed model in SAS 9.2 (SAS INSTITUTE 2008) with replication treated as a random variable. Treatment means were separated using Fisher protected least significant difference (LSD) at 0.05 probability level.

## Results and Discussion

A homogeneity test revealed a difference in the variances of 2009 and 2010. Therefore, the results from the two years are reported separately. Salt treatment reduced tiller number and leaf number compared to the untreated control in the 2009 and 2010 study (Table 1). Salinity resulted in an average tiller number reduction from 11 to 4.9; a 56 % reduction from the control ($P > 0.05$). Also, 'Wolfpack' showed 5 % more tiller number reduction than 'Tar Heel II' as a result of salt treatment. Plants treated with salts were shorter than the control in general, but was significant only for 'Wolfpack' in the 2010 study (Table 1). Plant height reductions under salinity stress also were reported in tall fescue and Kentucky bluegrass (ALSHAMMARY et al. 2004) as well as in warm-season grasses (MUSCOLO et al. 2003; CHEN et al. 2009). The different salinity response between two cultivars corroborates WIPFF and ROSE-FRICKER (2003).

Shoot and root biomass decreased by 58 and 63 %, respectively, in salt treatment as compared to the control (Table 2). No differences were detected between the two cultivars in shoot and root biomass changes compared to the control. Root/shoot ratio showed no differences between the salinity treatment and control, which may be explained by the positive quadratic regression relationship between root/shoot ratio and EC as reported by ALSHAMMARY et al. (2004). With a quadratic relationship, intermediate salinity levels may show higher root/shoot ratios, however, the ratio reduces to the control value at high salinity levels. Therefore, despite the reduction of shoot and root biomass due to salt effects, root/shoot ratio is not a reliable indicator in evaluation of salinity tolerance of cultivars.

The TAR and LAR of the plants in salinity treatment were lower than the control in both 2009 and 2010 as indicated by the slope of tiller number and leaf number changes over time (Fig. 1), where greater slope indicates a higher rate. The tiller appearance rate (TAR) (0.4 $week^{-1}$) and leaf appearance rate (LAR) (0.35 $week^{-1}$) of tall fescue plants under salinity stress were significantly lower than

Table 1. Growth features of two tall fescue cultivars as affected by salinity at the onset of 4-leaf seedling stage.

| Treatment | 2009 | | | 2010 | | |
|---|---|---|---|---|---|---|
| | Tiller number[a] | Leaf number[b] | Plant height[c] (cm) | Tiller number | Leaf number | Plant height (cm) |
| 'Tar Heel II' | 8.8 | 7.0 | 14.47 | 11.6 | 7.1 | 13.07 |
| 'Tar Heel II' + Salts | 5.8 | 6.5 | 14.13 | 3.8 | 5.7 | 11.71 |
| 'Wolfpack' | 10.5 | 7.2 | 13.70 | 13.0 | 7.2 | 15.39 |
| 'Wolfpack' + Salts | 6.4 | 6.5 | 13.25 | 3.5 | 5.3 | 12.13 |
| $LSD_{0.05}$ | 3.1 | 0.5 | 1.51 | 2.0 | 0.5 | 2.24 |

[a] Tiller number, total tiller number per seedling.
[b] Leaf number, number of leaves on the main shoot of the seedling.
[c] Plant height, measured to the tip of the youngest mutual leaf on the main shoot of the seedling.

Table 2. Biomass of two tall fescue cultivars as affected by salinity at the onset of 4-leaf seedling stage.

| Treatment | 2009 | | | 2010 | | |
|---|---|---|---|---|---|---|
| | Shoot (g) | Root (g) | Root/shoot | Shoot (g) | Root (g) | Root/shoot |
| 'Tar Heel II' | 27.5 | 9.73 | 0.35 | 37.7 | 16.31 | 0.44 |
| 'Tar Heel II' + Salts | 19.4 | 6.23 | 0.32 | 8.7 | 3.19 | 0.38 |
| 'Wolfpack' | 28.5 | 9.76 | 0.34 | 40.1 | 15.45 | 0.39 |
| 'Wolfpack' + Salts | 19.7 | 6.69 | 0.34 | 8.8 | 3.10 | 0.35 |
| $LSD_{0.05}$ | 2.3 | 1.28 | 0.05 | 3.4 | 1.18 | 0.08 |

the TAR and LAR of control, 1.90 week$^{-1}$ and 1.0 week$^{-1}$, respectively. In both years, TAR was more affected by stresses than LAR. Similar results were reported in a warm-season grass *Chloris gayana* Kunth (Cordoba et al. 2001). The decreasing leaf appearance rates in barley (*Hordeum vulgare* L.) and wheat (*Triticum aestivum* L.) showed an obvious change in slope, a break point between stage one and stage two after certain days of exposure to salinity (Harris et al. 2010). The LAR results from this study were similar to that reported by Harris et al. (2010), although development stage could not be clearly established as in barley or wheat. There is a direct link between reduced leaf number and tiller number in tall fescue under saline conditions. It is also noticeable that without salt treatments, 'Wolfpack' had higher leaf and tiller numbers than 'Tar Heel II', but under salt treatment, 'Wolfpack' showed more reduction than 'Tar Heel II'.

Salt treatment reduced uptake of $K^+$ and $Mg^{2+}$, but increased the uptake of $Na^+$ and $Ca^{2+}$ in the shoot in both 2009 and 2010 (Table 3). Iron content in shoot tissues was not affected by salt treatment. Shoot $Na^+$ increased by approximately 1.5 times, while $Ca^{2+}$ increased by 2 times in 2010 and 7 times in 2009 (Table 3). Since $Na^+$ and $Ca^{2+}$ was applied in a 1:1 ratio (W/W), it appeared that tall fescue shoot selectively took $Ca^{2+}$ under salinity stress. These results agree with those of Lunt et al. (1961) on 'Alta' tall fescue, and those of Torello and Rice (1986) on red fescue (*F. rubra* L.) and Kentucky bluegrass.

'Wolfpack' showed more increase of $Ca^{2+}$ and more decrease of $Mg^{2+}$ in leaf tissues than 'Tar Heel II' under saline conditions (Table 3) indicating that $Ca^{2+}/Mg^{2+}$ ratio may be used as an indicator of different salinity tolerance in tall fescue. In 2009, $Ca^{2+}/Mg^{2+}$ ratio of both cultivars was around 4.5 without salt treatment, and was 31.1 and 37.1 for 'Tar Heel II' and 'Wolfpack', respectively, in salt treatment. In 2010, $Ca^{2+}/Mg^{2+}$ ratio of the two cultivars was 4.7 and 4.9 without salt treatment, and was 18.9 and 25.5 for 'Tar Heel II' and 'Wolfpack', respectively, in salt treatment. Lower $Ca^{2+}/Mg^{2+}$ ratio seems to be related to better salt tolerance. The effects of salinity on mineral nutrients in leaves vary with species. For example, high exogenous $Na^+$ causes decreases of $K^+$, $Ca^{2+}$, and $Mg^{2+}$ in the leaves of Kentucky bluegrass (Qian et al. 2001), and decrease of $K^+$ concentration in the leaves of chewing fes-

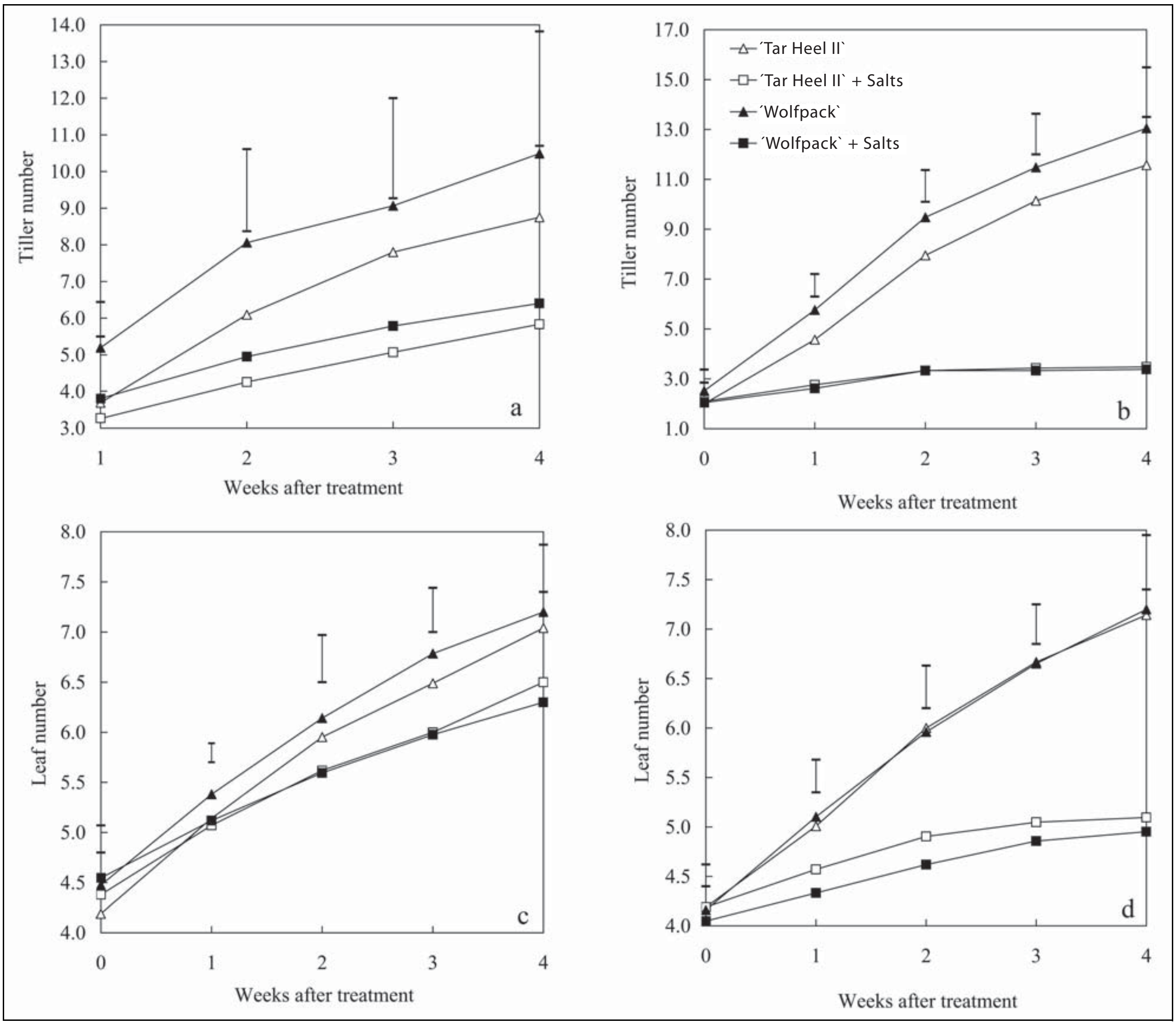


Fig. 1. Tiller and leaf appearance of two tall fescue cultivars irrigated with full strength Hoagland solution and Hoagland solution plus 16 g $L^{-1}$ NaCl and $CaCl_2$ in equal amounts at 4-leaf seedling stage. a) and c), results from 2009. b) and d), results from 2010. Vertical bars indicate least significant difference at 0.05 probability level at a given week after treatment.

cue (*F. rubra* L.) (Khan and Marshall 1981). On the other hand, Hu and Schmidhalter (1998) reported that $K^+$, $Ca^{2+}$ and $Mg^{2+}$ concentrations were increased by $Na^+$ in wheat.

The $K^+$ content in roots was not affected by salt treatment in 2009 but decreased in 2010 (Table 3). Salinity treatment increased the $Na^+$ uptake in roots by approximately 2.5 times compared with the control in both 2009 and 2010 (Table 3). Unlike the case in shoots, $Ca^{2+}$ content in roots did not increase in salt treatment. This may be the results of accelerated transport of $Ca^{2+}$ to the shoot under saline conditions, which might be involved in the mechanism of countering the toxic effect of $Na^+$. The $Mg^{2+}$ levels in salt treated roots decreased compared to the control although significant only in the 2010 study. The $Fe^{2+}$ content in roots also showed a general decreasing trend in response to salt treatment but was significant only for 'Wolfpack' in 2009 and 'Tar Heel II' in 2010.

Nutrient uptake of $K^+$, $Na^+$, $Ca^{2+}$, and $Mg^{2+}$ in the shoot under salinity stress may be affected by antagonism between the nutrients at the sites of uptake, the inhibitive effect of $Na^+$ on transport of $K^+$ and $Mg^{2+}$ in the xylem (Lynch and Läuchli 1984, 1985), and by $H^+$-ATPase activity and hormones in active transportation (Gronwald et al. 1990; Van Steveninck 1972). The understanding of nutrient uptake affected by salinity is further complicated by the different growth and development patterns of the leaf and tiller, and by the nutrient movement between tissues and organs. The complexity also is demonstrated by differences among species, cool-season vs warm-season, and glycophytes vs halophytes.

Table 3. Shoot and root tissue nutrients analysis (in mg $kg^{-1}$) of two tall fescue cultivars as affected by salt at the onset of 4-leaf seedling stage.

| | 2009 | | | | | 2010 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Treatment | K | Na | Ca | Mg | Fe | K | Na | Ca | Mg | Fe |
| **Shoot tissue** | | | | | | | | | | |
| 'Tar Heel II' | 615 | 275 | 995 | 219 | 525 | 554 | 359 | 1750 | 375 | 370 |
| 'Tar Heel II' + Salts | 578 | 416 | 5625 | 181 | 445 | 520 | 545 | 3409 | 180 | 397 |
| 'Wolfpack' | 604 | 294 | 1011 | 222 | 447 | 553 | 330 | 1829 | 375 | 356 |
| 'Wolfpack' + Salts | 579 | 421 | 6707 | 181 | 393 | 521 | 545 | 3441 | 135 | 377 |
| $LSD_{0.05}$ | 21 | 26 | 338 | 7 | NS | 5 | 41 | 442 | 5 | NS |
| **Root tissue** | | | | | | | | | | |
| 'Tar Heel II' | 566 | 933 | 4176 | 166 | 914 | 394 | 1059 | 8034 | 184 | 915 |
| 'Tar Heel II' + Salts | 581 | 2247 | 3357 | 154 | 795 | 378 | 2580 | 7965 | 160 | 804 |
| 'Wolfpack' | 570 | 953 | 3910 | 160 | 900 | 406 | 1143 | 8958 | 198 | 855 |
| 'Wolfpack' + Salts | 582 | 2330 | 3733 | 155 | 760 | 378 | 2593 | 8413 | 158 | 808 |
| $LSD_{0.05}$ | 14 | 122 | 710 | NS | 129 | 16 | 108 | 743 | 22 | 93 |

In conclusion, nutrient content in shoot and root tissues are differently affected under salinity stress. Since the nutrient uptake involves many steps and is very delicately regulated, it is important to study those essential elements and the function in stress-specific conditions, such as salinity. Results from this study show $K^+$, $Ca^{2+}$ and $Mg^{2+}$ are important in maintaining the ionic balance and salinity stress tolerance in tall fescue.

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Addresses of authors: Yang Gao and Deying Li (corresponding author), Department of Plant Sciences, North Dakota State University, Fargo, ND 58108, USA, e-mail: (corresponding author): deying.li@ndsu.edu.