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Impact of a basal diet of hay and fodder beet supplemented with rapeseed, linseed and sunflowerseed on the fatty acid composition of milk fat

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Abstract

The content of fatty acids (FAs) in milk from cows fed a control diet comprised of hay ad libitum and 15 kg fodder beet, supplemented with either 1 kg ground rapeseed (RAP1), 1 or 1.4 kg ground linseed (LIN1 or LIN1.4), or 1 or 1.4 kg ground sunflowerseed (SUN1 or SUN1.4), was determined using high resolution gas chromatography. The concentration of saturated fatty acids (SFAs) in milk fat from cows fed the control diet was very high and decreased almost linearly as a function of increased daily intake of oleic, linoleic and α-linolenic acids of the oilseeds. The highest concentrations of unsaturated fatty acids (UFAs) and conjugated linoleic acids (CLAs) were found in milk from cows fed the SUN1.4 diet. A supplement of linseed induced the highest content of α-linolenic acid in milk fat. Sunflowerseed was the best of the three oilseeds studied for counterbalancing the high content of SFAs and the relatively low content of UFAs in milk fat.

Keywords: Milk fat; Fatty acid; Hay and fodder beet; Rapeseed; Linseed; Sunflowerseed

1. Introduction

During winter, hay and fodder beet are common components of a basal diet for dairy cows in the lowland regions of Europe. Few studies have been carried out to evaluate the influence of such a diet on the fatty acid composition of milk fat. Generally, the concentration of SFAs is very high in milk fat from cows fed fodder beet and hay. It is well established that an inclusion of plant or fish oil, rich in UFAs, into animal diets modifies the fatty acid composition of body tissues and products of animal origin such as milk fat. Dietary supplements of rapeseed, soybean and linseed oils (Dhiman et al., 2000; Lawless, Murphy, Harrington, Devery, & Stanton, 1998; Reklewska et al., 2002), rapeseed press cake, full-fat rapeseed or oil-rich rapeseed cake (Jahreis, Steinhart, Pfalzgraf, Flachowsky, & Schöne, 1996) or fish meal (AbuGhazaleh, Schingoethe, Hippen, Kalscheur, & Whitlock, 2002) or fish oil supplemented with sunflowerseed or flaxseed (AbuGhazaleh, Schingoethe, Hippen, & Kalscheur, 2003) have been shown to decrease the concentration of SFAs and to increase that of UFAs including conjugated linoleic acids (CLAs) in milk fat. Extruded soybeans and fish oil fed alone or in combination doubled or increased the total concentration of CLAs two- to three-fold (AbuGhazaleh et al., 2002). The effect of those supplementations is proportional to the fat dose. Chilliard, Ferlay, and Doreau (2001) reported increased CLA values of 1.5–2.7 g $100\,g^{-1}$ in milk fat from cows fed supplemented diets. Recently, Bell and Kennelly (2002) published a value of 5.63 g $100\,g^{-1}$ for the CLAs in milk fat from cows fed a patented high fat diet. However, large amounts of fat in the diet of ruminants is not recommended, as it has a negative effect on the activity of the rumen microflora, which results in a significant drop in the milk yield (Song, Huang, & Choi, 1998), and contents of milk fat (Bauman & Griinari, 2001; Dhiman et al., 2000) and milk protein (Lawless et al., 1998).

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UFAs are known to have hypocholesterolemic effects and therefore it is desirable to elevate the content of C18:1, C18:2 and C18:3 FAs and to lower the SFA content. A high content of CLAs is also advantageous because of their particular physiological properties (Banni, Murru, Angioni, Carta, & Melis, 2002). In a double-blind, randomised, cross-over clinical study, the consumption of food from animals fed extruded linseed diets led to significant modifications in the concentration of CLAs in human plasma (Weill et al., 2002).

The increase in UFAs in milk fat obtained after supplementation of a control diet with oilseed also has an effect on the rheological properties of milk products, such as improving the spreadability of butter (Frede, Precht, Pabst, & Philipzcyk, 1992) or providing a more desirable consistency (lower friability) in hard type cheese (Jaros, Ginzinger, Tschager, Leitgeb, & Rohm, 2001). Studies undertaken at our institute have also proved that the tougher consistency (harder friability) of winter cheese can be lowered by oilseed supplementation (Stoll, Sollberger, & Schaeren, 2001, 2002; Sollberger, Schaeren, & Stoll, 2001).

The aim of the present experiment was to investigate the effect on milk FA composition after supplementation of a control diet with ground oilseeds containing either high concentrations of oleic (rapeseed), linoleic (sunflowerseed) or α-linolenic acid (linseed). Many earlier studies included dietary sources of oleic, linoleic, or α-linolenic acid although the majority did not include the different fat sources in a single study.

2. Materials and methods

2.1. Cows and treatments

Twelve primiparous and 21 multiparous cows (Red Holstein, Holstein and Brown Swiss) in mid lactation were attributed to three groups depending on their lactation stage, milk yield and milk composition and randomly assigned to one of the three diets. The main characteristics of the feeding strategies and the mean values of the daily intake by the individual cows are summarised in Table 1. All animals were fed a control

Table 1
Characteristics of the different diets (mean values for all individual cows in each treatment group)[a,b]

Treatment	Control diet Hay ad libitum and 15 kg beet	Supplement to the control diet (per cow per day)				
		Ground rapeseed	Ground sunflowerseed		Ground linseed	
		1.0 kg (RAP1)	1.0 kg (SUN1)	1.4 kg (SUN1.4)	1.0 kg (LIN1)	1.4 kg (LIN1.4)
Daily intake, kg dry matter day^{-1}						
Hay	12.5	13.5	14.0	13.2	12.9	12.0
Fodder beet	3.0	3.1	2.9	3.2	3.0	3.1
Total basal diet	15.6	16.6	16.8	16.4	15.8	15.1
Oilseed	0.0	0.92	0.95	1.27	0.87	1.24
Protein concentrate	0.9	0.8	0.5	0.5	0.6	0.5
Cereal mix	4.3	2.2	2.4	1.7	2.1	1.4
Mineral mix	0.2	0.3	0.3	0.4	0.3	0.4
Total diet supplements	5.5	4.2	4.1	4.0	3.8	3.6
Total diet	21.1	20.8	20.9	20.4	19.7	18.7
Fat-, energy- and APIN, APIE-intake per day in the total ration						
Fat (g)	549	937	960	1129	775	893
NEL (MJ)	139	140	140	137	130	124
NEL, MJ kg^{-1} dry matter	6.59	6.73	6.69	6.72	6.64	6.64
Crude protein (g)	3187	3064	2915	2847	2846	2724
APIE (g)	2189	2060	1989	1899	1935	1803
APIN (g)	2072	1957	1852	1811	1818	1737
Feed efficiency						
NEL, MJ kg^{-1} ECM	3.39	3.34	3.44	3.58	3.16	3.11
APIE, g kg^{-1} ECM	59.3	53.6	53.1	53.3	51.7	49.5
APIN, g kg^{-1} ECM	55.3	50.3	48.5	50.2	47.6	46.9

[a] MJ = Megajoule.
[b] ECM: Energy corrected milk; NEL: Net energy for milk production; APIN: Absorbable protein in small intestine synthesized from degraded dietary nitrogen in the rumen; APIE: Absorbable protein in small intestine synthesized from energy available in the rumen.

diet consisting of hay ad libitum and 15 kg of fodder beet for 2 weeks. From week 3 to 5, the control diet was supplemented daily with either 1.0 kg of ground rapeseed (RAP1, 0.92 kg dry matter), sunflowerseed (SUN1, 0.95 kg DM) or linseed (LIN1, 0.87 kg DM). From week 6 to 7, the amounts of sunflowerseed or linseed were increased by 0.4 kg to 1.27 DM day^{-1} (SUN1.4) and 1.24 kg DM day^{-1} (LIN1.4), respectively. The various diets were denoted: control, RAP1, SUN1, SUN1.4, LIN1 or LIN1.4. The diets were always supplemented individually for each cow with a cereal mix and a protein concentrate based on the average milk yield, milk composition, animal body weight and feed intake of the previous week, according to INRA recommendations (Jarrige, 1989).

The stage of lactation or a low carry over from an earlier diet could modify the results of the FA composition in milk fat because of the unusual feature of the experimental design (no control group during the full length of the experiment). Nevertheless, the impact of the stage of lactation of cows in mid-lactation on the FA composition is known to be very small (Palmquist, Beaulieu, & Barbano, 1993). In our experience, although a slight carry over from the previous treatment cannot be completely excluded, a significant carry over from one treatment to another seems to be unlikely after 2 weeks feeding.

2.2. Sampling and sample treatment

Milk yield and milk composition were measured weekly and the results are given for the period when the optimal effect of the diet was realised, at week 2 (control diet), week 4 (RAP1, LIN1, SUN1) and week 7 (LIN1.4, SUN1.4). The 88 milk samples were stored at −18°C. Prior to analysis, they were centrifuged and the resulting cream was churned at ~5°C. After filtering off the resulting molten butter on a hydrophobic filter (Schleicher Schuell no 597 hy 1/2), the pure milk fat was collected.

2.3. Method of analysis

Quantitative determination of the fatty acid composition was carried out by high resolution gas chromatography with flame ionisation detection according to Collomb and Bühler (2000). After dissolution of milk or fodder fat in hexane, the glycerides were *trans*-esterified to the corresponding methyl esters of fatty acids by a solution of potassium hydroxide in methanol. The FAs were separated on a capillary column CP-Sil 88 (100 m length, 0.25 mm internal diameter, 0.20 μm film thickness) and quantified using nonanoic acid as an internal standard. The results were expressed in absolute values, as g fatty acids per 100 g fat.

The contents of fat, protein and lactose in milk were measured by mid infrared spectrometry according to IDF (2000). The fat content in oilseed was determined by the acid hydrolysis method according to the Schweizerisches Lebensmittelbuch (1982).

2.4. Statistical analysis

Analysis of variance (ANOVA) and pair wise comparisons of mean values with Fisher's LSD test were performed with Systat for Windows version 9.0 (Anonymous, 1999).

3. Results and discussion

In the following text, only the impact of the cows' intake of fat from the three supplemented oilseeds (and not of the fat of the control diet) on the fatty acid composition of milk fat will be discussed.

3.1. Characteristics of the three oilseeds

The main characteristic of the fatty acid composition of the oilseeds fed to the cows was the high concentration of oleic acid in rapeseed, linoleic acid in sunflowerseed and α-linolenic acid in linseed (Table 2). As expected, cows in the RAP1 group had the highest daily intake of oleic acid (Table 3). Except for the cows in the SUN1 or SUN1.4 groups, the daily intake of linoleic acid was low and similar in all the other diets. Cows in the SUN1 or SUN1.4 groups had the highest daily intake of linoleic acid. The daily intake of α-linolenic acid was very low in the RAP1, SUN1 or SUN1.4 groups and very high in the LIN1 or LIN1.4 groups.

3.2. Milk yield and milk composition

Milk yield and milk composition were not significantly affected by the different supplements (Table 4). However, the SUN1.4 diet gave protein and fat concentrations in the milk which were about 0.1 and 0.35 g 100 g^{-1}, respectively, lower than that in the milk from the other diets. Milk fat commonly decreases when cows are fed diets high in free oil (Banks, Clapperton, Kelly, Wilson, & Crawford, 1980) or when the diet is supplemented by PUFA oils in free form (Selner & Schultz, 1980). In contrast, when dietary oil was supplied through oilseed, milk fat content was either not affected (Mohamed, Satter, Grummer, & Ehle, 1988), increased (DePeters, Taylor, Franke, & Aguire, 1985) or decreased (Lawless et al., 1998). The *trans*-isomer C18:1 t10 FA has been shown to decrease milk fat content (Romo, Casper, Erdman, & Teter, 1996). Recently, Baumgard, Matitashvili, Corl, Dwyer, and Bauman (2002) discussed the biohydrogenation theory

of milk fat reduction and concluded that the C18:2 t10c12 CLA is also a potent inhibitor of milk fat synthesis. The mechanism by which *trans*-isomers reduced milk fat content is not yet known. Dhiman et al. (2000) suggested that, compared to pure oil supplements, the slow release of oil during ruminal digestion of oilseeds decreased the accumulation and amount of *trans*-C18:1 FA leaving the rumen and thereby led to a reduction in milk fat content.

Table 2
Fatty acid composition (g $100\,g^{-1}$ fat) of the three oilseeds used for supplementation of diet fed to cows[a]

Fatty acid (FA)	Ground rapeseed	Ground sunflowerseed	Ground linseed
C12	<0.01	<0.01	0.01
C14	0.06	0.07	0.08
C15	0.02	0.02	0.02
C16	4.07	5.68	5.30
C16:1	0.20	0.07	0.07
C17	0.04	0.04	0.05
C18	1.59	3.78	3.50
C18:1 t10	0.04	0.19	0.05
C18:1 c9	**54.67**	15.20	17.55
C18:1 c11	3.27	0.52	0.71
C18:2 c9c12	17.95	**53.62**	14.21
C18:2 c9c15	0.02	<0.01	0.09
C18:3 c6c9c12	0.05	<0.01	0.19
C18:3 c9c12c15	8.88	0.14	**46.63**
C20	0.54	0.21	0.12
C20:1 c11	1.21	0.10	0.14
C22	0.32	0.55	0.12
C24	0.09	0.17	0.08
∑saturated[b]	6.75	10.52	9.29
∑C12, C14 & C16	4.14	5.75	5.39
∑C18:1	57.98	15.91	18.32
∑C18:2	17.96	53.62	14.30
∑C18:3	8.93	0.15	46.82
∑unsaturated[c]	86.27	69.84	79.65
∑monounsaturated[d]	59.39	16.08	18.53
∑polyunsaturated[e]	26.89	53.76	61.12

[a] Bold font, indicates the most important fatty acids.
[b] C12, C14, C15, C16, C17, C18, C20, C22 and C24.
[c] C16:1, C18:1 t10, C18:1 c9, C18:1 c11, C18:2 c9c12 C18:2 c9c15, C18:3 c6c9c12, C18:3 c9c12c15 and C20:1 c11.
[d] C16:1, C18:1 t10, C18:1 c9, C18:1 c11 and C20:1 c11.
[e] C18:2 c9c12, C18:2 c9c15, C18:3 c6c9c12 and C18:3 c9c12c15.

3.3. Composition of individual fatty acids (FAs) in milk fat

The extent to which the feeding of oilseed altered the FA composition of milk depends on a number of factors including the degree of ruminal biohydrogenation, the composition of the non-lipid component of the diet, the influence of the lipid source on microbial FA synthesis and de novo synthesis of FAs in the mammary gland, the stage of lactation, and the desaturase activity in the intestine and mammary gland (Kennelly, 1996). Recently, Peterson, Kelsey, and Bauman (2002) showed that there were large differences between individual cows as regards biohydrogenation and desaturase activities; consequently, during the course of diet experiments, these differences can lead to variations in the CLA content between cows on the same diet. Owing to the large number (70) of FAs analysed, the results of the individual FAs were presented in three tables: the major individual FAs (Table 5), *trans*-FAs and CLAs (Table 6) and eicosenoic and docosenoic FAs (Table 7).

3.3.1. Influence of rapeseed, as a rich oleic acid source, on the individual fatty acids of milk fat

Supplementation of the control diet with 1 kg rapeseed which is rich in oleic acid (Table 2), resulted in important changes in the FA composition of the milk fat. The cows' daily intake of oleic acid in the RAP1 diet was high (258 g) compared to the other diets (59–106 g) (Table 3). Oleic acid from fodder in the rumen was either not hydrogenated (Morris, 1970), isomerised to C18:1 *trans*-FAs with double bonds at position 6 through 16 of the carbon chain, or hydrogenated directly to C18:0 FA (Mosley, Powell, Riley, & Jenkins, 2002). In our study, feeding the RAP1 diet resulted in lower concentrations of the SFAs C10–C16 and higher concentrations of stearic, oleic (Table 5) and minor FAs (Tables 6 and 7) compared to the control diet. Similar results were obtained by Sollberger, Schaeren, and Stoll

Table 3
Fat content of the three oilseeds (g kg^{-1} dry matter) and daily intake (g per day per cow) of the predominant unsaturated fatty acids in the diet of cows supplemented with oilseeds[a,b]

Fatty acid (FA)	Ground rapeseed	Ground sunflowerseed		Ground linseed	
	1.0 kg (RAP1)	1.0 kg (SUN1)	1.4 kg(SUN1.4)	1.0 kg(LIN1)	1.4 kg(LIN1.4)
Fat, g kg^{-1} dry matter	513	551	—	388	—
Daily intake (g)					
Oleic acid	**258**	80	106	59	84
Linoleic acid	85	**281**	**375**	48	68
α-Linolenic acid	42	1	1	**157**	**224**

[a] Bold font, indicates the most important fatty acids.
[b] See text and Table 1 for details of diets.

Table 4
The yield and composition of milk from cows fed on different diets[a,b,c]

Treatment	Control diet Hay ad libitum and 15 kg beet		Supplement to the control diet (per cow per day)									
			Ground rapeseed		Ground sunflowerseed				Ground linseed			
			1.0 kg (RAP1)		1.0 kg (SUN1)		1.4 kg (SUN1.4)		1.0 kg (LIN1)		1.4 kg (LIN1.4)	
	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x
Milk yield ECM, kg day^{-1}	30.26	5.14	30.95	3.79	29.59	3.77	27.66	3.44	29.88	6.55	28.45	5.95
Fat, g 100 g^{-1}	4.02	0.61	4.05	0.46	4.13	0.50	3.69	0.42	4.19	0.48	4.18	0.55
Protein, g 100 g^{-1}	3.35	0.28	3.39	0.24	3.38	0.17	3.26	0.18	3.37	0.18	3.29	0.25
Lactose, g 100 g^{-1}	4.90	0.17	5.00	0.18	4.92	0.16	4.92	0.17	4.98	0.16	4.99	0.14

[a] ECM = energy corrected milk.
[b] $\bar{X}$ = mean value; s_x = standard deviation.
[c] See text and Table 1 for details of diets.

Table 5
Major fatty acids of milk fat (g 100 g^{-1} fat) from cows fed different diets[e,f,g]

Fatty acid (FA)	Control diet Hay ad libitum and 15 kg beet		Supplement to the control diet (per cow per day)									
			Ground rapeseed		Ground sunflowerseed				Ground linseed			
			1.0 kg (RAP1)		1.0 kg (SUN1)		1.4 kg (SUN1.4)		1.0 kg (LIN1)		1.4 kg (LIN1.4)	
	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x
C4	3.1	0.32	3.1	0.21	3.0	0.25	3.3	0.32	3.1	0.39	3.3	0.38
C6	2.2	0.21	2.3	0.14	2.2	0.21	2.1	0.26	2.3	0.28	2.2	0.24
C8	1.4^{ab}	0.15	1.4^{ab}	0.08	1.4^{abc}	0.15	1.2^{c}	0.19	1.5^{a}	0.20	1.3^{bc}	0.18
C10	3.6^{a}	0.50	3.3^{ab}	0.21	3.1^{b}	0.46	2.5^{c}	0.48	3.5^{ab}	0.47	2.8^{c}	0.43
C12	4.5^{a}	0.70	3.8^{b}	0.30	3.6^{b}	0.53	2.8^{c}	0.51	4.0^{ab}	0.47	3.0^{c}	0.43
C14	12.0^{a}	0.92	11.3^{ab}	0.72	10.9^{b}	1.01	9.6^{c}	0.96	11.5^{ab}	0.64	9.8^{c}	0.76
C15	1.4^{a}	0.25	1.2^{b}	0.16	1.2^{b}	0.13	0.9^{c}	0.07	1.2^{b}	0.19	0.9^{c}	0.10
C16	31.1^{a}	2.35	24.5^{bc}	2.60	25.1^{b}	2.39	20.7^{d}	2.55	26.2^{b}	2.94	22.1^{cd}	2.58
C18	5.4^{c}	1.08	9.1^{ab}	2.18	8.8^{ab}	1.25	9.8^{ab}	1.36	8.7^{b}	1.45	10.7^{a}	1.94
C18:1 c9	11.3^{c}	1.96	16.6^{a}	1.78	15.9^{ab}	3.07	17.9^{a}	3.18	14.0^{b}	1.73	17.1^{a}	2.61
C18:2 c9c12	1.8^{b}	0.32	1.6^{b}	0.30	2.2^{a}	0.40	2.6^{a}	0.38	1.5^{b}	0.28	1.6^{b}	0.18
C18:3 c9c12c15	0.7^{c}	0.10	0.7^{c}	0.11	0.7^{c}	0.10	0.7^{c}	0.12	1.2^{b}	0.20	1.6^{a}	0.25

[e] $\bar{X}$ = mean value; s_x = standard deviation.
[f] c = *cis*; values within a row not showing a common superscript (a > b > c > d) differ significantly, $P \leqslant 0.01$.
[g] See text and Table 1 for details of diets.

(2001) in milk from cows fed 1 or 1.5 kg rapeseed. These authors reported a decrease in the concentration of the C10, C12, C14 and C16 FAs and an increase in the concentration of stearic and oleic acids when supplementing the control diet with rapeseed. Jaros et al. (2001) also found lower concentrations of palmitic acid and higher concentrations of stearic and oleic acids on replacing 2 kg of grains in the diet by 1 kg of shredded rapeseed. In the latter study, the ratio C16:0–C18:1, which was proposed by Ulberth (1989) as a simple indicator of fat firmness, decreased from 1.64 in milk fat from cows fed the control diet to 0.58 in milk fat from cows fed the diet supplemented with rapeseed. In our study, the ratio C16 to $\sum$C18 : 1 was reduced from 2.24 in milk fat from the control diet to 1.17 in milk fat from cows fed the RAP1 diet. Sollberger, Schaeren and Stoll (2001) obtained a similar value (2.50) for the ratio of C16 to $\sum$C18 : 1 in the control diet and found that it decreased to 1.16 in milk fat from cows fed a diet supplemented with 1 kg rapeseed and to 1.03 when the diet was supplemented with 1.5 kg rapeseed. Other authors (Jahreis et al., 1996) have also reported increased concentrations of stearic and oleic acids and reductions in SFAs in milk from cows fed rapeseed press cake, full-fat rapeseed, or oil-rich rapeseed cake. The increased oleic acid content led to a better spreadability of butter (Jahreis et al., 1996).

3.3.2. *Influence of sunflowerseed, as a rich linoleic acid source, on the individual fatty acids of milk fat*

Sunflowerseed is a rich source of linoleic acid and contains only trace amounts of α-linolenic acid (Table 2).

Table 6
Trans-C18:2 and C18:1, CLA and minor *cis*-C18:1 fatty acids of milk fat (g 100 g^{-1} fat)[f,g,h]

Fatty acid (FA)	Control diet Hay ad libitum and 15 kg beet		Supplement to the control diet (per cow per day)									
			Ground rapeseed		Ground sunflowerseed				Ground linseed			
			1.0 kg (RAP1)		1.0 kg (SUN1)		1.4 kg (SUN1.4)		1.0 kg (LIN1)		1.4 kg (LIN1.4)	
	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x
C16:1t	0.05^{d}	0.02	0.06^{cd}	0.01	0.07^{bcd}	0.02	0.17^{a}	0.04	0.06^{cd}	0.01	0.09^{b}	0.03
C18:1 t6–8	0.08^{e}	0.07	0.30^{b}	0.05	0.23^{c}	0.05	0.37^{a}	0.05	0.15^{d}	0.04	0.22^{c}	0.04
C18:1 t9	0.16^{d}	0.06	0.34^{b}	0.04	0.30^{b}	0.05	0.45^{a}	0.07	0.22^{c}	0.03	0.29^{b}	0.03
C18:1 t10–11	1.18^{c}	0.75	1.59^{c}	0.29	2.10^{b}	0.38	4.23^{a}	0.74	1.39^{c}	0.36	2.18^{b}	0.40
C18:1 t12	0.11^{d}	0.03	0.30^{c}	0.03	0.40^{b}	0.10	0.58^{a}	0.09	0.26^{c}	0.06	0.38^{a}	0.06
C18:1 t13-14 + c6-8	0.30^{d}	0.06	0.67^{c}	0.07	0.80^{c}	0.16	1.05^{b}	0.14	0.85^{c}	0.19	1.33^{a}	0.22
C18:1 t16 + c14	0.14^{d}	0.03	0.31^{c}	0.04	0.38^{b}	0.08	0.50^{a}	0.08	0.37^{bc}	0.08	0.53^{a}	0.07
C18:1 c11	0.38^{ab}	0.11	0.44^{a}	0.06	0.36^{ab}	0.11	0.34^{b}	0.07	0.32^{b}	0.06	0.32^{b}	0.04
C18:1 c12	0.12^{e}	0.02	0.17^{d}	0.02	0.39^{b}	0.09	0.64^{a}	0.09	0.21^{d}	0.04	0.31^{c}	0.04
C18:1 c13	0.05^{c}	0.03	0.07^{ab}	0.01	0.07^{ab}	0.02	0.08^{a}	0.02	0.06^{bc}	0.01	0.08^{ab}	0.01
C14:1c	1.19^{a}	0.29	1.03^{ab}	0.30	1.07^{ab}	0.19	0.99^{ab}	0.20	1.01^{ab}	0.28	0.82^{b}	0.20
C16:1c	1.39^{a}	0.34	1.09^{b}	0.32	1.09^{b}	0.25	0.93^{b}	0.15	1.02^{b}	0.25	0.90^{b}	0.19
C18:2 ttNMID	0.06^{d}	0.03	0.08^{cd}	0.01	0.08^{bc}	0.01	0.11^{b}	0.01	0.09^{bc}	0.02	0.13^{a}	0.03
C18:2c9t12 + c,c-MID + t8c13	0.20^{d}	0.03	0.27^{c}	0.02	0.31^{bc}	0.04	0.35^{ab}	0.05	0.29^{c}	0.04	0.37^{a}	0.05
C18:2 c9t13 + (t8c12)	0.11^{c}	0.03	0.21^{b}	0.03	0.25^{b}	0.06	0.33^{a}	0.08	0.24^{b}	0.05	0.38^{a}	0.07
C18:2 c9t11 (CLA)	0.51^{c}	0.20	0.62^{bc}	0.10	0.82^{b}	0.15	1.71^{a}	0.33	0.53^{c}	0.14	0.82^{a}	0.19
C18:2 t9t11 (CLA)	0.02^{b}	0.01	0.02^{b}	0.00	0.03^{b}	0.01	0.05^{a}	0.01	0.02^{b}	0.01	0.02^{a}	0.00
C18:2 t11c13 + c9c11 (CLA)	0.015^{d}	0.006	0.016^{cd}	0.005	0.017 ^{d}	0.008	0.027^{b}	0.01	0.025^{bc}	0.013	0.052^{a}	0.014
C18:2 t11c15 + t9c12	0.11^{c}	0.05	0.12^{c}	0.02	0.13^{c}	0.03	0.16^{c}	0.03	0.25^{b}	0.07	0.51^{a}	0.08

[f] CLA = conjugated linoleic acid; t = *trans*; c = *cis*; t = *trans*; tt NMID = *trans*, *trans* non-methylene interrupted diene; cc MID = *cis*, *cis* methylene interrupted diene.

[g] n = number of samples analyzed; $\bar{X}$ = mean value; s_x = standard deviation; values within a row not showing a common superscript (a > b > c > d > e) differ significantly, $P \leqslant 0.01$.

[h] See text and Table 1 for details of diets.

Table 7
Eicosenoic and docosenoic fatty acids of milk fat (g 100 g^{-1} fat)[d,e,f]

Fatty acid (FA)	Control diet Hay ad libitum and 15 kg beet		Supplement to the control diet (per cow per day)									
			Ground rapeseed		Ground sunflowerseed				Ground linseed			
			1.0 kg (RAP1)		1.0 kg (SUN1)		1.4 kg (SUN1.4)		1.0 kg (LIN1)		1.4 kg (LIN1.4)	
	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x
C20:1 c5	0.01^{b}	0.00	0.01^{b}	0.00	0.01^{b}	0.00	0.011^{a}	0.00	0.01^{b}	0.00	0.01^{b}	0.00
C20:1 c9	0.09^{c}	0.01	0.16^{a}	0.02	0.11^{bc}	0.02	0.12^{b}	0.02	0.10^{bc}	0.02	0.10^{bc}	0.01
C20:1 c11	0.03^{b}	0.01	0.07^{a}	0.01	0.03^{b}	0.01	0.04^{b}	0.01	0.03^{b}	0.00	0.03^{b}	0.00
C20:2 c,c (n−6)	0.03	0.00	0.03	0.00	0.03	0.01	0.03	0.01	0.03	0.00	0.02	0.00
C20:3 (ω6)	0.08^{ab}	0.01	0.07^{ab}	0.01	0.08^{a}	0.02	0.08^{a}	0.01	0.07^{b}	0.01	0.06^{c}	0.01
C20:3 (ω3)	0.01^{c}	0.00	0.01^{c}	0.00	0.01^{c}	0.01	0.01^{c}	0.00	0.02^{b}	0.01	0.03^{a}	0.01
C20:4 (ω6)	0.12^{a}	0.02	0.11^{ab}	0.02	0.12^{ab}	0.04	0.10^{ab}	0.01	0.10^{ab}	0.02	0.09^{b}	0.03
C20:5 EPA (ω3)	0.07^{ab}	0.01	0.07^{b}	0.02	0.07^{b}	0.01	0.05^{c}	0.01	0.07^{ab}	0.01	0.09^{a}	0.02
C22:5 DPA (ω3)	0.10	0.02	0.10	0.01	0.11	0.06	0.09	0.02	0.10	0.02	0.11	0.02
C22:6 DHA (ω3)	0.01^{b}	0.00	0.01^{b}	0.00	0.01^{b}	0.00	0.01^{b}	0.00	0.01^{b}	0.00	0.01^{a}	0.001

[d] $\bar{X}$ = mean value; s_x = standard deviation; c = *cis*; EPA = c5,c8,c11,c14,c17-eicosapentaenoic acid ; DPA = c7,c10,c13,c16,c19-docosapentaenoic acid; DHA = c4,c7,c10,c13,c16,c19-docosahexaenoic acid.

[e] Values within a row not showing a common superscript ($a > b > c$) differ significantly, $P \leqslant 0.01$.

[f] See text and Table 1 for details of diets.

The daily intake of linoleic acid for cows fed the SUN1 or SUN1.4 diet (281 and 375 g) was high compared to that for cows fed the other diets (48–85 g) (Table 3). The high concentration of linoleic acid in the SUN1 and SUN1.4 diets led to increased concentrations of this FA in milk fat. Compared to the control diet, the feeding of SUN1 or SUN1.4 diets led to increases in the concentrations of stearic, oleic and linoleic acids in milk

fat and a reduction in the concentrations of SFAs (especially from C10 to C16) (Table 5). Increased concentrations of most *trans*-(C18:1 and C18:2) and C18:1 c12 FAs were also found. Loor, Bandara, and Herbein (2002) found a high correlation between the C18:1 *trans*-isomers (t11, t13+t14, t15 and t16) in milk fat and level of linoleic acid in the diet. In vitro studies with pure cultures of rumen microorganisms also indicated that several *cis*-isomers of C18:1, especially c11 and c12, were formed during isomerization and hydrogenation of linoleic acid in the rumen (Hazlewood, Kemp, Lander, & Dawson, 1976). In our study, the concentration of the C18:1 c12 FA was highest in milk fat from cows fed the SUN1.4 diet. In milk fat from cows fed the SUN1 or SUN1.4 diets, the most interesting observation concerned the high concentrations of C18:1 t10+C18:1 t11 FAs and of C18:2 c9t11 FA. It is well known that linoleic acid is isomerised to the conjugated linoleic acid, C18:2 c9t11 by *cis*-9, *trans*-11 isomerase and then hydrogenated by *Butyrivibrio fibrisolvens* to *trans*-vaccenic FA in the rumen (Kepler, Hirons, McNeill, & Tove, 1966). These initial steps occur rapidly. The hydrogenation of *trans*-vaccenic acid to stearic acid appears to involve a different group of organisms and occurs at a slow rate (Griinari, Chouinard, & Bauman, 1997). For this reason, *trans*-vaccenic acid typically accumulates in the rumen and is therefore the main FA responsible for the formation of the CLA which occurs by desaturation of the *trans*-vaccenic acid in the mammary gland (Griinari et al., 2000). The conjugated linoleic acid, C18:2 c9t11, which is regarded as the most effective FA in cancer prevention as well as being antiatherogenic, immunomodulatory, growth-promoting and lean body mass-enhancing (Banni et al., 2002; Ip et al., 1999), generally represents more than 82% of the total CLA concentration in milk fat (Chin, Liu, Storkson, Ha, & Pariza, 1992). The high concentration (e.g. as high as 1.71 g $100\,g^{-1}$ fat) of this CLA in milk fat from cows fed SUN1.4 could provide a basis for health-promoting milk products.

Some authors described close correlations between the concentrations of *trans*-vaccenic and the CLA, C18:2 c9t11, in milk fat (Jiang, Bjoerck, Fondén, & Emanuelson, 1996). These correlations were found under experimental conditions using different oil supplements (Palmquist, 2001). Gerson, John, and King (1985) found that decreasing the proportion of fibre, and increasing the starch content, of the diet led to a slow final hydrogenation step and to an increased concentration of *trans*-vaccenic acid in the rumen as the main hydrogenation product. Palmquist and Schanbacher (1991) reported that high fat diets interfered with the terminal hydrogenation step in the rumen, and that the content of *trans*-vaccenic acid increased substantially in milk from cows fed high grain diets. According to Precht and Molkentin (1996), the concentration of *trans*-vaccenic acid normally corresponds to approximately 90% of the total concentration of the two isomers C18:1 *trans*-10 and C18:1 *trans*-11 which could not be separated under the chromatographic conditions used in the current study. In our study, the ratio of C18:2 c9t11 to *trans*-vaccenic acids (90% of the $\sum$C18:1 t10-11) was 0.48 in milk from cows fed the control diet and ranged from 0.42 to 0.45 in milk from cows fed the oilseed diets. In the study of Piperova et al. (2000), cows fed a ration supplemented with 5% soybean oil produced a milk fat with 1.70 g *trans*-vaccenic acid $100\,g^{-1}$ fatty acid methyl ester (FAME) and 0.54 g CLA $100\,g^{-1}$ FAME; this corresponded to a ratio of C18:2 c9t11 to *trans*-vaccenic of 0.32. The ratio C16 to $\sum$C18 : 1, which is an indicator of fat firmness (Ulberth, 1989), was reduced from 2.24 in the control diet to 1.20 and 0.79 in the milk from cows fed the SUN1 or SUN1.4 diets, respectively.

3.3.3. *Influence of linseed, as a rich α-linolenic acid source, on the individual fatty acids of milk fat*

In contrast to rapeseed and sunflowerseed, linseed contains α-linolenic acid as the main PUFA (Table 2). The cows' daily intake of this FA was high in LIN1 or LIN1.4 diets (157 or 224 g, respectively) compared to other diets (1–42 g) (Table 3). The high daily intake of α-linolenic acid by cows fed the LIN1 or LIN1.4 induced increases of 65% and 125%, respectively, of this FA in milk fat (Table 5). The pathway for the hydrogenation of C18:3 c9c12c15 in the rumen involves an initial isomerization to a conjugated triene (C18:3 c9t11c15), followed by reduction of double bonds at carbons 9, 15 and 11 to yield the FAs C18:2 t11c15, C18:1 t11, and C18:0, respectively (Wilde & Dawson, 1966). The α-linolenic acid does not appear to produce the conjugated linoleic acid, C18:2 c9t11, as an intermediate. Compared to the control diet, feeding LIN1 or LIN1.4 diets led to lower concentrations of C10–C16 FAs and higher concentrations of oleic acid, stearic acid, most of the *trans*-FAs (particularly C18:1 t10–11, C18:1 t13–14+c6–8 and C18:2 t11c15+t9c12) and CLAs (C18:2 c9t11, C18:2 t11c13+c9c11) in the milk fat (Table 6). Weill et al. (2002) also showed that feeding extruded linseed reduced the concentration of C16:0 and increased the concentrations of stearic, linoleic, α-linolenic acids and CLAs in milk fat. In our study, there was no significant difference in the concentration of linoleic acid between the milks from cows fed LIN1 or LIN1.4. Recently, Loor et al. (2002) analysed the concentration of C18:2 t11c15 FA in the rumen fluid from cows fed a diet supplemented with canola and soya bean oils. Their results confirmed that C18:2 t11c15 was the major C18:2 *trans*-isomer produced by hydrogenation of α-linolenic acid. In our study, the highest concentration of the combined C18:2 t11c15 and C18:2 t9c12 FAs was found in milk fat from cows fed LIN1 or LIN1.4; it was

attributed to the high concentration of C18:2 t11c15. The increase in the content of CLAs in milk fat by supplementation of a base diet with linseed was confirmed by Aii et al. (1999). Recently, AbuGhazaleh et al. (2003) reported a high increase in the content of CLAs in milk fat by supplementation of a fish oil diet with linseed. The ratio of C16:0 to $\sum$C18 : 1 was reduced from a value of 2.24 in milk from cows fed the control diet to 1.47 or 0.97 in milk from cows fed LIN1 or LIN1.4 diets, respectively.

3.3.4. *Comparison of the impact of the three oilseeds on the individual fatty acids of milk fat*

The concentration of the SFAs, C10–C16, in milk fat decreased almost linearly in the following order: LIN1 > RAP1 > SUN1 > LIN1.4 > SUN1.4 (Table 5). The concentrations of the pentadecanoic (C15) and palmitic (C16) acids were significantly ($P \leqslant 0.01$) higher in milk fat from cows fed the control diet than in milk fat from cows fed diets supplemented with each of the oilseeds. The lowest concentrations of these two FAs were found in milk from cows fed LIN1.4 or SUN1.4. The concentration of stearic acid was highest in milk from cows fed a diet supplemented with LIN1.4, i.e. 81% higher than that of the control diet. The high concentration of C18 in milk fat from cows fed LIN1.4 is associated with the high biohydrogenation activity in the rumen of the cow which is stimulated by feeding lipids rich in C18-UFAs (Dhiman, Zanten, & Satter, 1995).

Oleic acid (C18:1 c9) was present at highest concentration in milk fat from cows supplemented with SUN1.4 and was present at a higher concentration in milk fat from cows fed RAP1 than in milk fat from cows fed LIN1 or SUN1, in accordance with the C18:1 cows' daily intake. Increased dietary supply of C18:2 and C18:3 has also been shown to increase the concentration of C18:1 in milk through ruminal biohydrogenation (Dhiman et al., 1995). The concentration of linoleic acid (C18:2 c9c12) was highest in milk from cows fed SUN1 or SUN1.4, in accordance with the cows' daily intake of this FA (281 and 375 g, respectively). Similar concentrations of this FA were found in milk from cows fed the control diet or diets supplemented with each of the other oilseeds. Dhiman et al. (2000) also found a relatively high increase in the concentration of C18:2 in milk fat by feeding raw soybeans (linoleic acid concentration in soybean fat: 50.7 g 100 g^{-1} FAME), compared with other treatments (e.g. soybean or linseed oils); this result was confirmed by AbuGhazaleh et al. (2002) with extruded soybeans. In our study, the concentration of α-linolenic acid (C18:3 c9c12c15) was only modified in milk from cows fed linseed, a trend in accordance with the high daily intake of this FA in cows fed LIN1 or LIN1.4 (daily intake: 157 and 224 g, respectively). Reklewska et al. (2002) found an increase of about 30% in the concentration of α-linolenic acid in milk from cows fed low levels of linseed (daily intake: 21 g).

The concentration of most of the *trans*-FAs generally increased in parallel with the daily intake of oleic, linoleic and α-linolenic acids, in the LIN1, RAP1, SUN1, LIN1.4 or SUN1.4 diets, respectively (Table 6). However, at oilseed intakes of 1 kg, the highest concentration was found in milk from cows fed LIN1 for the C18:2 t11c15 + C18:2 t9c12 and C18:1 t13-14 + C18:1 c6-8 FAs.

All the eicosenoic and docosenoic FAs were present at very low concentrations in all milks (Table 7). However, the eicosenoic and docosenoic acids present at highest concentration in milks from cows fed the different diets were: C20:4 (ω6) with the control diet; C20:1 *cis*-9 and *cis*-11 with the RAP1 diet; C20:3 (ω3), 20:5 c5c8c11c14c17 (eicosapentaenoic acid, EPA) and C22:6 c4c7c10c13c16c19 (docosapentaenoic acid, DHA) with the LIN1.4 diet. Plant seed oils usually contain C20:1 *cis*-11, often abbreviated as C20:1, as the major eicosanoic FA isomer (Aitzetmüller, 1999). In our study, the low concentration of all eicosenoic and docosenoic FAs found in milk fat was in agreement with the results reported by Mansbridge and Blake (1997). The increase in the concentration of very long chain FAs in milk fat, such as EPA and DHA, can be better realized by feeding fish oil (Chilliard et al., 2001).

3.4. *Comparison of the impact of the three oilseeds on groups of fatty acids of milk fat*

The concentrations of groups of fatty acids found in milk fat from cows fed the different diets are presented in Table 8. Here again, the concentration of the SFAs decreased while that of UFAs, *trans*-FAs excluding CLAs and C18:1 *t* FAs, increased almost linearly as the diet was changed from LIN1 to RAP1 to SUN1 to LIN1.4 or to SUN1.4. Since high concentrations of dietary C12, C14 and C16 SFAs are considered as risk factors for cardiovascular diseases, the FA composition of milk fat from cows fed hay ad libitum and 15 kg fodder beet (control diet) should be considered negatively from a nutritional point of view. Hence, a reduction in the concentration of these FAs in milk fat, through supplementation of the fodder with oilseed, improved the quality of the milk. The highest increase in the concentration of the C18:1 *t* FAs in milk fat when feeding the SUN1.4 diet was related to the highest daily intake of linoleic acid with this diet. Similarly, the highest increase in the concentration of the C18:2 *t* FAs (excluding CLAs) in milk fat when feeding the LIN1.4 diet was related to the highest daily intake of α-linolenic acid.

In the current study, diet did not affect the total concentration of branched chain FAs in the milk fat. Lund (1991) suggested that feeding rations low in energy

Table 8
Groups of fatty acids in milk fat (g 100 g^{-1} fat)[e,f,r]

Fatty acid (FA)	Control diet Hay ad libitum and 15 kg beet		Supplement to the control (diet per cow per day)									
			Ground rapeseed		Ground sunflowerseed				Ground linseed			
			1.0 kg (RAP1)		1.0 kg (SUN1)		1.4 kg (SUN1.4)		1.0 kg (LIN1)		1.4 kg (LIN1.4)	
	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x	$\bar{X}$	s_x
∑saturated[g]	67.7[a]	2.77	62. 7[b]	1.54	62.0[bc]	3.26	55.5[d]	3.86	64.9[b]	2.13	59.1[c]	1.57
∑C12, C14 & C16	47.6[a]	2.72	39.6[b]	3.06	39.5[b]	3.30	33.1[c]	3.73	41.6[b]	2.91	35.0[c]	2.31
∑branched FA[h]	2.1	0.17	1.9	0.14	2.0	0.18	1.9	0.13	1.9	0.22	2.0	0.19
∑unsaturated[i]	21.1[c]	2.70	27.9[b]	1.80	28.9[b]	3.98	35.2[a]	4.59	25.1[b]	2.37	31.1[ab]	2.56
∑monounsaturated[j]	17.1[d]	2.26	23.7[b]	1.69	23.8[b]	3.43	28.7[a]	3.99	20.5[c]	1.91	25.1[b]	2.51
∑polyunsaturated[k]	4.0[c]	0.60	4.2[bc]	0.45	5.1[b]	0.68	6.4[a]	0.81	4.6[b]	0.71	6.0[a]	0.61
∑CLA[l]	0.6[c]	0.21	0.7[bc]	0.11	0.9[b]	0.16	1.8[a]	0.34	0.6[c]	0.15	0.9[b]	0.20
∑*trans* without CLA[m]	2.5[d]	1.10	4.1[c]	0.42	4.8[c]	0.78	7.9[a]	1.05	3.9[c]	0.79	6.0[b]	0.85
∑C18:1 t[n]	1.9[d]	0.96	3.3[c]	0.39	3.9[b]	0.69	6.8[a]	0.94	2.9[c]	0.62	4.5[b]	0.64
∑C18:2t without CLA t[o]	0.5[d]	0.13	0.7[c]	0.05	0.8[c]	0.11	1.0[b]	0.14	0.9[bc]	0.17	1.4[a]	0.22
∑C18:1	13.9[d]	2.24	20.9[bc]	2.06	21.0 [bc]	3.33	26.2[a]	3.97	17.8[c]	1.93	22.8[b]	2.47
∑C18:2	2.8[c]	0.51	3.0[c]	0.31	3.9[b]	0.57	5.3[a]	0.74	3.0[c]	0.49	4.0[b]	0.47
∑ω3[p]	1.1[c]	0.15	1.1[c]	0.14	1.1[c]	0.20	1.1[c]	0.15	1.7[b]	0.30	2.5[a]	0.30
∑ω6[q]	2.4[d]	0.40	2.6[cd]	0.34	3.6[b]	0.50	4.4[a]	0.52	2.5[cd]	0.40	2.9[c]	0.23

[e] $\bar{X}$ = mean value; s_x = standard deviation; ∑ = sum of the concentrations; CLA = conjugated linoleic acid ; t = *trans*; tt NMID = *trans*, *trans* non-methylene interrupted diene ; cc MID = *cis*, *cis* methylene interrupted diene; EPA = c5,c8,c11,c14,c17-eicosapentaenoic acid; DPA = c7,c10,c13, c16,c19-docosapentaenoic acid; DHA = c4,c7,c10,c13,c16,c19-docosahexaenoic acid.

[f] n = number of samples analyzed; values within a row not showing a common superscript (a > b > c > d) differ significantly, $P \leqslant 0.01$.

[g] C4, C5, C6, C7, C8, C10, C12, C12 iso, C12 aiso, C13 iso, C14, C14 iso, C14 aiso, C15, C15 iso, C16, C16 iso, C16 aiso ; C17, C17 iso, C17 aiso, C18, C19, C20, C22.

[h] C12 iso, C12 aiso, C13 iso, C14 iso, C14 aiso, C15 iso, C16 iso, C16 aiso, C17 iso, C17 aiso.

[i] C10:1, C14:1 ct, C16:1 ct, C17:1 t, C18:1 t4, C18:1 t5, C18:1 t6-8, C8:1 t9, C18:1 t10-11, C18:1 t12, C18:1 t13-14 + c6-8, C18:1 c9, C18:1 c11, C18:1 c12, C18:1 c13, C18:1 t16 + c14, C18:2 ttNMID, C18:2 t9t12, C18:2 c9t13 + (t8c12), C18:2 c9t12 + (ccMID + t8c13), C18:2 t11c15 + t9c12, C18:2 c9c12, C18:2 c9c15, C20:1 t, C18:3 c6c9c12, C20:1 c5, C20:1 c9, C20:1 c11, C18:3 c9c12c15, C18:2 c9t11, C18:2 t11c13 + c9c11, C18:2 t9t11, C20:2 c,c (ω-6), C20:3 (ω-6), C20:3 (ω-3), C20:4 (ω-6), C20:5 (EPA) (ω-3), C22:5 (DPA) (ω-3), C22:6 (DHA) (ω-3).

[j] C10:1, C14:1 ct, C16:1 ct, C17:1 t, C18:1 t4, C18:1 t5, C18:1 t6-8, C8:1 t9, C18:1 t10-11, C18:1 t12, C18:1 t13-14 + c6-8, C18:1 c9, C18:1 c11, C18:1 c12, C18:1 c13, C18:1 t16 + c14, C20:1 t, C20:1 c5, C20:1 c9, C20:1 c11.

[k] C18:2 ttNMID, C18:2 t9t12, C18:2 c9t13 + (t8c12), C18:2 c9t12 + (ccMID + t8c13), C18:2 t11c15 + t9c12, C18:2 c9c12, C18:2 c9c15, C18:3 c6c9c12, C18:3 c9c12c15, C18:2 c9t11, C18:2 t11c13 + c9c11, C18:2 t9t11, C20:2 cc (ω-6), C20:3 (ω-6), C20:3 (ω-3), C20:4 (ω-6), C20:5 (EPA) (ω-3), C22:5 (DPA) (ω-3), C22:6 (DHA) (ω-3).

[l] CLA total (∑C18:2 –c9t11, –t11c13, -c9c11, -t9t11)

[m] C14:1t, C16:1t, C17:1 t, C20:1t, C18:1 *trans* + C18:2 *trans*.

[n] C18:1 (∑-t4, - t5, -t6-8, -t9, -t10-11, -t12, -t13-14).

[o] C18:2 ttNMID, C18:2 t9t12, C18:2 c9t13 + (t8c12), C18:2 c9t12 + (ccMID + t8c13), C18:2 t11c15 + t9c12.

[p] C18:2 t11c15, C18:2 c9c15, C18:3 c9c12c15, C20:3 (ω-3), C20:5 (EPA) (ω-3), C22:5 (DPA) (ω-3), C22:6 (DHA) (ω-3).

[q] C18:1 t12, C18:1 c12, C18:2 t19t12, C18:2 c9t12, C18:2 c9c12, C18:3 c6c9c12, C20:2 cc, C20:3 (ω-6), C20:4 (ω-6).

[r] See text and Table 1 for details of diets.

(starch) and rich in fibre led to a more intensive activity of rumen bacteria, as reflected, *inter alia*, by a high percentage of branched-chain fatty acids in the milk fat.

The concentrations of CLAs in milk fat did not significantly increase by feeding the RAP1 and LIN1 diets, despite the high intakes of linoleic acid (85 or 48 g, respectively) and of α-linolenic acid (42 or 157 g, respectively) (Table 3) of these diets. Feeding the LIN1.4 or SUN1 diets gave similar levels of CLAs (0.89 and 0.87 g 100 g^{-1} fat, respectively) despite very different intakes of linoleic acid (68 or 281 g, respectively) or α-linolenic acid (224 or 1 g, respectively). When the diet was changed from SUN1 to SUN1.4, a 33% increase in the daily intake of linoleic acid (from 281 to 375 g; α-linolenic acid: 1 g) increased the total CLA content by a factor of 2 (from 0.87 to 1.79 g 100 g^{-1} fat, respectively). Dhiman et al. (2000) observed the same effect in milk from cows fed a soybean oil supplement and concluded that it may be caused by incomplete biohydrogenation of the CLA in the rumen and its escape from the rumen to the lower digestive tract. Indeed, UFAs were generally converted in the rumen to saturated FAs within a short time through ruminal biohydrogenation but the extent of lipolysis and biohydrogenation in the rumen decreased with increasing amounts of substrate (Jenkins, 1993).

Compared to the control diet, a similar total concentration of ω3 FAs was found in milk fat when

feeding the RAP1, SUN1 or SUN1.4 diets but the concentration of these FAs increased to much higher values when feeding the LIN1 or LIN1.4 diets (Table 8). The total concentration of the ω6 FAs was similar in milk fat from cows fed the control diet, LIN1 or RAP1 diets, increased to slightly higher values when feeding the LIN1.4 diet, and to much higher values when feeding the SUN1 or SUN1.4 diets. The ratio of ω3−ω6 FAs in milk fat from cows fed hay ad libitum and fodder beet (0.43) was not changed when feeding the RAP1 diet (0.41). It increased to 0.69 or 0.86 when feeding the LIN1 or LIN1.4 diets, respectively, and decreased to values of 0.30 or 0.24 when feeding the SUN1 or SUN1.4 diets, respectively. A recommended ratio of ω3−ω6 FA of <0.20 is generally considered highly valuable from a nutritional point of view (Deutsche Gesellschaft für Ernährung, Österreichische Gesellschaft für Ernährung, Schweizerische Gesellschaft für Ernährungsforschung, Schweizerische Vereinigung für Ernährung, 2000). In our study, the lowest ratio of ω3–ω6 FAs was obtained in milk fat from cows fed the SUN1.4 diet; however, this value was still higher than the recommended maximum value.

4. Conclusions

Supplementation of a basic diet, consisting of hay ad libitum and 15 kg fodder beet, with oilseeds improved the milk quality from a nutritional point of view. Sunflowerseed resulted in the greatest nutritional enhancement of milk fat composition, owing to its high content of linoleic acid. Compared to the control diet, sunflowerseed supplementation resulted in a large reduction in the content of saturated FAs and an increase in the levels of MUFAs and PUFAs. The highest content of ω3 FAs in milk fat was, however, obtained when supplementing the control diet with linseed. The nutritional enhancement (reduction in the concentration of the SFAs and increases in MUFAs, PUFAs and CLAs) was, however, dependent of the total dietary intake of fat. For example, a 33% increase in the daily intake of linoleic acid (from 281 to 375 g; α-linolenic acid 1 g) increased the total CLA content by a factor of 2 (from 0.87 to 1.79 g 100 g^{-1} fat, respectively). This effect may be caused by incomplete biohydrogenation of the CLAs in the rumen and its escape from the rumen to the lower digestive tract.

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