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RESEARCH ARTICLE

Assessing Genetic Diversity of Newly Developed Winter Maize (*Zea mays* L.) Inbred Lines

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Understanding the genetic diversity among the breeding materials is fundamental consideration for any crop improvement programme. Ninety seven newly developed winters maize inbred lines along with thirteen released inbreds were evaluated to assess the genetic diversity based on morphological traits. Analysis of variance revealed significant differences for 13 characters studied. All the inbred lines were grouped into fifteen clusters with ten solitary clusters. The D^2 statistics displayed that cluster I being largest group, with maximum inbred lines (37) followed by Cluster II (24), Cluster III (16) Cluster IV (14) and Cluster V (9). The maximum inter-cluster distance was observed between cluster V and cluster XV (26.96) followed by cluster IV and XV (26.12), cluster V and XII (24.55) suggesting higher probability of heterotic combinations if parents selected from these pairs of groups. Cluster IV has the highest intra-cluster distance (11.59). Maximum genetic divergence as per cent was contributed by 100 kernel weight (39.45) followed by days to anthesis (22.64), grain filling duration (10.31) and grain yield (10.50). On the basis of per se performance, intra and inter cluster distance, inbred lines IMLSB-2005, IMLSB-1000-2, IMLSB-182-1, IMLSB-719-1, IMLSB-164-1, IMLSB-457-2, IMLSB-2083, IMLSB-1298-2, IMLSB-1298-5 and IMLSB-246-2 were identified that might be used in maize improvement programme to develop superior hybrid combinations.

Key Words: Diversity, D^2 analysis, Inbred lines, Principal component analysis, Winter maize

Introduction

Maize (*Zea mays* L.) is one of the most important cereal crops of the world known as "Queen of Cereals" because of its highest yield potential and wider adaptability which plays a pivotal role in food security of many developing countries. Maize is currently produced on nearly 160 million hectares in 125 developing countries with a production of 850 million tonnes (Anonymous, 2016). In India, maize is emerging as third most important crop after rice and wheat. It is worldwide important crop used as a food, feed and as a source of raw material for several industries. This is also considered as a model genetic organism with immense genetic diversity (Prasanna, 2012). The introduction of new hybrids resilient to changing climates *viz*., low temperature during winter season, off-season disease and pests with high productivity has made maize a profitable alternative for small farmers in U.P., Bihar, Andhra Pradesh and Karnataka. In India, winter maize could help to meet the industrial requirements consistently throughout the year. Winter maize has emerged as an important crop in the non-traditional areas including major winter maize growing states *i.e.* Andhra Pradesh, Bihar, Tamil Nadu, Karnataka, Maharashtra and West Bengal. There is potential to increase the production of maize by increasing the area under winter maize in the coming years as winter maize has a higher yield at 4 MT/hectare as against 2.5MT/hectare for *Kharif* maize. Due to inbuilt tolerance for biotic and abiotic stresses under winter ecology the winter maize inbred lines with high yield potential can be utilised for developing more productive hybrids and provide more adaptability to different agro-climatic conditions. Since opportunities are limited for further expansion of maize area, future increases in maize supply will be achieved through the intensification and commercialization of current maize production systems (Krishnamoo and Mohan, 2017). Genetic improvement of a crop is pivoted on the strength of genetic diversity within the crop species.

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Assessing genetic diversity and relatedness among breeding materials has a preponderant role in a breeding program. Development of improved inbred lines and identifying suitable parental combinations to generate high performing hybrids is the leading task of maize breeders (Semagn *et al,* 2012). Morphological traits are the functional manifestation of underlying genetic constitution of an organism hence they constitute an important set of markers to assess the genetic diversity. Therefore, characterization of genetic diversity of maize germplasm or inbred lines is of great importance in hybrid maize breeding (Xia *et al.,* 2005). For effective management of genetic diversity, there is need of well-characterized germplasm and genetic pools classified into different clusters based on genetic diversity (Wende *et al,* 2013). Quantification of magnitude of genetic diversity among the germplasm has become possible with the help of advance biometrical technique; *viz*., multivariate analysis, based on D^2 statistics and principal component analysis (PCA). In view of the above, present investigation was conducted to assess the genetic diversity among newly developed inbred lines of winter maize based on morpho-physiological and yield traits using D^2 statistics and principal component analysis (PCA).

Material and Methods

Ninety seven new inbred lines of winter maize were developed through pedigree method from the diverse source of germplasm. These inbred lines along with thirteen released inbred lines (Table 1) were evaluated in randomized complete block design (RCBD) with three replications for morpho-physiological and yield traits during winter season of 2014-15 and 2015-16 at Regional Maize Research and Seed Production Centre, (ICAR-IIMR), Begusarai-851129 (Bihar). Each inbred line was sown in a single row of 4 m spaced at 60 cm with interplant distance of 25 cm. All recommended agronomic practices were followed to raise a good crop. Observations were recorded on thirteen morpho-physiological and yield characters**.** In each line, five plants were randomly selected for recording observations on plant height, ear height, ear length, ear girth, kernel rows per ear, kernels per row and 100-kernel weight. Observations on days to anthesis, days to silk, anthesis–silking interval (days), grain filling duration (days) and days to maturity were recorded on plot basis. The period after pollination to 75 per cent dry husk maturity was considered as grain filling duration. Observations were also recorded on three qualitative traits i.e. kernel colour (Y= yellow, O= orange, LY= light yellow, W= white, CW= creamy white), kernel type (D= dent, F= flint, SF= semi-flint, SD= semi-dent) and kernel size (S= small, M= medium, B= bold). Pooled mean over years was utilized for analysis of variance (ANOVA) and used to quantify the genetic differences among the genotypes. Data were subjected to Mahalanobis (1936) D^2 statistical analysis extended by Rao (1952) and Principal Component Analysis (Pearson*,* 1901*)*. Intra-cluster and inter-cluster distance, cluster mean and contribution of each trait to the divergence were estimated as suggested by Singh and Chaudhary (1985) using INDOSTAT software and significant means were compared using significant differences at P 0.05 and 0.01.

Results and Discussion

A significant variability was exhibited among the inbred lines for morphological characters as well as yield components. Variability for grain type (flint, semi dent, dent), grain colour (white, creamy white, yellow and orange), kernel size (small to very bold) and 100 kernel weight (14.73-38.27g) was exhibited by the genotypes under study which play a key role for targeted trait improvement. The analysis of variance (Table 2) revealed the significant differences among the genotypes for all the characters studied indicating that the experimental materials were genetically divergent to each other. This shows that there is sufficient scope for selection of lines with specific traits amongst the available inbred lines aimed to enhance the genetic potential of maize.

All the inbred lines were grouped into fifteen (Table 3) clusters having variable number of entries. Cluster I with 37 genotypes had the maximum number of genotypes followed by cluster II with 24, cluster III with 16, cluster IV with 14 and cluster V with 9 genotypes while rests of the clusters were solitary entry clusters demonstrating the impact of selection presence in increasing the genetic diversity. The formation of solitary clusters may be due to total isolation preventing the gene flow or intensive natural/ human selection for diverse adaptive complexes. The clustering pattern of inbred lines revealed that the inbred lines had significant genetic divergence among themselves. Similar results were reported by Bhusal *et al*. (2016) and Ranawat *et al.* (2013). In a similar study, Shrestha (2016) grouped sixty maize inbreds into six major groups through cluster and principal component analysis and concluded that the presence of high level of diversity among the inbred lines grouped

Table 1. Detail of winter maize inbred lines, source of germplasm and kernel characters

S. No.	Name of Inbred line	Source of germplasm	Institute of source germplasm	Kernel colour	Kernel type	Kernel size	S. No.	Name of Inbred line	Source of germplasm	Institute of source germplasm	Kernel colour	Kernel type	Kernel size
1	IMLBG-3-1	VH1266	CIMMYT	O	F	M	56	IMLBG-763-1	HQPM-1F2	IIMR	O	F	M
2	IMLBG-13-1	VH112935	CIMMYT	LY	SD	S	57	IMLBG-800-1	Pro4794F2	IIMR	O	SF	M
3	IMLBG-16-1	VH112940	CIMMYT	W	F	M	58	IMLBG-801-2	Bio-9681F2	IIMR	O	SF	M
4	IMLBG-22-1	ZH114207	CIMMYT	O	SF	S	59	IMLBG-814-2	Bio-9681F2	IIMR	O	SD	B
5	IMLBG-23-2	VH112922	CIMMYT	O	F	M	60	IMLBG-825-2	Bio-9681F2	IIMR	O	SF	M
6	IMLBG-43-2	ZH112700	CIMMYT	O	F	B	61	IMLBG-961-1	NK6240F2	IIMR	O	SF	M
7	IMLBG-46-1	VH101421	CIMMYT	O	SD	M	62	IMLBG-975-2	NK6240F2	IIMR	O	F	M
8	IMLBG-49-2	VH112450	CIMMYT	O	D	M	63	IMLBG-976-2	P3522F2-1	IIMR	O	F	M
9	IMLBG-55-2	VH112934	CIMMYT	Y	F	VS	64	IMLBG-1000-2	P3522F2	IIMR	O	F	B
10	IMLBG-58-1	VH112948	CIMMYT	O	F	M	65	IMLBG-1052-1	NQPM-Pool	IIMR	O	F	M
11	IMLBG-66-1	ZH111659	CIMMYT	O	F	S	66	IMLBG-1333	WNC18242	IIMR	O	SF	B
12	IMLBG-81-1	ZH112645	CIMMYT	O	F	B	67	IMLBG-1334	WNC18261	IIMR	O	SF	M
13	IMLBG-91-2	ZH112606	CIMMYT	O	F	B	68	IMLBG-1364	WNC19053	IIMR	O	SF	S
14	IMLBG-93-2	VH112933	CIMMYT	Y	F	M	69	IMLBG-1382	WNC19207	IIMR	Y	SD	B
15	IMLBG-100-1	VH11152	CIMMYT	Y	F	M	70	IMLBG-2005	ZL11258	CIMMYT	Y	SD	B
16	IMLBG-103-1	ZH112656	CIMMYT	Y	SF	M	71	IMLBG-2025	VL054794	CIMMYT	W	SD	B
17	IMLBG-106A-2	ZH116117	CIMMYT	Y	F	M	72	IMLBG-2032	VL05616	CIMMYT	W	SF	M
18	IMLBG-114-1	VH1275	CIMMYT	O	SD	B	73	IMLBG-2039	VL108305	CIMMYT	Y	SF	M
19	IMLBG-123-1	VH101429	CIMMYT	O	F	B	74	IMLBG-2045	VL105544	CIMMYT	Y	SD	B
20	IMLBG-141-2	ZH111929	CIMMYT	O	F	M	75	IMLBG-2051	VL108880	CIMMYT	O	F	M
21	IMLBG-147-1	ZH111450	CIMMYT	O	D	B	76	IMLBG-2077	VL108727	CIMMYT	Y	SF	B
22	IMLBG-156-2	ZH12421	CIMMYT	O	D	B	77	IMLBG-2083	VL1018391	CIMMYT	O	SF	M
23	IMLBG-164-1	ZH111948	CIMMYT	Y	SD	B	78	IMLBG-2086	VL0512418	CIMMYT	O	SF	M
24	IMLBG-173-2	ZH111948	CIMMYT	O	F	B	79	IMLBG-2092	ZL11884	CIMMYT	O	F	M
25	IMLBG-182-1	VH112552	CIMMYT	O	F	B	80	IMLBG-2093	ZL124332	CIMMYT	LY	SD	B
26	IMLBG-183-1	VH12280	CIMMYT	O	F	M	81	IMLBG-2094	ZL124478	CIMMYT	O	F	M
27	IMLBG-197-1	VH121028	CIMMYT	Y	SF	S	82	IMLBG-2096	ZL124351	CIMMYT	O	F	M
28	IMLBG-201-1	VH121038	CIMMYT	O	F	M	83	IMLBG-2097	ZL124430	CIMMYT	Y	SD	M
29	IMLBG-210-2	VH101411	CIMMYT	O	F	M	84	IMLBG-2102	ZL124372	CIMMYT	Y	SF	M
30	IMLBG-219-2	VH112650	CIMMYT	Y	F	B	85	IMLBG-2103	ZL124384	CIMMYT	O	F	B
31	IMLBG-231-1	ZH116132	CIMMYT	Y	W	M	86	IMLBG-2108	ZL124485	CIMMYT	O	F	M
32	IMLBG-246-2	VH113021	CIMMYT	O	D	B	87	IMLBG-2115	ZL124382	CIMMYT	Y	F	S
33	IMLBG-254-1	VH112906	CIMMYT	Y	SD	M	88	IMLBG-2128	ZL11863	CIMMYT	W	SD	M
34	IMLBG-269-1	VH112993	CIMMYT	O	F	B	89	IMLBG-2132	ZL11589	CIMMYT	W	SD	M
35	IMLBG-274-1	VH1279	CIMMYT	Y	F	M	90	IMLBG-2135	ZL126611	CIMMYT	W	SD	M
36	IMLBG-282-2	VH121043	CIMMYT	Y	F	M	91	IMLBG-2145	ZL114846	CIMMYT	W	SD	M
37	IMLBG-285-2	VH121055	CIMMYT	O	F	B	92	IMLBG-2147	ZL113897	CIMMYT	W	SF	S
38	IMLBG-301-2	VH12157	CIMMYT	O	F	S	93	IMLBG-2150	ZL113672	CIMMYT	W	SD	M
39	IMLBG-306-2	VH121082	CIMMYT	O	D	B	94	IMLBG-2152	ZL113792	CIMMYT	W	SF	M
40	IMLBG-310-1	AH1222	CIMMYT	O	F	B	95	IMLBG-2166	VL109126	CIMMYT	O	D	B
41	IMLBG-310-2	AH1222	CIMMYT	O	SD	M	96	IMLBG-1298-2	900MF2	IIMR	Y	SD	M
42	IMLBG-334B-1	Check4	CIMMYT	O	F	M	97	IMLBG-1298-5	900MF2	IIMR	O	SD	B
43	IMLBG-343-2	VH126	CIMMYT	Y	F	M	98	HKI-193-1	HKI-193-1	HAU	Y	F	m
44	IMLBG-406-1	VH112650	CIMMYT	O	SD	B	99	HKI-163	HKI-163	HAU	O	SD	M
45	IMLBG-406-2	VH112650	CIMMYT	O	SD	B	100	BML-6	BML-6	ANGRAU	Y	SD	M
46	IMLBG-428-2	VH1293	CIMMYT	Y	SD	B	101	BML-7	BML-7	ANGRAU	O	SD	B
47	IMLBG-457-2	ZH111688	CIMMYT	y	SD	B	102	LM-16	LM-16	PAU	O	SD	B
48	IMLBG-481-2	VH1277	CIMMYT	O	F	M	103	LM-13	LM-13	PAU	O	SD	B
49	IMLBG-507-1	VH11124	CIMMYT	O	D	M	104	LM-14	LM-14	PAU	O	F	S
50	IMLBG-592-2	ZH112687	CIMMYT	O	F	B	105	HKI-1105	HKI-1105	HAU	Y	SF	M
51	IMLBG-617-1	CP838F2	IIMR	O	SF	M	106	HKI-1128	HKI-1128	HAU	Y	SF	M
52	IMLBG-667-1	S 900MF2	IIMR	Y	S	M	107	UMI-1200	UMI-1200	TNAU	Y	SF	M
53	IMLBG-678	Bio-9637F2	IIMR	Y	SF	B	108	UMI-1201	UMI-1201	TNAU	Y	D	B
54	IMLBG-719-1	DKC9081F2	IIMR	O	D	B	109	UMI-1210	UMI-1210	TNAU	O	SD	M
55	IMLBG-722-1	DKC9081F2	IIMR	O	F	M	110	BML-15	BML-15	ANGRAU	O	SF	M

Table 2. Analysis of variance for maturity and yield traits in winter maize inbred lines

Source of Variation	df	Mean Squares												
		DOA	DOS	ASI	DM	GFD	PH	EH	EL	EG	KR	K/R	100KW	GY(q/h)
Replicate	2	0.039	1.973	2.239	6.221	3.648	61.131	0.820	2.036	1.330	2.583	15.626	0.094	53.135
Treatments	109	81.625**	84.327**	3.613**	102.390**	95.847**	1461.872**	370.505**	10.280**	3.634**	6.601**	56.162**	67.601**	98.118**
Error	218	1.226	2.025	0.872	3.928	5.349	79.266	31.505	1.770	0.861	1.674	5.730	0.781	3.434

*Significant at 5% level, ** Significant at 1% level
DOA= Days to anthesis, DOS= Days to silk, ASI=Anthesis-Silking interval, GFP= Grain filling duration, DM= Days to maturity, PH= Plant height, EH= Ear height, EL= Ear length, EG= Ear girth KR=kernel rows per ear, K/R= Kernels per row, 100kw= 100 kernel weight, GY(q/h)= Grain yield q/h,

Table 3. Distribution of 110 maize inbred lines in 15 different clusters

Name of Cluster	No. of inbred lines	Name of inbred lines present in a cluster
I	37	IMLSB-173-2, IMLSB-231-1, IMLSB-667-1, IMLSB-269-1, IMLSB-2115, IMLSB-2094, IMLSB-800-1, IMLSB-976-2, IMLSB-310-1, IMLSB-2039, IMLSB-507-1,IMLSB-274-1, IMLSB-201-1, IMLSB-678, IMLSB-2103, IMLSB-722-1, IMLSB-2102, IMLSB-2093, IMLSB-975-2, IMLSB-814-2, IMLSB-49-2, IMLSB-2032, IMLSB-43-2, IMLSB-2147, IMLSB-23-2, IMLSB-343-2, IMLSB-1052-1, BML-6, IMLSB-428-2, IMLSB-2135, IMLSB-763-1, BML-7, IMLSB-254-1, IMLSB-825-2, IMLSB-592-2, IMLSB-2086, IMLSB-2145
II	24	IMLSB-197-1, IMLSB-2150, IMLSB-1334, IMLSB-961-1, IMLSB-100-1, IMLSB-481-2, IMLSB-22-1, IMLSB-16-1, IMLSB-81-1, IMLSB-617-1, LM-16, IMLSB-13-1, IMLSB-2108, IMLSB-147-1, IMLSB-103-1, IMLSB-114-1, IMLSB-2152, IMLSB-58-1, IMLSB-91-2, IMLSB-2045, IMLSB-106A-2, IMLSB-46-1, IMLSB-156-2, HKI-193-1
III	16	IMLSB-801-2, IMLSB-2132, IMLSB-2096, IMLSB-334B-1, IMLSB-1333, IMLSB-219-2, IMLSB-210-2, IMLSB-306-2, IMLSB-2097, IMLSB-2166, IMLSB-310-2, IMLSB-1382, IMLSB-2092, IMLSB-183-1, IMLSB-141-2, IMLSB-282-2
IV	14	IMLSB-2051, IMLSB-2077, IMLSB-406-2, IMLSB-406-1, IMLSB-2083, IMLSB-164-1, IMLSB-457-2, IMLSB-246-2, IMLSB-1298-5, IMLSB-1298-2, IMLSB-301-2, IMLSB-2128, IMLSB-285-2, IMLSB-93-2
V	9	UMI-1201, UMI-1210, LM-13, UMI-1200, BML-15, HKI-163, HKI-1128, HKI-1105, LM-14
VI	1	IMLSB-182-1
VII	1	IMLSB-719-1
VIII	1	IMLSB-123-1
IX	1	IMLSB-1000-2
X	1	IMLSB-3-1
XI	1	IMLSB-2025
XII	1	IMLSB-55-2
XIII	1	IMLSB-2005
XIV	1	IMLSB-66-1
XV	1	IMLSB-1364

into divergent clusters. The clustering pattern (Fig. 1) revealed that some genotypes that originated from the source germplasm of different geographical regions had been grouped into the same cluster. On the other hand, the genotypes that originated in one region had been distributed into different clusters, indicating that genotypes with same geographic source germplasm could have undergone change for different characters under selection. This could be due to selection pressure, genetic drift and environment, which created greater diversity rather than genetic distance during the developmental phase (Marsan *et al*, 1998; Senior *et al*, 1998; Wende *et al*, 2013). Discrepancies in classification of germplasm based on pedigree relatedness were earlier reported by Dhliwayo *et al* (2009) and Yang *et al.* (2011). These might resulted due to the fact that majority of the inbred lines involved in the current study were developed from maize germplasm obtained from CIMMYT. Therefore, there might be exchange of breeding materials among different CIMMYT breeding programs, justifying the alignment of some inbred lines from different origin in the same clusters.

The intra and inter-cluster distance ($D = \sqrt{D^2}$) values were worked out from divergence analysis and are listed in Table 4. It revealed that the highest inter cluster distance was observed between cluster V and cluster XV (26.96) followed by cluster IV and XV (26.12), V and XII (24.55) indicating more diverse genetic makeup of the inbred lines included in the respective pairs of clusters. The genotypes in cluster V and XV were more diverse for days to maturity and genotypes in cluster IV for days to anthesis and days to silking whereas, genotypes in cluster V and XII were more diverse for 100 kernel weight. Therefore, parents of these clusters

Table 4. Average intra-cluster (diagonal bold) and inter- cluster distances (d values above diagonal) and D^2 values (below diagonal) among newly developed winter maize inbred lines

Clusters	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV
I	8.41	11.40	11.74	13.23	14.87	14.39	9.85	15.13	10.10	11.73	10.14	19.05	13.89	18.22	20.95
II	129.96	8.71	16.60	15.09	17.57	17.02	12.00	16.33	11.23	15.03	13.36	13.41	17.62	12.53	15.36
III	137.83	275.56	9.66	18.00	17.29	13.19	13.03	16.10	14.30	12.12	12.13	25.75	14.67	23.21	25.33
IV	175.03	227.71	324.00	11.59	15.74	22.87	16.47	23.28	16.40	14.61	16.06	20.97	20.98	19.38	26.12
V	221.12	308.70	298.94	247.75	10.25	23.00	15.98	23.75	19.30	18.17	15.51	24.55	19.62	21.51	26.96
VI	207.07	289.68	173.98	523.04	529.00	0.00	13.10	5.53	10.18	16.85	13.51	22.79	10.28	24.52	20.12
VII	97.02	144.00	169.78	271.26	255.36	171.61	0.00	12.70	9.92	17.33	12.76	18.31	10.94	19.81	18.80
VIII	228.92	266.67	259.21	541.96	564.06	30.58	161.29	0.00	8.56	19.36	13.93	19.63	9.91	23.78	16.84
IX	102.01	126.11	204.49	268.96	372.49	103.63	98.41	73.27	0.00	14.47	10.63	15.25	11.16	18.96	16.63
X	137.59	225.90	146.89	213.45	330.15	283.92	300.33	374.81	209.38	0.00	12.57	24.08	20.12	19.82	25.63
XI	102.82	178.49	147.14	257.92	240.56	182.52	162.82	194.04	113.00	158.00	0.00	19.92	12.70	19.54	20.31
XII	362.90	179.83	663.06	439.74	602.70	519.38	335.26	385.34	232.56	579.85	396.81	0.00	22.30	14.26	10.91
XIII	192.93	310.46	215.21	440.16	384.94	105.68	119.68	98.21	124.55	404.81	161.29	497.29	0.00	25.35	21.78
XIV	331.97	157.00	538.70	375.58	462.68	601.23	392.44	565.49	359.48	392.83	381.81	203.35	642.62	0.00	15.15
XV	438.90	235.93	641.61	682.25	726.84	404.81	353.44	283.59	276.56	656.90	412.50	119.03	474.37	229.52	0.00

can be chosen for hybridization programme. Cluster IV has the highest intra-cluster distance (11.59). The inbred lines belonging to the clusters separated by high statistical distance could be used in hybridization programme for obtaining a wide spectrum of variation among the segregates and to obtain high heterosis. The distance between the clusters VI and VIII was lowest (5.53) followed by the distance between the clusters I and VII (9.85) indicating that the inbred lines belonging to these clusters were comparatively less diverse. This is probably because genotypes in these clusters may possess common alleles governing the characters.

The genetic differences between clusters were reflected in their cluster means. Cluster mean values for 13 morphological and yield related characters are presented in Table 5. The cluster means of inbred lines revealed that the highest mean value for days to anthesis and days to silking were reported in cluster IV. Required minimum days to anthesis, silking and high grain yield were observed in cluster VIII indicating the early inbreds with high yield in this group. Zaman and Alam (2013) also reported genotypes ranking first in a cluster for early days to maturity, days to 50% silking, and tasseling, The solitary cluster XI revealed the highest cluster mean for plant height, ear height and ASI and low mean for grain yield which indicated the usefulness of inbred line for fodder yield instead of grain yield, whereas lowest mean for ASI was reported by cluster XIV which is desirable trait for high seed setting to enhance the grain yield and such material may be tested under drought and water logging stress. The cluster XIII reported the highest mean for ear girth, kernel rows per ear and kernels per row which are desirable trait for grain yield. Similarly, cluster IX reported highest mean for ear length and grain yield. Shazia *et al.* (2017) also reported genotypes with highest grain yield in one cluster while grouping of 47 maize genotypes. Highest cluster mean for 100 kernel weight was revealed by the cluster III whereas, solitary cluster VII reported highest mean for grain filling duration. The lowest maturity duration was reported by solitary cluster VI which also reported third rank for mean grain yield among the clusters. The results of the study showed that desirable genotypes may be selected from the respective cluster for utilization in breeding programme and improvement of the specific trait. Similar results have also been reported by Shrestha (2016) Bhusal *et al.* (2016), Shazia *et al.* (2017) *and* Suryanarayana *et al.* (2017). The clusters contributing maximum to D^2 values are to be given greater emphasis for deciding the clusters for the purpose of further selection and hybridization. The cluster means and characters contributing towards genetic divergence (Table 5) showed that maximum genetic divergence in percent was contributed by 100 kernel weight (39.45) followed by days to anthesis (22.64), grain filling duration (10.31) and grain yield (10.50). In a similar study Suryanarayana *et al.* (2017) also reported that grain yield was one of the major component traits for contributing towards genetic divergence. The least and negligible contribution towards divergence was observed by ear girth (0.55%).

Principal Component Analysis (PCA) used to visualize genetic distance and relatedness between

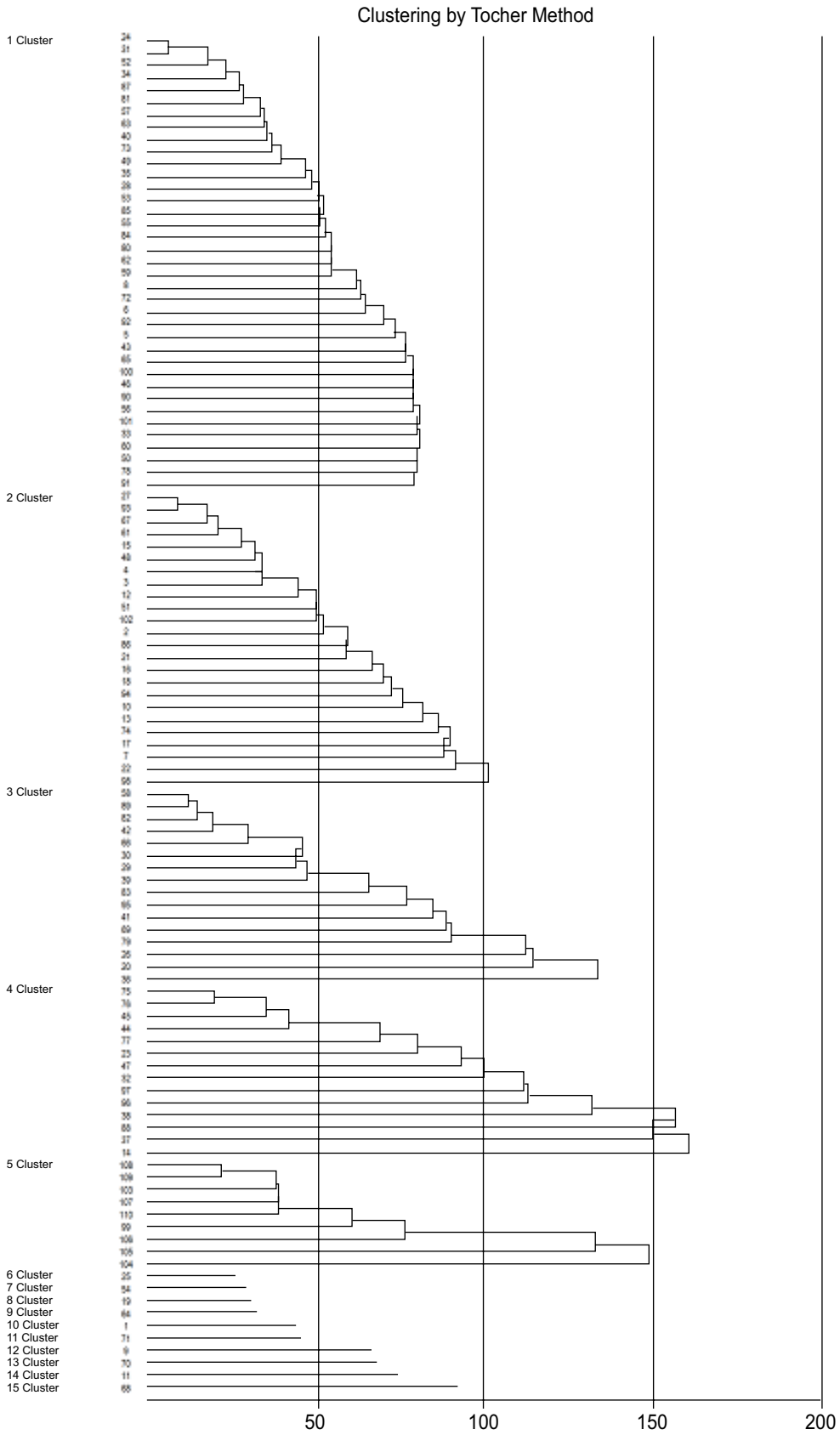

Fig. 1. Clustering of winter maize inbred lines by Tocher's method

Table 5. Cluster means and per cent contribution of characters towards divergence in newly developed winter maize inbred lines

Character	Clusters															Per cent contribution
	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV	
Days to anthesis	105.72	102.83	105.96	112.98	110.00	96.33	99.00	93.33	100.67	112.67	105.67	97.00	97.33	104.33	90.33	22.64
Days to silking	109.67	106.76	109.40	116.74	115.52	99.00	102.67	97.67	104.33	115.33	111.33	101.00	102.33	106.33	94.67	2.3
Anthesis-silking interval	3.95	3.93	3.44	3.76	5.52	2.67	3.67	4.33	3.67	2.67	5.67	4.00	5.00	2.00	4.33	2.1
Days to maturity	151.25	147.46	150.19	154.86	161.41	139.67	156.00	140.00	142.00	141.00	151.33	146.00	153.33	146.33	140.67	4.27
Grain filling duration	41.59	40.88	40.79	38.33	51.41	40.67	53.33	42.33	37.67	25.67	40.00	45.00	51.00	40.00	46.00	10.31
Plant height (cm)	106.19	80.18	100.56	112.37	118.67	111.33	82.00	125.00	123.23	110.57	151.10	100.23	141.67	57.77	78.33	5.2
Ear height (cm)	38.16	29.92	36.54	38.63	47.41	44.90	26.43	44.00	42.90	37.77	70.57	41.67	57.23	20.77	32.00	0.67
Ear length (cm)	9.94	8.65	9.25	10.48	10.22	9.27	12.23	11.00	13.20	6.30	9.93	10.00	12.77	7.23	8.67	0.58
Ear girth (cm)	12.24	11.41	12.43	12.25	12.07	12.73	13.93	12.67	12.40	10.30	11.40	10.40	15.00	8.23	10.50	0.55
Kernel rows per ear	12.08	11.93	11.97	12.66	11.04	13.07	11.87	12.67	12.33	11.07	11.53	14.00	14.00	8.90	10.67	0.27
Kernels per rows	17.00	14.58	15.20	18.55	13.48	21.27	18.50	22.33	22.87	10.77	10.97	18.00	31.43	12.33	13.33	1.53
100 kernel weight (gm)	27.45	22.46	33.77	23.08	26.56	33.47	28.00	30.33	26.33	29.43	28.13	14.73	31.40	17.53	18.73	39.45
Grain yield (q/h)	28.01	23.88	26.67	29.67	20.71	30.09	31.40	30.33	31.51	25.87	19.72	24.85	28.81	9.83	15.40	10.13

populations. The Principal component analysis can be used to uncover similarities between variables (Venujayakanth *et al*, 2017). In the present study principle component (PC) analysis partitioned the total variance (Table 6) into 5 PCs contributing maximum to the total diversity among the genotypes due to various traits. In PC analysis, first 3 PCs were found to contribute 75.89% of the total variation. The traits contributing most heavily to variation in PCI were 100 kernel weight (-0.976), plant height (-133) and ear girth (-0.121) with negative loading similarly traits, days to 50 per cent anthesis (0.830), days to maturity (0.421) and grain filling duration (0.296) with positive loading and ear height (-0.144) with negative loading contributed significantly towards total variation in PCII. In PCIII, grain yield (-0.649), plant height (-0.340), days to maturity (-0.320), ear length (-0.298), ear girth (-0.269) with negative loadings and days to anthesis (0.276) with positive loading were causing variability among genotypes. Therefore, the results showed that traits like days to anthesis, days to maturity, 100 kernel weight, grain filling duration ear length, grain yield as whole contributed most strongly towards variability among genotypes, indicating that more emphasis should pay

Table 6. Principal component analysis for morphological traits in winter maize inbreds

Characters	PCI	PCII	PCIII	PCIV	PCV
Days to anthesis	0.014	0.830	0.276	0.378	0.201
Days to silking	0.014	0.060	-0.097	-0.135	0.014
Anthesis-silking interval	0.000	0.000	0.000	0.000	0.000
Days to maturity	-0.058	0.421	-0.320	-0.346	-0.161
Grain filling duration	-0.026	0.296	-0.187	-0.630	-0.095
Plant height (cm)	-0.133	0.016	-0.340	0.049	0.770
Ear height (cm)	-0.005	-0.144	-0.077	-0.051	0.348
Ear length (cm)	-0.028	-0.059	-0.298	0.000	0.114
Ear girth (cm)	-0.121	-0.007	-0.269	-0.068	0.157
Kernel rows per ear	0.063	-0.091	-0.020	0.192	0.062
Kernels per rows	-0.026	0.069	-0.208	0.046	0.026
100 kernel weight (gm)	-0.976	-0.032	0.171	0.011	-0.079
Grain yield (q/h)	-0.078	0.062	-0.649	0.524	-0.403
Canonical roots	3510.198	3112.714	1277.573	1040.338	543.505
Percent of Variance	33.718	29.900	12.272	9.993	5.221
Cumulative proportion of Variance	33.718	63.617	75.889	85.882	91.103

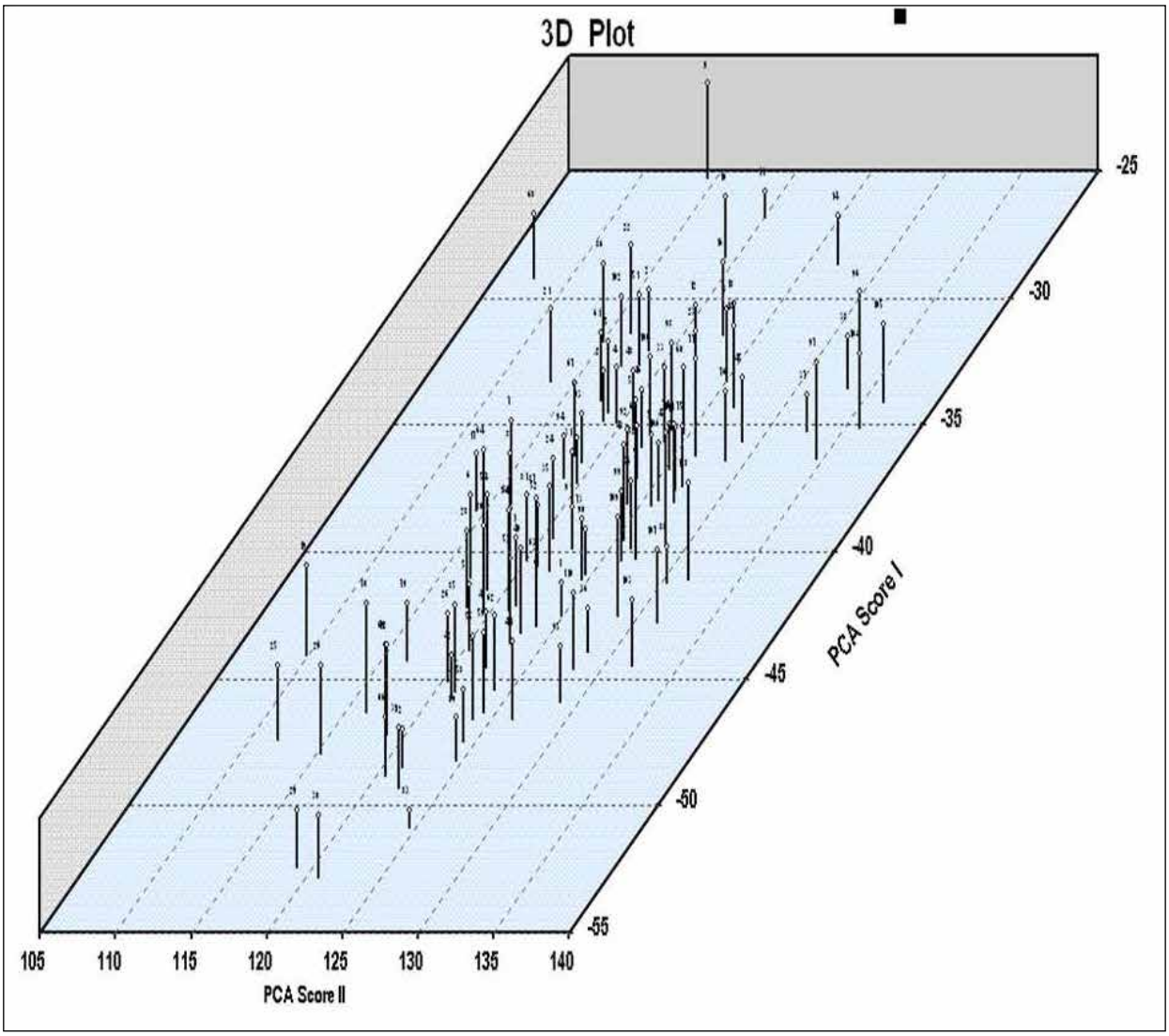

Fig. 2. 3D plot diagram showing Principle Component Analysis score of newly developed winter maize inbred lines.

on these traits while selecting the inbreds lines for their further use in breeding programme. The 3D bi-plot representation of PCI score and PCII score showed the genotypes falling in same cluster were present closer to each other in diagrams. The inbred lines in solitary clusters were present distantly from the other genotypes which have desirable mean value for traits i.e. IMLSB-3-1 (least grain filling duration), IMLSB-66.1(low plant height), IMLSB-182-1 (high plant height a desirable trait for forage), similarly lines with high intra cluster mean and with desirable mean value i.e. IMLSB-49-2 (high grain yield), IMLSB-93-2 (least grain filling duration). IMLSB-183-1(higher grain yield and 100 kernel weight) and IMLSB-975-2 (higher grain yield and 100 kernel weight indicating their usefulness in breeding programmes. Hafiz *et al.* (2015), Shazia *et al.* (2017) and Suryanarayana *et al.* (2017) also reported similar results and highlighted the usefulness of PCA in selection of maize genotypes *via* choosing of traits that strongly contributed in genetic variation of maize germplasm.

Conclusion

Selection of contrasting inbred lines for hybridization as reflected from D^2 statistics and principal component analysis (PCA) would ensure greater chances of obtaining high heterotic hybrids. The results of the present investigation will be very handy in the selection of genetically diverse and agronomically superior inbreds for hybridization programme. The genetic divergence of

parental varieties defines the manifestation of heterosis, and the heterotic pattern is determined by the genetic divergence of 2 parental lines. Therefore, crossing schemes comprising the more distant maize genotypes might allow for greater success in the production of genetic variability and thus might maximize the exploitation of heterosis and segregation (Molin *et al*, 2013). On the basis of inter cluster distances and *per se* performance, inbred line IMLSB-2005, IMLSB-1000-2, IMLSB-182-1, IMLSB-719-1, listed in solitary cluster XIII, IX, VI and Cluster VII could be used for developing hybrids. Similarly, on the basis of *per se* performance and intra cluster distance inbred line of cluster IV *viz*. IMLSB-164-1, IMLSB-457-2, IMLSB-2083, IMLSB-1298-2, IMLSB-1298-5 and IMLSB-246-2 may be used for developing new material. Inter-crossing of divergent groups would lead to greater opportunity for crossing over, which may release hidden variability by breaking linkage. Progenies derived from such diverse crosses are expected to show wide spectrum of genetic variability. Hence, hybridization of inbred lines from cluster IV with Cluster III, IX and XIII might be used in single as well as multiple crossing programmes for development of better hybrids and populations.

References

Anonymous (2014) *India maize summit Report*, pp 1-26.

Anonymous (2016) *Director's Review*, Indian Institute of Maize Research, New Delhi, pp 1-2.

Bhusal TN, GM Lal, S Marker and GJ Synrem (2016) Discrimination of maize (*Zea mays* L.) inbreds for morpho-physiological and yield traits by D^2 statistics and principal component analysis (PCA). *Asian J. Bio. Sci.* **11**(1): 77-84.

Dhliwayo T, Pixley K, Menkir A and Warburton M (2009) Combining ability, genetic distances, and heterosis among elite CIMMYT and IITA tropical maize inbred lines. *Crop science* **49**: 1201-1210.

Hafiz SBM, J Farooq, T Bibi and T Mahmood (2015) Cluster and principle component analyses of maize accessions under normal and water stress conditions. *J. Agric. Sci.* **60**(1): 33-48.

Krishnamoo N and P Mohan (2017) Cultivation and new production strategies of maize (*Zea mays* L.) in India. *Paripex International Journal of Research*. **6**(1): 265-266.

Mahalonobis PC (1936) On the generalized distance in statistics. *Proc. Nat. Acad. Sci. India* **12**: 49-55.

Molin D, C Coelho, D Máximo, F Ferreira, J Gardingo and R Matiello (2013) Genetic diversity in the germplasm of tropical maize landraces determined using molecular markers. *Genet. Mol. Res.* **12**: 99-114.

Marsan PA, P Castiglioni, F Fusari, M Kuiper and M Motto (1998) Genetic diversity and its relationship to hybrid performance in maize as revealed by RFLP and AFLP markers. *Theor Appl Genet* **96**: 219-227.

Pearson K (1901) On lines planes of closest fit to systems of points in space. *Philosophical Magazine* **2**: 559–572.

Prasanna BM (2012) Diversity in global maize germplasm: characterization and utilization. *J. Biosci.* **37**: 843-855.

Ranawat A, SK Singh, R Singh, PK Bhati and A Sharma (2013) Morphological diversity analysis in QPM and non-QPM maize (*Zea mays* L.) genotypes. *Journal of Crop and Weed* **9(2):** 32-35.

Rao CR (1952) Advanced biometrical methods in biometric research. *John Wiley and Sons* Inc., New York, pp 357-363.

Semagn K, C Magorokosho, BS Vivek, D Makumbi, Y Beyene, S Mugo, BM Prasanna and ML Warburton (2012) Molecular characterization of diverse CIMMYT maize inbred lines from eastern and southern Africa using single nucleotide polymorphic markers. *BMC Genomics* **13**: 1-11.

Senior M, J Murphy, M Goodman and C Stuber (1998) Utility of SSRs for determining genetic similarities and relationships in maize using an agarose gel system. *Crop Sci* **38**: 1088-1098.

Shazia G, ZA Dar, AA Lone, MA Khan, MA Bhat, G Ali, N Yousuf and RA Lone (2017) Genetic diversity in maize (*Zea mays* L.) inbred lines from Kashmir. *Int. J. Pure App. Biosci.* **5**(1): 229-235.

Shrestha J (2016) Cluster analysis of maize inbred lines. *Journal of Nepal Agricultural Research Council* **2**: 33-36.

Singh RK and BD Chaudhary (1985) Biometrical methods in quantitative genetic analysis. *Kalyani Publishers,* New Delhi. pp 229-252.

Suryanarayana L, MR Sekhar, DR Babu, AV Ramana and V Srinivasa Rao (2017) Cluster and principal component analysis in maize. *Int. J. Curr. Microbiol. App. Sci*. **6**(7): 354-359.

Venujayakanth B, AS Dudhat, B Swaminathan and ML Anurag (2017) Assessing crop genetic diversity using principle component analysis: a review, *Trends in Biosciences* **10**(2): 523-528.

Wende A, H Shimelis, J Derera, W Mosisa, J Danson and MD Laing (2013) Genetic interrelationships among medium to late maturing tropical maize inbred lines using selected SSR markers. *Euphytica* **191**: 269-277.

Xia XC, JC Reif, AE Melchinger, M Frisch, DA Hoisington, D Beck, K Pixley and ML Warburton (2005) Genetic diversity among CIMMYT maize inbred lines investigated with SSR markers. *Crop Sci*. **45**: 2573-2582.

Yang X, S Gao, S Xu, Z Zhang, BM Prasanna, L Li, J Li and J Yan (2011) Characterization of a global germplasm collection and its potential utilization for analysis of complex quantitative traits in maize. *Mol Breeding* **28**: 511-526

Zaman MA and MA Alam (2013) Genetic diversity in exotic maize (*Zea mays* L.) hybrids. *Bangladesh J. Agril. Res*. **38**(2): 335-343.