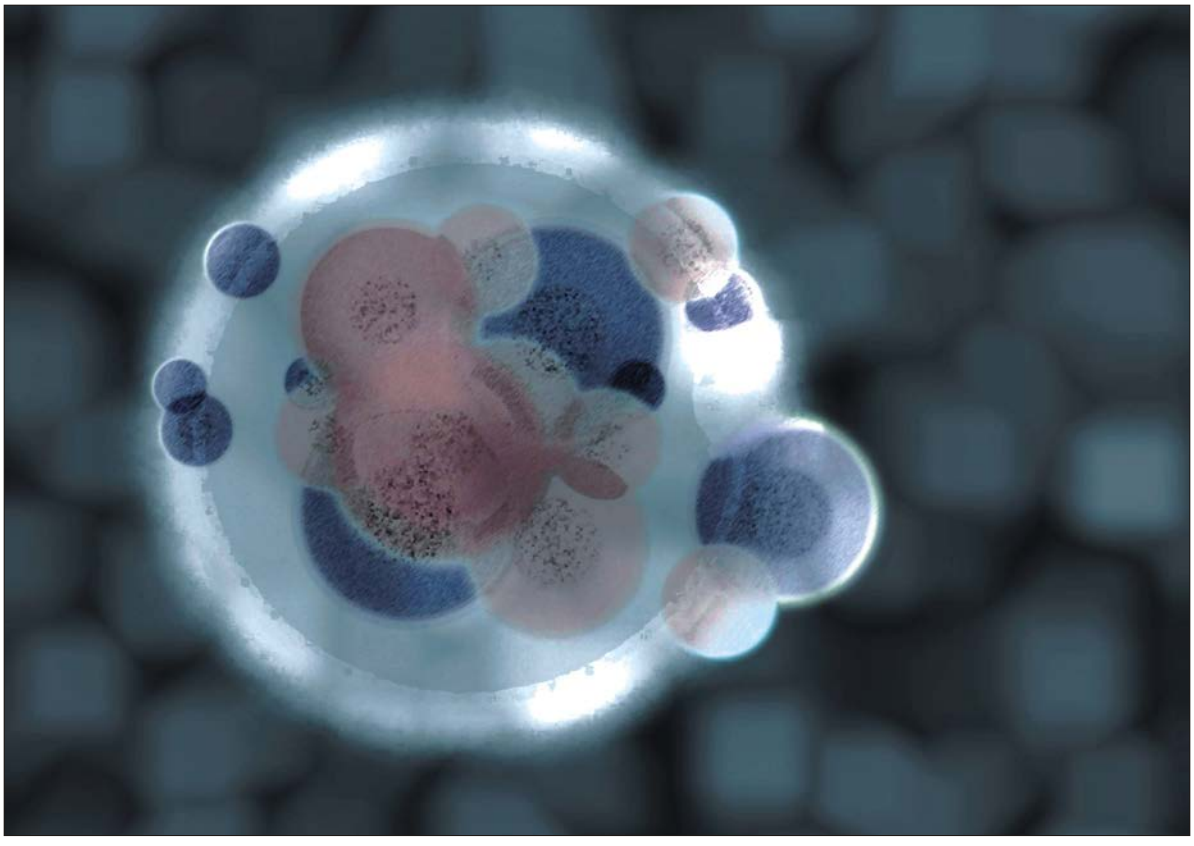

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Accumulation and Role of Ions (Ca^{2+}, Mg^{2+}, SO_4^{2-}) on Salt Tolerance in *Triticum turgidum* L.

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Abstract: Understanding the mechanisms of salt tolerance and the physiological and biochemical factors are very important in plant crops. Salinity-minerals (Ca^{2+}, Mg^{2+}, SO_4^{2-}) interactions, which have been shown to be important in plants grown in saline conditions, were studied in durum wheat (*Triticum turgidum* L.). Two genotypes of durum wheat, one salt-tolerance (ICDW751) and the other salt-sensitive (ICDW324) were grown in nutrient solution containing 0, 50, 100 and 150 mM. NaCl. Amount of minerals of Ca^{2+}, Mg^{2+} and SO_4^{2-}were determined in the shoot and roots. Usually, salinity reduced Ca^{2+} and Mg^{2+} contents in shoot and root of both genotypes, mainly in the salt-sensitive genotype. Apparently, SO_4^{2-} content in shoot increased as the medium salinity increased, especially in the salt- tolerance genotype. Salinity increased significantly Na^+/Ca^{2+} ratio and sodium adsorption ratio (SAR) = $Na^+/(Ca^{2+}+Mg^{2+})^{½}$ of shoot and root in both genotypes, mainly in the salt-sensitive genotype. Thereby, we suggested that ratio of Na^+/Ca^{2+} and especially amount of SAR in durum wheat (*Triticum turgidum* L.) are suitable for discrimination salt-tolerance genotype from salt-sensitive genotype.

Key words: *Triticum turgidum*, salinity, accumulations of Ca^{2+}, $Mg^{2+}$and SO_4^{2-}, tolerance genotype

INTRODUCTION

Deleterious effects of salinity are thought to result from low water potentials, ion toxicities, nutrient deficiencies, or a combination of these factors. Nutrient deficiencies can occur in plants when high concentrations of Na^+ in the soil reduce the amounts of available Ca^{2+} and Mg^{2+} or when Na^+ displaces membrane-bound Ca^{2+} (Khan *et al*., 2000). Calcium is an essential element in all plant (Marschner, 1995). Calcium can have a protective effect on plant growth and survival under high salinity (Davenport *et al*., 2005). Ca^{2+} uptake and concentration decreases in plant cells and tissues as the external Na^+ concentration increase (Lazof and Bernstein, 1999). Also, in wheat reduction of the Ca^{2+} concentration due to salinity was reported by Meneguzzo *et al*. (2000). Furthermore, Allen *et al*. (1995) reported that Na^+ influx to cell by calcium can be inhibits in durum wheat. Lazof and Bernstein (1999) reported that accumulation and influx in Ca^{2+} to cell decreases by salinity as to increasing external Na^+, accumulation and influx in Na^+ was increased whilst accumulation and influx in Ca^{2+} decreased. Houshmand *et al*. (2005) reported that in durum wheat, shoot Ca^{2+}/Na^+ ratio increased with increase in NaCl concentration of the growth medium In durum wheat, Meneguzzo *et al*. (2000) indicated that Ca^{2+} content in cell was decreased by salt stress that this decrease can be role of the signal calcium inhibits in mechanism of tolerant to salinity. In durum wheat, reports for the effect of salinity on the Mg^{2+} content not found, nevertheless in the other plants are the reports. For example, Khan *et al*. (2000) reported that Mg^{2+} content was reduced in shoots of *Atriplex griffithii* var. stocksii plants grown at high salinity. Also, in response to such stress conditions the concentrations of the major essential ion such as Mg^{2+} in *Plantago coronopus* (L.) transiently decreased (Koyro, 2005). The Mg^{2+} contents of both stem and root greatly increased in *Aloe vera* plants, however leaf Mg^{2+} content had no obvious change in the presence of salinity (Jin *et al*., 2007). The combined inhibitory effects of both Ca^{2+} and K^+ on Mg^{2+}uptake may have led to the relatively poor Mg^{2+}status of alfalfa plants grown at salt stress (Grieve *et al*., 2004). In mostly studied, for the effect of salinity on the accumulation of SO_4^{2-} in plants is very little attention. Nevertheless, More and Manchanda (1992) reported that in table pea plants, salinity of due to salt of chloride, causes reduction of SO_4^{2-} content in shoot and leaves and increasing it's in roots. Furthermore, White and Broadley (2001) indicated in soil that SO_4^{2-} could be inhibits Cl^- uptake in to the plants. On the base of these concepts, the object of present research is to evaluate which are the effects of salinity stress on the Ca^2,Mg^{2+}

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and SO_4^{2-} accumulations for finding correlation between accumulation elements and salt tolerance in durum wheat.

MATERIALS AND METHODS

This experiment was conducted at the Azad University of Islamic, Branch Science and Research, Tehran-Iran in 2005. Seeds of two durum wheat [*Triticum turgidum* L. subsp. durum (Desf.) Husn.] genotypes (preliminary studies at germination stage showed that ICDW751 from Syria and ICDW324 from Iran are salt tolerant and salt sensitive genotypes, respectively) were provided by Agricultural Biotechnology Research Institute Iran (ABRII). A factorial experiment with two factors of genotypes with two levels and salinity with four levels (0, 50, 100 and 150 mM NaCl) was used. The treatment combinations were replicated three and arranged in a completely randomized design. Seed were surface sterilized in sodium hypochlorite solution 5% and rinsed with distilled water. Their were placed on the half strength nutrition solution (Clark nutrition) in the greenhouse under day/night temperatures of 22±2°C /15±2°C and day length of 13 h. After 3 days the nutrient solutions were replaced with full strength nutrition solution (Hadi *et al*., 2007). The solution were aired automatically 15 min h^{-1} and were renewed every 6 days. Salt stress was initiated 21 days after seed germination, by gradual adding NaCl to the nutrient solutions. To avoid osmotic shock, NaCl was added twice daily to increments of 50 mM until the final concentrations of 100 and 150 mM NaCl were achieved. Plants were harvested 30 days after commencing treatments, separated into shoot and root washed with distilled water. Plants samples were oven dried (75°C to constant mass) and weighed. Plant samples were ashed at 500°C for 3 h. Inorganic ions were then extracted with 10 mL 1M H_2SO_4 and the volume of each sample was standardized to 100 mL and Na^+, Ca^{2+} and Mg^{2+} concentrations of the solutions were determined using an atomic absorption spectrophotometer (Kamnev *et al*., 1997). SO_4^{2-} concentrations of the solutions were determined using an Ion chromatograph (Cataldi *et al*., 2003).

RESULTS AND DISCUSSION

Calcium is a non-toxic inorgamic nutrient and has a function of detoxification under saline medium (Jin *et al*., 2007). Figure 1 shows the effects of salinity on the Ca^{2+} accumulation at shoot (A) and root (B) in two durum wheat genotypes. The results show that as the medium salinity increased, Ca^{2+} contents of shoot and root in two genotypes were decreased significantly (Fig. 1a, b).

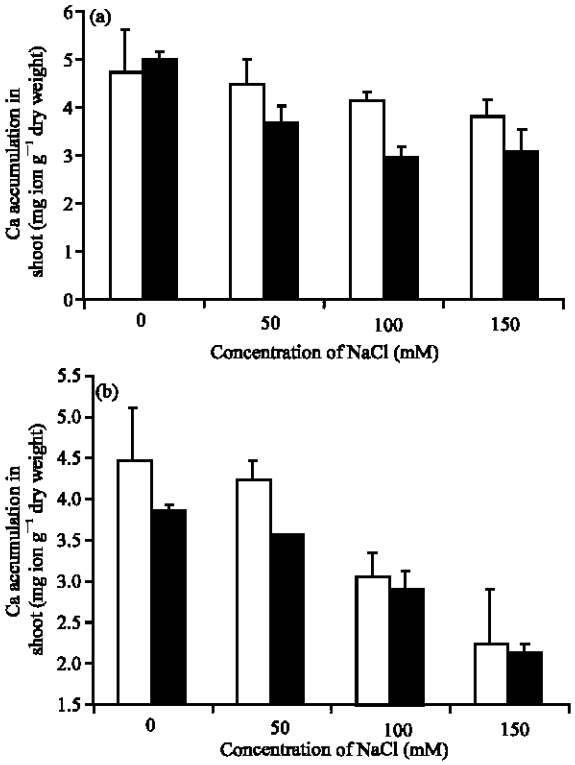

Fig. 1: The effects of salinity on the Ca^{2+} accumulation at shoot (a) and root (b) in two durum wheat genotypes. In each salt treatment from left to right are genotypes of ICDW751 and ICDW324, respectively. Vertical bars indicate±Standard error

Meneguzzo *et al*. (2000) reported similar results in wheat. This response was undoubtedly the result of increasing concentrations of Na^+ in the nutrition solution as this cation may not only reduce Ca^{2+} activity in solution, but also may displace Ca^{2+} from its extracellular binding sites within plant organs and further disrupt Ca^{2+} acquisition. Also, Jin *et al*. (2007) indicated that salinity restricted Ca^{2+} uptake and transport from root. Thereby Ca^{2+} contents in all plant parts decreased under salt stress. Furthermore, the results show that Ca^{2+} content in ICDW751 genotype (shoot and root) had the higher than ICDW324 genotype then grown in 150 mM medium (Fig. 1). Jin *et al*. (2007) reported that in *Aloe vera* under salt stress, Ca^{2+} contents of leaf and stem decreased and Ca^{2+} content of leaf in salt-tolerance genotype was three times higher than that in salt-sensitive genotype. Thereby, it seems that high Ca^{2+} contents of shoot and root in ICDW751 genotype can be related to salt-tolerance in this genotype. Hasegawa *et al*. (2000) indicated that content increase of Ca^{2+} intercellular could be cause decrease influx in Na^+ and also increase selection of K^+ respect to Na^+ and thus improve effects of

salinity. On the other hand, Hadi *et al.* (2007) reported that in durum wheat, Na^+ content of shoot and root in salt sensitive genotype had the higher than the salt tolerance genotype whilst K^+ content in salt sensitive genotype had the lower than the salt tolerance genotype than grown in 150 mM NaCl. Therefore, it seems that high accumulation of Ca^{2+} intercellular in the ICDW751 genotype has been improved effects of salinity. Also, Hasegawa *et al.* (2000) indicated that Na^+ with Ca^{2+} by competitive inhibits influx in calcium canals and also by Na^+ displaces membrane-bound Ca^{2+} can be cause turbulence of cell membrane. Thereby, it seems that salt-tolerance genotype (ICDW751) have lower influx of Na^+ than salt-sensitive genotype (ICDW324). Furthermore, the results show that shoots Ca^{2+} content of both genotypes in all salt concentrations had grater than root (Fig. 1). Ramoliya *et al.* (2004) reported similar results in *Salvadora persica.* They also indicated that Ca^{2+} was transferred from roots to leaves at high salinities. Therefore, the increased Ca^{2+} content may reduce the toxicity of Na^+ in leaves. Thereby, it is reasonable to believe that higher salt-tolerance in ICDW751 genotype are due to the higher Ca^{2+} content in shoot and root of plants. These results confirm the importance of Ca^{2+} interaction with salinity stress and indicate differences in both durum wheat genotypes response. The results show that Mg^{2+}contents of shoot and root in two genotypes grown in 150 mM NaCl were decreased significantly in comparison with control (Fig. 2). Nearly, new report that shows the effects of salinity on Mg^{2+} uptake and accumulation is not in wheat and it is very little in the crop. Nevertheless, Datta *et al.* (2007) reported that in mangroves plants, salinity imposed reduction in Mg^{2+} uptake and accumulation in leaves. Datta *et al.* (2007) indicated that in mangroves plants, salinity affects ion accumulation of Mg^{2+} in leaves, thereby membrane permeability and chlorophyll synthesis. In inverse, Jin *et al.* (2007) reported that in *Aloe vera* plants, Mg^{2+} contents of both stem and root greatly increased and leaf Mg^{2+} content had no obvious change and in the presence of salinity. Donovan *et al.* (1997) reported that uptake and transport of Mg^{2+} relative to Na^+ increased. They indicated that maintaining leaf Mg^{2+} might be especially necessary at the highest salinity because it is a cofactor for tonoplast ATPases. In addition, Marschner (1995) reported that maintaining leaf Mg^{2+} might also be necessary for protein translation in the presence of high Na^+. On the other hand, Ramoliya *et al.* (2004) reported that the role of magnesium in chlorophyll structure and as an enzyme cofactor, another important role of magnesium in plants is in the export of photosynthesis, which is impaired and leads to enhanced

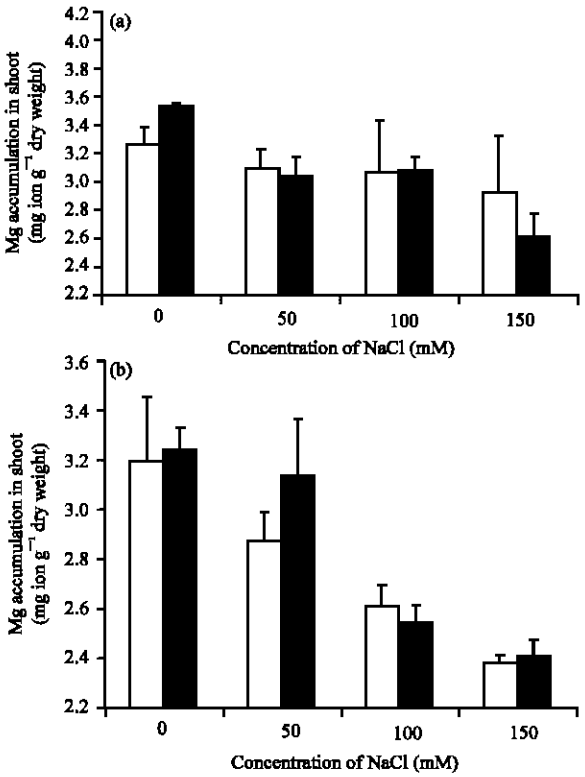

Fig. 2: The effects of salinity on the Mg^{2+} accumulation at shoot (a) and root (b) in two durum wheat genotypes. In each salt treatment from left to right are genotypes of ICDW751 and ICDW324, respectively. Vertical bars indicate±Standard error

degradation of chlorophyll in magnesium deficient source leaves, resulting in increased oxygenase activity of ribolose bisphspate carboxylase. Therefore, it seems that the decreases of Mg^{2+} content of shoot, which is due to increasing of salinity could be decreased in the export of photosynthesis in durum wheat. The results show that salinity had affected significantly on the SO_4^{2-} contents of in both genotypes, however it had an uncertain trend (Fig. 3). Also, as the medium salinity increased, SO_4^{2-} content of shoots in ICDW751 genotype was increased significantly in comparison with control (Fig. 3). Inverse, SO_4^{2-} content of root at the 50 and 100 mM NaCl concentrations of medium was decreased significantly in comparison with control (0 mM NaCl). Very little attention has been given to the influence of salinity on sulfate uptake and accumulation in crops. Nevertheless, Gupta *et al.* (2003) reported that as the soil salinity increased, the SO_4 contents increased in chrysanthemum leaves. Also, Carter *et al.* (2005) reported that in *Celosia argentea* under salt stress, SO_4^{2-} increased in plant tissues. In addition, Grieve *et al.* (2001) found similar

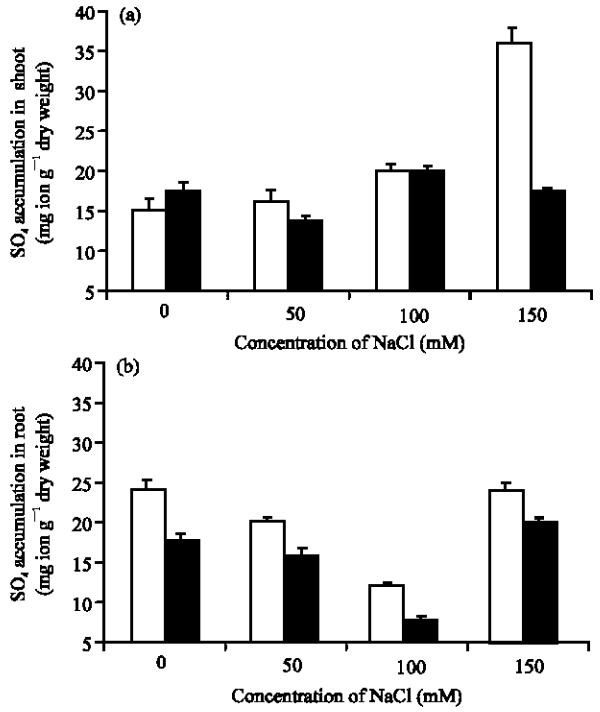

Fig. 3: The effects of salinity on the SO_4^{2-} accumulation at shoot (a) and root (b) in two durum wheat genotypes. In each salt treatment from left to right are genotypes of ICDW751 and ICDW324, respectively. Vertical bars indicate ± Standard error

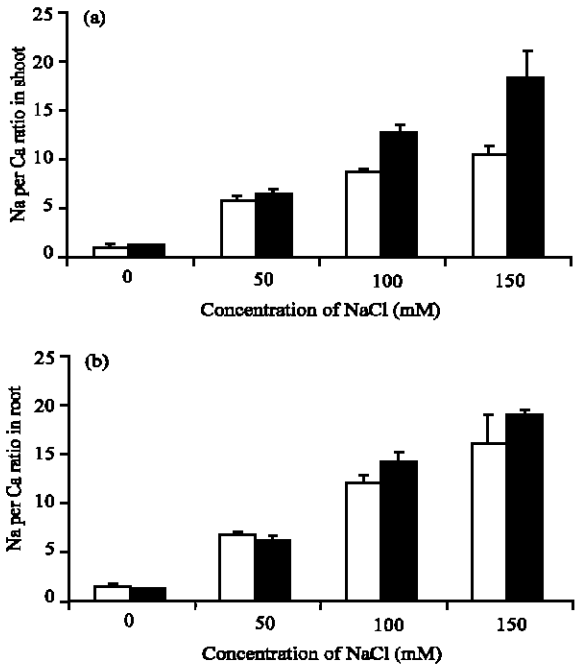

Fig. 4: The effects of salinity on the Na^+/Ca^{2+} ratio at shoot (a) and root (b) in two durum wheat genotypes. In each salt treatment from left to right are genotypes of ICDW751 and ICDW324, respectively. Vertical bars indicate ± Standard error

responses with a study of nine leafy vegetables. They also found that *Beta vulgaris*, *Cichorium endivia* and *C. intybus* showed an increase in SO_4^{2-} uptake as substrate salinity increased. In this study, results show that SO_4^{2-} content of shoot and root in the ICDW751 genotype had the higher than ICDW324 genotype then plants grown in 150 mM NaCl (Fig. 3). There it seems that high SO_4^{2-} content of shoot and root in ICDW751 genotype could be relative with high salt-tolerance this genotype. Because, White and Broadley (2001) indicated that in alfalfa plants under salt stress, SO_4^{2-} had inhibition of Cl^- uptake. On the other hand, Hadi *et al.* (2007) reported that in durum wheat, Cl^- content in salt-tolerance genotype had the lower than salt-sensitive genotype then plants grown 150 mM NaCl. So, lower Cl^- concentration in this genotype could be lower toxicity and thereby this genotype had the higher salt-tolerance. Furthermore, understanding the mechanisms of salt tolerance and the physiological and biochemical factors that impact these processes are very important in the selection of tolerant crop varieties. For example, ions ratios and interaction between they are very important in plants. The majority of these interactions are directly related to the concentrations and ratios of ions in the root zone. The influence of these external ion ratios on ion accumulation by salt-stressed plants is complex. The results show that as the medium salinity increased, Na^+/Ca^{2+} ratios of shoot and root in both genotypes were increased (Fig. 4). However, this ratios in ICDW751 genotype is the lower than ICDW324 genotype (Fig. 4). Houshmand *et al.* (2005) reported that higher Ca^{2+}/Na^+ ratio in genotype of tolerant to salinity than other genotypes may be accounted for as one of the reasons for high grain yield production of this durum wheat genotype under salt-stressed field experiment. Thereby, it seems that lower Na^+/Ca^{2+} ratio in ICDW751 genotype than ICDW324 genotype may be accounted for as one of the reasons for high salt-tolerant of this durum wheat genotype under salt-stressed. Further, Jin *et al.* (2007) indicated that in *Aloe vera* plants, genotype of salt tolerant maintained significantly lower Na^+/Ca^{2+} ratios than genotype of sensitive and lesser degree of membrane injury, lower Na^+/Ca^{2+} ratio and the salt-induced enhancement of osmotic adjustment in *Aloe vera* indicate that the relatively salt tolerant cultivar had a higher prevention capacity for a large and permanent efflux of Ca^{2+}. Thereby, it seems that may be

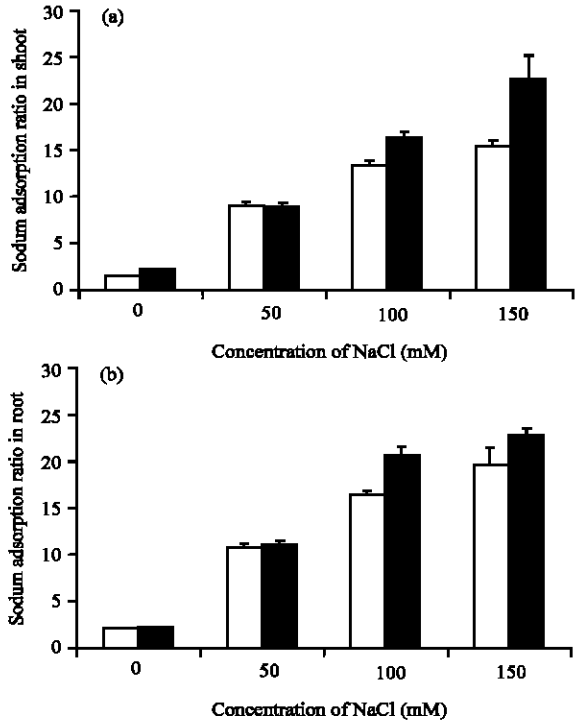

Fig. 5: The effect of salinity on the Sodium Adsorption Ratio (SAR) = $Na^+/ (Ca^{2+}+Mg^{2+})^{½}$ at shoot (a) and root (b) in two durum wheat genotypes. In each salt treatment from left to right are genotypes of ICDW751 and ICDW324 respectively. Vertical bars indicate±Standard error

lower Na^+/Ca^{2+} ratio under salt stress the main osmotic adjustment for durum wheat is accumulating inorganic cations in root and shoot. Furthermore, sodium adsorption ratio (SAR) = $Na^+/(Ca^{2+}+Mg^{2+})^{½}$ uses in relations between water and soil especially salinity conditions (Cramer, 2002). SAR maybe used in plants. Thereby, we used SAR for durum wheat. The results show that as the medium salinity increased, amount of SAR at the shoot and root in both genotypes was increased significantly (Fig. 5). Mean comparison between both genotypes indicates that ICDW751 genotype had significantly lower amount of SAR at the shoot and root than ICDW324 genotype (Fig. 5). On the other hand, on based early results (Hadi *et al*., 2007) ICDW751 and ICDW324 are known as salt-tolerance and salt-sensitive genotypes, respectively. Thereby, it seems that amount of SAR can be used as a parameter for salt tolerant in durum wheat. We observed significant differences between ion concentrations of shoot and root in both genotypes. The single cycle selection process that led to the salt tolerance in ICDW751 genotype most likely affected a biochemical process where probably a single gene product, such as an enzyme change in a metabolic pathway, occurred. This genotype should be suitable for planting on saline soils of the Iran for improved yields.

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