Turkish Journal of Agriculture and Forestry
http://journals.tubitak.gov.tr/agriculture/
Research Article

Turk J Agric For
(2015) 39: 876-886
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doi:10.3906/tar-1412-106

Tomato salt tolerance: impact of grafting and water composition on yield and ion relations

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Received: 24.12.2014 • **Accepted/Published Online:** 08.06.2015 • **Printed:** 30.11.2015

Abstract: We evaluated the salt tolerance of tomato cv Big Dena under both nongrafted conditions and when grafted on Maxifort rootstock, under a series of 5 salinity levels and 2 irrigation water composition types. The salinity levels of the irrigation water were −0.03, −0.15, −0.30, −0.45, and −0.60 MPa osmotic pressure (corresponding to specific electrical conductivity values of 1.2, 4.0, 8.5, 12, and 15.8 dS m^{-1}, respectively). We salinized the irrigation water with either a mixture of salts with a predominant composition consisting of Na^+–Ca^{2+}–Cl^- salts, a composition typical of coastal Mediterranean ground waters or, alternatively, a salt composition that was of mixed Na^+–Ca^{2+}–SO_4^{2-}–Cl^- ions, a water composition more typical of interior continental basin ground waters such as those of the California Central Valley in the US. We determined that there were no statistically significant differences in tomato salt tolerance (fruit yield) relative to water type. This result indicates that in the range of Cl^- concentrations tested in our experiment (up to 150 mmol L^{-1}), Cl^- is not an important factor in tomato yield reduction associated with salinity. The grafted Big Dena on Maxifort tomato plants exhibited increased yield both under control and elevated salinity levels relative to the nongrafted Big Dena plants. In contrast to absolute yield relationships, expression of salt tolerance in terms of relative yield, as salt tolerance is commonly expressed, provides the conclusion that grafted Big Dena on Maxifort tomato plants are slightly less salt tolerant than nongrafted Big Dena plants. Our data also indicate that, for tomato, decreased yield under saline conditions is well related to increased leaf Na^+ concentrations.

Key words: Chloride salinity, grafting, irrigation, salt tolerance, sulfate salinity, tomato

1. Introduction

Salinity is a major abiotic plant stress that is increasing worldwide. Increasing salinization of agricultural soils is aggravated by an increasing scarcity of fresh water and thus a need to utilize more saline waters for irrigation. Salinity has an adverse impact on agriculture as it can cause large losses in crop productivity, thus threatening world food security. Salinity affects plant growth by imposing both osmotic and specific ion stresses (Castillo et al., 2007). Increasing the salt tolerance of crops through plant breeding could increase the sustainability of irrigation with low-quality water by reducing the need for leaching and allowing the use of poorer quality water (Gawad et al., 2005). Intensive farming practices with limited crop rotation have also been considered to potentially contribute to increased soil salinity (King et al., 2010). High concentrations of NaCl disrupt the plant osmotic balance and result in a decrease in plant water uptake and closing of stomatal apertures, leading to transpiration inhibition (Munns and Tester, 2008).

Breeding new salt-tolerant crop varieties is one strategy to alleviate the impacts of salinity on crop production, but success has been limited (Flowers, 2004). Grafting has been utilized to obtain plants with higher fruit quality and production (Lee, 1994). It is thus of great interest to know whether the grafting technique is a valid strategy for either improving the salt tolerance in tomato (Santa-Cruz et al., 2001) or increasing yield under saline conditions. Despite the initial objective of vegetable grafting to improve crop resistance to soil-borne diseases, the yield increase of grafted vegetables has been directly linked to improvement of tolerance to abiotic stresses (including low and high temperatures, salinity, flooding), enhancement of nutrient and water uptake, and delayed senescence (Zhao and Simonne, 2008). Grafting has also been utilized to reduce infection by soil-borne diseases caused by pathogens (Biles et al., 1989), to increase plant resistance to low temperatures (Tachibana, 1982, 1988, 1989), and is documented to increase water use efficiency for field tomatoes irrigated with the drop and furrow method (Semiz and Yurtseven, 2010).

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A number of studies have reported on the response of grafted plants to salinity. Interpretation of the results can be confusing as some studies did not evaluate salt tolerance as such but rather reported on the yield of grafted versus nongrafted plants under saline conditions. Additionally, plants have been grafted onto their own rootstock as well as onto other rootstocks. Grattan and Maas (1985) determined that grafting the rootstock of soybean cultivar Lee, a salt tolerant cultivar that excludes Cl^- in the leaves, onto salt-sensitive cultivars that accumulate Cl^- in the leaves reduced Cl^- in the leaves and enhanced salt tolerance. In addition, grafting one variety onto itself did not significantly alter Cl^- accumulation in any of the four cultivars tested (Grattan and Maas, 1985). The impact of grafting on the salt tolerance of soybean thus appears related to the characteristics of the rootstock rather than grafting itself.

Plant salt tolerance is commonly described by the decline in yield as related to increasing salinity or the salinity level at which yield starts to decline (Maas and Hoffman, 1977; Ayers and Westcot, 1985; Mass and Grattan, 1999; Grieve et al., 2012). Relative salt tolerance is defined as yield under saline conditions divided by yield under nonsaline conditions. Plants that have increased yield at a specific salinity level may thus be either more salt tolerant, more vigorous at all salinity levels, more vigorous and more salt tolerant, or more vigorous and less salt tolerant. Determination of salt tolerance thus requires that plants be grown under a range of salinity conditions.

Watermelon grafted onto an apparently more salt tolerant rootstock demonstrated increased vegetative growth relative to nongrafted plants in a 2-week-long study with NaCl as the salinizing solution (Goreta et al., 2008). Huang et al. (2009) examined the effect of grafting on cucumber salinized with NaCl. They noted a greater number of fruit and a greater fruit yield when salt-tolerant rootstocks were grafted onto a sensitive variety as compared to self-grafted plants of the sensitive variety. Their data indicated somewhat greater yield on the tolerant/sensitive-grafted plants relative to the self-grafted ones, but additionally our calculations showed a large increase in their salt tolerance (as defined above). Estan et al. (2005) examined the effect of grafting various rootstocks on tomato fruit yield after salinizing with NaCl. They observed no difference in fruit yield on self-grafted versus nongrafted Jaguar cultivars under salinizing treatments but they did observe increased yield under control conditions and increased salt tolerance when other rootstocks were grafted onto Jaguar cultivars. They attribute the increased salt tolerance of the other rootstocks to the regulation of both Na and Cl transport by the rootstocks. Recently, Di Gioia et al. (2013) reported on the response of Maxifort rootstock grafted on a salt-sensitive heirloom variety. They recorded an almost 50% higher yield for the grafted plants under control conditions but no differences at salinity levels of 20 and 40 mM NaCl. In contrast, Arnold rootstock grafted to the heirloom variety had an approximately 10% increase in yield under control conditions but was about 25% greater relative to the nongrafted heirloom at 20 mM salinity.

Increased tolerance to salinity was related to reduced Na^+ in the vegetation with no change in Cl^- (Goreta et al., 2008), reduced concentrations of Na^+ and Cl^- (Estan et al., 2005), and increased K^+ with lower Na^+ and Cl^- (Huang et al., 2009). Edelstein et al. (2011), in a study with one salt level (EC 1.9 dS m^{-1}), determined that melon grafted on pumpkin rootstock decreased plant Na^+ relative to nongrafted and self-grafted melon, while plant Cl^- concentrations were similar among the rootstocks. Di Gioia et al. (2013) did not report on Cl, but their data related salinity response of Arnold grafted plants to Na partitioning into older leaves as compared to the nongrafted heirloom. They did not provide data on Na accumulation in Maxifort grafted plants. Since Cl was not determined, the Cl accumulation of Maxifort and Armstrong was not discussed.

Tomato is one of the most important horticultural crops in the world. Tomato production is very concentrated in semiarid regions, where saline waters are frequently used for irrigation and salinity problems are most severe. For example, more than 30% of the world tomato production comes from countries around the Mediterranean Sea and about 20% from California (FAO, 1995). The increasing salinity in the groundwater in these regions and the decreased availability of fresh water for food production means that there is a critical need to evaluate options for increasing crop salt tolerance.

The most popular tomato rootstock cultivars commercially available in the US are Maxifort and Beaufort, both released by De Ruiter Seeds (Bergschenhoek, the Netherlands). Maxifort and Beaufort are reported to be resistant to tomato mosaic virus, fusarium root rot and fusarium crown rot, corky root, verticillium, and nematodes (King et al., 2010). Zhao and Simonne (2008) also stated that a few seed companies can currently provide tomato rootstocks in the US. Maxifort (De Ruiter Seeds) is one of the most popular rootstocks for greenhouse tomato production in the US because of its prominent disease resistance, high grafting compatibility, and strong vigor. Improved yield performance related to rootstock selection had no impact on the fruit quality attributes of grafted tomatoes (measured as firmness, pH, soluble solids, titratable acidity, and concentrations of lycopene and minerals) despite increased production (Khah et al., 2006). We found no data on the effect of Maxifort grafting on the yield of high production commercial tomato varieties under saline conditions and no information on Na and Cl plant accumulation related to use of Maxifort as a grafting rootstock.

The objective of this study was to determine the effects of the salinity (expressed as osmotic potential, OP) of irrigation water dominated by either chloride (Cl^-) or mixed sulfate (SO_4^{2-}) and Cl^- anions with mixed Ca^{2+} and Na^+ salts on the yield of Big Dena (a widely used commercial variety) grafted onto Maxifort and nongrafted Big Dena tomato in greenhouse sand culture. We also wanted to evaluate the salt tolerance of this grafted combination and relate yield to leaf ion composition and total soluble solids.

2. Materials and methods

Commercial tomato seedlings were purchased from Bevo Farms (Milner, BC, Canada). Grafted seedlings consisted of Maxifort rootstocks and Big Dena scions. Nongrafted seedlings were of the variety Big Dena (whole plant). Earlier, Estan et al. (2005) established that self-grafting and nongrafted tomato plants had comparable yield, similar to the findings of Grattan and Mass (1982) with soybean. The OP levels examined in our study were –0.003 (control, with nutrition added at the same concentrations as the treatments), –0.15, –0.30, –0.45 and –0.60 MPa. The compositions of the major ions of the various irrigation waters are presented in Table 1, along with the electrical conductivities of the waters. The compositions were calculated using the ExtractChem computer model to achieve the target OP levels (Suarez and Taber, 2007). Modified half Hoagland's solution (plant nutrient solution) was prepared and added to the irrigation reservoirs as 0.17 KH_2PO_4, 0.75 $MgSO_4{\cdot}7H_2O$, 2.0 KNO_3, and 0.25 $CaSO_4{\cdot}2H_2O$ mM with micronutrients, also expressed in mM, of 0.34 KH_2PO_4, 0.050 Fe (as sodium ferric diethylenetriamine pentaacetate), 0.023 H_3BO_3, 0.005 $MnSO_4$, 0.0004 $ZnSO_4$, 0.0002 $CuSO_4$, and 0.0001 H_2MoO_4. The salts added to achieve the target concentrations of major ions were NaCl, $CaCl_2{\cdot}2H_2O$, $Na_2SO_4{\cdot}10H_2O$, and $MgSO_4{\cdot}7H_2O$. The prepared solutions for each tank were analyzed and the concentrations of the individual tanks were adjusted such that they varied by less than 5% from the values reported in Table 1. The pH of the water was adjusted with HCl to maintain pH and nitrate concentration during the experiment.

The sand tanks (1.2 × 0.6 × 0.5 m deep) contained washed sand having an average bulk density of 1.4 Mg m^{-3} (g cm^{-3}) with a sand volume of 0.29 m^3. At saturation, the sand had an average volumetric water content of 0.34 $m^3\ m^{-3}$, thus storing 100 L of solution in each tank. The experimental design consisted of five OP levels (including the control) and two salt compositions with three replications, for a total of 30 tanks.

The salt treatments were prepared to represent two water types, either equal (in $mmol_c\ L^{-1}$) concentrations of Ca^{2+} and Na^+ with Cl^- as the anion, designated as a chloride water, or a mixed salt solution better representing arid zone interior valleys of the US (and world), with a relatively high SO_4^{2-} concentration, $Na^+ > Ca^{2+}$ at high salinity, and increasing Mg^{2+} with increasing salinity, designated as a sulfate–chloride water. The specific compositions used for both water types at varying osmotic pressures are given in Table 1. With increasing salinity, it is necessary to increase the Cl^-/SO_4^{2-} ratio as the SO_4^{2-} concentration is constrained by gypsum solubility, which is the same trend as observed in natural waters of mixed sulfate and chloride type.

Table 1. Irrigation water composition.

Osmotic potential, MPa	Cations ($mmol_c\ L^{-1}$)				EC, dS m^{-1}	pH	Anions ($mmol_c\ L^{-1}$)			
	Na^+	K^+	Ca^{2+}	Mg^{2+}			SO_4^{2-}	Cl^-	PO_4^{-2}	NO_3^-
Control (–0.03)	1.5	3.0	4.0	2.0	1.200	4.90	2.0	3.0	1.0	5.0
–0.15 Cl^-	16.0	3.0	16.0	2.0	3.988	4.92	2.0	29.0	1.0	5.0
–0.30 Cl^-	36.0	3.0	36.0	2.0	8.260	4.92	2.0	69.0	1.0	5.0
–0.45 Cl^-	55.5	3.0	55.5	2.0	12.02	4.93	2.0	109.0	1.0	5.0
–0.60 Cl^-	75.0	3.0	75.0	2.0	15.84	4.94	2.0	148.0	1.0	5.0
Control (–0.03)	1.55	3.0	4.0	2.0	1.200	4.90	2.0	3.0	1.0	5.0
–0.15 SO_4^{2-}–Cl^-	15.7	3.0	14.7	7.6	4.334	4.93	19.8	20.8	1.0	5.0
–0.30 SO_4^{2-}–Cl^-	32.0	3.0	32.0	16.0	8.875	4.93	45.5	45.5	1.0	5.0
–0.45 SO_4^{2-}–Cl^-	51.0	3.0	34.9	25.5	12.20	4.92	56.9	75.0	1.0	5.0
–0.60 SO_4^{2-}–Cl^-	74.0	3.0	36.0	33.0	15.82	4.93	65.4	105.5	1.0	5.0

The experimental treatment for each tank was randomly selected for the factorial design. Two grafted and two nongrafted seedlings were sown in each tank. All tanks were flood-irrigated with the same amount of water at a frequency of once per day. Approximately 500 L was applied to each tank during each irrigation (5 times the holding capacity of the sand), fully saturating the sand and re-equilibrating the soil water salinity to that of the irrigation water. The drainage water flowed back to the irrigation water reservoir. The amount of water consumed each day (approximately 0.3 L per tank) represents a small percentage of the water held by the sand (100 L at saturation); thus we can assume that the electrical conductivity (EC) of the soil water equals the EC of the irrigation water. Two weeks after planting the seedlings, salts were applied to the water reservoirs used for irrigation (900 L capacity) and all subsequent irrigations utilized these waters. The total soluble solids (TSS) or brix, which is the soluble solid contents, of the tomato juice was measured by refractometer, with measurements expressed in percent, i.e. grams of solid per 100 mL of solution, as commonly reported.

The irrigation waters were analyzed for Na K, Ca, Mg, S, Fe, Mn, Cu, and Zn after acidification with analytical-grade nitric acid using PerkinElmer Optima 3300DV ICP OES (inductively coupled plasma optical emission spectroscopy) (PerkinElmer Corp, Waltham, MA, USA). The chloride analyses were done by amperometric titration using a Labconco chloridometer (Labconco, Kansas City MO, USA) and NO$_3^-$ was analyzed spectrophotometrically with a Hitachi model 100-20 (Hitachi Corp, Japan) at a 210 nm wavelength. The analyzed solutions were within 5% of the target values in Table 1. The plant and fruit samples were washed in deionized water, dried in a forced-air oven at 70 °C for 72 h, and ground in a Wiley mill to pass a 60-mesh screen. Total S, total P, Ca, Mg, Na, and K of the leaf and fruit tissue were determined from nitric–perchloric acid digests of the tissues by ICP OES. The fruit and leaf Cl$^-$ was determined on nitric–acetic acid extracts by amperometric titration. Statistical analyses of all data were performed using SPSS 16.0 (SPSS Inc., Chicago, IL, USA).

3. Results and discussion

3.1. Yield

The tomato fruit yields had no significant differences according to salt type, as shown in Table 2 ($P > 0.05$). The Cl$^-$ type and SO$_4^{2-}$–Cl$^-$ type irrigation water compositions had comparable Na$^+$ concentrations at each OP level,

Table 2. Fruit yield, fruit weight, and total soluble solids (TSS).

	Osmotic potential (MPa)	–0.003	–0.15	–0.30	–0.45	–0.60	Average
Yield kg plant^{-1}	Grafted Cl	8.31	6.93	6.03	3.90	2.51	
	Grafted SO$_4$	6.93	5.85	4.58	3.80	3.57	
	Average	7.62 Aa	6.39 Aa	5.30 ABa	3.85 Ba	3.04 BCa	5.202 a
	Nongrafted Cl	5.41	4.85	4.05	3.20	2.19	3.833 b
	Nongrafted SO$_4$	4.83	4.64	4.09	2.72	2.35	
	Average	5.12 Ab	4.75 Ab	4.07 ABb	2.96 Ba	2.27 BCa	
	↓ a, → A, Salinity $P < 0.001$, Grafting $P < 0.001$, Interaction (Grafting × Salinity) $P < 0.05$						
Fruit weight, g fruit^{-1}	Grafted Cl	196.47	193.28	131.92	76.89	71.62	
	Grafted SO$_4$	193.07	166.96	148.54	79.23	70.36	
	Average	194.77Aa	180.12 Aa	140.29 Ba	78.06 Ca	70.99 Ca	132.384 a
	Nongrafted Cl	197.48	159.67	109.79	73.35	66.00	
	Nongrafted SO$_4$	162.83	153.76	102.15	73.5	57.48	
	Average	180.16 Ab	156.72 Ab	105.97 Bb	73.43 Ba	61.74 Ca	120.002 b
	Average (salinity)	187.465 A	168.419 B	123.098 C	75.745 D	66.365 D	
	↓ a, → A, Salinity $P < 0.001$, Grafting $P < 0.001$, Interaction (Grafting × Salinity) $P < 0.05$						
TSS, brix, %	Grafted Cl	3.80	3.97	5.13	7.00	7.03	
	Grafted SO$_4$	2.97	4.67	4.87	5.87	7.23	5.253 b
	Nongrafted Cl	4.07	4.60	4.67	7.93	8.20	
	Nongrafted SO$_4$	3.47	4.67	5.20	7.60	7.80	5.820 a
	Average (salinity)	3.575 C	4.475 B	4.967 B	7.100 A	7.567 A	
	↓ a, → A, Salinity $P < 0.001$, Grafting $P < 0.005$						

Different lower case letters denote statistical significance among columns (salinity treatments), and upper case letters refer to statistical significance between rows (either grafted or nongrafted).

while the SO_4^{2-}–Cl^- type irrigation waters had lower Cl^- concentrations. For example, the Cl^- concentration at OP −0.60 MPa in the mixed anion water is comparable to the Cl^- concentration of the −0.45 MPa OP level (Table 1). The lack of a statistically significant response to water type in our study indicates that Cl^- ion toxicity was not an important aspect of tomato yield reduction with increasing salinity, consistent with the findings of Goreta et al. (2008) on watermelon and Edelstein et al. (2011) on melon and *Cucurbita*. Our results are consistent with the concept that, for many plants, Na^+ is the primary cause of ion-specific damage (Tester and Davenport, 2003). However, this cannot be generalized as Cl^- toxicity also exists for some crops such as avocado, citrus, grape, and strawberry (Grieve et al., 2012). Moreover, Colla et al. (2012) observed increased salt tolerance with Na_2SO_4 as compared to NaCl salts on cucumber.

The yield under nonsaline (control) conditions in our experiment was approximately 50% greater for the grafted as compared to the nongrafted plants (Figure 1). This is not surprising as grafting has been promoted and adopted based on increased yield relative to nongrafted plants. These values are also comparable to the increases seen by Di Gioia et al. (2013) under control conditions when they grafted Maxifort to an heirloom variety. Previous investigators examining nonsaline conditions have reported that yields of grafted plants increased relative to nongrafted plants. For example, in greenhouse and open field studies conducted by Khah et al. (2006), grafted tomato plants had a higher yield than nongrafted tomato plants.

Under nonsaline conditions in our study, the yield differences between grafted and nongrafted plants were at a maximum, with the differences decreasing somewhat as salinity increased (Figure 1). However, even at 150 $mmol_c$ L^{-1} salt (with 75 mM Na), the grafted plants still had a higher yield. This is in contrast to some other studies with grafted plants such as watermelon, where the differences in yield were reported as greatest at the higher salinity levels (Goreta et al., 2008), and studies where grafting did not improve yield under saline conditions. For example, Di Gioia et al. (2013) examined the salinity response of salt-sensitive heirloom tomato (*Solanum lycopersicum* L.), both nongrafted and grafted onto interspecific tomato hybrid rootstocks (*S. lycopersicum* × *S. habrochaites*) Maxifort and Arnold under increasing NaCl conditions. In the presence of 20 mM NaCl, plants grafted onto Arnold provided a marketable yield of 23.5% (on average) higher than Maxifort-grafted or nongrafted heirloom. Maxifort grafted to an heirloom variety had no increased yield relative to the nongrafted heirloom even under relatively mild salinity (20mM NaCl, corresponding to an EC of 2.0 dS m^{-1}). The specific response may thus depend on the relative ability of the rootstock to exclude Na or Cl relative to the shoot variety. For example, Grattan and Maas (1985) determined that soybean rootstock Lee excluded Cl while others did not and related this to soybean tolerance to salinity. Some grafting combinations increased yield as well as salt tolerance and some did not, as observed for tomato.

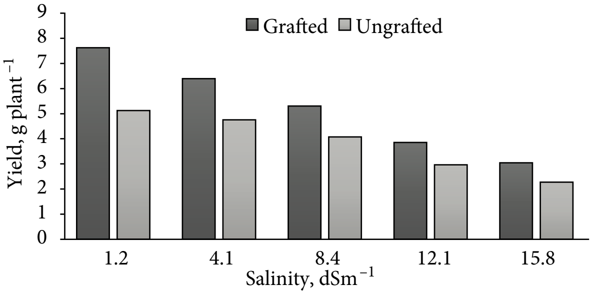

Figure 1. Mean fruit yield of grafted and nongrafted plants as related to irrigation water salinity. Results of the SO_4^{2-}–Cl^- and Cl^- type irrigation waters were combined as they were not statistically significant ($P > 0.05$).

Statistical analyses of our yield data showed that the grafting and salinity interactions were significant ($P < 0.05$) when comparing the average grafted to the average nongrafted yield (Table 2). The largest yields were obtained from the −0.003 MPa (control) and the −0.15 MPa OP levels (where −0.15 MPa corresponds to an electrical conductivity of approximately 4.1 dS m^{-1}). Above −0.15 MPa OP, there was a continuous decrease in yield with increasing salinity, with the yield differences between the grafted and nongrafted cultivars statistically significant at −0.3 MPa OP. Above −0.3 MPa OP, there were no statistically significant differences in yield between the grafted and nongrafted plants ($P > 0.05$). The yield differences between the grafted and nongrafted plants were 2.4, 1.64, 1.23, 0.89, and 0.77 kg fruit $plant^{-1}$ for the −0.003 (control), −0.15, −0.30, −0.45, and −0.60 MPa OP treatments, respectively. Salinity (high OP) adversely affected both grafted and nongrafted plants.

In a study with cucumber plants grafted onto bottle gourd rootstock, Huang et al. (2009) showed a decrease in yield with increasing salinity for both grafted and nongrafted plants, but in their study the grafted plants lost less yield with increasing salinity as compared to the nongrafted plants, indicating a greater salt tolerance.

3.2. Salt tolerance

Plant salt tolerance is typically expressed as the relative yield related to root zone salinity (Maas and Hoffman, 1977; Grieve et al., 2012). The commonly used piecewise linear model consists of a threshold salinity above which there is a yield decline, and a slope value representing the decrease in yield per unit of salinity (typically percent yield decline per unit of salinity, expressed as EC in units of dS m^{-1}). This model is widely used not only for reporting crop salt tolerance but also for recommendations regarding suitable crops or varieties to plant under saline conditions (Ayers and Westcot, 1985; Maas and Grattan, 2009; Grieve et al., 2012).

Santa-Cruz et al. (2002) evaluated the salt tolerance of two shoot tomato genotypes, Moneymaker, an excluder, and UC-82B, an includer (relative to NaCl accumulation), grafted onto a commercial hybrid tomato (cv Kyndia). They determined that the UC-82B Kyndia-grafted plants were more salt tolerant than the UC-82B self-grafted plants despite lower yields under control conditions. In contrast, the Moneymaker Kyndia-grafted plants had comparable yield and salt tolerance to the self-grafted Moneymaker plants. Data from Di Gioia et al. (2013) for Maxifort-grafted plants indicated markedly less salt tolerance as compared to the heirloom variety. In addition, De Gioia et al. (2013) reported that, in the presence of moderate salinity conditions (20 mM of NaCl), the rootstock Arnold showed greater yield than the rootstock Maxifort, yet at higher salinity levels (40 mM of NaCl), vegetable grafting did not enhance the yield.

This means that the salt tolerance response of shoot–rootstock grafting depends on the shoot characteristics as well as the rootstock. Thus, depending on the rootstock and shoot characteristics, grafting may or may not improve salt tolerance. Estan et al. (2005) determined that Jaguar shoots grafted onto 5 rootstocks generally resulted in improved yield (and salt tolerance as evaluated by one salt level, 50 mM), but the improvement varied and depended on the rootstock selected.

The relative yield data for our grafted and nongrafted tomato plants is shown in Figure 2. We used Extract Chem (Suarez and Taber, 2007) to calculate OP from concentrations and to convert the salinity values from OP to EC, as EC is the unit generally used to express salt tolerance; however, plants are considered to respond to OP not EC. Conversion of EC values from soil water to EC_e, the EC of a saturation extract, can be done by multiplying the values in Figure 2 by 0.472, the inverse of the ratio of the water content of the saturation paste to the water content of the sand at field capacity.

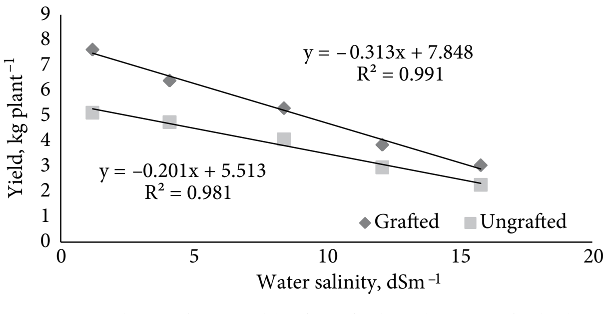

Figure 2. Relative fruit yield of grafted and nongrafted plants as related to irrigation water salinity. Results of the SO_4^{2-}–Cl^- and Cl^- type irrigation waters were combined as they were not statistically significant ($P > 0.05$).

For both the grafted and nongrafted plants, we did not find a salinity threshold; any increase in salinity above the control resulted in yield loss and a simple linear model fit the data well for both grafted and nongrafted plants (Figure 2). The yield decreases with increasing salinity (compared to the control treatment) for grafted tomatoes were 16.1%, 30.5%, 49.5%, and 60% and were 7.0%, 20.5%, 42.2%, and 55.7% for nongrafted tomato, respectively. Thus, in terms of salt tolerance, the nongrafted plants had more salt tolerance because they had less loss in relative yield at all salinity levels as compared to the grafted plants (Figure 2). The fitted linear model for our data indicated that the slope (yield decline in percent per unit of salinity increase) was −3.63 for nongrafted plants and −4.12 for grafted plants. If we convert the soil water EC data to corresponding EC_e values, the slope is −7.69 for nongrafted plants and −8.73 for grafted plants. Based on these data, and using the salt tolerance model as shown in Figure 2, we concluded that the salt tolerance of Big Dena tomato plants grafted onto Maxifort rootstock was slightly lower than that of nongrafted Big Dena plants due to the difference in the slope values. Grafting per se did not increase salt tolerance when grafting Big Dena to Maxifort rootstock. This result is similar to De Gioia et al. (2013), except that they observed a large decrease in salt tolerance on grafted Maxifort relative to nongrafted while our decrease was relatively low. This conclusion is relevant to developing methods to increase salt tolerance. However, from a production viewpoint, the grafted plants still had larger absolute yields at all salinity levels (Figure 1) and would thus be preferred under moderately saline conditions.

3.3. Mean fruit weight

The fruit weights (g/fruit) are presented in Table 2 along with the statistical analyses. As with total yield, there were no significant differences between fruit weights in relation to water type; thus, the subsequent analysis utilized the means of both water types for grafted and nongrafted data. Salinity ($P < 0.001$), grafting ($P < 0.001$), and the interaction of salinity and grafting were all significant ($P < 0.05$), as shown in Table 2. Consistent with the trend in yield with salinity, mean fresh fruit weight also significantly decreased with increasing salinity relative to the control, starting at the −0.30 MPa OP level (Table 2). The fruit weight differences between the grafted and nongrafted plants were 7.5%, 13.0%, 24.5%, 6.0%, and 13.0% for the control, −0.15, −0.30, −0.45, and −0.60 MPa OP level treatments, respectively. In all instances, the fruit weights of the grafted plants were greater than the fruit weights of the nongrafted plants. Fruit weight differences (grafted vs. nongrafted) were not statistically significant for the last two OP levels ($P > 0.05$), which is again similar to the differences seen in total yield (discussed above). These results confirm that the yield decrease with increasing salinity is mainly caused by the decrease in fruit weight. There was a continuous decrease in fruit weight with increasing salinity for both grafted and nongrafted plants. Our results under nonsaline conditions are consistent with those of Zhao and Simonne (2008); they reported an overall increase in yield for grafted as compared to nongrafted tomato plants under nonsaline conditions.

3.4. Total soluble solid content

There were no significant differences in TSS content from irrigation with the two different water types (Table 2). Subsequently, we combined the data from the two water types for further analysis. The TSS content of the fruit increased with increasing salinity for both grafted and nongrafted treatments (expressed as brix% in Table 2). While increased TSS is desirable in terms of quality considerations, it is nonetheless an indicator of plant stress. Statistical analyses showed that salinity ($P < 0.001$) and grafting treatment ($P < 0.05$) were statistically significant for TSS content (Table 2). These results indicate that increasing salinity significantly increases the TSS of the tomato juice extracted from the fruit. The TSS of the grafted plants increased from 3.4% in the control to 7.1% under the highest salinity treatment, and for nongrafted plants, from 3.8% to 8.0%. TSS was higher for nongrafted plants at all but one salinity level, but significantly different concentrations between the grafted and nongrafted plants were determined only at the highest salinity treatment (Table 2). Flores et al. (2010) determined that TSS increased under salinity for tomato (Moneymaker) grafted onto Radja rootstock while Moneymaker grafted onto itself showed a slight decrease in TSS with salinity despite being less salt tolerant, and De Gioia et al. (2013) found no effect of grafting on TSS, but at relatively low salinity.

Increasing TSS in tomato with increasing irrigation water salinity, as we observed, is consistent with the findings of many earlier studies (Mizrahi et al., 1988; Dorais et al., 2000; Yurtseven et al., 2005; among others). Our data indicate that moderate salinity increases can achieve the desired increase in TSS without yield loss for nongrafted plants.

3.5. Leaf ion composition

Plant response to salinity can be evaluated by consideration of the change in plant ion composition with increasing salinity, as ion toxicity is a very important component of plant salt tolerance (Munns and Tester, 2008), especially in photosynthesizing leaf tissue. There were no statistically significant differences in the Na^+ content of the leaves of the grafted and nongrafted plants in relation to irrigation water composition (Table 3). Differences in leaf Na^+ as related to water composition were not expected because the irrigation waters types had comparable Na^+ concentrations at each salinity level.

The data in Table 3 indicate that Na^+ uptake increased with increasing salinity as expected for both grafted and nongrafted plants, but, more importantly, grafted plants had much less Na^+ uptake into the leaves as compared to nongrafted plants. The differences in Na^+ leaf content were statistically significant at the –0.15, –0.45, and –0.60 MPa osmotic levels (Table 3). Regulation of Na^+, either by exclusion at the root interface or by restriction of Na^+ translocation from the roots to the leaves, is a mechanism attributed to increased salt tolerance. In this instance, the data indicate that the Maxifort rootstock excluded Na^+ better than the nongrafted Big Dena even under nonsaline conditions. The decreased Na^+ in the grafted plants as compared to the nongrafted plants is consistent with the plant response, specifically increased yield, increased fruit size, and decreased TSS. These data indicate that increased Na^+ concentrations have a role in the response of tomato to salt stress. Earlier, Estan et al. (2005) for tomato, Goreta et al. (2008) for watermelon, Huang et al. (2009) for cucumber, and Zhu et al. (2008) for cucumber all reported that their grafted plants resulted in reduced Na^+ concentrations in shoot or fruit under salt stress. De Gioia at al. (2013) reported a decrease in Na^+ content of shoot and fruit for grafted tomato.

Grafted and nongrafted plants had comparable Cl^- leaf concentrations, as shown in Table 3 (only one salinity level was statistically significantly different). These data indicate that Cl^- accumulation does not explain the greater yield of grafted tomato plants as compared to nongrafted plants under saline conditions. Furthermore, the grafted plants (with Maxifort rootstock) did not exclude Cl^- more efficiently than the nongrafted plants. These results are in contrast to those reported earlier for other grafting combinations of Jaguar cv tomato (Estan et al., 2005), watermelon (Goreta et al., 2008), and cucumber (Huang et al., 2009). They all reported that grafting decreased Cl^- (as well as decreased Na^+ and higher yields) as compared to nongrafted under salt stress. Since both Cl^- and Na^+ were lower in the grafted plants in their studies, they were not able to determine if Na^+, Cl^-, or both were the critical growth-limiting ion. In contrast, our studies had similar leaf Cl^- and differing leaf Na^+ levels when comparing grafted and nongrafted plants.

There was, as expected, a statistically significant ($P < 0.001$) difference between the leaf Cl^- content of plants grown in Cl^- type waters and those grown in SO_4^{2-}–Cl^- type waters for all salinity treatments. The leaf Cl^- contents of the controls were not significantly different, and both water type controls had equal Cl^- content in the irrigation water. Since the tomato yields from the treatments irrigated with the Cl^- as compared to the SO_4^{2-}–Cl^- type waters were not

Table 3. Leaf ion analysis.

	Osmotic potential (MPa)	–0.003	–0.15	–0.30	–0.45	–0.60
Leaf Ca, mmol^{-1} kg	Grafted Cl	1765.67	2044.33	2317.33	2262.67	2573.00
	Nongrafted Cl	1794.00	2012.67	2267.33	2116.67	2447.00
	Average (Cl)	1779.8 aC	2028.5 aBC	2292.3 aAB	2189.7 aB	2510.0 aA
	Grafted SO_4	1754.67	2099.00	2083.67	1720.67	1645.67
	Nongrafted SO_4	1866.33	2032.00	2017.33	1474.33	1561.33
	Average (SO_4)	1810.5 aAB	2065.5 aA	2050.5 bA	1597.5 bB	1603.5b B
	↓ a, → A, Salinity P < 0.001, Salt P < 0.001, Interaction (Salt × Salinity) P < 0.001					
Leaf Mg, mmol^{-1} kg	Grafted Cl	441.00	478.33	229.33	263.37	305.67
	Nongrafted Cl	518.33	660.00	362.67	379.67	414.33
	Average (Cl)	479.7 aA	569.2 aA	296.0 aAB	321.7 bAB	360.0 bAB
	Grafted SO_4	408.67	332.67	640.33	592.00	872.00
	Nongrafted SO_4	614.67	460.67	571.67	770.67	947.67
	Average (SO_4)	511.7 aBC	396.7 bC	606.0 aB	681.3 aB	909.3 aA
	↓ a, → A, Salinity P < 0.001, Salt P < 0.001, Interaction (Salt × Salinity) P < 0.001					
Leaf Na, mmol^{-1} kg	Grafted Cl	30.50	71.03	108.87	175.33	167.67
	Grafted SO_4	29.83	74.43	77.50	177.67	134.33
	Average (grafted)	30.2 aCB	73.2 bB	93.2 aB	176.5 bA	151.0 bA
	Nongrafted Cl	38.77	112.97	137.67	250.33	228.67
	Nongrafted SO_4	36.57	109.03	93.23	331.00	126.67
	Average (nongrafted)	37.7 aD	111.0 aC	115.5 aC	290.7 aA	177.7 aB
	↓ a, → A, Salinity P < 0.001, Grafting P < 0.001, Interaction (Grafting × Salinity) P < 0.001					
Leaf K, mmol^{-1} kg	Grafted Cl	746.67	662.33	682.67	713.00	627.00
	Nongrafted Cl	674.67	631.00	676.67	542.00	402.00
	Average (Cl)	710.7 aA	646.7 aA	679.7 aA	627.5 bA	514.5 aB
	Grafted SO_4	854.00	710.67	618.00	898.00	614.00
	Nongrafted SO_4	737.00	643.67	712.00	890.00	589.33
	Average (SO_4)	795.5 aA	677.2 aAB	665.0 aAB	894.0 aA	601.7 aB
	↓ a, → A, Salinity P < 0.001, Salt P < 0.001, Interaction (Salt × Salinity) P < 0.001					
Leaf P, mmol^{-1} kg	Grafted Cl	335.67	284.33	88.83	76.53	91.13
	Nongrafted Cl	263.67	159.67	52.83	54.87	66.33
	Average (Cl)	299.7 aA	222.0 aB	70.8 bC	65.7 bC	78.7 bC
	Grafted SO_4	388.67	186.00	155.00	162.00	216.33
	Nongrafted SO_4	294.33	84.77	210.00	93.20	148.10
	Average (SO_4)	341.5 aA	135.4 aB	182.6 aB	127.6 aB	182.2 aB
	Average (grafted)	362.2 aA	235.2 aB	121.9 aC	119.3 aC	153.7 aC
	Average (nongrafted)	279.0 bA	122.2 bB	131.4 aB	74.0 aB	107.2 aB
	↓ a, → A, Salinity P < 0.001, Grafting P < 0.001, Interaction (Grafting × Salinity) P < 0.001, (Salt × Salinity) P < 0.001					
Leaf S, mmol^{-1} kg	Grafted Cl	513.67	721.00	597.33	428.67	486.00
	Nongrafted Cl	437.00	671.67	495.67	429.00	407.66
	Average (Cl)	475.3 aB	696.3 aA	546.5 bB	428.8 bB	446.8 bB
	Grafted SO_4	586.00	575.33	696.67	759.00	637.00
	Nongrafted SO_4	454.00	542.33	729.00	649.67	520.67
	Average (SO_4)	520.0 aB	558.8 bB	712.8 aA	704.3 aA	578.8 bA
	↓ a, → A, Salinity P < 0.001, Salt P < 0.001, Interaction (Salt × Salinity) P < 0.001					
Leaf Cl, mmol^{-1} kg	Grafted Cl	521.00	964.33	1516.00	1919.67	1992.00
	Nongrafted Cl	364.33	852.33	1433.33	2046.33	1802.00
	Average (Cl)	442.7 aD	908.3 aC	1474.7 aB	1983.0 aA	1897.0 aA
	Grafted SO_4	449.00	608.67	689.00	1276.00	1159.00
	Nongrafted SO_4	337.00	458.00	752.00	1590.33	1185.33
	Average (SO_4)	393.0 aE	533.3 bDE	720.5 bCD	1433.2 bA	1172.2 bB
	Average (grafted)	485.0 aD	786.5 aC	1102.5 aB	1597.8 bA	1575.5 aA
	Average (nongrafted)	350.7 aE	655.2 aD	1092.7 aC	1818.3 aA	1493.7 aB
	↓ a, → A, Salinity P < 0.001, Grafting P < 0.001, Interaction (Grafting × Salinity) P < 0.001, (Salt × Salinity) P < 0.001					

Different lower case letters denote statistical significance among columns (salinity treatments), and upper case letters refer to statistical significance between rows (either grafted and nongrafted or chloride and sulfate–chloride waters).

Table 4. Fruit ion analysis.

	Osmotic Potential (MPa)	–0.003	–0.15	–0.30	–0.45	–0.60
Fruit Ca, mmol^{-1} kg	Grafted Cl	57.37	51.40	54.77	35.80	38.10
	Nongrafted Cl	59.73	54.33	58.5	38.63	19.23
	Average (Cl)	58.55Ab	52.87 Aa	56.63 Aa	37.22 Ba	28.67 Ba
	Grafted SO$_4$	74.97	57.57	45.63	28.90	20.8
	Nongrafted SO$_4$	68.23	63.37	45.63	28.90	20.83
	Average (SO$_4$)	71.60 Aa	60.47 Ba	45.32 Cb	35.95 CDa	24.57 Da
	Average (salinity)	65.08 A	56.67AB	50.98 B	36.58 C	26.62 D
	↓ a, → A, Salinity $P < 0.001$, Interaction (Salt × Salinity) $P < 0.01$					
Fruit Mg, mmol^{-1} kg	Grafted Cl	79.60	61.37	47.97	43.60	45.33
	Nongrafted Cl	73.40	57.23	49.50	38.87	40.57
	Grafted SO$_4$	80.73	67.60	63.93	55.20	54.17
	Nongrafted SO$_4$	87.60	73.93	66.70	46.40	51.80
	Average (salinity)	80.33 A	65.03 B	57.03 B	45.52 C	48.00 C
	→ A, Salinity $P < 0.001$					
Fruit Na, mmol^{-1} kg	Grafted Cl	27.87	37.37	40.90	40.03	37.40
	Nongrafted Cl	27.07	39.80	42.53	41.13	46.73
	Grafted SO$_4$	27.27	34.13	41.73	35.33	39.13
	Nongrafted SO$_4$	28.63	34.43	43.00	45.20	46.50
	Average (salinity)	27.71 B	36.43 A	42.04 A	40.43 A	42.44 A
	→ A, Salinity $P < 0.001$					
Fruit K, mmol^{-1} kg	Grafted Cl	1210	1235	1006	968	984
	Nongrafted Cl	1094	1081	1008	826	881
	Average (Cl)	1152 bA	1158 aA	1007 aAB	897 aB	933 aB
	Grafted SO$_4$	1534	1231	1122	1038	1018
	Nongrafted SO$_4$	1299	1244	1052	861	870
	Average (SO$_4$)	1417 aA	1238 aB	1088 aBC	950 aBC	944 aBC
	Average (salinity)	1284 A	1198 B	1047 C	923 D	938 CD
	↓ a, → A, Salinity $P < 0.001$, Salt $P < 0.001$, Interaction (Salt × Salinity) $P < 0.05$					
Fruit P, mmol^{-1} kg	Grafted Cl	177.3	145.7	10.00	124.00	102.30
	Nongrafted Cl	153.33	15.00	107.43	101.27	102.30
	Average (Cl)	165.33 bA	130.3 bB	106.22 aB	112.63aB	102.17 aB
	Grafted SO$_4$	232.33	147.33	112.67	121.33	125.67
	Nongrafted SO$_4$	178.33	154.33	107.13	93.83	103.67
	Average (SO$_4$)	205.33 aA	150.83 aB	109.90 aC	107.58 aC	114.67 aC
	Average (salinity)	185.33 A	140.58 B	108.06 C	110.11 C	108.42 C
	↓ a, → A, Salinity $P < 0.001$, Salt $P < 0.005$, Grafting $P < 0.001$, Interaction (Salt × Salinity) $P < 0.05$					
Fruit S, mmol^{-1} kg	Grafted Cl	61.27	54.30	54.93	46.37	43.90
	Nongrafted Cl	61.27	54.30	54.93	46.37	43.90
	Average (Cl)	165.33 bA	130.33 bB	106.22 aB	112.63 aB	102.17 aB
	Grafted SO$_4$	77.23	65.60	58.23	61.17	50.03
	Nongrafted SO$_4$	71.90	67.50	57.87	46.67	46.47
	Average (SO$_4$)	74.57 aA	66.55 aB	58.05 aC	53.42 aCD	48.25 aD
	Average (salinity)	68.36 A	62.23 B	56.25 C	51.01 D	47.41 D
	↓ a, → A, Salinity $P < 0.001$, Salt $P < 0.005$, Grafting $P < 0.001$, Interaction (Salt × Salinity) $P < 0.05$					
Fruit Cl, mmol^{-1} kg	Grafted Cl	167.33	250.67	250.00	330.67	282.00
	Nongrafted Cl	133.33	229.00	263.67	223.33	243.33
	Average (Cl)	150.33 aB	239.83 aA	256.83 aA	277.00 aA	262.67 aA
	Grafted SO$_4$	215.33	190.00	205.00	217.33	212.33
	Nongrafted SO$_4$	161.00	207.00	193.33	194.33	196.33
	Average (SO$_4$)	188.17 aA	198.5b A	199.17 bA	205.83 bA	204.33 bA
	Average (salinity)	169.25 B	219.17 A	228.00 A	241.42 A	233.50 A
	↓ a, → A, Salinity $P < 0.001$, Salt $P < 0.005$, Grafting $P < 0.001$, Interaction (Salt × Salinity) $P < 0.05$					

Different lower case letters denote statistical significance among columns (salinity treatments), and upper case letters refer to statistical significance between rows (either grafted and nongrafted or chloride and sulfate–chloride waters).

significantly different despite the significant differences in Cl^- leaf content, these data provide additional support for the conclusion that Cl^- toxicity is not a factor limiting tomato yield under saline conditions.

Potassium is a critical element for plant growth. Yield reductions with increased salinity are often associated with corresponding reductions in K^+ plant organ content or reductions in leaf Na^+/K^+ ratios. There was, as expected, a general trend of decreasing leaf K^+ content with increasing irrigation water salinity, as shown in Table 3. There were no significant differences between the grafted and nongrafted plants, but the grafted plants had slightly higher K^+ concentrations (Table 3), perhaps related to their lower Na^+ values (K^+/Na^+ selectivity). There were also no significant differences between the K^+ content of leaves in treatments irrigated with the two different water types at all salinity levels except –0.45 MPa OP. Earlier studies have also noted a relationship between increased shoot or fruit K^+ in grafted plants when compared to nongrafted plants and related this to yield under NaCl salt stress, but they also observed decreased Na^+ and Cl^- in the grafted plants (Estan et al., 2005; Goreta et al., 2008; Huang et al., 2009). However, they could not readily separate the roles of these two ions in impacting salt tolerance as they used one salt type, NaCl, in their studies.

Significant differences in P leaf composition were recorded for the treatments, as shown in Table 3. The grafted plants had larger P concentrations as compared to the nongrafted plants, suggesting more efficient P root uptake or transport within the grafted plants. This may be related to the increased vigor of the Maxifort rootstock. Additionally, the leaf P concentrations were significantly greater in the treatments irrigated with the mixed SO_4^{2-}–Cl^- type waters as compared to the Cl^- waters for osmotic potentials greater than –0.15 MPa. These data are matched by the elevated leaf Ca^{2+} concentrations resulting from the elevated Ca^{2+} in the Cl^- type irrigation waters, as compared to the Ca^{2+} in the SO_4^{2-}–Cl^- type waters, also at osmotic pressures above –0.15 MPa (statistically significant, $P < 0.05$). The inverse correlation between leaf Ca^{2+} and leaf P may be explained by suppression of P transport under elevated plant Ca^{2+} for the treatments that used Cl^- water compositions. Phosphorus concentrations in leaves were substantially below the 100–180 mmol kg^{-1} P level considered optimal for plant growth (Marschner, 1995) for several salinity levels of the Cl^- type waters.

3.6. Fruit ion composition

Fruit ion composition data are presented in Table 4. There were statistically significant differences in fruit Ca concentrations obtained from use of both water types at high salinity (Table 4), explained by the increased Ca in the irrigation water of the Cl^- type water. Mg concentrations were greater in the fruit grown with the SO_4^{2-}–Cl^- water versus the Cl^- water, especially at high salinity. This is explained by the increased Mg content of the SO_4^{2-}–Cl^- irrigation water compared to the Cl^- water, especially at high salinity (Table 1). Grafted plants also resulted in lower Mg in the fruit as compared to nongrafted plants (Table 4). There were no statistical differences in Na content of the fruit, either with increasing salinity, different water types, or grafted versus nongrafted (Table 4). This is in contrast to the leaf data where Big Dena grafted on Maxifort rootstock had lower leaf Na relative to nongrafted plants. The grafted plants had higher fruit K and higher fruit P for both water types when compared to the nongrafted plants (Table 4), consistent with the leaf K and P data discussed earlier.

3.7. Conclusions

The grafted tomato plants exhibited increased yield both under control and elevated salinity conditions relative to the nongrafted plants. In contrast to the absolute yield relationships, expression of salt tolerance in terms of relative yield provides the conclusion that nongrafted plants are slightly more salt-tolerant than grafted plants. Our data indicate that, for tomato, decreased yield under saline conditions is well related to increased leaf Na^+ concentrations. The grafted plants also had reduced leaf Na^+ contents relative to the nongrafted plants at all salinity levels. Grafting using Maxifort rootstock is thus an option to increase tomato yield regardless of irrigation water salinity. Increased irrigation water Cl^- was not related to changes in fruit yield, suggesting that yield reduction from Cl^- toxicity is not a consideration for tomato. Additionally, the grafted Big Dena on Maxifort rootstock plants had an increased yield but slightly higher leaf Cl^- concentrations as compared to the nongrafted Big Dena tomato plants, again indicating that Cl^- ion concentration was not a factor in yield loss under saline conditions.

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