



Estimation of Genetic Diversity among Bread Wheat (*Triticum aestivum* L. em. Thell) Genotypes

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Triticum aestivum L. em. Thell, known as bread wheat, is a vital staple crop globally, contributing significantly to caloric and protein intake. Its hexaploid nature, comprising three genomes (AA, BB, DD), resulted from natural hybridization, enhancing its agricultural significance. The advent of high-yielding cultivars during the Green Revolution drastically increased wheat yields, and its adaptability and self-pollinating characteristics further solidified its importance in food production. Genetic diversity within *Triticum aestivum* is crucial for improving traits such as stress tolerance and yield. This study highlights the necessity of estimating genetic variability among wheat genotypes, utilizing 24 genotypes. The study assesses the genetic parameters and diversity of various morpho-physiological traits in bread wheat genotypes, focusing on their variability and potential for genetic improvement. Key genetic parameters including the coefficient of variation (CV), genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), and genetic advance as a

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percentage of mean (GAM) were estimated for traits such as days to heading, plant height, peduncle length, and grain yield. Days to heading exhibited low variability, while plant height showed considerable genetic variation, indicating a good potential for improvement. The wheat genotypes were grouped into five distinct clusters based on Mahalanobis divergence and Tocher's method, revealing significant genetic diversity. Cluster I, comprising eighteen genotypes, displayed the highest intra-cluster distance, while Clusters III and V showed the greatest inter-cluster distance. Trait analysis across clusters highlighted variations in days to heading, plant height, grain yield, and other traits, emphasizing the genetic diversity and potential for selective breeding in wheat.

Keywords: Bread wheat; Mahalanobis diversity; cluster analysis; variability; genetic diversity.

1. INTRODUCTION

Triticum aestivum L. em. Thell, commonly known as bread wheat, is a crucial staple food globally, providing a significant portion of daily caloric and protein intake [1]. This hexaploid species, with three genomes (AA, BB, DD), originated through natural hybridization, which is key to its importance in agriculture [2,3]. The species' large chromosomes make it well-suited for genetic studies and observation of variations [4,5,6,7,8]. The implementation of high-yielding cultivars, such as those introduced during the Green Revolution, had a substantial impact on wheat yields [9,10]. Adaptability to diverse environments and its self-pollinating nature further emphasize its significance in food production [11]. The genetic diversity within *Triticum aestivum* plays a critical role in enhancing traits like abiotic stress tolerance [12]. Overall, *Triticum aestivum* stands as a cornerstone crop in global food security and agricultural research. Estimating variability and genetic diversity among bread wheat genotypes is crucial for several reasons. Firstly, genetic diversity provides the raw material for natural selection and adaptation to changing environments [13]. By assessing genetic diversity among bread wheat genotypes, researchers can identify variations that may confer traits such as drought tolerance, yield increase, and disease resistance [14]. This information is invaluable for wheat breeders as it allows them to select genotypes with desirable characteristics for further breeding programs [14]. Additionally, genetic diversity plays a vital role in the adaptability of bread wheat to different environmental conditions [13]. Studies have shown that increased genetic diversity in bread wheat enhances its ability to adapt divergently, ensuring its survival and productivity in various ecological niches [13]. Moreover, genetic diversity is essential for improving the overall quality of bread wheat [15]. For instance, the

Glu-score of individual wheat genotypes, which is influenced by genetic diversity, significantly contributes to the bread-making quality of wheat [15]. Understanding the genetic diversity among bread wheat genotypes allows for the selection of genotypes with optimal protein compositions that are crucial for baking quality [15]. Furthermore, genetic diversity is closely linked to the potential for adaptation to biotic and abiotic stresses [16]. Bread wheat germplasm with wide genetic diversity has a higher likelihood of withstanding various stresses, ensuring food security and sustainability in agricultural production systems [16]. Assessing genetic diversity among bread wheat genotypes also aids in the development of breeding programs aimed at enhancing specific traits such as drought tolerance [17]. Studies have utilized techniques like amplified fragment length polymorphisms (AFLPs) and microsatellite markers to evaluate genetic diversity and relationships among different genotypes, providing valuable insights for breeding programs [17,18]. By incorporating genetic diversity information, breeders can select genotypes with the desired traits and improve the overall resilience and productivity of bread wheat varieties [18]. Furthermore, genetic diversity analysis allows for the identification of stable genotypes with consistent performance across different environments [19]. Evaluating the stability and performance of advanced bread wheat genotypes helps in selecting varieties that exhibit reliable performance under varying conditions, contributing to sustainable agricultural practices [19]. Additionally, genetic diversity studies enable the classification of bread wheat genotypes into distinct groups based on morphological traits, facilitating targeted breeding efforts. Understanding the variability among genotypes aids in the efficient utilization of genetic resources for developing improved wheat varieties. This study was conducted to estimate variability and genetic diversity among 24 bread

wheat genotypes for use in various breeding programs, improving quality traits, ensuring adaptability to changing environments. By leveraging genetic diversity information among the present set of genotypes, researchers and breeders can develop resilient and high-performing wheat varieties that meet the demands of modern agriculture and contribute to global food security.

2. MATERIALS AND METHODS

2.1 Experimental site, Climate and Weather

The present investigation was carried out at the NEBCRC, Pantnagar, Uttarakhand, India. Twenty-four wheat genotypes were taken for present investigation (Table 1) and experiment was laid down in RBD with four replications. Each entry was planted in three-meter-long four rows plot. The rows were spaced 20 cm apart for timely sown and for late sown also. The 24 bread wheat genotypes were used for evaluation during *Rabi* 2016 and *Rabi* 2017. These genotypes were subjected to evaluation in a replicated trial using Randomized Block Design (RBD) at NEBCRC. The type of soil texture, which is generally 1.0 to 1.5m deep, high water table, shallow depth and calcareous nature are key features of the soil in this area. The favorable climatic conditions for normal growth of experimental crop include 15–30°C temperature throughout the crop duration with equitable distribution of rain.

2.2 Statistical Analysis

2.2.1 Estimation of variability parameters

To assess and measure the genetic diversity among the genotypes for the traits being examined, both genotypic and phenotypic variation coefficients were calculated according to the method proposed by Burton and De Vane, Sivasubramanian and Madhavamenon [20,21] classified GCV and PCV values as low, moderate and high on basis of following range: 0

to 10% - Low; 10 to 20% – Moderate; 20% and above: High. Genetic advance is estimated by selecting five per cent of the superior progeny and was calculated by using the following formula.

$$\text{Genetic Advance (GA)} = ih^2\sigma_p$$

The genetic advance as a percentage of the mean was classified into three categories: low, moderate, and high, as outlined by Johnson et al. [22]. Specifically, 0-10% was considered low, 10-20% was moderate, and above 20% was high.

2.2.2 Estimation of genetic divergence

Genetic divergence among 24 wheat genotypes was assessed by analyzing data for sixteen traits using methodology of Mahalanobis [23]. The steps involved were:

1. The genotypes were evaluated in replicated trials.
2. Observations were recorded for days to heading, days to maturity, grain filling duration, plant height (cm), spike length (cm), peduncle length (cm), tiller number per meter square, no. of spikelets per spike, no. of grains per spike, biological yield per plot (g), 1000 grain weight (g), grain yield per plot, harvest index (%), canopy temperature depression (CTD) and normalized difference vegetation index (NDVI). Variances and covariances were then calculated.
3. The D^2 values were calculated using a specific formula.

$$D^2 = W_{ij}(\bar{X}_i^1 - \bar{X}_i^2)(\bar{X}_j^1 - \bar{X}_j^2)$$

Where,

W_{ij} = represents the inverse of the estimated variance-covariance matrix.

$(\bar{X}_i^1 - \bar{X}_i^2)(\bar{X}_j^1 - \bar{X}_j^2)$ = represents the differences in the means of the two populations.

Table 1. Wheat genotypes used in experiment

Sl.	Genotype	Sl.	Genotype	Sl.	Genotype	Sl.	Genotype
1	Kenya TK18	7	Dharwar Dry	13	IC 252874	19	Ariana 66
2	Raj 4037	8	PBW 343	14	Sonora 64	20	STW 598874
3	Salembo	9	IC 212185	15	Jebelmara 131	21	C 306
4	Giza 155	10	BWL 0814	16	Karim	22	Giza168
5	T64 2W	11	Heines Peko	17	Redfife	23	HD 2967
6	Bacanora	12	Raj 3765	18	CusParula	24	Sunstar

4. To determine the contribution of each trait to overall divergence, the frequency with which each trait ranked first was calculated using the following

$$d_i = Y_i^j - Y_i^k$$

Where, d_i = is the mean deviation in the population, $Y_i^j - Y_i^k$ = Values for characters in population.

5. Genotypes were grouped into different clusters using Tocher's method [24]

6. The formula for calculating the average intra-cluster and inter-cluster distances was provided by Singh and Chaudhary [25].

The average intra-cluster distance was calculated as $= \sum D_i^2 / n$

Where, $\sum D_i^2$ = is the sum of distances between all possible combinations (n) within a cluster.

The average inter-cluster distance was estimated as $= \sum D_i^2 / (n_1 \times n_2)$

Where, $\sum D_i^2$ = is the sum of distances between all possible combinations of genotypes in the two clusters, and n_1 and n_2 are the number of genotypes in the first and second clusters, respectively.

7. A cluster diagram was created to illustrate the distances between clusters and genotypes based on the methods described above.

3. RESULTS AND DISCUSSION

3.1 Estimation of Genetic Parameters for Morphological and Physiological Characters

The genetic parameters for various morpho-physiological characters in bread wheat were assessed, highlighting the extent of variability and genetic potential within the genotypes. Key parameters considered include the coefficient of variation (CV), genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), and genetic advance as a percentage of the mean (GAM). These metrics provide insights into the potential for genetic improvement of the traits under study.

The trait of days to heading exhibits a low coefficient of variation (CV) at 1.03%, with moderate genotypic (GCV) and phenotypic (PCV) coefficients of variation at 10.08% and 10.13%, respectively. The genetic advance as a percentage of mean (GAM) is 20.55%, indicating a moderate scope for genetic improvement through selection. Similarly, days to maturity show very low CV (0.77%) and low GCV (5.16%) and PCV (5.21%), with a GAM of 10.46%. This suggests that while there is limited phenotypic variation, the genetic variation is sufficient to allow for some improvement in breeding programs. Plant height displays a CV of 1.74%, with moderate GCV (17.03%) and PCV (17.12%), and a substantial GAM of 34.73%. This indicates a broad range of genetic variation, making it a promising target for selection. Peduncle length, with a CV of 3.12%, GCV of 13.42%, PCV of 13.78%, and GAM of 26.78%, also shows considerable genetic variability, suggesting good potential for enhancement through selective breeding. The number of tillers per meter square has a CV of 6.61%, GCV of 11.38%, PCV of 13.20%, and GAM of 20.09%. This reflects moderate genetic variation and potential for improvement. Spike length, with higher CV (8.44%), low GCV (8.22%), and moderate PCV (11.77%), coupled with a GAM of 11.78%, suggests a modest scope for genetic gains through selection. Spikelets and Grains per Spike: The number of spikelets per spike shows a CV of 4.64%, lower GCV (5.39%), and PCV (7.11%), and a GAM of 8.38%, indicating limited genetic variation. In contrast, the number of grains per spike has a CV of 6.14%, GCV of 10.34%, PCV of 12.02%, and GAM of 18.24%, reflecting a broader genetic variability and better potential for improvement. Grain weight per spike demonstrates higher variability with a CV of 7.49%, GCV of 16.40%, PCV of 18.03%, and a significant GAM of 30.58%, suggesting a high potential for genetic enhancement. Similarly, 1000 kernel weight has a CV of 2.57%, GCV of 14.16%, PCV of 14.38%, and GAM of 28.56%, indicating substantial genetic variability and scope for improvement. Biological yield shows a CV of 6.83%, with higher GCV (16.69%) and PCV (18.00%), and a GAM of 31.73%, suggesting considerable genetic variability and potential for yield improvement. Grain yield per plot, with a CV of 5.80%, GCV of 13.60%, PCV of 14.75%, and GAM of 25.70%, indicates substantial genetic variation and good prospects for selection-based improvement. The harvest index displays a CV of 5.75%, higher GCV (19.47%) and PCV (20.31%), and a significant

GAM of 38.25%, indicating a wide range of genetic variability and high potential for genetic gains. Grain filling duration, with a CV of 3.58%, GCV of 8.12%, PCV of 8.87%, and GAM of 15.23%, reflects moderate genetic variation. Canopy temperature depression at various growth stages shows varying degrees of variability. At heading, it has a CV of 11.99%, GCV of 13.24%, PCV of 17.73%, and GAM of 20.25%. At anthesis, the CV is 13.27%, with GCV of 13.07%, PCV of 18.58%, and GAM of 18.86%. At maturity, the CV is 13.63%, GCV is 11.85%, PCV is 18.21%, and GAM is 15.81%. These values indicate moderate to high genetic variability. The normalized difference vegetation index (NDVI) at heading shows low CV (1.87%), GCV (2.03%), PCV (2.72%), and GAM (3.10%). At anthesis, the CV is 2.11%, GCV is 4.05%, PCV is 4.51%, and GAM is 7.44%. At maturity, NDVI exhibits higher CV (4.39%), GCV (8.50%), PCV (9.64%), and GAM (15.37%). These indices suggest moderate genetic variability and potential for selective breeding. Results of this study closely align with studies conducted by Alemu et al., Balkan, Bayisa et al., Ferede et al., Seyoum et al. [26–30].

3.2 Grouping of Wheat Genotypes into Different Clusters

The grouping of twenty-four wheat genotypes into five distinct clusters using Mahalanobis divergence and Tocher's clustering method [24] presents a detailed landscape of genetic diversity and trait variation. Cluster I, the largest, comprises eighteen genotypes: Raj 4037, Giza 155, T64 2W, Bacanora, Dharwar Dry, PBW 343, BWL 0814, Heines Peko, Raj 3765, IC 252874, Sonora 64, Jebelmara 131, Ariana 66, STW 598874, C 306, Giza 168, HD 2967, and Sunstar. Cluster II includes three genotypes: Redfife, Ariana 66, and STW 598874, showing some overlap with Cluster I. Clusters III, IV, and V are solitary, each containing a single genotype: IC 212185, Kenya TK18, and Salemo, respectively, highlighting their unique genetic profiles.

The intra-cluster distances (Table 2) reveal the homogeneity within each cluster, with Cluster I having the highest intra-cluster distance (513.13), indicating substantial variability among its members. Cluster II follows with a distance of 477.20. The solitary clusters III, IV, and V have an intra-cluster distance of 0.00, as expected due to the presence of a single genotype in each. Inter-cluster distances (Table 2) are essential for understanding the genetic divergence between clusters. The largest inter-cluster distance is between Clusters III and V (3915.35), followed by Cluster II and V (3425.19), indicating significant genetic dissimilarity between these groups. Conversely, Clusters I and II are the closest with a distance of 1651.30, suggesting relatively similar genetic makeup. Fig. 1 shows a dendrogram of genetic distances between genotypes.

Trait analysis of cluster mean across clusters (Table 3) shows variations in days to heading, with Cluster II having the longest duration (116.917 days) and Cluster I the shortest (94.806 days). Days to maturity follow a similar trend, with Cluster II again taking the longest (152.833 days) and Cluster I the shortest (137.403 days). Plant height varies significantly, with Cluster III exhibiting the tallest plants (133.75 cm) and Cluster IV the shortest (90.25 cm). Peduncle length is highest in Cluster III (45.625 cm) and lowest in Cluster IV (34.625 cm). Tiller number per meter square ranges from 1030 in Cluster IV to 669 in Cluster V. Spike length is relatively consistent across clusters, with minor variations, the highest being in Cluster IV (10.393 cm) and the lowest in Cluster III (9.5 cm). The number of spikelets per spike peaks in Clusters II and V (20) and is lowest in Cluster III (18). Grain yield traits also vary, with the number of grains per spike highest in Cluster V (55.5) and lowest in Cluster III (41.5).

Genotype serial numbers are used instead of names. Names can be identified with the serial numbers of Table 1.

Table 2. Intra cluster (diagonal) and inter-cluster distances for five clusters in wheat

	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
Cluster I	513.13	1651.30	1404.87	1910.73	1282.14
Cluster II		477.20	917.29	1253.95	3425.19
Cluster III			0.00	2801.32	3915.35
Cluster IV				0.00	2929.92
Cluster V					0.00

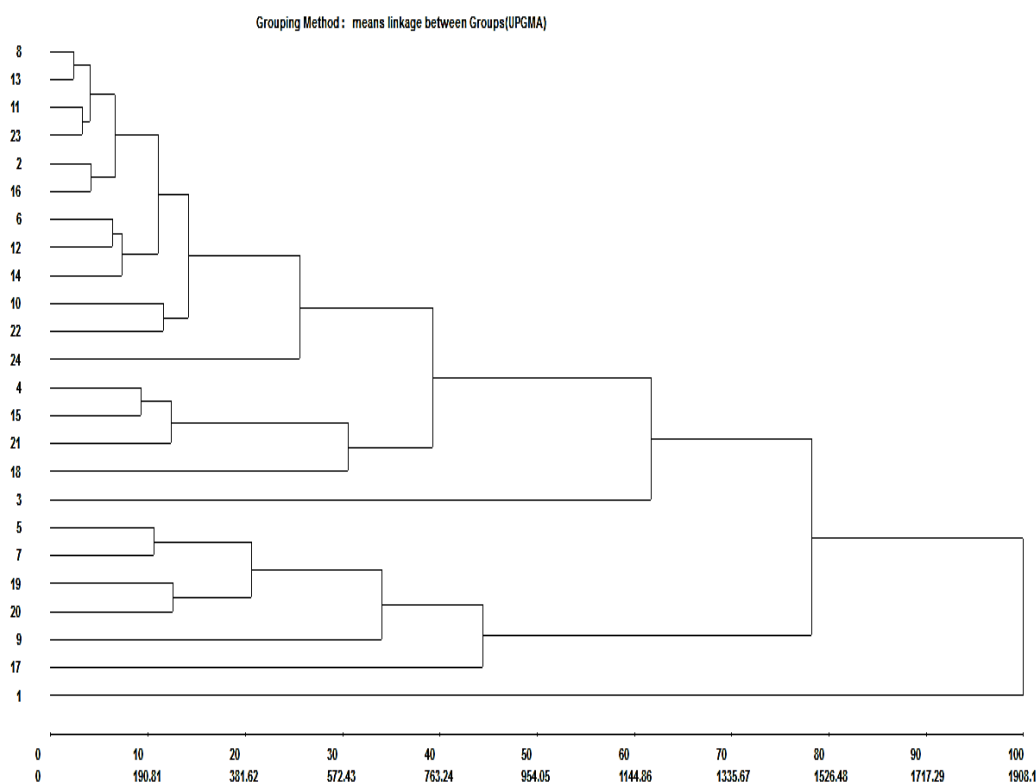


Fig. 1. Dendrogram obtained from mahalanobis Diversity matrix

Table 3. Per se performance of diverse clusters with respect to different traits

	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
Days to heading	94.806	116.917	98	122	97
Days to maturity	137.403	152.833	141	158	136.75
Plant height	103.507	130.575	133.75	90.25	92.375
Peduncle length	42.028	41.042	45.625	34.625	36
Tiller number per meter square	816.542	943	871	1030	669
Spike length	10.129	9.917	9.5	10.393	10
No. of spikelets per spike	19.222	20	18	21	20
No. of grains spike	49.014	44.583	41.5	43	55.5
Grain weight per spike	1.859	1.683	1.49	1.53	2.2
1000 Kernel weight	43.865	33.287	33.175	39.35	46.51
Biological yield	2880.556	2482	2913	1961	1756.25
Grain yield per plot	1125.944	886.167	780	790	1132
Harvest index (%)	39.391	38.789	26.788	40.28	64.74
Grain filling duration	42.597	35.917	43	36	39.75
CTD heading	4.729	4.4	4.3	5.5	4.8
CTD anthesis	4.729	4.4	4.3	5.5	4.8
CTD maturity	1.36	1.15	0.925	1.325	1.475
NDVI heading	0.783	0.784	0.788	0.76	0.79
NDVI anthesis	0.713	0.708	0.698	0.668	0.738
NDVI maturity	0.225	0.241	0.2	0.22	0.225

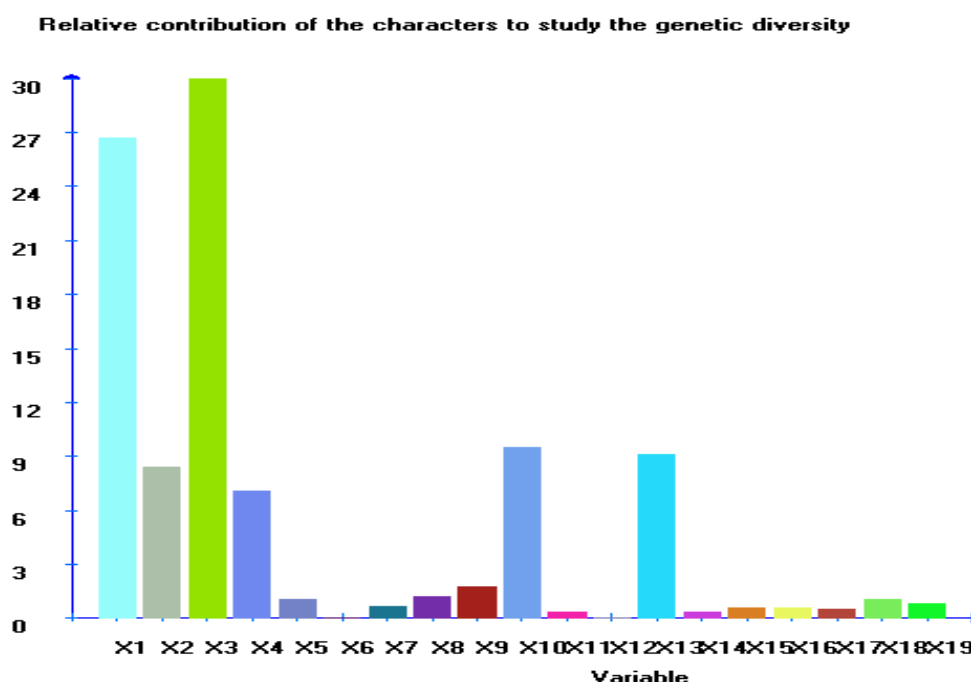


Fig. 2. Graphical presentation of characters showing contribution to total divergence. Where, X1-Days to heading, X2-Days to maturity, X3-Plant height, X4-Peduncle length, X5-Tiller number per meter square, X6-Spike length, X7-No. of spikelets per spike, X8- No. of grains per spike, X9-Grain weight per spike, X10-1000 grain weight , X11-Biological yield , X12-Grain yield per plot, X13-Harvest index (%), X14-CTD heading, X15-CTD anthesis, X16-CTD maturity, X17-NDVI heading, X18-NDVI anthesis, X19-NDVI maturity

Grain weight per spike follows a similar pattern, highest in Cluster V (2.2 g) and lowest in Cluster III (1.49 g). The 1000 kernel weight is greatest in Cluster V (46.51 g) and least in Cluster II (33.287 g). Biological yield is highest in Cluster III (2913 g) and lowest in Cluster V (1756.25). Grain yield per plot ranges from 1132 in Cluster V to 780 in Cluster III. The harvest index is highest in Cluster V (64.74%) and lowest in Cluster III (26.788%). Grain filling duration is longest in Cluster III (43 days) and shortest in Cluster II (35.917 days). Canopy temperature depression (CTD) at heading, anthesis, and maturity shows minimal variation across clusters. NDVI values at heading, anthesis, and maturity also exhibit slight differences, indicating uniformity in vegetative vigor and greenness among the clusters. Results of diversity and cluster analysis closely align with the results of Bisht et al., Datta et al., Siddique et al., Suchitra and Lal, Tapaswini et al. [31–35] in wheat and other crop species.

The contribution of various traits (Fig. 2) to genetic diversity shows that plant height (30.08%) and days to heading (26.77%) are the most significant contributors. Other traits such as 1000 kernel weight (9.53%), harvest index

(9.13%), and days to maturity (8.43%) also play crucial roles. Lesser contributions come from traits like spike length (0.03%) and grain yield per plot (0.0001%).

4. CONCLUSION

The comprehensive analysis of genetic parameters and diversity in bread wheat genotypes underscores the substantial variability and genetic potential for improvement in key traits. The low variability in traits like days to heading (CV: 1.03%, GAM: 20.55%) and maturity (CV: 0.77%, GAM: 10.46%) contrasts with the high genetic variation observed in plant height (CV: 1.74%, GCV: 17.03%, GAM: 34.73%), peduncle length (CV: 3.12%, GCV: 13.42%, GAM: 26.78%), and grain yield per plot (CV: 5.80%, GCV: 13.60%, GAM: 25.70%), indicating targeted areas for genetic enhancement. The clustering of genotypes into distinct groups based on Mahalanobis divergence and Tocher's method highlights the significant genetic diversity present. The highest intra-cluster variability observed in Cluster I (513.13) and the considerable inter-cluster distances, particularly between Clusters III and V (3915.35), suggest

diverse genetic backgrounds among the genotypes. Trait analysis across clusters reveals significant differences, particularly in days to heading (Cluster I: 94.806 days, Cluster II: 116.917 days), plant height (Cluster III: 133.75 cm, Cluster IV: 90.25 cm), and grain yield (Cluster V: 1132, Cluster III: 780), which are critical for breeding programs. The findings provide valuable insights into the genetic structure of wheat populations, guiding future breeding efforts to improve yield and other agronomic traits through selective breeding.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models and text-to-image generators have been used during writing or editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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