Italian Journal of Animal Science

ISSN: (Print) 1828-051X (Online) Journal homepage: https://www.tandfonline.com/loi/tjas20

In situ and *in vitro* nutritional evaluation of rumen-protected lipids

F. Rossi, A.M. Pulimeno & F. Masoero

To cite this article: F. Rossi, A.M. Pulimeno & F. Masoero (2005) *In situ* and *in vitro* nutritional evaluation of rumen-protected lipids, Italian Journal of Animal Science, 4:sup2, 156-158, DOI: 10.4081/ijas.2005.2s.156

To link to this article: https://doi.org/10.4081/ijas.2005.2s.156

In situ and *in vitro* nutritional evaluation of rumen-protected lipids

F. Rossi, A.M. Pulimeno, F. Masoero

Istituto Scienze degli Alimenti e della Nutrizione, Università Cattolica del Sacro Cuore, Italy

Corresponding author: Filippo Rossi. Istituto Scienze degli Alimenti e della Nutrizione. Via Emilia Parmense 84, 29100 Piacenza, Italy – Tel: +39 0523 599286 – Fax: +39 0523 599259 – E-mail: filippo.rossi@unicatt.it

RIASSUNTO – Valutazione di lipidi rumino-protetti *in situ e in vitro. E' stata determinata la degradabilità ruminale in situ e la digeribilità in vitro di sette diversi integratori lipidici rumino-protetti mediante saponificazione. La degradabilità ruminale è risultata bassa (inferiore al 40% dopo 48 ore di incubazione) ed inversamente proporzionale al contenuto in ceneri dei prodotti, mentre la digeribilità in vitro non è risultata correlata alla composizione dei lipidi rumino-protetti, al loro profilo acidico o alla degradabilità ruminale.*

Key words: lipids, rumen degradation, digestibility, in vitro.

INTRODUCTION – Rumen-protected lipids are a class of products which is increasingly used in ruminant nutrition even if the results are not homogeneous. The different results may be due to different analytical or technological characteristics. Aim of this work was therefore to compare the *in situ* rumen behaviour of different soaps as well as their *in vitro* intestinal digestibility.

MATERIAL AND METHODS – Seven different types of rumen-protected lipids commercially available in Italy, identified as S1, S2, S3, S4, S5, S6, plus tallow soaps were used. The samples were analyzed for lipid and mineral content, fatty acid pattern and free fatty acids (FFA). *In vitro* intestinal digestibility was assessed by putting 1 g of the sample in an HCl 0.01 N solution (pH 2) at 39°C for 1 hour. After this period of time, 1 ml of NaOH 1N and 30 ml of a pancreatin solution (0.5M KH_2PO_4 buffer at pH 7.8 and 6g/l enzyme, Sigma P-1750) were added. The whole was incubated at 39°C for 24 hours; at the end, the sample residue was collected with the filtration method then dried in an oven in vacuum conditions at 35°C and finally analyzed for its lipid content. The difference between the initial and final lipid content, taking 100 as the reference value, provides an estimate of lipid *in vitro* digestibility. Rumen degradability was measured by means of the *in situ* technique using nylon bags with a size of 12.5x8 cm and pore size of 46 mm, filled with 3.5 g of sample. 3 Italian Frisian cows with rumen fistula housed indoors in controlled environmental conditions were used. The following diet was administered during all the trial: 8 kg grass hay; 5 kg corn silage; 2 kg concentrate (18% crude protein as fed). The bags were placed into the rumen before the morning meal and incubated for 8, 24 and 48 hours. Two replicates for each incubation period/animal and kinetic were obtained for each sample; the number of kinetic was two. The randomized block was used as statistical model considering the effect of the sample, of the cow and of the kinetics. Once removed, the bags were washed with cold water and then placed in a 1% carboxymethylcellulose solution in order to detach the rumen micro-organisms adhering to the lipid substrate and wash them out. They were then placed in an oven at 35°C till a constant weight could be obtained. The results thus obtained were statistically analyzed using PROC GLM and PROC CORR of the SAS software.

RESULTS AND CONCLUSIONS – With the only exception of the S4 product, all supplements show similar fatty acids profile and may be included in two different categories (Table 1):
- S2 - S3 - S4 - S5: essentially made of palmitic and oleic acid with varying saturated/unsaturated fatty acid ratio;
- S1 - S6 - tallow soaps: made of palmitic, stearic and oleic acid with strong prevalence of saturated over unsaturated fatty acids.

Intestinal digestibility: the values are medium-high and fall within the range of variability observed by Doreau and Ferlay (1994). The S3 product shows the highest while the S1 product shows the lowest digestibility; this difference is not easy to explain since S6 and the tallow derivatives - with an acid profile which is almost identical to S1 - do not show any significant differences compared with S3. Even if Sauvant and Bas (2001) have reported that there is a difference in the digestibility of the individual fatty acids, no correlations were found between the *in vitro* digestibility and the lipid content or the saturated/unsaturated fatty acid content of the products. However, the work of Doreau and Ferlay (1994) shows that the *in vivo* lipid digestibility depends on several factors which have different modes of action: it increases dramatically from C12 to C16 but decreases in the case of greater lengths, especially from C20 onwards. Among the C18 acids, the oleic acid has greater digestibility than the stearic acid or linoleic and linolenic. This contrasting role of chain length and level of unsaturation may account for the absence of significant correlations.

Table 1. Lipid content and fatty acids composition (% of total fatty acids) of rumen-protected fats.

	S1	S2	S3	S4	S5	S6	Tallow
Lipids (% s.s.)	80.37	77.75	83.18	91.03	74.81	81.71	78.89
Ash (% s.s.)	19.63	22.25	16.82	8.97	25.19	18.29	21.11
Capric acid C 10:0	-	0.1	-	-	0.22	0.1	-
Lauric acid C 12:0	0.7	1.68	0.3	-	2.56	0.6	0.1
Miristic acid C14:0	1.2	1.88	0.8	1.5	2.12	1.2	1.0
Palmitic acid C 16:0	42	51.23	30.8	44.0	17.56	44.0	44.1
Stearic acid C 18:0	26.1	3.92	2.4	5.0	2.86	27.6	27.9
Oleic acid C 18:1	18.8	33.09	46.2	40.0	61.91	16.8	19.5
Linoleic acid C 18:2	9.8	6.68	13.1	9.5	8.73	8.5	6.5
Linolenic acid C 18:3	0.2	0.21	0.5	-	0.69	0.3	0.3
Arachidic acid C 20:0	1.1	0.22	-	-	0.33	-	0.5
Arachidonic acid C 20:4	-	-	4.8	-	-	-	0.0
Free fatty acids (%)	6.68	3.14	9.18	10.61	1.52	10.00	10.08
Satured fatty acids (%)	71.1	59.03	34.30	50.50	25.65	73.50	73.60
Unsatured fatty acids (%)	28.8	39.98	64.60	49.50	71.33	25.60	26.31

Table 2 shows that the resistance of the investigated lipid supplements to rumen degradation is high as shown by the low rumen degradability which does not exceed 40% even after 48 hours. The disappearance of the lipids from the nylon bags does not show a linear trend: hydrolysis is initially (8h) low, probably corresponding to the non-saponified percentage. After 24 hours the disappearance of the lipids from the nylon bags, however, only shows a minor increase as compared to the value at 8 hours. Longer incubation periods cause greater degradability probably because the lipolytic bacteria have more time to act. Both at 8 and 24 hours of incubation the S2 product shows the lowest rumen degradability while the S4 product shows the highest, probably because of the different ash content and, therefore, a different saponification degree between the two products.

Table 2. *In vitro* digestibility and in situ rumen degradability of lipids in several rumen protected fat supplement.

Products	Digestibility (%)	Rumen degradability (%) of fats		
		8h	24h	48h
S 1	75.22^{a}	26.99^{De}	27.46^{Bb}	33.94^{Dd}
S 2	80.08^{ab}	17.27^{Aa}	20.16^{Aa}	24.69^{Aa}
S 3	86.41^{b}	26.05^{Cdde}	26.97^{Bb}	30.56^{Cc}
S 4	77.32^{ab}	33.12^{Ef}	34.12^{Cc}	39.23^{Ef}
S 5	77.93^{ab}	25.21^{Ccd}	25.52^{Bb}	37.56^{Ee}
S 6	79.02^{ab}	21.51^{Bb}	25.92^{Bb}	27.09^{Bb}
Tallow	79.68^{ab}	23.87^{BCc}	26.73^{Bb}	33.30^{Dd}
SE	3.123	0.593	0.777	0.446

$^{a, b, c, d, e, f}$($P<0.05$); $^{A, B, C, D, E}$($P<0.01$).

Statistical analysis indeed shows an inverse correlation between the ash content and degradability at 24 hours ($r=-0.81$; $P<0.03$). At the shorter incubation time the statistical significance of this correlation is lower ($r=-0.70$; $P<0.08$). No significant correlation was found between the unsaturation level or the acid profile of the products and their intestinal or ruminal behaviour. The percentage of FFA shows a positive but poor correlation with rumen degradation at 24 hours of incubation ($r=0.63$; $P<0.13$).

Doreau and Ferlay (1994) have found a negative correlation between the saturated fatty acid content and rumen digestibility as well as a positive correlation for unsaturated fatty acids. In both cases, however, the correlation values obtained in this work do not confirm this hypothesis.

As conclusion, the rumen-protected lipids tested can be classified in two major classes:

- high saturated fatty acid content: S1, S2, tallow soaps;
- high palmitic and oleic acid content: S2, S3, S4 and S5.

Their rumen digestibility is low and related to the extent of the saponification process rather than to the presence of a particular fatty acid or saturation level. The *in vitro* intestinal digestibility, which range from 75 to 86%, does not depend on the fat content, on the acid composition of the products or on the amount of lipids which are not salified with Ca++.

REFERENCES – **Doreau**, M., Ferlay, A., 1994. Digestion and utilization of fatty acids by ruminants. Anim. Feed Sci Technol.. 45: 379-396. **Sauvant**, D., Bas, P., 2001. La digestion des lipids chez le ruminant. INRA Prod. Anim. 14: 303-310.