



Genetic resources for enhancing drought tolerance from a mini-core collection of spring bread wheat (*Triticum aestivum* L.)

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ABSTRACT. An enhanced level of drought tolerance in wheat (*Triticum* spp.) may be reached through combining agronomic and physiological traits associated with grain yield under drought conditions. We aimed to explore valuable diversity for the drought tolerance, existed in the core collection of Iranian spring bread wheat landraces. A number of 206 spring bread wheat accessions along with the check cultivar were assessed for grain yield, drought-adaptive traits, and estimated drought tolerance criteria during 2016-17 and 2017-18 growing seasons. Analysis of data using the best linear unbiased predictions (BLUPs) approach revealed that the genotype x environment (GE) interactions accounted for the highest variation in grain yield (36.23%) followed by 1000-kernel weight (35.39%), heading date (21.4%), days to maturity (16.38%), and plant height (5.83%). Using the hierarchical cluster analysis and developed pattern heat map based on the values for the agronomic traits and drought resistance indices, the accessions clustered into nine groups of different sets of agronomic and drought tolerance characteristics. Several accessions with high yield potential, early heading, optimal plant stature and high drought tolerance groups were identified. Three drought selection criteria of stress tolerance index (STI), geometric mean productivity (GMP) and mean productivity (MP) were more effective in identifying accessions producing higher yield under both drought and irrigated conditions. The superior accessions identified in this study may be explored further for breeding new wheat cultivars with enhanced level of drought tolerance.

Keywords: wheat; landraces; phenotypic diversity; grain yield; agronomic traits; stability performance.

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Introduction

Wheat is an important crop and one of the most essential commodities in the global market (Curtis & Halford, 2014). Many studies have emphasized increasing wheat production to meet growing global demand (Tilman, Balzer, Hill, & Befort, 2011; Ray, Mueller, West, & Foley, 2013; Mohammadi, 2018). According to Food and Agriculture Organization (FAO, 2018), 746.6 million tons of wheat was produced globally in 2018, to which Iran contributed about 13.5 million tons. In recent decades, predicting severe droughts in the Mediterranean basin and similar regions, has become more complicated due to unfavorable climate changes. Breeding drought tolerant cultivars is considered as the most effective approach towards a sustainable wheat production in the Mediterranean dryland regions. Constructing a core collection, as a subset of entries representing the most of diversities available in the entire collection, facilitates efficient utilization of genetic resources in breeding programs (Upadhyaya et al., 2009; Wang et al., 2013).

Several traits proven to contribute in improving grain yield under water-limited environments. Earliness is considered as an important trait for increasing yield productivity in the dry Mediterranean conditions, where wheat plants are affected by terminal drought and heat stresses (Reynolds, Dreccer, & Trethowan, 2007; McIntyre et al., 2010; Sharma, Upadhyaya, Manjunatha, Rao, & Thakur, 2012; Shavrukov et al., 2017; Crespo-Herrera et al., 2018). Drought tolerance in wheat may be evaluated using most recommended yield-based drought tolerance criteria (Dodig, Zoric, Kandic, Perovic, & Šurlan-Momirović, 2012). The stress susceptibility index was suggested by Fischer and Maurer (1978) to screen drought resistant/susceptible genotypes. Later, the mean productivity proposed by Rosielle and Hamblin (1981) and the tolerance index defined by Hossain, Sears, Cox, and Paulsen (1990) were applied for the evaluations of drought tolerance. Fernandez (1992 quoted in Mohammadi, 2016), introduced the drought stress tolerance index and geometric mean productivity for the assessment of drought tolerance in crop breeding programs. The mentioned indices are determined based on mathematical relationships between yields under drought and irrigated conditions.

Significant genotype x environment (GE) interactions affecting grain yield, limit the progress of selecting superior genotypes in crop breeding programs (Reddy, Rathore, Reddy, & Panwar, 2011). Many studies have investigated positive and negative effects of GE interactions on yield and its related traits under different growing conditions (Yan, Cornelius, Crossa, & Hunt, 2001; Alwala, Kwolek, Mcpherson, Pellow, & Meyer, 2010; Luo et al., 2015). The GE interaction effects have been analyzed using several statistical linear (Yates & Cochran, 1938; Eberhart & Russell, 1966) or multivariate methods (Zobel, Wright, & Gauch Jr., 1988; Gauch Jr., 1992; Yan, Hunt, Sheng, & Szlavnic, 2000; Yan & Kang, 2002). Among the multivariate methods, the GGE biplot, provides a powerful tool for graphical analysis of GE interactions in multi-environment trials (METs), that reveals a clear demonstration of genotype and GE interactions. In addition, GGE biplot provides an easy-to-use method for analyzing yield stability and mega-environment investigations (Yan et al., 2000; Yan & Kang, 2003).

This study aimed to assess grain yield, important agronomic characteristics, yield stability performance, and to explore valuable diversity for drought tolerance existed in the core collection of Iranian spring bread wheat landraces to use in the wheat breeding program.

Material and methods

Experimental layout and data recorded

A core collection of spring bread wheat landraces including 206 accessions of Iranian origin were evaluated along with a local check cultivar (Rijaw - Table 1) for their agronomic performance and drought tolerance. The diversity core collection was developed at CIMMYT by examining 2,403 Iranian bread wheat landraces through field evaluations for heat and drought tolerance and molecular screening based on SNP markers (Crossa et al., 2016). This collection assembled at CIMMYT, which represents 10% prediction core set of the Iranian landraces. The selection of the core set was based on the reliability measure of VanRaden (2008). The core set may serve as training population to predict the remaining accession in the original collection (Crossa et al., 2016). To further evaluate the provided core collection for drought tolerance under dryland conditions, we carried out two field experiments under rainfed and supplemental irrigation conditions during two cropping seasons of 2016/17 and 2017/18. Irrigated experiments received two irrigations, each with 30 mm using sprinkler system, at anthesis to mid-grain filling period, to mitigate terminal drought stress in the study. In the first year, entries were planted in a two-row plot with two m length and 0.2 m row spacing in a non-replicated trial (due to seed limit) and, in the second year, with the same plotting area, entries were evaluated in experiments based on an alpha-lattice design with two replications. The local check spring wheat cultivar, Rijaw, was repeated 13 times in each trial, distributed throughout the field to provide measures for adjusting data collected from the field trials.

Experiments were undertaken at Sararood dryland agricultural research station (34°19' N, 47°17' E; 1,351 m a.s.l), the main station for breeding crops targeting regions with moderate cold climates in the west of Iran. The soil texture was silty-clay-loam at the research site. Weeds were controlled by herbicide and hand weeding as required. Fertilizers were used at rates of 50 kg N ha⁻¹ and 50 kg P₂O₅ ha⁻¹ at the time of planting. Agronomic and phenological traits were recorded for all accessions. Heading date (DHE) was recorded when about 50% of spikes in each plot were fully emerged. Days to maturity (DMA) was recorded when about 50% of peduncles in each plot turned yellow. Plant height (PLH) was recorded for each accession at maturity stage. After harvest, grain yields were recorded as kg ha⁻¹ for each plot, and 1000-grain weight (TKW) was determined for each accession using a seed counter.

Statistical analysis

Variance components and data description

In each trial, data were subjected to the best linear unbiased predictions (BLUPs) analysis. Then, the BLUPs data in form of adjusted means were subjected to estimate variance components. Variance components of genotype, environment and GE interactions were estimated for each studied trait.

The phenotypic diversity index (*H*), as described by Shannon (1948), was calculated for the core collection based on each studied trait. Values calculated for mean and standard deviation (SD) were used for classifying accessions into five distinct groups: Group I included entries with values between mean ± SD; Group II comprised entries with values greater than mean + SD; Group III consisted of entries with values greater than

mean + 2SD; Group IV included entries with values less than mean - SD; and Group V consisted of entries with values less than mean - 2SD. Later, the proportion of the different groups for each trait was determined and the H index was calculated using the following equation.

$$H = - \sum_{i=1}^n P_i \ln P_i$$

where: n , \ln , and P_i stand for number of groups, natural logarithm, and the relative frequency of the i th group in the collection, respectively.

To estimate the phenotypic and genotypic correlations among the studied traits, the following variance and covariance formula were applied:

$$r_g = \frac{Cov_g 1,2}{\sqrt{(\sigma_g^2 1)(\sigma_g^2 2)}}$$

$$r_p = \frac{Cov_p 1,2}{\sqrt{(\sigma_p^2 1)(\sigma_p^2 2)}}$$

where: r_g and r_p are genotypic and phenotypic correlation coefficients, respectively; $Cov_g 1,2$ and $Cov_p 1,2$ represent genotypic and phenotypic covariances for attributes 1 and 2, respectively; $\sigma_g^2 1$ and $\sigma_g^2 2$ are genotypic variances for attributes 1 and 2, respectively; $\sigma_p^2 1$ and $\sigma_p^2 2$ represent phenotypic variances of attributes 1 and 2, respectively.

Selection indices for drought tolerance

Based on BLUP's data on grain yield under drought and irrigated conditions, selection drought tolerance indices included 1) stress tolerance index (STI; Fernandez, 1992 quoted in Mohammadi, 2016) = $[(Y_s)(Y_p)]/(Y_{pm})^2$; 2) geometric mean productivity (GMP; Fernandez, 1992 quoted in Mohammadi, 2016) = $\sqrt{Y_s Y_p}$; 3) mean productivity (MP: Rosille and Hamblin) = $(Y_s + Y_p)/2$; 4) tolerance index (TOL; Hossain et al., 1990) = $(Y_p - Y_s)$; and stress susceptibility index (SSI; Fischer & Maurer, 1978) = $[1 - (Y_s/Y_p)]/(1 - SI)$, where SI (stress intensity) = $(1 - Y_{sm}/Y_{pm})$, where Y_s and Y_p represent the accessions yields under drought and irrigated conditions, respectively, and Y_{sm} and Y_{pm} stands for mean yields of accessions under drought and irrigated environments, respectively.

A scatter plot using Y_s (X-axis) and Y_p (Y-axis) was developed to group the evaluated accessions, as suggested by Fernandez (1992 quoted in Mohammadi, 2016). The X-Y plane was divided into four groups of A, B, C and D by drawing intersecting lines through mean values of Y_s and Y_p . Group A, consisted of accessions expressing superiority under both Y_s and Y_p conditions; while group B, contained accessions performed well under irrigated condition; group C, included accessions producing relatively higher performance under drought condition; and in group D, accessions with low yield under both conditions were located.

A principal component analysis (PCA) based on collected data and estimated drought tolerance selection indices were applied to characterize drought tolerance in the examined accessions.

Cluster and biplot analyses

The hierarchical cluster and discriminant analyses based on the studied traits and estimated drought tolerance selection indices were applied to classify accessions in the examined core collection.

The GGE biplot methodology, as suggested by Yan et al. (2001), was applied for graphical analysis of genotype by environment interactions using MET data. The which-win-where pattern analysis, integrating both yield and stability performances, was used to identify ideal accessions in the study.

A genotype by trait (GT) biplot analysis, as suggested by Yan and Rajcan (2002), was applied to reveal variations among the accessions and trait profiles of the core collection.

The variance components, GGE and GT bipots were completed using the R software (R Core Team, 2016) with the packages of GEA-R (Pacheco et al., 2015) and Meta-R (Alvarado et al., 2015). The scatter plot, principle component analysis (PCA), cluster and discriminate analyses were performed using the SPSS software (IBM SPSS Statistics V21.0).

Results and discussion

Weather conditions

Two cropping seasons during 2016-17 and 2017-18 varied in the amount of annual precipitations and its seasonal distribution (Figure 1), providing different growth conditions that lead to a wide range of yield productivity. The annual precipitations recorded on 2016-17 and 2017-18 cropping seasons were 492.1 and 521.2 mm, respectively. Although, the annual precipitation during growth seasons exceeded the long-term average (445 mm per year), but crops experienced severe terminal drought due to the lack of rainfall coincided with high temperature during grain filling period. The monthly averages of minimum and maximum temperatures were -4.8 and 34.3°C, respectively, in 2016-17, and -3.6 and 30.8°C, respectively, in 2017-18. No rainfall received in October and November, resulted in late germination till mid-December, 2016. Crops were also affected by low temperature at stem elongation stage in February, 2017. In the both cropping seasons, drought and high temperature are dominant factors during grain-filling stages, with high potential impact on crop productivity. This phenomenon often shortened grain filling period, reducing grain weight that resulting in yield loss under the rainfed conditions of Iran.

Variance components and data description

Variations explained by GE interactions accounted for the highest variance in grain yield (36.23%) followed by 1000-kernel weight (35.39%), heading date (21.4%), days to maturity (16.38%) and plant height (5.83%). In contrast, genotype expressed the highest variations in heading date (56.7%) followed by 1,000-kernel weight (19.87%), days to maturity (17.65%), grain yield (15.48%) and plant height (7.40%). The highest variations explained by environments were observed in plant height (86.59%), followed by grain yield (47.77%), 1000-kernel weight (42.85%), days to maturity (24.62%) and heading date (2.80%), respectively (Table 1).

Within the core collection, mean grain yield for the evaluated accessions ranged from 2,543 to 4,233 kg ha⁻¹, with an overall mean of 3,395 kg ha⁻¹ (Table 1). The TKW ranged from 30.6 to 45.2 g with an overall mean of 37.9 g; plant height measured from 54 to 117 cm with an overall average of 98 cm; heading date varied from 130-155 days with the average heading days of 143; and days to maturity varied from 177-189 days with an overall mean of 183 days (Table 1).

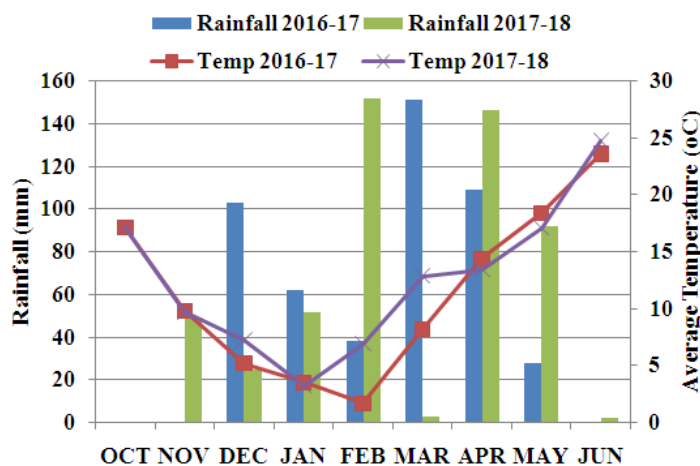


Figure 1. Monthly rainfall patterns and average temperatures recorded during the two cropping seasons.

Table 1. The variance components estimated for genotype, environment and GE interaction, and some overall statistics based on the BLUP's data for grain yield and agronomic traits collected from the spring wheat core collection grown across four environments.

Variance components	BLUP_DHE		BLUP_DMA		BLUP_PLH		BLUP_TKW		BLUP_YLD	
	Estimate	%	Estimate	%	Estimate	%	Estimate	%	Estimate	%
Genotype (G)	36.36	56.7	8.78	17.65	78.64	7.40	11.91	19.87	162699.14	15.48
Environment (E)	1.78	2.8	12.24	24.62	919.95	86.59	25.68	42.85	501987.07	47.77
G x E	13.72	21.4	8.15	16.38	61.93	5.83	21.21	35.39	380717.88	36.23
Grand Mean	143.1		182.9		99.7		37.9		3395.3	
Range	130-155		177-189		54-117		30.6-45.2		2543-4233	

DHE: days to heading; DMA: days to maturity; PLH: plant height; TKW: thousand kernel weight.

Traits distribution and accession frequencies

The percentages of accessions that exceeded the collection mean, 'mean+SD', 'mean+2SD' or 'mean-SD' and 'mean-2SD' for each trait is presented in Table 2. In the case of grain yield and TKW, the percentages greater than 'mean+SD' and 'mean+2SD' were used, while for the phenological traits, the 'mean-SD' and 'mean-2SD' were applied, as earliness is desired under drought conditions. In the case of grain yield under stress condition, 15.9 and 1.4% of the accessions showed values greater than the 'mean+SD' and 'mean+2SD', respectively. That is, 32 accessions exceeded the 'mean+SD', compared with only two or three accessions exceeded the 'mean+2SD' (Table 2). For grain yield under irrigated condition, 16.9 and 2.9% of the accessions yielded higher than the 'mean+SD' and 'mean+2SD', respectively. In the case of TKW, 16.4% of the accessions were greater than 'mean+SD', while 2.4% of accessions exceeded the 'mean+2SD'. For plant height, 13.5% of accessions were taller than 'mean+SD', and only 0.5% were taller than 'mean+2SD'. Around 17 and 1% of accessions, respectively, entered heading stage earlier than the collection 'mean-SD' and 'mean-2SD'. In the case of days to maturity, 16.4 % of accessions showed earlier maturity than the 'mean-SD' (Table 2).

In the case of drought tolerance indices, 16.4 and 2.9% of accessions with STI values greater than collection 'mean+SD' and 'mean+2SD', respectively, were identified as the most drought tolerant accessions across years. For MP, 15.9 and 1.9% of accessions exceeded the 'mean+SD' and 'mean+2SD', respectively, indicating superiority of these accessions for productivity under both drought and irrigated conditions. Similarly, 12.1% of accessions had GMP values greater than 'mean+SD'. TOL and SSI indices, indicating drought resistance may serve as important traits for selecting accessions resistant to severe drought stress conditions. 14.5 and 3.9% of accessions with TOL values less than collection 'mean-SD' and 'mean-2SD' were identified, respectively. Similar trends were also found for SSI, showing the capacity of the examined collection for selecting drought resistant accessions.

Phenotypic diversity within the core collection

The Shannon's diversity index (H) calculated for the studied traits (Table 3), indicated considerable diversity within the core collection across all test environments. The average phenotypic diversity value for the core collection was 1.0. The highest H value was found for the grain yield under irrigated condition (1.049) followed by grain yield under drought stress condition (1.0), plant height (0.993), days to maturity (0.992), 1,000-kernel weight (0.943) and heading date (0.917).

Phenotypic and genotypic correlations

Figure 2 presented the correlation heat map among traits recorded both under drought and irrigated conditions. Significantly positive genotypic and phenotypic correlations were observed between TKW and grain yield (Figure 2A), indicating that accessions with higher TKW also produced higher grain yield in the evaluated core collection. The significant negative correlation between TKW and days to heading, suggested that accessions with early heading tend to have higher TKW. Therefore, selection based on TKW may lead to increased grain yield under drought conditions. Plant height showed positive correlation with phenological traits, suggesting that accessions with early heading and maturity tend to have low to moderate plant height.

Under irrigated conditions, heading date showed significantly positive phenotypic and genotypic correlations with plant height and days to maturity; and negative correlation with TKW and grain yield (Figure 2B). Plant height and TKW also showed significant positive genotypic as well as phenotypic correlations with grain yield.

Table 2. Percentage of spring bread wheat accessions identified as better than the collection average for the studied traits across environments.

Groups of diversity	Traits					
	Ys	Yp	DHE	DMA	PLH	TKW
G1 (Mean ± SD)	66.2	61.8	68.1	66.7	66.7	67.6
G2 (> Mean + SD)	15.9	16.9	13.0	12.6	13.5	16.4
G3 (> Mean +2SD)	1.4	2.9	1.0	2.9	0.5	2.4
G4 (< Mean - SD)	13.5	17.4	16.9	16.4	14.5	13.0
G5 (< Mean - 2SD)	2.9	1.0	1.0	1.4	4.8	0.5

Ys: grain yield under drought condition; Yp: grain yield under irrigated condition; DHE: days to heading; DMA: days to maturity; PLH: plant height; TKW: thousand kernel weight; LSD: least significant difference.

Table 3. Shannon-Weiner diversity index (H) and group frequencies for the studied traits in the spring bread wheat core collection.

Groups of diversity	Traits					
	Ys	Yp	DHE	DMA	PLH	TKW
Group I (Mean \pm SD)	0.273	0.297	0.262	0.270	0.270	0.264
Group II (> Mean + SD)	0.293	0.301	0.266	0.261	0.271	0.297
Group III (> Mean + 2SD)	0.061	0.103	0.045	0.103	0.026	0.090
Group IV (< Mean - SD)	0.271	0.304	0.301	0.297	0.280	0.266
Group V (< Mean - 2SD)	0.103	0.045	0.045	0.061	0.146	0.026
H	1.000	1.049	0.917	0.992	0.993	0.943

Ys: grain yield under drought condition; Yp: grain yield under irrigated condition; DHE: days to heading; DMA: days to maturity; PLH: plant height; TKW: thousand kernel weight; LSD: least significant difference.



Figure 2. Phenotypic (below diagonal) and genetic (above diagonal) correlations between grain yield and agronomic traits for 207 examined spring bread wheat accessions across two cropping seasons under drought (A) and irrigated (B) conditions. Ys: grain yield under drought condition; Yp: grain yield under irrigated condition; DHE: days to heading; DMA: days to maturity; PLH: plant height; TKW: thousand kernel weight.

Selection for yield and stability performance

Due to significant effects found for GE interactions on grain yield and other studied traits, GGE biplot analysis provided more insights on genotypes stability, environments effect and relationships among environments as well as among the investigated traits. The GGE biplot constructed for grain yield captured 90.3% of the total variations (Figure 3). Using the which-win-where pattern, the biplot was divided into seven sectors, with two sectors each comprising two environments (Figure 3A), which each sector representing different mega-environments (ME). Accession no. 102 was the winning genotype in the ME represented by the environments YR17 and YI17; while accession no. 191 was the best performed genotype in test environments YR18 and YI18. These findings demonstrated a significant separation between two cropping seasons having two different top-yielding genotypes, while the water regime conditions in each year were not significantly interacted with the genotype effect.

Spring bread wheat accessions in the core collection with stable above-or below-average mean yield were screened by GGE biplot (Figure 3B). Of these, accessions no. 57, 133, 15, 16, 60, 14, 130, and 128 were found stable with high mean yield. Conversely, accessions no. 145, 30, 98, 40, 6, and 8 were identified as stable but with low grain yield production. Accessions no. 57, 133, 130, 161, 15, and 128 were identified as ideal accessions with high mean yield and high stability performances across environments (Figure 3C).

Trait relations and traits profiles of accessions

To better understand the relationships among the studied traits and to demonstrate traits profiles for the examined accessions, a GT-biplot was constructed based on data collected across environments (Figure 4). The constructed biplot explained 60.1% of the total variations. Accession no. 204 with the highest values for DHE and DMA, was the latest in heading and maturity among the evaluated accessions (Figure 4A). Other accessions, with most late flowering and maturity included 206, 125, 63, 201, 202, and 127. In contrast, the accession no. 184 with the highest values for YLD, TKW, and PLH found to be most early in heading and maturity. Other accessions with early heading included 73, 111, 200, 136, 71, 67, 189, 124, 17, 133, 23, and

137. The obtuse angles between DHE and grain yield vectors as well as between DMA and grain yield vectors indicated that in our trials, phenological traits DHE and DMA negatively affected grain yield (Figure 4B). In contrast, TKW and PLH both showed positive effects on the grain yield.

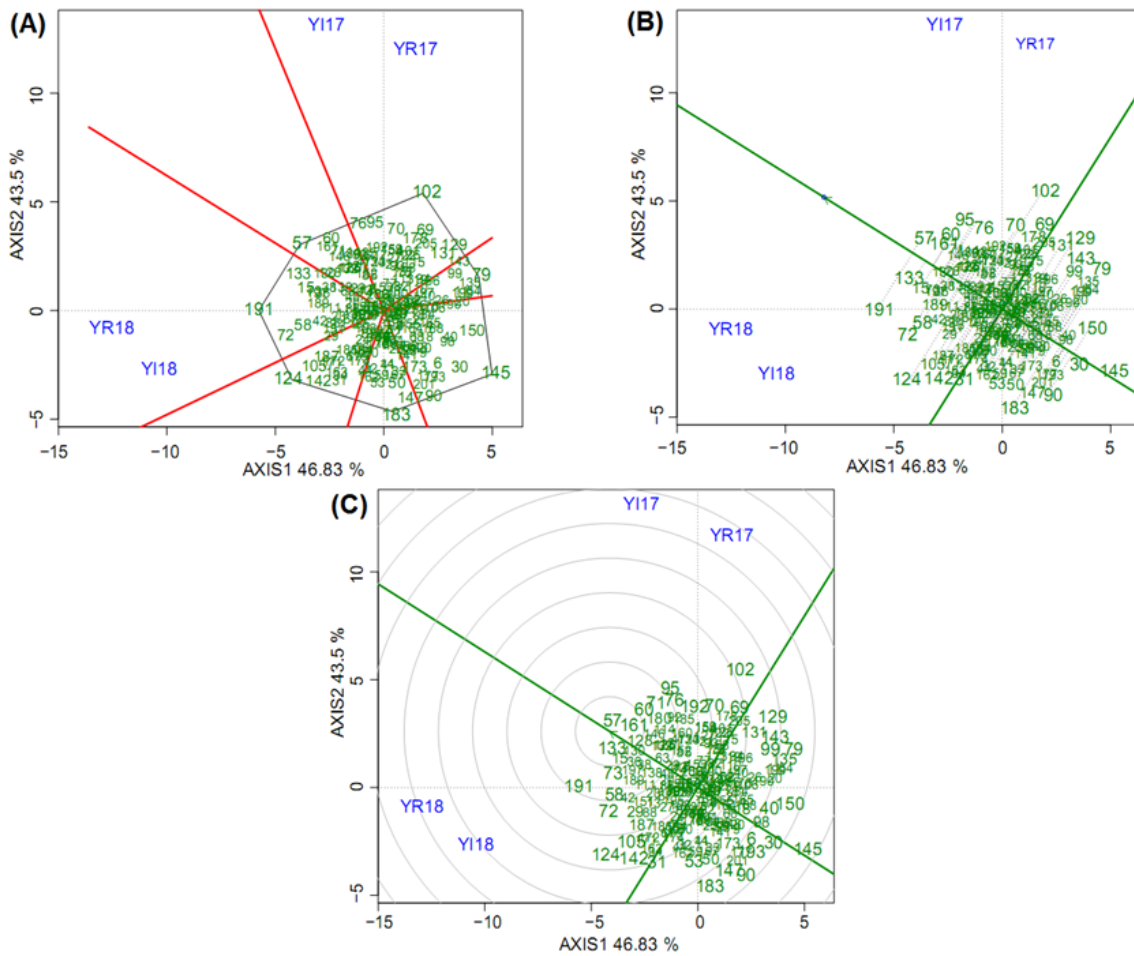


Figure 3. The which-win-where pattern (A), integrating yield and stability performance (B) and ideal accessions (C) based on GGE biplot analysis for grain yield of 207 spring bread wheat accessions across four test environments. Entries 1-207 represent the accessions number; environmental codes YR17, YR18 stands for drought conditions in 2016/17 and 2017/18 cropping seasons, respectively; and YI17 and YI18 stands for irrigated conditions in 2016/17 and 2017/18 cropping seasons, respectively.

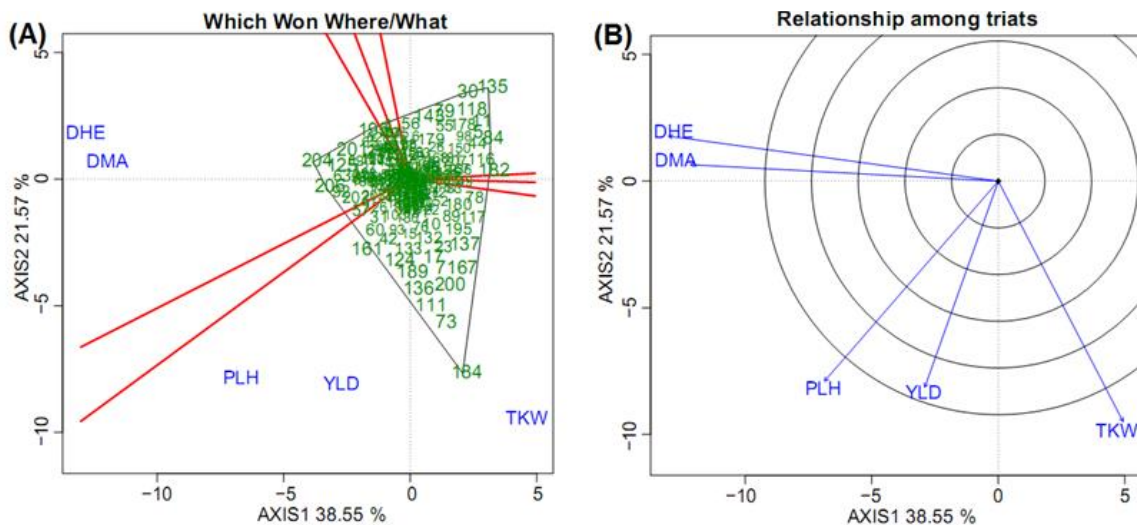


Figure 4. The GT biplot showing (A) which-for-what pattern and (B) relationships among the studied traits for 207 spring bread wheat accessions across four test environments. YLD: grain yield; DHE: days to heading; DMA: days to maturity; PLH: plant height; TKW: thousand kernel weight.

Quantifying drought tolerance for the core collection

Although in overall, a linear relationship was observed between grain yields of accessions under both environmental conditions across years, there was sufficient dispersal indicating that all accessions did not respond similarly to drought conditions (Figure 5A). Thus, as previously reported, a genotype with high yield performance under drought conditions may not essentially perform well under non-stress conditions (Fernandez, 1992 quoted in Mohammadi, 2016). Therefore, breeders need to distinguish between genotypes which produce high yield under drought conditions due to their intrinsic high yield potential from those producing higher yield due to enhanced drought tolerance. Simultaneous consideration of grain yield potentials of wheat accessions under drought and irrigated conditions, with drought tolerance indices such as STI, GMP, MP, TOL and SSI, may facilitate identifying genotypes best fitted for growing and producing higher yield under stressed conditions. Genotypes with higher STI values for example, possess valuable characteristics that prevent yield loss under drought conditions, and then will be desirable genotypes if their productivity is high. Figure 5A present a scatter plot based on grain yields under both drought and irrigated conditions which allowed separation of accessions into four distinct groups, designated as groups A, B, C, and D. Accessions classified in group A were recognized as desirable, performed well under both conditions. 79 accessions were placed in group A with the highest drought tolerance (Figure 5B). 21 accessions performed well only under irrigated conditions and classified in group B, with average STI values less than unit. 24 accessions performed well just under drought conditions which classified as group C with average STI values less than unit; Finally, 84 accessions with poor performance under both experimental conditions were classified as group D. This group of accessions may be discarded as undesired genetic materials with no value for improving drought tolerance.

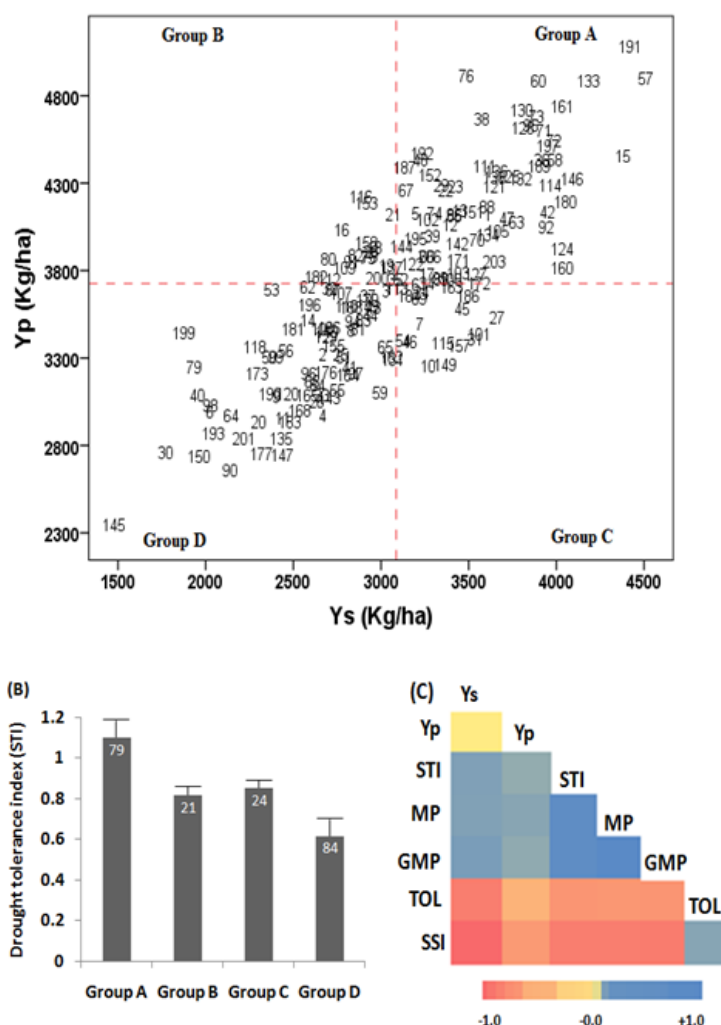


Figure 5. (A) Accession groups based on grain yield under rainfed (Ys) and irrigated (Yp) conditions. Groups A, B, C and D are defined according to Fernandez (1992 quoted in Mohammadi, 2016); (B) drought tolerance index and standard error for accessions included in the groups A, B, C and D; (C) heat map showing Pearson's correlation coefficients among drought resistance indices and grain yields of 207 spring bread wheat accessions across two cropping seasons.

Among the estimated drought selection criteria, STI, GMP and MP were found to be significantly associated ($p < 0.01$; Figure 5C) with grain yields under both conditions, and proved to be effective in selection of germplasms with enhanced level of drought tolerance. Accessions with the highest values for STI, GMP and MP indices mainly included genotypes in group A that may be regarded as accessions with high grain yield potential and most promising for enhancing drought tolerance in wheat cultivars.

The PCA analysis constructed based on estimated drought tolerance indices for 207 accessions, revealed interesting relationships among estimated indices and grain yield (Figure 6). The first and second factors captured 72.4 and 27.3% of total variations, respectively; together explained up to 99.7% of variations observed in the examined core collection. There were strong positive associations among STI, GMP, MP and grain yields under both studied conditions, which appeared to be prominent characteristics for a set of accessions in the examined core collection. These indices were negatively correlated with TOL and SSI indices, found to be associated with a different set of accessions. The set of accessions characterized by STI, GMP and MP indices are considered as the most drought tolerant genotypes. In contrast, the set of accessions characterized by the SSI and TOL indices identified as the most drought susceptible genotypes (Figure 6).

Pattern map of accessions for agronomic performance and drought tolerance

The results of hierarchical cluster analysis for the 207 examined accessions and the pattern heat map developed based on the studied traits and drought tolerance indices, allowed identifying nine distinct accession groups that may be explored as potential genetic materials in spring wheat breeding programs (Figure 7). Accessions showing a good level of grain yield, drought tolerance, and agronomic characteristics were selected. The group G-1 consisted of 39 accessions characterized with low yield, late heading, late maturity and susceptibility to drought condition. A few accessions i.e., 2, 81, 165, 166, 167 and 168 in this group were found to be earlier in heading and accessions no. 147 and 143 showed high values for TKW. In general, agronomic characteristics and drought tolerance in this group ranked below the average of accessions examined in the core collection. Group G-2 comprised of 37 accessions, that showed values below the average in terms of agronomic characteristics and drought tolerance; however, this group ranked

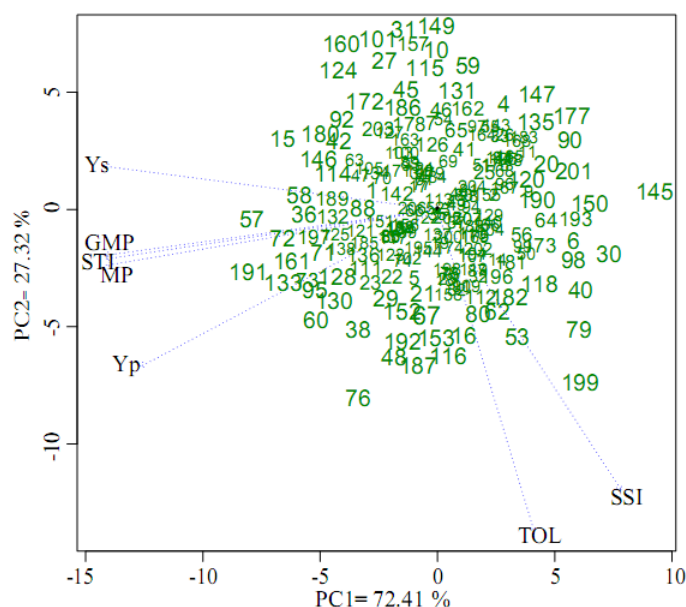


Figure 6. PCA-based biplot showing relationships among drought tolerance indices and grain yields of 207 wheat accessions under both rainfed and irrigated conditions across two cropping seasons. Ys: grain yield under drought condition; Yp: grain yield under irrigated condition; STI: stress tolerance index; MP: mean productivity; GMP: geometric mean productivity; TOL: tolerance index; SSI: stress susceptibility index.

above the group G-1. In this group few accessions were selected as desirable genotypes for further use in wheat breeding programs. For example, accessions no. 110, 188, 170, 110 and 170 were characterized as genotypes with high yield potential as well as high drought tolerance. Accessions no. 19 and 37 showed higher TKW, accessions no. 202 and 170 were among the earliest in entering heading stage; and accessions no. 198, 53, 87 and 75 were early at maturity. Furthermore, in this group, the accessions no. 204, 206, 202 and 188 with the lowest values for SSI index were found to be most resistant to drought conditions. The group G-3 included

14 accessions with mean grain yield, heading, maturity and drought tolerance below the average, while having plant height and TKW greater than the average. Some exceptions were observed in this group including accession no. 99 with high yield and drought tolerance; accessions no. 40, 64, 50, and 99 with lowest TKW; accessions no. 190, 150 and 20 showing most early heading; and accessions no. 199, 150 and 50 with most early maturity date. The fourth group (G-4) consisted of seven accessions including check cultivar. This group was mainly characterized as being early in heading and maturity; while scored as below average for other studied traits. The accession no. 5 in this group produced acceptable yield with moderate drought tolerance, most early heading, low plant height and low TKW. The next group (G-5) consisted of 32 accessions, mainly characterized as high yielding under irrigated condition with higher TKW under drought conditions. Some accessions in this group i.e., 22, 29, 23, 117, 152 may be considered as ideal genotypes due to good agronomic characteristics such as: high yield potential under both studied conditions, high drought tolerance, early heading and high TKW values. The sixth group (G-6) included 24 accessions, of which, all well performed under both conditions and expressed high drought tolerance. Accessions in group G-6 mainly were early in heading and maturity, expressed higher plant height and higher TKW. The next group (G-7) consisted of 33 accessions with desirable agronomic characteristics and drought tolerance greater than the average. The group G-8 comprised 13 accessions characterized with higher grain yields under both studied conditions, higher drought tolerance, early heading and maturity, lower plant height and TKW higher than the average. The last accession group (G-9) consisted of eight accessions with grain yield under stressed conditions higher than irrigated conditions, resistant to drought, high plant height, moderate heading and maturity, and TKW lower than the average.

In our experiment, there was remarkable interaction between genotype, environment and their interaction for grain yield and other studied traits. The cropping seasons varied in total rainfall and their monthly distribution and temperature (minimum and maximum during crop growth; Figure 1) which provided different growing conditions, leads to terminal drought stress that was coincided with terminal heat stress. However, this climatic condition is a typical phenomenon in the Mediterranean conditions including west of Iran. Considerable phenotypic diversities were observed among the examined accessions that deserved further evaluations and utilizations in wheat breeding programs. Several cluster-based groups of accessions were identified with certain superiority in terms of grain yield and other important characteristics. The superior wheat accession groups included: groups G-4 (accessions with early heading and maturity), G-5 (accessions with high grain yield under irrigated condition and high TKW), G-6 (accessions with high yields and high tolerance to drought, early heading and maturity), G-8 (accessions with high grain yield under both conditions, high drought tolerance, early heading and maturity) and G-9 (accessions with high yield under stress conditions, resistant to drought, high plant stature, and moderate in heading and maturity). The characterized trait-specific groups will facilitate the effective utilization of accessions in wheat breeding programs (Basu, Ramegowda, Kumar, & Pereira, 2016; Mohammadi, Etminan, & Shoshtari, 2019).

Traits such as TKW, PLH, DHE, and DMA with higher heritability than grain yield may be used for indirect selection. Such traits strongly correlated with grain yield, are recommended for enhancing grain productivity under Mediterranean environments (McIntyre et al., 2010; Gizaw, Garland-Campbell, & Carter, 2016). The analyses of phenotypic and genotypic correlations are useful to understand the nature of correlations and inheritance of the traits (Lopes et al., 2015). In this research, for the studied traits, genotypic correlations were slightly greater than their corresponding phenotypic correlations, suggesting strong effects of environmental conditions on the studied traits (Johnson, Robinson, & Comstock, 1955). The robust negative phenotypic and genotypic correlations between grain yield and heading date indicated the possibility of selecting accessions with high grain yield and early heading from the examined core collection. Previous studies suggested that selection for early heading, by minimizing the effect of terminal drought stress, will enhance yield productivity under stressed conditions (Vita et al., 2007; González-Ribot, Opazo, Silva, & Acevedo, 2017). In addition, selection for high 1000-kernel weight, because of close association with grain yield, may also lead to higher grain productivity. Biplot graphical analysis revealed that yield was positively affected by TKW and PLH; and negatively affected by DHE and DMA (Figure 4). Researchers are keen to find new traits that may serve for indirect selection of genotypes possessing high grain yield potential (Fufa et al., 2005; Gutierrez, Reynolds, Raun, Stone, & Klatt, 2012). Several studies have endorsed the selection for higher TKW to enhance grain yield (Morgounov et al., 2010; Tian, Jing, Dai, Jiang, & Cao, 2011; Zheng et al., 2011; Aisawi, Reynolds, Singh, & Foulkes, 2015). Furthermore, breeding novel genetic materials with early maturity and higher grain yield has been among the most important goals of wheat breeding programs targeting environmental conditions that plants are exposed to terminal drought and heat stresses (Motzo & Giunta,

2007; Vita et al., 2007; Morgounov et al., 2010; Kamran, Randhawa, Pozniak, & Spaner, 2013; Chen et al., 2016; Mondal et al., 2016). However, in some cases, the higher grain productivity is found not to be related with early heading in wheat (Chairi et al., 2018; Flohr et al., 2018). This may be partly explained by the reduced time available for partitioning assimilates from source to sink in wheat (Royo et al., 2007; Zhou et al., 2007).

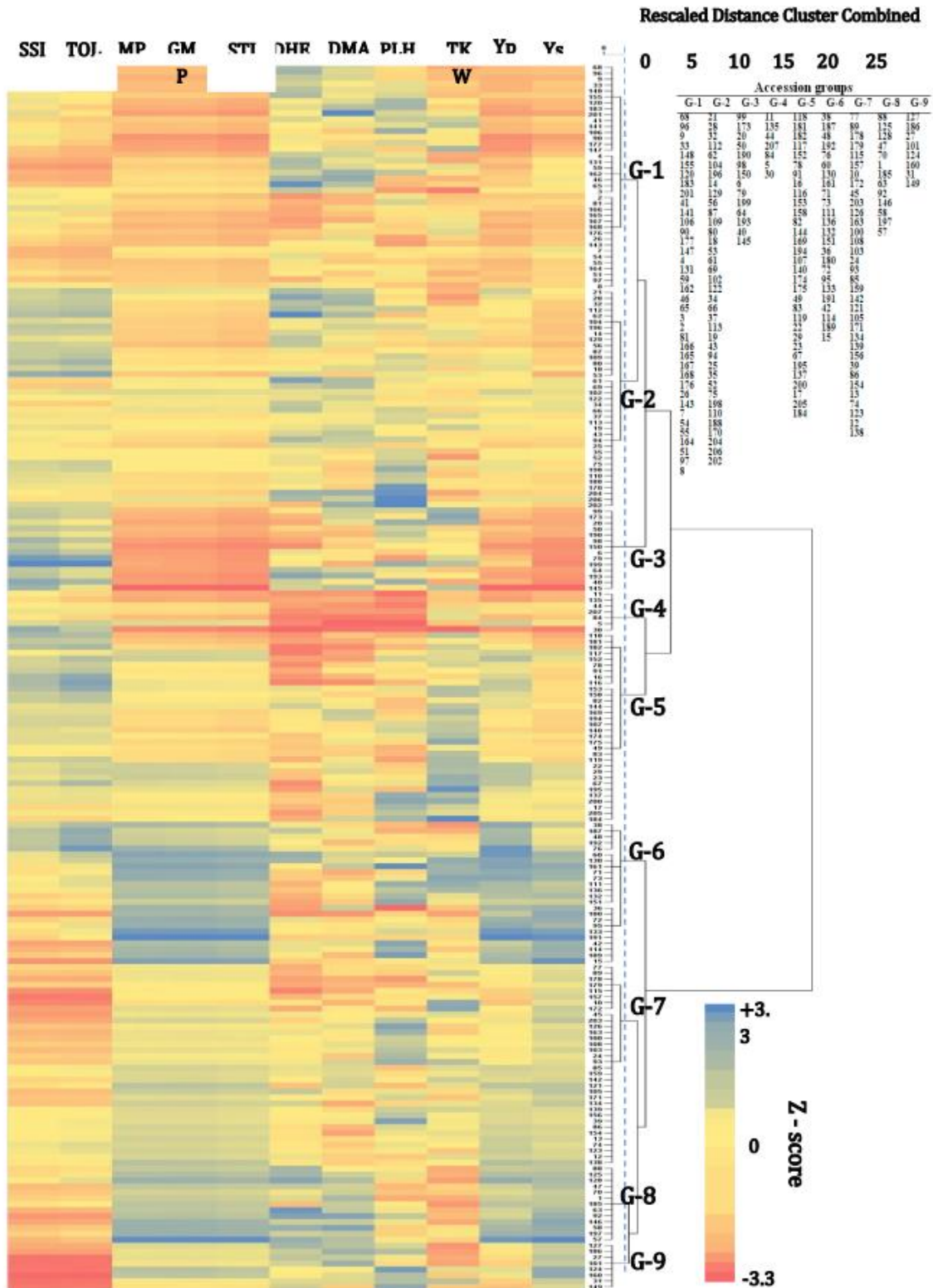


Figure 7. Hierarchical cluster analysis dendrogram (Ward Jr.'s [1963] method) and pattern map based on the values for the agronomic traits and drought resistance indices in the 207 spring bread wheat accessions. Dashed line represents the cut-off line for cluster (nine groups) according to discriminant analysis. Ys, yield under rainfed condition; Yp, yield under irrigated condition; TKW, 1,000-kernel weight; PLH, plant height; DHE, heading date; DMA, days to maturity; STI, drought tolerance index; GMP: geometric mean productivity; MP: mean productivity; TOL, tolerance index; SSI, stress susceptibility index.

The region where this study was performed is characterized as Mediterranean conditions with high variations in the amount and distribution of monthly rainfall in consequent years. This, resulted in high GE interactions and low crop stability from year to year, that represents a complex mega-environment (Mohammadi, Haghparast, Amri, & Ceccarelli, 2010). In this study, we identified a set of wheat accessions i.e., 57, 133, 15, 16, 60, 14, 130, AND 128 with high yield stability performance. However, the examined core collection exhibited a high variation for yield stability, including accession possessing extreme desirable phenotypic characters that present novel genetic materials to expand resources available for improving drought tolerance in wheat.

Breeding for yield under drought prone environments is difficult and received most attentions by wheat breeding programs targeted Mediterranean conditions (Acevedo & Silva, 2007; Mohammadi et al., 2014; González-Ribot et al., 2017). This study demonstrated the benefit of utilizing high yielding accessions with early heading, high grain weight, optimal plant stature and more tolerant to drought. Breeding for grain yield should include genotypes which integrate high yield stability with drought tolerance (Mohammadi, Sadeghzadeh, Armion, & Amri, 2011).

Based on the results, drought selection indices of STI, GMP, and MP found to be more effective for screening accessions with high yield potentials under both drought and irrigated environments. This finding was in agreement with previous studies (Mohammadi, 2016; Nouri, Etminan, Silva, & Mohammadi, 2011), that verified the effectiveness of STI, GMP, and MP indices for screening accessions with high yield under both drought and irrigated conditions.

Conclusion

Several groups of accessions with high stability performance possessing different phenotypic backgrounds i.e., earliness, plant height, grain weight and high level of drought tolerance were identified, that can assist in selection for grain yield, particularly in early generations under drought conditions. A set of accessions characterized by STI, GMP and MP were considered as the most drought tolerant genotypes, while another set of accessions characterized by low values of SSI and TOL identified as accessions resistant to drought conditions. As a conclusion, this study identified accessions with outstanding stable performance, expressing desirable traits in different test environments, which presents plant germplasm suitable for constructing genetic networks on important traits using high throughput genotyping technologies.

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