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Full Length Research Paper

Effects of salt stress on pigment and total soluble protein contents of three different tomato cultivars

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In this study, the effects of salt stress on pigment and total soluble protein contents were investigated in different varieties of tomato (*Lycopersicon esculentum* Mill.). The seedlings of *L. esculentum* viz. Hazera, Dalli Tokat and Argy were treated with NaCl at 25, 50, 100, 125, 150 and 200 mM concentrations for 96 h with 24 h interval. Pigment and total soluble protein contents of all tomato cultivars were significantly decreased by salt stress depending on time intervals and salt concentrations. Decreasing of pigment and total soluble protein contents were more evident in Hazera under short time salt exposure. Pigment content of Argy plants were less affected by salt concentration and exposure time. The results of this study suggest that the Argy cultivars are relatively better protected under salt stress conditions than Dalli tokat and Hazera cultivars.

Key words: Salt stress, *Lycopersicon esculentum* Mill., chlorophyll, carotenoid, total soluble protein.

INTRODUCTION

Salty soils extensively exist in arid and semi-arid climate regions of the world and cause salt stress in plants (Khan et al., 2001). Salinity is an important abiotic stress factor seriously affecting plant productivity and survival (Eker et al., 2006). Plants can be classified into two groups, glycophytes and halophytes according to their survival ability in salinity conditions (Lee et al., 2005). Growth and development of glycophytes are negatively affected but halophytes tolerate high salt concentrations (Parvaiz and Satyavati, 2008).

Salinity is an environmental stress and high ion concentration in rhizosphere, ion toxicity and water limitation are major causes of stress. In general, salt stress is sourced by Na salts, especially NaCl. High concentration of soluble salts in growing media causes salt stress and plants exposure to water scarcity due to physiological drought. When a plant is exposed to salt stress, chemical potential, activity and salt concentration is higher than normal limits (Porgali, 2001).

Changes in protein hydration are one of the results of high ion amounts in salt stress in plant cells. Salinity reduces both RNA amounts due to changes in cytoplasmic RNAaz activity and DNA levels as a result of disruption of synthesis mechanism. The reason for decrease in the leaf area in *Phaseolus vulgaris* plants subjected to NaCl was connected with inhibition of RNA-protein metabolic pathway (Yu and Rengel, 1999). Total soluble protein content was not affected in *Lupinus angustifolius* plant exposed to both drought and salt stress, but the decrease in protein content was shown in root, young and old leaves of *Helianthus annuus* and *Coleus blumei* plants (Dos Santos et al., 1999; Gilbert et al., 1998; Yu and Rengel, 1999). Similarly the decline in total soluble protein content was showed in *Lycopersicon esculentum*, *Oryza sativa*, *Vicia faba, Amaranthus tricolor* and *Brugiera parviflora* plants under NaCl stress (Al-aghabary et al., 2004; Alamgir et al., 1999; Gadallah, 1999; Parida and Das, 2005; Parvaiz and Satyavati, 2008; Wang and Nil, 2000). Increase in total soluble protein content was reported at high NaCl concentration

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and decrease at low concentration in *Pancratium maritimum* plants (Khedr et al., 2003). In contrast to this, plants increases in protein contents were reported in *Arabidopsis thaliana* and *Fragaria* × *ananassa* cv. *Camarosa* (El-Baz et al., 2003).

Net photosynthesis, transpiration rate and stomatal conductance are significantly affected by salt stress due to changes in chlorophyll content and chlorophyll fluorescence, damage of photosynthetic apparatus and chloroplast structure (Abd El Baki et al., 2000; Fidalgo et al., 2004; Kao et al., 2003; Pinheiro et al., 2008). It was reported that both chlorophyll a (Chl-a) and chlorophyll b (Chl-b) amounts decreased with NaCl application in *Zea mays* and *Carthamus tinctorius* plants (Sepehr and Ghorbali, 2006, Siddiqi et al., 2009). However, in *Hibiscus esculentus* plants exposed to salt stress, significant increases were only found in Chl-a content but Chl-b was not significantly affected by salt stress (Ashraf et al., 2003). Chl-a, Chl-b, Chl-a/b and carotenoid (Car) contents showed increase and decrease depending on exposure time of NaCl exposure in *Ricinus communis* plants (Pinheiro et al., 2008). Some physiological responses to salt stress have been used in determining salt tolerance of plants. Plant hormone levels (Yurekli et al., 2001, 2004), antioxidant enzyme activities (Yurekli and Porgali, 2006), pigment contents (Ashraf et al., 2003; Essa and Dawood, 2001; Sarwat and El-Sherif, 2007; Siddiqi et al., 2009), osmotic potential reduction (Cha-um et al., 2009), gas exchange characteristics (Ashraf et al., 2003), total soluble protein contents (Demiral and Turkan, 2006; Yurekli and Porgali, 2006) and proline (Essa and Dawood, 2001; Porgali and Yurekli, 2005) amounts were determined in different tolerant and sensitive plant varieties at wide range of salt concentrations.

The aims of the present work were (1) to determine effects of salt stress on pigment (Chl-a, -b, and Car) and total soluble protein contents in *L. esculentum* viz. Hazera, Dalli Tokat and Argy cultivars (2) to find the differences of salt response in these cultivars.

MATERIALS AND METHODS

Plant material and growth conditions

In this study *L. esculentum* viz. Hazera, Dalli Tokat and Argy were used in experimental analyses. These cultivars were grown at 26±2°C and 60±10% relative humidity and daylight photoperiod condition in seedling manufactory (Zirve Seedling, Fethiye, Turkey) for 20 days and then transported to greenhouse of Ahi Evran University. During the study the average temperature and photoperiod were 19±1 (night) /23±1°C (day), 14/10 h light/dark, respectively and relative humidity varied between 60 - 70%. After this time all seedlings were transferred to Ecomat Mulching, made from oil palm fibres (Ecofibre technology, Malaysia). Plants were irrigated with Hoagland nutrient solution 50 ml/per plant daily until it was 30 days old. After planting, the Ecomat were exposed to distilled water containing 0 (control), 25, 50, 75, 100, 125, 150 and 200 mM NaCl respectively for 96 h with 24 h interval and then pigment and total soluble protein contents were determined in leaf tissues of tomato cultivars. Plant leaves were harvested at 24, 48, 72 and 96 h respectively and were stored at -80°C for the next step of chemical analysis.

Total soluble protein content assay

Total soluble protein content was measured according to Bradford (1976) using bovine serum albumin (BSA) as a protein standard. Fresh leaf samples (1 g) were homogenized with 4 ml Na-Phosphate buffer (pH 7.2) and then centrifuged at 4°C. Supernatants and dye were pipetting in spectrophotometer cuvettes and absorbances were measured using a Uv-vis spectrophotometer (PG instruments T80) at 595 nm.

Pigment content assay

Chlorophyll-a, chlorophyll-b and carotenoid content assays were performed according to Arnon (1949). 200 mg fresh leaves were homogenized in 8 ml 80% acetone with homogenizer. Homogenates were centrifuged at 4°C for 15 min (3000 rpm). Supernatants were used for the analysis of pigments. Absorbances were determined at 645, 652, 663 and 470 nm respectively and the following equations were used for calculations (Litchtenthaler and Wellburn, 1983).

Total Chl: $A_{652} \times 27.8 \times 20$ /mg leaf weight
Chl-a: $(11.75 \times A_{663}-2.35 \times A_{645}) \times 20$ /mg leaf weight
Chl-b: $(18.61 \times A_{645}-3.96 \times A_{663}) \times 20$ /mg leaf weight
Car: $[(1000 \times A_{470}-2.27 \times$ Chla$-81.4 \times$ Chl b$)/227] \times 20$ /mg leaf weight

Statistical analysis

Differences in pigment and total soluble protein contents in leaf tissues belonging to different tomato cultivars at eight NaCl concentrations were determined using ANOVA, with means separation by Duncan's test using SPSS 15 software at a significance level $P \leq 0.05$.

RESULTS AND DISCUSSION

Pigment content

In this study, the responses of three different tomato cultivars to salt stress were compared with regard to pigment and total soluble protein. Although, pigment contents of seedlings decreased considerably with the addition of NaCl on the growth media, protein contents did not show same changes in three varieties of tomato. The changes in pigment contents were affected by exposure time and salt concentrations. Total Chl, Chl-a, Chl-b and Car contents of tomato plants under NaCl stress are shown in Tables 1 - 4. Compared to control, total chlorophyll contents decreased by all of NaCl concentrations (except for 150 mM) in Hazera cv. (Figure 1). Maximum decrease in total chlorophyll content was shown at 50 mM NaCl concentration (68%) and this was followed by 25 mM NaCl application in Hazera. In contrast to Hazera, total chlorophyll contents of Dalli Tokat and Argy cultivars increased at 24 h of NaCl

Table 1. Total soluble protein and pigment (Total Chl, Chl-a, Chl-b and Car) contents of tomato plants under NaCl stress at 24 h.

	Cultivars	NaCl concentrations (mM)							
		Control	25	50	75	100	125	150	200
Total protein	Hazera	72.86±5,86^{b}	60.29±0.99^{a}	60.86±1.84^{a}	61.24±1.82^{a}	62.38±1.01^{a}	62.00±0.33^{a}	63.33±1.49^{a}	60.67±0.95^{a}
	D. Tokat	58.38±1.01^{a}	64.67±0.69^{a}	63.33±3.07^{a}	58.76±1.01^{a}	59.14±2.31^{a}	58.19±1.63^{a}	60.67±0.83^{a}	58.38±0.76^{a}
	Argy	67.52±1.49^{a}	64.67±1.16^{a}	59.71±1.65^{a}	62.19±1.33^{a}	56.86±1.51^{a}	65.24±3.82^{a}	58.38±4.55^{a}	60.67±2.39^{a}
Total Chl	Hazera	3.56±0.49^{A,b}	2.98±0.37^{A,b}	2.12±0.36^{A,a}	3.37±0.06^{A,b}	3.23±0.25^{A,b}	3.53±0.20^{B,b}	3.70±0.16^{B,b}	3.11±0.07^{A,b}
	D. Tokat	3.01±0.36^{A,ab}	4.42±0.70$^{A.c}$	3.70±0.14^{B,bc}	3.89±0.14^{B,bc}	3.19±0.18^{A,ab}	3.41±0.10^{B,ab}	2.51±0.08^{A,a}	3.18±0.12^{A,ab}
	Argy	2.71±0.08^{A,a}	3.06±0.14$^{A.b}$	3.14±0.12^{B,b}	3.07±0.14^{A,b}	3.50±0.06^{A,c}	3.04±0.06^{A,b}	3.54±0.15^{B,c}	3.64±0.06^{B,c}
Chl-a	Hazera	2.13±0.23^{A,b}	1.85±0.17^{A,b}	1.34±0.22^{A,a}	2.06±0.02^{A,b}	2.02±0.13^{A,b}	2.14±0.10^{B,b}	2.23±0.07^{B,b}	1.94±0.03^{A,b}
	D.Tokat	1.90±0.19^{A,b}	2.20±0.17^{A,bc}	2.24±0.04^{C,c}	2.30±0.06^{B,c}	1.97±0.08^{A,bc}	2.09±0.05^{B,bc}	1.57±0.05^{A,a}	1.98±0.07^{A,bc}
	Argy	1.71±0.05^{A,a}	1.92±0.08^{A,b}	1.94±0.05^{B,b}	1.93±0.08^{A,b}	2.16±0.03^{A,c}	1.91±0.03^{A,c}	2.14±0.07^{B,c}	2.23±0.03^{B,c}
Chl-b	Hazera	0.92±0.16^{A,a}	0.73±0.12^{A,a}	0.52±0.09^{A,a}	0.85±0.03^{A,a}	0.78±0.07^{A,a}	0.89±0.06^{B,a}	0.94±0.05^{B,a}	0.76±0.02^{A,a}
	D. Tokat	0.72±0.10^{A,a}	1.37±0.31^{A,b}	0.93±0.05^{B,a}	1.02±0.04^{B,a}	0.80±0.06^{A,a}	0.86±0.03^{B,a}	0.63±0.02^{A,a}	0.79±0.04^{A,a}
	Argy	0.67±0.02^{A,a}	0.75±0.04^{A,ab}	0.80±0.04^{B,bc}	0.74±0.04^{A,ab}	0.86±0.02^{A,cd}	0.74±0.02^{A,a}	0.91±0.05^{B,d}	0.89±0.02^{B,cd}
Car	Hazera	2.38±0.19^{A,a}	2.58±0.17^{A,a}	2.55±0.01^{A,a}	2.42±0.06^{A,a}	2.59±0.07^{A,a}	2.41±0.06^{B,a}	2.38±0.06^{B,a}	2.55±0.04^{A,a}
	D. Tokat	0.72±0.08^{A,ab}	1.00±0.13^{A,d}	0.87±0.02^{C,bcd}	0.95±0.03^{B,cd}	0.78±0.03^{A,abc}	0.84±0.02^{A,bcd}	0.64±0.02^{A,a}	0.79±0.03^{A,abc}
	Argy	0.65±0.02^{A,cd}	0.73±0.03^{A,cd}	0.75±0.02^{B,ab}	0.71±0.04^{A,d}	0.83±0.01^{A,bc}	0.73±0.01^{B,cd}	0.84±0.03^{B,a}	0.81±0.01^{A,bcd}

*****Means followed by the capital letters in same rows do not differ significantly at P ≤0.05 (resulted by Oneway-ANOVA, separated by Duncan test). ** Means followed by the lower case in same columns do not differ significantly at P ≤0.05 (resulted by Oneway-ANOVA, separated by Duncan test).

applications (except for 150 mM NaCl concentration in Dalli Tokat).

While the lowest level of total chlorophyll in Dalli Tokat was determined at 150 mM, all other applications caused increase in total chlorophyll contents compared to control. Statistically significant differences were only observed at 25, 50 and 75 mM of salt application. Maximum and minimum changes of total chlorophyll were determined at 25 and 200 mM of salt concentration in Dalli Tokat, respectively (Table 1, Figure 2). As seen in Table 1, compared to control, salt treatment caused a marked increase in total chlorophyll contents of Argy plants 24 h after NaCl exposure ($p \leq 0.05$). Total chlorophyll content was found to increase in all treatments ranging from 11 - 26% at 25 and 200 mM exposure and this fluctuation were significantly different from the control at 24 h (Table 1).

Application of salt stress caused a significant increase in Chl-a content of Dalli Tokat and Argy cvs. However, Chl-b contents showed significant increases in Argy(except at 125 mM) and only at 25 mM salt application in Dalli Tokat ($p \leq 0.05$). Maximum increases of Chl-a and Chl-b contents were determined as 23% at 200 mM in Argy and 47% at 25 mM in Dalli Tokat.

The most affected variety was found in Hazera for 48 h of salt stress. In this time the total chlorophyll content was significantly found in low level compared to control at all seven applications and these reductions were calculated as 14 - 41% in salt stress seedling. In Dalli Tokat, increasing total chlorophyll until 75 mM application was followed by a fluctuating trend in other salt treatments. As compared to control, the total chlorophyll showed 1.3 - 2.5 fold increase in Argy seedling. Chl-a, Chl-b and Car contents decreased in Hazera while it increased in Argy plants. However Chl-a,

Table 2. Total soluble protein and pigment (total Chl, Chl a, Chl b and Car) contents of tomato plants under NaCl stress at 48 h.

	Cultivars	NaCl concentrations (mM)							
		Control	25	50	75	100	125	150	200
Total protein	Hazera	62.57±0.99^{e}	59.14±0.87^{d}	55.33±1.16^{bc}	55.14±0.87^{bc}	53.81±0.38^{b}	57.81±0.19^{cd}	47.71±1.44^{a}	52.48±0.76^{b}
	D. Tokat	60.10±0.69^{e}	56.09±0.76^{d}	53.24±0.69^{abc}	56.29±0.66^{d}	53.81±0.50^{c}	53.62±1.06^{bc}	51.33±1.06^{ab}	50.95±0.38^{a}
	Argy	59.33±1.06^{c}	59.71±0.33^{c}	50.19±1.10^{a}	53.81±0.50^{b}	50.00±0.99^{a}	59.71±1.51^{c}	54.19±0.69^{b}	55.71±0.22^{b}
Total Chl	Hazera	4.29±0.13^{B,d}	3.17±0.09^{A,b}	2.93±0.17^{A,ab}	3.71±0.15^{A,c}	3.23±0.07^{A,b}	3.31±0.20^{A,bc}	2.57±0.12^{A,a}	2.69±0.09^{A,a}
	D. Tokat	3.15±0.08^{A,cd}	4.35±0.19^{A,e}	3.63±0.31^{B,d}	2.60±0.06^{A,bc}	1.82±0.03^{A,a}	1.96±0.16^{A,ab}	4.50±0.05^{A,e}	2.54±0.19^{B,bc}
	Argy	2.26±0.17^{A,a}	2.97±0.07^{A,b}	3.43±0.10^{A,cd}	4.16±0.12^{A,e}	3.26±0.07^{A,bc}	5.70±0.03^{B,f}	3.81±0.15^{B,de}	3.91±0.18^{C,e}
Chl-a	Hazera	2.41±0.03^{B,e}	1.99±0.05^{A,bc}	1.84±0.10^{A,ab}	2.24±0.05^{A,de}	2.04±0.03^{A,c}	2.06±0.09^{A,cd}	1.65±0.07^{A,a}	1.73±0.05^{A,a}
	D.Tokat	1.95±0.04^{A,c}	2.50±0.04^{A,d}	2.18±0.13^{B,c}	1.65±0.04^{A,b}	1.14±0.02^{A,a}	1.21±0.10^{A,a}	2.51±0.01^{A,d}	1.62±0.13^{B,b}
	Argy	1.46±0.11 ^{A,a}	1.85±0.04^{A,b}	2.10±0.05^{B,bcd}	2.39±0.05^{A,ef}	2.03±0.04^{A,bc}	2.56±0.04^{B,f}	2.26±0.06^{B,cde}	2.31±0.07^{C,de}
Chl-b	Hazera	1.16±0.06^{B,d}	0.77±0.03^{A,b}	0.72±0.04^{A,ab}	0.93±0.05^{A,c}	0.77±0.02^{A,b}	0.81±0.07^{A,bc}	0.61±0.03^{A,a}	0.63±0.02^{A,a}
	D. Tokat	0.80±0.02^{A,bc}	1.17±0.08 ^{A,d}	0.94±0.10^{A,c}	0.63±0.02^{A,ab}	0.46±0.01^{A,a}	0.51±0.04^{A,a}	1.26±0.03^{A,d}	0.62±0.04^{B,ab}
	Argy	0.53±0.04^{A,a}	0.74±0.02^{A,b}	0.87±0.03^{A,c}	1.11±0.04^{A,d}	0.81±0.02^{A,bc}	1.88±0.05^{B,e}	1.00±0.05^{B,d}	1.01±0.06^{C,d}
Car	Hazera	0.94±0.01^{B,d}	0.76±0.02^{A,b}	0.72±0.04^{A,b}	0.86±0.02^{A,c}	0.78±0.01^{A,b}	0.77±0.03^{A,b}	0.64±0.02^{A,a}	0.65±0.02^{A,a}
	D. Tokat	0.75±0.02^{A,b}	1.05±0.03^{A,c}	0.91±0.07^{B,c}	0.68±0.01^{A,b}	0.46±0.01^{A,a}	0.52±0.04^{A,a}	1.04±0.01^{A,c}	0.54±0.03^{B,a}
	Argy	0.53±0.04^{A,a}	0.73±0.01^{A,b}	0.86±0.02^{B,cd}	0.93±0.02^{A,d}	0.79±0.01^{A,bc}	1.16±0.02^{B,e}	0.93±0.03^{B,d}	0.94±0.03^{C,d}

*****Means followed by the capital letters in same rows do not differ significantly at P ≤0.05 (resulted by Oneway-ANOVA, separated by Duncan test). ** Means followed by the lower case in same columns do not differ significantly at P ≤0.05 (resulted by Oneway-ANOVA, separated by Duncan test).

Chl-b and Car contents of Dalli Tokat were followed fluctuating trend by different salt concentrations. Salt stress was less effective on pigment contents of Argy plants compared to control group; maximum increases in Chl-a, Chl-b and Car contents were detected as 43, 72 and 55% at 125 mM NaCl concentration, respectively (Table 2). While maximum reduction in Chl-b contents were determined in Hazera (89% at 150 mM), maximum decreases in Chl-a and Car contents were determined in Dalli Tokat (71 and 63% at 100 mM).

As seen in Table 3, compared to control, NaCl treatments caused statically significant reductions in total Chl contents of Dalli Tokat and Argy cultivars at 72 h after treatment (except for Dalli Tokat at 25 mM). The lowest levels of total Chl contents were found in Dalli Tokat at 150 mM (155%) and 75 mM (103%) concentrations. While statically significant increases were determined in Hazera at 25 and 100 mM salt concentrations, other applications caused significant reductions in total Chl content ($p \leq 0.05$) (Figure 3).

Although 25, 100, 125 and 150 mM salt treatments significantly increased Chl-b content, Chl-a and Car contents, it showed significant increases only at 25 and 100 mM salt treatment in Hazera plants. This is different from Hazera, Chl-a, Chl-b and Car contents of Dalli Tokat and Argy plants as they significantly decreased at 72 h after treatment (except for at 25 mM in Dalli Tokat). 96 h after salt treatment, the reductions in total Chl content were shown at only 125 and 200 mM NaCl treatment while other salt concentrations caused statistically significant increase ($p \leq 0.05$). There were maximum decreases in Dalli Tokat

Table 3. Total soluble protein and pigment (total Chl, Chl-a, Chl-b and Car) contents of tomato plants under NaCl stress at 72 h.

	Cultivars	NaCl concentrations (mM)							
		Control	25	50	75	100	125	150	200
Total Protein	Hazera	46.19±5.44^{ab}	52.29±2.93^{b}	42.95±2.02^{a}	45.43±3.49^{ab}	43.71±0.66^{ab}	47.90±1.25^{ab}	46.54±0.57^{ab}	49.66±1.78^{ab}
	D. Tokat	72.14±1.98^{c}	62.33±1.84^{abc}	67.52±1.06^{bc}	54.76±3.80^{ab}	57.43±13.51^{abc}	54.00±0.87^{ab}	52.86±0.87^{ab}	49.62±1.33^{a}
	Argy	52.67±7.84^{ab}	43.90±2.50^{ab}	42.57±0.87^{ab}	49.62±0.19^{ab}	47.90±5.69^{ab}	39.14±2.16^{a}	63.14±15.72^{b}	58.19±1.90^{ab}
Total Chl	Hazera	2.44±0.05^{A,c}	3.68±0.18^{B,d}	1.80±0.04^{A,a}	2.15±0.10^{B,b}	4.19±0.10^{C,e}	2.20±0.08^{A,bc}	1.87±0.01^{B,a}	1.69±0.04^{A,a}
	D. Tokat	3.56±0.14^{B,d}	3.31±0.05^{B,d}	2.53±0.05^{B,c}	1.76±0.12^{A,b}	1.95±0.00^{A,b}	2.49±0.15^{A,c}	1.39±0.05^{A,a}	2.33±0.07^{B,c}
	Argy	3.93±0.06^{C,e}	2.50±0.05^{A,a}	3.30±0.07^{C,d}	2.89±0.06^{C,c}	2.93±0.04^{B,c}	2.72±0.05^{B,b}	2.86±0.03^{C,bc}	2.92±0.02^{C,c}
Chl-a	Hazera	1.59±0.03^{A,d}	2.20±0.08^{B,e}	1.14±0.03^{A,b}	1.38±0.06^{A,c}	2.25±0.05^{C,e}	0.90±0.04^{A,a}	0.78±0.01^{A,a}	1.08±0.03^{A,b}
	D.Tokat	2.18±0.06^{B,d}	2.04±0.02^{B,d}	1.64±0.03^{B,c}	1.11±0.08^{B,b}	1.22±0.00^{A,b}	1.57±0.09^{B,c}	0.87±0.03^{B,a}	1.49±0.04^{B,c}
	Argy	2.32±0.03^{B,e}	1.60±0.03^{A,a}	2.04±0.04^{C,d}	1.83±0.04^{C,bc}	1.85±0.02^{B,c}	1.74±0.03^{B,b}	1.82±0.02^{C,bc}	1.88±0.02^{C,c}
Chl-b	Hazera	0.56±0.01^{A,b}	0.94±0.06^{B,c}	0.44±0.01^{A,ab}	0.51±0.02^{A,ab}	1.37±0.02^{C,d}	1.78±0.09^{B,f}	1.34±0.01^{C,e}	0.41±0.01^{A,a}
	D. Tokat	0.89±0.04^{B,e}	0.84±0.02^{B,e}	0.59±0.01^{B,d}	0.44±0.03^{A,b}	0.50±0.00^{A,bc}	0.61±0.04^{A,d}	0.36±0.01^{A,a}	0.57±0.02^{B,cd}
	Argy	1.02±0.01^{C,f}	0.60±0.01^{A,a}	0.83±0.02^{C,e}	0.71±0.01^{B,cd}	0.72±0.01^{B,d}	0.65±0.01^{A,b}	0.69±0.01^{B,bcd}	0.68±0.00^{C,bc}
Car	Hazera	0.60±0.01^{A,ab}	0.83±0.03^{B,bc}	0.47±0.01^{A,a}	0.52±0.02^{A,ab}	0.93±0.02^{C,c}	0.44±0.27^{A,a}	0.62±0.02^{B,ab}	0.44±0.01^{A,a}
	D. Tokat	0.84±0.02^{B,d}	0.82±0.00^{B,d}	0.60±0.01^{B,c}	0.51±0.03^{A,b}	0.51±0.00^{A,b}	0.60±0.03^{A,c}	0.40±0.01^{A,a}	0.61±0.02^{B,c}
	Argy	0.88±0.01^{B,e}	0.61±0.01^{A,a}	0.80±0.01^{C,d}	0.68±0.01^{B,b}	0.72±0.01^{B,c}	0.69±0.01^{A,bc}	0.70±0.01^{C,bc}	0.67±0.01^{C,b}

*****Means followed by the capital letters in same rows do not differ significantly at P ≤ 0.05 (resulted by Oneway-ANOVA, separated by Duncan test). ** Means followed by the lower case in same columns do not differ significantly at P ≤ 0.05 (resulted by Oneway-ANOVA, separated by Duncan test).

and Argy plants with the NaCl exposure determined at 200 mM (39 %) and 100 mM (34 %) salt concentrations, respectively (Table 4). Compared to control group, statistically significant differences were determined in total Chl contents of all tomato cultivars at 96 h (except for Dalli Tokat at 75 and 125 mM NaCl application).

Maximum decrease in Chl-a was seen in Dalli Tokat (45 %, at 200 mM), while Chl-b and Car contents were detected in Hazera at 200 mM (Table 4).

Chl (a and b)and Car are main photosynthetic pigments and they play important role in photosynthesis. The changes in the amount of pigment were evaluated as the changes in photosynthesis. Changes of pigment contents under salt stress are used as parameter for selection of tolerant and sensitive cultivars in crop plants (Eryilmaz, 2007). In this study, we determined the effect of salt stress on tomato varieties in both different concentrations and exposure times. Pigment responses of all tomato cultivars were found different from each other in different exposure times. At 24 h of salt treatment, pigment contents of Dalli Tokat and Argy plants continued to increase, but that of Hazera did not change. Increased total Chl content under salt stress was also observed in Cucumis sp. Galia F_1, wheat, broad bean and rice plants (Abd elSamad 1993a; 1993b; Kusvuran et al., 2008). The increased content of photosynthetic pigments was seen during plant growth. According to Misra et al., (1997) an increase in Chl content was observed in salt stressed plants and this may be due to an increase in the number of chloroplasts in the stressed leaves. It was thought that the

Table 4. Total soluble protein and pigment (total Chl, Chl-a, Chl-b and Car) contents of tomato plants under NaCl stress at 96 h.

	Cultivars	NaCl concentrations (mM)							
		Control	25	50	75	100	125	150	200
Total protein	Hazera	58.76±1.37^{bc}	61.62±0.50^{c}	57.81±1.06^{b}	55.52±0.19^{b}	58.57±0.33^{bc}	57.05±1.93^{b}	51.33±0.19^{a}	50.95±0.83^{a}
	D. Tokat	73.43±1.19^{cde}	72.29±0.87^{cd}	76.29±0.87^{e}	70.00±1.98$^{b\,c}$	74.95±2.12^{de}	70.95±0.19^{bc}	65.62±0.50^{a}	67.52±0.69^{ab}
	Argy	69.43±5.49^{b}	57.81±0.50^{a}	56.29±0.99^{a}	54.76±1.66^{a}	59.33±1.33^{a}	59.71±0.87^{a}	55.14±0.33^{a}	56.67±0.69^{a}
Total Chl	Hazera	2.80±0.09^{A,c}	3.16±0.10^{A,d}	3.15±0.15^{A,d}	4.08±0.08^{B,e}	4.71±0.15^{C,f}	2.31±0.04^{A,b}	3.40±0.14^{A,d}	1.90±0.03^{A,a}
	D. Tokat	2.85±0.04^{A,b}	3.18±0.04^{A,c}	3.22±0.12^{A,c}	2.09±0.03^{A,b}	4.17±0.05^{B,d}	2.69±0.05^{B,b}	3.04±0.05^{A,c}	2.04±0.01^{B,a}
	Argy	3.58±0.10^{B,c}	3.30±0.02^{A,b}	4.29±0.10^{B,d}	4.65±0.05^{C,e}	2.67±0.01^{A,a}	3.23±0.12^{C,b}	4.07±0.11^{B,d}	3.15±0.01^{C,b}
Chl-a	Hazera	1.78±0.06^{A,c}	1.96±0.06 ^{A,d}	1.95±0.09^{A,d}	2.39±0.02^{B,e}	2.50±0.05^{B,e}	1.48±0.02^{A,b}	2.08±0.07^{A,d}	1.23±0.02^{A,a}
	D.Tokat	1.85±0.03^{A,c}	2.02±0.02^{A,d}	2.04±0.07^{A,d}	1.32±0.02^{A,a}	2.41±0.02^{B,e}	1.69±0.03^{B,b}	1.93±0.03^{A,c}	1.32±0.01^{B,a}
	Argy	2.20±0.04^{B,c}	2.03±0.01^{A,b}	2.43±0.03^{B,de}	2.51±0.03^{C,e}	1.70±0.01^{A,a}	1.99±0.07^{C,b}	2.37±0.03^{B,d}	1.96±0.00^{C,b}
Chl-b	Hazera	0.68±0.02^{A,b}	0.79±0.03^{A,c}	0.79±0.04^{A,c}	1.05±0.03^{B,d}	1.33±0.06^{B,e}	0.54±0.01^{A,a}	0.85±0.04^{B,c}	0.45±0.01^{A,a}
	D. Tokat	0.66±0.01^{A,b}	0.76±0.01^{A,c}	0.78±0.04^{A,c}	0.53±0.01^{A,a}	1.12±0.02^{B,d}	0.68±0.01^{B,b}	0.73±0.01^{A,c}	0.49±0.00^{B,a}
	Argy	0.89±0.03^{B,c}	0.84±0.01^{A,bc}	1.16±0.04^{B,d}	1.29±0.01^{C,e}	0.65±0.00^{A,a}	0.82±0.03^{C,bc}	1.07±0.04^{C,d}	0.78±0.01^{C,b}
Car	Hazera	0.70±0.02^{A,b}	0.75±0.02^{A,bc}	0.76±0.03^{A,bc}	0.92±0.01^{B,d}	1.02±0.02^{B,e}	0.55±0.01^{A,a}	0.82±0.03^{B,c}	0.49±0.01^{A,a}
	D. Tokat	0.70±0.01^{A,b}	0.75±0.01 ^{A,c}	0.78±0.02^{A,c}	0.54±0.01^{A,a}	0.99±0.01^{B,d}	0.67±0.01^{B,b}	0.75±0.01^{A,c}	0.52±0.01^{B,a}
	Argy	0.85±0.02^{B,c}	0.79±0.00^{A,b}	0.97±0.02^{B,e}	0.98±0.01^{C,e}	0.66±0.00^{A,a}	0.80±0.02^{C,b}	0.96±0.01^{C,d}	0.76±0.01^{C,b}

*Means followed by the capital letters in same rows do not differ significantly at P ≤ 0.05 (resulted by Oneway-ANOVA, separated by Duncan test). ** Means followed by the lower case in same columns do not differ significantly at P ≤ 0.05 (resulted by Oneway-ANOVA, separated by Duncan test).

continuation of total Chl content of Argy plants under short time salt stress using NaCl treatments for 24 h did not caused a negative impact on total Chl content in Argy plants. This was because pigment synthesis continued in these plants. After 72 and 96 h of salt treatment, it was determined that Hazera plants acclimated to salt stress because this plant increased their pigment synthesis to regulate the adverse effect of salt stress. Thus, we can say that Hazera was affected by short term salt stress, but was overcome by long time exposure at 72 and 96 h. We thought that Argy was relatively less affected by salt application than Dalli Tokat and Hazera cvs based on Chl, Chl a and b and Car basis. Essa and Dawood (2001) reported that six soybean genotypes responded differently to salt stress. These researchers selected tolerant genotypes with regard to less reduction in chlorophyll content. According to Essa and Dawood (2001), chlorophyll contents could be a parameter for selecting salt tolerant genotypes of soybean. Similarly, Siddiqi et al. (2009) reported that only net CO_2 assimilation rate was the most effective indicator for salt tolerant genotype selection in spite of salinity caused decreases on leaf Chl a and b contents of ten safflower accessions. Pigment contents were reduced by salt applications in proso millet (*Panicum miliaceum*) accessions which have different salt tolerance characteristics and it was reported that Chl-a contents can be related to the salt tolerance of *P. miliaceum* (Sabir et al., 2009).

Total soluble protein content

Compared to control, salt treatment during 24 h caused significant decrease of total soluble

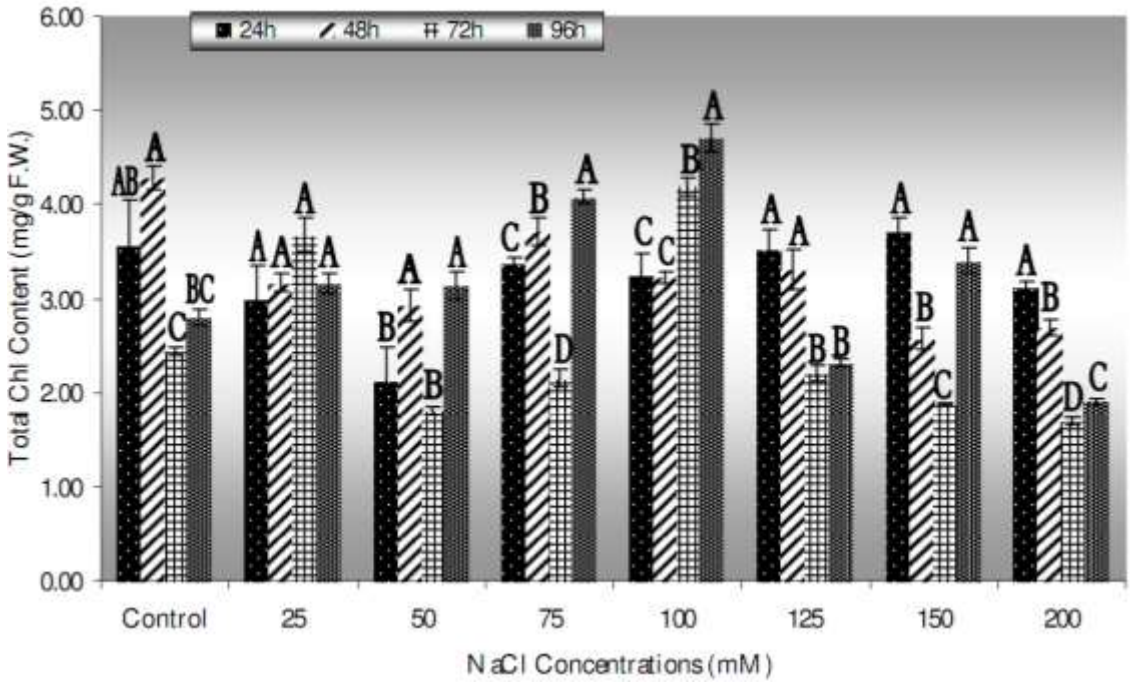

Figure 1. Effect of NaCl concentrations on total Chl content of *L. esculentum* Mill. cv. Hazera.

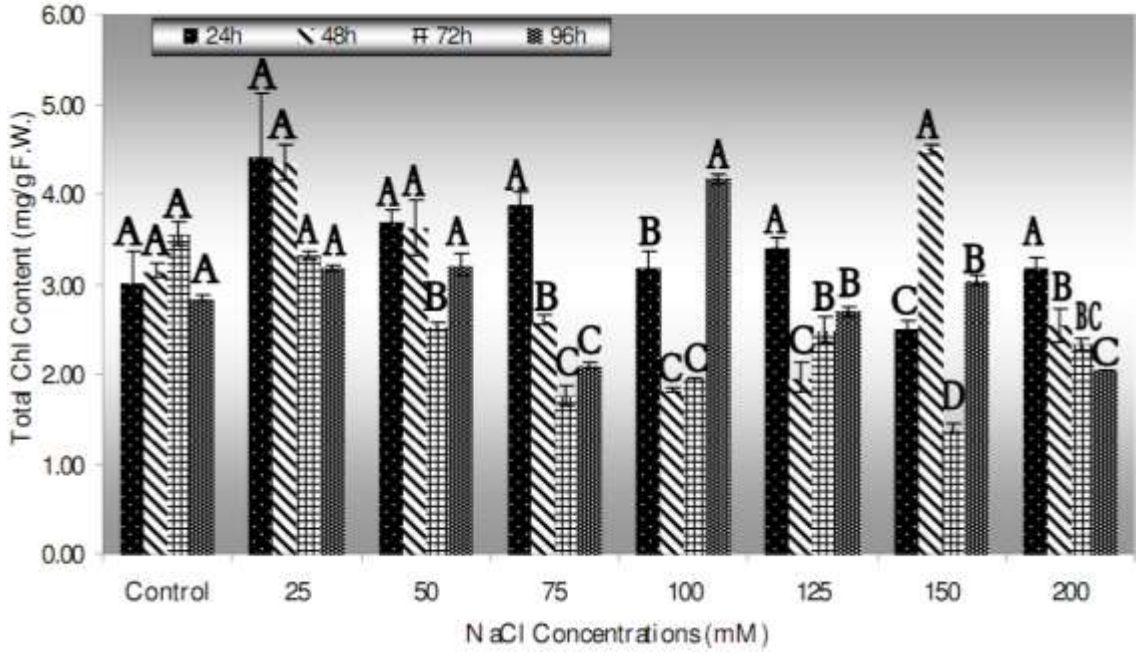

Figure 2. Effect of NaCl concentrations on total Chl content of *L. esculentum* Mill. cv. Dalli Tokat.

protein content only in Hazera plants ($p \leq 0.05$) and maximum decrease in total soluble protein content was determined at 25 and 200 mM NaCl concentrations (Table 1). As seen in Table 2, after 48 h salt application, total soluble protein contents of all tomato cultivars were significantly decreased (except for Argy 25, and 125 mM NaCl concentrations). The major decrease in protein content was detected in Hazera (31%) at 125 mM salt

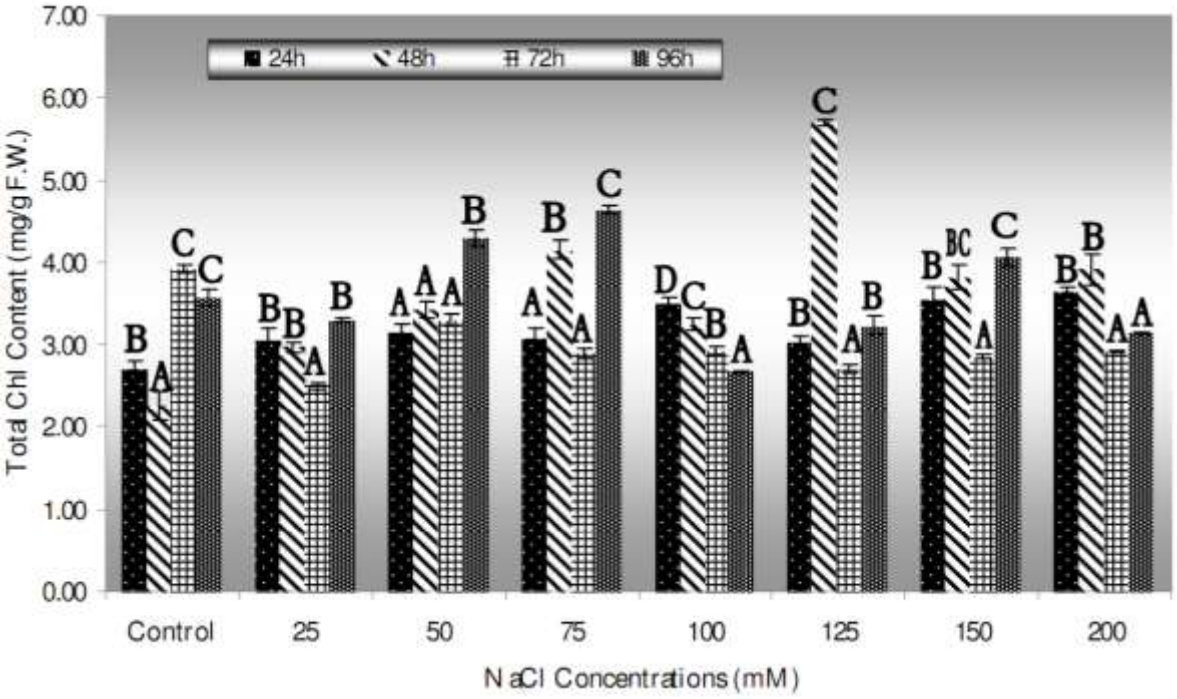

Figure 3. Effect of NaCl concentrations on total Chl content of *L. esculentum* Mill. cv. Argy.

application and this reduction was followed by Argy (19%) at 100 mM salt application (Table 2). In Dalli Tokat, maximum decrease was detected as 18 % at 200 mM NaCl concentration compared to control. The total soluble protein contents of Hazera and Argy cultivars did not show any statistical significant differences 72 h after salt treatment (Table 3), while 96 h after treatment significant reductions in protein contents were detected in Argy plants (Table 4). Decreases in protein content were high in Argy at all NaCl concentrations. Compared to control, maximum reduction in protein content was between 16 and 27 % at 96 h after salt treatment.

One of the mechanisms affected by salt stress in plants was protein synthesis. It is known that soluble protein content is an important indicator of physiological status of plants. At 24 h after salt treatment decrease, total soluble protein content was more evident in Hazera (Table 2). This situation demonstrated that Hazera plants were first affected by salt stress. So we thought that Dalli Tokat and Argy plants could tolerate 24 h salt application. Similar with our findings, Sibole et al. (1998) and Yurekli et al. (2004) reported that short term NaCl stress severely reduced leaf protein contents in *Phaseolus vulgaris* plants. 48 h after salt treatment, decrease in total soluble protein content was most visible in Hazera as with previous time. On the other hand, it was seen that 48 h salt application affected Dalli Tokat and Argy plants too. 72 h after salt treatment, we determined that Hazera plants were relatively acclimated to NaCl stress when compared with 24 and 48 h NaCl exposure. This was due to reduction of their total soluble protein contents, which were found to be lower than short time exposure. But In Dalli Tokat plants, this reduction was especially determined as 7 - 45 % compared with control. Compared to control, protein content of Argy plants exhibited statistically significant increase at 150 mM NaCl and decrease at 100 mM NaCl concentrations. Mohammadkhani and Heidari (2008) detected that drought stress caused initial increase due to expression of new stress proteins and subsequent decrease due to severe decrease in photosynthesis in two different maize cultivars. We thought that fluctuating trend in total soluble protein content can be sourced from synthesis of specific stress proteins. Yurekli et al. (2004) reported that total soluble protein content significantly decreased in salt sensitive *Phaseolus vulgaris* but increased in salt tolerant *P. acutifolius* plants. Similarly, Porgali and Yurekli (2005) reported that compared with control plants, protein amount in salt sensitive *L. esculentum* plants decreased with the salt application. In salt tolerant *L. pennellii* plants, total protein amount was more than control plants. Demiral and Turkan (2006) detected that while total soluble protein content of salt tolerant *O. sativa* cv. Pokkali plants increased with salinity, sensitive (*O. sativa* cv. IR-28) rice cultivars showed a decrease under salt stress. Similar results were reported in salt tolerant cultivars of barley, sunflower, finger millet and rice plants (Parvaiz and Satyavati, 2008). However, Ashraf and Fatima (1995) reported that there were not significant differences in leaf soluble protein contents in salt

sensitive and salt tolerant *Carthamus tinctorius*. Similarly, Qasim et al. (2004) reported that salt stress did not affect total soluble protein content of salt tolerant and sensitive canola cultivars. These different results on salt stress showed that the responses to salt stress depends on plant species even in varieties of same plant species, plant developmental stage, duration and severity of the salt application (Parvaiz and Satyavati, 2008).

In this study, when total soluble protein and pigment content were evaluated together we thought that *L. esculentum* cv. Hazera was relatively sensitive to short term salt stress and this cultivar may acclimate rapidly to salty environments. Dalli Tokat plants were not affected by short term salt stress and we can evaluate this cultivar as intermediate (Ashraf and O'leary, 1999). Ashraf and O'leary (1999) reported that the highest increase in protein content was found in salt sensitive wheat cv. Potohar while the lowest was in salt tolerant S24. Other examined wheat cultivars were defined as intermediate by these researchers. The results of this study suggest that the Argy cultivars are relatively better protected under salt stress conditions than Dalli tokat and Hazera cultivars.

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