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Early response of the cotton (*Gossypium hirsutum*) genotypes against drought stress

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Abstract

Cotton is an important cash crop of almost all developing countries including Pakistan. It is backbone of textile industry. Cotton yields are stagnant for the last several years due to a number of factors including: lack of agronomic practices, shortage of rain, biotic and abiotic stresses. Drought stress, poses the most important constraint to plant survival and crop productivity. It is responsible for shedding of small squares on large scale, resulting in a decrease in flowering. The main objective of this study was to evaluate the response of cotton genotypes at seedlings stage against drought stress. The experiment was conducted under Randomized Complete Block Design using factorial arrangements with three replications. The treatment plant of the experiment included two factors genotype (eleven) and drought levels (control, 50% drought and 75% drought stress). After 50 days the data was recorded on the seedling parameters (root and shoot length, root and shoot fresh & dry weight, relative water contents, excised leaf water loss). It was found that DPL-70010N, DPL-2775 and A-8100 were drought tolerant in both conditions. It's important to note, that these genotypes are non-Bt, so it can be concluded that non Bt varieties are comparatively more drought tolerant as compared to the non-Bt and this is useful information for cotton breeding programs for drought tolerance.

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Introduction

Cotton is an important cash crop of Pakistan and other developing countries. It is also known as white gold. Since ancient times humans have cultivated cotton plants for fiber collection, clothing and other byproducts i.e. cocking oil and animal khal. Cotton is responsible for livelihood of many families around the globe (Baffes, 2004). Its seed is used to get cottonseed oil (Khalid *et al.,* 2011). Cotton is supposed to be the backbone of Textile industry (Khan *et al.*, 2009).

Grown in more than 100 countries, cotton is a heavily traded agricultural commodity, with over 150 countries involved in exports or imports of it. Cotton yields are stagnant for the last several years due to a number of factors including: lack of agronomic practices, shortage of rain, biotic and abiotic stresses. Even in recent years cotton is showing negative trend with respect to its production and further usage (Ali *et al.,* 2012).

During the life period of a plant, it passes through many biotic and abiotic stresses. Among abiotic stresses, drought and salinity are very important. Salinity in topsoil, subsoil and drought is one of the major abiotic environmental stresses to crop production (Soomro *et al.,* 2011). One third population of the world is suffering due to shortage of water (Noorka *et al.,* 2013). Agricultural drought is the deficiency of sufficient moisture required for a typical plant development and growth to complete the life cycle (Manivannan *et al.,* 2008). Water scarcity, poses the most significant constraint to crop survival. Drought severely affects crop development with substantial decreases in crop growth rate and biomass accumulation. The main consequences of drought in crop plants are reduced cell expansion, cell division, leaf size, stem elongation and root proliferation.

It also disturb stomatal oscillations, plant water and nutrient relations resulting in diminished crop productivity, and water use efficiency (Farooq *et al.*, 2009). Drought causes impaired growth and development of cotton plants (Cakir, 2004; Pettigrew, 2004). It causes disturbance in the structural root parameters like root number, volume and density which ultimately affects aerial plant parts (Cahn *et al.,* 1989). It also causes reduction in leaf size, leaf number and its function (Rucker *et al.,* 1995). Shoot and root lengths are seedling growth traits which are also negatively affected by drought stress (Khodarahmpour, 2011). The plant water relation is maintained by traits like relative water contents, transpiration rate and water potential, any imbalance in one of negatively affects plant water relation (Anjum *et al.,* 2011).

Stress tolerance is genetically controlled and linked with different morphological and physiological traits of crop plants (Singh, 2004). There is need to breed drought tolerant cultivars (Farooq *et al.*, 2009; Mahajan and Tuteja, 2005 and Yordanov *et al.*, 2000). The main objective of this study was to evaluate the response of cotton genotypes at seedlings stage in drought stress. These data could then be used to analyze the susceptibility or tolerance of a cultivar to drought's stress.

Materials and methods

Experimental site and Plant Material

Present study was done in the green house of Department of Plant Breeding and Genetics, University of Agriculture Faisalabad. The plant material was comprised of eleven cotton (*Gossypium hirsutum*) genotypes (Maria Delmar, DPL-26, A-8100, LA-OKRA, HA-118, BT-122, CULTURE-605, DPL-2775, DELCOT-226, DPL-70010-N and BT-IR-3701) both Bt and non-Bt genotypes. Polythene bags (6 inches × 4 inches size) were filled with 1.3kg of soil in each bag. The physical properties of the soil were; EC 3.74, pH 7.88, TSS 37.4 and SAR 6.39. Three seeds were sown in each bag at depth of 2cm. After seedling emergence, thinning was practiced and left with only one healthy seedling per bag.

Treatments

The experiment was conducted under Randomized Complete Block Design with factorial treatment arrangements (two factor factorial with genotype and Drought levels). Three water levels including control (normal irrigation), 50% of field capacity and 75% of field capacity were used. All the bags were watered equally till the emergence of first true leaf.

At emergence of first true leaf, both drought treatments were applied to each set. Drought was maintained up to 50 days of seedling emergence.

Data Collection

After 50 days root (RL) and shoot length (SL) were measured with measuring tape. Other parameters being measured were fresh root (FRW) and shoot weight (FSW), dry root (DRW), shoot weight (DSW) number of nodes (NON) and number of leaves (NOL). Also relative water contents (RWC) and excised leaf water loss (ELWL) were measured using following formulas.

RWC = ((Fresh leaf weight – Dry leaf weight)/ (Turgid weight - Dry leaf weight)) × 100

ELWL = (Fresh Leaf Weight – Wilted Leaf Weight)/ Dry Leaf Weight

Statistical Analysis

Analysis of variance (Stell *et al.*, 1980) using Gen Stat 10th edition was done to check the effect of treatment and genotypes on plant parameters. Biplot was also drawn to check genotype-trait-treatment interplay usig Gen Stat 10th edition (Yan *et al.*, 2007).

Results

Analysis of Variance

Analysis of variance showed high variation for root length, shoot length and number of nodes for drought levels and non-significant with respect to genotypes and genotypes to drought levels interaction. Whereas fresh shoot weight, dry shoot weight, fresh root weight, dry root weight, relative water content and excised leaf water loss showed significant differences for drought levels, genotypes and their interaction. An exception to the above results was observed in number of leaves where both genotypes and drought levels were significant but their interaction was non-significant.

Biplot Analysis for identification of drought tolerant and susceptible genotypes.

Under control conditions

In biplot two things are important one is the vector length of the trait and the second is cosine angle among the traits. The longer the vector length the more discriminatory is that trait and vice versa. In case of cosine angle the smaller the angle between the two traits the more linked the both traits are and vice versa. DRW is important trait under control conditions as is evident from the longest vector length followed by FRW and RL. DRW, FRW and RL.

These traits have more promontory effect on plant performance under control conditions as these lies in the first quadrate. Although SL, NON, FSW, DSW and NOL are also having positive effect on plant performance but their contribution is lower as compared to the above mentioned traits. Whereas ELWL and RWC have negative association with the plant performance under control conditions. Table 1.

Table 1. Combined analysis of variances for traits under consideration, of Cotton Genotypes

S.O.V	D.F	RL	SL	NON	NOL	FSW	DSW	FRW	DRW	RWC	ELWL
Treat.	2	286.099**	548.884**	78.9242**	94.4091**	90.1692**	6.7144**	98.8847**	5.1980**	10941.4**	13.2294**
Gen.	10	53.487^{NS}	6.533^{NS}	2.4273^{NS}	4.1121**	2.7943**	0.2047**	5.9813**	0.3751**	78.9**	0.8279**
Treat × Gen	20	29.636^{NS}	6.619^{NS}	1.5909^{NS}	2.2758^{NS}	1.3659**	0.1527**	6.1264**	0.2132**	127.5**	1.1676**
Error	33	22.475	3.759	1.2879	1.2121	0.0564	0.0103	0.0530	0.0041	8.3	0.0596
C.V%		19.05	12.34	17.38	22.43	9.03	13.84	9.22	10.83	7.29	11.88

Whereas RL stands for root length, SL for shoot length, FSW for fresh shoot weight; DSW for dry shoot weight, FRW for fresh root weight, DRW for dry root weight, RWC for relative water contents, ELWL for excised leaf water loss, Treat. for treatment; Gen. for genotype; CV for coefficient of variation. NS for non-significant, and ** for highly significant at $p<0.01$.

While describing the trait genotypes association it is important to note that genotypes which are present away from the origin in the positive direction of a discriminatory trait are performing best with respect to that trait whereas genotypes which lies away from the origin in the opposite direction of a discriminatory trait are performing low. MARIA-DELMAR, DPL-26, DPL-700010-N and BT-TR-3701 showed good growth.

In comparison performance of LA-OKRA, HA-118, BT-122, DECLCOT-226, CULTURE-605 and DPL-2775 was low respectively (Fig 1). Looking at the trait and genotype association it was observed that MARIA-DELMAR, DPL-26, A-8100 showed high scores for RL, FRW and DRW. DPL-70010-N and BT-TR-3701 showed high scores for SL, NON, FSW, NOL and DSW. LA-OKRA and HA-118 have high scores for RWC. DPL-2775, CULTURE-605, DELCOT-26 and BT-122 have high ELWL. RL, DRW and FRW have strong positive association with each other as these lies close together. Similarly SL, NON, FSW, DSW and NOL have positive association among themselves. RWC and ELWL have positive association with each other and have negative association with rest of the traits as is evident from Fig. 1.

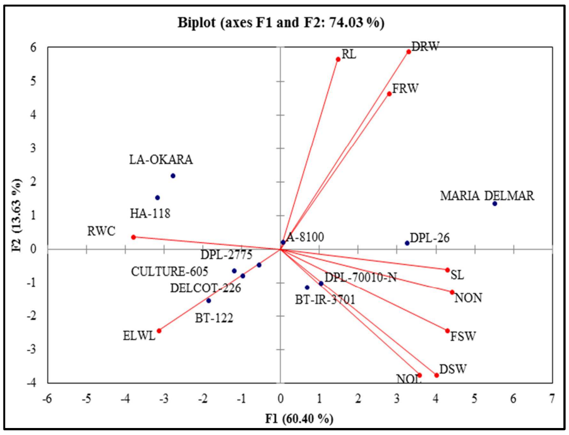

Fig. 1. Biplot analysis under control conditions.

Under 50% drought stress of field capacity

RWC and all other plant attributes contributed positively for plant survival under 50% drought stress except NOL and RL and ELWL. Vectors of NOL and RL lie very close to the origin hence their contribution is doubtful and non-reproducible. Whereas ELWL showed negative association with all other traits as its vector lies opposite to the vectors of other traits. RWC has maximum positive contribution as to the plant survival under 50% drought stress as is evident from the Fig 2. Among genotypes MARIA-DELMAR, DPL-2775, CULTURE-605 and DPL-26 are showing stress tolerance. On contrary A-8100, LA-OKRA, DELCOT-26, BT-4R-3701, HA-118 and BT-122 are susceptible and could not tolerated 50 % drought stress.

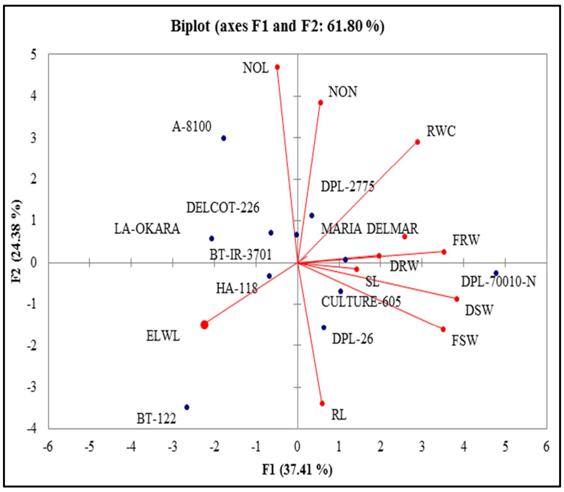

Fig 2. Biplot analysis under treatment 1 (50% drought of field capacity).

Those genotypes which are not performing better under 50% drought also have negative association with almost all the plants traits except ELWL. As all these genotypes lie opposite to the vector of FRW, DSW, SL, FSW and RWC.

Whereas genotypes which showed better drought tolerance have positive association with all these traits and lies in the same quadrate of the biplot i.e. DPL-2775 showed high score for RWC, MARIA-DELMAR for DRW and FRW, DPL-0010-N for SL and DSW, CULTURE-605 and DPL-26 for FSW. HA-118 and BT-122 showed high ELWL hence are prone to drought stress. These genotypes also showed low RWC, DRW, DSW, FRW, FSW and SL.

Under 75% drought stress of field capacity

Under 75% drought stress RL, DRW, RWC and FRW traits are contributing positively to the plant survival. However NOL, ELWL, FSW, DSW and SL showed negative association with plant performance under 75% drought stress.

Whereas NON did not show clear positive or negative association with either side. From the Fig. 3 it is evident that RL, DRW, RWC and FRW have positive association among themselves and negative association with other traits. Similar NOL, FSW, SL and DSW have positive association with one another whereas negative association with other traits.

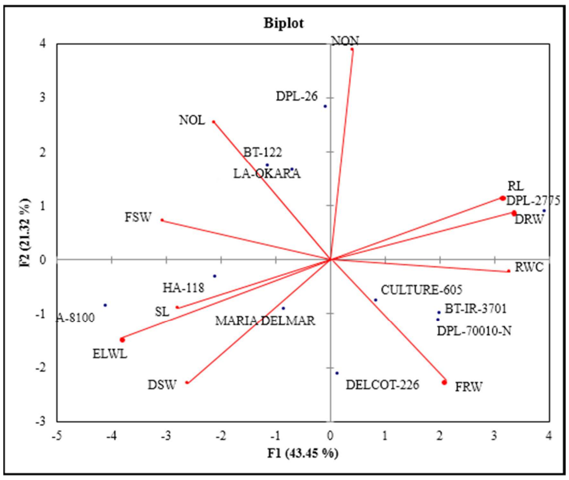

Fig 3. Biplot analysis for treatment 2 (75% drought of field capacity).

While talking about the trait genotypes association it was observed that under 75% drought stress BT-122, LA-OKRA, HA-118, A-8100 and MARIA-DELMAR showed very poor performance in comparison to CULTURE-605, BT-IR-3701 and DPL-70010-N which gave good performance. Whereas DPL-26 and DELCOT-226 showed intermediate drought tolerance as these lie very close to the origin in Fig. 3. With respect to traits genotypes association it was observed that DPL-2775, CULTURE-605, BT-IR-3701, DPL-70010-N and DELCOT-226 showed high scores for RWC, FRW, DRW and RL. MARIA-DELMAR showed high score for DSW, HA-118 and A-8100 for SL and ELWL, LA-OKRA, BT-122 and DPL-26 for NOL and FSW.

Discussion

Analysis of variance showed significant difference among genotypes for most of the traits showing high level of variability except RL, SL, NON and NOL. It indicated that these sets of genotypes did not exhibit much variation for these traits. Further the treatments varied significantly for all traits which indicated that the treatments have influenced the genotypes for almost all the traits. Treatments and genotypes interaction also influenced majority of the traits except four RL, SL, NON and NOL. Hence there is no variation for root length, shoot length, number of nodes and number of leaves in these set of cotton genotypes. Whereas maximum variation for fresh shoot weight, dry shoot weight, fresh root weight, dry root weight, relative water content and excised leaf water loss (Table 1).

These results indicated that genotypes present in this study have narrow genetic makeup with respect to root length, shoot length, number of nodes and number of leaves. Whereas genetic regions controlling fresh shoot weight, dry shoot weight, fresh root weight, dry root weight, relative water content and excised leaf water content are not alike across genotypes. Basal *et al.* (2005) also reported variation existed in for root and shoot related traits i.e. fresh root weight, fresh shoot weight, dry root weight and dry shoot weight along with RWC and ELWL. Khalid *et al.* (2011) also reported that fresh and dry, shoot and root weights, relative water contents and excised leaf water loss are related to drought tolerance.

Genotype-trait-environment interplay was also studied using Biplot analysis under drought as well as normal conditions. As biplot is an efficient way to study the trait genotype association and to study a large number of environments, genotypes and traits together. It gives us best information while dealing with multi trait, multi genotype and multi environment data (Yan *et al.*, 2007).

Under control conditions root related parameters contributed more to the plant growth and survival. As good root growth is necessary for establishment of crop stand and root helps in uptake of water and mineral elements necessary for plant growth and plays role in drought tolerance (Brunner *et al.*, 2015). Excised leaf water loss and relative water contents are stress related traits, under normal condition plant does not have to make these adjustments. Hence under normal conditions ELWL and RWC showed negative association with all other plants traits and plant growth (Fig 1). Because assurance of least excised leaf water loss and maintenance of the high relative water contents are energy consuming process and plant have to make these adjustments at the cost of growth and development (Siddique *et al.*, 2000). Drought tolerance based on reduced growth in different characters of cotton plant under water deficit conditions (Pettigrew, 2004; Khalid *et al.*, 2011) have been used to screen drought tolerant lines under stress conditions.

Drought stress shifts the normal development process of a plant towards stress tolerance. Stress tolerant plants tries to adjust their solute concentration to adjust their solute-water balance in the cell to maintain cell's turgor pressure (Snowden *et al.,* 2014). Under 50% drought stress overall growth reduction was observed as compared to the control treatments however some genotypes i.e. DPL-70010-N, MARIA-DELMAR and CULTURE-605 showed better growth. These genotypes recorded good fresh shoot weight, dry shoot weight, fresh root weight and better relative water contents. These traits helped them survive better under drought stress whereas other genotypes i.e. A-8100 and LA-OKRA did not have these adjustments and hence could not tolerated drought stress (Fig 2).

Under 75% drought stress DPL-70010-N, BT-IR-3701 and CULTURE-605 genotypes showed better survival as compared to the other genotypes and MARIA-DELMAR, A-8100 and H-118 showed drought sensitive behavior (Fig 3). It was observed that root linked traits i.e. root length, fresh root weight, dry root weight were contributing more to drought tolerance as compared to the shoot related traits which were more associated with drought sensitive behavior (Fig 3). Previously Basal *et al.* (2005) also reported that root related traits are more important for drought tolerance. Also Hassan *et al.* (2015) reported that the root length is more important traits in drought tolerance as roots help plants to uptake water and nutrients essential for its growth. Also roots helps to maintain water balance by uptake of water from the soil. Hence deep root system favors plant to cope drought stress.

Relative water content is used as a measure to differentiate drought tolerant and drought sensitive cultivars (Parida *et al.*, 2008; Ahmad *et al.*, 2009). Excise leaf water loss is also used a selelction criteria for drought tolerance (Basal *et al.,* 2005). Relative water content and water loss in excised leaf showed that varieties/lines differed from each other and some of them were distinctly more tolerant than others. Under 50% and 75% drought stress both traits (RWC and ELWL) were oppositely linked to each other as the vector direction for both traits was opposite in the biplot (Fig 2 & Fig 3). It indicated that genotypes having high relative water contents showed least excised leaf water loss and vice versa. Hence drought tolerant genotypes DPL-70010-N and CULTURE-605 showed good relative water contents and low excised leaf water loss. Similar drought sensitive genotypes showed more excised leaf water loss and low relative water contents i.e. MARIA-DELMAR, A-8100 and HA-118 (Fig 2 & 3).

Conclusion

The study concluded that for selection under normal conditions growth related attributes i.e. root length, shoot length, root fresh weight, shoot fresh weight, root dry weight and shoot dry weight should be focused while for drought tolerance root related traits, relative water contents and excised leaf water loss should be focused. Biplot is favorable approach to study traits, genotypes and environments interaction simultaneously. Further DPL-70010-N and CULTURE-605 were found drought tolerant lines whereas A-8100, MARIA-DELMAR and HA-118 were drought sensitive lines.

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Authors contribution

MSI (conduction of research work and paper write up), GMT (conduction of research work and data analysis), RS (guided for planning and implementation of experiment, data analysis and paper write up and correspondence of paper), SJ (data analysis and paper write up) SJ (data analysis), MA (Paper write up), MZI (Supervision of Work).

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