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Interview

with prof. Beatrice Pool-Zobel

In this issue we are glad to introduce Professor Beatrice Pool-Zobel, Head of the Department of Nutritional Toxicology and Director of the the Institute of Nutrition, Friedrich Schiller University Jena, in Germany.

As Head of the Department of Nutritional Toxicology, Professor Pool-Zobel is involved in studies on the intracellular effects of risk factors for colon cancer by detection of strand breaks and oxidised DNA bases with the Comet Assay or by determining the capability of these compounds to induce damage in relevant genes (p53,APC) using primary colon cells.

From cancer research to nutrition research

Beatrice Pool-Zobel is also a keen sports woman: she runs three to four times a week and does aerobics. "I run for fun, but I try to do it as much as possible, even when I attend a congress. Often, I practice early in the morning, before I start to work. However, I have no ambitions to run a marathon. I do it just for fun and to feel good."
She is also a great traveller. "I very much like to travel, all over the world. For a congress I often have to go more than once to the same place, but when I am on vacation I never go to the same place twice, I always choose a new destination. When I travel, I read a lot about the country I am visiting. Not just tourist guides, but fiction, based on the histories e.g.of Alaska or Africa, a story that is situated in Los Angeles."
Anyone who has attended a lecture by Beatrice Pool-Zobel will have noticed that she pays a lot of attention to the visual presentation of her work. "When I have to make a presentation, I spend time to make it as attractive as possible. In fact, I like very much to design things using the computer, it is a kind of a hobby, and it is a modern kind of handwork like embroidering wich I liked to do years ago, when I had more time."

► She also studies the effects of phyto-protectants on specific signal transduction pathways in colon cells and which consequences this may have in complex patterns of gene expression and cellular protein profiles. Furthermore, Professor Pool-Zobel is involved in the development of biomarker methods indicative of cancer protection by nutritional factors and in the establishment of these methods for human dietary intervention studies and for molecular epidemiological investigations. Finally, the Department studies the impact of genetic polymorphisms on the efficacy of nutritional intervention. This includes research on gene-environment interactions by determining reduction of genetic damage and induction of chemopreventive enzymes following intervention with plant foods.
Professor Pool-Zobel studied at the Karl-Ruprecht Universität in Heidelberg, where she specialised in biochemistry and genetic toxicology. She started her scientific career as a cancer researcher in the Deutsches Krebsforschungszentrum in Heidelberg. Here her main interests were in assessing gentetic effects of environmental chemicals in tumor target tissues, and enhancing the understanding of how mutations caused by active metabolites of thes chemicals can be important for the cancer process. In 1990 she became the head of the toxicology department at the Bundesforschungsanstalt für Ernährung (BFE) in Karlsruhe. In 1995 she switched to the Department of Molecular

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Preface

by Dr. Anne Franck

Prebiotics may increase your sense of well-being

With this new issue after the summer holiday break we would like to update you about recent scientific developments in the field of prebiotic dietary fibres, their nutritional benefits – especially with regard to gut function – and their use in baby food, as well as in feed and pet food applications.
Prebiotics are an exciting concept in human nutrition. They stimulate selectively the growth of bifidobacteria and lactobacilli in the gut, and they increase the body's natural resistance to invading pathogens. Thereby, they could prevent episodes of diarrhoea, an effect that is under evaluation.
Inulin and oligofructose improve bowel habits and significantly increase the absorption of calcium in the body. Other areas of research involve the impact of prebiotics on both colonic and systemic disorders, including ulcerative colitis, colon cancer, hyperlipidemia, liver disease, osteoporosis and compromised immune function.
In a recent study involving subjects on holiday, oligofructose significantly increased the sense of well-being, an interesting observation that could lead to new developments in the area of functional foods.
Consuming foods containing oligofructose may limit the seriousness of common illnesses in young children and the need for treating them with antibiotics, as reported at the 3rd ORAFTI Research Conference.
Oligofructose may stimulate the vaccine-response in infants.
More and more scientific data on animal feed indicate that prebiotics may improve growth rate and feed conversion, may provide an alternative to growth promoters, and may prevent intestinal disorders and infections.
We invite you to browse through this issue for more information on these topics.

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Toxicology and Carcinogenesis of the Institute for Nutritional Physiology, also at he BFE in Karlsuhe. Since July 1998, Professor Pool-Zobel heads the Department of Nutritional Toxicology at the Friedrich Schiller University in Jena.
"When I switched from cancer research to nutrition research, I began to look at exposure factors for their genotoxic effects, and at the interaction between risk factors and protective factors. In nutrition research, you have to acknowledge not only the risk factors, as you would in environmental or toxicological research. Instead risks must be evaluated by taking into account all the food in the diet. Of course, this will includes protective factors as well. Since I was interested in the colon, where the most important process that takes place is fermentation, I was also interested in how positive fermentation profiles of prebiotics and dietary fibre were responsible for protective effects of pro- and prebiotics."
Professor Pool-Zobel is an honorary member of the International Union Against Cancer and a member of the BENEO®Scientific Committee and has received the prestigious award "Dr. Rainer Wild Preis" for her research on beneficiary effects of nutrition in cancer prevention. She is a member of national and international organisations, and she sits on the editorial and advisory boards of several scientific journals.
"Our food may be at the origin of one-third of all cancers. The consumption of more vegetables and fruits on the one hand, and of less meat and animal fat on the other hand, is essential to decrease disease risk. The underlying reasons and the detailed mechanisms of the protective activities of plant foods are not fully understood. Although the identification of specific chemopreventive food substances is very difficult, it is well established that certain non-digestible food ingredients have a chemopreventive activity. I am particularly interested in seeing how pre- and probiotics interact with relevant colon carcinogens. Mainly, the work of my group is aimed at studying the impact of the fermentation products on human colon cells and to assess the modulation of gene expression in these epithelial cells. Colon cancer is a frequent, diet-related disease, for which individual carcinogenic risk factors have not been identified conclusively.

Colon cells are particularly susceptible to toxic compounds of the faeces, due to the long periods of physiological exposure. However, little is known about the consequences of activating and inactivating enzymes in colon cells. I have been involved in developing systems that would enable us to investigate the effects of environmental compounds on the DNA of target tumour tissue. One major step was the development of the single-cell microgel electrophoresis technique (Comet Assay), one of the techniques we have been using for the past twelve years. It is a rapid and very sensitive fluorescent microscopic method for examining DNA damage and repair at the level of the individual cell. Since the introduction of the Comet Assay in 1988, by R. Tice and N.P. Singh, a number of improvements have greatly increased the flexibility and utility of this technique for detecting various forms of DNA damage and repair in virtually any eukaryotic cell. Because of the complexity of cancer initiation and progression, many types of interaction may be envisaged. One possible mechanism is the detoxification of genotoxins in the gut. We are investigating whether the proteins and the enzymes of the so-called chemopreventive system are altered. These enzymes, such as glutathione-S-transferase (GST) and UDP-glucuronosyltransferase, play an important role in inactivating or detoxifying several risk factors.The rationale underlying our studies is based on the hypothesis that if colon epithelial cells have higher detoxifying activity or are more potent in repairing DNA damage, they will be more resistant to colon cancer risk factors and this will retard initiation and progression of cancer. This mechanism has been shown experimentally in animals with use of the colon carcinogen 1,2-dimethylhydrazine (DMH) in rats, and by determining endpoints that range from tumours to induction of DNA damage. Several lactic acid bacteria, such as Lactobacillus acidophilus, and bifidobacteria decrease DNA damage in the gut. Ingestion of prebiotics results in a different spectrum of fermentation products, including the production of high concentrations of short-chain fatty acids, which may enhance GST expression. We are looking in human colon cells for modulation of gene expression. We are studying, for instance, the effect of fermention products, as well as the effect of model compounds like butyrate, proprionate, acetate and lactate on cellular events of early carcinogenesis, genotoxicity and cytotoxicity in rat distal colon cells."

You studied especially colon cancer. But did you also do research on other types of cancers ?

"We are presently performing a study using lignans and isoflavonoids. These polyphenolic compounds have been suggested to be chemopreventive on account of their antioxidative properties. In this context, it is important to have knowledge of their ability to reduce oxidative stress within target cells of tumorigenesis. We are investigating their activities in human breast and prostate cells. As soon as we know a little bit more about these cells and the possibility of specific active fermentation products being active locally, and being distributed systemically, we will look into these mechanisms as well. The consequences of our colon cancer work could also be to analyse similar effects in systemically affected target tissues of carcinogenesis. The possible effects would be dependent on the systemic distribution of the protective factors we identified in the colon".

What do you expect for the future?

"I hope we will understand better the interaction between gut fermentation products and colon epithelial cells. We also need to know more about the interaction of pre- and probiotics; the synbiotic effect. Then, we should be able to use the results from our gene expression profiles for developing biomarkers. This biomarker development will be very important in the future. So, we are now looking at the impact of fecal water on colon cells to see if we can use it as an effective biomarker to determine whether pre- and probiotics can be of benefit for humans. Exogenous nutritional factors modulate the fecal contents, leading to an enhanced or reduced burden of toxic and carcinogenic factors. These factors are thought to contribute to colon cancer by inducing mutations or enhancing proliferation in colon cells. Fecal water has been shown to cause these effects in model systems, and thus could be the basis for valuable biomarker approaches. Our investigations are aimed at determining geno- and cytotoxicity of fecal water in human colon cell lines in vitro. We are developing techniques for their applicability as biomarker tests during dietary intervention studies. We have now determined baseline toxic activities and calculated inter and intra-individual, and experimental coefficients of variation for fecal water from different subjects consuming similar or different diets. However, following intervention with certain protective dietary regimens (e.g. lignan-containing bread), significant reduction of fecal water-induced genotoxicity has been observed. Therefore, in spite of the expected and observed degrees of variation in this methodology, effective experimental protocols may still lead to detectable modulation of the level of toxic and genotoxic effects. Also, biomarker development using colon cells from biopsy samples is very important and should be performed in high-risk groups. We are using this type of approach in SYNCAN, the EU-funded project on synbiotics and colon cancer risk. ■

Topic

State of the Art: The Effects of Inulin and Oligofructose on Gut Function

Non-digestibility in the Small Intestine

The beta configuration of the anomeric C-2 in the fructose monomers makes inulin-type fructans non-digestible by human enzymes, which are mostly specific for alpha osidic linkages (Roberfroid & Delzenne 1998; Roberfroid & Slavin 2000; Cherbut 2001). Studies with human ileostomy subjects by Bach Knudsen & Hessov (1995), Ellegard et al. (1997) and Anderson et al. (1999) have demonstrated that inulin and oligofructose are practically indigestible in the small intestine of man. Average recovery at the terminal ileum is about 90% of the material fed. Similarly, when gut contents are aspirated from the terminal ileum after test meals containing oligofructose, almost 90% is recovered (Molis et al. 1996). The small loss of inulin-type fructans during the passage through the small intestine is probably due to anaerobic fermentation by the microbial population colonising the ileum, as was shown by Bach Knudsen & Hessov (1995).

Inulin and oligofructose pass through the small bowel without degradation and without influencing the absorption of nutrients or minerals. They do not appear to have any marked effect on cholesterol or bile acid excretion in the small bowel, and they do not increase the viscosity of the aqueous phase (Ellegard et al. 1997). Their primary effect in the small intestine is to increase bulk (Schneeman, 1999).

A study with dogs by Buddington et al. (1999) indicated that a diet containing oligofructose was associated with heavier small intestines, increased mucosal surface and increased mucosa, especially in the proximal portion of the intestine, and increased capacity for nutrient absorption. These changes may reduce the risk of enteric infections or aid in the treatment of intestinal diseases, particularly those involving reduced nutrient absorption. In mice fed diets with 10% inulin or oligofructose, the small intestine was longer and heavier compared with mice fed diets with 10% cellulose. Rates of glucose transport and absorption of leucine, proline and glycyl-sarcosine were lower in mice fed diets with 10% inulin or oligofructose. Since inulin and oligofructose are not digested, these changes in intestinal length and transport functions must have been mediated by other signals. These findings show that it is possible that the responses of different amounts and types of non-digestible oligosaccharides (NDO) are of greater magnitude in more proximal regions of the gastrointestinal tract than what is evident from the distal colon or stool samples (Buddington et al. 2000).

Selective Fermentation in the Large Bowel

Carbohydrates that are not digested and utilised in the small intestine reach the colon, where they can be partly degraded by the microflora. The monomeric composition, the type of glycosidic linkage, the degree of polymerisation, the solubility and the structural arrangement of the carbohydrates are factors of impor-

tance for the degree of fermentation (Nyman 2001).
Inulin and oligofructose are not recovered in the stools (Molis et al. 1996; Alles et al. 1996, 1997; Castiglia-Delavaud et al. 1998), indicating that they are fermented completely in the colon. That inulin and oligofructose are fermented by bacteria colonising the large bowel is supported by a large number of in vitro and in vivo studies (for reviews, see Roberfroid & Delzenne 1998; Roberfroid & Slavin 2000; Jenkins et al. 1999; Cummings & Macfarlane 2001).
Using gas and short-chain fatty acid production as endpoints, Wang & Gibson (1993) showed that human fecal slurries fermented oligofructose along with a wide range of other carbohydrates, but that inulin and oligofructose stimulated the growth of bifidobacteria selectively, whilst maintaining populations of potential pathogens at relatively low levels. Pure-culture studies confirmed the enhanced ability of bifidobacteria to utilise these substrates in comparison with glucose. These findings were subsequently confirmed in diet-controlled human studies (Gibson et al. 1995); Roberfroid et al. 1998). These studies showed that the intake of inulin or oligofructose modifies the composition of the fecal microbiota significantly by stimulating the growth of bifidobacteria, which become by far the most numerically predominant bacterial group. In addition, feeding inulin or oligofructose reduces the count of bacteroides, fusobacteria and clostridia. These effects last as long as inulin or oligofructose are consumed. After 2 weeks on a control

Worldwide official recognition of inulin and oligofructose as dietary fibre

Can inulin and oligofructose be labelled under the 'dietary fibre' group in the nutrition panel of foods ? This question has kept us much busier than we expected since the early nineties.
Today, we can rely on official statements from practically all countries where we are operating, all of which confirm that indeed, inulin and oligofructose can be labelled as dietary fibre. The first countries to confirm this were the European countries. The last European country to accept and confirm was the United Kingdom. The new Food Standards Agency confirmed, in September 2000, that the AOAC methods for dietary fibre analysis, including that for inulin and oligofructose, are now accepted for food labelling. This concluded a period of more than 5 years of discussions with the authorities.

Recently, in May 2001, the Australian authorities have proposed new regulations, soon to be adopted in a final form, which review the existing labelling regulations. They explicitly accept inulin and oligofructose for dietary fibre labelling. Here too, the time between our official request and the conclusion was long: no less than 6 years.
In another set of recent developments, several official bodies in the USA have confirmed this status. AOAC International had already published its opinions (Prosky, 1999), AACC has now edited its Report on the Definition of Dietary Fiber, stating explicitly that both inulin and oligofructose should be included. A recent report of the Institute of Medicine of the National Academy of Sciences (USA) has come forward with new definitions for Dietary Fiber, Added Fiber and Total Fiber. Here, it is clear that inulin and oligofructose are included in the Total Fiber group for nutrition labelling, and can be considered "dietary" or "added" fiber depending on whether it is present intrinsically or added.
Similar confirmations have been obtained in South America and Asia. These lead to one general conclusion; that the scientific and regulatory authorities worldwide have accepted both inulin and oligofructose for dietary fibre labelling on foods.

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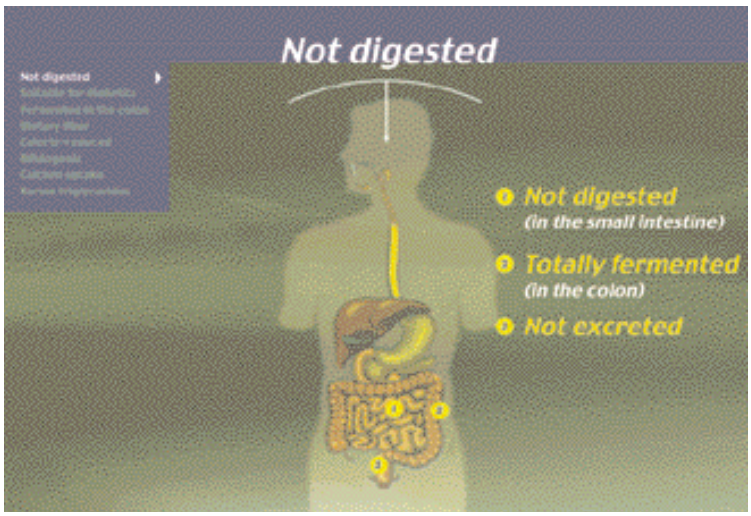
unsupplemented diet following the study period, the composition of the fecal flora is still different from the control, indicating that the changes disappear progressively. Inulin and oligofructose may therefore be classified as prebiotics, that is, non-digestible food ingredients that affect the host beneficially by stimulating the growth and/or the activity of one or a limited number of bacteria in the colon selectively and thus improve host health (Gibson & Roberfroid 1995; Roberfroid & Delzenne 1998; Roberfroid & Slavin 2000; Cummings & Macfarlane 2001; Cummings et al. 2001; Roberfroid 2001). Cummings & Macfarlane (2001) reported the results of a study showing that the prebiotic effects of inulin



and oligofructose may be seen at the critical interface between the mucosa and surface-associated bacteria.

Production of Short-Chain Fatty Acids and Other Metabolites

The major products of prebiotic metabolism are short-chain fatty acids (SCFA), the gases hydrogen and carbon dioxide (and methane in ~50% of the population), and bacterial cell mass.

When incubated in vitro with a human fecal flora, inulin and oligofructose produce SCFA (Wang & Gibson, 1993). This effect has been demonstrated also in rats harbouring a human intestinal flora. In these animals fed a 10% fibre diet, cecal concentration of total SCFA was higher with inulin than with fibre from cereals or vegetables. Fermentation of inulin promoted production of butyrate, the molar ratio of which increased to 27% with inulin as compared to about 10% with insoluble or partly soluble fibre (Roland et al, 1995). A similar butyrogenic effect was reported with oligofructose (Campbell et al. 1995,1997). This indicates that populations other than the bifidobacteria also benefit, as bifidobacteria do not produce butyrate. In addition to butyrate, production of acetate, propionate and lactate is enhanced by fermentation of inulin and oligofructose.

From in vivo animal studies, it can be concluded that supplementing a diet with non-digestible oligosaccharides (NDO) decreases the cecal pH, increases the size of the cecal pool of SCFA, with acetate the primary acid, followed by butyrate and propionate (Roberfroid & Slavin 2000). Related to this increase in the pool of short-chain carboxylic acids is the effect of some NDO on the intestinal tissue leading to hyperplasia of the mucosa and increased wall thickness in the small intestine and the cecum (Campbell et al. 1997). This effect is accompanied by an increase in blood flow.

It is largely accepted that SCFA, especially butyrate, play an essential role in maintenance of colonic mucosa integrity, by acting on metabolism, proliferation and differentiation of the different epithelial cell types. It may affect apoptosis (Hughes et al. 2001). It is possible that certain bacteria also influence epithelial cell functioning directly (Cherbut 2001). Ceco-colonic mucosa contained greater amounts of sulphomucins and smaller amounts of sialomucins in heteroxenic rats (harbouring a human intestinal flora) fed an inulin-containing diet as compared to a sucrose-containing diet. The very small amount of mucin that was recovered in the colonic mucosa suggests that, in the presence of the bacterial flora and associated with inulin in the diet, mucin was released extensively from the mucosa to the colonic lumen. These variations in the cecal and colonic mucosa are important to consider, as it has been reported that sulphomucin can increase potential resistance to attacks by bacterial enzymes. The change in mucin composition may contribute to the protective effect of these non-digestible carbohydrates in the incidence of intestinal diseases such as inflammation or certain forms of colon cancer (Fontaine et al. 1995).

Delzenne et al. (2000) investigated whether the fermentation of oligofructose in the gut produced other metabolites that may contribute to the physiological effects of dietary

fructans. Rats were fed a standard diet or the same diet enriched with 10% oligofructose for 4 weeks. Oligofructose almost doubled the concentration of putrescine in the cecal contents, and the concentration of polyamines (spermidine, spermine and putrescine) in the cecal tissue was significantly greater than in controls.

Gases and Intestinal Discomfort

In case of consumption of high amounts of prebiotics, the gases carbon dioxide and hydrogen (and methane in ~50% of the population), which are inevitable products of fermentation, can lead to unwanted gastro-intestinal symptoms such as bloating, flatulence and abdominal pain (Cummings et al. 2001). Mostly, these symptoms are mild. The effects are more pronounced with the short-chain oligosaccharides than with long-chain inulin (Rumessen et al. 1998; Carabin et al. 1999).

A series of clinical studies has been reported which shows that up to 20g/day of inulin or oligofructose is well tolerated. In children aged 10 to 13 years, doses of oligofructose up to 9g/day were well tolerated. It was further observed that oligofructose was better tolerated when incorporated into solid food as opposed to liquid, and when given in divided doses throughout the day (Brighenti et al. 1999; Carabin et al. 1999; Roberfroid & Slavin 2000).

Bulking Effect

The consensus report from the EU-funded ENDO project (Van Loo et al. 1999) stated that there is strong and consistent evidence of an effect of NDO on bowel habit, which results in a typical fecal bulking effect and in a normalisation of stool frequency (aiming at daily defecation). Stool bulking is considered to be protective against colon cancer, probably due to reduced exposure of the colonic mucosa to carcinogens (Hughes et al. 2001).

In most studies, there was a moderate increase in fecal bulk following ingestion of inulin or oligofructose (Roberfroid 1997). Their bulking capacity, an increase of 1-2 g of stool per g of oligofructose or inulin ingested, results mainly from increases in microbial biomass in the colon. Alongside the increase in excretion of dry matter, there is a significant increase in fecal nitrogen (Gibson et al.1995). The effect on stool output depends on the dose and on the amount of fibre in the diet (Brighenti et al. 1999).

Inulin and Oligofructose increase fecal water content (Gibson et al. 1995; Kleesen et al. 1997; Castiglia-Delavaud et al. 1998; Den Hond et al. 2000), stimulate bowel movements and increase stool frequency, particularly in slightly constipated subjects.

Cummings et al. (2001) concluded that the effect of inulin and oligofructose is moderate and similar to that produced by rapidly fermented polysaccharides, such as pectin and gums.

The first demonstration of a bulking effect was the controlled-diet study by Gibson et al. (1995), which showed that at 15g/day inulin and oligofructose increased stool output significantly, from 136 to 154 g/day and from 92 to 123 g/day, respectively.

The increase in stool frequency was observed clearly when inulin was incorporated into the diet of healthy but chronically, slightly constipated elderly human volunteers (Kleessen et al. 1997). The subjects had only one or two bowel movements per week. They received lactose or inulin for a period of 19 days: 20g/day from days 1 to 8, gradually increased to 40g/day from days 9 to 11, and kept at this dose from days 12 to 19. Only inulin stimulated the growth of bifidobacteria and suppressed other organisms such as enterococci (in number) and enterobacteria (in frequency). Inulin showed a significant effect on stool frequency and reduced functional constipation with only mild discomfort. The

stool frequency in seven of ten patients was increased to eight or nine per week, independent of the amount of inulin. Stools were soft, but diarrhea was not observed. In two other patients, the effect was dose-dependent. The percentage of dry fecal matter decreased significantly in response to 20g/day of inulin, corresponding to an increase in the water content.

Den Hond et al. (2000) studied the effect of high-performance inulin (with higher average chain length) on mild constipation in healthy volunteers with low stool frequency (one stool every two to three days). Subjects were administered inulin at 15g/day for 2 weeks. There was a significant increase in stool frequency (from 4 to 6.5/week) and fecal bulk increased by 1.5-2g per gram of inulin ingested. Gastric emptying rate, orocecal transit time, total transit time and intestinal permeability were not affected. This suggests that the chain length is of only minor importance for fecal bulking capacity. Menne et al. (2000) observed a change in stool frequency and appearance (softer) with oligofructose at only 8g/day. Tominaga et al. (1999) reported effects for oligofructose at 3g/day on stool frequency and quantity in healthy female volunteers. Also in patients with ileal pouch-anal anastomosis, receiving oligofructose at 15g/day, stool frequency and fecal weight were increased (Alles et al. 1997).

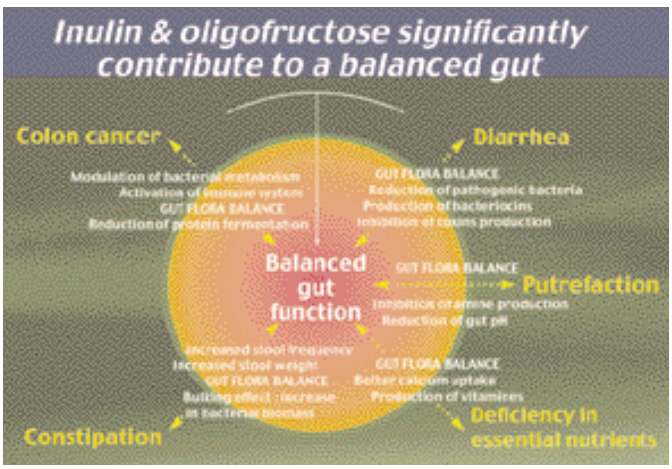
Inulin and oligofructose may have an inhibitory effect on diarrhea, especially when it is associated with intestinal infections (Roberfroid & Slavin 2000). This may be related to the inhibitory effect of bifidobacteria on Gram-positive and Gram-negative bacteria that has been reported by Wang & Gibson (1993) but this effect has not been demonstrated convincingly in humans.

Cummings et al. (2001b) report a randomised, double-blind, placebo-controlled study in 224 healthy subjects travelling to high and medium-risk destinations for travellers' diarrhea: 42% of the travellers experienced diarrhea and the consumption of 10g oligofructose per day failed to prevent it, but did give subjects an increased sense of well-being and less irritable bowel symptoms while on holiday. A likely reason for the failure of oligofructose may lie in the fact that it primarily affects the large intestinal flora, whereas many pathogens affect the small bowel. Moreover, it is equally likely that subjects experience episodes of diarrhea for reasons other than simple infection (food intolerance, alcohol use, etc.).

Effect on Uremia and Nitrogen/Urea Disposal

Feeding rats a diet supplemented with 10% inulin or oligofructose enhances fecal nitrogen excretion. In parallel, renal excretion of nitrogen is reduced significantly. The total nitrogen balance is not affected by the treatment (Delzenne et al. 1995; Younes et al. 1997).

This occurs because these non-digestible carbohydrates serve as an energy source for the intestinal bacteria, which require also a source of nitrogen for protein synthesis during growth. However, it seems unlikely that these carbohydrates exert any noticeable effect on protein digestibility in the small intestine. Their osmotic effect in the small intestine accelerates the transfer of urea into the distal ileum and the large intestine, where a highly ureolytic microflora may proliferate. When fermentable carbohydrate intake is high, the amount of ammonia may become insufficient to sustain maximal bacterial growth. In such conditions, blood urea constitutes the largest and the most readily available source of nitrogen for bacterial protein synthesis in the cecum. Younes et al. (1997) have shown that this transfer is proportional to cecal size and to uremia, and that the breakdown of large amounts of carbohydrate increases the incorporation of urea nitrogen into bacterial proteins.



In the presence of ammonia and amino acids, ureagenesis in the liver is inhibited by propionate, an important end-product of bacterial fermentation of inulin-type fructans. In fact, feeding fermentable carbohydrate leads to greater absorption of SCFA through the cecal wall, followed by extensive uptake by the liver, where propionic acid is liable to decrease the intra-mitochondrial concentration of carbamylphosphate by decreasing ATP content.

Some studies have shown that consumption of non-digestible carbohydrates results in a higher fecal excretion of nitrogen in humans (Roberfroid & Delzenne 1998; Roberfroid & Slavin 2000).

Caloric Value

As inulin-type fructans are not utilised in the small intestine but fermented in the colon, their energy value depends on their fermentability, energy lost as microbial biomass, hydrogen, methane and fermentation heat, efficiency of SCFA and lactate utilisation (Cherbut 2001). Their available energy content is only 40-50% that of a digestible carbohydrate. A caloric value of 1.5 kcal/g for labelling purposes has been proposed (Roberfroid 1997, 1999, 2000; Carabin & Flamm 1999). Molis et al. (1996) used the factorial method to determine a caloric value of 2.3 kcal/g. Direct measurements of the metabolisable energy content by whole-body indirect calorimetry gave similar energy values in humans (Castiglia-Delavaud et al. 1998).

Moreover, inulin and oligofructose may interact with the digestion of proteins and fats, thus lowering the caloric value of the diet as a whole. Therefore, they can be classified as low-energy food ingredients. The daily intake of these carbohydrates is likely to remain relatively small, probably often not more than 5% of total daily calorie intake.

Labelling as Dietary Fibre

Dietary fibre has been defined as the skeletal remains of plant cells resistant to hydrolysis by human alimentary enzymes, and later expanded to include

polysaccharides asssociated with the cell wall, such as the gums. Inulin and oligofructose are metabolised in the human gut in the same way as dietary fibre. Because they are not hydrolysed nor absorbed in the stomach and small intestine, they reach the colon where they are fermented almost completely by the resident microflora. Their energy value is consequently significantly lower than that of carbohydrates absorbed in the small intestine. Their fermentation in the colon affects stool output, gut microflora, as well as integrity of the mucosal barrier. They also induce systemic effects, e.g. on blood lipids.

Because of the natural occurrence of inulin and oligofructose in vegetables and fruits and because of their physiological effects, they have been considered as part of the dietary fibre complex and labelled as such. The classical methods of analysis for dietary fibre do not determine inulin or oligofructose, but accurate official methods (AOAC) do exist that allow their separate analytical determination in foodstuffs (Hoebregs, 1997). Several legal authorities have confirmed that inulin and oligofructose may be labelled as dietary fibre (Prosky 1998; Coussement 1999).

Inulin and oligofructose are resistant to hydrolysis and digestion in the upper part of the gastro-intestinal tract and are subsequently fermented in the colon. There is strong evidence in humans for an effect of inulin and oligofructose increasing fecal bulk and frequency, and promoting the selective growth of bifidobacteria at the expense of other micro-organisms. Inulin and oligofructose share many properties with the materials classically referred to as dietary fibre (Roberfroid 2000).

What is required now is exploration of these changes in the colonic function in relation to both colonic and systemic disorders and diseases for which laboratory studies already have indicated possible benefits. These areas include a.o. colon cancer, ulcerative colitis, intestinal infections, and possibly compromised immune function. A useful effect in any of these situations would amply justify continued interest in inulin and oligofructose as dietary ingredients (Jenkins et al. 1999).

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Monitor

In the following pages we offer you the latest nutritional information on chicory inulin and oligofructose, summarised from key articles in major scientific journals.

Prebiotics stimulate intestinal flora in preterm and term infants

Several recent studies have shown that the addition of prebiotic oligosaccharides to infant formula promotes the growth of bifidobacteria in the colon.

Knol et al. (2001) compared the proportion of bifidobacteria in the stool flora of term infants fed with a standard infant formula (SF) and those fed a formula with prebiotic fructo- and galacto-oligosaccharides and hydrolysed whey proteins (PBF). In the PBF-fed group, the frequency of bifidobacteria increased from 30.7% at 0-2 weeks to 59.4% at 6 weeks, whereas no significant change was observed (from 27.5% at 0-2 weeks to 31.7% at 6 weeks) in the SF-fed group. At 6 weeks the percentage of bifidobacteria in the PBF-fed group was closer to the level that has been reported for breast-fed infants (79%).(The bifidobacteria frequency is expressed as a percentage of total bacteria counts.)

Rigo et al. (2001) compared a new formula (NF), containing galacto- and fructo-oligosaccharides (0.4g/100ml), with data from breast-fed (BF) infants and infants fed SF. Fourteen healthy term infants obtained NF from birth to the age of 2 months. The growth and quality of growth of the NF-fed infants were similar to those seen in BF infants and SF-fed infants. The weight gain composition was 61% lean body mass, 37.2% fat mass and 1.5% bone mineral mass. Volumetric bone mineral density index decreased from 11.1 to 10.1. This slight decrease, due to a stronger increase in bone area than in bone mineral density during this period of rapid growth, was similar to results obtained with BF-fed and SF-fed infants. The average frequency of bifidobacteria in fecal samples was 21% at day 3, 53% at 3 weeks, 56% at 6 weeks and 50% at 9 weeks, indicating a rapid and significant bifidogenic effect of NF and the ability of NF to maintain a stable bifidobacterial flora.

These results were confirmed in a study by Moro et al. (2001). They investigated the dose dependence of this bifidogenic effect in 63 infants, who received a formula supplemented with 4g/l or 8 g/l galacto-oligosaccharides and fructo-oligosaccharides. The influence on fecal flora and stool characteristics (pH, frequency and consistency) was more pronounced with the 0.8 g/l supplement, but reached the level of significance only for stool consistency.

Boehm at al. (2001) investigated the bifidogenic effect of oligosaccharides (1g/dl) in 15 formula-fed preterm infants compared to a group fed fortified, pasteurised human milk. The oligosaccharide mixture was clearly bifidogenic and was accompanied by changes in stool characteristics to resemble those found in the group fed fortified, pasteurised human milk.

Knol J., Poelwijk E.S., van der Linde E.G.M., Wells J.C.K., Brönstrup A., Kohlschmidt N., Wirth S., Schmitz B., Skopnik H., Schmelzle H. & Fusch C. (2001). Stimulation of endogenous bifidobacteria in term infants by an infant formula containing prebiotics. Journal of Pediatric Gastroenterology and Nutrition, 32, 399.

Rigo J., Pieltain C., Studzinski F., Knol J. & Bindels J.G. (2001). Clinical evaluation in term infants of a new formula based on prebiotics, beta-palmitate and hydrolysed proteins. Journal of Pediatric Gastroenterology and Nutrition, 32, 402.

Moro G., Minoli I., Mosca F., Jelinek J., Stahl B & Boehm G. (2001). Dosage effect of oligosaccharides (OS) on fecal flora and stool characteristics in term infants. Journal of Pediatric Gastroenterology and Nutrition, 32, 401.

Boehm G., Casetta P., Lidestri M., Negretti M., Jelinek J., Stahl B. & Marini A. (2001). Effect of dietary oligosaccharides (OS) fecal bifidobacteria in formula fed preterm infants. Journal of Pediatric Gastroenterology and Nutrition, 32, 393.

Prebiotics stimulate vaccine-response of infants

This double-blind randomised controlled study examines the effects on the immune response after measles vaccination of an infant cereal with milk supplemented with a prebiotic mixture of chicory fructo-oligosaccharides and inulin (Prebio1).

Fifty infants 8 months old receiving a mixed feeding (breast, formula and solids) were given the cereals during a period of 10 weeks, one group with the supplement, a control group without. Four weeks after introduction of the cereals, all infants were vaccinated with the live attenuated measles vaccine. Post-vaccination IgG antibody levels were significantly higher in the supplement group (96% IgG positivity rate vs. 88% in the control group). Mild reactions were observed more often in the supplement group. No differences in growth or overal health status were observed.

Firmansyah A., Pramita G.D., Carrié Fässler A.L., Haschke F., Link-Amster H. (2000). Improved humoral immune response to measles vaccine in infants receiving infant cereal with fructo-oligosaccharides. Journal of Pediatric Gastroenterology and Nutrition, 31, S2, Abstract 521.

Effect of synbiotics on catch-up growth in preschool children

This double-blind, randomised study was designed to evaluate the incidence and duration of sickness in mild to moderately malnourished children who received a nutritional supplement with and without synbiotics (L. acidophilus, B. infantis and oligofructose), and to determine gastro-intestinal tolerance and growth. A total of 626 children 1 to 6 years of age in Brazil, Mexico, Spain and Portugal participated in the study. The children were 1 SD and 3 SD below the median of weight-for-height. Incidence and duration of sick episodes, antibiotic use, supplement intake and stool frequency and consistency were evaluated monthly for four months.

The number of days sick decreased significantly in both feeding groups. The decrease was more pronounced for younger (1-2 years) than for older children. Both groups experienced catch-up growth, with a significant increase in normalised percentiles for weight and height. For children aged 3-5 years with at least one sick episode, the number of days sick was lower in the group receiving synbiotics. The number of days of constipation were fewer in the synbiotics group.

Fisberg M., Maulen I., Vasquez E., Garcia J., Comer G.M. & Alarvon P.A. (2000). Effect of oral supplementation with and without synbiotics on catch-up growth in preschool children. Journal of Pediatric Gastroenterology & Nutrition, 31, S2, Abstract 987.

Inhibiting effect of inulin and oligofructose on cancer metastases

This study investigated the effect of 15% inulin (RAFTILINE®HP) or oligofructose (RAFTILOSE®P95) on lung metastases of transplantable liver tumours in mice. At 47 days after tumour transplantation, 59% of mice in the control group were bearing lung metastases, in comparison to only 36% in the inulin-fed group and 35% in the oligofructose-fed

group. The total number of lung metastases was 37 in the control group, 18 in the inulin-fed group and 6 in the oligofructose-fed group.

This inhibiting effect on the development of metastases may be explained by changes in the colonic microflora (increase of bifidobacteria), by decreased serum glucose and lipid levels (which may influence the cancer cells directly in such a way that they lose their energy, agressiveness and invasiveness). Inulin and oligofructose also may stimulate beneficial changes in the composition and structure of the basement membrane in capillary vessels, which is the most important barrier in the metastatic cascade, thus potentially inhibiting the development of metastases.

Taper H.S. & Roberfroid M.B. (2000). Inhibitory effect of dietary inulin or oligofructose on the development of cancer metastases. Anticancer Research, 20, 4291-4294.

Protective mechanisms of probiotics and prebiotics in the development of colon tumours

This article discusses the protective mechanisms of pro- and prebiotics in the development of colon tumours. One mechanism to explain the anti-carcinogenic effects of pre- and probiotics may be the detoxification of genotoxins in the gut. New animal studies showed that short-lived metabolite mixtures isolated from milk fermented with strains of Lactobacillus bulgaricus and Streptococcus thermophilus are more effective in deactivating etiologic risk factors of colon carcinogenesis than are cellular components of microorganisms. Ingestion of probiotics leads to the excretion of urine with low concentrations of components that are genotoxic in human colon cells and high concentrations of components that induce oxidised DNA bases. Ingestion of prebiotics results in a different spectrum of fermentation products, including the production of high concentrations of butyrate and other short-chain fatty acids. It has been shown that butyrate may inhibit the genotoxic activity of nitrosamides and hydrogen peroxide in human colon cells. There are many experimental observations regarding the protective effect of pro- and prebiotics against colon tumours, but in humans there is no evidence available on whether they can prevent the initiation of colon cancer. Therefore, further research is clearly needed to quantify any beneficial effects for prevention of human colon cancer.

Wollowski I., Rechkemmer G. & Pool-Zobel B.L. (2001). Protective role of probiotics and prebiotics in colon cancer. American Journal of Clinical Nutrition, 73 (Suppl.), 451S-455S.

Stimulation of apoptosis by chicory fructans in the rat colon

Hughes et al. studied apoptosis (programmed cell death) and bacterial metabolism as a possible mechanism involved in the protective effects of chicory oligofructose and long-chain inulin. They fed 18 rats for a 3-week period with either a basal diet (44% energy as fat), or a basal diet with 5% of oligofructose (RAFTILOSE,P95) or 5% of inulin (RAFTILINE,HP). All animals were then dosed with 1,2-dimethylhydrazine and sacrified after 24h. The mean number of apoptotic cells per crypt was significantly higher in the colon of rats fed chicory fructans compared to those fed the basal diet alone. The apopototic index was slightly higher in animals fed inulin. This suggests that the fructans exert protective effects at an early stage in the onset of cancer, as the supplements were effective soon after the carcinogen insult. No significant site-specific effect was evident, even though mean results showed that the apoptotic indices were higher in the distal colon from both dietary groups. There were no significant dietary effects on bacterial enzyme activities or ammonia concentration, despite a trend towards increased colonic beta-glucosidase and reduced ammonia concentration with the fructan-containing diet.

Hughes R. & Rowland I.R. (2001). Stimulation of apoptosis by two prebiotic chicory fructans in the rat colon. Carcinogenesis, 22, 43-47.

Non-digestible carbohydrates and carcinogenesis

This paper presents the implications for colorectal cancer of the effects on gut physiology and function of non-digestible carbohydrates (non-starch polysaccharides or dietary fibre, resistant starch and non-digestible oligosaccharides).

The intact carohydrates may have direct effects in the colon by increasing fecal bulk, decreasing transit time and binding substances such as bile acids and carcinogens that may have a preventive action on colorectal cancer. These direct effects are confined largely to the non-starch polysaccharides. Many of the effects of carbohydrates in the gut that may influence colon cancer are indirect effects on the gut microflora composition, the gut bacterial metabolism and the production of short-chain fatty acids. New studies are reported that show that the administration of non-digestible oligosaccharides and resistant starch may prevent DNA damage in the colon mucosa of animals.

Rowland I. (2001). Non-digestible Carbohydrates and Gut Function: Implications for Carcinogenesis. In: McCleary B.V. & Prosky L. (ed.), Advanced Dietary Fibre Technology, Blackwell Science Oxford, 226-231.

Inhibitory effect of inulin and oligofructose on colon carcinogenesis

This article reviews a number of recent animal studies on the potential inhibitory properties of inulin and oligofructose on colon carcinogenesis.

Chicory inulin and oligofructose added to the diet of rats leads to a significant reduction in the number of colonic aberrant crypt foci (ACF), which are considered to be early precursor lesions of colon tumours in rodents and humans. The degree of ACF inhibition was more pronounced in animals fed inulin, in comparison with oligofructose, which is probably due to the slower fermentation rate of inulin, allowing it to be fermented in the more distal part of the colon. The tumour-inhibitory effect seems to be dose-related. Also the time of administration may be an important factor: the inhibitory effect was more important when inulin was given during the promotion phase of carcinogenesis.

Several studies described a synbiotic effect on carcinogenesis when the administration of inulin or oligofructose was combined with Bifidobacterium longum.

The protective effect of fructans proceeds through the selective modulation of microflora, thus developing a favourable micro-environment in the gut. This may involve the modulation of specific bacterial enzymes such as beta-glucuronidase. Moreover, prebiotics increase the production of short-chain fatty acids in the colon, especially butyrate, by microbial fermentation.

The EU-funded SYNCAN project, which started in 2000, is investigating the potential colon cancer-preventing activity of pre- and synbiotics in human volunteers.

Franck A. (2000). The potential inhibitory properties of inulin and oligofructose on colon carcinogenesis. Agro-Food-Industry Hi-Tech, 11, 9-11.

Prebiotics and calcium absorption

The stimulation of mineral absorption by inulin-type fructans has been observed repeatedly in rat studies. The use of different models has shown an increase in the absorption of calcium, magnesium and iron at the level of the large intestine, as well as an increased

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calcium uptake into the bone tissue, resulting in improved bone mineral density. Inulin prevented ovariectomy-induced loss in the bone structure and therefore strengthened the bones.

These findings were confirmed in human experiments. Two earlier human feeding studies showed a significant positive effect, with an increase in Ca absorption of 26% (with 15g/day of chicory oligofructose in adults) and 58% (with 40g/day of chicory inulin in young adults). So far, positive effects on calcium absorption in humans seem to occur under conditions of increased calcium requirements. Long-term studies in humans are required to confirm the effects on skeletal development or bone health.

At least part of the stimulating effect of inulin and oligofructose on mineral absorption might be attributed to the production of short-chain fatty acids. The extent of the effect seems to be related to the type of carbohydrate, the rate of fermentation by the intestinal flora, and the ingested dose.

Franck (2000) reports a recent human study demonstrating that the intake of only 8g/day of enriched inulin (RAFTILOSE®Synergy1) during 3 weeks significantly increases the fractional and absolute Ca absorption by 18% and 90 mg/day, respectively, in adolescent girls.

Scholz-Ahrens K.E., Schaafsma G., van den Heuvel E. G.H.M. & Schrezenmeir J. (2001). Effects of prebiotics on mineral metabolism. American Journal of Clinical Nutrition, 73 (Suppl.), 459S-464S.

Franck A. (2000). Prebiotics and Calcium Absorption, Functional Foods 2000 Conference Proceedings, Leatherhead Publishing, 108-113.

Effects of fructans on lipid metabolism

The addition of 10% oligofructose (RAFTILOSE®P95) to the diet of rats can decrease lipogenesis in the liver by lowering the activity of key enzymes regulated only through modifications of gene expression. The mechanism by which such non-digestible nutrients modify hepatic metabolism remains to be clarified. In humans, some studies performed in normo- and hyperlipidemic patients showed a hypo-triglyceridemic effect of inulin, whereas other studies showed no effect. In the interpretation of human studies, one has to take into account the duration of the treatment, the dietary intake of carbohydrates compared with lipids, and the serum lipid composition at the beginning of treatment. Studies need to be performed in hyperglyceridemic patients, in whom lipogenic homeostasis could be disturbed, to determine the relevancy of fructans in decreasing lipogenesis in humans.

Delzenne N.M. & Kok N. (2001). Effects of fructan-type prebiotics on lipid metabolism. American Journal of Clinical Nutrition, 73 (Suppl.), 456S-458S.

Prebiotics and human health

This article gives the state-of-the-art and future perspectives concerning prebiotics and human health. It is well established that ingestion of prebiotics can elevate indigenous bifidobacterium and lactobacillus levels in the colon, and that they have the potential to improve health through the actions of fermentation end-products, including butyrate.

Further clinical trials are needed to demonstrate the positive effects on health of prebiotics and synbiotics. These studies would be strengthened by understanding mechanisms of function, and by the application of molecular tools for characterising alterations in the microbial profiles of the colonic microbes.

An ideal prebiotic could be developed for specific applications by using a check list that identifies the following parameters: (a) how rapidly the prebiotic will be utilised; (b) where in the tract it will be utilised; (c) to what extent it will be fermented; (d) which particular indigenous microbes will be enhanced and which will be suppressed; (e) identification of the fermentation products.

The development of novel prebiotics that are target and site-specific will allow added health benefits to be achieved. It is envisaged that poor health conditions that are linked to the indigenous microbes can be influenced by the application of prebiotics and synbiotics.

Conway P.L. (2001). Prebiotics and human health: the state-of-the-art and future perspectives. Scandinavian Journal of Nutrition, 45, 13-21.

Current state of knowledge concerning prebiotics

This review article presents the current state of knowledge concerning prebiotics, with emphasis on the criteria used for classification, mechanisms of selective growth stimulation and physiologic effects.

Of the currently available food ingredients, the non-digestible oligosaccharides are the only known components for which convincing evidence has been reported in favour of a prebiotic effect. The inulin-type fructans, inulin and oligofructose, are the prebiotics that have been investigated most extensively for their nutritional properties.

In pure culture, most species of bifidobacteria are adapted to the utilisation of these fructans even if other bacteria are capable of metabolising them. These studies with a single species of bacteria are of limited use unless their results are supported by studies using mixed cultures. Indeed, as many components of the gut microbiota as possible should be measured to indicate a true prebiotic effect. Simple stimulation of bifidobacteria is insufficient to demonstrate such an effect. New methods are being applied extensively to human gut microbiology and promise the degree of reliability required to detect subtle changes in colonic microflora composition, and to correlate such changes with health benefits. The potential applications of inulin-type fructans for reducing the risk of colon carcinogenesis as well as for improving calcium bioavailability and lipid homeostasis are of particular interest.

Roberfroid M.B. (2001). Prebiotics : preferential substrates for specific germs? American Journal of Clinical Nutrition, 73 (Suppl.), 406S-409S.

General properties of inulin and oligofructose

This article gives an overview of the production process and the general properties of inulin-type fructans. They can be used for either their nutritional advantages or technological properties, but they are often applied to offer a double benefit: an improved organoleptic quality (improved taste and texture) and improved nutritional composition (fat and sugar replacement). An overview is given of their applications in foods and drinks. Furthermore, the nutritional and physiological properties and the potential effects to the well-being and health (prebiotic effect, stool-bulking effect, protection against intestinal disorders and infections, improvement of calcium availability, reduction in serum triglycerides and insulin levels, inhibition of colon carcinogenesis in rats), are described.

Because of these properties, chicory inulin and oligofructose are legally classified as food or food ingredient (not as additive) in most countries. Most countries have further confirmed that they can be labelled as "dietary fibre" for food labelling.

Franck A.M.E. (2000). Inulin and Oligofructose. In: Gibson G. & Angus F. (ed.), LFRA Ingredients Handbook, Prebiotics and Probiotics, Leatherhead Publishing, 1-18.

Recent Scientific Research on Inulin and Oligofructose

Increased Calcium Uptake

Abrams presented the effect of a new enriched inulin (RAFTILOSE®Synergy1) on calcium absorption in healthy adolescent girls of near puberty (11-14 y.) receiving diets containing 1500 mg/day of calcium. The study used a randomised, double-blind, cross-over design. Calcium intake was increased by the addition of two daily servings of 240 ml of calcium-fortified orange-juice. For 3 weeks all subjects consumed 8g/day of RAFTILOSE®Synergy1 (Syn1), or oligofructose (RAFTILOSE®P95), or a placebo (sucrose), separated by a 2-week wash-out period. Calcium absorption was measured at the end of each 3-week study period.

Syn1 led to a significant increase in fractional calcium absorption (from 32.3% to 38.2%; a relative increase of 18%) and in total calcium absorption (+ 90mg/day), but this was not seen with oligofructose. There was no difference in urinary calcium excretion. If even part of this additional calcium was utilised for bone mineral production, it could lead to a significant increase in peak bone mineral density during this critical period. Assuming that this additional calcium could be retained daily during the 2 years of maximal adolescent bone mineral accumulation, the net increase in bone mineral mass indeed could be 65g, an average gain of 5.5% attributable to the consumption of RAFTILOSE®Synergy1.

The fact that oligofructose at the same dose did not induce a significant effect on calcium absorption suggests that the chain-length distribution of the fructan is of importance to obtain an optimal effect, especially at a rather low daily dose. Although a benefit was seen at all calcium absorption levels, the data suggest that the maximum benefit may occur in subjects with the lowest calcium absorption values.

Griffin I.J., Davila P.M. & Abrams S.A., Non-digestible oligosaccharides and calcium absorption in girls with adequate calcium intakes.

Prebiotics and Immunology in Children

The preliminary findings of several recent human clinical trials indicate that prebiotics may prove to be a useful dietary adjunct in the management of gastrointestinal and systemic conditions that are related to an adequate interaction between our microbial environment and our immunologic relationship to it. An important mechanism of these prebiotic agents is the induction of immunological responses from the gut-associated lymphoid system.

Saavedra presented the results of a recent controlled, double-blind, randomised, longitudinal study among 123 healthy children aged between 4 and 24 months attending day care centres. The children received a regular cereal or cereal supplemented with oligofructose (RAFTILOSE®P95; a daily average of 1.1g/day), for three months. All subjects exhibited normal growth, no significant differences were observed in the occurrence of flatulence or stool frequency and consistency. A significantly lower frequency of reported emesis, regurgitation and discomfort with bowel movements was detected in the supplemented group. No differences in incidence of loose stools or diarrhea were reported. However, significant differences were noted in the symptoms, medical visits and day care absenteeism associated with diarrhea.

Episodes of cold symptoms with fever, rhinorrhea and cough were reported less frequently in the supplemented group. Antibiotic prescription associated with respiratory illness also was significantly lower in the supplemented group.

These results suggest an ameliorating effect of the regular ingestion of a prebiotic added to a weaning food in this healthy population. The effect may be different in populations with different immunological status or different intestinal flora.

Saavedra J.M., Human studies with prebiotics: clinical and immunologic implications.

Probiotics and Prebiotics in Pediatrics

Breast-feeding is the Gold Standard for infant feeding. However, the majority of infants a few weeks old are fed with a bovine milk-based formula. One among the many differences between human and bovine milk is the influence on the development of the gastro-intestinal flora: the flora of breast-fed children being richer in bifidobacteria and lactobacilli. The absence of oligosaccharides, the third largest component in human milk, from bovine milk is likely to account for the differences in colonic flora. The oligosaccharide content and concentration in human milk varies continuously and many factors affect the final composition, making it impossible for industry to mimic nature. However, if the changes in chemical composition cannot be mimicked, the effect and function can be imitated.

The addition of lactobacilli to infant formula results in a gastro-intestinal flora that is dominated by lactobacilli, comparable to that in breast-fed infants. Also, some aspects of breast-fed infants such as aspects of stools change accordingly. The addition of probiotics to infant formula has been shown to decrease the incidence and severity of episodes of infectious diarrhea. The bacteria however need to be administered in a viable way, necessitating specific guidelines for formula preparation.

The prebiotic concept, in which oligosaccharides are added to the infant formula, seems a "more physiologic" approach and no special precautions or guidelines are needed for the preparation of the formula. The addition of galacto- and fructo-oligosac-

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charides to bovine milk-based infant formula has been shown to have a bifidogenic effect, i.e. it stimulates the growth of intestinal bifidobacteria and lactobacilli. The chain length of inulin and oligofructose is of importance, since the longer it is, the more sustained the fermentation pattern becomes.

Vandenplas Y., Oligosaccharides in infant formula.

Non-digestible Oligosaccharides and Defense Functions

Experiments have shown that changes in the populations of the gastro-intestinal bacteria of laboratory rodents in response to feeding diets with non-digestible oligosaccharides (NDO) modulate immune system functions and increase resistance to carcinogens that promote colorectal cancer and to challenges with pathogenic organisms.
Buddington examined if and how fructans and other NDO enhance defense functions. Several studies showing that fructans enhance the resistance to health challenges were discussed, including resistance to lumenal pathogens, resistance to noxious chemicals in the diet (drugs, pesticides, etc.), resistance to carcinogens and reduced growth of spontaneous and transplantable tumours, enhancement of the mucosal barrier, improved surviving after systemic challenges with pathogens, and better recovery of disturbed gastro-intestinal tract (GIT) ecosystems.
The review also examined the defense mechanisms and responses to fructans and dietary fibres. The research with animal models has revealed three lines of host defense. The first is the assemblages of bacteria resident in the different regions of the GIT. The second is the multilayered mucosal barrier that acts as selective filter to exclude pathogens, hazardous chemicals and other potential challenges to health. The third consists of the systemic mechanisms that must recognise and eliminate any potentially harmful organisms or chemicals that manage to pass the mucosal barrier.
The lactic acid-producing bacteria (LAB) are considered to be immuno-modulatory and influence the GIT and systemic defense functions directly and indirectly. Corresponding with this, supplementing the diet with fructans or other NDO that increase the densities and metabolic capacities of the LAB enhances defense mechanisms of the host, increases resistance to various health challenges and accelarates recovery of the GIT after disturbances.

Buddington R.K., Kelly-Quagliana K., Buddington K.K. & Kimura Y., Non-digestible Oligosaccharides and Defense Functions: Lessons Learned from Animal Models.

Anti-cancer Properties of Inulin and Oligofructose

Recent research with experimental animal models revealed that chicory inulin and oligofructose added to the diet in concentrations of 5% to 15% had significant anti-carcinogenic properties. They prevent formation of azoxymethane (AOM)-induced aberrant crypt foci and tumours in the colon. The effects are dose-related. Inulin is active in the promotion phase, but more active when present during initiation and promotion. Ongoing studies are now directed at elucidating whether the combination of long and short-chain inulin, each putatively fermented in different portions of the large gut, could have even more efficient protective effects. The protective effect is enhanced by additional supplementation with bifidobacteria. This type of synbiotic treatment is also effective when the bifidobacteria and the prebiotics are given after the initiation phase, also indicating a suppressive effect.
Studies with transgenic min-mouse containing a nonsense mutation in the murine APC-gene have shown that inulin and oligofructose may also modulate the occurrence of colon tumours that are not chemically induced.
These effects may be due to the stimulation of bifidobacteria and the production of lactate, butyrate and acetate, which have been shown to be anti-genotoxic in the colon and to reduce AOM-induced tumours. Butyrate has several biochemical/toxic effects in tumour cells mediated by altering gene expression. It inhibits cell proliferation, it may induce apoptosis of already transformed cells, it modulates GST/glutathione expression and modifies histone acetylation. It may also increase secretion of mucin, a barrier that can deactivate carcinogens, thus protecting the epithelial cells.
There is some evidence that pro- and prebiotics also may inactivate carcinogens in the colon by modification of toxifying and detoxifying enzymes, such as glutathione-S-transferase (GST). In order to confirm the experimental data in humans and to obtain more insight into these effects in the colon, dietary intervention studies relating biomarkers of reduced risk to inulin consumption are needed.
That is the aim of the SYNCAN project, a three-year EU-funded research project which started in March 2000; eight leading teams from six EU countries are collaborating with ORAFTI as coordinator.
As first action, various synbiotic combinations will be investigated using in vitro fermentation techniques in order to identify which offer the greatest competitive advantages in the colonic ecosystem, especially in distal parts of the colon where most colon cancers occur. Various mechanistic hypotheses will be tested.
The anti-carcinogenic effects of a specific synbiotic (10% RAFTILOSE®Synergy1 combined with 108/g Lactobacillus GG and 108/g Bifidobacterium BB12) will be studied in an animal model for colon carcinogenesis over a 1 year period. Three types of biomarkers will be quantified: immunological, genetic and cytotoxic ones. Central to the SYNCAN project will be a dietary intervention study with human subjects: Dukes B cancer patients and polypectomised volunteers, using the different biomarkers. Volunteers of each group will be given either a placebo or the test synbiotic for a period of 6 months.

Pool-Zobel B., Van Loo J., Rowland I. & Roberfroid M, Review on experimental evidence investigating the potential of the prebiotic carbohydrates inulin and oligofructose to reduce the risk of colon cancer.

Van Loo J., Synbiotics and Reduction of Colon Cancer Risk in Humans: The SYNCAN Project. ■

This page is entirely reserved for your FAQs. It is meant to give answers to the most important questions that may arise.

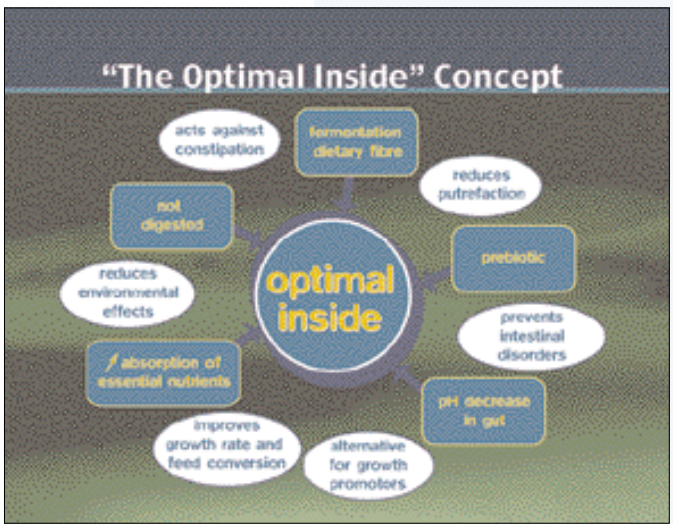
Do inulin and oligofructose have benefits for use in animal feed ?

The simple answer is yes. There are several good reasons why this is the case, which we have summarised as the "Optimal Inside" concept. This concept is based on the findings of a research program initiated by ORAFTI, which has already brought us very encouraging data.
These prebiotic feed ingredients are not digested in the gastro-intestinal (GI) tract. Monogastric animals, which include swine, poultry, rabbits, horses, calves, dogs and cats, do not have intestinal enzymes capable of digesting inulin or oligofructose, which are fermented by the microflora that inhabit the GI tract. Research by ORAFTI has demonstrated that the place where fermentation occurs depends very much on the animal species, its life-stage and the type of prebiotic present in the feed. In domestic pets (dogs, cats), the fermentation happens mainly in the large intestine. In young pigs, however, oligofructose is fermented completely before the gut contents reach the colon whereas fractions of inulin survive passage through the ileum.
The major end products of fermentation, mainly by Lactobacillae and Bifidobacteria, are gases and short-chain fatty acids. The release of these acids into the intestinal lumen lowers the pH of the gut, making it less suitable for the growth of pathogenic micro-organisms. As this happens in the ileum with certain types of animal, like swine, calves and poultry, it will make these animals less susceptible to bacterial infections. At the same time, these circumstances contribute to a better availability of minerals and a better digestibility of proteins in the digestive tract. All these factors together can result in an improved growth-rate and feed conversion.

Today, a lot of organic acids are used in feed formulations in order to achieve a lower pH in the gut and thus a better digestibility of the feed. Prebiotics offer the advantage that they are fermented gradually during their passage through the digestive tract, and will thus steadily release organic acids. Prebiotics could partly replace or be added together with organic acids in order to obtain similar effects. They both act as a preventative agent against the development of pathogenic bacteria and can be considered as a valid and natural alternative to the use of antibiotics.
For other types of animal, like dogs and cats, the benefits are derived mainly from the effects in the large intestine. The boost of saccharolytic bacteria at the expense of the proteolytic flora results in a lower formation of putrefactive and toxic substances and thus leads to a healthier gut. At the same time, the consumption of inulin and oligofructose leads to a reduced stool volume compared to that resulting from the use of traditional fibre sources such as beet fibre.

We have developed a range of products for animal feed use, called RAFTIFEED®. These products consist of inulin and oligofructose with different chain lengths and variable chain-length profiles. Our research program is exploring which prebiotic is most effective and efficient in the specific digestive system of individual species.

In conclusion, the challenge for the future is not so much the need to demonstrate the positive effects of RAFTIFEED® prebiotics on animals, as this is already confirmed by convincing data from recent research, but the need to find the right product and right dosing regime for different species at different life-stages, and to demonstrate the economic justification for incorporation in animal feed. ■



Agenda

Disneyland Paris, France
October 17-19, 2001

International Symposium on Functional Foods, Scientific and Global Perspectives

Organised by International Life Sciences Institute – ILSI Europe.

The objectives of the meeting are :

• to review current world view on the scientific basis of funtional foods and to identify areas of agreement and disagreement,

• to identify unifying concepts and illustrate with relevant examples,

• to review current scientific support for biomarkers to link functional food consumption to quality of life and/or health,

• to review the communication requirements from a scientific, consumer and regulatory point of view,

• to identify new trends in functional food science.

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functional.sympo@ilsieurope.be
Website: www.ilsi.org/europe.html

Malle (Antwerp), Belgium
November 13, 2001

Do we eat healthy ? Bioactive Substances in our Food

Organised by Working Group KVCV-Food.

Speakers include :

Ir Wim Caers – ORAFTI :

Non-digestible oligosaccharides.

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Fax: +32/ 3 820 27 34
E-mail: labrom@uia.ua.ac.be

London, United Kingdom
November 5-7, 2001

Food Ingredients Summit

Organised by United Business Media International

Topics to be discussed include:

• encapsulating and coating,
• taste and flavours,
• replacer ingredients,
• processing technologies,
• texture,
• food safety and quality,
• consumer awareness.

The Master Class session will explore the impact of the internet on the relationship between suppliers, manufacturers, retailers and consumers.

Info : Miller Freeman BV
PO Box 200
3600 AE Maarssen
The Netherlands
Tel : +31 / 346 559 444
Fax : +31 / 346 573 811
Website : www.fi-events.com

The Hague, The Netherlands
March 5-7, 2002

Functional Foods 2002, Europe's Premier Conference on Science, Technology and Marketing

Organised by Leatherhead Food RA & BV Industrial Promotions International

Topics to be discussed in the workshops include:

• cardiovascular health,
• gut health,
• women's/men's health,
• bone health,
• mental health,
• immune function.

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Colophon

Active Food Scientific Monitor is published by ORAFTI, a daughter company of RAFFINERIE TIRLEMONTOISE (B), which is part of the SÜDZUCKER Group (D).
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