

## Interactive effects of salinity and ozonated water on yield components of cucumber

E. Peykanpour<sup>1</sup>, A.M. Ghehsareh<sup>1</sup>, J. Fallahzade<sup>2</sup>, M. Najarian<sup>1</sup>

<sup>1</sup>Department of Soil Science, Isfahan (Khorasgan) Branch, Islamic Azad University, Isfahan, Iran

<sup>2</sup>Young Researchers and Elite Club, Isfahan (Khorasgan) Branch, Islamic Azad University, Isfahan, Iran

### ABSTRACT

A greenhouse experiment was conducted to investigate the effects of salt stress and ozone on yield and yield components of cucumber. The treatments were defined by a two-factorial design of three irrigation water salinity levels (2, 4 and 6 dS/m) and three ozone concentrations (0, 0.5 and 1 ppm). After germination, cucumber seeds were grown in pots containing soil, coco peat and perlite and fed with Hoagland's solution and were imposed with salt and ozone treatments. The results showed that salinity of irrigation water significantly reduced fruit yield of cucumber owing to a decrease both in fruit number and fruit weight. Also, statistical testing showed significant reductions in vitamin C, fresh and dry weight of shoot and root with increasing water salinity. Ozonated water improved fruit yield by increasing fruit number, but had no significant effect on fruit weight. Application of ozone caused significant increases in fresh and dry weight of shoot and leaf area but caused a significant decrease in the vitamin C. Generally, using ozonated water irrigation can induce positive effect on cucumber under salt stress. In conclusion, the saline water in combination with ozone could be recommended in strategic management to grow cucumber (without impaired effects on crop quantity).

**Keywords:** salinization; irrigated land; *Cucumis sativus*; vegetable; L-ascorbic acid

The progressive salinization of irrigated land with saline water can limit the future of agriculture in many areas of the world (Estañ et al. 2005). The irrigation water salinity in Iran is the major factor limiting plant growths. Soil salinity is a serious constrain to crop production in many areas of Iran where the poor quality of irrigation water is commonly associated with high temperature and reduced rainfall. Because salinity water is among the most limiting factors in agriculture, it is important to study the potential effects of this stress. Soil salinity induces osmotic stress by inducing both the accumulation of toxic levels of Na and Cl ions and the uptake prevention of essential nutrients such as K, Ca and NO<sub>3</sub> (Munns 2002). Also osmotic stress led to the reduction of water uptake capability of roots and consequently inhibition of plant growth and a reduction in the

crop productivity (Netondo et al. 2004). Soil salinity inhibits plant growth, with considerable reductions in plant biomass (Rameeh et al. 2004).

Ozone is unstable in solution (Beltrán 2004). It has been effectively used to control growth of microorganism, food and drinking water treatment (Wang et al. 2006), treatment of fruits and vegetables to increase shelf-life of the products (Guzel-Seydim et al. 2004). Ozone has a potential for enhancing crop yield (Zheng et al. 2007). In a greenhouse experiment Chan et al. (2007) concluded that irrigation by ozonated water for one month increased the leaf area and fresh weight of Pak Choi and Chinese spinach. However, Graham et al. (2011) reported that single applications of aqueous ozone had no effect on leaf area and shoot dry weight. However, pathogen levels were significantly reduced in all treatments with aqueous

doi: 10.17221/170/2016-PSE

ozone. Ohashi-Kaneko et al. (2009) suggested that ozonated water can be used to sterilize different water sources (e.g., drain water, river water, rain water and underground water), in order to control pathogens during the early growth stage of plants. The effects of salt stress (Huang et al. 2009) and ozonated water (Ohashi-Kaneko et al. 2009) on the growth and productivity of vegetables have been studied separately by many authors, but the combined effects of ozone and salt stress on cucumber are not available in the literature. Therefore, the present study aims at investigating combined ozone and salt stress effects on: fruit yield, biomass of shoot and root, leaf area and vitamin C of cucumber.

## MATERIAL AND METHODS

In order to study salt stress and ozonated water effects on cucumber yield and yield components, a greenhouse experiment was conducted in the Khorasgan University in 2014. Average temperatures of day and night were 26°C and 18°C, respectively, in greenhouse. Cucumber seeds (storm) were sown in a box of coco-peat substrate and young plants at four leaf stages were transferred in 10 L pots filled with soil (80%) and leca + perlite (20%). One cucumber plant was cultivated per pot. Modified Hoagland's solution was used for irrigation of the pots. Electrical conductivity of nutrient solution was 1.5 dS/m. pH of this solution was set at 5.6 with nitric acid. The treatments were defined by a two-factorial design of three irrigation water salinity levels (2, 4 and 6 dS/m) and three ozone concentrations (0, 0.5 and 1 ppm). The nine treatment combinations were replicated five times and arranged in a randomized complete block design. The salt solution was prepared by nutrient solution and NaCl and the electrical conductivity of different salinity levels was adjusted by a direct reading conductivity meter. Ozonated water was generated with an electrolytically ozonated wa-

ter generator. The plants were exposed to ozone concentrations via irrigation with nutrient solution. For preparation of 0.5 and 1 ppm O<sub>3</sub>, ozone was added to nutrient solution for 5 s and 10 s, respectively. Some fruit yield indexes including fruit weight, fruit length, fruit number and fruit yield were measured. Also, some growth indices including fresh and dry weight of shoot and root, leaf number, leaf area were determined at the end of growth period. The concentration of vitamin C was measured by applying the method described by Ruck (1963).

**Statistical analysis.** Two-way ANOVA was used to assess the effects of ozonated water and salts stress on all analysed plant properties. Means were compared by the Duncan's test at  $P < 0.05$ . Statistical procedures were carried out using the software package SAS 9.1 for Windows (Cary, USA).

## RESULTS

Soil analysis results are shown in Table 1. The soil belongs to the non-saline soil with a neutral reaction and the amount of lime which is relatively high.

**The ozonated water and salts stress levels effects on fruit traits.** Significant mean squares of the ozonated water were determined for fruit number, fruit yield and vitamin C indicating significant differences of these traits at three ozonated water levels (Table 2). However, the effects of ozonated water on fruit length and fruit weight were not significant (Table 2). Fruit number and fruit yield were increased at high ozonated water levels, but the concentration of vitamin C was decreased. Although, fruit number and fruit yield were increased by about 35% at 1 ppm O<sub>3</sub> as compared with control, the concentration of vitamin C was decreased by 44%. According to Table 2, significant mean squares of the salts stress levels were determined for all fruit traits. A decrease in these traits was noted as the salinity of irrigation water

Table 1. Some chemical and physical properties of experimental soil

EC (dS/m)	pH	N (%)	P (mg/kg)	K (mg/kg)	OC (%)	CEC (cmol <sub>+</sub> /kg)	CaCO <sub>3</sub> (%)	Texture	ρ <sub>b</sub> (g/cm <sup>3</sup> )	Porosity (%)
1.42	8.22	0.08	28	153	1.7	19	42	Sandy loam	1.5	56

EC – electrical conductivity; OC – organic matter; CEC – cation exchange capacity; ρ<sub>b</sub> – bulk density

Table 2. ANOVA model and comparison of cucumber fruit traits for different treatments

Treatment	Fruit length (cm)	Fruit weight (g)	Fruit number	Fruit yield (kg/plant)	Vitamin C (mg/100 g)	
<b>Ozone comparison</b>						
mean values plant properties for each treatment						
Control	13.09	177.33	26.20 <sup>b</sup>	4.71 <sup>b</sup>	10.53 <sup>a</sup>	
0.5 ppm O <sub>3</sub>	13.00	176.75	33.60 <sup>a</sup>	6.04 <sup>a</sup>	7.20 <sup>b</sup>	
1 ppm O <sub>3</sub>	12.91	176.35	35.33 <sup>a</sup>	6.35 <sup>a</sup>	5.95 <sup>c</sup>	
<b>Salt stress comparison (dS/m)</b>						
2	14.10 <sup>a</sup>	195.82 <sup>a</sup>	39.40 <sup>a</sup>	7.71 <sup>a</sup>	8.95 <sup>a</sup>	
4	12.71 <sup>b</sup>	172.44 <sup>b</sup>	31.27 <sup>b</sup>	5.39 <sup>b</sup>	7.68 <sup>b</sup>	
6	12.19 <sup>c</sup>	162.17 <sup>c</sup>	24.47 <sup>c</sup>	4.00 <sup>c</sup>	7.05 <sup>c</sup>	
<b>Statistical analysis</b>						
	<i>df</i>	mean square				
Ozone (O <sub>3</sub> )	2	0.11	3.69	352.96 <sup>**</sup>	11.45 <sup>**</sup>	84.22 <sup>**</sup>
Salt stress (S)	2	14.67 <sup>**</sup>	4460.32 <sup>**</sup>	838.49 <sup>**</sup>	52.82 <sup>**</sup>	14.09 <sup>**</sup>
O <sub>3</sub> × S	4	0.16	1.36	50.19 <sup>**</sup>	1.96 <sup>**</sup>	5.40 <sup>**</sup>
Error	36	1.55	178.07	6.16	0.25	0.44
CV (%)		9.58	7.55	7.82	10.50	8.43

Mean values plant properties for each treatment. In each column, different letters indicate that values are significantly different ( $P < 0.05$ ), according to the Duncan's test. \* $P < 0.05$ ; \*\* $P < 0.01$

increased. Compared with 2 ds/m, fruit length, fruit weight, fruit number, fruit yield and vitamin C were 14, 17, 38, 48 and 21.2% lower at 6 ds/m of salinity level, respectively (Table 2). Although, the interaction of ozonated water × salts stress on fruit length and weight were not significant, there was

a significant ozonated water × salts stress interaction on the fruit number, fruit yield and vitamin C (Table 2). The highest fruit number and yield was obtained at the third level of ozonated water (1 ppm) and 2 ds/m of salinity level (Table 3) and the treatment of control (without ozone) and 6

Table 3. Effects of different treatment on some parameters of cucumber

Treatment		Fruit length (cm)	Fruit weight (g/plant)	Fruit number (per plant)	Fruit yield (kg/plant)	Vitamin C (mg/100 g)
Ozone (O <sub>3</sub> )	salt stress (dS/m)					
	2	14.08	196.73	31.80 <sup>c</sup>	6.25 <sup>c</sup>	12.94 <sup>a</sup>
Control	4	12.82	172.58	25.60 <sup>de</sup>	4.44 <sup>d</sup>	9.86 <sup>b</sup>
	6	12.36	162.70	21.20 <sup>f</sup>	3.44 <sup>e</sup>	8.79 <sup>c</sup>
0.5 ppm	2	14.12	195.58	39.80 <sup>b</sup>	7.78 <sup>b</sup>	7.50 <sup>d</sup>
	4	12.86	172.96	33.20 <sup>c</sup>	5.74 <sup>c</sup>	7.12 <sup>de</sup>
	6	12.00	161.70	27.80 <sup>d</sup>	4.58 <sup>d</sup>	6.97 <sup>de</sup>
1.0 ppm	2	14.10	195.15	46.60 <sup>a</sup>	9.10 <sup>a</sup>	6.42 <sup>ef</sup>
	4	12.44	171.78	35.00 <sup>c</sup>	6.00 <sup>c</sup>	6.05 <sup>fg</sup>
	6	12.20	162.12	24.40 <sup>e</sup>	3.96 <sup>de</sup>	5.38 <sup>g</sup>

In each column, different letters indicate that values are significantly different ( $P < 0.05$ ), according to the Duncan's test

doi: 10.17221/170/2016-PSE

Table 4. ANOVA model and comparison of plant properties for different treatments

Treatment	Shoot		Root		Leaf number	Leaf area (cm <sup>2</sup> )	
	fresh weight	dry weight	fresh weight	dry weight			
(g)							
<b>Ozone comparison</b>	Mean values plant properties for each treatment						
Control	99.74 <sup>c</sup>	38.00 <sup>c</sup>	1.98 <sup>b</sup>	0.99 <sup>a</sup>	32.87 <sup>c</sup>	87.32 <sup>c</sup>	
0.5 ppm O <sub>3</sub>	158.85 <sup>b</sup>	54.57 <sup>b</sup>	3.00 <sup>a</sup>	1.08 <sup>a</sup>	37.47 <sup>b</sup>	131.94 <sup>b</sup>	
1 ppm O <sub>3</sub>	176.82 <sup>a</sup>	70.44 <sup>a</sup>	1.79 <sup>b</sup>	0.66 <sup>b</sup>	38.47 <sup>a</sup>	145.33 <sup>a</sup>	
<b>Salt stress comparison (dS/m)</b>							
2	169.29 <sup>a</sup>	69.77 <sup>a</sup>	3.24 <sup>a</sup>	1.23 <sup>a</sup>	41.33 <sup>a</sup>	139.14 <sup>a</sup>	
4	145.22 <sup>b</sup>	55.83 <sup>b</sup>	2.31 <sup>b</sup>	0.89 <sup>b</sup>	36.87 <sup>b</sup>	132.46 <sup>b</sup>	
6	120.90 <sup>c</sup>	37.41 <sup>c</sup>	1.24 <sup>c</sup>	0.61 <sup>c</sup>	30.60 <sup>c</sup>	93.00 <sup>c</sup>	
<b>Statistical analysis</b>							
Sources of variation	<i>df</i>			mean square			
Ozone (O <sub>3</sub> )	2	24391.8**	3946.3**	6.40**	0.72**	133.8**	13835.2**
Salt stress (S)	2	8783.5**	3952.8**	14.97**	1.46**	436.1**	9331.3**
O <sub>3</sub> × S	4	748.1**	36.21	1.44**	0.12**	46.7*	1814.0**
Error	36	89.89	48.12	0.25	0.03	15.56	4.63
CV (%)		6.53	12.76	22.12	19.05	10.88	1.77

Mean values plant properties for each treatment. In each column, different letters indicate that values are significantly different ( $P < 0.05$ ), according to the Duncan's test. \* $P < 0.05$ ; \*\* $P < 0.01$

ds/m of salinity level had the lowest number and fruit yield. However, the highest concentration of vitamin C was observed at control (without ozone) and 2 ds/m of salinity level (Table 3).

**The ozonated water and salts stress levels effects on growth indices.** According to Table 4, significant ( $P < 0.01$ ) mean squares of the ozonated water were determined for all growth indices. A significant increase in fresh and dry weight of shoot, leaf number and leaf area was noted as the ozonated water levels increased (Table 3). Treatment with 1 ppm O<sub>3</sub> increased fresh and dry weight of shoot, leaf number and leaf area by 77, 85, 17 and 66%, respectively, comparing to control. The salt stress resulted in a significant ( $P < 0.01$ ) decrease in all growth indices. Statistical testing indicated that with higher levels of water salinity the fresh and dry weight of shoot, fresh and dry weight of root, leaf number and leaf area were significantly decreased (Table 4). Compared with 2 ds/m, fresh and dry weight of shoot and fresh and dry weight of root were 29, 46, 62 and 50% lower at 6 ds/m

of salinity level, respectively (Table 4). The treatment with 6 ds/m of salinity level decreased leaf number and leaf area by 26% and 33%, respectively, comparing to 2 ds/m. In most cases, there were a significant interaction between ozonated water and salts stress levels on the growth indices (Table 4). The highest fresh weight of shoot, leaf number and area was obtained at the third level of ozonated water (1 ppm) and 2 ds/m of salinity level and the lowest of these traits was obtained at control (without ozone) and 6 ds/m of salinity level (Table 5). However, fresh and dry weight of root was higher in 0.5 ppm ozonated water and 2 ds/m of salinity level than in the other treatments (Table 5), although in some cases, the differences were marginally significant.

## DISCUSSION

The present study demonstrated that the application of ozone concentration (especially 1 ppm)

Table 5. Effects of different treatment on some parameters of cucumber

Treatment		Shoot		Root		Leaf number (in plant)	Leaf area (cm <sup>2</sup> )
Ozone (O <sub>3</sub> )	salt stress (dS/m)	fresh weight	dry weight	fresh weight	dry weight		
		(g)					
Control	2	139.96 <sup>d</sup>	50.74	2.83 <sup>b</sup>	1.33 <sup>ab</sup>	35.00 <sup>cde</sup>	101.65 <sup>f</sup>
	4	92.06 <sup>e</sup>	39.89	1.76 <sup>c</sup>	0.89 <sup>c</sup>	33.40 <sup>de</sup>	89.06 <sup>h</sup>
	6	67.21 <sup>f</sup>	23.38	1.36 <sup>c</sup>	0.75 <sup>cd</sup>	30.20 <sup>e</sup>	71.26 <sup>i</sup>
0.5 ppm	2	176.55 <sup>b</sup>	70.49	4.17 <sup>a</sup>	1.47 <sup>a</sup>	42.40 <sup>ab</sup>	135.09 <sup>d</sup>
	4	160.41 <sup>c</sup>	54.92	3.56 <sup>a</sup>	1.19 <sup>b</sup>	38.00 <sup>bcd</sup>	146.90 <sup>c</sup>
	6	139.59 <sup>d</sup>	38.30	1.30 <sup>c</sup>	0.58 <sup>de</sup>	32.00 <sup>e</sup>	113.82 <sup>e</sup>
1.0 ppm	2	191.37 <sup>a</sup>	88.09	2.71 <sup>b</sup>	0.90 <sup>c</sup>	46.60 <sup>a</sup>	180.68 <sup>a</sup>
	4	183.20 <sup>ab</sup>	72.68	1.61 <sup>c</sup>	0.59 <sup>de</sup>	39.20 <sup>bc</sup>	161.41 <sup>b</sup>
	6	155.88 <sup>c</sup>	50.55	1.06 <sup>c</sup>	0.49 <sup>e</sup>	29.60 <sup>e</sup>	93.88 <sup>g</sup>

In each column, different letters indicate that values are significantly different ( $P < 0.05$ ), according to the Duncan's test

increased the fruit number, fruit yield, leaf area and fresh, dry weight of shoot. Ohashi-Kaneko et al. (2009) found that flower bud number, flower number, shoot fresh weight, shoot and root dry weight were greater tomatoes treated with ozonated water (1 ppm concentration) than for control. The root respiration by ozonated water might be related to increased biomass productivity of cucumber plants. On the other hand, ozone in water irrigation is well known to autolysis and generates active oxygen species (H<sub>2</sub>O<sub>2</sub>, O<sub>2</sub> and OH) (Tomiyasu et al. 1985). Therefore, the exposure of roots with high level of oxygen is probably associated with the promotion of biomass productivity. According to the study on the tomato reported by Ohashi-Kaneko et al. (2009), ozonated water did not harm plants (tomato) but did promote their growth. Sterilization by ozonated water might have decreased root, consequently increasing water and mineral uptake by plant root (Ohashi-Kaneko et al. 2009). Chan et al. (2007) concluded that irrigation by ozonated water for one month increased the leaf area and fresh weight of Pak Choi and Chinese spinach. However, Graham et al. (2011) reported that single applications of aqueous ozone had no effect on leaf area and shoot dry weight. Bou Jaoudé et al. (2008) reported that a low level of ozone concentration reduced the number of leaves, flowers and pods, leaf area and dry matter of soybean crops. An EC of 1.8 dS/m can be usually harmful to vegetable crops due to salt stress

(De Pascale and Barbieri 1995). Many works found that the yields components of vegetables decreased with increase in irrigation water salinity (Rameeh et al. 2004, Huang et al. 2009). The results showed that with higher levels of water salinity the fruit number, fruit yield, fresh and dry weight of shoot, fresh and dry weight of root, leaf number and leaf area were significantly decreased. These results agree with the findings of Huang et al. (2009) who showed that salinity significantly reduced fruit yield of cucumber owing to a decrease both in mean fruit weight and fruit number. However, salinity caused a reduction in the number of fruits (Beyenne and Hunter 2003, Trajkova et al. 2006).

In conclusion, it is proposed that ozonated water can be used for irrigation of cucumber plant. Moreover, ozonated water does not harm plants. Overall, the use of ozonated water could provide a useful tool to improve fruit yield of cucumber under salinity stress.

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doi: 10.17221/170/2016-PSE

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Received on March 4, 2016

Accepted on June 24, 2016

*Corresponding author:*

Dr. Ahmad Mohammadi Ghehsareh, Islamic Azad University, Department of Soil Science, Isfahan (Khorasgan) Branch, Isfahan; e-mails: amohammadi@khuisf.ac.ir, mghehsareh@yahoo.com