THE ANALYSIS OF GENOTYPE × ENVIRONMENT INTERACTION USING RAPESEED (*BRASSICA NAPUS L.*) BY GGE BIPLOT METHOD

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ABSTRACT
This study was aimed to asses seed yield performances of 16 rapeseed genotypes in randomized complete block designs (RCBD) with three replications at four Agricultural Research Stations of cold and mid-cold regions over two years in Iran (2015-2017). GGE biplot analysis indicated that the first two components explained 83% of seed yield variations. Genotype, location and their interaction explained 18%, 52% and 30%of the total GE variation, respectively. In this research, a graphically represented GGE biplot analysis enabled selection of stable and high-yielding genotypes for all investigated locations, as well as genotypes with specific adaptability. The GGE biplot analysis was adequate in explaining GE interaction for seed yield in rapeseed. It can be concluded that genotypes G2, G4 and G13 had the highest mean seed yield and stability in four investigated locations. For specific adaptability, G13 was recommended for Isfahan, Karaj and Kermanshah and G4 for Mashhad.

Keywords: Mid- Cold, GGE Bi-Plot, Performance, Specific Adaptability, Stability

مجلة العلوم الزراعية العراقية -2020 :51 (5):1337-1349 مطهري وآخرون

تحليل التداخل لجيني × البيئة في السلجم (*Brassica napus* L.) باستعمال طريقة GGE Biplot

علي رضا مطهري اسلام مجيدي هرفان بهرام علي زاده ومحمود خسروشلي

المستخلص

تم فحص أداء إنتاجية الحبوب لـ 16 نمطًا وراثيًا من بذور السلجم حسب تصميم القطاعات الكاملة العشوائية (RCBD) مع ثلاثة مكررات في أربع محطات للبحوث الزراعية في المناطق الباردة والمتوسطة على مدى عامين في إيران (2015–2017). أشار تحليل GGE biplot إلى أن المكونين الأولين أوضحا 83٪ من تغيرات محصول البذور. أوضح التركيب الوراثي والموقع وتفاعلهم 18 ٪ و 52 ٪ و 30 ٪ من إجمالي تباين GE ، على التوالي. في هذا البحث ، أتاح تحليل GGE biplot الممثّل بيانياً اختيار الأنماط الجينية المستقرة وذات العائد المرتفع لجميع المواقع التي تم فحصها ، فضلاً عن الأنماط الجينية ذات القدرة على التكيف المحددة. كان تحليل GGE biplot مناسبًا في شرح تفاعل GE لبذور السلجم. يمكن الاستنتاج أن الطرز الوراثية G2 و G4 و G13 كان لها أعلى متوسط إنتاجية وثبات في أربعة مواقع تم فحصها. من أجل التكيف المحدد ، تمت التوصية بـ G13 لأصفهان وكرج وكرمانشاه و G4 في مشهد

الكلمات المفتاحية: تفاعل جنرال إلكتريك ، مؤامرة GGE ثنائية ، بذور اللفت ، القدرة على التكيف المحدد ، الاستقرار.

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INTRODUCTION

Rapeseed (*Brassica napus* L.), which contains more than 40 % oil content, is one of the important oil crops after soybean in the world and has an excellent source of protein with its meal (17,30). About 90% of the requirements for vegetable oils in Iran are provided by imports (44) costing huge amounts of foreign exchange. Local production of edible oil is hardly enough to meet 10% of the total requirement. There is a strong need for domestic production to increase vegetable oil production by increasing the production area of oilseed crops among which rapeseed is of great importance and potential (4). According to FAO statistics, rapeseed with 75431 ha harvested area, 1836 kg/ha yield and 138489 tons production in 2016 crop year has recently been exploited to raise oil production in Iran (6). Stability of cultivars in different environmental conditions is very important in crop breeding programs. Because of the different response of the cultivars to environmental changes, their performance fluctuates from one environment to another. Therefore, genotype × environment interaction is a challengeable issue among plant breeders (34). Generally, each genotype has the maximum production potential in a given environment but by assessing the compatibility and stability of cultivars, it is possible to identify genotypes in different environments that have acceptable performance in all environments (31). Stable genotypes have the same reaction in different environments (15,29). Therefore, identification of such genotypes by various stability parameters is one of the important goals in breeding crops. Development of hybrids and varieties with high and stable genetic potential for seed and oil yield are the main goals in rapeseed breeding (20). Yield performance as a complex trait usually is affected by the environmental effect (E), genotypic effect (G) and their interaction (GEI). Although more than 80 % of the total variance caused by environment, genotype and genotype × environment interaction cause 10 % of each variation (39). Up to now, different methods introduced for stability analysis in crop plants by various researchers. In multi-environmental trials (METs), there are usually a large number of genotypes tested in number of locations and years, Therefore, it is difficult to determine genotype × environment response patterns without the need for graphical illustration of data (41). GGE biplot is a novel multivariate analysis method and a user-friendly graphical tool for analysis of two-way data to evaluate genotype × environment interaction and stability analysis (39). This method is recently used for stability analysis in different crops such as sunflower (25), wheat (9), barley (36,38), rapeseed (3), maize (23) and sugar beet (10). In rapeseed, many studies have been done on the stability of the crop (2,5,11,20,21,24,27,43). The rapeseed breeding program and the study of the stability and compatibility of cultivars in Iran are carried out in two warm and cold regions of the north and south of the country. 1) Spring cultivars for arid and warm-humid regions and 2) winter cultivars for cold and mid-cold regions (2). The objectives of this study were to evaluate genotype × environment interaction and stability of 16 winter rapeseed genotypes across four locations in Iran using GGE biplot model and to find high yield and favorable genotype(s) with specific adaptability within the investigated test locations.

MATERIALS AND METHODS

Plant materials and experimental designs

Seventeen genotypes consisting ten winter rapeseed obtained from foreign institutes with six Iranian lines and Okapi as check variety were arranged in randomized complete block design (RCBD) with three replications at four Agricultural Research Stations of cold and mid-cold regions of Iran during two years (2015-17). Geographical characteristics of experimental sites and characteristics of the genotypes were show in Tables 1 and 2. Standard practices were followed to raise a good crop in each location. Harvest of each genotype was done by hand at physiological maturity stage after removal of marginal effects and then seed yield was measured.

Table 1. Geographical characteristic of the experimental locations

Locations	Altitude (m)	Longitude	Latitude	Rainfall (mm)	Soil texture
Isfahan (ISF)	1612	51.26	36.32	125	loam
Mashhad (MAS)	1050	59.15	35.43	212	loam
Kermanshah (KER)	1346	47.26	34.08	538	loam
Karaj (KAR)	1300	57.50	35.48	250	Clay loam

Table 2. Description of winter rapeseed genotypes used in the experiment

Genotypes code	Genotypes Name	Type	Origin
G1	Zorica	Hybrid	Serbia
G2	Zlatna	Hybrid	Serbia
G3	ES Hydromel	Hybrid	France
G4	ES Alonso	Hybrid	France
G5	ES Darko	Hybrid	France
G6	ES Lauren	Hybrid	France
G7	ES Kamilo	Hybrid	France
G8	ES Mercure	Hybrid	France
G9	ES Artist	Hybrid	France
G10	HL3721	Open pollinated	Iran
G11	Ahmadi	Open pollinated	Iran
G12 (Check)	Okapi	Open pollinated	France
G13	Nafis (L72)	Open pollinated	Iran
G14	HL2012	Open pollinated	Iran
G15	Nima (SW102)	Open pollinated	Iran
G16	L1192	Open pollinated	Iran

Statistical analysis

Outlier detection and normality test of data were done before variance analysis using Grubbs' test and Shapiro-Wilk test by Statgraphics software (33). Also, Bartlett's test for homogeneity of variances is used to test that variances are equal across all location ($x^2 = 10.38^{ns}$). Then data were subjected to combined analysis of variance (ANOVA) using SAS software (28) and treatment means were compared by LSD test. Genotype × environment interaction and GGE Biplot analysis was performed using GGE biplot software (41). Based on singular value decomposition (SVD) of tester-centered data of the first two principal components Model I of Yan and Kang (39) was chosen to construct various biplots. This model is used for dataset in which all testers have the same unit, such as a genotype × environment table of a single trait like seed yield in this research.

The model is: $Y_{ij} - u - \beta_j = g_{i1}\, e_{1j} + g_{i2}\, e_{2j} + \varepsilon_{ij}$

RESULTS AND DISCUSSION

Analysis of variance (ANOVA)

Combined analysis of variance showed significant effects for genotypes, environments and their interactions. Therefore, investigated genotypes had different response to the environments. Mean comparisons indicated significant differences between check variety and investigated genotypes for seed yield. Genotypes G4 and G2 had significant yield increase at 5% compare to the check variety, while genotypes G10 and G6 had significant yield decrease at 1% compare to the check variety (Table 3). Sohrabi *et al.*, in grouping of 12 promising winter rapeseedlines in cold regions of Iran using cluster analysis (19) showed that there were significant year×location and year×location×genotype effects Similar results were reported by Tahira *et al.*, (35) and Mortazavian and Azizinia (22).

Table 3. Biennial mean comparison for seed yield (kg/ha) and PCA-scaled entry scores of winter rapeseed genotypes in different locations

Genotype code	Yield (kg/ha)	PC1	PC2
G1	3735 C	318.742	148.464
G2	4175 B	-841.135	-364.755
G3	3925 C	-150.901	364.381
G4	4200 B	-783.516	-741.150
G5	3716 C	132.276	301.328
G6	3427 F	781.800	538.096
G7	4107 C	-494.486	-48.775
G8	3879 C	-107.027	-62.334
G9	3653 C	316.056	495.674
G10	3501 E	730.216	-414.859
G11	3800 C	165.248	25.229
G12 (Check)	3820 C	-153.696	-70.496
G13	4103 C	-769.386	765.646
G14	3803 C	27.2	127.163
G15	3616 C	791.802	-814.761
G16	3981 C	36.808	-248.852
LSD 5%=295.32			

GGE biplot analysis

GGE biplot analysis of rapeseed genotypes showed that the first two components explained 83% of seed yield variations. Out of which, 49% belonged to the first component and 34% to the second component. Based on GGE biplot analysis, total GGE variance explained by entry (genotype), tester (location) and their interaction was 18%, 52% and 30%, respectively. A significant seed yield variation explained by environment (52 %) revealed that locations tested in this research were diverse, with great differences for environmental or tester effects causing the most variation in seed yields of investigated rapeseed genotypes. Yan (42) states that the environment determines a larger portion in explaining the total variance compare to G and GE. Similar results were reported in sunflower (25) and rapeseed (26).The GGE biplot and which-won-where pattern based on seed yield data of 16 rapeseed genotypes was show in Fig.1. It was based on model 1, in which the singular value is entirely partitioned into entries so that the entries have the unit of the original data. This model is preferred when the focus is to compare entries (genotypes). Polygon view of the GGE biplot, showing which genotype yielded best in which locations (39).Based on entry-focused biplot, G4 and G2 were best genotypes for Mashahd location and G13 was best for Karaj, Kermanshah and Isfahan locations (Fig. 1).

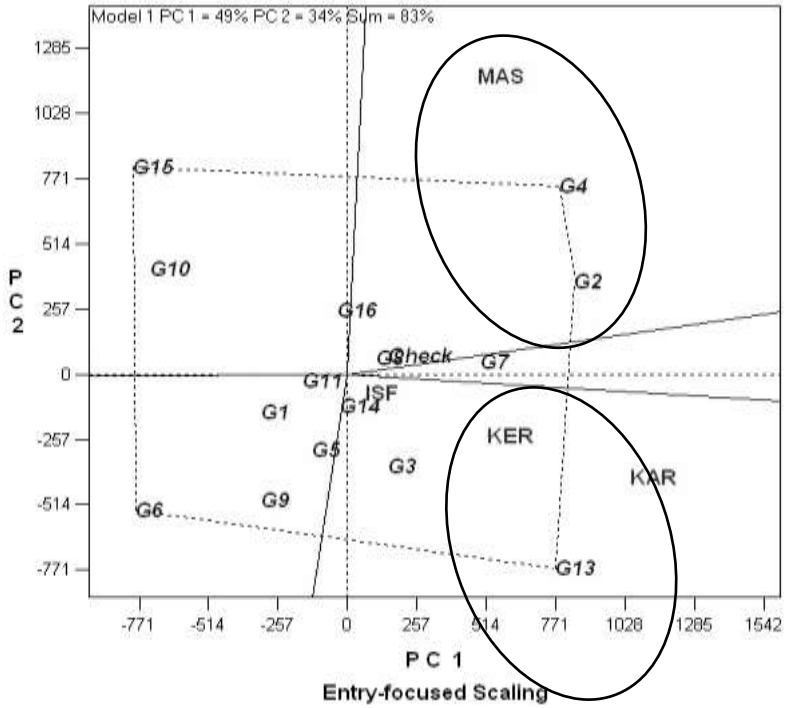

Fig. 1. GGE biplot and which-won-where pattern based on seed yield data of 16 rapeseed genotypes

Average tester coordination (ATC) is a very important function, which defines an average tester and draws it on the biplot. This biplot indicates the mean yield and stability of genotypes together. The parallel lines help rank the genotypes based on mean yield. The arrow of the ATC-abscissa points toward the direction of increasing mean yield, and the arrow of the ATC-ordinate point to greater GE interaction or instability (39). Based on ATC biplot, genotypes G2, G4 and G13 with the highest mean seed yield, which are close to ATC ordinate may be given here as stable genotypes across four investigated locations (Fig. 2). High correlation between entry projections onto ATC axis and entry main effects was calculated 0.97, which means a good approximation of the genotype main effects. This result was in accordance with findings Kang (14) and Yan and Kang (39).

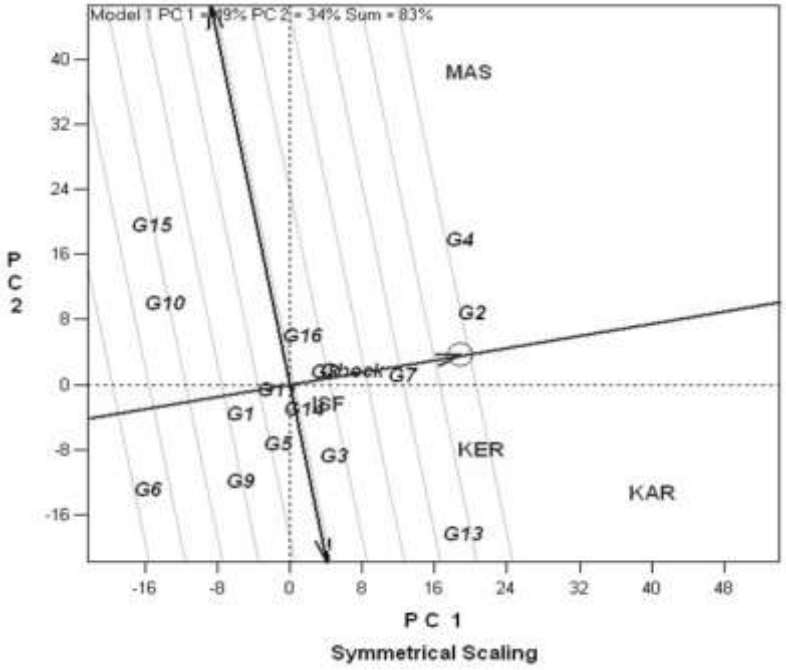

Fig.2. Average tester coordination (ATC) view of the GGE biplot based on the entry-focused scaling

Comparison of all investigated locations (testers) with ideal tester as the most discriminating and absolutely representative tester is represented in Fig 3. Accordingly, Karaj and Kerman were the most representative, whereas Mashahd and Isfahan were the least representative of the average environment.

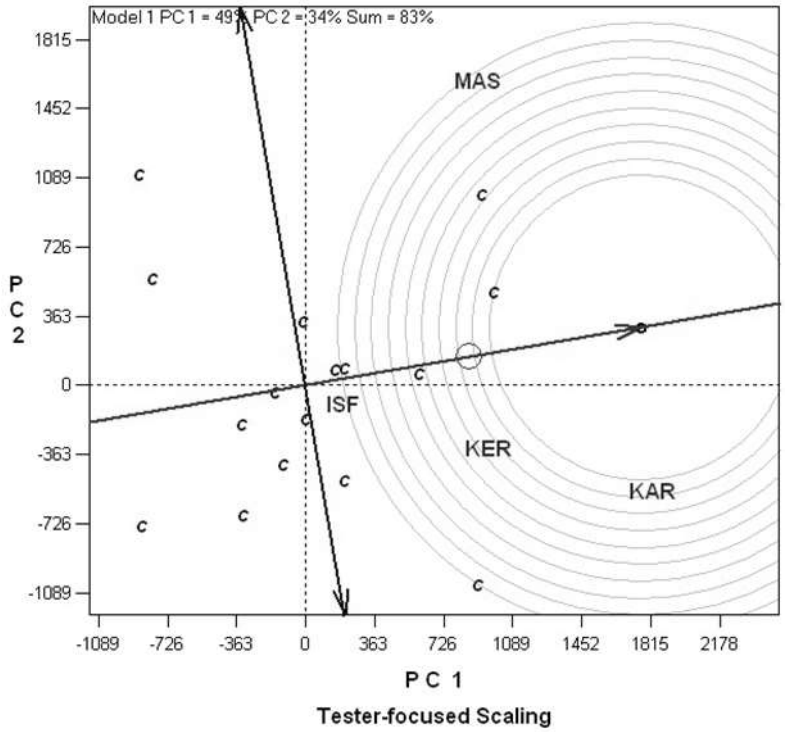

Fig. 3. Comparison of four investigated locations with ideal tester by tester-focused scaling using GGE biplot

The high distinguishing ability is an important criterion for assessing the test environment. The test environment lacking the distinguishing ability does not provide any kind of information about the genotypes and, thus, the test environment is ineffective. Another criterion, which is similar to the ability to distinguishing ability, is the representativeness of the test environment as an example of the target environment. This criterion is difficult to measure, and therefore, an average environment should be defined and use it as a reference. An ideal test location (environment) should be both discriminating and representative (39). In the other words, by using GGE biplot based on tester-focused scaling and ideal test environment, desirable environments (Karaj and Kerman) are used to identify superior genotypes, and unsustainable environments (Mashahd and Isfahan) can be used to eliminate unstable genotypes. Comparison of all investigated genotypes with ideal entry (genotype) by entry-focused scaling is show in Fig. 4. An ideal genotype in this biplot defined by a small circle with the arrow mark specified as the highest-yielding genotype in all environments. There is no such ideal genotype in reality. Other genotypes are ranked based on their distance from this ideal genotype. Genotypes G2, G7 and G4 were ranked highest mean yield and stable in all locations compare to the others.

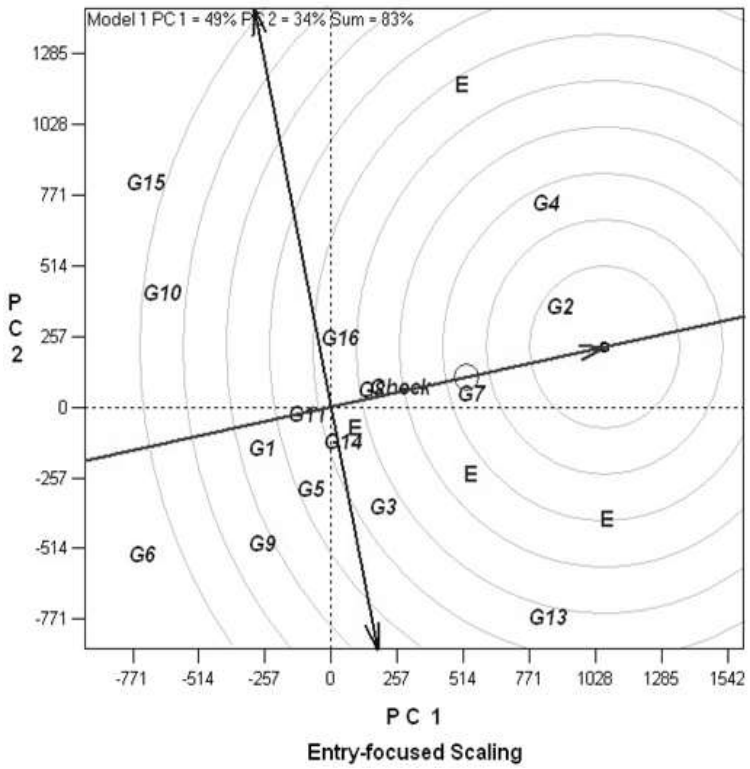

Fig. 4. Comparison of all investigated genotypes with ideal entry (genotype) by entry-focused scaling using GGE biplot

The vector length (Fig. 5) shows the ability to distinguish genotypes in that environment (39). Therefore, Karaj and Kerman locations were determined as discriminant environments. Kroonenberg (18) concluded that the cosine of the angles between the two environments shows the estimated correlation coefficient between them. This is confirmed by the correlation coefficients in Table 4. Karaj and Kerman locations had highest significant correlation coefficient (0.610) compare to the others. Isfahan location with smallest vector length showed least discriminating ability and determined as undesirable location for selection of superior genotypes.

Table 4. Correlation coefficients of the experimental locations by entry-focused scaling using GGE biplot

Locations	ISF	MAS	KER	KAR
ISF	**1**			
MAS	**-0.035^{ns}**	**1**		
KER	**0.122^{ns}**	**0.067^{ns}**	**1**	
KAR	**0.128^{ns}**	**0.133^{ns}**	**0.610****	**1**

ns: Non significant, **: Significant at 1% level

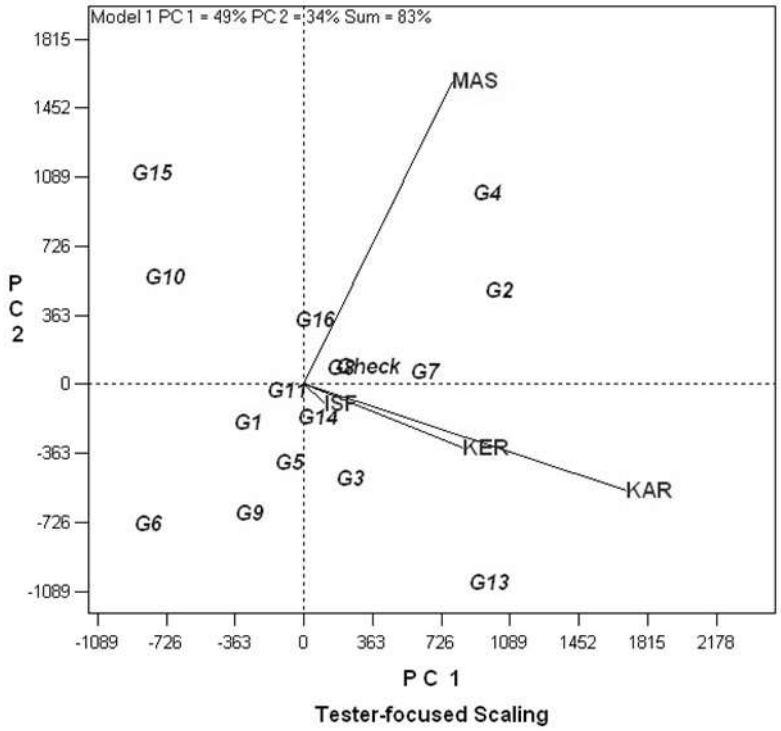

Fig. 5. Interrelationship among investigated locations by entry-focused scaling using GGE biplot

Specific adaptability

Figures 6-9 shows performance of all genotypes in a specific location. Specific adaptability is one of the favorites of breeders to introduce a most adapted and stable genotype to a given location. This is easily possible using GGE biplot. Based on entry-focused scaling biplots, genotype G13 was the best for Isfahan, Karaj and Kerman locations (Fig. 6, 7 and 8), while genotype G4 was the best for Mashhad location (Fig. 9). These results have already been confirmed in Fig. 1 by which-won-where pattern.

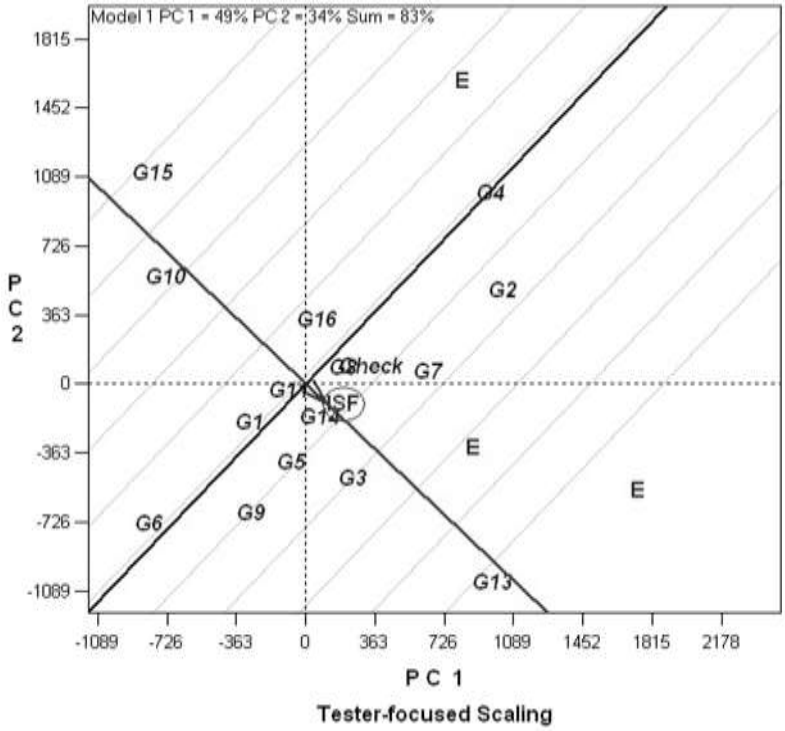

Fig. 6. Comparison of all genotypes' performance in specific location (Isfahan) by entry-focused scaling using GGE biplot

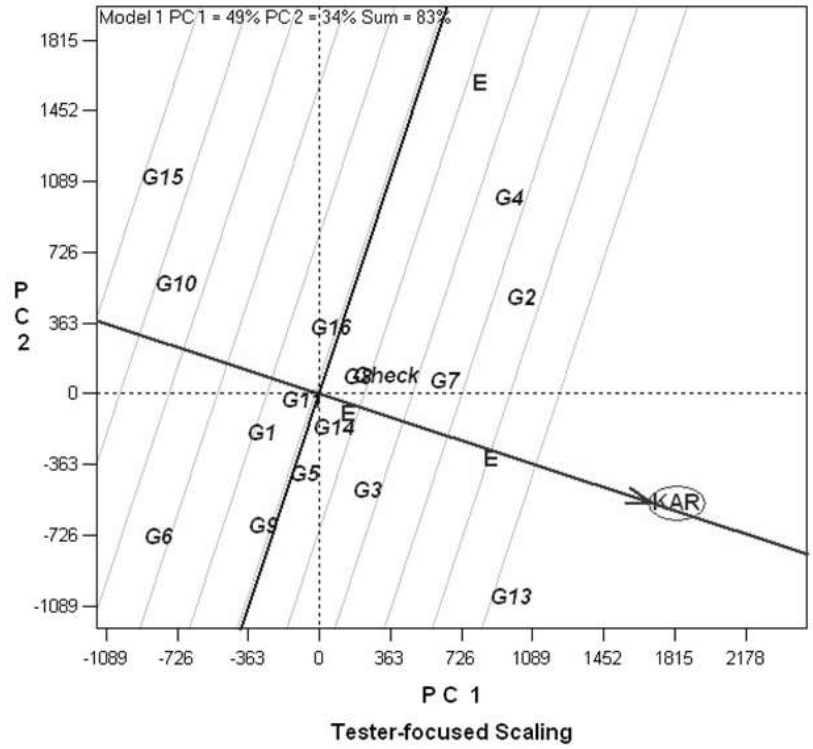

Fig. 7. Comparison of all genotypes' performance in specific location (Karaj) by entry-focused scaling using GGE biplot

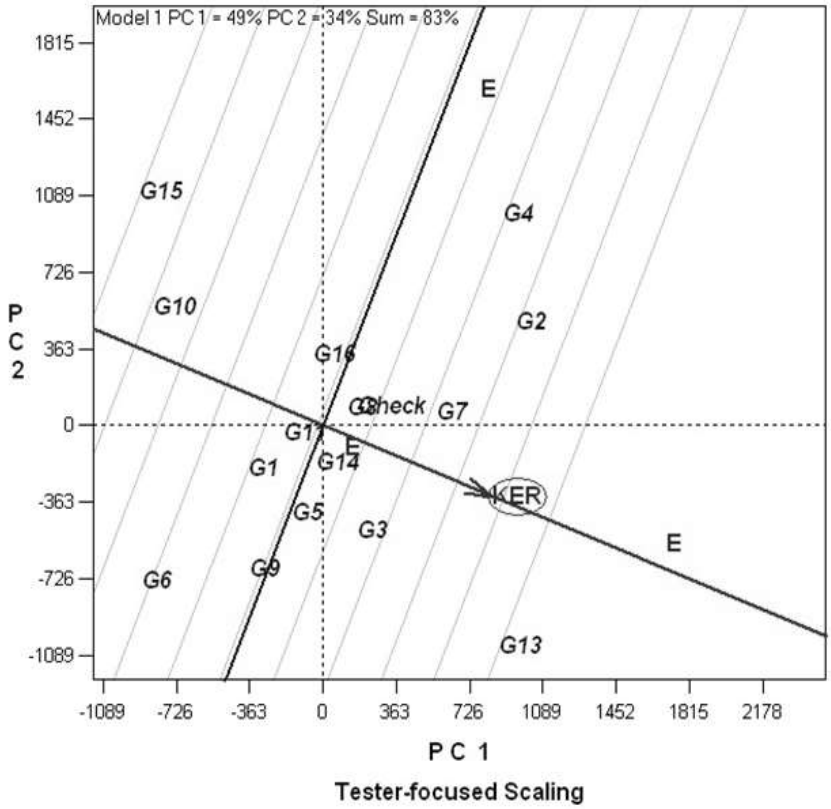

Fig. 8. Comparison of all genotypes' performance in specific location (Kerman) by entry-focused scaling using GGE biplot

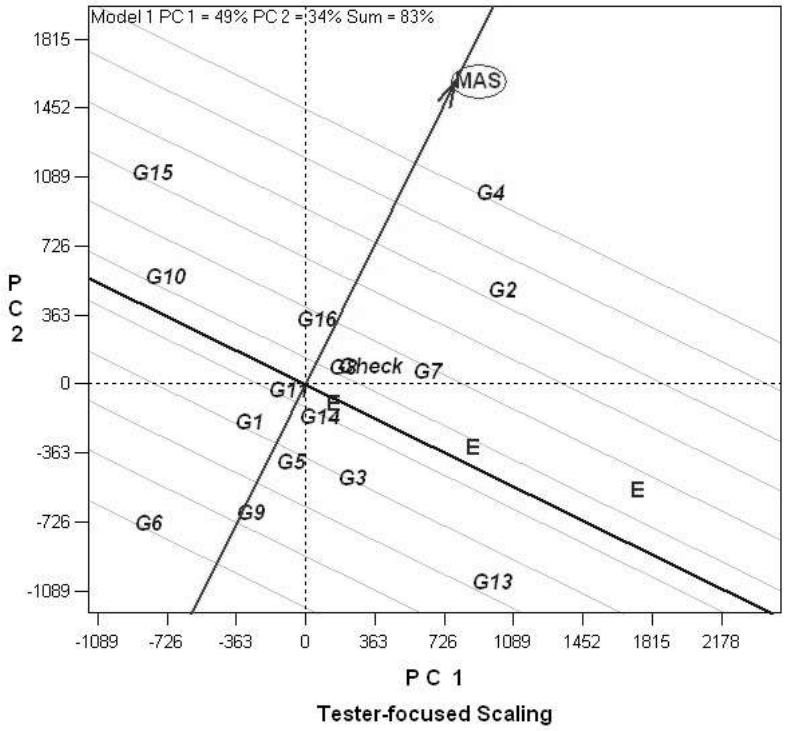

Fig. 9. Comparison of all genotypes' performance in specific location (Mashhad) by entry-focused scaling using GGE biplot

Investigation of genotype × environment interaction and selection of best and stable genotypes is one of the most important stages of breeding programs (13).Successful breeding programs focus on high yeild performance at the initial stage under non-stress conditions (26).In most breeding programs, despite the exact trials and effective selections, genotype×environment interaction causes instability performance of genotypes in different environments, so, improvement of quantitative traits such as seed yield remains a serious problem for breeders (8,40). Evaluation of genotypes in multi-environmental trials plays a key role in reducing genotype × environment interaction and selection of superior genotypes (1,7,39,45).Considering the influence of different factors such as biotic and abiotic stresses, minimum temperature of winter regions, the role of rapeseed in crop rotation and classification of climatic regions of Iran for the cultivation of rapeseed, identification and determination of adapted and stable genotypes with high seed yield for different environments is very important (12). In a study of 36 spring rapeseed genotypes over two years in Iran, the stable genotypes were different based on type I, II or III stability concepts (2). Jafari *et al.*(12) used Eberhart and Russell's regression method (coefficient of regression=1 and non-significant deviation from regression line) to study the seed yield stability of winter rapeseed lines for cold and mid-cold regions in Iran and reported significant interaction of genotype×year×location. However, stability should be considered as an important aspect of the variety comparison trials, because the genotype × environment interaction can reduce any selection progress during a breeding pragram. Since researchers used one of the stability methods (23,25,32,36) or combined methods (3,16,24,37) in their studies to find high-yielding and stable genotypes, the GGE biplot method was preffered and used in this study. Obviously, the selected genotypes will be introduced into the target locations for further studies in order to demonstrate their superiority to the control cultivar at the farmer's condition. In this research, a graphically represented GGE biplot analysis enabled selection of stable and high-yielding genotypes for all investigated locations, as well as genotypes with specific adaptability. The GGE biplot analysis is adequate in explaning G×E interaction for seed yield in rapeseed.Genotypes G2, G4 and G13 had the highest mean seed yield and stability in four investigated locations. For specific

adaptability, G13 can be recommended for Isfahan, Karaj and Kerman and G4 can be recommended for Mashhad.

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