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# Salinity Induced Effects on Growth Parameters, Chemical and Biochemical Characteristics of Two Forage Sorghum (Sorghum bicolor L.) Cultivars

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Abstract: Plant production stability under saline condition compared to normal has had limited success due to lack of knowledge on salt tolerance mechanisms. To evaluate the changes in chemical and biochemical characteristics (content of free proline and total protein, Na+ and K+) and growth parameters (leaf area, plants height, total tiller number, leaf number, shoot and root weight, percent of emergence, growth rate and growth power) of sorghum under salinity stress, a pot experiment was carried out at Shiraz University, Shiraz, Iran in 2010. Four salinity levels containing 3, 6, 9, 12 ds m<sup>-1</sup> using NaCL and without using NaCL (as control) and also two sorghum cultivars consist of Pegah (Iranian cultivar) and Speedfeed were evaluated in a factorial experiment design with three replications. Results showed that salinity decreased accumulation of K+, emergence percentage and all growth parameters but increased amount of Na+, proline and total protein content in shoot and leaves of treated plants. Multiple regression, stepwise selection, selected root weight and their ratio (shoot/root), leaf area and leaf number per plant, tiller number per plant, photosynthesis rate and also content of K<sup>+</sup> as most significant factors for contributing in shoot weight. Stepwise selection and correlation coefficient cleared that root weight is the most important factor in plant shoot production under salinity condition and breeding program must be carried out to screen cultivars with higher root density in order to increase resistance to salinity. Based on the results, Speedfeed is proper cultivar to be used in breeding programs to achieve higher salinity resistance by screening or crossing programs.

Key words: Forage sorghum, proline, salinity, total protein, biochemical characteristics

### INTRODUCTION

During growth and development, plants often encounter adverse environmental conditions, including drought, salinity, extreme temperature, nutrient deficiency, pathogens and so on (Li et al., 2010). Soil salinity has been recognized as one of the most serious problems for agriculture in arid and semi-arid areas of the world. It is known that one crop may be highly tolerant at one stage of development and sensitive during another stage. Saline soils are estimated to cover about 5-10% of the world's arable land and the area affected by salinity is increasing steadily, in part due largely to mismanaged irrigation (Krishnamurthy et al., 2007). Soil salinity drastically reduces the productivity of most crops although to a varying extent across species (Munns et al., 2002). Several attempts have been made to overcome tile effects of salinity on germination and seedling development of different species such as wheat, sorghum, barley, oat, carrot and tomato.

Sorghum (Sorghum bicolor L.), an important failsafe crop in the global agroecosystem, is the fifth most important grain crop grown worldwide, which is unusually tolerant of low input levels, an essential trait for arid and semiarid areas in temperate and tropical regions. Recently, its significance is increasing as a biofuel crop for its high yield of biomass and broad adaptation to different kinds of environmental regions (Paterson et al., 2008). It's also emerging as a genetic model for tropical grasses based on its small and well-characterized genome, low level of gene duplication and close relationship to the larger and more complex genomes of maize and sugarcane (Paterson et al., 2008, 2009). Also, sorghum is a major grain and forage crop and was previously characterized as moderately tolerant to salinity (Igartua et al., 1995). It is considered relatively more salt tolerant than maize, the cereal crop ranking first in productivity globally (Maas, 1985) and so sorghum has the potential as a crop for salt affected areas (Igartua et al., 1994). The presence of large genotypic variation for tolerance to salinity reported in sorghum

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(Maiti et al., 1994) offers a good scope for integrating tolerance characteristics into appropriate breeding programs to improve crop productivity on saline soils (Krishnamurthy et al., 2007).

Efforts to enhance crop yields under salinity stress have had a limited success because available knowledge of the mechanisms of salt tolerance has not been completed and also not turned into useful selection of genotypes. Attempts have been made to evaluate salt tolerance at germination and emergence stages in grain sorghum (Igartua et al., 1994) and large genotypic differences were reported, but this early evaluation appears to have little relation with overall performance under saline conditions (Munns et al., 2002).

Though Na<sup>+</sup> exclusion and grain K<sup>+</sup>/Na<sup>+</sup> ratios have been suggested to be reliable traits for selecting salt tolerant crops (Munns and James, 2003; Munns *et al.*, 2002), the value of that trait has not been used in a large scale in sorghum (Poustini and Siosemardeh, 2004).

This study was conducted to evaluate effects of salinity on growth parameters and powers, biochemical traits and photosynthesis rate of two forage sorghum cultivars for better understanding of tolerance mechanisms to salinity.

## MATERIALS AND METHODS

The surface layer of soil (0-30 cm) was collected from Bajgah, located in the province of Fars, south of Iran (coordinated: 29°43′ N and 52°35′ W). The main characteristics of soil were: pH 7.09; electrical conductivity, 0.0513 ds m<sup>-1</sup>; total organic mater, 1.04%; Total N, 0.060 g kg<sup>-1</sup>; Available P, 12.5 µg g<sup>-1</sup> and soil water capacity, 23.27%. Based on the analysis of soil 70 mg N kg<sup>-1</sup> soil and 20 mg P kg<sup>-1</sup> soil were added to soil.

**Experimental procedures:** A factorial experiment on the bases of completely randomized design was established with two factors and three replications. The first factor was cultivar that was consisted of Pegah (Iranian cultivar) and Speedfeed and the second factor was salinity levels containing without adding NaCl to soil (as control), 3, 6, 9 and 12 ds m<sup>-1</sup> salinity.

The pot experiment was carried out at a greenhouse in Agriculture School, Shiraz University, Shiraz, Iran in 2010. The pots with 23 cm diameters, 20 cm heights were filled with 3 kg washed and sieved soil. Before planting seeds were treated with ethanol 98% for about 20 second and then three times were washed with distilled water.

Four levels of salinity were applied prior to sowing through a one-time application of deionized water with 3, 6, 9 and 12 ds m<sup>-1</sup> NaCl and without adding NaCl (as control). Ten seeds of each cultivar were sown in each

pot in equal distance from each others. Pots were weighted daily and based on decreasing the amount of water in pots, water was added to pots soils until gain to soil field capacity (23.2%, w/w).

# Plant analyses

**Growth parameters:** Leaf number at two stage, total tiller number, plants height with a ruler and leaf area of plants using following formula: Leaf area of plant = Maximum leaf heights×maximum leaf diameters×0.75 were measured. Shoot and root weight of plants were determined after their harvesting.

Emergence, growth rate and growth power: After complete germination period of plants, emergence percent based on the germinated number of seeds number per pot ratio was obtained. After calculating the emergence percent, number of remained plants per pot was countered and percent of surviving power for plants subjected to control plant numbers in emergence was calculated at three stages. For growth power, plants of each pot visually scaled from 1 (lowest growth power) to 5 (highest growth power). Results of emergence and growth rate are presented in Fig. 1.

**Photosynthesis rate:** During the vegetative stage, Photosynthesis rate was conducted on the youngest fully expanded leaf at stem elongation stage on two sorghum cultivars. Net  $CO_2$  assimilation, was assessed on intact leaves using the LiCor 6400 gas exchange system (Lincoln, NE, USA). During the reproductive stage Photosynthesis rate was measured at flowering (65-70 day after sowing) on flag leaves after 1-2 h of acclimation in a growth cabinet, under a light intensity of about  $1000 \ \mu \text{mol} \ \text{m}^{-2} \, \text{s}^{-1}$ , relative humidity of 70% and 29°C to ensure measurement under stable conditions.

Free proline content: Free proline was extracted from fresh leaves according to the method of Bates  $\it et~al.~(1973)$ . Leaves samples (0.5~g) were homogenized in 10~mL of 3%~(w/v) aqueous sulphosalicylic acid then the homogenate was filtered through Whatman No. 2 filter paper. Two milliliters of filtrate was then mixed in a test tube with 2 mL acid ninhydrin and 2 mL glacial acetic acid and incubated in a  $100^{\circ} C$  water bath for 1 h. The reaction was terminated by placing the mixture in an ice bath. It was then extracted with 4 mL toluene. The absorbance was recorded at 520 nm and the proline concentration was determined as  $(\mu g~g^{-1}~FW)$  using a standard curve.

**Total protein content:** The protein content was estimated according to the method of Bradford (1976), using Bovine Serum Albumin (BSA) as a standard and observance of 595 nm.

**Determination of Na+ and k+ content:** One hundred and fifty milli-grams of finely ground shoot sample was digested in 4 mL of concentrated sulfuric acid with 0.5% selenium powder at 360°C for 75 min on a block digester and the digest was diluted to 75 mL. Exchangeable K and Na were estimated (Sahrawat *et al.*, 2002) using an atomic absorption spectrophotometer (Varion model 1200, Australia).

**Statistical analysis:** Statistical Analysis Software (SAS 9.1) was applied for all analysis containing, two ways ANOVA, mean comparison, Pearson correlation and multiple regressions. Mean comparison was carried out based on Least Significant Difference (LSD) and multiple regression was based on stepwise selection.

# RESULTS

**Photosynthesis rate:** Analysis of variance showed statistical significant difference between cultivars and among salinity levels (p<0.01). Photosynthesis Rate (PR) in Pegah cultivar (19.22) was higher than Speedfeed

(18.55). Salinity decreased the photosynthesis rate of plants. Highest (PR) was measured for control (34.24) and lowest PR was observed for 9 salinity levels and there was no significant difference between 6 and 9 levels. Fitted regression lines for two cultivars showed that the most variation of data ( $R^2 = 0.97$  and  $R^2 = 0.94$ , respectively) could interpret by these lines (Fig. 3).

Chemical and biochemical parameters: Free proline content (Pr): measuring free proline content showed no significant difference between cultivars and salinity×cultivar interaction but difference among salinity levels was significant (p<0.01). Highest amount of Pr was recorded for 9 salinity level and the lowest one was recorded for control. The difference between 3 and 6 salinity levels was not significant.

**Total protein contents:** Protein content of Pegah (8.17) was significantly lower than content of Speedfeed (9.13). In general, salinity enhanced protein concentration of treated plants but no significant different was observed

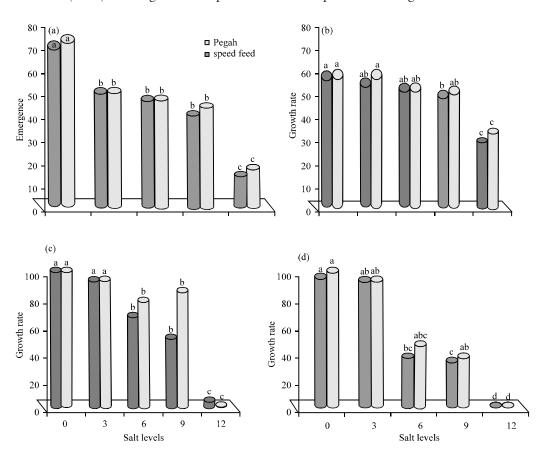


Fig. 1(a-d): Percent of emergence (a) growth rate at first (b) second (c) and third (d) measuring. Different alphabets are significantly different at p<0.01

Table 1: Analysis of variance and mean comparison for main and interaction effect of tiller number and also leaf number and height of plants at two different time of sampling

time of sampl	ıng				
Treatments	Leaf number (first)	Height (cm) (first)	Total tiller number	Leaf number (second)	Height (cm) (second)
Cultivars					
Pegah (V1)	3.86a	13.20a	1.67a	3.60a	15.19a
Speedfeed (V2)	d (V2) 3.66a		1.60a	3.33a	13.42b
Salinity					
0 (S1)	6.83a	24.75a	3.50a	6.50a	29.643a
3 (S2)	5.17b		15.08b 2.17c		17.717b
6 (S3)			1.50c	3.00c	13.252c
9 (S4)			1 d	2.67c	10.925d
12 (S5)	0.00e	0.00d	0.00e	0.00d	0.00e
Interaction					
V1S1	6.67a	24.0a	3.67a	6.33ab	29.330a
V1S2	5.33b	13.5b	2.00bc	5.33ab	15.593c
V1S3	4.33bc	9.50c	1.33cd	2.67c	11.607d
V1S4	3.00d	9.17c	1.00d	2.33c	10.570d
V1S5	0.00e	0.00d	0.00e	0.00d	0.000e
V2S1	7.00a	25.50a	3.33a	6.67a	29.957a
V2S2	5.00b	16.67b	2.33b	5.00b	19.840b
V2S3	3.67cd	14.00b	1.67b-d	3.33c	14.897c
V2S4	2.67d	9.83c	1.00d	3.00c	11.280d
V2S5	0.00e	0.00d	0.00e	0.00d	0.000e
Analysis of Variance					
Cultivar (V)	$ns\P$	*	ns	ns	*
Salt level (S)	***	***	***	ole ole	oks oks
V×S	ns	ns	ns	ns	ns

<sup>¶</sup>ns: Not significant; \*\*\*: Significant at p<0.001; \*\*: significant at p<0.01 and \*: significant at p<0.05. The means with the same letters are not significantly different (Duncan's multiple ranges test; p<0.05)

between control and 3 ds m<sup>-1</sup> level. On the other hand, content of protein in Pegah for control was higher than 3 ds m<sup>-1</sup> level of salinity.

K<sup>+</sup>, Na<sup>+</sup> and K/Na ratio: Main effects of cultivar and salinity levels showed significant difference for both Na<sup>+</sup> and K<sup>+</sup> but just salinity showed significant difference for K/Na ratio. The only significant interaction between cultivar×salinity was obtained for Na<sup>+</sup>. Pegah cultivar had higher Na<sup>+</sup> (7.35%), K<sup>+</sup> (5.2%) and K/Na (8.47%) ratio than Speedfeed. Salinity decreased amount of K<sup>+</sup> and K/Na ratio but increased amount of Na<sup>+</sup>. Highest amount of Na<sup>+</sup> (220.17 mm kg<sup>-1</sup>), K<sup>+</sup> (503.10 mm kg<sup>-1</sup>) and K/Na ratio (5.02) were measured for 9 ds m<sup>-1</sup>, control and control but the lowest ones were observed in control, 6 and 9, respectively (Table 3).

# **Growth parameters**

**Leaf Number (LN):** Measuring leaf number in two different times showed that numbers of leaves were decreased from first measuring to second measuring generally. In both time of measuring, Pegah had higher leaf number per plant than Speedfeed but the difference was not significant. Salinity decreased number of leaves per plant at both measurements. Maximum number of leaves per plant was recorded for control and the minimum number was observed in 9 ds m<sup>-1</sup> salinity level in both measurements, respectively. At first time measuring, significant difference between 6 and 9 ds m<sup>-1</sup> levels of

salinity was observed but this difference was not obtained for second measurement (Table 1).

**Plant height:** Plants height of Pegah was significantly (p<0.01) higher than Speedfeed (17.54 and 13.119% at first and second measurements, respectively). Control plants showed maximum height (24.75 and 29.64 cm respectively) and 9 ds m<sup>-1</sup> stressed plants showed minimum height (9.5 and 10.92 cm respectively). The difference between 6 and 9 ds m<sup>-1</sup> salinity levels was not significant at first measuring (Table 1).

**Total tiller number per plant:** There was no significant difference between two cultivars related to total number of tillers per plant but salinity showed significant difference. Control had maximum tiller number per plant but 9 ds m<sup>-1</sup> level of salinity had minimum number (Table 1).

Shoot weight, root weight and shoot/root ratio: Analysis of variance showed no significant difference between cultivars for shoot weight, root weight and shoot/root ratio. With increase salinity level, shoot weight and root weight was decreased. Highest shoot (3.19 g) and root (2.29 g) weight were obtained in control while the lowest ones were measured for 9 level of salinity. Minimum shoot/root ratio was observed in 9 ds m<sup>-1</sup> salinity but it had no difference with 3 ds m<sup>-1</sup> salinity level. On the other hand, maximum ratio of shoot/root was observed in

Table 2: Analysis of variance and mean comparison for main and interaction effect of shoot and root weight and their ratio

Treatments	Root weight (g)	Shoot weight (g)	Shoot/Root
Cultivars			
Pegah (V1)	0.79a	1.06a	1.22a
Speedfeed (V2)	0.73a	0.96a	1.23a
Salinity			
0 (S1)	2.2883a	3.1917a	1.37504a
3 (S2)	1.0567b	1.2783b	1.2232ab
6 (S3)	0.3683bc	0.4750c	1.30678a
9 (S4)	0.1133c	0.1233d	1.08712b
12 (85)	0.0000c	0.0000d	0.00000c
Interaction			
V1S1	2.3333a	3.1967a	1.3132ab
V1S2	0.9133bc	1.0567c	1.2261abc
V1S3	0.3300bc	0.4567d	1.3359ab
V1S4	0.1000c	0.1167d	1.1576bc
V1S5	0.0000c	0.0000f	0.0000d
V2S1	2.2433a	3.1867a	1.4369ab
V2S2	1.2000b	1.5000b	1.2206abc
V2S3	0.4067bc	0.4933d	1.2777ab
V2S4	0.1267c	0.1300d	1.0167bc
V2S5	0.0000c	0.0000f	0.0000d
Analysis of Variance			
Cultivar (V)	ns	ns	ns
Salt level (S)	***	***	***
V×S	ns	ns	ns

Table 3: Analysis of variance and mean comparison for main and interaction effect of proline, photosynthesis rat and chemical composition

Treatments	Total protein (mg g <sup>-1</sup> FW)	Proline (µg g <sup>-1</sup> )	$Na^+ (mm kg^{-1})$	K+ (mm kg-1)	K/Na
Cultivars					
Pegah (V1)	8.17b	0.35a	129.38a	368.23a	2.69a
Speedfeed (V2)	9.13a	0.24a	120.52b	350.62b	2.48b
Salinity					
0 (S1)	6.54c	0.225c	100.557d	503.097a	5.0223a
3 (S2)	6.72c	0.298b	132.083c	483.937b	3.6771b
6 (S3)	9.48b	0.321b	171.977b	434.297c	2.5313c
9 (S4)	11.85a	0.385a	220.170a	375.815d	1.7206d
12 (S5)	0	0.000d	0.0000e	0.0000e	0.0000e
Interaction					
V1S1	6.61d	0.230d	99.947e	497.23a	4.9878a
V1S2	5.85d	0.297c	135.290d	467.29b	3.4643c
V1S3	9.00cd	0.31bc	175.303c	419.30c	2.3956d
V1S4	11.22b	0.370 a	236.407a	369.30d	1.5632e
V1S5	0.00e	0.000 e	0.0000f	0.0000e	0.0000f
V2S1	6.48d	0.220d	101.167e	508.96a	5.0568a
V2S2	7.58d	0.300c	128.877d	500.59a	3.8899b
V2S3	9.96c	0.333d	168.650c	449.29b	2.6669d
V2S4	12.49ab	0.400a	203.933b	382.33d	1.8780e
V2S5	0.00e	0.0000e	0.0000f	0.0000e	0.0000 f
Analysis of Vari	ance				
Cultivar (V)	**	ns		**	ns
alt Level (S)	**	***	etc etc	***	***
V×S	ns	ns	**	ns	ns

Ins: Not significant; \*\*\*: Significant at p<0.001; \*\*: significant at p<0.01 and \*: significant at p<0.05. The means with the same letters are not significantly different (Duncan's multiple ranges test; p<0.05)

control while the difference among control, 3 and 6 salinity levels were not significant (Table 2).

**Leaf Area (LA):** Results of leaf area measuring at two times showed that cultivars had no significant difference at first time but had significant difference at second time measuring and Speedfeed showed higher leaf area (13.5%). Leaf area decreased with increasing salinity

levels and during times of measurements. Highest leaf area was observed in control while the lowest was recorded for 9 ds m<sup>-1</sup> salinity level. Regression lines separately were fitted for Pegah and Speedfeed cultivars and they could interpret most of the variations at first (Pegah;  $R^2 = 0.86$  and Speedfeed;  $R^2 = 0.83$ ) and also second (Pegah;  $R^2 = 0.85$  and Speedfeed;  $R^2 = 0.96$ ) measuring (Fig. 2).

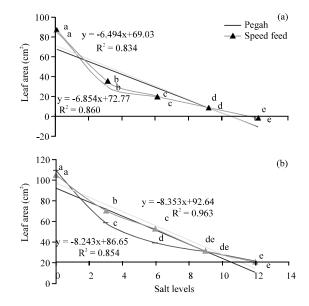


Fig. 2: Leaf area at first (a) and second (b) time measuring

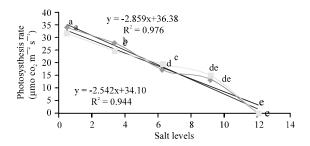


Fig. 3: Photosynthesis rate of two sorghum cultivars under different salinity levels

Table 4: Results of stepwise selection for indicating most important

variable affecting shoot weight

Step	Entered variable	Partial R2	Model R2	p-value
1	Root weight	0.9829	0.9829	< 0.0001
2	LA (Second)	0.0048	0.9877	0.0032
3	Tiller number	0.0036	0.9913	0.003
4	Sh/R	0.0009	0.9922	0.0969
5	K <sup>+</sup> content	0.0043	0.9964	< 0.0001
6	LN (First)	0.0005	0.997	0.0602
7	LA (First)	0.0005	0.9974	0.0573
8	PR	0.0003	0.9977	0.1137

LA: Leaf area, Sh/R: Shoot per root ratio, LN: Leaf numbers and PR: Photosynthesis rate

# Emergence, growth rate and growth power

**Percent of emergence:** Results showed that emergence percent of Speedfeed was more than that in Pegah but the difference was not significant. Maximum emergence (71.67%) was obtained in control plants but the minimum was observed for 12 ds m<sup>-1</sup> salinity level (15%). Among 3, 6 and 9 levels of salinity significant difference were not observed (Fig. 3).

**Growth rate:** Results of three times countering plant number after emergence and comparing with emergent percent in control showed that cultivars was not different in growth rate but salinity levels were significantly different. Growth rate in control plants was 100% at all measurements but in other salinity levels was decreased during the measurements. Salinity level of 12 ds m<sup>-1</sup> showed 52.77% growth rate at first time measurement but at second time was nearly zero and finely at last was completely zero. Significant measuring difference between control and other salinity levels except 12 ds m<sup>-1</sup> was not significant at first time measurement but significant different between control with 6 and 9 ds m<sup>-1</sup> was significant at second and third times (Fig. 3).

**Growth power:** Recording results of growth power at four times showed that there no significant different between cultivars at all recording while the differences among salinity levels were significant. Average growth power at fist time recording for 12 ds m<sup>-1</sup> was 1.7 while it had minimum value (1) at other recordings. Maximum value for growth power was recorded for control plants at all recordings. Difference between control and 3 ds m<sup>-1</sup> salinity level was not significant at first, second and third times but significant at fourth time. Growth powers for all saline treatments were decreased during recording periods (Fig. 4).

**Multiple regressions:** Because of being first variable entered in model (p<0.001) based on Stepwise selection for shoot weight, root weight is the most important indicator of shoot weight in this multiple regression. After root weight, second leaf area (p = 0.003), tiller number (p = 0.003), Shoot/Root ratio (p = 0.096), content of  $K^+$ (p<0.001), first leaf number (p = 0.060), first leaf area (p = 0.057) and photosynthesis rate (p = 0.113) entered in model, respectively (Table 4).

**Correlation coefficient:** Mean of the morphological, biochemical and chemical traits were used for correlation coefficient analyzing. Correlations between shoot weights with other variables except concentration of  $Na^+$  were significantly positive but correlation of shoot weight with  $Na^+$  was negative and not significant (r=0.07). Highest correlation of traits with shoot weight was belonged to root weight. Content of  $Na^+$  showed significant correlation just with proline content (r=0.96), shoot per root ratio (r=0.72) and  $K^+$  (r=0.68). In general, all correlation among traits except correlations of  $Na^+$  content with shoot and root weight were positive (Table 5).

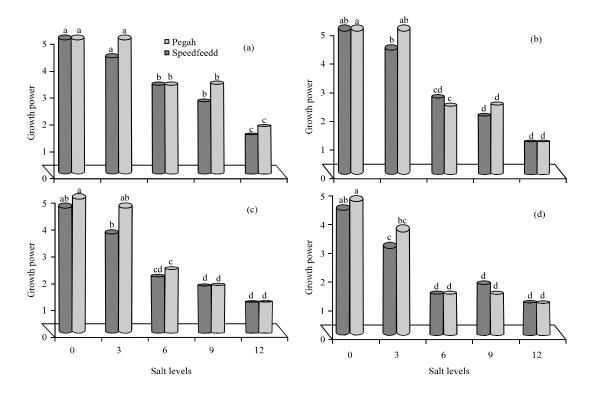


Fig. 4(a-d): Growth power of sorghum cultivars under salinity levels at first (a) second (b) third (c) and fourth time measuring. Different alphabets are significantly different at p<0.001

Table 5: Correlation coefficient among measured traits	Table 5	: Correlation	coefficient	among	measured traits
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Trait	LA	LN	height	Root (gr)	Sh/R	proline	$K^{+}$	$Na^+$	K/Na	PR	TN	sw
LA	1											
LN	0.83**	1										
height	0.91**	0.95*	1									
Root (gr)	0.91**	0.78**	0.82**	1								
Sh/R	0.60**	0.81**	0.78**	0.49**	1							
proline	0.14	0.5**	0.44*	0.074	0.80**	1						
K	0.62**	0.89**	0.85**	0.54**	0.95**	0.79**	1					
Na	0.02	0.33	0.3	-0.062	0.72**	0.96**	0.68**	1				
K/Na	0.87**	0.96**	0.96**	0.79**	0.81**	0.44**	0.88**	0.278	1			
PR	0.81**	0.89**	0.87**	0.79**	0.82**	0.50**	0.84**	0.345	0.91**	1		
TN	0.89**	0.90**	0.93**	0.81**	0.72**	0.362	0.77**	0.21	0.91**	0.89**	1	
SW	0.92**	0.75**	0.81**	0.99**	0.48**	0.05**	0.51**	-0.074	0.77**	0.76**	0.79**	1

LA: Leaf area, LN: Leaf numbers, RW: root weight, Sh/R: Shoot per root ratio, PR: Photosynthesis rate, SW: Shoot weight and TN: Tiller numbers

# DISCUSSION

Results indicate that photosynthesis rate decreased by increase salinity levels. Abiotic stresses such as salinity stress cause impairment in the balance between light capture in leaf of stressed-plants and its utilization. This extra energy of light results in production of Reactive Oxygen Species (ROS) that are so dangerous for chloroplast. ROS damage chloroplast and its photosystems consist of PSÉ and PSIT and finally results in decrease photosynthesis. On the other side less decreasing of photosynthesis rate in plants under stress conditions is an indicator for more salinity resistance.

Pegah significantly showed more photosynthesis rate that is may be due its higher resistance of its chloroplastic cells to salinity. There are many studies report decreasing photosynthesis rate under salinity stress in crops such as rice (Yeo *et al.*, 1985) sorghum, linseed (Nasir Khan *et al.*, 2007) and other plants species.

Salinity caused increases the amount of free proline in stressed-plants leaves which salinity levels of 9 ds m<sup>-1</sup> showed the highest amount. Proline is an osmoregulate and has a key role in osmotic adjustment. Salinity usually is companied with declining water absorption in root. This situation causes loss of water and turgor in plants cells that induces osmotic stress. One of the most important

plant responses to osmotic stresses is higher accumulation of osmoregulators such as proline. Accumulation of proline in plants cells induces higher osmotic adjustment and inhibiting water loss on cells. In addition to this proline can detoxify some kind of ROS such as singlet oxygen. Thus, proline is an important component for stress protection in plants. Higher amount of proline in salinity treatments of this study indicate accuring osmotic stress in this condition. On the other side, there was no significant difference between cultivars implying that both of them have same response to salinity related to proline. Free proline found to be increased in studies on wheat (Goudarzi and Pakniyat, 2009), tobacco (Kuznetsov and Shevyakova, 1997), Arabidopsis (Nanjo et al., 1999) and other plants.

Salinity increased total protein content and Speedfeed showed higher amount of protein. With occurring stresses, plants respond by expression or over-expression of some stress related genes and since the final product of most of these genes are different proteins, higher accumulation of protein can caused more protection in plants. Higher protein content could be an appropriate indicator for higher resistance of plants and because of higher protein in Speedfeed; this cultivar is an eligible cultivar to achieve breeding aims for higher resistance to salinity.

Stressed-plants had higher accumulation of Na+ indicates higher uptake of Na<sup>+</sup> in this situation, although higher levels of salinity causes higher uptake of Na<sup>+</sup>. On the other hand, salinity decreased amount of K<sup>+</sup> in plants implying that higher levels of salinity and Na+ have a negative effect on absorption and accumulation of K+. Both K<sup>+</sup> and Na<sup>+</sup> are ions that have important effect in osmotic adjustment. Higher accumulation of Na<sup>+</sup> has positive effect on osmotic adjustment but extra amount of this ion can be very dangerous for plant cells and therefore, one of important negative effects of salinity in plants is high amount of this ion. Also, K+ has an osmoregulating role and decreasing content of this ion under salinity stress has negative effects on plants cells. Salinity reduced K/Na ratio implying negative effect of salinity on K<sup>+</sup> and its positive effects on Na<sup>+</sup> uptakes. Speedfeed cultivar showed higher amount of K<sup>+</sup> and lower content of Na+ in its leaves that is may be due to higher controlling of ion uptake in this cultivar. Mahmood et al. (2010) reported higher accumulation in Na<sup>+</sup> and lower accumulation in K<sup>+</sup> under saline condition.

All growth parameters were decreased with respect to enhancing salinity levels. Decreasing the amount of growth parameters under saline condition in this study probably is due to inhibiting effects of salinity on photosynthesis and also higher use of energy to control the ion uptake in root and higher accumulation of proline. Comparing effects of salinity on emergence and growth parameters indicated that salinity had lower effects on emergent percentage than growth parameters. Pegah cultivar had higher stem height but the leaf area of Speedfeed was higher. On the other hand, the difference between cultivars for other growth parameters was not significant. Due to importance of leaves in photosynthesis, leaf area is more proper factor than height and therefore, Speedfeed is more appropriate than Pegah under saline condition.

Percent of emergence for 3, 6 and 9 ds m<sup>-1</sup> salinity levels were not different but 12 ds m<sup>-1</sup> salinity level and control had minimum and maximum percent, respectively. Plants growth rate in the first time of measuring was not significant between control and 3, 6 and 9 ds m<sup>-1</sup> salinity levels but 12 ds m<sup>-1</sup> showed different growth rate. During salinity period, growth rate of plants under salinity levels were decreased and plants under 12 ds m<sup>-1</sup> salinity level treatment were completely destructed. On the other hand, growth power of plants under saline condition showed significant difference with control in all measuring times. The results indicate that severity and lasting of the salinity are of important factors for plats in order to response to this stress. Speedfeed cultivar showed higher emergence percent and also percent of growth and growth rate than Pegah indicating higher resistance of this cultivar to salinity that is may be due to higher control in ion (Na<sup>+</sup> and K<sup>+</sup>) uptakes.

Results of multiple regressions based on stepwise selection showed that most important factor for shoot weight of sorghum under salinity condition is root weight factor. As well as root weight, shoot per root ration entered in the model as an effective variable affects shoot weight of sorghum. Salinity causes limiting available water in soils by increasing content of Na<sup>+</sup> and other substance in soil and then, absorbing water from soil becomes hard for plants roots. Higher density of root under saline condition can be so helpful for more resistance of plants bye increasing absorbing surface of root. Two stages of leaf area and first stage measuring leaf number and also photosynthesis rate were entered to the model that is implying the importance of photosynthesis rate and ability to continue of plants on producing higher shoot weight. Stepwise selection also indicated important effect of higher K<sup>+</sup> accumulation by entering this variable in the model. Results of correlation coefficient confirmed results of multiple regressions since highest coefficient correlation of traits was belonged to shoot and root weights.

### CONCLUSION

Salinity showed deleterious effects on growth parameters and caused higher accumulation of Na<sup>+</sup> and proline and also total protein content but lower accumulation of K<sup>+</sup> and decreased photosynthesis rate, indicating that severity of salinity can causes high negative effects on productivity of plants. Stepwise selection and correlation analysis showed that root density is the most important factor for plant productivity. Speedfeed in general had higher power of survive, growth and emergence and it can be a favorite cultivar for selecting or crossing programs with Pegah in order to gain more salinity resistance.

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