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Multivariate analysis of genetic divergence in sugarcane using mahalanobis D² method

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Abstract

The present study evaluated genetic divergence among 30 sugarcane genotypes under environment using Mahalanobis D² statistics. Significant variability among genotypes was observed, indicating substantial genetic diversity. Based on D² values, the genotypes were grouped into three distinct clusters following Tocher's method, revealing the presence of wide genetic divergence independent of eco-geographical origin. The clustering pattern suggested the possibility of selecting genetically diverse parents for hybridization to obtain superior recombinants. Among the sixteen characters studied, brix percentage contributed the maximum towards total genetic divergence, followed by other quality and yield traits, indicating its importance in differentiating genotypes. The results emphasized that inter-cluster crosses involving genotypes from highly divergent clusters would be effective for improving yield and quality traits. Overall, the study highlighted the usefulness of D² analysis in identifying diverse genotypes for efficient sugarcane breeding programs under environment conditions.

Keywords: Sugarcane, *Saccharum* spp., genetic divergence, Mahalanobis's D² statistics, cluster distance, hybridization

Introduction

Agriculture serves as a critical bridge between natural ecosystems and human civilization and its advancement increasingly depends on precise analytical tools for crop improvement (Dodiya *et al.*, 2025) [7, 6]. In this context, the application of multivariate statistical techniques particularly the Mahalanobis D² method has become essential for assessing genetic divergence in crops like sugarcane (Dodiya *et al.*, 2023) [5]. Rather than relying solely on conventional agronomic practices, modern research emphasizes the identification and utilization of genetically diverse parents to enhance breeding efficiency and yield potential. Studies on sugarcane demonstrate that evaluating multiple traits simultaneously enables a more comprehensive understanding of genetic variability, thereby facilitating the development of superior cultivars through informed selection and hybridization strategies. Sugarcane (*Saccharum* spp. L.) is one of the most significant agro-industrial crops, playing a vital role in the global sugar economy and renewable energy sector. Apart from sugar production, it serves as a primary raw material for ethanol, cogeneration of electricity, and various by-products such as molasses, bagasse, and press mud. (Moore, P. H. and Botha, F. C. 2014) [17]. Its ability to accumulate high levels of sucrose in the stalk and produce large biomass under favorable conditions makes it a unique crop among field crops. Globally, Brazil leads in production, while India has the largest area under cultivation and ranks second in total production. In India, sugarcane serves as a major cash crop, contributing significantly to the agricultural economy, especially in states like Uttar Pradesh, Maharashtra, Gujarat, and Karnataka.

Botanically, sugarcane belongs to the family Poaceae and comprises complex polyploid (Sreenivasan, T. V., Ahloowalia, B. S. and Heinz, D. J. 1987) [31] and highly heterozygous genotypes derived from interspecific hybridization. (Jackson, P. A. (2005) [12] Modern cultivars mainly originate from *Saccharum officinarum* crossed with wild species such as *S. spontaneum*, resulting in improved yield and adaptability. Despite its economic importance, sugarcane productivity is often constrained by a narrow genetic base and complex genome structure.

Today's com-mercially grown sugarcane cultivars developed through interspecific and intervarietal hybridization programs with the crop known to carry the highest and variable number of chromosomes (usually an octabloid with $x = 10$ and aneuploidy) among the commonly grown crop plants (Prasanna *et al.*, 2005) ^[20]. Genetic diversity is a fundamental requirement for any crop improvement program, as it provides the raw material for selection and hybridization. (Singh, R. K. and Chaudhary, B. D. 1985) ^[28] The presence of wide genetic variation enables breeders to select superior genotypes and develop improved cultivars with desirable traits. (Poehlman, J. M. and Sleper, D. A. 1995) ^[23]. In sugarcane, genetic diversity is particularly important due to its polyploidy and highly heterozygous nature, which influence trait expression and inheritance patterns. (Gravois, K. A. and Milligan, S. B. 1992) ^[10]. Multivariate statistical techniques (Rahman *et al.* 2013) ^[24] especially Mahalanobis D² analysis, have been widely employed to assess genetic divergence among genotypes. (Mali, S. C. and Patel, A. I. 2017) ^[21]. This approach considers multiple traits simultaneously and provides a reliable estimate of genetic distances, (Sharma and Singh 2015) ^[27] facilitating the classification of genotypes into distinct clusters. (Kumar *et al.* 2011) ^[14] The selection of parents from genetically distant clusters is expected to produce higher heterosis and wider variability in segregating populations. (Arunachalam, 1981) ^[2]. In addition to conventional breeding approaches, modern biotechnology tools such as CRISPR-Cas9 genome editing are revolutionizing sugarcane improvement by enabling precise and targeted genetic modifications. While multivariate analysis using the Mahalanobis D² method helps identify genetically diverse parents for hybridization, CRISPR-Cas9 complements this approach by directly introducing desirable traits at the molecular level. This technology allows researchers to edit genes associated with yield, disease resistance, stress tolerance, and sugar content with high accuracy and efficiency. In complex polyploid crops like sugarcane, CRISPR-Cas9 offers significant potential to accelerate breeding programs by overcoming limitations of traditional selection methods (Dodiya *et al.* 2025, Dodiya *et al.* 2025) ^[7, 6]. When integrated with genetic divergence studies, genome editing provides a powerful framework for developing superior sugarcane varieties tailored to meet future agricultural demands.

In addition, understanding the contribution of individual traits towards total genetic divergence helps in identifying key characters responsible for variability. (Nair, N. V., Nair, S. and Sreenivasan, T. V. 2002) ^[18]. Such information is valuable in formulating effective breeding strategies aimed at improving both yield and quality parameters. Brown *et al.* (1978) ^[3]. Therefore, detailed assessment of genetic diversity using appropriate statistical tools is crucial for the identification of promising genotypes (Ishlam *et al.* 2011) ^[11] and the development of superior sugarcane varieties suited to diverse agro-climatic conditions.

Materials and Methods

The present investigation was carried out during Kharif 2023-24 at the Research Farm, Main Sugarcane Research Station, Navsari Agricultural University, Navsari, Gujarat, India. The experimental material comprised thirty genetically diverse sugarcanes (*Saccharum* spp. L.) genotypes. The experiment was laid out in a Randomized

Block Design (RBD) with three replications Each genotype was grown in recommended agronomic practices to ensure uniform crop growth.

Observations were recorded on sixteen quantitative and quality traits, namely germination percentage at 45 days after sowing (DAS), number of tillers at 120 days (000/ha), number of shoots at 240 days (000/ha), brix percentage, sucrose percentage, purity percentage, CCS percentage at 10 and 12 months, sucrose percent in cane, Fibre percentage, single cane weight (kg), millable height (cm), cane girth (cm), cane yield (t/ha), number of millable canes (000/ha), and sugar yield at harvest (t/ha). (Patel K.K *et al.* 2018) ^[22]

The data were subjected to analysis of variance (ANOVA) as per standard procedures to test the significance of differences among genotypes. Genetic divergence among genotypes was assessed using Mahalanobis D² statistics, and clustering of genotypes was performed using Torcher's method.

Table 1: Details of experimental material used for the study.

Sr. No.	Genotypes	Sr. No.	Genotypes	Sr. No.	Genotypes
1.	Co 86002	11.	CoN 18071	21.	CoN 19071
2.	Co 97009	12.	CoN 15072	22.	CoN 18072
3.	Co 7527	13.	Co 09004	23.	VSI 12121
4.	Co 6304	14.	CoM 20082	24.	VSI 3102
5.	Co 8338	15.	CoN 13073	25.	2014 N 1361
6.	CoC 671	16.	CoN 20071	26.	Co 11015
7.	Co 419	17.	VSI 18121	27.	Co 86032
8.	CoN 14072	18.	CoN 13072	28.	CoN 04131
9.	CoN 15071	19.	CoN 07072	29.	CoN 91132
10.	CoN 05071	20.	Co 62175	30.	Co 86249

Statistical Analysis

Analysis of variance, estimation of standard error and critical difference were carried out as per Panse and Sukhatme (1946) ^[19]. Test of significance for difference between characters' means was carried out by referring to the F- table given by Snedecor and Cochran (1967) ^[30]. Genetic divergence among the studied genotypes based on statistical differences was estimated by Mahalanobis's D² procedure using SPSS software (Statistical Package for Social Studies). Data obtained regarding genotypes classification in different clusters and results of inter and intra clusters D² values between clusters, as well as mean of intracusters D² values of different clusters were used for discussion and to draw conclusion.

Results and Discussion

Genetic divergence (D² analysis)

The genetic divergence among thirty genotypes of sugarcane was estimated based upon observation of eleven quantitative characters. (Tiwari, D. K. and Singh, R. K. 2015) ^[32]. The Mahalanobis's D² statistics was computed for all possible pair of populations under investigation. Verma *et al.* (2014) ^[34].

Distribution of genotypes into clusters

The thirty genotypes of sugarcane grouped into three clusters on the basis of relative magnitude of D² values following Tocher's method (Rao, 1952) ^[25], cluster I was the largest cluster having twenty-seven, fourteen and twenty genotypes. (Ali *et al.* 2008) ^[1] In the clustering pattern revealed low genetic divergence among most of the sugarcane genotypes as a single large cluster accommodated

27 out of the 30 genotypes. (Khan, A. Q. and Singh, J. R. 2001) ^[13] This cluster included commonly cultivated and widely adapted genotypes such as Co 86002, Co 97009, Co

6304, Co 8338, several CoN series lines, VSI entries and Co 11015, indicating close genetic similarity and comparable performance under environment conditions.

Table 2: Cluster composition of thirty genotypes of sugarcane

Sr. No.	Cluster	Total number of genotypes	Genotypes
1	I	27	Co 86002, Co 97009, Co 7527, Co 6304, Co 8338, CoC 671, Co 419, CoN 14072, CoN 15071, CoN 05071, CoN 15072, CoM 20082, CoN 13073, CoN 20071, VSI 18121, CoN 13072, CoN 07072, Co 62175, CoN 19071, VSI 12121, VSI 3102, 2014 N 1361, Co 11015, Co 86032, CoN 04131, CoN 91132, Co 86249
2	II	2	CoN 18071, CoN 18072
3	II	1	Co 09004

The remaining genotypes were grouped into two small clusters, with two genotypes (CoN 18071 and CoN 18072) forming a separate cluster II and Co 09004 appearing as a solitary cluster I. The occurrence of such small and mono-genotypic clusters suggests the presence of genetically distinct material. These genotypes may serve as useful parents in hybridization programmes to enhance variability. (Falconer, D. S. and Mackay, T. F. C. 1996) ^[9].

Intra and inter cluster distance

Intra and inter cluster distance (D) between all possible pairs of clusters were computed. (Singh, R. K., Singh, R. B. and Singh, S. P. 1994) ^[29]. Intra and inter cluster average (D) values are summarized in Table 3.

The inter- and intra-cluster D2 values in E1 revealed the extent of genetic divergence among the clusters. The intra-cluster distance was highest in cluster II (169.42), followed by cluster I (87.64), indicating that genotypes within cluster II were more heterogeneous compared to those within cluster I (Table 3). The cluster III showed an intra-cluster distance of zero, as it consisted of a single genotype, reflecting complete uniformity within the cluster. The inter-cluster distances indicated considerable genetic divergence among the clusters (Tyagi *et al* 1998) ^[33].

Table 3: Inter and intra cluster D2 values for different clusters

Cluster	I	II	III
I	87.64	308.92	185.59
II		169.42	292.37
III			0.00

The maximum inter-cluster distance was observed between cluster I and cluster II (308.92), suggesting that genotypes belonging to these two clusters were highly divergent and could be effectively exploited as parents in hybridization programmes to obtain higher heterosis. (Miller *et al* 1996) ^[16] This was followed by the distance between cluster II and cluster III (292.37), also indicating substantial divergence. The inter-cluster distance between cluster I and cluster III (185.59) was comparatively lower but still indicated moderate genetic divergence. Overall, the wide inter-cluster distances, particularly involving cluster II, suggest that crosses between genotypes from cluster II and those from clusters I or III would be promising for generating broad genetic variability and superior recombinants under environment conditions.

Conclusion

Genetic divergence analysis indicated varying levels of diversity. The presence of distinct and mono-genotypic

clusters suggests the availability of genetically diverse parents for hybridization. High inter-cluster distances indicate the potential for obtaining superior recombinants through crossing of divergent genotypes. Traits such as brix percentage and CCS contributed significantly to genetic divergence, highlighting their importance in genotype differentiation.

References

1. Ali A, Khan SA, Iqbal J. Genetic diversity in sugarcane genotypes. *Pakistan Journal of Botany*. 2008;40(6):2707-2714.
2. Arunachalam V. Genetic distance in plant breeding. *Indian Journal of Genetics and Plant Breeding*. 1981;41:226-236.
3. Brown AHD. Isozymes, plant population genetic structure and genetic conservation. *Theoretical and Applied Genetics*. 1978;52(4):145-157.
4. Burton GW. Quantitative inheritance in grasses. *Proceedings of the 6th International Grassland Congress*. 1952;1:277-283.
5. Dodiya RD, Barad AH, Pathan NP, Raghunandan BL. Trichogramma: a promising biocontrol agent. *International Journal of Economic Plants*. 2023;10:192-199.
6. Dodiya RD, Italiya JV, Patel PS, Pathan NP, Barad AH. CRISPR/Cas9-mediated gene disruption: a novel strategy against fall armyworm. *Farming Management*. 2025;10(1).
7. Dodiya RD, Koosi ST, Chaudhary FK, Patel PS, Barad AH. CRISPR/Cas9: a new frontier in biocontrol strategies for insect pest management. In: *Bio-control agents for sustainable agriculture: diversity, mechanisms and applications*. Singapore: Springer Nature Singapore; 2025. p. 511-530.
8. Dodiya R, Patel P, Pathan N, Deb S. Temporal patterns of aphid infestations in coriander. *Journal of Agricultural Ecology*. 2025;20:77-83.
9. Falconer DS, Mackay TFC. *Introduction to quantitative genetics*. 4th ed. Longman; 1996.
10. Gravois KA, Milligan SB. Genetic relationships in sugarcane. *Crop Science*. 1992;32(2):327-331.
11. Islam MS, Miah MAS, Begum MK. Genetic variability and character association in sugarcane. *Bangladesh Journal of Agricultural Research*. 2011;36(1):51-60.
12. Jackson PA. Breeding for improved sugarcane productivity. *Field Crops Research*. 2005;92(2-3):277-290.
13. Khan AQ, Singh JR. Genetic divergence in sugarcane. *Indian Journal of Genetics*. 2001;61(2):123-126.

14. Kumar N, Singh R, Singh PK. Genetic divergence in sugarcane using Mahalanobis D^2 statistics. *Progressive Agriculture*. 2011;11(2):289-293.
15. Mahalanobis PC. On the generalized distance in statistics. *Proceedings of the National Institute of Sciences of India*. 1936;2:49-55.
16. Miller JD, Tai PYP, Glaz B. Genetic variation and breeding potential of sugarcane. *Crop Science*. 1996;36(4):1037-1043.
17. Moore PH, Botha FC. Sugarcane: physiology, biochemistry, and functional biology. Wiley-Blackwell; 2014.
18. Nair NV, Nair S, Sreenivasan TV. Genetic variability in sugarcane. *Sugar Tech*. 2002;4(1-2):45-50.
19. Panse VG, Sukhatme PV. Statistical methods for agricultural workers. 2nd ed. Indian Council of Agricultural Research; 1946. p. 123-126.
20. Parasanna K, Ashok M, Parasannajeit M. Genetic divergence in sugarcane. *Indian Sugar*. 2005;53(3):33-38.
21. Patel DU, Mali SC, Patel AI. Genetic diversity analysis in sugarcane. *Journal of Sugarcane Research*. 2017;7(2):45-52.
22. Patel KK, Patel DU, Mali SC. Genetic variability and character association in sugarcane. *International Journal of Chemical Studies*. 2018;6(5):1560-1564.
23. Poehlman JM, Sleper DA. Breeding field crops. 4th ed. Iowa State University Press; 1995.
24. Rahman MM, Islam MS, Hossain M. Multivariate analysis in sugarcane for yield traits. *Journal of Agricultural Science*. 2013;5(3):30-35.
25. Rao CR. Advanced statistical methods in biometric research. John Wiley and Sons; 1952. p. 357-363.
26. Sharma ML, Singh R. Multivariate analysis in sugarcane. *Indian Sugar*. 2002;52(4):257-262.
27. Sharma RK, Singh PK. Genetic variability and divergence studies in sugarcane. *International Journal of Agricultural Science*. 2015;11(1):67-72.
28. Singh RK, Chaudhary BD. Biometrical methods in quantitative genetic analysis. Kalyani Publishers; 1985.
29. Singh RK, Singh RB, Singh SP. Genetic divergence in sugarcane. *Indian Journal of Genetics and Plant Breeding*. 1994;54(2):189-194.
30. Snedecor GW, Cochran WG. Statistical methods. Oxford and IBH Publishing Co; 1967.
31. Sreenivasan TV, Ahloowalia BS, Heinz DJ. Cytogenetics and breeding of sugarcane. *Critical Reviews in Plant Sciences*. 1987;6(2):117-160.
32. Tiwari DK, Singh RK. Genetic divergence analysis in sugarcane using D^2 statistics. *Electronic Journal of Plant Breeding*. 2015;6(3):789-795.
33. Tyagi SD, Singh DN, Singh R. Genetic divergence in sugarcane. *Indian Sugar*. 1998;48(4):257-260.
34. Verma RS, Singh PK, Singh J. Genetic variability and divergence in sugarcane genotypes. *Sugar Tech*. 2014;16(2):123-130.