

*Full Length Research Paper*

# Salinity response of some tomato rootstocks at seedling stage

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The research aims to identify the response of some commercial tomato rootstocks namely Beaufort, Body, Heman, Resistar, Spirit, Vigomax, Yedi to salinity stress with the screening method. The study assays not only non-grafted (cv. Gokce) but also self-grafted plants as control plants in order to demonstrate grafted union effect. The plants were placed in styrofoam on horizontal pots and grown in aerated water culture. Salination was initiated one week after the transplantation of seedlings to the pots. Salinity level was gradually increased up to 300 mM by adding 50 mM NaCl one day interval and plants were left for 10 days at that salinity level. Plants were removed before salt treatment and at 150 and 300 mM NaCl level. They were classified according to the severity of leaf symptoms caused by salinity levels. In addition, scale scoring, leaf number, stem length and diameter, leaf area, total root length per plant and leaf catalase (CAT) and peroxidase (POX) enzyme contents were measured. Furthermore, the plants were separated into shoots and roots for dry matter assimilation. The results demonstrated that salt stress decreased plant vigor, and rootstocks yielded better performance than non- and self-grafted treatments. Beaufort and Yedi appeared as more tolerant genotypes than the others under stress condition. CAT activity decreased while POX activity increased with increased salinity levels and both activities occurred higher in grafted treatments. The study concludes that grafting may be considered as an alternative strategy to enhance salt tolerance in tomato; however, the effects of the grafting may vary for different rootstock genotypes.

**Key words:** Plant growth, enzyme, grafting, NaCl, screening.

## INTRODUCTION

Salinity, as the most important stress factor detrimental to the performance of agricultural products creates a high risk in greenhouses compared to open-field cultivation (Anac and Eryuce, 2003). In Turkey, greenhouses are located at intensive agricultural areas and most of them are small scale family enterprises. Unfavorable drainage in addition to high temperature and evapotranspiration, the obligation to use low quality water, unbalanced and intense fertilization by the producers (Kaplan et al., 1997; Sonmez and Kaplan, 2004), penetration of sea water into irrigation water on the coast line (Anac and Eryuce, 2003) are the major factors that cause to salinity in greenhouses. Salinity problem may also occur in soilless

culture because of both low quality irrigation water use and rapid accumulation of salt in the nutrition solution into small pot volumes (Sonneveld et al., 1999; Li, 2000). Regular and conscious fertilization, organic substance use, removal of excess water by means of drainage, washing the soil with extra irrigation, deep-tilling, and replacing the top soil layer from time to time (Sevgican, 2002) in greenhouses; and interim washes, alternate use of salty and clean water (Adams and Ho, 1989) and suitable fertilization regimes (Navarro et al., 2000) in soilless culture could be the solutions to overcome this problem. However, these are not easy and practical solutions; they are time consuming and expensive indeed. In addition, recovering the salinity which occurs due to the salinity of irrigation water seems not so possible. Under the current conditions with a higher necessity of saving from water and fertilizer use; the safest and most effective solution appears as using

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resistant plants to salty conditions, and grafted plants are critical in this context.

Grafting applications on vegetable began in the Asian countries such as China, Japan and Korea who failed to overcome the soil-borne disease (*Fusarium*) and nematode problem (Kurata, 1994). Its use and spread increased rapidly in time as an affective alternative to methyl bromide (MeBr) (Lee, 1994; Leonardi and Romano, 2004). In our country, the first commercial grafted seedlings were introduced in 1998 and their annual production quantity increased to 72 million seedlings by the end of 2010 (34 million watermelons, 28 million tomatoes, 7.5 million eggplants, 2 million cucumbers, 300 thousand melons, and 200 thousand peppers) (Öncel, 2010, nursery consultant, Antalya, personal communication).

Although the actual reason for using grafted plants in vegetable culture is to ensure the plant's resistance against biotic stress conditions (against soil-borne diseases such as *Fusarium*, *Verticillium*, root corky and nematode); it is also known that grafted seedlings increase resistance/adaptation against abiotic stress conditions (that is, low temperature, high temperature, drought, flooding and salinity) (Romero et al., 1997; Edelstein et al., 1999; Fernandez-Garcia et al., 2003). Grafted seedlings are able to uptake more water and nutrients from the soil and transfer them to the upper portion compared to non-grafted seedlings as they have longer and denser roots (Kovalev, 1990; Ra et al., 1995). Nevertheless, it is also known that the cytokinins synthesized abundantly in the roots are moved to the upper portion by xylem juice, thus increasing plant growth and productivity (Lee and Oda, 2003). Also it is claimed that under the stress conditions such as salinity, grafted plants develop various mechanisms to shut down  $\text{Cl}^-$  ion uptake and reduce  $\text{Cl}^-$  absorption by roots or retain  $\text{Na}^+$  ion in the roots in order to prevent excessive accumulation of  $\text{Na}^+$  and  $\text{Cl}^-$  ions on the leaves (Romero et al., 1997; Santa-Cruz and Cuertero, 2001; Fernandez-Garcia et al., 2003).

Being exposed to an environmental stress such as salinity stress impairs the balance between the plant's production and consumption of reactive oxygen types, and oxidative damages occur as a result. The electrons which leak from the electron transport chain in mitochondria and chloroplasts interact with  $\text{O}_2$  and create "active oxygen radicals" such as superoxide ( $\text{O}_2^{\cdot-}$ ), hydroxyl (OH), hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) and singular oxygen ( $1\text{O}_2$ ). These reactive radicals give oxidative damage to lipids, proteins, enzymes and nucleic acids, and impact the normal metabolism. In order to cope with it, plants have developed an "antioxidant defense system" which cleans these radicals. Antioxidant defense system contains enzymes such as superoxide dismutase (SOD), peroxidase (POX), catalase (CAT), glutathione reductase (GR), and nonenzymatic antioxidants such as polyamines,

phenols, carotenoids, abscisic acid and proline (Halliwell and Gutteridge, 1989; Bowler et al., 1992; Edreva, 1998). In general, plants' exposure to oxidative stress causes to an increase in gene products and substrates related with antioxidant defense system. The effectiveness of such a configuration of the plant, and basic antioxidant levels create the basis of the distinction of whether the plant is sensitive to or durable against stress (Harper and Harvey, 1978; Wise and Naylor, 1987). Higher antioxidant defense enzyme activities under salinity stress compared to non-grafted and self-grafted plants may be an indicator for the durability of grafted plants against salinity stress (Hea et al., 2009).

This study examines the reactions of certain most commonly used commercial tomato rootstocks against salinity. Plant growth parameters, biomass (dry weight) quantities and antioxidative enzyme activities of grafted and non-grafted plants under salty and salt-free conditions have been identified, and the study has been conducted in water culture as it facilitates establishing salt reaction. In this context, the identification and use of salt-resistant rootstocks at the places with salinity problem will obviously yield great benefits for the tomato producer.

## MATERIALS AND METHODS

The research has been conducted at the Department of Horticulture, Faculty of Agriculture in Ege University (38°27' 16.2" N; 27°13' 17.8" E; altitude 33 m). The experiment was carried out in a PE-covered bi-tunnel greenhouse (min. and max. temperature 20.8 to 25.9°C; relative humidity 46.2 to 81.7%; daily solar radiation 2.6 to 10.2 MJ m<sup>-2</sup>). In the research, which employs tomato as the plant material, the most commonly available commercial rootstocks in the market namely Beaufort, Body, Heman, Resistar, Spirit, Vigomax and Yedi have been assayed, CV.Gokce (191) tomato F<sub>1</sub> hybrid has been used as the scion; non-grafted and self-grafted Gokce/Gokce combination has been accepted as the control group in order to observe the grafting unit activity. The properties of rootstocks and culture type in the research are given in Table 1.

The research has been conducted in water culture. The seedlings supplied from a seedling company as grafted in single stem with "Splice (Tube) Grafting" method were planted on 17 September, 2007, in the foam plates placed in light-proof brown plastic lateral pots (75 × 23 × 16 cm) with 18.75 cm distance in a manner that the seedling roots completely touch the nutrition solution, after the peat + vermiculite mixture on their roots were washed.

The assay has been realized in three repetitions according to randomized parcels experimental design.

The plants were fed with nutrient solution prepared according to Day (1991) for 1 week from the date of plantation (N 210, P 40, K 250, Ca 150, Mg 50, Fe 2, Mn 0.75, B 0.4, Zn 0.50, Cu 0.10 and Mo 0.05 (mg/l)), and the nutrient solution used by the plant within this period was supplemented with 2 days interval (EC:2.0 to 2.5 dS m<sup>-1</sup>, pH:5.5 to 6.0) and the aeration of the solution with aquarium pump was ensured. One week after planting (25 September, 2007), the 1st removal was performed and the treatment of salt on the remaining seedlings other than the control group was initiated. Salt treatment was realized by giving the plants a new solution prepared by adding 50 mM salt (NaCl) to the nutrient solution. The salinity

**Table 1.** Properties of used rootstocks and cultivar in experiment.

Name	Product firm	Disease resistant*	
Body F1	Seminis	ToMV, Va, Fol:2, Mi, Pl	<i>L. esculentum</i> x <i>L. hirsutum</i>
Beaufort F1	De Ruiters seed	ToMV, Va, Fol:0-1, For, Pl, Vd, Ma, Mi, Mj	<i>L. esculentum</i> x <i>L. hirsutum</i>
Heman F1	Syngenta	ToMV, Va, Fol:2, Mi, For, Pl	<i>L. esculentum</i> x <i>L. hirsutum</i>
Resistar F1	Hazera	ToMV, Va, Fol:2, Mi, Pl, For	<i>L. esculentum</i>
Spirit F1	Nunhems	ToMV, Va, Fol:2, Mi, Pl, For,	<i>L. esculentum</i>
Vigomax F1	De Ruiters seed	ToMV, Va, Fol:2, Mi, For, Pl	<i>L. esculentum</i> x <i>L. hirsutum</i>
Yedi RZ F1	Rijk Zwaan	ToMV, Va, Fol:2, Mi, Pl, For	<i>L. esculentum</i> x <i>L. hirsutum</i>
191 (Gokce) F1	Zeraim Gedera	ToMV, Va, Fol:1-2, C	<i>L. esculentum</i>

\*Fol: *Fusarium oxysporium* (0, 1, 2 races), For: *Fusarium oxysporium* f.sp. Radicis, V: Verticillium wilt Va: *Verticillium albo-atrum*, Vd: *Verticillium dahliae*, ToMV: Tomato Mosaic Virus, Pl: root rot, Mi: root-knot nematode (*Meloidogyne incognita*), Mj: *Meloidogyne javanica*, Ma: *Meloidogyne arenaria*, C: *Clodisporium*.

level of the nutrient solution has been modified as 50, 100, 150, 200, 250 and 300 mM with one day intervals and the plants were grown for 10 days under 300 mM salt stress condition. The plants in the control group were fed with normal nutrient solution during the same period. 2nd removal (28 September, 2007) was realized at the end of 150 mM salt treatment, and the 3rd removal (10 October, 2007) at the end of 300 mM salt treatment.

The plants removed at 150 and 300 mM levels were classified for their salt tolerance by the visual appearance. Plants were scored for severity of salt susceptibility by 1 to 5 scale. Accordingly, (1) is scored as undamaged, healthy leaves or slightly involuted normal green leaves, (2) completely involuted green leaves, (3) moderately-excessively damaged dry leaves in addition to involuted green leaves, (4) (50 to 80%) drying damage on most leaves (50 to 80%), (5) drying damages on all leaves (Dasgan et al., 2002). After scoring, all leaves on the seedlings were counted (pieces plant<sup>-1</sup>), and the seedling length (cm plant<sup>-1</sup>) and the stem thickness (mm) at the stem's middle section and between nodes were measured respectively by means of a tape measure and calipers. On two seedlings which belong to each treatment, all of the leaves were scanned with a scanner in their original sizes, and their pictures were transferred to computer in jpeg format. The areas of the mentioned leaves (m plant<sup>-2</sup>) were identified with Sigma Scan Pro program. After the root and shoot portion fresh weight measurements, the parts of seedlings were dried in an incubator at 65°C, their dry weights were measured and their total biomasses (g plant<sup>-1</sup>) were calculated. Total root lengths (m plant<sup>-1</sup>) were identified with the Newman method (Newmann, 1966).

The activities of peroxidase (POX) and catalase (CAT) among antioxidant enzymes were inspected on the sample leaves taken from 150 mM NaCl during the 2nd removal and 300 mM NaCl during the 3rd removal. 50% w/v gelatin and 0.15 M Na-phosphate-citrate buffer containing DAB (diaminobezidinetetrahydrochloride-dihydrate) solution and 0.6% H<sub>2</sub>O<sub>2</sub> was added to the samples diluted for POX activity, and the absorbance change at 465 nm was observed for 3 min (Herzog and Fahimi, 1973). In CAT enzyme activity analysis, reaction mixture contained 0.05 M Na-phosphate buffer (pH 7.0), 3% H<sub>2</sub>O<sub>2</sub> and 1 mM EDTA, and the consumption of H<sub>2</sub>O<sub>2</sub> at 240 nm was monitored for 3 min (Bergmeyer, 1970). In both specific enzyme activities, 1 enzyme unit was calculated as the  $\mu\text{mol}^{-1}\text{ml}^{-1}$  H<sub>2</sub>O<sub>2</sub> quantity consumed per minute, and the enzyme unit was indicated as mg protein<sup>-1</sup> g fresh weight<sup>-1</sup>.

According to the data obtained from the research, variance analysis has been realized on computer by using TARIST statistical analysis package program, and least significant difference (LSD) test has been conducted at 5% importance level in order to identify

the differences between the averages.

## RESULTS

### Salinity damage scale class

The rootstocks responded widely to the salinity stress as judged from the visual appearance; however genotypes have gathered together in the scale classes 1.0 to 3.3. At 150 mM NaCl salinity level, the highest damage was observed with non-grafted plants and self-grafted plants. *Lycopersicon esculentum* rootstocks namely Resistar and Spirit showed the second highest damage after non-grafted and self-grafted plants. A similar situation was also observed, with a higher and more evident damage, on the plants which were exposed to 300 mM NaCl salinity level. Under moderate and high salinity stress, rootstocks Vigomax and Yedi in scale class-1 were least affected by the NaCl treatment (Table 2).

### Number of leaves

A statistical difference has not been observed between the treatments in terms of the leaf count during the seedling removal before beginning to salt treatment. During the 2nd removal, among salt-free plants, Vigomax and Yedi rootstocks had the maximum leaf number with 9.7 pieces, while the highest number of leaves of the plants under salinity stress was obtained from Vigomax rootstock as 9.3. During the 3rd removal, the highest number of leaves was identified on Yedi, Spirit and Body rootstocks, while the lowest number of leaves was identified with non-grafted and self-grafted plants in salt-free treatment. For 300 mM salinity stress, the highest number of leaves was obtained from Vigomax rootstock as 11.0 and *L. esculentum* rootstocks (Resistar, Spirit) and plants (non and self grafted) had lowest leaf number when they compared with *L. esculentum* x *L. hirsutum*

**Table 2.** Leaf damages according to salt scale.

Treatment	2nd removal	3rd removal
Non-grafted	3.0 <sup>a</sup>	3.3 <sup>a</sup>
Self-grafted	2.7 <sup>ab</sup>	3.0 <sup>a</sup>
Beaufort	1.3 <sup>cd</sup>	1.7 <sup>b</sup>
Body	1.7 <sup>cd</sup>	1.3 <sup>b</sup>
Heman	1.7 <sup>cd</sup>	1.3 <sup>b</sup>
Resistar	2.0 <sup>bc</sup>	2.7 <sup>a</sup>
Spirit	2.0 <sup>bc</sup>	2.7 <sup>a</sup>
Vigomax	1.0 <sup>d</sup>	1.0 <sup>b</sup>
Yedi	1.0 <sup>d</sup>	1.0 <sup>b</sup>
LSD (0.05)	0.738	0.809

Different letters within columns indicate statistical differences ( $p < 0.05$ ).

**Table 3.** Changes on leaf number (pieces plant<sup>-1</sup>).

Treatment	1st removal -NaCl	2nd removal		3rd removal	
		-NaCl	+NaCl	-NaCl	+NaCl
Non-grafted	8.0	8.7 <sup>ab</sup>	8.0b <sup>c</sup>	11.3 <sup>c</sup>	7.3 <sup>c</sup>
Self-grafted	8.0	8.7 <sup>ab</sup>	7.7 <sup>c</sup>	11.3 <sup>c</sup>	9.0 <sup>bc</sup>
Beaufort	8.7	8.3 <sup>b</sup>	8.0b <sup>c</sup>	13.3 <sup>ab</sup>	10.3 <sup>ab</sup>
Body	8.7	9.0 <sup>ab</sup>	8.0b <sup>c</sup>	14.0 <sup>a</sup>	9.7 <sup>ab</sup>
Heman	8.7	9.3 <sup>ab</sup>	9.0 <sup>ab</sup>	12.0 <sup>bc</sup>	10.3 <sup>ab</sup>
Resistar	8.3	9.3 <sup>ab</sup>	8.3 <sup>abc</sup>	12.0 <sup>bc</sup>	9.3 <sup>ab</sup>
Spirit	8.0	9.3 <sup>ab</sup>	9.0 <sup>ab</sup>	14.0 <sup>a</sup>	9.7 <sup>ab</sup>
Vigomax	8.3	9.7 <sup>a</sup>	9.3 <sup>a</sup>	13.7 <sup>ab</sup>	11.0 <sup>a</sup>
Yedi	8.0	9.7 <sup>a</sup>	8.7 <sup>abc</sup>	14.3 <sup>a</sup>	10.7 <sup>ab</sup>
LSD (0.05)	ns	1.095	1.044	1.953	1.747

ns: Non-significant. Different letters within columns indicate statistical differences ( $p < 0.05$ ).

hybrids. The average number of leaves was counted as 8.3 during the 1st removal; as 9.2 for the salt-free plants and 8.4 for salt-treated plants with 8.4% decrease during the 2nd removal; as 12.9 for salt-free plants and 9.7 for salt-treated plants with 24.7% decrease during the 3rd removal (Table 3).

### Seedling length

During the first removal before salt treatment, seedling length was identified as 16.2 cm in average and a statistical difference has not been observed between the treatments. During the 2nd removal, the average seedling length of salt-free plants was identified as 21.6 cm and the shortest plants were obtained from non-grafted Gokce treatment. Self-grafted plants followed that. The tallest seedling was identified as Beaufort rootstock with 23.2 cm. Under 150 mM salinity stress, plant size varied between 20.5 cm (Vigomax) and 14.4 cm (Non-grafted); the average size was identified as 17.7 cm. During the

3rd removal, the lengths of salt-free plants varied between 37.2 cm (Resistar) and 55.2 cm (Spirit), and those of 300 mM salt-treated plants between 25.2 cm (Heman) and 16.0 cm (Non-grafted). During the 3rd removal, the average length of the control group was identified as 44.9 cm and that of salt-treated group as 21.1 cm. During the research, the change in seedling length according to salinity became more evident. At 150 mM salinity stress, the seedling length decreased by 17.6% compared to the control group, while the decrease in the seedling length under high salinity stress was identified as 53.1%, comparably higher than that of 150 mM salinity stress (Table 4).

### Stem thickness

For 150 mM salinity stress, the thickest stem diameter was obtained from rootstock Yedi, while it appeared on Yedi, Resistar and Heman rootstocks under 300 mM salinity stress. With salinity application, non-grafted and

**Table 4.** Changes on seedling length (cm plant<sup>-1</sup>).

Treatment	1st removal		2nd removal		3rd removal	
	-NaCl	-NaCl	+NaCl	-NaCl	+NaCl	
Non-grafted	14.2	18.2 <sup>b</sup>	14.4 <sup>c</sup>	37.2 <sup>de</sup>	16.0 <sup>c</sup>	
Self-grafted	15.5	20.7 <sup>ab</sup>	15.5 <sup>c</sup>	37.9 <sup>de</sup>	19.9 <sup>b</sup>	
Beaufort	16.3	23.2 <sup>a</sup>	16.3 <sup>bc</sup>	48.0 <sup>bc</sup>	21.0 <sup>b</sup>	
Body	17.3	22.7 <sup>a</sup>	19.0 <sup>ab</sup>	43.6 <sup>cd</sup>	22.5 <sup>ab</sup>	
Heman	16.7	21.3 <sup>ab</sup>	18.7 <sup>ab</sup>	37.9 <sup>de</sup>	25.2 <sup>a</sup>	
Resistar	15.7	22.0 <sup>a</sup>	16.7 <sup>bc</sup>	37.2 <sup>e</sup>	20.2 <sup>b</sup>	
Spirit	17.0	22.0 <sup>a</sup>	18.7 <sup>ab</sup>	55.2 <sup>a</sup>	21.7 <sup>b</sup>	
Vigomax	16.7	21.3 <sup>ab</sup>	20.5 <sup>a</sup>	54.3 <sup>ab</sup>	22.2 <sup>ab</sup>	
Yedi	16.5	22.7 <sup>a</sup>	20.2 <sup>a</sup>	53.3 <sup>ab</sup>	21.1 <sup>b</sup>	
LSD (0.05)	ns	3.687	2.881	6.444	3.405	

ns: Non-significant. Different letters within columns indicate statistical differences (p<0.05).

**Table 5.** Changes on average stem thickness (mm plant<sup>-1</sup>).

Treatment	1st removal		2nd removal		3rd removal	
	-NaCl	-NaCl	+NaCl	-NaCl	+NaCl	
Non-grafted	4.0 <sup>ab</sup>	5.1 <sup>c</sup>	3.5 <sup>d</sup>	5.9 <sup>e</sup>	4.2 <sup>c</sup>	
Self-grafted	4.2 <sup>ab</sup>	5.2 <sup>b</sup>	4.1 <sup>cd</sup>	6.2 <sup>cde</sup>	4.4 <sup>bc</sup>	
Beaufort	3.9 <sup>ab</sup>	6.1 <sup>a</sup>	4.9 <sup>abc</sup>	7.5 <sup>a</sup>	5.2 <sup>ab</sup>	
Body	3.9 <sup>ab</sup>	6.0 <sup>ab</sup>	5.3 <sup>ab</sup>	7.0 <sup>ab</sup>	5.2 <sup>ab</sup>	
Heman	3.7 <sup>ab</sup>	5.4 <sup>abc</sup>	4.8 <sup>bc</sup>	5.9 <sup>de</sup>	5.3 <sup>a</sup>	
Resistar	3.6 <sup>b</sup>	5.3 <sup>abc</sup>	4.8 <sup>bc</sup>	5.9 <sup>de</sup>	5.3 <sup>a</sup>	
Spirit	3.8 <sup>ab</sup>	6.2 <sup>a</sup>	5.0 <sup>ab</sup>	6.4 <sup>bcd</sup>	5.2 <sup>ab</sup>	
Vigomax	3.6 <sup>ab</sup>	6.1 <sup>a</sup>	5.5 <sup>ab</sup>	6.5 <sup>bcd</sup>	5.4 <sup>a</sup>	
Yedi	4.3 <sup>a</sup>	6.2 <sup>a</sup>	5.8 <sup>a</sup>	6.7 <sup>abc</sup>	5.3 <sup>a</sup>	
LSD (0.05)	0.691	0.837	0.915	0.754	0.743	

ns: Non-significant. Different letters within columns indicate statistical differences (p<0.05).

self-grafted plants constituted the plants with the thinnest stems. Upon the examination of the change in the average plant diameter according to salinity levels, it has been identified that the stem diameter decreased 15.2 and 22.8% respectively under 150 and 300 mM salinity stress, compared to those of control plants (Table 5).

### Leaf area

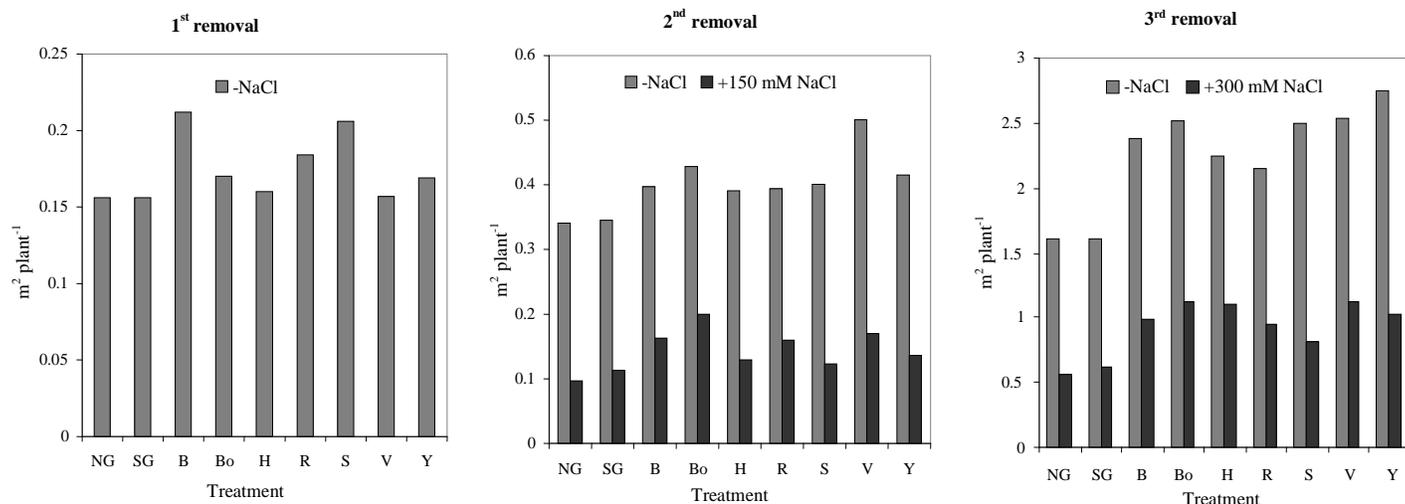
The average leaf area was identified as 0.175 m plant<sup>-2</sup> during the 1st removal; as 0.401 m plant<sup>-2</sup> for the salt-free plants and 0.144 m plant<sup>-2</sup> for salt-treated plants during the 2nd removal; and finally as 2.255 m plant<sup>-2</sup> for salt-free plants and 0.925 m plant<sup>-2</sup> for salt-treated plants during the 3rd removal. Both under 150 and 300 mM salinity stress, the highest leaf areas were obtained from Body and Vigomax rootstocks. The smallest leaf area at both salinity levels was obtained from non-grafted and self-grafted plants. Under high NaCl stress *L. esculentum*

rootstocks showed less leaf area than *L. esculentum* × *L. hirsutum* hybrids. The average leaf area under 150 and 300 mM salinity stresses respectively decreased by 64.2 and 58.9% compared to their control groups (Figure 1).

### Total root length

During the removal of the 1st plants with 27.2 m average root length, a statistical difference has not been observed with respect to root length among the averages. During the 2nd removal, the average root length was identified as 69.1 m for salt-free plants and 44.9 m for salt-treated plants. At the plants which were exposed to 150 mM salinity stress, Yedi rootstock gave the highest root with 67.9 m, and Vigomax rootstock followed it.

During the 3rd removal, the average root length under salt-free condition appeared as 92.6 m, and the longest roots were obtained Beaufort and Vigomax rootstocks respectively under salt-free and 300 mM salt-treated



**Figure 1.** Leaf areas of seedlings according to removal time and salt application. NG: Non-grafted, SG: Self-grafted, B: Beaufort, Bo: Body, H: Heman, R: Resistar, S: Spirit, V: Vigomax, Y: Yedi.

**Table 6.** Changes on total root length (m plant<sup>-1</sup>).

Treatment	1st removal		2nd removal		3rd removal	
	-NaCl	-NaCl	-NaCl	+NaCl	-NaCl	+NaCl
Non-grafted	20.0	54.2 <sup>c</sup>	26.9 <sup>d</sup>	64.4 <sup>c</sup>	36.7 <sup>c</sup>	
Self-grafted	23.1	52.8 <sup>c</sup>	30.3 <sup>cd</sup>	76.3 <sup>bc</sup>	39.9 <sup>c</sup>	
Beaufort	25.8	89.9 <sup>ab</sup>	50.7 <sup>abc</sup>	102.8 <sup>ab</sup>	62.4 <sup>ab</sup>	
Body	28.4	75.8 <sup>abc</sup>	49.6 <sup>abc</sup>	117.9 <sup>a</sup>	64.7 <sup>ab</sup>	
Heman	24.9	59.5 <sup>c</sup>	41.6 <sup>bcd</sup>	90.1 <sup>abc</sup>	63.4 <sup>ab</sup>	
Resistar	27.5	51.3 <sup>bc</sup>	37.8 <sup>cd</sup>	91.1 <sup>abc</sup>	58.2 <sup>ab</sup>	
Spirit	27.4	67.9 <sup>abc</sup>	39.5 <sup>bcd</sup>	87.1 <sup>abc</sup>	56.3 <sup>b</sup>	
Vigomax	36.3	92.7 <sup>a</sup>	60.5 <sup>ab</sup>	96.5 <sup>ab</sup>	71.6 <sup>a</sup>	
Yedi	31.4	78.2 <sup>abc</sup>	67.9 <sup>a</sup>	106.9 <sup>ab</sup>	59.7 <sup>ab</sup>	
LSD (0.05)	ns	32.659	21.228	31.165	14.329	

ns: Non-significant. Different letters within columns indicate statistical differences ( $p < 0.05$ ).

environments. In both conditions, the shortest roots were obtained from non-grafted plants and *L. esculentum* rootstocks namely Resistar and Spirit followed them. The decrease in total root length compared to control plants under the damage of salinity was measured as 34.9% under 150 mM stress and 38.4% under 300 mM salinity stress (Table 6).

### Total dry weight

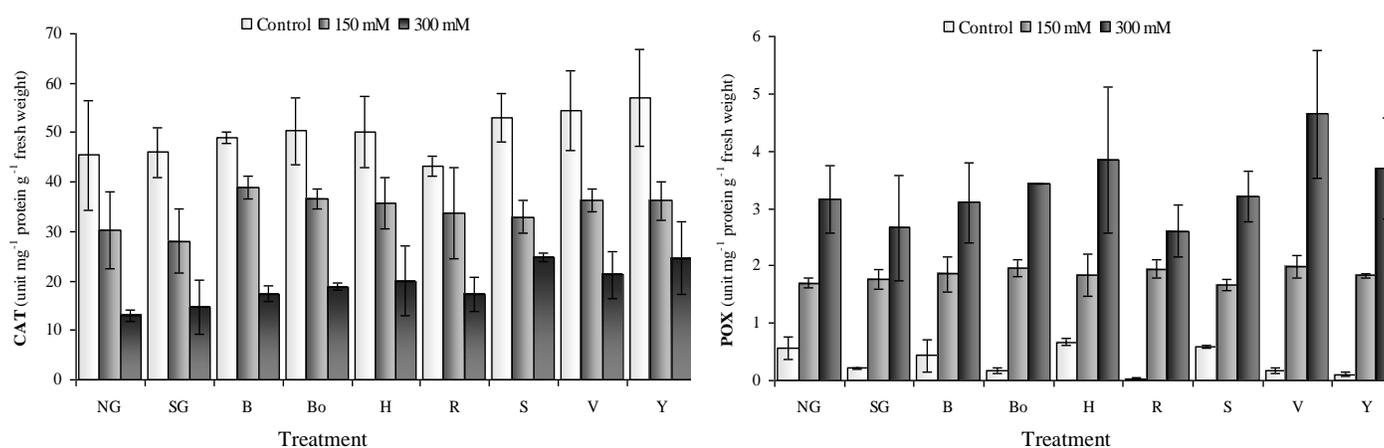
During the 1st removal, the treatment did not cause a statistical difference on total dry weight of the plant. During the 2nd removal, the highest total dry weight among salt-free plants was obtained with Yedi and Spirit

rootstocks and that under 150 mM salinity stress from Yedi rootstock. During the 3rd removal, Spirit and Beaufort rootstocks had the highest dry weight among salt-free plants; and Heman and Vigomax among those under 300 mM salinity stress. The lowest values at both removals were obtained from non-grafted plants, and then from self-grafted plants. *L. esculentum* rootstocks (Resistar and Spirit) showed the second lowest dry matter content although they were in same statistical group with other rootstocks except Vigomax. During the 2nd removal, the total average plant dry weight of control plants was 3.51 g, while this value decreased by 20.6% and became 2.78 g during 150 mM salinity application. During the 3rd removal, the mentioned values respectively appeared as 8.99 and 3.88 g with 56.8%

**Table 7.** Changes on total dry weight of seedling (g plant<sup>-1</sup>).

Treatment	1st removal		2nd removal		3rd removal	
	-NaCl	-NaCl	-NaCl	+NaCl	-NaCl	+NaCl
Non-grafted	1.21	2.62 <sup>b</sup>	1.60 <sup>c</sup>	6.65 <sup>d</sup>	2.43 <sup>c</sup>	
Self-grafted	1.46	2.95 <sup>ab</sup>	2.00 <sup>c</sup>	8.15 <sup>cd</sup>	3.30 <sup>bc</sup>	
Beaufort	1.75	3.78 <sup>ab</sup>	2.84 <sup>b</sup>	10.04 <sup>a</sup>	3.98 <sup>ab</sup>	
Body	1.73	3.74 <sup>ab</sup>	3.01 <sup>ab</sup>	9.32 <sup>abc</sup>	4.33 <sup>ab</sup>	
Heman	1.50	3.34 <sup>ab</sup>	2.92 <sup>b</sup>	8.75 <sup>abc</sup>	4.68 <sup>a</sup>	
Resistar	1.68	3.31 <sup>ab</sup>	2.80 <sup>b</sup>	8.42 <sup>bc</sup>	3.78 <sup>ab</sup>	
Spirit	1.78	3.94 <sup>a</sup>	3.08 <sup>ab</sup>	10.23 <sup>a</sup>	3.81 <sup>ab</sup>	
Vigomax	1.62	3.69 <sup>ab</sup>	3.19 <sup>ab</sup>	9.49 <sup>abc</sup>	4.61 <sup>a</sup>	
Yedi	1.74	4.16 <sup>a</sup>	3.59 <sup>a</sup>	9.85 <sup>ab</sup>	3.99 <sup>ab</sup>	
LSD (0.05)	ns	1.242	0.585	1.566	1.269	

ns: Non-significant. Different letters within columns indicate statistical differences ( $p < 0.05$ ).



**Figure 2.** Effects of NaCl concentration of nutrient solution on leaf CAT and POX activities. NG: Non-grafted, SG: Self-grafted, B: Beaufort, Bo: Body, H: Heman, R: Resistar, S: Spirit, V: Vigomax, Y: Yedi. Standard error of means are shown as vertical bars.

decrease (Table 7).

### Antioxidant enzyme activity

CAT enzyme activity varied depending on the use of rootstock, while the highest values were obtained with Yedi rootstock under salt-free conditions and from Beaufort and Spirit rootstocks respectively, under 150 and 300 mM salt-treated conditions; and the lowest values were obtained with non-grafted and self-grafted plants respectively. About CAT enzyme activity, 25.6, 28.3 and 90.1% increases compared to non-grafted plants, and 24.3, 38.6 and 68.2% increases compared to self-grafted plants were obtained with Beaufort and Spirit rootstocks respectively under salt-free, 150 and 300 mM NaCl levels. The average CAT enzyme activity of the

salt-free plants was identified as 49.8 units mg<sup>-1</sup> protein g<sup>-1</sup> fresh weight; the enzyme activity under 150 mM NaCl conditions decreased by 31.1%, and that under 300 mM NaCl conditions by 61.7% (Figure 2).

The activity of the POX enzyme employed during the removal of H<sub>2</sub>O<sub>2</sub> produced by chloroplast was identified as 0.327, 1.839 and 3.371 unit mg<sup>-1</sup> protein g<sup>-1</sup> fresh weight, respectively during salt-free, 150 and 300 mM NaCl levels, and POX enzyme activity at 300 mM salinity concentration occurred at 1.8 and 10.3% times higher compared to 150 mM and salt-free conditions respectively. The increase of POX activity due to salinity was more with rootstock plants compared to others and the highest POX activity under 150 and 300 mM NaCl level was obtained from Vigomax rootstock. With the use of Vigomax rootstock, 16.8 and 12.3% in 150 mM NaCl level compared to non-grafted and self-grafted

plants, and 47.3 and 74.4% higher enzyme activity at 300 mM level were obtained compared to the use of non-grafted and self-grafted plants respectively (Figure 2).

## DISCUSSION

The results obtained from the study have shown that the non-grafted and self-grafted Gokce was more impacted by NaCl treatment compared to grafted plants. It is known that grafted plants are able to uptake more water and nutrients from the root environment compared to non-grafted or self-grafted plants due to grafted plants' stronger and denser root structure, which increases internal plant hormones and as a result the rate of photosynthesis, thus inciting plant growth and development, and positively contributing to resistance against stress conditions (Lee, 1994; Ahn et al., 1999; Cohen et al., 2002). Nevertheless, the performances of rootstocks in terms of resistance against salinity stress also differed compared to each other. Among the rootstocks, interspecific tomato hybrids (*L. esculentum* × *L. hirsutum*) especially Vigomax and Yedi were least affected by NaCl treatment than tomato hybrids (*L. esculentum*). This reaction of grafted plants is associated with both the genetic structure, root characteristics and rootstock/scion compatibility, growth period and growth method (Lee, 1994; Romero et al., 1997; Leonardi and Romano, 2004). Among the tomato hybrids rootstocks, Spirit showed more salinity tolerance than Resistar. This result may be associated with different responses such as inclusion or exclusion mechanism to salinity of *L. esculentum* genotypes (Perez-Alfocea et al., 1993).

Under stress conditions, the biomass values which increase depending on the rootstock genotype (Chouka and Jebari, 1999; LiFei et al., 2006; GuWen et al., 2006), became one of the reasons for the gradual expansion of grafting technique employment in vegetable growing (Rivero et al., 2003; Khah, 2005). Bletsos et al. (2003) indicated that depending on the increase in plant development, the grafted plants showed a better vigor compared to non-grafted plants with regard to the plant length and stem diameter. Also in this study, the use of rootstock increased plant vigor under salinity stress; the decline in total plant dry weight in low and high salt concentrations decreased with rootstock use. This is supported by the previous studies which indicate that salt tolerance may be increased by grafting especially the rootstock genotype which are resistant to salt (Santa-Cruz et al., 2002; Martinez-Rodriguez et al., 2002; Estan et al., 2005).

Degradations are known in the characteristics of the plants grown (Shannon and Grieve, 1999; Romero-Aranda et al., 2001; Heuvelink et al., 2003) and in their total root lengths (Bourgeais and Guerrier, 1992; Sweby et al., 1994) under salty conditions. This can also be explained with the osmotic dehydration and increasing

transpiration which occur as a result of the osmotic stress caused on the plant by the increasing NaCl concentration. Osmotic dehydration decreases the cell's water and osmotic potential, and causes a reduction in cell volume and expansion rate. As a result of increasing transpiration, the drying of the plants buds and leaves results to a loss of weight (Lewit, 1980). During the study, decreases in leaf number, seedling length and leaf area (Perez-Alfocea et al., 1993; Chouka and Jebari, 1999; Bletsos et al., 2003; Khah, 2005) have been observed under salinity stress. The previous studies conducted on tomato indicating that the dry weight of plant decreases in salty conditions also support our findings (Al-Karaki, 2000; Santa-Cruz et al., 2002; Schwarz 2003).

High CAT activity obtained from rootstock plants under salty conditions indicate that rootstocks act as an agent to keep active oxygen radicals (Hea et al., 2009). Zhu et al. (2004) stated that CAT activity, as one of the effective enzymes in preventing cellular impacts in a manner similar to that in our findings, decreases together with the increase in salinity. Singha and Choundri (1990) indicated that H<sub>2</sub>O<sub>2</sub> quantity on the leaves of rice increased as a result of reducing CAT activity due to salinity. Higher POX level obtained under salty conditions indicates that the plant is capable of removing H<sub>2</sub>O<sub>2</sub> from the environment in a quicker manner (Kraus and Fletcher, 1994; Upadhyaya et al., 1989). In addition, the increase in POX activity under salty conditions is a selection criteria used for the identification of the species resistant or sensitive to salt (Shalata and Tal, 1998) and it is possible to say that mainly Vigomax rootstock with high activity at 300 mM salinity level, and Yedi and Heman rootstocks are prospective in this regard.

Evaluation of all findings obtained from the research suggest that grafting may be a valid strategy that can be used for increasing salt tolerance of tomato (Khah, 2005; Colla et al., 2006; Oztekin et al., 2007). Among the used rootstocks, interspecific tomato hybrids (*L. esculentum* × *L. hirsutum*) mainly Vigomax and Yedi have been identified as more resistant against salinity stress compared to other *L. esculentum* × *L. hirsutum* and *L. esculentum* genotypes due to the higher performances they represented in the measured parameters.

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