

Full Length Research Paper

# The examination of Na-Ca effect on some qualitative and quantitative characters in durum wheat plants

Ragbet Ezgi Duran\*, Yasemin Coskun and Cigdem Savaskan

Biology Department, Faculty of Arts and Sciences, Suleyman Demirel University, 32260 Isparta, Turkey.

Accepted 19 May, 2011

The effect of salt stress (NaCl) on shoot height (cm), root length (cm), dry and fresh weight (g), chlorophyll a and b, total chlorophyll ( $\text{mg}^{-1}$ ) and carotenoid amount was investigated in this study. In addition, the positive effects of  $\text{Ca}^{+2}$  (20 mM) were also investigated. *Triticum durum* Desf. Mirzabey, Kunduru-1149, and DH-6 and DH-8, derived from Kunduru-1149 using wide hybridization, were used as plant materials in this study. Arnon-Hoagland solution was used as food source for the plants and various NaCl concentrations (0, 50, 150 and 200 mM) and  $\text{Ca}^{+2}$  (as  $\text{CaCl}_2$ ) were added to Arnon-Hoagland solution. The research was completed at the end of the fifth week and the effect of NaCl on the root length of genotypes was found to be significant ( $P < 0.05$ ).  $\text{Ca}^{+2}$  caused an increase in root length in all NaCl applications, except for the 200 mM NaCl, whereas shoot height decreased with increasing salt concentration, except for the 50 mM NaCl. This character also increased with  $\text{Ca}^{+2}$  application significantly. Dry and fresh weight of the plant decreased with increasing salt concentration.  $\text{Ca}^{+2}$  ameliorated dry and fresh weight of the plant at 50 mM NaCl in all the genotypes and NaCl doses, except for the Kunduru-1149 and DH-6. The highest increase in total chlorophyll amount was found in 50 mM NaCl +  $\text{CaCl}_2$  in DH-8 (from 17.29 to 20.14  $\text{mg g}^{-1}$ ). However, the rate of increase in the amount of carotenoid by the addition of  $\text{Ca}^{+2}$  was also determined for each genotype.

**Key words:** *Triticum durum* Desf., salinity, calcium, chlorophyll, carotenoid.

## INTRODUCTION

A plant cell, which contains many essential elements in micro and macro amounts, does not need  $\text{Na}^+$  ions.  $\text{Na}^+$  and  $\text{Cl}^-$  ions taken by plant roots from saline soils show toxic effects by blocking enzyme activity in cells. These ions accumulate in plant tissues and affect the growth and synthesis of cell pigments (chlorophyll and carotenoids) negatively (Taban et al., 1999; Duran et al., 2010). Calcium affects plant development under salt stress positively, enhances membrane integration and provides selective control of ion uptake and transport (Lauchli, 1990).  $\text{Na}^+$  permeability of cell membrane can be decreased by  $\text{Ca}^{+2}$  and prevented by  $\text{Na}^+$  accumulation, which is already taken by the cell with passive transport (Hoffman et al., 1989).

Reid and Smith (2000) investigated the limit of calcium effect on salt toxicity and reported that application of 150

mM NaCl on wheat (*Triticum aestivum* L.) reduced plant growth significantly, but improved plants when Ca concentration was added to the nutrient medium, thereby increasing it up to 2.34 mM. However, they also reported that increasing Ca to 10 mM did not affect growth. Also, Cramer (1992) investigated the effect of 10 mM Ca on the kinetic value of leaf length under the 75 mM NaCl condition and showed that Ca was more responsive on Dekalb hybrid XL75 genotype than Pioneer hybrid 3906 genotype in decreasing leaf elongation.

Arshi et al. (2005) investigated the effect of NaCl and  $\text{CaCl}_2$  on growth parameters, ionic relations and proline level of senna (*Cassia angustifolia*) plant in a pot culture experiment, using NaCl,  $\text{CaCl}_2$  and the combined salt of NaCl +  $\text{CaCl}_2$ . They showed that the combined treatments of NaCl +  $\text{CaCl}_2$  applied at different stages reduced the biomass, but this reduction was less than the one observed with NaCl treatments alone. However, proline accumulation in the leaves was 8 times higher than that in the controls with a treatment of 160 mM NaCl + 10 mM  $\text{CaCl}_2$ , whereas it was 5 times higher with NaCl

\*Corresponding author. E-mail: rezgiduran@gmail.com. Tel: 0090 246 211 41 52. Fax: 0090 246 237 11 06.

alone. Other scientists reported similar results on the ameliorative effect of calcium (Kaya et al., 2007; Manivannan et al., 2007; Nedjimi and Daoud, 2009).

Salinity and alkalinity are the important problems of Turkey's land, while salt accumulation in irrigated soils is one of the main factors that diminish crop productivity. In recent years, drainage problem has occurred due to increasing irrigation and the quality of irrigation water problems. Wheat is an important crop plant in Turkey. Therefore, to have information about salt tolerance of wheat genotypes is important in order to select genotypes for areas exposed to salt stress.

The effect of two different concentrations (100 and 200 mM) of sodium chloride (NaCl) on some quantitative and qualitative features in durum wheat genotypes (Mirzabey, Altin, Kunduru-1149, DH-6 and DH-8) was investigated in an earlier study by Duran et al. (2010). In this study, the plants belonging to durum wheat genotypes (*T. durum* Desf. Mirzabey, Kunduru-1149, DH-6 and DH-8) were grown under salt stress conditions (50, 150 and 200 mM NaCl), supplementing it with  $\text{Ca}^{2+}$  to determine genotype responses to Na-Ca relationship, following shoot height, root length, dry-fresh weight and chlorophyll-carotenoid amount.

## MATERIALS AND METHODS

This study was carried out with four *T. durum* Desf. cvs., namely: Mirzabey, Kunduru-1149, DH-6 and DH-8. Mirzabey is a modern semi-dwarf genotype, while Kunduru-1149 is an old landrace, improved by selection. Seeds of these genotypes were obtained from the Field Crops Central Research Institute, Ministry of Agriculture and Rural Affairs in Ankara. DH-6 and DH-8 (doubled haploid = DH) were derived from Kunduru-1149 via wide-cross hybridization (Savaskan, 1997). They are different genotypes and their differences are determined using the method of molecular genetic analysis by Hakki et al. (2001).

### The growth of donor plants

Seeds were sown in pots containing peat and then plants were kept in a plant growth chamber, belonging to the laboratory of Plant Tissue Culture and Biodosimetry, Department of Biology, Süleyman Demirel University, for 35 days, at 51 to 54% humidity and 18°C in 16/8 h photoperiod (12 klux). Arnon-Hoagland nutrient solution (NS) was used for the plants. Also, the treatment groups of NaCl (0, 50, 150 and 200 mM) and 20 mM  $\text{CaCl}_2$  were supplied to plants according to the application doses. The control group was supplied with only nutrient solution, while the application group was supplied with nutrient solution plus salt and  $\text{Ca}^{2+}$  as follows: 1) Control (0 mM NaCl + NS); 2) 50 mM NaCl + NS; 3) 50 mM NaCl + NS +  $\text{CaCl}_2$  (20 mM); 4) 150 mM NaCl + NS; 5) 150 mM NaCl + NS +  $\text{CaCl}_2$  (20 mM); 6) 200 mM NaCl + NS; 7) 200 mM NaCl + NS +  $\text{CaCl}_2$  (20 mM).

The doses of NaCl and  $\text{CaCl}_2$  were applied with NS after 10 days of sowing. The control group was supplied with only nutrient solution without any Na and  $\text{Ca}^{2+}$  treatments. Arnon-Hoagland solution was diluted three times before it was applied each time to the plants. Irrigation was performed in regular periods of treatments. After 35 days of sowing, 10 plants were selected from each of the application groups and the root length and shoot height were

measured, after which the dry and fresh weight was recorded. Also, the chlorophyll and carotenoid amounts were analysed.

### Records of dry and fresh weight

Plants were harvested after seven weeks for fresh weight measurement and then kept in the dark for 24 h at 70°C for dry weight measurement (Al-Mefleh and Tadros, 2010).

### Measurements of root length and shoot height

Root length and shoot height of each plant for the four genotypes were measured with a ruler.

### Determination of photosynthetic pigments

Chlorophyll-a and b and carotenoids were measured by extracting fresh leaf tissue (0,1 g) with potassium carbonate ( $\text{KCO}_3$ ) in 15 ml 80% acetone. Subsequently, the extract was put in a centrifuge tube, where 5 ml acetone was added and centrifuged at 3000 rpm for 5 min. The absorbance of the supernatant was read at 663, 645 and 450 nm, using a spectrophotometer (Jenway 6300) (Strain and Svec, 1966; Duran et al., 2010). Chlorophylls a and b were calculated using the following formulae:

$$\text{mg chlorophyll a / g tissue} = [12.7 (D663) - 2.69 (D645)] (V / 1000 W)$$

$$\text{mg chlorophyll b / g tissue} = [22.9 (D645) - 4.68 (D663)] (V / 1000 W)$$

Where, D = absorbance, V = final volume of the 80% acetone and W = fresh weight of tissue extracted (0,1 g) (Witham et al., 1971).

### Statistical analysis

The experiment was established with a randomized complete block design, performed three times for each application group and SPSS 15.0 software programme was used for the statistical analysis. Means were compared with Duncan's multiple range test (Duncan's test).

## RESULTS

Durum wheat genotypes showed differences significantly in all the treatment groups in this research ( $P < 0.01$ ) (Table 1). Also, root length, shoot height, fresh weight and dry matter, carotenoids ( $P < 0.05$ ), total chlorophyll, and chlorophyll-a and -b amounts ( $P < 0.01$ ) of the genotypes were found to be significant statistically in all the NaCl treatment groups (Table 1). As it can be seen in Table 1, interaction between genotypes and salinity (NaCl) was found to be important, except for the amount of carotenoids (Table 1) ( $P < 0.01$ ). Also, the relationship of salinity and calcium (Ca) functions in genotypes was found to be statistically important, as already investigated in all characters in this research (Table 1).

The amounts of chlorophyll a and b decreased when the genotypes were treated with 50 mM NaCl ( $P < 0.05$ ) according to the control group (Figures 1 a and b). In

**Table 1.** The variation analysis of plant characters belonging to durum wheat genotypes affected by treatment subject in this study.

Subject	d.f	F Value						
		Root	Shoot	Dry matter	Total chlorophyll	Carotenoids	Chlorophyll-a	Chlorophyll-b
Genotype	4	115.103**	102.056**	104.512**	3046.098**	22.502**	1874.238**	594.742**
NaCl	2	96.076**	86.161**	138.957**	1235.780**	8.326*	756.300**	247.178**
Genotype x NaCl	8	5.072*	4.103*	18.964**	182.788**	2.675ns	104.932**	48.702**
Genotype x NaCl x Ca	12	18.065**	12.106**	22.076**	198.172**	4.646*	112.985**	65.825**

\*\*0.01 significant level; \*0.05, significant level; ns, not significant.

contrast, the value of these pigments increased again in all genotypes except in Kunduru-1149, when the genotypes were treated with 50 mM NaCl + CaCl<sub>2</sub>. The highest increasing amount of these genotypes in chlorophyll-a and -b was observed in DH-8, respectively (Figure 1c). In DH-8, chlorophyll-a was affected by salinity (11.16 mg g<sup>-1</sup>) (Figure 1b), while this amount was increased to 13.25 mg g<sup>-1</sup> again with Ca<sup>+2</sup> treatment (Figure 1c). Also, in DH-6, the amount of chlorophyll-b increased from 4.65 (Figure 1b) to 6.85 mg g<sup>-1</sup> (Figure 1c) with Ca<sup>+2</sup> treatment effects.

In the genotypes, the amount of chlorophyll-a and -b was found to be lower in the treatment group of 150 mM NaCl than in the control group (P<0.05) (Figure 2a and b), but the treatment of 150 mM NaCl increased the amount of chlorophyll-b in Kunduru-1149 (Figure 2a). Also, the treatment of 150 mM NaCl+Ca positively affected chlorophyll-a and -b amounts (P<0.01) in all the genotypes, except in Kunduru-1149 (Figure 2b). Ca<sup>+2</sup> gave the most effective results in increasing chlorophyll-a and -b amounts in Mirzabey and DH-8, respectively. However, chlorophyll-a changed from 10.75 to 12.15 mg g<sup>-1</sup> in Mirzabey and chlorophyll-b also changed from 5.95 to 6.60 mg g<sup>-1</sup> in DH-8 (Figures 2a and b).

The treatment of 200 mM NaCl decreased the amounts of chlorophyll-a and -b in all genotypes investigated in this study (P<0.05) (Figure 3a). In contrast, the treatment of 200 mM NaCl+ CaCl<sub>2</sub> ameliorated the amount of chlorophyll-a and -b importantly in all genotypes, except in Mirzabey statistically (P<0.01) (Figure 3b). In the 200 mM NaCl + CaCl<sub>2</sub> group, an increasing amount of chlorophyll-a and -b was observed in Kunduru-1149. In this genotype, increasing results were found in chlorophyll-a and -b from 9.04 to 11.59 mg g<sup>-1</sup> and from 4.24 to 6.05 mg g<sup>-1</sup>, respectively (Figures 3a and b).

Negative effect of salinity in the amount of total chlorophyll was observed in all genotypes and was found statistically (P<0.05) (Table 2). The highest total chlorophyll amount was found in DH-8 under salinity effect in the control group and in almost all the treatment groups (Table 2).

Total chlorophyll amount was affected by Ca<sup>2+</sup> in all treatments, except in 150 mM NaCl + CaCl<sub>2</sub> and 200 mM NaCl + CaCl<sub>2</sub> in Kunduru-1149 and Mirzabey, respectively (Table 2). Total chlorophyll was ameliorated by 150

mM NaCl + CaCl<sub>2</sub> treatment in DH-8 from 17.29 to 20.14 mg g<sup>-1</sup> and for 200 mM NaCl + CaCl<sub>2</sub> treatment in Kunduru-1149 from 14.52 to 17.60 mg g<sup>-1</sup> (Table 2).

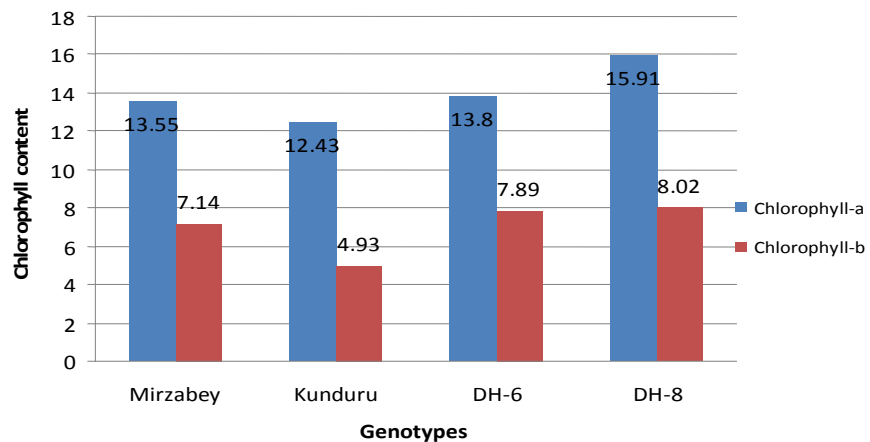
NaCl affected the amount of carotenoids in the genotypes negatively, but this situation was not found to be statistically important (Figure 4). It was observed that Ca<sup>+2</sup> reduced the negative effect of salinity stress on the amount of carotenoids in all the genotypes, except in DH-6 and Kunduru-1149 treated after 50 mM NaCl and also in Kunduru-1149 and Mirzabey after 150 mM NaCl treatments (Figure 4).

The changes of root length and shoot height after treating genotypes with NaCl and CaCl<sub>2</sub> are shown in Tables 3 and 4. Root length was found to be lower in all the genotypes after NaCl treatment than in the control group (P<0.05). Ca<sup>+2</sup> had positive effect on root length in all the NaCl treatment groups, except for the highest level of NaCl treatment group (Table 3) (Figure 5). It was also effective for shoot height in all the treatment groups significantly (P<0.01) in the genotypes (Table 4).

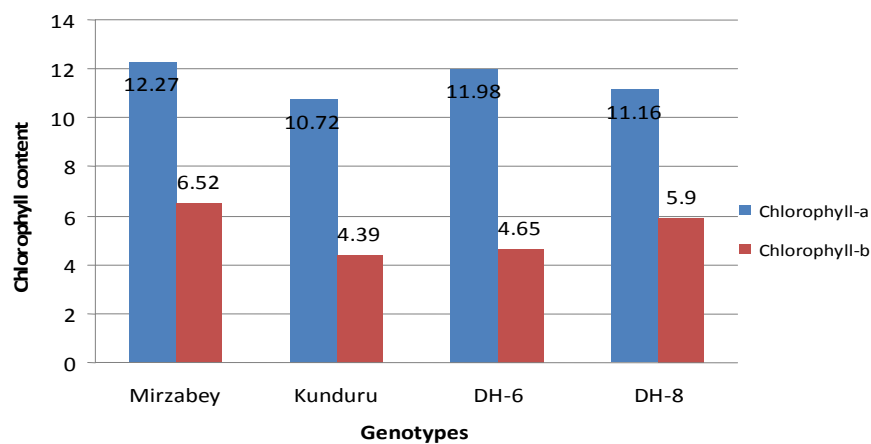
As it can be seen in Figures 6 and 7, fresh and dry weights of the plants reduced when the NaCl concentration increased in the treatments (P<0.05) in this study. The lowest value of fresh weight in plants was found in Mirzabey as 44.19%. Also, for the dry weight, the lowest amount of plant value was in Kunduru-1149 in the 200 mM NaCl treatment group (18.19%). Ca<sup>+2</sup> ameliorated the damage of NaCl treatments in all the genotypes, except in Kunduru-1149 and DH-6 in the 50 mM NaCl salinity. The highest fresh weight of plant was found as 0.699 g/plant in 50 mM NaCl + CaCl<sub>2</sub> and the highest dry weight of plant was found as 0.117 g/plant in 150 mM NaCl + CaCl<sub>2</sub> treatment group.

## DISCUSSION

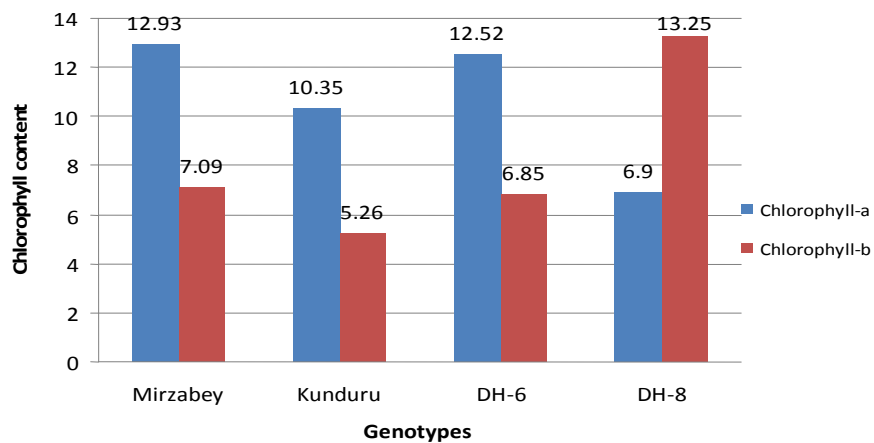
Soil salinity is a major factor limiting plant productivity and affecting about 95 million hectares world wide (Szabolcs, 1994). Saboora et al. (2006) warn that there is a dangerous trend of a 10% per year increase in saline areas throughout the world. There are areas, totaling 1,518,722 hectares in Turkey, which exist with salt and sodium (Rural Services General Management, 2006). Reclamation of this land is slow, difficult and expensive



a

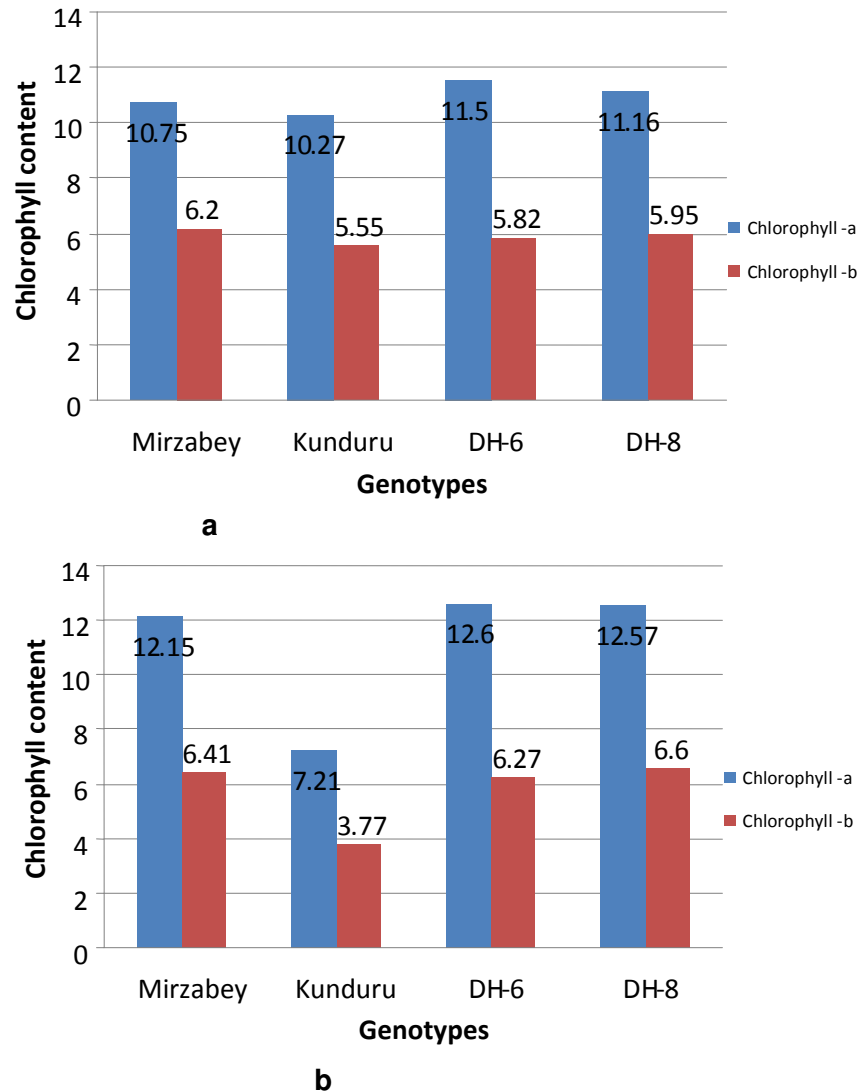


b



c

**Figure 1.** Chlorophyll-a and -b amount of genotypes for 50 mM NaCl and 50 mM NaCl + CaCl<sub>2</sub> application groups. (a) Control group; (b) 50 mM NaCl application group; (c) 50 mM NaCl+ CaCl<sub>2</sub> application group.

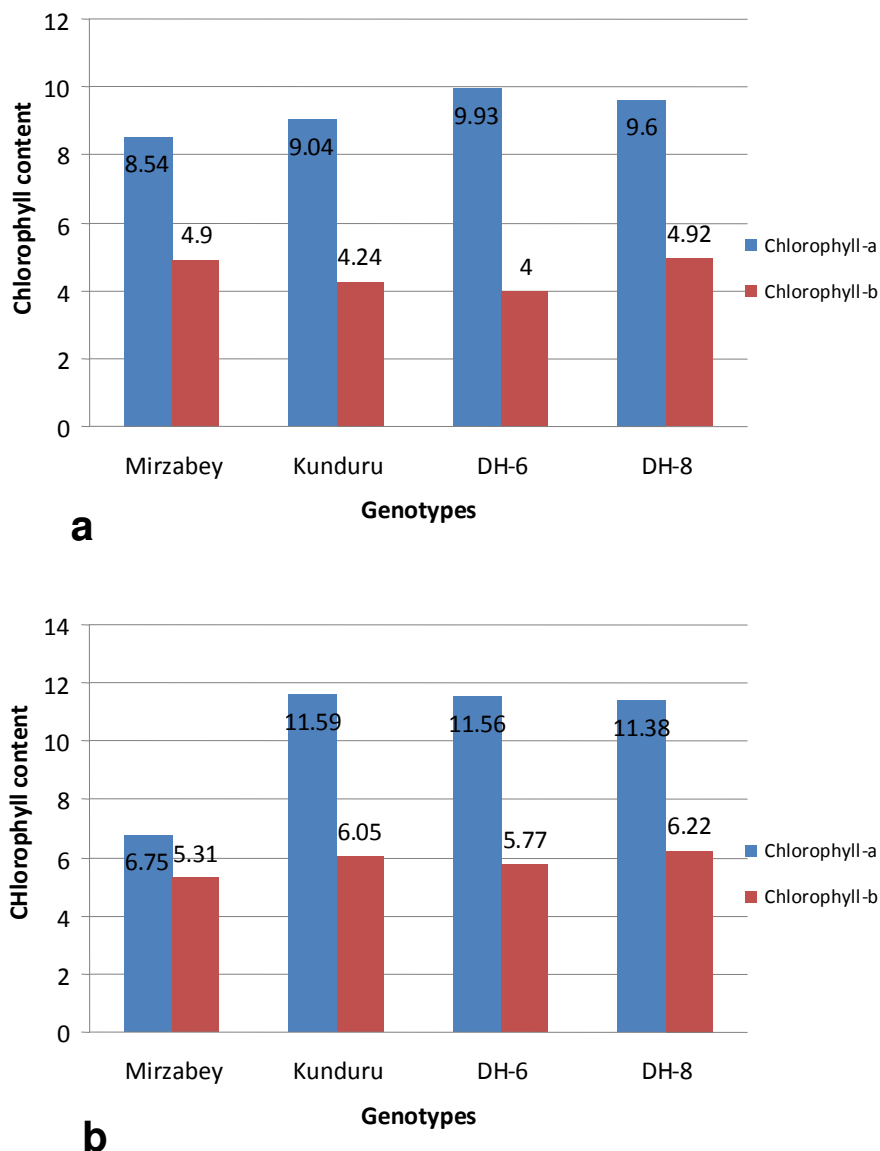


**Figure 2.** Chlorophyll-a and -b content of genotypes for 150 mM NaCl and 150 mM NaCl + CaCl<sub>2</sub> application groups. (a) 150 mM NaCl application group; (b) 150 mM NaCl+ CaCl<sub>2</sub> application group.

Therefore, it is important to produce salt-resistant plants. In Turkey, the durum wheat sowing area of 13.35 million hectares, producing 3.74 million tons and 213 kg per hectare yield, also has an important position (Anonymous, 2009). Turkey is one of the major producer countries of durum wheat. There is approximately 30% of durum wheat in the total wheat planting area. After the 1990s, this rate fell significantly and decreased to around 15% in 2007 (Anonymous, 2007). Reasons for this decrease are that salty areas increased rapidly and the production of high yielding bread wheats which have the ability of better adaptation. Therefore, determining salt-tolerant genotypes of durum wheat is very important.

Many researches have considered that Ca<sup>2+</sup> play a nursery role in plants for salt stress conditions, affecting ion transportation in cells and protecting the cell

membrane. Jaleel et al. (2007) stated that salinity could be an inhibition of root development by altering the external water potential and increasing ion toxicity and instability in plants. As it can be seen in Tables 3 and 4 of this study, 50, 150 and 200 mM doses of NaCl were importantly hazardous to the root development of durum wheat plantlets statistically. However, negative effects of NaCl were reduced by Ca addition and following this procedure, the length of root increased again in 50 and 150 mM doses of NaCl treatments significantly. Also, in 200 mM dose of NaCl treatment, only the roots of Kunduru-1149 were lengthened by addition of Ca. In a previous study conducted by Manivannan et al. (2007), they observed that root length and shoot height of mung bean (*Vigna radiata*) were much more longer in the combination of NaCl + CaCl<sub>2</sub> treatment than in NaCl



**Figure 3.** Chlorophyll-a and -b content of genotypes for 50 mM NaCl and 50 mM NaCl + CaCl<sub>2</sub> application groups. (a) 200 mM NaCl application group; (b) 200 mM NaCl + CaCl<sub>2</sub> application group.

and/or CaCl<sub>2</sub> treatments. Reduction of shoot height in various plants using NaCl was reported by many researches (Meneguzzo and Navarilzzo, 1999; Meloni et al., 2004). In this study, the treatment group of 50 mM NaCl increased shoot height slightly when compared with the control group.

Sotiropoulos and Dimassi (2004) reported that it might be possible to increase shoot height in plants with low level of NaCl treatment. Also, Flowers and Lauchlii (1983) supported the idea mentioning that positive effect of low level dose of NaCl in shoot height could be reasoned from osmolarity. In this study, plant shoot height was reduced in the treatment groups of 150 and 200 mM doses, while it increased importantly after using Ca in

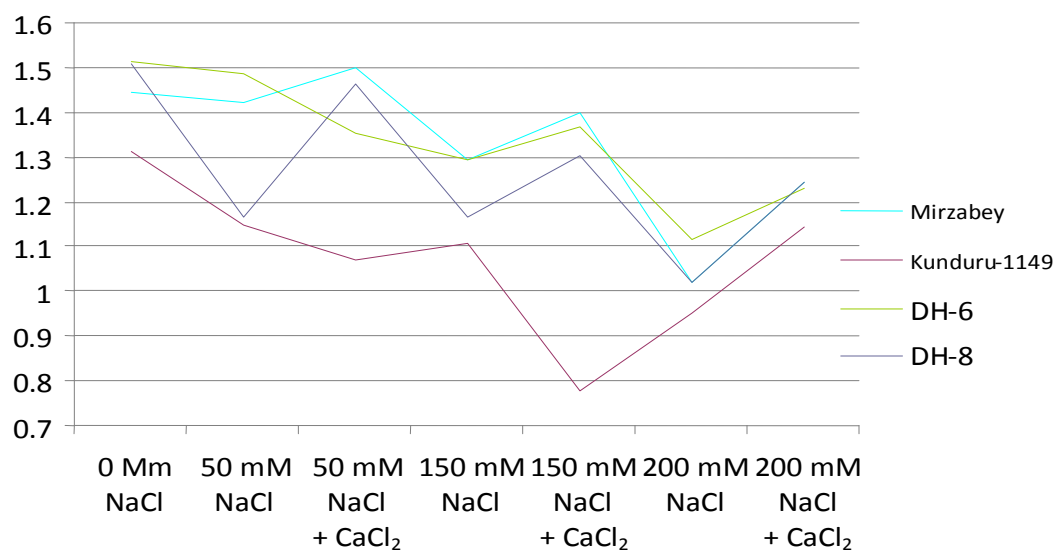
durum wheat genotypes (Table 4). This increase had also observed by Khavari-Nejad and Chaparzadeh (1998) in their previous study about *Medicago sativa*, by Manivannan et al. (2007) for *V. radiata* and Jaleel et al. (2008) for *Phyllanthus amarus*.

Researches about salinity in crop plants were commonly focused on the photosynthetic system in plants. Arzani (2008) found that the net photosynthesis rate and growth of plant reduced because of salinity. Misra et al. (1997) and Munns et al. (2002) also found that various parameters about photosynthesis, such as the potential of leaf moisture, the composition of pigment, the heat of leaf and the relative water content of leaf, were changed by salinity.

**Table 2.** Effects of NaCl and Ca<sup>2+</sup> applications on total chlorophyll amount in leaf tissues (g) taken from 35-days durum wheat plants.

Genotype	Total chlorophyll content						
	0 mM NaCl (control)	50 mM NaCl	50 mM NaCl + CaCl <sub>2</sub>	150 mM NaCl	150 mM NaCl + CaCl <sub>2</sub>	200 mM NaCl	200 mM NaCl + CaCl <sub>2</sub>
Mirzabey	20.69 <sup>bc</sup> ±0.40	18.78 <sup>d-g</sup> ±0.35	20.01 <sup>bcd</sup> ±0.17	16.94 <sup>ghi</sup> ±0.34	18.55 <sup>d-g</sup> ±0.36	13.44 <sup>mno</sup> ±0.35	12.05 <sup>op</sup> ±0.35
Kunduru-1149	17.36 <sup>f-i</sup> ±0.47	15.10 <sup>j-m</sup> ±0.70	15.60 <sup>f-i</sup> ±0.50	15.81 <sup>h-k</sup> ±0.31	10.98 <sup>p</sup> ±0.22	13.28 <sup>no</sup> ±0.45	17.63 <sup>e-h</sup> ±0.62
DH-6	21.69 <sup>b</sup> ±1.00	16.62 <sup>h-j</sup> ±0.82	19.37 <sup>cde</sup> ±0.28	17.33 <sup>f-i</sup> ±0.77	18.86 <sup>c-g</sup> ±0.17	13.93 <sup>lmn</sup> ±0.49	17.32 <sup>f-i</sup> ±0.36
DH-8	24.93 <sup>a</sup> ±0.82	17.29 <sup>f-i</sup> ±0.44	20.14 <sup>bcd</sup> ±0.24	17.11 <sup>ghi</sup> ±0.18	19.16 <sup>c-f</sup> ±0.13	14.52 <sup>k-n</sup> ±0.13	17.60 <sup>e-h</sup> ±0.17

\*The values that are followed by the same letter do not differ statistically at a significance level of 5% ( $P < 0.05$ ).

**Figure 4.** Effects of NaCl and Ca<sup>2+</sup> applications on carotenoids amount in leaf tissues (g) taken from 35-days durum wheat plants.

The amount of photosynthetic pigment (chloroplast) is affected by the many physiological and environmental factors. The amount of chlorophyll is one of the very important parameters for determining salt stress in plant development. Chlorophyll amount has been reduced in high concentration of salinity treatments according to

the control groups in muskmelon (Franco et al., 1993), bean (Gadallah, 1999) and in tomatoes (Toffouo et al., 2010). In this study, total chlorophyll amount decreased significantly in all the durum wheat genotypes and NaCl applications under salt stress (Table 1).

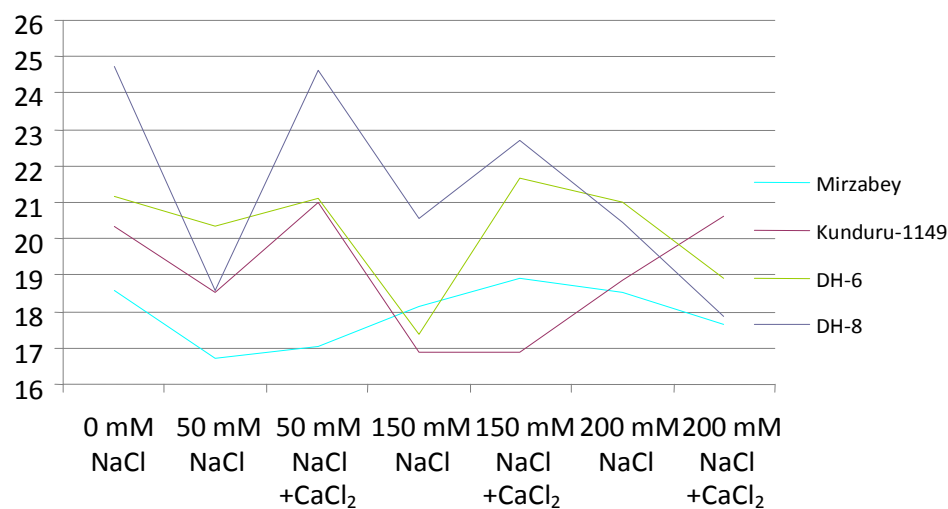
The disruption of metabolic activities under salt

stress, affects the chlorophyll activation negatively, because it reduces the uptake of macronutrient elements, such as Ca, K, N, P and Mg. Both parameters that are related with the total chlorophyll and carotenoids were reduced in the NaCl treatment groups significantly; however, Ca<sup>2+</sup> alleviated the negative effects of NaCl on the

**Table 3.** Effects of NaCl and Ca applications on root length in 35-days plants of durum wheat genotypes.

Genotype	Root length (cm)						
	0 mM NaCl (control)	50 mM NaCl	50 mM NaCl + CaCl <sub>2</sub>	150 mM NaCl	150 mM NaCl + CaCl <sub>2</sub>	200 mM NaCl	200 mM NaCl + CaCl <sub>2</sub>
Mirzabey	18.60 <sup>efg</sup> ±0.45	16.70 <sup>g</sup> ±0.52	17.03 <sup>fg</sup> ±0.25	18.13 <sup>f</sup> ±0.26	18.93 <sup>def</sup> ±0.40	18.53 <sup>efg</sup> ±0.22	17.66 <sup>fg</sup> ±1.02
Kunduru-1149	20.33 <sup>cde</sup> ±0.48	18.53 <sup>efg</sup> ±0.34	20.98 <sup>bc</sup> ±0.32	16.86 <sup>g</sup> ±0.36	16.90 <sup>g</sup> ±0.32	18.86 <sup>def</sup> ±0.30	20.60 <sup>cd</sup> ±0.46
DH-6	21.16 <sup>bc</sup> ±0.35	20.36 <sup>cde</sup> ±0.38	21.10 <sup>bc</sup> ±0.44	17.40 <sup>fg</sup> ±0.22	21.66 <sup>bc</sup> ±0.25	21.00 <sup>bc</sup> ±0.36	18.93 <sup>def</sup> ±0.26
DH-8	24.76 <sup>a</sup> ±0.35	18.56 <sup>efg</sup> ±0.28	24.65 <sup>a</sup> ±0.26	20.56 <sup>cd</sup> ±0.32	22.68 <sup>b</sup> ±0.24	20.45 <sup>cde</sup> ±0.28	17.86 <sup>fg</sup> ±0.26

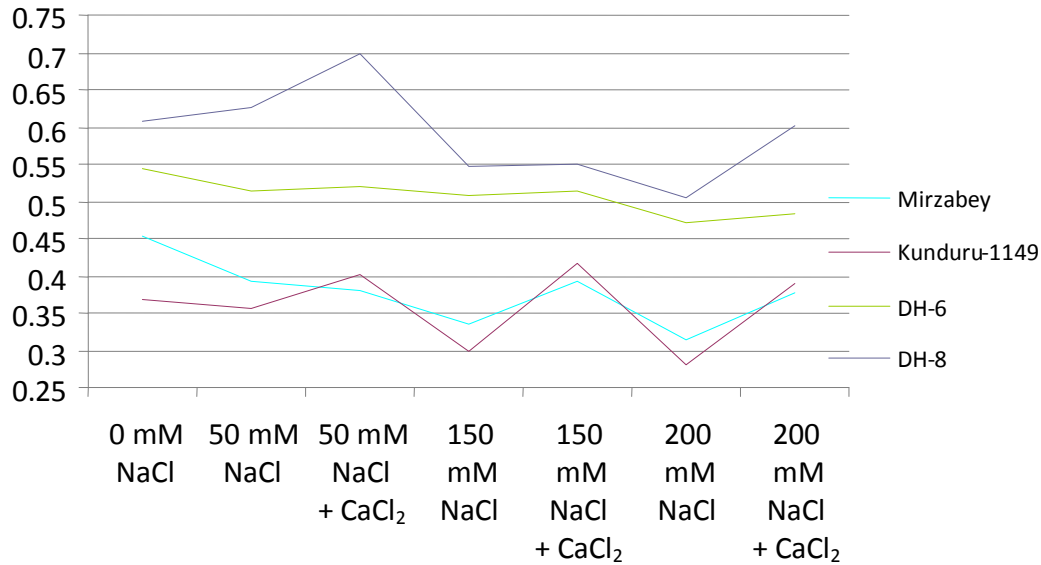
\*The values that are followed by the same letter do not differ statistically at a significance level of 5% (P<0.05).

**Figure 5.** Effects of NaCl and CaCl<sub>2</sub> applications on root length (cm) in 35-days plants.**Table 4.** Effects of NaCl and Ca applications on shoot height in 35-days plants of durum wheat genotypes.

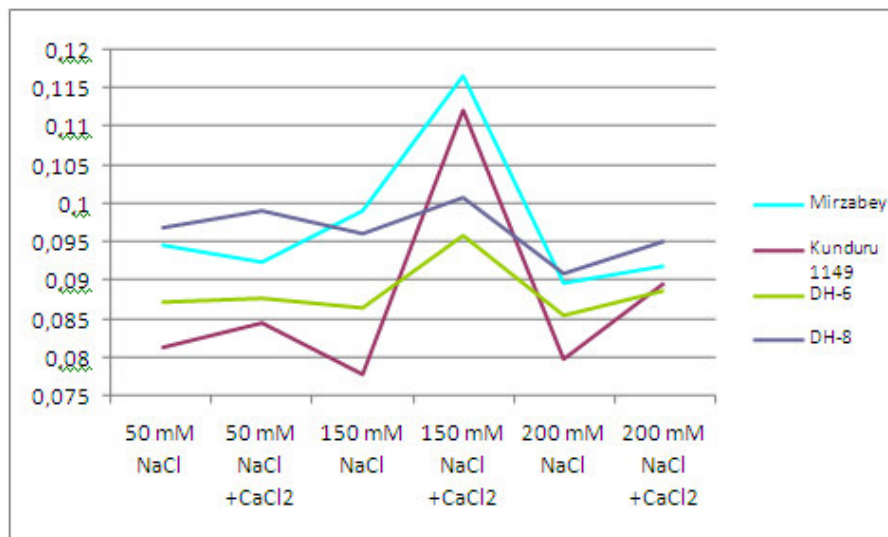
Genotype	Shoot height (cm)						
	0 mM NaCl (control)	50 mM NaCl	50 mM NaCl + CaCl <sub>2</sub>	150 mM NaCl	150 mM NaCl + CaCl <sub>2</sub>	200 mM NaCl	200 mM NaCl + CaCl <sub>2</sub>
Mirzabey	29.33 <sup>gh</sup> ±0.30	29.90 <sup>g</sup> ±0.32	30.03 <sup>g</sup> ±0.34	27.36 <sup>hi</sup> ±0.40	29.93 <sup>g</sup> ±0.42	26.76 <sup>i</sup> ±0.28	29.53 <sup>gh</sup> ±0.32
Kunduru-1149	30.83 <sup>fg</sup> ±0.25	32.76 <sup>ef</sup> ±0.26	37.20 <sup>cd</sup> ±0.34	30.10 <sup>g</sup> ±0.28	35.96 <sup>de</sup> ±0.35	30.76 <sup>fg</sup> ±0.28	34.40 <sup>ef</sup> ±0.30
DH-6	31.16 <sup>fg</sup> ±0.26	31.13 <sup>fg</sup> ±0.24	38.76 <sup>abc</sup> ±0.24	25.56 <sup>i</sup> ±0.50	40.23 <sup>a</sup> ±0.36	30.20 <sup>g</sup> ±0.26	39.56 <sup>a</sup> ±0.24
DH-8	33.23 <sup>ef</sup> ±0.26	36.60 <sup>cde</sup> ±0.40	37.60 <sup>bcd</sup> ±0.26	31.80 <sup>fg</sup> ±0.30	35.64 <sup>de</sup> ±0.34	26.30 <sup>i</sup> ±0.32	31.56 <sup>fg</sup> ±0.32

\*The values that are followed by the same letter do not differ statistically between them in a significance level of 5% (P<0.05).





**Figure 6.** Effects of NaCl and CaCl<sub>2</sub> applications on fresh weight in 35-days plants of durum wheat genotypes.



**Figure 7.** Effects of NaCl and CaCl<sub>2</sub> applications on dry weight in 35-days plants of durum wheat genotypes

amount of total chlorophyll and carotenoids, besides chlorophyll-a and -b in the genotypes considered in this study. The amount of chlorophyll-a increased by 18.72% in DH-8 and also chlorophyll-b increased by 47.31% in DH-6 with CaCl<sub>2</sub> application under 50 mM NaCl salinity (Figure 1). The amount of chlorophyll-a increased maximally by 13.02% in Mirzabey and chlorophyll-b also increased maximally by 10.92% in DH-8 with Ca<sup>2+</sup> application under the 150 mM NaCl salinity (Figure 2). Ca<sup>2+</sup> was ameliorated with the highest degree of chlorophyll-a and -b amounts under 200 mM NaCl salinity in

Kunduru-1149 genotype, by 28.20 and 42.68%, respectively (Figure 3). The highest amelioration with the amount of total chlorophyll of Ca<sup>2+</sup> was found in DH-6 as 16.48% (using 50 mM NaCl + CaCl<sub>2</sub>), while in DH-8, it was found as 11.98% (using 150 mM NaCl + CaCl<sub>2</sub>) and in Kunduru-1149, it was found as 32.75% (using 200 mM NaCl + CaCl<sub>2</sub>) (Table 2). Various nursery effects were found in genotypes of Ca<sup>2+</sup>, but the highest rate of nursery was observed in DH-8. Kaya and Higgs (2003) stated that KNO<sub>3</sub> supplied the amounts of K, as well as the chlorophyll content, in leaves and roots and alleviated the

stress parameters in bell pepper.

Ebert et al. (2002) reported that the chlorophyll content of guava leaf (*Psidium guajava* L.) was lower in 60 mM NaCl than in 30 mM NaCl treatment, while the chlorophyll concentration and photosynthesis rate increased when they added Ca(NO<sub>3</sub>)<sub>2</sub> to the treatment. This increase in plants was 143% under 30 mM NaCl stress and 10 mM Ca(NO<sub>3</sub>)<sub>2</sub> treatment group. Whittington and Smith, (1992) stated that external application of high doses of calcium, reduces the permeability of cell membranes against Na<sup>+</sup> and so prevents the accumulation of sodium by passive transport in the cell and plant. Yeo and Flowers (1983) stated that application of potassium increased the chlorophyll pigment in the young leaves of plants under saline conditions. As such, we thought that calcium showed the same effect in this study.

In photosynthetic organisms, carotenoids play a vital role in the photosynthetic reaction centre. They either participate in the energy-transfer process, or protect the reaction center from auto-oxidation. However, salinity caused a decrease in the carotenoids content of plants. In this study, the maximum amelioration of the carotenoid content with Ca<sup>2+</sup> was found in all NaCl application groups in DH-8.

Many researchers reported that salinity significantly reduced dry and fresh matter accumulation in various plants (Irshad et al., 2002; Ghoulam et al., 2002; Daşgan et al., 2002). Reduction in dry weight reflects the increased metabolic energy cost and reduced carbon gain. It also reflects salt impacts on tissues (Karimi et al., 2005) reduction in photosynthetic rates (Ziska et al., 1990; Ashraf, 2004) and attainment of maximum salt concentration obtained by the fully expanded leaves (Hu et al., 2000). Ca<sup>2+</sup> application partially resulted in the amelioration of adverse effects of high NaCl in sorghum growth, despite the fact that supplemental effect of Ca<sup>2+</sup> increased osmolality in a study on salinity-calcium interactions on corn (Cramer, 1992). Similar results were obtained in this study and Ca ameliorated the negative effect of NaCl on the dry and fresh weight of the plant. The results showed that there was clear effect of salinity concentration on the plant growth and pigment contents of durum wheat genotypes. It was observed that the high levels of salinization induced a significant decrease in the contents of pigment fractions (chlorophyll a and b) and consequently, on the total chlorophyll content, dry and fresh weight, shoot height and root length as compared with the control plants. However, Ca<sup>2+</sup> alleviated the negative effects of NaCl on these parameters considered in this study. Although this improvement was seen at varying rates for each genotype, the most tolerant and most positive effect of Ca<sup>2+</sup> was thought to occur in DH-8.

## REFERENCES

Al-Mefleh NK, Tadros MJ (2010). Influence of water quantity on the yield, water use efficiency, and plant water relations of *Leucaena*

- leucocephala* in arid and semi arid environment using drip irrigation system. Afr. J. Agric. Res. 5(15): 1917-1924.
- Anonymous (2007). Turkish Statistical Institute. Crop production statistics database. <http://www.tuik.gov.tr/bitkiselapp/bitkisel.zul>
- Anonymous (2009). Turkish Statistical Institute. Crop production statistics database. <http://www.tuik.gov.tr/bitkiselapp/bitkisel.zul>
- Arshi A, Abdin MZ, Iqbal M (2005). Ameliorative effects of CaCl<sub>2</sub> on growth, ionic relations, and proline content of senna under salinity stress. J. Plant. Nutr. 28(1): 101-125.
- Arzani A (2008). Improving salinity tolerance in crop plants: a biotechnological view. *In vitro* Cell. Dev. Biol. Plant, 44: 373-383.
- Ashraf M (2004). Some important physiological selection criteria for salt tolerance in plants. *Flora*, 199: 361-376.
- Cramer GR (1992). Kinetics of maize leaf elongation. II. Response of a Na-excluding cultivar and a Na-including cultivar to varying Na/Ca salinities. J. Exp. Bot. 43: 857-864.
- Daşgan HY, Aktas H, Abak K, Cakmak I (2002). Determination of screening techniques to salinity tolerance in tomatoes and investigation of genotype responses. *Plant Sci.* 163: 695-703.
- Duran RE, Coşkun Y, Savaşkan Ç (2010). The effect of salt on some quantitative and qualitative features in durum wheat genotypes (*Triticum durum* Desf.). *SDU J. Natural Appl. Sci.* 14(1): 17-22.
- Ebert G, Eberle J, Ali-Dinar H, Lüdders P (2002). Ameliorating effects of Ca(NO<sub>3</sub>)<sub>2</sub> on growth, mineral uptake and photosynthesis of NaCl-stressed guava seedlings (*Psidium guajava* L.). *Scientia Horticulturae*, 93: 125-135.
- Flowers TJ, Lauchli A (1983). Inorganic plant nutrition. V3. Sodium versus potassium: Substitution and compartmentation. *Encycl. Plant Physiol.* 15B: 651-681.
- Franco JA, Esteban C, Rodriguez C (1993). Effect of salinity on various growth stages of muskmelon cv. Revival. *J. Hort. Sci.* 68: 899-904.
- Gadallah MAA (1999). Effects of proline and glycinebetaine on *Vicia faba* responses to salt stress. *Biol. Plant.* 42: 249-257.
- Ghoulam C, Foursy A, Fores K (2002). Effects of salt stress on growth inorganic ions and proline accumulation in relation to osmotic adjustment in five sugar beet cultivars. *Environ. Exp. Bot.* 47: 39-50.
- Hakki E, Savaskan C, Akkaya MS (2001). Genotyping of anatolian doubled-haploid durum lines with SSR markers. *Euphytica*, 122: 257-262.
- Hoffman R, Tufariello J, Bisson MA (1989). Effect of divalent cations on Na<sup>+</sup> permeability of *Chara corallina* and freshwater grown *Chara buckelii*. *J. Exp. Bot.* 40: 875-881.
- Hu Y, Schnyder H, Schmidhalter U (2000). Carbohydrate deposition and partitioning in elongation leaves of wheat under saline soil conditions. *Aust. J. Plant Physiol.* 27: 363-370.
- Irshad M, Yamamoto S, Eneji AE, Endo T, Hona T (2002). Urea and manure effect on growth and mineral contents of maize under saline conditions. *J. Plant Nutr.* 25(1): 189-200.
- Jaleel CA, Kishorekumar A, Manivannan P, Sankar B, Gomathinayagam M, Rajaram P (2008). Salt stress mitigation by calcium chloride in *Phyllanthus amarus*. *Acta Bot. Croat.* 67(1): 53-62.
- Jaleel CA, Gopi R, Manivannan P, Panneerselvam R (2007). Antioxidative potentials as a protective mechanism in *Catharanthus roseus* (L.) G. Don. plants under salinity stress. *Tur. J. Bot.* 31: 245-251.
- Karimi J, Ghorbanli M, Heidari H, Khavari-Nejad RA, Assareh MH (2005). The effects of NaCl on growth, water relations, osmolytes and ion content in *Kochia prostrata*. *Biol. Planta*, 49: 301-304.
- Kaya C, Higgs D (2003). Supplementary potassium nitrate improves salt tolerance in bell pepper plants. *J. Plant Nutr.* 26(7): 1367-1382.
- Kaya C, Tuna AL, Ashraf M, Altunlu H (2007). Improved salt tolerance of melon (*Cucumis melo* L.) by the addition of proline and potassium nitrate. *Environ. Exp. Bot.* 60: 397-403.
- Khavari-Nejad RA, Chaparzadeh N (1998). The effects of NaCl and CaCl<sub>2</sub> on photosynthesis and growth of alfalfa plants. *Photosynthetica*, 35: 461-466.
- Lauchli A (1990). Calcium, salinity and plasma membrane. In: *Calcium in Plant Growth and Development* (Leonard RJ and Hepler PK Eds.). The American Society of Plant Physiologists Rockville, M.D. 26: p. 35.
- Manivannan P, Jaleel CA, Sankar B, Somasundaram R, Murali PV,

- Sridharan R, Panneerselvam R (2007). Salt stress mitigation by calcium chloride in *Vigna radiata* (L.) WILCZEK. *Acta Biologica Cracoviensia Series Botanica*, 49(2): 105–109.
- Meloni DA, Gulotta MR, Martínez CA, Oliva MA (2004). The effects of salt stress on growth, nitrate reduction and proline and glycinebetaine accumulation in *Prosopis alba*. *Braz. J. Plant Physiol.* 16(1): 39-46.
- Meneguzzo S, Navarilzzo I (1999). Antioxidative responses of shoots and roots of wheat to increasing NaCl concentrations. *J. Plant Physiol.* 155: 274–280.
- Misra AN, Sahu SM, Misra M, Singh P, Meera I, Das N, Kar M, Shau P (1997). Sodium chloride induced changes in leaf growth, and pigment and protein contents in two rice cultivars. *Biol. Plant.* 39: 257–262.
- Munns R (2002). Comparative physiology of salt and water stress. *Plant Cell Environ.* 25: 239-250.
- Nedjimi B, Daoud Y (2009). Ameliorative effect of CaCl<sub>2</sub> on growth, membrane permeability and nutrient uptake in *Atriplex halimus* subsp. *schweinfurthii* grown at high (NaCl) salinity. *Desalination*, 249: 163-166.
- Reid RJ, Smith FA (2000). The limits of sodium/calcium interactions in plant growth. *Aust. J. Plant Physiol.* 27: 709-715.
- Rural services General Management (2006). General envanter abstract. Summary of general inventory (01.01.2006). [http://www.khgm.gov.tr/ENVANTER/en\\_genel.htm](http://www.khgm.gov.tr/ENVANTER/en_genel.htm)
- Saboora A, Kiarostami K, Behroozbayati F, Hashemi SH (2006). Salinity (NaCl) tolerance of wheat genotypes at germination and early seedling growth, Pakistan. *J. Biol. Sci.* 9(11): 2009-2021.
- Savaskan C, Ellerbrook C, Fish LJ, Snape JW (1997). Doubled haploid production in Turkish durum wheats using crosses with maize. *Plant Breed.* 116: 299-301.
- Sotiropoulos T, Dimassi K (2004). Response to increasing rates of boron and NaCl on shoot proliferation and chemical composition of *in vitro* kiwifruit shoot tip cultures. *Plant Cell Tissue Organ Cult.* 79: 285-289.
- Strain SS, Svec WA (1966). Extraction, separation, estimation and isolation of chlorophylls. In the *Chlorophylls*, Vernon L.P.: Seely G.R. Academic Press N.Y. pp. 21-66.
- Szabolcs I (1994). Soils and salinization. In: Pessarakli M (Ed), *handbook of plant and crop stress*. Marcel Dekker, New York. pp. 3-11.
- Taban S, Güneş A, Alpaslan M, Özcan H (1999). Sensitivity of Various Maize (*Zea mays* L. cvs.) Varieties to Salinity. *Trend J. Agric. For.* 3: 625-633.
- Toffouo VD, Nouck AH, Dibong SD, Amougou A (2010). Effects of salinity stress on seedlings growth, mineral nutrients and total chlorophyll of some tomato (*Lycopersicon esculentum* L.) cultivars. *Afr. J. Biotechnol.* 9(33): 5366-5372.
- Whittington J, Smith FA (1992). Calcium-salinity interactions affect ion transport in *Chara corallina*. *Plant Cell Environ.* 15: 727-733
- Witham FH, Blaydes D, Devlin RM (1971). *Experiments in plant physiology*. Van Nostrand Reinhold, New York, pp. 1-11.
- Yeo RA, Flowers JT (1983). Vertical differences in toxicity of sodium ions in rice leaves. *Plant Physiol.* 59: 189-195.
- Ziska LH, Seemann JR, Dejong TM (1990). Salinity induced limitation on photosynthesis in *Prunus salicina*, a deciduous tree species. *Plant Physiol.* 93: 864- 870.