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RESEARCH ARTICLE

Stability analysis of promising sugarcane genotypes for cane and sugar yields using the AMMI model and GGE biplot

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Abstract

The sugarcane clone G 2008019 is a general cross progeny of CoV 92102 that outyielded the checks in initial breeding trials. To assess its stability and yield potential under saline stress, multi-environment trials comprising 20 environments were conducted from 2022 to 2024. It produced the highest mean cane yield of 123.41 t/ha with a commercial cane sugar percentage of 13.06, representing a 27.46%, 28.89% and 31.79% increase over the checks Co 86032 (96.82 t/ha), CoG 94077 (93.64 t/ha) and CoG 95076 (95.75 t/ha) respectively. Further, it exhibited a 29.35%, 35.09% and 34.16% increase in sugar yield over the checks, respectively. From the AMMI (additive main effects and multiplicative interactions) and GGE (genotype × genotype interaction with environment) biplots, the performance and stability of the test genotypes were ascertained. The ANOVA analysis showcased a significant contribution of genotypes to total variation followed by the genotype × environment interaction and the environment itself. The yield potential and the stability of G 2008019 were confirmed through minimal ASV values and higher values for cane yield and juice-based quality traits. The biplots of AMMI I, AMMI II and GGE confirmed the constancy of G 2008019. Additionally, the sugarcane clone G 2008019 possessed good jaggery qualities, including 65.36% juice recovery, 91.84% juice purity, 11.32% jaggery recovery and 15.21 t/ha jaggery yield. The mean fibre content of the clone was 13.25%. These combined results indicate the suitability of clone G 2008019 for further utilization in the breeding cycle.

Keywords

AMMI stability analysis; G 2008019; GGE biplot; sugarcane

Introduction

Sugarcane belongs to the grass family and is capable of storing sugar in its stem. It is cultivated in approximately 110 countries under varied weather regimes. Its products serve as inputs for several food and non-food industries. Among sugarcane-producing countries, Brazil leads with an annual production of 752.8 million tonnes (MT) of sugar. Globally, India is the second-largest producer of cane, with a 15.39% share. It supplies 80% of inputs to sugar industries and 35% to bioethanol companies (1). The sugar industry holds significant economic importance because (i) it provides direct employment to 50 million growers (ii) it generates 0.5 million indirect jobs in associated industries (iii) it meets domestic and international sweetener demands through white sugar, jaggery and khandsari. The key factors determining the jaggery quality are variety, juice recovery and juice purity (2).

This implies that not all high-yielding varieties are suitable for jaggery production, necessitating targeted trials to assess juice characteristics. Typically, the fibre content in sugarcane ranges from 8 to 14% (3). Higher fibre content increases the amount of cane required to produce the same volume of sugar. Therefore, assessing fibre content in a sugarcane clone before commercialization is crucial. The demand for high-yielding and high-quality sugarcane varieties continues to rise due to the increasing demand for sugar and the introduction of blended petrol initiatives. In sugarcane breeding, a single variety cannot meet all farmers' requirements, as sugarcane is grown in both tropical and subtropical climatic regimes. Therefore, the identification of location-specific parents, their hybridization and offspring evaluation are essential (4).

Soil salinity (SS) is an accumulating abiotic stress that reduces the area of agricultural lands, and the problem gets aggravated year after year. SS negatively impacts the quality and productivity of agricultural produce (5). It is estimated that 20% of global cultivable lands are salinized (6). The issue of SS is more severe in agricultural regions where supplementary irrigation is frequent. A significant portion of Indian farmlands is located in tropical and subtropical regions, where the number of supplementary irrigations is higher than other areas. Mismanagement of irrigation in these lands leads to secondary salinization (6). Tamil Nadu is a major hub of the tanning industry in India, with most tanneries located in the Vellore, Ranipet and Tirupattur districts (7). Issues related to the treatment of tannery wastes and their indiscriminate disposal degrade water quality for both drinking and agricultural use, further increasing soil salinity (8). SS reduces soil and water quality and hinders plant development and productivity (9, 10). Sugarcane is no exception, as SS lowers its productivity. Therefore, breeding new sugarcane varieties suited to SS-affected conditions is essential for sustaining sugarcane cultivation.

Keeping this in view, the sugarcane clone G 2008019 along with a few checks were tested in salt-affected areas (20 environments) in four cropping years (2021, 2022, 2023, and 2024) to assess the stability for yield performance.

Materials and Methods

The fluff-grown seedlings of the biparental cross synthesized during the hybridization year 2008 were tested for various agronomic traits, including quantitative and quality traits, from 2009 to 2020. After several breeding cycle tests, 10 clones were evaluated in the advanced yield trial. In the breeding evaluation trials, yield-and quality-contributing traits such as cane yield, commercial cane sugar percentage and sugar yield were considered. Among them, clone G 2008019 exhibited significant results for both yield and quality. A multi-environment trial was conducted across 20 locations from 2021 to 2024 to assess its performance under salinity stress. In the SS trials, soil and irrigation-water characteristics were assessed (11). The ranges for soil pH and EC were 8.0 to 8.5 and 0.1 to 0.3, respectively. For water, the pH ranged from 7.1 to 8.0 and EC ranged from 2.3 to 5.9, respectively. The sodium absorption ratio (SAR) varied between 1.62 and 4.58, while HCO^3 ranged from 6.8 to 10.8.

The checks used in the experiments were Co 86032, CoG 95076 and CoG 94077. The check Co 86032 is predominantly grown in Tamil Nadu. Check CoG 95076 is tolerant to tannery effluents, while the check CoG 94077 is a jaggery variety. Since the tested clone G2008019 was evaluated for high yield, jaggery quality and suitability for SS, all these checks were included in the experiments. To establish a healthy crop stand, the recommended crop spacing, fertilizer dose (275:112.5 kg of N and K_2O/ha), irrigation intervals (once every 7-10 days) and plant protection measures were adopted. In each environment, three replications were laid out. At harvest, traits such as cane yield per hectare (CYH), commercial cane sugar percent (CCSP), sugar yield per hectare (SYP), Brix (%), pol (%) and purity (%) were recorded. CYH was determined by weighing the clean canes (devoid of green tops) on a plot basis and later converting the values to a hectare basis.

For measuring other traits, 20 full canes were selected from each replication. The green tops and leaf sheaths were removed and the canes were weighed. These canes were crushed before microbial degradation using an electrified sugarcane crusher. The Brix (%), pol (%), CCSP and PP were estimated according to ICUMSA methods (12, 13). The filtered and clarified juice was utilized to measure the brix with a handheld refractometer (0-28 Brix°) (Erma, Tokyo, Japan). The PP was measured using an Autopol I Automatic Polarimeter (Rudolph Research Analytical). The formula used was pol factor × correction factor. As per the methodology, the data on pol (%) and Brix (%) were used to calculate the CCSP (14). The sugar yield per hectare (SYH) was calculated using the data for CYH and CCSP as follows:

$$SYH = (CYH x CCSP)/100 \quad \text{(Eqn. 1)}$$

The purity percentage is the ratio between pol (%) and Brix (%). The ratio between the quantity of clean cane crushed and juice yield is considered the juice recovery percentage. The ratio between pol (%) and Brix (%) was used to determine the purity percentage. After the juice extraction, the bagasse was dried completely and weighed. The weight difference between fresh clean cane crushing and dried bagasse was measured. The average value across locations and replications was documented as the fibre content of the clone.

Statistical analysis

The mean data of 60 trials were utilized for statistical analyses. The first and second-order statistics (mean and standard error) were worked out using Microsoft Excel 365. For performing AMMI analysis (15). The procedure of (16) was utilized for GGE estimations. For stability analyses, the R studio Metan package was utilized.

ASV: AMMI stability value

The ASV was computed by adopting the formula and the stability of the tested genotypes was assessed based on these ASVs (17).

$$ASV = \sqrt{\left\{\left(\frac{SSIPCA1}{SSIPCA2}\right) \times (IPCA1Score)\right\} + (IPCA2Score)^2} \quad \text{(Eqn. 2)}$$

SSIPCA1 refers to the sum of squares for IPCA1 and SSIPCA2 denotes IPCA2. A lower ASV indicates greater stability of the tested sugarcane genotype. The IPCA scores (either negative or positive) were used to identify location-specific genotypes.

Yield stability index (YSI)

A genotype's yield potential and stability are used to compute YSI (YSI = RASV + RY). RASV represents a genotype's AMMI-derived ASV, while RY refers to a genotype's yield rank. A genotype is considered stable and high - yielding when it has low RY and RASV values (18).

Results and Discussion

Sugarcane is a semi-perennial cash crop grown under diverse climatic conditions. Its yield potential is challenged by a variety of abiotic stress factors, such as soil salinity, drought, high temperatures, frost and sodicity. SS is one such major abiotic stress. Vellore district of Tamil Nadu is the hub of leather tanneries. The wastewater discharged from these tanneries in the long term causes soil salinity (8). Sugarcane is a major cash crop grown in these districts and its yield is reduced due to SS. Sugarcane can sustain its growth under moderate salinity levels (up to 1.7 dSm^{-1} (19)). Higher salinity levels reduce sugarcane yield by modifying various metabolic processes (20, 21). It is well known that a high-yielding sugarcane variety released for commercial cultivation may not always perform well under SS and vice versa. Therefore, SRS, Melalathur, is working on the development of location-specific SS-tolerant sugarcane varieties to sustain sugarcane productivity in these districts. The following is one of the findings of its multi-environmental experiments conducted from 2022 to 2024 under SS.

Multi-environment-experiments under SS (mean of 60 experiments)

The promising sugarcane clone G2008019 was tested under SS on multi-environment trials along with the check varieties Co 86032, CoG 95076 and CoG 94077. The evaluation trials were conducted in 20 different SS environments. The clone G 2008019 recorded a CYH of 123.41 t/ha, which was higher than that of the salt-tolerant variety Co G 95076. The increase in CYH was 31.79%. Likewise, it exhibited 27.46% and 28.89% higher yields than the other checks (Co 86032 and CoG 94077, respectively). The clone G 2008019 had a mean CCSP of 13.06%, which was 1.48%, 4.82% and 1.79% higher than those of the checks (Co 86032, CoG 95076 and CoG 94077). Because of its higher CYH and CCSP, the clone G2008019 outyielded the checks for SYP. It produced a mean SYH of 16.12 t/ha, which was higher than that of the checks (Table 1). The clone G 2008019 exhibited notable jaggery quality traits (Table 2). The juice purity, juice recovery and jaggery recovery were 91.84%, 11.32% and 15.21%, respectively. Because of these parameters, the clone G 2008019 produced high-quality jaggery at 15.21 t/ha. Earlier studies revealed that jaggery yield potential and quality depends on variety, juice recovery and juice purity (2). Juice purity determines the quality of jaggery, as high juice purity is not dependent on chemical juice clarifiers. Reduced use of chemical clarifiers improves the shelf life of jaggery, which fetches a good market price for growers. Sugarcane fibre content normally ranges from 8 to 14% (3). A lower fibre content may increase susceptibility to stress factors, while excess fibre increases the burden on millers, as it negatively affects sugar content and its recovery. The tested clone G 2008019 had a fibre content of 13.25% and therefore, is considered optimal (Table 2).

AAMI and GGE biplot analysis for yield and stability

To understand the magnitude of the interaction between genotypes and environments, statistical techniques such as GGE and AMMI biplots are commonly used. AMMI analysis predicts the interaction between environment and

Table 1. Mean yield performance of the sugarcane clone G 2008019 in the multi-environment-based SS experiments*

Genotypes	CYH	CCSP	SYH
G 2008019	123.41 ± 7.34	13.06 ± 0.58	16.12 ± 6.37
Co 86032 (C)	96.82 ± 12.51	12.87 ± 0.71	12.46 ± 8.55
CoG 94077 (C)	93.64 ± 9.76	12.83 ± 0.63	12.01 ± 7.26
CoG 95076 (C)	95.75 ±8.22	12.46 ± 0.81	11.93 ± 8.41
% increase over Co 86032	27.46	1.48	29.35
% increase over CoG 94077	28.89	4.82	35.09
% increase over CoG 95076	31.79	1.79	34.16

* Mean of 20 environments ± SE, CYH: cane yield per hectare; CCSP: commercial cane sugar percent; SYH: sugar yield per hectare

Table 2. The jaggery quality parameters of the sugarcane clone G 2008019 in the multi-environment-based SS experiments*

Parameters	Genotypes			
	G 2008019	Co 86032 (C)	CoG 95076(C)	CoG 94077(C)
Juice recovery (%)	65.36±2.46	61.0±7.36	56.00±6.33	57.2±6.22
Brix (%)	21.87±0.73	21.6±0.93	20.74±0.81	20.8±0.59
Purity (%)	91.84±1.84	90.36±1.75	89.65±1.54	88.43±1.64
Jaggery recovery (%)	11.32±0.49	10.75±0.82	10.23±0.92	10.51±0.76
Fibre content (%)	13.25±1.85	13.36±2.55	13.72±2.34	14.13±2.13
Jaggery yield (t/ha)	15.21±1.21	12.35±1.71	11.23±1.42	11.46±1.27
% Increase jaggery yield over checks		23.16	35.44	32.72

* Mean of 20 environments ± SE

genotypes in multi-environment trials with interaction effects inferred from the biplots. However, in AMMI analysis, drawing inferences on genotype ranking is not possible, as stability alone is considered for testing genotypic suitability (22, 23). Therefore, in this study, GGE biplots were generated alongside AMMI biplots to provide both genotype and environment rankings. Additionally, ideal environments were identified (24). A significant effect due to genotypes, environment and their interactions was observed. Among the sources of variation, genotypic variation was the most significant. The main effect due to genotypes explained 95.48%, 95.39% and 57.36% of the total variation for CYH, SYH and CCSP respectively. A pronounced influence of the environment and its interaction with the genotypes was observed in the expression CCSP, with effects of 13.12% and 29.52%, indicating statistical significance. A similar result was previously reported (25). In multi-environment trials, total variations are explained through principal components (PCs). In this SS evaluation experiment, the data were represented by four PCAs. The first two principal components (PC1 and PC2) accounted for 72.87% for CYH, 73.70% for SYH and 88.99% of CCSP (Table 3). The effect of PC3 was non-significant for CYH and SYH, while the impact of PC 4 was zero for all traits.

The clone G 2008019 displayed its significance for CHY, SYH and quality traits in the SS trials. The sugarcane test genotypes were yield-ranked for yield (RY) based on their performance under SS environments. To evaluate the stability of these genotypes, the ASV was employed. To estimate ASV scores, with a view to justifying the interaction effects between genotypes and the environments, the environmental scores pertaining to PCA 1 and PCA 2 were considered. In the stability analysis experiment, a genotype is considered promising when it has a high yield with a minimal ASV (26). For CYH, SYH and CCSP, G 2008019 exhibited minimal ASV values, indicating its stability under SS environments. Based on their ASV values, the sugarcane genotypes were ranked (rASV). To derive the genotypic index

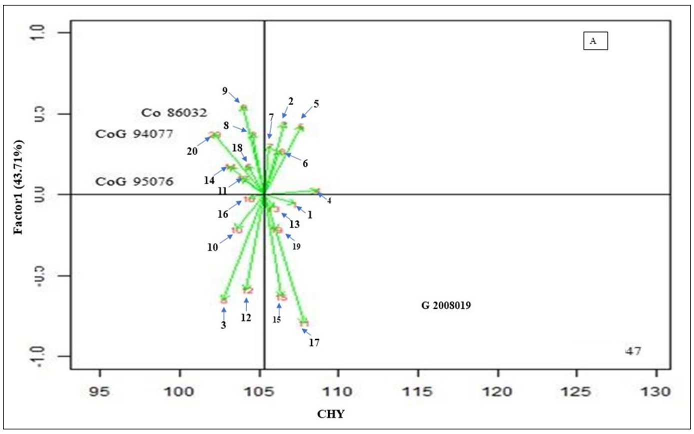

Fig. 1. (A). AMMI biplot PC1 (factor 1) vs. CYH where red font numbers indicate SS environments (E1– E20).

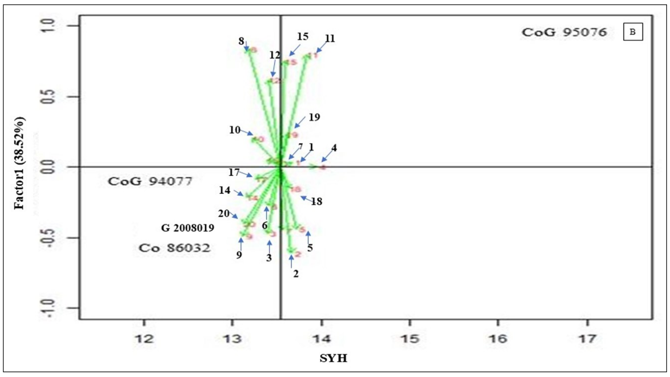

Fig. 1. (B). AMMI biplot PC1 (factor 1) vs. SYH where red font numbers indicate SS environments (E1– E20).

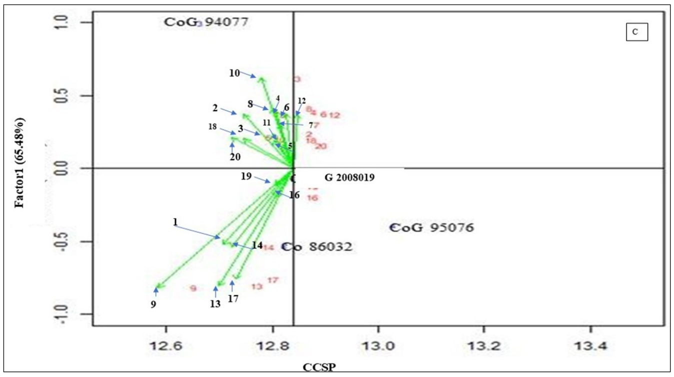

Fig. 1. (C). AMMI biplot PC1 (factor 1) vs. CCSP where red font numbers indicate SS environments (E1– E20).

Table 3. Analysis of variance of main and interaction effects for metric traits in sugarcane (AMMI)

Source	DF	CYH		SYH		CCSP	
		MS	VE %	MS	VE %	MS	VE %
Environment	19	38.1426**	1.25	0.67484**	1.41	0.04172 ***	13.12
Genotype	3	13842.26735***	95.48	267.27152***	95.39	1.28015***	57.36
Interaction (Env x Genotype)	57	31.0238**	3.27	0.5264**	3.19	0.03714***	29.52
PC1	21	39.8515**	43.71	0.68251*	38.52	0.06107***	65.48
PC2	19	30. 4372**	29.16	0.60924*	35.18	0.02362***	23.51
PC3	17	26.24062^{NS}	21.67	0.41732^{NS}	21.92	0.00581***	4.85
PC4	15	0^{NS}	0	0^{NS}	0	0^{NS}	0
Residuals	160	18.5209	0	0.31726	0	0.00281	0

CYH: cane yield per hectare; CCSP: commercial cane sugar percent; SYH: sugar yield per hectare; DF = degrees of freedom; MS = mean sum of square; VE% = variability explained in percentage; * = significance at 5%; ** = significance at 1% and *** = significance at 0.5% NS = non-significance for stability, rY and rASV were summed (27). The YSI and rYSI values for genotype G 2008019 (6 and 1, respectively) indicate its suitability for SS environments (Table 3).

In this experiment, AMMI I and II biplots were produced to infer the concurrent influences of environments and genotypes. Genotypic effects are inferred based on the alignment of genotypes relative to the biplot's vertical line; those aligned with the vertical axis exhibited greater main effects, whereas genotypes positioned horizontally indicated equal interaction between environment and genotype. PCA 1 contributed significantly to CYH, SYH and CCSP, accounting for 45.5%, 40.79% and 69.32% of the variance, respectively (Fig. 1, 2). The biplot of CYH versus PC1 indicates that environments E5, E2, E7, E6, E13, E1, E15, E19 and E17 are greater influence. Environments with greater main effects for SYH were identified as E15, E19, E11, E18, E5, E2 and E7, while those for CCSP were identified as E3, E6, E8, E20, E4, E7, E12, E18, E2, E16 and E19. Combined, the biplot information revealed that environments E7, E2 and E19 exerted significant effects. Environments plotted near the origin are assumed to exert minimal or negligible interaction and, therefore, will not significantly influence the trait expression of genotypes. For CYH and SYH, environments E4, E1 and E13 are located closer to the origin, indicating minimal interaction effects on these traits. Similarly, environments E19, E18 and E20 exerted minimal interaction effects on the expression of CCSP. Finally, the sugarcane genotypes G 2008019, CoG 94077 and CoG 95076 displayed greater main effects for CYH, SYH and CCSP.

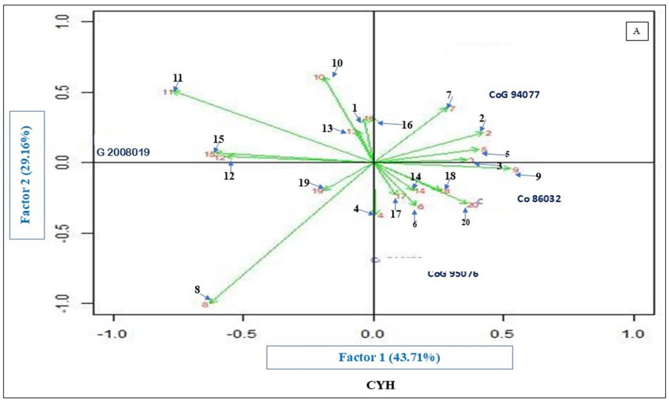

Fig. 2. (A). AMMI biplot PC1 (factor 1) vs. PC2 (factor 2) for CYH where red font numbers indicate SS environments (E1–E20).

Mean vs. stability

Using the mean versus stability biplots from these SS experiments, the average performance of the sugarcane genotypes can be effectively assessed. The average-environment coordinates (AECs) are critical in defining the biplot axes. The AEC, drawn as a line through the origin (under the condition of single value portioning of 1), classifies the tested genotypes along the abscissa (vertical axis) and ordinate (horizontal axis). The mean versus stability biplot analysis explained 98.79%, 97.74% and 91.51% of the variation attributable to genotypic main effects and genotypes × environment interactions for the tested traits (Fig. 3). Along the abscissa, an arrow denotes the better-performing and stable genotypes (24). Based on these considerations, genotype G 2008019 is identified as both stable and high - performing for CYH, SYH and CCSP.

Ranking of genotypes

The ranking of genotypes plays a crucial role in selecting promising candidates in a multi-environment-based experiment. The following two criteria are considered for the selection of promising genotypes, (i) plotting in the arrow end circle AEC abscissa and (ii) placing on the left side of the vertical axis (24). The biplots depicting genotype ranking confirmed that G 2008019 is the most productive and stable genotype for CYH, SYH and CCSP (Table 4, Fig. 4).

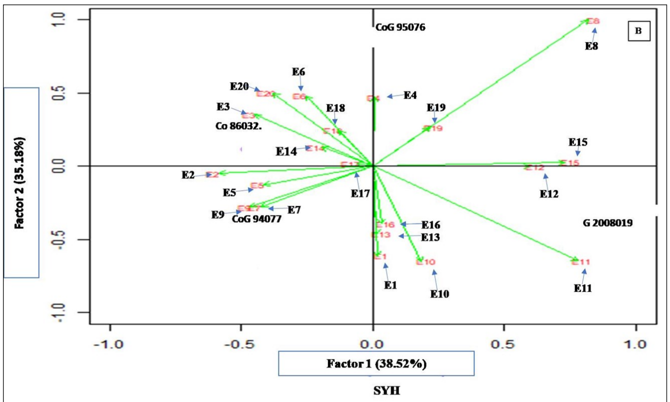

Fig. 2. (B). AMMI biplot PC1 (factor 1) vs. PC2 (factor 2) for SYH where red font numbers indicate SS environments (E1–E20).

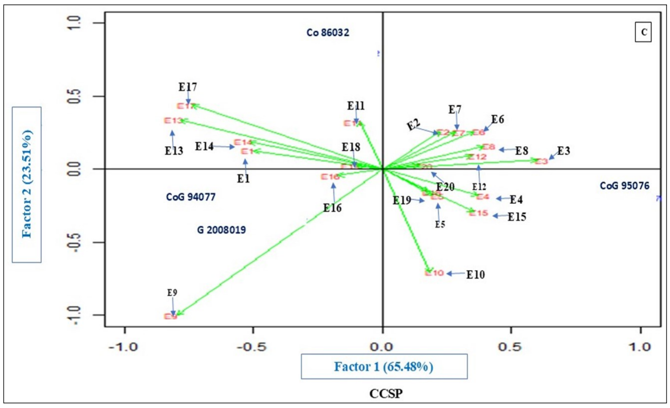

Fig. 2. (C). AMMI biplot PC1 (factor 1) vs. PC2 (factor 2) for CCSP where red font numbers indicate SS environments (E1–E20).

Table 4. The details of the yield and stability ranking of the sugarcane genotypes

Genotype	CYH						CCSP						SYH					
	M	rY	ASV	rASV	YSI	rYSI	M	rY	ASV	rASV	YSI	rYSI	M	rY	ASV	rASV	YSI	rYSI
G 2008019	123.41	1	2.26	1	2	1	13.03	1	0.54	1	2	1	16.42	1	0.62	1	6	1
Co86032 (C)	96.82	2	2.35	2	4	2	12.84	2	0.72	2	4	2	12.24	2	1.41	4	5	2
CoG94077 (C)	93.64	4	3.37	3	7	4	12.81	3	0.98	3	6	3	11.78	3	1.03	3	6	3
CoG95076 (C)	95.75	3	4.72	4	7	3	12.43	4	1.76	4	8	4	11.78	4	1.02	2	2	4

YHC: cane yield per hectare; CCSP: commercial cane sugar percent; SYH: sugar yield per hectare M = mean of the traits over 20 environments; rY=rank of yield; ASV= AMMI stability values; rASV = rank of AMMI stability values; YSI = yield stability index; rYSI = rank of the YSI

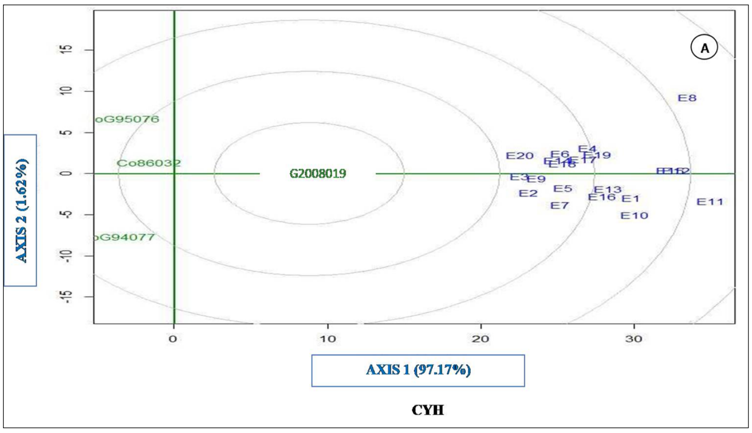

Fig. 3. (A). Mean vs. stability biplot for CYH where E1, E2, E3… denotes SS environments (E1- E20).

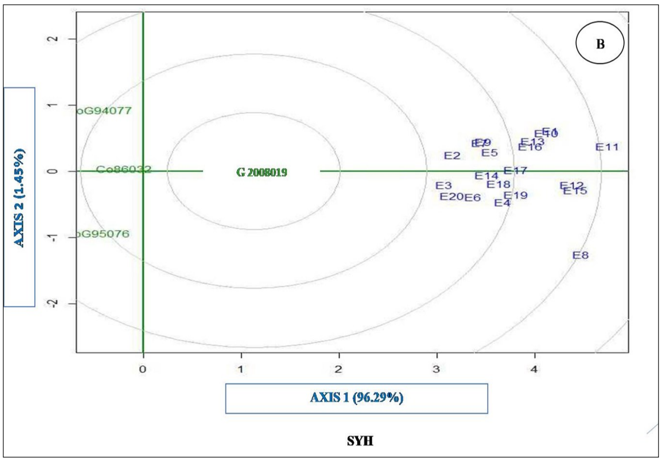

Fig. 3. (B). Mean vs. stability biplot for SYH where E1, E2, E3… denotes SS environments (E1- E20).

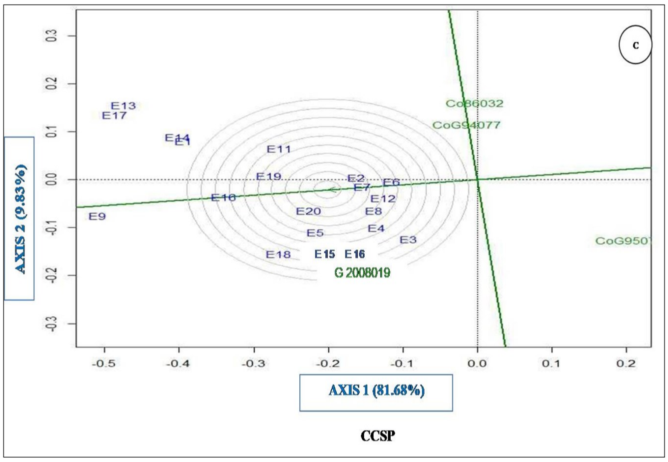

Fig. 3. (C). Mean vs. stability biplot for CCSP where E1, E2, E3… denotes SS environments (E1- E20).

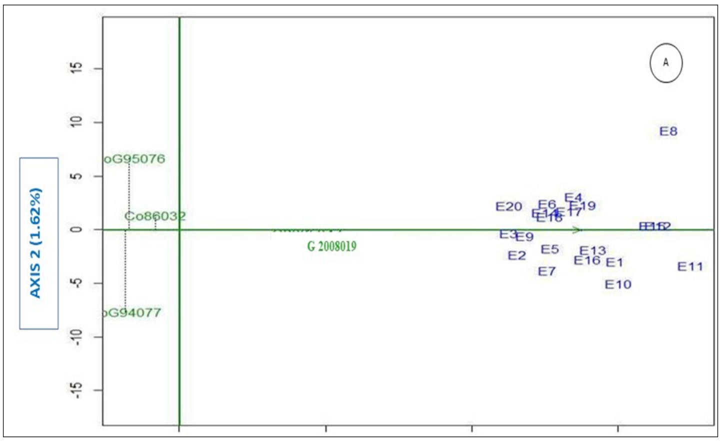

Fig. 4. (A). Ranking biplot of sugarcane genotypes for CYH where E1, E2, E3…denotes SS environments (E1-E20).

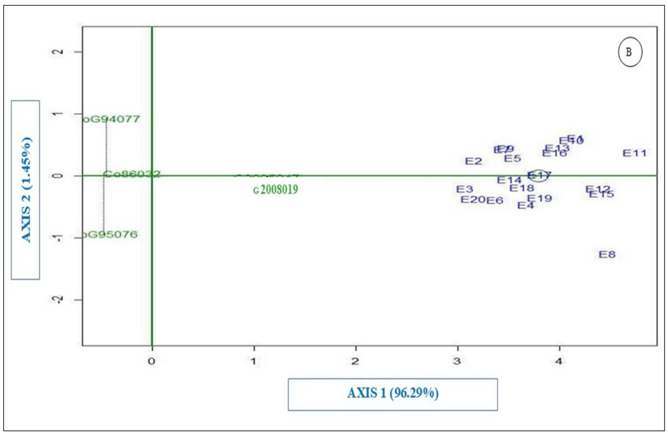

Fig. 4. (B). Ranking biplot of sugarcane genotypes for SYH where E1, E2, E3… denotes SS environments (E1-E20).

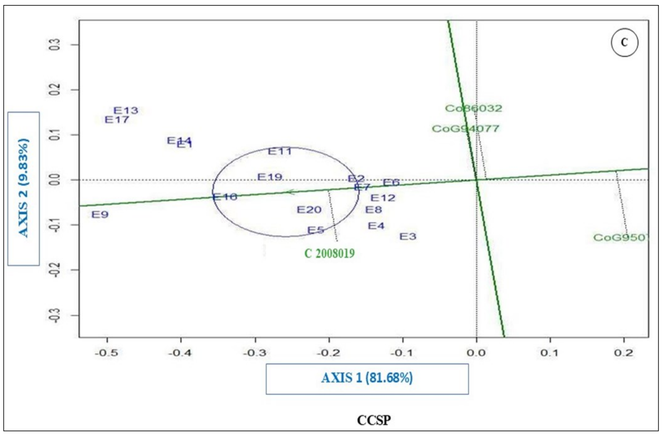

Fig. 4. (C). Ranking biplot of sugarcane genotypes for CCSP where E1, E2, E3…denotes SS environments (E1-E20).

Conclusion

The multi-environment-based experiment under salinity stress helped to identify a better-performing and stable sugarcane genotype G 2008019 for white sugar and jaggery production. The supremacies of this clone under SS are (i) enhanced yield (27.46 to 31.79%) (ii) increased sugar yield (29.35 to 35.09%); (iii) optimum fibre content (13.25%); (iv) good jaggery qualities (juice recovery: 65.36%; juice purity: 91.84% and jaggery recovery: 11.32%). Owing to these advantages this clone can be considered for large-scale seed multiplication, big mill test and proposed for a variety release for cultivation under salinity situations for white sugar and jaggery production.

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Authors' contributions

NAS did conceptualization and methodology of the experiment, collected, analyzed the data and wrote the first draft of the manuscript. RS conducted experiments; reviewed and edited the manuscript. MS conducted experiments and collected the data. JN collected the data. All authors had read and agreed to the published version of the manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None

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