National Enzyme Company

Forsyth, Missouri, USA

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TNO Nutrition and Food Research

Zeist, Netherlands

Presents New Research on Digestion

The First Quantitative Evidence Proving The Efficacy Of Supplemental Enzymes

Background

The underlying premise for taking a digestive enzyme supplement is the capacity for better nutrient absorption through enhanced digestion. National Enzyme Company has been advocating this application for over 70 years. Throughout these years, we have collected a plethora of anecdotal and qualitative information backing the use of digestive enzymes. While we were convinced of the efficacy of fungal digestive enzymes, we have been lacking the quantitative information that would prove the same under disinterested scientific scrutiny. To achieve this goal, we embarked on a collaborative effort with TNO of the Netherlands to prove the efficacy of supplemental, fungal digestive enzymes for the digestion of food and absorption of nutrients.

The Netherlands Organisation for Applied Scientific Research TNO (TNO Nutrition and Food Research) based in Zeist, Netherlands, is a reputable scientific organization whose mission is to be a link of knowledge between fundamental research and its applications to food, drugs and agrochemicals. To quantify the efficacy of supplemental enzymes, TNO proposed a series of experiments using a computer controlled dynamic gastrointestinal model (TIM). TIM is a unique patented technology, developed by TNO, that simulates the conditions of the human stomach and the small intestine. Using this technology, we accurately replicated the dynamic environment of the human stomach and the small intestine when food is being digested and absorbed. The stellar feature of this system is that it allowed for sampling at various times during the digestive process, which enabled us to gather information in *real time* about the extent of digestion and absorption of food under various conditions.

The protocols for TIM have been validated. The various studies that have been performed using TIM are well documented in literature (see references p.7).

The first step in this project was the formulation of a digestive enzyme blend that was generic, yet also effective. To this end, NEC formulators created a blend of fungal digestive enzymes that is the basis of all our digestive enzyme products. In other words, we chose a basic blend of proteases, carbohydrases and lipases. This blend was tested under two sets of conditions to observe digestability and absorption of nutrients. The two conditions were perfect human digestion and impaired human digestion (explained in next section). The TIM system was fed a meal (standard FDA type) with and without NEC digestive enzymes. The extent of digestion was monitored by sampling nutrients (glucose and nitrogen) at various times and at different points in the GI tract.

Below is a graphical representation of the TIM system and the corresponding parts of the GI tract.

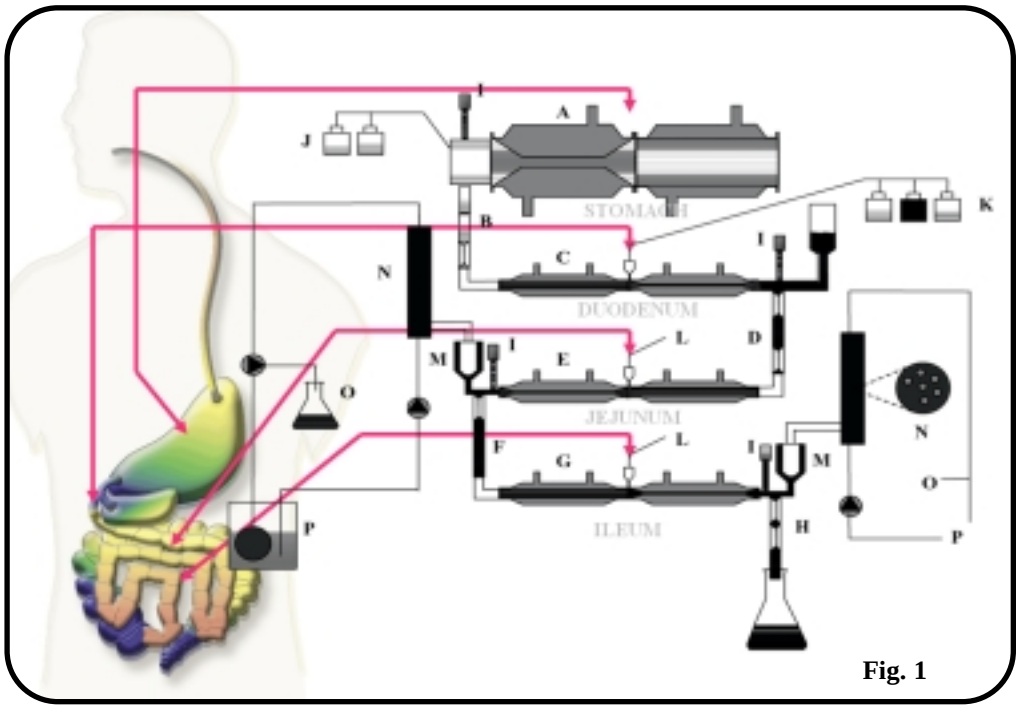


Fig. 1

Figure 1. Schematic diagram of the dynamic, multi-compartmental model of the stomach and small intestine (TIM-1): A. gastric compartment; B. pyloric sphincter; C. duodenal compartment; D. peristaltic valve; E. jejunal compartment; F. peristaltic valve; G. ileal compartment; H. ileo-caecal valve; I. pH electrodes; J. gastric secretion bottles with acid and enzymes; K. duodenal secretion bottles with bile, pancreatin, bicarbonate; L. secretion of bicarbonate to control the intestinal pH; M. pre-filter system; N. hollow fibre semi-permeable membrane system; O. water absorption system; P. closed dialysing system.

Study Protocol & Test Conditions

The study was performed in TNO's dynamic, multi-compartmental system of the stomach and small intestine (TIM) as depicted in Figure 2. The model simulated very closely the successive dynamic conditions in the stomach and small intestine of healthy human adults with normal gastric and intestinal secretions and of human adults with impaired digestion due to lower levels of gastric and intestinal secretions. In other words, a perfect digestive system and an extremely impaired digestive system were chosen as test conditions. The two extremes were tested because the digestive capabilities of most humans fall somewhere in between.

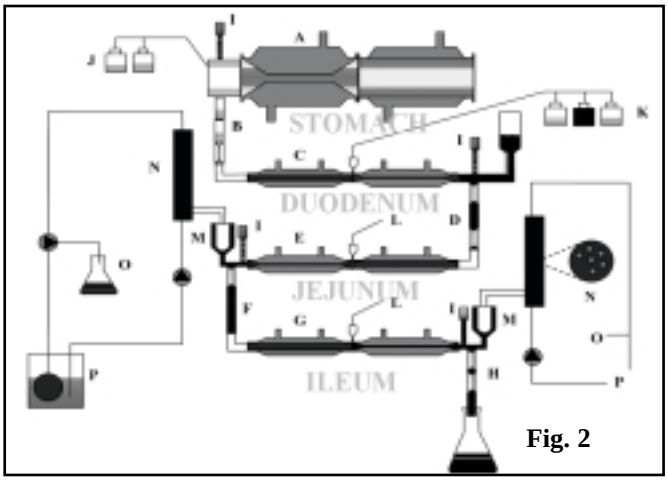


Fig. 2

Figure 2. Schematic diagram of the dynamic, multi-compartmental model of the stomach and small intestine (TIM-1): A. gastric compartment; B. pyloric sphincter; C. duodenal compartment; D. peristaltic valve; E. jejunal compartment; F. peristaltic valve; G. ileal compartment; H. ileo-caecal valve; I. pH electrodes; J. gastric secretion bottles with acid and enzymes; K. duodenal secretion bottles with bile, pancreatin, bicarbonate; L. secretion of bicarbonate to control the intestinal pH; M. pre-filter system; N. hollow fibre semi-permeable membrane system; O. water absorption system; P. closed dialysing system.

For each TIM run, 170 g of the standardized FDA-type of test meal (proteins, carbohydrates and fats) were mixed with 100 ml drinking water and 70 ml artificial saliva. Four different types of TIM experiments were performed in duplicate with this meal as follows:

(a) meal without the digestive enzyme blend under perfect digestive conditions

(b) meal with addition of digestive enzyme blend under perfect digestive conditions

(c) meal without the digestive enzyme blend under 70% reduced gastric and intestinal secretion (impaired digestion)

(d) meal with addition of digestive enzyme blend under 70% reduced gastric and intestinal secretion (impaired digestion)

The secretion products of the human digestive system consisting of gastric juice with enzymes, pancreatin, bile and bicarbonate were added to the system at the appropriate times. The pH was monitored and maintained at physiological conditions and peristalsis was mechanically simulated. The gastric emptying and intestinal passage time were mimicked as per human conditions.

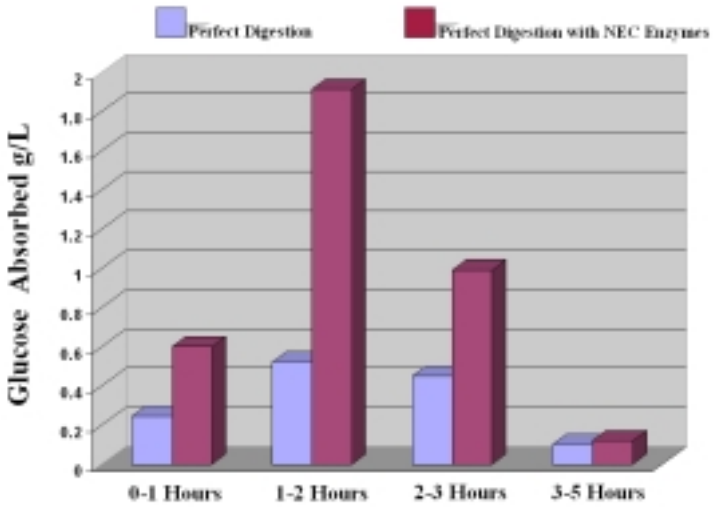
Sampling and Analysis

Samples were collected over a 5 hour span at two points in the small intestine: jejunum (upper small intestine) and ileum (lower small intestine). The samples represented the extent of digestion during each of the test conditions mentioned above. Samples were collected using a method called dialysis, which closely approximates the absorption of nutrients through the lumen of the gut. Thus, only nutrients, and not undigested food, passed through and were collected as samples. Analysis of these samples for glucose and nitrogen content correlated directly with the extent of carbohydrate and protein digestion respectively in the GI tract.

Results - Perfect Digestive Condition

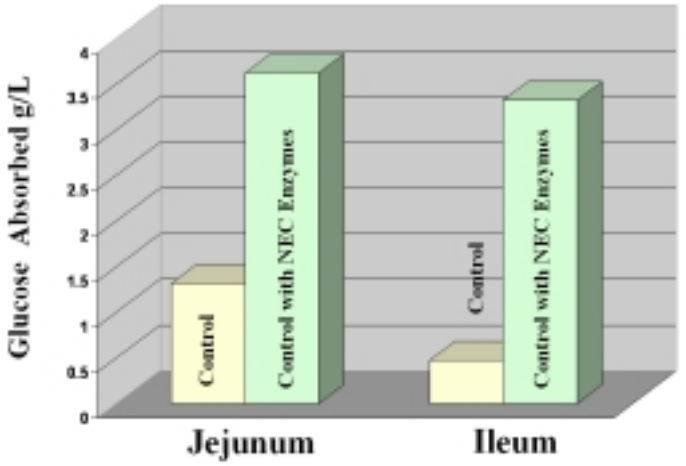
The following graphs compare the digestion of carbohydrates and proteins under ***perfect digestive conditions,*** tested with and without the aid of the NEC fungal digestive enzyme blend.

Digestion of Carbohydrates in Jejunum



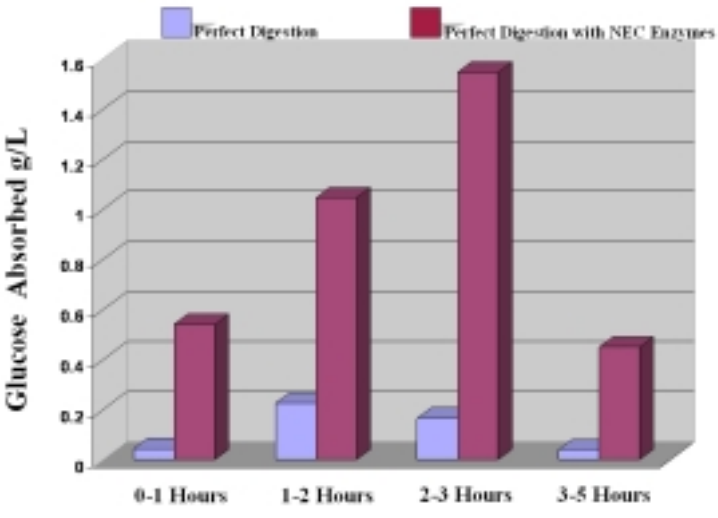
Graph 1: Shows a substantial increase in the level of digestion of carbohydrates in the lumen of the jejunum over a 5 hour span with NEC fungal digestive enzyme blend even under perfect digestive conditions.

Carbohydrate Digestion:
Small Intestine



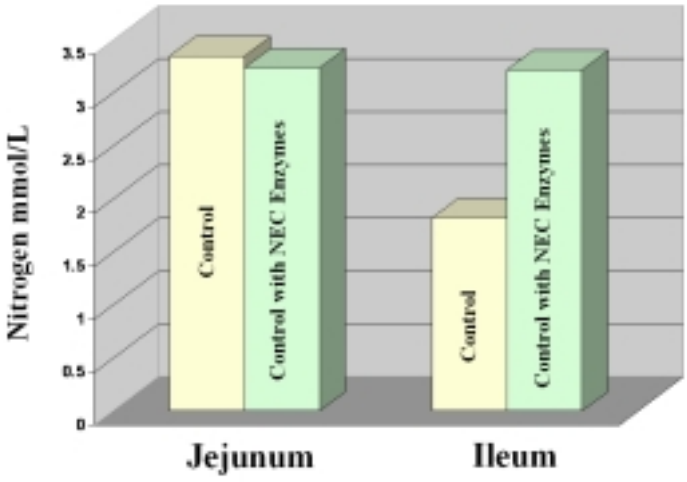
Graph 3: The total digestion of carbohydrates is increased nearly 4 fold in the small intestine with NEC fungal digestive enzymes even under perfect conditions.

Digestion of Carbohydrates in Ileum



Graph 2: Shows a substantial increase in the level of digestion of carbohydrates in the lumen of the Ileum over a 5 hour span with NEC fungal digestive enzyme blend even under perfect digestive conditions.

Protein Digestion:
Small Intestine

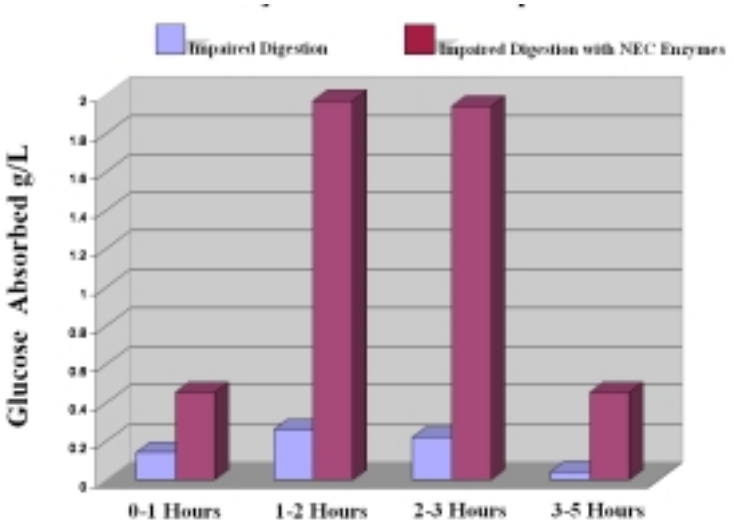


Graph 4: The digestion of proteins is slightly increased in the ileum and remains essentially the same in the jejunum with NEC fungal digestive enzymes under perfect conditions.

Results - Impaired Digestive Conditions

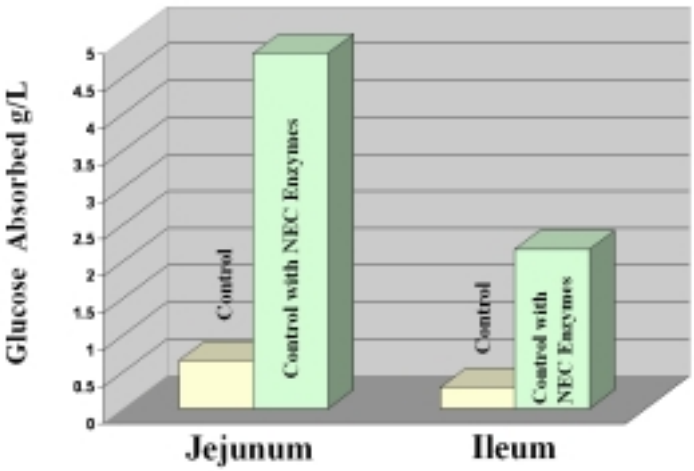
The following graphs compare the digestion of carbohydrates and proteins under ***impaired digestive conditions*** tested with and without the aid of NEC fungal digestive enzyme blend.

Digestion of Carbohydrates in Jejunum



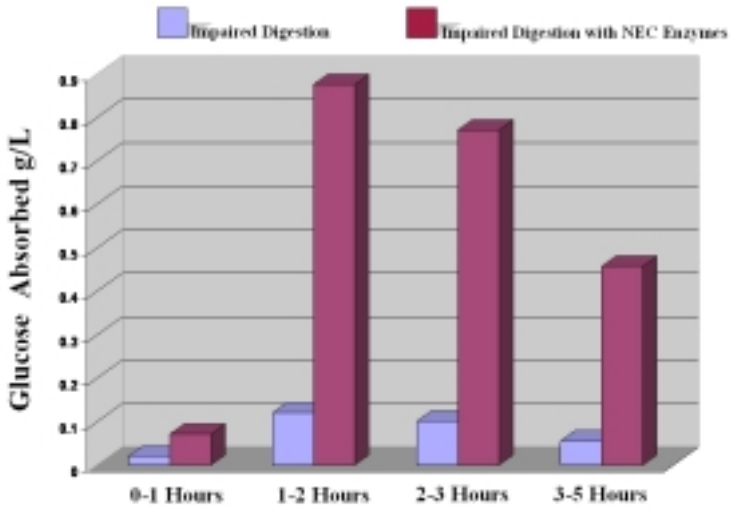
Graph 5: Shows a substantial increase in the level of digestion of carbohydrates in the lumen of the jejunum over a 5 hour span with NEC fungal digestive enzymes under impaired conditions

Carbohydrate Digestion: Small Intestine



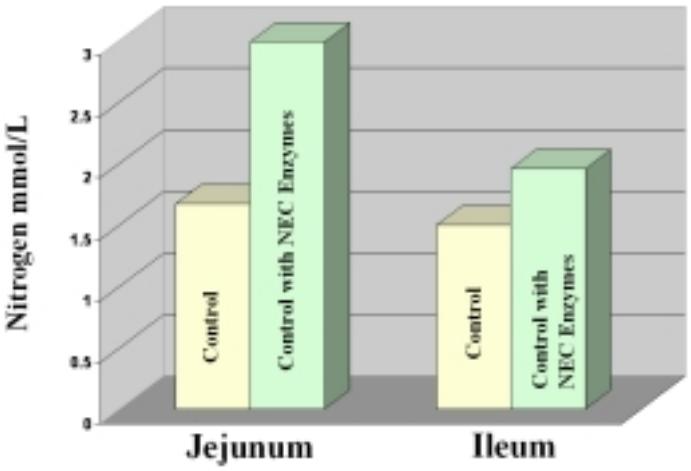
Graph 7: The total digestion of carbohydrates is increased about 7 fold in the small intestine with NEC fungal digestive enzymes under impaired conditions

Digestion of Carbohydrates in Ileum



Graph 6: Shows a substantial increase in the level of digestion of carbohydrates in the lumen of the Ileum over a 5 hour span with NEC fungal digestive enzymes under impaired conditions

Protein Digestion: Small Intestine



Graph 8: The digestion of proteins increases significantly in the small intestine with NEC fungal digestive enzymes under impaired conditions

Discussion

The aim of the study was to determine the efficacy of an NEC digestive enzyme supplement on the digestibility of proteins and carbohydrates, and the bioaccessibility of nutrients under the following conditions:

1) Healthy human adult digestion (perfect conditions)

2) Impaired digestion. 70% reduced gastric and intestinal secretions

Results show that NEC fungal digestive enzymes improve the digestibility and bioaccessibility of proteins and carbohydrates in the lumen of the small intestine, not only under impaired digestive conditions, but also in healthy human digestion. Furthermore, the test meal fed to the TIM system was an FDA recommended meal, which is smaller in macronutrient content and total calories than the typical American diet. The conservative amount of food used in the experiments, and the corresponding results obtained, further testify to the use of fungal digestive enzyme supplements.

The activity of any digestive enzyme supplement in the small intestine presupposes that the enzymes in the supplement survive the acidity of the stomach. From the above experiments we have established that NEC fungal digestive enzymes do survive the acidity of the stomach and work in the small intestine. In fact, our research has demonstrated that NEC fungal enzymes not only survive the acidity of the stomach, but also are active in that harsh environment where most other types of enzymes are inactivated. Therefore, there is no scientific basis for enterically coating NEC fungal enzymes for general digestive applications. Such action would be detrimental to the consumer since the coating would prevent the enzymes from working in the stomach.

This research is the first comprehensive study that shows that NEC fungal digestive enzymes substantially increase the level of digestion in the lumen of the small intestine and bioaccessibility of proteins and carbohydrates. These results not only validate the use of digestive enzymes in cases of impaired digestion, but also show that most healthy adults can benefit by using a digestive enzyme supplement.

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References

- Havenaar, R. (1999). The model selection tool. Dairy Industries International, 64 (6): 33-36.
- Havenaar, R. and Minekus, M. (1996). Simulated assimilation. Dairy Industries International 61(9): 17-23.
- Havenaar, R., Minekus, M. and Speckmann, A. (1995). Efficacy of Natuphos® phytase in a dynamic computer-controlled model of the gastro-intestinal tract. Proceedings European Symposium on Feed Enzymes, Noordwijkerhout, Netherlands. pp 211-212.
- Krul, C., Humblot, C., Philippe, C., Vermeulen, M., Van Nuenen, M., Havenaar, R., Rabot, 5. (2002). metabolism of sinigrin (2-propenyl glucosinolate) by the human colonic microflora in a dynamic in vitro large-intestinal model. Carcinogenesis 23:1009-1016.
- Krul, C., Luiten-Schuite, A., Tenfelde, A., Van Ommen, B., Verhagen, H. and Havenaar, R. (2001). Antimutagenic activity of green tea and black tea extracts studied in a dynamic in vitro gastrointestinal model. Mutation Research; 474: 71-85.
- Krul, C., Luiten-Schuite, A., Baan, R., Verhagen, H., Mohn, G., Feron, V., and Havenaar, R. (2000). Application of a dynamic in vitro gastrointestinal tract model to study the Availability of food mutagens, using heterocyclic aromatic amines as model compounds. Food and Chemical Toxicology 38: 783-792.
- Larsson, M., Minekus, M., and Havenaar, R. (1997). Estimation of the bioavailability of iron and phosphorus in cereals using a dynamic in-vitro gastrointestinal model. J. Sci. Food Agric. 73: 99-106.
- Marteau, P., Minekus, M., Havenaar, R. and Huis in 't Veld, J.H.J. (1997). Survival of lactic acid bacteria in a dynamic model of the stomach and small intestine: Validation and the effects of bile. J. Dairy Sci. 80: 1031-1037.
- Minekus, M. (1998). Development and validation of a dynamic model of the gastrointestinal tract. PhD Thesis, University of Utrecht; Elinkwijk b.v., Utrecht, Netherlands.
- Minekus, M., Marteau, P., Havenaar, R. and Huis in 't Veld, J.H.J. (1995). A multi compartmental dynamic computer-controlled model simulating the stomach and small intestine. Alternatives to Laboratory Animals (ATLA) 23: 197-209.
- Minekus, M., Smeets-Peeters, M., Bernalier, A., Marol-Bonnin, S., Havenaar, R., Marteau, P., Airic, M., Fonty, G., and Huis in 't Veld, J.H.J. (1999). A computer controlled system to simulate conditions of the large intestine with peristaltic mixing, water absorption and absorption of fermentation products. Appl. Microb. Biotechn. 53: 108-114.
- Minekus, M. and Havenaar, R. (1996). In vitro model of an in vivo digestive tract. United States Patent; nr. 5,525,305, dated June 11, 1996.
- Minekus, M. and Havenaar, R. (1998). Reactor system. European Patent No. 0642382. Eur. Patent Bulletin 98/07, Art. 97(4) and (5) EPC, dated 11.02.98.
- Smeets-Peeters, M.J.E. (2000). Feeding FIDO: Development, validation and application of a dynamic in vitro model of the gastrointestinal tract of the dog. PhD Thesis Wageningen University. Universal Press, Veenendaal, The Netherlands.
- Smeets-Peeters, M.J.E., Minekus, M., Havenaar, R., Schaafsma, G., Verstegen, M.W.A. (1999a). Description of a dynamic in vitro model of the dog gastrointestinal tract and an evaluation of various transit times for protein and calcium. ATLA 27: 935-949.
- Smeets-Peeters, M., Havenaar, R., Schaafsma, G. and Verstegen, M. (1999b). Validation of a dynamic computer-controlled in vitro model of the canine gastrointestinal tract. In: Proceedings World Congress on Alternatives to Laboratory Animal Science, Bologna, Italy.
- Smeets-Peeters, M, Watson, T., Minekus, M., Havenaar, R. (1998). A review of the physiology of the canine digestive tract related to the Development of in vitro systems. Nutrition Research Reviews 11: 45-69.
- Venema, K., Van Nuenen, M., Smeets-Peeters, M., Minekus, M. and Havenaar, R. (2000). TNO's in vitro large intestinal model: an excellent screeening tool for functional food and pharmaceutical Research. ErnahrungfNutrition 24 (12): 558-564.
- Zeijdner, E. and Havenaar, R. (2000). The fate of orally administrated compounds during passage through the gastrointestinal tract simulated in a dynamic in vitro model (TIM). European pharmaceutical Contractor, Febr. isue: 76-81.
- Zeijdner, E.E. and Mohede, I.C.M. (1999). Latest tool for screening new clinical foods. A dynamic, computer-controlled model of the human gastrointestinal tract is the most up-to-date technology for testing new foods. New World Health 1999/2000: 105.a
- Arkbge, K., Verwei, M., Havenaar, R., Witthˆft, C. (2003). Folic acid and (6S)-5-methyltetrahydrofolate bioaccessibility decreases after addition of folate-binding protein to yogurt as studied in a dynamic in vitro gastrointestinal model. J. Nutr. 133: 3678-3683.
- Verwei, M., Arkbge, K., Havenaar, R., Van den Berg, H., Witthˆft, C. and Schaafsma, G. (2003). Folic acid and 5-Methyl-tetrahydrofolate in fortified milk are bioaccessible as determined in a dynamic in vitro gastrointestinal model. J. Nutr. 133: 2377-2383.
- Verwei, M., Arkbge, K., Mocking, H., Havenaar, R. and Groten, J. (2004). The binding of folic acid and 5-methyltetrahydrofolate to folate-binding proteins during gastric passage differs in a dynamic in vitro gastrointestinal model. J. Nutr. 134: 31-37.
- Van Nuenen, H.M.C., Meyer, P.D., Venema, K. (2003). The effect of various inulins and Clostridium difficile on the metabolic activity of the human colonic microbiota in vitro. Microbial Ecology in Health and Disease 15 (2-3): 137-144.
- Venema, K. Van Nuenen, H.M.C., Van den Heuvel, E.G., Pool, W., Van der Vossen, J.M.B.M. (2003).The effect of lactulose on the composition of the intestinal microbiota and short-chain fatty acid production in human volunteers and a computer-controlled model of the proximal large intestine. Microbial Ecology in Health and Disease, 15 (2-3): 94-105.

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