

DECIPHERING GENETIC DIVERSITY FOR YIELD AND AGRO - MORPHOLOGICAL TRAITS IN TROPICAL MAIZE INBRED LINES (*Zea mays* L.)

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ABSTRACT

This study aimed to determine genetic parameters and the magnitude of association between yield and morpho-agronomical traits in maize to improve the efficiency of selection in future breeding programs for high-density planting. The experiment was conducted during *Kharif* 2023 at the Maize Research Centre, Agricultural Research Institute, Rajendranagar, Hyderabad, using a Randomized Block Design with two replications. A total of 52 tropical maize inbred lines were evaluated for various morpho-agronomical traits, including number of leaves above and below the ear, tassel branch number, tassel main axis length, plant height, ear height, leaf angle, leaf length, leaf width and total number of leaves per plant, all of which influence the leaf and stem architecture of maize. The results revealed that traits such as plant height, total number of leaves, leaf length, leaf width, number of kernels per row, number of rows per cob, and shelling percentage exhibited moderate to high values of genotypic and phenotypic coefficients of variation, heritability, and genetic advance as percent of mean. These traits also showed positive and significant correlations along with direct effects on yield per cob, indicating their importance in selection. Based on these findings, the inbred lines GP88, PFSR29, PFSR32, PFSR51, BML6 and BML20 were identified as promising genotypes for use in further breeding programs aimed at improving yield under high-density planting.

Keywords: Maize, correlation, path analysis, genetic parameters, leaf angle, plant architecture, tassel morphology

Maize, with its widespread cultivation across millions of hectares worldwide, stands out as a staple food source and a key component of various industries. It has become an industrial crop due to its wide-ranging applications in food, feed, and as a raw material for various industries (Shiferaw *et al.*, 2011). To meet the growing food demand, crop productivity must be doubled by 2050, requiring an enhanced rate of 2.4% per year (Ray *et al.*, 2012). Consequently, improving yield has become a major objective in recent years. The main aim of any plant breeding program is to develop promising genotypes that exceed current ones in economic yield.

Genetic diversity is essential in plant breeding as it involves creating hybrids derived from lines of different origins typically show more heterosis than those produced from closely related strains. In the present study, multivariate analysis tools, such as D² statistics introduced by Mahalanobis and described by Rao (1952), along with PCA, were utilized for genetic divergence analysis. PCA is a valuable tool for evaluating phenotypic diversity, identifying genetically distant clusters of genotypes,

and pinpointing key traits that influence overall variation among genotypes. It provides a reliable means of discerning differences within the population by facilitating the natural grouping of genotypes. D² statistics developed by Mahalanobis (1936) is the most efficient tool for evaluating genetic divergence among a specified set of genotypes (Murthy and Arunachalam, 1966).

MATERIAL AND METHODS

The field experiment was carried out at the Maize Research Centre, Agricultural Research Institute, Rajendranagar, during the *kharif* season of 2023, using a RBD. Fifty-two genotypes were evaluated in two replications and each genotype was sown in two rows of 4 m length and a plant-plant and row-row spacing of 60 X 20 cm was followed. Data was collected from five randomly chosen plants for 15 characters viz., no. of leaves above the ear, no. of leaves below the ear, tassel branch number, tassel main axis length (cm), plant height (cm), ear height (cm), leaf angle of the leaf near the ear, leaf length of the leaf near the ear (cm), leaf width of

Table 1 : PCA for 17 yield and its attributing traits

Character	PC1	PC2	PC3	PC4	PC5
Days to 50% tasseling	0.0744	0.144	0.066	0.155	0.094
Days to 50% silking	0.0267	0.0393	0.0099	0.069	0.052
Plant Height (cm)	0.1143	-0.084	0.400	0.341	-0.050
Ear Height (cm)	0.0495	-0.091	0.415	0.301	-0.127
Leaf Angle	-0.7775	-0.313	0.181	0.173	0.102
No.of Leaves above ear	0.0624	-0.066	0.28	-0.0943	0.324
No.of Leaves below ear	0.0149	0.143	-0.053	0.301	-0.521
Total Number of Leaves	0.010	0.008	0.002	0.00	0.014
Leaf Length (cm)	-0.0747	0.1086	0.0700	0.322	-0.224
Leaf Width (cm)	0.0038	0.136	0.334	0.066	0.063
Tassel Branch Numbe	-0.38	-0.539	0.040	0.002	0.249
Tassel Main Axis Length (cm)	-0.0308	-0.139	-0.168	0.367	-0.234
No.of Kernels Per Row	-0.054	-0.127	0.196	-0.467	-0.456
No.of Rows Per Cob	0.174	0.605	0.157	0.0916	0.0087
Test Weight	0.1736	-0.145	0.506	-0.112	0.159
Shelling%	-0.367	0.251	-0.0079	-0.079	-0.080
Yield/Cob	-0.1407	0.180	0.288	-0.382	-0.414
Eigen value	4.23	2.601	2.128	1.683	1.453
%Var	39.88	15.42	13.60	9.126	6.12
Cum.Var	39.88	55.31	68.91	78.0	84.15

the leaf near the ear (cm), total no. of leaves per plant, no. of rows per cob, no. of kernels per row, 100 kernel weight(g), shelling percentage(%), grain yield per plant (g). While data on days to 50% tasseling and days to 50% silking was taken on plot basis. The values from five plants were averaged for each replication and the mean values were used for statistical analysis.

STATISTICAL ANALYSIS

For genetic divergence studies data were analyzed using Mahalanobis D^2 -statistics (Mahalanobis, 1936), and the intra-cluster and inter-cluster distances, along with the cluster mean and the contribution of each characteristic to the divergence, were calculated following the method proposed by Singh and Chaudhary(1985). Clustering of genotypes was done by using Tocher's method (Rao, 1952). The cluster distance was estimated by the formula given by Singh and Chaudhary (1977).PCA was performed using the "Facto Mine R" package (Husson *et al.*, 2020), "R" software version 4.1.3 (2022) was used to generate PCA biplot.

RESULTS AND DISCUSSION

Principle component analysis:

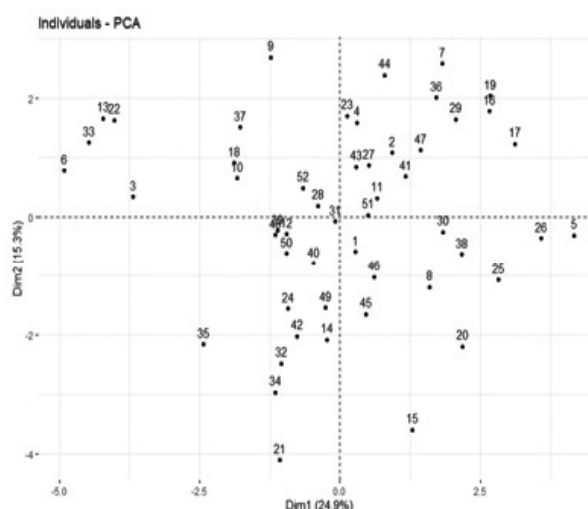
PCA was performed to minimize the complexity of multivariate data by identifying the key yield traits that make up the majority of variation and representing them as new variables (Principal Components, PCs). The aim was to capture as much information as possible in the first few components (Abdi & Williams, 2010; Stauffer *et al.*, 1985). Out of the seventeen traits included in the PCA, five principal components (PCs) with eigenvalues greater than 1 accounted for the majority of the variation, explaining approximately 88.62% of the variability present in the 52 inbred lines (Table 1). Therefore, these five PCs were prioritized for further analysis in this study. The percentage of total variation explained by PC1, PC2, PC3, PC4, and PC5 were 39.88%, 15.42%, 13.6%, 9.12%, and 6.12%, respectively (Table 1). A cutoff value of 0.3 for the absolute magnitude of eigenvector coefficients was used to identify important traits (Badu-Apraku *et al.*, 2006). Traits with coefficients greater than 0.3 were considered influential due to their substantial effect,

while traits with coefficients below 0.2 were deemed to have little influence on the overall variation (Laude & Carena, 2015; Sharifi *et al.*, 2018).

PC1 was responsible for 39.33% in the overall variation. The traits that had the most positive impact to PC1 included no. of rows per cob, plant height, test weight, days to 50% tasseling, no. of leaves above the ear, ear height and days to 50% silking. This component represented the weighted average of these traits, which are key determinants of yield, and therefore had the largest contribution to variation. PC2 explaining 15.42% of the total variation, showed negative coefficients for plant height and test weight. PC3, which explained 13.60% of the variation, was primarily influenced by the no. of rows per cob with a positive contribution. PC4 accounted for 9.12% of the variation, with major positive contributions from tassel main axis length, plant height, and leaf length. Lastly, PC5 explained 6.12% of the variation, with the number of leaves above the ear being the main contributing trait.

To examine the diversity among the genotypes (inbred lines), a biplot of the first two PCs was created. The inbred lines were spread across all quadrants of the biplot, indicating diversity in the data set. Lines located closer to the origin were considered to be more similar to each other and less diverse, while those further from the origin were regarded as more genetically distinct. Inbred lines GP82, PFSR 137, MGW 325, and GP 27, which were positioned far from the origin, were observed as the most diverse among the inbred lines.

Fig. 1 : Biplot dispersion graph of 52 maize inbred lines



The analysis of the principal components highlighted the significant contribution of traits such as no. of rows per cob, plant height, test weight, days to 50% tasseling, no. of leaves above the ear, ear height and days to 50% silking to the overall variability. Therefore, these traits should be prioritized in selection processes for future hybridization programs, followed by other traits. Traits that did not contribute significantly to the variation within the inbred lines under study can be excluded from similar analyses. Consequently, the traits that exhibit high positive or negative loadings in the PCA are particularly important in defining the clusters, as they have played a major role in contributing to genetic diversity.

Table 2. The inbred lines were grouped into clusters based on the Mahalanobis D² statistic.

Cluster number	No. of genotypes	Genotypes included
1	6	PFSR 137, GP 82, GP 94, GP 311, GP 125, BML 20
2	3	PFSR 49, 7 11, MGW 270
3	12	PFSR 32, BML 14, PFSR 132, PFSR 29, BML 6, GP 83, MGC 161, MGC 61, CML 286, PFSR 95, GP 170, CML 451
4	8	GP 35, MGW 419, MGC 331, GP 27, PFSR 90, PFSR 3, MGC 157, BML 45
5	5	MGW 360, PFSR 92, GP 2, 3122, PFSR 130
6	6	PFSR 1, PFSR 129, GP 88, BML 7, PFSR 51, PFSR 104
7	4	MGW 325, BML 11, D1, MGC 137
8	8	MGC 90, P852, MGW 316, MGC 89, MGW 334, GP 86, MGC 6, 3070

Table 3. Mean values for yield and yield-associated traits in various clusters of inbred lines.

Clusters	DTT	DTS	PH	EH	LA	LAE	LBE	TNL	LL	LW	TBN	TML	KPR	RPC	TW	SH	Y/c
1	59.08	61.25	116.31	47.10	31.33	5.05	5.64	10.69	57.49	7.09	10.38	22.67	13.54	19.97	15.33	73.80	77.91
2	61.00	63.33	135.33	51.92	29.67	5.53	4.63	10.17	48.87	6.06	4.41	21.66	13.37	8.83	20.5	66.42	53.35
3	63.71	65.96	128.27	53.94	41.50	5.72	5.72	11.43	54.25	7.20	8.72	20.57	15.49	17.20	21.88	71.14	73.67
4	59.93	62.25	117.66	49.59	40.00	5.58	4.64	10.21	48.4	6.56	5.73	19.73	18.69	11.23	22.13	70.02	71.63
5	55.70	58.20	139.13	61.10	45.00	5.62	5.14	10.76	53.18	6.12	4.62	22.36	14.28	9.35	28.9	68.87	63.31
6	62.00	64.25	125.31	53.48	28.00	5.52	5.89	11.41	58.02	6.62	6.81	21.22	15.38	13.23	22.75	69.59	75.31
7	53.63	55.75	108.13	45.06	65.00	5.23	4.75	9.98	45.16	6.35	8.24	21.13	20.94	11.89	19.75	70.4	69.79
8	60.00	62.19	104.63	41.53	47.31	4.98	4.51	9.49	47.43	6.89	9.49	19.56	15.77	14.06	20.88	72.27	72.61

Where, DTT-Days to 50% tasseling, DTS-Days to 50% silking, LAE-Number of leaves above the ear, LBE-Number of leaves below the ear, TBN-Tassel branch number, TML-Tassel main axis length (cm), PH-Plant height(cm), EH-Ear height(cm), LA-Leaf angle of the leaf near the ear, LL-Leaf length of the leaf near the ear(cm), LW-Leaf width of the leaf near the ear(cm), TNL-Total number of leaves per plant, RPC-Number of rows per cob, KPR-Number of kernels per row, TW-Test weight(g), SH-Shelling percentage(%), GYPP- Grain yield per plant (g).

Table 4. Average D2 values for intra-cluster (diagonal) and inter-cluster (above the diagonal) distances between the inbred lines.

CLUSTERS	1	2	3	4	5	6	7	8
1	40.9	197.32	181.06	294.57	316.33	323.66	749.57	835.22
2	197.32	49.59	191.17	259.09	323.17	300.63	662.38	767.29
3	181.06	191.17	112.32	212.51	166.87	240.93	666.03	787.01
4	294.57	259.09	212.51	193.87	308.60	392.6	1039.66	1189.66
5	316.33	323.17	166.87	308.96	0.00	203.01	420.41	586.94
6	323.66	300.63	240.93	392.6	203.01	306.66	512.32	706.52
7	749.57	662.38	666.03	1039.66	420.41	512.32	0.00	352.48
8	835.22	767.29	767.29	1189.66	586.94	706.52	352.48	0.00

CLUSTER ANALYSIS

The 52 inbred lines were categorized into eight clusters (Table 2 & Fig. 2), with Cluster 3 being the largest, containing twelve genotypes, followed by Clusters 4 and 8, each with eight genotypes, indicating genetic similarity among the lines in these clusters. The remaining clusters 1, 2, 5, 6, and 7, each contained fewer inbred lines, suggesting greater uniqueness among the genotypes in these groups. The clustering pattern revealed a significant level of genetic variation within the inbred lines, as they were distributed across eight distinct clusters.

Intra-cluster distances were generally smaller than inter-cluster distances (Table 4 & Fig. 3), indicating that inbred lines within the same cluster exhibited less genetic diversity. Cluster 6 had the largest intra-cluster distance (306.66), followed by Cluster 4 (193.87), Cluster 3 (112.32), Cluster 2 (49.59) and Cluster 1 (40.9). The intra-cluster distances for Clusters 5, 7, and 8 were recorded as 0.00, suggesting no diversity within these clusters. The highest inter-cluster distance (1189.66) was observed between Clusters 4 and 8, followed by distances between Clusters 4 and 7 (1039.66), Clusters 1 and 8 (835.22),

and Clusters 3 and 8 (787.01), signifying higher genetic variation between these clusters. Selecting inbred lines from Clusters 1, 4, 7, and 8 could be beneficial for hybrid development due to the significant genetic differences among them. The smallest inter-cluster distances were observed between Clusters 3 and 5 (166.87), Cluster 1 and 3 (181.06), and Cluster 3 and 2 (191.17), showing the genetic relatedness between the inbred lines within these clusters.

The mean values for different traits showed the greatest variation across clusters (Table 3). Cluster 3 exhibited the highest mean values for traits such as days to 50% tasseling, days to 50% silking, plant height, ear height, no. of leaves above the ear and leaf width. Cluster 1 had the highest mean values for yield per cob, shelling percentage, no. of rows per cob, tassel main axis length and tassel branch number. Cluster 6 recorded the highest means for the no. of leaves below the ear, total number of leaves, and leaf length. Cluster 7 had the highest mean for the no. of kernels per row, while Cluster 8 displayed the highest mean for leaf angle.

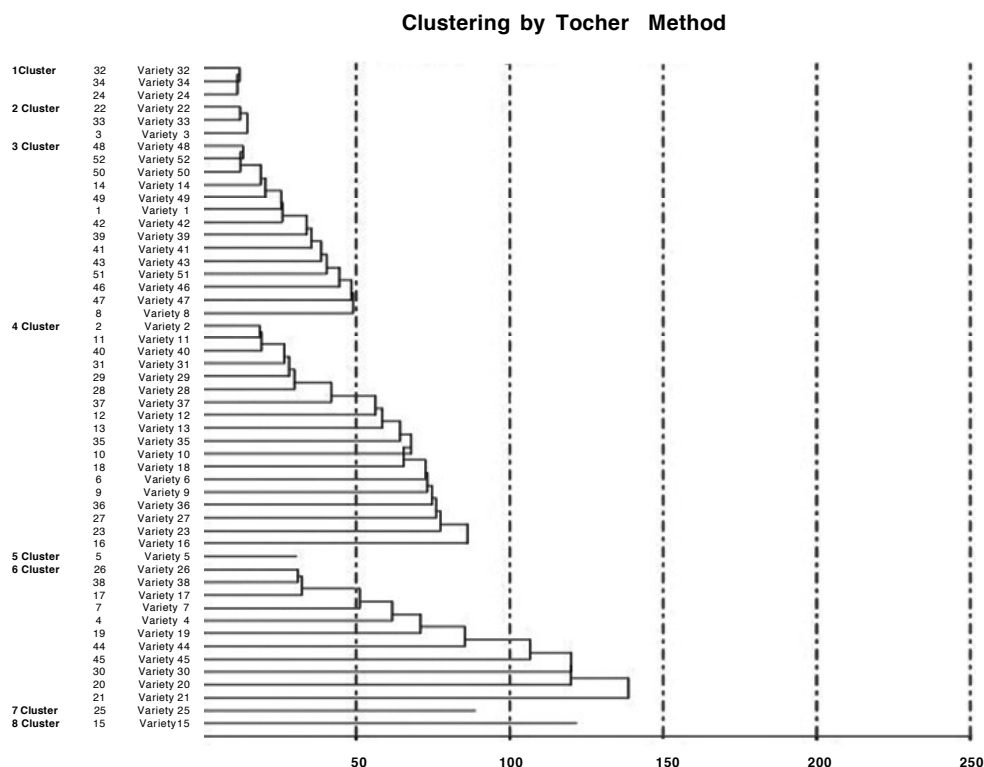


Fig. 2. Clustering pattern by Tocher's method

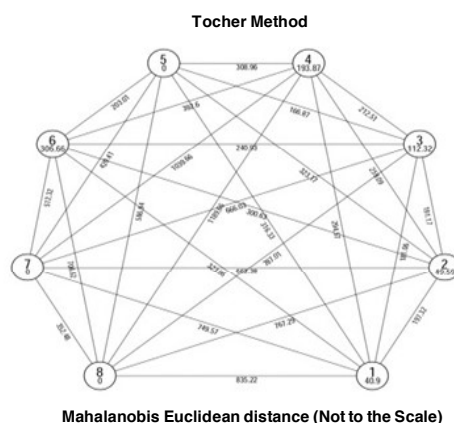


Fig. 3. Mahalanobis and Euclidean distances calculated using Tocher's meth

CONCLUSION

The results indicated that five principal components, with eigenvalues greater than 1, accounted for most of the variability, approximately 88.62%, in the traits. A two-dimensional biplot showed that inbreds GP82, PFSR 137, MGW 325, and GP 27 were located far away from the origin, indicating their diversity in comparison with other genotypes, they also formed distinct clusters in the D^2 statistics. D^2 statistics displayed that cluster III, comprises 12 maize genotypes followed by cluster IV (8), VIII (8), I (6), VI (6), V (5), VII (4) and II (3). Cluster VI had the highest intra-cluster distance of 306.6, while Clusters V, VII, and VIII showed zero intra-cluster distance, indicating homogeneity within those clusters. The greatest inter-cluster distance was observed between Cluster IV and VIII (1189.66), followed by Cluster IV and VII (1039.66), and Cluster I and VIII (835.22). Both PCA and D^2 statistics highlighted the significant contributions of traits such as the no. of rows per cob, plant height, test weight, days to 50% tasseling, tassel branch number, number of leaves above the ear, ear height, and days to 50% silking towards overall variability. The findings imply that the inbreds (GP 88, PFSR 137, MGW 325, and GP 27) studied are highly diverse, implying that crossing these inbreds from different clusters could result in high heterotic hybrids for increased grain yield.

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