A critical analysis of osmotic and ionic effects of salinity in two barley cultivars

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Abstract

Salinity is an important constraint to crop productivity in many agricultural areas, especially in arid and semi-arid regions. Despite intensive research on plant physiological responses to salinity, the relative importance of ion excess and water deficit to yield reduction under field condition remains unclear. This may be further complicated in soil-grown plants where the effect of soil physical properties may interact with the soil solution to determine the soil water potential and water uptake. A factorial experiment comprising different concentrations of NaCl, $CaCl_2$ and concentrated nutrient solution was conducted using two varieties of barley (*Hordeum vulgare*) in a completely randomised design with six replicates. Addition of NaCl or $CaCl_2$ strongly inhibited plant growth by up to 60%, but there was no significant difference between the effects of NaCl and $CaCl_2$.The concentrated nutrient solution also reduced the growth of the plants. The Na^+ concentration of plant tissues increased with soil EC, but Clipper accumulated less Na^+ than Sahara. Dry matter production of both cultivars declined as tissue Na^+ increased with Clipper being less tolerant to high Na^+ than Sahara. Photosynthesis rate declined as soil EC increased, but there was no significant difference in the responses to NaCl, $CaCl_2$ or to concentrated nutrient solution. In conclusion, growth of the barley was reduced by salinity primarily due to an osmotic stress following by excess ionic toxicities over the time. Sahara was more tolerant to induced salinity at all levels than Clipper. Na^+ exclusion did not always reflect salt tolerance. Osmotic stress is the predominant limiting factor in terms of plant growth.

Key words

Sodium toxicity, chloride toxicity, nutrient solution, salt tolerance

Introduction

Salinity is an important factor that reduces crop productivity in many agricultural areas, especially in arid and semi-arid regions. Salinity reduces plant growth through both non-specific, or osmotic effects and ion-specific mechanisms. While Munns et al (1995) proposed a two-phase model of salt injury, where growth is initially reduced by osmotic stress and then by Na^+ toxicity, it is difficult to assess with any confidence the relative importance of the two mechanisms to yield reduction because they overlap. Additionally, this may be further complicated in soil-grown plants where the effect of soil physical properties may interact with the soil solution to determine the soil water potential and water uptake. The objectives of this research were to evaluate the mechanism of salt tolerance in barley and quantify the injury of seedlings suffered under NaCl and $CaCl_2$ stress employed.

Materials and Methods

A factorial experiment examined the effect of different concentrations of NaCl (25, 50, 75 and 100 mM), $CaCl_2$ (12.5, 25, 37.5 and 50 mM) and modified concentrated Hoagland nutrient solution (2X, 4X and 8X strength generated from macro elements) on the growth of two varieties of barley (*Hordeum vulgare*), Clipper and Sahara. The nutrient solutions were used to examine the effect of osmotic stress independently of Na^+ or Cl^- toxicity. The experiment was a completely randomised design with six replicates. It was carried out in a greenhouse in which day temperatures ranged from 25 to 30 ^{0}C and night temperatures were from 15 to 20 ^{0}C. Quantum flux peaked at 350 mmol m^{-2} s^{-1} at midday. The surface 15 cm layer of a loamy soil from Roseworthy, SA, was collected, air-dried, ground to pass through a 5-mm mesh screen, and thoroughly mixed. Five barley seeds were sown in each pot and thinned to one per pot 3 days after emergence. The salt treatments were added with irrigation water to bring up the moisture level to field capacity. Non-destructive measurements of plant growth were made 2-3 times per week using a LemnaTec 3D plant scan analyser. Logistic curves were fitted to the data and the instantaneous growth rates calculated from the slope of the curve. Plant tissues were analysed for nutrients by Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES). Gas exchange studies were carried out using a LI-COR 6400 portable gas exchange system. Leaf temperature was maintained at 25^{0}C, light intensity was set at 500 ?molm^{-2} s^{-1} with and the CO_2 was set at 400 ?mol mol-$^{-1}$. Leaf to air VPD was maintained at 1.1 KPa. Plants were harvested after 50 days of growth. At the completion of the experiment, soil samples were taken and the electrical conductivity (EC) was measured on a 1:5 soil:water extract.

Results

Addition of NaCl or $CaCl_2$ strongly inhibited plant growth, but there was no significant difference between NaCl and $CaCl_2$ (Fig 1a). For example, 100mM of NaCl or 50mM of $CaCl_2$ decreased shoot dry weight by 60%, compared to the control treatment. The addition of the most concentrated nutrient solution (8X) resulted in a soil EC that was approximately equal to that of 35 mM NaCl, but overall the response to EC generated by the nutrient solutions was not significantly different to that of the salt solutions. There was a significant reduction in dry weight from different levels of induced salinity (P<0.05) but the effect of the two salts and between varieties were not significant.

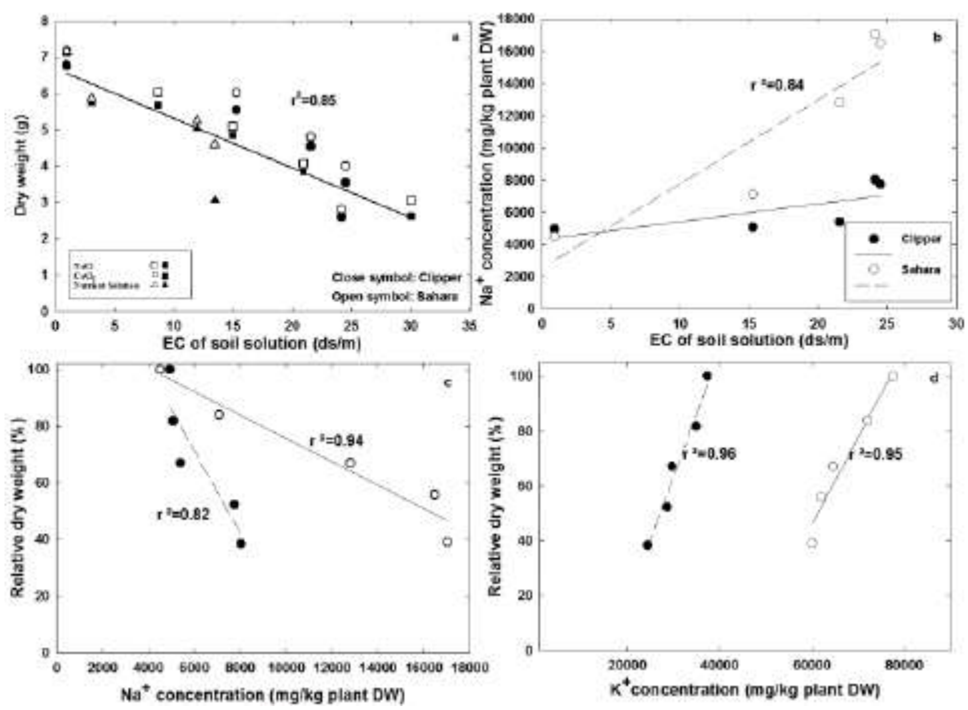

Figure 1. The effect of different levels of salinity on a) dry matter yield (g pot^{-1}) generated by NaCl, $CaCl_2$ and concentrated nutrient solution b) the relationship between Na^+ concentration in plant and EC of soil solution, c) the relationship between Na^+ concentration in the shoot and relative shoot dry matter and d) the relationship between the K^+ concentration of shoot and relative shoot dry matter production after 7 weeks of growth at 25, 50, 75 and 100 mM NaCl. Values are means (n=6 for a and n=3 for b, c and d).

The Na^+ concentration of plant tissues increased with soil EC (Fig 1b), but Clipper accumulated less Na^+ than Sahara. Plant Na^+ concentration increased by 62% in Clipper and 280% in Sahara when 100mM of

NaCl was applied compared to the control treatment (Fig 2b). Dry matter production of both cultivars declined as tissue Na^+ increased with Clipper being less tolerance to high Na^+ than Sahara. Increased salinity reduced (P<0.05) K^+ concentration by 34% in Clipper and 22% in Sahara (Fig 1d). Clipper had lower concentrations of K^+ than Sahara.

Increasing the level of salinity, either from NaCl or $CaCl_2$, significantly reduced leaf area over time and there was no significant difference between the sources of salinity (Fig 2). After 40 days of salt stress leaf area was reduced by 32 and 30% in Clipper and by 25 and 15% in Sahara grown at 100 mM NaCl and 50 mM $CaCl_2$ respectively. The concentrated nutrient solution reduced the green leaf area of Clipper to levels similar to those of the salt treatments, but the reduction in Sahara was not as great as the salt treatments.

The reduction in growth started at day 10 for both varieties but from day 25 settled down to a reduced steady rate in Clipper. NaCl reduced the rate of growth more than $CaCl_2$ in both varieties. With an increase from 25 to 100 mM NaCl and 12.5 to 50 mM $CaCl_2$, growth decreased 136 and 52% in Clipper and 17% and 30% in Sahara respectively after 30 days of growth. Decreases in plant growth with increasing salinity resulted in a lower net accumulation of leaf area (Fig. 2).

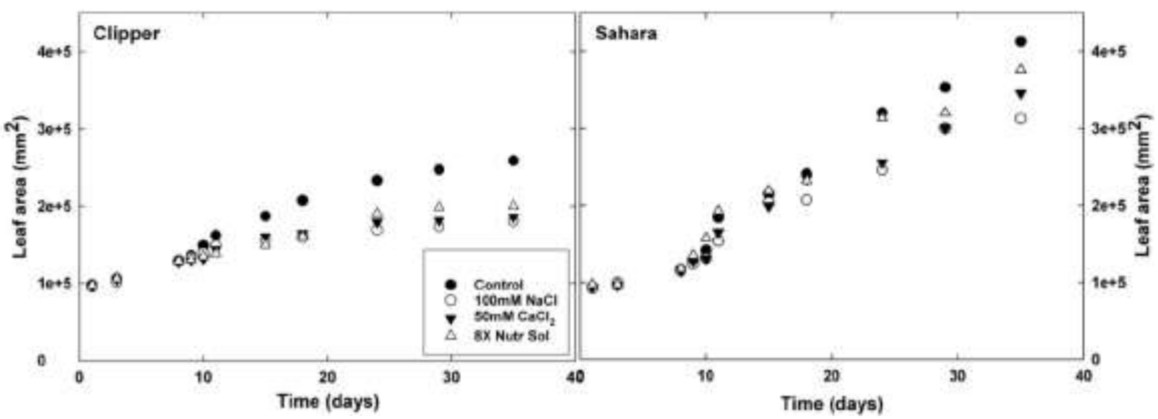

Figure 2. Changing in total leaf area over time of two varieties of barley at 100mM NaCl, 50mM $CaCl_2$ and concentrated nutrient solution. The leaf area was measured non-destructively using a LemnaTec 3D plant scan analyser. Values are means (n=6).

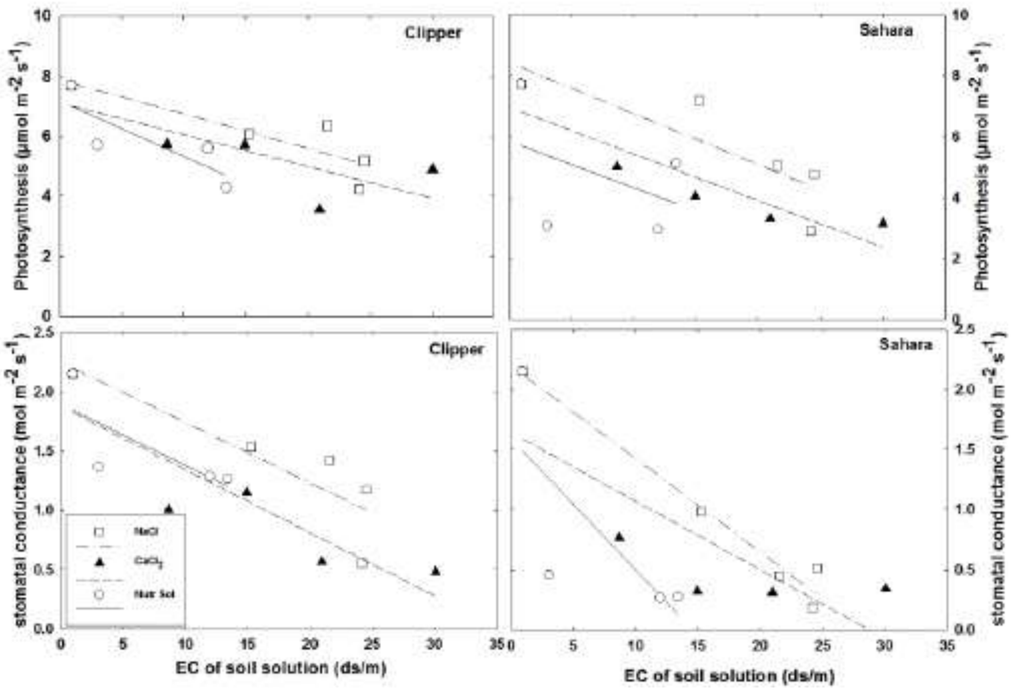

Figure 3. Effect of different salinity levels generated by NaCl, $CaCl_2$ and concentrated nutrient solution on photosynthetic parameters at 48 days after sowing for two varieties of barley. Values are mean (n=4).

Photosynthesis rate declined as soil EC increased, but there was no significant difference in the responses to NaCl, $CaCl_2$ or to concentrated nutrient solution (Fig 3).

Discussion

The solution electrical conductivity was the most important source of variation in growth irrespective of whether this was caused by NaCl, $CaCl_2$ or concentrated nutrient solution. This was also observed in the changes in leaf area where the responses to NaCl, $CaCl_2$ and nutrient solution were similar, especially in Clipper. The osmotic potential of the soil solution decreases as the EC increases and therefore the level of osmotic stress significantly increased as the concentration of NaCl, $CaCl_2$ or nutrient solution increased. Shoot Na^+ increased as the concentration of the NaCl increased, but despite this the dry matter response to EC in the NaCl treatment was the same as that observed with the addition of $CaCl_2$ or concentrated nutrient solution. This suggests that the dry weight reduction was mostly due to osmotic stress.

Sahara was less able to exclude Na^+ than Clipper. It accumulated Na^+ to concentrations up to 3 times higher than Clipper but produced higher biomass at a given Na^+ concentration, indicating it had a greater level of tolerance to high tissue concentration of Na^+ than Clipper. Ion accumulation and osmotic stress may not necessarily be independent phenomena because the degree of Na^+ exclusion may influence the osmotic potential near the root surface. Vetterlein *et al.* (2004) found that the osmotic potential of the rhizosphere was lower in a high Na^+ excluding line of maize and this caused a greater osmotic stress in the high excluding line.

The capacity of the plant to increase the leaf area depends on both its capacity to avoid the increase in ion concentration in leaf tissues as on the maintenance of higher rates of leaf production than of leaf death (Munns and Termaat 1986). When cells reach maximum salt sequestering capacity, salt concentration starts to rise in the cell walls, preceding cell death by dehydration, or rises in the cytoplasm causing toxicity (Munns and Passioura 1984). The reduction in photosynthesis can be due to either stomatal or non-stomatal factors (Araus *et al.* 2002, El-hendawy *et al.* 2005).

In conclusion growth of the barley was reduced by salinity primarily due to an osmotic stress following by excess ionic toxicities over the time. While Sahara had significantly greater concentrations of Na^+ in the leaves, it showed a higher level of tissue tolerant compared to Clipper and it was less sensitive to osmotic stress. The greater Na^+ exclusion of Clipper did not confer greater salinity tolerance. It is possible that exclusion of Na^+ reduced the growth of Clipper compared to Sahara, due to accumulation of Na^+ in the rhizosphere. Osmotic stress is the predominant limiting factor in terms of plant growth.

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