

Genetic Divergence of Sugarcane under Waterlogging Conditions

Bal Krishna^{1*}, Dharam Nath Kamat¹, Juli Kumari² and Divya Prakash¹

¹Department of Plant Breeding and Genetics, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar 848 125, India

²Department of Botany, Tilka Manjhi Bhagalpur University, Tatarpur, Bhagalpur, Bihar 812 007, India

*Corresponding Author E-mail: bk151219@gmail.com

Received: 17.06.2017 | Revised: 29.07.2017 | Accepted: 4.08.2017

ABSTRACT

Genetic divergence for twenty-five traits were studied in sixteen phenotypically diverse midlate maturing sugarcane genotypes, which were planted in RBD in three replications at D. R.P.C.A.U. Research Farm, Pusa, Bihar during spring season 2016-2017. The study indicated the presence of sufficient amount of diversity among the genotypes under study. The clustering of 16 clones into five clusters for morphological and quality characters revealed divergence between the genotypes. Five clusters were formed on grouping of clones based on Mahalanobis D^2 distances. The result indicated that, higher estimate of inter cluster distance between cluster III and IV indicated wide genetic diversity between these two groups. Thus, genotypes with high index for specific characters that fall into different clusters could be intercrossed to have maximum hybrid vigor and good number of useful segregants while lower inter cluster distance was noticed between cluster IV and V. This might indicate the close relationship and likelihood between genotype groups within these clusters. The results also indicated that cluster III, IV and V involved highly diverse genotypes, can be chosen for recombination breeding and isolating desirable genotypes combinations. Cluster III (CoP11440) was found superior for quality characters. Among five clusters, cluster II was the largest with nine genotypes followed by cluster I with four genotypes. The study also revealed that, the cluster II (BO155, CoP2061, CoP14439, CoP15439, BO154, CoP12438, CoP09437, CoP14438 and CoP15441) performed superior for sugar yield and cane yield under waterlogging condition. Hence these genotypes can be selected and advanced through further breeding programme.

Key words: Sugarcane, Waterlogging, Genetic divergence, Mahalanobis's D^2 statistics, Cluster distance

INTRODUCTION

Sugarcane is an important agro-industrial crop of India. It is grown in 5.30 million hectare with total production of 366.8 million tonnes and productivity of 69.1 tonnes/ha. whereas in Bihar it is grown in an area of 3.02 lakh hectare with production of 14.90 lakh tonnes

and productivity 50 tonnes/ha. (2014-15, Indian sugar February, 2016). The cultivated varieties of sugarcane are interspecific hybrids involving at least three species, *S. officinarum*, *S. barberi* and *S. spontaneum* which themselves represent complex polyploidy.

Cite this article: Krishna, B., Kamat, D.N., Kumari, J. and Prakash, D., Genetic Divergence of Sugarcane under Waterlogging Conditions, *Int. J. Pure App. Biosci.* 6(1): 210-218 (2018). doi: <http://dx.doi.org/10.18782/2320-7051.4085>

The chromosome number among varieties varies from $2n = 100$ to 120. It is for this reason the sugarcane varieties are botanically described as *Saccharum spp.* complex hybrid. The heterozygous and polyploid nature of this crop has resulted in generation of greater genetic diversity. A basic understanding of the genetic diversity that exists in the germplasm available for breeding is fundamental to the success of a breeding programme. This knowledge can be useful in the utilization and management of genotypes and indeed genes in the breeding gene pool. In heterosis breeding programme the diversity of parents is always emphasized. More diverse the parent within a reasonable range, better the chances of improving economic characters under consideration in the resulting offspring. In sugarcane, for example, crosses could be planned between genotypes from divergent backgrounds to maximize heterosis while increasing genetic diversity in the gene pool.

The success of a sugarcane breeding programme lies in the proper choice of rich and genetically diverse parents. The accurate quantification of the genetic diversity of major agricultural crops is important both scientifically and socio-economically. Concern has often been expressed that the practice of modern intensive plant breeding leads inevitably to a reduction in both diverse agricultural practices and genetic diversity of crops. One way of averting this impending problem is to broaden the genetic base of the breeding programme. In general, inclusion of diverse parents in hybridization programme will improve the chances of desirable segregants for yield and other characters in the progeny. The search of genetically diverse parents can be based on geographical origin, morphological traits, and pedigree data or molecular markers data.

So, present study was undertaken to assess the extent of genetic divergence of some important traits of midlate sugarcane clones under subtropical India for waterlogging condition.

MATERIALS AND METHODS

Study area

The experiment was conducted with sixteen genotypes received from Sugarcane Research Institute, D.R.P.C.A.U. Pusa, Samastipur, Bihar during *spring season* of 2016-2017. The experimental plot is situated between 25.97° N latitude and 85.66° E longitudes at 51.80 m above mean sea level.

Treatments and experimental design

The sixteen sugarcane clones viz. BO91, BO154, CoP 09437, CoP 11439, CoP 11440, CoP 12438, CoP 12439, CoP 13438, CoP 13439, CoP 14438, CoP 14439, CoP 15439, CoP 15440, CoP 15441, BO 155 and CoP 2061 were evaluated. The trial was laid out in randomized block design with three replications. All the experimental material introduced from SRI, Pusa, Bihar and planted in spring season 2016-2017 under waterlogging condition. Equal number of three budded set of each clones were planted.

Data collected

Data were collected for yield attributing traits viz. germination percentage at 45 DAP (Days After Planting), number of shoots at 120 DAP. (000/ha), plant height at 150, 240, 360 days (cm), number of fully emerged leaves at 30 days and 60 days after waterlogging, leaf area index before waterlogging, at 30 and 60 days after waterlogging, number of nodes with aerial roots, cane diameter at harvest (cm), number of shoots at 240 DAP (000/ha), number of millable canes at harvest (000/ha), single cane weight (Kg), cane yield (t/ ha) and sugar yield (CCS t/ha) at harvest and juice quality traits viz. brix, Pol and Purity at 10 & 12 months stage (%), CCS % at 10 and 12 months stage.

Mahalanobis D^2 statistic⁸

In the present study, Mahalanobis D^2 statistic was employed to know the clustering pattern by using dendrogram and to compare the cluster pattern based on canonical variate analysis.

The square of the Mahalanobis' generalized distance between any two populations is given by the formula:

$$\Delta^2 = \sum \delta_i \delta_j \lambda_{ji}$$

Where,

Δ^2 = square of the generalized distance

λ_{ji} = reciprocal of the common dispersion matrix ij

$\partial_i = (\mu_{i1} - \mu_{i2})$

$\partial_j = (\mu_{j1} - \mu_{j2})$

Where, μ = Vector of mean values for all the characters.

The formula for the estimation of distance for sample is

$D2P = d1 S^{-1} d$

Where,

$D2P$ = Square of the distance considering P values

d = Vector of observed distances of mean values of all the characters = $(x_{i1} - x_{i2})$

x_i = Vector of the mean values of all the characters

S^{-1} = Inverse of variance and co-variance matrix.

Since inverting matrix is complicated, the original corrected variables (X_i) were transformed to non-corrected variable (λ). So the computation of D^2 values reduced to simple summations of the squares of the difference between the values of the two populations.

The transformations were done by pivotal condensation method. These newly transformed uncorrected variables were used to calculate the square of the distance using the formula.

$D2 = (\gamma_{i1} - \gamma_{i2})^2$

Where, γ = Vector of transformed mean values.

Clustering of genotypes

Clustering was done by adopting dendrogram technique described by Tocher's¹² (Rao, 1952) method utilizing the $D2$ values and the dissimilarity coefficient values.

Intra and inter cluster distance

The intra and inter cluster distances were calculated by adopting the formula given by Panse and Sukhatme⁹.

Contribution of each character towards genetic divergence

The character contribution towards genetic divergence was computed using the method given by Singh and Choudhary¹⁴. In all the combinations, each character was ranked on the basis of $d_i = y_{ij} - y_{ik}$ values.

Where,

d_i = mean deviation

y_{ij} = mean value of the jth genotype for the ith character and

y_{ik} = mean value of the kth genotype for the ith character

Rank 'I' is given to the highest mean difference and rank 'P' is given to the lowest mean difference

Where,

P is the total number of characters.

Finally, the number of times that each character appeared in the first rank was computed and per cent contribution of characters towards divergence was estimated using the formula

Percentage contribution of character X = $(N \times 100) / M$

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

M = All possible combinations of number of genotypes considered.

RESULT AND DISCUSSION

Clustering pattern

Based on the D^2 values sixteen sugarcane genotypes involved under the study were grouped into five clusters (Table:1 and fig: 1) using Tocher's method given by Rao¹² indicating the presence of sufficient amount of diversity among the genotypes under study. Of the five clusters, cluster II was largest comprising nine genotypes followed by cluster I with four genotypes, while cluster III, IV and V were solitary type. The formation of solitary clusters may be due to isolation preventing the gene flow or intensive natural or human selection for diverse adaptive complexes. These accessions may be unique and useful in breeding.

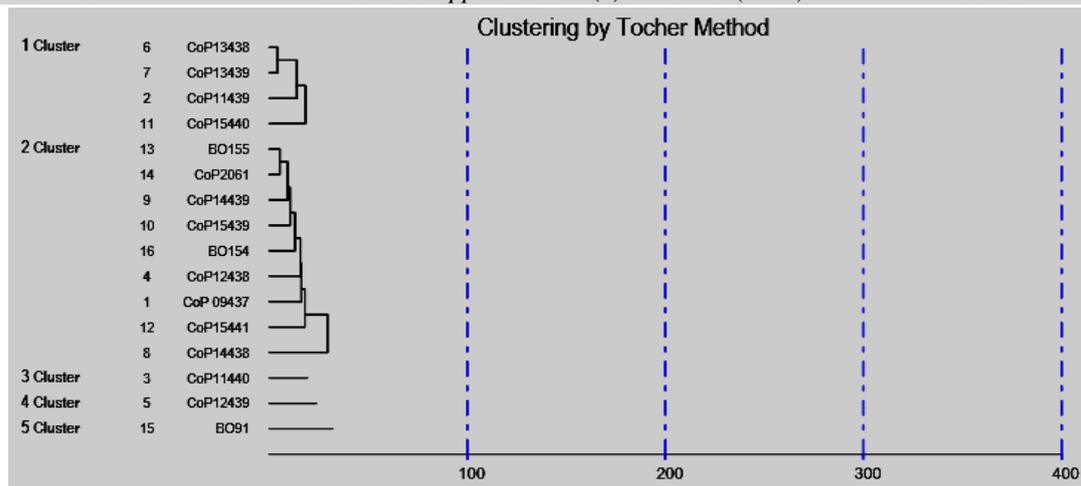


Fig. 1: Dendrogram of 16 different sugarcane genotypes based upon mean of 25 yield contributing and quality characters

In general, the clustering pattern revealed the existing pedigree relationships between the genotypes. The genotypes with common parentage clustered together as in the case of genotypes, CoP12438 (BO91 × Co1158), CoP09437 and CoP14438. However, this kind of relationship was not always true as the two genotypes evolved from crosses involving different parentages were clustered together. For instance, the genotypes CoP13438 (Lg 9505 PC), CoP13439 (Co62232 GC) and CoP11439 (Co88039 GC) are grouped together into cluster I though they are derived from different parents. One of the most important problems in sugarcane breeding is the narrow genetic base of the parents, that is, the high frequency of common ancestors in their genealogy which has led to a high degree of progeny inbreeding and reduces variability¹⁰. However, it is noticed that the clustering pattern of genotypes in the present study indicated its poor correspondence with the pedigree of the sugarcane genotypes as the cluster analysis grouped the genotypes with greater similarity for agronomic characters but not with its genealogical similarity. Hence they did not necessarily include the genotypes derived from common pedigree in to single cluster. This poor correspondence could be attributed to heterozygosity at the parent selection of hybrids under different agro-climatic conditions and the selection pressure applied on some characters to evolve these

varieties. This finding has applied value in that use of genotypes derived from same parental combination as specific combiners need not always lead to close breeding due to existence of genetic diversity between them.

Clustering pattern of genotypes of the same habitat into different clusters could be due to different genetic nature of the parents from which they were derived or due to the selection pressure applied on some characters to evolve these.

Average inter and intra cluster distances

Inter cluster D^2 values (Table 3 and fig: 2) ranging from 85.15 to 21.439 suggest moderate genetic divergence among the genotypes under investigation. Higher estimate of inter cluster distance between cluster III and IV indicated wide genetic diversity between these two groups. Thus, genotypes with high index for specific characters that fall into different clusters could be intercrossed to have maximum hybrid vigor and good number of useful segregants. Lower inter cluster distance was noticed between cluster IV and V. This might indicate the close relationship and likelihood between genotype groups within these clusters. Similar observations have been reported by earlier workers like Hooda *et al*⁷., Bhakshiram and Hemaprabha^{2,3}, Srivastava *et al*¹⁵., Ahmed *et al*¹., and Sanghera *et al*¹³.

Intra cluster D^2 values of cluster I was 52.93 and cluster II was 56.17 indicating that genotypes in cluster I and cluster II exhibited

relatively least differences among themselves. Same trends were reported by Bhakshiram and Hemaprabha², and Hemaprabha and Bhakshiram⁶. Present investigation revealed that the solitary cluster III, cluster IV and

cluster V were the most diverse cluster as many clusters showed highest inter cluster distance from these. It suggests that selected genotypes of these two clusters have potential as parents in further breeding programmes.

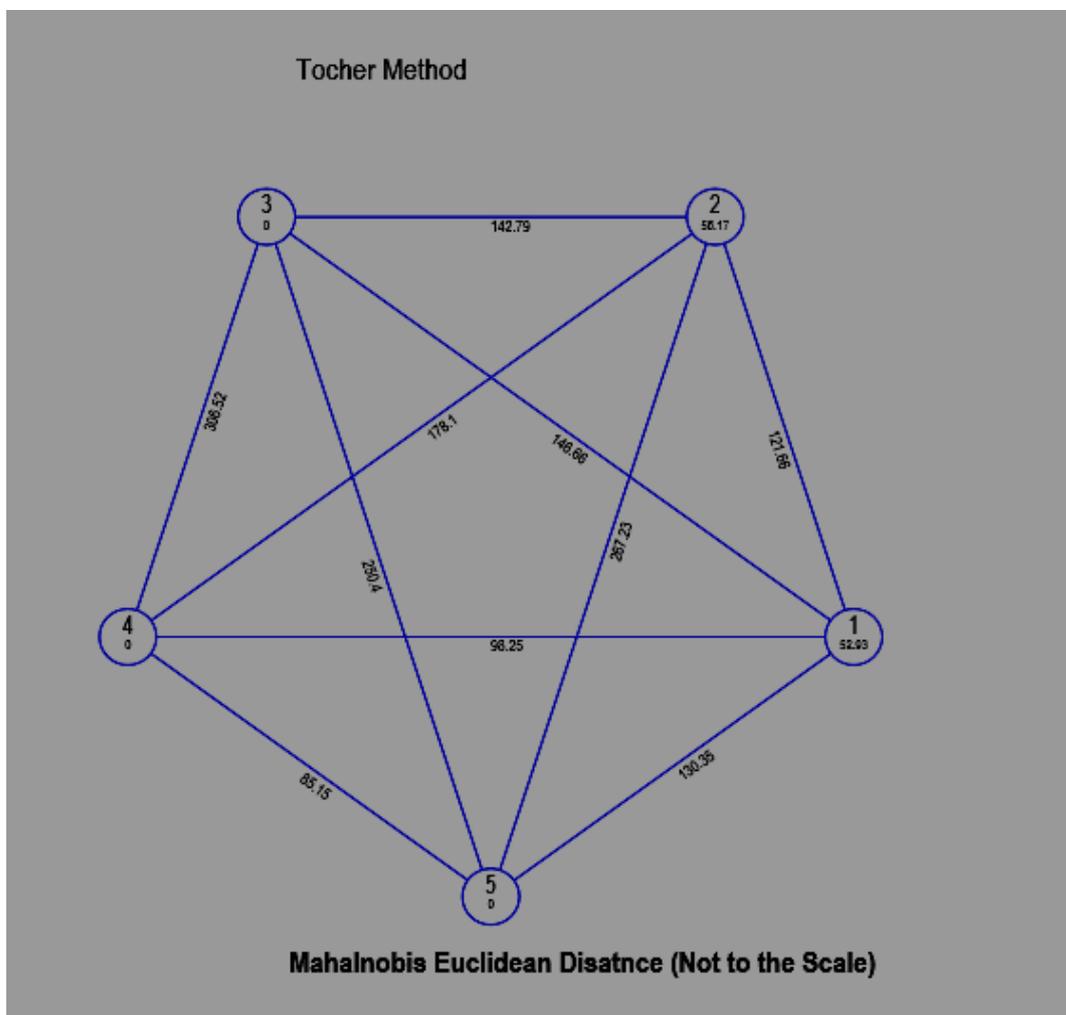


Fig. 2: Cluster analysis for 16 different genotypes

Contribution of characters to the divergence

Average inter and intra cluster D^2 values are furnished in Table: 3. Among the twenty-five characters studied, the most important character contributing to the divergence includes CCS % at 12 months' stage followed by leaf area index at 60 days after waterlogging, leaf area index at 30 days after waterlogging, plants height at 150 DAP (day after planting), number of fully emerged leaves at 60 days after waterlogging and leaf area index before waterlogging. These results

were in accordance with the findings of earlier workers for single cane weight, cane yield and quality attributes like Brix per cent, pol per cent and purity per cent¹¹, for cane yield and purity per cent¹⁵ and Sanghera *et al*¹³.

Analysis of cluster means

Mean values (Table 4) obtained from varying number of genotypes in each cluster although cannot be compared statistically to get relative idea of diversity among the clusters, it helps to group the clusters according to their average performance for a particular character. A critical study of cluster means for different

traits indicated that cluster II registered high cane yield (t/ha) as well as sugar yield (CCS t/ha), number of nodes with aerial roots, single cane weight (kg), cane diameter at harvest (cm), leaf area index before waterlogging, leaf area index at 30 days after waterlogging, number of fully emerged leaves at 30 and 60 days after waterlogging, while the cluster III was considered as the one with high quality characters like brix %, purity %, pol % at 10 and 12 months stage respectively^{5,17}. Cluster IV comprising the CoP 12439 genotype categorized as a cluster with high number of shoots at 240 DAP, number of millable cane at harvest (000/ha), leaf area index at 60 days after waterlogging, plant height at 150, 240, 360 DAP respectively. The cluster V were found high germination percentage at 45 DAP and number of shoots at 120 DAP (000/ha).

In general it could be inferred from the present investigation that, clusters II, III, IV and V were superior for important characters viz., cane yield, sugar yield and quality attributes and genotypes included in these diverse clusters hold good as parents for obtaining potential hybrids through creating large variability for desirable characters and

agrees with earlier inferences by Gill and Tripathi⁵, Hooda *et al*⁷, Hemaprabha and Bhakshiram⁶, Bhakshiram and Hemaprabha⁴, Srivastava *et al*¹⁵, Ahmed *et al*¹, Sanghera *et al*¹³, and Tahir *et al*¹⁶.

The most significant point that emerged from the present divergence investigations is that the clusters III and IV not only registered highest inter cluster distance between them but also moderate and farthest inter cluster distance values with all other clusters. The genotypes of these two clusters and also the genotypes of other superior clusters like II and V could be beneficially employed in future breeding programmes to improve both cane and sugar yield.

Hence, it is worth to note that in calculating cluster means, the superiority of a particular genotype with respect to a given character gets diluted by other genotypes that are related and grouped in the same cluster. Hence, apart from selecting genotypes from the clusters which have high inter-cluster distance for hybridization, one can also think of selecting parents based on extent of genetic divergence with respect to a particular character of interest.

Table 1: Distribution of sugarcane genotype to different cluster by Tocher Method

Clustar no.	Genotypes
I	CoP13438, CoP13439, CoP11439, CoP15440
II	BO155, CoP2061, CoP14439, CoP15439, BO154, CoP12438, CoP09437, CoP15441, CoP14438
III	CoP11440
IV	CoP12439
V	BO91

Table 3: Contribution of each character to the divergence in sugarcane genotypes

Sl. No	Characters	No. Of Ranked first	Per cent contribution
1	Germination percentage at 45 DAP	0.01	0.00
2	Number of shoots at 120 DAP (000/ha)	0.01	0.00
3	Number of Shoot at 240 DAP	0.01	0.00
4	Number of millable cane at harvest (000/ha)	0.01	0.00
5	Sugar yield (CCS t/ha).	0.01	0.00
6	Cane yield (t/ha).	0.01	0.00
7	Brix % at 12 months stage.	0.01	0.00
8	Purity % 10 months stage	0.01	0.00
9	Purity % at 12 months stage.	0.01	0.00
10	Pol % at 10 months stage.	1.67	1.67
11	Pol % at 12 months stage.	0.01	0.00
12	Brix % at 10 months stage.	0.83	0.83
13	Number of nodes with aerial roots	0.83	0.83
14	Plants height at 150 DAP (cm)	11.67	11.67
15	Plant height at 240 DAP (cm)	0.83	0.83
16	Plants height at 360 DAP(cm)	0.01	0.00
17	Single cane weight (kg)	1.67	1.67
18	Cane diameter at harvest (cm)	0.01	0.00
19	Leaf area index before water logging	5.00	5.00
20	Leaf area index at 30 days after waterlogging	13.33	13.33
21	Leaf area index at 60 days after waterlogging	24.17	24.17
22	No. of fully emerged leaves at 30 days after waterlogged	2.50	2.50
23	No. of fully emerged leaves at 60 days after waterlogged	10.00	10.00
24	CCS % at 10 months stage.	0.83	0.83
25	CCS % at 12 months stage.	26.67	26.67

Table 2: Average intra and inter Cluster values for 5 cluster formed due to 16 genotypes

Cluster	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
Cluster I	52.93				
Cluster II	121.66	56.17			
Cluster III	146.66	142.79	0.00		
Cluster IV	98.25	178.10	306.52	0.00	
Cluster V	130.35	267.23	250.40	85.15	0.00

Table 4: The mean value of 25 characters for 5 clusters formed by 16 sugarcane genotypes

Characters	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
Germination percentage at 45 DAP	35.29	38.66	30.33	42.33	44.50
Number of shoots at 120 DAP (000/ha)	91.80	96.92	77.33	101.82	110.33
Number of Shoot at 240 DAP	135.52	142.34	120.00	157.20	148.00
Number of millable cane at harvest (000/ha)	78.10	86.62	60.00	92.87	78.15
Sugar yield (CCS t/ha).	61.89	76.35	49.00	67.46	54.03
Cane yield (t/ha).	6.97	8.71	6.12	7.63	6.59
Brix % at 12 months stage.	18.78	18.89	20.07	18.73	19.67
Purity % 10 months stage	87.01	87.46	87.93	87.87	86.83
Purity % at 12 months stage.	86.87	87.57	89.57	87.60	89.37
Pol % at 10 months stage.	15.72	16.04	16.66	15.82	15.83
Pol % at 12 months stage.	16.35	16.56	17.97	16.42	17.58
Brix % at 10 months stage.	18.07	18.34	18.93	18.00	18.23
Number of nodes with aerial roots	5.92	6.44	6.33	5.67	5.67
Plants height at 150 DAP (cm)	86.79	83.98	87.78	108.66	74.44
Plant height at 240 DAP (cm)	192.20	208.74	170.62	228.36	206.18
Plants height at 360 DAP(cm)	206.58	218.86	186.44	248.00	217.99
Single cane weight (kg)	0.79	0.88	0.81	0.73	0.72
Cane diameter at harvest (cm)	2.22	2.47	2.43	2.21	2.14
Leaf area index before water logging	1.79	2.24	1.73	1.97	1.67
Leaf area index at 30 days after waterlogging	2.41	2.92	2.17	2.78	2.07
Leaf area index at 60 days after waterlogging	2.81	3.39	2.22	3.41	2.82
No. of fully emerged leaves at 30 days after waterlogged	8.58	9.85	8.33	8.67	8.00
No. of fully emerged leaves at 60 days after waterlogged	9.92	11.52	9.67	9.67	9.67
CCS % at 10 months stage.	10.97	11.05	11.50	10.91	10.85
CCS % at 12 months stage.	11.22	11.40	12.50	11.31	12.22

CONCLUSIONS

High genetic diversity was noticed among the genotypes under study. The per cent contribution of characters indicated that CCS % at 12 months' stage was major contributor towards the divergence followed by leaf area index at 60 days after waterlogging, leaf area index at 30 days after waterlogging, number of fully emerged leaves at 60 days after waterlogging and leaf area index before waterlogging. The clones were grouped into

clusters II was the largest comprising of nine genotypes followed by cluster I with four genotypes while three clusters III, IV and V were solitary with single genotype.

The intra cluster distance values indicated that the cluster II comprising nine genotypes had highest genetic diversity. The inter cluster D^2 values also ranged widely from 85.15 between clusters IV and V to 306.52 between clusters III and IV. It was observed that cluster III was placed distantly from

cluster IV and V. And these clusters also recorded highest mean values for one or more characters. Therefore, crosses can be effected between genotypes of cluster III with genotypes of cluster II, IV and V to yield wide spectrum of variability for different characters which would help to identify superior genotypes and also to improve more than one superior character simultaneously.

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