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Influence of Salinity and Sodicity on Nodulation and Nitrate Reductase Activity in Berseem (*Trifolium alexandrinum* L.)

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Abstract

The present findings indicate that a concentration dependent decrease in nodulation viz. number, fresh weight, dry weight, leghemoglobin content and nitrate reductase activity was noticed in two germplasms of berseem viz. JHB-146 and JB-5 when grown in the field and exposed to artificial salinity and sodicity. The percentage reduction over control was observed to go up with increasing salt stresses and with progressive stages of growth and development. Salinity demonstrated a relatively more drastic effect than sodicity. This variable inhibitory effect of four salts NaCl, $NaHCO_3$, Na_2SO_4 and $CaCl_2$ proved more pronounced in JB-5 than in JHB-146 revealing the latter germplasm to be more tolerant, it is suggested that nodulation and nitrate reductase activity may serve as valuable indices of salt stress experienced during growth and development.

Key words : Berseem, Nodulation, Nitrate reductase activity, Salinity, Sodicity.

Salinity and sodicity are the major abiotic stresses, which adversely affect the plant growth and leads to poor productivity. Two schools of thought have disputed the relative importance of osmotic effects and specific ion effects. Both salinity and sodicity impose a significant limitation on plant productivity by adversely affecting the host plants, root nodule bacteria, symbiotic development and nitrogen fixation capacity. Active nitrogen fixing root nodule of legumes possesses pink pigment hemoglobin called leghemoglobin (Lb), whose concentration is positively correlated with nodule nitrogen fixation. However, fully developed nodules that had formed under stress free conditions continue to fix nitrogen when subjected to salt stress. Nitrate reductase , which converts nitrate into nitrite is the best key enzyme in the process of nitrate utilization, a significant step in plant growth, protein metabolism and yield. The NR activity inhibition has been reported in several crop species under salinity and sodicity. Hence,an attempt was made to elucidate the impact of salinity and sodicity on nodulation and NR activity in berseem.

Methods

The seeds of two germplasms of berseem procured from Indian Grassland and Fodder Research Institute (I.G.F.R.I.), Jhansi (UP) and Jawaharlal Nehru Krishi Vishwa Vidyalaya (J.N.K.V.V.), Jabalpur (MP), the field experiment was conducted in 2004 to evaluate the effect of salinity and sodicity on nodulation and nitrate reductase activity; 0.1% (wt/vol) $HgCl_2$ sterilized seeds of tolerant JHB-146 and sensitive JB-5 germplasms of berseem were sown in equal 1 m^2 area plots hoving properly manured soil. Salinity and sodicity were induced by using NaCl, $NaHCO_3$, $Na_2SO_4$and $CaCl_2$ formula (1) to maintain 0, 8, 12, 16, 20 and 24 ECe levels. The constancy of the ions from leaching was maintained by flooring the beds by a polythene sheet at a depth of 30 cm. direct reading conductivity meter was used to used to cross check the degree of salt stress. The excessive accumulation of soluble salts has prevented by watering the plants due to evaporation in the rhizosphere. The plants were harvested and analyzed at different durations i.e. 60, 90, 120 days after sowing (DAS). Number of nodule and weight of nodules per plants were measured after detaching them from roots. Lb content of nodule was measured (2). Fresh leaves were plucked from each plot and used for assay of nitrate reductase (3).

Results and Discussion

The accumulated salts in rhizosphere not only inhibit various metabolic processes but also enzy-

Table 1. Effect of soil salinity on nodulation and nitrate reductase activity in JB-5 and JHB-146 germplasms of berseem.

		Growth stages on DAS					
		60		90		120	
Parameters	Treatment	JB-5	JHB-146	JB-5	JHB-146	JB-5	JHB-146
No.of nodules	Control	24	25	25	26	30	31
	8 ECe	23	24	27	29	28	29
	12 ECe	23	24	24	25	26	28
	16 ECe	21	22	23	24	25	27
	20 ECe	22	23	22	23	24	26
	24 ECe	21	22	21	22	21	25
Fresh weight of nodules (g)	Control	–	–	0.20	0.24	0.24	0.40
	8 ECe	–	–	0.19	0.23	0.24	0.39
	12 ECe	–	–	0.18	0.22	0.22	0.37
	16 ECe	–	–	0.17	0.21	0.19	0.35
	20 ECe	–	–	0.16	0.20	0.18	0.25
	24 ECe	–	–	0.15	0.20	0.17	0.23
Dry weight of nodules (g)	Control	–	–	0.113	0.122	0.132	0.240
	8 ECe	–	–	0.112	0.121	0.131	0.240
	12 ECe	–	–	0.111	0.120	0.130	0.190
	16 ECe	–	–	0.108	0.117	0.128	0.180
	20 ECe	–	–	0.107	0.114	0.126	0.165
	24 ECe	–	–	0.100	0.113	0.085	0.150
Lb content (µg/g, fresh weight)	Control	40	41	47	48	54	55
	8 ECe	37	44	44	51	52	53
	12 ECe	35	39	40	44	47	48
	16 ECe	29	37	37	41	44	45
	20 ECe	25	31	34	37	37	42
	24 ECe	17	29	29	36	34	40
Nitrate reductase activity (NO_2/h/g fresh weight)	Control	2.13	2.18	2.88	2.89	2.88	2.90
	8 ECe	1.65	1.66	2.73	2.77	2.51	2.53
	12ECe	1.41	1.44	2.69	2.73	2.47	2.50
	16 ECe	0.93	0.95	2.39	2.69	2.14	2.16
	20 ECe	0.92	0.93	2.19	2.39	1.90	2.00
	24 ECe	0.86	0.89	1.90	2.00	1.10	1.60

matic activities in the plants. the present findings on nodulation indicate a reduction in number, fresh weight and dry weight of nodules per plant irrespective of durations and germplasms (Table 1). In general, number, fresh weight and dry weight of root nodules progressively decreased at all salinity levels (Table 1). These inhibitory effects of salinity have also been reported earlier (4). Nodule mass (fresh and dry) could not be determined due to small size at 60 DAS but nodule mass increased form 90 to 120 DAS at all salinity levels, which indicates the senescence of nodules when plants were reaching to maturity. It is also observed that inhibitions in number and mass of nodules are due to reduced rate of cell division.

Table 1 indicates that the Lb content decreased with increasing salinity levels from 8 to 24 ECe and increased up to 120 DAS irrespective salinity sodicity. This could be due to ageing nodule because of irreversible oxidation of Lb. These results are similar in the earlier reports (5). Salinity and sodicity enhanced earlier senescence of nodules and further restricted the development of pigment in newly formed nodules at later growth stages.

The activity of NR declined with increasing salinity levels in both germplasms at all three observations. These results are in the conformity with earlier reports (6). However, it was consistently higher in germplasm JHB-146 as compared to JB-5 at all salinity levels. Further, higher salinity levels (12 to 24 ECe) have depressed the NR activity, which has resulted in lesser nitrate assimilation in plants. This might be due to inhibition in nitrate uptake from soil. Reduction in NR activity ieads to slow protein synthesis, which ultimately reflected in terms of growth. The

suppression in nodulation and NR activity was found higher in germplasm viz. JB-5 than JHB-146. Reduction in pink pigment may be ascribed to depressed synthesis, enhanced oxidation of the pigment and degradation of heme protein due to increased proteolytic activity. decreased chlorophyll contents under salt stress leads to reduced supply of photosynthesis, may be one of the cause of reduced amount of Lb because their formation depends more on the availability or proper need of photosynthates. The cause of NR activity inhibition may be due to dissociation of FAD in the leaves or depletion of endogenous nitrate pool.

The variable inhibitory effect of salinity and sodicity proved to be more pronounced in JB-5 than JHb-146 revealing the latter germplasm to be tolerant. It is suggested that nodulation and NR activity may serve as valuable indices of salt stress experienced during growth and development.

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