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Research Article

Assessment of genetic divergence through multivariate analysis in chilli (*Capsicum annuum* L.)

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Abstract
An experiment was conducted to analyze the genetic diversity among 63 genotypes for ten quantitative and six qualitative characters in chilli at Horticultural Research Station, Lam, Guntur, Andhra Pradesh. The analysis of variance revealed significant differences among the genotypes for all the characters studied. Based on hierarchial cluster analysis, the 63 genotypes were grouped into 8 clusters. Among the clusters, cluster II was the largest containing 18 genotypes followed by cluster III with 15 genotypes and cluster VIII had only one genotype. The highest inter cluster distance was observed between cluster IV and VIII (7941.635) whereas the lowest was observed between cluster VI and VIII (2836.497). The Cluster VII exhibited highest intra cluster distance (614.548) and the lowest was observed in cluster VIII (0.00). The maximum contribution towards genetic divergence was shown by fruit diameter (44.14%) followed by yellow carotenoids (16.90%), red carotenoids (10.45%), ascorbic acid (10.19%) and capsaicin (9.17%). The principal component analysis revealed that first six principal components with eigen value more than one were observed to contribute 76.83 per cent towards the total variability. Among the six principal components, PC_1 contributed maximum towards variability (25.06%). Considering diversity pattern and horticultural performance, the genotypes Warangal chapatta, LCA-702, LCA-724, LCA-756, LCA-353 and LCA-716 were identified as promising parents and could be utilized for efficient hybridization in chilli.

Key words
Chilli, genetic divergence, hierarchical cluster analysis, principal component analysis.

Introduction

Chilli (*Capsicum annuum* L.) is known as the universal spice of India and has diverse utilities as a spice, condiment, culinary supplement, medicine, vegetable and ornamental plant. The important chilli growing states are Andhra Pradesh, Karnataka, Maharashtra, Orissa, Tamil Nadu and Madhya Pradesh. 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 (Farhad *et al*., 2010; Khodadabi *et al.,* 2011). 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 than those from closely related parents (Lahbib *et al*., 2012). Ward's minimum variance dendrogram (Hierarchial cluster analysis) creates sub group within a cluster, so relative position of the genotypes within the clusters can be examined by the dendrogram distance. In case of D^2 analysis, one can only know the intra-cluster distance but not relative position of the genotypes in the respective cluster. Principal component analysis facilitates in-depth analysis of genetic divergence between genotypes in terms of spatial distance. Thus, main objective of this study was to analyze the potential genetic diversity among genotypes of chilli and to classify the genotypes into different groups based on cluster analysis and principal component analysis and selection of suitable genotypes for further chilli hybridization programme.

Materials and methods

The present investigation was carried out during *kharif* 2012-13 at Horticultural Research Station, Lam, Guntur with 63 genotypes of chilli in a randomized block design with two replications. 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 of 4 m 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 parameters considered

for the study were plant height (cm), number of primary branches plant^{-1}, days to 50 per cent 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 yield plant^{-1} (g), ascorbic acid (mg/100g), oleoresin (%), capsaicin (%), total color value (ASTA units), red carotenoids (%) and yellow carotenoids (%). Analysis of variance was carried out as per the procedure given by Panse and Sukhatme (1957). Agglomerative heirarchial clustering technique (Ward's minimum variance) was followed for cluster analysis as given by Anderberg (1993) and principal component analysis (PCA) as per Jackson (1991).

Results and 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 1). The per cent contribution towards genetic divergence by all the 16 contributing characters is presented in Table 2. The maximum contribution towards genetic divergence was shown 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%), per cent fruit set (0.31%), yield per plant (0.20%) and average dry fruit weight (0.05%). Selection for divergent parents based on these characters will be useful for heterosis breeding in chilli. The characters like number of primary branches per plant and days to 50 per cent flowering had no contribution towards genetic divergence.

The 63 genotypes were grouped into 8 clusters of which cluster II was the largest containing 18 genotypes followed by cluster III with 15 genotypes and cluster IV with 10 genotypes (Table 3 and Figure 1). The cluster V consisted of 6 genotypes followed by cluster VII with 5 genotypes, cluster I and VI with 4 genotypes and cluster VIII with 1 genotype (Warangal chapatta). This random distribution of genotypes indicated absence of parallelism between geographical and genetic diversity. Farhad *et al*. (2010) reported six clusters with 45 chilli genotypes and Shrilekha *et al.* (2011) reported seven clusters with 38 genotypes and Lahbib *et al.* (2012) grouped 11 landraces into three clusters and these findings support the results of this investigation.

The intra- and inter- cluster distance represent the index of genetic diversity among clusters (Table 4 and Figure 2). Of the 8 clusters formed, the mean intra-cluster Euclidean^{2} distance values ranged from a minimum of 0.00 in cluster VIII to a maximum of 614.548 in cluster VII. The intra cluster distance in other clusters *viz.*, cluster VI (236.271), cluster II (278.175), cluster III (291.256), cluster V (301.821), cluster IV (370.385) and cluster I (464.183) was found to be intermediate.. The high intra-cluster distance in cluster VII indicated 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 (7941.635) followed by cluster V and VIII (7542.904) and cluster III and VIII (6889.590), the minimum between cluster II and III (420.652). 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 cluster II and III (420.652) which can be used for backcross breeding programmes. The genotypes of cluster III and IV (547.233) and cluster I and II (578.721) 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. The findings of inter and intra cluster distances are in conformity with earlier works of Roy and Sorma (1996), Mishra *et al*., (2004), Farhad *et al*. (2010), Kumar *et al*. (2010), and Lahbib *et al.* (2012). 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. Agglomerative cluster analysis revealed wide genetic distance (inter cluster) between the genotypes of cluster IV (LCA-353, LCA-716, LCA-756, LCA-724, LCA-703, Punjab Gucchedar, Pusa Sadabahar, LCA-714, Pant C-1 and LCA-710) and VIII (Warangal chapatta); cluster V (LCA-357, LCA-713, LCA-758, LCA-760, LCA-738 and LCA-728) and VIII (Warangal chapatta). The crossing between genotypes of cluster IV & VIII and V & VIII can be exploited for the development of heterotic hybrids in future breeding programmes.

Cluster I showed high mean values for number of fruit per plant; cluster III for dry fruit yield per plant; cluster IV for number of primary branches per plant, per cent fruit set, ascorbic acid, oleoresin

and capsaicin; Cluster V for fruit length, total color value, red and yellow carotenoids; cluster VII for plant height; Cluster VIII for fruit diameter, average dry fruit weight and number of seeds per fruit and the cluster VI showed lower mean value for days to 50% flowering (Table 5). Genotypes of clusters IV and V which showed better performance for quality traits can be used in breeding programme for introgression of their desired genes into the high yielding varieties. The clusters I, III, IV, V, VII and VIII were found superior for one or more characters. Therefore, a multiple crossing programme can be proposed involving genotypes from these clusters to isolate superior segregants in advanced generations with high genetic yield potential and other desirable characters in chilli.

In the present investigation, the principal components with eigen values more than one were retained and less than one were considered as non-significant. The first six principal components with eigen values more than one contributed 76.83 per cent towards the total variability. The first PC explained 25.06 per cent of the total variability in the set of all variables and remaining ones accounted for progressively lesser amount of variation (Table 6) and the characters viz., number of seeds fruit^{-1}, total color value, ascorbic acid, number of fruits plant^{-1}, average dry fruit weight and fruit diameter significantly loaded in PC_1 and contributed more towards variability.

The hierarchial cluster analysis and principal component analysis confirmed the findings of each other. Results of cluster analysis based on PCA scores were compared with the results of the principal component analysis on a visual aid in desecrating clusters in the 2D and 3D scattered diagrams (Figures 3 & 4). The genotypes falling in same cluster were present closer to each other in scattered diagram thereby confirming the results of cluster analysis. 2D and 3D graphs showed wide divergence between Warangal chapatta and LCA-724, LCA-756, LCA-353, LCA-716, Aparna which are also distantly placed with LCA-702 signifying their usefulness in chilli breeding to develop high heterotic hybrids. Utilization of principal component analysis combined with clustering of Ward's method in genetic divergence studies of chilli was supported by Thul *et al.* (2009), Kadri *et al.* (2009), Farhad *et al*. (2010), Sudre *et al*. (2010), Shrilekha *et al.* (2011) and Lahbib *et al.* (2012).

Both the methods of grouping revealed a single concept of non-correspondence of genetic divergence and geographic diversity. In a broad sense both the methods of classifying the genotypes into different groups are equally useful but hierarchial cluster analysis gave an additional advantage of identifying sub-clusters of the major groups at different levels so that each small group can be critically analyzed and exploited in the breeding programmes. All the 63 genotypes were grouped into 8 clusters. The genotypes Warangal chapatta, LCA-702, LCA-724, LCA-756, LCA-353 and LCA-716 showed maximum inter-cluster distance in cluster analysis and principal component analysis. So they can be exploited for the development of heterotic hybrids in future breeding programmes. From this study, it may be concluded that a wide range of variation for almost all the economically important traits are observed in this crop. This implies a great potential for breeding through hybridization programme or direct use as variety for successful chilli production. Further, one or two promising genotypes from different clusters may be chosen for further genetic studies either by way of diallel or line x tester analysis. The genetically divergent genotypes may be used as mapping populations to detect diversity at molecular level and also to identify molecular markers linked to desirable traits for marker assisted selection (MAS).

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References

Anderberg, M.R. 1993. Cluster Analysis for Application, Academic Press, New York.

Asati, B.S. and Yadav, D.S. 2004. Diversity of horticultural crops in north eastern region. *ENVIS Bull Him Eco*., **12**: 1-11.

Farhad, M., Hasanuzzaman, M., Biswas, B.K., Arifuzzaman, M. and Islam, M.M. 2010. Genetic divergence in chilli (*Capsicum annum L.*). *Bangladesh Res. Pub. J*., **3**: 1045-1051.

Guerra, E.P., Destro, D., Miranda, L.A. and Montalvan, R. 1999. Parent selection for intercrossing in food type soybean through multivariate genetic divergence. *Acta Sci*., **21**(3): 429-437.

Jackson, J.E. 1991. A User's Guide to Principal Components, John Wiley and Sons Inc., New York.

Kadri, M.B., Eşiyok, D. and Turhan, K. 2009. Patterns of phenotypic variation in a germplasm collection of pepper (*Capsicum annuum* L.) from Turkey. *Spanish J. Agri. Res*., **7**: 83-95.

Khodadabi, M., Fotokian, M.H. and Miransari, M. 2011. Genetic diversity of wheat genotypes based on cluster and principal component analysis for breeding strategies. *Australian J. Crop Sci*., **5**(1): 17-24.

Kumar, B.M., Kantti, D. and Mallikarjunaiah, H. 2010. Genetic divergence in chilli accessions. *Electronic J. Plant Breed*., **1**(5): 1363-1366.

Lahbib, K., Bnejdi, F. and Mohamed, El.G. 2012. Genetic diversity evaluation of pepper (*Capsicum annuum* L.) in Tunisia based on morphologic characters. *African J. Agri. Res*., **7**: 3413-3417.

Mishra, A.C., Singh, R.V. and Ram, H.H. 2004. Studies on genetic divergence in capsicum (*Capsicum annuum* L.) in Uttaranchal. *Capsicum and Eggplant Newsl*., **23**: 45-48.

Roy, A. and Sorma, R.N. 1996. Multivariate analysis in chilli (*Capsicum annuum* L.). *Annals of Agril. Res*., **17**(2): 130-132.

Shrilekha, M., Lal, R.K., Darokar, M.P. and Khanuja, S.P.S. 2011. Genetic variability in germplasm accessions of *Capsicum annuum* L. *American J. Plant Sci*., **2**: 629-635.

Sudre, C.P., Goncalves, L.S.A., Rodrigues, R., Amaral, J.A.T., Riva-Souza, E.M. and Bento, C.S. (2010) Genetic variability in domesticated *Capsicum* spp as assessed by morphological and agronomic data in mixed statistical analysis. *Genet.Mol. Res*., **9**: 283-294.

Thul, S.T., Lal, R.K., Shasany, A. K., Darokar, M.P., Gupta, A.K., Gupta, M.M., Verma, R. K. and Khanuja ,S.P.S. 2009. Estimation of phenotypic divergence in a collection of Capsicum species for yield-related traits. *Euphytica.*

Table 1. Analysis of variance for 16 characters in chilli

		Mean sum of squares		
S.No.	Character	Replications	Genotypes	Error
1	Plant height (cm)	28.097	563.376**	43.543
2	Primary Branches/ Plant (no.)	0.701	1.117**	0.219
3	Days to 50% Flowering	1.341	25.422**	3.954
4	Fruit Set (%)	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 content (mg/100g)	4.371	4326.548**	100.724
9	Oleoresin (%)	0.944	6.103**	0.572
10	Capsaicin Content (%)	0.000007	0.022**	0.0006
11	Color 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	Dry fruit yield per plant (g)	2143.226	3553.576**	541.662

Table 2. Relative contribution of different characters towards genetic divergence in chilli

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

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Table 3. Clustering pattern of 63 chilli genotypes

Cluster No.	No. of genotypes	Name of genotypes
1 Cluster	4	G-3, LCA-620, LCA-706 and Aparna
2 Cluster	18	LCA-206, LCA-718, LCA-436, LCA-750, LCA-704, LCA-705, LCA-754, Super-10, LCA-305, LCA-315,LCA-762, LCA-752, LCA-424, Phule Jyoti, LCA-709, LCA-746, Pandava and LCA-732
3 Cluster	15	G-4, LCA-736, LCA-748, LCA-334, LCA-625, LCA-712, LCA-715, LCA-722, LCA-730, LCA-726,LCA-744, LCA-235, LCA-734, LCA-742 and LCA-740
4 Cluster	10	LCA-353, LCA-716, LCA-756, LCA-724, LCA-703, Punjab Gucchedar, Pusa Sadabahar, LCA-714, Pant C-1 and LCA-710
5 Cluster	6	LCA-357, LCA-713, LCA-758, LCA-760, LCA-738 and LCA-728
6 Cluster	4	G-5, LCA-708, LCA-711 and CA-960
7 Cluster	5	LCA-707, HC-28, LCA-720, KT-1 and LCA-702
8 Cluster	1	Warangal Chapatta

Table 4. Average intra (bold) and inter cluster Euclidean^2 values of eight clusters in chilli

Cluster	I	II	III	IV	V	VI	VII	VIII
I	464.183	578.721	605.103	683.154	1345.526	1044.877	1567.917	5913.015
II		278.175	420.652	647.227	646.62	697.290	754.180	5358.971
III			291.256	547.233	611.918	1263.919	1194.593	6889.590
IV				370.385	787.896	1749.573	1623.893	7941.635
V					301.821	1715.499	959.175	7542.904
VI						236.271	837.344	2836.497
VII							614.548	4293.503
VIII								0.000

Table 5. Mean performance of yield plant^{-1} 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
I	89.800	3.475	30.500	43.750	245.925	1.336	8.290	141.889	8.921	0.377	39.415	0.065	0.026	0.945	66.975	170.701
II	80.303	3.539	31.861	53.333	164.242	1.429	8.576	118.119	8.561	0.303	74.351	0.124	0.072	1.101	59.350	141.749
III	95.247	3.573	32.500	51.533	196.493	1.159	9.490	107.158	8.892	0.257	61.616	0.114	0.056	1.093	62.437	174.847
IV	80.785	4.140	31.000	55.800	190.840	1.026	7.322	142.443	9.687	0.448	69.922	0.109	0.074	0.688	48.320	128.144
V	88.617	4.000	28.917	44.833	176.883	1.138	9.675	97.753	9.235	0.279	118.284	0.181	0.119	0.935	57.617	149.652
VI	69.375	2.750	28.875	45.875	111.325	1.955	7.178	105.921	7.690	0.255	62.259	0.112	0.056	1.432	74.075	129.154
VII	107.090	3.420	32.700	46.100	102.820	1.730	9.378	78.713	7.964	0.343	107.112	0.168	0.111	1.478	63.080	117.981
VIII	1060.30	2.800	34.000	32.500	49.800	3.175	8.710	90.000	9.610	0.295	1050.00	0.040	0.035	3.350	152.500	107.300

PH – Plant Height (cm), NPBP –Number of Primary Branches Plant^{-1} (no.) , DFF – Days to 50 per cent Flowering , FSP – Fruit Set Per cent, NFP – Number of Fruits Plant^{-1}, 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 Fruit^{-1}, YP –Yield Plant^{-1}(g)

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Table 6. Eigen values, proportion of the total variance, cumulative per cent variance and component loading of different characters in chilli

	PC_1	PC_2	PC_3	PC_4	PC_5	PC_6
Eigene Value (Root)	4.010	2.583	1.952	1.524	1.173	1.052
% Var. Exp.	25.059	16.141	12.197	9.525	7.329	6.574
Cum. Var. Exp.	25.059	41.201	53.398	62.923	70.252	76.826
Plant height (cm)	0.126	0.092	0.182	0.522	0.139	0.445
Number of primary branches plant^{-1}	-0.162	0.130	-0.247	-0.181	0.249	0.512
Days to 50 per cent flowering	0.064	-0.019	0.046	0.474	0.262	-0.077
Fruit set per cent	-0.180	0.076	-0.030	0.010	-0.711	0.214
Number of fruits plant^{-1}	-0.339	0.133	-0.095	0.109	-0.013	-0.257
Fruit diameter (cm)	0.316	-0.411	0.100	-0.019	-0.219	0.017
Fruit length (cm)	0.136	0.358	0.327	-0.194	0.064	0.412
Ascorbic acid (mg/100g)	-0.374	0.098	-0.131	0.272	-0.166	0.106
Oleoresin (%)	-0.074	-0.299	-0.017	0.533	-0.059	0.023
Capsaicin (%)	-0.190	-0.124	-0.402	-0.053	0.375	0.079
Total colour value (ASTA Units)	0.397	0.133	-0.298	0.018	-0.098	-0.062
Red carotenoids (%)	0.197	0.397	-0.264	0.100	0.007	-0.378
Yellow carotenoids (%)	0.149	0.250	-0.517	0.121	-0.082	0.139
Average dry fruit weight (g)	-0.335	0.184	0.325	-0.038	0.235	-0.232
Number of seeds fruit^{-1}	0.419	0.098	0.127	0.050	0.156	-0.053
Yield plant^{-1} (g)	-0.012	0.509	0.224	0.171	-0.157	-0.105

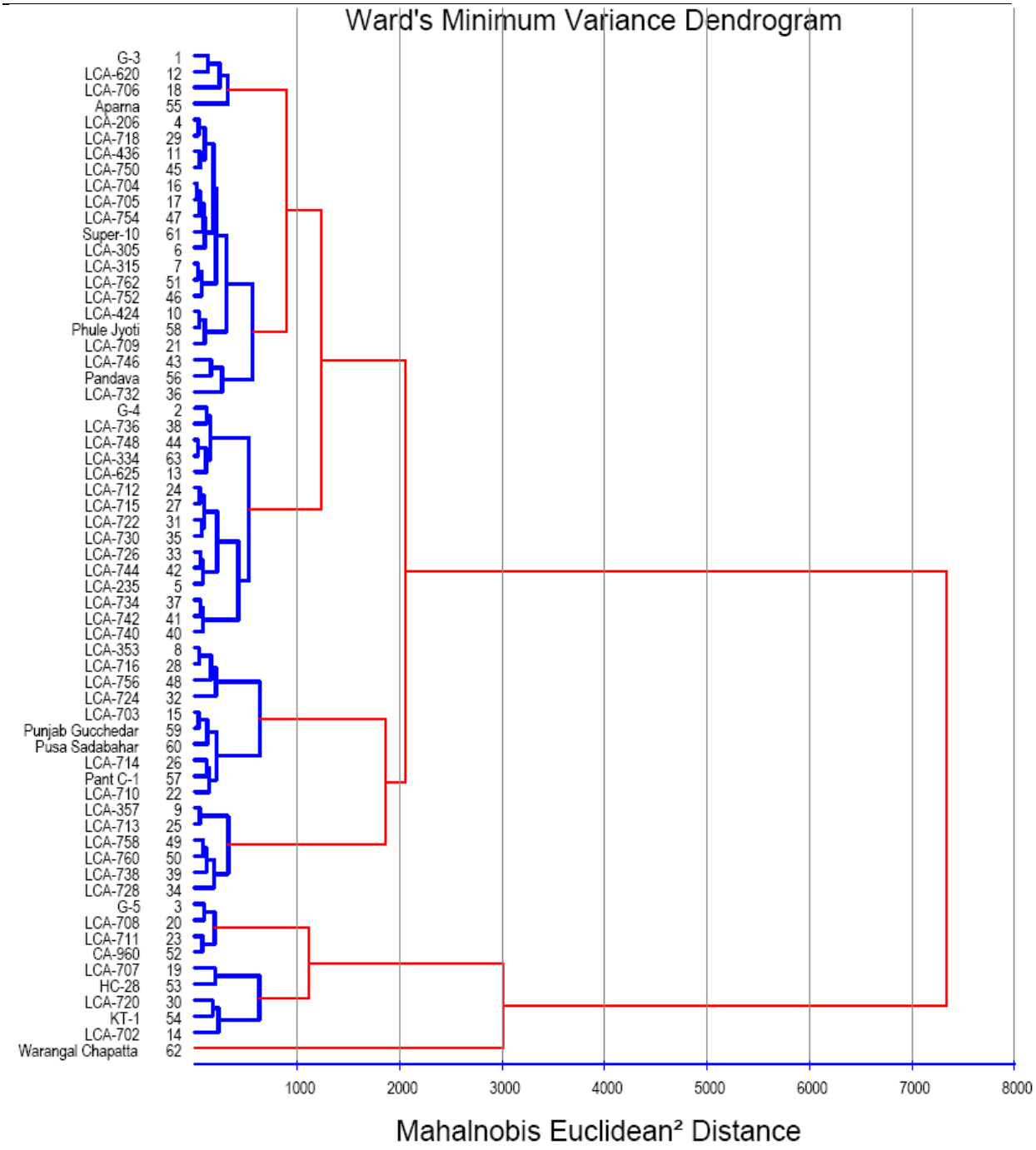

Figure 1. Dendrogram showing relationship of 63 chilli genotypes in eight clusters based on Euclidean^2 distance

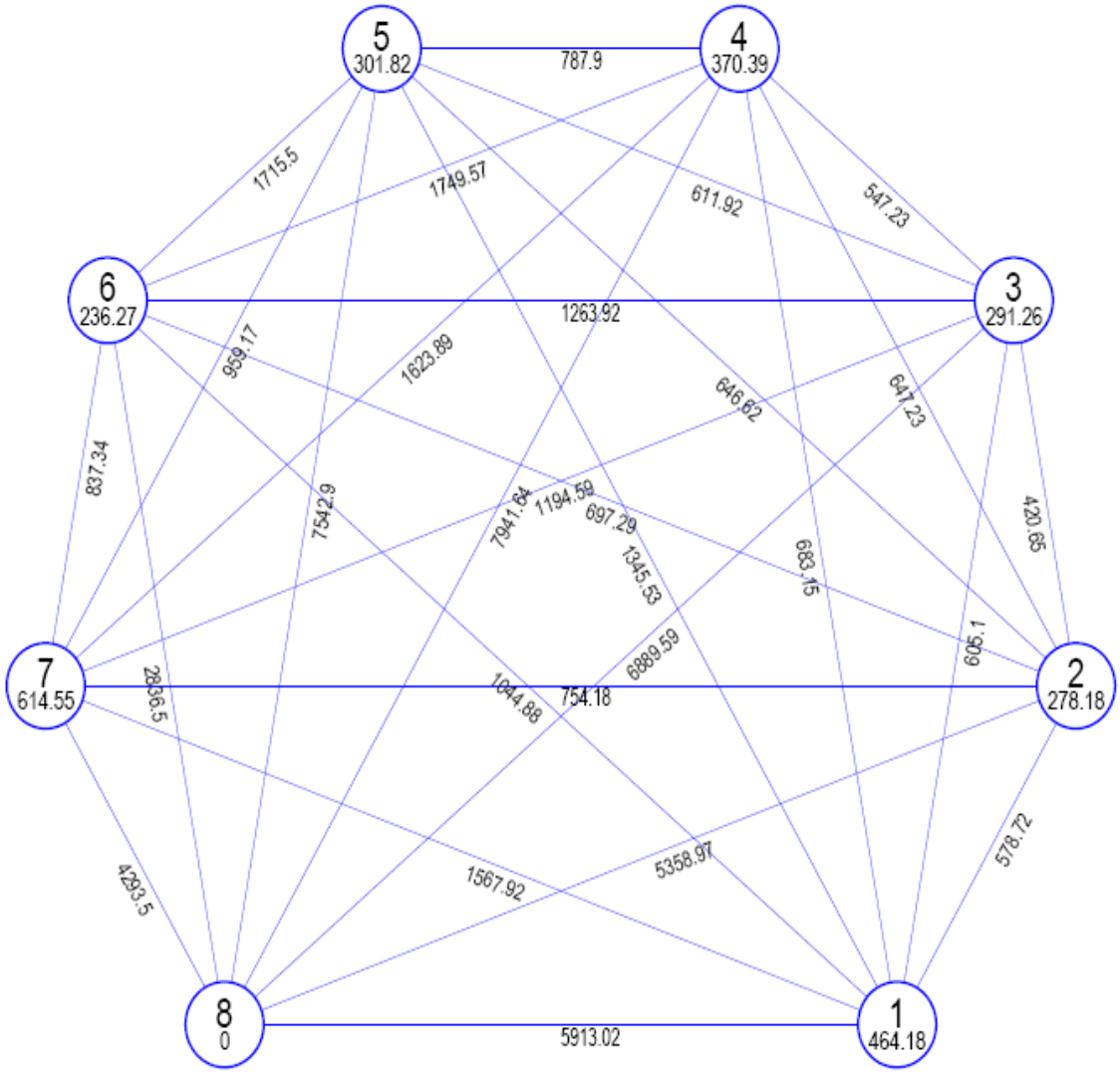

Figure 2. Intra-and inter-cluster distance of 63 chilli genotypes in eight clusters based on Euclidean^2 distance

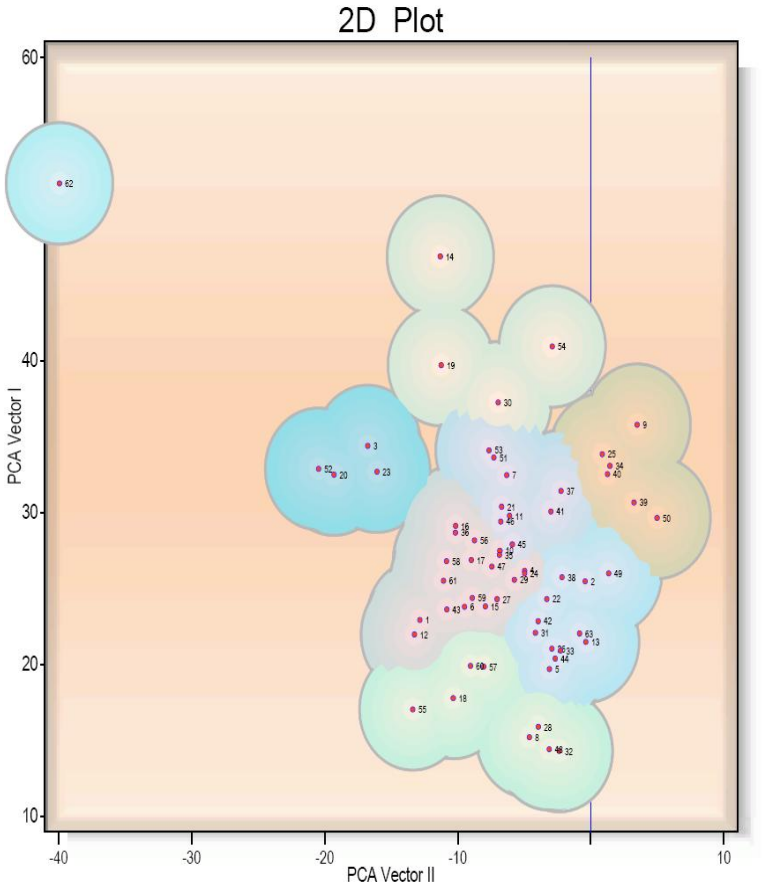

Figure 3. Two dimensional graph showing relative position of 63 chilli genotypes based on PCA scores

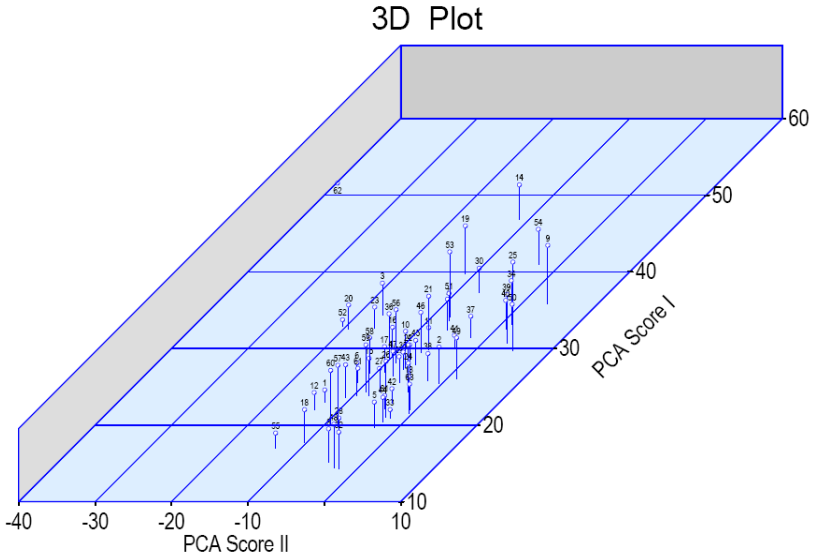

Figure 4. Three dimensional graph showing relative position of 63 chilli genotypes based on PCA scores