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OSMOTIC ADJUSTMENT FOR TWO WHEAT VARIETIES

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Abstract

The physiological basis of salt tolerance of wheat Giza 164 (relatively salt tolerant) and Sakha 69 (relatively salt sensitive) was assesed using sand culture technique; the treatments of NaCl were 0, 20, 40 and 80 mol m~3 in full strength of Arnon nutrient solution. The results showed that increasing NaC1 concentration in the rooting medium had a marked effect on water and solute potentials for leaf and root of both two varieties. Whereas, Sakha 69 had a marked lower water and solute potentials in root, Giza 164 had a marked lower water and solute potentials in leaf. On the contrary, Giza 164 maintained high turgor potential in leaf, while Sakha 69 maintained high turgor potential in root.

Data also showed that Na and C1 ions contributed the major parts in osmoregulation in leaves and roots of both two varieties. However, Giza 164 contained higher concentrations of inorganic solutes in leaves, while Sakha 69 contained higher concentrations of inorganic solutes in roots. Also, Giza 164, generally, accu~nulated more proline in leaves and mots at all levels of salinity as compared with Sakha 69.

The relatively salt tolerant (Giza 164) maintained low water and solute leaf potentials and high leaf proline content to relatively resist salt injury.

INTRODUCTION

Salinity is one of the important factors affecting growth and yield of most crops. Reggiani *et al.* (1994) reported that shoot and root growth of wheat cultivars which differ in salt sensitivity was progressively inhibited by increasing NaCl concentration up to 150 mM in irrigation water. Mansour *et al.* (1993) found that sodium concentration in both roots and shoots of wheat seedlings increased proportionally with increasing salinity. On the other hand, Glenn *et al.* (1994) reported that tolerant plants (*Atriple Canescens*) were associated with restricted accumulation of Na in the shoots mainly due to osmotic adjustment. However, it is known that the ability of plants to accumulate solutes in their tissues is commonly known as "osmotic adjustment" which is believed to reduce the water potential in plant cells. A number of inorganic and organic components have been identified which increase in their concentration in tissues of higher plants subjected to saline stress. Among these Na, K, Cl as well as proline and total free amino acids "compatible solutes" are known to be osmotically active (Reda, 1996). Lu and Yi (1992) and Naqvi *et al.* (1994) observed that proline accumulation in wheat seedlings is an adaptive response to NaCl stress. Reda (1996) added that salt tolerant wheat variety showed higher level of proline than sensitive variety subjected to NaCl stress condition. This may agree with results of Wrench *et al* .(1977) who suggested that proline has a protective function in the cytoplasm. This amino acid has been suggested by Wated *et al.* (1983) and Amer (1989) to play an adaptive role in tolerance of plant cells to NaCl salinity by increasing the concentration of cultural osmotic components in order to equalize the osmotic potential of the cytoplasm (Flowers *et al.*, 1977).

The present work was carried out to draw comparisons between water relations and inorganic and organic solutes for two wheat varieties differ in salt tolerance, explanations of possible mechanisms involved are considered.

MATERIALS AND METHODS

Grains of two varieties of wheat (*Triticum aestivum*), Sakha 69 and Giza 164, were obtained from Agricultural Research Station at Giza. Giza 164 was shown to be relatively salt tolerant as compared with Sakha 69 (Gawish *et al.*, 1998). Natural sand was washed thoroughly by HCl then by distilled water. Polyethylene pots of 15 cm diameter which had a hole at the bottom, to facilitate flushing, were used and received a quantity of 2 kg of washed dried sand per pot. Twenty grains of wheat were planted and seedlings being thinned 7 days later to 8 plants per pot. The treatments of salinity were 0, 20, 40 and 80 mol m^{-3} NaCl in full strength of Arnon nutrient solution. The experiment was planned in a complete randomized design with treatments having three replicates each. The pots were regularly flushed by nutrient solutions of different salinity treatments 7 days after the start of experiment.

After 30 days, a fully expanded leaf and a main root were taken from each plant for analysis of water potential (Yw) using water potential apparatus (Chasw. Cock and Sons, Birmingham, UK). Solute potential (Ys) was determined using an Osmometer TP 10 B (Camlab Limited). Turgor potential (Yt) was mathematically derived from the difference between solute and water potentials (Ashraf, 1989).

Samples of leaves and roots were dried at 70 °C and then ground. Dry samples were used to assay inorganic and organic solutes. Among these, Na and K were determined according to Chapman and Pratt (1961), Ca and Mg being assayed using Elmer atomic absorption spectrophotometer as described by Cottenie *et al* (1982). Cl was determined according to Greenway (1963). NO_3 was determined according to Johnson and Ulrich (1950). Free proline content was estimated using the method of Bates (1973) and the total free amino acids being evaluated according to Lee and Takahashy (1966).

RESULTS AND DISCUSSION

1. Water relationship parameters:

The data for the three water relationship parameters for leaf and root of the two studied varieties; water potential Yw (-Mpa), solute potential Ys (-Mpa) and turgor potential Yt (Mpa) are presented in Tables 1 and 2. Turgor potential (Yt) was calculated, as previously mentioned, as the difference between water potential and solute potential. Increasing the concentration of NaCl salinity had a marked effect on the leaf and root water potentials of both varieties. This result is in agreement with that obtained by Ashraf (1989). Also, varieties showed marked different responses to increasing salt concentration. Sakha 69 had markedly higher leaf water potential than Giza 164 at both concentrations of 40 and 80 mol m^{-3} NaCl. On the contrary, Giza 164 had markedly higher root water potential than Sakha 69 at all salt concentrations.

As regard to leaf and root solute potentials, data showed that increasing NaCl concentration in the rooting medium was,generally, followed by a gradual decrease in leaf and root solute potentials. Similar results were also obtained by Ashraf (1989). This decrease in solute potential should be mainly attributed to the decrease in water potential or the increase in dissolved solutes as a result of uptake of salt (Statyar, 1961). Also, varieties had different responses to increasing NaCl concentration. Sakha 69 had markedly higher leaf solute potential, while, Giza 164 had markedly higher root solute potential. These results show clearly that salt tolerant variety (Giza 164) had higher Yw and Ys in root than the salt sensitive variety (Sakha 69) to aviod physiological drops. In fact, the markedly lower leaf osmotic potential in Giza 164, compared to that of Sakha 69, may be related to the fact that halophytic as well as glycophytic plant species may adjust to high salt concentration by lowering tissue osmotic potential with increased uptake of solutes (Flowers *et al.*, 1977).

The leaf turgor potential representing varieties responses to increasing NaCl concentrations, since it was markedly different. Giza 164 maintained higher leaf turgor potential, while Sakha 69 maintained higher root turgor potential at all salt concentrations tested. In fact, higher leaf turgor potentials have been considered a principal factor for maintaining growth at high salinities (Greenway and Munns, 1980).

Table 1. Leaf water potential (Yw), solute potential (Ys) and turgor potential (Yt) of the two studied wheat varieties at different levels of NaCl salinity.

Potential	NaCl salinity as mol m^{-3}			
	0	20	40	80
Sakha 69				
Yw (- Mpa)	3.64	3.78	5.05	6.32
Ys (- Mpa)	4.72	4.76	5.89	6.46
Yt(Mpa)	1.08	0.98	0.84	0.14
Giza 164				
Yw (- Mpa)	1.74	3.70	5.68	8.99
Ys (- Mpa)	3.43	5.25	7.19	10.44
Yt (Mpa)	1.69	1.55	1.51	1.45

Table 2. Root water potential (Yw), solute potential (Ys) and turgor potential (Yt) of the two studied wheat varieties at different levels of NaCl salinity.

Potential	NaCl salinity as mol m^{-3}			
	0	20	40	80
Sakha 69				
Yw (- Mpa)	2.94	3.04	3.28	3.73
Ys (- Mpa)	3.74	3.80	3.86	4.23
Yt(Mpa)	0.80	0.76	0.58	0.50
Giza 164				
Yw (- Mpa)	2.08	2.14	2.43	2.97
Ys (- Mpa)	2.46	2.38	2.59	3.07
Yt (Mpa)	0.38	0.24	0.16	0.10

2. Inorganic and organic solute:

The osmotic contribution of the inorganic and organic solutes are presented in Tables 3 and 4. It was assumed that Na, K, Ca, Mg, Cl and NO_3 were used as the total inorganic solute, while the free proline and total free amino acids were used as the total organic solute.

Tables 3 and 4 cleared that Na and Cl contributed the major part in osmoregulation in leaves and roots of both varieties, while NO_3 contribution was minimal. The contribution of inorganic solute to the total solute concentration ranged between 81.2 and 89.9 % in leaves of Sakha 69 and ranged between 80.7 and 92.5 % in leaves of Giza 164 when the NaCl salinity increased from 0 to 80 mol m^{-3} NaCl, respectively .For roots, it ranged from 84 to 95.3 % in Sakha 69 and from 76.4 to 85.7 % in Giza 164. These results show that inorganic solute in leaves of Giza 164 was higher, whereas that of Sakha 69 was lower. Opposite trend was observed for roots. consequently comparisons between varieties can be based only on Na and Cl concentrations in the leaves. These comparisons between varieties with extreme differences of salt tolerance support the view that high electrolytes concentration in leaves are of adaptive value. In this connection, Shannon (1978) indicated that tolerance was associated with restricted accumulation of Na in the shoots. Also, Bernstein (1961) concluded that electrolytes provide the major contribution to osmotic pressure in most experiments at high salinity.

For organic solute, addition of NaCl to the rooting medium markedly increased the leaves and roots proline content in both varieties . Similar results were also observed by Somal and Yapa (1998). Varieties differed markedly in leaves and roots proline content. Giza 164 ,generally, accumulated more proline in leaves and roots at all salt concentrations as compared to Sakha 69 . This result is in agreement with that obtained by El-Leboudi *et al* (1997). In this concern, Wrench *et al* (1977) suggested that proline is associated with enzymes in such a way to serve as a protective function in the cytoplasm. Also,total content of free amino acids in Giza 164 for leaves and roots was higher, whereas that of Sakha 69 was lower. The corresponding relative values to control were 104, 116, and 111 % in leaves and 92, 86 and 69 % in roots of Giza 164, while it reached 85, 88 and 68 % in leaves and 92, 77 and 67 % in roots of Sakha 69 when the salinity were 20, 40 and 80 mol m-3, respectively. Such amino acids were suggested by Watad *et al* (1983) to play an adaptive role in the tolerance of plant cells to NaCl salinity by increasing the concentration of cultural osmotic components in order to equalize the osmotic potential of the cytoplasm (Flowers *et al.*, 1977).

Table 3. Inorganic and organic solutes in leaves of the two studied wheat varieties at different levels of NaCl salinity.

Solutes (mM) In tissues' water	NaCl salinity as mol m-3			
	0	20	40	50
Sakha 69				
Inorganic solutes:				
Na^+	107.62	229.63	472.41	678.98
K^+	434.55	356.13	337.55	248.11
Ca^{++}	149.47	127.78	136.18	101.47
Mg^{++}	286.84	237.04	250	225.49
Cl^-	474.43	588.42	798.72	1052.2
NO_3^-	104.75	81	65.33	47.2
Total inorganic solutes	1557.66	1620	2060.19	2353.45
Organic solutes:				
Free proline	9.45	10.23	16.65	21.54
Total free amino acids	350.92	299.2	310	240
Total organic solutes	360.37	309.43	326.65	261.54
Giza 164				
Inorganic solutes:				
Na^+	87.55	432.51	742.18	1226.01
K^+	314.8	292.66	318.47	251.79
Ca^{++}	117.56	125.92	140.13	191
Mg^{++}	218.65	237.35	267.52	262.01
Cl^-	313.76	697.59	1069.35	1936.3
NO_3^-	68.91	56.83	51.57	43.16
Total inorganic solutes	1121.23	1842.86	2589.22	3910.27
Organic solutes:				
Free proline	7.45	12.09	21.6	30.83
Total free amino acids	259.09	270.29	300.85	288
Total organic solutes	266.54	282.38	322.45	318.83

Table 4. Inorganic and organic solutes in roots of the two studied wheat varieties at different levels of NaCl salinity .

Solutes (mM) In tissues' water	NaCl salinity as mol m^{-3}			
	0	20	40	80
Sakha 69				
Inorganic solutes:				
Na$^+$	169.67	204.92	420.37	1176.05
K$^+$	241.4	129.39	137.41	199.66
Ca^{++}	231.7	188.52	248.62	368.85
Mg^{++}	187	153.69	184.16	266.39
Cl$^-$	189.63	234.36	373.51	923.57
NO$_3^-$	49.57	33.05	37.43	52.88
Total inorganic solutes	1068.94	943.93	1401.46	2987.44
Organic solutes:				
Free proline	5.23	6.11	9.32	13.14
Total free amino acids	197.27	182.31	151.22	132.71
Total organic solutes	202.5	188.42	160.6	145.85
Giza 164				
Inorganic solutes:				
Na$^+$	134.65	233.32	328.83	443.36
K$^+$	172.22	109.03	93.37	88.56
Ca^{++}	160.45	112.9	84.03	82.24
Mg^{++}	136.82	102.64	91.04	63.05
Cl$^-$	132.44	170.17	225.67	372.5
NO$_3^-$	23.47	15.61	14.01	15.39
Total inorganic solutes	760.02	743.69	836.92	1065.11
Organic solutes:				
Free proline	5.1	8.08	14.32	21.13
Total free amino acids	228.71	211.44	196.82	155.22
Total organic solutes	233.81	219.52	211.14	176.35

CONCLUSIONS

In agreement with previous findings, results suggest that the salt sensitive plants (Sakha 69) accumulated more Na and Cl in roots than the relatively salt tolerant plant (Giza 164), whereas the latter variety transported more Na and Cl to shoots to reduce the adverse effect of Na and Cl on root growth. The relatively salt tolerant plants seemed to be able to withstand the unfavorable effect of high Na and Cl accumulation in roots and consequently on growth through higher rates of Na and Cl translocation to shoots.

Moreover, Giza 164 accumulated more proline and total free amino acids in leaves and roots than that of Sakha 69 to equalize the osmotic potential of the cytoplasm.

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ضبط الاسموزية لصنفين من القمح

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أجريت تجربة معملية باستخدام بيئة رملية علي صنفين من القمح جيزة ١٦٤ (متحمل نسبيا للملوحة) وسخا ٦٩ (حساس نسبيا للملوحة) وذلك لدراسة الأسس الفسيولوجية لعملية التحمل الملحي، وقد تم إضافة ملح كلوريد الصوديوم بتركيزات صفر ، ٢٠ ، ٤٠ ، ٨٠ مليمول/لتر إلي المحلول الغذائي المستخدم.

أوضحت النتائج المتحصل عليها أن زيادة تركيز ملح كلوريد الصوديوم في البيئة كان له تأثير ملحوظ علي كل من الجهد المائي والاسموزي لكل من الورقة والجذر لكلا الصنفين، حيث انخفض الجهد المائي والاسموزي في الصنف جيزة ١٦٤ في الورقة بينما حدث ذلك في الصنف سخا ٦٩ في الجذر، كذلك احتفظ الصنف جيز ١٦٤ بجهد انتفاخ عالي في الورقة بينما احتفظ الصنف سخا ٦٩ بجهد انتفاخ عالي في الجذر.

كما أوضحت النتائج أيضا أن كل من أيوني الصوديوم والكلوريد ساهما مساهمة كبيرة في عملية ضبط الاسموزية في كل من الورقة والجذر لكلا الصنفين ومع ذلك فان الصنف جيزة ١٦٤ كان عالي في محتواه من الصوديوم والكلوريد في الأوراق بينما العكس في الصنف سخا ٦٩ حيث كان محتواه عالي منهما في الجذور.

أيضا أظهرت النتائج أن الصنف جيزة ١٦٤ كان محتواه من البرولين في الأوراق والجذور أعلا من الصنف سخا ٦٩ وهذا مما يؤيد بان الصنف المتحمل نسبيا للملوحة (جيزة ١٦٤) قد احتفظ بجهد مائي واسموزي منخفض في الأوراق ومحتوي عالي من البرولين وذلك لمقاومة الضرر الناتج من ارتفاع الأملاح في البيئة الخارجيه.