

Effect of Salinity (NaCl + CaCl₂) and some Soil Additions on Growth, flowering and chemical composition of rose of China (*Hibiscus rosa-sinensis* L.) plant

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ABSTRACT

A pot experiment was carried out under the full sun at the Experimental Farm of Hort Res. Inst., Giza, Egypt during 2014 and 2015 seasons to explore the role of both humic acid (HA) at the rates of 0, 5 and 10 cm³/l and effective microorganisms (EM) biostimulant at the rates of 0, 3 and 5 cm³/l when applied individually every month as soil drench on growth, flowering and chemical composition of Chinese hibiscus (*Hibiscus rosa-sinensis* L.) plants cultivated in 25-cm-diameter plastic pots filled with about 6 kg of clay and sand soil mixture (1:1, v/v) salinized with an equal mixture of NaCl and CaCl₂ (1:1, w/w) at 0, 2000, 4000 and 6000ppm concentrations. The results show that the means of all vegetative and root growth traits were descendingly decreased with increasing salinity concentrations to reach the minimal values by the highest level (6000 ppm) with significant differences compared to control means in the two seasons, but they were gradually increased as a result of increasing the application rate of both HA and EM biostimulant regardless of salinity level. In general, humic acid treatments recorded better vegetative and root growth than EM ones in most cases of both seasons. However, the best vegetative and root growth in the two seasons was attained by plants grown in soil mixture free from salinity and treated with the high rate of either HA (10 cm³/l) or EM (5 cm³/l), as these two interaction treatments gave the utmost high averages at all over control and all other interactions in both seasons. Similarly, were those results of salt resistance index percentage (SRI %), flower diameter and flower fresh and dry weights. The content of chlorophyll a, b, carotenoids, N, P and K % in the leaves were gradually decreased with raising salinity concentration, but was progressively increased as the rate of either HA or EM was elevated. The opposite was the right regarding the contents of Na, Ca, Cl and proline amino acid, which were raised as the rate of salinity, HA and EM was increased, except for of Cl content that was gradually diminished with rising the application rate of both HA and EM. This pronouncedly showed the role of both HA and EM in restricting the absorption of Cl element that may cause toxicity for plants when exist at high concentration. Hence, it is recommended to treat the rose of China (*Hibiscus rosa-sinensis* L.) plants grown in saline soil, monthly with either HA (10 cm³/l) or EM biostimulant (5 cm³/l) to improve their growth and quality under such condition. Key words: Rose of China (*Hibiscus rosa-sinensis* L.), salinity, humic acid, effective microorganisms, vegetative and root growth.

Key words: *Hibiscus rosa-sinensis*, NaCl , CaCl₂, flowering and chemical composition

Introduction

Rose of China or Chinese hibiscus (*Hibiscus rosa-sinensis* L.) is a large beautiful evergreen shrub that belongs to Fam. Malvaceae nearly glabrous, grown up to 5-7 m height, leaves usually simple, ovate to 8-10 cm long, not lobed, but toothed or nearly entire, mostly grown in subtropical and tropical regions as ornamental for its profuse large very showy flowers which are born solitary on the leaf axils and also in glass- houses for summer bloom. It is propagated by cuttings, grafting or layering (Bailey, 1976).

Salinity is still the major environmental factor limiting plant growth and productivity (Allakhverdiev, 2000), especially those plants are considered sensitive to salinity of which rose of China plant that tolerates salinity up to 650 ppm (Kratsch *et al.*, 2008). However, amending the soil with nutritive-or bio-preparations may help plants to grow well under saline conditions. It this concern, Doak *et al.*, (2005) found that humic acid at either 3.5 or 7.0 g/100 m² of *Agrostis palustris*

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turf reduced the negative effects of NaCl at 6000 ppm, whereas Na and proline content in the herb was increased. Likewise, Abdel-Fattah *et al.*, (2008) reported that Tifway Bermudagrass can tolerate salinity of irrigation water up to 16000 ppm if treated with humic acid at 20 ml/l as a soil drench which greatly improved growth, density, colour and chemical constituents under salinity stress. Parallel observations were also detected by El-Khateeb *et al.*, (2011) on *Acacia saligna*, Abdel-Fattah *et al.*, (2012) on *Ficus benjamina*, Kasmani *et al.*, (2013) on *Pistachio*, Abdel-Moniem *et al.*, (2014) on *Tecoma stans* and Gad *et al.*, (2015) on okra (*Hibiscus esculantus*).

In addition, EM as a commercial biostimulant containing more than 60 selected strains of "effective microorganisms", viz., photosynthetic bacteria, lactic acid bacteria, yeast, actinomycetes and various fungi that improve growth and health of plants (Primavesi, 1999), may play a vital role in reducing the harmful effect of salinity on plant growth and production. This was documented by Tawila *et al.*, (2015) who revealed that application of 3 ml/l EM to sanitary water caused additional improvement in various vegetative and root growth parameters of Neem (*Azadirachta indica*), even with 100 % sanitary water treatment which recorded growth rates better than control one in the presence of such biostimulant (EM). The 3 ml of EM treatment significantly improved chlorophyll a, b and carotenoids content in the leaves, as well as total soluble sugars, N, P and K in the stem, leaves and roots, but markedly decreased lead (Pb) and cadmium (Cd) content in the different parts of the plant, indicating the ability of such biostimulant in bioremediation of sanitary water.

The other beneficial effects of EM bio-preparation on plants were postulated by Khan *et al.*, (2006) on *Albizia procera*, Khan *et al.*, (2011) on *Dalbergia sissoo*, Mohammed *et al.*, (2013) on *Coffea arabica*, Khan *et al.*, (2014) on *Acacia auriculiformis* and Abdel-Galeil *et al.*, (2015) on cv. Siwi cv. date palm.

However, this work was done in doer to investigate the effect of either humic acid or EM application at various levels on growth and chemical composition of Chinese hibiscus transplants planted in different levels of saline soil.

Materials and Methods

A pot experiment was consummated in the full sun at the Experimental Farm of Hort. Res. Inst., Giza, Egypt during the 2014 and 2015 seasons to find out the role of humic acid and EM individual application on reducing the harmful effect of soil salinity on growth, flowering and chemical constituents of Chinese hibiscus plant.

Therefore, the young uniform transplants of *Hibiscus rosa-sinensis* L. at a length of 15 ± 1 cm were planted on March, 1st for the two seasons in 25-cm-diameter plastic pots (one transplant/pot) filled with about 6 kg of an equal mixture of clay and sand. The chemical and physical analysis of the used clay and sand are shown in Table (a).

Table a: The chemical and physical properties of the used clay and sand in the two seasons.

Soil texture	Particle size distribution (%)				S.P.	E.C. (ds/m)	pH	Cations (meq/l)				Anions (meq/l)		
	Coarse Sand	Fine sand	Silt	Clay				Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺	HCO ₃ ⁻	Cl ⁻	SO ₄ ⁻
Clayly	7.54	22.28	30.55	39.63	55.31	2.33	8.16	7.82	2.12	15.4	0.75	6.6	8.2	11.29
Sandy	81.53	9.55	0.42	8.5	22.88	3.5	7.69	10.5	1.56	30.6	0.57	4.5	23.1	15.63

A pure salt of NaCl was mixed well with a pure one of CaCl₂ at the ratio of 1:1, by weight, and then added thoroughly by to the previously mentioned soil mixture before filling the pots at the concentrations of 0, 2000, 4000 and 6000 ppm. After one month from planting, the soil mixture was monthly drenched with either humic acid solution at the rates 0, 5 and 10 cm³/l or effective microorganisms (EM) solution at the rates of 0, 3 and 5 cm³/l, starting from the first of April till the first of September for each season. A total of 216 transplants were used in a 4 x 6 factorial experiment for each season based on a completely randomized design with 3 replicates and 3 plants for each replicate (Mead *et al.*, 1993).

At the end of each season (on October, 1st), data were recorded as follows: plant length (cm), stem diameter at the base (cm), number of branches/plant, number of leaves/plant, leaf area (cm²),

root length (cm) and the salt resistance index (%) was calculated from the following equation indicated by Wu and Huff (1983):

$$\text{SRI (\%)} = \text{Mean root length of salt treated plant} / \text{mean root length of control one} \times 100.$$

Besides, the first flower diameter, as well as fresh and dry weights (g) of leaves, roots and first flower were also measured. In fresh leaf samples, photosynthetic pigments (chlorophyll a, b and carotenoids, mg/g f.w.) were determined according to the method of Yadava (1986), while in dry ones the percentages of nitrogen (Black, 1956), phosphorus (Luatanab and Olsen, 1965), potassium, sodium and calcium (Jackson, 1973) were assessed. Moreover, the content of chloride (mg/100 g. f.w.) and proline (mg/g. f.w.) was evaluated using the methods described by Jackson, (1973) and Bates *et al.*, (1973), respectively.

Data were then tabulated and statistically analysed using SAS Institute program (2009) and followed by Duncan's New Multiple Range Test (Steel and Torrie, 1980) to compare the differences among treatment means.

Results and Discussion

Effect of salinity, humic acid, EM and their interactions on:

Vegetative and root growth parameters:

Data presented in Tables (1, 2, 3, 4, 5, 6, 7 and 8) show that the means of plant length (cm), stem diameter (cm), No. branches and leaves/plant, leaf area (cm²), root length (cm), as well as fresh and dry weights of leaves and roots (g) were progressively decreased with increasing salinity concentration to reach the minimal values by the highest level (6000 ppm) with significant differences compared to control means in the two seasons. These results may be due to the deleterious effects of salinity in reducing the cell volume at a constant cell number (Handreck and Black, 2002). Likewise, Devitt *et al.*, (2005) stated that mechanism of salt may result in cell division inhibitory and hence, reduce the rate of plant development. Furthermore, Jou *et al.*, (2006) demonstrated that ATPase participates in the endoplasmic reticulum- Golgi mediated protein sorting machinery for both housekeeping function and compartmentalization of excess Na⁺ under high salinity. Recently, Sabbagh *et al.*, (2014) stated that salinity has a two-phase effect on plant growth, an osmotic effect due to salts in the outside solution and ion toxicity in a second phase due to salt build-up in transpiring leaves.

Table 1: Effect of salinity, humic acid, EM and their interactions on plant height (cm). of *Hibiscus rosa-sinensis* L. during 2014 and 2015 seasons

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	62.33b	64.51ab	66.75a	64.53a	61.76b	63.50a	64.78a	63.35a
2000	60.48bc	62.90b	65.32a	62.90ab	53.48d	55.10cd	56.69c	55.09b
4000	55.00cd	57.21c	59.43bc	57.21b	46.33ef	48.26e	49.15e	47.91c
6000	46.31e	48.16de	50.04d	48.17c	41.59g	42.85fg	44.10f	42.86d
Mean	56.03c	58.20b	60.39a		50.79b	52.43ab	53.68a	
Second season: 2015								
0	61.83b	62.97ab	65.23a	63.34a	62.10b	63.88a	65.76a	63.91a
2000	58.19c	60.00bc	62.71ab	60.03b	51.48de	52.81d	54.50c	52.93b
4000	51.85e	53.93d	56.67cd	54.15c	44.49f	46.33e	46.00e	45.61c
6000	43.50g	45.27fg	47.33f	45.37d	39.95h	41.00gh	42.21g	41.05d
Mean	53.84b	55.54b	57.99a		49.51b	51.01ab	52.12a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

On the other hand, the means of all previous characters were gradually increased as a result of increasing the rate of either humic acid or EM irrespective of salinity concentration. This may indicate the role of humic acid in increasing the availability of nutrients in the soil through influencing on soil

microbial activity (as it can provide microbes with energy), improving nutrients retention in the soil and enhancing the water holding capacity (Doak *et al.*, 2005). As for EM, it contains more than 60 selected strains of effective microorganisms that improve growth and health of plants (Primavesi, 1999). Further, Janas (2009) mentioned that EM is characterized by a wide spectrum of activity and complex effect on plant living environment. Thus, it may be used as foliar treatments or as soil drenches. Its effects include plant disease resistant, yield creating and protective which were observed in many industrial, medicinal and ornamental plants. It also creates humus and regulates basic relations in the soil. So, it is used in many countries, on a large scale in organic production of agricultural crops.

Table 2: Effect of salinity, humic acid, EM and their interactions on stem diameter (cm) of *Hibiscus rosa-sinensis* L. plants during 2014 and 2015 seasons

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	0.90c	0.96bc	1.20a	1.02a	0.90c	0.98b	1.13a	1.00a
2000	0.87cd	0.98bc	1.13ab	0.99ab	0.86cd	0.91bc	1.03ab	0.93b
4000	0.80d	1.00b	1.10ab	0.97b	0.80d	0.90c	1.00b	0.90b
6000	0.67e	0.93d	1.00b	0.83c	0.71d	0.80d	0.91bc	0.81c
Mean	0.81c	0.94b	1.11a		0.82c	0.90b	1.02a	
Second season: 2015								
0	0.87bc	0.93b	1.12a	0.97a	0.84bc	0.95b	1.09a	0.96a
2000	0.82c	0.94b	1.06ab	0.94ab	0.80c	0.86bc	0.97ab	0.88b
4000	0.77cd	0.85bc	0.97b	0.86b	0.74cd	0.82bc	0.95b	0.84b
6000	0.63d	0.76bc	0.83c	0.74c	0.66d	0.75cd	0.83bc	0.75c
Mean	0.77cd	0.87b	1.00a		0.76c	0.85b	0.96a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 3: Effect of salinity, humic acid, EM and their interactions on number of branches per *Hibiscus rosa-sinensis* L. plant during 2014 and 2015 seasons

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	3.33cd	5.33b	6.67a	5.11a	3.67c	4.33b	5.78a	4.59a
2000	3.33cd	4.26c	6.33ab	4.64ab	3.56c	4.00b	5.51a	4.36ab
4000	2.78de	3.67cd	5.00bc	3.82b	3.33c	4.00b	4.18b	3.84b
6000	2.00e	3.00d	3.78cd	2.93c	3.10c	3.00c	3.33c	3.14c
Mean	2.86c	4.07b	5.45a		3.42b	3.83b	4.70a	
Second season: 2015								
0	3.37c	4.76b	6.27a	4.80a	3.50b	4.17b	5.42a	4.36ab
2000	3.17d	3.99bc	5.63ab	4.26b	3.25bc	3.80b	5.13a	4.06ab
4000	2.51de	3.32c	4.56b	3.46c	2.98c	3.33b	3.90b	3.40b
6000	1.90e	2.41de	2.78de	2.36d	1.86d	2.37cd	2.51c	2.15c
Mean	2.74c	3.62b	4.81a		2.90c	3.42b	4.24a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

In general, humic acid treatments recorded better results than EM ones in most cases of the two seasons regardless of salinity level, indicating its ability to reduce the harmful effects of salinity on growth more than EM, to some extent.

In relation to the effect of interaction treatments, data presented in Tables (from 1 to 8) exhibit that the best vegetative and root growth were attained by plants grown in a soil mixture free from salinity and treated with the high rate of either humic acid (10 cm³/l) or EM (5 cm³/l), as these two interaction treatments gave the highest recodes at all over control and other interactions in both

seasons. However, the superiority in most cases of both seasons was the combining between planting in fresh soil (free from salinity) and drenching with humic acid at 10 cm³/l rate.

Improving growth of Chinese hibiscus plants under salinity stress in the presence of either humic acid or EM may indicate their role in decreasing the harmful effects of salinity, especially if they were applied at higher rate. These gains go in line with those postulated by Doak *et al.*, (2005) on *Agrostis palustris*, Abdel-Fattah *et al.*, (2008) on Tifway bermudagrass and El-Khateeb *et al.*, (2011) who noticed that humic acid (powder) at the rate of 4 g/pot gave the highest fresh and dry weights of *Acacia saligna* leaves under the different irrigation intervals. The irrigation treatment every 3 or 5 days combined with humic acid gave the highest value of plant height, and fresh and dry weights of leaves. Similarly, Kasmani *et al.*, (2013) elicited that root dry weight and relative water content in pistachio plants were increased by humic acid treatments (500-1500 ppm) in combination with severe drought stress. On *Hibiscus esculantus*, Gad *et al.*, (2015) claimed that humic acid (5-15 %) as soil drench decreased soil pH and increased the availability of cobalt an micronutrients.

Table 4: Effect of salinity, humic acid, EM and their interactions on number of leaves per *Hibiscus rosa-sinesis* L. plant during 2014 and 2015 seasons

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	70.36c	78.37b	96.51a	81.75a	78.00bc	81.51b	90.33a	83.28a
2000	58.89d	70.56c	78.37b	69.27b	61.18d	69.09c	76.95bc	69.67b
4000	45.33e	63.40d	65.38cd	58.04c	53.76de	59.00d	68.67cd	60.48c
6000	43.67e	56.63d	57.73d	52.68d	41.96e	46.33e	59.59d	49.29d
Mean	54.56c	67.24b	74.50a		58.73b	63.98b	73.89a	
Second season: 2015								
0	67.85bc	73.39b	87.36a	76.20a	73.28b	76.85ab	85.50a	78.54a
2000	55.81cd	66.95bc	74.10b	65.62b	56.45c	64.56bc	73.16b	64.72b
4000	43.00de	57.86cd	61.71c	54.19c	46.83cd	56.10c	64.60bc	55.84c
6000	38.79e	49.20d	54.82cd	47.60d	38.00d	42.73d	55.11c	45.28d
Mean	51.36c	61.85b	69.50a		53.64c	60.06b	69.59a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 5: Effect of salinity, humic acid, EM and their interactions on leaf area (cm²) of *Hibiscus rosa-sinesis* L. plants during 2014 and 2015 seasons

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	47.50ab	48.76a	49.00a	48.42a	47.50a	47.50a	48.33a	47.78a
2000	41.67d	45.10bc	46.31b	44.36b	38.71d	40.73c	42.58b	40.67b
4000	39.73e	41.50d	43.33c	41.52b	32.75e	37.00de	38.67d	36.14c
6000	29.00h	35.77g	36.91f	33.89c	25.76g	26.48g	29.50f	27.25d
Mean	39.48b	42.78ab	43.89a		36.18g	39.93ab	39.77a	
Second season: 2015								
0	48.82ab	51.63a	51.94a	51.13a	48.93a	50.33a	50.89a	50.05a
2000	43.15c	46.70b	47.76b	45.87b	39.50c	42.40bc	44.50b	42.13b
4000	38.34d	42.50cd	43.68c	41.57c	33.92de	36.16d	37.36cd	35.81c
6000	30.72e	37.11d	37.25d	35.03d	24.69f	27.00e	28.56e	26.75d
Mean	40.51b	44.49ab	45.16a		36.76b	38.97ab	39.59a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 6: Effect of salinity, humic acid, EM and their interactions on root length (cm) on *Hibiscus rosa-sinesis* L. plants during 2014 and 2015 seasons

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	48.00c	53.36ab	56.91a	52.76a	46.96bc	51.60ab	54.45a	51.00a
2000	43.70d	50.90bc	52.63b	49.08ab	42.50c	50.33b	51.00ab	47.94ab
4000	33.50gh	38.77f	41.00e	37.76b	31.10e	38.61d	40.76cd	36.82b
6000	23.30i	30.42h	34.66g	29.46c	21.36g	26.30f	28.63ef	25.43c
Mean	37.13b	43.36ab	46.30a		35.48b	41.71ab	43.71a	
Second season: 2015								
0	46.67bc	51.73ab	54.10a	50.83a	45.75bc	51.33ab	53.87a	50.32a
2000	40.35c	47.36b	48.43b	45.38ab	38.95c	45.50bc	48.60b	44.35ab
4000	30.10e	35.15d	37.50cd	34.25b	28.60e	35.17d	37.09cd	33.62b
6000	20.63g	25.00f	30.46e	25.36c	19.33g	23.96f	26.61ef	23.30c
Mean	34.44b	39.81ab	42.62a		33.16b	38.99ab	41.54a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 7: Effect of salinity, humic acid, EM and their interactions on fresh and dry weights of *Hibiscus rosa-sinesis* L. leaves during 2014 and 2015 seasons.

Salinity level (ppm)	Fresh weight (g)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	37.21c	42.00b	50.73a	43.31a	39.46bc	41.80b	45.17a	42.14a
2000	31.28cd	37.35c	41.46b	36.70b	30.50e	34.50d	37.49c	34.16b
4000	27.48d	33.50cd	34.00cd	31.66bc	26.13f	28.33ef	31.88de	28.78c
6000	23.07e	27.91d	29.96d	26.98c	21.22h	23.10g	25.21fg	23.18d
Mean	29.76c	35.19b	39.04a		29.33c	31.93b	34.94a	
Second season: 2015								
0	35.31bc	39.76b	45.96a	40.34a	35.46b	35.46b	42.85a	39.34a
2000	28.45cd	33.21c	37.85b	33.17b	27.91d	27.91d	34.21bc	31.18b
4000	24.68e	29.78cd	31.35c	28.60bc	23.70e	23.70e	29.00cd	26.40c
6000	20.90f	23.71e	26.51d	23.71c	18.93f	18.93f	22.75e	20.95d
Mean	27.34c	31.62b	35.42a		26.50c'	29.70b	32.20a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 7: Cont.

Salinity level (ppm)	Dry weight (g)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	8.34c	9.45b	11.26a	9.68a	7.62b	8.10ab	8.71a	8.14a
2000	7.10cd	8.50c	9.21b	8.27b	5.67cd	6.67c	7.10bc	6.48b
4000	6.02d	7.43cd	7.36cd	6.94bc	4.88de	5.38d	5.93cd	5.40c
6000	5.11e	6.03d	6.75d	5.96c	4.09e	4.40de	4.64de	4.38d
Mean	6.64b	7.85ab	8.65a		5.57b	6.14ab	6.60a	
Second season: 2015								
0	7.92bc	9.15ab	10.58a	9.22a	7.10b	7.95ab	8.65a	7.90a
2000	6.46cd	7.61c	8.50b	7.52b	5.58c	6.28bc	6.79b	6.22b
4000	5.56de	6.83cd	7.14c	6.51bc	4.71d	5.25cd	5.80c	5.25c
6000	4.81e	5.31e	6.00d	5.37c	3.76e	4.22de	4.41d	4.13d
Mean	6.19b	7.23ab	8.06a		5.29b	5.93ab	6.41a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 8: Effect of salinity, humic acid, EM and their interactions on fresh and dry weights of *Hibiscus rosa-sinesis* L. roots during 2014 and 2015 seasons.

Salinity level (ppm)	Fresh weight (g)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	31.68c	35.18ab	37.62a	34.83a	26.92b	29.33a	30.78a	29.01a
2000	28.23cd	33.60b	34.00b	31.94ab	23.50bc	26.59b	28.76ab	26.28ab
4000	23.46e	25.42de	27.10d	25.33b	16.33d	21.60cd	22.80c	20.24b
6000	15.79g	20.07f	21.64ef	19.17c	12.00e	14.81de	15.96d	14.26c
Mean	24.79c	28.57ab	30.09a		19.69b	23.08ab	24.58a	
Second season: 2015								
0	30.01bc	33.50ab	35.78a	33.13a	25.61b	28.10a	29.37a	27.69a
2000	26.51c	30.35bc	32.31b	29.72b	20.89c	24.71b	26.28ab	23.96b
4000	20.85de	23.00d	24.29cd	22.71c	16.00d	19.33cd	20.91c	18.75c
6000	14.26f	18.17e	19.72de	17.38d	11.41e	13.30de	14.89d	13.20d
Mean	22.93b	26.26ab	28.03a		18.48b	21.36ab	22.86a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 8: Cont.

Salinity level (ppm)	Dry weight (g)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	12.36b	13.65ab	14.46a	13.49a	8.62b	9.38a	9.86a	9.29a
2000	11.03c	13.10b	13.23ab	12.45ab	7.38c	8.51b	9.21a	8.37ab
4000	9.04e	9.75d	10.50cd	9.76b	5.23d	6.91cd	7.30c	6.48b
6000	6.17g	7.80f	8.19ef	7.39c	3.86e	4.74de	5.12d	4.41c
Mean	9.65b	11.08ab	11.60a		6.27b	7.39ab	7.87a	
Second season: 2015								
0	11.10b	12.21ab	13.25a	12.19a	8.17b	8.96a	9.41a	8.85a
2000	9.81c	11.16b	11.56b	10.84ab	6.65d	7.87c	8.32ab	7.61b
4000	7.71de	8.50d	8.83cd	8.35b	5.12e	6.15de	6.53d	5.93c
6000	5.25f	6.67ef	7.30e	6.41c	3.65f	4.20ef	4.75ef	4.20d
Mean	8.47b	9.64ab	10.24a		5.90b	6.80ab	7.25a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Salt resistance index:

From data averaged in Table (9), it can be concluded that the percent of salt resistance index (SRI %), as a real indicator for salinity tolerance was descendingly decreased as the level of salinity was raised, but was ascendingly increased with elevating the application rate of either humic acid or EM with significant differences relative to control in the two seasons. This of course, mainly attributed to that salinity treatments caused a gradual decrement in the root length, whereas humic acid and EM ones caused a progressive increment in such parameter, as shown before in Table (6).

However, the mean of this character was more than 100 % in the two seasons for plants grown in fresh soil (non saline) or saline one (2000 ppm) and fertilized with either humic acid or EM at any rate. The opposite was the right concerning the higher salinity level, where 4000ppm level declined the mean of SRI % to less than 70 % and 6000 ppm one decreased it to less than 50 % in both seasons, while the sole application of either humic acid or EM elevated these declined means to more than 75-80 % for 4000 ppm saline treatment and to more than 50-60 % for 6000 ppm one in the two seasons. This may indicate the role of both humic acid and EM on improving the tolerance ability of the sensitive Chinese hibiscus plants to salinity more than 2000 ppm.

On the same line was the finding of Abdel-Fattah *et al.*, (2012) whom noted that *Ficus benjamina* plants can tolerate salinity of irrigation water up to 6000 ppm with food performance if they were fertilized with 100 cm³ of phosphorene/plant.

Table 9: Effect of salinity, humic acid, EM and their interactions on salt resistance index (%) of *Hibiscus rosasinesis* L. plants during 2014 and 2015 seasons.

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	100.00b	111.17a	118.56a	108.92a	100.00b	112.20a	117.75a	109.98a
2000	91.04bc	106.04ab	109.65ab	97.24ab	85.14c	99.45b	106.23ab	96.94b
4000	69.79cd	80.77c	85.42bc	73.39b	62.52d	76.88cd	81.07c	73.49c
6000	48.54e	63.38d	72.21cd	54.35c	42.25f	52.37e	58.17de	50.93d
Mean	77.34c	90.34b	96.46a		72.48b	85.23ab	90.81a	
Second season: 2015								
0	100.00b	110.84a	115.92a	108.92a	100.00b	112.20a	117.75a	109.98a
2000	86.46bc	101.48ab	103.77ab	97.24ab	85.14c	99.45b	106.23ab	96.94b
4000	64.50d	75.32c	80.35c	73.39b	62.52d	76.88cd	81.07c	73.49c
6000	44.20e	53.57de	65.27d	54.35c	42.25f	52.37e	58.17de	50.93d
Mean	73.79c	85.30b	91.33a		72.48b	85.23ab	90.81a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Flowering traits:

A similar trend to those obtained in case of vegetative and root growth parameters and the percent of salt resistance index was also attained regarding flower diameter (cm), fresh and dry weights (g) as shown in Tables (10 and 11), where the means of these criteria were gradually decreased as a result of increasing salinity concentration, but were progressively increased with rising the rate of both humic acid and EM. So, the widest flowers and heaviest flower fresh and dry weights were achieved in the two seasons by planting in soil mixture free from salinity and amended with either humic acid or EM biostimulant at the highest rate of each.

These results could be discussed and interpreted as aforementioned in case of vegetative and root growth parameters. A parallel observation was also detected by Gad *et al.*, (2015) who pointed out that humic acid at 10 % as soil drench gave the highest figures of *Hibiscus esculantus* plant followed by 15 % rate, while the level of 5 % gave the lowest figures.

Table 10: Effect of salinity, humic acid, EM and their interactions on flower diameter (cm) of *Hibiscus rosasinesis* L. plants during 2014 and 2015 seasons.

Salinity level (ppm)	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	9.76c	10.89b	12.47a	11.04a	9.63a	10.71b	12.33a	10.89a
2000	9.13cd	10.18bc	11.00b	10.10b	9.01cd	9.97bc	10.89b	9.96b
4000	8.11d	8.67cd	9.17cd	8.65c	7.99d	8.36cd	9.09cd	8.48c
6000	6.73e	7.10de	7.68d	7.17d	6.31e	6.73de	7.52d	6.85d
Mean	8.43b	9.21ab	10.08a		8.24c	8.64b	9.96a	
Second season: 2015								
0	9.20c	10.25b	11.72a	10.39a	9.10bc	10.12b	11.89a	10.37a
2000	8.63cd	9.62bc	10.36b	9.54ab	8.50c	9.56bc	10.91ab	9.66ab
4000	7.62d	8.46cd	8.60cd	8.23b	7.52cd	8.31c	8.67bc	8.17b
6000	6.33e	6.68de	7.50d	6.84c	5.93d	6.92d	7.16d	6.67c
Mean	7.95b	8.75ab	9.55a		7.76c	8.73b	9.66a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 11: Effect of salinity, humic acid, EM and their interactions on fresh and dry weights of *Hibiscus rosa-sinensis* L. flowers during 2014 and 2015 seasons.

Salinity level (ppm)	Fresh weight (g)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	2.52d	3.05b	3.76a	3.11a	2.52bc	2.97b	3.73a	3.07a
2000	2.40e	2.66c	2.84bc	2.63b	2.36c	2.53bc	2.67bc	2.52b
4000	2.31e	2.52d	2.57cd	2.47bc	2.19cd	2.31c	2.57bc	2.36bc
6000	2.04f	2.30e	2.38e	2.24c	2.04d	2.20cd	2.36c	2.20c
Mean	2.32b	2.63ab	2.89a		2.28b	2.50ab	2.83a	
Second season: 2015								
0	2.37bc	2.87b	3.50a	2.91a	2.40bc	2.76b	3.46a	2.87a
2000	2.21c	2.40bc	2.49bc	2.37b	2.18c	2.35bc	2.41bc	2.31b
4000	2.11cd	2.23c	2.31c	2.22b	2.06c	2.10c	2.27bc	2.14b
6000	1.86d	2.15cd	2.17cd	2.06c	1.80c	1.96c	2.15c	1.97c
Mean	2.14b	2.41ab	2.62a		2.11b	2.29b	2.57a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Table 11: Cont.

Salinity level (ppm)	Dry weight (g)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
First season: 2014								
0	0.31bc	0.36b	0.46a	0.38a	0.31a	0.34ab	0.42a	0.36a
2000	0.28c	0.32bc	0.32bc	0.31b	0.28b	0.29b	0.31b	0.29b
4000	0.27c	0.30bc	0.31bc	0.29b	0.26b	0.27b	0.29b	0.27b
6000	0.23c	0.26c	0.28c	0.26c	0.22c	0.24bc	0.26b	0.24c
Mean	0.27b	0.31ab			0.27b	0.29ab	0.32a	
Second season: 2015								
0	0.31bc	0.33ab	0.41a	0.35a	0.26b	0.29ab	0.36a	0.30a
2000	0.26b	0.27b	0.28b	0.27b	0.21bc	0.24b	0.25b	0.23b
4000	0.23b	0.25b	0.28b	0.25b	0.20bc	0.22b	0.23b	0.22b
6000	0.21c	0.22bc	0.25b	0.23b	0.16c	0.19bc	0.21bc	0.19c
Mean	0.25b	0.27b	0.31a		0.21b	0.24ab	0.26a	

Means followed by the same letter in a column or row do not differ significantly according to Duncan's New Multiple Range *t*-Test at *P* = 0.05.

Chemical constituents:

It is clear from data presented in Tables (12, 13 and 14) that chlorophyll a, b and carotenoids content in the leaves (mg/g f.w.), as well as the percentages of nitrogen, phosphorus and potassium were descendingly decreased with increasing salinity level, but they were augmentatively raised as the rate of either humic acid or EM biostimulant was raised. Therefore, the highest content of such constituents was gained by combining between planting in fresh (salinized) soil mixture and the highest rate of either humic acid (10 cm³/l) or EM (5 cm³/l), whereas the least one was obtained by binding between cultivating in soil mixture salinized with 6000 ppm level and void of either humic acid or EM. This means that the individual applying of humic acid or EM at any rate markedly improve the content of the aforementioned constituents in the leaf tissues.

The opposite was the right in relation to the percentages of sodium and calcium, chloride (mg/100 g f.w.) and the free amino acid proline (mg/g f.w.), which were progressively increased in response to increasing either salinity level or humic acid and EM rate. So, the highest content of these constituents was recorded by connecting between the highest level of salinity and the higher one of either humic acid or EM biostimulant, with the exception for chloride content, that was gradually decreased with increasing the application rate of either humic acid or EM, and consequently the

highest record of Cl content were scored by planting in a soil mixture salinized with 6000 ppm level + applying either humic acid or EM at zero rate combined treatment, while the minimal content was found due to planting in a soil mixture free from salinity and applying both humic acid and EM at the highest rate. This clearly indicate the role of both humic acid and EM biostimulant in limiting the absorption of Cl element, which may cause toxicity for plant tissues when exist at high concentration.

Increasing the content of some chemical constituents in the leaves of hibiscus plants treated with either humic acid or EM may be ascribed to their role in activating the release of more nutrients necessary for healthy growth. In this connection, El-Khateeb *et al.*, (2011) elicited that the highest total carbohydrates in the roots of *Acacia saligna* was obtained with humic acid treatments (4 g/pot) regardless of irrigation interval. Kasmani *et al.*, (2013) cleared that chlorophyll content in the leaves of pistachio plants was increased by humic acid treatments (500-1500 ppm) regardless of subjecting them to severe drought stress. Gad *et al.*, (2015) reported that the combining between 7.5 ppm cobalt and 10 % humic acid resulted the highest mineral content (N, P, K, Mn, Zn and Cu) and chemical constituents (total proteins, total soluble solids, total carbohydrates, vitamins C and A) of okra (*Hibiscus esculantus*) pods. Tawila *et al.*, (2015) mentioned that 3 ml/l EM treatment improved chlorophyll a, b and carotenoids content in the leaves, as well as total soluble sugars in the stem, leaves and roots of Neem seedlings, plus reducing the content of Pb and Cd in the different parts of the seedlings, indicating the ability of such biostimulant in bioremediation of waste water.

Table 12: Effect of salinity, humic acid, EM and their interactions on pigments and nitrogen % in the leaves of *Hibiscus rosa-sinensis* L. plants during 2015 season.

Salinity level (ppm)	Chlorophyll a (mg/g f.w.)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	3.351	3.673	3.958	3.661	3.283	3.299	3.751	3.444
2000	2.339	2.731	3.295	2.788	2.297	2.591	2.647	2.512
4000	1.831	2.031	2.418	2.093	1.740	1.933	2.280	1.984
6000	1.676	1.956	2.145	1.926	1.569	1.810	2.043	1.807
Mean	1.299	2.598	2.954		2.222	2.408	2.680	
	Carotenoids (mg/g f.w.)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	1.508	1.751	1.765	1.675	1.475	1.436	1.705	1.539
2000	1.253	1.365	1.485	1.368	1.035	1.170	1.186	1.134
4000	0.833	0.946	1.089	0.956	0.783	0.855	1.053	0.897
6000	0.765	0.807	0.990	0.854	0.718	0.816	0.910	0.815
Mean	1.090	1.217	1.332		1.003	1.069	1.216	

Table 12: Cont.

Salinity level (ppm)	Chlorophyll b (mg/g f.w.)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	1.173	1.385	1.468	1.342	1.159	1.190	1.305	1.218
2000	0.846	0.967	1.155	0.989	0.805	0.911	1.046	0.921
4000	0.630	0.710	0.849	0.730	0.609	0.677	0.803	0.696
6000	0.583	0.664	0.735	0.661	0.548	0.569	0.700	0.606
Mean	0.808	0.932	1.052		0.780	0.837	0.964	
	N (%)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	2.31	3.14	3.57	3.01	2.23	2.86	2.93	2.67
2000	2.02	2.68	3.11	2.60	1.84	2.37	2.51	2.24
4000	1.07	2.25	2.63	1.98	1.00	1.38	1.75	1.38
6000	0.97	1.64	1.98	1.53	0.87	1.43	1.49	1.26
Mean	1.59	2.43	2.82		1.49	2.01	2.17	

Table 13: Effect of salinity, humic acid, EM and their interactions on phosphorus, potassium, sodium and calcium % in the leaves of *Hibiscus rosa-sinensis* L. plants during 2015 season.

Salinity level (ppm)	P (%)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	0.747	0.807	0.943	0.832	0.682	0.733	0.848	0.754
2000	0.708	0.750	0.789	0.749	0.631	0.656	0.701	0.663
4000	0.315	0.348	0.536	0.400	0.284	0.395	0.485	0.388
6000	0.134	0.167	0.268	0.190	0.135	0.158	0.243	0.179
Mean	0.476	0.518	0.634		0.433	0.486	0.569	
	Na (%)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	0.35	0.38	0.46	0.40	0.37	0.46	0.55	0.46
2000	0.46	0.60	0.71	0.59	0.60	0.69	0.71	0.67
4000	0.65	0.71	0.80	0.72	0.71	0.80	0.84	0.78
6000	0.84	0.84	0.93	0.87	0.82	0.89	0.91	0.87
Mean	0.58	0.63	0.73		0.63	0.71	0.75	

Table 13: Cont.

Salinity level (ppm)	K (%)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	1.533	1.697	1.826	1.685	1.368	1.493	1.590	1.484
2000	1.250	1.386	1.424	1.353	1.127	1.215	1.233	1.192
4000	1.036	1.139	1.215	1.130	0.911	1.003	1.103	1.006
6000	0.628	0.701	0.984	0.771	0.569	0.660	0.861	0.697
Mean	1.112	1.231	1.362		0.994	1.093	1.197	
	Ca (%)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	0.319	0.575	0.648	0.514	0.317	0.495	0.567	0.460
2000	0.501	0.638	0.731	0.623	0.431	0.568	0.681	0.560
4000	0.648	0.759	0.819	0.742	0.558	0.676	0.703	0.646
6000	0.924	0.863	1.000	0.929	0.895	0.750	0.863	0.836
Mean	0.598	0.709	0.800		0.550	0.622	0.704	

Table 14: Effect of salinity, humic acid, EM and their interactions on chloride and proline content of *Hibiscus rosa-sinensis* L. plants during 2015 season.

Salinity level (ppm)	Cl (mg/100 g f.w.)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	5.85	4.76	3.45	4.69	5.56	4.36	3.35	4.42
2000	7.31	8.37	5.49	7.06	6.94	7.78	5.34	6.69
4000	8.79	9.23	6.51	8.18	8.36	8.59	6.33	7.76
6000	11.70	9.75	7.89	9.78	10.43	9.10	7.69	9.07
Mean	8.41	8.03	5.84		7.82	7.46	5.68	
	Proline (mg/ g f.w.)							
	Humic acid (cm ³ /l)				EM (cm ³ /l)			
	0	5	10	Mean	0	3	5	Mean
0	0.227	0.246	0.336	0.270	0.231	0.251	0.353	0.278
2000	0.306	0.339	0.378	0.341	0.335	0.340	0.478	0.384
4000	0.395	0.451	0.497	0.448	0.389	0.467	0.657	0.504
6000	0.442	0.610	0.856	0.636	0.445	0.535	0.753	0.578
Mean	0.343	0.412	0.517		0.350	0.398	0.560	

From the previous findings, it can be advised to treat the Chinese hibiscus plants, monthly with either humic acid (10 cm³/l) or EM biostimulant (5 cm³/l), if planted in saline (NaCl + CaCl₂) soil up to 6000 ppm to improve their growth and performance.

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