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Effect of the Lactation Months on Milk Composition of the Second-Parity Lacaune Ewes

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Abstract. This research aims to determine the chemical composition of Lacaune ewe milk produced in Estonia, as well as the correlations and the influence the stages of lactation have on milk. The study was carried out on fifty-one second-parity ewes. The analysis involved a total of 178 milk samples collected monthly from the second to the seventh month of lactation. Milk analyses included the determination of the contents of total solids, fat, total protein, casein, casein index, lactose, ash, P, Ca, K, Na, Mg, somatic cell count and pH. The Lacaune ewes' milk contained on average 18.62% total solids, 7.75% fat, 5.74% total protein, 4.32% casein, 4.76% lactose, 0.89% ash, 160.26 mg 100 g^{-1} Ca, 140.07 mg 100 g^{-1} P, 135.21 mg 100 g^{-1} K, 46.44 mg 100 g^{-1} Na, 17.66 mg 100 g^{-1} Mg. Overall means for casein index, pH value and somatic cell scores were 75.35%, 6.61 and 12.62, respectively. It was found that the month of lactation significantly affected almost all monitored traits except somatic cell score, casein index and Ca content. The contents of total solids, fat, total protein, casein, ash, P, and Mg increased, while the lactose content, and pH value decreased with the advancing lactation. Sodium content was highest and potassium content lowest value during mid-lactation. Producers must take into account that the composition of Lacaune ewe milk depends on the stage of lactation and may, therefore, affect the production process and the final quality of the product.

Key words: ewe milk, chemical composition, stage of lactation.

INTRODUCTION

The Lacaune breed has become one of the world's high-yielding ovine milk breeds, with average daily milk yields of 1.75 L (Giambra et al., 2014; Pesantez-Pacheco et al., 2018). Since dairy sheep farming is not traditional in Estonia, ewe milk products are a niche market, which is mainly influenced by the growth of consumer interest in different dairy products. Ewe milk is considered a delicacy in many countries, and ewe dairy products have gained popularity among consumers due to the quality and nutritional

value of the products (Balthazar et al., 2017). Compared to the milk of other domestic mammals, high nutritional value of ewe milk is contributed to the higher concentrations of proteins, fats, vitamins, and minerals (Park et al., 2007; Barłowska et al., 2011; Balthazar et al., 2017). Ewe milk is mainly processed into traditional fine cheeses for gourmet and export markets, as well as yoghurt and ricotta (Milani & Wendorff, 2011; Pulina et al., 2017). The high levels of protein, fat, and calcium are important in cheese-making (Moatsou et al., 2004). The same reasons make ewe milk very suitable for yoghurt making. Ewe milk yoghurt possesses high gel strength and minimal syneresis, it can be produced without the need for added milk solids or stabilizers (Milani & Wendorff, 2011).

The composition of ewe milk is mainly influenced by breed, nutrition, environment and stage of lactation (Pulina et al., 2005; Park et al., 2007; Komprej et al., 2012; Inostroza et al., 2020). The stage of lactation has a significant effect on daily milk yield, pH value, somatic cell count and the content of total solids, fat, protein, casein, lactose, Ca and P (Kuchtík et al., 2017). However, studies have shown mixed results in examining the relationship between milk composition and lactation progression. There is no information on the composition and properties of ewe milk produced in Estonia. The wider purpose of this pilot study was to give an overview of the composition of Lacaune ewes' milk produced in local conditions to encourage potential farmers to take up dairy sheep farming.

The composition and the physico-chemical properties of milk vary depending on production conditions and the individual characteristics of particular animals. Fluctuations in the composition and quality of milk are also reflected in the quality of products. This research aims to identify changes in the composition and properties of milk during the lactation period in Estonia. The outcome of this work would benefit the production and valorisation efficiency of ewe milk.

MATERIALS AND METHODS

Animals

The Lacaune dairy sheep breed was introduced in Estonia in 2017, when 84 purebred Lacaune Lait sheep were imported from France. A semi-intensive system was implemented on the family-operated farm (with year-round free-range housing in modern buildings and seasonal milk production). During the sample collecting period, all ewes were kept in one flock under identical conditions under permanent veterinary supervision. The ewes did not show any signs of serious health issues, although some minor health issues were observed during the sample period and it was necessary to exclude some ewes from the study and include others from the same study group.

Lambing took place from January to March 2019. During the lactation, the ration was constant, ewes were fed twice a day with 1.1 kg of concentrate per day and *ad libitum* first cut mixed hay. The quantity and composition of the ration fed to the animals was the same throughout the sampling period. The concentrate contained 62.5% of full grain oat, 35.7% of rape seed cake, and 1.8% of vitamins and minerals. The DM content of the feed was 88%, that of crude protein 15% and 10.92 MJ kg^{-1} of metabolizable energy (ME). Hay contained 85% of dry matter, 10% of crude protein and 4.5% of minerals.

Ewes were milked twice a day at 7:00 and 17:00 using a Panazoo 1×12 parallel milking parlour (-39 kPa; 180 pulsation min^{-1}). Average milk yield was 1.8 kg per day in the first half of lactation and 0.8 kg per day in the second half of lactation, measured at the bulk milk tank. Most of the lactating ewes dried off at the end of August.

Milk sampling

Fifty one second-parity Lacaune Lait ewes, which were divided into two 25−26 ewes groups based on lambing time, participated in the study. Milk samples were collected monthly from 15 ewes within the group, from 30 ewes in total. Milk sampling started from 30 days postpartum and then monthly during the lactation period (from February to August). Milk samples were collected only at the morning milking. The total number of samples collected was 178. Individual milk samples (volume at least 250 mL) were immediately transported to the laboratory in a cooler at 4 °C. Milk sub-samples to determine fat, total protein (TP), lactose contents and somatic cell count (SCC) were preserved with Broad Spectrum Microtabs® II (Bronopol < 44%, Natamycin < 2%). Milk sub-samples for the determination of total solids (TS), casein, ash, Ca, P, K, Na and Mg contents were frozen at –20 °C using 15 and 50 mL polypropylene centrifuge tubs.

Compositional analysis

Milk fat (%), TP (%), lactose (%) and SCC ($\times 10^3$ mL^{-1}) were estimated in the Milk Analysis Laboratory of the Estonian Livestock Performance Recording Ltd with Analysers (CombiFOSS, Denmark) using the international standard IDF 141; EVS-EN ISO 13366-2 and work package PL-PR-2.

TS, casein, ash and pH were estimated in the Laboratory of the Chair of Food Science and Technology of the Estonian University of Life Sciences. TS (%) content was determined gravimetrically by oven drying at 102°C to constant weight according to the standard ISO 6731:2010. Casein (%) content was analysed by the Kjeldahl method (ISO 17997-2:2004/IDF 29-2:2004) using the wet ashing device Digestor™ 2508 and analyser Kjeltec™ 2300 (FOSS, Denmark). Ash (%) content was determined gravimetrically by incineration in a muffle furnace at 550°C (AOAC 945.46). Contents of calcium (Ca, mg 100 g^{-1}), potassium (K, mg 100 g^{-1}), sodium (Na, mg 100 g^{-1}) and magnesium (Mg, mg 100 g^{-1}) were measured according to the atomic absorption spectrometric method (ISO 8070/IDF 119:2007) by using the ContrAA® 700 device (Analytik Jena AG, Germany). The phosphorus (P, mg 100 g^{-1}) content was determined according to the standard ISO 9874:2006/IDF 42:2006 using the spectrometer Specord® 250 Plus (Analytik Jena AG, Germany). Active acidity (pH) was measured with the pH-meter SevenCompact™ S210 equipped with the electrode InLab® Expert Pro (Mettler Toledo, U.S.A).

The casein index was calculated according to the following formula (Buccioni et al., 2015):

$$Casein\ index\ \% = \left(\frac{Total\ casein\ content}{Total\ crude\ protein\ content}\right) \times 100 \quad (1)$$

To achieve a normal distribution, the somatic cell counts (SCC) were converted to a somatic cell score (SCS) using a log base 2 function (Ali & Shook, 1980):

$$SCS = \log_2\left(\frac{SCC}{100}\right) + 3 \quad (2)$$

Statistical analyses

Statistical analyses were performed with the statistical package R 4.0.3 (R Core Team 2021). Effect of lactation month and random effect of animals to the milk fat, TP, casein, casein index, SCS, lactose, pH, TS, ash and mineral content were studied by Linear Mixed-Effects Model (GLMM). Emmeans and multcomp packages were used to carry out pairwise comparison of the groups. Tukey's multiple comparison post-hoc test was used to determine the groups least square mean differences at the significance level of $\alpha = 0.05$. Relationships between variables and their statistical significances were calculated by rcorr function in Hmisc package (Harrell & Dupont, 2021) and reported as Pearson correlation coefficients. Significance probability levels were denoted as: * - $P < 0.05$, ** - $P < 0.01$ and *** - $P < 0.001$. In addition, principal component analysis (PCA) was performed with the procedure fviz pca in factoextra package (Kassambara & Mundt, 2020) using ewe milk composition records (fat, TP, lactose, SCS, TS, pH, ash, casein, Ca, P, K, Na and Mg) as variables and lactation month as factor.

RESULTS AND DISCUSSION

Milk composition

The ewe milk had higher contents of TS, fat, TP, casein, lactose and ash in comparison with both goat and cow milk (Park et al., 2007; Balthazar et al. 2017). This study showed that milk samples from Lacaune ewes contained on average 18.62% total solids, 7.75% fat, 5.74% total protein and 4.76% lactose (Table 1). Considerably higher contents of total solids (19.34%) and fat (8.10%) for the same breed, and distinctly lower protein (5.22%) and lactose (4.43%) contents have been found from bulk tank milk in a Brazilian study (Fava et al., 2014). Similar total solid content (18.61%) was reported in a study conducted in the Czech Republic on Lacaune ewes (individual samples, $n = 18$) at 127 days of lactation (Kuchtík et al., 2017) and in a mixture of milk (18.7%) from different breeds of sheep (Hanuš et al., 2015). The average casein and ash contents in our study were 4.32% and 0.89%, respectively (Table 1). Similar results have been found for the same breed by Kaminarides & Anifantakis (2004) and Kuchtík et al. (2017) and for the Sarda breed by Bittante et al. (2017). The mean casein index in our study was quite low, 75.35%. Commonly the casein portion of ewe milk amounts to around 80% of the total milk protein (Hejtmánková et al., 2012; Balthazar et al., 2017). The average Ca content (160.26 mg 100 g^{-1}) of Lacaune milk in this study was significantly lower (Table 1) than in some published results (Kaminarides & Anifantakis, 2004; Panayotov et al., 2018) and somewhat higher than in a Czech Republic study (Kuchtík et al., 2017). The average P content (140.07 mg 100 g^{-1}) in the current study was higher than the data reported in earlier research (Kuchtík et al., 2017; Panayotov et al., 2018). A similar trend was also found for the K content by Panayotov et al. (2018). The average K content of Lacaune milk in this study (135.21 mg 100 g^{-1}) was higher than in the milk of other ewe breeds (Park et al., 2007; Balthazar et al., 2017). The average Na and Mg contents in this study (46.44 mg 100 g^{-1} and 17.66 mg 100 g^{-1}, respectively) confirm the results from earlier research with the Lacaune (Kaminarides & Anifantakis 2004; Panayotov et al., 2018) and other ewe breeds' milk (Park et al., 2007; Balthazar et al., 2017; Aldalur et al., 2019).

Of the basic components, the fat content varied the most (CV, 19.72%), and the ash content the least over the lactation period (CV, 6.47%) (Table 1). The lactose and the

casein index (CV, 7.53% and CV, 6.67% respectively) were also less variable. The variation in minerals contents was greater for Na (CV, 29.92%), K (CV, 24.89%) and Mg (CV, 21.23%) contents than for other analysed minerals. Corresponding well to the findings from earlier studies on Lacaune ewes' milk (Kaminarides & Anifantakis, 2004; Kuchtík et al., 2017; Panayotov et al., 2018), the average pH value in the present study was 6.61 and was most stable (CV, 1.72%). SCS had the highest variation (CV, 24.24%). The average number of SCS (12.62) is close to that of a Spanish study (Rovai et al., 2015) and is slightly higher than in some others studies of Lacaune milk (Barillet et al., 2001; Kuchtík et al., 2017). High SCC could refer to udder infection or some content of colostrum in the first month milk sampling. Barillet et al. (2001) and Kuchtík et al. (2017) reported that SCC can be caused by clinical, chronic or subclinical mastitis or could be linked to a reduced milk yield.

Table 1. Descriptive statistics of milk chemical composition and the coefficients of variation (CV) over the lactation period (n = 178)

Milk characteristics	Mean	SD	Minimum	Maximum	CV (%)
Fat (%)	7.75	1.53	4.23	14.75	19.72
TP (%)	5.74	0.86	4.18	8.65	15.00
Casein (%)	4.32	0.72	2.81	6.96	16.72
Casein index[1] (%)	75.35	5.03	52.32	86.53	6.67
SCS	12.62	3.06	7.32	20.89	24.24
Lactose (%)	4.76	0.36	3.42	5.44	7.53
pH	6.61	0.11	6.34	6.98	1.72
TS (%)	18.62	2.04	14.80	27.73	10.93
Ash (%)	0.89	0.06	0.75	1.11	6.47
P (mg 100 g^{-1})	140.07	19.92	80.65	206.13	14.22
Ca (mg 100 g^{-1})	160.26	18.09	118.00	229.90	11.29
K (mg 100 g^{-1})	135.21	33.65	80.50	195.90	24.89
Na (mg 100 g^{-1})	46.44	13.89	19.30	78.20	29.92
Mg (mg 100 g^{-1})	17.66	3.75	8.40	27.30	21.23

[1]Casein index: casein to protein ratio.

In the present study, the strongest and most statistically significant correlation was found between TP and casein (r = 0.91; P < 0.001) (Table 2). The TS content had a similar relationship with fat (r = 0.846; P < 0.001), TP (r = 0.846; P < 0.001) and casein (r = 0.833; P < 0.001) contents. Similar results were obtained in a study of Araucana creole ewe´s milk (Inostroza et al., 2020). In our study, the fat content had a strong positive correlation with TP (r = 0.766; P < 0.001) and casein (r = 0.757; P < 0.001). Additionally, all correlations between lactose and TS, fat, TP, casein and ash content were negative (P < 0.001), which is consistent with another study of Lacaune (Kuchtík et al., 2017) and Araucana creole ewe´s milk (Inostroza et al., 2020).

The pH value correlated positively with SCS (r = 0.429; P < 0.001) and negatively with casein and TS (r = –0.422; P < 0.001, r = –0.392; P < 0.001 respectively). This can be explained by the health condition of the udder. In case of mastitis, with an increased number of the SCC, changes in the production of milk in the mammary gland occur, e.g., a decrease in the casein to TP ratio also causes a fall in the acidity of milk. Similar relationship between the SCC and pH value has been found in ewe milk in Italy (Albenzio et al., 2004) and in Sarda ewes' milk (Paschino et al., 2019).

Table 2. Pearson correlation coefficients between milk parameters of Lacaune ewes

	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.
1. Lactation month	1													
2. Fat	0.518 ***	1												
3. TP	0.725 ***	0.766 ***	1											
4. Casein	0.672 ***	0.757 ***	0.910 ***	1										
5. Casein index	0.042	0.141	0.001	0.409 ***	1									
6. SCS	-0.054	-0.055	-0.081	-0.192 *	-0.271 ***	1								
7. Lactose	-0.643 ***	-0.592 ***	-0.678 ***	-0.583 ***	0.069	-0.332 ***	1							
8. pH	-0.399 ***	-0.284 ***	-0.357 ***	-0.422 ***	-0.239 **	0.429 ***	0	1						
9. TS	0.527 ***	0.846 ***	0.846 ***	0.833 ***	0.13	-0.243 **	-0.429 ***	-0.392 ***	1					
10. Ash	0.224 **	0.359 ***	0.456 ***	0.326 ***	-0.207 **	0.197 **	-0.447 ***	0.151 *	0.267 ***	1				
11. P	0.255 ***	0.371 ***	0.468 ***	0.443 ***	0.033	0.007	-0.197 **	-0.306 ***	0.440 ***	0.479 ***	1			
12. Ca	0.151*	0.319 ***	0.371 ***	0.305 ***	-0.074	-0.151 *	-0.033	-0.136	0.371 ***	0.534 ***	0.537 ***	1		
13. K	-0.329 ***	-0.08	-0.282 ***	-0.247 ***	0.023	0.306 ***	0.032	0.223 **	-0.199 **	-0.08	-0.013	-0.04	1	
14. Na	0.569 ***	0.194 **	0.452 ***	0.358 ***	-0.119	0.055	-0.483 ***	-0.133	0.215 **	0.331 ***	0.141	0.015	-0.677 ***	1
15. Mg	0.770 ***	0.496 ***	0.690 ***	0.621 ***	0.003	-0.009	-0.496 ***	-0.405 ***	0.524 ***	0.345 ***	0.330 ***	0.197 **	-0.462 ***	0.637 ***

Significance probability levels are denoted as: * - $P < 0.05$, ** - $P < 0.01$ and *** - $P < 0.001$.

Correlations between the mineral content and major composition parameters of the milk were mainly moderate, but statistically significant. Higher contents of TS, fat and casein and lower contents of lactose were associated with higher contents of the analysed minerals except for K, which was lower in this case. The milk samples' major components had the strongest correlations with Mg content (Table 2). The Mg content was positively correlated with TS ($r = 0.524$; $P < 0.001$), fat ($r = 0.496$; $P < 0.001$), TP ($r = 0.690$; $P < 0.001$), casein ($r = 0.621$; $P < 0.001$), ash ($r = 0.345$; $P < 0.001$), P ($r = 0.330$; $P < 0.001$) and Na ($r = 0.637$; $P < 0.001$) contents and negatively with lactose ($r = –0.496$; $P < 0.001$), pH value ($r = –0.405$; $P < 0.001$) and K ($r = –0.462$; $P < 0.001$).

Effect of Lactation Stage

The study confirmed that the stage of lactation has a significant effect on the contents of all of the recorded components of milk, except for SCS, casein index and calcium content (Tables 2 and 3). The contents of TS, fat, TP and casein in milk increased over the lactation ($r = 0.527$; $P < 0.001$, $r = 0.518$; $P < 0.001$, $r = 0.725$; $P < 0.001$ and $r = 0.672$; $P < 0.001$ respectively), which is comparable to reports on different breeds (Othmane et al., 2002) and the Lacaune breed (Kuchtík et al., 2017), but these indicators in this study were higher than those reported for Bovec, Istrian Pramenka and Improved Bovec breeds (Komprej et al., 2012). In contrast to the TS, fat, TP and casein contents, lactose content decreased significantly during lactation in the current research ($r = –0.643$; $P < 0.001$). The same finding has been observed in studies in the Czech Republic (Kuchtík et al., 2017) and Greece (Termatzidou et al., 2020). The lactose contents in the milk of healthy animals remained fairly constant, while the decrease in the lactose content is may be attributed to udder health issues (mastitis). This suggestion is supported by the negative correlation found between lactose and SCS ($r = –0.332$; $P < 0.001$) (Table 2). However, at the sixth month of lactation the lactose content decreased while the SCS increased (Table 3). The month of lactation had a slight ($r = 0.224$), but statistically significant ($P < 0.01$) effect on the ash content, which was higher at the end of lactation. Differences in the ash content can be explained by the reduction in the milk yield over the course of lactation. Robles Jimenez et al. (2020) showed that maximum milk yield is achieved on the second month of lactation, after which the yield begins to decrease. However, the variation in the mineral contents of ewe milk depends also on several others factors such as breed, geographical location, diet, stage of lactation, parity, and farming practices (Chia et al., 2017).

The month of lactation had no significant effect on SCS ($P = 0.471$), which is in line with the findings by Kuchtík et al. (2017), where SCS in Lacaune ewe milk insignificantly increased as lactation advanced. On the other hand, Matutinovic et al. (2011) reported a significant increase in SCS during lactation in the milk from an indigenous breed in sub-Mediterranean area. In our study, the mean SCS was lowest in the fifth month of lactation, differing significantly from the first two and the sixth month of lactation ($P < 0.05$).

The pH value of ewe milk decreased ($P < 0.001$) consistently during lactation (Table 3). The same result was found by Pavić et al. (2002) and Sahan et al. (2005). It might be explained by an increase in protein content (Pellegrini et al., 1997), and especially with an increase in casein ($r = −0.422$) and P ($r = −0.306$) content during lactation. Changes in the concentration of salts and proteins, mainly the caseins in the

milk also affect the acidity level (Merlin Junior et al., 2015). Kuchtík et al. (2017) reported very stable pH values in the milk of Lacaune ewes over the lactation, and the stage of lactation had no significant effect on the pH value.

Table 3. Least square means and coefficients of variations (CV) of Lacaune ewe milk parameters in different months of lactation

		Month of lactation					
		2	3	4	5	6	7
	n	29	30	30	30	30	29
Fat (%)	Mean	7.10ab	6.91a	7.8b	7.43ab	8.89c	9.13c
	CV%	14.48	17.49	14.26	13.11	20.61	15.33
TP (%)	Mean	4.9a	5.19a	5.68b	5.79b	6.4c	6.65c
	CV%	7.05	10.19	9.26	7.01	12.27	12.53
Casein (%)	Mean	3.72a	3.86a	4.2b	4.32b	4.93c	4.97c
	CV%	10.86	9.83	9.89	8.38	14.48	15.45
Casein index[1] (%)	Mean	75.71a	74.97a	74.11a	74.72a	77.35a	74.87a
	CV%	6.92	8.75	5.41	6.95	4.51	6.83
SCS	Mean	13.61a	13.16a	11.96ab	11.37b	13.07a	12.81ab
	CV%	13.45	18.47	31.52	29.27	26.14	21.55
Lactose (%)	Mean	4.96a	4.96a	4.91a	4.76b	4.48c	4.36c
	CV%	3.54	3.92	5.55	4.17	9.09	6.42
pH	Mean	6.7a	6.64b	6.62bc	6.58cd	6.56d	6.57cd
	CV%	1.37	1.19	1.80	1.30	1.84	1.80
TS (%)	Mean	17.32a	17.54ab	18.84cd	18.46bc	19.76de	20.40e
	CV%	6.85	7.95	6.06	6.40	13.21	11.23
Ash (%)	Mean	0.87a	0.89ab	0.9ab	0.89ab	0.9b	0.92b
	CV%	5.80	4.85	5.46	6.02	7.49	7.89
P (mg 100 g^{-1})	Mean	132.87a	137.94ab	136.31ab	141.28ab	145.14ab	148.6b
	CV%	22.64	13.92	10.67	9.94	12.23	11.67
Ca (mg 100 g^{-1})	Mean	156.55a	156.79a	157.46a	164.27a	164.13a	163.71a
	CV%	9.80	13.04	9.27	8.74	14.30	11.16
K (mg 100 g^{-1})	Mean	179.08a	149.19b	109.74cd	102.25c	120.62d	152.41b
	CV%	3.60	21.10	9.55	8.98	26.63	9.84
Na (mg 100 g^{-1})	Mean	26.15a	40.49b	52.85cd	56.58c	51.07cd	50.44d
	CV%	18.29	26.50	18.22	13.77	23.11	17.09
Mg (mg 100 g^{-1})	Mean	11.53a	16.36b	18.14c	18.79c	20.27d	20.89d
	CV%	14.29	17.35	9.04	10.46	11.21	9.73

[1]Casein index: casein to protein ratio.

Different superscripts indicate significant differences ($P < 0.05$, Sidak multiple comparisons post-hoc test).

Only a weak correlation was found between the month of lactation and the Ca content ($r = 0.151$; $P = 0.044$) (Table 2, 3). A stable level of Ca over the lactation period has also been found by Sevi et al. (2004), but a constantly increasing Ca content in ewe milk over the lactation has been reported in studies of Lacaune ewes by Kuchtík et al. (2017) and by Abilleira et al. (2010) for the Latxa breed's milk. These different results may have been obtained due to different feeds, parity or climate. Kuchtík et al. (2017) reported that increase in the content of Ca and P during lactation were mainly affected by the gradual increase of TP and casein contents, which were also found in our study. The P content increased slightly during lactation; the differences between the first and

last months of lactation were significant ($P < 0.05$; Table 3). This is consistent with the trends reported by Kuchtík et al. (2017) and Panayotov et al. (2018) for the milk of Lacaune and Awassi breed ewes (Sahan et al., 2005). Contrary results have been found in ewe milk of the Dorset breed, where a decrease in P content during lactation was recorded (Wohlt et al., 1981).

A significant ($P < 0.001$) positive correlation was found between the lactation month and both Mg and Na contents ($r = 0.770$ and $r = 0.569$ respectively) and a negative correlation with K ($r = –0.329$), whilst Sahan et al. (2005) found significant changes only in the Na content and no significant changes in either the K or Mg contents during lactation. The mean Na content of ewe milk was higher in mid-lactation milk (Table 3). As the Na content in the milk increased, there was a decrease in the K content. A similar trend was observed elsewhere (Wohlt et al., 1981; Sahan et al., 2005). The highest average Na content and the lowest average K content were found in the fifth months of lactation.

A PCA assessment was carried out for all of the evaluated lactation months and 13 variables, which qualify milk composition. Individual traits showed high variability in the composition of milk according to the month of lactation. However, when individual components were averaged and visualized in a bi-plot (Fig. 1), lactation months can be differentiated according to the concentration of milk components.

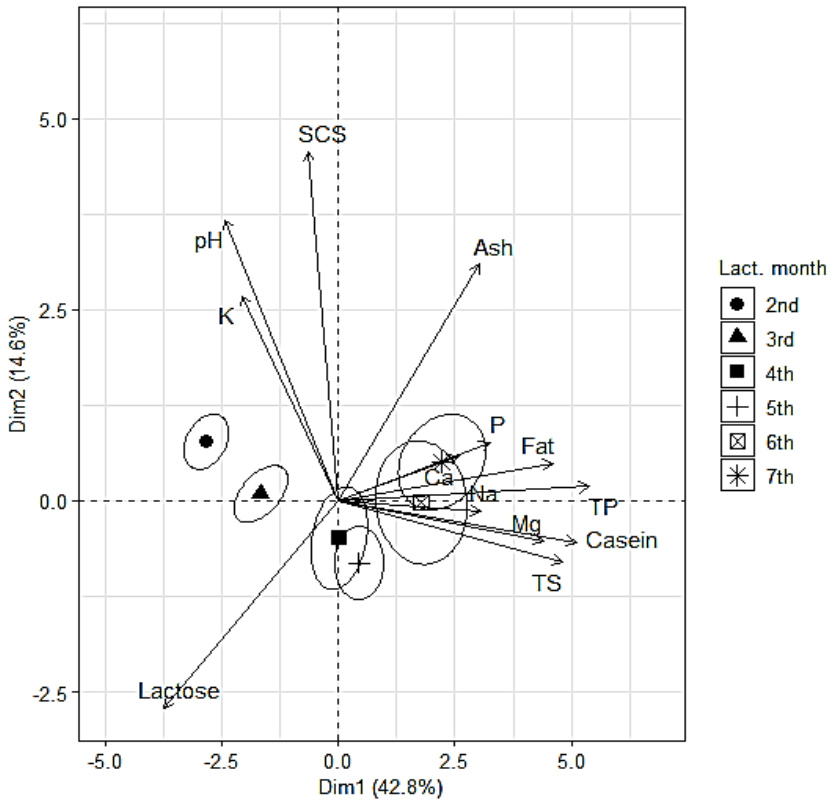

Figure 1. Bi-plot of principal component analysis with month of lactation (2nd–7th) and milk components in Lacaune ewes (TP – total protein; TS – total solids; SCS – somatic cell score; Dim 1 and 2 – principal components).

As the result of the PCA, six evaluated lactation months showed that 42.8% of the total variability was attributed to the first principal component (PC). The variables related to the mineral content of the milk (except K) as well as fat, casein and TP all contribute to PC1 with higher values. The PC2 aggregated 14.6% of the total variance and was related to SCS and also to pH and K. Together these two PC-s can explain over half (57.4%) of the variance.

From the location of the lactation months in Fig. 1, it can be concluded that some lactation months differ from the others, especially the second and third ones. The bi-plot reveals that the fat, casein, TP and mineral content of the milk (except K) increases throughout the lactation months. This can be explained by the changes in the milk yield within the lactation period. The milk yield was not recorded currently, but numerous studies have shown that the lactation curve of ewes is highest at the beginning of lactation, and then starts to decrease steadily (Ruiz et al., 2000; Oravcová et al., 2006; Komprej et al., 2012; Elvira et al., 2013; Inostroza et al., 2020; Robles Jimenez et al. 2020). However, according to the PC2, higher milk pH value, SCS and K were registered on the second month of lactation. In the comparison of the lactation months, the PCA showed that milk composition is quite similar on the fourth and fifth lactation months and as well as on the sixth and seventh months.

CONCLUSIONS

Lacaune ewes' milk in Estonia contained high levels of total solids, including fat, protein, casein and minerals (ash). The most variable milk traits were Na, K, SCS, Mg and fat contents and the least variable were pH value, ash and lactose contents and the casein index. The lactation stage significantly affected almost all of the measured milk traits, except for the casein index, SCS and Ca content. TS, fat, TP, casein, ash, P, and Mg contents increased, while the lactose content, and pH values decreased with advancing lactation. The mean values for K were higher and for Na lower at the beginning and at the end of lactation. Principal component analysis showed that there were small differences in the milk compositional characteristics between the fourth and fifth and between the sixth and seventh months of lactation.

Higher TS content, including the casein content, requires a higher yield of the cheese and an improvement in the structural properties of the fermented beverages in the second half of the lactation period. However, the content of Ca and P, which affects the coagulation properties of milk, remained stable or increased slightly within the lactation months. Therefore, an analysis of the coagulation properties of ewe milk during lactation would be necessary. Our results suggest that it would be possible to differentiate milk according to the lactation stage to harvest milk with key-traits for certain products.

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