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GROUPING BREAD WHEAT GENOTYPES AND LINES BASED ON SOME MORPHOLOGICAL TRAITS USING MULTIVARIATE ANALYSIS

M. PASANDI[1], M. JANMOHAMMADI[1*], Z. MOVAHEDI[2], N. SABAGHNIA[1]

*E-mail: jmohamad@alumni.ut.ac.ir

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ABSTRACT. The knowledge about the extent of variability among bread wheat (*Triticum aestivum* L.) genotypes is a high value for the genetic improvement programs and the efficient genetic diversity utilization of plant materials. The objective of this research was to assess the morphological characteristics of 56 common wheat genotypes which were planted under field condition and their morphological traits were recorded. The principal components (PC) analysis, factor analysis and clustering procedure were applied to group genotypes according to similarity on the basis of the measured traits. Results showed that the first two PCs explain 41% of the total variation. The PC and factor analyses grouped genotypes into four groups while the cluster analysis grouped them into five distinct clusters. The cluster I had good resistance to yellow rust and salinity while the cluster II, had good resistance to drought, cold and salinity. The cluster III had high tolerance to cold stress and low temperatures but the cluster IV had good tolerance to drought, brown rust and yellow rust. Also, cluster V had various properties including tolerance abiotic stresses, resistance to biotic stresses, resistant to lodging and semi-dwarf property. The results of this research will support efforts of conservation and utilization of genotypes in bread wheat breeding programs.

Key words: Genetic diversity; Morphological characters; Multivariate analyses; *Triticum aestivum* L.

INTRODUCTION

Bread wheat (*Triticum aestivum* L.) is an important cool season cereal crop, and plays an important role in the food and nutritional security in many areas of world. Being a staple food, it has occupied a lot of area in Iran and the need and importance of wheat is increasing day by day due to increase in population (Mollasadeghi *et al.* 2012). An important objective of Iran is to reduce the dependence on imported wheat by enhancing average grain yield and production and

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[1] Department of Agronomy and Plant Breeding, Faculty of Agriculture, University of Maragheh, Maragheh, Iran
[2] Department of Agronomy and Plant Breeding, Faculty of Agriculture, Malayer University, Malayer, Iran

increasing productivity is a national target to fill the gap between wheat consumption and production. This target can be done by increasing cultivation area as well as increasing yield per unit area but, it is nearly impossible to increase area under wheat due to competition with other crops (Moghaddam *et al.* 1998; Aharizad *et al.* 2012). Therefore, increase yield is possible by better crop management techniques and introducing high yielding cultivars along with resistance against biotic and abiotic stresses.

The plant breeding efforts have narrowed the genetic base of bread wheat, which could jeopardize future genetic improvement programs. Also, an intensive selection has further reduced genetic diversity, narrowing the germplasm base available for future breeding advances (Tanksley and McCouch, 1997). Cultivation of such plant materials entails a risk due to genetic vulnerability and changes in environmental conditions may impose stress on plants and they could not cope with these situation and their performance may significantly reduce (Reif *et al.,* 2005). Nevertheless, plant breeding does not inevitably lead to a loss of genetic diversity and it could be reduced by intensive selection can be counterbalanced by introgression of novel germplasm. Genetic divergence analysis estimates the extent of diversity existed among selected plant materials and knowing precise information on the nature and magnitude of genetic diversity helps the plant breeder in choosing the diverse parents for purposeful hybridization (Hailegiorgis *et al.,* 2011).

There are different genetic resources for bread wheat improvement and among those; landraces have been the preferred ones due to their genetic homozygosity. The characterization of such plant materials is crucial for exploiting the genetic variability and tolerance to stresses (Aliu and Fetahu, 2010). However, genetic diversity in cultivated cultivars is also essential for successful breeding and creation of new improved genotypes. The evaluation of phenotypic diversity to identify groups with similar genotypes is important for utilizing genetic resources for investigation the diversity of breeding materials (Franco *et al.,* 2001). Knowing this information is essential for determining diverse parental combinations and creating segregating generations with high genetic variation for selection process in breeding programs. Criteria for the estimation of genetic diversity can be morphological traits and the success of a breeding program depends upon the amount of genetic variability of these traits present in the plant materials.

There are different statistical techniques covering grouping genotypes to evaluate genetic variability for breeding programs (Manly, 2004). Leilah and Al-Khateeb (2005) used several statistical methods to investigate the association among wheat traits. The principal components (PC) analysis explores complex data sets and transforms a number of associated variables into a smaller number of PCs and used in wheat (Khodadadi *et al.,* 2011). Factor analysis has been used to identify traits related to wheat and grouping genotypes (Moghaddam *et al.,* 1998). Cluster analysis can be used to identify genotypes which can be classified into main clusters based on similarity or dissimilarity indices and is useful for parental selection in wheat breeding program (Aharizad *et al.,* 2012).

This study was undertaken as a practical trial in order to determine the dependence relationship among genetic diversity of fifty-six Iranian cultivars of bread wheat by using certain multivariate statistical procedures. The aim of this investigation is to help wheat breeders, how to select suitable parental could be efficiently used in breeding programs.

MATERIALS AND METHODS

Fifty-six genotypes of bread wheat listed in *Table 1* were planted using based on a randomized complete block design with three replications. Tillage of all plots was performed prior to sowing date and fertility is constrained by low organic matter. For chemical fertilization, 60 kg ha^{-1} of nitrogen, 30 kg ha^{-1} of phosphorus and 20 kg ha^{-1} of potassium were applied and the weeds were controlled chemically. Sowing was done by hand in plots with six rows 2.5 m in length and 25 cm apart. The planted plot size was 3.75 m^2 and the harvested plot size was 3.75 m^2 and the harvested was 2.5 m^2. Proper cultural practices were applied to the trial through out the growing season according to local practices. Ten plants were tagged randomly to measure stem diameter, plant height, leaf number at flowering, leaf length of flag length, leaf width of flag length, tiller number, internode length, peduncle length, spike length, floret number per spikelet, spikelet number, grain number per spike, length of awn, grain diameter and grain length. The number of days to anthesis or flowering, thousand seed weight and grain yield of each plot were recorded as well is possible.

Table 1 - The name and code of 56 bread wheat genotypes

Code	Name	GH	Maturity
G1	Niknezhad	Spring	Medium
G2	Alvand-1	Facultative	Early to Medium
G3	Shahpasand	Winter	Late
G4	Pishtaz	Spring	Medium
G5	Marvdasht	Spring	Medium
G6	Golestan	Spring	Late
G7	MS-81-14	Winter	Early
G8	C-85-11	Facultative	Medium
G9	Sepahan	Winter	Medium
G10	C-84-55-B	Facultative	Medium

Code	Name	GH	Maturity
G11	Chamran	Spring	Early
G12	Norstar	Winter	Late
G13	Karaj-3	Winter	Medium
G14	Sabalan	Winter	Early
G15	Arta	Spring	Medium
G16	Alborz	Spring	Early
G17	Bayat	Spring	Early
G18	C-85-8	Winter	Medium-early
G19	Roshan-1	Spring	Medium
G20	C-85-9	Facultative	Medium
G21	Arvand	Spring	Early
G22	Hirmand	Spring	Early
G23	Zagros	Spring	Early
G24	Shiroodi-1	Spring	Early
G25	Zarin	Facultative	Late
G26	Azar-2	Winter	Early
G27	C-85-5513	Winter	Medium
G28	Alvand-2	Facultative	Early to Medium
G29	Veerynac	Spring	Early
G30	C-85-5512	Winter	Medium-late
G31	Roshan-2	Spring	Medium
G32	Ghods	Facultative	Medium
G33	Shahi cross	Winter	Medium-late
G34	Toos	Facultative	Medium-early
G35	Moghan-1	Spring	Early
G36	CDC-Ospray	Winter	Late
G37	Falat-1	Spring	Early
G38	Tajan	Spring	Early
G39	Chanab	Spring	Medium
G40	Sorkhtokhm	Spring	Medium
G41	C-84-5502	Winter	Medium
G42	Omid	Winter	Medium- late
G43	Akbari	Spring	Medium
G44	Tabasi	Spring	Early
G45	Shiraz	Spring	Medium
G46	Line A	Spring	Early
G47	Mahdavi-1	Spring	Medium
G48	Mahdavi-2	Spring	Early
G49	Roshan BC	Spring	Medium- early
G50	Shahriar	Winter	Medium-late
G51	Bahar	Spring	Medium-early
G52	Kavir	Spring	Medium
G53	Shiroodi-2	Spring	Medium
G54	Falaat-2	Spring	Early
G55	Sistan	Spring	Medium-early
G56	Saison	Spring	Late

GH= growth habit

The obtained dataset was first subjected for normality test by the Shapiro-Wilk normality test. The PC analysis was used and plot of first two principal components was drawn. The factor analysis method was performed to identification of the most important factors and after extraction; the matrix of factor loading was submitted to a varimax orthogonal rotation. Pair-wise distances between the genotypes based on Pearson's correlation similarity index on the raw data of measured agronomic traits were calculated. The average linkage cluster analysis was used to group the tested genotypes in the experiment. The experimental data were statistically analyzed for the above mentioned statistical methods using the Statistica version 7.0 (StaSoft, 2004), SPSS version 13.0 (SPSS, 2004) and Minitab version 14.0 (2005).

RESULTS

In the principal component (PC) analysis, the first two PCs (having eigenvalues > 1) explained about 41% of the observed variation, were, respectively, linked to variables related to grain number contributing traits (grain number, floret number and tiller number) and yield potential (grain diameter, grain length and thousand seed weight) characteristics (results are not shown). Based on the projection of genotypes in the first principal plan (*Fig. 1*) one can distinguish four groups as: the first group in the upper-left part comprises 12 genotypes, which most of them are the medium maturity landraces or commercial cultivars and mainly characterized by high tolerance to low temperatures such as Norstar (G12) and Sabalan (G14) cultivars. On the opposite side, a second group is composed by the genotypes such as Chamran (G11) and Karaj-3 (G13), which have spring growth habit and are characterized by tolerance to environmental stresses specially drought and salinity (*Fig. 1*). Note that these two groups of bread wheat genotypes are clearly distinguished by the first axes of PC. The other group in the lower-left part comprises MS-81-14 and C-85-8 lines, which are the landraces introduced for early and medium maturity and mainly characterized by high tolerance to cold, drought and salt stresses (*Fig. 1*).

It could be concluded that 15 of our bread wheat genotypes, which included in this section, have good potential for the mentioned stresses. On the opposite side (lower-right part), the forth group is composed by the eleven genotypes, which have medium or late maturity and are characterized by their resistance to brown rust and yellow rust diseases (*Fig. 1*).

The plot of varimax rotated scores of first two factors (*Fig. 2*) showed that 19 genotypes including cold tolerant genotypes were grouped to each other in upper-right side of plot. These genotypes are good candidates for genetic improvement of cold tolerant genotypes in future breeding attempts. Of 13 genotypes, were located in the upper-left side of *Fig. 2*, were heat, drought and salt tolerant genotypes. Also, the genotypes of lower-right side of plot are clearly distinguished by their properties such as early of medium maturity as well as resistance to drought, salinity and low temperatures (*Fig. 2*). Finally, the forth group of genotypes of *Fig. 2* were those that were resistant to diseases such as yellow and brown rusts. The morphological-based factor analysis plot of *Fig. 2* has a good fit to the PC analysis plot of *Fig. 2* and gave results in good agreement with the genotype grouping obtained from the PC analysis. The factor analysis is applied as a data reduction or structure detection method and after the PCs has been determined, and in order to facilitate the interpretation, the analysis involves a rotation of the PCs. Varimax rotation is the most popular rotation method, which is simplifies data interpretation because, each original variable tends to be associated with one of PCs, and each PC indicated only a small number of variables. Therefore, it seems that grouping of bread wheat genotypes based on factor analysis is more interpretable than PC analysis.

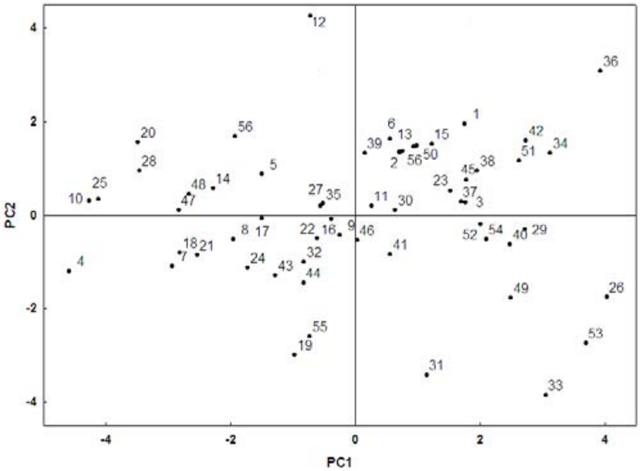

Figure 1 - Principal component (PC) analysis two-dimensional plot of 56 bread wheat genotypes based on morphological trait values

In the cluster analysis, all 56 bread wheat genotypes were grouped into five different categories with dendrogram slice in 0.00015 of Pearson's correlation similarity (*Fig. 3*). In cluster I, genotypes G1, G6, G16, G23, G29, G36 and G45, had good resistance to some diseases yellow rust and some abiotic stresses such as salinity. This cluster was superior for the mentioned characteristics. In cluster II, genotypes G3, G13, G14, G15, G22, G33, G38, G50 and G51, had good resistance abiotic stresses such as drought, cold and salinity (*Fig. 3*). The genotypes of cluster III (G12, G26 and G42) had high tolerance to cold stress and low temperatures but the genotypes of cluster IV (G2, G4, G5, G7, G8, G9, G10, G41, G49, G53, G54 and G56) had good tolerance some abiotic stresses such as drought and had good resistance to some diseases brown and yellow rust. Finally, the other remained genotypes which are grouped as cluster V had various properties including tolerance abiotic stresses, resistance to biotic stresses, resistant to lodging and semi-dwarf property. The morphological-based dendrogram (*Fig. 3*) has a good fit to the factor analysis and gave results in good agreement with the genotypes grouping obtained from the PC analysis. The generated dendrogram sequestrated wheat genotypes into clusters which exhibit high homogeneity within a cluster and high heterogeneity among clusters.

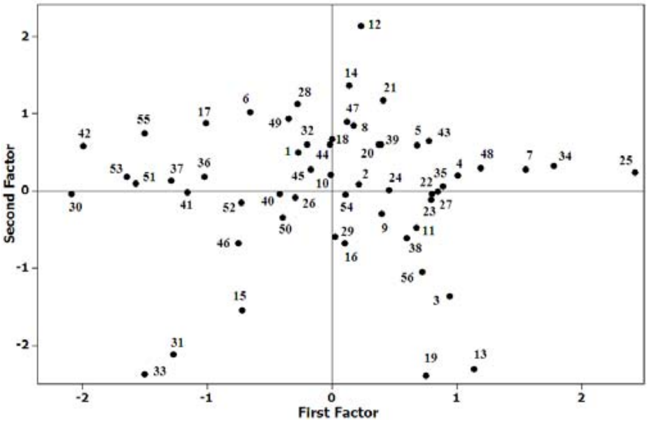

Figure 2 - Factor analysis two-dimensional plot of 56 bread wheat genotypes based on morphological trait values

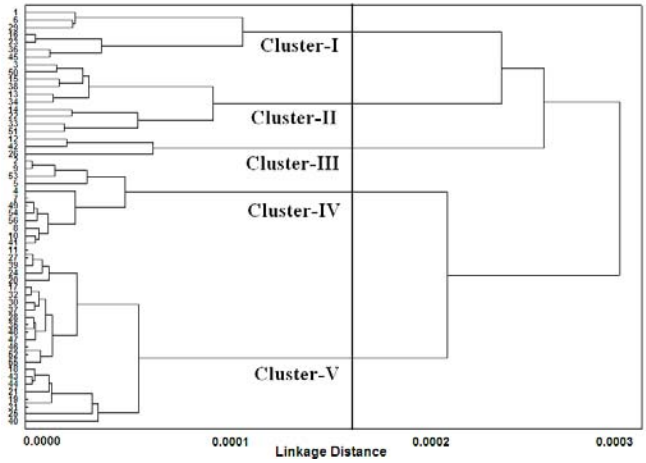

Figure 3 - Dendrogram of cluster analysis based on similarity matrix of 56 bread wheat genotypes

DISCUSSION

The results of the morphological analyses that bread wheat genotypes have high variability and were separated to several distinct groups. This high variability might be affected by various factors; selections made by breeders according to aims and environmental conditions (Dreisigacker *et al.* 2005). This might be the reason for genetic differences among the origin of plant materials or due to natural selection such as diseases and pests. The results of present study indicated that some genotypes from the same origins close to each other are genetically similar to each other and were grouped in the same sector in PC analysis or same cluster in cluster analysis. Similar results on genetic diversity in the morphologic characterization of bread wheat were reported by Van Beuningen and Bush (1997) and Mollasadeghi *et al.* (2012) and in the molecular characterization were reported by Strelchenko *et al.* (2004) and Wei *et al.* (2005).

The moderate association between genetic variations estimated using PC analysis, factor analysis and clustering procedure can be explained by a range of factors. The cluster analysis provides a wider usage of observed total variation than the PC analysis or factor analysis due to plotting of only two first components or factors. Thus, the cluster analysis depicts more precisely than the PC analysis and factor analysis the relationships between the lines and cultivars. The association between estimates is also influenced by the fact that most of the survived variation detected by PC analysis or factor analysis. The results of factor analysis are more favorable than PC analysis due to varimax rotation (Mohamed, 1999). In addition, the rotated components via varimax method could be interpreted from the opposition of few variables with positive loadings to few variables with negative loadings and searched for a linear combination of the original factors such that the variance of the squared loadings is maximized.

The observed high variation among bread wheat genotypes due to morphological traits would be reflecting their genotypic differences or can be explained on the basis of the environmental factors (Moriss, 2009). The morphological traits are subject to both natural and artificial selection, aside from their high environmental dependence. Therefore, it is clear that if there is an association between the loci controlling the targeted morphological trait, the influence of environmental factors will be low (Mollasadeghi *et al.,* 2012). According to Dreisigacker *et al.,* (2004) there may be some reasons for the above problem; (i) selection based on one environment has not been performed long enough to determine the various genotypes, (ii) fitness responsible genes to one environmental condition are not unique to that environment and may confer fitness to the other environments, and (iii) adaptation to a different environments is not based on accretion of random genes but rather on a limited set of specific genes.

The used multivariate procedures helped in finding contrasting wheat groups for future breeding programs. In our study, genetic similarity as estimated through the PC analysis or factor analysis and clustering method via morphological traits showed relatively the similar results. Crossing of different clusters' genotypes, characterized by tolerance to abiotic stresses and resistance to biotic stresses can be used to produce breeding populations to be used in recurrent selection for improved and combining these resistance to biotic and abiotic stresses. According to Chahal and Gosal (2002), the most important traits in the first PC influence the clustering more than the other no important traits in the first PC. Therefore, in our investigation, differentiation of the bread wheat genotypes into different clusters was because of relatively high contribution of few traits rather than small contribution from each trait. Distribution structure of the genotypes into five distinct clusters indicated the presence of considerable genetics diversity among the genotypes for most of the traits under consideration.

In the cluster analysis, genotypes of cluster I, for resistance to yellow rust and salinity; genotypes of cluster II, for resistance to drought, cold and salinity; genotypes of cluster III for resistance to cold stress and low temperatures; genotypes of cluster IV for resistance to drought, brown rust and yellow rust; and cluster V for resistance to various stresses including biotic and abiotic stresses as well as resistant to lodging and semi-dwarf property.

CONCLUSION

Fifty-six bread wheat genotypes grouped into five clusters showed maximum inter cluster diversity the large magnitude of genetic variability among genotypes shows the presence of excellent opportunity to bring about the improvement through wide hybridization by crossing genotypes in different clusters. The present study provided significant information helpful in future genetic improvement programs of bread wheat.

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