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Michel M. Doreau, Cécile Martin. Effects of linseed lipids fed as rolled seeds, extruded seeds or oil on organic matter and crude protein digestion in cows. *Animal Feed Science and Technology*, 2009, 150 (3-4), pp.187-196. 10.1016/j.anifeedsci.2008.09.004 . hal-02658550

**HAL Id: hal-02658550**

**<https://hal.inrae.fr/hal-02658550>**

Submitted on 30 May 2020

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Contents lists available at ScienceDirect

# Animal Feed Science and Technology

journal homepage: [www.elsevier.com/locate/anifeedsci](http://www.elsevier.com/locate/anifeedsci)

## Effects of linseed lipids fed as rolled seeds, extruded seeds or oil on organic matter and crude protein digestion in cows

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### ARTICLE INFO

#### Article history:

Received 11 January 2008

Received in revised form 16 September 2008

Accepted 19 September 2008

#### Keywords:

Linseed oil

Linseed

Extrusion

Ruminal digestion

Cow

### ABSTRACT

Effects of fatty acids of linseed in different forms, on ruminal fermentation and digestibility were studied in dry cows fitted with ruminal and duodenal cannulas. Four diets based on maize silage, lucerne hay and concentrates (65/10/25 dry matter (DM)) were compared in a  $4 \times 4$  Latin square design experiment where the diets were: control diet (C), diet RL supplied 75 g/kg DM rolled linseeds, diet EL supplied 75 g/kg DM extruded linseeds, and diet LO supplied 26 g/kg DM linseed oil and 49 g/kg DM linseed meal. The diets did not differ in total organic matter (OM) and fibre digestibility, in forestomach and intestinal OM digestibility, and in duodenal N flow. Microbial N duodenal flow tended to be lower for RL versus C diet ( $P < 0.1$ ). Extrusion did not reduce ruminal crude protein (CP) degradation *in vivo* and *in situ*. Volatile fatty acid concentration and pattern, and protozoa concentration in the rumen, did not vary among diets. Results confirm the absence of a negative effect of a moderate supply of linseed on rumen function, as well as no effect of extrusion on its ruminal CP degradability.

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### 1. Introduction

Linseed is an important source of  $\alpha$ -linolenic acid. Diets supplemented with fatty acids (FA) from linseed lead to higher milk concentrations of  $\alpha$ -linolenic acid and the *cis*9, *trans*11 isomer of conjugated

**Abbreviations:** ADF, acid detergent fibre; C, control diet; CLA, conjugated linoleic acid; DM, dry matter; EL, diet C + extruded linseeds; FA, fatty acid; LO, diet C + linseed oil + linseed meal; NAN, non-ammonia N; NDF, neutral detergent fibre; OM, organic matter; PDI, protéines digestibles dans l'intestin; PPB, purine and pyrimidine bases; RL, diet C + rolled linseeds; TD, theoretical degradability; UFL, unité fourragère lait; VFA, volatile fatty acids.

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linoleic acid (CLA), that are beneficial to human health (Loor et al., 2005). Hence the use of linseeds in dairy cow diets is increasing.

However, a major concern is potential negative effects of FA from linseed on ruminal digestion. Several experiments in the 1980s showed strong negative effects of 50–70 g/kg diet dry matter (DM) linseed oil on ruminal digestion in sheep (Ikwuegbu and Sutton, 1982; Sutton et al., 1983). This effect was ascribed partly to a large drop in the protozoa population and partly to a shift of volatile fatty acid (VFA) composition towards propionate. In contrast, a recent experiment in dairy cows has shown that a supply of 30 g/kg DM linseed oil did not decrease whole tract organic matter (OM) or fibre digestibility, and moderately decreased forestomach OM digestibility (Ueda et al., 2003). Other experiments with sheep or cattle fed linseeds did not show decreases in OM total tract digestibility (Wachira et al., 2000; Petit et al., 2002). These results show that effects of linseed oil may depend on level of inclusion and/or of level of feeding, which is higher in cows than in sheep and suggest that linseed oil may have a more depressing effect on digestibility than linseed. This second hypothesis is supported by the general assessment that vegetable oil has a more depressing effect on digestion when fed in the triglyceride form than when it is in the seed (Jouany et al., 2000), possibly due to a more rapid release of oil from the cell in the rumen. Extrusion, which is often used to improve the nutritive value of protein in seeds, may give intermediate results since it disrupts most cells leading to increased oil release compared to whole or rolled seeds.

The present experiment aimed to evaluate effects on forestomach and intestinal OM digestion and protein metabolism of three forms of linseed, being free oil, rolled seeds, and extruded seeds, in comparison with a control diet.

## 2. Materials and methods

### 2.1. Animals, design, and treatments

Four multiparous dry Holstein cows were fitted with rumen cannulae (polyvinyl chloride and polyamide, internal diameter 120 mm) and duodenal cannulae (plastisol, T-shaped with a gutter-type base). Surgery had been completed aseptically at least 1 year before the experiment under general anaesthetics using halothane (ICI Pharma, Paris, France). Surgical procedures, post-surgical care and animal management during the experiment were in accordance with French national legislation on care and use of laboratory animals, and with international recommendations (Canadian Council on Animal Care, 1993). The body weight of the cows at the beginning of the experiment was  $644 \pm 40$  kg and the experimental design was a  $4 \times 4$  Latin square with 5-week experimental periods. Measurements occurred during the last 10 days of each period, noted below as d1–d10. Treatments were: (1) control diet (C); (2) diet C supplied with 75 g/kg DM rolled linseeds (RL); (3) diet C supplied with 75 g/kg DM extruded linseeds (EL); (4) diet C supplied with 26 g/kg DM linseed oil and 49 g/kg DM linseed meal (LO). The ingredient and chemical composition of the experimental diets are in Table 1. Diets were given in limited amounts (i.e., 0.90 of individual voluntary intake determined in a pre-experimental period) to avoid refusals and differences in intake among diets. Diet composition was calculated to meet the net energy and digestible crude protein (CP) requirements of cows according to the French Unité Fourragère Lait (UFL) and Protéines Digestibles dans l'Intestin (PDI) systems (INRA, 1989). All diets were comprised of (g/kg DM) 600 maize silage, 100 lucerne hay, 140 dairy concentrate, 150 experimental concentrates, 5 urea and 5 mineral–vitamin premix (Galaphos Midi Duo GR, CCPA, Aurillac, France). Extruded linseeds were supplied as Croquelin® (Valorex, Combourtillé, France) which is an extruded mixture of (g/kg DM) 500 linseeds, 300 wheat bran and 200 sunflower meal. Extrusion used a one-screw extruder at 120°C after a low-temperature conditioning with steam. In the RL diet, the linseeds of the same batch as that used to make Croquelin® was rolled (Model Futura 5.5 CV, S.A. Toys, Montoire, France). Particle size, determined by dry sieving, was  $0.226 > 2.0$  mm; 0.620 between 0.8 and 2.0 mm, 0.094 between 0.4 and 0.8 mm, 0.043 between 0.2 and 0.4 mm, and  $0.017 < 0.2$  mm. In the LO diet, proportions of linseed oil and meal were calculated to provide the same mean FA content in the mixture as rolled and extruded linseeds. In order to balance the ingredient composition, wheat bran and sunflower meal were supplied in appropriate proportions in the RL and LO diets. The control diet included the same experimental concentrates as in the LO diet except that linseed oil was replaced

**Table 1**  
Ingredient and chemical composition of the experimental diets<sup>a</sup>.

	Diet <sup>b</sup>				
	C	RL	EL	LO	S.E.M.
Ingredient composition (g/kg DM)					
Maize silage	600	600	600	600	
Alfalfa hay	100	100	100	100	
Concentrate mixture <sup>c</sup>	140	140	140	140	
Rolled linseeds	–	75	–	–	
Extruded mixture of linseed, wheat bran and sunflower meal <sup>d</sup>	–	–	150	–	–
Linseed oil	–	–	–	26	
Linseed meal, expeller	45	–	–	49	
Wheat bran	45	45	–	45	
Sunflower meal, solvent	30	30	–	30	
Ground maize grain	30	–	–	–	
Urea	5	5	5	5	
Mineral-vitamin mixture <sup>e</sup>	5	5	5	5	
Chemical composition (g/kg DM)					
OM	947.2 <sup>a</sup>	942.6 <sup>b</sup>	943.0 <sup>b</sup>	943.1 <sup>b</sup>	0.03
Crude protein	147.9 <sup>a</sup>	143.5 <sup>b</sup>	144.1 <sup>b</sup>	145.7 <sup>c</sup>	0.02
NDF	333.8 <sup>a</sup>	355.0 <sup>c</sup>	338.7 <sup>b</sup>	331.1 <sup>a</sup>	0.09
ADF	188.8 <sup>a</sup>	203.3 <sup>c</sup>	192.4 <sup>b</sup>	187.6 <sup>a</sup>	0.05
Fatty acids	26.4 <sup>a</sup>	49.9 <sup>b</sup>	50.1 <sup>c</sup>	50.1 <sup>c</sup>	0.01

<sup>a</sup> Means with different superscripts differ ( $P < 0.01$ ).

<sup>b</sup> C = control; RL = rolled linseeds; EL = extruded linseeds; LO = linseed oil + linseed meal.

<sup>c</sup> Composition (g/kg): wheat grain 200, barley grain 200, dehydrated beet pulp 300, rapeseed meal (solvent) 150, soybean meal (solvent) 87, beet molasses (liquid) 20, limestone 8, dicalcium phosphate 1, magnesium oxide 5, sodium chloride 5, binding agent 1, vitamin and trace element mixture 5.

<sup>d</sup> Croquelin® (Valorex, Combourtillé, France).

<sup>e</sup> Composition (g/kg): Ca, 200; P, 45; Mg, 45; Na, 50; Cu, 1.3; Zn, 6.0; Mn, 3.5; I, 0.08; Co, 0.032; Se, 0.020; vitamin A, 600,000 IU; vitamin D3, 120,000 IU; vitamin E, 1,300 IU.

by maize grain. It also contained linseed meal in order to ensure the same protein composition as for the other diets.

Maize silage was offered in equal portions at 09:30 and 16:30 h. Hay and concentrates, including oil, were offered at 09:00 h.

## 2.2. Measurements and analyses

The DM content of the feeds was measured at 103°C for 24 h every day for maize silage and twice a week for the other feeds. Total tract apparent digestibility was measured by total faecal collection for 6 days (d1–d6) in collection bins placed on the floor. A 0.01 representative sample of faeces was collected every day, and its DM content was measured at 103°C for 24 h. Another aliquot of 0.005 of faeces was sampled daily, stored at –20°C, and pooled among days before freeze-drying. Urine was separated from faeces using a device maintained around the vulva, and collected for 6 days (d1–d6) in a 20-l bottle containing 500 ml of 0.10 sulphuric acid. A 0.005 aliquot was collected daily and preserved at 4°C before being pooled among days for N analysis. Samples of each feed were collected twice weekly, pooled and stored at ambient temperature. Samples of feeds (i.e., hay, soybean meal, grain mixture pellet) and freeze-dried faeces were ground and analysed for OM, N, neutral detergent fibre (NDF), acid detergent fibre (ADF) and total FA content. Organic matter was determined by mass difference before ashing at 550°C for 6 h. The NDF was analysed using heat-stable amylase but not sodium sulphite and expressed exclusive of residual ash (aNDFom) and the ADF was analysed exclusive of residual ash (ADFom) (Van Soest et al., 1991). N was analysed in feeds, faeces and urine using a Kjeldahl method (AOAC, 1990, ID 954.01). Total FA contents of feedstuffs were determined from methyl esters by gas–liquid chromatography (model CP-3800, Varian, Les Ulis, France). Fatty acid methyl esters were prepared according to the method of Sukhija and Palmquist (1988).

Duodenal flow was measured via the double marker method (Faichney, 1980) using  $\text{YbCl}_3$  and cobalt–ethylene diamine tetra-acetic acid (Co–EDTA) continuously infused in aqueous solution for 8 days. A solution containing the markers (Yb: 1 g/L, Co–EDTA: 2 g/L) was infused into the rumen at a rate of 100 ml/h through individual tubes. After 4 days for ruminal marker concentration equilibration, sixteen 250 ml samples were collected every 1.5 h for 48 h (d4–d6). Immediately after each sampling, liquid and solid phases were obtained from a 100 ml duodenal sample by squeezing through a 250  $\mu\text{m}$  mesh-size nylon cloth. Duodenal samples were preserved at  $-20^\circ\text{C}$  until analysis. A fraction of fresh duodenal samples (i.e., total, liquid phase, solid phase) was analysed for DM, OM and N using the described methods. Ammonia N was determined using an automated colorimetric technique (Van Eenaeme et al., 1969) and Co was analysed by atomic absorption spectrophotometry (Analyst 400 spectrophotometer, PerkinElmer, Bois d'Arcy, France) at 240.7 nm with a air/acetylene flame. The remaining fraction of duodenal samples was freeze-dried and ground, and then Yb was assayed by atomic absorption spectrophotometry at 398.8 nm with an acetylene/ $\text{N}_2\text{O}$  mixture flame, while purine and pyrimidine bases (PPB) were assayed by HPLC (Alliance, Waters, Milford, MA) according to Lassalas et al. (1993). The sum of PPB was used as the reference for nucleic acid content. Three representative 1 kg samples of ruminal content were collected by hand at 09:00, 11:30, and 14:00 h on d8. Samples were immediately squeezed by hand through a 250  $\mu\text{m}$  mesh-size nylon cloth and the liquid filtrate was quickly moved to the laboratory to obtain bacterial samples by extraction from the liquid filtrate by successive centrifugations at  $1000 \times g$  for 15 min and at  $27,000 \times g$  for 30 min at  $4^\circ\text{C}$ . Bacterial samples were freeze-dried, ground and analysed for N and PPB. Duodenal flows and the reconstitution factor were calculated from Yb and Co–EDTA concentrations in total duodenal content, liquid phase and solid phase (Faichney, 1980). Effective duodenal flows of OM, N, and ammonia N were calculated using the reconstitution factor. Duodenal bacterial N flow was calculated using the ratios between PPB and N in bacteria and at the duodenum.

On 2 consecutive days of the experimental period (i.e., d8 and d9) at 09:00 h (i.e., immediately before feeding) and 11:00 h (i.e., 2 h after feeding), a 100-ml rumen liquid sample was obtained via a tube introduced into the ruminal ventral sac through the rumen cannula, and pH was immediately measured. Subsamples were stored after preservation with a 0.05 *o*-phosphoric acid solution (1 ml/9 ml of rumen fluid) for VFA analysis, with a fixation solution (50 ml glycerol, 2 ml 0.25 formaldehyde, 48 ml distilled water) at 1 ml/ml of rumen fluid for protozoal counting, and with a 0.20 NaCl solution at 4 ml/ml of rumen fluid for ammonia N determination. Samples used for ammonia N and VFA analysis were immediately stored at  $-20^\circ\text{C}$ , and the samples used for protozoal counts were stored at  $4^\circ\text{C}$ . Protozoa were counted in a Dollfuss cell (Maillièrre, Aubière, France). The VFA concentration were determined by gas–liquid chromatography (Jouany, 1982), and ammonia N was analysed as described previously.

Blood was collected from the caudal vein at 08:30 h at the end of each period (d10). After centrifugation at  $1000 \times g$ , plasma was sampled and frozen until urea analysis, which was conducted on an autoanalyzer using diacetylmonoxime, by the improved method of Sahnoune et al. (1991).

Ruminal *in situ* measurements were used to obtain the CP degradability of rolled linseeds, of Croquelin®, the mixture of wheat bran and sunflower meal (60/40 on a DM basis) and linseed meal. Samples of the feedstuffs were used as fed to the cows, and were not ground before being placed into the *in situ* bags. Measurements were made on all four cows at week 3 of period 4, so that the mean value was an average of the degradation obtained with the 4 diets (i.e., one cow/diet). Nylon bags (pore size, 46  $\mu\text{m}$ ; Ankom, Fairport, NY, USA) containing 3 g of feeds were placed in the rumen of each cow at 09:00 h and removed after 2, 4, 7, 10, 17, 24 and 48 h of incubation (i.e., 2 bags/cow and per sampling time for each feed). Kinetics of degradation of DM and CP were fitted to an exponential model (Ørskov and McDonald, 1979) as

$$D(t) = a + b(1 - e^{-ct})$$

where  $D$  = degradation rate of feedstuff,  $a$  = rapidly degradable fraction (g/kg),  $b$  = slowly degradable fraction (g/kg) and  $c$  = degradation rate of  $b$  ( $\text{h}^{-1}$ ) and  $t$  is the time of incubation. The theoretical

**Table 2**Dry matter and crude protein degradation<sup>a</sup> of feedstuffs<sup>b</sup>.

	Rolled linseeds	Wheat bran + sunflower meal (60/40)	Extruded mixture of linseed, wheat bran and sunflower meal (50/30/20) <sup>c</sup>	Linseed meal	S.E.M.
Dry matter					
<i>a</i> (g/kg)	165 <sup>a</sup>	238 <sup>b</sup>	187 <sup>a</sup>	115 <sup>c</sup>	9.5
<i>b</i> (g/kg)	702 <sup>a</sup>	417 <sup>b</sup>	426 <sup>b</sup>	601 <sup>a</sup>	28.5
<i>c</i> (h <sup>-1</sup> )	0.0780 <sup>ab</sup>	0.0930 <sup>a</sup>	0.0555 <sup>b</sup>	0.0682 <sup>b</sup>	0.00531
TD	0.534 <sup>a</sup>	0.595 <sup>b</sup>	0.600 <sup>b</sup>	0.607 <sup>b</sup>	0.0142
Crude protein					
<i>a</i> (g/kg)	43 <sup>a</sup>	44 <sup>a</sup>	23 <sup>b</sup>	4 <sup>b</sup>	2.2
<i>b</i> (g/kg)	630 <sup>a</sup>	541 <sup>ab</sup>	475 <sup>b</sup>	595 <sup>a</sup>	9.1
<i>c</i> (h <sup>-1</sup> )	0.0769 <sup>a</sup>	0.0976 <sup>b</sup>	0.0515 <sup>c</sup>	0.0550 <sup>c</sup>	0.00369
TD	0.681 <sup>a</sup>	0.744 <sup>c</sup>	0.724 <sup>bc</sup>	0.694 <sup>ab</sup>	0.0073

<sup>a</sup> *a*, *b* and *c* are the parameters of the exponential equation between degradation and time. *a*, rapidly degradable fraction; *b*, slowly degradable fraction; *c*, degradation rate of *b*; TD, theoretical degradability calculated using a particulate outflow rate from the rumen of 0.06 h<sup>-1</sup>.

<sup>b</sup> Means with different superscripts differ (*P*<0.05).

<sup>c</sup> Croquelin® (Valorex, Combourtillé, France).

degradability (TD) was calculated as:

$$TD = a + b \times c / (c + k)$$

with particle passage rate from the rumen (*k*) assumed to be 0.06 h<sup>-1</sup>.

### 2.3. Statistical analysis

All data except *in situ* measurements were analysed as a 4 × 4 Latin square using PROC MIXED of SAS (1997). The statistical model included the effect of cow as random with period and treatment as fixed effects. Differences between means were determined using the Student's–Newman–Keuls test. Significance was declared at *P*<0.05 and trends were discussed if 0.05<*P*<0.1. *In situ* measurements were analysed by one-way analysis of variance. The analysed effect was the cow, but it is noted that cow and diet are confounded.

## 3. Results

### 3.1. Degradation of feedstuffs

The theoretical DM degradability was similar for linseed meal and the wheat bran/sunflower meal mixture and Croquelin®, and lower for rolled linseeds (Table 2). The theoretical CP degradability was lowest for rolled linseed and highest for the wheat bran/sunflower meal mixture.

### 3.2. Digestibility, ruminal metabolism and intestinal digestion

Total tract and forestomach digestibility did not differ among diets for DM, OM and NDF (Table 3). Nitrogen flows (i.e., intake, excreted, urinary, retained) did not differ among diets (Table 4). Duodenal nonammonia N was similar among diets, but microbial N flow tended (i.e., *P*=0.07) to a lower flow for RL versus C, diets EL and LO being intermediate, whereas nonmicrobial N flow did not vary among diets. As a consequence, the ratio of microbial N flow and DM intake tended to be lower (*P*=0.06) for RL versus the C diet. Efficiency of microbial synthesis, expressed as the ratio of microbial N flow and OM digested in the rumen, tended to be lower (*P*=0.06) for RL than for the other diets.



**Table 3**

Intake, total tract apparent digestibility, and ruminal and intestinal organic matter digestibility in cows fed linseed in different forms<sup>a</sup>.

	Diet <sup>b</sup>				S.E.M.
	C	RL	EL	LO	
DM intake (kg/d)	10.5	10.0	9.9	9.9	0.24
Total tract apparent digestibility					
DM	0.695	0.700	0.696	0.704	0.0078
OM	0.718	0.720	0.718	0.724	0.0073
NDF	0.527	0.558	0.541	0.525	0.0119
ADF	0.448	0.486	0.471	0.452	0.0150
Forestomach OM digestibility					
g/kg OM intake	533	598	518	517	36.4
g/kg OM totally digested	742	830	721	715	47.7
Intestinal OM digestibility					
g/kg OM intake	185	122	200	206	34.9
g/kg OM totally digested	258	170	279	285	47.7
Forestomach NDF digestibility					
g/kg NDF intake	400	476	456	419	28.4
g/kg NDF totally digested	759	851	845	800	55.1
Intestinal NDF digestibility					
g/kg NDF intake	127	82	85	106	29.3
g/kg NDF totally digested	241	149	155	200	54.9

<sup>a</sup> For all variables, differences among diets were not significant (i.e.,  $P>0.05$ ).

<sup>b</sup> C = control; RL = rolled linseeds; EL = extruded linseeds; LO = linseed oil + linseed meal.

Ammonia concentration in the rumen did not vary among diets, both before and after the morning feeding (Table 5). This result was consistent with the absence of difference in plasma urea among the diets. The VFA concentrations and proportions of VFA did not differ among diets either before or after the morning feeding (Table 5). The protozoal concentration in rumen fluid was also not affected by diet, before and after morning feeding, either in terms of total population or of the two main classes (i.e., *Isotrichidae* and *Ophryoscolecidae*).

**Table 4**

Nitrogen balance, N ruminal digestion and intestinal digestibility in cows fed linseed in different forms<sup>a</sup>.

	Diet <sup>b</sup>				S.E.M.
	C	RL	EL	LO	
N intake (g/d)	243	238	235	236	1.2
N in faeces (g/d)	76	75	70	72	1.9
N in urine (g/d)	136	142	142	132	5.3
N retained (g/d)	31	21	22	33	7.3
Duodenal NAN <sup>c</sup> (g/d)	204	171	176	188	9.2
Microbial (g/d)	107	82	92	94	5.3
Nonmicrobial (g/d)	97	89	84	94	9.5
Duodenal NAN (g/kg N intake)	854	721	757	800	40.5
Microbial N (g/kg DM intake)	104	81	93	97	5.0
Microbial N (g/kg OMDR <sup>d</sup> )	22.2	14.5	20.1	20.2	1.60
Nonmicrobial N (g/kg DM intake)	9.2	9.0	8.5	9.5	0.76
N intestinal disappearance	0.630	0.559	0.591	0.620	0.0305

<sup>a</sup> For all variables, differences among diets were not significant (i.e.,  $P>0.05$ ). All trends (i.e.,  $P<0.10$ ) referred to in the text.

<sup>b</sup> C = control; RL = rolled linseeds; EL = extruded linseeds; LO = linseed oil + linseed meal.

<sup>c</sup> NAN, non-ammonia N.

<sup>d</sup> OMDR, OM apparently digested in the rumen.

**Table 5**Ammonia, volatile fatty acids and protozoa in rumen fluid and plasma urea of cows fed linseed in different forms<sup>a</sup>.

	Diet <sup>b</sup>				
	C	RL	EL	LO	S.E.M.
Ammonia in rumen fluid (mg/L)					
Before feeding	337	378	332	319	37.4
2 h after feeding	775	797	756	731	47.2
Volatile fatty acids in rumen fluid (mmol/L)					
Before feeding	87	88	83	83	6.0
2 h after feeding	131	122	115	121	6.5
Volatile fatty acids in rumen fluid (mol/100 mol)					
Before feeding					
Acetate	66.1	70.2	68.7	69.3	2.34
Propionate	17.6	16.6	16.6	16.7	0.82
Butyrate	10.0	9.1	9.8	9.7	0.47
2 h after feeding					
Acetate	64.3	63.7	64.6	63.4	1.22
Propionate	20.4	21.0	19.9	20.9	1.18
Butyrate	10.8	11.2	10.9	11.3	0.37
Protozoa in rumen fluid (10 <sup>3</sup> mL <sup>-1</sup> )					
Before feeding					
Total	186	205	197	178	34.6
<i>Isotrichidae</i>	4.4	5.5	3.5	5.8	0.65
<i>Ophryoscolecidae</i>	182	199	193	172	34.8
2 h after feeding					
Total	157	167	111	137	39.7
<i>Isotrichidae</i>	3.5	3.5	2.0	3.8	0.87
<i>Ophryoscolecidae</i>	154	164	109	133	39.8
Plasma urea (mg/L)	202	202	216	228	19.4

<sup>a</sup> For all variables, differences among diets were not significant (i.e.,  $P > 0.05$ ).<sup>b</sup> C = control; RL = rolled linseeds; EL = extruded linseeds; LO = linseed oil + linseed meal.

## 4. Discussion

### 4.1. Digestibility and ruminal digestion

Previous experiments have shown negative effects of linseed oil on total tract and forestomach OM and, especially, fibre digestibility (Cottyn et al., 1971; Ikwuegbu and Sutton, 1982; Sutton et al., 1983; Broudiscou et al., 1994) with sheep fed at maintenance receiving more than 50 g/kg diet DM of linseed oil. However, this is not consistent with results of Ueda et al. (2003) with a supplement of 30 g/kg DM linseed oil in lactating cows, and of Machmüller et al. (2000) and Sutter et al. (1999) who reported total tract digestibility in steers and bulls receiving 20–30 g/kg diet DM oil from linseeds, and did not find any decrease in OM and NDF digestibility due to linseed supply. Similarly, Wachira et al. (2000) did not show any difference between sheep fed diets supplemented with linseeds and palm oil calcium salts. Three hypotheses can be proposed to explain these differences, being the effect of linseed on ruminal digestion depends on the level of linseed FA supply, it depends on the feeding level (i.e., at maintenance, negative effects of linseed on digestion can be higher than in high-producing animals due to a higher retention time of digesta in the rumen at low intake), the effect depends on the form of linseed. In the present study, total tract and forestomach digestibility was not modified in dry cows by a supplement of 0.026 oil from linseed, regardless of the oil form. In another study with dairy cows, Martin et al. (2008) observed a decrease in total tract digestibility with a supply of 0.05 oil from linseed fed crude, extruded, or free oil in dairy cows. These results suggest that the first hypothesis (i.e., effect of linseed level) is the most likely, and that effects of feed intake and the form of feeding the lipids is not likely to be important. However, Gonthier et al. (2004) did observe an increase



in total (but not forestomach) digestibility of OM and fibre when feeding a 27 g/kg DM linseed oil supplement.

The absence of a difference in total tract and forestomach OM and fibre digestibility due to extrusion of linseeds is partly due to the absence of general effect of linseed incorporation in this experiment on ruminal fermentation. In addition, Gonthier et al. (2004) and Martin et al. (2008) with linseeds, Ferlay et al. (1992); Albro et al. (1993) and Petit et al. (1997) with soybeans or rapeseeds, did not find any effect of extrusion on forestomach and total tract OM or fibre digestibility. Likewise, with oil and whole seeds, Pallister and Smithard (1987) did not find differences in digestion of rapeseed oil *versus* oil included in rapeseeds while, more recently, Martin et al. (2008) did not find differences between linseed oil and oil included in linseeds.

#### 4.2. Parameters of ruminal digestion

A high supply of linseed oil has been shown in the literature to increase propionic acid at the expense of acetic and butyric acid; as a consequence the proportion of propionic acid increases from less than 0.20 in control diets to 0.37 in diets containing more than 50 g/kg DM linseed oil (Ikwuegbu and Sutton, 1982; Sutton et al., 1983). Linseed oil has a more pronounced effect than oils from other oleaginous seeds (see Jouany et al. (2000) for review). Even when forestomach digestibility was not reduced by a supply of oil from linseeds, propionate increased at the expense of either butyrate (Machmüller et al., 2000) or acetate (Ueda et al., 2003; Gonthier et al., 2004). Our study is one of the few, along with Sutter et al. (1999), to report no modification in proportions of ruminal VFA. Our oil supply was apparently too low to modify ruminal fermentation.

A well-known characteristic of linseed oil is its defaunating effect in the rumen, with coconut being the only other oil to have a similar effect (see review by Doreau and Ferlay, 1995). When large amounts of linseed oil are fed to sheep, protozoa almost disappear (Czerkawski et al., 1975; Ikwuegbu and Sutton, 1982), which could be the cause of the decrease in butyrate proportion (Jouany and Ushida, 1999). The decrease in protozoa is of lower extent when lipids are included in linseeds, as shown with cattle (Machmüller et al., 2000; Ueda et al., 2003) and with sheep at maintenance (Purser and Moir, 1966). The absence of an effect on protozoa concentration in the present study is consistent with the absence of change in butyrate proportion, and is another factor in the absence of modification of rumen fermentation.

#### 4.3. Nitrogen metabolism

The form of the supply of linseed lipids in the diet did not modify duodenal N flow, which is consistent with general effects of lipids on ruminal N metabolism as described in the review by Doreau and Ferlay (1995). The absence of an effect of lipids was not surprising, given that there was no difference in forestomach OM digestion which is consistent with the results of the *in situ* degradation study which suggested that rolled linseeds are only slightly protected against ruminal degradation. However, it is surprising that extruded seed was not protected against microbial degradation, as it has been shown that extrusion limits ruminal CP degradation, especially due to its temperature effect (review by Poncet et al., 2003). The 120°C temperature used in our study was probably not high enough to limit CP degradation. Although extrusion has been used to protect CP of leguminous grains, such as peas, lupins and soybeans (Poncet and Rémond, 2002) from ruminal degradation, the effect on oilseeds rich in lipids has seldom been observed. *In vivo*, Ferlay et al. (1992) did not observe CP protection in rapeseeds extruded at 140°C, while Gonthier et al. (2004) did not observe any difference with linseeds extruded at 155°C. With the latter product, Ouellet et al. (2003) observed an increase in CP degradation due to extrusion. The mechanical action of extrusion may be reduced by the oily nature of the seed, even when, as in our experiment, linseeds are extruded together with bran and sunflower meal. This may explain the similar degradation of extruded linseed and low-temperature expelled linseed meal in this experiment.

## 5. Conclusions

A moderate supply of lipids from linseeds does not decrease the extent of forestomach and total tract digestion in cattle fed a diet based on maize silage, alfalfa hay and concentrate. This is consistent with effects on dairy cows (Loor et al., 2005) and fattening cattle (Sutter et al., 1999; Normand et al., 2005). The extrusion process used in our study did not succeed in protecting proteins from ruminal degradation, possibly due to the low temperature chosen to avoid a risk of degradation, or oxidation, of fatty acids. Extrusion did not improve the protein value of linseeds.

## Acknowledgements

This experiment received funding from Valorex (Combourtillé, France) and from the ACTA (Association pour la Coopération Technique Agricole, Paris, France). The authors thank the various INRA staffs whose skills were particularly appreciated, especially F. Anglard and D. Mathevon for animal care, feeding and sampling, P. Amblard, B. Chauveau-Duriot, M. Fabre and L. Genestoux for laboratory analyses, and Y. Rochette for statistical analyses.

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