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Grain yield stability analysis of maize (*Zea mays* L.) hybrids under different drought stress conditions using GGE biplot analysis

M. R. Shiri

Agricultural and Natural Resources Research Center of Ardebil Province, Moghan, Iran.
Author's E-mail address: mohammadrezashiri52@gmail.com

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ABSTRACT

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Drought stress is the most important environmental constraint contributing to grain yield instability of maize (*Zea mays* L.). Evaluation of maize genotypes under different stresses would be useful for identifying genotypes that combine stability with high yield potential for stress-prone areas. This study was conducted to estimate grain yield stability of maize hybrids and to identify hybrids that combine stability with high yield potential across stress and non-stress environments. Seven maize hybrids were tested in three consecutive growing seasons under four irrigation regimes (E1 = well-watered; E2 = water deficit at the vegetative growth stage; E3 = water deficit at flowering; E4 = water deficit at grain-filling) at Ultan Agricultural Research Station, Moghan, Iran. Combined analysis of variance showed that environments, genotype and genotype × environment (GGE) interaction effects were highly significant. Genotype and genotype × environment analysis using GGE biplot explained 94.7% of the total grain yield variation. The GGE biplot analysis ranked maize hybrids with above-average yield across growing seasons (SC704 > SC724 > SC703 ≈ SC720 > SC647) and grain yield stability (SC700 > TWC600 > SC724). According to the variation in maize hybrids as well as G × E interaction sources, hybrids SC704 and SC724 in environments E1, E2 and E4 as well as hybrid SC647 in E3 were the superior hybrids and had better specific adaptation. The hypothetical ideal genotype's biplot indicated that hybrid SC704 had higher grain yield and yield stability and was better adapted to all the test environments.

Keywords: drought stress, ideal genotype, specific adaptation, vegetative growth stage, yield stability

INTRODUCTION

Maize (*Zea mays* L.) is a versatile crop that adapts easily to a wide range of production environments (Gerpacio and Pingali, 2007). Maize is also the third most important crop in the world, after wheat and rice, in terms of growing area, production and grain yield (Shiri *et al.*, 2010).

Drought is the most important environmental constraint to maize grain yield stability. Grain yield stability is influenced by the capacity of a genotype to react to environmental conditions, which is determined by the genotype's genetic structure. Improved grain yield and stability in maize cultivars have been attributed to increased drought tolerance. Extensive testing of maize hybrids developed for drought prone conditions, under both severe and mild drought stress, as well as in optimal growing environments, would be useful for identifying hybrids that combine high grain yield potential and stability (Meseka *et al.*, 2008).

Genotype × environment (G×E) interaction alters the relative grain yield of genotypes in different environments and makes it difficult to select superior genotypes (Gauch, 2006; Cornelius and Crossa, 1999). Generally, different genotypes behave differently because of differences in gene responses or in their potential performance in different environments (Brandiej and Meverty, 1994). G×E interaction decreases the correlation between genotype and phenotype, which in turn reduces the progress of genotype selection, especially under drought stress conditions. Stability analysis is the most important method used to discover the nature of G × E interaction by which stable and consistent genotypes can be identified and selected (Cornelius and Crossa, 1999; Perkins and Jinks, 1971).

Different approaches (including single-variable, multivariate and non-parametric methods) have been suggested for evaluating G × E interaction and identifying stable genotypes (Becker and Leon, 1988; Karimizadeh *et al.*, 2006). Although employing and calculating non-parametric and single-variable parametric methods is easy, they do not perfectly interpret the multi-dimensional and complicated nature of G × E interaction. Therefore, multivariate analysis methods have been proposed to solve the problem (Moreno-Gonzalez *et al.*, 2004).

Among multivariate analysis methods, biplot methods are based on principal component analysis (Yan *et al.*, 2000; Kempton, 1984; Gauch and Zobel, 1997; Gabriel, 1971). Different versions of biplot methods based on multivariate statistics have been introduced and widely used in agricultural research by plant breeders for graphical analysis of G × E interaction (Yan and Tinker, 2006; Gauch, 2006; Yan *et al.*, 2000).

GGE biplot is a specific version of a biplot that provides information on genotype main effects and G × E interaction at the same time. In contrast to typical multivariate stability analysis methods where only G × E interaction is considered, this method includes genotype main effects as well. Various studies have shown that in most stability analysis experiments, the main effect of environment is high, while variations determined by the main effect of genotype and G × E interaction that are recommendable and interpretable are low. Since the environment is not a controllable factor, in the GGE biplot method, genotypic and G × E interaction sources of variation are used to obtain more reliable results (Yan *et al.*, 2000, 2001).

Because it graphically displays G × E interaction effects, the GGE biplot method helps plant breeders to easily assess genotypic stability and combinations of genotypic stability and yield in different environments. It also allows assessing the relationship between environments and facilitates the re-arrangement of target environments in plant breeding programs. This method has been applied for G × E interaction analysis to evaluate genotypes in multi-environment trials of wheat (Yan and Hunt, 2002; Yan *et al.*, 2001), maize (Choukan, 2011; Fan *et al.*, 2007), soybean (Yan and Rajcan, 2002), barley (Dehghani *et al.*, 2006), cotton (Dimitrios *et al.*, 2008; Blanche and Myers, 2006) and durum wheat (Mohammadi *et al.*, 2010).

Determining and grouping of target environments in plant breeding programs is one of the most important applications of the GGE biplot method. The environments being evaluated are grouped into different groups with the same genotype reaction. Environment grouping using GGE biplot analysis has been reported for different crops such as wheat (Yan and Tinker, 2006; Kaya *et al.*, 2006), durum wheat (Mohammadi *et al.*, 2010; 2012; Letta *et al.*, 2008), barley (Mohammadi *et al.*, 2009), soybean (Yan and Rajcan, 2002), rice (Samonte *et al.*, 2005) and maize (Choukan, 2011).

Genotype × environment interactions are common under drought and make breeding progress difficult. They may originate from environmental variation in the timing and severity of moisture stress and genetic variation in flowering time (Bänziger and Cooper, 2001). Therefore, plant breeders repeat experiments in different locations and years (environments) to assess environmental effects and obtain more reliable results.

The purpose of this study was to analyze G × E interaction using the GGE biplot method to evaluate maize hybrids, environments, and the relationships between hybrids and environments, as well as to identify ideal hybrids and recommend adapted hybrid(s) that are suitable for different water-stressed environments in the Moghan region of northwestern Iran.

MATERIALS AND METHODS

This research was carried out at the Agricultural and Natural Resources Research Center of Ardebil Province (Moghan) in northwestern Iran (39° 41′ N and 47° 32′ E; altitude: 45-50 masl). According to the Pars Abad, Moghan, synoptic station, it is a semi-arid region with mild winters and hot summers, with a maximum temperature of 31.4 °C in August and a minimum temperature of 1.4 °C in January. The average annual precipitation is 389.5 mm.

Seven maize hybrids (SC704, SC703, SC700, SC720, SC647, SC724 and TWC600) were evaluated under four irrigation regimes (in four separate trials) using a randomized complete block design with three replications in three consecutive growing seasons. The four irrigation regimes included: E1 = well-watered (irrigation based on the crop's water requirement and farmers' practice in the region); E2 = water deficit at the vegetative growth stage (no irrigation from post-emergence to tasseling, and continuing irrigation to physiological maturity); E3 = water deficit during flowering (no irrigation from tasseling to the end of pollination; irrigation is resumed pre and post flowering); E4 = water deficit during grain-filling (irrigation to the end of pollination; after that, no irrigation until physiological maturity).

The first irrigation was applied to all trial plots to ensure uniform germination, emergence and establishment. The inlet and outlet water mass was measured by flume in order to determine the amount of water consumed. Each plot consisted of four rows, 5.76 m long, with 75-cm row spacing and 18-cm intra-row distance between plants. When plants reached the 4- to 5-leaf stage, the plots were thinned to obtain a final density of 75000 plants ha^{-1}. Grain yield was measured and recorded from the two middle rows in each plot after removing 25 cm from both ends of each row. Grain yield (t ha^{-1}) for each hybrid was adjusted to 14% grain moisture content.

Prior to the combined analysis of variance, Bartlet's test of homogeneity of variances and the normality test for data were performed using Minitab software (version 14.0). Combined analysis of variance (ANOVA) was performed to invesigate the effects of genotype (G), environment (E) and genotype × environment (G × E) interaction using SAS software (version 9.0).

Genotype and genotype × environment (GGE) biplot analyses were conducted using GGE biplot software (Yan *et al.*, 2000; Yan and Kang, 2003) to assess grain yield stability and identify superior genotypes. GGE biplot analysis was also used to generate graphs for:

(i) comparing environments to the ideal environment;
(ii) the "which-won-where" pattern;
(iii) environment vectors.

The angles between environment vectors were used to judge correlations (similarities/dissimilarities) between pairs of environments (Yan and Kang, 2003).

RESULTS AND DISCUSSIONS

The combined analysis of variance for grain yield revealed highly significant effects of environment, genotypes and G × E interaction (Table 1). Results indicated variation in the grain yield performance of different hybrids in different environments and suggested that it would be more appropriate to select superior maize hybrids based on a combination of high average grain yield and good yield stability than on average grain yield alone. A large proportion (71.33%) of total variation in grain yield was caused by the environment. Large sum of squares for environment indicated that the environments were diverse, and that large differences among environmental means caused most of the variation in grain yield.

Genotype × environment interaction accounted for 9.24% of the total variation in grain yield, while genotype accounted for only 2.37% (Table 1). The G × E interaction sum of squares was 3.9 times larger than that for genotypes, indicating that there were substantial differences in hybrid responses across environments.

Table 1. Combined analysis of variance for grain yield of maize hybrids.

S.O.V.	df	SS	MS	% of total
Environment (E)	11	1239.368	112.670**	71.33
Replication/E	24	163.354	6.806	9.40
Genotype (G)	6	41.265	6.877**	2.37
G × E	66	160.624	2.434**	9.24
Error	144	133.012	0.924	7.65

** Significant at the 1% probability level.

Although G × E interaction was highly significant, the combined analysis of variance could not justify hybrid grain yield stability. Therefore, G × E interaction effects should be evaluated using suitable statistical methods to identify stable hybrids. The graphical GGE biplot method was employed to investigate environmental variation and interpret G × E interaction.

To draw the biplot, data obtained from the multivariate models of hybrids and environments should be used simultaneously in one figure. This GGE biplot is shown to effectively identify the GEI pattern of the data. A GGE biplot is 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 or environment-standardized G × E data (for a single trait) (Yan *et al.*, 2001; Mohammadi *et al.*, 2010).

GGE biplot analysis was also performed on the 3-year average grain yield of maize hybrids under four irrigation regimes (as environments). Results showed that the GGE biplot explained 94.7% of genotype main effects and the G × E interaction. The primary (PC1) and secondary (PC2) components explained 59.8 and 34.3% of genotype main effects and G × E interaction, respectively (Fig. 1).

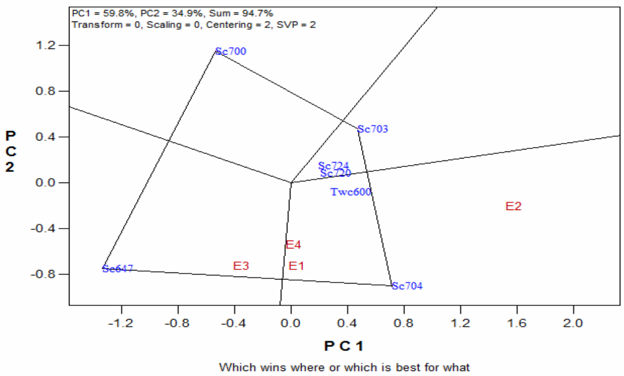

Fig. 1. Polygons of GGE biplot method for grouping environments.

The GGE biplot is also used to draw the polygon for G × E interaction effect from which different interpretations can be derived. The polygon is formed by connecting the markers of the genotypes that are farther away from the biplot origin such that all other genotypes are contained in the polygon. The polygon view of a biplot is the best way to visualize the patterns of interaction between genotypes and environments, and to effectively interpret a biplot. It clearly shows which genotype won in which environments, thus facilitating mega-environment identification (Yan *et al.*, 2000; Dimitrios *et al.*, 2008).

The polygon of the seven maize hybrids under four irrigation regimes is shown in Fig. 1. Other researchers have also used this method (Choukan, 2011; Sabaghnia *et al.*, 2008; Yan *et al.*, 2000). In Fig. 1, hybrids SC704, SC703, SC647 and SC700 are located at the top of the polygon. These hybrids are the strongest or weakest hybrids in terms of grain yield in some or all environments, as they are located at the maximum distance from the biplot center. In E1 (well-watered), E2 (water deficit at the vegetative growth stage) and E4 (water deficit at grain-filling), SC704 and SC703 had the highest grain yield and were considered the superior hybrids in the test environments. Of course, hybrids SC724, SC720 and TWC600 were not significantly different from these hybrids. In E3 (water deficit during flowering), SC647 had the highest grain yield. Although hybrid SC700 was located at the top of the polygon, it produced low grain yield in all test environments.

Grain yield performance and stability were evaluated using an average environment coordination (AEC) method (Yan et al., 2001; Yan and Hunt, 2002). In this method, an average environment, represented by a small circle, is defined by the average PC1 and PC2 scores of all environments. A line is then drawn that passes through this average environment and the biplot origin; this line is called the average environment axis and points to higher average grain yield. The line that passes through the origin and is perpendicular to the AEC (average environment coordination) with double arrows represents the stability of genotypes. A line that passes in either direction away from the biplot origin, on this axis, indicates greater G × E interaction and reduced grain yield stability (Yan and Hunt, 2002). In the present study, an average tester coordinate curve (Fig. 2) was drawn based on the mean grain yield values in three growing seasons to evaluate hybrid yield and stability.

According to Fig. 2, hybrids TWC600, SC720 and SC724 had average grain yield and high stability, whereas hybrid SC704 had high grain yield and average stability. Hybrid SC700 had low grain yield and high stability, while SC647 had average grain yield and very low stability. Generally, it seems that hybrid SC704 with acceptable stability and good grain yield is the best hybrid (Fig. 2).

The GGE biplot analysis allows comparing the test genotypes to a reference genotype. This method specifies the position of an ‘‘ideal’’ genotype that

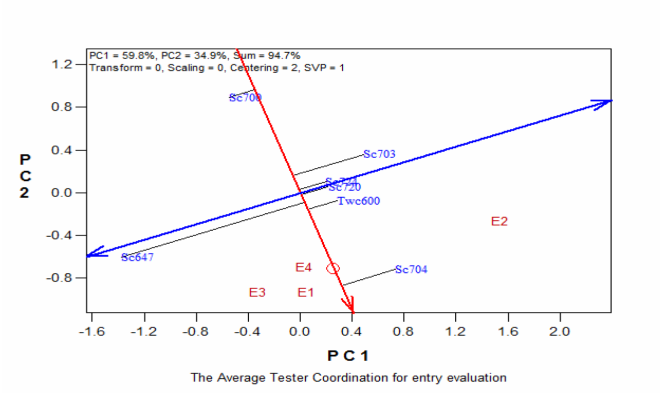

Fig. 2. Evalution of seven maize hybrids based on both yield and stability performance in different environments.

has the highest average value of all genotypes and is absolutely stable, i.e., it expresses no G × E interaction. The hypothetical ideal genotype, however, is determined based on the most stable genotype with the maximum grain yield. This genotype is determined as the genotype with the maximum length on the average vector of higher yielding genotypes, and play minimum in the G × E interaction phenomenon.

In Fig. 3, the hypothetical ideal genotype is shown as a small circle on the axis of average genotype yield. To use the ideal genotype as the measurement center, concentric circles were drawn in the biplot to graphically determine the distance between the test genotypes and the ideal one (Fig. 3). A genotype that is located at the center of the circles or is the genotype closest to the hypothetical genotype is considered a superior genotype with high grain yield and good yield stability. Hybrid SC704 was closest to the hypothetical ideal genotype and therefore identified as the best hybrid, while hybrid SC700 was extremely far away from it and thus not in the ideal hybrid category.

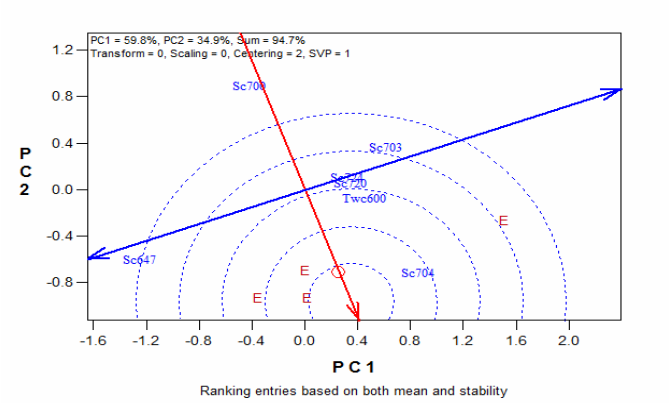

Fig. 3. GGE-biplot for comparing the test hybrids with the ideal genotype.

Results of the graphical analysis of G × E interaction effects revealed that environments justified the large proportion of observed variations in the G × E interaction matrix. The GGE biplot method was used successfully for grouping different environments in this study, as well as identifying stable genotypes with good adaptation to different environmental conditions.

Evaluation of correlation coefficients between test environments could clarify the relationships among environments and enlighten future planning and experiments. In the event of a strong positive correlation between two or more environments, the considered experiments could be conducted in one environment and the results obtained generalized to the others (Yan and Kang, 2003). In the graph drawn for this purpose, the cosine of the angle between environment vectors stands for correlation intensity. If it is null, the correlation between them is +1. On the other hand, cosine 90° stands for a null correlation, while cosine 180° stands for a correlation of -1.

The vectors of E1 and E4 created a very small angle and their correlation was close to +1, which implies that these environments had a strong positive correlation with each other (Fig. 4). Therefore, the results of environment E1 (well-watered) could be applied in the case of environment E4 (no irrigation during grain-filling) with more reliability. On the other hand, the correlation between E3 (water deficit during flowering) and E2 (water deficit at the vegetative growth stage), as well as the correlation of E1 (well-watered) and E4 (water deficit during grain-filling) with E2 (water deficit at the vegetative growth stage) were almost null. This implies that the reactions of the test hybrids in environments E3 and E2 were not the same. However, the reactions of the hybrids in E2 were not the same as those in E1 and E4. Therefore, the results of environments were independent and could not be generalized.

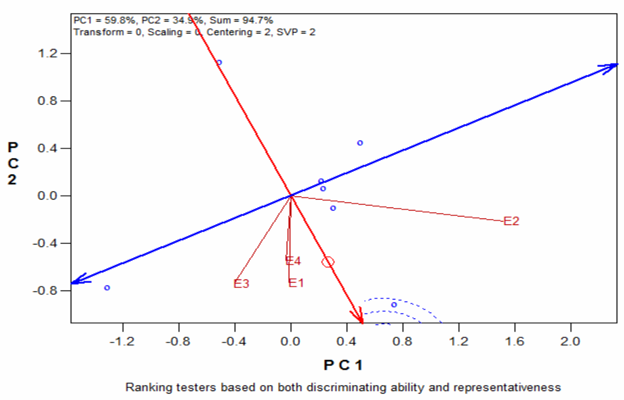

Fig. 4. GGE biplot for comparing the test environments to the ideal environment based on grain yield and yield stability of maize hybrids and the relationships among environments.

CONCLUSION

Employing multivariate analysis methods for evaluating of G × E interaction effects is a strong and useful approach, as it effectively analyzes the complicated and multi-dimensional nature of G × E interaction. The GGE biplot method facilitates the interpretation of results by using multivariate analysis methods, two-dimensional graphs and appropriate data analysis. For this reason, it is highly recommendable as a suitable method for yield stability analysis.

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