

Effects of replacing rapeseed cake with linseed cake in a corn-grass silage-based diet for milking cows*

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The aim of this study was to evaluate the substitution of rapeseed cake by an equivalent amount of linseed cake in the diets of dairy cows, and the effects of such equivalent substitution on the proportion of feed consumed, milk yield and chemical composition, fatty acid structural composition in their milk fat, and the metabolic profile of the cows' metabolism. The studies were conducted on 16 Polish Holstein-Friesian cows, divided into 2 groups [n= 8] using the analogue method. Cows from the control group received an addition of rapeseed cake, while animals from the experimental group received linseed cake as a by-product of "cold" oil extraction. The cows were maintained in a

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free-stall system in a separated compartment fitted with the Calan-gate system and fed according to TMR systems. Following 70 days there were no significant differences in the daily milk fat, protein, lactose, and yield between the control vs. experimental groups. Additionally, the biochemical parameters in the blood serum of cows did not show negative effects of either diet on the animals' health condition.

Rapeseed and linseed cake have similar feeding value. The slight improvement in the fatty acid structure in the milk fat of cows receiving linseed cake is of no greater practical value. Thus, in European conditions rapeseed cake should be preferred, as its availability is wider and its price is lower.

KEY WORDS: biochemical parameters / fatty acids / linseed cake / milk / rapeseed cake

Supplementing the diets of animals with oilseed plants in order to modify the profile of fatty acids in milk fat is a strategy which has been known for several decades [Chilliard and Ferlay 2004, Jóźwik *et al.* 2010a, Strzałkowska *et al.* 2009ab, Szmatoła *et al.* 2013]. On one hand, the unsaturated fatty acids contained in oilseed plants attenuated the concentration of saturated fatty acids in milk, while on the other hand this lead to an increased share of fatty acids such as, among others, 18:1 cis-9, 18:3 n-3 and the conjugated linoleic acid [CLA] cis-9, trans-11, [Frelich *et al.* 2012, Lerch *et al.* 2012]. A change in the concentration of milk fat fatty acids accentuates the nutritive value of milk. A properly balanced diet is important for ruminants.

Intensive energy turnover, under conditions of limited body reserves, causes high- yielding cows to be more susceptible to numerous pathogenic factors, especially during early lactation. The organism of a high-yielding dairy cow is often in the state of disturbed homeostasis caused principally by the generation and accumulation of free radicals [Jóźwik *et al.* 2004, 2010b, 2012a, Abuelo *et al.* 2015]. The oxidative stress was the principal factor causing dysfunction of the immune system of the organism and impairing the response to inflammatory conditions especially in the perinatal period [Spears and Weiss 2008]. Extracted rapeseed meal is a commonly used source of protein in high-milking ruminants' feed.

In order to improve the dietetic value of the milk of ruminants by the supplementation of a diet with either the seed of oily plants [linseed and rapeseed] or oils rich in non-saturated fatty acids, allows the modification of the profile of milk fatty acids, and sometimes leads to changes in biochemical mechanisms within cows.

Linseed and rapeseed cakes are also a rich source of polyphenol compounds, including benzoic and cinnamyl acids, coumarin, flavonoids and others, which act as antioxidants [Terpinc *et al.* 2012]. Linseed is a very rich source of lignans, which have a favourable effect on the hormonal metabolism, principally in relation to sex hormones, and show antioxidant properties [Murkies *et al.* 1998]. However, these plants contain some anti-nutritional factors, especially rapeseed [glucosinolates or erucic acid].

Thus, the aim of the study was to evaluate the influence of linseed and rapeseed on milk yield and chemical composition, fatty acid structure in milk fat, and the metabolic profile of the cows' metabolic organism.

Material and methods

Animals

The studies were conducted on Polish Holstein-Friesian cows selected during their second lactation, weighing on average 689 kg (± 17). The cows were between day 95 and day 110 of lactation, and were divided into 2 groups ($n=8$ animals in each), using the analogue method, taking into consideration the following parameters: number of lactation days, current daily milk yield, and the health condition of the mammary gland, estimated on the basis of the somatic cell count in milk. All experimental procedures fully met the ethical, legal and experimental conditions approved by the Local Ethics Commission No. 27/2009. There was no conflict of interest.

Nutrition of cows and registration of the daily feed consumption and milk yield

The daily diet was balanced in such a way as to ensure that an *ad libitum* consumption covered the requirements of the cows for both maintenance and production, estimated at a level of 35 kg milk/cow/day containing 4.0% fat and 3.5% protein (Tab. 1 and 2). Water was provided *ad libitum*. The cows from the control group received an addition of rapeseed cake, while animals from the experimental group received linseed cake, with equivalency of fat composition. Both types of cake were received as a by-product of “cold” oil extraction. The cows were maintained in a free-stall system, in a separated part of the cow-shed in which other animals, not participating in the experiment, were also housed. This separated compartment was fitted with the Calan Gate System (American Calan; NH, USA), ensuring electronically regulated individual access to the feed, which made possible precise control of the nutrition of every cow over the whole period of the experiment. The amounts of feed consumed and milk obtained were registered electronically. All the components of the diet were mixed in an experimental Super Data Ranger feeder wagon, which ensured uniformity of the feed offered. The TMR mixture was offered in quantities ensuring that the leavings would amount to less than 5%, calculated on a dry matter basis (5% orts on an as fed basis). The leavings were removed and weighed once daily. The experiment lasted 70 days. Representative feed samples were collected once a week and stored after freezing. Collective samples were analysed once a fortnight. Samples of leavings were collected and analysed analogously.

Milking, field registration, sampling of milk and blood

The cows were milked twice daily, at 6.00 and 18.00 hours. The amount of milk obtained was registered individually for each cow at each milking, using measurers installed in the milking parlour. The milk was analysed for the content of basic components on the basis of representative milk samples, taken individually from each cow at the beginning of the experiment, at monthly intervals, and at the end of the experiment. The milk samples were drawn into propylene containers with a preservation compound (Microtabs II, Bentley) and next transferred to the laboratory.

Parallel to the milk samples taken from each cow for basic chemical analyses, 100 mL of milk was collected for the determination of the fatty acid profile. Following cooling, these samples were transported to the laboratory, where they were frozen and stored until analysis at a temperature of -28°C.

Chemical analysis and estimation of nutritive value of feeds and milk.

The feed samples were analysed for the content of basic components using standard methods [AOAC, 2001]. On the basis of the chemical composition the nutritive value was estimated for feeds composing the daily diet according to the INRA system [2001], using the computer software PrevAlim 3.22 (Version professionnelle intégrale, INRA 1988-2004, France). The content of milk components was determined using the Milkoscan FT2 apparatus (Foss Analytical, Hillerød, Denmark), while the somatic cell count was determined using IBCM apparatus (Bentley Instruments, Chaska, USA).

Blood samples from the jugular vein (9 mL) were collected by an authorized veterinarian before the morning feed.

The biochemical parameters in blood serum were estimated using the COBAS INTEGRA® 400 plus system (Roche Diagnostics Ltd., Rotkreuz, Switzerland).

Fatty acid analyses

Fatty acids were extracted from milk samples with a chloroform-methanol mixer (2:1 v/v) by the method emulated by Folch, Lees, & Sloane Stanley [1957]. Fatty acid methyl esters were analysed using a gas chromatograph (GC-7890 Agilent, Santa Clara, USA) equipped with a 60 m Hewlett-Packard-88 capillary column (Agilent J&W GC Columns, Santa Clara, USA) with a 0.25 mm inner diameter and a 0.20 µm film thickness. A 1 µL sample was injected at a split ratio of 1:40. Helium was used as the carrier gas at a flow rate of 50 mL.min⁻¹. The injector and detector were maintained at 260°C. Column oven temperature was programmed to increase from 140°C [held for 5 min] at a rate of 4°C min⁻¹ to 190°C and then to 215°C at a rate 0.8°C.min⁻¹. Individual fatty acids were identified by comparing retention times to those of a standard mixture (Supelco 37 Component FAME Mix, 47885-U - 10 mg.ml⁻¹ in methylene chloride, analytical standard, Sigma-Aldrich Co.) and expressed as a percentage of total fatty acids.

Statistical analysis of results

All of the data were analysed by the least-squares mean method using the GLM procedures in SAS, Version 8e for MS Windows [SAS, SAS/STAT 1999-2001] using the model which included the fixed effect of the interaction group × date of sampling; a P value of <0.05 was taken as significant.

Results and discussion

The results in Tables 1 and 2 showed the chemical composition and feeding value components of the diets based on feed rations and concentrate mixture. After 70 days

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Table 1. Chemical composition, feeding value and components of the diets based on feed rations

Component	Dry matter (%)	In dry matter								
		crude protein (%)	crude fibre (%)	NDF (g/kg)	ADF (g/kg)	ether extract (%)	UFL (units)	PDIE (g/kg)	PDIN (g/kg)	%
Corn silage	34.82	6.13	24.75	49.02	29.95	2.50	0.90	68	50	27.62
Wilted grass silage	26.64	22.17	25.17	43.06	32.98	1.76	0.85	69	82	17.58
Corn seed silage	65.74	10.86	13.85	37.26	20.35	4.10	1.25	122	85	11.72
Concentrate mixture (see Tab. 2)	83.00	20.70	8.23	10.91	21.40	3.34	1.02	126	154	43.08
Whole diet (TMR)	57.76	15.78	16.43	30.18	23.74	2.92	0.98	99.49	100.00	100.00

NDF – Neutral Detergent Fibre; ADF – Acid Detergent Fibre; UFL – equivalent of 1kg of air dried barley; PDIN – g of protein that is synthesised in the rumen from breakdown of dietary nitrogen and is absorbed in the small intestine; PDIE – g of protein that is synthesised in the rumen from breakdown of dietary energy and is absorbed in the small intestine.

Table 2. Chemical composition and feeding value components of concentrate mixture

Component	Dry matter (%)	In dry matter									
		crude protein (%)	crude fibre (%)	NDF (g/kg)	ADF (g/kg)	ether extract (%)	UFL (units)	PDIE (g/kg)	PDIN (g/kg)	in concentrate mixture (%)	
										Gr 1	Gr 2
Rapeseed cake	89.56	31.32	21.83	46.59	26.51	10.65	1.06	160	253	26.51	-
Linseed cake	89.66	39.38	12.83	23.30	14.70	13.37	1.06	170	237	-	21.31
Triticale	81.33	10.72	2.69	11.83	-	1.70	1.21	108	88	28.91	33.71
Soya extract, meal	84.39	41.83	11.39	37.41	21.74	2.00	1.16	249	360	25.03	25.03
Ketomix E18 ¹	47.12	13.00	-	-	-	-	1.98	-	-	11.89	11.89
Chalk	-	-	-	-	-	-	-	-	-	1.83	1.83
Co-bind A-Z ²	-	-	-	-	-	-	-	-	-	0.57	0.57
Sodium bicarbonate	-	-	-	-	-	-	-	-	-	2.40	2.40
Witamix KW ³	-	-	-	-	-	-	-	-	-	2.86	2.86
Concentrate mixture	83.00	20.70	8.23	10.91	21.40	3.34	1.02	126	154	100.0	100.0

¹Energy supplement (3.36 UFL/kg DM).

²Neutralizer of mikotoxins.

³Macro (g/kg): Ca – 120; P – 60; Mg – 50; Na – 110; micro-elements [mg/kg]: Mn – 4000; Zn – 9500; Cu – 1150; J – 90; Co – 25; Se – 45; vitamins (mg/kg): D3 – 120 000; E – 4000; K3 – 50; B1 – 150; B2 – 100; B6 – 50; B12 – 550; calcium pantothenate – 300; niacin – 2500; folic acid – 30; vitamin A – 1000.

UFL – equivalent of 1kg of air dried barley; PDIN – g of protein that is synthesised in the rumen from breakdown of dietary nitrogen and is absorbed in the small intestine; PDIE – g of protein that is synthesised in the rumen from breakdown of dietary energy and is absorbed in the small intestine.

during the experiment there were no significant differences in the daily milk yield between the control and experimental groups (Tab. 3). In both groups of cows the daily milk yield attenuated with the progress of the experiment to a similar degree, i.e. by 14.8% and 14.4%, in the control and experimental groups, respectively (Tab. 3). Moreover, no significant differences were observed in the daily production of the basic milk components, i.e. fat, protein and lactose.

Table 3. Daily milk yield and milk components (kg/d)

Items	DM intake (cow/d)	Yield/day							
		milk	ECM	total solids	SNF	fat	protein	lactose	casein
Start of experiment									
Group1									
LSM	18.99	29.8	31.7	3.611	2.580	1.153	0.922	1.386 ^a	0.686
SE	1.08	1.7	1.8	0.050	0.057	0.099	0.038	0.028	0.034
Group2									
LSM	19.94	31.3	33.8	3.534	2.498	1.278	0.914	1.318 ^b	0.658
SE	1.08	1.7	1.8	0.055	0.060	0.099	0.040	0.029	0.036
End of experiment									
Group1									
LSM	16.18	25.4	28.2	3.637	2.601	0.967	0.968	1.345	0.717
SE	1.08	1.7	1.9	0.056	0.060	0.099	0.040	0.030	0.036
Group2									
LSM	17.07	26.8	30.7	3.652	2.617	1.098	1.012	1.304	0.745
SE	1.08	1.7	1.9	0.053	0.057	0.099	0.038	0.029	0.034

^{ab}Within column means bearing different superscripts differ significantly at $P \leq 0.05$.

ECM – energy corrected milk; SNF – solid not fat.

Of interest is the significant ($p < 0.05$) decrease of urea concentration in the milk of cows receiving a diet supplemented with linseed cake, remaining within the preferred reference interval (Tab. 4) from 294 to 228 mg/L. Theoretically, the level of urea should be similar in both groups, as the diets were balanced to ensure that they were isocaloric and isonitrogen (Tab 1 and 2).

Despite the lack of significant differences in the somatic cell count in milk (Ln SCC), a slight tendency to decrease their amount in the milk of cows fed a diet containing rapeseed cake was observed (Tab. 4). A reverse tendency was observed for the content of lactose in milk.

No clear differences, either in the content or in the profile of fatty acids in the milk fat obtained from cows of both groups were observed (Tab. 5). This was caused by the relatively low fat concentration (below 3%) in the diet's total dry matter, as well as by the amount of fat introduced additionally into the diets in the form of both cakes (ca. 1.2% in the diet's dry matter). Taking into consideration the amount of dry matter consumed by the cows, the amount of oil consumed in the form of cakes amounted to a mean of ca. 234g and 200g, at the beginning and at the end of the experiment, respectively. Thus, the comparatively small amount of fat consumed by the cows could not significantly affect either the content of fatty acids or their profile in the milk fat (Tab. 5).

In the present investigation the nutrition of cows using a diet of the given composition did not lead to significant changes in the fatty acid profile of milk fat (Tab. 5). Significant differences did occur in the share of linoleic acid (18:3 n-3) in the pool of fatty acids; this share was ca. 100% higher in the milk fat from cows fed a diet containing linseed cake. Significant ($p < 0.05$) increases in the content of palmitic acid (16:1) and MUFA occurred in the milk fat obtained from cows at the beginning and the end of the experimental period, and showed only small final variability. This suggested that

between cows randomly assigned to the two experimental groups genetic differences in the profile of milk fat fatty acids existed. The differences in the content of decanoic (10:0), oleic (18:1, n-9) and pentadecanoic (15:0) acids occurred only at the start of the experiment; indeed, on the day the experiment concluded such differences were not observed. In the milk obtained from cows in the control group a significant ($p < 0.05$) decrease was observed in the share of acids from the n-6 family, while in the experimental group the share of fatty acids from the n-3 family increased by about 100%. These changes had a favourable effect on the AI (*atherogenic index*) and TI (*thrombogenic index*) content, as well as n-6/n-3, the share of which decreased in the milk obtained from cows fed a linseed cake supplement. In this group of cows a tendency was also observed to decrease the share of the total amount of acids with a hypercholesterolemic effect (12:0; 14:0 and 16:0) in the overall pool of fatty acids.

On the basis of the biochemical parameters determined in the blood serum of cows, characterising the profile of liver, kidneys, heart, lipids and bones, we propose that the diets used had no negative effect on the overall animals' health condition (Tab. 6).

The absence of significant differences in the milk yield and net gain of milk components suggests that the diets containing rapeseed and linseed cakes were of a similar nutritive value. The adding of plant oils, in the form of whole seeds or by-products such as cakes or liquid oils, to the diets of cows, led to lower feed consumption, and thus additionally to a lower daily milk yield and lower milk fat

Table 4. Chemical composition of milk, acidity, content of urea and SCC

Item	Fat (%)	Total protein (%)	Casein (%)	Lactose (%)	Total solids (%)	SNF (%)	Citric acid (%)	FFA (%)	Acidity (%SH)	Urea (mg/L)	Ln SCC
Start of experiment											
Group1	3.86	3.30	2.46	4.90 ^a	12.81	9.16	0.166	0.496	6.28	212	5.09
LSM	0.27	0.13	0.11	0.09	0.17	0.19	0.012	0.057	0.41	21	0.17
SE											
Group2	4.13	3.30	2.39	4.64 ^b	12.58	8.91	0.176	0.487	6.44	249	4.98
LSM	0.27	0.14	0.12	0.09	0.18	0.20	0.013	0.060	0.43	22	0.15
SE											
End of experiment											
Group1	3.80	3.42	2.54	4.76 ^c	12.87	9.20	0.158	0.806	6.86	294 ^a	5.06
LSM	0.27	0.14	0.12	0.10	0.19	0.20	0.013	0.061	0.44	22	0.19
SE											
Group2	4.13	3.62	2.67	4.60 ^d	13.00	9.29	0.187	0.876	7.22	228 ^b	5.29
LSM	0.27	0.13	0.12	0.09	0.18	0.19	0.012	0.058	0.41	21	0.18
SE											

^{ab} Within column means bearing different superscripts differ significantly at $P \leq 0.05$.
SNF – solid not fat, FFA – free fatty acids.

Table 5. Fatty acids in milk fat (g/100 g milk fat)

Item	Start of experiment				End of experiment			
	Group 1		Group 2		Group 1		Group 2	
	LSM	SE	LSM	SE	LSM	SE	LSM	SE
6:0	2.69	0.30	2.82	0.32	2.40	0.32	2.26	0.30
8:0	2.71	0.32	2.83	0.34	2.32	0.34	2.78	0.32
10:0	2.70 ^a	0.20	2.04 ^b	0.21	2.99	0.21	2.78	0.20
12:0	3.35	0.28	3.00	0.30	3.91	0.30	3.51	0.28
14:0	11.03	0.52	11.21	0.55	12.91	0.56	12.19	0.53
15:0	1.47 ^a	0.21	2.33 ^b	0.22	2.12	0.22	1.71	0.21
16:0	31.43	1.18	29.52	1.25	33.52	1.26	31.29	1.19
16:1	1.81 ^a	0.17	2.26 ^b	0.18	1.77 ^a	0.18	2.25 ^b	0.17
18:0	12.58	0.81	11.10	0.85	10.46	0.86	10.47	0.81
18:1 n9	25.47 ^a	1.23	28.89 ^b	1.30	24.00	1.31	26.79	1.24
18:2 n6	2.73	0.18	2.71	0.19	2.50	0.19	2.60	0.18
18:3 n3	0.30	0.09	0.27	0.10	0.25 ^a	0.10	0.51 ^b	0.09
20:4 n6	0.22	0.03	0.17	0.03	0.15	0.03	0.21	0.03
20:5 n3	0.18	0.05	0.12	0.05	0.08	0.05	0.14	0.05
SCFA	8.10	0.51	7.69	0.54	7.70	0.54	7.82	0.51
LCFA	41.50	1.63	43.28	1.73	37.45	1.74	40.73	1.64
MCFA	49.09	1.74	48.33	1.84	54.23	1.86	50.96	1.76
MUFA	27.28 ^a	1.26	31.15 ^{bc}	1.33	25.77 ^a	1.34	29.04 ^c	1.27
PUFA	3.44	0.22	3.28	0.23	2.99	0.24	3.46	0.23
SFA	67.96	1.45	64.86	1.54	70.62	1.55	66.98	1.47
AI	2.67	0.23	2.32	0.24	3.17 ^a	0.25	2.63 ^b	0.23
TI	3.44	0.29	2.92	0.30	3.79 ^a	0.31	3.02 ^b	0.29
n-6	2.95	0.14	2.88	0.16	2.65 ^a	0.16	2.81 ^b	0.14
n-3	0.48	0.17	0.39	0.12	0.33 ^a	0.11	0.65 ^b	0.08
MUFA/SFA	0.40	0.02	0.48	0.03	0.39	0.04	0.43	0.03
PUFA/SFA	0.05	0.01	0.05	0.01	0.04	0.01	0.05	0.01
PUFA/MUFA	0.13	0.02	0.11	0.01	0.12	0.01	0.12	0.01
n-6/n-3	6.15	0.12	7.38	0.15	8.03 ^a	0.11	4.32 ^b	0.09

^{abc}Within rows means bearing different superscripts differ significantly at $P \leq 0.05$.

percentage content. A similar effect was observed by Lerch *et al.* [2012], who audited extruded linseed oil meal or rapeseed oilmeal, rapeseed cake or whole rapeseeds, not processed (the addition of fat reached 2.5-3.0% in the diet dry matter), in cows' diet composed of grass and hay. In the present study, however, the share of oil, introduced with rapeseed and linseed cakes, amounted to ca. 1.2% of the diet's dry matter, while the total fat content in the diet reached 3% of the dry matter. The majority of studies show that the share of plant fat introduced into the diet for cows was x2-3 higher [Martin *et al.* 2008, Oba *et al.* 2009]. Benchar *et al.* [2012] demonstrated that the addition of linseed oil to the diet of cows at a level of 4% of diet dry matter had no negative effect on the processes taking place in the rumen, diet digestibility or milk yield; the daily milk yield increased linearly with the increasing level of linseed oil. However, the results of some studies have shown that an addition of linseed oil resulted in a decrease of feed consumption at a level of 3% of diet dry matter, which was accompanied by decreased digestibility of the organic matter of the whole diet [Martin *et al.* 2008].

Table 6 Concentration of biochemical parameters in serum

Item	Start of experiment				End of experiment				Range
	group 1		group 2		group 1		group 2		
	LSM	SE	LSM	SE	LSM	SE	LSM	SE	
Albumin g/L	40.8	0.8	39.4	0.8	41.4	0.8	41.4	0.8	27-43
Alkaline phosphatase U/L	50.0	7.0	38.8	7.1	69.1	7.1	55.3	7.1	29-99
Alkaline aminotransferase U/L	27.8	1.3	25.0	1.3	30.8	1.3	28.3	1.3	17-37
Asparagine aminotransferase U/L	101.1	12.9	83.3	12.9	126.5	12.9	104.1	12.9	48-100
Bilirubin µmol/L	1.55	0.24	1.61	0.24	1.34	0.24	1.41	0.24	1.7-5.1
Calcium mmol/L	2.50	0.06	2.50	0.06	2.36	0.06	2.49	0.06	1.98-2.5
Cholonoesterase U/L	40.5	12.7	30.0	12.7	104.8	12.7	103.6	12.7	?
Cholesterol mmol/L	5.37	0.34	5.19	0.34	5.55	0.34	5.93	0.34	2.3-6.6
Kreatinine kinase U/L	138.1	10.1	120.9	10.1	124.6	10.1	109.9	10.1	44-228
Chloridium mmol/L	100.3	1.0	98.9	1.0	99.6	1.0	98.2	1.0	?
Creatinine µmol/L	58.1 ^a	2.0	66.2 ^b	2.0	57.2 ^a	2.0	64.7 ^b	2.0	62-97
Gamma-glutamyl transferase U/L	34.3	3.6	38.0	3.6	40.2	3.6	42.9	3.6	20-48
Glucose mmol/L	3.43	0.07	3.57	0.07	4.21 ^a	0.07	4.52 ^b	0.07	2.1-3.9
Iron µmol/L	26.6	2.0	26.2	2.0	25.9	2.0	29.3	2.0	10-29
Potassium mmol/L	4.78	0.11	4.61	0.11	4.87	0.11	4.76	0.11	4.0-5.3
Lactate mmol/L	2.62	0.34	2.98	0.34	2.70	0.34	3.37	0.34	1.11-8.33
Cholesterol LDL mmol/L	1.08	0.17	1.02	0.17	1.25	0.17	1.57	0.17	?
Hepatic lipase U/L	6.35	0.34	6.06	0.34	6.76 ^a	0.34	7.91 ^b	0.34	?
Magnesium mmol/L	0.96	0.03	1.02	0.03	1.01	0.03	1.01	0.03	0.7-1.1
Sodium mmol/L	142.5	1.3	141.3	1.3	142.1	1.3	141.3	1.3	139-144
Phosphorus mmol/L	1.88	0.11	1.78	0.11	1.83	0.11	1.85	0.11	1.5-2.9
Total protein g/L	77.5	1.6	80.1	1.6	74.6 ^a	1.6	78.6 ^b	1.6	59-77
Triglycerides mmol/L	0.12 ^A	0.01	0.15 ^B	0.01	0.10	0.01	0.12	0.01	0.0-0.2

^{a,b,A,B} Within rows means bearing different superscripts differ significantly at: small letters – P≤0.05, capitals – P≤0.01.

The lower urea level in the milk of cows from the experimental group may have been caused by a lower rate of protein deamination in the rumen, with a simultaneously larger pool of available energy, determined by the higher digestibility of crude fibre contained in the cakes, of which a considerable share is easily soluble in water [Warrand *et al.* 2005], and a lower proportion of energy lost in the form of methane [Martin *et al.* 2008].

The slight variations in the lactose content in milk did not indicate the presence of subclinical udder inflammation [Jóźwik *et al.* 2004, 2010b, Strzałkowska *et al.* 2010].

Corn and grass silages are characterised by a comparatively low level of ether extract and the fatty acids it contains, particularly unsaturated acids from the n-3 and

n-6 family. According to Sterk *et al.* [2011] the content of unsaturated fatty acids in linseed and rapeseed is x10-15 higher than the content of these acids observed in corn silage, x50-60 higher by comparison with grass silage, and x5 higher vs. alfalfa silage.

Changes in the structure of fatty acids in milk are related to the pool of these acids occurring within the feed. Rapeseed contains considerable amounts of acid cis-9 18:1, while linseed oil is characterised by a high quantities of 18:3 n-3 [Lerch *et al.* 2012a]. In linseed oil the share of linoleic acid (18:3 n-3) and of CLA constitutes 50-60% of the total amount of fatty acids [Benchar *et al.* 2012]. For this reason, in the present investigation within milk obtained from cows fed a diet supplemented with linseed cake the content of acid 18:3 n-3, as well as acids of the n-6 family, was twice as high as that recorded in the milk of cows receiving rapeseed cake.

The fat contained in rapeseed had no effect on the content of 18:3 n-3 in milk. A similar effect was reported by Collombo *et al.* [2004] and Frelich *et al.* [2012]. The results of some studies indicate that fat contained in products obtained from rapeseed, compared to other oleic plants, causes a slight increase in the concentration of acid trans 18:1 in milk [Glasser *et al.* 2008]. The results of studies conducted by Lerch *et al.* [2012b] have shown that the addition of both linseed and rapeseed increases the content of the trans form of fatty acids, regardless of the feeding season.

Milk's fat fatty acid structure depends not only on the type of fat present in the seeds of various oleic plants, but also on the processing methods used. The results of studies conducted by Lerch *et al.* [2012a] demonstrate that a similar percentage of fat content, and its quantity in the daily milk yield, may be obtained when using both whole, unprocessed rapeseeds and rapeseed cake, obtained when the oil is obtained through "cold" extraction. However, the addition of whole rapeseed attenuated the amount of CLA in milk. The same authors [Lerch *et al.* 2012b] have shown that the addition of rapeseed cake, extruded linseed or whole rapeseeds, unprocessed, leads to an increased concentration in milk of fatty acids synthesized *de novo*, but simultaneously caused an increase in 18:0 and cis-9 18:1 acids.

The structure of milk fatty acids is also affected by the physical structure and type of fodder [Benchaar *et al.* 2012]. This relationship has also been confirmed by Samkowa *et al.* [2009]. The degree of biohydrogenation of unsaturated fatty acids in the rumen depends on the physical structure of the diet, determined by the ratio of fodder dry matter to concentrated dry matter [Dang Van *et al.* 2011, Puppel *et al.* 2014, Sun and Gibbs 2012]. This indicator accentuates with the increasing share of fodders in the diet, which is reflected in milk – higher concentration of 18:0 and a lower transfer of 18:2 n-6 and 18:3 n-3 from the diet to milk.

The increased level of aspartate aminotransferase in the blood serum of cows from both groups suggests increasing of metabolic disorders in the liver, which could lead even to its damage. According to an earlier study, transferases demonstrated considerable liver specificity, and an increase in their level indicates that liver hepatocytes are damaged [Oprządek *et al.* 2005, Józwick *et al.* 2012b]. In the present study the activity of this enzyme remained within the known reference values. Also the

level of bilirubin, remaining below the reference levels, did not indicate that the animals' livers were damaged. The level of glucose, exceeding the upper level of the given norms, indicated only that the animals' energy requirements were fully covered. Diabetes could occur if the glucose level in blood serum exceeded 5.8 mmol/L [Hasegawa *et al.* 1999]. The significantly higher level of this component in the blood serum of the experimental animals was undoubtedly related to the lowered level of urea in the milk of the cows.

The presence of specific plant proteins in cow silage-based diet is an essential problem in the aspect of potential presence the allergenic determinants in milk and products derived from it. Some linseed proteins are able to induce clinical symptoms of food allergy in sensitized individuals [Leon 2002]. However, rapeseed oil is free from allergy-inducing proteins [Gylling 2006]. These data are in the contrast to the commonly used soy proteins with high allergenic properties [Wróblewska 2007].

Both rapeseed and linseed cakes are valuable high protein supplements in dairy cattle diets. The addition of linseed cake to the diet of cows, compared to rapeseed cake, resulted in a slight improvement of the fatty acid structure in milk fat. However, the improvement of the profile of milk fatty acids to such a degree is possibly of no greater practical value. Thus, in European conditions rapeseed cake should be preferred, as its availability is not only incomparably wider than that of linseed, but it is also constantly improving, as it is a by-product of an increasingly popular method of extracting oil without the use of chemical solvents. The use of rapeseed cake as a high protein supplement [substitution for soya oilmeal] in the diet of dairy cows renders it possible to obtain a high yield of milk, characterised by high nutritive value, and simultaneously containing a comparatively high concentration of functional components. Further investigations are needed.

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