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INVESTIGATION OF GENOTYPE × ENVIRONMENT INTERACTION IN CHICKPEA GENOTYPES USING AMMI AND GGE BIPLOT ANALYSIS

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ABSTRACT

Genotype by environment (GE) interaction is considered to be among the major factors limiting the efficiency of breeding programs. Fifteen chickpea genotypes were evaluated to study their adaptability and stability in eight environments of South East of Turkey. The experiment was carried out in randomized complete block design with three replications in two locations in over three years. In this study, AMMI and GGE biplot analyses were used in the evaluation of test environments and genotypes. The AMMI analysis showed that the effects of genotype, environment and genotype × environment interaction were significant ($P<0.01$) on grain yield. The results of AMMI analyses indicated that chickpea grain yield performances were highly affected by environmental effect followed by the magnitude of GEI and genotype contributed the least effect. The polygon view of the GGE biplot showed that environments used this study belonged to two mega-environments, with different winning genotypes G2 (FLIP03-128C) and G12 (FLIP09-51C). The GGE biplot also revealed that E6 (2015 Diyarbakır winter sowing) was the most discriminating environment for grain yield of chickpea genotypes. According to the AMMI, GGE biplot and linier regression models, considering simultaneous average yield and stability, G1 (EN 934) and G10 (FLIP 03-110C) genotypes were the best genotype all the environments. However, G2 (FLIP03-128C), G3 (FLIP03-28C) and G5 (X03TH130) genotypes can be regarded as adapted to a wide range of environments.

Keywords: Environment, GGE biplot, GE interaction, Ideal genotype, Stability

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is Turkey's most important food legume crop, including approximately 53.3% of the area grown to food legumes in the country. Chickpea is traditionally grown under rain-fed conditions in most of the country. Chickpea productivity remained incapable due to the lack of improved varieties (early maturing, biotic and abiotic stress tolerant, and high yielding genotypes), poor soil fertility and genotype × environment interaction. Selection of many crop varieties including chickpea suitable for the production environment is often challenged by existence of genotype × environment interaction in the variety development process. (Asfaw et al., 2009). Therefore, multi-environment yield trials (MET) are essential because of the existence of genotype × environment (GE) interactions and the MET have to be done each year to evaluate genotypes for grain yield and other important traits ((Ebdon and Gauch, 2002). The development of high yielding cultivars with wide adaptability is the basic target of plant breeders. However, this target is made more complicated by genotype × environment interaction (GEI) (Gauch and Zobel, 1996).

Obtained yield data from multi environment yield trails (MET), are usually quite large. It is difficult to understand the general pattern of the data without some kind of graphical presentation. The Biplot technique provides a powerful solution to this problem (Gabriel, 1971). A biplot that displays the GGE of a MET data, referred to as a GGE Biplot (graphical method), is an ideal tool for MET data analysis (Yan et al., 2001). Therefore, the AMMI and GGE biplot models are described as strong tools for effective analysis and comment of multi environment data structure in breeding programs (Yan et al., 2000; Samonte et al., 2005). The AMMI and GGE biplot have frequently been used for explaining GE interaction and to determine high yielding and wide adaptability cultivars. These two statistical analyses (AMMI and GGE) have broader relevance for agricultural researchers because they pertain to any two-way data matrices, and such data emerge from many kinds of experiments (Naroui et al., 2013).

The difference from AMMI is that GGE biplot analysis is based on environment-centered PCA, whereas AMMI analysis ascribe to double-centred PCA. The data structure that AMMI and GGE biplot analyses require is a two-way data matrix, such as number of genotypes tested in a number of environments. These analyses combine two statistical procedures: analysis of variance (ANOVA) and principal component analysis (PCA) (Gauch, 2006). GGE biplot methodology allows visual examination of GE interaction pattern of multi-environmental data based on two concepts. First, yield is measured as the combined effect of G, E, and GE. Second, GGE biplot technique separates two principal components, PC1 and PC2, which are also referred to as primary and secondary effects, respectively. The principal components are derived from subjecting environment-centered yield data (the yield variation due to GGE) to singular value decomposition. Then the pattern of genotypic response across environments can be graphically determined in a GGE biplot (Yan and Tinker, 2006).

MATERIALS AND METHODS

The experiments were conducted to determine the yield performances of 12 advanced lines and three commercial varieties of chickpea (Table 2) across two rain-fed locations (Adıyaman and Diyarbakır) during the 2013, 2014 and 2015 cropping seasons (Table 1). The experimental layout was a randomized complete block design with three replications at each location and year. The plot area was four rows of 6 m long with inter-row spacing of 0.3 m. Depending on weather, the genotypes were planted in the first week of November as winter sowing and in the first week of March as spring sowing. The sowing rate was 450.000 seeds per hectare. For fertilized plots, 150 kg per hectare DAP (Di ammonium phosphate which contains 18 % N and 46 % P_2O_5) was applied at planting time by sowing machine. Weeds were chemically controlled, and the harvests were made with the plot harvester in the last week of June. Grain yield was recorded from two central rows in each plot.

Statistical analyses were done using GenStat 12th edition statistical software. Before undertaking the combined analysis of variance over environments, the homogeneity of residual variance test and normality tests was undertaken using the residual plot procedure of GenStat 12th edition statistical software residual plot procedure. The genotypes were considered as fixed variables while environments were regarded as random variables and combined ANOVA was conducted. The model for a GGE biplot (Yan et al., 2000) based on singular value decomposition (SVD) of first two principal components is:

$$Y_{ij} = \mu + \beta_j + \sum_{n=1}^{k} \lambda_n \xi_{in} \eta_{in} + \varepsilon_{ij}$$

where Y_{ij} is the mean of genotype i in environment j; μ is the grand mean; β_j is the environment j main effect; n is the singular value; $\lambda_{n,}$ ζ_{in} and η_{in} are, respectively, singular value, genotype eigenvectors, and environment eigenvectors for nth interaction principal component ; and ε_{ij} is the residual effect. The GGE biplots were generated using the first two symmetrically scaled principal components (PC) for average tester coordinate (ATC), polygon view and vector view biplots (to visualize the correlations among environments or genotypes). The vector view biplots were compared with Pearson's correlation coefficients. These graphic analyses were performed using the GGE biplot software which is as an excellent tool that has many applications other than determining stability of performance (Yan, 2001).

Table 1. Environments used in the study and their main characteristics

Code	Environment	Year	Sowing time	Longitude (E)	Latitude (N)	Altitude (m)	Rainfall (mm/year)	Soil type
E1	Diyarbakır	2013	Winter	40°.13′	37°.91′	600	680	Clayey loamy
E2	Adıyaman	2013	Winter	37°.27′	37°.76′	678	970	Sandy loamy
E3	Diyarbakır	2014	Winter	40°.13′	37°.91′	600	357	Clayey loamy
E4	Diyarbakır	2014	Spring	40°.13′	37°.91′	600	209	Clayey loamy
E5	Adıyaman	2014	Winter	37°.27′	37°.76′	678	459	Sandy loamy
E6	Diyarbakır	2015	Winter	40°.13′	37°.91′	600	584	Clayey loamy
E7	Adıyaman	2015	winter	37°.27′	37°.76′	678	919	Sandy loamy
E8	Diyarbakır	2015	Spring	40°.13′	37°.91′	600	287	Clayey loamy

Table 2. Origin and pedigrees of chickpea genotypes tested in eight environments

Genotype Code	Genotype names	Origen	Genotype Code	Genotype names	Origen
G1	EN 934	Ankara /Turkey	G9	FLIP 03-131C	ICARDA
G2	FLIP03-128C	ICARDA	G10	FLIP 03-110C	ICARDA
G3	FLIP03-28C	ICARDA	G11	FLIP 03-104C	ICARDA
G4	FLIP 05-57C	ICARDA	G12	FLIP09-51C	ICARDA
G5	X03TH130	ICARDA		Arda	Diyarbakır/Turkey
G6	X03TH164	ICARDA		Diyar-95	Diyarbakır/Turkey
G7	FLIP03-112C	ICARDA		Azkan	Eskişehir/Turkey
G8	FLIP 06-111C	ICARDA			

RESULTS AND DISCUSSION

Additive main effect and multiplicative interaction (AMMI) analysis

Results of linear regression and AMMI variance analysis for grain yield of 15 chickpea genotypes tested in eight environment are presented in Table 3. AMMI analysis indicated variation among E, G and G×E showed highly significant different at level P< 0.01. The partitioning of SS indicated that environment effect was a predominant source of variation followed by GE and genotype effect. The environment effect was approximately 4.5 times higher than GEI, which suggests the possible existence of different environment groups. In genotype variation, E explains most of the variation, when variations of G and G×E are usually smaller (Yan, 2002). These results were also confirmed by many researchers (Hinsta et al., 2011; Hinsta and Abay, 2013; Gebremedhin et al., 2014). The average grain yield of each environment and genotype are given in Table 4. Environment grain yield ranged from 1.99 t ha^{-1} (E5) to 4.05 t ha^{-1} (E1), while genotype grain yield ranged from 2.51 t ha^{-1} (Diyar 95) to 3.28 t ha^{-1} (G4).

Table 3. AMMI analysis of variance for grain yield of 15 chickpea genotypes tested at eight environment

Source of variation	Df	SS	MS	F	%SS explained
Total	359	254.90	0.710		
Genotypes (G)	14	15.73	1.124	11.64**	6.17
Environments (E)	7	172.26	24.608	61.44**	67.58
Block	16	6.41	0.401	4.15	
Interaction (G×E)	98	38.88	0.397	4.11**	15.25
IPCA1	20	18.19	0.910	9.42**	46.78
IPCA2	18	8.73	0.485	5.03**	22.45
IPCA3	16	6.31	0.394	4.08**	16.23
Residuals	44	5.65	0.128	1.33	
Error	224	21.63	0.097		

*, ** Significance at respectively 5% and 1% level probability

Also, results of AMMI analysis indicated that the first three AMMI (IPCA1 to IPCA3) were highly significant (P < 0.01). The AMMI with IPCA1 and IPCA2 is the best predictive model for cross validation of the yield variation explained by the GEI (Tamene et al., 2013). The application of AMMI model for partitioning of GEI (Table 3) also revealed the first three principal component axis (IPCAs) of AMMI were highly significant (P<0.001) using an approximate F-statistic (Gollob, 1968). In this study, the first and second principal component axis explained 46.78 and 22.45% of GEI sum of squares, respectively. The mean squares for the IPCA1 and IPCA2 cumulatively contributed to 69.03% of the total GEI. The model was adequate enough to explain the total genotype × environment interaction component (Yan and Rajcan, 2002).

In Figure 1, the horizontal blue line showed the interaction score of zero and the vertical blue line indicated the grand mean yield. X-coordinate indicates the main effects (means) and the y-coordinate indicates the effects of the interaction (IPCA1). In the biplot, seven chickpea genotypes (G1, G2, G3, G5, G8, G10 and G12) and three environments (E1, E3, and E6) located on the right side of the right side of the blue vertical line (Figure 1). These were considered as high yielding genotypes and environments. Values closer to the origin of the axis (IPCA1) provide a smaller contribution to the interaction than those that are further away. Accordingly, the AMMI1graph shows that Arda and G1 genotypes stood out with the lowest IPCA1 scores (Figure 1). This indicates that these were least involved with the interaction, and are therefore the most stable. However, only the yield of G1 genotypes was above-average. On the other hand, the genotypes G7 and G4 were the most unstable, G4 with the highest average yield. Some of the environments stood out with a small contribution to the interaction (E3, E6 and E8); with an intermediate contribution (E1, E5 and E7); and with a high contribution (E2 and E4) (Figure 1). Only in environments E1, E3, and E6 averages were recorded above the overall averages (3.01 t ha^{-1}), indicating that these were favourable environments to obtain high means. The most ideal genotype should combine high yield and stable performance across a range of production environments. Among the four high yielding genotypes G1, G2, G3, G4, G5 and G10; G1 and G10 genotypes can be best evaluated based on stability and grain yield with combined low absolute PC1 score and high yield (Figure 1).

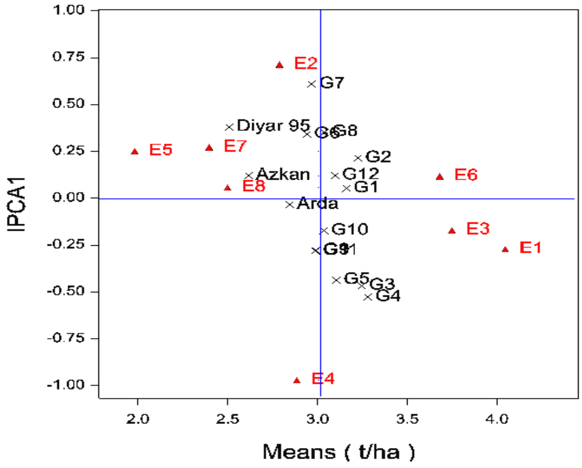

Figure 1. Biplot analysis of GEI based on AMMI 1 model for the PCA1 scores and grain yield

According to Eberhart and Russell (1966), a stable genotype is one with a high mean, a regression coefficient of unity (bi = 1), and a minimum deviation from the regression coefficient (S^2di) = 0 or close to these values of non-significant deviation. Thus, this analysis allows the identification of stable genotype for trait across environments and of genotypes that are most responsive to favorable or unfavorable environment. In the present study, Genotypes G1, G2, G3 and G10 had higher mean yield, unit regression coefficient (bi=1) and non-significant S^2di (Table 4). Thus they were found to be stable, high yielding genotypes which can be adapted to all the environments. The Linier regression model does not critically analyze the interaction of genotypes in specific environments and does not help to identify promising genotypes which should be recommended in a specific environment.

Table 4. Mean performance and stability parameters for grain yield of chickpea genotypes (as per the Eberhart and Russell model).

Genotype	**Grain yield (t/ha)**									**Stability parameters**	
	E1	**E2**	**E3**	**E4**	**E5**	**E6**	**E7**	**E8**	**Mean**	**bi**	**S^2di**
G1	4.48	3.16	3.94	2.93	2.17	3.89	2.21	2.51	3.16	1.05	0.20
G2	4.41	3.34	4.04	2.77	2.14	3.82	2.08	3.20	3.23	1.07	0.76
G3	4.36	2.58	4.00	3.58	2.15	3.77	2.72	2.82	3.25	1.00	0.45
G4	4.32	3.16	4.45	3.82	1.87	3.89	2.14	2.61	3.28	1.26	0.83
G5	4.48	2.36	3.96	3.25	2.05	3.65	2.60	2.50	3.11	1.12	0.46
G6	4.06	2.65	3.58	2.29	2.03	3.49	2.68	2.75	2.94	0.87	0.50
G7	4.36	3.31	3.22	2.36	2.08	3.61	2.54	2.37	2.98	0.93	0.96
G8	4.30	3.01	4.17	2.31	1.75	3.79	2.73	2.26	3.04	1.22	0.68
G9	4.14	2.29	3.39	3.15	1.97	3.66	2.79	2.57	2.99	0.91	0.58
G10	3.95	2.78	3.77	3.22	1.87	3.61	2.58	2.50	3.04	0.96	0.15
G11	4.51	2.39	3.48	2.97	2.03	3.69	2.21	2.63	2.99	1.10	0.47
G12	2.40	3.05	4.12	3.50	2.35	4.11	2.79	2.49	3.10	0.51	2.76
Arda	4.03	2.52	3.78	2.60	1.91	3.67	1.99	2.27	2.85	1.14	0.11
Diyar 95	2.60	2.37	3.27	2.27	1.82	3.28	2.33	2.14	2.51	0.59	0.59
Azkan	4.27	2.88	3.06	2.38	1.59	3.30	1.56	1.89	2.62	1.19	0.83
Mean	4.05	2.79	3.75	2.89	1.99	3.68	2.40	2.50	3.01		
CV(%)	10.0	10.5	9.2	12.7	11.9	8.7	11.6	7.2			
LSD $_{(0.05)}$	0.68**	0.49**	0.58**	0.61	0.40**	0.53**	0.47**	0.30**			

G= genotypes; E= environments.; bi= regression coefficient ; S^2di= deviation from the regression coefficient

Polygon view of the GGE biplot

The polygon view of the GGE-biplot analysis helps one detect cross-over and non-crossover genotype-by-environment interaction and possible mega environments in multi-location yield trials (Yan et al., 2007). The GGE Biplot graphic analyses of the fifteen chickpea genotypes tested at eight environments are presented in Figure 2. Rays in figure 2 divided the biplot into four sectors. The environments were located in two sectoral areas, while the genotypes were located in all four sectors. The genotypes found at vertex of the sectors are the most profitable genotypes of that sector. (Yan and Tinker, 2006). Three environments (E1, E2 and E3) located in the first sector and the vertex genotype for this sector was G2. The rest of environments (E4, E5, E6, E7 and E8) fell into the second sector and the vertex genotype for this sector was G12. The other vertex genotypes (Diyar 95 and Azkan) without any environment in their sectors were not the highest yielding genotypes at any environment rather they were the poorest genotypes of all or some environments. Thus these genotypes (G2, G12, Diyar 95 and Azkan) are accepted specifically adapted. G10 and G11 genotypes were closest to the center of origin, that is, these genotypes had low variation in GEI. (Abinasa et al., 2011).

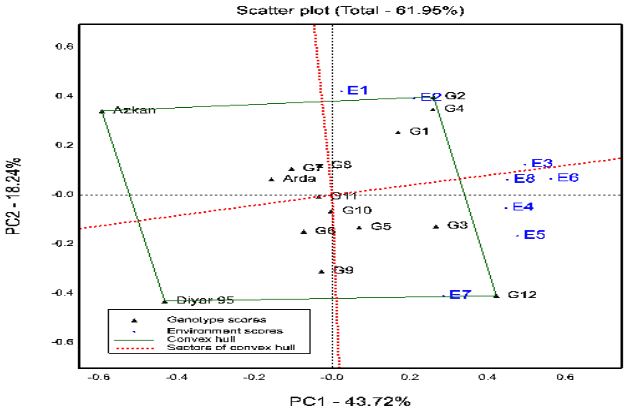

Figure 2. Polygon views of the GGE biplot based on symmetrical scaling of 15 chickpea genotypes across eight environments.

Relationship among test environments

The vector view of a GGE biplot provides a summary of the interrelationships among the environments (Yan, 2002). Provided that the biplot explained an adequate amount (≥50%) of the total variation, the correlation coefficient between any two environments is reliable (Yan et al., 2000). Furthermore, the length of an environmental vector is an estimation of discriminating power of the environment (Yan et al., 2007). Accordingly, the results of the present study revealed that the first principal component (PC1) and the second (PC2) respectively clarified 43.72% and 18.24% of the variance (Fig. 3). The two principal component axis (PC1 and PC2) together clarified 61.95% of the total variance. So this biplot can be used for extracting interrelationships among the environments. A long environmental vector represents a high capacity to discriminate the genotypes. With the longest vectors from the origin, environment E6 was the most discriminating of the genotypes, while E3, E4, E5 and E8 were moderately discriminating. However, with the shortest vector from the origin, E1 provided little or no information about the genotype differences. Furthermore, the vector view of the GGE-biplot provides a brief summary of the interrelationships among the environments. Two environments are positively correlated if the angle between their vectors is <90°, negatively correlated if the angle is >90°, independent if the angle is 90° (Yan and Tinker, 2006). Based on this, E3, E4, E5, E6 and E8 environments were positively correlated because all of the angles among their vectors were smaller than 90°. However, the angle between vectors of tester E1 and E4, E2 and E5 were approximately 90°, and were not correlated (Fig. 3).

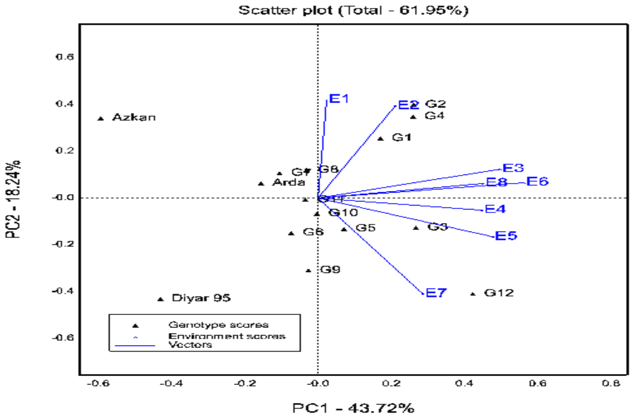

Figure 3. GGE biplot graph showing relationships between test environments

Evaluation of environments based on the ideal environment

An ideal environment is representative and has the highest discriminating power (Yan and Tinker, 2006). The ideal environment is located in the first concentric circle in the environment-focused the GGE biplot, and the environments that are close to the ideal environment are defined as the desired environments. Based on this, E6 located in the first concentric circle and has been the most ideal environment (Figure 4). Thus, genotype evaluation in E6 environment maximized the observed genotypic variation among genotypes for grain yield of the tested chickpea genotypes. E3, E8 and E4 environments were close to the ideal environment (E1), respectively and these environments has been identified as desirable environments. This difference between environments can be related to soil fertility, climate changes and other environmental variations from year to year.

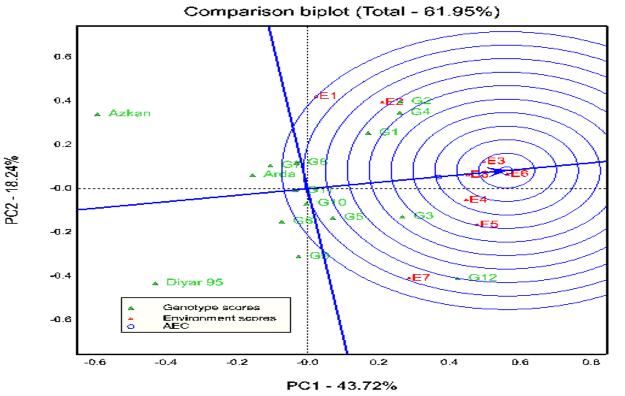

Figure 4. GGE biplot graph based on environment-focused scaling for comparison the environments with the ideal environment

Evaluation of genotypes based on the ideal genotype

An ideal genotype is defined as one of the highest yielding across the test environments and is definitely stable in performance (Yan and Kang, 2003). In the genotype-focused the GGE biplot analyses, concentric circles are drawn to help visualize the distance between each genotype and the ideal genotype (Naroui Rad et al., 2013). An ideal genotype is located in the first concentric circle of GGE biplot graphic and the genotypes that are close to the ideal genotype are defined as the desired genotypes. Based on these, placed near to the first concentric circle, genotype G4 was closer to the ideal genotype position and it can be used as reference for genotype evaluation (Figure 5). G2, G1 and G3, which were close to G4, were the more desirable genotypes than other chickpea genotypes. In spite of this, Azkan and Diyar 95 commercial cultivars were more undesirable than other chickpea genotypes and they were adapted to specific environments. Four genotypes (G11, G10, G5 and G8) were located near the biplot orgin and they were less sensitive to the environmental change.

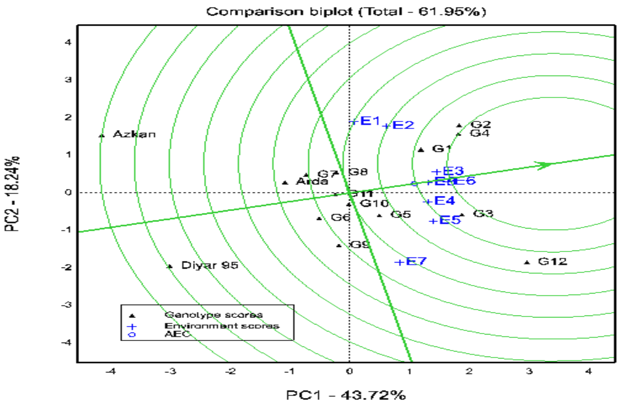

Figure 5. GGE biplot graph based on genotype-focused scaling for comparison of genotypes with ideal genotype

Mean performance and stability of genotypes

The mean yield performance and stability of genotypes was evaluated by an average environment coordination (AEC) method (Yan, 2001; Yan and Hunt, 2000; Yan, 2002). In the average environmental coordinate (AEC) system, AEC X axis (PC1) passes through the biplot origin with an arrow indicating the positive end of the axis and indicate the mean performance axis of genotypes. The ATC Y-axis passes through the biplot origin and is perpendicular to the ATC X-axis. This axis indicates the stability axis (PC2) (Figure 6). Based on these, istatistically, the stable genotypes located near the AEC X axis (PC1) with PC2 scores of almost zero. According to Figure 6, genotypes with above average yield were from G5 to G12 and located on the right side of the biplot origin, while genotypes with blow average yield were from G9 to Azkan cultivar and located on left side of the biplot origin. The genotype G12, Diyar 95 and Azkan were less stable because of the high PC2 values and they were adapted for specific environments. In respect to total environment, the stability and high yield should be considered together when making the selection. Because G1, G3, G5 and G10 genotypes were closest to zero in respect to PC2, these genotypes were more stable with above average yield. Therefore, the genotypes G1, G3, G5 and G10 with stable and high yield can be considered as commercial for the Southeast Anatolia Region in Turkey.

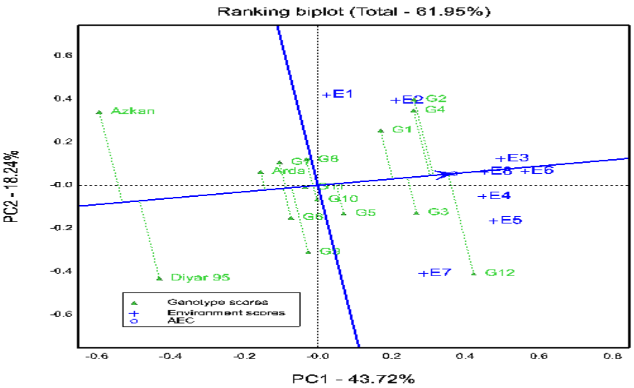

Figure 6. Average environment coordination (AEC) views of the GGE biplot based on environment-focused scaling for the means performance and stability of genotypes.

CONCLUSIONS

From the present investigation, it is concluded that multiple methods were employed to analyze stability. Those methods provided a good understanding of the adaptation level of chickpea genotypes across a diverse range of environments. The results of AMMI analyses indicated that chickpea grain yield performances were highly affected by environmental effect followed by the magnitude of GEI and genotype contributed the least effect. The AMMI and GGE biplot analysis permitted estimation of interaction effect of a genotype in each environment and it helped to identify genotypes best suited for specific environments, while liner regression analysis allowed only information about the stability status of a genotype. GGE biplot analysis showed that the polygon view of a biplot is the best way to visualize the interaction models between genotypes and environments. The polygon views of the GGE biplot pointed out that there existed two possible mega environments. The first mega environment consisted of three environments (E1, E2 and E3), the seccond mega environment consisted of five environments (E4, E5, E6, E7 and E8). In addition, the discriminating power vs. representativeness view of the GGE biplot has been an effective tool for test environments evaluation. Environment E6 and E3 were the most discriminating for grain yield of the tested chickpea genotypes. According to the AMMI, GGE biplot and linier regression models, considering simultaneous average yield and stability, G1 (EN 934) and G10 (FLIP 03-110C) genotypes were the best genotype all the

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