UMR 7282

Enzymologie Interfaciale et Physiologie de la Lipolyse

Enzymology at Interfaces and Physiology of Lipolysis

Production of digestive lipases for *in vitro* digestion models

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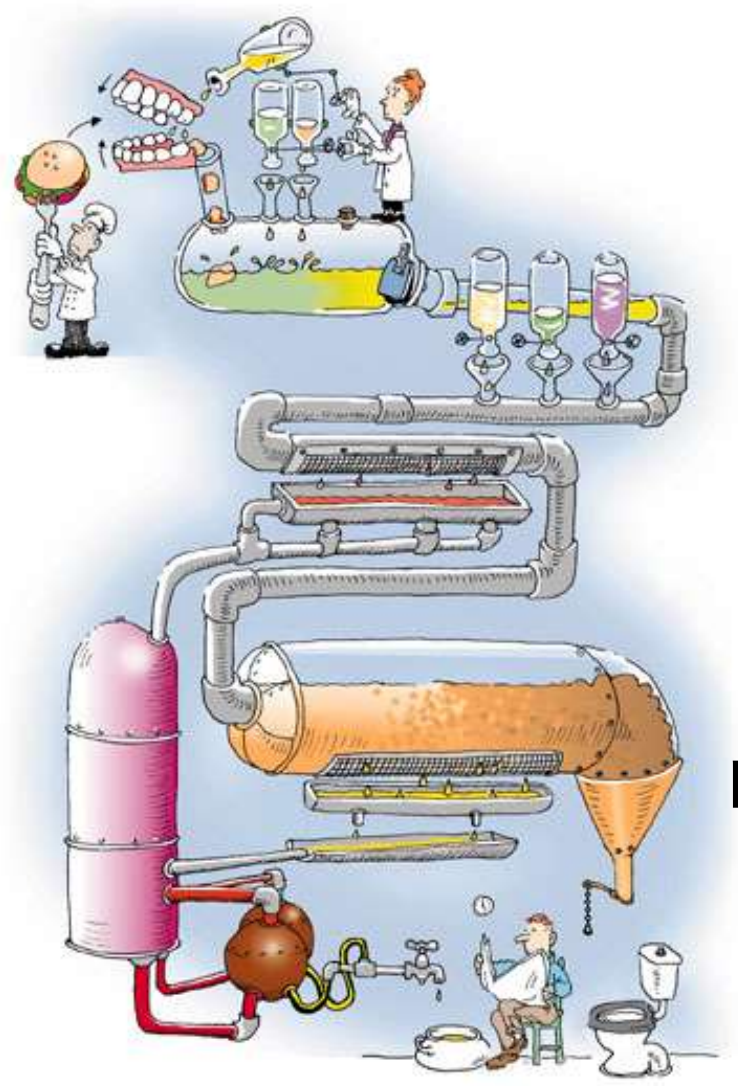
In vitro digestion models: why ?



Useful tools for studying:

Digestibility

The fraction of a substance that is mechanically and chemically degraded into smaller components

Bio-accessibility

The fraction of a substance that is available for digestion and intestinal absorption

Bio-availability

The quantity or fraction of the ingested substance that has reached the systemic circulation or target organ

of foods without, or before, performing animal and human studies (cost and ethical constraints)

To establish *in vitro-in vivo* correlations

In vitro digestion models: domains of application

Food

Ex: EU COST Infogest

Food & Function (2014) 5(6):1113-1124.

PAPER

A standardised static *in vitro* digestion method suitable for food – an international consensus†

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Pharma

Ex: LFCS consortium

JOURNAL OF PHARMACEUTICAL SCIENCES (2012) 101(9):3360-3380 DOI 10.1002/jps

RESEARCH ARTICLE

Toward the Establishment of Standardized *In Vitro* Tests for Lipid-Based Formulations, Part 1: Method Parameterization and Comparison of *In Vitro* Digestion Profiles Across a Range of Representative Formulations

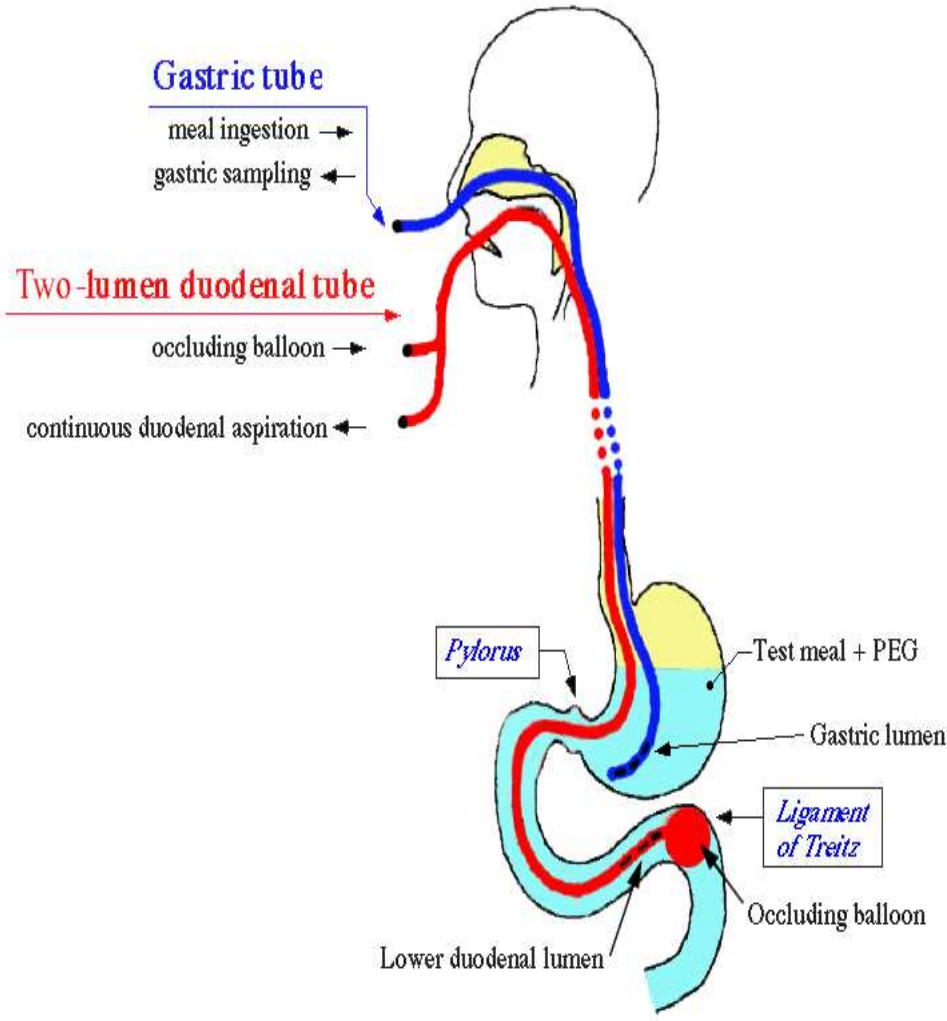
HYWEL D. WILLIAMS,[1] PHILIP SASSENE,[2] KAREN KLEBERG,[2] JEAN-CLAUDE BAKALA-N'GOMA,[3] MARILYN CALDERONE,[4] VINCENT JANNIN,[5] ANNABEL IGONIN,[6] ANETTE PARTHEIL,[7] DELPHINE MARCHAUD,[5] EDUARDO JULE,[6] JAN VERTOMMEN,[6] MARIO MAIO,[7] ROSS BLUNDELL,[4] HASSAN BENAMEUR,[6] FRÉDÉRIC CARRIÈRE,[3] ANETTE MÜLLERTZ,[2] CHRISTOPHER J. H. PORTER,[1] COLIN W. POUTON[8]

Oral lipid-based formulations of lipophilic drug, screening of lipase inhibitors,…

In vitro digestion models are based on data collected during studies of test meal digestion in humans

Quantitative studies of digestive lipase secretion and contribution to the lipolysis of dietary triglycerides

Carrière et al. *Gastroenterology* (**1993**) 105:876–888
Carrière et al. *Am. J. Physiol.* (**2001**) 281:G16-G28
Carrière et al. *Clin. Gastroenterol. Hepatol.* (**2005**) 3:28-38



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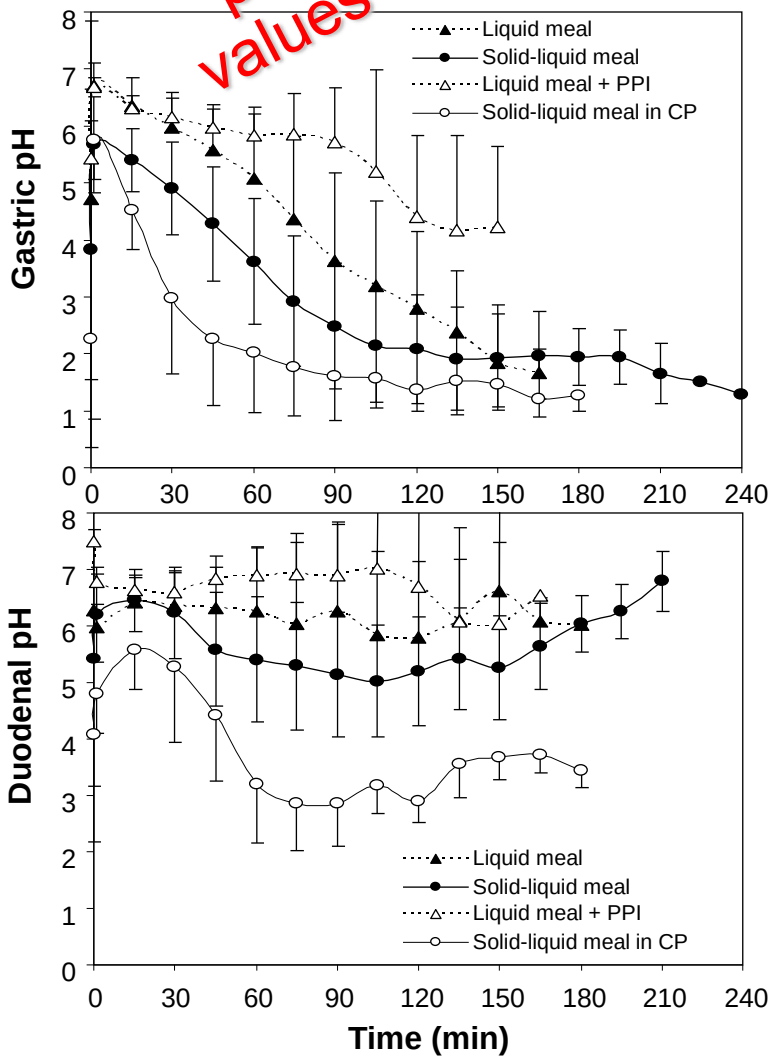
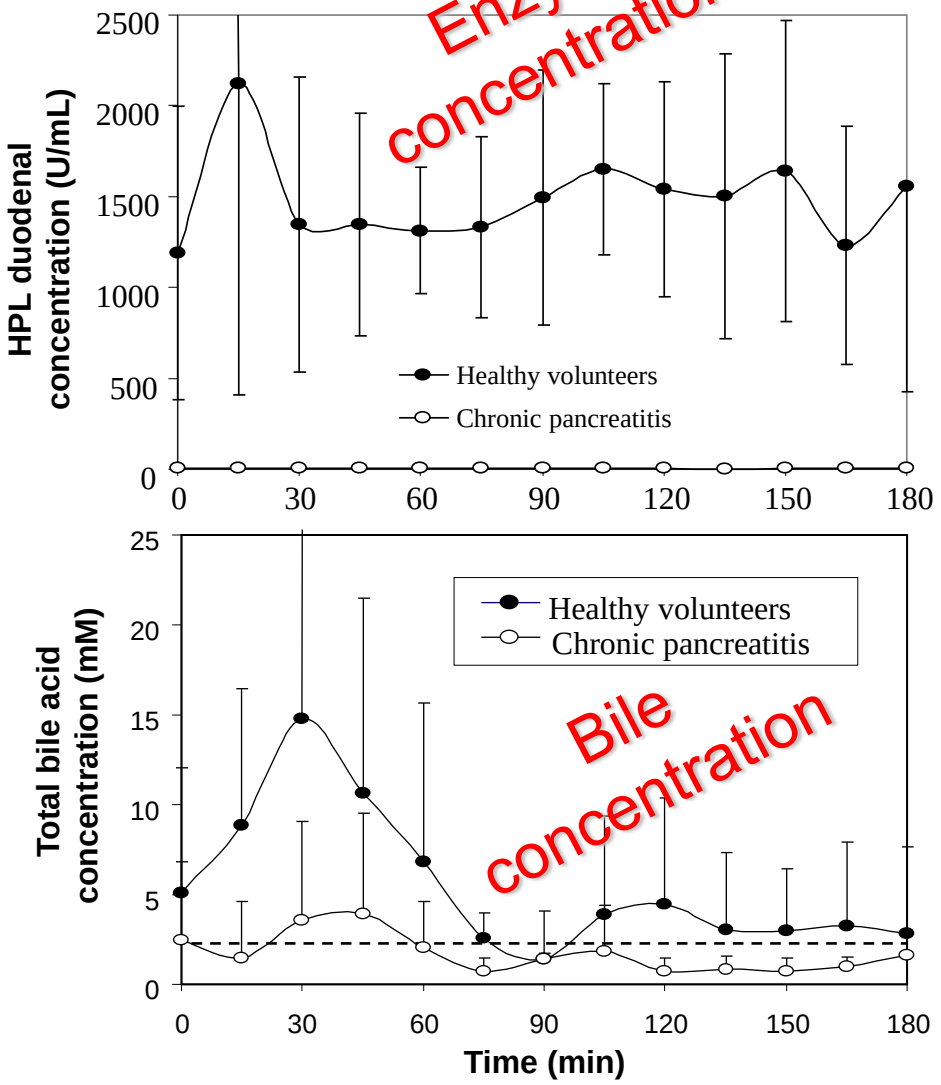
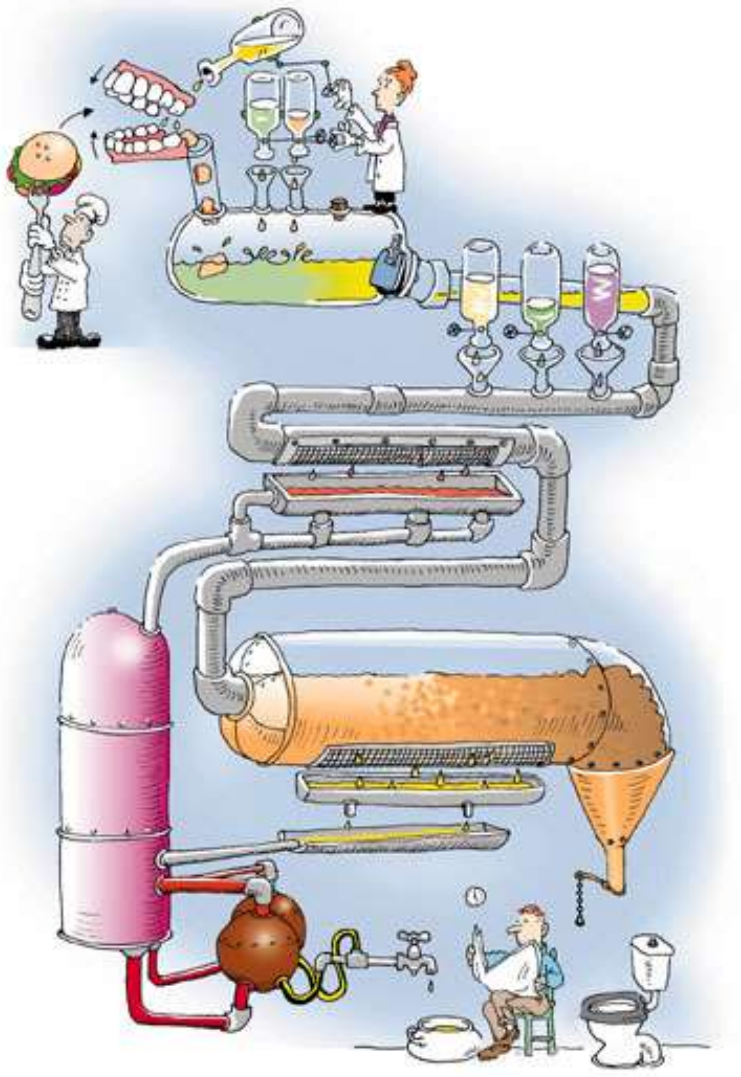




In vitro digestion models: various types

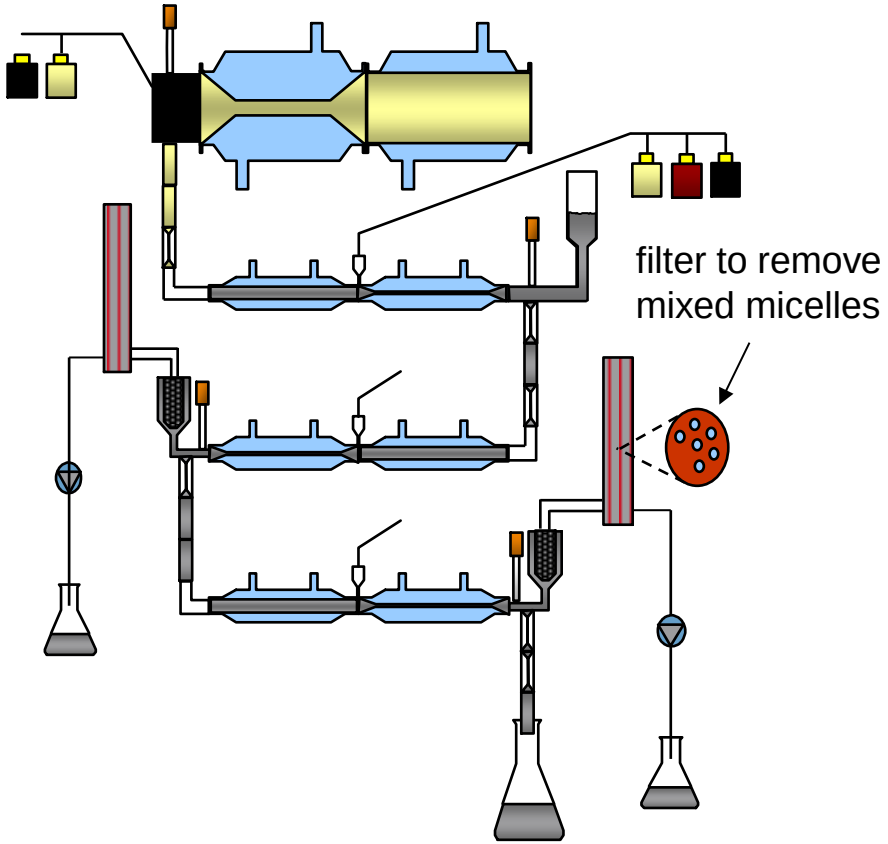


Dynamic vs. Static

- Simple static
  (ex: pH stat)
- Gastric models
- Two compartmental
  (ex: two-step static model with gastric and intestinal phases)
- Multi compartmental
  (ex: oral, gastric, intestinal and colonic phases)

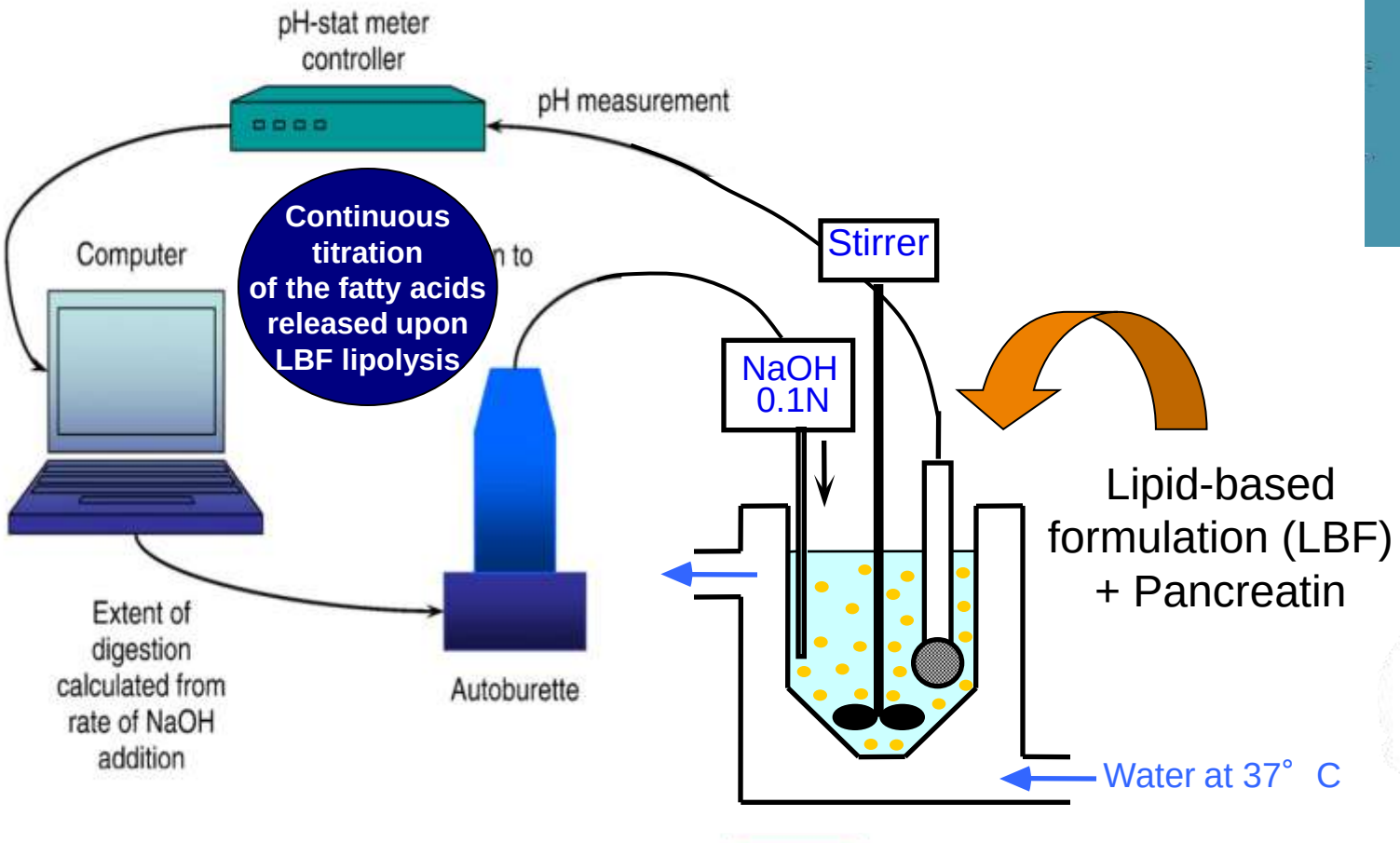
Dynamic *in vitro* digestion models

TIM gastro-small intestinal model (TNO)



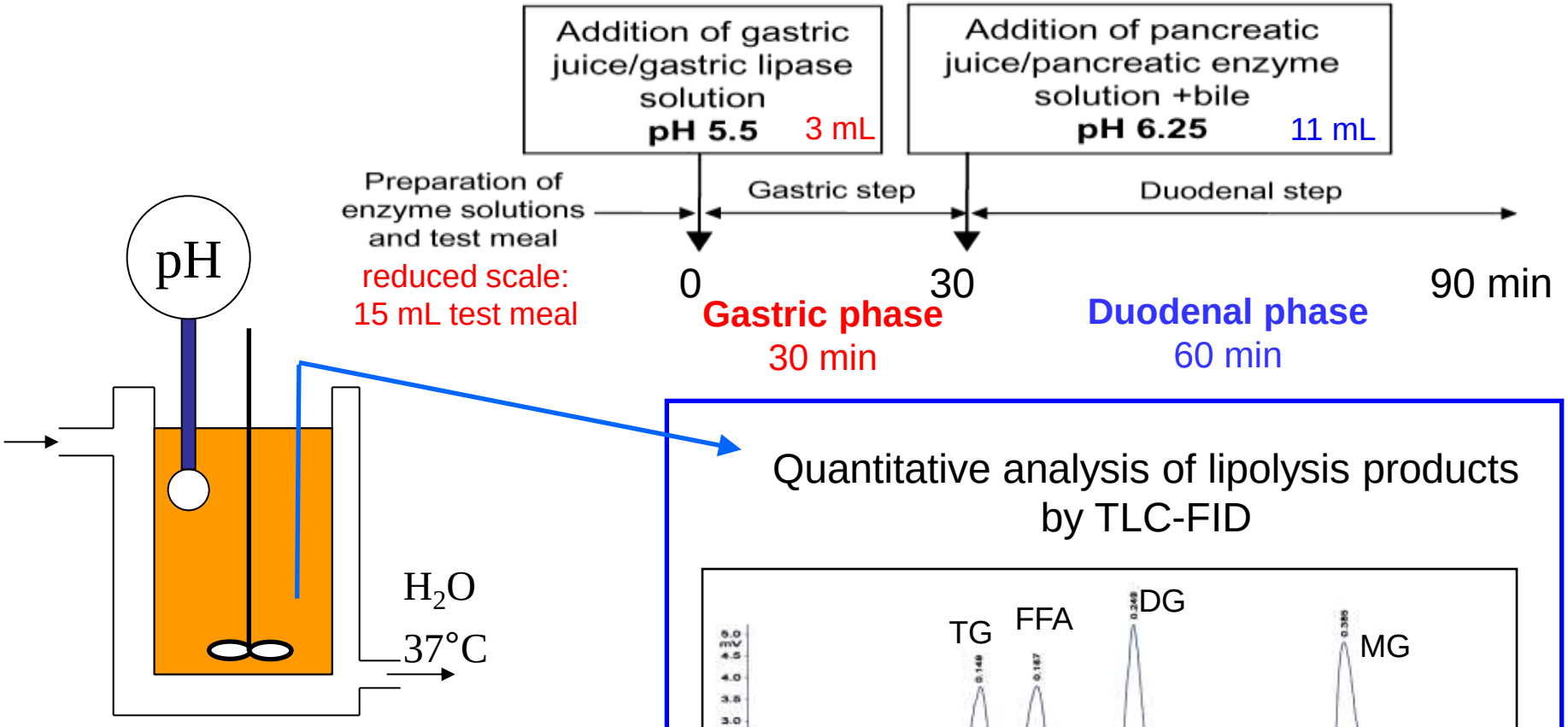
Slide kindly provided by Mans Minekus, TNO

The *in vitro* model chosen by the LFCS consortium: One-step static / intestinal digestion using a pHstat equipement



Williams et al. *J. Pharm. Sci.* (**2012**) 101(9):3360-3380

In vitro simulation of gastrointestinal lipolysis using a two-step static in vitro digestion model



Quantitative analysis of lipolysis products by TLC-FID

Lipolysis level (%)

Carrière et al. Gastroenterology (1993) 105:876–888
Carrière et al. Am. J. Physiol. (2001) 281:G16-G28
Capolino et al., *Food Digestion* (2011) 2:43–51

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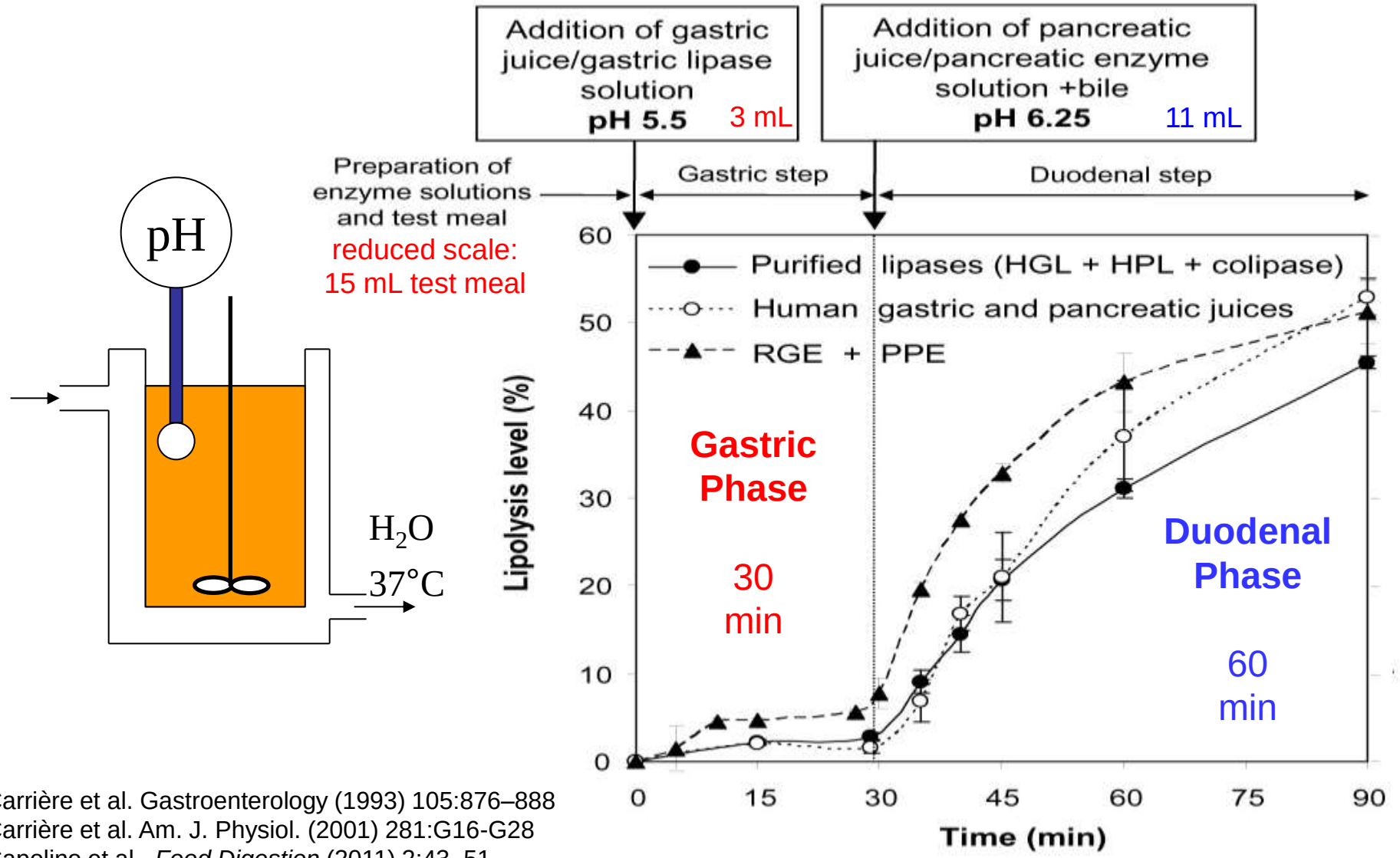

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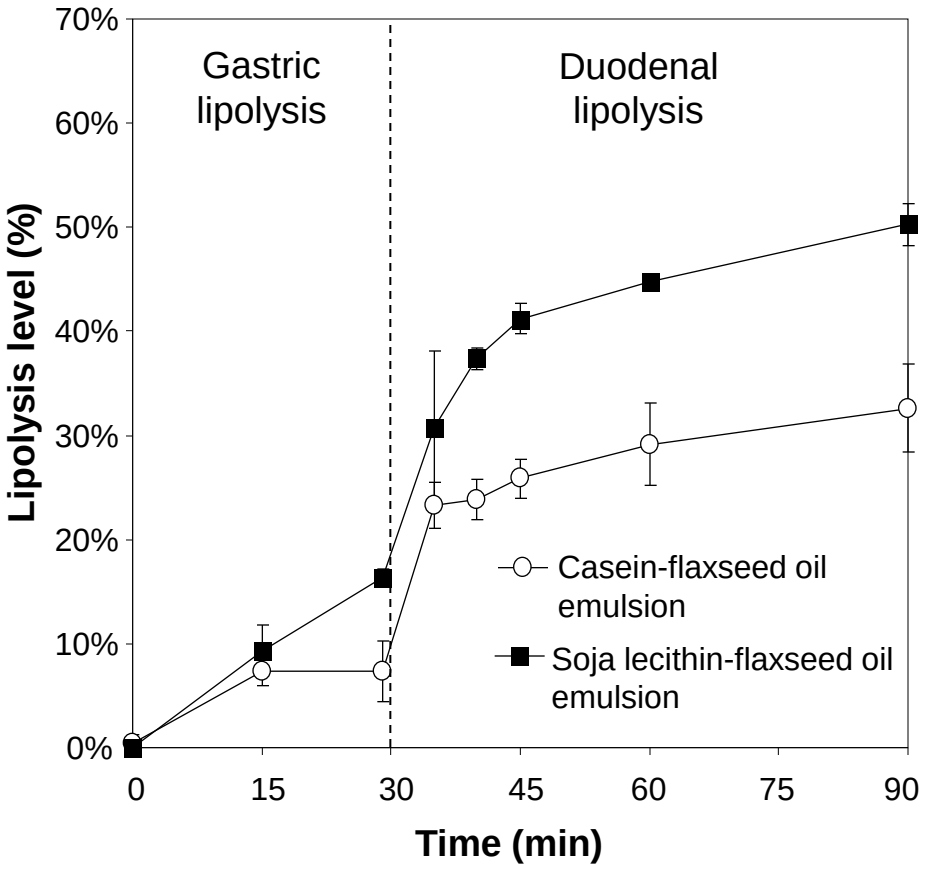
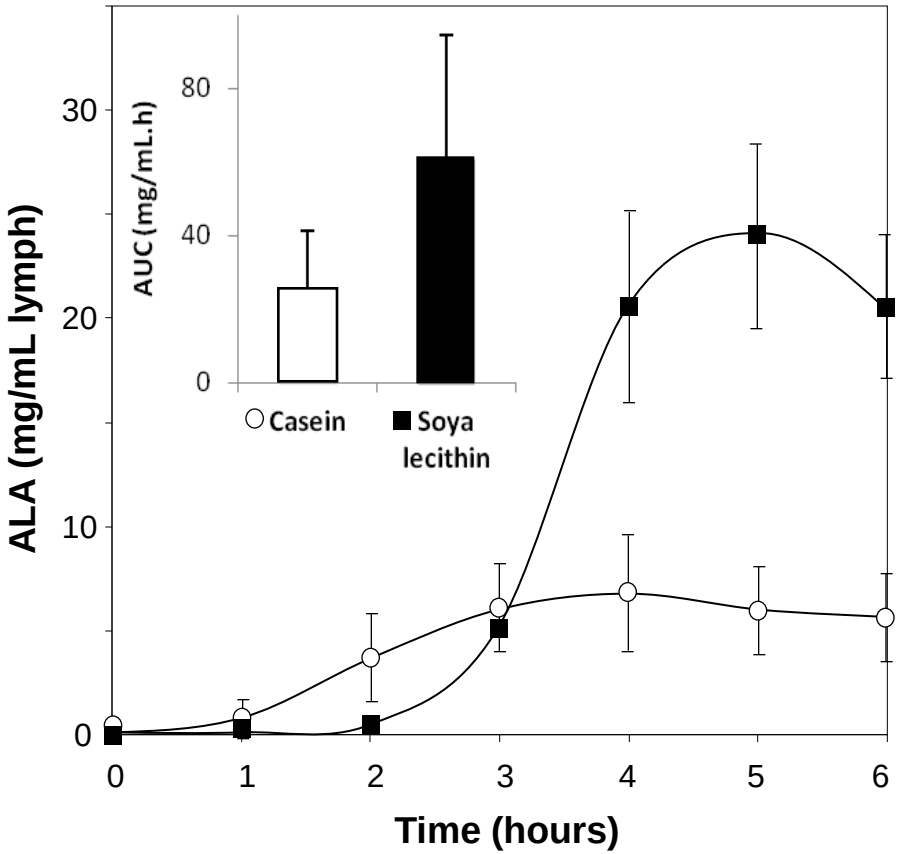
In vitro – *in vivo* correlations

In vitro digestion of flaxseed oil



Absorption in rats of α-linolenic acid (ALA) from flaxseed oil



Couëdelo et al. *Food & Function* (**2015**) 6:1726-1735

Sources of enzymes for *in vitro* digestion

- Human gastric and pancreatic juices

  Carrière et al. *Gastroenterology* (**2000**) 119:949–960

- Native lipases (HGL, HPL) purified from human gastric and pancreatic juices

  Carrière et al. *Gastroenterology* (**2000**) 119:949–960

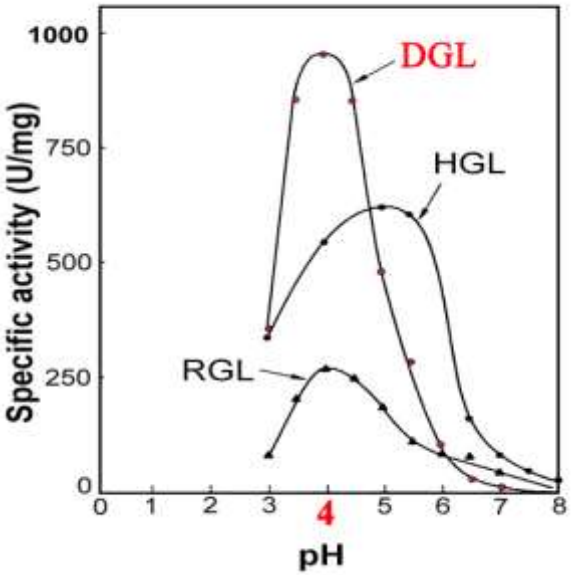
- Recombinant human lipases

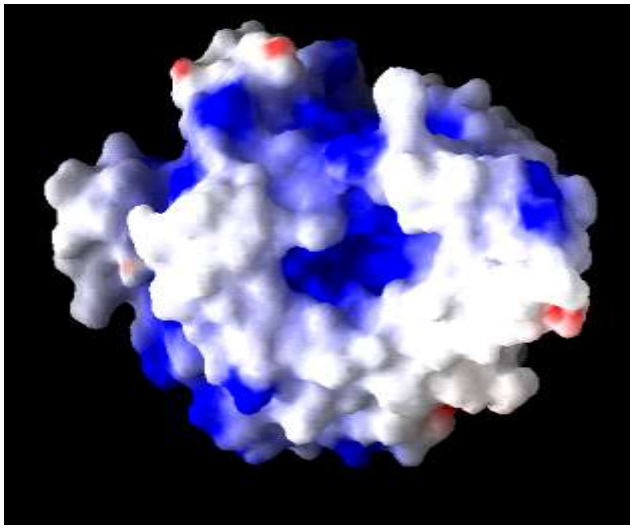
  rHPL in insect cells (Thirstrup et al. *FEBS Letters* (**1993**) 327:79-84)
  rHPL in *Pichia pastoris* (Belle et al. *Biochemistry* (**2007**) 46:2205-2214)

  rHGL in *yeast* (Bodmer et al. *BBA* (**1981**) 909:231-244)
  rHGL in insect cells (Canaan et al. *Protein Expression & Purification* (**1998**) 14:23–30

Sources of enzymes for *in vitro* digestion

Production of recombinant dog gastric lipase (r-DGL)
in transgenic corn (Meristem Therapeutics)





Sources of enzymes for *in vitro* digestion

- Native enzymes from animal sources

Capolino et al., *Food Digestion* (**2011**) 2:43–51

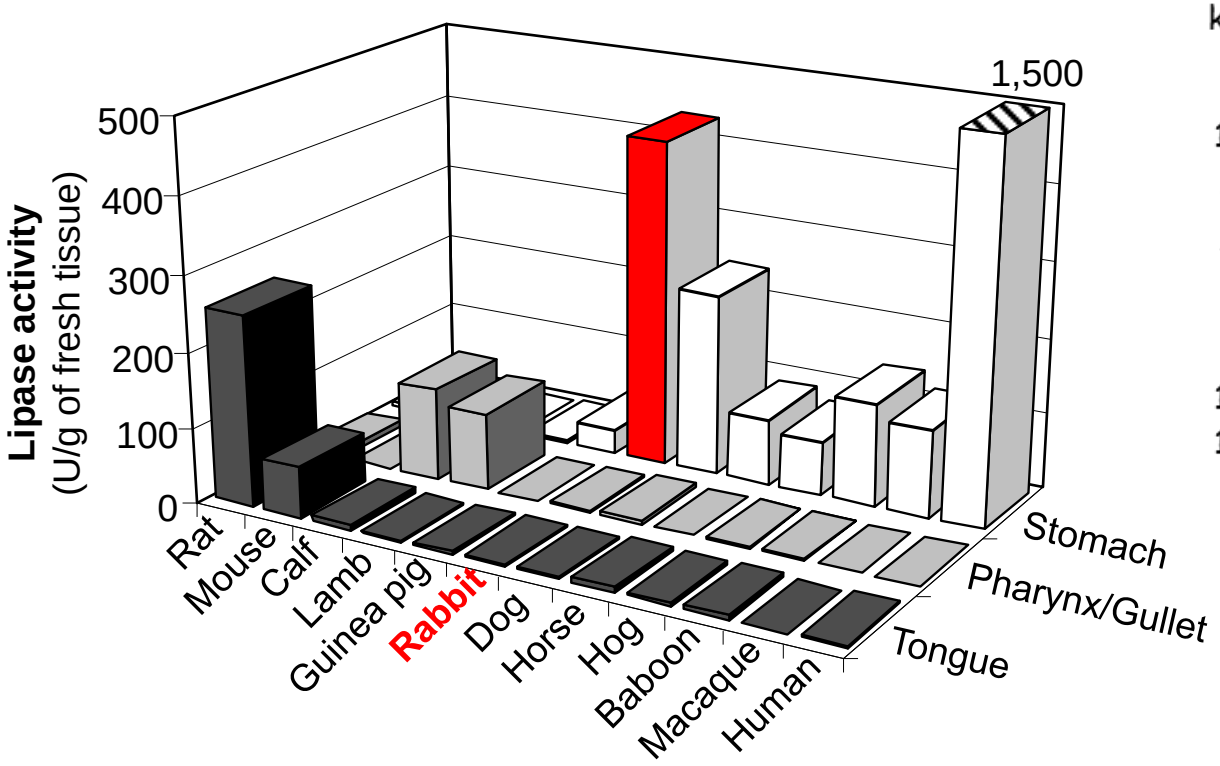
Porcine pancreatic extracts

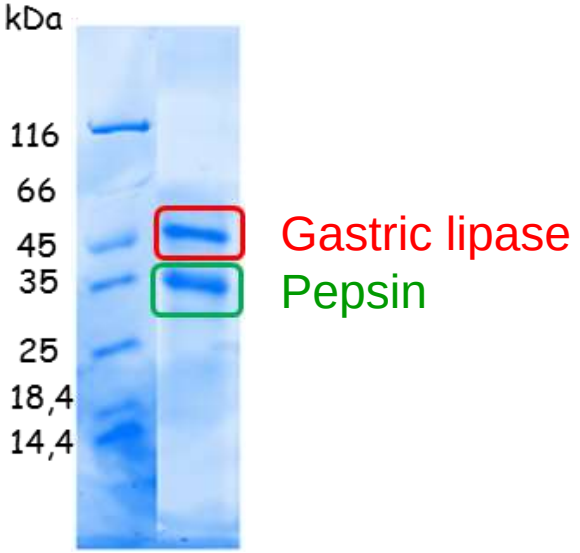
Sources of enzymes for *in vitro* digestion

- Native enzymes from animal sources

Capolino et al., *Food Digestion* (**2011**) 2:43–51

Rabbit gastric extracts





Aknowledgements

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