

**MULTIVARIATE ANALYSIS OF QUANTITATIVE CHARACTERS VARIABILITY IN ETHIOPIAN BARLEY (*Hordeum vulgare* L.) LANDRACE: BASED ON REGIONS AND ALTITUDE**

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Barley (*Hordeum vulgare* L.) is Ethiopia's most important highland cereal crop and widely growing in most part of the country. Based on high levels of genetic and phenotypic diversity, the country is considered as center of diversity for barley. Assessments of the amount of genetic variation within and among populations are crucial for effective and efficient genetic improvement of the crop. Hence, this study was conducted to assess the diversity of barley landraces collected from various altitudes and regions of Ethiopia. A total of 585 barley landraces and 10 checks were evaluated using augmented randomized complete block design consisting of six blocks. All the 585 landraces were planted in un-replicated plots and the 10 checks were replicated six times (ones in each block) to estimate an error variance. Data on 13 quantitative characters were subjected to calculation of descriptive statistics, ANOVA and multivariate analysis (Unweighted Pair Group Method Analysis (UPGMA) cluster analysis and principal component analysis). There were significant differences (ANOVA,  $P < 0.01$ ) among landraces for plant height, 1000-seed weight, number of seeds per spike, days to heading and days to maturity. All the genotypes were grouped into five clusters where 74.02% of the accessions (433) fall in cluster I, IV and V. Early matured accessions were grouped in cluster I, while late matured, high yielding and tall accessions were clustered in cluster IV. The highest intra-cluster distance was 23.12 for

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cluster III whereas the highest inter-cluster distance was 57.37 between cluster IV and V. The first three principal components contributed 51.76% of the total variations observed among the genotypes. Principal component one (PC1) alone had contributed 22.56% of the total variations mainly due to plant height, 1000-seed weight, grain yield and peduncle length in their respective order. Principal component two (PC2) contributed 18.94% of the total variations mainly through spike density, number of kernels per spike, spike weight and days to maturity in their descending order. Principal component three (PC3) had contributed 10.94% of the total variations through total number of tillers per plant, number of seed-bearing tillers, days to 90% maturity and days to 50% heading. Altitude of the original landrace collection sites also significantly impacted the various quantitative characteristics studied. Regional differentiations were also evident among the landrace collections. These results reveal the existence of significant agro-morphological variations among the landraces included in this study. Based on the characters considered and populations evaluated, the marked diversity observed among the barley landraces in Ethiopia could be utilized in future crop improvement for various agronomically important traits. The information generated complements the robust barley breeding program of competitive, stable and climate-resilient varieties of end users' preferences in different agro-ecologies of Ethiopia.

*Keywords:* Barley, cluster analysis, genetic variation, landraces, principal component analysis

#### INTRODUCTION

Barley (*Hordeum vulgare* L.) belongs to the tribe *Triticeae* of the grass family *Poaceae* and has been cultivated for the last many years in Ethiopia (HARLAN, 1969). It is grown in almost all parts of the country; however, Arsi, Bale, Gojam, Gonder, Shewa, Tigray, Wellega and Wello are the main barley producing regions accounting for more than 85% of the country's barley production (LAKEW *et al.*, 1997). In the country, it is the fifth most important cereal crop after tef, maize, sorghum and wheat in terms of area coverage and production, in a given order (LAKEW *et al.*, 1997). Ethiopia is also considered as the center of diversity for barley (*Hordeum vulgare* L.) due to high levels of genetic and phenotypic diversity and strong genetic differentiation from Asian and north African populations (VAVILOV, 1951; HARLAN, 1969; ORABI *et al.*, 2007) and this attributed to the presence of diversity in altitude, soil types, climate, topography and geographical isolation for a long period of time (LAKEW *et al.*, 1997). Availability of appropriate genetic resources is very crucial for any crop improvement and evaluation of genetic resources for various traits and the assessment of the amount of genetic variation among and within populations are useful to perform more effective and efficient genetic improvement (HAUSSMANN *et al.*, 2004).

Genetic diversity studies can be used to determine the extent of variability in a breeding program and to identify parental lines for hybridization and introgression of desirable genes into the available genetic base (MOHAMMADI and PRASANNA, 2003; CHAKRAVORTY *et al.*, 2013). Moreover, genetic diversity studies are important to identify core collections for conservation and methodologies useful in diversity studies, including measures of genetic distance (similarity statistics), multivariate procedures (cluster analysis, principal component analysis, principal

coordinate analysis, and multidimensional scaling), types of data for multivariate analysis (if individual or combined data should be used), and the different tests to identify true clusters were extensively reviewed methodologies (MOHAMMADI and PRASANNA, 2003).

Multivariate analysis refers to statistical technique that is widely used to analyse data which arise from more than one variable. Among the multivariate techniques, cluster analysis, principal component analysis (PCA), principal coordinate analysis (PCoA) and multidimensional scaling (MDS) are commonly employed techniques (GRAHIC *et al.*, 2013; MARUTHI *et al.*, 1999; MOHAMMADI and PRASANNA, 2003). Clustering is also used to summarize information on relationships between objects by grouping similar units so that the relationships may be easily understood and communicated. Cluster analysis is a multivariate analysis and it is widely used to describe genetic diversity based on similarities or differences among genotypes (PEETERS and MARTINELLI, 1989). As morphological and physiological variations routinely occur in crop species, PCA eliminates redundancy in data sets and gives the reliable patterns of distribution (ADAMS, 1995).

In Ethiopia, the barley landraces, which have been collected from different parts of the country, has been conserved *ex situ* in the Ethiopian Biodiversity Institute (EBI) gene bank. The value of the collected germplasm in the gene bank depends on the information generated through characterization and evaluation of different traits. However, majority of the collected materials were not yet systematically characterized in detail for their morphological and genetic diversity (ABEBE *et al.*, 2010).

Therefore, this study was initiated with the objective of assessing the existing quantitative agro-morphological variability using large scale testing materials collected since 1964 from 13 major barley growing zones and different altitudinal ranges using multivariate data analysis techniques. This will serve to expand varietal choice options for researchers in developing barley cultivars with better agronomic performances and resistance to biotic stresses for a climate change resilient agricultural system.

## MATERIALS AND METHODS

### *Genetic materials*

For the study a total of 585 barley landraces collected since 1964 to 2017 from 13 agro-ecological zones of Ethiopia with altitudinal ranges from 1430 to 2950 m asl (meters above sea level), and conserved *ex-situ* in the Ethiopian Biodiversity Institute (EBI) gene bank were used. The landraces were selected based on the region they originated from and altitude (Table 1). Ten checks, six of which were obtained from Holetta Agricultural Research Center (HARC) (HB-42, Shegie, HB-1966, HB-1964, HB-1307 and Ardu-12-60B) and the remaining four (Aruso, Abdane, Dafo and Guta) that were obtained from Sinana Agricultural Research Center (SARC) were also included. These checks were selected from improved varieties released in the country and commonly grown around and adapted to the experimental sites. Ninety eight percent of materials used in the study were landraces and majority of them (98.6%) have complete geographic coordinates and passport data (Table 1). The treatments were coded C1 –C10 for ten checks and as T1-T585 for 585 landrace accessions in ascending order based on the passport code.

Table 1. Regions, altitude (meter above sea level (masl)) of origin and number of accessions used for this study

Region/Zone	Number of accessions by altitude groups (m.a.s.l)				Total accessions
	Group I (≤1500)	Group II (1501-2000)	Group III (2001-2500)	Group IV (>2501)	
Arsi	-	10	45	28	83
Bale	2	14	25	5	46
Gamo Gofa	-	17	14	5	36
Gojam	-	8	39	14	61
Gonder	1	11	20	13	45
Hadiya	-	1	11	9	21
Hararghe	-	18	30	9	57
Jimma	1	14	5	-	20
Shewa	1	12	23	39	75
Sidama		10	4	3	17
Tigray	2	27	24	11	64
Wellega		8	16	5	29
Wello		9	11	11	31
Total	7	159	267	152	585

#### Experimental Site

Experiments were conducted at SARC on-station and at Bale-Goba on-farm research site, in south-east highland of Ethiopia. Sinana is located at a distance of 463 km South-East of Addis Ababa and 33 km east of Robe town (capital city of Bale zone) to the east. Sinana is located at an altitude of 2,400 m asl and located at 7°7'N latitude and 39°40'E longitude. The area is characterized by cambic vertisol with pH ranges of 6.3-7.0 (slightly acidic) at the depth of 0-15cm. The monthly averages of minimum and maximum temperatures are 9.42 and 21.16°C, respectively. Total annual rainfall for the last 5 consecutive years ranged from 750-1000mm (average 860mm). Bale-Goba is located 60 km from Sinana and located at 7°0'N latitude and 39°59'E longitude and an altitude of 2,743 masl. The soil of the area is characterized by pellic and chromic vertisol. Both areas represent the highest altitude for barley production areas and have bimodal rainfall pattern.

#### Experimental procedures

The experiments were laid out in augmented randomized complete block design (FEDERER and RAGAVARAO, 1975) consisting of six blocks in which the 585 landraces were planted in un-replicated plots and the ten checks were replicated six times (ones in each block) to estimate an error variance. The plot size used was one row with 1.75 m length and 0.2 m space between rows. Seeds were planted manually at seeding rate of 100 kg/ha when adequate moisture was available. At SARC, the seeds were planted on 11 August 2018 and 14 August 2019 and at Bale-Goba on 21 August 2018 and on 17 August 2019. Recommended agronomic practices like fertilizer application and weed control were applied to both experiments (SARC, 2004).

*Quantitative data collected*

Phenological and morphological characteristics were determined according to barley descriptors (IPGRI, 1994) based on plant based and plot based traits (Table 2). For plant based traits i.e. plant height, total number of tillers per plant, number of effective tillers (seed-bearing) per plant, number of kernels per spike, spike length, spike density, spike weight and peduncle length were considered. Ten randomly selected plants from central part of row were tagged at the early stage and measured timely according to the traits used. The averages were used for the analysis. For plot based traits, 1000- seed weight and grain yield per plant were taken from the whole row for each accession and converted into per hectare bases for the analysis. In addition days to heading and days to maturity were counted from emergence to 50% heading and 90% physiological maturity, respectively.

Table 2. List of quantitative characters recorded along with their code and description

Character	Code	Character descriptions
Days to 50% heading (count)	DH	Recorded as the number of days from planting to the date on which approximately 50% tillers had produced spikes..
Days to 90% maturity (count)	DM	Recorded as the number of days from planting up to the time when 90% of the plants in a row had reached physiological maturity (the stage when colour of plant changed from green to golden yellow and its tillers could break easily with hands).
Grain filling period (count)	GFP	Recorded as the difference between days to physiological maturity and days to heading.
Number of tillers per plant (count)	NTPP	Recorded as total number of tillers per plant for each accession.
Number of fertile tillers per plant (count)	NFTPP	Recorded as the total number of seed-bearing tillers for each accession'
Number of seeds per spike (count)	NSPS	Recorded as the total number of kernels in the main spike for each accession.
Peduncle length (cm)	PL	Measured as the length of peduncle in centimeters from the last node to the base of spike for each accession.
Spike length (cm)	SL	Recorded as length of the spike in cm on the tallest culm (excluding awn).
Spike density	SD	Ratio between the numbers of seeds per spike over spike length.
Spike weight	SW	Weight of the main plant spike in gram (g).
Plant height (cm)	PH	Height in centimeter from the soil surface to the tip of the spike (awn excluded) of the tallest culm.
Thousand seed weight (g)	TSW	Recorded as weight in grams of 1000 seeds randomly taken from each accession at 12.5% moisture content.
Grain yield	GY	The grain yield of each accession after moisture content was adjusted to 12.5% and expressed in grams.

*Data analysis*

All quantitative data were analyzed using Genstat v15 (VSN International, GenStat.co.uk) and NCSS (2019). Data were analyzed using the restricted maximum likelihood (REML) model to fit a mixed model with standard controls and experimental site as a fixed effect and non-replicated accessions as random effects (COMADRAN *et al.*, 2008). The checks were used for estimating error variance in mixed models. The REML model produced best linear

unbiased predictions (BLUPs), which can handle unbalanced data while accounting for differences in the amount of data available for each accession (ETTEN *et al.*, 2008). A mixed model procedure was employed to fit analysis of variance of the form:

$$Y_{ij} = \mu + \alpha_i + \beta_j + e_{ij}$$

Where;  $Y_{ij}$  is response variable;  $\mu$  is general mean;  $\alpha_i$  is the fixed effect of  $i^{\text{th}}$  checks and random effect of germplasms,  $\beta_j$  is the random effect of  $j^{\text{th}}$  block and  $e_{ij}$  is random error.

Analysis of variance for accessions and the region was made for 13 quantitative characteristics as described in Table 3. Estimates of  $\sigma^2_g$  (genetic variance component for genotypes (landraces and checks)),  $\sigma^2_c$  (genetic variance component for checks) and  $\sigma^2_t$  (genetic variance component for tests (accessions)) were obtained by computing the obtained sum of squares to their expectancies, as shown below;

$$\sigma^2_g = \frac{MSg - MSe}{Block}$$

$$\sigma^2_c = \frac{MSc - MSe}{Block}$$

$$\sigma^2_t = \frac{MSt - MSe}{Block}$$

Where,  $MS_g$  is genotypes (landraces + checks) mean square,  $MS_e$  is error mean square,  $MS_c$  is checks mean square and  $MS_t$  is test (accession) mean square. The mean squares of the thirteen regions were tested against pooled mean squares of accessions within regions. The pooled mean squares for accessions within regions of origin and the mean squares of accessions within each region were tested against the pooled within region error mean squares.

Means, ranges of means, and percent coefficient of variation (computed as a ratio of standard deviation of each characteristic to the corresponding entire data mean and expressed as a percentage) for all the characteristics were computed for each region of origin and for all the data. The regional means were compared using Duncan's multiple range test (AYANA and BEKELE, 2000).

Table 3. ANOVA table for sum of squares and their respective expectancies for the statistical genotype model (FEDERER and RAGAVARAO, 1975)

Source of variation	DF	Mean square	Expected mean square
Accessions (test (t))	t-1	MSt	$\sigma_t^2 + \sigma_e^2$
Genotypes (g)	g-1	MSg	$\sigma_g^2 + \sigma_e^2$
Checks (c)	c-1	MSc	$\sigma_c^2 + \sigma_e^2$
Tests vs. checks (t vs. c)	1	MSt vs. C	$\sigma_t^2 + \sigma_c^2 + \sigma_e^2$
Blocks (b)	b-1	MSb	-
Error	(b-1)(c-1)	MSe	$\sigma_e^2$
Total	n-1		$\sigma_g^2 + \sigma_b^2 + \sigma_e^2$

MSt = mean square of accessions (tests), MSg = mean square of genotypes, MSc = mean square of checks, MSb = mean square of blocks, MSt vs. c = mean square of tests vs. checks, MSe = mean square of error,  $\sigma_g^2$  = genotypic variance component,  $\sigma_t^2$  = accessions variance component,  $\sigma_e^2$  = expected error variance (MSe).

### *Cluster analysis*

The data was standardized to mean of zero (0) and a variance of one (1) before computing multivariate ANOVA in which two or more variables were analyzed at a time to avoid differences in scales. Five hundred eighty five accessions and thirteen regions of origin were grouped into respective classes. The values of Pseudo F statistic (PSF) and Hotellin's pseudo  $T^2$  statistic were used for defining optimum number of clusters. Cluster analysis was undertaken using the hierarchical cluster analysis.

### *Principal component analysis (PCA)*

The PCA was performed to reduce the number of variables into a few correlated components that can explain much of variability and to estimate the relative importance and contribution of every quantitative character to the total variance and illustrate the existing agromorphological diversity among the 585 barley landraces. It was computed using the correlation matrix to define the pattern of variation among landraces based on the means of quantitative characters. It also helps to identify characters that load the most in explaining the observed variation.

## RESULTS

### *Analysis of variance*

The analysis of variances indicated the presence of significant variations ( $P \leq 0.001$ ) among accessions (tests), genotypes (accessions and checks), checks, accessions (tests) *vs.* checks for 13 quantitative characters evaluated. However, there was no significant difference in grain filling period in genotypes and checks, total tiller per plant, fertile tillers per plant and spike weight in checks and number of fertile tillers per plant, spike density and weight in accessions *vs.* checks (Table 4).

The analysis also revealed that there were significant differences ( $P \leq 0.001$ ) among regions of origin in the 585 barley landraces for all quantitative characters studied (Table 5). However, within-region differences were significant ( $P \leq 0.001$ ) among the landraces within thirteen regions of origin for thirteen quantitative characters except within two regions for days to maturity and peduncle length in two regions, and for grain filling period in three regions (Table 5). In terms of altitudinal classes, significant differences ( $P \leq 0.001$ ) were observed among landraces were observed for all quantitative characters assessed except grain filling period in altitude class I (<1500 meter above sea level (masl)) and peduncle length in altitude class II (between 1501-2000 masl) (Table 5).

Duncan's multiple tests were computed for region and altitude class means for all quantitative characteristics as shown in Table 6. Differences were observed for quantitative characters among regions and altitude classes. Notably, much more regional and altitudinal differentiation among landraces was observed for tiller per plant, plant height, number of seeds per spike, days to heading and days to maturity. These were the most quantitative characters which contributed variance among landraces. Furthermore diverse regions of origin and altitude classes favored the development of different quantitative Characters.

In this study we have also observed that, there were high percent of coefficients of variation (CV) among regions and within each region of origins of landraces for grain yield, spike weight, spike density and number of seeds per spike. The highest CV was observed in

Table 4. Analysis of variance of 13 quantitative characteristics studied in barley landraces

Source	DF	DH	DM	GFP	PH	NTPP	NFTPP	PL	SL	SD	SW	NKPS	TSW	GY
Accessions (Tests)	584	58.63**	110.86**	50.03**	94.84**	6.37**	3.99**	12.74**	2.03**	6.72**	0.22**	274.29**	62.94**	298.60**
Genotypes	594	66.11**	119.75**	50.03 <sup>ns</sup>	100.09**	6.30**	3.95**	12.89**	1.21**	7.10**	0.24**	291.90**	64.42**	198.44**
Checks	9	449.61**	417.95*	87.40 <sup>ns</sup>	448.65**	2.09 <sup>ns</sup>	1.34 <sup>ns</sup>	22.60**	6.61**	16.06**	0.43 <sup>ns</sup>	631.19**	164.80**	140.50**
Tests vs. Checks	1	5.69*	1230.05*	214.38**	207.17**	42.07*	11.25 <sup>ns</sup>	67.90*	6.19*	2.10 <sup>ns</sup>	0.18 <sup>ns</sup>	2149.30*	31.76*	174.92**
Block	5	2.29	23.04	22.52	358.64	94.51	2.47	30.10	4.14	1.40	44.37	64.51	53.21	286.27
Error	45	17.51	48.32	74.51	44.66	0.67	0.44	9.22	0.64	0.14	0.22	71.34	19.76	831.00
Total	648	876.03	1274.37	1781.39	1133.24	124.33	73.62	751.31	214.31	276.91	529.26	1701.50	3102.77	1998.46
Mean		67.93	112.60	44.60	96.60	4.10	3.20	29.60	8.31	4.71	1.57	37.40	31.01	60.87
SE		0.20	2.60	0.20	0.30	0.05	0.04	0.20	0.04	1.20	0.48	0.40	0.20	2.96
CV (%)		6.16	6.17	19.33	6.92	19.12	20.56	10.26	9.66	20.49	25.31	22.58	14.33	26.16

\* \*\* ns = significant at P = 0.05; P = 0.01 level, and non-significant respectively; MS<sub>t</sub> = mean square of test (accessions), MS<sub>e</sub> = mean square of genotypes, MS<sub>c</sub> = mean square of checks, MS<sub>b</sub> = mean square of blocks, MS<sub>t</sub> vs. c = mean square of tests vs. checks, MS<sub>e</sub> = mean square of error,  $\sigma^2_g$  = genotypic variance component,  $\sigma^2_a$  = accessions variance component,  $\sigma^2_e$  = expected error variance (MSe), DH = Days to 50% heading, DM = Days to maturity (days), GFP = Grain filling period (days), NFTPP = Number of fertile tiller per plant (number), NKPS = Number of kernels per spike (number), NTPP = Number of tiller per plant (number), PH = Plant height (cm), PL = Peduncle length (cm), SL = Spike length (cm), SD = Spike density, SW = Spike weight, TSW = Thousand seed weight (g), GY = Grain yield per plant (g).

Table 5. Analysis of variances for 13 regions and 4 altitude classes of origin based on 13 quantitative characters of barley landraces

Region	DF	DH	DM	GFP	PH	NTP	NFTPP	PL	SL	SD	SW	NKPS	TSW	GY
Arsi	82	35.60**	76.04**	60.73*	72.19**	4.42**	2.62**	9.79**	1.92**	5.36**	0.19**	224.57**	49.02**	1372.94**
Bale	45	19.54**	73.42**	67.25**	80.23**	4.39**	2.54**	13.14**	1.31**	4.96**	0.12**	145.69*	35.43**	1926.56*
Gamo Gofa	35	11.89*	57.81**	51.19 <sup>ns</sup>	79.17*	4.70**	2.59*	13.93**	2.21**	5.10**	0.14*	216.65*	74.85**	2470.20**
Gojam	60	93.63**	135.22**	87.60**	136.83**	8.61**	6.41**	14.00*	0.19**	2.27**	0.19**	104.25*	67.12**	2052.98**
Gonder	44	122.33**	159.01**	142.39**	53.91**	8.71**	4.76**	12.95**	1.01**	3.76**	0.21**	208.20**	59.13**	2106.96**
Hadiya	20	26.72*	73.81 <sup>ns</sup>	52.07 <sup>ns</sup>	111.92**	4.32**	4.25**	12.72 <sup>ns</sup>	4.25**	8.86**	0.31*	303.42**	91.22**	1081.10*
Hararghe	56	20.98*	80.82**	51.04*	50.10*	3.65**	1.64**	7.29*	1.38**	5.21**	0.26**	183.26**	27.44**	1586.39**
Jimma	19	14.01**	89.71 <sup>ns</sup>	83.90 <sup>ns</sup>	64.58**	3.79**	1.87**	11.05 <sup>ns</sup>	2.56**	8.24**	0.21*	365.25**	27.44**	1212.21*
Shewa	74	61.45**	97.94**	78.22**	72.20**	8.67**	5.08**	13.56**	2.07**	4.89**	0.23*	209.54**	27.44**	1390.05**
Sidama	16	66.50**	131.60**	76.87**	84.19*	3.97**	2.82**	21.50**	2.34**	5.22**	0.55**	201.91*	33.41**	1939.22**
Tigray	63	36.32**	107.01**	62.82**	99.53**	7.20**	5.28**	10.39**	1.21**	2.36**	0.17**	98.69**	36.04**	1789.86**
Wellega	28	40.77**	117.52**	90.38**	74.93**	5.55**	2.73**	8.52**	1.19**	3.61**	0.14**	198.28**	69.63**	1886.49**
Wello	30	46.29**	70.18**	83.44**	12.02**	6.18**	5.01**	15.77**	2.69*	6.89**	0.24**	235.54**	52.96**	1090.46**

## Altitude classes

<1500	6	81.22**	146.25*	90.03 <sup>ns</sup>	151.03*	6.20**	7.06**	8.31*	0.82*	2.11*	0.18*	117.10*	87.44**	1300.55*
1501-2000	158	40.03**	90.48**	68.89**	96.89**	6.14**	4.05**	13.59 <sup>ns</sup>	2.17**	7.01**	0.19**	267.10**	63.24**	1749.18**
2001-2500	266	54.16**	109.61**	84.02**	100.68**	5.99**	3.65**	11.76**	2.25**	7.17**	0.21*	277.40**	69.07**	1558.76**
>2500	151	64.87**	106.15**	105.13**	78.07**	7.22*	4.37**	13.20**	1.56**	5.55**	0.20**	258.34**	43.92**	1929.89**

\* \*\*, ns = significant at P = 0.05; P = 0.01 level, and non-significant respectively; DH = Days to 50% heading, DM = Days to maturity (days), GFP = Grain filling period (days), NFTPP = Number of fertile tiller per plant (number), NKPS = Number of kernels per spike (number), NTPP = Number of tiller per plant (number), PH = Plant height (cm), PL = Peduncle length (cm), SL = Spike length (cm), SD = Spike density, SW = Spike weight (g), TSW: Thousand seed weight (g), GY = Grain yield per plant (g).

Table 6. Regional and altitude classes means for 13 quantitative characters of barley landraces

Region	DH	DM	GFP	PH	NTPP	NFTPP	PL	SL	SD	SW	NKPS	TSW	GY
Arsi	68.94c	114.90bc	45.95a	98.94a	4.10b	3.08cd	30.30b	8.39b	4.92bc	1.66a	39.74b	31.06b	64.83c
Bale	64.76h	109.70f	44.96b	96.38c	3.98b	3.04d	29.43c	7.87d	5.18bc	1.58ab	48.44a	30.71c	78.30a
Gamo Gofa	65.68e	107.90i	42.25g	98.18bc	4.12b	3.15cd	29.50d	8.57ab	4.66c	1.55ab	38.33d	29.63c	70.44b
Gojam	65.45ef	108.90gh	43.5c	95.45f	4.77a	3.60ab	29.85cd	8.71a	3.15f	1.28c	36.56d	34.91a	69.73b
Gonder	67.00d	109.80ef	42.81f	94.32g	5.01a	3.76a	29.23ef	8.53ab	3.74d	1.39b	39.42c	31.9ab	60.19d
Hadiya	69.98b	114.60c	44.67c	97.3cd	3.98b	3.16cd	29.66cd	8.39b	4.42d	1.64a	34.78e	32.53ab	57.38e
Hararghe	69.75bc	113.90d	44.16d	98.89b	3.49cd	2.55f	29.83cd	7.45e	6.66a	1.69a	31.09f	26.81de	53.35g
Jimma	65.32f	109.50g	44.20cd	99.22a	3.62c	2.86e	29.78cd	8.29c	4.98bc	1.63a	31.11e	31.28b	55.66f
Shewa	71.47a	116.60a	45.16b	98.01c	4.74a	3.42c	30.02c	8.27c	5.04bc	1.66a	39.13b	29.47d	63.93cd
Sidama	69.67bc	112.30d	42.33g	98.22b	4.03b	2.90d	30.57a	8.19cd	4.89c	1.48b	38.62c	27.7de	62.16cd
Tigray	65.18g	110.80e	45.64ab	90.36h	4.62b	3.52b	27.42f	8.80a	3.16f	1.38b	37.11d	34.26a	69.14b
Wellega	64.95h	108.70h	43.71de	97.05cd	3.90bc	3.14cd	30.54a	8.68a	3.70e	1.41b	39.66bc	33.82a	65.71c
Wello	69.98b	115.50b	45.47ab	96.86d	4.06b	3.27c	30.27b	8.17cd	5.59b	1.66a	43.10b	27.21de	64.69c
<b>Altitude classes</b>													
<1500	64.15bc	106.40c	42.23c	94.32b	5.17a	3.99a	29.35ab	8.31a	3.45b	1.36b	28.24c	34.89a	50.71d
1501-2000	65.79b	109.70b	43.88b	95.72ab	4.19ab	3.19bc	29.13ab	8.37a	4.42ab	1.48ab	35.09b	31.69b	61.41c
2001-2500	67.38b	112.30ab	44.92a	97.29a	4.19ab	3.16b	29.73a	8.34a	4.54a	1.53a	36.10b	31.57b	82.05a
>2500	70.50a	115.00a	44.48ab	96.45a	4.49a	3.37b	29.98a	8.27ab	4.98a	1.63a	39.71a	29.25c	69.49b

DH = Days to 50% heading, DM = Days to maturity (days), GFP = Grain filling period (days), NTPP = Number of fertile tiller per plant (number), NFTPP = Number of kernels per spike (number), NKPS = Number of tiller per plant (number), PH = Plant height (cm), PL = Peduncle length (cm), SL = Spike length (cm), SD = Spike density, SW = Spike weight (g), TSW = Thousand seed weight (g), GY = Grain yield per plant (g).

Table 7. Percent coefficient of variations for 13 quantitative characters in barley landraces based on regional and altitude classes

Region	DH	DM	GFP	PH	NTPP	PL	SL	SD	SW	NKPS	TSW	GY
Arsi	5.77	5.42	16.62	6.18	14.55	9.19	10.43	22.42	21.84	20.78	13.16	44.34
Bale	3.41	7.20	17.89	5.30	14.27	9.84	7.89	24.05	19.65	23.58	12.16	45.47
Gamo Gofa	3.72	5.27	16.72	6.55	14.22	11.70	10.42	32.32	22.58	30.46	12.76	31.18
Gojam	7.07	6.95	21.21	10.47	14.61	10.42	7.89	35.13	26.14	28.47	14.75	44.08
Gonder	9.15	6.79	24.69	6.78	16.13	11.86	8.40	30.42	23.16	27.98	15.11	40.92
Hadiya	6.41	5.38	15.87	6.02	11.50	9.40	8.60	21.45	27.29	21.49	14.18	38.89
Hararghe	5.58	5.63	15.99	5.29	12.68	8.71	11.29	19.62	20.33	19.29	14.43	38.17
Jimma	3.77	7.79	18.51	4.50	22.54	7.95	8.08	24.23	23.66	21.18	9.08	49.20
Shewa	5.65	5.57	19.27	7.02	19.12	11.13	9.40	17.77	23.81	16.98	14.15	50.43
Sidama	6.26	5.36	18.09	7.43	16.33	9.30	8.36	29.07	27.92	25.57	11.93	35.42
Tigray	4.83	6.07	16.50	7.18	10.84	10.38	7.40	28.49	27.64	20.97	12.05	40.48
Wellega	5.34	6.58	20.02	4.90	9.76	8.57	8.71	30.94	19.73	22.57	10.55	37.85
Wello	6.60	6.67	19.94	4.24	18.33	11.77	11.08	30.07	23.83	24.95	15.36	60.82
<b>Altitude classes</b>												
<1500	3.42	6.84	17.42	7.75	12.13	7.79	6.21	23.83	20.70	23.31	13.84	27.13
1501-2000	5.25	6.70	18.40	6.08	12.86	14.10	9.57	25.33	21.73	20.38	12.25	40.59
2001-2500	5.95	5.98	18.73	7.14	14.81	14.16	9.05	27.74	23.11	24.91	13.74	41.20
>2500	6.87	6.07	20.32	6.67	17.95	20.28	9.57	22.24	25.24	22.09	15.56	50.25

DH = Days to 50% heading, DM = Days to maturity (days), GFP = Grain filling period (days), NTPP = Number of fertile tiller per plant (number), NKPS = Number of kernels per spike (number), NTPP = Number of tiller per plant (number), PH = Plant height (cm), PL = Peduncle length (cm), SL = Spike length (cm), SD = Spike density, SW = Spike weight (g), TSW = Thousand seed weight (g), GY = Grain yield per plant (g).

grain yield, spike density, number of seeds per spike, spike weight, total tillers per plant, seed bearing tillers per plant and grain filling period in some of the regions and altitudinal classes (Table 7).

#### *Variation within regions of origin and altitudinal classes*

The estimates of genetic variation among regions of collection of landraces indicated highly significant differences ( $P < 0.001$ ) for all quantitative characteristics assessed in all regions, except spike weight in nine regions, grain yield in Bale, Jimma and Wello, days to maturity, peduncle length and 1000-seed weight in Hararghe (Table 8).

Analysis of estimation of genetic variances based on altitude classes (Table 8) indicated significant differences ( $P < 0.001$ ) among altitude classes for all quantitative characters except for number of tillers per plant, peduncle length, spike density and spike weight at altitude class I (<1500 masl) and number of fertile tillers at altitude class IV (>2500 masl). High genetic variations were also observed within each altitude class for the quantitative characteristics studied.

#### *Cluster analysis for landraces*

Hierarchical cluster analysis technique was used to see the aggregation patterns of 585 barley landraces. Accordingly, all the landraces were grouped into five clusters as shown in Table 9. Numbers of landraces per cluster varied from 52 landraces in cluster III to 168 landraces in cluster I. Means for quantitative characters of accessions in each cluster are presented in Table 10. Cluster I consist of 168 landraces, which is 28.72% of the total experimental materials. It is characterized as having landraces with moderately longer plant height and light seed and early maturing types. Landraces grouped under cluster I were distributed over all regions and majority of them were found in altitudinal class II (1501-2000) and III (2001-2500). Cluster II accounts for 17.09% of experimental materials with smaller number of tillers per plant and shorter grain filling period. Landraces with long maturity period, higher number of tillers, long spikes and relatively high grain yield were grouped in cluster III which account for 8.89% (52 landraces) of experimental materials. Landraces under cluster III distributed over 10 regions. Cluster IV had 154 landraces (26.32% of population) characterized by landraces with longer grain filling period, days to 90% maturity, plant height, peduncle and heavier seeds. Cluster V included 111 landraces (18.97% population) which comprised moderate plant height and number of tillers per plant, longer peduncle and spike (Table 10).

#### *Cluster analysis for regions of origin*

Regional cluster analysis categorized the thirteen regions of origin of barley landraces into five clusters based on thirteen quantitative characteristics studied as shown in Table 10. Hararghe, Sidama and Tigray were grouped under cluster I which is characterized with moderately longer plant height and light seed and early maturing types. Cluster II characterized by accession with smaller number of tillers per plant and shorter grain filling period in which regions like Gonder, Wello and Wellega were grouped. Landraces with long maturity period, higher number of tillers, long spikes and relatively high grain yield were grouped in cluster III in which four regions namely Bale, Gojam, Hadiya and Jimma were clustered.

Table 8. Estimates of genotypic variances for regions of origin and altitude classes based on quantitative characters

Region	DH	DM	GFP	PH	NTPP	NFTPP	PL	SL	SD	SW	NKPS	TSW	GY
Arsi	13.21**	26.35**	45.53**	22.19**	27.36**	0.21**	3.85*	1.23*	4.14**	0.06 <sup>ns</sup>	39.48**	13.21**	645.34**
Bale	21.15**	34.17**	37.83*	14.18**	19.58**	0.16*	4.17**	0.57**	3.41**	0.11 <sup>ns</sup>	43.38**	5.42*	658.56 <sup>ns</sup>
Gamo Gofa	43.21**	12.03*	27.62*	13.61*	0.23**	0.03*	5.05*	0.22**	2.83**	0.02 <sup>ns</sup>	71.40**	32.08**	198.78**
Gojjam	33.36**	26.67**	37.96**	18.35*	0.11**	0.02**	2.89*	0.17*	1.05**	0.08 <sup>ns</sup>	74.34**	12.64*	135.97**
Gonder	23.17**	42.35**	18.41**	39.13**	0.09**	0.37**	0.87*	0.25*	2.46**	0.11**	47.85**	11.34*	150.00**
Hadiya	17.65**	9.13*	13.39*	22.17**	0.17*	0.01*	4.65 <sup>ns</sup>	0.31**	7.96**	0.11 <sup>ns</sup>	45.06**	13.04*	583.20*
Hararghe	11.23**	1.58 <sup>ns</sup>	6.41*	14.21**	0.13*	0.26*	5.84 <sup>ns</sup>	0.16*	3.50**	0.14**	7.81**	4.37 <sup>ns</sup>	102.24**
Jimma	30.21**	31.42*	28.03*	19.11*	0.11*	0.08*	1.74*	0.24*	6.78**	0.06 <sup>ns</sup>	66.41**	7.62*	167.21 <sup>ns</sup>
Shewa	19.53**	22.89**	42.47*	47.86**	1.29*	0.26*	3.35*	0.71**	4.09**	0.07*	77.48**	51.77**	755.95**
Sidama	35.00**	72.65**	35.88*	36.69**	3.23*	0.07**	8.79**	1.15**	3.20**	0.38 <sup>ns</sup>	45.01**	31.24**	158.22**
Tigray	34.56**	16.47**	51.03*	57.52*	1.31**	0.32**	2.94*	0.31**	1.55**	0.02 <sup>ns</sup>	48.64**	4.40**	100.65**
Wollega	31.11**	33.23**	21.45**	37.49**	0.53**	0.47*	7.58*	0.24*	2.30**	0.06*	55.54**	8.48**	144.27**
Wello	19.57**	26.05*	20.46*	32.63*	4.02*	0.01*	9.88*	0.84*	3.67**	0.08 <sup>ns</sup>	38.86**	15.03**	283.96 <sup>ns</sup>
<b>Altitude classes</b>													
<1500	29.58**	27.94**	30.62**	41.91*	0.28 <sup>ns</sup>	0.11*	3.04 <sup>ns</sup>	0.72*	1.43 <sup>ns</sup>	0.10 <sup>ns</sup>	34.79**	30.66**	805.15*
1501-2000	38.01**	31.38**	29.29**	18.51**	1.17**	0.05*	8.65*	0.45*	5.76**	0.08**	55.07**	25.06**	953.78**
2001-2500	19.65**	32.64**	26.15**	41.61**	1.56*	0.15*	7.17**	0.78**	5.56**	0.08**	63.24**	21.99**	918.66**
>2500	18.08**	22.57**	21.99*	46.46**	0.48*	0.11 <sup>ns</sup>	7.53*	0.83**	4.30**	0.09**	44.75**	14.81**	128.05**

\*, \*\*, ns = significant at P = 0.05; P = 0.01 level, and non-significant respectively; DH = Days to maturity (days), GFP = Grain filling period (days), NFTPP = Number of fertile tiller per plant (number), NKPS = Number of kernels per spike (number), NTPP = Number of tiller per plant (number), PH = Plant height (cm), PL = Peduncle length (cm), SL = Spike length (cm), SD = Spike density, SW = Spike weight (g), TSW = Thousand seed weight (g), GY = Grain yield per plant (g).

Table 9. Distribution of 585 barley accessions over five clusters by region of origin and altitude classes

Region	Clusters					Number of accessions
	I	II	III	IV	V	
Arsi	35	10	5	16	17	83
Bale	10	12	5	12	7	46
Gamo Gofa	3	16	5	9	3	36
Gojam	7	4	8	26	16	61
Gonder	11	3	8	17	6	45
Hadiya	5	2	1	5	8	21
Hararghe	30	20	-	3	4	57
Jimma	6	4	-	7	3	20
Shewa	33	11	7	10	14	75
Sidama	2	7	-	3	5	17
Tigray	5	2	8	29	20	64
Wellega	5	2	2	16	4	29
Wello	16	7	3	1	4	31
Total	168	100	52	154	111	585
Altitude classes (masl)						
I (<1500)	37	29	6	40	31	143
II (1501-2000)	63	25	23	59	34	204
III (2001-2500)	46	23	19	55	25	168
IV (>2500)	22	23	4	-	21	70
Total	168	100	52	154	111	585

Table 10. Summary of cluster mean of regions of origin of barley accessions for the quantitative characters

Characters	Cluster means				
	I	II	III	IV	V
Days to 50% heading	65.45	68.02	66.15	69.50	72.87
Days to 90% maturity	102.55	104.93	124.41	124.90	107.10
Grain filling period	37.10	36.91	40.94	54.40	53.53
Plant height	101.26	101.51	97.53	107.61	100.45
Number of tiller per plant	3.99	3.72	8.60	4.45	4.90
Number of fertile tillers per plant	2.92	2.65	6.41	3.28	3.57
Peduncle length	35.04	34.53	35.19	35.56	35.81
Spike length	7.12	6.95	8.07	7.91	8.34
Spike density	4.70	5.80	7.71	3.42	4.45
Spike weight	1.54	1.66	1.97	1.28	1.41
Number of kernels per spike	49.72	44.59	30.91	24.79	27.06
Thousand seed weight	30.79	30.28	35.20	40.08	39.42
Grain yield	61.23	65.71	60.83	69.14	62.16
Regions	Hararghe, Sidama, Tigray	Gonder, Wellega, Wello	Bale, Gojam, Hadiya, Jimma	Arsi, Shewa	Gemo Gofa
Number of regions	3	3	4	2	1

Cluster IV consisted of Arsi and Shewa, in which landraces with longer grain filling period, longer plant height, moderately high peduncle length and heavier seed were grouped. Cluster V was comprised of one region (Gamo Gofa) having landraces with moderate plant height and number of tillers per plant, longer peduncle and spike length.

Regarding distance between accessions, on average the highest intra-cluster distance (distance within clusters) was 23.12 for cluster III followed by cluster I. This indicated that genotypes in the same cluster (in cluster III and I) had sufficient distances with each other for recombination in future cultivar development. The lowest intra-cluster distance was 6.23 for genotypes within cluster V indicating the genotypes within this cluster are more similar (Table 11).

On the other hand, the highest inter-cluster distances (distance between clusters) were 57.37 between clusters IV and V, 49.20 between cluster II and V and 45.48 between cluster III and V. The lowest inter-cluster distances were 15.36 between cluster III and V and 21.17 between cluster I and II (Table 11).

Table 11. Average intra (**Bold**) and inter-cluster (off diagonal) distances among five clusters.

Cluster	I	II	III	IV	V
I	<b>22.32</b>				
II	21.17	<b>19.21</b>			
III	31.41	29.81	<b>23.12</b>		
IV	28.52	26.96	15.36	<b>20.61</b>	
V	33.15	49.20	45.48	57.37	<b>6.23</b>

#### *Principal component analysis for landraces*

The principal component analysis (PCA) based on thirteen quantitative characters studied revealed that the first three principal components (PCs) with Eigen values greater than one accounted for 51.75% of the total variations among landraces as shown in Table 12. The relative magnitude of Eigen values from the first PCs (22.56%) indicated that, the traits such as plant height, 1000- seed weight, grain yield per plant and peduncle length posed the greater contribution in the positive direction while days to 50% heading loaded heavily in the negative direction. Similarly, number of seeds per spike, days to 90% maturity and spike length contributed major variation in the second PCs. Third PC explained 10.25% of total variation with high loadings from number of total tiller followed by fertile tillers per plants and days to 90% maturity.

#### *Principal component analysis for regions*

The PCA for regions explained that 36.68% of total variation among regions was attributed to the first three PCs having Eigenvectors greater than one as shown in Table 12. Number of fertile tiller per plant, plant height and days to 50% heading contributed for major variation in first PC. The second PC contributed 11.82% of total variation in which peduncle length, days to maturity and grain filling period had greater loading, while traits loading heavily on third PC (8.69%) were thousand seed weight, days to heading, and number of seeds per spike.

Table 12. Eigenvectors, total variance, cumulative variance and eigenvalues for quantitative characters for accessions, regions of origin and altitude classes

Characters	Eigen vector for accessions			Eigen vector for regions			Eigen vector for altitude classes		
	PC1	PC2	PC3	PC1	PC2	PC3	PC1	PC2	PC3
Days to heading	-0.309	0.227	0.258	0.367	0.135	0.346	-0.239	0.245	-0.080
Days to maturity	-0.115	0.329	0.300	0.112	0.329	0.185	-0.136	0.340	-0.025
Grain filling period	0.034	0.199	0.141	-0.037	0.303	-0.039	0.033	0.202	0.036
Plant height	0.534	0.224	0.017	0.339	0.299	-0.190	0.214	0.163	0.168
Tiller per plant	0.114	-0.103	0.580	0.028	0.146	0.095	0.118	0.009	-0.257
Fertile tiller per plant	0.103	-0.128	0.564	0.421	0.123	0.082	0.113	-0.027	-0.226
Peduncle length	0.313	0.210	0.174	-0.037	0.343	-0.224	0.258	0.293	-0.143
Spike length	-0.086	-0.280	0.078	0.067	-0.123	0.059	-0.055	-0.325	0.003
Spike density	-0.057	-0.459	-0.138	0.025	0.017	0.051	-0.046	0.071	0.008
Spike weight	0.089	-0.405	0.061	0.026	-0.102	-0.152	0.226	-0.093	-0.021
Number of kernels per spike	-0.108	0.427	-0.128	-0.136	0.213	0.284	-0.130	0.281	0.364
Thousand seed weight	0.441	-0.103	0.095	-0.066	0.181	0.366	0.301	-0.001	-0.247
Grain yield per plant	0.391	0.014	-0.142	0.189	0.310	-0.174	0.221	0.031	0.245
Eigen value	4.286	3.598	1.947	5.496	4.018	2.956	4.989	<b>3.850</b>	<b>3.055</b>
% of total variance	22.56	18.94	10.25	16.17	11.82	8.69	14.67	<b>11.32</b>	<b>8.99</b>
% cumulative variance	22.56	41.50	51.75	16.17	27.99	36.68	14.67	25.99	34.98

#### Principal component analysis for altitudinal classes

The first three PCs with Eigenvectors greater than one were considered for altitudinal classes and accounted for 34.98% of the total variance (Table 12). For the first PC accounting 14.67% of the total variance, 1000 seed weight, peduncle length, and plant height had major contributions. Traits such as days to maturity, number of seeds per spike and spike length were the most loading in the second principal component. Similarly, number of seeds per spike, total tiller per plant and number of fertile tillers per plant had greater loading in the third principal component.

#### DISCUSSION

In any crop improvement program, genetic gains require high heritability of important agronomic traits of the crop (SINGH, 2001). Existing variations of quantitative characters are influenced by crop genetic factors, environment and their interactions. These data indicate a high level of morphological (quantitative traits) variations in Ethiopian barley landraces within each region and altitude of collection that could be exploited by breeding programmes.

In this study we found significant variation in terms of phenological, morphological and agronomic characters among the tested landraces revealed the potential opportunities to exploit the existing genetic resources through selection to improve barley crop. For instance, the genetic variability observed in days to heading, days to maturity time and grain filling period offer greater opportunity for developing varieties that are suitable for different agro-ecologies of

Ethiopia that differ in growing-period length. Previous studies on genetic variability of barley landraces collections similarly indicated significant variations in quantitative characters (ASSEFA and LABUSCHAGNE, 2004; ABAY *et al.*, 2009; HADADO *et al.*, 2009; ABEBE *et al.*, 2010; MUHE and ALEMAYEHU, 2011; HAILU *et al.*, 2016).

Analysis of diversity pattern in terms of region of origin and altitude classes for quantitative characters indicated the existence of significant phenotypic variation within regions showing the presence of differences in agro-ecological conditions across the regions contributing for the observed differences. Characteristics which are important for farmers and are used as a selection criterion showed relatively high phenotypic variance within regions of origin. Though their main focus was on qualitative characters, similar results were reported by previous researches on certain quantitative characters by ENGELS (1994), DEMISSIE and BJORNSTAD (1996), and FEKADU *et al.* (2018) similar reports were presented.

Altitudinal gradient for phenotypic diversity analysis showed significant differences for all characters indicating presence of genotypic variations, especially in an altitude classes III (2001-2500) and IV (>2500). The altitude groups II and IV comprised the major barley producing regions of the country. As mentioned earlier, though they used small quantities of landraces collected from limited areas, in terms of altitudinal variations, previous studies by ENGELS (1994), DEMISSIE and BJORNSTAD (1996) and FEKADU *et al.* (2018) on barley landraces reported similar observations.

The detected high morphological variation among landraces for regions of collection and different altitudinal classes suggested that the morphological variations in Ethiopian barley landraces strongly influenced by environmental factors resulting in variations in characters with regions and altitude classes from where accessions were collected. Phenotypic diversity among Ethiopian barley landraces was also reported by NEGASSA (1985), ASFAW (1988), CROSS (1994), ENGELS (1994) and KEBEBEW *et al.* (2001). These observed large genetic variations within regions could be due to the nature of farmers' selection forces operating in similar manner across geographic regions which attributed to less difference between regions of origin for landraces. The other reason could be high seed exchanges among the farming societies from different localities and in some cases even to a longer distances resulting in high gene flow among regions. The observed less genetic variations in some regions could be related with weather conditions which increases environmental influences and increases the occurrence of specific phenotypes adapted to the prevailing climatic and soil conditions (JARADAT *et al.*, 2004). Also, as reported in DEMISSE and BJORNSTAD, 1996), regions with high levels of different environmental stresses (e.g. frost, drought) tend to have more homogenous genotypes and less variation among genotypes.

With similar pattern as regions, partitioning of total genetic variation was conducted for altitude classes and resulted in high genetic variation within altitude classes. As altitudes within a region cover a wide range there was no population differentiation observed among altitude classes which could be associated with the ease of movement along altitude class within regions and there were possibilities to cover all altitude classes with short distance accompanied by high gene flow as compared to regions. In this study we observed that high genetic variations were obtained in an altitude class II and III, which included the major barley growing areas of the country. Similar results were reported by previous researchers who conducted experiment on

Ethiopian barley landraces (ENGELS, 1994; DEMISSE and BJORNSTAD, 1996; ABEBE *et al.*, 2010) in which they found high variations in areas between 2,000 to 3,000 masl. KEBEBEW *et al.* (2001) also reported the reduction in diversity at altitude above 2600 masl for barley landraces and ABAY *et al.* (2009) reported positive and significant association between diversity index and altitude as well as low temperature. HADADO *et al.* (2009) also mentioned reduction of areas of barley cultivation as altitude decreased confirming that barley is a cool climate crop.

The Duncan's multiple range testing for regional means over all the quantitative characteristics indicated much more regional differentiation for yield and yield components like number of seeds per spike, number of fertile tillers per plant, total tillers per plant and spike length. Such more diverse regions and altitude favoured the development of different quantitative characteristics. Landraces from Bale and Wellega zones showed shorter days to heading and mature significantly earlier than those from other regions. The means for number of tillers per plant for the landraces from Gonder, Gojam and Shewa were significantly higher than those from the other regions ( $p \leq 0.05$ ).

Landraces from Hadiya, Hararghe and Jimma zones are inferior in grain yield than those from other regions. Landraces from Gojam, Tigray, Gonder, Arsi and Bale are characterized by heavier thousand seed weight. The means number of seeds per spike were high for landraces from Bale, Wello, Arsi, Wellega, Shewa, Gonder and Gojam than those of most of the rest of the regions.

The landraces from Bale, Gojam, Wellega and Gamo Gofa zones were not significantly different from each other for days to heading, maturity time and grain yield and could be a good source of early heading and yield genes for which there is an urgent need in Ethiopia. Early maturing traits are particularly important for barley cultivation in areas prone to low moisture stress or drought (where there is a limited amount of rainfall) and a short growing season.

Cluster analysis grouped landraces with greater morphological (phenotypic) similarities, however, it did not consider in including all landraces from the same or adjacent regions. Similar phenomenon was reported by (ABEBE *et al.*, 2010; ENYEW *et al.*, 2019; MEKONNON *et al.*, 2015; ZAKOYA and BENKOVA, 2004), who described that clustering of landraces based on the morphological (agronomic) characters showed no distinct regional grouping patterns in which landraces from same or adjacent regions could appear in different cluster classes. Characterization of landraces and clustering of them on the basis of their morphological and genetic similarity helps in identification and selection of the best parents for hybridization (SOUZA and SORRELLS, 1991). Hence, grouping of landraces using multivariate analysis such as UPGMA clustering would be valuable for the breeders in such a way that the most promising landraces in the population may be selected from different clusters for crop improvement.

The results of inter-cluster analysis revealed that the genotypes had wide genetic divergence with each other indicating existence of high probability for recombination. Results of this study from cluster analysis also strengthen the availability of genotypic variability even within clusters similar to previous researches on barley landraces by DERBEW *et al.*, (2013), TAHIR (2016) and ENYEW *et al.* (2019) in which they reported the existence of wide genetic divergence among the landraces expected to manifest maximum heterosis in crossing and wide genetic variability

The PCA was performed to estimate the relative importance and contribution of each quantitative trait to the total variance and illustrate the agronomic diversity among the Ethiopian landraces. The results of PCA suggested that traits, viz. spike length, number seeds per spike, plant height and large tiller per plant, were the principal discriminatory characteristics of the Ethiopian barley landraces. The findings are in line with the local farmers' perception and criteria for selection of a cultivar in high mountain areas (ABAY *et al.*, 2009; HADADO *et al.*, 2009). Similar conclusions, in terms of patterns of variability in barley traits via PCA, were obtained by several previous researchers (DEMISSIE and BJORNSTAD, 1996; ABEBE, *et al.*, 2010; HAILU *et al.*, 2016; FEKADU *et al.*, 2018).

Breeders aim to select superior genotypes on the basis of phenotypic expression. However, for the quantitative characters, genotypes are influenced by environment, thereby affecting the phenotypic expression. Information regarding the nature and extent of association of morphological characteristics to accessions would be helpful in selecting desirable traits and improving yield, a complex characteristic for which direct selection is not effective.

The large phenotypic variation of quantitative characters observed in this study and previous research reports (ENGELS, 1994; ASSEFA and LABUSCHAGNE, 2004; ABAY *et al.*, 2009; HADADO *et al.*, 2009; ABEBE *et al.*, 2010; MUHE and ALEMAYEHU, 2011; HAILU *et al.*, 2016) on barley germplasm could be ascribed to many factors. One important factor is the fact that barley is grown in diverse agro-ecological conditions, being influenced by rainfall, altitude, growing period and edaphic factors. Other important factors such as socio-cultural, historical and economic system differences among the population who are cultivating barley likely contribute to its variation (DEMISSIE and BJORNSTAD, 1996; LAKEW *et al.*, 1997; SHEWAYRGA and SOPADE, 2011; ABRAHA *et al.*, 2013). The various physical, biological, and human factors, as well as complex interactions among such factors all seem to have contributed to the wide range of variation of the current barley landraces in the country (HADADO *et al.*, 2009; ABEBE *et al.*, 2010).

The overall diversity of Ethiopian barley landraces used in this study provided confirmation of widespread statistically significant quantitative trait (phenotypic) variation at both regional, within-regional level and altitude of collection. The diverse agro-ecologies of Ethiopia coupled with the long years cultivation of barley landraces under diverse socio-economic and cultural situation and diverse soil factors attributed to the occurrence of highly diverse phenotypes and support the conclusion that Ethiopia is an important secondary centre of diversity for barley (HARLAN, 1976; LAKEW *et al.*, 1997; ABEBE *et al.*, 2010). This diversity is rather evenly distributed over the major barley producing regions of the country. However, the study showed that there is a decreasing diversity towards lower as well as higher altitudes. This means that to capture the most genetic diversity one should concentrate on the medium altitudes between 2000 and 3000 meter above sea level, which correspond with the best growing conditions for barley in Ethiopia. Because of the presence of stronger natural selection pressure towards the extremes of the altitudinal range, there is expectation to get certain desirable landraces, for instance for abiotic stress tolerance, such as frost resistance or drought, or biotic stress like barley shoot fly tolerance, to be found at higher frequencies in these areas. As a result, the enormous diversities identified will provide barley researchers with information and new opportunities for selection, breeding and creation of improved barley genotypes.

### CONCLUSION AND IMPLICATION TO IMPROVE BARLEY IN ETHIOPIA

Ethiopia is endowed with huge barley landrace genetic resources making it as one of the major centers of diversity. These large collections of barley landraces are divergent and rich with various unique traits. However, the magnitude of these genetic divergences is not quantified yet very well. As a result, the country has not benefitted from its own genetic resources at large. Therefore, this study was undertaken to estimate the genetic distance among the studied genotypes in terms regions of origin and altitude classes. ANOVA showed highly significant differences among the tested accessions for all 13 quantitative characters indicating the presence of genetic variability in the accessions studied. The clustering patterns of the accessions based on 13 quantitative characters grouped them into 5 distinct clusters of different sizes. On average the maximum genetic distances among the studied barley landraces were 57.37 suggesting presence of diversity between these groups. Hence, maximum variation in the subsequent generations is expected from crosses that involve parents from the clusters characterized by maximum inter-class distances. According to PCA, the major quantitative traits contributing 51.75% for these observed variations among the studied accessions include plant height, grain yield per plant, 1000-seed weight, number of kernels per spike, number of seed-bearing tillers per plant, days to heading and maturity. Based on this result, the landraces could be used as contrasting parents for further breeding programs. Yield of these landraces could also be improved by either crossing these divergent landraces with each other or by selection focusing on major agronomic traits.

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### REFERENCES

- ABAY, F., A., BJORNSTAD, M., SMALE (2009): Measuring on Farm Diversity and Determinants of Barley Diversity in Tigray: Northern Ethiopia. *Momona Ethiopian Journal of Sci.*, 1: 44-66.
- ABEBE, T.D., A.M., BAUER, J., LEON (2010): Morphological diversity of Ethiopian barleys (*Hordeum vulgare* L.) in relation to geographic regions and altitudes. *Hereditas*, 147: 154-164.
- ABRAHA, A., A.K., UHLEN, ABAY F., S., SAHLSTROM, A., BJORNSTAD (2013): Genetic variation in barley enables a high quality injera, the Ethiopian staple flat bread, comparable to tef. *Crop Sci.*, 53(5): 2040-2050.
- ADAMS, M.W. (1995): An estimate of homogeneity in crop plants with special reference to genetic vulnerability in dry season, *Phaseolus vulgaris*. *Euphytica*, 26: 665-679.
- ASAW, Z. (1988): Variation in the morphology of the spike within Ethiopian barley, *Hordeum vulgare* L. (Poaceae). *Acta Agricultura Scandinavica*, 38: 277-288.

- ASSEFA, A., M.T., LABUSCHAGNE (2004): Phenotypic variation in barley (*Hordeum vulgare* L.) landraces from north Shewa in Ethiopia. *Biodiversity and Conservation*, 13: 1441-1451.
- AYANA, A., E., BEKELE (2000): Geographic patterns of morphological variation in sorghum (*Sorghum bicolor* (L.) Moench) germplasm from Ethiopia and Eritrea: Quantitative characters. *Euphytica*, 115: 91-104.
- CHAKRAVORTY, A., P.D., GHOSH, P.K., SAHU (2013): Multivariate Analysis of Phenotypic Diversity of Landraces of Rice of West Bengal. *American Journal of Experimental Agriculture*, 3(1): 110-123.
- COMADRAN, J., J., RUSSELL, F., EEUWIJK (2008): Mapping adaptation of barley to drought environments. *Euphytica*, 161: 35-45.
- CROSS, R. (1994): Geographical trends within a diverse spring barley collection as identified by agro-morphological and electrophoretic data. *TAG*, 88: 597- 603.
- DEMISSE, A., A., BJORNSTAD (1996): Phenotypic diversity of Ethiopian barleys in relation to geographic regions, altitudinal range, and agro-ecological zones: as an aid to germplasm collection and conservation strategy. *Hereditas*, 124: 17-29.
- DERBEW, S., E., URAGE, H., MOHAMMED (2013): Genetic Variability in Barley (*Hordeum vulgare* (L.) Landrace Collections from Southern Ethiopia. *Int. J. Sci. Res.*, 2: 353-366.
- ENGELS, J.M.M. (1994): Genetic diversity in Ethiopia in relation to altitude. *Gen.Res. Crop Evol.*, 41: 67-73.
- ENYEW, M., T., DEJENE, B., LAKEW, F., WOREDE (2019): Clustering and principal component analysis of Barley (*Hordeum vulgare* L.) landraces for major morphological traits from North Western Ethiopia. *Int. J. Agric. Sci. Food Tech.*, 5(1): 058-063.
- ETTEN, J., M., FUENTES, L., MOLINA (2008): Genetic diversity of maize (*Zea mays* L. ssp. mays) in communities of the western highlands of Guatemala: Geographical patterns and processes. *Gen. Res. Crop Evol.*, 55: 303-317.
- FEDERER, W.T., D., RAGAVARAO (1975): On augmented design. *Biometrics*, 31: 35-39.
- FEKADU, A., F., WOLDESENBET, S., TENAW (2018): Qualitative traits variation in barley (*Hordeum vulgare* L.) landraces from the Southern highlands of Ethiopia. *Int. J. Biodiv. Conser.*, 10: 258-264.
- GRAHIC, J., F., GASI, M., KURTOVI, L., KARI, M., IKI, D., GADZO (2013): Morphological evaluation of common bean diversity in Bosnia and Herzegovina using the Discriminant analysis of Principal Components multivariate method. *Genetica*, 45: 963-977.
- HADADO, T.T., D., RAU, E., BITOCCHI, R., PAPA (2009): Genetic Diversity of barley (*Hordeum vulgare* L.) landraces from the central highlands of Ethiopia: comparison between the Belg and Meher growing seasons using morphological traits. *Gen. Res. Crop Evol.*, 56: 1131-1148.
- HAILU, A., S., ALAMEREW, M., NIGUSSIE, E., ASSEFA (2016): Genetic variability, heritability and genetic advance for yield and yield related traits in barley (*Hordeum vulgare* L.) germplasm. *World J. Agric. Sci.*, 12: 36-44.
- HARLAN, J.R. (1969): Ethiopia: a center of diversity. *Economic Botany*, 23:309-314.
- HARLAN, J.R. (1976): Barley. In: Simmonds, N.W. (Eds.) *Evolution of crop plants*, Longman Press, UK, pp. 93-98.
- HAUSSMANN, B.I.G., H.K., PARZIES, T., PRESTERL, Z., SUSIC, T., MIEDANER (2004): Plant genetic resources in crop improvement. *Plant Genetic Resources*, 2: 3-21.
- IPGRI (International Plant Genetic Resource Institute) (1994): Description of barley (*Hordeum vulgare* L.). International Plant Genetic Resources Institute, Rome, Italy.
- JARADAT, A., M., SHAHID, A., AL MASKRI (2004): Genetic diversity in the Batini barley landrace from Oman: I. spike and seed quantitative and qualitative traits. *Crop Sci.*, 44: 304 315.
- KEBEBEW, F., Y., TSEHAY, T., MCNEILLY (2001): Morphological and farmers cognitive diversity of barley (*Hordeum vulgare* L. (Poaceae)) at Bale and North Shewa of Ethiopia. *Gen. Res. Crop Evol.*, 48: 467-481.

- LAKEW, B., T., SEMEANE, F., ALEMAYEHU, H., GEBRE, S., GRANDO, J.A.G., VAN LEUR, S., CECCARELLI (1997): Exploiting the diversity of barley landraces in Ethiopia. *Gen. Res. Crop Evol.*, *44*:109-116.
- MARUTHI, S.G., M.D., NARASIMHA, M., VANAJA, R.P., RAGHURAM (1999): A multiple selection index for selecting sunflower genotypes using principal component analysis. *Indian Journal of Dryland Agricultural Research Development*, *14*: 93–103.
- MEKONNON, B., B., LAKEW, T DESSALEGN (2015): Morphological diversity and association of traits in Ethiopian food barley (*Hordeum vulgare* L.) landraces in relation to regions of origin and altitude. *Journal of Plant Breeding and Crop Science*, *7*: 44-54.
- MOHAMMADI, S.A., B.M., PRASANNA (2003): Analysis of genetic diversity in crop plants-salient statistical tools and considerations. *Crop Sci.*, *43*: 1235–1248.
- MUHE, K., A., ASSEFA (2011): Diversity and agronomic potential of barley (*Hordeum vulgare* L.) landraces under variable production system in Ethiopia. *International Research Journal of Plant Science*, *2*: 305-310.
- NEGASSA, M. (1985): Patterns of phenotypic diversity in an Ethiopian barley collection and the Arussi-Bale Highland as a center of origin of barley. *Hereditas*, *102*: 139-150.
- ORABI, J., G., BACKES, A., WOLDAY, A., YAHYAOU, A., JAHOOOR (2007): The Horn of Africa as a centre of barley diversification and a potential domestication site. *TAG*, *114*: 1117-1127.
- PEETERS, J.P., J.A., MARTINELLI (1989): Hierarchical clustering analysis as a tool to manage variation in germplasm collection. *TAG*, *78*: 42-48.
- SHEWAYRGA, H., P.A., SOPADE (2011): Ethnobotany, diverse food uses, claimed health benefits and implications on conservation of barley landraces in North Eastern Ethiopia highlands. *J. Ethnobiol. Ethnomed.*, *7*(19):1-15.
- SINGH, B.D. (2001): *Plant breeding: principles and methods*. Kalyani Publishers, New Delhi, India.
- TAHIR, Z., A., ASSEFA, A. SENTAYEHU, W., GEBRESELASSIE (2016): Phenotypic diversity in Ethiopian food barley (*Hordeum vulgare* L.) genotypes. *World Journal of Agricultural Sciences*, *12*: 308-314.
- VAVILOV, N.I. (1951): The origin, variation, immunity and breeding of cultivated plants. *Soil Sci.*, *72*: 482.
- ZAKOYA, M., M., BENKOVA (2004): Genetic diversity of genetic resources of winter barley maintained in the gene bank in Slovakia. *Czech J. Genet. Plant Breeding*, *40*: 118-126.

**MULTIVARIACIONA ANALIZA VARIJABILNOSTI KVANTITATIVNIH OSOBINA  
KOD POPULACIJA ETIOPSKOG JEČMA (*Hordeum vulgare* L.): NA OSNOVU  
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## Izvod

Ječam (*Hordeum vulgare* L.) je najznačajnija žitarica viših predela u Etiopiji i široko je rasprostranjena u većem dijelu zemlje. Ukupno 585 populacija ječma i 10 standarda ocenjeno je korišćenjem proširenog randomiziranog kompletnog blok dizajna koji se sastojao od šest blokova. Svih 585 populacija posađeno je na parcelama bez ponavljanja, a 10 standarda je ponovljeno šest puta (u svakom bloku po jedan) kako bi se procenila varijansa greške. Podaci o 13 kvantitativnih osobina korišćeno je za izračunavanje deskriptivne statistike, ANOVA-e i multivarijacijske analize (UPGMA i analiza glavnih komponenti). Bilo je značajnih razlika (ANOVA,  $P < 0,01$ ) među biljkama za visinu biljke, težinu 1000 zrna, broj zrna po klasu, broj dana do klasanja i broj dana do zrenja. Svi genotipovi su grupisani u pet klastera gde 74,02% uzoraka (433) spada u klastere I, IV i V. Ranostasni uzorci grupisani su u klaster I, dok su kasnostasni, visoko prinostni i visoki uzorci grupisani su u klaster IV. Najviša udaljenost unutar klastera bila je 23,12 za klaster III, dok je najveća udaljenost između klastera bila 57,37 između klastera IV i V. Prve tri glavne komponente doprinele su 51,76% ukupnim varijacijama među genotipima. Samo glavna komponenta (PC1) je doprinela 22,56% ukupnim varijacijama, uglavnom zbog visine biljke, mase 1000 zrna, prinosa zrna i dužine stabljike. Glavna komponenta PC2 doprinela je sa 18,94% ukupnim varijacijama uglavnom kroz gustinu klasa, broj zrna po klasu, težinu klasa i broj dana do pune zrelosti. Glavna komponenta PC3 doprinela je sa 10,94% ukupnim varijacijama ukupnim brojem stabljika po bokoru, brojem stabljika sa ozrnenim klasovima, brojem dana do 90% zrelosti i brojem dana do 50% klasanja. Nadmorska visina izvornih mesta sakupljanja populacija takođe je značajno uticala na različite proučavane kvantitativne osobine. Regionalne razlike vidljive su i među kolekcijama populacija. Ovi rezultati otkrivaju postojanje značajnih agro-morfoloških varijacija među populacijama uključenim u ovo istraživanje. Na osnovu proučavanih osobina ocenjenih populacija, značajan diverzitet koji je primećen među populacijama ječma u Etiopiji mogla bi se iskoristiti u budućem poboljšanju useva za razne agronomski značajne osobine. Dobijene informacije dopunjuju robusni program oplemenjivanja ječma u cilju stvaranja konkurentnih, stabilnih i klimatski otpornih sorti koje preferiraju krajnji korisnici u različitim agroekološkim uslovima Etiopije.

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