

Development of in vitro rumen model to measure digestibility of rumen by-pass fat

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- Belonging to the NuScience group - that is known in Europe for the production of high quality feed concepts for feed composers.
- Well known for its R&D activities in animal production
- Has its own laboratory, in order to perform bioactivity and (gastrointestinal) simulation assays.

STSM-Belgium



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Brussels



Gent

Aim: to build a model for *in vitro* assessment of rumen by-pass fat coated/encapsulated products digestibility

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Technical note: A modified three-step in vitro procedure to determine intestinal digestion of proteins

S. Gargallo, S. Calsamiglia and A. Ferret

J Anim Sci 2006. 84:2163-2167.
doi: 10.2527/jas.2004-704

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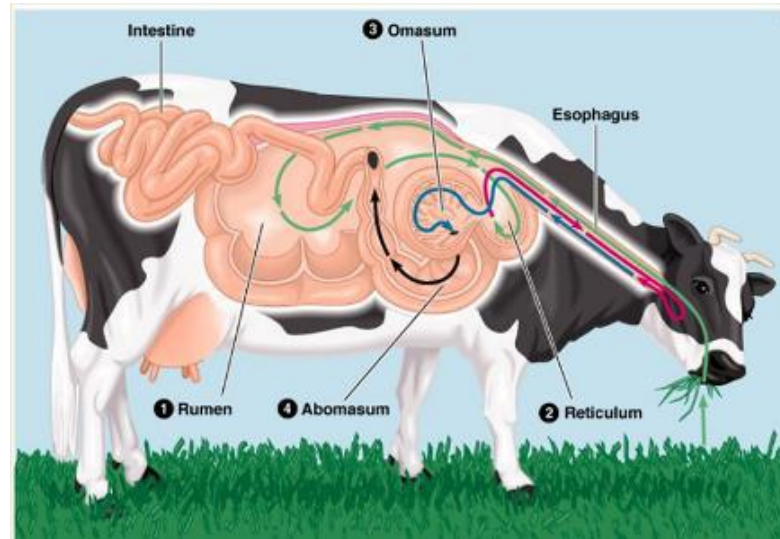
A three-step in vitro procedure for estimating intestinal digestion of protein in ruminants

S. Calsamiglia and M. D. Stern

J Anim Sci 1995. 73:1459-1465.



Three *in vitro* steps for fat digestion



Step 1
Rumen fluid

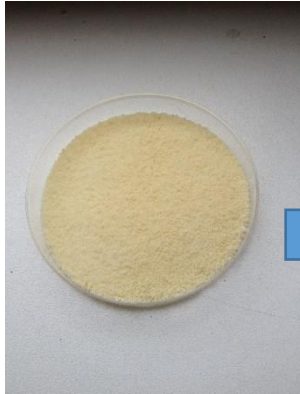


Step 2
Simulated
gastric fluid



Step 3
Simulated
intestinal fluid

Materials and Methods



Bypass fat
products



Nylon bags
Pore size 25 μm



Shaker 39°C
95 rpm



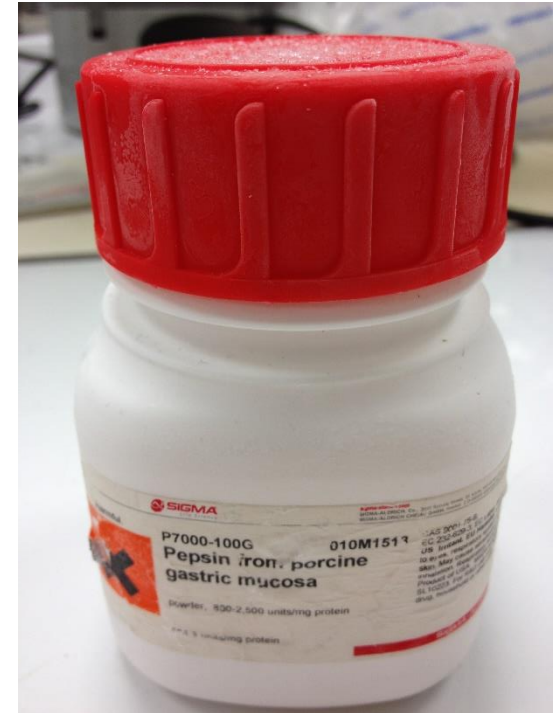
Freeze dryer



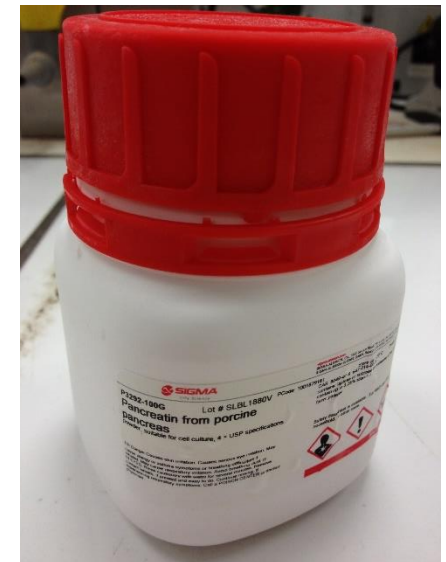
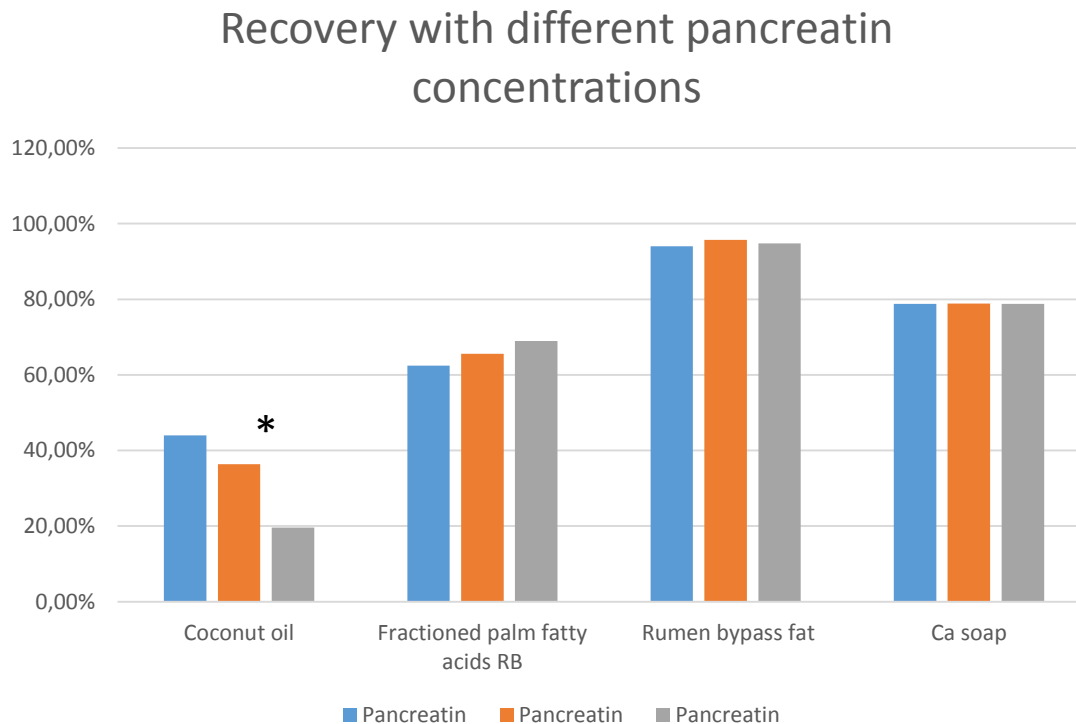
Soxhlet

Step 2: Pepsine 1g/L in 0.0125 M HCl pH 1.9

Sample	Sample weight, g	Pepsin + HCl	Freezdry recovery
Coconut oil	0,5	1 h	98,18%
Fractioned palm fatty acids RB	0,5	1 h	99,92%
Rumen bypass fat	0,5	1 h	99,74%
Ca soap	0,5	1 h	96,12%

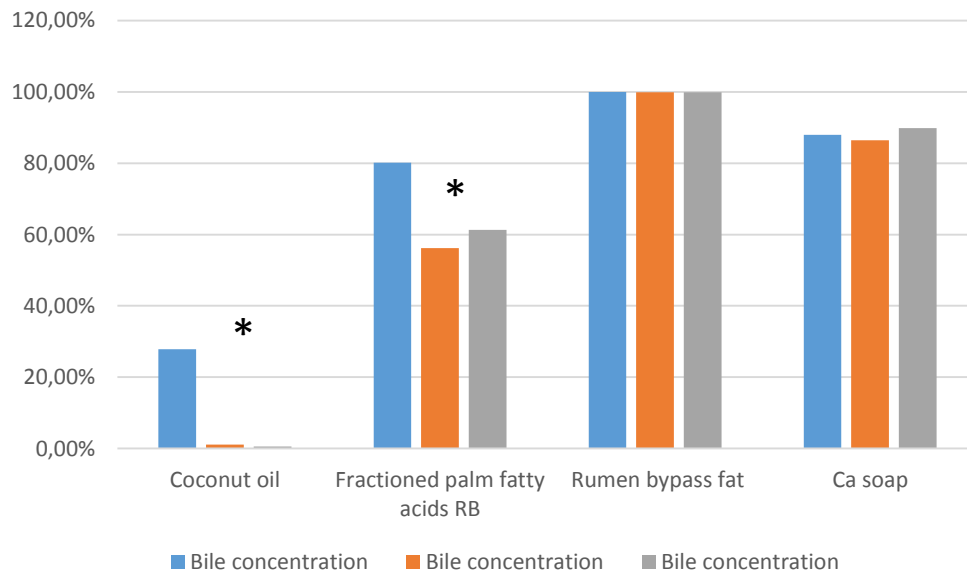


Sample recovery step 2 + step 3 with different pancreatin concentrations at pH 7.8 after 24h incubation

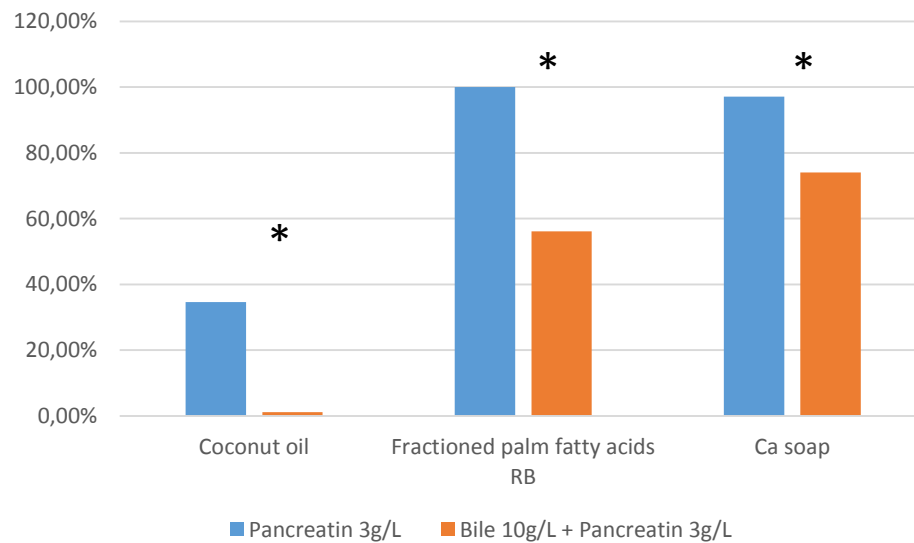


Sample recovery after step 2 + step 3 with different bile concentrations

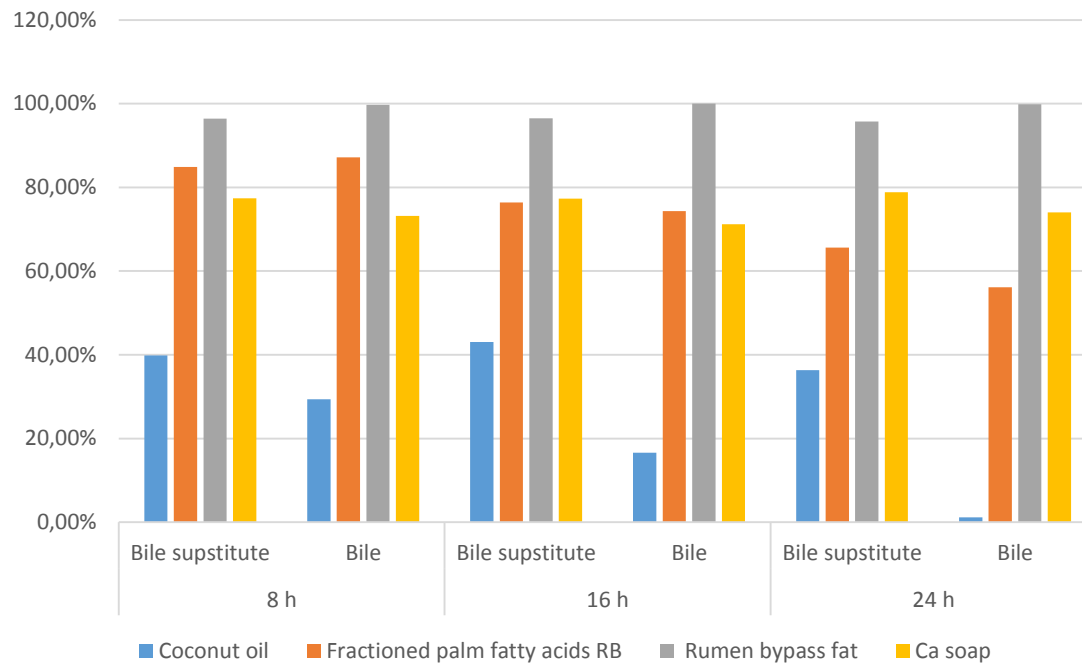
- Minimal 1,7 g/L of bile salts
- Amount of bile?



Sample recovery after step 2 + step 3 with or without Bovine bile



Recovery of samples at different time points, Bile substitute vs. Bovine bile



Phospholipid (lecithin)

Bile salt (taurocholate)



Three *in vitro* steps for digestion of fat

Step 1

Rumen fluid
Rumen fluid+
Artificial saliva+
Anaerobic
conditions
39°C
8h



Step 2

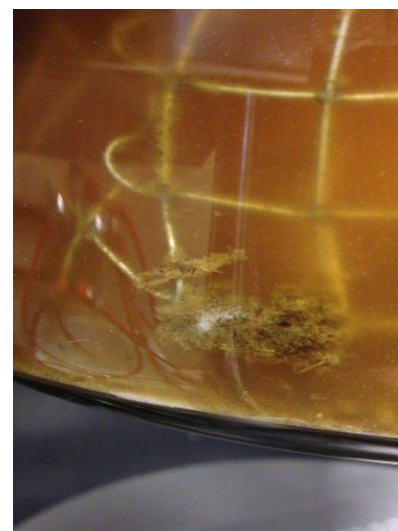
Simulated gastric
fluid
0.0125 M HCl
pH 1.9+
1 g/L Pepsin
39°C
1h



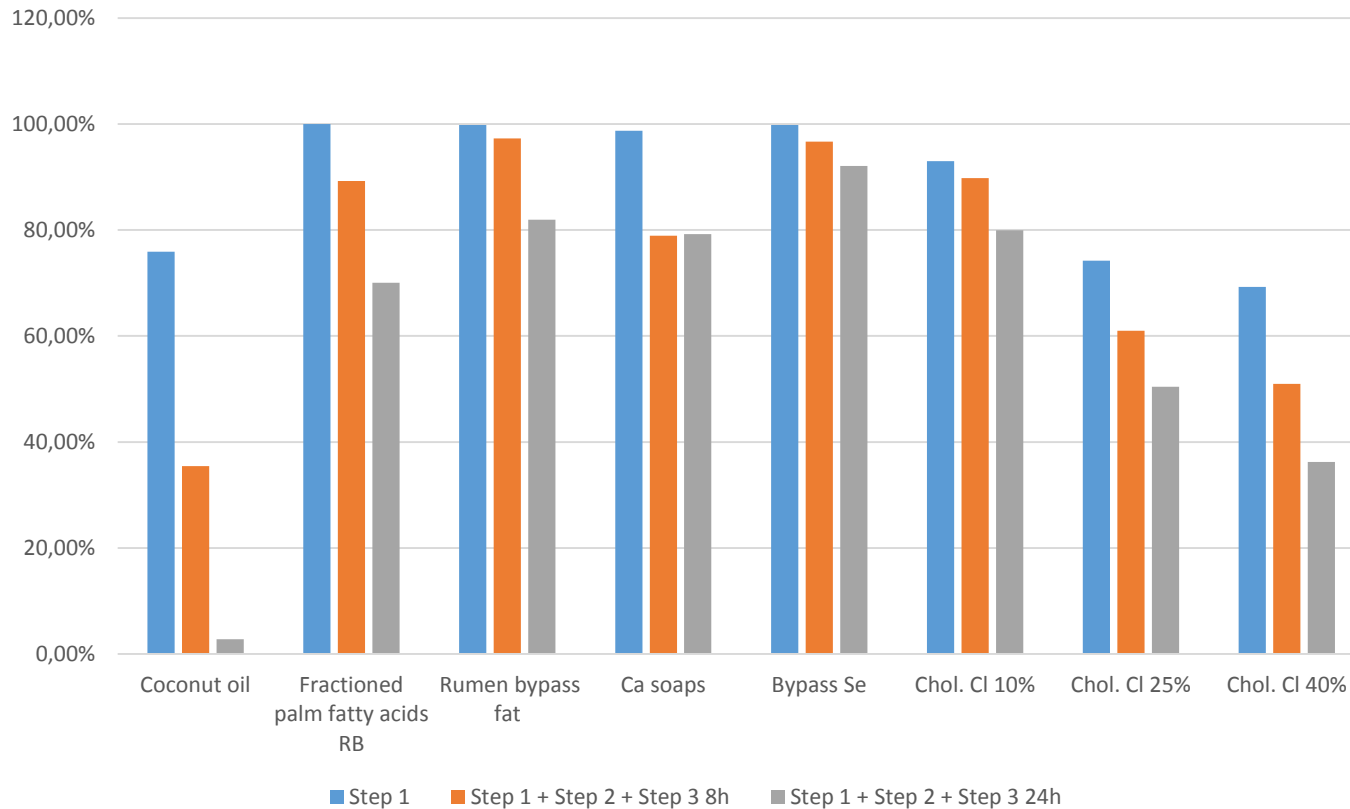
Step 3

Simulated
intestinal fluid
0.5 M phosphate
buffer
pH 7.8+
3 g/L Pancreatin+
10 g/L Bovine bile
39°C
24 h

Step 1 Rumen fluid incubation



Recovery of various fat products after in vitro digestion with simulated rumen, gastric and intestinal fluid



Comparison of various fat products recovery after *in vivo* and *in vitro* digestion

Product	Rumen phase		Gastric + Intestinal phase		
	<i>in vivo</i> 8h	<i>in vitro</i> 8h	<i>in vivo</i> 8-12h	<i>in vitro</i> Step3 8h	<i>in vitro</i> Step 3 24h
Chol. Cl 10%	89.9 %	92.97%	95.12%	96.05%	85.08%
Chol. Cl 25%	83.1 %	74.23%	53.79%	78.61%	65.63%
Chol. Cl 40%	61.0 %	69.27%	71.2%	68.87%	50.72%
Bypass Se	100.0 %	99.84%	95.07%	96.34%	91.74%

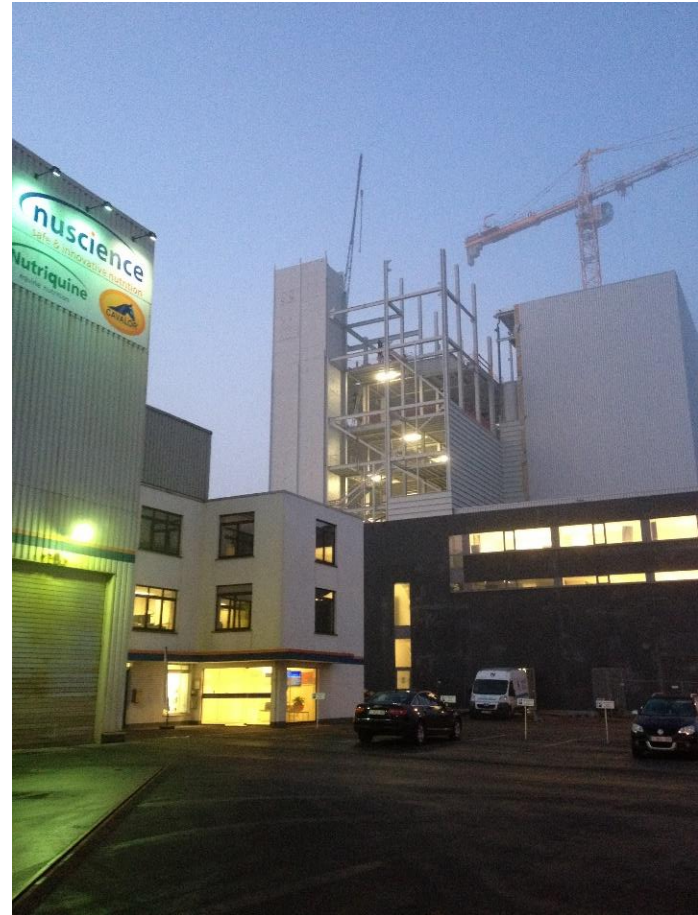
Conclusions

- we have successfully upgraded *in vitro* model of protein digestion and made it more appropriate for determination of fat digestion.
- this model works well with rumen bypass fat products
- it should be improved if natural feeds rich in fat are to be used

STSM timespan 6 weeks.



January 16th 2015



February 27th 2015



Site Melle

Biocentrum Agri-Vet

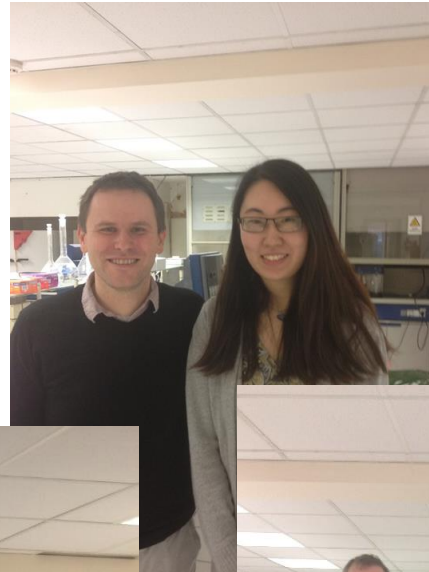
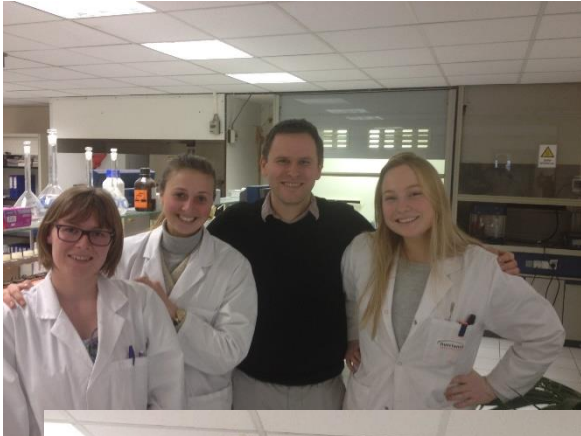


Vakgroep Dierlijke
Productie
(FBW)

Vakgroep Plantaardige
Productie
(FBW)



Nutrition Sciences N.V. - lab





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- Research was funded by Nutrition Sciences N.V.



Thank you for your attention.