



Evaluate the Genetic Divergence and Principal Component Analysis in Bread Wheat (*Triticum aestivum* L.)

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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

The current investigation was carried out on some bread wheat (*Triticum aestivum* L.) genotypes throughout spring season to evaluate their heat tolerance via Cluster Analysis and principal component analysis (PCA). The experiment was accomplished in an augmented block design with 60 genotypes and three replications. Evaluations were carried out on 26 quantitative traits. Cluster

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analysis showed five clusters, cluster I with 56 genotypes and clusters II, III, IV and V with only one genotype each. The clusters II, III, IV and V have only one genotype each, so their intra-cluster distances were zero. The intra-cluster distance for cluster I was 57.879. The maximum and minimum inter cluster distance was found between cluster II and III (267.377) and between cluster I and II (86.469), respectively. Cluster I showed the earliest (76.689 days) average for early maturity (days to 50% heading) and cluster III showed the maximum (27.664) average for grain yield (grain yield per plant). PCA indicated that the five principal components (PC1 to PC5) accounted for 65.61% of the total variance. PC1 accounted for 11.51% of the total variance and showed positive factor loading for almost traits. Harvest index, grain yield per plant, flag leaf width and leaf rolling showed the highest factor loadings for PC1. As a result of the foregoing data and analysis, it is possible to conclude that there is great potential for effective genetic improvement for grain yield and correlated traits in the present wheat genotypes.

Keywords: *Wheat; genetic divergence; cluster analysis; morphological traits.*

1. INTRODUCTION

Wheat (*Triticum aestivum* L., $2n=42$) is the most important cereal in the world. Wheat belongs to the family Poaceae (Gramineae) and tribe Triticeae containing more than 15 genera and 300 species including wheat and barley. *T. aestivum* is a segmental allohexaploid ($2n = 6x = 42$, AABBDD) originated in the Fertile Crescent area of South-Western Asia its geographical centre of origin and spreaded globally for cultivation and consumption. It is an allohexaploid (AABBDD, $2n = 6x = 42$) that arose by hybridization between a cultivated tetraploid wheat *T. turgidum* (AABB, $2n = 4x = 28$) and the wild goatgrass *Aegilops tauschii* (DD, $2n = 2x = 14$).

The nutritional composition of the wheat grain varies somewhat with differences in climate and soil. On an average, the kernel contains 10-12 per cent water, 65-70 per cent carbohydrates, 9-12 per cent protein, 1-2 per cent fat, 1-1.8 per cent minerals, and 2-2.2 per cent crude fibres. Thiamine, riboflavin, niacin, and small amounts of vitamin A are present, but the milling processes removes most of those nutrients with the bran and germ.

The yield and productivity of wheat are seriously threatened by high temperatures. In India, wheat cultivation suffers significant injuries each year due to high temperature stress [1]. As wheat is a crop that grown in the winter, it needs an extended period of low temperatures to attain highest grain. According to Wahid et al. [2] and Sareen et al. [3] high temperature stress is the outcome of a temperature increase that lasts longer than a threshold and has the potential to permanently harm plant growth and physiological development.

In the hybridization programme, D2 statistics analysis is used to select genetically dissimilar parents. P.C. Mahalanobis introduced the concept of D2 statistics in 1936. Rao utilised this approach to estimate genetic divergence in plant breeding. It is used to assess the degree of diversity and identify the relative contribution of each component characteristic to overall divergence. Genetic divergence is important in plant breeding because hybrids from different lines exhibit more heterosis than closely related parents. Principal Component Analysis (PCA) is a statistical technique that transforms a set of correlated variables into a smaller set of uncorrelated variables, called principal components. These components retain most of the variance of the original data, allowing for a simplified analysis without losing important information. This experiment was undertaken to assess genetic divergence among wheat genotypes using cluster analysis and PCA to select desirable parents in hybridization programmes.

2. MATERIALS AND METHODS

The current investigation entitled was conducted at three different locations; namely

1. Crop research rm, Nawabganj (UP)
2. Crop research farm, Araul (UP)
3. Crop research farm, Daleep Nagar (UP).

during crop season of Rabi 2021-2022 and 2022 2023 year under normal (non-stressed) and late sown (heat-stressed) conditions. The field experiments were planted comprised 60 germplasm of bread wheat. Furthermore, the our experimental trials were conducted in 12 environments (E1 to E12), which included six non-stressed (NS) and six heat-stressed (HS)

List 1. Details of bread wheat accessions used in the study

Sr. No.	Genotypes	Sr. No.	Genotypes
1.	K-1711	31.	K-2105
2.	K-1903	32.	K-2109
3.	K-1805	33.	K-0307
4.	K-1907	34.	K-0607
5.	K-1910	35.	K-1803
6.	K-2003	36.	K-1317
7.	K-0306	37.	PBW-852
8.	K-0402	38.	DBW-173
9.	K-2107	39.	HD-3388
10.	K-2121	40.	HD-2359
11.	K-8962	41.	K-9644
12.	K-9351	42.	K-2101
13.	K-9465	43.	KRL-213
14.	K-8027	44.	KRL-19
15.	K-2103	45.	PBW-826
16.	K-1006	46.	DBW-187
17.	K-1616	47.	HD-3392
18.	K-1905	48.	HD-2967
19.	K-1809	49.	DBW-107
20.	K-1908	50.	DBW-222
21.	K-2001	51.	PBW-833
22.	K-2007	52.	HD-3399
23.	K-9107	53.	PBW-835
24.	K-9162	54.	KRL-210
25.	K-9533	55.	KRL-1-4
26.	K-2108	56.	K-2010
27.	K-9423	57.	KRL-283
28.	K-8434	58.	DBW-350
29.	K-7903	59.	HD-3086
30.	K-2104	60.	WH-1142

environments. The field experiments were laid out in Augmented Block Design (ABD). Each experimental plot consisted of three rows of 2m length by 20 cm between rows covering an area of 2 m x 0.20m x 3 (1.2 m²) under irrigated condition. The observations on 26 physio-morphological and grain yield related traits were recorded viz., Days to 50% flowering(D50H), Number of spikelets per spike (NSPS), Plant height (cm) (PH), Physiological maturity (days) (PM), Spike bearing tillers per plant (SBTPP), Plant biomass (g) (PB), Peduncle length (cm) (PL), Number of grains per spike (NGPS), Flag leaf length (cm) (FLL), Number of grains per plant (NGPP), Flag leaf width (cm) (FLW), Grain length(mm) (GL), Flag leaf area (cm²) (FLA), Grain width(mm) (GW), Chlorophyll content (CC), LW ratio of grains (GLWR) , Canopy temperature depression (°C) (CTD), Grain yield /plant(g) (GYP), Plant waxiness (0-10) (PW), Harvest index (%) (HI), Leaf rolling (0-10 scale) (LR), 1000- grain weight(g) (GW), Grain filling

period (days) (GFP), Protein content (%) (PC), Spike length (cm) (SL), Gluten content (%) (GC).

3. RESULTS AND DISCUSSION

3.1 Genetic Divergence

The F test (ANOVA) for genotype was significant for all traits, indicating the presence of genetic variability among the genotypes.

Supported by the pseudo-F-test, the 60 genotypes were grouped into 5 non-overlapping clusters. The distribution of 60 bread wheat genotypes in 5 clusters is presented in Table 1.

The clustering pattern of the sixty genotypes were grouped into five different non-overlapping cluster. Cluster I had highest number of genotypes (56) followed by cluster II (1), cluster III (1), cluster VI (1), cluster V (1). This indicated presence of considerable diversity in the

genotype. The main groups in the genetic divergence analysis typically comprised genotypes of different origins. However, genotypes from the same origin or geographical area were also shown to be clustered together. The instance of grouping of genotypes of different origin or geographic region in same cluster were frequently observed. This implied that there is no correlation between genetic and geographic diversity. The estimates of intra and inter-cluster distance represented by D^2 values are given in Table 2. The minimum intra cluster distance (0.00) was found in cluster II, cluster III, cluster IV, cluster V and maximum was found for cluster I (57.879). The maximum inter-cluster distance was found between cluster II to III (267.377) followed by cluster III to IV (201.540), cluster II to V (172.747), cluster I to I (142.980), cluster IV to V (137.586). The minimum inter-cluster D^2 value found in case of cluster I to II (86.469) followed by cluster I to IV (86.769), cluster I to IV (88.334), cluster II to IV (101.141), cluster III to V (112.853). The higher inter-cluster distance indicated greater genetic divergence between the genotypes of those clusters, while lower inter-cluster values between the clusters suggested that the genotypes of the clusters were not much genetically diverse from each other.

These results are in close conformation with the findings of Rahman et al. [4] and Kumar et al. [5] Pandey et al. [6] and Abdelghany et al. [7].

A look on table 3 showed that cluster means for the different traits indicated considerable differences between the clusters. The entire cluster from cluster I to cluster V had average mean performance for most of the characters.

Cluster I showed earliest mean value for day to 50 per cent flowering (76.689 day), cluster II

showed earliest mean value for the grain filling period (29.536 day), cluster II showed maximum mean value for flag leave length (23.579), cluster III showed maximum mean value for flag leave width (1.988), cluster II showed maximum mean value for flag leave area (46.832) cluster IV showed maximum mean value for plant height (101.590), cluster III showed maximum mean value peduncle length (36.941), cluster V showed maximum mean value for spike bearing tiller per plant (12.415), cluster III showed maximum mean value for chlorophyll content (30.244), cluster I maximum mean value for canopy temperature depression (7.864), cluster III showed maximum mean value for plant waxiness (5.694), cluster II showed maximum mean value for leave rolling (5.596), cluster I showed maximum mean value for spike length (10.768), cluster I showed maximum mean value for no. of spikelet per spike (20.413), cluster III showed maximum mean value for physiological maturity (117.984), cluster I showed maximum mean value for plant biomass (51.717), cluster V showed maximum mean value for no. grain per spike (46.519), cluster III showed maximum mean value for no. of grain per plant (489.685), cluster I showed maximum mean value for grain length (6.778), cluster V showed maximum mean value for grain width (2.869), cluster I showed maximum mean value for length/width ratio (2.439), cluster I showed maximum mean value for test weight (39.125), cluster III showed maximum mean value for grain yield per plant (27.664), cluster III showed maximum mean value for harvest index (55.976), cluster IV showed maximum mean value for protein content (12.203), cluster III showed maximum mean value for gluten content (7.263). These results are in close conformation with the findings of Khalid et al. [8] Abdelghany et al. [7] and Khalid et al. [9].

Table 1. Cluster analysis

Clusters	Size	Genotypes
I	56	DBW-350, PBW-833, HD-3399, KRL-213, KRL-1-4, DBW-173, K-0402, WH-1142, KRL-19, K-2104, K-2108, KRL-283, K-1908, HD-2359, K-8962, K-9533, K-1907, DBW-107, PBW-826, K-1711, K-2109, K-0607, K-0306, K-1809, PBW-852, K-2101, K-1805, PBW-835, K-1317, K-2105, KRL-210, DBW-187, K-1910, K-2103, HD-3086, K-2010, K-9644, K-9351, HD-3388, HD-2967, K-1905, K-2007, DBW-222, K-9162, K-1616, HD-3392, K-2107, K-2001, K-1006, K-1903, K-7903, K-1803, K-9423, K-8027, K-0307, and K-9107
II	1	K-2003
III	1	K-2121
IV	1	K-8434
V	1	K-9465

Table 2. Average inter and intra cluster distance

	cluster 1	cluster 2	cluster 3	cluster 4	cluster 5
cluster I	57.879	86.469	142.980	88.334	86.769
cluster II		0.000	267.377	101.141	172.747
cluster III			0.000	201.540	112.853
cluster IV				0.000	137.586
cluster V					0.000

Table 3. Average cluster mean for 26 traits

	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
D50H	76.689	76.931	77.027	76.774	76.856
GFP	29.685	29.536	29.573	29.560	29.555
FLL	23.033	23.579	23.249	22.882	23.009
FLW	1.981	1.964	1.988	1.950	1.973
FLA	45.790	46.832	46.788	45.154	45.999
PH	100.009	100.288	100.975	101.590	99.925
PL	36.309	35.957	36.941	36.451	35.704
SBTPP	12.305	12.272	12.212	12.294	12.415
CC	29.556	29.317	30.244	29.534	29.935
CTD	7.864	7.783	7.557	7.756	7.581
PW	5.446	5.631	5.694	5.626	5.726
LR	5.425	5.596	5.583	5.335	5.375
SL	10.768	10.726	10.483	10.662	10.661
NSPS	20.413	20.202	20.098	20	20.057
PM	117.199	117.559	117.984	117.841	117.705
PB	51.717	50.657	50.987	51.604	50.077
NGPS	44.391	44.793	44.343	43.901	46.519
NGPP	425.896	441.773	489.685	455.669	467.843
GL	6.778	6.746	6.710	6.740	6.716
GW	2.813	2.835	2.849	2.844	2.869
GLWR	2.439	2.411	2.383	2.398	2.365
TW	39.125	38.555	38.568	38.863	38.413
GYPP	27.006	27.364	27.664	27.401	27.142
HI	53.767	55.777	55.976	54.923	55.722
PC	11.998	12.083	12.175	12.203	12.226
GC	7.255	7.252	7.263	7.257	7.247

4. PRINCIPAL COMPONENT ANALYSIS

In the present study, the first eight PC accounted for 65.62% (Table 4). Besides that, one of the premises of PCA is that the first two PC must account for at least 80% of the total variance.)

PC1 accounted for 11.51 % of the total variance and almost all studied characters showed positive loading in this principal component. The factor loading of principal components showed that, PC1 accounted maximum variability for characters like harvest index, grain yield per plant, flag leaf width, leaf rolling, protein content, chlorophyll content, grain L/W ratio, days of 50% heading, grain length, gluten content and grain yield per plant. These results agreed in Khodadadi et al. [10] Rymuza et al. [11] Sareen

et al. [12] Hamam et al. [13] Adilova et al. [14] Farheen et al. [15] Bhatti et al. [16] Khalid et al. [8] and Kumar et al. [17].

In the present investigation, PCA was performed for twenty-six yield and yield component traits in wheat lines. The principal components with eigenvalues more than 1 and which explained at least 5 per cent of the variation in the data should be considered. Eigen value measures the amount of variation explained by a particular factor out of the total variation. The factor loading of a principal component is the correlation between the scores determined by this principal component and the values or means of each original variable. The eigen values from PCA determines the number of factors to be retained which accounts for most of the variability in the

original data set. The principal components with higher eigen values and variables which had high factor loading were considered as best representative of system attributes. The sum of all eigen values is always equal to the number of variables.

In our study, first eight principal components had eigen value greater than one and they cumulatively explained 65.61 per cent of the total variation present in the original data set. So, these eight principal components were considered important for further explanation. The first principal component explained 11.51 per cent while, the second, third, fourth, fifth, sixth, seventh and eighth principal component exhibited 10.03 per cent, 9.65 per cent, 8.81 per cent, 7.81 per cent, 6.48 per cent, 5.99 per cent and 5.30 per cent variability, respectively among the lines for the traits under study (Table 4). The first principal component accounts for as much of the variability in the data as possible, and each succeeding component accounts for as much of the remaining variability as possible.

Scree plot explains the percentage of variation associated with each principal component and is

obtained by drawing a graph between principal component numbers (X- axis) and percentage of variation explained (Y-axis). The Principal Component 1 showed 11.51 per cent variability with eigen value 2.99 which then declined gradually. From the graph, the maximum variation was observed in Principal Component 1.

The result of the PCA explained the genetic divergence of wheat lines. Eigen values assess the importance and role of each component to total variation, while the factor loading indicates the scale of contribution of every origin variable with which each principal component is associated. Within each principal component, only highly loaded factors or traits were retained for further explanation. Component matrix revealed that Principal Component 1 showed high positive loading for harvest index (0.435), grain yield per plant (0.293), flag leaf width (0.214), leaf rolling (0.195), protein content (0.174) and chlorophyll content (0.159). Principal Component 2 enabled high positive loading for grain LW ratio (0.499), days of 50% heading (0.351), grain length (0.245), gluten content (0.268) and grain yield per plant (0.231). The

Table 4. Principal component analysis for yield and yield related traits

	Eigen value	Variance percent	Cumulative variance percent
PC1	2.993	11.513	11.513
PC2	2.608	10.033	21.547
PC3	2.509	9.653	31.200
PC4	2.291	8.814	40.015
PC5	2.032	7.818	47.833
PC6	1.687	6.489	54.323
PC7	1.557	5.992	60.315
PC8	1.378	5.302	65.618
PC9	1.199	4.611	70.229
PC10	1.101	4.237	74.467
PC11	1.001	3.853	78.320
PC12	0.811	3.119	81.439
PC13	0.809	3.113	84.553
PC14	0.673	2.588	87.142
PC15	0.612	2.354	89.497
PC16	0.565	2.173	91.670
PC17	0.499	1.920	93.591
PC18	0.397	1.529	95.120
PC19	0.357	1.376	96.496
PC20	0.292	1.126	97.623
PC21	0.274	1.057	98.680
PC22	0.197	0.759	99.440
PC23	0.116	0.446	99.886
PC24	0.014	0.055	99.942
PC25	0.011	0.041	99.984
PC26	0.004	0.015	100.000

Table 5. Factor loadings of principal components

	PC1	PC2	PC3	PC4	PC5
D50H	0.000	0.351	0.000	0.128	0.000
GFP	0.123	0.000	0.000	0.152	-0.144
FLL	0.313	0.000	0.393	0.000	0.000
FLW	0.214	0.000	0.129	-0.278	0.184
FLA	0.371	0.000	0.380	-0.181	0.117
PH	-0.265	0.000	-0.146	-0.349	0.000
PL	-0.107	-0.119	0.000	-0.527	0.000
SBTPP	0.000	0.000	0.117	0.000	-0.103
CC	0.159	-0.242	0.000	0.000	0.000
CTD	-0.109	-0.185	0.286	0.000	-0.288
PW	0.000	0.000	-0.135	0.000	-0.211
LR	0.195	0.000	0.000	0.161	-0.207
SL	-0.141	0.000	0.247	0.274	0.000
NSPS	0.000	0.000	0.415	0.171	0.270
PM	0.000	0.122	-0.309	0.238	0.000
PB	-0.312	0.126	0.187	0.213	0.241
NGPS	0.195	0.000	-0.161	0.000	0.307
NGPP	0.167	-0.128	-0.216	0.000	0.281
GL	0.000	0.245	0.169	0.000	-0.367
GW	0.000	-0.447	0.000	0.249	0.000
GLWR	0.000	0.499	0.000	-0.224	-0.174
TW	-0.190	0.104	0.000	-0.214	0.228
GYPP	0.293	0.231	-0.119	0.141	0.000
HI	0.435	0.000	-0.210	-0.111	-0.235
PC	0.174	0.166	0.000	0.000	0.123
GC	0.000	0.268	0.000	0.000	0.340

prominent traits contributing maximum variability and desegregating in different principal components have the tendency to remain together which may be kept into consideration during utilization of these characters in crop improvement programme as a donor for the associated traits.

5. CONCLUSION

Based on the above result of genetic diversity with the help of principle component analysis it could be concluded for all characteristics Cluster I had highest number of genotypes (56). The minimum intra cluster distance (0.00) was found for II to V and maximum was found for cluster I (57.879). The maximum inter-cluster distance was found between cluster II to III (267.377). The minimum inter- cluster D² value found in case of cluster I to II (86.469). Cluster I showed earliest mean value for day to 50 per cent flowering

(76.689 day) and most important character grain yield per plant cluster III showed maximum mean value for (27.664). Principal component analysis (PCA) indicated that the five principal components (PC1 to PC5) showed 65.61 per cent of the total variability. Thus, this finding indicated that these traits could utilize in various breeding as well as improvement programmes. The information may further help the breeder in formulation appropriate strategy aimed at getting higher yield and character improvement in wheat.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

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

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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