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Effect of different concentrations of diluted seawater on yield and quality of lettuce

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This study was carried out to investigate the effects of irrigating lettuce (*Lactuca sativa* L. cv. Funly) with different concentrations of diluted seawater (0%, 2.5%, 5%, 10%, 15%, 20%) on the fresh yield, marketable yield and quality (DM, total soluble solids, titratable acidity, total sugar, vitamin C, NO_3-N, protein, and total oxalate content). The experiments were conducted in a greenhouse in the autumn of 2012. The fresh yield, marketable yield, and DM of lettuce irrigated with 2.5% and 5% seawater were similar to those of control, but these parameters decreased in response to 10% seawater, and the lowest values were obtained in response to 20% seawater. The 2.5% seawater treatment had no effect on the vitamin C and NO_3-N content, but both significantly decreased when lettuce was irrigated with seawater concentrations higher than 2.5%. Total soluble solids, total sugar, and protein content significantly increased in response to low salinity (2.5% and 5%) but decreased in response to increasing seawater stress. The titratable acidity values remained unchanged under the various saline conditions. Irrigation with diluted seawater did not affect the total oxalate content up to a concentration of 5%, but increasing the concentration of seawater above 5% increased oxalate content. The results of this study demonstrated that low concentrations of seawater are suitable for lettuce production and lettuce can be grown successfully using diluted seawater at concentrations of 2.5% and 5%.

Key words: *Lactuca sativa*, lettuce plant, marketable yield, salinity, seawater stress, sugar.

INTRODUCTION

Soil salinity in arid and semi-arid regions is one of the most important factors limiting agricultural production (Neumann, 1995). Significant reductions in plant growth occur due to salt stress, but these reductions vary according to plant species. The salt tolerance of vegetable varieties is important because monetary return is higher when compared with other field crops (Shannon and Grieve, 1999). Fresh lettuce (*Lactuca sativa* L.) leaves are rich in minerals. Lettuce is categorized as being moderately salt tolerant. Salinity levels higher than 2.0 and 2.6 dS m^{-1} reduce fresh yield and plant growth, respectively (De Pascale and Barbieri, 1995).

Salinity does not always originate within the soil; in some cases, it originates from irrigation water. Agricultural production, especially in arid and semi-arid regions where water requirements cannot be met by rainfall alone, commonly requires irrigation. Surface and ground waters, which can be used for irrigation, contain varying quantities of dissolved salts. In Mediterranean environments, vegetable crops are often cultivated near coastal areas where soil salinization often occurs (Beltrao et al., 2000; Vallejo et al., 2003). The extensive use of well water results in the intrusion of seawater, and water becomes increasingly brackish (Miceli et al., 2003). In many areas, the availability of high-quality water is limited. The use of low-quality water results in an increase in soil salinity (Incrocci et al., 2006), which may have negative effects on growth and yield of crops (Chartzoulakis and Klapaki, 2000).

The yield and quality of vegetable crops are adversely affected by environmental factors such as drought and high salinity in the root zone (Goyal et al., 2003). Plants under salt stress conditions tend to decrease their water uptake and change their nutrient absorption ratios (Miceli et al., 2003; Tzortzakis, 2009). According to Al-Maskri et al. (2010), salinity is one of the main factors limiting plant growth and yield. In lettuce, salt stress negatively affects plant growth and production of DM (EL-Abagy et al., 2012). Increasing salt concentrations in irrigation water may lead to a significant decrease in lettuce growth, yield, marketable yields, weight and amount of DM (Miceli et al., 2003; Mekki and Orabi, 2007; Al-Maskri et al., 2010). However, the results of studies by Andriolo et al. (2005) and Ünlükara et al. (2008) showed that plant DM increased with increasing salinity. Salinity is also able

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to produce improvements in vegetable quality despite lowering growth efficiency (Francois and Maas, 1994), and improvements in quality are particularly important for consumer satisfaction (Colla et al., 2006).

Nitrogen, one of many elements required by all plants for growth, is used by plants in relatively large quantities. Lettuce is one of many vegetables with leaves that accumulate nitrate (Santamaria, 2006). Nitrogen deficiency is a growth limiting factor for plants (Tuncay et al., 2011). In cases of natural salinity, Na is often accompanied by chloride, which competes with nitrate (Kafkafi et al., 1982). However, high chloride content in water may reduce the absorption of nitrates and reduce their accumulation in leaves (Miceli et al., 2003). Oxalic acid is synthesized in conjunction with the reduction of nitrate, and it balances pH of plant cells (Raven, 1985). Oxalic acid is synthesized by a wide range of plants (Rahman and Kawamura, 2011). Masters et al. (2005) reported that plants growing in (high) saline areas accumulated secondary compounds (oxalate, tannins). The oxalate content in halogeton (*Halogeton glomeratus*) was greatly increased by adding NaCl to the nutrient solution (Williams, 1960).

Freshwater resources available for agriculture have been declining in quantity and quality. Therefore, the use of lower quality water for irrigation purposes is inevitable to maintain economically viable crops (Mekki and Orabi, 2007). In recent years, there has been increasing interest among agricultural scientists and planners in the use of seawater (at least diluted) for crop irrigation (Liu et al., 2003). However, the effect of seawater on lettuce growth has not been sufficiently researched. In this study, the potential for using seawater for the cultivation of lettuce was examined, and changes that may occur in yield and quality of lettuce irrigated with different concentrations of seawater were evaluated.

MATERIALS AND METHODS

The experiment was conducted in a greenhouse at the University of Uludag (40°02´ N, 28°23´ E; 22 m a.s.l.), Turkey, during the autumn of 2012. The plant analyses were carried out in a laboratory at the TAT Canned Company Inc., Bursa, Turkey.

Plant material, growth conditions, and salinity treatments

Lactuca sativa L. cv. Funly was used as the plant material. Seedlings were obtained from the Agromar Seedlings Nursery (Karacabey, Bursa, Turkey). Seedlings were transplanted in the beginning of October 2012 to plastic pots. The plastic pots (40 cm depth and 30 cm diameter) were filled with sandy loam soil (71% sand, 19% loam, 10% clay, 0.6% organic matter, and 0.09% total N, pH 7.1, electrical conductivity [EC] 0.4 dS m^{-1}). Phosphorus and K fertilizers were added to the soil prior to planting the seedlings, at a rate of 6.0 and 3.0 g per pot of super phosphate (16% P_2O_5) and potassium sulfate (48% K_2O), respectively. Nitrogen fertilizer was supplied in two equal portions at a rate of 0.6 g per pot in the form of ammonium nitrate (33.5% N) before planting and 30 d after planting as described by Mekki and Orabi (2007). The plants were grown in an unheated greenhouse with a day/night average temperature of 18 °C and an average relative humidity of 75% during the growing season.

Plants were irrigated with tap water for 15 d until mid-October, and then they were irrigated with various levels of diluted seawater. Plants were watered with seawater during the last 40 d of the study. The irrigation water volume was determined by the pot weight (Ünlükara et al., 2008; Cemek et al., 2011). The seawater was obtained from Mudanya, Bursa (40°22´ N, 28°54´ E), Turkey, and diluted with deionized water. In the present study, six treatments were utilized including tap water (control), 2.5%, 5%, 10%, 15%, and 20% seawater. The EC and salt concentration of control, 2.5%, 5%, 10%, 15%, and 20% seawater were 0.3, 1.2, 2.1, 3.7, 5.3, and 6.8 dS m^{-1} and 0.00590%, 0.0560%, 0.0117%, 0.216%, 0.316%, and 0.436%, respectively.

After harvest, soil samples of 250 mL from each pot for various treatments were taken for EC determination, average values (based on dry soil) were determined as 0.6, 1.3, 2.3, 3.5, 5.4, and 7.1 dS m^{-1} for control, 2.5%, 5%, 10%, 15%, and 20% seawater, respectively.

Harvest, yield, and quality parameter analysis

All the plants were harvested 55 d after seedlings were transplanted (40 d after initiation of salinization). The average plant fresh yield (g $plant^{-1}$) was calculated by dividing the total weight of plants by the number of plants. The marketable yield (g $plant^{-1}$) was calculated by weighing edible leaves.

Edible leaves were rinsed in tap water and deionized water and then sliced into pieces. The fresh samples were dried in oven at 70 ºC for 48 h, and dry weight was measured. The content of the total soluble solids in the fresh samples was determined using a refractometer (Abbe-type refractometer, model 60/DR Bellingham and Stanley, Kent, UK). Titratable acidity was estimated using the titrimetric method, and was expressed as a percentage of citric acid. For the analysis of the total sugar content, the Luff-Schoorl method was used (Gormley and Maher, 1990). The vitamin C (ascorbic acid) content in the fresh lettuce samples was determined using the titration method (AOAC, 1980). Nitrates (NO_3-N) in the fresh lettuce leaves were measured by spectrophotometric method, using a Shimadzu UV-1208 spectrophotometer (Shimadzu Co., Kyoto, Japan) at a wavelength of 410 nm (Fresenius et al., 1988). Plant N was determined using the Kjeldhal method, as described by AOAC (1990), and percentage N was converted to crude protein multiplying by 6.25. The total oxalate was analyzed according to Baker (1952), using a method of extraction with hydrochloric acid, precipitation as calcium oxalate from the deproteinized extract and titration with potassium permanganate.

Experimental design and statistical analysis

The experiment was arranged in a completely randomized design with six replicates for each treatment. There was one plant in each pot and four pots in each replicate. Data were subjected to ANOVA using Minitab 14.0 software. The significance of differences among treatments was tested using the least significant difference (LSD) method. The differences were judged significant at $P < 0.05$, according to the F-test. Regression analysis was performed on the relationships between irrigation water salinity and yield or other lettuce quality parameters.

RESULTS AND DISCUSSION

Fresh yield and marketable yield are important economic determining factors. Increases in fresh yield are particularly important for grower satisfaction. Andriolo et al. (2005) reported that a salinity level above 2.0 dS m^{-1} reduced lettuce fresh yield ('Vera'); therefore, for commercial purposes, salinity level should be kept below 2.0 dS m^{-1}. In the present study, lettuce fresh yield and marketable yield were significantly affected by irrigation with different concentrations of diluted seawater. There was a significant quadratic relationship between irrigation water salinity and fresh yield, and a third-degree polynomial relationship existed between irrigation water salinity and marketable yield (Figure 1). Compared with control, no significant differences were found in fresh yield and marketable yield at low seawater levels (2.5%, 1.2 dS m^{-1} and 5%, 2.1 dS m^{-1}). These results were similar to Cemek et al. (2011), who reported that the highest yield was achieved at a low irrigation water salinity and soil salinity, while the lowest yield was obtained at a high irrigation water salinity and soil salinity. With 10% seawater treatment (3.7 dS m^{-1}), there were considerable decreases in fresh yield and marketable yield in comparison to the control, and as the seawater concentration increased above this level, yields decreased further. The lowest fresh yield and marketable yield occurred in response to 20% seawater treatment (6.8 dS m^{-1}, Table 1). According to Al-Maskri et al. (2010), salinity is one of the main factors limiting plant growth and crop yield. Salinity in soil or irrigation water can reduce plant growth, to interfere nutrient balance, and to reduce crop yields (Francois and Maas, 1994; Rouphael et al., 2006; Tzortzakis, 2009). In other studies, an increase in salinity of nutrient solution was associated with a reduction in marketable yield and average plant fresh weight of lettuce ('Ballerina' and 'Severus') (Miceli et al., 2003).

Our results showed that DM decreased due to the exposure to seawater, and it decreased dramatically starting with 10% seawater treatment. Results of the regression analysis demonstrated that there was a quadratic relationship between irrigation water salinity and DM (Figure 1). The dry matter from seawater stressed lettuce plants at 10%, 15%, and 20% treatments decreased significantly in comparison to control. The lowest DM content in lettuce plants was obtained in response to 20% seawater treatment. Similar results were obtained by Miceli et al. (2003), Mekki and Orabi (2007), Al-Maskri et al. (2010), and EL-Abagy et al. (2012). These studies demonstrated significant decreases in lettuce DM as the salt concentrations in the irrigation water increased. However, several studies using other lettuce varieties, such as 'Vera' (Andriolo et al., 2005) and 'Crispa' (Ünlükara et al., 2008), found that increases in salt concentration positively affected DM. This study showed that at the low salinity levels (control, 2.5%, and

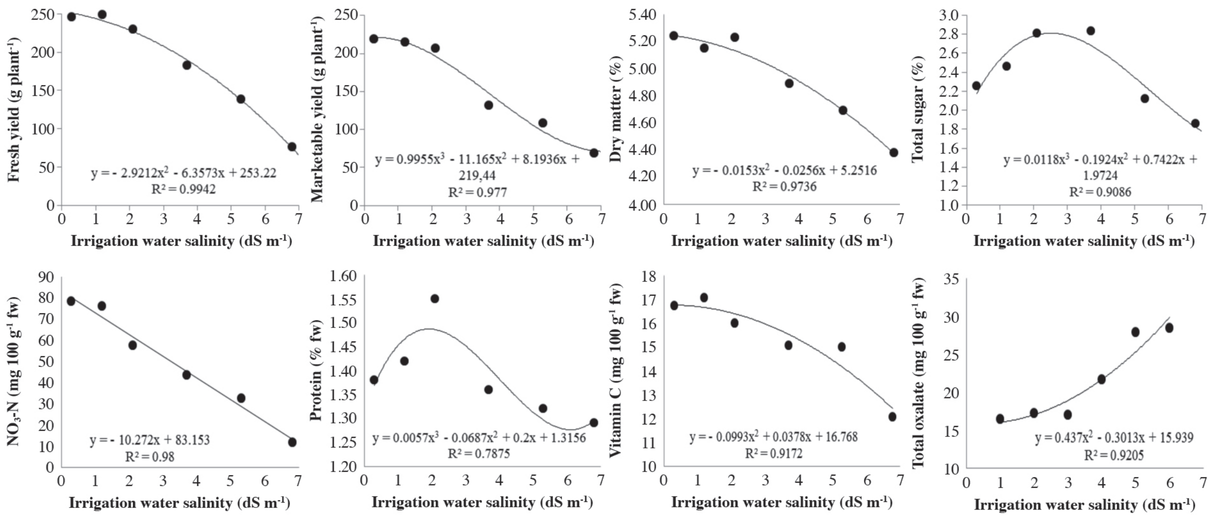

Figure 1. Relationships between irrigation water salinity and yield and some quality parameters of lettuce.

Table 1. Fresh yield and marketable yield, dry matter, total soluble solid content, titratable acidity, total sugar, NO_3-N, protein, vitamin C, and total oxalate of lettuce in response to different seawater treatments.

Treatments	Fresh yield	Marketable yield	Dry matter	Soluble solid	Titratable acidity	Total sugars	Protein	NO_3-N	Vitamin C	Total oxalate
	g plant^{-1}		%			%, fw		mg 100 g^{-1} fw		
Control (0.3 dS m^{-1})	245.59a[1]	218.77a	5.24a	3.48bc	0.065	2.25c	1.38bc	78.39a	16.75ab	16.50c
2.5% seawater (1.2 dS m^{-1})	248.42a	215.03a	5.15a	3.76b	0.065	2.46b	1.42b	76.06a	17.06a	17.31c
5% seawater (2.1 dS m^{-1})	230.28a	206.25a	5.23a	4.13a	0.063	2.81a	1.55a	57.60b	16.01b	17.09c
10% seawater (3.7 dSm^{-1})	182.93b	131.76b	4.89b	3.33c	0.064	2.83a	1.36cd	43.37c	15.09c	21.75b
15% seawater (5.3 dS m^{-1})	138.55c	108.42c	4.69c	3.30c	0.064	2.12d	1.32de	32.59d	15.01c	27.91a
20% seawater (6.8 dS m^{-1})	75.74d	68.94d	4.38d	3.23c	0.067	1.86e	1.29e	11.62e	12.09d	28.51a
LSD at 5%	19.17	14.24	0.20	0.28	ns	0.12	0.05	3.95	0.77	2.92

[1]Means followed by the same letter within each column are not significantly different according to Duncan's multiple range tests ($P < 0.05$).
ns: Non-significant; fw: fresh weight.

5% seawater), differences in DM were not significant (Table 1). Mekki and Orabi (2007) also reported that there were no significant differences in DM contents between moderately saline water and the control irrigation in prickly oil lettuce.

The total soluble solid content of lettuce plants significantly increased with increasing salinity until a certain point; solids were highest at 5% salt concentration (Table 1). A similar effect was reported in tomato (*Lycopersicon esculentum* Mill.) and melon (*Cucumis melo* L.) plants by Shannon and Grieve (2000). Pasternak et al. (1986) reported that application of saline water during specific growth stages may improve quality of many fruits and vegetables by increasing the content of total soluble solids. In this study, significant decreases in soluble solids content began at the 10% seawater treatment, and this content continued to decrease, particularly at the highest level of seawater (20%), when compared to the control (Table 1).

The values for titratable acidity and sugar also play an important role in determining crop quality (Moretti et al., 1998). As shown in Table 1, titratable acidity values remained unchanged in response to increasing saline concentrations. The different seawater treatments also had no effect (positive or negative) on lettuce titratable acidity. However, our results indicated a positive relationship between total sugar content and low seawater concentrations (control, 2.5%, 5%, and 10%) (Figure 1). In previous reports, application of saline water during fruiting of melon and tomato increased sugar content (Shannon and Grieve, 2000). At concentrations of 5-10% seawater, sugar content reached its maximum value. At increased seawater concentrations (15%, 20%) there was a decrease in sugar content, with the lowest in plants stressed with 20% seawater. Results demonstrated a third order polynomial relationship between irrigation water salinity and total sugar content (Figure 1). These results were in agreement with Amuthavalli et al. (2012), who found that treatment with NaCl decreased total sugar content in *Gossypium*. Application of NaCl decreased quality of strawberry (*Fragaria vesca* L.) fruits due to a decreased concentration of sugars, organic acids, and soluble solids (Keutgen and Pawelzik, 2007).

Nitrogen is an important component for many structural characteristics, and N deficiency may be a limiting factor for plant growth (Tuncay et al., 2011). In this study, the highest NO_3-N (nitrate) contents in lettuce plants were found in response to control and 2.5% seawater treatments; NO_3-N concentrations were not significantly different between these two salinity levels (Table 1). At 5% seawater treatment, there was a considerable decrease in NO_3-N concentration in comparison to control treatment, and NO_3-N concentration decreased as seawater stress increased. A negative linear relationship was observed between salinity of irrigation water and NO_3-N content (Figure 1). In comparison with the control, the lowest NO_3-N content was identified in response to 20% seawater treatment. These results were in agreement with Gabr (1999), who also found that NO_3-N concentration significantly decreased as salinity increased. According to Totawat and Saxena (1974), excess salts reduce the total amount of N in plants. Miceli et al. (2003) reported that lettuce plants under salt stress conditions may decrease their water uptake and change the absorption ratio of nutrients. Additionally, Turhan et al. (2013) indicated that NO_3-N concentration significantly decreased in salt-stressed spinach (*Spinacia oleracea* L.) plants.

Salt stress conditions also led to the accumulation of other nitrogenous compounds, such as amino acids, proteins, and polyamines, which are often correlated with salt tolerance (Mansour et al., 2000). Protein contents of lettuce plants were also strongly affected by the various seawater treatments. A third-degree polynomial relationship between irrigation water salinity and protein content was observed (Figure 1). An increased salt concentration (control, 2.5%, 5%) resulted in an increase in the protein content, with the highest protein content found in 5% seawater stressed plants (Table 1). In other studies, Ejaz et al. (2012) in sugarcane (*Saccharum officinarum* L.) and Shahba et al. (2010) in tomato roots found that application of salt stress resulted in an increase in protein content. However, increasing seawater concentrations to 10% or 15% resulted in a sharp decrease in protein content. This reduction was primarily observed at the highest seawater concentration (20%). A similar decrease in protein content as NaCl increased was also reported by Azooz et al. (2004), Dagar et al. (2004), and Gulen et al. (2006).

Vitamin C (ascorbic acid) is a water soluble antioxidant. Several studies have demonstrated that ascorbic acid plays an important role as a plant growth regulator (Garg and Kapoor, 1972; Conklin, 2001). Vitamin C is vital to protect against negative effects of salt stress in tomato plants (Shalata and Neumann, 2001). Data related to vitamin C content of lettuce plants grown under various seawater concentrations is presented in Table 1. There was a quadratic relationship between irrigation water salinity and vitamin C (Figure 1). Vitamin C contents were highest in response to 2.5% seawater concentration and to control treatment. Compared with control, vitamin C concentration of seawater-stressed plants significantly decreased in the treatments higher than 2.5%. The lowest vitamin C concentrations were found in response to 20% seawater treatment. Pitura and Michalojc (2012) found that excessive salt concentrations in crop soils due to increasing N rates significantly reduced vitamin C content in butterhead lettuce and leafy celery; however, the opposite relationship was observed with kale leaves. As described by Tas et al. (2005), leaf ascorbic acid concentrations were not affected by salinity concentrations in lettuce (var. *longifolia*).

Masters et al. (2005) reported that plants growing in saline areas accumulate compounds (oxalate, tannins) that can adversely affect palatability. Oxalic acid in leaves is detrimental to human health because it decreases Ca^{2+} absorption, and it also creates a bitter taste. Therefore, it is desirable to decrease its content by adjusting the cultivation methods (Raven, 1985). In this study, there were no significant differences in oxalate production in the lettuce plants exposed to low seawater stress (2.5% and 5%) compared to the control. However, there was a considerable increase in oxalate production in response to 10% seawater treatment that continued with 15% seawater treatment. The highest total oxalate content in lettuce plants was obtained in response to 20% seawater treatment (Table 1). There was a quadratic relationship between irrigation water salinity and total oxalate content (Figure 1). These results were similar to those of Williams (1960), who demonstrated that oxalate content in halogeton (*Halogeton glomeratus* [M. Bieb.] Ledeb.) greatly increased when NaCl was added to the nutrient solution. Rahman et al. (2008) observed that application of 100 mM NaCl resulted in slightly higher soluble oxalate content in plants compared to those lacking additional NaCl. However, the opposite results was obtained by Carvalho et al. (2009) in purslane (*Portulaca oleracea* L.), in which a sharp decrease in the total oxalic acid content was observed in response to an increase in salt concentration in the purslane plant.

CONCLUSIONS

Results of the current study indicated that low concentrations of seawater in irrigation water showed no significant effects on lettuce yield and quality components. Low amounts of salt are necessary in irrigation water in order to plants reach their optimal yield. In the present study, low concentrations of seawater (2.5% and 5%) in the irrigation water showed no negative effects on fresh yield or quality of lettuce crop. In addition, irrigation with low concentrations of saline water did not lead to an increase in salt concentrations in soil. However, it should be stressed that short-term applications do not fully reflect salt accumulation in soil. Longitudinal studies for the determination of true salt accumulation in soil will provide more accurate results.

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