Screening of sorghum (*Sorghum bicolor* L) genotypes under various levels of drought stress

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Abstract
Sorghum is an important fodder crop and plays an important role in the Pakistan dairy industry. High yielding and superior quality sorghum varieties should be produced to meet the domestic needs. In arid and semiarid regions, drought is a major and serious constraint to sorghum production and adversely influences sorghum growth and germination. There are many approaches to combat the drought stress. Among these approaches breeding of crops contributes towards increase in yield under stress condition like drought stress by making the plant tolerant against stress. Ten genotypes of sorghum were evaluated at seedling stage to determine the genotypic variation among them on the basis of tolerance against drought stress and impact of drought on fodder quality. Three levels of water (100%, 75%, and 50% field capacity) were applied to the genotypes. The experiment was carried out in wire house following a triplicate completely randomized design with factorial arrangements. The data were recorded after 20 days of sowing on following traits such as root and shoot length, root shoot fresh and dry weight, leaf area, crude protein, relative water content, total ash contents and chlorophyll contents. Significant differences were found among the genotypes for all traits. The genotypes NARC-11 followed by Sorgh-11 gave better response in all levels of drought stress while F-114 gave poor response in all levels of drought stress.

Keywords: sorghum, drought, genotypes and quality

Introduction

Sorghum (*Sorghum bicolor*) is a multiuse crop grown for feed, food and bioenergy. Sorghum can be cultivated in numerous environments due to its wider adaptation property. Sorghum is a model crop for a more concerned crop improvement program in agriculture to utilize marginal lands, to meet energy and food demands which might be increased in the near future (Bibi et al, 2012a). As compared to other cereal crops sorghum considered to be more tolerant to different stresses including drought, heat, flooding and salinity (Ejeta and Knoll, 2007; Ali et al, 2011) however, crop cultivated in rainfed zones is highly effected by drought stress (Kebede et al, 2001).

Plant abiotic stresses such as drought stress threaten stable global food availability as the increase in world population and per capita food consumption. Drought or any other abiotic stresses cause the reduction of yield and plant growth. They limit the photosynthesis and consequently, limited availability of photosynthetic assimilates and energy to the plant. It is necessary for plants to use this limited supply of nutrients to gain maximum advantage under stress. Under drought stress conditions, plant should increase the uptake of water, which is usually more available in deep soil (Xiong et al, 2006). The major goal of plant breeders and physiologists in sorghum is to identify and understand the mechanism of drought tolerance which include ability to maintain stomatal opening at lower level of leaf water potential, high osmotic adjustment and prolific root system (Ranjendran et al, 2011).

Almost every developmental stage of the plant is effected by drought stress. However, damaging effects of drought was more prominent when it coincided with various growth stages such as germination; seedling root length, shoot length and flowering (Rauf, 2008; Khayatnezhad et al, 2010). Under water deficit conditions, plants urgently need available water in root zone, and tolerant genotypes will extract water from deep layers of soil (Xiong et al, 2006). Generally, it has been observed that drought tolerant crop species has longer roots with more root density (Kaydan and Yagmur, 2008; Achakzai, 2009). Dhanda et al (2004) reported that the decrease in water availability affects the crop production at different growth stages but generally resulted in decreased coleoptile length, higher root:shoot ratio and longer roots. These water sensitive growth stages may be exploited to differentiate genotypes on the basis of their resistance to drought stress. Among these critical stages, drought stress at seedling stage has been exploited for screening germplasm in various crops i.e. Wheat (Xiong et al, 2006; Balota et al, 2008), sorghum (Bibi et al, 2010; Achakzai, 2011; Ali et al, 2011;

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Qadir et al, 2015) maize (Hajibabaee et al, 2012; Qayyum et al, 2012).

The objective of this research was to assess response of 10 different genotypes against drought stress at seedling stage.

Materials and Methods

Ten sorghum genotypes collected were sown in the wire house to find out the effect of water stress on various morphological and quality characters of forage sorghum. All the experimental plants were sown in metallic trays following completely randomized design with factorial arrangements with three replications maintaining 4.5 cm plant to plant distance and 6.5 cm row to row distance. Three levels of water, one normal and two drought levels (75% and 50% of normal) were applied. Three plants of each entry per replication were uprooted after 20 days of sowing for data recording.

The seedlings were washed, dried on paper towels and data regarding shoot length, root length, fresh shoot weight, fresh root weight, dry shoot weight, dry root weight, leaf area were recorded.

Relative water content (%)

Relative water contents were calculated using the formula (Teulat et al, 2003)

RWC (%) = FW- DW/TW-DW ×100, where RWC = Relative water contents, FW = Fresh weight, DW = Dry weight, TD = Turgid weight

Crude protein (%)

Protein % was calculated by multiplying N-contents with a factor 6.25 (AOAC, 1996). Crude protein % = 100 x 6.25 [(ml N/10 H_2SO_4 neutralized by NH_3 × 0.0014 × total diluted volume) / (weight of sample × ml of dil digested material distilled)].

Total ash (%)

The mineral elements as a group were determined in a sample by burning off the organic matter and weighing the residue, which was called ash. It is calculated using the calculation (AOAC, 1965).

Ash % = 100 (weight of ash / weight of sample)

Chlorophyll contents (SPAD value)

SPAD-502 plus chlorophyll meter (Konica Minolta) was used to determine the chlorophyll contents.

Statistical analysis

Standard statistical procedures were used to analyze the data and means were compared by least significance difference test.

Results

Results showed that drought levels significantly reduced the shoot length, shoot fresh and dry weight, root fresh and dry weight, leaf area, total ash contents, relative water contents and chlorophyll contents. All genotypes showed variable response against drought stress. Table 1 showed that at all water regimes NARC-11 had highest value for shoot length followed by sorgh-11 and minimum value for shoot length was obtained in F-114. Data regarding root length (Table 1) showed that in first water regime (100 water applied) NARC-11 had maximum root length (11.25 cm) followed by sorgh-11 while minimum was observed in F-114 (7.15 cm). At second (75% of normal) and third water regime (50% of normal), NARC-11 gave better root length followed by sorgh-11 as compared to other genotypes and minimum root length was produced by F-114 and F-113. Data regarding shoot fresh weight (Table 1) revealed that NARC-11 had maximum shoot fresh weight and F-114 produced minimum shoot fresh weight at all drought levels. Data (Table 2) depicted that at all drought levels highest shoot dry weight was showed by NARC-11 followed by sorgh-11 and minimum by F-114. Results (Table 2) showed that maximum root fresh and dry weights at all water regimes were given by NARC-11 followed by sorgh-11 while minimum were observed in F-114. Table 3 indicated that at first water regime (100% water applied) NARC-11 had highest value (13.61cm2) for leaf area followed by NOOR and minimum value was observed in FA-08 (11.50 cm2). NARC-11 showed the maximum leaf area (9.36 cm^2) followed by NOOR and lowest was observed by FA-08 (7.68 cm^2) at second water regime (75% of normal). At 50% of normal NARC-11 had the highest leaf area (7.43 cm^2) followed by NOOR and minimum value was observed in FA-08 (5.30 cm^2). Table 3 showed that at 1st water level (100% of normal) NOOR showed highest value for total ash contents and minimum value was observed in FA-08. While at second and third drought

Table 1 - Comparisons of sorghum genotypes for shoot length, root length and fresh shoot weight under three water regimes.

	Shoot length (cm)			Root length (cm)			Fresh shoot weight (g)		
	Drought levels			Drought levels			Drought levels		
Genotypes	100%	75%	50%	100%	75%	50%	100%	75%	50%
FS-08	24.25 ef	21.05 ij	16.65 mn	10.10 k-m	10.55 i-l	12.25 d-f	0.81 g-i	0.75 j-l	0.66 no
FSD-11	23.15 fg	21.75 hi	15.56 no	9.75 l-n	11.75 e-g	13.45 bc	0.9 d-f	0.84 f-h	0.72 k-m
NOOR	25.60 c-e	23.00 f-h	18.33 kl	9.65 mn	9.25 no	12.8 cd	0.9 c-e	0.84 e-g	0.72 lm
F-114	21.15 i	19.75 jk	14.35 o	7.15 q	8.35 p	10.90 h-k	0.78 i-k	0.74 j-m	0.63 o
F-113	25.95 cd	22.91 f-h	17.01 lm	9.60 mn	8.35 p	11.65 f-h	0.93 b-d	0.87 e-g	0.72 j-m
NARC-11	28.35 a	24.95 de	19.55 k	11.25 g-i	13.75 b	15.15 a	0.99 a	0.87 e-g	0.75 j-m
AARI-10	25.50 c-e	23.30 fg	16.33 mn	8.71 op	9.75 l-n	13.50 bc	0.84 e-g	0.78 h-j	0.72 l-n
AARI-08	27.50 ab	21.50 i	17.21 lm	8.51 op	10.35 j-m	11.09 g-j	0.87 d-f	0.81 g-i	0.69 m-o
FA-08	26.35 bc	22.05 g-i	17.12 lm	8.22 p	9.25 no	13.68 b	0.96 a-c	0.87 d-g	0.72 j-m
Sorgh-11	27.45 ab	24.86 de	19.32 k	10.50 i-l	12.51 de	13.70 b	0.96 ab	0.84 e-g	0.75 j-m

Table 2 - Comparisons of sorghum genotypes for fresh root weight, dry shoot weight and dry root weight under three water regimes.

Genotypes	Fresh root weight (g) Drought levels 100%	75%	50%	Dry shoot weight (g) Drought levels 100%	75%	50%	Dry root weight (g) Drought levels 100%	75%	50%
FS-08	0.147 f	0.135 gh	0.117 jk	0.147 f	0.135 gh	0.117 jk	0.147 f	0.135 gh	0.117 jk
FSD-11	0.135 gh	0.123 ij	0.117 jk	0.135 gh	0.123 ij	0.117 jk	0.135 gh	0.123 ij	0.117 jk
NOOR	0.147 f	0.129 hi	0.115 jk	0.147 f	0.129 hi	0.115 jk	0.147 f	0.129 hi	0.115 jk
F-114	0.114 jk	0.099 l	0.075 m	0.114 jk	0.099 l	0.075 m	0.114 jk	0.099 l	0.075 m
F-113	0.117 jk	0.099 l	0.075 m	0.117 jk	0.099 l	0.075 m	0.117 jk	0.099 l	0.075 m
NARC-11	0.234 a	0.213 b	0.189 c	0.234 a	0.213 b	0.189 c	0.234 a	0.213 b	0.189 c
AARI-10	0.141 fg	0.132 g-i	0.108 kl	0.141 fg	0.132 g-i	0.108 kl	0.141 fg	0.132 g-i	0.108 kl
AARI-08	0.195 c	0.174 d	0.141 fg	0.195 c	0.174 d	0.141 fg	0.195 c	0.174 d	0.141 fg
FA-08	0.132 g-i	0.123 ij	0.099 l	0.132 g-i	0.123 ij	0.099 l	0.132 g-i	0.123 ij	0.099 l
Sorgh-11	0.213 b	0.195 c	0.162 e	0.213 b	0.195 c	0.162 e	0.213 b	0.195 c	0.162 e

level (75% and 50% of normal) NOOR showed the highest value while AARI-08 showed lowest value for this trait. Table 3 results showed that at normal level highest value was observed by NARC-11 with lowest value of FA-08 for crude protein. While at 2nd and 3rd drought level F-114 produced lowest protein contents with highest protein contents of NARC-11. Results (Table 4) showed that at 1st water regime (100% water applied) NARC-11 had highest value for relative water contents while minimum value was observed in FS-08. At 2nd and 3rd water regime (75% and 50% of normal) NARC-11 showed the highest value for relative water contents while minimum value was observed by F-114. (50.28%). Data regarding chlorophyll contents (Table 4) showed that different genotypes had different values of chlorophyll contents at all levels of drought. Among the all genotypes NARC-11 and sorgho-11 had highest values for chlorophyll contents while minimum values were observed in AARI-08.

Discussion

Drought stress cause the reduction of expression of traits shoot length, fresh shoot weight, dry shoot weight, fresh root weight, dry shoot weight, relative water contents, leaf area, total ash contents and chlorophyll contents. The results are similar as Foyer et al (1994), Ali et al (2011), and Bibi et al (2010, 2012a). Crude protein increase due to water stress and most genotypes showed the increase in root length which shows that root length is less effected than shoot length. Similar findings were observed by Younis et al (2000), Bibi et al (2010), and Qadir et al (2015).

The root is the one which face the water stress firstly, the increase in root length but decrease in root weight is due to thin roots. Which is due to restriction in cell division caused by water stress. Root play a key role in tolerating water stress by reducing leaf expansion and promoting root growth. Ali et al (2011) reported that root length at seedling stage provides a fair estimate about the root growth in field. Shoot length was decreased due to water stress in genotypes. Our findings are in agreement with Achakzai (2009), Khakwani et al (2011), Bibi et al (2010), and Qadir et al (2015). They also found significant decrease in shoot length under water stress conditions. Reduction in shoot length occurred due to less water absorption and decrease in external osmotic potential created by water stress (Kaydan and Yagmur, 2008).

The decrease in fresh shoot weight during the drought period was due their small leaf size to minimize transpiration, ultimately plant fresh weight also reduced. Drought stress significantly affected rapidly going mitotic division to produce new biomass. Our results are in agreement with Achakzai (2009) and Bibi et al (2010) under normal and water stress conditions who also showed significant decrease in fresh shoot weight under limited water supply. Dry shoot weight was also decreased significantly in sorghum under water stress conditions as reported by Achakzai (2009) and Khakwani et al (2011). Under water stress fresh and dry root weight was also decreased significantly due to water stress in sorghum. Our findings are in accordance with Shiralipour and West (1984), Bibi et al (2012a), and Qayyum et al (2012), who also

Table 3 - Comparisons of sorghum genotypes for leaf area, total ash contents and crude protein under three water regimes.

Genotypes	Leaf area (cm^2) Drought levels 100%	75%	50%	Total ash contents (%) Drought levels 100%	75%	50%	Crude protein (%) Drought levels 100%	75%	50%
FS-08	12.04 c-e	7.91 i-k	6.35 mn	12.33 gh	11.30 j	9.83 m-o	3.19 no	3.99 g-i	5.01 d
FSD-11	11.92 de	7.68 i-l	6.43 mn	13.44 d-f	10.71 j-l	9.48 n-p	3.22 m-o	4.15 fg	5.23 bc
NOOR	13.06 ab	7.81 i-k	6.55 mn	15.33 a	13.99 cd	10.33 k-m	3.39 k-m	4.08 gh	4.91 d
F-114	12.98 ab	8.78 f-h	5.80 o	14.49 bc	11.21 j	9.01 pq	3.41 kl	3.86 i	4.86 d
F-113	12.16 c-e	8.41 g-i	6.97 lm	15.10 ab	11.48 ij	9.99 l-n	3.41 kl	4.30 ef	5.35 a-c
NARC-11	13.61 a	9.36 f	7.43 j-l	14.65 a-c	13.84 c-e	9.34 n-q	3.61 j	4.37 e	5.50 a
AARI-10	11.68 de	8.76 f-h	6.07 n	13.14 e-g	12.93 f-h	8.84 pq	3.48 j-l	4.12 gh	5.20 c
AARI-08	12.36 b-d	7.74 i-l	6.43 mn	15.14 ab	10.11 mn	8.59 q	3.54 jk	4.09 gh	5.35 a-c
FA-08	11.05 e	9.13 fg	7.10 k-m	12.23 hi	11.07 jk	9.33 n-q	3.09 o	3.96 hi	4.98 d
Sorgh-11	12.85 a-c	8.24 h-j	6.55 mn	14.33 bc	12.46 gh	9.09 o-q	3.35 l-n	4.31 ef	5.40 ab

Table 4 - Comparisons of sorghum genotypes for relative water contents and chlorophyll contents under three water regimes.

	Relative water contents (%) Drought levels			Chlorophyll content (SPAD value) Drought levels		
Genotypes	100%	75%	50%	100%	75%	50%
FS-08	65.56 e	53.28 gh	49.86 k-o	17.31 a-d	14.85 c-i	12.18 i-l
FSD-11	66.33 c-e	51.00 i-l	48.92 n-p	16.36 a-g	14.35 d-j	12.14 i-l
NOOR	67.83 bc	51.33 i-k	49.73 l-p	16.56 a-f	14.21 e-j	11.49 j-l
F-114	66.21 de	50.28 k-n	46.89 q	17.41 a-c	14.07 e-j	10.25 k-i
F-113	67.41 b-d	52.46 hi	48.66 op	17.01 a-e	14.41 d-j	11.7 j-l
NARC-11	69.36 a	55.26 f	50.81 j-l	18.56 a	15.4 b-h	13.93 f-j
AARI-10	67.46 b-d	50.46 j-m	50.30 k-n	17.21 a-d	15.09 b-i	12.19 i-l
AARI-08	68.33 ab	51.86 h-j	48.34 pq	15.51 b-h	13.21 h-k	10.05 l
FA-08	66.46 c-e	50.99 i-l	49.23 m-p	15.96 a-h	13.45 g-j	11.49 j-l
Sorgh-11	68.88 ab	54.34 fg	50.30 k-n	18.04 ab	15.2 b-h	13.16 h-k

found that fresh and dry root weight was decreased during the drought period. Leaf area was most effected due to water stress and is decreased as the water stress increased. Qadir et al (2015) reported the similar results. Reduction in leaf area is due to minimizing the loss through evapotranspiration. So the leaf area cannot increased under water stress.

Relative water content (RWC) is a key indicator of the degree of tissue and cell hydration, which is vital for optimal physiological functioning and growth processes. Under drought condition relative water contents usually decreased significantly as water stress increased subsequently. Leaf enlargement, stomatal opening and associated leaf photosynthesis are essential physiological and morphological processes. These processes directly affected due to the reduction of leaf turgor potential which was due to loss of water from leaf tissue. Our findings are in agreement with Oregan et al (1993) and Lonbani and Arzani (2009). Their results also showed that relative water contents were decreased during the drought period. Relative water contents had been utilized as a selection criterion for drought tolerance by many plant breeders. Several studies have shown that maintaining a relatively high RWC during drought stress is indicative of drought tolerance (Jamaux, 1997; Altinkut et al, 2001; Colom and Vazzana, 2003). Increasing the level of drought stress significantly reduced the ash contents, as previously reported in barley, wheat and sorghum plants exposed to different water levels (Araus et al, 1998; Voltas et al, 1998; Cabrera-Bosquet et al, 2009; Bibi et al, 2012b, Qadir et al, 2015). Increase in crude protein was also observed under water stress. Similar results were observed in sorghum (Bibi et al, 2012b) and soybean (Ibrahim and Kandil, 2007). Drought stress caused the decrease in chlorophyll contents. The reason for decrease in chlorophyll contents due to drought stress might be due to production of reactive oxygen species that causes lipid peroxidation and consequently disrupt the chloroplast structure (Foyer et al, 1994). Khayatnezhad et al (2011) reported that plant showed different physiological effects under drought stress and this amount of damage depends upon the plant tolerance and stress intensity.

Conclusion

Results showed that among the genotypes, NARC-11 and sorgh-11 showed better performance in normal as well as drought conditions. Due to their drought tolerance properties these genotypes will be used in future breeding program for producing drought tolerant genotypes.

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