CHAPTER 3

PHENOTYPIC DIVERSITY FOR MORPHOLOGICAL AND AGRONOMIC TRAITS IN TRADITIONAL ETHIOPIAN HIGHLAND MAIZE ACCESSIONS

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3.1 ABSTRACT

Farmers in the highlands of Ethiopia have developed locally adapted maize varieties for more than 300 years. In order to assess the phenotypic diversity among traditional Ethiopian highland maize accessions, a total of 180 accessions were evaluated for agro-morphological traits in a replicated randomized complete block design. The accessions varied significantly for all of the measured traits. Cluster analysis revealed the presence of four major clusters. Accessions collected from the different regions were distributed over all the phenotypic clusters, reflecting wide variation within a particular region, but low differentiation among regions. The first principal component, which explained 40.4% of the total variation, was due to days to tasseling and silking, plant and ear height, leaf length and days to maturity. Traits directly selected by farmers (yield, kernels per row, rows per ear, and ear height) had the highest phenotypic coefficients of variation (PCV), whereas indirectly selected traits (ear diameter, days to tasseling and silking) showed lower PCV values. Number of kernels per row had high heritability and genetic advance as percent of the mean and could be used as selection criterion to increase grain yield. Overall, the study indicated the existence of ample trait diversity in highland maize accessions, which can be exploited by hybridization and selection.

Key words: Ethiopia, correlation, heritability, highland maize, phenotypic diversity

3.2 INTRODUCTION

Since its early domestication in Mexico, maize was introduced to many regions of the world where it has become adapted to a wide range of climates and agronomic conditions. It is believed that maize was first introduced to Ethiopia in the 16th or 17th century (Hafnagel, 1961). Since its introduction, it has gained importance as a food and feed crop in Ethiopia. Averages of the 2000/2001 national production estimates of the Central Statistical Authority (CSA, 2001) indicate that maize, with 1.4 million ha and 2.52 million t, accounts for about 20.9% of the total area and 32.6% of the gross annual grain production.

The highland zone of Ethiopia covers 20% of the total land devoted to maize cultivation and more than 30% of small-scale farmers in this region depend on maize for their livelihood (CSA, 1998). Despite its importance in the highlands of Ethiopia, only two improved open-pollinated maize varieties were developed in the past 10 years (Twumasi-Afriyie *et al*., 2001). The cultivars grown by highland farmers may not be as productive as commercial hybrids but they possess many useful adaptive traits, which have helped them to thrive in the difficult highland environments. They could, therefore, be useful material in the development of superior hybrids or open-pollinated varieties suited to this region.

In view of this fact, the Highland Maize Germplasm Collection Mission was launched in 1998 throughout the highlands of Ethiopia in collaboration with CIMMYT (Twumasi-Afriyie *et al*., 2001). As part of this project, 287 maize accessions were collected from farmers' fields throughout the highland regions of Ethiopia. Currently, there is no information on the extent of morphological and genetic variation among these accessions.

The objectives of this study, therefore, were (i) to evaluate and characterize these accessions for agro-morphological traits, (ii) to assess the extent of phenotypic and genotypic variability, heritability (broad sense) and expected genetic advance, and (iii) to classify and identify groups of similar accessions by means of cluster and principal component analysis. This information will be useful to identify genotypic groups for breeding purposes and to select a representative sample for molecular marker analysis.

3.3 MATERIALS AND METHODS

3.3.1 Plant materials and field evaluation

A total of 180 maize accessions collected from the Northern, Southern and Western highlands of Ethiopia were used in this study. The accessions were grown at Alemaya University in Ethiopia during the 2002 main cropping season in a randomized complete block design with two replications. Each accession was grown in two row plots. Each row had 25 plants, which constituted 44444 plants ha $^{-1}$, which is recommended for the testing site. From each accession, 20 competitive plants were selected at random to record 15 agro-morphological traits (Table 3.1).

3.3.2 Statistical analysis

The mean values of sampled observations for 15 agro-morphological data were analyzed using SAS (SAS, 1993). Statistical measures of variability such as genotypic coefficient of variability (GCV), phenotypic coefficient of variability (PCV), broad sense heritability (h^2), genetic advance as percent of the mean (GA), and genotypic and phenotypic correlations were calculated according to Singh and Chaudhary (1977). The standardized mean values (mean of each trait was subtracted from the data values and divided by the standard deviation) were used to perform cluster analysis (CA) and principal component analyses (PCA) using NCSS 2000 (Jerry, 2000) statistical software. The unweighted pair group method with arithmetic average (UPGMA) was used as clustering technique.

3.4 RESULTS

3.4.1 Morphological and agronomic variability

Analysis of variance revealed highly significant differences among the accessions for all the traits studied (Table 3.1). There was a wide range of expression across the accessions for all of the traits, including a 79 day range in maturity, 28 day range in male flowering, 155 cm range in plant height and 251 g range in 1000 seed weight. Grain yield exhibited one of the widest ranges (424.2 to 7313.2 kg ha^{-1}), possibly due to the specific adaptation of these accessions to various highland environments.

3.4.2 Phenotypic and genotypic coefficients of variation, heritability and genetic advance as percent of the mean

The PCV, GCV, h^2 and GA are presented in Table 3.2. Ear height showed the highest genotypic variability followed by number of kernels per row and yield ha^{-1}. The lowest GCV was recorded for ear diameter. The differences between GCV and PCV for all traits, except yield ha^{-1}, leaf width, 1000 seed weight and ear length were small indicating that these traits were less influenced by environment. High h^2 estimates were noted for the morphological traits (number of leaves, days to maturity, tasseling and silking and plant height). The lowest h^2 estimate of 17% was recorded for yield ha^{-1}. The GA that could be expected from selecting the top 5% of the accessions, varied from 10.1% for leaf width to 38.7% for ear height.

Table 3.1 Means, standard deviation of the means (St Dev), ranges and mean squares for 15 agro-morphological traits measured in 180 maize accessions

Traits	Mean	St Dev	Minimum	Maximum	Variety mean square
Days to tasseling	65.8	5.8	48.5	76.0	67.2**
Days to silking	71.1	5.3	56.5	80.5	55.8**
Plant height (cm)	218.3	28.5	155.0	310.0	1621.2**
Ear height (cm)	124.8	28.2	71.5	274.5	1584.0**
Leaf length (cm)	71.6	8.3	49.5	100.8	136.7**
Leaf width (cm)	9.00	0.8	6.4	12.8	1.3**
Number of leaves	6.1	0.3	5.2	6.8	0.1*
Foliage rating	6.1	1.0	3.0	7.0	2.0**
Days to maturity	144.4	15.0	108.0	186.5	452.2**
Ear diameter (cm)	3.9	0.3	3.3	4.9	0.5*
Ear length (cm)	18.1	1.7	11.2	22.0	11.4**
Rows per ear (no)	10.9	1.8	6.5	14.0	2.5**
Kernels per row (no)	28.6	5.8	18.0	41.0	30.1**
1000 seed weight (g)	298.1	36.0	159.0	410.0	2804.0*
Yield (kg ha^{-1})	2841.0	17.1	424.2	7313.2	1.9**

** & * Significant at $p = 0.01$, and $p = 0.05$, respectively.

Table 3.2 Estimates of phenotypic and genotypic coefficients of variability, heritability and genetic advance as percent of mean

Traits	PCV (%)	GCV (%)	h^2 (%)	GA (%)
Days to tasseling	9.3	8.3	78.5	17.0
Days to silking	7.9	7.0	77.8	14.3
Plant height (cm)	14.2	11.8	70.1	24.4
Ear height (cm)	25.8	18.8	53.0	38.7
Leaf length (cm)	13.5	9.2	45.8	18.9
Leaf width (cm)	11.6	4.9	17.7	10.0
Number of leaves (no)	13.9	12.9	86.9	26.6
Foliage rating	19.3	12.4	40.9	25.4
Days to maturity	10.8	9.9	84.1	20.4
Ear diameter (cm)	7.3	4.9	44.7	10.1
Ear length (cm)	12.2	5.7	21.6	11.7
Rows per ear (no)	19.4	13.2	46.4	27.2
Kernels per row (no)	22.0	18.3	69.5	37.8
1000 seed weight (g)	15.7	6.7	18.1	13.8
Yield (kg ha^{-1})	40.6	16.7	17.0	13.5

3.4.3 Genotypic and phenotypic correlations

There were significant genotypic and phenotypic correlations among the various traits (Table 3.3). At genotypic level yield was negatively and significantly correlated with all of the morphological traits, but positively and significantly correlated with all of the agronomic traits. When correlations between morphological traits were taken into account, all of the values between various trait pairs were significant and positive at both phenotypic and genotypic levels. Seed weight appears to contribute substantially to yield at the genotypic level, as the strongest positive and significant correlation of yield was recorded for this trait (r= 0.67).

Table 3.3 Genotypic (above diagonal) and phenotypic (below diagonal) correlation coefficients between 15 agro-morphological traits measured in 180 maize accessions

	DTS [a]	DSK	PLH	ERH	LFL	LFW	NRL	ERD	ERL	RWE	KLR	DYM	SDW	FGR	YDH
DTS		0.99	0.74**	0.74**	0.81**	0.95**	0.46**	0.19**	0.62**	0.22**	0.19**	0.74**	0.19**	0.83**	-0.48**
DSK	0.96**		0.76**	0.76**	0.83**	1.01**^{s}	0.02 ^{ns}	0.18**	0.63**	0.24**	0.22**	0.76**	0.20**	0.81**	-0.46**
PLH	0.53**	0.51**		0.99**	0.97**	0.93**	0.49**	0.33**	0.70**	0.52**	0.36**	0.73**	0.41**	0.88**	-0.44**
ERH	0.45**	0.44**	0.77**		0.99**	0.93**	0.47**	0.42**	0.60**	0.56**	0.40**	0.76**	0.65**	0.86**	-0.61**
LFL	0.49**	0.47**	0.63**	0.53**		1.34**	0.52**	0.34**	0.68**	0.52**	0.36**	0.85**	0.51**	0.95**	-0.59**
LFW	0.40**	0.41**	0.40**	0.33**	0.46**		0.09 ^{ns}	0.67**	0.94**	0.56**	0.49**	0.84**	0.44**	1.02**	-0.64**
NRL	0.14*	0.02 ^{ns}	0.21**	0.10 ^{ns}	0.20**	0.08 ^{ns}		0.26**	0.41**	0.43**	0.24**	0.41**	0.30**	0.83**	0.29**
ERD	0.10^{ns}	0.12 ^{ns}	0.22**	0.24**	0.14*	0.12 ^{ns}	0.03 ^{ns}		0.22**	0.40**	0.51**	0.18**	0.77**	0.19**	0.23**
ERL	0.19 **	0.21**	0.25**	0.26**	0.16*	0.05 ^{ns}	0.04 ^{ns}	0.23**		0.53**	0.39**	0.53**	0.68**	0.50**	0.17*
RWE	0.15*	0.17*	0.29**	0.27**	0.21**	0.22**	0.06 ^{ns}	0.32**	0.04 ^{ns}		0.95**	0.29**	0.64**	0.24**	0.23**
KLR	0.17*	0.19**	0.27**	0.27**	0.21**	0.17*	0.07 ^{ns}	0.34**	0.24**	0.65**		0.18**	0.60**	0.06 ^{ns}	0.41**
DYM	0.58**	0.60**	0.57**	0.52**	0.53**	0.32**	0.21**	0.09 ^{ns}	0.26**	0.15*	0.14*		0.39**	0.78**	-0.35**
SDW	0.05 ^{ns}	0.05 ^{ns}	0.12 *	0.13*	0.10 ^{ns}	0.16*	0.05 ^{ns}	0.31**	0.17*	0.19**	0.19**	0.14*		0.20**	0.67**
FGR	0.47**	0.50**	0.52**	0.45**	0.48**	0.33**	0.23**	0.12 ^{ns}	0.25**	0.08 ^{ns}	0.06 ^{ns}	0.49**	0.19**		-0.36**
YLD	-0.19 **	-0.19 **	-0.08 ^{ns}	-0.05 ^{ns}	-0.15*	-0.29**	0.04 ^{ns}	0.25**	0.08 ^{ns}	0.14*	0.22**	-0.17*	0.04 ^{ns}	-0.13*	

[a] DTS - days to tasseling, DSK - Days to silking, PLH - Plant height (cm), ERH - ear height, LFL - Leaf length (cm), LFW - Leaf width (cm), ERD - Ear diameter (cm), ERL - Ear length (cm), RWN - rows per ear (no), KLR - Kernels per row (no.), DYM - Days to maturity, SDW - 1000 seed weight (g), YDH - Yield per hectare (kg).

* Significant at $p = 0.05$, ** significant at $p = 0.01$, ^{ns} not significant

3.4.4 Cluster analysis

The trait means for the four clusters generated by UPGMA as clustering technique are given in Figure 3.1 and Figure 3.2. Cluster I was the biggest with 73 accessions, 90% of them were collected from the Southern and Western regions of Ethiopia. Accessions in this cluster expressed high values for morphological traits (days to tasseling, silking and maturity) and for all agronomic traits (ear diameter, ear length, rows per ear, kernels per row and grain yield). Cluster II contained 68 accessions (42.6, 27.9 and 29.5%, collected from Northern, Western and Southern Ethiopia, respectively). Accessions in this cluster expressed high values in days to tasseling, silking and maturity (Figure 3.1) but low mean values for all of the agronomic traits (Figure 3.2).

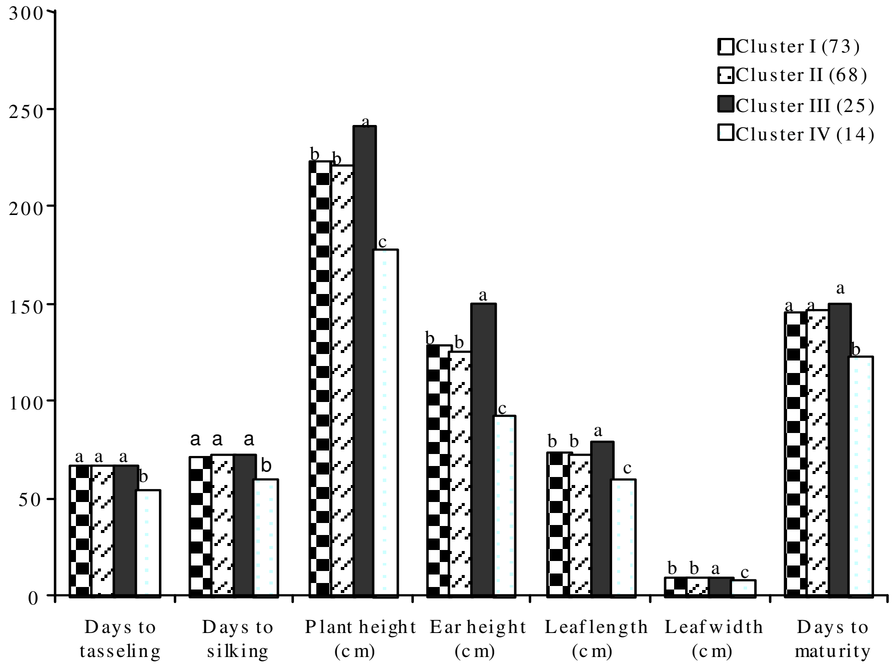

Figure 3.1 Mean values of morphological traits for traditional Ethiopian highland maize accessions for the four clusters generated through UPGMA as clustering techniques. Clusters having the same letter within each trait are not statistically significant at $p = 0.05$. Number in parenthesis (in the legend) indicates accessions

grouped in each cluster.

Cluster III contained 25 accessions (collected equally from Northern, Western and Southern Ethiopia) were tall and late maturing plants that had broad and long leaves. This cluster also gave the lowest mean values for all of the agronomical traits. In contrast, Cluster IV, comprising of 14 accessions, had low mean values for all of the morphological traits. High values were observed for all of the agronomic traits. Seventy percent of the accessions in cluster IV were collected from the Northern part of the country.

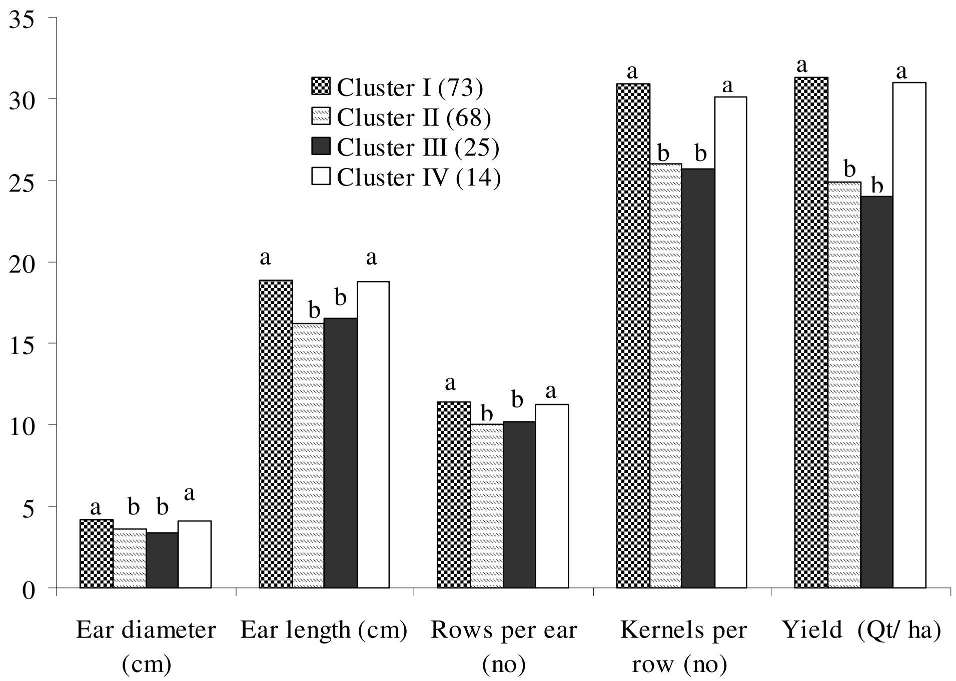

Figure 3.2 Mean values of agronomic traits for traditional Ethiopian highland maize accessions for the four clusters generated through UPGMA as clustering techniques. Clusters having the same letter within each trait are not statistically significant at p = 0.05. Number in parenthesis (in the legend) indicates accessions grouped in each cluster.

3.4.5 Principal component analysis

The first five principal components explained 75.1% of the total variation, with the first three components, with eigenvalues higher than 1.0, accounting for 62.8% of total variation (Table 3.4). Morphological traits such as days to tasseling and silking, plant and ear height, leaf length and days to maturity, were the major discriminatory traits associated with the first principal components axis, which accounted for 40.4% of the total variation, while agronomic traits (number of kernels per row, number of rows per ear, 1000 seed weight, ear diameter and yield) were important traits associated with the second principal component, which accounted for 15% of the total variation. The third principal component, which explained 7.4% of the total variation, was dominated by number of leaves, leaf width and grain yield.

Table 3.4 Eigenvector, eigenvalues, individual and cumulative percentage of variation explained by the first five principal components (PC) after assessing agro-morphological traits in 180 maize accessions

Traits	PC1	PC2	PC 3	PC 4	PC 5
Days to tasseling	-0.33	0.19	0.02	-0.04	-0.29
Days to silking	-0.33	0.18	0.02	-0.04	-0.30
Plant height (cm)	-0.35	0.01	0.01	-0.06	0.03
Ear height (cm)	-0.34	-0.02	-0.05	0.03	0.04
Leaf length (cm)	-0.34	0.07	-0.07	-0.06	0.17
Leaf width (cm)	-0.28	0.06	-0.37	0.08	0.23
Number of leaves	-0.13	-0.02	0.61	-0.49	0.38
Foliage rating	-0.30	0.15	0.26	0.04	0.16
Days to maturity	-0.32	0.13	0.11	0.04	-0.06
Ear diameter (cm)	-0.14	-0.40	0.00	0.29	0.19
Ear length (cm)	-0.19	-0.15	0.28	0.42	-0.49
Rows per ear	-0.18	-0.43	-0.29	-0.39	-0.02
Kernels per row	-0.17	-0.48	-0.23	-0.29	-0.21
1000 seed weight (g)	-0.13	-0.33	0.09	0.50	0.42
Yield (kg ha^{-1})	0.09	-0.42	0.43	-0.06	-0.26
Eigenvalue	6.1	2.3	1.1	1.0	0.9
Individual variation in %	40.4	15.0	7.4	6.5	5.8
Accumulated variation in %	40.4	55.4	62.8	69.3	75.1

3.5 DISCUSSION

Knowledge of the existing genetic variation and association between various agro-morphological traits and their heritability is vital for any breeding program. The accessions collected from different highlands of Ethiopia showed considerable variability for all examined morphological and agronomic traits (Table 3.1). Similarly, Lucchin *et al*. (2003) found significant differences within and between populations for all the traits measured in their study aimed to characterize 20 Italian maize populations for 34 morphological and agronomic traits. The broad range in the means of accessions for the various traits implies great possibility for the development of inbred lines, hybrid and/or open-pollinated varieties. The wide range in days to maturity (108 to 186.5) for example, suggest flexibility for the development of cultivars for the various highlands of Ethiopia with differing rainfall and length of growing season.

Genetic traits such as the genotypic coefficient of variability, heritability and genetic advance provide estimates of genetic variation of quantitative traits. Of all the traits evaluated in this study, grain yield appears to combine high values of PCV, intermediate GCV and low h^2 (Table 3.2). This is in agreement with the report of (Rebourg *et al*., 2001) in maize. Hallauer and Miranda (1988) summarized numerous estimates of heritability in maize. These range from less than 0.3 for grain weight and kernel depth to between 0.5 and 0.7 for plant height, ear height, kernel row number and days to flower. The relatively low estimates for yield indicate that selection for this trait in maize would be more difficult. In contrast, high h^2 values with increments in the range of about 69.5 to 86.9% were noted for most morphological traits. Greater than 60% for heritability for plant and ear height has been previously reported in maize by Rebourg *et al*. (2001). On the other hand, number of kernels per row exhibited moderate PCV, GCV and high h^2 and GA as percentage of the mean (Table 3.2), which indicated that it is under additive genetic control. Simple selection of the plants bearing higher number of kernels per row may lead to success in improving the trait up to 37.8%. This result was in agreement with previous studies of Arias *et al.* (1999) and Kumar and Kumar (2000). Therefore, highland maize breeders in Ethiopia should give more importance to kernels per row as selection criteria to increase grain yield.

The majority of the genotypic correlation coefficients were positive and highly significant (Table 3.3). However, only correlation coefficients greater than 0.71 or smaller than – 0.71 have been suggested to be biologically important (Skinner *et al.*, 1999), as more than 50% of the variation in one trait is predicted by the other (Snedecor and Cochran, 1980). In this study, such important correlations (at genotypic level) were found between days to 50% tasseling and days to 50% silking (0.99), plant height and ear height (0.99), plant height and leaf length (0.97), ear diameter and 1000 seed weight (0.77), and number of rows per ear and number of kernels per row (0.95). The genotypic correlations between morphological traits (plant height, ear height and days to maturity) with yield were negative and significant (Table 3.3), indicating that the possibility of developing high yielding varieties with short plant height, medium ear height and early maturing.

Unlike the present study where the accessions were grouped into a few clusters (Figure 3.1 and 3.2), Taba *et al.* (1998) showed the formation of 12 non-overlapping clusters by evaluating 249 Caribbean maize accessions. The limited clustering observed in this study may be attributed to several factors: (i) maize being open-pollinated, there is continuous gene exchange between adjacent fields, (ii) local farmers acquire new seeds from distant sources to meet their requirements, and (iii) there is a continuous seed supply by research organizations, non-governmental organizations and agricultural offices in these regions. Accessions collected from the Western and Southern regions (receiving high annual rainfall and long growing period) had the highest mean values for morphological traits. On the other hand, accessions collected from the Northern region (characterized by low annual rainfall and a short growing period) expressed the lowest mean values for all of the morphological traits. This result suggests that rainfall and growing season are the most important environmental factors differentiating traditional Ethiopian highland maize accessions. A similar result was reported by Ayana and Bekele (2000) in their study of morphological variation in sorghum collected from Ethiopia and Eritrea and noted that regional mean for plant height and days to maturity increases from North to South and from East to West, which followed the rainfall, temperature and seasonal patterns in Ethiopia (Tato, 1964).

The existence of broad morphological and agronomic diversity among the Ethiopian highland maize accessions is further substantiated by principal component analysis (Table 3.4), which indicated that the major contributing traits to the total variation are fairly distributed across morphological and agronomical traits. The major role of morphological traits in phenotypic variation is consistent with the work of Alika *et al.* (1993).

This study confirmed that traditional Ethiopian highland maize accessions display large amounts of variation for studied agro-morphological traits. The broad trait diversity evident among the maize accessions suggests ample opportunities for the genetic improvement of the crop through selection directly from the accessions and/ or the development of inbred lines for future hybrid programs. Grouping accessions into morphologically similar, and most likely genetically similar groups (Souza and Sorrells, 1991) is helpful for selecting parents for crossing. In addition, the study allowed the selection of representative accessions from different areas of Ethiopia, which will be studied using molecular markers. Therefore, the grouping of accessions by phenotypic diversity in the present study and the data from AFLP and SSR markers analysis will be used to classify the highland maize accessions into genetically related groups, which could be used for various breeding, collection and conservation programs in the highlands of Ethiopia.