Agriculture for Sustainable Development 2(1):32-35, 2014/Article *ISSN 2347-5358 (Print)/ 2349-2228 (Online)*

Agriculture for Sustainable Development

http://www.sustainableagriculture.in

IMPACT OF BRSSINOLIDE IN PHYSIOLOGICAL AND BIOCHEMICAL ATTRIBUTES IN RICE UNDER SALINE STRESS

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Received: 19.02.2014 **Revised accepted: 23.03.2014**

ABSTRACT

An experiment was conducted with five rice varieties *viz*. GR-7, GR-11, GR-12, Dandi and Gurjari were grown in petridish for 10 and 15 DAG. Petridish was treated with NaCl (@ 100 mM and 200 mM), brassinolide (@ 4µM and 8 µM) and NaCl levels were also supplemented with the brassinolide levels. Control maintained with water. Saline stress affected seedling germination, fresh weight and dry weight significantly at the first place. Biochemical attributes e.g., mineral constituents and free amino acids content were also affected. The Na^+ ion concentration was increased where as K^+ ion was decreased with salinity levels. Supplementation of NaCl with brassinolide significantly decreased Na^+, thereby toxicity level and increased K^+ in the salt stressed plants of all the varieties. Salinity treated seedlings has been observed higher amino acids content as compared to control. Brassinolide was found to have a positive impact on physiological and biochemical attributes in all the varieties under saline stress.

Keywords:
Brassinolide,
Saline stress,
Amino acid

INTRODUCTION

Rice is the most important crop of India and second most important crop of the world. It is planted on about one-tenth of the earth's arable land and is the single largest source of food energy to half of humanity particularly in Asia where rice is the staple food. Salinity is one of the most vital abiotic stresses especially for rice, which is mostly grown under irrigated conditions. Rice is a salt sensitive crop. Under saline conditions, growth of rice plant varies depending on the particular growth stage i.e., starting from germination and ending to maturation. Attempts have been made to alleviate environmental stresses by applying phytohormones (Rao *et al.*, 2002). Brassinosteroids (BRs) (Anuradha and Rao, 2003; Ozdemir *et al.*, 2004) are variously involved in improving salinity tolerance of plants subjected to salinity stress. They are steroidal plant hormones with growth promoting activity (Mandava, 1988). They were at first recognized from the fact that pollen extracts from several species of genus *Brassica* resulted in overexpansion of internodes in dotted bean and termed brassins due to their action on inducement of growth. Brassinolide (BL) and castasteron are two important BRs often occurring in higher plants. Brassinolide has emerged as a new phytohormones with pleiotropic effect (Sasse, 1997), effect of various physiological processes like germination, growth, flowering, senescence and biochemical processes which confered resistance to plant against various abiotic stresses.

MATERIALS AND METHODS

Seeds materials of five rice varieties were surface sterilized with 0.1% $HgCl_2$ and were placed in petridishes lined with Whatman No.1 filter paper. Seeds were then treated with two levels of NaCl @ 100 mM, 200 mM and two levels of BL @ 4µM, 8 µM and thereafter Nacl solutions were also supplemented with BL levels to count the ameliorating effect of BL under saline stress in plants. Seeds were grown in distilled water for control and three replications were maintained for each treatment. For both physiological and biochemical studies, observations were recorded at 10 DAG and 15 DAG. Germination (%), fresh weight and dry weight (gm) of seedlings were studied following the methods of AOSA, 1981 and ISTA, 1985. Mineral constituents (Na^+ and K^+ ratio) and free amino acids content were studied by methods of Jackson, 1973 and Lee and Takahashi, 1966 respectively.

RESULTS AND DISCUSSION

The results revealed that germination percentage decreased with increased salinity levels and supplementation of the saline solution with brassinolide considerably reduced the inhibitory effect of salinity on seed germination. The

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germination percentage was highest under the variety V_2 (GR 11) and it remained at par with the variety V_4 (Dandi) at 10 DAG. Significantly the lowest germination was observed under the variety Gurjari and the second lowest germination was found in the variety GR 12 (Table-1 and Figure-1). In treatment combinations, V_2T_5 (GR-11 variety treated with 8µM BL) was found significantly the highest treatment combination in term of germination percentage and the lowest was observed under the treatment combination V_5T_3 (Gurjari variety treated with 200 mM NaCl). At 15 DAG also similar results were noticed except in the treatment combinations (Figure-2). Salinity had an adverse effect on seedling germination because of excess sodium as well as chloride ions on the mobilization of seed reserves and inactivation of hydrolyzing enzyme system during germination due to which redicle emergence could not occur. Brassinolide considerably helped to reduce the inhibitory effect of salinity on seed germination. There was considerable increase in the seedling weight as well due to the brassinolide treatment as compared to the untreated seedlings when grown in saline condition. The interaction with V × T also found significant. Gurjari variety recorded significantly highest fresh weight and dry weight compared to other varieties and the lowest seedling weight was recorded under the variety GR 12.

Figure 1: Effect on germination percentage of rice (10 DAG) by levels of NaCl (salinity) and brassinolide treatments

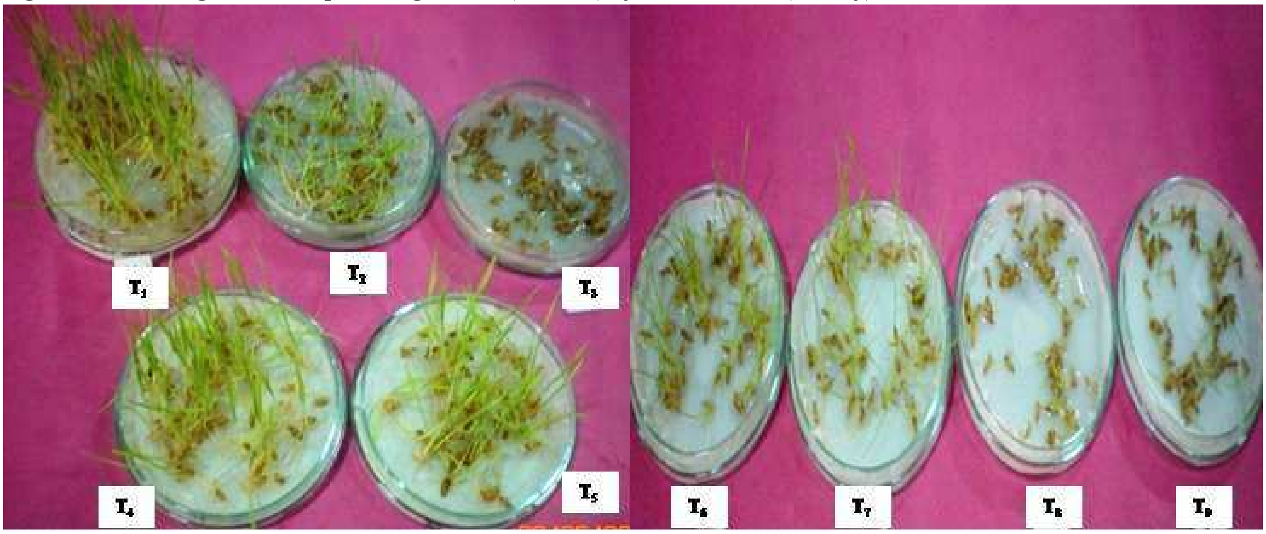

Figure 2: Effect on germination percentage of rice (15 DAG) by levels of NaCl (salinity) and brassinolide treatments

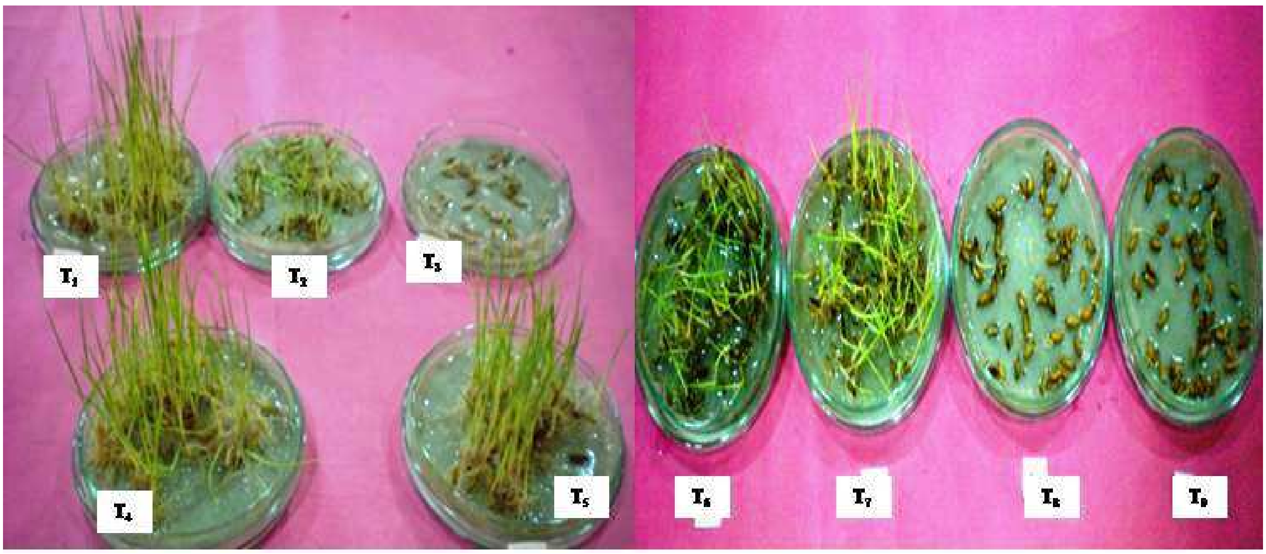

Table 1 Impact of BL on physiological characteristics in rice seedlings under saline stress

Variety (V)	Germination (%)		Fresh weight (mg)		Dry weight (mg)	
	10 DAG	15 DAG	10 DAG	15 DAG	10 DAG	15 DAG
GR-7	86.1	89.5	365.6	383.9	174.6	189.1
GR-11	87.7	90.6	340.5	362.4	157.3	168.8
GR-12	84.1	88.9	306.1	321.4	115.0	131.2
Dandi	84.9	89.1	393.3	418.8	173.4	190.7
Gurjari	80.9	84.8	399.9	424.5	179.4	198.8
S. Em±	0.4	0.4	1.5	1.7	1.05	0.8
CD *(P=0.05)*	1.2	1.2	4.2	4.8	2.9	2.2
Treatments (T)						
T_1-Control	91.3	95.3	457.1	482.7	192.0	204.9
T_2-100 mM NaCl	81.2	85.2	317.4	349.7	149.1	160.5
T_3-200 mM NaCl	67.9	73.5	256.1	270.2	114.3	130.1
T_4-4 µM BL	94.3	96.9	493.2	511.6	203.5	225.2
T_5-8 µM BL	96.3	97.9	498.1	521.9	207.0	230.4
T_6-100 mM NaCl+4 µM BL	84.8	90.2	335.2	358.6	161.2	177.1
T_7-100 mM NaCl+8 µM BL	87.0	92.5	341.2	364.9	164.7	179.3
T_8- 200 mM NaCl+4 µM BL	78.3	80.9	274.4	285.8	123.1	136.4
T_9-200 mM NaCl+8 µM BL	81.5	84.5	277.3	294.6	124.8	137.3
S. Em±	0.6	0.6	2.0	2.3	1.4	1.0
CD *(P=0.05)*	1.6	1.6	5.7	6.5	3.9	2.9
V × T						
S. Em±	1.27	1.24	4.49	5.19	3.15	2.34
CD *(P=0.05)*	3.57	3.49	12.66	14.61	8.88	6.60

Inorganic ions *e.g.*, K^+ and Na^+ play intrinsic role for the development of salt tolerance in plants growing under saline stress condition. It was found that the Na^+ ion concentration was increased where as K^+ ion was decreased with salinity levels. The V × T interaction clearly indicated the increase in Na^+ ion content and decrease in K^+ ion content in all the varieties with saline levels. At 10 DAG and 15 DAG, Dandi showed the highest Na^+ ion content (0.88%) whereas GR-12 (0.43%) and Dandi (0.55%) showed the highest K^+ ion content, respectively. On the other hand, Gurjari recorded the lowest Na^+ and K^+ content (0.74% and 0.32% respectively) at 10 DAG. Consistent results were also found at 15DAG except that GR-7 recorded significantly lowest K^+ content (0.45%). K^+ ion recorded highest under the treatment T_5 (Table-2). Supplementation of NaCl with brassinolide significantly decreased Na^+, thereby toxicity level and increased K^+ in the salt stressed plants of all the varieties. The lower Na^+/K^+ ratio is a good indication of salt tolerance. Brassinolide application helped to promote the process of osmotic adjustment, which is achieved by more accumulation of organic compatible solutes and lower accumulation of inorganic ions. Both Na^+ and K^+ played a significant role to osmoregulatory process. Brassinolide was found to have a positive impact on free amino acids content in the seedlings. Minimum free amino acids content was observed under control condition. Among the different treatment combinations (V × T) significantly the higher order value of free amino acids content was noticed under the treatment combinations V_5T_9 (Gurjari variety treated with NaCl and BL supplementation) and The lowest amino acids content was observed under the treatment combination V_2T_1 (GR-11 varieties treated under control condition). Gurjari recorded the highest free amino acids content significantly and GR-12 showed significantly lowest at both the time intervals. But at 15 DAG, opposite results were noticed in the interaction effect (Table-2). Similar results also reported by Simon-Sarkadi *et al.* (2002); Nguyen *et al*. (2003).

Table 2 Impact of BL on biochemical constituents in rice seedlings under saline stress

Variety (V)	Na^+ (%)		K^+ (%)		Free amino acids (µg mg^{-1})	
	10 DAG	15 DAG	10 DAG	15 DAG	10 DAG	15 DAG
GR-7	0.80	0.97	0.36	0.45	12.83	12.51
GR-11	0.79	0.96	0.39	0.53	12.78	12.49
GR-12	0.87	0.98	0.43	0.54	12.62	12.42
Dandi	0.88	1.13	0.41	0.55	13.75	13.40
Gurjari	0.74	0.96	0.32	0.49	15.94	15.51
S. Em±	0.00	0.00	0.00	0.00	0.05	0.06
CD *(P=0.05)*	0.01	0.01	0.00	0.00	0.16	0.17
Treatments (T)						
T_1-Control	0.14	0.27	0.40	0.55	12.10	11.86
T_2-100 mM NaCl	1.13	1.36	0.31	0.44	13.08	12.81
T_3-200 mM NaCl	1.36	1.78	0.28	0.39	13.30	12.97
T_4-4 µM BL	0.22	0.57	0.46	0.59	13.89	13.62
T_5-8 µM BL	0.23	0.38	0.47	0.60	14.09	13.67
T_6-100 mM NaCl+4 µM BL	0.92	1.06	0.41	0.53	13.81	13.51
T_7-100 mM NaCl+8 µM BL	0.94	1.05	0.41	0.54	13.89	13.55
T_8- 200 mM NaCl+4 µM BL	1.21	1.26	0.35	0.48	14.01	13.68
T_9-200 mM NaCl+8 µM BL	1.19	1.29	0.36	0.49	14.09	13.73
S. Em±	0.00	0.00	0.00	0.00	0.07	0.08
CD *(P=0.05)*	0.01	0.01	0.01	0.01	0.21	0.23
V×T						
S. Em±	0.01	0.01	0.00	0.00	0.17	0.18
C. D. (P=0.05)	0.02	0.03	0.02	0.02	0.48	0.52

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