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SALINITY TOLERANCE IN RICE AS AFFECTED BY APPLICATION OF ABSCISIC ACID AND SALICYLIC ACID

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Abstract

A hydroponic study to evaluate the role of exogenous application of abscisic acid (ABA) and salicylic acid (SA) levels (T_0 = no spray, T_1 = only 1 Mm SA, T_2 = only 2 µM ABA and T_3 = combine spray of 1 Mm SA + 2 µM ABA) against the salt stress on rice grown under saline (70 mM NaCl) and non-saline (0 mM NaCl) was conducted. Evaluation was done on various morphological (root length, shoot length, root fresh, shoot fresh weight, root dry weight, shoot dry weight, root shoot ratio) and biochemical parameters (sodium and potassium contents). Statistical analysis of data revealed that application of ABA and SA significantly improved the salinity tolerance in rice. The response of plant to salinity tolerance was maximum where SA and ABA were applied separately as compared to their combined or no application.

Introduction

Rice (*Oryza sativa*) is the staple food of nearly one-half of world's population, contributes over 20% of the total calorie intake of human beings. But in Asia, where 95% of the world's rice is produced and consumed, it contributes 40 to 80% of the calories to the Asian diet (Alizadeh 2011). Basmati rice is traditionally grown in the Himalayan foothill regions of India and Pakistan (Bligh 2000). Rice has been reported as being salt-susceptible at both vegetative and reproductive stages leading to more than 50% reduction in productivity when grown in a soil having ECe 6.65 dS m^{-1} (Zeng *et al*. 2001).

Environmental abiotic stresses, such as drought, extreme temperature, cold, heavy metals or high salinity, severely impair plant growth and productivity worldwide (Anjum *et al*. 2011). Salt stress in soil or water is one of the major abiotic stresses especially in arid and semi-arid regions and can severely limit plant growth and yield. Excessive quantity of salts in the soil is a major threat to the agriculture as plants face water stress, nutrient imbalance and ionic toxicity (Farouk 2011) which ultimately resulted in yield reduction. Extent of salinity is increasing in Pakistan as it is situated in arid region where evapotranspiration exceeds precipitation and poor quality irrigation water is continuously adding salts into the soil. Salt stress can lead to stomatal closure, which reduces CO_2 availability in the leaves and inhibits carbon fixation, exposing chloroplasts to excessive excitation energy, which in turn could increase the generation of reactive oxygen species (ROS) and induce oxidative stress (Parvaiz and Satyawati 2008). Growth inhibition is a common response to salinity and plant growth is one of the most important agricultural indices of salt stress tolerance (Parida and Das 2005).

Under the scenario of high population growth rate there is increasing demand of food and fibre, it is therefore necessary to get more yield from the marginal soils by improving salt tolerance of crops. Various types of chemicals, such as osmoprotectants, growth regulators and

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stress signaling molecules are being successfully used to induce the tolerance in plants against several biotic and abiotic stresses (Farooq *et al*. 2010).

Many studies have reported the role of salicylic acid (SA) induced stress tolerance in plants. For example, SA has been found to induce drought tolerance in wheat (Singh and Usha 2003). Sakhabutdinova *et al*. (2003) reported that SA induced resistance in wheat seedlings against salinity. In addition, the application of 0.05 mM SA also improves plant growth after salt stress and caused the accumulation of ABA and proline in wheat. However, numerous studies have demonstrated that the effect of exogenous SA application depends on various factors, including the species and developmental stage, the mode of application and the concentration of SA for salt stress alleviation (Horvth *et al*. 2007). ABA is very important agent in the mechanisms of resistance and adaptation in plants against various abiotic stress conditions (Bakhsh *et al*. 2011).

None or very little research work might have been conducted to study the mutual influence of SA and ABA on salinity tolerance of rice. The present study was carried out to evaluate the role of combined application of ABA and SA against the salt stress in rice in comparison to their separate application with the objectives to know the effect of salt stress on rice, to test the efficacy of ABA and SA to induce tolerance against salt stress and to compare the effect of ABA and SA on vegetative growth and biochemical insight of rice plants.

Materials and Methods

To evaluate the effect of SA and ABA on rice, the current experiment was conducted under salinized and non-salinized hydroponic conditions at College of Agriculture, Dera Ghazi Khan, Pakistan, during the month of May, 2012. The experiment was laid out in CRD in factorial arrangement having five replications. Different treatments (T_0 = no spray, T_1 = only 1 Mm SA, T_2 = only 2 µM ABA and T_3 = combine spray of 1 Mm SA + 2 µM ABA) were used through foliar application on rice grown under saline (70 mM NaCl) and non-saline (0 mM NaCl) hydroponio plastic pots containing Yoshida solution as a nutrient medium. For this purpose a nursery of rice (*Oryza sativa* L.) was raised and healthy and uniform size seedlings of 30 days were transplanted into plastic pots. NaCl salt was applied in two equal doses to get the final salt level (70 mM NaCl) in the Yoshida solution. The pH of solution was maintained up to 6 - 6.5, applying acid (H_2SO_4) and base (NaOH). The two chemicals were sprayed three days after salt application. Plants were harvested 32 days after transplanting and the evaluation was done on the basis of various morphological traits (root length, shoot length, root fresh weight, shoot fresh weight, root dry weight, shoot dry weight, root shoot ratio) and biochemical parameters (sodium and potassium contents) were recorded. Plants were separated into root and shoot. The root and shoot length (cm) was measured with the help of meter rod and then root and shoot fresh weight (g) was taken with the help of electrical balance. Root and shoot material was placed in an oven at 70^{o}C to get their constant dry weight. Shoot : root ratio was calculated. For the extraction of Na^+ and K^+, dry leaves of the rice plants were selected and were chopped in to small pieces. Then, 0.5 g chopped rice leaf from each replicate was soaked in test tubes containing 15 ml distilled water and were lid with platinum sheet to save from evaporation losses during boiling process. These test tubes containing soaked plant material, were heated in water bath at 100^{0}C for 20 minutes. The material after cooling was sieved to feed on flame photometer to get Na^+ and K^+ contents in leaf. K : Na ratio was calculated.

Results and Discussion

Salt stress significantly reduced growth and quality parameter except leaf sodium contents which was increased by salt application and application of SA and ABA improved all the growth and quality parameters except sodium contents in the leaf of the rice plant. Data (Table 1) regarding shoot length of rice indicated that salinity (70 mM NaCl) has significantly reduced the shoot length as compared to non-saline (0 mM NaCl). But, ABA and SA application significantly improved the shoot length of rice grown under saline and non-saline media. Data presented that treatment T_1 where only SA (1 mM) and treatment T_2 where only ABA (2 µM) were sprayed, produced higher shoot lengths as compared to T_0 (control spray) and T_3 (combined spray of SA (1 mM) + ABA (2 µM), respectively. Similar trend was observed under stress-full and non-stress conditions. Significant decrease in root length of rice was observed under salt stress. Furthermore, exogenous SA and ABA application showed significant effect on root length of rice under both saline and non-saline conditions (Table 1). Maximum root length was obtained in T_1 where SA and treatment T_2 where ABA was sprayed, respectively in comparison to treatments where hormones were not supplied or applied in combination (Table 1).

Shoot and root fresh weight of the rice reduced significantly under saline in comparison to non-saline conditions. The exogenous SA and ABA application showed significant influence on fresh weight of shoot and root under normal and stress conditions. Maximum fresh weight was obtained where only 1 mM SA and only 2 µM ABA sprayed, as compared to all other treatments (Table 1). Salinity stress reduced the shoot and root dry weight to a significant extent when compared with non-saline conditions. Application of SA (1 mM) and ABA (2 µM) significantly increased the dry weights of shoot and root under normal and saline conditions. The data (Table 1) showed that maximum dry weight was obtained in the treatments where only SA (1 mM) and only ABA (2 µM) sprayed and this value was higher in comparison to treatments where SA (1 mM) and ABA (2 µM) not applied or applied in combination (Table 1).

Shoot : root remained unaffected by SA (1 mM) and ABA (2 µM) application both under saline and non-saline conditions. However, individual spray of SA and ABA showed slightly higher shoot: root as compared to control and their combined application (Table 1).

Na^+ concentration in rice leaves under salt stress was observed to be significantly higher than non-saline conditions. The SA and ABA applications reduced the Na^+ concentration under saline and non-saline conditions. Higher concentration of Na^+ was recorded in T_1 where SA (1 mM) and treatment T_2 where ABA (2 µM) were sprayed respectively in comparison to treatments where SA (1 mM) and ABA (2 µM) were not supplied or applied in combination (Table 2). The concentration of K^+ in leaf of rice plant grown in the presence of NaCl was significantly lower in comparison to that where salt was not added. The application of SA = 1 mM and ABA = 2 µM significantly increased the K^+ concentration both under non-saline and saline conditions. Higher concentration of K^+ was observed where SA (1 mM) and ABA (2 µM) were applied individually in comparison to control and their combined application (Table 2). Presented in Table 2 show that salt stress significantly reduced the $K^+ : Na^+$ in comparison to non-saline conditions. SA and ABA applications enhanced $K^+ : Na^+$ to a significant extent under saline and non-saline conditions. K^+: Na^+ was higher in treatmentsT_1 and T_2 were applied as compared to T_0 and T_3 where hormonal spray was not applied or applied in combination, respectively (Table 2). The data (Fig.1) also shows that dry matter production is positively correlated with reduced Na^+ and increased K^+ uptake.

The exogenous applications of ABA and SA influenced the plant growth both in the presence and absence of salt stress. Growth enhancement by ABA and SA treatment is widely reported in the literature in wheat (Farouk 2011), in rice (Bakhsh *et al*. 2011), in barley (El-Tayeb 2005).

Table 1. Growth parameters of rice affected by exogenous SA and ABA application.

Treatments (T)	Growth parameters													
			Root length (cm)		Shoot fresh weight (g/plant)		Root fresh weight (g/plant)		Shoot dry weight (g/plant)		Root dry weight (g/plant)		Shoot: Root	
	S_0	S_1	S_0	S_1	S_0	S_1	S_0	S_1	S_0	S_1	S_0	S_1	S_0	S_1
T_0 (No spray)	39.00 c	17.00 f	19.00 c	10.00 e	10.40 c	5.20 e	6.40 c	2.80 e	6.40 c	2.80 e	1.00 c	0.80 c	2.05 ab	2.13 a
T_1 (1 mM SA)	44.66 a	24.40 d	24.66 a	13.00 d	14.50 a	9.00 c	10.67 a	6.20 c	10.67 a	6.20 c	4.00 a	2.80 b	1.81 c	1.87 bc
T_2 (2 µM ABA)	45.40 a	23.40 d	26.00 a	13.20 d	15.40 a	9.20 c	11.20 a	6.80 c	11.20 a	6.80 c	4.40 a	2.60 b	1.74 c	1.77 c
T_3 (1 mM SA+2 µM ABA)	42.00 b	20.60 e	22.00 b	8.00 f	12.75 b	7.00 d	9.00 b	4.20 d	9.00 b	4.20 d	2.75 b	1.40 c	1.90 abc	2.06 ab
Means	42.76 a	21.35 b	22.91 a	11.05 b	13.26 a	7.60 b	9.31 a	5.00 b	9.31 a	5.00 b	3.03 a	1.90 b	1.87 a	1.96 a

S_0 = (0 mM NaCl), S_1 = (70 mM NaCl).

Table 2. Quality parameters of rice affected by exogenous SA and AB application.

Treatments (T)	Quality parmeters					
	Na^+ (m mol/g)		K^+ (m mol/g)		K^+: Na^+	
	S_0 (0 mM NaCl)	S_1 (70 mM NaCl)	S_0 (0 mM NaCl)	S_1 (70 mM NaCl)	S_0 (0 mM NaCl)	S_1 (70 mM NaCl)
T_0 (No spray)	8.20 c	13.80 a	4.80 c	1.60 d	0.59 bc	0.11 c
T_1 (1 mM SA)	5.33 ef	6.40 de	9.50 a	6.60 b	1.93 a	1.03 b
T_2 (2 µM ABA)	4.60 f	7.60 cd	10.20 a	6.80 b	2.23 a	0.90 b
T_3 (1 mM SA + 2 µM ABA)	8.00 c	10.20 b	7.25 b	3.40 c	0.91 b	0.24 e
Means	6.53 b	9.50 a	7.93 a	4.60 b	1.41 a	0.57 b

Current results indicate that root length, shoot length, root fresh weight, shoot fresh weight, root dry weight, shoot dry weight, root : shoot, potassium, sodium and potassium : sodium increased due to application of ABA and SA on rice both under saline and non-saline conditions. Although, individual ABA and SA applications improved plant shoot and root length of rice as compared to control and the plant treated with combined ABA and SA. Root length, root number, and root cortex thickness of salt stressed rice seedlings increased as a result of ABA before exposure to soil salinity (Cha-um *et al*. 2007).

Chemicals like ABA and SA enhanced salt tolerance by also altering root growth. However, the response differed depending on both the salt stress regime and the level of salt resistance of the genotype. The individual ABA and SA applications responded significantly in root and shoot fresh weights compared with control treatment and also with those plants treated with combine ABA and SA in current experiment. This improvement in fresh weight of the shoot and root might be due to the improvement in plant water contents by increasing water use efficiency (WUE) and the photosynthetic efficiency by ABA and SA applications under salt stress. The function of ABA in root growth, development, formation of root hair, and lateral roots in rice has recently been reported (Chen *et al*. 2006).

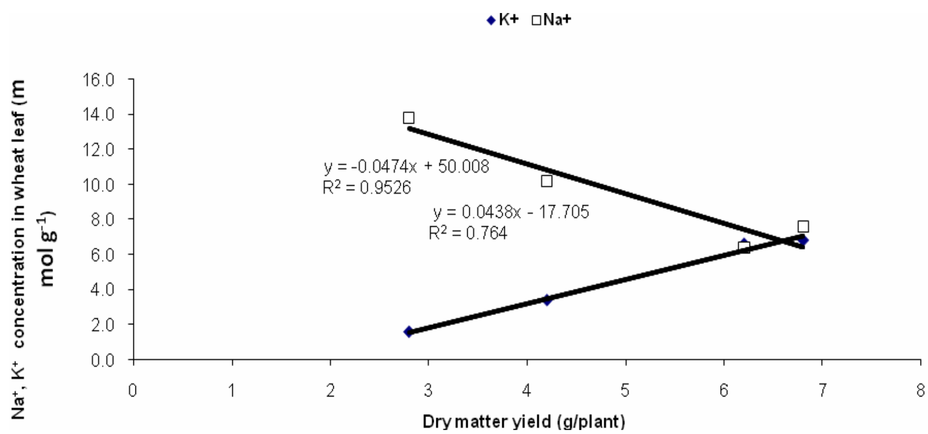

Fig.1. Correlation between dry matter yield (g plant) and K^+ and Na^+ concentrations in wheat leaf under hydroponic saline conditions.

The total shoot dry weight was more where ABA and SA applied separately as compared to control treatment and also in the treatment where both ABA and SA were applied in combination. ABA has been reported to increase growth and total dry weights in *Cucurbita pepo* (Tanino *et al*. 2002). The spray of ABA and SA has been reported to be involved in improving the shoot and root dry weights of the plant by resulting in increased WUE and xylem water potential of barley in the presence of salt (Fricke *et al*. 2004).

The individual ABA and SA application significantly increased the K^+ concentration both under non-saline and saline conditions. The present study indicates that Na^+ concentration in rice leaves was significantly reduced by ABA and SA application. Lower Na^+ concentration in plant body is an indicator of salinity tolerance. The alleviation of Na^+ toxicity might be due to diluted salt effects. It is evident of the present work that K^+ : Na^+ was badly affected by salt stress as compared to non-saline conditions indicating increased uptake of Na^+ as compared to K^+. ABA and SA applications into saline soil enhanced K^+ : Na^+ to a significant extent under saline and non-saline conditions in rice plant.

From the current experiment it is concluded that separate ABA and SA applications significantly increased all growth parameters including growth and dry matter of rice under normal as well as saline conditions as compared to no or their combined spray. The major mechanism responsible for inducing salt tolerance in rice increased K^+ uptake and decreased Na^+ uptake that enhanced K^+: Na^+ selectivity ratio in leaves.

Reference

Alizadeh MR 2011. Effect of paddy husked ratio on rice breakage and whitness during milling process. Aus. J. Crop Sci. **5**: 562-565.

Anjum SA, Xiao-yu X, Long-chang W, Saleem MF, Chen M and Lei W 2011. Morphological, physiological and biochemical responses of plants to drought stress. Afr. J. Agric. Res. **6**: 2026-2032.

Bakhsh I, Awan I, Sadiq M, Niamatullah M, Zaman KU and Aftab M 2011.Effect of plant growth regulator application at different growth stages on the economical yield potential of coarse rice (*Oryza sativa* L). J. Anim. Plant Sci. **21**: 612-616.

Bligh HFJ 2000. Detection of adulteration of basmati rice with non-premium long-grain rice. Int. J. Food Sci. Tech. **35**: 225-265.

Cha-um S, Roytakul S, Sathung T, Maijandang A, TengsiriwattanaS and Kirdmanee S 2007. Exogenous glucose and abscisic acid pretreatment in indica rice (*Oryza sativa* L. spp. *indica*) responses to sodium chloride salt stress. J. Plant Sci. **2**:141-152

Chen CW, Yang YW, Lur HS, Tsai YG and Chang MC 2006. A novel function of abscisic acid in the regulation of rice (*Oryza sativa* L.) root growth and development. Plant Cell Physiol. **47**: 1-13.

El-Tayeb MA 2005. Response of barley grains to the interactive effect of salinity and salicylic acid. Plant Growth Regul. **45**: 212-224.

Farooq M, Wahid A, Lee DJ, Cheema SA and Aziz T 2010. Comparative time course action of the foliar applied glycinebetaine, salicylic acid, nitrous oxide, brassinosteroids and spermine in improving drought resistance of rice. J. Agron. Crop Sci. **196**: 336-345.

Farouk S 2011. Osmotic adjustment in wheat flag leaf in relation to flag leaf area and grain yield per plant. J. Plant Physiol. Biochem. **7**: 117-138.

Fricke, W, G Akhiyarova, D Veselov and G. Kudoyarova 2004. Rapid and tissue-specific changes in ABA and in growth rate in response to salinity in barley leaves. J. Exp. Bot. **55**: 1115-1123.

Horvath E, Szalai G and Janda T 2007.Induction of abiotic stress tolerance by salicylic sacidsignalling. J. Plant Growth Regul. **26**: 290-300.

Parida AD and Das AB 2005. Salt tolerance and salinity effect on plants; a review.Ecotoxicol Environ. saf. **60**: 324-349.

Parvaiz A and Satyawati S 2008.Salt stress and photobiochemical response of plants-a review. Plant soil environ.**54**: 89-99.

Sakhabutdinova AR, Fatkhutdinova DR, Bezrukova MV and Shakirova FM 2003. Salicylic acid prevents the damaging action of stress factors on wheat plants. Bulg. J. Plant Physiol. (*Special Issue)*: 314-319.

Singh B and Usha K 2003. Salicylic acid induced physiological and biochemical changes in wheat seedlings under water stress. Plant Growth Regul. **39**: 137-141.

Zeng L, Shannon MC and Lesch SM 2001. Timing of salinity stress affects rice growth and yield components. Agric. Water Manage. **48**: 191-206.

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