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Abstract: Grain yield of durum wheat (*Triticum turgidum* L. var. durum) under Mediterranean conditions is frequently limited by both high temperature and drought during grain filling. Genotype-by-environment (GE) interaction and genotype-by-trait (GT) data were analyzed for agronomic performance of durum wheat breeding lines. Data were obtained from 18 durum wheat breeding lines and two cultivar checks (Zardak and Sardari) for their agronomic performance under three different climate locations (moderate, warm and cold winters) and two moisture regimes (rainfed and two supplemental irrigations conditions) in two cropping seasons (2006 and 2007) in Iran. Analysis of GE interaction data based on multiple traits showed that the environment (combination of year-location- moisture regimes) effect was always the most important source of trait variation, accounting for 58.6 to 98.4% of the total variation. Biplot analysis of the studied traits revealed that (i) the locations tended to discriminate genotypes in dissimilar fashions, and (ii) the relationships among traits were not consistent over the locations, where they facilitated visual genotype comparisons and selection at each location.

Key words: Agronomic traits, Biplot analysis, Durum wheat, Genotype-by-environment interaction, Genotype discrimination.

Superior genotypes must be evaluated on the basis of multi-environment trials (MET) and multiple traits to ensure that the selected genotypes have acceptable performance in variable environments within the target region and to meet the many-facets of the demand from the farmers, producers, processors, and the consumers. For this reason, MET are conducted throughout the world for major crops every year in which multiple traits and characteristics are usually recorded (Yan and Rajcan, 2002). Improvement of agronomic traits has been the primary objective of breeders/agronomists for many years under variable environments. Breeders have also measured and selected for grain yield and most related traits such as kernel weight, plant height, and other related traits (Link et al., 1999; Garcia del Moral et al., 2003; Maman et al., 2004; Rubio et al., 2004). All these traits are affected by the growing environment as well as by genetic factors, and numerous studies have described the genotype-by-environment (GE) interactions (Humphreys et al., 1994; Doehlert and McMullen, 2000; Doehlert et al., 2001). However, evaluation of genotypes across diverse environments and over several years is needed in order to identify spatially and temporally stable genotypes that could be recommended for release as new cultivars and/or for use in the breeding programs (Sharma et al., 2010).

The success of durum wheat in Iran, as a food security crop, is largely due to its good ability and capacity to yield well under drought-prone, marginal and under poor management conditions where other crops would fail. However, large differential genotypic responses occur under varying environmental conditions (Bokanga et al., 1994; Mkumbira et al., 2003). This phenomenon is referred to as the GE interaction, and is important in plant breeding programs (Kang, 1998). An understanding of the cause of the GE interaction can help identify superior genotypes based on traits. Usually, a number of genotypes are tested over a number of sites and years and multiple traits are recorded, and it is often difficult to determine the pattern of genotypic performance across environments. Numerous methods have been used to understand the causes of the interactions, although strategies may differ in overall appropriateness. Different methods usually lead to similar conclusions for a given dataset (Flores et al., 1998). Yan et al. (2000), using a site regression model (SREG) combined G and GE interaction, proposed a GGE biplot, constructed from the first two principal components

Table 1. General description of the experimental sites.

Climate	Moderate		Warm		Cold	
Location	Kermanshah		Ilam		Maragheh	
Year	2005–2006	2006–2007	2005–2006	2006–2007	2005–2006	2006–2007
Latitude	34º 19′ N	–	33º 41′ N	–	38º 52′ N	–
Longitude	47º 07′ E	–	46º 35′ E	–	45º 30′ E	–
Altitude (m)	1351	–	975	–	1400	–
Rainfall(mm)	515	552	574	470	382	418
Soil type	Clay-loam	–	loam	–	Clay-loam	–
Temperature*						
Max	38.6	39	41	41	34.6	30.2
Min	−8	−11.6	−2.4	−2.8	−25.5	−25
Average	11.7	10.4	13.9	13.8	6	4.7
No. of days below 0ºC	80	89	38	41	140	148

* From October to June.

derived from the singular value decomposition (SVD) of the environment-centered data. The objectives of the present research were (i) to investigate the efficacy of the different climate testing locations using the biplot technique (Yan et al., 2000) and (ii) to evaluate durum wheat genotypes on the basis of multiple traits as well as to study the interrelationships among durum wheat traits using the genotype-by-trait (GT)-biplot technique.

Materials and Methods

Eighteen durum wheat experimental lines [G1 (44-16-2-4), G2 (25-25-1-5), G3 (40-11-2-3), G4 (20-16-1-4), G5 (18-18-1-4), G6 (74-23-3-5), G7 (73-16-3-5), G8 (29-18-2-1), G9 (71-7-3-5), G10 (57-11-3-1), G11 (43-25-2-4), G12 (19-17-1-4), G13 (409), G14 (42), G15 (278), G16 (Gcn//Stj/Mrb3), G17 (Ch1/Brach//Mra-i) and G18 (Lgt3/4/Bcr/3/Ch1//Gta/Stk)] selected from the joint collaborative project of Iran/ICARDA along with two checks, the bread (G19, "Sardari") and durum (G20, "Zardak") wheat cultivars, were evaluated for two successive cropping seasons (2005−2006 and 2006−2007) at three locations differing in winter temperature regimes (moderate, warm and cold), under rainfed and supplementary irrigated conditions.

The three different climate locations, representative of major rainfed durum wheat growing areas, are located in the moderate (Kermanshah), warm (Ilam location) and cold (Maragheh location) regions of Iran. Additional information on the experimental sites is given in Table 1. In each environment (combination of year-location-moisture regime) the experimental layout was a randomized complete-block design with three replications. At each location the genotypes were sown under both rainfed and low irrigated (with two supplementary irrigations: (i) 25 mm supplied at early flowering and (ii) 25 mm at mid-anthesis stages) conditions. At each location, seeds of each genotype were planted in 6 rows 6 m long

and 20 cm apart (plot size=7.2 m^2). Fertilizer application was 41 kg N ha^{-1} and 46 kg P$_2$O$_5$ ha^{-1} at planting. The most important traits recorded for each genotype at each location and in both rainfed and irrigated conditions during two cropping seasons were: grain yield (YLD), thousand kernel weight (TKW), plant height (PH), days to heading (DH) and days to maturity (DM). Days to heading was designated as the day until 50% of the plants in the plot had at least one open flower. Days to maturity was when 50% of the plants in the plot had yellow leaves. The plant height was measured for each genotype at physiological maturity stage. Following harvest, grain yield and TKW were measured. The data recorded were subjected to analysis of variance (ANOVA). Ordination biplot (Delacy et al., 1996; 2000) analysis was performed on genotype-by-environment interaction and genotype-by-trait data using IRRISTAT statistical software (IRRI, 2005). For each trait a biplot was constructed by plotting the first principal component (PC1) scores of the genotypes and the environments against their respective scores for the second principal component (PC2) that result from singular value decomposition (SVD) of environment-centered to study the GE interaction of each trait and to identify superior genotypes (Yan et al. 2000; Yan and Kang 2003). In the GT biplot, a vector is drawn from the biplot origin to each marker of the traits to facilitate visualization of the relationships among the traits. The correlation coefficient between any two traits is approximated by the cosine of the angle between their vectors. Acute angles show a positive correlation, obtuse angles show a negative correlation and right angles no correlation. The length of the vector describes the discriminating ability of the trait. A short vector may indicate that the trait is not related to other traits, that there is a lack of variation or that it is not suitable for genotype discrimination.

Table 2. ANOVA and variability of agronomic traits between genotypes (G), environment (E) and GE interaction for 20 durum wheat across 12 different environments.

Trait	Source	df	MS	% (G+E+GE)
Yield	Environment	11	6984958 **	68.8
	Genotype	19	165145 **	2.8
	G×E	209	151957 **	28.4
TKW	Environment	11	413.6 **	58.6
	Genotype	19	49.5 **	12.1
	G×E	209	10.9 **	29.3
PH	Environment	11	9138.9 **	74.1
	Genotype	19	1057.0 *	14.8
	G×E	209	72.2 **	11.1
DH	Environment	11	13399 **	98.4
	Genotype	19	12.5 **	0.2
	G×E	209	10.2 **	1.4
DM	Environment	11	16043 **	94.6
	Genotype	19	40.5 **	0.4
	G×E	209	44.3 **	5.0

TKW: thousand kernel weight; PH: plant height; DH: days to heading; DM: days to maturity. *, ** Significant at 5% 1% level of probability, respectively.

Results

1. Genotype × environment interaction analysis for each agronomic trait

The results of ANOVA for each trait [grain yield (YLD), 1000-kernel weight (TKW), plant height (PH), days to heading (DH) and days to maturity (DM)] over all environments (combination of year-location- moisture regimes) are given in Table 2, which presents an overall picture of the relative magnitudes of the G, E and GE variance terms. For all agronomic traits, the environment was always the most important source of variation, accounting for 58.6 to 98.4% of total variance (G+E+GE).

The relative magnitude of the GE interaction with respect to the variability explained by (G+E+GE) for each trait is also given in Table 2. The variation caused by the GE interaction was larger than the variation among genotypes for all traits (except for PH), suggesting the possible existence of different mega-environments. The large variation due to E for each trait, which is irrelevant to genotype evaluation and mega-environment investigation, justifies the selection of biplots based on the site regression model for MET analysis (Yan et al., 2000).

(1) A: Grain yield (YLD)

The visualization of the "which-won-where" pattern of MET data is important for studying the possible existence of different mega-environments in a region (Gauch and Zobel, 1997; Yan et al., 2000, 2001). The polygon view of a biplot is the best way to visualize the interaction patterns between genotypes and environments and to effectively interpret the results of the biplot (Yan and Kang, 2003). The first two principal components (PC1 and PC2) obtained by singular value decomposition (SVD) of the environment-centered data explained 57.6% of the total variability attributable to G+GE of yield data (Fig. 1A). The biplot enabled visual comparison of the locations and genotypes studied and their interrelationships. Genotypes G5 and G18 were identified as the highest yielding genotypes at moderate location, as they were grouped together in one sector of the polygon. The winning genotypes at warm location under both rainfed and irrigated conditions were G17 and G20, while the genotypes G6, G16 and G19 were better adapted to cold location. The genotype G11 was the poorest yielding, as it was located farthest from all location markers. The biplot showed that the three different climate locations can be differentiated from each other and that they can discriminate genotypes in opposite directions. The three locations at 2006–2007 as indicated by the relative length of their vectors rather than 2005–2006, were better for identifying high yielding and adapted genotypes.

(2) B: Thousand kernel weight (TKW)

Fig. 1B is a biplot with a polygon view for TKW and it represents the data of 20 durum wheat entries at 12 different environment combinations. The PC1 and PC2 explained 61.6% of the total variability attributed to (G+GE). The vertex genotypes for TKW are G1, G4, G8, G9, G15 and G17, and the environments fell into the three sectors. Therefore, it seems that genotype G1 had the highest values of TKW at five out of six environments at 2006–2007 cropping season, while the G4 had the highest value of TKW at all environments during 2005–2006 cropping season, indicating that grouping of genotypes was rather under year effect than location effect. In 2006–2007 the moderate location under rainfed conditions made up a single group and the genotype G15 was winner for that, followed by G13 and G18. The vertex genotypes G8, G9 and G17 had the poorest TKW in most of the environments.

(3) C: Plant height (PH)

Fig. 1C presents a polygon view, which indicates that the genotype G2 had the highest plant height under rainfed conditions at cold location as well as under irrigated conditions at warm location. The genotype G3 had the highest PH at moderate location under both rainfed and irrigated conditions in 2006–2007. Genotype G5 had the highest PH in most environments. The breeding lines from ICARDA (G16, G17 and G18) as well as G7 had short PH in most environments. The biplot of PH unlike the biplot of TKW indicated that both year and location effects are important in grouping the genotypes.

(4) D: Days to heading (DH)

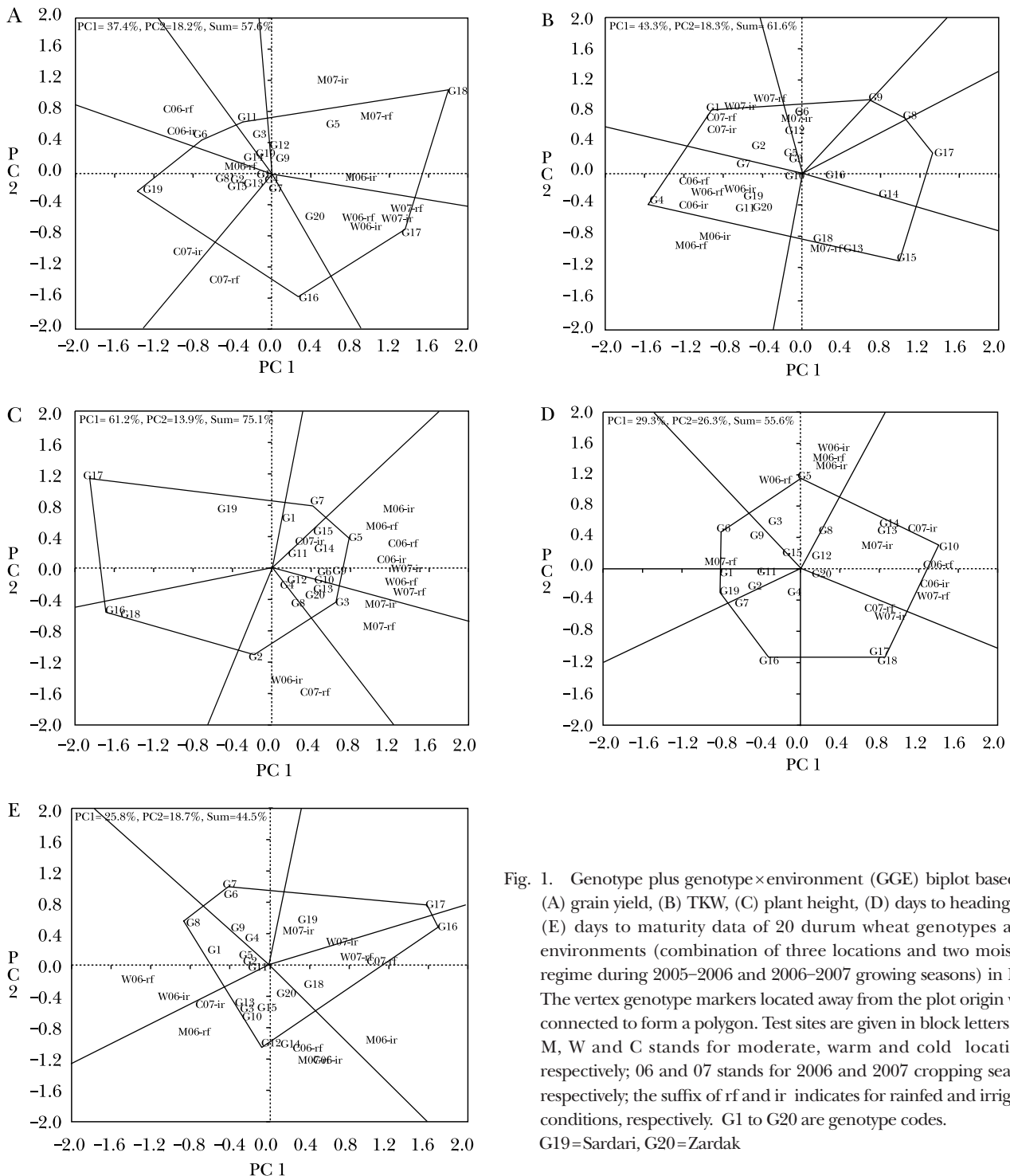Fig. 1D shows a polygon view of a biplot for DH. The proportion of total variation explained by the first two PC

Fig. 1. Genotype plus genotype×environment (GGE) biplot based on (A) grain yield, (B) TKW, (C) plant height, (D) days to heading and (E) days to maturity data of 20 durum wheat genotypes at 12 environments (combination of three locations and two moisture regime during 2005−2006 and 2006−2007 growing seasons) in Iran. The vertex genotype markers located away from the plot origin were connected to form a polygon. Test sites are given in block letters.
M, W and C stands for moderate, warm and cold locations, respectively; 06 and 07 stands for 2006 and 2007 cropping seasons respectively; the suffix of rf and ir indicates for rainfed and irrigated conditions, respectively. G1 to G20 are genotype codes.
G19=Sardari, G20=Zardak

axes was 55.6%. The genotypes and environments fell into six sectors. The vertex genotypes based on DH were G5, G6, G10, G16, G18 and G19. The vertex genotype for each sector is the one that had the most DH for the environments that fall within that sector. The genotypes G10 and G18 followed by G17 had the highest DH at the cold location (in both cropping seasons) and at warm locations (in the 2007 cropping season), while G5 had the highest DH at moderate and warm locations in the 2006 cropping season. G6 had also the highest DH under

rainfed conditions at moderate location. The vertex genotypes G16 and G19 had the lowest DH at most environments indicating that these genotypes tend to flower before other genotypes at all environments. The genotypes G4, G11, G12, G15, and G20 which were near to origin of the biplot had average DH and are more stable than the vertex genotypes.

(5) E: Days to maturity (DM)

Fig. 1E presents a polygon view of a biplot for DM trait, for which the first two PC axes accounted for 44.5% of the

total variability attributable to (G+GE) of DM data. The genotype G12 had the highest value of DM at cold and moderate locations, while G8 was the latest maturing genotype at the warm location in 2005–2006. G16 was the latest maturing genotype under rainfed condition at warm and cold locations in 2006–2007, while G17 was the latest one under irrigated conditions at warm and moderate locations during the 2006–2007 cropping season. The genotype G7 followed by G6 were the earliest in maturity under most environments. The genotypes near to origin biplot (G2, G5 and G11) had average maturities.

2. Genotype-by-trait (GT) biplots and trait relationship analyses

The GT-biplot for each of the three different locations (including two years and two moisture regimes) and based on multiple traits explained between 48.4% (moderate location) to 68.0% (warm location) of the total variation of the standardized data (Fig. 2). This relatively low proportion reflects the complexity of the relationships among the measured traits. Nevertheless, the fundamental patterns among the traits should be captured by the biplots (Kroonenberg, 1995; Yan and Kang, 2003). In the GT-biplot, a vector is drawn from the biplot origin to each marker of the traits to facilitate the visualization of the relationships between and among the traits. Coefficient of correlation (r) between any two traits is approximated by the cosine of the angle between their vectors (Yan and Rajcan, 2002) (e.g., $r=\cos 180^{\circ}=-1$, $\cos 0^{\circ}=1$, and $\cos 90^{\circ}=0$). The GT biplot was used to compare genotypes on the basis of multiple traits and to identify genotypes that possess several desirable traits. Traits with longer vectors are more responsive in genotype discrimination; traits with shorter vectors are less responsive; and those located at the biplot origin are not responsive.

(1) A: Moderate location

Fig. 2A shows a GT-biplot which was used to study relationship among multiple traits and to identify genotypes that were particularly desirable relative to specific (several) trait(s) at moderate locations. The proportion of total variation explained by the first two PC axes was 48.4%. The most outstanding relationships revealed by the biplot were: (i) a strong positive association between PH at both rainfed and irrigated conditions, and between yields under both rainfed and irrigated conditions, as indicated by the acute angles between their vectors; (ii) a strong negative association between PH and yield, between PH and TKW, and between yield and DH as indicated by the obtuse angle between their vectors; (iii) a positive association between DM at both rainfed and irrigated conditions, between yield with TKW under rainfed conditions, and between yield and TKW under irrigated conditions; and (iv) a near-zero correlation between DH under both rainfed and irrigated conditions,
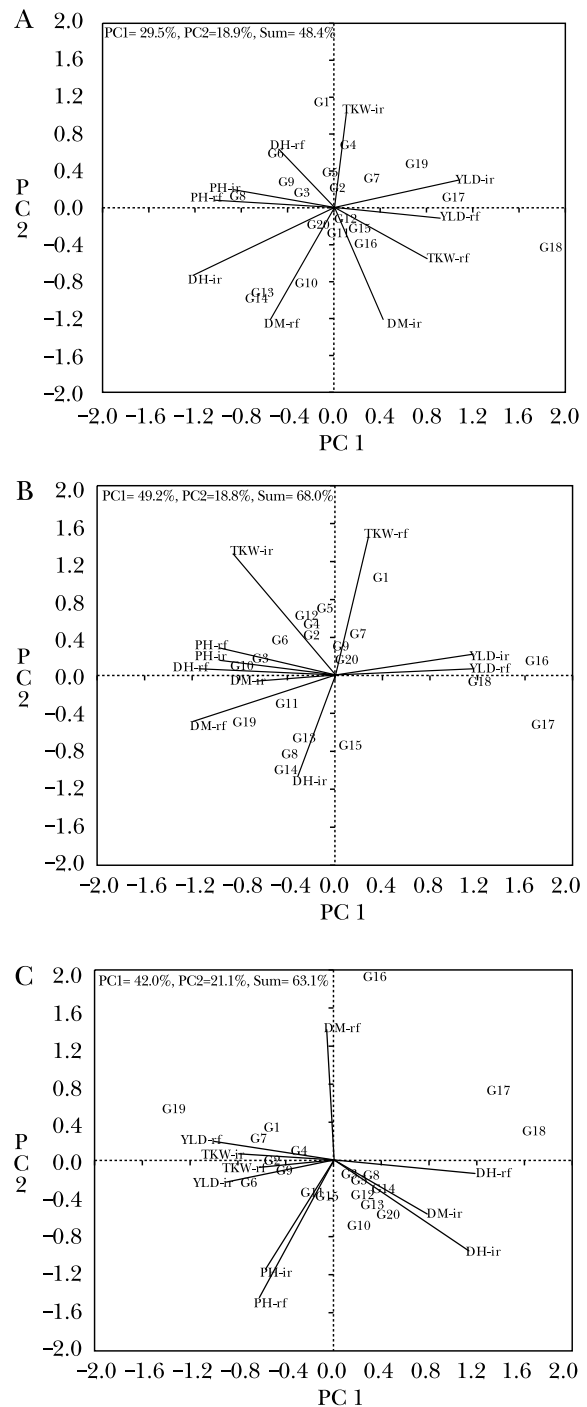

Fig. 2. Vector view of genotype×trait biplot summarizing the interrelationship among the traits studied under rainfed and irrigated conditions at (A) moderate (B) warm and (C) cold locations. YLD: grain yield; TKW: 1000-kernel weight; PH: plant height; DH: days to heading; DM: days to maturity; the suffix of rf and ir represent for rainfed and irrigated conditions, respectively; G1 to G20 are genotype codes.
G19＝Sardari, G20＝Zardak

and between yield and DM under both rainfed and irrigated conditions, as indicated by the right angle between their vectors. Correlation coefficients among these traits can further support these results and indicate

the value of the biplot in displaying the relationships among traits. However, an exact match is not to be expected, because the biplot describes the interrelationships among all traits on the basis of overall pattern of the data, whereas correlation coefficients only describe the relationship between two traits (Yan and Rajcan, 2002). The GT-biplot can be used to compare genotypes on the basis of multiple traits and to identify genotypes that are particularly good for certain trait(s). The GT-biplot also can be used to discriminate genotypes based on each trait (Yan and Rajcan, 2002). The best genotypes based on grain yield at both growing conditions were G17 followed by G18, while the G8 had the highest PH under both rainfed and irrigated conditions. The genotypes G4 and G1 had the highest TKW under irrigated conditions, while the G18 had the highest TKW under rainfed conditions. The genotypes G10, G13 and G14 tended to mature late. The genotypes near to origin of the biplot had an average performance based on multiple traits under both rainfed and irrigated conditions at moderate location.

(2) B: Warm Location

Fig. 2B is a GT-biplot showing the relationships among traits at warm location. From this biplot, the most prominent relations are: (i) strong positive associations between grain yield at both rainfed and irrigated conditions, between PH at both rainfed and irrigated conditions, and between DM under both rainfed and irrigated conditions; and (ii) negative correlations between grain yield with PH, DM and DH; between DH under irrigated conditions and TKW under both conditions. The three breeding lines (G16, G17 and G18) had the best performance based on grain yield under both rainfed and irrigated conditions, while G1 was the best with regard to TKW under rainfed conditions and G2, G4, G5 and G12 were superior for TKW under both rainfed and irrigated conditions. The genotypes G11 and G19 tend to mature late while the genotypes G16, G17 and G18 were early maturing.

(3) C: Cold location

Fig. 2C is a GT-biplot based on first two PC axes which explained 63.1% of the (G+GE) variation at the cold location. In the biplot, a strong positive association was found between PH at both rainfed and irrigated conditions, as found in case of the other two locations, indicating that the relationship between PH of tested genotypes are not affected by the environment effects (i.e., year, moisture regimes and locations with different climates), as already shown by ANOVA, where the G effect was greater than the GE effect. A positive correlation was also found between TKW and grain yield at both rainfed and irrigated conditions, indicating that TKW and yield are more associated at cold location than at the two other locations (moderate and warm). A negative correlation was found between DM under both rainfed and irrigated conditions, showing that the genotypes with late maturity under

rainfed conditions were not late maturing under irrigated conditions. This differed from the results obtained in the two other locations. The genotypes G16, G17 and G18 were late in maturity at the cold location, while these genotypes at moderate and warm locations were early maturing. The negative association observed between DM and PH at cold location was not found for the other two locations. The best genotypes based on grain yield and TKW at cold location were G1, G2, G6, G7, G9 and G19 whereas the best one based on PH were G11 and G15.

Discussion

We assessed the agronomic performance of 18 breeding lines in comparison with two checks, one durum wheat ("Zardak") and one bread wheat ("Sardari") under divergent environments in Iran where durum wheat is grown. The biplot analysis method enabled a visual comparison of the locations and genotypes, and their interrelationships. Based on each trait different genotypes emerged as winners in different locations, suggesting different mega-environments for each trait. On the basis of grain yield, the genotype G18 was the most suitable under the moderate environment of Iran, where this location made up a single mega-environment on the basis of yield potential, whereas the G17 was the best yielder at the warm location (second mega-environment) and the cold location made up the third mega-environment where the genotypes G6, G16 and G19 were the winners. The biplots have shown that the genotypes from ICARDA (G16, G17, G18) are more adapted to warm and moderate locations of Iran, while at cold location the best genotypes were G19 ("Sardari"), G2, G6, G7 and G9. These genotypes produced the highest yield at their locations, and then can be considered as having specific adaptation to their respective environments. The genotype G19 had the height TKW at the cold and moderate locations whereas at warm location the G5 and G12 were the best. The PH was not affected by the moister regime, where a close correlation was found between the responses of genotypes for this trait under both rainfed and irrigated conditions, although this trait discriminated genotypes at different locations. For instance, the G8 was the tallest genotype at the moderate location, while G3 and G10 at the warm location and G1 and G15 at the cold location had the highest PH. This indicated that the ranking of genotypes based on PH was predominantly affected by location. Similar results were also observed for other traits (DH, TKW and DM), indicating that the biplots correctly show the variation among traits and that they were able to discriminate genotypes in opposite directions. Similar reports on GT biplots (Yan and Kang, 2003; Peterson et al., 2005; Egesi et al., 2007; Fernandez-Aparicio et al., 2009) demonstrated that the GT biplot is an excellent tool for visualizing genotype-by-trait data and revealing the interrelationships among the crop traits. It also provides a

tool for visual comparison among genotypes on the basis of multiple traits (Yan and Kang, 2003). The GT-biplot can be used in independent culling based on multiple traits and in comparing selection strategies. The biplots displayed the patterns of variability of the genotypes, the locations, and their interactions. Interrelationships among agronomic characteristics allowed the identification of best genotypes for several traits. The development and release of high yielding genotypes with good agronomic attributes and adapted to different zones could allow the expansion of the areas grown to durum wheat in Iran. Supplemental or full irrigation can increase significantly grain yield of durum wheat mainly in warm and moderate cold winter areas. This study has allowed to refine durum wheat breeding strategy by identifying different target environments, the possible parental material and some desirable agronomic traits to be used during the selection process.

In this study, the durum wheat ("Zardak") and even the bread wheat ("Sardari") checks were outyielded by promising durum wheat lines. However, in the highlands of Iran, there is a need to develop better-adapted and higher-yielding cultivars to compete with bread wheat and increase the area devoted to durum wheat. In developing countries, such as Iran, cultivars have mostly been selected in favorable environments and then introduced with technological packages (e.g., mineral fertilizer, pesticides, irrigation) designed to significantly improve the growing environment. Fortunately, the results of less than 12 yr of breeding efforts jointly with between ICARDA and the Dryland Agricultural Research Institute, Iran, led to the release of stable durum genotypes with high-yielding ability in minimum inputs conditions.

Acknowledgment

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