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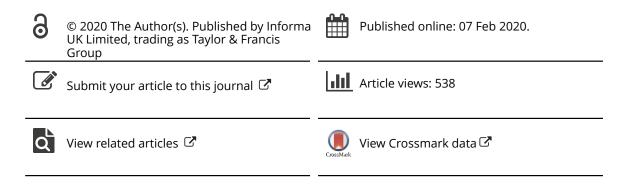
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Finding best exotic breed proportion in crossbred lactating sheep kept under farmers' conditions in Ethiopia determined by use of nested Legendre polynomials with limited data

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ABSTRACT

The present study was conducted to estimate milk production performance and fit lactation curves for groups of ewes of Local and of Awassi crosses, with a variable blood level, reared under farmer's environment. The Weigh-Suckle-Weigh method plus hand milking was used to estimate milk yield for ewes. A total of 466 observations from 115 ewes were used. Estimated least-squares adjusted means for the milk production over 120 days were 0.56 kg day⁻¹ (Local), 0.67 (<30% Awassi), 0.86 (30–50% Awassi), and 0.96 (>50% Awassi). Groups with 30–50% Awassi and >50% Awassi ewes produced significantly (p < 0.05) more milk than Local ewes. Significant differences were observed between <30% Awassi and >50% Awassi crossbred groups. The best crosses (>50% Awassi) produced over 70% more milk than the local ewes which demonstrates the potential that exists in increasing milk production through the initiated crossbreeding programme with sheep in Ethiopia.

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Test-day; local sheep breeds; Awassi crossbred; lactation curve; Legendre polynomial

Introduction

In Ethiopia, the major source of milk for human consumption comes from cattle, followed by camels (CSA, 2013). Small ruminants are mainly kept by smallholder farmers as a source of income from meat, milk and wool (Legesse et al., 2008; FAO, 2009; Abebe et al., 2013; Asresu et al., 2013). In the pastoral system of the Afar region to the north of Ethiopia, sheep are commonly used for milk in as well as for meat and skin (Getachew et al., 2010; Mirkena et al., 2011). The use of sheep milk has also been reported as important in southern Ethiopia (Legesse et al., 2008; Mekasha et al., 2016), and in South-Wollo (DBARC, 2011), in the central highlands of Ethiopia.

Over the years, there has been a fragmentation of land with less land per household, in the highlands and midlands of the country. In these situation farmers in the highlands seem to switch from cows to small ruminants, especially sheep, as they are easier to rear and have multipurpose roles (Abebe, 2012). Keeping sheep for milk production and promoting products developed from them will have advantages for smallholder farmers owning little land, and in food insecure areas. An increase in demand for milk and dairy products in rural and urban areas of Ethiopia is also observed (Mekasha et al., 2016); for direct consumption, for making butter, and to make the local drink 'hashara' (Getachew et al., 2010) by boiling sheep milk in water with roasted coffee hulls.

Due to the increasing demand for milk and milk products, there is an interest to increase milk production by genetic means. Genetically improved ewes would also improve the environment for the lambs resulting in higher pre-weaning growth (Ünal et al., 2007). Improved growth potential and subsequent survival of lambs also depends on the shape of the lactation curve of the ewe (Peniche et al., 2015).

To genetically improve production of sheep in Ethiopia a crossbreeding project has been implemented with various exotic meat and wool breeds, particularly Awassi. Awassi was imported from Israel and has been well accepted by producers. To date, most studies have focused on growth, wool and reproduction performances of native and crossbred sheep (Gizaw et al., 2007; Gizaw and Getachew, 2009; Getachew et al., 2013). However, when it comes to the milk production and the potential for genetic improvement through the current community-based sheep breeding programme (CBBP) in the central highlands of Ethiopia, there is a

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	Villages (Zone)	s (Zone)	
Study areas characteristics	Faji (North Shoa)	Chiro (South Wollo)	
Number of herds	16	18	
Number of ewes	55	60	
Distance from Addis-Ababa, km	120	501	
Altitude, m.a.s.l.ª	2770	1500–3700	
Latitude and longitude	10°00N-39°00 E	11°00N–39°00 E	
Rainfall, mm	920	700–1200	
Rainy season, pattern	June–September, bi-modal	June–September, bi-modal	
Temperature (annual), °C	14.4	13	

Table 1. Study area characteristics as well as number of herds and ewes observed in the two villages included in the study.

^am.a.s.l. = meters above sea level.

knowledge gap. Especially, there is a need for detailed information pertaining to the milk production potential of crosses with various levels of exotic blood. In developing countries like Ethiopia where limited resources are available for data recording, getting information is a big challenge. For traits like test day milk yield which is measured repeatedly, Legendre polynomials; a mathematical approach to model the average lactation curve; are widely used (Mrode, 2014; Schaeffer, 2016). Therefore, the objective of the present study was to estimate milk production performance and fit lactation curves of Awassi crosses relative to native sheep breeds reared under farmer's environment in the central highlands of Ethiopia based on registered testday milk yields using Legendre polynomials. Since gathering information under farmer's conditions is a challenge, efficient use of data is important, and we have presented an approach that allows comparisons of milk yield in different parts of the lactation given a limited number of ewes and observations per ewe.

Material and methods

Study area, genetic groups, and herd management

This study was carried out in two villages taking part in CBBP, in Faji (North Shoa zone) and Chiro (South-Wollo zone), of the Ethiopian central highlands (Table 1). Various genotypes involved in CBBP were included in this study. These were bred by smallholder farmers, locally organized as cooperative breeding groups. The local breeds were the Menz (http://eth.dagris.info/ node/2448) and Wollo sheep breeds that are indigenous to the selected study areas, classified as short fat-tailed, dual purpose breeds used for meat and wool and reared in the subalpine and cold highlands agro-ecological zones of Ethiopia (Gizaw et al., 2007).

Awassi is a fat-tailed meat and milk producing breed in common use around the Mediterranean Sea, particularly in Israel (http://afs.okstate.edu/breeds/sheep/ awassi/). In this study, the indigenous Menz ewes were considered as Local (0% Awassi) including the limited number of records of Wollo breed ewes. The Awassi crossbred ewes were categorized based on their Awassi blood percentage (<30% Awassi, 30-50% Awassi, >50% Awassi). Milk production from the various genotypes was measured on farm by trained local people and the first author. All animals were eartagged and housed in shaded open front barns. They were fed clover, straw and green fodder (maize and natural pasture) during the rainy seasons. Crop residues, hay, and often oat (Avena Sativa) straw and vetch (Vicia sativa) grass were commonly fed during the dry season. During crop harvesting, sheep had access to feed crop aftermath. Some farmers also gave supplementary feeds for the pregnant and nursing ewes.

Breeding rams

Breeding rams from three governmental farms: Debre-Berhan Agricultural Research Center (DBARC), Debre-Berhan Sheep Breeding and Multiplication Center (DB R1), and Amed-Guya Sheep Breeding and Multiplication Center (AG R2) had been allocated to local cooperatives. Each breeding ram was assigned a mating group of 20– 35 ewes. After three years of use, rams were culled and replaced with other rams from one of the governmental farms. Ewes were first mated at around 12 months of age. In both study areas, natural mating was practised throughout the year.

A local breeding cooperative is organized in groups of 6–12 (more in some cases) households based on neighbourhood and use of common grazing area. The Bureau of Agriculture and Rural Development (http://www.amhboard.gov.et/) is responsible for the dissemination of the selected Local x Awassi crossbred rams to villages. One 75% ($\frac{3}{4}$ Awassi × $\frac{1}{4}$ Menz) crossbred ram, and rarely one 50% ($\frac{1}{2}$ Awassi × $\frac{1}{2}$ Menz), is given for free to each group. The group of farmers is responsible for use and care of the breeding ram. Breeding rams are rotated both within the group, among groups of farmers, and

across the villages, to avoid mating between relatives (to minimize inbreeding) and to widen the gene pool (Gizaw and Getachew, 2009).

Data structure

Data used in this study were collected from Local (L) and all available Awassi x Local (AL) crossbred ewes kept under farmers' conditions for the production years 2015–2017 from the smallholder farms in the study areas. The ewes were at different ages. A total of 34 herds from the two villages were used for the study. A total of 466 records at different lactation stages or days in milk (DIM) were used from Local x Awassi crossbred ewes and Local breed ewes (Table 2).

Measuring milk yield

Milk measurement started from the 2nd (earlier for few) week after parturition. Most ewes were measured 4 times for milk yield during lactation. Lambs were separated from their mothers the evening before test day. In the morning, at least 12 h later, one half-udder was hand milked until it felt empty and the milk weighed. The other half udder was suckled by the lamb. The Weigh-Suckle-Weigh (WSW) method plus hand milking was used to estimate milk production as described by Benchohra et al. (2013). Test-day milk yield (TDMY) was then taken to be twice the sum of the hand milked yield and that consumed by lamb, following the methods suggested by ICAR (2002).

Statistical model and estimation

The analysis was performed mainly in two steps, first identification of the significant fixed effect for TDMY, Kg d⁻¹ was done using GLM procedure of SAS[®] and followed by estimation of variance components from Proc mix and finally in R programming to test significances of genetic groups for TDMY and fit lactation curves.

Important fixed effects for TDMY were identified, using the GLM procedure of SAS[®]. In addition to the identified fixed effects, data were analysed with fixed Legendre polynomials to model lactation curves nested

Table 2. Number ewes with records in each genetic group.

			Parity		
Genetic group	Number of ewe	Number of records	1	2	≥3
0% Awassi	46	196	39	66	94
<30% Awassi	19	85	11	32	43
30–50% Awassi	19	77	5	27	41
>50% Awassi	31	108	24	24	60
Total	115	466	79	149	238

within 4 genetic groups. Individual animals were included as a random effect. Regression coefficients for Legendre polynomials (up to order 3) were fitted as suggested by Schaeffer (2016) by use of mixed model and R (R Core Team, 2018) was chosen due to its ease of computing, data managing, and graphic display. Test days (*t*) with t_{min} (3rd day), the earliest test day, and t_{max} (147th day), the latest test day, were transformed to a normalized scale using $x = -1 + 2(t - t_{min})/(t_{max} - t_{min})$. The coefficients of the Legendre polynomial used were: d0 = 0.7071, d1 = 1.2247x, $d2 = -0.7906 + 2.3717x^2$, and $d3 = -2.8062 + 4.6771x^3$.

Predicted TDMY of observed ewes were fitted and tested for significance where graphs are above zero.

In matrix notation, the model was:

$$y = Xb + Zu + e$$

where \mathbf{y} is the vector of observations for daily milk yield in kg (TDMY); **b** is a vector of main fixed effects of: 2 villages (Faji, Chiro); 3 parities (first, second, later); 3 year-seasons of lambing (long rainy season, dry season, short rainy season); and 16 fixed regression coefficients for test days fitting lactation curves with the d0, d1, d2, d3 within the four genetic groups (i = 1, 2, 3, and 4): 0% Awassi (Local), <30% Awassi, 30-50% Awassi, and >50% Awassi; X is a design matrix assigning the fixed effects to the observations, including information on village, parity, year-season of lambing, genetic group, and transformed stage of lactation through the Legendre polynomial coefficients within genetic group; **u** is a vector of random effects of the 115 individual ewes (ID) included in the study, taken as independently distributed with same variance; Z is a matrix assigning the random effect of ewe (u) to its observations in y; and e is the vector of random independent residual effects.

The model assumptions were:

$$Cov(y, y') = V = ZGZ' + R,$$

Cov ($\boldsymbol{u}, \boldsymbol{u}'$) = **G** = $\sigma_{ID}^2 I$, where *I* is a 115*115 identity matrix, and σ_{ID}^2 is the variance component for ewes, and Cov ($\boldsymbol{e}, \boldsymbol{e'}$) = **R** = $\sigma_e^2 I$, where *I* is a 466*466 identity matrix.

The variance components (σ_{lD}^2 and σ_e^2) of **G** and **R** were estimated using the Proc Mixed procedure of SAS[®] with the model above. σ_{lD}^2 was estimated to be 0.08 kg², and σ_e^2 to be 0.04 kg². This approach was chosen due to the limited data available and incomplete pedigree of local ewes needed to run a random regression model. Given the estimated variance components, the **G**, **R**, and **V** were calculated as above and used in further calculations.

R software was used to estimate fixed effects of the model and carry out statistical testing. The b were

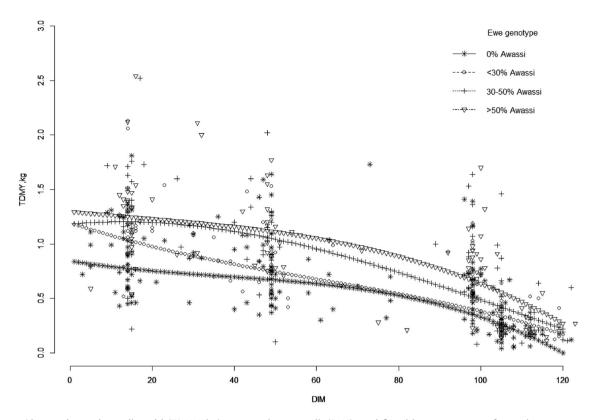


Figure 1. Observed test-day milk yield (TDMY, kg) against days in milk (DIM), and fitted lactation curves for each genetic group.

estimated with Generalized Least Squares means: $\hat{\boldsymbol{b}} = (\boldsymbol{X}' \boldsymbol{V}^{-1} \boldsymbol{X})^{-1} \boldsymbol{X}' \boldsymbol{V}^{-1} \boldsymbol{y}$, where $\hat{\boldsymbol{b}}$ is a 21*1 vector including 5 estimated fixed effects of: village, parity and yearseason of lambing, in addition to the 4*4 = 16 $\hat{\boldsymbol{b}}$ -s to establish the form of the lactation curves for the different genotypes for test day milk yield. Variance of this estimator is: **var** ($\hat{\boldsymbol{b}}$) = ($\boldsymbol{X}' \boldsymbol{V}^{-1} \boldsymbol{X}$)⁻¹.

Calculation of lactation curves and averages

For ewes of each genetic group i = 1, 2, 3, and 4, the leastsquares mean (LSM) yields for all lactation days t = 1, 2, ..., 120, making up the lactation curve was computed with: $\tilde{y}ii = Li \hat{b}$, where L*i* is a 120*21 matrix with d0, d1, d2, d3 for each of the 120 days in the genetic group *i*'s positions of the matrix **X**; and averaged over the main effects of village, parity, and year-season of lambing(i.e. the $\tilde{y}i$ is a vector with 120 estimated TDMY values for group *i*).

The LSM daily milk yield for an ewe in genetic group *i* over the 120 first days of lactation was calculated as follows:

$$\overline{\tilde{y}}i = \frac{1}{120} \sum_{t=1}^{120} \tilde{y}i(t) = k' Li \ \hat{\boldsymbol{b}}$$

where **k** is a vector with 120 equal elements: $\mathbf{k'} = \begin{bmatrix} \frac{1}{120}, \frac{1}{120}, \dots, \frac{1}{120} \end{bmatrix}$.

Comparison of daily TDMY and sub period yields of lactation

The three ranges of 5 test days (11–15, 46–50, 101–105 DIM; Figure 1.) with most observations were used for the calculation of LSM yields of three sub-periods of lactation per genetic group. These average yields for different lactation periods were calculated as follows:

$$\overline{\tilde{y}}i_{\text{early}} = \frac{1}{5} \sum_{t=11}^{15} \tilde{y}i(t) = \mathbf{k}_{\text{early}} Li \ \mathbf{\hat{b}}$$

with
$$\mathbf{k}_{early}$$
 being: $\mathbf{k}_{early}' = \begin{bmatrix} 0, 0, \dots, 0, \frac{1}{5}, \frac{1}{5}$

0, 0, ..., 0], with the $\frac{1}{5}$ – elements in position 11–15 of the vector with a total of 150 elements, the rest of

the elements being 0.

Similarly:

í

$$\overline{\tilde{y}}i_{mid} = \frac{1}{5} \sum_{t=46}^{50} \tilde{y}i(t) = k_{mid}'$$
 Li \hat{b}

and,

$$\bar{\tilde{\chi}}i_{\text{late}} = \frac{1}{5} \sum_{t=101}^{105} \tilde{y}i(t) = k_{\text{late}} \, Li \, \hat{b}$$

with \mathbf{k}_{mid} and \mathbf{k}_{late} defined according to range of test days.

		L	LSM contrasts \pm SE		
Genetic group	$LSM \pm SE$	< 30% Awassi	30–50% Awassi	>50% Awassi	
0% Awassi <30% Awassi 30–50% Awassi >50% Awassi	$\begin{array}{c} 0.56 \pm 0.08 \\ 0.67 \pm 0.09 \\ 0.87 \pm 0.07 \\ 0.96 \pm 0.07 \end{array}$	0.11 ± 0.07	$0.31 \pm 0.09^{*}$ 0.19 ± 0.10	$\begin{array}{c} 0.40 \pm 0.08^{*} \\ 0.28 \pm 0.09^{*} \\ 0.09 \pm 0.08 \end{array}$	

Table 3. Estimated least-square means (LSM) of test-day milk yield (kg) over 120 days in milk for 4 genetic groups and estimated contrasts between groups.

*p < 0.05.

Testing of differences between group of ewes

LSM differences between genetic groups (1 vs. 2, for example) of ewes over the whole lactation were found as:

$$L \hat{b}_{12} = \overline{\tilde{y}} 1 - \overline{\tilde{y}} 2 = \mathbf{k}' L 1 \, \mathbf{\hat{b}} - \mathbf{k}' L 2 \, \mathbf{\hat{b}} = \mathbf{k}' (\mathbf{L} 1 - \mathbf{L} 2) \mathbf{\hat{b}}$$

and correspondingly for selected sub-period and genetic groups (1 vs. 2 shown):

$$\overline{\tilde{y}}1_{\text{early}} - \overline{\tilde{y}}i2_{\text{early}} = \frac{1}{5}\sum_{t=11}^{15} \tilde{y}1(t) - \frac{1}{5}\sum_{t=11}^{15} \tilde{y}1(t)$$
$$= k'_{\text{early}}(L1 - L2)\hat{\boldsymbol{b}}$$

The variance of the differences between the average daily milk yield for genetic groups 1 and 2 in the first 150 days was calculated as:

$$\operatorname{var}(\overline{\tilde{y}}1 - \overline{\tilde{y}}2) = \operatorname{var}(L \ \hat{b}_{12}) = k'(L1 - L2) \operatorname{var}(\hat{b})(L1 - L2)' k$$
$$= SE_{12}^{2}$$

and similarly for other groups and time periods. SE is the standard error of the estimated difference. A 95% confidence interval for the difference was calculated using a t-distribution with the number of ewes as degrees of freedom:

$$L \hat{b}_{12} \pm 1.987 * \sqrt{SE_{12}}$$

Similar confidence intervals were calculated for all presented estimated differences, replacing SE_{12} with the relevant standard errors in each case. LSM differences between genetic groups are taken as non-significant (NS) at a 5% level if their confidence interval includes 0.

Results

Observed TDMY and fitted lactation curves for the genetic groups are shown in Figure 1. The LSM of testday milk yield from 120 days adjusted was 0.56, 0.67, 0.87, and 0.96 kg day⁻¹ for groups with 0% Awassi, <30% Awassi, 30–50% Awassi, and >50% Awassi, respectively (Table 3). The estimated contrasts between the four studied genetic groups over the entire lactation and their standard errors are given in Table 3. The groups >50% Awassi and 30–50% Awassi produced significantly (p < 0.05) more than the Local (0% Awassi) group, while there were no significant differences between Local and groups with < 30% Awassi ewes. Significant differences were also observed between <30% Awassi and >50% Awassi cross bred groups.

Contrasts between the genetic groups were also calculated in the periods with most observations (11–15, 46–50, and 101–105 DIM, Figure 1). At days 11–15, the estimated milk yield tended to increase with Awassi blood percentage of ewes (Table 4). The >50% Awassi group had LSM test-day milk yield of 1.25 kg day⁻¹, followed by the 30–50% Awassi groups of ewes. In this period, the 0% Awassi group (Local) produced significantly (p < 0.05) less test-day milk than the three studied genetic groups of ewes (<30%, 30–50%, and >50% Awassi), while no significant differences were found between groups with <30%, 30–50% and >50% Awassi.

At 46–50 days after lambing, the LSM of groups with 30–50% Awassi and >50% Awassi produced significantly

Table 4. Estimated least-square means (LSM) and standard errors (\pm SE) of test-day milk yield (kg) over 3 sub-periods of lactation (DIM)for 4 genetic groups and estimated contrasts between groups per period.

Genetic group		LSM contrasts 11–15 DIM \pm SE		
	$LSM \pm SE$	<30% Awassi	30–50% Awassi	>50% Awassi
0% Awassi	0.78 ± 0.09	$0.27 \pm 0.10^{*}$	0.43 ± 0.11*	0.48 ± 0.11*
<30% Awassi	1.05 ± 0.11		0.16 ± 0.12	0.21 ± 0.12
30–50% Awassi	1.20 ± 0.08			0.05 ± 0.11
>50% Awassi	1.25 ± 0.09			
			LSM contrasts 46–50 DIM ± SE	
0% Awassi	0.67 ± 0.09	0.08 ± 0.09	$0.38 \pm 0.10^{*}$	0.45 ± 0.11*
<30% Awassi	0.76 ± 0.11		$0.30 \pm 0.12^{*}$	$0.36 \pm 0.12^{*}$
30–50% Awassi	1.06 ± 0.08			0.06 ± 0.11
>50% Awassi	1.12 ± 0.09			
			LSM contrasts 101–105 DIM ± SE	
0% Awassi	0.29 ± 0.09	0.07 ± 0.08	0.16 ± 0.10	0.30 ± 0.09*
<30% Awassi	0.35 ± 0.11		0.09 ± 0.11	$0.23 \pm 0.10^{*}$
30–50% Awassi	0.45 ± 0.08			0.14 ± 0.09
>50% Awassi	0.59 ± 0.09			

*p < 0.05.

(p < 0.05) more than the two groups with either 0% Awassi or < 30% Awassi (Table 3). Likewise, after mid-lactation (101–105 DIM) only the higher Awassi% crossbred ewe group (>50%) showed significant differences from local and <30% Awassi (Table 3). The groups of ewes with >50% Awassi showed the highest TDMY in this period (0.96 kg day⁻¹). The average TDMY mainly decreased from the first to the last selected sub-periods of lactation. Overall, the group of ewes with >50% Awassi showed the highest average test-day milk yield in all three periods. However, there was no significant increase observed with the increase of Awassi blood level (%) neither for the entire nor in the selected days of sub-periods of lactation.

Discussion

In the present study, milk yields of groups of ewes of Menz and Wollo (Local) were compared with crossbred ewes having a variable percentage of Awassi. Using a limited number of ewes (n = 115) and records (n = 466)recorded over the entire lactation under field conditions in Ethiopia, the approach with modelling of lactation curves by genetic group had power enough to give some clear recommendations as to what breed percentages to be given preference with respect to milk yield. The group of ewes with a percentage of >50% Awassi produced consistently more milk than the Local breeds 0.40 kg day⁻¹ over the entire calculated 120 days of lactation, or 70% more. The 30-50% Awassi group produced 0.31 kg day⁻¹ (55% more) over the Local ewes. However, the >50% Awassi group did not improve significantly over the entire as well as sub periods of lactation over the 30-50% Awassi group. Considering the current management system at farmers' level, 30-50% Awassiewes suits best. If improved management can be provided, increasing Awassi percentage could be better for milk production.

The milk production of the Local group (0.56 kg day⁻¹) was more than double of that reported for the Afar breed (0.224 kg day⁻¹) in Ethiopia (Mirkena et al., 2011) and higher than what Mekoya et al. (2009) reported for Menz sheep breed (0.21 kg day⁻¹).

Use of Legendre polynomials within breed group allowed to utilize data for animals from different villages, parities and time of lactation period. Modelling of the contemporary group effect through village was chosen because each household had only a few ewes each (ranged from 1–30). A clear peak in the curve for most groups, expected to happen around 3–4 weeks after lambing (Assan, 2015) is lacking. Such a peak was only visible for the 30–50% Awassi group. These patterns made it difficult to compare the groups for their persistency, but also to compare them on average lactation yield (by integrating daily yield under the curve over the lactation). Comparison of lactation yield was done at 120 days, while the higher percentage Awassi groups milked longer. To become less dependent on the trajectory in comparisons, we chose to compare at lactation time points with most data.

To get Local x Awassi crossbreed ewes with 30–50% blood level in the field, it is necessary to disseminate breeding rams with a variable blood level of Awassi (25–75%). Local ewes (0% Awassi) could well be mated with rams with 75% Awassi initially. Thereafter, ewes with intermediate Awassi percentages could be mated to rams with 50% or 25% Awassi, or some intermediate percentage. The 50%Awassi rams would be the easiest to produce at the present stage. If a selection scheme for rams of a synthetic breed combining Local and Awassi is initiated, based on daughters' performances and BLUP, or based on genomic selection, proven individual rams of this type could be distributed.

Conclusions

With limited number of ewes and records from the field, modelling of the lactation curve within genetic groups can be used to draw some inference as to what breed percentage ewes should be given preference. The best performing ewes produced consistently more than local breeds over the course of the lactation, amounting to an average production improvement of close to 70% over Local ewes. This study shows the potential that exists for increasing milk production of ewes through the initiated crossbreeding programme with sheep in Ethiopia. A future evaluation could also rely on other traits than milk, like lamb survival, or udder morphometry to reach a more definite conclusion including large set of data. Further detailed economic analyses could also be required.

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Disclosure statement

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