







Principal component analysis (PCA) and genetic diversity studies for discrimination of genotypes and yield parameters in ICRISAT Pearl Millet [Pennisetum glaucum (L.) R. Br.] accessions

Aananthi N1*, A Sheeba2, R Annamalai3, D Shoba4 & M Arumugam Pillai3

¹Department of Plant Breeding and Genetics, Agricultural College and Research Institute, Madurai 625 104, Tamil Nadu, India ²ICAR - Krishi Vigyan Kendra, Aruppukottai 626 107, Tamil Nadu, India

³Department of Genetics and Plant Breeding, VOC Agricultural College and Research Institute, Killikulam, Vallanadu 628 252, Tamil Nadu, India ⁴Agricultural Research Station, Tamil Nadu Agricultural University, Kovilpatti 628 501, Tamil Nadu, India

*Correspondence email - aananthi.n@tnau.ac.in

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Abstract

PCA and cluster analysis were used to assess 50 pearl millet genotypes for 26 attributes to identify associations among individuals as well as their traits and patterns of variation. Among 26 PCs (Principal Components), eight unveiled eigenvalues greater than one, which accounted for 78.29 % of the total variability among the traits. The first component (PC1) had a great maximum variability of about 18.53 % with the highest eigenvalue. It was observed that PC1 revealed maximum variation in comparison with other 25 PCs, whereas PC2 to PC8 showed gradual reduction in variability with values 17.38, 11.71, 8.17, 7.37, 5.63, 5.0 and 4.49 percent respectively. Therefore, selecting lines and traits based on PC1 would be beneficial. The factor loading of PCs exhibited that PC1 accounted for maximum variability for traits like NTrPP (number of tillers per plant), LAI (leaf area index) and GY (grain yield) per plant. PC2 counted for 17.38 % of the total variance and exhibited the greatest variability for number of leaves per plant (NLPP). PC3 showed 11.71 % variability and taken maximum variability for leaf:stem ratio (LSR). PC4 exhibited 8.17 % of variability and possessed maximum variability for plant height (PtH), leaf width (LfW), flag leaf width (FgLW) and stem girth (SG) which pointed out the huge impact in the total variation of the genotypes. Cluster analysis recorded cluster III had desirable mean values for most of the traits studied. The genotype, viz. IP 8327 from cluster III would be used as parent for pearl millet improvement program.

Keywords: genetic diversity; grain and fodder yield; principal component analysis; pearl millet

Introduction

Pearl millet (P. glaucum (L.) R. Br.) is distinctly a cross-pollinated crop due to its protogynous flowering. Globally it ranks as the sixth most important cereal crop and is grown extensively by farmers with less resources across the Indian subcontinent and Africa that are arid or semi-arid (1). It is a cereal dietary grain having nutrient-dense, commonly used as feed and forage, fuel, construction materials as well as a source of biofuel (2). In India, pearl millet ranks fourth among cereals with an area of 9.4 million ha with an annual production of 10.1 million tones with an average productivity of 1069 kg/ ha (3). Accounting for 43.3 % of the world's land area and 42 % of global production, it is extensively grown across various regions. With a strong adaptability to challenging environmental conditions, it also showcases substantial genetic diversity (4). In contrast to other grains like rice, wheat, maize, sorghum and barley it may come up in harsh climates and offer farmers financial advantages (5). In Tamil Nadu, it spans 59956 hectares, yielding 1.46 lakh t with a productivity of 2437 kg / hectare (6). Since crops are cultivated in

low input conditions with marginal lands, the productivity is less than 1374 kg / ha (7). The increasing demand for pearl millet is attributed to its adaptability to different climatic conditions and its exceptional nutritional benefits (8).

GYs are low in pearl millet predominantly since the crop is frequently cultivated under low input conditions of subsistence cultivation in marginal environments. Breeders focus on improving yield by using advanced breeding techniques for adapting to changing climatic conditions, which involves discovering new abiotic and biotic resistance genes from unexplored genetic diversity. In pearl millet breeding, hybrids produced from diverse genotypes exhibit greater heterosis and produce novel high-yielding varieties. Hence, it is necessary to grow high yielding cultivars through heterosis breeding as a potential way to deal with increment in the pearl millet production in India. GY is an unpredictable character and relies upon number of component characters, which are quantitatively inherited. Thus, before starting any breeding programmes, intensive information on the nature and greatness of genetic

variability and degree of relationship between yield and different components is extremely essential. Having access to the fullest genetic diversity among parents is vital for creating superior cultivars. Diverse germplasms are essential for developing new varieties in crop improvement. Using an appropriate divergent scale to classify genotypes is vital for producing superior progeny, as yield is a multifaceted trait affected by several factors and environmental conditions. PCA is a method used to identify and reduce traits, thereby optimizing the efficiency of the selection process (9). It is a nonparametric, multivariate approach used for evaluation of genetic variation that aids in identification of characters which contributes to classifying genotypes into separate groups based on common traits or characteristics (10). Hence, the present investigation was carried out on ICRISAT pearl millet accessions with an objective to examine yield and yieldrelated traits, uncovering the maximum variability within the overall variability, to classify pearl millet germplasm accessions based on various characteristics and their relationships.

Materials and Methods

The field experiment was performed on the Killikulam Agricultural College and Research Institute's farm. The experiment was conducted in a Randomized Block Design with two replications (Fig 1.). During kharif 2019, seeds of fifty pearl millet entries (Table 1) received from ICRISAT, Hyderabad were directly sown in plots maintaining a row spacing of 45 cm and a plant spacing of 15 cm. The field size adopted was 3 × 3 m. Each plot is comprised of six rows. Normal agronomic practices and plant protection measures were followed during crop period. For grin yield (GY) study, the crop was harvested at maturity stage; conversely, for green fodder yield (GFY) and forage quality, the plant was harvested at days to 50 percent flowering stage (DFyF). Twentysix agronomic attributes, namely DFyF, plant height (PtH), peduncle length (PeL), number of nodes per plant (NNdPP), Number of Tillers per plant (NTrPP), number of productive tillers per plant (NPrTP), internode length (InL), leaf length (LfL), Leaf width (LfW), Leaf Area Index (LAI), flag leaf length (FgLL), flag leaf

Table 1. Pearl millet genotypes used in this study collected from ICRISAT, Hyderabad

Sl. No	Genotypes	Sl. No	Genotypes
1	IP3080	26	IP15320
2	IP3476	27	IP15321
3	IP3604	28	IP15322
4	IP3613	29	IP15341
5	IP3616	30	IP15342
6	IP3625	31	IP15343
7	IP3627	32	IP15344
8	IP3628	33	IP15348
9	IP3636	34	IP15351
10	IP3645	35	IP15369
11	IP3663	36	IP15710
12	IP3665	37	IP17428
13	IP5836	38	IP20273
14	IP8327	39	IP20339
15	IP10437	40	IP20346
16	IP11839	41	IP20347
17	IP11840	42	IP20348
18	IP15257	43	IP20350
19	IP15285	44	IP20379
20	IP15288	45	IP20539
21	IP15290	46	IP20540
22	IP15301	47	IP20585
23	IP15302	48	IP21226
24	IP15306	49	PT4806
25	IP15307	50	PT4181



Fig. 1. Field view.

width (FgLW), flag leaf area (FgLA), Number of leaves per plant (NLPP), Stem girth (SG), spike length (SpL), leaf:stem ratio (LSR), seed weight per spike (SWS), 1000 grain weight (TGW), GY, GFY, dry fodder yield (DFY), ash content (AC), brix content (BC), crude protein (CP) and crude fibre (CF) were studied.

Observations were made from ten randomly selected plants in all replications, in accordance with standard procedures. The data were assessed using the multivariate approach to genetic divergence employing D2 statistics (1). The genotypes were grouped using Tocher's approach and the average inter and intra cluster D² mean values were assessed according to the method specified (11). PCA was carried out to identify the patterns of variation (12, 13). The PCs with eigenvalues exceeding one were selected (14). The correlations between the original traits and the associated PCs were computed. Statistical analysis was carried out using the software STAR 2.0. The PCA was computed using the equation, PC1= $P\Sigma 1$ ajXj; where, ajXj = Linear coefficient - Eigen vectors. The agglomerative clustering method using the Euclidean distance measure was used for cluster analysis. Statistical software like Rstudio, Meta R and GENRES was used to statistically analyze the mean values of each genotype.

Results and Discussion

Assessment of genetic divergence

The analysis of variance (ANOVA) showed that the mean sum of squares resulting from genotypes was significantly high for all the traits studied, indicating the existence of variability (Table 2). Tocher's approach was used to group the 50 genotypes into 11 clusters (Table 3). Among 11 clusters, cluster I (30), cluster II (8) and cluster VIII (3 genotypes) are polygenotypic and the remaining clusters were monogenotypic revealing distinctiveness in genes. Solitary clusters showed that these genotypes exhibit considerable genetic diversity for several traits and might not have the parallelism between genetic diversity and geographical diversity. These results were comparable to earlier reports where fifty germplasms were grouped into sixteen clusters in pearl millet (15, 16). The intracluster distance of clusters II, VIII and I exhibited maximum divergence with descending order. To obtain a wide range of variation among the segregants, hybridization programs could make use of genotypes from distinct clusters that are separated by a high statistical distance. Inter cluster distance showed greater genetic divergence between cluster XI and IV followed by cluster XI and I, cluster XI and VII and cluster XI and VIII (Table 4). Hence, the genotypes of cluster XI could be

Table 2. ANOVA for yield and yield contributing traits

-	Traits	Mean sum o	f squares
Replic	ations (df = 1)	Genotypes (df = 49)	Error (df = 49)
DFyF	17.98	47.63**	8.61
PtH	11.80	7066.77**	30.38
PeL	35.57	239.2**	3.49
NNdPP	29.94	7942.66**	16.63
FgLL	0.23	110.9**	4.06
FgLW	0.02	1.36**	0.05
FgLA	12.24	440.48**	17.73
NTrPP	9.05	30.52**	2.35
NPrTP	0.80	27.04**	0.86
SpL)	0.11	107.49**	26.81
SWS	20.26	67.3**	1.46
TGW	18.50	199.94**	0.36
RL	29.31	72.36**	33.16
GY	49.32	4792.36**	13.95

^{**} Significance at 1% level, * Significance at 5% level. TGW = Test grain weight, ,

Table 3. Distribution of 50 pearl millet genotypes in D² cluster for grain yield and other component traits

Clusters	Number of genotypes	Name of the genotypes
I	30	IP3604, IP3636, IP3625, IP15288, IP21226, IP3663, IP15301, IP20350, IP15306, IP3627, IP15322, IP3616, IP20273, IP15342, IP3080, IP20339, IP15321, IP20348, IP20379, IP3476, IP15710, IP15320, IP17428, IP20585, IP5836, IP3665, IP15369, IP20346, PT4881, IP15285
II	8	IP10437, IP11839, IP15348, IP15307, IP15290, IP20347, IP15341, IP20540
Ш	1	IP3613
IV	1	IP15257
V	1	IP3645
VI	1	IP3628
VII	1	PT4806
VIII	3	IP11840, IP15351, IP15343
IX	1	IP15344
Χ	1	IP20539
ΧI	1	IP8327

Table 4. Inter cluster and intra cluster distance for 50 pearl millet genotypes based on grain yield traits

Clusters	ı	II	III	IV	V	VI	VII	VIII	IX	Х	ΧI
	26.12	35.16	34.63	31.86	37.18	34.48	35.34	63.8	44.79	43.27	106.36
II		29.91	35.64	40.32	40.41	40.99	46.77	45.33	41.38	36.15	93.11
III			0	43.72	19.93*	25.44	41.31	64.96	41.61	33.24	84.28
IV				0	51.29	35.32	40.33	61.01	48.75	57.64	110.45*
V					0	32.95	36.83	73.66	34.4	31.72	86.43
VI						0	36.91	68.99	44.44	50.83	89.05
VII							0	76.91	34.45	55.33	105.03
VIII								29.51	67.87	62.86	100.55
IX									0	42.3	83.97
Χ										0	89.97
XI											0

used in hybridization with genotypes of clusters viz., I, VII and VIII for getting superior heterotic Fis and transgressive segregants with substantial amount of genetic variability in the successive generations. From these results, the genotypes from cluster I (IP3604, IP3636, IP3625, IP15288, IP21226, IP3663, IP15301, IP20350, IP15306, IP3627, IP15322, IP3616, IP20273, IP15342, IP3080, IP20339, IP15321, IP20348, IP20379, IP3476, IP15710, IP15320, IP17428, IP20585, IP5836, IP3665, IP15369, IP20346, PT4881, IP15285) and genotypes from cluster VII (PT4806) and VIII (IP11840, IP15351, IP15343) could be selected and used as donors for hybridization with genotype IP8327 in the cluster XI for getting superior genotypes. Therefore, to create high-yielding cultivars, hybridization programs could use the genotypes of the most varied clusters as parents. Similar views were expressed in kodo millet (17) and pearl millet (18-20). In this study, genotypes from various regions were put together into a single cluster rather than being grouped according to their place of origin or geographic location which is in line with the previous findings (19, 25).

While considering the cluster mean values, cluster VIII exhibited higher values for NNdPP, NTrPP, NPrTP, FgLW and FgLA indicating the genotypes of this cluster viz., IP11840, IP15351and IP15343 promised its contribution in developing high GY coupled with high GFY. Cluster IX with the genotype IP15344 exerted higher cluster mean for DFyF and FgLL which could be used to develop an early genotype with increased grain yield. Cluster IV with the genotype IP15257 expressed higher cluster mean value for root lodging (RL). Cluster X with the genotype IP20539 contributed higher cluster mean for PtH and could be useful in bringing high GY coupled with fodder yield (Table 5). Previous findings for SpL and GY per plant (19 and 21) were also corroborated with the present findings. This was supported by (22) for SWS, TGW. Earlier reports (23) revealed the similar findings for NNdPP and NTrPP (24) forFgLW, FgLA and NPrTPP (25) and for RL, DFyF and PtH. All the maximum and minimum values for the cluster mean were grouped in different clusters. Recombination breeding between genotypes of different clusters could be taken up to unite the desirable

 Table 5. Cluster mean for 50 pearl millet genotypes based on grain yield traits

Traits	DFyF	PŧĦ	PeL	Adpun	FgLL	FgLW	FgLA	NTrPP	NPrTP	SpL	SWS	TGW	RL	λS
_	57.59	194.4	14.35	60.31	23.77	27.69	46.32	7.91	7.29	21.25	13.99	9.51	14.94	97.16
=	53.38	238.64	19.7	139.42	23.18	28.95	48.02	12.03	11.5	22.72	11.26	8.99	14.64	128.09
Ξ	56	206.82	39.7	56.6	19.75	14.3	19.1	13.35	12.9	19.01	17.05	12.13	10.26	219.94
2	62	120.33	3.52	71.7	25.27	16	27.5	7.6	8.6	20.47	15.02	11.07	40.26	129.1
>	28	256.21	45.11	37	27.42	36.5	68.05	14	13.85	27.92	13.66	9.39	10.26	187.01
5	57.5	145.34	6.99	32.4	20.32	23.8	32.46	10.45	12.3	25.02	19.85	8.2	10.26	242.01
5	54.9	131.45	18.45	30.65	39.29	48.5	129.55	8.9	7.2	18.44	14.16	12.95	20.26	101.43
III	57.67	202.76	13.99	275.1	22.98	21.12	32.06	17.72	15.95	22.22	9.19	7.85	16.93	144.52
×	66.5	274.01	26.01	81.2	40.41	43.1	118.4	7.7	9.2	23.67	17.18	11.15	35.26	157.6
*	54.5	382.3	39.6	93.5	9.54	20.9	13.55	7.5	8.45	12.18	14.85	9.72	10.26	125.55
₹	92	345.4	39.3	157.8	37.17	33.8	84.72	14.7	12.3	58.6	38.59	19.29	10.26	473.76
Mean	58.46	227.06	24.25	94.15	26.28	28.61	56.35	10.89	10.87	24.68	16.80	10.93	17.60	182.38
Min	53.28	120.33	3.52	30.65	9.54	14.3	13.55	6.8	7.2	12.18	9.19	7.85	10.26	97.16
Мах	66.5	382.3	45.11	275.1	40.41	48.5	129.55	17.72	15.95	58.6	38.59	19.29	40.26	473.76

characters of different accessions. Recombination breeding among genotypes from dissimilar clusters can be pursued to combine the desirable attributes of various accessions. Thirtyone pearl millet genotypes were categorized into five clusters where the average intra cluster distance was 0.00 for cluster V while 51.37 for cluster II (26). Cluster II had the greatest intracluster distance (382.29), followed by cluster III (41.84) (20) and it was concluded that the selection of parents and breeding choices mainly rely on the contribution of traits to divergence. The dominance of the NNdPP was evident in the current study. The ascendancy of NNdPP (35.10) followed by GY (18.61) and PtH (15.10) in divergence determined the clusters VIII with genotypes (IP11840, IP15351, IP15343), cluster XI (IP8327) and cluster X (IP 20539) (Table 6). As the genotypes in these clusters possessed high mean values for most of the agronomic traits those genotypes could be a source of genes for yield improvement.

PCA

PCA is a multivariate analysis that helps to examine relationships among various quantitative traits in studies of variance and numeral classification (27). In this study, PCA was performed based on GY, fodder yield and key characters linked to quality in 50 ICRISAT pearl millet accessions. Among the 26 traits analyzed, the initial eight PCs had eigenvalues greater than 1.00, accounting for approximately 78.29 of the total cumulative variability among the traits (Fig. 2, Table 7). Every character under study made a constructive contribution to PC1, except for AC (%), BC (%) and CP (%). PC1 recorded 4.82 eigen value and 18.53 % of total variability. The traits like PtH, LSR, total fresh yield, total dry yield, CP %, carbohydrates % and ash % were the critical components explained 43.159 of the total variability and 3.492 of the eigen value (28). It was observed that, the progressive decrease in variability was found in PC2, PC3, PC4, PC5, PC6, PC7 and PC8 with values of 17.38, 11.71, 8.17, 7.37, 5.63, 5.0 and 4.49 percent respectively. A gradual decline in variability was observed from PC2 to PC8 with corresponding values of 17.38 %, 11.71 %, 8.17 %, 7.37 %, 5.63 %, 5.0 %, and 4.49 % respectively (29, 19). Therefore, it would be beneficial to choose lines and traits from PC1. Comparable results from the PCA indicated that the third PC, among the nine PCs, had eigenvalues exceeding one, explaining 68.71 % and 86.28 % of the total variability respectively.

The factor loadings of the PCs indicated that PC1 accounted for the highest variability in traits like NTrPP, LAI and GY per plant. Meanwhile, PC2 contributed 17.38 % of the total variance, showing the most variation in NLPP. PC2 had an eigenvalue of 1.945 and accounted for 17.683 % of the total variance. While PtH, total fresh yield and dry yield contributed

negatively, the remaining traits exhibited positive contributions to the variation in PC2 (28). PC3 accounted for 11.71 % of the variability, with the highest variation observed in LSR. PC4 explained 8.17 % of the variability, expressing the maximum variation in PtH, LfW, FgLW and SG. PC1 demonstrated the greatest variability, encompassing traits such as PtH, SpL, LfSL, LfBL and LfBW contributing 34.17 % to the total variability (29). The trait PtH influenced more in PC1 (30). The genetic diversity analysis of 31 hybrid pearl millet revealed that PC1 accounted for 37.44 % of the total variability, primarily influenced by GY and its associated traits (26). Likewise, the PCA classified 68 pearl millet germplasm accessions based on yield and yield-related traits into eight principal components, with PC1 accounting for 25.27 % of the total variability (27). PC1 had the highest amount of variance (36.2 %) for the traits viz., yield per plant, productive tillers per plant and fodder yield per plot for 40 pearl millet genotypes (31).

The distribution of pearl millet accessions, influenced by distinct variables from first three components (with accessions arranged by their plot numbers), was spread across different groups, highlighting the genetic diversity among the pearl millet accessions. PC1 and PC2 were plotted in a biplot to assess the associations among pearl millet accessions based on yield and quality-related traits (Fig. 3). The genotypes such as IP15351, IP3613, IP15348, IP20347, IP11840, IP15290, IP15369 and IP15341 created a cluster in the right upper corner of the biplot, showing positive values of both the PCs and the characteristics viz. PeL, NTrPP, NPrTP, LAI, LSR, GFY, DFY and NNoPP placed in the same quadrant influencing the variation. PC1 and PC3 were plotted against each other in a biplot to visualize the distribution of genotypes and their contribution to trait variation (Fig. 4). Genotypes namely IP15306, IP11840, IP15351, IP10437, IP15344, IP15307, IP15285 and IP203347 created a group in the biplot's upper right corner, displaying favourable PC and trait scores for LSR, DFY, GFY, LAI, NTrPP, NPrTP, FgLW, FgLL and DFyF positioned in the same quadrant influencing the variation. In Fig. 5, PC2 and PC3 were plotted against each other in biplot. Genotypes viz. IP15307, IP11840, IP15343, IP15351, IP15348, IP20339, IP20348, IP15290, IP20273, IP11839, IP3080, IP15320, IP15710, IP20347 and IP3616 established a group in the biplot's upper right corner, displaying desirable PC and trait rankings for DFY, GFY, LAI, NNdPP and NTrPP positioned in the same quadrant influencing the variation.

Cluster analysis provides a hierarchical distribution of accessions, which is used to identify significant relationships among genotypes and to select the best parents for breeding programs aimed at developing superior hybrids (28). In the

Table 6. Contribution of grain yield traits towards divergence in 50 pearl millet genotypes

Traits	Number of times ranked first	Contribution per cent
DFyF	16	0.20
PtH	185	15.10
PeL	67	5.47
NNdPP	430	35.10
FgLL	8	0.65
FgLW	42	3.23
FgLA	62	5.06
NTrPP	6	0.49
NPrTP	1	0.08
SpL	13	1.06
SWS	43	3.52
TGW	56	4.57
RL	84	6.86
GY	228	18.61

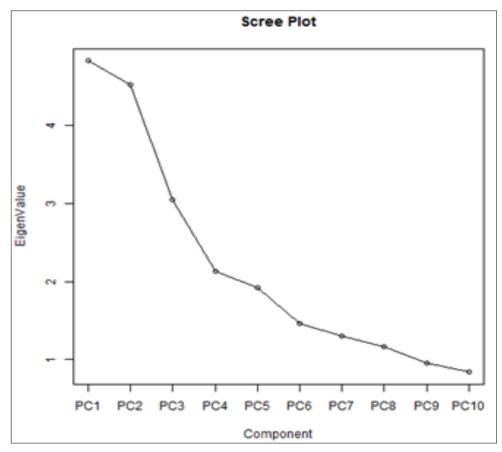
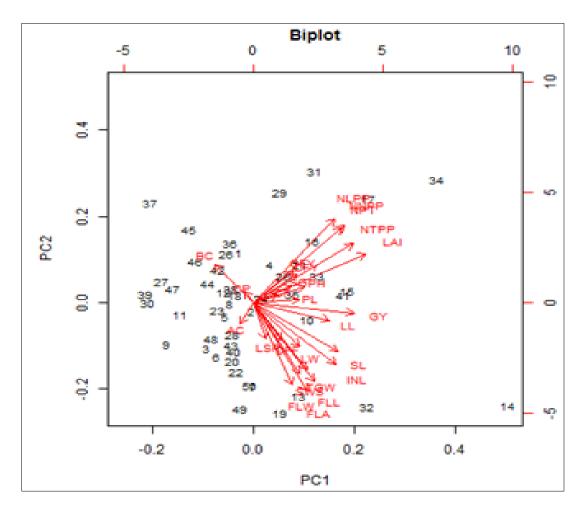


Fig. 2. Scree plot for different PCs in ICRISAT pearl millet accessions.

Table 7. Eigen value, contribution of variability for the principal component of variance in pearl millet accessions

Statistics	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Standard deviation	2.1949	2.126	1.7448	1.4576	1.384	1.2105	1.1407	1.0804
Percentage of Variance	18.53	17.38	11.71	8.17	7.37	5.63	5.0	4.49
Cumulative Percentage of Variance	18.53	35.91	47.62	55.79	63.16	68.8	73.8	78.29
Eigen Values	4.82	4.60	3.04	2.12	1.91	1.46	1.30	1.17
DFyF	0.0874	-0.1415	0.0626	-0.0906	0.2912	-0.1474	0.1369	-0.5876
PtH	0.1581	0.0601	-0.2434	0.352	0.2003	-0.1086	-0.0641	-0.1484
PeL	0.1434	0.0115	-0.4026	0.2506	0.0311	0.0343	0.1007	0.0314
NNdPP	0.2831	0.2919	0.1097	0.0035	-0.0055	-0.0648	-0.2391	-0.1329
NTrPP	0.3105	0.221	0.0553	-0.0561	0.0106	0.3041	0.0977	-0.1056
InL	0.2571	-0.2318	0.1203	-0.1011	-0.2628	-0.1027	0.0929	0.1405
LfL	0.2375	-0.0679	0.0536	-0.1033	-0.376	-0.2628	0.3281	0.0117
LfW	0.1438	-0.1659	0.0596	0.3646	-0.0291	-0.1739	-0.1254	0.2471
LAI	0.349	0.1839	0.1017	0.1171	-0.1895	-0.1556	-0.0497	0.046
FgLL	0.1887	-0.297	0.182	0.0863	-0.0808	0.1851	0.0028	-0.0944
FgLW	0.12	-0.3102	0.1723	0.3023	-0.0315	0.1737	-0.0623	0.0015
FgLA	0.1654	-0.3328	0.1813	0.2461	-0.0429	0.2152	-0.0135	-0.0716
NLPP	0.2516	0.3149	0.0995	0.0249	-0.082	-0.0192	-0.1416	-0.0659
SG	0.1153	0.064	-0.3001	0.4125	0.1361	0.0295	0.171	-0.1188
LSR	0.038	-0.1354	0.3351	-0.0404	0.1149	-0.0648	0.2419	-0.3585
GFY	0.1241	0.1194	0.2583	-0.1143	0.321	-0.0685	0.137	0.2941
DFY	0.1352	0.1074	0.2987	-0.0868	0.3535	-0.0399	-0.0211	0.1143
SL	0.2648	-0.1854	-0.1814	-0.2393	0.0388	-0.0835	-0.1431	0.1226
NPrTP	0.2763	0.2803	-0.0309	-0.0638	-0.0894	0.2843	0.1209	-0.0226
SWS	0.1438	-0.2676	-0.2922	-0.262	0.1408	-0.0793	0.0206	0.1248
TGW	0.1668	-0.2522	-0.1691	-0.2209	0.2494	0.1126	-0.0193	-0.0722
AC	-0.0423	-0.0826	-0.0941	-0.1128	-0.4738	-0.025	-0.0978	-0.2096
BC	-0.1186	0.1442	-0.1266	-0.0611	-0.1738	0.209	0.3551	-0.2354
CP	-0.0265	0.0423	0.0197	0.1483	0.0366	-0.3768	0.6075	0.1791
CF	0.0758	0.0296	-0.0947	-0.0486	-0.0199	-0.5535	-0.2836	-0.2878
GY	0.3145	-0.0396	-0.2872	-0.2498	0.0553	0.1128	0.117	0.0933



 $\textbf{Fig. 3}. \ \textbf{The biplot of pearl millet accessions for PC1 and PC2}$

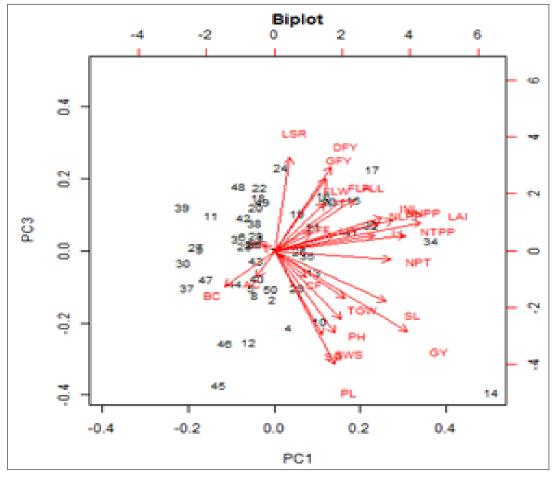


Fig. 4. The biplot of pearl millet accessions for PC1 and PC3.

Table

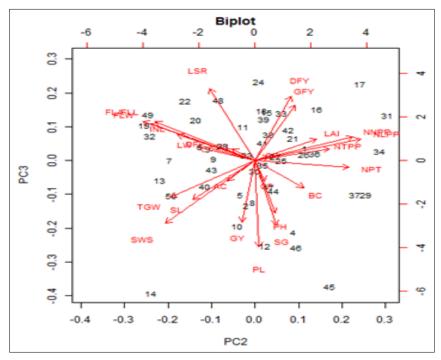


Fig. 5. The biplot of pearl millet accessions for PC2 and PC3.

current study, fifty pearl millet accessions were divided into five separate clusters using the hierarchical clustering method based on average linkage distance (Fig. 6, Table 8). Five distinct clusters were formed, namely Cluster I, II, III, IV and V, comprising 24, 16, 1, 5 and 4 accessions respectively. The mean values of different traits across pearl millet germplasm clusters are given in Table 9. Cluster I indicated greatest mean value for the trait LSR (1.55) and cluster II for AC (22.56 g). Cluster III had highest mean value for PeL (39.3 cm), LfL (52.81 cm), LfW (3.42 cm), FgLL (37.17), FgLW (3.38), FLAI (84.72), SpL (58.6 cm), SWS (38.59 g), TGW (19.29), Ph

(345.4 cm) and GY (473.76 g). Cluster IV had the lowest mean value for PtH (194.43 cm) and highest for NNdPP (221.66), NTrPP (16.43), INL (18.97 cm), LAI (0.23), NLPP (201.08), GFY(1007.4 g), DFY (311.18 g) and NPrTP (15.74). Cluster V exhibited low mean value for DFyF (50.62). As a result, the genotypes in those go clusters could be exploited for the imminent breeding programs. Earlier findings with 31 pearl millet genotypes also reported a similar grouping pattern. The classification of genotypes into distinct clusters confirmed the existence of variation among them (26).

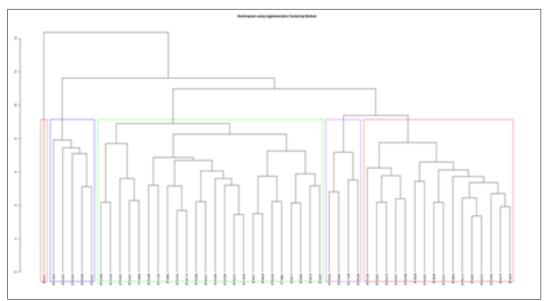


Fig. 6. Phenotypic dendrogram generated using agglomerative clustering method in pearl millet accessions.

Table 8. Hierarchical cluster grouping in pearl millet accessions

Clusters	Cluster size	Name of the genotype in clusters
I	24	IP3080 IP3613 IP3616 IP3627 IP3645 IP3665 IP5836 IP10437 IP11839 IP15285 IP15288 IP15301 IP15302 IP15306 IP15320
		IP15344 IP15348 IP15369 IP15710 IP20346 IP20347 IP21226 PT4806 PT4881
		IP3476 IP3604 IP3625 IP3628 IP3636 IP3663 IP15257 IP15307 IP15321 IP15322 IP15342 IP20273
II	16	IP20339 IP20348 IP20350
		IP20379
III	1	IP8327
IV	5	IP11840 IP15290 IP15341 IP15343 IP15351
V	4	IP17428 IP20539 IP20540 IP20585

Table 9. Cluster means of different traits of pearl millet accessions

Traits	Clusters	Min	Max	Mean	Std. Dev
	1	45.5	70	60.15	6.32
	2	39	62	55.72	6.4
DFyF	3	65	65	65	
	4	38	63.5	51.4	11.15
	5	44	54.5	50.62	4.85
	1	131.45	318.52	213.56	46.49
	2	120.33	235.03	169.49	35.12
PtH	3	345.4	345.4	345.4	
	4	132.43	261.22	194.43	47.54
	5	261.3	382.3	312.7	50.87
	1	5.21	45.11	18.98	10.06
	2	3.52	29.5	11.09	7.08
PeL)	3	39.3	39.3	39.3	
,	4	5.88	28.14	16.24	10.36
	5	13.2	42.85	27.44	15.98
	1	26.25	162.5	74.63	41.3
	2	21.2	176.8	62.42	38.17
NNdPP	3	157.8	157.8	157.8	30.11
WWATT	4	123.2	291.8	221.66	75.27
	5	51.6	101.5	78.78	22.93
	1	5	16.75	9.87	3.22
	2	4.05	11.9	7.47	2.34
NTrPP	3	14.7	14.7	14.7	
	4	14.3	18.5	16.43	1.88
	5	3	9	5.88	2.84
	1	11.61	28.62	19.6	4.01
	2 3	10.02	24.52	18.47	3.7
InL	3	24.66	24.66	24.66	
	4	15.94	25.21	18.97	3.81
	5	7.51	14.21	10.47	2.86
	1	18.47	60.66	41.34	11.51
	2	19.21	56.9	39.42	11.25
LfL	3	52.81	52.81	52.81	11.25
LIL	4	32.74	55.22		8.4
				42.81	
	5	18.5	27.41	23.16	4.26
	1	2.28	4.34	3.14	0.52
	2	1.36	3.97	2.8	0.59
LfW	3	3.42	3.42	3.42	
	4	1.72	3.34	2.63	0.64
	5	2.49	3.88	3.04	0.59
	1	0.03	0.25	0.11	0.07
	2	0.02	0.19	0.07	0.04
LAI	3	0.17	0.17	0.17	
	4	0.13	0.44	0.23	0.12
	5	0.03	0.11	0.06	0.04
	1	15.25	40.41	26.34	7.12
	2	14.82	37.85	23.83	5.86
FgLL	2 3	37.17	37.17	37.17	3.00
I BLL	J				E EO
	4	14.44	28.1	21.57	5.58
	5	9.5	21.16	12.98	5.55
	1	1.43	4.85	3.14	0.88
E 1111	2	1.47	3.48	2.48	0.62
FgLW	3	3.38	3.38	3.38	_
	4	1.44	3.57	2.34	0.82
	5	1.97	2.63	2.2	0.29
	1	19.1	129.55	59.14	30.13
	2	19.81	71.38	40.74	15.91
FgLA	3	84.72	84.72	84.72	
•	4	17.67	59.15	34.6	16.14
	5	12.71	37.7	20.2	11.8
	1	28.8	155.9	76.49	40.17
	2	33.7	121.5	65.39	25.61
NLPP	3	93.9	93.9	93.9	20.01
INEL I	4	138.5	299	201.08	66.85
	''	57.5	105.5	74.88	21.12
	5				
	1	2.12	7.78	4.05	1.3
	2	1.3	4.25	2.33	0.94
SG	2 3 4	5.76	5.76	5.76	
	4	2.55	6.34	3.81	1.61
	5	4.76	7.7	6.15	1.24
	1	1.29	1.71	1.55	0.13
	2	1.3	1.61	1.42	0.1
LSR	2 3	1.34	1.34	1.34	V.±
LOK			± 1		
LSK	4	1.28	1.65	1.43	0.13

	1	113.5	1290	704.15	279.8
	2	153.5	1678	587.53	423.4
GFY	3	669.5	669.5	669.5	
	4	390.5	2278.5	1007.4	767.82
	5	171.5	1110	580.25	443.66
	1	81.3	709.43	231.7	136.56
	2	77.4	325.8	170.98	74.33
DFY	3	221.7	221.7	221.7	
	4	167.5	559.6	311.18	152.38
	5	99.1	184.3	138.43	37.14
	1	12.33	32.7	22.08	5.53
	2	17.67	29.96	22.1	3.45
SpL	3	58.6	58.6	58.6	
	4	13.86	27.13	22.26	5.63
	5	9.88	21.48	14.75	5.03
	1	3.7	13.85	8.74	3.02
	2	3.6	12.3	7.58	2.46
ND-TD					2.40
NPrTP	3	12.3	12.3	12.3	1 74
	4	14	18.5	15.74	1.74
	5	3	13.05	7.12	4.61
	1	8.24	27.77	14.67	5.5
	2	7.74	19.85	13.35	3.49
SWS	3	38.59	38.59	38.59	
	4	7.88	11.7	10.04	1.84
	5	6.85	14.85	11.85	3.57
	1	6.39	17.45	10.61	2.87
	2	3.38	12.67	8.59	2.16
TGW	3	19.29	19.29	19.29	
	4	5.67	11.81	8.79	2.45
	5	5.38	9.72	7.22	1.82
	1	5	40	17.25	8.06
	2	4	44	22.56	11.09
AC	3	18	18	18	
	4	14	22	18	3.16
	5	12	22	18	4.32
	1	4.13	14.9	8.48	2.93
	2	4.92	16.69	8.78	3.46
ВС	3	4.58	4.58	4.58	
	4	5.53	14.07	8.69	3.39
	5	4.97	16.59	9.29	5.41
	1	2	35	16.99	8.92
	2	2.5	26	13.75	8.12
СР	3	10.5	10.5	10.5	0.12
СР	4	5.5	15.5	9.4	3.78
		9			
	5		31.5	18.62	9.93
	1	6.91	11.28	8.89	1.2
C.F.	2	7.17	10.76	8.9	1.08
CF	3	10.76	10.76	10.76	
	4	7.93	11.11	9.34	1.51
	5	8.83	10.85	9.86	1
	1	46.5	219.94	121.77	48.25
	2	50.58	242.81	101.56	50.12
GY	3	473.76	473.76	473.76	
	4	108.67	188.27	156.5	30
	5	41.73	125.55	75.91	39.33

Conclusion

Based on genetic diversity and principal component analysis, it could be found that the pearl millet genotypes such as IP8327, IP3628, IP3613, IP20347, IP15341 and IP11840 are divergent and can be exploited as parents in hybridization programs. Traits such as GY, NTrPP, PtH, NNdPP, AC, DFY and SG expressed higher heritability and maximum genetic divergence. Therefore, these characters could be prioritized for the choice of parents. The current study also points out for selecting suitable parents in a population improvement program, emphasizing genetic divergence analysis over geographical distances.

With respect to genetic diversity analysis, the cluster II had the highest intra cluster distance and cluster IV and XI exhibited the greatest inter cluster distance for GY and yield component traits. Based on cluster mean performance, the genotypes present in the clusters viz., XI (IP8327), VI (IP3628), III (IP3613), V (IP3645) had superior GY performance. Based on biometrical mean performance and diversity cluster mean values, the genotypes IP8327, IP3628, IP3613 and IP15341 had greater yield performance and good forage quality. Regarding PCA analysis, among the 26 traits analyzed, eight PCs had eigenvalues greater than 1.00, accounting for approximately 78.29 % of the total cumulative variability. PC1 had a great maximum variability of about 18.53 % with the highest eigenvalue and PC1 accounted for maximum variability for traits like NTrPP, LAI and GY per plant. Cluster analysis classified the genotypes into five distinct clusters viz. I, II, III, IV and V consisting of 24, 16, 1, 5 and 4 genotypes with cluster III with desirable mean values for PeL, LfL, LfW, FgLL, FgLW, LAI, SpL, SWS, TGW, PtH and GY and cluster IV for NNdPP, NTrPP, INL, LAI, NLPP, GFY, DFY and NPrTP as desirable traits. The genotypes from these clusters could be employed for the genetic enhancement of pearl millet with GY and forage yield.

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Authors' contributions

NA and RA conceived the concept and wrote the manuscript. DS and MAP gave idea for design the diagrams and tables. AS assisted in writing research paper, design the diagram and final draft. NA, RA, DS, MAP, AS revised and finalized the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

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

Ethical issues: None

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