SALINITY - CALCIUM INTERACTION IN WHEAT AND BARLEY PLANTS

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ABSTRACT

Salinity-calcium interactions, which have been shown to be important in plants grown in dryland saline soils of the Farm of Agricultural Research Station, Sabahia, Alexandria, were studied in two species differing in salt tolerance. In solution culture, wheat showed a greater reduction in growth and a higher incidence of foliar Ca deficiency symptoms than barley when grown under $MgSO_4$ or Na_2SO_4 plus $MgSO_4$ salt stress. Amendment of the saline solution with Ca to increase the Ca/(Na+Mg) ratio ameliorated the effects of salt, but more so in wheat than in barley. At least part of the difference in salt tolerance between the two species must therefore relate to species differences in the interaction of salinity and Ca nutrition. The greater response of wheat to Ca was not due to a lower Ca status in leaf tissue; on the contrary, although Ca amendments improved tissue Ca/(Na+Mg) ratios in both species, salinized wheat had equivalent or higher calcium content, and higher Ca/(Na+Mg) ratios than did barley. The higher calcium requirement of wheat is apparently specific to a saline situation; at low salinity, wheat growth was not reduced as extensively as that of barley as Ca/(Na+Mg) ratio was decreased. High night-time humidity dramatically improved wheat growth under saline conditions, but increasing the calcium concentration of the saline solution had no effect on growth in the high humidity treatment. Membrane leakage from leaf tissue of wheat grown under saline conditions was increased compared to tissue from non-saline plants. Plants grown in Ca-amended saline solutions showed no increase in membrane leakage. These results confirm the importance of calcium interaction with salinity stress, and indicate difference in species response.

Keywords: Wheat, barley, magnesium, calcium, salinity, humidity, membrane leakage.

INTRODUCTION

Despite a tendency for salt exclusion, the mineral nutrition of non-halophytes is influenced by the presence of salt, often as a consequence of ion interactions. For example, Na-K and Cl-NO_3 interactions are well documended (Abdel-Sabour, 1999; Abdel-Aziz and Reda, 2000 and Greenway and Munns, 1980). Interactions of Ca with other ions at high salinity are also known to occur and low Ca/Na concentration (or activity) ratios resulted in reduced growth and in some cases tissue Ca deficiencies (Gawish *et al*., 1999; Grieve and Maas, 1988; Kent and Läuchli, 1985; Maas and Grieve, 1987 and Mahammed *et al*., 1987). Calcium/cation ratios appear to be particularly relevant in serpentine or solonetzic soils (Khalil *et al*., 1999; Ashraf and Naqvi, 1991; Carter *et al*., 1979 and Proctor *et al*., 1981) and in

sulfate-dominated saline soils (Mona, 1999; Ashraf and Waheed, 1992 and Janzen and Chang, 1987).

Calcium has been known for some time to alleviate the effects of salts (Amer, 1999; Ashraf and Neilly, 1992 and LaHaye and Epstein, 1969), possibly by maintenance of ion selectivity of membranes (Somal and Yapa, 1998; El-Etriby, 1990; Greenway and Munns, 1980). Recently, it has been shown that the high ionic strength of saline solutions displaces Ca from the membranes of root cells (Abdel-Rahman, 1997; Benlloch *et al.*, 1994; Cramer *et al.*, 1985; Lynch and Läuchli, 1988 and Lynch *et al.*, 1987), possibly contributing to salinity-induced Ca deficiencies.

Plant response to an increase in Ca under saline conditions has often been compared against very low calcium levels (e.g. 0.1 mM) which are not generally representative of saline soils (El-Sayed, 1997; Reggiani *et al.*, 1994; Kawasaki and Moritsugu, 1978; Mahammed *et al.*, 1987; Abdel-Mawly and El-Sayed, 1999). It has been suggested that crop response to specific ions in artificially salinized media is often over estimated because of the use of these unusually low Ca levels (Reda, 1996; Shehata *et al.*, 1994; Maas and Grieve, 1987). Even so, Ca concentrations in some saline soils, although quite high, are insufficient to prevent reductions in tissue Ca content (Abdel-Rahman *et al.*, 1996; Abdel-Hady *et al.*, 2001; Janzen and Chang, 1987 and Lynch and Läuchli, 1985). Further, saline Egyptian soils and Canadian prairies are often dominated by Na and Mg sulfate (El-Noemani, 1996; Mona *et al.*, 2000 and Fowler and Hamm, 1980), which when compared to NaCl, may exert a more severe reduction in growth (El-Sayed and Ghaly, 1996 and Moulay and Mohamed, 2000). High sulfate levels may decrease available Ca through precipitation reactions, which may, in part, be responsible for reduced Ca levels in cereals on these soils (Mostafa, 1996; Shehata and Darwiesh, 2001 and Janzen and Chang, 1987).

In view of the relatively meager information on crop response to this type of salinity, two species differing in salt tolerance were compared in solution cultures which were typical in composition of sulfate-dominated dryland saline soils. Plant response to Ca was determined and related to the salinity tolerance of each crop.

MATERIALS AND METHODS

Seeds of wheat (*Triticum aestivum* L.) Cv. Sakha 8 and barley (*Hordeum vulgare* L.) cv. Giza 123 were germinated in water on filter paper and transferred to vermiculite-filled plastic beakers (one seedling each) having highly perforated bottoms. The beakers were mounted in a platform suspended above containers of nutrient solution each with a rooting volume of nine plants per 14-L of solution. Wheat and barley were grown in separate containers except in Expt. 1A, where 8 plants of each species were grown together in 11-L containers. (In the latter, the growth of non-salinized wheat was substantially less than that of barley, a situation not as evident when the species were grown separately). Solutions were aerated continuously and pH was adjusted to 6.5 every other day with H_2SO_4. Solutions were topped up

with water daily, and were replaced once a week. The base (non-saline) solution was made up in deionized water, and was composed of, in mM, 0.2 NH_4, 2.0 Mg, 2.5 NO_3, 1.5 K, 2.75 SO_4, 0.2 PO_4, 2.5 to 4.0 Ca (concentration varied slightly with experiment), 3.0 CL, and in µM, 50 Fe (as Fe-EDDHA), 6.0 B, 0.8 Mn, 0.5 Zn, 0.15 Cu, and 0.15 Mo. Manganese toxicity was observed in preliminary experiments, and silicate (as Na_2SiO_3 at 26 µM) was routinely added to alleviate this problem (Lewin and Reimann, 1969).

Salinity was imposed by the addition of Na_2SO_4, $MgSO_4$, $CaSO_4$, and $CaCl_2$ to the base solution. Sodium, Mg, SO_4, and Cl concentrations were typical of moderately to severely saline soils, according to the classification of Fowler and Hamm (1980) based on the saturated paste extraction method. Calcium concentrations varied according to treatment and experiment. With the exception of $CaSO_4$, salts were added gradually in increments of 10 to 15 mM/d of total salt. Because of its relatively poor solubility, $CaSO_4$ was dissolved overnight in fresh nutrient solution, to which the other salts were then added. Final salt concentrations were maintained at a constant value through the duration of the treatment period. Unless otherwise stated, treatments were started 11 days after seedling, and continued for 22 days. Treatment solution water potentials were determined with thermocouple psychrometers (JRD Merrill) and the mineral content of solutions was verified with plasma emission spectroscopy. All cation ratios are expressed on a molar basis.

Plants were maintained in a greenhouse. Containers of plants were rotated periodically within the growing space to ensure uniformity. Plants were rinsed at the crown with deionized water every other day to prevent salt accumulation.

A series of five experiments was conducted. Each experiment was repeated once, sometimes with a few minor variations. The first experiment tested the effects of a combination of Na_2SO_4 plus Mg SO_4 salinity on wheat and barley growth, and the effects of Ca on the response to salinity. Plants were grown in either a non-saline solution, or one of two saline solutions which differed in Ca concentration and consequently in the Ca/(Na+Mg) ratio. For convenience, the two saline treatments were termed "saline" (low Ca) or "amended saline" (high Ca). In trial A, the Ca concentrations of the non-saline and saline solutions were the same (4.6 mM and representative of saturation extracts from non-saline soils (Fowler and Hamm, 1980). The Ca concentration of the amended saline treatment was 11 mM, typical of saturation extracts of saline soils (Fowler and Hamm, 1980). In trial B, the Ca concentrations of the saline treatment was 10 mM, and that of the amended saline treatment 19 mM, the latter being approximately twice the concentration found in saturation extracts of saline soils. Experiment 2 was similar, except that $MgSO_4$ alone replaced the Na_2SO_4 plus $MgSO_4$ combination. Experiment 3 examined the effects of the Ca/(Na+Mg) ratio on wheat and barley growth under non-saline conditions. Plants were grown in the base solution with low concentrations of Na, Mg and Ca adjusted to the appropriate Ca/(Na+Mg) ratio.

Since wheat showed a greater response to Ca than did barley under saline conditions, further work was conducted to characterize the nature of

the response in wheat. The effect of night-time humidity on the response to salinity and Ca was examined in Experiment 4. Groups of plants were grown in non-saline, saline or amended saline solutions as in Experiment 1, and either enclosed at night in plastic lined with damp paper towelling, or left at ambient humidity. Humidities were maintained at vpd's of either 0.2-0.1 KPa (90-95% rh) or 0.8-1.0 KPa (50-60% rh). Humidity was measured with an aspirated wet-bulb, dry-bulb thermometer. In Experiment 5, the effects of salinity and Ca on leakage of UV-absorbing substances from wheat leaf tissue was examined. Plants were grown in non-saline, saline or amended saline solutions as described in Experiment 1. Twenty leaf sections, each 2 cm in length, were cut from lamina on the main shoot of plants from each treatment. After rinsing for 1 h in distilled water, the samples were drained and shaken in 25 mL distilled water at 25°C for 24 h. Absorbance at 280 nm (A) was then determined for each solution (Evenhuis and De Waard, 1978; FAO, 1980). The samples were frozen at -70°C for at least 4 h, allowed to thaw, and shaken for 2-3 h. Absorbance was again determined at 280 nm (A`). The relative leakage ratio (RLR) was calculated as RLR = A/A`.

On termination of each experiment (except 5), leaf area (lamina only) was determined for each plant with a leaf area meter (Li-Cor LI-3000). In Experiments 1, 2 and 3, leaf sheaths, leaf lamina, and occasionally root dry weight was also determined for each plant after oven drying (Cottenie *et al.*, 1982).

In some cases the mineral content of the dried leaf lamina tissue was measured. Since it was not feasible to analyze each replicate separately, the tissue of each plant was ground and pooled within each treatment. In on instance (experiment 1) the mineral content of very young leaves was also determined. Here, the leaf sheaths were cut open and both the youngest emerging and oldest unemerged leaves were withdrawn. Samples were then pooled for analysis. After nitric-percholic acid digestion, samples were analyzed using plasma emission spectroscopy. All solution and tissue mineral analyses were conducted by Alexandria Soil Salinity Testing Laboratory. Variance in the mineral content was estimated using mean values obtained from repeated experiments.

Statistical analysis of the tabulated data was performed using analysis of variance (ANOVA) (Steel and Torrie, 1980 and Snedecor and Cochran, 1981). Comparisons of the two species in terms of the percent change in leaf area or dry weight due to salinity were assessed by ANOVA (SAS Institute Inc., 1988) after values for each replicate (plant) in the saline treatment were converted to percentages of the mean value of the non-saline treatment.

RESULTS

At the relatively low Ca concentration of 4.6 mM, the growth of both species was reduced by salt (Table 1,A). Wheat showed a significantly greater reduction ($P<0.001$) in leaf area and plant dry weight than did barley when expressed as a percentage of non-saline values. The Ca/(Na+Mg) ratio in this saline treatment was 0.07. The amended saline treatment containing 11 mM Ca increased the Ca/(Na+Mg) ratio to 0.18. Compared to the non-

amended saline treatment, leaf area was increased 138% in wheat and 25% in barley. Plant dry weight increased 42% in wheat but was not affected in barley.

The experiment was repeated at similar Ca/(Na+Mg) ratios to those just described, but at higher salt and Ca concentrations (Table 1,B). In the saline treatment (10 mM Ca,Ca/(Na+Mg) ratio of 0.09), leaf area and plant dry weight (as a percentage of non-saline values) was again reduced to a greater extent (P<0.001) in wheat than in barley. An increase in the Ca/(Na+Mg) ratio to 0.18, achieved by increasing the Ca concentration to 19 mM, improved the leaf area and dry weight of wheat when compared to the non-amended salt solutions. This increase in growth was apparent despite a small decrease in the solution water potential (-30, -350, and -380 KPa in non-saline, saline, and amended saline treatments, respectively). The Ca amendment, however, has no significant effect on barley growth. These trends were also apparent in the number of visibly Ca-deficient leaves. Young emerging leaves of tillers, and to a lesser extent, the main shoot, showed collapse and necrosis of the distal portion. Symptoms were often evident in the early stages of emergence, with subsequent growth being normal. Even so, substantial loss of area was evident in these leaves. Neither species showed Ca deficiencies under non-saline conditions. Wheat had a greater number of Ca-deficient leaves than barley in the presence of salt and Ca amendments reduced the number of Ca-deficient leaves from 15.9$\pm$0.8 to 5.7$\pm$0.7 per plant in wheat and from 4.3$\pm$0.9 to 0$\pm$0 per plant in barley (means of 9 plants $\pm$ S.E.).

Table 1: Effect of Na and MgSO$_4$-salinity and Ca on leaf area and plant dry weight of wheat and barley. Values are the means of 16 plants (A) or 9 plants (B). Experiment 1.

	Na	Mg	Ca	Wheat[a]		Barley[a]	
	(mM)			Leaf area (cm^2)	Plant dry Weight (g)	Leaf area (cm^2)	Plant dry Weight (g)
(A)							
Non-saline	<0.1	3	4.6	269 a	1.38 a	527 a	3.22 a
Saline	31	31	4.6	86 c	0.82 c	330 c	2.35 b
Amended saline	31	31	11.0	203 b	1.12 b	413 b	2.46 b
(B)							
Non-saline	<0.1	3	4.6	410 a	2.90 a	502 a	4.29 a
Saline	46	61	10	196 c	1.95 c	415 b	3.82 b
Amended saline	46	61	19	287 b	2.37 b	423 b	3.98 b

[a]**Mean separation within columns for each experiment by Duncan's multiple range test, 5% level.**

Under saline conditions, the Ca content of the leaves decreased, particularly in barley (Table 2). Sodium and Mg content increased with increasing in both species, with Mg being the highest in wheat and Na being the highest in barley. Amendment of the salt solution with additional Ca significantly increased Ca in barley and reduced Mg in wheat. Sodium was not affected in either species. The leaf Ca/(Na+Mg) ratio was reduced by salinity, more so in wheat than in barley. Calcium amendment increased this ratio in wheat but not in barley.

Table 2: Effect of Na and MgSO₄-salinity on cation content of wheat and barley leaves.

Species	Ca/(Na+Mg)[b] (m*M*)	Na	Mg	Ca	Ca/(Na+Mg)
		(µ mol/g dry weight)			
Wheat					
Non-saline	1.61	8 d	129 d	178 a	1.30 a
Saline	0.19	131 b	448 a	92 b	0.16 c
Amended saline	0.29	107 bc	316 b	118 b	0.28 b
Barley					
Non-saline	1.61	24 cd	117 d	178 a	1.26 a
Saline	0.19	454 a	225 c	50 c	0.07 d
Amended saline	0.29	478 a	205 cd	90 b	0.13 cd

[a]Mean separation within columns by Duncan's multiple range test, 5% level. Each value is the mean of 8, 8 and 7 replicates in wheat and 4, 4 and 3 replicates in barley in non-saline, saline and amended saline treatments, respectively. Each replicate consisted of at least 9 plants.

[b]Average of values ranging from 1.50-1.72, 0.21-0.17, and 0.28-0.30 in non-saline, saline and amended saline treatments, respectively.

Salinity and Ca also influenced the mineral status of young emergent leaves of wheat (barley was not measured). The Ca/(Na+Mg) ratios of the youngest emerging plus oldest unemerged leaves were 0.24+0.01, 0.05+0.005 and 0.09+0.01 in non-saline, saline and amended saline treatments, respectively (values are means of three consecutive trials, each with 9 plants, + S.E.).

The trend in plant response to MgSO₄ alone (Experiment 2) was similar to that of a combination of MgSO₄ and Na₂SO₄. In the first trial (Table 3,A), salinity (5 m*M* Ca, Ca/Mg ratio of 0.15) reduced the leaf area of wheat more than that of barley (*P*<0.05) when expressed as a percentage of non-saline values; shoot dry weight was reduced in wheat but was not significantly affected in barley. Amendment of the saline solution with Ca (11 m*M*, Ca/Mg ratio of 0.33) increased wheat leaf area and shoot dry weight compared to the non-amended treatment, but had no significant effect on barley. The second trial (Table 3,B) conducted at somewhat higher salt and Ca concentrations, produced similar results. Salinity (9 m*M* Ca, Ca/Mg ratio of 0.22) again reduced wheat leaf area more than that of barley (*P*<0.05), but influenced dry weight to the same extent in both species (*P*>0.05). Calcium amendment (16 m*M* Ca, Ca/Mg ratio of 0.39) increased wheat leaf area and shoot dry weight compared to non-amended plants, but had no significant effect on barley.

Species showed Ca deficiency symptoms under non-saline conditions, but both showed symptoms in the saline treatments. In the second trial, for example, salinity produced 5.3+0.4 Ca-deficient leaves per plant in wheat and 2.1+0.4 in barley; amendment with Ca reduced the number of deficient leaves to 0.2+0.2 in both species (means of 16 plants + S.E.).

Table 3: Effect of MgSO$_4$-salinity and Ca on leaf area and shoot dry weight per plant of wheat and barley. Values are   the means of 16 plants. Experiment 2.

	Mg	Ca	Wheat[a]		Barley[a]	
	(mM)		Leaf area (cm^2)	Shoot dry weight (g)	Leaf area (cm^2)	Shoot dry weight (g)
(A)						
Non-saline	3	5	273 a	1.32 a	406 a	1.93 a
Saline	33	5	185 c	0.97 c	331 b	1.76 a
Amended saline	33	11	230 b	1.07 b	353 ab	1.90 a
(B)						
Non-saline	3	4	295 a	1.31 a	480 a	2.26 a
Saline	41	9	225 b	1.09 c	423 b	1.96 b
Amended saline	41	16	279 a	1.22 b	392 b	2.01 ab

[a]**Mean separation within columns for each experiment by Duncan's multiple range test, 5% level.**

In the absence of salinity and at an average solution water potential of -100 KPa (Experiment 3), leaf area and shoot dry weight decreased more in barley than in wheat with decreasing Ca/(Na+Mg) ratio of the solution. In the first trial (Table 4,A) of four Ca/(Na+Mg) ratios (1.29, 0.49, 0.33 and 0.07), barley leaf area was significantly reduced at Ca/(Na+Mg) ratios of 0.33 and 0.07. Wheat showed a reduction in leaf area only in the 0.07 treatment. A reduction in shoot dry weight was apparent in barley at the 0.07 ratio but reductions were not at all evident in wheat. The second trial (Table 4,B) of three Ca/(Na+Mg) ratios (1.29, 0.13 and 0.07) produced similar results. Barley leaf area and shoot dry weight were reduced at the 0.07 Ca/(Na+Mg) ratio but wheat leaf area was unaffected.

Table 4: Effect of low Na, Mg and Ca concentrations on leaf area and shoot dry weight per plant of wheat and barley. Values are the means of 9 plants. Experiment 3.

	Na	Mg	Ca	Wheat[a]		Barley[a]	
	(mM)			Leaf area (cm^2)	Shoot dry weight (g)	Leaf area (cm^2)	Shoot dry weight (g)
(A)[b]							
	<0.1	3	4.0	610 a	3.31 a	627 a	4.28 a
	<0.1	6	3.0	613 a	3.27 a	653 a	4.39 a
	<0.1	6	2.0	600 a	3.23 a	559 b	4.20 a
	1.2	6	0.5	503 b	2.97 a	475 c	3.41 b
(B)							
	<0.1	3	4.0	229 a	0.76 a	388 a	1.14 a
	0.5	6	0.86	221 a	0.83 a	341 a	1.04 a
	1.0	6	0.46	200 a	0.79 a	136 b	0.76 b

[a]**Mean separation within columns for each experiment by Duncan's multiple range test, 5% level.**
[b]**Experiment A terminated after 37 days rather than 22 days.**

Decreases in the Ca/(Na+Mg) ratio also increased the number of Ca-deficient leaves. In the first trial, a Ca/(Na+Mg) ratio of 0.33 produced 0.5$\pm$0.3 Ca-deficient leaves per plant in wheat and 3.1$\pm$0.8 in barley; a ratio of 0.07 resulted in 0.9$\pm$0.3 and 9.1$\pm$1.1 deficient leaves in wheat and barley, respectively (means of 9 plants $\pm$ S.E.). Deficient leaves were not apparent at the two higher ratios. A Ca/(Na+Mg) ratio of 0.13 in the second trial produced 0.9$\pm$0.4 and 1.7$\pm$0.5 Ca-deficient leaves in wheat and barley respectively; a ratio of 0.07 resulted in 8.7$\pm$1.4 and 15.3$\pm$1.3 Ca-deficient wheat and barley leaves, respectively (means of 9 plants $\pm$ S.E.). Again, no deficiencies were evident at the highest Ca/(Na+Mg) ratio.

Night-time humidity had no effect on the leaf area of wheat grown at low salinity, but did influence salinized plants (Experiment 4, Table 5). The salt x humidity interaction was significant at 0.6% and 1.6% probability levels in trials A and B, respectively. High humidity improved the growth of salinized plants (8.6 mM Ca, Ca/(Na+Mg) = 0.08 and reduced the number of Ca-deficient leaves. Calcium amendment (19.1 mM Ca, Ca/(Na+Mg) = 0.19) improved leaf area and reduced the number of Ca-deficient leaves at low humidity compared to non-amended plants. No significant effect of Ca amendment on leaf area was observed at high humidity, but the number of Ca-deficient leaves was reduced.

Humidity did not appear to influence the leaf Ca/(Na+Mg) ratios. At low humidity, the Ca/(Na+Mg) ratio was 1.34$\pm$0.08, 0.17$\pm$0.03 and 0.29$\pm$0.04 in non-saline, saline and amended saline treatments, respectively (each value is the mean of the two trials in Table 5, each with nine plants, $\pm$ S.E.). Ratios at high humidity, in the same order, were 1.35$\pm$0.02, 0.18$\pm$0.02 and 0.30$\pm$0.04.

Table 5: Effect of night-time humidity, Na and MgSO$_4$-salinity, and Ca on leaf area and number of Ca-deficient leaves of wheat. Values are the means of 9 plants. Experiment 4.

Night humidity vpd (KPa)		Na	Mg	Ca	Trial A[a],[b]		Trial B[a]	
			(mM)		Leaf area (cm^2)	Ca-deficient leaves (plant^{-1})	Leaf area (cm^2)	Ca-deficient leaves (plant^{-1})
0.8-1.0	Non-saline	<0.1	3	2.6	867 a	0 e	350 a	0 d
	Saline	51	51	8.6	420 d	28 a	152 d	17.0 a
	Amended saline	51	51	19.1	591 c	11 c	251 b	2.5 d
0.1-0.2	Non-saline	<0.1	3	2.6	905 a	0 e	353 a	0 d
	Saline	51	51	8.6	646 bc	22 b	206 c	14.5 b
	Amended saline	51	51	19.1	690 b	6 d	236 bc	5.0 c

[a]**Mean separation within columns by Duncan's Multiple Range Test, 5% level.**
[b]**Experiment terminated after 43 days rather than 22 days.**

Leakage of UV-absorbing substances from leaf tissue of wheat was influenced by salinity (Experiment 5, Table 6). Salinity increased RLR by 21%. Amendment of the saline solution with Ca resulted in no significant change in RLR compared to non-saline plants.

Table 6: Effect of saline solutions on the relative leakage ratio (RLR) of wheat leaves. Experiment 5.

	Na	Mg	Ca	RLR[a],[b]
	(m*M*)			
Non-saline	<0.1	3	2.6	0.0329 b
Saline	51	51	11	0.0398 a
Amended saline	51	51	21	0.0295 b

[a]**Values are means of the third and fourth leaves to emerge, each replicated 30 times (i.e. 30 plants) in two experiments (n = 60).**
[b]**Mean separation by Duncan's multiple range test, 5% level.**

DISCUSSION

Barley growth was not reduced as extensively as that of wheat in the presence of salt, supporting the view that barley is the more salt tolerant species (Ashraf and Naqvi, 1991; Abdel-Aziz and Reda, 2000; Maas and Hoffman, 1977). However, the Ca concentration of the salt solution had a marked effect on the relative salt tolerance of the two species, improving the growth of wheat but not barley. Hence, the difference in growth between the two species under saline conditions was due, in part, to species differences in the interaction of salinity and calcium nutrition.

In work with salinity-Ca interactions, Ca/cation ratios are often increased by progressively decreasing the salt concentration as the Ca concentration is increased, thereby maintaining a more consistent osmotic potential in the treatment solution. This may result, however, in an overestimation of the response to an increase in the Ca/cation ratio because of the concurrent reduction in Na or other saline cations. Despite a small reduction in osmotic potential, an increase in Ca concentration at constant Na plus Mg, or Mg alone, clearly promoted growth of wheat. This indicates that Ca was indeed limiting growth in the saline treatment.

Changes in the cation ratios of the treatment solutions were generally reflected in the cation ratios of leaf tissue. The Ca/(Na+Mg) ratios of both species decreased under saline conditions, but increased significantly with additional Ca only in wheat. Wheat consistently had higher Ca/(Na+Mg) ratios than barley in saline and amended saline treatments, even though growth under salinity was less than that of barley and showed a greater response to additional Ca. Wheat would appear to have higher Ca requirements than barley in the presence of salt, although this was not reflected in species differences in the Ca relations of bulked leaves. Bulked leaf samples, however, do not discriminate between expanding and expanded leaves, which may be quite different in ion content (Reggiani *et al.*, 1994; Moulay and Mohamed, 2000; Greenway and Munns, 1980). The substantially lower Ca/(Na+Mg) ratios (by 70-80 percent) of young, emergent leaves of wheat compared to older leaves suggests that the Ca nutrition of leaves in the crown is relatively poor, a situation often encountered in low or non-transpiring organs. Salinity appears to exacerbate the problem by reducing the Ca/(Na+Mg) ratio even further, inducing Ca deficiency symptoms in those leaves. Species comparisons of Ca/(Na+Mg) ratios in emerging leaves were

not conducted, but may prove useful in explaining species differences in the susceptibility of those leaves to Ca deficiency under saline stress.

The relative response of wheat and barley to Ca/(Na+Mg) ratio under non-saline conditions was the reverse of that found at high salinity. In this case, wheat showed fewer Ca-deficient leaves and less extensive growth reductions than barley. Cation antagonism under non-saline conditions is well known to influence tissue cation ratios (Benlloch *et al.*, 1994; Abdel-Hady *et al.*, 2001; Mengel and Kirkby, 1987). Magnesium will depress Ca uptake (Shehata *et al.*, 1994; Shehata and Darwiesh, 2001; Ohno and Grunes, 1985), leading to Ca deficiency and growth reductions (Ashraf and Neilly, 1992; Abdel-Sabour, 1999; Kawasaki and Moritsugu, 1979; Key *et al.*, 1962; Madhok and Walker, 1969). Differences in Ca nutrition among species or genotypes, however, have rarely been characterized (Ashraf and Waheed, 1992; Amer, 1999; Mona *et al.*, 2000; Clarkson and Hanson, 1980). The greater Ca requirement of wheat compared to barley appears to be unique to saline conditions, and is not related to a greater susceptibility of wheat to low Ca/(Na+Mg) and Ca/Mg ratios *per se.*

The modulation of the salt tolerance of wheat by Ca was influenced by humidity. High 24 h humidity generally promotes growth and reduces transpiration and salt uptake rates, irrespective of salinity level (El-Noemani, 1996; Gawish *et al.*, 1999; Hoffman and Jobes, 1978; Lauter and Munns, 1987; Pitman, 1984). In the case of wheat, a proportional increase in the growth of plants in both non-saline and saline conditions at high 24 h humidity results in no change in salt tolerance (Reda, 1996; Khalil *et al.*, 1999; Hoffman and Jobes, 1978). In contrast, the results showed that high night-time humidity increased growth, but only in salinized plants, indicating an increase in salt tolerance under these conditions. The promotive effect of additional Ca on growth was not evident in the high humidity treatment. This suggests that an interaction between night-time humidity and Ca nutrition occurs under saline conditions, and is supported by the observation of reduced susceptibility of young leaves to Ca deficiency at high humidity.

Salinity-induced increases in leakage of UV-absorbing substances from wheat leaves were found to be ameliorated by additional Ca in the growing medium. Unlike previous measurements of salt-induced membrane leakiness, where low-salt leaf tissue has been incubated in saline solutions (El-Sayed, 1997; Mona, 1999; Somal and Yapa, 1998; Leopold and Willing, 1984), we have shown changes in leakiness based on *in vivo* levels of salt and Ca. Calcium effects on leakage are not confined to saline situations, since increases in leakage were apparent also at low salinity when Ca/(Na+Mg) ratios wer low. It is conceivable that those changes in membrane leakiness related to Ca concentration or Ca/cation ratios may contribute to differences in growth.

There is an increasing awareness of the significance of Ca in salt tolerance. Differences in Ca nutrition at high salinity have recently been shown to occur among genotypes of barley (Asaad *et al.*, 1998; Lynch and Läuchli, 1985), rice (Abdel-Rahman, 1997; Abdel-Rahman *et al.*, 1996; Grieve and Fujiyama, 1987; El-Sayed and Ghaly, 1996) and sorghum (Mostafa, 1996; Abdel-Mawly and El-Sayed, 1999; Grieve and Maas, 1988), and

between salt-selected and unselected cells of alfalfa (El-Etriby, 1990; Stavarek and Rains, 1984).

CONCLUSION

Salt tolerance appears to be related to improved Ca concentrations or Ca/cation ratios in the tissue of tolerant plants. Based on our findings, differences between species do not seem to be related to differences in Ca concentrations in the tissue, but rather to differences in Ca utilization or requirements. In this respect, salt-sensitive species may be more dependent on Ca availability than are salt tolerant species. Breeding or selection programs for salt tolerance must therefore take into account the critical role of Ca, with appropriate consideration given to the Ca relations of each species.

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التفاعل بين الملوحة والكالسيوم فى نباتات القمح والشعير

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*** مركز البحوث الزراعية ـ المعمل المركزى للمبيدات ـ محطة بحوث وقاية النبات ـ الصباحية ـ الاسكندرية ـ مصر

مــن المعــروف أن الكالسيوم يعتبــر عنصــراً هامــاً للنباتــات الناميــة فــى الأراضــى الملحية . لذلك فقد تـم دراسـة التفاعـل بين عنصـر الكالسيوم والملوحـة لصـنفين مـن القمـح والشعير ومعرفة مدى قدرتهم على تحمل الملوحـة فى وجود عنصـر الكالسيوم فى محلـول مغذى .

أوضحت النتائج أن القمح أظهر نقصــاً كبيراً فى النمو فى المحلـول المغـذى حيث تأثر تأثراً كبيراً وظهرت على أوراقه أعراض نقص الكالسيوم فـى وجود تركيز مرتفع مـن أملاح كبريتات الماغنسيوم وكبريتات الصوديوم وعلى العكس من ذلك فإن نبات الشـعير نمـا نمواً طبيعياً فى وجود الأملاح ولم تظهر على أوراقه أعراض نقص الكالسيوم .

أظهرت النتائج أنه عند إضافة الكالسيوم للمحلول الملحـى أمكن التغلـب على تـأثير الأملاح وزادت نسبة الكالسيوم / (الصوديوم + الماغنسيوم) ، وحدث استجابة كبيرة لنبـات القمح لعنصر الكالسيوم حيث عمل الكالسيوم على تحسين نسبة الكالسيوم / (الصوديوم + الماغنسيوم) فى أنسجة كل من القمح والشعير .

أوضــحت النتــائج أن نسـبة الملوحـة بـالقمح تكـون متكافئة مـع المحتـوى العـالى للكالسيوم ، وكذلك الاحتياجات المرتفعة للقمح من الكالسيوم تكون دالة لحالة الملوحة بالتربة .

أدت الرطوبة المرتفعة ليلاً الـى تحسين نمو القمـح تحت الظروف الملحية ، بينمـا زيادة تركيز الكالسيوم فى المحلول المغذى الملحـى لـم يؤثر علـى النمو فـى حالـة الرطوبـة المرتفعة ، كذلك ازداد الغشاء المنفذ لأنسجة الورقة النامى تحت الظروف الملحية عنهـا فى أنسجة الورقة تحت الظروف الغير ملحية . كذلك فإن نمو النباتـات فـى المحلول الملحـى المحتوى على عنصر الكالسيوم يوضح عدم زيادة الغشـاء المنفذ للأوراق وهذا يؤكد أهميـة تفاعـل عنصـر الكالسـيوم للتغلـب علـى تركيز الملوحـة بالتربـة ، ويختلـف تبعـاً لاخـتلاف واستجابة الأصناف المختلفة للنباتات .