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## Research Article

### Assessing genetic diversity of maize genotypes for transpiration efficiency

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#### Abstract

Transpiration Efficiency (TE), the biomass produced per kilogram of water transpired is an ideal parameter for measuring genetic variation in crop water use efficiency. Assessing the genetic diversity and variability in maize for this character will help us to understand the most efficient genotypes for water use. Studies on 89 maize genotypes for two years using six characters revealed significant differences among the genotypes and wide range of variation for all the characters. High variability was observed for total biomass, seed yield and TE-seed. High heritability and genetic advance as per cent of mean was observed for all the characters except water transpired. Eighty nine genotypes were grouped into 10 clusters and total biomass contributed maximum towards genetic divergence. TE seed was positively and significantly correlated with all the characters studied except water transpired. As there is a lot of variability present in the maize genotypes and the desirable characters reported high coefficients of variability, heritability, genetic advance and positively high correlation among the characters, the chance of selecting superior genotypes are more. Moreover, crossing between different clusters of clusters will enhance the chance of transgressive segregants. Maize genotypes Z 32-87, NSJ-176, DTL-2, Z101-68, HKI-1040-4, NSJ-189, LM-16, HKI-1025, NSJ-2011-26, NSJ-2011-37, DTL-3, DTL-4-1 HKI-1332, Z-60-72 and PSRJ-13038 were found to be superior for TE biomass and TE seed.

#### Key words

Transpiration efficiency, Maize, Genetic variation

#### INTRODUCTION

The ability of crop to produce high yield per unit of available water is potentially important in affecting profitability and yield, in both irrigated and rainfed production system. Genetically, increasing crop water use efficiency is an effective strategy for increasing yield in dry environment (Condon *et al.*, 2004; Blum 2009) as forecasts of increasing scarcity of water for agriculture remains a strong motivation for improving crop water use efficiency. Transpiration Efficiency (TE) defined as the biomass production per unit of water transpired is the preferred measure for examining potential genetic variation in crop water use efficiency (WUE). Transpiration efficiency is WUE of plants alone (no soil water losses).

Almost a century ago, Briggs and Shantz (1913) showed that crop species differ in their transpiration efficiency.

Since then, the  $C_3$  and  $C_4$  photosynthetic pathways have been elucidated, and differences in transpiration efficiency have been related to them. Subsequent studies have identified genetic variation in transpiration efficiency within a species, examples include groundnut (Ratnakumar *et al.*, 2009; Halilou *et al.*, 2015), cowpea (Ismael and Hall 1992; Halilou *et al.*, 2015), wheat (Farquhar and Richards 1984; Blum 2009) and sorghum (Hammer *et al.*, 1997; Xin *et al.*, 2009) and breeding efforts have been made to include TE in the improved germplasm (Udayakumar *et al.*, 1998). Jackson *et al.*, (2015) reported a significant variation in whole plant transpiration efficiency among 51 genotypes of sugarcane (*Saccharum spp.*) or closely related germplasm in pot culture experiment. Phenotypic values for whole plant TE ranged from 5.7 to 8.6 g/Lin sugarcane. So, it is important to assess the genetic

variation for TE over an entire crop cycle and to determine whether there is a large genotype by water regime interaction for TE in maize.

Maize is a diverse and highly cross pollinated crop. There exist a good number of works on genetic diversity of maize, The genotypes experimented in this study were not studied for TE and water requirement so far. Analysis of genetic diversity is an important step for better understanding and utilization of germplasm. The present investigation was undertaken with a view to quantify the genetic variation for water use and transpiration efficiency in 87 maize genotypes and two varieties, to estimate genetic variance, heritability and genetic advance and to investigate the nature and magnitude of genetic diversity in 89 maize genotypes.

## MATERIALS AND METHODS

Eighty seven maize genotypes of diverse origin were used for the study. Seeds of these maize genotypes were obtained from National Bureau of Plant Genetic Resources (NBPGR), Regional Station, Hyderabad, Directorate of Maize Research (DMR), New Delhi, CIMMYT, Regional Centre, Hyderabad, Central Research Institute for Dry Land Agriculture (CRIDA), Hyderabad and Maize Research Station, Hyderabad (**Table 1**). Two varieties (Harsha and Varun) from Maize Research Centre, PJTSAU, Hyderabad were also used in the experiment. The experiment was laid out in Randomized Complete Block Design with three replications (RCBD). The plants were grown in pots during *Kharif -2014 and 2015* (June–Oct.) under rain out shelter facility. The minimum temperature was 19° C and the maximum was 36.6° C and relative humidity and sun shine hours during crop growth period varied from 44.0 % to 96.0 % and 0.0 to 10.3 hrs respectively.

Growth observations and water balance in pots quantified components of whole-plant TE. Transpiration efficiency was determined by a high throughput gravimetric method (Xin *et al.*, 2008) with modification. Whole plant level TE was determined gravimetrically in 18-liter plastic pot filled with a mixture of red soil and farmyard manure (21 kg). Recommended dose of fertilizer and standard agronomic practices were adopted. Two seeds were planted per pot and thinned to one plant at 7 d after emergence. The pots were then covered from both ends with poly bags. A slit was cut in the top bag to permit seedling growth. The slit was further sealed with a piece of clear adhesive tape. The poly bags were tightly fixed onto the pots with an elastic band (Photo-1). Dry soil was placed on top of the poly bag around the plant to avoid heating of poly bag. The initial weight was recorded. The pots were weighed every 5 days (from 7 days after covering with poly bags) and measured quantity of water was supplemented through a funnel placed into the poly bag and again sealed with tape after watering. Duration of maize genotypes used in the experiment were 110-120 days.

When the plants reached maturity, they were harvested at soil level and final pot weight was recorded. Individually plants were partitioned into leaves, stem and cobs. Dry weights were recorded after keeping the plant parts in hot air oven at 60° C till the constant weight were attained. Cobs were sun dried. Seed was separated from cob and seed yield was recorded. Total water transpired was calculated by subtracting the final pot weight from the initial weight and then adding the amount of water that has been applied at regular interval. TE biomass was calculated by dividing the above ground dry biomass by the amount of water transpired. TE seed was calculated by dividing the seed yield by the amount of water transpired.

**Table 1. Source of genotypes**

Institute	No.	Genotype names
NBPGR	40	RJR-049, RJR-55, RJR-42, NSJ-315, RJR-163, NSJ-366, NSJ-245, RJR-159, RJR-270, RJR-198, RJR-288, RJR-208, RJR-247, PSR13187, PSR13255, PSR13247, RJR-328, RJR-363, RJR-385, PSRJ13122, PSRJ13041, PSRJ13007, PSRJ13099, PSRJ13059, NSJ-211, NSJ-176, NSJ-189, NSJ-155, PSRJ13086, PSRJ13038, PSRJ13154, RJR-068, RJR-075, RJR-115, SNJ-2011-37, SNJ-2011-26, SNJ-2011-03, SNJ-2011-104, SNJ-2011-102, SNJ-2011-70
DMR	20	HKI-161, HKI-163, HKI-164-7-4, HKI-1035-10, HKI-1011, HKI-3-4-6ER, HKI 1025, HKI 209, HKI 1332, HKI 766(0), HKI 577, HKI 1040-4, HKI 46, HKI 325-17AN, HKI 47, HKI-L287, LM5, LM13, LM14, LM16
CIMMYT	19	Z60-87, Z40-19, Z61-34, Z59-9, Z59-11, Z101-68, Z32-12, Z93-194, Z49-7, Z93-154, Z101-15, Z59-41, Z60-72, Z32-87, Z93-170, Z40-183, Z49-65, Z96-5, Z162-9
CRIDA	8	DTL-1, DTL-2, DTL-3, DTL-4, DTL-4-1, DTL-10, DTL-11, DTL-12
Maize Research Station, Hyd.	2	Harsha, Varun (Composite variety)

## RESULTS AND DISCUSSION

Analysis of variance was carried out separately for two years for the six characters studied in maize and as coefficients of variations were low pooled analysis of variance was carried out and given in **Table 2**. Pooled

analysis of variance revealed highly significant mean sum of squares due to genotypes for all the characters indicating significant differences among 89 genotypes of maize. Significant mean sum of squares due to environment indicates the presence of significant differences among

the environments for all the characters studied.

The estimates of range, mean, phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability ( $h^2$ ) and genetic advance as per cent of mean are presented in **Table 3**. Total biomass ranged from 60.45 to 159.78 with a mean of 108.24 gm whereas seed yield ranged from 18.18 to 59.15 with a mean of 37.35 gm. Harvest index ranged from 21.28 to 44.31 with a mean of 34.26%. Range of water transpired in 89 maize genotypes

was from 24.45 to 34.15 with a mean of 31.34 kg/plant. The maize genotypes transpired 8.0 to 11.3 kg/plant during pre anthesis stage and 24.5 to 34.1 kg/plant from sowing to grain harvest indicating that 32.6 to 33.1% of water is transpired during pre anthesis stage. TE biomass ranged from 2.55 to 6.24 with a mean of 4.40 g/kg. TE seed ranged from 0.67 to 2.57 with a mean of 1.56 g/kg. Genetic diversity present in the material is evidenced from the wide range of variation observed for all the characters and the probability of selecting desirable genotypes will be

**Table 2. Analysis of variance for plant traits and TE in pooled data**

Source	df	Total biomass	Seed Yield	Harvest Index	Water Transpired	TE <sub>biomass</sub>	TE <sub>seed</sub>
Replications	2	24.850	8.068	1.211	0.049	0.009	0.008
Environment	1	39720.28	674.60	1353.63	132020.80	1967.27	290.08
Interaction	1	2.051	0.418	0.345	0.162	0.005	0.001
Genotypes	88	3308.6**	807.7**	202.9**	13.9**	4.21**	1.31**
Error	176	59.447	20.171	11.896	1.726	0.299	0.111
SEm		7.1	12.02	10.1	4.2	12.4	21.4
CV (%)		5.1	1.83	1.4	0.5	0.2	0.1
CD (5%)		8.7	5.10	3.9	1.5	0.6	0.4

\*Significant at 5% level, \*\* significant at 1% level



Maize plant pot covered with poly bag



Pot with dry soil and rubber band to seal



**Photo 1. Transpiration efficiency experiment setup under rainout shelter facility**

high. It is needed to partition the observed variability into heritable and non-heritable components by the means of GCV and PCV to understand the influence of environment on the expression of different characters.

The estimates of PCV were higher than that of GCV for all the characters indicating the effect of environment but the narrow difference between GCV and PCV indicated little influence of environment on these characters. PCV ranged from 6.19 (water transpired) to 35.88 (TE seed) and GCV ranged from 4.56 (water transpired) to 30.67 (seed yield). High genotypic coefficient of variability was observed for total biomass, seed yield and TE seed indicating high variability for these characters among the genotypes and there is a great scope for the improvement of these characters by direct selection among the genotypes. Medium variability was observed for the characters harvest index and TE biomass which indicated the variation for these characters was medium among the genotypes and there is a need for improvement of base population to increase the genetic variability and to fix the favourable alleles. Vashistha *et al.*, (2013) also reported a similar result. Low variability was recorded by water transpired indicated there is a less difference among the genotypes for this character.

For assessing the heritable variation, the magnitude of heritability is the most important aspect in the breeding material which has close bearing on the response to

selection. Heritability was high for all the characters except water transpired and it ranged from 54.16 (water transpired) to 90.11 (total biomass) which indicated that these characters were relatively less influenced by environmental conditions and phenotypic selection would be effective for the improvement of these characters. The higher values of heritability of traits are indicative that the selection can be made on the basis of these traits (Ali *et al.*, 2012).

Genetic advance as per cent of mean ranged from 6.91 (water transpired) to 58.82 (seed yield) and it was high for all the characters except water transpired which indicated that these characters are governed by additive gene action and selection would be effective for the improvement of these characters. High heritability coupled with high genetic advance as per cent of mean was observed for all the characters except water transpired which indicated that most likely the high heritability might be due to additive gene effects hence; it could be improved by simple selection methods like pureline selection, mass selection, progeny selection or family selection. Rahman *et al.*, (2015), Vashistha *et al.*, (2013), Nataraj *et al.*, (2014), Haydar *et al.*, (2015), reported analogous kind of result for this trait. Most of the characters in the present study recorded high variability, heritability and genetic advance as per cent of mean indicating additive gene action for the expression of these characters and scope of simple selection for the improvement of these characters.

**Table 3. Estimates of variability, heritability and genetic advance as per cent of mean for six characters**

S. No.	Character	Range		Mean	PCV (%)	GCV (%)	h <sup>2</sup> (%)	GA as % of mean (5%)
		Minimum	Maximum					
1.	Total biomass (g/pl)	60.45	159.78	108.24	22.65	21.50	90.11	42.04
2.	Seed Yield (g/pl)	18.18	59.15	37.35	32.94	30.67	86.68	58.82
3.	Harvest Index (%)	21.28	44.31	34.26	19.30	16.47	72.79	28.95
4.	Water Transpired (kg/pl)	24.45	34.15	31.34	6.19	4.56	54.16	6.91
5.	TE <sub>biomass</sub> (g biomass/kg water)	2.55	6.24	4.40	22.15	18.34	68.53	31.27
6.	TE <sub>seed</sub> (g seed/kg water)	0.67	2.57	1.56	35.88	28.77	64.29	47.52

PCV and GCV: Phenotypic and genotypic coefficient of variation, h<sup>2</sup>: Heritability in broad sense, GA: Genetic Advance

Multivariate analysis using Mahalanobis D<sup>2</sup> statistic provides a useful tool for measuring the genetic diversity with respect to all the characters considered together. Analysis of variance for dispersion showed a highly significant mean sum of squares due to genotypes. Test of significance using Wilk's criterion revealed highly significant V statistics. The D<sup>2</sup> values were computed for all the possible pairs of combinations from the mean values of 89 maize genotypes to 6 characters. Ranking character wise D<sup>2</sup> values and adding the ranks for each character for all the genotypes had done to identify the characters that

contributed maximum towards divergence. Contribution of different characters towards genetic divergence is presented in **Table 4**. Total biomass contributed the highest (60.80%) towards genetic divergence followed by seed yield (25.20%), water transpired (7.61%), harvest index (3.78%), TE total biomass (0.84%) and TE seed (1.76%). Present results are agreement with those of Marker and Krupakar (2009). High % contribution of total biomass towards genetic diversity indicated this character can be used as parameters in selecting genetically diverse parents for hybridization.



**Table 4. Contribution of each character to divergence**

Source	Times Ranked 1st	Contribution %
1. Total Biomass (g/pl)	2381	60.80
2. Seed Yield (g/pl)	987	25.20
3. Harvest Index (%)	148	3.78
4. Water Transpired (kg/pl)	298	7.61
5. $TE_{total\ biomass}$ (g biomass/kg water)	33	0.84
6. $TE_{seed}$ (g seed/kg water)	69	1.76

Eighty nine genotypes were grouped into 10 clusters. The compositions of different clusters along with number of genotypes are given in **Table 5**. Cluster II was the largest with 24 genotypes followed by cluster III with 23 genotypes, cluster I with 16 genotypes, cluster V with 14 genotypes and cluster VIII with 7 genotypes and remaining clusters were solitary. Presence of solitary clusters indicated the extreme phenotypic performance in positive or negative directions for one or the other characters included in the present study. The genotypes from different sources were grouped together in different clusters revealed that there was no parallelism between

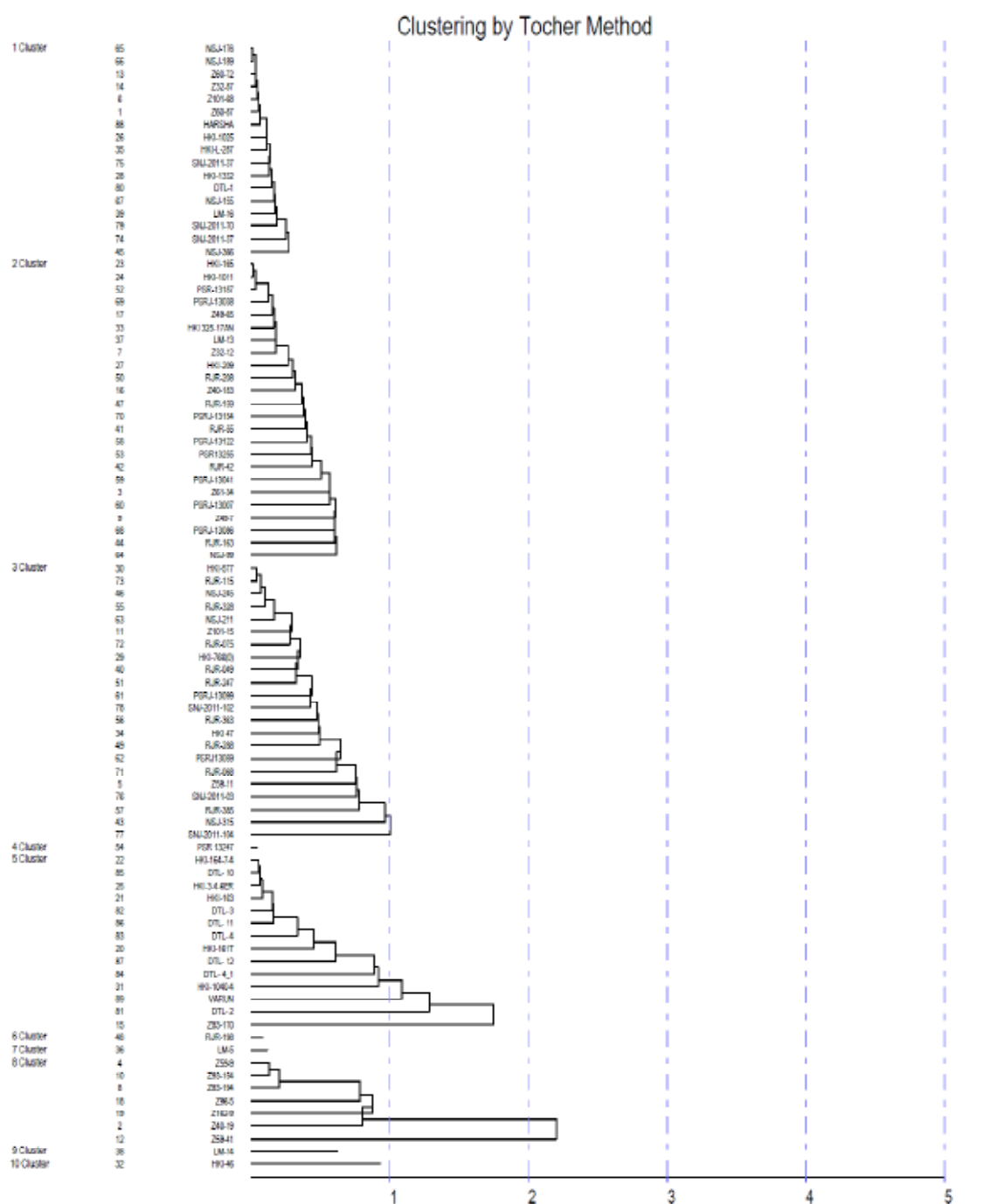
genetic diversity and geographic diversity. The nature of selection forces operating under one eco-geographical region seemed to be similar to that of other regions since the accessions from different geographical regions were grouped together into same clusters. This would be due to the similarity of objectives and conditions under which the types were bred and domesticated in different localities. By observing the cluster composition, it was evident that the accessions of same source were scattered into different clusters. The existence of wide genetic diversity among the accessions chosen from the same geographical region was thus obvious.

**Table 5. Grouping of genotypes into different clusters by Tocher method**

Cluster	No.	Genotype names
Cluster I	16	NSJ-176, NSJ-189, Z60-72, Z32-87, Z101-68, Z60-87, HARSHA, HKI-1025, HKI-L-287, SNJ-2011-26, HKI-1332, DTL-1, NSJ-155, LM-16, SNJ-2011-70, SNJ-2011-37
Cluster II	24	NSJ-366, HKI-165, HKI-1011, PSR-13187, PSRJ-13038, Z49-65, HKI 325-17AN, LM-13, Z32-12, HKI-209, RJR-208, Z40-183, RJR-159, PSRJ-13154, RJR-55, PSRJ-13122, PSR13255, RJR-42, PSRJ-13041, Z61-34, PSRJ-13007, Z49-7, PSRJ-13086, RJR-163
Cluster III	23	HKI-577, RJR-115, NSJ-245, RJR-328, NSJ-211, HKI-161, RJR-075, HKI-766(0), RJR-049, RJR-247, PSRJ-13099, SNJ-2011-102, RJR-363, HKI 47, RJR-288, PSRJ13059, RJR-068, Z59-11, SNJ-2011-03, RJR-385, NSJ-315, SNJ-2011-104
Cluster IV	1	PSR 13247
Cluster V	14	HKI-164-7-4, DTL- 10, HKI-3-4-6ER, HKI-163, DTL- 3, DTL- 11, DTL- 4, DTL- 12, DTL- 4_1, HKI-1040-4, VARUN, DTL- 2, Z93-170, Z101-15.
Cluster VI	1	RJR-198
Cluster VII	1	LM-5
Cluster VIII	7	Z59-9, Z93-154, Z93-194, Z96-5, Z162-9, Z40-19, Z59-41
Cluster IX	1	LM-14
Cluster X	1	HKI-46

**Table 6. Inter and intra Cluster Distances on the basis of D<sup>2</sup> cluster analysis**

Cluster	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI	Cluster VII	Cluster VIII	Cluster IX	Cluster X
Cluster I	<b>0.467</b>	3.443	8.981	6.372	2.779	6.699	8.974	11.820	12.447	12.862
Cluster II		<b>1.303</b>	3.647	1.761	4.177	1.825	3.212	6.392	7.635	12.066
Cluster III			<b>1.746</b>	3.643	11.064	3.472	3.516	4.968	13.996	22.488
Cluster IV				<b>0.000</b>	5.307	0.588	1.006	4.902	5.088	11.328
Cluster V					<b>2.543</b>	5.682	8.518	12.779	7.300	7.473
Cluster VI						<b>0.000</b>	1.341	4.408	5.352	11.669
Cluster VII							<b>0.000</b>	4.653	8.142	16.731
Cluster VIII								<b>3.617</b>	16.461	25.541
Cluster IX									<b>0.000</b>	2.491
Cluster X										<b>0.000</b>



**Fig.1. Grouping of genotypes by Tocher method**

Procedure suggested by Tocher (Rao, 1952) was used to group 89 genotypes into ten clusters. The pattern of distribution of 89 genotypes into various clusters is indicated in table 5

The wide divergence noticed might also be indicative of crop adaptation for wide environmental conditions under which this crop was grown. Inter and intra cluster genetic distance (D) values among five clusters are presented in **Table 6**. Inter cluster D values ranged

from 0.598 (IV and VI) to 25.54 (VIII and X). These findings are in conformity with the findings of Marker and Krupakar (2009). Medium inter cluster distance between cluster III and X and VIII and X suggested genotypes belonging to the clusters separated by the high statistical

distance could be used in recombination breeding for obtaining high heterotic responses and better segregants. Intra cluster values ranged from 0 to 3.62 (VIII). Maximum intra cluster distances were recorded by cluster VIII and V indicated these cluster had the accessions with varied genetic divergence while accessions of clusters III and V showing minimum intra cluster distance genetically resembled to each other and might have come from common gene pool. Intra cluster distances were zero in the solitary clusters. The inter-cluster

distances were larger than the intra-cluster distances which indicated wider genetic diversity among the genotypes of different groups. Debnath (1987) obtained a larger inter-cluster distance than the intra-cluster distance in a genetic variability in maize. Similar results were also obtained by Abedin and Hossain (1990) in maize. Clusters with comparatively less magnitude of divergence showed instability, while widely divergent clusters remained distinct in different environments (Raut *et al.*, 1985).

**Table 7. Cluster-wise mean table for the six traits studied in 89 genotypes of maize**

Cluster	Total biomass	Seed Yield	Harvest Index	Water Transpired	TE <sub>biomass</sub>	TE <sub>seed</sub>
Cluster I	128.763	52.497	40.956	31.879	5.081	2.130
Cluster II	108.060	36.490	33.860	31.684	4.384	1.520
Cluster III	80.662	27.452	34.289	30.807	3.414	1.204
Cluster IV	102.750	26.533	26.562	31.807	3.977	1.105
Cluster V	136.037	46.599	34.103	32.049	5.411	1.923
Cluster VI	103.683	27.250	26.275	31.287	4.583	1.200
Cluster VII	93.950	22.000	23.305	33.370	3.630	0.810
Cluster VIII	83.540	22.162	26.312	28.131	3.709	0.940
Cluster IX	130.600	27.800	21.280	33.710	5.085	1.075
Cluster X	156.300	41.300	26.430	33.470	6.000	1.575

**Table 8. Phenotypic (upper diagonal) and genotypic (lower diagonal) correlation matrix.**

Character	Total Biomass	Seed Yield	harvest Index	Water transpired	TE (biomass)	TE (seed)
Total biomass	1.000	<b>0.831**</b>	<b>0.256</b>	<b>0.449</b>	<b>0.887**</b>	<b>0.691**</b>
Seed Yield	0.859**	1.000	<b>0.733**</b>	<b>0.315</b>	<b>0.768**</b>	<b>0.902**</b>
Harvest Index	0.314	0.745**	1.000	<b>0.024</b>	<b>0.270</b>	<b>0.751**</b>
Water Transpired	0.579	0.429	0.101	1.000	<b>0.257</b>	<b>0.191</b>
TE <sub>biomass</sub>	0.989**	0.858**	0.324	0.431	1.000	<b>0.795**</b>
TE seed	0.820**	0.999**	0.797**	0.334	0.824**	<b>1.000</b>

\*Significant at 5% level, \*\* significant at 1% level

Cluster mean values were given in **Table 7** and ranged from 80.66 (III) to 156.30 g/pl (X) for total biomass. Seed yield ranged from 22 (VII) to 52.5 g/pl (I). Harvest index ranged from 21.28 (IX) to 40.96 (I). Water transpired ranged from 28.13 (VIII) to 33.71 Kg/pl (IX). TE Biomass ranged from 3.41 (III) to 6.00 g/kg (X). TE seed ranged from 0.81 (VII) to 2.13 g/kg (I). Based on cluster means Singh and Chaudhari (2001) also reported a wide range of variation for grain yield and its components in maize. Similarly, Marker and Krupakar (2009) have also assessed the range of variability of 16 genotypes for 14 different traits in maize.

Knowledge about the nature and extent of association among different biometrical characters will be useful to identify the key characters for which the selections can be fruitfully made. Genotypic and phenotypic correlation coefficients were estimated and given in Table 8. Magnitude of genotypic correlations were high

compared to corresponding phenotypic correlations for all the characters which indicated that there was a strong inherent genetic association between the various characters studied and phenotypic correlations were reduced by significant interaction of environment. TE seed is significantly positively correlated with total biomass (0.82), seed yield (0.99), harvest index (0.80) and TE biomass (0.82). TE biomass was positively and highly significantly correlated with total biomass (0.99) and seed yield (0.86). Seed yield (0.75) was positively highly significantly correlated with harvest index and total biomass (0.86) was highly significantly positively correlated with seed yield. High magnitude of genotypic correlations indicated that there was a strong inherent genetic association between the various characters studied. Correlation among component characters revealed that strong positive associations among desirable component characters, hence the selection criteria should consider all these characters. Maize genotypes Z 32-87,

NSJ-176, DTL-2, Z101-68, HKI-1040-4, NSJ-189, LM-16, HKI-1025, NSJ-2011-26, NSJ-2011-37, DTL-3, DTL 4-1 HKI-1332, Z-60-72 and PSRJ-13038 were found to be superior for TE biomass and TE seed.

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