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Salinity Induced Biochemical Changes in Berseem During Germination

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Abstract

Berseem cultivars JHB-146, Wardan (tolerant), BL-10 and JB-5 (susceptible) were analyzed to various levels of salinity stress (control, 4.0, 8.0 and 12.0dS/m) to elucidate the certain biochemical basis of salt tolerance during germination at seedling stage. The results indicate that increase in salt stress occupied a decrease in total sugar and an increase in malondialdehyde and phenol contents in all cultivars. However, tolerant cultivars showed a greater amount of sugar, phenols and lesser MDA content as compared to susceptible cultivars.

Key words : Berseem, Seeds, Germination, Salts, *In vitro* conditions.

Berseem (*Trifolium alexandrinum* L.) is an important *rabi* fodder crop of India. Understanding the physiological, biochemical and molecular mechanisms involved in imparting salt tolerance is most crucial for development of stress tolerant genotypes. Among abiotic stresses, salinity is an important forage yield reducer, which delays germination at lower levels and response may vary with type of salinity and species or cultivars. Salt stress is one of the major problems in irrigated agriculture, particularly in berseem grown areas. The increasing pressure of population and dwindling land resources had made it necessary to get better production from saline soils. High concentrations of salts have deterimental effect on biochemical activities of forage crops, excessive amounts cause death of growing plants. Thus exploitation of genetic variability in cultivated species may be made to see the possibility of developing salt tolerant crops (1).The mechanisms, which impart salt tolerance to some and sensitivity to others, however have not been fully worked out in forage crops. Hence, the present research work was undertaken to find out the biochemical basis of salinity stress tolerance in berseem cultivars during germination at seedling stage *in vitro* conditions.

Methods

The study work was conducted in March 2003. Seeds of berseem cultivars were procured from different Agricultural Institutes of India viz. I.G. F. R. I., Jhansi (UP), P. A. U, Ludhiana (Punjab) and J. N. K.V. V., Jabalpur (MP). The seeds of four cultivars of berseem were screened for their relative salinity tolerance. Cultivars JHB-146 and Wardan were identified as tolerant, whereas BL-10 and JB-5 were found to be susceptible. Seeds of the four cultivars were surface sterilized with 0.1% $HgCl_2$ solution for one minute and thoroughly washed with distilled water. The saline solutions were prepared by taking NaCl, $NaHCO_3$,Na_2SO_4 and $CaCl_2$ (2). The seeds of different cultivars were germinated at different levels of salinity viz. control, 4. 0,8.0, and 12.0 dS/m in sterilized germination boxes lined with Whatman filter paper no.1 and kept in BOD incubator at 26 ± C temperature. The experiment was terminated on day 10. Ten day old seedlings were subjected to various analyses. The quantitative estimations of total sugar, malondialdehyde and total phenols were estimated by their procedures described earlier (3—5).

Results and Discussion

The data on total sugar is represented in Table 1. The results indicate that total sugar content showed a decreasing trend with increasing levels of salinity from control to maximum (12.0 dS/m). At maximum salinity level, higher total sugar content was observed in JHB-146 (30.0 mg) and Wardan (26.20 mg) than BL-10 (19.90 mg) and JB-5 (14.90 mg). Salinity stress induced a reduction in sugar content has also been reported earlier (6).

Table 1. Effect of salinity of total sugar, malondialdehyde (MDA) and total phenol of berseem.

Cultivars	Salinity (ds/m)	Total sugar (mg/g dry/wt)	MDA (μ mol/g fr wt)	Total phenol (mg/g dry wt)
	Control	36.00	0.0027	4.96
JHB-146	4.0	34.90	0.0032	7.07
(Tolerant)	8.0	32.00	0.0035	10.00
	12.0	30.00	0.0039	11.00
	Control	39.00	0.0024	5.56
Wardan	4.0	34.00	0.0027	6.86
(Tolerant)	8.0	30.83	0.0032	9.85
	12.0	26.20	0.0036	10.35
	Control	26.25	0.0019	4.10
BL-10	4.0	24.90	0.0030	6.4
(Susceptible)	8.0	21.90	0.0040	7.79
	12.0	19.90	0.0068	8.00
	Control	25.00	0.0015	4.85
JB-5	4.0	21.90	0.0026	5.47
(Susceptible)	8.0	17.50	0.0034	6.89
	12.0	14.90	0.0059	8.95
C.D. at 5%				
Salinity (S)		1.10	0.0016	1.05
Cultivar (C)		1.12	0.0017	1.07
S × C		2.23	0.0041	2.85

Increasing trend of MDA content, a measure of lipid peroxidation, was noticed in all the cultivars with increasing levels of salinity (Table 1). At 12 dS/m, the values were lower in tolerant ones, viz. JHB-146 (0.0039 μ mol) and Wardan (0.0059 μ mol) compared to susceptible ones, viz BL-10 (0.0068 μ mol) and JB-5 (0.0059 μ mol). Salinity stress induced an increase in MDA has also been confirmed earlier (7). Free radicals and other active derivatives of oxygen are inevitable product of natural redox reaction in various cellular compartments under different saline conditions. The free radicals mediated peroxidation of unsaturated fatty acids also referred as lipid peroxidation is a chain reaction occurring mainly in biomembranes.

Our findings indicate that the amount of total phenols increased in all cultivars with rise in salinity levels ,and the tolerant maintained a higher level of phenols at all levels of salinity (Table 1). Salt stress induced an increase in phenol has been examined earlier (8). It is also reported that phenol constitutes a part of cellular solutes providing a reducing environment to the system. Salt stress exerts its effect through membrane peroxidation, which indicated that oxygen free radicals are found during stress. Hence, higher phenol accumulation in tolerant cultivars could be a cellular adaptive mechanism for scavening oxygen free radicals and preventing subcellular damage during stress. Thus higher amount of sugar and phenols and lower content of MDA might prove to be useful for screening berseem cultivars to withstand salinity.

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