PRINCIPAL COMPONENT AND CLUSTER ANALYSIS FOR QUANTITATIVE TRAITS TO IDENTIFY HIGH YIELDING GENOTYPES OF PEARL MILLET [PENNISETUM GLAUCUM (L.) R. BR.]

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SUMMARY

The 40 accessions of pearl millet were evaluated in randomized block design with three replications at experimental farm, SKN College of Agriculture, Johner (Rajasthan) during Kharif 2019. The wide range of genetic distinction for yield and yield component traits was recorded in the germplasm considered under study. The first four principal components having eigen value greater than one were extracted from the mean of 10 traits in 40 accessions with 67.61% variances. A variance of 25.71, 16.87, 13.32 and 11.69 % were extracted from first, second, third and fourth principal components, respectively. Hierarchical clustering technique based on ten quantitative trait data using Ward's method grouped 40 genotypes into five clusters with clear differentiation for different quantitative traits. Mostly accessions were not assembled into the same cluster based on their geographical origins. Based on PCA analysis, the first principal component (PC1) was the most important which accounted 25.71 % of variation with the major contributors traits biological yield per plant, plant height, panicle length, grain yield per plant and 1000-grain weight. Based on cluster analysis, genotypes of different origin like RIB-12141, Jhukarana, H-77/833-2, G-73-107, RIB-9215, RIB-16332, Desi panchu, AICRP(PM)-21, RIB-10011, AICRP(PM)-292 were grouped in cluster I therefore, breeders can use these genotypes directly as inbred lines and also for further hybridization with low yielding clusters like cluster II and V for yield improvement in pearl millet.

Key words: Pearl millet, PCA, Diversity, germplasm, genotypes

Pearl millet (Pennisetum glaucum (L.) R. Br., syn. Cenchrus americanus (L.) Morrone), a C4 grass, is a highly cross-pollinated diploid (2n = 2x = 14) with excellent photosynthetic efficiency and biomass production potential (Varshney et al., 2017). Considering the diversity and present-day distribution, a defused belt stretching from western Sudan to Senegal as the center of origin for pearl millet from where it spread to India. It is grown by subsistence farmers on the marginal agricultural land of Sub-Saharan Africa and South-East Asia with diverse and harsh climatic conditions where other cereal crops generally fail (Haussmann et al., 2012; Yadav et al., 2016). It is staple food for 90 million poor populations and preferred as main source of dietary energy with protein for the majority of people of western India particularly Rajasthan and Gujarat, endowed with dry and desert land with less rainfall. Pearl millet is cultivated in about 24.2 million hectares worldwide

with a production of 16.3 million tones. It occupies the fourth rank in India after wheat, rice and maize with cultivation in 7.4 million ha area producing 9.13 mt at the average productivity of 1237 kg ha⁻¹ (Anonymous, 2019). Based on climatic conditions and rainfall pattern the pearl millet growing area of India has been divided into three different zones (A1, A and B). The A zone consists of northern and part of northwestern India, B zone consists of peninsular India, receiving >400 mm of annual rainfall and A1 zone consists of parts of north-western India receiving less than 400 mm annual rainfall. Pearl millet grain has relatively high nutritional value compared other cereals in terms of both protein content and amino acid composition (Dayakar et al., 2017). Its grains are highly nutritious, with 8-19% protein, high starch (62.8 to 70.5 g/100 g), high fiber (1.2 g/100 g) and also rich in vitamin B-complex with higher levels of grain Fe and Zn (Velu et al., 2007; Nambiar et al. 2011; Govindaraj *et al.*, 2013; Govindaraj *et al.*, 2020). In addition, it is also a high quality forage crop because of its lower hydrocyanic acid content than sorghum and richness in protein, calcium and phosphorus with low fiber, lignin and oxalic acid content (Tako *et al.*, 2015).

Despite the clear importance of pearl millet, the production and productivity level of this staple crop are very low because it is grown in rainfed conditions, marginal production environments with minimal use of commercial inputs. Genetic improvement in quantitative traits like grain yield in any crop can be achieved through a clear understanding of the nature and amount of variability present in the breeding materials and their utilization in its genetic improvement. Pearl millet germplasm exhibits a wide range of valuable genetic variability for agronomic traits, tolerance to biotic and abiotic stresses (Stich et al., 2010; Bashir et al., 2014), although less utilized in breeding program (Yadav et al., 2009). Therefore, assessing the extent of variability for the economically important traits and identification of promising germplasm in untapped genetic resources of pearl millet is essential for its genetic improvement. In a crop improvement programme, observations are taken on several traits because of their inter-relationships; however, a breeder may be interested in selecting only few important traits in which the perfection is needed. A number of statistical procedures have been proposed from time to time for selection of important characters.

Step-wise regression analysis and Principal component analysis can be used by researchers for the purpose because it reduced the number of variables. Further genetic diversity assessment can be useful to identify contrasting parental materials to enhance heterozygosity in hybrids. These techniques have been used frequently in pearl millet (Pucher *et al.*, 2015, Animasaun *et al.*, 2017, Mithlesh *et al.*, 2020). Therefore, the present study was undertaken to evaluate pearl millet germplasm, study the relationship between different genotypes by clustering and PCA approach and to identify different germplasm suitable for further hybridization programme.

MATERIALS AND METHODS

A total of 40 accessions of pearl millet germplasm were grown in randomized block design with three replications at experimental farm, SKN College of Agriculture, Jobner, Jaipur (Rajasthan), India during *Kharif* 2019. The germplasm lines were collected from All India Coordinated Pearl millet Improvement Project (AICPMIP), Rajasthan Agricultural Research Institute, Durgapura, Jaipur (Table 1). The geographical location of experimentation was 450 meters above mean sea level on North latitude 26°05' and East longitude 75°28' in agro climatic zone III A (semi- arid eastern plain zone) in Rajasthan. The soil of experimental site was loamy sand in texture with a pH of 7.5 and climatic condition falls under the

TABLE 1 Details of experimental material

S.	Accession Name	Source	Origin	S. No.	Accession Name	Source	Origin
No.							
1.	J-2340	JAU, Gujarat	India	21.	IC-102793	NBPGR, New Delhi	India
2.	H-77/833-2	HAU, Hisar	India	22.	IC-139869	NBPGR, New Delhi	India
3.	RIB-15137	SKNAU, RARI, Durgapura	India	23.	IC-939029	NBPGR, New Delhi	India
4.	RIB-15131	SKNAU, RARI, Durgapura	India	24.	IC-332703	NBPGR, New Delhi	India
5.	G-73- 107	JAU, Gujarat	India	25.	Ardi Dungri ka bas	Local, RARI, Durgapura	India
6.	HBL-11	HAU, Hisar	India	26.	Desi panchu	Local, RARI, Durgapura	India
7.	RIB-16308	SKNAU, RARI, Durgapura	India	27.	Jhukarana local	Local, RARI, Durgapura	India
8.	RIB-16332	SKNAU, RARI, Durgapura	India	28.	NKD/YSR-2994	Local, RARI, Durgapura	India
9.	RIB-6031	SKNAU, RARI, Durgapura	India	29.	Thakarana	Local, RARI, Durgapura	India
10.	RIB-7056	SKNAU, RARI, Durgapura	India	30.	NBPGR-45	NBPGR, New Delhi	India
11.	RIB-8016	SKNAU, RARI, Durgapura	India	31.	AICRP(PM)-21	AICRP, Mandor (Jodhpur)	India
12.	RIB-8079	SKNAU, RARI, Durgapura	India	32.	AICRP(PM)-23	AICRP, Mandor (Jodhpur)	India
13.	RIB-8089	SKNAU, RARI, Durgapura	India	33.	AICRP(PM)-25	AICRP, Mandor (Jodhpur)	India
14.	RIB-8127	SKNAU, RARI, Durgapura	India	34.	AICRP(PM)-31	AICRP, Mandor (Jodhpur)	India
15.	RIB-9215	SKNAU, RARI, Durgapura	India	35.	AICRP(PM)-236	AICRP, Mandor (Jodhpur)	India
16.	RIB-10011	SKNAU, RARI, Durgapura	India	36.	AICRP(PM)-238	AICRP, Mandor (Jodhpur)	India
17.	RIB-11006	SKNAU, RARI, Durgapura	India	37.	AICRP(PM)-292	AICRP, Mandor (Jodhpur)	India
18.	RIB-12141	SKNAU, RARI, Durgapura	India	38.	AICRP(PM)-302	AICRP, Mandor (Jodhpur)	India
19.	RIB-11591	SKNAU, RARI, Durgapura	India	39.	ICMB-97444	ICRISAT, Hyderabad	India
20.	RIB-12171	SKNAU, RARI, Durgapura	India	40.	ICMB-00111	ICRISAT, Hyderabad	India

category of semi-arid region, characterized by less than 400 mm of annual average rainfall. Each line was sown in two rows, 4.0 m in length with 0.45 m interrow spacing following standard cultivation practices. Fertilizers were applied at a rate of 100 kg nitrogen and 40 kg phosphorus per hectare. Two life-saving irrigations (30 DAS and 60 DAS) were given. The observations were recorded for 10 quantitative traits such as days to 50% flowering, days to maturity, plant height (cm), number of productive tillers per plant, panicle length (cm), panicle diameter (cm), 1000-grain weight (g), biological yield per plant (g), harvest index (%) and grain yield per plant (g). Observations were recorded on five randomly selected competitive plants for each entry, in each replication for all the characters, except days to 50% flowering and days to maturity which were recorded on plot basis. The mean data were subjected to analysis of variance following the method suggested by Panse and Sukhatme (1985). The major descriptive statistics such as mean, range and coefficient of variation for each traits were computed using excel sheet program. Principal component analysis is a simple non parametric technique for extracting significant information from confounding data sets. With expose minimum efforts, this gives a roadmap for how to spruce down a multifaceted data set to simplified structures that often underlines it. The objective of principal component analysis is to identify the minimum number of components, which can explain maximum variability out of the total variability and also to rank germplasm on the basis of PC scores. PCA was performed using the statistical package SPSS 16.0 version by using data reduction approach under factor analysis. Cluster analysis was done using the Wards method of hierarchical clustering technique (Ward, 1963) and the accessions were grouped based on similarity matrix.

RESULTS AND DISCUSSION

The analysis of variance for the experiment revealed that presence of significant differences among 40 genotypes for all the ten characters (Table 2). The wide range of descriptive statistics indicating the presence of high variability in genotypes studied (Table 3). Range of variation was highest for plant height, days to maturity, harvest index, biological yield per plant, panicle length and grain yield per plant. On the basis of coefficient of variation higher variability was observed for plant height followed by panicle length, grain yield per plant, 1000-seed weight and biological yield per plant. Higher range, coefficients of variation

and large differences in mean values for most of the characters revealed that sufficient diversity existed among the genotypes and traits. The present findings were similar with previous reports in pearl millet (Anuradha *et al.*, 2018; Sharma *et al.*, 2018 and Mahendrakar *et al.*, 2019).

In present investigation, the mean data of ten quantitative traits was subjected to principal component analysis that follows a data reductionist approach involving a linear combination of optimally-weighted observed variables and helps in identifying the plant traits that contribute most towards the total variation. The first four principal components having eigen value greater than one were extracted from the mean of 10 traits and they explained 67.61% variance in pearl millet germplasm lines (Fig. 1). The first principal component (PC1) was the most important and accounted 25.71 % of variation. The major contributors for variation observed in first principle component were biological yield per plant, plant height, panicle length, grain yield per plant and 1000-grain weight. A variance of 16.87, 13.32 and 11.69 per cent were extracted from second, third and fourth principal components, respectively. The variations in PC2 were mainly due to grain yield per plant, harvest index, effective tiller per plant and days to maturity. PC3 imparted 13.32 per cent variance mainly through days to maturity, effective tiller per plant, panicle diameter and panicle length. Likewise major contributors to the variation observed in PC4 were days to 50% flowering, 1000-grain weight, days to maturity, panicle length and harvest index (Table 4, 5). The results indicated the role of traits (specific to each PC) which contributed more towards genetic divergence in discriminating the genotypes of pearl millet. The present study was in agreement with the PCA traits analysis of Animasaun et al., (2017), Sangwan et al., (2019), Mithlesh et al., (2020) in pearl millet. The three-dimensional view of all quantitative traits based on principal components to represent the variability in pearl millet is also present in Fig. 2.

PCA mainly contributes towards consideration of several traits simultaneously in the selection of materials with the added advantage of selectively rejecting traits by virtue of their duplication leads to not only greater labour but also causes loss of precision in the selection process when large numbers of characters are considered together. Since the principal components are based on correlation matrix, it stands to reason that only a few of the characters are considered providing the same extent of information as well as precision (Chaudhary *et al.*, 2015). The PC 1 and PC 2 involving characters of major economic

TABLE 2										
Analysis of variance for yield and other quantit	ative characters in pearl millet									

Source of Variation	DF	DFF	DM	PH	ETP	PL	PD	SW	BYP	HI	GYP
Replications	2	0.499	3.265	18.426	0.001	13.017	0.002	0.219	24.863	37.327	0.144
Treatments (Genotypes)	39	8.854*	107.926*	1943.717*	0.064*	16.702*	0.094*	2.143	66.835*	64.924*	13.102*
Error	78	1.689	4.367	42.643	0.007	7.775	0.009	0.63	14.55	41.606	3.889

^{**}Significant at 1% level, * Significant at 5% level.

DFF: Days to 50% flowering; DM: Days to maturity; PH: Plant height; ETP: Effective tillers per plant; PL: Panicle length; PD: Panicle diameter; GW: 1000 grain weight; BYP: Biological yield per plant; HI: Harvest index; GYP: Grain yield per plant.

TABLE 3
Descriptive statistics for yield and other quantitative characters in pearl millet

Characters	Minimum	Minimum Maximum		Std. Deviation	Coefficient of Variation	
DFF	45.33	54.33	49.62	1.717	3.46	
DM	71.00	90.00	82.48	5.997	7.27	
PH	75.45	165.51	124.56	25.453	20.43	
ETP	1.34	1.91	1.55	0.145	9.35	
PL	10.18	22.16	16.38	2.359	14.40	
PD	1.33	2.11	1.64	0.177	10.79	
GW	4.58	8.00	5.99	0.750	12.52	
BYP	30.21	46.46	37.82	4.720	12.48	
HI	32.30	51.33	39.63	4.652	11.74	
GY	12.07	22.12	14.81	2.089	14.11	

DFF: Days to 50% flowering; DM: Days to maturity; PH: Plant height; ETP: Effective tillers per plant; PL: Panicle length; PD: Panicle diameter; GW: 1000 grain weight; BYP: Biological yield per plant; HI: Harvest index; GYP: Grain yield per plant.

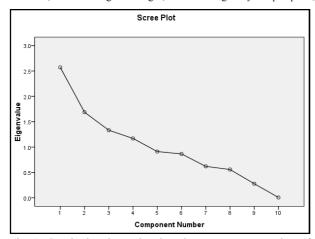
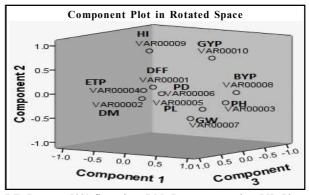


Fig. 1. Graph showing eginvalues in response to number of components for the estimated variables of pearl millet.

importance *i.e.* grain yield, harvest index, biological yield per plant, plant height, panicle length and 1000-grain weight and explaining 42.58 per cent variance were used to study the clustering/ divergence pattern of the 40 genotypes evaluated.

Cluster analysis is an important technique to classify the data which facilitates for dividing the genetic material into various homogenous groupings. It facilitates to group the genotypes on the basis of morpho-genetic traits which facilitate the selection of



DF: Days to 50% flowering; DM: Days to maturity; PH: Plant height; ETP: Effective tillers per plant; PL: Panicle length; PD: Panicle diameter; GW: 1000 grain weight; BYP: Biological yield per plant; HI: Harvest index; GYP: Grain yield per plant.

Fig. 2. PCA three dimensional plot showing 10 quantitative traits of pearl millet.

diverse lines for the crossing purpose. Cluster analysis provides an opportunity to yielding desirable segregants from crossing between the selected desirable diverse lines and bring together different gene combinations. Hierarchical clustering technique based on ten quantitative trait data using Ward's method resulted grouped 40 genotypes of pearl millet into five clusters (Fig. 3). Mean values of five clusters for different quantitative traits are given in Table 6. It was clearly

observed that cluster I possessed genotypes with high panicle length, 1000-grain weight, harvest index, grain yield per plant and early maturity. Similarly, cluster II comprised germplasm lines with medium panicle length, panicle diameter, 1000-seed weight and harvest index with low effective tillers per plant and grain yield per plant. These results were in agreement with the previous findings by Sangwan et al., (2019) in pearl millet. Cluster III comprised most of the genotypes with higher plant height and 1000-grain weight. Similarly, grouping in cluster IV including genotypes with early flowering habits with high panicle length and diameter showed the findings in close agreement with previous reports of Kumari et al., (2016) and Mithlesh et al., (2020). The genotypes early and dwarf were grouped in cluster V (Fig.3). For improving the seed yield the genotypes like RIB-12141, Jhukarana, H-77/833-2, G-73-107, RIB-9215, RIB-16332, Desi

TABLE 4
Eigenvalues, and per cent variance Explained in different PCs for 10 quantitative traits in pearl millet

Components	Eigenvalues	% of Variance	Cumulative %
1	2.571	25.714	25.714
2	1.688	16.877	42.590
3	1.332	13.322	55.913
4	1.170	11.699	67.612
5	0.912	9.122	76.734
6	0.865	8.649	85.383
7	0.620	6.197	91.580
8	0.558	5.575	97.155
9	0.278	2.785	99.940
10	0.006	0.060	100.000

TABLE 5
The estimated compound matrix in first four PCs of pearl millet genotypes

Characters	PC1	PC2	PC3	PC4	Communalities
DFF	-0.203	0.186	0.042	0.630	0.474
DM	0.051	0.300	0.677	0.376	0.693
PH	0.862	0.044	0.008	-0.003	0.745
ETP	-0.011	0.369	0.660	-0.162	0.598
PL	0.684	-0.009	0.249	0.199	0.570
PD	0.025	0.084	0.411	-0.565	0.496
GW	0.336	-0.437	0.033	0.463	0.519
BYP	0.886	0.171	-0.190	-0.124	0.865
HI	-0.481	0.747	-0.214	0.129	0.852
GY	0.434	0.799	-0.350	0.009	0.950

DFF: Days to 50% flowering; DM: Days to maturity; PH: Plant height; ETP: Effective tillers per plant; PL: Panicle length; PD: Panicle diameter; GW: 1000 grain weight; BYP: Biological yield per plant; HI: Harvest index; GYP: Grain yield per plant.

panchu, AICRP(PM)-21, RIB-10011, AICRP(PM)-292 in cluster I would be crossed with genotypes in cluster II (RIB-11006, RIB-11591, IC-102793, NKD/ YSR-2994, RIB-8079, RIB-8127, AICRP(PM)-23) and V (J-2340, HBL-11, RIB-12171, RIB-7056, RIB-16303, RIB-6031, RIB-8089, ICMB-97444, ICMB-00111). These lines also used as inbred line for further hybrid development programme. The results of cluster analysis for various yield and agro-morphological characters suggested clear differentiation of germplasm lines with some exceptions. The initial assessment of genetic materials to enable identification of potent parents for hybridization programme based on morphological data is easy, simple and can be considered as a universal approach for evaluating genetic diversity among genotypes. The results clearly indicate that cluster means of different clusters identify the characters to be chosen for hybridization and parents for different desirable traits can be easily chosen from clusters based on their merit. These results are in agreements with the findings of Drabo et al., (2013), Sankar et al., (2014), Chaudhary et al., (2015), Kumari et al., (2016) and Sangwan et al., (2019). Genotypes from different source/origin falls under the same cluster, so grouping did not happened on the basis of origin or geographical location. These

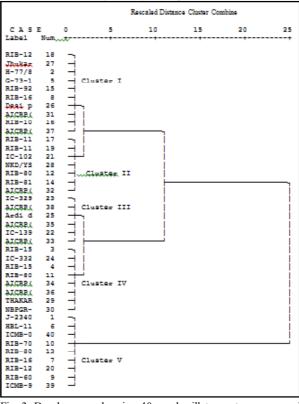


Fig. 3. Dendrogram showing 40 pearl millet genotypes grouped using Ward Method.

Cluster	No. of genotypes	Name of genotypes	DFF	DM	РН	ETP	PL	PD	GW	BYP	НІ	GYP
I	10	RIB-12141, Jhukarana, H-77/833-2, G-73-107, RIB-9215, RIB-16332, Desi panchu, AICRP (PM)-21, RIB-10011, AICRP(PM)-292	51.97	85.97	126.95	1.52	18.47	1.50	6.97	39.65	49.25	17.45
II	7	RIB-11006, RIB-11591, IC-102793, NKD/ YSR-2994, RIB-8079, RIB-8127, AICRP (PM)-23	50.33	83.56	115.22	1.46	16.91	1.64	6.16	34.90	39.19	13.53
Ш	6	IC-329029, AICRP(PM)-302, Ardi dungri ka bas, AICRP(PM)-236 , IC-139869, AICRP(PM)-25	48.67	79.73	159.77	1.66	16.98	1.65	6.29	41.97	36.70	15.31
IV	8	RIB-15137, IC-332703, RIB-15131, RIB-8016, AICRP(PM)-31, AICRP(PM)-238, Thakrana, NBPGR-45	49.71	80.54	145.22	1.54	17.95	1.66	5.87	40.43	39.64	15.96
V	9	J-2340, HBL-11, RIB-12171, RIB-7056, RIB-16303, RIB-6031, RIB-8089, ICMB-97444, ICMB-00111	49.26	80.89	87.36	1.59	14.08	1.66	5.68	32.96	42.32	13.75

 ${\it TABLE~6} \\ {\it Mean~of~five~clusters~of~pearl~millet~germplasm~for~yield~and~yield~component~traits}$

DFF: Days to 50% flowering; DM: Days to maturity; PH: Plant height; ETP: Effective tillers per plant; PL: Panicle length; PD: Panicle diameter; GW: 1000 grain weight; BYP: Biological yield per plant; HI: Harvest index; GYP: Grain yield per plant.

results are in agreement with the findings of Burson et al., (2015), Animasaun et al., (2017) and Mithelesh et al., (2020) who observed that accessions of pearl millet did not necessarily assemble into the same cluster based on their geographical origins. Clustering of pearl millet accessions together regardless of their source supports the possibility of a common progenitor but separation by geographical or ecological isolation mechanisms (Jauhar, 1981).

For any hybridization programs in pearl millet, the choice of suitable diverse parents based on genetic divergence analysis would be more rewarding than the choice based on the geographical distances. Present findings revealed that for improving the seed yield the different genotypes like RIB-12141, Jhukarana, H-77/ 833-2, G-73-107, RIB-9215, RIB-16332, Desi panchu, AICRP(PM)-21, RIB-10011, AICRP(PM)-292 (cluster I) would be crossed with genotypes like RIB-11006, RIB-11591, IC-102793, NKD/YSR-2994, RIB-8079, RIB-8127, AICRP(PM)-23, J-2340, HBL-11, RIB-12171, RIB-7056, RIB-16303, RIB-6031, RIB-8089, ICMB-97444, ICMB-00111 in cluster II and V and also used these genotypes as inbred line for further hybrid development programme. Based on the relative contributions of different characters; grain yield, harvest index, biological yield per plant, plant height, panicle length and 1000-grain weight were found the best discriminatory characters.

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