Efficiency of salicylic acid on germination and growth of chickpea (*Cicer arietinum* L.) under salt stress

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Abstract

The experiment was conducted to study the effect of Salicylic acid ith 4 levels (50, 100, 150 and 200 mg/L and salinity stress by Nacl with4 levels (0.0, 0.1, 0.2 and 0.3 M.) on germination percent, Vigor, germination speed, Plumule Length, Radicle Length, seedling wet and dry weight and the salinity resistance index in chickpea (Cicer arietinum L.).

The experiment dealt with the effect of using the growth regulator "Salicylic acid" (SA) at concentrations: 50, 100, 150 and 200 mg/L under four levels of salinity: (0.0, 0.1, 0.2 and 0.3 Molar (M)). of NaCl, the study showed that the Salinity reduced the germination and growth of chickpea seedlings with the increase in the intensity of the salinity level until it reached 0.0 at the 0.2 and 0.3 M. Salicylic acid reduced the damage caused by salinity, and increased all study parameters under salinity levels, the highest percentage of germination (97.3 and 92.6%) was observed when the concentrations of 100 and 150 mg/L of Salicylic acid.

The salinity resistance index STI showed a significant decrease in this indicator with an increase in the salinity intensity from 1.00 at the level of 0.0 molar NaCl to 0.00 at the level of 0.3 molar NaCl.

Introduction

Chickpea (*Cicer arietinum* L.) is one of the important leguminous crops, and it is one of the most widely consumed legumes and widespread in the tropics, semi-tropics and temperate regions, but drought conditions are one of the most influential factors determining the growth, survival and production of chickpeas (10) Chickpea is the third most important legume crop In production in the world besides dry beans and peas, from a nutritional point of view chickpeas are considered a healthy functional food for its acceptable role in the processing of protein and carbohydrates, which together constitute 80% of the total dry matter weight of the seeds (6) and various sugars, nutritionally important unsaturated fatty acids and B vitamins And various elements (12) that can potentially help reduce the risk of chronic diseases

Climate changes are threatening agricultural production due to the multiplication of abiotic stresses, especially high temperature, drought and increased salinity. The ability of the plant to withstand harsh environmental conditions, research has indicated that SA regulates the signal of the plant’s response to abiotic stress such as drought (7), heat (11) and osmotic stress caused by salinity (4) due to its ability to influence the regulation of hormonal balance and osmotic balance For cells by stimulating some compounds such as proline, glycine, soluble sugars and amines, which maintain osmotic balance, in addition to its role in various physiological processes such as regulating ion uptake and stomata movement (15). Salicylic acid and its chemical composition (Orthro_ Hydroxy Benzoic Acid) are considered plant hormones that produce and play an important role in regulating most plant bio-activities such as plant growth, photosynthesis and flower arrangement. In the last two decades, the acid has been of interest to scientists and researchers because it is related to the defense systems inside the plant against stress, especially under saline conditions, by reducing the production of oxidizing systems and reducing the damage caused by these free oxygen radicals (8).

The study aims to determine the best concentration of salicylic acid (SA) to increase the salt tolerance of chickpea plants (*Cicer arietinum* L.).

Materials and methods:

The experiment was carried out in the laboratory of the Department of Field Crops / College of Agriculture and Forestry / University of Mosul to study the effect of the interaction of salicylic acid and different levels of NaCl on the percentage and vigor of germination and seedling growth of Chickpea (Cicer arietinum L.).

ISSN 2072-3875

The experiment study the effect of two factors: the first; the concentrations of salicylic acid (SA) in four levels (200,150,100,50,0.0 mg/L, and the second factor salt stress by Nacl with four concentrations (0.0, 0.1, 0.2, 0.3 M). Salicylic acid were prepared by dissolving each in a liter of distilled water for each concentration of complete dissolution, while the Nacl was prepared by dissolving a gram molecular weight of NaCl in a liter of distilled water According to the following equation: $M \times MWt = g/L$.

The data were statistically analyzed according to complete random design (C.R.D) using the SAS (2004) .The experiment included the following measurements:

2.1- Germination percentage: The calculation of germination percentage was based on the following equation

Germination % = (number of germinated seeds / total number of seeds used x 100) mentioned before (9).

2.2- Germination Vigor: The germination Vigor was calculated according to the following equation

Germination strength = % germination x (Plumule Length + Radicle Length) according to what is between it (13).

2.3- Germination speed (seed per day): The germination speed was calculated according to the following equation

Germination speed (seeds / day) = (number of seeds germinated / number of days since germination began)

2.4- Plumule Length: The Plumule Length was measured with a 30 cm graduated ruler

2.5- The length of the radicle: The length of the radicle was measured using a ruler 30 cm.

2.6- Seedling fresh weight (mg): The fresh weight was measured after 12 days of germination using a sensitive scale.

2.7 - Seedling dry weight (mg): The dry weight was measured after 12 days of germination using a sensitive balance after drying the samples at a temperature of 70°C for 42 hours using an electric oven.

2.8- Salinity resistance index (STI) index (STI): It was estimated according to the following equation:

(STI) = dry weight of control seedlings x dry weight of salt-treated seedlings.

(Dry weight of seedlings of comparison treatment)2

mentioned by (2)

Results and discussion:

3.1- Percentage of germination (%): Figure (1) shows the effect of salicylic acid concentrations on the percentage of germination of chickpea seeds, which indicates that there were significant differences for all concentrations of salicylic acid that led to a significant increase in the percentage of germination. An average of 42% for the trait in the comparison treatment, while under salinity levels, the results showed that all salt concentrations led to a decrease in proportion to the intensity of salinity, which amounted to 0.0% at levels 0.2 and 0.3 molar NaCl, while 75% at the level of 0.1 molar NaCl, while concentrations of salicylic acid led The inhibitory effect of salt was significantly reduced, and it was 90%, 97.3%, 92.6%, and 75.0% for concentrations 50, 100, 150 and 200, respectively, under 0.1 molar level of salinity, and by increasing the level of salinity, the effect of salicylic acid decreased to be better than the comparison (0.0%). It

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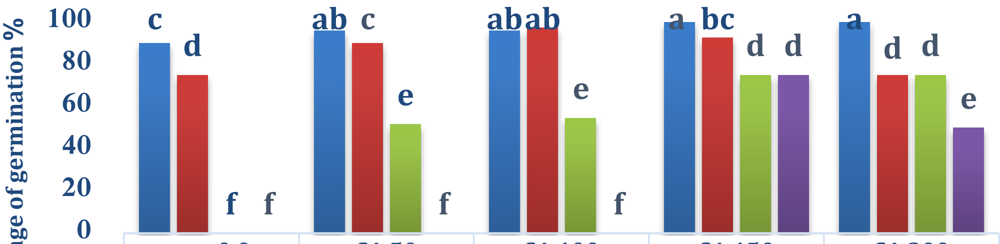
gave a germination percentage of 51.6, 54.6, 75.0 and 75.0 for the above mentioned concentrati ons, respectively.



	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	90	96	96	100	100
NaCl 0.1	75	90	97.3	92.6	75
NaCl 0.2	0	51.6	54.6	75	75
NaCl 0.3	0	0	0	75	50

Figure (1) The efficiency of salicylic acid on the percentage of germination of chickpea seeds under different levels of (NaCl) salt after 9 days.

These results are in agreement with many researchers, including (20) who showed that SA significantly stimulated germination under salinity levels. It was observed under the level of 100-150 mM of NaCl that only 50% of the seeds got germination, but in the presence of (0.05-0.5 mM) of NaCl. SA, the germination rate increased to 80%, the effect of SA stimulating germination under high saline conditions is by reducing oxidative damage through the stimulation of antioxidants (18) and perhaps because of its high regulation of translation, elongation factors and the formation of proteases (19) and (5)) And that salinity leads to an increase in the absorption of Cl and Na and its accumulation in the seed and an increase in its toxic, growth-inhibiting effect due to its obstruction of the vital and plant processes, especially division and elongation, which play the main role in growth. The researcher pointed out that salicylic acid works to remove the effect of salinity toxicity on germination and stimulate the activity of enzymes inside the seed.

3.2- germination Vigor: Figure (2) shows that there are significant differences in the concentrations of salicylic acid in the germination strength of chickpea seeds in a significant way, it was (8.48) in the concentration 50 mg/L and gradually decreased with increasing concentration to 6.2, 6.1, 5.3 at concentrations 100, 150 and 200 mg/L respectively from salicylic acid. Under salinity levels, germination Vigor decreased, reaching 3.1, 0.0 and 0.0 for levels of 0.1, 0.2 and 0.3 molar NaCl. While the interaction of SA with salinity showed a significant effect in reducing salinity damage, especially at 0.1 molar level of NaCl, which reached Germination Vigor was 7.0, 7.1, 5.4 and 4.6 at concentrations 50, 100, 150 and 200 mg/L SA, respectively. At the highest levels (0.2 mol) of salinity, the best was the concentration 150 mg/L of SA

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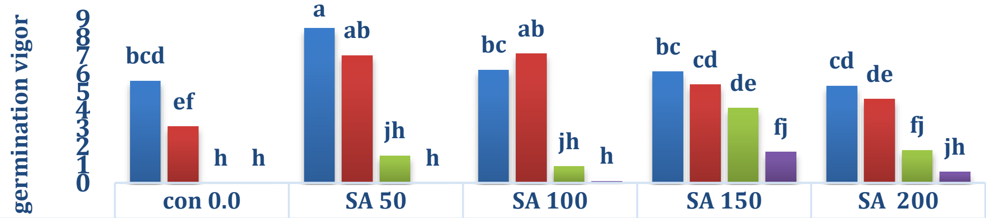
, where the germination Vigor reached 4.1 under this The level of salinity and the effect decreased below the level of 0.3 molar NaCl.



	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	5.6	8.48	6.2	6.1	5.33
NaCl 0.1	3.1	7	7.1	5.4	4.6
NaCl 0.2	0	1.5	0.9	4.1	1.8
NaCl 0.3	0	0	0.1	1.7	0.6

NaCl 0.0 NaCl 0.1 NaCl 0.2 NaCl 0.3

Figure (2) Efficiency of salicylic acid on germination vigor of chickpea seeds under different levels of salinity (NaCl)
9 days later

These results are consistent with what was indicated (19) that salicylic acid (SA) has basic levels that differ greatly between plant species to more than 100 times, and this discrepancy can be observed among members of the same family, either it is an inhibitor of germination or it increases the strength of germination and is due This is due to the concentrations of SA used. Concentrations more than 1 mM (equivalent to 138 mg/L) delay and even inhibit germination, depending on the plant species (18). The negative effect of germination in some types of seeds may be due to the stimulation of SA to oxidative stress. It was observed that the level of hydrogen peroxide (H2O2) increased to three times when treated with 1-5 mM of SA (20). Perhaps the effect of SA in the expansion and elongation of cells is due to its role in increasing auxins. And cytokines, which lead to a rapid and significant increase in cell division in meristematic cells (14).

3.3- Germination speed (seeds/day): Figure (3) shows the effect of salicylic acid concentrations on the germination speed of chickpea seeds under different levels of salinity. There were significant differences for all concentrations of salicylic acid compared to the comparison treatment in the germination speed of chickpea seeds, where the highest rate of this trait at 150 and 200 mg/L was 11.11 The lowest rate was 10.0 seeds. Day -1 in the control treatment, under salinity levels, the germination speed was significantly reduced, especially the high ones (0.2 and 0.3 molar) until it reached 0.0 seeds. Day-1, while 0.1 M concentration showed the least significant decrease in germination speed of 8.33 seeds. Day -1. By using salicylic acid, the negative effect of salinity was decreased. The interaction of SA and salt concentrations showed a significant effect on germination speed of 10.0, 10.8, 10.29 seeds per day when concentrations of 50, 100 and 150 mg/L of SA were interacted with the 0.1 molar level of SA, respectively. Sodium chloride salt, while the effect decreased to 5.7, 6.0, and 8.3 for the above concentrations below the level of 0.2 molar NaCl. and to 0.0 seeds. Day-1 at the level of 0.3 molar NaCl, while concentrations 150 and 200 mg/L reduced the inhibitory effect of salt and showed a speed of 8.33 and 5.56 seeds. Day-1 male (3) The reason for the decrease in the rate of germination and its speed may be due to the inability of the seeds to germinate due to Damage to the embryonic organs and the high osmotic pressure that hinders the absorption of water by the Radicle Length. He pointed out (21) that the negative effects of salinity come through some of the effects it causes, such as water shortage and disturbance of ionic balance

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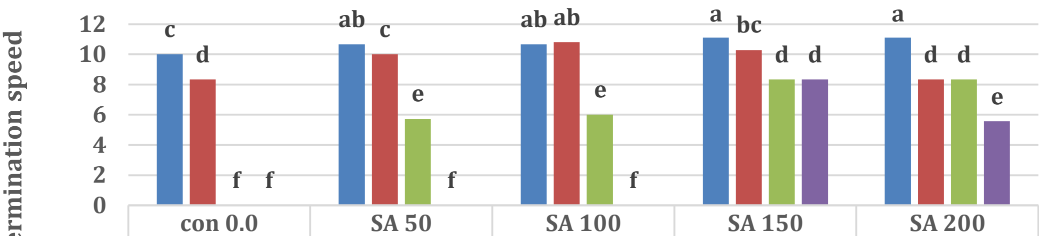
due to the increase of Cl and Na elements, causing a reduction in the rate and speed of germination.

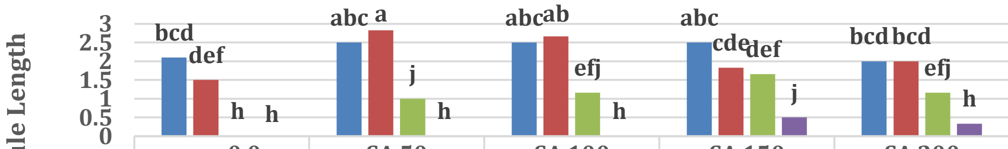


	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	10	10.67	10.67	11.11	11.11
NaCl 0.1	8.33	10	10.8	10.29	8.33
NaCl 0.2	0	5.74	6	8.33	8.33
NaCl 0.3	0	0	0	8.33	5.56

NaCl 0.0 NaCl 0.1 NaCl 0.2 NaCl 0.3

Figure (3) Efficiency of salicylic acid on germination speed in chickpea seeds under different levels of salinity (NaCl) after 9 days

3.4-Plumule Length: Figure (4) shows a non-significant increase in the concentrations of 50-150 mg/L of salicylic acid and all salinity levels showed inhibition of the Plumule Length except for the 0.1 Muller level which reached (1.5 cm) and the comparison was 2.1 cm. While the interactions of SA with the salinity level of 0.1 molar of NaCl showed an increase of the best and significantly was the concentration of 50 mg/L (2.8 cm), while the increase was not significant for the other concentrations when compared to the salt level of 0.1 molar and under high levels of salinity SA reduced the inhibitory effect of salinity For levels of 0.2, 0.3 M, especially concentration 150 mg/L. The negative effect of salinity on germination is related to the processes of nutrient absorption, embryo development, and the occurrence of ionic toxicity, which negatively affects germination and its speed and inhibition of seedling growth. Inhibition with salicylic acid and cause modification in the decrease in the length of the droplet caused by salinity.

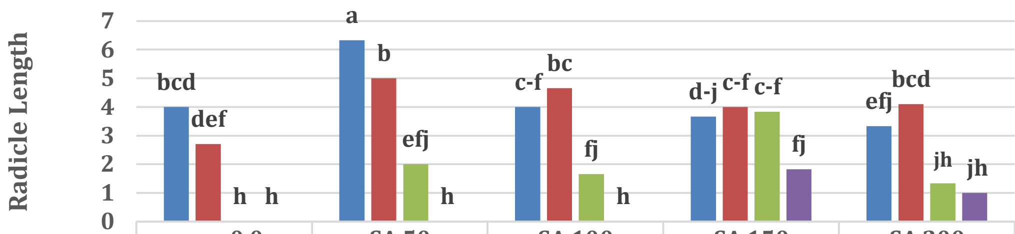


	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	2.1	2.5	2.5	2.5	2
NaCl 0.1	1.5	2.83	2.66	1.83	2
NaCl 0.2	0	1	1.16	1.66	1.16
NaCl 0.3	0	0	0	0.5	0.33

NaCl 0.0 NaCl 0.1 NaCl 0.2 NaCl 0.3

Figure (4) Efficiency of salicylic acid along the Plumule Length of chickpea seedlings under different salinity levels (NaCl) after 9 days (cm)

5.3- Radicle Length: (cm): Figure (5) shows a significant increase at the concentration of 50 and 100 mg/L in Radicle Length under the 0.0 molar salt level, while salinity levels showed a significant decrease in Radicle Length that led to its inhibition at high levels (0.2 and 0.3 molar NaCl), while the interaction of SA with NaCl was reduced. The salinity of the inhibitory effect of the root, especially the concentration 50 and 100 mg/L, was 5.0 and 4.66 cm, respectively, under the 0.1 molar NaCl level, while the effect of SA decreased at the salinity levels of 0.3 and 0.2 and this is consistent with what was observed (20) who found that the length of the root was decreased. with salinity, while salicylic acid increased Radicle Length by 60, 56 and 48% when overlapping with salinity levels of 50, 100 and 150 mM mM, respectively. These results also agree with (5) and this may be due to the role of SA in its effect on stimulating the production of plant hormones of auxins and cytokinins, which leads to a rapid and significant increase in cell division in meristematic cells and an increase in the efficiency of water use and the absorption of nutrients (14).

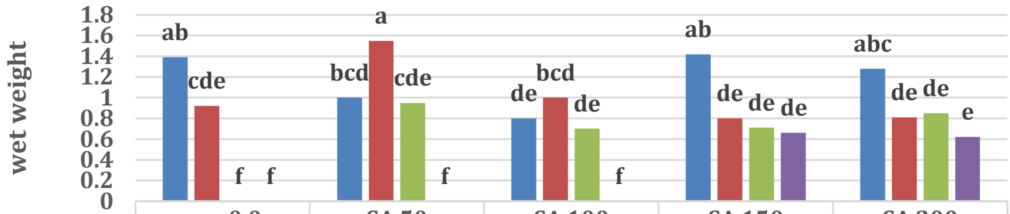


	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	4	6.33	4	3.66	3.33
NaCl 0.1	2.7	5	4.66	4	4.1
NaCl 0.2	0	2	1.66	3.83	1.33
NaCl 0.3	0	0	0	1.83	1

NaCl 0.0 NaCl 0.1 NaCl 0.2 NaCl 0.3

Figure (5) Efficiency of salicylic acid along chickpea seedling Radicle Length s under different salinity levels (NaCl) after 9 days (cm)

3.6- seed Fresh weight (mg): Figure (6) shows that there was no significant effect of salicylic acid concentrations on the fresh weight of chickpea seedlings at the saline level of 0.0 molar, but the concentration of 150 mg/L of SA led to a non-significant increase in the fresh weight of the seedlings, while the salinity levels showed a decrease in the fresh weight of up to Inhibition is below 0.2 and 0.3 M NaCl levels. While SA at a concentration of 50 mg/L promoted a significant increase in fresh weight, while the other interactions of SA concentrations had no significant effect on the characteristic of fresh weight except that they reduced the inhibitory effect of salinity, especially in high concentrations of SA (150 and 200 mg/L), and this is consistent with what It was mentioned (19) that salicylic acid (SA) has basic levels that may differ greatly even between parts of the same plant. The effect of SA stimulating germination and seedling growth under high saline conditions is by reducing oxidative damage through the stimulation of antioxidants, up-regulation of translation, formation of proteases, enzymes of the glycolytic cycle and glycolysis (18), which suggested that SA stimulates release from dormant states. Increasing the soft weight of the seedling by stimulating the production of plant hormones and increasing cell division in meristematic cells rapidly and significantly (14).

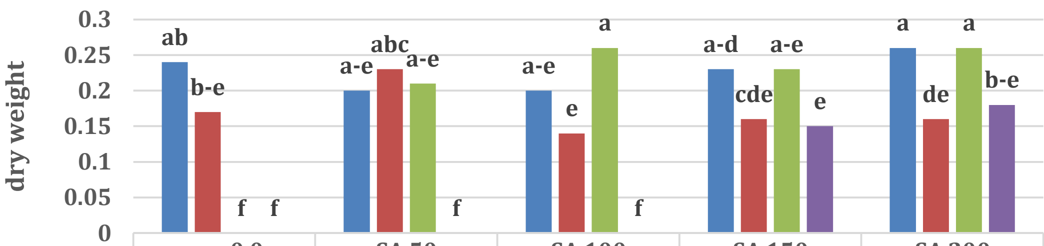


	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	1.39	1	0.8	1.42	1.28
NaCl 0.1	0.92	1.55	1	0.8	0.81
NaCl 0.2	0	0.95	0.7	0.71	0.85
NaCl 0.3	0	0	0	0.66	0.62

NaCl 0.0 NaCl 0.1 NaCl 0.2 NaCl 0.3

Figure (6) Efficiency of salicylic acid on wet weight of chickpea seedlings under different salinity levels (NaCl) after 9 days

3.7- Dry weight of seedling (mg): Figure (7) shows the effect of salicylic acid concentrations on the dry weight of seedling under different levels of salinity, and it was noted from the results that there was no significant effect of salicylic acid concentrations on the dry weight of chickpea seedlings at the saline level of 0.0 molar, but the concentration of 200 mg/L of SA led to An insignificant increase in the dry weight of the seedlings, while the salinity levels showed a significant decrease in the dry weight, which reached inhibition under the levels of 0.2 and 0.3 molar NaCl. However, SA reduced the inhibitory effect of salinity, especially below 0.1 and 0.2 Muller level, and this agrees with what was mentioned previously regarding the fresh weight of chickpea seedlings for the current study and agrees with what researchers (20) and (16) and mentioned (17) that soaking Chickpea seeds with salicylic acid mitigate the negative effects on seedling germination and growth under salt stress by increasing antioxidant enzymes, promoting cell division and increasing the efficiency of nutrient absorption.



	con 0.0	SA 50	SA 100	SA 150	SA 200
NaCl 0.0	0.24	0.2	0.2	0.23	0.26
NaCl 0.1	0.17	0.23	0.14	0.16	0.16
NaCl 0.2	0	0.21	0.26	0.23	0.26
NaCl 0.3	0	0	0	0.15	0.18

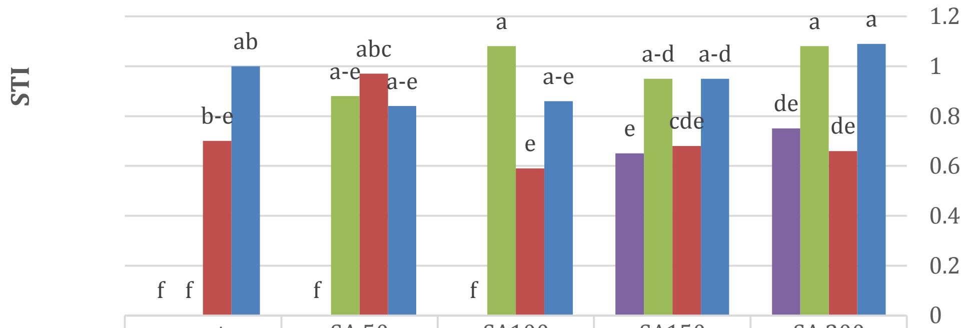
NaCl 0.0 NaCl 0.1 NaCl 0.2 NaCl 0.3

Figure (7) Efficiency of salicylic acid on dry weight of chickpea seedlings under different levels of salinity (NaCl) after 9 days.

3.8- Salinity Resistance Index (STI): Figure (8) shows that salinity negatively affected the growth of seedlings and lowered the salinity resistance coefficient as the salt concentration increased, as the best treatments were the comparison in the absence of salt (1.00), and by increasing the intensity of salinity to 0.1 molar, the salinity resistance index decreased insignificantly (0.7), but by increasing the salinity intensity to 0.2 and 0.3 molar, the salinity resistance index decreased significantly amounting to (0.00). The

ISSN 2072-3875

interaction of salicylic acid with salinity showed a positive effect by mitigating the harmful effect of salinity. Concentrations of 50 mg/L of SA showed an increase in the salinity resistance index of 0.97 under the 0.1 molar salt level, and by increasing the salinity levels (0.2 and 0.3 molar), salicylic acid showed a significant superiority in All concentrations used are from (SA) especially high concentrations; 100, 200,150 mg/l at the salinity level of 0.2 molar reached 0.95, 1.08 and 1.08, respectively, while the concentrations of SA 150 and 200 mg/l showed a significant increase in the resistance value under the salinity level of 0.3 molar. The results of this study indicate the ability of salicylic acid to mitigate the harmful effect of salinity, as the concentrations used showed their resistance to salinity through the salinity resistance index, which increased with the increase of SA concentrations. This indicates that salicylic acid improved the characteristics of seedling growth, including dry weight (Figure 7). This was reflected in the high salinity resistance coefficient (STI).



	cont	SA 50	SA100	SA150	SA 200
NaCl0.0	1	0.84	0.86	0.95	1.09
NaCl0.1	0.7	0.97	0.59	0.68	0.66
NaCl0.2	0	0.88	1.08	0.95	1.08
NaCl0.3	0	0	0	0.65	0.75

Figure (8) Efficiency of salicylic acid on salinity resistance index (STI) of chickpea seedlings under different levels of salinity (NaCl) after 9 days.

The salinity reduced the growth of chickpea seedlings, but salicylic acid significantly enhanced the growth and vigor of germination and agreed with (5) who noted that the 0.1 molar level of NaCl affected the germination. And the growth of seedlings, but their use of salicylic acid mitigated the harmful effect of salinity, and they showed that salicylic acid works to remove the effect of salinity toxicity on germination and stimulate the activity of enzymes inside the seed.

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 ISSN 2072-3875