



Grain yield stability analysis of soybean genotypes by AMMI method

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Article Info

Accepted:
9 Nov. 2016

Keywords:
AMMI Stability
Value (ASV), GGE
biplot, Grain yield

ABSTRACT

The additive main effects and multiplicative interaction (AMMI) model was used to analysis the grain yield stability of 20 soybean genotypes in four locations (Karaj, Gorgan, Moghan and ShahreKord) of Iran. Experiments were carried out based on randomized complete block design (RCBD), with three replications in 2014-2015. Result revealed that the grain yield was significantly influenced by environments (E), genotypes (G) and $G \times E$ interactions. Principal component analysis (PCA) declared three components which explained up to 90% of $G \times E$ sum square (IPCA1, IPCA2 and IPCA3 with 70.72%, 18.99% and 10.60%, respectively). AMMI multivariate method identified two genotypes (No.13 and No.8) with grain yield of 2789 and 2702 kg.ha⁻¹ respectively, which were stable genotypes in different environments. The study concluded that the AMMI model is a practical and effective alternative for crop breeders to screen stability of soybean genotypes for different environments.

INTRODUCTION

Soybean (*Glycine max*, L. Merrill) is one of the most important crops in the world because of its high oil content and nutritional value (Vaughan and Geissler 2008). Soybean is a crop with essential amino acids pattern that come close to satisfying the needs of the human diet (Osho et al. 1995).

Genotype and environment interaction plays a key role in phenotypic expression, and must be estimated and considered when indicating cultivars for breeding program. It is therefore necessary to study the genotype \times environment interaction (GEI) to identify the genotypes that are stable in different environments (Calvino et al. 2003). Because of $G \times E$ interaction, the process of identification of stable genotype is difficult. Although the plant breeders have observed genetic differences for adaptability, they have been unable to fully exploit these differences in breeding stable genotypes. This has been largely due to the problem of defining and measuring phenotypic stability. Various attempts

were made to characterize the behaviors of genotypes in response to varying environments (Farshadfar and Sutka 2003). The presence of significant $G \times E$ for quantitative traits such as yield can seriously limit the feasibility of selecting superior genotypes (Flores et al. 1998). Therefore, identification of yield contributing traits, and knowledge of the $G \times E$ interactions and yield stability is important for breeding new cultivars with improved adaptation to the environmental constraints prevailing in the targeted environments. Study of genotype \times environment interaction is important to plant breeders because it can limit the progress in the selection process; hence it is a basic cause of differences among genotypes for yield stability (Asad et al. 2009).

Among multivariate statistical methods, the additive main effects and multiplicative interaction (AMMI) model is a powerful multivariate method for multi-environmental trials (Romagosa and Fox 1993). This technique incorporates both additive and multiplicative components into an integrated, powerful least squares analysis (Gauch 1992; Voltas et al. 1999). Plots showing both the genotypes and the environments simultaneously can be of great assistance in this respect, and are called biplots (Gabriel 1971; Rubio et al. 2004). The GGE biplot analysis is another method which integrates the genotype and genotype by environment effects in the evaluation of cultivars. The GGE that uses graphic axes identifies superior cultivars in the mega environments (Akcura et al.

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2011). The AMMI allows for a large set of technical interpretations and uses a principal component to interpret cultivar performance by integrating the use of ANOVA and PCA. The AMMI analysis combines additive components in a single model for the main effects of genotype and environment as well as multiplicative components for the interaction effect. The graphic analyses bring out phenotypic stability, genotypic behavior of the cultivars and environments that optimize performance (Miranda et al. 2009).

Radi et al. (2003) evaluated five soybean genotypes under different locations and years. Their result revealed that seed yield remarkably affected by varying locations and years. Rao et al. (2002) tested 12 soybean genotypes and found significant genotype, year, and location interaction effects for yield. Ramana and Satyanarayana (2006) tested 16 soybean genotypes for protein, oil, and yield in five different environments and found mean sum of squares due to genotype and environment (linear) were significant. Non-linear components of genotype \times environment were significant for all traits. Similarly, Gurm et al. (2009) found strong significant environment, genotypes, and genotype \times environment effects for grain yield, oil, and protein of twenty soybean genotypes tested at six different locations.

The objective of this study was to evaluate the yield stability of soybean genotypes in different locations using the additive main effects and multiplicative interaction (AMMI) and genotype and genotype-by-environment interaction (GGE) biplot models.

MATERIALS AND METHODS

Planting Materials and Testing Locations

Twenty Soybean (*Glycine max* (L.) Merr.) genotypes were evaluated at four locations (Karaj, Gorgan, Moghan and ShahreKord) during 2014/2015 in Iran (Table 1). Field experiments were set as a randomized complete block design with four replications. Plots were five rows of 3m length spaced 0.5 m apart. Planting was done using drilling method and the plants were later thinned down to a spacing of 5cm between plants within the rows. Accepted cultural practices were applied at each location. Currently accepted levels of management and cultural practices for soybean were applied in all performance trials. Seed harvested from each plot was recorded as kilograms per plot and converted to tones per hectare at 13% seed moisture contents.

Data analysis

The grain yield data were subjected to combined analysis of variance, mean comparison using Duncan's multiple range test (DMRT) by

statistical software's SPSS and EXCEL. In the first step of combined analysis across locations, a two-factor ANOVA was performed (genotype and location, as main effects) in order to observe the importance of interactions between genotypes and locations. Genotype by environment interaction sum of squares was partitioned according to AMMI model (Gauch 1992). To determine the effects of GEI on yields, the data were subjected to AMMI and GGE Biplot analysis using SAS and GenStat softwares.

AMMI stability value (ASV)

The AMMI stability value (ASV) as described by Purchase et al, (2000) was calculated as follows:

$$ASV = \sqrt{\left(\frac{IPCA1_{SS}}{IPCA2_{SS}} (IPCA1_{score})\right)^2 + (IPCA2_{score})^2}$$

Where, SSIPC1/SSIPC2 is the weight given to the IPC1 value by dividing the IPC1 sum of square on the IPC2 sum of square. The larger the IPCA (interaction principal component analysis) scores, either negative or positive, the more specifically adapted a genotype is to certain environments, smaller IPCA scores indicate a more stable genotype across environments.

Table 1. Description and evaluation of soybean

Genotype number	Pedigree
1	Liana (8) \times Telar
2	Liana (14) \times Telar
3	Liana (16) \times Telar
4	L6 (6) \times Hacheston
5	Williams \times G.non-Photo (1)
6	Williams \times G.non-Photo (2)
7	Williams \times G.non-Photo (5)
8	Williams \times G.non-Photo (7)
9	Williams \times G.non-Photo (9)
10	Williams \times G.non-Photo (10)
11	Williams \times G.non-Photo (11)
12	Will.82 \times L87- 0174 (1)
13	Will.82 \times L87- 0174 (2)
14	Will.82 \times L87- 0174 (3)
15	L17 \times Lavina (3)
16	Williams \times Steele (2)
17	Williams \times Steele (3)
18	Williams \times Steele (4)
19	L8. P79
20	Williams (Check)

RESULTS AND DISCUSSION

The study of the stability by using AMMI

The significant effect of genotype \times environment interaction by the grain yield analysis of variance based on AMMI stability (Table 2) showed that the genotypes had different performance in different environments. According

Table 2. Analysis of Variance of AMMI Model for Yield in Four Environments

Source of Variation	df	SS	SS (%)	MS
Total	239	119472330.1		
Treatment	79	10869721.3		
Genotype	19	19112496.8	17.5	1005920.8**
Environment	3	19433038.2	17.8	6477679.4**
Genotype × Environment	57	70151886.3	64.5	1230734.8
IPCA ₁	22	49618120.4	70.7	2255369.1**
IPCA ₂	20	13095872.5	18.6	654793.6**
IPCA ₃	18	7438617.0	10.6	413256.5**
Residual	16	7886.3	0.00	492.9 ^{ns}
Polled Error	158	10756104.2		68076.6

to table 2, 17.5 percent of the genotype and also 17.8 percent of environment allocated the total sum of squares, while the effect of genotype × environment interaction allocated 64.5% of this amount which is about 3.7 fold more than the effect of genotypes. Environment was also significant at 1% probability level. Significant differences among genotypes were indicated that the genotypes performance in all test environments were different. Asfaw et al. (2009) evaluated grain yield of 11 genotypes at four sites for three cropping seasons in Ethiopia. AMMI analysis showed that grain yield variation due to environments, genotypes and GEI were highly significant ($p < 0.01$). Their reports illustrated that the environments explained the greater proportion (61.08%) of total yield variation followed by GEI (34.13%) and genotypes (4.79%), indicating the necessity for testing soybean varieties at multi-locations and over years. Sudaric et al. (2006) evaluated fourteen soybean cultivars at 15 environments in Croatia. In the combined ANOVA, genotypes, environments and GEI were significant. Genotype by environment interaction is generally of less importance than effects of genotypes and years for grain yield. Gurmu et al. (2009) studied genotype × environment interaction (GE) and stability of twenty soybean genotypes for grain yield at six environments. There are strong significant ($P < 0.01$) environment, genotype and GEI effects, and environment and GEI captured larger portion of the total sum of squares.

Principal Component Analysis (PCA)

In order to analyze the effect of genotype × environment using of AMMI model, principal component analysis was performed on the remaining matrix which the three first principal components were significant at 1% probability level (Table 3). Principal component analysis (PCA) declared three components which explained up to 90% of G × E sum square (IPCA₁, IPCA₂

and IPCA₃ with 70.72%, 18.99% and 10.60%, respectively). Birla and Ramgiry (2015) used AMMI model to analysis the yield and yield component traits data of 25 soybean genotypes grown in three environments. Main effects due to environments (E) and genotypes (G) were found significant for the all traits. The G × E interaction was significant for most of the traits. Principal Component Analysis1 (PCA 1) and PCA 2, statistically significant ($P < 0.01$) for all the traits. The highest contribution for PCA 2 was (36.2%) for grain yield per the plant.

Table 3. Yield and the first and second principal components

Genotype	Yield	IPCA ₁	IPCA ₂	IPCA ₃
1	2098.51	-0.35	8.43	-2.03
2	2763.48	18.39	-8.76	-11.10
3	2553.15	5.70	21.98	1.41
4	2859.17	18.38	2.36	9.19
5	2147.37	-1.77	-2.74	9.39
6	2227.73	5.46	14.67	-14.65
7	2817.57	23.98	4.88	1.41
8	2702.84	7.26	-0.19	8.86
9	2418.56	18.38	-7.74	-5.11
10	2357.19	-6.39	-23.01	1.61
11	2541.27	-1.40	-8.17	-11.12
12	2887.93	14.56	-13.89	7.88
13	2789.21	-8.43	5.45	4.57
14	2573.30	-13.63	-0.89	-8.59
15	2784.24	-24.28	12.39	-3.33
16	2761.53	-23.90	-7.92	-3.44
17	2920.11	-18.03	0.12	3.59
18	3141.16	-14.08	0.42	21.82
19	2772.66	-8.97	-5.95	-12.30
20	3094.29	9.21	8.59	1.96

AMMI Stability Value (ASV)

In this research, for the study of the stability of genotypes, the AMMI stability value (ASV) was used. The results are presented in table 4. In this method, the genotype is stable that has less ASV.

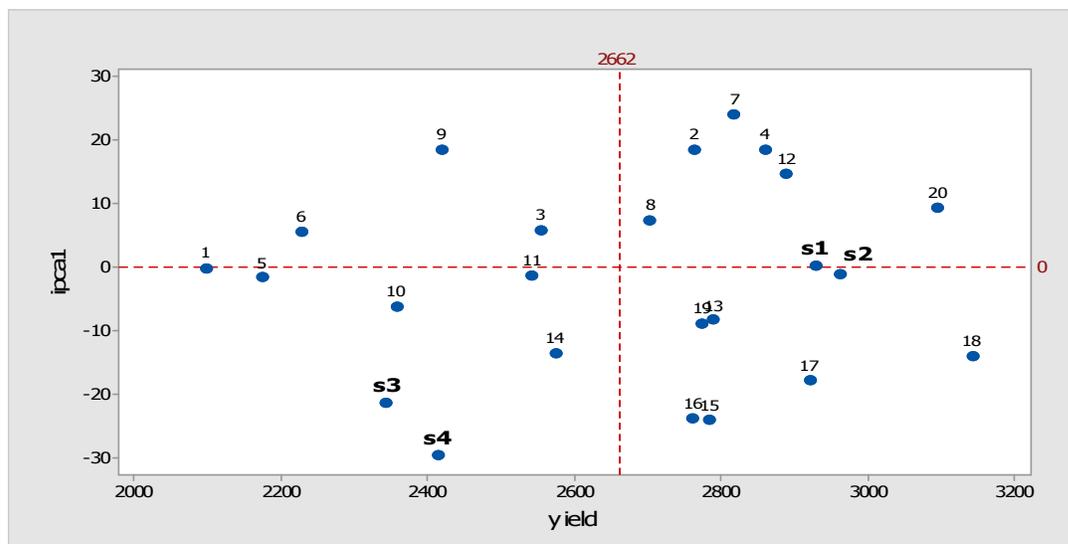


Figure 1. Biplot of mean of genotypes, Environments and the first its principal component (AMMI₁). S1 to S4 are Karaj, Gorgan, Moghan and ShahreKord, respectively.

According to this point, in this procedure, genotypes 5, 1, 11, 8 and 13 had the lowest ASV's, but among them genotypes 8 and 13 with a higher performance than the grand mean were identified as stable genotypes. Also, genotype 15 by having the highest ASV, was known as the most unstable genotype and genotypes 16, 7 and 2 were after it, respectively. Karimizadeh et al. (2007) to study the effect of genotype × environment by using AMMI in corn genotypes, used different stability parameters. They introduced ASV as the best one due to the accuracy of the results.

The study of AMMI biplot model

To easily understand of AMMI model, biplot was drawn (Figure 1). In this figure the vertical line in the middle of biplot, represents the grand mean of one year. Right side genotypes and locations of this line had higher yield than the grand mean. According to the description, genotypes 18, 20, 17, 12, 4, 7, 13, 15, 19, 2, 16 and 8, respectively, by being in right vertical line in the mid of biplot, consecrated most to least yield mean, respectively. Due to mentioned biplot, genotype 1 had the lowest yield. Also among the locations, Gorgan and Karaj had high yield and Moghan had the lowest yield. In addition, the horizontal axis represents in the middle of the graph shows IPCA1 = 0 (AMMI = 0), indicating that there is no interaction. Therefore, the genotypes that were at the center of biplot, by having close to zero interaction effect, had more general stability. So genotypes 13, 19, 8, 11, 5 and 1 have lower interaction with each other, but genotypes 13, 19 and 8 with a higher average yield than the grand mean, are selected as the genotypes with desirable stability. Tukamuhabwa et al (2012) evaluated soybean lines in five sites and three

seasons in Uganda. Results of AMMI analysis indicated the presence of a scale genotype-by-environment interaction for soybean grain yield.

Figure 2 shows biplot of the first and second principal components of interaction for genotypes. This biplot explains a total of 89.38% of variation in the data. So that the first principal component has a greater proportion (72.70%) from the second

Table 4. AMMI Stability Value (ASV) of Soybean

Genotype	Yield Mean	ASV
1	2098.51	8.45
2	2763.48	36.81
3	2553.15	24.61
4	2859.17	35.81
5	2147.37	4.39
6	2227.73	18.14
7	2817.57	46.87
8	2702.84	14.11
9	2418.56	36.56
10	2357.19	26.14
11	2541.27	8.61
12	2887.93	31.53
13	2789.21	17.27
14	2573.30	26.51
15	2784.24	48.80
16	2761.53	47.13
17	2920.11	35.05
18	3141.16	27.37
19	2772.66	18.42
20	3094.29	19.86

Table 5. Yield and the first, second and third principal components of S1 to S4 Locations

Location	Location Code	Yield	IPCA ₁	IPCA ₂	IPCA ₃
Karaj	S1	2928.67	0.20	0.16	0.04
Gorgan	S2	2961.70	-1.17	37.87	-9.97
Moghan	S3	2343.87	-21.40	-22.78	-24.74
ShahreKord	S4	2413.52	-29.65	-4.21	28.75

principal component (18.66%), respectively (Table 3).

In biplot of figure 2, genotypes 15, 16 and 10 had great and unstable interactions. Also, genotypes 12, 9, 7 and 3 have great, positive and unstable interactions and genotypes 1, 13, 19, 5, 11, 8 had the least interactions that among them, genotypes, 8, 13 and 19 due to higher performance than the grand mean, were identified as genotypes with good and wide adaptation.

In the survey of specific adaptation and determine the most appropriate genotypes for locations according to the biplot (Figure 2) genotypes 15, 16, 10, 12, 9, 7, 3 located in the vertices of the polygon with the highest response in their proper places. In fact, they have most adaptation with their proper places.

Sections created by vertical lines on the sides represent suitable genotypes and locations. In fact, the genotypes that have been at the vertex, are suitable genotypes for the location or locations where are lied. In this biplot, Gorgan (S2) located in the first section that the winner genotype is the

genotype 3. In the second section of the biplot, genotype 16 located at the vertex that is the winner genotype for Moghan (S3) and ShahreKord (S4). There was no winner genotype for Karaj (S1), due to locating at the top center of biplot.

In analysis of genotype response pattern based on two principal components, consideration of angle between the environmental vector is useful for the interpretation of environmental similarities. The acute angle between the two environmental vectors indicating high environmental correlation. AMMI method is an efficient method due to capable of graphical interpretation. Since at AMMI1 model, genotypes 13, 19 and 8 were identified as genotypes with suitable adaptation and high yield; and in AMMI2 model, genotypes 8, 13, 19 were identified as good and stable genotypes with high yield, also according to the AMMI stability value (ASV), genotypes 13 and 8 were identified as stable genotypes with high yield, generally, genotypes 13 and 8 introduced as the best genotypes with yield 2789.21 and 2702.84 kg per hectare, respectively (Table 5). Amira et al (2013) evaluated six genotypes of soybean at ten locations in Nigeria for grain yield and stability. The analysis of variance revealed significant ($P <$

Table 6. Joint Selection for Yield and Stability of Soybean Genotypes

Genotype	Yield	Yield Rank	Adjusted Yield Rank	Adjusted Rank	Joint Effect of Yield and Stability	Stability	Stability Variance
1	2098.51	1	-3	-2	-2	0	52835.2
2	2763.48	11	1	12	4	-8	468234.2
3	2553.15	7	-1	6	6	0	122214.1
4	2859.17	16	1	17	9	-8	500281.1
5	2147.37	2	-2	0	0	0	137698
6	2227.73	3	-2	1	-7	-8	319185.6
7	2817.57	15	1	16	8	-8	1008868.28
8	2702.84	9	1	10	2	-8	911348.9
9	2418.56	5	-1	4	-4	-8	492632.7
10	2357.19	4	-2	2	-6	-8	572253.1
11	2541.27	6	-1	5	1	-4	232245.1
12	2887.93	17	1	18	10	-8	630919.4
13	2789.21	14	1	15	13	-2	182537.4
14	2573.30	8	-1	7	-1	-8	419210.4
15	2784.24	13	1	14	6	-8	557975.9
16	2761.53	10	-1	9	9	0	57343.8
17	2920.11	18	1	19	11	-8	332610.5
18	3141.16	20	2	22	14	-8	881323.6
19	2772.66	12	1	13	13	0	120613.5
20	3094.29	19	2	21	13	-8	565766.2

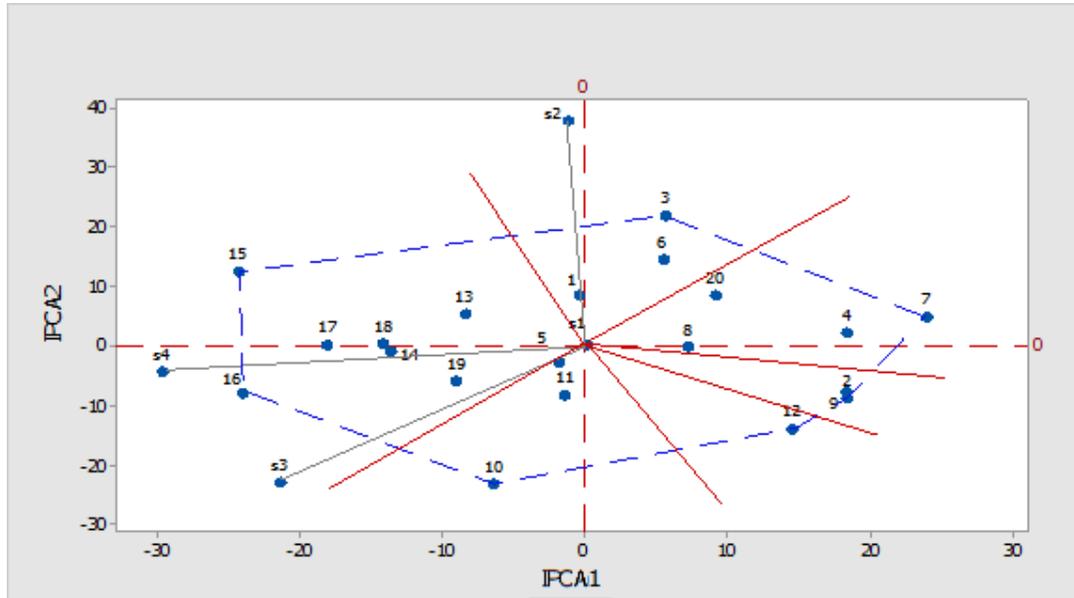


Figure 2. Biplot based on AMMI2 Model (S1 to S4 are Karaj, Gorgan, Moghan and ShahreKord, respectively)

0.05) GEI effect. Mean grain yield of the soybean genotypes ranged from 1148 kg ha⁻¹ to 1584 kg ha⁻¹. Adiea and Krisnawatia (2015) tested twelve

soybean genotypes in eight locations in Indonesia in 2013 to assess its potential yield. The seed yield range was 2.15 t ha⁻¹ to 2.79 t ha⁻¹ with an average of 2.54 t ha⁻¹. Four soybean were known as stable genotypes.

Simultaneous Selection for Yield and Stability

In this method the genotypes that are selected had high yield and stability. So genotypes 18, 20, 17, 12, 4, 7, 13, 15, 19, 2, 16, 8 were known as superior genotypes. Among these, genotypes 18, 19, 20 and 13 due to having joint the highest yield and stability than other genotypes, were superior genotypes. Genotypes 6, 10, 9 and 1 by having the lowest amount of performance and stability were as

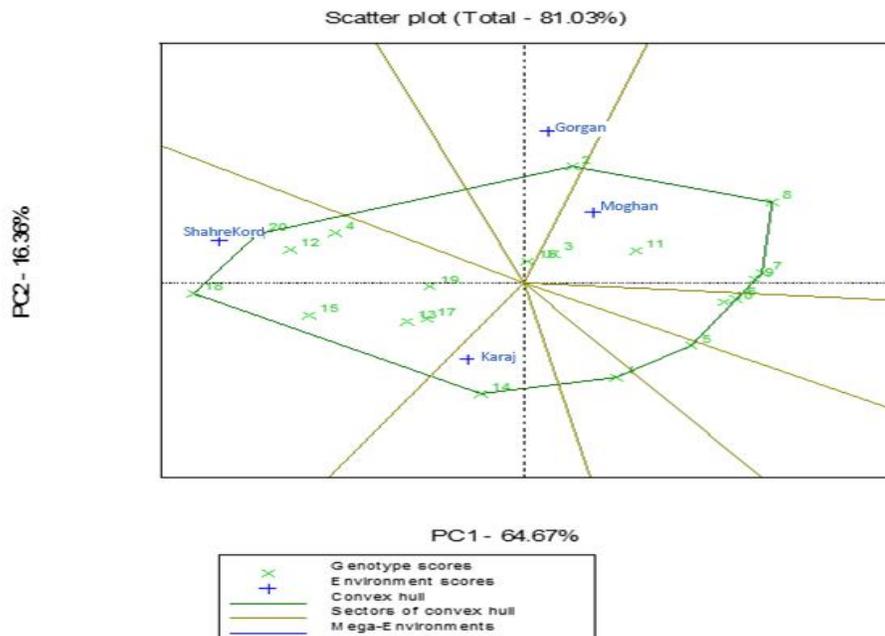


Figure 3. Polygon graph for determination of mega environments and the best genotypes for each location

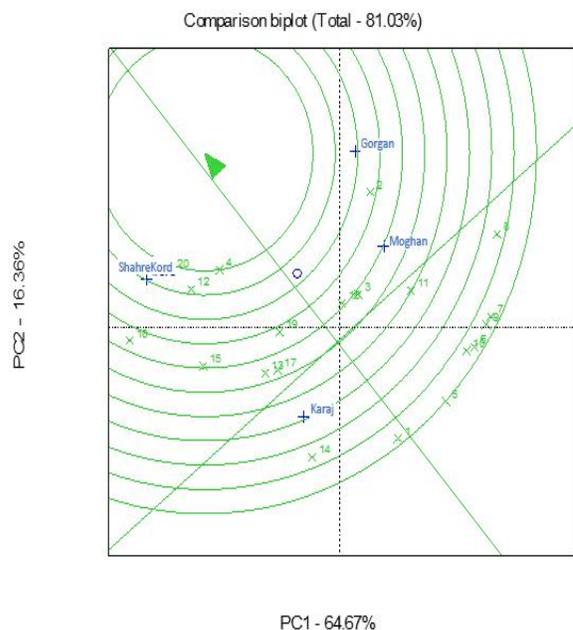


Figure 4. Determination of the best genotype by GGE biplot

the weakest genotypes (Table 6).

Stability analysis of genotypes using multivariate analysis and graphical GGE biplot

In this study, based on GGE biplot, in all biplots that are interpreted completely in the following, total of two principal components is 81.03%. In other words, each biplot explained 81.03% of yield variations (by GGE model). Sousa et al. (2015) evaluated twenty-seven early-cycle soybean genotypes. Significant and complex genotype-by-environment interactions were observed. The AMMI model presented greater efficiency by retaining most of the variation in the first two main components (61.46%), followed by the GGE biplot model (57.90%), and factor analysis (54.12%).

Determining mega environments and the best genotype in each location

A polygonal seen in figure 3 which created by connecting the farthest genotypes to each other and provides comprehensive information. With this biplot and polygon located within its, the best mega environments and genotypes can be identified for any location. In this biplot by lines that have become vertical to the polygons, eight sections are seen. The presence of genotypes in the vertex of polygons, represent the best genotypes for locations that located in the polygons. Genotypes 2, 20, 18, 14, 1, 5, 7 and 8 are at the vertex of the polygons. The Moghan is located in a section that genotypes 8 and 7 are in its vertex. This means that genotypes

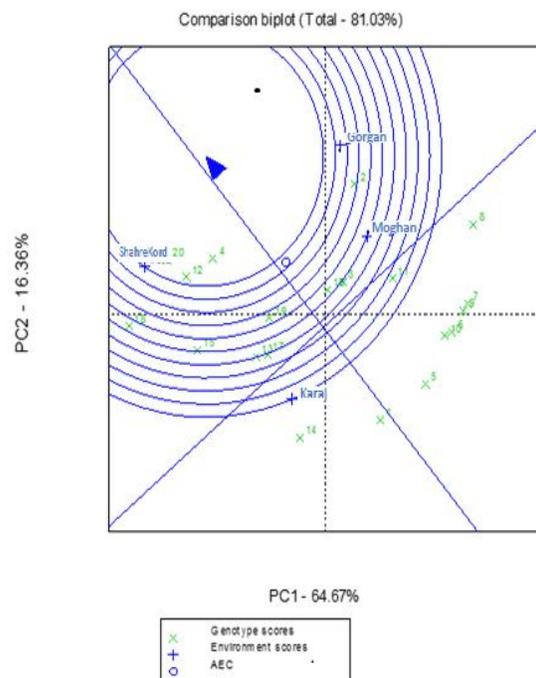


Figure 5. Determination of the best environments by graphical method

8 and 7 are the best genotypes for the Moghan. The Karaj is located in a place that genotype 14 is at its vertex. The ShahreKord is located in a place that genotypes 20 and 18 are at its vertex and the Gorgan is located in a place that genotype 2 is at its vertex. Obua et al (2013) tested combined yield of the 24 soybean genotypes across 15 site-environments in Ugandan mega-environments. The results indicated that Maksoy 3N had the highest mean yield of 1,135 kg ha⁻¹. The GGE scatter plot indicated that all the five sites can be classified as one mega environment with the best genotype being Maksoy 3N. Tukamuhabwa et al (2012) reported that by AMMI analysis, from the environmental focusing plot, the five multi-locations tested were grouped into two putative mega environments for soybean production.

The most important fact is that there is no environment within the sectors that genotypes 4 and 5 are at its vertex which suggests that genotypes 4 and 5 had no priority in any mega-environment. These biplots have the property of environmental grouping, as well. In fact, one can identify different mega-environments. In this biplot, Gorgan, Moghan, Karaj and ShahreKord was identified as the first, second, third and fourth mega-environment, respectively.

Identifying the ideal genotype using GGE biplot graphical method

As can be seen in figure 4, center of concentric circles where is located the superior genotype. So genotype 4 by being in the center of

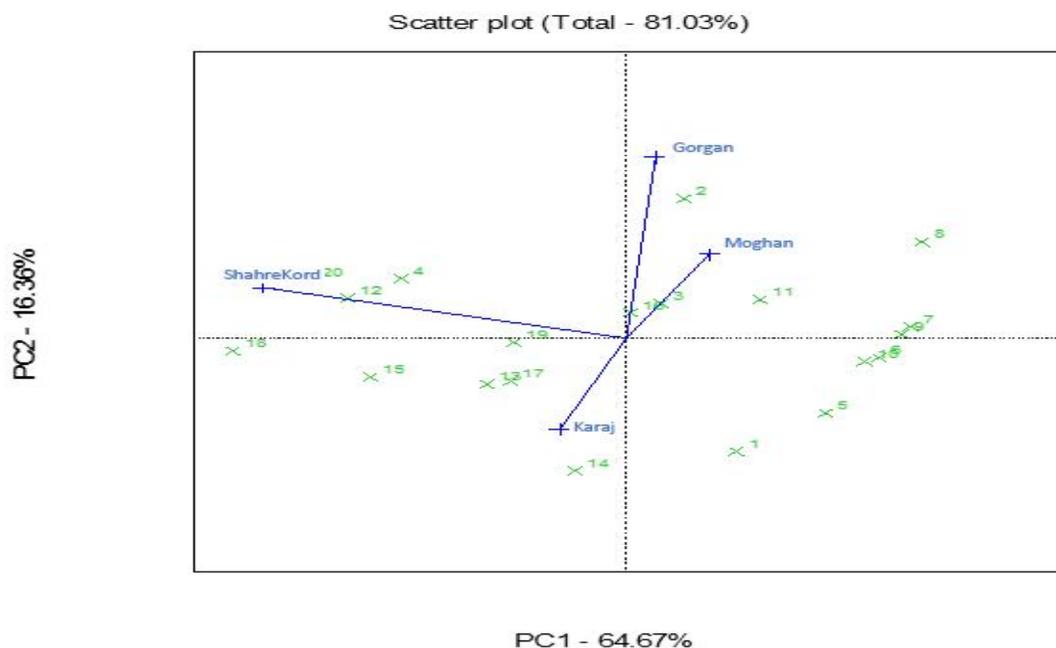


Figure 6. Study of relationships among environments by graphical method

circle is as the ideal genotype and genotypes 20 and 12 are then in the next rank. Interestingly, genotype 4 was in good condition with some of stability methods.

Determine the ideal environment using GGE biplot graphical methods

According to the figure 5, ShahreKord, among the four tested locations, known as the ideal location, due to being near the center of concentric circles. Gorgan, Moghan and Karaj locations were in the next group and due to the great distance that Karaj located from the center of concentric circles, detected as the weakest location. It should be noted that an ideal environment shows the ideal genotype response pattern.

The survey of relationships between environments by using GGE biplot graphical method

In this biplot, the correlation between the environments is determined through the angle that exists between them. In fact, angles that are less than 90° , had positive correlations. The angle of 90° between the vectors represents the independence of the environment. Angle greater than 90° represent a negative correlation between the environments. According to the explanation and Figure 6, there is a high positive correlation between Gorgan and Moghan, due to the low angle among them, which represents the similar responses of genotypes in these locations. In other words, the genotypes had high yield in Moghan, they show first ranks in Gorgan, too. Moghan and Karaj locations with close to zero correlation, result

in the creation of independent yield of genotypes at these places. According to this fact that in this study, the locations of Moghan and Gorgan have close correlation and with this in mind that these results be replicated in a similar way in a few years, in order to reduce costs, one of these places be used.

CONCLUSION

Due to the fact that in different methods, genotypes that ranked first place in each parameter, were different, but considering all methods, there were genotypes that in most of the methods were rated as stable genotypes and were in stable situations in the graphics. Based on multivariate methods of AMMI model, genotypes 13, 19 and 8 were identified as stable genotypes. Four mega environments were identified in GGE biplot. It is suggested that genotypes 8, 19 and 13, which were selected as superior genotypes in terms of yield and stability in many methods, become used in the experiments of comparative and extension studies and become accessible for farmers, in the future. In the end, it is suggested, stable experiments done in more than two years, allowing the calculation of repeatability.

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Journal sponsorship

Azarian Journal of Agriculture is grateful to the [University of Maragheh](#) and its faculty members for their ongoing encouragement, support and assistance.