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EFFECTS OF SALINITY ON GERMINATION AND SOME GROWTH PARAMETERS IN THREE CULTIVARS OF *Zea mays* L.

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ABSTRACT

In this research, responses of three maize (*Zea mays* L.) cultivars (GS 308, DK 585 and P.3167) were compared for seed germination percentage, length of coleoptile and root, total fresh and dry weight of coleoptile and root, and absolute water content under the conditions of absence (control, distilled water) as well as presence (75-375 mM NaCl) of various of salinity. As a result of this study, germination and other growth parameters decreased significantly with increasing salinity level. Germination was decreased significantly at 75, 150 and 300 mM NaCl in GS 308, P.3167 and DK 585 cultivars, respectively. The root tissue was more affected by salinity than coleoptile tissue. Based on our findings, GS 308 maize cultivar may accept probably more sensitive than DK 585 and P.3167 cultivars.

Keywords: *Zea mays* L., NaCl stress, germination, growth parameters

Zea mays L.'IN ÜÇ ÇEŞİDİNDE ÇİMLENME VE BAZI BÜYÜME PARAMETRELERİ ÜZERİNE TUZLULUĞUN ETKİLERİ

ÖZET

Bu araştırmada, üç mısır (*Zea mays* L.) çeşidi (GS 308, DK 585 and P.3167), farklı tuz konsantrasyonlarının (75-375 mM NaCl) varlığında ve yokluğunda (kontrol, distile su) tohum çimlenme yüzdesi, koleoptil ve kök uzunluğu, koleoptil ve kök taze ve kuru ağırlığı ve su içeriği için karşılaştırılmıştır. Çimlenme; GS 308, DK 585 ve P.3167 çeşitlerinde sırasıyla 75, 150 ve 300 mM NaCl'de önemli düzeyde azalmıştır. Bu çalışmanın sonucu olarak, çimlenme ve diğer büyüme parametreleri tuz seviyesinin artmasıyla önemli düzeyde azalmıştır. Koleoptil dokusuna göre kök dokusu tuzluluktan daha fazla etkilenmiştir. Bulgularımız temelinde, GS 308 mısır çeşidinin, DK 585 ve P.3167 mısır çeşitlerine göre muhtemelen çok daha hassas olarak kabul edilebilir.

Anahtar kelimeler: *Zea mays* L., NaCl stresi, çimlenme, büyüme parametreleri

1. INTRODUCTION

Salinity is a major abiotic stress reducing the yield of a wide variety of crops all over the world [1]. Due not only to climatic conditions, but also to intensive, poorly controlled agriculture, the surface of NaCl-affected soils has continuously increased during the past decades.

Germination of seeds is greatly influenced by salinity [2]. In the agricultural areas affected by salt, seed germination and seedling growth emergence may be reduced, delayed and spread over time. Salinity is reported to decrease as well as delay germination of most of the crops. Lower levels of salinity delayed germination whereas higher levels in addition, reduced the final percentage of seed germination [3]. In *Triticum durum* Desf. cultivars (Belikh, salt-resistant and Cando, salt-sensitive), final germination percentage was not affected by 100 mM NaCl and it was significantly reduced in response to the highest concentration (300 mM) of NaCl [4].

Since salt stress involves both osmotic stress and ionic stress, growth suppression is related to directly total concentration of soluble salts or osmotic potential of soil water [5]. Suppression of growth occurs in all plants, but their tolerance levels and rates of growth reduction at lethal concentrations of salt vary widely among different plant species. Many plants develop mechanisms either to exclude salt from their cells or to tolerate its presence within the cells. Most plants respond to salt tolerance as glycophytes and salt tolerance may depend on the capacity to maintain absolutely low levels of Na^+ and Cl^- in tissues [6]. Maize (*Zea mays* L.) is a glycophytic plant that is sensitive to growth inhibition and damage by salt stress. In Turkey, production of maize among other cereals is 2.1 million tones [7]. Degree of salt tolerance of maize is a moderately sensitive. Maximum salinity level at which maize has no yield loss is 1.7 dS/m [8]. The physiological mechanisms have often been considered to be primarily responsible for the growth inhibition induced by salinity: turgor pressure reductions in expanding tissues and direct effects of accumulated salt on critical metabolic steps in dividing and expanding cells [9]. A successful crop stand depends on the establishment of young seedlings. Prolonged exposure to substrate salinity results in an extremely poor stand [10] caused by seedling mortality [11]. This may be more pronounced in the case of glycophytes owning to their high sensitivity to salinity [12].

Some maize cultivars are grown as a second yield in Southeastern Anatolia Region of Turkey. In this region, soil salinity is one of the most common stress problems in crop production under irrigation. Such areas affected by salt need to introduction of salt tolerant crops with some appropriate soil amelioration treatments. The aim of the present study was to determine the effect of salinity on the three maize (*Zea mays* L.) cultivars (GS 308, DK 585 and P.3167) evaluated for their salt tolerance/sensitivity on the basis of magnitude of seed germination and growth parameters.

2. MATERIALS and METHODS

2.1. Seed Material

In this research, the seeds of three maize (*Zea mays* L.) cultivars (GS 308, DK 585 and P.3167) were obtained from Ankara Seed Breeding Certification Center.

2.2. Germination and Growth Conditions

The selected seeds were surface sterilized in 3% sodium hypochlorite solution for 15 min. Then, seeds were rinsed with distilled water three times. Six salt concentrations [0 (distilled H_2O), 75, 150, 225, 300 and 375 mM NaCl] were used based on a preliminary test for salt tolerance of the cultivars. Seeds were imbibed with six different salinity concentrations for 12 h. The imbibed seeds were then germinated on two folds of filter paper placed in germination cups. The germination cups were placed to growth chamber using a completely randomized block design. Six replicates of 15 seeds were used for each treatment. Seed germination and seedling growth at these salt concentrations were tested in growth chamber at 25±1°C in dark for 6 days. A seed was considered to have germinated at the emergence of radicle [13].

2.3. Determination of Germination and Seedling Growth Parameters

Germination counts were noted every day during six days. The experiment was finished at the end of sixth days. The data for coleoptile and root lengths were recorded. Also, fresh weights (FW) of coleoptile and root samples were determined. The samples were then dried at 80°C for 48 h, and their dry weight (DW) was recorded.

The final germination percentage was calculated as the ratio between the number of seeds germinated and the total number of seed used x 100. The germination rate index (GRI) was calculated from formula described by Bouton et al. [14] and modified by Fowler [15]:

$$GRI = (G_3/3) + (G_6/6)$$

where G_3 and G_6 are germination percentages at 3 and 6 d after initiation of germination.

The absolute water content (AWC) of coleoptile and root was calculated using following formula:

$$AWC\ (\%) = [(FW-DW)/FW] \times 100$$

2.4. Statistical Analysis

A completely randomized block design was used with six replicates. Germination percentage data were arcsine transformed before statistical analysis. Statistical variance analysis of all the data was performed using SPSS, version 10, and compared with Duncan's multiple range at 0.05 level.

3. RESULTS AND DISCUSSION

Genetic variability within a species offers a valuable tool studying mechanism of salt tolerance. One of these mechanisms depends on the capacity for osmotic adjustment that allows growth to continue under saline conditions [9]. One approach to reducing the deleterious effects of salinity on crop production is the development of salt-tolerant cultivars [16].

Seed germination is one of the most critical periods for a crop subjected to salinity [17]. In this study, final germination percentage was decreased by increasing NaCl levels (Fig. 1a). Germination in all the cultivars was not observed at 375 mM NaCl. Therefore, this concentration was not added to statistical evaluation. First decrement was found significantly at 75, 150 and 300 mM NaCl in GS 308, P.3167 and DK 585 cultivars, respectively. *Arabidopsis* (a glycophytic plant) seed germination was greatly impaired at salt concentrations at or above 75 mM [18]. This decrease is probably the result of a physico-chemical effect, either osmotic or toxic [13]. Seed reserves are utilized in the growth of the embryo and the elongation of young tissues and involve the turnover and de novo synthesis of macromolecules. Germinating seeds in saline media exhibit a lowered and delayed production of radicle and plumule [19]. While germination was not observed at or above 225 mM in GS 308, was observed at 300 mM in DK 585 and P.3167. The 300 mM NaCl was highest salinity level which germination was observed for P.3167 (31.1%) and DK 585 (58.9%). Salt stress inhibits seed germination through a limitation of water uptake arresting radicle emergence, although a toxic influence of salt cannot be excluded [17]. Differences between cultivars were significant level at control and 75 mM NaCl. In contrast, there is no difference between DK 585 and P.3167 against salinity at 150 mM NaCl. Also, the difference between these cultivars was observed at 225 and 300 mM NaCl. Germination rate index decreased significantly with increasing salinity level of germination medium (Fig. 1b). Some researchers [6, 20-22] have reported that rate index and percentage of seed germination are affected adversely as salinity (especially NaCl) increases and the osmotic potential of the germination medium decreases. Çiçek and Çakirlar [23] suggested that determining salt tolerance of two maize cultivars at -0.2, -0.5 and -0.8 MPa NaCl:$CaCl_2$ (2:1) culture solutions is not possible at the end of germination periods for 5 days. In contrast to this result, based on our findings, the salinity tolerance of maize cultivars in final germination may classify probably as sensitive (55.6% at 75 mM in GS 308), moderately sensitive (31.1% at 300 mM in P.3167) and moderately tolerant (58.9% at 300 mM in DK 585) in studied NaCl concentrations. This salinity tolerance classification was determined on the basis of the salinity levels resulting in a reduction in final germination at 6 d.

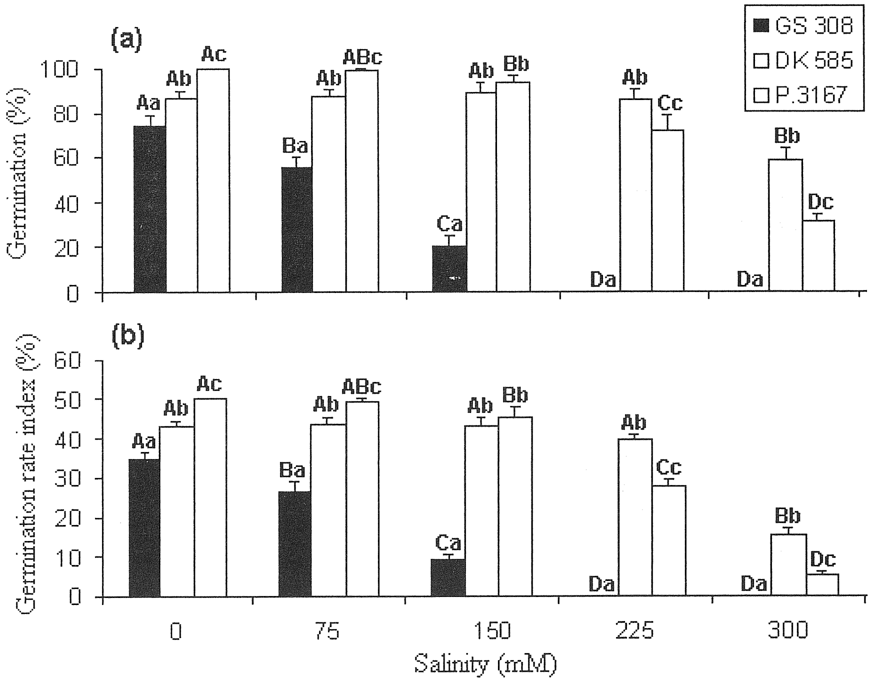

Fig. 1. Final germination percentages (a) and germination rate index (b) of three maize cultivars as a response to six salinity (NaCl) levels at 25°C. Different capital letters above column represent significantly differences of treatment levels within a cultivar ($P \leq 0.05$). Different letters above column represent significantly difference among cultivars within a treatment ($P \leq 0.05$). Bars represent mean ± SE.

Some of the morphological changes in salinity are reduction of shoot [24] and root length [25]. In *Arabidopsis*, higher than 50 mM of NaCl inhibits plant growth and eventually kills the plants [18]. In our study, the coleoptile and root lengths (CL and RL, respectively) of three maize cultivars decreased significantly by increasing NaCl levels (Fig. 2a and b): The root tissue was more effected than coleoptile tissue. In contrast to our finding, in soybean plants, it was found that growth of shoot was more sensitive to salinity than root growth [26]. Salinity affected more significantly the two parameters of GS 308 than that of DK 585 and P.3167. While the CL of P.3167 was higher than other cultivars at control, 75 and 150 mM NaCl, the CL of DK 585 was higher than P.3167 at 300 mM NaCl. The RL of DK 585 cultivar was higher than other cultivars at high salinity concentrations as 225 and 300 mM NaCl. Plant growth is ultimately the direct result of massive and rapid expansion of the young cells produced by meristematic divisions. The growth of cells is primarily correlated with turgor potential, decreased turgor is the major cause of inhibition of plant growth under saline conditions. Cell expansion in plant tissues can be inhibited by salinity [9]. In two maize cultivars (C 6127 and DK 623), CL and RL decreased significantly by increasing salt treatment levels, -0.5 and -0.8 MPa [23].

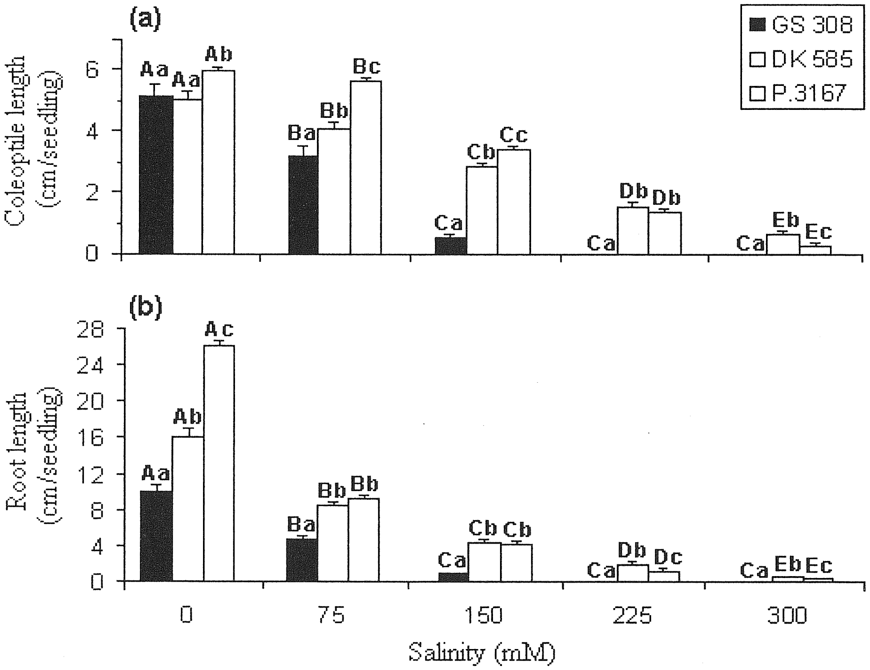

Fig. 2. Effects of salinity on coleoptile **(a)** and root **(b)** length of three maize cultivars. Letters and bars are same as in Fig. 1.

The deleterious effect of salinity was suggested as a result of water stress, ion toxicities, ion imbalance, or combination of all these factors [27]. Salt stress results in a considerable decrease in the fresh and dry weight, absolute water content of stems, roots, and leaves [21, 28-30]. In present study, salinity treatment resulted in decreasing the fresh weight (FW), dry weight (DW) and absolute water content (AWC) of the coleoptile (Fig. 3) and root tissues (Fig. 4) as compared to their respective control values in three cultivars at different levels of salinity. AWC was not changed at 75 mM NaCl compared to control. Decrease in FW, DW and AWC at various levels of salinity in both coleoptile and root was more in GS 308 as compared to that of other cultivars. Note that even at 300 mM salinity level, DK 585 and P.3167 cultivars were able to maintain the water content at a higher salinity level as compared to that of GS 308 at 150 mM salinity in both coleoptile and root. In control, 75, and 150 mM salinity levels, P.3167 had more FW and DW of coleoptile and root than other maize cultivars. Absolute water content of coleoptile decreased significantly in all cultivars at 150 mM and higher salinity levels whereas AWC of root decreased significantly in only GS 308 at 150 mM NaCl. In high salinity (300 mM), root AWC of DK 585 was higher significantly than other cultivars. According to coleoptile AWC, there is difference between DK 585 and P.3167 at 300 mM NaCl. Higher water content in coleoptile and root of sensitive cultivar under control suggest varied behaviors for water uptake during germination of seeds differing in salt tolerance. The deleterious effect of salinity on seedling growth was greater in root DW than coleoptile DW. The similar result was reported in *Phaseolus aureus* cultivars by Misra and Dwivedi [22]. In green gram (*Phaseolus aureus*) cultivar SML-32 (salt sensitive), increasing levels of salinity decreased seed germination and caused pronounced decrease in root FW and DW, and absolute water content in shoot and root tissues compared to cultivar T-44 (salt tolerant) [22]. In our study, with increasing salinity there was a decrease in FW and DW, and water uptake in three maize cultivars. In the cultivar GS 308, the magnitudes of such a decrease were more as compared to that of cultivars DK 585 and P.3167. Saneoka et al., [31] reported that leaf, stem and shoot dry weight in two wheat lines (SARC and Pothowar) decreased by 200 mM NaCl, whereas the leaf dry weight was more affected by salinity than the stem and shoot dry weight. It is suggested that decrease in seed germination and depression in seedling vigor under saline stress is attributed to decrease water uptake followed by limited hydrolysis of food reserves in storage tissues as well as due to impaired translocation of food reserves from storage tissue to developing embryo axis [32, 33].

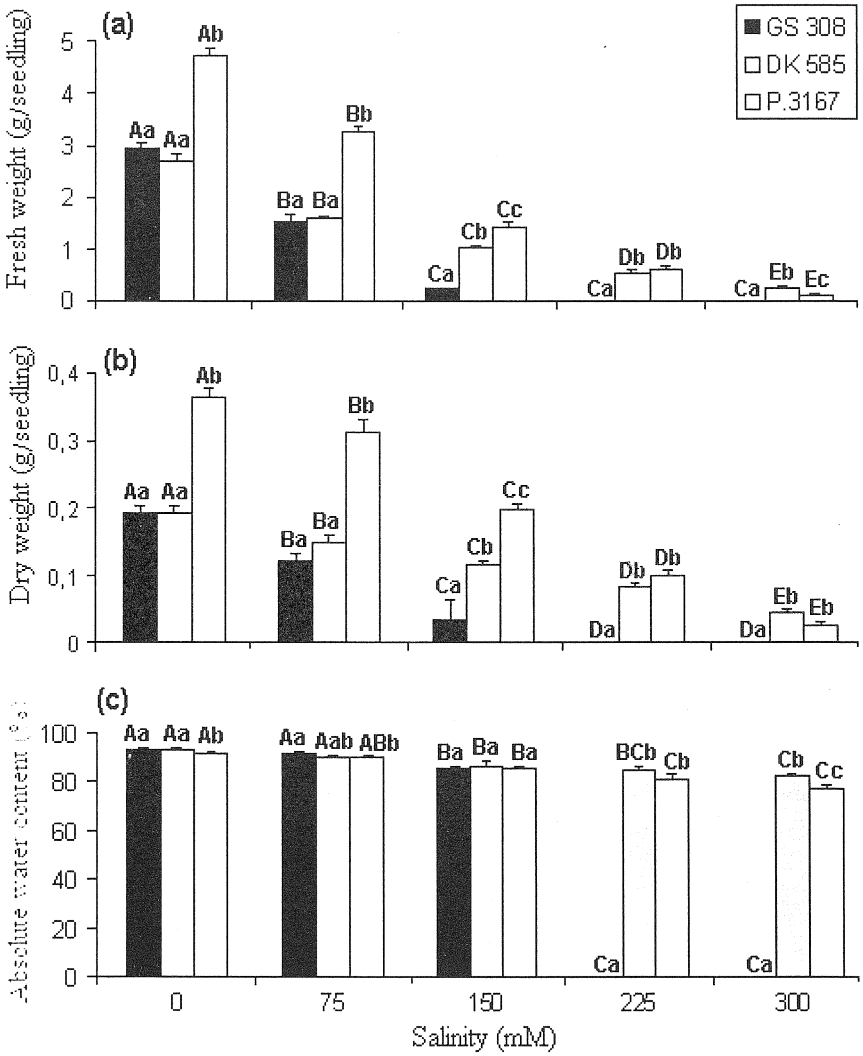

Fig. 3. Effects of salinity on coleoptile fresh weight **(a)**, dry weight **(b)** and absolute water content **(c)** of three maize cultivars. Letters and bars are same as in Fig. 1.

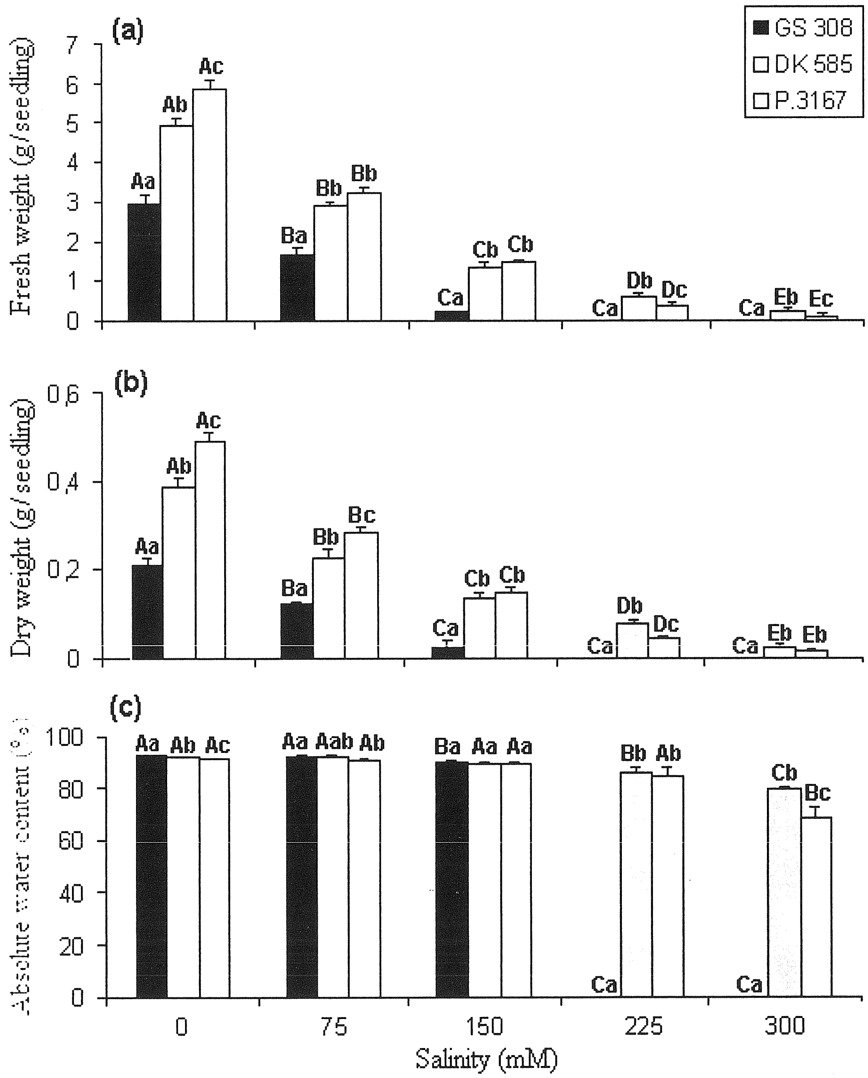

Fig. 4. Effects of salinity on root fresh weight **(a)**, dry weight **(b)** and absolute water content **(c)** of three maize cultivars. Letters and bars are same as in Fig. 1.

In conclusion, the results of this study demonstrate that salt tolerance during germination and early seedling growth exists within maize cultivars. Based on our findings, GS 308 maize cultivar may accept probably more sensitive than DK 585 and P.3167. However, classification for salt tolerance of maize cultivars should be supported by physiological and biochemical studies.

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