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In Vitro evaluation of sorghum (*Sorghum bicolor* Var. Moench) genotypes under drought condition

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ABSTRACT

Drought is a serious concern in agriculture, affecting yields all around the world, and is becoming a major source of food scarcity in many nations. The genetic potential of twelve different sorghum genotypes for drought tolerance and germination was evaluated at Sardarkrushinagar Dantiwada Agricultural University in 2018. Water stress was created by employing non-ionic PEG-6000 to lower water potential. The experiment was set up using a complete randomized design with three replications. The experiment included twelve distinct sorghum genotypes with four different PEG-6000 treatments ((S1) 0%, (S2) 5%, (S3) 10%, and (S4) 15% treatment). Our goal in this study was to evaluate 12 sorghum genotypes based on physiological responses as well as drought tolerance. Germination percentage, shoot length, root length, shot dry weight, root dry weight, seedling vigour index, and Root: Shoot ratio were all traits evaluated. Significant differences were observed among the treatments in all 12 sorghum genotypes and genotypeDS-183 was more tolerant to simulated drought stress with PEG-6000 than other sorghum genotypes.

Keywords: Sorghum, drought, germination percentage, seedling vigour index, root: shoot ratio

INTRODUCTION

Sorghum (*Sorghum bicolor* Var. Moench) is a multifunctional crop used for food, animal feed, and industry. Although sorghum is more resistant to heat, drought, salt, and floods than other cereal crops, it is nonetheless susceptible to many stress at different crucial plant growth periods. Apart from that, moisture stress has a significant impact on crop growth, development as well as in production during, at the time of anthesis in rainfed areas.Abiotic stress is the leading cause of crop loss worldwide and decreases 50% yields of mostly all crops. (Ejeta and Knoll, 2007; Ali *et al.,* 2011).The damaging effects of water stress were more noted when it coincided with various growth stages such as germination, seedling shoot length, root length, and flowering (Tsago *et al.,* 2014). They limit photosynthesis and consequently, limited availability of photosynthetic assimilates and energy to the plant. Plants must use this limited supply of nutrients to their maximum advantage to survive under stress. For germplasm screening, another option is to use polyethylene glycol (PEG) solutions to create water stress (Khodarahmpour, 2011; Rajendran *et al.* 2011). Polyethylene glycol with a molecular mass of 6000 or more is non-ionic, water-soluble polymers that are unlikely to permeate intact plant tissues quickly. Because of the reduced osmotic potential, this solution makes it difficult for the roots to absorb water. Water stress can affect germination by delaying the initiation of germination, slowing the rate of germination, or decreasing the final percentage of germination. Sorghum is known for its moderate drought tolerance and might be used instead of agriculture in areas with little rainfall. Plant physiologists and breeders have been working to identify and understand the processes of drought tolerance in sorghum, which include a germination percentage at different levels of PEG with root and shoot traits, seed vigor, and Root: shoot ratio (Rajendran *et al.,* 2011). However, data on this moderate drought tolerance during seed germination and seedling establishment is still preliminary and ambiguous. Drought stress influences practically every step of plant growth. The negative consequences of this stress were more noticeable when it occurred throughout different phases of growth, such as germination, establishment, and flowering (Khayatnezhad *et al.,* 2010; Tsago *et al.,* 2014). Artificially created water a stress environment is used to provide the opportunity in selecting superior genotypes out

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of a large population. Based on these grounds, the experiment was carried out to categorize the sorghum germplasm against drought stress; select suitable accessions for drought tolerance, and also to determine the suitability of various seedling traits for the selection of tolerant or susceptible genotypes to drought stress.

MATERIALS AND METHODS

Twelve sorghum genotypes (DS-105, DS-178, DS-179, DS-180, DS-181, DS-182, DS-182, DS-184, DS-185, DS-186, DS-148, and DS-172) were collected from the sorghum research station, Dessa, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Banas Kantha, Gujarat, India. This investigation was performed experiment under a completely randomized design with three replications in 2018 at the Department of genetics and plant breeding, SDAU, Sardarkrushinagar, (Gujarat). Five plants of each genotype from each replication and treatment were evaluated for germination percentage (GP), shoot length (SL), root length (RL), shoot dry weight (SDW), root dry weight (RDW), root: shoot ratio (R/S) and seed vigour index (SVI). After carefully seedling germination and dividing it into roots and shoots, length-based characteristics were recorded with measuring tape. Dry shoot and root weights were obtained by placing shoots and roots in separate paper bags and drying them at 70°C for a uniform dry weight. The weight of both the average dry shoot and root was then measured.

Seeds were treated with five different concentrations of PEG-6000 (Polyethylene Glycol) 100 ml of 5%, 10%, 15%, and 20% along with control (100 ml of water). Then after the 8^{th} day's observations were recorded, germination percentage, shoot length, root length, shoot dry weight, root dry weight, seedling vigour index, and root: shoot ratio were recorded.The recorded data were subjected to analysis of variance Steel *et al.* (1997).Response of the different sorghum genotypes under water stress was statistical analysis using ANOVA in CRD design.

RESULTS AND DISCUSSION

Germination percentage

Data related to germination percentage (Table 1) were found osmotic stress induced a considerable drop in germination percentage in all genotypes, in the present investigation, the genotype DS-183 found the highest germination (100.0%) percentage as compared to the other genotypes in control.

Table 1: Effect of different levels of PEG on germination percentage of sorghum genotypes

Genotypes	Germination percentage			
	Control	5% PEG	10% PEG	15% PEG
DS-105	90.00	83.33	73.33	56.67
DS-178	96.67	86.67	70.00	50.00
DS-179	93.33	86.67	76.67	50.00
DS-180	93.33	80.00	70.00	50.00
DS-181	90.00	83.33	76.67	56.67
DS-182	86.67	73.33	60.00	40.00
DS-183	100.00	96.66	80.00	60.00
DS-184	93.33	90.00	76.66	50.00
DS-185	93.33	86.67	76.66	50.00
DS-186	90.00	76.67	66.66	43.33
DS-148	96.67	86.67	73.33	50.00
DS-172	93.33	83.33	73.33	56.67
S.Em. ±	2.72	3.04	2.72	1.92
C.D. at 5%	NS	8.88	7.94	5.62
C.V. %	5.07	6.24	6.48	6.52

Although genotypes DS-183 again was found having significantly higher germination percentage (96.6 and 60.0%) as compared to other genotypes at 5% PEG-6000 level and 15% PEG-6000 level, respectively. Genotype DS-183 remains at par with the genotypes DS-105, DS-181, and DS-172. Among 12 genotypes, genotype DS-182 recorded the lowest **V.G.**

germination percentage (40.0%) at 15% PEG-6000 treatment. At 10% PEG-6000 concentration, genotypes DS-183 and DS-182 showed the highest and lowest germination percentage as compared to other genotypes, respectively. Reduced germination percentage due to no absorption of water by seed and slow metabolic activity during germination under water stress conditions Rezende *et al.* (2017). Drought-tolerant genotypes often get the maximum germination and high survival rates. The tolerant germplasm's greater germination rates might be attributed to their ability to absorb water even under PEG-induced water stress conditions. The results of the current study were in agreement with Bobade *et al.*(2019). in wheat. This was most likely due to the low hydraulic conductivity of the environment, where PEG-6000 makes water unavailable to seeds, affecting the seed imbibition process, which is crucial for germination.

Shoot length (cm)

All the genotypes revealed a consistent pattern, namely a decrease in shoot length with increasing PEG concentrations (Table 2). Maximum shoot length was found at the control as compared to other PEG-6000 levels. At 5% PEG concentration shoot length was highest in DS-183 (1.33 cm) and DS-184 (1.28 cm) and at 15% PEG concentration genotypes DS-183 and DS-184 observed similarly highest shoot lengths of 0.40cm. The lowest length was found in genotypes DS-182 (1.28 cm) at the control condition and at 15% PEG-6000 level also same genotype DS-182 shows the lowest shoot length of 0.30 cm. Although genotype DS-183 again registered a higher value of shoot length (0.64 cm) as compared to DS-105, DS-179, DS-180, DS-181, DS-182, DS-185, DS-186, and DS-172 at 10% levels of PEG-6000.

Table 2: Effect of different levels of PEG on shoot length of sorghum genotypes

Genotypes	Shoot length (cm)			
	Control	5% PEG	10% PEG	15% PEG
DS-105	1.40	1.17	0.57	0.30
DS-178	1.60	1.23	0.60	0.30
DS-179	1.50	1.13	0.57	0.40
DS-180	1.50	1.08	0.54	0.40
DS-181	1.52	1.14	0.57	0.40
DS-182	1.27	1.03	0.53	0.30
DS-183	1.64	1.33	0.64	0.40
DS-184	1.61	1.28	0.64	0.40
DS-185	1.57	1.17	0.54	0.37
DS-186	1.37	1.07	0.57	0.30
DS-148	1.60	1.23	0.62	0.40
DS-172	1.60	1.17	0.55	0.30
S.Em. ±	0.04	0.04	0.02	0.01
C.D. at 5%	0.12	0.10	0.05	0.03
C.V. %	4.86	5.19	4.89	5.34

Root length (cm)

The data of the mean root length present in (Table3) differed significantly and revealed differences in various sorghum genotypes with different levels of PEG 6000. The root length in all the genotypes increased from 0% to 15% PEG-induced water stress. The genotype DS-183 was a statistically higher value of root length (3.23 cm) and it was at par with genotypes DS-181, DS-184, DS-185, DS-148, and DS-172 at the control condition. However, genotype DS-181 observed a higher value of root length (2.57 cm) as compared to DS-180, DS-186, DS-182, DS-178, and DS-172 and the lowest root length was found in the genotypeDS-182 (2.00 cm) at the level of5% PEG-6000. Whereas at 10% PEG 6000 level genotype DS-183 was at par with the genotypes DS-179, DS 181, DS 184, DS 185, and DS 148. At 15% PEG 6000 level DS 181 was again found significantly superior for the value of root length over to DS-105, DS-179, DS-180, DS-182, DS-185, DS-172, and DS-186 other than these genotypes showed difference non-significant with genotype DS-181. Shoot length and PEG concentration has a significantly negative correlation and positive correlation was noted between shoot length and root length as indicated that an increase in root length helps in increase of shoot length (Basha *et al.,* 2015). Baloch *et al.,* (2012) reported a significant decrease in shoot length under different PEG levels occurred due to an increased osmotic pressure of water which reduced the absorption of water and decreased external osmotic potential. The result are supported by Deshmukh and Dhumal (2011) in sorghum. Drought stress had a greater impact on hypocotyl length than on radical growth, indicating that hypocotyl length was more susceptible to drought stress. As a result, radical and hypocotyl development represent the plant's capacity to adjust to drought stress. Thus, root development is important for plant survival during a drought, and drought-tolerant plants have large root growth and reduced shoot growth. The genotypic diversity in response to water deficiency stress is indicated by the genotypic variability in the declining trend of osmotic regulation. Similar findings were reported by Kumar*et al.,* (2011).

Table 3: Effect of different levels of PEG on root length of sorghum genotypes

Genotypes	Root length (cm)			
	Control	5% PEG	10% PEG	15% PEG
DS-105	2.80	2.43	1.63	1.30
DS-178	2.97	2.37	1.53	1.40
DS-179	3.00	2.40	1.67	1.30
DS-180	2.87	2.33	1.57	1.27
DS-181	3.07	2.57	1.77	1.50
DS-182	2.80	2.00	1.43	1.17
DS-183	3.23	2.53	1.77	1.47
DS-184	3.13	2.43	1.70	1.43
DS-185	3.10	2.47	1.67	1.37
DS-186	2.90	2.33	1.53	1.27
DS-148	3.10	2.40	1.70	1.40
DS-172	3.17	2.37	1.62	1.37
S.Em. ±	0.07	0.06	0.05	0.04
C.D. at 5%	0.20	0.18	0.13	0.11
C.V. %	3.95	4.53	4.74	4.92

Shoot dry weight (mg)

The data on the mean shoot dry weight (Table 4) revealed that the genotypic differences among the different genotypes were found significant at different levels of PEG-6000. Seedling dry weight decreased continuously when osmotic stress increased and the lowest dry weight was found in 15% PEG-6000 concentration.The shoot dry weight was highest at control conditions in all genotypes ranging from 1.37 cm in DS-183 to 1.19 cm in DS-180. At 5% PEG-6000 level genotypes DS-183, DS-184, and DS-148 registered similarly higher (1.28 mg) shoot dry weight and lowest shoot dry weight (1.12 mg) observed in genotype DS-182.

Carefully inspection of values related to the shoot dry weight at 10% PEG-6000 levels revealed that genotype DS-183 again registered significantly higher shoot dry weight (0.90 mg)over genotype genotypes DS-105, DS-180, DS-182, DS-185, and DS-186 although the rest of all other genotypes were found at par with each other. At 15% PEG 6000 level genotype DS-183 (0.60 mg) once again found a significant difference in shoot dry weight it is at par with genotype DS-183 and the lowest shoot dry weight (0.49 mg) was recorded in genotype DS-182.

Under osmotic stress, seedling dry weight increased, which might be attributed to the deposition of organic and inorganic solutes as well as greater growth due to osmotic adjustment. Reduced biomass under drought stress tolerance index was observed in several plant species reported by Saenseе*et al.* (2012) in sunflower. Drought stress severely inhibited the rapid proceeding of mitotic cell division to create new biomass in the plant, leading to a reduction in shoot dry weight. The result was in agreement with the findings of Bibi *et al.,* (2012) in sorghum.

Table 4: Effect of different levels of PEG on shoot dry weight of sorghum genotypes

Genotypes	Shoot dry weight (mg)			
	Control	5% PEG	10% PEG	15% PEG
DS-105	1.29	1.20	0.83	0.55
DS-178	1.33	1.27	0.87	0.55
DS-179	1.30	1.26	0.85	0.57
DS-180	1.19	1.24	0.81	0.52
DS-181	1.35	1.25	0.87	0.57
DS-182	1.27	1.12	0.77	0.49
DS-183	1.37	1.28	0.90	0.60
DS-184	1.36	1.28	0.89	0.58
DS-185	1.35	1.24	0.81	0.55
DS-186	1.27	1.18	0.79	0.53
DS-148	1.31	1.28	0.84	0.57
DS-172	1.27	1.27	0.86	0.56
S.Em. ±	0.03	0.03	0.02	0.01
C.D.at 5%	0.10	0.09	0.07	0.04
C.V. %	4.34	4.23	4.49	4.39

Root dry weight (mg)

It was crystal clear from the perusal of (Table 5) that the genotypic differences were found statistically significant among various sorghum genotypes with different levels of PEG-6000 concentration. Under control conditions, the genotype DS-183 had significantly higher root dry weight (1.91 mg) and it was statistically at par with all the genotypes except DS-182 (1.62 mg). At 5% PEG-6000 level genotype, DS-183 again found a maximum value of root dry weight (1.93 mg) over to rest of all genotypes except genotype DS-184, and the lowest RDW (1.24 mg) was found in genotype DS-182. The genotypes DS-183 and DS-184 registered the same value of root dry weight (1.3mg) which was at par with the genotypes DS-105, DS-178, DS-179, DS-181,DS-185, and DS-172 at 10% PEG-6000 concentration. At 15% PEG-6000 level genotype DS-183 again observed a superior value of root dry weight (0.95 mg)other than genotypes except forDS-179 and DS-184 and the lowest was RDW (0.74 mg) observed in genotype DS-182. Many plant breeders have used dry root weight (DRW) as a drought tolerance selection criteria. The pattern by which various portions of the root contribute to overall water transport is a complicated character that depends on root structure, root anatomy, and the pattern by which different parts of the root lead to improved water transport. The dry weight of roots was decreased during the drought period to minimize translocation of photosynthates. Dry root weight has been utilized as a selection criterion for drought tolerance. The results were similar to the findings of Qadir *et al.* (2015) and Rezende *et al.* (2017) in sorghum.

Table 5: Effect of different levels of PEG on root dry weight of sorghum genotypes

Genotypes	Root dry weight (mg)			
	Control	5% PEG	10% PEG	15% PEG
DS-105	1.85	1.65	1.26	0.81
DS-178	1.87	1.76	1.22	0.85
DS-179	1.86	1.67	1.23	0.88
DS-180	1.80	1.56	1.18	0.75
DS-181	1.84	1.61	1.25	0.78
DS-182	1.62	1.24	1.11	0.74
DS-183	1.91	1.93	1.30	0.95
DS-184	1.88	1.81	1.30	0.89
DS-185	1.84	1.78	1.28	0.81
DS-186	1.80	1.54	1.17	0.75
DS-148	1.83	1.70	1.19	0.81
DS-172	1.84	1.57	1.22	0.80
S.Em. ±	0.05	0.05	0.03	0.02
C.D. at 5%	0.13	0.14	0.10	0.07
C.V. %	4.23	4.91	4.77	4.92

Data showed that the genotype DS-183 proved the superior value of seedling vigour index (470.6) as compared to other genotypes and the lowest SVI (351.6) found in genotypeDS-182 at the control condition. However, genotype DS-183 observed a higher value of seedling vigour index (348.0) over to rest of all other genotypes except genotypeDS-184 at 5% PEG-6000 concentration. At 10% and 15% PEG-6000 levels DS-183 found a significant higher seedling vigour index (200.1 and 112.2, respectively) over to rest of all genotypes except genotype DS-181 remains at par with DS-183 only at 15% PEG-6000 level and lowest SVI (124.4 and 58.7) was recorded in genotype DS-182 at 10% and 15% PEG-6000, respectively. The degree and percentage of seed establishment are extremely important in determining yield and maturation duration. A decrease in seedling vigour index with increasedlevels of drought has been also observed. The effect on seedling vigour index depends on the seedling length and germination percentage. Under drought conditions, germination percent decreased and seedling length has been reduced. A similar result was found by Rauf, (2008) and Rajendran *et al.* (2011) in sorghum.

Table 6: Effect of different levels of PEG on seedling viogur index of sorghum genotypes

Genotypes	Seedlingviogur index			
	Control	5% PEG	10% PEG	15% PEG
DS-105	378.00	300.34	161.56	91.77
DS-178	441.67	311.34	149.44	86.23
DS-179	420.00	306.65	171.18	85.12
DS-180	407.33	272.71	147.12	83.59
DS-181	412.77	308.40	178.85	107.88
DS-182	351.67	221.62	124.46	58.75
DS-183	470.67	348.02	200.19	112.23
DS-184	442.96	321.17	186.80	91.86
DS-185	435.33	315.02	168.57	86.67
DS-186	384.40	260.36	139.36	73.51
DS-148	454.33	314.70	177.69	98.05
DS-172	445.00	294.03	158.87	94.88
S.Em. ±	13.81	10.15	5.77	4.45
C.D. at 5%	40.31	29.62	16.84	13.00
C.V. %	5.69	5.90	6.11	8.65

Root: shoot ratio

Data revealed that the different genotypes were significantly different with different PEG-6000 levels (Table 7). Increase in root: shoot ratio was continuously recorded when increased PEG-6000 concentration from 0% to 15% and maximum value was recorded in 15% PEG concentration. In the control condition, the sorghum genotype DS-182 showed a numerically higher value of root: shoot ratio (2.2) followed by the genotypes DS-186, DS-181, DS-179, and DS-105, and the lowest ratio (1.86) found in genotype DS-178. At 5% PEG-6000 level genotype DS-181 observed a maximum root: shoot ratio (2.25)followed by genotypes DS-186 and DS-179. Carefully examining the values related to the root: shoot ratio revealed that two genotypes DS-181 and DS-185 had similar resultswith significantly higher root: shoot ratio (3.12) and it was at par with the genotypes DS-105, DS-179, DS-180, DS-183, and DS-172 at 10% PEG-6000 concentration however at 15% PEG-6000 level genotype DS-172 found the significant higher root: shoot ratio (4.49) over the rest of genotypes and the lowest ratio (3.12) was found in genotype DS-180. Plants in stress conditions often decrease biomass production and contribute more biomass to roots, maintaining a higher root: shoot ratio as an adaptation trait to confer drought resistance. High root: shoot ratio was found to increase water uptake and have a positive effect on yield under stress. Similar results of increased root shoot ratio were recorded by Queiroz *et al.,* (2019).

Table 7: Effect of different levels of PEG on root: shoot ratio of sorghum genotypes

Genotypes	Root: shoot ratio			
	Control	5% PEG	10% PEG	15% PEG
DS-105	2.00	2.09	2.88	4.01
DS-178	1.86	1.92	2.55	4.31
DS-179	2.01	2.12	2.94	3.22
DS-180	1.91	2.17	2.93	3.12
DS-181	2.01	2.25	3.12	3.71
DS-182	2.20	1.94	2.69	3.85
DS-183	1.98	1.90	2.78	3.63
DS-184	1.94	1.91	2.68	3.55
DS-185	1.98	2.11	3.12	3.80
DS-186	2.11	2.18	2.71	4.16
DS-148	1.93	1.94	2.75	3.29
DS-172	1.98	2.03	2.97	4.49
S.Em. ±	0.08	0.08	0.12	0.18
C.D. at 5%	NS	NS	0.35	0.53
C.V. %	6.37	7.06	7.29	8.32

In the present study from all the observations recorded it may be concluded that a twelve sorghum genotypes, DS-183 performed better under drought conditions as compared to the rest of the other genotypes.The perusal of the data under laboratory conditions revealed that the genotype DS-183 was marked by maximum germination percentage (100%), shoot length (1.64 cm), root length (3.23 cm) as well as highest root and shoot dry weight (1.37 and 1.91 mg per plant), seed vigour index (470.67) and root: shoot ratio (1.98) under control (0% PEG-6000) level. Increasing the concentration of PEG-6000 sharply decreased the percentage of seed germination and some other characteristics. Highest value of germination percentage (60 %), shoot length (0.40 cm), root length (1.47 cm), shoot dry weight (0.60 mg), root dry weight (0.95 mg), seed vigour index (112.23) was recorded in genotype DS-183 while highest value root: shoot ratio (4.49) was found in genotype DS-172 at 15% PEG-6000 level.

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