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Effect of Salt Stress and Salicylic Acid on Growth of Marigold (*Tagetes erecta* L.) Plants

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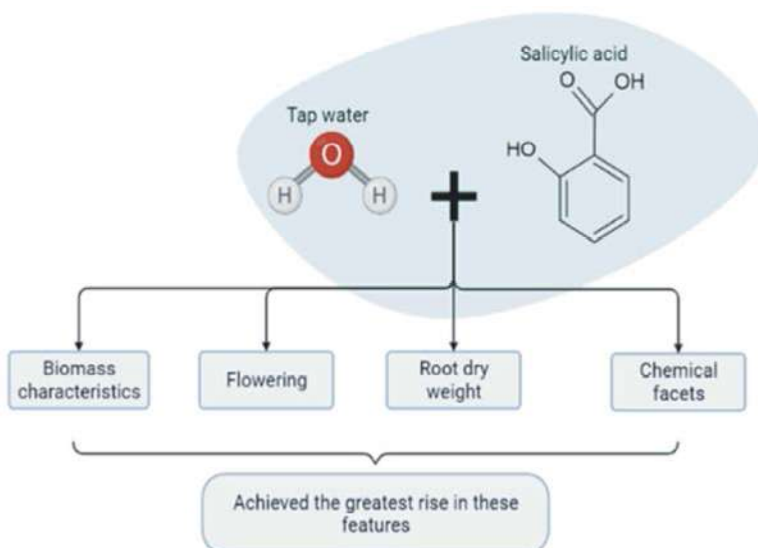
Abstract: The current study was conducted on marigold plants (*Tagetes erecta, L.*) grown in pots in Antoniadis Gardens, Research Branch and Ornamental Plants in Alexandria - Egypt during the 2019 and 2020 growing seasons to explore the effect of salicylic acid at concentrations of 0.00, 25.0, 50.0 and 75.0 mg. l^{-1} as a foliar spray on vegetative growth, flowering and chemical major constituents of Marigold under several EC, 0 (tap water), 2.0, 4.0 and 6.0 dsm^{-1} of irrigation water salinity stress. The results showed that raising the salicylic acid concentration led to significant increases in all growth indices. Plant height (64.37cm), number of branches per plant (18.05), number of leaves (300.34), dry weight of leaves per plant (9.41g), number of flowers per plant (14.38), flower fresh weight per plant (8.16 g). The amount of chlorophyll A (21.22) and B (23.64) in leaves (mg.100 g^{-1} F.W.) and the percentage of total carbohydrates in leaves (12.19 %) were both enhanced. Adding 75.0 mg. l^{-1} of salicylic acid per plant, paired with EC and tap water irrigation, achieved the greatest in these features. Increased salicylic acid treatments at any irrigation water salinity level contributed to a significant decline in Na^+ content, proline content and membrane stability index in leaves *Tagetes erecta* L. plant.

Key words: Marigold · Salt stress · Ssalicylic acid · Membrane stability index · Proline



Schematic diagrams for the both factors. (A)Water salinity (tap water=control; 2, 4 and 6 $dS.m^{-1}$). (B) Salicylic acid (SA) (0.0, 25, 50 and 75 mg. l^{-1}).

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INTRODUCTION

The Marigold plant *Tagetes erecta* L. is a blooming plant that blooms during the summer months (family: Asteraceae). Tagetes is a genus containing about 40-50 species that can be found in Mexico and Guatemala, as well as other nations [1]. Thousands of distinct cultivars in lovely yellow and orange hues are widely cultivated as an easy-to-grow bedding plant for mass display, pots, borders and window and porch boxes [2]. The florets in the heads are generally hermaphrodite. Oil glands may be found on the leaves of all marigold species and the blooms are employed in pharmaceuticals, processed foods and the confectionery sector. Its essential oil is known as an antiparasitic, antispasmodic, antibiotic, antibacterial and antiseptic [3]. Also, all marigold species have oil glands on their leaves; the oils are pungent [4]. It grows quite well in clay and sandy soils with enough good drainage and bright sun [5]. Most crop plants are thought to be harmed by saline water irrigation. When grown under saline irrigation, even salt-resistant plants produce less. However, because saline water is abundant in various parts around the world, it is self-evident that it is necessary to develop agro-administration programs that include salty water. Salinity has been identified as a key element impacting crop productivity and agricultural sustainability in arid and semi-arid regions. On the other hand, irrigation with saline water without better monitoring, such as adding fresh water, would result in soil deterioration, which would have a detrimental influence on crop production and land management [6]. Because of their effect on quality and production, salinity is regarded as one of the most significant constraints on crops around the world Ali *et al*. [7], Li *et al*. [8] and Rodríguezes *et al*. [9] found that irrigating maize plants with saltwater or increasing the salt concentration in the soil solution negatively influenced growth and yield. Plant height, leaf area and the fresh and dry weights of aerial organs, as well as the number of flowers flower diameter, the dry weight of flowers and the number of buds, are all affected by salt stress. According to Kamkari *et al.* [10] increasing salt stress to 200 mM NaCl on pot marigold decreased the above-mentioned features by 89, 73, 94, 52, 90, 46, 78, 67, 83 and 69%, respectively. The effects of salt on lavender *(Lavandula angustifolia Miller*) were investigated by Khorasaninejad *et al*. [11]. They found that salinity stress of 0, 25, 50, 75 and 100 mM NaCl lowered plant development metrics significantly (stem length, shoot wet weight and root wet and dry weight). Taher *et.al.*, [12] treated sunflower cultivars for 30 days under ideal conditions using 50, 150 and 250 mM NaCl, they found that salt stress lowered characteristics like plant height, leaf area and fresh weight in all types. Salicylic acid (SA) is an endogenous phenolic compound produced by root cells in a large number of plants and it plays a variety of roles in plant growth and development as a para-hormonal substance that regulates a variety of physiological processes in plants Shakirova, *et al.* [13], including plant growth and development, ion uptake, photosynthesis, germination and defensive responses [14]. It's thought to act as a natural thermogenesis indicator inducing flowering in many plants and controlling ion uptake by roots and stomatal transmission [15]. Plants' physiological and biochemical traits are also regulated when they are exposed to abiotic stress [16]. Under stressful conditions, SA enhances pigment content and reduces the negative effects of stressors [17].

As a result, spraying salicylic acid can protect plant against a variety of stresses, including salinity. Salicylic acid is a crucial component of plant disease resistance and participates in the plant response to harsh environmental conditions, according to several studies. Yildirim *et al*. [18] investigated the effects of foliar salicylic acid (SA) treatments (0.0, 0.25, 0.50 and 1.0 mM) on cucumber growth, chlorophyll content and mineral content under salt stress (0, 60 and 120 mM sodium chloride) (NaCl), they discovered that foliar sprays of SA led in higher shoot fresh weight, shoot dry weight, root fresh weight and root dry weight, as well as plants that were less salt stressed. Under salt stress, SA treatments boosted shoot diameter and leaf number per plant. In either saline and non-saline regimes, 1.0 mM SA treatment had the highest chlorophyll concentration. Zarghami *et al*. [19] investigated the effect of salicylic acid on petunia plant stress tolerance. The use of 2.0 mM salicylic acid also reduced electrolyte loss.

The major goal of this study would be how three different concentrations of salicylic acid (SA) and two various rates of saline irrigation affected vegetative growth, blooming and chemical constituents of *Tagetes erecta* L. plants alone and in combination throughout the growth period.

MATERIALS AND METHODS

Plant Materials and Growing Conditions: The pot empirical study was carried out at Antoniadis Research Branch, Horticulture Research. Institute, Alexandria, Egypt throughout the two successive growing seasons of

2019 and 2020. The aim of this study the effect of foliar spray of salicylic acid (SA) with different concentrations on plant growth, flowers production and the chemical constitutes such as chlorophyll, carbohydrates, Na^+, proline and electrolyte leakage of *Tagetes erecta* L. (local variety, orange flowers) plants grown under salt stress. Seeds of *T. erecta,* L. for the two years' experiments were obtained from Horticulture Research Institute, Agricultural Research Centre, Ministry of Agriculture, Egypt.

Seeds of *T. erecta* L. were sown on 18, 24th March 2019 & 2020 respectively in pots of 40 cm diameter, the growing medium used for seeds germination was clay + sand a mixture of (3:1 v/v). Uniform seedlings of 60 days old (20 cm in height) were transplanted individually into a clay pot of 30 cm diameter, which filled with 10 kg of clay soil, with bottom holes to drain excess irrigation water. The used medium contained 210, 17 and 746 mg/l of N, P and K, respectively, according to the chemical analysis, pH of 7.9 and (EC) was 3.1 $dS.m^{-1}$. Calcium superphosphate "16% P_2O_5" (4 gm per plant) but was added during soil preparation at final transplanting. Ammonium nitrate "33.5% NH_4" (3 gm per plant) and potassium sulfate "48% K_2O, 2 gm per plant) were added after 10 days from transplanting [20]. After a month from transplanting the plants were irrigated with saline water, to the final of pots were irrigated with 300 cm^3 capacity of the soil used every three days the growing media. Water saline irrigation was performed with four salinity rates: control (tap water = 0.310 $dS.m^{-1}$), 2, 4 and 6 $dS.m^{-1}$ of sodium chloride (NaCl). Which achieved was dissolved four salinity ratings 0.763, 1.526 and 2.289 g/liter of salt (NaCl) in tap water. Exogenously, the plants were given four different doses of SA solutions (T_0 (just tap water), T_1= 25, T_2=50 and T_3=75 (mg/l= ppm) as recommended by Eraslan *et al*. [21] and Basit, *et al*. [22]. The solutions were foliar spray on the marigold's aerial parts. SA was sprayed monthly three times throughout the growing season until the run -off point after transplanting (before the reproductive stage). Each time, a total amount of 100 cc of SA solution was sprayed on each plant. The regular agricultural activities were done as plant needed. In both seasons, the study was concluded in November 2019 and 2020, respectively.

Morphological Measurements: The data recorded for the growth measurements included; plant height (cm), number of branches $plant^{-1}$, number of leaves, dry weight of leaves $Plant^{-1}$(g), number of flowers $Plant^{-1}$, flower wet weight. $plant^{-1}$ (g) and root dry weight. $plant^{-1}$ (g).

Botanical Estimates: Leaf chlorophyll a, b (mg. g^{-1} leaf fresh weight) were measured using the method provided by Moran [23], carotene contents (mg.100g^{-1} L.F.W.) according to Wellburn [24] and total carbohydrate (percent) in the foliage was determined using the technique described by Hedge *et al.* [25]. Also, chemical elements were analyzed on a dry-weight basis such as the nitrogen (N %) content was determined by the distillation in the micro-Kjeldahl method, phosphorus (P%)content was colorimetrically determined by using the Vando-Molybdate-Yellow method and potassium content (K%) was determined by using flame spectrophotometry according to method of Chapman and Partto [26] and Sodium contents Na^+ (mg. l^{-1}) of dry matter in the leaves were determined according to Piper [27]. Proline content (mg/g) in the leaves was determined according to Bates *et al*. [28] and The membrane stability index (MSI%) was determined according to Sairam *et al*. [29].

The Layout of Experimental Treatments: The layout of the experiment was a factorial experiment in randomized complete blocks design. The study contained 16 treatments [4 rates of salinity x 4 concentration of salicylic acid (SA)] with three replicates. Each experimental unit contained 6 plants. The data were examined statistically using the methods outlined by Snedecor and Cochran [30] with L.S.D. at 5% used to compare treatment means.

RESULTS AND DISCUSSION

Effect of Saline Irrigation Water and Salicylic Acid on Biomass and Biochemical Characteristics of Marigold Plant

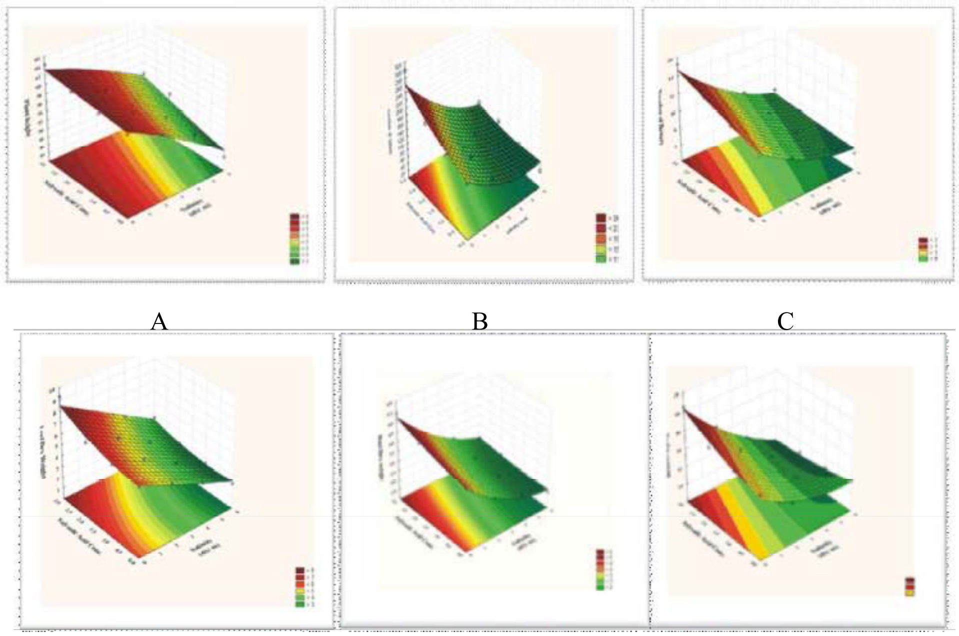
The Effect of Water Salinity, Salicylic Acid and Their Combination Treatments on Biomass Characteristics: The result in Table (1) and Figure (1) reveal that spraying *Tagetes erecta* plants with salicylic acid separately or in combine with different levels of saline water had a significant influence on the studied foliage growth parameters: plant height (cm), no. of branches per plant, leaves no. per plant and dry weight of leaves during both growth seasons (g). Table 1 shows that in the first and second seasons, salt water with EC, 2, 4 and 6 $dS.m^{-1}$ considerably reduced botanical development as compared to tap water, showing the lowest values with the high level of saline water. The decrease in vegetative growth characteristics resulting from soil salinity might be attributed to the inhibition of both meristematic activity and elongation of cells Neiman [31] and Tuna *et al*. [32] also reported that decrease in plant height with increasing

Table 1: The effect of water salinity, salicylic acid application and their interaction on plant height, number of branches/ plant, number of leaves/ plant and dry weight of leaves of *Tagetes erecta* L. during the two seasons of 2019 and 2020

	Salicylic acid (SA, mg.l^{-1}) (B)									
	Plant height (cm)									
	(1st) season					(2nd) season				
Water salinity (EC, dS.m^{-1}) (A)	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	60.85	61.30	62.09	64.36	62.15	60.58	61.21	61.88	64.38	62.01
2.00	57.43	58.39	59.41	59.99	58.80	57.17	58.19	59.09	59.70	58.54
4.00	54.00	54.05	54.23	55.07	54.34	53.94	54.20	54.47	55.58	54.55
6.00	45.20	48.13	52.26	53.30	49.72	45.17	48.08	52.33	53.02	49.65
Mean	54.37	55.47	57.00	58.18	56.25	54.21	55.42	56.94	58.17	56.19
$LSD_{0.05}$		A= 0.23 B=0.23 A×B=0.46					A=0.55 B=0.55 A×B= 1.10			
	Number of branches/ plant									
	0.00	25.0	50.0	75.0	mean	0.00	25.0	50.0	75.0	Mean
Tap water	13.077	13.790	15.800	18.190	15.214	13.083	13.667	15.667	17.917	15.083
2.00	6.477	6.800	11.333	12.543	9.288	6.467	7.000	11.611	12.233	9.328
4.00	4.810	5.000	5.233	5.550	5.148	4.583	4.767	4.933	5.067	4.838
6.00	1.767	2.327	2.757	3.823	2.668	2.000	2.206	2.383	3.067	2.414
Mean	6.533	6.979	8.781	10.027	8.080	6.533	8.649	128.19	9.571	7.916
$LSD_{0.05}$		A=0.286 B=0.286 A×B= 0.571					A=0.255 B=0.255 A×B=0.509			
	Number of leaves/ plant									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	154.84	172.44	207.79	297.62	208.17	154.50	176.58	202.58	303.18	209.21
2.00	116.78	123.57	132.28	142.21	128.71	114.25	126.08	134.92	144.08	129.83
4.00	90.66	92.98	93.99	106.66	96.07	88.08	93.07	97.00	101.75	94.98
6.00	46.84	70.88	84.20	87.21	72.28	47.63	71.32	78.25	86.08	70.82
Mean	102.28	114.97	129.56	158.42	126.31	101.12	116.76	128.19	158.77	126.21
$LSD_{0.05}$		A=4.68 B=4.68 A×B= 9.35					A=9.12 B=9.12 A×B=18.25			
	Leaves dry weight / plant (g)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	6.3067	6.5833	6.8033	9.2800	7.2433	6.3260	6.5360	6.8873	9.5367	7.3215
2.00	5.2233	5.6767	5.9100	6.0533	5.7158	5.0717	5.3600	5.7100	5.9417	5.5208
4.00	3.7600	3.9133	4.2000	4.9633	4.2092	3.7650	3.9183	4.1800	4.8533	4.1792
6.00	2.2300	2.5167	2.7133	3.6267	2.7717	2.1623	2.5433	2.7433	3.5333	2.7456
Mean	4.3800	4.6725	4.9067	5.9808	4.9850	4.3313	4.5894	4.8802	5.9662	4.9418
$LSD_{0.05}$		A=0.087 B=0.087 A×B= 0.174					A=0.062 B=0.062 A×B=0.125			

L.S.D $_{(0.05)}$ = Least significant differences at 0.05 probability level

salinity levels may be due to inadequate water uptake and hence, relative water content (RWC) was significantly decreased and resulted in limited water availability for the cell extension process. Similar results obtained by many researches as El- Kouny *et al*. [33] on roselle (*Hibiscus sabdariffa,* L.) and El-Naggar *et al*. [34] on *Matricaria chamomilla* plants. Compared to the untreated control of the two growing seasons, the salicylic acid (SA) application (25.0, 50.0 and 75.0 ppm) produced significant increases in vegetative growth characteristics of *Tagetes erecta* plants. Overall, the concentration of salicylic acid (75.0 ppm) for both seasons, led to a rest increase in plant height. Most of the time, the various concentrations of salicylic acid treatments improved vegetative growth and made a big difference in the growth of plants compared to plants that weren't treated. Salicylic acid (SA) is a non-enzymatic antioxidant messenger molecule that regulates plant physiology during stress [35]. Apart from protoplasm production, division and elongation of meristem cells, stimulating apical meristems may also enhance protein and carbohydrate biosynthesis. Yildirim *et al*. [18] on cucumber and Zarghami *et al*. [19] on Petunia plants observed similar results. Several growth characteristics, such as plant height (cm), number of branches per plant, number of leaves per plant and dry leaf weight, were found to be optimum when salicylic acid (SA) 75.0 ppm was used in conjunction with tap water (giving values of



A B C

D E F

Fig. 1: The effect of water salinity, salicylic acid application and their interaction on (A): plant height (cm), (B): number of leaves/ plant, (C): number of flowers/ plant, (D): dry weight of leaves (g), (E): root dry weight (g) and(F): proline content (mg/g) of *Tagetes erecta*, L. during both seasons

64.36 cm, 18.19, 297.62 and 9.28 g, respectively in the 1st season and 64.38cm, 17.92, 303.18 and 9.54 g, respectively, in the 2nd season for the above-mentioned parameters.). Because salicylic acid (SA) and water salinity work together to encourage plant growth and dry matter accumulation, this could be why these results were found. Reduced growth could also be caused by salt buildup in the soil due to increments application of saline water, which raises the osmotic stress experienced by plant tissue cells and reduces water absorption, resulting in decreased cambium activity and lower cell maturity [36, 19].

The Effect of Water Salinity, Salicylic Acid and Their Combined Treatments on Flowering: The obtained results revealed that the mean of number of flowers. plant^{-1} and flower wet weight. plant^{-1}(g) were markedly improved as a result of spraying *Tagetes erecta* L. with salicylic acid compared to the control (Table 2) and Figure (1). In both seasons, the number of flowers per plant and flower fresh weight per plant decreased as the EC concentration increased, as shown in Table (2) and Figure (1). In the two growing seasons, the lowest ones were recorded in plants treated with saline water 6.0 dS.m^{-1} EC, which produced 6.85 and 5.85 flowers/plant and 1.45 and 1.86g, respectively, as compared to the control. It's possible that the decline in flower quantity seen after utilizing high salinity up to EC, 8 dS.m^{-1} is connected with an increase in respiration and an inhibition of photosynthetic system in all cells, resulting from the occurrence and accumulation of detrimental ions (Na^+ and Cl^-). Likewise, similar results are documented On poinsettia Yelanich and Biernbaum [37] and reported similar results, as did Kamkari *et al*. [38] pot marigolds. In our study, the use of 75.0 ppm salicylic acid (SA) with tap water treatment gave significantly higher flowers than those of other treatments. The increase in blossom number and fresh weight as a result of employing a sufficient salicylic acid (SA) concentration combined with tap water could be attributed to the role of salicylic acid, which is required for protein and cytokinin synthesis and hence impacts cell division. and to the increase and activation the formed roots. This stimulates absorption of the essential elements for flowers initiation and development. These results are similar to those obtained by Zarghami *et al*. [19] with petunia plants.

The Effect of Water Salinity, Salicylic Acid and Their Combination Treatments on Root Dry Weight: The heaviest of dry weight of roots / plant was obtained by applying salicylic acid (SA) at 75.0 ppm, giving 3.54 and 3.52g compared with the control 2.83 and 2.88 g in the

Table 2: The effect of water salinity, salicylic acid application and their interaction on number of flowers / plant, flower fresh weight/ plant, root dry weight/ plant of *Tagetes erecta* L. during the two seasons of 2019 and 2020

	Salicylic acid (SA , mg.l^{-1}) (B)									
	Number of flowers / plant									
	(1st) season					(2nd) season				
Water salinity (EC, dS m^{-1}) (A)	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	10.83	12.27	14.05	15.33	13.12	9.83	11.27	13.05	14.33	12.12
2.00	9.00	9.39	9.89	10.03	9.58	8.00	8.39	8.89	9.03	8.58
4.00	8.11	8.44	8.55	9.00	8.53	7.11	7.44	7.55	8.00	7.53
6.00	6.00	6.39	7.17	7.83	6.85	5.00	5.39	6.17	6.83	5.85
Mean	8.49	9.12	9.92	10.55	9.52	7.49	8.12	8.92	9.55	8.52
$LSD_{0.05}$		A= 0.25 B=0.25 A×B=0.49					A=0.24 B=0.24 A×B= 0.49			
	Flower fresh weight/ plant (g)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	6.5467	7.0700	7.5900	7.9800	7.2967	7.0200	7.4867	8.0733	8.3467	7.732
2.00	4.7467	5.3033	5.6800	5.9400	5.4175	5.1267	5.6567	6.1133	6.4667	5.841
4.00	2.6433	3.6067	4.0733	4.3733	3.6742	2.9700	3.9200	4.4967	4.7833	4.043
6.00	0.7867	1.1733	1.7700	2.0667	1.449	1.1200	1.7300	2.1567	2.4467	1.863
Mean	3.6808	4.2883	4.7783	5.0900	4.4594	4.0592	4.6983	5.2100	5.5108	4.8696
$LSD_{0.05}$		A=0.286 B=0.286 A×B= 0.571					A=0.042 B=0.042 A×B=0.084			
	Root dry weight/ plant (g)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	4.1433	4.4900	4.6967	5.4233	4.6883	4.1427	4.3007	4.6690	5.1667	4.5698
2.00	2.9800	3.1467	3.3300	3.6500	3.2767	3.0693	3.1867	3.4367	3.6460	3.3347
4.00	2.3767	2.4867	2.6200	2.8633	2.5867	2.5353	2.6667	2.8133	2.8767	2.7230
6.00	1.8133	1.9333	2.1133	2.2367	2.0242	1.7900	1.8787	2.0917	2.4000	2.0401
Mean	2.8283	3.0142	3.1900	3.5433	3.1440	2.8843	3.0082	3.2527	3.5223	3.1669
$LSD_{0.05}$		A=0.026 B=0.026 A×B= 0.051					A=0.038 B=0.038 A×B=0.075			

L.S.D. (0.05) = Least significant differences at the 0.05 level of probability

both growing seasons, respectively. (Table 2 and Figure 1). Salty water irrigation of EC, 2 and 4 dSm^{-1} considerably reduced the maximum dry weights of roots/plant, when compared to (tap water). The plants that were irrigated with EC 6 dS.m^{-1} saline water had the lowest root dry weight, with 2.024 and 2.040 g in 1st and 2nd seasons, respectively. This finding may be attributed to the fact that under high salinity conditions, the dry weight of the roots was lower because the cells didn't divide as a response to the toxic effects of Na^{+} and Ca^{+} ions in the cytoplasm of root cells. This led to a decrease in root cell division and their elongation [39]. These results are in general line with what was Sutarno [40] wrote about *Amaranthus paniculatus*, stated that the interaction of highest levels of salicylic acid (SA) combined with tap water was the most effective for increasing root, dry weight in both seasons. The large increase may due to the application of salicylic acid (SA) at low salinity (tap water) enhances the vegetative development of the employed plants, resulting in more biosynthesizes being utilized and thus the root weight becoming improved. These findings are consistent with those of El-Kouny *et al*. [33] who studied *Hibiscus sabdariffa L*. plants. There are two bioactive concentration windows for SA in the root system; at low levels, it acts as a developmental regulator and at high levels, SA acts as a stress hormone [41]. Most of the evidence reviewed herein, suggests that SA's hormetic abilities act to stimulate growth at low doses and to inhibit growth at high doses [42].

The Effect of Water Salinity, Salicylic Acid and Their Combination Treatments on Chemical Facets

Leaf Nitrogen (N), Phosphorus (P) and Potassium Contents (K) (%): Table (3) shows that salicylic acid treatment had a considerable influence on leaf nitrogen, phosphorus and potassium levels. In both seasons, the concentration of macro elements (N, P and K) in leaves (percent) grew progressively as salicylic acid (SA) increased. During study it was found that 75 mg. l^{-1} salicylic acid (SA) produced the best results when compared to the other treatments. These findings could be explained by salicylic acid's role in the creation

Table 3: The effect of water salinity, salicylic acid application and their interaction on Leaf nitrogen(N), Phosphorus(P) and Potassium contents(K) (%) of *Tagetes erecta* L. during the both seasons of 2019 and 2020

	Salicylic acid (SA , mg. l^{-1}) (B)									
	Nitrogen content of leaves (N%)									
	(1^{st}) season					(2^{nd}) season				
Water salinity (EC, dS.m^{-1}) (A)	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	2.146	2.172	2.239	2.622	2.295	0.618	0.664	0.724	0.744	0.6875
200	2.011	2.030	2.062	2.098	2.050	0.425	0.478	0.525	0.571	0.5001
4.00	1.908	1.940	1.972	1.998	1.955	0.201	0.295	0.356	0.394	0.3115
6.00	1/485	1.692	1.789	1.839	1.701	0.0167	0.098	0.122	0.160	0.0991
Mean	1.888	1.959	2.016	2.139	2.000	0.315	0.384	0.432	0.467	0.3996
$LSD_{0.05}$	A= 0.109 B= 0.109 A×B= 0.217					A=0.0042 B=0.0042 A×B=0.0059				
	Phosphorus content of leaves (P%)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	0.668	0.685	0.719	0.889	0.741	0.081	0.083	0.089	0.098	0.088
2.00	0.604	0.629	0.645	0.657	0.634	0.045	0.050	0.051	0.057	0.051
4.00	0.522	0.540	0.557	0.577	0.549	0.040	0.043	0.047	0.054	0.046
6.00	0.446	0.467	0.488	0.503	0.476	0.032	0.032	0.038	0.043	0.036
Mean	0.560	0.5803	0.602	0.657	0.599	0.049	0.052	0.056	0.063	0.055
$LSD_{0.05}$	A= 0.027 B= 0.027 A×B= 0.054					A= 0.0153 B=0.0153 A×B=0.0306				
	Potassium content of leaves (K%)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	1.2627	1.2933	1.4160	2.042	1.502	1.362	0.685	0.673	0.656	0.844
2.00	1.188	1.202	1.232	1.247	1.217	1.030	0.611	0.734	0.754	0.782
4.00	1.151	1.165	1.170	1.177	1.166	0.811	0.660	0.638	0.689	0.700
6.00	1.056	1.099	1.123	1.137	1.104	0.663	0.705	0.629	0.664	0.661
Mean	1.165	1.190	1.235	1.401	1.248	0.966	0.665	0.669	0.691	7.47
$LSD_{0.05}$	A= 0.060 B= 0.060 A×B=0.120					A= 0.134 B=0.134 A×B=0.189				

L.S.D. (0.05) = Least significant differences at the 0.05 level of probability

of membrane phospholipids, sugar phosphates, nucleotides and co-enzymes. As a result, the N, P and K content of photosynthetic pigments increased. This result agrees well with Khan *et al.* [39]. Furthermore, when concentrations increased, the leaf content of nitrogen (N), phosphorus (P) and potassium (K) decreased in both seasons. The minimum was produced by irrigation water with an EC of 6 dS.m^{-1} EC compared to tap water during the two growing seasons. Bernstien *et al*. [44] on ornamental plants and Shaban [45] on Tagetes minuta found similar results. Significant interaction effect between salicylic acid (SA) and irrigation water salinity treatments of N, P and K were discovered in both seasons. Mixing tap water and a large amount of salicylic acid (SA), the highest values% was achieved (75 mg. l^{-1}). Exogenously administered SA enhanced the ATPs content in the shoot and roots of four wheat genotypes under NaCl stress, according to Zhang *et al*. [46]. Furthermore, when salicylic acid (SA) was applied to the shoots of control and stressed plants, the potassium level reduced, whereas it increased in the plant. It may also be due to the replacement of potassium ions (K^+) in the leaves tissue by sodium ions (Na^+). These findings are consistent with those of Khan *et al*. [39] on xxxx and Shaban [45] on *Tagetes minuta*. Refat *et al*. [47] shows that increasing soil salinity from 4 to 12 dB.m^{-1} reduces nutrient absorption from sunflower plant leaves, spraying salicylic acid on the leaves improves nutrient absorption because salicylic acid increases plant dry matter and prevents sodium absorption. This lowers sodium ion significant harm in plant tissues while enhancing N, P, K, Ca and Mg absorption.

Chlorophyll A, B, (mg. g^{-1} Leaf Fresh Weight) Carotene (mg.100g^{-1} Leaf Fresh Weight) and Carbohydrates Contents of Leaves (%): In comparison to the control plants presented in Table (4) salicylic acid treatment significantly enhanced the leaves chlorophyll A, B, Carotene content (mg.100g^{-1} L.F.W.) and carbohydrates content (percent) in the two seasons. There was a steady

Table 4: The effect of water salinity, salicylic acid application and their interaction on Chlorophyll A, B, Carotene content of leaves and total carbohydrates content of leaves of *Tagetes erecta,* L. during the two seasons of 2019 and 2020

	Salicylic acid (SA , mg.l^{-1}) (B)									
	Chlorophyll (A) content of leaves (mg.100g^{-1} FW)									
	(1st) season					(2nd) season				
Water salinity (EC, dS m^{-1}) (A)	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	17.107	18.139	18.901	21.224	18.843	10.307	11.339	12.101	14.424	12.043
2.00	14.475	14.673	15.246	16.020	15.104	7.675	7.873	8.446	9.220	8.304
4.00	12.039	13.397	13.847	14.146	13.357	5.239	6.597	7.047	7.346	6.557
6.00	8.093	9.657	10.618	11.195	9.891	1.293	2.857	3.818	4.395	3.091
Mean	12.928	13.967	14.653	15.646	14.299	6.128	7.167	7.853	8.846	7.499
$LSD_{0.05}$		A= 0.471 B=0.471 A×B=1.625					A= 0.331 B=0.331 A×B= 0.663			
	Chlorophyll (B) content of leaves (mg.100 g^{-1} FW)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	15.316	16.318	18.069	23.635	18.335	12.116	13.118	14.869	20.435	15.135
2.00	11.787	12.666	13.107	14.041	12.900	8.587	9.466	9.907	10.841	9.700
4.00	8.987	9.828	10.219	10.888	9.981	5.787	6.628	7.019	7.688	6.781
6.00	5.288	6.203	7.179	8.707	6.844	2.088	3.003	3.979	5.507	3.644
Mean	10.345	11.254	12.144	14.318	12.015	7.145	8.054	8.944	11.118	8.815
$LSD_{0.05}$		A=1.254 B=1.254 A×B= 2.508					A=1.347 B= 1.347 A×B=2.694			
	Carotene content of leaves (mg.100g^{-1}F.W.)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	14.384	15.045	15.954	17.456	15.800	8.155	7.587	9.813	10.829	9.096
2.00	11.899	12.284	12.664	13.484	12.583	8.241	9.9576	12.406	9.3484	9.988
4.00	9.937	10.279	10.669	11.307	10.549	9.112	10.038	9.250	9.139	9.387
6.00	6.557	7.284	8.831	9.728	8.099	10.175	8.856	7.314	8.660	8.751
Mean	10.694	11.223	12.029	12.994	11.735	8.921	9.109	9.696	9.494	9.305
$LSD_{0.05}$		A=0.31486 B= 0. 31486 A×B= 0.62973					A=1.5864 B= 1.5864 A×B=3.1724			
	Total carbohydrates content of leaves (%)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	10.056	10.347	10.697	12.194	10.824	6.456	6.747	7.097	8.594	7.224
2.00	9.466	9.612	9.739	9.906	9.681	5.866	6.012	6.139	6.306	6.081
4.00	8.776	8.843	8.997	9.203	8.955	5.176	5.243	5.397	5.603	5.355
6.00	6.167	7.559	8.126	8.528	7.595	2.567	3.960	4.526	4.928	3.995
Mean	8.616	9.091	9.390	9.958	9.264	5.016	5.491	5.790	6.358	5.664
$LSD_{0.05}$		A=0.369 B=0.369 A×B= 0.738					A=0.408 B=0.408 A×B=0.817			

$L.S.D_{(0.05)}$ = Least significant differences at 0.05 probability level

decline in chlorophyll and carbohydrate content was seen as the salinity concentration in the irrigation water increased. The lowest values of chlorophyll a, b and carbohydrates was recorded plants in treated with water saline at EC 6.0 dS.m^{-1} which equal 9.89, 6.84, 809.98 and 7.60 compared to (18.84, 18.34, 15.710 and 10.82) the control treatment in both seasons. The highest significant increases of Chlorophyll A, B, Carotene and carbohydrates in leaves were found in plants sprayed from 75.0 ppm salicylic acid with tap water. In both seasons, average Chlorophyll A, B, Carotene and carbohydrates content were observed at 21.224, 23.635, 17.456, 12.194 and 14.42, 20.44, 1082.92 and 8.594, respectively. This increase may be due to the effects of salicylic acid (SA) on stimulating the growth and enhancing leaf production which probably had higher chlorophyll biosynthesis in the leaves tissues and consequently more carbohydrate production. Some researchers believe salicylic acid is a new plant growth hormone. Salinity stress reduced the rate of photosynthesis, ribulose-bisphosphate carboxylase activity (RUBPC) and chlorophyll concentration, according to Mittova *et al*. [48]. The loss in chlorophyll caused by salt stress was reported by Yamane *et al*. [49], who determined that the injury in chloroplasts is dependent on light and that H_2O and OH are responsible for the negative effects of salt stress on chlorophyll content and chloroplast ultra-structure. In general, when plants are subjected to salt stress, the stomata are closed and photosynthetic efficiency decreases Parida *et al.* [50]. According to several studies, when plants are exposed to salt stress, their total chlorophyll and carotene content decreases and their photosynthetic efficiency decreases as the stomata are closed [51]. These findings match those of Naiem *et al*. [52] on spearmint and marjoram plants and Hussein *et al*. [53] on maize plants. Because SA-treated plants produced more cytokinin, which is engaged in increased pigment levels by the methodologies of chlorophyll production and mitigation of chlorophyll collapse, plants treated with SA alone and with SA+NaF exhibited higher levels of all investigated pigments [54]. Hadi *et al*. [55] found that SA reduced the negative effects of salinity stress (reduced the amount of chlorophyll a, b, carotenoids, proline, protein and soluble sugars) and increased the amount of plant pigments and plant growth on white bean plants under salinity stress using different forms (soil, foliar and priming. High salinity decreased the carbohydrate percentage, as indicated in this study which may be related to a decrease in chlorophyll content and inhibition of carbohydrate biological processes, disturbance of nitrogen [56].

Sodium Content (Na^+ mg. g^{-1}): Salicylic acid (SA) foliar treatments at various concentrations (0.0, 25.0, 50.0 and 75.0 mg. l^{-1}) reduced leaf content of Na ppm in both seasons, as shown in Table (5). Furthermore, there was a positive correlation between increased irrigation saline dosage and increased leaf content of Na^+ within both seasons The greatest amounts were reported in leaves of the plants irrigated with EC, 6 dSm^{-1}, which equaled 135.07 and 128.76 mg. g^{-1}, respectively, during the first and second seasons. Irrigation with EC 6 dSm^{-1} saline water without adding salicylic acid (SA) resulted in the highest leaves Na of *Tagetes erecta* L. with 152.67 and 146.34 ppm for both seasons, respectively, compared to other treatments. Meanwhile, irrigating the plants with tap water laced with salicylic acid (SA) at a concentration of 75.0 ppm had the opposite effect. It lowered the Na+ ppm of the leaves to just 92.96 and 86.67 ppm in the both seasons, respectively. Similar findings were discovered in the Eucalyptus hybrid by Singh [57]. These findings could be explained by an excess of Na ions in root cells, which can enter through multiple channels via plasma membrane ATP use and subsequently be translocated and stored in leaf tissues [58]. A similar pattern of results was discovered by Mostafa [59] and Koyro [60].

Proline Content (mg. g^{-1}): Salicylic acid (SA) had a negative effect on proline content (mg/g) in the dried leaves of the two seasons, according to the data in Table (5) and Figure (1). Furthermore, when the level of salicylic acid (SA) increased, proline content (mg/g) fell progressively and consistently. In the first and second seasons, proline content decreased from 14.17 (control) to 12.20 mg/g at 75 ppm salicylic acid (SA) and from 16.37 to 14.40 mg/g, respectively. Significant changes in proline content were also seen in plants receiving various irrigation water treatments (Table 4); raising the salt level in the water increased the proline contents (mg/g) in dried leaves in general. In the first and second seasons, plants irrigated with the large level of salt concentration (EC 6 $dS.m^{-1}$) had the highest mean proline values of 16.30 and 18.50 mg/g, respectively. Respectively. On the other hand, plants irrigated with tap water had the lowest mean proline value of 10.61 and 12.81 for both growing seasons. Respectively. The interaction between salicylic acid (SA) and salinity concentrations, the data in Table (4) show that significant changes in proline (mg/g) were found in the leaves of plants receiving various combinations of salicylic acid (SA) and water salinity. In the first and second seasons, plants irrigated with salty water at EC6 $dS.m^{-1}$ without receiving salicylic acid (SA) paired with EC, 6 $dS.m^{-1}$ had the greatest proline concentrations (mg/g) in the leaves, reaching 18.50 and 20.70 mg/g, respectively, when compared to other treatments. Because of the significant increase in proline accumulation in plants irrigated with high salt rates, it's possible that proline plays a role in plant salinity tolerance. Greenway and Munns [61] defined this role by stating that proline can be thought of as an osmotic pressure stabilizer within the cell. Maraim [62] and Marcum and Murdoch [63] both came to the conclusion that proline could play a significant role in cytoplasmic osmotic adjustment. In terms of the influence of diverse combinations of saline irrigation water and salicylic acid (SA)concentrations, EL-Sayed [64] on Murrya and Jaiswal *et al*. [65] on soybean plants and EI-Shanhorey, *et al*. [66] on Jatropha reported similar results.

Membrane Stability Index (MSI) of Leaves (%): Table (5) shows that the membrane stability index (MSI) of leaves (percent) went down with the high level of foliar application of salicylic acid in the two seasons. The control treatment had the lowest MSI values, which

Table 5: The effect of water salinity, salicylic acid application and their interaction on sodium content of leaves proline content of leaves (mg. g^{-1}) and membrane stability index of leaves (%) of *Tagetes erecta L.* during the two seasons of 2019 and 2020

	Salicylic acid (SA, mg. l^{-1}) (B)									
	Sodium content of leaves (Na mg. l^{-1})									
	(1st) season					(2nd) season				
Water salinity (EC, dS.m^{-1}) (A)	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	106.223	104.933	98.817	92.960	100.733	100.130	98.349	92.518	86.670	94.417
2.00	113.450	111.800	109.498	107.309	110.514	107.340	105.504	102.909	101.277	104.258
4.00	122.107	120.923	118.490	115.827	119.337	116.860	114.624	111.902	109.798	113.296
6.00	152.667	136.517	126.66	124.421	135.068	146.337	130.218	120.078	118.390	128.756
Mean	120.296	117.014	114.897	113.445	116.413	117.604	112.171	106.851	104.107	110.182
$LSD_{0.05}$		A= 2.059 B=2.059 A×B=4.119					A=1.764 B=1.764 A×B= 3.528			
	Proline content of leaves (mg.g^{-1})									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	11.191	10.870	10.662	9.716	10.610	13.391	13.07	12.862	11.916	12.809
2.00	12.069	11.807	11.517	11.416	11.702	14.269	14.007	13.717	13.616	13.902
4.00	14.933	14.212	12.854	12.57	13.641	17.133	16.412	15.054	14.766	15.841
6.00	18.504	16.038	15.548	15.11	16.300	20.704	18.238	17.748	17.311	18.500
Mean	14.174	13.232	12.645	12.202	13.063	16.374	15.431	14.845	14.402	15.263
$LSD_{0.05}$		A=0.695 B=0.695 A×B= 1.389					A=0.723 B=0.723 A×B=1.446			
	Membrane stability index (MSI) of leaves (%)									
	0.00	25.0	50.0	75.0	Mean	0.00	25.0	50.0	75.0	Mean
Tap water	52.012	52.414	53.17	53.2377	52.7084	53.601	53.655	53.826	54.157	53.809
2.00	49.534	50.1483	50.473	51.049	50.3011	52.000	53.355	52.646	53.343	52.586
4.00	48.137	48.661	49.315	49.4137	48.8817	50.612	50.774	51.075	51.635	51.024
6.00	45.593	46.142	47.176	47.7203	46.6578	47.007	48.613	49.213	49.637	48.618
Mean	48.819	49.3413	50.0335	50.3552	49.6373	50.805	51.349	51.690	52.193	51.5093
$LSD_{0.05}$		A=0.0002 B=0.0002 A×B=0.0004					A=0.097 B=0.097 A×B=0.195			

$L.S.D_{(0.05)}$ = Least significant differences at 0.05 probability level

were 48.819% and 50.805% in both years. Treatment of EC 6 dS.m^{-1} was the most important, where the highest salinity levels gave the highest value of the membrane stability index. Table (4) shows that the MSI of leaves rose as the concentration of salt in irrigation water rose in both seasons. As for the interaction, the data show that leaves' membrane stability index (MSI) decrease significantly when salicylic acid concentration went up. This is because water saltiness at all degrees (EC, 2 to 6 dS.m^{-1},) reduced the MSI at the presence of salicylic acid (25.0 ~ 75.0 mg. l^{-1}, range). Both seasons, plants irrigated with tap water and sprayed with salicylic acid (SA) at 75.0 mg. l^{-1} had the best results. NaCl stress can reduce nutrient uptake by the root of the plant, resulting in decreasing plant growth (dry aerial biomass), chlorophyll a, b, total carbohydrates and proline. Application of salicylic acid under salinity stress increases water holding capacity and inhibits the plants from excessive uptake of Na^+. Therefore, it prevents Na+ toxicity in plants. This study showed that the lowest salinity level (2.0 &4.0 dS.m^{-1}EC) and application of salicylic acid, especially at 75.0 ppm, had the highest effect on ameliorating biomass and biochemical characteristics of the plant [67]. In previous studies, SA has been shown to positively affect the membrane stability index. This is because salinity lowers the osmotic pressure, which means there is less water available. SA did a good job of raising the osmotic pressure when there was a lot of salt in the water. This is important for restoring cell swelling. Chinnusamy and Zhu [68] said that plants need positive turgor in order to grow and open their stomata. These membranes are very sensitive to changes in their environment and stress has been shown to cause damage to them in plants [69].

CONCLUSIONS

The study results on the growth characteristics, flower quality and chemical contents of Marigold plants indicated that the optimal salicylic acid (SA) spraying concentrations were 75 ppm and irrigation with tap water (control). Furthermore, the data suggest that foliar spraying with sprayed salicylic acid (SA) from 25 to 75 mg. l^{-1} concentration is effective. Finally, the findings suggest that SA could be employed to improve plant growth and quality in saline environment in the future. Saline water concentrations of 2 and 4 ds.m^{-1} were suitable for marketing the plant, allowing it to achieve its goal.

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