



## Exploitation of Germplasm for Plant Yield Improvement in Cotton (*Gossypium hirsutum* L.)

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**ABSTRACT:** Diverse genetic resources are vital for ample production for imminent challenges. The current study was aimed to find pattern of genetic diversity among 50 cotton genotypes and selection of diverse genotypes for upcoming cotton breeding programs. Data was recorded for fourteen quantitative attributes. All attributes exhibited tiny difference phenotypic coefficient of variation (PCV) and respective genotypic coefficient of variation (GCV), suggesting that environment had less effect on all the recorded attributes. High expected genetic advance coupled with higher heritability (broad sense) were recorded for plant height, number of bolls per plant, weight per boll and plant yield, indicating the existence of additive gene action hence selection on phenotypic basis might be productive. Correlation studies revealed positive and significant association for most of the attributes. Plant height, number of monopodia and sympodia/plant, and number of bolls/plant contributed well towards considerable principal components (PCs) and were highly related to plant yield (seed cotton yield per plant). The projection of cotton genotypes on 1<sup>st</sup> and 2<sup>nd</sup> PC exhibited population structure. Best diverse genotypes were IUB-222, SB-149, CIM-612, CIM-598, CIM-506, RS-1 and VS-1. The outcomes for this experiment are helpful for planning forthcoming cotton breeding programs particularly in Pakistan.

**Key Words:** Biodiversity, Principal component analysis, Phenotypic coefficient of variation, Genotypic coefficient of variation, Heritability

**INTRODUCTION:** Conserving genetic diversity in agriculture is not optional, but essential to maintain natural resources which uphold agriculture. Plant breeders often make use of germplasm to develop improved genotypes for the upcoming environmental conditions that completely outclass the previous genotypes in terms of performance.

The variability for economic attributes in the given germplasm is vital for gratifying exploitation following selection and breeding (Sajjad, *et al.*, 2011). Evaluation of biodiversity is of prime importance for analysis of genetic miscellany in genotypes and introgressive hybridization of favorable attributes from sundry germplasm into the

existing genetic base (Thompson and Nelson, 1998).

The assessment of biodiversity could be utilized on lineage records morphological attributes and molecular markers (Malik, *et al.*, 2014). Biodiversity based upon morphological attributes has been widely evaluated in various crops, including cotton (Ahmad, *et al.*, 2012), to plan for successful breeding programs. In future, cotton yield will heavily rely on development of genotypes with superior capability of yield and quality of cotton seed along with tolerance to living and non-living stress factors. The concept of broadening the genetic base of cotton germplasm was overlooked previously as a consequence of this reduction in biodiversity (Chen and Du, 2006). It has been contemplated that reduction in cotton yield and fiber characteristics is the outcome of a narrow genetic base in the elite cotton germplasm (Tyagi, *et al.*, 2014). So for the improvement of elite genotypes or launching of any breeding strategy, genetic variation is of supreme significance. Variation in germplasm assists in boosting up the yield along with accumulation of favorable combinations that can be used in forthcoming breeding program (Saeed, *et al.*, 2014).

When dealing with a large number of genotypes, multivariate biometrical techniques are mostly utilized to assess biodiversity irrespective of data set. Among these biometrical techniques, principal components analysis (PCA), principal coordinate analysis (PCoA), cluster analysis and multidimensional scaling (MDS) are mostly utilized by the plant breeders (Brown-Guedira, *et al.*, 2000).

Being hierarchical analytical technique, cluster analysis is of limited usefulness to assess the pattern of biodiversity. This hierarchical analysis will more useful when the variables are non-linearly associated (Lessa, 1990). Most plant breeders are

utilizing PCA and PCoA to assess pattern of variation present among genotypes rather than cluster analysis. With the help of PCA, each genotype can be allocated to single group and it also imitates the significance of humongous contributor to the entire variation at every axis of differentiation (Saeed, *et al.*, 2014). Genetic variability for morphological attributes has been assessed by employing PCA, which lead to the detection of phenotypic variation in cotton (Esmail, *et al.*, 2008 and Li, *et al.*, 2008).

Biodiversity assessment based merely on morphological data set requires a very high level of accuracy of field experimentation through proper design and analysis. Keeping in view the vitality of genetic diversity for cotton breeding, the present study was conducted with high a precision level. In this investigation, a set of fifty cotton genotypes was evaluated; (i) to assess the extent of variability for fourteen parameters, (ii) to explore grouping pattern (iii) to notify genetically diverse but agronomically important genotypes.

**MATERIALS AND METHODS:** The study was conducted on fifty cotton (*Gossypium hirsutum* L.) genotypes (Figure 2), collected from Cotton Research Station, Multan, during normal cropping season 2013 following Randomized Complete Block Design (RCBD) with three replications at Four Brothers Research Farm, Multan. For each entry, 4.5 meter × 1.5 meter of plot size was used consisting of two rows set at a distance of 29.5 inches. Plant to plant distance within rows was 11.8 inches. The seedlings were thinned fifteen days after planting, leaving one plant per hill. Recommended doses of urea (100 kg/acre) and DAP (50 kg/acre) fertilizers were applied to raise homogenous crop. At the time of maturity, ten randomly selected plants where chosen from each line and then recorded for fourteen attributes namely plant height (cm), number of monopodia and

sympodia branches/plant, number of bolls/plant, weight/boll (g), plant yield (g), ginning out turn (%), lint index (%), seed index (%), chlorophyll contents (%), fiber length (mm), fiber strength (g/tex), micronaire value (ug/inch) and uniformity index (%). Plant height was recorded from 1<sup>st</sup> node to shoot apex. Number of sympodia and monopodia was recorded by counting the fruiting and vegetative branch respectively. Total chlorophyll contents were recorded with the help of SPAD meter by taking readings from fresh green leaves. Mature fully open bolls were taken and counted for boll number/plant and as well as weight/boll. Individual plant yield was calculated by weighing seed cotton obtained from all bolls. Seed index was calculated by weighing hundred fuzzy seeds of cotton on electrical digital weighing balance. All fiber quality attributes was measured using apparatus HVI-1000 from laboratory of Four Brothers Group Pakistan. This device gives an accurate profile of raw fiber. Lint index was obtained by weighing 100 seeds in grams, however, below mentioned formula was applied to calculate lint index from individual plant. To calculate lint percentage, dry and clean seeds were picked from bolls of each and every plant followed by weighing and then ginning outturn (G.O.T) was calculated by the below mentioned formula following Khan *et al.*, (2010). Broad sense heritability and genotypic & phenotypic variance was also calculated by following formula as reported by Khan, *et al.*, (2010).

$$\text{Lint index} = \frac{(\text{Seed Index} \times \text{Lint \%})}{(100 - \text{Lint \%})}$$

$$\text{G. O. T \%} = \frac{\text{Weight of lint in a sample}}{\text{Weight of seed cotton sample}} \times 100$$

Genotypic and phenotypic variance was also calculated by using following formula.

$$\text{Genotypic variance (Vg)} = \frac{\text{Genotype mean square (GMS)} - \text{Error mean square (EMS)}}{\text{Number of replication (r)}}$$

$$\text{Phenotypic variance (Vp)} = Vg + Ve/r$$

$$\text{Heritability (H}^2\text{)} = Vg/Vp$$

**Statistical Analysis:** Analysis of variance (ANOVA) for RCBD was performed for all the recorded data as reported by Steel *et al.*, (1997) through Microsoft Excel. To assess the pattern of diversity present among the cotton genotypes PCA was conducted as outlined by Saeed *et al.*, (2014) through PAST software.

**RESULTS AND DISCUSSION:** Analysis of variance revealed considerable difference amongst given genotypes pointing out significant amount of genetic variability. High amount of variability in present experimental material courtesy higher values of mean and range for nearly all the traits. On the basis of range, difference was more for traits viz., plant height, plant yield, number of bolls/plant and number of sympodia/plant. Our outcomes were in accordance with the results of Erande *et al.*, (2014) and Khan *et al.*, (2014).

**Coefficient of Variation and Variance of Components:** The estimates of genetic attributes such as coefficient of variability (CV%) genotype mean square (MS), environmental variance ( $\sigma^2_e$ ), phenotypic variance ( $\sigma^2_p$ ), genotypic variance ( $\sigma^2_g$ ), phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), environmental coefficient of variation (ECV), Broad sense heritability ( $H^2$ ), and Genetic Advance (GA) are given in Table I.

The values of genotypic variance were less than the phenotypic variance for all the attributes. Maximum genotypic and phenotypic variance was exhibited by plant yield with values of 878.2 and 1031.2 respectively while micronaire values showed lowest genotypic and phenotypic variance with values of 0.15 and 0.164 correspondingly. Similarly values of environmental variance were also less as compared to GV and PV expect for fiber

strength. The outcome of these findings revealed that environmental variations were greater enough to modify gene expression organizing the attribute under examination. Selection pressure could be applied on the attributes to notify classy genotypes.

As in case of genotypic and phenotypic variance, similar results were obtained for GCV, PCV and ECV. Lower magnitude of difference was observed between them indicating that they are nearly similar to each other. These outcomes indicate that these traits are less affected by environment and selection could be effectual. Baloch *et al.*, (2014) and Dhivya *et al.*, (2014) also reported alike results in their investigations. Maximum estimates of GCV and PCV were observed for plant yield, plant height and bolls per plant respectively. Whereas micronaire value and lint index exhibited minimum values for GCV and PCV (Table I). These attributes may be a poor index for selection of genotypes because of less values of genotypic and phenotypic coefficient variation. Bechere *et al.*, (2014) and Dinakaran *et al.*, (2012) also reported low values of GCV and PCV for micronaire value and lint index.

**Heritability and Genetic Advance:** Heritability values are valuable in forecasting

the estimated improvement to be attained through the procedure of selection. Heritability estimates along with GCV offers a dependable guesstimate of amount of genetic advance to be anticipated through phenotypic selection. Heritability values > than 80% are high while values in between 79% and 60% are moderately high whereas values less than 59% are considered as low (Singh, 1983). Genetic advance (GA) refers to the betterment of traits in genotypic value for novel population as compared with the base population in 1 cycle of selection at a particular selection intensity (Singh, 1983). High heritability (broad sense) and genetic advance (GA) was noted for attributes *viz.*, plant height, number of bolls/plant, weight/boll and plant yield. The high estimates of heritability and genetic advance indicating that there could be predominance of additive gene action and are not much prejudiced by environmental changes, hence could be selected for cotton improvement through these traits. These observations were in agreement with the results of Dinakaran *et al.*, (2012). High heritability and low genetic advance was noted for uniformity index, micronaire value and lint index indicating very little scope of breeding through selection.

**Table 1: Attributes of genetic variability**

	Range	CV%	MS	EV	GV	PV	GCV	PCV	ECV	H <sup>2</sup>	GA
<b>Plant Height</b>	73.67-213	18.01	2074.6	109.8	654.6	764.76	17.53	18.94	7.17	85.64	62.99
<b>No. of Monopodia/plant</b>	0.17-5.17	13.20	4.23	0.435	1.26	1.701	69.36	80.42	40.69	74.4	1.99
<b>No. of Sympodia/ plant</b>	6.33-27.5	18.05	66.19	8.11	19.36	27.47	35.647	42.46	23.078	70.47	24.23
<b>Bolls/plant</b>	11.83-71.33	11.67	333.50	24.70	102.5	127.63	40.104	44.63	19.64	80.64	71.22
<b>Weight/boll</b>	1.76-3.84	15.95	0.502	0.035	0.155	0.191	15.37	17.04	7.34	81.42	73.3
<b>Plant Yield</b>	28.91-173.2	16.66	2787.7	153.0	878.2	1031.2	45.36	49.16	18.93	85.16	56.33
<b>G.O.T</b>	32.25-48.08	7.32	26.306	1.208	8.36	9.57	7.15	7.65	2.71	87.38	5.56
<b>Lint Index</b>	3.43-6.21	13.60	1.241	0.137	0.37	0.505	12.839	15.04	7.84	72.83	1.06
<b>Chlorophyll Contents</b>	36.88-55.05	8.45	45.755	12.08	11.22	23.3	7.252	10.45	7.525	64.15	38.21
<b>Seed Index</b>	5.17-9.82	12.98	2.39	0.19	0.74	0.925	12.45	13.97	6.33	79.43	1.57
<b>Fiber Length</b>	22.8-29.67	6.22	8.09	0.934	2.38	3.321	5.85	6.90	3.66	71.87	2.69
<b>Fiber Strength</b>	26.45-39.35	8.06	2.77	5.43	5.2	7.97	7.4338	9.20	5.4316	81.23	37.55
<b>Micronaire Value</b>	3.4-5.6	7.97	0.456	0.018	0.15	0.164	7.828	8.29	2.747	89.03	0.74
<b>Uniformity Index</b>	73.9-83.4	2.76	14.902	1.45	4.50	5.88	2.637	3.01	1.458	76.58	3.87

**Principal Component Analysis:** Out of 14 principal components 5 PCs showed eigen value more than 1. These 5 PCs, contributed 76.65% of total variability present among the given cotton genotypes. Whereas, remaining 9 components contributed 23.34% towards total variability. First and second PC exhibited 45.89% variability (Table 2) so these PCs were given the due importance for further explanation. First PC had a high match with plant yield (0.4388), plant height (0.3721), bolls per plant (0.3569) and number of sympodia per plant (0.3494) (Figure 1). IUB-222 (2.82), SB-149 (2.79), RCA-2 (1.95), SITARA-12 (1.509), NS-161 (1.299), SAYBAN-202 (1.182), SUN-1 (0.978), BGC-09 (0.854), AURIGA-213 (0.823) and AGC-777 (0.820) had higher values for first PC (Figure 2). Whereas, 2<sup>nd</sup> PC had a high match to lint index (0.3641) and weight per boll (0.3357) figure 1. Second PC was more related to VS-1 (2.022), LEADER-1 (1.74), CIM-482 (1.46), KS-389 (1.202), RS-1 (1.099), NS-161 (0.989), AA-904 (0.960), AGC-777 (0.956), CIM-473 (0.835) and SAYBAN-201 (0.803) (Figure 2). From due significant principal components it was obvious that plant yield and plant height had maximum values only for 1<sup>st</sup> PC. For that reason, this component could be considered as a suitable production component and can be useful to notify genotypes having good production ability.

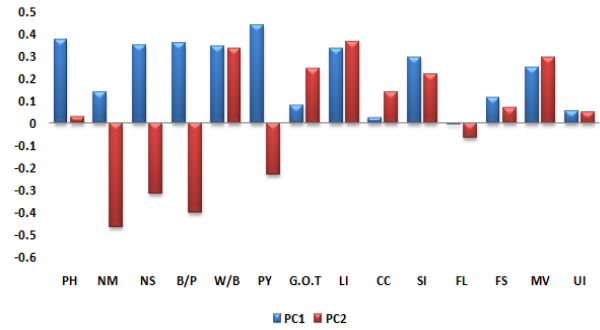


Figure 1: Factor loadings for traits

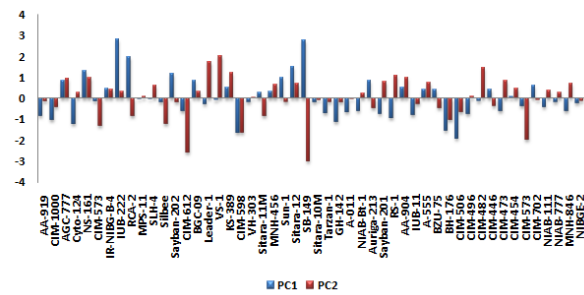


Figure 2: Factor loadings for genotypes

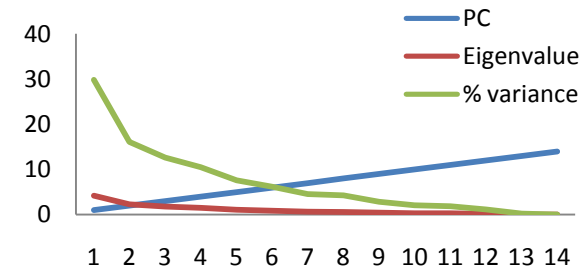


Figure 3: Scree Plot

Table 2: Eigen value and % Variance

PC	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Eigen value	4.17	2.249	1.765	1.470	1.070	0.8709	0.6352	0.5972	0.4030	0.2957	0.2605	0.1617	0.0376	0.0059
% Variance	29.82	16.06	12.61	10.50	7.64	6.22	4.53	4.26	2.87	2.11	1.86	1.15	0.27	0.05

**Scree Plot:** Scree plot exhibited variance percentage associated with each principal component attained by drawing a graph between eigen value and PC numbers. PC1 showed 29.82% variability followed by PC2 with 16.06% having eigen values of 4.17 and 2.24 respectively. Elbow resemblance stripe

was attained which after 5<sup>th</sup> PC tend to straight. After this, very little amount of variation was observed in each PC (Figure 3). From both graph it was clear that highest variation was present in first PC. So genotypes could be selected from this PC and could be utilized for future cotton breeding program.

**Spread out Plot:** The spread out plot of principal component of cotton genotypes revealed that closely located genotypes on graph are perceived as alike when rated on given attributes (Figure 4). Farthest the distance from point of origin more diversified will be the genotypes and vice versa. Figure 4 showed that most cotton genotypes in present investigation situated close to each other on the graph indicating narrow genetic background of cotton genotypes. This might be because of extensive breeding for a limited number of traits. Genotypes such as NIAB-777, VH-303, MPS-11 and NIAB-111, NIAB-Bt-1, CIM-496 and MNH-846, CIM-473 and AA-919, Tarzan-1, IUB-11 clogged very near to each other and as well as very close to the point of origin, hence of less breeding value and less diversified. On the other hand, genotypes which clogged at vertex of the polygon are farthest from point of origin hence more diversified and of high breeding value. IUB-222, SB-149, CIM-612, CIM-598, CIM-506, RS-1, VS-1 clogged at the vertex of polygon (Figure 4).

**Genotype by Trait Analysis:** The evaluation and notification of outclass genotypes for different traits were carried out by using biplot (Figure 4). RCA-2, BGC-09, SUN-1 and SAYBAN-202 were found in close vicinity of plant height and number of sympodia per plant; hence these genotypes are more related to these traits. AGC-777 and NS-161 were found in close proximity of lint index, weight/boll and seed index, these traits had a high match for these genotypes. AA-904 and KS-389 were found near to micronaire value. A-555, MNH-456 was found near to G.O.T. Moreover, SITARA-11M and CIM-612 were clogged near number of monopodia per plant.

**Relationship among Attributes:** The line between the point of origin of a biplot and marked point of any attribute is known as attribute vector or trait vector. The Cosine angle between attribute vectors decide affiliation among attributes. Attributes are

positively associated courtesy cosine angle less than  $90^\circ$  while if cosine angle is greater than  $90^\circ$  then negative association takes place between the attributes. If cosine angle between traits is nearly  $90^\circ$  then, they will act autonomously (Malik *et al.*, 2013). Biplot analysis of all gathered data of cotton genotypes showed significant as well as non-significant association among different traits (Figure 4). As yield, is of supreme importance in cotton so due significance was given to plant yield (seed cotton yield/plant). High positive but non-significant association (0.76212) was observed between plant height and plant yield (Table 3) with cosine angle less than right angle indicating increase in plant height could cause slight increase in plant yield. Bechere *et al.*, (2014) and Bibi *et al.*, (2011) also reported similar results. Therefore it is proposed that cotton breeders should be very vigilant in selection program based upon correlation between plant yield and plant height. Positive but non-significant association (0.458) was noted between plant yield and number of monopodia/plant (Table 3). These findings were in accordance with the work of Bechere *et al.*, (2014), Bibi *et al.*, (2011) and Salahuddin *et al.*, (2010b) but Sambamurthy (1999) obtained contrasting results indicating positive but significant association between these two parameters. This might be due to difference in genetic composition of investigational material. Therefore it could be concluded that the association of plant yield and number of monopodia per plant is weak and it could not be a better selection criterion. Highly positive and considerable (0.768) association was noted between plant yield and number of sympodia/plant (Table 3). The findings of the authors were in complete conformity with the outcomes of Salahuddin *et al.*, (2010a) and Soomro *et al.*, (2008). Therefore it may be concluded that selection based on number of sympodia/plant will be helpful for increasing plant yield. Highly positive and significant

association (0.892) was also found between number of bolls/plant and plant yield indicating that selection for higher plant yield based on number of bolls/plant would be fruitful. Ahsan *et al.*, (2011); Bibi *et al.*,

(2011) and Hussain *et al.*, (2010) also found similar results. Positive and significant (0.765) association was also noted between weight per boll and plant yield. Alkuddsi *et al.*, (2013); Kazerani, (2012) and Makhdoom *et al.*, (2010)

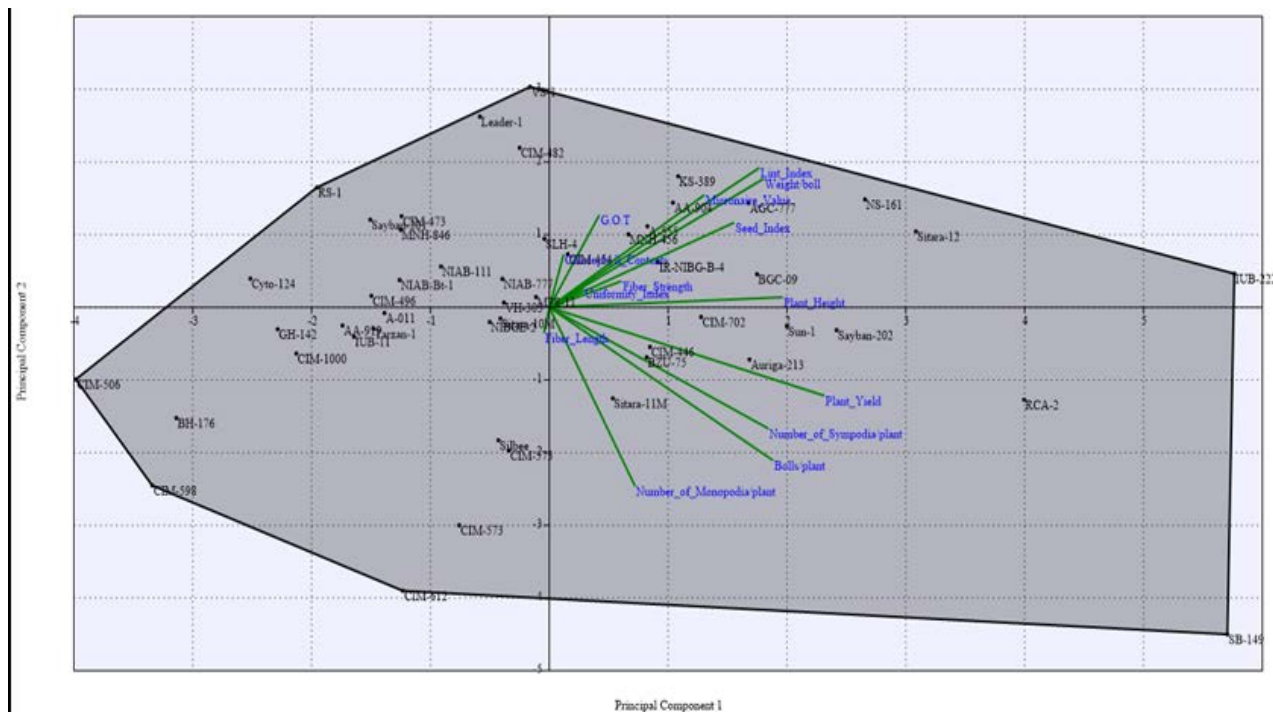


Figure 4: Biplot analysis

Table 3: Correlation analysis of studied attributes

	Plant Height	Number of Monopodia/plant	Number of Sympodia/plant	Bolls/plant	Weight/boll	Plant Yield	G.O.T	Lint Index	Chlorophyll Contents	Seed Index	Fiber Length	Fiber Strength	Micronaire Value	Uniformity Index
<b>Plant Height</b>	0.000													
<b>Number of Monopodia/plant</b>	0.152	0.000												
<b>Number of Sympodia/plant</b>	0.509	0.301	0.000											
<b>Bolls/plant</b>	0.443	0.572	0.781	0.000										
<b>Weight/boll</b>	0.519	-0.086	0.218	0.142	0.000									
<b>Plant Yield</b>	0.7621 <sup>NS</sup>	0.458 <sup>NS</sup>	0.768 <sup>**</sup>	0.892 <sup>**</sup>	0.481 <sup>**</sup>	0.000								
<b>G.O.T</b>	0.064	-0.054	-0.114	0.042	0.183	0.091 <sup>NS</sup>	0.000							
<b>Lint Index</b>	0.458	-0.072	0.202	0.176	0.718	0.424	0.567	0.000						
<b>Chlorophyll Contents</b>	0.097	-0.125	-0.053	-0.017	0.101	0.6785 <sup>*</sup>	-0.008	-0.048	0.000					
<b>Seed Index</b>	0.496	-0.116	0.252	0.140	0.602	0.364	-0.301	0.527	0.013	0.000				
<b>Fiber Length</b>	0.077	0.097	0.065	-0.064	-0.112	-0.672 <sup>*</sup>	-0.035	0.019	0.012	0.076	0.000			
<b>Fiber Strength</b>	0.092	-0.082	0.148	0.111	0.169	0.265	-0.061	0.119	0.089	0.271	-0.093	0.000		
<b>Micronaire Value</b>	0.297	-0.124	0.159	0.190	0.438	0.311 <sup>*</sup>	0.265	0.480	0.208	0.280	-0.219	0.060	0.000	
<b>Uniformity Index</b>	0.116	0.049	0.013	-0.028	0.058	0.032 <sup>*</sup>	0.002	0.122	0.031	0.202	0.642	-0.169	0.078	0.000

also reported similar results. Micronaire value showed positive and significant correlation (0.311) with plant yield. It might be concluded that coarse fiber can result by selecting cotton genotypes with higher plant yield. Our outcomes are in accordance with the results of Tabasum *et al.*, (2012). Low positive but non-significant association (0.091) was found between G.O.T and plant yield in present study. Some researcher found high positive association between them; this could be due to diverse genotypic composition of cotton genotypes. Chlorophyll contents also showed positive and significant association (0.6785) with plant yield. Our outcomes are in agreement with the results of Karademir *et al.*, (2009) who too noted positive and considerable association between these two parameters. Positive correlation (0.265) was also observed between plant yield and fiber strength suggesting that any improvement in fiber strength leads to increase in plant yield, while plant yield was observed negatively correlated (-0.672) with fiber length indicating that improvement in fiber length may cause reduction in plant yield, these findings were in consistence with the outcomes of Azhar *et al.*, (2004). Positive association (0.032) was also observed between uniformity index and plant yield. Khan *et al.*, (2014) also reported similar results.

**CONCLUSION:** Genetic diversity and selection are regarded as bread and butter for plant breeder. Successful breeding program is courtesy of effective selection. Hence critical know how about genetic diversity, heritability, genetic advance and correlation between different morphological attributes and their association with genotypes gives steadfast basis for cotton betterment. The attributes like plant height, number of monopodia/plant, number of sympodia/plant, number of bolls/plant, weight/boll, plant yield (seed cotton yield/plant), ginning out turn, seed index, lint index, fiber uniformity index, fiber strength, micronaire value and fiber length

could be used as morphological markers. Principal component analysis helped in the identification of diversified cotton genotype i.e. IUB-222, SB-149, CIM-612, CIM-598, CIM-506, RS-1, LEADER-1 and VS-1, hence these genotypes might be utilized in breeding program.

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