Genetic Diversity Analysis of Tepi Surroundings Coffee (*Coffea arabica* L.)
Germlasm Accessions using Quantitative Traits in Ethiopia

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**ABSTRACT**

Ethiopia is the homeland and center of genetic diversity of Arabica coffee (*Coffea arabica* L). A field experiment on evaluation of 93 Tepi surroundings coffee germplasm accessions including 5 standard checks was conducted using Augmented Design of four blocks at Tepi National Spices Research Center during 2016 cropping seasons. Data on 22 morphological agronomic characters was obtained. The germplasm accessions differed significantly for the 10 of 22 morphological agronomic characters indicating the prevalence of variability among the coffee germplasm accessions studied. Furthermore, the first eight principal components explained 74 percent of the total variation prevalent within the germplasm accessions, out of which 21 percent was explained by the first principal component. Average linkage cluster analysis using Mahalanobis (D\(^2\)) distance for the 22 characters grouped the 93 accessions in to 5 clusters. The number of accessions per cluster ranged from 1 in cluster V to 56 in cluster I. The clustering pattern of the accessions revealed the prevalence of genetic diversity in the Tepi surroundings (southwestern) coffee for the characters considered. The maximum inter-cluster distance was observed between clusters I and V while the minimum was observed between clusters II and III. The study highlighted the possibility of using accessions of the distant clusters as potential candidates for the genetic improvement of southwestern Ethiopian coffee through crossing and selection.

**Key words:** *Coffea arabica*, Cluster analysis, Genetic divergent, Principal component analysis

**INTRODUCTION**

Coffee is a perennial field crop which belongs to the genus *Coffea* in the *Rubiaceae* family, and is mostly grown in the tropical and subtropical regions (Berthaud and Charrier, 1988). About 124 species of the genus Coffea have been identified so far (Davis et al., 2012). *Coffea arabica* is known to be one of the most important beverages in the world and is a very important source of foreign exchange for many countries (Labouisse et al., 2008).

Ethiopia is the primary center of origin and center of genetic diversity of *Coffea arabica* L. and the existence of such genetic diversity provides immense opportunity for coffee improvement (Fikadu et al., 2008). According to CSA, (2017), the estimated area of land covered by coffee in Ethiopia is about 700474.69 ha, where as the estimated annual national production of clean coffee is about 469901.12 tons with average productivity of 669.6 kg ha\(^{-1}\). Ethiopia remains the largest producer of coffee in Africa and is the fifth largest coffee producer in the world next to Brazil, Vietnam, Colombia and Indonesia, contributing to about 4.2% of the total world coffee production (ICO, 2016).

The crop is mainly produced in the Southern, South Western and Eastern parts of the country. The wider agroclimatic conditions and soil factors offer the country to grow diverse Arabica coffee, which accounts about 80% of the world coffee trade. Arabica coffee grows under very diverse environments including altitude of 550–2600m and annual rainfall of 1000–2000mm (Mesfin and Bayetta, 1987). In view of the present situation where our coffee genetic resource is under serious threat of extinction mainly due to deforestation, replacement of traditionally grown landraces of coffee by improved varieties, environmental degradation and change in land use (Gole and Teketay, 2001). Jimma Agricultural Research Center (JARC) has been collecting coffee germplasm from different coffee growing regions of Ethiopia and so far about 6721 accessions have been conserved at JARC and its sub-centers is a typical evidence for the huge and...
untapped coffee genetic wealth of coffee in the country (Taye, 2010). Effective germplasm conservation strategy and immediate measures are crucial to reduce such loss of coffee genetic resources and improve the productivity of the crop by developing high productive coffee varieties. Other major factors such as predominant use of unimproved local coffee landraces, as well as conventional husbandry and processing practices, which in turn seriously hampers the overall national coffee production and productivity of the smallholder coffee farmers in the country (Taye, 2010). Information about genetic diversity within and among genotypes of any crop is fundamental to estimate the potential of genetic gain in a breeding program and for effective conservation of available genetic resources (Sakiyama, 2000). It may also be important for selecting promising parental lines in hybrid variety development (Barbosa et al., 2003). Such information could also serve as a bench mark for future assessment of genetic erosion (Hammer et al., 1996).

Several workers have estimated the extent of genetic diversity present from the different sources of Arabica coffee germplasm collections. For example, a study by Kebebe and Bellachew (2008) on Arabica coffee collections from Hararge indicated the presence of high genetic diversity. Similarly, the genetic diversity analysis carried out by Yigzaw (2005) by employing morphological characters, biochemical characteristics and molecular markers on coffee Arabica genotypes from Ethiopia displayed the existence of genetic diversity.

The collected coffee accession is not systematically characterized to identify desirable growth characters for future breeding program and remains unknown to breeders from Tepi surroundings (Bench-maji and Sheka zones) earlier. Thus, the present study was carried out with the objective of estimating the genetic diversity among some Tepi surroundings coffee germplasm accessions using morphological traits thereby avoid handling of large number of duplicates and of facilitating their use in coffee Arabica breeding programs.

MATERIALS AND METHODS

Experimental material, design and management

The experiment was conducted on 93 coffee accessions germplasms including the five standard checks at Tepi National Spices Agricultural Research Center, Southwest Ethiopia. Tepi is located at a a latitude of 7° 3' N and longitude of 35° 18' E. and at an altitude of 1200masl. The mean annual rainfall of the area is 1678 mm per annum well distributed over eight months with an average maximum and minimum air temperatures of 30°C and 16°C, respectively and Soil PH=6.9-8 with fine textured 30-80% clay soil. The experimental design used was augmented design of four blocks. A plot consisted of a single row with 12 trees. Spacing between both rows and plants was 2m, (plot area of by 2x2 =4m²). All the management practices such as shading, weeding and fertilization were uniformly applied to all plots as per the recommendation (Endale et al., 2008).

Data collection

During the course of this study, data on 22 quantitative characteristics namely; includes leaf length (cm), leaf width (cm), leaf area (cm²), bean length (mm), bean width (mm), bean thickness (mm), fruit length (mm), fruit width (mm), fruit thickness (mm), hundred bean weight (gm), yield (kg/ha), plant height(cm), stem diameter(cm), number of main stem nodes (no), canopy diameter (cm), average internodes of stem (cm), length of primary branches (cm), number of primary branches, number of secondary branches, height up to first primary branches (cm), number of node on primary branch (no) and coffee leaf rust (%) (IPGRI, 1996).

Statistical analysis

The variance (ANOVA) was analyzed using SAS version 9.2 (SAS, 2010) based on augmented design. Least Significant Difference (LSD at P=0.05) were employed to identify accessions that are significantly different from each other. In this study, 22 morphological characters that showed statistically significant variations among the accessions were used for clustering and principal component analysis. The data were subjected to cluster analysis so as to determine the variability among the accessions. Hierarchical clustering was employed using the similarity coefficients among the 93 coffee accessions. Clustering was performed using the proc cluster procedure of SAS version 9.2 (SAS, 2010) by employing the method of average linkage clustering strategy of the observation. The numbers of clusters were determined by following the approach suggested by Copper and Milligan (1988) by looking into three statistics namely Pseudo F, Pseudo t² and cubic clustering criteria. Genetic divergence between clusters was calculated using the generalized Mahalanobis’s (D²) statistics (Mahalanobis, 1936) using the equation:

\[
D_p^2 = (X_i - X_j)^T S^{-1} (X_i - X_j)
\]

Where, \(D_p^2\) = the distance between any two groups i and j; \(X_i\) and \(X_j\) = the p mean vectors of accessions i and j, respectively. \(S^{-1}\) = the inverse of the pooled covariance matrix.

The D² values obtained for pairs of clusters was tested for significance at 0.05 and 0.01 level of significance against the tabulated values of chi square test t² for p degrees of freedom, where p is the number of variables considered (Singh and Chaudhary, 1987). Principal component analysis was also performed by employing SAS version 9.2 (SAS, 2010).

RESULTS AND DISCUSSION

Analysis of variances

Analysis of variance of 22 quantitative traits revealed significant difference among the accessions in leaf length, leaf width, leaf area, number of primary branches, fruit length, bean length, bean width, bean thickness, hundred bean weight and clean bean yield. The variability present for important traits in the present study clearly proved the possibility to bring considerable improvement mainly in coffee yield and coffee leaf rust resistance through selection and hybridization. Bayetta (1997) reported high genetic variability within the Arabica coffee population for yield, CBD (coffee berry disease) resistance and
growth characters. The existence of variability among Arabica coffee accessions was further confirmed by many authors who reported significant differences among coffee germplasm accessions collected from major coffee growing regions of the country (Ermias, 2005; Yigzaw, 2005; Olika et al., 2011).

Cluster analysis

The $D^2$ value based on the mean of coffee germplasm accessions resulted in classifying the 93 accessions into five groups (Table 1). Cluster I and II were the largest with 56 (60.2%) and 23 (24.7%) germplasm accessions respectively, followed by Cluster IV with 7 germplasms (7.5%), cluster III with 6 germplasm accessions (6.5%) and one accession (1.1%) into cluster V. This indicates that the tested coffee germplasm accessions were highly divergent. The five checks and 28 accessions from Bench maji and 23 accessions from Sheka Zone were grouped into cluster 1, indicating that germplasm was moving freely between the two zone. All the 23 accessions of Sheka zone are in this cluster. All other clusters contained different number of only Bench-maji accessions. This indicates divergence of coffee accessions from the two zones, although almost equal numbers of accessions from the two zones were grouped together in cluster I. However, the classification of coffee accessions from Bench maji zone into five clusters, suggests the existence of high genetic diversity within these collections. The diversity between the two collection zones and within Sheka zone can be exploited in future breeding to broaden the genetic base of the crop and develop new varieties.

The maximum inter cluster distance was between clusters I and V (1374.42) followed by between clusters II and V (1061.33), I and IV (883.54) and also between cluster III and IV (823.633). The minimum inter cluster distance was observed between clusters II and III (239.85) followed by I and II (314.34) and III and IV (331.873) (Table 2). The chi-square test for similarity of germplasm accessions resulted in classifying the 93 accessions into five groups (Table 1).

The first principal component which accounted for 21% of the total variation among the accessions for 22 quantitative traits (Table 3). The first principal component accounted for 74% of the total variation among the accessions. The second principal component accounted for 21% of the total variation among the accessions. The third principal component accounted for 21% of the total variation among the accessions. The fourth principal component accounted for 21% of the total variation among the accessions. The fifth principal component accounted for 21% of the total variation among the accessions. The first principal component which accounted for 21% of the total variation among the accessions. The second principal component accounted for 21% of the total variation among the accessions. The third principal component accounted for 21% of the total variation among the accessions. The fourth principal component accounted for 21% of the total variation among the accessions. The fifth principal component accounted for 21% of the total variation among the accessions. The first principal component which accounted for 21% of the total variation among the accessions. The second principal component accounted for 21% of the total variation among the accessions. The third principal component accounted for 21% of the total variation among the accessions. The fourth principal component accounted for 21% of the total variation among the accessions. The fifth principal component accounted for 21% of the total variation among the accessions. The first principal component which accounted for 21% of the total variation among the accessions. The second principal component accounted for 21% of the total variation among the accessions. The third principal component accounted for 21% of the total variation among the accessions. The fourth principal component accounted for 21% of the total variation among the accessions. The fifth principal component accounted for 21% of the total variation among the accessions.
Table 3: Principal components for 22 morphological traits of 93 coffee accessions.

<table>
<thead>
<tr>
<th>Traits</th>
<th>Prin1</th>
<th>Prin2</th>
<th>Prin3</th>
<th>Prin4</th>
<th>Prin5</th>
<th>Prin6</th>
<th>Prin7</th>
<th>Prin8</th>
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</thead>
<tbody>
<tr>
<td>LL</td>
<td>0.31</td>
<td>0.07</td>
<td>-0.32</td>
<td>-0.23</td>
<td>-0.03</td>
<td>0.16</td>
<td>0.02</td>
<td>-0.14</td>
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<tr>
<td>LW</td>
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<td>0.22</td>
<td>-0.36</td>
<td>-0.19</td>
<td>-0.08</td>
<td>0.14</td>
<td>-0.15</td>
<td>0.14</td>
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<tr>
<td>LA</td>
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<td>0.19</td>
<td>-0.38</td>
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<td>0.16</td>
<td>-0.09</td>
<td>0.03</td>
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<tr>
<td>PH</td>
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<td>0.34</td>
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<td>0.29</td>
<td>0.20</td>
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<td>HUFPB</td>
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<td>-0.09</td>
<td>0.05</td>
<td>-0.18</td>
<td>0.51</td>
<td>0.07</td>
<td>0.18</td>
<td>0.50</td>
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<td>-0.18</td>
<td>0.34</td>
<td>-0.14</td>
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<td>0.29</td>
<td>0.20</td>
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<td>0.48</td>
<td>-0.05</td>
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<tr>
<td>GD</td>
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<td>-0.14</td>
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<td>0.37</td>
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<tr>
<td>LPB</td>
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<td>0.08</td>
<td>0.26</td>
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<td>0.24</td>
<td>-0.03</td>
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<tr>
<td>NPB</td>
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<td>0.29</td>
<td>0.20</td>
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<tr>
<td>NNPB</td>
<td>0.04</td>
<td>0.34</td>
<td>0.13</td>
<td>0.11</td>
<td>0.11</td>
<td>-0.12</td>
<td>-0.15</td>
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<tr>
<td>CD</td>
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<tr>
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</tr>
<tr>
<td>FT</td>
<td>0.32</td>
<td>-0.19</td>
<td>0.01</td>
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<td>-0.13</td>
<td>-0.34</td>
<td>0.06</td>
<td>0.29</td>
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<tr>
<td>BL</td>
<td>0.08</td>
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<td>-0.03</td>
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<td>0.42</td>
<td>0.26</td>
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<tr>
<td>BW</td>
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<td>-0.08</td>
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<tr>
<td>BT</td>
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<td>-0.11</td>
<td>-0.05</td>
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<td>0.05</td>
<td>-0.03</td>
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<td>CLR</td>
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<td>0.34</td>
<td>-0.17</td>
<td>0.08</td>
<td>-0.02</td>
<td>0.07</td>
<td>0.37</td>
<td>0.14</td>
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<tr>
<td>HBW</td>
<td>0.18</td>
<td>0.08</td>
<td>-0.22</td>
<td>0.25</td>
<td>0.05</td>
<td>0.01</td>
<td>0.45</td>
<td>-0.04</td>
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<tr>
<td>Yld</td>
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<td>0.37</td>
<td>0.10</td>
<td>-0.07</td>
<td>-0.04</td>
<td>-0.32</td>
<td>0.21</td>
<td>0.07</td>
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<tr>
<td>Eigen value</td>
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<td>2.98</td>
<td>2.07</td>
<td>1.99</td>
<td>1.38</td>
<td>1.3</td>
<td>1.10</td>
<td>1.00</td>
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<tr>
<td>Percent variation</td>
<td>0.21</td>
<td>0.13</td>
<td>0.094</td>
<td>0.09</td>
<td>0.06</td>
<td>0.06</td>
<td>0.05</td>
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<tr>
<td>Cumulative</td>
<td>0.21</td>
<td>0.34</td>
<td>0.44</td>
<td>0.53</td>
<td>0.59</td>
<td>0.65</td>
<td>0.69</td>
<td>0.74</td>
</tr>
</tbody>
</table>

Where LL=leaf length, LW=leaf width, LA=leaf area, FL=fruit length, BL=bean length, HBW=hundred bean weight Yld=green bean yield, PH=plant height, NPB=number of primary branches, LPB=number of primary branches, NSB=number of secondary branches, CD=canopy diameter, FW=fruit width FT=fruit thickness; FL=fruit length, BL=bean length, HBW=bean weight; BT=bean thickness; CLR=Coffee leaf rust; GS=Girth of stem.

Conclusions

Almost all clusters showed a highly significant (P<0.01) difference among each other. Germplasm accessions from Bench-maji Zone were more divergent than selections of Sheka Zone though relatively greater number of selections were considered from Bench-maji Zone. The significant inter-cluster distances between clusters indicated that there is a high opportunity for obtaining transgressive segregates and maximize heterosis by crossing germplasm accessions belonging to these clusters. Therefore, the grouping of accessions by multivariate methods could be of considerable practical value to the coffee breeders so that representative accessions could be chosen from such clusters for selection and hybridization programs. It is also possible to state that quantitative characters studied significantly contributed to the elucidation of genetic diversity prevalent in the region could be used as a selection criterion for improving the productivity of the crop since they represent the lion’s share in the variability of the coffee population in the specified area. Furthermore, additional traits of interest and molecular techniques may be very useful in order to further confirm the present encouraging result that indicated the presence of considerable variations within Southwestern Ethiopia coffee populations that provides immense potential for the development of improved varieties from the local landraces for the area.

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REFERENCES


