Effects of Irrigation Water Salinity on the Growth, Gas Exchange Parameters, and Ion Concentration of Hot Pepper Plants Modified by Leaching Fractions

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Abstract. In this experiment, the responses of plant growth, gas exchange parameters, and ion concentration to different levels of irrigation water salinity (EC_{iw} of 0.9, 1.6, 2.7, 4.7 and $7.0 \text{ dS}\cdot\text{m}^{-1}$) and leaching fractions (LFs of 0.17, 0.29) were investigated in hot pepper plants. The pot experiment was conducted using a completely randomized block design with four replications in a rain shelter. Results showed that the height of the hot pepper plants decreased as the EC_{iw} was increased from 25 d after transplanting (DAT) and increased when the LF was increased from 55 DAT. Neither the EC_{iw} nor the LF influenced the root length. An increase in the EC_{iw} caused the suppression of the stem diameter (SD); leaf length; leaf area; leaf chlorophyll content (CCI); dry biomass of roots, stems, and leaves; net photosynthesis (P_n); stomatal conductance (g_s); transpiration rate (T_r); and intercellular CO_2 concentration (C_i). An increase in the LF caused the SD, leaf length, leaf area, and dry biomass of stems and leaves to increase. However, the dry biomass of roots and the P_n , g_s , T_r , and C_i were not significantly affected by the LF, except for the C_i measured on 23 DAT and the T_r on 76 DAT. The Na^+ concentrations in the roots and stems increased, whereas the K^+/Na^+ ratios decreased as the EC_{iw} increased. An increase in the LF led to a decrease in the Na^+ concentration of the roots and stems, whereas there was an increase in the K^+ concentration in the stems and the K^+/Na^+ ratios in the roots and stems. Collectively, an increase in the EC_{iw} had an adverse effect on plant growth and gas exchange and led to the accumulation of the Na^+ concentration in the roots and stems, whereas an increase in the LF enhanced plant growth, leaf transpiration, and K^+ concentration and reduced the accumulation of the Na^+ concentration in the roots and stems. We suggest that higher quantity of water should be applied in higher saline irrigation for satisfactory performance for hot pepper growth.

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In many plants, salinity has been found to set a number of biochemical and physiological mechanisms in motion, leading to a corresponding reduction in plant growth (Hatamnia et al., 2013; Hossain and Dietz, 2016). For instance, salinity restricts water uptake by roots, which quickly inhibits the growth rate, along with a suite of changes in metabolic activity of photosynthesis and biosynthesis of photosynthesis pigments (Munns, 2002). Salinity can induce the absorption of excessive mineral nutrition, causing an ionic imbalance as a result of the accumulation of particular salts within the plant, and the consequence restriction of g_s , photosynthesis, and C_i in plants (Hannachi and Van Labeke, 2018; Munns, 2002). Salinity stress is also revealed as oxidative stress and has a number of deleterious effects (Azuma et al., 2010). The analysis of minerals in the plant tissues

and the relationship between the mineral concentrations in the leaves and osmotic stresses are also often examined to understand the mechanism of salt tolerance (Munns and Tester, 2008). The mechanisms used to defend a variety of plant species against salt stress may be dissimilar (Greenway and Munns, 1980), with environmental conditions and cultural practices potentially also influencing these mechanisms.

Hot peppers, one of the most popular vegetables used in sauces and condiments, as well as an important source of vitamin C in the human diet, are particularly susceptible to salinity stress (Aktas et al., 2006; Lycoskoufis et al., 2005). Lycoskoufis et al.'s aforementioned study shows that both P_n and g_s are reduced by a high level of salinity. Salt-stressed peppers also show a severe reduction in growth, water uptake, and ionic balance (Cabañero et al., 2004; Hannachi and Van Labeke, 2018; Navarro et al., 2003; Yang and Guo, 2018). Nevertheless, the effects that salt stress have on plants are still unclear because of the complex nature of salt stress in plants (Aktas et al., 2006).

Irrigation with saline water leads to a successive accumulation of salts in the soil. One of the methods adopted used to reduce salt concentration in the soil involves the application of extra water to compensate for the leaching of salts from the root zone (Aktas et al., 2006). A suitable LF can maintain favorable root-zone salinity under varying levels of irrigation water salinity (Dudley et al., 2008), and consequently enhance plant growth. An improved understanding of the mechanisms enabling hot pepper plants to adapt to salt stress and maintain growth under a suitable LF is incredibly important to ensure the successful production of hot peppers. In addition, to better understand the mechanisms by which LFs affect growth under varying EC_{iw} , the K^+ , Na^+ , and Ca^{2+} concentrations and K^+/Na^+ ratio in hot pepper roots and stems were also studied.

The aim of this study is to evaluate the effects of EC_{iw} on growth (i.e., plant height; SD; leaf length; leaf area; and the dry weight of roots, stems, and leaves), gas exchange (i.e., P_n , g_s , T_r , and C_i), and the uptake of ions (i.e., the Na^+ , K^+ , and Ca^{2+} concentrations and K^+/Na^+ ratio) by hot pepper plants and to test whether LFs are able to modify the effects of the EC_{iw} with respect to the aforementioned parameters.

Materials and Methods

Experimental setup. The experiment was conducted in a rain shelter at the Agrometeorology Research Station at Nanjing University of Information Science and Technology in Nanjing City, Jiangsu Province in China (lat. 32.2° N , long. 118.7° E , altitude 14.4 m) from 28 Apr. to 22 July 2015. The white plastic pots used were 27 cm high, with the top and bottom diameters of the pots measuring 26 and 22 cm , respectively. A 2-cm hole was drilled in the bottom of each pot to collect the drainage water. Each pot

was filled with 11 kg of air-dried soil with a sandy loam texture consisting of sand (75.7%), silt (20.4%), and clay (3.9%) sifted through a 5-mm sieve. The bulk density of soil is $1.47 \text{ g}\cdot\text{cm}^{-3}$. The field water capacity and wilting point are 0.27 and $0.04 \text{ (cm}^3\cdot\text{cm}^{-3}\text{)}$, respectively. The available N, P, and K contents of the soil were 28.0, 16.3, and $47.7 \text{ mg}\cdot\text{kg}^{-1}$, respectively.

The hot pepper plants (Bocuiwang cultivar) were transferred to plastic pots with one plant per pot on 28 Apr. 2015. Each pot was saturated with tap water before the transplanting took place. All the plants were then irrigated using tap water at 5 DAT, with 0.9 L for each pot. After the plants were established (10 DAT), saline water treatments with two LF_s were initiated.

Five levels of saline irrigation water (EC_{iw} of 0.9, 1.6, 2.7, 4.7, and $7.0 \text{ dS}\cdot\text{m}^{-1}$) and two LF_s of 0.17 and 0.29 were used and each treatment was repeated four times. The 40 pots were subjected to a completely randomized block design. A half strength Hoagland solution was used as a nonsaline treatment (in $\text{mmol}\cdot\text{L}^{-1}$: $2.0 \text{ Ca}(\text{NO}_3)_2 \times 4\text{H}_2\text{O}$, 2.0 KNO_3 , $0.5 \text{ NH}_4\text{NO}_3$, $0.5 \text{ MgSO}_4 \times 7\text{H}_2\text{O}$, $0.25 \text{ KH}_2\text{PO}_4$ and in $\mu\text{mol}\cdot\text{L}^{-1}$: 40 Fe-EDTA , $25 \text{ H}_3\text{BO}_3$, $2.0 \text{ MnCl}_2 \times 4\text{H}_2\text{O}$, $2.0 \text{ ZnSO}_4 \times 7\text{H}_2\text{O}$, $0.5 \text{ CuSO}_4 \times 5\text{H}_2\text{O}$, 50 KCl , $0.075 \text{ (NH}_4\text{)}_6\text{Mo}_7\text{O}_{24} \times 4\text{H}_2\text{O}$, $0.15 \text{ CoCl}_2 \times 6\text{H}_2\text{O}$) (Heeg et al., 2008), which added an EC of $0.9 \text{ dS}\cdot\text{m}^{-1}$ to the irrigation water used in all the treatments. To achieve the desired treatment salinity levels, 1:1 milli equivalent concentrations of NaCl and CaCl_2 were added to the half-strength Hoagland solution.

The amount of applied irrigation water was 120% and 140% of the evapotranspiration (ET) for the LF of 0.17 and 0.29, respectively, according to the equation proposed by Letey et al. (2011). Evapotranspiration (g) was calculated as follows:

$$\text{ET} = W_n - W_{n+1} + (I - D) \times \rho, \quad (1)$$

where W_n and W_{n+1} are the pot weights before the n^{th} and $(n + 1)^{\text{th}}$ irrigation (g). D and I are the amounts of applied drainage and irrigation water (L), respectively, and ρ is the water bulk density ($1000 \text{ g}\cdot\text{L}^{-1}$).

Right before each irrigation event, all pots were weighed and the crop ET and the amount of irrigation water for each pot were calculated. A glass bottle was set underneath each pot to collect the leachates. The volume of leachate for each pot was measured using graduated cylinder after each irrigation event. The plants were irrigated every 2–5 d.

Plant measurements. Plant heights were obtained using a tape measure every 7–17 d. The diameters of the plant stems were measured with a digital vernier caliper (PD 151; Prokit's Industries Co., China) at the end of the experiment. All the leaf lengths and the maximum leaf widths of each plant were also measured at the end of the experiment. The leaf area was then estimated by summing the length $\times$ maximum width of each leaf and multiplying by a factor of 0.54 (our measurement). The roots of each plant were washed in fresh water. The maximum length of root

was measured by a ruler. Plant materials, i.e., the roots, leaves, and stems, were then dried in an oven at 70°C to obtain the constant dry weight.

The CCI and gas exchange of the leaves. The CCI of the leaves was obtained using an SPAD 502 chlorophyll meter (Minolta, Tokyo, Japan) on 17, 26, 69, and 84 DAT. The SPAD 502 calculates an index in SPAD units (dimensionless), which is an objective measurement for leaf greenness (Qiu et al., 2015). Readings were taken from five leaves in the upper layers of each plant and the average CCIs were used for each pot.

The gas exchange parameters of the leaves including the P_n rate, leaf g_s , T_r , and C_i were obtained using an LI-6400 photosynthesis system (LI-COR, Lincoln, NE). Measurements of three–five fully grown leaves per treatment were made at 9:00–11:00 AM on 23, 39, and 76 DAT (sunny days), with a fixed PPFD level of $1200 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$.

The Na^+ , K^+ , and Ca^{2+} concentrations of hot pepper roots and stems. The dried roots and stems were ground into powder. The ground plant tissue samples were broken down with concentrated HNO_3 , heated using a heating block, and finally dissolved in 5% (v/v) high-purity HNO_3 . The Ca^{2+} , Na^+ , and K^+ concentrations at the end of experiment were measured using Inductively Coupled Plasma–Optical Emission Spectrometry (ICP-OES, Perkin Elmer Optima 8000). Procedural standard reference materials, sample replications, and blanks were randomly inserted for quality control.

Statistical analysis. Two-way variance analyses were made using the general linear model-univariate procedure with an SPSS (Version 21; IBM Corp., Armonk, NY) to determine the effects of the EC_{iw} and LF on plant height, SD, leaf length, leaf area, the dry weight of roots, stems, and leaves, the P_n , g_s , T_r , C_i and the Na^+ , K^+ , and Ca^{2+} concentrations of the hot pepper roots and stems. Any significant difference among the treatment means was compared using the Duncan's multiple range tests at significant level of $P < 0.05$.

Results

Effects of the EC_{iw} and LF on plant growth

Plant height and SD. The effects of the EC_{iw} and LF on the plant height were analyzed and compared statistically within treatments (Table 1). The plant height increased rapidly until 37 DAT, increasing slowly after that because of reproductive growth. The plant height started to decrease significantly ($P < 0.05$) at 25 DAT with the increasing EC_{iw} , whereas it started to increase significantly ($P < 0.05$) at 55 DAT with an increase in the LF. The plant height in the EC_{iw} of $7 \text{ dS}\cdot\text{m}^{-1}$ diminished by 20.1% as compared with the EC_{iw} of $0.9 \text{ dS}\cdot\text{m}^{-1}$ across the LF at the end of the experiment. There were no interactive effects between the EC_{iw} and LF in terms of plant height throughout the experimental period. There was a significantly negative linear correlation between

the plant height and the EC_{iw} for both LF_s, except for 44 DAT in the LF of 0.17 and 13 DAT in the LF of 0.29.

The SD measured at the end of the experiment under various levels of EC_{iw} and LF is shown in Table 2. An increase in the EC_{iw} significantly ($P < 0.001$) decreased the SD. The application of the EC_{iw} of $7.0 \text{ dS}\cdot\text{m}^{-1}$ caused the SDs to be significantly ($P < 0.001$) reduced by 16.4% as compared with the EC_{iw} of $0.9 \text{ dS}\cdot\text{m}^{-1}$ for both LF_s. High LF significantly ($P < 0.01$) increased the SD by 6.6% across the EC_{iw} . There was no significant interaction between the EC_{iw} and the LF in terms of the SD. There was, however, a significant ($P < 0.01$) negative linear relationship between the SD and the EC_{iw} , regardless of the LF ($\text{SD} = -0.31 \text{ EC}_{\text{iw}} + 11.97$, $R^2 = 0.73$, $n = 10$).

Leaf length and leaf area. The leaf length and leaf area declined significantly ($P < 0.05$) as the EC_{iw} was increased (Table 2). Across the EC_{iw} treatments, the leaf length and leaf area in the LF of 0.29 were significantly ($P < 0.05$) higher than those in the LF of 0.17. There was no significant interactive effect between the EC_{iw} and the LF in terms of leaf length and leaf area. However, a significant negative linear relationship between the leaf area and the EC_{iw} was found for both LF_s (for LF of 0.17, $Y = -0.345 \text{ EC}_{\text{iw}} + 0.333$, $R^2 = 0.91$, $P < 0.05$ and for LF of 0.29, $Y = -0.364 \text{ EC}_{\text{iw}} + 0.386$, $R^2 = 0.99$, $P < 0.001$). Neither was there any notable difference between the slopes of the regression functions, whereas the intercept in the LF of 0.29 was significantly higher than that in the LF of 0.17.

Root length and the dry weight of roots, stems, and leaves. Neither the EC_{iw} nor the LF had any significant effect on the root length of the hot peppers. An increase in the EC_{iw} apparently led to a significant decrease in the dry weight of roots, stems, and leaves (Table 2), especially when the EC_{iw} was high. For both the LF_s, the dry weight of roots, stems, and leaves in the EC_{iw} of $7.0 \text{ dS}\cdot\text{m}^{-1}$ decreased by 54.4%, 55.4%, and 51.3%, respectively, when compared with the EC_{iw} of $0.9 \text{ dS}\cdot\text{m}^{-1}$. The LF had no effect on the dry weight of roots. The dry weight of stems and leaves did increase because of the increase in the LF, however. Across the EC_{iw} , the dry weight of the stems and leaves in the LF of 0.29 increased by 10.2% and 11.5%, respectively, as compared with the LF of 0.17. There was no significant interactive effect between the EC_{iw} and the LF with regard to the dry weight of roots, stems, and leaves. A significant ($P < 0.001$) negative linear correlation was observed in the dry weight of the roots, stems, and leaves and the EC_{iw} , regardless of the LF (for roots, $Y = 0.55 \text{ EC}_{\text{iw}} + 6.24$, $R^2 = 0.87$, $n = 10$; for stems, $Y = 1.68 \text{ EC}_{\text{iw}} + 20.47$, $R^2 = 0.91$, $n = 10$; and for leaves, $Y = 1.25 \text{ EC}_{\text{iw}} + 15.74$, $R^2 = 0.93$, $n = 10$).

Effects of the EC_{iw} and LF on the CCI and gas exchange of the hot peppers. Neither the EC_{iw} nor the LF had any significant effect on the CCI on 16 DAT. On 69 and 84 DAT, it decreased with an increase in the EC_{iw}

Table 1. Dynamics of mean values of plant height (cm) under varying irrigation water salinity levels (EC_{iw} , $dS\cdot m^{-1}$) and leaching fractions (LFs), and the output of the two-way analysis of variance (ANOVA) with regard to the height of the hot peppers.

	Days after transplanting						
	13	25	37	44	55	67	84
EC_{iw}							
0.9	25.0	35.5 a	41.6 ab	42.4 ab	46.1 a	51.6 a	54.1 ab
1.6	25.1	35.7 a	42.1 a	45.1 a	49 a	52.7 a	55.9 a
2.7	24.0	34.4 ab	40.1 ab	43.4 a	45.7 ab	49.3 ab	53.4 ab
4.7	24.9	32 ab	37.9 b	38.4 bc	41.6 bc	46.6 bc	49.6 b
7.0	23.3	31.3 b	33.9 c	35.1 c	38.8 c	43.1 c	43.3 c
LF							
0.17	24.3	32.9	38.2	40.2	43.7 b	46.9 b	49.6 b
0.29	24.6	34.8	40	41.4	45.6 a	50.3 a	52.3 a
ANOVA							
LF	NS	NS	NS	NS	*	*	*
EC_{iw}	NS	*	***	***	***	***	***
$LF \times EC_{iw}$	NS	NS	NS	NS	NS	NS	NS

NS, *, ***Nonsignificant or significant differences between means at 0.05 and 0.001 level of probability, respectively.

Table 2. Mean values of plant growth variables under varying irrigation water salinity levels (EC_{iw} , $dS\cdot m^{-1}$) and leaching fractions (LFs), and the output of the two-way analysis of variance (ANOVA) for plant growth variables.

Factors	Stem diam (mm)	Leaf length (cm)	Leaf area ($m^2/plant$)	Maximum root length (cm/plant)	Dry weight (g)		
					Root	Stem	Leaf
EC_{iw}							
0.9	11.6 a	6.3 a	0.32 a	29.8	5.5 a	19.7 a	14.3 a
1.6	11.6 a	6.2 a	0.32 a	32.4	5.9 a	18.0 a	14.0 ab
2.7	10.9 ab	6.1 ab	0.26 b	32.0	4.5 b	14.3 b	12.5 b
4.7	10.8 b	6.0 b	0.18 c	31.7	3.4 c	13.1 b	9.8 c
7.0	9.7 c	5.5 c	0.12 d	30.3	2.5 d	8.8 c	7.0 d
LF							
0.17	10.6 b	5.9 b	0.22 b	31.6	4.2	14.1 b	10.9 b
0.29	11.3 a	6.1 a	0.26 a	30.9	4.6	15.5 a	12.2 a
ANOVA							
LF	**	*	**	NS	NS	*	*
EC_{iw}	***	***	***	NS	***	***	***
$LF \times EC_{iw}$	NS	NS	NS	NS	NS	NS	NS

NS, *, **, ***Nonsignificant or significant differences between means at 0.05, 0.01, and 0.001 level of probability, respectively. Different letters within a column for each experimental factor indicate significant difference at $P < 0.05$ by Duncan's multiple range tests.

(Table 3), whereas a significant reduction appeared only in the EC_{iw} of $7.0 dS\cdot m^{-1}$. The CCI in the EC_{iw} of $7.0 dS\cdot m^{-1}$ on 84 DAT decreased by 5.8% to 14.3% as compared with 69 DAT. Across the EC_{iw} , the CCI in the LF of 0.29 was significantly higher than that in the LF of 0.17 on 26, 69, and 84 DAT. Throughout the experiment, no significant interaction was observed between the EC_{iw} and the LF in terms of CCI.

Variations in the P_n , g_s , T_r , and C_i over three measurements taken between 9:00 and 11:00 AM under varying levels of EC_{iw} and LF during 23–76 DAT are illustrated in Table 4. An increase in the EC_{iw} caused a significant ($P < 0.001$) decrease in the P_n , g_s , T_r , and C_i . The LF had no effect on the P_n , g_s , T_r , and C_i over the three measurements, except for the C_i on 23 DAT and the T_r on 76 DAT. Significant interactive effects were observed for the P_n on 23 and 39 DAT, for the g_s on 39 DAT, and for the T_r on 76 DAT. Significantly negative linear relationships between the P_n , T_r , and g_s and the EC_{iw} were observed over the three measurements and the LF had no effect on these parameters, except for the T_r on 76 DAT.

Effects of the EC_{iw} and LF on the K^+ , Ca^{2+} , and Na^+ concentrations of hot pepper roots and stems. Table 5 shows the K^+ , Ca^{2+} , and

Na^+ concentrations and the K^+/Na^+ ratio in the hot pepper roots and stems under different salinity and LF treatments. The K^+ and Na^+ concentrations in the roots increased significantly, whereas the K^+/Na^+ ratios decreased significantly as the EC_{iw} increased. The Ca^{2+} concentrations in the roots was not affected by either the EC_{iw} or the LF.

In the stems, an increase in the EC_{iw} caused a significant decrease in the K^+ concentration and the K^+/Na^+ ratio, but a significant increase in the Ca^{2+} and Na^+ concentrations. An increase in the LF apparently caused a significant decrease in the Na^+ concentration in the roots and stems, but an increase in K^+ concentration in the stems and the K^+/Na^+ ratio in the roots and stems. Significant negative linear correlations between the dry weight and the K^+ and Na^+ concentrations in the roots, as well as between the dry weight and the Na^+ concentrations in the stems were found, whereas the linear relationship between the dry weight of stems and the K^+/Na^+ ratio in the stems was positive (Table 6).

Discussion and Conclusions

In this study, the growth of hot pepper plants was found to be reduced when the

Table 3. Variation of leaf chlorophyll content under varying irrigation water salinity levels (EC_{iw} , $dS\cdot m^{-1}$) and leaching fractions (LFs), and the output of the two-way analysis of variance (ANOVA) for leaf chlorophyll content.

Factors	Days after transplanting (d)			
	17	26	69	84
EC_{iw}				
0.9	62.9	62.3	72.9 a	75.8 a
1.6	62.3	62.0	72.2 a	75.8 a
2.7	63.3	61.7	72.4 a	74.9 a
4.7	61.8	62.4	72.3 a	73.7 a
7.0	63.6	62.3	68.8 b	61.9 b
LF				
0.17	62.0	61.1 b	70.8 b	70.7 b
0.29	63.5	63.1 a	72.7 a	74.1 a
ANOVA				
LF	NS	***	*	**
EC_{iw}	NS	NS	**	***
$LF \times EC_{iw}$	NS	NS	NS	NS

NS, *, **, ***Nonsignificant or significant differences between means at 0.05, 0.01, and 0.001 level of probability, respectively. Different letters within a column indicate significant difference at $P < 0.05$ by Duncan's multiple range tests.

EC_{iw} was higher than $1.6 dS\cdot m^{-1}$ and reduction was more significant as the EC_{iw} further increased to $7.0 dS\cdot m^{-1}$ (Table 2). The dry weight of roots, stems, and leaves in the EC_{iw} of $7.0 dS\cdot m^{-1}$ was reduced by 51.3% to 54.4%, as compared with that of EC_{iw} of $0.9 dS\cdot m^{-1}$. A similar response in the EC_{iw} of $8 dS\cdot m^{-1}$ was reported by Lycoskoufis et al. (2005), in which salinity stress (recirculating the nutrient solution) reduced the dry weight in the stems and roots in the pepper plants by 50%, compared with that when standard nutrition ($1.9 dS\cdot m^{-1}$) was used. Azuma et al. (2010) showed, however, that the dry weight of the whole hot pepper plant in the EC_{iw} of $13.3 dS\cdot m^{-1}$ was only reduced by 29% relative to the control (a nutrient solution containing no NaCl). These results show that the response may, in fact, vary with different cultivars. Niu et al. (2010) found that the reduction of the dry weight of shoots in an EC_{iw} of $4.1 dS\cdot m^{-1}$ ranged from 22% to 92% for eight chili peppers (five cultivars of *Capsicum annuum* L., two cultivars of *Capsicum chinense*, and one accession of *C. annuum*). The maximum length of roots was unaffected by the EC_{iw} and LF because the pot size was limited, which is different from the report by Shivakumara et al. (2017). However, the plant height, SD, leaf length, and leaf area were also markedly diminished when the EC_{iw} was higher than 1.6 or $2.7 dS\cdot m^{-1}$. The inhibition of leaf development is largely because of the osmotic effect of the salinity (Munns and Tester, 2008), and the mechanism behind decreased leaf growth, which is independent of carbohydrate supply and water status, must be regulated by long distance signals in the form of their precursors or hormones (Munns et al., 2000). In addition, water deficit caused by salinity is one of the major reasons behind the suppression of plant growth in saline soils (Bhatt et al., 2008).

Plants irrigated with saline water usually accumulate inorganic ions in vacuoles, decreasing cell water potential (Yang et al.,

Table 4. Effects of irrigation water salinity (EC_{iw} , $dS\cdot m^{-1}$) and leaching fractions (LFs) on photosynthesis rate (P_n , $\mu mol\ CO_2\ m^{-2}\cdot s^{-1}$), leaf stomatal conduction (g_s , $mol\ H_2O\ m^{-2}\cdot s^{-1}$), transpiration rate (T_r , $mmol\ H_2O\ m^{-2}\cdot s^{-1}$), and intercellular CO_2 concentration (C_i , $\mu mol\ CO_2\ mol^{-1}$) measured on 23, 39, and 76 d after transplanting (DAT), and the output of the two-way analysis of variance (ANOVA) for the aforementioned parameters.

	23 DAT				39 DAT				76 DAT			
	P_n	g_s	T_r	C_i	P_n	g_s	T_r	C_i	P_n	g_s	T_r	C_i
EC_{iw}												
0.9	12.33 a	0.21 a	4.86 a	235.1 a	24.32 a	0.92 a	10.44 a	313.1 b	22.06 a	1.18 a	10.39 a	304.3 ab
1.6	10.79 b	0.17 b	3.99 b	227.7 a	24.76 a	0.94 a	9.67 ab	329.3 a	21.27 a	1.22 a	10.12 ab	307.9 a
2.7	9.91 b	0.16 b	3.83 b	230.5 a	23.41 a	0.84 a	9.51 b	328.6 a	20.80 a	0.92 b	9.63 b	299.3 b
4.7	7.91 c	0.12 d	2.93 c	227.3 a	21.27 b	0.52 b	8.36 c	310.0 b	20.63 a	0.73 c	8.21 c	292.7 c
7.0	5.24 d	0.06 d	1.64 d	204.4 b	19.06 c	0.42 c	7.59 d	298.0 c	17.75 b	0.55 d	7.27 d	289.9 c
LF												
0.17	9.21	0.14	3.33	231.5 a	22.66	0.75	9.10	314.5	20.82	0.88	8.18 b	298.9
0.29	9.26	0.15	3.57	218.5 b	22.47	0.71	9.13	314.0	20.19	0.95	10.07 a	298.8
ANOVA												
LF	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	***	NS
EC_{iw}	***	***	***	***	***	***	***	***	***	***	***	***
$LF \times EC_{iw}$	*	NS	NS	NS	*	*	NS	NS	NS	NS	***	NS

NS, *, **, ***Nonsignificant or significant differences between means at 0.05, 0.01, and 0.001 level of probability, respectively. Different letters within a column for each experimental factor indicate significant difference at $P < 0.05$ by Duncan's multiple range tests.

2009). Excessive amounts of ions enter the plant, restricting photosynthesis and growth rate (Munns, 2002). Excessive Na^+ has an adverse effect on plant growth (Niu et al., 1995), whereas K^+ is of great importance in a number of physiological processes (e.g., turgor maintenance, enzyme activation, stomatal behavior, osmotic adjustment, cell expansion, and membrane polarization) (Hatamnia et al., 2013). High K^+ and low Na^+ in the cytoplasm play a critical role for the maintenance of a series of enzymatic processes (James et al., 2006). Salinity stress causes excessive Na^+ and a reduction in K^+ in plant tissues (Li et al., 2010), which may cause nutritional imbalance.

In this study, we observed that the Na^+ concentration increased and the K^+ concentration and K^+/Na^+ ratio decreased in the stems (Table 5), indicating competitive inhibition between the absorption of K^+ and Na^+ in hot peppers under higher salinity stress (Li et al., 2010). There is also a correlation between the accumulation of Na^+ in plants under salt stress and growth reduction (Garcia-Legaz et al., 2005) which was also confirmed in this study (Table 6). Ashraf and Ahmad (2000) have shown that the inhibition uptake of K^+ and Ca^{2+} in the root zone under salt stress might be another reason for plant growth reduction. However, in this study, the Ca^{2+} concentration in the root zone was unaffected by the EC_{iw} and the K^+ concentration in the roots increased as the EC_{iw} increased, indicating that the uptake of mineral nutrition in the root zone was not the limiting factor that affects plant growth, which can be attributed to the addition of the half-strength Hoagland solution to the saline water in our experiment.

Plant height, the width of plant stems, leaf length, leaf area, and the dry weight of stems and leaves were higher in the higher LF, indicating that higher LF alleviated the inhibition of salinity stress on plant growth. There was a correlation between the decline in the P_n and reduced plant growth (Hatamnia et al., 2013). However, a decrease in the LF did not always lead to a reduction in the P_n , length, and dry weight of roots, but they did

Table 5. Effects of irrigation water salinity (EC_{iw} , $dS\cdot m^{-1}$) and leaching fractions (LFs) on the K^+ ($mg\cdot g^{-1}$ DW), Ca^{2+} ($mg\cdot g^{-1}$ DW), and Na^+ ($mg\cdot g^{-1}$ DW) concentrations and K^+/Na^+ ratio of hot pepper roots and stems.

	Roots				Stems			
	K^+	Ca^{2+}	Na^+	K^+/Na^+	K^+	Ca^{2+}	Na^+	K^+/Na^+
EC_{iw}								
0.9	11.13 bc	14.01	3.01 d	3.94	16.82 ab	12.01 c	0.23 d	76.2 a
1.6	10.67 c	15.94	5.95 c	1.84	16.67 ab	12.72 bc	0.71 d	24.17 b
2.7	10.81 c	15.79	7.81 b	1.43	20.21 a	16.83 a	1.57 c	14.14 c
4.7	12.15 ab	16.61	8.53 b	1.45	12.88 b	15.55 ab	2.95 b	5.02 cd
7	13.13 a	16.05	12.6 a	1.05	14.1 b	16.77 a	4.75 a	3.43 d
LF								
0.17	11.29	15.78	8.07 a	1.67 b	13.77 b	13.92	2.66 a	16.55 b
0.29	11.84	15.58	7.28 b	2.47 a	17.87 a	15.85	1.78 b	22.59 a
ANOVA								
LF	NS	NS	*	**	**	NS	**	*
EC_{iw}	***	NS	***	***	**	*	***	***
$LF \times EC_{iw}$	NS	NS	NS	**	*	NS	*	NS

NS, *, **, ***Nonsignificant or significant differences between means at 0.05, 0.01, and 0.001 level of probability, respectively. Different letters within a column for each experimental factor indicate significant difference at $P < 0.05$ by Duncan's multiple range tests.

Table 6. Linear correlation between dry weight of roots and stems and the Na^+ concentration, K^+ concentration, and K^+/Na^+ ratio in root and stems, regardless of the leaching fraction.

Dependent variable (Y)	Independent variable (X)	Function	Determination coefficient (R^2)
Root dry weight	K^+ concentration in roots	$Y = -1.02X + 16.14$	0.65**
Root dry weight	Na^+ concentration in roots	$Y = -0.35X + 7.03$	0.75**
Stem dry weight	Na^+ concentration in stems	$Y = -2.13X + 19.14$	0.90***
Stem dry weight	K^+/Na^+ ratio in stems	$Y = 11.6X + 11.94$	0.65**

, *Significance of line at $P < 0.01$ and 0.001, respectively.

increase in the electrical conductivity of soil-saturated paste extract (EC_e) (Qiu et al., 2017a). The lower osmotic potential of soil water due to higher root-zone EC, Na^+ stress in roots and stems, and the inhibition of K^+ in stems (Table 5) can result in a lower uptake of water by the roots and the T_r , in turn, reduces growth (Romero-Aranda et al., 2001).

The EC_{iw} could strongly affect the gas exchange parameters. That salinity stress can cause a dramatic reduction of P_n was also observed in our study, especially when a large quantity of EC_{iw} is used at the vegetative growth stage. It is generally believed that a reduction of the P_n in plants irrigated with saline water might be because of a decrease in partial pressure in terms of C_i , caused by

stomatal closure or nonstomatal limitation (Bethke and Drew, 1992; Silva et al., 2010), thus limiting g_s and photosynthetic CO_2 assimilation (Downton et al., 1990; Yang and Lu, 2005). Nonstomatal factors include the effects of osmotic stress, biochemical constituents (Parida and Das, 2005; Sultana et al., 1999), the contents of photosynthetic pigments (Koyro, 2006; Ma et al., 1997), and the energy spent in managing the distribution and allocation of absorbed ions at different cellular levels (Ahmed et al., 2012). Many studies have shown that nonstomatal limitations might prevail over stomatal limitations with high levels of salinity (Qin et al., 2010). This study shows that, across the LF, there was a considerable reduction (53.4% to 70.5%) of g_s in the EC_{iw} of 7.0 $dS\cdot m^{-1}$

compared with that of EC_{iw} of $0.9 \text{ dS}\cdot\text{m}^{-1}$, but the reduction of C_i was small (3.8% to 13.1%) over the three measurements. This shows that a decrease in g_s does not diminish C_i and that the decrease in the P_n might be because of both stomatal and nonstomatal factors. The significant decrease in the CCI observed on 69 and 84 DAT (Table 3), the increase of salinity in the drainage water and soil, and the increase of Na^+ concentration in the high EC_{iw} treatments reflect the nonstomatal limitations in this study. The reduction of the P_n shows that salt built up in the leaves with adverse effects on photosynthesis (Hatamnia et al., 2013). It is well known that a reduction in P_n is positively related to a decrease in g_s and T_r (Lu et al., 2009; Qin et al., 2010; Wang et al., 2011). The notable reduction in g_s and T_r (Table 4) is not only a negative consequence of excessive salt but also a representative of the adaptive mechanisms in place to cope with it (Clark et al., 1999; Flanagan and Jefferies, 1989; Koyro, 2006).

The LF had no effect on the P_n and g_s over time. The C_i under the two LFs reached a similar level on 39 and 76 DAT. Interestingly, the C_i was significantly higher in the LF of 0.17 than that of 0.29 on 23 DAT (at the vegetative growth stage). This phenomenon might be explained by greater changes in the mesophyll demand for CO_2 in the lower LF in a normally functioning leaf, where ambient CO_2 concentration is a constant at this stage (Mott, 1988). Across the EC_{iw} , the LF had no effect on the T_r on 23 and 39 DAT. However, the inhibitory effect on the T_r was more severe in the low LF on 76 DAT (Table 4). An increase in the LF did not lead to an increase in the g_s (Table 4), the length or dry weight of the roots (Table 2), and is, therefore, likely to have been caused by the higher osmotic potential of the soil water and, consequently, the higher availability of soil water for the plants (Chen et al., 2016). This result might also partly explain our previous study concerning the reduction of the daily ET of the hot peppers under the lower LF from 65 DAT (Qiu et al., 2017a).

In summary, the present study demonstrates that growth and gas exchange were both markedly inhibited by the EC_{iw} . The SD; leaf length; leaf area; the dry weight of the roots, stems, and leaves; and the P_n , g_s , T_r , and C_i were significantly reduced as the EC_{iw} increased. An increase in the EC_{iw} led to an increase in Na^+ concentrations, whereas there was a decrease in the K^+/Na^+ ratio in the roots and stems.

A higher LF removed more salt out of the root zone, which in turn led to a corresponding increase in plant growth. However, the LF did not significantly affect either the dry weight of roots or the gas exchange, except for the C_i measured on 23 DAT and the T_r on 76 DAT. An increase in the LF caused a reduction in the Na^+ concentration in the roots and stems, whereas there was an increase in the K^+ concentration in the stems and the K^+/Na^+ ratio in the roots and stems.

Our research showed that high LF can relieve Na stress and encourage the growth of hot pepper plants. Therefore, higher saline water should be applied with higher quantity of water for satisfactory performance for hot pepper production. However, there was a time lag for the LF effect on leaf transpiration and plant growth, also on ET, EC_e , and yield [see our previous studies (Qiu et al., 2017a, 2017b)], due to the gradual building up of soil salinity during the growth season, which deems further studies for the other crop. This result is valuable for saline irrigation and salinity management, especially in arid and semiarid region where fresh water is shortage. If crops have short-term growth stage and LF have no favorable short-term effect on gas exchange, plant growth, and yield, saline water with lower LF might be adopted for irrigation.

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