Application of Multivariate Analysis on Morphometric Traits to Characterize the Sheep Populations Reared in the Central Rift Valley of Ethiopia

Ashebir Worku[1] and Aberra Melesse[2#]

[1]Oromia Agricultural Research Institute, AdamiTullu Agricultural Research Center, P.O. Box 35 Batu;
[2]Hawassa University, School of Animal and Range Sciences, P.O. Box 1798, Hawassa
[#]Corresponding author: a_melesse@uni-hohenheim.de

ABSTRACT

The study was conducted in Adam-Tullu Jiddo-Kombolcha (ATJK), Bora, Asasa and Kofele districts situated at the Central Rift Valley of Oromia, to describe the morphological diversity of sheep populations by applying multivariate analysis. Morphometric traits were taken from 523 ewes of various age groups. The traits scored were live weight (LW), body length (BL), height at withers (WH), heart girth (HG), height at rump (RH), chest depth (CD), chest width (CW), rump length (RL), rump width (RW), head length (HL), head width (HW), ear length (EL), canon circumference (CC), canon length (CL), tail length (TL), tail circumference (TC) and neck length (NL). Results revealed that district had a highly significant effect on most traits except RL, RW, and HL. The age effect was also significant for all traits except CC, CL and TC. The sheep of Kofele had the highest LW, BL and TL values while those of the Asasa showed the lowest RH, and WH being the shortest amongst the studied sheep populations. The highest WH was noted in Kofele sheep being the tallest ($p<0.01$) from Asasa and Bora sheep. The longest EL was observed in Asasa and Kofele sheep being higher ($p<0.01$) than those of ATJK and Bora. Most morphometric traits increased with advancing age. Body length, TC, CW, TL and LW were identified as the most discriminating variables to differentiate the four district sheep populations. All Mahalanobis distances were significant ($p<0.01$) being the shortest between sheep of ATJK and Bora (0.76) while the longest was observed between those of Asasa and Bora (3.60). Three canonical variables (CAN) were determined of which CAN1 and CAN2 accounted for 69.0 and 27.0% of the total variations, respectively. The CAN1 loaded highly for RH while CAN2 weighted for CD. About 72.3%, 68.6%, and 64.7% of original sheep were correctly classified into their respective source population of Asasa, Kofele and Bora districts. However, only 36.4% of the original ATJK sheep were assigned into their respective population while the remaining being misclassified to other sheep populations. The present work revealed that characterization of indigenous livestock based on morphological traits using a multivariate analysis is a viable option in regions where molecular tools are inaccessible. We recommend initiation of a community-based breeding program for sustainable utilization and conservation of Kofele sheep.

Keywords: Central rift valley; districts; indigenous sheep; morphometric traits; multivariate analysis

INTRODUCTION

Sheep rearing is one of the most important means of livelihood and food security for majority of the rural population, especially in developing countries. Given its proximity to the Arabian Peninsula, Ethiopia is considered as a genetic corridor for the introduction of livestock species including sheep to the African continent (Muigai and Hanotte, 2013). There are 14 traditional sheep populations and nine identified sheep breeds (Gizaw *et al.* 2007) and with a population of 39.89 million heads of sheep out of which about 70.3 and 29.7% are females and males, respectively (CSA, 2019/20).

The 14 Ethiopian sheep populations are broadly categorized according to their tail phenotypes as thin tailed (one breed), fat-tailed (11 populations), and fat-rumped (two populations) (Gizaw*et al.*, 2007). The short fat-tailed population mainly inhabits the sub-alpine regions; long fat-tailed sheep are predominant in mid- to high-altitude environments; and fat-rumped sheep are distributed in dry lowland areas (Gizaw *et al.*, 2007).Study conducted by Edea *et al.* (2018) using a high-density genome-wide SNP analyses on five local sheep revealed that Ethiopian sheep populations are roughly clustered according to their geographic distribution and tail phenotype. Accordingly, short fat-tailed,long fat-tailed and fat-rumped sheep are distributed in very cool high altitude, mid to high-altitude, and arid low-altitude, respectively. Recent study based on genome-wide scans of Bonga sheep revealed the existence of several candidate regions spanning genes that were not reported before in prolific sheep (Tera *et al.*, 2019).

Indigenous livestock biodiversity serve as apoolfor genes that confer disease resistance, specific product qualities likefatty acid composition or milk composition, resistance todraught and high temperatures. Indigenous sheep in Ethiopia play multifarious roles including sources of income, meat, skin, and coarse fleece. They provide an economic buffer in the event of crop failures especially under marginal productivity under low and erratic rainfall, severe land erosion, frost, and water logging regions. In some areas such as the cool alpine and arid lowlands where crop production is not a viable economic option, sheep production is a sole choice for sustainable livelihood of the rural community. Improvement of sheep productivity through proper husbandry and breeding is needed to meet the increasing demand of dietary protein from animal products.It is thus necessary to first explore the genetic potential of the existing sheep genetic resources at national, regional and/or district level through well designed characterization studies (FAO, 2012).

The first step of the characterization of local genetic resources is based on the knowledge of variation in the morphological traits (FAO, 2012). Linear body measurement traits have been suggested as objective measures of body conformation in sheep, which mainly influences market value of meat sheep in traditional markets (Gizaw *et al.*, 2008). In this regard, characterization studies based on morphological traits has contributed to the designing of viable breeding and conservations programs for sustainable utilization of the indigenous livestock genetic resources. Multivariate analyses of morphological traits have been reportedas suitabletools in assessing the genetic variations within and between populations and can effectively discriminate different population ecotypes when all measured morphological variables are considered simultaneously (Härdle and Simar, 2015). These kinds of studies are commonly reported in small ruminant animals such as goats and sheep in many countries of the world (Yakubu and Ibrahim, 2011; Dudusola *et al.*, 2019).

In Ethiopia, characterization of animal genetic resources has largely been limited to description of production systems and phenotype classification of traditional breeds using simple statistical tools such as correlation and regression analysis (Getachew *et al.*, 2009; Melesse *et al.*, 2013; Taye *et al.*, 2016; Tesfay *et al.*, 2017). In a recent study, Wagari *et al.* (2020) reported the existence of new sheep ecotype in western part of Ethiopia by applying multivariate statistical tools. However, to the authors' knowledge, no recent information is available in the literature on the morphological characterization of the indigenous sheep populations of the study area by applying a multivariate analysis. Moreover, morphological distances between sheep populations in the study areas have not been yet established which could serve as a possible basis for genetic improvement and conservation programs. The current study was therefore conducted to describe the indigenous sheep populations of four districts reared in the central rift valley of Oromia based on their morphometric traits by applying multivariate statistical tools.

MATERIALS AND METHODS

Site selection and sampling procedures

The study was conducted in four districts drawn from East Shewa and West Arsi zones of Oromia, Ethiopia. First, the relevant second hand information was gathered from Agriculture and Rural Development office of livestock. Based on the collected information, multi-stage purposive sampling techniques were applied to select the representative districts, kebeles and households. In the first stage, four districts namely Adam-Tullu Jiddo-Kombolcha (ATJK) and Bora from East Shewa zone, and Asasa, and Kofele from West Arsi zone were identified and selected purposively based on their potential for sheepproduction. In the second stage, based on distribution of sheep population, 3 kebeles from each district (total of 12 kebeles) were selected purposively. In the final stage, the households within kebeles who possess at least three matured sheep of both sexes and had long enough experiences in rearing sheep, were randomlyselected based on the proportional to the population size of the selected kebeles. Collectively, 523 ewes were sampled from the four districts. The agro-climatic characteristics of the studied districts and number of sampled ewes are provided in Table 1. The owner's recall method along with dentition classes (pairs of permanent incisors, PPI) were used to estimate the ages of sheep. Thus, sheep with 1PPI, 2PPI, 3PPI and 4PPI were classified in the age groups of yearling, 2-year-old, 3-year-old and 4-year-old, respectively (Ebert and Solaiman, 2010).

Table 1. Agro-climatic characteristics and sample size of households and sheep across each district

Districts	GPS coordinates	Altitude (m.a.s.l)	Agro-ecology coverage	Annual Rainfall(mm)	Sampled ewes
ATJK	07^o 55'N, 39^o45'E	1643	90% LL, 10% ML	700	89
Bora	08°39′ N, 39°50′ E	1880	85% LL, 15% ML	800	85
Kofele	07^o 00' N, 38^o45'E	2695	90% HL, 10% ML	2500	178
Asasa	07°06′ N, 39°12′ E	2367	65% ML, 35% HL	1970	171

ATJK = Adam-Tullu Jiddo-Kombolcha; LL = lowland <1500 m a.s.l;
ML = midland =1500-2300 m a.s.l; HL = highland >2300 m a.s.l

Data collection procedures

Data were scored on 17 morphometric traits following the descriptor list of FAO (2012) for phenotypic characterizations of sheep. Accordingly, the following traits were measured: live weight (LW), body length (BL), height at withers (WH), heart girth (HG), height at rump (RH), chest depth (CD), chest width (CW), ramp length (RL), rump width (RW), head length (HL), head width (HW), ear length (EL), canon circumference (CC), canon length (CL), tail length (TL), tail circumference (TC) and neck length (NL). Wooden made ruler fitted with sliding height bars were used to measure withers height. The LW was taken using a suspended weighing scale with 100 kg capacity by placing each animal in self-devised holding harness. All other linear measurements were taken in the morning before sheep were released for grazing by using measuring tapes made of textile material. Measurements were also restricted to healthy and non-pregnant sheep.

Data analysis

After double-checking for any types of errors or outliers, data were subjected to GLM procedures of Statistical Analysis System (SAS 2012, ver. 9.4)to analyze quantitative variables to determine effects of class variables (district and age) and their interactions. When F-test declared significant, multiple least square means were then compared with Tukey-Kramer test. The degree of morphological similarity or divergence among the indigenous sheep populations was determined using the multivariate analysis.

Dendrogram was constructed based on distances between the sheep populations of the four districts using the average linkage method of the hierarchical cluster analysis to group the flocks into their morphological similarity. Moreover, the stepwise discriminant analysis was applied using the STEPDISC procedure to determine the morphometric traits that have the most discriminating power to separate the studied sheep populations. The relative potential of the morphometric variables in discriminating the foursheep populations was assessed using the level of significance, F-statistic and partial R^2.

The canonical discriminant analysis was then performed on the identified traits with the highest discriminating power using the CANDISC procedure to compute the squared Mahalanobis distances between class means, univariate (ANOVA) and multivariate (MANOVA) analysis, canonical variables with eigenvalues, standardized canonical coefficients and canonical structures. The TEMPLATE and SGRENDER procedures were used to create a plot of the first two canonical variables in a scatter graph for visual interpretation. The discriminant analysis of the DISCRIM procedure was also conducted to determine the classification of sheep into their source populations using the quadratic discriminant function for unequal covariance matrices within classes after conducting the Bartlett's homogeneity test. The cross-validationoption was finally applied to evaluate the accuracy of the classification with a minimum bias. All multivariate analyses were performed using the Statistical Software of SAS (2012, ver. 9.4).

RESULTS

Effect of district and age on morphometric traits

Leastsquare means of morphometric traits, as well as the significance of the district and age effects, are presented in Table 2. The effect of district was highly significant for most traits studied except RL, RW, and HL. Accordingly, the Kofele sheep had higher LW, BL and TL values than the other populations. The Kofele sheep were further characterized withhigher HG than ATJK and Asasa sheep populations. The Asasa sheep had the lowest BL, CC, and TCas compared with other sheep populations. They had also the lowest RH, and WH values compared with Bora and Kofele sheep being the shortest amongst the studied sheep populations. The highest HW was noted in sheep of Kofele, which differed ($p<0.01$) from those of Asasa and Bora. The longest EL was observed in Asasa and Kofele sheep being higher ($p<0.01$) than those of ATJK and Bora. The effect of age was also significant for all traits except CC, CL and TC. Accordingly, most of the morphometric traits increased with the advancing age of the sheep. The interaction effect was significant for HG, WH, BL, CD, RL, HL, EL, and CL.

Table 2. Least square means of the morphometric traits as affected by district and age (N = 523)

Traits	Districts (D)				Age (A)				Sources of variation		
	ATJK	Asasa	Bora	Kofele	1PPI	2PPI	3PPI	4PPI	D	A	D*A
Live weight	26.0^{b}	25.9^{b}	25.9^{b}	28.1^{a}	23.6^{d}	25.2^{c}	27.8^{b}	29.0^{a}	<0.001	<0.001	0.689
Heart girth	70.0^{b}	68.6^{bc}	70.2^{ab}	71.5^{a}	67.0^{d}	68.9^{c}	71.6^{b}	72.8^{a}	<0.001	<0.001	0.006
Height at withers	61.9^{ab}	61.2^{b}	62.4^{a}	62.7^{a}	61.1^{c}	61.9^{bc}	62.3^{ab}	62.9^{a}	<0.001	<0.001	0.009
Body length	62.0^{b}	59.4^{c}	62.2^{b}	63.9^{a}	60.3^{c}	61.2^{bc}	62.6^{ab}	63.5^{a}	<0.001	<0.001	0.018
Height at rump	63.5^{ab}	62.5^{b}	63.9^{a}	64.3^{a}	62.9^{b}	63.2^{b}	63.9^{ab}	64.3^{a}	<0.001	0.006	0.101
Chest depth	30.4^{ab}	29.3^{c}	30.2^{bc}	31.1^{a}	29.1^{c}	30.1^{bc}	30.6^{b}	31.3^{a}	<0.001	<0.001	0.004
Chest width	17.6^{ab}	17.9^{a}	17.2^{b}	17.9^{a}	17.0^{b}	17.9^{a}	17.7^{a}	18.0^{a}	0.007	0.001	0.295
Rump length	19.6	19.5	19.4	19.7	19.1	19.6	19.6	19.8	0.082	0.097	0.024
Rump width	17.0	16.6	17.2	16.9	16.4^{b}	17.1^{ab}	17.0^{ab}	17.1^{a}	0.108	0.007	0.620
Head length	16.5	16.4	16.7	16.4	16.2^{b}	16.4^{ab}	16.6^{ab}	16.8^{a}	0.884	<0.001	0.002
Head width	10.9^{ab}	10.6^{b}	10.6^{b}	11.1^{a}	10.4^{b}	10.8^{ab}	11.0^{a}	11.1^{a}	0.004	<0.001	0.194
Ear length	11.2^{b}	11.7^{a}	11.2^{b}	11.9^{a}	11.0^{b}	11.4^{ab}	11.7^{a}	11.8^{a}	0.007	<0.001	0.059
Canon circumference	6.99^{a}	6.56^{b}	6.93^{a}	6.79^{a}	6.83	6.93	6.72	6.79	0.002	0.902	0.007
Canon length	12.3^{a}	11.9^{b}	12.4^{a}	12.1^{ab}	12.1	12.2	12.2	12.2	0.004	0.732	0.445
Tail length	32.6^{b}	32.7^{b}	31.7^{b}	34.8^{a}	29.6^{c}	34.9^{a}	33.0^{b}	34.2^{ab}	<0.001	<0.001	0.003
Tail circumference	16.8^{a}	14.4^{b}	17.0^{a}	16.6^{a}	16.0	16.1	16.0	16.6	<0.001	0.233	0.098
Neck length	22.6^{ab}	22.4^{b}	22.6^{ab}	23.2^{a}	22.2^{b}	22.3^{b}	23.0^{ab}	23.3^{a}	0.006	<0.001	0.633

$^{a-d}$Means with different superscript letters between columns across districts and age are significant; ATJK = Adam-Tullu Jiddo-Kombolcha

Results of multivariate analysis

The dendrogram (Figure 1) showed two clusters: cluster one identified Asasa sheep as an independent group and cluster two includes all other sheep with two sub-clusters in which Bora and ATJK grouped in one while those of Kofele in the other.

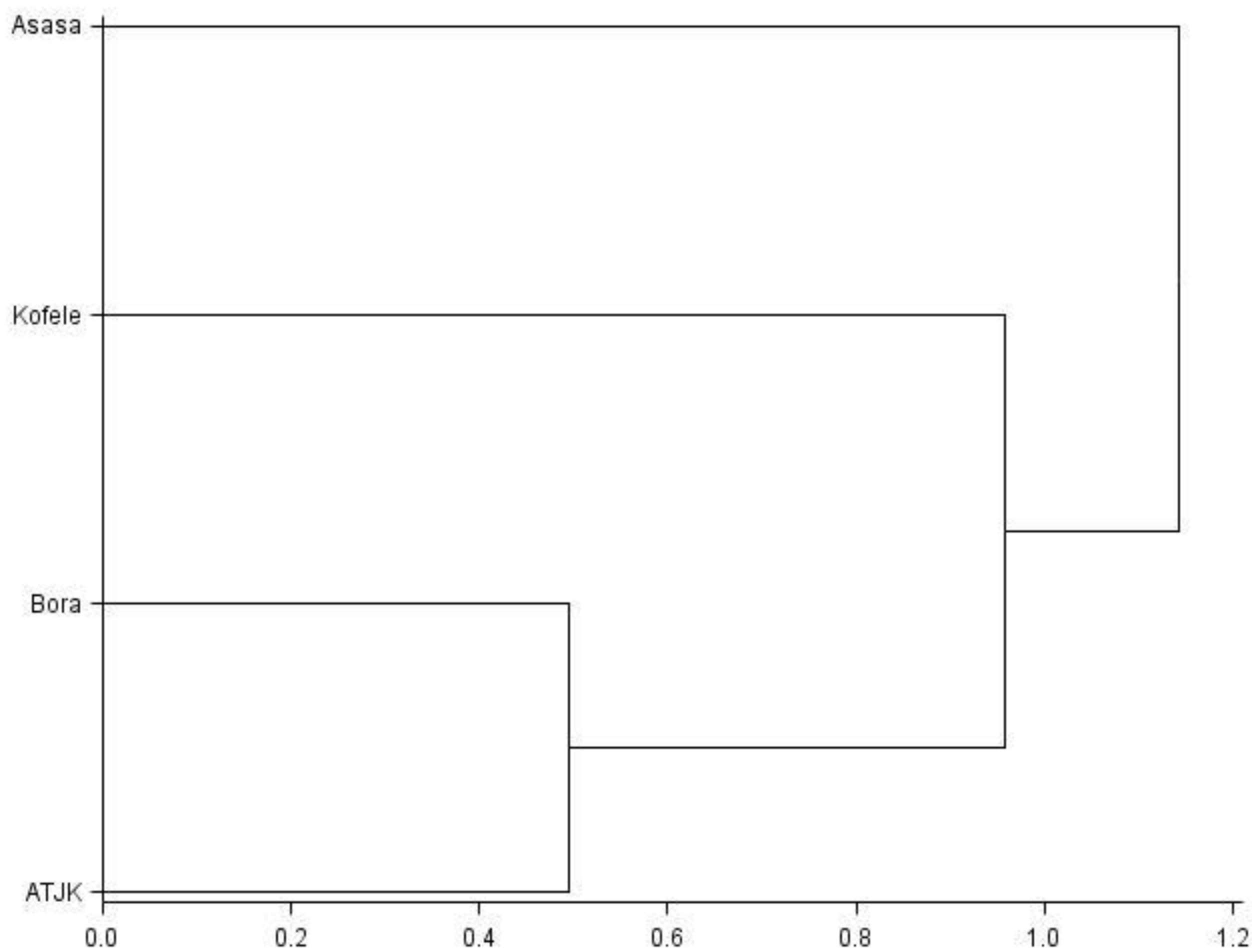

Figure 1. Dendrogram based on average linkage distances among the indigenous sheep populations of the four districts by using 15 morphometric traits (ATJK = Adam-TulluJiddo-Kombolcha district)

Seventeen quantitative variables were subjected to the STEPDISC procedure and fifteen were identified as the best discriminating variables in stepwise selection summary (Table 3). Wilk's lambda test confirmed that all the selected variables had significant ($p<0.001$) contribution to discriminate the total population into separate groups. Among them, BL, TC, CW, TL and LW were identified as the most discriminating variables to differentiate the four sheep populations.

Table 3.Selection summary of morphomteric traits using the stepwise discriminant analysis

Variables entered	Partial R^2	F Value	Pr > F	Wilks' Lambda	Pr < Lambda	ASCC	Pr > ASCC
Body length	0.149	29.60	<.0001	0.850	<.0001	0.050	<.0001
Tail circumference	0.072	13.15	<.0001	0.789	<.0001	0.071	<.0001
Chest width	0.057	10.24	<.0001	0.744	<.0001	0.088	<.0001
Tai length	0.037	6.52	0.0002	0.716	<.0001	0.100	<.0001
Live weight	0.034	5.88	0.0006	0.692	<.0001	0.110	<.0001
Chest depth	0.048	8.35	<.0001	0.659	<.0001	0.122	<.0001
Rump width	0.034	5.86	0.0006	0.637	<.0001	0.132	<.0001
Head length	0.027	4.65	0.0032	0.620	<.0001	0.139	<.0001
Height at rump	0.033	5.69	0.0008	0.599	<.0001	0.147	<.0001
Height at withers	0.025	4.32	0.0051	0.584	<.0001	0.154	<.0001
Ear length	0.023	3.85	0.0096	0.571	<.0001	0.160	<.0001
Canon length	0.023	3.84	0.0098	0.558	<.0001	0.167	<.0001
Head width	0.021	3.57	0.0141	0.546	<.0001	0.173	<.0001
Canon circumference	0.015	2.46	0.0623	0.537	<.0001	0.176	<.0001
Heart girth	0.015	2.52	0.0569	0.530	<.0001	0.180	<.0001

ASCC = average squared canonical correlation

All the fifteen variables were subjected to canonical discriminant analysis using the CANDISC procedure. The univariate statistics confirmed that each quantitative variable in sampled populations is a significant ($p<0.01$) contributor to the total variation (data not shown). The hypotheses that assumes districts' means are equal in the populations were tested using multivariate statistics and was rejected by Wilk's Lambda (F-value = 7.77, $p<0.001$).

Table 4 shows significant Mahalanobis distances between flocks based on morphometric measurements sorted by mean distances. All Mahalanobis distances were significant ($p<0.001$) being the shortest between ATJK and Bora sheep (0.76) followed by that of ATJK and Kofele (0.79). On the other hand, the longest distances was noted between Asasa and Bora (3.60) followed by that of Asasa to Kofele (2.61). The third longest distance was observed between Bora and Kofele sheep (1.76).

Table 4.Mahalanobis distances and significant levels among the sheep populations of the four districts based on morphometric traits

Districts	ATJK	Asasa	Bora	Kofele
ATJK	0	1.88***	0.76**	0.79***
Asasa		0	3.60***	2.61***
Bora			0	1.76***
Kofele				0

*** = $p<0.0001$; ** = $p<0.001$; ATJK = Adam-TulluJiddo-Kombolcha

The canonical discriminant analysis further derived a linear combination of the variables that has the highest possible multiple correlation with the groups (Table 5). The variable that is defined by the linear combination is the first canonical variable (CAN1). The process of extracting CAN2, CAN3, etc.that is needed for the separation purposes will be repeated until the number of canonical variables equals the number of classes/groups minus one. In the present study, there were four districts (ATJK, Asasa, Bora and Kofele), thus there would be 4 − 1 = 3 possible CANs(CAN1, CAN2 and CAN3) needed for separation purposes. This is evident in Table 5 where CAN1 and CAN2 explained 69.0% and 27.0% of the total variation, respectively being highly significant (p<0.0001). The third CAN (CAN3) accounted for only 4% of the total variation and its contribution to the total variation was insignificant (p = 0.3919). The correlation between CAN1 and the sheep populations sampled from the four districts was relatively high (0.568), with the canonical variables being highly significant (p<0.0001).Moreover, the respective eigenvalues foe CAN1 and CAN2 were 0.524 and 0.205, which together accounted for 0.729 (96.3%) of the cumulative variance.Table 5 further displayedthe likelihood ratio test rejecting the hypothesis that the current canonical correlation and all smaller ones are zero, except CAN3.

Table 5. Summary of canonical correlations, eigenvalues and likelihood ratios

Functions	Canonical correlations	Eigenvalues			Likelihood ratios	Approximate F-value	Pr>F
		Eigenvalue	Proportion	Cumulative			
CAN1	0.568	0.524	0.69	0.69	0.530	7.77	<0.0001
CAN2	0.387	0.205	0.27	0.96	0.807	3.98	<0.0001
CAN3	0.098	0.028	0.04	1.00	0.973	1.06	0.3919

CAN1 = canonical variable 1; CAN2 = canonical variable 2; CAN3 = canonical variable 3

Figure 2 shows a plot built with the first two canonical variables illustrating the relationships between sheep belonging to the four districts. The plot clearly showed that CAN2 discriminates between the Asasa sheep in one group while the CAN1 best discriminates among the ATJK and Bora sheep populationsin another group. The Kofele sheep fall between the two canonical variables (CAN1 and CAN2). Nevertheless, a noticeable overlapping has been observed among the three sheep populations suggesting morphological similarity among them.

As indicated in Table 6, the first canonical variable CAN1 loaded highly for RH, TC, and BL with canonical discriminant function scores of 0.765, 0.661 and 0.565, respectively. On the other hand, the CAN2 loaded for CD, TL, HW, CW, LW and ELwith canonical discriminant function scores of 0.530, 0.479, 0.462, 0.306, 0.287 and 0.255, respectively. Results of canonical structures were also in line with that of total standardized canonical coefficients in which BL, TC and RH dominated CAN1, while TL, LW, CD, HW, EL and CW showed the largest influence on CAN2 with comparable values of canonical structures.

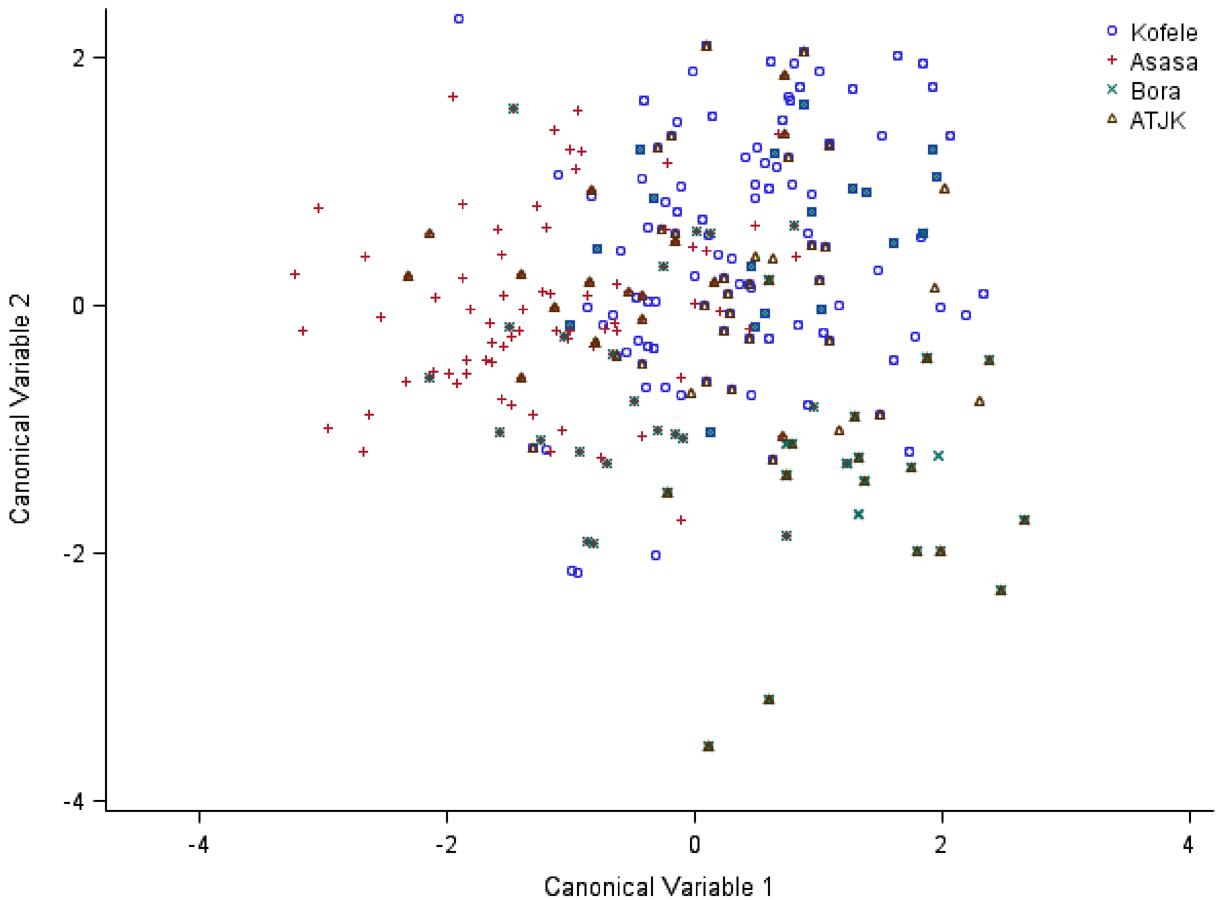

Figure 2. Canonical representation of the central rift valley sheep populations across the four districts based on morphometric traits

Discriminant analysis assumes that the individual group covariance matrices are equal (homogeneity within covariance matrices) and by default it uses the linear discriminant function for classification unless specified otherwise. In the current discriminant analysis, equality of covariance matrices within groups was tested using Bartlett's test of homogeneity for all traits and was significant ($\chi^2 = 926$; $p<0.0001$). Thus, the null hypothesis that assumes all four covariance matrices within the sheep populations are equal was rejected (Table 7). Based on these results, the appropriate discriminant function to be applied for the classification of the four sheep populations would be the quadratic discriminant function.

Table 6.Standardized canonical coefficients and canonical structures based on morphometric variables of indigenous sheep populations sampled from four districts

Variables	Standardized canonical coefficients		Canonical structures	
	CAN1	CAN2	CAN1	CAN2
Live weight	-0.589	0.287	0.208	0.465
Heart girth	0.256	-0.470	0.331	0.285
Height at withers	-0.531	0.226	0.315	0.233
Height at rump	0.765	-0.292	0.410	0.216
Body length	0.565	0.405	0.595	0.370
Chest depth	0.465	0.530	0.415	0.412
Chest width	-0.574	0.306	-0.098	0.365
Rump width	0.103	-0.483	0.231	-0.131
Head length	-0.365	-0.194	0.012	0.001
Head width	0.006	0.462	0.155	0.403
Ear length	-0.246	0.255	-0.118	0.396
Canon circumference	0.258	-0.234	0.323	-0.020
Canon length	-0.005	-0.417	0.277	-0.060
Tail length	0.177	0.479	0.077	0.509
Tail circumference	0.661	-0.102	0.593	-0.0013

Most important variables (WEIGHT) within the CAN1and CAN 2 are indicated with bold CAN1 = canonical variable 1; CAN2 = canonical variable 2;

Table 7. Test for the homogeneity of covariance matrices

Districts	Covariance matrix rank	Log of covariance determinant	Overall homogeneity test results
ATJK	15	11.6	$\chi^2 = 926$
Asasa	15	11.2	DF = 360
Bora	15	9.42	P-value = <0.0001
Kofele	15	12.3	
Pooled within districts	15	13.3	

ATJK = Adam-Tullu Jiddo-Kombolcha

As presented in Table 8, 72.3% of Asasa sheep were correctly classified into their respective group with 12.7, 8.43 and 6.63% being misclassified to ATJK, Bora and Kofele sheep populations, respectively. Likewise, 68.6% of Kofele sheep were correctly assigned to their source population while the remaining 11.1, 14.0 and 6.40 being misclassified to ATJK, Asasa and Bora sheep populations, respectively. The quadratic discriminant function was also able to differentiate the Bora sheep from others with 64.7% correct classification into their original source population with the remaining 3.53, 15.3 and 16.5% being misclassified to ATJK, Asasa and Kofele sheep populations, respectively. On the other hand, only 36.4% of the original ATJK sheep were correctly classified into their respective population while the majority (about 63.6%) of the remaining being misclassified to the other sheep populations. Overall, 60.5% the sheep populations were correctly assigned to their source population using the quadratic discriminant function with 15 variables.

Table 8. Percent of individual sheep classified into source populations and cross-validation of the classification using morphometric variables (values in brackets are number of sheep)

Districts	ATJK	Asasa	Bora	Kofele	Total
Classified					
ATJK	36.4 (32)	15.9 (14)	27.3 (24)	20.5 (18)	88
Asasa	12.7 (21)	72.3 (120)	8.43 (14)	6.63 (11)	166
Bora	3.53 (3)	15.3 (13)	64.7 (55)	16.5 (14)	85
Kofele	11.1 (19)	14.0 (24)	6.40 (11)	68.6 (118)	172
Error count rate	0.636	0.277	0.353	0.311	0.395
Priors	0.250	0.250	0.250	0.250	
Cross-validated					
ATJK	22.7 (20)	19.3 (17)	29.6 (26)	28.4 (25)	88
Asasa	18.7 (31)	60.2 (100)	11.5 (19)	9.64 (16)	166
Bora	14.1 (12)	21.2 (18)	45.9 (39)	18.8 (16)	85
Kofele	18.6 (32)	14.5 (25)	9.30 (16)	57.6 (99)	172
Error count rate	0.773	0.398	0.541	0.424	0.534

ATJK = Adam-Tullu Jiddo-Kombolcha

The classification accuracy of the discriminant analysis was further cross-validated and indicated an overall 46.6% success rate. The error-count estimates gave the proportion of misclassified observations in each group being highest in ATJK sheep and lowest in Asasa. Accordingly, 19.3, 29.6 and 28.4% of ATJK sheep were misclassified to Asasa, Bora and Kofele, respectively (Table 8). The overall rate of error count estimate for the classification was 39.5% while it was 53.4% for cross-validation method. It would be worthwhile to note that the cross-validation method achieves a nearly unbiased estimate but with a relatively large variance.

DISCUSSION

Morphometric traits as affected by district and age

District showed a significant ($p<0.001$) effect on all quantitative traits except RL, RW, and HL. The observations are consistent with the reports of Wagari *et al.* (2020) for sheep populations of western part of Ethiopia. The Asasa sheep were characterized by short height as measured by their WH and RH (both represent height), with the body being close to the ground, which might correspond to their adaptation to mountainous terrains. Moreover, the Asasa sheep were described by small body size as measured by their BL, CC, and TC values being the lowest compared with the other sheep populations. Sheep of Kofele district were the heaviest possessing the longest body size as measured by their LW and BL, respectively. This may suggest that the productivity of Kofele sheep related to its adaptation to the highland altitudes, an environment that favors sheep keeping. The longest EL was observed in Asasa and Kofele sheep, which is consistent with the observations of Wagari *et al.* (2020), who reported a similar value (11.8 vs. 11.9 cm) for Guduru sheep reared in western part of Ethiopia. Abdallah and Omar (2017) reported EL values for Awassi ewes ranging from 19.5 to 22.3, which is almost twice as long as the ear length of the Ethiopian sheep reported here. Ear length has been related to the adaptation potential of small ruminant animals in a specific production environment such as exposure to extreme heat stress conditions (Elbeltagy *et al.*, 2016).

The overall average LW for all districts (26.5 kg) in the present study is similar to that of Wagari *et al.* (2020) who reported 26.4 kg for female sheep populations from Dendi, Guduru and MidaKegn districts of western Ethiopia. Melesse *et al.* (2013) also reported similar LW, WH and BL values for female sheep populations reared in five administrative zones of southern Ethiopia. However, the same authors reported lower HG for the same sheep than observed in the average values of the current study. Likewise, Wagari *et al.* (2020) reported higher HG, WH, BL and RH values for female sheep than observed in the average of values these traits. Taye *et al.* (2016) also reported higher HG, WH, and RH for Doyogena female sheep compared with the average values of the current study. The manifestation of such variations in the indicated quantitative traits in the literature might be attributed to differences in the management practices among the communities and availability of feed and water resources. The indigenous sheep populations are also mostly bred at low selection intensity, and might be subject to high natural selection pressure resulting in considerable variations among indigenous livestock populations with respect to the expression of the morphometric traits.

Except CC, CL and TC, other morphometric traits significantly increased with the age of the sheep suggesting that these traits were little influenced by season (wet and dry season) which is mainly characterized by the availability of feed resources among others. This observation is consistent with those of Mohammed *et al.* (2018) and Wagari*et al.* (2020) who reported increase of all measured morphometric traits with the age of the sheep reared in North Eastern and western parts of Ethiopia, respectively. Yakubu (2013) reported a significant effect of age on all the morphological characters except for EL.Edea*et al.* (2009) and Taye *et al.*(2016) reported a non-significant effect of age on TC of local sheep, which is in good agreement with current finds. The interaction of district with age showed a significant effect for HG, WH, BL, CD, RL, HL, EL, and CL which suggests that some districts influence the development of these traits with the age of the sheep. In this regard, Kofeleagro-climate, which is dominated by high elevation, appeared to favor the sheep productivity for most of the studied morphometric traits. Moreover, the significant interaction effects of district by age may indicate the separate rankings of the sheep populations under ages investigated.

Multivariate analysis of morphometric traits

The stepwise discriminant analysis identified fifteen significant discriminatory traits (to discriminate among the sheep populations of the four districts) with BL, TC, CW, and TL showing the highest discriminating power. These identified morphomtetric traits in the studied sheep could be utilized as references to develop future conservation and breeding strategies. These observations are consistent with the reports of Abdallah and Omar (2017) who reported similar traits that had discriminating power in differentiating the Awassi female sheep breed in three regions. Most of these traits were also significant contributors to discrimination among geographical regions in Spanish Assaf (Legaz *et al.*, 2011) and Ethiopian (Melesse *et al.*, 2013) indigenous sheep breeds.

The application of cluster analysis identified two clusters in which cluster one displayed the Asasa sheep as a separate independent group and cluster two embraces all other sheep with two sub-clusters in which sheep of Bora and ATJK grouped in one and those of Kofele in the other. The grouping of Bora and ATJKsheep populations collectively under one sub-cluster could be explained by the fact that these two ecotypes tend to have comparable values in most of the morphometric traits. Moreover, both sheep populations share similar geographical terrains that cover most (over 80%) of the lowland regions of the studied districts (Table 1). This observation indicates the existence of genetic similarity among sheep populations due to gene flow caused by the exchange of flocks through common market outlets. Moreover, the shortest Mahalanobis distance was noted between sheep of ATJK and Bora districts further confirming their genetic similarity.

On the other hand, the cluster analysis indicated that the Asasa sheep were separately groupedfar away from the threesheep populations, which could be explained by the fact that this sheep population tend to have the shortest height and the smallest body size, which apparently differentiated them into a separate group. Besides, this observation has been further supported by the results of the Mahalanobis distances in which the Asasa sheep had the longest distances (ranging from 1.88 to 3.60) with the other three sheep populations. Separate grouping is an indication that the sheep populations of the Asasa district may possess different morphometric qualities and characteristics that might be attributed mainly to geographical origin of the ecotypes. Differences in the origin of farm animals could influence phenotypic response based on potential for additive genes controlling the quantitative traits (Mulyono *et al.*, 2009).

Most of the discriminating variables in the present study are similar to those reported by Wagari *et al.* (2020) for sheep of western Ethiopia, that of Abdallah and Omar (2017) for Awassi sheep of Palestine, and Yunusa *et al.* (2013) for Nigerian sheep populations. Canonical discriminant analysis finds linear combinations of the quantitative variables that provide maximal separation between classes or groups. Determining the morphological distances will help to comprehend the genetic diversity of the indigenous animal populations and enable to initiate suitable breeding programmes that are useful for the conservation of the animal geneticresources.

The Wilks' Lambda, which presents the ratio of within-group variability to total variability on the discriminating variables, is an inverse measure of the importance of the discriminant functions (Huberty and Olejnick, 2006). In the present study, the value of Wilks' Lambda for the sampled population was 0.53 (53.0%), which indicates that almost half (47.0%) of the variability in the discriminator variables was because of the differences between the sheep populations rather than variation within the population. In this regard, the discriminant analysis carried out provided complementary information in which about 59.7% of the individual sheep were correctly classified to their source population, which indicates the existence of genetic heterogeneity of sheep populations across populations for those variables included in the discriminant analysis. However, only about 36.0% of the original ATJK sheep were correctly classified into their respective population while the majority (about 74.0%) of the remaining being misclassified to the other sheep populations. This observation may suggest a strong intermingling of the ATJK sheep with the other ecotypes with almost similar misclassification rates (15.9to 27.3%) across the three districts (Table 8). In agreement with the current findings, Yakubu and Ibrahim (2011) reported on Nigerian indigenous sheep population that 41.2% of Uda sheep were misclassified as Yankasa sheep and 35.4% of Yankasa as that of Uda sheep.

The Mahalanobis distance is the most commonly used distance measure for quantitative traits of livestock breeds. All the Mahalanobis distances in the current study were significant which is consistent with the findings of Birteeb *et al.* (2013) and Wagari *et al.* (2020) for Northern Ghana sheep and western Ethiopia sheep, respectively.The significant differences among distances indicated that differences among sheep populations are important for the classification process. It also signifies theexistence of variations among the quantitative traits of the studied populations. The longest Mahalanobis distance occurred between Asasa and Bora sheep populations (3.60) possibly due to geographical isolation of these herds, consequently limiting the flow of genes across generations. The shortest Mahalanobis distance observed between ATJK and Borasheep populations may suggest that that they share similar phenotypic similarities, which might have been resulted from non-selection, continuous inbreeding, and admixture among these populations due to migration or ram exchange programs over several generations. Moreover, geographic proximity may have facilitated gene flow between the ATJK and other populations particularly to that of Bora sheep.

Yakubu and Ibrahim (2011) reported a Mahalanobis distance of 1.79 between Uda and Yankasa sheep, which is consistent with the current results observed between Bora and Kofele sheep (1.76). The Mahalanobis distances between Asasa and Kofele sheep populations in the current study are comparable to those reported by Wagari *et al.* (2020) among Dendi and Guduru sheep; but were much lower than those reported by Abdallah and Omar (2017) for Awassi sheep. Such large variations might arise in the methods applied for computing the Mahalanobis distances. For example, pairwise distances computed using canonical discriminant analysis might be different from the one analyzed using the discriminate function that produces squared Mahalanobis distances (Abdallah and Omar, 2017). Moreover, the number of samples used in the discriminant analysis would influence the outcome of the Mahalanobis distances being higher in smaller sample size than in larger (Melesse *et al.*, unpublished data).

It is apparent that for the total traits used in this study both canonical variables were adequate to explain the total variation (96.0%), indicating large reduction in sample space, with little loss (4.0%) to explain the total variation. Most of studies dealing with goats suggested that canonical analysis could minimize sample space with a loss of similar to this value (Jimcy *et al.* 2011; Arandas *et al.*, 2017).In a study conducted by Traoré *et al.* (2008) for Burkina Faso sheep and Legaz *et al.* (2011) for Spanish Asaf sheep, all the canonical variables extracted were found to be significantly different and are consistent with the current results. In the present study, CAN1 and CAN2 explained 69.0% and 27.0% of the total variation while the third CAN (CAN3) accounted for only 4% of the total variation being its contribution insignificant ($p = 0.3919$). Therefore, the third CAN does not contribute much significantly in the discrimination process as compared to that of CAN1 and CAN2. The current observations are lower than that of Wagari *et al.* (2020) who reported that the first canonical variable accounted for 80.6% of the variation for sheep of western part of Ethiopia. On the other hand, Ogah (2013) reported 59.7 and 40.3% of the total variation for CAN1 and CAN2, respectively, which is lower than found in the present study. The observed variations for each canonical variable in the literature might be due to differences in genetic makeup of the sheep studied and management practices applied by different countries.

The standardized canonical coefficients indicated the partial contribution of each variable to the discriminant function, controlling for other attributes entered in the equation. The first canonical variable CAN1 loaded highly for RH, which is in good agreement with the observations of Birteeb *et al.* (2013) and Wagari *et al.* (2020) who reported RH as a principal trait to differentiate the Sahel from the Djallonke sheep and the four local sheep populations in western Ethiopia, respectively. The CAN2 highly loaded for CD as important discriminant variable among other traits. Results of canonical structures were also in line with that of total standardized canonical coefficients. Wagari *et al.* (2020) also reported a strong influence of EL and WH on CAN1 that differs from the present observation. Age, genetic make,and production environments of the givensheep populations might be responsible for differences reported by various authors.

Overall, 60.5% the sheep populations were correctly assigned to their source population using the quadratic discriminant function with 15 morphometric variables, which indicates the existence of heterogeneity among the sheep populations across districts for those variables included in the discriminant analysis. Nevertheless, the overall average correct classification for sampled populations was relatively low with high proportion of misclassification rates. The large number of misclassified individuals was particularly observed in ATJK sheep,which indicates a higher degree of intermingling with the other sheep ecotypes. The existence of genetic variation within and between populations is essential for the populations to adapt to frequently changinglocal climate and to respond to artificial selection successfully (Toro *et al.*, 2011).

Wagari *et al.* (2020) correctly classified 71.2% of Dandi sheep to their genetic group, which is similar to those of Asasa sheep in which 72.1% of them were assigned to their original group. The same authors also reported the classification of 69.2% of MidaKegn sheep into their genetic group, which is consistent to the present study where68.6% of Kofele sheep were correctly assigned to their source group. About 60.3% (semi-arid), and 38.5% (dry subhumid) of indigenous goats of Limpopo province in South Africa were correctly allocated into their original agro-ecological zone by Selolo *et al.* (2015), which is comparable with the current findings for Bora and ATJK sheep populations (with the respective values of 64.7 and 36.4%). The classification of Bora and ATJK sheep into their source population was also in good agreement with those of Yakubu and Ibrahim (2011), who reported that 61.5% and 33.5% of Balami and Uda sheep breeds were assigned to their source population.

Animal genetic resources in developing countries like Ethiopia are components of biological diversity, which play a significant role in meeting the food requirement of these nations where the effect of climate change has been a major challenge to the main stay of the rain fed agriculture. Between-breed diversity has beentraditionally considered as a major criterion to be taken into account when setting priorities for conservation of domestic animal breeds. In the present study, both Asasa and Kofele sheep have shown significant deviations from each other as well as from the other two sheep populations. The Kofele sheep were particularity superior in most of economic important traits including LW, BL, HG and CD.

CONCLUSIONS

District and age had a significant effect on most of the morphometric traits studied. The highest live weight, height at withers and body length was noted for Kofele sheep being the tallest among the other sheep with a large body dimension. Sheep of ATJK and Bora were clustered together while those of Asasa were grouped distantly. Among the studied morphometric traits, body length, tail length and circumference, chest width, and live weight were identified as the best discriminating variables to differentiate the four sheep populations. All pairwise Mahalanobis distances were significant being the shortest between sheep of ATJK and Bora districts and the longest among those of Asasa and Bora. The CAN1 and CAN2 explained 69.0% and 27.0% of the total variation, respectively. The CAN1 highly loaded for rump height while CAN2 for chest depth with the respective discriminant function scores of 0.765 and 0.530. Most of the sheep populations were correctly assigned into their districts of origin except those of ATJK. However, the canonical discriminant analysis has showed a visible overlapping among the four district sheep populations indicating the existence of morphological homogeneity among them. The current findings further revealed that the Kofele sheep were particularity found to be superior in most of economic important traits and would be justifiable to consider this unique ecotype in a community-based breeding program as a conservation strategy for its sustainable utilization.

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Conflict of interest

Authors declared that there is no conflict of interests.

Authors' contribution: AW prepared the proposal; collected all data in the field; entered all data to spread sheet; prepared the first draft manuscript. AM reviewed the proposal; supervised the field research activity; run all statistical analysis; prepared the final manuscript.

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