Methods to assess effects of prebiotics in humans

Lieselotte Cloetens[1,2], Willem F. Broekaert[3,4], Björn Åkesson[1], Gunilla Önning[1], Jan A. Delcour[3], Kristin Verbeke[2]

[1]Biomedical Nutrition, Pure and Applied Biochemistry, Lund University, Lund, Sweden
[2]Laboratory of Digestion and Absorption, TARGID, Catholic University of Leuven, Leuven, Belgium
[3]Laboratory of Food Chemistry and Biochemistry and Leuven Food Science and Nutrition Research Center (LFoRCe), Catholic University of Leuven, Leuven, Belgium
[4]Fugeia NV, Leuven, Belgium

Prebiotics

non-digestible food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon, and thus improve host health

(Gibson et al., Nutr Res Rev 2004;17:259-275)

Health-promoting bacteria: bifidobacteria and lactobacilli

Health-related effects:

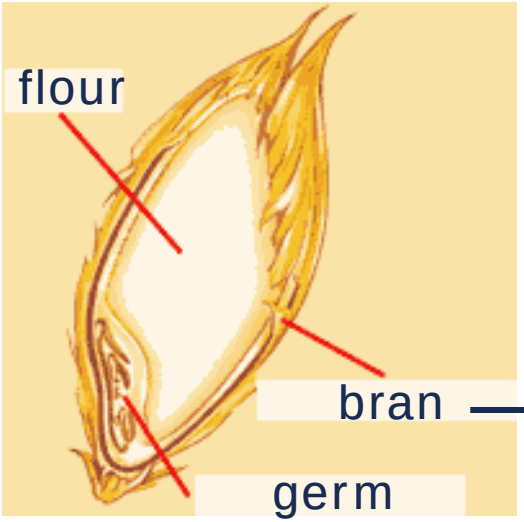
- effects on gastrointestinal transit
- improvement of mineral absorption
- lipid-lowering effects
- reduced risk of colon cancer
- reduced protein fermentation

Prebiotics

Fructo-oligosaccharides (FOS) and inulin: well-known prebiotics

Arabinoxylan-oligosaccharides (AXOS): prebiotic potential?

= degradation products of arabinoxylan

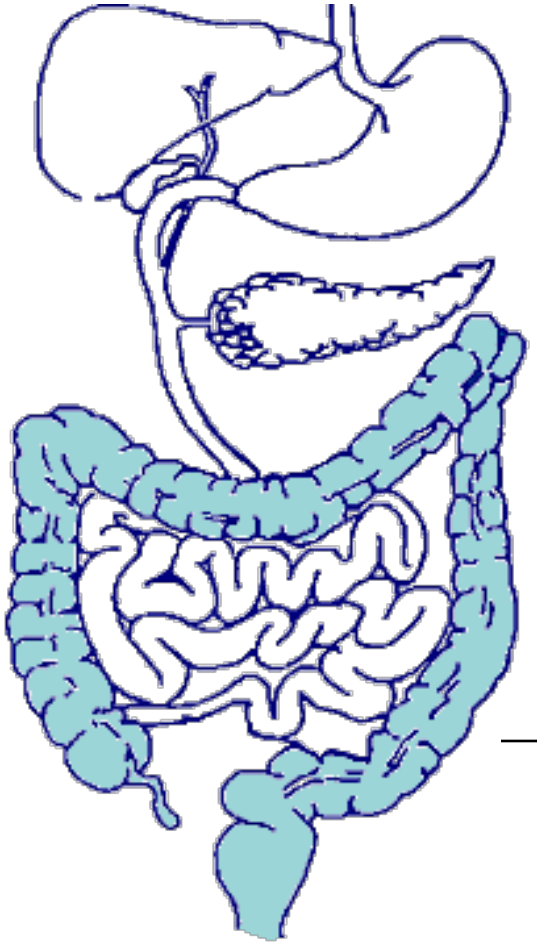


outer layer of a cereal:
20-25% arabinoxylans

Methods

- Gastrointestinal parameters
  - Motility and digestion
- Prebiotic effects
  - Microbial composition
  - Metabolic activity of the microbiota

Motility and digestion



dietary intake and digestive processes

substrate availability

microbial composition

colonic transit time

colonic fermentation metabolites

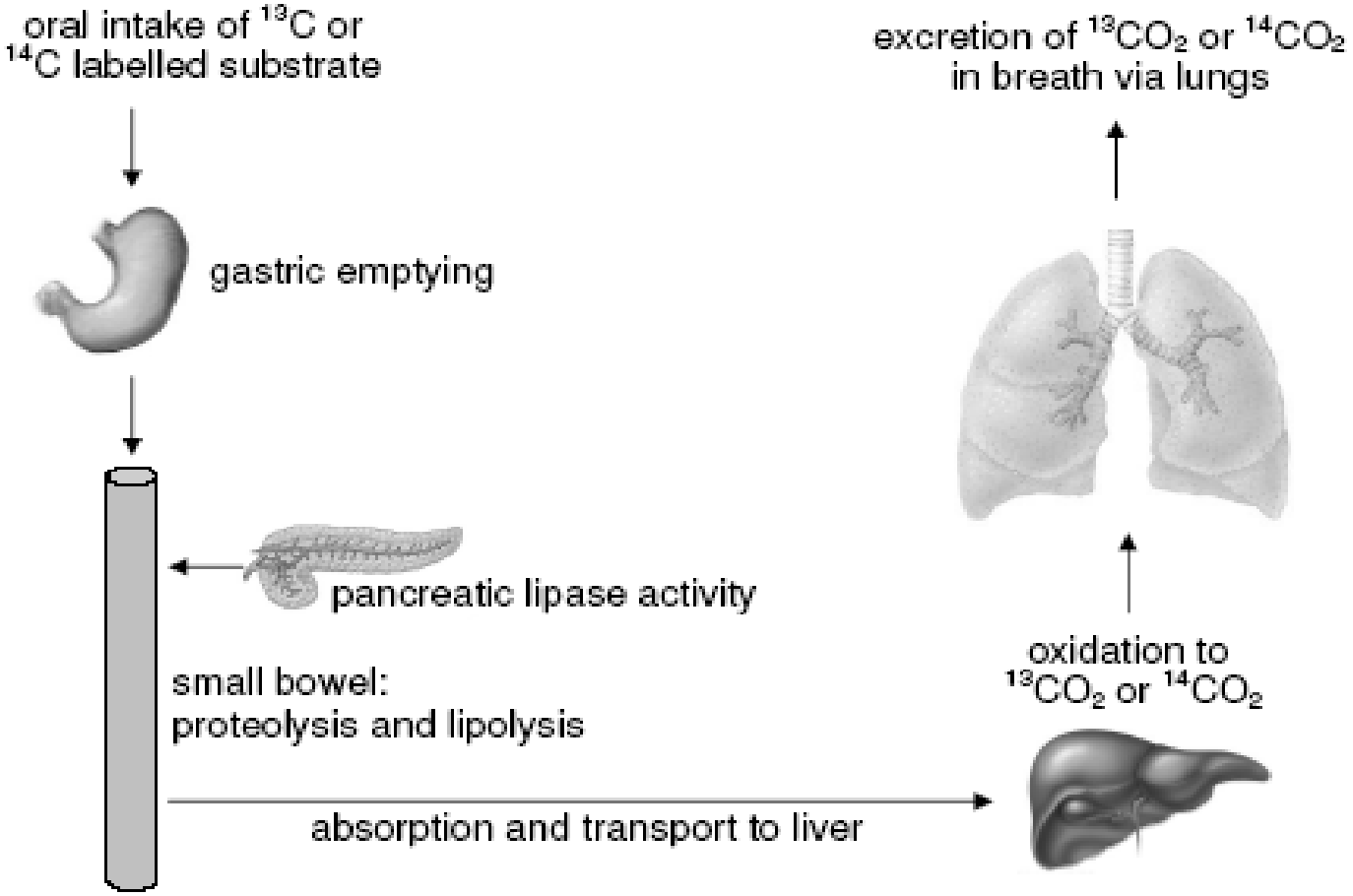
Cloetens et al., e-SPEN 2008

Digestive processes

- In the proximal GI-tract:
  - gastric emptying rate (GE)
  - lipid digestion
  - protein digestion
  - oro-caecal transit time (OCTT)
  - (total gastrointestinal transit)

  - prebiotics can alter digestion and absorption of other nutrients: **physicochemical properties** (viscosity, water-holding capacity and osmolarity)

Breath tests



GI parameters

parameter	labelled substrate	dose	sample collection
gastric emptying[1]	[^{13}C]-octanoic acid or [^{14}C]-sodium octanoate	91mg 74kBq	4h
oro-caecal transit time[2,3]	lactose-[^{13}C]-ureide or inulin-[^{14}C]-carboxylic acid	500mg 74kBq	10h
protein digestion[4]	egg protein intrinsically labelled with [^{13}C]-leucine	200mg	6h
lipid digestion[5]	[^{13}C]-mixed triglyceride	250mg	6h

[1] Ghoos et al. Gastroenterology 1993
[2] Geypens et al. J Nucl Med 1999
[3] Verbeke et al. Aliment Pharmacol Ther 2005
[4] Evenepoel et al. J Nutr 1997
[5] Vantrappen et al. Gastroenterology 1989

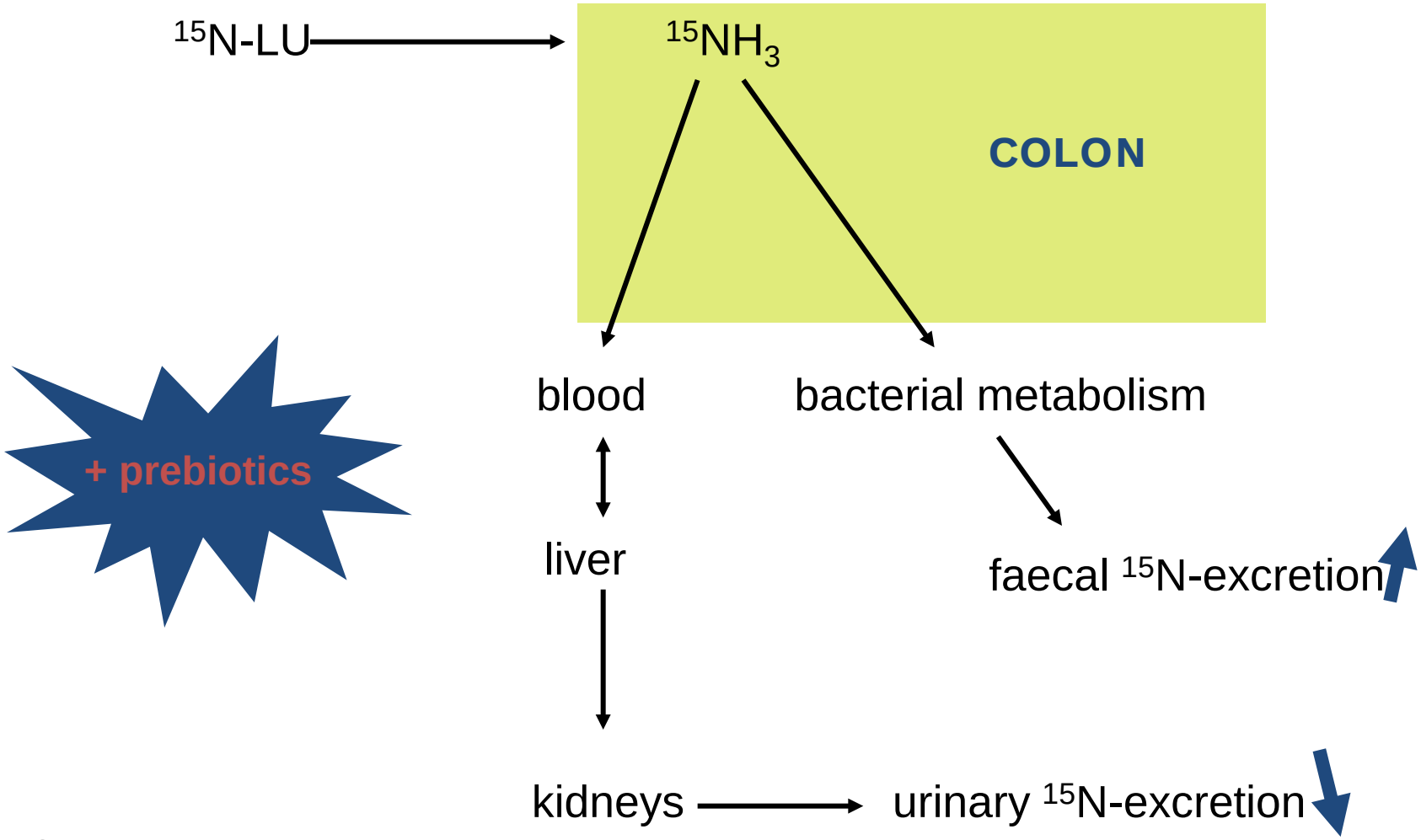
Methods

- Gastrointestinal parameters
  - Motility and digestion
- Prebiotic effects
  - Microbial composition
  - Metabolic activity of the microbiota

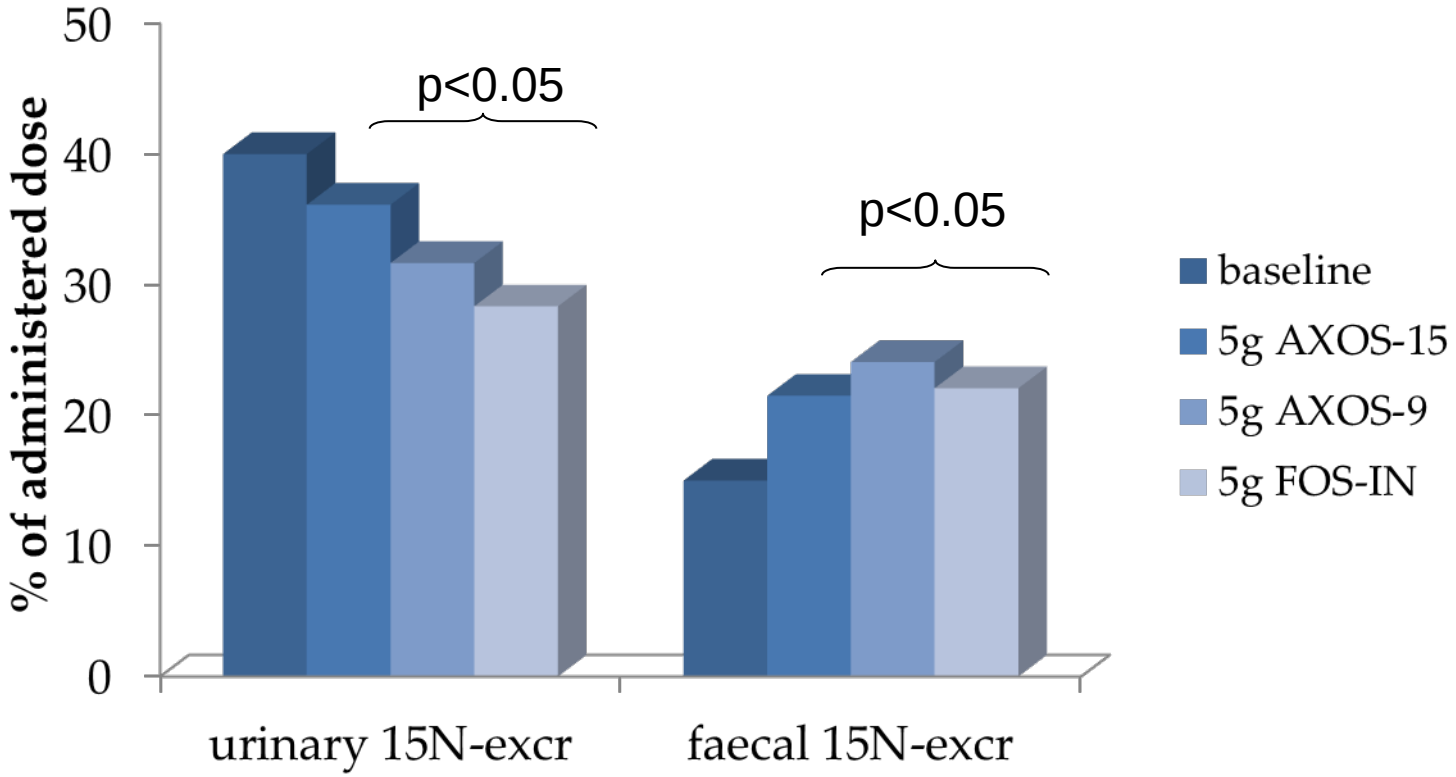
Metabolic activity of microbiota

- **Lactose-[^{15}N,^{15}N']-ureide** to measure colonic **ammonia** metabolism

Lactose-[^{15}N,^{15}N']-ureide



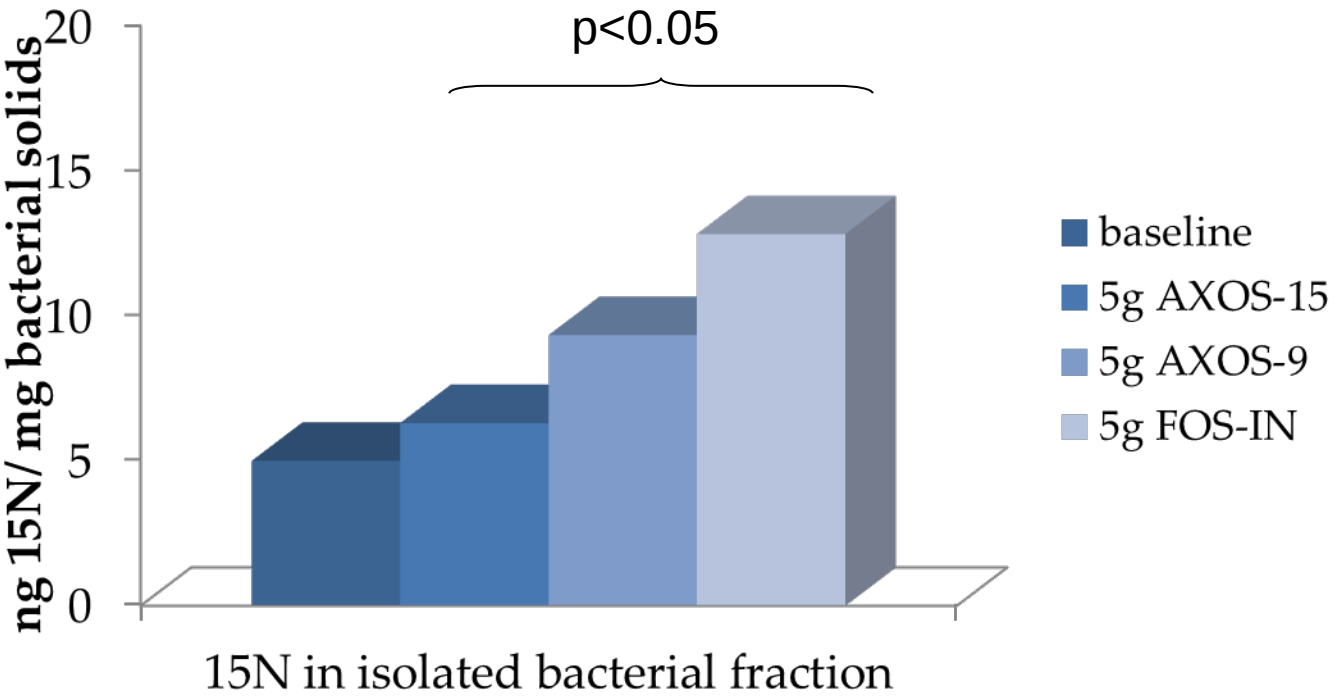
Urinary and faecal ^{15}N-excretion



significant shift from urinary to faecal ^{15}N-excretion

Cloetens et al., Am Coll Nutr 2008
Cloetens et al., Br J Nutr 2010

^{15}N-excretion in bacterial fraction



significant increased uptake of nitrogen by bacteria due to stimulation of bacterial growth/activity

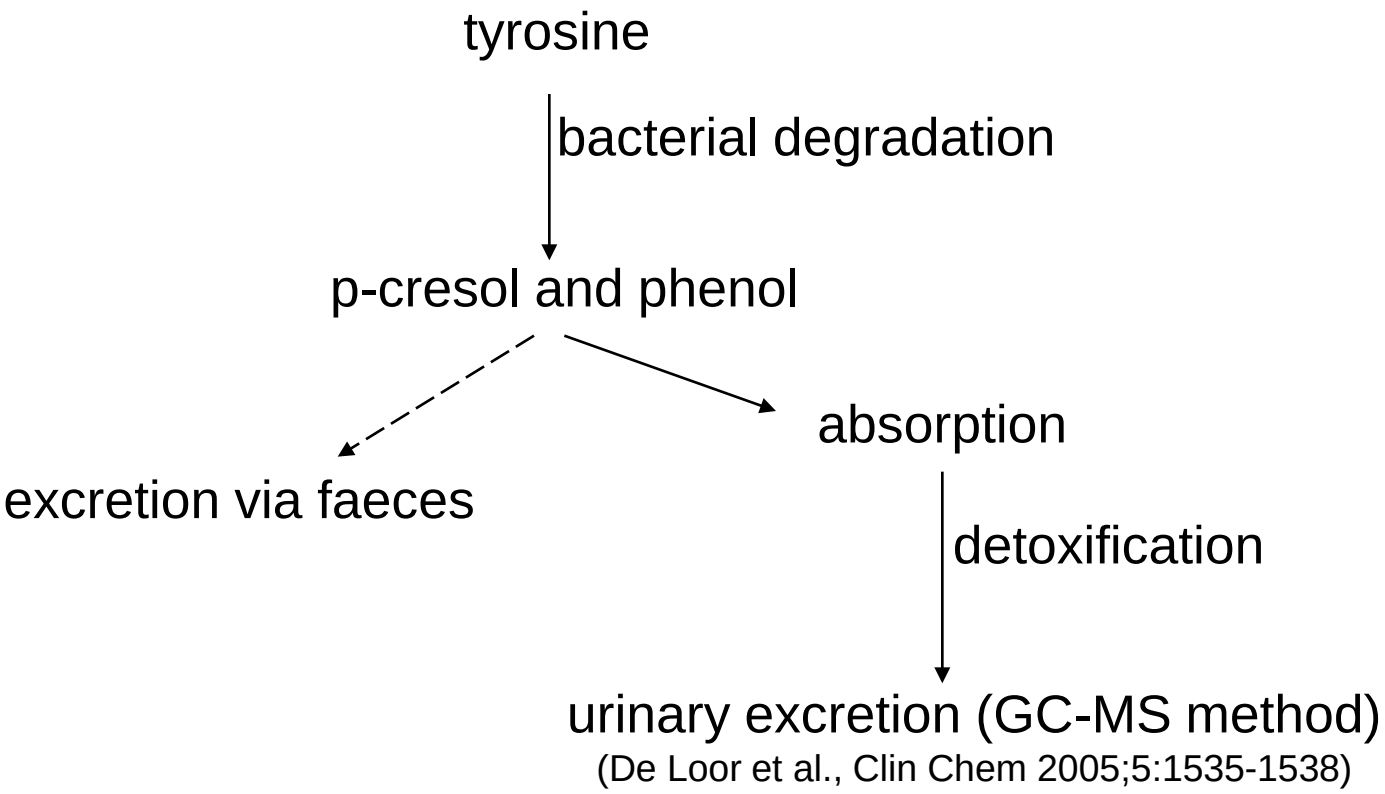
Cloetens et al., Am Coll Nutr 2008
Cloetens et al., Br J Nutr 2010

Metabolic activity of microbiota

- **Lactose-[^{15}N,^{15}N']-ureide** to measure metabolic **ammonia** metabolism

- **P-cresol and phenol** to measure **protein** fermentation

Phenolic compounds



urinary excretion of p-cresol and phenol is a measure of the extent of colonic protein fermentation

Phenolic compounds

- Prebiotics: decreased proteolytic activity, less p-cresol and phenol excretion
  - increased carbohydrate fermentation = saccharolytic activity
  - increased uptake and assimilation of nitrogen
  - decreased colonic pH
  - reduced protease activity

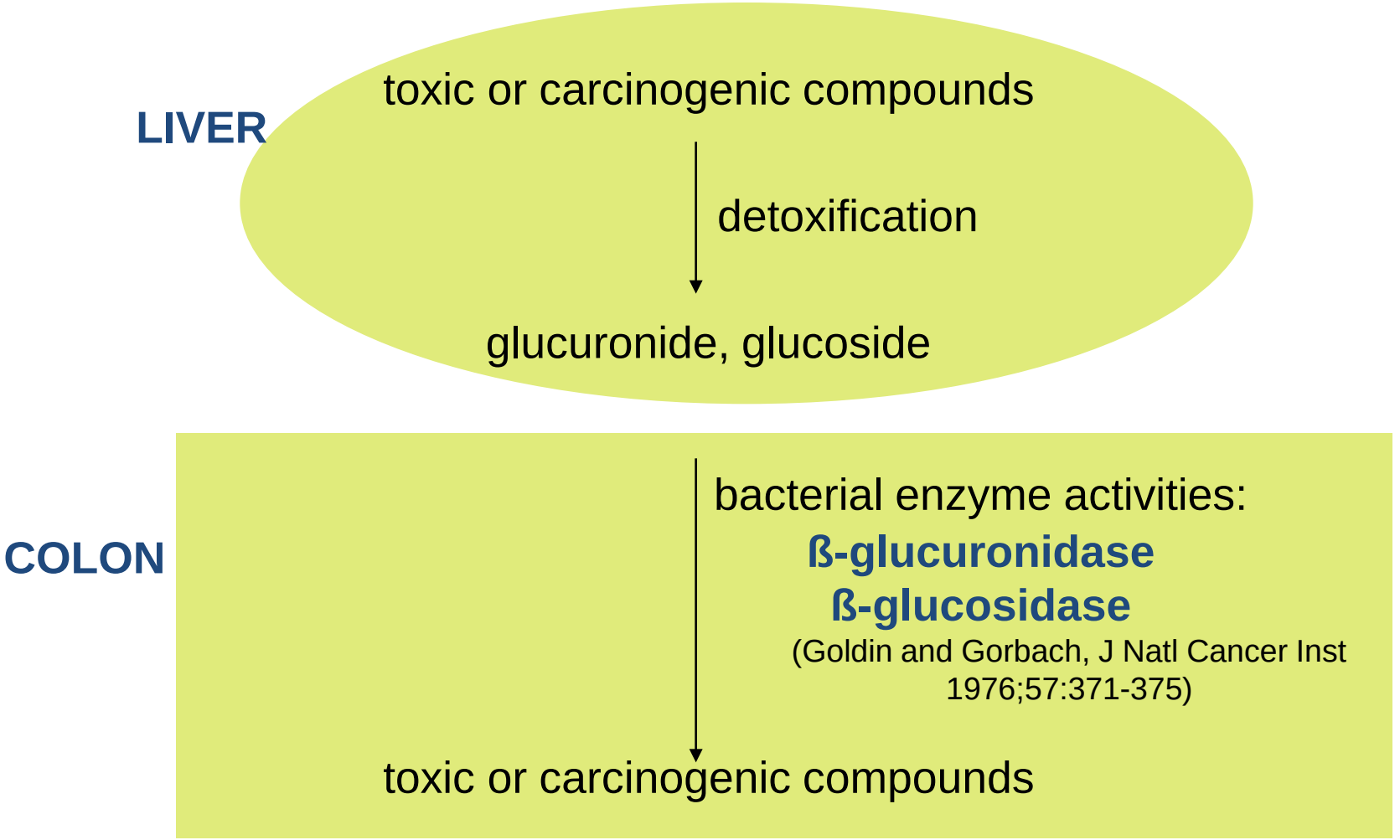
- Results
  - significantly decreased amount of p-cresol after 10g AXOS

Cloetens et al., Br J Nutr 2010

Metabolic activity of microbiota

- **Lactose-[^{15}N,^{15}N']-ureide** to measure metabolic **ammonia** metabolism
- **P-cresol and phenol** to measure **protein** fermentation
- **Bacterial enzyme activities:** β-glucuronidase and β-glucosidase

Bacterial enzymatic activities

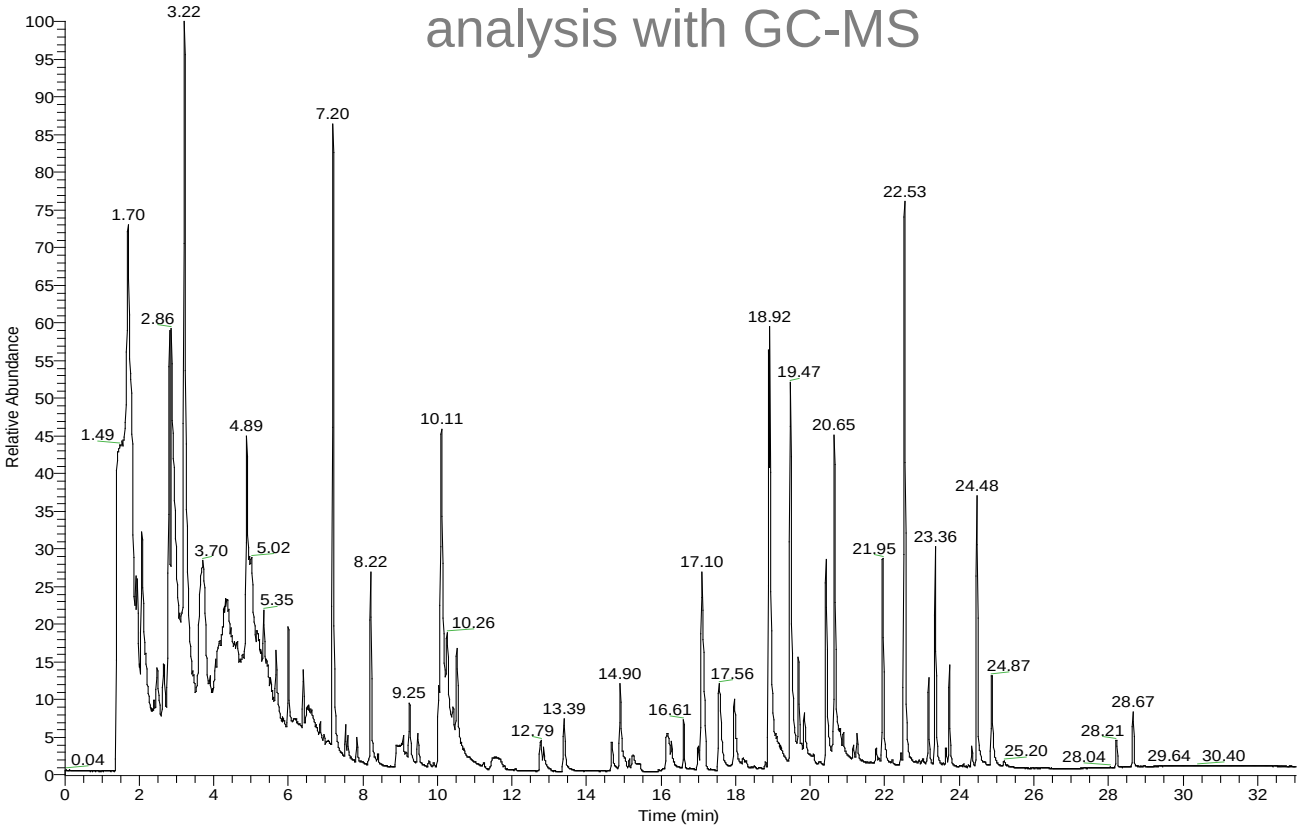


Metabolic activity of microbiota

- **Lactose-[^{15}N,^{15}N']-ureide** to measure metabolic **ammonia** metabolism
- **P-cresol and phenol** to measure **protein** fermentation
- **Bacterial enzyme activities:** β-glucuronidase and β-glucosidase
- **Metabolic profiles** of volatile organic compounds

Volatile organic compounds

analysis with GC-MS

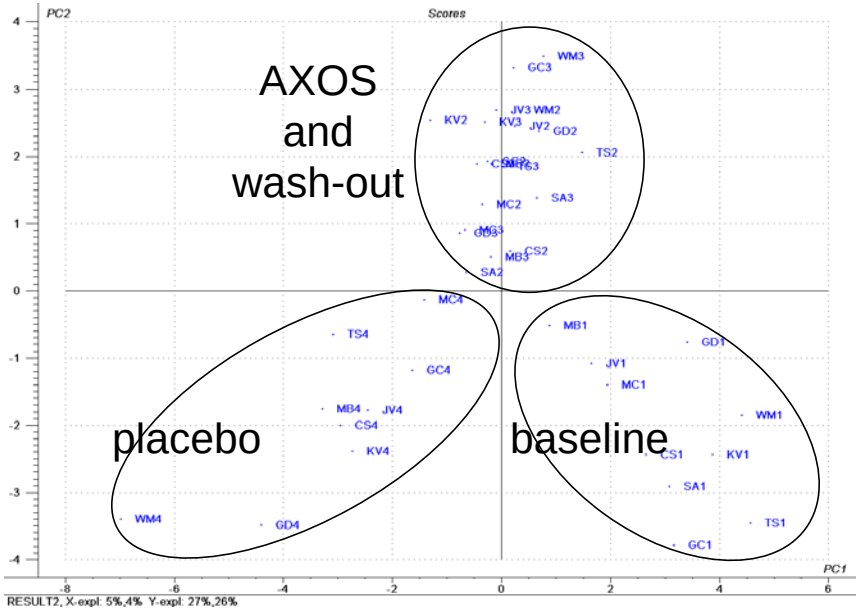


±60 volatile organic components per sample

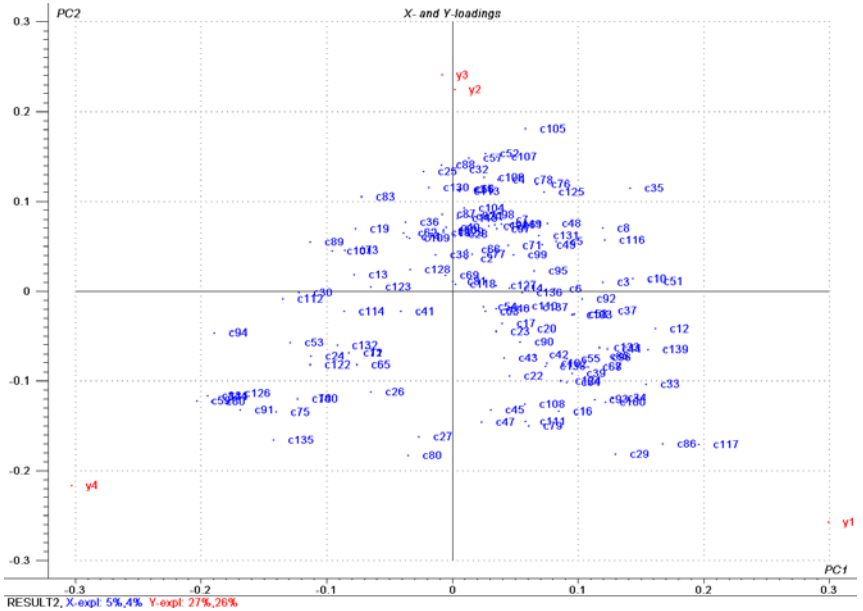
Metabolic profiles

multivariate data analysis

score plot



loading plot



discrimination was explained by differences in diet

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