# THE RELATIONS OF SOME MILK INDICATORS OF ENERGY METABOLISM IN COW, GOAT AND SHEEP MILK<sup>\*</sup>

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The milk indicators (MIs) of energy metabolism, protein (CP), fat (F) and acetone (AC) determine and also influence the qua lity of milk. Deterioration in their values point to animal fertility disorders. Thus, these MIs can be used for monitoring and preventing the negative energy balance (NEB) in animals at the beginning part of lactation. In this way both animal health and milk quality can be improved. For this reason, selected relations between the energy metabolism indicators (AC and the fat/protein (F/CP) ratio) were studied in bulk cow (Czech Fleckvieh (CF), n = 40; Holstein (HN), n = 36), goat (White short-haired (WS), n = 40) and sheep (Tsigai (TS), n = 40) milk samples in the first half of lactation. The cows were fed a total mixed ration (TMR) based on maize, alfalfa and clover silage with brewers draff and concentrates. The goats and sheep grazed on a semicultural grazing growth and fattened up by concentrates. The differences in CP, F, F/CP and AC were significant for both breed and species (P < 0.05). The highest AC (12.68 (8.73 xg) mg.l<sup>-1</sup>) was in TS. The F/CP variability was relatively low and in AC in contrast high as is typical for health indicators, from 54.5% (HN) to 98.9% (WS). Whereas the AC means (TS, CF 7.77 (6.34 xg) and HN 2.03 (1.60 xg) and WS 7.55 (3.50 xg) mg.l<sup>-1</sup>) can be considered species specific, the same is not true for their variability. Positive relations were found between AC and F/CP in cows CF and HN (r = 0.34, P < 0.05; r = 0.15, P > 0.05) and in sheep (r = 0.40, P < 0.05). A negative relation was surprisingly found for goat milk (r = -0.22, P > 0.05). This shows good agreement in the informative ability for both the indicators of energy metabolism not only in cow but also in sheep milk. Further studies are necessary in the case of goat milk. The F/CP ratio is accessible from routine milk recording, in contrast to the less frequent AC examination.

cow; goat; sheep; bulk milk sample; energy metabolism indicator; acetone; fat/crude protein ratio

#### INTRODUCTION

The fat/protein (crude protein) coefficient (F/CP) for milk is considered an indicator of cow energy metabolism, as production of both depends first of all on this part of the total metabolism (Table 1; H e u e r et al., 2001). For this reason it is possible to estimate the energy surplus or malnutrition due to fodder ration in relation to milk yield capacity, primarily in the risk first third of lactation. With nutrition energy deficiency, the milk protein content usually decreases (K i r c h g e s s n e r et al., 1985, 1986) while the fat content may increase at first. Other opinions also consider the F/CP ratio as an indicator of the technological suitability of milk, especially for the cheese-making process (Table 1; A g a b r i e l et al., 1990, 1991; B i r o et al., 1992).

According to the majority of studies an urea concentration in milk as a final product of protein catabolism is thought to be the indicator of nitrogen and energy metabolism (Baker et al., 1985; Kirchgessner et al., 1985, 1986; Garcia, Linn, 1997; Mottram et al., 2002; Johnson, Young, 2003; Kuchtík, Sedláčková, 2003). In the case of unfavourable milk urea (U) variability dairy cow longevity is also endangered in addition to reproductive performance (Haraszti, Zöldag, 1990; Butler et al., 1996; Hanuš et al., 1993b (Fig. 1), 2004a; Rajala-Schultz et al., 2001; Hojman et al., 2004; Miglior et al., 2006). U and AC values are often higher in high milk yielding dairy cows (Hanuš et al., 2007; Janů et al., 2007). Urea and AC have been investigated in dairy goats and sheep in some papers (Hanuš et al., 2004b, 2008; Stella et al., 2007; Genčurová et al., 2008a,b).

The concentrations of milk ketones or AC (Steger et al., 1972; Andersson, 1984, 1988; Andersson, Lundström, 1984a,b;

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Table 1. Interpretation of the fat content/crude protein content (F/CP) ratio in cow milk (modified according to: Agabriel et al., 1991, 1992; Bíro et al., 1992; Fürll et al., 1992; Geishauser and Ziebell, 1995; Schulz, 1997; Gasteiner, 2000)

Interpretation:	Too low F/CP	Suitable F/CP	Too high F/CP				
- In relation to the nutritional physiology of dairy cows (individual milk samples):							
For Holstein dairy cows	< 1.05	from 1.05 to 1.18	> 1.18				
For combined and milk breeds (in Germany)	< 1.1	from 1.1 to 1.6	> 1.6				
	- shortage of structural fiber in feeding ration		- shortage of energy, risk of ketosis				
- In relation to cheesemaking technology (bulk and tank milk samples):							
Milk in general (in France)	< 1.10	from 1.10 to 1.20	> 1.20				
		- the best technological values					

A n d e r s s o n, E m a n u e l s o n, 1985; E m a n u e l s o n, A n d e r s s o n, 1986; G r a v e r t et al., 1986; G u st a f s s o n, E m a n u e l s o n, 1993; G e i s h a u s e r et al., 1997, 1998; H a n u š et al., 2001b (Fig. 2); E n j a l b e r t et al., 2001; M o t t r a m et al., 2002; W o o d et al., 2004) are suitable indicators of animal energy metabolism in the sense of final degree of catabolism of body fat reserves and ketose growth. This implies these metabolic milk indicators, which allow the negative energy balance (NEB) in animals, mostly cows to be successfully monitored during the risk period in the beginning of lactation, affect the milk processability as well. Milk fermentation ability which is important to some parts of milk processing,



Fig. 1. The relation of acetone levels in dairy cows (individual milk samples) to some reproduction indicators in the first third of lactation (Říha, Hanuš, 1999; Hanuš et al., 2004a)

is significantly negatively influenced by unfavourable milk U and acetone (AC) values (Hanuš et al., 1993a,c). In cows with an increased AC in the second and third months of lactation (0.25 mmol.l-1) was found out a significant negative correlation to the amount of energy received through fodder (r = -0.47 and -0.42, resp.) and milk yield capacity (r = -0.30, Gravert et al., 1991; Miettinen, 1994). It follows from this that a high milk AC indicates a labile metabolism. However, the quality of silage also has a significant influence on the acetone content of milk. The heritability coefficient for AC in milk was shown to be 0.30 (Gravert et al., 1991) for the first three months of lactation and was thus similar to the coefficient of milk yield capacity. For this reason, it was recommended that AC content assessment in milk as an energy balance indicator should be added to the measurements used in milk recording including determination of the breed value. Earlier, a positive correlation (r = 0.30; P < 0.001) between log AC and the F/CP coefficient in cow milk had been found (Hanuš et al., 2004a). This coefficient confirmed the mentioned informative value of both indicators. These facts apply primarily to cow metabolism and



Fig. 2. Scheme of acetone (ketone) concentration increase in milk or other body liquids such as blood or urine of ruminants with ketosis degree (Hanuš et al., 2001b, 2004a)

milk production. However, there is a lack of information in the literature on these relations in the milk in small ruminants.

A number of studies (E m er y et al., 1964; Miettinen, 1995; Green et al., 1999; Gasteiner, 2003; Tedesco et al., 2004; Stella et al., 2007) describe positive modifications in early lactation metabolism and corresponding possible energy deficits by addition of various glucoplastic or hepatoprotective substances (as propyleneglycol, monensin, *Silybum marianum*, *Saccharomyces cerevisiae*) to feeding rations in cows and small ruminants as well.

The milk indicators (MIs) of animal energy metabolism as protein (CP), fat (F) and acetone (AC) or urea (U) determine and influence milk quality essentially. Their deteriorated values point out the animal fertility disorders (K a u p p i n e n, 1984) as lactation is part of reproduction. Therefore, the aim of this study was to compare the relations of the energy MIs in cow, goat and sheep milk in order to standartise interpretation and determine the applicability of the suggested diagnostic methods for the milk production of small ruminants, as well. This kind of milk production is carried out in the CR principally in less favourable areas (LFA) with generally greater problems in securing energy fodder sources as opposed to cow milk production which is practised in LFA but more often in agriculturally very favourable areas with better energy sources.

#### MATERIALS AND METHODS

## Sets of bulk milk samples from selected biological species and breeds of ruminants

(a) Bulk milk samples (MSs; small groups of animals, from 4 to 6 animals in one sample, which was valid for all species; n = 40) were obtained in the spring and summer season (three months) from one dairy herd with Czech Fleckvieh cattle (CF; *Bos primigenius* f. *taurus*, L, 1758). The cows were on different lactations according to the herd profile. They were kept in LFA and milked with pipeline milking equipment twice a day. The dairy cows were fed by total mixed ration (TMR) based on: maize silage 13 kg; trifolium silage 9 kg; whole spindle maize silage (LKS) 5 kg; brewery draff 3 kg; concentrates 6 kg per cow and day. The average daily milk yield was 26.17 kg. The milk samples were collected in the first half of lactation.

(b) Bulk MSs (n = 36) were obtained in the winter and spring season (three months) from one dairy herd of Holstein cattle (HN; *Bos primigenius* f. *taurus*, L, 1758). The cows were kept in a good productive area and were on different lactations according to profile of whole herd. They were milked in a milking parlour twice a day. The dairy cows were fed by TMR based on: maize silage 15 kg, alfalfa silage 10 kg, whole spindle maize silage (LKS) 5 kg, brewery draff 3 kg, alfalfa hay 1 kg, dried whey 0.3 kg and concentrates with feeding yeasts 5 kg per cow per day. The average daily milk yield was 28.11 kg. The milk samples were collected in the first half of lactation, where the risk of negative energy balance and negative impact on the reproductive performance of dairy cows can be important.

(c) Bulk MSs (n = 40) were obtained in spring and summer season (four months) from one goat herd of White Short-haired breed (WS; *Capra aegagrus* f. *hircus*, L, 1758). The goats were on different lactations according to herd profile and were kept in LFA. The goats together with sheep grazed on half culture grass growth (composition with the dominance: *Poa pratensis*, *Dactylis glomerata*, *Elytrygia repens*, *Trifolium repens* and *Taraxacum officinale*) with supplementation of concentrates in the milking parlour. The average daily milk yield was 3.10 kg. The milk samples were collected in the first half of lactation. The animals were milked twice a day.

(d) Bulk MSs (n = 40) were obtained in the spring and summer season (four months) from one sheep flock of Tsigai (TS; *Ovis aries*, L, 1758) breed. Sheep were on different lactations according to flock profile. They were kept in LFA. The sheep together with goats grazed on the same half culture grass growth with supplementation of concentrates. The average daily milk yield was 0.55 kg. The milk samples were collected in the first half of lactation. Sheep were milked twice a day in a milking parlour.

#### Chemical milk analyses

The MSs were transported unpreserved and under cold conditions ( $\leq 8^{\circ}$ C) to an accredited testing laboratory (EN ISO 17 025, number 1340, 124/2004) immediately after sampling. The samples were analysed using the reference methods for the following indicators: fat content (F; g.100  $g^{-1}$ ; %); crude protein content (CP; Kjeldahl total N  $\times$  6.38; g.100 g<sup>-1</sup>; %); acetone concentration (AC; mg. $l^{-1}$ ). Fat values were investigated by the Gerber's method in accordance with CSN 57 0530 and divided by specific weight values which were investigated using Mohr's hydrostatic scale. CP was determined by Kjeldahl's method (instrument 2200 Kjeltec Auto Distillation, Foss-Tecator AB, Sweden) according to standard CSN 57 0530. The F and CP analyses results were also compared and completed using Milko-Scan 133B (Foss Electric, Denmark) investigation by species specific calibrations (according to Z eng 1996). Recently the FT-MIR technology has been confirmed as useable for ketone measurement in milk (H a n s e n, 1999; Heuer et al., 2000; Roos et al., 2006). Various effective stable tests for ketone determination in milk and urine have also been successfully compared and evaluated (Geishauser et al., 1997; Hanuš et al., 2001b; Carrier et al., 2004). In this study the

milk AC was investigated by spectrophotometry at 485 nm wavelength using Spekol 11 instrument (Carl Zeiss Jena, Germany). The acetone was absorbed in an alkaline solution of KOH with salicylaldehyde (O'M o o r e, 1949; Vojtišek, 1986) due to 24 hours of microdiffusion. The Spekol was calibrated by the five points on the scale with an increase of AC from 1 to 20 mg.<sup>1–1</sup>.

#### Statistical evaluation of data sets

The milk AC values were used in their raw form and after their logarithmic transformation to normalise the data (M e l o u n, M i l i t k ý, 1992, 1994; H a n u š et al., 2001a,b). The basic statistical characteristics were calculated for all values of indicators in data sets (n = 4) as: arithmetical mean (x); geometrical mean (xg); median (m); standard deviation (sd); variation coefficient (vx). The data sets were evaluated using linear and nonlinear regression with the Microsoft Excel programme. Variation, determination and regression coefficients, correlation coefficients and indices were compared for dairy cow breeds (n = 2) and among biological kinds of milk of selected species (n = 3). The cow results were mostly used as reference for small ruminants.

#### **RESULTS AND DISCUSSION**

### The characteristics of energy indicators in the biological milk sorts

The mean differences in the observed milk indicators (F, CP, F/CP, and AC) were predominantly significant between breeds and species, as expected (Tables 2 and 3, observations A, B, C, D). The highest values for acetone (TS 12.68 (8.73 xg) mg.l<sup>-1</sup>) were unambiguously in the componently most concentrated sheep milk with the typically lowest milk yield capacity among the compared species. The variability of the F/CP indicator was relatively low with values around approximately 10% for cows and 20% for small ruminants in bulk MSs. However, it was undoubtedly decreased due to the method of sampling as opposed to the possible values of individual animals. The variability of the AC indicator was high as is typical for the health indicators and ranged from 54.5% (cow HN) to 98.9% (goat). Whereas the mean values in the components, especially in the major ones (F and CP) but also acetone (CF cow 7.77 (6.34 xg) and HN 2.03 (1.60 xg), goat WS 7.55  $(3.50 \text{ xg}) \text{ mg.l}^{-1}$  and sheep TS), can be considered as approximately genetically specific, this is not valid in terms of their variability. In earlier results (Hanuš et al., 2005) significant differences were found in the mean values of both milk energy indicators (AC and F/CP) between the biological types of milk. In acetone cow  $\approx$  goat < sheep and in the F/CP ratio cow < sheep < goat. Here the results (Table 2) show the order for AC sheep (TS) > goat (WS) or cow (CF and HN) and for F/CP goat (WS 1.51) > sheep or cow (TS and HN 1.21 or CF 1.08). The significant difference in AC between goats and sheep (xg 3.50 < 8.73 mg.l<sup>-1</sup>; Tables 2 and 3) under conditions of equivalent nutrition could be explained perhaps only by sheep lower milk yield, where similar quantity of acetone has to be more concentrated into smaller daily milk volume.

According to D e l g a d o - P e tr i ň e z et al. (2009)the average F/CP ratio can vary in Florida dairy goats from 1.54 to 1.69 in the studied kinds of rearing systems

Table 2. Statistics of various milk kinds in energy metabolism indicators

		F	СР	F/CP	AC	log AC
Milk kind		(g.10	0g <sup>-1</sup> )		(mg. l <sup>-1</sup> )	
	n	40	40	40	40	40
	X	3.71	3.45	1.08	7.77	0.8018
	xg					6.34
Cow-Czech	sd	0.458	0.172	0.127	4.942	0.2960
Fleckvieh	VX	12.3	5.0	11.8	63.6	
(CF)	min	3.01	3.13	0.90	1.11	0.0453
	max	4.70	3.85	1.38	26.49	1.4231
	m	3.54	3.45	1.05	8.00	0.9028
	n	36	36	36	36	36
	x	4.06	3.36	1.21	2.03	0.2042
	xg					1.60
Cow –	sd	0.418	0.114	0.108	1.106	0.3717
(HN)	VX	10.3	3.4	8.9	54.4	
(1114)	min	3.22	3.10	0.97	0.08	-1.0969
	max	4.93	3.55	1.39	4.61	0.6637
	m	3.99	3.36	1.20	1.95	0.2901
	n	40	40	40	40	40
	x	4.82	3.27	1.48	7.55	0.5446
	xg					3.50
Goat – White	sd	1.047	0.252	0.313	7.468	0.7614
(WS)	VX	21.7	7.7	21.2	99.0	
((()))	min	3.21	2.69	1.03	0.01	-2.0000
	max	7.53	3.87	2.29	30.70	1.4871
	m	4.49	3.25	1.44	5.47	0.7300
Sheep – Tsigai (TS)	n	40	40	40	40	40
	x	7.60	6.25	1.21	12.68	0.9410
	xg					8.73
	sd	1.810	0.543	0.223	8.527	0.4495
	vx	23.8	8.7	18.4	67.2	
	min	4.57	4.67	0.78	0.82	-0.0862
	max	11.44	7.47	1.63	34.41	1.5367
	m	7.56	6.08	1.25	11.94	1.0767

F - fat; CP - crude protein; F/CP - fat/crude protein ratio;

AC – acetone; n – number of observations; x – arithmetical mean; xg – geometrical mean; sd – standard deviation; vx – variation coefficient; min – minimum; max – maximum; m = median

Table 3. Significance of differences among species and breed milk

		F	СР	F/CP	AC	log AC
CF – HN	t	3.40	2.54	4.72	6.73	7.69
	sign.	**	*	***	***	***
CF – WS	t	6.07	3.65	7.39	0.16	1.97
	sign.	***	***	***	ns	ns
CF – TS	t	13.01	30.68	3.21	3.11	1.62
	sign.	***	***	**	**	ns
HN – WS	t	4.04	1.97	4.84	4.33	2.40
	sign.	***	ns	***	***	*
HN – TS	t	11.32	30.86	0.04	7.34	7.64
	sign.	***	***	ns	***	***
WS-TS	t	8.30	31.06	4.35	2.83	2.80
	sign.	***	***	***	**	**

F - fat; CP - crude protein; F/CP - fat/crude protein ratio; AC - acetone; CF - cow Czech Fleckvieh; HN - cow Holstein; WS - goat White Short-haired; TS - sheep Tsigai; t - t value; sign. - significance; ns P > 0.05; \* P < 0.05; \* P < 0.01 and \*\*\* P < 0.001

during the first five weeks of lactation. Our average result was 1.48. These authors also reported markedly higher average fat content in their data set. In contrast, Z e n g , E s c o b a r (1996) and Z e n g et al. (1997) found lower fat averages of 4.08 and 2.91%. Their F/CP ratio averages were 1.28 and 0.89 in Alpine together with Nubian goats and Alpine goats, which also differ from our results. It is clear that there can be great differences in F/CP ratio between goat breeds under various rearing conditions. This fact is also confirmed by B r a g h i e r i et al. (2007) in sheep. The average F/CP ratio varied from 1.30 to 1.37 in Merino derived sheep in various rearing systems. Sheep F/CP average ratio was 1.21 in our data set, which is also different. Probably it will be necessary to estimate specific



Fig. 3. The relation between F/CP ratio and acetone concentration (log AC) in individual milk samples in Holstein dairy cows during their first third of lactation and summer sampling period (Hanuš et al., 2005)

F/CP diagnostic limits according to local conditions in herds and flocks of small ruminants.

# The mutual relations of the energy milk indicators in various biological sorts of milk

In cows the milk AC at the beginning of lactation is closely related to the reproduction indicators which are evidently deteriorated with growing AC value (Fig. 1). Thus, it is a good indicator of the fertility disorders. With respect to energy metabolism a very close correlation was found out between AC in milk and the F/CP ratio in HN cows in the first third of lactation in the summer season (Fig. 3; r = 0.48; P < 0.001). This indicates the very good informative value of both indicators.

In this study an interesting positive relation was found between the AC and F/CP energy indicators in cows of the CF breed (Table 4; Fig. 4; r = 0.34; P < 0.05). This is in accord with the previous results

	Relationship between	Relationship type			- 2	
Milk kind	indicators	linear	nonlinear	Equation	R <sup>2</sup>	r
	$AC \times F/CP$	LIN		y = 0.0086x + 1.0105	0.1105	0.33 <sup>ns</sup>
Cow CF	$\log AC \times F/CP$	LIN		y = 0.1251x + 0.9767	0.0847	0.29 <sup>ns</sup>
	$AC \times F/CP$		EXP	$y = 1.0055 e^{0.008x}$	0.1180	0.34*
	$AC \times F/CP$	LIN		y = 0.0085x + 1.1899	0.0077	0.09 ns
Cow HN	$\log AC \times F/CP$	LIN		y = 0.0358x + 1.1999	0.0152	0.12 <sup>ns</sup>
	$\log AC \times F/CP$		POL2	$y = -0.0459x^2 + 0.0258x + 1.21$	0.0224	0.15 <sup>ns</sup>
Goat WS	$AC \times F/CP$	LIN		y = -0.0093x + 1.5469	0.0487	-0.22 ns
	$\log AC \times F/CP$	LIN		y = -0.0373x + 1.4973	0.0082	-0.09 ns
	$AC \times F/CP$		EXP	$y = 1.5143e^{-0.0061x}$	0.0490	0.22 <sup>ns</sup>
	$AC \times F/CP$	LIN		y = 0.0091x + 1.0930	0.1225	0.35 *
Sheep TS	$\log AC \times F/CP$	LIN		y = 0.0868x + 1.1273	0.0307	0.18 <sup>ns</sup>
	$AC \times F/CP$		POL2	$y = 0.0006x^2 - 0.0062x + 1.1606$	0.1639	0.40 *

Table 4. The regression relationships between energy metabolism indicators in bulk milk samples in various ruminant species in first half of lactation

CF - cow Czech Fleckvieh; HN - cow Holstein; WS - goat White Short-haired; TS - sheep Tsigai; LIN - linear; POL2 - multinominal of second degree; EXP - exponential;  $R^2$  - determination coefficient; r - correlation coefficient or index;  $^{ns}P > 0.05$ ;  $*P \le 0.05$ 



Fig. 4. The regression relation between acetone concentration and F/CP ratio in bulk milk of Czech Fleckvieh (CF) cows in first half of lactation



Fig. 6. The regression relation between acetone concentration and F/CP ratio in bulk milk of goat of White Short-hired breed (WS) in first half of lactation

(Fig. 3) and indicates the relatively good informative value for the two milk indicators of energy metabolism. With the growth of milk AC by 10 mg.l<sup>-1</sup> (i.e. approximately the growth value moving the cow indicated as healthy to subclinical ketosis, H a n uš et al., 2001b) the value of F/CP ratio grows by 0.086. Furthermore, it can be said that as much as 11.8% of the variation in the F/CP ratio is explainable by variations in AC. The results also show that the relationship between F/CP and AC in cows can be markedly closer in individual milk samples (Fig. 3; correlation 0.48 > 0.34) from classical milk recording than bulk samples.

Unlike the CF cows, a positive relation was found in HN cows but this was insignificant and relatively less close (Table 4; Fig. 5; r = 0.15; P > 0.05). This relation is markedly less close than the previous results (Fig. 3). This can be explained by the fact that the AC value dispersion in this group (which obviously represented cows in more balanced nutritional conditions with respect to their milk yield capacity) was much lower, 4 to 8 times than the compared groups. E.g. it was only a quarter of the CF cows (Table 2). The mean value of milk AC was also markedly lower in the HN group than the CF cows (Tables 2 and 3).

The relation of the energy indicators in goat (WS) milk was surprisingly negative (Table 4; Fig. 6; r = -0.22;



Fig. 5. The regression relation between acetone concentration and F/ CP ratio in bulk milk of Holstein (HN) cows in first half of lactation



Fig. 7. The regression relation between acetone concentration and F/CP ratio in bulk milk of sheep of Tsigai breed (TS) in first half of lactation

(0.22); P > 0.05) even with a relatively higher mean value and variability of AC (Table 2). However, in our previous paper (H a n u š et al., 2004b), a positive correlation (r = 0.32; P < 0.01) was found for goat (WS) milk between AC and fat production during the first 100 days in milk. This shows that even in goat milk comparable relations between the energy indicators in comparison to the cow metabolism could exist. An interesting fact was also a positive correlation (r = 0.34; P < 0.01) between the AC and urea in goat milk. However, the urea concentration here is only partially an indicator of energy metabolism and imitates the level of the nitrogen metabolism of animals to a large extent (K i r c h g e s s n e r et al., 1985, 1986). The results in goat milk are thus not definite.

The relation of the energy indicators in sheep (TS) milk was relatively close and significantly positive (Table 4; Fig. 7; r = 0.40; P < 0.05). It was accompanied by higher values and variability of AC in sheep milk (Table 2) as well. This indicates the good accord of information for both indicators of energy metabolism. With AC increase in sheep milk by 10 mg.l<sup>-1</sup> the F/CP ratio value increases by 0.082, which is comparable with the conditions in CF cows (Table 2). It can be further stated that as many as 15.2% of the variability in F/CP ratio is explainable as variability in AC.

The frequent insignificance of even closer physiological relations in these results was caused by lower (but not low) numbers of observations in the individual groups. In general, the monitoring of the possible unfavourable impact of NEB (Negative Energy Balance) on the fertility of female of small ruminants is not as great as in dairy cows. This is due to the different kind of seasonal reproduction physiology in small ruminants. However, in this case too it has a relationship to milk quality and yield. The metabolic milk indicators according to which the NEB of cows can be controlled also affect technological properties and consequently the processability of milk. E.g. (H a n u š et al., 1993a,c) found a weaker but still significant negative correlation of urea and acetone contents to the souring capacity in samples of native cow milk (fermentation ability, or yoghourt test; r = -0.23; P < 0.05; and -0.21; P < 0.05) although the artificial surpluses of these undesirable metabolites in milk resulted in reduction souring capacity only in strong, practically physiologically as well as pathologically unreal concentrations. Therefore it is probable that the reduction of souring capacity in natural milk is not always caused by direct effects of these undesirable metabolites themselves, but rather by the overall changed composition and properties of milk in animals suffering from energy deficiency. The high ketone levels themselves in the body fluids then, as known, can also affect negatively the immunity capacities of the organism, e.g. phagocytic abilities of leucocytes.

#### CONCLUSIONS

The findings of positive correlations confirm the informative ability of these two milk indicators of energy metabolism not only in cow but also in sheep milk. Further studies are necessary in goat milk as for the compared results are not definite. Another question remains about specifying for both AC and F/CP suitable discrimination limits for diagnostics in small ruminants. The approximate estimates have been published for cows (Gravert et al., 1986, 1991; Gustafsson, Emanuelson, 1993). The F/CP ratio indicator is regularly accessible to farmers for practical use from the routine milk recording unlike the less frequent examinations for milk AC. Overall the results can be used to prevent problems connected with serious negative energy balance in the first half of lactation in the ruminant species studied here and contribute to the improving of animal health and milk quality. These problems affect the animals mostly in the first third of lactation, that is, the animals that produce the decisive volume of milk delivery in the herd (flock) to processing for milk foodstuffs.

#### REFERENCES

- AGABRIEL, C. COULON, J.B. MARTHY, G. CHENEAU, N.: Facteurs de variation du taux protéique du lait de vache. (Factors influencing proteins in cow milk.) INRA Productions Animales, 3, 1990: 137–150. (in French)
- AGABRIEL, C. COULON, J.B. MARTHY, G.: Facteurs de variations du rapport des teneurs en matiéres grasses et protéiques du lait de vache. (Factors influencing fat and protein content in cow milk.) INRA Productions Animales, *4*, 1991: 141–149. (in French)
- ANDERSSON, L.: Concentrations of blood and milk ketone bodies, blood isopropanol and plasma glucose in dairy cows in relation to the degree of hyperketonaemia and clinical signs. Zentralblatt fur Veterinarmedizin A, 31, 1984: 683–693.
- ANDERSSON, L.: Subclinical ketosis in dairy cows. Veterinary Clinical of North America: Food Animal Practice, *4*, 1988: 233–251.
- ANDERSSON, L. LUNDSTRÖM, K.: Milk and blood ketone bodies, blood isopropanol and plasma glucose in dairy cows, methodical studies and diurnal variations. Zentralblatt für Veterinarmedizin A, 31, 1984a: 340–349.
- ANDERSSON, L. LUNDSTRÖM, K.: Effect of energy balance on plasma glucose and ketone bodies in blood and milk and influence of hyperketonaemia on milk production of postparturient dairy cows. Zentralblatt für Veterinarmedizin A, 31, 1984b: 539–547.
- ANDERSSON, L. EMANUELSON, U.: An epidemiological study of hyperketonaemia in Swedish dairy cows: determinatnts and the relation to fertility. Preventive Veterinary Medicine, 3, 1985: 449.
- BAKER, L.D. FERGUSSON, J.D. CHALUPA, W.: Response in urea and true protein of milk to different protein feeding schemes for dairy cows. Journal of Dairy Science, 78, 1985: 2424–2434.
- BÍRO, D. LABUDA, J. CABADAJOVÁ, M.: Factors influencing cow milk production and fat to protein content ratio. Živočišná výroba (Czech Journal of Animal Science), 37, 1992: 521–528. (in Slovak)
- BRAGHIERI, A. PACELLI, C. VERDONE, M. GI-ROLAMI, A. – NAPOLITANO, F.: Effect of grazing and homeopathy on milk production and immunity of Merino derived ewes. Small Ruminant Research., 69, 2007: 95–102.
- BUTLER, W.R. CALAMAN, J.J. BEAM, S.W.: Plasma and milk urea nitrogen in relation to pregnancy rate in lactating dairy cattle. Journal of Animal Science, 74, 1996: 858–865.
- CARRIER, J. STEWART, S. GODDEN, S. FETROW, J.
   RAPNICKI, P.: Evaluation and use of three cowside tests for detection of subclinical ketosis in early postpartum cows. Journal of Dairy Science, *87*, 2004: 3725–3735.
- CSN 57 0530: Methods for testing of milk and milk products. Prague, Czech Republic, CNI Prague, 1973. (in Czech)
- DELGADO-PETRÍŇEZ, M. GUTMÁN-GUERRERO, J.L. MENA, Y. – CASTEL, J.M. – GONZÁLEZ-REDONDO, P. –

CARAVACA, F.P.: Influence of kid rearing system on milk yield, kid growth and cost of Florida dairy goats. Small Ruminant Research, *81*, 2009: 105–111.

- EN ISO/IEC 17025: Conformity assessment General requirements for the competence of testing and calibration laboratories. CNI Prague, 2005. (In Czech)
- EMANUELSON, U. ANDERSSON, L.: Genetic variation in milk aceton in Swedish dairy cows. Journal of Veterinary Medicine Series A, 33, 1986: 600–608.
- EMERY, R.S. BURG, N. BROWN, L.D.: Detection, occurrence and prophylactic treatment of borderline ketosis with propylene glycol feeding. Journal of Dairy Science, 47, 1964: 1074.
- ENJALBERT, F. NICOT, M.C. BAYOURTHE, C. MON-COULON, R.: Ketone bodies in milk and blood of dairy cows: Relationship between concentrations and utilization for detection of subclinical ketosis. Journal of Dairy Science, 84, 2001: 583–589.
- FÜRLL, M. DECKERT, W. SCHÄFER, M. WEHLITZ, A.: Lipolyse und Ketogenese bei Milchkühen - Beobachtungen im Laktationsverlauf. (Lipolysis and ketogenesis in dairy cows – observations during lactation course.) Monatshefte für Veterinar-Medizin, 47, 1992: 119–124. (in German)
- GARCIA, A. LINN, J.: Let's keep milk urea test results in perspective. Hoard's Dairyman, 25, 1992: 597.
- GASTEINER, J.: Ketose, die bedeutendste Stoffwechselerkrankung der Milchkuh. (Ketosis as most considerable metabolism disorder in dairy cows.) In: BAL Gumpenstein Bericht, 27. Viehwirtschaftliche Fachtagung, Juni, 2000: pp. 11–18.
- GASTEINER, J.: Einsatz glukoplastischer Verbindungen in der Milchviehfütterung. (Use of glucoplastic matters in dairy cattle feeding.) In: BAL Gumpenstein Bericht, 30. Viehwirtschaftliche Fachtagung, April, 2003: pp. 61–63. (in German)
- GEISHAUSER, T. LESLIE, K.E. DUFFIELD, T. EDGE, V.: An evaluation of milk ketone tests for the prediction of left displaced abomasum in dairy cows. Journal of Dairy Science, 80, 1997: 3188–3192.
- GEISHAUSER, T. LESLIE, K.E. DUFFIELD, T. SAN-DALS, D. – EDGE, V.: The association between selected metabolic parameters and left abomasal displacement in dairy cows. Journal of Veterinary Medicine Series A, 45, 1998: 499–511.
- GEISHAUSER, T. ZIEBELL, K.L.: Fett/Eiweiss-Quotient in der Milch von Rinderherden mit Vorkommen von Labmagenverlagerungen. (Fat/protein coefficient in milk of cattle herds with abomasal displacement.) Deutsche Tierärztliche Wochenschrift, *102*, 1995: 469–494. (in German)
- GENČUROVÁ, V. HANUŠ, O. HULOVÁ, I. VYLETĚLOVÁ, M. – JEDELSKÁ, R.: The differences of selected indicators of raw milk composition and properties between small ruminants and cows in the Czech Republic. Výzkum v chovu skotu/Cattle Research, L, 2008a: 10–19.

- GENČUROVÁ, V. HANUŠ, O. VYLETĚLOVÁ, M. LANDOVÁ, H. – JEDELSKÁ, R.: The relationships between goat and cow milk freezing point, milk composition and properties. Scientia Agriculturae Bohemica, 39, 2008b: 324–328.
- GRAVERT, H.O. JENSEN, E. HEFAZIAN, H. PABST, K. – SCHULTE–COERNE, H.: Umweltbedingte und genetische Einflüsse auf den Acetongehalt der Milch. (Environmental and genetic impacts on milk acetone content.) Züchtungskunde, 63, 1, 1991: 42–50 (in German).
- GRAVERT, H.O. LANGER, R. DIEKMANN, L. PABST, K. – SCHULTE–COERNE, H.: Ketonkörper in Milch als Indikatoren für die Energiebilanz der Milchkühe. (Ketone bodies in milk as indicators for energy balance in dairy cows.) Züchtungskunde, 58, 1986: 309–318. (in German)
- GREEN, B.L. McBRIDGE, B.W. SANDALS, D. LESLIE, K.E. – BAGG, R. – DICK, P.: The impact of a Monensin controlled – release capsule on subclinical ketosis in the transition dairy cow. Journal of Dairy Science, 83, 1999: 333–342.
- GUSTAFSSON, A.H. EMANUELSON, U.: Milk acetone as indicator of hyperketonaemia in dairy cows – the critical value revised. EAAP Congress Aarhus, Denmark, 1993: 443.
- HANSEN, P.W.: Screening of dairy cows for ketosis by use of infrared spectroscopy and multivariate calibration. Journal of Dairy Science, *82*, 1999: 2005–2010.
- HANUŠ, O. BEBER, K. FICNAR, J. GENČUROVÁ, V. GABRIEL, B. BERANOVÁ, A.: Relationship between the fermentation of bulk milk sample, its composition and contents of some metabolites. Živočišná výroba (Czech Journal of Animal Science), 38, 1993a: 635–644. (in Czech)
- HANUŠ, O. BJELKA, M. TICHÁČEK, A. JEDELSKÁ, R. – KOPECKÝ, J.: Substantiation and usefulness of transformations in data sets of analyzed milk parameters. In: Proceedings: VÚCHS Rapotín, 2001a: pp. 122–137. (in Czech)
- HANUŠ, O. FRELICH, J. JANŮ, L. MACEK, A. ZAJÍČKOVÁ, I. – GENČUROVÁ, V. – JEDELSKÁ, R.: Impact of different milk yields of cows on milk quality in Bohemian spotted cattle. Acta Veterinaria Brno, 76, 2007: 563–571.
- HANUŠ, O. FRELICH, J. KRON, V. ŘÍHA, J. POZDÍŠEK, J.: The checking of the body condition score, health state and nutrition of dairy cows and improvement of their reproduction functions. Prague, Czech Republic. Zemědělské informace, UZPI Praha, 2004a. (in Czech)
- HANUŠ, O. GENČUROVÁ, V. FICNAR, J. GABRIEL, B. – ŽVÁČKOVÁ, I.: The relationship of urea and protein in bulk milk to some breeding factors. Živočišná výroba (Czech Journal of Animal Science), 38, 1993b, 61–72. (in Czech)
- HANUŠ, O. GENČUROVÁ, V. PONÍŽIL, A. HLÁSNÝ, K. – GABRIEL, B. – MÍČOVÁ, Z.: The effects of year season, urea, acetone and nitrate additions and native content of microelements on cow's milk fermentation. Živočišná výroba (Czech Journal of Animal Science), 38, 1993c: 753–762.

- HANUŠ, O. GENČUROVÁ, V. VYLETĚLOVÁ, M. LANDOVÁ, H. – JEDELSKÁ, R. – KOPECKÝ, J.: The comparison of relationships between milk indicators in different species of ruminants in the Czech Republic. Výzkum v chovu skotu (Cattle Research), L, 2008: 35–44.
- HANUŠ, O. ŘÍHA, J. GENČUROVÁ, V. JEDELSKÁ, R. – KOPECKÝ, J.: Composition and quality of goat's milk, relationships of specific parameters and effects of some management factors in less favourable areas. Výzkum v chovu skotu /Cattle Research, XLVI, 2004b: 6–19.(in Czech)
- HANUŠ, O. TRAJLINEK, J. HLÁSNÝ, J. GENČUROVÁ, V. – KOPECKÝ, J. – JEDELSKÁ, R.: Ketosis problem, its diagnostic and monitoring. In: Rearing and breeding of cattle for competition production: proceedings of the seminar VÚCHS Rapotín, 2001b: 105–113. (in Czech)
- HANUŠ, O.: Annual report of research project NAZV QE 0040 (Research Institute for Cattle Breeding), Prague, Czech Republic, 2005.
- HARASZTI, J. ZÖLDAG, L.: Die diagnostische Rolle der Ketonurie in der Vorhersage von Fortpflanzungsstörungen der Kühe. (The diagnostic importance of ketonuria for the prognosis of reproductive disorders in dairy cows.) Wiener Tierärztliche Monatschrift., 77, 1990: 377–380. (in German)
- HEUER, C. LUINGE, H.J. LUTZ, E.T.G. SCHUKKEN, Y.H. – VAN DERMAAS, J.H. – WILMINK, H. – NOORD-HUIZEN, J.P.T.M.: Determination of acetone in cow milk by Fourier transform infrared spectroscopy for the detection of subclinical ketosis. Journal of Dairy Science, *83*, 2000: 575–582.
- HEUER, C. VAN STRAALEN, W.M. SCHUKKEN, Y.H.: Prediction of energy balance in high yielding dairy cows with test-day information. Journal of Dairy Science, 84, 2001: 471–481.
- HOJMAN, D. KROLL, O. ADIN, G. GIPS, M. HANO-CHI, B. – EZRA, E.: Relationships between milk urea and production, nutrition and fertility traits in Israeli dairy herds. Journal of Dairy Science, 87, 2004: 1001–1011.
- JANŮ, L. HANUŠ, O. FRELICH, J. MACEK, A. ZAJÍČKOVÁ, I. – GENČUROVÁ, V. – JEDELSKÁ, R.: Influences of different milk yields of Holstein cows on milk quality indicators in the Czech Republic. Acta Veterinaria Brno, 76, 2007: 553–561.
- JOHNSON, R.G. YOUNG, A.J.: The association between milk urea nitrogen and DHI production variables in vestern commercial dairy herds. Journal of Dairy Science, *86*, 2003: 3008–3015.
- KAUPPINEN, K.: Annual milk yield and reproductive performance of ketotic and non-ketotic dairy cows. Zentralblatt für Veterinarmedizin A, 31, 1984: 694–704.
- KIRCHGESSNER, M. KREUZER, M. ROTH-MAIER, D.A.: Milk urea and protein content to diagnose energy and protein malnutrition of dairy cows. Archives of Animal Nutrition, 36, 1986: 192–197.
- KIRCHGESSNER, M. ROTH-MAIER, D.A.– RÖHRMOSER, G.: Harnstoffgehalt in Milch von Kühen mit Energie-

bzw. Proteinmangel und anschliessender Realimentation. Zeitschrift für Tierphysiologie. (Urea contents in milk of cows with energy and protein restriction and subsequent realimentation.) Tiernährung und Futtermittelkunde, *53*, 1985: 264–270. (in German)

- KUCHTÍK, J. SEDLÁČKOVÁ, H.: Composition and properties of milk White Short-haired goats on the third lactation. Czech Journal of Animal Science, 48, 2003: 540–550.
- MELOUN, M. MILITKÝ, J.: Statistical processing of experimental data by personal computer. Part IIA, Pardubice, Czech Republic, 1992: 102. (in Czech)
- MELOUN, M. MILITKÝ, J.: Statistical processing of experimental data. Prague, Czech Republic. Plus 1994: 839. (in Czech)
- MIETTINEN, P.V.A.: Relationship between milk acetone and milk yield in individual cows. Journal of Veterinary Medicine A, *41*, 1994: 102–109.
- MIETTINEN, P.V.A.: Prevention of bovine ketosis with glucogenic substance and its effect on fertility in Finnish dairy cows. Berliner und Münchener Tierärztliche Wochenschrift, *108*, 1995: 14–19.
- MIGLIOR, F. SEWALEM, A. JAMROZIK, J. LEFEBVRE, D.M. – MOORES, R.K.: Analysis of milk urea nitrogen and lactose and their effect on longevity in Canadian dairy cattle. Journal of Dairy Science, *89*, 2006: 4886–4894.
- O'MOORE, L.B.: A quantitative diffusion method for the detection of acetone in milk. Veterinary Research, *31*, 1949: 457–458.
- MOTTRAM, T. VELASCO–GARCIA, M. BERRY, P. RICHARDS, P. – GHESQUIERE, J. – MASSON, L.: Automatic on-line analysis of milk constituents (urea, ketones, enzymes and hormones) using biosensors. Comparative Clinical Pathology, *11*, 2002: 50–58.
- RAJALA-SCHULTZ, P.J. SAVILLE, W.J.A. FRAZER, G.S.
  WITTUM, T.E.: Association between milk urea nitrogen and fertility in Ohio dairy cows. Journal of Dairy Science, 84, 2001: 482–489.
- ROOS DE, A.P.W. BIJGAART VAN DEN, H.J.C.M. HOR-LYK, L. – JONG DE, G.: Screening for subclinical ketosis in dairy cattle by Fourier transform infrared spectrometry. Final programme and Technical presentations, 35<sup>th</sup> ICAR Session, Kuopio, Finland, 3<sup>rd</sup> – 10<sup>th</sup> June, 2006: 53–59.
- ŘÍHA, J. HANUŠ, O.: The relationships between individual reproduction parameters and milk composition in first 120 lactation days. Výzkum v chovu skotu /Cattle Research, *XLI*, 1999: 3–17. (in Czech)
- SCHULZ, T.: Ohne formeln und Tabellen die Leistung gesteigert. (Without patterns and tables milk yield to improve.) Top Agrar, *5*, 1997: 20–22. (in German)
- STEGER, H. GIRSCHEWSKI, H. PIATKOWSKI, B.: Die Beurteilung des Ketosisstatus laktierender Rinder aus der Konzentration der Ketokörper im Blut und des Azetons in der Milch. (Ketosis state evaluation in cattle during lactation according to concentration of ketone bodies in blood

and acetone in milk.) Archiv für Tierernährung, 22, 1972: 157–162. (in German)

- STELLA, A.V. PARATTE, R. VALNEGRI, L. CIGALINO, G. – SONCINI, G. – CHEVAUX, E. – DELL'ORTO, V. – SAVOINI, G.: Effect of administration of live Saccharomyces cerevisiae on milk production, milk composition, blood metabolites, and faecal flora in early lactating dairy goats. Small Ruminant Research, 67, 2007: 7–13.
- TEDESCO, D. TAVA, A. GALLETTI, S. TAMENI, M. – VARISCO, G. – COSTA, A. – STEIDLER, S.: Effects of sylimarin, a natural hepatoprotector, in periparturient dairy cows. Journal of Dairy Science, 87, 2004: 2239–2247.
- VOJTÍŠEK, B.: The determination of acetone in milk, colostrum, blood and urine of dairy cows by microdiffusion method. Veterinářství, 36, 1986: 394–396. (in Czech)

- WOOD, G.M. BOETTCHER, D.F. KELTON, D.F. JANS-EN, G.B.: Phenotypic and genetic influences on test-day measures of acetone concentration in milk. Journal of Dairy Science, 87, 2004: 1108–1114.
- ZENG, S.S.: Comparison of goat milk standards with cow milk standards for analyses of somatic cell count, fat and protein in goat milk. Small Ruminant Research, *21*, 1996: 221–225.
- ZENG, S.S. ESCOBAR, E.N.: Effect of breed and milking method on somatic cell count, standard plate count and composition of goat milk. Small Ruminant Research, 19, 1996: 169–175.
- ZENG, S.S. ESCOBAR, E.N. POPHAM, T.: Daily variations in somatic cell count, composition, and production of Alpine goat milk. Small Ruminant Research, 26, 1997: 253–260.

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### Vztahy některých mléčných ukazatelů energetického metabolismu v kravském, kozím a ovčím mléce

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Mléčné ukazatele energetického metabolismu (bílkoviny (CP), tuk (F), aceton (AC)) významně ovlivňují kvalitu mléka. Jejich zhoršené hodnoty poukazují na poruchy plodnosti zvířat. Mohou být využity pro monitoring a prevenci negativní energetické bilance zvířat v počátku laktace. Tak lze zlepšovat zdraví zvířat a kvalitu mléka. Byly studovány vztahy mezi ukazateli energetického metabolismu (AC a poměr tuk/bílkoviny (F/CP)) v bazénovém mléce kravském (Czech Fleckvieh (CF), n = 40; Holstein (HN), n = 36), kozím (White short-haired (WS), n = 40) a ovčím (Tsigai (TS), n = 40). Vzorky pocházely z první poloviny laktace. Krávy byly krmeny směsnou krmnou dávkou (TMR) na bázi kukuřičné, vojtěškové a jetelové siláže s pivovarským mlátem a koncentráty. Kozy a ovce byly paseny na polokulturním pastevním porostu a přikrmeny koncentráty. Průměrné rozdíly CP, F, F/CP a AC byly mezi plemeny a druhy významné (P < 0.05). Nejvyšší AC (12,68  $(8,73 \text{ xg}) \text{ mg.}^{-1})$  byl u TS. Variabilita F/CP byla poměrně nízká a u AC naopak vysoká, jak je typické pro zdravotní ukazatele, od 54,5% (HN) do 98,9% (WS). Zatímco průměry AC (TS, CF 7,77 (6,34 xg) a HN 2,03 (1,60 xg) a WS 7,55 (3,50 xg) mg.l<sup>-1</sup>) lze považovat za druhově specifické, nemusí tomu tak být u jejich variability. Byly nalezeny pozitivní vztahy mezi AC a F/CP u krav CF a HN (r = 0.34, P < 0.05; r = 0.15, P > 0.05) a u ovce (r = 0.40, P < 0.05). Negativní vztah byl překvapivě v kozím mléce (r = -0.22, P > 0.05). Uvedené indikuje dobrou vypovídací schopnost pro oba ukazatele energetického metabolismu nejen v kravském, ale také ovčím mléce. V kozím mléce jsou třeba další studie. Poměr F/CP je dostupný z rutinní kontroly mléčné užitkovosti, na rozdíl od méně častého vyšetření na AC.

dojnice; koza; ovce; bazénový vzorek mléka; ukazatel energetického metabolismu; aceton; poměr tuk/hrubé bílkoviny

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