HortScience 44(1):138–144. 2009.

Hypersensitivity of *Ranunculus asiaticus* to Salinity and Alkaline pH in Irrigation Water in Sand Cultures

Luis A. Valdez-Aguilar[1,2]**, Catherine M. Grieve, and James Poss**
U.S. Department of Agriculture, Agricultural Research Service, U.S. Salinity Laboratory, 450 West Big Springs Road, Riverside, CA 92507

Michael A. Mellano
Mellano & Company, 734 Wilshire Road, Oceanside, CA 92057

Additional index words. flower fields, salinity, sprouting percentage, tuberous root, water quality, water reuse

Abstract. **Ranunculus, grown as a field crop in southern and central coastal California, is highly valued in the cut flower and tuberous root markets. However, concerns regarding the sustainability of ranunculus cultivation have arisen when the plantations are irrigated with waters of marginal quality because the viability of the tuberous roots may be compromised. A study was initiated to evaluate the effect of saline irrigation waters, with and without pH control, on the growth of plants and tuberous roots of ranunculus. Treatments consisted of four irrigation water solutions with increasing concentration of Ca^{2+}, Mg^{2+}, Na^+, SO_4^{2-}, and Cl^- to meet an electrical conductivity (EC) of 2, 3, 4, and 6 $dS\cdot m^{-1}$ and pH 6.4. The 3, 4, and 6 $dS\cdot m^{-1}$ solutions were replicated with uncontrolled pH, which averaged 7.8 over the trial. Ranunculus 'Yellow ASD' and 'Pink CTD' seedlings were transplanted into greenhouse sand tanks and irrigated twice daily with treatment solutions. Shoot dry weight of plants irrigated with 2 $dS\cdot m^{-1}$ solutions was 7.20 g and 6.66 g in 'Yellow ASD' and 'Pink CTD', respectively; however, increasing EC from 2 to 3 $dS\cdot m^{-1}$ induced an 83% and 78% decrease, respectively. Tuberous root fresh weight of control plants, 7.45 g and 8.42 g for 'Yellow ASD' and 'Pink CDT', respectively, was decreased by 82% and 89% when EC was 6 $dS\cdot m^{-1}$. High pH of irrigation water caused an additional decrease in shoot dry weight and tuberous root weight. In control plants, 83% and 76% of tuberous roots of 'Yellow ASD' and 'Pink CTD', respectively, that were transplanted in the following season produced new shoots; however, tuberous roots sprouting percentage from plants irrigated with EC 4 $dS\cdot m^{-1}$ water decreased to 42.9% and 58.3% and to 11.1% and 45.0% with EC 6 $dS\cdot m^{-1}$. The hypersensitivity of ranunculus to salinity was associated with a significant decrease in Ca^{2+} and K^+ tissue concentration. In 'Yellow ASD', Ca^{2+} decreased from 202 $mmol\cdot kg^{-1}$ in control plants to 130 $mmol\cdot kg^{-1}$ in plants irrigated with 3 $dS\cdot m^{-1}$ solutions and pH 6.4. In 'Pink CTD', the decrease was from 198 $mmol\cdot kg^{-1}$ to 166 $mmol\cdot kg^{-1}$. Potassium was similarly affected. Compared with control plants (405 $mmol\cdot kg^{-1}$), shoot Na^+ concentration was increased by 101% in 'Yellow ASD' and by 125% in 'Pink CTD' when irrigated with 6 $dS\cdot m^{-1}$ water. Salt sensitivity of ranunculus, as determined by growth of the flowering stems and viability of the tuberous roots, was increased by irrigation with alkaline waters, which was associated with additional increases in Na^+ and Cl^- tissue concentration and decreased iron accumulation. Hypersensitivity to salinity makes ranunculus crop a poor candidate for water reuse systems; however, further research is warranted to elucidate the possibility of enhancing its tolerance to salinity by supplemental Ca^{2+} and K^+ and acidification of irrigation water.**

Ranunculus, *Ranunculus asiaticus* (L.), is a geophyte with tuberous roots that is adapted to the hot and dry summers of its natural habitat in the Mediterranean region. It is highly valued as bedding or potted plant (Karlsson, 2003), but it is also cultivated as a field-grown cut flower as a result of its attractive inflorescences on sturdy long stems (Karlsson, 2003). Ranunculus can be propagated by seeds or by tuberous roots (Margherita et al., 1996; Meynet, 1993), although tuberous roots are preferred because time to flowering is shorter and the number of flowers per stem is higher than in seed-propagated plants (Meynet, 1993).

Ranunculus is grown outdoors during the fall and winter for spring blooming in Mediterranean climates such as southern and central coastal California. The crop is also cultivated in Europe, South Africa, Israel, and Japan (Meynet, 1993). In Carlsbad, CA, ranunculus flower fields attract over 150,000 visitors in the spring, contributing importantly to the local economy. In addition, the tuberous roots are harvested after shoot senescence, stored during the summer for dormancy release, and marketed for garden plantations or flower beds for the next season.

Most horticultural crops, including flower crops, are glycophytes (Greenway and Munns, 1980) because they have evolved under conditions of low soil salinity. Cut flower growers are reluctant to use poor-quality water for irrigation because floricultural species are believed to be highly sensitive to salinity. However, economically important species such as *Antirrhinum*, *Celosia*, *Chrysanthemum*, *Gypsophyla*, *Helianthus*, *Limonium*, and *Matthiola* have been cultivated successfully with irrigation waters of moderate to high salinity (Carter and Grieve, 2008; Carter et al., 2005a, 2005b; Friedman et al., 2007; Grieve et al., 2006; Shillo et al., 2002).

Water of good quality is limited, especially in the arid and semiarid regions of the world as a result of the competition among agricultural, urban, industrial, environmental, and recreational groups. These users, now faced with less dependable supplies of fresh water, are turning to recycled, degraded waters as a valuable alternative for crop irrigation. Recently, concerns regarding the sustainability of ranunculus cultivation in California have arisen because of the loss of tuberous roots quality when the plantations are irrigated with recycled municipal wastewaters, which may have a high alkalinity and contain salts and other contaminants that limit plant growth. To the best of our knowledge, no information describing the salt tolerance and response to alkalinity in irrigation water of ranunculus is available. The objective of this study was to evaluate the effect of saline irrigation waters with and without pH control on growth, quality, resprouting ability of the tuberous roots, and nutrient concentration of ranunculus plants cultivated in sand cultures.

Materials and Methods

Ranunculus 'Yellow ASD' and 'Pink CTD' (The Flower Fields®, Carlsbad, CA) were grown in greenhouse sand tanks at the U.S. Salinity Laboratory, Riverside, CA. On 17 Nov. 2006, 12 seedlings of each cultivar were transplanted into each of 21 sand tanks. Seedlings were 3 inches tall and had three to four fully developed leaves at transplanting time. The tanks had a volume of 360 L (1.2 m × 0.6 m × 0.5 m deep) and contained washed sand with a bulk density of 1.7 $Mg\cdot m^{-1}$. Plants were irrigated with a base solution prepared with Riverside municipal water and containing 3 mM KNO_3, 0.34 mM KH_2PO_4, 50 μM Fe-DTPA, 23 μM H_3BO_3, 5 μM $MnSO_4$, 0.4 μM $ZnSO_4$, 0.2 μM $CuSO_4$, and 0.1 μM H_2MoO_4 in addition to the ions shown in Table 1 for the 2.0-$dS\cdot m^{-1}$ treat-

---

Received for publication 10 July 2008. Accepted for publication 19 Sept. 2008.

Special thanks to The Flower Fields® and Carltas Company in Carlsbad, CA, for providing plant materials for this study and to Don Layfield for tissue analysis.

[1] Current address: Centro de Investigación en Química Aplicada, Blvd. Enrique Reyna Hermosillo 140, Saltillo, Coah., México 25253.

[2] To whom reprint requests should be addressed; e-mail lavaldez@ciqa.mx.

Table 1. Target ion concentration of irrigation water solutions at varying electrical conductivity (EC).

	Concn (meq·L^{-1})				
EC (dS·m^{-1})	Ca^{2+}	Mg^{2+}	Na^{+}	SO$_4^{2-}$	Cl^{-}
2	4.3	2.6	5.4	6.1	3.2
3	6.1	7.4	13.2	9.6	15.4
4	7.8	12.1	20.9	13.1	27.5
6	11.4	18.7	32.3	20.0	42.0

ment. Irrigation waters were applied twice daily using an autocompensated drip irrigation system. Each sand tank had three irrigation lines with drippers every 20 cm and each dripper delivered 0.6 L·h^{-1}. Each irrigation had a duration of 15 min to saturate the sand, after which the solutions drained to 765-L reservoirs for reuse in the next irrigation.

Treatments, imposed 1 week after transplant, consisted of solutions with added Ca^{2+}, Mg^{2+}, Na^{+}, SO$_4^{2-}$, and Cl^{-} to provide an electrical conductivity (EC) of 2, 3, 4, and 6 dS·m^{-1} (Table 1) plus the base solution mentioned previously. Solutions were prepared to mimic estimates of the ion compositions of water imported from the Colorado River to the San Diego Subregion (California Department of Water Resources, 2004) and on simulations of what these water compositions would be on further concentrations by plant–water extractions and by evaporation (Suarez and Simunek, 1997). Solution pH was maintained at 6.4 by H$_2$SO$_4$ addition. Each treatment was replicated three times and experimental units with 12 plants of each cultivar were distributed randomly in the greenhouse. An auxiliary experiment was conducted concurrently with plants irrigated with EC 3, 4, and 6 dS·m^{-1} waters, in which pH was uncontrolled and averaged 7.8 over the course of the trial. Constant ECs were maintained by replenishing daily water lost by evapotranspiration using Riverside municipal water. For experiment duration, saline waters were analyzed for ion composition twice, 30 and 60 d after experiment initiation, by inductively coupled plasma optical emission spectrometry (Perkin-Elmer Corp., Waltham, MA). Chloride was analyzed on nitric–acetic acid extracts by coulometric–amperometric titration. Salts were replenished as necessary to maintain solution target ion concentrations.

Standard meteorological measurements were recorded hourly in the greenhouse with a Class I agrometeorological station. Daily air temperature ranged from 10.2 to 34.5 °C (average, 22.4 °C), whereas night temperature ranged from 8.9 to 23.2 °C (average, 16.2 °C). Relative humidity ranged from 40.5% to 49.0% (average, 44.7%). Average photosynthetic active radiation (*PAR*) was 320.1 μmol·m^{-2}·s^{-1}, whereas noontime average *PAR* was 690.2 μmol·m^{-2}·s^{-1}.

Stems were harvested from 5 Feb. to 22 Feb. 2007 when the petals of the terminal flower were completely extended. The number of flowering stems per plant, length of the flowering stem, and number of flower buds and leaves per flowering stem were recorded. The diameter of the flowering stem was measured at sand surface. Diameter of the terminal flower was also measured. The flowering stems were washed twice in deionized water, blotted dry, placed in paper bags in an oven at 70 °C for 72 h, weighed, and ground in a Wiley mill to pass a 20-mesh screen (20 holes per inch^2). At experiment termination, leaves from the rosette of each plant were collected, placed in bags, dried, and weighed. Shoot dry weight was calculated by adding the dry weight of the foliage plus the dry weight of the flowering stems produced. Concentration of total sulfur, total phosphorus, Ca^{2+}, Mg^{2+}, Na^{+}, K^{+}, iron, and zinc in shoot tissues was determined on nitric acid digestions as indicated previously as well as Cl^{-}. Tuberous roots were harvested and allowed to lose excessive moisture for 2 weeks in the greenhouse after which fresh weight was measured. Tuberous roots from plants irrigated with saline waters (EC 2, 3, 4, 6 dS·m^{-1} and pH 6.4) were stored at room temperature from July to Nov. 2007 for dormancy release. On 14 Nov. 2007, these tuberous roots were planted in an outdoor sand tank, which was divided into three sections. Seven bulbs, with homogeneous diameter, of each cultivar were planted 5 cm deep in each section and irrigated with a complete nutrient solution (2 dS·m^{-1}). Starting 14 d after transplant, tuberous roots exhibiting the development of new shoots emerging above the sand surface were recorded. Sprouting percentage was calculated daily by dividing the number of tuberous roots with shoot emergence by the total number of tuberous roots transplanted.

Data recorded were analyzed with SAS Version 8.02 (SAS Institute, Inc., 2001) as a one-way analysis of variance (ANOVA) considering each EC plus pH combination as a treatment. Another ANOVA was performed for solutions with EC 3, 4, and 6 dS·m^{-1} to detect differences resulting from pH effect at each EC. Means were separated using Tukey's multiple comparison test at $P < 0.05$.

Results

Plant growth and flower production. Plant growth of both ranunculus cultivars was significantly reduced by increasing salinity. As EC increased from 2 to 3 dS·m^{-1}, shoot dry weight of 'Yellow ASD' was decreased from 7.20 g to 1.20 g; however, additional salt stress had no further effect (Table 2). Ranunculus 'Pink CTD' exhibited similar responses (Table 2). When EC of irrigation water was 3 dS·m^{-1} and pH 7.8 shoot dry weight was further decreased, indicating that alkalinity induced an additional stress on plant growth (Table 2).

Salinity-induced reduction in shoot growth negatively impacted the production of flowering shoots. Ranunculus 'Yellow ASD' had reduced number of flowering stems, from 2.90 to 1.06, and 'Pink CTD' from 3.17 to 0.88 when irrigation water EC increased from 2 to 3 dS·m^{-1} (Table 2). Additional increase in EC did not result in further reduction of the number of flowering shoots. The combined effects of salinity and high pH of irrigation water caused an additional reduction in the production of flowers in 'Yellow ASD' when EC was 3 and

Table 2. Effect of increasing electrical conductivity (EC) and pH in irrigation water on plant growth attributes of ranunculus 'Yellow ASD' and 'Pink CTD'.

	Shoot dry wt (g)			Number of flowering stems			Number of flower buds/ flowering stem			Flowering stem length (cm)			Flower diam (mm)		
	Yellow ASD														
EC (dS·m^{-1})	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8
2	7.20 a[z]	[y]	—	2.90 a		—	2.99 a		—	63.3 a		—	72.0 a		—
3	1.20 b	*	0.64 b	1.06 b	*	0.68 b	1.38 bc	NS	1.17 bc	49.1 b	NS	50.5 b	64.4 ab	NS	63.4 ab
4	1.49 b	*	0.42 b	1.29 b	*	0.49 b	1.74 b	NS	1.28 bc	49.4 b	*	40.4 c	63.5 ab	*	49.1 c
6	1.25 b	*	0.35 b	0.75 b	NS	0.33 b	1.68 b	*	1.00 c	45.1 bc	*	33.7 c	59.0 b	*	42.3 c
	Pink CTD														
EC (dS·m^{-1})	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8
2	6.66 a		—	3.17 a		—	2.75 a		—	64.3 a		—	72.7 a		—
3	1.44 bc	*	0.74 c	0.88 c	NS	0.81 c	2.03 ab	NS	1.22 b	51.4 b	*	45.1 bc	66.3 ab	NS	59.5 b
4	2.09 b	*	0.46 c	1.53 b	*	0.53 c	1.84 ab	*	1.16 b	51.3 b	*	39.9 c	63.1 ab	NS	46.1 c
6	1.00 b	*	0.54 c	0.77 c	NS	0.58 c	1.42 b	NS	1.26 b	47.3 bc	*	41.0 c	64.7 ab	NS	57.8 bc

[z] Means followed by different letter, including both pH treatments, are significantly different at $P < 0.05$ according to Tukey's multiple comparison test.

[y] A pairwise comparison to detect the pH effect at the respective EC was performed separately from analysis of variance; NS, *, nonsignificant and significant at $P < 0.05$ according to Tukey's multiple comparison test.

4 $dS{\cdot}m^{-1}$; however, the response was not consistent in 'Pink CTD' (Table 2).

Table 3. Effect of increasing electrical conductivity (EC) and pH in irrigation water on flowering shoot attributes and tuberous root weight of ranunculus 'Yellow ASD' and 'Pink CTD' grown in sand tanks.

	Flowering stem dry wt (g)			Flowering stem diam (mm)			Number of leaves per flowering stem			Tuberous root fresh wt ($g/plant^{-1}$)		
	Yellow ASD											
EC ($dS{\cdot}m^{-1}$)	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8
2	3.73 a	z,y	—	4.09 a		—	5.44 a		—	7.45 a		—
3	1.82 b	NS	1.13 bcd	2.66 bc	NS	2.43 cd	2.64 b	NS	2.35 b	3.15 b	*	1.32 b
4	1.56 bc	*	0.71 cd	2.94 b	*	2.00 de	2.75 b	NS	2.38 b	2.26 b	NS	1.32 b
6	1.36 bc	*	0.45 d	2.49 bcd	*	1.73 e	3.36 b	NS	2.50 b	1.54 b	*	0.45 b
	Pink CTD											
EC ($dS{\cdot}m^{-1}$)	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8	pH 6.4		pH 7.8
2	3.84 a		—	4.17 a		—	5.18 a		—	8.42 a		—
3	1.89 b	NS	1.20 bc	3.19 b	NS	2.54 cd	3.35 ab	NS	2.34 b	2.43 b	NS	1.85 b
4	2.06 b	*	0.72 c	3.09 bc	*	2.22 d	3.28 ab	NS	2.73 b	2.50 b	*	0.94 b
6	1.57 bc	NS	1.01 bc	2.64 bcd	NS	2.54 cd	2.87 b	NS	2.61 b	2.11 b	*	0.50 b

[z] Means followed by different letter, including both pH treatments, are significantly different at $P < 0.05$ according to Tukey's multiple comparison test.

[y] A pairwise comparison to detect the pH effect at the respective EC was performed separately from analysis of variance; NS, *, nonsignificant and significant at $P < 0.05$ according to Tukey's multiple comparison test.

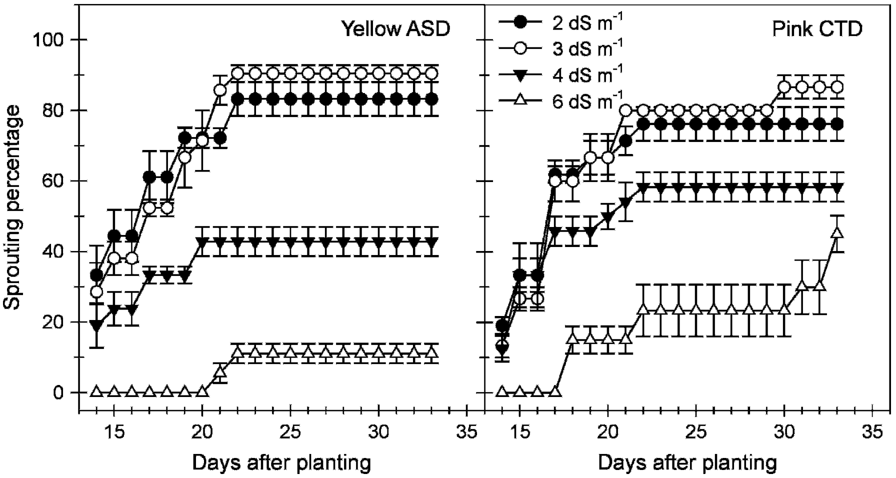

Fig. 1. Cumulative sprouting percentage of tuberous roots of ranunculus 'Yellow ASD' and 'Pink CTD' as affected by electrical conductivity (EC) of irrigation water in the previous season. Bars represent the SE of the mean (n = 3). Mean comparison of data collected at Day 33 indicated a significantly decreased sprouting rate when EC was higher than 4 $dS{\cdot}m^{-1}$ according to Tukey's multiple comparison test at $P < 0.05$.

The number of flower buds per flowering shoot was significantly decreased by high EC. Compared with control plants, 'Yellow ASD' significantly reduced the number of flower buds when plants were irrigated with EC 3 $dS{\cdot}m^{-1}$ water; however, higher EC had no further effect on the number of flower buds (Table 2). Salinity treatment had a lower effect on the number of flower buds produced by 'Pink CTD' compared with 'Yellow ASD' because bud number was significantly decreased only when EC of irrigation water was 6 $dS{\cdot}m^{-1}$ (Table 2). Irrigation with saline water and pH 7.8 did not consistently affect the number of buds produced (Table 2).

Despite the severe reduction in plant growth and in the number of flowering stems and flower buds produced, stem length and flower diameter were less affected by high EC than other quality components. Stem length and flower diameter of both cultivars decreased significantly as EC increased from 2 to 3 $dS{\cdot}m^{-1}$ in solutions with pH 6.4 (Table 2); however, in relative terms, the decrease was much lower, 22% and 11% in 'Yellow ASD' and 20% and 9% in 'Pink CTD', respectively, as compared with the 83% and 78% decrease in shoot dry weight in both cultivars. High pH of irrigation water was detrimental for stem length and flower diameter only when combined with ECs of 4 or 6 $dS{\cdot}m^{-1}$ in 'Yellow ASD', whereas in 'Pink CTD', irrigation with alkaline water caused a significant additional decrease in stem length but no effect on flower diameter (Table 2). Both ranunculus cultivars met the commercial standard stem length requirement (41 cm) for cut flowers when irrigation water had a pH 6.4, even when they were irrigated with 6 $dS{\cdot}m^{-1}$ waters. Other important parameters, however, were significantly reduced when EC was 6 $dS{\cdot}m^{-1}$: dry weight, number of leaves, and diameter of flowering stems (Table 3).

Tuberous root weight. Biomass of tuberous roots was decreased in response to increasing irrigation water EC (Table 3). Tuberous root weight of 'Yellow ASD' and 'Pink CDT' significantly decreased when EC was increased from 2 to 3 $dS{\cdot}m^{-1}$. The combination of high salinity and high pH induced an EC-dependent response. In 'Pink CTD', an alkaline pH combined with EC higher than 4 $dS{\cdot}m^{-1}$ was associated with a significant decrease in tuberous root weight compared with plants irrigated with water with pH 6.4 (Table 3).

Tuberous root sprouting percentage. As determined by the capacity to produce new shoots, quality of tuberous roots collected from plants irrigated with saline waters in the previous season was significantly affected. The emergence of new shoots in 'Yellow ASD' 22 d after planting under nonsaline conditions was of 83%, 90%, 41%, and 11% for the tuberous roots collected from plants irrigated in 2006 with solutions with ECs of 2, 3, 4, and 6 $dS{\cdot}m^{-1}$, respectively (Fig. 1). Over the same range of salinity treatments, emergence of new 'Pink CTD' shoots was 76%, 87%, 58%, and 45%, respectively.

Mineral ion relations in flowering shoots. Calcium concentration in shoots of 'Yellow ASD' and 'Pink CTD' irrigated with water of pH 6.4 was significantly decreased with increasing salinity; however, this response was not significant at pH 7.8 (Fig. 2). Magnesium internal concentration was unaffected by increasing EC in irrigation water of pH 6.4; however, in 'Yellow ASD', shoot Mg^{2+} concentration increased significantly as salinity increased when plants were irrigated with water of pH 7.8 (Fig. 2).

Potassium tissue concentration significantly decreased when EC was 3 $dS{\cdot}m^{-1}$ or higher with no effect of irrigation water pH (Fig. 2). In contrast, Na^+ and Cl^- tissue concentration was significantly increased with increasing EC in irrigation water (Fig. 3); however, the increase was more remarkable in plants irrigated with saline water of pH 7.8. Total sulfur and total phosphorus concentration in both cultivars decreased with increasing salinity in water with pH 6.4 (Figs. 3–4), but this trend was significant only for 'Yellow ASD'.

There was an increasing tendency in phosphorus and sulfur tissue concentration in plants irrigated with saline water and pH 7.8 (Figs. 3–4). This trend was significant for sulfur in 'Yellow ASD' and for phosphorus in 'Pink CTD'. The concentration of phosphorus was not significantly correlated with sulfur concentration (data not shown), but the P/Cl^- and S/Cl^- ratios decreased significantly with increasing salinity for plants irrigated with pH 6.4 water (Table 4).

Tissue iron concentration was not significantly affected by increasing EC; however, there was a pH effect because plants irrigated with saline water of pH 7.8 exhibited a lower concentration (Fig. 4). In addition, in 'Yellow ASD', tissue zinc concentration was significantly reduced by increasing EC in plants

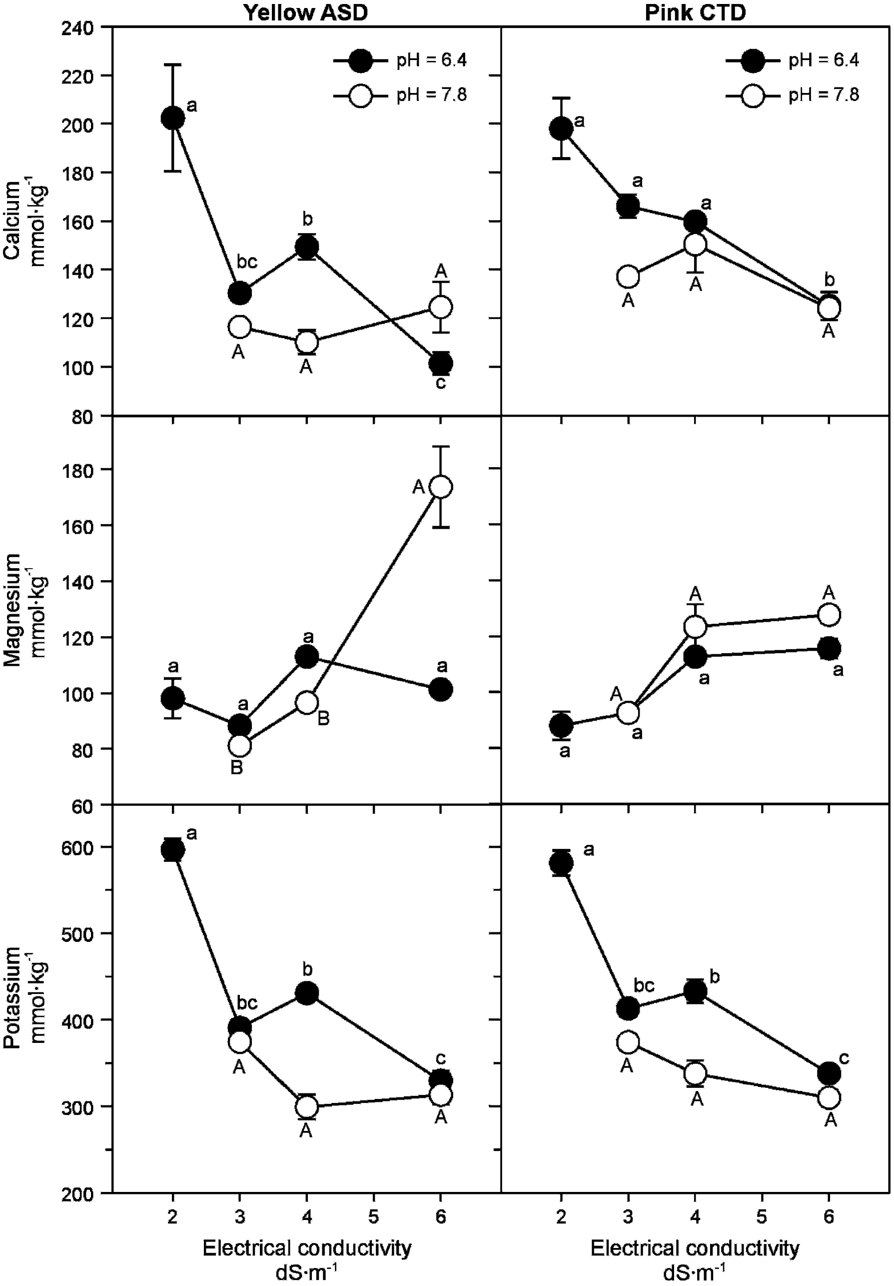

Fig. 2. Effect of increasing electrical conductivity (EC) and pH in irrigation water on Ca^{2+}, Mg^{2+}, and K^{+} concentration in flowering shoots of ranunculus ‘Yellow ASD’ and ‘Pink CTD’ at experiment termination. Bars represent the SE of the mean (n = 3). Means followed by the same letter indicate a nonsignificant difference for plants irrigated with water with pH 6.4 (lower case letter) and pH 7.8 (upper case letters) according to Tukey’s multiple comparison test ($P < 0.05$).

irrigated with water of pH 6.4, but not at pH 7.8 (Fig. 4).

Discussion

The fact that a slight increase in EC of irrigation water (from 2 to 3 dS·m^{-1}) imposed a significant decrease in plant and tuberous root growth suggests that ranunculus is hypersensitive to salinity; however, despite the decreased production of flowering shoots under saline irrigation, flower quality was acceptable. This was because ‘Yellow ASD’ plants irrigated with EC 4 dS·m^{-1} water showed no effect on flower diameter, whereas the stem was within acceptable length. Commercial standard stem length requirement is 41 cm for cut flowers (Barr, 1992). Flower quality in ‘Pink CTD’ was maintained even when plants were irrigated with 6 dS·m^{-1} water. Nonetheless, other important parameters were significantly reduced by high EC: dry weight, number of leaves, and diameter of flowering stems were reduced between 35% and 51% in ‘Yellow ASD’ and between 24% and 51% in ‘Pink CTD’. The combination of salinity with alkaline pH caused additional reduction in plant growth (shoot dry weight) and flower quality (stem length).

Irrigation with saline water affected the viability of ranunculus tuberous roots when EC was higher than 4 dS·m^{-1}. The decreased shoot emergence from tuberous roots was not related to the decrease in fresh weight because there was not a significant difference in sprouting capacity when plants were irrigated with 3 dS·m^{-1} water (although fresh weight was decreased by 58% in ‘Yellow ASD’ and 71% in ‘Pink CTD’). In contrast, despite the comparable fresh weight decrease (≈70% in both cultivars), there was a significant reduction in sprouting capacity on tuberous roots when plants were irrigated with 4 dS·m^{-1} water. Thus, sprouting capacity was affected only when plants were irrigated in the previous season with water with EC higher than 4 dS·m^{-1} regardless of tuberous root fresh weight. This suggests that not only biomass accumulation, but also other factors such as dormancy depth, may be playing a role in the emergence of new shoots. No information has been published as to the effect of salinity on dormancy of bulbs or bulb-like organs; however, bulb dormancy shares the same physiological basis of seed dormancy because both are related to endogenous levels of abscisic acid (ABA) (Kim et al., 1994; Yamazaki et al., 1999). In wheat seeds, studies have demonstrated that increasing NaCl concentration results in increased dormancy (Siddiqui et al., 2006), which may be associated to an increase in ABA in response to salinity (Srisvatava, 2001). In addition, it has been shown that dormancy depth of *Allium wakegi* bulbs is increased by high concentrations of externally applied ABA (Yamazaki et al., 1999). Therefore, because salinity increases ABA internal concentrations and ABA is related to dormancy depth, the decreased sprouting rate of tuberous roots of ranunculus observed in the present experiment may be a result of increased dormancy depth as a result of higher salinity. Further research is necessary to elucidate the effect of salinity on ABA synthesis and dormancy of bulbs and tuberous roots.

Increasing salinity has been reported to induce a decrease in tissue Ca^{2+} concentration (Grieve et al., 2004; Tuna et al., 2007). The decrease reported may range between 5% and 7% in *Celosia argentea* when EC was increased from 2 to 4 dS·m^{-1} (Carter et al., 2005b). However, in the present study, we report a decrease in Ca^{2+} much higher than that and at lower EC; Ca^{2+} concentration was decreased by ≈36% when EC was increased from 2 to 3 dS·m^{-1} in ‘Yellow ASD’ and from 2 to 6 dS·m^{-1} in ‘Pink CTD’. The decrease in Ca^{2+} is attributed to its displacement from the extracellular binding sites induced by Na^{+} (Cramer et al., 1985). A decreased activity of Ca^{2+} resulting from higher ionic strength (Grattan and Grieve, 1999; Hu and Schmidhalter, 2005) and the competition with external ions under high EC (Suarez and Grieve, 1988) also induces decreased Ca^{2+} concentration in plant tissues. The hypersensitivity to relatively low salinity levels observed in the present study may be related to the inability of ranunculus plants to acquire Ca^{2+} from nutrient solution and/or the displacement from plasma membrane even at

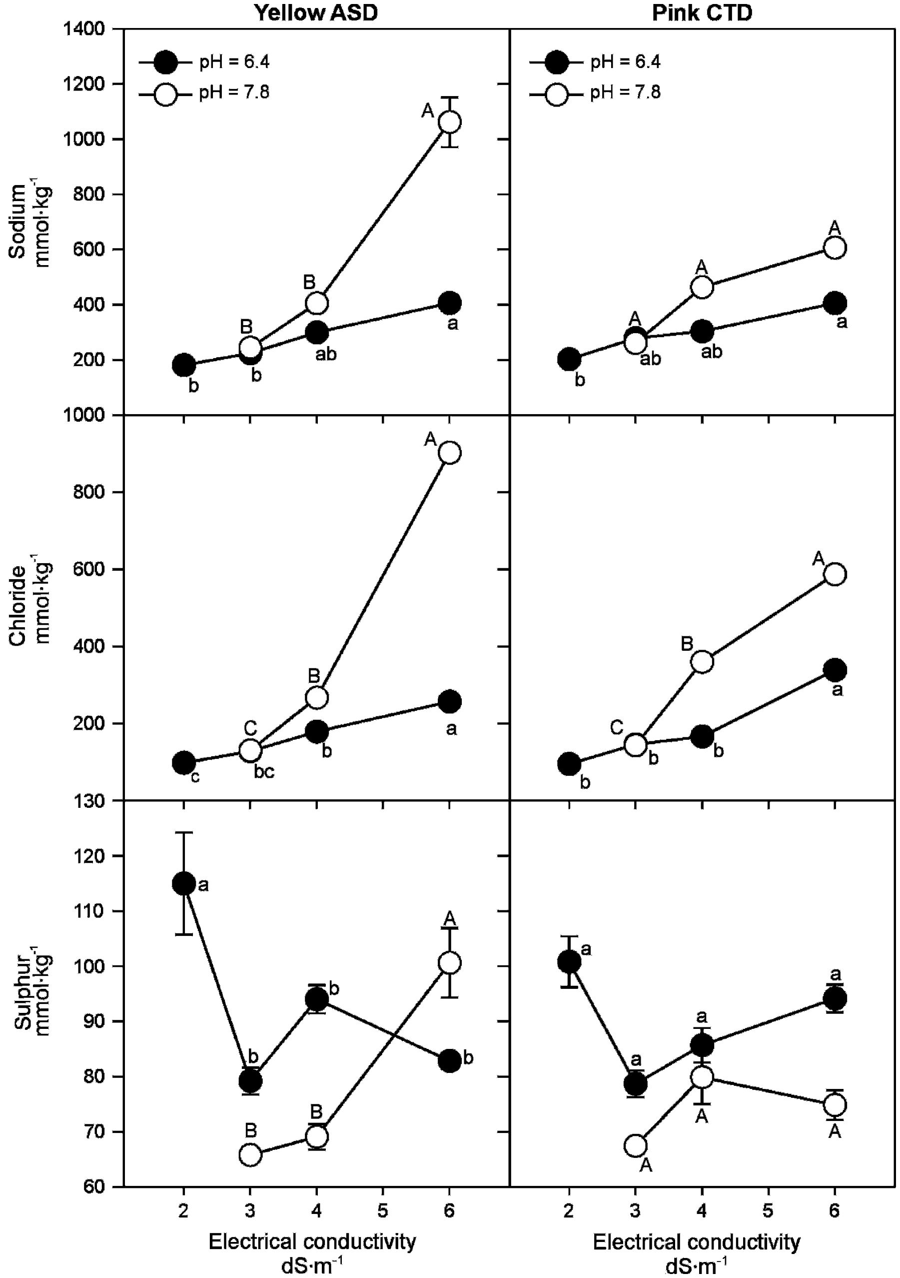

Fig. 3. Effect of increasing electrical conductivity (EC) and pH in irrigation water on Na^+, Cl^-, and total sulfur concentration in flowering shoots of ranunculus ‘Yellow ASD’ and ‘Pink CTD’ at experiment termination. Bars represent the SE of the mean ($n = 3$). Means followed by the same letter indicate a nonsignificant difference for plants irrigated with water with pH 6.4 (lower case letter) and pH 7.8 (upper case letters) according to Tukey’s multiple comparison test ($P < 0.05$).

relatively low EC. Cessna et al. (2007) indicated that Ca^{2+} influx to the cytosol serves as a signaling intermediate for tolerance to salinity; in addition, Ca^{2+} functions as a messenger, mediating responses to hormones (Reddy and Reddy, 2004) such as ABA. Thus, the decreased Ca^{2+} concentration observed in the present study, even at EC as low as 3 dS·m^{-1}, could have affected the adaptive responses of ranunculus to salinity, including responses to ABA.

In plants irrigated with high-salinity water, a decrease in tissue K^+ concentration has been related to a decrease in K^+ ion activity when external Na^+ concentration is high (Grattan and Grieve, 1999). In *Celosia argentea* (Carter et al., 2005a) and *Limonium perezii* (Carter et al., 2005b), K^+ tissue concentration was decreased by 14% to 20% when EC of irrigation water was increased from 2 to 4 dS·m^{-1} and by 30% when EC was increased from 2 to 6 dS·m^{-1}. However, our results indicate that ranunculus was more sensitive to salinity because it exhibited a comparable decreased K^+ uptake at relatively low EC; in ‘Yellow ASD’, K^+ decreased by 35% and in ‘Pink CTD’ by 29% when EC was increased from 2 to 3 dS·m^{-1}. Therefore, the hypersensitivity of ranunculus to salinity may also be explained by the rapid decrease in K^+ uptake. However, the salinity-induced Ca^{2+} deficiency may have a more important role in susceptibility to salinity as suggested by Liu and Zhu (1997), who reported that supplemental external Ca^{2+} suppressed growth defect even at low K^+ in a salt overly sensitive mutant of *Arabidopsis thaliana*.

Magnesium concentration was unaffected by increasing EC in irrigation water at pH 6.4, suggesting a remarkable uptake regulation despite the 3.7-fold increase in external Mg^{2+} concentration. However, at pH 7.8, Mg^{2+} concentration in ‘Yellow ASD’ increased significantly as salinity increased, suggesting that alkaline pH combined with higher external Mg^{2+} ionic strength may affect the regulatory Mg^{2+} uptake mechanism. Islam et al. (1980) reported that shoots of bean plants, and other species, grown at pH 8.5, exhibited increased Mg^{2+} concentration compared with plants grown at pH 5.5. Marschner (1995) recalculated the data from Islam et al. (1980) and observed that the uptake of cations was enhanced at pH 8.5, whereas the uptake of anions decreased, suggesting that this was as a result of a decrease in the efficiency of the H^+ pump at high H^+ concentration in the external medium. In the present study, we detected increasing Mg^{2+} concentration in plants irrigated with water of pH 7.8 only with EC 6 dS·m^{-1}, which may be the result of the combination of the higher pH, as suggested by Marschner (1995), and a higher external Mg^{2+} concentration.

Total phosphorus and total sulfur concentration most likely decreased because of the interaction of phosphates (phosphorus was maintained constant in all treatments) and sulfates with the increasing external concentration of Ca^{2+}, producing insoluble forms of phosphorus and sulfur (Grattan and Grieve, 1999). Another possibility involves interactions whereby Cl^- effectively reduces phosphorus (Papadopoulos and Rendig, 1983) and sulfate uptake (Papadopoulos, 1987; Papadopoulos et al., 1985). This is supported by the significant decrease in P/Cl^- and S/Cl^- ratios observed when EC was increased, suggesting a competition between phosphorus and sulfur with Cl^-.

The decreased growth observed when alkaline pH of saline water was used for irrigation may be associated with an increase in Na^+ and Cl^- concentration and a decrease in iron concentration. Sodium and Cl^- concentration increased with increasing EC in irrigation water; however, the increase was more remarkable in plants irrigated with saline water of pH 7.8, suggesting that plants were under higher Na^+ and Cl^- toxicity stress at higher pH. The higher external Na^+ concentration may have outcompeted K^+ for uptake sites (Marschner, 1995) and displaced Ca^{2+} from root cell membranes (Cramer et al., 1985; Maas and Grieve, 1987), causing the increased Na^+ and decreased K^+ and Ca^{2+} concentration observed in the present study. The fact that increasing salinity had no effect on iron concentration suggests that there may be a concentration effect as a result of the reduced growth (Römheld, 2000), although the decrease at high pH indicates that iron may have been unavailable for uptake as reported under alkaline conditions (Alhendawi et al., 1997).

In conclusion, both cultivars of ranunculus were hypersensitive to irrigation with saline water and high pH induced an additional decrease in plant growth. The marked decrease in plant growth exhibited by plants irrigated with 3 dS·m^{-1} water was associated with a severe reduction in Ca^{2+} and K^{+} concentration in plant tissues and an increase in Na^{+} and Cl^{-} concentration. Further research should explore the possibility of decreasing the sensitivity of ranunculus to salinity through supplemental Ca^{2+} and K^{+} applications. Irrigation with water of alkaline pH caused additional decrease in plant growth, which was associated with an increased shoot concentration of Na^{+} and Cl^{-} and a decrease in iron concentration. The percentage of sprouting tuberous roots was also decreased by increasing EC; however, this was not associated with the decreased biomass when EC was increased from 2 to 3 dS·m^{-1}, suggesting that other physiological responses may play a role in tuberous roots dormancy. If the continued cultivation of ranunculus in California must depend on the use of degraded waters for irrigation, other options must be considered to reduce salinity such as blending recycled waters with high-quality waters and to reduce alkalinity by controlling irrigation water pH by acid injection, addition of organic soil amendments, or the use of acid reaction fertilizers.

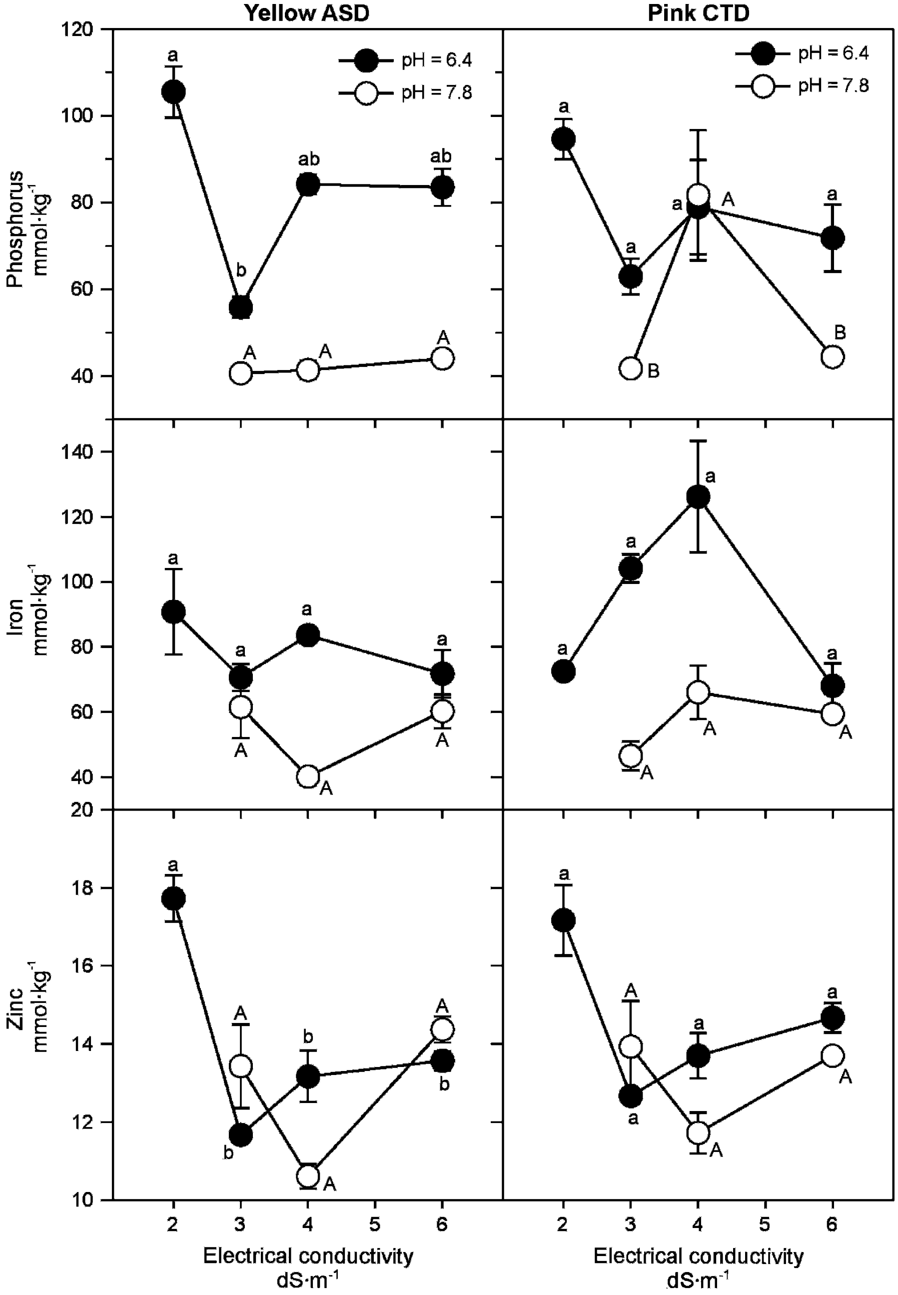

Fig. 4. Effect of increasing electrical conductivity (EC) and pH in irrigation water on total phosphorus, iron, and zinc concentration in flowering shoots of ranunculus ‘Yellow ASD’ and ‘Pink CTD’ at experiment termination. Bars represent the SE of the mean (n = 3). Means followed by the same letter indicate a nonsignificant difference for plants irrigated with water with pH 6.4 (lower case letter) and pH 7.8 (upper case letters) according to Tukey’s multiple comparison test ($P < 0.05$).

Table 4. Effect of increasing electrical conductivity (EC) and pH in irrigation water on the P/Cl^{-} and S/Cl^{-} ratios in flowering shoots of ranunculus ‘Yellow ASD’ and ‘Pink CTD’.

	Yellow ASD				Pink CTD			
	Ratio P/Cl^{-}		Ratio S/Cl^{-}		Ratio P/Cl^{-}		Ratio S/Cl^{-}	
EC (dS·m^{-1})	pH 6.4	pH 7.8	pH 6.4	pH 7.8	pH 6.4	pH 7.8	pH 6.4	pH 7.8
2	1.08 a^{z}	—	1.18 a	—	1.00 a	—	1.07 a	—
3	0.48 b	0.31 a	0.62 b	0.51 a	0.47 b	0.29 a	0.57 b	0.47 a
4	0.44 b	0.16 b	0.53 b	0.26 b	0.46 b	0.29 ab	0.52 b	0.22 b
6	0.32 b	0.05 c	0.33 b	0.11 c	0.21 b	0.09 b	0.28 b	0.12 c
Analysis of variance^{y}	***	**	***	***	***	***	***	***

^{z}Means followed by different letter are significantly different at $P < 0.05$ according to Tukey’s multiple comparison test.

^{y}, **, ***Significant at $P < 0.01$ and $P < 0.001$, respectively.

Literature Cited

Alhendawi, R.A., V. Römheld, E.A. Kirkby, and H. Marschner. 1997. Influence of increasing bicarbonate concentration on plant growth, organic acid accumulation in roots and iron uptake by barley, sorghum, and maize. J. Plant Nutr. 20:1731–1753.

Barr, C. 1992. The kindest cuts of all: How to evaluate new crops. Greenhouse Manager 11:82–84.

California Department of Water Resources. 2004. California’s Groundwater Bul. 118.

Carter, C.T. and C.M. Grieve. 2008. Mineral nutrition, growth, and germination of *Antirrhinum majus* L. (snapdragon) when produced under increasingly saline conditions. HortScience 43:710–718.

Carter, C.T., C.M. Grieve, and J.A. Poss. 2005a. Salinity effects on emergence, survival, and ion accumulation of *Limonium perezii*. J. Plant Nutr. 28:1243–1257.

Carter, C.T., C.M. Grieve, J.A. Poss, and D.L. Suarez. 2005b. Production and ion uptake of *Celosia argentea* irrigated with saline wastewaters. Scientia Hort. 106:381–394.

Cessna, S.G., T.K. Matsumoto, G.N. Lamb, S.J. Rice, and W.W. Hochstedler. 2007. The externally derived portion of the hyperosmotic shock-activated cytosolic calcium pulse mediates adaptation to ionic stress in suspension-cultured tobacco cells. J. Plant Physiol. 164: 815–823.

Cramer, G.R., A. Lauchli, and V.S. Polito. 1985. Displacement of Ca^{2+} by Na^{+} from the plasmalemma of root-cells—A primary response to salt stress. Plant Physiol. 79:207–211.

Friedman, H., N. Bernstein, M. Bruner, I. Rot, Z. Ben-Noon, A. Zuriel, R. Zuriel, S. Finkelstein,

N. Umiel, and A. Hagilandi. 2007. Application of secondary-treated effluents for cultivation of sunflower (*Helianthus annuus* L.) and celosia (*Celosia argentea* L.) as cut flowers. Scientia Hort. 115:62–69.

Grattan, S.R. and C.M. Grieve. 1999. Salinity-mineral nutrient relations in horticultural crops. Scientia Hort. 78:127–157.

Greenway, H. and R. Munns. 1980. Mechanisms of salt tolerance in nonhalophytes. Annu. Rev. Plant Physiol. 31:149–190.

Grieve, C.M., J.A. Poss, and C. Amrhein. 2006. Response of *Matthiola incana* to irrigation with saline wastewaters. HortScience 41:119–123.

Grieve, C.M., J.A. Poss, S.R. Grattan, D.L. Suarez, S.E. Benes, and P.H. Robinson. 2004. Evaluation of salt-tolerant forages for sequential water reuse systems. II. Plant-ion relations. Agr. Water Mgt. 70:121–135.

Hu, Y. and U. Schmidhalter. 2005. Drought and salinity: A comparison of their effects on mineral nutrition of plants. J. Plant Nutr. Soil Sci. 168:541–549.

Islam, A.K.M.S., D.G. Edwards, and C.J. Asher. 1980. pH optima for crop growth. Plant Soil 54:339–357.

Karlsson, M. 2003. Producing ravishing ranunculus. Greenhouse Prod. News January:44–48.

Kim, K.S., E. Davelaar, and G.-J. De Klerk. 1994. Abscisic acid controls dormancy development and bulb formation in lily plantlets regenerated in vitro. Physiol. Plant. 90:59–64.

Liu, J. and J.-K. Zhu. 1997. An *Arabidopsis* mutant that requires increased calcium for potassium nutrition and salt tolerance. Proc. Natl. Acad. Sci. USA 94:14960–14964.

Maas, E.V. and C.M. Grieve. 1987. Sodium-induced calcium deficiency in salt-stressed corn. Plant Cell Environ. 10:559–564.

Margherita, B., C. Giampiero, and D. Pierre. 1996. Field performance of tissue-cultured plants of *Ranunculus asiaticus* L. Scientia Hort. 66:229–239.

Marschner, H. 1995. Mineral nutrition of higher plants. 2nd Ed. Academic Press, San Diego, CA.

Meynet, J. 1993. Ranunculus, p. 603–610. In: De Hertogh, A.A. and M. Le Nard (eds.). The physiology of flower bulbs. Elsevier, Amsterdam, The Netherlands.

Papadopoulos, I. 1987. Effects of residual soil salinity resulting from irrigation with sulphate waters on lettuce. Plant Soil 97:171–177.

Papadopoulos, I. and V.V. Rendig. 1983. Interactive effects of salinity and nitrogen on growth and yield of tomato plants. Plant Soil 73:47–57.

Papadopoulos, I., V.V. Rendig, and F.E. Broadbent. 1985. Growth, nutrition and water uptake of tomato plants divided roots growing in differentially salinised soil. Agron. J. 75:676–700.

Römheld, V. 2000. The chlorosis paradox: Fe inactivation as a secondary event in chlorotic leaves of grapevine. J. Plant Nutr. 23:1629–1643.

Reddy, V. and A.S.N. Reddy. 2004. Proteomics of calcium-signaling components in plants. Phytochemistry 65:1745–1776.

SAS Institute, Inc. 2001. SAS/STAT software changes and enhancements through release 8.0.2. SAS Institute, Cary, NC.

Shillo, R., M. Ding, D. Pasternak, and M. Zaccai. 2002. Cultivation of cut flower and bulb species with saline water. Scientia Hort. 92:41–54.

Siddiqui, Z.S., S.S. Shaukat, and A.U. Zaman. 2006. Alleviation of salinity-induced dormancy by growth regulators in wheat seeds. Turk. J. Bot. 30:321–330.

Srisvatava, L.M. 2001. Plant growth and development. Hormones and environment. 1st Ed. Academic Press, San Diego, CA.

Suarez, D.L. and C.M. Grieve. 1988. Predicting cation ratios in corn from saline solution composition. J. Expt. Bot. 39:605–612.

Suarez, D.L. and J. Simunek. 1997. UNSATCHEM: Unsaturated water and solute transport model with equilibrium and kinetic chemistry. Soil Sci. Soc. Amer. J. 61:1633–1646.

Tuna, A.L., C. Kaya, M. Ashraf, H. Altunlu, I. Yokas, and B. Yagmur. 2007. The effects of calcium sulphate on growth, membrane stability and nutrient uptake of tomato plants under salt stress. Environ. Expt. Bot. 59:173–178.

Yamazaki, H., T. Nishijima, Y. Yamato, M. Koshioka, and H. Miura. 1999. Involvement of abscisic acid (ABA) in bulb dormancy of *Allium wakegi* Araki I. endogenous levels of ABA in relation to bulb dormancy and effects of exogenous ABA and fluridone. Plant Growth Regulat. 29:189–194.