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Effect of Salinity on Growth and Biochemical Parameters of Berseem (*Trifolium alexandrinum* L.)

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Abstract

Eleven genotypes of berseem (*Trifolium alexandrinum* L.) were evaluated under different levels of salinity (control, 8.0 and 12.0 dS/m) created by using salts viz. NaCl, $NaHCO_3$,Na_2SO_4 and $CaCl_2$ for 10 days after moistening the seeds for primary test (screening) on percentage germination and dry weight of seedlings. Out of these two most tolerant (JHB-146 and Wardan) and two most susceptible (BL-10 and JB-5) varieties were further investigated for physiological basis of salt stress tolerance. At maximum salinity stress (12.0 dS/m), there were comparatively more accumulation of sugars, chlorophylls and free amino acids along with higher catalase activity.
Key words : Berseem, Salinity, Chlorophylls, Sugars, Amino acids.

Salinity is one of the major problems in irrigated agriculture, particularly in berseem growing areas because high concentrations of salt have detrimental effect on plant growth, excessive amounts cause death of growing plants and use of poor quality water for irrigation and poor drainage. Therefore, the identification of suitable genotypes for this purpose requires an efficient screening of genotypes for salt tolerance to sustain crop productivity and achieve food security. The mechanism which imparts salt tolerance to some genotypes and sensitivity to others, has not fully worked out in berseem varieties. Soil salinity affects various plants physiological and biochemical processes depending on salt type and concentrations, plant genotype, growth rate and environmental conditions (1).

In the present study an attempt was made to examine the growth and some biochemical changes in berseem seedlings induced by salinity.

Methods

The present experiment was conducted on two most tolerant (JHB-146 and Wardan) and two most susceptible (BL-10 and JB-5) varieties selected from a set of 11 varieties viz. JB-2, JB-5, JB-1, BL-10, BL-1, HFB 600, HFB 100-1, JHB-146, Wardan, Mescavi and Sh-69 on the basis of germination percentage and dry weight of seedlings. Seeds of the four genotypes were surface sterilized with 0.1% $HgCl_2$ solution for one minute and thoroughly washed with distilled water. The saline solutions were made by taking salts viz. NaCl, $NaHCO_3$,Na_2SO_4 and $CaCl_2$ following the method of Richards (2). The seeds of four different varieties were germinated in different levels of salinity (control, 8.0 and 12.0 dS/m) in sterilized petridishes lined with Whatman filter paper no.1 in BOD incurator at 26 C temperature; 10 ml of saline solution was added in each petridish containing 30 seeds and carried in triplicate. The seedlings were allowed to grow for 10 days. Dry shoot samples were subjected to various analyses like total sugars (3) and total free amino acids (4). Total chlorophyll and enzyme catalase were assayed in fresh samples by the procedure of Arnon (5).

Results and Discussion

Salinity is known to exert depressive effects on physiological functions and energy generating biochemical processes in berseem and other fodder crops. There was a gradual reduction of seedlings in all four genotypes with increasing level of salinity (Table 1). Moreover, the relative rate of the loss was directly determined by the concentration of salt. Further, varietal differences in salt tolerance take some

Table 1. Effect of salinity levels on some morphological and biochemical parameters on 10 days old seedlings of berseem.

Varieties	Salinity (dS/m)	Dry wt of seedling (mg)	Chlorophylls (mg/g fw)	Total sugars (mg/g dw)	Free amino acids (mg/g dw)	Catalase (per g fw)
JHB-146	control	75.60	0.495	28.50	36.50	111.50
(Tolerant)	8.0	65.25	0.440	24.50	32.50	140.50
	12.0	52.60	0.365	20.00	26.00	165.20
Wardan	control	89.0	0.480	27.40	35.80	108.70
(Tolerant)	8.0	80.53	0.422	23.00	29.50	138.70
	12.0	62.50	0.330	20.00	22.22	170.50
BL-10	control	69.80	0.405	25.15	26.50	145.18
(Susceptible)	8.0	50.20	0.292	18.50	18.50	130.50
	12.0	36.50	0.167	15.00	10.50	105.25
JB-5	control	74.80	0.388	24.16	23.70	170.70
(Susceptible)	8.0	40.73	0.254	19.55	13.70	130.78
	12.0	30.50	0.150	15.50	09.90	097.50
CD at 5%	Salinity (S)	Variety (V)	S × V			
Dry wt of seedlings	1.950	2.223	3.940			
Chlorophylls	0.017	0.2000	0.045			
Total sugars	1.450	1.600	2.950			
Free amino acids	1.080	1.250	2.155			
Catalase	1.610	1.755	3.025			

time to show as the main difference in growth rate between seedlings exposed to various levels of salinity appeared between day 6 and 9 after moistening. This is because up to this time the seedling becomes less dependent on the cotyledons. The possible causes of varietal differences most likely involve ion transport properties and cellular com- partmentation. These in turn affect leaf water relations and could affect metabolic processes such as photosynthesis. The present findings indicated that minimum reduction in growth was observed in JHB-146 and Wardan, whereas the maximum reduction was observed in BL-10 and JB-5 genotypes. The genotype and salinity interaction was recorded as significant. This reduction under salinity levels may be due to inhibition of hydrolysis of reserve food material and its translocation to growth axis (6).

The total chlorophylls in different genotypes of berseem (Table 1) decreased with increasing levels of salinity and the more decrease was noticed in susceptible cultivars BL-10 and JB-5. The present findings are in conformity of earlier reports (7). The less reduction of chlorophyll pigments in tolerant cultivars might have caused more dry matter accumulation in berseem genotypes.

Total soluble sugars gradually declined in all four cultivars with increasing levels of salinity (Table 1). Significant differences were observed between genotypes of tolerant and susceptible groups for total sugars. The reduction in sugar content under salinity stress might be due to lower amylase activity with causes reduced hydrolysis of reserve polysaccharides in cotyledons. Salt induced reduction in sugar content has been reported (8). At higher levels of salinity more accumulation of sugars were observed in tolerant varieties. The possible role of sugar might be a readily available energy source.

The salinity has differential effects on amino acid spectrum. Total free amino acids content showed a declining trend in both tolerant and susceptible genotypes of berseem. These findings are in accordance with earlier reports (9). Comparatively more accumulation of amino acids in tolerant genotypes might be due to high enzyme activity under test condition and decreased availability of amino acids might also be due to denaturating of enzymes involved in amino acid formation. The characteristic changes in the levels of free amino acids have been observed in leaves of plant exposed to salt stress. The most obvious of them was an increase in proline, the role of cytoplasmic osmoticum counter balancing the effect of salts and play a significant role in salt tolerance of the variety. Hence, in salt stressed plants amino acids and amides accumulate at the expense of structural and functional proteins and balance between soluble amino acids and proteins is changed due to disturbed metabolism of the berseem, which results in the form of suppressed growth.

It was found that salinity caused an increase in catalase in all varieties as compared to control sets. The activity of catalase was more in tolerant genotypes as compared to susceptible genotypes at 12 dS/m. Similar findings were reported earlier (10). High level of antioxidative enzyme is a part of biochemical and physiological make up involved in salt tolerance and ameliorate the oxidative damage arising from salt stress. Higher activity of catalase in tolerant genotypes may help in protecting the membrane from lipid, peroxidation due to H_2O_2, which is a toxic metabolite produced by plants in various pathways.

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