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Mineral composition of lucerne (*Medicago sativa*) and white melilot (*Melilotus albus*) is affected by NaCl salinity of the irrigation water

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Most glycophytic forage legumes exhibit large reductions in biomass in response to saline growing conditions, although differences among species in salt tolerance exist. As several plant metabolic processes are affected by salinity, the nutritive value of these species to animals may be altered. Mineral concentration is an important component of plant nutritive value, and was evaluated in lucerne and white melilot under salt concentrations of 0, 55 and 110 mM NaCl in a glasshouse. Lucerne consistently accumulated higher concentrations of Na and Cl than white melilot. Calculated NaCl concentrations per unit DM reached 125 g/kg in lucerne and 39 g/kg in white melilot when irrigated with water containing 110 mM NaCl. In leaves of both species the Ca and Mg concentration decreased ($P < 0.0001$) with increasing salinity level, and there was an interaction ($P < 0.0001$) for these elements in which lucerne stems had decreases. The P concentration increased ($P < 0.0001$) in both species and the K concentration was affected only in lucerne stems. Among trace elements, Zn and Fe had marked decreases. Leaf and stem Zn concentration decreased in both species and Fe decreased more in leaves than in stems, but most analyzed microelements remained at acceptable levels for animal production. Detrimental changes in the mineral composition of forage species in response to salinity can be minimized by selecting appropriate species, such as white melilot. This may lead to improved animal production if biomass production, voluntary animal intake and digestibility can be maintained.

1. Introduction

A number of strategies have been considered in attempting to achieve economic returns from the increasing area of salt affected land in agricultural regions. One of them is implementation of agricultural systems where salt tolerant plant species are used as a source of forage for grazing livestock (Masters et al., 2001, 2005a; Barrett-Lennard, 2003). Most of these plant species are halophytes, which have limitations as animal feeds such as a high salt concentration and secondary compounds (Masters et al., 2007). High salt intake reduces feed intake and feed retention time in the rumen due to an accelerated flow of fluids, and can cause mineral imbalances, especially in Ca and Mg (Moseley, 1980; Godwin and Williams, 1986; Garg and Nangia, 1993). For instance, in sheep a drop in production is accentuated when dietary content of NaCl is higher than 0.1 kg/1.0 kg DM (Jackson et al., 1971; Moseley and Jones, 1974; Masters et al., 2005b). Therefore, productive forage

Abbreviations: EC, electrical conductivity; TDS, total dissolved solids; CP, crude protein; DM, dry matter; Sa, salinity; Fo, forage species.

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Table 1
Characteristics[a] of the rainwater and the irrigation solutions (mean ± SD).

	pH	EC (dS/m)	Salinity (‰)[b]	TDS[c] (mg/L)
Rainwater	7.2 ± 0.33	0.12 ± 0.015	0.1 ± 0.00	57 ± 7.9
Nutrient solution 0 mM of NaCl	6.6 ± 0.10	1.62 ± 0.188	0.8 ± 0.10	796 ± 96.9
Nutrient solution 55 mM of NaCl	6.6 ± 0.05	7.24 ± 0.611	3.9 ± 0.35	3870 ± 375.3
Nutrient solution 110 mM of NaCl	6.6 ± 0.07	11.60 ± 0.800	6.5 ± 0.61	6420 ± 604.7

[a] Measurements were with a pH meter ATi Orion and an EC meter ORION and are the average of 7 samples.
[b] ‰ means parts per thousand.
[c] TDS means total dissolved solids.

species with low salt concentrations are essential in order to improve animal production and to avoid a reliance on low NaCl supplementary feedstuffs. Some species of legumes possess some degree of tolerance to salinity, with lucerne being the most widely studied in this regard (Rejili et al., 2008; Bao et al., 2009; Wang and Han, 2009).

A point to consider in forage species for use on saline land is their nutritive value to the animal, including the mineral concentration which varies under salinity stress as a consequence of processes related to ion movement, such as competition, substitution, exclusion, accumulation and compartmentation. Thus, in plants exposed to salinity stress, the concentration of some minerals can become excessive and others can fall into marginal or deficient concentrations either for the plants or for the animals which eat them. Such information is particularly scant for legume forage species. Our research focuses on two glycophytic legumes species known to differ in tolerance to salinity, being lucerne (*Medicago sativa* L.) and white melilot (*Melilotus albus* Medik.) exposed to relatively long periods of salinity with the expectation that the mineral fraction of the species will change and, for some minerals, the concentration will fall outside the range of acceptable levels for animal production when the plants are exposed to high concentration of NaCl in the irrigation water.

2. Materials and methods

Two forage legume species were chosen according to their different tolerances to salinity, being *Melilotus albus* cultivar "El Domador", a biennial species considered moderately salt tolerant, and *Medicago sativa* cultivar "Eureka", a perennial species considered moderately sensitive to salinity.

The plants were grown with saline irrigation water using NaCl as the main salt, and sampled over a period of 8 mo at times when the two species were at similar phenological stages to facilitate comparisons at similar stages of maturity.

2.1. Establishment

The growing substrate was comprised of sterilized coarse washed sand and peatmoss in the ratio 1.6:1 by volume, fertilized with 0.5 g of calcium hydroxide, 0.9 g of calcium carbonate and 1.0 g of Nitrophoska 12-5-14 per liter of mixture. The substrate had a pH of 6.8, salinity ($EC_{1:5}$) 621 μS and total dissolved solids of 299 mg/l, with measurements taken at 25 °C. A total of 864 free draining nursery tree pots with dimensions of 50 mm × 50 mm × 120 mm (*i.e.*, 200 ml effective capacity) were filled with substrate and arranged in free-draining trays with capacity for 36 pots which composed each experimental unit.

Before sowing, white melilot and lucerne seeds were inoculated with Alfalfa Group commercial *Rhizobium* (Nodulaid™ from Becker Underwood Pty. Ltd. Somersby, New South Wales, Australia). Three seeds were sown in each pot and, 5 d after emergence, plants were thinned to one seedling. Trays were located in a glasshouse equipped with automatic cooling system at Roseworthy Campus, The University of Adelaide, in Adelaide, South Australia.

The experiment was conducted mainly during winter and spring (sown 1 February 2004) and no shade cloth was used after the first 60 d of establishment. Daily temperature for the experiment averaged 16 °C ± 4.5.

2.2. Nutrient solution

The base nutrient solution consisted of quarter strength Hoagland's solution, except for Ca and Mg which were at full strength, according to the protocol for forage production under salt stress (Smith, 1996). All fertilisers used were analytical reagent grade and their concentrations in the irrigation solution were: 0.25 mM KH_2PO_4, 1.25 mM KNO_3, 5.0 mM $Ca(NO_3)_2{\cdot}4H_2O$, 2.0 mM $MgSO_4{\cdot}7H_2O$, 11.3 μM H_3BO_3, 2.3 μM $MnCl_2{\cdot}4H_2O$, 0.2 μM $ZnSO_4{\cdot}7H_2O$, 0.08 μM $CuSO_4{\cdot}5H_2O$, 0.004 μM $(NH_4)_6Mo_7O_{24}{\cdot}4H_2O$ and, 30.0 μM Fe–EDTA. Stock solutions were prepared with deionised water, and the irrigation solution with rainwater which was filtered through a carbon filter. Salt treatments imposed were 0, 55 and 110 mM NaCl and Table 1 shows the characteristics of the irrigation solutions and rainwater as a complement of the concentration in mM applied.

Rainwater irrigation was maintained for 17 d after germination, followed by 4 d of irrigation with nutrient solution and, at day 26, salt treatments were gradually imposed until the desired concentrations were reached. The frequency of watering was once every 2 d for the first 10 d after germination and post-harvest and then once per day, always allowing free drainage of the solution to avoid salt accumulation in the substrate. Irrigations occurred by placing a copper pipe trident in the pot