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Genetic divergence for qualitative traits of fruit yield and its components in custard apple (*Annona squamosa* L.). Indigenous accessions from northern Bastar of Chhattisgarh

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Abstract
The field experiment was conducted at Research cum Instructional Farm, College of Agriculture and Research Station, Singarbhat, Kanker, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.) during *Kharif* 2013-14. The experiment was laid out in Randomized Block Complete Design with four replication, *in-situ* sixty genotypes of about 10 year age were selected for investigation, which were found as wild seedling plants on forest land and rice bunds in Kanker District-(C.G.). The treatments comprised of sixty genotypes for with different qualitative character *viz* total soluble solid, total sugar, reducing sugar, non-reducing sugar, titration acetic acid, keeping days, pulp ratio, pulp seed ratio, peel percentage and fruit yield per plant. The D^2 analysis classifies the genotypes into relatively homogeneous groups in such a way that within cluster diversity is minimized and between clusters diversity is maximized. The respective genotypes from diverse clusters can be utilized in breeding programme. Cluster analysis for qualitative traits showed that all genotypes were grouped into 8 clusters using non-hierarchical Euclidean cluster analysis (Table 4.15 and Fig. 4.5). For qualitative parameter, maximum number of genotypes were grouped into cluster VIII (14) followed by cluster III (12). Intra and Inter-Cluster distances among 8 clusters for qualitative traits in custard apple are presented in Table 4.16. The highest intra-cluster distance was recorded for cluster VI (48.65) followed by cluster II (46.79) and the smallest intra cluster distance was recorded in cluster III (0.00) followed by cluster V (8.15) for qualitative traits. Custard apple is a predominantly cross-pollinated (protogyny) crop and no information is available suggesting a change in its breeding behavior under varied environmental conditions. Discrimination of desirable parents for a successful hybridization programme is a difficult task for a plant breeder. Generally, geographical diversity has been considered as a measure of genetic diversity. However, this is an inferential criterion and it may not be so effective in quantifying or differentiating among populations.

Keywords: D^2, genetic divergence, custard apple, indigenous accessions, Bastar and Chhattisgarh

Introduction
Custard apple (*Annona squamosa* L.), is an important dry-land fruit crop in India and belongs to family 'Annonaceae' having chromosome number 2n=14. It is also known as *Sitaphal* or *Sharifa* The fruit tree belongs to tropical climate and is native of tropical America and surrounding regions. *Annona* means year's harvest and *squamosa* means scaly referring to the scale like structure of the fruit surface. It is cultivated in Maharashtra, Gujarat, Madhya Pradesh, Andhra Pradesh, Chhattisgarh, Karnataka, Bihar, Orissa, Assam and Tamil Nadu. Besides India, it is common in China, Philippines, Egypt and Central Africa. In India, it is presently grown in an area of about 29.87 thousand hectares with a production of 228.37 metric tones and the average productivity is 765 q/ha and it ranges from 673 q/ha in Andhra Pradesh to 685 q/ha in Maharashtra (2014-15). Chhattisgarh and Maharashtra occupies 55.74 per cent of the total area in the country. Whereas Gujarat covered 5.34 thousand hectare and the average productivity is 768 q/ha (Chandra, 2010) [2]. Chhattisgarh, state of India occupies an area of approximately 7.99 thousand hectare with an annual production of 39.73 metric tones having the productivity of 497.25 q/ha under custard apple. In the range of forest scattered across Jagdalpur, Beejapur, Dantewada, Kanker, Dhamtari, Rajnandgaon, Durg,

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Jashpur, Surguja and Bilaspur districts, only Kanker district is blessed with natural biodiversity of the custard apple. Its wild land races are found distributed all along as a natural stand over an area of about 7.20 thousand hectare with an annual production of 35.60 metric tones having the productivity of 494.45 q/ha (Anonymous, 2013) [1]. The edible portion or pulp is creamy or custard like, granular, with a good blend of sweetness, possessing pleasant flavor and mild aroma have a universal liking, being rich in carbohydrates 23.0 g per 100 g fruits. The fruit is reported to have moisture 70.5 g, protein 1.6 g, fat 0.4 g, minerals 0.9 g, fiber 3.1 g, calcium 17.0 mg, phosphorous 47.1 mg, iron 1.5 mg, thiamine 0.07 mg, riboflavin 0.17 mg, niacin 1.30 mg, Vitamin C 37.0 mg and energy 104 Kcal Gopalan et al., (1987) [4]. The evolution of custard apple through natural and human se lection in diverse elevation zones and under different cropping systems with involvement of honey bees being the carrier of cross pollination has resulted in a wide variety of locally adapted land races. These land races have evolved over years to fit into local cropping patterns and diverse end uses and represent a wide range of patterns of crop diversity. The knowledge of patterns of genetic variation of a crop species in any given region or country is very important for planning future germplasm exploration missions and thereafter it's efficient utilization in crop improvement programme. Assessment of genetic variability for yield and its components is useful to predict the extent of improvement possible for fruits yield and other important characters.

Materials and Methods

An experiment comprised of 60 genotype accessions (Table-1) was conducted at 10 years old *In-situ* plants at Northern Bastar, Research cum Instructional Farm College of Agriculture & Research Station, Singarbhat, Kanker, Indira Gandhi Krishi Vishwavidyalaya, Raipur (Chhattisgarh) during *Kharif* 2013-14. The experiment was laid out in Randomized Complete Block Design (RCBD) with four replications recommended package of practices were applied to raise the normal crop. Observations were recorded on one randomly selected competitive plants from each genotype, in each replication on 10 qualitative characters *viz.* total soluble solid, total sugar, reducing sugar, non-reducing sugar, titration acetic acid, keeping days, pulp ratio (%), pulp seed ratio, peel percentage and fruit yield per plant (kg). The statistical analysis for D^2 statistic originally developed by Mahalanobis (1928) [8] and further elaborated by Rao (1952) [10] was applied for assessment of genetic divergence between the populations. The D^2 values between any two populations were estimated from the sample on the basis of characters.

Results and Discussion
Genetic divergence

In plant breeding, genetic diversity plays an important role because hybrids between lines of diverse origin, generally display a greater heterosis than those between closely related parents. Genetic diversity arises due to geographical separation or due to genetic barriers to cross-ability. The choice of genetically diverse parents for hybridization is an important feature of crop improvement programme for getting desirable segregates. The multivariate analysis based on Mahalanobis D^2, Tocher Method Euclidean cluster analysis is used for divergence analysis. Multivariate analysis by means of Mahalanobis D^2 statistic is a powerful tool in quantifying the degree of divergence between biological populations at genotypic level to assess the relative contribution of different components to the total divergence. The D^2 analysis classifies the genotypes into relatively homogeneous groups in such a way that within cluster diversity is minimized and between clusters diversity is maximized. The respective genotypes from diverse clusters can be utilized in breeding programme.

Cluster constellation

Cluster analysis for qualitative traits showed that all genotypes were grouped into 8 clusters using non-hierarchical Euclidean cluster analysis (Table-2). For qualitative parameter, maximum number of genotypes were grouped into cluster VIII (14) followed by cluster III (12). The minimum number of genotypes was found in cluster V (3) followed by cluster I(4). Grouping pattern of genotype indicated that genotypes within each cluster were similar type plants than the genotypes in other cluster. Clustering pattern of genotypes recognized that there is no influence of ecological location on divergence because parental populations were collected from different ecological area.

Intra and Inter-Cluster distances for qualitative traits

Intra and Inter-Cluster distances among 8 clusters for qualitative traits in custard apple are presented in Table-3. The highest intra-cluster distance was recorded for cluster VI (48.65) followed by cluster II (46.79) and the smallest intra cluster distance was recorded in cluster III (0.00) followed by cluster V (8.15) for qualitative traits. These results revealed that genotypes included in largest intra- cluster distances were relatively more diverse than those of smallest intra- cluster distance, it means further screening will be required to select superior genotypes because of similar type less diverse genotypes.

The highest inter-cluster distance was recorded between cluster V and cluster III (4166.05) followed by cluster VI and III (4091.02), whereas, lowest inter-cluster distance was recorded between cluster IV and cluster V (52.47) followed by cluster VII and cluster VIII (62.20) for qualitative traits. The highest inter- cluster distance revealed the presence of genetic wide diversity in between groups leads to selection of such lines for hybridization and production of elite type progenies. The least inter-cluster distance revealed close relationship between the genotypes and hence, may not be emphasized upon to be used in hybridization programme. Narrow genetic base does not give rise to produce vigorous progeny.

Percent contribution to variation or mean performance of genotypes for qualitative traits

Cluster wise mean performance of genotypes for qualitative traits in custard apple is presented here in table-4. The information revealed that cluster I didn't show highest cluster mean for any trait, but it showed lowest cluster mean for total soluble solid, total sugar, reducing sugar, pulp ratio and pulp-seed ratio. Cluster II showed highest mean value for acidity, whereas, it showed lowest mean value for non-reducing sugar. Cluster III showed highest mean performance for pulp ratio and peel percentage. Cluster IV showed the maximum cluster mean for non-reducing sugar, whereas, it showed lowest cluster mean for keeping days and fruit yield per plant. Cluster V showed maximum cluster mean for peel percentage. Cluster VI and cluster VII did not show highest and lowest mean value for any of the trait. Cluster VIII showed highest mean value for total soluble solid, total sugar, reducing sugar, keeping days, pulp-seed ratio, peel percentage, fruit yield per plant, whereas, it showed lowest mean value for acidity.

Genotypes of cluster VIII showed highest mean value for most of the qualitative traits, so, it can be concluded that genotypes of cluster VIII will give good opportunity to researcher to be used as parents for developing good variety or hybrids of superior fruit quality, whereas, genotypes of cluster III could be used for increasing pulp ratio and peel percentage.

Percent contribution to variation is presented in table-4. Data on percent contribution to genetic divergence showed a high percentage of variation for non-reducing sugar (51.30), pulp-seed ratio (35.88), total sugar (5.03), acidity (3.56), total soluble solids (1.47), keeping days (shelf life) (1.36) and fruit yield per plant (0.90)whereas, rest of the characters had very less percentage contribution to variation.

Cluster wise promising genotypes (qualitative traits)
Cluster wise promising genotypes for qualitative traits in custard apple are presented in Table-5. Results showed that in cluster I, genotype IGCA-21 is promising for most of the qualitative traits, therefore, it can be selected as parent for further research programme to improve fruit quality traits. Genotypes IGCA-21, IGCA-11, IGCA-4, IGCA-2, IGCA-3, IGCA-12, IGCA-13 and IGCA-10 are most promising for pulp-seed ratio and peel percentage from cluster I, II, III, IV, V, VI, VII and cluster VIII respectively. Varieties from geographically diverse localities are generally included in hybridization programme presuming genetic diversity. The

problem of selection may further be simplified if one could identify the characters responsible for discrimination between parents. The reports available on this aspect in custard apple are rather scanty. Therefore, the present investigation was aimed at ascertaining the nature and magnitude of genetic diversity among a set of custard apple genotypes.

Genetic divergence analysis by using cluster statistics is a powerful tool in quantifying the degree of divergence between biological populations and to assess the relative contribution of different components to the total divergence. The genetic architecture of a population is controlled by the breeding behavior of the individuals. Changes in breeding systems have accelerated the genetic divergence in natural population. Custard apple is a predominantly cross-pollinated (protogyny) crop and no information is available suggesting a change in its breeding behavior under varied environmental conditions. Discrimination of desirable parents for a successful hybridization programme is a difficult task for a plant breeder. Generally, geographical diversity has been considered as a measure of genetic diversity. However, this is an inferential criterion and it may not be so effective in quantifying or differentiating among populations. These results are in general agreement with the findings of Rao (1974) [11], George *et al.* (2005) [3], Le *et al.* (1998), Liu ShiBiao (2000) [7], Samal *et al.* (2002) [12], Liu and Chen (2002) [6] and Mariguele and Silva (2010b) [9].

Table 1: The list of experimental material used for correlation and path analysis in custard apple

S. No.	Name of Genotypes	S. No.	Name of Genotypes
1.	IGCA-1	31.	IGCA-31
2.	IGCA-2	32.	IGCA-32
3.	IGCA-3	33.	IGCA-33
4.	IGCA-4	34.	IGCA-34
5.	IGCA-5	35.	IGCA-35
6.	IGCA-6	36.	IGCA-36
7.	IGCA-7	37.	IGCA-37
8.	IGCA-8	38.	IGCA-38
9.	IGCA-9	39.	IGCA-39
10.	IGCA-10	40.	IGCA-40
11.	IGCA-11	41.	IGCA-41
12.	IGCA-12	42.	IGCA-42
13.	IGCA-13	43.	IGCA-43
14.	IGCA-14	44.	IGCA-44
15.	IGCA-15	45.	IGCA-45
16.	IGCA-16	46.	IGCA-46
17.	IGCA-17	47.	IGCA-47
18.	IGCA-18	48.	IGCA-48
19.	IGCA-19	49.	IGCA-49
20.	IGCA-20	50.	IGCA-50
21.	IGCA-21	51.	IGCA-51
22.	IGCA-22	52.	IGCA-52
23.	IGCA-23	53.	IGCA-53
24.	IGCA-24	54.	IGCA-54
25.	IGCA-25	55.	IGCA-55
26.	IGCA-26	56.	IGCA-56
27.	IGCA-27	57.	IGCA-57
28.	IGCA-28	58.	IGCA-58
29.	IGCA-29	59.	IGCA-59
30.	IGCA-30	60.	IGCA-60

Table 2: Distribution of different clusters for qualitative traits of fruit yield and its components in custard apple during the year 2013-14 and 2014-15

Cluster number	Number of genotypes	Genotypes
I	4	IGCA-5, IGCA-6, IGCA-7, IGCA-9
II	8	IGCA-8, IGCA-14, IGCA-15, IGCA-18, IGCA-26, IGCA-28, IGCA-31, IGCA-36
III	12	IGCA-1, IGCA-2, IGCA-3, IGCA-4, IGCA-10, IGCA-11, IGCA-12, IGCA-13, IGCA-19, IGCA-20, IGCA-21, IGCA-22,
IV	5	IGCA-17, IGCA-34, IGCA-35, IGCA-38, IGCA-39
V	3	IGCA-25, IGCA-32, IGCA-33
VI	6	IGCA-42, IGCA-45, IGCA-46, IGCA-51, IGCA-52, IGCA-54
VII	8	IGCA-24, IGCA-29, IGCA-43, IGCA-44, IGCA-47, IGCA-49, IGCA-55, IGCA-56
VIII	14	IGCA-16, IGCA-23, IGCA-27, IGCA-30, IGCA-37, IGCA-40, IGCA-41, IGCA-48, IGCA-50, IGCA-53, IGCA-57, IGCA-58, IGCA-59, IGCA-60

Table 3: Intra and Inter-Cluster distances and D^2 values among 8 clusters for qualitative traits of fruit yield and its components in custard apple during the year 2013-14 and 2014-15

Cluster Number	I	II	III	IV	V	VI	VII	VIII
I	33.84	123.75	721.50	1327.86	1602.26	1683.85	1359.08	1399.72
II		46.79	794.89	1199.96	1445.68	1369.65	1394.31	1553.06
III			0.00	3742.72	4166.05	4091.02	3902.15	3976.95
IV				39.53	52.47	142.09	111.11	260.65
V					8.15	109.19	126.14	300.53
VI						48.65	379.16	656.61
VII							20.75	62.20
VIII								19.08

Bold figures represent intra-cluster distance

Table 4: Cluster wise means performance of genotypes for qualitative traits of fruit yield and its components in custard apple during the year 2013-14 and 2014-15

Cluster numbers	Total Soluble Solids (° Brix)	Total Sugar (%)	Reducing Sugar (%)	Non-reducing Sugar (%)	Acidity (%)	Keeping Days	Pulp Ratio	Pulp- Seed Ratio	Peel Percentage	Fruit Yield per Plant (kg)
I	21.27✱✱	16.61✱✱	12.88✱✱	4.91	0.40	3.34	46.23✱✱	2.43✱✱	31.37✱✱	104.61
II	21.58	17.34	13.75	3.80✱✱	0.42✱	3.59	47.98	2.54	33.45	104.24
III	25.65	23.00	18.81	4.19	0.31	5.15	53.04✱	8.37	36.00✱	148.79
IV	21.69	17.30	13.60	6.04✱	0.40	3.25✱✱	46.29	2.55	31.59	103.66✱✱
V	25.50	23.15	17.67	5.47	0.32	4.25	53.00	8.09	36.00✱	145.50
VI	21.91	20.74	16.23	5.00	0.35	3.50	49.69	3.61	34.00	104.50
VII	21.56	20.15	15.20	4.95	0.34	5.00	48.00	2.69	34.00	146.75
VIII	28.07✱	23.45✱	19.30✱	4.15	0.22✱✱	6.50✱	53.00	12.65✱	36.00✱	152.25✱
% Contribution towards genetic divergence	1.47	5.03	0.11	51.30	3.56	1.36	0.28	35.88	0.11	0.90

✱ Highest values**;** ✱✱ Lowest values

Table 5: Cluster wise promising genotypes of genotypes for qualitative traits of fruit yield and its components in custard apple during the year 2013-14 and 2014-15

S. No.	Cluster Number	I	II	III	IV	V	VI	VII	VIII
1.	Total Soluble Solids (°Brix)	IGCA-21	IGCA-22	IGCA-10	IGCA-2	IGCA-13	IGCA-20	IGCA-12	IGCA-3
2.	Total Sugar (%)	IGCA-2	IGCA-21	IGCA-3	IGCA-22	IGCA-1	IGCA-20	IGCA-12	IGCA-11
3.	Reducing Sugar (%)	IGCA-20	IGCA-3	IGCA-2	IGCA-1	IGCA-21	IGCA-22	IGCA-4	IGCA-19
4.	Non-Reducing Sugar (%)	IGCA-7	IGCA-6	IGCA-16	IGCA-23	IGCA-12	IGCA-9	IGCA-5	IGCA-2
5.	Acidity (%)	IGCA-21	IGCA-22	IGCA-11	IGCA-20	IGCA-10	IGCA-1	IGCA-3	IGCA-12
6.	Keeping Days (Shelf life)	IGCA-21	IGCA-22	IGCA-4	IGCA-1	IGCA-11	IGCA-10	IGCA-9	IGCA-3
7.	Pulp Ratio	IGCA-21	IGCA-20	IGCA-22	IGCA-1	IGCA-2	IGCA-3	IGCA-4	IGCA-5
8.	Pulp -Seed Ratio	IGCA-21	IGCA-11	IGCA-4	IGCA-2	IGCA-3	IGCA-12	IGCA-13	IGCA-10
9.	Peel Percentage	IGCA-21	IGCA-11	IGCA-4	IGCA-2	IGCA-3	IGCA-12	IGCA-13	IGCA-10
10.	Fruit Yield / Plant (kg)	IGCA-21	IGCA-3	IGCA-11	IGCA-1	IGCA-4	IGCA-2	IGCA-10	IGCA-22

Conclusions

Genetic divergence analysis by using cluster statistics is a powerful tool in quantifying the degree of divergence between biological populations and to assess the relative contribution of different components to the total divergence. The genetic architecture of a population is controlled by the breeding behaviour of the individuals. Changes in breeding systems have accelerated the genetic divergence in natural population. Custard apple is a predominantly protogynous crop and no information is available suggesting a change in its breeding behaviour under varied environmental conditions. Discrimination of desirable parents for a successful hybridization programme is a difficult task for a plant breeder. Generally, geographical diversity has been considered as a measure of genetic diversity. However, this is an inferential criterion and it may not be so effective in quantifying or differentiating between populations.

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