Ingestion of probiotic *Bacillus coagulans* GBI-30 6086 increases plant protein digestion in a dynamic, computer-controlled *in vitro* model of the small intestine (TIM-1)

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Script of the presentation

- the TNO *in vitro* models of the GI tract at Maastricht University

- validation examples of the TIM models
  - survival of probiotics
  - protein digestion

- effect of BC30 on protein digestion
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A model to simulate the GI tract requirements for such models
• simulating successive dynamics
• sampling in time at different intestinal segments
• validation in vitro versus in vivo

In the late 1980's, TNO started the development of the TNO dynamic in vitro models of the GI tract, nick-named TIM
Successive dynamic conditions in the GI tract

- peristalsis, gastric acid, salivary and gastric enzymes, gastric emptying
- secretion of digestive enzymes, bile, intestinal transit time
- absorption of (digestion) products and water
- dense active microbiota, microbial metabolites
- biotransformation enzymes
- cecum and colon transit
TIM-1
gastric-small-intestinal model

1. stomach 6. gastric secretion
2. duodenum 7. intestinal secretion
3. jejunum
4. ileum
5. ileo-caecal valve

8. pH electrodes
9. prefilter
10 absorption system
Schematic view of TIM-1 with sampling ports

1. intestinal content concentration in time
2. dialysis flow concentration in time
3. dialysate (jej, il) total amount in time
4. ileal delivery total amount in time
Peristaltic movements

Gastro-intestinal transit
Stomach and ileal delivery and pH curves

- adult liquid
- adult semi-solid
- baby

- gastric pH
- duodenal pH
- jejunal pH
- ileal pH
TIM-systems available

- human models: babies, adults, elderly
  - healthy vs. patients
  - average vs. extremes
- dog model  Functional Intestinal Dog model
- pig model
- pre-ruminant calf model
- chicken model  Chicken ALIMENTary tract model
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Validation of the TIM-systems

TIM-1 has been validated with respect to
- protein digestion
- carbohydrate digestion
- fat digestion
- survival of probiotic microorganisms
- bioaccessibility of (fat-soluble) vitamins

TIM-2 has been validated with respect to
- the composition of the microbiota
- the metabolic activity of the microbiota
Validation survival LAB during GI passage

_in vitro_ (i.v.) versus humans (h) J. Dairy Sci. 80:1031, 1997

![Graph showing cumulative delivery of LAB in vitro and in humans over time.](image)
Protein digestion; peptides and AA absorption
Ileal digestibility; *in vivo* (fistulated pigs) *vs.* *in vitro* (TIM system)

\[ y = 0.9882x \]
\[ R^2 = 0.9728 \]

- skimmilk
- wheat
- rapeseed
- meat and bone
- faba beans
- barley
- wheat
- soy isolate
- skimmilk + wheat
- skimmilk
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### Protein digestion – degree of hydrolysis (DH)

**α-amino nitrogen (AAN) and total nitrogen**

<table>
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<th>A</th>
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<table>
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<tbody>
<tr>
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<td>7</td>
<td>1-4</td>
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</tbody>
</table>

![Diagram](image)
TIM-1 experimental set-up

* 3 protein sources:
  - pea
  - soy
  - rice

* with/without BC30
  - $1 \times 10^9$ CFU in the duodenum

* samples from jejunal and ileal dialysates for ‘absorption’, but also from ileal efflux for mass balance
Protein digestion – AAN, TN, TN/AAN ratio

- AAN is increased in presence of BC30 for all three proteins
- factor increase ~ 2 – 2.7
Protein digestion – AAN, **TN**, TN/AAN ratio

- TN is increased in presence of BC30 for all three proteins
- factor increase ~ 1.2 – 1.6
Protein digestion – AAN, TN, TN/AAN ratio

- Due to this a decrease in TN/AAN ratio for all three proteins
- factor \( \sim 1.2 - 2.4 \) (0.8 – 0.4)
- Indicating higher degree of hydrolysis

Experimental set-up 2\textsuperscript{nd} experiment

* meal
* BC30 added as spores in gastric compartment
* (separate goal: germination)

* samples from jejunal and ileal dialysates for ‘absorption’
Ratio of amino acids and dipeptides BC30/no BC30

Overall increase in amino acids and dipeptides absorption*

* larger peptides not present in the metabolomics platform
Ratio of amino acids and dipeptides BC30/no BC30

Even release in amino acids and dipeptides absorption in the colon*↓
* larger peptides not present in the metabolomics platform↓ sampled in the lumen of the model, not dialysate

Wrap up and
Take home messages
Conclusions

- Validated *in vitro* models of the gastrointestinal tract are great tools to understand the effects of probiotics and food components on bioavailability of nutrients (here amino acids).
- BC30 enhances digestion of plant-proteins

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Thank you very much for your attention