

INFLUENCE OF SALT ON THE GROWTH, NITROGENASE AND AMMONIA ASSIMILATING ENZYMES OF *AZOLLA PINNATA* R. Br.

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THE *Azolla-Anabaena* symbiosis represents potentially an ideal biofertilizer for rice production due to its high nitrogen fixing ability and rapid growth¹. Benefit of *Azolla* as a biofertilizer depends upon its high salt tolerance of rice field standing water. Very little work concerning the effect of salinity on *Azolla-Anabaena* complex has been done². In our earlier studies we reported that salinity affected the growth, nitrogen content of the complex and pigmentation of the symbiont also^{3,4}. The present communication deals with the influence of salt concentration on the biomass production, nitrogen fixation and ammonia assimilating enzymes of *Azolla pinnata-Anabaena azollae* complex.

Azolla pinnata R. Br. plants were grown in nitrogen-free liquid medium⁵. The total salt concentration of the medium was 770 ppm. Sodium chloride was added to the culture medium to make the final concentration of the media to 1000, 1300, 1900, 2500 and 3100 ppm, respectively. About three grams of actively growing *Azolla* plants were inoculated into one litre of the test medium. Cultures were incubated at $25 \pm 2^\circ\text{C}$ under fluorescent light for 16 h photoperiod. Plants were harvested after 21 days and their biomass recorded. Nitrogenase activity was measured by the acetylene reduction assay⁶. The three principal ammonia assimilating enzymes, viz., glutamine synthetase (GS), glutamate dehydrogenase (GDH) and glutamate synthase

(GOGAT) activities were assayed from the crude enzymes⁷⁻⁹ of *Azolla-Anabaena* complex.

Increasing salt concentrations in the media were associated with a reduction in the biomass and acetylene reduction activity (table 1). The growth of *Azolla* in the presence of lower concentrations (1000 and 1300 ppm) was compared to that of the control. Significant reductions in biomass production and acetylene reduction activity (ARA) were recorded from 1900 ppm and above salt concentration in the culture medium. Even at low salt concentration (1000 ppm), the GS activity was reduced by 6%, while GDH and GOGAT remained unaffected (table 1). Progressive reduction in GS activity was recorded as the salt concentration of the media increased. At 1900 ppm of salt level, the GS activity was reduced by 39% while at 3100 ppm of the salt level in the culture medium, GS activity was reduced by 77%. The GDH and GOGAT activities of the complex were not much affected. The maximum reductions recorded at the highest concentration of salt level (3100 ppm) treated *Azolla-Anabaena* complex was 8% and 13% in GDH and GOGAT, respectively.

In the *Azolla-Anabaena* association, both the partners possess the capacity for glutamate synthesis through GDH or by GS-GOGAT pathway. The host *Azolla* contributed about 90% of GS and 10% of GDH activities¹⁰. The present study clearly showed that higher salt concentration in the medium showed reduced nitrogen fixing activity with the corresponding reduction in the GS activity. GS and nitrogenase interact in blue green algae and the general consequence is that GS is involved directly or indirectly in nitrogenase regulation¹¹.

One of the authors (KR) is thankful to UGC, New Delhi for financial assistance.

Table 1 Influence of salt on the growth, nitrogenase and ammonia assimilating enzymes of *Azolla-Anabaena* complex

Total salt concentration of the medium (ppm)	Fresh weight (g)	ARA nmol C ₂ H ₄ formed/g fresh weight/h	GS ^a	GDH ^b	GOGAT ^b
770	25.9	685	386	39	28
1000	25.7	680	361	39	28
1300	25.3	668	327	39	28
1900	17.6	530	234	38	28
2500	10.8	411	131	36	27
3100	7.2	213	89	34	26
CD at 5%	1.3	76			

^aμmol γ glutamyl hydroxamate formed/mg protein/min. ^bμmol NADPH oxidized/mg protein/min.

12 March 1988

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AGROPYRON SPECIES AS COLLATERAL HOSTS FOR BLACK RUST OF WHEAT

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DURING the last several years occurrence of black rust [*Puccinia graminis* var. *tritici* (Pers.) Eriks. &

Henn.] was observed on *Agropyron yukonense* Scribn. & Merrill, *A. imbricatum* Roem. & Schult., *A. tauri* Boiss. & Bal., *A. intermedium* var. *trichophorum* (Link.) Haloe., *A. libanoticum* Hack. ex Kneuk., *A. scabriglume* (Hackel) L. Parodi, *A. dasystachium* (Hook) Scribn. and *A. batalinii* (Krassn.) Roshev., maintained at the IARI Regional Station, Wellington. The occurrence of black rust was observed on *A. yukonense* throughout the year, while on the other *Agropyron* spp. this occurred mainly during *kharif* season (June–October). Prasada¹ observed the occurrence of uredinial and telial stages on leaves, stems and ears of *A. semicostatum* and *A. longearistatum* in the Shimla hills. However, on inoculation, there was no infection on the differentials of *P. graminis tritici*. This led him to propose the rust as a variety of *P. graminis* and proposed the name *P. graminis* var. *agropyri* (Pers.) Mehta and Prasada.

The rust was isolated from the uredinial pustules occurring on leaves or stems of *Agropyron* spp. and cross-inoculations were carried out on cv Agra local or wheat. Isolates from all the *Agropyron* spp. infected Agra local wheat heavily. The isolates were further tested on 12 International Standard Differentials² and on 3 supplementary differentials for race identification. Reaction types produced by single spore cultures of the isolates on differential hosts are presented in table 1.

The isolate from *A. yukonense* produced reaction types identical to those of pathotype 117A, while those from other *Agropyron* spp. produced reaction types identical to those of pathotype 40A. The pathotype 40A is the most common on wheat in the Nilgiri hills, though in case of pathotype 117A, the

Table 1 Infection types produced by *Puccinia graminis tritici* pathotypes from *Agropyron* spp.

Pathotypes	International differentials ²												Supplementary differentials		
	a	b	c	d	e	f	g	h	i	j	k	l	m	n	o
117A (S.S.)	4	4	0;	0;	4	4	4	4	4	4	4	0; -1	4	4	0;
<i>Agropyron</i> <i>yukonense</i> isolate	4	4	0	0	4	4	4	4	4	3	4	0;	4	4	0
40A (S.S.)	4	4	4	4	4	4	4	4	4	0; -1	4	0; -1	4	4	4
<i>Agropyron</i> spp. isolate	4	4	4	4	4	4	4	4	4	0;	4	0;	4	4	4

a-Little club; b-Marquis; c-Reliance; d-Kota; e-Arnautka; f-Mindum; g-Spelmar; h-Kubanka; i-Acme; j-Einkorn; k-Vernal; l-Khapli; m-Charter; n-Yalta; o-E 535.