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GENETIC VARIABILITY IN COTTON FOR WATER-DEFICIT TOLERANCE

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Increasing water shortage for irrigation is a major constrain to sustainable cotton production. Cotton plant observes reduction in seed cotton yield as well as fibre quality when subjected to drought conditions. Physiological as well as agronomic traits provide useful information for water-deficit tolerance. To find out genotypes having better drought tolerance, 45 genotypes of *Gossypium hirsutum* L. were studied under two moisture levels i.e. well water and water-deficit stress conditions for two years (2013 and 2014). The experiment was conducted using split plot design under RCBD. All the genotypes behaved differently under both control and water-deficit stress. Genotype × Environment Interaction (GEI) of cotton genotypes with two water levels (Environments) were studied for some selected agro-physiological traits i.e. water potential, osmotic potential, pressure potential and seed cotton yield using AMMI biplot analysis. Results showed that the genotypes VH-291, FH-329, FH-153, IR-6, FH-159, VH-289, FH-322, MNH-886, S-15 and FH-207 are either stable or showing positive interaction with water deficit conditions for most of the traits under studied. These genotypes can be used in further breeding program for developing varieties suitable for cultivation under water deficit conditions whereas; NS-131, AA-703 and KZ 191 interacted undesirably with water-deficit stress.

Keywords: *Gossypium hirsutum* L., breeding, seed cotton yield, abiotic stress, turgor potential.

Abbreviations: SCY: Seed cotton yield, WP: Water potential, OP: Osmotic potential, PP: Pressure potential, S: Stress, NS: Non-stress

INTRODUCTION

Water scarcity is increasingly becoming a serious threat to the world's economy. Most of the countries across the world are facing problems due to water shortage. Response of a plant to drought stress condition depends upon the stage of the growth and environmental conditions (Cattivelli *et al*., 2008). Furthermore, responses to drought stress are tremendously different conferring to the genetic architecture of a plant. Water quality as well as availability affect the physiological processes and growth of all plants, as water is the major constituent of vigorously growing plants, it ranges from 70-90% of plant fresh weight (Gardner *et al*., 1983). In general, drought stress is the condition where the water and turgor potential of the plant are reduced sufficiently to inhibit normal functioning of the plant (Hsiao, 1973). Cotton is the most important fiber crop in world but its production is fluctuating due to various biotic and abiotic stresses. Among the abiotic stresses, water-deficit stress is accepted as the most devastating cause which limits the fibre yield as well as quality in cotton (Saleem *et al*., 2015). Drought tolerance mechanism in plants is genetically controlled and is linked with many physiological and morphological traits (Singh, 2004). Numerous studies have been conducted on several drought related biochemical, physiological as well as agronomic traits for different plant species and have been suggested as selection criteria in plants for water-deficit tolerance (Brito et al., 2011; Rahman *et al*., 2008). The requirement for this success involves determination of the degree of genetic variability within a plant species for these parameters and their comparative impact on economic yield (Cooper, 1999). When cotton plants are exposed to water-deficit stress, the leaves showed big decline in leaf water potential and relative water contents (Nayyar *et al*., 2006). Thus, a greater leaf relative water content, lower transpiration rate and lesser excised leaf water loss have been accepted as selection criteria for breeding plants against drought stress conditions (Rahman *et al*., 2000). Numerous biometrical methods including univariate as well as multivariate have been developed to measure the variability among genotypes (Akcura *et al*., 2005). Amongst, the most extensively used is AMMI (additive main effects and multiplicative interactions) model's PCA1 and PCA2 (Principal Components 1 and 2 respectively) scores for each genotype (Gauch *et al.*, 2008). Keeping in view the fluctuating environmental conditions, the current study was planned to govern the genetic variability existing amongst cotton germplasm linked with water-deficit tolerance.

MATERIALS AND METHODS

The plant material was comprised of 45 Bt upland cotton (*Gossypium hirsutum* L.) genotypes. Seed of these genotypes was collected from their respective research stations/institutes in Pakistan. The seed of some genotypes were also provided by Department of Plant Breeding and Genetics, University of Agriculture Faisalabad, Pakistan. The experimental work was conducted at research area of plant physiology section, Ayub Agricultural Research Institute, Faisalabad, Pakistan. These genotypes were studied under two moisture levels i.e. well-watered and water-deficit stress during two years 2013 and 2014. During the year 2013, total irrigation water applied was 609 mm and 307 mm to well-watered and water-deficit conditions respectively and additional moisture of 245 mm was received in the form of precipitation. During the year 2014, the total irrigation water applied to well water and water-deficit condition was 563 mm and 290 mm respectively while precipitation received was 389.7 mm in the form of rain. The sowing dates were 24th March and 8th April during 2013 and 2014 respectively. The experiment was planted in split plot design under RCBD with three replications. Moisture levels were kept in main plots and genotypes in subplots. In each replication there was one row for each genotype. The distance between row to row and plant to plant were kept at 75 and 30 cm respectively. The distance between stress and non-stress plots was 100 cm while between different replications of a plot were 90 cm. There were ten plants in each row. Recommended agronomic practices were applied during both seasons. Data were recorded for seed cotton yield (g), water potential (Ψw), osmotic potential (Ψs) and pressure potential (Ψp). Seed cotton was picked at 180 days after sowing and was dried under sunlight for one day and weighed. Physiological traits were measured on the fully expanded young leaf at 40-45 days after the 1st irrigation, when all the genotypes were at least 50% flowering. Leaf water potential (Ψw) was measured with a pressure chamber (Model 600, Pressure Chamber Instrument, PMS International, UK) adopting the method described by Scholander *et al.* (1964). The leaf lamina was sealed off in the pressure chamber and pressure was applied using nitrogen cylinder till free sap was visible at the protruding end of the petiole. This counteracting pressure was regarded approximately equal to water potential of the leaf tissues and also to the tension which originally existed in the xylem sap. Leaf samples (flag leaves) were collected between 6.00 and 9.00 a.m. (to avoid evaporation losses) and leaves were placed in the pressure chamber as quickly as possible. The leaves used to measure the water potential were frozen in a freezer (-20°C) and later they were thawed and cell sap was collected by pressing the leaf tissue with the help of a glass rod. Cell sap was collected in Eppendorf tubes and a drop of sap was used directly to measure osmotic potential using cryoscopic osmometer (Osmomat 030-D, Cryoscopic osmometer printer, Genatec). The pressure potential was calculated by the formula given by Hopkin (1999) as the difference between water potential and osmotic potential values.

Pressure potential (Ψp) = water potential (Ψω) - osmotic potential (Ψs)

***Statistical analysis*:** Data were evaluated for significance differences among treatments, genotypes and treatments × genotypes using analysis of variance described by Steel *et al*. (1996). AMMI biplot analysis was used for studying GEI as it was given by Gauch *et al*., (2008). For conducting the AMMI biplot, year-treatment combinations were considered as four environments i.e NS2013 and NS2014 for well-watered while S2013 and S2014 for water-deficit condition during 2013 and 2014, respectively. The analysis were performed using R software.

RESULTS

Analysis of variance (Table 1) indicates significant differences among genotypes, treatments and genotype × environment interaction (GEI). In AMMI biplot, distance from origin (0, 0) indicates the interaction of genotypes with environments.

Table 1. Degrees of freedom and mean squares for environment and genotype effects on agro-physiological traits.

SOV	DF	SCY	WP	OP	PP
Env	3	54866.365**	24.217**	30.498**	0.629**
Rep (Env)	8	353.474*	0.007*	0.025*	0.020*
Gen	44	6116.597**	1.076**	1.168**	0.130**
Gen:Env	132	1193.368**	0.476**	0.511**	0.166**
Residuals	352	31.146	0.006	0.022	0.030

SCY: Seed cotton yield, WP: Water potential, OP: Osmotic potential and PP: Pressure potential of upland cotton.

***Seed cotton yield*:** Biplot for SCY among 45 genotypes under four different drought environments was plotted between PC1 and PC2 which contributed 97.7% of total interaction (Fig. 1). The genotypes which were present at origin of graph, i.e. FH-320, MNH-886 and FH-153 are stable. The NS2013 was the most interactive environment due to the longest spoke length followed by NS2014, S2013 and S2014. The genotypes, including S-15, FH-155, FH-322 and IR-6 at S2013; MNH-888, FH-207 and FH-173 at NS2013; FH-4243, FH-153 and FH-329 at S2014 and FH-118, VH-283, IR-4 and FH-161 at NS2014 environments had a strong positive interaction. Conversely, the genotypes VH-259, VH-283, IR-4 and FH-118 at S2013; IR-3701, IR-901, AS-01 and CRS-456 at S2013; FH-142, KZ-189 and FH-173 at S2014 and NS2014 had a strong negative interaction. In addition, 2013 (NS2013 and S2013) environment vectors displayed a greater angle with 2014 (NS2014 and S2014) vectors indicating that 2013 and 2014 conditions had different interaction with genotypes for seed cotton yield. Greater angle of NS2013 and S2013

vectors with NS2014 and S2014 vectors indicated that 2013 and 2014 condition had shown different interaction with genotypes for seed cotton yield.

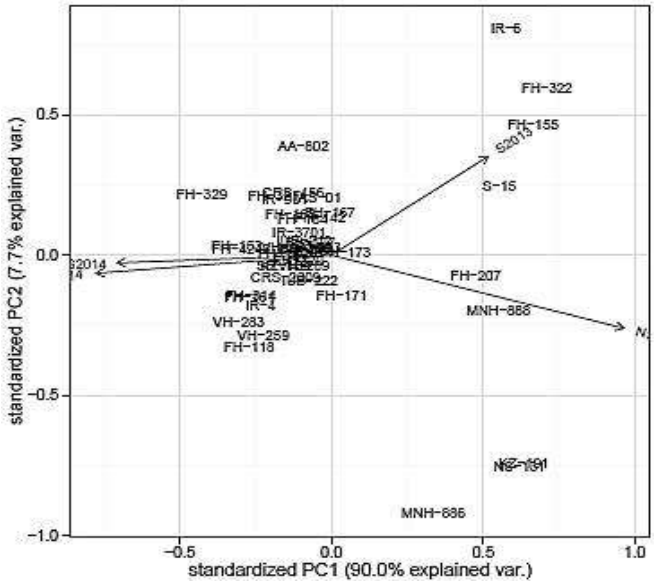

Figure 1. AMMI Biplot for Seed cotton yield.

***Water potential*:** Biplot for water potential under four drought environments plotted between PC1 and PC2 showed 74.8% of total interaction (Fig. 2). S2014 was the most interactive environment followed by NS2013, S2013 and NS2014 for water potential of genotypes on the basis of spoke length differences. All the four environments interacted differentially with genotypes for water potential. VH-291, KZ-181, FH-314 and FH-161 in S2013; FH-329, FH-318, FH-142 and A-ONE in NS2013; IR-4, FH-4243, FH-171 and FH-173 in S2014 and KZ-189 and FH-159 in NS2014 had positive interaction with the respective environments. While the genotype AA-802 in S2013; NS-131, AA-703 and FH-322 in NS2013; FH-320, FH-171 and AA-802 in NS2014; SB-149, S-15, KZ-189 and MNH-886 in S2014 had strong negative interaction with the corresponding environments. All the genotypes under different environments interacted differentially for water potential as represented by dispersive and no overlapping graph.

***Osmotic potential*:** Contribution of interaction representing PC1 and PC2 was 74.2% for osmotic potential at four different irrigation water levels, i.e. S2013, NS2013, S2014 and NS2014 (Fig. 3). Vector of S2013 was most interactive followed by NS2013, S2014 and NS2014. Interactive strength of S2014 and NS2014 environments was nearly similar but different from S2013 and NS2013. Genotypes FH-314, VH-291, KZ-181 and FH-161 in S2013; FH-4243, AA-802 and FH-329 in NS2013; VH-289 and FH-159 in S2014 and FH-173, NS-131 and KZ-191 in NS2014 had strong positive interaction for osmotic potential with respective environments. Whereas IR-4, FH-321, A-ONE and FH-142 in case of S2013; FH-207, IR-6 and FH-322 in NS2013; KZ-189, VH-283, FH-155 and FH-318 in S2014 and FH-320, AS-01 and IR-3701 in NS2014 had strong negative interaction with the respective environments.

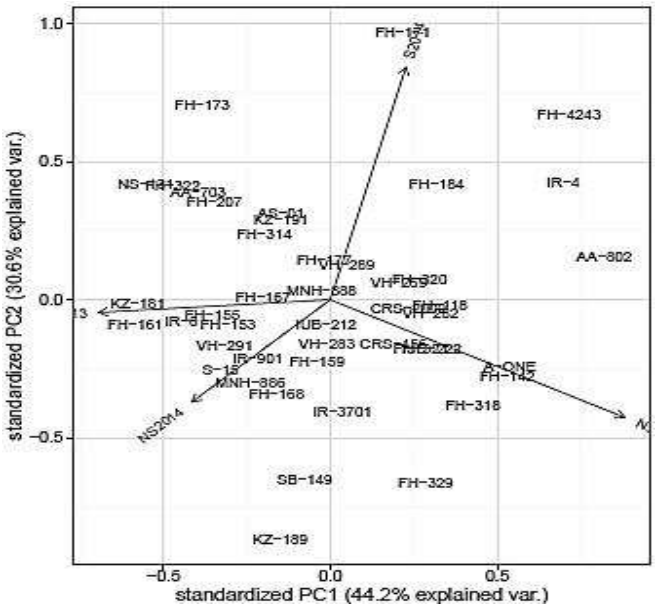

Figure 2. AMMI Biplot for Water potential.

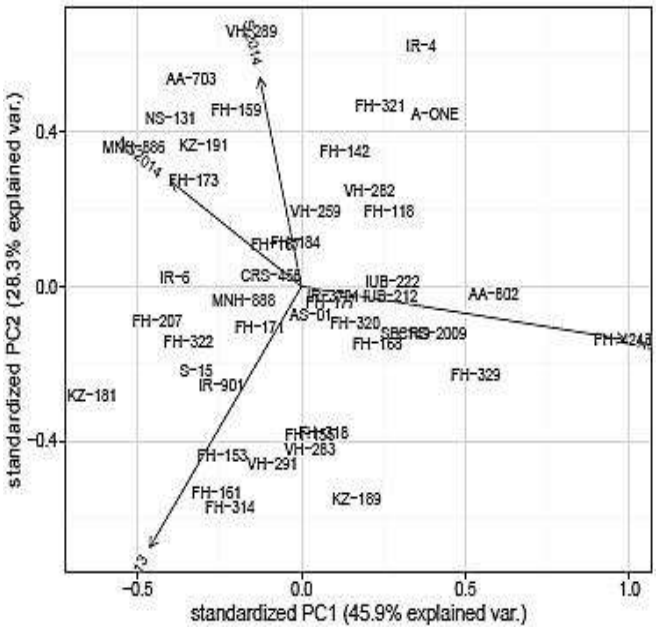

Figure 3. AMMI Biplot for osmotic potential.

***Pressure potential*:** In Figure 4, biplot analysis exhibited 83.9% interaction, between PC1 and PC2, for pressure potential (PP) under S2013, NS2013, S2014 and NS2014. S2013 vectors were the most interactive followed by NS2013,

S2014 and NS2014 respectively on the basis of their respective spoke lengths. SB-149, FH-321, FH-320, FH-168 and FH-173; FH-142, MNH-886, FH-318 and CRS-456; FH-4243 and FH-322; FH-329, IUB-212 and FH-167 had strong positive interaction with S2013, NS2013, S2014 and NS2014 respectively. Strong negative interaction was observed for FH-171, MNH-888 and FH-153 at S2013; for VH-289 KZ-181 and FH-142 at S2014; for KZ1-191, FH-320 and IR-3701 at NS2014.

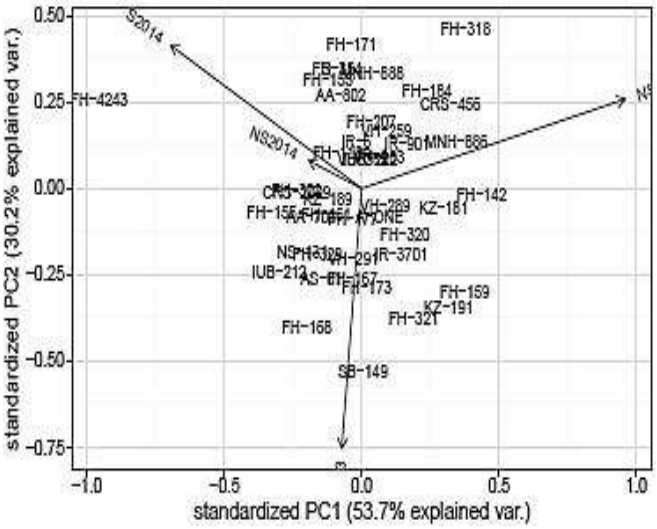

Figure 4. AMMI Biplot for pressure potential.

DISCUSSION

The effects due to water-deficit stress depend upon the severity as well as duration of the stress, the growth stage of the plant at which stress is imposed and the genotype of the plant (Kramer, 1983). The negative impact of water-deficit stress on the physiology, growth, and yield of cotton plant was recently studied by Loka *et al.* (2012). This study discusses the effects of drought stress on reproductive development of cotton (*Gossypium hirsutum* L.). The indeterminate growth habit and perennial nature of cotton plant results in occurrence of different stages of flowering and fruiting simultaneously. This uncertainty has contributed to conflicting reports on which stage of crop development is more sensitive to water deficit stress. Whilst, according to Orgaz *et al.* (1992) the water deficit stress during the peak flowering had most negative effects on seed cotton yield. In contrast, many reports (Plaut *et al.*, 1992; Radin *et al.*, 1992; De Cock *et al.*, 1993) specified that boll development period, specifically after the effective flowering, is the most sensitive period to water-deficit stress in cotton. In our study we skip the irrigations for stress environment at initiation of flowering, 50% flowering and boll development. The genotypes FH-320, MNH-886 and FH-153 were stable for all environments while the genotypes FH-155, FH-4243, FH-153, IR-6, FH-322, S-15 and FH-329 S2014 had strong positive interaction with water deficit conditions for seed cotton yield. Remaining genotypes showed lower yield under four environments. Lint yield in cotton is normally reduced due to reduced boll setting, mainly because of flowers and boll abortions when stress occurs during reproductive stage of cotton plant (Pettigrew, 2004). The genotypes VH-291, FH-314, FH-161, IR-4, FH-4243, FH-171 and FH-173 showed higher value for water potential under water deficit condition. Leaf water potential is a reliable indicator of plant water balance (Karamanos, 2003). It had been seen that plant response in terms of leaf water potential was much dependent on irrigation applied. The leaf water potential of the stressed plants was significantly lower than that of non-stressed plants (Loannis *et al.*, 2015). The genotypes which maintained higher value of water potential are considered tolerant against water deficit conditions (Silva *et al.*, 2013). Genotypes FH-314, VH-291, VH-289, FH-159 and FH-161 had strong positive interaction with water deficit conditions (S2013 and S2014) for osmotic potential. We observed higher negative values of water and osmotic potential for sensitive varieties than tolerant (Sayar *et al.*, 2008). CRS-456, FH-171, FH-155 and FH-207 had strong positive interaction with water deficit conditions i.e. S2013 and S2014 for pressure potential. Osmotic adjustment is considered to be a major acclimation response under water deficit condition because by increasing the solute concentration in cell, it maintains the Ψ_w gradients required to confirm the continued uptake of water during the stress period (Oosterhuis and Wullschleger, 1988). This process involves the accumulation of organic acids, sugars and ions in the cytosol to decrease the Ψ_s and subsequently, help to maintain leaf Ψ_w near optimum levels (Zhu *et al.*, 1997).

***Conclusion*:** The genotypes VH-291, FH-329, FH-153, IR-6, FH-159, VH-289, FH-322, MNH-886, S-15 and FH-207 are suitable for cultivation under water deficit conditions whereas; NS-131, AA-703 and KZ 191 are not favorable for water-deficit condition.

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