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Genetic Diversity Analysis through Phenotypic Assessment in Bt-Cotton Germplasm

M. A. MUGHERI, A. W. BALOCH, M. BALOCH⁺⁺, T. A. YASIR*, N. GANDAHI, G. H. JATOI**, A. M. BALOCH*** M. ALI****, I. A. BALOCH

Department of Plant Breeding and Genetics, Sindh Agriculture University, Tandojam, Pakistan

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Abstract: The present research was carried-out to analysis the genetic diversity in a set of 26 Bt-cotton genotypes for seed cotton yield and its associated traits. Results showed that genotypes differed significantly $P \le 0.01$ for all the traits, registering the significant genetic variability among the genotypes for further evaluation. With regards to mean performance, varieties BH-180 and KZ-389 displayed better performance in terms of plant height, bolls plant-1, seed cotton yield plant-1, monopodial branches plant-1 and sympodial branches length, suggesting that these genotypes can further be exploited for various breeding programs to improve upland cotton genotypes. In cluster analysis, all genotypes divided into nine small clusters, cluster one consisted of five genotypes which were characterized as high yielding genotypes suggesting that genotypes of this cluster could be utilized for future breeding programs in order to get better yielding varieties. In total, first three principal components accounted 75.90% variability, which is considered very high and could be utilized for further breeding programs in cotton. The obtained results may provide a helpful guide for selecting specific genotypes with distinct genetic backgrounds in cotton breeding programs.

Keywords: Genetic diversity, seed cotton yield, Gossypium hirsutum, Bt-cotton.

1. <u>INTRODUCTION</u>

The cotton crop is widely cultivated due to the fact that cultivars upland cotton (Gossypium hirsutum L.) meet the 90% of world's cotton demand (Wendel et al., 1992). The cotton crop is also very important in Pakistan because it sustains the economy by means of foreign exchange and employment. The production of cotton has been maintained since several years throughout the world, and at present, it is being grown in nearly more than 100 countries. The top four cotton producing nations are China, USA, India and Pakistan, which account nearly 2/3 of world's cotton acreage (Dahab et al., 2013). Crop genetic diversity is considered as the critical element in the production of crops, and it is also important to identify the genetic diversity in gene pool for the genetic conservation and breeding programs. Hence, it is essential to know the relationships among the plant varieties and genetic diversity in order to recognize complexity of gene pool and also to identify the gaps in the genotype collections (Baloch et al., 2014). In order to get the superior genotypes, it has become very necessary to properly exploit the germplasm in hybridization programs and also to adding the new germplasms; these practices are enough in order to create the sufficient variations (Li, et al., 2008). The variability in the germplasm causes resistance against both abiotic as well as biotic stresses. For getting the desirable genetic variability, various plant breeding procedures have been applied

such as hybridization, exotic germplam and the polyploidy (Esmail *et al.*, 2008). Therefore, in the current study, genetic diversity was estimated for various agronomical traits which can further be utilized in different breeding programs.

2. <u>MATERIALS AND METHODS</u>

The current experiment was conducted at the Botanical Garden, Sindh Agriculture University, Tandojam in order to assess genetic variability of some agronomical traits in Bt cotton (Gossypium hirsutum L.) germplasm. In a total, 26 Bt-cotton genotypes (Table-2) were grown in Randomized Complete Block Design with three replications. The distance of 30 and 75 cm was kept between plant to plant and row to row, respectively. All recommended practices were applied at appropriate time. The traits studied were plant height (cm), monopodial branches plant⁻¹, sympodial branch plant⁻¹, sympodial branch length (cm), bolls palnt⁻¹, boll weight (g), seed index (100-seed weight, g) and seed cotton yield plant⁻¹(g). The analysis of variance, mean comparisons and principal component analysis were derived through Statistix v. 10 software, while cluster analysis was carried out through NTSys-pc ver. 1.2 software. software by putting matrix (0 and 1). For obtaining matrix, the recorded phenotypic data was converted into 0 and 1 matrix as suggested by Shakhatreh et al. (2010) and upland cotton genotypes

*College of Agriculture, Bahauddin Zakaria University, Bahadur Sub-Campus Layyah

⁺⁺Corresponding author's email: munaizabaloch@yahoo.com

^{**}Department of Plant Pathology, Sindh Agriculture University, Tandojam, Pakistan

^{***}Department of Horticulture, Sindh Agriculture University, Tandojam, Pakistan

^{****}Department of Biotechnology, Sindh Agriculture University, Tandojam, Pakistan

grouped into three groups based on their mean and standard deviations. The three groups were:

- 1. Mean -1 standard deviation.
- 2. Mean -1 to +1 standard deviation.
- 3. Mean +1 standard deviation

3. <u>RESULTS AND DISCUSSION</u>

Analysis of variance and mean performance:

The analysis of variance for all the characters is given in (Table-1). The obtained results revealed that genotypes differed significantly at P ≤ 0.01 probability level for plant height, monopodial branches plant⁻¹, sympodial branches plant⁻¹, sympodial branches length, bolls plant⁻¹, boll weight, seed index and seed cotton yield plant⁻¹, suggesting that studied materials possess useful genetic resources for variety of traits thus can extensively be used for upcoming breeding programs. Almost similar results have also been reported by Baloch et al. (2014). The data regarding mean performance of genotypes is given in (Table-2). The data revealed that genotype BH-180 produced the tallest plants of 146.30 cm while BZU-75 recorded the shortest plants measuring of 108.67 cm as compared to the rest of the genotypes. In case of monopodial branches plant⁻¹, the genotype KZ-389 produced maximum (3.33) number of monopodial branches plant⁻¹;however, no monopodial branches plant⁻¹ was found in BZU-75, NIAB-1, CIM-602, VH-282 and CEMB-33 genotypes. For sympodial branches plant⁻¹, FH-118 produced the highest number of sympodial branches plant⁻¹ (27.66), whereas Tarzen-402 produced the lowest number of sympodial branches plant⁻¹ (17.33). With regards to sympodial branches length, the variety KZ-389 measured the longest branch length (34.077 cm) against the other genotypes, while the shortest (11.10 cm) branch length was measured from AGC-777. With respect to bolls plant⁻¹, the variety BH-180 set maximum number of bolls plant⁻¹ (46.16) while minimum (28.16) number of bolls plant⁻¹ were obtained from AGC-777.AGC-777 weighed bigger bolls of 4.15g, while the smaller bolls were observed in variety BH-180 (3.17 g). The variety Tarzen-402 gave higher seed index (7.46 g), however, the variety FH-118 gave the lower seed index(6.20 g). The variety BH-180 produced maximum seed cotton yieldplant⁻¹ (148.08 g), whereas the variety FH-118 gave the lowest seed cotton yieldplant⁻¹ (122.23 g). In general, varieties BH-180 and KZ-389 displayed better performance in terms of plant height, bolls plant⁻¹, seed cotton yield plant⁻¹, monopodial branches plant⁻¹ and sympodial branches length, indicating that these cotton genotypes offer great potential for further evaluation.

Cluster analysis:

Cluster analysis has been used as most widely technique in order to classify the different genotypes into homogeneous groups. It works on a matrix of similarity (or dissimilarity) indexes for all possible pairs of genotypes (Ghaderi et al., 1980). Cluster analysis was performed to study the patterns of groupings of genotypes. The dendrogram (Fig-1) was generated from the UPGMA (Un-weighted Pair Group Method with Arithmetic Mean) clustering method of genotypes based on Euclidean distances. The cluster analysis classified the 26 Bt cotton genotypes into 9 small clusters, reflecting the presence of wide genetic diversity among the tested genotypes. Based on obtained results, it is suggested that the genotypes clustered together into cluster one, possessing desirable gene combinations for seed cotton yield plant⁻¹, offering that these Bt cotton genotypes could be used in future breeding programs in order to improve seed cotton yield. It has been suggested that genotypes grouped together into cluster eight should not be used in cotton breeding programs since the genotypes of that cluster contain undesirable gene recombination for seed cotton yield and its related traits. It is also recommended that hybridization program should be avoided between cluster one with cluster eight because later cluster do not possess reliable gene combinations for yield and morphological traits. Similar to our results, Xian et al. (2012) also reported that genetic diversity analysis divided 38 cotton genotypes into two groups with similar genetic background.

Principal component analysis:

The conservation and exploitation of genetic resources could be achieved by partitioning the total variance into its components. It also offers a chance for utilization of proper germplasm in crop development for specific plant characters (Pecetti et al., 1990). PCA is an important tool to get parental materials for successful breeding strategies (Nazir et al., 2013). In this study, out of total eight, first three principal components were extracted having Eigen value more than one. However, these principal components (PCs) contributed 75.90% of the total variability amongst the cotton genotypes assessed for various yield and its associated traits while remaining five principal components contributed only 24.10% towards the total diversity for this set of cotton genotypes. First three principal components explained 75.90% variability, which is considerably high and can be utilized for further breeding programs in cotton. The positive and negative loading reveals the occurrence of positive and negative association trends between the components and the variables. Therefore, the given characters (Table 1-3) which load high positively and negatively contributed more to the genetic variability and they were the ones that most distinguished the clusters. As usual, it is customary to choose one variable from known groups. Hence, for the first group boll weight value is best choice, which had the largest loading from component one, seed index for the second, plant height for the third group, sympodial branches plant⁻¹ for the fourth group, monopodial branches plant⁻¹ for the fifth group, sympodial branches length for the sixth group, bolls plant⁻¹ for the group seventh and seed cotton yield plant⁻¹ for the eighth group. Recently, Elci *et al.* (2014) also derived PCA on morphological data in Turkish cotton varieties where PCA indicated the relationships of genotypes in a more significant manner showing that PCA should be used along with the cluster to achieve a better perceptive of relationships among genotypes.



Fig-1 Tree diagram of 26 upland cotton genotypes for 8 characters using hierarchical cluster analysis (Ward's Method).

Source of	Degree of	Characters						
variation	Freedom	Plant height	Monopodial branches plant ⁻¹	Sympodial branches plant ⁻¹	Sympodial branches length			
Replications	2	6.640	3.57692	0.9744	6.9882			
Genotypes	25	264.797**	2.16821**	15.3215**	69.9882**			
Error	50	30.808	0.28359	2.9877	3.5551			
Source of	Degree of		Characters					
variation	Freedom	Bolls plant ⁻¹	Boll weight	Seed index	Seed cotton yield plant ⁻¹			
Replications	2	58.5054	0.13702	0.00628	204.334			
Genotypes	25	49.7927**	0.16212**	0.26759**	150.098**			
Error	50	4.7787	0.0016	0.0455	13.134			

	Table-1 Mean so	quares from a	nalysis of v	variance for	various ti	raits in B	t cotton genotypes.
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Note: ** = Significant at 0.01 level of probability.

Table-2 Mean performance of Bt-cotton genotypes for various traits

Genotypes	Plant	Monopo-dial	Sympodial	Sympodial	Bolls	Boll	Seed	Seed cotton
	height	branches plant	branches plant ⁻¹	branches	plant ⁻¹	weight	index	yield plant ⁻
	(cm)	-1		length (cm)		(g)	(g)	¹ (g)
BH-180	146.30	1.33	22.00	11.26	46.16	3.17	6.76	148.08
BS-52	141.80	2.00	25.66	19.42	35.33	3.67	6.63	130.60
NS-161	131.37	0.33	19.66	14.02	33.83	3.73	6.33	127.92
Soyaben	131.52	1.33	23.66	18.66	35.05	3.72	6.30	131.89
KZ-389	129.80	3.33	22.66	34.07	41.00	3.35	6.46	139.02
GH-142	129.33	0.33	24.33	12.90	35.50	3.58	6.46	128.51
Leader-1	120.33	0.33	23.66	7.03	36.50	3.57	6.60	131.59
AGC-777	123.67	0.66	23.3	11.10	28.16	4.15	6.30	118.25
Trend-1	121.92	1.33	23.00	9.88	36.50	3.57	6.76	131.75
BZU-75	108.67	0.00	21.00	15.65	35.00	3.67	6.43	129.77
Sitara-12	130.00	1.00	20.00	11.70	36.00	3.64	7.20	132.54
Tarzen-402	125.00	1.66	17.33	1.60	37.83b	3.57	7.46	136.43
NIAB-1	115.00	0.00	19.33	12.46	34.66	3.64	6.46	128.08
CEMB-44	113.00	1.66	20.00	14.86	31.66	3.91	6.26	123.70
CIM-602	111.00	0.00	20.33	14.46	36.66	3.55	6.40	131.45
AA-919	113.67	0.66	21.00	13.56	32.00	3.85	6.30	125.36
SB-149	122.00	0.66	22.00	15.28	37.66	3.54	6.56	135.04
MM-58	115.67	2.00	21.66	14.98	45.66	3.19	6.60	147.77
SLH-4	111.00	0.66	24.00	16.50	37.66	3.57	6.66	135.31
Soyaben-202	122.33	0.33	23.66	18.16	37.50	3.52	6.50	133.73
VH-282	132.33	0.00	19.66	13.60	31.66	3.89	6.20	124.26
JS-1V	117.33	0.66	24.66	14.36	33.33	3.76	6.26	126.91
CEMB-33	117.33	0.00	22.33	14.36	31.66	3.88	6.30	124.30
FH-118	117.67	0.33	27.66	12.86	30.33	3.95	6.20	122.23
Tarzen-3	123.00	2.33	23.66	13.10	35.66	3.54	6.33	127.04
AA-904	116.33	1.00	22.00	12.43	36.66	3.23	6.23	122.67
LSD (5%)	9.1028	0.8733	2.8347	3.0922	3.5851	0.3245	0.3048	5.9435

Characters	Eigenvectors								
Characters	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	
Plant height	-0.2116	-0.3224	0.6466	0.2105	-0.5028	-0.3649	0.0493	0.0240	
Monopodial branches plant ⁻¹	-0.3353	-0.2756	0.2856	-0.1141	0.8098	-0.2413	-0.0014	-0.0555	
Sympodial branches plant ⁻¹	0.0549	-0.5657	-0.1570	0.6757	0.1077	0.4287	-0.0136	-0.0880	
Sympodial branches length	-0.0951	-0.5592	-0.0190	-0.6841	-0.2083	0.3936	-0.0623	-0.0110	
Bolls plant ⁻¹	-0.5153	0.0504	-0.1804	-0.0059	-0.0548	0.1229	0.5692	0.5978	
Boll weight	0.4966	-0.0630	0.3098	-0.0921	0.1392	0.0885	-0.2395	0.7486	
Seed index	-0.2675	0.4246	0.5247	0.0682	0.0465	0.6604	-0.1231	-0.1217	
Seed cotton yield plant ⁻¹	-0.4971	0.0240	-0.2675	0.0702	-0.1096	-0.1042	-0.7727	0.2366	
Eigenvalues	3.38706	1.60614	1.07494	0.81725	0.57447	0.37777	0.11320	0.04918	
Percent of variance	42.3	20.1	13.4	10.2	7.2	4.7	1.4	0.6	
Cumulative percent of variance	42.3	62.4	75.9	86.1	93.2	98.0	99.4	100.0	

Table-3 Vector loadings and explained percentage variance by the 8 PCs.

CONCLUSION

Highly significant differences were observed among Bt cotton genotypes evaluated for all the 8 studied traits. The results of the current study show the presence of genetic diversity among the selected Bt cotton genotypes. Parents from divergent clusters can be used for hybridization in order to separate useful recombinants in the segregating generations. This information might be used in the genetics and breeding programs for improvement of upland cotton genotypes.

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