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Organoleptic Characterization of Some Limu Coffee (*Coffea arabica* L.) Germplasm at Agaro, Southwestern Ethiopia

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ABSTRACT

The aim of this study was to characterized and to estimate the extent of genetic variation and character association of organoleptic quality attributes of *Coffea arabica* accessions from Limu (Jimma, Ethiopia). Forty nine coffee germplasm accessions which have little or no information about their genetic variability together with two checks were planted in the field at Agaro Agricultural Research Sub Center, Ethiopia from 2004 to 2009. Simple lattice design with two replications was used in this particular study. Variances component method was used to estimate genetic variation, heritability and genetic advance. Relationship among traits was also estimated using standard method. The germplasm accessions differ significantly for most of the traits. Analysis of variance, variance components, phenotypic and genotypic associations, cluster analysis and principal components were computed for the sensorial quality attributes studied. The results depicted significant variations among coffee accessions for cup quality attributes studied, except aromatic intensity, bitterness, astringency and body. There was high phenotypic coefficient of variation for astringency and bitterness. This is in contrast to the low phenotypic coefficients of variation recorded for aromatic intensity and body. In principal component analysis, the first three principal components with eigen values greater than one explained 81.4% of the total variation. The first two principal components accounted with percent variability of 52.87 and 17.77%, respectively explained 70.64% of the total variability among the coffee germplasm. These were grouped into three genetically divergent clusters and three uncorrelated principal components. In general, our findings show that more than half percent of the Limu accessions had similar quality attributes with the standard checks. The results also confirmed the presence of variability in most quality attributes among the Limu coffee accessions and this could be exploited in the future genetic improvements.

Key words: *Coffea arabica*, cluster analysis, genetic divergent, variability, correlation analysis

INTRODUCTION

The Ethiopian coffee is also important source of coffee genetic resources for the world coffee industry. As a matter of fact, Ethiopia is the single known center of origin and genetic diversity for arabica coffee (*C. arabica*) (Wintgens, 2004). It is cultivated in most parts of the tropics, accounting for 80% of the world's coffee market, about 70% of the production and it is also important

source of income and employment in developing countries like Latin America, Africa and Asia (Anthony *et al.*, 2001).

When assessing organoleptic quality, one has to take into account that consumers have a specific taste according to their nationality that leads to an unreliable definition of organoleptic quality. Expert assessors can describe organoleptic quality profile. It is a complex procedure that uses some specific descriptors. At least five expert assessors have to be trained to use the vocabulary. Flavor obtained in a coffee cup is the result of multiple aromatic compounds present in the coffee (more than 800 in the roasted coffee) (Belitz *et al.*, 2004). Since measurement of the composition in 800 aromatic compounds present in roasted coffee is not a viable method to assess coffee organoleptic quality, development of indirect predictors of coffee organoleptic quality through biochemical compounds is found inevitable and efforts are underway (Leroy *et al.*, 2006).

The success of a new variety of Arabica coffee (*Coffea arabica* L.) depends to an important extent on its liquors. Selection for these traits is however constrained by the prevalence of large genotype-by-environment interactions in connection with the low genetic variability characteristics of this species (Agwanda *et al.*, 2003). However, Van der Vossen (1985) reported non-significant genotype x environment interaction effects on quality characters, such as bean size and cup quality. However, Gichimu and Omondi (2010a) reported non-significant genotype x environment interaction effects in coffee. Organoleptic quality attributes variation was accounted for genetic, environment and genetic x environment interaction. Therefore, the presence of strong genotype x environment interaction for arabica coffee quality, limits development of wide adapting varieties, initiates coffee quality mapping, conservation of coffee genetic resources and establishment of core collections with respective origins (Getu, 2009).

In contrast to the low average national coffee yield, the released coffee varieties can give high average clean coffee yields ranging from 1200 to 2600 and 800 to 2400 kg ha^{-1} under on- research and on-farm conditions, respectively (Bayeta *et al.*, 1998). Efficient utilization of the genetic potential held in germplasm collections requires detailed knowledge about the collections (Beuselinck and Steiner, 1992), including characterization, evaluation and classification. The research findings depict the significant interactions between coffee genotypes and environment, demonstrating the need for local coffee landrace development program (Bellachew and Labouisse, 2006). In this regard, apart from some observations based on the variety trials, there has been no systematic diversity analysis carried out in Limu coffee germplasm collection and this might have resulted in the handling of a large degree of duplicated germplasm collection. Similarly, there is no detailed information on the extent and nature of interrelationships among characters. This study was, therefore, carried out with the specific objectives to characterize some Limu coffee germplasm accessions based on organoleptic quality attributes, to estimate the extent of phenotypic and genotypic correlations between pairs of characters in the study coffee germplasm accessions, to estimate the genetic differences among the coffee genotypes. These would enable to cluster them into different homogenous groups using organoleptic quality attributes and to estimate the extent of phenotypic and genotypic variability, heritability and genetic advance expected under selection in future coffee breeding program.

MATERIALS AND METHODS

Study site: The experiment was conducted at Oromiya Estate in Jimma District, at Agaro Agricultural Research Sub Center, Ethiopia from 2004 to 200. It is located at 45 km in the southwest of Jimma town at an altitude of 1630 m.a.s.l. It is situated at 7°50'35"N latitude and

36°35'30"E longitude. The mean annual rainfall of the area is 1616 mm with average maximum and minimum temperatures of 28.4 and 12.4°C, respectively (Elias, 2005).

Experimental material, design and management: The study was carried out during the main season in 2009/10 on batch II forty-nine Limu coffee trial of six-year old including two standard checks (744 and F-59) (Table 1). The coffee germplasm accessions were collected in 2003 from the potential and representing areas in the Limu-Kossa Wereda of Jimma Zone and were planted in the field in August 2004. The experiment was laid out in a 7×7 simple lattice design with 2 replication and seven accessions per incomplete block. Six trees per accessions were planted in 2×2 m spacing. All the management practices such as shading, weeding and fertilization were uniformly applied to all plots as per the recommendation.

Experimental procedure and data collection: Ripe Red coffee cherries were handpicked. Before pulping fully ripened and healthy berries were separated from foreign materials and unripe green cherries and green bean coffee samples were prepared during 2009/2010 cropping season. A total of 98 samples were prepared from forty nine accessions (six trees per accessions bulked together). The samples were prepared from six trees per accession per replication at peak harvest period. The samples were carefully prepared using wet processing method (pulping, fermentation, and drying) and prepared for sensorial analysis using the following procedures.

Table 1: Geographical origin of coffee accessions used in the study

Accession	Region	Zone	Wereda	Collection place	Altitude
L-1/2003	Oromiya	Jimma			
L-2/2003					
L-3/2003					
L-4/2003			Limu kossa	Miaa	1670
L-5 /2003					
L6 /2003					
L7 /2003	Oromiya	Jimma	Limu kossa	Cheraki	1640
L8 /2003					
L9 /2003 L10/2003	Oromiya	Jimma	Limu kossa	Babo	1610
L11/2003	Oromiya	Jimma			
L12 /2003					
L13 /2003 L14/2003 L15/2003		Limu kossa	Kosa sate farm	1610-1850	
L16/2003 L17/2003					
L20/2003 L22/2003 L23/2003	Oromiya	Jimma	Limu kossa	Tenebo	1650
L24/2003 L25/2003	Oromiya	Jimma	Limu kossa	Buya	1650-1680
L26/2003 L27/2003 L28/2003					
L29/2003 L30/2003	Oromiya	Jimma	Limu kossa	Ajamo	1680
L32/2003 L33/2003 L34/2003	Oromiya	Jimma	Limu kossa	Genji	1640-1720
L35/2003 L47/2003 L48/2003					
L36/2003 L37/2003	Oromiya	Jimma	Limu kossa	Bidaru	1640
L38/2003 L39/2003	Oromiya	Jimma	Limu kossa	Gindacha	1640
L40/2003 L41/2003 L42/2003	Oromiya	Jimma	Limu kossa	Algae	1690
L43/2003 L44/2003	Oromiya	Jimma	Limu kossa	Kelecha	1670-1690
L45/2003 L46/2003					
L49/2003 L50/2003 51/2003	Oromiya	Jimma	Limu kossa	Sombo	1710-1720
744	SNNP			Bonga	1770
F-59	SNNP			Bonga	1770

Pulping: Fully ripened beans of berries were separated from the skin and pulp by using a pulping machine that squeezes the berries between fixed and moving surfaces.

Fermentation and drying: The beans were then stored in a fermentation tank for 48 h till first washing was made. Then, samples were stored for 24 h for final washing. The samples were placed on mesh wire under sun for drying. During drying, the moisture content of the bean wasmeasured by moisture tester H-E50 to maintain the moisture level at 10-12% for all samples uniformly. About 300 g of green coffee bean samples were prepared per accession per replication separately for each accession for organoleptic quality characteristics analysis. To attain homogenous bean size and healthy beans for organoleptic quality analysis, samples were screened through a mesh sieve 15 (5.95 mm). Then, samples on and above screen 15 were used for analysis. The sensorial quality analyses were carried out at the Jimma Agricultural Research Center by a group of well trained cup tasters/panelists of the center.

Roasting and grinding: The roaster machine, probatBRZ6, was first heated at about 160-200°C. About 150-200 g of green coffee bean sample prepared per accession per replication was used for roasting. When the roast starts to crackle (burst open), the gas were turned down. When the coffee was considered medium roast (7 min on average) it was tipped out into the cooling try. Cold air was blown through the coffee to produce rapid cooling-off. When the roast was cool (4 min on average), it was blown to remove the loose silver skins before grinding. Variability in roasting was controlled by measuring weight loss. The weight loss found was between 8 and 10% and these matches from medium to dark roasting degree reported by Agwanda *et al.* (2003). About 12 g medium seized ground coffee was prepared using Mahlkoing electrical grinder with middle adjustment.

Brewing: Soon after grinding, coffee powder weighing about 8 g was placed in a cup with a capacity of 180 mL. Then, boiling water was poured on to the ground coffee up to about half way in the cup. Soon after, volatile aromatic quality and intensity parameters were recorded by sniffing. Then, the contents of the cup were stirred to ensure an infusion of all coffee grounds. The cup was then filled to the brim with boiled water. The brew was made ready for panelists within 8 min.

Cup tasting: This was carried out once the beverage cooled to around 60°C (drinkable temperature). Two cups per sample were prepared for tasting session. The genotypes and the replicates were arranged at random. Aroma (aromatic quality and intensity), flavor, acidity, bitterness and astringency were scored using scales ranging from 0 to 5. Typical flavor was assessed as an after taste aromatic quality that could vary from winy to flowery (winy, fragrant, floral, citrus, moka, spicy and others). There was also an overall standard for liquor quality based on the above attributes that ranged from 0 to 5 scales. Mean of each variable by the panel was used for statistical analysis (ISO, 1992).

Data analysis: The data were statistically analyzed using lattice analysis of variance format design by using the statistical procedures described by Gomez and Gomez (1984). All statistical and data processing were performed Using XLSTAT (XLSTAT, 2008), Computer program, SAS (SAS, 2002) version 9.2 software and Genes version 7.0 soft ware. For characters having significant mean differences, the difference between treatment means was compared using LSD at 5% probability level.

The phenotypic variances and coefficients of variations were estimated as per Singh and Chaudhry (1985). Heritability in the broad sense for all organolyptic quality attributes was computed using the formula suggested by Singh and Chaudhry (1985). The genetic advance expected under selection, assuming selection intensity of the superior 5% of the genotypes was estimated in accordance with the methods illustrated by Allard (1960).

Phenotypic and genotypic correlations were computed following the method described by Singh and Chaudhry (1985).

Clustering of the 49 accessions for organolyptic quality attributes was performed using the proc cluster procedure of SAS version 9.2 by employing the method of average linkage clustering strategy of observations. The number of clusters was determined by following the approach suggested by Copper and Milligan (1988) by looking into three statistics namely, pseudo F, pseudo t^2 and the Cubic Clustering Criteria (CCC). The number of cluster was decided where the CCC and pseudo F statistics combined with a small value of the pseudot^2 statistics and large pseudo t^2 statistics for the next cluster fusion.

Genetic divergence between clusters was determined using the generalized Mahalanobis's D^2 statistics. Mahalanobis (1936) developed this method to determine divergence prevailing among population in terms of generalized group distance-deleted (Sharma, 1998)-deleted. Testing the significance of D^2 values obtained for a pair of clusters were taken as the calculated value of chi-square (χ^2) and tested against the tabulated value of χ^2 for P degrees of freedom (P is the number of organolyptic quality attributes) (Singh and Chaudhry, 1985) at appropriate probability level, that was considered.

RESULTS AND DISCUSSION

The mean square values for the studied traits ranged between 0.045 and 0.248 (Table 2). The Analyses of Variance (ANOVA) revealed the presence of significant differences among the Limmu coffee accessions for the organoleptic quality attributes (acidity, flavor and overall standard). However, ANOVA showed non-significant differences among the accessions for aromatic intensity, astringency, body, and bitterness (Table 2).The significant variations among coffee accessions for the quality traits indicated the existence of variability to have an effective selection for improvements. Similarly, different studies indicated the existence of high genetic diversity of arabica coffee for quality specifically in major coffee growing areas in Ethiopia (Yigzaw, 2005; Getu, 2009) and in Kenya (Walyaro, 1983). Similarly, Alsemaan *et al.* (2011) reported the existence of genetic diversity within *Rosa damascena* cultivated in Syria and recommended Almarahl and Bab Alnayrab accessions to be used to broaden the production of rose oil. In addition, Parthiban *et al.* (2011) found variability in Jatropha for some character studied. Gichimu and Omondi (2010b) studied on morphological characterization of five newly developed lines of Arabica coffee and two commercial cultivars in Kenya and they reported non significant difference for internodes length. However, Dar and Sharma (2011) obtained highly significant difference among the genotypes for all the quality traits studied in tomatoes.

In this study, we recorded genetic variance values ranging 0.00 to 0.09 and phenotypic variances between 0.06-0.15 (Table 3). Genetic variance of quality attributes was observed less as compared to environmental variance except aromatic quality (Table 3). The results revealed that bitterness, astringency and body variation was mainly influenced by environment. According to Verma and Agarwal (1982), heritability values greater than 50% are considered as high, whereas values less than 20% are low and values between 20 and 50% as medium. Accordingly, higher

Table 2: Mean squares of genotypes for organoleptic quality traits

	Mean square	
Trait	Genotype	Error
Aromatic intensity	0.056^{ns}	0.047
Aromatic quality	0.147**	0.062
Acidity	0.248**	0.095
Astringency	0.045^{ns}	0.096
Bitterness	0.051^{ns}	0.066
Body	0.046^{ns}	0.043
Flavor	0.187*	0.114
Overall standard	0.208**	0.103

*,**significant at p<0.05, 0.24 and p<0.01, 0.34, respectively

Table 3: Variability parameters for organoleptic quality trait in coffee accessions

Trait	σ^2g	σ^2P	H^2	PCV%	GCV%	GA	GA%
Aromatic intensity	0.01	0.11	12.11	9.09	3.28	0.06	1.90
Aromatic quality	0.09	0.15	64.74	11.95	9.51	0.49	14.90
Acidity	0.06	0.15	43.64	11.92	7.81	0.33	10.20
Astringency	0.00	0.07	0.00	110.24	0.00	0.00	0.00
Bitterness	0.00	0.06	0.00	37.50	0.00	0.00	0.00
Body	0.00	0.07	0.00	8.37	0.00	0.00	0.00
Flavor	0.05	0.12	39.70	10.56	6.79	0.25	7.80
Overall standard	0.06	0.16	40.77	12.48	7.86	0.33	10.30

σ^2g: Genotypic variance, σ^2P: Phenotypic variance, H^2: Heritability, GCV: Genotypic coefficients of variation, PCV: Phenotypic coefficients of variation, GA: Genetic advance, GA%: Genetic advance as percent of mean

estimates of broad sense heritability were observed for aromatic quality (64.74%) and medium broad sense heritability values were recorded for acidity (43.64%), over all standards (40.77%) and flavor (39.70%). However, aromatic intensity, astringency, bitterness and body were grouped under low heritability range. Variations of genotypes for bitterness, astringency and body were due to environmental factors and thus broad sense heritability was estimated to be small. Thus, the study depicted that bitterness, astringency and body are the appropriate parameters to control variation due to roasting problem during liquoring. Therefore, these attributes are less important to evaluate organoleptic performance of genotypes.

Phenotypic coefficients of variation values ranging from 8.37 to 37.50% and genotypic coefficient of variation values ranging from 0.00 to 9.51% were obtained Table 3). The organoleptic cup quality attributes were influenced by both environment and genotype. Genotypes respond differently for quality attributes and their magnitude is measured by Phenotypic Coefficient of Variation (PCV) and Genotypic Coefficient of Variation (GCV) values greater than 20% are regarded as high, whereas values less than 10% are considered to be low and values between 10 and 20% to be medium (Deshmukh *et al*., 1986).

The organoleptic cup quality attributes were influenced by both environment and genotype. Genotypes respond differently for quality attributes and their magnitude is measured by Phenotypic Coefficient of Variation (PCV) and Genotypic Coefficient of Variation (GCV) values greater than 20% are regarded as high, whereas values less than 10% are considered to be low and values between 10 and 20% to be medium (Deshmukh *et al*., 1986).

High Phenotypic Coefficient of Variation (PCV) was recorded for astringency and bitterness. Medium phenotypic coefficient of variation was obtained for aromatic quality, acidity, flavor and over all standard. In contrast, low values of phenotypic coefficients of variation also recorded for aromatic intensity and body. All quality attributes categorized under low GCV range. Environmental variation had large effect on the expressions of astringency, bitterness and body where difference between Genotypic Coefficients of Variation (GCV) and Phenotypic Coefficients of Variation (PCV) was relatively large. Thus, selection on phenotypic basis of these attributes may not be effective for the genetic improvement. However, relatively, small difference was obtained for other attributes indicating both environmental and genetic variation contributed towards expressions of the traits and selections on phenotypic basis for this trait may improves the population for coffee quality.

High heritability, genotypic coefficient of variation coupled with high genetic advance as percent of mean for aromatic quality attribute implies the potential for the coffee genotype improvement through selection.

Although the phenotypic coefficients of variation were greater than genotypic coefficients of variation for quality attributes aromatic intensity, aromatic quality, acidity, flavor and over all standard a narrow gap was found between PCV and GCV values, indicating little influence of environment in the expression of quality trait. The present study is in agreement with the study by Getu (2009).

The phenotypic coefficient of variability were although greater for quality attributes aromatic intensity, aromatic quality, acidity, flavor and over all standard than respected genotypic ones but a narrow gap was found indicating little influence of environment in the expression of quality trait. The present study is in agreement with the study by Getu (2009).

We obtained phenotypic association values ranging between -0.51 and 66 (Table 4). Significant positive phenotypic association was observed among aromatic quality, aromatic intensity, over all standard, body, acidity, and flavor (Table 4). Attributes which correlated strongly and positively with flavor were considered as good cup quality attributes. Flavor revealed negative association with bitterness. Therefore, aromatic quality, aromatic intensity, acidity, body and over all standard were described as good cup quality attributes, whereas bitterness were considered as poor cup quality attributes. Body was positively and significantly associated with good cup quality attributes such as aromatic intensity, aromatic quality, flavor and overall standards. Expression of characters of crop plants is correlated due to genotypic and/or environmental factors. Similarly, Yigzaw (2005) indicated positive association among good cup quality attributes. Moreover, Agwanda *et al.* (1999) identified flavor as an all round organoleptic attribute to be considered during selection to develop superior coffee genotypes which is in support of present finding. In addition, direct observable phenotypic association of characters resulted from genotypic and/or environmental correlations (Falconer and Mackay, 1996). Therefore, these organoleptic quality attributes contributed to good final cup quality. Thus, considering good cup quality attributes like flavor during selections would improve future target population for quality. Similar to present finding, Umamaheswari and Mohanan (2011) identified internodal length, vine length, number of inflorescences per plant and leaf area as lead characters in *Vanilla planifolia* Andrews and they recommended that due weightage can be given to these characters in improvement of this species.

The results of genotypic association for organoleptic quality attributes were given in Table 4 and we recorded the values ranging between 0.68 and 0.99. In this study, positive and significant genotypic association was observed among aromatic intensity, aromatic quality, acidity,

Table 4: Senso-phenotypic (above diagonal) and genotypic (below diagonal) correlation

Trait	AI	AQ	AC	AS	BI	BO	OVS	FL
AI	1	0.54**	0.28*	0.05	-0.11	0.36**	0.40**	0.33*
AQ	0.97**	1	0.33*	0.24	-0.05	0.38**	0.66**	0.61**
AC	0.99**	0.58**	1	0.09	-0.51**	0.11	0.52**	0.49**
AS	0.98**	0.91**	0.99**	1	0.12	0.18	0.22	0.19
BI	0.94**	0.99**	0.91**	0.91**	1	0.07	-0.09	-0.17
BO	0.97**	0.90**	0.97**	0.93**	0.99**	1	0.43**	0.39**
OVS	0.92**	0.92*	0.68**	0.96**	0.93**	0.95**	1	0.91**
FL	0.90**	0.93*	0.90**	0.99**	0.98**	0.99**	0.87**	1

*, **significant at $p<0.05$, 0.24 and $p<0.01$, 0.34, respectively. AI: Aromatic intensity, AQ: Aromatic quality, AC: Acidity, AS: Astringency, BI: Bitterness, BO: Body, FL: Flavor, OVS: Over all standard

astringency, bitterness, body, flavor and overall standard. Likewise, Yigzaw (2005) and Getu *et al.* (2009) indicated similar positive association among good cup quality attributes in Arabica coffee genotypes. Agwanda *et al.* (1999) indicated that flavor has got strong genetic association with preference and therefore it is the best selection criteria for the genetic improvement of Arabica coffee liquor quality. Moreover, flavor used for discriminating among varieties because it was found as an all round beverage quality attribute which in corporate other aromatic attributes and well associated with good cup quality attributes like acidity and body. Nikhila *et al.* (2008) reported that length of primary branches, number of primary branches, intermodal length and bush spread are the character that should be given premium importance while carrying out crop improvement programmes in robusta coffee.

Cluster distributions of 49 coffee genotypes using organoleptic quality traits is given in Table 5. The number of accessions classified in each cluster was 31, 17 and 1, in cluster I, II and II, respectively. Cluster I composed of 31 (63.27%), Cluster II 17 (34.69%) accessions and Cluster III 1 (2.04%), indicating how closely related different genotypes were grouped together. The first group comprised 31 coffee accessions that were characterized by low organoleptic quality. The second cluster comprised 17 that were characterized by medium in organoleptic quality and the third remaining cluster comprised of one coffee accession which was relatively superior in organoleptic quality. The present study was in agreement with previous findings that reported by Dessalegn *et al.* (2008) and on forty-two Ethiopian collections of Arabica coffee genotypes.

Mean organoleptic quality attributes of clusters for eight organoleptic quality attributes in 49 coffee accessions was given in Table 6 and it ranged 0.25 to 4.58. Cluster III and I was found relatively different because distant and divergent groups constituted these clusters and thereby characterized by extreme values of organoleptic quality attributes. High mean values of aromatic intensity, aromatic quality, acidity, body and flavor and overalls standard characterized cluster III while the lowest mean values of these attributes characterized in turn cluster I. Mean of clusters therefore, indicated the presence of two quite different groups (I and III) consisted of contrasting sensorial quality attributes. Cluster II was average for good cup quality attributes except astringency. The released coffee varieties (744 and F-59) were used as check to identify Limu coffee accession that perform similar to the check. The two checks were grouped in clusterI, indicating more than half percent of Limu accessions had similar quality attributes with the standard checks. The present finding corroborates with that of Yigzaw (2005) and Dessalegn *et al.* (2008).

Table 5: Cluster distribution of the 49-coffee genotypes for organoleptic quality traits

Cluster I		Cluster II	Cluster III
744*	L-27/2003	L-05/2003	L-12/2003
F-59*	L-29/2003	L-06/2003	
L-01/2003	L-32/2003	L-09/2003	
L-02/2003	L-33/2003	L-16/2003	
L-03/2003	L-35/2003	L-17/2003	
L-04/2003	L-38/2003	L-20/2003	
L-07/2003	L-39/2003	L-23/2003	
L-08/2003	L-40/2003	L-24/2003	
L-10/2003	L-43/2003	L-28/2003	
L-11/2003	L-44/2003	L-30/2003	
L-13/2003	L-45/2003	L-34/2003	
L-14/2003	L-46/2003	L-36/2003	
L-15/2003	L-47/2003	L-37/2003	
L-22/2003	L-49/2003	L-41/2003	
L-25/2003	L-51/2003	L-42/2003	
L-26/2003		L-48/2003	
		L-50/2003	----
	31 (% 63.27)	17 (% 34.69)	1 (2.04%)

*Represents the check varieties

Table 6: Mean organoleptic quality attributes in three clusters

Cluster	AI	AQ	AC	AS	BI	BO	FL	OVS
I	3.28*	3.22*	3.00*	0.29**	0.25**	3.18*	2.99**	3.03*
II	3.39	3.42	3.53	0.23	0.08	3.33	3.39	3.42
III	3.71**	4.38**	4.29**	0.25	0.00*	3.46**	4.33**	4.58**

*,** represent the lowest and highest mean values, respectively. AI: Aromatic intensity, AQ: Aromatic quality, AC: Acidity, AS: Astringency, BI: Bitterness, BO: Body, FL: Flavor, OVS: Over all standard

Squared mahalanobis distance between clusters for organoleptic quality attributes indicated that most of the clusters were highly significantly distant from each other (Table 7). Significant distance analysis and association of genotypes to specific cluster group indicated the presence of genetically distant materials and the association of genotypes with specific cluster group of distinct characteristics. Cluster I showed the maximum and significant genetic distance (38.82) from Cluster I. The result also indicated that cluster III had maximum genetic distance from cluster I. Cluster III the inter cluster distances between clusters II and III, I and III in that order were found to be significant. This confirmed the presence of genetically diverse coffee materials for the quality characters as pointed out by Dessalegn *et al.* (2008). The significant inter cluster distances indicated that there is a high opportunity for obtaining transgressive segregates and maximizing hetrosis by crossing accessions belonging to different clusters as there is a higher probability that unrelated genotypes would contribute unique desirable alleles at different loci (Peeters and Martinelli, 1989). Similarly, Rohman *et al.* (2004) also reported that high divergence between the clusters for 25 sorghum varieties which were grouped in to four clusters.

Similarly, Souza and Sorrells (1991) pointed out that categorizing germplasm accessions in to morphologically similar, more particularly genetically similar groups is useful for selecting parents

for crossing. Iftekharuddaula *et al.* (2002) reported that clustering pattern was not influenced by the geographic origin rather it was influenced by the pedigree of the breeding lines in rice which is again in support of present study. In contrast to present study, Falconer (1981) reported that genetic diversity has probably arisen through diversity in origin (geographical separation), ancestral relationship, gene frequencies and morphology. Zhuang *et al.* (2011) also reported that the clustering analysis done on Persian Wheat (*Triticum turgidum* ssp. carthlicum) accessions using EST-SSR Markers suggested that most of the accessions with adjacent geographic origins had the tendency to cluster together which is also in contrast to our finding. Souza and Sorrells (1991) pointed out that categorizing germplasm accessions in to morphologically similar, more particularly genetically similar groups is useful for selecting parents for crossing. Falconer (1981) reported that genetic diversity has probably arisen through diversity in origin (geographical separation), ancestral relationship, gene frequencies and morphology. These workers indicated that plants differing in either one or more of these factors would differ by a significant number of genes. Iftekharuddaula *et al.* (2002) reported that clustering pattern was not influenced by the geographic origin rather it was influenced by the pedigree of the breeding lines in rice. On the other hand, Zhuang *et al.* (2011) reported that the clustering analysis done on Persian Wheat (*Triticum turgidum* ssp. carthlicum) accessions using EST-SSR Markers suggested that most of the accessions with adjacent geographic origins had the tendency to cluster together. Three principal components (PC) PC1, PC2 and PC3 with eigen values of 4.23, 1.42 and 1.01, respectively, explained 81.37% of the total variance (Table 8). The first two principal components PC1 and PC2, with percent variability of 52.87 and 17.77%, respectively, explained 70.64% of the total variance. All the organoleptic quality attributes had contribution to genotype classification. However, some of the characters had relative values closer to unity in the first Principal Component (PC1) and thus contributed more to the classification (Chahal and Gosal, 2002). Aromatic quality, acidity, flavor

Table 7: Mahalanobis distance (D^2) of the three clusters for organoleptic quality trait

Clusters	I	II
II	4.66ns	
III	38.82**	23.21**

*D^2 significant at p = 0.05 (χ^2) = 14.07, **D^2 = Significant at p = 0.01 (χ^2) =18.48

Table 8: Eigenvalues, total variance, cumulative variance, and eigenvector for quality traits in Limu coffee germplasm accessions

	Eigen vector		
Trait	I	II	III
Aromatic intensity	0.29	0.38	-0.39
Aromatic quality	0.41	0.20	-0.33
Acidity	0.43	-0.23	-0.02
Astringency	-0.08	0.65	-0.13
Bitterness	-0.26	0.52	0.11
Body	0.24	0.28	0.83
Flavor	0.46	0.01	0.07
Overall standard	0.47	0.04	0.11
Eigen value	4.23	1.42	1.01
% of total variance	52.87	17.77	10.73
% cumulative variance	52.87	70.64	81.37

and over all standard had higher score as compared to others and contributed the highest variability in PC1. Therefore, these quality attributes were the cases of diversity among the coffee genotypes.

CONCLUSION

There has been no compressive and systematic quality analysis carried out in Limu coffee germplasm collections, though some promising selections were identified based on yield performances and disease resistances. The present study was undertaken using batch II involving 49-Limu coffee accessions with the prime aim to characterize them based on organoleptic quality traits and thereby determine the extent of genetic diversity. The findings showed significant differences among the coffee accessions for most quality characters, indicating the existence of organolepitc variability for future improvements. Moreover, there was high phenotypic coefficient of variation for astringency and bitterness. Medium phenotypic coefficient of variation was obtained for aromatic quality, acidity, flavor and over all standard. This is in contrast to the low phenotypic coefficients of variation recorded for aromatic intensity and body. It must be known that Limu coffee is of a restricted environmental importance. In an attempt to identify and develop suitable coffee varieties, it is necessary to include additional accessions and evaluate them over seasons based on disease resistance, yield performance, quality standards and other biochemical characteristics.

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