Abstract

The research aimed is to determine the yield stability of 15 cassava genotypes using GGE biplot. The study was conducted at five locations starting in November 2010 to August 2011. The results showed that environment, genotype, and genotype-by-environment interactions significantly affect the yield. Environment gives the most effect (64.69%), followed by genotype-by-environment interaction effect (6.53%), and genotype effect (4.94%). CMM 03038-7 has the highest yield among other clones, and higher than the controls clones (UJ5, Malang 6, and Adira 4), but not significantly different from Malang 4. GGE biplot identified that CMM 03038-7 is the most stable clones with high yield.

Keywords: multi-environment trial; genotype-by-environment interaction; stability analysis; GGE biplot

Nomenclature

GGE  Genotype + Genotype-by Environment
CMM  Cross Manihot Malang
GEI  Genotype-by-Environment Interaction
PC   Principle Component
SVD  Singular Value Decomposition
AMMI Additive Main Effect dan Multiplicative Interaction

* Corresponding author. Kartika Noerwijati Tel.: +62-34-180-1468; fax: +62-34-180-1496.
E-mail address: tika_iletri@yahoo.com

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1. Introduction

Approximately 70% of cassava production in Indonesia is edible to food. Cassava demand has increased with the development of industries that use cassava as raw materials including bioethanol industry. The efforts to increase cassava production must be done so that the need of cassava as a food was not bothered where cassava is a priority commodity in the food diversification program to support food security. Cassava demand has increased 5% to 7% per year. This can be achieved through increased productivity of 3% to 5% per year and the expansion of planting area approximately 10% to 15% per year [1].

Cassava tuber yield is strongly influenced by environmental conditions, varieties, and harvesting time. The magnitude of genotype-by-environment interaction (GEI) is the cause of the cassava clones should be planted in a suitable environment so that the potential yield of a genotype to be maximum. Genotype testing in various environments will help to identify cultivar with broad adaptation and specific adaptations [2].

There are three types of GEI, i.e (1) no interaction, (2) no crossover intercation, and (3) crossover interaction [3]. Crossover interactions are the most important for breeders as they imply that the choice of the best genotype is determined by the environment [4]. Only crossover interaction affects genotype selection. To determine crossover interaction, both genotype (G) and GEI must be considered [3].

Multilocation trials is very important to be done with the purpose of which are (1) to compare the genotypes performance in multi environments and in the specific environment, (2) to estimate GEI component to measure its impact on the heritability and selection, (3) to select test sites and determines the environment in a broader scope, (4) to identify genotypes with specific adaptations, and (5) to determine breeding objectives [3].

GGE biplot technique is an analytical technique of GEI involving the effect of genotype (G) and the effect of genotype-by-environment (GE) [3]. The biplot is constructed by the first two symmetrically scaled principal components (PC1 and PC2) derived from singular value decomposition (SVD) of multi-environment trial data. Biplots can be multidimensional, but two-dimensional biplots, using only the first and the second PCs, are most common [5].

The analyses of GEI data include three major aspects: (1) mega-environment analysis, (2) test-environment analysis, and (3) genotype evaluation [3]. The “which-won-where” view of GGE biplot is an effective visual tool in mega-environment analysis [5]. GGE biplot technique (developed after AMMI biplot technique) has several advantages when compared with AMMI techniques include (1) the graph of the GGE biplot better than AMMI1 graph in mega-environment analysis and genotype evaluation because it explains more G + GE, and has the inner product of property of biplot, (2) the discriminating power vs representativeness view of the GGE biplot is effective in evaluations test environments, which is not possible in AMMI analysis [6].

GGE biplot can be used to analysis of the mega-environment [7,8,9], genotype evaluation [10], test-environment evaluation [7,11], and analysis of heterotic patterns [12]. These aspects cause the GGE biplot become very popular as the most comprehensive tool in quantitative genetics and plant breeding.

The commonly used GGE biplot is based on the Sites Regression (SREG) linear-bilinear (multiplicative) model [8,13]. In the SREG model, the main effect of genotype (G) plus GE interaction were absorbed into the bilinear terms [14], which are the two sources of variation that are relevant to cultivar evaluation [15]. These factors are graphically shown through a GGE biplot, which is used in the visual evaluation of both genotypes and environments [16].

The purpose of this research is to determine the stability of fresh tuber yield of 15 cassava genotypes grown in five locations using GGE biplot technique.
2. Material and method

The research was conducted in November of 2010 to August of 2011 at five locations: Kediri, Ponorogo, Probolinggo, Malang, and Mojokerto (East Java, Indonesia). The research was conducted using a randomized block design with three replications. The treatments are 15 cassava genotypes consisting of eleven cassava clones namely CMM 03025-43, CMM03036-7, CMM 03036-5, CMM 03038-7, CMM 03094-12, CMM 03094-4, CMM 03095-5, CMM 02040-1, CMM 02033-1, CMM 02035-3, CMM 02048-6, and four control varieties, namely UJ5, Malang 6, Malang 4, and Adira 4.

Cassava is planted in a plot size of 5 m × 5 m with a spacing of 100 cm × 80 cm. Cassava cuttings about 20 cm long are planted with the vertically position of cuttings. Fertilization was given twice, at one month after planting with a dose of 100 kgs ha⁻¹ Urea + 100 kgs ha⁻¹ SP36 + 100 kgs ha⁻¹ KCl, and at three months after planting with 100 kgs ha⁻¹ Urea. Weeding was performed twice, at one and three months after planting. The activities to improve the ridge were carried out before fertilization. Removal shoots with leaves two best buds performed at two months after planting. Harvesting was done when the plant was 10 months. Observations were made on fresh tuber yield per plot. Tuber yield per plot converted to fresh tuber yield per hectare.

Combined analysis of variance was performed on the fresh tuber yield data. The equation for the combined analysis of variance is

\[ \bar{Y}_{ij} = \mu + e_j + g_i + (ge)_{ij} + e_{ij} \]  

where \( \bar{Y}_{ij} \) is the yield mean of the \( i^{th} \) genotype at the \( j^{th} \) environment (\( i = 1,2,...,v \) dan \( j = 1,2,...,s \)), \( v \) is the number of genotypes, \( s \) is the number of environments, \( \mu \) is the grand mean, \( g_i \) is the genotype effect, \( e_j \) is the environment effect, \( (ge)_{ij} \) is the genotype-by-environment interaction effect, and \( e_{ij} \) is the error associated to the model.

If there was a significantly different in genotype-by-environment interaction was followed by GGE biplot analysis to assess the stability of tuber yield of 15 cassava genotypes that tested. The equation for GGE biplot is

\[ \bar{Y}_{ij} = \mu + e_j + \sum_{k=1}^{t} \lambda_k \alpha_k \gamma_{jk} + e_{ij} \]  

where \( \bar{Y}_{ij} \) is the yield mean of the \( i^{th} \) genotype at the \( j^{th} \) environment (\( i = 1,2,...,v \) dan \( j = 1,2,...,s \)), \( v \) is the number of genotypes, \( s \) is the number of environments, \( \mu \) is the grand mean, \( e_j \) is the environment effect, \( \lambda_k \) is the eigenvalue of the PCA analysis axis \( k \), \( \alpha_k \) (\( \alpha_k,\alpha_k,\ldots,\alpha_k \)) and \( \gamma_{jk} \) (\( \gamma_{jk},\gamma_{jk},\ldots,\gamma_{jk} \)) are the genotype and environment principle component scores for axis \( k \), \( t \) is the number of principle component retained in the model, and \( e_{ij} \) is the error.

3. Result and discussion

The results of the combined analysis of variance showed that environment, genotype and genotype-by-environment interactions significantly affect fresh tuber yield. Environment was the most important source for tuber yield variation (64.69%), followed by genotype-by-environment interaction effect (6.53%), and the genotype effect (4.94%) (Table 1). It showed that cassava tuber yield is strongly influenced by environmental factors. However, the large environment effect is not relevant to cultivar evaluation [3]. Only genotype (G) and interaction genotype-by-environment (GE) are relevant to cultivar evaluation. Therefore, for cultivar evaluation, it is essential to remove environment effect (E) from data and focus on G and GE. Further explained that another important point in cultivar evaluation is that G and GE effect must be considered simultaneously to make any meaningful selection decisions.

GGE biplot analysis divides the sum of squares of genotype plus genotype-by-environment interactions into several principle components that are PC1, PC2, and PC3, each of which has a proportion of 55.98%, 24.44%, and 10.27% of the sum of squares genotype and interaction. However, only the first two components (PC1 and PC2), which have a significant effect (Table 2). PC1 and PC2 values both the genotype and the environment are presented in Table3.
Table 1. Combined anova for fresh tuber yield of fifteen cassava genotypes tested at five environments.

<table>
<thead>
<tr>
<th>Source of variance</th>
<th>Df</th>
<th>Sum of Square</th>
<th>Means Square</th>
<th>F value</th>
<th>Proportion (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Env</td>
<td>4</td>
<td>49843.52</td>
<td>12460.88</td>
<td>263.46**</td>
<td>64.69+</td>
</tr>
<tr>
<td>Rep (Env)</td>
<td>10</td>
<td>2495.11</td>
<td>249.51</td>
<td>5.28**</td>
<td></td>
</tr>
<tr>
<td>Genotype</td>
<td>14</td>
<td>3806.41</td>
<td>271.89</td>
<td>5.75**</td>
<td>4.94+</td>
</tr>
<tr>
<td>Genotype-by- Env</td>
<td>56</td>
<td>5029.75</td>
<td>89.82</td>
<td>1.90**</td>
<td></td>
</tr>
<tr>
<td>Plant Number</td>
<td>1</td>
<td>134.29</td>
<td>134.29</td>
<td>2.84</td>
<td></td>
</tr>
<tr>
<td>Error</td>
<td>134</td>
<td>6337.89</td>
<td>47.30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>219</td>
<td>77050.72</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

** Siginificantly different (p < 0.01)
+ Proportion to total sum of square

GGE biplot is presented with two principle components explaining a total of 80.42% GGE variation (PC1 55.98% and PC2 24.44%) (Table 2). The first principle component (PC1) is represented in X axis. The genotype that have higher PC1 values are considered be more productive. The second principle component (PC2) is represented on the Y axis and describes the genotype stability.

Table 2. GGE analysis for fresh tuber yield.

<table>
<thead>
<tr>
<th>Principle Component</th>
<th>Eigenvalues</th>
<th>Percentage (%)</th>
<th>Porcentaje (%)</th>
<th>Probability</th>
</tr>
</thead>
<tbody>
<tr>
<td>PC1</td>
<td>5205.53 **</td>
<td>55.9805</td>
<td>55.980</td>
<td>0.00000</td>
</tr>
<tr>
<td>PC2</td>
<td>2272.55 **</td>
<td>24.4391</td>
<td>80.420</td>
<td>0.00016</td>
</tr>
<tr>
<td>PC3</td>
<td>954.89</td>
<td>10.2689</td>
<td>90.689</td>
<td>0.10670</td>
</tr>
<tr>
<td>PC4</td>
<td>734.72</td>
<td>7.9013</td>
<td>98.590</td>
<td>0.17428</td>
</tr>
<tr>
<td>Residual</td>
<td>131.13</td>
<td>1.4102</td>
<td>100.000</td>
<td>0.97117</td>
</tr>
<tr>
<td>Total</td>
<td>9298.82</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

** Significantly different (p < 0.01)

3.1. Performance of different genotypes at specific environment

Figure 1 shows the comparison of relative performance of all genotypes at S1 (Kediri). The first step is to make a straight line passing through the biplot origin to S1 marker, to make the S1 axis. The second step is to make a line was perpendicularly drawn from each genotype toward the S1 axis. The line which passed through the biplot origin and was perpendicularly to S1 axis, separates genotypes that have yield higher than the mean from genotypes that have yield lower than the mean [3, 8]. Three genotypes that have highest yield at S1 are Malang 4 (G3), CMM 03036-7 (G6), and CMM 03038-7 (G8), whereas genotypes that have low fresh tuber yield at S1 are UJ5, CMM 02048-6 (G15), CMM 03095-5, (G11), and CMM 03094-12 (G9). There are six genotypes that have fresh tuber yield above the mean, and nine genotypes below the mean (Fig.1).

The distance from the biplot origin to the marker of an environment is called the environment’s vector. The length of the vector is a measure of the environment’s ability to discriminate among genotype. The short vector, relative to the biplot sizes, implies that all genotypes tend to have similar yield in the associated environment [3].

3.2. Relative adaptation of a specific genotype across environment

One of the activities in the genotype testing was to determine which environment is most suitable for a genotype. GGE biplot can be used for this purpose. A line was drawn pass through the biplot origin and the marker of G8 (G8 is used as an example), which may be called the G8 axis. The environment are ranked along to the G8 axis in the direction indicated by the arrow. Based on Fig.2, G8 performed well at S1, followed by S4, S3, S5 and S2.
A line that is perpendicularly to the G8 vector and pass through the biplot origin separates the environment in which G8 yielded below the mean from those in which G8 yielded above the mean. At all environments, G8 always had yield above the mean of each environment (Fig. 2).

Table 3. The mean of fresh tuber yield (t ha⁻¹), PC1 and PC2 values of fifteen genotypes and five environments.

<table>
<thead>
<tr>
<th>Genotypes</th>
<th>The mean of fresh tuber yield (t ha⁻¹)</th>
<th>PC1</th>
<th>PC2</th>
</tr>
</thead>
<tbody>
<tr>
<td>UJ5 (G1)</td>
<td>25.22</td>
<td>-1.64</td>
<td>-0.68</td>
</tr>
<tr>
<td>Malang 6 (G2)</td>
<td>32.58</td>
<td>1.33</td>
<td>0.91</td>
</tr>
<tr>
<td>Malang 4 (G3)</td>
<td>37.79</td>
<td>3.37</td>
<td>-0.96</td>
</tr>
<tr>
<td>Adira 4 (G4)</td>
<td>31.51</td>
<td>0.28</td>
<td>2.35</td>
</tr>
<tr>
<td>CMM 03025-43 (G5)</td>
<td>26.91</td>
<td>-0.91</td>
<td>0.07</td>
</tr>
<tr>
<td>CMM 03036-7 (G6)</td>
<td>31.52</td>
<td>1.10</td>
<td>0.87</td>
</tr>
<tr>
<td>CMM 03036-5 (G7)</td>
<td>29.93</td>
<td>-0.82</td>
<td>2.59</td>
</tr>
<tr>
<td>CMM 03038-7 (G8)</td>
<td>37.52</td>
<td>2.69</td>
<td>0.55</td>
</tr>
<tr>
<td>CMM 03094-12 (G9)</td>
<td>24.40</td>
<td>-1.32</td>
<td>-1.64</td>
</tr>
<tr>
<td>CMM 03094-4 (G10)</td>
<td>34.55</td>
<td>1.81</td>
<td>-2.28</td>
</tr>
<tr>
<td>CMM 03095-5 (G11)</td>
<td>23.95</td>
<td>-2.24</td>
<td>-0.04</td>
</tr>
<tr>
<td>CMM 02040-1 (G12)</td>
<td>28.13</td>
<td>-0.92</td>
<td>-1.30</td>
</tr>
<tr>
<td>CMM 02053-1 (G13)</td>
<td>29.49</td>
<td>0.04</td>
<td>-0.86</td>
</tr>
<tr>
<td>CMM 02055-3 (G14)</td>
<td>24.16</td>
<td>-0.68</td>
<td>1.10</td>
</tr>
<tr>
<td>CMM 02048-6 (G15)</td>
<td>26.69</td>
<td>-2.10</td>
<td>-0.68</td>
</tr>
<tr>
<td>Kediri (S1)</td>
<td>54.84</td>
<td>4.65</td>
<td>1.76</td>
</tr>
<tr>
<td>Ponorogo (S2)</td>
<td>7.79</td>
<td>0.45</td>
<td>0.41</td>
</tr>
<tr>
<td>Probolinggo (S3)</td>
<td>31.00</td>
<td>2.25</td>
<td>2.70</td>
</tr>
<tr>
<td>Malang (S4)</td>
<td>37.08</td>
<td>3.63</td>
<td>-4.11</td>
</tr>
<tr>
<td>Mojokerto (S5)</td>
<td>18.28</td>
<td>1.28</td>
<td>0.37</td>
</tr>
</tbody>
</table>

Fig. 1. The performance of different genotypes at S1 (Kediri).

3.3. Comparison of two genotypes performance all environments

With the same principle, the performance of two genotypes can be compared on the GGE biplot. To compare two genotypes, for example, G4 and G10, a line that connected the markers of G4 and G10 was drawn, and a line that was perpendicularly to the first line that passes through the biplot origin was drawn. The perpendicular line divides the environment into two groups, with each genotype yielding better than the other within its respective side of the
perpendicular line. Fig.3 showed that one environment, S3, are on the same side with genotype G4. Thus, G4 would yield better than G10 at S3, while G10 yielded better at the other environment (namely S1, S2, S4 and S5).

Fig. 2. The relative adaptation of a specific genotype in different environment.

Fig. 3. The comparison of two genotypes.

3.4. Identification of the best genotypes for each environment

The polygon of the GGE biplot provides an effective and elegant tool for visualizing the “which-won-where” pattern. The polygon not only shows the best genotype for each test environment but also devides the test environment into several groups [3]. Two criteria are required to suggest existence of different mega-environments. First, there are different winning genotypes in different test environments. Second, the among-group variation
should be significantly greater than the within-group variation, a common criteria for clustering [17]. Graphically, different mega-environments should consist of test environments.

The polygon is drawn joining the genotype that are located farthest from the biplot origin (0,0) that served as corners of polygon, so that all genotypes are located in the polygon. Then, perpendicular line to each side of the polygon is drawn passing through the biplot origin, so the environment are divided into several sectors, each sector with different corner genotype. Within a sector, genotype which is located at the top of polygon is the best genotypes in all environments that located in the sector [5,3].

There are formed eight sectors with the corner genotypes are G8, G4, G7, G11, G15, G9, G10, and G3. Only three sectors contained environment, thus formed three mega-environments (Fig.4). Mega-environment is the group of environments that share the same best genotype(s) identified as being located at the corner of polygon [8]. Based on mega-environment that formed can be seen that Adira 4 (G4) is high yielding genotype at Probolinggo (small mega-environment). Malang 6 (G2) and CMM 03036-7 (G6) are high yielding genotypes at Kediri (S1) and Ponorogo (S2). Malang 4 (G3), CMM 03038-7 (G8), and CMM 03094-4 (G10) are high yielding genotypes at Malang (S4) and Mojokerto (S5). And vice versa, genotype and environment at two sectors that are opposite to the farthest distance showed that genotype had the largest negative interactions in the environment. For example, CMM 02048-6 (G15) has a low tuber yield at Kediri (S1) and Probolinggo (S3). CMM 03095-5 (G11) has low tuber yield in Ponorogo (S2) (Fig.4). A genotype planted outside its mega-environment frequently suffers yield reduction [17].

Deviding the target environment into different mega-environments and developing different genotypes in different mega-environment is the best way to utilize genotype-by-environment interaction [3]. Mega-environment is formed can be simple or complex [7].

![Fig. 4. The mega-environment and their winning genotypes.](image)

### 3.5. Ideal genotype identification

Estimation of yield and genotype stability was done using AEC (Average Environment Coordinate) [18], where the average environment is defined by the average values of PC1 and PC2 of all environments and is presented with a red circle on the biplot graph. A line passing through the origin of biplot and AEC point was made and is called the average environment axis (AEA). The line which is perpendicular to the AEA line and pass through the origin is called the average ordinate environment (AOE). This line separates genotypes that have fresh tuber yield higher than grand mean (right of the line) with genotypes that have a fresh tuber yield lower than grand mean (left of the line). Genotypes that have higher tuber yield than the grand mean are Malang 6 (G2), Malang 4 (G3), Adira 4 (G4), CMM
03036-7 (G6), CMM 03038-7 (G8), and CMM 03094-4 (G10). Genotypes that have lower tuber yield than the grand mean are UJ5 (G1), CMM 03025-43 (G5), CMM 03036-5 (G7), CMM 03094-12 (G9), CMM 03095-5 (G11), CMM 02040-1 (G12), CMM 02033-1 (G13), CMM 02035-3 (G14), and CMM 02048-6 (G15) (Fig.5).

Stability of the genotypes depends on their distance from the AEA line. The genotypes closer to the AEA line are more stable than others. The results of this study indicate that the most stable genotype and high tuber yield is CMM 03038-7 (G8). Genotype CMM 03025-43 (G5) and CMM 03095-5 (G11) are also a stable genotype, but it have low tuber yield. Other genotypes that relative stable and have high tuber yield are Malang 6 (G2) and CMM 03036-7 (G6).

Ideal genotypes are genotypes that should have large PC1 scores (high mean yield) and small (absolute) PC2 score (high stability) [5, 7, 3]. Projection of the ideal genotype on the AEA line is equal to the longest vector of all genotypes and its projection on the AOE line is obviously zero [19]. Based on criteria of ideal genotype as already mentioned above, the closest genotype to the ideal genotype is CMM 03038-7 (G8) (Fig.6).

![Fig. 5. GGE biplot base symmetrical scale with AEC.](image)

3.6. Identification of ideal test environments

Ideal environments are environments that should have small (absolute) PC2 scores (more representative of the overall environment) and large PC1 score (more power to discriminate genotype in terms of the genotypic main effect) [5, 7]. Ideal environment can be used to as a reference for genotype selection in the multi-environment trial [20]. The projection of ideal environment on the AEA line is equal to the longest vector of all environment (the most discriminating environment) and its projection on the AOE line axis was zero, meaning that is absolutely representative of average environments [19]. The ideal environment that represented by the green small circle is the most discriminating of genotypes and yet representativeness of the other test environments. Accordingly, S1 (Kediri) is the test environment that is closest to the ideal environment, so it can be categorized that Kediri is the most ideal environment compare to other environments (Fig.7)
4. Conclusion

According to GGE biplot analysis, CMM 03038-7 (G8) is the most stable clone across five environments with high tuber yield, and Kediri (S1) is the ideal environment to discriminating genotypes.

As has been reported previously by many authors, GGE biplot can make biplot analysis of MET data very easy, informative, and interesting.
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References