

Chemical composition, physical traits and fatty acid profile of goat milk as related to the stage of lactation

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The chemical composition and physical traits (selected) of milk, and fatty acid profile of milk fat were determined throughout the progress of lactation II in 20 Polish White Improved goats. From October to May goats were fed corn silage, hay and carrot *ad lib.*, concentrate according to their milk yield, with mineral and vitamin premix, chalk and NaCl. From June to September hay was substituted by fresh grass. The diets were balanced according to INRA feeding standards. Milk samples were taken from each goat on lactation day 60, 120 and 200 (lactation stage 1, 2 and 3, respectively). With the progress of lactation highly favourable changes took place in the most desirable monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids content of a total of fatty acids. The coefficients of the calculated Pearson's correlation showed that the concentration of MUFA and PUFA (including conjugated linoleic acid – CLA) which are most desirable from the nutritive point of view, is negatively correlated with the daily milk yield.

KEY WORDS: fatty acids / goats / milk / lactation

Milk is one of the essential products in the human diet, rich in nutritive components. Although the production and consumption of cow milk is the largest throughout the world, one may observe a growing demand for milk of other farm animals, such as goats, which is recognized in developed countries as a “niche” product [Kanwal *et al.* 2004, Haenlein and Wendorff 2006, Krzyżewski *et al.* 2009].

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Although several authors have examined the nutritive value of goat milk taking into consideration various factors [Pilar *et al.* 1998, Andrade and Schmidely 2006, Matsushita *et al.* 2007, Pandya and Ghodke 2007, Strzałkowska *et al.* 2008], little is known about its composition and physical traits in relation to the stage of lactation. Therefore, the present study aimed at evaluating the chemical composition, selected physical traits and fatty acid profile of goat milk throughout the lactation.

Material and methods

Animals, feeding, sampling

The study was carried out on 20 Polish White Improved goats in their second lactation, maintained at the Experimental Farm of the Institute of Genetics and Animal Breeding, Jastrzębiec, Poland. The goats were fed diets formulated according to Institut National de la Recherche Agronomique [INRA-IZ, 2001] standards which met all their individual nutritive requirements. The winter diet (October to May) consisted of corn silage, hay, carrot and concentrate fed according to the milk yield, and mineral and vitamin premix, chalk and NaCl. During the summer season (June to September) hay was substituted by fresh grass. Goats were machine-milked twice a day. Milk samples were taken from morning and afternoon milking of each goat, on day 60, 120 and 200 (about the peak of lactation, mid lactation, and the end of lactation) – 1, 2 and 3 lactation stage, respectively.

Chemical composition and physical traits of milk

Each milk sample was analysed for the content of fat, protein, casein, total solids, solids non fat, lactose, urea, citric acid and free fatty acids. Moreover, the density, acidity and freezing point were determined. The contents of components and parameters of milk were estimated using a MilkoScan FT2.

Fatty acid profile

Frozen milk samples were freeze-dried for 48 h and then extracted with chloroform-methanol and water (4:2:1,v/v) mixtures. The lower layer was hydrolysed and free fatty acids were extracted [Czauderna *et al.* 2001, Czauderna and Kowalczyk 2001], The praecipitate obtained was dissolved in a mixture of dibromoacetophenone and triethylamine, after which acetic acid was added to stop the derivatization reaction. The derivatizing procedure for standards (Sigma life Science 2008-2009) was the same as for biological samples. Derivatized samples were filtered through a 0.2 µm membrane filter (Whatman) and the solutions obtained were injected onto chromatographic columns.

Analyses of the dibromoacetophenacyl ester of fatty acids were carried out on HPLC systems Series 200 PERKIN ELMER, USA. The development of the gradient elution system, collection and data integration were performed with the Turbochrom Workstation Ver. 6.1.2 software. The separation was performed on Spheri-5 RP-18, 5

μ , 220 x 4.6 mm columns (PERKIN ELMER, USA). All solvents were degassed under vacuum and then maintained flushed with 99.996 helium (PRAXAIR, Poland). The column temperature was maintained at 35°C and the eluted dibromoacetophenylacyl esters of fatty acids were detected at 242 nm. Elution was performed using a concentration of a mixture of methanol (MeOH) and acetonitril-water (ACN-H₂O, 40-60, v/v). The elution of dibromoacetophenacyl ester of 3:0-20:5 fatty acids was completed within 40 min at a flow-rate of 2.6 ml/min.

Statistical

Statistical evaluation of results was done with the GLM procedure using the model which included fixed effect of a stage of lactation. The Pearson's correlations were calculated with the CORR procedure [SAS, SAS/STAT 1999-2001].

Results and discussion

Chemical composition and physical traits of milk

Changes observed in the yield and chemical composition of milk throughout the lactation are shown in Table 1. The daily milk yield differed clearly between stages of lactation. The protein content of milk increased with the progress of lactation, while the content of fat was the lowest about lactation day 120 (stage 2).

Similar dynamics in the milk protein and milk fat production over lactation in sheep and goats was reported by Aganga *et al.* [2002] and Soryal *et al.* [2005]. In the present study the increasing concentration of milk protein was accompanied by a significant ($P<0.01$) change in the content of its most important fraction – casein – the content of which, as compared to the lactation stage 1, increased by 0.44 per cent units (Tab. 1). Similar regularity in the concentration of casein in the milk of goats was observed by Soryal *et al.* [2005]. The increase in concentration of protein and fat in subsequent stages of lactation had a direct effect on the content of total solids ($P<0.05$) and solids-non-fat ($P<0.01$). As regards the remaining traits of milk, significant differences ($P<0.01$) were observed in the titrimetric acidity (SH) of milk. One may assume that the increased concentration of protein, including the casein fraction which to a considerable degree decide about the acidity of milk, was the direct cause of such change.

During the subsequent stages of lactation a significant ($P<0.01$) increase was recorded in the freezing point of milk. One should emphasize that the values obtained for this indicator in the present study markedly exceeded those reported by other authors. Park *et al.* [2007] reported the freezing point for goat milk to range between -0.540 and -0.570°C, while in the milk of animals considered and discussed here the freezing point occurred much lower, and amounted to -0.609°C, -0.596°C and -0.625°C for three subsequent lactation stages, respectively. Such low values for the freezing point may be related to the fact that the concentration of individual milk components increased with the progress of lactation.

Table 1. Least squares means (LSM) and their standard errors (SE) for daily milk yield, milk chemical composition and milk physical traits in goats across the three stages of lactation

Item	Stage 1		Stage 2		Stage 3	
	LSM	SE	LSM	SE	LSM	SE
Daily milk yield (kg)	1.08 ^A	0.06	1.38 ^B	0.04	0.74 ^C	0.05
Fat (%)	3.67	0.18	3.38	0.21	3.85	0.24
Protein (%)	2.98 ^A	0.08	3.12 ^A	0.09	3.66 ^B	0.11
Casein (%)	2.41 ^A	0.07	2.48 ^A	0.08	2.85 ^B	0.09
Total solids (%)	11.89 ^a	0.22	11.83 ^a	0.26	12.75 ^b	0.30
Solids-non-fat (%)	8.31 ^A	0.10	8.34 ^A	0.12	8.86 ^B	0.14
Lactose (%)	4.42 ^a	0.04	4.29 ^b	0.04	4.40 ^{ab}	0.05
Urea (mg/%)	127.6 ^A	27.85	295.6 ^B	33.26	315.7 ^B	38.16
Citric acid (%)	0.09 ^A	0.01	0.10 ^A	0.01	0.06 ^C	0.01
FDP (-°C)	0.609 ^A	2.28	0.596 ^B	2.73	0.625 ^C	3.13
FFA mMol/L)	0.74 ^a	0.06	0.96 ^b	0.08	1.05 ^b	0.08
Density (g/L)	1025.9 ^A	0.52	1025.7 ^A	0.62	1029.8 ^B	0.71
Acidity (SH)	6.10 ^A	0.55	5.88 ^A	0.66	7.44 ^B	0.76

^{aA} Within rows means bearing different superscripts differ significantly at: small letters – P<0.05; capitals – P<0.01.

FFA – free fatty acids.

FDP – freezing point.

Changes in the milk fat content were accompanied by a significant (P<0.05) increase in the level of free fatty acids (FFA), the maximum of which (1.05 mmol/l), was observed in milk from lactation day 200) – Table 1. This indicates that fat present in the milk during the final stage of lactation is more susceptible to lipolysis, which may be caused by enzymes naturally present in the mammary gland as well as by bacterial enzymes [Gajdusek *et al.* 1993, Collins *et al.* 2003]. The results reported by Eknes *et al.* [2006] indicate that if the FFA content of milk exceeds 2.0 mmol/l the characteristic „goat flavour” is clearly stronger and may prove unacceptable to some consumers. The FFA level is an indicator of the process of lipolysis in milk. A negative energy balance occurring during the early stage of lactation, leading to lipolysis of the fatty tissue, may intensify processes that have a negative effect on the palatability traits of milk. During the peak of lactation even 30-40% of the body fat may be subjected to lipolysis, causing changes in the milk FFA content. Over that time milk fat is synthesized from fatty acids obtained through the hydrolysis of the fat tissue. Simultaneously, the concentration of FFA is high (principally C6:0-C9:0), what leads to undesirable changes in the milk taste and flavour [Chilliard *et al.* 2003, Eknes *et al.* 2006]. Recently Eknes *et al.* [2009] reported, that adding palmitic acid to the diet for goats reduces the undesirable FFA content of milk. Moreover, an addition of the acid in question leads to an increase in the blood serum cholesterol content, what has a positive effect on the stabilization of the milk fat globule membrane (MFGM). Fat globules in such a situation are less susceptible to lipolysis. The resulting lower FFA concentration positively affects the palatability of milk.

Fatty acids profile

The goat milk fat is composed of several hundred fatty acids, the share of which in the total FA pool is considerably differentiated. The share of five of those acids (C10:0, C14:0, C16:0, C18:0, C:18-1 cis) comprises over 75% of total FA in milk [Park *et al.* 2007]. In the present study, the percentage of capric, myristic, palmitic, oleic and

Table 2. Least squares means (LSM) and their standard errors (SE) for percentages of individual fatty acids and their groups in relation to the sum of fatty acids considered as 100

Fatty acid	Stage 1		Stage 2		Stage 3	
	LSM	SE	LSM	SE	LSM	SE
C4:0	1.37	0.23	0.88	0.25	1.56	0.30
C6:0	5.93 ^A	0.38	5.35 ^{AB}	0.43	4.25 ^B	0.53
C8:0	6.17 ^A	0.21	5.39 ^B	0.23	5.03 ^B	0.28
C10:0	15.74 ^A	0.36	14.43 ^B	0.41	13.55 ^B	0.50
C12:0	6.20 ^A	0.19	5.60 ^B	0.21	6.59 ^A	0.26
C12:1	0.13 ^A	0.01	0.08 ^B	0.02	0.24 ^C	0.02
C14:1	0.22 ^A	0.03	0.17 ^A	0.03	0.43 ^B	0.04
C14:0	11.62	0.32	11.15	0.37	12.30	0.44
C16:0	21.58	0.54	22.19	0.61	21.19	0.74
C16:1	0.40 ^A	0.06	0.69 ^B	0.07	0.84 ^B	0.08
C17:0	0.45	0.03	0.46	0.04	0.41	0.05
C18:0	8.58 ^A	0.48	9.07 ^A	0.55	5.87 ^B	0.67
C18:1 <i>cis</i>	16.56 ^A	0.64	18.41 ^B	0.73	19.88 ^B	0.88
C18:1 <i>trans</i>	2.21 ^{AB}	0.28	1.72 ^A	0.31	3.13 ^B	0.38
C18:2	0.99 ^A	0.11	1.75 ^B	0.12	1.98 ^B	0.15
C18:3	0.66 ^A	0.12	1.30 ^B	0.14	0.92 ^{AB}	0.17
CLA	0.13 ^A	0.04	0.18 ^A	0.05	0.41 ^B	0.06
C20:3	0.26	0.02	0.25	0.03	0.25	0.03
C20:4	0.69 ^A	0.05	0.77 ^A	0.06	0.99 ^B	0.07
C20:5	0.09	0.02	0.13	0.03	0.17	0.03
SFA	77.65 ^A	0.87	74.52 ^B	0.99	70.74 ^C	1.21
MUFA	19.52 ^A	0.82	21.08 ^B	0.93	24.52 ^B	1.14
PUFA	2.82 ^A	0.21	4.39 ^B	0.24	4.73 ^B	0.29
SCFA	29.22 ^A	0.94	26.06 ^B	1.06	24.38 ^B	1.29
MCFA	40.61	0.86	40.34	0.98	42.01	1.19
LCFA	30.18 ^A	1.08	33.59 ^B	1.22	33.60 ^B	1.49
Σ C10-18; C18:1	74.09 ^A	0.60	75.25 ^A	0.68	72.79 ^B	0.83

^{aA} Within rows means bearing different superscripts differ significantly at: small letters – P<0.05; capitals – P<0.01.

CLA – conjugated linoleic acid (rumenic acid); SFA- saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; SCFA – short-chain fatty acids; MCFA – medium-chain fatty acids; LCFA – long-chain fatty acids.

stearic acids in the total FA occurred similar to that reported by the authors cited. Significant differences ($P < 0.01$) were observed during different stages of lactation, the lowest value being recorded in milk sampled on lactation day 200 (Tab. 2).

One should emphasise that the fat present in goat milk is a rich source of short-chain fatty acids (SCFA – C6:0, C8:0, C10:0), which are synthesized *de novo* in the mammary gland [Chilliard *et al.* 2006]. The share of those acids in the pool of FA composing the goat milk fat is more than twice as high as in cow milk (about 18% vs. 8%) – Chilliard *et al.* [2006], Sanz Sampelayo *et al.* [2007]. In the present study the level of SCFA in the goat milk reached its maximum (29.22%) at the start of lactation (stage 1) and next dropped reaching a minimum (24.38%) during stage 3. The results obtained differ from those reported by Soryal *et al.* [2005], who observed a high content of SCFA at the beginning, and then in the last stage of lactation. The SCFA concentration in the milk of goats is important, as it decides about the palatability and sensory properties of milk and dairy products obtained [Eknes *et al.* 2009, Talpur *et al.* 2009].

A characteristic trait distinguishing goat milk from cow milk is the relation between the lauric (C12:0) and capric (C10:0) acid (0.46 vs. 1.16% of sum of acids) [Haenlein and Wendorff 2006]. This is an important indicator, as it may be used to detect falsifications of goat milk with cow milk. In the milk of the goats examined here the values of this indicator during individual stages of lactation amounted to 0.38, 0.38 and 0.49, for lactation stage 1, 2 and 3.

In the fat of goat milk, saturated fatty acids (SFA) are the dominating group and their share in the milk fat ranges from about 67% [Rodriguez-Alcala *et al.* 2009] to about 75% [Žan *et al.* 2006]. In the present study a significant ($P < 0.01$) decrease in the SFA content was observed with the progress of lactation (Tab. 2). Within the SFA pool the C16:0 constitutes the largest item and in the goat milk analysed in the present study the share of this acid in the total FA pool amounted to 21.58%, 22.19%, 21.19% for lactation stage 1, 2 and 3, respectively. The high concentration of the acid in the goat milk fat is not a characteristic of this animal species, but of a majority of mammals [Haenlein and Wendorff 2006]. In turn, the share of monounsaturated fatty acids (MUFA) in the total FA pool of the goat milk fat, may range from about 20% [Žan *et al.* 2006] to about 32% [Talpur *et al.* 2009]. Among the MUFA group, the oleic acid (C18:1) is characterized by the highest content, what is typical for a majority of mammals [Haenlein and Wendorff 2006]. In the present study a significant ($P < 0.01$) increase was observed in the level of oleic acid with the progress of lactation. Similar relations were reported by Tsiplakou and Zervas [2008].

Polyunsaturated fatty acids (PUFA) are, due to their favourable effect on the health of consumers, the most valuable group among FA and comprise 3-5% of a total FA pool [Rodriguez-Alcala *et al.* 2009]. In the milk fat of the goats examined in the present study, the content of PUFA increased significantly ($P < 0.01$) during the progress of lactation and their maximum share (4.73%) was observed in stage 3. Among the PUFA group the conjugated linoleic acid (CLA) is one of the most

valuable. Both the acid itself and its isomers (principally *cis-9*, *trans-11* and *trans-10*, *cis-12*), are characterized by an exceptionally high biological activity [Gulati *et al.* 1997, Parodi 2002, Zhang *et al.* 2006]. One should emphasise, that in the human

Table 3. Pearson correlation coefficients of daily milk yield, milk fat content and fatty acids in goat milk

	Milk yield	fat	C10:0	C12:0	C14:0	C16:0	C18:1c	SCFA	MCFA	LCFA	SFA	MUFA	PUFA	C10-C18	CLA
Milk yield	1.00														
Fat		1.00													
C10:0			1.00												
C12:0				1.00											
C14:0					1.00										
C16:0						1.00									
C18:1c							1.00								
SCFA								1.00							
MCFA									1.00						
LCFA										1.00					
SFA											1.00				
MUFA												1.00			
PUFA													1.00		
S C10-C18														1.00	
CLA															1.00

*p<0.05; **p<0.01;
 CLA – conjugated linoleic acid (rumenic acid); SFA- saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; SCFA – short-chain fatty acids; CFA – medium-chain fatty acids; LCFA – long-chain fatty acids.

diet milk fat is the principal source of CLA and covers 75% of the daily requirement of the human organism for this compound. The CLA content of milk is determined principally by the nutrition of the animal. The results of D'Urso *et al.* [2008] indicate that also other factors, such as stage of lactation, breed and age of animal, affect the CLA concentration of goat milk. If the animals are grazing the synthesis of isomer *cis-9, trans-11*-CLA in the goat's mammary gland may reach even 91% of the total amount of CLA present in the milk fat [Kay *et al.* 2004]. In the study presented here, the content of CLA in the goat milk increased significantly ($P < 0.01$) with the progress of lactation, but it was lower than that reported by Park *et al.* [2007]. The coefficients of the calculated Pearson's correlation show that the concentrations of MUFA and PUFA (including CLA), *i.e.* the fatty acids most desirable from the nutritive point of view, are negatively correlated with the daily milk yield (Tab. 3).

Summarizing, one should emphasise, that the changes in the chemical composition and physical properties of goat milk, observed in the present study, correspond with those reported by other authors [Fekadu *et al.* 2005, Zhang *et al.* 2006, Park *et al.* 2007]. Thus, one may consider them as typical for a standard goat lactation. With the progress of lactation highly favourable changes take place in the concentration of the most desirable FA, *i.e.* MUFA and PUFA. Those fatty acids are the source of what is known as functional compounds and have a favourable effect on the organism of consumers [Gajdusek *et al.*, 1993].

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Skład chemiczny, cechy fizyczne i profil kwasów tłuszczowych mleka kóz w przebiegu laktacji

Streszczenie

Badania przeprowadzono na 20 kozach rasy polskiej białej uszlachetnionej, będących w drugiej laktacji. Dawki pokarmowe zestawiono zgodnie z normami INRA. W sezonie zimowym zwierzęta żywiono dawką złożoną z kiszonki z kukurydzy, siana, marchwi i koncentratu, w ilości dostosowanej do wydajności mleka. Ponadto kozy otrzymywały dodatki mineralno-witaminowe oraz kredę i sól pastewną. W sezonie letnim kiszonkę z kukurydzy zastąpiono zielonką z traw. Kozy doiono mechanicznie dwukrotnie w ciągu dnia. Próbkę mleka każdej kozy pobrano w 60, 120 i 200 dniu laktacji (stadium 1, 2 i 3). Wraz z przebiegiem laktacji stwierdzono korzystne zmiany w profilu kwasów tłuszczowych (FA) w mleku – rósł udział kwasów jednonienasyconych (MUFA) i wielonienasyconych (PUFA). Oszacowane wskaźniki korelacji Pearsona wskazują, że koncentracja kwasów najkorzystniejszych z punktu widzenia wartości odżywczej, tj. MUFA, PUFA, w tym sprzężonego kwasu linolowego (CLA), była ujemnie skorelowana z dobową wydajnością mleka.