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Effect of anti-salts in the chemical properties of the pomelo seedlings leaves under salt stress

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Abstract. The study was conducted in the district of Al-Hilla, Al-Hakkana region for the period from 1/9/2020 till 1/5/202 in a fabric canopy to search the effect of anti-salts in the chemical properties of pomelo seedlings under salt stress by spraying the acids (salicylic, ascorbic, proline) at a concentration of 300 mg . L^{-1} in addition to the standard treatment (comparison) by irrigation four levels of saline water, which are (0,2,4,6 ds. $M^{-1})$ using sodium chloride salt, Chemical measurements were carried out(Nitrogen, phosphorous, potassium, iron, proline, chloride, sodium). The results showed the superiority of the treatment with salicylic acid at a concentration of (300) mg . L^{-1} in all the studied characteristics, and ascorbic acid at a concentration of (300) mg . L^{-1} had a significant superiority in the characteristics under study compared with the standard treatment. The experiment was carried out by factor according to the Randomized Complete Block Design (R.C.B. D) and with three replications by three seedlings for the experimental unit with two factors (4 * 4). The number of experiment treatments was (16) treatments distributed randomly to (144) seedlings of one and a half years old, and the results were analyzed using an (ANOVA table) according to the EXCEL program. The statistical differences between the treatments were tested using the least significant difference LSD at a probability level (0.05).

Keywords: pomelo; salinity; antisalinity; stress; chemical properties.

1. INTRODUCTION

Citrus is one of the evergreen fruit trees and belongs to the family Rutaceae, which includes a number of genera, the most important of which are Citrus. It grows in tropical and subtropical regions, and Southeast Asia is its original home and in Iraq cultivation of most Citrus is spread in the central and southern regions [1]. The pomelo fruit (Citrus grandis) is one of the species belonging to the genus (Citrus), as its fruits contain high amounts of carbohydrates, about 17.5%, as well as mineral salts and acids Organic, fiber, vitamins, and low in protein [2],[3]. The salinity prepare is one of the most important problems facing agriculture on a global scale, particularly in arid and semi-arid regions [4]. Iraq Prepare is at the forefront of the Arab and Asian countries in terms of the total area affected by salinity [5], and it is needful to use some alternative technologies at the present time that work to increase the tolerance of pomelo trees to salinity in order to reduce its damage, including salinity antibodies of various types.

Salicylic acid is one of the plant hormones of a nature phenolic and antioxidant, which works to systematize many physiological processes, including flower stimulation, balance of compounds, regulation of ions absorption, stomatal movement, and photosynthesis [6],[7]. Studies have shown that there are many alternative non-salicylic means to improve the salt tolerance of plants, including external treatment with some Osmo protective material such as proline as well as anti-oxidants such as Ascorbic acid [8]. Due to the lack of studies on the use of these compounds in their effect on reducing salt stress on pomelo seedlings and thus on plant growth This study was carried out with the aim of the following:





ISSN: 2789-6773

- 1- The possibility of improving the salt tolerance of pomelo seedlings through the use of different antisaline individually.
- 2- Knowing the true effect of the types of anti-saline individually and determining the best compound of anti-saline compounds (salicylic, prolin, Ascorbic) in giving the best plant growth.
- 3- Knowing the physiological and chemical effects of both salinity and its antagonists used in the experiment.

2. MATERIALS AND METHODS

2.1. Experiment location

The seedlings were obtained from the certified citrus production nursery returning the Iraqi Ministry of Agriculture / General Directorate of Horticulture and Forests in the Holy Karbala Governorate / Al-Hindiyah District. Nearly homogeneous seedlings were selected planted in an anvil 8 kg, consisting of its soil from mixed soil and petmos, and the seedlings were placed in a canopy covered with sarsan. The experimental parameters were applied to them, the service operations were carried out on them, including irragation, removing jungle manually, adding humic acid and duckson by watering method, and treatment of fungi rivus by means of foliar spray in a homogeneous manner throughout the duration of the experiment. The experiment included 144 seedlings of pomelo grafted on the origin of Citrus aurantium and followed the experiment of its workers according to the design of complete random sectors with two factors (4 * 4) and three replications, As the experiment included 16 experimental units, three seedlings for each experimental unit in one replicate, and the factors were as follows:

1- *The first factor*: irrigation with four levels of saline water, which is (0,2,4,6 ds. M⁻¹) using sodium chloride salt, and the concentrations were determined by the EC device, and The salinity levels are symbolized as follows:

 $S0 = 0 \text{ ds. } M^{-1}$

 $S1 = 2 ds. M^{-1}$

 $S2 = 4 ds. M^{-1}$

 $S3 = 6 \text{ ds. } M^{-1}$

2- *The second factor*: spraying with anti-saline at a concentration of (300) mg. L⁻¹ includes (without Spraying, proline, ascorbic acid, and salicylic acid), and these concentrations are within the recommended limits, as the seedlings were sprayed with anti-salinity with 10 sprays between one spray and another (10) days, and symbolizes the antisaline as follows:

A0 = Comparison (without spray)

A1 = Proline acid at a concentration of 300 mg. L⁻¹

A2 = ascorbic acid at a concentration of 300 mg. L⁻¹

A3 = salicylic acid at a concentration of 300 mg. L⁻¹

The experiment was completed a month after the date of the last spray, in which samples were taken for analysis and the results were recorded, then the variance was analyzed by the EXCEL program, and the differences between the averages were compared according to the test of Less significant difference (L.S.D) at a probability level of 0.05 [9].

2.2. Studied traits

- 1. Nitrogen: Estimation of it using the Microkjeldahl device described before [10].
- 2. *Phosphorous*: It was determined using ammonium molybdate and ascorbic acid, and the measurement was taken by a Spectrophotometer at a wavelength of 620 nm [10].
- 3. Potassium: Determination of sodium and potassium content of leaves, stems and roots in the digested sample as reported in [11], by using the Flame Photometer.





ISSN: 2789-6773

- 4. Iron: was determined By the Absorption Atomic Spectrophotometer according to the method [12],[13].
- 5. Proline: was determined by Following the method of [14].
- 6. chloride: The chloride content of leaves, stems and roots was estimated in the digested sample diluted by calibration with (0.05) standard silver nitrate using potassium chromate index [15].
- 7. Sodium: Estimate the sodium and potassium content of leaves, stems and roots in the digested sample, as mentioned in [11] using the Flame Photometer.

3. RESULTS AND DISCUSSION

3.1. Effect of anti-salts under salt stress on chemical properties

It is evident from Tables (1,2,3,4) that there is a significant effect of anti-salts in all chemical characteristics except for potassium mentioned in Table No. (3) as the treatments of salicylic acid were significantly superior, then ascorbic, then proline and then comparison, respectively. The effect of salicylic acid gave the highest content of (nitrogen, phosphorous, potassium, iron) in the leaves, while the treatment of ascorbic acid had a significant effect compared with the standard treatment.

The effect of salicylic acid being one of the plant hormones can encourage the building of the hormones gibberellin and auxin and may also encourage the process of gene expression to build many chemical compounds, including auxins and gibberellins[16] in addition to encouraging it in building carbohydrates and chlorophyll resulting from increased growth.

The effect of ascorbic acid being an antioxidant maintains the important enzymes that prevent the destruction of chlorophyll and increase growth by increasing the content of the leaves of carbohydrates. This may be attributed to the role of salicylic acid in increasing the absorption of ions, including nitrogen [17], in the form of nitrate and increasing the effectiveness of the enzyme Nitrate reductase [18], and this may be attributed to the role of salicylic acid in increasing the efficiency of the photosynthesis process and increasing the absorption of ions [19] .These results are consistent with findings [20] in apple seedlings.

The same results also showed that the salinity of irrigation water has a significant effect on the chemical properties, as the high concentrations caused a decrease in the rate of characteristics, and the lowest rate was recorded when irrigation with the salinity level (6) ds.M⁻¹ while the highest rate was recorded when irrigation with the salinity level (2) dS. M⁻¹ which significantly surpassed the rest of the treatments.

The results of Table No. (5) showed the presence of a significant effect of anti-salinity, as the treatment of proline acid significantly outperformed the rest of the treatments in the rate of proline, that the effect of proline acid gave the highest content of proline in the leaves, followed by salicylic and ascorbic treatments and the comparison respectively, and the results agree with [21].

The high content of proline in leaves due to its relationship with the regulation of osmotic pressure, as exposure of plants to environmental stresses leads to the accumulation of some nitrogenous compounds as an adaptive phenomenon, including proline because it is osmotic-activated as it restores the balance of the coenzyme NADP and NADPH or works to protect the enzymes from the risk of water stress or saline [22], as it is compatible with components of the cytoplasm and is used by plants as a nitrogen reserve [23].

Tables (6,7) (chloride ion, sodium) show that the salinity of irrigation water has a significant effect on chloride and sodium ions, as the treatment (S3) was superior to irrigation at the level of salinity of 6 dS. M⁻¹ reached the lowest rate when irrigating the salinity level (0) dS. M⁻¹, and these results agree with [24], and a significant effect was found for antisaline, as the comparison treatment (A0) was significantly superior to ascorbic, salicylic, and proline, respectively, and gave the highest values for chloride and sodium ion rate.





ISSN: 2789-6773

Table 1. The effect of anti-salts under salt stress in the percentage of Nitrogen (%) in leaves

salts anti-salts	S0	S1		S2	S3	rate
A0	1.640	1.700		1.630	1.510	1.620
A1	1.760	1.850		1.710	1.640	1.740
A2	1.940	2.030		1.820	1.710	1.875
A3	2.110	2.270		2.060	1.890	2.083
Rate	1.863	1.963		1.805	1.688	
L.S.D 0.05	S=0.027			A=0.027	S*A=	0.054

Table 2. The effect of anti-salts under salt stress in the percentage of Phosphorous (%) in leaves

salts anti-salts	S0	S1		S2	S3	rate
A0	0.386	0.3	91	0.378	0.367	0.381
A1	0.402	0.414		0.395	0.381	0.398
A2	0.411	0.422		0.406	0.393	0.408
A3	0.452	0.464		0.443	0.411	0.443
Rate	0.413	0.423		0.406	0.388	
L.S.D 0.05	S=0.003			A=0.003	S*A=	=0.007

Table 3. The effect of anti-salts under salt stress in the percentage of potassium (%) in leaves

salts anti-salts	S0	S1		S2	S3	rate
A0	1.171	1.036		1.096	1.031	1.151
A1	1.159	1.197		1.141	1.092	1.147
A2	1.181	1.214		1.163	1.117	1.169
A3	1.209	1.245		1.192	1.146	1.198
Rate	1.180	1.240		1.148	1.097	
L.S.D 0.05	S=0.057	,		A=n.s	S*/	4=n.s

Table 4. Effect of anti-salts under salt stress in the iron content in leaves (mg. Kg -1 dry weight)

salts	S0	S	1	S2	S3	rate
anti-salts	11.5			• • •		
A0	41.6	43	5.4	39.6	37.5	40.5
A1	43.8	45.6		42.4	39.4	42.8
A2	44.4	446.4		43.5	40.2	43.6
A3	45.5	47.4		44.2	41.5	44.7
Rate	43.8	45.7		42.4	39.7	
L.S.D 0.05	S=0.244			A=0.244	S*A=n.	S





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Table 5. The effect of anti-salts under salt stress in the proline content in leaves (mg.g⁻¹ dry weight)

salts	S0	S1	S2	S3	rate
anti-salts					
A0	2.2	3.1	4.2	5.0	3.6
A1	2.7	4.6	5.8	6.5	4.9
A2	2.4	3.4	4.6	5.3	3.9
A3	2.5	4.0	5.1	5.8	4.3
Rate	2.4	3.7	4.9	5.7	
L.S.D 0.05	S=0.057		A=0.057	S*A=0.1	113

Table 6. Effect of antisalants under saline stress in the chloride ion content in leaves (%)

salts anti-salts	S0	S1	S2	S3	rate
A0	1.2	1.5	2.3	2.6	1.9
A1	0.6	0.8	1.4	1.8	1.1
A2	1.0	1.3	1.9	2.3	1.6
A3	0.9	1.0	1.7	2.0	1.4
Rate	0.9	1.2	1.8	2.2	
L.S.D 0.05	S=0.11		A=0.11	S*A=	n.s

Table 7. Effect of antisalants under saline stress in the sodium ion content in leaves (%)

salts anti-salts	S0	S1	S2	S3	rate
A0	0.203	0.247	0.352	0.424	0.307
A1	0.148	0.172	0.269	0.340	0.232
A2	0.188	0.223	0.337	0.406	0.289
A3	0.170	0.201	0.291	0.386	0.262
Rate	0.177	0.21	0.312	0.389	
L.S.D 0.05	S=0.006		A=0.006	S*A	=0.012

4. CONCLUSIONS

- 1- Irrigation of pomelo seedlings with water at a salinity level of 0, 2 dS.m⁻¹ gave good vegetative growth and did not show symptoms of nutritional deficiency compared to seedlings irrigated with water of 4, 6 dS.m⁻¹,. Which negativel affected in the leaves content of mineral elements (nitrogen, phosphorous, potassium, iron), which had a significant impact on the increased of proline accumulation of in Leaves as a result of osmotic stress, and the intensity of the effect increases with increasing levels of salinity, which makes us conclude that they can be used as indicators of the tolerance of pomelo seedlings to salinity in later studies.
- 2- Salicylic acid treatment led to an improvement in the mineral content of leaves, nitrogen, phosphorous, potassium, iron) for seedlings under experimental conditions.
- 3- The results showed that spraying seedlings with salicylic acid was more clear and effective in reducing the harmful effect of salinity of irrigation water, followed by spraying with ascorbic and then proline.





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