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Assessment of Genetic Divergence in Tomato (*Solanum lycopersicum* L.) through Clustering and Principal Component Analysis under Mid Hills Conditions of Himachal Pradesh, India

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A B S T R A C T

The nature and magnitude of genetic divergence was estimated in 35 genotypes of tomato using Mahalanobis D^2 – statistics. The genetic material revealed considerable amount of diversity for all the characters investigated. All the genotypes were grouped into 4 clusters. Maximum number of genotypes was accommodated in cluster III. The intra cluster distance was maximum in cluster III (3.103) and minimum in cluster IV (2.435). The inter cluster distance was found maximum to the tune of 4.790 between cluster I and IV and minimum (2.765) between cluster II and IV, indicating that hybridization between the genotypes from cluster I and IV can be utilized for getting superior recombinants/transgressive segregants in segregating generations of tomato. Principal component (PC) analysis depicted first four PCs with Eigen-value higher than 1 contributing 72.97% of total variability for different traits. The PC-I showed positive factor loadings for for most of the traits except fruit shape index, number of locules per fruit, pericarp thickness and harvest duration.

Introduction

Tomato *(Solanum lycopersicum* L.) is one of the important vegetables grown throughout the world and occupying prime position among processed vegetable. It is one of the most popular vegetable in India and is grown in tropical, subtropical and mild cold climate regions. Varsality of tomato in fresh and processed form plays major role in its rapid and wide spread adoption as an important food commodity. Tomato is most remunerative cash crop of mid hills of Himachal Pradesh being grown as an off season vegetable for fresh market and supply the produce to the plains of northern India. Longer harvesting period and off season production of tomato make this crop more suitable for cultivation in mid-hills conditions. The productivity of tomato grown in the region is much less than its potential yield due to the non availability high yielding disease and insect pest resistant cultivar for growing in hilly areas. Realizing this, there is a need for continuous crop improvement in tomato which can be achieved by isolating superior breeding lines/varieties having desirable horticultural traits and insect- pest

resistance. Progress in breeding for economic characters often depends upon the availability of germplasm representing a diverse genetic origin and has crucial role in sustaining and strengthening the food and nutrition security of the country. Estimation of genetic distance is one of appropriate tools for parental selection in tomato hybridization programs. Appropriate selection of the parents is essential to be used in crossing to enhance the genetic recombination for potential yield increase. Some appropriate methods, factor analysis, cluster analysis and PCA, for parental selection and genetic diversity identification. D^2 statistics offers a reliable technique to estimate the genetic divergence available in the population (Mahalanobis, 1936).

Principal component analysis helps researchers to distinguish significant relationship between traits. The main advantage of using PCA over cluster analysis is that each genotype can be assigned to one group only. Hybridization programme involving genetically diverse parents belonging to different clusters would provide an opportunity for bringing together gene constellations of diverse nature. Following hybridization, these parental combinations can possibly produce progenies with elevated genetic variability, thereby increasing chances of creating superior genotypes with traits of interest (Crossa and Franco, 2004). For those traits, where selection is not responsive and non-additive gene effects are playing major role in the expressions, hybridization between diverse parents on the basis of their mean performance to get superior hybrids or transgressive segregants or partitioning of additive genetic variation and non additive genetic variation in segregating generations will be useful. Therefore, studies on genetic divergence will be helpful in identification of better parents. Keeping this in view, present investigation was carried out on 35 genotypes of tomato to study the nature and magnitude of genetic divergence.

Materials and Methods

The present investigation was carried out at the experimental farm of the Department of Vegetable Science, Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh during *Kharif* season of 2013. Thirty five genotypes of tomato including one check Solan Lalima were laid out in a Randomized Complete Block Design with three replications. The genotypes along with their sources are presented in Table 1. The plot size was 2.0 m × 1.8 m with 90 cm and 30 cm spacing between rows and plants respectively. The standard cultural practices recommended in the Package of Practices of Vegetable Crops were followed to produce a healthy crop stand (Anonymous, 2013).

Data were recorded on ten randomly selected plants from each genotype and each replication and their means were worked out for statistical analysis. The mean values of data were subjected to analysis of variance as described by Gomez and Gomez (1983). The observations were recorded on days to 50% flowering, number of fruits per cluster, number of fruits per plant, average fruit weight (g), fruit shape index, number of locules per fruit, pericarp thickness (mm), plant height (cm), harvest duration (days), internodal diatance (cm), days to marketable maturity, total soluble solids (˚Brix), ascorbic acid content (mg/100g) and fruit yield per plant (kg).

The data were subjected to Mahalanobis's D^2 statistics (Mahalanobis 1936). Treating D^2 as the generalized statistical distance between a pair of populations (genotypes), all populations were grouped into number of clusters according to method described by (Rao, 1952). Principal component analysis

was done using computer software Microsoft Excel along with XLSTAT.

Results and Discussion

The analysis of variance revealed highly significant differences among the genotypes for all the characters studied, indicating the existence of wide genetic divergence among them. On the basis of performance of various traits, the clustering pattern of 35 diverse genotypes of tomato has been presented in the table 2. All the genotypes were grouped into 4 clusters. Maximum number of genotypes was accommodated in cluster III (10) followed by cluster I (9), cluster IV (9) and cluster II (7), respectively. Average of inter and intra cluster divergence (D^2) values have been presented in the table 3. The diagonal figures in the table represent the intra cluster distances. The intra cluster distance was maximum in cluster III (3.103) and minimum in cluster IV (2.435), whereas, highest inter cluster distance (4.774) was recorded between I and IV and lowest (2.767) was observed between cluster II and IV. Since crossing of genotypes belonging to same cluster do not expect to yield superior hybrids or segregants, inter cluster distances were also worked out. The cluster means for various horticultural traits have been presented in the table 4. Minimum days taken to 50% flowering were recorded in cluster I (30.67). Maximum number of fruits per cluster was recorded in cluster II (5.87). Maximum number of fruits per plant was recorded in cluster IV (35.83) followed by cluster II (35.71), cluster I (16.09) and cluster III (13.51). Maximum average fruit weight was recorded in cluster IV (64.34) followed by cluster III (62.41), cluster I (52.71) and cluster II (48.28). Maximum fruit shape index values for fruit shape index were recorded in cluster III (1.10) followed by cluster I (1.01), clusters II (0.93) and cluster IV (0.88). Minimum number of locules per fruits was recorded in cluster III (2.98). Maximum pericarp thickness was recorded in cluster IV (6.16). Maximum plant height was recorded in cluster IV (168.78) followed by cluster II (131.79), cluster III (85.44) and cluster I (84.35). Maximum harvest duration was recorded in cluster IV (36.67) followed by cluster II (35.95), cluster I (28.96) and cluster III (27.53). Minimum internodal distance was recorded in cluster II (9.55) followed by cluster III (9.64), cluster I (9.67) and cluster IV (10.92). The minimum days to marketable maturity was recorded in cluster I (68.56) followed by cluster II (70.43), cluster IV (71.78) and cluster III (74.67). Maximum total soluble solids were recorded in cluster IV (4.16) followed by cluster III (3.82), cluster II (3.59) and cluster I (3.59). Maximum ascorbic acid content was recorded in cluster III (24.02) followed by cluster IV (23.14), cluster II (19.91) and cluster I (18.50). Highest fruit yield per plant was recorded in cluster IV (2.18) followed by cluster II (1.63), cluster III (0.82) and cluster I (0.82). Information on genetic diversity was also used to identify the promising diverse genotypes, which may be used in further breeding programmes. Genotypes from same centre of origin were placed in separate clusters, indicating wide genetic diversity among them. This may be due to frequent exchange of germplasm between different geographical regions. The inter cluster distance was maximum between cluster I and IV and minimum between cluster II and IV, indicating that hybridization between the genotypes from cluster I and IV can be utilized for getting superior recombinants/transgressive segregants in segregating generations of tomato.

Furthermore, for getting the reliable conformity on the basis of cluster means, the important cluster for different traits were i.e. cluster I for days to 50% flowering and days to marketable maturity.

Table.1 List of tomato genotypes studied along with their sources

Sr. No.	Genotype	Source
1	EC-1749/3	NBPGR, New Delhi
2	EC-8910-155	NBPGR, New Delhi
3	EC-37239	NBPGR, New Delhi
4	EC-191531	NBPGR, New Delhi
5	EC-191535-3	NBPGR, New Delhi
6	EC-267727	NBPGR, New Delhi
7	EC-535580	NBPGR, New Delhi
8	EC-620370	NBPGR, New Delhi
9	EC-620374	NBPGR, New Delhi
10	EC-620375	NBPGR, New Delhi
11	EC-620378	NBPGR, New Delhi
12	EC-620383	NBPGR, New Delhi
13	EC-620396	NBPGR, New Delhi
14	EC-620397	NBPGR, New Delhi
15	EC-620398	NBPGR, New Delhi
16	EC-620400	NBPGR, New Delhi
17	EC-620402	NBPGR, New Delhi
18	EC-620407	NBPGR, New Delhi
19	EC-620410	NBPGR, New Delhi
20	EC-620424	NBPGR, New Delhi
21	EC-620434	NBPGR, New Delhi
22	EC-620435	NBPGR, New Delhi
23	JTS-1-1	RHRS, Jachh
24	JTS-1-3	RHRS, Jachh
25	JTS-7-6	RHRS, Jachh
26	JTS-10-1	RHRS, Jachh
27	JTS-10-2	UHF, Nauni, Solan
28	JTS-10-3	UHF, Nauni, Solan
29	JTS-10-10	RHRS, Jachh
30	LE-79-5	RHRS, Bajaura
31	BT-1	UHF, Nauni, Solan
32	BT-10	UHF, Nauni, Solan
33	Yalabingo	UHF, Nauni, Solan
34	Arka Keshav	IIHR, Bangalore
35	Solan Lalima (Check Variety)	UHF, Nauni, Solan

Table.2 Clustering pattern of 35 genotypes of tomato on the basis of genetic divergence

Cluster	Number of genotypes	Genotypes
I	9	EC-620383, EC-620397, EC-620398, EC-620400, EC-620407, EC-620410, EC-620424, EC-620434, BT-1
II	7	EC-8910-155, EC-191531, EC-191535-3, EC-535580, JTS-10-3, JTS-10-10, LE-79-5
III	10	EC-620370, EC-620374, EC-620375, EC-620378, EC-620396, EC-620402, EC-620435, JTS-1-3, JTS-7-6, Arka Keshav
IV	9	EC-1749/3, EC-37239, EC-267727, JTS-1-1, JTS-10-1, JTS-10-2, BT-10, Yalabingo, Solan Lalima

Table.3 Average intra and inter cluster distance (D2)

Cluster	I	II	III	IV
I	2.477			
II	3.255	2.733		
III	2.982	4.244	3.103	
IV	4.774	2.767	4.697	2.435

Table.4 Cluster mean for different characters among 35 genotypes of tomato

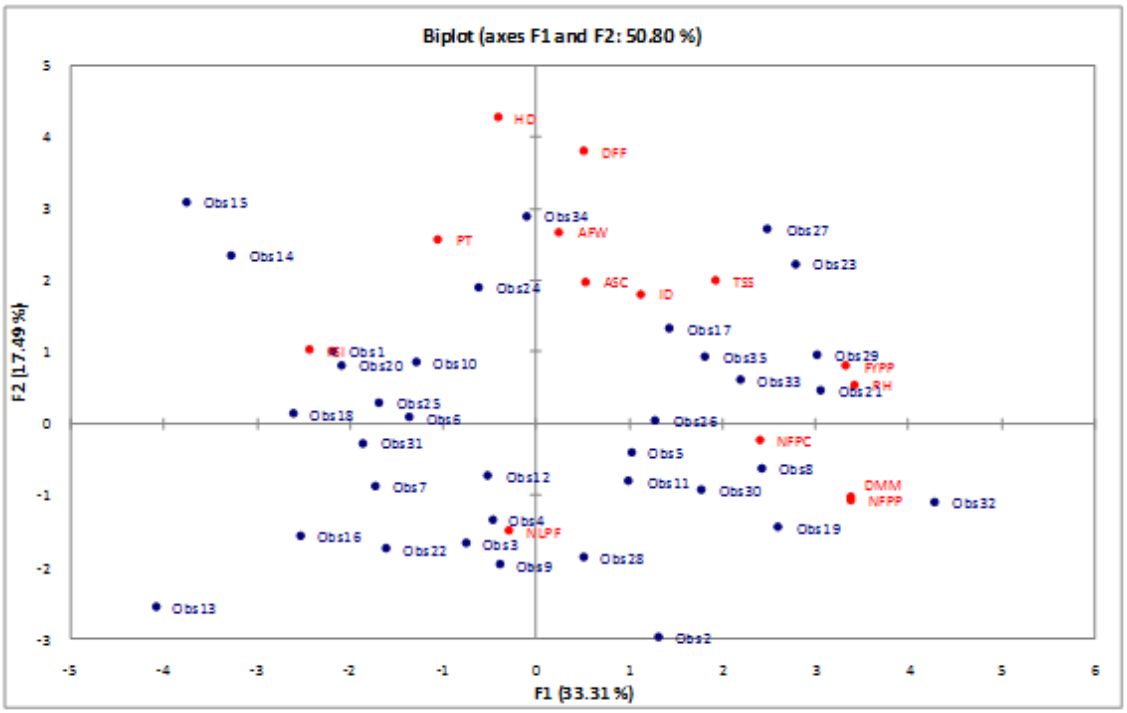
Characters	Clusters			
	I	II	III	IV
Days to 50% flowering	30.67	32.05	33.13	33.15
Number of fruits per cluster	4.61	5.87	4.14	5.30
Number of fruits per plant	16.09	35.71	13.51	35.83
Average fruit weight (g)	52.71	48.28	62.41	64.34
Fruit shape index	1.01	0.93	1.10	0.88
Number of locules per fruit	3.43	3.28	2.98	3.24
Pericarp thickness (mm)	5.17	5.72	5.35	6.16
Plant height (cm)	84.35	131.79	85.44	168.78
Harvest duration (days)	28.96	35.95	27.53	36.67
Internodal distance (cm)	9.67	9.55	9.64	10.92
Days to marketable maturity	68.56	70.43	74.67	71.78
Total soluble solids (^{o}Brix)	3.59	3.59	3.82	4.16
Ascorbic acid content (mg/100g)	18.50	19.91	24.02	23.14
Fruit yield per plant (kg)	0.82	1.63	0.82	2.18

Table.5 Principal component for 35 genotypes on 14 characters in tomato

	DFF	NFPC	NFPP	AFW	FSI	NLPF	PT	PH	HD	ID	DMM	TSS	ASC	FYPP	Eigen value	Variability (%)	Cumulative %
PC1	0.139	0.646	0.908	0.067	-0.659	-0.080	-0.284	0.923	0.911	0.305	-0.111	0.518	0.144	0.896	4.663	33.31	33.310
PC2	0.742	-0.046	-0.206	0.523	0.202	-0.293	0.504	0.108	-0.201	0.355	0.834	0.391	0.386	0.158	2.449	17.49	50.805
PC	0.485	-0.221	0.133	-0.486	-0.387	0.507	-0.489	0.013	-0.115	0.105	0.405	-0.266	0.307	-0.188	1.573	11.24	62.044
PC4	0.120	0.112	-0.140	0.413	-0.283	0.693	0.337	0.153	-0.156	0.180	-0.008	-0.333	-0.636	0.205	1.530	10.93	72.969

DFF-Days to 50% flowering, NFPC-Number of fruits per cluster, NFPP-Number of fruits per plant, AFW-Average fruit weight (g), FSI-Fruit shape index, NLPF-Number of locules per fruit, PT-Pericarp thickness (mm), PH-Plant height (cm), HD-Harvest duration (days), ID-Internodal distance (cm), DMM-Days to marketable maturity, TSS-Total soluble solids (^{o} Brix), ASC-Ascorbic acid content (mg/100g), FYPP-Fruit yield per plant (kg)

Fig.1 Bi-plot of tomato genotypes for first two principal components



Cluster II for the traits *viz.*, number of fruits per cluster, number of fruits per plant and internodal distance, cluster III for fruit shape index and ascorbic acid content. Cluster IV for average fruit weight, pericarp thickness, plant height, harvest duration, total soluble solids and fruit yield per plant. The genotypes having wide genetic base and desirable characteristics can be involved in intra-specific crosses which would lead to transmission of good genetic gain for various traits including yield. Earlier workers like Rai *et al.*, (1998), Mohanty and Prusti (2001), Mehta *et al.*, (2007), Shashikant *et al.*, (2010), Pathak and Kumar (2011), Narolia and Reddy (2012) and Reddy *et al.*, (2013) have also indicated the significance of genetic divergence in tomato.

Principal component analysis (PCA)

PCA reflects the importance of the largest contributor to the total variation at each axis of differentiation. The eigen values are often used to determine how many factors to retain. The sum of the eigen values is usually equal to the number of variables. Therefore, the present study revealed that out of 14 principal components (PCs), four *viz.*, PC-1, PC-II, PC-III and PC-IV had Eigen values >1 and contributed for 72.97% of total cumulative variability among different genotypes (Table 5). The contribution of PC-I towards variability was highest (33.31%) followed by PC-II, PC-III and PC-IV which contributed 17.49%, 11.24% and 10.93% variability respectively. The PC-I showed positive factor loadings for most of the traits except fruit shape index, number of locules per fruit, pericarp thickness and harvest duration while PC-II indicated positive factor loading for days to 50% flowering, average fruit weight, fruit shape index, pericarp thickness, plant height, internodal distance, harvest duration, total soluble solids, ascorbic acid content and fruit yield per plant. Traits which contributed

positive factor loadings towards PC-III were days to 50% flowering, number of fruits per plant, number of locules per fruit, plant height, internodal distance, harvest duration and ascorbic acid content. PC-IV indicated highest positive factor loading for number of locules per fruit followed by average fruit weight and pericarp thickness. It is evident that fruit yield per plant shows higher contribution to PC-I and chief contributors to PC-II. Number of locules per fruit contributed maximum share in PC-III and PC-IV. These results clearly indicated that PC (s) analysis in parallel to characterization of genetic resources also highlighted certain traits for exercising selection of interest for practical breeding purposes. Similar results were found in earlier article of Krasteva and Dimova (2007). In further support to our findings, Merk *et al.,* (2012) reported that first two PC (s) explained 28% and 16.2% of the variance and were heavily weighted by measures of fruit shape and size in tomato.

The first two principal components who contributed 50.80% towards total variance were plotted on PC-I x-axis and PC-II on y-axis to detect the association between different clusters (Fig. 1). It can be seen that fruit yield per plant was significantly positive correlated with plant height, number of fruits per cluster and harvest duration.

In conclusion, present genetic divergence studies grouped thirty five genotypes of tomato into four clusters.

The cluster I and IV were found most divergent, therefore genotypes from these clusters could be selected for hybridization to develop promising F_1 hybrids or transgressive segregants in succeeding generations. Principal component (PC) analysis depicted first four PC (s) with Eigen-value higher than 1 contributing 72.97% of total variability for different traits. The PC-I showed positive factor loadings for for most of the traits except fruit shape index, number of locules per fruit, pericarp thickness and harvest duration.

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