

Original Research Article

Agro-morphological Characterization of Proso millet Germplasm for Utilization

ABSTRACT

Aims: In this study, 634 accessions of proso millet were evaluated to assess phenotypic diversity using morpho-agronomic traits, aiming to identify specific donors for desired traits.

Study design: Mention the design of the study here.

Place and Duration of Study: During the Kharif season of 2021, a total of 634 proso millet germplasm accessions were characterized, alongside three control varieties, as part of the Institute Project (IIMR/CI/2021-2026/150) at ICAR-IIMR in Hyderabad.

Methodology: The experimental design employed was an augmented block design, and four check varieties, namely GPUP 8, TNAU 145, TNAU 164, and TNAU 202, were included.

Results: The results revealed significant variation across all the traits studied. Principal Component Analysis (PCA) demonstrated that the first three principal components accounted for 60% of the total variation. PC1 accounted for 27.3% of the variation, followed by PC2 (22.6%) and PC3 (10%). PC1 was primarily influenced by Leaf blade length (cm), Peduncle length (cm), Panicle length (cm), Plant height (cm), and grain yield (g plant⁻¹). Furthermore, the study identified 21 accessions with superior performance in multiple traits (3-6 traits).

Conclusion: These accessions can serve as valuable genetic resources for improving yield and quality traits. The selected multi-trait donors can be directly released for cultivation, serving as a short-term breeding goal. This approach has the potential to enable farmers to achieve high remuneration by harvesting increased yields.

Keywords: Proso millet; germplasm characterization, PCA, trait donors

1. INTRODUCTION

Proso millet (*Panicum miliaceum*) is an ancient crop that was domesticated over 10,000 years ago in China [1]. Research indicates that proso millet was independently domesticated in multiple regions and has since spread across Europe and Asia. Throughout centuries of cultivation, the crop has adapted to diverse climatic conditions, leading to significant population differentiation. The germplasm collected from various regions is likely to harbor a wide range of genetic alleles. Key proso millet-growing countries, including Russia, China, Germany, the USA, and India, have collected and conserved germplasm in their gene banks, recognizing its importance as a foundational resource in crop breeding programs. Proso millet possesses valuable nutritional attributes, as it is rich in protein, starch, dietary fiber, and several trace elements such as magnesium, iron, and calcium [2]. Moreover, it has been associated with potential health benefits, including the prevention of arteriosclerosis, gastrointestinal cancer, coronary heart disease, and other ailments [3]. In recent years, there has been a growing interest in the use of millet, particularly in food applications as a gluten-free ingredient [4]. Proso millet is extensively cultivated in countries such as China, Korea, India, Pakistan, the United States, and Russia. It serves as a food source in South and Southeast Asian countries, while in the United States, it is predominantly used as bird feed [5].

The rapid and widespread adoption of proso millet during its early domestication can be attributed to its short growing duration and resilience to biotic and abiotic stress [6], [7]. Additionally, proso millet is rich in protein [8], and recent studies have reported the presence of high levels of antioxidants and secondary

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metabolites with health benefits [9],[10]. Analyses of the C4 pathway in proso millet have revealed its greater efficiency in photosynthesis under lower gradients of transport metabolites and in varying light intensities [11]. Moreover, proso millet exhibits high water and nitrogen use efficiency [12].

Despite its desirable attributes, proso millet has become a contingent crop over time. However, it possesses immense potential in the food industry and health food sector, making it a suitable crop under the current climate change regime. The limited availability of information on the characterization of proso millet accessions in terms of yield, agronomic traits, and quality hampers its utilization and practicality. This tetraploid crop has demonstrated considerable genetic plasticity, enabling it to adapt to diverse geographical regions, ranging from sea level to altitudes of 3500 meters above sea level [13]. This geographical adaptation has resulted in significant crop diversity of proso millet worldwide. To fully harness the potential of this crop, it is crucial to undertake detailed characterization of these collections.

In India, proso millet is cultivated in several states, including Tamil Nadu, Maharashtra, Karnataka, Andhra Pradesh, Bihar, Madhya Pradesh, and Uttar Pradesh, covering an area of 0.41 lakh hectares and yielding 0.22 lakh tonnes of grains [14]. However, the crop area in Northeastern India is not accounted for in the present data. The primary challenge in proso millet cultivation lies in achieving higher yield levels. To address this issue, the Indian proso millet program has initiated a focused breeding program aimed at enhancing both yield and protein content in proso millet.

The National Bureau of Plant Genetic Resources (NBPGR) in New Delhi houses a total of 1007 indigenous collections of proso millet. These collections serve as crucial genetic resources for the ongoing proso millet improvement program conducted at the ICAR-Indian Institute of Millets Research (ICAR-IIMR). ICAR-IIMR is mandated to act as a National Active Germplasm Site for all millet crops. It is an apex body in India for research and development in millets. At present, ICAR-IIMR holds a collection of 1556 accessions collected from various sources, nationally and internationally. This study was conducted with the following objectives to characterize 634 proso millet accessions based on important agronomic traits. To develop a database of traits that can be utilized by researchers in the proso millet improvement program. To identify trait-specific germplasm that can be used as trait donors by proso millet breeders. By achieving these objectives, the study aims to enhance the understanding of proso millet's genetic diversity and facilitate targeted breeding efforts for improved varieties.

2. MATERIAL AND METHODS

2.1 The experimental design and layout

During the Kharif season of 2021, a total of 634 proso millet germplasm accessions were characterized, alongside three control varieties, as part of the Institute Project (IIMR/CI/2021-2026/150) at ICAR-IIMR in Hyderabad. The characterization process took place in the experimental fields of ICAR-Indian Institute of Millets Research (IIMR), located at geographical coordinates 17.3207°N latitude, 78.3959°E longitude, and an elevation of 476.5 meters above mean sea level.

The experimental design employed was an augmented block design, and four check varieties, namely GPUP 8, TNAU 145, TNAU 164, and TNAU 202, were included. The sowing of accessions took place in the third week of July, with a plot size of 1-meter row length and 60 cm distance between rows. Two weeks after thinning, the distance between plants was maintained at 10 cm. Fertilizers were applied at a rate of 80 kg/ha of nitrogen and 40 kg/ha of P₂O₅ during the crop's growth, while all necessary agricultural practices were followed to ensure a healthy crop stand. Regular irrigation was provided to maintain adequate moisture levels, and measures were taken to protect the crop from weeds, pests, and diseases.

2.2 Agro-morphological traits studied

The study involved characterizing the germplasm for 11 quantitative traits, namely the number of basal tillers, days to 50% flowering, flag leaf blade length, flag leaf blade width, peduncle length, panicle length,

panicle width, plant height, days to maturity, grain yield per plant, and 100-seed weight (Table 1). The trait descriptors provided by the International Board for Plant Genetic Resources (IBPGR, 1985) were followed to ensure consistency in trait assessment [15].

Table 1. Quantitative traits and measurement units

Organ	Trait	Abbreviation	Unit
Phenology	Days to 50% flowering	DAYS_FLW	days
	Days to maturity	DAYS_MAT	days
Plant	Number of basal tillers	BASAL_TILLERS	Number
	Plant height	PLANT_HT	cm
Leaf	Leaf blade length	LEAF_BLD_LEN	cm
	Leaf blade width	LEAF_BLD_WID	cm
Flower	Panicle length	PANICLE_LEN	cm
	Panicle width	PANICLE_WID	cm
Seed	100_Seed weight	100_SD_WT	g
	Grain yield	GRN_YLD_PLN	g plant ⁻¹

2.3 Statistical analysis

The analysis of variance (ANOVA) was conducted using the augmented randomized complete block design (RCBD) package of R (version 3.4.2), following the method described by Aravind et al. (2021). Pearson's correlation coefficient (r) was employed to determine the relationships between the studied traits. Factor analysis was performed based on the Pearson correlation matrix and Euclidean distances. The latent roots or eigenvalues for all principal components were calculated and presented. To assess diversity, the Shannon-Weaver diversity index (H) and principal component analysis (PCA) were generated using the PAST4 software, as outlined by [16].

3. RESULTS

3.1 Descriptive statistics and diversity indices for 634 proso millet germplasm accessions

Table 2 presents the summary statistics of the 11 studied traits, including mean, standard error of mean, standard deviation, range, and coefficient of variance. The results indicate a significant variability in all the studied traits. The number of basal tillers ranged from 1 to 24, with an average of 6.9. The days to 50% flowering varied from 36 to 60 days, with an average of 47.5 days. The flag leaf blade length measured at the time of flowering ranged from 12.3 cm to 45.8 cm, while the flag leaf blade width had a mean value of 0.94 cm, ranging from 0.23 cm to 2.9 cm. The peduncle length exhibited a minimum value of 5.3 cm and a maximum value of 15.7 cm, with an average of 2.83 cm. The panicle characteristics, such as panicle length (mean = 25.9 cm; range = 12.4 cm – 46.7 cm) and panicle width (mean = 3.3 cm; range = 1.2 cm - 7.0 cm) were also exhibiting the greatest variability with extreme values.

Table 2. Summary statistics of quantitative traits of 634 acc. of Proso millet germplasm characterized during Kharif 2021

SN	Trait	Mean	SEM	SD	CV	Minimum	Maximum
1.	Number of basal tillers	7.0	0.1	3.3	48.0	1.0	24.0

2.	Days to 50% flowering (days)	47.5	0.2	5.3	11.2	36.0	60.0
3.	Flag leaf blade length (cm)	23.3	0.2	4.1	17.6	12.3	45.9
4.	Flag leaf blade width (cm)	0.9	0.0	0.4	39.7	0.2	2.9
5.	Peduncle length (cm)	8.1	0.0	1.3	15.5	5.3	15.8
6.	Panicle length (cm)	25.9	0.2	4.0	15.3	12.4	46.7
7.	Panicle width (cm)	3.3	0.0	0.9	27.1	1.2	7.0
8.	Plant height (cm)	77.4	0.6	16.1	20.8	33.3	151.7
9.	Days to maturity (days)	75.4	0.3	6.4	8.5	59.0	89.0
10.	Grain yield (g/plant)	4.8	0.1	3.6	74.1	1.1	18.4
11.	1000-Seed weight (g)	0.5	0.0	0.1	16.7	0.1	0.8

SEM: Standard Error of mean, SD: Standard deviation, CV: coefficient of variation

The plant height with a range of 33.3 cm to 151.6 cm, had a mean of 77.3 cm. The main yield and yield attributing traits like grain yield and 100 seed weight also showed the presence of a vast variability. On the one hand, the grain yield with an average of 4.7 g per plant ranged from a minimum of 1.1 g to a maximum of 18.4 g per plant, while on the other hand, 100 seed weight was ranging from 0.06 g to 0.82 g with a mean value of 0.46 g.

The frequency distribution of proso millet accessions for various levels of studied traits along with the checks is shown in figure 1. The distribution of checks around the mean for almost all traits can be observed.

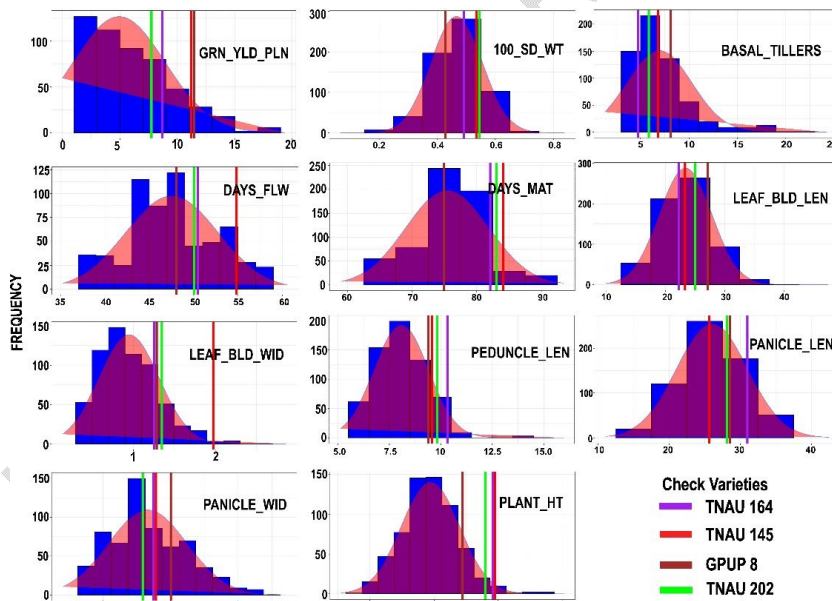


Fig 1. Histogram showing frequency distribution of studied quantitative traits in 634 Proso millet germplasm.

The shannon-weaver diversity index for studied traits ranged from 6.3 for grain yield to a maximum of 7.51 for 100 seed weight (figure 2).

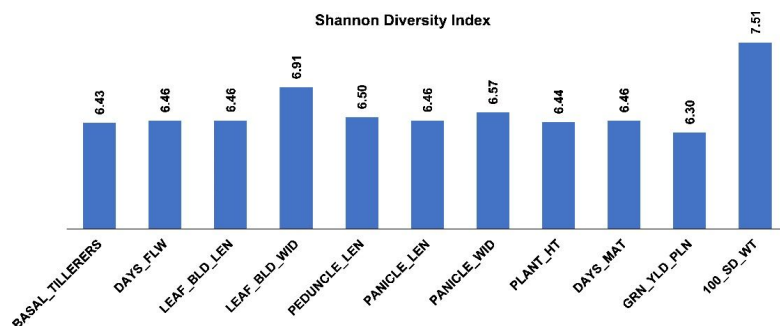


Figure 2. Diversity among 634 acc. of proso millet germplasm based on Shannon diversity index (H')

3.2 Analysis of variance

Analysis of variance (ANOVA) using augmented design indicated the presence of significant ($P < 0.05$) and non-significant ($P > 0.05$) mean sums of squares for studied traits for different sources of variation (Table 2). The block effects eliminating the treatments were non-significant ($P > 0.05$) for all the traits studied. The treatment effects ignoring blocks were highly significant ($P < 0.01$) for days to 50% flowering, days to maturity, and peduncle length indicating treatments had a significant effect on these traits, and the differences between the treatments were not due to chance.

The mean sums of squares due to germplasm versus checks were significant ($P < 0.05$) for all traits except for number of basal tillers, and panicle width. The germplasm had a significant effect on all of the traits except for number of basal tillers, and panicle width. The differences between the germplasms were not due to chance.

Table 3. Analysis of variance using augmented block design for 634 Proso millet germplasm

Source	Df	NBT	DF	DM	GY	LBL	LBW	PanL	PanW	PedL	PH	100SW
Block (eliminating Treatments)	7	4.86ns	7.46ns	19.1ns	17.86ns	12.3ns	0.06ns	39.8ns	1.19ns	0.62ns	296.2ns	0.01ns
Treatment (ignoring Blocks)	637	11.2 ns	28.94 **	42.58 **	13.88 ns	16.93 ns	0.16ns	16.15ns	0.81 ns	1.7 **	314.85 ns	0.01 ns
Check	3	16.43 ns	64.7 **	137.36 **	28.38 ns	36.94 ns	0.92 **	38.39 ns	0.94 ns	1.28 ns	706.12 ns	0.02 **
Germplasm	633	11.17 ns	28.31 **	40.72 *	12.64 ns	16.81 ns	0.14ns	15.8 ns	0.81 ns	1.57 **	259.2 ns	0.01 ns
Germplasm vs. Check	1	12.73ns	319.2 **	934.8 **	754.6 **	31.8 ns	8.3 **	171.2*	1.6 ns	85.64 **	34369**	0.03*
Error	21	8.87	8.63	17.77	31.74	16.95	0.09	31.43	0.72	0.53	306.19	0.0046
Check vsgermplasm CD at 5%		7.34	7.24	10.4	13.89	10.15	0.76	13.83	2.09	1.8	43.15	0.17

Df degree of freedom, 100SW 100 seed weight (g), DFF Days to 50% flowering (days), LBL Leaf Blade Length (cm), LBW Leaf Blade Width (cm), GY Grain Yield (g plant⁻¹), NBT Number of basal tillers, PanL Panicle Length (cm), PanW Panicle Width (cm), PedL Peduncle Length (cm), PH plant height (cm); ns $P > 0.05$; * $P \leq 0.05$; ** $P \leq 0.01$

3.3 Principal Component Analyses

Principal component analysis (PCA) of the data recorded on characterization of 11 traits from 634 proso millet germplasm accessions indicated that the first three principal components explained 60 % of the variation (Table 4). The ~~total of 11 principal components were~~total of 11 principal components

was estimated. The eigenvalue of PC1 was 3.0, while it was 0.24 for PC11. The eigen values of PC4-PC11 were less than 1, hence, only the first three PCs were considered to assess the diversity. The first principal component (PC1) explained 27.3 % of the variation, followed by PC2 (22.6 %), and PC3 (10 %). The important traits that were positively related and contributed to the variation in PC1 are: Leaf blade length (cm), Peduncle length (cm), Panicle length (cm), Plant height (cm) and grain yield (g per plant) (Table 5).

Days to 50 % flowering, days to maturity and leaf blade width are negatively related. These traits contribute maximum to the variation explained by PC2. The leaf blade length and peduncle length are positively related and contribute maximum variability to PC3.

Table 4. Eigenvalues (Latent roots) and variability of non-rotated values of principal components

Principle component	Eigenvalue	Proportion of variance Explained	Components cumulative proportion (%)
PC1	3.006	0.273	27.3
PC2	2.489	0.226	50
PC3	1.104	0.100	60
PC4	0.918	0.083	68.3
PC5	0.830	0.075	75.9
PC6	0.642	0.058	81.7
PC7	0.564	0.051	86.8
PC8	0.496	0.045	91.4
PC9	0.429	0.039	95.3
PC10	0.282	0.026	97.8
PC11	0.240	0.022	100

Table 5. Non-rotated component loadings (values of principal component traits of Proso millet)

Trait/Variable	PC1	PC2	PC3	PC4	PC5
1. BASAL_TILLERS	0.18	0.04	-0.82	-0.12	-0.02
2. DAYS_FLW	-0.04	-0.56	-0.08	0.18	-0.17
3. LEAF_BLD_LEN	0.37	0.09	-0.07	-0.45	0.09
4. LEAF_BLD_WID	0.19	-0.38	0.30	-0.40	0.08
5. PEDUNCLE_LEN	0.38	-0.07	0.40	-0.04	0.02
6. PANICLE_LEN	0.39	0.12	-0.06	0.10	-0.59
7. PANICLE_WID	0.23	0.37	0.19	0.19	-0.46
8. PLANT_HT	0.45	-0.26	-0.03	0.05	0.01
9. DAYS_MAT	0.14	-0.52	-0.15	0.25	-0.11

10. GRN_YLD_PLN	0.39	0.18	-0.09	-0.12	0.40
11. 100_SD_WT	0.28	0.10	0.03	0.68	0.48

Bold values indicate the traits which contributed maximum to principal component.

The distribution of traits among the principal components explains the existence of greater variability among the studied accessions. The scree plot, score plot, loading plot, and biplot of principal component analysis of 634 accessions of proso millet are shown in Figure 3. It is obvious from the scree plot that only the first three components explained maximum of diversity existed in the studied germplasm lines.

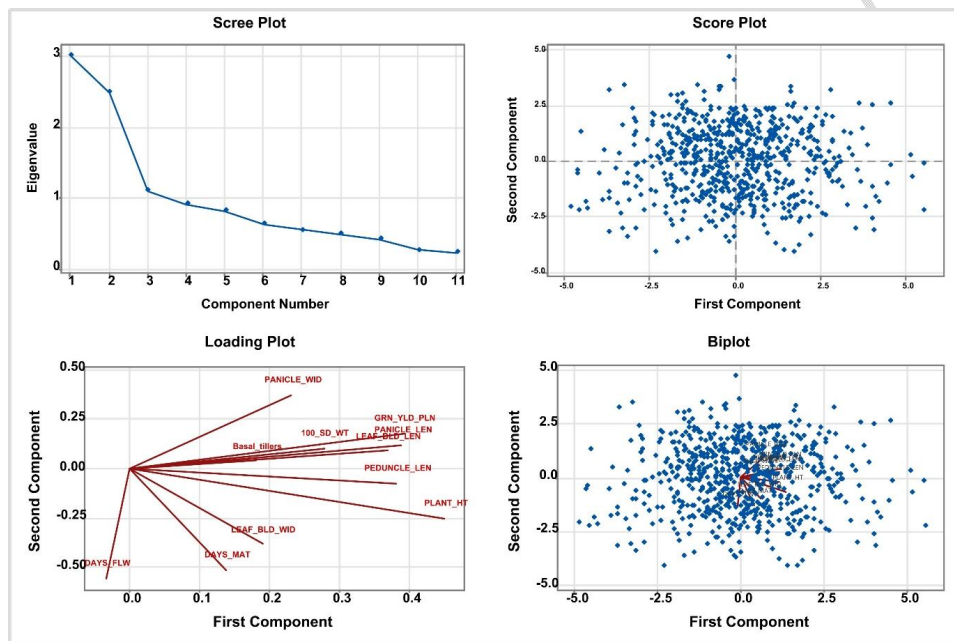


Fig3. Scree plot, Score plot, Loading plot and biplot of principal component analysis of 554 accessions of Proso millet. 100SW 100 seed weight (g), DF Days flowering (days), FLL Flag Leaf Length (cm), FLW Flag Leaf Width (cm), GY Grain Yield (g plant⁻¹), LReCL Lowest receme length (cm), NBT Number of basal tillers, PanL: Panicle Length (cm), PanW Panicle Width (cm), PedL Peduncle Length (cm), PH plant height (cm)

3.4 Frequency distribution of qualitative traits

The frequency distribution of lines for qualitative traits showed that the dominant type among the populations was recorded (Table 6). In the qualitative data, the following traits were the most frequent: Erect growth habit (444 acc.), Absence of leaf sheath pigmentation (630 acc.), Sparse leaf sheath pubescence (333 acc.), Absence of ligule pubescence (639 acc.), Glabrous leaf blade pubescence (559 acc.), Arched inflorescence shape (443 acc.), Presence of culm branching (378 acc.), Absence of lodging

(677 acc.), Absence of seed shattering (661 acc.), Intermediate panicle compactness (303 acc.), Golden yellow grain color (470 acc.), and oval grain shape (379 acc.).

Table 6: The frequency distribution of qualitative traits of 634 acc. of Proso millet germplasm characterized under Institute Project (IMR/CI/2021-2026/150) during Kharif 2021

S.No	Category	Plant growth habit
1	Erect	444
2	Decumbent	239
3	Prostrate	1
	Category	Leaf sheath pigmentation
1	Absent	630
2	Present	54
	Category	Leaf sheath pubescence
1	Sparse	333
2	Glabrous	248
3	Strong	103
	Category	Ligule pubescence
1	Absent	639
2	Present	45
	Category	Leaf blade pubescence
1	Glabrous	559
2	Sparse	107
3	Strong	18
	Category	Inflorescence shape
1	Arched	443
2	Diffused	214
3	Globose elliptic	27
	Category	Culm branching
1	Present	378
2	Absent	306
	Category	Lodging
1	Absent	677
2	Present	7
	Category	Seed shattering
1	Absent	661
2	Present	23
	Category	Panicle compactness
1	Intermediate	303
2	Open	282
3	Compact	99
	Category	Grain colour
1	Golden yellow	470
2	Grey	140
3	Light Grey	35
4	Dark grey	23
5	Cream	15
6	Grey	1
	Score/State	Grain shape
1	Oval	379
2	Elliptical	305

3.5 Potential germplasm identified

There are 442 acc. of trait specific proso millet germplasm identified. In which, 82 acc. are multi-trait specific germplasm for multi-trait specific germplasm for 2-6 traits. The list of 21 acc. identified with multi-trait specific germplasm for 3-6 traits are given Table 7.

Table 7: Multi-trait specific Proso millet germplasm identified under Institute Project (IIMR/CI/2021-2026/150) during Kharif 2021 from 634 acc. characterized

S.No.	Crop Name	MGB No	IC No/ Other ID	Multi-trait
1	Proso millet	PRO 1185	IC0484157	Flag leaf blade length (>30.00 cm) Flag leaf blade width, (>2.00 cm) and Panicle length (>32.00 cm)
2	Proso millet	PRO 165	TNPM 471	Days to 50% flowering (>40.00 days), Grain yield (>15.00 g/plant and 1000-Grain weight (>5.50 g)
3	Proso millet	PRO 208	TNPM 516	Panicle length (>32.00 cm), Panicle width (>5.00 cm), Plant height (>120.00 cm) and Grain yield (>15.00 g/plant
4	Proso millet	PRO 221	TNPM 529	Peduncle length (>11.00 cm), Panicle length (>32.00 cm) and Plant height (>120.00 cm)
5	Proso millet	PRO 226	TNPM 534	Panicle length (>32.00 cm), Plant height (>120.00 cm) and Grain yield (>15.00 g/plant
6	Proso millet	PRO 302	TNPM 615	Days to 50% flowering (>40.00 days), Plant height (<50.00 cm) and Date of harvest (<65.00 days)
7	Proso millet	PRO 325	TNPM 638	Days to 50% flowering (>40.00 days), Flag leaf blade length (>30.00 cm), Panicle length (>32.00 cm), Panicle width (>5.00 cm)
8	Proso millet	PRO 330	TNPM 643	Days to 50% flowering (>40.00 days), Plant height (<50.00 cm) and Date of harvest (<65.00 days)
9	Proso millet	PRO 356	TNPM 669	Days to 50% flowering (>40.00 days), Plant height (<50.00 cm) and Date of harvest (<65.00 days)
10	Proso millet	PRO 359	TNPM 672	Number of basal tillers (>15.00), 1000-Grain weight (>5.50 g), Days to 50% flowering (>40.00 days), Plant height (<50.00 cm) and Date of harvest (<65.00 days)
11	Proso millet	PRO 38	TNPM 336	Days to 50% flowering (>40.00 days), Panicle length (>32.00 cm), Panicle width (>5.00 cm) and 1000-Grain weight (>5.50 g)
12	Proso millet	PRO 49	TNPM 347	Days to 50% flowering (>40.00 days), Panicle width (>5.00 cm), Plant height (<50.00 cm) and Date of harvest (<65.00 days)
13	Proso millet	PRO 50	TNPM 348	Days to 50% flowering (>40.00 days), Panicle width

S.No.	Crop Name	MGB No	IC No/ Other ID	Multi-trait
				(>5.00 cm), Plant height (<50.00 cm), Date of harvest (<65.00 days) and 1000-Grain weight (>5.50 g)
14	Proso millet	PRO 593	IC0483576	Days to 50% flowering (>40.00 days), Flag leaf blade length (>30.00 cm), Peduncle length (>11.00 cm), Panicle length (>32.00 cm) and Date of harvest (<65.00 days)
15	Proso millet	PRO 618	IC0483602	Days to 50% flowering (>40.00 days), Plant height (<50.00 cm), Date of harvest (<65.00 days), Flag leaf blade length (>30.00 cm), Panicle length (>32.00 cm) and 1000-Grain weight (>5.50 g)
16	Proso millet	PRO 660	IC0483867	Days to 50% flowering (>40.00 days), Date of harvest (<65.00 days), Grain yield (>15.00 g/plant)
17	Proso millet	PRO 690	IC0484165	Number of basal tillers (>15.00), Flag leaf blade length (>30.00 cm) and Panicle length (>32.00 cm),
18	Proso millet	PRO 715	IC0484177	Number of basal tillers (>15.00), Panicle length (>32.00 cm) and 1000-Grain weight (>5.50 g)
19	Proso millet	PRO 791	IC0483718	Number of basal tillers (>15.00), Panicle length (>32.00 cm), Days to 50% flowering (>40.00 days) and Date of harvest (<65.00 days)
20	Proso millet	PRO 919	IC0483859	Number of basal tillers (>15.00), Days to 50% flowering (>40.00 days) and Date of harvest (<65.00 days)
21	Proso millet	PRO 968	IC0483913	Days to 50% flowering (>40.00 days), Plant height (<50.00 cm) and Date of harvest (<65.00 days)

4. DISCUSSION

4.1 Field evaluation of germplasm lines

Genetic characterization of a large set of available germplasm accessions conserved across the global genebanks is useful for researchers worldwide[12]. Phenotypic characterization helps breeders, geneticists, and botanists in identifying promising trait donors for use in crop improvement activities towards developing high-yielding, climate-resilient, and pest- and disease-resistant cultivars.

In the present study, a high variability was observed for the studied traits, and trait-specific germplasm lines were identified for future use in the development of cultivars for higher grain yield and earliness. Earlier researchers have also reported higher variability in studied proso millet germplasm for morpho-agronomic traits [17],[18]. The observed high coefficient of variation within the studied accessions for a few traits explains the presence of high variability for these traits.

4.2 PCA

Principal component analysis was carried out to study the traits associated with the variability components and the accessions with specific traits contributing maximum towards the variability. Leaf blade length (cm), Peduncle length (cm), Panicle length (cm), Plant height (cm), and grain yield (g plant⁻¹) were the important traits contributing to variability explained by PC1. This indicates that the accessions which fall in this region of the score plot of PCA could be used as potential donors of these traits.

On the other hand, the traits such as Days to 50 % flowering, days to maturity and leaf blade width were mainly contributing to PC2. Hence, the accessions falling in this region may be used for the development of cultivars with early flowering and maturity in proso millet. The distinctly identified accessions through the PCA may be either used for hybridization programmes as parents or may be directly released for commercial cultivation after multi-location evaluation for stability of the genotype.

The current study ascribed 60% of the total variations observed across the 11 quantitative traits to the first three PCs with eigenvalues greater than one. The PCA result shows that the 11 quantitative traits examined were discovered the inherent uniqueness among the studied proso millet accessions. Similar kind of results were reported by [17] where the variability in PC1 was contributing to a maximum extent by days to 50% flowering, plant height, flag leaf blade length, flag leaf sheath length, inflorescence length, number of nodes per main stem, days to maturity, grain yield, and protein content. Similarly, [17] also reported that first three PCs contributed majorly to diversity in the studied accessions. In another study by [19], the PCA was conducted on 18 starch quality traits in 95 proso millet accessions. The first three principal components accounted for 83.3% of the variation.

4.3 Potential germplasm

Identification of the germplasm accessions by tagging trait/traits is an invaluable information to the researchers in tailoring the new varieties with improved performance. It will help in utilizing trait specific germplasm in breeding programs to dissect the traits at genetic/molecular level. The present study has identified 21 potential accessions tagged with superior performance for more than one trait. The accession IC0483602 was identified as

potential donor for six specific traits. The accessions, TNPM 672, TNPM 348 and IC0483576 were identified as potential donors of more than five traits together in a single accession. The accessions are useful to develop lines with high panicle length, early flowering, 100 g weight etc.

Trivedi et al 2017 [20] mentioned that trait-specific germplasm is important genetic material for crop improvement, climate resilience breeding, and improving food and nutritional security in a sustainable way. Similarly, the trait-specific accessions identified with two/three/four traits in the present study will serve as ready reckoner for the improvement of targeted traits in the crop improvement programs of barnyard millet as potential donor parents specifically for the traits like earliness and high grain yielding ability. Similar kind of results were reported by [21] in foxtail millet, 26 accessions with three desirable trait combinations were identified giving useful donor lines for grain yield, high iron, Zinc, and Calcium content, and early flowering. Khairwal et al., in 2007 [22] identified trait-specific lines for vigor, extra early flowering, early maturity, plant height, high productive tillers, long panicle, thick panicle, and high grain yield and mentioned that the developed trait-specific gene-pools will serve as a core-subset for future breeding programs in pearl millet.

4. CONCLUSION

The study revealed presence of significant genetic variation for the studied eleven quantitative traits in proso millet. PCA analysis showed that the first three principal components accounted for 60% of the total variation. Additionally, the study also identified 21 multiple traits (3-6 traits) donor accessions which may directly be used for proso millet improvement by breeders.

REFERENCES

- [1] Lu H, Zhang J, Liu K, Wu N, Li Y, Zhou K, Ye M, Zhang T, Zhang H, Yang X, Shen L, Xu D, Li Q. Earliest domestication of common millet (*Panicum miliaceum*) in East Asia extended to 10,000 years ago. *Proceedings of the National Academy of Sciences of the United States of America* 2009;106(18):7367–72. <https://doi.org/10.1073/pnas.0900158106>
- [2] Cho Y-I, Chung J-W, Lee G-A, Ma K-H, Dixit A, Gwag J-G, Park Y-J. Development and characterization of twenty-five new polymorphic microsatellite markers in proso millet (*Panicum miliaceum* L.). *Genes Genom* 2010;32(3):267–73. <https://doi.org/10.1007/s13258-010-0007-8>
- [3] Saleh ASM, Zhang Q, Chen J, Shen Q. Millet Grains: Nutritional Quality, Processing, and Potential Health Benefits: Millet grains.... *Comprehensive Reviews in Food Science and Food Safety* 2013;12(3):281–95. <https://doi.org/10.1111/1541-4337.12012>
- [4] Zhu F, Wang S. Physicochemical properties, molecular structure, and uses of sweetpotato starch. *Trends in Food Science & Technology* 2014;36(2):68–78. <https://doi.org/10.1016/j.tifs.2014.01.008>

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- [5] Cavers PB, Kane M. The Biology of Canadian Weeds: 155. *Panicum miliaceum* L. *Can J Plant Sci* 2016;96(6):939–88. <https://doi.org/10.1139/cjps-2015-0152>
- [6] Gyulai G, Humphreys M, Lagler R, Szabo Z, Toth Z, Bittsanszky a., Gyulai F, Heszky L. Seed remains of common millet from the 4th (Mongolia) and 15th (Hungary) centuries: AFLP, SSR and mtDNA sequence recoveries. *Seed Science Research* 2006;16(3):179–91. <https://doi.org/10.1079/ssr2006251>
- [7] Gyulai F. THE HISTORY OF BROOMCORN MILLET (*Panicum miliaceum* L.) IN THE CARPATHIAN-BASIN IN THE MIRROR OF ARCHAEOBOTANICAL REMAINS I. FROM THE BEGINNING UNTIL THE ROMAN AGE
Archaeobotany or palaeo-ethnobotany is the D ÄEULGJLQJ ´UROH ZLWKLQ WKH V \ VWHP RI VFLH. *Columella - Journal of Agricultural and Environmental Sciences* 2014;1(1):29–38
- [8] Kalinova J, Moudry J. Content and Quality of Protein in Proso Millet (*Panicum miliaceum* L.) Varieties 2006. <https://doi.org/10.1007/s11130-006-0013-9>
- [9] McSweeney MB, Seetharaman K, Ramdath DD, Duizer LM. Chemical and Physical Characteristics of Proso Millet (*Panicum miliaceum*)-Based Products. *Cereal Chemistry* 2017;94(2):357–62. <https://doi.org/10.1094/CCHEM-07-16-0185-R>
- [10] Kim JK, Park SY, Yeo Y, Cho HS, Kim YB, Bae H, Park CH, Lee Dr. JH, Park Dr. SU. Metabolic profiling of millet (*Panicum miliaceum*) using gas chromatography-time-of-flight mass spectrometry (GC-TOFMS) for quality assessment. *Plant OMICS* 2013;6(1):73–8
- [11] Wang Y, Bräutigam A, Weber APM, Zhu XG. Three distinct biochemical subtypes of C4 photosynthesis? A modelling analysis. *Journal of Experimental Botany* 2014;65(13):3567–78. <https://doi.org/10.1093/jxb/eru058>
- [12] Habiyaremye C, Matanguihan JB, D'Alpoim Guedes J, Ganjyal GM, Whiteman MR, Kidwell KK, Murphy KM. Proso Millet (*Panicum miliaceum* L.) and Its Potential for Cultivation in the Pacific Northwest, U.S.: A Review. *Frontiers in Plant Science* 2017. <https://doi.org/10.3389/fpls.2016.01961>
- [13] Baltensperger DD. Foxtail and proso millet. *Progress in New Crops: Proceedings of the Third National Symposium Indianapolis, Indiana, 22-25 October, 1996* 1996;(Hag 1989):182–90
- [14] Rajasekaran R, Francis N. Genetic and genomic resources for improving proso millet (*Panicum miliaceum* L.): a potential crop for food and nutritional security. *Nucleus (India)* 2021;64(1):21–32. <https://doi.org/10.1007/s13237-020-00331-2>
- [15] IBPGR. Descriptors for *Panicum miliaceum* and *P. sumatrense*. Rome, Italy: IBPGR; 1985
- [16] Hammer Ø, Harper DA, Ryan PD, others. PAST: Paleontological statistics software package for education and data analysis. *Palaeontologia Electronica* 2001;4(1):9
- [17] Vetriventhan M, Upadhyaya HD. Diversity and trait-specific sources for productivity and nutritional traits in the global proso millet (*Panicum miliaceum* L.) germplasm collection. *The Crop Journal* 2018;6(5):451–63. <https://doi.org/10.1016/j.cj.2018.04.002>
- [18] Nithyashree K, Vijayalaxmi KG. Study on Physical Properties of Minor Millets. *IJECC* 2023:156–62. <https://doi.org/10.9734/ijecc/2023/v13i11663>
- [19] Li K, Zhang T, Narayanamoorthy S, Jin C, Sui Z, Li Z, Li S, Wu K, Liu G, Corke H. Diversity analysis of starch physicochemical properties in 95 proso millet

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(*Panicum miliaceum* L.) accessions. *Food Chemistry* 2020;324:126863.
<https://doi.org/10.1016/j.foodchem.2020.126863>

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- [20] Trivedi AK, Arya L, Verma SK, Tyagi RK, Hemantaranjan A. Evaluation of barnyard millet diversity in central Himalayan region for environmental stress tolerance. *J Agric Sci* 2017;155(10):1497–507.
<https://doi.org/10.1017/S0021859617000545>
- [21] Upadhyaya HD, Ravishankar CR, Narasimhudu Y, Sarma NDRK, Singh SK, Varshney SK, Reddy VG, Singh S, Parzies HK, Dwivedi SL, Nadaf HL, Sahrawat KL, Gowda CLL. Identification of trait-specific germplasm and developing a mini core collection for efficient use of foxtail millet genetic resources in crop improvement. *Field Crops Research* 2011;124(3):459–67.
<https://doi.org/10.1016/j.fcr.2011.08.004>
- [22] Khairwal I, Yadav S, Rai K, Upadhyaya H, Kachhawa D, Nirwan B, Bhattacharjee R, Rajpurohit B, Dangaria C, others. Evaluation and identification of promising pearl millet germplasm for grain and fodder traits. *Journal of SAT Agricultural Research* 2007;5(1):1–6

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