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GENETIC DIVERGENCE AMONG CHILLI (*CAPSICUM ANNUUM* L.) GENOTYPES BASED ON QUANTITATIVE AND QUALITATIVE TRAITS

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ABSTRACT
Genetic divergence among sixty three genotypes of chilli was assessed using Mahalanobis D^2 statistic for sixteen characters at Horticultural Research Station, Lam, Guntur, Andhra Pradesh. The analysis of variance revealed significant differences among the genotypes for all the characters studied indicating considerable diversity in the material. Based on Mahalanobis D^2 statistic, the sixty three genotypes were grouped into 8 clusters. The maximum contribution towards genetic divergence was by fruit diameter (44.14%) followed by yellow carotenoids (16.90%), red carotenoids (10.45%), ascorbic acid (10.19%) and capsaicin (9.17%). The mutual relationships between the clusters revealed that inter-cluster distance values were greater than intra-cluster values. Among the clusters, clusters III and V were the largest containing 17 genotypes followed by cluster IV (11) whereas the clusters VI, VII and VIII were mono genotypic (1 genotype). The highest inter cluster distance was observed between clusters IV and VIII (4139.41) whereas the lowest was observed between clusters I and III (117.25). Cluster V (434.43) has exhibited highest intra cluster distance and the lowest was observed in clusters VI, VII and VIII (0.00). D^2 cluster analysis revealed wide genetic distance (inter cluster) between the genotypes of cluster IV (LCA-353, LCA-716, LCA-756, LCA-724, LCA-714, Pusa Sadabahar, Pant C-1, LCA-758, G-4, LCA-738 and LCA-760) and VIII (Warangal chapatta) and the crossing between genotypes of these two clusters can be exploited for the development of heterotic hybrids in future breeding programmes.

KEY WORDS: *Capsicum annuum*, chilli, D^2 statistic, clustering, genetic divergence.

INTRODUCTION

Chilli (*Capsicum annuum* L., 2n = 24) a member of the *Solanaceae* family has originated from South and Central America. It is an indispensable spice due to its pungency, taste, appealing colour and flavor and has its unique place in the diet as a vegetable cum spice crop. India is the largest producer, consumer and exporter of chilli in the world with an annual production of 1.30 million tonnes from 0.79 million ha with production share of 22.72% (Indian Horticulture Database, 2013). Andhra Pradesh leads the country in its production, productivity and export followed by Karnataka, West Bengal, Madhya Pradesh and Orissa.

Capsicinoids and carotenoids are the major chemical constituents of chilli fruits and add commercial value to the crop. The carotenoids contributing to fruit colour act as dietary precursors of vitamin A and play an important role in the regulation of vision, growth and reproduction. Among carotenoids 'capsanthin, capsorubin and capsanthin 5,6 – epoxide are responsible for the final red colour (Davies *et al*., 1970). Pungency (heat) is an important quality attribute of hot pepper besides colour. The nature of pungency has been established as a mixture of seven closely related alkyl vanillyl amides, collectively referred as "Capsaicinoids". Among capsiacinoids, capsaicin (8-methyl-N-vanillyl-6-enamide) and dihydrocapsaicin account for more than 80 and determine the pungency (Bosland and Votava, 2000). The degree of pungency varies widely with the genotypes of five cultivated species (Kumar *et al*., 2006) and range from less than 0.05% in the mildly pungent types to as high as 1.3% in the hottest chillies. The 'capsaicin' is an alkaloid present in the placenta of the fruit, which can directly scavenge various free radicals (Reddy and Lokesh, 1992; Kogure *et al*., 2002; Bhattacharya *et al.*, 2010) and has diverse prophylactic and therapeutic uses in Allopathic and Ayurvedic medicine (Sumathy and Mathew, 1984). The pharmaceutical application of capsaicinoids is attributed to its antioxidant, anticancer, antiarthritic and analgesic properties (Prasad *et al.*, 2006). Chilli is a good source of vitamin C (ascorbic acid) used in food and beverage industries (Bosland and Votava, 2000). It has also acquired a great importance because of the presence of 'oleoresin', which permits better distribution of color and flavor in foods. Apart from developing traditional varieties through conventional breeding, exploitation of heterosis for yield and yield attributing characters through hybridization is also important in crop improvement. Screening of available germplasm helps in studying the variability and diversity and identification of superior parents for use in hybridization. A wide variability in chilli fruit morphology, pungency, bearing habit and crop duration is found

throughout India (Asati and Yadav, 2004). Genetic divergence existing in the population helps in the selection of suitable parents for utilization in any crop breeding programme leading to reduction in the number of crosses (Guerra *et al.,* 1999). The information on the nature and degree of genetic divergence is essential for the breeder to choose the right type of parents for hybridization in heterosis breeding (Patel *et al*., 1989). Moreover, evaluation of genetic diversity is important to know the source of genes for a particular trait within the available germplasm (Tomooka, 1991). In order to benefit transgressive segregation, the knowledge of genetic distance between parents is necessary (Khodadabi *et al*., 2011). Hybrids produced from distantly related parents are expected to exhibit higher heterosis and minimize the inherent field genetic vulnerability (Moll *et al*., 1962; Ramanujam *et al*., 1974) than those from closely related parents. The knowledge of characters influencing divergence is important for a breeder to plan a successful breeding programme. Thus, the present study was undertaken to assess the genetic diversity in 63 genotypes of chilli (*Capsicum annuum* L.) and to identify suitable donors for a successful breeding programme in this crop. Mahalanobis's D^2 statistic of multivariate analysis is recognized as a powerful tool in quantifying the degree of genetic divergence among the populations and has been utilized in this study.

MATERIALS & METHODS

The experiment was carried out with 63 genotypes of chilli (Table 1) at Horticultural Research Station, Lam, Guntur, Andhra Pradesh, India. The site of the experiment at Lam is situated on 16.28^0 North latitude and 80.44^0 East longitude at an altitude of 31.5 m above mean sea level which falls under humid tropical climate and the soils of the experimental site are rich black cotton soils.

TABLE 1: List of chilli genotypes used in the experiment and their source

Treatment	Accession Number	Source	Treatment	Accession Number	Source
$T_{1.}$	G-3	HRS, Lam farm, Guntur	$T_{34.}$	LCA-728	HRS, Lam farm, Guntur
$T_{2.}$	G-4	HRS, Lam farm, Guntur	$T_{35.}$	LCA-730	HRS, Lam farm, Guntur
$T_{3.}$	G-5	HRS, Lam farm, Guntur	$T_{36.}$	LCA-732	HRS, Lam farm, Guntur
$T_{4.}$	LCA-206	HRS, Lam farm, Guntur	$T_{37.}$	LCA-734	HRS, Lam farm, Guntur
$T_{5.}$	LCA-235	HRS, Lam farm, Guntur	$T_{38.}$	LCA-736	HRS, Lam farm, Guntur
$T_{6.}$	LCA-305	HRS, Lam farm, Guntur	$T_{39.}$	LCA-738	HRS, Lam farm, Guntur
$T_{7.}$	LCA-315	HRS, Lam farm, Guntur	$T_{40.}$	LCA-740	HRS, Lam farm, Guntur
$T_{8.}$	LCA-353	HRS, Lam farm, Guntur	$T_{41.}$	LCA-742	HRS, Lam farm, Guntur
$T_{9.}$	LCA-357	HRS, Lam farm, Guntur	$T_{42.}$	LCA-744	HRS, Lam farm, Guntur
$T_{10.}$	LCA-424	HRS, Lam farm, Guntur	$T_{43.}$	LCA-746	HRS, Lam farm, Guntur
$T_{11.}$	LCA-436	HRS, Lam farm, Guntur	$T_{44.}$	LCA-748	HRS, Lam farm, Guntur
$T_{12.}$	LCA-620	HRS, Lam farm, Guntur	$T_{45.}$	LCA-750	HRS, Lam farm, Guntur
$T_{13.}$	LCA-625	HRS, Lam farm, Guntur	$T_{46.}$	LCA-752	HRS, Lam farm, Guntur
$T_{14.}$	LCA-702	HRS, Lam farm, Guntur	$T_{47.}$	LCA-754	HRS, Lam farm, Guntur
$T_{15.}$	LCA-703	HRS, Lam farm, Guntur	$T_{48.}$	LCA-756	HRS, Lam farm, Guntur
$T_{16.}$	LCA-704	HRS, Lam farm, Guntur	$T_{49.}$	LCA-758	HRS, Lam farm, Guntur
$T_{17.}$	LCA-705	HRS, Lam farm, Guntur	$T_{50.}$	LCA-760	HRS, Lam farm, Guntur
$T_{18.}$	LCA-706	HRS, Lam farm, Guntur	$T_{51.}$	LCA-762	HRS, Lam farm, Guntur
$T_{19.}$	LCA-707	HRS, Lam farm, Guntur	$T_{52.}$	CA-960	HRS, Lam farm, Guntur
$T_{20.}$	LCA-708	HRS, Lam farm, Guntur	$T_{53.}$	HC-28	HAU, Hisar
$T_{21.}$	LCA-709	HRS, Lam farm, Guntur	$T_{54.}$	KT-I	IARI, Katrain
$T_{22.}$	LCA-710	HRS, Lam farm, Guntur	$T_{55.}$	Aparna	HRS, Lam farm, Guntur
$T_{23.}$	LCA-711	HRS, Lam farm, Guntur	$T_{56.}$	Pandava	Local collection, Guntur
$T_{24.}$	LCA-712	HRS, Lam farm, Guntur	$T_{57.}$	Pant C-1	GBPUA&T, Pantnagar
$T_{25.}$	LCA-713	HRS, Lam farm, Guntur	$T_{58.}$	Phule Jyoti	MPKV, Rahuri
$T_{26.}$	LCA-714	HRS, Lam farm, Guntur	$T_{59.}$	Punjab Gucchedar	PAU, Ludhiana
$T_{27.}$	LCA-715	HRS, Lam farm, Guntur	$T_{60.}$	Pusa Sadabahar	IARI, New Delhi
$T_{28.}$	LCA-716	HRS, Lam farm, Guntur	$T_{61.}$	Super-10	Local collection, Guntur
$T_{29.}$	LCA-718	HRS, Lam farm, Guntur	$T_{62.}$	Warangal Chapata	Local collection, Warangal
$T_{30.}$	LCA-720	HRS, Lam farm, Guntur	$T_{63.}$	LCA-334	HRS, Lam farm, Guntur
$T_{31.}$	LCA-722	HRS, Lam farm, Guntur			
$T_{32.}$	LCA-724	HRS, Lam farm, Guntur			
$T_{33.}$	LCA-726	HRS, Lam farm, Guntur			

The genotypes studied in a randomized block design were replicated twice. The nursery was raised during last week of July and the seedlings were transplanted at a spacing of 75 cm × 30 cm in a row 4m length during first fortnight of September. Each row consisted of 12 plants, of which five competitive plants were selected at random for recording the observations. The crop was raised as per the recommended package of practices. The observations were recorded on plant height (cm), number of primary branches plant^{-1}, days to 50 % flowering, per cent fruit set, number of fruits plant^{-1}, fruit diameter (cm), fruit length (cm), average dry fruit weight (g), number of seeds fruit^{-1} and dry fruit yield plant^{-1} (g), ascorbic acid (mg 100g^{-1}), oleoresin (%), capsaicin (%), total color value (ASTA units), red carotenoids (%) and

yellow carotenoids (%). The red ripe fruits were sun dried and ground in an electronic grinder and passed through a 0.5 mm sieve and the dry chilli powder was used to measure biochemical constituents except Vitamin 'C' content, for which mature green fruits were used. The following procedures were used for estimating the biochemical constituents.

1. Ascorbic acid (mg /100g)

Ascorbic acid content of mature green fruits was estimated by volumetric method (Sadasivam and Balasubramanian, 1987). Dye solution was prepared by dissolving 42 mg of sodium bicarbonate in distilled water taken into 200 ml volumetric flask, to which 52 mg of 2-6 dichlorophenol indophenol was added and the volume was made up to 200 ml with distilled water. Stock solution was prepared by dissolving 100 mg ascorbic acid in 100 ml of 4% oxalic acid solution and 10 ml of this stock solution was diluted to 100 ml with 4% oxalic acid to get the working standard of 100 mg per ml.

5 ml of the working standard solution was pipetted into a 100 ml of conical flask to which 10 ml of 4% oxalic acid was added. The contents were titrated against the dye (V_1ml) to get a pink end point. The chilli sample (5 g) was extracted in 4% oxalic acid and the volume was made up to 100 ml and the contents were centrifuged. 5 ml of this supernatant was pipetted out, to which 10 ml of 4 per cent oxalic acid was added and titrated against the dye (V_2 ml). The ascorbic acid content was calculated using the formula given below.

$$\text{Ascorbic acid (mg/100 g)} = (0.5\text{ mg} \div V1) \times (V2 \div 5\text{ml}) \times (100\text{ml} \div \text{Wt. of the sample}) \times 100$$

2. Oleoresin (%)

The oleoresin content was estimated as per the procedure given by Ranganna (1986). Finely mashed 25g chilli powder was transferred to a glass column, which was plugged by cotton plug on its narrow end. A thin layer of cotton was placed over chilli powder in the glass column and 25 ml of acetone was added. After all the acetone was decanted, 25 ml acetone was added each time till a total of 250 ml acetone was added to the contents. After decantatation, the resulting red colored liquid in beaker contains all the principle constituents of chilli. The collected filtrate was transferred to a 250 ml volumetric flask and the volume was made up with acetone. The chilli extract was transferred to a 250 ml beaker of known weight (W_1 g) and was kept in water bath at 50-60ºC for 15-30 minutes so that acetone gets evaporated. Then, weight of the beaker along with contents was recorded as W_2 g. The weight of the oleoresin content in the 25 g chilli powder was calculated and expressed in percentage using the given formula.

$$\text{Oleoresin content (\%)} = \left((W2 - W1) \div \text{Weight of sample}\right) \times 100$$

3. Capsaicin (%)

The capsaicin content of fruits was estimated by colorimetric method described by Bajaj *et al*., (1980). 0.5g dry chilli powder was weighed into glass-stoppard test tube; 10ml dry acetone (add 25g anhydrous sodium sulphate to 500ml of acetone at least one day before use) was added into the test tube and kept overnight for extraction. Next day samples were centrifuged at 10000 rpm for 10min to get clear supernatant. 1ml of the supernatant was taken into a test tube and evaporated to dryness in a hot water bath. Then, the residue was dissolved in 5ml of 0.4% of NaOH solution and 3ml of 3% phosphomolybdic acid was added. The contents were shaken and left undisturbed for 1hr. After 1hr, the solution was quickly filtered into centrifuge tubes to remove any floating debris, and then centrifuged at 5000rpm for 15min. The clear blue coloured solution was directly transferred into the cuvette and absorbance was read at 650nm along with a reagent blank. A standard graph was prepared using 0-200µg pure capsaicin. Simultaneously 0.2, 0.4, 0.6, 0.8 and 1ml of working standard solution (stock standard capsaicin solution was prepared by dissolving 50mg capsaicin in 50ml of 0.4% NaOH solution (1000µg/ ml) and working standard solution prepared by diluting the 10ml of the stock standard to 50ml with 0.4% NaOH solution (200µg/ ml)) was taken into new test tubes and proceeded as mentioned above. Per cent capsaicin calculated using the formula mentioned below

$$\text{Capsaicin content (\%)} = (\mu\text{g capsaicin} \times 100 \times 100) \div (1000 \times 1000 \times 1 \times 0.5)$$

4. Total color value (ASTA units):

Total extractable colour of fruits (ASTA- American Spice Trade Association units) was estimated as per the procedure given by Rosebrook *et al.,* (1968). 100mg of sieved fine chilli powder was weighed into a volumetric flask. Acetone was added and flask was closed tightly with stopper, then contents were kept for 16h at room temperature in dark and shaken intermittently. Solution was filtered using Whatman filter paper and final volume was made up to 100ml. Absorbance of final extract was read at 460nm using acetone as blank. ASTA color units were calculated as per the formula given below,

$$\text{ASTA} = (\text{Absorbance at 460 nm} \times 16.4) \div (\text{Weight of sample in g})$$

5. Determination of yellow and red fractions in chilli powder:

Total red (C^R; capsanthin, capsorubin and capsanthin-5, 6-epoxide) and yellow (C^Y; zeaxanthin, violaxanthin, antheraxanthin, β-cryptoxanthin, β-carotene and

cucurbitaxanthin A) carotenoid isochromic fractions were estimated following protocol of spectrophotometric method (Hornero-Mendez and Minguez-Mosquera, 2001). Dried chilli fruits were ground into a fine powder and 100mg of dried powder was extracted four times with 25ml acetone until the complete exhaustion of the color. The extract was filtered and transferred to 50ml volumetric flask and the volume was made up with acetone. The samples absorbance was read at two wavelengths *i.e.*, 472 and 508nm using acetone as blank. The red and yellow fractions were calculated using the following formulae.

$$C^R\,(\mu g/ml) = \left((A508 \times 2144.0) - (A472 \times 403.3)\right) \div 270.9$$

$$C^Y\,(\mu g/ml) = \left((A472 \times 1724.3) - (A508 \times 2450.1)\right) \div 270.9$$

The analysis of variance was carried out as per the procedure given by Panse and Sukhatme (1957).The character contribution towards genetic divergence was computed using the method given by Singh and Chaudhary (1977). Percentage contribution towards genetic divergence was calculated using the following formula

Percentage contribution of the character = (N × 100) ÷ M

Where, N = Number of genotype combinations where the character was ranked first.

M = All possible combinations of number of genotypes considered.

The genetic divergence was worked out among the genotypes using Mahalanobis D^2 statistics (Mahalanobis, 1936) and the D^2 values were calculated as

$$D^2_{ij} = \sum_{t=1} (Y_i^t - Y_j^t)^2$$

Where, Y_i^t is uncorrelated mean value of i^{th} genotype for character 't'

Y_j^t is uncorrelated mean value of j^{th} genotype for character 't'

D^2_{ij} is D^2 between i^{th} and j^{th} genotypes.

The genotypes were grouped into different clusters by employing Tocher's method as outlined by Rao (1952). For grouping of genotypes, D^2 values of all combinations of each genotype were arranged in ascending order of magnitude in a tabular form as described by Singh and Chaudhary (1977).To start with, two populations having the closest distance from each other were considered, to which the third population having the smallest D^2 value from the first two populations was added. Similarly, the next nearest fourth population was considered and this procedure was continued. At certain stage when it was felt that after adding a particular population there was an abrupt increase in the average D^2, that population was not considered for including in that cluster. The genotypes of the first cluster were then eliminated and the rest were treated in a similar way. This procedure was continued till all the genotypes were included into one or other cluster.

The average intra and inter cluster distances were calculated by the formula given by Singh and Chaudhary (1977).

Square of intra- cluster distance = $\Sigma Di^2 / n$

Square of inter- cluster distance = $\Sigma Di^2 / n_i\, n_j$

Where,

ΣDi^2= Sum of distance between all possible combinations.

n= Number of all possible combinations

n_i= Number of entries in cluster i

n_j= Number of entries in cluster j

RESULTS & DISCUSSION

The analysis of variance (ANOVA) revealed significant differences among 63 genotypes for quantitative and qualitative traits indicating the existence of variability among genotypes for characters studied (Table 2).

TABLE 2. Analysis of variance for various characters in chilli (*Capsicum annuum* L.)

S.No.	Character	Mean sum of squares		
		Replications	Genotypes	Error
1	Plant height (cm)	28.097	563.376**	43.543
2	Number of primary branches per plant	0.701	1.117**	0.219
3	Days to 50 per cent flowering	1.341	25.422**	3.954
4	Fruit set per cent	176.198*	501.725**	39.198
5	Number of fruits per plant	409.320	9125.453**	634.339
6	Fruit diameter (cm)	0.024**	0.276**	0.0007
7	Fruit length (cm)	0.956*	6.022**	0.234
8	Ascorbic acid (mg/100g)	4.371	4326.548**	100.724
9	Oleoresin (%)	0.944	6.103**	0.572
10	Capsaicin (%)	0.000007	0.022**	0.0006
11	Total colour value (ASTA Units)	35.914	1234.578**	32.894
12	Red carotenoids (%)	0.000096	0.0032**	0.000046
13	Yellow carotenoids (%)	0.000179*	0.0020**	0.000032
14	Average dry fruit weight (g)	0.00002	0.369**	0.028
15	Number of seeds per fruit	1.28	580.326**	80.323
16	Yield per plant (g)	2143.226	3553.576**	541.662

*: Significant at 5 % level; **: Significant at 1 % level

These findings are in accordance with the results of many earlier works (Farhad *et al.*, 2010; Kumar *et al.*, 2010; Shrilekha *et al*., 2011; Yatung *et al*., 2014).

The per cent contribution towards genetic divergence by all the 16 contributing characters is presented in table 3 & figure 1. The maximum contribution towards genetic divergence was by fruit diameter (44.14%) followed by yellow carotenoids (16.90%), red carotenoids (10.45%), ascorbic acid (10.19%), capsaicin (9.17%), fruit length (3.07%), total color value (2.10%), number of fruits per plant (1.43%), oleoresin (0.87%), number of seeds per fruit (0.61%), plant height (0.51%), fruit set % (0.31%), yield per plant (0.20%), average dry fruit weight (0.05%) whereas, remaining characters like number of primary branches per plant and days to 50 % flowering had no contribution towards genetic divergence.

TABLE 3. Relative contribution of different characters towards genetic divergence in chilli (*Capsicum annuum* L.)

Source	Times Ranked 1st	Contribution %
1. Plant height (cm)	10	0.51
2. Number of primary branches plant-1	0	0.00
3. Days to 50 per cent flowering	0	0.00
4. Fruit set per cent	6	0.31
5. Number of fruits plant-1	28	1.43
6. Fruit diameter (cm)	862	44.14
7. Fruit length (cm)	60	3.07
8. Ascorbic acid (mg /100g)	199	10.19
9. Oleoresin (%)	17	0.87
10. Capsaicin (%)	179	9.17
11. Total colour value (ASTA)	41	2.10
12. Red carotenoids (%)	204	10.45
13. Yellow carotenoids (%)	330	16.90
14. Average dry fruit weight (g)	1	0.05
15. Number of seeds fruit-1	12	0.61
16 Yield plant-1 (g)	4	0.20

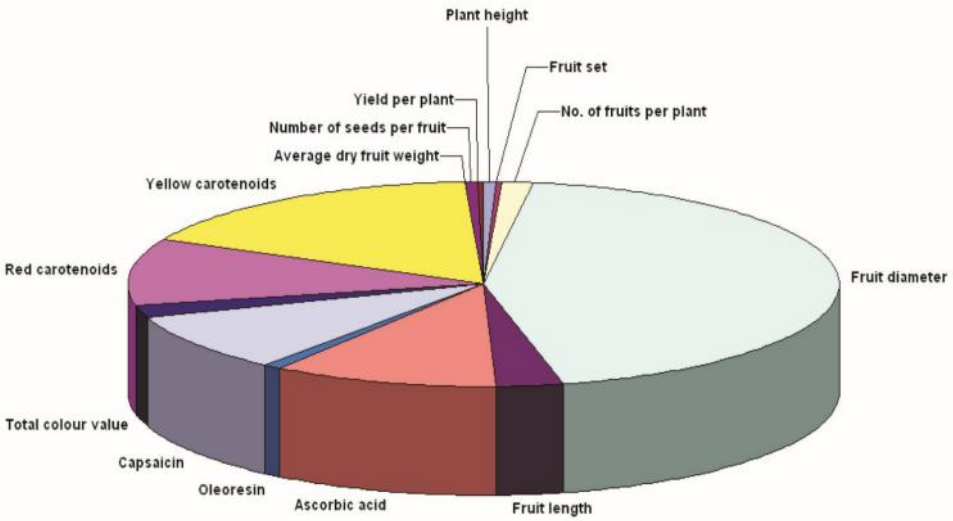

FIGURE 1: Relative contribution of different characters towards genetic divergence

The sixty three genotypes were grouped into 8 clusters (Table 4 and Figure 2). Clusters III and V were the largest containing 17 genotypes, followed by cluster IV (11), cluster I (8), cluster II (7). The clusters VI, VII and VIII were solitary clusters with genotypes LCA-706, Aparna and Warangal chapatta respectively showing zero intra-cluster D^2 values. The formation of distinct solitary clusters may be due to the fact that geographic barriers preventing gene flow and intensive natural and human selection for diverse and adoptable gene complexes must be responsible for this genetic diversity. The pattern of grouping of genotypes into different clusters was random and indicated that there is no parallelism between genetic divergence and geographical divergence of genotypes. Therefore, selection of genotypes for hybridization should be based on genetic diversity rather than geographical diversity. Vani *et al*. (2007) reported fourteen clusters with 55 genotypes, Dutonde *et al*. (2008) observed seven clusters with 40 accessions, Farhad *et al*. (2010) reported six clusters with 45 chilli genotypes, Shrilekha *et al*. (2011) reported seven clusters with 38

genotypes, Lahbib *et al*. (2012) grouped 11 landraces into three clusters and Yatung *et al*. (2014) observed six clusters with 30 chilli genotypes and these findings support the results of this investigation.

TABLE 4: Clustering of 63 chilli (*Capsicum annuum* L.) genotypes

Cluster	No. of genotypes	Name of genotypes
1 Cluster	8	LCA-704, LCA-705, LCA-754, LCA-206, LCA-718, LCA-750, LCA-715 and LCA-730
2 Cluster	7	LCA-748, LCA-334, LCA-235, LCA-744, LCA-726, LCA-722 and LCA-712
3 Cluster	17	LCA-315, LCA-762, LCA-436, LCA-734, LCA-752, LCA-424, LCA-305, Phule Jyoti , LCA-709, LCA-703, Punjab Gucchedar, Super-10, LCA-736, LCA-742 , LCA-740, LCA-625 and LCA-710
4 Cluster	11	LCA-353, LCA-716, LCA-756, LCA-724, LCA-714, Pusa Sadabahar, Pant C-1, LCA-758, G-4, LCA-738 and LCA-760
5 Cluster	17	LCA-357, LCA-713, LCA-728, KT-1, HC-28, Pandava, LCA-707, LCA-720, LCA-732, LCA-711, G-5, LCA-746, LCA-708, LCA-702, CA-960, LCA-620 and G-3
6 Cluster	1	LCA-706
7 Cluster	1	Aparna
8 Cluster	1	Warangal Chapatta

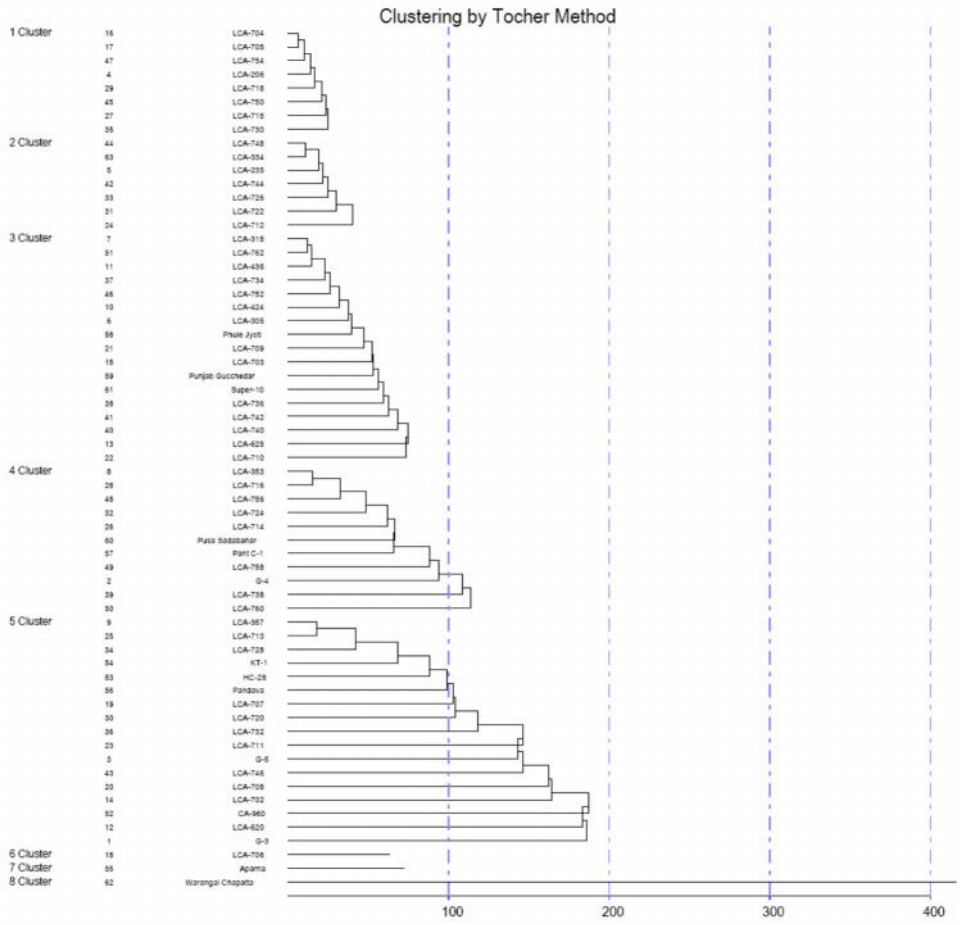

FIGURE 2: Dendrogram showing relationship of 63 chilli (*Capsicum annuum* L.) genotypes in eight clusters based on Mahalanobis' D^2 values

The intra- and inter- cluster distance represent the index of genetic diversity among clusters (Table 5 and Figure 3). Of the 8 clusters formed, the mean intra-cluster D^2 distance values ranged from a minimum of 0.00 (clusters VI, VII and VIII) to a maximum of 434.43 (cluster V). The intra cluster distance in other clusters *viz.*, cluster IV (259.50), cluster III (163.60), cluster II (93.56), cluster I (62.86), was in between this range. The high intra-cluster distance in cluster V indicates the presence of wide genetic diversity among the genotypes present within this cluster. The maximum inter-cluster distance was observed between cluster IV and VIII (4139.41) followed by cluster II and VIII (3633.27), cluster VI and VIII (3323.09) and cluster VII and VIII (3149.32), the minimum between clusters I and III (117.25). The hybrids of distant genotypes are reported to yield better (Kumar *et al*., 2010) and thus crosses between the genotypes from cluster IV and VIII can be used in chilli breeding to achieve maximum heterosis and to obtain heterotic hybrids and desirable segregants. The minimum inter-cluster distance was observed between genotypes of clusters I and III (117.25) which can be used for backcrossing programmes. The genotypes of cluster I and II (134.54) and cluster II and III (206.58) also have recorded minimum inter-cluster distance. The lowest inter-cluster distance between these cluster pairs suggested close proximity of genotypes of one cluster with those of the other cluster in respect of their genetic constitution. Several earlier reports (Mishra *et al*., 2004; Ajjapplavara, 2009; Kumar *et al.,* 2010; Suryakumari *et al*., 2010; Pandit *et al.,* 2010; Yatung *et al*., 2014) also indicate the presence of a high genetic divergence among chilli genotypes in their respective experiments. The genotypes grouped into the same cluster presumably diverge very little from one another and crossing of genotypes belonging to the same cluster is not expected to yield desirable segregants. Consequently, a crossing programme should be conducted with putative parents. Thus, crosses between the members of clusters separated by inter-cluster distances are likely to be beneficial for further improvement. D^2 cluster analysis revealed wide genetic distance (inter cluster) between the genotypes of cluster IV (LCA-353, LCA-716, LCA-756, LCA-724, LCA-714, Pusa Sadabahar, Pant C-1, LCA-758, G-4, LCA-738 and LCA-760) and VIII (Warangal chapatta). The crossing between genotypes of cluster IV & VIII can be exploited for the development of heterotic hybrids in future breeding programmes.

TABLE 5: Average intra (bold) and inter cluster D^2 values of eight clusters in chilli (*Capsicum annuum* L.)

Cluster	1 Cluster	2 Cluster	3 Cluster	4 Cluster	5 Cluster	6 Cluster	7 Cluster	8 Cluster
1 Cluster	62.86	134.54	117.25	292.68	331.58	260.02	309.56	2842.57
2 Cluster		93.56	206.58	265.05	551.16	272.09	337.29	3633.27
3 Cluster			163.60	317.45	364.72	307.18	431.22	2967.46
4 Cluster				259.50	668.82	374.99	509.29	4139.41
5 Cluster					434.43	600.47	722.07	2343.89
6 Cluster						0.00	376.38	3323.09
7 Cluster							0.00	3149.32
8 Cluster								0.00

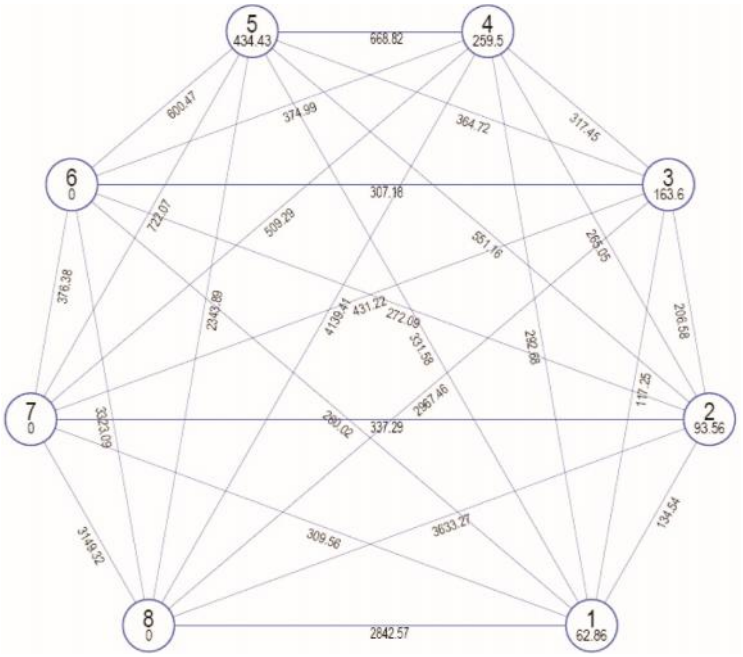

FIGURE 3. Intra-and inter-cluster distance of 63 chilli (*Capsicum annuum* L.) genotypes in eight clusters based on Mahalanobis' D^2 values

Cluster III earned highest cluster mean value for yellow carotenoids (0.08) (Table 6). On the other hand, Cluster IV produced highest mean value for no. of primary branches (3.95) and yellow carotenoids (0.08). Cluster V had the highest mean value for red (0.14) and yellow carotenoids (0.08) and cluster VI showed highest mean value for plant height (107.35), number of fruits per plant (480.00), yield per plant (204.18), capsaicin content (0.45) and had lowest mean value for days to 50% flowering (28.50). Cluster VII recorded highest mean value for per cent fruit set (56.00), fruit length (9.92) and ascorbic acid content (223.22) while cluster VIII recorded maximum fruit diameter (3.18), maximum dry fruit weight (3.35), more number of seeds per fruit (152.50), maximum oleoresin content (9.61) and high total colour value (105.00). The genotypes in cluster VI were flowered earlier and recorded higher yield. Genotypes of clusters V, VI and VIII showed better performance for quality traits. These clusters can be used in breeding programme for introgression of their desired quality genes into the high yielding varieties.

TABLE 6. Mean performance of yield per plant and its component characters in various clusters of chilli

Cluster No.	PH	NPBP	DFF	FSP	NFP	FD	FL	AA	O	C	TCV	RC	YC	ADFW	NSF	YP
1 Cluster	85.38	3.51	30.63	55.13	155.31	1.33	9.11	99.44	9.32	0.28	64.75	0.11	0.06	1.06	59.35	139.97
2 Cluster	89.58	3.77	31.86	52.71	199.99	1.09	9.09	109.83	8.75	0.24	52.05	0.11	0.04	1.00	57.13	166.78
3 Cluster	84.44	3.52	32.68	47.44	174.91	1.32	9.12	99.77	8.52	0.33	75.32	0.13	**0.08**	1.07	60.45	150.73
4 Cluster	84.37	**3.95**	30.50	54.41	197.86	0.99	7.66	143.41	9.53	0.37	81.63	0.13	**0.08**	0.77	50.89	138.19
5 Cluster	89.57	3.55	31.00	48.82	140.30	1.64	8.47	114.68	8.55	0.32	87.60	**0.14**	**0.08**	1.28	65.85	143.33
6 Cluster	**107.35**	3.00	**28.50**	48.00	**480.00**	1.24	6.98	118.35	9.15	**0.45**	43.14	0.12	0.03	0.77	73.30	**204.18**
7 Cluster	82.00	3.60	31.00	**56.00**	159.00	1.32	**9.92**	**223.22**	5.96	0.27	20.58	0.01	0.04	1.05	58.40	132.34
8 Cluster	106.30	2.80	34.00	32.50	49.80	**3.18**	8.71	90.00	**9.61**	0.30	**105.00**	0.04	0.04	**3.35**	**152.50**	107.30

Bold values indicate maximum mean performance

Where

PH – Plant Height (cm), NPBP –Number of Primary Branches per Plant (no.) , DFF – Days to 50 per cent Flowering , FSP – Fruit Set Per cent, NFP – Number of Fruits per Plant, FD – Fruit Diameter (cm), FL – Fruit Length (cm), AA – Ascorbic Acid (mg/100g), O – Oleoresin (%), C – Capsaicin (%), TCV – Total Color Value (ASTA units), RC – Red Carotenoids (%), YC – Yellow Carotenoids (%), ADFW – Average Dry Fruit Weight (g), NSF – Number of Seeds per Fruit, YP –Yield per Plant (g)

CONCLUSION

D^2 cluster analysis revealed wide genetic distance (inter cluster) between the genotypes of cluster IV (LCA-353, LCA-716, LCA-756, LCA-724, LCA-714, Pusa Sadabahar, Pant C-1, LCA-758, G-4, LCA-738 and LCA-760) and VIII (Warangal chapatta) and the crossing between genotypes of these two clusters can be exploited for the development of heterotic hybrids in future breeding programmes. The clusters III, IV, V, VI, VII and VIII were found superior for one or more characters. Therefore, a multiple crossing programme can be proposed involving genotypes from these clusters for the development of superior segregants in advanced generations with high yield potential combined with better quality in chilli.

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