Response Of Lettuce To Humic Acid And Salt Stress Condition Under Soilless Culture System

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ABSTRACT: The study was carried out at the private Farm of Borg El Arab, Alexandria, Egypt, during the seasons of (2016/2017 and 2017/2018) to study the effect of humic acid on lettuce yield under salt stress condition using soilless culture system. The experiments were carried out in a Factorial experiment consisted of sixteen treatments arranged in a Randomized Complete Block Design (RCBD) with three replicates for each treatment. The humic acid rates (0, 100, 200 and 400 mg/l) were arranged in the main plots, whereas, the four salt stress rates (control, 400, 800 and 1200mg/l) were arranged in the sub plots. The results indicated that, the yield and its components gradually increased with the increasing of humic acid rate at 400 mg/l such as, (head diameter, head fresh weight, head dry weight and yield/fed). Humic additions led to increase NPK content of lettuce leaves, total carbohydrates percentage and vitamin C content as compared with control treatment which recorded significantly decreased in all studied characteristics in both seasons. On another hand, salinity treatments significantly decreased all yield and its components whereas, sodium chloride at 1200 mg/l recorded the negative effect on head diameter, head fresh weight, head dry weight and yield/fed, also, it caused a significant decrease in chemical composition such as (N,P,K,Ca, Mg and Na), total carbohydrates percentage and vitamin C content of lettuce plants as compared with control treatment which recorded significantly increased in head diameter, head fresh weight, head dry weight and yield/fed, chemical composition i.e. (N, P, K, Ca, Mg and Na), total carbohydrates percentage and vitamin C content during both seasons.

Keywords: lettuce, humic acid, salt stress, yield, chemical composition.

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INTRODUCTION: Lettuce (*Lactuca sativa* L.) belongs to the family *Compositae* which is one of the most important vegetable crops in the world. It is one of the very important cash and food vegetable crops. Lettuce is more susceptible to nutrient deficiencies than most crop plants because of their shallow root system **(Yeshiwas *et al.*, 2018).** Its production and cultivation area in recent years has been increased due to marketability, medicinal properties and having various vitamins. It contains various vitamins such as A, C and B_9 as well as minerals such as calcium, phosphorus, potassium, manganese and iron, also, plays an important role in food digestion due to its high cellulose content. Lettuce is rich in antioxidants carotenoids, caffeic acid, and flavanols which are anti-carcinogenic **(Viacava *et al.*, 2014).** It also, provides some dietary fiber, carbohydrates, protein and a small amount of fat. Lettuce also, provides calcium, iron and copper, vitamins and minerals largely found in the leaf.

Hydroponics or soilless farming referred to the method of growing plants where essential nutrients components are provided in water **(Kim *et al.*, 2013; Falmata *et al.*, 2020).** This system involves the placement of plant roots in either a static or continuously aerated nutrient solution **(Nguyen *et al.*, 2016 and Shohael *et al.*, 2017).** Efficient nutrient regulation and efficient water use are the main advantages of hydroponics **(Resh 2016).**

Salt stress is one of the most widespread environmental concerns that restricted plant growth and function, especially in arid and semi-arid regions. About 2000 ha of arable land around the world are losing their productivity daily due to salinization. Salt stress in many crops reduces yield by 10-25% **(Shahid *et al.*, 2018).** Lettuce is a moderately salt-sensitive plant with electrical conductivity (EC) of 1.3 dS m^{-1} that salinity stress negatively affects its growth and yield **(Fernandez *et al.*, 2016).** The application of salt stress to increase the productivity of lettuce in long term application will decline soil fertility.

Several studies have evaluated the effect of the organic matter content on the fertility of soils. The humic substances, the major component of soil organic matter, have both direct and indirect effects on plant growth (**Sangeetha and Singaram, 2006**). The indirect effect of humic acid improves physical, chemical and biological condition of soil (**Halime *et al.*, 2011**). Whereas, the direct effects are those that require the uptake of humic substances into the plant tissue resulting in various biochemical actions exerted at the cell wall, membrane or cytoplasm and mainly of hormonal nature (**Chen *et al.*, 2004 and Eyheraguibel *et al.*, 2008**). On the other hand, humic acid has beneficial effects on nutrient uptake by plants and was particularly important for transportation and availability of macro and micro nutrients (**Anonymous, 2010**).

The main objective of this study is to investigate of the effect of humic acid on the productivity and quality of lettuce under salt stress condition in soilless culture system.

MATERIALS AND METHODS

The experiments were conducted at a private Farm of Borg El Arab, Alexandria, Egypt, during the seasons of (2016/2017 and 2017/2018) to study the effect of humic acid on lettuce yield growing under salt stress condition using soilless culture system. Lettuce (*Lactuca Sativa* L.) cv. Batvia green and cv. Osely were planted on 17^{th} of October and harvested on 8^{th} of December in the two seasons of (2016/2017 and 2017/2018). seeds were sown singly in seedling tray (ST) ($2.5\times2.5\times3.5$ cm^3) filled with commercial peat moss and vermiculite. These trays were kept moist at 25±2°C until germination. Seedlings were kept in seedling trays for 22 days when the plants consist of three true leaves and afterwards transplanted according to a Randomized Complete Block Design (RCBD) with three replicates for each treatment to the cups(9cm) full of peat moss and vermiculite at rates (1:1), these cups were perforated from below and then put on perforated plastic pipes of size (2 m length × 0.4 m width) , the distance of the cultivation was 20 cm and the number of plants in each channel were 8 plants. The numbers of channel frames in the entire experiment were 32 (16 for each level). Humic acid levels (0, 100, 200 and 400 mg/l) were arranged in the main plots, whereas, the four salt stress levels (control, 400, 800 and 1200mg/l) were arranged in the sub plots. To impose salinity, sodium chloride (NaCl) salt was used in this experiment. Seedlings were fertilized with full strength Allen cooper solution (Allen cooper, 1979). The pH level of the treatments were maintained at 6.5 to 8.0. Cultivation channels were sterilized with a solution of (HCL 10%) and then each channel filled with 8 liter of water then the treatments solutions. Each channel connected with induvidual tank (fertigation tank) then connected with air pump to renew the pipes air.

Reconditioning of plants before treated:
Seedlings in the cups were treated inside channels with regular water for three days without adding the treatments and then recommended treatments were added to each channel. The plants were harvested after 52 days from culture.

Table (1): Weights (g) of pure substances to be dissolved in (1000 liters of water to give theoretically ideal concentrations:

Substance	Formula	Weight (g)
Potassium dihydrogen phosphate	KH_2PO_4	263
Potassium nitrate	KHO_3	583
Calcium nitrate	$Ca (NO_3)_24H_20$	1003
Magnesium sulphate	$Mg SO_4.7H_20$	513
EDTA iron	$[CH_2 N(CH_2.C00)_2]_2$ FeNa	79
Manganous sulphate	$MnSO_4.H_20$	6.1
Boric acid	HBO_3	1.7
Copper sulphate	$CuSO_45H_20$	0.39
Ammonium molybdate	$(NH_4)_6Mo_70_{24}4H_20$	0.37
Zinc sulphate	$ZnSO_4.7H_20$	0.44

Data recorded

A) Yield and yield components

- Head diameter (cm)
- Head fresh weight (g)
- Head dry weight (g)
- Yield (t/fed)

B) Chemical composition:

The NPK contents as percentages were determined in the leaves of lettuce. The dry weight was determined by drying the heads of lettuce in a drying chamber to a constant weight at 75°C for 72 hour according to **Tandon (1995).** After dryness, the plant samples were milled and stored for analysis as reported. However, 0.5g of the tubers powder was wet-digested with H_2SO_4–H_2O_2 mixture according **(Lowther, 1980)** and the following determinations were carried out in the digested solution to determine the following:

- **Nitrogen content (N %):**
  Total nitrogen was determined in digested plant material colorimetrically by Nessler`s method **(Chapman and Pratt, 1978)**. Nessler's solution (35 KI/100 ml distilled water (d. w.) + 20 g $HgCl_2$ / 500 ml d. w.) +120 g NaOH / 250 ml d. w. Reading was achieved using wave length of 420 nm and N was determined as percentage as follows:
  % N = NH4 % x 0.776485
- **Phosphorus content (P %):**
  Phosphorus was determined by the Vanadomolyate yellow method as given by **Jackson (1973)** and the intensity of color developed was read in spectrophotometer at 405nm wave length.
- **Calcium, magnesium, sodium and potassium content (Ca, Mg,Na and K%):**
  Calcium, magnesium, sodium and potassium were determined according to the method described by method **Jackson (1973)** using Beckman Flame photometer.
- **Vitamin C (Ascorbic acid):**
  The ascorbic acid content of the juice was determined by titration with 2, 6-dichlorophenol-indo-phenol **(AOAC, 1985)** and calculated as milligrams per 100 ml of juice.
- **Total carbohydrates:**
  Total carbohydrates were determined, quantitatively, in lettuce by Anthron method according to **Mahadevan and Sridhar (1986)** as follows:

  Extraction was carried out by grinding dry matter in Mahadavan buffer (sodium citrate buffer, pH 6.8). Extracts were homogenized for 3 minutes and centrifuged at 4000 rpm for 15 min. the supernatant was then used to determine total carbohydrates.

Statistical analysis

The obtained data were subjected to the proper method of statistical analysis of variance as described by **Gomez and Gomez (1984).** The treatment means were compared using the revised least significant differences (L.S.D.) test at 0.05 and 0.01 levels of probability.

RESULTS AND DISCUSSION

A) Yield and yield components

The obtained results in **Table (2), fig. (1 to 8)** showed that all treatments of humic acid caused a marked effect on head diameter, head fresh weight, head dry weight and yield/fed. of lettuce plants during both seasons. It is quite clear from data that increasing the application rate of humic acid up to 400 mg/l significantly increased head diameter (26.16 and 29.30 cm), head fresh weight (382.56 and 428.46 g), head dry weight (76.51 and 85.69 g) and yield /fed. (30.90 and 34.61 t/fed), as compared with plants grown under control conditions which recorded the lowest mean values of head diameter (19.07 and 21.36 cm), head fresh weight (278.88 and 312.35g), head dry weight (55.78 and 62.47g), and yield /fed. (22.53 and 25.23 t/fed), during both seasons. The increases in shoots characteristics might be due to the influence of humic acid which provides nutrient minerals that involve in plants bioactivities and finally leads to growth induction (**Abdel Mawgaud *et al.*, 2007**). Also, Similar finding was achieved by **Winsor and Schwarz (1990)** who reported that humic acid contains cytokinins and their application resulted in increasing endogenous cytokinin and auxin levels which possibly leading to improve yield.

Table (2): Effect of humic acid levels and NaCl concentration on head diameter, head fresh and dry weight and yield/fed. of lettuce plants during 2016/2017 and 2017/2018 seasons.

Treatments mg/l		**Head diameter (cm)**		**Head fresh weight (g)**		**Head dry weight (g)**		**Yield (t/fed.)**	
		2016/17	**2017/18**	**2016/17**	**2017/18**	**2016/17**	**2017/18**	**2016/17**	**2017/18**
A) Humic acid									
Control		19.07d	21.36d	278.88d	312.35d	55.78d	62.47d	22.53d	25.23d
100		21.19c	23.73c	309.86c	347.05c	61.97c	69.41c	25.03c	28.03c
200		23.55b	26.37b	344.30b	385.61b	68.86b	77.12b	27.81b	31.15b
400		26.16a	29.30a	382.56a	428.46a	76.51a	85.69a	30.90a	34.61a
$LSD_{(0.05)}$		**0.13**	**0.15**	**0.60**	**0.68**	**0.12**	**0.13**	**0.17**	**0.20**
B) NaCl									
Control		30.48a	34.14a	445.67a	499.15a	89.13a	99.83a	36.00a	40.32a
400		24.38b	27.31b	356.53b	399.32b	71.31b	79.86b	28.80b	32.26b
800		19.51c	21.85c	285.23c	319.45c	57.04c	63.89c	23.04c	25.81c
1200		15.60d	17.48d	228.18d	255.56d	45.64d	51.11d	18.43d	20.64d
$LSD_{(0.05)}$		**0.13**	**0.15**	**0.60**	**0.68**	**0.12**	**59.45**	**0.17**	**0.20**
Interaction (AXB)		**	**	**	**	**	**	**	**
HA	**NaCl**								
Control	Control	25.84	28.95	377.89	423.24	75.58	84.65	30.53	34.19
	400	20.68	23.16	302.31	338.59	60.46	67.72	24.42	27.35
	800	16.54	18.52	241.85	270.87	48.37	54.17	19.54	21.89
	1200	13.23	14.82	193.48	216.70	38.70	43.34	15.63	17.50
100	Control	28.71	32.16	419.88	470.26	83.98	94.05	33.92	37.99
	400	22.97	25.73	335.90	376.21	67.18	75.24	27.13	30.39
	800	18.38	20.58	268.72	300.97	53.74	60.19	21.70	24.31
	1200	14.70	16.46	214.98	240.78	43.00	48.16	17.37	19.45
200	Control	31.91	35.73	466.53	522.51	93.30	104.50	37.69	42.21
	400	25.52	28.59	373.23	418.01	74.64	83.60	30.15	37.77
	800	20.42	22.87	298.58	334.41	59.72	66.88	24.12	27.01
	1200	16.34	18.30	238.86	267.53	47.77	53.50	19.30	21.61
400	Control	35.45	39.70	518.37	580.57	103.67	116.11	41.87	46.90
	400	28.36	31.76	414.69	464.46	82.94	92.89	33.50	37.52
	800	22.69	25.41	331.75	371.57	66.35	74.31	26.80	30.02
	1200	18.15	20.33	265.41	297.25	53.08	59.45	21.44	24.01
$LSD_{(0.05)}$		**0.15**	**0.17**	**0.70**	**0.78**	**0.14**	**0.16**	**0.20**	**0.23**

Values with the same alphabetical letters, within a comparable group of means , don't significant differ, using L.S.D test at 0.05 level.

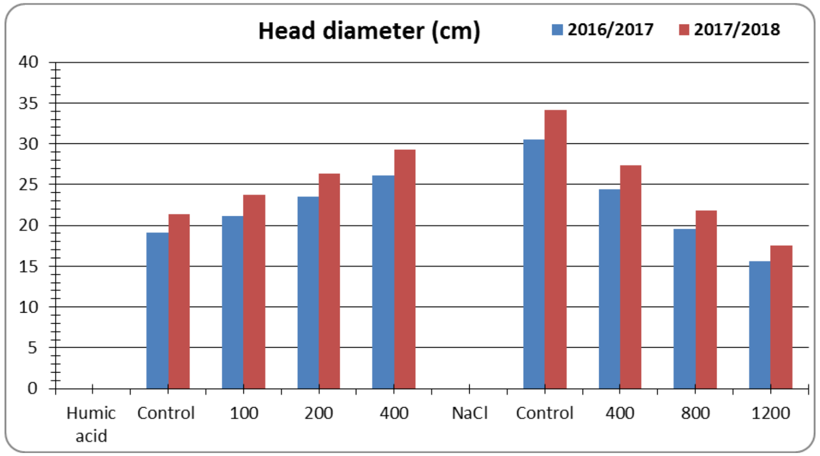

Fig. (1): Effect of humic acid levels and NaCl concentration on head diameter during 2016/2017 and 2017/2018 seasons.

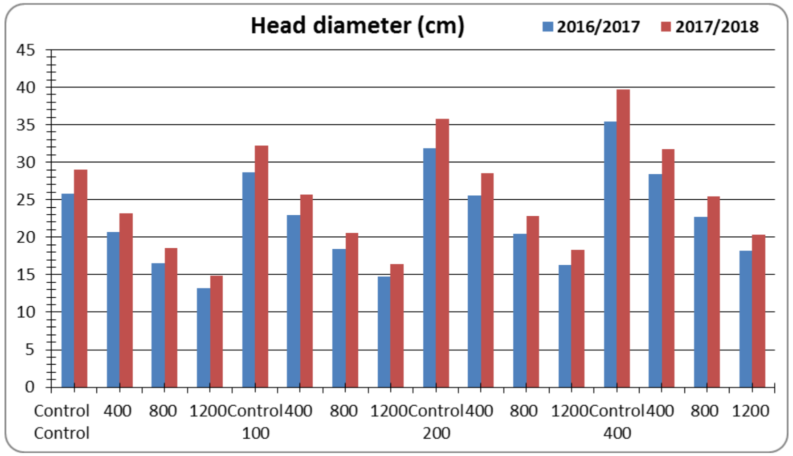

Fig. (2): Interaction between humic acid levels and NaCl concentration on head diameter during 2016/2017 and 2017/2018 seasons.

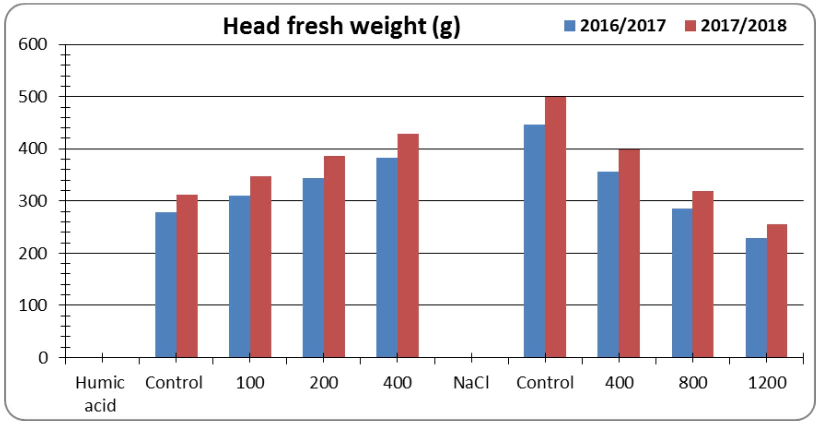

Fig. (3): Effect of humic acid levels and NaCl concentration on head fresh weight during 2016/2017 and 2017/2018 seasons.

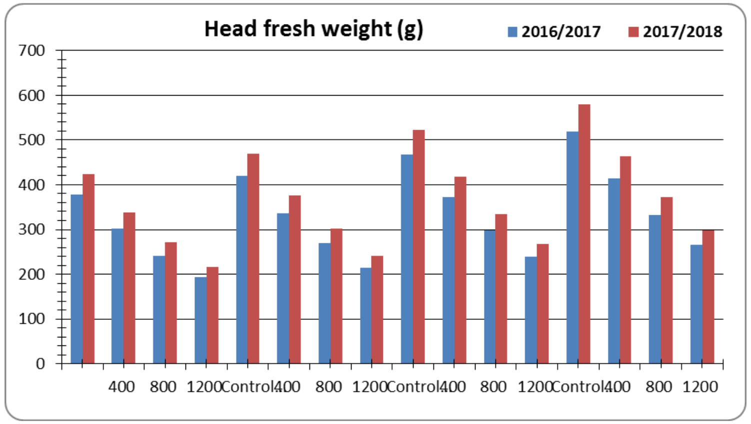

Fig. (4): Interaction between humic acid levels and NaCl concentration on head fresh weight during 2016/2017 and 2017/2018 seasons.

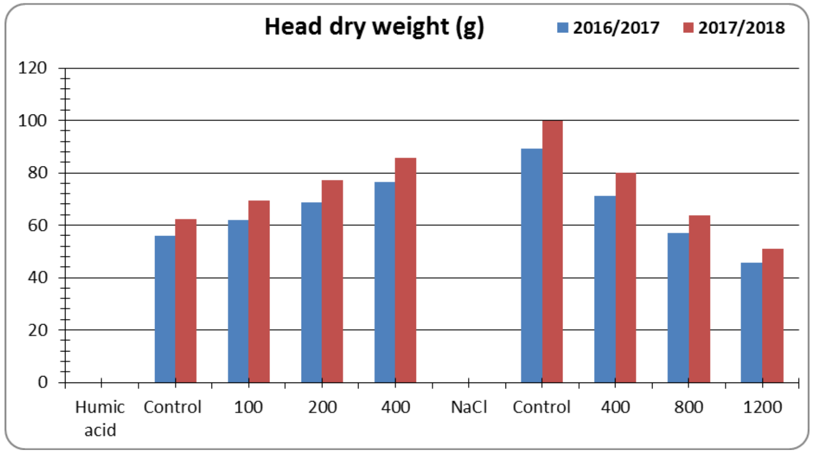

Fig. (5): Effect of humic acid levels and NaCl concentration on head dry weight during 2016/2017 and 2017/2018 seasons.

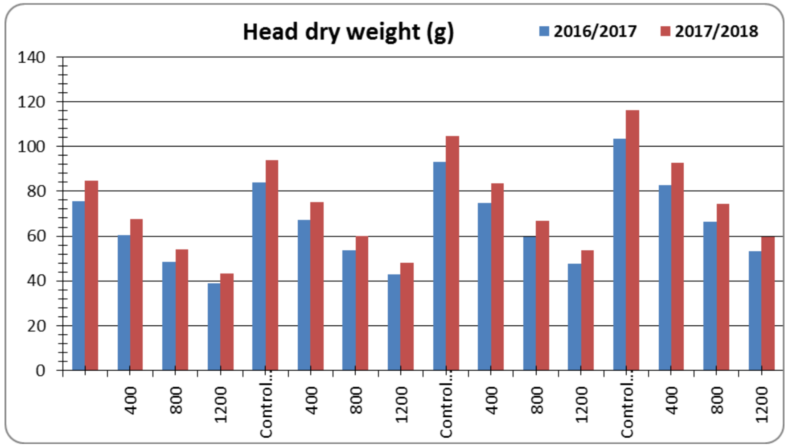

Fig. (6): Interaction between humic acid levels and NaCl concentration on head dry weight during 2016/2017 and 2017/2018 seasons.

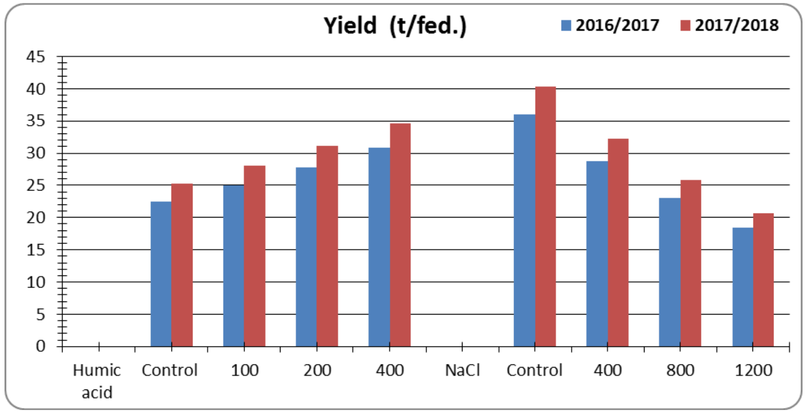

Fig. (7): Effect of humic acid levels and NaCl concentration on yield (t/fed.) during 2016/2017 and 2017/2018 seasons.

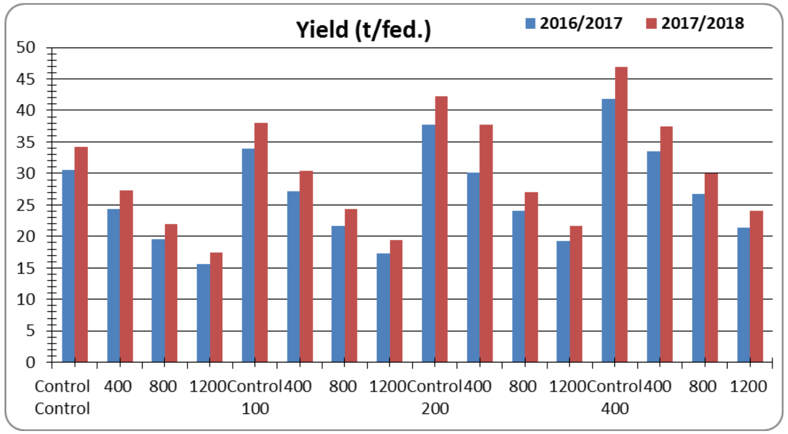

Fig. (8): Interaction between humic acid levels and NaCl concentration on yield (t/fed.) during 2016/2017 and 2017/2018 seasons

On the contrary, all salinity treatments significantly decreased head diameter, head fresh weight, head dry weight and yield (t/fed.) of lettuce plants as compared with control treatment, during both seasons. However, control treatment gave the highest mean values of head diameter (30.48 and 34.14 cm), head fresh weight (445.67 and 499.15 g), head dry weight (89.13 and 99.83 g), and yield/fed. (36.00 and 40.32 t/fed.), as compared with high concentration of NaCl (1200 mg/l) which recorded the lowest mean values of head diameter (15.60 and 17.48 cm), head fresh weight (228.18 and 255.56g), head dry weight (45.64 and 51.11g), and yield/fed (18.43 and 20.64 t/fed), during both seasons. The findings were similar to those reported by **Bar-Yosef *et al*. (2005)**. Number of leaves was reduced significantly with increasing salinity levels, which confirms the results of **Ünlükara *et al*. (2008)** but this is in contrast to the findings of **Andriolo *et al*. (2005)** who reported that number of lettuce leaves was not affected by salinity treatments. Plant dry matter was significantly reduced with increasing salinity but this is inconsistent with the results of **Ünlükara *et al*. (2008)** who found that plant dry matter content increased with increasing salinity for the salinity range studied. Lettuce yield response to the three salinity levels in soilless culture system solutions was similar with the findings of **Karam *et al*. (2005)**. **Ünlükara *et al*. (2008)** also reported that salinity reduced the yield of lettuce in a constant manner. In addition, gas exchange rates, stomatal conductance and product quality were reduced by salinity level **(De Pascale & Barbieri, 1995)**. These may be attributed to low uptake or decreased xylem transport of calcium or to disturbed partitioning of cations in plant tissues at high concentration of sodium ions in the solution **(Sonneveld, 1988).**

The interaction between humic acid and salinity treatments was highly significant on head diameter head fresh weight, head dry weight and yield/fed of lettuce plants during both seasons.

Table (2): Effect of humic acid levels and NaCl concentration on head diameter, head fresh and dry weight and yield/fed. of lettuce plants during 2016/2017 and 2017/2018 seasons.

Treatments mg/l		Head diameter (cm)		Head fresh weight (g)		Head dry weight (g)		Yield (t/fed.)	
		2016/17	2017/18	2016/17	2017/18	2016/17	2017/18	2016/17	2017/18
C) Humic acid									
Control		19.07d	21.36d	278.88d	312.35d	55.78d	62.47d	22.53d	25.23d
100		21.19c	23.73c	309.86c	347.05c	61.97c	69.41c	25.03c	28.03c
200		23.55b	26.37b	344.30b	385.61b	68.86b	77.12b	27.81b	31.15b
400		26.16a	29.30a	382.56a	428.46a	76.51a	85.69a	30.90a	34.61a
$LSD_{(0.05)}$		**0.13**	**0.15**	**0.60**	**0.68**	**0.12**	**0.13**	**0.17**	**0.20**
D) NaCl									
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800		19.51c	21.85c	285.23c	319.45c	57.04c	63.89c	23.04c	25.81c
1200		15.60d	17.48d	228.18d	255.56d	45.64d	51.11d	18.43d	20.64d
$LSD_{(0.05)}$		**0.13**	**0.15**	**0.60**	**0.68**	**0.12**	**59.45**	**0.17**	**0.20**
Interaction (AXB)		**	**	**	**	**	**	**	**
HA	**NaCl**								
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	400	22.97	25.73	335.90	376.21	67.18	75.24	27.13	30.39
	800	18.38	20.58	268.72	300.97	53.74	60.19	21.70	24.31
	1200	14.70	16.46	214.98	240.78	43.00	48.16	17.37	19.45
200	Control	31.91	35.73	466.53	522.51	93.30	104.50	37.69	42.21
	400	25.52	28.59	373.23	418.01	74.64	83.60	30.15	37.77
	800	20.42	22.87	298.58	334.41	59.72	66.88	24.12	27.01
	1200	16.34	18.30	238.86	267.53	47.77	53.50	19.30	21.61
400	Control	35.45	39.70	518.37	580.57	103.67	116.11	41.87	46.90
	400	28.36	31.76	414.69	464.46	82.94	92.89	33.50	37.52
	800	22.69	25.41	331.75	371.57	66.35	74.31	26.80	30.02
	1200	18.15	20.33	265.41	297.25	53.08	59.45	21.44	24.01
$LSD_{(0.05)}$		**0.15**	**0.17**	**0.70**	**0.78**	**0.14**	**0.16**	**0.20**	**0.23**

Values with the same alphabetical letters, within a comparable group of means , don't significant differ, using L.S.D test at 0.05 level.

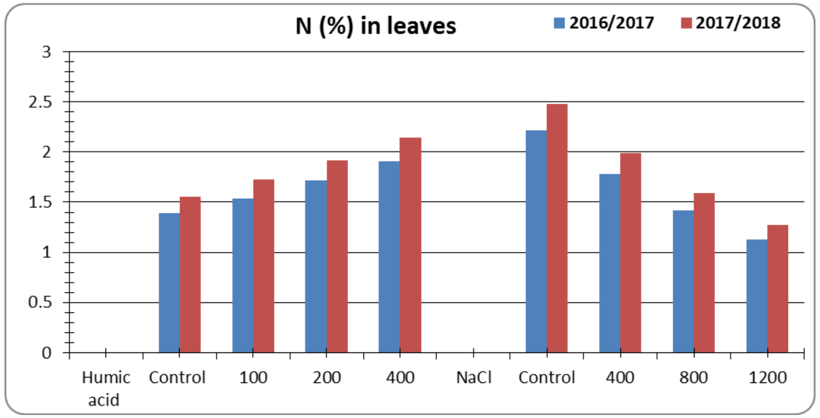

Fig. (9): Effect of humic acid levels and NaCl concentration on N (%) in leaves during 2016/2017 and 2017/2018 seasons.

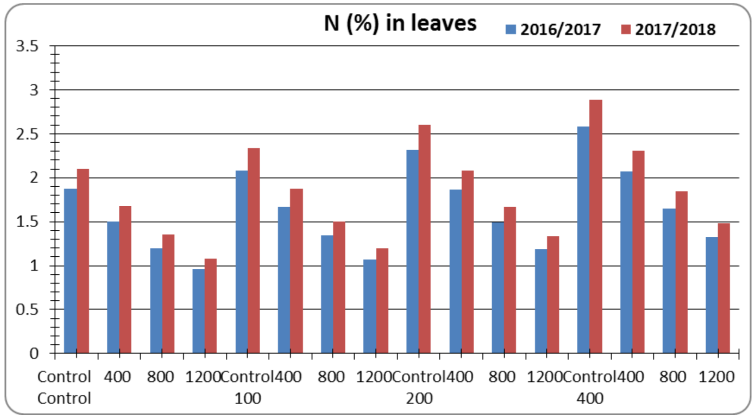

Fig. (10): Interaction between humic acid levels and NaCl concentration on N (%) in leaves during 2016/2017 and 2017/2018 seasons.

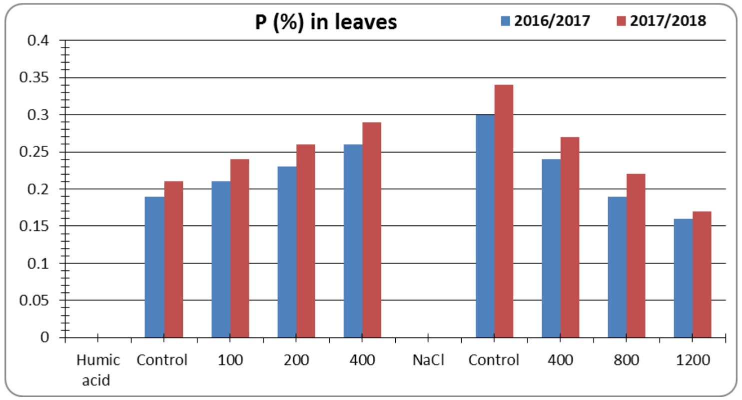

Fig. (11): Effect of humic acid levels and NaCl concentration on P (%) in leaves during 2016/2017 and 2017/2018 seasons.

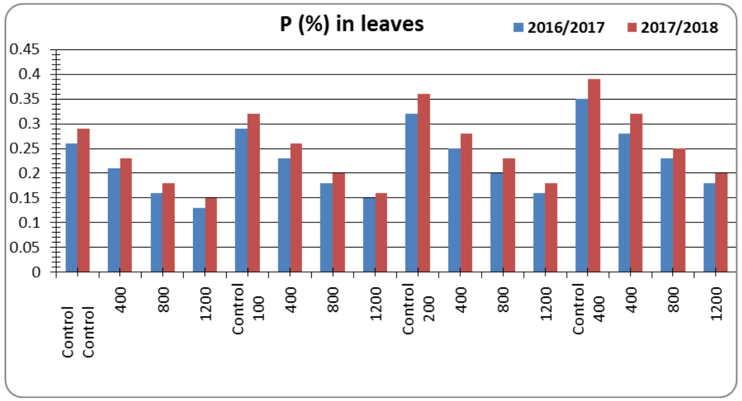

Fig. (12): Interaction between humic acid levels and NaCl concentration on P (%) in leaves during 2016/2017 and 2017/2018 seasons.

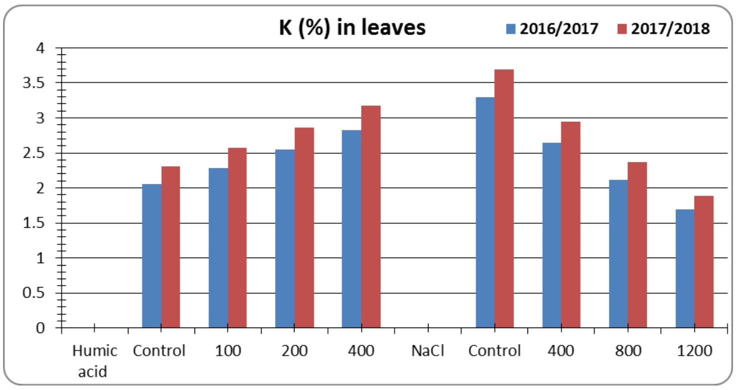

Fig. (13): Effect of humic acid levels and NaCl concentration on K (%) in leaves during 2016/2017 and 2017/2018 seasons.

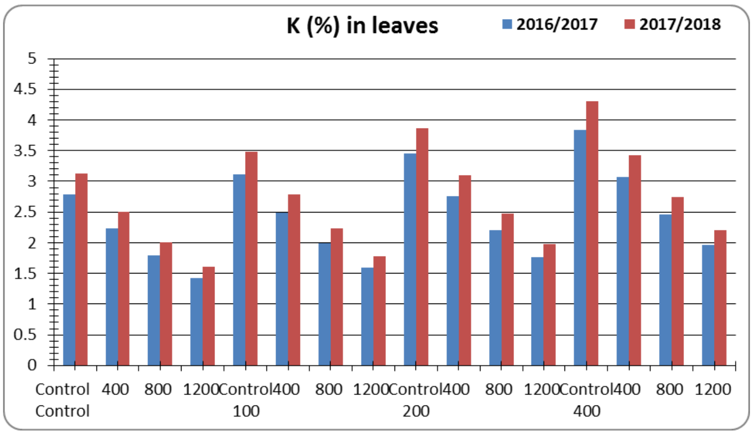

Fig. (14): Interaction between humic acid levels and NaCl concentration on K (%) in leaves during 2016/2017 and 2017/2018 seasons.

- Vitamin C (mg/ 100 g f.w.)

Significant variation was recorded for vitamin C of lettuce with application of different levels of humic acid is presented in Table (4) and Fig. (15 and 16). It was revealed that the higher doses of humic acid up to 400 mg/l showed higher vitamin C content (35.87 and 40.17mg /100g f.w.), followed by humic acid rate 200 mg/l (30.20 and 33.82 mg /100g f.w.), as compared with plants grown under control conditions which recorded the lowest mean values of vitamin C content (26.15 and 29.29 mg /100g f.w.), during both seasons. The increase in quality character (vitamin c) might be due to the growth promoting substances which could have accelerated synthesis of carbohydrate, vitamins and other quality characters. These results are in conformity with those of Fageria et al. (1992) and Singh and Singh (1992). Since humic acid was reported to stimulate photosynthesis (Nardi et al., 2002), there must have been more assimilates for the plants which increased their vitamin C.

In this respect, all salinity treatments were significant decreased vitamin C content in leaves as compared with control treatment, during both seasons. However, control treatment gave the higest mean values of vitamin C content in leaves (41.78 and 46.80), followed by 400mg/l NaCl (33.43 and 37.44 mg /100g f.w.), as compared with high concentration of NaCl (1200 mg/l) which recorded the lowest mean values of vitamin C in leaves (21.39 and 23.96 mg /100g f.w.), during both seasons. These are in agreement of those (Parida and Das 2005), they reported that salt stress led to a significant increase in the level of electrolyte leakage in many crops. These results are concordant with those of Karlidag et al. (2011) who determined that salinity facilitated the maintenance of membrane functions. This facilitation could be attributed to the induction of antioxidant responses and elevated Ca uptake that protects the plant from the oxidative damage and increase vitamin C content.

The interaction between humic acid and salinity treatments was highly significant on vitamin C content in leaves during both seasons.

- Total carbohydrates (%)

Results in Table (4) and Figs. (17 and 18) indicated that increasing humic acid rate increased total carbohydrates percentages, as compared with control treatment during both seasons. It was found that application of humic acid (400mg/l) resulted in significantly greater of total carbohydrates percentages (36.01 and 40.33 %), followed by humic acid rate 200mg/l (32.41 and 36.30%), as compared with control treatment which recorded the lowest mean values of total carbohydrates percentages (26.25 and 29.40%), during both seasons. Humic acid compounds may have a variety of biochemical effects either at cell wall, membrane level or in the cytoplasm, including improved photosynthesis, carbohydrates formation and respiration rates in plants, better protein synthesis and plant hormone like activity (Chen and Aviad, 1990).

Table (4): Effect of humic acid levels and NaCl concentration on Vitamin C and Total carbohydrate percentages in leaves during 2016/2017and 2017/ 2018 seasons.

Treatments mg/l		**Vitamin C (mg/ 100 g f.w.)**		**Total carbohydrate (%)**	
		2016/2017	2017/2018	2016/2017	2017/2018
A) Humic acid (HA)					
Control		26.15c	29.29c	26.25d	29.40d
100		29.05bc	32.54bc	29.17c	32.67c
200		30.20b	33.82b	32.41b	36.30b
400		35.87a	40.17a	36.01a	40.33a
$LSD_{(0.05)}$		**3.01**	**3.37**	**0.35**	**0.40**
B) NaCl					
Control		41.78a	46.80a	41.95a	46.99a
400		33.43b	37.44b	33.56b	37.59b
800		24.66c	27.62c	26.85c	30.07c
1200		21.39d	23.96d	21.48d	24.06d
$LSD_{(0.05)}$		**3.01**	**3.37**	**0.35**	**0.40**
Interaction (AXB)		**ns**	**ns**	**	**
HA	**NaCl**				
Control	Control	35.43	39.68	35.57	39.84
	400	28.34	31.75	28.46	31.87
	800	22.67	25.39	22.77	25.50
	1200	18.14	20.32	18.21	20.40
100	Control	39.37	44.09	39.53	44.27
	400	31.50	35.28	31.62	35.42
	800	25.20	28.22	25.30	28.33
	1200	20.15	22.57	20.24	22.67
200	Control	43.74	48.99	42.92	49.19
	400	34.99	39.19	35.13	39.35
	800	19.66	22.02	28.11	31.48
	1200	22.40	25.08	22.48	25.18
400	Control	48.60	54.43	48.80	54.65
	400	38.88	43.55	39.04	43.72
	800	31.10	34.84	31.23	34.98
	1200	24.88	27.87	24.98	27.98
$LSD_{(0.05)}$		**3.47**	**3.89**	**0.41**	**0.46**

Values with the same alphabetical letters, within a comparable group of means , don't significant differ, using L.S.D test at 0.05 level.

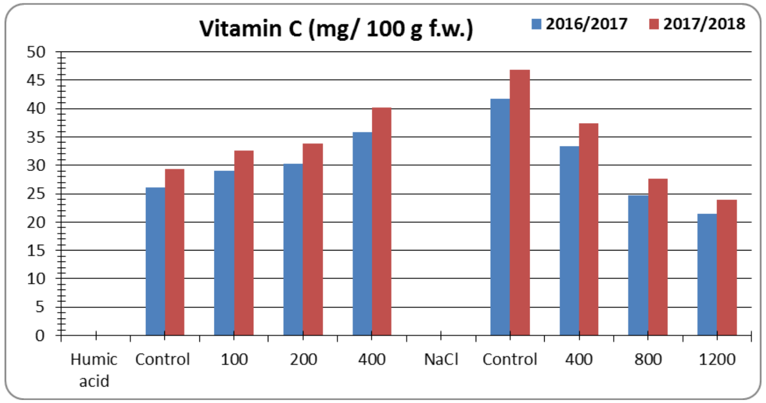

Fig. (15): Effect of humic acid levels and NaCl concentration on vitamin C (mg/ 100 g f.w.) during 2016/2017 and 2017/2018 seasons.

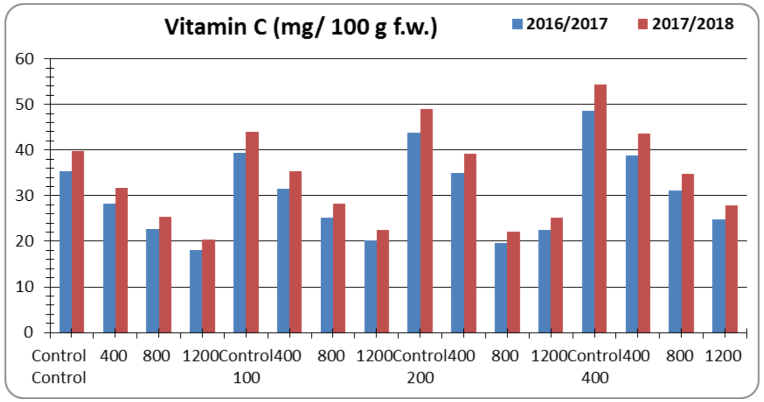

Fig. (16): Interaction between humic acid levels and NaCl concentration on vitamin C (mg/ 100 g f.w.) during 2016/2017 and 2017/2018 seasons.

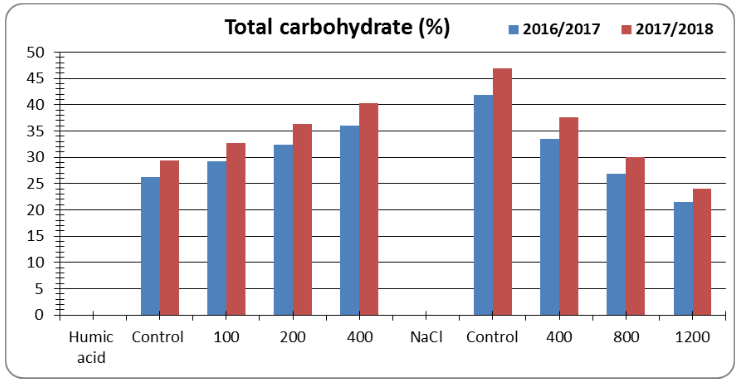

Fig. (17): Effect of humic acid levels and NaCl concentration on total carbohydrates (%) during 2016/2017 and 2017/2018 seasons.

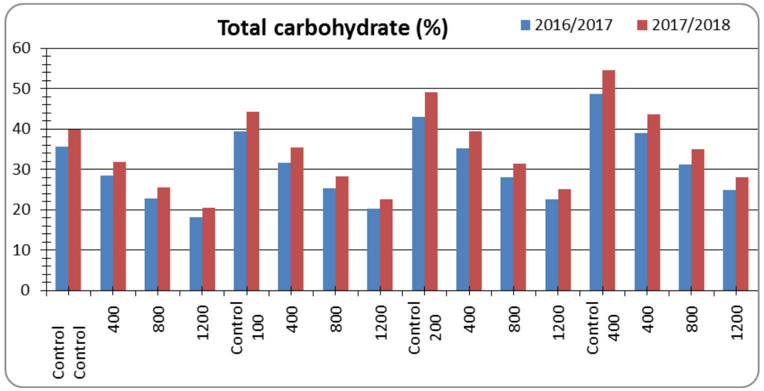

Fig. (18): Interaction between humic acid levels and NaCl concentration on total carbohydrates (%) during 2016/2017 and 2017/2018 seasons.

In this respect, all salinity treatments were significant decreased total carbohydrates percentages in leaves as compared with control treatment, during both seasons. However, control treatment gave the highest mean values of total carbohydrates percentages in leaves (41.95 and 46.99), followed by 400mg/l NaCl (33.56 and 37.59 mg /100g f.w.), as compared with high concentration of NaCl (1200 mg/l) which recorded the lowest mean values of total carbohydrates percentages in leaves (21.48 and 24.06 mg /100g f.w.), during both seasons. However, as reported by **Ashraf and Harris (2004),** the role of carbohydrates in the salinity tolerance is not clear and further investigations are needed to conclude that they are universally associated with salt tolerance, because the variations in the accumulation of these compounds could vary among species. In our material, the tissue levels of total sugars were not affected by salinity and treatment applications, in fact all plants showed similar concentrations. These results indicated that treated plants did not show salinity stress under the conditions applied.

The interaction between humic acid and salinity treatments was highly significant on total carbohydrates percentages content during both seasons.

Calcium (%) in leaves

Results in **Table (5) and Figs. (19-20)** show that positive effect of all humic acid treatments on calcium content in leaves of lettuce plants compared with untreated plants during both seasons. However, humic acid up to 400 mg/l recorded the highest mean values of calcium content in leaves (1.16 and 1.30 %), followed by humic acid rate 200 mg/l (1.05 and 1.18 %), as compared with plants grown under control conditions which recorded the lowest mean values of calcium content in leaves (0.85 and 0.95 %), during both seasons.

Furthermore, all salinity treatments were significant decreased calcium content in leaves as compared with control treatment, during both seasons. However, control treatment gave the highest mean values of calcium content in leaves (1.36 and 1.52 %), followed by 400mg/l NaCl (1.08 and 1.21 %), as compared with high concentration of NaCl (1200 mg/l) which recorded the lowest mean values of calcium content in leaves (0.69 and 0.78%), during both seasons. Salt stress disturbs the uptake of essential mineral nutrients such as K^+ and Ca^{2+}, as Na^+ competitively inhibits K^+ and ca^{2+} transport through membranes (**Zhao *et al*., 2007**). The results showed a reduction of calcium content in shoots as a result of salt stress, and this reduction is most probably due to the competition of Na^+ for the same cation transporters (**Azevedo-neto and Tabosa, 2000; Neocleous *et al*., 2014**).

The interaction between humic acid and salinity treatments was highly significant on calcium content in leaves during both seasons.

4.4.8 Magnesium (%) in leaves

Results in **Table (5) and Figs. (21-22)** it was found that, with humic acid, magnesium percentage of lettuce leaves was significant increased as compared with control treatment, during both seasons, where humic acid up to 400 mg/l recorded the highest mean values of magnesium content in leaves (0.77 and 0.87 %), followed by humic acid rate 200 mg/l (0.69 and 0.78 %), as compared with plants grown under control conditions which recorded the lowest mean values of magnesium content in leaves (0.56 and 0.63 %), during both seasons.

Likewise, all salinity treatments were significantly decreased magnesium content in leaves as compared with control treatment, during both seasons. However, control treatment gave the highest mean values of magnesium content in leaves (0.90 and 1.01 %), followed by 400mg/l NaCl (0.72 and 0.81 %), as compared with high concentration of NaCl (1200 mg/l) which recorded the lowest mean values of magnesium content in leaves (0.46 and 0.52%), during both seasons. These are in accordance with those **(Csaba *et al*., 2015)**. They reported that salinity affect photosynthetic carbon uptake, but reduces moisture accumulation during storage of lettuce heads. Salt stress tends to reduce stomatal conductance in a short period after exposure. A higher conductance enables a better carbon dioxide supply for a sustained photosynthetic assimilation, resulting in a smaller reduction of biomass production. A direct correlation between stomatal conductance and salt stress tolerance was also observed in maize cultivars (Azevedo-neto *et al*., 2004).

The interaction between humic acid and salinity treatments was highly significant on magnesium content in leaves during both seasons.

Sodium (%) in leaves

Results illustrated in **Table (5) and Figs. (23-24)** show the effect of different doses of humic acid on sodium content in leaves of lettuce as compared with control treatment, during both seasons. Increasing humic acid rate decreasing sodium content in leaves, where humic acid up to 400 mg/l recorded the lowest mean values of sodium content in leaves (1.61 and 1.81 %), followed by humic acid rate 200 mg/l (2.26 and 2.53 %), as compared with plants grown under control conditions which recorded the highest mean values of sodium content in leaves (4.37 and 4.89 %), during both seasons.

On another side, all salinity treatments significantly increased sodium content in leaves as compared with control treatment, during both seasons. However, control treatment gave the lowest mean values of sodium content in leaves (1.90 and 2.13 %), followed by 400mg/l NaCl (2.50 and 2.80 %), as compared with high concentration of NaCl (1200 mg/l) which recorded the highest mean values of sodium content in leaves (3.90 and 4.37%), during both seasons. Lettuce cultivar growth in saline conditions showed an increase in Na^+ concentration. Therefore, two main strategies of salt stress tolerance can be considered, i.e. salt exclusion and salt sequestration, the latter one is used by lettuce cultivar **(Csaba *et al.*, 2015).** T In a recent study on different cultivars of barley, **Shabala *et al*. (2010)** conclude that after one week of salt treatment (320 mM NaCl), shoot Na^+ content of the tolerant variety was about 20 % higher than in the sensitive genotype. In the first phase of the salt stress the rapidly accumulating Na^+ is an osmolite with low energy cost in the leaf vacuoles for the adjustment of cell turgor, and ultimately of tissue growth under the hyperosmotic stress condition imposed by salinity (**Munns and Tester, 2008; Shabala *et al*., 2010**).

The interaction between humic acid and salinity treatments was highly significant on sodium content in leaves during both seasons.

Table (5): Effect of humic acid levels and NaCl concentration on Ca, Mg and Na percentages in leaves during 2016/2017 and 2017/2018 seasons.

Treatments		In leaves					
		Ca (%)		Mg (%)		Na (%)	
		2016/2017	2017/2018	2016/2017	2017/2018	2016/2017	2017/2018
A) Humic acid							
Control		0.85d	0.95d	0.56d	0.63d	4.37a	4.89a
100		0.94c	1.06c	0.63c	0.70c	3.18b	3.56b
200		1.05b	1.18b	0.69b	0.78b	2.26c	2.53c
400		1.16a	1.30a	0.77a	0.87a	1.61d	1.81d
$LSD_{(0.05)}$		**0.01**	**0.01**	**0.004**	**0.01**	**0.01**	**0.02**
B) NaCl							
Control		1.36a	1.52a	0.90a	1.01a	1.90d	2.13d
400		1.08b	1.21b	0.72b	0.81b	2.50c	2.80c
800		0.87c	0.97c	0.57c	0.64c	3.12b	3.50b
1200		0.69d	0.78d	0.46d	0.52d	3.90a	4.37a
$LSD_{(0.05)}$		**0.01**	**0.01**	**0.004**	**0.01**	**0.01**	**0.02**
Interaction (AXB)		**	**	**	**	**	**
HA							
Control	Control	1.15	1.28	0.76	0.85	2.71	3.04
	400	0.92	1.03	0.61	0.68	3.87	4.34
	800	0.74	0.83	0.49	0.55	4.84	5.42
	1200	0.59	0.66	0.39	0.44	6.05	6.78
100	Control	1.28	1.43	0.85	0.95	2.20	2.47
	400	1.02	1.14	0.68	0.76	2.76	3.09
	800	0.82	0.92	0.54	0.60	3.45	3.86
	1200	0.66	0.74	0.44	0.49	3.31	4.82
200	Control	1.42	1.59	0.94	1.05	1.57	1.76
	400	1.14	1.27	0.75	0.84	1.96	2.20
	800	0.91	1.02	0.60	0.67	2.45	2.74
	1200	0.73	0.82	0.48	0.54	3.06	3.43
400	Control	1.58	1.76	1.05	1.17	1.12	1.25
	400	1.26	1.41	0.84	0.94	1.40	1.57
	800	1.01	1.13	0.67	0.75	1.75	1.96
	1200	0.81	0.90	0.54	0.60	2.18	2.45
$LSD_{(0.05)}$		**0.0063**	**0.00782**	**0.0053**	**0.0066**	**0.0158**	**0.01836**

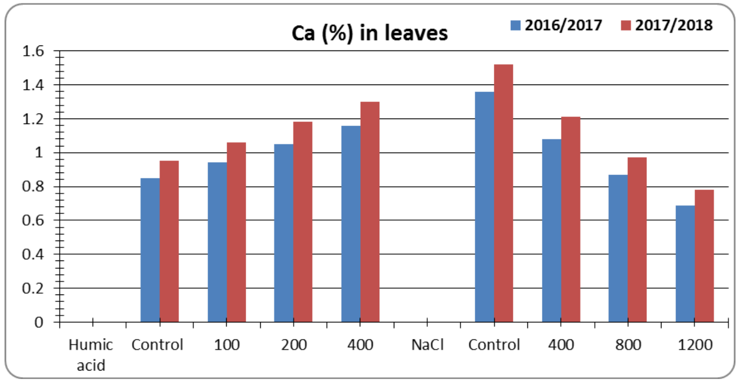

Fig. (19): Effect of humic acid levels and NaCl concentration on Ca (%) in leaves during 2016/2017 and 2017/2018 seasons.

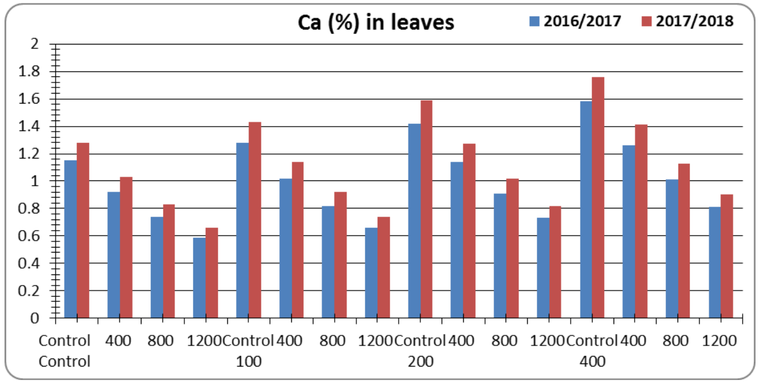

Fig. (20): Interaction between humic acid levels and NaCl concentration on Ca (%) in leaves during 2016/2017 and 2017/2018 seasons.

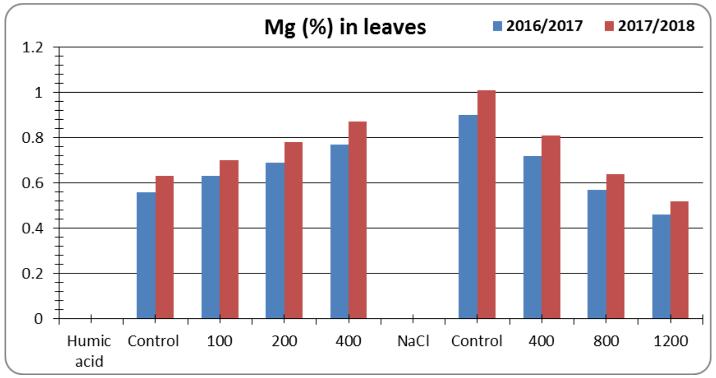

Fig. (21): Effect of humic acid levels and NaCl concentration on Mg (%) in leaves during 2016/2017 and 2017/2018 seasons.

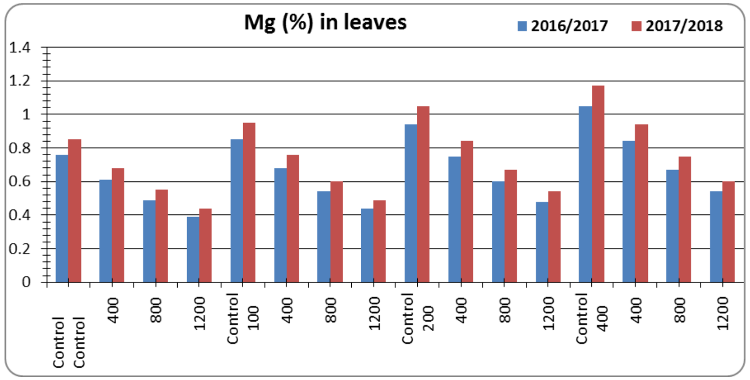

Fig. (22): Interaction between humic acid levels and NaCl concentration on Mg (%) in leaves during 2016/2017 and 2017/2018 seasons.

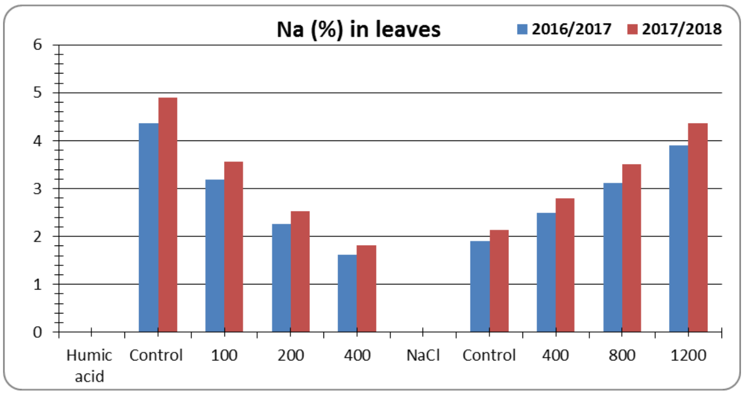

Fig. (23): Effect of humic acid levels and NaCl concentration on Na (%) in leaves during 2016/2017 and 2017/2018 seasons.

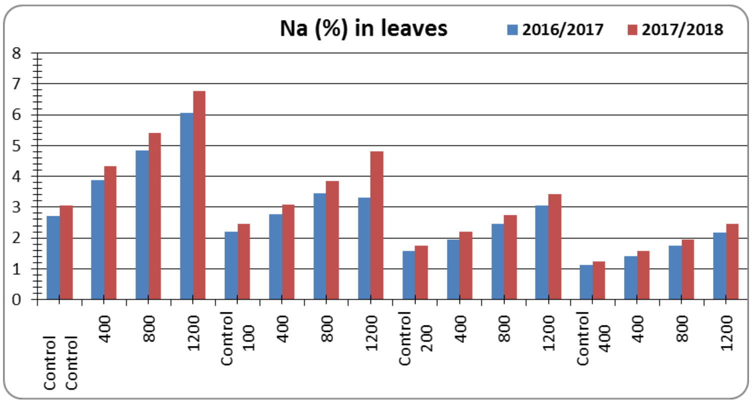

Fig. (24): Interaction between humic acid levels and NaCl concentration on Na (%) in leaves during 2016/2017 and 2017/2018 seasons.

CONCLUSION

Humic substances play a vital role in fertility and plant nutrition. Plants grown on solution which contain adequate humic acid (HAs) are healthier produce higher yields; and the nutritional quality of harvested foods and feeds are superior. Humic acid has contributed a rich source to the growth of plants. Therefore we can conclude that plants of lettuce sown using soilless culture system and received fertilization of humic acid (400 mg/l) gave the best results compared to other treatments. Levels of salinity partially ameliorated the deleterious effects of salinity stress on plant growth and improved cell membrane stability and nutrients uptake of lettuce under salinity stress.

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الملخص العربى

إستجابة الخس لحامض الهيوميك وظروف الإجهاد الملحى تحت نظام الزراعة بدون تربة

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أجريت هذه الدراسة بمزرعة خاصة ببرج العرب، الإسكندرية ، مصر ، خلال موسمي (٢٠١٦/٢٠١٧ و ٢٠١٧/٢٠١٨) لدراسة تأثير حمض الهيوميك على محصول الخس تحت ظروف الإجهاد الملحي باستخدام نظام الاستزراع بدون تربة. . أجريت التجارب في تجربة عاملية تتكون من ستة عشر معاملة مرتبة في تصميم قطاعات كاملة العشوائية (RCBD) بثلاث مكررات لكل معاملة ، حيث أن مستويات حمض الهيوميك (٠ ، ١٠٠ ، ٢٠٠ و ٤٠٠ مجم / لتر) تم ترتيبها في القطع الرئيسية ، في حين تم ترتيب معاملات الإجهاد الملحي (الكنترول ، ٤٠٠ ، ٨٠٠ و ١٢٠٠ ملجم / لتر) في القطع تحت الرئيسية. أشارت النتائج إلى أن المحصول ومكوناته يزداد تدريجياً مع زيادة معدل حامض الهيوميك بمقدار ٤٠٠ ملجم / لتر مثل (قطر الرأس ، الوزن الرطب للرأس ، الوزن الجاف للرأس والمحصول / فدان) ، وكذلك زيادة المحتوي الكيميائي مثل NPK ، النسبة المئوية للكربوهيدرات الكلية ومحتوى فيتامين ج مقارنة بمعاملة الكنترول والتي سجلت نقصًا معنويًا لهذه الصفات في هذه الدراسة في كلا الموسمين. من ناحية أخرى ، أدت جميع معاملات الملوحة إلى انخفاض معنوي في المحصول ومكوناته حيث سجل كلوريد الصوديوم عند ١٢٠٠ ملجم / لتر تأثيرًا سلبيًا على قطر الرأس والوزن الرطب للرأس والوزن الجاف للرأس والمحصول / فدان ، كما تسبب في انخفاض معنوي في المكونات الكيميائية مثل NPK ، النسبة المئوية للكربوهيدرات الكلية ومحتوى فيتامين C في نباتات الخس بالمقارنة مع معاملة الكنترول التي سجلت زيادة معنوية في قطر الرأس والوزن الطازج للرأس والوزن الجاف للرأس والمحصول / فدان والتركيب الكيميائي مثل NPK ونسبة الكربوهيدرات الكلية وفيتامين C المحتوى خلال كلا الموسمين.