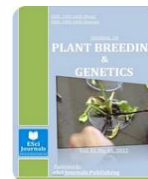




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MULTI-ENVIRONMENT TRIAL ANALYSIS OF FOOD BARLEY IN ETHIOPIA USING AMMI AND GGE BILOT METHODS

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ABSTRACT

Food barley released varieties were tested in 2012 for performance across major environments in Ethiopia consisting of 12 varieties Diribe, Tilla, Abbay, Biftu, Defo, Dinsho, Mulu, Setegn, Misiratch, Basso, Mezezo and local checks over six locations Gergera, Estayish, Shambu, Arjo, Robe and Sinana. The objective was to determine genotype by environment interaction using AMMI and GGE biplot, compare the two models for identifying the adaptable and stable genotypes. Sinana was identified as the high yielding environment and MULU the high yielding variety with mean yields of 3466.31 and 3137.67 kg/ha, respectively. The mean yield at Estayish was lower (1535 kg/ha) than other environments whereas lower yield (2212.16 kg/ha) was also obtained from the variety DINSHO. The AMMI analysis of Variance indicated that 47% of the total sum of squares is attributed to the Environmental effect, 8% to the genotypic effect and 25% to the interaction. The first three principal components of the GEI explained 81% of the variation. Genotypes Basso, Biftu and Setegn were the most stable whereas Diribe was unstable. Variety Mulu was identified as the winner genotype by AMMI model whereas Diribe was identified as the winner by the GGE model. GGE model better explains the which-won-where scenario and hence preferred to AMMI model. The discriminating and representative view of the GGE biplot depicted that Sinana and Shambu are discriminating environments whereas Sinana, Estayish and Gergera are representative environments. Therefore, Sinana is the ideal environment for discriminating genotypes and representing other environments for selecting ideal genotypes.

Keywords: Multi-environment, GGE biplot, stability, AMMI, interaction, adaptability.

INTRODUCTION

Barley is a cool-season crop which grows at altitudes of about 3000 meter above sea level and commonly cultivated in stressed areas where soil erosion, occasional drought or frost limits the growth of other crops (Bekele et al., 2005). Ethiopia is the second largest barley producer in Africa, next to Morocco, accounting for about 25 percent of the total barley production in the continent (FAO, 2014). Barley production and consumption has a longstanding tradition in Ethiopia where the country is considered the centre of diversity

or secondary origin of the crop with more than 15,000 accessions conserved in the gene bank.

In plant breeding programs, genotypes are evaluated in multi-environment trials (METs) by testing their performance across environments and selecting the best genotypes in specific environments. However, the selection of superior genotypes in multi-environment trials usually results in genotype-by-environment interactions that often complicate the interpretation of results obtained and reduce efficiency in selecting the best genotypes (Annicchiarico and Perenzin, 1994).

This interaction is due to the changes in genotype's relative performance across environments, as a result of differential responses of the genotypes to various abiotic and biotic factors (Dixon and Nukene, 1997). Hence, a

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significant Genotype by Environment (GE) interaction for a quantitative trait like grain yield can complicate the identification of superior genotypes for both improved crop development and new crop introduction.

Statistical techniques have been proposed to facilitate the interpretation of GEI from MET's. The most commonly used statistical methods for analyzing GEI is the two-way cross-classification analysis of variance (ANOVA). However, while this technique can adequately explain only the main effects and identify GEI as a source of variation, it fails to analyze the inherent effects of GEI. This is due to the additive nature of the ordinary ANOVA model does not allow it to analyze a non-additive interaction component and other statistical approaches are therefore required to identify the relationships of interaction.

Gauch and Zobel (1988) compared the performance of the ANOVA method with the regression method and found that ANOVA fails to detect a significant interaction component and the regression approach accounts for only a small portion of the interaction sum of squares only when the pattern fits a specific regression model.

The AMMI model has been suggested to be an effective method because it captures a large portion of the GE sum of squares and uniquely separates main and interaction effects as required for most agricultural research purposes (Gauch, 2006). It has proved to be a powerful tool used by researchers to evaluate a number of genotypes established in a number of environments, identify stable and adaptable genotypes and determine the magnitude of GEI (Crossa, 1990). As a result, Grüneberg et al. (2005) reported that the AMMI model was a highly efficient multivariate tool for the analysis of MET data. Likewise, the most well-known and appealing component of AMMI analysis is the graphical display of the results in a very informative biplot (AMMI1) which shows both main and interaction effects for both genotype and environment (Zobel et al., 1988). Yet, the AMMI1 biplot does not have the most important property of a true biplot, namely the inner-product property. In addition, the AMMI1 biplot does not display the discriminating ability and representativeness view of a biplot which is effective in evaluating test environments. This has been recognized by Yan et al. (2000) who adopted the proposal of Gabriel (1971) by using the biplot technique to display the genotype main effect plus genotype-by-environment interaction (G+GE) of a METs data and called it the GGE biplot.

GGE biplot is a graphical tool which displays, interprets and explores two important sources of variation, namely genotype main effect and GE interaction of MET data (Fan et al., 2007; Yan et al., 2000). GGE biplot analysis considers that only the G and GE effects are relevant and that they need to be considered simultaneously when evaluating genotypes. The GGE biplot has therefore been used in crop variety trials to effectively identify the best-performing genotype across environments, identify the best genotypes for mega-environment delineation, whereby specific genotypes can be recommended to specific mega-environments and evaluate the yield and stability of genotypes (Yan and Kang, 2002; Yan and Tinker, 2006). The relative versatility of the GGE biplot, especially in mega-environment analysis and genotype selection, is worthy of being exploited for selection of genotypes for specific environments. More importantly, it would assist in guiding the direction of varietal development for stable ecology-based selections.

The differences between the GGE biplot and AMMI methods are; firstly, AMMI stands for the additive main effect and multiplicative interaction (Gauch Jr, 1992), and GGE stands for genotype main effect plus GE interaction (Ma et al., 2004). Secondly, the GGE biplot analysis is based on the environment-centred principal component analysis (PCA), whereas AMMI analysis is established on double centred PCA (Kroonenberg, 1995). However, according to Yan and Tinker (2006), AMMI could be misleading if used for the purpose of "which-won-where" (i.e., identification of mega-environments as well as their winning genotypes). Also, Ding et al. (2007) asserted that the GGE biplot is superior to the AMMI, because it provides a lot of more visual interpretations than the AMMI, by allowing the visualization of any crossover GE interaction which is usually essential to breeding programs.

Several multi-environment trial studies have compared the AMMI and GGE biplot analyses to obtain an effective tool for analyzing GEI and have come out with differing results. Kandus et al. (2010) found the AMMI model was the best model to describe the GEI in maize. Stojaković et al. (2010) and Stojaković et al. (2010) also found out that the models provided similar results. Moreover, Rad et al. (2013) indicated that both models performed equally using data on bread wheat while Samonte et al. (2005) found the AMMI and GGE biplot analyses complementing one another. Contrary to these findings, Yan et al. (2007) compared the GGE biplot and AMMI

analyses and concluded that the GGE biplot was superior to the AMMI biplot in mega-environment analysis and genotype evaluation. The main objectives of this study were therefore, to determine the magnitude and patterns of G×E interaction effects in food barley using the AMMI and GGE biplot methods, to observe mean performance and stability of 12 food barley genotypes and to compare GGE biplot and AMMI analysis and determine the most suitable method for evaluating MET of food barley in Ethiopia.

MATERIALS AND METHODS

The experiment was conducted at different locations representing the south-eastern, North-eastern and western parts of Ethiopia in 2012 to evaluate the released varieties of food barley for yield and stability across contrasting environments in major parts of barley growing areas in Ethiopia. The locations were Gegera and Estayish in North Wolo Zone of Amhara region in Northeastern Ethiopia, Arjo and Shambu in East and Horoguduru Wolega Zones of Oromia Region in the Western Ethiopia and Robe and Sinana in Bale Zone of Oromia Region in the Southeastern part of Ethiopia. The varieties included 11 improved varieties of food barley such as Diribe, Tilla, Abay, Biftu, Defo, Dinsho, Mulu, Setegn, Misirach, Basso, Mezezo and local checks. The trials were laid out in Randomized Complete Block Design with three replications. The plot size was 1.2 x 2.5m (3m²) with row spacing of 20 cm. the central four out of the six rows were used as harvestable plots for yield and then converted to the hectare. All the agronomic and pest management practices were employed as per the recommendation in the areas.

Additive Main Effect and Multiplicative Interaction (AMMI) Method: The additive main effects and multiplicative interaction (AMMI) method integrates analysis of variance and principal component into a unified approach (Bradu and Gabriel, 1978; Gauch, 1988). AMMI method first fits the additive main effects of genotypes and environments by the usual analysis of variance and then describes the non-additive part, genotype by environment interaction, by principal component analysis. Stable genotypes for each environment were selected by AMMI and principal component axes (PCAs) were extracted and statistically tested by Gollob (1968) F-test procedure (Vargas and Crossa, 2000). These components from AMMI analysis were used to obtain a biplot of the main effect of means versus the first Interaction Principal Component

Analysis Axis (IPCA1). IPCA1 the pattern of response of G, E, and GEI were then identified. The AMMI equation according to Gauch and Zobel (1988) for T genotypes and S environments is;

$$Y_{ij} = \mu + g_i + e_j + \sum_{k=1}^n \lambda_k \alpha_{ik} \gamma_{jk} + \epsilon_{ij}$$

Where, Y_{ij} is the mean yield of the i^{th} genotype in the j^{th} environment; μ is the general mean g_i is the i^{th} genotypic effect; e_j is the j^{th} location effect; λ_n is the eigenvalue of the PCA axis n; α_{in} and γ_{jn} are the i^{th} genotype j^{th} environment PCA scores for the PCA axis n; θ_{ij} is the residual; n' is the number of PCA axis retained in the model. Therefore, the interaction effect can be calculated as;

$$(txE)_{ij} = y_{ij} - y_{i.} - y_{.j} - y_{..}$$

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$$(GxE)_{ij} = y_{ij} - y_{i.} - y_{.j} - y_{..}$$

GGE Biplot Method: Mathematically, biplot may be regarded as a graphical display of matrix multiplication and for this study, the GGE biplot method outlined by Yan (2002) was used to display the G and GE interaction patterns in the data. The which-won-where pattern (which is an intrinsic property of the GGE biplot rendered by the inner-product property of the biplot, of food barley genotype by environment data set was also visually presented. Additionally, GGE biplot was used to identify high yielding and adapted barley genotypes as well as suitable test environments. The best barley genotypes were represented by large principal component scores, PCA1 (high grain yield) and small principal component scores, PCA2 (high stability) (Yan, 2001). Genotypes that had PCA1 scores greater than 0 were identified as higher yielding and those that had PCA1 scores less than 0 were identified as lower yielding. PCA1 scores greater than 0 detected the genotypes of interest (i.e. adaptable or higher yielding), while PCA1 scores less than 0 discriminated the non-adaptable ones (Zerihun, 2011). PCA2, which was related to genotypic stability or unstable, divided the genotypes of interest based on their scores. The model for the GGE biplot based on singular value decomposition (SVD) of the first two principal components is:

$$Y_{ij} - \mu - \beta_j = \lambda_1 \xi_{i1} \eta_{j2} + \varepsilon_{ij}$$

Where Y_{ij} is the measured mean of genotype i in environment j , μ is the grand mean, β_j is the main effect of environment j , $\mu + \beta_j$ being the mean yield across all genotypes in environment j , λ_1 and λ_2 are the singular values (SV) for the first and second principal component (PCA1 and PCA2) respectively, ξ_{i1} and ξ_{i2} are are eigenvectors of genotype i for PCA1 and PCA2 respectively, η_{1j} and η_{2j} are eigenvectors of environment j , for PCA1 and PCA2 respectively, ε_{ij} is the residual associated with genotype i in environment j .

Data analysis: data were subjected to analysis after checking for required assumptions of normality, homogeneity of variance using respective tests. The analysis was performed using R statistical software.

RESULTS

The mean yields of 12 barley genotypes grown in six environments are presented in Table 1. On average, grain yield of genotypes ranges from 2212.16 kg ha⁻¹ for DINSHO genotype to 3137.67 kg ha⁻¹ for MULU

genotype. In addition to these, half of the genotypes had mean above the average grain yield while the other half were below the average. Among the environments, Arjo, Estayish and Robe had below the mean average yields. The highest yields were recorded for environments Sinana followed by Gergera with mean yields of 3466.306 and 3322.4 kg ha⁻¹, respectively. Varieties in figure 1 exhibit interactions across the test environments with the possible existence of crossover interaction. This indicates that the data signifies a remarkable genotype by environment interaction (GEI) and require further investigation to understand the magnitudes and patterns of interactions.

The analysis of variance for grain yield (kg ha⁻¹) of twelve barley genotypes tested in six environments showed 47.29% of the total sum of squares was attributable to environmental effects, 8.14% to genotypic effects, and 25.87% to G x E interaction as shown in Table 2. The analysis revealed that variances due to environments, genotypes and GEI interactions were highly significant ($P < 0.01$). The large variation due to the environment is an indication of diversity among environments and the highly significant variation of GEI is an indication of changes in the rank of genotype performance across environments.

AMMI Model: The first three interaction principal components were highly significant ($p < 0.01$) which implied that the interaction of barley genotypes with six environments was predicted by the first three components of genotypes and environments. The results further indicated that the first two interaction principal components (IPCA1 and IPCA2) were very important in explaining the interactions while the third (IPCA3) less significant compared with IPCA1 and IPCA2, and the rest IPCA's were not significant. IPCA1 explained 47 % of the variability relating to GEI while IPCA2 explains 32.6% of the GE interaction. Both IPCA1 and IPCA2 comprise 79.6 % variations in the GE interactions.

The AMMI biplot analysis for barley yield grown in six environments was presented in Figure 2. The x-axis shows the main effects while the y-axis shows the first PCA axis and revealed differential responses of genotypes to the study environments. The figure depicts that LOCAL_CHECK, BASSO and MEZEZO were close to the origin (low IPCA1 score) and hence considered stable genotypes compared with others.

DAFO had the largest positive interaction scores while TILA had a relatively negative interaction. Environment Estayish and Robe were positively related to the interaction while Shambu is negatively related to

poorly performing environment. Gergera, Sinana and Arjo can be considered stable environments. Among the group of genotypes which had a negative interaction, MULU had a relatively high mean yield.

Table 1. Average grain yield (kg ha⁻¹) of 12 barley genotypes in 6 environments compared with the rest of the environment.

| Genotypes | Environments | | | | | | mean |
|-------------|--------------|---------|---------|----------|----------|----------|---------|
| | Arjo | Shambu | Gergera | Estayish | Sinana | Robe | |
| DIRIBE | 2785.690 | 1901.56 | 3105.0 | 776.667 | 4842.583 | 1666.583 | 2513.01 |
| TILA | 2483.767 | 4520.28 | 2307.5 | 975.167 | 3676.667 | 1823.667 | 2631.17 |
| ABBAY | 2015.623 | 2991.05 | 3268.8 | 1908.00 | 3948.833 | 2469.500 | 2766.97 |
| BIFTU | 1998.387 | 2969.69 | 3654.7 | 1918.67 | 4137.833 | 2986.167 | 2944.23 |
| DAFO | 2154.810 | 1569.12 | 3341.0 | 1337.83 | 2828.417 | 2810.750 | 2340.32 |
| DINSHO | 1720.723 | 1885.81 | 3006.8 | 1596.33 | 2783.500 | 2279.750 | 2212.16 |
| MULU | 2589.830 | 4462.61 | 3990.5 | 2138.50 | 3463.000 | 2181.583 | 3137.67 |
| SETEGN | 1906.367 | 2510.36 | 3124.7 | 1234.33 | 3222.833 | 2332.083 | 2388.44 |
| MISRATCH | 1644.467 | 4115.41 | 4030.2 | 933.167 | 3682.333 | 1805.250 | 2701.80 |
| BASSO | 1785.947 | 2992.74 | 3676.5 | 1330.17 | 2824.667 | 2368.333 | 2496.39 |
| MEZEZO | 2260.080 | 3283.88 | 2854.5 | 2372.50 | 3469.667 | 2411.083 | 2775.29 |
| LOCAL_CHECK | 1761.570 | 3104.80 | 3508.7 | 1898.67 | 2715.333 | 2611.417 | 2600.08 |
| Mean | 2092.272 | 3025.61 | 3322.4 | 1535.00 | 3466.306 | 2312.181 | 2625.63 |
| CV% | 23.90656 | 26.3951 | 19.532 | 32.6474 | 14.85685 | 11.17023 | |

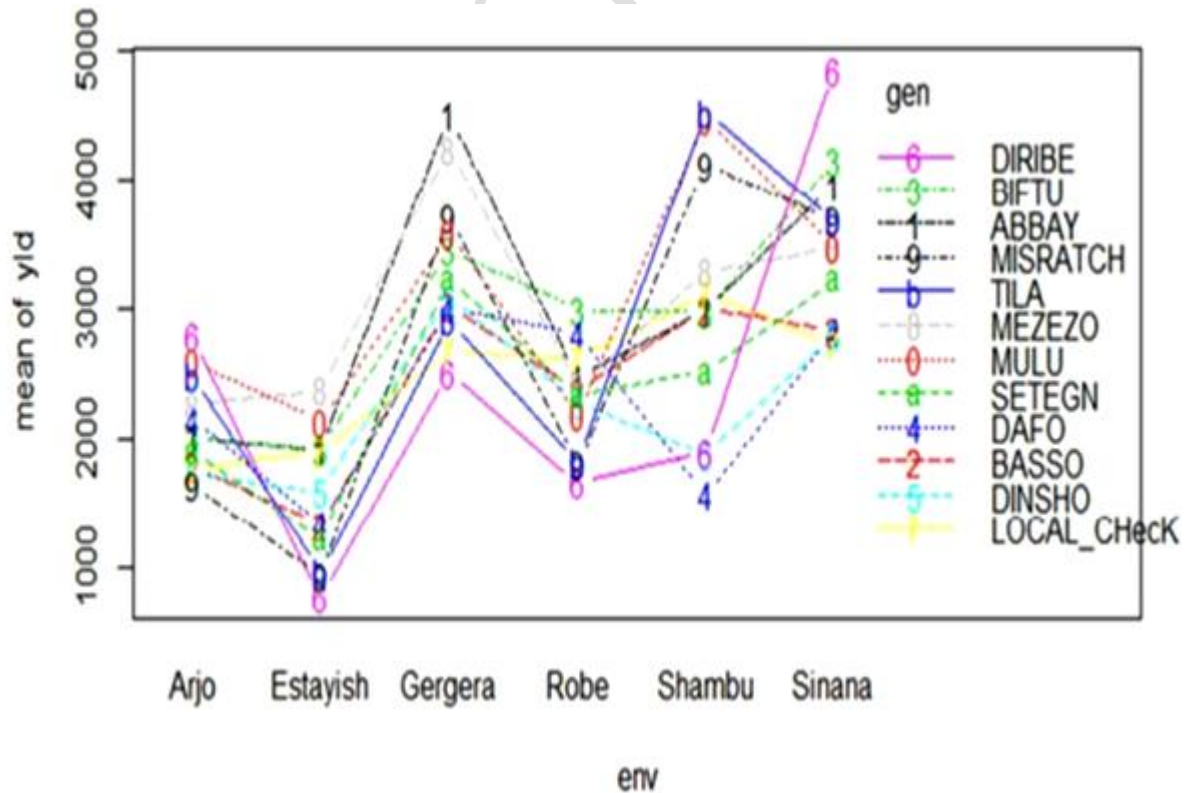


Figure 1. Average yield performance 12 barley genotypes across six environments.

Table 2. Additive main effect and multiplicative interactions (AMMI) analysis of variance for barley grain yield (kg ha⁻¹) across environments.

| Source | DF | Sum of Squares | Mean of Square | Variation Explained (%) |
|-----------------|-----|----------------|----------------|-------------------------|
| Environment (E) | 5 | 105278618 | 21055723.60** | 47.29 |
| Genotype (G) | 11 | 18128643 | 1648058.46** | 8.14 |
| G x E | 55 | 57580614 | 1046920.26** | 25.87 |
| IPCA 1 | 15 | 27074627 | 1804975.13** | 12.16 |
| IPCA 2 | 13 | 18760617 | 1443124.39** | 8.43 |
| IPCA 3 | 11 | 7615573 | 692324.82* | 3.42 |
| Residual | 132 | 41626631 | 315353.27 | 18.70 |
| Total | 215 | 222614506 | 1035416.31** | |

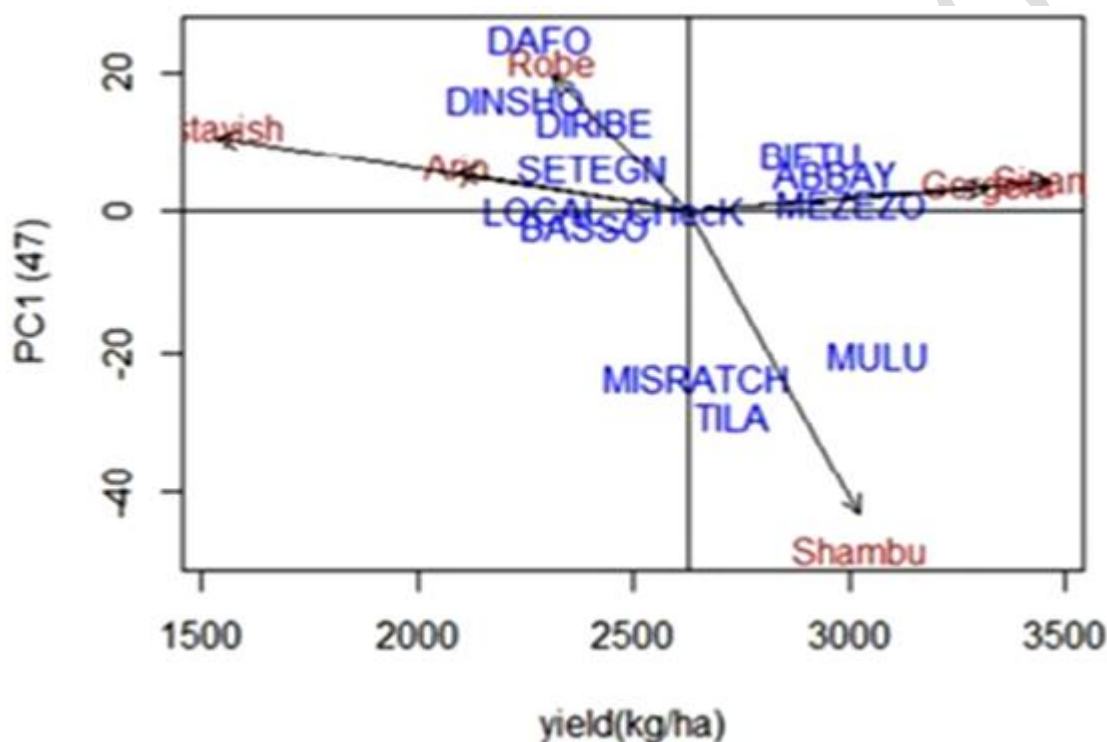


Figure 2. AMMI biplot for 12 barley genotypes across 6 environments.

GGE Biplot Analysis The partitioning of GGE through GGE biplot analysis showed that PCA1 and PCA2 accounted for 45.76% and 25.22% of GGE sum of squares respectively for grain yield, explaining a total variation of 70.98 as shown in Figure 3. The GGE biplot revealed the best genotypes under different environments and identified genotype DIRIBE as the best genotype in the environments Sinana and Arjo. Genotype MEZEZO was best for environment Gergera, Estayish and Robe while MULU and MISRATCH for Shambu. Genotype DIRIBE gave the highest average yield (largest PCA1 scores), but was unstable over the

environments, due to its high absolute PCA2 scores. In contrast, BIFTU yielded poorly in all environments, as indicated by its small PCA1 scores (low yielding) and relatively small PCA2 scores which make it relatively stable genotype. The average yield of genotypes MULU, MISRATCH, TILA, MESEZEZO and ABBAY were below the mean average (PCA1 scores < 0), as shown in Figure 3, and were thus classified as the non-adaptable genotypes. On the other hand, genotypes DIRIBE, DAFO, and DINSHO, with PCA1 scores > 0 were detected as the genotypes of interest (i.e. adaptable or higher yielding). Figure 4 shows GGE of genotypes for both average yield

and stability performance over environments. The line passing through the biplot origin is called the average environment coordinate (AEC). Closer to concentric

circle indicates higher mean yield. The line which passes through the origin and is perpendicular to the AEC with double arrows represents the stability of genotypes.

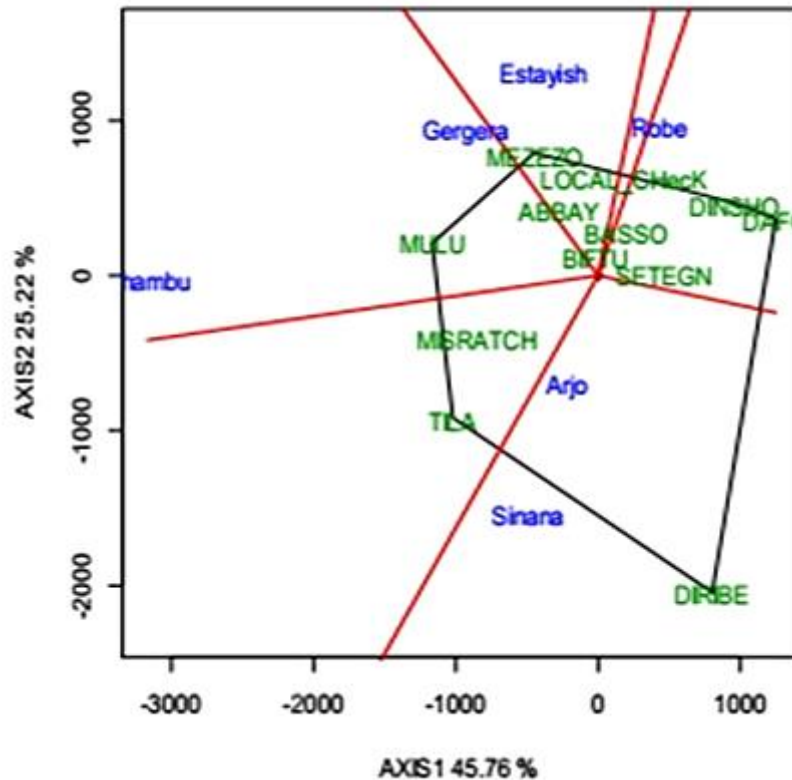


Figure 3. Which-won-where/what of GGE biplot graph.

However, DIRIBE which had the longest projection from the AEC x-axis was highly unstable genotype whereas BASSO, BIFTU and SETEGN were very stable genotypes compared to the others. The double arrowed line also separated genotypes with the above average mean yield (DIRIBE, DAFO, DINSHO, SETEGN, BASSO and LOCAL_CHECK) from genotypes with the below average mean yield (MULU, MISRATCH, TILA, MEZEZO, ABBAY and BIFTU).

The discriminating power versus representativeness view of the GGE biplot as shown in Figure 5 showed that test environments Shambu and Sinana with the longest projection from the biplot origin were found to be the environments with more discriminating power that they

provided much information about the differences among genotypes. On the other hand, Arjo, with its shortest vector from the biplot origin, was found less discriminating of the test genotypes. Test environments Gergera, Estayish and Sinana were found to be more representative of other test environments due to the fact that they have smaller angles with the Average Environment Axis (AEA). Sinana was therefore identified as an ideal environment that has both discriminating abilities of the genotypes and representative of the other test environments. Thus, environment Sinana can be used to effectively select superior barley genotypes that can perform consistently across environments.

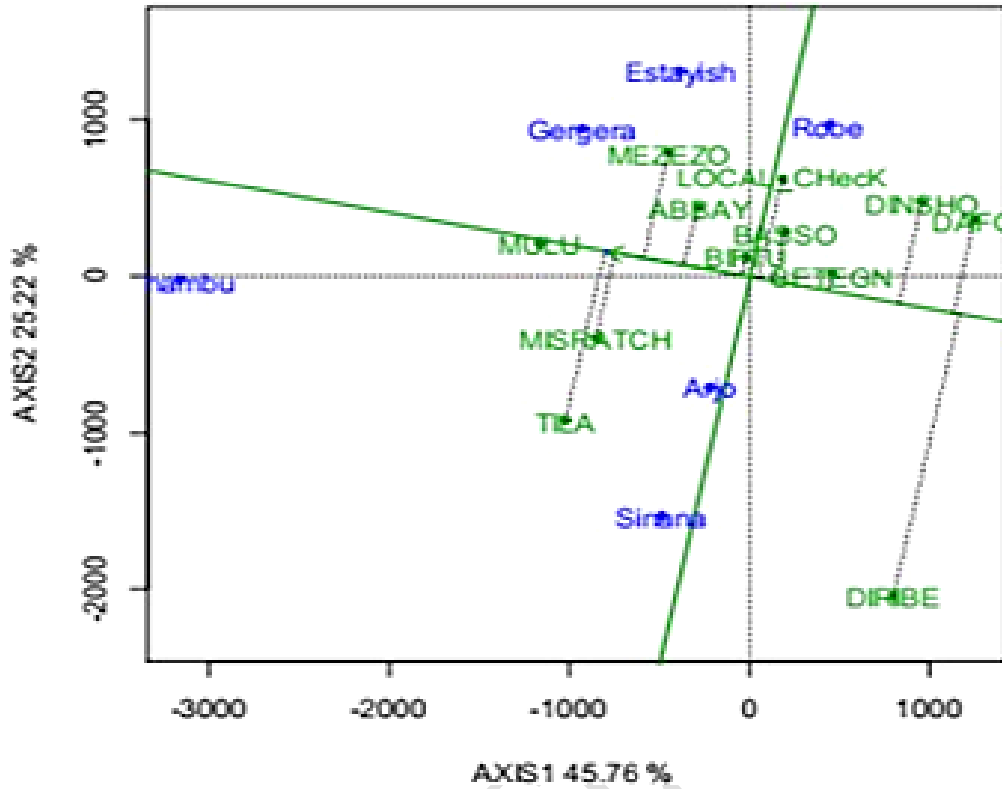


Figure 4. Mean vs Stability of the GGE biplot.

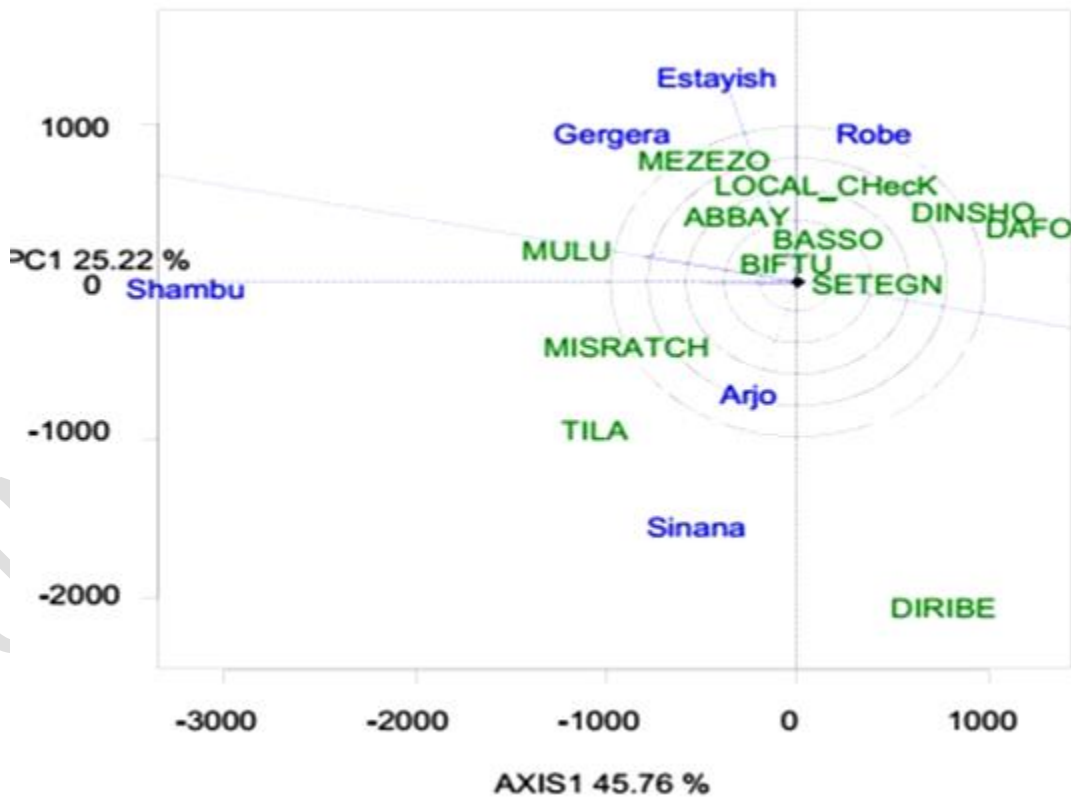


Figure 5. Discriminativeness vs Representativeness of the GGE biplot graph.

DISCUSSION

This study revealed that GEI is a significant source of variation in the barley multi-environment trial. The observed pattern of GE interaction for grain yield of barley suggests that genotypes respond differently in different environments, hence the need for biplot analysis which allows visual interpretation of GE interaction and facilitates genotype recommendations in MET. Subsequently, two types of biplots (AMMI1 and GGE) were used to graphically display, interpret and explore important sources of variation, namely genotype main effect and GE interaction of MET data, to identify the genotypes which were superior or had adapted well in each environment based on their mean performance and stability and also to evaluate test environments for effective genotype evaluation based on their discriminating ability and representativeness.

In this study, AMMI1 explained 47% of the variation while GGE biplot explained 70.98% of the G+GE total variance. In this study, the GGE biplot explains more variation than AMMI1 and hence, considered as an accurate presentation of G+GE of the mean yield of barley data. This may be due to the fact that AMMI1 biplot (Zobel et al., 1988) has been proven to be very efficient in detecting important sources of variation of GE interaction effects and has also been adjudged as either superior or equal to GGE biplot analysis (Gauch, 2006), it is not able to effectively display the relative performance of each genotype in each environment (i.e., does not have the most important property of a true biplot, which is the inner-product property). Therefore, the performance of a given genotype in a given environment cannot be accurately visualized even if it fully displays the data. In mega-environment analysis and genotype evaluation, GGE biplot is superior to AMMI1 bi-plot Yan et al. (2007) while the AMMI1 biplot is better viewed as a tool for presenting conclusions rather than as a tool for discovering which-won-where patterns. But GGE was criticized by Gauch (2006) for not being able to reveal which-won-where patterns if more than two PCs are required to approximate the data.

With regard to visualizing the mean performance and the stability of the genotypes simultaneously, AMMI1 biplots identified MULU as the highest yielding genotype showing high absolute interaction with Shambu environment. But according to GGE biplot, DIRIBE was identified as highest yielding genotype. In addition, BASSO was considered as the most stable genotype

though not highyielding by both biplots.

Evaluation of the test environments for effective selection of superior genotypes is one of the most important features of GED and biplot analysis. Yet, the AMMI1 biplot (Zobel et al., 1988) displays the test environments by their main effects E and IPC1 scores, but provides no information on the environment's ability in identifying superior genotypes, only the GGE biplot is able to optimize genotype selection based on its discriminating ability and representativeness view (Yan et al., 2007). Thus, the GGE biplot was able to identify Shambu as the ideal environment having a long vector length (discriminating ability) and a small angle (representativeness) to the average environment axis (AEA) and selecting MULU as a superior genotype that can perform consistently across good environments.

CONCLUSIONS

In general, this study revealed that GGE and AMMI1 biplots are useful techniques to detect the existence of GE interaction between twelve food barley genotypes across six environments. AMMI model identifies MULU as the winner genotype while GGE model identifies DIRIBE as the winner genotype. Since GGE biplot explains more variation in the interaction effect, it may be preferred than AMMI biplot. In both models, MULU and DIRIBE perform better than the LOCAL_CHECK.

The performance of a given genotype in a given environment was more accurately displayed by the GGE biplot compared to the AMMI1 biplot. The reason for this assertion is that, the which-won-where view of the GGE biplot proved to be a more effective visual tool in the mega-environment analysis and genotype evaluation, because it explained more G+GE and depicted the inner product property of a biplot whereas the AMMI1 biplot provides no information on the environment's ability in identifying superior genotypes: only the GGE biplot is able to optimize genotype selection based on its discriminating ability and representativeness view. Although both methods have proved to be important tools that can be used to effectively analyze and interpret GE interactions, the GGE biplot analysis provides a better innovative approach to the interpretation of genotype by environment interactions and this will enable breeders to effectively design the dissemination strategy for new barley genotypes.

ABBREVIATIONS

AMMI, Additive main effect and multiplicative interaction; GGE, genotype and genotype by

environment interaction.

COMPETING INTERESTS

The authors declare that they have no competing interests.

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AVAILABILITY OF DATA AND MATERIALS

All datasets and software used for supporting the conclusions of this article can be available upon request of the funding institutes.

AUTHORS' CONTRIBUTIONS

Girma Fana designed, coordinated this research and drafted the manuscript. Diriba Tadese carried out data analysis. Hiwot Sebsibe carried out the experiment. Ramesh Verma assisted in germplasm provision and manuscript edition. The authors read and approved the final manuscript.

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