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Statistical analysis for genotype stability and adaptability in maize yield based on environment and genotype interaction models

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Abstract

Current analysis investigates genotype x environment interaction and stability performance of grain yield with nine maize genotypes in seven environments. ANOVA revealed highly significant (p-value<0.001) data for genotypes, environments and their interactions. Only PC1 (45.4%) and PC2 (35%) were significant ($p \leq 0.05$). Genotype G7 had a specific adaptation to environment E7, whereas genotypes G2 and G3 were adapted to environment E1, and genotypes G8 and G9 to environment E5. Dataset was divided into group A, composed of E5 and E7, and group B composed of E1, E2, E3 and E6. Genotypes G1, G2, G3 and G6, belonging to group B, were the most productive. Further, no environment fell into the G4, G5, G7, G8 and G9 sectors, denoting these genotypes as the poorest ones across environments. GGE biplot indicated that genotype G4 was highly unstable, whereas G3 very stable. In addition, G2 was more desirable due to its small contribution to both G and GE. On the other hand, G4 and G9 were more undesirable due to large contribution to either G or GE. Finally, genotypes G2 and G9 were very different. Their dissimilarity may be due to difference in mean yield and/or in GEI.

Keywords: AMMI analysis; GGE biplot; Cultivar recommendations

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1 Introduction

Our food source heavily depends on cereals which have nourished humans since their domestication thousands of years ago. They continued to be the most important source of calories for most human beings (Daryanto et al., 2016). Several cereal crops are used for food. Wheat, rice and maize are important human food sources, accounting for 94% of all cereal consumption (Ranum et al., 2014). The consumption of major cereal grains (i.e., wheat, rice and maize) varies widely by region: while wheat is the preferred cereal in Europe, rice is the major cereal in Asia and maize predominates in Africa and the Americas (Ranum et al., 2014).

Maize (Zea mays L.), also called Indian corn, is the third most important cereal crop in the world after wheat and rice (Choudhary & Kumar, 2013) since it represents 34% of total global cereal production (FAOSTAT, 2014). It originated in southern Mexico (Matsuoka et al., 2002) and spread across the Americas and subsequently to Europe, Africa and Asia (Rótolo et al., 2015). Maize belongs to the Poaceae family and is primarily a cross pollinated species, a feature that has contributed towards its broad morphological variability and geographical adaptability (Ishaq et al., 2015).

Maize, a major crop grown for food, feed, and fuel (Sharma et al., 2012; Ranum et al., 2014; Wang et al., 2016), is cultivated worldwide with large differences in yield (Ranum et al., 2014). Maize's range of cultivation stretches between 50°N to 40°S, and at altitudes varying from sea level to 3300 meters above sea level (Ishaq et al., 2015). Environmental conditions strongly influence agricultural production and cause wide variations in crop yield (Akbarpour et al., 2014). Globally, climate variability accounts for roughly a third (~32-39%) of the observed yield variability for several cereals, including maize (Ray et al., 2015).

Besides environmental fluctuations, yield is highly influenced by many genetic factors (Akter et al., 2014). Maize exhibits extensive molecular and phenotypic variation (Tenaillon et al., 2001; Wright et al., 2005; Flint-Garcia et al., 2005). Morphologically maize exhibits a greater diversity of phenotypes than perhaps any other grain crop (Wallace et al., 2014) due to its artificial selection by domestication (Wright et al., 2005). Moreover, standing height of maize genotypes at flowering may range between 0.5 and 5 meters; they mature between 60 and 330 days after planting; produce between 1 and 4 ears per plant; between 10 and 1800 kernels per ear and yield from 0.5 to 23.5 tons of grain per hectare (Ortiz et al., 2010).

As a cash crop and a model biological system, maize has a long history as a model for the study of genetics, evolution, and domestication (Wei et al., 2007; Wallace et al., 2014). Its genetic diversity has been used to analyze the molecular basis of phenotypic variation and to improve agricultural efficiency and sustainability (McMullen et al., 2009). Thus, analysis of genotype interaction with environmental conditions would improve information on the adaptability and stability performance of genotypes which influence the selection and recommendation of cultivars (Bose et al., 2014; Hongyu et al., 2014).

Expansion of crops into varied agro-climatic conditions leads to relatively different performances of the same cultivars when they are evaluated in different environments (Silveira et al., 2013). The yield trial is one of the most common experiments in agricultural research, typically testing a number of genotypes in several environments (Gauch, 2006). Under the same management conditions, variations in the yield performance of cultivars are attributed to the effect of the genotype × environment (G×E) interaction (Dingkuhnet al., 2006).

The performance of a crop is the result of the cultivar's genotype (G), the environment in which it grows (E), and the interaction between G and E (GEI) (Akter et al., 2015). Interaction between these two explanatory variables gives an insight towards the identification of the genotype suitable for specific environments, which may constitute good opportunities (Bornhofen et al., 2017). However, GEI impairs the selection of cultivars, because it changes the genotypic performance across environments (Mohamed, 2013) and minimizes the size of the association between phenotypic and genotypic rates (Alwala et al., 2010).

Several statistical methods are available to minimize the effect of G × E interaction on the selection of cultivars and the prediction of the phenotypic response to environmental changes (Silveira et al., 2013). Two frequently used statistical analyses have been the Additive Main Effects and Multiplicative Interaction (AMMI) model and the Genotype main effects and Genotype X Environment interaction effects (GGE) model (Gauch, 2006).

In recent years, the quantification of GEI and yield stability studies involving several cereal crops have been done (Benin et al., 2012; Forkman & Piepho, 2014; Pereira et al., 2014; Carvalho et al., 2016). Current assay evaluates the genotypic stability and adaptability of nine maize genotypes in seven environments using AMMI and GGE models.

2 Material and Methods

Data Collection

The yield maize was obtained from the International Maize and Wheat Improvement Center (CIMMYT; http://data.cimmyt.org/dvn/dv/cimmytdatadvn). Assays with nine genotypes (coded G1 to G9) from Tropical Three Way Crosses White Late Normal and QPM Hybrid Trial (TTWCLWQN) were conducted in seven environments, in 2013 (Table 1).

Table 1 – Geographic information, planting and harvesting dates for sites in which yield maize was evaluated in 2013.

Code	Site	Latitude	Longitude	Altitude	Planting Date	Harvest Date
E1	Costa Rica: Concepción de Pilas, Buenos Aires, Puntarenas	09°60'N	83°27'W	571	5/14/2013	9/17/2013
E2	Costa Rica: El Parque, Los Chiles, Alajuela	10°57'N	84°40'W	77	7/2/2013	11/3/2013
E3	Costa Rica: La Gloria, Concepción de Pilas, Buenos Aires, Puntarenas	09°50'N	83°27'W	430	5/14/2013	9/17/2013
E4	Costa Rica: Veracruz, Pejibaye, Pérez Zeledón	09°60'N	83°32'W	535	5/15/2013	9/18/2013
E5	Mexico: Villaflores, Chiapas	15°45'N	92°16'W	600	6/7/2013	12/26/2013
E6	Mexico: Zacapalco, Tepalcingo, Mor.	18°37'N	98°59'W	1043	7/6/2013	11/28/2013
E7	Guatemala: Cuyuta, Masagua	14°15'N	90°00'W	48	6/26/2013	10/18/2013

Data Analysis

Maize yield data for nine genotypes in seven environments were subjected to Additive Main Effects and Multiplicative Interaction (AMMI) and Genotype main effects and Genotype-Environment interaction (GGE) models using R program (R Core Team, 2016). These models combined analysis of variance (AOV) and singular value decomposition (SVD), also known as principal components analysis (PCA). While AMMI model (called doubly-centered PCA) applies SVD to data minus the genotype and environment means, the GGE model (called environment-centered PCA) applies SVD to data minus the environment means (Gauch, 2006).

AOV and SVD were invented by Fisher (1918) and Pearson (1901), respectively. However, Kempton (1984) was the first to publish an article on agriculture using AMMI and GGE, which triggered interest on the models (Gauch, 2006). These models are often applied to yield-trial data which use any two-way data matrices originating from several types of experiments. Thus, the data structure is a two-way factorial design with a number of genotypes tested within several environments.

Initially, adaptability and phenotype stability analyses were performed by AMMI method described by Zobel et al. (1988), using the statistical model below:

$$Y_{ij} = \mu + g_i + e_j + \sum_{K=n}^{n} \lambda_k \alpha_{ik} \gamma_{jk} + r_{ij} + \varepsilon_{ij'} \qquad (1)$$

where Y_{ij} is the mean response of genotype i in environment j; μ is the overall mean; g_i is the fixed effect of genotype i (i = 1, 2, ... g); e_j is the random effect of environment j (j = 1, 2, ... e); ε_{ij} is the average experimental error; G × E interaction is represented by factors; ε_k is a unique value of the k^{th} interaction principal component analysis (IPCA), (k = 1, 2, ... p, where p is the maximum number of estimable main components), ε_{ik} is a singular rate for the i^{th} genotype in the k^{th} IPCA, ε_{jk} is a unique rate of the j^{th} environment in the k^{th} IPCA; r_{ij} is the error for G × E interaction or AMMI residue (noise present in the data); and k is the characteristic non-zero roots, k = [1, 2, ... min (G, E - 1)].

Subsequently, a study of adaptability and phenotype stability of the biplot graphic was designed by combining the orthogonal axes of IPCAs. The biplot term refers to a type of graphic that contains two categories of points, which specifically refers to genotypes and environments. The biplot graphic interpretation was based on the variation caused by the main additional effects of genotype and environment, and the multiplication effect of G × E interaction. In the case of GE interaction, the biplot was interpreted by observing the magnitude and sign of the scores of genotypes and environments, for the axis (axes) of interaction.

The abscissa in AMMI1 biplot represents the main effects (overall average of the variables of the genotypes evaluated) and the ordinate is the first interaction axis (IPCA1). In this case, the lower the IPCA1 rate (absolute rates), the lower is its contribution to G × E interaction, and, therefore, a more stable genotype. The ideal genotype is that with high productivity and IPCA1 rates close to zero. An undesirable genotype has low stability associated with low productivity. In AMMI2 biplot, the points of stable genotypes and environments (with little contribution to the sum of squares of GE interaction (SSGxE)) lie near the origin.

Finally, GGE graph analysis was also performed to evaluate adaptability and phenotypic stability, described in Yan et al. (2000), using the statistical model below:

$$Y_{ij} - \mu_j = \sum_{K=n}^{n} \lambda_k \alpha_{ik} \gamma_{jk} + r_{ij} + \varepsilon_{ij}, \qquad (2)$$

where Y_{ij} is the mean response of genotype i in environment j; μ_j is the mean for environment j; ε_k is the unique rate of the kth interaction principal component analysis (IPCA), (k = 1, 2, ... p, where p is the maximum number of estimable main components); ε_{ik} is a singular rate for the ith genotype in the kth IPCA; ε_{jk} is a unique rate of the jth environment in the kth IPCA; r_{ij} is the error for G × E interaction or AMMI residue (noise present in the data); ε_{ij} is average experimental error; G × E interaction is represented by the factors; k is the characteristic non-zero roots, k = [1, 2, ... min (G, E - 1)].

GGE biplots display both G and GE, which are the two sources of variation relevant to cultivar evaluation. They are constructed by plotting the primary effect scores of each genotype (x axis) and each site against their respective secondary effect scores (y axis) such that each genotype and each test site are represented by a “marker” (Yan et al., 2001). The commonly used GGE biplot is based on the Sites Regression (SREG) linear-bilinear (multiplicative) model (Cornelius et al., 1996). GGE analysis may include two aspects: (i) mega-environment analysis and (ii) genotype evaluation (Yan & Kang, 2003).

In order to evaluate if a biplot model is adequate, Yan & Tinker (2006) proposed the “information relation (IR)” that displays patterns in a double entry table, where we can assume, for example, a table that contains genotypes and environments. The maximum number k=min(E, G-1) of PCs is required to represent this table. If there is any correlation between the environments, then the ratio of explained variance should be greater than 1/k for the first PCs, if there is no correlation between the environments, the ratio of the explained variance for each PC is exactly equal to 1/k be less than or equal to 1/k for other PCs (Yan & Tinker, 2006; Yan, 2011). The ratio of total variation explained by each PC multiplied by k is the way of calculating the IR for each PC. It is interpreted as follows: PC with IR < 1 has no pattern or information and a PC with IR ≥ 1 contains patterns of association between environments. If only the first two PCs have an IR ≥ 1, then the data patterns are adequately represented by a biplot with dimension 2 (Yan & Tinker, 2006).

3 Results and Discussion

AMMI Analysis and Biplots

Maize yield ranged between 3.2 and 12.05 t ha-1 for the different environments, and between 6.65 and 8.79 t ha-1 for the different genotypes (Table 2). The overall average maize yield was 7.46 t ha- 1. Bergamaschi et al. (2006) obtained an average grain yield of approximately 10 t ha-1 in irrigated maize plots, with a high level of management, in southern Brazil. However, the average grain yield was less than 6 t ha-1 in non-irrigated plots. Raw data of maize yield revealed that the most productive environment was E1 for all genotypes; the least being E7. In average, the most productive genotype was G2 and the least productive was G9.

AMMI analysis of variance showed that the effects of sources of variation, genotype, environment, and GEI were significant for maize yield (Table 3). This analysis of variance also showed that 80.36% of total SS was attributable to environmental effects, namely, 6.12% to genotypic effects and 8.04% to GEI. As a rule, environment explains up to 80% of the variation in multi-environment trials, while genotype and GEI usually represent approximately 10 - 15% of each variation (Gauch & Zobel, 1996). A large SS for environments indicates that the environments were distinct, with large differences among

Table 2 – Mean yield-trial data of maize (t ha^{-1}) of nine genotypes (G1 to G9) tested at seven locations (E1 to E7) in 2013.

Genotype	Test Environments							Mean
	E1	E2	E3	E4	E5	E6	E7	
G1	13.60	8.02	8.56	6.96	7.72	10.09	3.49	8.35
G2	14.25	8.39	8.98	6.95	7.63	11.69	3.64	8.79
G3	12.33	8.05	8.98	5.68	6.98	10.80	3.04	7.98
G4	11.68	3.31	8.00	6.04	5.28	11.18	2.46	6.85
G5	11.70	7.16	7.38	5.38	5.67	8.82	3.33	7.06
G6	12.40	7.40	7.73	5.42	7.15	10.00	3.30	7.63
G7	10.29	5.72	6.49	6.42	7.40	9.26	3.37	6.99
G8	11.44	4.76	7.56	5.89	7.44	7.47	3.07	6.80
G9	10.78	4.35	7.57	4.98	8.17	7.60	3.12	6.65
Mean	12.05	6.35	7.92	5.97	7.05	9.66	3.20	7.46

Table 3 – Analysis of variance for the AMMI model by fixed model of grain yield (t ha^{-1}) for maize trial and decomposition of the sum of squares of genotype (G) and environment (E) interactions.

Source	DF	SS	MS	Explained (%)	Accumulated (%)
Environments (E)	6	1292.57	215.429***	-	-
Genotypes (G)	8	98.42	12.302***	-	-
G x E	48	129.40	2.696***	-	-
PC1	13	58.76	4.52***	45.4	45.4
PC2	11	45.30	4.12***	35.0	80.4
PC3	9	12.19	1.35NS	9.4	89.8
PC4	7	6.827	0.97 NS	5.3	95.1
PC5	5	4.69	0.94 NS	3.6	98.7
PC6	3	1.64	0.55 NS	1.3	100.0
Mean (t ha^{-1}) = 7.46					
Coefficient of Variation (%) = 11.90					

DF = Degree of Freedom; SS = Sum of Square; MS = Mean Square; *** significant at 0.1%; ** significant at 1%; * significant at 5%; NS = not significant (p-value > 0.05).

environmental means which caused most variations in maize yield. In addition to environmental heterogeneity, significant differences between the genotypes are also indicated, since responses were not coincident in test environments. Genotypes were therefore characterized as environmentally-induced changes. With the method of Gollob (1968) it was verified that PC1 and PC2 were significant ($p \leq 0.05$), which implies predictions of GEI by the first two PCs. Thus, it was possible to explain 80.4% of the interaction sum of squares, at 45.4% and 35% for PC1 and PC2, respectively. The coefficient of variation was 11.90%, indicating good experimental accuracy in the test set.

AMMI1 biplot shows E1 and E6 as the most productive environments, whilst E7 was the least productive (Figure 1). In general, genotypes had similar maize yield, but the most and the least productive was G2 followed by G1, and G8 followed by G9, respectively. Maize yield of genotypes G1, G2, G3 and G6 were above average, while E1, E3 and E6 were above average among the environments.

In the AMMI2 biplot, genotypes G1, G5, G6 and G7 were the most stable and contributed less to GEI, since they were positioned near the origin of the biplot (Figure 2). The other genotypes were unstable and contributed more to GEI. In fact, they were farther off the origin. On the other hand, E3 and E4 respectively contribute less to GEI, or rather, it was not only stable but also the largest contributor to the genotypes' phenotype stability. If genotypes and environments are positioned close to one other in the biplot, they are positive associations with specific adaptation enabling the establishment of agronomic zones with relative ease (Silveira et al., 2013). Genotype G7 had a specific adaptation to environment E7, whereas genotypes G2 and G3 were adapted to environment E1, and genotypes G8 and G9 to environment E5.

Figure 1 – AMMI1 biplot (main effect vs PC1) for maize productivity data (t ha^{-1}) with nine genotypes (G) and seven environments (E).

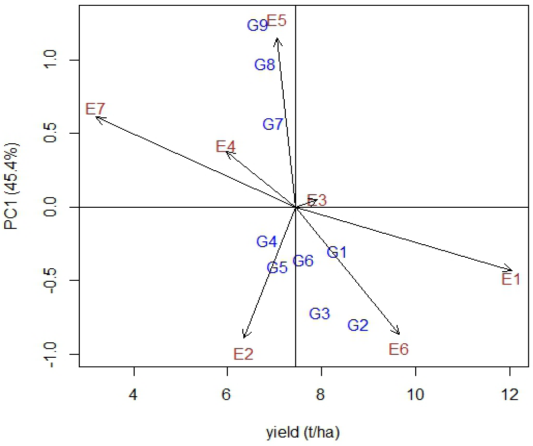
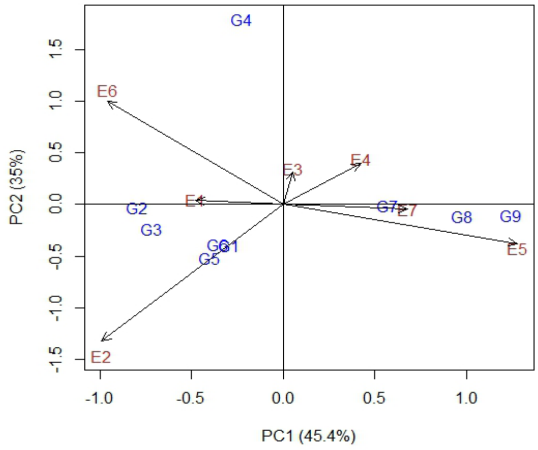


Figure 2 – AMMI2 biplot (PC1 vs PC2) for maize productivity data (t ha^{-1}) with nine genotypes (G) and seven environments (E).



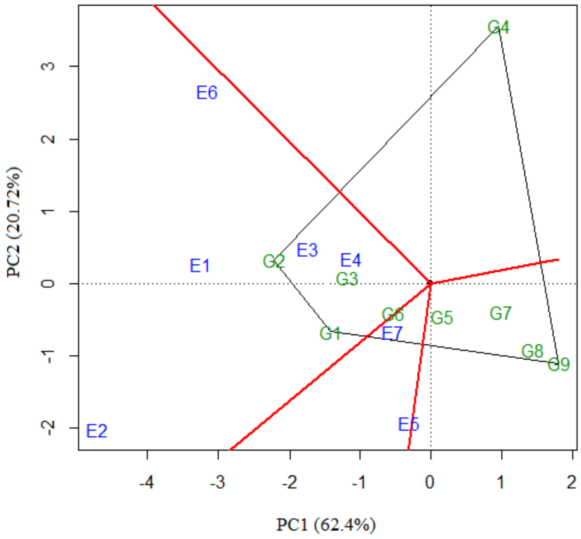
Mega-environment Analysis by GGE biplot

While GGE biplot of PC1 scores explained 62.4%, PC2 scores explained 20.72% of the total G+GE variation, totalizing 83.12% (Figure 3). One attractive feature of the GGE biplot is to graphically show the ‘which-won-where’ pattern of a genotype environment two-way data (Yan et al., 2000), revealing mega-environments. The latter are defined as a group of sub-regions that consistently share a single genotype or a group of similar genotypes specifically adapted and the best in performance (Gauch & Zobel, 1996; Yan & Rajcan, 2002).

Mega-environments on biplot consist of an irregular polygon and a set of straight lines that radiate from the biplot origin to intersect each of the polygon sides at right angles (Yan, 2011). Vertices are genotypes makers and they radiate lines that perpendicularly intersect a polygon side representing hypothetical environments. Thereby, the radiate lines divide the biplot into sectors. There is a vertex genotype for each sector, indicating a nominal winner for environments falling into that sector (Yan, 2011).

The vertex genotypes which formed the polygon were G1, G2, G4 and G9 (Figure 3). The nine environments were cut into two groups by the radiate lines: (i) group A composed of E5 and E7; (ii) group B composed of E1, E2, E3 and E6. G1 and G2 are vertex genotypes in the sector named group B, where G3 and G6 are also included. Genotypes belonging to group B were the most productive. No genotype is the vertex or belong to group A. Since no environment fell into the sectors of G4, G5, G7, G8 and G9, this fact denoted that these genotypes were the poorest across the environments.

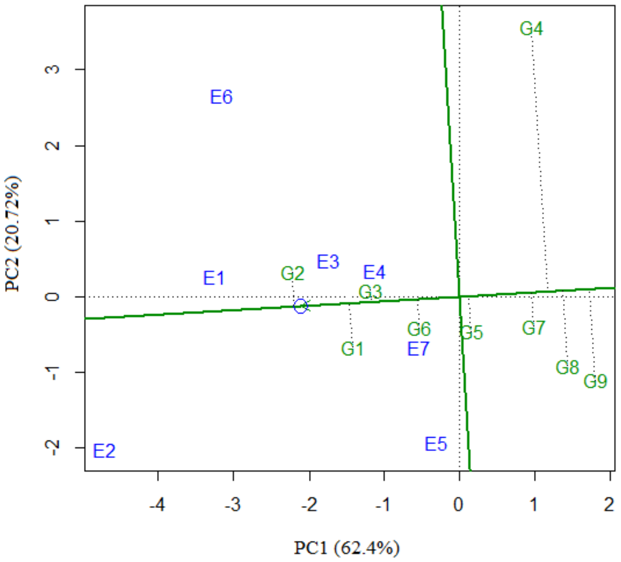
Figure 3 – Mega-Environment Delineation by GGE Biplot for maize productivity data (t ha^{-1}) with nine genotypes (G) and seven environments (E).



Genotype Evaluation by GGE biplot

The single-arrowed line is the average-environment coordination (AEC) abscissa and it points to higher mean yield across environments. Thus, G2 had the highest mean yield, followed by G1, whereas G9 had the least mean yield (Figure 4). The genotypes are therefore classified according to their average productivity as follows G2 < G1 < G3 < G6 < mean < G5 < G7 < G4 < G8 < G9. Further, the double-arrowed line is the AEC ordinate and it points to greater variability (poorer stability) in either direction. Whereas G4 is highly unstable, G3 was very stable.

Figure 4 – Mean performance and stability of genotypes by GGE Biplot with the average-environment coordination (AEC) for maize productivity data (t ha^{-1}) with nine genotypes (G) and in seven environments (E).



An ideal genotype should have high mean performance plus high stability across environments (Yan & Tinker, 2006). The genotype should be in the center of the concentric circles, corresponding to absolutely stability. However, genotypes closer to the concentric circles are more desirable than others external to it. Consequently, G2 is more desirable and G4 and G9 are the least undesirable. Further, G2 provided small contribution to both G and GE due to its nearness to the biplot contribution; G4 provided a significant contribution to either G or GE due to its longer vectors to the biplot origin. Furthermore, genotypes G1, G2 and G3 are quite similar, whereas G2 and G9 are very different. Their dissimilarity may be due to difference in mean yield and/or in GEI.

From the information ratio (IR ≥ 1) for the two models containing the patterns in the main components, the IR1 = 4.37 (0.624 x 7) in GGE biplot versus IR1 = 3.18 (0.454 x 7) of AMMI, GGE biplot explained greater proportion of the variation of the data than the PC1 of the GGE biplot (table 4). In the AMMI analysis, the first two components (PC1 and PC2) explained 80.4% of the total variation of the G × E, already in the GGE biplot analysis, the first two components explained 83.12% of the total variation. The biplot GGE was the best model for this dataset, it explained greater variations with the first two components, therefore, the interpretations of the biplot plots are the most reliable to represent the patterns in the data on genotypes and environments.

Figure 5 – Adaptability and stability with Centroid Method (ideotype) by GGE Biplot for maize productivity data (t ha-1)

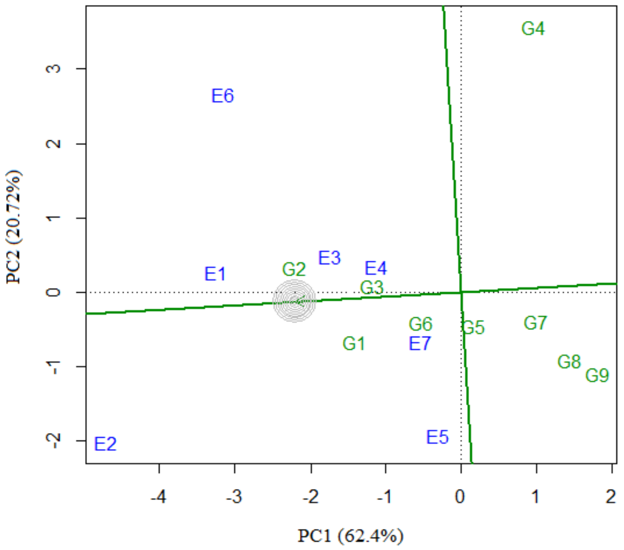


Table 4 – Singular values and explained variation of the two main components (PCs) for the models AMMI and GGE biplot.

Models	Singular values		Explained variation	
	1	2	PC1	PC2
AMMI	2.49	1.17	45.4%	35%
GGE biplot	6.88	3.96	62.4%	20.72%

4 Conclusions

The AMMI and GGE biplot models were effective in the study of the genotype VS environment interaction. Both methods achieved satisfactory results, the difference was minimal from model to model, even so, the GGE biplot model was the most efficient because it explained the greater variation of the first two components (83.12%), therefore, to represent the standards in the applied data, the interpretations of the biplot graphics are the most reliable.

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