

Original Research Article

Multivariate and Association Analysis for Yield and Yield attributing traits in Quality Protein Maize (QPM) Inbred Lines

ABSTRACT

The present investigation reveals the diversity existing among thirty inbred lines of **Quality Protein Maize (QPM)** in terms of yield and yield attributing traits. The study further elucidates the mutual association among the various morphological traits recorded among the inbred lines. The inbred lines were evaluated during the *Rabi* seasons of 2016-17, 2017-18 and 2018-19. The analysis of variance calculated over the mean performances of the inbred lines across three *rabi* seasons revealed significant differences among the inbred lines in terms of yield and yield attributing traits. The diversity among the inbred lines were further determined using cluster analysis which classified the inbred lines into 3 phylogenetically distinct groups. Additionally, a principal component analysis was performed which revealed three principal components (i.e., PC I, II and III) elucidating eighty six percent of the total observable variance among the inbred lines, with traits like grain yield, cob length, cob diameter, **number** of grain rows per cob, **number** of grains per row and **number** of grains per cob contributing to nearly half of the total variance explained by the **Principal Component Analysis (PCA)**. The correlation as well as path coefficient analysis performed for the various traits further indicated significant influence of morphological traits like cob length, cob diameter, **number** of grain rows per cob and **number** of grains per cob over the observable grain yield per plant. Overall, the observations from the current investigation can be helpful in identifying superior parental lines to be used in future hybrid maize development programs.

Keywords: Correlation, Cluster analysis, Inbred Lines, Path Analysis, Principal Component Analysis, Quality Protein Maize.

1. INTRODUCTION

Maize (*Zea mays* L.) is one of the most ubiquitous, widely grown and consumed cereals; ranking third in production next to wheat and rice. Besides, staple food for human consumption and quality feed for animals, maize serves as a basic raw material and ingredient to many industrial products like starch, oil, protein, alcoholic beverages, food sweeteners, pharmaceutical, cosmetic, film, textile, gum, package and paper industries etc. [1]. As a result of various uses of maize for different purposes and increasing population its demand is continually increasing. In terms of nutrition, **Quality Protein Maize** (QPM) is described as superior than its normal maize counterparts as it is induced with higher lysine and tryptophan contents [2]. Therefore, for food security along with higher nutritive value, QPM can play vital role in developing nations. Furthermore, maize being a cross pollinated plant, the crop shows immense heterotic potential which can be exploited by selecting superior inbred parents endowed with desirable morphological traits [3].

Development of superior inbred lines through various breeding programs can prove to be challenging unless the various inter-association among the desirable traits are known to the breeder. Additionally, availability of genetically divergent parents is a pre-requisite for any breeding program. Thus, inbred lines are the sine qua non and needs to be evaluated for their diverged gene pool for hybrid variety development. The genetic variation and its analysis are important components in planning and executing breeding programs [4].

Keeping, the above objectives in view, the current investigation aims at analyzing the diversity existing among a population of Quality Protein Maize inbred lines. Furthermore, the study also focuses on determining the influence of important morphological traits upon each other in terms of their phenotypic expression.

2. MATERIAL AND METHODS

This experimental material comprised of thirty Quality Protein Maize (QPM) inbred lines out of which thirteen of the inbred lines were obtained from the International Maize and Wheat Improvement Center (CIMMYT), Hyderabad, India and seventeen from the Indian Institute of Maize Research (IIMR), Ludhiana, India (Table 1). The experiment was carried out in Randomized Block Design (RBD) with three replications. The crop was grown during three (2016-17, 2017-18 and 2018-19) consecutive *rabi* seasons from mid of November to mid of April at Experimental Farm of University of Calcutta, Baruipur, South 24 Parganas, West Bengal, India (Latitude 22°52'N, Longitude 88° 52'E), and pooled performance of the thirty inbred lines across three *rabi* seasons were analyzed. The QPM inbred lines were sown with 2m of length and 60cm row to row and 20 cm plant to plant spacing. Normal inter culture operations were practiced throughout the growing period. Harvesting was performed manually when the plants had attained its full physiological maturity in each season.

Data was recorded on five randomly selected plants from each replication for eleven yield traits like- DT: Days to tasseling-50%, DS: Days to silking -50%, PH: Plant height (cm), EH: Ear height (cm), CL: Cob length (cm), CD: Cob diameter (cm), GRC: No. of grain rows per cob, GR: No. of grains per row, GRC: No. of grains per cob, GW:100 grain weight (gm) and GYP: Grain yield per plant (gm) was calculated.

Descriptive statistics, Correlation coefficient and path analysis were analyzed with SPAR 2.0 software. The cluster analysis was performed using the unweighted pair-group method with arithmetic averages (UPGMA) based on the Euclidean distance matrix [5] and principal component analysis was done according to [6]. Both the analysis were done using IBM SPSS 20.0 software package.

Table 1. Details of collected QPM Inbred Lines and their source

CODE	INBREDS	SOURCE	CODE	INBREDS	SOURCE
IM 1	CML 161	CIMMYT	IM 16	CML161-D	IIMR
IM 2	CML 163	CIMMYT	IM 17	CML163-D	IIMR
IM 3	CML171	CIMMYT	IM 18	CML165	IIMR
IM 4	CML193	CIMMYT	IM 19	CML167	IIMR
IM 5	CML502	CIMMYT	IM 20	CML169	IIMR
IM 6	CML504	CIMMYT	IM 21	CML170	IIMR
IM 7	CML505	CIMMYT	IM 22	CML179	IIMR
IM 8	CML507	CIMMYT	IM 23	CML451 Q	IIMR

IM 9	CML508	CIMMYT	IM 24	DMR QPM 102	IIMR
IM 10	CML509	CIMMYT	IM 25	DMR QPM 103	IIMR
IM 11	CML510	CIMMYT	IM 26	DMR QPM 03-104	IIMR
IM12	CML511	CIMMYT	IM 27	DMR QPM 03-113	IIMR
IM 13	CML537	CIMMYT	IM 28	DMR QPM 03-121	IIMR
IM 14	CML 153	IIMR	IM 29	H.K.I 163	IIMR
IM 15	CML154-2	IIMR	IM 30	H.K.I 193 - 1	IIMR

3. RESULTS AND DISCUSSION

In combined pooled analysis of variance, significant variations were observed among the inbred lines for all the traits under study (Table 2), suggesting the importance of their genetic value in order to identify the best genetic makeup for a particular condition and scope for effective selection. The inbred lines showed wider range of variability particularly higher range of variability was observed in the traits like number of grains per cob, grain yield per plant, plant height and ear height. When the genotype interaction was tested against the interaction components it was observed that the true genetic variance was present for all the traits evaluated. Such diversity can be utilized in future breeding programs. The, results were further confirmed by the pooled mean performance of thirty QPM inbred lines (Table 3). Similar results were also reported by [7, 8].

For improvement of more than one trait in any crop subsequently determining the mutual correlations among the various traits can prove to be helpful to the breeder. Such information can help the breeder in selecting more than one trait in each generation. In the current study important correlations among the various traits were observed (Table 4). The current analysis revealed that grain yield per plant was positively correlated at both genotypic and phenotypic levels with traits like cob length, cob diameter, number of grain rows per cob, number of grains per row, number of grains per cob and, hundred grain weight. Thus, grain yield per plant is determined through these yield attributing characters, which can be exploited for indirect selection for grain yield in future breeding programs. Similar observations were also made by [9, 10]. Interestingly, grain yield was also positively correlated with plant and ear height, similar studies were also made by other researchers [7, 11]. Another important yield attributing character is number of grains per cob and in the present study the trait was positively associated with cob length, cob diameter, number of grain rows per cob, number of grains per row. Similarly, all these yield attributing characters were interrelated while their interrelationships were observed. Thus, positive selection for these traits can indirectly help in increasing the number of grains observable in a single cob. Similar observations were also made by [8,10]. In case of the flowering traits, it was observed that days to 50 % tasseling was significantly and positively correlated with number of days to 50% silking. Thus, such observation indicates that synchronization in tasseling and silking which can be expected resulting in better pollination and grain set. Similar observations were also made by [8, 11].

The path coefficient analysis further corroborates the results observed in correlation analysis. The path analysis (Table 5) reveals that the yield attributing traits like number of grain rows per cob, number of grains per row and hundred grain weight showed high positive direct effects over grain yield; which was at par with results observed in correlation analysis. The observation further emphasizes the role of these three characters towards the improvement of grain yield in maize. Similar to the findings of our study, other researchers also have reported the same [12, 13, 14].

Table 2. Mean Sum of Squares from Combined Analysis of Variance for Grain Yield and its Attributing Traits in Thirty Inbred Lines of QPM

S.No.	Traits	Season (df=2)	Genotypes (df=29)	Genotypes × Season (df=58)	Pooled Error (df=174)
1.	DT (50%)	3788.27 **	164.39 *** ^{TT}	24.57 **	8.87
2.	DS (50%)	4147.24 **	168.82 *** ^{TT}	28.84 **	10.50
3.	PH (cm)	88856.55 **	5012.96 *** ^{TT}	989.37 **	470.35
4.	EH (cm)	52401.74 **	2112.23 *** ^{TT}	545.19 **	247.55
5.	CL (cm)	143.66 **	32.34 *** ^{TT}	10.02 **	2.40
6.	CD (cm)	76.08 **	9.84 *** ^{TT}	4.21 **	1.83
7.	GRC	67.27 **	15.15 *** ^{TT}	3.91**	1.43
8.	GR	2752.92 **	183.02 *** ^{TT}	63.93 **	15.13
9.	GC	531579.83 **	56248.65 *** ^{TT}	11927.69 **	3595.92
10.	GW(g)	97.84 **	118.76 *** ^{TT}	18.25 **	5.82
11.	GYP(g)	41162.24 **	2736.87 *** ^T	1360.11 **	246.74

*, ** = Significant at 0.05 and 0.01 levels, respectively; d.f. = degree of freedom

π , π indicates significant genotype MS at 5% and 1% probability when tested against the genotype x season interaction component

Table 3. Mean based traits variation in thirty QPM Inbred Lines

S. No	Characters	Pooled Mean	Range		S.E.	C.V.
			Min	Max		
1.	DT (50%)	90.12	81.44	99.00	0.80	1.54
2.	DS (50%)	93.63	85.02	102.78	1.04	1.92
3.	PH (cm)	169.35	116.22	220.07	7.31	7.48
4.	EH (cm)	86.36	58.38	118.21	5.47	10.98
5.	CL (cm)	13.26	9.52	17.07	0.56	7.27
6.	CD (cm)	12.68	10.97	16.30	0.48	6.59
7.	GRC	12.77	10.45	15.70	0.42	5.65
8.	GR	21.94	14.19	31.99	1.31	10.32
9.	GC	290.78	180.52	516.32	20.94	12.47
10.	GW(g)	25.59	19.39	34.65	0.84	5.69
11.	GYP(g)	77.92	53.34	115.35	5.81	12.92

Note: **DT:** Days to tasseling-50%, **DS:** Days to silking -50%, **PH:** Plant height (cm), **EH:** Ear height (cm), **CL:** Cob length (cm), **CD:** Cob diameter (cm), **GRC:** No. of grain rows per cob, **GR:** No. of grains per row, **GRC:** No. of grains per cob, **GW:**100 grain weight (gm) and **GYP:** Grain yield per plant (gm)

Table. 4. Genotypic and Phenotypic correlations of different traits of Thirty QPM Inbred Lines (pooled over 3 rabi seasons)

Trait s	Attri but e	DT	DS	PH	EH	CL	CD	GRC	GR	GC	GW	GYP
DT	G	1.000										
	P	1.000										
DS	G	0.986 **	1.000									
	P	0.957 **	1.000									
PH	G	0.231	0.187	1.000								
	P	0.151	0.100	1.000								
EH	G	0.233	0.186	0.746 **	1.000							
	P	0.139	0.098	0.697 **	1.000							
CL	G	0.123	0.045	0.279	0.306 *	1.000						
	P	0.120	0.040	0.181	0.180	1.000						
CD	G	-0.197	-0.235	0.174	0.188	0.511 **	1.000					
	P	-0.125	-0.155	0.075	0.087	0.495 **	1.000					
GRC	G	-0.085	-0.158	0.101	0.117	0.287	0.833 **	1.000				
	P	-0.091	-0.152	0.092	0.101	0.346 **	0.621 **	1.000				
GR	G	-0.259	-0.381 *	0.298	0.279	0.708 **	0.718 **	0.592 **	1.000			
	P	-0.236	-0.344 *	0.247	0.239	0.700 **	0.565 **	0.575 **	1.000			
GC	G	-0.176	-0.302	0.421 *	0.414 *	0.652 **	0.836 **	0.801 **	0.935 **	1.000		
	P	-0.165	-0.275	0.380 *	0.372 *	0.664 **	0.695 **	0.757 **	0.924 **	1.000		
GW	G	-0.060	-0.035	0.248	0.303	0.422 **	0.065	-0.501 **	0.055	-0.105	1.000	
	P	-0.060	-0.032	0.196	0.221	0.387 **	0.025	-0.344 *	0.090	-0.034	1.000	
GYP	G	-0.121	-0.209	0.587 **	0.579 **	0.786 **	0.721 **	0.486 **	0.843 **	0.825 **	0.403 **	1.000
	P	-0.136	-0.200	0.449 **	0.436 **	0.717 **	0.543 **	0.445 **	0.765 **	0.756 **	0.321 **	1.000

G = Genotypic correlation, **P** = Phenotypic correlation; *, ** = significant at 0.05 level and 0.01 levels, respectively

Note: **DT:** Days to tasseling-50%, **DS:** Days to silking -50%, **PH:** Plant height (cm), **EH:** Ear height (cm), **CL:** Cob length (cm), **CD:** Cob diameter (cm), **GRC:** No. of grain rows per cob, **GR:** No. of grains per row, **GRC:** No. of grains per cob, **GW:** 100 grain weight (gm) and **GYP:** Grain yield per plant (gm)

Table.5. Pooled path analysis for different agromorphological traits in Thirty QPM Inbred Lines										
Traits	DT(50 %)	DS(50%)	PH(cm)	EH(cm)	CL(cm)	CD(cm)	GR/C	GR	GC	GW(g)
DT(50%)	-0.210	0.165	0.042	0.002	0.005	-0.004	-0.002	-0.075	-0.047	-0.012
DS(50%)	-0.201	0.172	0.028	0.001	0.002	-0.005	-0.003	-0.109	-0.079	-0.006
PH(cm)	-0.032	0.017	0.276	0.011	0.028	0.010	0.005	0.127	0.128	0.060
EH(cm)	-0.029	0.017	0.243	0.013	0.028	0.010	0.005	0.129	0.126	0.070
CL(cm)	-0.025	0.007	0.173	0.008	0.045	0.015	0.007	0.222	0.190	0.075
CD(cm)	0.026	-0.027	0.090	0.004	0.022	0.031	0.013	0.179	0.199	0.005
GRC	0.019	-0.026	0.061	0.003	0.016	0.019	0.021	0.182	0.217	-0.067
GR	0.050	-0.059	0.110	0.005	0.032	0.017	0.012	0.317	0.264	0.017
GC	0.035	-0.047	0.123	0.005	0.030	0.021	0.016	0.293	0.286	-0.007
GW(g)	0.013	-0.005	0.085	0.005	0.017	0.001	-0.007	0.028	-0.010	0.194
Residual are 0.245; bold diagonal value represents direct effects										

For determining the diversity among the inbred lines, a cluster analysis was performed, (Table 6A) which revealed that the genotypes can be classified into three clusters based on seventy percent similarity coefficient, as well as for a clear understanding, a dendrogram was drawn through hierarchical cluster analysis based on **unweighted pair-group method with arithmetic averages** (UPGMA) and Euclidean distance (Fig.1). Appreciable amount of variation among the cluster means for different characters presented in (Table 6B) also suggested the existence of diversity. Cluster number I and II was observed to be the largest consisting of forty-seven and forty-three genotypes respectively whereas, cluster number III was the smallest group comprising of only three inbred lines (Table 7).

The analysis further revealed that the cluster number I and III exhibited maximum inter cluster distance indicating that genotypes belonging to cluster number I are significantly different from the inbred lines belonging to cluster number III. Such diversity can be exploited in future breeding programs aimed at developing heterotic hybrids. The cluster analysis further revealed that the average grain yield per plant was maximum for the inbreds belonging to cluster number III. Similarly, other important yield attributing characters like number of grains per cob, number of grains per row, number of grain rows per cob, cob diameter, cob length were also maximum for the inbreds belonging to cluster III. In cluster number III the inbred IM 14 outperformed the other two inbreds in terms of grain yield per plant and yield related traits like, number of grains per row, number of grains per cob, number of grain rows per cob. Thus, the inbred can be designated as promising parent in future breeding programs.

For determining the contribution of individual traits towards the overall variation observed in the cluster analysis; a **Principal Component Analysis** (PCA) was performed (Table 8). The PCA revealed cumulative variance of 86 % to the total variation observed in the population which could be explained through three principal components i.e., PC 1, 2 and 3 exhibiting eigen value greater than one, as observable in scree plot (Fig.2) The first principal component (PC1) explaining 48 % of the total variance was comprised of important traits like grain yield per plant, number of grains per row, number of grain rows per cob, cob diameter, cob length, plant height, ear height. The observation revealed that yield along with major yield attributing characters are represented in PC 1. Thus, maximum variation for yield and yield attributing traits can be expected within the current population under study. The analysis further revealed that flowering traits like days to 50 % tasseling and days to 50% silking were constituted in the second principal component (PC 2), whereas (PC 3) was comprised of single morphological trait i.e., grain weight. Overall, the maximum proportion of variability with higher positive factor loading was seen on first principal component. Our findings were in accordance with the earlier studies made by [15, 16, 17].

Table 6. Inter cluster distance and cluster means			
<i>A. Inter Cluster distance among Quality Protein Maize Inbreds</i>			
Cluster	I	II	III
I	0	87.448	259.869
II	87.448	0	177.243
III	259.869	177.243	0
<i>B. Cluster means of the respective 11 characters</i>			
Characters	I	II	III
DT (50%)	88.27±4.22	92.23±3.65	89.62±0.49
DS (50%)	91.59±4.22	96.26±3.22	91.80±0.67
PH(cm)	162.20±19.13	168.39±21.15	206.84±9.51
EH(cm)	81.29±13.29	86.35±12.92	110.14±5.99
CL(cm)	13.24±1.52	12.57±1.62	16.36±0.95
CD(cm)	12.74±0.64	12.11±0.54	14.90±1.04
GRC	13.01±0.98	11.93±0.71	15.26±0.31
GR	23.96±1.99	17.87±2.02	30.18±1.33
GC	311.34±21.74	225.60±22.51	477.31±28.96
GW(g)	25.02±3.71	26.55±3.43	24.09±2.11
GYP(g)	80.15±12.59	67.77±11.60	111.51±4.41
<i>Note: Values are the mean ± SD</i>			
Note: DT: Days to tasseling-50%, DS: Days to silking -50%, PH: Plant height (cm), EH: Ear height (cm), CL: Cob length (cm), CD: Cob diameter (cm), GRC: No. of grain rows per cob, GR: No. of grains per row, GRC: No. of grains per cob, GW:100 grain weight (gm) and GYP: Grain yield per plant (gm)			

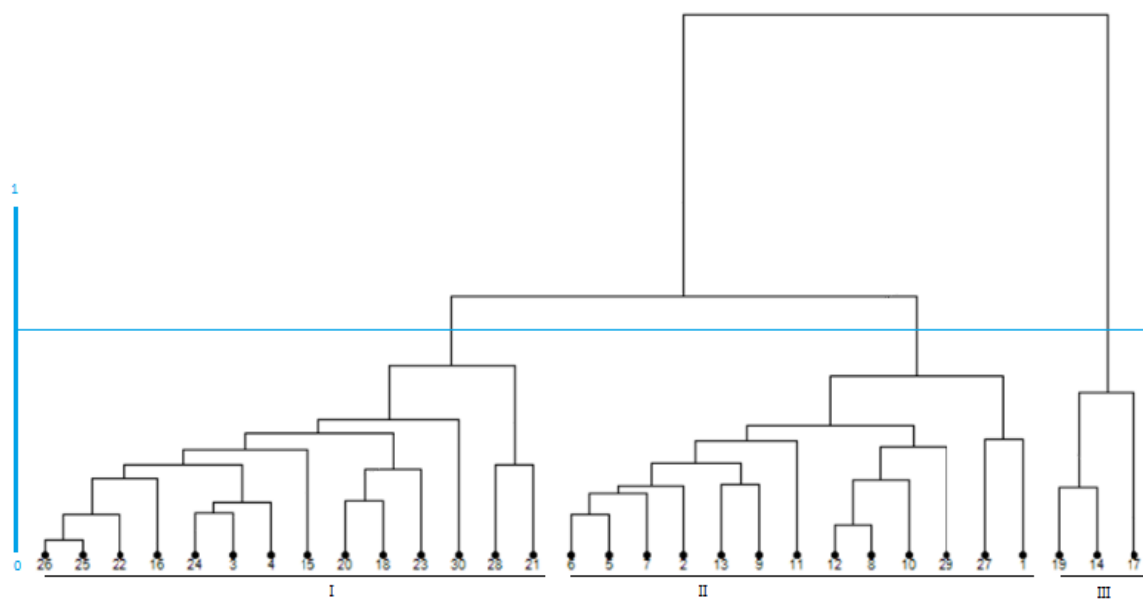


Fig. 1: Dendrogram showing the clustering pattern of 30 QPM inbred lines using hierarchical cluster analysis (UPGMA Method and Euclidean distance)

Table 7. Distribution of 30 QPM Inbred lines among three clusters			
Cluster No.	No. of inbred lines	Percentage of contribution	Inbred lines
I	14	47	IM 3, IM 4, IM 15, IM 16, IM 18, IM 20, IM 22, IM 23, IM24, IM 25, IM26, IM 30, IM 21, IM 28
II	13	43	IM 1, IM 2, IM 5, IM 6, IM 7, IM 8, IM 9. IM 10, IM 11 IM 12, IM 13, IM 27, IM 29
III	3	10	IM 14. IM 17, IM 19

Table 8. PC of different agro-morphological traits with Factor loadings in QPM Inbred Lines (Pooled over 3 rabi seasons)

PARAMETERS	COMPONENTS		
	1	2	3
Eigenvalue	5.309	2.520	1.696
% of Variance	48.259	22.905	15.419
Cumulative %	48.259	71.164	86.584
TRAITS			
DT (50%)	-0.117	0.775	0.589
DS (50%)	-0.209	0.790	0.548
PH(CM)	0.718	0.533	-0.118
EH(CM)	0.719	0.542	-0.140
CL(CM)	0.819	0.351	-0.135
CD(CM)	0.788	-0.221	0.173
GRC	0.667	-0.392	0.547
GR	0.883	-0.246	-0.020
GC	0.923	-0.263	0.192
GW	0.244	0.503	-0.771
GYP	0.922	0.093	-0.190

DT: Days to tasseling-50%, DS: Days to silking -50%, PH: Plant height (cm), EH: Ear height (cm), CL: Cob length (cm), CD: Cob diameter (cm), GRC: No. of grain rows per cob, GR: No. of grains per row, GRC: No. of grains per cob, GW:100 grain weight (gm) and GYP: Grain yield per plant (gm)

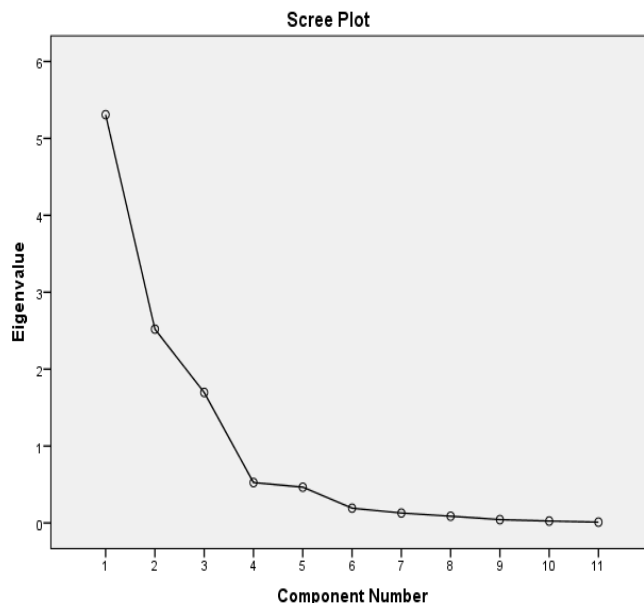


Fig. 2: Scree Plot showing variation of eigen values

4. CONCLUSION

The current study revealed significant effect of environment upon all the traits evaluated. Thus, variation in performance of inbred lines with respect to seasonal variations must be taken under consideration during future breeding programs. Additionally, by substituting the genotype x environment interaction component from the genotype MS it was observed that significant genetic differences were present among the inbreds in terms of all the traits evaluated. Such genetic differences can be exploited by maize breeders during future breeding programs. The diversity observed was further elucidated through cluster analysis which classified the inbred lines into separate groups. Inbreds belonging to different clusters can be selected for hybridization in order to ensure maximum genetic diversity between the two parents for achieving high heterosis for yield and its attributing traits. The current study further revealed the inter association among important agro-morphological traits which can be utilized in future breeding programs aimed at parental line development in maize. The study revealed significant influence of yield attributing traits like number of grains per row, number of grain rows per cob, number of grains per cob, hundred grain weight, cob length and cob diameter upon grain yield per plant, such correlations can be exploited for indirect selection of yield through one or more of the attributing characters. Lastly, the principal component analysis revealed that more than half of the variation observed among the inbred

lines analyzed in the current study was contributed by important morphological traits having direct or indirect influence upon yield. Thus, the current population of inbred lines can prove to be instrumental for developing heterotic F1 hybrids in maize.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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