



RESEARCH ARTICLE

# Multivariate analysis, trait association and biochemical profiling of finger millet germplasm for identification of potential genotypes for bioethanol production

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## Abstract

Finger millet is a climate-resilient, nutrient-rich crop well-suited to marginal environments. Recently, its lignocellulosic biomass has gained attention as a potential feedstock for biofuel production. Despite Odisha being a hotspot for landrace diversity, limited studies have evaluated finger millet germplasm for biofuel-relevant traits using multivariate analysis. The present study characterized 30 finger millet genotypes collected from AICRP on small millets, Berhampur, Odisha, during *Khari* 2022. Observations were recorded for 8 morphological traits relevant to biofuel potential. Principal Component Analysis (PCA) and correlation matrix analysis were employed to identify traits contributing most to variability and to group genotypes with superior biofuel-related characteristics. The first three principal components exhibited 76.88 % of the total variation. A statistically significant positive correlation was observed between grain yield per plant and the number of ears per plant at both genotypic ( $r = 0.916$ ) and phenotypic ( $r = 0.866$ ) levels, indicating the potential for indirect selection on the basis of ears per plants to enhance yield. Based on mean performances and PCA results, 6 promising genotypes were selected for further biochemical analysis. Estimation of cellulose, hemicellulose and lignin content revealed that genotype G30-GN-2-2 excelled in both grain yield and bioethanol potential, highlighting its suitability for use in future biofuel-oriented breeding programs. This fills a critical research gap by integrating multivariate tools to identify promising dual-purpose finger millet genotypes. The findings support the potential of finger millet as a renewable biofuel resource and offer a strategic framework for breeding programs aimed at enhancing both yield and bioenergy efficiency.

**Keywords:** correlation; cellulose; hemicellulose; lignin; PCA

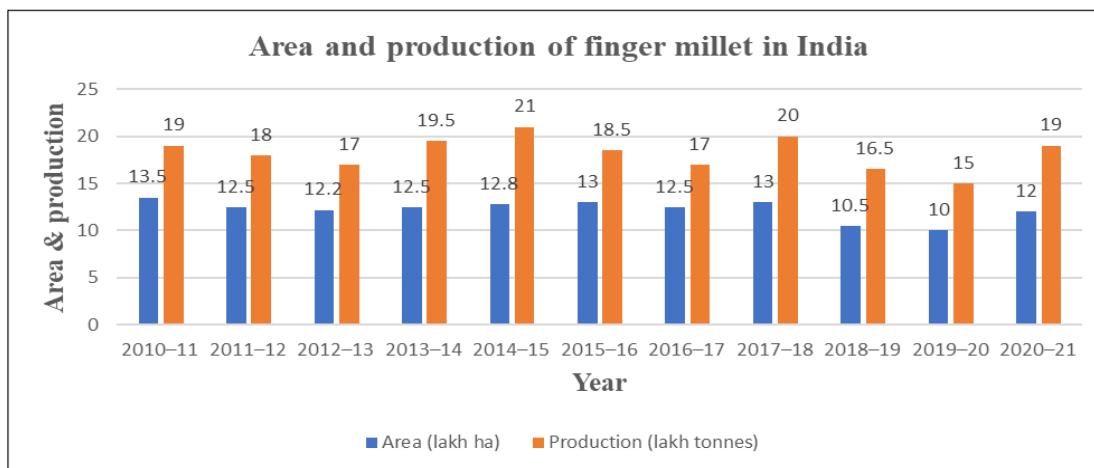
## Introduction

Finger millet (*Eleusine coracana* (L.) Gaertn.), a primitive cereal crop of the Poaceae family, is predominantly cultivated in arid and semi-arid regions of Africa and Asia. It plays a critical role in food and nutritional security due to its high resilience to climatic stress, low input requirements and exceptional nutritional profile, including rich contents of calcium, dietary fibre and essential amino acids (1, 2). In India, finger millet is largely grown in the states of Karnataka, Odisha, Tamil Nadu and Maharashtra, with Odisha being one of the centres of genetic diversity for this crop, harbouring numerous landraces adapted to distinct agroecological zones. The trends in area, production and productivity of finger millet in India have been presented in Fig. 1a & b (3).

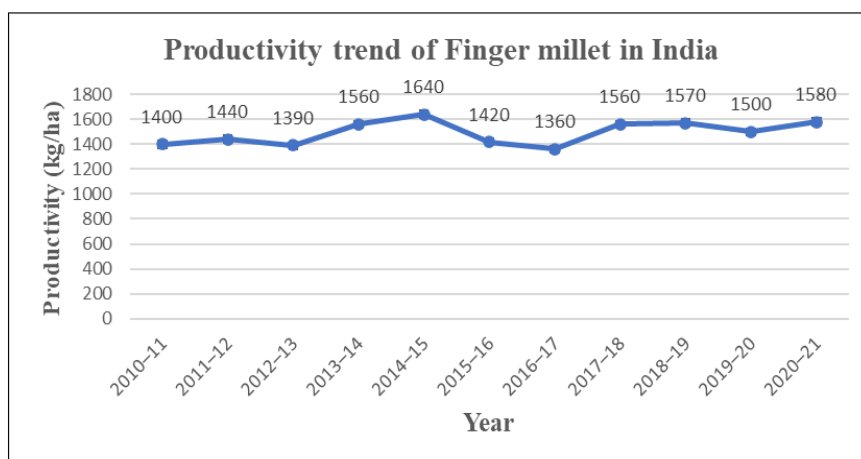
While the importance of finger millet as a staple and health food is well established, its potential as a dual-purpose crop particularly for bioethanol production from the grain is gaining

attention in recent years. The grain starch comprising mainly of starch (> 60 %) with very less lignin content (< 15 %), which could serve as a promising renewable feedstock for first-generation bioethanol production (4 - 6). Ethanol production from finger millet grain presents a sustainable and eco-friendly alternative to fossil fuels, leveraging its high starch content and adaptability to marginal lands. Utilizing finger millet for bioethanol supports energy security while promoting the cultivation of underutilized crops. Its dual-purpose use for food and fuel enhances resource efficiency. Moreover, millet-based bioethanol production contributes to rural economy and reduces greenhouse gas emissions, aligning with global efforts to achieve sustainable energy goals (7).

Despite these advantages, systematic studies on the lignocellulosic potential of finger millet genotypes remain limited. The identification of high-yielding genotypes with favourable biomass composition is crucial for developing varieties that can



**Fig. 1a.** Trends of area and production of finger millet in India.



**Fig. 1b.** Trends of productivity of finger millet in India (low to high).

serve both food and biofuel purposes. Recent advances in multivariate statistical techniques such as PCA, correlation matrix analysis and path coefficient analysis offer powerful tools to dissect complex trait relationships and identify key contributors to variability. These methods facilitate the clustering of genetically diverse germplasm and assist in selecting promising lines with dual-purpose potential (7-9).

Moreover, trait association studies help in understanding the interrelationships between grain yield and its components, which is essential for indirect selection in breeding programs. For instance, traits such as the number of ears per plant, plant height and finger length have been positively associated with grain yield, while biochemical parameters like cellulose and hemicellulose content have been linked to biomass convertibility (10). Therefore, integrating morphological, agronomic and biochemical profiling through multivariate analysis holds significant promise for identifying elite germplasm suitable for dual-purpose utilization.

In this context, the present study was undertaken to evaluate a diverse set of 30 finger millet genotypes collected from the AICRP on Small millets, Berhampur, Odisha. The objectives were to assess morphological and yield-related traits, quantify lignocellulosic components and apply multivariate and correlation-based approaches to identify genotypes with superior bioethanol potential. The outcomes aim to contribute to bioenergy-oriented breeding strategies while supporting sustainable agriculture and renewable energy development.

## Materials and Methods

### Germplasm collection and experimental design

The present study commenced with the collection of finger millet germplasm from AICRP on small millets, Nutri Crops Research Station, Berhampur (Odisha). A total of 30 germplasm accessions were procured from the research station, selected based on their local adaptability, traditional cultivation and representation of the broad genetic variability inherent to Odisha's agro-ecological landscape.

During the *Kharif* season of 2022, the collected germplasm was evaluated under field conditions at the Experimental Block-II (EB-II), Department of Plant Breeding and Genetics, Odisha University of Agriculture and Technology (OUAT), Bhubaneswar, located at 20°52' N latitude and 82°52' E longitude. The experimental layout followed a Randomized Block Design (RBD) with three replications. Each genotype was sown at an inter-row and intra-row spacing of 20 cm × 10 cm, respectively. Standard agronomic practices were adopted throughout the crop growth period to ensure uniform crop stand and expression of genetic potential. Field preparation involved 2-3 ploughings followed by levelling and nutrient management was carried out by applying fertilizers at the rate of 40:20:20 kg/ha N:P:K, along with the incorporation of organic manure. Manual weeding was undertaken at 20 and 40 days after sowing (DAS) and protective irrigation was applied during dry spells to support crop establishment and growth. Harvesting was performed at physiological maturity, varying slightly depending on the genotype and environmental conditions. The list of germplasm under investigation has been presented in Table 1. Observations were recorded for traits contributing to both productivity and suitability for biofuel applications.

**Table 1.** Finger millet germplasm utilized for the study (Source: AICRP on small millets, Nutri-Crops Research Station, Berhampur, Odisha)

Sl. No.	Genotypes	Sl. No.	Genotypes
1	Mut-3	16	G39-GG-2-3
2	Mut-4	17	GPU-157
3	V3-VG-3	18	PR-202
4	V21-VE-3-7	19	P5-400
5	MR-33	20	RES-110
6	BM-107-2	21	RAU-8
7	BM-110-2	22	SRS-2
8	MG-1-3	23	VL-149
9	G5-GG-2-1	24	VL-322
10	G18-GG-2-2	25	VR-708
11	G20-GG-2-4	26	VR-768
12	G22-GE-3-2	27	VR-822
13	G24-GE-3-4	28	VR-849
14	G26-GN-1-2	29	Suvra
15	G30-GN-2-2	30	Nilanchala

### Statistical analysis

The data for various quantitative traits *i.e.*, days to 50 % flowering, days to maturity, plant height (cm), ear length (cm), number of fingers per ear, number of ears per plant, 1000-seed weight (g) and grain yield per plant (g) were observed and recorded from five different plants from each treatment and all the replications. These observations were subjected to statistical analysis to determine the significance of variation among the genotypes. Analysis of variance (ANOVA) was performed following the standard procedure appropriate for a Randomized Block Design (RBD) to assess the presence of statistically significant differences among the treatments. The mean values were further compared using the Least Significant Difference (LSD) test at a 5 % level of significance ( $p < 0.05$ ) to differentiate treatment effects. All statistical computations were carried out using WINDOSTAT software, version 9.30.

### Principle component analysis (PCA)

PCA is a multivariate statistical technique used to convert a set of correlated variables into a smaller set of uncorrelated variables, known as principal components (PCs), thereby reducing data dimensionality while retaining maximum variability. The analysis involves standardization of data, computation of the covariance matrix and extraction of eigenvalues and eigenvectors to identify major axes of variation. PCA enables efficient clustering of genotypes based on their performance, facilitating the identification of superior lines with favourable biofuel-related traits. By minimizing trait redundancy and highlighting key sources of variation, PCA provides a clearer understanding of genotype-trait associations and supports strategic selection in breeding programs. The analysis was performed using WINDOSTAT software version 9.30.

### Correlation analysis

Pearson's correlation coefficients were computed to assess the strength and direction of associations among key agronomic and biofuel-related traits. This analysis helped to identify interrelationships among traits contributing to yield and biofuel

potential. The correlation matrix provided insights into direct and indirect influences, aiding in the selection of trait combinations for effective breeding strategies. The correlation matrix was generated using the KAU GRAPES software.

### Biofuel related biochemical analysis

Cellulose and hemicellulose contents of best performing germplasms were quantified using High-Performance Liquid Chromatography (HPLC) with an Aminex HPX-87H BioRad column (Shimadzu, Japan). Biomass (stover and kernel) samples were subjected to a three-step Soxhlet extraction using hexane, ethanol and water, followed by acid hydrolysis with 72 %  $H_2SO_4$  and autoclaving. The resulting hydrolysate was filtered and analysed *via* HPLC, while acid-insoluble and soluble lignin were estimated using gravimetric methods and UV-Vis spectrophotometry, respectively.

## Results and Discussion

### Analysis of variance

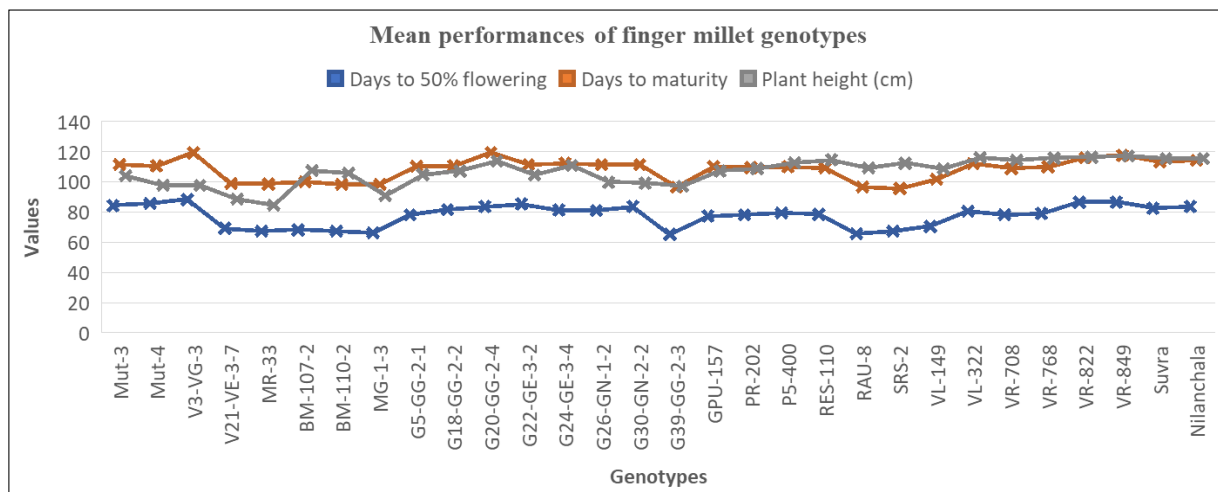
ANOVA indicated highly significant differences among the genotypes which suggested ample amount of genetic variability present among the genotypes for all the traits assessed in the study. These findings confirm that the differences observed for each trait were statistically meaningful, reflecting a wide range of genetic diversity within the evaluated germplasm (Table 2).

### Mean performance of genotypes

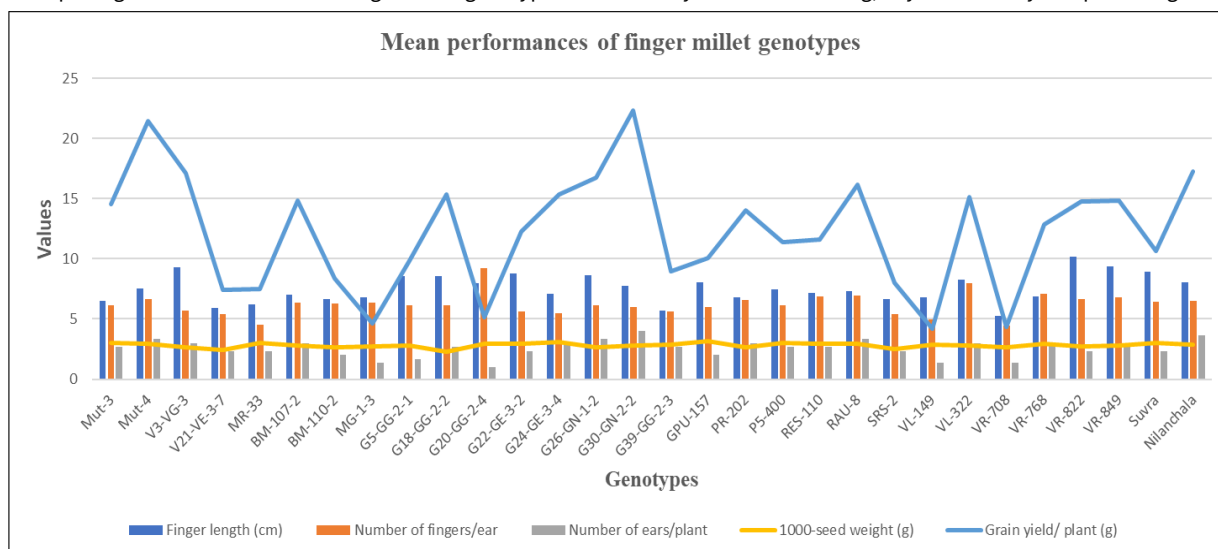
The mean performance analysis of finger millet genotypes revealed significant variability across major agronomic traits, indicating considerable genetic diversity (Fig. 2 a & b). Days to 50 % flowering ranged from 65 to 83 days, with early flowering observed in G39-GG-2-3 and RAU-8 ( $\approx 65$  days). Days to maturity varied from 96 to 112 days, with the shortest duration also recorded in G39-GG-2-3 and RAU-8 ( $\approx 96$  days). Plant height ranged from 79.60 cm to 112.42 cm, while ear length varied between 6.82 cm and 11.44 cm. The number of ears per plant was highest (3-4) in genotypes G30-GN-2-2, Mut-4

**Table 2.** Analysis of variance for various morphological traits of finger millet

Sl. No.	Source	Replication	Treatment	Error
	Degrees of freedom			
1	Days to 50 % flowering	2	29	58
2	Days to maturity	32.643	163.740**	36.665
3	Plant height (cm)	82.242	153.439**	59.529
4	Finger length (cm)	31.307	233.282**	75.999
5	Number of fingers per ear	0.006	4.195**	0.365
6	Number of ears per plant	0.060	2.622**	0.190
7	1000 seed weight (g)	0.108	1.571**	0.036
8	Gran yield per plant (g)	0.015	0.112**	0.040
		0.429	68.866**	0.837



**Fig. 2a.** Morphological characterization of finger millet genotypes based on days to 50 % flowering, days to maturity and plant height.



**Fig. 2b.** Characterization of finger millet genotypes based on yield-related morphological traits: ear length, number of fingers per ear, number of ears per plant, 1000-seed weight and grain yield per plant.

and G26-GN-1-2. Genotypes G20-GG-2-4, VL 322 and VL 768 exhibited the maximum number of fingers per ear (7-9). Grain yield per plant ranged from 7.40 g to 22.31 g, with G30-GN-2-2 (22.31 g), Mut-4 (21.48 g) and Nilanchala (17.27 g) performing best (Table 3). These results underscore the superior yield potential of genotypes Mut-4 and G30-GN-2-2 which demonstrated consistently high mean performance across multiple traits and can be considered promising lines for further improvement programs.

### Genetic parameters

Phenotypic coefficients of variation (PCV) were marginally higher than genotypic coefficients (GCV) for all traits, indicating minimal environmental influence. High PCV and GCV were observed for grain yield per plant (PCV: 39.61 %, GCV: 38.90 %) and number of ears per plant (PCV: 28.96 %, GCV: 27.98 %), reflecting strong genetic control (Fig. 3). Grain yield also recorded the highest heritability (96.44 %) and genetic advance (as percentage of mean 5 %) (78.70 %), followed by number of ears per plant (93.43 %, 55.73 %) and finger length (77.78 %, 27.25 %). High heritability coupled with high genetic advance suggests additive gene action and the effectiveness of selection. These traits present significant scope for genetic improvement through breeding.

### Principal component analysis

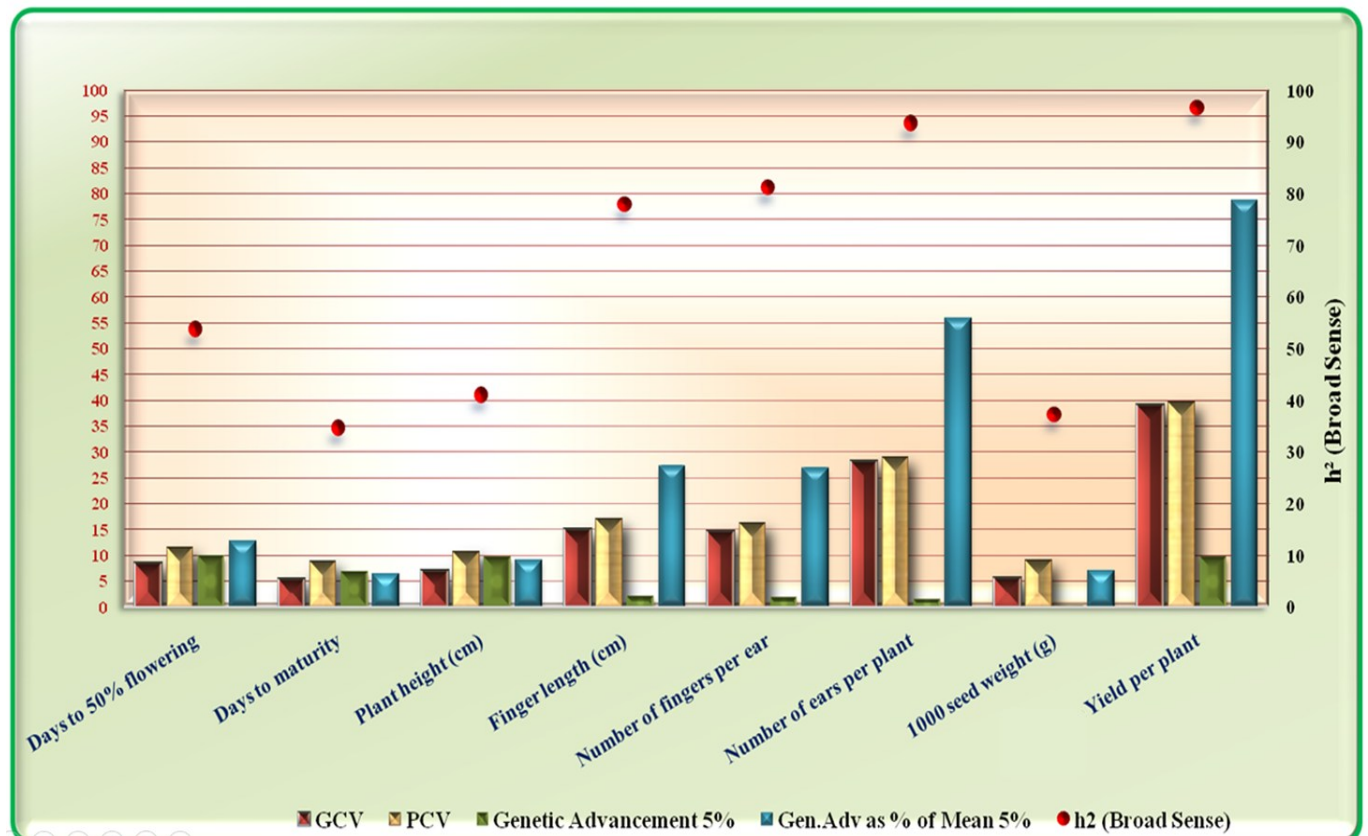
PCA effectively reduced data dimensionality by converting correlated traits into uncorrelated principal components (PCs). Out

of 8 extracted components, three had eigenvalues greater than one and collectively explained 76.88 % of the total variability among the finger millet genotypes. The first component (PC1) alone accounted for 44.59 % of the variation while PC2 and PC3 contributed 19.88 % and 12.40 %, respectively (Table 4). These findings highlight the key traits driving genetic diversity and their relevance for selecting dual purpose genotypes with potential for bioethanol production.

The PCA screen plot (Fig. 4a) and biplot (Fig. 4b-d) generated for the present study was divided into 4 distinct quadrants. Notably, Quadrants 1 and 4 were found to represent genotypes exhibiting superior performance across most of the traits under investigation. This suggests that genotypes located in these quadrants demonstrate favorable trait profiles, making them ideal candidates for further selection, hybridization or pureline development. The germplasm that consistently showed high performance, specifically genotypes [genotype no.] Mut-4 [2], G20-GG-2-4 [11], VR-822 [27], VR-849 [28] and G30-GN-2-2 [15] were predominantly located in Quadrants 1 and 4. This observation further supports the validity of these genotypes as promising selections for breeding purposes. The positioning of these genotypes in the favorable quadrants of the biplot enhances their potential for improving the desired traits, thereby confirming their authenticity as superior genetic material for future breeding programs. The results from the PCA biplot offer valuable insights into the genetic diversity of the studied germplasm and identify

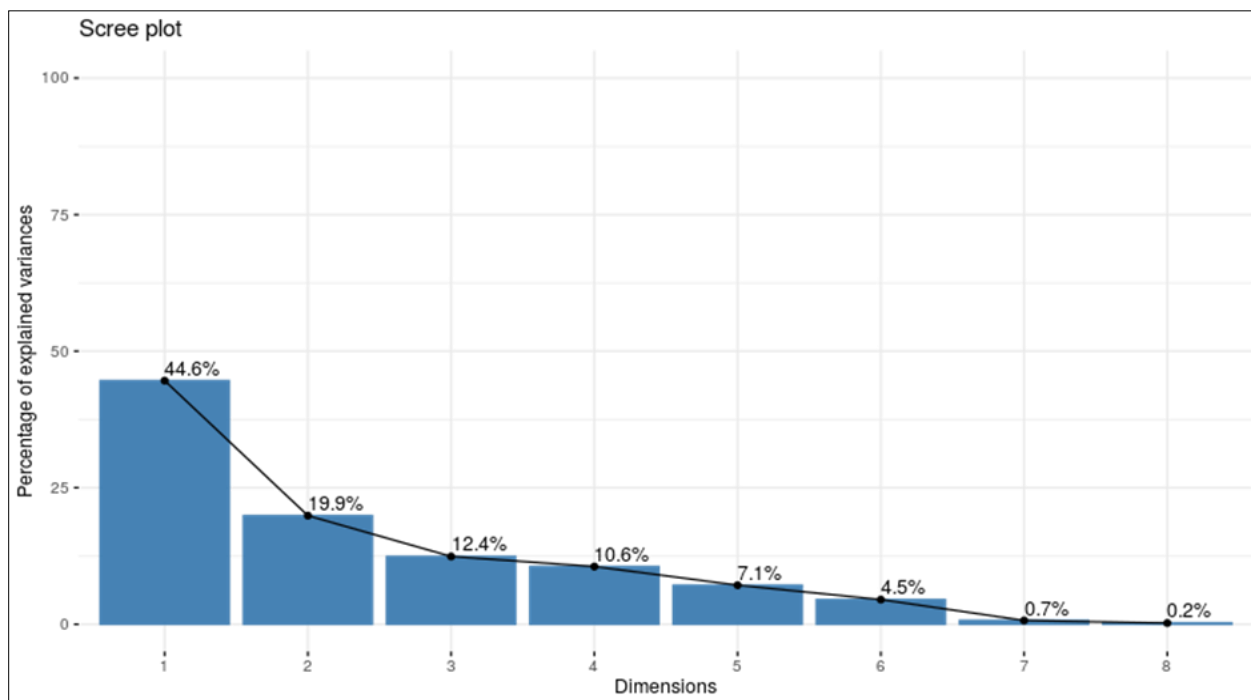
**Table 3.** Mean performance of different genotypes of finger millet for yield and yield contributing traits

Sl. No.	Genotypes	Days to 50 % flowering	Days to maturity	Plant height (cm)	Finger length (cm)	Number of fingers/ears	Number of ears/plants	1000-seed weight (g)	Grain yield/plant (g)
1	Mut-3	84.33	111.67	103.94	6.50	6.13	2.67	3.04	14.56
2	Mut-4	85.67	110.67	97.65	7.50	6.67	3.33	2.94	21.48
3	V3-VG-3	88.33	119.33	97.89	9.30	5.67	3.00	2.64	17.10
4	V21-VE-3-7	69.33	99.00	88.63	5.90	5.40	2.33	2.46	7.43
5	MR-33	67.33	98.67	84.51	6.20	4.53	2.33	3.00	7.53
6	BM-107-2	68.33	100.00	107.41	7.00	6.33	3.00	2.77	14.85
7	BM-110-2	67.33	98.33	105.68	6.67	6.27	2.00	2.67	8.39
8	MG-1-3	66.33	98.33	90.98	6.80	6.33	1.33	2.75	4.60
9	G5-GG-2-1	78.33	110.33	104.91	8.53	6.13	1.67	2.79	9.81
10	G18-GG-2-2	81.67	110.67	107.37	8.57	6.13	2.67	2.29	15.33
11	G20-GG-2-4	83.67	119.67	113.88	7.97	9.20	1.00	2.92	5.16
12	G22-GE-3-2	85.33	111.67	104.70	8.80	5.60	2.33	2.96	12.28
13	G24-GE-3-4	81.33	112.33	111.03	7.07	5.47	3.00	3.06	15.39
14	G26-GN-1-2	81.00	111.67	99.83	8.63	6.13	3.33	2.65	16.74
15	G30-GN-2-2	83.67	111.67	99.23	7.77	6.00	4.00	2.79	22.31
16	G39-GG-2-3	65.33	96.67	97.06	5.67	5.60	2.67	2.86	8.96
17	GPU-157	77.33	110.00	107.32	8.07	6.00	2.00	3.13	10.07
18	PR-202	78.33	109.67	108.85	6.77	6.60	3.00	2.62	14.01
19	P5-400	79.33	110.00	112.75	7.47	6.13	2.67	2.99	11.40
20	RES-110	78.67	109.33	114.55	7.13	6.87	2.67	2.93	11.60
21	RAU-8	65.67	96.67	109.29	7.30	6.93	3.33	2.92	16.16
22	SRS-2	67.33	95.67	112.51	6.63	5.43	2.33	2.51	8.01
23	VL-149	70.67	101.67	108.77	6.77	4.97	1.33	2.90	4.22
24	VL-322	80.67	112.00	116.01	8.30	8.00	3.00	2.82	15.12
25	VR-708	78.33	109.00	114.54	5.23	4.47	1.33	2.63	4.36
26	VR-768	79.00	110.00	115.89	6.90	7.10	3.00	2.96	12.86
27	VR-822	86.67	116.00	116.50	10.20	6.67	2.33	2.74	14.75
28	VR-849	86.67	117.67	117.33	9.40	6.80	3.00	2.82	14.84
29	Suvra	82.67	113.33	115.41	8.90	6.40	2.33	3.02	10.65
30	Nilanchala	83.67	114.33	115.68	8.07	6.53	3.67	2.87	17.27
Mean		77.74	108.2	106.67	7.53	6.22	2.56	2.82	12.24
CV (%)		7.79	7.13	8.17	8.02	7.01	7.42	7.14	7.47
SEm		3.5	4.45	5.03	0.35	0.25	0.11	0.12	0.53
CD at 5 %		9.9	12.61	14.25	0.99	0.71	0.31	0.33	1.49
CD at 1 %		13.17	16.78	18.96	1.31	0.95	0.41	0.44	1.99
Minimum		65.33	95.67	84.51	5.23	4.47	1	2.29	4.22
Maximum		88.33	119.67	117.33	10.2	9.2	4	3.13	22.31

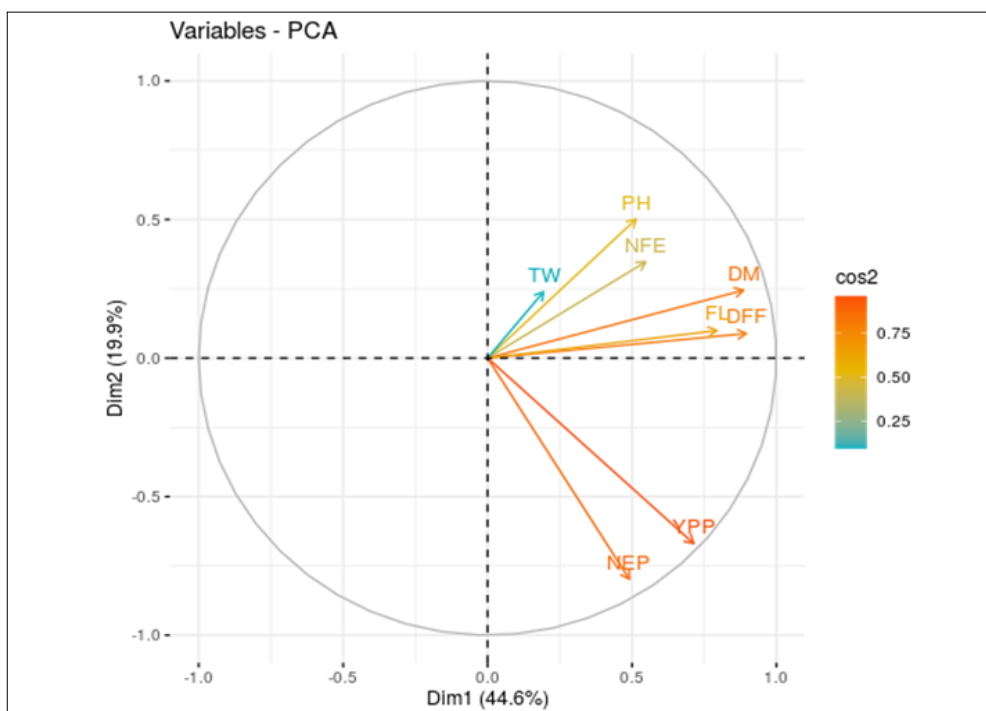
**Fig. 3.** Graphical representation of mean, range and genetic parameter estimates for the traits of finger millet.

**Table 4.** Principal components, eigen values (latent root), per cent variability, cumulative per cent variability and component loading of different characters

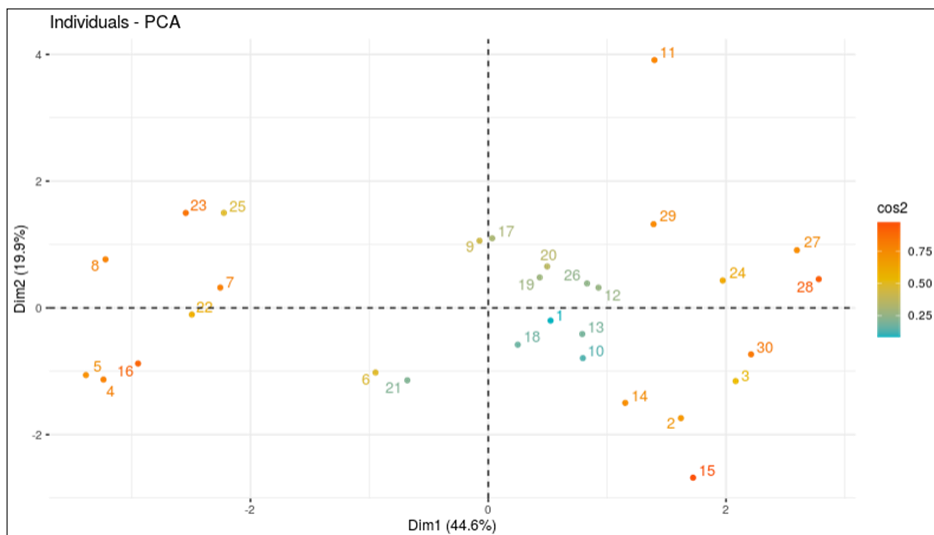
Sl. No.		PC 1	PC 2	PC 3	
		Eigene value (root)	3.568	1.590	0.992
		Percent variability (%)	44.598	19.881	12.404
		Cumulative percent variability (%)	44.598	64.479	76.882
1	Days to 50 % flowering	-0.474	0.07	-0.164	
2	Days to maturity	-0.469	0.194	-0.149	
3	Plant height (cm)	-0.272	0.397	0.105	
4	Finger length (cm)	-0.42	0.079	-0.278	
5	Number of fingers per ear	-0.29	0.275	0.172	
6	Number of ears per plant	-0.26	-0.633	0.221	
7	1000 seed weight (g)	-0.103	0.19	0.88	
8	Gran yield per plant (g)	-0.378	-0.531	0.101	



**Fig. 4a.** Scree plot showing percentage of variances for all the eight PCs studied.

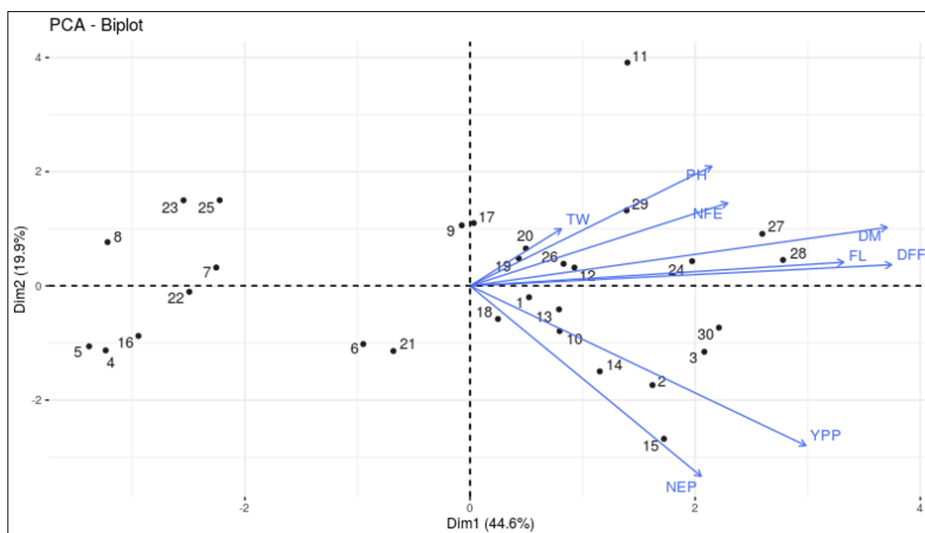


**Fig. 4b.** PCA for eight traits under study in finger millet genotypes (DFF: days to 50 % flowering; DM: days to maturity; PH: plant height; FL: finger length; NFE: number of fingers per ear; NEP: number of ears per plant; TW: test weight; YPP: gran yield per plant).



**Fig. 4c.** PCA for genotypes of finger millet under study.

(The number denotes the code assigned to each genotype. *i.e.*, **1:** Mut-3; **2:** Mut-4; **3:** V3-VG-3; **4:** V21-VE-3-7; **5:** MR-33; **6:** BM-107-2; **7:** BM-110-2; **8:** MG-1-3; **9:** G5-GG-2-1; **10:** G18-GG-2-2; **11:** G20-GG-2-4; **12:** G22-GE-3-2; **13:** G24-GE-3-4; **14:** G26-GN-1-2; **15:** G30-GN-2-2; **16:** G39-GG-2-3; **17:** GPU-157; **18:** PR-202; **19:** P5-400; **20:** RES-110; **21:** RAU-8; **22:** SRS-2; **23:** VL-149; **24:** VL-322; **25:** VR-708; **26:** VR-768; **27:** VR-822; **28:** VR-849; **29:** Suvra; **30:** Nilanchala)



**Fig. 4d.** Genotype by trait biplot presenting the association between PC1 and PC2 for 30 genotypes and the biofuel related morphological traits under study (DFF: Days to 50 % flowering; DM: Days to maturity; PH: Plant height; FL: Finger length; NFE: Number of fingers per ear; NEP: Number of ears per plant; TW: Test weight; YPP: Grain yield per plant) (The number denotes the code assigned to each genotype. *i.e.*, **1:** Mut-3; **2:** Mut-4; **3:** V3-VG-3; **4:** V21-VE-3-7; **5:** MR-33; **6:** BM-107-2; **7:** BM-110-2; **8:** MG-1-3; **9:** G5-GG-2-1; **10:** G18-GG-2-2; **11:** G20-GG-2-4; **12:** G22-GE-3-2; **13:** G24-GE-3-4; **14:** G26-GN-1-2; **15:** G30-GN-2-2; **16:** G39-GG-2-3; **17:** GPU-157; **18:** PR-202; **19:** P5-400; **20:** RES-110; **21:** RAU-8; **22:** SRS-2; **23:** VL-149; **24:** VL-322; **25:** VR-708; **26:** VR-768; **27:** VR-822; **28:** VR-849; **29:** Suvra; **30:** Nilanchala).

key genotypes that could contribute to the enhancement of targeted traits through selection and hybridization efforts.

### Correlation analysis

Correlation analysis revealed that genotypic correlation coefficients were consistently higher than phenotypic ones, indicating a strong genetic basis for trait associations. Grain yield per plant exhibited a highly significant positive correlation with number of ears per plant at both genotypic ( $r$ ) (0.916) and phenotypic ( $r$ ) (0.865) levels, identifying it as a key contributor to yield improvement (Table 5). Significant positive correlations were also observed between grain yield and traits such as days to 50 % flowering, days to maturity and finger length, suggesting their utility as indirect selection indices. Days to 50 % flowering was strongly correlated with days to maturity ( $r$  0.567,  $r^2$  0.992), finger length ( $r$  0.512,  $r^2$  0.812) and plant height ( $r$  0.212,  $r^2$  0.556), reflecting interconnected growth and yield traits (Table 5). These relationships highlight the potential for

simultaneous improvement of yield and its components through targeted selection.

### Biofuel related biochemical analysis

Out of the 30 genotypes evaluated for various traits related to yield and biofuel production efficiency, 6 genotypes were identified as the most promising based on their superior mean performance and PCA analysis. These genotypes namely Mut-4, G20-GG-2-4, VR-822, VR-849 and G30-GN-2-2 and Nilanchala demonstrated consistent high performance across the traits under study, making them suitable candidates for further analysis and breeding efforts aimed at improving biofuel production. To assess their potential for biofuel-related applications, grain samples of these 6 selected genotypes were further analyzed for key biochemical traits associated with biofuel production, specifically the percentages of cellulose, hemicellulose and lignin in the grain. The biofuel related biochemical analysis revealed considerable variation in

**Table 5.** Estimates of phenotypic and genotypic correlation coefficients among eight quantitative traits in finger millet

Characters	Days to 50 % flowering	Days to maturity	Plant height	Finger length	No. of fingers per ear	No. of ear per plant	1000-seed weight	Grain yield per plant
Days to 50 % flowering <sup>P</sup>	1	0.567**	0.212*	0.512**	0.252*	0.218*	0.139	0.433**
Days to 50 % flowering <sup>G</sup>	1	0.922**	0.556**	0.812**	0.348**	0.320**	0.139	0.622**
Days to maturity <sup>P</sup>		1	0.243*	0.473**	0.287**	0.132	0.125	0.314**
Days to maturity <sup>G</sup>		1	0.701**	0.927**	0.546**	0.204	0.243*	0.539**
Plant height <sup>P</sup>			1	0.272**	0.326**	0.022	0.186	0.077
Plant height <sup>G</sup>			1	0.382**	0.550**	0.012	0.096	0.081
Ear length <sup>P</sup>				1	0.321**	0.192	0.031	0.420**
Ear length <sup>G</sup>				1	0.436**	0.217*	0.018	0.489**
No. of fingers per ear <sup>P</sup>					1	0.076	0.144	0.197
No. of fingers per ear <sup>G</sup>					1	0.096	0.153	0.234*
No. of ears per plant <sup>P</sup>						1	0.020	0.865**
No. of ears per plant <sup>G</sup>						1	0.013	0.916**
1000-seed weight <sup>P</sup>							1	0.021
1000-seed weight <sup>G</sup>							1	0.046
Grain yield per plant <sup>P</sup>								1
Grain yield per plant <sup>G</sup>								1

\* Significant at 5 % level; \*\* Significant at 1 % level; <sup>P</sup>: Phenotypic correlation, <sup>G</sup>: Genotypic correlation

lignocellulosic composition relevant to biofuel potential (Table 6). Cellulose content ranged from 60.12 % (G20-GG-2-4) to 67.27 % (G30-GN-2-2), with VR-822 (66.26 %) and Mut-4 (62.9 %) also recording high values. Hemicellulose content varied between 2.05 % (VR-822) and 4.09 % (G20-GG-2-4). Lignin content was lowest in G20-GG-2-4 (11.85 %) and highest in VR-822 (14.50 %). Extractives ranged from 13.13 % (G30-GN-2-2) to 16.05 % (G20-GG-2-4). Among all genotypes, G30-GN-2-2, VR-822 and Mut-4 demonstrated favourable profiles for biofuel-related biochemical traits *i.e.*, higher cellulose and hemicellulose content and lower lignin content. These biochemical constituents are critical for determining the efficiency of biofuel conversion, as they directly influence the process of biomass degradation and subsequent biofuel yield. By focusing on these genotypes, the most favorable biochemical compositions for biofuel production are aimed to be identified, which could ultimately lead to the development of high-yielding, biofuel-efficient varieties for sustainable energy production.

The present study provides a comprehensive assessment of genetic variability, trait associations and biochemical composition in 30 finger millet genotypes, with the primary objective of identifying promising dual-purpose lines suitable for both grain yield and bioethanol production. The findings offer critical insights into the potential of finger millet as a sustainable bioenergy crop while preserving its role in food security. The highly significant differences observed among genotypes cross all studied traits in the ANOVA clearly indicate the presence of substantial genetic variability within the evaluated germplasm. Such diversity is essential for effective selection in breeding programs and supports the feasibility of identifying genotypes with superior performance for biofuel-related agronomic traits (11, 12). The mean performance analysis further highlighted the presence of considerable phenotypic variability across traits like flowering time, maturity, plant height, ear length and grain yield (13, 14). Early flowering and maturity were observed in genotypes G39-GG-2-3 and RAU-8, making them potentially valuable for short-duration cropping systems or environments with

terminal drought stress. Meanwhile, genotypes such as G30-GN-2-2, Mut-4 and Nilanchala exhibited the highest grain yield per plant, underscoring their productivity potential. Two important yield-contributing traits *i.e.*, number of ears per plant and number of fingers per ear exhibited high performances in genotypes like Mut-4 and VL 768, which enhances their utility as parents in breeding programmes.

The estimation of genetic parameters revealed that PCV values were only marginally higher than their respective GCV values, suggesting minimal environmental influence and reinforcing the heritable nature of the traits studied. Traits such as grain yield per plant and number of ears per plant recorded high PCV and GCV, along with high heritability and genetic advance. These findings indicate that these traits are under strong additive genetic control and thus, direct selection could be effective in improving them. The presence of high genetic gain further substantiates their value as key selection criteria for yield improvement (15).

PCA served as a robust multivariate tool to dissect the complex inter-relationships among traits and to identify those contributing most significantly to variation. Out of 8 principal components extracted, the first three accounted for 76.88 % of the total variability, with PC1 alone explaining 44.59 %. Most of the traits had strong loadings on PC1, indicating their combined influence on genetic diversity. These traits are especially relevant for dual-purpose selection, where both biomass quality and agronomic performance are targeted (16, 17). The PCA biplot provided a graphical illustration of genotype distribution across trait space, with quadrants 1 and 4 highlighting genotypes that exhibited favourable trait combinations. Notably, Mut-4, G20-GG-2-4, G30-GN-2-2, VR-822 and VR-849 consistently appeared in these quadrants, validating their potential as elite selections for breeding programs. Their placement reflects superior performance across both yield and biomass quality-related traits, reinforcing their dual-purpose nature (11).

**Table 6.** Biofuel related biochemical analysis of finger millet grain samples

S. No.	Genotypes	Cellulose %	Hemicellulose %	Lignin %	Extractives %
1	Mut-4	62.9	3.31	12.76	15.55
2	G20-GG-2-4	60.12	4.09	11.85	16.05
3	G30-GN-2-2	67.27	2.29	13.96	13.13
4	VR-849	61.25	3.89	12.25	15.95
5	VR-822	66.26	2.05	14.5	13.17
6	Nilanchal	61.12	3.89	12.55	15.05

The correlation analysis offered further insights into trait interdependencies. Grain yield per plant showed a strong and significant positive correlation with number of ears per plant at both genotypic and phenotypic levels, marking it as a key determinant of yield performance (18, 19). Additional positive associations were observed between grain yield and traits such as days to 50 % flowering, days to maturity and finger length, suggesting that these can be used as indirect selection criteria. The strong correlation between days to flowering and traits like plant height and ear length reflects the coordinated expression of developmental and reproductive traits, providing opportunities for synchronized selection in early-generation breeding (20-22).

To validate the dual-purpose potential, a biochemical evaluation was conducted for six genotypes identified as top performers through agronomic and multivariate analyses. From the 30 germplasm under study 6 best performing lines were selected based on their mean performances and PCA analysis and were subjected to biofuel related biochemical characterization. The biochemical profiling revealed substantial variation in lignocellulosic traits, which are pivotal for assessing bioethanol production efficiency (23). Cellulose content which is a key determinant of fermentable sugar yield was highest in G30-GN-2-2 followed closely by VR-822 and Mut-4. While high cellulose content is desirable, lignin content must remain moderate, as excessive lignin impedes enzymatic hydrolysis during biomass conversion (10, 24). Genotype G20-GG-2-4 exhibited the lowest lignin content, making it highly suitable for biofuel processing. Hemicellulose variation was relatively narrow but still meaningful, with G20-GG-2-4 displaying the highest value, potentially enhancing saccharification efficiency. The extractive content, which can affect downstream processing, was within acceptable limits *i.e.*, 10-15% across genotypes.

Genotypes G30-GN-2-2, VR-822, Mut-4 and G20-GG-2-4 emerged as promising candidates for dual-purpose use, demonstrating a desirable combination of high grain yield and favorable lignocellulosic composition. Among these, G30-GN-2-2 stood out, exhibiting superior performance in both grain productivity and bioethanol potential, underscoring its suitability for future biofuel-oriented breeding programs. These results underscore the potential of finger millet in integrated food and energy systems. Furthermore, the ability to pinpoint such promising genotypes highlights the effectiveness of integrating classical phenotyping with biochemical profiling and multivariate statistical analysis for targeted crop improvement.

## Conclusion

The present study successfully identified significant genetic variability among 30 finger millet genotypes for both morphological and biofuel-related traits through comprehensive multivariate, correlation and biochemical analyses. Genotypes such as G30-GN-2-2, VR-822, Mut-4 and G20-GG-2-4 demonstrated superior performance in grain yield and favourable lignocellulosic profiles, establishing their potential as dual-purpose lines suitable for food and bioethanol production. Among them, G30-GN-2-2 distinctly outperformed others, demonstrating exceptional grain yield and high bioethanol potential, thereby reinforcing its potential as a key candidate for future breeding programs focused on biofuel development. The observed strong heritability, high genetic advance and trait associations further validate their use in targeted

breeding programs. The integration of agronomic, genetic and biochemical data enabled the identification of elite finger millet genotypes with potential for both food security and sustainable biofuel production. These dual-purpose genotypes represent valuable genetic resources for future breeding programs aimed at enhancing productivity and renewable energy potential in marginal cropping systems. Looking ahead, future research should focus on validating these genotypes under multi-location trials, exploring their biomass conversion efficiency through enzymatic saccharification studies and incorporating molecular markers linked to lignocellulosic traits to accelerate the development of climate-resilient, energy-efficient finger millet cultivars.

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## Authors' contributions

Conceptualization was done by PM, MKM and TKM. Material preparation, conduct of experiment, data collection and analysis were performed by PM and RRKR. The first draft of the manuscript was written by PM and checked by DL. Statistical analysis was done by SRP, MRM and KCP. The manuscript was reviewed, read and edited with significant contributions by DL and TKM. All authors read and approved the final 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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