

BODY CONDITION SCORE AND MILK FATTY ACID COMPOSITION IN EARLY LACTATION OF CZECH FLECKVIEH COWS

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Abstract

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The development of body condition score (BCS) and milk fatty acid composition and its mutual relationships were evaluated in the first 4 weeks of lactation of 50 Czech Fleckvieh cows. Average BCS values ranged from 4.14 at calving to 3.6 points in the 4th week of lactation. The percentage of saturated fatty acids (SFA) tended to slightly increase (69.1 to 71.3%), whereas the percentage of unsaturated fatty acids (UFA) tended to decrease (30.9 to 28.7%). The animals used were divided to three groups according to the BCS change between calving and week 4 of lactation. The cows with a minimum BCS loss during the first 4 weeks postpartum (–0.03 points) showed higher (+2.16 to 5.24%; $P < 0.05$) SFA proportions than those having BCS loss more than 1 point. An opposite tendency was observed for UFA, with the highest (+1.23 to 5.26%; $P < 0.05$) proportion found in the animals with the greatest BCS loss (≥ 1 point). These results indicate that high proportions of milk UFA related with a deep negative energy balance expressed by the highest BCS loss (≥ 1 point). The correlations between BCS at calving, BCS in the 1st and 2nd week of lactation, and proportion of fatty acid groups determined in the 4th week ranged from $r = 0.29$ to $r = 0.41$ ($P < 0.05$).

milk fat, dairy cow, body condition, fatty acid, Czech Fleckvieh

The intensity of cows' metabolism changes considerably in early lactation. However, it is important to monitor its changes during the entire transition period (Adamski *et al.*, 2011). Body condition score (BCS) is the 2nd most frequent indicator used for evaluation of the nutritional and health status of cows. It is a source of valuable information about the level of body energy supply correlated with milk performance, reproduction, and health of cows (Bastin *et al.*, 2007). A higher BCS at calving is associated with increased milk fat contents in early lactation (Roche *et al.*, 2009). However, these effects of BCS at calving were not confirmed in a study by Jílek *et al.* (2008), but the level of BCS in the first month of lactation was shown to be associated with fertility traits in cattle. BCS changes during lactation are more important

than its current status (Roche *et al.*, 2009). Therefore, it is necessary to minimize BCS losses during the first weeks of lactation to maintain an adequate level of milk and reproduction performance (Roche *et al.*, 2009).

Milk composition is affected by a number of different factors like genetics (Hanuš *et al.*, 2011), nutrition (Dai *et al.*, 2011), and the energy status of cows (Friggens *et al.*, 2007). The content of milk fat (F) in the first 2 months of lactation may represent a better indicator of negative energy balance (NEB) than the content of protein (P) (De Vries and Veerkamp, 2000). A milk F/P ratio was also previously recommended as a suitable NEB indicator (Heuer *et al.*, 1999). Optimum F/P values range from 1.2 to 1.4, whereas values outside this range indicate a higher risk of health problems

(Čejna and Chládek, 2005). Low F/P values are associated with a higher incidence of acidosis, whereas high values (> 1.5) considerably increase the risk of ketosis, ovarian cysts (Heuer *et al.*, 1999), and mastitis (Winding *et al.*, 2005).

Most of milk fat (97 to 98%) is composed of glycerol and fatty acids (FA) esters (Jensen, 2002). A number of studies have been previously aimed at the content and composition of FA in bovine milk (Ducháček *et al.*, 2011) and beef (Bartoň *et al.*, 2010). The content and composition of milk FA are influenced by nutrition (Dai *et al.*, 2011), breed, lactation stage, parity, season (Barłowska *et al.*, 2009), and cows' individuality (Samková *et al.*, 2008). Short- and medium-chain FA are *de novo* synthesised in the mammary gland, whereas long-chain FA are transported to the mammary gland by the blood in the form of non-esterified FA originating directly from the diet (Harvatine *et al.*, 2009), or released from adipose tissue (Samková *et al.*, 2008). During lactation, less than 10% of milk FA are mobilized from the fat depots except the period of NEB, when this proportion is considerably higher (Bauman and Griinari, 2001). Generally, there are several main FA groups: saturated (SFA), unsaturated (UFA), monounsaturated (MUFA), and polyunsaturated FA (PUFA). SFA and UFA comprise 67.11 to 70.76% and 25.69 to 32.68% of the total milk FA, respectively (Schennink *et al.*, 2009). Lock and Shingfield (2004) reported the average proportions of milk MUFA and PUFA as being 20 to 25% and 5%, respectively. Significantly higher SFA and lower UFA contents in milk were observed in winter compared to summer (Barłowska *et al.*, 2009).

The greatest changes in the content and composition of milk FA occur in early lactation, when cows may suffer from NEB (Ducháček *et al.*, 2012). There is an increased synthesis of FA $C_{4:0}$ – $C_{12:0}$ in this period (Garnsworthy *et al.*, 2006). The highest contents of $C_{18:0}$ and $C_{18:1}$ were found at the beginning of lactation (Bear, 1991). Komprda *et al.* (2001) reported that the content of $C_{14:0}$ and $C_{18:0}$ markedly increased in the first phase of lactation, whereas the contents of FA with carbon numbers up to 13 remained similar. Cows in NEB mobilize their fat reserves and therefore, the percentages of $C_{16:0}$ and $C_{18:0}$ increase (Stoop *et al.*, 2009). The content of $C_{18:1}$ in early lactation is high due to the increased absorption of FA – the products of adipose tissue hydrolysis – from the blood (Bauman and Griinari, 2003). The content of SFA increased whereas MUFA and PUFA decreased by day 90 of lactation, which indicated the compensation of NEB (Komprda *et al.*, 2005). However, Van Knegsel *et al.* (2007) reported that cows producing more UFA are less susceptible to severe NEB.

Study of the literature revealed that NEB influences the BCS, F content, F/P ratio, and FA content in early lactation. If the total content of milk fat is affected, then the contents of individual FA and FA groups might be affected as well. Based on mentioned knowledge we hypothesize, that greater

BCS change as a result of more intensive NEB is also reflected in FA composition changes in the milk fat. The objective of this study was to evaluate the relationships among BCS changes and milk FA contents as indicators of Czech Fleckvieh cows NEB in the first 4 weeks after calving.

MATERIAL AND METHODS

A total of 50 Czech Fleckvieh cows were included in the experiment analysis – 10, 16, 15, and 9 in the first, second, third, and fourth and subsequent lactation, respectively. All animals were bred at one herd and calved within the period of 40 days (October to November). The average daily milk yield ranged from 25.7 to 28.6 l of milk, with the standard deviation ranging from 6.26 to 6.64 l. The cows were loose housed in a cubicle straw-bedded barn and fed a total mixed ratio (TMR). The ingredient composition of the diet corresponded to the level of the daily milk yield. BCS was evaluated at calving and subsequently weekly to the 4th week postpartum (a total of 5 times; BCS₀, BCS₁, BCS₂, BCS₃, BCS₄) on a 5-point scale with 0.25-point increments (Ferguson *et al.*, 1994). For further evaluation of the effect of BCS changes on FA composition, the animals were assigned to three groups based on the BCS loss in the first month of lactation: A – BCS loss ≥ 1 points ($n = 13$); B – BCS loss from 0.5 to 0.75 points ($n = 21$); C – BCS loss ≤ 0.25 points ($n = 16$). Proportional milk samples from each cow were collected from the afternoon milking in accordance with the ICAR methodology of milk performance recording at day 7, 14, 21, and 28 after calving. A total of 200 milk samples collected during the first 4 weeks of lactation were analyzed. The analysis of milk samples comprised the determination of basic components (% F, % P) and FA as well as FA group contents. Fat and protein contents were determined using Milcoscan 133B (N. Foss Electric; Denmark). When determining milk FA composition, firstly the milk fat was extracted using the standard Röss-Gottlieb method (gravimetric) in accordance with EN ISO 1211 (2002). The extract was obtained using a water-based solution of ammonia, ethanol, diethylether and petrolether. FA methyl esters were prepared by potassium hydroxide catalysed methylation and extracted into heptane. Gas chromatography of FA methyl esters was performed using the Master GC (DANI Instruments S.p.A.; Italy) (split regime, FID detector) on a column with polyethylene glycol stationary phase (FameWax – 30 mm \times 0.32 mm \times 0.25 μ m). Helium was used as the carrier gas at a flow rate of 5 ml/min. The temperature programme used for GC was as follows: 50 °C (2 min), after which the temperature was increased to 230 °C at 10 °C/min (8 min), the temperature of the detector being 220 °C. Gravimetric contents (mg/100g of milk) and percentages of the total FA were determined in 34 individual FA, SFA, UFA, MUFA, and PUFA (UFA = MUFA + PUFA). The data were evaluated

by the statistical software SAS 9.1. (SAS/STAT® 9.1., 2004) using MEANS, GLM and CORR procedures. The model equation used for the evaluation of the relationship between BCS and FA group contents was as follows:

$$Y_{ij} = \mu + BCS_{4-0i} + e_{ij},$$

where:

Y_{ij} dependent variable (SFA, UFA, MUFA, and PUFA content);

μ mean of the dependent variable;

BCS_{4-0i} fixed effect of the i^{th} group in accordance with the BCS change between calving and the 4th week of lactation ($i = BCS_{4-0} \geq -1$ point, $n = 13$; $BCS_{4-0} -0.5$ to -0.75 points, $n = 21$; $BCS_{4-0} \leq -0.25$, $n = 16$);

e_{ij} random residual error.

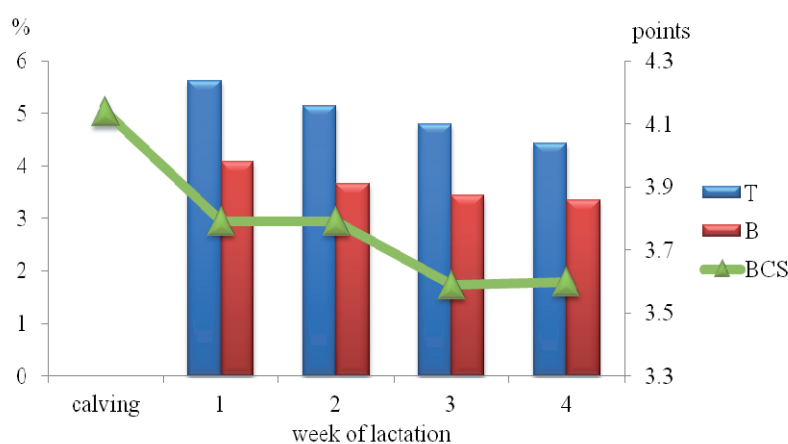
The significance of differences was assessed using the t-test method, and the probability level $P < 0.05$ was considered as the statistically significant.

RESULTS AND DISCUSSION

The Czech Fleckvieh cows were in NEB during observed lactation period as evidenced by the loss of BCS. The greatest BCS changes occurred at the very beginning of lactation. Different lengths of BCS loss periods are reported in various studies. Maršálek *et al.* (2008) observed the most important BCS changes up to month 3 of lactation. In our study, particularly high BCS values (average 4.14 points) were detected at the beginning of lactation. Jílek *et al.* (2008) recommended higher BCS for Czech Fleckvieh cows at calving, since they observed a positive effect of BCS higher than 3.5 points in the first month after calving on reproduction parameters and milk yield. In our study, we

I: Basic statistics for fat and protein contents, milk yield, and body condition score

Variable	Lactation week	N	\bar{x}	s_d
Fat (%)	1	47	5.63	0.94
	2	46	5.14	0.77
	3	50	4.8	0.82
	4	48	4.42	0.91
Protein (%)	1	50	4.08	0.33
	2	49	3.66	0.33
	3	50	3.44	0.26
	4	50	3.35	0.26
Milk yield (kg)	1	38	25.7	6.34
	2	47	27.37	6.47
	3	41	27.99	6.7
	4	45	28.61	6.72
BCS (point)	0	50	4.14	0.42
	1	49	3.79	0.46
	2	50	3.79	0.62
	3	50	3.59	0.45
	4	41	3.6	0.51



1: Development of body condition score (BCS) and milk fat and protein contents

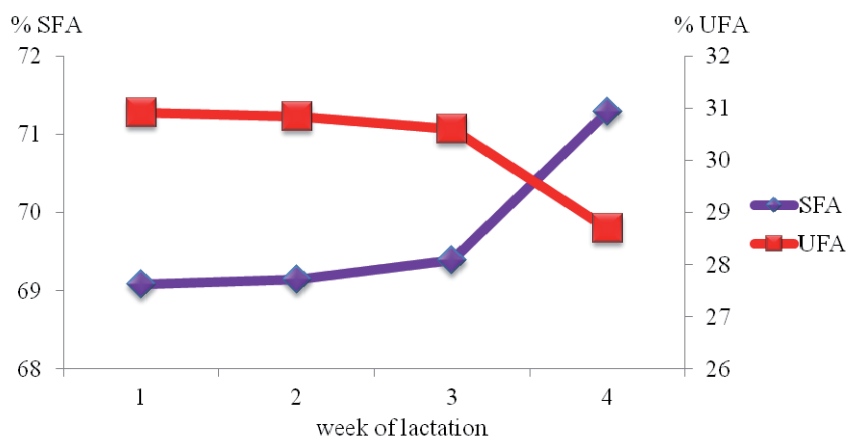
observed a considerable BCS change from 4.14 to 3.6 points during the entire first month of lactation (Tab. I., Fig. 1).

The onset of the lipomobilization syndrome is associated with the hydrolysis of subcutaneous fat and it is reflected in milk composition during the reduction of BCS (Contreras *et al.*, 2010), especially in the content and composition of milk fat. Basic statistics of milk composition (% F, % P, daily milk yield) and BCS in monitored cows are given in Tab. I. All the variables were quite homogeneous, as evidenced by standard deviation (s_d). During the period observed, a marked decrease was demonstrated in F (from 5.63% in week 1 to 4.42% in week 4) and P (from 4.08% in week 1 to 3.35% in week 4). Fig. 1 shows the development of the average % F, % P, and BCS. The optimum F/P ratio values ranged from 1.2 to 1.4, whereas higher values indicate NEB (Čejna and Chládek, 2005). The F/P ratios found in this study ranged from 1.31 in week 4 to 1.41 in week 2 of lactation, which is the value evidencing energy deficiency (NEB). The F/P ratio observed in week 1 was also high (1.38) and approached the critical value for NEB. However, this value detected was not the highest, which may be explained by the gradually increasing NEB related

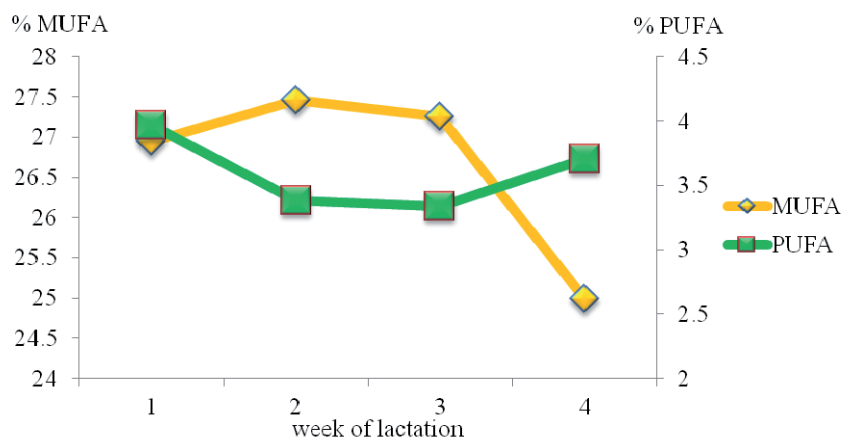
to growing daily milk yields during the first lactation week. The F/P ratios indicate the presence of NEB and the lipomobilization of fat reserves in cows. Our results are in agreement with those reported by Bergk and Swalve (2011).

Percentages content of 34 FA and 4 FA groups (SFA, UFA, MUFA, and PUFA) were determined in milk samples. As all the cows used in our study calved within a period of 40 days, the effect of season on SFA and UFA contents (Barłowska *et al.*, 2009) was eliminated. The average SFA and UFA proportions ranged from 69.08 to 71.29% and from 30.92 to 28.71%, respectively, which is in agreement with a study by Schennink *et al.* (2009). The proportions of MUFA and PUFA ranged from 24.99 to 27.45% and from 3.33 to 3.97%, respectively. Thus, the proportions of MUFA were by 2.5% higher and the proportions of PUFA by 1% lower than those reported by Lock and Shingfield (2004). The trends of SFA, UFA, MUFA, and PUFA changes are demonstrated in Fig. 2 and 3.

SFA proportions slightly increased during the period analysed, whereas UFA proportions decreased (Fig. 2). MUFA decreased especially in weeks 3 and 4 (Fig. 3), which may have been associated with the recovery of energy balance



2: Development of SFA and UFA proportion (%)



3: Development of MUFA and PUFA proportion (%)

II: Mean BCS in different cow groups and different lactation weeks

Group	N	BCS 0	BCS 1	BCS 2	BCS 3	BCS 4	BCS ₄₋₀
(≥ -1 point)	13	4.37	3.77	3.69	3.5	3.15	-1.21
(-0.5 to -0.75)	21	4.15	3.7	3.63	3.56	3.49	-0.67
(≤ -0.25)	16	3.94	3.92	3.83	3.70	3.91	-0.03

BCS 0 – body condition score at calving; BCS 1-4 – body condition score in week 1, 2, 3 and 4 of lactation; BCS₄₋₀ – difference between the body condition score at calving and week 4 of lactation.

III: Fatty acid composition % (LSM ± SE) in the milk from cows with different BCS changes

	BCS ₄₋₀	N	Week of lactation			
			I.	II.	III.	IV.
SFA	-1.21	13	69.07 ± 1.3 ^a	71.19 ± 1.45	67.99 ± 1.08 ^a	71.29 ± 1.12 ^a
	-0.67	21	68.57 ± 1.5 ^a	68.74 ± 1.67	71.17 ± 1.25	72.78 ± 1.3
	-0.03	16	73.55 ± 1.5 ^b	73.35 ± 1.67	73.23 ± 1.25 ^b	75.49 ± 1.3 ^b
UFA	-1.21	13	30.93 ± 1.3 ^a	28.8 ± 1.46	32 ± 1.08 ^a	28.68 ± 1.13 ^a
	-0.67	21	31.43 ± 1.5 ^a	31.26 ± 1.68	28.8 ± 1.25	27.19 ± 1.3
	-0.03	16	26.42 ± 1.5 ^b	26.57 ± 1.68	26.74 ± 1.25 ^b	24.48 ± 1.3 ^b
MUFA	-1.21	13	27.03 ± 1.41	25.53 ± 1.38	28.72 ± 1.06 ^a	25.6 ± 1.06 ^a
	-0.67	21	27.31 ± 1.63	28.14 ± 1.6 ^a	25.56 ± 1.22	23.85 ± 1.23
	-0.03	16	23.51 ± 1.63	23.33 ± 1.6 ^a	23.62 ± 1.22 ^b	21.15 ± 1.23 ^b
PUFA	-1.21	13	3.9 ± 0.25 ^a	3.27 ± 0.2	3.27 ± 0.12	3.08 ± 0.23
	-0.67	21	4.12 ± 0.29 ^a	3.12 ± 0.23	3.25 ± 0.14	3.34 ± 0.27
	-0.03	16	2.91 ± 0.29 ^b	3.24 ± 0.23	3.11 ± 0.14	3.32 ± 0.27

SFA – saturated fatty acids; UFA – unsaturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; BCS₄₋₀ – difference between the body condition score at calving and week 4 of lactation; a,b – P < 0.05, different superscript letters confirm statistical significance of difference among rows.

(Ducháček *et al.*, 2011). The tendency towards SFA increase and UFA decrease with continuing lactation has also been reported in previous studies (Pešek *et al.*, 2005; Samková *et al.*, 2008).

Group means for BCS in different lactation weeks (BCS0 to BCS4) as well as the change of BCS between calving and week 4 (BCS₄₋₀) are presented in Tab. II.

The lowest BCS decrease was associated with the highest SFA proportion in all 4 weeks of lactation. The differences in SFA proportions between the cows with the lowest BCS change (≤ -0.25 point) and the remaining groups were from +4.48 to +4.98% (P < 0.05) in week 1, from +2.16 to +4.61% in week 2, from +2.06 to 5.24% (P < 0.05) in week 3, and from +2.71 to +4.2% (P < 0.05) in week 4 (Tab. III).

Generally, greater extent of NEB (BCS loss ≥ 1 point) resulted in fluctuating SFA proportions compared to moderate NEB animals (BCS loss from 0.5 to 0.75 points) with higher milk SFA as early as week 1 of lactation (P < 0.05). Opposite trends were observed for UFA. The differences between groups ranged from -4.51 to -5.01% (P < 0.05) in week 1, from -2.23 to -4.69% in week 2, from -2.06 to -5.26% (P < 0.05) in week 3, and from -2.71 to -4.2% (P < 0.05) in week 4 (Tab. III). As with SFA, greater extent of NEB (BCS loss ≥ 1 point) resulted in fluctuating UFA proportions compared to moderate NEB animals (BCS loss from 0.5 to 0.75 points). Also, higher UFA in the first 2 weeks of lactation was observed in dams with a BCS loss from 0.5 to 0.75 points compared to

those with a BCS loss ≥ 1 point. These results lacked statistical significance, but they confirmed findings reported by Van Knegsel *et al.* (2007) that cows with moderately intensive NEB had higher UFA contents in milk in weeks 1 and 2 of lactation than animals with severe NEB (BCS loss ≥ 1 point). As studies by Samková *et al.* (2008) and Pešek *et al.* (2005) showed, UFA proportions decreased as early as week 3 in all the groups. We report that the greater the BCS loss in the first 4 weeks of lactation, the higher the milk UFA proportions were observed (P < 0.05). Thus, increased SFA and decreased UFA proportions are more closely related to the low intensity of NEB than to lactation weeks, as stated by Samková *et al.* (2008) and Pešek *et al.* (2005).

Within UFA, the most pronounced changes were observed in MUFA proportions. The differences in MUFA proportions between the animals with a BCS loss ≤ 0.25 and the remaining groups were from -3.52 to -3.8% in week 1, from -2.2 to -4.81% (P < 0.05) in week 2, from -1.94 to -4.81% (P < 0.05) in week 3, and from -2.7 to -4.45% (P < 0.05) in week 4. The trend observed in weeks 3 and 4 are similar to those found in UFA.

Unlike in SFA and UFA, no trends were detected for PUFA. Significant differences were observed only in week 1 of lactation. This may be explained by the relatively low PUFA proportions corresponding to those reported by Pešek *et al.* (2005). SFA and UFA appeared to be good indicators of NEB. Thus,

the proportions of SFA and UFA of 72% and 28%, respectively, may be considered as the threshold values for intensive NEB occurrence. These values also correspond to the entire lactation results as reported by Lock and Shingfield (2004).

In addition, correlations were calculated to evaluate the relationships between FA groups and BCS. Significant correlations ($P < 0.05$) were found between BCS after calving and FA group proportions in week 4 of lactation ($r = 0.3$ to 0.32). Moderate positive correlations ($P < 0.05$) were also detected between BCS determined in weeks 1 and 2 and FA group proportions in week 4 of lactation ($r = 0.29$ to 0.41). Furthermore, moderate negative correlations ($P < 0.05$) were calculated between FA group proportions in week 1 and BCS in week 4 of lactation ($r = -0.46$ to -0.29).

CONCLUSIONS

Body condition score as well as F and P percentages content in milk were reduced in Czech Fleckvieh cows during the first 4 weeks

of lactation. The F/P ratio in the first 3 weeks of lactation reached the threshold indicating the occurrence of NEB, whereas in week 4 this ratio decreased. The proportion of milk SFA tended to increase in the period observed (69.08 to 71.29%), whereas the opposite tendency was found for UFA (30.92 to 28.71%). Different trends in FA group proportions were detected when the animals were assigned to groups according to the BCS change between calving and week 4 of lactation. The milk from cows with a low BCS loss contained more SFA compared to those with a BCS loss ≥ 1 point. An opposite tendency was observed for UFA, which was significantly higher in animals with a BCS loss ≥ 1 point. Within UFA, similar tendencies were found for MUFA, but not for PUFA. The results confirmed the relationships between the BCS development of Czech Fleckvieh cows after calving and the composition of their milk. The parameters of milk composition can be used as an indicator of animal metabolic status as well as an indicator related to nutrition and food quality.

SUMMARY

The objective of this study was to evaluate the relationships among BCS changes and milk FA contents as indicators of Czech Fleckvieh cows NEB in the first 4 weeks after calving. A total of 50 Czech Fleckvieh cows were included in the experiment analysis – 10, 16, 15, and 9 in the first, second, third, and fourth and subsequent lactation, respectively. The BCS of cows was evaluated before calving and during the first four weeks of lactation. The animals observed were divided to three groups according to the BCS change between calving and week 4 of lactation. From proportional milk samples were analysed basic milk components (% F, % P). Gas chromatography was used for determination of FA groups in milk (saturated – SFA, unsaturated – UFA, monounsaturated – MUFA, polyunsaturated – PUFA). The data were evaluated with the statistical software SAS 9.1. (SAS/STAT 9.1., 2004) using the linear model with the fixed effect groups of BCS change.

Body condition score as well as F and P percentages content in milk were reduced in Czech Fleckvieh cows during the first 4 weeks of lactation. The percentage of SFA tended to slightly increase (69.1 to 71.3%), whereas the percentage of UFA tended to decrease (30.9 to 28.7%). The cows with a minimum BCS loss during the first 4 weeks postpartum (-0.03 points) exhibited higher ($+2.16$ to 5.24% ; $P < 0.05$) SFA proportions than those having BCS loss ≥ 1 point. An opposite tendency was observed for UFA, with the highest ($+1.23$ to 5.26% ; $P < 0.05$) proportion found in the animals with the greatest BCS loss (≥ 1 point). Within UFA, similar tendencies were found for MUFA, but not for PUFA. The high proportions of milk UFA is thus in relation with a deep negative energy balance and marked BCS loss. The correlations between BCS at calving, BCS in the 1st and 2nd weeks of lactation, and proportions of fatty acid groups determined in the 4th week ranged from $r = 0.29$ to $r = 0.41$ ($P < 0.05$). All the results confirmed the relationships between the BCS development of Czech Fleckvieh cows after calving and the composition of their milk.

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