Dietary Conjugated Linoleic Acids Alter Adipose Tissue and Milk Lipids of Pregnant and Lactating Sows

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ABSTRACT Conjugated linoleic acids (CLA) have been shown to affect fatty acid synthesis in various tissues. The objective of the study was to compare the effect of a commercial source of CLA with a linoleic acid–enriched oil (LA), supplied to 12 multiparous sows during gestation and lactation, on adipose tissue and milk fatty acid composition. The CLA isomers detected in the CLA oil were (in order of magnitude) *c*9,*t*11; *t*10,*c*12; *c*9,*c*11; *t*9,*t*11/*t*10,*t*12 and *c*10,*c*12 and amounted to 58.9 g/100 g fat. Biopsies were taken from the backfat on d 7 and 97 of gestation and milk samples were collected on d 2, 9, 16 and 23 after farrowing. Collection of colostrum and mature milk samples took place at 1100 h for sows who farrowed in the morning or at 1500 h for those who farrowed in the afternoon. All major CLA isomers in the supplement were transferred to the tissue and milk fat and, compared with the LA group, significantly increased saturated fatty acid and decreased monounsaturated fatty acid levels in the tissue and milk. These findings suggest a distinct involvement of CLA in the de novo fatty acid synthesis and desaturation process in the adipose tissue and mammary gland. Estimated transfer efficiency of dietary CLA isomers was 41–52% for the backfat and 55–69% for the mature milk. The incorporation and uptake efficiency seemed to be selective with the highest values found for *c*9,*t*11-CLA. Overall, dietary CLA supplementation of sows during gestation and lactation markedly altered backfat and milk fatty acid composition. J. Nutr. 130: 2292–2298, 2000.

KEY WORDS: • *sows* • *conjugated linoleic acids* • *gestation and lactation* • *milk* • *adipose tissue*

Conjugated linoleic acids (CLA),[1] a naturally occurring group of dienoic derivatives of linoleic acid, have been shown to have a variety of effects in animal models (Belury and Kempa-Steczko 1997, Chin et al. 1994, Dugan et al. 1999, West et al. 1998). In addition to the reported anticarcinogenic activity (Ip et al. 1994, Scimeca et al. 1994), the protective effects against atherosclerosis (Lee et al. 1994) and prevention of endotoxin-induced growth suppression (Miller et al. 1994), interest in CLA has grown because of additional health-related effects. Data from rodents indicated that CLA reduced body fat by several mechanisms, including a reduced energy intake and increased metabolic rate (West et al. 1998). Experimental data of Dugan et al. (1997) suggested that CLA acted as a repartitioning agent in pigs. Recently, Ostrowska et al. (1999) confirmed these effects and found a dose-dependent quadratic effect on lean tissue deposition, which was maximized at 5 g/kg dietary CLA.

Additionally, consumption of CLA during lactation has been shown to increase the concentration of CLA in the rat's milk (Chin et al. 1994). In dairy cows, infusion of CLA into the abomasum altered milk fatty acid composition and markedly depressed the total content and yield of milk fat (Chouinard et al. 1999, Loor and Herbein 1998). Compared with ruminants, sow's milk is devoid of short-chain fatty acids and CLA; instead, it is dominated by oleic, palmitic and linoleic acids (Darragh and Moughan 1998, Jahreis et al. 1999). The levels of those fatty acids have been shown to be affected most by dietary CLA (Loor and Herbein 1998).

Because the fatty acid composition of colostral and mature milk depends on the type of fat supplied by the diet (Pettersen and Opstvedt 1991) and in view of the results from studies in cows and rats, it seems likely that supplementing the sow diet with CLA may result in CLA enrichment of sow's milk. Because milk is the sole or main source of nutrients for piglets during the first 2–4 wk of life, it is important that the nutrients in milk meet the substantial requirements for structural and functional development of the rapidly growing neonate. Essential fatty acids (EFA) of the (n-3) and (n-6) families are particularly important for the development of the brain and retina and are integral constituents of the central nervous system (Alessandri et al. 1998, Goustard et al. 1999). Because of the many similarities between pigs and humans (Purvis et al. 1982), results of dietary supplementation with CLA in sows may serve as a model for the effects of CLA supplementation on human milk composition. Therefore, the objectives of this study were to determine the influence of a commercial source of CLA supplied in the gestation and lactation period on the incorporation rate into adipose tissue during pregnancy and into milk fat during lactation.

MATERIALS AND METHODS

Experimental diets and animals. Two basal gestation and lactation diets were formulated; they were supplemented with either a

[1] Abbreviations used: CLA, conjugated linoleic acids; FAME, fatty acid methyl esters; LA, linoleic acid–enriched oil; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids.

0022-3166/00 $3.00
Manuscript received 7 January 2000. Initial review completed 3 March 2000. Revision accepted 21 April 2000.

linoleic acid (LA)- or CLA- enriched oil (2 g/100 g diet) (**Table 1**). A commercial source of CLA was used to supplement the diet (SELIN-CLA, Grünau Jllertissen GmbH, Germany). The LA was derived from sunflower oil and served as the source material to produce CLA. The basal diets contained no fat supplements and were formulated to meet Swiss requirements for sows during gestation and lactation (Boltshauser et al. 1993).

Multiparous sows ($n = 12$; Swiss Large White) were mated to three boars (Swiss Large White) by artificial insemination and assigned on the day of mating to one of the two experimental treatments (LA; $n = 5$, CLA; $n = 7$). The animals were blocked by body weights into five blocks with an extra pig in two blocks. Treatment was assigned randomly within block with the stipulation that the extra pigs be assigned to the CLA treatment. Sows were fed individually twice a day and had free access to water. Until farrowing, the total daily ration varied from 2.8 to 3.5 kg. The amount was adjusted according to a body condition score determined weekly. In the 35-d lactation period, all pigs were fed 4.5 kg of the lactation diet; for each suckling pig exceeding a litter size of eight, an additional 200 g feed was offered. Feed consumption for each sow during gestation and lactation was recorded, as were the number of piglets born per sow, birth weight and weaning weights. Litters were not adjusted in size. Two sows were removed from the study after farrowing (one from each treatment) for medical reasons unrelated to the dietary treatments. All procedures involving animals were approved by the Swiss Federal Committee for Animal Care and Use.

TABLE 1

Ingredient composition of gestation and lactation diets[1,2]

	Gestation diet		Lactation diet	
Item	LA	CLA	LA	CLA
		g/kg		
Barley	480.00		520.00	
Wheat			150.00	
Corn			100.00	
Whole corn plant	200.00			
Dry sugar beet pulp	100.00			
Oat	80.00		40.00	
Wheat bran	30.00		12.00	
Soybean meal	10.00		50.00	
Potato protein			20.00	
Yeast	10.00		10.00	
Blood meal			26.00	
Meat and bone meal	50.26		32.00	
Calcium carbonate	5.84		6.70	
NaCl	4.14		4.00	
Lysine · HCl	1.22		2.70	
DL Methionine	0.06			
L Threonine (98%)	0.48		0.60	
Vitamin-mineral premix[3]	4.00		4.00	
Color grit[4]	2.00		2.00	
Linoleic acid (60%)[5]	20.00		20.00	
CLA[6]		20.00		20.00

[1] Gestation diet was formulated to contain 12 MJ digestible energy, 10 g crude protein, 0.50 g lysine, 0.34 g methionine-cystine, 0.42 g threonine and 0.08 g tryptophan per MJ digestible energy.

[2] Lactation diet was formulated to contain 13.5 MJ digestible energy, 12 g crude protein, 0.70 g lysine, 0.39 g methionine-cystine, 0.49 g threonine and 0.13 g tryptophan per MJ digestible energy.

[3] Supplied the following nutrients per kilogram of diet: 1.2 mg all-*trans* retinol, 0.006 mg cholecalciferol, 9.9 mg vitamin E, 2.8 mg riboflavin, 1.3 mg vitamin B-6, 0.015 mg vitamin B-12, 0.2 mg vitamin K_3, 102 mg pantothenic acid, 10 mg niacin, 0.48 mg folic acid, 84 mg Fe as Fe-sulfate, 0.56 mg I as $Ca(IO)_3$, 0.2 mg Se as Na_2Se, 9.2 mg Cu as $CuSO_4$, 81 mg Zn as ZnO_2, 2.5 mg Mn as MnO_2, 196 g choline, 0.99 mg biotin.

[4] Color supplement added to the diets to avoid feeding mistakes.

[5] The linoleic-enriched oil was derived from sunflower oil and served as source material to produce the CLA-enriched oil (SELIN-CLA: Grünau Jllertissen GmbH, Germany). The fatty acid composition of the linoleic enriched oil was (g/100 g total fatty acids): myristic [14:0], 0.14; palmitic [16:0], 5.59; stearic [18:0], 2.37; eicosanoic [20:0], 0.24; behenic [22:0], 0.42; palmitoleic [16:1(n-7)], 0.13; oleic [18:1(n-9)], 23.86; eicosenoic [20:1(n9)], 0.18; erucic [22:1(n-9)], 0.21; linoleic [18:2(n-6)], 65.79; and linolenic acid [18:3(n-3)], 0.75.

[6] The fatty acid composition of the CLA enriched oil (SELIN-CLA) was (g/100 g total fatty acids): myristic [14:0], 0.24; palmitic [16:0], 4.37; stearic [18:0], 1.67; eicosanoic [20:0], 0.18; behenic [22:0], 0.27; palmitoleic [16:1(n-7)], 0.17; oleic [18:1(n-9)], 29.48; eicosenoic [22:1(n-9)], 0.13; and linoleic [18:2(n-6)], 3.99. The following CLA isomers were detected: *cis* (*c*), *trans* (*t*) *c*9, *t*11, 20.33; *c*9,*c*11, 5.59; *t*10,*c*12, 21.73; *c*10,*c*12, 1.33; and *t*9,*t*11/*t*10,*t*12, 9.96.

Measurements and sampling. A shot-biopsy was taken from the backfat (5 cm caudal to the last rib and also 5 cm off the midline) on d 7 and 97 of gestation. Biopsies were frozen immediately and stored at −20°C. On d 2 (colostrum), 9, 16 and 23 (mature milk) after farrowing, milk samples were collected. The milk collections took place at 1100 h for sows who farrowed in the morning or at 1500 h for those who farrowed in the afternoon. Milk (10 mL) was obtained by milking several udders after milk let-down was induced by intramuscular injection of 3 mL oxytocin (Oxytocin-20, Graeub, Switzerland). Samples were frozen immediately for lipid extraction and fatty acid analysis.

Sample analysis. Milk and adipose tissue lipids were extracted as described by Winter (1963) with minor modifications. Briefly, 5 g of milk and 300 mg of adipose tissue were homogenized in 1 mL of triundecanine (internal standard) and 60 mL of dichloromethane/methanol (1:2, v/v) for 30 s. After 15 min, the filtered sample was added to 1 mL $MgCl_2$ (20 g/L) and 20 mL water. The organic phase containing the lipid extract was removed and reduced in volume under vacuum (550×10^2 Pa at 40°C). The fatty acid methyl esters (FAME) were prepared by transesterification by methanolic sodium hydroxide and boron trifluoride according to the method of Metcalfe and Smith (1961). The FAME were determined using a gas chromatograph (HP 5860 A GC; Hewlett-Packard, Urdora, Switzerland), equipped with a flame ionization detector and separated on a 30 m × 0.32 mm i. d. Supelcowax TM 10 fused-silica capillary column (Supelco, Bellefonte, PA). The oven temperature was as follows: initial temperature 170°C for 1 min; raised to 250°C at 2.5°C/min; 250°C held for 7 min. The detection temperature was at 250°C and split at 250°C.

From the same lipid extract CLA-FAME were separated on the same gas chromatograph mentioned above but using different oven temperatures, i.e., initial temperature 200°C for 15 min; raised to 250°C with 20°C/min; 250°C held for 10 min. The detection temperature was at 250°C and split at 250°C. CLA standards (Matreya, Pleasant Gap, PA) and CRM 163 (Commission of the European Communities, Community Bureau of Reference, Brussels, Belgium) were used to validate the measurements. The following CLA isomers were measured: *cis*9,*trans*11 (*c*9,*t*11)-, *trans*10,*cis*12 (*t*10,*c*12)-, *cis*9,*cis*11 (*c*9,*c*11)- and *cis*10,*cis*12 (*c*10,*c*12)-18:2. The *trans*9,*trans*11 (*t*9,*t*11) and *trans*10,*trans*12 (*t*10,*t*12)-18:2 could not be separated and are reported as the sum of both isomers.

Statistical analysis. The fatty acid profile of the adipose tissue samples (biopsies) and milk were analyzed with the PROC MIXED procedure of SAS (1998) with treatment and time as fixed effects and block (block = sow's body weight at mating) as random factor (Littell et al. 1998). Data for preweaning growth were analyzed by the General Linear Models procedure of SAS (1998) with treatment as fixed effect and block as random factor. Differences with probability levels of $P \leq 0.05$ were considered significant. Data are reported as least-square means ± SEM.

RESULTS

The oil added to the diet LA was composed mainly of linoleic [18:2(n-6); 65.79 g/100 g total fatty acid] and oleic acid [18:1(n-9); 23.86 g/100 g total fatty acid]. The CLA isomers detected in the CLA-enriched oil were *c*9,*t*11; *t*10,*c*12; *c*9,*c*11; *c*10,*c*12 and *t*9,*t*11/*t*10,*t*12 and amounted to 58.9 g/100 g of total fatty acids. The fatty acid profile of the dietary lipid

TABLE 2

Lipid content and fatty acid composition of gestation and lactation diets[1]

Item[2]	Gestation diet		Lactation diet	
	LA	CLA	LA	CLA
Lipid, *g/kg diet*	45.07	46.98	49.40	49.15
	g/100 g total fatty acids			
14:0 (myristic)	0.44	0.44	0.34	0.32
16:0 (palmitic)	15.18	13.29	15.43	12.69
18:0 (stearic)	4.74	3.04	4.79	2.88
20:0 (eicosanoic)	0.35	0.82	0.40	0.32
22:0 (behenic)	0.72	0.45	0.44	0.24
SFA	21.43	18.03	21.41	16.45
16:1(n-7) (palmitoleic)	0.63	0.60	0.48	0.43
18:1(n-9) (oleic)	25.60	27.30	26.69	27.10
20:1(n-9) (eicosenoic)	0.56	0.56	0.57	0.37
22:1(n-9) (erucic)	0.66	0.45	ND[3]	ND
MUFA	27.45	28.91	27.74	27.90
18:2(n-6) (linoleic)	48.88	26.06	48.46	28.41
18:3(n-3) (α-linolenic)	2.23	2.04	2.38	2.46
CLA-isomers[4]				
*c*9,*t*11	ND	8.34	ND	8.37
*c*9,*c*11	ND	2.52	ND	2.41
*t*10,*c*12	ND	8.75	ND	8.80
*c*10,*c*12	ND	0.62	ND	0.60
*t*9,*t*11/*t*10,*t*12	ND	4.74	ND	4.61
PUFA	51.12	53.06	50.85	55.65
16:1(n-7)/16:0	0.04	0.05	0.03	0.03
18:1(n-9)/18:0	5.40	8.99	5.57	9.42

[1] Only fatty acids that accounted for ≥0.1 g/100 g total are presented.

[2] Fatty acids were designated by the number of carbon atoms followed by the number of double bonds. The position of the first double bond relative to the methyl (n) end of the molecule was also indicated. The sums of the main fatty acid series are represented as follows: SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.

[3] ND, not detected.

[4] *c,* cis; *t,* trans.

reflected the fat supplementation (**Table 2**). The CLA gestation and lactation diets were lower in palmitic (16:0), stearic (18:0) and linoleic acid and slightly higher in oleic acid than the respective LA diets. None of the measured CLA isomers were detected in the LA diets.

Birth weights and weaning weights. There were no significant differences in feed consumption, weight gain (gestation) or weight loss (lactation) by sows among treatment groups (data not shown). Similarly, pig birth weights (1.56, 1.58 ± 0.13kg; $P = 0.87$, for LA and CLA groups, respectively) and weaning weights (7.91, 8.26 ± 0.20 kg; $P = 0.32$, for LA and CLA groups, respectively) did not differ between treatments.

Fatty acid composition of the backfat. The fatty acid profile of the backfat biopsies at d 7 and 97 of gestation are presented in **Table 3**. The content of the main fatty acids at d 7 of gestation did not differ between treatments. Inclusion of the CLA in the gestation diet increased the level of myristic acid (14:0), palmitic acid and total SFA [treatment (T) × gestation day (D) interaction: $P \leq 0.01$ for each], whereas that of oleic acid and total monounsaturated fatty acids (MUFA) (T × D interaction: $P \leq 0.02$ for each) were decreased. The eicosanoic (20:0), palmitoleic [16:1(n-7)] and eicosenoic acid [20:1(n-9)] levels were not affected by dietary CLA. At d 97 of gestation, the contents of linoleic and arachidonic acid [(20:4(n-6)] were significantly greater in the LA than in the CLA group (T x D interaction: $P \leq 0.05$ for each), and α-linolenic acid [18:3(n-3)] concentration tended to be lower only in the CLA group (T × D interaction: $P = 0.07$).

At d 7 of gestation, sows had no detectable amounts of CLA isomers in the backfat. After 97 d of gestation, CLA isomers supplied by the diet were incorporated. The most abundant was *c*9,*t*11 and in decreasing order *t*10,*c*12 > *t*9,*t*11/*t*10,*t*12 > *c*9,*c*11-18:2. The *c*10,*c*12 CLA isomer was present only at trace levels.

Fatty acid composition of colostrum and mature milk. Regardless of dietary treatment, mature milk had markedly more SFA and less polyunsaturated fatty acids (PUFA) compared with colostrum [lactation week (W) effect: $P < 0.01$ for each], whereas total MUFA level did not differ between colostral and mature milk (W effect: $P = 0.20$; **Table 4**). The presence of CLA in the lactation diet increased the stearic and eicosenoic acid contents (T effect: $P \leq 0.04$ for each) whereas those of linoleic and arachidonic acid were lower in the colostrum and mature milk (T effect: $P < 0.01$ for each); myristic acid was increased and palmitoleic acid was lower only in the mature milk (T × W effect: $P \leq 0.04$ for each). The levels of palmitic, oleic acid and α-linolenic acids did not differ between the two treatment groups, but were lower in the mature milk than in the colostrum (W effect: $P < 0.01$ for each).

In treatment LA, none of the measured CLA isomers were detected in the milk (Table 4). In contrast, sows of the CLA treatment group excreted CLA in the colostral and mature milk. Total CLA concentration in the colostrum was significantly higher than in the mature milk (W effect: $P < 0.01$). The level of the *c*9,*t*11 and the *trans*,*trans* isomers (*t*9,*t*11/*t*10,*t*12) in the mature milk was higher (W effect: $P \leq 0.02$ for each), whereas that of the *c*9,*c*11 and *t*10,*c*12 was lower than in the colostrum (W effect: $P < 0.01$ for each). The *c*10,*c*12 isomer was present in the colostrum, but not in the mature milk.

DISCUSSION

The main findings of this study relate to fat metabolism, which seemed to be affected markedly by the dietary CLA supply. The relationship between dietary fat and the fatty acid profile in swine tissues is well documented (Camara et al. 1996, Morgan et al. 1992, Mourot et al. 1994). Under fattening conditions and therefore positive energy balance, dietary polyunsaturated fats of the (n-6) and (n-3) families are readily incorporated into body lipids (Fontanillas et al. 1997, Warnants et al. 1999). Thus, a clear dose-response relationship exists between dietary PUFA supply and tissue level, which results in lower MUFA deposition without affecting SFA content (Bee et al. 1999).

In this study, the fatty acid profile of backfat tissue of pregnant sows was not affected after a 90 d of supplementation with a linoleic acid–rich dietary fat (Table 3), except for a slight increase in the PUFA content, which was related predominantly to the high linoleic acid content of the diet. The reason for the lack of effect over time seen in the LA group can be attributed to the similar fatty acid profile of the gestation and lactation diets offered to all sows before their assignment to this experiment. Therefore, no change in the fatty acid profile of the backfat lipid was anticipated because the backfat tissue composition already reflected the diet; even with tissue turnover, it continued to reflect the diet throughout this study. On the contrary, dietary CLA supply in the gestation period

TABLE 3

Fatty acid composition of backfat tissue at d 7 and 97 of gestation in sows fed diets containing a linoleic acid (LA)- or conjugated linoleic acids (CLA)-enriched oil[1]

		Day of gestation			*P*-value[2]		
Item[3]	Treatment	7	97	SEM	T	D	T × D
		g/100 g total fatty acids					
14:0	LA	1.60	1.60	0.06			
	CLA	1.57	2.51	0.06	<0.01	<0.01	<0.01
16:0	LA	24.50	24.38	0.33			
	CLA	24.28	27.76	0.32	0.01	<0.01	<0.01
18:0	LA	13.35	13.24	0.76			
	CLA	14.06	16.41	0.69	0.11	0.09	0.07
20:0	LA	0.34	0.31	0.02			
	CLA	0.39	0.33	0.02	0.25	0.17	0.66
SFA	LA	40.12	39.95	0.91			
	CLA	40.60	47.62	0.85	0.02	0.01	0.01
16:1(n-7)	LA	2.53	2.51	0.12			
	CLA	2.45	2.22	0.11	0.29	0.72	0.15
18:1(n-9)	LA	40.02	38.82	1.03			
	CLA	40.29	32.02	0.95	0.06	<0.01	0.01
20:1(n-9)	LA	1.49	1.23	0.13			
	CLA	1.57	1.17	0.12	0.94	<0.01	0.43
MUFA	LA	44.29	42.81	1.16			
	CLA	44.58	35.63	1.08	0.07	<0.01	0.02
18:2(n-6)	LA	13.35	14.90	0.55			
	CLA	11.88	11.20	0.50	0.01	0.38	0.04
20:3(n-6)	LA	0.19	0.17	0.04			
	CLA	0.21	0.20	0.04	0.56	0.59	0.90
20:4(n-6)	LA	0.13	0.21	0.02			
	CLA	0.15	0.13	0.02	0.42	0.19	0.05
18:3(n-3)	LA	0.91	0.86	0.03			
	CLA	1.01	0.85	0.03	0.37	<0.01	0.07
CLA-isomers[4]							
*c*9,*t*11	LA	ND[5]	ND				
	CLA	ND	1.40	0.09		<0.01	
*c*9,*c*11	LA	ND	ND				
	CLA	ND	0.34	0.02		<0.01	
*t*10,*c*12	LA	ND	ND				
	CLA	ND	1.16	0.07		<0.01	
*c*10,*c*12	LA	ND	ND				
	CLA	ND	0.06	0.01		0.06	
*t*9,*t*11/*t*10,*t*12	LA	ND	ND				
	CLA	ND	0.63	0.04		<0.01	
PUFA	LA	15.59	17.24	0.55			
	CLA	14.84	16.76	0.51	0.44	0.01	0.81
16:1(n-7)/16:0	LA	0.10	0.10	0.01			
	CLA	0.10	0.05	0.01	0.10	0.07	0.08
18:1(n-9)/18:0	LA	3.01	2.98	0.16			
	CLA	2.97	2.01	0.15	0.06	0.03	0.03

[1] Results are presented as least-square means and SEM.
[2] Effects of treatment (T), gestation day (D), or that of interaction between treatment and gestation day (T × D).
[3] Fatty acids were designated by the number of carbon atoms followed by the number of double bonds. The position of the first double bond relative to the methyl (n) end of the molecule was also indicated. The sums of the main fatty acid series are represented as follows: SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.
[4] *c, cis*; *t, trans.*
[5] ND, not detected.

markedly increased myristic, palmitic and total SFA content of the backfat lipid, but decreased that of oleic acid and total MUFA (Table 3). The level of total SFA was likely determined by the rate of the de novo synthesis because the SFA content of the CLA and LA gestation diets was similar, differing by only 2 g/100 g total fatty acids.

Palmitoleic and oleic acid originate from uptake of dietary lipid and desaturation from a portion of the respective saturated fatty acids, which is regulated by the fatty acyl-CoA Δ^9-desaturase complex. The palmitoleic to palmitic acid [16:1(n-7)/16:0] ratio in the adipose tissue of the CLA group decreased from d 7 (0.10) to 97 (0.05) of gestation and was comparable to the dietary ratio (0.05), implying that the ratio was dictated primarily by the dietary lipids. On the contrary, in the LA group, the ratio did not change over the experimental period in the backfat (0.10) and was higher than in the diet (0.04), suggesting an increased desaturation activity. The oleic to stearic acid [18:1(n-9)/18:0] ratio in the adipose tissue of both treatments was lower than in the diet. In the LA group, the ratio was virtually identical at d 7 and 97 of gestation, but

TABLE 4

Effect of dietary supplementation of linoleic acid (LA) or conjugated linoleic acids (CLA) on fatty acid composition of sow colostrum and mature milk[1]

		Lactation week[2]					*P*-value[3]		
Item[4]	Treatment	1	2	3	4	SEM	T	W	T × W
		g/100 g total fatty acids							
14:0	LA	2.01	3.42	3.72	3.71	0.31			
	CLA	2.32	5.22	4.75	4.84	0.25	0.06	<0.01	0.04
16:0	LA	22.76	31.06	30.17	32.04	1.01			
	CLA	24.69	35.64	32.78	34.64	0.87	0.15	<0.01	0.70
18:0	LA	5.67	5.10	4.71	4.04	0.47			
	CLA	9.07	8.67	8.90	8.53	0.38	<0.01	0.03	0.46
SFA	LA	30.64	40.24	39.26	40.49	1.08			
	CLA	36.93	50.72	47.36	48.95	0.87	<0.01	<0.01	0.53
16:1(n-7)	LA	4.64	9.54	8.42	10.99	0.55			
	CLA	3.42	5.08	4.22	4.29	0.44	<0.01	<0.10	<0.01
18:1(n-9)	LA	34.80	30.31	29.97	28.69	1.36			
	CLA	32.20	25.70	29.45	27.70	1.09	0.30	<0.01	0.61
20:1(n-9)	LA	0.23	0.26	0.36	0.34	0.05			
	CLA	0.61	0.43	0.52	0.48	0.04	0.04	0.04	0.02
MUFA	LA	39.79	40.47	42.63	40.56	0.98			
	CLA	36.56	31.47	34.37	32.58	0.79	<0.01	0.20	0.28
18:2(n-6)	LA	25.84	16.75	15.86	16.86	0.82			
	CLA	17.57	11.23	12.08	11.96	0.66	<0.01	0.01	0.06
20:2(n-6)	LA	0.62	0.38	0.38	0.38	0.04			
	CLA	0.43	0.27	0.30	0.19	0.03	0.05	<0.01	0.26
20:4(n-6)	LA	1.11	0.68	0.50	0.39	0.04			
	CLA	0.62	0.39	0.36	0.36	0.03	<0.01	<0.01	<0.01
18:3(n-3)	LA	1.46	0.89	0.89	0.91	0.04			
	CLA	1.31	0.89	0.93	0.90	0.03	0.61	<0.01	0.29
total CLA[5]	LA	ND[6]	ND	ND	ND				
	CLA	6.03	4.91	4.42	5.04	0.19	<0.01	<0.01	<0.01
CLA-isomers[7,8]									
*c*9,*t*11	CLA	34.66	38.35	40.36	38.31	0.36		<0.01	
*c*9,*c*11	CLA	11.69	10.93	10.83	10.52	0.18		<0.01	
*t*10,*c*12	CLA	36.22	32.40	32.58	32.29	0.66		<0.01	
*c*10,*c*12	CLA	2.41	ND	ND	ND	0.33		<0.01	
*t*9,*t*11/*t*10,*t*12	CLA	15.00	16.98	16.20	18.72	0.37		0.02	
PUFA	LA	29.56	19.29	18.10	18.95	0.99			
	CLA	26.51	17.81	18.27	18.47	0.79	0.41	<0.01	0.43
16:1(n-7)/16:0	LA	0.21	0.31	0.28	0.35	0.02			
	CLA	0.14	0.14	0.13	0.12	0.02	<0.01	<0.01	<0.01
18:1(n-9)/18:0	LA	6.31	5.99	6.50	7.21	0.20			
	CLA	3.59	2.98	3.32	3.23	0.16	<0.01	0.04	0.09

[1] Results are presented as least-square means and SEM.
[2] Lactation wk 1 refers to sample of colostrum and lactation wk 2, 3 and 4 refer to mature milk collected on d 9, 16 and 23 after farrowing.
[3] Effects of treatment (T), lactation week (W), or that of interaction between treatment and lactation week (T × W).
[4] Fatty acids were designated by the number of carbon atoms followed by the number of double bonds. The position of the first double bond relative to the methyl (n) end of the molecule was also indicated. The sums of the main fatty acid series are represented as follows: SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.
[5] Sum of the *c*9,*t*11;*c*9,*c*11;*t*10,*c*12;*c*10,*c*12 and *t*9,*t*11/*t*10,*t*12 isomers.
[6] ND, not detected.
[7] *c, cis*; *t, trans.*
[8] Expressed as g/100 g total CLA.

decreased over time from 2.97 to 2.01 in the CLA group. These findings might indicate that the presence of CLA in the diet depressed synthesis of oleic acid in the adipose tissue lipids, possibly by decreasing stearoyl-CoA desaturase activity.

In the backfat tissue of the LA group, CLA isomers were not detected on either d 7 or 97 of gestation, confirming results of earlier studies with growing finishing pigs that tissue is devoid of CLA if not supplied by the diet (Kramer et al. 1998). Conversely, in the CLA group at d 97 of gestation, all measured isomers could be detected in the adipose tissue. To estimate the utilization of CLA isomers for lipid synthesis in the adipose tissue, we calculated a discrimination factor (CLA isomers to PUFA ratio in the adipose tissue as a percentage of the ratio in the diet) as proposed by Pettersen and Opstvedt (1991) for *trans* fatty acids. The discrimination factors were similar for the *c*9,*c*11 (0.42), *t*10,*c*12 (0.41) and *t*9,*t*11/*t*10,*t*12 (0.41) CLA isomers (**Table 5**). By contrast, the *c*9,*t*11 (0.52) isomer was incorporated into the adipose tissue at a higher level.

Sugano et al. (1997) reported that CLA was transported from the intestine as chylomicrons similarly to other fatty acids such as linoleic acid. However, lymphatic absorption was lower than that of linoleic acid and differed among individual CLA isomers. Additionally, the amount of the various isomers

TABLE 5

Ratio of the percentage of CLA isomers of the total polyenic fatty acid content of backfat, colostrum and mature milk lipids of pigs to that of the respective dietary fats[1]

	Backfat	Colostrum	Mature milk[2]
*c*9,*t*11[3]	0.52 ± 0.04	0.53 ± 0.03	0.69 ± 0.02
*c*9,*c*11	0.42 ± 0.03	0.63 ± 0.03	0.66 ± 0.03
*t*10,*c*12	0.41 ± 0.04	0.53 ± 0.02	0.55 ± 0.02
*c*10,*c*12[4]		0.55 ± 0.11	
*t*9,*t*11/*t*10,*t*12	0.41 ± 0.04	0.42 ± 0.01	0.56 ± 0.03

[1] Results are presented as means ± SEM.
[2] Average values of lactation wk 2, 3 and 4.
[3] *c, cis*; *t, trans.*
[4] Discrimination factors for backfat and mature milk are not calculated because the *c*10,*c*12 isomer was present only at trace levels.

in the chylomicrons as well as the distribution in the three stereospecific positions (*sn*-1, *sn*-2, *sn*-3) in the triacylglycerol molecule differed. In addition to uptake (and consequently availability) for lipid synthesis, other mechanisms such as rate of mobilization and reuptake could contribute to the explanation of their low proportion in adipose tissue triacyglycerols compared with the diet.

As reported in previous studies (Fritsche et al. 1993, Hartmann and Holmes 1989, Pettersen and Opstvedt 1991), regardless of diet, colostrum had a higher percentage of PUFA and lower level of SFA than mature milk. However, in contrast to these earlier studies, the MUFA level of the colostrum was not different from that of mature milk. The distinct difference between the treatments in the total SFA content of colostral and mature milk was due mainly to the markedly higher stearic acid content in the CLA compared with the LA group. The effect was independent of lactation stage.

When sows are not in a negative energy balance, the fatty acids in the milk are derived from the following two sources: *1*) the blood lipids, both endogenous and dietary fatty acids, and *2*) de novo synthesis in the mammary glands (Boyd and Kensinger 1998). As reviewed by Hartmann and Holmes (1989), most of the fatty acids in the colostral and mature milk lipids are found to reflect closely those of blood triacylglycerols, which in turn are influenced by the type of fat ingested by the sow. The fatty acid profile of the milk lipids suggested an increased synthesis of stearic acid by elongation from palmitic acid and/or by de novo synthesis in the mammary gland of the CLA-fed sows because dietary fat intake, and consequently intake of the respective fatty acids, in the lactation period was similar for both groups. In addition, the palmitoleic to palmitic acid and oleic to stearic acid ratios were lower in the CLA group compared with the LA group and corroborate the effects found in adipose tissue (Table 3).

In the lactation diet, no detectable levels of arachidonic acid were present; however, the amount excreted by the mammary gland was significantly different between treatments. Arachidonic acid derives primarily from the diet or from synthesis from linoleic acid via γ-linolenic acid (18:3, n-6) and dihomo-γ-linolenic acid 20:3(n-6) (Enser 1984) by one elongation and two specific desaturation steps (Δ^6-and Δ^5-desaturase). The question arises whether the lower arachidonic acid level in the milk of the CLA group was due to the lower dietary supply of the substrate or the result of inhibition of one or both of the specific desaturases. Belury and Kempa-Steczko (1997) suggested that CLA may compete with linoleic acid for the Δ^6-desaturase because radiolabeled CLA was desaturated to a similar extent as radiolabeled linoleic acid. In support of these findings, they reported reduced arachidonic acid in the liver, which might have been the effect of lower conversion of linoleic to arachidonic acid. Interestingly, in their in vitro assay, CLA was converted to an unidentified 18:3 product, which might not be a suitable substrate for elongation or desaturation by Δ^5-desaturase.

Sow's milk is naturally devoid of CLA, if not supplied by the diet (Jahreis et al. 1999). The accumulation of the different CLA isomers identified, reflected that of the diet. However, contrary to the fat biopsy, the *c*10,*c*12 isomer could be detected only in the colostrum and not in the mature milk. The estimated transfer or incorporation efficiency of the different isomers from the diet to the lipids excreted by the mammary gland was lower in the colostrum than in the mature milk (Table 5). Compared with the dietary supply via the lactation diet and in agreement with the results found for adipose tissue, relatively more *c*9,*t*11 was excreted than *c*9,*c*11; *t*9,*t*11/*t*10,*t*12 and *t*10,*c*12 in the mature milk. In the colostral milk, the transfer seemed to be higher for the *c*9,*c*11 followed by the *c*10,*c*12; *c*9,*t*11; *t*10,*c*12 and *t*9,*t*11/*t*10,*t*12 CLA isomers. This finding is in contrast to the previously estimated incorporation (adipose tissue) and transfer efficiency rate (mature milk) and might be the effect of the transition state of the mammary gland at the beginning of the lactation period. However, the overall transfer efficiency seemed to be higher in sows compared with cows in which the transfer rate was estimated to reach 22–26% for most of the infused isomers (Chouinard et al. 1999).

In conclusion, the data show that feeding sows a CLA-enriched diet during gestation and lactation can increase the concentration of CLA and markedly affect the fatty acid composition of backfat tissue, colostrum and mature milk lipids. The effect on the lipid composition was similar for adipose tissue and milk. Compared with the LA group, significantly more SFA and less MUFA were deposited in the tissue and excreted in the milk, whereas changes in the PUFA content were minimal, except for a significant decrease in the arachidonic acid level. Estimated transfer efficiency for adipose tissue and mature milk ranged from 41 to 52% and 55 to 69%, respectively, and seemed to be higher than that measured in cows.

ACKNOWLEDGMENTS

The author is grateful for the donation of the CLA and linoleic-enriched oil by Grünau Jllertissen GmbH, Germany. I also would like to thank Pierre-Alain Dufey, Silviane Gentil, Martin Jost, Peter Stoll, Fritz Wüthrich and Bruno Wirz for their excellent technical assistance.

LITERATURE CITED

Alessandri, J. M., Goustard, B., Guesnet, P. & Durand, A. (1998) Docosahexaenoic acid concentrations in retinal phospholipids of piglets fed an infant formula enriched with long-chain polyunsaturated fatty acids: effects of egg phospholipids and fish oils with different ratios of eicosapentaenoic acid to docosahexaenoic acid. Am. J. Clin. Nutr. 67: 377–385.

Bee, G., Messikommer, R. & Gebert, S. (1999) Dietary fats and energy levels differently affect tissue lipogenic enzyme activity in finishing pigs. Fett Lipid 101: 336–342.

Belury, M. A. & Kempa-Steczko, A. (1997) Conjugated linoleic acid modulates hepatic lipid composition in mice. Lipids 32: 199–204.

Boltshauser, M., Jost, M., Kessler, J. & Stoll, P. (1993) Fütterungsempfehlungen und Nährwerttabellen für Schweine. Landwirtschaftliche Lehrmittelzentrale, Switzerland.

Boyd, R. D. & Kensinger, R. S. (1998) Metabolic precursors for milk synthesis. In: The Lactating Sow (Verstegen, M.W.A., Moughan, P. J. & Schrama, J. W., eds.), pp. 71–95. Wageningen Pers, Wageningen, The Netherlands.

Camara, M., Mourot, J. & Fevrier, C. (1996) Influence of two dairy fats on lipid

synthesis in the pig: comparative study of liver, muscle and the two backfat layers. Ann. Nutr. Metab. 40: 287–295.
Chin, S. F., Storkson, J. M., Albright, K. J., Cook, M. E. & Pariza, M. W. (1994) Conjugated linoleic acid is a growth factor for rats as shown by enhanced weight gain and improved feed efficiency. J. Nutr. 124: 2344–2349.
Chouinard, P. Y., Corneau, L., Barbano, D. M., Metzger, L. E. & Bauman, D. E. (1999) Conjugated linoleic acids alter milk fatty acid composition and inhibit milk fat secretion in dairy cows. J. Nutr. 129: 1579–1584.
Darragh, A. J. & Moughan, P. J. (1998) The composition of colostrum and milk. In: The Lactating Sow (Verstegen, M.W.A., Moughan, P. J. & Schrama, J. W., eds.), pp. 3–21. Wageningen Pers, Wageningen, The Netherlands.
Dugan, M.E.R., Aalhus, J. L., Jeremiah, L. E., Kramer, J.K.G. & Schaefer, A. L. (1999) The effects of feeding conjugated linoleic acid on subsequent pork quality. Can. J. Anim. Sci. 79: 45–51.
Dugan, M.E.R., Aalhus, J. L., Schaefer, A. L. & Kramer, J.K.G. (1997) The effect of conjugated linoleic acid on fat to lean repartitioning and feed conversion in pigs. Can. J. Anim. Sci. 77: 723–725.
Enser, M. (1984) The chemistry, biochemistry and nutritional importance of animal fats. In: Fats in Animal Nutrition (Wiseman, J., ed.), pp. 23–51. Butterworths, London, UK.
Fontanillas, R., Barroeta, A., Baucells, M. D. & Codony, R. (1997) Effect of feeding highly *cis*-monounsaturated, *trans*, or n-3 fats on lipid composition of muscle and adipose tissue of pigs. J. Agric. Food Chem. 45: 3070–3075.
Fritsche, K. L., Huang, S. C. & Cassity, N. A. (1993) Enrichment of omega-3 fatty acids in suckling pigs by maternal dietary fish oil supplementation. J. Anim. Sci. 71: 1841–1847.
Goustard, L. B., Guesnet, P., Durand, G., Antoine, J. M. & Alessandri, J. M. (1999) n-3 and n-6 fatty acid enrichment by dietary fish oil and phospholipid sources in brain cortical areas and nonneural tissues of formula-fed piglets. Lipids 34: 5–16.
Hartmann, P. E. & Holmes, M. A. (1989) Sow lactation. In: Manipulating Pig Production II (Barnett, J. L. & Hennessy, D. P., eds.), pp. 72–97. Australasian Pig Science Association, Albury, NSW, Australia.
Ip, C., Scimeca, J. A. & Thompson, H. J. (1994) Conjugated linoleic acid. A powerful anticarcinogen from animal fat sources. Cancer 74: 1050–1054.
Jahreis, G., Fritsche, J., Mockel, P., Schone, F., Moller, U. & Steinhart, H. (1999) The potential anticarcinogenic conjugated linoleic acid, *cis*-9,*trans*-11 C18: 2, in milk of different species: cow, goat, ewe, sow, mare, woman. Nutr. Res. 19: 1541–1549.
Kramer, J.K.G., Sehat, N., Dugan, M.E.R., Mossoba, M. M., Yurawecz, M. P., Roach, J. A., Eulitz, K., Aalhus, J. L., Schaefer, A. L. & Ku, Y. (1998) Distributions of conjugated linoleic acid (CLA) isomers in tissue lipid classes of pigs fed a commercial CLA mixture determined by gas chromatography and silver-ion high-performance liquid chromatography. Lipids 33: 549–558.
Lee, K. N., Kritchevsky, D. & Pariza, M. W. (1994) Conjugated linoleic acid and atherosclerosis in rabbits. Atherosclerosis 108: 19–25.
Littell, R. C., Henry, P. R. & Ammerman, C. B. (1998) Statistical analysis of repeated measures data using SAS procedures. J. Anim. Sci. 76: 1216–1231.
Loor, J. J. & Herbein, J. H. (1998) Exogenous conjugated linoleic acid isomers reduce bovine milk fat concentration and yield by inhibiting de novo fatty acid synthesis. J. Nutr. 128: 2411–2419.
Metcalfe, L. D. & Smith, J. F. (1961) The rapid preparation of fatty acid esters for gas chromatographic analyses. Anal. Chem. 33: 363–364.
Miller, C. C., Park, Y., Pariza, M. W. & Cook, M. E. (1994) Feeding conjugated linoleic acid to animals partially overcomes catabolic responses due to endotoxin injection. Biochem. Biophys. Res. Commun. 198: 1107–1112.
Morgan, C. A., Noble, R. C., Cocchi, M. & McCartney, R. (1992) Manipulation of the fatty acid composition of pig meat lipids by dietary means. J. Sci. Food Agric. 58: 357–368.
Mourot, J., Peiniau, P. & Mounier, A. (1994) Effets de l'acide linoleique alimentaire sur l'activité des enzymes de la lipogenese dans les tissus adipeux chez le porc [Effects of dietary linoleic acid on lipogenesis enzyme activity in adipose tissue in the pig]. Reprod. Nutr. Dev. 34: 213–220.
Ostrowska, E., Muralitharam, M., Cross, R. F., Bauman, D. E. & Dunshea, F. R. (1999) Dietary conjugated linoleic acids increase lean tissue and decrease fat deposition in growing pigs. J. Nutr. 129: 2037–2042.
Pettersen, J. & Opstvedt, J. (1991) *Trans* fatty acids. 4. Effects on fatty acid composition of colostrum and milk. Lipids 26: 711–717.
Purvis, J. M., Clandinin, M. T. & Hacker, R. R. (1982) Fatty acid accretion during perinatal brain growth in the pig. A model for fatty acid accretion in human brain. Comp. Biochem. Physiol. 72: 195–199.
SAS Institute Inc. (1998) SAS User's Guide: Statistics, version 6.12. SAS Institute, Cary, NC.
Scimeca, J. A., Thompson, H. J. & Ip, C. (1994) Effect of conjugated linoleic acid on carcinogenesis. Adv. Exp. Med. Biol. 364: 59–65.
Sugano, M., Tsujita, A., Yamasaki, M., Yamada, K., Ikeda, I. & Kritchevsky, D. (1997) Lymphatic recovery, tissue distribution, and metabolic effects of conjugated linoleic acid in rats. J. Nutr. Biochem. 8: 38–43.
Warnants, N., Van Oeckel, M. J. & Boucque, C. V. (1999) Incorporation of dietary polyunsaturated fatty acids into pork fatty tissues. J. Anim. Sci. 77: 2478–2490.
West, D. B., Delany, J. P., Camet, P. M., Blohm, F., Truett, A. A. & Scimeca, J. (1998) Effects of conjugated linoleic acid on body fat and energy metabolism in the mouse. Am. J. Physiol. 44: R667–R672.
Winter, E. (1963) Über ein neues Verfahren zur Bestimmung und Untersuchung von Fetten in Lebensmitteln. Z. Lebensm.-Unters.-Forsch. 123: 205–210.