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Effect of saline stress on root system of different wheat genotypes

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ABSTRACT
Overview of root system may clarify some aspects of breeding crops under saline stress condition. In an experiment studied 8 genotypes of wheat (spring cultivars) consist of Quds, Shiraz, Sorkh-Tokhm, Sistani, Mahooti, Falat, Bam and Arg at 3 levels of salinity stress with six replications. The present work, conducted in green-house of Zanjan University in the process of constant condition (T=27±2 °C, L:D= 16:8, RH=60±5 %) in 2011. Variation of few root attributes such as, Shoot Dry Weight (SDW), Root Dry Weight (RDW), the Ratio of Root Dry Weight /Shoot Dry Weight (R/S), Root Area (RA), Root Volume (RV), Root Area Per Volume (RAPV), Root Density (RD), the Average of Root Diameter (ARD), Root Area Index (RAI), Special Root Area (SRA), Root Area Density (RAD), Root Volume Ratio (RVR), and Specific Root Mass (SRM) evaluated before pollination step. The results showed that Sistani and Sorkh-tokhm Genotype Showed Highest resistance in compare to other cultivars. Therefore, selection and breeding traits on wheat would be possible to find the more consequential types for further crop products.

Introduction

Wheat (*Triticum aestivum* L.) is one of the most important agricultural products all over the world. It provides around 20% of main food human beings as a valuable nutrients (Salunkhe et al. 1985). One of the most consequential problems affecting on the yield of crop plants is salt restore in the soil (Heidari et al. 2007). Thus, identification of wheat with high salinity-tolerance would be acceptable to find better cultivars in versus of this aspect which improve the quality of soil in order to decrease the salinity effects and increase the production in saline condition. In agreement with genetic diversity of plants can be used in breeding programs designed in the issue of salt tolerance, when the appropriate characteristics are selected (Kingsbury and Epstain 1986). Root plant system plays the real role of developed feature in front of soil salt (Oyanagi et al. 1991). Ganjali et al (2007) reported that root developed system (root number, root length, and growth speed) has straight correlation with saline tolerance in different varieties and plants. Rafigh et al (2006) tested the morphologic parameters concerned to wheat germination in stress condition (fresh and dry weight of buds, dry weight of root and shoot, root and shoot length, and their number in normal and stress conditions). The results showed that fresh weight of bud, dry weight of shoot and roots, root and shoot decrease in salt stress.

There is evidence that root dry weight decreases as well as shoots for the sake of negative effects of salt stress (Zadoorian et al 2011). An internationally important plant breeding program was conducted largely by selection of above-ground organs. Wheat roots were rarely studied by breeders in national programs (Waines, and Ehdaie 2007) in fact, the breeding of root system has been delayed rather than shoot, and using the root characters as a selection index for indirect breeding of yield in the stress condition resulted in less developments (Seifeldin et al. 2003, O'Toole and Bland 1987). As the breeding and best selection has caused the increase of shoot yield and it can increase the yield of root and guarantee the success of critical stress conditions (Ganjali et al. 2007). This study aims to recognize some of the best genotypes of wheat with valuable traits in saline stress.

Materials and methods

The experiments were carried out in greenhouse of agricultural college Zanjan University. Eight genotypes Arg, Bam, Falat, Mahooti, Sistani, Sorkh-Tokhm genotypes (salt-tolerant genotypes) and Shiraz and Quds genotypes (salt-sensitive genotypes) selected this study. Three levels of salt stress (0, 0.5 and 1 gram of NaCl per kg of soil) used in the mixture by soil. The experimental units were plastic pots (vases) with the same diameter (10 cm), height=30 cm and capacity= 1700 grams (of soil). Field capacity was determined 23 and water for each pot was 80% of field capacity so that watering was conducted every other day. The present work surveyed parameters of Shoot Dry Weight (SDW), Root Dry Weight (RDW), the Ratio of Root Dry Weight/Shoot Dry Weight (R/S), Root Area (RA), Root Volume (RV), Root Area Per Volume (RAPV), Root Density (RD), the Average of Root Diameter (ARD), Root Area Index (RAI), Special Root Area (SRA), Root Area Density (RAD), Root Volume Ratio (RVR), and Specific Root Mass (SRM). Up to the initiation of pollination, the pots were broken before pollination (65 days after cropping) and divided in 2 parts: shoots and roots. Washing roots was performed in 2 stages: firstly, were washed by root washing machine then, washed carefully by hand to remove any exterior material on their surface. Thereafter, roots volumes were calculated by subtraction of primary volume of remained (existed) water before after immersing them in scalar cylinder. Then gained data were noted down.

In order to make a close contrast which is essential for the operation of scanner and software- the mixture of water and dyeing black color was put into the plate of each sample. As a result, the root completely became black after 24 hours. Water and alcohol were mixed with the rate 4:1 (water=4, alcohol=1) to preserve (keep) the root until analyzing and put it into the plastic bags containing each sample. The root were put into the grass tray and then were took photographed by canon scanner, model LIDE 600E with the clarity 4800*960 dpi. The photos were analyzed by scanner and with GSA Image Analyzer. In order to measure the dry weight of sample, each one was put in the oven (temperature=75 c) for 24 hours (Zhao et al. 2005) and dry weight of root was measured by digital scale with the accuracy=0.0001 in each pot, dry weight of shoot was calculated in gram unit. Data variance analysis was done by SAS software and comparison of data mean was carried out by Dancan's multiple-range test.

Results

According to the results of variance analysis the effect of salt stress is the significant for the most of root characters. It denotes that at least one level of salt stress has had a significant effect on the most of characters. More ever, in the most of them, there is a significant difference, at least, between 2 genotypes in the view to measured characters.

Root Volumep

The rate of root volume decreased in 0.5 gr and 1 gr treatment comparing to the control (treatment) was observed about 16% and 25%, relatively. With a view to the calculated statistics (the statistical data) a significant difference was observed between the mean of root volume of tolerant and sensitive genotypes. Sistani genotype had the most volume of root (weather in severe stress or in temperate stress) which had significant difference with 2 sensitive genotypes, I e, Shiraz and Quds. The ratio of the highest root volume in Sistani genotype to Quds genotype was 4.9 times. declining rate of the assessed characters was obvious in different genotypes. So that, it was significant in 1 gr of salinity level, rather than control. The Sistani and Quds genotypes had the most rate of this character (relatively, 6.96 and 5.59 $cm^{3)}$ and the most tolerant genotypes had significant different with Quds and Shiraz genotypes (1.98 and 1.42 cm^3, relatively). Due to different characters of root such as volume, the variation of different plant varieties declares their difference in salt stress resistance. So that, these differences can be used for the selection of genotypes and genotypes (Gregory 1988). These characteristics are more in tolerant genotypes, so it can denote their more ability to absorb water and nutrients and to keep swelling in the stress condition that is essential for plant to grow better.

Root Area

The studies showed that there is a significant difference between genotypes and different salt treatments with a view to the root area.

In this research the descending process of characters variations in different studied genotypes is clearly obvious. The biggest size of root area is related to the tolerant genotypes, i.e, Sistani and Mahooti and Arg and the least belonged to the sensitive genotypes, i.e, Quds and Shiraz. So that, this decrease was significant in salinity levels equal to 1 gr in comparison with control the surface of root system of plants is an important and effective parameter to determine the rate of water and nutrients absorption, too. In salt stress, the decrease rate of root area in Sistani and Quds genotypes was 3% and 1% relatively. A significant different between the root surface mean of tolerant and sensitive genotypes was observed. So that, its ratio was 1.09 and 1.08, relatively in Sistani, Shiraz and Arg/ Shiraz, and it were 1.15 and 1.14 relatively, in Sistani/Quds and Arg/Quds. Probably, the reason of root surface decrease –that is caused by stress- in the verified genotypes, may be related to roots remove (rotting).

Root Dry Weight

The effect of salt stress on the root dry weight is significant at the p=1%. Due to the gained results, it can be expressed that Sistani and Arg genotypes have more ability for photosynthesis. This is because of their priority in some characteristics like the root and the shoot dry weight. These genotypes could have more dry weight in different salt treatments. The Sorkh-Tokhm genotype had an intensive decline in both root and dry weight- under temprature stress condition. But these weights increased may be the gained resistance by mentioned genotype, after undergoing the stress. So that it became able to rebuilding itself. Perhaps, the decrease of root dry weight in high salinities can be relate to the allocation of produced carbon to the other metabolic ways which are effective in stress tolerance. As reported, the leaf development would cease at the beginning of salt stress. While, carbon absorption remains approximate to the normal amount, as well. The extra produced carbon may be stored and be used for osmotic regulation or it can be allocated to the root growth (Emam and Zavvareh 2005). Shiraz and Quds genotypes had the least root dry weight and its most amounts belonged to Sistani genotype. The ratio of root dry weight of Sistani genotype to Shiraz and Quds was observed about 4.4 and 4.3 relatively.

Shoot Dry Weight

A significant different was observed between different genotypes and different level of salinity, in this character. Decrease of this trait (29%) was observed further to the increase of salinity level. So that, the least amount of salinity level=1 gram of salt and among the genotypes was observed in Shiraz genotype. Some of the common reasons of the decrease of biomass in these conditions include the decrease of water absorption by plant and the toxic effects of Na+ and Cl- caused by their high concentrations in soil solution (Aslam and Muhammed 1972). According to the earned results, Arg, Sistani and Sorkh-Tokhm genotypes had the most shoot dry weight.

Table 1. Analysis of variation for studied traits.

S.O.V.	df	RV	RA	RDW	SDW	R/S	ARD	RD
salinity	2	*9.96	ns 0.03	**0.017	**0.42	ns 0.001	* 2.03	ns 0.00004
genotype	7	**32.98	**0.09	**0.053	**1.02	ns 0.005	** 5.06	*0.0001
salinity× genotype	14	ns 1.68	ns 0.01	ns 0.002	ns 0.04	ns 0.002	ns 0.354	ns 0.00005
Error	48	2.20	0.01	0.002	0.04	0.005	0.790	0.00007
C.V		16.12	4.76	16.44	14.43	20.50	18.6	12.27

*,** show significant effect at 5 and 1% statistically levels, respectively and ns shows no significant effect

RV: Root Volume, RA: Root Area, RDW: Root Dry Weight, SDW: Shoot Dry Weight, R/S: Root Dry Weight /Shoot Dry Weight, ARD: Average Root Diameter, RD: Root Density

And the least rate belonged to Shiraz and Quds genotypes. The highest mean rate (Sorkh-Tokhm) to the least mean rate (Shiraz) was observed about 4.78 folds. As these genotypes are superior to others in the characters like root volume, so, it is logical to have more shoot dry weight. Probably, the less rate of shoot dry weight in sensitive genotypes is because of the allocation of plant extra-cellular energy. Consumption to osmotic regulation under stress condition and further more consumption of ATP (Wyn and Gorham 1993). It is supposed that the light rate and so the photosynthesis (assimilation) rate and dry material accumulation decrease due to the decrease of leaf area and number, when salinity increases. More ever the dry weight of aerial part which is the sum of leaf and stem dry weight would decrease. The rate of shoot dry weight determines grain yield, as its one of the growth index (Romero et al. 2001). Thus the genotypes having more rates of this character can be supposed as the genotypes having more yield under stress condition.

Root Dry Weight /Shoot Dry Weight

The most ratio of root/ shot dry weight belonged to Shiraz genotype, and the least ratio belonged to Sorkh-Tokhm genotype and its rate was 1.6 time (folds) of Sorkh-Tokhm's genotype but other genotypes did not have any significant difference with a view to this character. The comparison of the means of this ratio in 8 genotypes of wheat in different rates and conditions of salt stress, showed that the earned ratios were approximately equal in the most genotypes, expect sensitive genotypes. But it was more in severe stress than temperate stress in sensitive genotypes, I e, Quds and Shiraz. It means that according to the reports the ratio of root/shoot dry weight is more in severe stress in comparison with temperate stress.

Average Root Diameter

The results of the study of average root diameter denoted that Sistani genotype had the most average root diameter and Shiraz and Quds genotypes had the least. The mean of this character in Sistani genotype was about 3.84 times (folds) in comparison with Quds genotype: In all, 0.5 gr and 1 gr of salinity caused the decrease of this character up to 15% and 27%, relatively.

Decrease of the volume and the diameter of root in saline condition may be because of less availability of photosynthates in shoot and water (osmotic) stress and the toxicity of ions because of salt accumulation (existence) around the roots. The comparison of the mean of average root diameter in wheat genotypes under different rates of salt stress showed that the increase of stress caused the decrease of average root diameter, significantly.

Root Density

Root density was calculated by the ratio of root dry weight/root volume. There was no significant difference between wheat genotypes. In the comparison of mean root density of wheat genotypes in different rates of salt stress, the root density of different genotype in stress levels did not fallow a fixed (constant) pattern. The most amount of root density was in normal condition for Bam genotype, in temperate stress for Arg genotype and in severe stress for Sorkh-Tokhm genotype. The most rate of root density mean belonged to Bam genotype and the least rate belonged to the Shiraz. The ratio of Bam/ Shiraz root density mean was equal to 1.52.

Discussion

Although several studies have denoted that root characters are genetical (Sexena., 2003), but there are many studied which emphasize that these characters are also affected by environmental parameters intensively and severly (Gregory., 1988). The researches have shown that there is genetic diversity in root character between wheat germplasms (Simane et al. 1993). A significant difference was observed in root characters of genotypes, too. This denotes that it is possible to select and breed these characters in order to make and produce salt stress tolerant lines in wheat. That obtained results of this experiment showed that the different levels of salinity caused the presence of negative effects on all of the characters which are effective on wheat growth. The most rate of characters decrease was observed in the salinity equal to per gram of salt per kilogram of soil.

Table2. Means comparison of studied traits in response to treatments.

		RV		RA (mm2)		RDW		SDW (g)		R/S		ARD (mm)		RD	
Arg	S1	7.5	ab	25000	a	0.26	a	1.2	ab	0.22	ab	0.001	a-e	0.035	abc
	S2	4.8	b-f	21000	a-d	0.17	a-f	1.1	bc	0.16	ab	0.0008	a-g	0.04	abc
	S3	4.2	c-g	18000	b-e	0.11	e-h	0.7	b-e	0.15	ab	0.0006	a-g	0.033	abc
Bam	S1	6.2	a-d	20000	a-e	0.22	abc	1.1	bc	0.02	ab	0.0011	abc	0.045	a
	S2	5.5	a-e	19000	a-e	0.16	b-f	0.9	bcd	0.02	ab	0.0008	a-g	0.033	abc
	S3	4.5	c-f	18000	b-e	0.13	c-g	0.7	cde	0.19	ab	0.0007	a-g	0.035	abc
Falat	S1	5.5	a-e	10000	e-f	0.13	c-g	0.7	cde	0.18	ab	0.0008	a-g	0.025	bc
	S2	3	e-h	17500	b-e	0.12	d-h	0.65	c-f	0.16	ab	0.0006	a-g	0.033	abc
	S3	3	e-h	17000	b-f	0.09	fgh	0.5	e-g	0.16	ab	0.0005	b-g	0.033	abc
Mahooti	S1	5.8	a-e	24000	abc	0.2	a-d	1.1	abc	0.17	ab	0.0009	a-g	0.035	abc
	S2	6	a-e	16000	c-e	0.16	a-f	0.9	bcd	0.17	ab	0.0012	a	0.035	abc
	S3	5.2	a-e	17000	b-e	0.15	c-f	0.7	cde	0.18	ab	0.00095	a-f	0.025	bc
Sistani	S1	7	abc	24000	abc	0.25	ab	1.1	abbc	0.23	ab	0.0011	abc	0.03	abc
	S2	7.8	a	23000	a-d	0.25	ab	0.9	bc	0.24	ab	0.0012	a	0.035	abc
	S3	6	a-e	22500	a-d	0.16	a-f	0.8	bcd	0.2	ab	0.0008	a-g	0.035	abc
Sorkh-tokhm	S1	5.8	a-e	19000	a-e	0.17	a-e	1.5	a	0.14	b	0.001	a-d	0.028	bc
	S2	4	d-h	15000	def	0.11	d-h	0.8	bcd	0.14	b	0.0008	a-g	0.035	abc
	S3	4.7	a-f	25000	ab	0.16	a-f	1	bc	0.15	ab	0.0009	a-g	0.03	abc
Shiraz	S1	2	fgh	14000	ef	0.05	gh	0.2	g	0.29	ab	0.0005	c-g	0.043	ab
	S2	1.9	fgh	14500	ef	0.04	gh	0.21	fg	0.17	ab	0.0003	g	0.025	bc
	S3	1.5	gh	9000	f	0.03	h	0.12	g	0.23	a	0.0004	dg	0.025	bc
Ghods	S1	1.5	gh	14000	ef	0.05	gh	0.25	fg	0.17	ab	0.0004	dg	0.02	c
	S2	1.4	gh	15000	c-f	0.045	gh	0.3	efg	0.15	ab	0.0003	g	0.035	abc
	S3	1.3	h	13000	ef	0.04	h	0.2	g	0.2	ab	0.0003	g	0.03	abc

RV: Root Volume, RA: Root Area, RDW: Root Dry Weight, SDW: Shoot Dry Weight, R/S: Root Dry Weight /Shoot Dry Weight, ARD: Average Root Diameter, RD: Root Density

According to means comparison in studied genotypes, Shiraz and Quds allocated the least rates to themselves, and had a significant different with tolerant genotypes. In this comparison, Shiraz genotype had a lower level rather than Quds genotype. It denotes that Shiraz genotype has more susceptibility under salt stress condition comparing to the normal condition. In fact, in the one hand the study of characters like shoot dry weight, root volume and root area, root dry weight and the significant superiority of Sistani and Sorkh-Tokhm genotypes in different salinity treatments, with the view of these characters can denote their priority in the stress condition and in the other hand, it can focus on the applicability of the studied character in the evaluations which were done in the case of salt tolerance in wheat. The superiority of these genotypes was obvious in other characters. Ultimately, it can be concluded that salinity tolerance is not a function of an organ's operation or a character of plant. But it's a resultant of most of the plants characters. The genotype which has superior in most of the characters related to salinity, can be appropriate in the stress condition. According to this, it can be expressed that the more number and amount of the mentioned characters in Sistani and Sorkh-Tokhm genotypes can result in the use f these genotypes as a salinity tolerant germplasm for the separation of salinity tolerant genotypes in the breeding programs.

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