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Phenotypic Characterization and Genetic Diversity of Sugarcane Varieties Cultivated in Northern Karnataka of India based on Principal Component and Cluster Analyses

Rakesh Tawadare¹

<https://orcid.org/0000-0003-3510-2309>

Devarajan Thangadurai^{1*}

<https://orcid.org/0000-0003-4126-163X>

Rayappa Bharamappa Khandagave²

<https://orcid.org/0000-0002-1007-4340>

Abhishek Mundaragi³

<https://orcid.org/0000-0001-6510-4829>

Jeyabalan Sangeetha⁴

<https://orcid.org/0000-0003-3798-3066>

¹Karnatak University, Department of Botany, Dharwad, Karnataka, India; ²S. Nijalingappa Sugar Institute, Belagavi, Karnataka, India; ³Davangere University, Department of Microbiology, Davangere, Karnataka, India; ⁴Central University of Kerala, Department of Environmental Science, Periyar, Kasaragod, Kerala, India.

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*Correspondence: drthangaduraid@gmail.com; Tel.: +91-836-2215314 (D.T.)

HIGHLIGHTS

- Genetic variation among 24 sugarcane varieties cultivated across Northern Karnataka in India.
- Phenotypic Characterization.
- PCA and AHC chemometric analyses.
- Co 86032 and CoC 671 were found to be elite varieties.

Abstract: Sugarcane is a major commercial crop grown in India and across the world. Hence, several elite varieties have been developed now-a-days to overcome many obstacles including abiotic stresses and diseases. The present study was undertaken to screen genetic variation among twenty four sugarcane varieties that are commonly cultivated across Northern Karnataka, India with reference to physicochemical characters. Experiment was conducted in triplicate following randomized complete

block design (RCBD) at S. Nijalingappa Sugar Institute, Belagavi, Karnataka, India during February 2016-17. Physiological parameters such as internode length, stalk height, plant height, stalk girth, number of internodes, single cane weight, single cane volume of juice, cane yield and recovery were investigated. Further, statistical techniques such as principal component analysis and agglomerative hierarchical clustering were performed to characterize the twenty four varieties. Among twenty four sugarcane varieties studied, Co 86032 and CoC 671 were found to be elite varieties with respect to sugar recovery and cane yield, whereas varieties such as Co 86032 and Com 0265 were found to be best with respect to cane yield only. Based on the results obtained, eight varieties, viz., Co SNK 09232, Com 0265, Co 86032, Co SNK 09293, Co SNK 07680, CoC 671, Co 13006 and Co 2001-15 were found to be good with respect to overall qualities. Further studies need to be involved with molecular marker that would help in identification of elite varieties which could substantially contribute to construction of genetic resources library that may in turn find maximum use in molecular breeding.

Keywords: Physicochemical characters; Principal component analysis; Agglomerative hierarchical clustering; Sugarcane.

INTRODUCTION

Sugarcane (*Saccharum*) is a genus characterized by high ploidy levels and composed of six species such as *S. officinarum*, *S. barberi*, *S. sinensi*, *S. spontaneum*, *S. robustum* and *S. edule* [1]. Assessment of genetic diversity of any food crop is considered to be most essential in breeding programs as it assists in selection and identification of desirable genotypes and diverse parental combination and for further improvement through selection in the segregating populations [2]. Sugarcane is widely cultivated across globe for various purposes, the foremost motive being sugar production in most of the under developed nations and as fuel with respect to production of ethanol and electricity in most of the developed nations [3]. Further *S. spontaneum* has significantly contributed in agronomically important traits such as increased biomass for cogeneration. It is apparent from previous reports that assessment of genetic diversity is essential in breeding programs for enhanced productivity. ICAR-Sugarcane Breeding Institute (ICAR-SBI), Coimbatore, India, is the pioneer in core collection and is the center that comprises all the germplasm and is actively involved in developing improved varieties for resistance against several abiotic and biotic stresses [4]. As many as 1410 clones have been produced in this context. The first interspecific hybrid Co 205 [between *S. officinarum* (Vellai; 2n=80) and *S. spontaneum* (Coimbatore local; 2n=64)] with wide adaptability was made and released for cultivation in subtropical India. S. Nijalingappa Sugar Institute based at Belagavi in Northern Karnataka, India, a subsidiary of ICAR-SBI is actively involved in developing several improved local varieties for farmers across this region. It has core collection of sugarcane and has witnessed some novel varieties for maximum yield and productivity.

Chemometric tools are often employed to understand and represent the complex data in to simple. Principal Component Analysis (PCA) and Agglomerative Hierarchical Cluster Analysis (AHC) are widely used tools in many experimental procedures for better understanding of data as PCA aids to minimize the variables and demonstrates the relationship among them [5]. Further, several studies indicate that multivariate statistical analysis, viz. PCA and AHC are more effective in evaluating genetic diversity among sugarcane varieties [6,7]. These techniques have been employed in assessment of genetic diversity among several other crops such as wheat, maize and chilli [8-10]. Generally, AHC is based on Euclidean distance method coupled with Ward's method [11]. In recent past, several studies have been reported on the sugarcane genetic diversity assessment using the AHC and PCA analyses [12-17].

A recent study by Govindaraj *et al.* [18] reported the genetic diversity analysis of sugarcane across Maharashtra, India wherein they assessed 41 germplasm collected from 29 districts of Maharashtra belonging to *S. spontaneum* and they noticed morphological and agronomical variability among the collected accessions. Several studies have also used advanced molecular markers in assessment of genetic diversity among the sugarcane cultivars [19]. The current study, however, the exploration and collection of local sugarcane cultivars cultivated across Northern Karnataka was undertaken to evaluate the genetic diversity based on morphological and biochemical characters. PCA and AHC were further used as statistical tools for efficient phenotypic characterization of sugarcane in identifying superior clones for maximized usage.

MATERIAL AND METHODS

Plant materials and field experiments

A total of twenty four sugarcane varieties maintained in sugarcane germplasm garden of S. Nijalingappa Sugar Institute (22°50' N; 108°14' E), Zardshapur, Belagavi, Karnataka, India have been taken for the present study during February 2016 - February, 2017. These twenty four sugarcane varieties were planted in a randomized block design with three replications and the varieties were planted in single rows of 2.5 m length, with 1.2 m row-spacing. Nine traits were investigated during the growing season (February, 2017) and observations were recorded on ten randomly selected plants of each variety for a range of physicochemical parameters, viz., internode length (cm), stalk height (cm), plant height (cm), cane girth (cm), number of internodes, single cane weight (kg), single cane volume of juice (l), cane yield (metric ton/hectare) and recovery (%) after sugarcane maturity, i.e., after twelve months (365 days) post-germination.

Statistical analysis

Ten plants per variety were selected and the mean values of the physicochemical measurements were used for statistical analysis. Collected data was subjected to variable transformation. The data matrix 24×9 was prepared and statistical analysis such as PCA and AHC were performed based on Pearson and Euclidean distance with wards method, respectively, using the statistical software (XLSTAT 2014.5.03, Addinsoft, USA).

RESULTS

Phenotypic characterization

Generally, sugarcane cultivation depends on the cane yield and sucrose content, and the cane yield was related to the traits of stalk height and stalk girth [20,21]. Nine traits include internode length (cm), stalk height (cm), plant height (cm), cane girth (cm), number of internodes, single cane weight (kg) and single cane volume of juice (litre), cane yield (metric ton/Acre) and recovery (%) which are important in sugarcane breeding programs. In this present study, high variability was observed for all the quantitative traits studied for twenty four sugarcane varieties. Table 1 shows the information related to the measurement techniques made for nine quantitative traits (Table 2).

Principal component analysis

By principal component analysis, nine traits were simplified to four principal components based on nine quantitative traits with cumulative variance contribution of 76.72%. Further, PC1 and PC2 contributed 47.50% of variance among the variables. However, PC1 and PC3 also exhibited a variance of 41.72%. The Eigen value of the

first principal component was 2.2263, the variance contribution rate was 24.73% followed by second principal component with Eigen value of 2.0487 and variance contribution rate of 22.764% and the Eigen value of the third principal component was found to be 1.525 with a variance contribution rate of 16.94%. Nevertheless fourth component also expressed the variability of 12.23% with Eigen value of 1.1012. Figure 3 describes the information on scree plot for the twenty four sugarcane varieties on principal components of 1-9.

It could be noted from the PC1 and PC2 biplot axes that stalk height and stalk girth were found to be most effective variables contributing 28.64 and 17.98 % respectively for PC1, whereas, cane yield (37.69%), single cane weight (25.98%) and plant height (10.49%) were best explained by PC2. Similarly, internode length, number of internodes, single cane volume of juice and recovery were effective variables for PC3 with a contribution of 27.53, 23.39, 22.35 and 16.42% respectively. Further, stalk height and internode length were closely related variables in PC1 which could be observed in figure 1. It is evident from the biplot axes PC1 and PC2 that the varieties such as Co SNK 07680, Co SNK 09227, Co SNK 09232, Co 2012-24, Co 2012-25, Co 2001-15 and Co SNK 081132 were found to be highest in stalk height and plant height as depicted in figure 2(b). Biplot axes PC1 and PC3 indicated that recovery and number of internodes are closely associated traits. Further, stalk height exhibited a negative correlation with single cane weight (Table 3 and 4).

Table 1. Physicochemical traits of 24 sugarcane cultivars of Northern Karnataka in India.

Physicochemical trait	Measurement technique
Height of single internode (cm)	Measured from top, middle, bottom and mean was taken
Stalk height (cm)	Length of stalk from tip to the bottom
Plant height (cm)	Length of the plant from top of leaf to bottom of bud
Stalk girth (cm)	Diameter of the stem at top, middle and bottom
Number of internodes	Number of internodes per stalk
Single cane weight (kg)	Weight of individual plant
Single cane volume of juice (l)	Juice volume per individual plant
Cane yield (Metric ton/acre)	Average weight of cane yield per hectare
Recovery (%)	Sugar concentration

Agglomerative Hierarchical Cluster Analysis

Figure 5 represents the dendrogram for the twenty four sugarcane varieties. AHC analysis grouped sugarcane varieties into three clusters, i.e., I, II and III using the Ward's method. Cluster I and II were more close to each other on contrary to cluster III. Cluster I consisted of 4 varieties, i.e., Co 2012-109, Co 06027, Co 0265 and Co 86032, wherein Co 2012-109 and Co 06027 showed 98% similarity.

Whereas cluster II consisted of 11 varieties, viz., Co 11024, Co 10023, Co 10024, CoC 671, Co SNK 0632, Co SNK 09268, Co 10027, Co 11023, Co SNK 7658, Co SNK 07337 and Co SNK 83495 and finally cluster III consisted of 9 varieties such as Co 2001-15, Co 13006, Co 2012-23, Co 2012-24, Co SNK 07680, Co SNK 09227, Co SNK 09293, Co SNK 09232 and Co SNK 0811324. When compared to PCA the varieties belonging to Cluster III were good at stalk height, internode length, plant height, number of internodes and recovery. Cluster I comprised varieties good at cane yield, single cane weight and stalk girth and finally, Cluster II constituted varieties good at single cane volume of juice and poor at other traits.

Table 2. Physicochemical characters studied for 24 sugarcane varieties cultivated across Northern Karnataka in India.

Variety	Internode length (cm)	Stalk height (cm)	Plant height (cm)	Stalk girth (cm)	Number of internodes	Single cane weight (Kg)	Single cane volume of juice (l)	Cane yield of (metric ton/hectare)	Recovery (%)
Co 2012-109	13.21±1.33	156.06±16.00	368.50±20.10	2.81±0.19	22.02±0.47	1.65±0.04	1.43±0.05	171.68±1.77	9.82±0.25
Co 06027	13.36±0.98	160.02±11.76	369.11±49.90	2.83±0.25	22.08±0.94	1.72±0.03	1.60±0.05	172.79±2.17	9.85±0.31
Co 11024	12.34±1.14	156.97±13.71	373.38±80.10	2.56±0.05	21.20±1.25	1.52±0.03	2.08±0.06	152.70±2.22	9.38±0.24
Co 10023	14.61±0.48	153.92±05.75	399.29±34.90	2.81±0.17	21.80±0.94	1.40±0.02	1.54±0.03	130.59±1.63	9.20±0.23
Co 10024	17.63±0.52	186.54±06.26	394.72±25.10	2.61±0.19	19.40±0.47	1.78±0.04	1.47±0.03	172.42±1.60	9.31±0.21
Co 2001-15	18.44±0.79	195.07±09.42	416.05±32.90	2.71±0.09	21.96±0.94	1.43±0.04	1.37±0.05	148.77±1.11	9.80±0.13
CoC 671	15.90±1.22	184.40±14.58	396.55±06.60	2.71±0.05	20.15±1.41	1.51±0.04	1.73±0.05	135.43±2.42	10.07±0.71
Co SNK 0632	18.19±1.56	187.45±18.67	407.82±32.00	2.81±0.12	21.90±0.82	1.53±0.03	2.18±0.02	120.75±1.03	9.85±0.32
Com 0265	15.60±0.32	178.31±03.80	397.76±20.10	3.01±0.05	22.65±1.25	1.92±0.04	1.46±0.04	199.78±0.96	9.30±0.61
Co SNK 09268	18.16±1.15	213.97±13.86	316.69±25.30	2.81±0.12	21.64±0.00	1.55±0.04	2.29±0.03	157.35±2.14	9.01±0.12
Co 13006	14.33±0.63	204.83±07.60	427.03±10.10	2.91±0.16	22.32±0.47	1.58±0.05	1.64±0.04	175.73±0.86	9.91±0.43
Co 10027	17.93±0.62	172.21±07.47	388.93±20.30	2.71±0.05	19.74±0.00	1.57±0.03	1.73±0.07	146.45±0.88	8.99±0.23
Co 2012-23	15.67±0.95	207.26±11.40	371.86±36.30	2.54±0.22	24.26±0.94	1.60±0.03	1.15±0.03	155.00±1.01	9.27±0.11
Co 2012-24	15.55±1.06	198.12±12.77	389.13±18.00	2.63±0.12	22.88±0.47	1.59±0.04	0.99±0.04	159.72±1.92	9.79±0.19
Co 11023	13.03±0.00	182.88±00.00	346.46±16.60	2.62±0.19	20.88±0.47	1.39±0.04	0.89±0.04	144.62±2.27	9.81±0.43
Co SNK 7658	10.67±1.15	185.93±13.86	349.50±20.10	2.82±0.14	20.76±0.47	1.48±0.04	1.10±0.05	138.05±1.38	9.75±0.32
Co SNK 07337	13.97±0.79	183.19±09.42	361.70±27.40	3.12±0.19	20.86±0.47	1.57±0.06	1.01±0.06	135.18±1.53	9.63±0.19
Co SNK 07680	17.65±0.84	212.14±10.06	411.48±40.20	2.52±0.09	21.52±0.47	1.62±0.03	1.01±0.06	162.74±2.07	9.94±0.12
Co SNK 09227	18.03±1.08	212.75±12.93	403.45±24.30	2.82±0.05	21.88±1.25	1.43±0.04	0.90±0.06	64.37±0.69	9.39±0.36
Co SNK 09293	17.83±0.96	213.36±11.49	448.06±30.20	2.82±0.09	22.22±0.94	1.73±0.06	0.85±0.05	77.87±0.66	9.91±0.44
Co SNK 09232	20.32±1.08	243.84±12.93	411.48±26.90	2.42±0.12	22.10±0.82	1.40±0.03	0.81±0.03	63.31±0.55	9.52±0.29
CO SNK 0811324	13.72±0.63	205.23±07.60	366.78±20.30	2.82±0.12	24.86±0.47	1.33±0.03	0.80±0.02	65.54±0.53	9.47±0.61
Co SNK 83495	15.14±1.26	195.07±15.14	343.41±18.80	2.42±0.09	21.96±0.94	1.20±0.05	0.79±0.04	47.52±0.89	9.83±0.81
Co 86032	15.29±0.12	189.99±01.44	328.27±08.90	2.82±0.12	23.22±0.82	1.88±0.06	1.44±0.04	79.16±0.67	9.97±0.74

Table 3. Pearson's Correlation coefficient among different physicochemical characters studied for 24 sugarcane varieties.

Variables	Internode length	Stalk height	Plant height	Stalk girth	Number of internodes	Single cane weight	Single cane volume of juice	Cane yield	Recovery
Internode length	1	0.602	0.459	-0.261	-0.073	0.046	0.039	0.068	-0.197
Stalk height	0.602	1	0.219	-0.289	0.301	-0.180	-0.427	0.125	0.053
Plant height	0.459	0.219	1	-0.001	-0.050	0.100	-0.106	0.170	0.159
Stalk girth	-0.261	-0.289	-0.001	1	0.052	0.395	0.208	0.268	-0.012
Number of internodes	-0.073	0.301	-0.050	0.052	1	-0.006	-0.286	0.316	0.071
Single cane weight	0.046	-0.180	0.100	0.395	-0.006	1	0.273	0.774	0.028
Single cane volume of juice	0.039	-0.427	-0.106	0.208	-0.286	0.273	1	-0.060	-0.254
Cane yield	0.068	0.125	0.170	0.268	0.316	0.774	-0.060	1	0.030
Recovery	-0.197	0.053	0.1589	-0.012	0.071	0.028	-0.254	0.030	1

Table 4. Principal components of 24 sugarcane varieties for nine physicochemical characters.

Variables	PC 1	PC 2	PC 3
Internode length	0.3812	0.2639	0.5248
Stalk height	0.5352	0.2570	-0.0013
Plant height	0.2219	0.3240	0.2700
Stalk girth	-0.4241	0.2086	-0.0887
Number of internodes	0.1351	0.2586	-0.4837
Single cane weight	-0.3729	0.5098	0.1026
Single cane volume of juice	-0.3865	-0.0779	0.4729
Cane yield	-0.1661	0.6140	-0.1074
Recovery	0.0700	0.0778	-0.4053

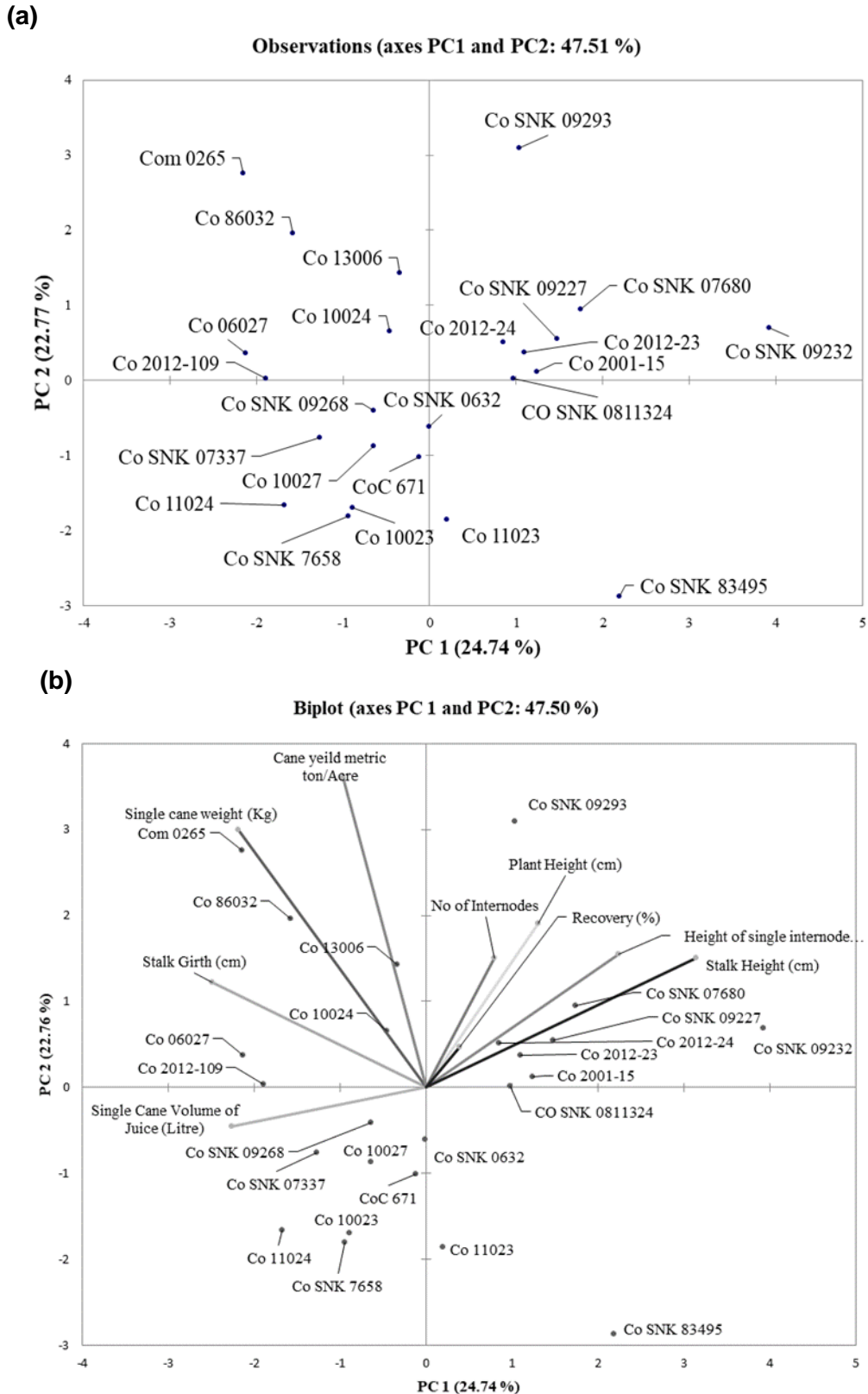


Figure 1. (a) Biplot representing 24 cultivars along the axes PC1 and PC2, (b) PCA loading plot for relationships between parameters for component PC1 and PC2.

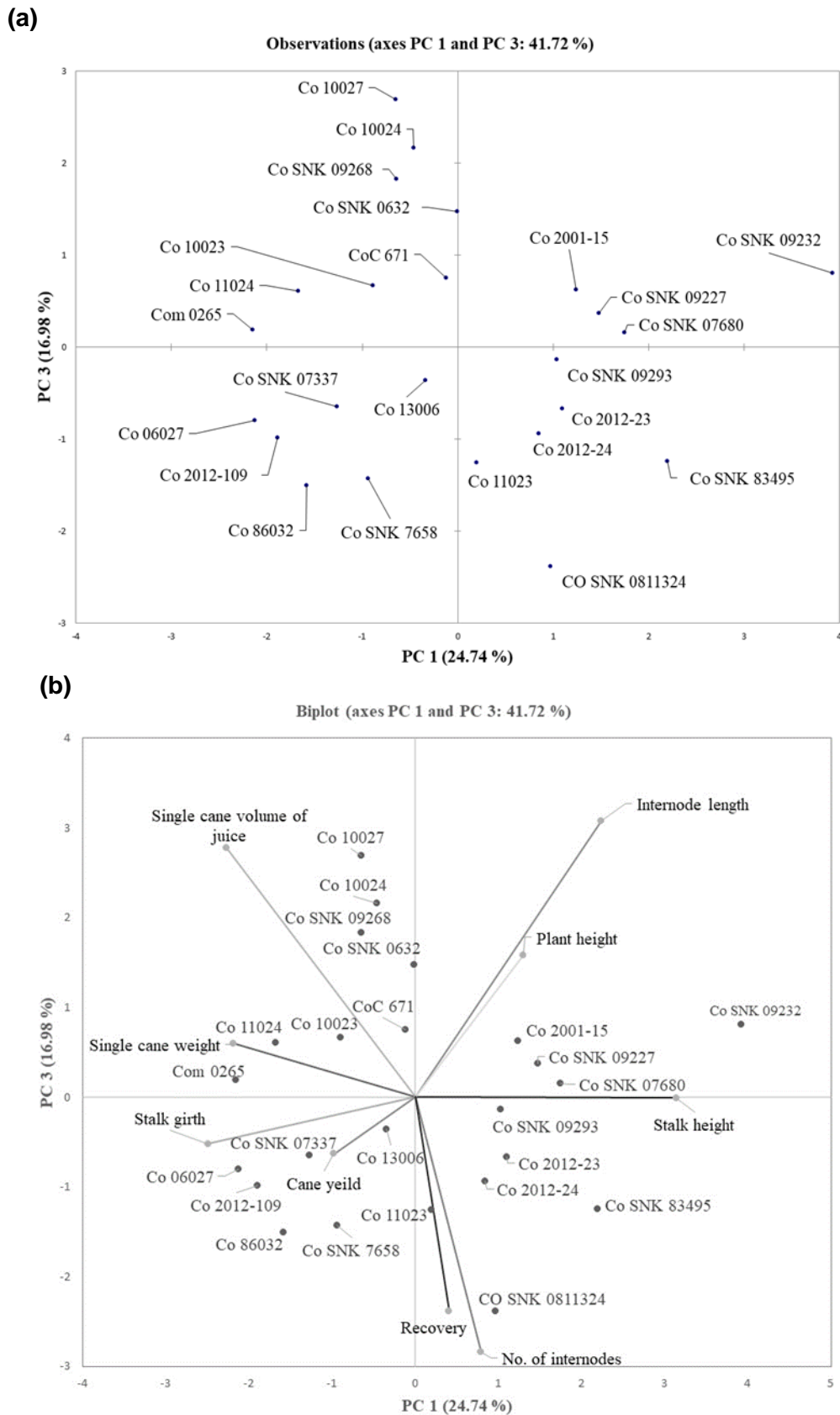


Figure 2. (a) Biplot representing 24 cultivars along the axes PC1 and PC3, (b) PCA loading plot for relationships between parameters for component PC1 and PC3.

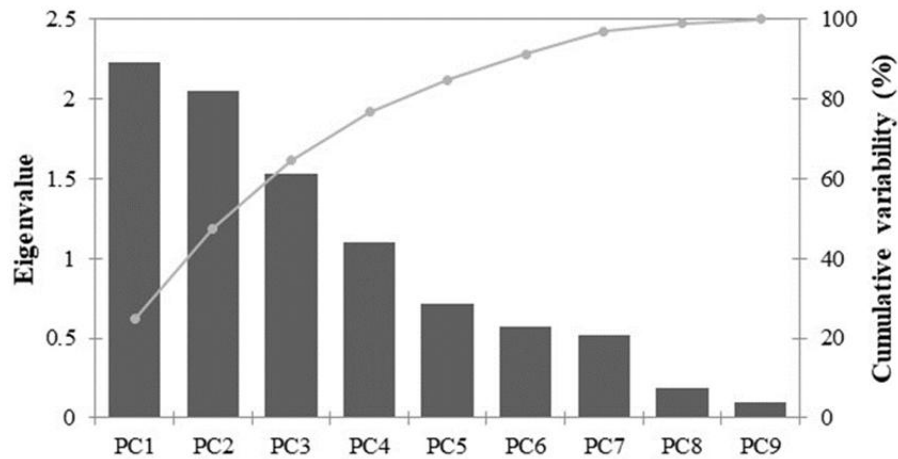


Figure 3. Scree plot describing the Eigen value and cumulative variance among the principal components.

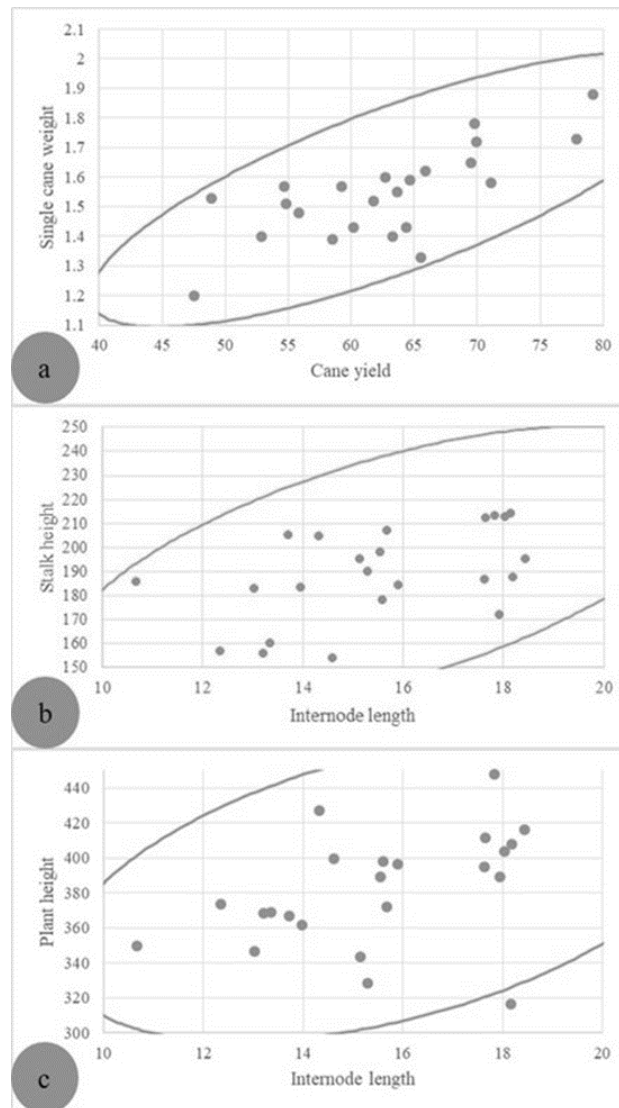


Figure 4. Scatter plot diagram of positive correlation observed between variables: (a) single cane weight vs. cane yield, (b) stalk height vs. internode length, and (c) plant height vs. internode length.

DISCUSSION

The results observed in the present study were in accordance with the previous literature. A recent study carried out by Arrey and Mih [6] reported similar observations, wherein their study involved assessment of genetic diversity among five sugarcane landraces of Cameroon with the aid of ten morphological quantitative traits. Pearson's correlation coefficient among nine variables was studied. Figure 4 explains scatter plot for the positive correlations observed between single cane weight and cane yield with a significant positive correlation coefficient value of 0.774 (Figure 4a).

Similarly, stalk height was found to be significantly correlated to internode length (0.602) followed by plant height that was in positive correlation with internode length with a value of 0.459 (Figure 4c). A study by Govindaraj *et al.* [18] made a similar observation wherein their study indicated that plant height had significant positive correlation with internode length (0.459). However, a negative correlation was observed among the single cane volume of juice and stalk height (-0.427).

Zhou *et al.* [21] investigated principal component and cluster analysis for 111 accessions of Guitang sugarcane germplasm based on 9 quantitative traits, wherein they observed 74.42% of cumulative variance for first four principal components. Smiullah *et al.* [22] reported genetic diversity assessment for ten genotypes using PCA and their study indicated cumulative variance of 67.3% for first two components. Similarly, another study by Zhang *et al.* [23] reported cumulative variance of 82.47% for first three components while studying ninety four *S. spontaneum* genotypes for seven traits. A most recent study indicated similar observations wherein total cumulative variance for first three components was 54.10, 26.70 and 13.10, respectively.

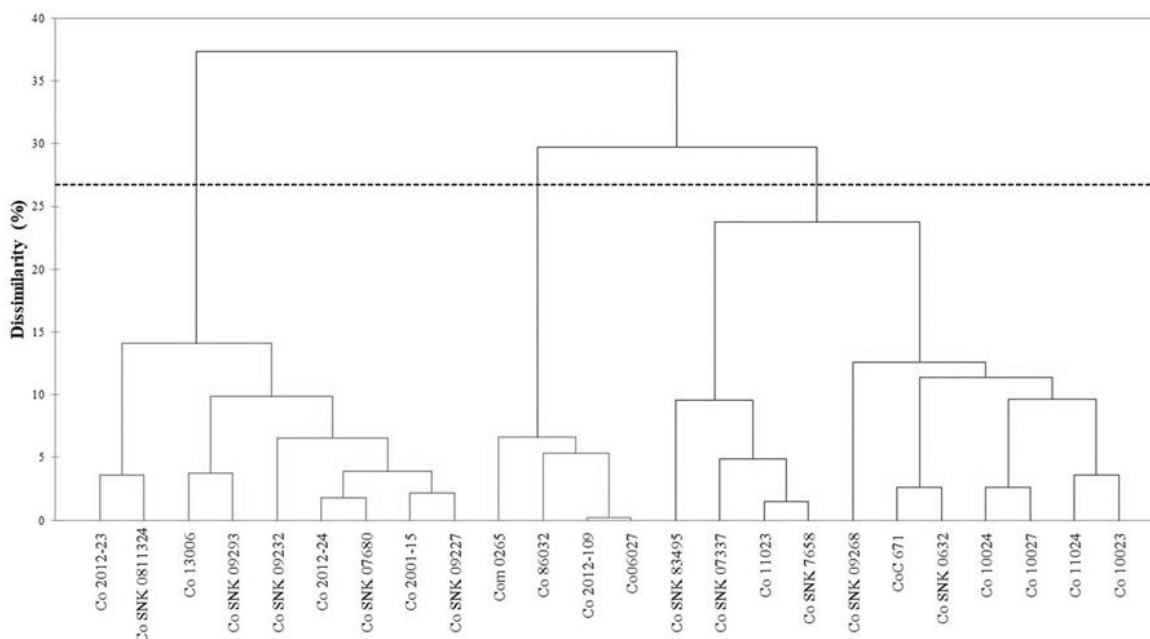


Figure 5. Dendrogram of cluster analysis for 24 sugarcane varieties of Northern Karnataka in India.

CONCLUSION

The sugarcane varieties which were studied and compared exhibited maximum variability, which could be utilized for further sugarcane crop improvement. Among the nine quantitative traits used in the present study, the PCA identified 4 principal components, contributing to 76.72% of variance. Thus, PCA reduces the number of variables in the data collection and provides useful information with multiple correlated variables further reducing screening cost which is beneficial in breeding and selection

process. AHC analysis categorized twenty four sugarcane varieties in to three categories. Both the PCA and AHC analyses implicated similar results. Based on the results of principal component and cluster analysis, 24 sugarcane varieties that are commonly cultivated across Northern Karnataka were grouped in three major groups depending on nine quantitative traits. Results indicated eight elite varieties, viz., Co SNK 09232, Com 0265, Co SNK 09293, Co 86032, Co SNK 09293, Co SNK 07680, CoC 671, Co 13006 and Co 2001-15 which could serve as potential varieties in terms of quality and vigor.

Conflicts of Interest: The authors declare no conflict of interest.

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