

Assessment of drought tolerance in various cotton genotypes under simulated osmotic settings

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Abstract

Pakistan's agriculture, especially the cotton area is facing serious threat of water shortage, which is negatively affecting the sizeable foreign reserves. Besides other irrigation management practices, selection of drought tolerant varieties can support to tackle the issue. The current study was aimed at the assessment of drought tolerance potential of various Bt cultivars of *Gossypium hirsutum* L. Under the current study, sixteen cotton cultivars were placed for germination in petri dishes under distinct osmotic potentials with seven different concentrations of PEG-6000 (i.e., 0, 5, 10, 15, 20, 25 and 27 percent, having osmotic potential of 0.0, -0.05, -0.148, -0.295, -0.491, -0.735 and -0.846 MPa respectively). The results revealed significant differences among various traits of all genotypes. It was observed that seed germination and root length increased up to concentration level of 25% PEG-6000 (at -0.735 MPa) whereas increment in shoot length stopped further. Root/shoot ratio increased until PEG concentration of 20% and then ceased. NIBGE-8 was the best performer under all simulated osmotic adjustments with maximum mean germination percentage of 62.86 %. The growth parameters of NIBGE-8 recorded on 12th and 18th days after sowing were noted as root length (6.87 and 9.9) cm, shoot length (5.9 and 6.37) cm, root/shoot ratio (1.03 and 1.23), root length-index (597 and 843) and shoot-vigor index (539 and 576) respectively. The results of study revealed that the genotypes NIBGE-8, NIBGE-9, BH-201 and RH-668 were found osmotic stress tolerant while Mubarak, CEMB-88 and DEEBEL were found highly sensitive to drought conditions.

Keywords: Bt Cotton, PEG-6000, Osmotic potential, Drought resistance

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Introduction

Cotton (*Gossypium hirsutum* L.) is the essential fiber crop of the world, highly demanded all over the world. Nearly 70 nations of the world grow cotton for their domestic use and/or for export purposes. It is grown mostly in warm as well as mild zones of the world. During 2019-20, cotton was cultivated on about 2.5 million hectares, having annual production of 9.86 million bales (480 lb, bale) with average yield of 618 kg ha⁻¹ (Economic Survey of Pakistan, 2020). Its contribution in GDP is 0.8% while its share in agriculture value addition is 4.1%. Harvesting of cotton is done by picking the cotton bolls from which seed is separated after ginning. Seed is utilized for extraction of oil and pressed seed is used for feeding livestock. Remaining fiber known as 'lint' is the entity of great importance which is further processed for the production of thread and ultimately for outfits production.

Large yield gaps exist between production and yield potentials. These yield gaps are normally due to poor cultural practices, shortage of quality seed and lack of inputs (fertilizer, insecticide and irrigation water) as well as biotic and abiotic stress especially the drastic environmental influences such as extreme drought and high temperature regimes. It is said that in future cotton will have to be grown under extreme water shortage and high temperatures in relation with other abiotic stresses (Dabbert and Gore, 2014). Cotton growing areas in Pakistan are mostly warm and arid regions having little rainfall in addition to high temperature during season of cotton (Riaz et al., 2013). Cotton yield depends upon various factors such as potential of cultivar, environmental fluctuations and overall cultural practices. These factors influence the crop individually as well as in combination, which ultimately decreases yield significantly (Romagosa and Fox, 1993).

Abiotic stresses especially drought and heat mutually creates adverse survival circumstances in the life cycle of cotton crop plant. Cotton crop has the ability to withstand under drought conditions but for higher yields it need adequate water supply of about 2,158 to 3,906 m³ every growing season (McWilliams, 2003). Thus, yield and production of cotton increases with intense rainfall pattern. Few developmental stages such as flowering initiation stage and boll developing phase require sufficient water supply (McWilliams, 2003). Plant needs water at different developmental stages during its life cycle, which critically depends on

plant water losses via transpiration and moisture quantity present in soil (Allen et al., 1998). Similarly plant developmental stage and time of irrigation as well as drought situation during cotton growing period, states the up and downs in the yield (Boman and Lemon, 2006). If drought stress remains for longer periods it effects plant height due to which short statured plant could be seen in field as compared to plant having sufficient water supply (Pace et al., 1999). Cotton boll weight, formation of seed inside the boll, seed and lint index as well as staple size, consistency, fiber development and its longevity are highly influenced under water stress circumstances (Wen et al., 2013).

Though cotton genotypes are significantly adjusted in particular environment and their selection is done after observing maximum potential in accordance to each specific desirable trait but, when it comes to shortage of water, these overwhelming challenges become hurdles in achieving maximum lint and seed yield which ultimately decreases the overall production. Survival of crop plant and increment in yield under such circumstances can be achieved by cultural and management practices such as by raising cover crops, sowing on beds, adopting zero tillage and using high efficiency irrigation practices to maintain soil moisture and tackle the water shortage at crucial stages. Besides these practices, selection of drought tolerant variety is vital to cover the gap of production. Keeping in view, these obstacles in cotton yield, the current study was planned to assess the drought tolerance potential of various Bt cotton cultivars using osmotic concentration technique.

Material and Methods

Experimental site and design

The experimental study was conducted to determine the effect of drought stress on yield and yield attributes of various cotton cultivars in the Laboratory located at the Department of Plant Breeding and Genetics, Faculty of Agriculture, Gomal University D.I Khan during the year 2017-18. The experiment was laid out under completely randomized design (CRD) having factorial arrangement with three replications.

Treatments and data recording

Sixteen cotton cultivars viz., BH-201, CEMB-55, CEMB-88, CIM-602, CIM-625, CIM-632, CYTO-179, CYTO-313, DEEBEL, FH-142, FH-152, FH-



326, Mubarak, NIBGE-8, NIBGE-9 and RH-668 were selected for assessment of their tolerance against drought.

Ten healthy, identical and lint free seeds of each cultivar were chosen. These seeds were placed for germination, one at bottom and other on top of filter papers in sterilized petri dishes to maintain concentrations at 27°C. The seeds were placed apart in each petri dish under seven different concentrations levels of PEG-6000 i.e., 0, 5, 10, 15, 20, 25 and 27% that generated osmotic potential of 0.0, -0.05, -0.148, -0.295, -0.491, -0.735 and -0.846 MPa respectively. The concentrations were applied in such a way that 2 mL of each concentration at bottom and 1 mL on upper side of filter paper were applied. Then the petri dishes were transferred to incubator having temperature set at 27 °C. In order to maintain the levels of osmotic potential each PEG-6000 concentration was applied to petri dishes after an interval of 48-72 hours. The PEG-6000 concentration required to achieve a specific osmotic potential was computed by the formula given by Michel and Kaufmann (1973) expressed as:

$$\Psi_s = -(1.18 \times 10^{-2}) \times C - (1.18 \times 10^{-4}) \times C_2 + (2.67 \times 10^{-4}) \times C \times T + (8.39 \times 10^{-7}) \times C_2 \times T$$

Where:

Ψ_s = osmotic potential (bar);

C = concentration (g L⁻¹ PEG-6000 in water);

T = temperature (°C).

For control, a solution with osmotic potential $\Psi_s = 0.0$ MPa was used.

All the treatments were observed for germination (%), shoot length (cm) and root length (cm) recorded at 12th and 18th day after sowing (DAS) and implementation of PEG-6000 concentrations. Mean values of all three replications for each of the parameters were calculated and presented in data tables. Various plants growth parameters were investigated as described below:

Germination (% age): Sprouted seed in each petri dish were considered as germinated seed and counted on 12th and 18th day after sowing. The germination percentage was computed as follows:

$$\text{Germination (\%)} = \left(\frac{\text{Number of germinated seed}}{\text{Number of seed sown}} \right) \times 100$$

Root and shoot length (cm): Five seedlings were taken out at random from each petri-dish without

disturbing the root of seedling on 12th and 18th day after sowing and the longest root was measured in centimeters from the collar to the tip and recorded. Similarly, the shoot lengths were also measured for each treatment and recorded accordingly.

Root to shoot ratio: Root lengths and shoot lengths recorded on 12th and 18th day after sowing were converted into root-shoot ratio as follows:

$$\text{Root-shoot Ratio} = \frac{\text{Root length}}{\text{Shoot length}}$$

Shoot and root vigor index: Shoot and root vigor indices were also computed at 12th and 18th day after sowing using the expression proposed by Abdul- Baki and Anderson, (1973).

Root length index (RLI) = Root length × Germination %

Shoot length index (SLI) = Shoot length × Germination %

Statistical analysis

All the data gathered were subjected to statistical analysis by following Fisher's analysis of variance method (Steel, 1997). Means were compared with LSD test at 5 % probability level as described by Gomez and Gomez (1984).

Results

All the data collected under the study were analyzed statistically. The calculated variances showed highly significant differences among the genotypes as well as the interactive effects among all the parameters investigated. These results are presented in following tables (Table-1 & Table-2).

Table-1. Analysis of variance for germination percentage

Source	DF	SS	MS	F
Genotype	15	8871.43	591.429	6.4E+30**
PEG	6	473239	78873.2	8.5E+32**
Genotype*PEG	90	6503.57	72.2619	7.8E+29**
Error	224	2.09E-26	9.31E-29	
Total	335	488614		

** means highly significant



Table-2. Mean squares for various seedling related traits

Source	DF	RL	SL	RSR	RLI	SVI
Genotype	15	40.5528**	15.4278**	0.12113**	381373**	162312**
PEG	6	2622.59**	1942.94**	78.9533**	21190000**	18510000**
Growth stage	1	1173.43**	161.171**	0.08371**	6717600**	848806**
Genotype*PEG	90	2.43476**	1.35541**	0.09697**	36570.8**	18408.1**
Genotype*Growth stage	15	0.28686**	0.82599**	0.07628**	5162.84**	4589.14**
PEG* Growth stage	6	82.382**	24.533**	7.28871**	735968**	175237**
Genotype*Growth- stage*PEG	90	0.18196**	0.32518**	0.06594**	1823.59**	1867.36**
Error	448	2.26E-30	9.95E-31	5.41E-32	1.27E-26	1.25E-26
Total	671					

** means highly significant

Germination percentage

It was observed that the germination percentage reduces as the PEG-6000 concentration increases and became zero at concentration level of 27%. Mean germination values at PEG-6000 concentrations levels of 0%, 5%, 10%, 15%, 20%, 25% and 27% which generated osmotic potentials of 0.0MPa, -0.05MPa, -0.148MP, -0.295MPa, 0.491MPa, -0.735 MPa and -0.846 MPa respectively were observed as 100%, 89.375%, 80.625%, 66.875% ,35%, 5.625% and 0% respectively (Table-3).

Table-3. Effect of different concentrations of PEG-6000 on germination %age of various cultivars

Genotype	Seed germination (%) at various PEG Concentrations							
	0%	5%	10%	15%	20%	25%	27%	Mean
BH-201	100	90	80	70	40	10	0	55.71
CEMB-55	100	80	80	70	40	0	0	52.86
CEMB-88	100	90	80	70	40	10	0	55.71
CIM-602	100	80	70	60	30	0	0	48.57
CIM-625	100	80	80	60	20	0	0	48.57
CIM-632	100	90	80	70	40	10	0	55.71
CYTO-179	100	90	80	60	20	0	0	50.00
CYTO-313	100	80	70	50	20	0	0	45.71
DEEBEL	100	100	90	80	50	20	0	62.86
FH-142	100	90	80	70	30	0	0	52.86
FH-152	100	90	80	70	40	0	0	54.29
FH-326	100	90	80	70	40	0	0	54.29
Mubarak	100	80	70	50	20	0	0	45.71
NIBGE-8	100	100	90	80	50	20	0	62.86
NIBGE-9	100	100	90	70	40	10	0	58.57
RH-668	100	100	90	70	40	10	0	58.57
Mean	100	89.4	80.6	66.9	35	5.63	0	53.93
Alpha 0.05	Standard Error for Comparison = 1.4045							

The results showed that the highest mean germination percentage under all PEG-6000 concentration was observed in the genotypes NIBGE-8 and DEEBEL having at par value of 62.86%, followed by NIBGE-9 and RH-668 with germination percentage of (58.57%), while the lowest germination percentage was shown

by CYTO-313 and Mubarak with statistically at par value of 45.71%. From these results, it is concluded that all cotton genotypes can germinate up to 20% (0.491 MPa) concentration of PEG-6000.

Root length

The results showed that increment in root length was observed until PEG-6000 concentration of 10%, beyond that level root length was reduced leading to complete cessation at concentration level of 27%. Maximum root length (12.1 cm & 14.8 cm) was observed at -0.148 MPa under both growth stages (after 12th & 18th DAS) (Table-4). Under entire concentrations of PEG-6000, NIBGE-8 was the best performer with maximum root length (9.9 cm) succeeded by NIBGE-9 (9.19 cm) while Mubarak was the least performer with minimum root length (6.26 cm).

Shoot length

Analysis of shoot length data showed inverse relationship among PEG-6000 concentrations and the shoot length in all genotypes i.e., the shoot length decreased with rise of PEG-6000 concentration. On 18th day after sowing the mean shoot lengths under all concentrations of PEG-6000 (i.e. 0, 5, 10, 15, 20, 25 and 27%) were observed as 11.98, 10.32, 8.38, 5.64, 3.28, 0 and 0 cm respectively (Table-5). Maximum mean shoot length was observed in genotype NIBGE-8 (6.37cm) followed by NIBGE-9 (6.24 cm) and lowest shoot length was recorded in genotype Mubarak (4.67 cm). The shoot length was completely ceased at 25% concentration having osmotic potential of -0.735 MPa.



Table-4. Effect of different concentrations of PEG-6000 on root length (cm) at two growth stages

Genotype	PEG-6000 concentration on 12 th DAS								PEG-6000 concentration on 18 th DAS							
	0%	5%	10%	15%	20%	25%	27%	Mean	0%	5%	10%	15%	20%	25%	27%	Mean
BH-201	8.4	9.3	13.7	9	5	0.3	0	6.53	12.2	13.6	15.7	13	5.7	0.6	0	8.69
CEMB-55	6.3	7.2	11.1	6.9	2.5	0	0	4.86	10.8	12.1	14.3	10.6	4	0	0	7.4
CEMB-88	5.3	6.5	10.1	6.1	1.9	0.1	0	4.29	9.4	10.9	13	9.1	4.5	0.2	0	6.73
CIM-602	8.2	9.1	13	8.6	4.5	0.2	0	6.23	12.1	13.5	15.5	13.1	6.7	0.5	0	8.77
CIM-625	8	8.9	12.9	8.3	4.4	0.1	0	6.09	12	13.3	15.5	12.9	7.1	0.2	0	8.71
CIM-632	7.5	8.2	12.2	8.1	4	0.1	0	5.73	11.4	12.8	15	12.6	6.3	0.2	0	8.33
CYTO-179	5.7	6.9	10.8	6.6	2.2	0	0	4.6	10.2	11.7	13.5	9.8	4.7	0	0	7.13
CYTO-313	5.9	7	11.1	6.7	2.4	0	0	4.73	10.3	11.7	13.8	10.1	5.4	0	0	7.33
DEEBEL	5.5	6.8	10.5	6.3	2	0.1	0	4.46	9.9	11.4	13.4	9.4	4.5	0.3	0	6.99
FH-142	7.7	8.4	12.3	8.2	4.1	0	0	5.81	11.4	12.9	15.1	12.4	6.2	0	0	8.29
FH-152	7.5	8.1	12	8.1	3.9	0	0	5.66	11.3	12.4	14.7	12	5.9	0	0	8.04
FH-326	7.9	8.7	12.7	8.3	4.2	0	0	5.97	11.7	13.1	15.3	12.8	6.1	0	0	8.43
Mubarak	4.7	5.8	9.9	5.4	1.2	0	0	3.86	9.3	10.5	12.7	8.7	2.6	0	0	6.26
NIBGE-8	8.7	9.9	14.3	9.4	5.4	0.4	0	6.87	12.9	14.4	17.1	14.1	9.5	1.3	0	9.9
NIBGE-9	8.5	9.5	13.9	9.1	5	0.2	0	6.6	12.5	14	16.5	13.6	8.6	0.9	0	9.44
RH-668	8.2	9.2	13.4	8.7	4.9	0.2	0	6.37	12.3	13.8	15.8	13.3	8.4	0.7	0	9.19
Mean	7.13	8.09	12.1	7.74	3.6	0.11	0	5.54	11.2	12.6	14.8	11.7	6.01	0.31	0	8.1
Alpha 0.05	Standard Error for Comparison = 0.0381								LSD = 0.1304							

Table-5. Effect of different concentrations of PEG-6000 on cotton shoot length (cm) at different growth stages

Genotype	PEG-6000 concentration on 12 th day after sowing								PEG-6000 concentration on 18 th day after sowing							
	0%	5%	10%	15%	20%	25%	27%	Mean	0%	5%	10%	15%	20%	25%	27%	Mean
BH-201	10.9	9.9	8	7.2	1.6	0	0	5.37	12.7	10.9	9	6.5	3.9	0	0	6.14
CEMB-55	9	8.7	6.9	4.5	0.9	0	0	4.29	11.6	10	8.1	5.2	3.1	0	0	5.43
CEMB-88	8.3	6.9	5.1	3.9	0.7	0	0	3.56	10.9	9.5	7.4	4.5	2.9	0	0	5.03
CIM-602	10.5	9.9	8.2	7.7	1.8	0	0	5.44	12.5	10.9	8.9	6.2	4.2	0	0	6.1
CIM-625	10	9.4	7.9	6.6	1.5	0	0	5.06	12.3	10.7	8.9	6.1	4.1	0	0	6.01
CIM-632	9.6	9.1	7.5	6.1	1.3	0	0	4.8	12	10.5	8.6	5.7	3.2	0	0	5.71
CYTO-179	8.7	8.2	6.4	4.1	0.9	0	0	4.04	11.4	9.8	7.6	4.8	2.6	0	0	5.17
CYTO-313	8.9	8.4	6.6	4.2	1	0	0	4.16	11.6	10.1	8.4	5.5	2.7	0	0	5.47
DEEBEL	8.5	8.1	6.4	4.1	0.8	0	0	3.99	11.2	9.7	7.4	4.7	3	0	0	5.14
FH-142	9.8	9.8	7.2	5.9	1.1	0	0	4.83	12.2	10.5	8.8	6	3.4	0	0	5.84
FH-152	9.7	9.7	7.1	5.7	1	0	0	4.74	12	10.2	8.5	5.7	3.1	0	0	5.64
FH-326	9.9	9.8	7.4	6.1	1.4	0	0	4.94	12.1	10.2	8.3	5.4	3.1	0	0	5.59
Mubarak	7.9	6.7	4.4	3.5	0.5	0	0	3.29	10.5	9.2	7.1	4.4	1.5	0	0	4.67
NIBGE-8	11.9	10.4	8.3	8.7	2	0	0	5.9	13.3	11.3	9.2	6.7	4.1	0	0	6.37
NIBGE-9	11.5	10.1	8	7.3	1.5	0	0	5.49	13	11.1	9.1	6.5	4	0	0	6.24
RH-668	10.4	9.8	7.3	6.2	1.4	0	0	5.01	12.4	10.6	8.9	6.4	3.7	0	0	6
Mean	9.72	9.06	7.04	5.74	1.21	0	0	4.68	12	10.3	8.39	5.64	3.29	0	0	5.66
Alpha 0.05	Standard Error for Comparison = 0.0509								LSD = 0.1743							

Root to shoot ratio

The result from data analysis of root to shoot ratio revealed that ratio was increasing with increase of PEG-6000 concentration. The maximum value of this ratio on 12th DAS and 18th DAS was observed at concentration levels of 20 % and 15% respectively. There was a decreasing trend in root to shoot ratio on 18th DAS at 20% concentration level while increasing trend at 15 % concentration level. However, on 18th DAS highest mean ratios were recorded for the genotypes NBGE-8 followed

by RH-668 while minimum ratios were observed for cultivar BH-201. It is evident from results that maximum root to shoot ratio was recorded at 20% PEG-6000 concentration on 12th DAS followed by 10% concentration and minimum ratios were recorded at 25%-27% concentrations. Highest ratios on 18th DAS were recorded at 15% concentration followed by 20% concentration while least ratios are recorded at 25-27% concentration due to ceasing of root or shoot growth at these potentials (Table-6).



Table-6. Effect of different concentrations of PEG-6000 on cotton root to shoot ratio at different growth stages

Genotype	PEG-6000 concentration on 12 th day after sowing								PEG-6000 concentration on 18 th day after sowing							
	0%	5%	10%	15%	20%	25%	27%	Mean	0%	5%	10%	15%	20%	25%	27%	Mean
BH-201	0.8	0.9	1.7	1.3	3.1	0	0	1.11	0.9	1.2	1.7	2	1.5	0	0	1.04
CEMB-55	0.7	0.8	1.6	1.5	2.8	0	0	1.06	0.9	1.2	1.8	2	1.5	0	0	1.06
CEMB-88	0.6	0.9	1.9	1.6	2.7	0	0	1.1	0.9	1.1	1.8	2	1.6	0	0	1.06
CIM-602	0.8	0.9	1.6	1.1	2.5	0	0	0.99	1	1.2	1.7	2.1	1.6	0	0	1.09
CIM-625	0.8	0.9	1.6	1.3	2.9	0	0	1.07	1	1.2	1.7	2.1	1.7	0	0	1.1
CIM-632	0.8	0.9	1.6	1.3	3.1	0	0	1.1	1	1.2	1.7	2.2	2	0	0	1.16
CYTO-179	0.7	0.8	1.7	1.6	2.4	0	0	1.03	0.9	1.2	1.8	2	1.8	0	0	1.1
CYTO-313	0.7	0.8	1.7	1.6	2.4	0	0	1.03	0.9	1.2	1.6	1.8	2	0	0	1.07
DEEBEL	0.6	0.8	1.6	1.5	2.5	0	0	1	0.9	1.2	1.8	2	1.5	0	0	1.06
FH-142	0.8	0.9	1.7	1.4	3.7	0	0	1.21	0.9	1.2	1.7	2	1.8	0	0	1.09
FH-152	0.8	0.8	1.7	1.4	3.9	0	0	1.23	0.9	1.2	1.7	2.1	1.9	0	0	1.11
FH-326	0.8	0.9	1.7	1.4	3	0	0	1.11	1	1.3	1.8	2.4	2	0	0	1.21
Mubarak	0.6	0.9	2.3	1.5	2.4	0	0	1.1	0.9	1.1	1.8	2	1.7	0	0	1.07
NIBGE-8	0.7	1	1.7	1.1	2.7	0	0	1.03	1	1.3	1.9	2.1	2.3	0	0	1.23
NIBGE-9	0.7	0.9	1.7	1.2	3.3	0	0	1.11	1	1.3	1.8	2.1	2.1	0	0	1.19
RH-668	0.8	0.9	1.8	1.4	3.5	0	0	1.2	1	1.3	1.8	2.1	2.3	0	0	1.21
Mean	0.73	0.88	1.73	1.39	2.93	0	0	1.09	0.94	1.21	1.76	2.06	1.83	0	0	1.12
Alpha 0.05	Standard Error for Comparison = 0.0229								LSD = 0.0785							

Root length index

Analysis of data on root length revealed that root length index at both growth stages was highest at osmotic potential of -0.148 MPa (10% PEG-6000 concentration) followed by -0.05 MPa (5% PEG-6000 concentration) while the least was recorded at -0.846 MPa (27% PEG-6000 concentration). Maximum mean root length index was recorded for genotype NIBGE-8 followed by NIBGE-9 while minimum root length index was recorded for genotype Mubarak at both developmental stages as well as at all concentrations of PEG-6000. At all levels of PEG-6000 (0.0, -0.05, -0.148, -0.295, -0.491, -0.735 and -0.846 MPa) mean root length index recorded on 12th DAS were 712.5, 726.81, 980.81, 521.62, 130.5, 1.18 and 0 while

1123.12, 1132.12, 1197.5, 797.62, 219.18, 3.62 and 0, respectively were recorded on 18th DAS (Table-7).

Shoot vigor index

The results of data analysis on mean shoot vigor index indicated that highest shoot vigor index was observed at both growth stages in NIBGE-8 followed by NIBGE-9 and least was recorded in Mubarak. At PEG-6000 concentrations 0, 5, 10, 15, 20, 25 and 27% mean values recorded for shoot vigor index on 12th DAS were 971.87, 812.12, 570.25, 388.5, 43.56, 0, and 0, respectively while at 18th DAS recorded mean values for shoot vigor index were 1198.12, 924.12, 677.37, 378.5, 105.75, 0, and 0, respectively (Table-8).



Table-7. Effect of different concentrations of PEG-6000 on cotton root length index at different growth stages

Genotype	PEG-6000 concentration on 12 th DAS								PEG-6000 concentration on 18 th DAS							
	0%	5%	10%	15%	20%	25%	27%	Mean	0%	5%	10%	15%	20%	25%	27%	Mean
BH-201	840	837	1096	630	200	3	0	515	1220	1224	1256	910	228	6	0	692
CEMB-55	630	576	888	483	100	0	0	382	1080	968	1144	742	160	0	0	585
CEMB-88	530	585	808	427	76	1	0	347	940	981	1040	637	180	2	0	540
CIM-602	820	728	910	516	135	0	0	444	1210	1080	1085	786	201	0	0	623
CIM-625	800	712	1032	498	88	0	0	447	1200	1064	1240	774	142	0	0	631
CIM-632	750	738	976	567	160	1	0	456	1140	1152	1200	882	280	2	0	665
CYTO-179	570	621	864	396	44	0	0	356	1020	1053	1080	588	94	0	0	548
CYTO-313	590	560	777	335	48	0	0	330	1030	936	966	505	108	0	0	506
DEEBEL	550	680	945	504	100	2	0	397	990	1140	1206	752	225	6	0	617
FH-142	770	756	984	574	123	0	0	458	1140	1161	1208	868	186	0	0	652
FH-152	750	729	960	567	156	0	0	452	1130	1116	1176	840	236	0	0	643
FH-326	790	783	1016	581	168	0	0	477	1170	1179	1224	896	260	0	0	676
Mubarak	470	464	693	270	24	0	0	274	930	840	889	435	52	0	0	449
NIBGE-8	870	990	1287	752	270	8	0	597	1290	1440	1539	1128	475	26	0	843
NIBGE-9	850	950	1251	637	200	2	0	556	1250	1400	1485	1088	344	9	0	797
RH-668	820	920	1206	609	196	2	0	536	1230	1380	1422	931	336	7	0	758
Mean	713	727	981	522	131	1.19	0	439	1123	1132	1198	798	219	3.63	0	639
Alpha 0.05	Standard Error for Comparison 3.8114								LSD 13.053							

Table-8. Effect of different concentrations of PEG-6000 on cotton shoot vigor index at different growth stages

Genotype	PEG-6000 concentration on 12 th DAS								PEG-6000 concentration on 18 th DAS							
	0%	5%	10%	15%	20%	25%	27%	Mean	0%	5%	10%	15%	20%	25%	27%	Mean
BH-201	1090	891	640	504	64	0	0	456	1270	981	720	455	156	0	0	512
CEMB-55	900	696	552	315	36	0	0	357	1160	800	648	354	92	0	0	436
CEMB-88	830	621	408	273	28	0	0	309	1090	855	592	315	76	0	0	418
CIM-602	1050	792	574	462	54	0	0	419	1250	872	623	372	105	0	0	460
CIM-625	1000	752	632	396	30	0	0	401	1230	856	712	366	68	0	0	462
CIM-632	960	819	600	427	52	0	0	408	1200	945	688	399	120	0	0	479
CYTO-179	870	738	512	246	18	0	0	341	1140	882	608	288	40	0	0	423
CYTO-313	890	672	462	210	20	0	0	322	1160	808	588	275	54	0	0	412
DEEBEL	850	810	576	328	40	0	0	372	1120	970	376	100		0	0	462
FH-142	980	882	576	413	33	0	0	412	1220	945	704	420	102	0	0	484
FH-152	970	873	568	399	40	0	0	407	1200	918	680	399	124	0	0	474
FH-326	990	882	592	427	56	0	0	421	1210	918	664	378	112	0	0	469
Mubarak	790	536	308	175	10	0	0	260	1050	736	497	220	30	0	0	362
NIBGE-8	1190	1040	747	696	100	0	0	539	1330	1130	828	536	205	0	0	576
NIBGE-9	1150	1010	720	511	60	0	0	493	1300	1110	819	455	160	0	0	549
RH-668	1040	980	657	434	56	0	0	452	1240	1060	801	448	148	0	0	528
Mean	972	812	570	389	43.6	0	0	398	1198	924	677	379	106	0	0	469
Alpha 0.05	Standard Error for Comparison 3.8569								LSD 13.209							

Discussion

The current research focused to investigate the drought tolerance in various genotypes of cotton. The data collection and analysis on various plant growth

parameters comprised on germination percentage, root and shoot lengths, root to shoot ratio and vigor indices. These parameters were statistically analyzed, and explored thoroughly in comparison with each other and among their interactions with a wide range of



osmotic potentials. It was found that the PEG-6000 concentration showed inverse relationship with germination percentage i.e., as concentration increases germination decreases. Drop in germination percentage was due to water stress, which alters the cell function and growth. Xue-yan et al., (2008) found that cellular extension and carbohydrates wall production highly altered and inhibited due to water stress. Water stress ultimately decreases cell enlargement due to turgescence reduction (Shalhevet et al., 1995). The concentrations of 25-27% creates fatal osmotic potential for germination upon which germination stops (Sidari et al., 2008; Khodarahmpour, 2011; Babu et al., 2014; Megha and Mummigatti, 2017). Tsaliki et al. (2019) and Jatoi et al. (2014) also revealed that sprouting of cotton genotypes decreased under increased drought intensity with application of PEG-6000. Lesser PEG applied greater will be germination as adverse effect upon germination is dependent upon the proportion of drought intensity and duration in addition to PEG-6000 used.

It was observed that enlargement in the root length continued until PEG-6000 concentration reached at the level of 10%. Sakthivelu et al. (2008); Khodarahmpour (2011) and Jatoi et al. (2014) also reported decrease in root length under various water deficit conditions. This can be due to the reason that under osmotic stress plant separately execute additional photosynthesis for the enlargement and development of root instead of shoots. It facilitates plant to acquire moisture through deep penetration into the soil whereas reduced shoot size decreases transpiration rate (Tonin et al., 2000; Maruti and Katageri, 2015; Megha and Mummigatti, 2017). Long roots play important role in provision of water to plant by extracting water from the deep zone of soil as long roots are found to withdraw more water per unit length of root from moist ground and withdrawal of water reduces with the reduction of soil water potential (Landjeva et al., 2008 and Babu et al., 2014).

Shortening of shoot length could be due to the fact that under drought stress, plant tends to get moisture from the deep zone of soil, for which root size, number of roots, mass of root and adjacent roots became large and expanded which causes ultimate reduction in shoot length. These results are in complete agreement with the findings of Landjeva et al.,(2008); Sakthivelu et al. (2008); Khodarahmpour (2011); Babu et al. (2014) and Megha and Mummigatti (2017) who observed that shoot biomass decreases due to increase

in root length volume, weight and lateral roots in search of moisture from deep soil layers. Declined length of shoot decreases transpiration rate due to decrease in surface area for water loss (Babu et al., 2014). Xue-yan et al. (2008) discovered that evaluation and selection of cotton genotypes can be carried out easily and rapidly for drought tolerance by the modification of osmotic conditions by means of PEG-6000. He subjected some cotton genotypes to artificial drought stress conditions for 12 hours by utilizing different concentrations of PEG at sprouting-, seedling-, cotyledon- and leaf formation stages. He observed varied amounts of osmotic stress tolerance and found that shoot development as well as 3-6 leaves formation phases were very crucial with respect to osmotic stress tolerance.

Higher ratios might be due to ultimate increment in mass of roots, which enabled the plant roots to extract more water due to increased photosynthetic activity of plant for the development of higher root biomass. Lower shoot length and biomass assisted in the prevention of higher water losses by decreasing transpiration rate per unit area of shoot. They may change to maintain existence under osmotic stress conditions instead of having contribution in yield (Khodarahmpour, 2011; Babu et al., 2014 and Megha and Mummigatti, 2017). Meneses et al. (2011) reported that osmotic potential below -0.4 MPa have drastic effects upon seed viability and seedling vigor. Likewise earlier studies; on cotton by Michel and Kaufmann (1973), on cowpea by Ogbonnaya et al. (2003) and on wheat by Landjeva et al. (2008) revealed that genotypes, which were tolerant to drought stress circumstances attains higher root to shoot ratio as compared to the susceptible cultivars. Megha and Mummigatti, (2017) narrated that root length index decreases as osmotic potential increases by using PEG-6000 in susceptible genotypes but shoot vigor index showed inverse relationship with PEG-6000 concentration. The reduction in shoot vigor index is probably due to lengthy root and shoot; smaller number of leaves and reduced seedling length. Xue-yan et al. (2008) also reported the similar results.

Conclusion

From the current investigation, it is concluded that various cotton varieties showed different behavior against stress. Some varieties had significant tolerance against drought stress generated by PEG-6000 at some levels. Water deficiency highly effects the survival of



seed and seedling development at different osmotic conditions. On basis of findings of this study, it is concluded that genotype NIBGE-8 was highly osmotic stress tolerant whereas cultivar Mubarak was highly sensitive to water stress. Furthermore, the results of the study revealed significance of PEG-6000 as synthetic stress inducer for quick evaluation and screening of drought tolerant cotton genotypes that can play key role in cotton breeding activities.

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Contribution of Authors

Gondal MR: Planned and managed the study, supervised data collection and analysis
Saleem MY: Conceived idea, designed research methodology, data collected and first draft write up
Rizvi SA: Interpreted data and final editing & approval of manuscript
Riaz A: Literature review and manuscript write up
Naseem W & Muhammad G: Literature review, data analysis and interpretation
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