Genetic Divergence for Morpho-physiological Traits and Yield Components Associated with Drought Tolerance in Maize (*Zea mays* L.) Inbreds

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Forty-one maize inbreds were evaluated for obtaining information regarding genetic divergence through multivariate analysis using Mahalanobis' D^2 statistic for a set of 20 divergent characters. The analysis of variance revealed significant differences among the genotypes for all the traits except canopy air temperature difference. The D^2 analysis revealed that genotypes exhibited considerable diversity and were grouped into nine clusters, where cluster I and V accommodated maximum and equal numbers of entries (11 each). The inter-cluster distance was highest between cluster III and cluster VIII followed by cluster III and VIII, II and VII, The maximum contribution towards genetic divergence was observed by 100 kernel weight followed by dry stover yield, relative water content, proline content and grain yield. Based on Tocher's clustering pattern of the genotypes, eight inbreds were suggested for inclusion into hybridization for obtaining high heterotic crosses.

Key Words: D^2 Statistic, Drought tolerance, Genetic Divergence, Maize, *Zea mays*

Introduction

Maize is one of the most important cereal crop in the world; it finds third important position in India after rice and wheat with enormous role in food and nutritional security. Drought is one of the major causes of yield losses in maize in the world. The risk of drought is particularly high in rainfed maize cultivation. Drought that coincides with the flowering period can cause serious yield instability at the farm level, because it allows no opportunity for farmers to replant or otherwise compensate for loss of yield. Even a single irrigation omission during one of the sensitive growth stages, may cause a 30-40% grain yield loss (Cakir, 2004. In order to develop genotypes with desirable traits, the breeders choose genetically distant parents, as genetic diversity plays an important role in plant breeding because hybrids derived from the lines of diverse origin display a greater heterosis than those between closely related strains. Estimation of genetic divergence also allows breeders to eliminate some parents in downsizing the core collections maintained and concentrate their efforts in a smaller number of hybrid combinations (Fuzzato *et al*., 2002). Several methods have been developed to study the extent of genetic divergence in the genotypes among which Mahalanobis' generalized distance (D^2) (Mahalanobis, 1936) is frequently used. D^2 analysis is a useful tool in quantifying the degree of divergence between biological populations at genotypic level and to assess relative contribution of different components to the total divergence both the inter- and intra-cluster levels. In order to utilize the material for further improvement, it is imperative to know the extent of diversity present among the inbreds. Keeping this in view, the present investigation was carried out to know the magnitude of diversity present in these introduced and locally developed inbreds for morpho-physiological and yield components related to drought tolerance and to select diverse parents with an aim to obtain heterotic crosses and wide array of combinations.

Material and Methods

The material for the present investigation consisted of 41 maize inbreds obtained from DMR, New Delhi; PAU, Ludhiana; CIMMYT, Mexico and SKUAST-Jammu (Table 1). The experiment was conducted under three environments (irrigated, limited irrigation and rainfed) during *kharif* 2005 at Research Farm of Division of Plant Breeding and Genetics, SKUAST-J, Chatha, Jammu. Each genotype was sown in a two row plot of 5 m length spaced at 60 cm with interplant distance of 25 cm. The experiment was laid in Randomized Block Design with three replications. The recommended package and practices were followed to raise a good crop. Observations were recorded on twenty characters**.** In each entry five plants were randomly selected and utilized to collect observations on plant height, ear height, leaves per plant, ears per plant, ear length, ear girth, kernel rows per ear, kernels per row, 100-kernel weight, grain yield per plant, dry stover yield per plant and biological yield per plant. Data for days to 50 per cent pollen shed, days to 50 per cent silk, anthesis–silking interval, days to maturity, number of barren plants per plot and canopy-air temperature

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Table 1. List of maize inbreds, pedigree and source

S.No.	Name	Pedigree	Source
1	LM5	(Tux Pool C2IC2-5-1-1-2-2-2-3-3-1 F)	PAU, Ludhiana
2	LM6	(MS Pool C2IC2-5-1-2-1-1-1-2-1-1 F)	PAU, Ludhiana
3	LM9	(MS Pool C2IC2-11-2-2-1-1-1-1-#-FSb-l-#-#)	PAU, Ludhiana
4	LM10	(Pop28TSR (S2)-13-#-#-6F-#-1-3-1-#-I-#-#-#)	PAU, Ludhiana
5	LM11	Sw 1-26-1-1-1-1-1-1-#-#	PAU, Ludhiana
6	LM12	J54.M.17 #-21-2-3-2-2-1-1⊗b	PAU, Ludhiana
7	CM105	Peru 330-#-#329c-1-f-#	DMR, New Delhi
8	CM111	Cuba 342-2-f-#-#	DMR, New Delhi
9	CM115	Eto 182-1-2-f-f-#-A bulk-#	DMR, New Delhi
10	CM116	Puerto Rico Gr-I-A 116-f-#	DMR, New Delhi
11	CM117	Cuba 11J-A46-f-#	DMR, New Delhi
12	CM121	A derivative from A dec-A257	DMR, New Delhi
13	CM122	(J 61 7-61-1-1-1-1-1 #)	DMR, New Delhi
14	CM125	(H 104-15-1-1-3g-1-1#)	DMR, New Delhi
15	CM132	B57E#-1-3-#-3-#-⊗-#-f-##	DMR, New Delhi
16	CM135	IPA-3-6-10-3-#-⊗-⊗-#-⊗	DMR, New Delhi
17	CM137	IPA-3-7-f-⊗-#-⊗-⊗ derived from DMR	DMR, New Delhi
18	CM138	IPA-21-10-f-#-⊗ derivative of AD609	DMR, New Delhi
19	CM139	{(Tarun x MSiY-63-1g-2-1-1-2##-1-1-1#FS2###}	PAU, Ludhiana
20	CM140	(J617-61-1-1-1-f-FS5-1-1###)	PAU, Ludhiana
21	CM142	IPA-34-62-f-1-1-1-1	DMR, New Delhi
22	CM143	(JS2 x J3022) H.S.43-2-1-1-2-#FS.2-#-#-1-#-b-#	PAU, Ludhiana
23	CM144	SE533: (Tarun x MSI)-Y63-ig-2-1-1-#-#-3-1-2-1-1-F$_1$s bulk-#-#-#	PAU, Ludhiana
24	CM207	Fla3H94-f-#-#2-1-1-b	DMR, New Delhi
25	CM 213	Fla6H13-f-#-#-1-1-1-1	DMR, New Delhi
26	Y-500	Pool-500# 2-2-1-1-1-1-1⊗b	SKUAST-JAMMU
27	Y-501	Pool-501# 5-3-2-1-1-1-1⊗b	SKUAST-JAMMU
28	CML228	EV88SUWAN1-SR(BC5)-201-1-1-b-b	CIMMYT, Mexico
29	CL02443	Pob24STEC1HC45-1-2-3-3-2-BB-f	CIMMYT, Mexico
30	Ib6105	Pob45-1-2-1-1-1-1-1⊗b	SKUAST-Jammu
31	Ib6118	C6-5-1-2-1-1-1-1⊗b	SKUAST-Jammu
32	Ib6135	Pob45-5-2-1-1-1-1-1⊗b	SKUAST-Jammu
33	Ib6140	Pob33-3-2-1-1-1-1-1⊗b	SKUAST-Jammu
34	Ib6144	Pob33-7-2-1-1-1-1-1⊗b	SKUAST-Jammu
35	Ib3010	Super1-1-2-1-1-1-1-1⊗b	SKUAST-Jammu
36	PMS-14	Super1-14-2-1-1-1-1-1⊗b	SKUAST-Jammu
37	M4-4	MRTxSuper1#-4-4-1-1-1-1-1⊗b	SKUAST-Jammu
38	M7-7	MRTxSuper1#7-7-1-1-1-1-1⊗b	SKUAST-Jammu
39	M8-3	MRTxSuper1#8-3-1-1-1-1-1⊗b	SKUAST-Jammu
40	M13-8	MRTxSuper1#13-8-1-1-1-1-1⊗b	SKUAST-Jammu
41	M16-5	MRTxSuper1#16-10-1-1-1-1-1⊗b	SKUAST-Jammu

difference were recorded on plot basis. Free proline content from leaf samples was estimated following the method of Bates *et al.* (1973). Relative water content was estimated using formula: RWC (%) = $[(F_w - D_w)/(T_w - D_w)] \times 100$. Where, F_w, D_w, and T_w stands for fresh weight, dry weight and turgid weight, respectively.

Pooled analysis of variance (ANOVA) was used to quantify the genetic differences among the genotypes. The multivariate analysis was performed through Window State Software using Mahalanobis' D^2 statistics (Mahalanobis, 1936). Treating D^2 as a generalized statistical distance, the criteria used by Toucher (Rao, 1952) was applied for determining the group constellation and clustering was done accordingly (Fig.1). The character-wise rank totals were used to calculate the per cent contribution of each character to the total divergence. Average inter- and intra-

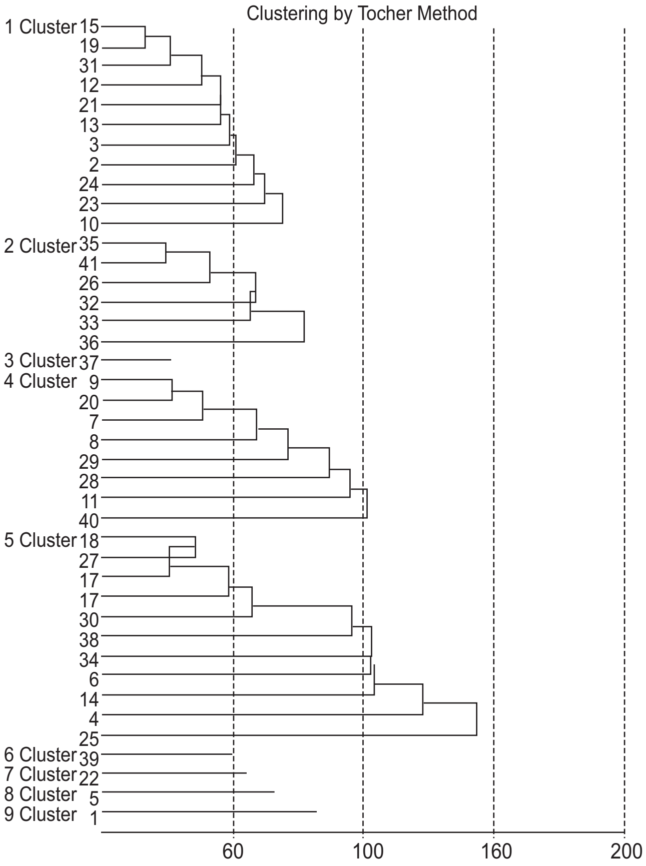

Fig. 1: Different clusters showing the position of inbreds (mentioned by serial number)

cluster distances were estimated as per the method given by Singh and Chaudhary (1985).

Results and Discussion

The analysis of variance revealed significant differences among genotypes for all the traits except canopy-air temperature difference (Table 2). Genotypes exhibited a wide range of variation for most of the traits except canopy-air temperature difference making possible the identification of lines with special characters. Similar results were also reported by Teixeira *et al.* (2002). The estimates of phenotypic coefficient of variation were greater than the genotypic coefficient of variation, indicating thereby the importance of environmental factors. High genotypic coefficient of variation (GCV) was found for number of barren plants, proline content, dry stover yield grain yield per plant, anthesis-silking interval (ASI), and ear height; moderate value of GCV were recorded for plant height, kernel rows per ear, 100-kernel weight and ear length.

The 41 genotypes were grouped into nine clusters (Fig. 1) using Tocher's method with variable number of entries (Table 3), suggesting adequate scope for selecting superior and diverse parents to be exploited for any breeding programme. Cluster I and V with 11 entries had the maximum number of genotypes followed by

Table 2. Estimates of mean, range, mean square, phenotypic and genotypic coefficient of variation, heritability and genetic correlation with grain yield for different characters in maize

Characters	Mean	Range		Mean Square	PCV (%)	GCV (%)	h^2 (broad sense)	rg with seed yield
		Min.	Max.					
Days to 50% pollen shed	59.02	52.00	66.17	82.938**	6.74	6.21	0.848	-0.440
Days to 50% silking	62.35	55.50	72.67	101.718**	7.07	6.51	0.848	-0.434
Anthesis silking interval	3.33	2.50	6.50	3.321**	29.55	20.60	0.486	-0.214
Days to maturity	93.87	83.67	108.67	160.385**	6.31	5.33	0.715	-0.160
Plant height (cm)	120.65	75.12	172.70	2878.41**	20.92	17.55	0.704	0.648
Ear height (cm)	53.43	32.30	83.93	856.202**	27.39	21.21	0.600	0.503
Number of leaves/ plant	10.93	9.50	13.03	3.823**	10.75	6.39	0.354	0.454
Number of barren plant/ plot	1.76	0.00	7.33	16.875**	83.61	60.89	0.640	-0.640
Number of ears/plant	1.10	0.62	1.33	0.121**	20.38	10.78	0.280	0.369
Ear length (cm)	11.43	7.70	15.85	22.00**	19.46	16.20	0.694	0.414
Ear girth (cm)	12.18	8.45	14.93	15.216**	14.43	12.79	0.786	0.800
Number of kernel rows	12.83	8.67	16.13	16.833**	14.76	12.69	0.739	0.622
Kernels/row	16.70	10.95	23.67	62.756**	22.66	18.55	0.670	0.372
100-kernel weight (g)	20.01	13.00	27.20	73.320**	17.78	17.41	0.959	0.688
Dry stover yield/ plant (g)	124.24	59.98	204.82	7836.555**	30.11	28.88	0.920	0.646
Biological yield/plant (g)	166.54	93.27	260.58	12114.983**	27.83	26.81	0.928	0.790
Canopy air temp. difference (°C)	0.00	-0.11	0.18	0.032	47.12	23.38	0.234	0.052
Relative water content (%)	75.00	59.00	88.06	339.794**	10.77	9.88	0.842	0.312
Proline content (mg/g fresh weight)	0.17	0.06	0.40	0.037**	49.91	45.35	0.826	-0.320
Grain yield/ plant (g)	42.31	19.87	69.65	893.056**	30.15	28.57	0.898	--------

*, ** Significant at P = 0.05 and 0.01, respectively

Table 3 Distribution of 41 maize inbreds into different clusters

Cluster number	Number of inbreds	Name of inbred
I	11	CM139, CM144, LM9, LM6, CM132, CM121, CM142, CM122, CM207, CM116, Ib6118
II	6	Ib3010, M16-5, Y-500, Ib6135, Ib6140, PMS14
III	1	M-4-4
IV	8	CML228, CL02443, CM140, CM111, CM115, CM105, CM117 M-13-8
V	11	Y-501, Ib6105, M-7-7, Ib6144, LM12, LM10, CM138, CM137, CM135, CM125, CM213
VI	1	M-8-3
VII	1	CM143
VIII	1	LM11
IX	1	LM5

Table 4. Average intra-clusters (diagonal and bold) and inter-cluster distances (D^2 values) among nine clusters in maize

Clusters	I	II	III	IV	V	VI	VII	VIII	IX
I	7.89	16.75	17.15	9.55	11.34	16.15	9.22	10.70	12.50
II		8.70	10.16	18.08	12.76	10.63	19.26	21.57	16.72
III			0.00	17.34	14.12	7.93	20.25	22.15	15.70
IV				9.89	12.94	17.10	11.82	13.37	14.05
V					10.83	12.91	12.99	15.12	14.78
VI						0.00	19.48	19.43	17.67
VII							0.00	10.56	14.99
VIII								0.00	18.07
IX									0.00

cluster IV with eight genotypes representing different eco-geographical regions. Further, cluster II had six genotypes while rests of the clusters were solitary entry clusters. Similar results were reported by Zheng and Wang (1995), Singh *et al.* (1999) and Kumar and Singh (2002). The clustering pattern revealed that the genotype originated from different geographical regions had been grouped in cluster IV which indicated that there was no association between genetic diversity and geographical diversity. Similar results were reported by Sridhar *et al.* (2002). On the other hand, the genotypes that originated in one region had been distributed into different clusters, indicating that genotypes with same geographic origin 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.

The intra- and inter-cluster distances (D^2 values) are presented in Table 4. Cluster V recorded maximum intra cluster distance followed by cluster I and cluster II, indicating existence of considerable genetic divergence among constituent genotypes. Hence parents within the cluster can be chosen for hybridization programme. The relative distance of each cluster from other clusters (inter-cluster distances) indicated greatest divergence between cluster III and cluster VIII (D=22.15) followed by cluster III and VIII (D=21.57), II and VII (D=20.25), VI and VII (D=16.09), indicating greater diversity between genotypes belonging to respective pairs of clusters, whereas clusters III and VI were closest (D=7.93) with minimum genetic distance.

The cluster means of genotypes (Table 5) revealed considerable genetic differences between the groups. The cluster II registered the highest mean value for plant height (155.74) ear height (70.76), leaves per plant (11.85), kernels per row (18.13), dry stover yield (162.29) and biological yield per plant (218.95), whereas, the highest average cluster mean was reported for yield components like grain yield per plant in cluster VI (69.65) followed by cluster III (59.87); for 100-kernel weight in cluster III (26.65) and II (25.64); for number of kernel rows in cluster VI (16.13) and III (16.00); for kernels per row in cluster II (18.13) followed by cluster V (17.63); ears per plant in cluster VI (1.27) and for shorter plant height in cluster IX (108.90). The highest cluster means for character important to drought tolerance were observed in cluster VI for relative water content (88.06) and cluster VII for proline content (0.26). The characters whose low value is desirable for drought tolerance, the lowest mean values were obtained in cluster V (92.20) and II (92.42)

Table 5. Cluster means and percent contribution of characters towards divergence in maize

Clusters Characters	I	II	III	IV	V	VI	VII	VIII	IX	Number of times ranked 1st	Contribution %
Days to 50% pollen shed	60.65	56.00	59.83	59.67	57.24	58.17	61.00	64.67	66.17	58	7.07
Days to 50% silking	64.15	59.22	62.67	62.58	60.58	61.17	64.00	68.00	72.67	1	0.12
Anthesis silking interval	3.50	3.22	2.83	2.92	3.33	3.00	3.00	3.33	6.50	37	4.51
Days to maturity	95.26	92.42	96.00	92.73	92.20	96.33	95.00	94.17	108.67	1	0.12
Plant height (cm)	110.30	155.74	131.00	109.76	120.28	129.25	109.13	119.43	108.90	1	0.12
Ear height (cm)	49.76	70.76	56.60	49.68	51.45	47.73	53.43	49.27	48.40	1	0.14
Number of leaves/ plant	10.61	11.85	10.72	10.51	11.04	10.38	11.60	11.33	10.77	3	0.40
Number of barren plant/ plot	2.67	0.72	0.00	2.15	1.03	0.00	2.67	0.17	7.33	23	2.85
Number of ears/plant	1.12	1.16	1.00	1.03	1.15	1.27	1.13	1.08	0.62	10	1.21
Ear length (cm)	10.92	12.59	10.08	10.80	12.19	9.73	9.33	13.62	9.75	13	1.59
Ear girth (cm)	11.14	13.92	14.33	10.86	12.83	14.93	10.75	13.12	12.00	14	1.71
Number of kernel rows	12.13	13.88	16.00	11.74	13.09	16.13	14.37	14.67	10.00	6	0.70
Kernels/row	16.33	18.13	15.45	15.83	17.63	16.73	14.83	16.33	12.33	12	1.51
100-kernel weight (g)	17.60	25.64	26.65	18.29	20.61	22.80	15.98	13.00	21.43	252	30.85
Grain yield/plant (g)	34.14	56.69	59.87	33.11	47.67	69.65	31.95	37.72	30.40	67	8.17
Dry stover yield/ plant (g)	105.99	162.29	116.40	103.94	142.35	150.12	114.87	101.38	73.83	127	15.61
Biological yield/plant (g)	140.13	218.98	176.27	137.05	190.02	216.77	146.82	139.10	104.23	10	0.15
Canopy air temp. difference (^{o}C)	0.00	0.00	0.04	-0.02	0.02	-0.03	-0.06	0.04	0.09	0	0.00
Relative water content (%)	71.84	75.39	84.92	81.73	73.67	88.06	59.00	68.54	67.54	96	11.71
Proline content (mg/g fresh weight)	0.16	0.10	0.16	0.24	0.17	0.08	0.26	0.10	0.16	94	11.46

for early maturity, cluster III (2.83) for ASI and cluster III (0.00) and VI (0.00) for barren plants. 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 characters contributing towards genetic divergence show that maximum genetic divergence was observed for 100-kernel weight (30.85%) followed dry stover yield (15.61%), relative water content (11.71%), proline content (11.46%) and grain yield per plant (8.17). Hence, 100-kernel weight, dry stover yield relative water content, proline content and grain yield were considered to be important traits. The least and negligible contribution towards divergence was observed for canopy air temperature difference (0.00%). Similar results were earlier reported by Kumar and Singh (2002), Teixeira *et al*. (2002) and Singh *et al*. (2005).

The data on inter-cluster distances and *per se* performance of genotypes may be used to select genetically diverse and agronomically superior genotypes. On the basis of inter cluster distances, the eight genotypes, *viz.*, M-4-4, Ib3010, M16-5, Y-500, Ib6135, Ib6140, PMS14 and LM11 listed in cluster II, III and Cluster VIII could be used to obtain the heterotic combinations in hybridization programme. Intercrossing of divergent groups would lead to greater opportunity for crossing over, which may release hidden variability by breaking linkage (Thoday, 1960). Progenies derived from such diverse crosses are expected to show wide spectrum of genetic variability. Hence, these genotypes might be used in single as well as multiple crossing programmes for development of hybrids.

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