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Statistical evaluation of genotype by environment interactions for grain yield in Millet (*penniisetum glaucum (L) R. Br*)

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----------**ABSTRACT**----------
Over the years, Pearl Millet has suffered set back in the production due mainly to the poor varieties used and poor environmental condition by famers which in turn lead to shortage of food production, poor commercialization and trade, market opportunity, unemployment among others. This study aimed at applying AMMI Model and GGE biplots in possessing the stability and adaptability of patterns of GE interaction in Pearl Millet varieties.The combined ANOVA and AMMI analysis for grain yield of forty (40) millet genotypes at 4 environments showed that environments, genotype and GxE interaction revealed highly significant (P<0.001) variations. The analysis also show that millet grain yield was significantly affected by environment E, which explained 33.20% of the total treatment (G+E+GE) variation, whereas the genotype G and GEI were significant accounted for 22.72% and 44.01% respectively. In additive variance, the portioning of (GE) SS data matrix by using AMMI analysis indicated that the two PCAs were significant (P<0.001). The first IPCA axis (IPCA1) accounted for 62.58% of the GxE interaction sum of squares, using 41 degree of freedom. The second IPCA axis (IPCA2) accounted for 30.71% of the interaction sum of squares using 39 degree of freedom. Both represent a total of 93.29% variation. Graphical display of genotype by environment interaction (GGE-biplot) based on the genotype ranking is shown on the graph of genotype so-called "ideal" genotype. genotype-focused scaling was depicted in order to detect the locations of genotypes, whereas the millet genotypes were divided into three groups based on their scores of PCA 1 and PCA 2: four stable and high yielding genotypes (G11,G7,G10 and G8), three stable low yielding genotypes (G12, G23, and G21). Genotype G11, G7, and G17, had specific adaptation to E2 and E4, and E1 and E3 is unfavorable environment. Variety G11 can thus be used as a reference genotype in cultivar evaluation follow by Variety G8, G7, G10, G27 as superior variety in this study. In our research both of AMMI and biplot model were successful in assessing the performance of genotype and the selection of best genotype were identical in both of them. We used both models to analyze 40 millet varieties in 4 environments and reported that the AMMI model and GGE biplot models were very useful in estimating the performance of millet genotype**.**

Key words: (MET) Multi-environmental trial, **(AMMI)** Additive main effects and multiplicative interactions, **(GEI)** genotype by environment interaction**, (PCA)** principal component analysis, Millet, biplot**.**

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I. INTRODUCTION

Agricultural research on finding high yield performance of Pearl Millet varieties is generally accepted as an important means of raising agricultural productivity, Commercialization and trade, market opportunity, farmers income and food security. But there are a lot of challenges that alleviate the growth in yield potential of Pearl Millet. The productivity and profitability of this crop is low, so is the income of small farmers, but through the identification of new improved Pearl Millet varieties would enhance and improving the income and food security of small scale farmers in the West African region.
Over the years, Pearl Millet has suffered set back in the production due mainly to the poor varieties used and poor environmental conditions by farmers which in turn lead to shortage of food production, poor commercialization and trade, market opportunity, unemployment among others.

Until recently, statistical analyses focuses is aimed at investigating the performance of Pearl Millet varieties in various locations and identify the superior varieties among different localities that can give high yield (increase production) with the bid to enhance commercialization and trade, create market opportunity, employment and food security to the Nations.

The yield variation due to changing environment is commonly referred to as genotype × environment interaction (G × E). G × E usually complicates the process of selecting superior genotypes. Consequently, multi-environment trials (METs) are widely used by plant breeders for evaluating the relative performance of genotypes over the target environments (Delacy *et al.*, 1996). A wide array of statistical techniques have been developed to study and reveal the nature of G×E interaction, e.g., joint regression(Finlay and Wilkinson, 1963; Eberhart and Russell,1966), additive main effects and multiplicative interaction (AMMI) (Gauch, 1992) and type B genetic correlation (Burdon, 1977). These methods are commonly used to analyze MET data and have also been applied in G×E interaction studies in many crops.

In genotype variation, E explains most of the variation, and G and G × E are usually small (Yan, 2002). However, only G and G × E interaction are relevant to variety evaluation, particularly when G × E interaction is determined as repeatable. Hence, Yan *et al.*(2000) deliberately put the two together and referred to the combination as GGE. Following the proposal of Gabriel (1971), the biplot technique was also used to display the GGE of MET data, and is referred to as a GGE biplot (Yan, 2001; Yan *et al.,* 2000).The GGE biplot is in fact a data visualization tool that graphically displays G × E interaction in a two way table (Yan *et al*., 2000). The GGE biplot is an effective tool for the following applications:1) Mega-environment analysis (e.g.; "which won-where" pattern), whereby specific genotypes can be recommended for specific mega environments(Yan and Kang, 2003) .2) Genotype evaluation (mean performance and stability), and. 3) Environmental evaluation (to discriminate Among genotypes in target environments).GGE biplot analysis is increasingly being used in G × E interaction studies in agricultural research.

AMMI is a multivariate technique for assessing the stability and adaptability of genotypes (Pacheco and Vencovsky, 2005). This method partitions the overall variation into G, E and G × E. 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. The experiment may or may not be replicated. These analyses combine two statistical procedures: analysis of variance (ANOVA) and principal component analysis (PCA) (Gauch, 2006)

The permutation of analysis of variance and PCA in the AMMI model together with forecast assessment is an important approach for better understanding GEI and obtaining better yield estimates. The interaction is explained in the form of a biplot display where, PCA scores are plotted against each other and it provides visual inspection and interpretation of the GEI components.

The purpose of this research was to apply GGE biplot, and AMMI techniques to study the patterns of G×E interaction in millet; to graphically display means, adaptability and stability of millet genotypes and environment.

II. MULTI – ENVIRONMENT TRIALS (METs)

Multi – locational trials often called Multi – environment trials are simply trials or experiments carried out in multiple environments or contexts. In Agriculture and related environmental and rural development research, METs are standard research tools. In fact, some scientific journals in Agriculture have rule that research is only acceptable for publication if results are available from several sites or seasons, that is from multiple environments.

Multilocational trials are mainly conducted to test and assess superior genotype from different environmental locations. They are used to ascertain which entries, if any, are superior to existing ones and to determine the stability of performance across sites and years. The data are also used to establish the area of adaptation in which the genotype will be recommended for cultivation. Critical issue in the conduct of Multilocational trials is how to select the test sites and, once they are chosen, using management conditions that will most efficiently identity superior entries, often multilocational trials are used to select cultivars with adaptation that perform well over a wide range of environments.

Developing the high yielding and good quality genotypes as well as more stable genotypes are very important for researchers. (Gauch H. G, 2006). The superior genotypes to deal with unpredictable environmental factors have been studied in MET. In most cases, GE interaction is observed, complicating selection for improved millets due to the effect of the environmental factors such as soil type, weather conditions etc. (Annicchiarico, 1997)

Due to the interaction noise in the experiment, yield trials for studying genotypes are carried out in numerous locations and in the course of several years. Data of such trials may have three principle tasks, to;

i. Evaluate accurately and to predict the yield on the basis of limited experimental data:
ii. Determine stability and explain variability in the response of genotype across locations: and
iii. Be a good guide for the selection of the best genotype. (Bobic V., *et al.*, 2010)

III. MATERIAL AND METHODS

Forty genotypes based on preliminary trial, were tested GxE trials at four locations. They were Sadore local, Kapielga, Toronia, Zatib, Zongo, HKP, CIVT, SoSan C-88, Taram, SoSank, ICMV IS 89305, ICMV IS 90311, Synthetic 1-2000, NKO x TC1, Guefoue 16, Indaina 05, NKK, Bongo short head, Manga Nara, Arrow, Tongo Yellow, PT732B, P1449-2, ¾ Ex-Borno, ¾ HK, ¾ Souna, Gwagwa, LCIC 9702, DMR 15, DMR 68, DMR 72, GB 8735, 99-72, TG102, T99B, T454, IBMV8401Mx68A4R4w, 01MisoNCD2-NE, 68Ax086R, and 99M59043Mw x 68A4R4MIB05. The varieties used in this study were obtained from researchers at national and international programs. In 2003, field trials were grown in Ghana, Mali, Senegal and Nigeria; Experiments were arranged in a randomized complete block design with four replications in each environment. The data has already been used for other purpose, it is secondary data.

IV. STATISTICAL ANALYSIS

Gauch (1988, 1992) has advocated the use of AMMI analysis for yield trials experiment, Gauch and Zobel (1988) compared the performance of AMMI analysis together with the ANOVA approach and regression method and from that ANOVA fail to detect a significant interaction component and the regression method accounts only a small portion of the interaction sum of squares only when the pattern fits a specific regression model.

The model AMMI model for G genotype and E environment is given as $Y_{ij} = \mu + \alpha_i + \beta_j + \sum_{k=1}^{m} \delta_{ik}\theta_k\gamma_{jk} + \varepsilon_{ij}$

$\varepsilon_{ij} \sim N(0, \sigma^2)$ $i = 1,2,..G,$ $J = 1,2,... E$

Where y_{ij} is the yield of the i^{th} genotype in the j^{th} environment, μ is the grand mean, $\alpha_i and \beta_j$ are the genotype and environment deviations from the grand mean respectively, θ_k is the Eigen value of the IPC analysis axis n ; $\delta_{ik} and \gamma_{jk}$ are the genotype and environment Eigen vectors for axis n ; n is the number of principal components retained in the model and ε_{ij} is the error term. The residual combines the PCA scores from the N-n discarded axis, when N= min $(g-1)(e-1)$ the other constrain in the model are $\sum_{I=1}^{k} \delta_{in}^2 = \sum \gamma_{jn}^2 = 1 \forall n$:

$$\sum_i \delta_{in}\delta_{in*} = \sum_j \gamma_{jn*} = 0, n \neq n^* \text{ and}$$

$$\theta_1 > \theta_{2.} > ...\theta_n > 0.$$

Thus the model can be reparamaterized as $Y_{ij} = \mu + g_i + e_j + Z_{ij}$

Where $Z_{IJ} = \sum \theta_n \delta_{in} \lambda_{jn} + \varepsilon_{ij}$

In this modified AMMI stability parameter, all significant IPCs were used. Crossa (1990) pointed out three main purposes of AMMI models: (i) model diagnosis — AMMI is more appropriate in the initial statistical analysis of yield trials because it provides an analytical tool for diagnosing other models as subclasses when these are better for a particular data set (Bradu and Gabriel, 1978); (ii) to clarify GEI — AMMI models summarize patterns and relations of genotypes and environments (Kempton, 1984; Zobel *et al*., 1988; Crossa *et al*. 1990); and (iii) to improve the accuracy of yield estimates — gains have been obtained in the accuracy of yield estimates that are equivalent to increasing the number of replicates by a factor of two to five (Zobel *et al*., 1988; Crossa *et al*., 1990) which can be used to reduce the costs by reducing the number of replications, to include more treatments in the experiment, or to improve efficiency in selecting the best genotypes. There are several possible AMMI models characterized by a number of significant principal component axes ranging from zero (AMMI-0, i.e. additive model) to min(g-1,e-1), where *g* is the number of genotypes and *e* is the number of environments. The full model (AMMI-F), with the highest number of principal component axes, provides a perfect fit between expected and observed data.

V. PRINCIPAL COMPONENT ANALYSIS

Principal component analysis is variable reduction technique; it is a linear combination of weighted observed variable and is uncorrelated and orthogonal; it also minimizes the sum of the squared perpendicular to the x-axis (not perpendicular to the fitted line).
Principal component analysis is the most frequently used multivariate method (Crossa, 1990; Purchase, 1997). Its aim is to transform the data from one set of coordinate axes to another, which preserves, as much as possible, the original arrangement of the set of points and concentrates most of the data structure in the first principal component axis. Various limitations have been noted for this technique (Perkins, 1972; Williams, 1972; Zobel *et al.,* 1988).

It was observed that the linear regression method use only one statistic to describe the pattern of response of a genotype across environments and most of the information is wasted as a result of accounting for deviation. Principal component analysis (PCA) is a generalization of linear regression that overcomes this difficulty by giving more than one statistic, the score on the principal component axes to describe the response of a genotype.

The model

$$Y_{ij} = \mu + \alpha_i + \beta_j + \sum_{k=1}^{m} \delta_{ik}\theta_k\gamma_{jk} + \varepsilon_{ij}$$

Where θ_k is the Eigen value of the PCA analysis k; δ_{ik} *and* γ_{jk} are the genotype and environment principal component scores for axis k; n is the number of principal component retained in the model and ε_j is the error term. There are many techniques which can perform similar work as PCA does, but we choose PCA rather than other techniques because when using fixed set of components there is no assurance that a small number of components will give a good reformation of the original data. PCA guarantees that the first components will perform better (mean square) work of reformation of the original data than any other linear model using only one component.It is also good at preserving distances between the points; the component scores give the most favorable linear multi-dimensional scaling. PCA also offer us uncorrelated component which are generally not independent component, for that you need independent component analysis (Stone, 2004). PCA is purely a descriptive technique; in itself it makes no forecast about what prospect data will look like. Eigen vectors are the weights in a linear transformation when computing principal component scores, while Eigen values indicate the amount of variance explained by each principal component for each factor.

GGE-biplot methodology, which is composed of 2 concepts, the biplot concept (Gabriel, 1971) and the GGE concept (Yan et al., 2000) was used to visually analyze the METs data. This methodology uses a biplot to show the factors (G and GE) that are important in genotype evaluation and that are also the source of variation in GEI analysis of METs data (Yan et al., 2000; 2001). The GGE-biplot shows the first 2 principal components derived from subjecting environment centered yield data (yield variation due to GGE) to singular value decomposition (Yan et al., 2000). In the current study, genotype-focused scaling was used in visualizing for genotypic comparison, with environment-focused scaling for environmental comparison. The statistical analysis was conducted using GenStat 16th edition.

VI. RESULT AND DISCUSSION

The combine ANOVA and AMMI analysis for grain yield show that environments, genotype and GxE interaction revealed highly significant (P<0.001) variations. The analysis also show that millet grain yield was significantly affected by environment E, which explained 33.20% of the total treatment (G+E+GE) variation, whereas the genotype G and GEI were significant accounted for 22.72% and 44.01% respectively.

In additive variance, the portioning of (GE)SS data matrix by using AMMI analysis indicated that the two PCAs were significant (P<0.001). The first IPCA axis (IPCA1) accounted for 62.58% of the GxE interaction sum of squares, using 41 degree of freedom. The second IPCA axis (IPCA2) accounted for 30.71% of the interaction sum of squares using 39 degree of freedom. Both represent a total of 93.29% variation.

The yield variation explained by environment indicated that the environments were not diverse, there are not large differences between environments, but it can also contributing to the variation in grain yield. In Table 2 the environments showed much variability in both main effect and interaction.

TABLE 1: ANOVA table for AMMI model

Source	d.f.	Sum of Squares	Mean Square	V.R.	F pr
Total	479	199147643	415757		
Treatments	159	199140547	1252456	56671.60	<0.001
Genotypes	39	45239963	1159999	52488.06	<0.001
Environments	3	66126020	22042007	880375.12	<0.001
Block	8	200	25	1.13	0.3407
Interactions	117	87774564	750210	33945.77	<0.001
IPCA 1	41	54927370	1339692	60618.87	<0.001
IPCA 2	39	26955839	691175	31274.55	<0.001
Residuals	37	5891355	159226	7204.71	<0.001
Error	312	6895	22		

The "which-won-where" pattern of the GGE biplot (Yan *et al.*, 2000) is the most suitable tool for mega-environments analysis in variety trials (Yan *et al.*, 2007). The "which-won-where" pattern of MET data is represented by a polygon formed by connecting the markers of genotypes that are further from a biplot origin, and a set of lines drawn from the biplot origin perpendicular to each side of the polygon. The perpendicular lines to the polygon sides divide the polygon sectors, each having its own winning cultivar which is the vertex genotype for that sector (Yan *et al.*, 2000). Seven out of the forty genotypes located in the vertex formed a seven-sided polygon having seven possible sectors (Figure1). The vertex genotype for each sector is the one that yielded the highest for the environments filling within that sector. Five of the sectors had no environments. The four environments fell into two sectors delineated by different winning genotypes. With the present figure G2, G6, G11, G7, G16, G35, G33 expressed a high interactive behavior (positive or negative). Whereas the environment E1 exhibited low interaction, E2 stood

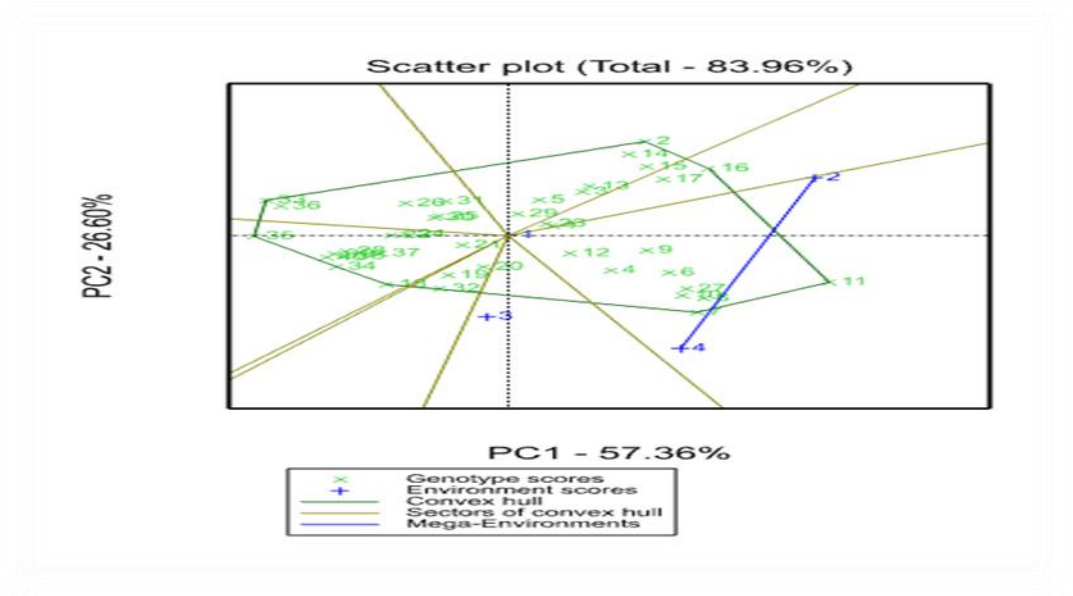

FIGURE 1. GGE biplot showing "which-won-where" the environment indicated by + and genotype by × respectively.

as intermediate between the three Genotype G16, G11 and G7 sectors indicating the existence of one mega location, according to this biplot, G16, G11 and G7 are expected to give the same yield at E2. Genotype G11 was the winning genotype at E4, although G7 is expected to give the same yield in E4.

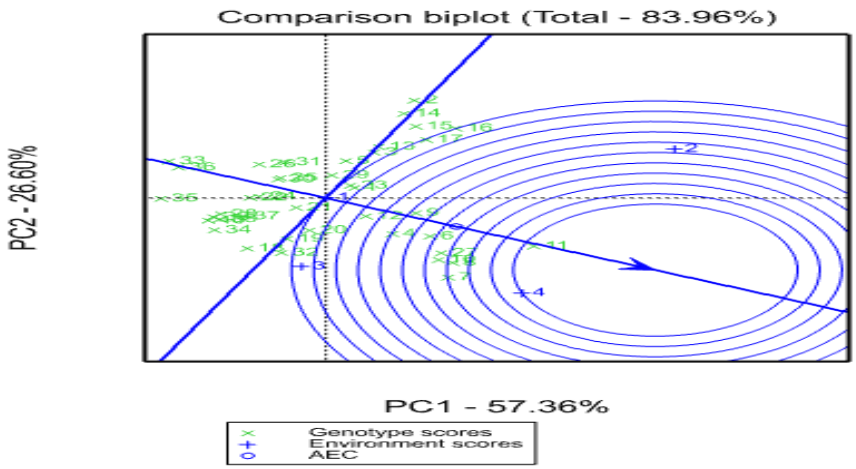

FIGURE 2. GGE biplot based on genotype focused scaling for comparison the genotypes with the ideal environment.

The vertex genotypes G18, G35, G33, G2 and G16 had no environment in their sector. The five genotypes were not the highest yielding ones at any of the test environments. G23 and G21 are located near to the plot origin and hence were less responsive than the vertex genotypes. The genotypes within the polygon and located nearer to plot origin are less responsive than vertex genotypes (Yan *et al.*, 2001). E2 and E4 have the best genotype as G11; so G11 is adaptable in both environments. The MET indicate the presence of different mega-environments, which is defined as the group of locations that consistently share the most suitable set of genotypes across years.

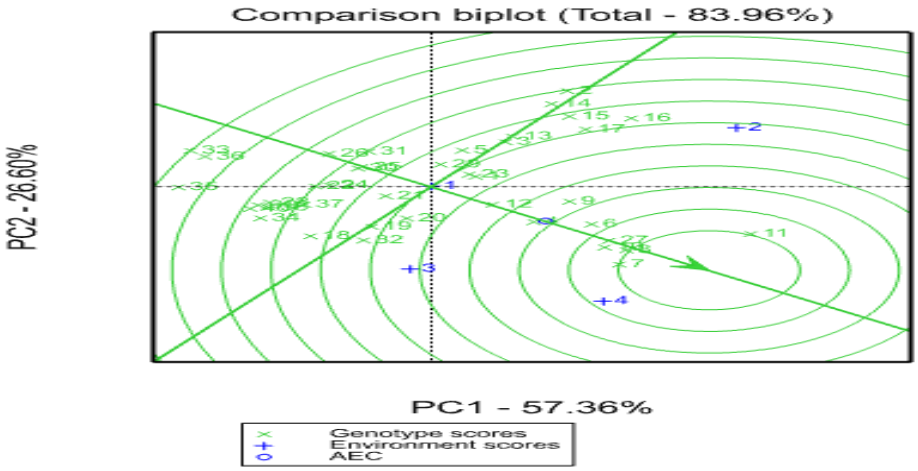

FIGURE 1. GGE biplot base on the comparison of environment relative to an ideal genotype.

An ideal genotype is defined as one that is the highest yielding across test environments and it's absolutely stable in performance that ranks the highest in all test environments (Yan and Kang, 2003) it should also possess both high mean performance and high stability within a mega-environment (Yan *et al.*,2007). Although such an ideal genotype may not exist in reality, it could be used as reference for genotype evaluation (Mitrovic et, al, 2012).

In Figure 3, a genotype is more desirable if it is located closer to ideal genotype (Kaya et al, 2006) the closer the genotype are G7, G8 and G11. Favorable genotypes are G10, G27, and G6. The ideal test environment should have large PC1 scores and small PC2 scores. Thus, using the ideal environment as the center, concentric circles were drawn to help visualize the distance between each environment as the ideal environment. (Yan et al 2000).

Figure 3. Indicated that E4 which fell near the center of concentric circles was an ideal test environment in terms of being the most representative of the overall environment and the most powerful to discriminate genotypes. Favorable environment is E2, while unfavorable environment is E1 and E3.

Yield performance and stability of genotypes were evaluated by an average environment coordination (AEC) method (Yan, 2001; Yan and Hunt, 2002; Yan, 2002). In this method, an average environment is defined by the average PC1 and PC2 scores of all environments, represented by a small circle (Figure 4). A line is then drawn to pass through this average environment and the biplot origin; this line is called the average environment axis and serves as the abscissa of the AEC. The ordinate of the AEC is the line that passes through the origin and is perpendicular to the AEC abscissa (Figure 4). Unlike the AEC abscissa, which has one direction, with the arrow pointing to greater genotype main effect, the AEC ordinate is indicated by a thick line or double arrows, and either direction away from the biplot origin indicates greater GEI effect and reduced stability. The AEC ordinate separates genotypes with below-average means from those with above-average means. Furthermore, the average yield of genotypes is approximated by the projections of their markers to the AEC abscissa. Figure give genotypes with above-average means were from G11 to G15, while genotypes below-average means were from G1 to G33. The length of the average environment vector (the distance from biplot origin and the average environment marker), relative to the biplot size, is a measure of the relative importance of genotype main effect vs. GEI. The longer it is, the more important is the genotype main effect, and the more meaningful the selection based on mean performance. For this study, the length of the average environment vector was sufficient to select genotypes based on yield mean performances. Genotypes with above-average means (i.e. from G11 to G15) could be selected, whereas the rest were discarded. On the other hand, genotype stability is very important, in addition to genotype yield mean. A longer projection to the AEC ordinate, regardless of the direction, represents a greater tendency of the GEI of a genotype, which means it is more variable and less stable across environments or vice versa. For instance, genotypes G11, G7, G10 and G8 were more stable as well as high

yielding. Conversely, G32, G15 and G16 were more variable, but high yielding. An ideal genotype should have the highest mean performance and be absolutely stable (i.e. perform the best in all environments). Such an ideal genotype is defined by having the greatest vector length of the high yielding genotypes and with zero GEI, as represented by an arrow pointing to it (Figure 4). Although such an ideal genotype may not exist in reality, it can be used as a reference for genotype evaluation. A genotype is more desirable if it is located closer to the ideal genotype. Thus, using the ideal genotype as the center, concentric circles were drawn to help visualize the distance between each genotype and the ideal genotype.

To rank the genotypes based on their performance in an environment, a line is drawn that passes through the biplot origin and the environment. This line is called the axis for this environment, and along it is the ranking of the genotypes. Figure 5 ranks the genotypes based on performance in E2. Genotypes G20 to G35 had lower than average yield, G27, G7, G8, G10 and G11 had near average yield, and all others had higher than average yields. The highest yielder in E2 was G16 and G11, and the lowest yielder G35.

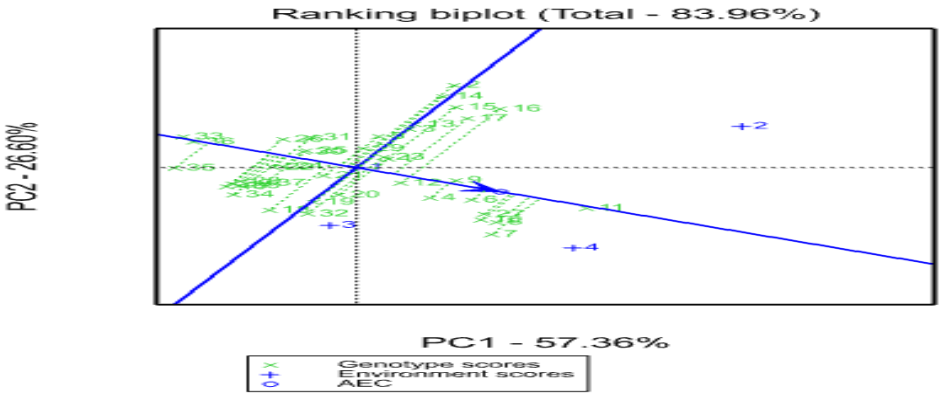

FIGURE 2. Environmet focused scaling

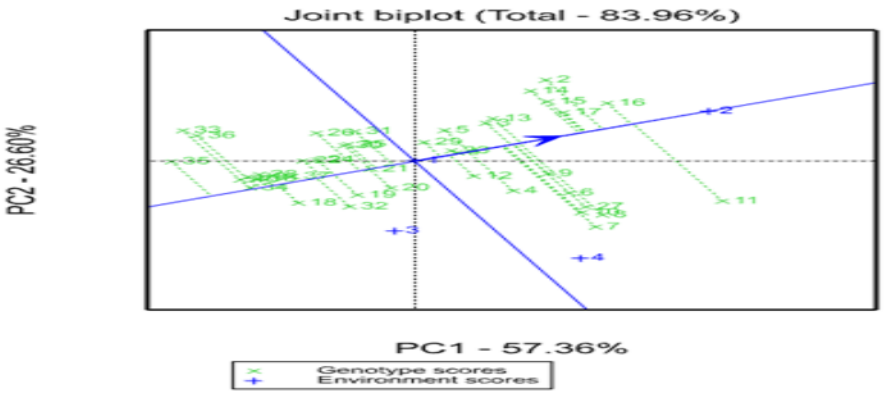

FIGURE 3. Genotype focused scaling.

TABLE 2: Environment means and scores

Environment	NE	Em	IPCAe[1]	IPCAe[2]
E1	1	487.1	-20.37803	32.96391
E2	2	1442.6	52.04641	12.37874
E3	3	599.3	-33.90875	-3.57007
E4	4	928.3	2.24037	-41.77257

Table 3: Environment means and variances

Location	No observed	Mean	Variance
1	120	487.1	116181
2	120	1442.6	535156
3	120	599.3	133275
4	120	928.3	333216
Margin	480	864.3	415757

Table 3 of first four AMMI selections per environment

Number	Environment	Mean	Score	1	2	3	4
2	E2	1442.6	52.05	G16	G11	G2	G14
4	E4	928.3	2.24	G11	G7	G8	G27
1	E1	487.1	-20.38	G19	G37	G18	G14
3	E3	599.3	-33.91	G19	G18	G37	G7

VII. CONCLUSION AND RECOMMENDATION

The application of AMMI and GGE biplot to millet multi-environmental grain yield trial facilitated the visual comparison and identification of the winning genotype in relation to the test environment, Based on the two analysis AMMI and GGE-biplot models, G11, G10, G7 and G8 characterized by high yield and stability, therefore, the G11 which is close to the ideal genotype, Two genotypes, G11and G7 were found suitable and adaptable for planting in E4 where as G11 is best in E2 and E4. AMMI analysis indicated that these two genotypes were able to produce high more stable as well as high yielding.Nevertheless, these two genotypes are to be recommended for specific planting at Mali and Senegal for their best shoot tips yield. Agriculturalist, policy markers have to search for genotypes that are stable and adaptable to E1 and E3.

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