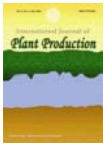

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Assessment of agro-physiological traits for salt tolerance in drought-tolerant wheat genotypes

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Abstract

Salt stress is one of the major constraints for wheat cultivation in Iran and leads to a considerable loss in crop yield each year. In high salinity soils, the reduced osmotic potential of soil solutes may cause physiological drought. In this study the salt tolerance of different drought-tolerant bread wheat genotypes were studied by examining various agronomic and physiological traits, including Na^+ and K^+ concentrations, the Na^+/K^+ ratio in leaf and spike, shoot dry weight, leaf greenness, stomatal conductance, leaf area, osmotic potential, relative water content (RWC) and grain yield. Two pot experiments were conducted using a completely randomized design with three replications. Wheat genotypes were grown in pots and irrigated either with tap water (EC=0.5 dSm^{-1}) or saline water (EC≈18 dsm^{-1}) as control and salt stress treatments, respectively. Significant differences were observed in all measured traits between control and stress treatments except for the spike potassium concentration. Differences between genotypes were significant for all traits except for RWC and osmotic potential. Among the different genotypes, one drought-tolerant genotype appeared salt tolerant, three were semi-salt-tolerant, one drought-sensitive genotype appeared semi-salt-sensitive, and two drought-tolerant genotypes appeared salt-sensitive and semi-salt-sensitive. This study shows that drought tolerance does not necessarily lead to salt tolerance. Some physiological traits including Na^+ content, leaf area, SPAD number, stomatal conductance and shoot dry weight, which are significantly correlated with grain yield and show remarkable variations among wheat genotypes, may be useful parameters for measuring the responses of other wheat genotypes to high-salinity soils in the field.

Keywords: Salt stress; Wheat; Na^+ content; Physiological characteristics.

Introduction

Salt and drought stresses have a lot of destructive effects on plants which are physiologically similar to each other (Sairam and Tyag, 2004). Several studies have addressed cross-talk between drought and salt stresses. Soil salinity is one of the major problems in vast areas of the world. Salt stress occurs in two phases: first, high concentration of soluble salts in the soil makes it harder for plant roots to extract water. This will put plant under osmotic stress. Water loss from plant cells influences the turgor and changes the size and membrane characteristics of the plant cells. Second, toxic concentrations of salts within the plant cells act as an ionic stress that limits the photosynthetic capacity and supply of carbohydrates for grain filling (Munns and Tester, 2008).

A primary response to water deficit in salt-tolerant genotypes is osmotic adjustment (Decosta et al., 2007). While Na^+ and Cl^- are sequestered in the vacuole of a cell, osmotic adjustment maintains the osmotic equilibrium by accumulation of various compatible osmolytes in cytoplasm such as K^+, proline, mannitol and glycinebetaine. It helps the plants keep their stomata open and continue their photosynthesis under salt stress (Munns and Tester, 2008).

Under soil salinity, high concentration of Na^+ competes with the uptake of other nutrients, especially K^+ as a necessary element. Salt-tolerant genotypes of wheat have a more efficient system for selective uptake of K^+ over Na^+ (Goudarzi and Pakniat, 2008). In these genotypes Na^+ and Cl^- are effectively excluded by roots or the excessive ions accumulate in the vacuole of leaf sheaths. The net sodium uptake and its ratio to potassium have a strong correlation with leaf area and crop yield (Zeng et al., 2003; Asgari et al., 2012).

Potassium is essential for various essential activities in plants such as maintenance of electric potential gradient, stomatal movements and activation of numerous enzymes and membrane proteins (Brito et al., 2008). Disturbance in potassium absorption lead to disturbances in photosynthesis and cell apoptosis via production of reactive oxygen species (ROS). ROS such as superoxide, hydrogen peroxide and hydroxyl radical result in oxidative damage to proteins, DNA and lipids. Peroxidation of membrane lipids in sensitive wheat varieties under salt stress leads to accumulation of hydrogen peroxide and malondialdehyde (Mandania, 2005) which can activate signal cascades to regulate the plant development under stress (Xiong et al., 2002). All these reactions accelerate developmental processes

and lead to decrease of leaf number and leaf area, shoot dry weight, tillage number and as a result the spike number, seed number and finally decrease of grain yield (Perwise, 2002; Hussain et al., 2003). However at low level of salinity, it is possible that decrease of leaf area and shoot biomass do not lead to grain yield reduction and the salinity should be reached to a threshold level to decrease the grain yield. The death of leaves is started by accumulation of ions in older leaves. The rate of leaf decline is important for plant survival under salinity. If the rate that new leaves are produced is greater than the death of old leaves and photosynthetic products are enough for flower and seed growth, the plant completed its growth period although the seed number is decreased (Munns et al., 2006).

Studies on the effects of salt stress on different wheat genotypes have shown that sodium and potassium content and their ratio and shoot dry matter are appropriate traits for screening wheat genotypes for salt tolerance (Goodarzi and Pakniat, 2008). It is also reported that genotype ranking in terms of shoot dry weight lead to the same result as grain yield ranking (Elhendawy et al., 2011).

In this research the wheat drought-tolerant genotypes were compared under severe salinity based on some agro-physiological traits and the importance of drought tolerance under salt stress condition was studied.

Methods and Materials

Growth conditions

Two experiments were carried out in two seasons (2010-10 and 2011-02) at the greenhouse of the Agricultural Biotechnology Research Institute of Iran (ABRII). The soil of the first experiment was loam (25% clay, 28% sand, 47% silt) with a soil EC of 3.5 dSm^{-1} and the soil of second experiment was silt clay loam (37% clay, 16% sand, 47% clay) with a soil EC of 2.8 dSm^{-1}.

Plant materials and treatments

Seven differently drought-tolerant wheat (*Triticum aestivum* L.) genotypes were used in the first experiment (Table 1). The seeds were kindly provided by Dr. Ehdaie and Dr. Reynolds. The genotypes No. 14, No. 49, Bam and Ghods were used in the second experiment. Bam as a salt-

tolerant and Ghods as a salt-sensitive varieties were obtained from the Seed and Plant Improvement Institute, Karaj, Iran. In both experiments five seeds of each genotype were sown in each 3 kg pot.

Table 1. Characteristics of the wheat genotypes were used in the first experiment.

Genotype	Characteristics
No.14	Landrace, from South West of Iran, drought-tolerant (Ehdaie et al., 2006)
No.49	Landrace, from East and center of Iran, drought-tolerant (Ehdaie et al., 2006)
C_4	Recombinant inbred line from Seri*Babax, drought-tolerant (Reynolds and Condon, 2007)
C_6	Recombinant inbred line from Seri*Babax, drought-tolerant (Reynolds and Condon, 2007)
C_{15}	Parental line of Seri*Babax, drought-sensitive(Reynolds and Condon, 2007)
C_{168}	Parental line of Seri*Babax, drought-tolerant (Reynolds and Condon, 2007)
C_{169}	Recombinant inbred line from Seri*Babax, drought-tolerant (Xue et al., 2008)

The genotypes were compared at two salinity levels for their salt tolerance (control EC_{water}: 0.5 dS m^{-1}, stress EC_{water} for the first experiment: 18 dS m^{-1} and for the second experiment: 15 dS m^{-1}). Treatments were replicated three times in a completely randomized factorial arrangement. Treatments were applied through irrigation with saline water when the third leaf emerged.

Tissue sampling

At Zadoks scale 47 (Zadoks et al., 1976), about 8 weeks after imposing salt stress (EC_{soil}=14 to 16 dSm^{-1}) two flag leaves per replicate were used for all physiological measurements except RWC that was done with the second leaf from the top. At physiological maturity (Zadoks 90) spike Na^+ and K^+, shoot dry weight and grain yield measured from four plants per replicate (EC_{soil}: 20 dSm^{-1}). All sampling was done at 9:00 to 11:00 am except stomatal conductance that was done at 7:00 am.

Agronomic measurements

Leaf area (LA) was determined nondestructively by measuring the length and greatest width of each leaf blade (assuming length×width×0.75) (El-Hendawi et al., 2009). Shoot dry weight, seed number and seed weight used for yield estimation were assessed after 48 hrs in 70 ˚C at physiological maturity. Salt sensitivity index (SSI) and salt tolerance index (STI) were calculated for each genotype using the following formula (Fischer and Maurer, 1978):

$$SSI = \frac{1-(GY_s / GY_p)}{1-D}$$

Where GY_s is the mean of genotype under salt stress and GY_p the mean of genotype under non-stress (control) conditions. D is the ratio of the overall mean of all genotypes under stress to the overall mean of all genotypes in control condition. Salt tolerance index (STI) was calculated for the grain yield of each genotype as:

$$STI = \frac{Y_s}{Y_c}$$

Where Y_s and Y_c are the means of the genotype yield under salt stress and control condition, respectively.

Physiological measurements

To compare the genotypes, Na^+ and K^+ content of flag leaf and spike were measured using standard flame photometry procedure (Munns et al., 2010). Leaf greenness (SPAD number), stomatal conductance and leaf osmotic potential were measured using SPADmeter (MINOLTA-502, Japan), Porometer (Delta-T AP4, England) and Osmometer (WESCOR C5022, USA) respectively. Leaf relative water content (RWC) was also measured as described by Danda and Seti (1998). Chlorophyll content was analyzed by spectrophotometry method (Porra, 2002).

Statistical analysis

Data were analyzed using SAS (version 9.0) statistical package. Mean comparisons were performed using least significant difference (LSD) test ($P<0.05$).

Results and Discussion

Agronomic traits

Analysis of variance showed that salt stress had a significant effect on spikelet number per spike, seed weight, seed number, shoot dry weight, grain yield, leaf area, chlorophyll and greenness of wheat genotypes (Table

2). Slicing of the significant interaction effects of salinity and genotypes showed that the genotypes respond differently to salt stress in seed number, yield, stomatal conductance and SPAD number (Table 2). Analysis of variance of the shoot dry weight, seed number, grain yield, stomatal conductance and leaf area in the second experiment were in agreement with the results of the first experiment (data not shown). On the basis of mean comparison, yield production was more affected in the more productive genotypes. No. 49 genotype had the least affected yield between them (Table 3). Grain yield of No. 49 was reduced by an average of 37% while it was reduced by an average of 79% for other genotypes. Similarly, Asgari (2012) reported that at salinity 16 dSm^{-1}, the grain yield was reduced by an average of 40% for Koohdasht as a tolerant variety and 62% for Tajan as a sensitive one.

Asgari (2012) concluded that spikelet number per spike had a positive significant correlation with grain yield under salt stress. Although in this study there was no significant interaction for spikelet number per spike under severe salinity, we found that seed number per spike had a significant variation between genotypes and positively correlated with grain yield under salt stress (Table 6). The decrease of seed number under salinity changed the sink-source relationship and led to the increase of seed weight but this increase could not compensate the decrease of seed number under severe salinity, thus a negative correlation was observed between seed weight and yield (Table 6).

Shoot dry weight was significantly different between genotypes. The highest shoot dry weight was belonged to genotype No. 49 and the lowest weight was observed in genotypes C_{15} and C_{169} (Table 3). Munns (2006) found that after 40 days of stress at EC of 15 dSm^{-1}, osmotic and ionic effects caused 75% and 20% biomass loss in wheat respectively.

Leaf area

There was a significant difference between genotypes for leaf area under stress and control condition (Table 2). No. 49 had the highest leaf area between genotypes under saline stress. It is presumed that high levels of Na^+ in leaf blades would enhance premature senescence of old leaves and inhibit photosynthetic performance of younger leaves (Benderradji et al., 2011). In salt-sensitive genotypes, accumulation of salt to toxic levels in photosynthesizing leaves causes them to fall and in tolerant ones decreases the leaf area (Munns et al., 2006).

Table 2. Analysis of variance for spikelet number per spike, 100-seed weight, seed number, shoot dry weight, yield, stomatal conductance (SC.), RWC, osmotic potential (OP), leaf area (LA), SPAD number and chlorophyll (Chl) attributes in salinity experiment.

		Sum of Square										
SOV	df	Spikelet No.	Seed w.	Seed No.	Shoot dw.	Grain Yield	SC.	RWC	OP	LA	SPAD No.	chl
Salinity (S)	1	331.5^{***}	7.6^{***}	2347.5^{**}	2.32^{***}	14041^{**}	19616^{**}	0.28^{**}	17.6^{***}	1176^{***}	125^{***}	2.3^{*}
Genotype (G)	6	137.9^{***}	8.1^{***}	1285.9^{*}	0.31^{***}	4341^{***}	12811^{*}	0.01^{ns}	0.77^{ns}	87.4^{**}	104^{***}	30.9^{***}
S*G	6	10.5^{ns}	9.4^{***}	723.8^{**}	0.16^{ns}	4608^{***}	7585^{**}	0.01^{ns}	0.36^{ns}	22.6^{ns}	25^{*}	5.2^{ns}
Error	28	30	4.4	897.3	0.443	106	1624	0.097	3.32	85.5	39	11.8
		Slicing of interaction: sum of square of G levels in each level of S										
Control	6		16.5^{***}	1931.2^{***}		8813^{***}	19197^{**}				33.4^{**}	
Salinity	6		0.97^{ns}	78.5^{*}		136^{***}	1170.8^{*}				95.6^{*}	

Table 3. Mean values of shoot dry weight (g), seed number, yield (g per plant), leaf area (cm^{-2}), SPAD number and OP (MPa) at EC=0.5 and 18 dS m^{-1}.

Genotype	Shoot Dry W.		Seed no.		Grain Yield		LA		SPAD No.		OP	
	N	S	N	S	N	S	N	S	N	S	N	S
No.14	1.11^{c}	0.35^{cd}	27.3^{d}	11^{b}	0.51^{d}	0.16^{bc}	19.2^{ab}	7.2^{bc}	37.3^{d}	38.6^{d}	-1.6^{b}	-2.9^{ab}
No.49	1.25^{b}	0.49^{a}	26.3^{d}	12^{b}	0.43^{e}	0.27^{a}	22.5^{a}	9.6^{a}	39.9^{bc}	46.2^{a}	-1.6^{b}	-2.7^{a}
C_4	1.38^{a}	0.42^{ab}	31.7^{c}	7.7^{d}	0.88^{a}	0.08^{c}	16.4^{bc}	6.6^{c}	40.1^{ab}	43.6^{bc}	-1.8^{c}	-3.4^{c}
C_6	1.22^{b}	0.42^{ab}	31.3^{c}	9.0^{c}	0.79^{b}	0.12^{bc}	17.6^{b}	6.6^{c}	41.3^{a}	43.0^{c}	-1.4^{a}	-2.9^{ab}
C_{15}	1.05^{d}	0.32^{d}	35.6^{ab}	10.3^{bc}	0.70^{c}	0.11^{c}	15.7^{c}	7.4^{bc}	40.6^{ab}	43.8^{bc}	-1.6^{b}	-3.0^{b}
C_{168}	1.12^{c}	0.37^{c}	36.7^{a}	16.5^{a}	0.73^{c}	0.19^{b}	17.7^{b}	7.4^{bc}	40.2^{ab}	44.2^{b}	-1.7^{bc}	-2.8^{a}
C_{169}	1.08^{c}	0.40^{bc}	34.3^{b}	11^{b}	0.74^{c}	0.18^{b}	17.4^{b}	8.1^{b}	38.5^{cd}	42.5^{c}	-1.6^{b}	-2.7^{a}

Means within a column followed by the same letter are not significantly different (P=0.05) according to LSD test.

Na^+ and K^+ content and Na^+/K^+ ratio

The sodium and potassium ions content in the flag leaf and spike varied significantly among the genotypes and a significant genotype × salinity interaction showed that the genotypes acted differently in sodium and potassium absorption under salt stress (Table 4). In the second experiment the same result for Na^+, K^+ content and their ratio was obtained except that there was no significant difference between genotypes for flag leaf K^+ content (data not shown). The highest sodium content of leaf was observed in C_{168} and C_{169} and the lowest one was observed in No. 14. There was a significant negative correlation between leaf Na^+ and grain yield (Table 6). As Benderradji (2011) mentioned, in salt-sensitive genotypes of wheat, sodium was less effectively excluded from the transpiration stream as it entered the leaf blade, so resulting in a higher sodium accumulation (Benderradji et al., 2011). The control of Na^+ exclusion from the xylem and thereby from the leaves by *Nax1* and *Nax2* loci lead to more leaf longevity and continuous photosynthesis under severe salinity (James et al., 2012).

The highest K^+ content of leaf belonged to genotypes No. 14, No. 49 and C_{15} and the lowest one to C_4. The leaf potassium content has been suggested as a weak index of salt tolerance compared to sodium content under field conditions (Elhendavi et al., 2009).

Table 4. Analysis of variance for Na^+, K^+ and Na^+/K^+ of leaf and spike in salinity experiment.

		Sum of Square					
SOV	df	Leaf Na^+	Leaf K^+	Leaf Na^+/K^+	Spike Na^+	Spike K^+	Spike Na^+/K^+
Salinity(S)	1	4626^{***}	734^{***}	0.72^{**}	3412.1^{***}	97.5^{ns}	2.41^{***}
Genotype(G)	6	3103^{***}	2495.8^{***}	0.452^{***}	6266.4^{***}	1302.4^{***}	3.66^{***}
S*G	6	851^{*}	2964.5^{***}	0.304^{***}	5265.5^{***}	169.9^{ns}	3.17^{***}
Error	28	1331	5196.4	0.137	641.5	751	0.39
Slicing of interaction: sum of square of G levels in each level of S							
Control	6	606.5^{ns}	1091.1^{**}	0.045^{ns}	80.5^{ns}		0.33^{**}
Salinity	6	3347.7^{***}	4369.2^{***}	0.711^{***}	11451^{***}		6.5^{***}

Wheat genotypes were different with regard to leaf and spike K^+/ Na^+ ratio (Table 5). The least leaf Na^+/K^+ ratio was observed in genotype No. 14 and the least spike Na^+/K^+ ratio was belonged to genotypes No. 14 and No. 49 (Table 5). The highest sodium content and the highest ratio of Na^+/K^+ in spike were found in C_4 and C_6 (Table 5). There was a significant negative correlation between leaf Na^+/K^+ ratio and seed number and a significant negative correlation between spike Na^+/K^+ ratio and seed number, spikelet number and yield (Table 6).

Table 5. Mean value of evaluated Na^+, K^+ and Na^+/K^+ of leaf and spike (mgg^{-1}) at EC=0.5 and 18 dS m^{-1}.

Genotype	Leaf Na^+		Leaf Ka^+		Leaf Na^+/K^+		Spike Na^+		Spike K^+		Spike Na^+/K^+	
	N	S	N	S	N	S	N	S	N	S	N	S
No.14	29.5^{d}	43^{d}	105.1^{c}	124.3^{a}	0.28^{e}	0.35^{d}	7.9^{e}	9.7^{e}	26^{b}	24.5^{d}	0.30^{e}	0.40^{e}
No.49	36.6^{c}	55.8^{b}	111.5^{b}	100^{c}	0.33^{b}	0.56^{bc}	9.1^{bc}	14^{d}	34.5^{a}	34.4^{b}	0.27^{cd}	0.41^{e}
C_4	35.7^{c}	47^{c}	95.5^{e}	81.4^{e}	0.37^{ab}	0.58^{b}	10.9^{b}	41^{a}	34.5^{a}	39.6^{a}	0.32^{c}	2.1^{a}
C_6	28.3^{d}	51.5^{c}	100.8^{d}	89.7^{d}	0.28^{e}	0.57^{b}	14^{a}	31^{b}	24.3^{bc}	32.2^{bc}	0.59^{a}	1.2^{b}
C_{15}	40.8^{b}	57^{b}	100.6^{d}	112.7^{b}	0.41^{a}	0.51^{c}	8.9^{bc}	19.2^{e}	21.3^{c}	30.5^{e}	0.42^{b}	0.63^{d}
C_{168}	34.1^{c}	57.4^{b}	108.9^{bc}	93.5^{cd}	0.31^{bc}	0.61^{b}	8.1^{c}	18.6^{e}	31.5^{a}	30.7^{e}	0.25^{d}	0.61^{d}
C_{169}	44.6^{a}	85^{a}	118.5^{a}	84.6^{e}	0.38^{ab}	1.00^{a}	10.1^{bc}	14.2^{d}	18.5^{c}	20^{d}	0.54^{a}	0.70^{c}

Means within a column followed by the same letter are not significantly different (P=0.05) according to LSD test.

Table 6. Correlation coefficients between SPAD number, stomatal conductance (SC), shoot dry weight (shoot dw), leaf area (LA), RWC, osmotic potential (OP), leaf Na^+, K^+ and Na^+/K^+, spike Na^+/K^+ (SNa^+/K^+), seed number (Seed N), seed weight (Seed W) and yield.

	SPAD	SC	Shoot dw	LA	RWC	OP	Na^+	K^+	Na^+/K^+	SNa^+/K^+	Seed N	Seed W
SC	-0.339*											
Shoot dw	-0.504**	0.555**										
LA	-0.619**	0.593**	0.829**									
RWC	-0.441**	0.574**	0.685**	0.721**								
OP	0.179^{ns}	-0.281^{ns}	-0.246^{ns}	-0.402**	-0.449**							
Na^+	0.398**	-0.555**	-0.647**	-0.675**	-0.679**	0.430**						
K^+	-0.345*	0.109^{ns}	0.155^{ns}	0.276^{ns}	0.131^{ns}	-0.334*	-0.203^{ns}					
Na^+/K^+	0.135^{ns}	-0.211^{ns}	-0.142^{ns}	-0.295^{ns}	-0.278^{ns}	0.592**	0.477**	0.329*				
SNa^+/K^+	0.079^{ns}	0.115^{ns}	0.386^{ns}	0.488*	0.115^{ns}	0.013^{ns}	0.220^{ns}	0.162^{ns}	0.227^{ns}			
Seed N	-0.509**	0.688**	0.404**	0.597**	0.524**	-0.364*	-0.455**	0.402**	-0.3*	0.597**		
Seed W	0.31*	-0.75**	0.023^{ns}	0.7**	0.46**	-0.36*	0.61**	0.56**	-0.31*	0.230^{ns}	0.21^{ns}	
Yield	-0.402**	0.705**	0.465**	0.614**	0.563**	-0.309*	-0.483**	0.215^{ns}	-0.079^{ns}	0.581**	0.769**	-0.35*

^{ns} non-significant.
* differences significant at P<0.05.
** differences significant at P<0.01.

The existence of genetic diversity for the traits in wheat genotypes is in agreement with the results obtained by Goudarzi and Pakniyat (2008), who reported variation of these traits in Iranian wheat varieties in response to salt stress.

Stomatal conductance

There was a significant difference between genotypes for stomatal conductance (Table 2). The highest stomatal conductance was recorded in genotype C_{168} but the least decrease under salt stress was observed in genotype No. 49 (Figure 1). There was a significant correlation between conductance and shoot dry weight (Table 7). According to Rahnama (2010), there was a genotypic variation in stomatal conductance among wheat varieties under salinity. They also found a positive relationship between stomatal conductance and relative growth rate under salt stress (James et al., 2008; Rahnama et al., 2010) because higher stomatal conductance followed by higher CO_2 assimilation rate at salinity (James et al., 2008).

Higher stomatal conductance allows photosynthesis to be continued under salt stress but at salinity higher than 15 $dS.m^{-1}$, stomatal closure in both sensitive and tolerant genotypes has a strong impact on current photosynthesis (James et al., 2002).

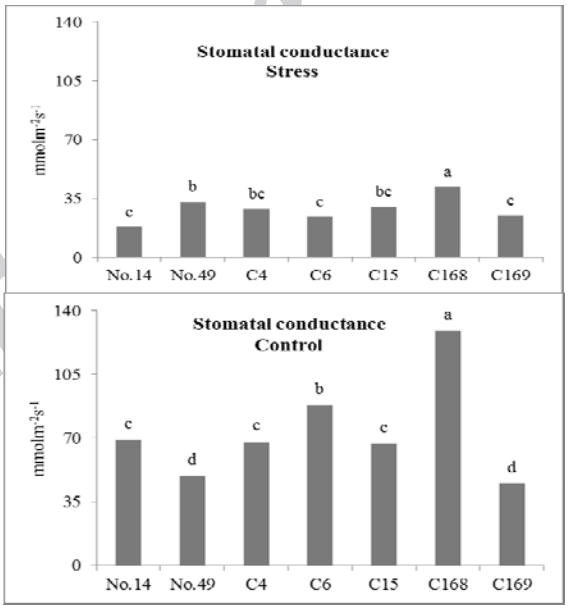

Figure 1. The levels of stomatal conductance of studied wheat genotypes under control and salt stress at EC_w=18 ds m^{-1} ($P<0.05$).

Relative water content (RWC)

There was a remarkable difference between stress and control treatments but there was no difference among genotypes (Table 2). Other studies in wheat and barley in a range of saline solutions have shown that turgor was unchanged but RWC decreased under salinity (Rivelli et al., 2002). This was because of an abnormal water absorption in osmoregulated leaves that were floated on distilled water, so a leakage of cytoplasmic solution into the appoplast happens. Thus, osmotic adjustment changes the relationship between turgor and RWC. Use of rehydrating condition for the whole plant is an offered method by Munns (2006) to study the leaf water status in a way that plant is placed in a dark and humidified atmosphere while its roots are in saline soil.

Osmotic potential

A significant difference was observed between stress and control conditions for osmotic potential but there was not any difference between the genotypes (Table 2). Increasing osmotic potential under salt stress can be due to high ion absorption and compartmentation of them in vacuole or the presence of osmolytes produced because of osmotic adjustment. Increase of osmotic potential in sensitive plants is due to decrease in turgor (Perida and Dus, 2005).

SPAD Number

Measuring greenness index using SPAD showed a significant difference between salt and control treatments (Table 2). The highest greenness was belonged to the genotype No. 49 and the least observed in the genotype No. 14 (Table 3). A positive and significant correlation was observed between chlorophyll amount and SPAD No. in both experiments (0.77 and 0.91, $P<0.01$). In sensitive genotypes the great chlorophyll destruction and photochemical malfunctions especially in photosystem II cause to a rapid decline of leaves (Zheng et al., 2009). A negative correlation between SPAD No. and leaf area (Table 6) was presumably due to the higher number of chloroplast per leaf area unit and the increase of chlorophyll density in smaller leaves. Increase in chlorophyll content under salt stress has already been reported (Akbari et al., 2012).

Genotype grouping on the basis of SST and SSI

In the first experiment, results from calculated SSI and SST showed that the genotype No. 49 had the highest SST and lowest SSI under salt stress (Table 7). The results revealed that from drought-tolerant genotypes only C_4 and C_6 were salt-sensitive and semi-sensitive respectively (SSI of 1.15, 1.08 and STI of 0.1 and 0.14 respectively). For other genotypes there was an association between their drought and salt tolerance (Table 7). Genotype No. 49 appeared salt-tolerant, No. 14, C_{168} and C_{169} were identified as semi-salt-tolerant and C_{15} identified as semi-salt-sensitive genotypes. Other studies on salt stress effect on wheat growth showed that Koohdasht and Atrak as semi-drought-tolerant genotypes were salt-tolerant but Tajan and Rasoul as drought-sensitive genotypes had low grain yield under salinity (Asgari et al., 2012).

Table 7. Average of evaluated SSI and SST at EC=18 $dS.m^{-1}$ in seven wheat genotypes.

genotype	SSI	SST
NO.14	0.82	0.34
NO.49	0.47	0.63
C_4	1.15	0.10
C_6	1.08	0.14
C_{15}	1.06	0.15
C_{168}	0.94	0.25
C_{169}	0.95	0.24

In the second experiment, grouping of the genotypes based on SST and SSI (Table 9) were in agreement with the results of the first experiment. The genotypes Bam and No. 49 were salt-tolerant, No. 14 was semi-salt-tolerant and Ghods was salt-sensitive. They were classified in salt-tolerance order according to SST and SSI as Bam > No. 49 > No. 14 > Ghods.

Table 9. Average of evaluated SSI and SST at EC=15 $dS.m^{-1}$ in four wheat genotypes.

genotype	SSI	SST
NO.14	1.04	0.20
NO.49	0.98	0.25
Bam	0.79	0.40
Ghods	1.20	0.12

Conclusion

Salt tolerance of wheat was associated with a low speed of sodium transport to shoot, a more efficient system for selective uptake of K^+ over Na^+ and maintenance of leaf greenness and stomatal conductance (Zeng et al., 2003). Therefore the salt tolerance of No. 49 might be due to the lower Na^+ content in leaf and spike, maintenance of leaf area, leaf greenness and shoot dry weight and less affected stomatal conductance leading to a higher grain yield. These traits have remarkable variations between seven genotypes, thus can be helpful as candidate traits for next studies at greater population of drought-tolerant genotypes for salt tolerance at different environments. Considering the reproducible results for salt tolerance of No. 49, it can be introduced as a candidate genotype to the plant breeders.

Altogether, drought tolerance does not necessarily lead to salt tolerance. At the first phase of salt stress, drought-tolerant genotypes have common mechanisms against osmotic stress. However it seems that different drought-tolerant genotypes may have various mechanisms against ionic stress under severe salinity leading to different degrees of salt tolerance.

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