

## Study of phenological diversity of wheat genotype by discriminant function analysis

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### ABSTRACT

High yield potential genotypes are required to increase the cultivation and production of wheat in Bangladesh. With a view to identify such genotypes, this study was conducted consisting 170 wheat (*Triticum aestivum* L.) genotypes. Seventeen quantitative variables were included in the descriptive analysis of which eight were considered in the multivariate analysis i.e. cluster analysis, discriminant function analysis (DFA). A wide range of variation was found for all most all the quantitative plant characters studied. According to the correlation study between plant characters showed that plants tend to emerge earlier in the phenological cycle become earlier in producing flowers and seed yield is higher in early emergence plants. Seed yield of wheat is the product of effective tiller plant<sup>-1</sup>, 1000 seed weight and harvest index. The genotypes were grouped into seven clusters using K-mean clustering. Maximum number of genotypes (52) was concentrated in cluster 6 followed by cluster 4 (29), 1 (23), 7 (23), 2 (19), 5 (17) and 3 (7). The clustering pattern of the genotypes revealed that genotypes under cluster 6 and cluster 7 were early flowering, early days to 50% flowering and early maturing with higher number of tiller plant<sup>-1</sup>. They also possess highest seed yield compared to other clusters. Classification matrix showed that 98.2% of the genotypes were correctly assigned to clusters. The genotype no 142 (2344) in group 1, the genotype no 94 (2080) in group 2, the genotype no 68 (1990) in group 3, the genotype no 46 (1688) in group 4, the genotype no 81 (2027) in group 5, the genotype no 165 (2481) in group 6 and the genotype no 53 (1751) in group 7 might be considered as more representative of their respective groups. The result clearly showed that the genotypes under group 6 and group 7 were early emergence, early flowering and produced higher seed yield. From the result it was observed that harvest index was placed at the top of the list of discriminatory variables with correlation coefficient of 0.943 under function 1. It indicates that harvest index played the most dominant role out of 8 variables in explaining the maximum variance in 170 genotypes by stepwise DFA.

### INTRODUCTION

The importance of genetic resources in breeding selection, agriculture, and ecology has been highlighted in recent years (Anonymous, 1999; Stoyanova et al., 1998). Genetic variety in common and durum wheat has decreased as a result of conventional breeding methods, which has limited the possibility of increasing production (Hadjiivanova et al., 2010). There is a greater need for new genetic material when wheat approaches its biological production limits (Hailegiorgis, 2011; Graybosch and Peterson, 2010; Lanning et al., 2010).

Wheat (*Triticum aestivum* L.) holds a prominent position among cereal crops worldwide, and in Bangladesh, it ranked 30th in wheat production in 2020 (World Agricultural Production 2020/2021). Despite occupying only about 4% of the total cropped area and 11% during the Rabi season, wheat contributes 7% to the total output of food cereals. To meet the increasing demand for food cereals in Bangladesh, efforts are being made to develop improved wheat varieties and cultivation practices with high yield potential that lower farmers' production costs. Hassan et al. (1998) reported significant variation in grain yield of

wheat genotypes grown under different management practices. Following the 2007 Intergovernmental Panel on Climate Change report, Islam (2009) and Poulton & Rawson (2011) stated that temperature in Bangladesh is increasing and will likely impact the future wheat productivity. Bangladesh already faces drought in the northwestern region and it is expected that the moderately drought affected areas will become severely drought prone areas within the next two decades.

The introduction of Mexican varieties like 'Sonora 64' and 'Kalyansona' in collaboration with CIMMYT in 1965 marked the initial stages of wheat cultivation in Bangladesh, driving its expansion in the region (BARI 2010). However, Bangladesh's heavy dependence on wheat imports by the time of independence in 1971, alongside shifting dietary preferences favoring wheat as a staple food, highlighted the need for local production. The release of 'Sonalika' in 1972 revolutionized wheat production with its fast maturation and high yield, reaching 80% adoption in the early 1980s (WRC 2009). In 1983, WRC-BARI introduced four more high-yielding varieties ('Ananda', 'Kanchan', 'Barkat', and 'Akbar') yielding between 2 to 3 tons ha<sup>-1</sup>, with 'Kanchan' emerging as the predominant variety by the early 1990s, gradually replacing 'Sonalika'. Additionally, 'Aghrani' and 'Protiva', two other high-yielding varieties, were endorsed by BNSB in 1987 and 1993, respectively. Wheat is an especially critical "staff of life" for the approximately 1.2 billion "wheat-dependent" to 2.5 billion "wheat-consuming" poor-men, women and children-who live on less than \$US 2/day (FAOSTAT, 2010). At the same time, climate change-induced temperature increases are likely to reduce wheat production in developing countries (where around 66% of all wheat is produced) by 20-30% (Esterling et al., 2007; Lobell et al., 2008; Rosegrant and Agcaoili, 2010).

Little research has been done in Bangladesh to find wheat cultivars that can help increase productivity. The need to swiftly disseminate stress-tolerant cultivars to growers and incorporate them into breeding programmes is highlighted by the increasing frequency of climatic threats. As such, focused research and the preservation of the

existing wheat gene pool are of utmost importance. The National Gene Bank is a vital domestic and international genetic resource that keeps a variety of wheat collections, including mutants from BARI Gom-26 (Odzhakova et al., 2007; Kolev and Stoyanova, 2005; Popova, 2003). Creating a database to measure plant genetic diversity begins with a thorough assessment of these collections (Angelova and Popova, 1998).

A comprehensive analysis was conducted on 170 wheat genotypes, focusing on 17 quantitative variables, with eight variables selected for multivariate analysis including discriminant function analysis (DFA), Mahalanobis distance, Classification matrix, Representative genotypes, Significant variations were observed in all studied plant characteristics. This variation signifies the immense potential and the assessment of crop diversity functions as an exceptional starting point for crop enhancement, as it furnishes a framework and a roadmap for selecting parental lines and devising a breeding program. Selecting the superior genotype from an extensive genetic pool based on their phenotypic expression poses a formidable challenge. Correlation analysis indicated that early emergence correlated with early flowering and higher seed yield. DFA further supported these findings, with harvest index playing a dominant role in grouping genotypes. Discriminatory functions effectively classified genotypes into clusters, with 98.2% accuracy.

The present investigation was conducted to evaluate the genetic diversity of 170 mutationally induced wheat genotypes through the implementation of multivariate analysis in order to effectively manage and incorporate them into breeding programs. Furthermore, this study aimed to analyze the variance components of these genotypes, with the intention of guiding future breeding programs for wheat. The outcomes of this study will be valuable in advancing genetic research efforts focused on enhancing wheat by utilizing agronomic traits.

## MATERIALS AND METHODS

### Plant Materials

One seventy accessions of Bari Gom-26 formed by mutations by ACI were the treatment variables in the experiment. A list of genotypes with their accession number and origin is given in Table 1. Seeds were collected from the Genetic Resources Unit of the ACI. The study was conducted with a view to evaluate all the accessions for their yield performance under irrigated condition. This chapter includes short description of location and site, soil, climate and weather, planting material, preparation of field, sowing of seeds, application of fertilizers, intercultural operations, data collection, procedure of recording data, data analysis etc.

### Field Experiment Site

The investigation took place at an elevation of 8.4 meters above sea level, with coordinates 24°05' N latitude and 90°16' E longitude. The site featured shallow Red-Brown Trace soil, known as the "Salna" soil series, classified by the USDA as a Paleudult in the Orchept suborder of the Inceptisol order. The soil, acidic with heavy clay content in the top 50 cm, was combined with neighboring floodplain alluvium for experimental purposes. Situated in a subtropical zone with moderate temperatures, low humidity, and minimal wind and rainfall, the seeds were treated with Provax-200 WP fungicide for optimal germination and fungal protection. Fertilizer, following WRC recommendations (100-27-10-20-1 kg/ha N-P-K-S-B), was applied with two-thirds N and full others at basal, with the rest after 21 days. Irrigation occurred at 21, 55, and 75 days after sowing (DAS), and intercultural tasks were performed as needed.

### Data Collection

Plant phenology data, including days to emergence, heading, and flowering, were recorded for each genotype in a row. Days to flowering were noted when at least one flower opened in each genotype. Additionally, plant height and number of leaves per plant were recorded at various times, along with dry weights of stem, leaf, and reproductive parts, number of tillers per plant, and leaf chlorophyll content using a SPAD meter. Yield and yield components, such as spikelet per plant, seed per spikelet, spikelet

length, spikelet color, seed weight, and grain yield, were also measured. Plant height was measured from the above-ground portion to the shoot tip using a cm scale at 20-day intervals starting from 20 days after sowing (DAS). Descriptive analysis, including range, mean, coefficient of variation, and skewness, along with frequency distribution, was used to assess and describe genotype performance for each trait.

### Statistical Analysis

The genetic variation of genotypes was analyzed using SPSS 16 following the method outlined by Rojas et al. (2000). Pearson's coefficient was employed to estimate correlations among various plant characteristics (Clifford and Stephenson, 1975). A comprehensive analysis was conducted on 170 wheat genotypes, focusing on 17 quantitative variables, with eight variables selected for multivariate analysis including discriminant function analysis (DFA), Mahalanobis distance, Classification matrix, Representative genotypes.

## RESULTS AND DISCUSSION

### Discriminant Function Analysis

Stepwise discriminant function analysis (DFA) was done to determine the set of discriminatory functions contributed in separating 170 genotypes into six distinct clusters. DFA was actually performed to know whether a particular set of plant characters for previously discussed 8 characters is useful in separating six clusters. DFA is particularly useful in defining groups of the genotypes as prior classification criteria. Moreover, it provides a graphical output illustrating the existence of groups (Singh et al., 1991).

The four discriminant functions that differentiated among clusters were obtained by the stepwise procedure. Table 1 summarizes the contribution of each of 4 canonical discriminant functions for explaining the variance along with their Eigen values (Latent root) and canonical correlation coefficient. Function 1 alone explained more than fifty percent which is 55.3% of total variance and function 2 explained 33.3% of the total variance. Hence, the function 1 and function 2 accounted for

a cumulative 88.6% of total of total variance. All discriminatory functions except function 4 were statistically significant at a probability level of 0.000 according to chi-square test. Function 4 was statistically significant at a probability level of 0.013. Table 2 summarizes the variables mostly contributed to the discriminatory functions along with their coefficient under each function. Results show that days to flowering, biomass, 1000 seed weight (g) and harvest index mostly contributed in grouping 170 genotypes. These four characters mostly explained 88.6% of total variance under function 1 and function 2. The coefficient of 1000 seed weight (0.948) was higher in function 1 than that in function 2. It meant that 1000 seed weight mostly explained 55.3% of total variance showed in function 1. On the other hand, days to flowering (0.977) was higher in function 2 indicating the contribution of this variable to function 2 was higher in explaining 33.3% on total variance. Table 3 describes the correlation of coefficient between 8 discriminatory variables and 4 discriminatory functions. From the result it was observed that harvest index was placed at the top of the list of discriminatory variables with correlation coefficient of 0.943 under function 1. It indicates that harvest index played the most dominant role out of 8 variables in explaining the maximum variance in 170 genotypes by stepwise DFA. Genotypes situated at the right side of the diagram (Figure 1) produced highest harvest index and that of the left side produced lowest harvest index based on X ordinate. Therefore, function 1 separated group (cluster) 7 and 3 very clearly from group 5 and 2 based on harvest index. Cluster 7 and 3 were highest in producing kernel. On the other hand, the genotypes scattered on the upper part of the diagram required more days to emergence and that of the lower part required lesser days to emergence based on Y ordinate. Therefore, function 2 separated group 7 and group 6 very clearly from group 3, 4 and group 5 based on days required to flowering. Group 6 and group 7 require very less days to emergence and others require more days.

### Mahalanobis distance

According to the Mahalanobis distance ( $D^2$ ) among clusters (Table 4) also calculated by DFA, the seven clusters were statistically different from

each other at 0.001 level. Among 7 clusters, cluster 3 showed the highest distant of 10.337 units with cluster 2 and 6.790 units with cluster 1. Cluster 5 showed the high distant of 10.335 units with cluster 3 and 6.193 units with cluster 1. Cluster 7 showed the high distant of 9.632 units with cluster 5 and 8.254 units with cluster 2. The distant units among other clusters were highly significant but very near to each other.

### Classification matrix

The classification matrix of 7 groups of wheat genotypes presented in Table 5 summarized the predictive ability of discriminatory functions when classifying the different groups of genotypes. Each genotype was assigned to a cluster based on discriminatory functions. In Table 5, these are compared to the actual cluster membership of each genotype. DFA is particularly informative because misclassified genotypes were identified and reassigned to the appropriate group.

In general, the discriminatory functions reached a high degree of precision for group classification. In all cases, more than 95.7% of the genotypes were correctly assigned to clusters and overall 98.2% of the genotypes were correctly classified. The degree of total precision was highly significant according to the Q statistical test (Hair et al., 1992), indicating the high discriminatory ability of the classification matrix (Table 5). In group 2, 3, 5 and 7, 100% of the genotypes were correctly classified.

In group 1, out of 22 genotypes 23 were correctly classified (95.7%). one misclassified genotype corresponded to group 4. In group 4, out of 29 genotypes 28 were correctly classified (96.6%). One misclassified genotype corresponded to group 6. On the other hand, in group 6, out of 52 genotypes 51 were correctly classified (98.1%). One misclassified genotype corresponded to group 1.

### Representative genotypes

Figure 2 showed the orientation of genotypes under each of 7 clusters. The relative position of genotypes indicated the cumulative response of variables representing of function 1 and function

2. Group centroid of each cluster represented the optimum values of function 1 and function 2 that was resulted from the cumulative effects of all genotypes oriented under that cluster based on their response to the optimum response of that group. The deviation of the genotypes in response of discriminating variables was very close to the group centroid and might be considered as the most representative (might not be the best) of that

group. Accordingly, the genotype no 142 (2344) in group 1, the genotype no 94 (2080) in group 2, the genotype no 68 (1990) in group 3, the genotype no 46 (1688) in group 4, the genotype no 81 (2027) in group 5, the genotype no 165 (2481) in group 6 and the genotype no 53 (1751) in group 7 might be considered as more representative of their respective groups (Table 6).

**Table 1:** Discriminant functions that distinguish between clusters of 170 wheat genotypes

Function	Latent root	Variance %		r <sup>2</sup> coefficient	Walk's $\lambda$	X <sup>2</sup>	df	P
		Function	Cumulative					
1	4.950 <sup>a</sup>	55.3	55.3	.912	.020	637.235	24	.000
2	2.977 <sup>a</sup>	33.3	88.6	.865	.121	345.637	15	.000
3	.949 <sup>a</sup>	10.6	99.2	.698	.480	119.913	8	.000
4	.068 <sup>a</sup>	.8	100.0	.253	.936	10.826	3	.013

a. = First 4 canonical discriminant functions were used in the analysis.

**Table 2:** Standardized canonical discriminant function coefficients of the plant characters mostly contributed in grouping 170 wheat genotypes

Discriminating variables	Discriminant Function			
	1	2	3	4
Days to flowering	-.278	.977	-.039	.240
Biomass	-.131	-.538	.120	.894
1000 seed wt. (g)	.948	.012	.045	.396
Harvest index	-.038	.230	.974	-.188

**Table 3:** Structure matrix representing correlation between sixteen discriminating variables and standardized canonical discriminant functions of 170 wheat genotypes

Discriminating variables	Discriminant Function			
	1	2	3	4
Harvest Index	.943*	.207	.038	.258
Days to flowering	-.209	.839*	-.112	.489
Days to Emergence <sup>a</sup>	-.171	.523*	-.184	.153
Days to 50% flowering <sup>a</sup>	-.059	.461*	-.131	.400
Effective Tiller/plant <sup>a</sup>	.005	.107*	.049	-.096
1000 seed weight (g)	-.007	.061	.993*	-.097
Straw yield (g plant <sup>-1</sup> )	-.363	-.304	.222	.852*
Seed yield (g plant <sup>-1</sup> ) <sup>a</sup>	.084	-.265	.196	.785*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions. Variables ordered by absolute size of correlation within function.

\* Largest absolute correlation between each variable and any discriminant function.

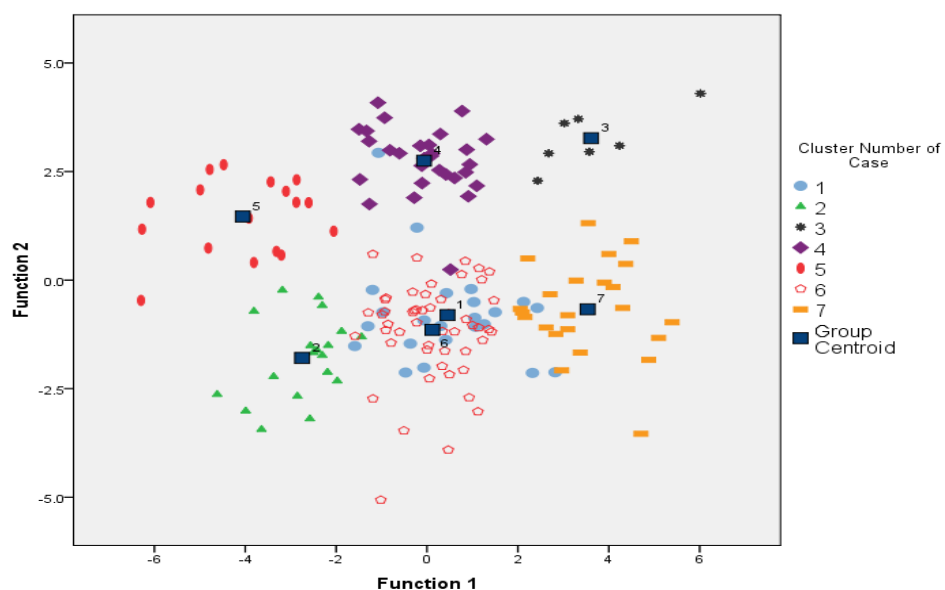
**Table 4:** Pairwise Mahalanobis distances ( $D^2$ ) between seven clusters of 170 wheat genotypes

Cluster	1	2	3	4	5	6	7
1	0.00						
2	4.855	0.00					
3	6.790	10.337	0.00				
4	4.276	6.059	4.889	0.00			
5	6.193	3.367	10.335	5.492	0.00		
6	3.061	3.943	7.214	4.083	5.690	0.00	
7	4.628	8.254	4.487	5.369	9.632	4.550	0.00

\*\* Distances differing from zero at a 99% confidence interval

**Table 5:** Classification matrix (Precision level) of seven groups of wheat genotypes, rows being observed category and columns being predicted category

Group	% Correct	1	2	3	4	5	6	7	Total no. observed
1	95.7	22	0	0	1	0	0	0	23
2	100	0	19	0	0	0	0	0	19
3	100	0	0	7	0	0	0	0	7
4	96.6	0	0	0	28	0	1	0	29
5	100	0	0	0	0	17	0	0	17
6	98.1	0	1	0	0	0	51	0	52
7	100	0	0	0	0	0	0	23	23
Total no. predicted	98.2	22	20	7	29	17	52	23	170

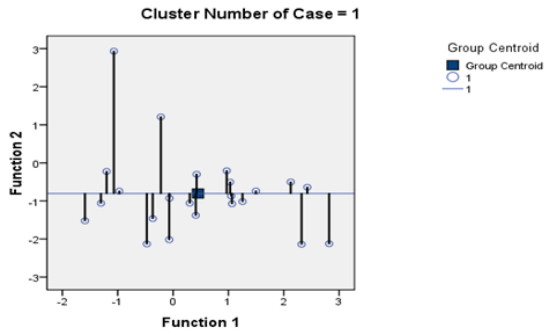


**Figure 1:** Graphical illustration of the discriminant function analysis of seven groups of 170 wheat genotypes. The accessions indicate the groups (clusters) obtained through cluster analysis.

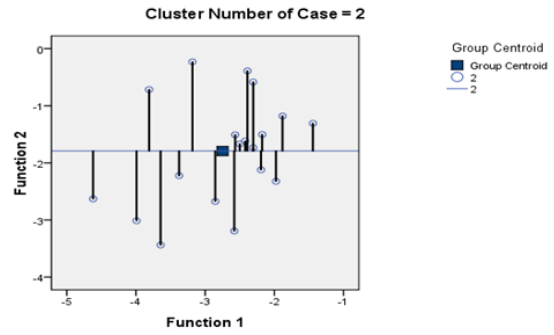
**Table 6:** Major plant characteristics of seven genotypes mostly representing their respective group

Group no.	Gen. no.	Acc. no.	Days to Emergence	Days to flowering	Days to 50% flowering	Effective Tiller/plant	1000 seed weight (g)	Biomass (g plant <sup>-1</sup> )	Harvest Index	Seed yield (g plant <sup>-1</sup> )
1	142	2344	4.92	73.85	75.53	5.10	41.35	16.17	54.42	8.80
2	94	2080	4.71	73.43	76.43	3.45	39.87	18.57	50.16	9.24
3	68	1990	5.33	75.63	77.57	2.67	42.17	14.94	59.37	8.87
4	46	1688	5.17	75.67	77.63	4.65	43.62	15.80	54.71	8.62
5	81	2027	5.11	76.14	79.78	3.13	41.75	18.26	50.09	9.01
6	165	2481	4.67	73.45	76.54	4.15	43.25	15.74	53.80	8.47
7	53	1751	4.92	74.42	76.63	4.32	43.03	15.45	58.89	9.10

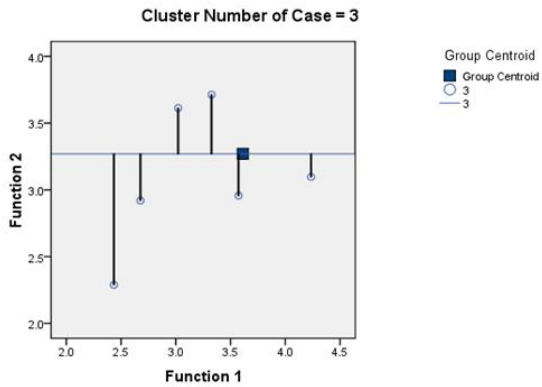
Canonical Discriminant Functions



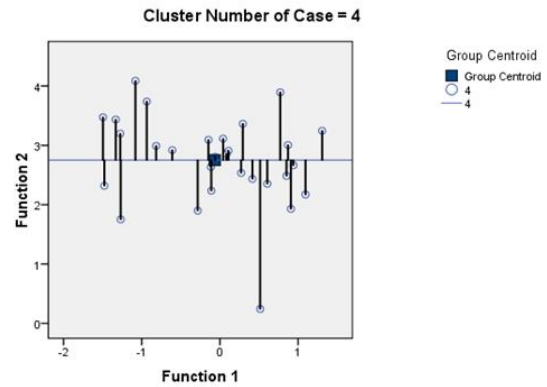
Canonical Discriminant Functions



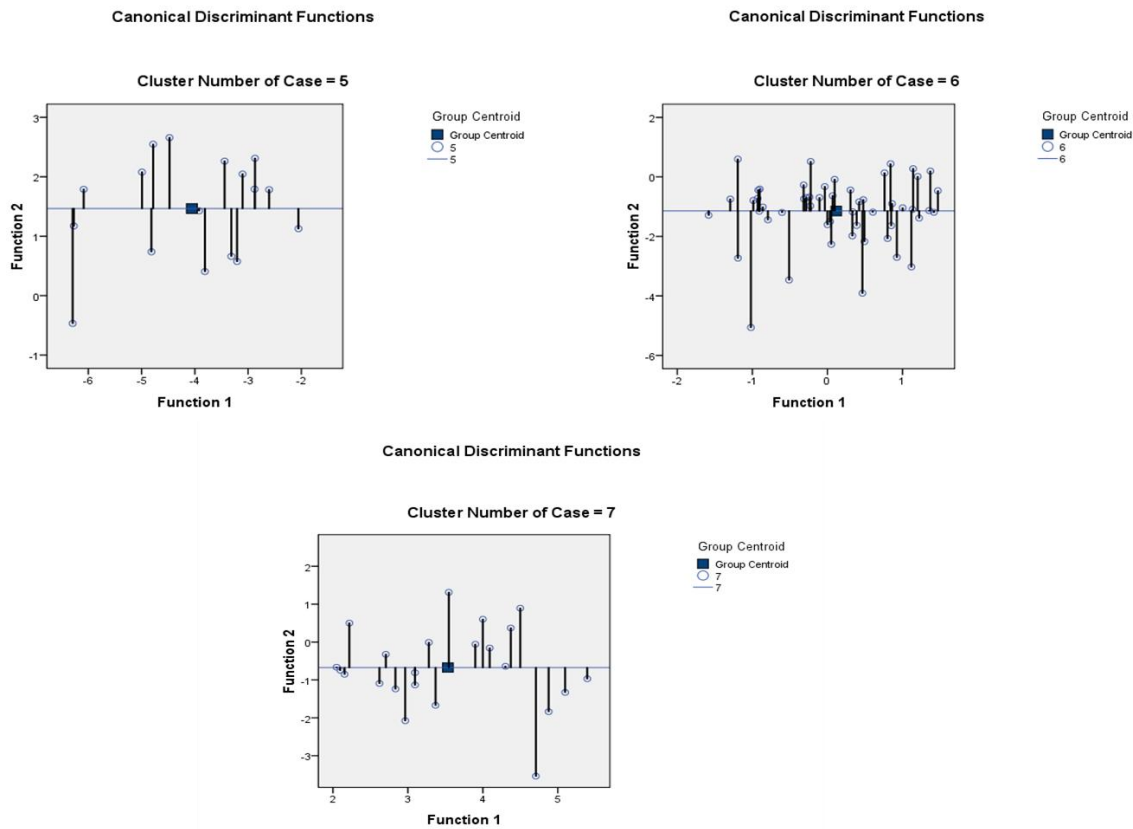
Canonical Discriminant Functions



Canonical Discriminant Functions







**Figure 2:** Graphical illustration of genotypes under each cluster by DFA based on 8 plant characters

## CONCLUSION

The study found significant variability among the genotypes in terms of characteristics such as effective tiller plant<sup>-1</sup>, days to flowering, biomass, 1000 seed weight, and seed yield. The genotypes were grouped into seven clusters based on cluster analysis, with distinct differences observed in traits and yield potential among the clusters. The discriminatory functions used for classification displayed a high degree of precision, with over 98% of genotypes correctly classified into their respective clusters. The study highlighted the contribution of specific variables like days to flowering, biomass, 1000 seed weight, and harvest index in grouping the genotypes effectively. These variables explained a significant portion of the total variance, indicating their importance in differentiating the genotypes based on their characteristics. Overall, the research underscores the importance of understanding the morpho-physiology, phenology, and yield performance of

wheat genotypes for effective classification and selection of promising varieties. Further studies with selected genotypes are recommended for potential commercial cultivation, and exploring molecular approaches could enhance the evaluation process for better agricultural outcomes.

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