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Joint effects of breed, parity, month of lactation, and cow individuality on the milk fatty acids composition

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Abstract

The aim of the study was to explain the effects of four animal factors - breed, parity, cow individuality (within breed phenotypic variation) and month of lactation on the composition of bovine milk fatty acids (FA) in a local dual-purpose Czech Fleckvieh breed as compared to the worldwide dairy Holstein breed. In total, 357 milk samples were analysed from 25 dairy cows of each breed during year-round testing. The variation in the individual FA was affected mainly by cows´ individuality (16-48) and month of lactation (3-18 %). The effects of breed and parity were limited (each about 2%). The animal related factors appeared significant also for FA groups. Greater differences in the explained variation of all factors were observed in the groups classified by the number of FA carbons (35.8, 54.4 and 44.8 % for C4 to C14, C16 and C18 to C24, respectively) and by the number of double bounds (45.4 % and 39.2 % for monounsaturated and polyunsaturated FA, respectively) as well. No differences in the explained variation were observed between the groups of saturated and unsaturated FA (46.8% and 45.9 %, respectively). In conclusion, from the viewpoint of nutrition it would be more convenient to classify FA by the number of carbons than by the usual grouping to saturated/unsaturated FA.

Key words: cow, milk, fatty acids, animal factors

Introduction

Bovine milk fat is often considered as not favorable to human health due to high content of saturated fatty acids (FA) (about 65 %) and *trans* isomers of unsaturated FA (about 5 %; Jensen, 2002). Both groups of FA are associated with cardiovascular diseases. As recent research shows, FA of animal origin are not as adverse as industrially produced *trans* fats (German et al., 2009). In addition, a relatively high proportion of oleic acid (about 18 %) occurs in bovine milk fat, which can contribute to a reduction of blood lipids (Lopez-Huertas, 2010). Also stearic acid at level of about 10% has beneficial biological effects (Kris-Etherton et al., 2005) in comparison to other saturated FA, like myristic and palmitic acid. Moreover, the milk fat contains in a small proportion (about 3 %) other nutritionally important FA like linoleic, α-linolenic and conjugated linoleic (CLA) acids (Lock and Baumann, 2004).

Researchers strived for decades to modify milk fat composition to achieve a higher proportion of nutritionally desirable FA (e.g. Kliem and Shingfield, 2016). Although, the FA composition is affected mainly by feeding (dietary) factors (Kalac and Samkova, 2010; Vranjes et al., 2010), animal (non-dietary) factors should not be ignored (Palmquist et al., 1993). Although all animal factors affecting milk FA composition have been

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previously reported, mostly one or two factors have been included in statistical models (e. g. Soyeurt et al., 2006; Poulsen et al., 2012). This approach may lead to partial misinterpretation of data, because of the mutual relations of each animal factor, both synergistic and antagonistic, need to be respected (Samkova et al., 2012). There is a limited number of published work that studied more than three factors. For example, De La Fuente et al. (2009) and Sojak et al. (2013) examined the variability of FA composition in ovine milk fat. Nevertheless, the results can hardly be compared to bovine milk fat due to the seasonal changes in small ruminants. The variability of FA composition in bovine milk fat taking into account more than two factors has been sporadically reported (Kelsey et al., 2003; Soyeurt et al., 2006; Schwendel et al., 2015).

Thus, the aim of this study was to determine the extent of variability, explaining joint effect of four tested animal factors (breed, parity, cow individuality and month of lactation) on the composition of bovine milk FA in dual-purpose Czech Fleckvieh and dairy Holstein breed.

Material and methods

Statement of institutional animal care

Ethical committee hereby declares that experiments performed in the present study are according Act No 246/1992 Coll., on the protection of animals against cruelty of the Czech Republic. With regard to the type of study, no special permission was required.

Sampling and feeding

The study was carried out on a farm (420 meters above sea level) located in the region of South Bohemia, Czech Republic. Thereat cows of two breeds, Czech Fleckvieh (dual-purpose) and Holstein (dairy) were housed together in one stable and were milked twice a day. Milk was sampled within the afternoon regular testing of milk efficiency twelve times a year.

Within each of 12 samplings, milk samples were taken according to breed, parity and month of lactation to get a balanced set of samples (Table 1 and 2). Twenty five cows of each breed were selected (30% of all lactating cows). From each cow 7 milk samples were obtained on average (5 to 10) during its lactation.

Table 1. Mean, standard deviation (SD), minimum, and maximum values for parity and days in milk of Czech Fleckvieh and Holstein cows

Item	Czech Fleckvieh (n = 188)			Holstein (n = 175)			P-value
	Mean±SD	Min.	Max.	Mean±SD	Min.	Max.	
Parity	2.03±0.79	1	4	2.07±0.76	1	5	0.0334
Days in milk	155±80	9	330	152±78	10	312	0.0646

P-value = probability

Table 2. Distribution of milk samples according to breed, parity and months of lactation

Months of lactation	Days in milk	Czech Fleckvieh				Holstein				Total
		Parity			Total	Parity			Total	
		1	2	≥3		1	2	≥3		
1	(<30)	2	4	5	11	2	3	5	10	21
2	(31-60)	4	6	5	15	4	5	6	15	30
3	(61-90)	6	6	8	20	5	6	8	19	39
4	(91-120)	6	8	6	20	5	9	7	21	41
5	(121-150)	6	9	7	22	5	9	6	20	42
6	(151-180)	6	9	8	23	6	9	7	22	45
7	(181-210)	6	7	6	19	5	8	6	19	38
8	(211-240)	6	7	5	18	4	8	4	16	34
9	(241-270)	6	8	5	19	5	9	4	18	37
10	(>271)	5	6	4	15	5	6	4	15	30
Total		53	70	59	182	46	72	57	175	357

Cows were fed under the same conditions during the whole year. Total mixed rations were formulated by the DLG-Futterwerttabellen, Wiederkäuer (1997) and calculated for the mean live weight 650 kg, milk fat content of 4.2 % and milk protein content of 3.5 %. Total mixed rations consisted of components widely used in the recent Czech farming practice - see Tables 3 and 4.

Analytical methods

Milk samples were immediately cooled after sampling and transported to the laboratory in a cool box. Milk samples were analysed for the fat, protein and lactose contents that were determined spectrophotometrically using a Milcoscan 4000 (Foss Electric, Hillerød, Denmark). Milk fat was extracted with petroleum ether from freeze dried milk samples. FA were re-esterified in isolated fat to their methyl esters by a methanolic solution of potassium hydroxide. Methyl esters of FA were determined by a gas-liquid chromatographic method (GLC) using an apparatus Varian 3300 (Varian Techtron, USA) under conditions according to Samkova et al. (2009).

The identification of FA was carried out using the analytical standards (Supelco, USA). In total, 45 FA were observed of which 33 were identified. The proportions of individual FA were calculated from the ratio of their peak area to the total area of all the observed acids.

Statistical analysis

The data were analyzed by Statistica CZ 6.1 (Statsoft CR) software using a general linear model with fixed effects of breed, parity and month of lactation and with random effect of cows nested in breed:

$$Y_{ijk} = \mu + B_i + I_j (B_i) + P_k + MOL_l + \varepsilon_{ijkl},$$

where

Y_{ijkl} = milk yield (kg/d), fat, protein and lactose content (g/100 g), proportion of individual milk FA (g/100g of FA), and groups of FA (g/100 g of FA); μ = mean; B_i = breed (i = Czech Fleckvieh, Holstein); I_j (B_i) = cow individuality (j = 1-50); P_k = parity (k = 1, 2, 3); MOL_l = month of lactation (l = 1-10; see Table 2), and ε_{ijkl} = residual error. For the groups comparison unequal N HSD test was used.

The total explained variance (coefficient of determination; R^2) and variance explained by four animal factors (factors variance) were calculated using sum of squares and were expressed in percent. R^2 was defined as [(1- (residual sum of squares/total sum of squares)) x 100], factors variance was defined as [(sum of squares of individual effects/total sum of squares) x 100].

Table 3. Components composition of average total mixed rations

Components composition	% of DM[1]
Maize silage	27.5
Grass silage	32.5
Hay	4.0
Mashed oats	6.0
Production mixture[2]	30.0

[1]DM = intake of dry matter was 18.4 kg/d.
[2]Production mixture composed of wheat, barley, extracted soybean meal, and salt, minerals and vitamins in proportion 32, 32, 32, and 4 %, respectively);
minerals and vitamins mixture consisted per kilogram:
210, 30, 100, 70 g of calcium, phosphorus, sodium, magnesium;
750, 30, 80, 2,730 mg of copper, selenium, iodine, vitamin E;
500,000 and 75,000 IU of vitamin A and D_3, respectively.

Table 4. Chemical composition of diet components

Item	Maize silage	Grass silage	Hay[1]	Mashed oats	Production mixture[2]
Dry matter (DM; g/kg)	356	327	897	870	884
Concentration (g/kg DM)					
Crude protein[3]	79.0	133.8	71.4	132.2	242.0
Crude fat	2.4	19.8	18.9	42.5	19.3
Crude fibre	179.5	262.1	309.2	141.4	39.1
Crude ash	1.5	8.5	3.2	33.6	75.2
NE_L (MJ/kg)[4]	6.62	4.93	4.68	6.83	8.02

[1]Permanent grassland hay from late cut with prevailing *Deschampsia cespitosa*, *Agrostis tenuis*, *Agrostis stolonifera*, *Alopecurus pratensis*.
[2]Production mixture consisted of 37, 31, 28 and 4 % (w/w) of wheat, barley, extracted soybean meal and a mixture of minerals and vitamins.
[3]Crude protein = N x 6.25.
[4]NE_L = Net Energy of Lactation (Sommer et al., 1994).

Results and discussion

The fat is a major variable constituent of milk and its composition is influenced by various factors (Schwendel et al., 2015). The group of animal factors includes breed, parity, stage of lactation (expressed in days in milk, weeks, or months) or milk yield and milk composition (Samkova et al., 2012). All these factors are within the breed affected by a phenotypic variation (cow individuality). It seems like the main role was assigned to a genetic variability (Arnould and Soyeurt, 2009) and to a physiology of milk production (Kay et al., 2005).

The results from our data analysed by a general linear model (Table 5) showed that the four animal factors tested ascribe a relatively high proportion of the total explained variation (R^2) of individual FA in milk fat of Czech Fleckvieh and Holstein cows ranging from 23.3 to 61.8 % (mean 42 %). The main factors were cow individuality (from 16 to 48 %; 28 %) and month of lactation (from 3 to 18 %; 9 %). Breed and parity affected the mean R^2 in a limited extent (each about 2 %).

The dominant role of month of lactation within the three factors (breed, parity, and days in milk)

Table 5. Distribution of the total variance for milk yield (kg/d), fat, protein and lactose content (g/100 g), milk fatty acids (FA), and groups of FA (g/100 g of FA) in a general linear model (GLM) involving animal factors

Item	Animal factors[1]								R^2 (GLM)[2]	
	Breed		Individuality		Parity		Month of lactation			
	%	P	%	P	%	P	%	P	%	P
Milk yield and composition										
Milk yield	8	***	31	***	1	*	34	***	76.2	***
Fat content	1	*	35	***	0#	ns	6	***	42.4	***
Protein content	9	***	32	***	1	**	29	***	72.1	***
Lactose content	0#	ns	40	***	1	ns	16	***	61.1	***
FCM[3]	6	**	30	***	0#	ns	32	***	71,7	***
ECM[4]	6	**	32	***	1	†	31	***	71,8	***
Individual FA										
C4:0	0#	ns	17	ns	1	ns	4	ns	23.3	*
C6:0	0#	ns	18	ns	1	ns	3	ns	24.1	*
C8:0	1	ns	19	*	1	ns	7	***	28.8	***
C10:0	4	***	20	***	1	*	12	***	37.4	***
C12:0	3	***	20	***	2	*	14	***	38.3	***
C14:0	1	**	24	***	2	*	15	***	41.3	***
C14:1	15	***	29	***	0#	ns	15	***	61.8	***
C16:0	2	***	40	***	2	***	9	***	54.4	***
C16:1	2	**	40	***	1	ns	4	*	47.9	***
C18:0	0#	ns	29	***	3	**	8	***	40.8	***
C18:1	0#	ns	26	***	2	*	18	***	46.2	***
C18:2*n-6*	0#	ns	48	***	2	***	5	***	54.9	***
C18:3*n-3*	0#	ns	39	***	5	***	4	**	47.0	***
CLA[5]	1	*	16	*	1	ns	9	***	28.2	***
Group of FA[6]										
SFA	0#	ns	29	***	2	**	16	***	46.8	***
UFA	0#	ns	27	***	2	**	17	***	45.9	***
MUFA	0#	ns	26	***	2	*	18	***	45.4	***
PUFA	1	ns	33	***	1	ns	5	**	39.2	***
C4-C14	2	**	18	**	2	*	15	***	35.8	***
C18-C24	0#	ns	26	***	2	**	16	***	44.8	***

P = probability; ns = not significant; * = p<0.05; ** = p<0.01; *** = p<0.001; # = % of total variance <0.5.
[1]Animal factors, in % = proportion of individual factors variance accounted for by total variance.
[2]R^2 (GLM), in % = proportion of total explained variance (coefficient of determination)
[3]FCM = Fat Corrected Milk (4 %).
[4]ECM = Energy Corrected Milk.
[5]CLA = C18:2 *cis*-9, *trans*-11.
[6]SFA = saturated FA; UFA = unsaturated FA; MUFA = monounsaturated FA; PUFA = polyunsaturated FA; C4-C14 = sum of C4 to C14 (even); C18-C24 = sum of C18 to C24; C18 = sum of C18:0, C18:1, C18:2*n-6*, C18:3*n-6*, C18:3*n-3*, and CLA.

was confirmed by Kelsey et al. (2003), who found the R^2 ranging from 2.2 to 35.8 %. Lower levels were probably caused by only one milk sampling, which did not allow for the testing of a cow individuality. The observed high proportion of R^2 was affected by all year round sampling and by factor of cow individuality. Cow individuality differences in FA proportion could be affected by various factors such as pedigree, health status or rumen biohydrogenation. Also different feed intake and obviously different response on feeding diet can play an important role. The effect of cow individuality on milk fat composition was also affirmed by Elgersma et al. (2006), who tested the response of individual cows on changes in feeding ration. They found that even if patterns in response to diet changes were similar, proportion of CLA differed among cows.

Table 6. Milk yield (kg/d), fat, protein, and lactose content (g/100 g), milk fatty acids (FA), and groups of FA (g/100 g of FA) depending on breed, cow individuality and parity

Number of milk samples	Breed[1]		Individuality[2]		Parity			Total			
	C	H	C	H	1	2	>3				
	186	170	25[#]	25[#]	98	142	116	Mean	Min.-Max.	SEM	RSD
Milk yield and composition											
Milk yield	19.03[a]	22.86[b]	11.4-26.1	10.8-29.6	18.90[a]	21.17[b]	22.13[b]	20.9	4.5-44.3	0.38	34.6
Fat	4.36	4.22	3.4-5.4	3.5-5.5	4.35	4.27	4.28	4.29	2.2-6.9	0.04	18.9
Protein	3.61[b]	3.36[a]	3.2-4.3	2.8-3.8	3.52	3.47	3.50	3.49	2.4-4.8	0.02	12.7
Lactose	4.82	4.78	3.9-5.2	4.4-5.0	4.86[b]	4.80[ab]	4.76[a]	4.80	3.3-5.5	0.02	6.3
FCM[3]	19.84[a]	23.43[b]	11.3-25.3	11.3-29.8	19.69[a]	21.76[b]	22.89[b]	21.6	3.7-47.4	0.39	33.7
ECM[4]	21.63[a]	25.23[b]	12.9-27.4	12.4-31.2	21.36[a]	23.55[b]	24.79[b]	23.3	4.1-47.6	0.40	32.2
Individual FA											
C4:0	2.01	2.03	1.7-2.3	1.6-2.3	1.96	2.02	2.07	2.02	0.8-3.7	0.02	20.6
C6:0	1.81	1.82	1.6-2.1	1.6-2.1	1.76[a]	1.81[ab]	1.86[b]	1.81	0.9-2.8	0.02	16.6
C8:0	1.35	1.33	1.2-1.5	1.1-1.5	1.29[a]	1.34[ab]	1.38[b]	1.34	0.6-2.2	0.01	16.6
C10:0	3.58[b]	3.38[a]	3.0-3.9	2.6-3.9	3.34[a]	3.47[ab]	3.62[b]	3.48	1.5-5.2	0.03	18.4
C12:0	4.35[b]	4.13[a]	3.6-5.1	3.1-5.0	4.07[a]	4.24[ab]	4.39[b]	4.24	1.8-6.3	0.04	18.9
C14:0	13.40[b]	13.07[a]	11.7-14.9	11.3-14.5	12.91[a]	13.31[ab]	13.44[b]	13.2	6.9-17.6	0.09	12.7
C14:1	0.98[a]	1.18[b]	0.8-1.2	0.8-1.5	1.00	1.14	1.06	1.08	0.3-1.9	0.01	25.8
C16:0	31.28[a]	32.69[b]	26.8-35. 7	27.1-39.1	31.24[a]	32.45[b]	31.94[ab]	31.9	22.1-43.6	0.02	12.2
C16:1	1.78[a]	1.88[b]	1.3-2.2	1.5-2.6	1.76[a]	1.87[b]	1.84[ab]	1.83	1.1-3.2	19.5	19.5
C18:0	8.96	8.66	6.7-10.7	6.5-11.9	9.30[b]	8.54[a]	8.75[ab]	8.82	3.9-17.3	0.11	24.4
C18:1	21.59	21.08	17.9-25.3	17.4-25.4	22.23[b]	20.90[a]	21.10[a]	21.3	12.4-36.0	0.21	18.5
C18:2*n-6*	1.60	1.59	1.2-2.5	1.2-2.0	1.56	1.58	1.63	1.59	0.7-3.2	0.02	20.7
C18:3*n-3*	0.39	0.38	0.3-0.6	0.3-0.5	0.41[b]	0.39[b]	0.36[a]	0.39	0.2-0.9	0.01	28.0
CLA[5]	0.41[b]	0.38[a]	0.3-0.6	0.2-0.5	0.42[b]	0.37[a]	0.40[ab]	0.39	0.1-0.9	0.01	38.3
Group of FA[6]											
SFA	69.12	69.49	63.2-73.2	65.0-74.0	68.32[b]	69.62[a]	69.71[a]	69.3	53.8-80.1	0.22	6.1
UFA	28.15	27.91	24.3-33.4	23.7-31.5	28.86	27.67	27.78	28.0	17.7-44.1	0.22	15.0
MUFA	24.92	24.76	21.4-28.7	21.0-28.9	25.62[b]	24.51[a]	24.57[a]	24.8	15.8-39.8	0.20	15.5
PUFA	3.24	3.15	2.6-4.7	2.5-3.9	3.24	3.16	3.21	3.20	1.4-6.4	0.03	20.5
C4-C15	30.58[b]	30.08[a]	27.8-32.8	26.2-32.9	29.52[a]	30.56[ab]	30.76[b]	30.3	16.8-40. 8	0.19	11.7
C18-C24	35.39	34.42	29.2-41.0	27.7-40.1	36.45[b]	34.19[a]	34.55[a]	34.9	17.8-56.3	0.32	18.0

[a, b] Means in breed and parity groups with different superscripts within a row differ ($P<0.05$); SEM = standard error of the mean; RSD = relative standard deviation (coefficient of variation), in % = [standard deviation/mean].100.
[1]C = Czech Fleckvieh; H = Holstein; [2#] = number of cows (range in cow means)
[3]FCM = Fat Corrected Milk (4 %); [4]ECM = Energy Corrected Milk.
[5]CLA = C18:2 *cis*-9, *trans*-11.
[6]SFA = saturated FA; UFA = unsaturated FA; MUFA = monounsaturated FA; PUFA = polyunsaturated FA; C4-C14 = sum of C4 to C14 (even); C18-C24 = sum of C18 to C24.

Milk fat composition is widely assessed by its FA groups. Most common evaluation is done by the presence of double bonds. In addition, milk fat could be evaluated according to the number of carbons. In our study, R^2 was evaluated both ways. With regards to the presence of double bonds, the testing of R^2 did not reveal any differences between saturated and unsaturated FA (46.8 *vs.* 45.9 %, respectively). Similarly, no differences were observed within each animal factor (Table 5). However, the differences were found according to the number of double bonds in the FA chain (45.4 and 39.2 % in monounsaturated and polyunsaturated FA, respectively). The lower value in polyunsaturated FA could probably be explained by a higher effect of feeding factors, which predominantly affected these FA (Coppa et al., 2013). As can be observed from data in Table 5, proportion of polyunsaturated FA was influenced by a variation in the cow individuality (33 %), while that of monounsaturated FA was affected by a cow individuality (26 %) and month of lactation (18 %).

The second way of FA classification is partially connected with the physiological pathway of FA synthesis: *de novo* and preformed FA (Vlaeminck et al., 2006; Harvatine et al., 2009). In our work, the differences in the groups classified by a number of carbons were observed. Those showed values of R^2: 35.8, 54.4 and 44.8 % for the groups with even saturated FA C4 to C14, C16 and C18 to C24, respectively. This was probably related to different heritability for the individual FA. Stoop et al. (2008) reported heritability coefficients of 0.31 to 0.54 and 0.09 to 0.21 for FA with short-/medium- (from C4 to C16) and long-chain (≥ C18), respectively.

Our data proved a statistically significant ($P<0.001$ and <0.01) effect of the breed on the individual FA up to C16 (excluding volatile FA) and consequently in the groups C4 to C14. This may suggest a more important effect of animal factors on short- and medium-chain FA than on the long-chain ones.

Significant effects of breed and cow individuality on CLA could be explained by the pathway of its synthesis. According to Mosley et al. (2006), 80 % of CLA is synthesized in the mammary gland. However, the low R^2 of CLA (28.2 %) indicates a lower proportion of animal factors. The prevailing proportion of the variation can be thus affected by feeding ration (e.g. Elgersma, 2015).

The composition of both, the individual FA and their groups in relation to breed, cow individuality and parity is collated in Table 6.

The inter-breed differences do not seem to be too high despite the statistical significance ($P<0.05$) observed among some individual FA and their groups. From the nutritional point of view, significantly higher proportion (32.7 *vs.* 31.3 %) of palmitic acid (C16:0) and lower proportion (0.38 *vs.* 0.41 %) of CLA were determined in milk fat of Holstein cows as compared with that of Czech Fleckvieh breed. However, both the breeds are supposed to have different feeding requirement, varying in milk yield and resulting in different feed intakes.

Cow individuality caused substantial differences within both the breeds. The extent of variability is characterized by wide ranges of FA proportions and relative standard deviations (coefficients of variation). It ranges for the individual FA between 12.2 % for C16:0 and 38.3 % for CLA.

Some reports suggested that breed differences in FA composition could be affected particularly by the fat yield per day (Soyeurt et al., 2007; Stoop et al., 2008). In milk of cattle breeds with considerably low milk yield or fat content there were mostly reported low proportions of saturated FA and thus higher proportion of unsaturated FA (Ferlay et al., 2006; Moioli et al., 2007). Limited differences in the proportions of quantitatively and nutritionally important FA were observed within the breeds with similar fat yield per day (Palladino et al., 2010). The differences could thus be affected by functional type (dairy Holstein vs. dual purpose Czech Fleckvieh) in association with rearing condition factors as seems to be in this work.

With regards to parity, the observed significant differences within FA composition were evident between primiparous and multiparous cows. No significant differences, except for linolenic acid (C18:3 *n-3*), were observed between dairy cows at second and further (≥3) lactations. According to the previously published data (e.g. Artegoitia et al., 2013; Stadnik et al., 2013), primiparous cows have nutritionally more desirable FA composition with lower proportion of saturated FA and higher proportion of unsaturated FA and CLA. The differences in the control of tissue mobilisation between multiparous and primiparous cows can be the reason

(Wathes et al., 2007). Both body and mammary gland development is not yet finished in primiparous cows in the post-calving period. This can cause a lower milk production and elevated utilisation of FA from both feeds and body reserves. As Miller et al. (2006) reported, activity of an enzyme synthase in mammary gland, which participates in FA production, is in primiparous cows low during the initial third of lactation and only during the final third of lactation reaches an activity usual in multiparous cows.

The effect of month of lactation is undoubtedly higher than parity. The most extensive changes occur particularly during the initial third of lactation (days 0-100), which was largely studied (Kay et al., 2005; Lake et al., 2007). In the works studying whole lactation period (days 0-305), the researchers (Garnsworthy et al., 2006; Mele et al., 2007) mostly sampled milk with a limited frequency. It could hardly give a true picture of the changes in details.

According to our data (Figure 1), more extensive changes occurred not only during the initial month of lactation, but also during the final third. The changes in the proportion of individual FA depended first of all on the number of carbons in FA chains which indicates the relation to the pathway of FA synthesis. The proportion of FA with C4 to C14 and C16 culminated during the second third of lactation, while FA with ≥18 carbons were at their minimum level. Somewhat different course during lactation we observed for CLA with the maximum proportion at the end of lactation period. These observations correspond to a large extent with the results of Craninx et al. (2008) or Micinski et al. (2012).

The obtained differences were probably affected by different metabolic requirements during the lactation period (Lake et al., 2007) and physiological state of dairy cows (Nielsen et al., 2003), which influenced the ratio between *de novo* and preformed FA. Such relation does not depend on diet

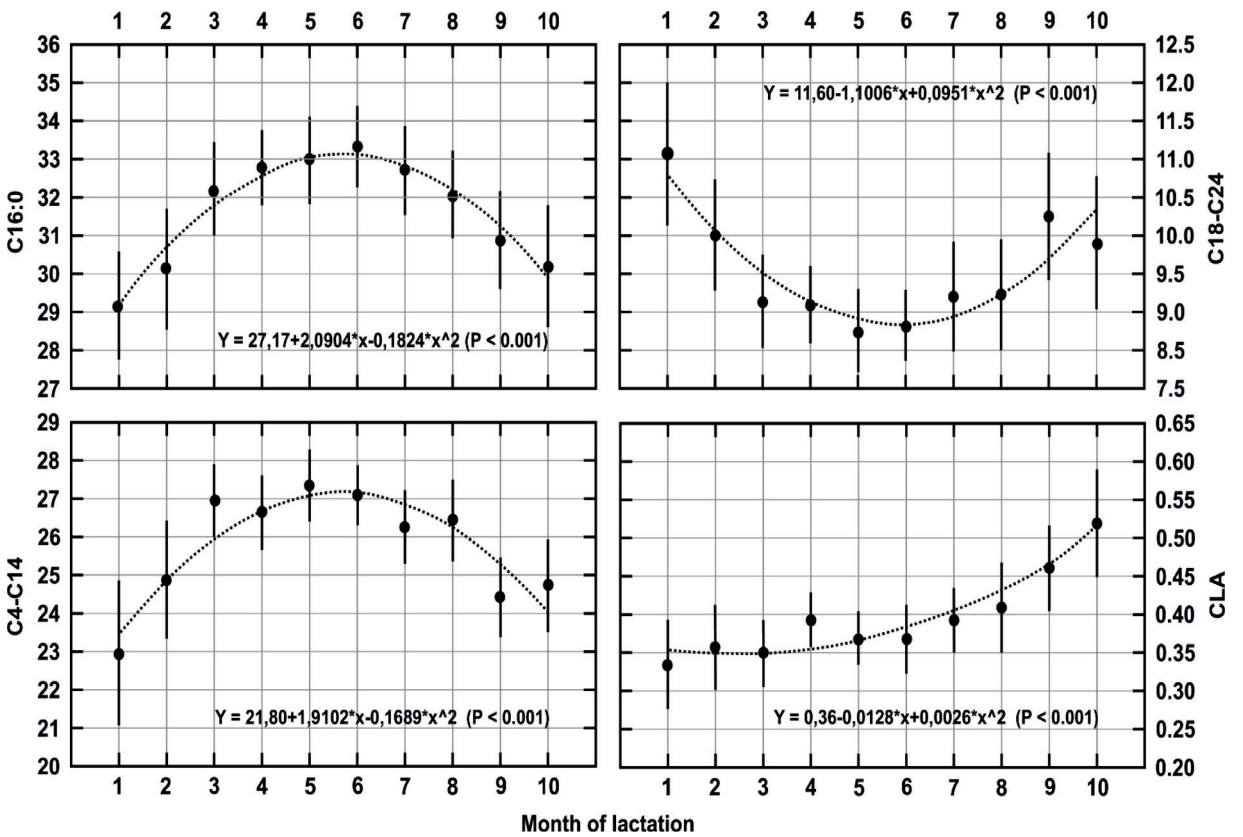

Figure 1. Proportion of fatty acids (FA) in C4-C14 (even), C16:0, conjugated linoleic acid (CLA), and C18-C24 (even) (y-axis: g/100 g of FA) depending on stage of lactation (x-axis: month of lactation); vertical bars denote 0.95 confidence intervals

(Garnsworthy et al., 2006). Negative balance of energy during the initial phase of lactation period results in limited production of *de novo* FA, proportion of which is low during this phase. Similar situation occurs during the finishing lactation period. The proportion of preformed FA can vary between 5 and 20 % in relation to dairy cow physiology.

Conclusion

The results of this study showed considerable effects of four animal factors on milk fat composition of two different breeds under the same rearing conditions. Whilst cows´ individuality and month of lactation were the main factors affected the variation in milk fatty acids, the effects of breed and parity were limited. Nevertheless, in particular the frequently discussed influence of the breed on milk fat fatty acid profile is often overlapped with the nutrition effects, whereas it can be considered as pure result here. Cow individuality caused substantial differences within both the breeds, which could be probably affected by different interactions between rearing conditions factors and cow individuality. Lower proportions of *de novo* fatty acids were determined during the initial and final third month of lactation as a possible result of negative energy balance. This could be an influential physiology phenomenon in these lactation periods. According to the above mentioned facts it is obvious that the work can bring some new insights to the current topic of influencing milk fat composition, which is important not only from a nutritional and health point of view, but also from a technological point of view.

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Zajednički utjecaj pasmine, rednog broja i mjeseca laktacije te utjecaja jedinke na sastav masnih kiselina mliječne masti

Sažetak

Cilj ovog istraživanja bio je objasniti utjecaj četiri čimbenika - pasmine, broja laktacije, utjecaja jedinke (fenotipska varijacija unutar pasmine) i mjeseca laktacije na sastav masnih kiselina (MK) u mliječnoj masti kravljeg mlijeka. Lokalna češka pasmina Fleckvieh uspoređivana je sa svjetski poznatom pasminom holstein. Ukupno je analizirano 357 uzoraka mlijeka prikupljenih tijekom jedne godine od 25 mliječnih krava svake pasmine. Varijacije sastava masnih kiselina uglavnom su bile pod utjecajem jedinke (16-48 %) i mjeseca laktacije (3-18 %), dok je utjecaj pasmine bio ograničen (oko 2 %). Faktori vezani za životinje također su se pokazali statistički značajnim za sastav MK. Značajnije razlike u varijaciji svih čimbenika utvrđene su u odnosu na broj ugljikovih atoma u MK (35,8, 54,4 i 44,8 % za C4 do C14, C16, C18 i C24) te u odnosu na broj dvostrukih veza (45,4 % jednostruko nezasićene i 39,2 % višestruko nezasićene MK). Pri tom nisu utvrđene značajnije razlike u sastavu zasićenih i nezasićenih MK (46,8 %, odnosno 45,9 %) između ispitivanih pasmina krava. Zaključno, s nutritivne točke gledišta bilo bi prikladnije MK klasificirati prema broju ugljikovih atoma, a ne prema dosad uvriježenoj podjeli prema zasićenosti/nezasićenosti.

Ključne riječi: krava, mlijeko, masne kiseline, utjecaj jedinke

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