Small Ruminant Research xxx (2011) xxx–xxx

Contents lists available at ScienceDirect

Small Ruminant Research

journal homepage: www.elsevier.com/locate/smallrumres

Multivariate characterisation of morphological traits in Assaf (Assaf.E) sheep

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A R T I C L E   I N F O

Article history:
Received 15 February 2011
Received in revised form 7 June 2011
Accepted 8 June 2011
Available online xxx

Keywords:
Assaf
Morphology
Mammary morphology
Zoometry
Multivariate analyses

A B S T R A C T

Seventeen body measurements in a total of 341 adult individuals (61 males and 280 females) in addition to eight udder scores collected during the period of maximum levels of lactation in the 280 females were used to morphologically characterize the Assaf.E breed both in magnitude and variability. Sampling included the two main environmental areas to check if the native dairy sheep breeds, namely Churra and Manchega, leaded to differences in the male-mediated absorption. Standard morphology of the Assaf.E breed was assessed with a live weight of 110.47 ± 12.51 kg and 75.74 ± 11.23 kg respectively for males and females. The sexual dimorphism (m/f) was 1.13 as expected, with males being 46% heavier than females. The coefficient of variation of all traits ranged from 3.73% to 15.00%, showing high uniformity. Canonical analyses and Mahalanobis distances showed that differences in body measurements between regions existed but they were small as expected in a unique breed. The breed has shown itself to be slightly longer than others with deeper udders and more angled teat placement. Some peripheral traits such as ear and tail size, usually considered important in the breed definition, have been shown to have a low, or null, relationship with other morphological traits. Even when homogeneity is found in the breed, there are still some small differences found between geographical areas as a consequence of the short history of the breed in Spain. However, after a 30 year history in Spain, the homogeneity of the Assaf.E breed allows this population to be defined as an authentic breed.

1. Introduction

Spanish sheep population numbers roughly 19 million heads, of which approximately 3.5 million are dairy sheep. Of that, 45% belong to foreign breeds and roughly 700,000 heads belong to the Spanish Assaf (Assaf.E) population (Ugarte et al., 2001). The Assaf sheep breed was created in Israel by crossbreeding the East Friesian (Milchschaf) with the Awassi breed (Goot, 1986). The importation of Assaf individuals into Spain started in 1977 and had finished by 2000. From this time and within the so considered closed Assaf.E population, the exchange of animals between farms, sometimes located in different areas, has been steady but irregular from a few animals to entire herds. This exchange has lead to a more homogeneous breed despite no action having been undertaken to achieve uniformity. At present, the breed is well developed with a number of dairy recording schemes (Jiménez and Jurado, 2005; Gutiérrez et al., 2007) that have recently merged into a single breeding organization officially recognized by the

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doi:10.1016/j.smallrumres.2011.06.005

Spanish Government. Although mating with other breeds stopped in 2000, since 2005 the organization controls the entry of new animals in the official flockbook in order to avoid new hybridization. Approximately 200,000 Assaf.E heads owned by roughly 300 farmers are currently registered in the official flockbook (http://www.assafe.es/).

The introduction of the Assaf sheep breed has occurred basically by the male-mediated absorption of Spanish native dairy sheep breeds such as Castellana, Churra, Manchega or, to a lesser extent, Latxa (Ugarte et al., 2002; Legaz et al., 2008). Recent molecular studies have tried to clarify the absorption process. Pedrosa et al. (2007) did not find differences at the mitochondrial DNA level among the Spanish Assaf and the Spanish native dairy sheep breeds, thus supporting the hypothesis of a male mediated absorption of native sheep during the formation of the Spanish Assaf breed. Legaz et al. (2008), using microsatellites, reported that Spanish local genetic background added to the Assaf.E breed mainly comes from the Churra, Castellana and Manchega sheep breeds. Although the homogenised appearance of the Assaf.E individuals, there still ought to be type differences according to geography, making the establishment of a standard for the breed difficult.

Even though the Assaf.E individuals are in high demand in other countries such as Greece, information on the population is still scarce (Gutiérrez et al., 2007; Legaz et al., 2008) and should be gathered first. Since the breed is the operation unit for the assessment of livestock diversity all over the world (Duchev and Groeneveld, 2006; Duchev et al., 2006; Simon, 1999), contributions to characterisation of local domestic animal populations are of major importance.

The first step to the characterisation of local genetic resources falls on the knowledge of the morphological trait variation (Azor et al., 2008; Delgado et al., 2001). Multifactorial analyses of morphological traits have been shown to be suitable to assessing variation within and can discriminate different population types when all measured morphological variables are considered simultaneously (Traoré et al., 2008a). These kinds of studies are commonly used with goats (Capote et al., 1998; Dossa et al., 2007; Herrera et al., 1996; Jordana et al., 1993; Zaitoun et al., 2005; Traoré et al., 2008b); however, multivariate analyses on morphological traits are rarely reported in sheep (Riva et al., 2004; Carneiro et al., 2010).

In this study, we analysed seventeen body measurements in 341 individuals (61 male and 280 female) ranging from 2 to 5 years old in addition to eight udder scores collected during the period of maximum levels of lactation in the 280 females. Samples were obtained throughout the area of influence of the Assaf.E breed in order to characterise the morphological variation in the whole population. The general aim was to contribute to the establishment of a standard for the breed both in magnitude and variability. The study will be also valuable as an example of introgression of a foreign population into local breeds.

2. Materials and methods

2.1. Data

During the summer of 2009, sampling was carried out on a total of 18 Assaf.E flocks, selected at random, located in 9 different provinces in central Spain which covers the main area of influence of the breed. Two main subareas can be defined according to geography and Spanish local sheep breeds: (a) Northern Central Spain (Castilla-León; R1); and (b) Madrid and surrounding provinces (R2). These two subregions are separated by the Central Mountainous Range. The main Spanish local sheep spread in R1 and R2 are, respectively, the Churra and the Manchega breeds. From these the first region involves roughly 85% of the heads, and, thus, the sampling aimed to be representative of this distribution.

A total of 280 female individuals (217 and 63 respectively for R1 and R2) from 2 to 4 years old, and 61 males (53 and 8 for R1 and R2, respectively) from 2 to 5 years old belonging to 18 flocks (14 and 4 flocks for R1 and R2, respectively) were scored for seventeen body measurements in addition to eight udder traits in the females. Live weight (LW) was obtained first. The zoometric variables measured were live weight (LW), head width (HW), chest width (CW), anterior croup width (ACW), posterior croup width (PCW), croup length (CL), thorax perimeter (TxP), cane perimeter (CP), head length (HL) cross height (CH), croup height (CrH), longitudinal diameter (LD), dorsoesternal diameter (DD), bicostal diameter (BD), tail width (TaW), ear length (EL), and ear width (EW).

The methodology used for measuring udder traits were those described by Labussiere et al. (1981) and used in ovine breeds from the Mediterranean basin. The morphological udder traits measured were udder length (UL), udder depth (UD), udder width (UW), cistern height (CiH), teat placement (TP), teat length (TL), teat width (TW), and teat angle (TA). LW was determined with scales, and measurements were carried out using a Lydthin stick, tape measure, and Vernier calliper. TP was subjectively scored in lateral view from 1 = turned backwards, 2 = vertical, 3 = a little forward, 4 = forward and 5 = much forward). Animals were put on a flat floor and held down by the respective owners with measurements being obtained by a single technician. Females were measured during the period of maximum levels of lactation.

2.2. Statistical analyses

Statistical analyses were carried out using the SAS/STAT package (1999). Basic statistics for the body measurements and qualitative traits were obtained using the PROC UNIVARIATE and PROC FREQ, respectively. PROC CORR was also used to compute the Pearson correlations between traits. The influence of the area on the body traits measured was assessed using the PROC GLM, fitting a model which includes the area effect with 2 levels, the flock effect with 18 levels (nested into the area effect), and the age effect with 3 (males) or 4 (females) levels beginning in the second year. Least square means and their corresponding standard errors were obtained for each body trait by flock level. The PROC CANDISC was used to perform canonical analyses to derive canonical functions, that is, linear combinations of the quantitative variables to summarise variation between flocks and compute the between-flocks Mahalanobis distance matrix. The association between the qualitative traits was assessed via a correspondence analysis using the PROC CORRESP. When necessary for descriptive purposes, canonical variables and correspondence analysis dimensions were plotted using Microsoft Excel™.

3. Results

Least-squared means for the analysed body traits both for males and females are provided in Table 1. This is the first time that the morphology of the Assaf.E breed has been assessed with an Assaf.E LW of 110.47 ± 12.51 kg and 75.74 ± 11.23 kg being obtained respectively for males and females (Table 1), higher than the 80–100 kg and 60–70 kg defined as the current breed standard. The measure of sexual dimorphism (m/f) has been included in Table 1 to express these differences between males and females. The global mean of this value (1.13) was as expected, with males being 46% heavier than females, as were values for the other variables, except for ears (EL and EW) which demonstrated no significant differences. The coefficient of variation of all traits ranged from 3.73% to 15.00%.

Table 1 also shows the significance of the age of the animal and area and flock effects on the traits analysed both in

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Table 1
Raw means, standard deviation (SD), coefficient of variation (CV) and significance of area, flock and age effects for each of the morphological and udder traits in both males and females.

	Males					Females					Sexual dimorphism (m/f)
	Statistics		Fixed effects			Statistics		Fixed effects			
	Mean	CV	Area	Flock	Age	Mean	CV	Area	Flock	Age	
Body traits											
LW (kg)	110.47	11.32%	NS	NS	NS	75.74	14.82%	NS	***	***	1.46
HW (cm)	14.52	4.75%	NS	**	*	13.01	4.11%	NS	***	**	1.12
CW (cm)	26.85	7.45%	NS	*	NS	22.86	6.23%	NS	*	***	1.17
ACW (cm)	21.83	8.34%	***	*	NS	20.43	5.57%	*	***	**	1.07
PCW (cm)	18.41	7.30%	NS	***	NS	17.18	5.81%	NS	***	NS	1.07
CL (cm)	23.97	4.41%	NS	*	*	22.25	5.53%	NS	*	NS	1.08
TxP (cm)	117.30	4.83%	NS	NS	NS	105.68	6.31%	***	***	***	1.11
CP (cm)	10.82	7.41%	NS	NS	NS	8.95	5.51%	NS	***	NS	1.21
HL (cm)	31.11	4.69%	NS	NS	NS	26.60	4.08%	NS	*	*	1.17
CH (cm)	83.51	3.82%	NS	*	*	74.10	4.52%	NS	***	NS	1.13
CrH (cm)	84.18	3.73%	NS	NS	*	75.94	4.31%	NS	***	NS	1.11
LD (cm)	82.16	4.81%	NS	**	NS	73.09	4.83%	NS	***	*	1.12
DD (cm)	38.82	5.61%	NS	NS	*	34.65	5.70%	***	**	**	1.12
BD (cm)	27.89	7.97%	*	NS	NS	25.51	9.93%	***	***	**	1.09
EL (cm)	17.90	9.99%	NS	NS	NS	18.19	9.28%	NS	***	NS	0.98
EW (cm)	10.33	11.23%	NS	NS	NS	10.12	8.00%	NS	NS	NS	1.02
TaW (cm)	14.45	14.12%	NS	***	NS	12.35	15.00%	***	***	**	1.17
Udder traits											
UL (cm)						11.26	13.73%	***	***	NS	
UD (cm)						19.53	16.30%	NS	***	***	
UW (cm)						9.81	13.67%	***	***	NS	
CiH (cm)						4.67	31.68%	NS	***	**	
TP (cm)						2.78	21.96%	NS	NS	NS	
TL (cm)						3.06	23.43%	**	***	NS	
TW (cm)						1.75	18.55%	NS	***	NS	
TA (grades)						63.25	16.85%	NS	***	*	

NS, non-significant. $n = 61$ for males except for Ew ($n = 29$) and $n = 280$ for females except for Ew ($n = 107$).
* Significance level: $p < 0.05$.
** Significance level: $p < 0.01$.
*** Significance level: $p < 0.001$.

males and females. The sexes were analysed separately due to the differences in sample size and, more importantly, to the high sexual dimorphism found. Area effect was only slightly significant for ACW and BD in males and in addition to TxP, DD and TaW in females, the flock effect having a stronger influence in general. In males, the flock effect was highly significant ($p < 0.001$) for TaW, while the age effect had only a significant effect ($p < 0.05$) for five (HW, CL, CH, CrH and DD) out of the seventeen analysed traits, showing a high homogeneity among males of different flocks. In contrast, the flock effect in females resulted non-significant only for EW, showing a high heterogeneity across herds. The age of the females showed different influences across traits varying between highly significant (LW, CW and TxP) and non-significant (PCW, CL, CP, for females, 95% confidence intervals were built for all morphological traits (Johnson and Welch, 1940; McKay, 1932). Males were only significantly more heterogeneous for ACW (8.34% vs. 5.57%) and CP (7.41% vs. 5.51%) with no differences in homogeneity being found within sex and between areas except very slightly for ACW and PCW in males, and CL in females. No differences in homogeneity between regions were found for the udder traits either (Table 2).

Least-squared means for the udder traits, as well as the significance of the flock and age effects, are also given in Table 1. These values are obviously conditioned for a high variety of uncontrollable environmental circumstances such as nutrition and time spent since the last milking event, and should not be used to define the breed. Area effect was only highly significant for UL and UW ($p < 0.001$). Again flock effect was highly significant except for TP, while age effect only seemed to condition the UD ($p < 0.001$), CiH ($p < 0.01$) and TA ($p < 0.05$) traits.

Significant correlations between traits are shown in Table 3 for males and females. Regarding females, all morphological traits showed significant correlations; however, the ear-related traits (similar as in males) showed a different behaviour. TaW was highly correlated to the other morphological traits while ears, both EW and EL, seemed to be independent of the size of the animal. Correlation between udder traits ranged from non-significant (22% of them) to 0.76 in absolute value. This characterises udder trait as an independent group of morphological traits. 38% of the correlations assessed between body and udder traits were non significant with 0.32 being the highest absolute value of the others. EW, EL and TL showed null or very low correlation with udder traits. Fig. 1 shows a bidimensional plot built with the first two canonical variables for body measurements. On the *Y*-axis, the udder traits are separated from most of the other morphological traits. Curiously, TaW is located in the centre of the cloud formed by those udder morphological traits. There is not a clear

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Table 2
Significant phenotypic correlations among all measured traits in males (above the diagonal line) and in females (below the diagonal line). Correlations higher than 0.25 are in bold.

	LW	HW	CW	ACW	PCW	CL	TxP	CP	HL	CH	CrH	LD	DD	BD	EL	EW	TaW	UL	UD	UW	CiH	TP	TL	TW	TA
LW	**1.00**	**0.47**	**0.66**	**0.47**	**0.68**	**0.56**	**0.83**	**0.54**	**0.60**	**0.48**	**0.44**	**0.45**	**0.62**	**0.68**			**0.41**								
HW	**0.40**	**1.00**	**0.41**		**0.38**	**0.33**	**0.40**	**0.32**	**0.34**	**0.39**	**0.31**		**0.59**	**0.33**											
CW	**0.75**	**0.43**	**1.00**	**0.28**	**0.62**	**0.49**	**0.61**	**0.39**	**0.46**	**0.44**	**0.42**	**0.28**	**0.45**	**0.48**			**0.34**								
ACW	**0.68**	**0.42**	**0.57**	**1.00**	**0.47**	**0.32**	**0.45**	**0.31**	**0.35**	**0.30**	**0.36**		**0.36**	**0.41**			**0.45**								
PCW	**0.56**	**0.36**	**0.53**	**0.62**	**1.00**	**0.49**	**0.58**	**0.49**	**0.49**	**0.44**	**0.46**	**0.40**	**0.53**	**0.43**	**0.38**	**0.45**	**0.42**								
CL	**0.42**	**0.21**	**0.43**	**0.36**	**0.30**	**1.00**	**0.41**	**0.36**	**0.32**	**0.39**	**0.49**	**0.29**	**0.43**	**0.30**	**0.32**	**0.38**	**0.30**								
TxP	**0.85**	**0.41**	**0.68**	**0.67**	**0.52**	**0.37**	**1.00**	**0.52**	**0.45**	**0.38**	**0.29**	**0.30**	**0.63**	**0.73**			**0.39**								
CP	**0.44**	**0.30**	**0.45**	**0.33**	**0.29**	**0.34**	**0.40**	**1.00**	**0.34**	**0.39**	**0.36**		**0.41**	**0.39**		**0.39**	**0.43**								
HL	**0.34**	**0.29**	**0.37**	**0.33**	**0.27**	**0.29**	**0.35**	0.22	**1.00**	**0.61**	**0.46**	**0.52**	**0.45**				**0.53**								
CH	**0.30**	**0.32**	**0.26**	**0.33**	**0.34**	**0.28**	**0.33**	**0.28**	**0.32**	**1.00**	**0.69**	**0.38**	**0.46**	**0.28**	**0.26**		**0.29**								
CrH	**0.42**	**0.31**	**0.31**	**0.41**	**0.40**	**0.31**	**0.41**	**0.33**	**0.31**	**0.84**	**1.00**	**0.31**	**0.37**	**0.26**	**0.40**	**0.62**									
LD	**0.50**	**0.34**	**0.38**	**0.42**	**0.48**	**0.35**	**0.42**	**0.33**	**0.38**	**0.36**	**0.39**	**1.00**		0.25			**0.32**								
DD	**0.63**	**0.41**	**0.51**	**0.60**	**0.52**	**0.33**	**0.70**	**0.39**	**0.37**	**0.52**	**0.54**	**0.38**	**1.00**		**0.53**										
BD	**0.67**	**0.41**	**0.56**	**0.56**	**0.40**	0.25	**0.72**	**0.31**	**0.25**	0.19	**0.30**	**0.30**	**0.52**	**1.00**	**1.00**										
EL		0.11			0.18					0.15	0.18	0.11			**1.00**	**0.81**	**0.28**								
EW		**0.27**	0.18	0.23	0.22			0.20		0.19	0.21		0.21	0.20	**0.62**	**1.00**									
TaW	**0.59**	0.22	**0.43**	**0.42**	**0.28**	0.17	**0.58**	0.22			0.12	0.21	**0.31**	**0.50**	−0.22		**1.00**								
UL	0.17			0.13		0.16	0.20			0.20	0.19		0.24	0.19			0.12	**1.00**							
UD	0.24	0.19	0.17	0.18	0.14	0.15	0.15		**0.26**	0.21	0.24	0.22	**0.32**						**1.00**						
UW	0.12			0.13	0.06		0.15			0.18	0.18		0.19	0.22				**0.32**	−0.11	**1.00**					
CiH	**0.27**	0.15	**0.27**	**0.26**	0.16	0.12	**0.26**		0.24	0.16	0.13	0.22	**0.27**	0.19					**0.52**	−0.23	**1.00**				
TP			−0.13																		**−0.26**	**1.00**			
TL											0.11		0.12					0.11	0.18	0.12	−0.17	0.13	**1.00**		
TW	0.13	0.16				0.16			0.11		0.11		0.12					0.16	0.23		−0.11	0.18	**0.76**	**1.00**	
TA	0.23	0.15	0.22	0.16	0.11	0.12	0.17		0.15	0.14	0.17	0.16	0.20		0.15				**0.32**	−0.20	**0.60**	**−0.27**	−0.22	−0.17	**1.00**

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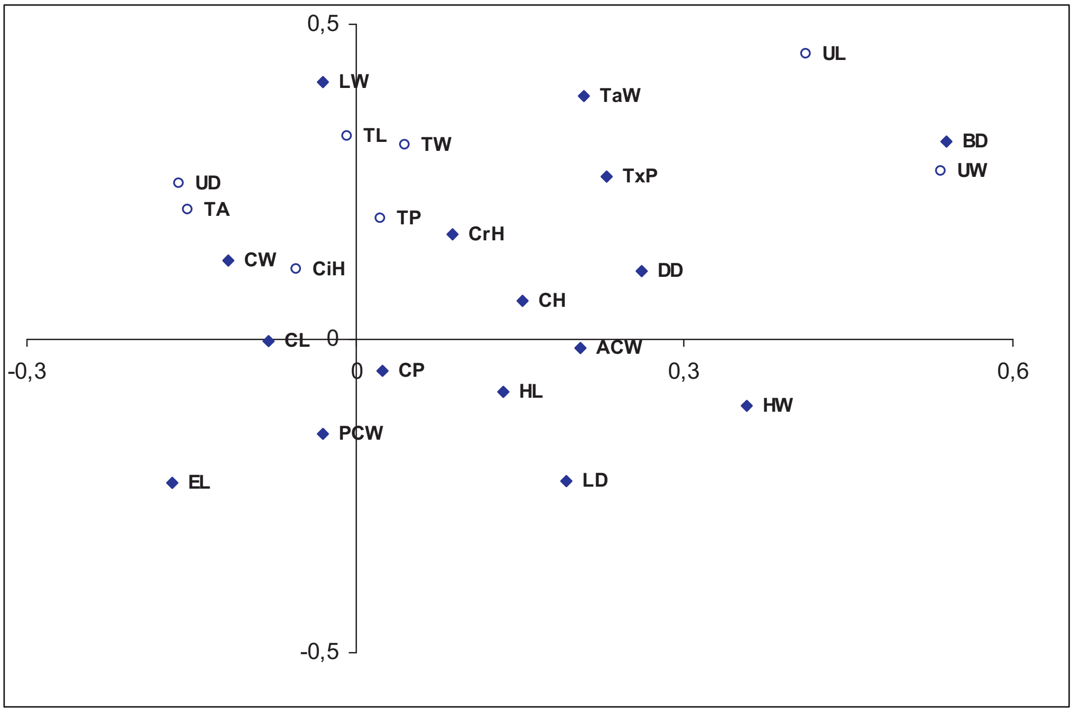

LW: Live Weight; HW: Head Width; CW: Chest Width; ACW: Anterior Croup Width; PCW: Posterior Croup Width; CL: Croup length; TxP: Thorax Perimeter; CP: Cane Perimeter; HL: Head Length; CH: Cross Height; CrH: Croup Height; LD: Longitudinal Diameter; DD: Dorsoesternal Diameter; BD: Bicostal Diameter (BD); TaW: Tail Width; EL: Ear Length; UL: Udder Length; UD: Udder Depth; UW: Udder Width; CiH: Cistern Height; TP: Teat Placement; TL: Teat Length; TW: Teat Width; TA: Teat Angle.

Fig. 1. Bi-dimensional plot illustrating the association between body measurements in Assaf.E sheep breed assessed via canonical analysis. Udder traits are marked with circles. LW: live weight; HW: head width; CW: chest width; ACW: anterior croup width; PCW: posterior croup width; CL: croup length; TxP: thorax perimeter; CP: cane perimeter; HL: head length; CH: cross height; CrH: croup height; LD: longitudinal diameter; DD: dorsoesternal diameter; BD: bicostal diameter (BD); TaW: tail width; EL: ear length; UL: udder length; UD: udder depth; UW: udder width; CiH: cistern height; TP: teat placement; TL: teat length; TW: teat width; TA: teat angle.

differentiation on the *X*-axis, although EL is identified as an outlier measurement.

Table 3 shows the significant Mahalanobis distances between the flocks based on morphological measurements. 96% of the pairwise distances were significant ($p < 0.05$) demonstrating that differences between flocks are important. The hypothesis that the flock means are equal in the analysed population was also tested using Wilks' Lambda. This parameter had a significant value ($p < 0.0001$) of 0.05058254 ($F = 3.65$; degrees of freedom = 272) thus showing that differences found between flocks were statistically different from zero. After ordering the flocks according to the average distances between their locations, two out of the four flocks belonging to the centre zone (R2) were designated as the farthest. The other two R2 flocks were located between the middle and the farthest distance.

Fig. 2 shows the bi-dimensional representation of the canonical variables associated with the individuals sampled from the two different regions considering morphological and udder traits both separately and jointly. Animals from the central region (R2) are shown with clear marks while the others appear in black. It can be observed that in all three figures there was a substantial amount of overlap between both groups in addition to a differentiation between regions on the *X*-axis which represents the most important canonical variable, i.e., R2 individuals appearing within the cloud of points though principally in the left region.

The canonical analysis carried out on the morphological traits identified seven statistically significant ($p < 0.001$) canonical variables that accounted for 28.0%, 19.7%, 12.5%, 9.5%, 8.1%, 6.7% and 6.0% of the total variation, respectively, adding up to 90.5% of that total variance. Fig. 3 shows a bi-dimensional plot built with the first two canonical variables illustrating the relationships between farms belonging to different provinces. This was done by assigning the same symbol to the farms found in the same province. Black squares are used for the farms in the central region (R2) and appear close to one other. Again, even when included in a common cloud, R2 farms tend to appear in the left area.

4. Discussion

There is an increasing interest in the characterisation of the Spanish Assaf.E breed population because of its spreading as a consequence of its high milk production (Gutiérrez et al., 2007; Ugarte et al., 2001). More than 30 years after the introduction of the first Assaf individuals into Spain, the breed has been greatly developed and currently constitutes a single population which is managed together under

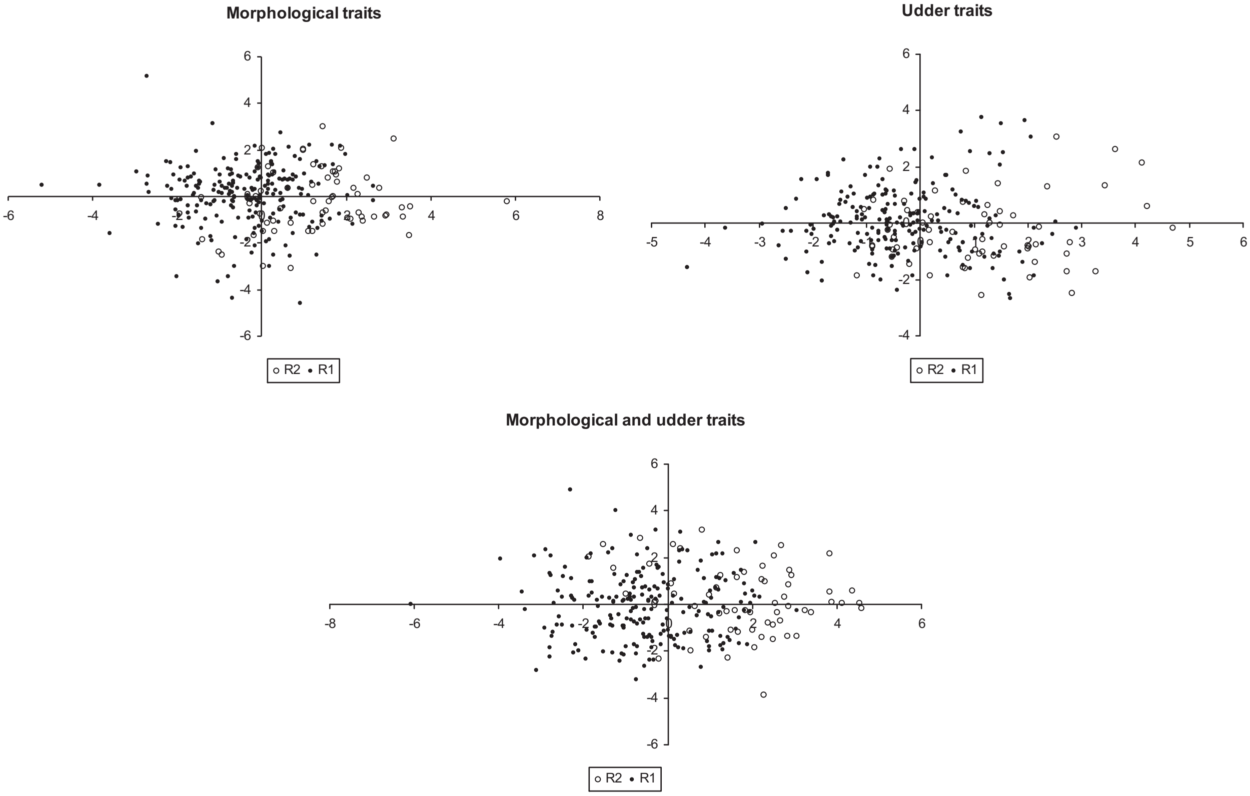

Fig. 2. Bi-dimensional representation of the canonical variables associated to the individuals sampled from two different regions, considering morphological and udder traits both separately and jointly. R1 black marks and R2 clear marks.

a unique breeding organization (Jiménez and Jurado, 2005; Jiménez and Jurado, 2010) interested in a morphological standardization. The standard morphotype may be further distinguished from the original breed in Israel given that it was introduced into Spain basically by the male-mediated absorption of Spanish native dairy sheep breeds such as Castellana, Churra, Manchega or Latxa (Ugarte et al., 2002; Pedrosa et al., 2007; Legaz et al., 2008). The time has come to determine its breed standard to facilitate the increasing exportation of Assaf.E individuals to other countries. These values would allow for the defining of the adult morphological standard of the Assaf.E breed from both the mean point of view as well as variability.

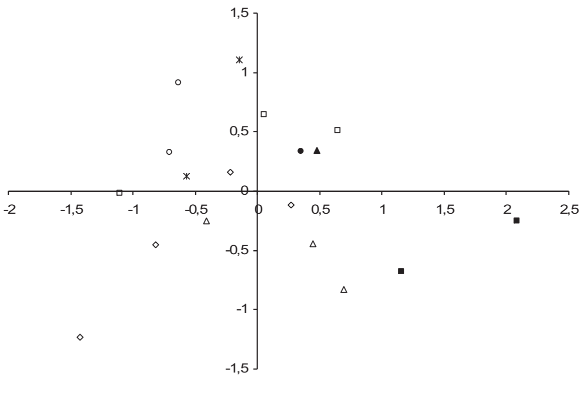

Fig. 3. Bi-dimensional plot illustrating the association between measured flocks in Assaf.E sheep breed assessed via canonical analysis. Flocks in the same province are marked with the same symbol. Black marks are used for region R2 (Madrid, black squares) and surrounding provinces.

The coefficient of variation was from 3.73% to 15.00%, with only EL, EW, LW and TaW higher than 10% which corresponds with a mean homogeneity within the breed (Herrera, 2007). Important information can be extracted from the mean coefficient of variation of the traits which showed that the variability of the zoometric measures was low. The overall coefficient of variation was similar in males (7.16) and females (7.03). However, when analyzing the 95% confidence intervals for the coefficient of variation of the traits, it can be concluded that, although sheep appear different between flocks, the overall heterogeneity of females is even lower than among males when viewed globally. Moreover, considering that sexual dimorphism was as expected, the actual Spanish Assaf.E population can be considered as having become a unique consistent population, even when its history reveals a high initial variability.

Area effect showed a very low significance in the analysed traits, leaving a more important influence for the flock effect nested into the area effect. Homogeneity within flock for males was higher than for females (Table 1). For males, flock effect was highly significant only for TaW, while for females it was significant for all the variables except for EW. The age effect was non significant for 5 and 10 variables respectively for males and females. The fact that there are fewer males could be an influencing factor in

Table 3
Significant Mahalanobis distances between the flocks, based on morphological measurements. Flocks belonging to the centre zone are in bold.

Flock	DGM	AV	HFM	OG	FA	HVV	HMG	EFR	SC	HP	MV	OLM	HBA	EC	RAC	SGA	AP	SLL
DGM																		
AV	3.628																	
HFM	3.612	4.995																
OG		6.459	5.043															
FA		3.254	4.466	3.467														
HVV	3.291	5.932	6.622		3.295													
HMG	3.781	7.476	3.469	3.441	4.062													
EFR	7.432	6.828	6.107	3.685	5.723	4.523	6.099											
SC	**6.325**	**4.567**	**5.857**	**7.991**	**7.459**	**9.005**	**7.483**	**8.341**										
HP	4.486	6.749	9.429	5.683	7.926	3.883	6.463	8.318	**8.849**									
MV	3.927	6.165	6.567		3.999	4.136	4.312	7.541	**6.314**	7.914								
OLM	8.191	6.157	5.701	8.767	10.320	11.850	10.418	7.618	**3.950**	9.827	9.694							
HBA	5.141	7.361	7.458		5.838	7.043	6.633	5.141	**10.146**	6.488	6.676	8.046						
EC	**4.242**	**7.316**	**9.480**	**5.832**	**8.453**	**5.889**	**9.739**	**10.184**	**9.272**	**7.526**	**7.660**	**11.433**	**12.150**					
RAC	7.696	7.290	6.356	8.709	7.250	7.942	8.980	9.380	**6.925**	9.346	9.698	7.732	14.835	**9.952**				
SGA	5.903	8.814	4.456	7.280	7.733	11.874	6.701	10.344	**6.821**	11.101	10.873	6.320	5.534	**14.398**	12.589			
AP	**8.487**	**7.603**	**9.488**	**9.799**	**12.764**	**10.058**	**13.301**	**8.261**	**6.239**	**6.751**	**12.943**	**5.355**	**12.784**	**6.233**	**6.614**	**13.383**		
SLL	**8.355**	**6.400**	**9.441**	**9.545**	**10.196**	**8.725**	**12.962**	**7.799**	**9.565**	**7.069**	**15.451**	**9.415**	**13.006**	**7.615**	**6.750**	**13.933**		
Mean	**5.633**	**6.294**	**6.385**	**6.592**	**6.638**	**6.938**	**7.207**	**7.254**	**7.359**	7.518	7.742	8.282	8.392	**8.669**	8.708	9.298	**9.379**	**9.764**

the signification; however, it seems as if there still existed important differences between flocks in ewes that do not exist in rams. Given that both the males and females included in this study were born in Spain after a couple of generations of mating within the new population, it seems that the selection via males within flock has lead to similar morphological animals, the females being less selected. Despite this, influence of flock effect on the variability seems to be in the expected range of any population including animals from the same breed. The area effect, nonetheless, including an important genetic component as it includes the effect of the breed mediated for absorption, did not result relatively significant enough to cause important heterogeneity in the data when compared with that originated by the flock effect.

Regarding udder traits, Table 1 shows that there was a higher variability for these traits than for the other morphological traits, with the coefficient of variation being from 13.67% to 31.68%. Part of this variability can be attributed to the flock effect, as described in the Churra breed (Fernández et al., 1995), though not to such an extent as the age effect. Again the area of influence did not greatly affect the traits nor their variability. It seems clear that environmental influence is higher in these traits with the obtained values not usually being considered as defining a standard morphotype of the breed. UL (11.3 cm) and TL (3.1 cm) were similar to those measured in other sheep breeds (Caja et al., 2002) such as Lacaune (11.3 cm and 2.9 cm respectively), Manchega (11.4 cm and 3.4 cm respectively) and Churra (9.30 cm and 3.83 cm respectively) (Fernández et al., 1995), although the udder was deeper, with wider cisterns, and longer and more angled teats as revealed by the mean values obtained in Assaf.E breed for UD (19.5 cm), CiH (4.7 cm), TW (1.75 cm) and TA (63°). This is in contrast with the respective values found by Caja et al. (2002) for the Lacaune breed (17.8 cm, 2.00 cm, 1.32 cm and 44°), for the Manchega breed (17.2 cm, 1.55 cm, 1.51 cm and 43°) and by Fernández et al. (1995) for the Churra breed (12.2 cm, 1.48 cm, 1.93 cm and 51°). Globally, it seems that udder can be considered as deeper and less harmonically angled than the Lacaune, Manchega and Churra breeds.

Regarding correlations, all morphological variables except those for udder, ears and tail were significant in females while in males 95% of them showed a high harmony degree (Herrera, 2007). TaW was moderately correlated to the other morphological traits while EL and EW seem to be independent of animal size. Udder traits showed important correlations between them but only those not including information on the teat had moderate correlations with the other morphological traits. Long ears and fat tail are usually associated by the farmers with the Assaf.E breed but only TaW seems to be somehow related with the size of the animal and the relationship between them with udder traits is still lower. Fig. 1 shows a bi-dimensional plot, accessed via canonical analysis, illustrating the association between body measurements in the Assaf.E sheep breed, with udder traits marked with circles. All these udder traits are distributed in an area within positive values of the *Y*-axis. Interestingly, TaW is located in the centre of the udder variables, while EL has the two lowest values on both coordinates showing complete independence with regards to other morphological and udder traits.

Several analyses have been conducted with the aim of describing the current similarities and differences between animals of the Assaf.E breed. Legaz et al. (2008) showed that animals in this population, wherever they were sampled, could be considered as belonging to the same population, although there may still remain some heterogeneity by flocks, most likely as a consequence of the breed used for introgression. Table 3 shows significant Mahalanobis distances between flocks based on morphological measurements sorted by mean distance, with the four flocks belonging to the R2 zone highlighted in bold. Two of those R2 flocks are the furthest from the geographical centre, and, of the other two, one is in the middle and the other is from the most remote. Figs. 2 and 3 lead to similar conclusions. Fig. 2 represents sampled animals using the first two canonical variables, with morphological and udder measurements, taken both individually and in conjunction, being taken into consideration; those marked in black symbols belong to the R1 region. While morphological differences can be mainly attributed mainly to genetic differences, udder traits are more likely to be affected by environmental influences in the area, namely management practices. No matter what area the information comes from, the pattern of the figure shows a uniform cloud even though individuals from the R2 region are located mainly in the negative values of the *X*-axis whereas R1 animals appear primarily in the positive zone. While globally it appears that the population can be seen as being uniform, when studying the individuals by regions, it is clear that some differences remain. The same conclusion is arrived at when looking at Fig. 3 which illustrates the association between measured flocks in the Assaf.E sheep breed assessed via canonical analysis. Flocks in the same province (more heavily influenced by the Churra or Manchega breed) are marked with the same symbol, and they can be grouped by region. Black marks are used for region R1: Madrid (black squares) and its surrounding provinces and are always found in the positive zone of the *X*-axis, with those in the province of Madrid being the most positive. Again, globally, flocks can be seen as a unique population; however, differences are found when their geographical situation, more heavily influenced by the Churra or Manchega breed, is revealed. Consequently, an exchange of artificial insemination sires between areas is recommended in order to achieve a more homogeneous spread of the same genetic base.

5. Conclusions

A morphological standard of the Assaf.E breed, needed but unknown to date, has been defined with this study. Some peripheral traits such as ear and tail size, usually considered important in the definition breed, have been shown to have a low, or null, relationship with other morphological traits. The breed has shown itself to be slightly longer than others with deeper udders and more angled teat placement. Even when homogeneity is found in the breed, there are still differences found between geographical areas as a consequence of the short history of the breed in Spain. However, after a 30 year history in Spain, the Assaf.E breed allows this population to be defined as being sufficiently homogeneous to be considered a breed different from the original Assaf from Israel. Given that a greater homogenization is expected after a few more generations have gone by, it is recommended that this study be repeated in the future. The present and future studies constitute a good example of introgression of a foreign population into local breeds.

Acknowledgments

Authors are indebted to the ASSAF.E farmers that collaborated in this study.

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