



## Research Article

## Effect of foliar spray with sulfosalicylic acid on morphological and yield traits of chickpea under salinity conditions

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**Abstract:** Salinity is one of the limiting environmental factors for crop production. Chickpea has special importance among the legumes especially in arid and semi-arid regions and is sensitive to salinity. Therefore, it becomes necessary to make a plan to mitigate the salinity effect on this plant. For this purpose, an experiment was conducted in the net house of Department of Botany, Kurukshetra University, Kurukshetra to investigate the role of sulfosalicylic acid (SSA) at different concentrations ( $10^{-4}$ ,  $10^{-5}$  and  $10^{-6}$  M) in overcoming salinity stress imposed on chickpea plants in natural conditions. Different salinity levels (0, 50 mM, 100 mM and 150 mM) were applied and caused a significant reduction in morphological and yield parameters. Our main findings are as follows: (1) Salt stress has detrimental effects on growth and physiology of plants. (2) Application of SSA at  $10^{-5}$  M was the most significant concentration in modulating the inhibitory effects of salt stress.

**Key words:** chickpea, foliar spray, morphological traits, salinity, sulfosalicylic acid, yield traits.

### Introduction

Over 6 % of the world's arable land is affected by salinity, which accounts for more than 800 mhectare of agricultural land (FAO, 2008). In India about 20-27% of world irrigated land and 6.8 mhectare of arable land is salt affected (Singh and Chatrath, 2001). A major problem in almost all the regions of the world, mainly in arid and semi-arid areas is salinity (Munns and James, 2003; Khan and Kaiser, 2006). Amid the stresses, eminent concentrations of salt in the soil can result in harsh detrimental factors, like poor germination, seedling establishment and crop yield (Iqbal and Ashraf, 2006). Plant growth reduction under salt stress is a commonly occurring phenomenon which is chiefly due to low soil water potential and an imbalance in the uptake of mineral nutrients and their accumulation within the plant (Hosseini and Thengane, 2007). Also, salt stress can disrupt growth and photosynthetic processes by causing changes in the accumulation of  $\text{Na}^+$ ,  $\text{Cl}^-$  and nutrients and interruption in water and osmotic potential (Nafees *et al.*, 2010). One of the common stresses is the salt stress which limits the crop plants productivity. Due to high salinity levels in the soil, more than 45 M ha (million hectares) of irrigated land have been damaged. Salt stress reduces osmotic potential of soil solution, induces oxidative stress and imbalance nutrients. It also reduces rate of photosynthesis and enhanced photorespiration rate which leads to an enhancement of production of reactive oxygen species (Parida and Das, 2005).

Generally, legumes have been found to be more sensitive to salinity and chickpea is also highly sensitive to salinity. In chickpea production, India is the world leader. It is an important source of human and animal food due to its high protein content (25.3–28.9% after dehulling). Various strategies have been employed to mitigate the deleterious effect of salt stress. Presently, the recommended strategies to overcome the adverse effects of salt stress include the use of tolerant cultivars, ameliorative water management and diverse cultural practices.

However, none of these approaches have been found to be fully effective under salt stress conditions. An alternative and technically similar approach to induce salt stress tolerance is the exogenous application of PGRs. This technique has gained significant importance during the past decade.

Salicylic acid belongs to a diverse group of plant phenolics. Salicylates from plant sources have been used in medicines since antiquity. In 1928, *Johann Buchner* in Munich, isolated for the first time a small amount of salicin, the glucoside of salicyl alcohol, from willow bar. Ten years later, *Raffaele Piria* named it Salicylic acid from the latin word *Salix* for willow tree. Salicylic acid caused the collapse of the transmembrane electrochemical potential of the mitochondria and the ATP-dependent proton gradient of tonoplast-enriched vesicles (*Macri et al.*, 1986). It promoted flowering in combination with

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other plant growth regulators (e.g. gibberellins). It is an effective inhibitor of ethylene biosynthesis, the effect being pH-dependent. As the salicylic acid contains carboxyl (-COOH) and hydroxyl (-OH) groups the ester derivatives such as acetate esters (acetylsalicylic acid or aspirin), methyl ester (methyl salicylate or oil of wintergreen) and phenyl esters (phenyl salicylate or salol) can be prepared from the salicylic acid. (The Columbia Electronic Encyclopedia, 6th ed. 2005, Columbia University Press.). Several workers reported that gentisic acid treatments caused the accumulation of antifungal PR-proteins such as P-23, P-32 and P-34 in tomato (Rodrigo *et al.*, 1993, Belles *et al.*, 1999). Salicylic acid (SA) and sulfosalicylic acid (SSA) induces resistance to cold stress in seedlings of Pepper (Benavides- Medoza *et al.*, 2002).

Chickpea (*Cicer arietinum* L.), is the third most extensively planted grain legume (D'amore *et al.*, 1996). Besides being an important source of human and animal food, the crop also plays an important role in the maintenance of soil fertility, particularly in arid regions (Saxena, 1990). A major constraint on chickpea production in India is soil salinity, predominately due to chloride and sulphate accumulation. Although some soils are naturally saline, the secondary salinization largely about by the use of irrigation systems, that is the greatest threat to legume sustainability in arid and semi-arid regions, where water supplies are limited, irrigation is essential to improve poor crop yields.

### Material and Methods

The certified seeds of *Cicer arietinum* L. were purchased from CCS Haryana Agriculture University, Hisar. The seeds were surface sterilized with 0.01% mercuric chloride solution followed by inoculation with *Rhizobium* and were sown in earthen pots (0.254 m in diameter) filled with sandy loam soil and farmyard manure (6:1) arranged under a simple randomized block design in the net house of the Botany Department of Kurukshetra University, Kurukshetra during the winter season (November–February). The temperature was 15–25°C and irrigation was done on alternate days during the experiment. At the stage of 30 days after sowing (DAS), the foliage of the plants was sprayed uniformly either with double distilled water (control), with different concentrations ( $10^{-4}$ ,  $10^{-5}$  or  $10^{-6}$  mol/L) of SSA dissolved in ethanol to elucidate the effect of exogenous SSA on plants. The plants were sampled at 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS to assess various morphological parameters and yield parameters.

### Morphological parameters

#### Shoot length

Shoot length was measured from the top of the plant to the base of the root using meter scale and expressed in centimeters.

#### Root length

Root length was measured from the base of the shoot length using meter scale and expressed in centimeters.

#### Plant fresh and dry weight

At each sampling stage, plants under each treatment were taken out carefully from the bags after washing away the sand adhering to roots with copious of tap water and then were separated into their component i.e. stem and root. Then all the separated stems are blotted with filter paper in order to remove moisture from their surface. Then their fresh weight was determined with the help of an electronic balance. All the stems were dried in labeled paper envelopes in an oven at 60° C till a constant weight was attained. Their dry weight was measured with the help of an electronic balance.

#### Number of branches/plant

Ten plants at random were selected from each treatment. Number of branches were counted manually on each branch of the plant and expressed as number of branches per plant.

#### Root fresh and dry weight

At each sampling stage, plants under each treatment were taken out carefully from the bags after washing away the sand adhering to roots with copious of tap water and then were separated into their component i.e. stem and root. Then all the separated roots are blotted with filter paper in order to remove moisture from their surface. Then their fresh weight was determined with the help of an electronic balance. All the stems were dried in labeled paper envelopes in an oven at 60° C till a constant weight was attained. Their dry weight was measured with the help of an electronic balance.

### Yield Parameters

#### Number of pods per plant

At harvest (130<sup>th</sup> DAS), approximate 9 plants (3 from each replicate) representing each treatment were randomly sampled and counted for the number of pods per plant.

#### Number of seeds per pod

25 pods from each treatment were randomly selected and computed to get number of seeds per pod.

#### Seed yield per plant and 100 seed mass

The pods from each replicate were cleaned, crushed and computed to assess seed yield per plant. 100 seeds were subsequently randomly picked per plant and weighed to record 100 seed mass.

#### Pod weight per plant

The pods from each replicate were sampled, cleaned and weighed to record pod weight per plant.

### Statistical analysis

Data in the tables and figures are expressed as mean  $\pm$  standard error. A mean of three reading was taken in every replication. Statistical analysis was done using Statistical Packages for Social Sciences (SPSS) version 16.0. Two-way ANOVA was used to test whether there was a significant difference in various estimations.

## Results

### Morphological parameters

#### Shoot Length

Among SSA treated plants, SSA 2 gave the highest shoot length over control. Application of SSA 2 increased the shoot length by 39.81, 31.23 and 26.12 percent at S<sub>1</sub> and 31.15, 27.18 and 20.58 percent at S<sub>2</sub> and 24.32, 21.81 and 19.88 percent at S<sub>3</sub> over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively. Furthermore, SSA 1 enhanced the shoot length by 24.95, 23.41 and 21.85 percent at S<sub>1</sub> and 19.47, 19.75 and 18.75 at S<sub>2</sub> and 15.28, 18.12 and 15.29 at S<sub>3</sub> over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively (Tables 1-3). However, plants treated with SSA3 had less significant effect on the shoot length over control at highest level of salinity in all the three sampling stages.

#### Root length

Among SSA treated plants, SSA 2 gave the highest root length over control under all salinity levels. Application of SSA 2 increased the root length by 35.38, 30.78 and 23.65 percent at S<sub>1</sub> and 30.16, 23.18 and 18.83 percent at S<sub>2</sub> and 22.09, 17.83 and 13.15 percent at S<sub>3</sub> over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively. Furthermore, SSA 1 enhanced the root length by 32.24, 26.98 and 20.41 percent over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively at lowest level of salinity. Regarding SSA 3, root length increased by 28.62, 24.18 and 16.81 percent at S<sub>1</sub> and 23.35, 21.55 and 17.23 percent at S<sub>2</sub> and 16.24, 14.23 and 11.29 percent at S<sub>3</sub> over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively

#### Number of branches per plant

Among SSA treated plants, SSA 2 gave the highest number of branches per plant over control under all salinity levels. When compared to control, SSA 2 increased the number of branches per plant by 42.83, 35.21 and 29.62 percent at S<sub>1</sub> whereas at S<sub>2</sub> 34.63, 27.85 and 24.13 percent and at S<sub>3</sub> level an increase of 23.18, 16.53 and 13.15 percent was observed on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively. Furthermore, SSA 1 enhanced the number of branches per plant by 35.32, 27.18 and 23.43 percent over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively at lowest level of salinity. Regarding SSA 3, number of branches per plant increased by 29.14, 25.34 and 17.18 percent at S<sub>1</sub> and 23.58,

20.12 and 14.73 percent at S<sub>2</sub> and 19.81, 13.72 and 11.34 percent at S<sub>3</sub> over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively (Tables 1-3).

#### Fresh weight of shoot

The soil administered with NaCl induced a significant reduction in the fresh weight of shoot in chickpea plants. The highest level (150 mM) of NaCl proved most toxic and it decreased the values for fresh weight of shoot by 32.81%, 37.26% and 43.28% at 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS compared with the respective control (Tables 1-3). The whole plant fresh weight increased significantly as plants matured up to 105<sup>th</sup> DAS under normal growth condition. Plants sprayed with SSA1 increased the fresh weight by 43.68, 39.89 and 36.08 per cent at S<sub>1</sub>, S<sub>2</sub> and S<sub>3</sub> over control respectively on 45<sup>th</sup> DAS (Table 1). Similarly, on 75<sup>th</sup> DAS, the same treatments were able to increase the fresh weight by 37.12, 33.18 and 29.94 percent over control (Table 2). However, their effectiveness somewhat slackened as plants matured.

#### Dry weight of shoot

It is evident that dry mass of shoot increased with the advancement in growth. Salinity stress significantly declined the shoot dry weight of chickpea as compared to control on all growth stages. However, out of different levels (50, 100, or 150 mM) of salinity, 150 mM of salinity proved most toxic and reduced the dry weight of shoot by 64.19%, 49.23 and 43.09% than respective control at 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS (Tables 1-3). Foliar spray treatment with sulfosalicylic acid (SSA) significantly enhanced shoot dry weight in salinity stressed chickpea when compared with those of untreated salinity stressed plants, but this enhancement was not above the control. Regarding SSA, SSA2, SSA1 and SSA3 treatments were able to increase the shoot dry weight by 40.16, 35.18 and 28.57 percent over control at S<sub>1</sub> respectively on 105<sup>th</sup> DAS (Table 3).

#### Fresh weight of root

The fresh weight of root decreased significantly as compared to control in chickpea plants under salinity stress on all growth stages and it was 44.18, 48.34 and 48.43 per cent at 150 mM salinity level over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively (Tables 1-3). In contrast to salinity stress, application of SSA significantly increased fresh weight of root in salinity stressed chickpea plants, but this increase was lower as compared to control. Among the groups treated with sulfosalicylic acid, SSA2 exhibited highest fresh weight of root followed by SSA1 and SSA3 and it was 45.63, 35.83 and 27.14 per cent over control respectively on 105<sup>th</sup> DAS at lowest level of salinity (Tables 1-3).

**Table 1:** Effect of SSA foliar treatment on morphological parameters grown under salinity stress at vegetative stage (45<sup>th</sup> DAS).

Treatments		Shoot length (cm)	Root length (cm)	Shoot weight (g)		Root weight (g)		No. of branches/plant
Salinity (mM)	SSA (mole/L)			Fresh	Dry	Fresh	Dry	
0	0	29.70±1.216 <sup>b</sup>	13.32±0.134 <sup>c</sup>	3.29±0.026 <sup>b</sup>	2.25±0.000 <sup>c</sup>	1.55±0.026 <sup>c</sup>	1.09±0.005 <sup>d</sup>	17.04±0.108 <sup>c</sup>
	0	24.29±0.391 <sup>d</sup>	12.56±0.153 <sup>def</sup>	2.51±0.062 <sup>f</sup>	1.83±0.005 <sup>e</sup>	1.32±0.005 <sup>ef</sup>	0.92±0.001 <sup>fg</sup>	13.55±0.453 <sup>d</sup>
50	10 <sup>-4</sup>	30.35±0.165 <sup>b</sup>	16.61±0.092 <sup>ab</sup>	3.29±0.045 <sup>b</sup>	2.36±0.097 <sup>b</sup>	1.67±0.045 <sup>b</sup>	1.23±0.005 <sup>b</sup>	18.34±0.561 <sup>b</sup>
	10 <sup>-5</sup>	33.96±1.318 <sup>a</sup>	17.00±0.370 <sup>a</sup>	3.61±0.036 <sup>a</sup>	2.87±0.117 <sup>a</sup>	1.81±0.051 <sup>a</sup>	1.29±0.036 <sup>a</sup>	19.35±0.802 <sup>a</sup>
	10 <sup>-6</sup>	25.41±0.639 <sup>d</sup>	16.15±0.134 <sup>b</sup>	2.95±0.081 <sup>cd</sup>	2.17±0.072 <sup>c</sup>	1.53±0.036 <sup>c</sup>	1.14±0.026 <sup>c</sup>	17.49±0.504 <sup>c</sup>
100	0	20.65±0.370 <sup>ef</sup>	9.66±0.140 <sup>i</sup>	2.35±0.015 <sup>s</sup>	1.26±0.036 <sup>i</sup>	0.86±0.051 <sup>h</sup>	0.70±0.010 <sup>i</sup>	9.51±0.128 <sup>s</sup>
	10 <sup>-4</sup>	24.67±1.045 <sup>d</sup>	12.09±0.339 <sup>fg</sup>	2.95±0.092 <sup>cd</sup>	1.84±0.010 <sup>e</sup>	1.41±0.026 <sup>d</sup>	0.89±0.010 <sup>g</sup>	11.96±0.386 <sup>f</sup>
	10 <sup>-5</sup>	27.08±0.561 <sup>c</sup>	12.57±0.442 <sup>def</sup>	3.29±0.045 <sup>b</sup>	1.99±0.052 <sup>d</sup>	1.53±0.010 <sup>e</sup>	0.95±0.015 <sup>f</sup>	12.80±0.278 <sup>e</sup>
	10 <sup>-6</sup>	21.79±0.567 <sup>e</sup>	11.92±0.298 <sup>g</sup>	2.73±0.036 <sup>e</sup>	1.68±0.036 <sup>f</sup>	1.35±0.015 <sup>de</sup>	0.78±0.020 <sup>j</sup>	11.75±0.422 <sup>f</sup>
150	0	16.95±0.380 <sup>g</sup>	10.68±0.020 <sup>h</sup>	2.21±0.072 <sup>h</sup>	0.81±0.020 <sup>j</sup>	1.15±0.056 <sup>g</sup>	0.85±0.026 <sup>h</sup>	8.15±0.242 <sup>h</sup>
	10 <sup>-4</sup>	19.54±0.546 <sup>f</sup>	12.73±0.391 <sup>de</sup>	2.90±0.062 <sup>d</sup>	1.48±0.041 <sup>h</sup>	1.31±0.030 <sup>ef</sup>	1.05±0.005 <sup>e</sup>	9.89±0.391 <sup>g</sup>
	10 <sup>-5</sup>	21.07±0.438 <sup>e</sup>	13.04±0.422 <sup>cd</sup>	3.01±0.072 <sup>c</sup>	1.59±0.010 <sup>g</sup>	1.33±0.045 <sup>ef</sup>	1.11±0.030 <sup>cd</sup>	10.04±0.201 <sup>g</sup>
	10 <sup>-6</sup>	17.02±0.442 <sup>g</sup>	12.41±0.468 <sup>efg</sup>	2.59±0.036 <sup>f</sup>	1.27±0.005 <sup>i</sup>	1.27±0.015 <sup>f</sup>	0.95±0.026 <sup>f</sup>	9.76±0.252 <sup>g</sup>
ANOVA F <sub>(12,26)</sub>		542.959	163.296	160.624	155.489	318.003	115.915	222.447
Salinity		511.556	457.103	306.439	998.233	266.668	618.121	964.709
Treatment		176.118	213.040	454.445	413.842	224.474	359.059	138.528
Salinity*treatment		9.681	8.943	5.111	12.082	26.216	9.216	13.371

**Table 2:** Effect of SSA foliar treatment on morphological parameters grown under salinity stress at flowering stage (75<sup>th</sup> DAS).

Treatments		Shoot length (cm)	Root length (cm)	Shoot weight (g)		Root weight (g)		No. of branches/plant
Salinity (mM)	SSA (mole/L)			Fresh	Dry	Fresh	Dry	
0	0	34.31±0.416 <sup>b</sup>	15.39±0.216 <sup>c</sup>	3.79±0.036 <sup>a</sup>	2.73±0.113 <sup>b</sup>	1.83±0.026 <sup>b</sup>	1.34±0.020 <sup>c</sup>	19.68±0.597 <sup>b</sup>
	0	28.06±0.149 <sup>e</sup>	13.09±0.349 <sup>def</sup>	2.74±0.026 <sup>f</sup>	2.15±0.062 <sup>d</sup>	1.45±0.036 <sup>e</sup>	1.13±0.020 <sup>f</sup>	15.65±0.639 <sup>c</sup>
50	10 <sup>-4</sup>	34.63±0.123 <sup>b</sup>	16.62±0.360 <sup>ab</sup>	3.31±0.102 <sup>b</sup>	2.73±0.010 <sup>b</sup>	1.86±0.062 <sup>b</sup>	1.39±0.005 <sup>b</sup>	19.90±0.427 <sup>b</sup>
	10 <sup>-5</sup>	36.82±1.127 <sup>a</sup>	17.12±0.721 <sup>a</sup>	3.76±0.010 <sup>a</sup>	2.96±0.117 <sup>a</sup>	1.99±0.045 <sup>a</sup>	1.55±0.066 <sup>a</sup>	21.16±0.113 <sup>a</sup>
	10 <sup>-6</sup>	28.23±0.153 <sup>e</sup>	16.26±0.103 <sup>b</sup>	2.93±0.040 <sup>de</sup>	2.61±0.102 <sup>c</sup>	1.65±0.036 <sup>c</sup>	1.26±0.030 <sup>d</sup>	19.62±0.051 <sup>b</sup>
100	0	24.55±0.062 <sup>f</sup>	10.23±0.201 <sup>h</sup>	2.55±0.020 <sup>g</sup>	1.74±0.015 <sup>f</sup>	0.95±0.026 <sup>h</sup>	0.86±0.036 <sup>i</sup>	10.83±0.468 <sup>f</sup>
	10 <sup>-4</sup>	29.39±0.612 <sup>d</sup>	12.51±0.407 <sup>ef</sup>	3.14±0.117 <sup>c</sup>	2.17±0.026 <sup>d</sup>	1.39±0.030 <sup>e</sup>	1.05±0.020 <sup>g</sup>	13.45±0.066 <sup>de</sup>
	10 <sup>-5</sup>	31.22±0.113 <sup>c</sup>	12.60±0.535 <sup>ef</sup>	3.39±0.134 <sup>b</sup>	2.86±0.041 <sup>a</sup>	1.44±0.020 <sup>e</sup>	1.11±0.015 <sup>f</sup>	13.85±0.263 <sup>d</sup>
	10 <sup>-6</sup>	24.61±0.736 <sup>f</sup>	12.43±0.201 <sup>f</sup>	2.81±0.081 <sup>ef</sup>	1.94±0.036 <sup>c</sup>	1.17±0.015 <sup>g</sup>	0.98±0.005 <sup>h</sup>	13.01±0.561 <sup>e</sup>
150	0	19.54±0.246 <sup>h</sup>	11.56±0.112 <sup>g</sup>	2.38±0.020 <sup>h</sup>	1.38±0.055 <sup>h</sup>	1.26±0.041 <sup>f</sup>	0.99±0.025 <sup>h</sup>	8.40±0.098 <sup>h</sup>
	10 <sup>-4</sup>	23.08±0.252 <sup>g</sup>	13.35±0.531 <sup>d</sup>	2.96±0.010 <sup>d</sup>	1.59±0.036 <sup>c</sup>	1.52±0.062 <sup>d</sup>	1.09±0.006 <sup>fg</sup>	9.65±0.262 <sup>g</sup>
	10 <sup>-5</sup>	23.80±0.406 <sup>fg</sup>	13.62±0.298 <sup>d</sup>	3.09±0.092 <sup>c</sup>	1.83±0.010 <sup>f</sup>	1.63±0.015 <sup>c</sup>	1.20±0.004 <sup>e</sup>	9.79±0.345 <sup>g</sup>
	10 <sup>-6</sup>	19.59±0.056 <sup>h</sup>	13.20±0.499 <sup>de</sup>	2.73±0.047 <sup>f</sup>	1.58±0.015 <sup>g</sup>	1.43±0.026 <sup>c</sup>	1.06±0.026 <sup>g</sup>	9.55±0.303 <sup>g</sup>
ANOVA F <sub>(12,26)</sub>		258.866	450.152	83.519	111.858	232.315	181.476	145.412
Salinity		1418.241	275.387	292.665	724.975	563.306	447.277	1699.258
Treatment		485.984	98.730	251.185	252.896	270.386	183.398	132.650
Salinity*treatment		17.703	4.422	6.014	24.458	5.154	11.054	16.370

**Table 3:** Effect of SSA foliar treatment on morphological parameters grown under salinity stress at pod filling stage (105<sup>th</sup> DAS).

Treatments		Shoot length (cm)	Root length (cm)	Shoot weight (g)		Root weight (g)		No. of branches/plant
Salinity (mM)	SSA (mole/L)			Fresh	Dry	Fresh	Dry	
0	0	42.68±1.541 <sup>b</sup>	16.45±0.355 <sup>c</sup>	4.56±0.144 <sup>b</sup>	3.16±0.087 <sup>c</sup>	1.92±0.056 <sup>c</sup>	1.46±0.045 <sup>b</sup>	21.04±0.685 <sup>ab</sup>
	0	35.19±0.730 <sup>d</sup>	14.49±0.314 <sup>d</sup>	3.86±0.045 <sup>d</sup>	2.67±0.005 <sup>e</sup>	1.57±0.020 <sup>e</sup>	1.22±0.010 <sup>de</sup>	16.73±0.499 <sup>d</sup>
50	10 <sup>-4</sup>	42.88±0.850 <sup>b</sup>	17.45±0.097 <sup>ab</sup>	4.65±0.072 <sup>b</sup>	3.61±0.134 <sup>a</sup>	2.18±0.021 <sup>b</sup>	1.56±0.036 <sup>a</sup>	20.65±0.411 <sup>b</sup>
	10 <sup>-5</sup>	44.38±0.360 <sup>a</sup>	17.92±0.468 <sup>a</sup>	4.99±0.098 <sup>a</sup>	3.74±0.117 <sup>a</sup>	2.29±0.098 <sup>a</sup>	1.62±0.000 <sup>a</sup>	21.69±0.819 <sup>a</sup>
	10 <sup>-6</sup>	37.50±0.442 <sup>c</sup>	16.93±0.488 <sup>bc</sup>	4.54±0.180 <sup>b</sup>	3.43±0.062 <sup>b</sup>	1.86±0.036 <sup>c</sup>	1.48±0.062 <sup>b</sup>	19.60±0.015 <sup>c</sup>
100	0	27.03±0.288 <sup>g</sup>	11.69±0.233 <sup>f</sup>	2.96±0.081 <sup>fg</sup>	2.11±0.063 <sup>g</sup>	0.99±0.025 <sup>h</sup>	0.97±0.010 <sup>g</sup>	11.51±0.020 <sup>g</sup>
	10 <sup>-4</sup>	32.09±0.144 <sup>e</sup>	13.76±0.561 <sup>c</sup>	3.61±0.087 <sup>e</sup>	2.93±0.092 <sup>d</sup>	1.38±0.020 <sup>f</sup>	1.16±0.030 <sup>ef</sup>	13.87±0.176 <sup>ef</sup>
	10 <sup>-5</sup>	32.59±0.385 <sup>e</sup>	13.89±0.463 <sup>de</sup>	3.74±0.144 <sup>de</sup>	2.98±0.118 <sup>d</sup>	1.67±0.074 <sup>d</sup>	1.21±0.045 <sup>de</sup>	14.29±0.504 <sup>e</sup>
	10 <sup>-6</sup>	28.76±0.442 <sup>f</sup>	13.70±0.428 <sup>e</sup>	3.59±0.072 <sup>e</sup>	2.89±0.072 <sup>d</sup>	1.45±0.058 <sup>f</sup>	1.13±0.046 <sup>f</sup>	13.21±0.227 <sup>f</sup>
150	0	20.27±0.163 <sup>f</sup>	12.07±0.489 <sup>f</sup>	2.58±0.036 <sup>h</sup>	1.79±0.045 <sup>f</sup>	1.39±0.023 <sup>f</sup>	1.11±0.044 <sup>f</sup>	8.96±0.294 <sup>f</sup>
	10 <sup>-4</sup>	23.37±0.823 <sup>h</sup>	13.52±0.561 <sup>c</sup>	3.12±0.123 <sup>f</sup>	2.06±0.087 <sup>gh</sup>	1.66±0.010 <sup>d</sup>	1.23±0.026 <sup>cd</sup>	10.05±0.036 <sup>h</sup>
	10 <sup>-5</sup>	24.29±0.288 <sup>h</sup>	13.66±0.185 <sup>c</sup>	3.65±0.025 <sup>e</sup>	2.42±0.083 <sup>f</sup>	1.73±0.075 <sup>d</sup>	1.29±0.036 <sup>c</sup>	10.14±0.422 <sup>h</sup>
	10 <sup>-6</sup>	20.31±0.127 <sup>i</sup>	13.43±0.288 <sup>c</sup>	2.93±0.056 <sup>g</sup>	1.92±0.081 <sup>hi</sup>	1.68±0.011 <sup>d</sup>	1.20±0.010 <sup>de</sup>	9.98±0.113 <sup>h</sup>
ANOVA F <sub>(12,26)</sub>		541.629	73.544	167.885	167.165	152.263	86.227	393.236
Salinity		2041.051	259.865	590.511	549.545	428.221	283.067	15.37.265
Treatment		204.315	66.379	151.698	161.966	232.227	96.245	90.598
Salinity*treatment		11.583	3.130(NS)	5.244	12.828	18.639	6.738	11.773

**Dry weight of root**

All the plants of the test crop showed noticeable reduction in dry weight of root in presence of salinity. Different levels of salinity (50, 100 or 150

mM) significantly decreased the values for dry weight of root in chickpea plants by 15.59%, 35.43%, and 22.18% at 45<sup>th</sup> DAS compared with

the control plants (Tables 1-3). Moreover, foliar spray treatments with SSA mitigated the inhibitory effects of salt stress. Among the groups treated with SSA, SSA2 recorded highest dry weight of root and it was 41.26, 37.26 and 32.46 percent at S<sub>1</sub> and 35.18, 28.52 and 24.41 percent at S<sub>2</sub> and 30.92, 21.41 and 16.43 percent at S<sub>3</sub> over control on 45<sup>th</sup>, 75<sup>th</sup> and 105<sup>th</sup> DAS respectively (Tables 1-3).

### Yield parameters

#### Number of pods per plant

In response to salinity stress, pods got aborted so their number decreased at different salinity levels and supplementation with sulfosalicylic acid ameliorated salt stress by increasing the number of pods per plant. It is evident from the results that number of pods per plant was significantly increased by sulfosalicylic acid used in the study when compared to control (Table 4) and maximum increase was observed in plants sprayed with SSA2 (48.15 percent) followed by SSA1 (40.63 percent) over control at 50 mM salinity level. SSA2 treated plants also exhibited significant increase in number of pods per plant whereby increasing it by 39.16 percent over control at 100mM salinity level. Besides, less significant differences were found in number of pods per plant among the groups treated with SSA3 at harvest time at 150 mM salinity level.

#### Number of seeds per plant

Salinity stress decreased the number of seeds per plant in chickpea plants and addition of salicylates increased the number of seeds per plant. It is clear from the results that number of seeds per plant was significantly increased by sulfosalicylic acid used in the study compared to control (Table 4) and maximum increase was registered in chickpea plants sprayed with SSA1 (45.19 percent) followed by SSA2 (43.65 percent) over control at 50 mM salinity level. Treatments like SSA2 also exhibited significant increase in number of seeds per plant

whereby increasing it by 35.19 percent over control at 100 mM salinity level at harvest time. On the other hand, less significant differences were found in number of seeds per plant among the groups treated with SSA1 at harvest time at 150 mM salinity level.

#### Seed weight per pod

The obtained results revealed that seed weight per pod was significantly decreased by salt stress and increased by sulfosalicylic acid used in the study compared to control (Table 4) and maximum increase was noticed in plants sprayed with SSA2 (48.15 percent) over control at 50 mM salinity level. Various treatments like SSA2 also exhibited significant increase in seed weight per pod whereby increasing it by 42.18 percent over control at 100 mM salinity level at harvest time. Besides, less significant differences were found in seed weight per pod among the groups treated with SSA1 at harvest time at 150 mM salinity level.

#### 100 Seed weight per plant

Salinity stress depreciated the 100-seed weight per plant in the present study and addition of salicylates elevated the 100-seed weight per plant. It is clear from the results that 100 seed weight per plant was significantly increased by sulfosalicylic acid used in the study compared to control (Table 4) and maximum increase was registered in chickpea plants sprayed with SSA2 (49.73 percent) followed by SSA1 (43.19 percent) over control at 50 mM salinity level. Plants treated with SSA2 also exhibited significant increase in 100 seed weight per plant whereby increasing it by 30.15 percent over control at 100 mM salinity level at harvest time. On the other hand, less significant differences were found in 100 seed weight per plant among the groups treated with SSA1 at harvest time at 150 mM salinity level.

**Table 4:** Effect of SSA foliar treatment on yield parameters grown under salinity stress in chickpea.

Treatments Salinity (mM)	SA (mole/L)	Number of pods/plant	Number of seeds/plant	Seed weight/pod (g)	100 seed weight (g)	Pod weight/plant (g)
0	0	12.95±0.262 <sup>c</sup>	26.35±0.321 <sup>d</sup>	0.582±0.012 <sup>c</sup>	21.416±0.266 <sup>d</sup>	8.672±0.200 <sup>c</sup>
0	0	10.43±0.396 <sup>d</sup>	23.88±0.432 <sup>c</sup>	0.512±0.007 <sup>d</sup>	18.359±0.132 <sup>f</sup>	6.271±0.237 <sup>c</sup>
50	10 <sup>-4</sup>	14.67±0.170 <sup>b</sup>	34.67±1.468 <sup>a</sup>	0.742±0.031 <sup>a</sup>	26.288±1.018 <sup>b</sup>	8.957±0.024 <sup>b</sup>
	10 <sup>-5</sup>	15.45±0.350 <sup>a</sup>	34.31±0.834 <sup>a</sup>	0.759±0.031 <sup>a</sup>	27.489±0.594 <sup>a</sup>	9.415±0.169 <sup>a</sup>
	10 <sup>-6</sup>	13.15±0.411 <sup>c</sup>	31.56±1.252 <sup>b</sup>	0.676±0.028 <sup>b</sup>	24.514±0.265 <sup>c</sup>	7.274±0.124 <sup>d</sup>
100	0	9.43±0.153 <sup>e</sup>	21.27±0.937 <sup>f</sup>	0.408±0.001 <sup>e</sup>	15.058±0.257 <sup>g</sup>	4.386±0.189 <sup>g</sup>
	10 <sup>-4</sup>	13.12±0.163 <sup>c</sup>	28.36±1.225 <sup>c</sup>	0.568±0.018 <sup>c</sup>	19.435±0.420 <sup>e</sup>	5.718±0.231 <sup>f</sup>
	10 <sup>-5</sup>	12.96±0.221 <sup>c</sup>	28.75±0.751 <sup>c</sup>	0.580±0.025 <sup>c</sup>	19.598±0.088 <sup>c</sup>	5.829±0.246 <sup>f</sup>
	10 <sup>-6</sup>	12.74±0.540 <sup>c</sup>	27.89±0.953 <sup>c</sup>	0.489±0.005 <sup>d</sup>	19.224±0.727 <sup>e</sup>	5.697±0.097 <sup>f</sup>
150	0	7.41±0.081 <sup>g</sup>	15.62±0.434 <sup>g</sup>	0.251±0.003 <sup>g</sup>	9.088±0.180 <sup>h</sup>	2.086±0.013 <sup>j</sup>
	10 <sup>-4</sup>	8.16±0.180 <sup>f</sup>	15.69±0.058 <sup>g</sup>	0.259±0.010 <sup>g</sup>	9.094±0.327 <sup>h</sup>	2.092±0.054 <sup>j</sup>
	10 <sup>-5</sup>	8.54±0.201 <sup>f</sup>	16.48±0.607 <sup>g</sup>	0.294±0.010 <sup>f</sup>	9.312±0.067 <sup>h</sup>	2.754±0.034 <sup>h</sup>
	10 <sup>-6</sup>	7.53±0.180 <sup>g</sup>	15.72±0.324 <sup>g</sup>	0.271±0.001 <sup>g</sup>	9.191±0.281 <sup>h</sup>	2.284±0.047 <sup>i</sup>
ANOVA F <sub>(12,20)</sub>		290.128	213.236	300.404	682.785	882.401
Salinity		953.838	727.789	1082.014	2524.572	3345.126
Treatment		233.288	107.226	126.734	199.850	216.019
Salinity*Treatment		36.890	26.382	24.692	62.249	63.472

### Pod weight per plant

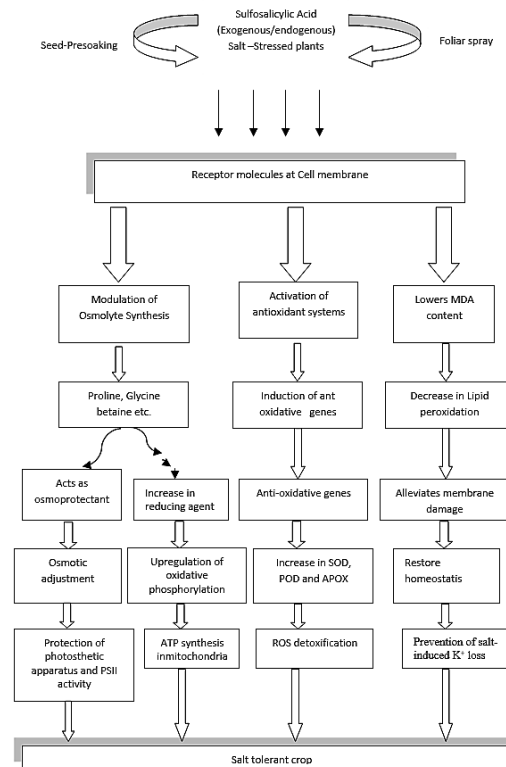
The results obtained in present study clearly indicated that pod weight per plant was significantly increased by sulfosalicylic acid used in the study compared to control (Table 4) and maximum increase was observed in chickpea plants sprayed with SSA2 (50.13 percent) followed by SSA1 (42.83 percent) over control at 50 mM salinity level. SSA2 treated plants also exhibited significant increase in pod weight per plant whereby increasing it by 32.89 percent over control at 100 mM salinity level. Besides, less significant differences were found in pod weight per plant among the groups treated with SSA1 at harvest time at 150 mM salinity level.

### Discussion

The growth parameters (fresh and dry weight of root and shoot, their lengths and number of branches per plant) decreased progressively with the rise of stress level, compared with the control (Table 1). These results are in agreement with those of Ghoulam *et al.*, (2002), who reported that salinity caused a marked reduction in growth parameters of sugar beet plants. Hasanuzzaman *et al.*, (2009) noticed a remarkable reduction in plant height and tiller number in *O. sativa* plants grown in saline soil. Likewise, SA treatment increased the growth of wheat plants under water stress (Singh and Usha 2003), barley (El-Tayeb 2005) and maize (Khodary 2004) under NaCl stress. Reductions in root length with salinity were also reported by Muharrem (2008). Soltani *et al.*, (2002) observed that root length was diminished by increasing the sodium chloride (NaCl) concentration. Plants grown with SA and without salt had more root length as compared with other treatments. Dolatabadian *et al.*, (2011) demonstrated that salinity stress significantly reduced plant height, total biomass and shoot and root weight. A number of studies also mentioned that salinity affects seed imbibitions and led reduction in growth and production of plants (Jamian *et al.*, 2014). Salinity decreased the root length of *Catharanthus roseus* (Jaleel *et al.*, 2007). In *Suaeda salsa*, plant height, number of branches and diameter of shoot were significantly affected by salt stress which was due to increased content of Na<sup>+</sup> and Cl<sup>-</sup> (Guan *et al.*, 2011).

Reduction in root and shoot lengths of plants, is one of the most commonly observed responses of salinity as earlier reported by Bernstein and Hayward (1958). Bruggeman *et al.*, (2003) reported that increasing levels of salinity adversely affected both root and shoots length of chickpea seedling and decreased with salinity. Depression of the shoot: root ratio in response to salinity of wheat plants when different N sources were applied has been reported by Drihem and Pilbeam (2002). Excess of salt concentrations in the root zone causes substantial changes in various morphological and physiological traits at various organizational

levels in the plant (whole plant, tissue and cellular levels). This is achieved through osmotic stress, specific ion toxicities, and ion imbalance (Ashraf and Harris, 2004; Parida and Das, 2005).



**Fig. 1.** Possible mechanism of action of SSA in alleviating salt stress in plants.

Grain yield, the ultimate objective of crop production, was negatively affected by salt stress. Lowest grain yield plant<sup>-1</sup> was recorded for plants that were grown without salicylates and were exposed to salt. The effects of salt stress on plants ultimately lead to reduction of yield of crop which is the most countable effect of salt stress in agriculture. Adriana *et al.*, (2003) reported that a salinity level of 4 dS m<sup>-1</sup> had substantially reduced yield components of chickpea varieties. In the present study, salt stress significantly reduced the growth and biomass yield of chickpea plants (Table 1), which is in harmony with earlier reports on different crops, such as wheat (*Triticum aestivum*) (Kausar *et al.*, 2013), tomato (*Lycopersicon esculentum*) (Abdel Latef and Chaoxing, 2011), pepper (*Capsicum annuum*) (Abdel Latef and Chaoxing, 2014) and rice (*Oryza sativa*) (Mostofa *et al.*, 2015). Baybordi (2009) in their experiment, salinity stress on canola plant, grain weight loss was reported. Reduction in chickpea seed yield by means of terminal drought stress has been previously reported by Behboudian *et al.*, 2001; Fallah *et al.*, 2005). Similar results obtained by Mafakheri *et al.*, (2010) reported that drought stress had considerable effects on the number of pods in chickpea plants. Nahar and Hasanuzzaman (2009) reported that yield

components of *V. radiata* were significantly affected by salinity stress. Number of pods per plant, seeds per pod and seed weight were negatively correlated with salinity levels. Grain yield reduction of rice due to salt stress is also reported by Linghe and Shannon (2000) and Gain *et al.*, (2004). From this study, it can be concluded that SSA can alleviate salinity stress in chickpea plants by modulating antioxidant machinery during the initial growth (figure 1).

### Conclusion

Out of different concentrations of sulfosalicylic acid, SSA<sub>2</sub> was the most effective in the tested parameters at lowest level of salinity. Furthermore, SSA<sub>3</sub> found to be non-significant in number of pods per plant over control at highest level of salinity. SSA<sub>1</sub> had non-significant effect in number of seeds per plant, seed weight per pod and pod weight per plant and over control at highest level of salinity.

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