



Morphological diversity valuation of exotic and advanced lines of bread wheat (*Triticum Aestivum L.*) via multivariate analysis

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Abstract

Morphological diversity assessment of 28 exotic and 8 advanced lines was carried out. The data of 12 different traits were subjected to different multivariate techniques. The correlation analysis revealed heavier positive correlation among 1000 grain weight with the yield, spikelet spike⁻¹, Normalized vegetation index (NDVI) and negative correlation with spike length, days to heading and maturity. NDVI and CTD (Canopy temperature depression) at booting stage revealed positive correlation with NDVI and CTD at anthesis stage. CTD and NDVI is a good indicator of a genotype's fitness against heat stress environment and this trait may be used as selection tools for developing heat and drought stress tolerant genotypes. Principal component analysis showed first 5 PCs having eigen value >1 explaining 78.1% of the total variability. The lines speared into 4 clusters. Cluster 1, 2, 3 comprised of exotic lines while all varieties in cluster 4 comprised of advanced lines. Therefore, the traits of the exotic lines should be used in advanced lines and vice versa during crossing schemes. Plot and tree diagrams demonstrated adequate diversity among the accession for various traits and some extent of association between different clusters. In all described entries disease reaction (leaf and yellow rust) ranged from 0-100S. The results concluded that morphological diversity in the deliberate material is planned by genotypes and this diversity could be utilized for further breeding of exotic lines with advanced lines to cope with climate change mitigation and food security.

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Introduction

Historically, wheat production in Pakistan can be allocated into three divergent eras, 1947-65 (prior to the release of semi-dwarf wheat), 1966-76 (green revolution period) and 1977 (post green revolution period). The green revolution led to high yielding wheat varieties with assembly of allelic combinations for few key traits. The successes of such varieties permit many wheat importing countries like Pakistan to become wheat-exporter. However, the disproportionate use of few successful varieties as parents for breeding new varieties led to the loss of locally adaptive genetic diversity. The decreases in genetic diversity in a population are standard as genetic bottlenecks (Marta *et al.*, 2015).

As result of global climate change, the restricted contact occurred in 20th century during the process of green revolution (1940s to 1960s) where breeding programs hinge on few lines for triggering gene pools. In wheat populations, this blockage is thought to lessen radical genetic diversity which justified the prominence on germplasm exchange and the use of exotic lines in existing advanced lines for improving the genetic miscellany.

Morphological characterization and evaluation of the diversity in wheat breeding has been extensively studied around the world. Exotic lines may have imperative role for food security not only as cradle of gene readily available for researchers but also because they perform well in marginal surroundings to the new advanced lines because advanced lines are frequently selected in promising environment (Tamoor *et al.*; 2014).

Realizing the importance of exotic lines for rehabilitating genetic diversity in existing advanced lines, a panel of 28 exotic and 8 advanced lines was characterized for sole yield and useful traits which would help in wheat improvement program. This kind of tryout will alive legacy of wheat genotypes and also permit researchers to judge the value of exotic and advanced lines in today's emergent disorders especially in heat and drought stress scenario.

Materials and methods

Research Area

The research was conducted in the research area of Wheat Research Institute, Ayub Agricultural Research Institute, Faisalabad (longitude 73° and 74° East, latitude 30° and 31.5° North, at an elevation of 605 feet above sea level) in mid-November 2016-17 and 2017-18 wheat cropping seasons.

Materials and Work Procedure

The material was planted in randomized complete block design with 2 replications. The plot size was 2.5m × 2 rows. NPK fertilizer was applied 120-90-60kg ha⁻¹. Full dose of phosphorus and potash was applied at sowing along with half of nitrogen, while the rest of the nitrogen was applied at tillering with first irrigation. Data on the morphological traits were recorded, i.e., 1000 grain weight (g), plant height (cm), days to heading (50%), days to maturity (50%), spikelets spike⁻¹, grains spike⁻¹, spike length (cm), normalized difference vegetation index (at booting and anthesis stages), canopy temperature (at booting and anthesis stages), (yield kg ha⁻¹) and disease reaction (0-100S). The two years observations were pooled for and an average of two readings was calculated for use in further analysis. For canopy temperature (°C), data was recorded with LT. 300 6th Sense Infrared Thermometer (IRT) and the canopy depression of each variety/line was calculated by formula before subjected to analysis.

Analysis

CTD=Initial value outside environmental temperature + Final value outside environmental temperature divided by two minus each recorded CT value. For recording the value of NDVI, green seeker (handheld-505) was used. Both the readings (CT and NDVI) were taken during sunny days with least wind speed at noon time during 11 a.m. to 1 p.m. when the dew dried off from the plant canopy. For disease infestation all the materials were surrounded by the spreader rows of highly susceptible variety (Morocco) while the rust severity and response was recorded according to the modified Cobb's scale described by Peterson *et al.*, (1973).

The average data of both the years were subjected to basic statistics, correlation analysis, cluster analysis and principal component analysis (PCA) using statistical software packages of SPSS version 12 and STATISTICA version 5.0. (Sneath and Sokal, 2014). Cluster analysis was performed using K-means clustering while tree diagram based on elucidation distances was developed by Ward's method. To find out the patterns of variability among genotypes and association between different clusters SPSS version 12 was used.

Results and discussion

The analysis of variance for all measured traits was carried out. The mean squares from analysis of variance given in table 1 indicated that genotypic differences were highly significant ($P \leq 0.01$) for all the measured traits indicating the presence of sufficient variability to identify potential genotypes (Khalil *et al.*, 2010, Duguid and Brule-Babel, 1994) also found highly significant genotypic differences among exotic and advanced lines in their findings.

Table 1. Analysis of variance for measured traits in exotic and advance lines

SOV	Df	Means of squares											
		G/W	PH	D/H	D/M	SP/SP	GR/SP	SpLn	NDVI (B)	CTD (B)	NDVI (A)	CT (A)	Yield (Kgha ⁻¹)
Treat	35	74.5**	165.3**	41.8**	34.9**	4.7**	49.5**	1.8**	0.0064**	4.1**	0.0046**	4.4**	1902501**
Error	35	6.4	6.4	0.4	0.7	0.4	0.3	0.2	0.0001	0.03	0.0001	0.1	44
F-value	-	11.6	25.9	104.5	48.6	11.8	147.1	9.8	60.2	120.7	80.6	89.0	1.1
CV%	-	7.8	2.4	0.6	0.6	3.8	1.2	3.5	1.3	1.6	1.0	1.9	108.8

**= $P \leq 0.01$, * = $P \leq 0.05$, G/W=1000grain weight (g), PH=plant height (cm), D/H=days to heading, D/M=days to maturity, SP/SP=spikelet spike⁻¹, GR/SP=grain spike⁻¹ SpLn=spike length (cm), NDVI (B)=normalized vegetation index at booting, CT (B)= canopy temperature at booting (C°), NDVI(A)=normalized vegetation index at anthesis and CT (A)=canopy temperature at anthesis (C°).

Traits Correlation Patterns

Simple correlation coefficients among 12 traits in both groups revealed a lot of significant and practical associations (table 2). The heavier positive correlation among 1000 grain weight with the yield and spikelet spike⁻¹ and negative correlation with spike length, days to heading, maturity and CTD were found which suggested that these traits are important for direct selection of high yielding genotypes. Gomez *et al.*, (2014) also suggested that these morphological attributes contribute to grain yield and yield components like grain weight by maintaining rate and

duration of days to heading and maturity. It was observed that the varieties have ranged between early to normal heading performed well in all environments that may be due to the fact that heading and maturity time is considered as a covariate to control yield and yield related components.

The negative correlation of 1000 grain weight with the spike length might be due to a low spike length genetic diversity among the genotypes. Likewise, plant height was deterrent traits for grain yield improvement because this trait exhibited.

Table 2. Correlation matrix for measured traits in exotic and advanced lines.

Variables	GW	PH	D/H	D/M	SP/SP	GR/SP	SpLn	NDVI (B)	CTD (B)	NDVI (A)	CTD (A)
PH	-0.04										
D/H	-0.22	0.14									
D/M	-0.37*	0.31	0.36*								
SP/SP	0.34*	0.22	-0.13	0.07							
GR/SP	0.21	-0.26	-0.31	-0.18	0.38						
SpLn	-0.11	-0.10	0.30	0.21	0.19	-0.04					
NDVI (B)	0.23	-0.08	-0.38*	-0.10	0.02	0.29	-0.37*				
CTD (B)	-0.48*	0.25	0.09	0.33	0.07	-0.17	0.23	-0.47**			
NDVI (A)	0.28	-0.04	-0.41*	-0.06	0.21	0.42**	-0.39*	0.78**	-0.50**		
CTD (A)	-0.19	-0.03	0.05	0.30	0.07	0.29	0.05	-0.24	0.48**	-0.21	
Yield	0.51**	-0.05	-0.46**	-0.16	0.19	0.34*	-0.45*	0.58**	-0.56**	0.55**	-0.15

Negative association with yield. El-Mohsen *et al.*, (2012) reported the same negative correlation between these two traits. There was positive correlation was observed between spikelet spike⁻¹ with the spike length and grain spike⁻¹. Reynolds *et al.*, (2001) found the same positive interrelation of these traits in Egyptian bread wheat varieties by additionally mentioning that these yield attributing traits are very important contributors to total yield and biomass. Similarly, NDVI and CTD at booting stage revealed positive correlation with NDVI and CTD at anthesis stage. CTD is a good indicator of a genotype's fitness against heat stress environment and further suggested that this trait may be used as selection tools for developing heat and drought stress tolerant genotypes. Freeman *et al.*, (2007) advocated in his experiments that CTD values obtained, almost in all stages the highest CTD values observed in drought tolerant genotypes, while, minimum values were related to drought sensitive genotypes indicating drought tolerant genotypes had cooler canopy and drought sensitive genotypes had hotter canopy in rainfed condition. Interesting results were found between yield and NDVI, CTD reading at booting and anthesis stages. In this scenario, NDVI at both stages highly significant positive association. The NDVI at booting, heading and flowering has been correlated to final wheat yield (Verhulst and Govaerts, 2010). The NDVI data provides information on chlorophyll content. A positive association of NDVI data with yield suggested that lines with high chlorophyll content at anthesis and grain filling stage had higher yield and could be used as an effective criterion for

screening genotypes in drought and heat stress conditions (Balota *et al.*, 1993; Scotford and Miller, 2005). Higher NDVI values during anthesis and grain filling could suggest the presence of stay-green which resulted in higher yield. Similarly, CTD showed the same pattern with the yield trait but with negative relationship. As the booting and then anthesis period progressed, NDVI values in varieties decreased while CTD increased (Mondal *et al.*, 2015) that is why CTD showed negative association with yield because the high temperature leads to terminal heat stress. These results are absolutely confirmatory with the finding of Mason and Singh (2014) who also declared positive correlations between grain yield and NDVI and negative correlation with CTD at different stages. Canopy temperatures had a negative incline, advocating that cooler canopies had association with higher yields because cooler canopies improve physiological and metabolic functions related to adaptation under stress. Such comebacks to canopy cooling have been testified by Pecetti and Damania (1994).

Principal Component Analysis

Principal component analysis tears the total variance into various factors which supports for conservation and manipulation of genetic resources and scheduling for deployment of applicable germplasm in crop improvement for particular plant traits (Zaman *et al.*; 2014). Out of total 12 traits studied, 5 traits were extracted having eigen value >1 and contributed about 78.1% variability. However, the remaining contributed only 21.9%, towards the total multiplicity (table 3).

Table 3. Principle component analysis of various traits in exotic and advanced lines.

Traits	PC1	PC2	PC3	PC4	Eigen values	Proportion (%)	Cumulative (%)
G/W	-0.299	0.023	0.166	0.489	3.9190	0.327	0.327
PH	0.099	0.150	-0.616	0.283	1.6780	0.140	0.466
D/H	0.294	-0.152	-0.124	0.221	1.4269	0.119	0.585
D/M	0.217	0.283	-0.472	-0.056	1.3180	0.110	0.695
SP/SP	-0.090	0.516	0.003	0.516	1.0278	0.086	0.781
GR/SP	-0.237	0.456	0.329	-0.141	0.7835	0.065	0.846
SpLn	0.249	0.116	0.304	0.374	0.5278	0.044	0.890
NDVI (B)	-0.387	0.013	-0.251	-0.193	0.3738	0.031	0.921
CTD (B)	0.360	0.315	-0.036	-0.175	0.3648	0.030	0.952
NDVI (A)	-0.404	0.141	-0.235	-0.100	0.2574	0.021	0.973
CTD (A)	0.165	0.509	0.144	-0.353	0.2104	0.018	0.991
Yield	-0.414	0.083	-0.118	0.022	0.1125	0.009	1.000

The PC I had considerable positive factor loadings for plant height, days to heading, days to maturity along with CTD value at both stages. Likewise the 2nd PC was dominated with positive factor for spikelet spike⁻¹, grains spike⁻¹, except and spike length in group₁ and CTD values at both stages in group 2. These results are in harmony with the results of correlation analysis which showed mostly positive association among traits. The results are confirmatory with [Gaju, 2007] findings that grains spike⁻¹ and spikelet spike⁻¹, plant height, days to heading, days to maturity and 1000 grain weight are contributing characters for the diversity with their considerable positive factor loadings. The PC III was explained by variation among genotypes due to grain spike⁻¹ and spike length. Similarly, PC IV was explicated by variation in spikelet spike⁻¹ and 1000 grain weight with their

considerable positive factor loadings. NDVI at booting appear as negative loading in this PC. Usually it is customary to choose one trait from these identified groups. Hence for spikelet spike⁻¹, grain spike⁻¹, and 1000 grain weight and NDVI at booting is the best choice. However spikelet spike⁻¹ and grain spike⁻¹ revealed a common parameter among the material in hand which could be utilized in scheming a breeding program. Presentation by biplots with their relative score plot provides spontaneous and quantitative classification of multidimensional data (Fig. 1). The Biplot represents the relationship between different plant attributes and genotypes. Biplot vectors close to each other depicted the correlation among those traits and the genotypes lie close to a particular trait vector represents the high performance of the genotypes for that trait

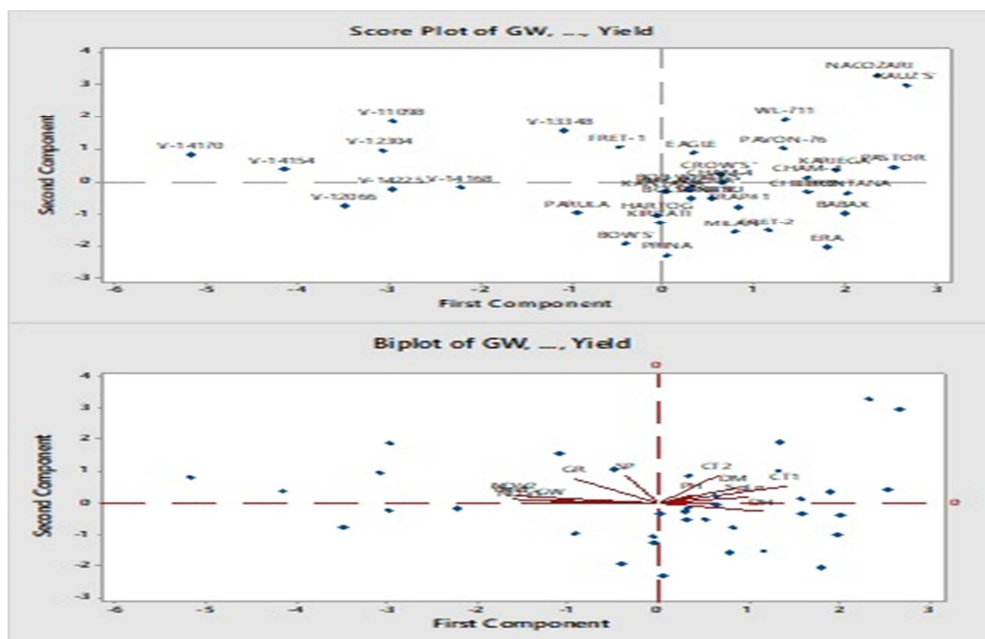


Fig. 1. Biplot and score plot presentation of exotic and advanced lines.

Cluster Analysis

Based on various traits, the measured traits separated into 4 clusters comprising 8, 10, 10 and 8 genotypes (table 4). The genotypes in cluster 1 demonstrated high plant height and CTD value at booting stage with lowest values of 1000 grain weight, spikelet spike⁻¹, grain spike⁻¹, spike length and NDVI readings at both stages. (table 5). Similarly, the 2nd cluster comprised of genotypes which took more number of days to

maturity with greater spike length but exhibited lower NDVI value at both stages. The members of 3rd cluster took more number of days to heading with high value of CTD at anthesis stage. Similarly cluster 4 was characterized by highest yield, 1000 grain weight and exhibited more NDVI readings at both stages. According to [Daniel *et al.*, 2011] the 49 genotypes were classified into 22 clusters with significant differences among all groups in all clusters.

Table 4. Germplasm accessions related to various cluster in exotic and advanced lines.

Cluster No.	No of accessions	Accessions
Cluster1	8	Babax , Parula, Fret-1, Cham-1, Pastor, Fret-2, Frontana, Trap#1
Cluster2	10	Bacanora, Bob White's', Bow's', Cham-4, Kariega, Milan , Eagle, WL-711,Tukuru, Kiritati,
Cluster3	10	Bolsena's', Nacozeni, Kautz's', Prina, Chilero, Crow's', Pavon-76, Hartog, Era, , Kantana
Cluster4	8	V-11098, V-12304, V-12066, V-14154, V-14225, V-14170, V-14168,V-13348

Table 5. Cluster centroids of studied traits in exotic and advanced lines.

Variables	Cluster1	Cluster2	Cluster3	Cluster4	Grand centroid
G/W	29.47	32.84	29.57	37.99	32.33
PH	111.25	105.25	104.75	105.00	106.39
DH	106.81	106.10	109.40	103.69	106.64
DM	147.38	148.20	147.30	145.81	147.24
S/P	16.13	16.60	16.50	17.38	16.64
G/S	44.94	46.00	49.40	53.06	48.28
SpLn	12.38	13.05	12.70	11.63	12.49
NDVI (B)	0.76	0.76	0.77	0.86	0.79
CTD(B)	12.83	11.95	12.28	10.25	11.86
NDVI (A)	0.73	0.73	0.74	0.81	0.75
CTD (A)	11.68	11.53	12.35	11.83	11.86
Yield	5507.50	5964.50	4790.00	7402.50	5856.25
Distances between cluster centroids					
Cluster1	0.00				
Cluster2	457.06	0.00			
Cluster3	717.55	1174.51	0.00		
Cluster4	1895.05	1438.03	2612.52	0.00	

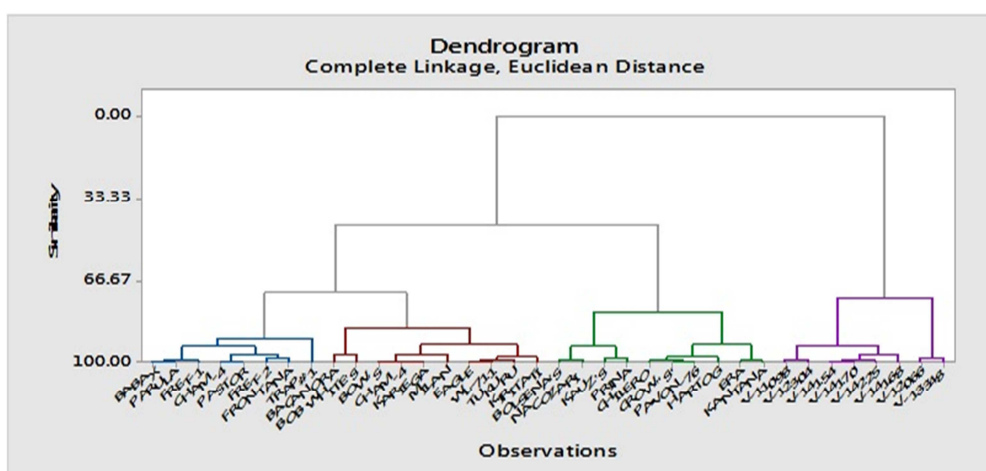


Fig. 2. Dendrogram showing the linkage of exotic (by name) and advanced lines (by V-number).

All the varieties in cluster 1, 2, 3 comprised of exotic lines while all varieties in cluster 4 comprised of advanced lines. Therefore, the traits of the exotic lines should be used in advanced lines and vice versa during crossing schemes. The lowest distance was recorded in cluster 3 which shows the occurrence of less genetic diversity within this cluster. The peak average inter cluster distance was recorded among cluster 3 and cluster 4 which had shown these clusters were genetically more divergent from each other than

any other clusters. Minimum inter cluster distance was observed between cluster 2 and cluster 3 indicating that the bread wheat genotypes in these clusters were not genetically assorted or there was little genetic diversity between these clusters. The wider distances between the clusters could be utilized in breeding program to develop transgressive segregants with a broad array of adaptability in traits deliberated. The dendrogram showed results comprising of main groups each of which is further

subdivided into sub groups (Fig. 2). The most different genotypes were Babax (exotic line) and V-13348 (advanced line).

Descriptive frequency of qualitative leaf and yellow rust disease data was noted (Fig. 3). In all described entries disease reaction (leaf and yellow rust) ranged from 0-100S. However, in leaf rust more frequency of TMS followed by TMR, 10MRMS and TS while in yellow rust 0 followed by TR, TS and TMS frequencies were observed collectively.

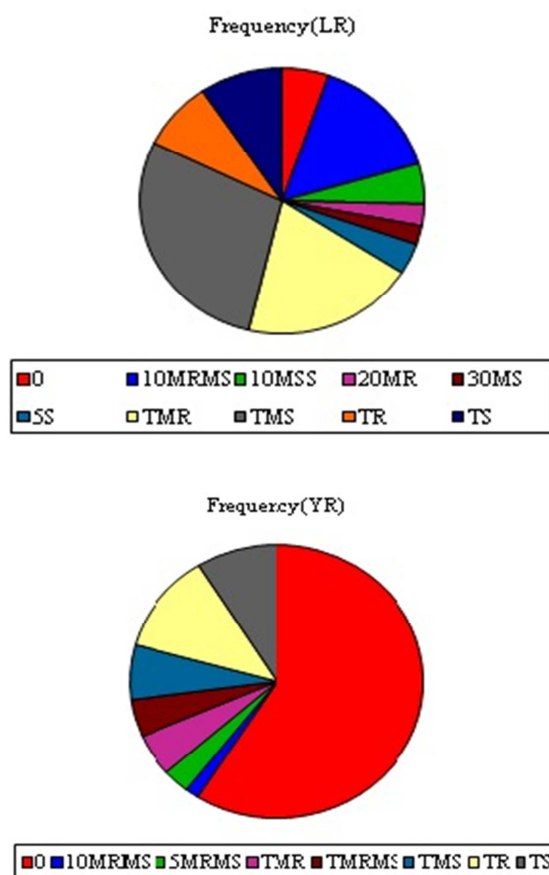


Fig. 3. Descriptive frequency of qualitative leaf and yellow rust disease data.

Conclusion

From the results of present exploration, it is concluded that grain yield can be enhanced by improving yield related components while plant height was deterrent traits for grain yield improvement. NDVI and CTD at booting stage revealed positive correlation with NDVI and CTD at anthesis stage. CTD found to be a good indicator of a genotype’s fitness against heat stress environment. A

positive association of NDVI data with yield suggested that lines with high chlorophyll content at booting and anthesis stage had higher yield. Therefore, both the traits could be used as an effective criterion for screening and developing heat and drought tolerant genotypes. Overall, this experiment accredits exotic and advanced lines meriting their usage towards improvement of stagnant wheat yield and disease resistance and to cope with climate change mitigation and food security.

References

Balota M, Amani I, Reynolds MP, Acevedo E. 1993. Evaluation of membrane thermostability and canopy depression as screening traits for heat tolerance in wheat. CIMMYT; Mexico. Wheat special report No. **20**, 1-26.

Daniel H, Mebrahtom M, Tsigie G. 2011. Genetic Divergence Analysis on some Bread Wheat Genotypes Grown in Ethiopia. Journal of Central European Agriculture **12**, 344-352

Duguid SD, Brule-Babel AL. 1994. Rate and duration of grain filling in five springwheat (*Triticumaestivum L.*) genotypes. Canadian Journal of Plant Science **74**, 681-686.

El-Mohsen AAA, Hegazy SRA, Taha MH. 2012. Genotypic and phenotypic interrelationships among yield and yield components in Egyptian bread wheat genotypes. Journal Plant Breeding and Crop Science **4**, 9-16.

Freeman KW, Raun WR, Johnson GV, Mullen RW, Stone ML, Solie JB. 2007. Late-season prediction of wheat grain yield and grain protein. Communication in Soil Science and Plant Analysis **34**, 1837-1852.

Gaju O. 2007. Identifying physiological processes limiting genetic improvement of ear fertility in wheat. PhD thesis, University of Nottingham 1-265.

Gomez D, Vanzetti L, Helguera M, Lombardo L, Frascina J, Miralles DJ. 2014. Effect of Vrn-1, Ppd-1 genes and earliness on heading time in Argentinean bread wheat cultivars. Field Crops Research **158**, 73-81.

- Khalil AL, Mahboob AS, Afzal A, Umar DM, Sher M, Pirzada AJ.** 2010. Comparative performance of wheat advance lines for yield and its associated traits. *World Applied Science Journal* **8**, 34-37.
- Marta SL, Ibrahim E , Peter SB, Sukhwinder S, Conxita R, Kursad O, Husnu A, Emel O, Fatih O, Alagu M, Tomohiro B, Prashant V.** 2015. Exploiting genetic diversity from landraces in wheat breeding for adaptation to climate change. *Journal of Experimental Botany* **66**, 3477-3486.
- Mason RE, Singh RP.** 2014. Considerations when deploying canopy temperature to select high yielding wheat breeding lines under drought and heat stress. *Agronomy* **4**, 191-201.
- Mondal S, Ravi S, Huerta-Espino J, Kehel Z, Autrique E.** 2015. Characterization of heat and drought stress tolerance in high yielding spring wheat. *Crop Science* **55**, 1-11.
- Pecetti L, Damania AB.** 1994. Geographic variation in tetraploid wheat (*Triticum turgidum* ssp. *Turgidum* convar. *durum*) landraces from two provinces in Ethiopia. *Genetics Research and Crop Evolution* **43**, 395-407.
- Peterson RF, Champbell AB, Hannah AE.** 1948. A diagrammatic scale for estimating rust intensity of leaves and stem of cereals **26**, 496-500.
- Reynolds MP, Ortiz-Monasterio JI, McNab A.** 2001. Application of physiology in wheat breeding. CIMMYT, Mexico 59-76.
- Scotford IM, Miller PCH.** 2005. Applications of spectral reflectance techniques in Northern European cereal production. A review of Biosystems engineering **90**, 235-250.
- Sneath PHA, Sokal RR.** 1975. Numerical Taxonomy- The Principles and Practice of Numerical Classification. Taylor and Francis Publishing Ltd. **24**, 263-268.
- Tamoor HM, Tariq Z, Akram JI, Attiq R, Ghulam R.** 2014. Estimation of some genetic parameters and inter-relationship of grain yield and yield related attributes in certain exotic lines of wheat (*Triticum aestivum* L.). *Journal of Biology Agriculture and Health Care*. **4**, 48-53.
- Verhulst N, and Govaerts B.** 2010. The normalized difference vegetation index (NDVI) green seeker TM handheld sensor: Toward the integrated evaluation of crop management. Part A: Concepts and case studies. CIMMYT, Mexico 1-12.
- Zaman M, Siddique NA, Rahman M, Abida MY, Islam MJ.** 2014. Divergence analysis of drought tolerant. *Bangladesh Journal of Agriculture Research* **39**, 385-396.