Ann Zootech (1992) 41, 137-143

Original article

Effects of supplementation with hydrogenated fish fat on digestion in dairy cows

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(Received 26 September 1991; accepted 6 January 1992)

Summary — Three dry cows received a diet based on maize silage according to a 3 x 3 Latin square design, without or with a fat supplement composed either of crystalline hydrogenated fish oil (rich in arachidic and behenic acids) or rapeseed oil (rich in unsaturated fatty acids). The fatty acid content of the diets was 1.5, 8.6 and 8.7% dry matter (DM) respectively. Fish oil supplementation did not decrease fibre digestion and did not modify ruminal fermentation, in contrast to rapeseed oil which altered ruminal fermentation. Digestibilities of lipids and fatty acids were lower when the diet was supplemented with fish oil than when the other 2 diets were fed because of the low digestibility of all fatty acids, whatever the chain length.

cow / digestion / lipids

Résumé — Effets d'une supplémentation en huile de poisson hydrogénée sur la digestion chez la vache. *Trois vaches taries ont été utilisées dans un schéma en carré latin. Elles ont reçu soit un régime témoin (T) à base d'ensilage de maïs, soit ce régime supplémenté avec de l'huile de poisson hydrogénée (HPH, riche en acides arachidique et béhénique) présentée sous forme cristalline, soit le régime témoin supplémenté avec de l'huile de colza (HC, riche en acides gras polyinsaturés). La teneur en acides gras de ces trois régimes était respectivement de 1,5; 8,6 et 8,7% de la matière sèche. La supplémentation en huile de poisson n'a pas modifié la digestibilité des parois végétales et les fermentations du rumen, contrairement à l'addition d'huile de colza : la digestibilité du NDF a été respectivement de 73,9; 72,8 et 63,5 pour les régimes T, HPH et HC. En revanche, la digestibilité apparente des lipides et des acides gras a été plus faible pour le régime HPH. Cette dernière a été respectivement de 71,7; 44,9 et 72,0% pour les régimes T, HPH et HC. La digestibilité des acides gras du régime HPH a été faible quelle que soit leur longueur de chaîne.*

vache / digestion / lipides

INTRODUCTION

Fish oils have seldom been used in feeding cows, primarily because of the drop in milk fat percentage observed with these supplements (review by Opstvedt, 1984). However, the few experiments on digestion in diets containing fish oils did not show marked disturbances in carbohydrate digestion, either when fish oils were hydrogenated (Sundstøl, 1974) or not (Sutton *et al*, 1975). There is no evidence of a decrease in ruminal digestibility when these kinds of lipids are added to the diet.

Moreover, the digestibility of such lipids in the small intestine appears high (Andrews and Lewis, 1970).

The recent trends in dairy cow nutrition (increasing the fat content of diets, without the necessity of maintaining a high butterfat content) have stimulated renewed interest in fish oil supplementation. In addition, the use of technological treatments such as crystallisation may be helpful in avoiding disturbances in rumen fermentation.

In this trial, the consequences of hydrogenated fish oil supplementation on fat digestibility in cows have been studied. In order to improve the interpretation of results, this fat-supplemented diet was compared: 1), to a control diet, without lipid addition; and 2), to a diet supplemented with rapeseed oil that is efficiently digested in the intestines but which is known to decrease fibre digestion in the rumen.

MATERIAL AND METHODS

Animals, experimental design and diets

Three non-lactating Holstein cows, weighing on average 753 kg and fitted with ruminal cannulae, were utilized in a 3 x 3 Latin square design. Each period consisted of 3 or 4 weeks for adaptation to the diets and 1 week for experimental measurements.

Three diets were fed to the cows (table I). The control diet (C) consisted of 70% maize silage and 30% concentrates on a dry matter (DM) basis, of urea and of mineral premix containing 14% Ca and 14% P. This diet was fed in amounts so that energy, nitrogen and mineral requirements were met. Diet HFO was diet C supplemented with 500 g of crystalline hydrogenated fish oil (Enerjet 98, Relasa, Sesena, Spain). Lipids of this product were 92.8% free fatty acids and 7.2% glycerides. Diet RO was diet C supplemented with 500 g rapeseed oil.

Table I. Intake and chemical composition of experimental diets.

	Diet		
	C	*HFO*	*RO*
Intake (g DM/d)			
Maize silage	4 340	4 340	4 340
Concentrates [1]	1 860	1 860	1 860
Urea	60	60	60
Mineral premix	100	100	100
Hydrogenated fish oil	–	500	–
Rapeseed oil	–	–	500
Chemical composition (g/100 g DM)			
Organic matter	92.9	93.5	93.5
NDF	39.6	36.6	36.6
ADF	16.9	15.6	15.6
Crude protein	14.2	13.1	13.1
Lipids	3.7	10.8	10.9
Fatty acids	1.5	8.6	8.7
Calcium	0.88	0.82	0.82

[1] Composition: 20% wheat, 20% barley, 30% beet pulp, 15% rapeseed meal, 7% soybean meal, 5% beet molasses, 1% limestone, 1% dicalcium phosphate, 0.5% magnesium oxide, 0.5% sodium chloride.

Chemical composition of diets is given in table I. Composition of hydrogenated fish oil and rapeseed oil fatty acids is shown in table II.

Diets including lipidic supplements were fed as a total mixed ration. Cows were fed twice daily in equal portions at 09.00 h and 16.00 h.

Measurements

Digestibility was measured by total collection of feces over a 5-day period. Two representative samples of feeds and feces were taken. The first was dried and used for analyses of ash, NDF and ADF; the second was frozen and then lyophilised for determination of lipids and fatty acids.

Samples of rumen liquor were taken from the ventral sac for 2 consecutive days at 09.00 h (before food distribution) and 11.30 h. PH was measured immediately; a sample was taken for volatile fatty acids (VFA) determination, with orthophosphoric acid as preservative.

Nylon bags (internal size: 6 x 11 cm, pore size: 46 μm) were filled with 3 g of dry maize silage without cobs that had been ground through a 0.8-mm screen. Triplicate bags were introduced into the ventral sac of the rumen at 09.00 h and incubated for 3, 6, 12, 24 and 48 h.

Analyses

Dry matter content of feeds and feces was determined by oven-drying at 80 °C for 48 h. Ashing was performed at 550 °C for 6 h. NDF and ADF were analyzed by the method of Goering and Van Soest (1970). The difference between NDF and ADF was considered as hemicellulose. Calcium was analyzed by atomic absorption spectrophotometry. Lipid and fatty acid composition of feed and feces were determined according to the method described by Bauchart *et al* (1990): lipids were first extracted by the method of Folch *et al* (1957), then in hexane – ethanol – hydrochloric acid (5:2:2, by vol), and then determined gravimetrically. Lipids were saponified and their fatty acids methylated. Methyl esters were separated by GLC at 195 °C using a glass capillary column coated with FFA phase. Methyl heptadecanoate was used as the internal standard.

Concentration and composition of VFA in rumen fluid were determined by GLC (Jouany, 1982).

Nylon bags were washed in cold water before drying at 80 °C for 48 h and weighing. The kinetics were adjusted to an exponential model: $D = a + b\,(1 - e^{-ct})$ where D is the percentage of DM disappearance at time t, a and b the rapidly and slowly degradable components, and c the fractional rate of degradation of component b (Ørskov and McDonald, 1979).

Statistical analyses were performed by analysis of variance with 3 factors: animal, period, treatment. When treatments differed significantly, comparisons between 2 treatments were made by a Duncan test. Results are expressed as mean and pooled standard error of the mean (SEM).

Table II. Fatty acid composition of lipidic supplements (g/100 g methyl esters).

	Hydrogenated fish oil	*Rapeseed oil*
C14:0 (myristic acid)	7.3	0.1
C16:0 (palmitic acid)	30.4	5.0
C18:0 (stearic acid)	22.2	1.7
C18:1 n–9 (oleic acid)	0.8	57.6
C18:2 n–6 (linolenic acid)	0.1	21.1
C18:3 n–3 (linolenic acid)	–	9.4
C20:0 (arachidic acid)	20.0	0.6
C22:0 (behenic acid)	11.9	0.4

RESULTS

Organic matter digestibility was significantly lower for diets HFO and RO than for diet C (table III). The cause of this drop in digestibility is not the same for diet HFO and for diet RO. Indeed, digestibility of all fibre fractions (NDF, ADF and hemicellulose) was significantly lower for diet RO than for diet C, but did not differ between diets HFO and C. On the contrary, apparent digestibility of all lipid fractions (total lipids, fatty acids, non-fatty acid lipids was not different between diets RO and C, but was

Table III. Apparent digestibility of nutrients (%).

	Diet			*SEM*
	C	*HFO*	*RO*	
Dry matter	77.7[a]	75.0[ab]	72.3[b]	0.31
Organic matter	81.6[a]	78.7[b]	76.3[b]	0.23
NDF	73.9[a]	72.8[a]	63.5[b]	0.51
ADF	68.3[a]	67.8[a]	55.7[b]	0.92
Hemicellulose	78.1[A]	76.4[A]	69.3[B]	0.25
Lipids	62.5	45.1	68.3	1.17
Fatty acids	71.7[A]	44.9[B]	72.0[A]	0.17
16C and 18C fatty acids	72.0[A]	47.8[B]	75.3[A]	1.02
20C fatty acid	–37.6	44.9	–27.2	16.61
22C fatty acids	31.3	33.7	– 3.1	8.00
Non-fatty acid lipids	55.9	45.2	54.4	3.66

Values are means for 3 cows per diet. SEM: standard error of the mean. 16C, 18C, 20C and 22C represent the sum of fatty acids with 16, 18, 20 and 22 carbons, respectively. Means on the same row with different superscripts differ significantly ([a, b]: $P < 0.05$; [A, B]: $P < 0.01$).

lower for diet HFO. The drop in digestibility between diets C and HFO for lipids, fatty acids and non-fatty acid lipids was 17.4, 26.8 and 10.7 percentage units, respectively. The trend was not significant for lipids and non-fatty acid lipids, due to a high SEM values, but was significant for fatty acids. The lower digestibility of fatty acids for diet HFO than for diets C and RO was because of a low digestibility of all fatty acids, including fatty acids with 16 and 18 carbons. Although means varied, digestibility of C20 and C22 did not differ significantly among diets. Lack of significance is explained by the extremely high SEM values, caused by the very low intake for diets C and RO.

The mean pH did not vary among diets (table IV). VFA concentration and composition show a non significant trend toward a decrease in concentration and in butyrate percentage, and a significant postprandial increase in propionate percentage for diet RO when compared to the other 2 diets.

Difference among diets was shown to be non-significant by the *in sacco* technique (table V), either for percentages of DM disappearance at different incubation times or for parameters of the mathematical model.

DISCUSSION

The decrease in organic matter and fibre digestibilities with addition of rapeseed oil to a maize silage diet was the same as in a previous trial (Doreau *et al*, 1991a). In this trial as in another experiment carried out with rapeseed (Doreau *et al*, 1991b), the decrease in digestibility is not related to a decrease in *in sacco* fibre degradation. As lipid supply generally causes no variation in retention time of particles in the rumen

Table IV. pH, concentration and composition of volatile fatty acids in rumen liquor at two sampling times, before and 2.5 h after feeding.

		Diet			*SEM*
		C	*HFO*	*RO*	
pH	09.00 h	6.91	6.98	7.13	0.025
	11.30 h	6.68	6.59	6.61	0.030
Volatile fatty acids (mMol/l)	09.00 h	57.1	51.3	42.8	1.70
	11.30 h	71.7	76.0	68.6	5.48
Acetate (mol/100 mol)	09.00 h	66.3	66.0	66.9	0.30
	11.30 h	61.7	61.6	61.4	0.23
Propionate (mol/100 mol)	09.00 h	14.9	14.3	15.5	0.14
	11.30 h	17.5[a]	17.7[a]	20.7[b]	0.19
Butyrate (mol/100 mol)	09.00 h	13.0	14.0	11.3	0.34
	11.30 h	13.7	14.2	11.2	0.30

Values are means for 3 cows per diet. SEM: standard error of the mean. Means on the same row with different superscripts differ significantly ($P < 0.05$).

Table V. Degradation of dry matter *in sacco*: percentages of disappearance at different times of incubation and parameters of the model.

	Diet			*SEM*
	C	*HFO*	*RO*	
Incubation time				
3 h	39.6	39.1	39.2	0.24
6 h	43.3	44.4	42.9	0.31
12 h	51.9	54.6	50.5	1.37
24 h	64.9	68.1	62.8	1.50
48 h	74.8	76.5	71.7	1.40
Parameters of the model				
a (%)	33.0	32.9	30.8	0.87
b (%)	48.7	45.5	49.7	2.22
c (%/h)	4.33	4.47	5.61	0.54

Values are means for 3 cows per diet. SEM: standard error of the mean.

(Ferlay *et al*, 1991), this lack of consistency could be explained by a negative effect of rapeseed oil on protozoal number and activity, as suggested by a decrease in butyrate proportion in rumen VFA. Protozoal action is not taken into account in *in sacco* measurements. Indeed, size of most protozoa species is higher than bag pore size.

The absence of changes in fibre digestion when hydrogenated fish oil was added to the diet may be because of saturated fatty acids, which are less determinant for microbial growth than unsaturated fatty acids (Maczulak *et al*, 1981) and to the high melting point of this fat, which may cause a by-pass of the rumen. However, when such fats are fed in large amounts, Sundstøl (1974) observed a decrease in fibre digestibility, showing that a high melting point does not prevent fat from altering ruminal fermentation. With crude fish oils,

changes in VFA composition obtained by several authors in cows receiving 225 to 300 g/d cod liver oil ranged from no effect (Pennington and Davis, 1975) to a large increase in propionic acid (Shaw and Ensor, 1959). The only complete digestion experiment did not show a decrease in ruminal fibre digestion (Sutton *et al,* 1975).

Although saturated fatty acids sometimes have a low digestibility (Jenkins and Jenny, 1989), the low digestibility of fatty acids in the HFO diet was surprising. Andrews and Lewis (1970) showed a high total digestibility of herring oil. They did not calculate digestibility of total 20- and 22-carbon fatty acids, but from their results it can be estimated to be more than 80%. In the same manner, Sutton *et al* (1975) noted a high total digestibility of fatty acids from cod liver oil. Børsting and Weisbjerg (1989) measured digestibility in the small intestine that was close to 80% for arachidic and behenic acids in cows fed protected fish oils. In contrast to an earlier idea that an increase in intake of fat caused increase in digestibility of fat, it is now thought that too much fat in the diet can depress digestibility (Tamminga and Doreau, 1991). It could be postulated that in this experiment the level of dietary arachidic and behenic acids is too high to prevent fat digestibility depression. However, Sundstøl (1974) observed high digestibility of ether extract when hydrogenated fish oil was fed to sheeps. As digestibility of all fatty acids in impaired in the present trial, the main hypothesis to explain this low digestibility is an incomplete micellar organization of fats. No evidence for this hypothesis has been shown either in ruminants or in monogastric animals. However, it should be noted that Børsting and Weisbjerg (1989) observed a lower bile acid secretion and a higher reabsorption when fish oil was fed to cows than when other fat sources were fed, although this had no consequence on digestibility of the oil.

CONCLUSION

This trial has shown the absence of negative effects of crystalline hydrogenated fish oil on rumen fibre digestion. The low intestinal digestibility of lipids has to be assessed by new experiments in which the causes of variation in intestinal absorption can be analyzed.

ACKNOWLEDGMENTS

The authors acknowledge the financial assistance of SCD Chauveau, La Brechoire, 49280 Cholet, France. Thanks are due to E Girard and D Thomas for care of animals and help in sampling and to G Sauvage, J Chabrot and J Fléchet for technical assistance.

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