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The Influence of Irrigation Water Salinity and Humic Acid on Nutrient Contents of Onion (*Allium cepa* L.)

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1. Introduction

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ABSTRACT

Humic acid (HA) efficiently enhances the uptake of nutrients of plants, especially on saline soil. In this study, some nutrient contents of onion in response to salinity and HA application were investigated, and effects of HA application on salinity resistance was evaluated. Research plots were established as a randomized factorial design with four replications on a lysimeter and each replication included 10 plants. Plants in the lysimeters were irrigated with tap water (control, EC: 0.3 dS m⁻¹) and four different doses of salinized water (EC: 2.0, 4.0, 6.0 and 8.0 dS m⁻¹). The HA (0 and 1.0 g kg⁻¹) was applied to the soil and mixed with the soil before planting. Increasing the

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levels of irrigation salinity decreased contents of K, Ca, N, P, Mg, Fe, Zn, Cu and B in onion bulbs; increased contents of Na, Cl and Mn. However, the highest content of K, Ca, and N in the bulbs were obtained by HA application under different salinity levels. Similarly, the soil application of HA positively was affected the P, Mg, Fe, Zn, B contents of the bulbs. While contents of Na, Mn, and Cu were not affected by soil application, Cl was decreased. The results showed that application of HA could partially reduce the harmful effects of salt, so HA can be used as an alternative method to improve product performance in saline conditions.

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Since the excess uptake of toxic ions occurs in salt impacted soils, reduce in plant growth and yield may observed in most cases (Grattan & Grieve 1999). Ions such as Na, Mg, Ca, Cl, HCO₃, SO₄, and B in high quantity are responsible for soil salinity and plant growth reduction. The effect of physiological drought is one of the essential implications of salinity in plants. Soil productivity is negatively affected by a decrease in nutrient absorption of plants due to high salt concentrations in the soil (Aşık et al 2009; Khaled & Fawy 2011).

Salinity may be originated from irrigation water (Vallejo et al 2003). Limitation in the availability of water with reasonable quality water in most arid and semi-arid places entails the saline water use for agricultural production in developing countries (Bekheet et al 2006; Kiremit & Arslan 2016). Many researches were conducted to examine the effect of different irrigation water salinity on yield, growth and nutrient contents of numerous crops (Semiz et al 2012; Neocleous et al 2014).

Onion ranks third after potato and tomato in terms of crop production areas in the world. There are several problems in onion production, one of which, in the arid and semi-arid regions, is the effect of salinity. Hanci

& Cebeci (2018) reported that onion was moderately sensitive to salinity. Initial yield decrease started at a threshold EC of 1.4 dS m^{-1} , and 50% yield reduction occurred at 4.1 dS m^{-1} (Grattan & Grieve 1999). Seed germination, seedling emergence, and seedling growth of onions were affected by soil property, the salinity of irrigation water, and irrigation method (Miyamoto et al 2010).

Humic substances are the subjects of studies in various areas of agriculture such as soil chemistry, fertility, plant physiology, and these materials can significantly improve plant growth and nutrient uptake (Paksoy et al 2010). According to Akıncı et al (2009), HA is a product contains many elements that develop the soil fertility, increase the availability of nutrient elements by holding them on mineral surfaces, and consequently affect plant growth and yield. Khaled & Fawy (2011) were reported that HA has positive impacts on the chemical, biological and physical properties of soils and nutrient uptake. Commercial products containing HA are mostly used in vegetable production; these products are applied to the soil along with liquid fertilizer (Hartz & Bottoms 2010).

The agricultural area affected by soil salinization necessitates some measures such as determination of the proper salt tolerance crop types or application of some materials to diminish the effect of salinity (Lynch & Lauchli 1988). As shown in some studies, there are positive correlations between HA and soil salinity. In this respect, Liu & Cooper (2002) said that treatments of HA might improve the plant response to the salinity since it may increase the absorption of some nutrients and decrease the uptake of some toxic substances. Kulikova et al (2005) also declared that HA might show anti-stress effects under abiotic stress conditions. Liu & Cooper (2002) also stated that more research should be carried out associated with HA application and its impacts on plant tolerance to salinity. Therefore, the present study was aimed to determine the effects of HA on the contents of some nutrients of onion grown under different salinity levels.

2. Material and Methods

2.1. Experimental site and biological material

The experiment was carried out in a greenhouse at the Mustafakemalpasa Vocational School (the coordinates are 40°02' N, longitude 28°23' E; and the altitude is 22 m above mean sea level), and the Department of Soil Sciences (laboratory), Uludag University, Turkey, during the spring and summer of 2013. *Allium cepa* L. cv. Banko was used in the experiment as the plant material. The onion seeds were obtained from the Unigen Seeds[®] (United Genetics Seeds Company, Inc., Mustafakemalpasa, Bursa, Turkey).

2.2. Treatments

This study was conducted in a completely randomized factorial design with two soil application doses of HA, (0 and 1.0 g kg⁻¹) and five levels of irrigation water salinity (EC) 0.3 (control, tap water), 2.0, 4.0, 6.0 and 8.0 dS m^{-1} . Each application consisted of four replications. There were ten plants in each lysimeter, with one lysimeter in each replicate.

2.3. Agronomic practices

During the first two weeks, all the treatments were irrigated with tap water. Irrigation treatments with saline water were imposed after two weeks from the planting date. Four salinity levels were prepared by adding a mixture of NaCl for watering the seedlings. It was necessary to maintain the quantity of the drainage water at 30-40% of the amount of nutrient solution applied; the excess water was drained through the holes in the base of the lysimeter. Humic acid obtained from liquid Deltahumate derived from leonardite (Delta Chemicals Inc., USA) was added to the soil and mixed with the soil before planting (Asık et al 2009). Seedlings of the onion cultivar were transplanted in metallic lysimeters (10 cm apart in rows of 25 cm in width) after 60 days from sowing in early March. Each lysimeter area was 0.26 m², 0.60 m depth and consisted of 10 onion plants. All of the lysimeters were randomized on benches in an unheated. Air-dried soil samples sieved through a 2 mm sieve and blended for homogeneity. Each lysimeter filled by 185 kg of soil. Some physical and chemical properties of the soil used for the experiments are presented in Table 1. Total of 140 kg ha⁻¹ N (as ammonium sulfate) and 60 kg ha⁻¹ P₂O₅ (as superphosphate) fertilizers were applied to the soil according to recommendations based on soil analyses. The amounts of mineral fertilizers were divided into two equal portions. The first portion was applied

during soil preparation and the second at 60 days after onion planting. The plants were grown in a greenhouse with a day/night average temperature of 21 °C and an average relative humidity of 70%.

2.4. Harvest and analyses

Maturity was generally observed when the crop leaves turned pale green. This was later followed by wilting and leaf fall. Whole plants were harvested in mid-July. The onion bulbs were then harvested carefully by digging them out using a hand shovel. The onion bulbs were rinsed under a tap and deionized water then sliced into pieces.

Property	Value	Method	Reference
Bulk density (g cm ⁻³)	1.44	Core	Rahimi et al (2011)
Texture (sand, silt, clay %)	29.10-48.20-24.70	Hydrometer	Soil (1951)
Organic matter (%)	1.90	Walkley-Black	Nelson & Sommers (1982)
pH (saturation)	7.80	1:2.5 water extract	Richards (1954)
EC (1:2.5 dS m ⁻¹)	0.45	1:2.5 water extract	Richards (1954)
Field capacity (%)	38.30	Pressure plate	Obi (1974)
Total N (%)	0.20	Kjeldahl	Bremmer (1965)
Available P2O5 (mg kg-1)	12.00	Olsen	Olsen et al (1954)
Available K ₂ O (mg kg ⁻¹)	278.00		
Available Ca (mg kg ⁻¹)	4055.00	Flame photometer	Pratt (1965)
Available Mg (mg kg ⁻¹)	578.00	-	
Available Fe (mg kg ⁻¹)	9.00		
Available Mn (mg kg ⁻¹)	8.10	Atomic absorption	Lindson & Normall (1078)
Available Zn (mg kg ⁻¹)	0.810	spectrophotometer	Lindsay & Norvell (1978)
Available Cu (mg kg ⁻¹)	2.17		

Table 1- Some physical and chemical properties of soils used in the study

The fresh samples were dried in a forced-air oven at 65 °C for 48 h. Plant total N (nitrogen) was detected with a Buchi K-437/K-350 digestion/distillation unit according to the Kjeldahl method (Bremmer 1965). Total elements of plant samples were digested with HNO₃ and H₂O₂ (Berghof MWS 2 DAP 60 K microwave oven). The total cations K (potassium), Ca (calcium), P (phosphorus), Na (sodium), Mg (magnesium), Fe (iron), Mn (manganese), Zn (zinc), Cu (copper) and B (boron) were analyzed from extracts using the ICP OES (Perkin Elmer OPTIMA 2100 DV) (Kacar 2014). Cl (chloride) was determined by titration with silver nitrate using a potassium chromate indicator (Chapman & Pratt 1962). The K, Ca, N, P, Na, Cl, Mg and Fe, Mn, Zn, Cu, B concentrations in the dry matter were expressed as percentages and ppm, respectively.

The data were subjected to analyses of variance using statistical programs (IBM SPSS Statistics for Windows). Duncan is multiple range test was used to group the means of irrigation water salinity, HA, and their interactions when the F-test was significant (P<0.05).

3. Results and Discussion

Potassium (K) is a primary plant nutrient, which is needed by the plants in large amount. It is available to the plants in the form of cation (K^+). Potassium is essential for a variety of process photosynthesis, fruit formation, winter hardiness and disease resistance (Behairy et al 2015). In the present study, when the salinity increased, the K concentration in the onion bulbs markedly decreased under saline conditions. Our findings revealed that the interaction effect due to the combined application of salt treatments and HA application on K concentration in the onion bulb was significant as shown in Table 2. When compared with the salt only, the K content in the onion was found to be higher in the application of HA. These results were in agreement with Rauthan & Schnitzer (1981) who reported that the HA treatment raised the uptake of K, N, P, Fe, and Zn thereby improving the nutritional status of the plant.

Both Ca and K play a significant role in plant growth-development and control numerous processes. On the other hand, it is emphasized that increased salt concentrations negatively affect Ca and K intake by plant (Turhan et al 2013). In this study, compared with the control, no significant differences were found in the Ca concentration at low salinity levels (2.0 dS m⁻¹), but increasing the salt concentration of the irrigation water above 2.0 dS m⁻¹ decreased the Ca content of bulbs. However, the effect of HA on the Ca level in the onion bulbs, under saline irrigation water, was significant (Table 2). The Ca content in the onion bulbs was found

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higher in the HA application under the saline water irrigation. In other words, the soil applications of HA may decrease the adverse effects of the increasing salinity in onion plants, and Ca play a significant role in the life of the plants, which is essential in their growth and development. According to the literature, HA stimulates roots, increases both available plant nutrients and nutrient uptake from the soil, and improves the plants' resistance to biotic and abiotic stress factors (Cimrin & Yılmaz 2005).

Tuostmonto	K	Ca	Ν	Р	Na	Cl	Mg	Fe	Mn	Zn	Си	В
Treatments ((%)	(%)	(%)	(%)	(%)	(%)	(%)	$(mg \ kg^{-1})$	$(mg \ kg^{-1})$	(mg kg ⁻¹)) (mg kg ⁻¹)	(mg kg ⁻¹)
NaCl 0HA0	2.81 b ^(x)	1.40 c	2.06 d	0.61	0.29	0.25 f	0.20	41.67	19.08 cd	26.84	7.33	7.50
NaCl ₀ HA ₁ NaCl ₂ HA ₀	2.91 a 2.62 c	1.90 a 1.38 c	2.47 b 2.37 c	0.66 0.62	0.28 0.36	0.25 f 0.29 e	0.24 0.22	41.17 40.83	21.50 bc 25.58 a	27.08 24.87	7.75 7.83	8.92 9.25
NaCl 2HA1 NaCl 4HA0	2.76 b 2.13 e	1.81 b 1.16 ef	2.65 a 1.80 e	0.63 0.43	0.35 0.49	0.27 f 0.33 d	0.23 0.19	46.33 33.00	22.17 b 18.67 d	27.92 20.36	7.83 7.00	9.75 7.17
NaCl 4HA1 NaCl 6HA0	2.48 d 1.79 g	1.26 d 1.14 f	1.69 f 1.37 g	0.48 0.34	0.49 0.53	0.30 e 0.50 b	0.22 0.17	33.67 31.50	23.42 ab 22.75 ab	25.92 14.25	7.33 5.58	10.33 5.83
NaCl 6HA1 NaCl 8HA0	1.86 f 1.16 i	1.22 de 0.75 g	1.31 h 1.19 j	0.36 0.20	0.51 0.67	0.43 c 0.72 a	0.19 0.16	36.58 36.17	22.42 b 23.25 ab	16.17 11.48	5.83 5.50	7.67 5.58
NaCl 8HA1	1.31 h	1.12 f	1.26 i	0.25	0.67	0.73 a	0.18	31.17	23.75 ab	11.5	6.50	7.17
NaCl (dS m ⁻¹)												
0.30 (Control)	2.86 a	1.65 a	2.51 a	0.63 a	0.29 e	0.25 e	0.22 ab	41.42 a	20.29 c	26.96 a	7.54 a	8.21 a
2.0	2.69 b	1.60 a	2.26 b	0.62 a	0.36 d	0.28 d	0.23 a	43.58 a	23.88 a	26.39 a	7.83 a	9.50 a
4.0 6.0	2.30 c 1.83 d	1.21 b 1.18 b	1.74 c 1.34 d	0.46 b 0.35 c	0.49 c 0.52 b	0.32 c 0.47 b	0.21 b 0.18 c	33.33 bc 34. 04 b	21.04 bc 22.58 ab	23.14 b 15.21 c	7.16 ab 5.71 b	8.75 a 6.75 b
8.0	1.23 e	0.93 c	1.22 e	0.22 d	0.67 a	0.72 a	0.17 c	30.67 c	23.50 a	11.49 d	6.00 b	6.38 b
Iumic acid (HA	, g kg ⁻¹)											
0.0 (Control)	2.10 b	1.17 b	1.76 b	0.44 b	0.47	0.42 a	0.19 b	35.43 b	21.87	19.56 b	6.65	7.07 b
1.0 NaCl HA	2.26 a ** **	1.46 a ** **	1.87 a ** **	0.47 a ** **	0.46 ** ns	0.32 b ** **	0.21 a ** **	37.78 a ** *	22.65 * ns	21.72 a ** *	7.05 * ns	8.77 a ** **
$\frac{\text{NaCl} \times \text{HA}}{\text{Small and ca}}$	**	**	**	ns	ns	**	ns	ns	*	ns	ns	ns

Table 2- The effects of salinity and humic acid on some nutrient contents in onion bulb

^(x), small and capital letters indicate significant differences among treatments; * and ** significant at the 5% and 1% of probability level, respectively; ns, non-significant

Onion plants take up a large amount of three essential nutrients, which are the basis of growth and development, such as N, P and K (Gharib et al 2016; Akhter et al 2017). Hussein et al (2015) reported a negative correlation between salinity level and N concentration. In this study, salt levels significantly influenced bulb N content. Increasing the levels of salinity were reduced the N contents. However, the interaction between salinity and N levels was significant, application of HA positively affected the N content of onion bulb. (Table 2). These results are consistent with other researchers reporting that HA increased N uptake (Eyheraguibel et al 2008; Al-Fraihat et al 2018). On the other hand, it is emphasized that increased salt concentrations negatively affect Ca and K intake (Turhan et al 2013).

In the last decade, researchers have attempted to address the problem of salinity and nutrient disorder. It is well accepted that salt-stress leads P deficiency in plants by reducing P uptake (Beltrano et al 2013). In this study, P content was highest at control and 2.0 dS m^{-1} salinity levels. P content of the onion bulbs was significantly decreased by irrigation with saline waters having the concentrations of 4.0, 6.0 and 8.0 dS m^{-1} , compared with the control (Table 2). Our findings revealed that the interaction effect between soil HA and salinity water treatments were found statistically non-significant as to K content of onion bulbs but, the soil applications of humus had a significant effect on the accumulation of P in onion bulbs. Although the applications of saline decreased the P content, the soil application of HA limited the decrease. P content gave the highest values by HA treatment. Mesut et al (2010) reported that HA enhances the uptake of some mineral nutrients by plants. The availability of phosphate and iron increased due to the humic application (Cimrin & Yılmaz 2005).

In our study, bulb Mg content was significantly influenced by NaCl levels. The Mg content did not decrease significantly until a water salinity value of 2.0 dS m⁻¹ was exceeded. Increasing the levels of salinity from 2.0 dS m⁻¹ to 4.0, 6.0, 8.0 dS m⁻¹ significantly were reduced the Mg contents. On the other hand, the soil application of HA positively affected the Mg content of the onion plant, although the effect of the interaction of the NaCl × HA application was insignificant (Table 2). The onions treated with the application of HA rather than the untreated plants observed the higher Mg contents. The findings of this investigation are in close conformity with those of Asık et al (2009).

The results indicated that the Fe, Zn, Cu and B content of onion bulbs were strongly affected by the salt treatments; with an increased salt concentration causing a decrease in the micronutrients (Table 2). However, there were no significant differences in the Fe and Zn between the onion plants that were given the two treatments (control and 2.0 dS m⁻¹). According to the analysis results, bulb Cu and B contents increased up to 4.0 dS m⁻¹, however increasing salt doses decreased these contents significantly. Similarly, Zhu et al (2004) reported that micronutrient deficiencies were very common under plants salt stress. Mazhar et al (2012) reported that, adding different levels of HA could alleviate the harmful effect of salinity. In another study, it was reported that humic substances in saline conditions affect plant some mineral intake and increase salt tolerance (Ouni et al 2014). These results might be because humic substances are improved nutrient uptake and ability to chelate soil nutrients (Mayhew 2004). In this connection, the soil application of HA positively affected the Fe, Zn, B contents of the onion bulb, although the effect of the interaction of the NaCl × HA application was insignificant (Table 2). The higher Fe, Zn and B contents were observed by the onions treated with the use of HA rather than the untreated plants. The results obtained are supported by previous studies, as stated by Salwa (2011) that HA essentially helps the movement of micronutrients from soil to plant. The application of HA affects the uptake of Fe, Zn, and Cu plants positively (Mayhew 2004; Eyheraguibel et al 2008; Al-Fraihat et al 2018). Grown under salt stress, Mn and Cu contents of onions were not significantly affected by supplemental HA. The research finding of Osvalde et al (2012), who indicated that the different humic substances application caused no changes in onion leaf Mn concentration, also supports this outcome.

4. Conclusions

In this study, the results show that different irrigation water salinity levels significantly affected the nutrient contents in onion bulb grown in a loamy soil. Increasing the levels of irrigation salinity decreased contents of K, Ca, N, P, Mg, Fe, Zn, Cu and B in onion bulbs while increased contents of Na and Cl. Humic acid had a significant role on increase some nutrient contents (K, Ca, N, P, Mg, Fe, Zn and B) in onion grown under different salinity levels. It could be concluded that HA has a positive effect on the onion by decreasing the negative aspects of salinity stress condition. Considering the nutrient contents in the onions, the application of 1.0 g kg⁻¹ soil humic acid can be recommended in loam soils of the arid and semi-arid regions where irrigation water is saline until 8 dS m⁻¹.

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