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Mechanisms of Salt Tolerance in Selected Wheat Cultivars

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ABSTRACT

An experiment in NaCl saline solution culture was conducted to observe the mechanisms of salt tolerance in three selected wheat cultivars, two Chinese (Bao-119 & cv. 86-6) and one Pakistani (FSD-85) differing in salt tolerance. Fifteen days old 20 seedlings of each cultivar in ½ strength Hoagland nutrient were subjected to incremental salt stress until the required salinity levels (0, 200-mol m^{-3}) was obtained. Dry matter yield (DMY) at 30 days, indole acetic acid (IAA), abscisic acid (ABA) and total free amino acid (AA) contents of leaves / roots at 29 days and dehydrogenase activity of root at 34 days were determined after initiation of the salt stress. Shoot and root (DMY), dehydrogenase activity of root and IAA contents both in leaves and root of all the cultivars decreased; whereas, ABA and AA contents increased. At saline treatment, cultivar FSD-85 produced maximum shoot DMY and Bao-119 minimum; whereas, in case of root, cv. 86-6 produced maximum and FSD-85 minimum DMY. Dehydrogenase activity of root of Bao-119 was more affected followed by FSD-85 and cv. 86-6. The extent of decrease of IAA contents in leaves and root of cv. 86-6 was more pronounced than other cultivars. Increase in ABA contents of shoot and root was more in FSD-85 followed by Bao-119 and cv. 86-6, respectively. The extent of increase of AA contents was more in the leaves of FSD-85. From this study, it was concluded that cultivar FSD-85 has the salt inclusion and cv. 86-6 salt exclusion mechanism. The tolerance of cultivars was in order of FSD-85> cv. 86-6 >Bao-119.

Key Words: Salinity; DMY; Dehydrogenase activity; IAA; ABA; ABA

INTRODUCTION

Salinity and sodicity has affected about 10% of the total world land (Szabolcs, 1991). Approximately 20 mha land deteriorates to zero production each year (Malcolm, 1993) mainly due to salinization. The salt affected area in Pakistan is estimated about 6.67 mha (Khan, 1998) of which 60% is saline sodic where as in Punjab saline sodic area is about 80% (Muhammad, 1983). Salt affected soils can be managed by reclamation, but due to less availability of good quality of water, low soil permeability and high cost of amendments, this approach is not feasible on a large scale (Qureshi *et al.*, 1990). Saline agriculture technology is an alternative approach for effective utilization of salt affected soils, which involves the cultivation of salt tolerant species/crop cultivars. This technology gives economic returns from salt affected soils and provide vegetative covers to soil which reduces evaporation and hence the rate of salinization (Qureshi & Barrett-Lennard, 1998). Study of response of plants/crops to salinity under naturally saline condition is not feasible due to extreme variability in soil salinity both spatially and temporarily (Richards, 1983). To avoid this problem, comparative differences for salt tolerance among crops/varieties can be studied under artificially salinized control conditions.

Salinity tolerance in wheat has been and is being extensively researched in Pakistan and elsewhere in the world, but still efforts to improve salt tolerance have been hampered by a number of factors, particularly the lack of understanding of the mechanisms of salt tolerance and interaction of salinity with various environmental factors with regard to plant growth. Wheat tolerance to salinity varies with the stage of plant growth, nature and level of salinity, duration of stress etc. (Qureshi *et al.*, 1990) and is affected by soil moisture, climate, nutrition and management practices (Maas & Hoffman, 1977). Different physiological traits such as selectivity for potassium, exclusion and/or compartmentation of sodium and chloride ions, an osmotic adjustment by accumulation of organic solutes have all been related to salt tolerance of crops plants (Wyn Jones & Storey, 1981). In this study, an attempt has been made to study the effect of NaCl salinity on yield, dehydrogenase activity, indole acetic acid (IAA) contents, abscisic acid (ABA) contents and total free amino acid contents and their relationship with salt tolerance of cultivars under consideration.

MATERIALS AND METHODS

Experiment was conducted in wire house of the department of Soil Science and Agricultural Chemistry, Zhejiang Agricultural University, Hangzhou, P. R. China, with natural daylight and day/night temperature of 17/7ºC, respectively. Sufficient healthy seeds of three wheat cultivars {2Chinese (Bao-119, cv. 86-6) and one Pakistani (FSD-85)} were soaked in 0. 2% fungicide solution for 18 h. After draining fungicide solution, the seed were washed thrice with tap water. Then seeds were sown in quartz sand in iron trays. The condition in trays kept moist with water and trays remained covered until the sprout came out and

waited for nine days. Thirteen days old 20 seedlings of each cultivar were transferred to 1 cm plugged holes in wooden covers over 32 L of ½ strength Hoagland and Amon (1950) nutrient solution in plastic containers. Ten holes were used for each cultivars and each hole having two seedlings. Fifteen days old seedlings were subjected to incremental salt stress. Salt concentrations were increased by 25 mol m^{-3} after every 12 h by adding NaCl to nutrient solution until the required salinity level (200 mol m^{-3}) was obtained in respective container. Thirty eight days old seedlings were subjected to full strength Hoagland nutrient solution. Solutions were renewed after every 7 days and pH 6.0-6.5 was daily maintained and loss of water was made regularly. Solutions were aerated for 9 h every day with air pump by splitting in to three equal parts and intervals. Twelve plants were harvested 30 days after salinization. The plants were washed for five minutes in running tap water followed by a quick rinse in distilled water. The plant tissue dried at 70ºC were weighed. Abscisic acid and indole acetic acid were determined 29 days after salinization by the method given by Weiler *et al.* (1986), total free amino acid after 29 days by the method given by Water Associates (1983). Dehydrogenase activity of the roots was determined after 34 days of salinization using the method as given below;

One-gram root sample was taken in test tube. Added 5 mL 0. 4% TTC and 5 mL 0. 1 M phosphate buffer solution. Then the sample was incubated for at 37ºC. After 3 h, samples were immediately taken out from incubator and added 2 mL 2N H_2SO_4 for stopping the enzyme reaction. Then ground the roots in pestle mortar with ethyl acetate up to no colour and made volume up to 50 mL. Then reading was taken at 485 nm wavelengths on spectrophotometer and calculated as under:

Reduced TTC (g. g^{-1}. F. Wt h^{-1}) = 50 x reading of sample / weight of sample x time (in h)

Where TTC= 2, 3, 5-triphenyl tetrazolium chloride

Statistical analysis was done by the methods given by Steel and Torrie (1980).

RESULTS AND DISCUSSION

The results of this study indicate, that salinity decreased the DMY of shoot and root, dehydrogenase activity of roots and IAA contents of leaves and roots, whereas increase in ABA and AA contents in both leaves and roots was observed in all the cultivars tested. Data in Table I revealed that saline treatment gave 62% DMY both of shoot and root. The cultivars FSD-85 produced maximum DMY of shoot (67% of control) with 28% coefficient of variation and followed by c v. 86 and Bao-119 whereas in root, cv. 86-6 was at the top (68% of control) with 29% coefficient of variation followed by Bao-119 and FSD-85. Reduction of DMY under saline condition is in agreement with those of Almansouri *et al.* (1999). Data in Table II revealed that saline treatment decreased and gave 67% dehydrogenase activity. Murumkar and Chavan (1987) and Mattioni *et al.* (1997) also reported reduction in root dehydrogenase activity. The root dehydrogenase activity was highest (75% of control) in cv. 86-6 with 20% coefficient of variation and lowest (57% of control) in Bao-119 with 39% coefficient of variation. The root dehydrogenase activity had positive correlation with shoot potassium contents and negative with root potassium contents at saline treatment as well as on cumulative means of two treatments (Pervaiz *et al.*, 2002). Data in Table III revealed that salinity decreased and gave 45 and 6% IAA contents both in leaves and roots, respectively. The decrease in IAA contents is in agreement with Wright (1978), Guinin and Brummet (1987), Ikeda *et al.* (1989), and Prakash and Prathapasenan (1990). The IAA contents of leaves were highest (94% of control) in Bao-119 with 4% coefficient of variation and lowest (11% of control) in cv. 86-6 with 11% coefficient of variation. As regard roots IAA contents, the highest (23.80% of control) in FSD-85 with 86% coefficient of variation and lowest (2. 47% of control) in cv. 86-6 with 135% coefficient of variation were found.

Data in Table IV revealed that salinity increased the ABA contents both in leaves and roots of all the cultivars. The ABA contents of leaves were highest (942% of control) in FSD-85 with 114% coefficient of variation and lowest (122% of control) in cv. 86-6 with 14% coefficient of variation. As regard roots ABA contents, the highest (331%

Table II. Effect of sodium chloride salinity on the dehydrogenase activity of roots of three wheat cultivars

NaCl (mol m^{-3})	Dehydrogenase activity (TTC g g^{-1}. Fresh weight. h^{-})			
	Bao-119	Cv.86-6	FSD-85	Average
0	514	470	475	486
200	291 (57)	352 (75)	338 (71)	327 (67)
Average	403	411	407	
Coefficient of variation (%)	39	20	24	

Figures in parenthesis are percentage of their respective control

Table I. Effect of sodium chloride salinity on dry matter yield of shoot and root of three cultivars

NaCl (mol m^{-3})	Dry weight of shoot (g)				Dry weight of root (g)			
	Bao-119	Cv.86-6	FSD-85	Average	Bao-119	Cv.86-6	FSD-85	Average
0	1.27	1.49	1. 39	1. 38	0.50	0. 45	0. 54	0. 50
200	0.74 (59)	0. 90 (60)	0. 93 (67)	0. 86 (62)	0.32 (63)	0. 30 (68)	0. 32 (60)	0.31(62)
Average	1.00	1.20	1.16		0.41	0.38	0.43	
Coefficient of variation (%)	37	35	28		32	29	37	

Figures in parenthesis are percentage of their respective control

Table III. Effect of sodium chloride salinity on indole acetic acid contents of leaves and roots of three wheat cultivars

NaCl (mol m^{-3})	Indole acetic acid contents (P mol g^{-1}. Fresh weight)							
	Leave				Root			
	Bao-119	**Cv.86-6**	**FSD-85**	**Average**	**Bao-119**	**Cv.86-6**	**FSD-85**	**Average**
0	843	2546	963	1451	255	276	82	204
200	792 (94)	274 (11)	886 (92)	651 (45)	8.42(3.30)	6.83(2.47)	19.52(23.80)	11.59 (6)
Average	818	1410	925		132	141	51	
Coefficient of variation (%)	4	11	6		132	135	86	

Table IV. Effect of sodium chloride salinity on abscisic acid contents of leaves and roots of three wheat cultivars

NaCl (mol m^{-3})	Abscisic acid contents (P mol g^{-1}. Fresh weight)							
	Leaves				Roots			
	Bao-119	**Cv.86-6**	**FSD-85**	**Average**	**Bao-119**	**Cv.86-6**	**FSD-85**	**Average**
0	53218	48284	14388	38630	3358	5873	10285	6505
200	80789 (152)	59005 (122)	135489 (942)	91761 (238)	7538 (224)	11776 (201)	34079 (331)	17798 (274)
Average	67004	53645	74939		5448	8825	22182	
Coefficient of variation (%)	29	14	114		54	47	76	

Table V. Effect of sodium chloride salinity on total free amino acids contents of leaves and roots of three wheat cultivars

NaCl (mol m^{-3})	Total free amino acids contents (ppm)							
	Leaves				Roots			
	Bao-119	**Cv.86-6**	**FSD-85**	**Average**	**Bao-119**	**Cv.86-6**	**FSD-85**	**Average**
0	3697	2953	2725	3125	2077	2211	2089	2126
200	4281 (116)	3864 (131)	3941 (145)	4029 (129)	1811 (87)	2463 (111)	2304 (110)	2193 (103)
Average	3989	3409	3333		1944	2337	2197	
Coefficient of variation (%)	10	19	26		10	8	7	

Figures in parenthesis are percentage of their respective control

of control) in FSD-85 with 76% coefficient of variation and lowest (201% of control) in cv. 86-6 with 47% coefficient of variation were found. The data is in agreement with those of Verma (1980), Clipson *et al.* (1988) and Cammue *et al.* (1989). The probable cause in increase of ABA contents might have been due to the accumulation of Na^+ contents, which caused the deficiency of water in plant tissues. Non-significant positive correlation (r = 0. 995) between ABA and Na^+ contents (Pervaiz *et al.,* 2002) of shoot of all the cultivars was observed. Inconsistent patterns between ABA contents and salt tolerance of cultivars on dry weight basis were observed. Accumulation of more ABA and Na^+ contents (Pervaiz *et al.,* 2002) and production of more DMY in case of FSD-85 might have been due to the stimulation of protein synthesis from glutamic acid/or accumulation of more Na^+ in shoot and survive under adverse condition is the genetic character of this cultivars. Accumulation of more ABA in the excised leaves of drought resistant line of both maize and sorghum than susceptible line has been reported by Larque-Saavedra and Wain (1976) and ABA as responsive element in salt tolerant indica rice by Sudhiranjan *et al.* (1998). But this review is not in agreement with the result of this experiment, because at 200 mol m^{-3} salinity cv. 86-6 showed moderate relative shoot DMY and least ABA contents whereas Bao-119 showed least relative shoot DMY and moderate ABA contents in its leaves. The results of these experiments are in agreement with the hypothesis of Pitman *et al.* (1974) who reported that effect of ABA depends on the species, growth condition and temperature. Salinity increased the AA contents both in leaves and roots of all the cultivars except in roots of Bao-119 (Table V). The AA contents of leaves were highest (145% of control) in FSD-85 with 26% coefficient of variation and lowest (116% of control) in Bao-119 with 10% coefficient of variation. As regard roots AA contents, the highest (111% of control) in cv. 86-6 with 8% coefficient of variation and lowest (87% of control) in Bao-119 with 10% coefficient of variation were found. The increase in AA contents is in agreement with those of Imam-ul-Haq (1983), Gorham *et al.* (1984b) and Mattioni *et al.* (1997). Higher the relative DMY of shoot and higher the AA contents of leaves and reverse were also true, and this is in agreement with those of Dubey and Rani (1989). Decrease in AA contents due to salinity was also reported by Gorham *et al.* (1985a) in the leaves of *Thinopyurum bessarabicum*. The roots relative AA contents at 200 mol m^{-3} salinity also showed consistent pattern with relative root DMY. Generally, increase in AA contents in leaves was more than in roots.

As regard the individual character of each cultivar, it was noted that FSD-85 has the salt inclusion mechanism because of production of best DMY in the presence of higher quantity of sodium and chloride in its shoot (Pervaiz *et al.,* 2002), ABA and AA contents in leaves and even moderate root dehydrogenase activity which are the best evidences of its salt tolerance/salt inclusion mechanism. The

cv. 86-6 showed salt exclusion mechanism, because of production of moderate DMY among the cultivars under consideration, and accumulation of least sodium and chloride in its shoot (Pervaiz *et al.,* 2002), least ABA and IAA and best root *Dehydrogenase* activity (Gupta & Kaur, 1970). The Cultivars Bao-119 was found sensitive due to minimum shoot DMY, minimum and maximum reduction in IAA contents of leaves and root dehydrogenase activity (Gupta & Kaur, 1970), respectively and moderate accumulation of sodium and chloride contents in shoot.

CONCLUSION

The cultivars, FSD-85 and cv. 86-6 showed salt inclusion and exclusion mechanism and found salt tolerant cultivars. Bao-119 was salt sensitive due to its low DMY/ root dehydrogenase activity and accumulation of more sodium and chloride contents in its shoot as compared to cv 86-6.

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