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# Effect of salicylic acid on growth and plant water status of sorghum genotype under salt stress

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#### Abstract

Physiological response of sorghum (*Sorghum bicolor* L.) genotype (HJ 513) against the foliar application of salicylic acid (SA) under salt stress was studied. Present investigation was carried out to evaluate the role of SA in mitigating the adverse effect of salt stress. Experiment was conducted in pots under screen house conditions. Salt stress was maintained by saturating the pots with desired levels of salts and foliar spray of SA (25 and 50 mg l<sup>-1</sup>) was made after 40 and 80 days after sowing (DAS). Salt stress adversely affected growth and physiological parameters in sorghum genotype. But foliar spray of SA improved plant growth and SA treated plants exhibited better adjustment under salt stress than untreated plants. Foliar spray of 50 mg l<sup>-1</sup> of SA was more significant compared to 25 mg l<sup>-1</sup>. Salicylic acid was sprayed to plants under salt stress showed higher growth rate and enhanced water status in sorghum plants. All growth parameter *viz.* plant height (cm), fresh and dry weight (g), leaf area (cm<sup>2</sup>) and leaf number showed declined trend but after application of salicylic acid, values were increased from 19.53 to 27.48, 11.36 to 15.65, 1.12 to 1.80, 50 to 78 and 4 to 5 respectively, with respect to control values at 10 dS m<sup>-1</sup>. High salt concentration increases the negative value of osmotic and water potential and decreases the value of relative water content. In nutshell, 50 mg l<sup>-1</sup> salicylic acid significantly ameliorates the deleterious effect of salt stress in sorghum genotype by maintaining the water status of plants.

Keywords: Salt stress, sorghum, salicylic acid, physiological responses, foliar application

#### Introduction

Sorghum genus is a flowering plant which belong to grass family *Poaceae*. It is the fifth most important cereal crop after rice, wheat, maize and barley. In India sorghum crop is popularly known as "Jowar". The grain is also commercially used for making starch, edible oil, dextrose (a sugar), alcoholic beverages and paste. The gluten-free grain is ground into a meal that is made into cakes, flatbreads and porridge. United States and southern Africa mainly grown sorghum for syrup manufacture, forage and sometimes for ethyl alcohol and for production of biofuel. The largest growers of sorghum are India, America and Nigeria. World covers 39.6 million hectare area, producing 57.79 million tones with a productivity of 1404 kg per hectare <sup>[1]</sup>. India contributes 9.45% of the world's sorghum production with 5.82 million hectare area and 5.39 million tonnes of total production <sup>[2]</sup>. The advantage of this cereal crop is that it can be cultivated in both *Kharif* and *Rabi* season and for dual purpose (food and fodder). The major constraints that reduce sorghum productivity are abiotic stresses. Sorghum is moderately salt tolerant, that is well adapted in arid and semi-arid regions where salinity is the major problem. Plant growth and productivity were adversely effected by salinity stress by affecting various physiological processes <sup>[3, 4]</sup>.

Salicylic acid is an important plant bioactive signaling molecule that plays a role in flowering in thermogenic plants, fruit yield, seed germination, ion uptake and transport, glycolysis, stomatal conductance, photosynthetic rate and transpiration. According to Senaratna *et al.*, <sup>[5]</sup>, growth and development of plant is controlled by salicylic acid. Salicin, the glucoside of salicylic alcohol, was isolated from willow bark in 1826 and then converted into an aromatic compound. Oxidation of this aromatic compound leads to the formation of salicylic acid. Application of salicylic acid enhances the cytokinin and IAA levels in plant under salt stress, which cause cell division in apical meristem, so that growth and productivity increases. It also accentuated the rate of photosynthesis by maintaining plant water status. Major significance of SA was observed in agriculture and horticulture in relation to plant tolerance. The systematic studies of sorghum in relation to salt tolerance are relatively few. Therefore, the present study was carried out to mitigate the effect of salinity by foliar application of salicylic acid in sorghum.

## Materials and methods

Seeds of *Sorghum bicolor* L. were collected from Forage Section, Department of Genetics and Plant Breeding, Chaudhary Charan Singh Haryana Agricultural University, Hisar, Haryana (India). Seeds were surface sterilized in 10% sodium hypochlorite solution and grown in pots containing 10 kg dune sand under screen house conditions. Before sowing pots were saturated with desired levels of salinity i.e. 5.0, 7.5 and 10.0 dS m<sup>-1</sup>. The control pots were irrigated with canal water. Hoagland nutrient solution was given at different intervals <sup>[6]</sup>. Salicylic acid (25 and 50 mg l<sup>-1</sup>) was applied exogenously with the help of manual sprayer after 30 and 70 days after sowing (DAS). Sampling was made after 10 days from each spray of SA i.e. 40 and 80 DAS.

# **Growth parameters**

Three plants from each treatment (three replicates) were undertaken to determine average plant height, fresh and dry weight, leaf area and leaf number at each sampling stages. Plant height was measured with the help of meter scale from surface of soil to the tip of the apical shoot. Shoot and root was separated and their fresh weights (FW) were directly determined. For dry weight (DW) determination, the shoot and roots were dried in hot air oven at 65 °C for 48 h and weighed. Leaf area of the detached leaves of each sample was measured with the help of leaf area meter (Model LI 3000, LI COR Ltd., Nebraska, USA) and expressed as sq. cm plant<sup>-1</sup>. Number of leaves per plant was counted and the data were expressed on average number of leaves per plant basis.

#### Plant water status

Osmotic potential was determined using psychrometric technique (Model 5199-B vapour Pressure Osmometer, Wescor Inc. logan, Utah, USA). The third leaf from the top was stored in air tight eppendorf tubes. The leaves were crushed at room temperature. A filter paper disc was immediately dipped in the sap and placed in the concave depression of the sample holder, avoiding the touching of wet disc on the outer surface of the sample holder. The sample slide was pushed gently in to the instrument and sealed the chamber by rotating the knob clockwise. After about one minutes a beep tone was sounded. The osmotic potential reading (mmole kg<sup>-1</sup>) displayed on the digital meter was recorded. The osmometer was calibrated by using osmolarity reference standards of sodium chloride (Wescor Inc, USA) and calculation was done as follows: 1000 mmol  $kg^{-1} = 2.5$ MPa ; 2.5 MPa = 25 bars.

Water potential of leaves was measured with the help of pressure chamber (Model 3005, Soil Moisture corporation, Santa Barbara, CA, USA). The third fully expanded leaf from the top was cut from the plant with the help of sharp edge knife and sealed in the pressure chamber one by one with the cut end protruding outside and the pressure was developed till the sap just appeared at end. That pressure (bar) was recorded as water potential i.e. 1 Bar = 0.1 MPa.

Leaf discs of 200 mg were cut and weighed immediately to record the fresh weight (FW) of the sample. Then, the leaf discs were hydrated to full turgidity by floating on de-ionized water in a closed petri-dish for 3-4 hours at room temperature. After 3-4 hours, the leaf discs were taken out of water and any surface moisture is removed quickly with filter paper lightly and immediately weighed to obtain fully turgid weight (TW). The leaf discs were dried in an oven at 80°C for 24 h and weighed the sample (after being cooled down in a desiccator) to determine dry weight (DW) of the leaf discs. The RWC

(%) was calculated by using the formula given by Weatherly  ${\space{12mu}}^{[7]}_{.}$ 

RWC (%) = 
$$\left(\frac{\text{Fresh weight - Dry weight}}{\text{Turgid weight - Dry weight}}\right) \times 100$$

Statistical analysis was carried out by using OPSTAT programme. Treatments were compared with CD values at 5% level of significance.

## **Results and discussion**

Salt stress induces the deleterious effect on plants by disturbing their metabolism which adversely affect the growth <sup>[4, 8]</sup>, physiological processes directly or indirectly. The previous literature indicated that deleterious effect was mitigated by the some chemicals which may be a hormone i.e. salicylic acid. Successful attempts were also made under salt stress by application of salicylic acid that induces tolerance on the basis of physiological studies in sorghum. The results observed in the present investigation have been discussed here with the recent literature available on the subject with the following heads:

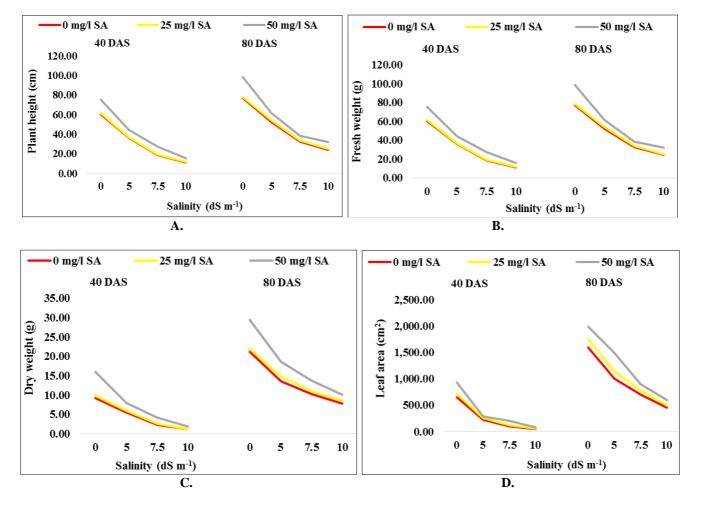
#### **Growth parameters**

Sorghum genotype (HJ 513) treated with salt stress (Fig 3) (control, 5, 7.5 and 10 dS m<sup>-1</sup>), showed significant decrease in plant height (Fig. 1 A), fresh and dry weight (Fig. 1 B and C), leaf area (Fig, 1 D) and leaf number (Fig. 1 E) with the increasing levels of salt stress. For example percent reduction in plant height (74%), fresh weight (81.1%), dry weight (87.7%), leaf area (92.3%) and leaf number (50%) was found highest at 10dS m<sup>-1</sup> of salt stress. The reduction in growth parameters was more at 80 DAS as compared to 40 DAS. Similar results were also obtained by Saied et al. <sup>[9]</sup>. Salts present in the soil reduced the imbibition of water by roots because of low solute potentials that cause changes in the cell metabolism, which was responsible for reduction in plant growth and development. This reduction may be due to harmful effects on physiological processes such as plant water status, ion homeostasis and photosynthesis <sup>[10]</sup>. Significant enhancement was attributed after foliar spray of salicylic acid (SA) i.e. 25 and 50 mg  $l^{-1}$ , but this increment was higher at 50 mg 1<sup>-1</sup> (Fig. 4). Maximum increase in percent was noticed in plant height (27.2%), fresh weight (45.8%), dry weight (78.7%), leaf area (95.7%) and leaf number (33.3%) at 7.5 dS m<sup>-1</sup>of salt stress in HJ 513, after application of 50 mg l<sup>-1</sup> of salicylic acid. The present investigation also depicted that foliar application of SA ameliorate the adverse effects of salt stress. The prevention of water loss from the leaves after foliar application of salicylic acid might be the main reason for the significant increase in growth parameters. This increase in fresh and dry weight of plant under salt stress +foliar application of SA might be due to increase in the nitrate reductase activity and nitrogen status [11]. Salicylic acid also enhance the level of ABA, which was helpful in tolerance mechanism. SA prevent the decrease in rate of synthesis of IAA and cytokinin content and maintained the plant growth <sup>[12]</sup>. The present results were confirmed with previous finding in maize plants <sup>[13]</sup>. Increase in leaf area after foliar application of SA was also confirmed by Khan and Abdullah<sup>[14]</sup>. All growth parameters were found higher at 80 DAS as compared to 40 DAS but trend was same. The interaction between both treatments (salt stress and salicylic acid) was found to be significant at 40 DAS and nonsignificant at 80 DAS.

# Plant water status

A significant reduction in plant water status was observed from control to 10 dS m<sup>-1</sup> of salt stress and values of water and osmotic potential becomes more negative i.e. -0.72 to -1.26 MPa and -0.65 to -0.93 MPa respectively. Value of osmotic and water potential decreased with increasing level of salt stress. Fig. 2 A and B showed that negative value of osmotic and water potential decreases with increasing level of salt stress. This decrease was due to the hydrolysis of macromolecules into smaller molecular like mono and disaccharides, amino acids mainly proline etc. are the main cause of high osmotic potential under salt stress <sup>[15]</sup>. Accumulation of salt ions inside the cell which disturb the osmotic balance in the cell, due to which absorption and translocation of water reduced. Kukreja et al. [16]; Nandwal et al. [17] proposed that value of osmotic potential lowered also due to the decreased availability of water to root, which ultimately reduce the relative water content in chickpea. Relative water content (Fig. 2 C) in terms of percent was reduced with increasing levels of salt stress from control to 10 dS m<sup>-1</sup>. Percent value of RWC i.e. 18.64% decreased under salt stress of 10 dS m<sup>-1</sup> in HJ 513. The decrease in relative water content was due to the accumulation of solute i.e. sodium and chloride ions, which also decreased the osmotic potential, resulting in decreased availability of water to root cells, ultimately lead to decreased relative water content. The present results were confirmed and explain by the precious literature of Kukreja *et al.* <sup>[16]</sup> and Nandwal *et al.* <sup>[17]</sup> in chickpea.

Osmotic and water potential value become less negative after the application of salicylic acid in plants. This increase in water potential was found in my research work after application of salicylic acid in sorghum genotypes, this increase was due to accumulation of high amount of organic and inorganic osmolyte which maintain the water balance by uptake of water. Same results were also obtained by Lee et al., <sup>[18]</sup>, in Arabidopsis. Plant water status in terms of osmotic and water potential was maintained after foliar application of SA and the results were more pronounced at 50 mg l<sup>-1</sup>. Values of osmotic and water potential declined significantly after application of salicylic acid and highest decrease in negative values from -0.91 to -0.74 MPa and -0.89 to -0.78 MPa at 7.5 dS m<sup>-1</sup> of salt level, respectively in HJ 513. Foliar application of salicylic acid also increases the relative water content in HJ 513 genotype of sorghum. Increase in RWC was observed after application of 50 mg l<sup>-1</sup> of salicylic acid i.e. 17.58 % at 10 dS m<sup>-1</sup> of salt stress, it probably occurs due to the membrane stability <sup>[19]</sup>, accumulation of osmolytes <sup>[20]</sup>, which make osmotic adjustment and maintain the desired level of relative water content in tissue.



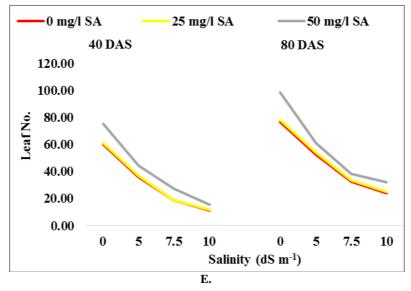


Fig 1: Effect of salicylic acid (25 and 50 mg/l) on sorghum genotype (HJ 513) at 40 and 80 DAS on plant height (A), Fresh weight (B), Dry weight (C), Leaf area (D) and no. of leaves (E) at different salt levels.

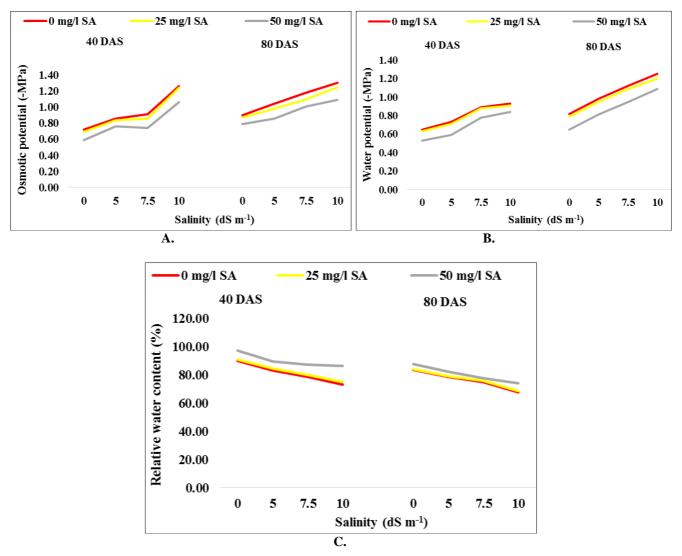


Fig 2: Effect of salicylic acid (25 and 50 mg/l) on sorghum genotype (HJ 513) at 40 and 80 DAS on osmotic potential (A), water potential (B) and relative water content (C) at different salt levels.

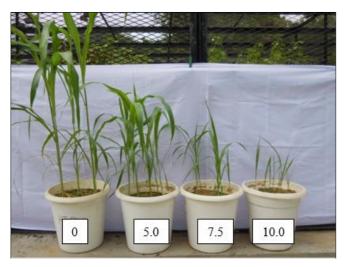


Fig 3: Effect of salt stress (0, 5.0, 7.5 and 10.0 dS m<sup>-1</sup>) on growth of sorghum genotype (HJ 513).



**Fig 4:** Effect of salicylic acid (0, 25 and 50 mg/l) on growth of sorghum genotype (HJ 513) at 10.0 dS mof salt stress.

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