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Full Length Research Paper

A preliminary approach to halo sensitivity of sorghum cultivars

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Effect of five salt concentrations viz, 0, 130, 160, 190, 220 and 250 mM on germination and seedling growth of sorghum cultivars (*Asian, Indian, Mycho* and *Summer graze*) was evaluated under laboratory condition. Analysis of variance (ANOVA) revealed significant differences for germination percentage, plumule and radicle growth, fresh and dry weight for different salt concentrations except moisture contents. Inter cultivars genetic variation and concentration percentage, plumule and radicle growth, fresh and concentration x cultivars interaction showed significant differences for all the parameters studied. Data for germination percentage, plumule and radicle growth, fresh and dry weight and moisture contents clearly demonstrated varietal differences. Though results clearly showed that salinity stress decreased all the parameters studied for sorghum cultivars significantly, tolerance of sorghum cultivars to such high levels of salt stress is worth mentioning and it can be suggested that sorghum can be tried for cultivation on moderately saline areas.

Key words: germination percentage, plumule and radicle growth, fresh and dry weight, moisture contents.

INTRODUCTION

Salinity is one of the fore most important abiotic factors that retard the activity of embryo in arid and semi arid regions. Most part of the land is under the influence of salinity and water logging. Crops growing under these saline regimes are stunted and weak resulting in low productivity (Ahmed, 2009; Ahmad et al., 2012). Although Pakistan is an agrarian economy yet we are facing scarcity of cereals and forages because of the aforementioned problems. Salt stress adversely affects plants at all stages of their life cycle.

Salinity affects seed germination by creating an external osmotic potential that prevents water uptake due to the toxic effects of sodium and chloride ions on the germinating seed (Khajeh-Hosseni, 2003; Kandil et al., 2012). Plants differ in their ability, to develop under saline conditions (Greenway and Munns, 1980; Kandil et al., 2012). Salinity compacts root length and plant height as the level of salt increases (Bashir et al., 2011; Saberi et

al., 2011; Kandil et al., 2012). Plant scientists are confronting a challenge of salinity and water logging and they are trying to find out salinity resistant valuable plants. The aim of the present study was to search out salinity stress tolerant sorghum cultivars for vast barren salt effected land of Pakistan. Various researchers studied the effect of salinity on sorghum (Asfaw, 2011; Bashir et al., 2011; Kafi et al., 2011; Kandil et al., 2012; El-Naim et al., 2012; Rani et al., 2012). Though the study is preliminary, yet it will provide important clues and guide line for researchers to study further in this regard.

MATERIALS AND METHODS

Seeds of sorghum cultivars (Asian, Indian, Mycho and Summer graze) obtained from open market were subjected to 4 salt concentrations (130, 190, 220 and 250 mM). Tap water was used

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Table 1. Mean squares analysis of the varience table for plumule and radicle growth, germination percentage (%), fresh and dry weight and moisture contents of sorghum cultivars.

Source	Degree of freedom	Plumule growth	Radicle growth	Germination (%)	Fresh weight	Dry weight	Moisture contents
Concentration	4	0.0000 ^S	*****				
Cultivars	3	0.0001 ^s	0.0000 ^S	0.0000 ^S	0.0000 ^S	0.0000 ^S	0.0053 ^S
Concentration x cultivars	12	0.0000 ^S	0.0048 ^S	0.0325 ^S	0.0000 ^S	0.0000 ^S	0.0011 ^S
Error	76						
Total	99						

S = Significant NS = non significant.

Table 2. Effect of salinity on plumule growth (cm) of sorghum cultivars.

Concentration (milli moles)	Asian	Indian	Mycho	Summer graze	Concentration means
Control	7.51 ^{ab}	7.68 ^a	6.80 ^b	5.84 ^c	6.96 ^a
160	5.00 [°]	1.05 ^{fgh}	3.71 ^d	1.99 ^e	2.94 ^b
190	0.64 ^{gh}	0.71 ^{gh}	1.81 ^{ef}	1.49 ^{efg}	1.16 [°]
220	0.60 ^h	0.48 ^h	0.49 ^h	0.58 ^h	0.54 ^d
250	0.53 ^h	0.48 ^h	0.32 ^h	0.38 ^h	0.43 ^d
Cultivars means	2.86 ^a	2.08 ^b	2.63 ^a	2.06 ^b	

Lsd value at 5% level of significance for concentration = 0.4286, cultivars = 0.3833 and concentration x cultivars interaction = 0.8571. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

as control. Seeds viability was checked prior to the start of the experiment. Seeds were placed equidistantly on two folded Whatman # 1 filter paper as seed bed in Petri dishes of equal size (5 cm). Each treatment was replicated 5 times with 10 seeds in each replicate. Equal volume of solutions was added to the Petri dishes from respective salt concentrations. The glass wares were properly washed and sterilized at 65°C for 24 h in oven prior to use. The dishes were incubated at 25°C for 72 h. After 72 h, data for germination percentage (%), plumule and radicle growth (cm) and fresh weight (gm) were collected. Dry weight (g) and moisture contents (%) were determined after the seedlings were dried in oven at 65°C for 72 h following Hussain (1989). Fisher analysis of variance technique (1985) and LSD test at 5% probability was applied on the data to compare the differences among treatment means (Steel and Torie, 1980).

RESULTS AND DISCUSSION

Effect on plumule growth (cm)

ANOVA for plumule growth revealed significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (6.96 cm) for plumule growth was recorded in control, followed by 160 (2.94 cm), 190 (1.16 cm), 220 (0.45 cm) and 250 (0.43 cm) mM salt concentration, respectively. Among cultivars, *Asian* showed maximum mean value (2.86 cm) for plumule growth, while cultivar *Summer*

graze showed minimum mean value (2.06 cm) for plumule growth. Differences between cultivar Asian and cultivar Mycho was not significant for plumule growth. Similarly, cultivar Indian and cultivar Summer graze showed non-significant differences. In concentrations x cultivars interaction, maximum mean value (7.68 cm) was recorded in control for cultivar Indian, while minimum (0.32 cm) for cultivar *Mycho* in 250 mM salt concentration (Table 2). Concentration dependent decrease was observed in plumule growth (cm) of sorghum cultivars (Figure 1). Jamil et al. (2006) reported that root and shoot lengths are important traits in salt stress sensitivity evaluation. Arrest of plumule growth of sorghum cultivars may be seen due to the toxic or injurious effects of different ions on embryo or may be due to osmotic disturbance uptake of ions was effected resulting in abnormal metabolism (Farooq and Azam, 2006; Maghsoudi-Moudi and Maghsoudi, 2008; Kazemi and Eskandari, 2011; Ahmad et al., 2012).

One may also conclude that ions of different charges bind with the domain of DNA or enzymes responsible for normal metabolism resulting in retarded plumule growth. Mechanisms of plant adaptation under salt stress are complex. The initial sites of the cell injury by any environmental stress firstly appears in cell membranes (Ashraf and Ali, 2008; Kafi et al., 2011). The imbibition of water was reduced due to the increased osmolality of salt



Figure 1. Effect of salinity on plumule and radicle growth (cm) of sorghum.

Concentration	Asian	Indian	Mycho	Summor arazo	Concentration
(milli moles)	Asian	mulan	wycho	Summer graze	means
Control	10.98 ^b	10.97 ^b	13.63 ^ª	10.57 ^b	11.54 ^ª
160	5.54 ^{cd}	2.57 ^{fhg}	7.09 ^c	4.08 ^{def}	4.82 ^b
190	2.23 ^{fgh}	2.78 ^{fgh}	5.45 ^{cd}	5.07 ^{de}	3.88 ^b
220	2.61 ^{fgh}	1.09 ^h	3.49 ^{efg}	2.39 ^{fgh}	2.37 ^c
250	2.02 ^{gh}	2.70 ^{fgh}	1.98 ^{gh}	3.05 ^{fg}	2.44 ^c
Cultivars means	4.67 ^{bc}	4.02 ^c	6.31 ^a	5.03 ^b	

Table 3. Effect of salinity on radicle growth (cm) of sorghum cultivars.

Lsd value at 5% level of significance for concentration = 0.9550, cultivars = 0.8541 and concentration x cultivars interaction = 1.910. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

solutions (Wahid et al., 1998). Among cultivars, Asian was more tolerable to salinity and showed maximum plumule growth (Figure 4). It may be concluded that cellular membrane permeability of cultivar Asian is less as compared to other cultivars (Ismail, 2003). Naseer (2001) reported that the reduction in shoot length is due to excessive accumulation of salt in the cell wall which modifies the metabolic activities and limits the cell wall elasticity. Further, secondary cell appears sooner and cell wall becomes rigid, as a consequence the turgor pressure efficiency in cell enlargement decreases. These processes may cause the shoot to remain small. Our findings are parallel to those of Jamil et al. (2006) on vegetables species, Rahman et al. (2008) on wheat, Baybordi and Tabatabaei (2009), Keshavarzi (2012) on Artemesia and Moradi and Zavareh (2013) on chickpea, thus confirmed our findings for plumule growth.

Effect on radicle growth (cm)

ANOVA for radicle growth exhibited significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (11.54 cm) for radicle growth was recorded in control which was significantly different from the rest of all the salt concentrations. Differences between 160 and 190 mM salt concentrations for radicle growth were non-

significant. Similarly, differences between 220 and 250 mM salt concentrations were also non significant for radicle growth. Cultivar *Mycho* showed maximum mean value (6.31 cm) for radicle growth, followed by *Summer graze* (5.03 cm), *Asian* (4.67 cm) and *Indian* (4.02 cm), respectively. In concentrations x cultivars interaction, maximum mean value (13.36 cm) for radicle growth was recorded in control for cultivar *Mycho* while minimum mean value (1.09 cm) was observed in 220 mM salt concentration for cultivar *Indian* (Table 3). Water stress condition caused marked reduction in radicle growth (Ahmad et al., 2012) (Figure 1). Salinity affects the seedling growth of plant by slow or less mobilization of reserved food, suspending the cell division, enlargement and injuring hypocotyls (Tezara et al., 2003).

The data on the average length of radicle and plumule shows that decrease in length of plumule was more pronounced as compared to radicle in all NaCl salt treatment. This indicated that plumule is more sensitive than radicle. It may be due to ability of the radicle system to control entry of ions as compared to plumule (Moradi and Zavareh, 2013). Stress tolerance is dependent upon the genetic and biochemical characteristics of the cultivars (Barakat, 2003).

Our study shows that cultivar *Mycho* is genetically well adapted to salt stress than other sorghum cultivars (Figure 4). Our findings are in complete accordance with the Keshavarzi (2012), Saberi et al. (2012) and Moradi

Concentration (milli moles)	Asian	Indian	Mycho	Summer graze	Concentration means
Control	86.00 ^{ab}	82.00 ^{abc}	96.00 ^a	76.00 ^{bcd}	85.00 ^a
160	68.00 ^{cde}	44.00 ^{h-k}	68.00 ^{cde}	40.00 ^{i-l}	55.00 ^b
190	64.00 ^{d-g}	54.00 ^{e-i}	58.00 ^{e-h}	44.00 ^{h-k}	55.00 ^b
220	66.00 ^{c-f}	36.00 ^{j-m}	48.00 ^{g-j}	20.00 ^{mn}	42.50 [°]
250	28.00 ^{k-n}	12.00 ⁿ	50.00 ^{f-j}	26.001 ^{mn}	29.00 ^d
Cultivars means	62.40 ^a	45.60 ^b	64.00 ^a	41.20 ^b	

Table 4. Effect of salinity on germination percentage (%) of sorghum cultivars.

Lsd value at 5% level of significance for concentration = 8.858, cultivars = 7.923 and concentration x cultivars interaction = 17.72. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

and Zavareh (2013) whom studied effect of salinity on *Trifolium*, *Artemesia* and chick pea, respectively, thus confirming our results regarding radicle growth.

Effect on germination (%)

ANOVA for germination percentage (%) exhibited significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (85.00%) for germination percentage (%) was observed in control while minimum in 250 mM salt concentration. Control showed significant differences for all the salt concentrations. Cultivar *Mycho* showed maximum mean value (64.00%) for germination percent-tage (%) while cultivar *Summer graze* showed minimum mean value (41.20%).

Differences between cultivar Mycho and cultivar Asian was non-significant for germination percentage (%). Similarly cultivar Indian and cultivar Summer graze was non-significantly different. In interaction of concentrations x cultivars, maximum mean value (96.00%) for germination percentage (%) was recorded in control for cultivar Mycho while minimum mean value (12.00%) was observed for cultivar Indian in 250 mM salt concentration (Table 4). Salinity concentrations are inhibitory for germination percentage of sorghum cultivars (Figure 3). Salinity exerts its undesirable effects through osmotic inhibition and ionic toxicity (Munns et al., 2006). The differences in germination percentage of forage sorghum cultivars may be due the genetic factors and heredity variation among the four sorghum cultivars (Figure 6). Our results regarding germination percentage of sorghum cultivars are confirmed by Kausar et al. (2012), Keshawarzi (2012), Saberi et al. (2012) and Moradi and Zavareh (2013) who studied ill effects of salinity on germination of sorghum, Artemesia, Trifolium and chick pea, respectively. Furthermore Anwar et al. (2001), Zia and Khan (2002) and Muhammad and Hussain (2010) also reported reduced germination under saline conditions in some medicinal plants that also strengthen our findings.

Effect on fresh weight (g)

ANOVA for fresh weight was significantly different for concentrations, cultivars and concentration x cultivars interaction (Table 1). Maximum mean value (1.29 g) for fresh weight was observed in control which was significantly different from all the salt concentrations. Differences among the salt concentrations were significant for fresh weight. Cultivar Indian showed maximum mean value (0.75 gm) for fresh weight. While cultivar Summer graze showed minimum mean value (0.32 g). In interaction of concentrations x cultivars, maximum mean value (1.77 g) for fresh weight was observed for cultivar Indian in control which was higher than control of salt concentrations while minimum mean value (0.11 g) was recorded for cultivar Indian in 250 mM salt concentrations (Table 5). Increase in salinity level caused simultaneous reduction of seedling fresh weights in all sorghum cultivars (Figure 2).

This is in line with previous reports in wheat (Afzal et al., 2005), sugar beet, cabbage, amaranth and pak-choi (Jamil et al., 2006) and in sorghum (Asfaw, 2011). Thus our findings regarding fresh biomass are confirmed. Reduction in fresh biomass at higher concentration might be due to poor absorption of water from the growth medium due to physiological drought (Muhammad and Hussain, 2010). Salt stress disturbed the biochemical machinery resulting in reduced biomass. Cultivar *Indian* has got good genetic variations for salinity tolerance thus showed increase in fresh biomass (Figure 4).

Effect on dry weight (g)

ANOVA for dry weight exhibited significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (0.53 g) for dry weight was recorded in control while minimum in 250 mM salt concentration. Among cultivars, *Asian* showed maximum mean value (0.31 g) for dry weight while minimum mean value (0.16 g) was recorded for *Summer graze*. In concentration x cultivars interaction, control of

Concentration	Asian	Indian	Mycho	Summer graze	Concentration
(milli moles)	Asian	mulan			means
Control	1.44 ^b	1.77 ^a	1.37 ^b	0.56 ^{ef}	1.29 ^a
160	0.84 ^c	0.81 ^{cd}	0.59 ^{de}	0.27 ^{ghi}	0.63 ^b
190	0.57 ^{ef}	0.56 ^{ef}	0.52 ^{ef}	0.39 ^{e-h}	0.51°
220	0.35 ^{fgh}	0.50 ^{efg}	0.27 ^{ghi}	0.19 ^{hi}	0.33 ^d
250	0.26 ^{hi}	0.11 ⁱ	0.22 ^{hi}	0.18 ^{hi}	0.19 ^e
Cultivars means	0.69 ^{ab}	0.75 ^a	0.60 ^b	0.32 ^c	

Table 5. Effect of salinity on fresh weight/seedling (gram) of sorghum cultivars.

Lsd value at 5% level of significance for concentration = 0.1161, cultivars = 0.1039 and concentration x cultivars interaction = 0.2323. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.



Figure 2. Effect of salinity on fresh and dry weight/seedling (gram) of sorghum.



Figure 3. Effect of salinity on germination and moisture contents (%) of sorghum.

cultivar *Mycho* represented maximum mean value (0.71 g) which was higher than control of salt concentrations while minimum mean value (0.03 gm) was observed for cultivar *Indian* in 250 mM salt concentration (Table 6). Increased salinity level caused simultaneous reduction of seedling dry weights in all the cultivars (Figure 2). Salt stress disturbed the biochemical machinery resulting in reduced dry biomass. Increase dry biomass of cultivar *Asian* indicated good genetic variations for salinity (Figure 4). This is in line with previous reports in wheat (Afzal et al., 2005), sugar beet, cabbage, amaranth and pak-choi (Jamil et al., 2006) and in sorghum (Asfaw,

2011) and we reported the same. The decrease in seedling dry weight with increasing salinity level is in contrast with the findings of Muhammad and Hussain (2010) who reported increase in seedling dry weight of *Lepidium sativum*, *Linum usitatissimum*, *Plantago ovata* and *Trigonella foenum-graecum* (Figure 5).

Effect on moisture contents (%)

ANOVA for moisture contents revealed non-significant differences for concentrations while differences were



Figure 4. Effect of salinity on plumule and radicle growth (cm) of sorghum cultivars.

Concentration	Asian	Indian	Mucho	Summor arozo	Concentration
(milli moles)	(milli moles)		wycho	Summer graze	means
Control	0.60 ^b	0.43 ^c	0.71 ^ª	0.36 ^{cd}	0.53 ^ª
160	0.42 ^c	0.32 ^{de}	0.29 ^{def}	0.12 ^{h-k}	0.29 ^b
190	0.23 ^{fg}	0.25 ^{efg}	0.23 ^{fg}	0.16 ^{ghi}	0.22 ^c
220	0.18 ^{gh}	0.11 ^{h-k}	0.14 ^{hij}	0.07 ^{jk}	0.13 ^d
250	0.11 ^{h-k}	0.03 ^k	0.09 ^{h-k}	0.08 ^{ijk}	0.08 ^d
Cultivars Means	0.31 ^ª	0.23 ^b	0.29 ^a	0.16 ^c	

Table 6. Effect of salinity on dry weight/seedling (gram) of sorghum cultivars.

Lsd value at 5% level of significance for concentration = 0.04454, cultivars = 0.03983 and concentration x cultivars interaction = 0.08907. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.



Cultivars

Figure 5. Effect of salinity on fresh and dry weight/seedling (gram) of sorghum cultivars.

significant for cultivars and between concentrations and cultivars interaction (Table 1). Maximum mean value (165.9 g) for moisture contents (%) was recorded in control. Cultivar *Indian* showed maximum mean value (195.4%) for moisture contents. While minimum mean value (128.9 g) was observed for cultivar *Asian*. In concentrations x cultivars interaction, maximum mean value (432.1%) for moisture contents was observed for cultivar *Indian* in control while minimum (98.8%) in 220 salt concentration for cultivar *Mycho* (Table 7). Moisture contents of seedling play an important role in various physiological processes including growth (Muhammad and Hussain, 2010; Ahmad et al., 2012). In the present

study, decrease in moisture contents was recorded with salinity treatments (Figures 3 and 6). Shadded and Zaidan (1989), Naqvi and Mahmood (1994) and Ahmad et al. (2012) recorded reduction in moisture contents with increase salt stress in *Trigonella*, *Sesbania sesban* and canola, respectively, thus confirmed our findings. The decrease in moisture contents with increasing salinity level is in contrast with the findings of Ibrar and Hussain (2003), Akhtar and Hussain (2008) and Muhammad and Hussain (2010) who reported increase in moisture contents with increase in moisture contents with increase in moisture contents with increase in moisture contents, respectively.

Concentration (milli moles)	Asian	Indian	Mycho	Summer graze	Concentration Means
Control	146.1 ^{cd}	324.1 ^ª	101.1 ^d	92.2 ^d	165.9
160	93.0 ^d	155.0 ^{bcd}	119.0 ^{cd}	147.9 ^{cd}	128.7
190	150.7 ^{cd}	163.3 ^{bcd}	178.0 ^{bcd}	136.2 ^{cd}	157.1
220	155.0 ^{cd}	195.0 ^{ьс}	98.8 ^d	241.3 ^{ab}	162.5
250	140.0 ^{cd}	139.5 ^{cd}	164.0 ^{bcd}	126.0 ^{cd}	142.4
Cultivars Means	128.9 ^b	195.4 ^ª	132.2 ^b	148.7 ^b	

Table 7. Effect of salinity on moisture contents (%) of sorghum cultivars.

Lsd value at 5% level of significance for Cultivars = 40.31 and Concentration x Cultivars interaction = 90.15. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.



Cultivars

Figure 6. Effect of salinity on germination and moisture contents (%) of sorghum cultivars.

This preliminary laboratory study suggests that the tested sorghum cultivars could be grown on marginally saline habitats due to their tolerance to moderate salinity at germination and seedling stage. However, further study is needed to test their salt tolerance under field conditions to assess the possibility of cultivation of sorghum cultivars on otherwise unproductive lands.

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