

# The effects of different amounts and types of fat on the level of conjugated linoleic acid in the meat and milk of sheep\*

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## ABSTRACT

The influence of the addition of rape seed oil, linseed oil and hydrogenated rape seed oil to the diet for milking ewes and growing lambs on the concentration of *cis*-9, *trans*-11 octadecadienoic acid, a conjugated linoleic acid, in milk and meat was investigated. In Experiment 1, 24 fattened lambs of 22±2 kg body weight were divided into four groups and fed control (without added fat) and experimental diets with the respective additions to the basic diet of 6% rape seed oil, linseed oil or hydrogenated rape seed oil. In experiment 2 four lactating ewes of 50±5 kg body weight were fed a basic diet for milking sheep consisting of 60% concentrate and 40% meadow hay (control group), which was supplied with 4, 8 or 10% of fat in dry matter for the experimental groups. Supplementing diets for fattened lambs with 6% rape seed oil, linseed oil or hydrogenated rape seed oil had no effect on the CLA content in meat. Rape seed oil and hydrogenated rapeseed oil had no effect on the CLA content in milk, whereas the addition of linseed oil significantly increased ( $P<0.05$ ) the CLA level in milk.

KEY WORDS: sheep, fat, conjugated linoleic acid, CLA

## INTRODUCTION

Conjugated linoleic acids (CLA) found in ruminant milk and meat are a by-product of incomplete biohydrogenation of lipids by ruminal bacteria (Chouinard

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et al., 2001). CLA refers to a mixture of positional and geometric isomers of octadecadienoic (linoleic) acid with conjugated double bonds (Ip et al., 1994). The most extensively investigated CLA isomer is *cis*-9, *trans*-11 octadecadienoic acid, which is thought to be the biologically active form. Conjugated linoleic acid is unusual among anti-carcinogenic compounds in that it acts to both reduce the incidence of tumors in laboratory animal models and is a cytotoxic agent to existing tumor cells. Meat from ruminants generally contains more CLA than meat from nonruminants (Ip, 1994; Bessa et al., 2000). In addition to meat, milk and other dairy products are good sources of CLA (Ip, 1994; Parodi, 1994). Because fat is commonly used to supply energy in the diets of high producing ruminants, it is necessary to determine which fat sources yield the most CLA in the rumen as well as in milk and meat fat. Because there is no evidence for conversion of 9-*cis*, 12-*cis*-octadecadienate to CLA (*via* an isomerase) in humans (Adlof et al., 2000), research needs to focus on maximizing CLA formation in animal products (i.e., from ruminants) that are consumed by humans. Therefore, the objective of this study was to determine the effect of three fats on CLA formation in sheep milk and meat.

## MATERIAL AND METHODS

### *Experiment 1*

Twenty-four fattened lambs of 22±2 kg body weight were divided into four groups and fed *ad libitum* a diet consisting of concentrate and meadow hay without added fat for the control group and with the addition of 6 % of rape seed oil, hydrogenated rape seed oil or linseed oil in dry matter to the concentrate for the experimental groups. Lambs were fattened until 40 kg. The composition of diets is shown in Table 1. Drinking water was available *ad libitum*. Consequently, special emphasis was placed on the effect of dietary fat on conjugated linoleic acid levels in the muscles of lambs.

TABLE 1

Composition of the concentrate fed to lambs (Experiment 1), %

Components	Level of fat rapeseed oil, linseed oil or hydrogenated rapeseed oil	
	0%	6%
Wheat meal	60	60
Rapeseed meal	18	18
Wheat bran	20	14
Minerals	2	2
Fat	0	6

## Experiment 2

Four lactating ewes of  $50 \pm 5$  kg body weight were assigned in a  $4 \times 4$  Latin square design to four diets differing in supplemental fat source. Each experimental period was 16 d and consisted of 12 days of adjustment followed by 4 days of sample collection. Sheep had free access to water and were fed twice daily following each milking. The daily basic ration (control group) consisted of forage and concentrate (60:40) and was supplemented for experimental groups with 0, 4, 8 and 10% in dry matter of rape seed oil, linseed oil or hydrogenated rape seed oil. The concentrate consisted of wheat meal, soyabean meal, wheat bran and minerals (Table 2). The energy value of the rations was 1.21 JPM, whereas the crude protein content was 120 g BTJN and 129 g BTJE. Sheep were milked twice daily at approximately 08.00 and 17.00, and yields were recorded.

TABLE 2

Composition of the diet for ewes (Experiment 2), %

Supplemented fat, %	0	4	8	10
Meadow hay	59.0	57.0	55.0	54.0
Wheat meal	21.0	20.0	19.0	19.0
Rapeseed meal	3.0	3.0	3.0	3.0
Wheat bran	15.0	14.0	13.0	12.0
Minerals	2.0	2.0	2.0	2.0
Fat	0.0	4.0	8.0	10.0

*Analysis*

The level of conjugated linoleic acid in muscles (*M. longissimus dorsi*) of lamb and milk of ewes was determined according to the procedures of Heinig et al. (1998), modified by Czauderna et al. (2001). All data were analyzed using SAS procedures (User's Guide, 1990).

## RESULTS

Feeding fat of vegetable origin to lambs had no significant influence on meat fat CLA content (Table 3). CLA content in milk increased ( $P < 0.05$ ) from 0.42  $\mu\text{g/g}$  in the control group to 0.61, 1.47 and 2.72  $\mu\text{g/g}$ , respectively, in groups with 4, 8 and 10% added linseed oil (Table 4). The addition of rape seed oil and hydrogenated rapeseed oil had no influence on CLA content.

TABLE 3

Conjugated linoleic acid level in meat,  $\mu\text{g/g}$ 

Meat	Control		Rape seed oil		Linseed oil		Hydrogenated rape seed oil	
	mean	CV	mean	CV	mean	CV	mean	CV
CLA	0.02	34.40	0.03	30.98	0.03	29.96	0.03	209.08

TABLE 4

Conjugated linoleic acid level in milk,  $\mu\text{g/g}$  of lyophilized samples

Supplemented fat, %	0		4		8		9		10	
	mean	CV	mean	CV	mean	CV	mean	CV	mean	CV
Rape seed oil	1.26	39.02	1.21	59.97	1.32	64.00	1.34	58.71	1.34	58.71
Linseed oil	0.42 <sup>b</sup>	40.40	0.61 <sup>a</sup>	58.63	1.47	103.06	2.72 <sup>ab</sup>	89.94	2.72 <sup>ab</sup>	89.94
Hydrogenated rape seed oil	1.58	58.82	1.28	64.06	1.53	48.98	1.15	89.39	1.15	89.39

means in rows with the same letter differ statistically significant <sup>a,b,c</sup> –  $P < 0.05$ 

## DISCUSSION

Supplementation of diets for ruminants with fat changes the nutrient supply and composition of milk and meat (Romo et al., 2000). Literature data suggest that it may be possible to reduce the incidence of some cancers in humans by increasing the amount of CLA consumed in the diet. Potentially beneficial effects on body composition and on immune function have also been reported. Less consistent have been reports of putative hypocholesterolemic actions of CLA (Williams, 2000). Supplementing lamb diets with a source of linoleic acid seems to be a successful method of increasing the CLA content of tissues (Mir et al., 2000). In our experiment, adding fat did not significantly influence the CLA level in meat, though we observed a tendency for the CLA level to increase (by 50%) but this was not significant ( $P > 0.05$ ) in lamb meat when the fat of rape seed oil, linseed oil or hydrogenated rape seed oil was added to the diets (Table 3). Because CLA is formed as an intermediate in the biohydrogenation of unsaturated fatty acids, hydrogenated rape seed oil, the source of most saturated fatty acids, had no influence on CLA content in either meat or in milk. Also rape seed oil added to sheep diets had no influence on CLA level in milk, whereas Mir et al. (1999) noted that feeding 4% rape seed oil to dairy goats greatly increased milk fat CLA content (by 204%). Experiments carried out by Desilets et al. (2000) indicate that regardless of the

form of added canola (calcium salts of fatty acids from canola oil, free canola oil or whole ground canola seed) supplementing diets with canola increased milk CLA contents. Feeding linseed oil (a C18:3-rich oil) greatly increases milk fat CLA content (Chilliard et al., 2000) and is as at least as efficient as C18:2-rich vegetable oils. The obtained results indicate a significant correlation between adding linseed oil and the CLA level in milk. In our experiment, the addition of linseed oil also significantly ( $P < 0.05$ ) affected the CLA concentration.

An increased level of conjugated linoleic acid can be achieved by feeding optimal fat in a ruminant's ration. Further experiments are needed to determine the proper fatty acid composition of fat added to ruminant diets.

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## STRESZCZENIE

### **Wpływ rodzaju i ilości dodanego do diety tłuszczu na poziom sprzężonego kwasu linolowego w mięsie i mleku owcy**

Sprzężony kwas linolowy (CLA) został ostatnio uznany jako potencjalny czynnik antykancerogenny, a jego jedynym naturalnym źródłem są produkty pochodzenia zwierzęcego. Celem przeprowadzonych doświadczeń było określenie wpływu dodatku oleju rzepakowego, oleju lnianego oraz uwodornionego oleju rzepakowego do dawki dla owiec na poziom sprzężonego kwasu linolowego w uzyskiwanym od nich mleku i mięsie. W pierwszym doświadczeniu 24 jagniąt o masie ciała  $22 \pm 2$  kg podzielono na 4 grupy i skarmiano dawkę kontrolną bez dodatku tłuszczu roślinnego, bądź z dodatkiem 6% oleju rzepakowego, oleju lnianego lub uwodorowanego oleju rzepakowego. W drugim doświadczeniu materiał doświadczalny stanowiły cztery owce mleczne o średniej masie ciała  $50 \pm 5$  kg. Doświadczenie przeprowadzono w układzie kwadratu łacińskiego,  $4 \times 4$ . Zwierzęta żywiono dawkami z dodatkiem 0, 4, 8 i 10% oleju rzepakowego, oleju lnianego lub uwodornionego oleju rzepakowego w suchej masie. Dodatek oleju rzepakowego oraz uwodornionego oleju rzepakowego nie miał wpływu na poziom CLA w mleku i w mięsie, natomiast olej lniany dodany do dawki dla owiec podniósł statystycznie istotnie ( $P < 0,05$ ) poziom CLA w mleku.