



American Journal of  
**Plant Physiology**

ISSN 1557-4539



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## Effect of Salt Stress on Seed Germination, Plant Growth, Photosynthesis and Ion Accumulation of four Tomato Cultivars

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### ABSTRACT

Plant growth and seed germination are severely affected by saline conditions. Local tomato cultivars could be better adapted to salt stress. For this reason, to test that, the effect of four levels of salinity (0, 50, 100 and 150 mM NaCl) on seed germination, plants growth (relative fresh and relative dry weight),  $K^+$  and  $Na^+$  content and photosynthetic rate of the four local cultivars (Heb, Ram and J1) and one commercial cultivar (Mar) was studied. Significant difference in  $G_{50}$  of Heb cultivar was seen at 50 and 100 mM NaCl when compared with the other four cultivars ( $p < 0.05$ ) and the only one achieved 50% germination at 150 NaCl. Salt stress reduced plant growth of all cultivars, but Ram and Mar cultivars were characterized as the most tolerant and sensitive, respectively. No significant difference was seen in  $K^+/Na^+$  ratio among four cultivars tested, but Ram showed the maximum value of 5.72 and 35.09 at 50 and 100 mM NaCl, respectively. Ram also showed better photosynthesis rate (5.1, 3.71) at 50 and 100 mM NaCl, respectively, than the other four cultivars.

**Key words:** *Solanum lycopersicum* mill,  $K^+/Na^+$ , fresh weight, dry weight, infrared gas analyzer

### INTRODUCTION

Tomato (*Solanum lycopersicum* Mill.) belongs to family Solanaceae is the second largest vegetables crop, widely cultivated all over the word and grown in approximately 4.3 million hectares with annual production in 2009 reached to 150 million tonnes (FAO, 2009).

In Palestinian territory, it considered as one of the most important vegetables, where it is cultivated in total area of 24,921 dunums with annual production reaches to 207,559 tons in 2008 (<http://www.pcbs.gov.ps>).

Salinity affects over 70 million hectare of agricultural land which is about 20% of irrigated land and about 2% of dry land (FAO, 2002). Currently, salinity affects about 33% of all irrigated lands in the world (Munns, 2005).

Several studies have investigated the effects of the salinity on tomato crop. As the response to salinity is genotype specific (Tester and Davenport, 2003; Maggio *et al.*, 2004), other new cultivars should be studied to evaluate their degree of tolerance to saline stress.

The tomato genotype and its developmental stages respond differently to salinity (Turhan *et al.*, 2009; Santa-Cruz *et al.*, 2002; Fernandez-Garcia *et al.*, 2004; Estan *et al.*, 2005). Increasing salinity is resulted in significant reductions in shoot weight, plant height and root length (Parida and Das, 2005; Hajer *et al.*, 2006).

Plants are affected by salinity through osmotic stress and ion toxicity (Munns, 2005). Presence of ions (mainly  $Na^+$  and  $Cl^-$ ) in the soil limits the availability of water to plant leading to osmotic stress. While, accumulation of these ions in leaves to detrimental level leads to ion toxicity.

Salinity also has an adverse impact on photosynthetic rate. Several authors showed the impact of salinity on chlorophyll contents (Khavari-Nejad and Mostofi, 1998).

Selection and breeding of salt tolerant plants is becoming one option to minimize the negative impact of salinity (Epstein *et al.*, 1980).

In Palestinian Territories, salinity is a major constrain in farming, where tomato production is adversely affected by moderate to high saline content in the soil (Dudeen, 2008).

Screening for local cultivars which were being grown for hundreds of years for now is an urging priority for Palestinian scientists. For this reason, the present investigation was undertaken to screen the level of salt tolerance in four local cultivars at seed germination level and to investigate the effect of salts on other physiological parameters.

## MATERIALS AND METHODS

**Plant materials and growth conditions:** Four tomato cultivars, Jenin1 (Jen1), Hebron (Heb), Ramallah (Ram) from the Palestinian Ministry of Environmental Affair and a commercial cultivar Maramand (Mar) kindly provided by Polytechnic University, Hebron in January, 2011 were used in the present study.

**Seed disinfection and germination:** Seeds were surface sterilized with 1% (v/v) sodium hypochlorite for 20 min, rinsed four times with distilled sterile water and briefly blotted on sterile Whatman paper. Twenty seeds from four cultivars, ten in each petri dish were germinated in 90 mm petri dishes containing different concentration of NaCl (0, 50, 100 and 150 mM) solidified with 8 g L<sup>-1</sup> Agar and incubated in light at 23±2°C.

A seed was regarded germinated by the emergence of radical. The number of germinated seeds was recorded daily for 14 days.

The Germination Rate (GT) were evaluated using the GT<sub>50</sub> parameter, defined as the time (days) required for the germination of 50% of the seeds.

Days needed for 50% germination of total seeds was calculated for the each replicate and averaged for the two replicates.

**Effect of salt concentration on plant growth:** Twenty one days old seedlings were transferred to ½ MS medium (Murashige and Skoog, 1962) in Magenta boxes with the correspondent salt concentration solidified with 8 g L<sup>-1</sup> agar for one month. These plantlets were transferred to Pots containing vermiculites and irrigated with Hoagland nutrients solution containing salt with correspondent concentration (0, 50, 100 and 150 mM NaCl) Fresh weight (FW), Dry Weigh (DW), ion contents and photosynthetic rate was determined on four month. For dry weight, Shoots were weighted and kept overnight in oven at 105°C. For photosynthetic rate, Infra red gas Analyzer was used. At the end, Na<sup>+</sup> and K<sup>+</sup> was determined using flame photometer after extraction of dry weight with Ammonium acetate.

**Statistical analysis:** Statistical analysis was performed using a Duncan multiple range test (for p<0.05) for a 95% confidence level to test for significant differences among treatments.

## RESULTS

**Effect of salt on seed germination:** In all four cultivars tested, seed germination was delayed in response to salt stress. There was a differential response among cultivars in time of response in

all treatments (Table 1). There was no significant difference in  $GT_{50}$  values among four cultivars grown without salt and the  $G_{50}$  ranged from 2.45-3.3 days. At 50 mM NaCl, the  $G_{50}$  value of Heb cultivar was 3.77 days and was the only one significantly different from the other four cultivar tested; J1(4.85), Ram (4.32) and Mar (5.81). At 100 NaCl, Heb showed the lowest  $G_{50}$  (7.22) followed by J1 (8.51), Ram (9.51) and Mar (11.5). Furthermore, at 100 mM NaCl, Heb cultivar showed the least  $G_{50}$  value (7.22) than the other four cultivars; Ram (9.51) than Mar (11.5). However, after 10 days of incubation at 150 mM NaCl, only seeds of the Heb cultivar achieved 50% germination (Table 1).

**Effect of salt concentration on plant growth:** It was observed that as the salt concentration increased, plant growth was reduced in all four cultivars. A clear difference was seen between plant grown under stress condition and the control in all cultivars. Furthermore, there was a slightly difference in the plant growth among four cultivars watered with Hoagland's medium containing 50,100 and this difference is clearly seen in plants grown under 150 mM NaCl and the control.

The data of relative fresh weight and relative dry weight to the control are presented in Table 2 and 3.

Ram cultivar showed the highest and significant accumulated fresh weight than other four cultivars at four levels of salinity. It was not significant only when compared with J1 at 50 mM salinity level. On the other hand, Mar cultivar showed the least value in relative FW (5.96) at 100 mM salinity level when compared with the other four cultivars. The major difference was seen in plants grown under 150 mM NaCl. In general, Ram cultivar showed the highest value at 100 mM salinity level than the other four cultivars. Furthermore, J1 and Mar cultivars failed to continue to grow at 150 mM salinity level (Table 2).

Ram cultivar showed the highest accumulative relative dry weight (37.18, 32.06) at 100 and 150 mM NaCl levels than other four cultivars, respectively. The commercial Mar cultivar was more sensitive than others and its value was 5.45 at 100 mM NaCl (Table 3).

Table 1: Effect of salinity on  $G_{50}$  values of four tomato cultivars grown under four levels of salinity

Cultivar	NaCl Conc. (mM)			
	0	50	100	150
J1	2.45±0.35 <sup>a</sup>	4.85±0.21 <sup>a</sup>	8.51±0.70 <sup>bc</sup>	0.0 <sup>b</sup>
RAM	3.00±0.70 <sup>a</sup>	4.32±0.46 <sup>ab</sup>	9.51±3.35 <sup>b</sup>	0.0 <sup>b</sup>
Heb	2.77±0.31 <sup>a</sup>	3.77±0.46 <sup>b</sup>	7.22±2.40 <sup>c</sup>	10.0±3 <sup>a</sup>
Mar	3.30±0.32 <sup>a</sup>	5.81±0.22 <sup>a</sup>	11.50±1.25 <sup>a</sup>	0.0 <sup>b</sup>

Values are Mean±SD of two independent experiments, Values in the same column followed by the different alphabets are significantly different according to DMRT at  $p<0.05$ ,  $G_{50}$ : Mean time (days) required for 50% of seed to germinate

Table 2: Accumulated relative fresh weight (g) of four tomato cultivars grown under four levels of salinity

Cultivar	NaCl Conc.(mM)			
	0	50	100	150
J1	100	36.70 <sup>a</sup>	15.6 <sup>bc</sup>	0.00 <sup>c</sup>
RAM	100	39.27 <sup>a</sup>	29.2 <sup>a</sup>	4.47 <sup>a</sup>
Heb	100	21.90 <sup>bc</sup>	18.8 <sup>b</sup>	1.28 <sup>b</sup>
Mar	100	26.42 <sup>b</sup>	5.96 <sup>d</sup>	0.00 <sup>c</sup>

Values in the same column followed by the different alphabets are significantly different according to DMRT at  $p<0.05$

Table 3: Accumulated relative DW (g) of four tomato cultivars grown under four levels of salinity

Cultivar	NaCl conc.(mM)			
	0	50	100	150
J1	100	55.30 <sup>a</sup>	20.62 <sup>b</sup>	0.00 <sup>c</sup>
RAM	100	44.64 <sup>ab</sup>	37.18 <sup>a</sup>	32.06 <sup>a</sup>
Heb	100	23.47 <sup>c</sup>	16.90 <sup>bc</sup>	9.01 <sup>b</sup>
Mar	100	34.67 <sup>b</sup>	5.45 <sup>c</sup>	0.00 <sup>c</sup>

Values in the same column followed by the different alphabets are significantly different according to DMRT at  $p < 0.05$

Table 4: Relative  $K^+/Na^+$  content of four tomato cultivars grown under four level of salinity

Cultivar	NaCl conc. (mM)			
	Heb	Ram	J1	Mar
50	23.78 <sup>c</sup>	57.20 <sup>a</sup>	38.9 <sup>b</sup>	45.80 <sup>a</sup>
100	24.08 <sup>b</sup>	35.09 <sup>a</sup>	30.5 <sup>a</sup>	29.48 <sup>ab</sup>

Values in the same column followed by the different alphabets are significantly different according to DMRT at  $p < 0.05$

Table 5: Effect of different salt concentration on photosynthetic rate ( $\mu\text{mol m}^{-2} \text{sec}^{-1}$ ) of four tomato cultivars

Cultivar	NaCl conc. (mM)			
	0	50	100	150
J1	2.92 <sup>c</sup>	0.44 <sup>c</sup>	0.80 <sup>d</sup>	0.00 <sup>c</sup>
RAM	7.15 <sup>a</sup>	5.10 <sup>a</sup>	3.71 <sup>a</sup>	3.24 <sup>a</sup>
Heb	4.45 <sup>b</sup>	2.51 <sup>b</sup>	2.91 <sup>b</sup>	1.97 <sup>b</sup>
Mar	6.28 <sup>a</sup>	0.43 <sup>c</sup>	1.20 <sup>c</sup>	0.00 <sup>c</sup>

Values in the same column followed by the different alphabets are significantly different according to DMRT at  $p < 0.05$

**Sodium and potassium content:** Sodium concentration increased in plant shoot with increasing salt stress, while potassium content decreased in plant shoot with increasing salt stress. At 50 mM NaCl no difference was seen between Ram and Mar where relative  $K^+/Na^+$  was 57.2 and 45.8 for Ram and Mar respectively. At the same NaCl concentration Ram and Mar showed higher  $K^+/Na^+$  than J1 and Heb cultivars, while at 100 mM salinity level, the only significant difference was between Ram and Heb cultivars. On the other hand, no significant difference was seen between Ram cultivar and J1 and Mar cultivars. In general, Heb cultivar showed the least  $k^+/Na^+$  at 50 and 100 Mm NaCl (Table 4).

Photosynthetic rate is also affected by an increase in salt concentration. It decreased in all cultivar tested as salt concentration increased. Under control conditions, no difference in photosynthetic rate was seen between Ram and Mar cultivars, but at the same conditions, significant difference was seen between Ram and Mar and the other two cultivars, J1 and Heb. At 50 and 100 mM salinity level, Ram was significantly difference in it photosynthetic rate 5.1 and 3.71, respectively than the other four cultivars. In general, Ram cultivar showed higher photosynthetic rate than the other cultivar in all salt treatments (Table 5).

## DISCUSSION

Agricultural production is highly affected by Salinity mainly due to presence of high concentration of  $Na^+$  and  $Cl^-$ . Tomato plant grown in arid or semi arid land is under a biotic stress

during their life cycle. Different mechanisms were used by some of these plants to tolerate these stresses depending on plant itself (Ahmad *et al.*, 2009).

In this study, four concentrations (50, 100 and 150) of NaCl were used in order to evaluate its effect on seed germination, growth parameters,  $K^+$ ,  $Na^+$  contents and photosynthetic rate of four local tomato cultivar (Heb, Ram and J1) and one commercial tomato cultivar (Mar). The results of current study indicated that, an increase of salt concentrations delayed seed germination of the four tomato cultivars especially at the highest concentration (150 mM). The cultivar Heb was less affected by salinity stress than the other four cultivars (Ram, J1 and Mar). This may be explained to partially osmotic or Ion toxicity as have been reported by others (Begum *et al.*, 1992; Croser *et al.*, 2001; Essa and Al-Ani, 2001) who reported that delay or prevention of seed germination may be due to partially osmotic or ion toxicity which can affect enzymes activity. The same explanation was also reported by other researches (Bewley and Black, 1982; Poljakoff-Mayber *et al.*, 1994; Caramer *et al.*, 1994; Mansour, 1994; Leopold and Willing, 1984; Perez-Alfocea *et al.*, 1993).

In our results, the saline conditions reduced the growth parameter such as fresh and dry weights of the four tested cultivars. These results were in conformity with other results reported by several authors (Taffouo *et al.*, 2010; Turhan *et al.*, 2009; Maggio *et al.*, 2007; Mohammad *et al.*, 1998; Hajer *et al.*, 2006). Others reported that physiological parameters like fresh weigh, dry weight, leaf area, plant stem and roots of tomatoes were reduced when grown in saline condition above 100 mM (Omar *et al.*, 1982). Satti and Al-Yahyai (1995) showed that when plant irrigated with nutrients containing different salt concentrations, the leaf and stem dry weights of tomato were also reduced significantly in contrast with control plants.

Plant tends to either accumulate  $Na^+$  in their vacuole or extrude them through their roots. We showed that  $K^+/Na^+$  ratio was decreased as salt concentration increased. High concentration of  $Na^+$  was seen in shoots of all cultivars tested. This was accompanied by decrease in  $K^+$  accumulation. Similar results was obtained by Al-Karaki (2000), who showed that increasing NaCl concentration in nutrient solution adversely affected tomato shoot and roots, plant height,  $K^+$  concentration and  $K^+/Na^+$ .

The control of  $Na^+$  accumulations and high  $K^+/Na^+$  ratios may enhance salt tolerance and the  $K^+/Na^+$  ratio has been used as a indicator by a number of authors to select salt tolerant in tomato crops (Dasgan *et al.*, 2002; Juan *et al.*, 2005).

Photosynthetic rate is also affected by salt stress. Ram have higher photosynthetic rate than the other four cultivars. Decrease in photosynthetic rate may be attributed to decrease in chlorophyll contents. Researchers like Khavari-Nejad and Mostofi (1998) and Hajer *et al.* (2006) also reported that tomato plant photosynthesis decreased when subjected to salt stress. Others, reported that stomatal closure and high NaCl concentration may be responsible for the decrease in chlorophyll content in cotton plants when were treated with NaCl (Meloni *et al.*, 2003). Salinity stress affect chlorophyll content because of its adverse effects on membrane stability as being reported by Ashraf and Bhatti (2000) and Al-Sobhi *et al.* (2006).

## CONCLUSION

Local cultivar Ram showed better salt tolerant than other cultivars. It also showed better photosynthesis when compared with others. In general, local cultivars performed better than commercial cultivar (maramand). This cultivar could be used later for any breeding program for improvement of local genotypes for salt stress.

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