

## EFFECTS OF SALINE WATER ON TOMATO UNDER SUBSURFACE DRIP IRRIGATION: NUTRITIONAL AND FOLIAR ASPECTS

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### ABSTRACT

A field experiment on the effects of drip irrigation (DI) and subsurface drip irrigation (SDI) with saline water (6.57 dS m<sup>-1</sup>) on three tomato cultivars (*Lycopersicon esculentum* Mill., cvs. Río Tinto, Río Grande and Nemador) was carried out with the purpose to quantify physiological responses. The aim was to improve irrigation water management under saline conditions of Tunisia. The trial was established in a silt-clayey soil with three regimes of irrigation: 100 %, 85 % and 70 % of crop water requirement. Results evidenced a significant difference between the two irrigation systems for the three cultivars. Growth parameters such as leaf area, chlorophyll content and mineral composition of leaves, petioles, stems and roots were affected significantly by the different treatments, particularly for Río Tinto and Nemador, being Río Grande the more adapted. The fruit was the organ less affected. Strong accumulation of Na<sup>+</sup> and Cl<sup>-</sup> accompanied a reduction in Ca<sup>2+</sup>, K<sup>+</sup>, Mg<sup>2+</sup> and P content in the case of DI. The distribution of these last necessary elements for plants nutrition under a strong accumulation of Na<sup>+</sup> and Cl<sup>-</sup> depends on the cultivar and changes from one organ to another. SDI can be included as an effective option for tomato production in Tunisia.

**Keywords:** Salt tolerance, tomato, localized irrigation, deficit irrigation, Tunisia.

### INTRODUCTION

Soil and water salinity in the arid regions are continuously increasing (Rus *et al.*, 2002). Globally, more than 770,000 km<sup>2</sup> of the lands are affected by secondary salinization, 20 % of the irrigated areas and about 2 % of the agricultural lands (FAO, 2000). In Tunisia, soils affected by salts cover about 1.5 million hectares, around 10 % of the total country area. About 30 % of irrigated areas are affected by salts in different degrees (Hachicha,

2007). Salinity is a major abiotic factor limiting plant growth and fruit yield (Parada *et al.*, 2006). It induces osmotic and toxic effects leading physiological, morphological and biochemical modifications; it causes growth inhibition, crop yield reduction, lower photosynthesis and respiration, nutritional deficiencies and inhibition of protein synthesis (Ashraf and Foolad, 2007). These phenomena have been observed in

agricultural and horticultural crops, including tomato (Juan *et al.*, 2005).

In order to overcome salinity problem, several works have been accomplished for selecting tolerant cultivars to abiotic constraints (De Malach and Pasternak, 1993; Claudivan *et al.*, 2005; Ouled Ahmad *et al.*, 2007). Juan *et al.* (2005) studying ten cultivars of tomato showed that Jaguar and Brilliant cultivars were the most tolerant to salt stress; they were characterized by a reduction of the uptake and accumulation of toxic ions in leaves. In the same way, Romero-Aranda *et al.* (2001) studied the behaviour of two cultivars of tomato under salt stress condition and showed that Daniella cultivar appears more adapted to salinity, based on total dry matter and leaf area production. In addition, it presents higher Na<sup>+</sup> content than Moneymaker cultivar, which allowed authors to conclude that Daniella cultivar possesses a leaf tissue more tolerant to the toxic ions accumulation. Thus, restriction of toxic ion uptake is an important adaptation character contributing to improved glycophytes tolerance to salinity (Romero-Aranda *et al.*, 2001).

Besides this physiological dimension, several studies have been conducted for development of irrigation systems for salinity management with drip irrigation (DI) using saline water (Meiri *et al.*, 1992; Simsek *et al.*, 2004; Dagdelen *et al.*, 2008). According to Cetin and Bilgel (2002), the DI permits an uniform and frequent application of water and a direct feeding of the plant at the root zone level, leading an increase of yield and saving water (Bozkurt *et al.*, 2006; Sezen *et al.*, 2008). According to Wang *et al.* (2007), DI improves tomato yield and reduces leaf burn (browning). However, this system may result in localized

accumulation of salts at the soil surface (Ayers and Westcot, 1985) due to increased evaporation. According to Hachicha *et al.* (2006), salt accumulates on the soil surface before migrate and reach the root zone when DI irrigation is used. Subsurface drip irrigation (SDI) has been developed to improve salinity management and water use efficiency. According to Phene *et al.* (1991) and Oron *et al.* (1998), SDI decreases the accumulation of salts at the root zone level of plants, producing an improved yield and fruit quality. This has been observed in tomato (Ayars *et al.*, 2001; Hanson *et al.*, 2004), onion (Enciso *et al.*, 2007), cotton (Detar, 2007), bean (Gençoglan *et al.*, 2006), potato (Patel and Rajput, 2008) and corn (Payero *et al.*, 2008). SDI improves water use efficiency (Camp *et al.*, 1998; Enciso *et al.*, 2005; Patel and Pandey, 2008; Hassanli *et al.*, 2008). Indeed, it permits the application of small amounts of water directly at the root zone level of the plant and the maintenance of high soil moisture in this area (Enciso *et al.*, 2007). Camp (1998) obtained improved yields when they irrigated more than thirty crops by different methods, including DI and SDI. According to studies of Hassanli *et al.* (2008) on corn irrigated by DI, SDI and sprinkler irrigation, SDI permits a better yield compared to other irrigation methods. Beside a reduction of evaporation, SDI increases leaves transpiration which improves the stomata opening and photosynthetic activity.

The purpose of this research was to evaluate the adaptive responses of three open-field tomato cultivars to different irrigation regimes and water location systems, and determine the more tolerant cultivar under the same conditions for further research.

## MATERIALS AND METHODS

### Plant material

The experiment was carried out in summer 2007 (from May to August). Three cultivars of tomato (*Lycopersicon esculentum* Mill.) were used: Río Grande, Río Tinto and Nemador. Río Grande is a cultivar adapted to mechanical harvest. It is characterized by a determined growth, a middle size canopy assuring a good cover of fruits (oblong, smooth, thick, 80 g, fruits very firm and having good acidity). The cultivar Río Tinto is characterized by a determined growth and red fruits slightly flattened of 100 to 180 g. Finally, Nemador possesses a determined growth, of round red fruits. Seeds were provided by the Laboratory of Seeds and Plant Control of the General Direction of Protection and Control of the Agricultural Production and Quality, Tunisia. These three cultivars were selected previously through a screening of 20 cultivars tested in germination experiments under saline condition.

### Experimental design

The experiment was located at the Cherfech Agricultural Experimental Station, 25 km north of Tunis in the Low Valley of Mejerda River. Climate of the region is Mediterranean with an annual rainfall close to 470 mm and an average yearly evapotranspiration of 1370 mm (Penman method). The experiment was set using emitters with filters and buried at 30 cm depth in SDI. Transplanting date was 2/05/2007 in single lines, using seedlings at the 5 - 7 leaves stage and according to a factorial design. Tomato plants were spaced 1 m between rows and 0.40 m between plants. The plot was divided in three areas corresponding to 3 regimes of irrigation water. Within each

parcel, plants were distributed in six rows, three by treatment; each line included 30 plants. Lines 1, 3 and 5 corresponded to the DI treatment, whereas lines 2, 4 and 6 corresponded to the SDI treatment.

### Irrigation water

Irrigation water came from a well. Its pH is 7.45; electrical conductivity (EC) is about 6.57 dS m<sup>-1</sup> and the sodium adsorption ratio (SAR) equals to 14. It is rich in NaCl (Na<sup>+</sup> = 35.65 me L<sup>-1</sup>, Cl<sup>-</sup> = 34.21 me. L<sup>-1</sup>, K<sup>+</sup> = 3.96; Ca<sup>2+</sup> = 7.73 me. L<sup>-1</sup> and Mg<sup>2+</sup> = 4.83 me L<sup>-1</sup>). Three regimes of water were applied: 100%, 85% and 70% of crop water requirement, according to climatic data (about 1434 mm). As well known there is an effect on soil and crop, according to the concentration of NaCl in water. The EC of the well water is the half of the range for the tomato crop, meaning that tomato would be over the tolerance threshold and probably affected up to 49% of yield reduction (Mass and Hoffman, 1977).

### Pigment extraction

Total Chlorophyll (Total Chl), Chlorophyll a (Chl a), and Chlorophyll b (Chl b) contents were determined according to the method of Torrecillas *et al.* (1984). Leaf tissues (200 mg) were kept in 5 ml of acetone at 4°C in dark. After 72 h the optic density of the extract was measured at 665 nm and 649 nm. Total Chl, Chl-a and Chl-b were calculated, according to the following equations and expressed as mg.g<sup>-1</sup> fresh weight (FW).

$$\text{Chlorophyll a} = 11.63 \times (\text{OD}_{665}) - 2.39 \times (\text{OD}_{649})$$

$$\text{Chlorophyll b} = 20.11 \times (\text{OD}_{649}) - 5.18 \times (\text{OD}_{665})$$

$$\text{Total chlorophyll} = 6.45 \times (\text{OD}_{665}) + 17.72 \times (\text{OD}_{649})$$

### Measurement of leaf area

It was measured with a planimeter type Delta-T Devices Ltd. For every plant, leaves were detached and introduced in the device. Leaf area was expressed in  $\text{cm}^2$ .

### Analysis of $\text{Na}^+$ , $\text{K}^+$ , $\text{Ca}^{2+}$ , $\text{Mg}^{2+}$ , $\text{Cl}^-$ , P in plant tissues

For chemical analyses tissues of leaf, root and fruit were used. The organic ions were extracted from dry matter by  $\text{HNO}_3$  at room temperature for 48 h.  $\text{K}^+$  and  $\text{Na}^+$  were analyzed by flame emission, using a Eppendorf spectrophotometer (JENWAY PFP7).  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  were determined by atomic absorption spectrophotometry.  $\text{Cl}^-$  was quantified by colorimetry with a Buchler chloridometer. Phosphorus was estimated by the chlorostannous molybdophosphoric blue colour method (Gericke and Kurmies, 1952).

### Soil analyses

Soil salinity was determined before the trial set and at the end of the crop cycle, every 20 - 40 cm depth at the root level. The soil/water ratio was 1/5. A significant linear relationship permitted to convert the electrical conductivity EC (1/5) to the electrical conductivity of the saturated paste extract (ECe) (Jammazi and Hachicha, 2002):

$$\text{ECe} = 5.853 * \text{EC} (1/5) - 0.262$$

$$(n = 134 \text{ and } R^2 = 0.91)$$

$$\text{with } 1.2 < \text{ECe} < 8.3 \text{ dS m}^{-1} \text{ and } 0.2 < \text{EC} (1/5) < 1.4 \text{ dS m}^{-1}$$

### Statistical analysis

Statistical processing was achieved by the software STATISTICA, Version 5 (Statsoft France, 1997). All the

parameters recorded were subjected to a factorial analysis of variance with three factors (cultivars, irrigation system and irrigation regime). Means comparison were carried out by the LSD test at the significant level of 0.05.

## RESULTS

### Effects on soil

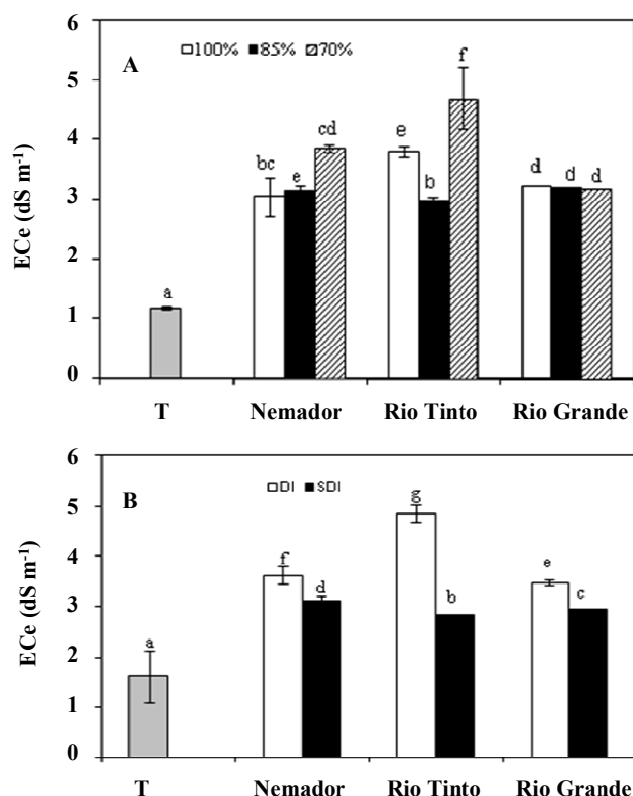
Figure 1 shows that soil salinity varied according to the irrigation regime. ECe was generally higher than the control treatment (100 % water requirement) for the 70% water regime (Fig. 1 A). The increase was highly significant ( $p \leq 0.05$ ) in the case of Río Tinto cultivar. The irrigation system had a significant effect on the soil salinity: ECe was significantly higher ( $p \leq 0.05$ ) in DI than in SDI, particularly in the case of Río Tinto (Figure 1 B).

### Effects on tomato crop

#### Growth effects

As seen in Table 1, highly significant ( $p \leq 0.01$ ) reduction of leaf area on the three cultivars with the regime 70% of water requirement occurred. The reduction in this lower regime treatment was 16.81%, 37.57% and 55.85% respectively for Río Grande, Río Tinto and Nemador compared to the control treatment (100 % of water requirement). The effect of the irrigation system was also significant ( $p \leq 0.01$ ) on the three cultivars. The leaf area was higher in Río Grande than in the other cultivars and in SDI than DI (Table 2).

The high chlorophyll content (Chl a, Chl b and total Chl) in the three cultivars confirmed the visual observation at lower amount of water applied (Table 1). The



**Figure 1.** Variation on electric conductivity (EC) of soil according to (A) three regimes of irrigation (100 %, 85 % and 70 % of water requirement and (B) two irrigation systems, drip irrigation (DI) and subsurface drip irrigation (SDI), for three tomato cultivars (Río Grande, Río Tinto and Nemador). Different letters indicate significant differences at  $p \leq 0.05$ . T= control.

total chlorophyll (a + b) was increased significantly from 0.54, 0.47 and 0.45  $\text{mg.g}^{-1}$  FW in the control (100% of water requirement) to 0.80, 0.66 and 0.69  $\text{mg.g}^{-1}$  FW in the more deficit water treatment (70% of water requirement), respectively for Río Grande, Río Tinto and Nemador. This significant increase ( $p \leq 0.01$ ) in chlorophyll (a + b) was also observed in the case of SDI in relation to DI for the three cultivars (Table 2).

#### *Effects on mineral nutrition*

The presence of salt in the irrigation water affected significantly ( $p \leq 0.05$ ) all organs of the three cultivars, where a higher content of  $\text{Na}^+$  in leaves of Río Grande, petioles of Río Tinto and stems and roots of the three cultivars, particularly Nemador was observed (Figure 2A). By comparing the distribution of this element in different plant organs, data showed that

**Table 1.** Effect of saline water (6.57 dS m<sup>-1</sup>) on Chl a, Chl b and Total Chl (mg g<sup>-1</sup> FW) and leaf area (cm<sup>2</sup>) of three tomato cultivars (Río Grande, Río Tinto and Nemador) under three irrigation regimes (100%, 85% and 70% of water requirement) after 117 days post transplanting.

Treatment	Chla	Chl b	Total chl	Leaf area
	-----mg g <sup>-1</sup> -----			cm <sup>2</sup>
<b>Río Grande</b>				
100%	0.41 ± 0.018 <b>g</b>	0.13 ± 0.016 <b>d</b>	0.54 ± 0.037 <b>c</b>	182.87 ± 3.70 <b>f</b>
85%	0.32 ± 0.011 <b>a</b>	0.08 ± 0.091 <b>a</b>	0.40 ± 0.103 <b>a</b>	213.03 ± 15.12 <b>g</b>
70%	0.45 ± 0.035 <b>h</b>	0.35 ± 0.016 <b>h</b>	0.80 ± 0.058 <b>g</b>	153.46 ± 18.87 <b>cd</b>
<b>Río Tinto</b>				
100%	0.37 ± 0.009 <b>d</b>	0.10 ± 0.004 <b>c</b>	0.47 ± 0.011 <b>b</b>	216.76 ± 20.26 <b>g</b>
85%	0.33 ± 0.004 <b>b</b>	0.28 ± 0.049 <b>e</b>	0.61 ± 0.044 <b>e</b>	213.03 ± 15.12 <b>g</b>
70%	0.36 ± 0.028 <b>cd</b>	0.33 ± 0.002 <b>g</b>	0.66 ± 0.040 <b>e</b>	134.85 ± 5.71 <b>b</b>
<b>Nemador</b>				
100%	0.37 ± 0.009 <b>e</b>	0.09 ± 0.011 <b>b</b>	0.45 ± 0.025 <b>b</b>	164.68 ± 16.56 <b>e</b>
85%	0.36 ± 0.016 <b>c</b>	0.27 ± 0.042 <b>e</b>	0.63 ± 0.058 <b>d</b>	148.87 ± 30.24 <b>c</b>
70%	0.39 ± 0.007 <b>f</b>	0.3 ± 0.018 <b>f</b>	0.69 ± 0.018 <b>f</b>	91.98 ± 24.59 <b>a</b>
Analysis of variance				
Effect of cultivar (C)	**	**	**	**
Effect of regime (R)	**	**	**	**
Interaction C X R	**	**	**	**

Within each column of the three cultivars of tomato all values are the mean of nine plants (n = 9) irrigated by different regimes. Mean ± SE with the different letters are significantly different at  $p \leq 0.05$  according to LSD test.

\*\* indicate significant differences at  $p \leq 0.01$  according to the analysis of variance.

Chl a: Chlorophyll a; Chl b: Chlorophyll b; Total Chl: Total Chlorophyll

fruits of the three cultivars appear to be less affected by the salinity of the irrigation water, particularly Río Tinto, whereas roots accumulated more Na<sup>+</sup> ions (Figure 2A). A significant ( $p \leq 0.05$ ) increase in roots was found, particularly in Nemador. However, Na<sup>+</sup> was significantly higher in DI than SDI and especially in the case of the 70 % water

treatment, for the three cultivars, particularly Río Grande cultivar (Tables 3 and 4). As shown in figure 2B, the Cl<sup>-</sup> content was increased significantly ( $p \leq 0.05$ ) in leaves, stems and particularly, in the petiole of the three cultivars and decreased significantly ( $p \leq 0.05$ ) in both roots and fruits for the three cultivars. Similar to the results obtained in Na<sup>+</sup>

**Table 2.** Effect of saline water ( $6.57 \text{ dS m}^{-1}$ ) on Chl a, Chl b and Total Chl ( $\text{mg g}^{-1}$  FW) and leaf area ( $\text{cm}^2$ ) of three tomato cultivars (Río Grande, Río Tinto and Nemador) irrigated by drip irrigation (DI) and subsurface drip irrigation (SDI) after 117 days post transplanting.

Treatment	Chl a	Chl b	Total Chl	Leaf area
	-----mg g <sup>-1</sup> -----			cm <sup>2</sup>
<b>DI</b>				
Río Grande	0.37 ± 0.021 <b>c</b>	0.18 ± 0.016 <b>a</b>	0.55 ± 0.004 <b>b</b>	173.8 ± 15.63 <b>d</b>
Río Tinto	0.29 ± 0.035 <b>a</b>	0.22 ± 0.011 <b>d</b>	0.51 ± 0.023 <b>a</b>	162.25 ± 7.47 <b>c</b>
Nemador	0.36 ± 0.014 <b>b</b>	0.21 ± 0.004 <b>c</b>	0.57 ± 0.018 <b>c</b>	119.00 ± 23.11 <b>a</b>
<b>SDI</b>				
Río Grande	0.42 ± 0.007 <b>e</b>	0.20 ± 0.018 <b>b</b>	0.62 ± 0.011 <b>d</b>	192.42 ± 13.05 <b>e</b>
Río Tinto	0.42 ± 0.007 <b>e</b>	0.25 ± 0.016 <b>f</b>	0.67 ± 0.023 <b>e</b>	178.14 ± 2.96 <b>d</b>
Nemador	0.39 ± 0.014 <b>d</b>	0.23 ± 0.002 <b>e</b>	0.62 ± 0.011 <b>d</b>	151.30 ± 16.01 <b>b</b>
Analysis of variance				
Effect of cultivar (C)	<b>**</b>	<b>*</b>	<b>*</b>	<b>**</b>
Effect of irrigation system (S )	<b>**</b>	<b>*</b>	<b>**</b>	<b>**</b>
Interaction C X S	<b>**</b>	<b>*</b>	<b>**</b>	<b>**</b>

Within each column of the three cultivars of tomato, all values are the mean of nine plants ( $n = 9$ ) irrigated by Drip irrigation (DI) and Subsurface drip irrigation (SDI). Mean  $\pm$  SE with different letters are significantly different at  $p \leq 0.05$  according to LSD test.

\*, \*\* indicate significant differences at  $p \leq 0.05$  and  $p \leq 0.01$  respectively according to the analysis of variance.

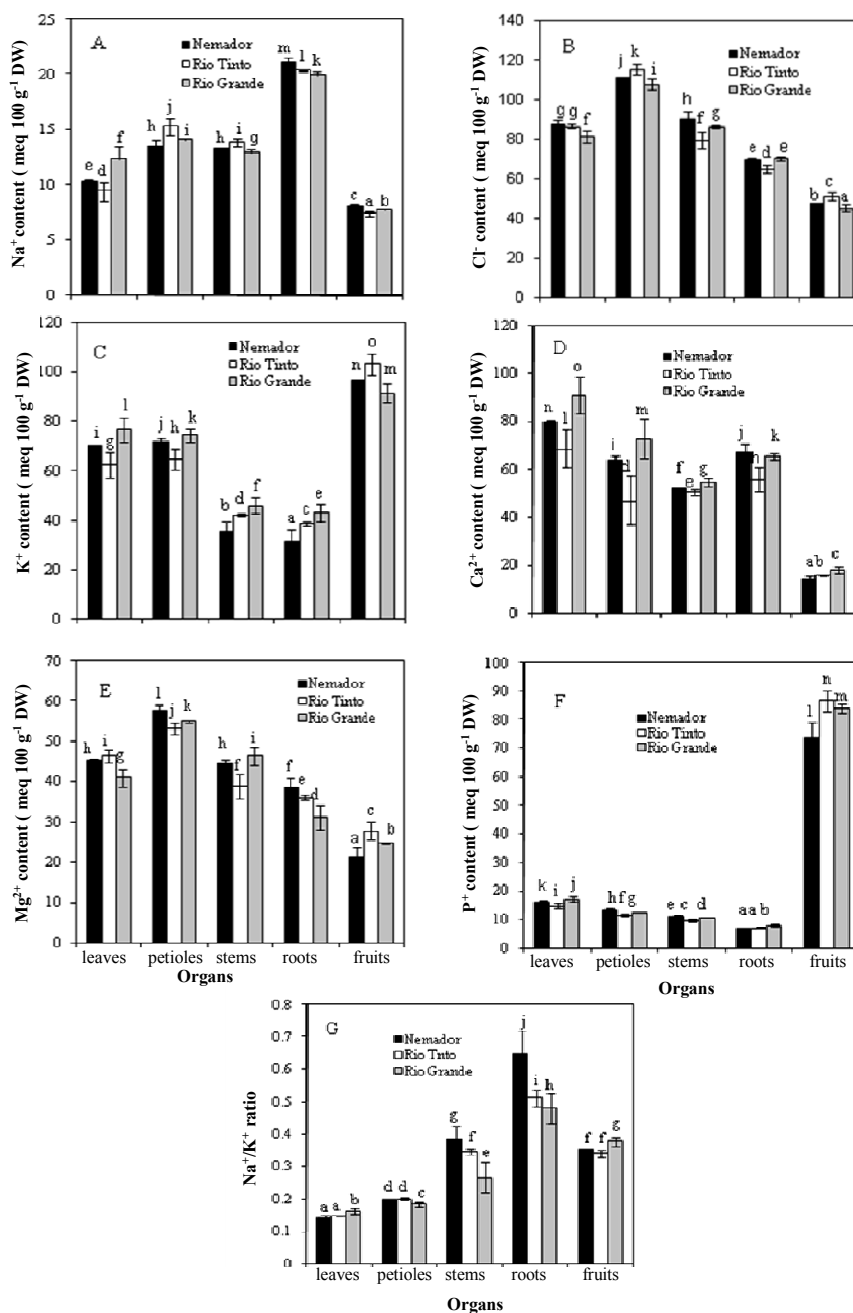
Chl a: Chlorophyll a; Chl b: Chlorophyll b; Total Chl: Total Chlorophyll

contents,  $\text{Cl}^-$  content is significantly higher ( $p \leq 0.01$ ) in the more deficit regime (70% of water requirement) compared with the control treatment (100% of water requirement) (Table 3) and also in the case of DI in relation to SDI for the three cultivars, particularly Nemador (Table 4).

At the same time,  $\text{K}^+$  content was significantly higher ( $p \leq 0.05$ ) in both leaves and petioles in Río Grande. The highest increase was found in fruits of the three cultivars (Figure 2C). As

shown in Table 3, the irrigation regime treatment significantly affected ( $p \leq 0.01$ )  $\text{K}^+$  ion.

The  $\text{K}^+$  content was lower in Río Tinto and Nemador when compared with Río Grande in the 70% of water requirement treatment and when compared with the control treatment (100% of water requirement). The irrigation system significantly affected ( $p \leq 0.01$ ) also  $\text{K}^+$  ion. This content was higher in SDI than in DI, particularly in Río Grande (Table 4).



**Figure 2.** Effect of saline water (6.57 dS m<sup>-1</sup>) on (A) Na<sup>+</sup>, (B) Cl<sup>-</sup>, (C) K<sup>+</sup>, (D) Ca<sup>2+</sup>, (E) Mg<sup>2+</sup>, (F) P<sup>+</sup> contents and (G) Na<sup>+</sup>/K<sup>+</sup> ratio of different organs of tomato cultivars (Río Grande, Río Tinto and Nemador). All values are the mean ± SE of nine plants (n = 9) and bars with different letters are significantly different at p ≤ 0.05 according to LSD test.



**Table 3.** Effect of saline water (6.57 dS m<sup>-1</sup>) on Na, Cl, K, Ca, Mg and P content (meq 100 g<sup>-1</sup> DW) and Na<sup>+</sup>/K<sup>+</sup> ratio of three tomato cultivars (Río Grande, Río Tinto and Nemador) under three regimes of irrigation (100%, 85% and 70% of water requirement).

Treatment	Na <sup>+</sup>	Cl <sup>-</sup>	K <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	P	Na <sup>+</sup> /K <sup>+</sup>
<b>Río Grande</b>							
100%	10.48 ± 0.252 <b>a</b>	74.05 ± 0.019 <b>a</b>	70.75 ± 3.015 <b>g</b>	76.68 ± 4.256 <b>h</b>	47.52 ± 3.318 <b>e</b>	25.65 ± 0.018 <b>c</b>	0.17 ± 0.013 <b>a</b>
85%	13.37 ± 0.188 <b>e</b>	79.72 ± 0.019 <b>cd</b>	64.92 ± 1.040 <b>f</b>	62.14 ± 2.800 <b>e</b>	42.67 ± 1.013 <b>d</b>	25.77 ± 0.154 <b>c</b>	0.31 ± 0.011 <b>d</b>
70%	16.27 ± 0.271 <b>h</b>	80.33 ± 2.640 <b>de</b>	63.23 ± 2.519 <b>d</b>	43.00 ± 0.919 <b>a</b>	28.80 ± 0.499 <b>b</b>	27.68 ± 1.953 <b>f</b>	0.38 ± 0.022 <b>e</b>
<b>Río Tinto</b>							
100%	10.79 ± 0.033 <b>b</b>	75.47 ± 0.451 <b>b</b>	65.25 ± 0.871 <b>ef</b>	65.79 ± 3.447 <b>f</b>	53.46 ± 0.880 <b>f</b>	25.16 ± 0.363 <b>b</b>	0.20 ± 0.008 <b>b</b>
85%	13.72 ± 0.433 <b>f</b>	78.55 ± 0.844 <b>c</b>	64.33 ± 0.626 <b>e</b>	53.72 ± 3.152 <b>c</b>	40.94 ± 0.210 <b>c</b>	26.35 ± 0.253 <b>e</b>	0.30 ± 0.035 <b>c</b>
70%	15.25 ± 0.447 <b>g</b>	84.16 ± 0.065 <b>f</b>	57.03 ± 1.867 <b>a</b>	43.77 ± 2.800 <b>a</b>	26.94 ± 0.812 <b>a</b>	26.41 ± 0.040 <b>e</b>	0.42 ± 0.017 <b>f</b>
<b>Nemador</b>							
100%	11.24 ± 0.286 <b>c</b>	74.97 ± 0.009 <b>ab</b>	65.25 ± 2.144 <b>ef</b>	69.52 ± 0.809 <b>g</b>	55.66 ± 2.43 <b>g</b>	26.22 ± 0.382 <b>de</b>	0.20 ± 0.008 <b>b</b>
85%	12.23 ± 0.621 <b>d</b>	80.97 ± 0.864 <b>c</b>	64.33 ± 1.670 <b>e</b>	58.68 ± 0.351 <b>d</b>	40.10 ± 0.803 <b>c</b>	25.85 ± 0.099 <b>cd</b>	0.38 ± 0.035 <b>e</b>
70%	16.14 ± 0.175 <b>h</b>	87.72 ± 2.570 <b>g</b>	57.03 ± 0.651 <b>a</b>	46.14 ± 1.297 <b>b</b>	28.53 ± 0.312 <b>b</b>	20.73 ± 0.845 <b>a</b>	0.43 ± 0.017 <b>g</b>
<b>Analysis of variance</b>							
Effect of regime irrigation (R)	*	**	**	**	**	*	**
Effect of cultivar (C)	**	**	**	**	**	*	**
Interraction (C) x (R)	**	**	**	**	**	*	**

Within each column of the three cultivars of tomato, all values are the mean of nine plants (n = 9) irrigated by different regimes. Mean ± SE with different letters are significantly different at  $p \leq 0.05$  according to LSD test.

\*, \*\* indicate significant differences at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively according to the analysis of variance.

DW: dry weight

**Table 4.** Effect of saline water (6.57 dS m<sup>-1</sup>) on Na, Cl, K, Ca, Mg and P content (meq 100 g<sup>-1</sup> DW) and Na<sup>+</sup>/K<sup>+</sup> ratio of three cultivars of tomato (Rio Grande, Rio Tinto and Nemador) irrigated by drip irrigation (DI) and subsurface drip irrigation (SDI).

Treatment	Na <sup>+</sup>	Cl <sup>-</sup>	K <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	P	Na <sup>+</sup> /K <sup>+</sup>
<b>DI</b>							
Rio Grande	16.35 ± 0.520 <b>e</b>	80.85 ± 1.126 <b>d</b>	61.02 ± 1.423 <b>c</b>	52.85 ± 2.865 <b>d</b>	35.40 ± 0.529 <b>a</b>	24.23 ± 0.156 <b>b</b>	0.29 ± 0.019 <b>a</b>
Rio Tinto	15.18 ± 0.304 <b>d</b>	82.14 ± 0.104 <b>e</b>	58.64 ± 0.256 <b>b</b>	44.47 ± 3.056 <b>a</b>	37.66 ± 1.068 <b>a</b>	25.90 ± 1.030 <b>d</b>	0.31 ± 0.011 <b>b</b>
Nemador	15.31 ± 0.216 <b>d</b>	83.88 ± 1.021 <b>f</b>	57.35 ± 1.167 <b>a</b>	49.06 ± 0.190 <b>b</b>	35.38 ± 0.538 <b>a</b>	23.21 ± 0.873 <b>a</b>	0.36 ± 0.030 <b>d</b>
<b>SDI</b>							
Rio Grande	10.40 ± 0.380 <b>a</b>	75.22 ± 1.235 <b>a</b>	71.59 ± 2.961 <b>f</b>	68.37 ± 5.939 <b>f</b>	43.93 ± 0.673 <b>b</b>	28.51 ± 1.330 <b>e</b>	0.28 ± 0.010 <b>a</b>
Rio Tinto	11.33 ± 0.272 <b>c</b>	76.64 ± 0.113 <b>b</b>	65.77 ± 1.151 <b>c</b>	50.38 ± 6.780 <b>e</b>	43.24 ± 1.162 <b>b</b>	26.04 ± 0.409 <b>d</b>	0.30 ± 0.001 <b>b</b>
Nemador	11.10 ± 0.110 <b>b</b>	78.55 ± 1.121 <b>c</b>	64.84 ± 1.800 <b>d</b>	61.16 ± 0.840 <b>e</b>	47.48 ± 1.836 <b>b</b>	25.32 ± 0.922 <b>c</b>	0.32 ± 0.012 <b>c</b>
<b>Analysis of variance</b>							
Effect of cultivar ( C )	*	*	**	*	ns	*	*
Effect of irrigation system ( S )	**	**	**	**	**	*	**
Interaction (C) x (S)	**	**	**	**	**	*	**

Within each column of the three cultivars of tomato, all values are the mean of nine plants (n = 9) irrigated by drip irrigation (DI) and subsurface drip irrigation (SDI). Mean ± SE with the different letters are significantly different at  $p \leq 0.05$  according to LSD test.

ns, \*, \*\* indicate non significant or significant differences at  $p \leq 0.05$  and  $p \leq 0.01$ , respectively according to the analysis of variance.

DW: dry weight

Ca<sup>2+</sup> content was, in general, higher in leaves than in other organs (Figure 2D) and higher in leaves of Río Grande than the other cultivars. As obtained for K<sup>+</sup> contents, Ca<sup>2+</sup> content decreased significantly ( $p \leq 0.01$ ) when decreasing the amount of water. The highest reduction was found with the more deficit water treatment of the three cultivars (Table 3). The irrigation system, in general, affected significantly ( $p \leq 0.01$ ) the Ca<sup>2+</sup> content in the three cultivars. This content was higher in SDI than DI, especially for cv. Río Grande (Table 4).

As observed in Figure 2E, Mg<sup>2+</sup> content was significantly increased ( $p \leq 0.05$ ) in shoots of plants, especially in petioles of the three cultivars. The effects of the irrigation regime on Mg<sup>2+</sup> content are shown in Table 3. This content was significantly decreased ( $p \leq 0.01$ ) in the 70% water treatment compared with the control treatment (100% of water requirement) for the three cultivars. However, Mg<sup>2+</sup> content was significantly higher ( $p \leq 0.01$ ) in SDI than DI for the three cultivars (Table 4).

High P content was notably observed in fruits of the three cultivars, particularly Río Grande and Río Tinto and exhibited a declining trend in other organs, especially in roots of the three cultivars (Figure 2F). When compared with the control treatment (100% of water requirement), this content was significantly increased ( $p \leq 0.05$ ) in Río Grande and decreased in Nemador with the 70% water treatment (Table 3). As observed in Table 4, effect of irrigation system was significant ( $p \leq 0.05$ ) for the P content and this content was higher in SDI than DI in the three cultivars. This significant increase was notably observed in Río Grande.

## DISCUSSION

Irrigation is an important practice in agriculture. Currently, competition for fresh water in the urbanization development, industry, leisure, and agriculture causes the decline of fresh water for irrigation (Qadir and Oster, 2004). On the other hand, large amounts of saline water resources in the world (Ma *et al.*, 2008) may be a good alternative. Thus, it is necessary and feasible to use saline water for agricultural irrigation if the appropriate crops, soil and water managements are applied (Oster, 1994). Our present study exposed several effects concerning the tested treatments in a Tunisian soil. Similar results have been confirmed by Mendlinger *et al.* (1994), Oron *et al.* (2002) and Hachicha *et al.* (2006). At the crop level our data documented a great reduction of leaf area in Nemador (55.85%) compared with Río Tinto (37.85%) and Río Grande (16.81%) cultivars with the regime of 70 % of water requirement (Table 1) and in DI (Table 2). Results regarding this growth parameter (leaf area) has been reported for a large number of crops, including tomato (Juan *et al.*, 2005; Maggio *et al.*, 2004; Maggio *et al.*, 2007; Zribi *et al.*, 2009), cowpea (Anyia and Herzog, 2004), pepper (González-Dugo *et al.*, 2007), rice (Cabuslay *et al.*, 2002), Chinese kale and Casin (Issarakraisila *et al.*, 2007). They reported that the observed reduction was due in part to a general size reduction of plants and in part to the fall of the bottom leaves induced by anticipated senescence.

The chlorophyll content was significantly increased with the 70% of water regime, using the saline water on the three cultivars. This phenomenon was more pronounced in Río Grande with the

most deficit treatment, compared with the control treatment (100% of water requirement) (Table 1). So, we can deduct that this cultivar is more tolerant than the other cultivars to deficit regime treatment. This is consistent with reports for other agricultural and horticultural crops, including winter triticale (Hura *et al.*, 2009) and potato (Teixeira and Pereira, 2007). As shown in Table 2, a significant reduction of chlorophyll content was observed in plants of the three cultivars irrigated by DI with saline water. This significant reduction was due to the greater accumulation of salts in DI than SDI. The accumulation of salts can be due to the increase of the evaporation rate near the soil surface in the case of DI (Hachicha *et al.*, 2006). Adverse effects observed on chlorophyll contents of the three cultivars, under salt stress, have previously been shown in rice (Yeo *et al.*, 1990), barley (Belkoudja *et al.*, 1994), tomato (Kaya *et al.*, 2001) and *Shinopsis quebracho colorado* (Meloni *et al.*, 2008). The present study shows a significant increase in  $\text{Na}^+$  and  $\text{Cl}^-$  with the water deficit treatment (70% of water requirement) for the three cultivars (Table 3). This significant increase lead sharply to reduce the  $\text{K}^+$ ,  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$  and P contents (Table 3) of the three cultivars. These results are in agreement with findings in tomato under saline stress (Pérez -Alfocea *et al.*, 1996; Al-Karaki, 2000; Dasgan *et al.*, 2002; Maggio *et al.*, 2004), rice, maize and soybean under drought stress (Tanguilig *et al.*, 1987) who reported that nutrients uptake by plants is decreased under drought stress conditions due to reduced transpiration, impaired active transport and membrane permeability, resulting in reduced root absorbing power. As observed in the present experiment, applying saline water continuously through DI might result in high salt accumulation in the three

cultivars (Table 4). Similar effects have also been shown previously by Ayers and Westcot (1985), DeMalach and Pasternak (1993), Oron *et al.* (2002) and Hachicha *et al.* (2006). Under conventional surface DI, salts can migrate downwards and reach the main root zone during precipitation and uniform distribution of water on the soil surface. This process may inhibit water and nutrient uptake, consequently causing adverse effects on crop growth and yield (Hanson and Bendixen, 1995; Oron *et al.*, 2002). This hypothesis was confirmed by the present study for Tunisian conditions (Table 4).

As a result, the irrigation of plants of the three cultivars with saline water indicate clearly a significant increase of  $\text{Na}^+$  contents in the roots of the three cultivars, particularly Nemador and a decline in fruits, leaves and stems, respectively (Figure 2A). The  $\text{Na}^+$  accumulation in roots of the three cultivars reveals the existence of inhibition mechanisms of  $\text{Na}^+$  transport from roots towards the shoots. This hypothesis was reported by Greenway and Munns (1980), Dasgan *et al.* (2002), Chartzoulakis *et al.* (2002), Maggio *et al.* (2004) and Juan *et al.* (2005) who suggested that the tolerance to saline stress at glycophytes is associated to the capacity of uptake restriction and transport of these ions from the root zone towards the shoot. In a similar way, Zandstra-Plom *et al.* (1998), studying sodium fluxes in sweet pepper plants grown under salt stress, found that sodium is preferably accumulated in roots and in pith cells in the lower part of the stem, and it plays a decisive role in the recirculation of sodium throughout the plant. In contrast to  $\text{Na}^+$ ,  $\text{Cl}^-$  concentrations increased in leaves and petioles of the three cultivars of tomato (Río Grande, Río Tinto and Nemador), indicating that the three cultivars were

unable to exclude  $\text{Cl}^-$  ions from leaves (Figure 2B). In addition, these results were found in agreement with Maggio *et al.* (2007) on tomato crop. Moreover, the transport of  $\text{Cl}^-$  occurs mainly in the transpiration stream (Wahome, 2003), which explains the high concentration of these ions in leaves and the occurrence of salt injury. Our present study showed that  $\text{Cl}^-$  ions are absorbed at higher rates than  $\text{Na}^+$  ions in different organs of the three cultivars (Figures 2A and 2B). Some similar results were observed by Marschner (1995) and Neocleous and Vasilakakis (2007).

As well known,  $\text{K}^+$  is essential for cell expansion, osmoregulation and cellular and whole plant homeostasis (Schachtman *et al.*, 1997; Patel *et al.*, 2009). The role of  $\text{K}^+$  in response to salinity is also well documented, where  $\text{Na}^+$  depresses  $\text{K}^+$  uptake (Fox and Guerinot, 1998; Patel *et al.*, 2009). The present study shows the increase of  $\text{K}^+$  in shoots of plants (leaves and petioles) of Río Grande compared to the other two cultivars (Figure 2C). The  $\text{K}^+$  content was increased in fruits and reduced in roots of the three cultivars (Figure 2C). Similar observations were indicated by Juan *et al.* (2005) on two cultivars of tomato (cv. Brillante and cv. Jaguar). The significant decrease of  $\text{K}^+$  in roots resulted in an increase of  $\text{Na}^+/\text{K}^+$  ratio in Nemador compared to the other cultivars (Figure 2G). These results corroborate previous data obtained with tomato (Alian *et al.*, 2000; Kaya *et al.*, 2001; Romero-Aranda *et al.*, 2001; Maggio *et al.*, 2007), melon (Botia *et al.*, 1998; Sivritepe *et al.*, 2003) as well as in celery (Pardossi *et al.*, 1999), eggplant (Chartzoulakis and Loupassaki, 1997) and pepper (Chartzoulakis and Klapak, 2000; Kaya *et al.*, 2009). Decrease of  $\text{K}^+$  in roots may provide a mechanism by which the three cultivars of tomato, particularly Nemador, achieve ionic balance following uptake of high  $\text{Na}^+$  concentrations in roots

(Slama, 1986, Pérez-Alfocea *et al.*, 1996, Al-Karaki, 2000; Dasgan *et al.*, 2002; Maggio *et al.*, 2007). In other way,  $\text{K}^+$  exhibited a rapid decrease in roots, while it increased in leaves, petioles and a high increase in fruits of three cultivars; it can be attributed to: (i) transfer of  $\text{K}^+$  from roots to leaves and fruits, (ii) an exchange of  $\text{K}^+$  ions with  $\text{Na}^+$  ions in root tissues, and (iii)  $\text{Na}^+$  could have directly interfered with  $\text{K}^+$  uptake. The accumulation pattern of  $\text{K}^+$  and  $\text{Na}^+$  in the three cultivars, particularly Río Grande belongs to group B and/ or group C plants in Marschner (1995) classification of ability of plants to substitute  $\text{Na}^+$  for  $\text{K}^+$ . In this classification, Marschner divided plants into four groups, A, B, C and D, depending upon whether  $\text{K}^+$  is mostly exchangeable with  $\text{Na}^+$ . Sodium has a positive effect on growth in A and B plants (mostly salt-tolerant plants). Group C plants contain very little  $\text{K}^+$  that can be substitutable with  $\text{Na}^+$  without a negative effect on growth, and group D plants exhibit no  $\text{K}^+/\text{Na}^+$  substitution mechanisms (salt-sensitive plants).

Calcium is important during salt stress, e.g. in preserving membrane integrity (Rengel, 1992), signalling in osmoregulation (Mansfield *et al.*, 1990) and influencing  $\text{K}/\text{Na}$  selectivity (Cramer *et al.*, 1987). In the present study,  $\text{Ca}^{2+}$  was transferred from roots to leaves at high proportion (Figure 2D). This increased  $\text{Ca}^{2+}$  content observed in leaves of the three cultivars, particularly Río Grande, may reduce the toxicity of  $\text{Na}^+$  in leaves. So we confirm results obtained by Cramer *et al.* (1987) on cotton, Caines and Shennan (1999), Romero-Aranda *et al.* (2001) and Juan *et al.* (2005) on tomato. As a result,  $\text{Ca}^{2+}$  fertilizers may mitigate  $\text{Na}^+$  toxicity to the plants. Besides the role of  $\text{Mg}^{2+}$  in chlorophyll structure and as an enzyme cofactor, another important role of  $\text{Mg}^{2+}$  in plants is found in the exportation of

photosynthates, which is impaired and leads to enhanced degradation of chlorophyll in Mg deficient source leaves, resulting in increased oxygenase activity of the RuBP carboxylase (Marschner and Cakmak, 1989).

The interaction between salinity and P is very complex and there is no clear cut mechanistic explanation for decreased, increased or unchanged P uptake in response to salinization in different species (Grattan and Grieve, 1992). However, it is known that P concentration is related to the rate of photosynthesis and it decreases the conversion of fixed carbon into starch (Overlach *et al.*, 1993); therefore, decrease of P in leaves will reduce shoot growth. A decrease of P concentration in root tissues, on the other hand, strongly stimulates the formation of root hairs and lateral roots in leguminous trees, rape, spinach, tomato and white lupin (Racette *et al.*, 1990). Decreased P in roots may have influenced the increase in root production.

The application of this research is concerned with the improvement of the water use and management under saline conditions of Tunisia; according our results, SDI can be included as an effective option for tomato production in combination to more tolerant cultivars like Río Grande. At the same time, physiological findings can be considered in cultivars improvement for the arid and semi-arid zones.

## CONCLUSIONS

Field experimentation permitted to quantitatively analyze the response of three tomato cultivars (Río Grande, Río Tinto and Nemador) to water deficit and to the water application method under irrigation with saline water. Growth

parameters such as leaf area, chlorophyll content and mineral composition of leaves, petioles, stems and roots were affected significantly by the different treatments, particularly for the cultivars Río Tinto and Nemador. Increasing water deficit reduced the contents of  $K^+$ ,  $Ca^{2+}$  and  $Mg^{2+}$  in all the three cultivars, while  $Na^+$ ,  $Cl^-$  and  $Na^+/K^+$  ratio were significantly increased in the different organs of the three cultivars. This strong toxic ion accumulation leads a different distribution of contents of  $Ca^{2+}$  and  $K^+$ ,  $Mg^{2+}$  and P in the different organs (leaves, petioles, stems, roots and fruits). Concerning water application method, SDI increased leaf area, chlorophyll,  $K^+$ ,  $Ca^{2+}$  and  $Mg^{2+}$  while reduced significantly  $Na^+$  and  $Cl^-$  for all the three cultivars compared to DI. So the differences between the three cultivars, exhibiting behaviour under Tunisian conditions according to published research, can be used to select for cultivar tolerance to salinity where Río Grande was the more adapted; while according our results, SDI can be included as an effective option for tomato production in combination to more tolerant cultivars, from the agronomic point of view.

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