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Saline effects on forage growth and quality

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SUMMARY – Birdsfoot trefoil (*Lotus corniculatus* L.) and tall fescue (*Festuca arundinacea* Schreb.) plants were grown hydroponically in pots (NFT system), with four NaCl levels: 0, 44, 87 and 174 mM (corresponding to 1, 4, 8 and 16 dS/m). Plants shoot dry weight, amino acids, proline and mineral content were analysed after 35, 78, 121 and 148 days of saline treatment. In the last sampling, dry weight was reduced in 44% and 77%, with respect to control, in birdsfoot trefoil and tall fescue, respectively, with higher salinity. At the first harvest, dry matter formation was correlated with K^+ (r=0.96) in the former and with Ca^{2+} (r=0.91) in the second species. The chloride content in dry matter increases with time of exposure and salinity level in both species, with a higher rate accumulation in birdsfoot trefoil. In tall fescue proline and sucrose contents were twice as high and well related with sodium (r=0.92) and chloride (r=0.98), respectively.

Keywords: Salinity, *Lotus corniculatus*, *Festuca arundinacea,* growth, quality.

RESUME – *"Effets de la salinité sur la croissance et la qualité des fourrages". Nous avons étudié la croissance de plantes de lotier corniculé (*Lotus corniculatus *L.) et de fétuque élevée (*Festuca arundinaceae *Schreb.) utilisant un système hydroponique (NFT) avec quatre niveaux de NaCl 0, 44, 87 et 174 mM (correspondant à 1, 4, 8 et 16 dS* m^{-1}*). Le poids de la matière sèche et la concentration en sels minéraux des parties aériennes, ont été déterminés après 35, 78, 121 et 148 jours de traitements salins. À la fin de l´expérience les poids de la matière sèche des plantes traitées avec 174mM NaCl ont subi une réduction de 44 et 77% respectivement en lotier corniculé et fétuque élevée. Au début de l´expérience (à la première récolte) nous avons constaté une corrélation entre le poids de la matière sèche et la teneur en* K^+ *(r=0,96) dans la première espèce et de* Ca^{2+} *(r=0,91) dans la dernière. La teneur en chlorure dans la matière sèche augmente avec le temps d'exposition et la teneur en NaCl de la solution nutritive dans les deux espèces, à un taux plus élevé dans le lotier corniculé. Dans la fétuque élevée les teneurs en proline et en sucres solubles sont 2 fois plus élevées et corrélées au sodium (r=0,92) et aux ions de chlore (r=0,98).*

Mots-clés : *Salinité,* Lotus corniculatus *L.,* Festuca arundinaceae *Schreb., croissance, qualité*.

Introduction

The ability of plants to adapt to environmental stress conditions is genetically determined, and includes biochemical, biological and physiological components.

In response to increased salinity plants have to lower the internal water potential to avoid wilting by taking up water. This is achieved by salt ion uptake and/or by synthesis of organic osmoprotective substances, like carbohydrates, free amino acids.

The aim of this study was to evaluate salt stress by means of biomass production and composition in two forage species for utilization in pastures in marginal and/or salt affected agricultural land or irrigated with brackish or saline water.

Materials and methods

Birdsfoot trefoil (*Lotus corniculatus* L. cv. San Gabriel) and tall fescue (*Festuca arundinacea* Schreb. cv. Demeter) plants were grown hydroponically in pots with sand using a NFT system, with half-strength Hoagland's solution (Hoagland and Arnon, 1950) supplemented with 0, 44, 87 and 174 mM NaCl (corresponding to A -1, B - 4, C - 8 and D -16 dS m^{-1}) and three replications. Salinity was imposed to 41 days-old plants in four increments 48 hours apart. The pH and conductivity of nutrient solutions were measure and adjusted as needed three times a week and the volume topped up with deionised water as required.

Harvests were made after 35, 78, 121 and 148 days of saline treatment and consist of cutting the shoot of six plants per pot. Samples were dried at 80℃ for 48 hours and dry weight calculated. Soluble protein and carbohydrates in plants were quantified only in the first sampling and mineral content in all harvests.

Chloride was determined in aqueous solution after incubation at 45℃ for one hour and quantified with a specific ion electrode. Nitrate was determined according to Cataldo *et al.* (1975) and mineral content were analysed by atomic flame spectrophotometry (Anonimous, 1976). Soluble protein was estimated by the method of Lowry *et al.* (1951) using BSA as a standard. Total and reduced soluble sugars were determined by the method of Somogyi (1952). Total free amino acids (Spies, 1957) and proline (Bates *et al.*, 1973) were quantified in shoot extracts using ninhydrin reagent and a standard curve was prepared with L-glycine and L-proline, respectively.

Statistical analysis was made using multiple linear regression with SPSS package and the Stepwise method to adjust the models. The level of significance was $P < 0.001$.

Results and discussion

The initial response of dry weight to salinity was similar in both species and there was a high correlation with K^+ (0.96) in *L. corniculatus* and Ca^{2+} (0.91) in *F. arundinacea*. This reduction was due to the osmotic effect of the salt outside the roots. The production of dry matter (Fig. 1) decreased in *L. corniculatus* after 148 days of salinity with 16 dS m^{-1}, but in *F. arundinacea* the effect of this salinity started earlier. In the last sampling dry weight was reduced in 44 and 77%, with respect to control, in birdsfoot trefoil and tall fescue, respectively. Salinity of 4 dS m^{-1} stimulated dry matter production in tall fescue.

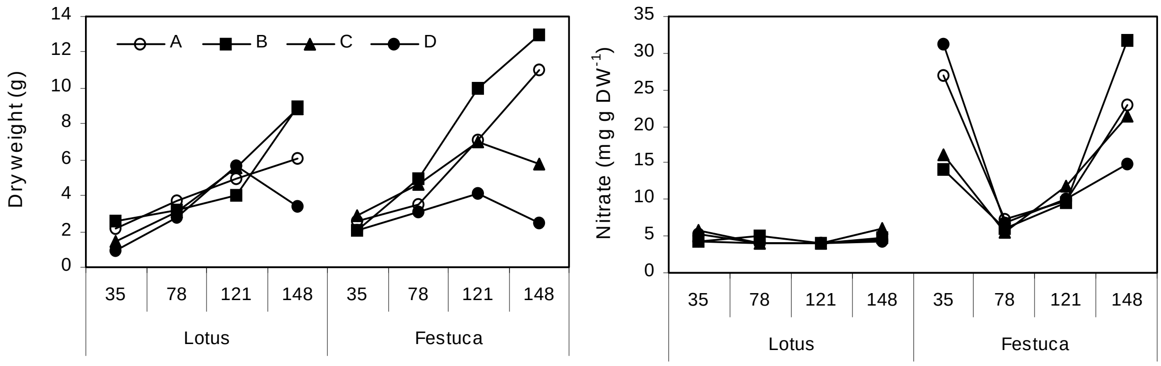

Fig. 1. Shoot dry matter production and nitrate content in *L. corniculatus* and *F. arundinacea* after 35, 78, 121 and 148 days of growth in four salt levels: A-1, B-4, C-8 and D-16 dS m^{-1}.

The assimilation of nitrate (Fig. 1) in birdsfoot trefoil was not affected by salinity. This may be due to higher nitrate assimilation in roots than in stems and leaves of Lotus (Márquez *et al.*, 2005). An adaptation that allows nitrate assimilation primarily in roots may be of importance for forage legumes as a way to overcome the loss of photosynthetic tissue due to grazing animals.

In tall fescue, nitrate assimilation decreased with salinity. The higher content was reached after 35 and 148 days of salinity. The assimilation in the last harvest may be associated to the rise of growth at environmental conditions (July).

Accumulation in shoot of sodium and chloride increased with salt level and time of treatment much more in birdsfoot trefoil than in tall fescue (Fig. 2), while water content, related to dry weight (data not shown), decreased in both species. As a result, the concentration of sodium in birdsfoot trefoil achieved values higher than those in nutrient solutions. This ion-excess effect of salinity (Greenway and Munns, 1980) is responsible for the growth reduction in long-term. In the first harvest we found that the sucrose content (data not shown) was highly correlated with Na^+ in birdsfoot trefoil (r=0.96) and tall fescue (r=0.73) and with Cl^- (r=0.98) in both. With salinity, sucrose content in tall fescue achieved twice the value in birdsfoot trefoil. Accumulation of sucrose in the shoot increases dry matter digestibility (Brown *et al*., 1963) and forage quality.

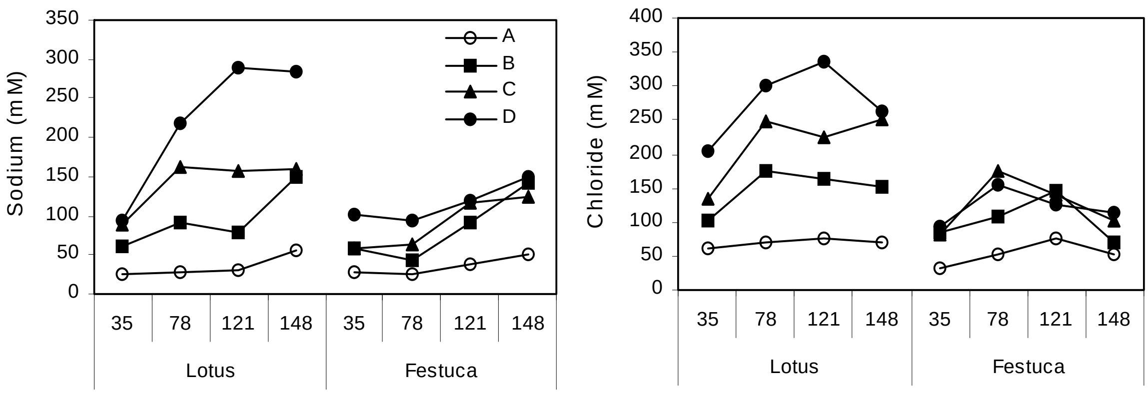

Fig. 2. Effect of salinity on shoot sodium and chloride content in both species. Time and salt levels as in Fig.1.

Amino acids content increased in older birdsfoot trefoil plants while in tall fescue the salt effect was noticed after 35 days of treatment (Fig. 3). In this first harvest there was a high correlation between Cl^- (r=0.96), Na^+ (r=0.84) and sucrose (r=0.90) in tall fescue, which can be attributed to an osmotic response.

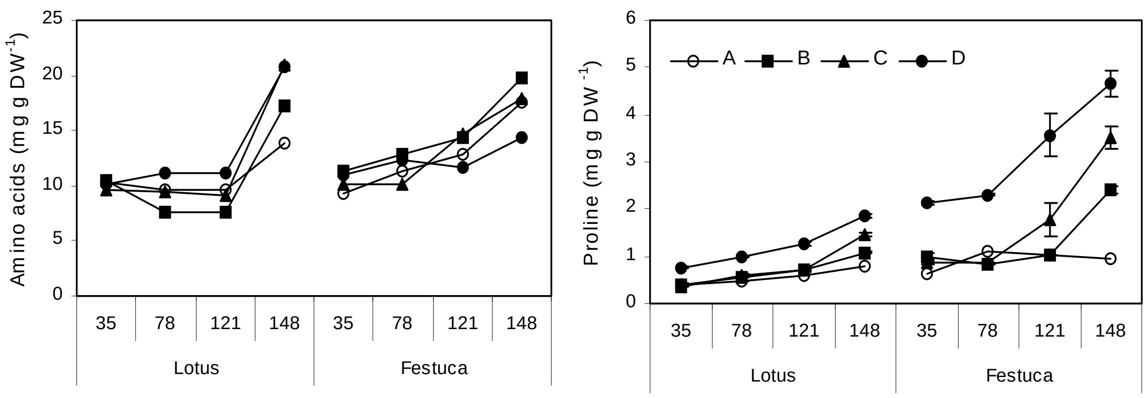

Fig. 3. Effect of salinity on shoot amino acids and proline content in both species. Time and salt levels as in Fig.1.

Compared to control, salt-treated plants had a 30% decrease in birdsfoot trefoil, whereas in fescue had a 20% increase in protein content after 35 days.

Proline content (Fig. 3) was significantly higher with 16 dS m^{-1} in both species, but in tall fescue there was an increase with 8 dS m^{-1} at the third and fourth harvest and with 4 dS m^{-1} in the last harvest. The lower levels of proline detected in the shoots don't exclude a potential role of nitrate nutrition in the osmotic response in the roots of birdsfoot trefoil, as it was verified in other plant species (McIntyre, 1997).

Table 1. Coefficients of the models adjusted for dry matter production and some quality parameters and correspondents coefficients of determination R^2

	Dry weight	Nitrate	Chloride	Amino acids	Proline
	(mg)		(mg g DW^{-1})		
Species	3.436	10.882	14.554	4.947	-
Salt	5.383	-	36.186	2.251	-0.831
Time	3.183	-16.401	15.995	-5.067	-0.590
Species x Salt	-0.916	-	-8.972	-1.352	-
Species x Time	-	-	-	-	-0.126
$Salt^2$	-0.684	0.338	-2.492	-	0.167
Salt x time	-0.526	-0.754	-	-	-0.122
$Time^2$	-	3.714	-3.111	1.475	0.149
R^2	0.759	0.815	0.853	0.869	0.911

Conclusions

Salinity affected the growth and composition in different way in both species. The high salt content in shoot makes birdsfoot trefoil less salt tolerant than tall fescue and it was the most sensitive component of forage quality. It seems that in birdsfoot trefoil the energy used for regrowth came mainly from N compounds, while in tall fescue it came from sucrose accumulated in roots. All these compounds may also play a role in osmoregulation.

Especially in tall fescue proline accumulation may play an important role in NaCl response of plants since the beginning of the experiment at the higher concentration.

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