Alleviating Effects of Foliar Spray of Salicylic Acid on Rice (*Oryza sativa* L.) Genotypes Under Wáter Deficient Conditions

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ABSTRACT

Water scarcity is a severe environmental constraint for agricultural productivity. Water stress conditions inside plant body affect many vital physiological and metabolic processes thereby cause losses in crop yield probably exceeds losses from other stresses. Present study is planned to investigate the ameliorating effects of salicylic acid (SA) foliar spray (100 ppm) on rice growth and physiological traits under water deficit conditions. Two rice genotypes (DR 92 and IR 50) with three replicates and six treatments including control (C), C+ SA, 80% field capacity (FC), 80% FC+ SA, 60% FC and 60% FC+ SA were selected for conducting experiment in pots. Salicylic acid foliar sprays were applied on vegetative stage twice with 3 days time interval. Foliar spray of SA enhanced the physiological traits, chlorophyll a, b and total, relative water content (RWC), proline and potassium (K) under control and water stress conditions (80% and 60% FC) in both genotypes. Foliar spray decreased the electrolyte leakage (EL) and sodium (Na) concentrations in both genotypes under water stress conditions. So, on the basis of results it may be concluded that foliar sprays of SA alleviated the negative effects of water deficient stress at vegetative stage and may cause beneficial effects on rice growth and productivity.

INTRODUCTION

Rice is an important staple crop and export commodity mostly grows on irrigated lands of Pakistan covering 2.7 million hectares with per annum production of about 6.5 million tons. Being a high water requiring crop puts huge pressure on already limited water resources of the country. Water scarcity is one of the most important abiotic stress/constraint for rice crop productivity and food security (Waraich *et al*., 2011). Water plays a fundamental role in different physiological functions and developmental processes of the plant. Water deficient conditions primarily disturb tissue water status brings biochemical changes, leads to the formation of secondary metabolites, endogenous reactive oxygen species (ROS) and causes damage to biological membrane (Munne-Bosch and Penuelas 2003, El-Tayeb and Ahmed 2007, Farooq *et al*. 2009a). These malfunctioning in physiological processes ultimately affect rice growth and yield. It is estimated that under extreme water stress conditions the greater 50% losses in yield may occur.

Phytohormones have been recognized as a strong tool for alleviating adverse effects of abiotic stresses. Among the phytohormones salicylic acid (SA) a naturally existing phenolic compound, has been increasingly recognized for improving plant abiotic stress tolerance via control of major plant metabolic processes, plant growth development, ripening and flowering (Erdei *et al*., 1996; Nazar *et al*., 2011; El-Esaw *et al*., 2017). The beneficial role of SA against abiotic stress has been mentioned in many earlier studies e.g., tolerance to high temperature stress in mustard, tolerance against chilling in maize and wheat, heavy metal tolerance in barley, salinity tolerance in mung bean and wheat, drought tolerance in wheat plants (Janda *et al*., 1999, 2014; Metwally *et al*., 2003; Singh & Usha, 2003; Tasgin *et al*., 2006; Waseem *et al*., 2006; Horvath *et al*., 2007;

Farooq *et al*., 2008; Hamid *et al*., 2010; Akhtar *et al*., 2013; Shaheen *et al*., 2018). This present study was therefore carried out to investigate the possible role of SA to induce water stress tolerance in rice.

In present study two rice (*Oryza sativa* L.) genotypes (IR 50 and DR 92) were investigated under water deficient conditions (Control, 80% F.C, 60% F.C) along with salicylic acid (SA) spray (100 ppm).

OBJECTIVES

- To study the physiological responses of rice genotypes under wáter deficient conditions.
- To study the ameliorative effect of foliar application of salicylic acid on physiological traits.

METHODOLOGY

The experiment was conducted in net house at vegetative stage during kharif using two rice (*Oryza sativa* L.) genotypes (DR-92 and IR-50). Surface sterilized seeds were sown in normal soil. Six weeks old seedlings were transferred to plastic pots containing 10 kg soil at different water stress levels (created in pots artificially 80% and 60% field capacity (F.C) along with control (water logged conditions as recommended for rice). Experiment has following groups of treatments C (control), C+SA (control+ 100 ppm salicylic acid), 80% (FC), 80% + SA (100 ppm), 60% (FC), 60%+ SA (100 ppm). Water stress was maintained at 80% and 60% FC by gravimetric method through watering every alternate day or whenever needed for almost five weeks. Hoagland nutrient solution (1/4th strength) was applied with irrigation water two times in a week. SA (100 ppm) was applied twice as foliar spray at vegetative stage. First spray was applied at 56 days old seedling, second spray was applied after 3 days of first spray at 60 days old seedlings, and samples were taken after three days of both spray doses for measuring physiological parameters.

Measurement of electrolyte leakage was done through the method described by Nayyar and Gupta (2006). Leaf samples were washed with distilled water and placed in closed vials containing 10 ml of distilled water 25˚C on a shaker for 24h and subsequently electrical conductivity of the solution (L1) was recorded. The same samples were then autoclaved at 120°C for 20min. After cooling the solution to room temperature, the final electrical conductivity (L2) was recorded.

For relative water content's (RWC) measurements was done by the method of Cornic (1994). Three leaves (flag leaf) from each treatment were taken. Fresh weight (FW) of each sample was recorded using a digital electrical balance (Chyo, MK-500C) and leaves were dipped in test tube containing distilled water for 24 hours. Then leaves were taken out, wiped with the tissue paper and the turgid weight (TW) was recorded. The samples were dried at 65°C for 72 h and dry weight (DW) of each sample was recorded.

Potassium (K) and sodium (Na) was determined using flame photometer (Jenway PFP 7).

Chlorophyll (green pigments) contents were determined by using the method given by Lichtenthaler (1987). 0.1 g fresh leaf sample was taken in 25ml test tube containing10 ml (80%) acetone. These test tubes were kept overnight in dark. Next day the filtrate was run on 663.2, 532.0 and 480 nm at spectrophotometer (Hitachi-150-20, Japan).

Leaf extraction was prepared for proline and total soluble sugars by grinding fresh leaf material of 0.5 g in 10 ml of 3% sulfo-salicylic acid. The sample material was filtered by using Whatman No. 40 filter paper.

The proline was determined according to the method of Bates *et al*. (1973). Two ml of the filtrate was taken in a 25 mL test tube containing 2 mL acid ninhydrin solution (1.25g ninhydrin in 30 mL of glacial acetic acid and 20 mL of 6 M orthophosphoric acid) and 2 mL of glacial acetic acid then test tubes were heated for 1 h at 100oC. Reaction was terminated in an ice bath, the reaction mixture was extracted with 10 mL toluene which form a chromophore. Absorbance of chromophore containing toluene was noted at 520 nm using above mentioned model of spectrophotometer. The proline concentration was calculated by using a standard curve developed by Analar grade proline.

Total soluble Sugar was determined by Riazi *et al*., 1985. The leaf extract (0.1ml) was taken in 25ml test tube then 6 ml of anthrone reagent (anthrone 150 mg was dissolved in 72% H_2SO_4) was poured into each tube and placed in boiling water bath for 10 minutes. The test tubes were then ice-cooled for 10 minutes, and incubated for 20 minutes at room temperature (25°C). Optical density was measured at 625 nm on a spectrophotometer (Hitachi, 220, Japan). The concentration of total sugars was estimated from the standard curve developed with glucose by using the above method.

RESULTS

Salicylic acid spray significantly ($p<0.05$) mitigated all the adverse effects of water stress. Foliar application of SA (100 ppm) increased the concentration of Total chlorophyll (T.ch), chlorophyll a (Chl. a) and chlorophyll b (Chl.b) under control, 80% and 60% FC osmotic stress in both genotypes. Potassium (K%), Relative water content (RWC%) decreased under wáter stress conditions. SA application enhanced these traits positively under stress conditions. Electrolyte leakage (EL%) increased under stress, whereas foliar applications of SA significantly ($p<0.05$) decreased EL% in C, 80%, 60% stress up to 23%, 36%, 40% in IR-50 and 21, 24 and 48% in DR 92 genotype and Proline concentration also improved under stress conditions. Sodium concentrations also decreased by SA application up to 21 and 59% in IR- 50 while 18 and 51% in DR-92 under control and 60% stress. So, it is concluded that foliar application of SA played a vital role in maintaining physiological processes under water stress conditions.

Keywords: Water stress, salicylic acid, rice, biochemical analysis.

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CONCLUSION

Based on the above study it can be concluded that Salicylic acid (SA) mitigates the negative effects of water deficient conditions in plants facing the less availability of water. Salicylic acid enhanced the metabolic activities in plants under stress to cope the situation and help in better growth of plant.

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