

## Applications of inulin and oligofructose in health and nutrition

NARINDER KAUR\* and ANIL K GUPTA

Department of Biochemistry and Chemistry, Punjab Agricultural University, Ludhiana 141 004, India

\*Corresponding author (Fax, 91-161-400945; Email, nkaur@rediffmail.com)

Inulin and oligofructose belong to a class of carbohydrates known as fructans. The main sources of inulin and oligofructose that are used in the food industry are chicory and Jerusalem artichoke. Inulin and oligofructose are considered as functional food ingredients since they affect physiological and biochemical processes in rats and human beings, resulting in better health and reduction in the risk of many diseases. Experimental studies have shown their use as bifidogenic agents, stimulating the immune system of the body, decreasing the levels of pathogenic bacteria in the intestine, relieving constipation, decreasing the risk of osteoporosis by increasing mineral absorption, especially of calcium, reducing the risk of atherosclerosis by lowering the synthesis of triglycerides and fatty acids in the liver and decreasing their level in serum. These fructans modulate the hormonal level of insulin and glucagon, thereby regulating carbohydrate and lipid metabolism by lowering the blood glucose levels; they are also effective in lowering the blood urea and uric acid levels, thereby maintaining the nitrogen balance. Inulin and oligofructose also reduce the incidence of colon cancer. The biochemical basis of these beneficial effects of inulin and oligofructose have been discussed. Oligofructoses are non-cariogenic as they are not used by *Streptococcus mutans* to form acids and insoluble glucans that are the main culprits in dental caries. Because of the large number of health promoting functions of inulin and oligofructose, these have wide applications in various types of foods like confectionery, fruit preparations, milk desserts, yogurt and fresh cheese, baked goods, chocolate, ice cream and sauces. Inulin can also be used for the preparation of fructose syrups.

[Kaur N and Gupta A K 2002 Applications of inulin and oligofructose in health and nutrition; *J. Biosci.* 27 703–714]

### 1. Introduction

About 15% of flowering plant species store fructans as a reserve in at least one of their organs during their life cycle (Hendry 1993). Fructan, in general is a term used for any carbohydrate in which fructosyl-fructose links constitute the majority of the glycosidic bonds. Fructans are linear or branched fructose polymers, which are either **b** 2 → 1 linked inulins or **b** 2 → 6 linked levans. Dicotyledonous species store inulin type fructans consisting of linear **b** (2 → 1) fructofuranosyl units. More complex and branched type fructans are common in monocots (Vijin and Smeeckens 1999).

### 2. Synthesis of fructans

Because fructans are synthesized from sucrose by repeated fructosyl transfer from a fructosyl donor, both inulins and levans usually, but not always, have a terminal glucose unit. The enzyme generally considered to be involved in plant fructan synthesis is sucrose-sucrose fructosyl transferase (EC 2.4.1.99) which catalyses the transfer of a fructose molecule from one sucrose molecule to another, leading to kestose formation (glucosyl-1, 2 fructosyl-1, 2 fructose). Chain elongation is mediated by either 1<sup>F</sup>- or 6<sup>F</sup> fructan-fructan-fructosyl (EC 2.4.1.100) transferase leading to inulin and levans respectively.

**Keywords.** Fructan; inulin; oligofructosaccharides; oligofructose

Abbreviations used: ACF, Aberrant crypt foci; AOM, Azoxymethane; CoA, coenzyme A; DP, degree of polymerization; GIP, glucose-dependent insulinotropic peptide; GLP-1, glucagon like peptide; LDL, low-density lipoprotein; ODC, ornithine decarboxylase; 1Q, 2-amino-3-methyl imidazo (4,5-F) quinoline; SFr, synthetic fructan; TAG, triacylglycerol.

Readers may consult Gupta and Kaur's review (2000) for further details on fructan synthesis.

Bacterial fructans are synthesized without a trisaccharide as an intermediate, but by sequential transfer of fructose from a fructosyl donor to the growing levan chains by levan sucrose (sucrose<sup>F</sup>6-fructosyl transferase) (Smeekens *et al* 1991). They have a much higher degree of polymerization (DP, up to 100,000) than plant inulins (up to 150).

### 3. Plant sources of fructans

Fructan containing plant species are found in a number of mono and dicotyledonous families such as *Liliaceae*, *Amaryllidaceae*, *Gramineae*, and *Compositae*. Parts of various fructan containing plant species like asparagus, garlic, leak, onion, Jerusalem artichoke, chicory, etc. are often eaten as vegetables (Van Loo *et al* 1995). Some important sources of inulin (together with their average inulin content) are given in table 1. However, only a limited number of species are suitable for industrial food and non food applications. Despite the high fructan content of the aerial parts of many gramineae, particularly of young seedlings (up to 70% of their dry wt.), grasses and cereals are not used for industrial extraction and processing of fructans. Conversely, in *Liliaceae*, *Amaryllidaceae* and *Compositae*, fructans (most exclusively inulins) are usually stored in organs such as bulbs, tubers and tuberous roots which because of the absence of interfering components, can be easily extracted and processed to purified products. The two species currently used by the industry to produce inulin belong to *compositae*: Jerusalem artichoke (*Helianthus tuberosus*) and chicory (*Cichorium intybus*), the latter being by far the most commonly used source (Debruyne *et al* 1992). Inulin is processed by the food industry to produce either short chain fructans, namely oligofructose (DP, 2–10; average 5) as a result of partial enzymatic (endo-inulinase EC 3.2.1.7) hydrolysis, or long chain fructans by applying industrial physical separation techniques (De Leenheer 1996). In addition to chicory inulin and its hydrolysate, the food industry also

produces a synthetic fructans. Basically they are sucrose molecules to the fructose units of which, 1, 2 or 3 additional fructose units have been added by **b**-(2, 1)-glycosidic linkage, using the transfructosyl activity of **b**-fructosidase, from *Aspergillus niger* (Crittenden and Playne 1996; Fishbein *et al* 1988). Such kind of fructans have also been reported in the medium of *Fusarium oxysporum* (Gupta and Bhatia 1980).

The average daily consumption of inulin and oligofructose has been estimated to be 1–4 g in the United States and 3–11 g in Europe (Van Loo *et al* 1995). However, no such study has been made in India. Chicory root is best known for its use as a coffee substitute. In chicory, inulin is stored as a reserve carbohydrate in the fleshy tap root and constitutes about 70–80% of root dry weight (Gupta *et al* 1985). An interesting thing about chicory is that depending upon the stage of its development, we can obtain either inulin or oligofructose, the latter being formed after hydrolysis of inulin by inulinase, after the emergence of inflorescence axis, which takes place after the root has fully developed (Kaur *et al* 1992). Both inulin and oligofructose can be further hydrolysed to fructose by exoinulinase. Use of a powerful exoinulinase can replace the multienzymic steps of producing fructose from starch (Gupta and Kaur 1997). Thus, chicory roots are a concentrated source of inulin and oligofructose. Inulin and oligofructose are officially recognized as natural food ingredients in most European countries and have a self-affirmed generally regarded as safe (GRAS) status in the United States. However, the synthetic fructans have been classified as a 'novel food' (EU Regulation on Novel Foods and Novel Food Ingredients 258/97) by the ad hoc authorities of the European Commission.

### 4. Inulin and oligofructose are prebiotics

A prebiotic is a non digestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon, that can improve the host's health (Gibson and Roberfroid 1995). In addition, a prebiotic may repress the growth of pathogens for overall beneficial health (Roberfroid 2001). Inulin and oligofructose cannot be digested except through bacterial activity; they can alter the composition of human gut flora by a specific fermentation which results in a community predominated by bifidobacteria (Hidaka *et al* 1986; Wang and Gibson 1993). Menne *et al* (2000) reported that Fn type of fructans had a similar prebiotic effect in humans as reported with GFn type oligofructose.

Because of the **b**-configuration of the anomeric C<sub>2</sub> in their fructose monomers, these fructans are resistant to hydrolysis by human digestive enzymes (**a**-glucosidase,

**Table 1.** Percent inulin content on fresh weight basis from some important sources.

Source	Inulin (%)
Garlic	15–20
Asparagus root	10–15
Salisfy	15–20
Jerusalem artichoke	15–20
Dahlia tubers	15–20
Chicory root	15–20

Data taken from Gupta and Kaur (1997).

maltase, isomaltase, sucrase) which are specific for  $\alpha$ -glycosidic bonds, and are thus classified as non-digestible oligosaccharides on the basis of both *in vitro* and *in vivo* data. Since the stomach hydrolysis of (inulin type) fructans is of limited physiological significance, these products pass undigested through the upper part of the gastrointestinal tract into the colon. This has been confirmed by *in vivo* studies of rats and humans. The most convincing studies were done with the use of an ileostomy model which provides a valuable alternative to the study of digestive physiology in man and which has often been used to quantify the small intestinal excretion of nutrients, carbohydrates in particular. It was found that 86–88% of the dose (10, 17, 30 g) of inulin and oligofructose was recovered in the ileostomy effluent which supported the idea that inulin and oligofructose are practically indigestible in the small intestine of man (Coudray *et al* 1997).

The small loss during the passage through the small intestine could be due to fermentation by the microbial population colonizing the ileum, a population known to be 100 times greater in the ileostomists than in normal individuals. The estimation of lactic acid and short chain carboxylic acids, the end products of the anaerobic fermentation of carbohydrates in the ileostomy effluents before and after inulin intake, explained the 12–14% loss of fructans in the small intestine. Another explanation was acid and/or enzymatic hydrolysis of the low molecular weight fructans, as it has been shown that they are more sensitive to stomach and/or small intestinal hydrolysis than the high molecular weight components (Coudray *et al* 1997).

### 5. Inulin and oligofructose are bifidogenic

Predominance of bifidobacteria in the large intestine is essential for the prevention of many diseases and for maintaining good health. One main strategy is the prebiotic approach – the use of selective carbohydrate substrates in the diet for the growth of indigenous bifidobacteria. To be effective, these carbohydrates must reach the colon undigested and unabsorbed in the upper gastrointestinal tract and be selectively utilized by the bacteria present there. Inulin and oligofructose are examples of such carbohydrates. The large intestine is most heavily colonized with up to  $10^{12}$  bacteria for every g of gut content. It has been demonstrated by *in vivo* and *in vitro* studies that in humans, fermentation of fructans leads to the selective stimulation of growth of the bifidobacteria populations (Gibson and Wang 1994; Gibson *et al* 1995; Roberfroid *et al* 1995; Cummings *et al* 2001). Hussein *et al* (1999) observed that oligofructose supplementation increased the number of bifidobacteria in dog faeces.

The studies of Gibson *et al* (1995) showed that oligofructose and inulin significantly modified the *in vivo* composition of the microbiota by stimulating the growth of bifidobacteria. In addition, ingestion of oligofructose significantly reduced the count of bacteroids, fusobacteria and clostridia (table 2). Similar human studies in adult European, Japanese and North American populations have been reported for inulin using different daily doses (Hidaka *et al* 1986; Buddington *et al* 1996; Kleessen *et al* 1997). It has been suggested that the beneficial effect of inulin could be due to the ability of bifidobacteria to change the colonic environment by inhibiting detrimental bacteria via the formation of bacteriocins, the successful competition for substrates or adhesion sites on the gut epithelium, and stimulation of the immune system (Miller-Catchpole 1989; Gibson *et al* 1995). Inulin and oligofructose also help in the absorption of certain ions and the synthesis of B-vitamins (Gibson and Roberfroid 1995). The positive effects of bifidobacteria on human health are summarized in figure 1.

### 6. Physiological effects of products of fermentation of inulin and oligofructose in the gastrointestinal tract

Colonic fermentation of inulin and oligofructose produces short chain carboxylic acids (acetate, butyrate and propionate), lactate and gases as products of digestion (Gibson *et al* 1995). These fructans are low energy food ingredients. Their energy content is only 40–50% of that of digestible carbohydrates, giving them a caloric value of 1.0–2.0 Kcal/g. Both inulin and oligofructose are used in the diet of obese persons. Up to 95% of the acids produced in the colon are absorbed, mainly in the ascending part of the colon.

From animal *in vivo* studies it was concluded that supplementing the diet with inulin type fructans decreased the cecal pH and increased the size of the cecal pool of short chain carboxylic acids, with acetate being the primary acid followed by butyrate and propionate. Moreover, butyrate is produced in higher concentration in inulin-fed

**Table 2.** Effect of feeding 15 g/day of sucrose, oligofructose and inulin on percent composition of intestinal microflora in humans.

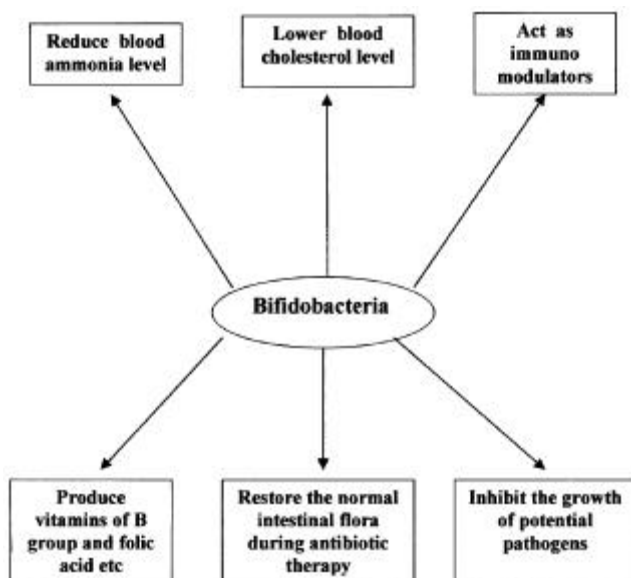
Microflora	Feed		
	Sucrose	Oligofructose	Inulin
Bacteroids	72	16	26
Bifidobacteria	17	82	71
Clostridia	2	1	0.3
Fusobacteria	9	1	3

Data from Gibson *et al* (1995).

than in control rats (Levrat *et al* 1991; Remesy *et al* 1992; Roland *et al* 1993; Younes *et al* 1996; Campbell *et al* 1997). Possibly, the increase in the pool size of these acids is related to the effect of the fructans on intestinal tissue, leading to hyperplasia of the mucosa and increased wall thickness, both in the small intestine (Oku *et al* 1984) and in the cecum (Campbell *et al* 1997), is accompanied by an increase in blood flow (Remesy *et al* 1993).

### 7. Effect of inulin and oligofructose on constipation

Constipation is an ailment encountered in elderly people. Many factors contribute to the development of constipation with aging, such as changes in diet and fluid intake, decline in the consumption of fibre-containing products, intake of drugs or laxatives, decrease in intestinal motility, and physical inactivity. Several studies in humans suggest that fermentation of carbohydrates stimulates colonic motility (Roberfroid 1993). Hidaka *et al* (1991) observed that the administration of oligofructose relieved constipation. Inulin ingestion improved constipation in 9 of 10 subjects. Abdominal discomfort, mainly flatulence, was reported rarely, and by only a few patients (Kleessen *et al* 1997). A significant increase in stool frequency was observed in healthy volunteers having one stool every 2–3 days by including inulin with DP more than 25 in the diet (Hond *et al* 2000).



**Figure 1.** Potential health promoting properties associated with bifidobacteria.

### 8. Effect of inulin and oligofructose on mineral absorption

A remarkable increase in the body absorption of calcium, a nutrient that helps to build and maintain structure of teeth and bones has been shown by ingredients extracted from chicory roots. Chicory inulin, which acts as a soluble fibre, increased the intake of calcium in the bone tissue which resulted in improved bone mineral density thus showing that it has the potential to prevent or postpone osteoporosis. Several investigators have demonstrated that rats fed with oligofructose and/or inulin absorbed more calcium and magnesium than control rats, despite an increase in total fecal mass (Delzenne *et al* 1995; Ohta *et al* 1994). Chronic ingestion of inulin or oligofructose decreased, or prevented the loss of bone calcium and phosphorus from the bones of gastrectomized rats (Ohta *et al* 1994), and the loss of bone mineral density by ovariectomized rats (Taguchi *et al* 1994).

Increased calcium absorption could be due to its increased availability by transfer of calcium from the small intestine into the large bowel and the osmotic effect of inulin and oligofructose that transfers water into the large intestine, thus allowing it to become more soluble (Carabin and Flamm 1999). Improved calcium availability in the colon could also be as a result of hydrolysis of calcium phytate complex by bacterial phytase releasing calcium (Lopez *et al* 2000). The improved absorption was associated with decreased pH of ileal, cecal and colonic contents, resulting in an increased concentration of ionized minerals: a condition that favours passive diffusion, hypertrophy of cecal walls and increased concentration of volatile fatty acids, bile acids, calcium, phosphorus, phosphate and to a lesser extent magnesium in the cecal contents (Levrat *et al* 1993). It was reported that although ingestion of oligofructose improved calcium and magnesium absorption in normal rats, only magnesium absorption was increased in cecectomized rats. This suggested that the effect of fermentation in the cecum was particularly important for calcium absorption (Ohta *et al* 1994). Coudray *et al* (1997) noted that inulin improved the absorption of calcium but not of magnesium, iron and zinc in humans. Van den Heuvel *et al* (1999) also reported an increased absorption of calcium in adolescents on oligofructose ingestion. Mechanisms by which ingestion of non-digestible carbohydrates improves mineral absorption is not very clear.

### 9. Influence of inulin and oligofructose on glycemia/insulinemia

The effect of inulin and oligofructose on glycemia and insulinemia are not yet fully understood, and available data are sometimes contradictory, indicating that these

effects may depend on physiological (fasting versus postprandial state) or disease (diabetes) conditions. Oligofructose given at the dose of 10% in the diet of rats for 30 days, reduced postprandial glycemia by 17% and insulinemia by 26% respectively. In rats fed 10% SFr for 3 months, the glycemic response to saccharose or maltose was reduced, possibly due to reduction of disaccharidase activity in the gastrointestinal tract (Oku *et al* 1984). In streptozotocin treated rats (diabetic), ingestion of a diet containing 20% oligofructose for 2 months decreased postprandial glycemia, despite a lack of modification of the glycemic or insulinemic response to a saccharose or maltose load. Similarly chronic ingestion of SFr (20 g/day for 4 weeks) did not modify fasting plasma glucose and insulin in healthy human volunteers, even if it lowered basal hepatic glucose production (Luo *et al* 1996). However, in diabetic subjects, taking 8 g of SFr/day for 14 days led to a decrease in fasting blood glucose (Yamashita *et al* 1984). When 10 g of artichoke inulin was added to 50 g of wheat-starch meal in healthy human subjects, the blood glycemic response was lower, despite no apparent interference by inulin on starch absorption (Rumessen *et al* 1990).

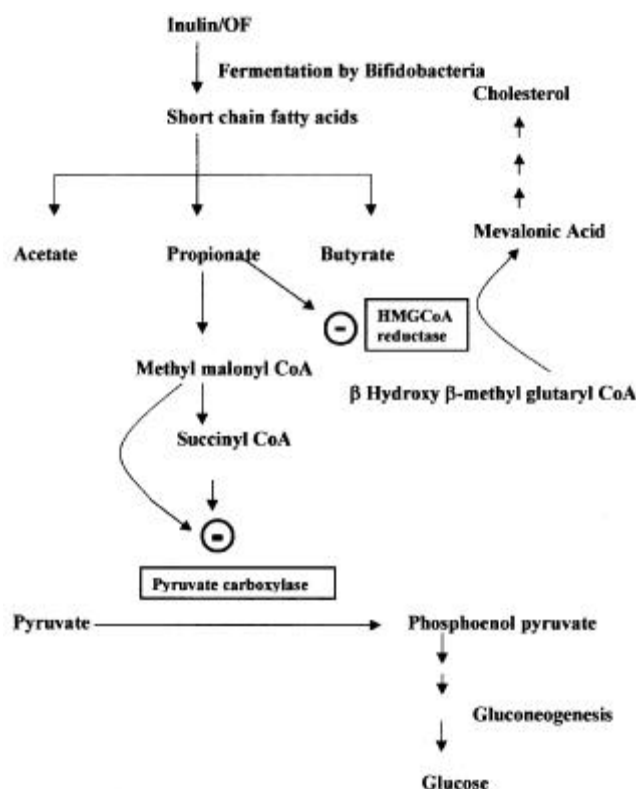
When rats are fed with 10% and 20% SFr in their diet for 6 weeks tests showed that mouth to anus transit time was shortened by 25 and 50%, respectively. This suggested a dose-dependent effect (Oku *et al* 1984; Tokunaga *et al* 1986). Possibly like other dietary fibres, inulin and oligofructose influence the absorption of macronutrients, especially carbohydrates, by delaying gastric emptying and/or shortening small-intestinal transit time.

The lower fasting glycemia has been reported in normal subjects fed with SFr (Luo *et al* 1996). The reduced hepatic gluconeogenesis induced by inulin and oligofructose intake could be mediated by the short-chain carboxylic acids, especially propionate. Propionate given in the diet of rats for 4 weeks reduced fasting blood glucose (Boillot *et al* 1995). Propionate also inhibited gluconeogenesis in isolated hepatocytes, probably via its metabolic conversion into methylmalonyl-coenzyme A (CoA) and succinyl-CoA, both of which are specific inhibitors of pyruvate carboxylase (Baird *et al* 1980) as depicted in figure 2. In addition, propionate enhanced glucose utilization by depleting hepatic citrate, an allosteric inhibitor of phosphofructokinase. Propionate may also influence hepatic glucose metabolism indirectly by lowering plasma fatty acid concentration, a factor known to be closely related to gluconeogenesis (Lee *et al* 1996).

### 10. Effect of dietary oligofructose on lipid metabolism

Inclusion of chicory roots (mainly inulin) in the diet of saturated fat fed rats significantly reduced the high

triglyceride content of blood and liver (Kaur *et al* 1988). A hypolipidemic effect was also observed in the livers of dexamethasone and ethanol-injected rats (Kaur *et al* 1988, 1989). Delzenne and Kok (1998) suggested that triacylglycerol (TAG) lowering effect of oligofructose occurs via reduction in very low density lipoprotein (VLDL)-TAG secretion from the liver as a result of the reduction in the activity of lipogenic enzymes, and in the case of fatty acid synthase via modification of lipogenic gene expression. Oligofructose decreased serum TAG when it was included in the standard, fibre free or high fat diet of rats. Addition of oligofructose in a carbohydrate rich diet reduced the *de novo* liver fatty acid synthesis (Delzenne and Kok 1999). Studies on incorporation of [<sup>14</sup>C] acetate into TAG in hepatocytes isolated from control and oligofructose-fed rats also supported the above results (Kok *et al* 1996). Hepatic glycerol-3-phosphate concentrations were significantly higher in oligofructose fed rats than in controls. This relative increase in glycerol-3-phosphate content of the liver might be due to its decreased utilization for fatty acid esterification. Indeed, the administration of oligofructose slightly but significantly, reduced hepatocyte capacity to esterify [<sup>14</sup>C] palmitate into TAG (Fiordaliso *et al* 1995).



**Figure 2.** Inhibition of pyruvate carboxylase by methyl malonyl CoA and succinyl CoA, and HMG CoA reductase by propionate. ⊖, inhibition.

Fatty acid availability is considered to be the rate limiting factor for TAG synthesis under physiological conditions (Stals *et al* 1994). The hypothesis of an increased fatty acid oxidation to explain a lower availability is unlikely as the activity of carnitine palmitoyl transferase I (CPTI), the rate-limiting enzyme in the hepatic mitochondrial *b*-oxidation, was not increased in the liver of oligofructose fed rats (MacGarey *et al* 1978). This is supported by the observation that mitochondria-enriched fraction prepared from the liver of oligofructose-fed rats had the same capacity to oxidize various fatty acids as those prepared from control rats (Kok *et al* 1996).

The activities of lipogenic enzymes were found to be lower in the livers of oligofructose fed rats compared with controls, suggesting that oligofructose feeding could decrease lipogenic flux and thus liver VLDL-secretion capacity (Arbeeney *et al* 1992; Gibbons 1990). The depression of the activity of all lipogenic enzymes and fatty acid synthase mRNA suggested that oligofructose modifies the gene expression of lipogenic enzymes (Hillgartener *et al* 1995).

Glucose-dependent insulinotropic peptide (GIP) and glucagon like peptide (GLP-1) are known to regulate postprandial insulin release and also to have direct insulin like actions on lipid metabolism (Morgan 1996). Serum GIP concentrations were more in oligofructose fed rats. Oligofructose also increased GLP-1 two-fold in the cecum of oligofructose fed rats as compared to the control. The latter increase is a consequence of cecal hypertrophy and might be related to the trophic effects of short chain fatty acids produced by oligofructose fermentation in the cecocolon (Roberfroid 1993). If the observations with dietary oligofructose or similar compounds could be confirmed in humans it would have profound importance, because hypertriglyceridemia is a known risk factor for coronary heart disease (Davignion and Cohn 1996). Daubioul *et al* (2000) reported that dietary enrichment with oligofructose can counteract both the fat mass development and hepatic steatosis in obese rats. After 10 weeks of oligofructose feeding, a significant reduction in the activity of malic enzyme in the liver was observed in these rats.

### 11. Effect of inulin and oligofructose on lipid parameters in humans

Attempts to reproduce similar effects of inulin and oligofructose in humans as observed in rats have generated conflicting results. This may be because of the much lower doses used in humans as a result of the adverse gastro-intestinal symptoms exhibited by most subjects consuming daily doses in excess of 30 g of inulin. Two studies that fed either oligofructose (20 g/d) or inulin

(14 g/d) observed no effect on fasting total, low density lipoprotein (LDL) or high density lipoprotein (HDL) cholesterol, or serum triglycerides. Two other studies that fed inulin either in a breakfast cereal (9 g/d), or as a powdered addition to beverages and meals (10 g/d) reported reductions in fasting triglycerides (27 and 19% respectively). In one of these studies, total and LDL cholesterol concentrations were also modestly reduced (Williams 1999). Jackson *et al* (1999) reported that daily addition of 10 g inulin in the diet significantly reduced the fasting insulin concentrations in 54 healthy middle aged men and women during the 8 week test period. This diet also lowered the plasma TAG levels particularly in subjects in whom fasting TAG levels were greater than 1.5 mmol/l. In animal studies, inhibition of hepatic fatty acid synthesis has been identified as the major site of action for the triglyceride-lowering effects of inulin. Since this pathway is relatively inactive in humans unless a high carbohydrate diet is fed, future attempts to demonstrate lipid lowering effects of inulin should consider the choice of subjects, the duration of the study and the background diet, as these important variables may influence the enzymes (Williams 1999). It becomes more significant in the Indian context because their diet is rich in carbohydrates.

### 12. Effect on cholesterolemia

The effect on cholesterolemia is controversial. SFr has been shown to lower serum total and LDL cholesterol in non-insulin dependent diabetic patients, but not in healthy subjects (Luo *et al* 1996; Yamashita *et al* 1984). Both short term (3 weeks) and long term (16 weeks) administration of fructans decreased total cholesterol level in the serum of rats (Gupta *et al* 1986; Fiordaliso *et al* 1995). Oligofructose influenced neither the absorption of dietary cholesterol nor the excretion of cholesterol or bile acids in ileostomic subjects (Ellegard *et al* 1997). The role of short chain carboxylic acids in these effects is difficult to establish because either in isolation or in mixture, these acids have antagonistic effects on the cholesterol metabolism. Acetate has been related with the hypercholesterolemia observed in healthy patients receiving lactulose (Jenkins *et al* 1991), whereas propionate, which lowers serum cholesterol when added to the diet of rats, may decrease cholesterol synthesis by inhibiting hydroxymethylglutaryl-CoA reductase (Rodwell *et al* 1976; Illman 1988) as shown in figure 2. Demigne *et al* (1995) suggested that a mechanism of action of oligofructose was associated with the modulation of *de novo* cholesterol synthesis by short chain fatty acids produced by the gut microflora during the fermentation process.

Kaur *et al* (1991) reported the cholesterol lowering effect of an inulin rich diet in caffeine-fed rats. Though similar results are not available in human beings, use of chicory

as a coffee substitute is well known (Pazola and Cieslak 1979).

### 13. Effect on uremia and nitrogen/urea disposal

Feeding rats a diet supplemented with inulin and oligofructose (10%) for a few weeks decreased uremia, in both normal and nephrectomized rats (Delzene *et al* 1995; Younes *et al* 1997). Dietary inulin effectively enhanced fecal nitrogen excretion and reduced renal excretion of nitrogen in rats (Younes *et al* 1995). Inulin and oligofructose serve as an energy source for intestinal bacteria which during growth, also require a source of nitrogen for protein synthesis. However, it seems unlikely that inulin-type fructans exert any noticeable effect on protein digestibility in the small intestine (Levrat *et al* 1993). When the fermentable carbohydrate intake is high, the amount of ammonia required to sustain maximal bacterial growth may become insufficient, and blood urea is then required as a ready source for bacterial protein synthesis in the cecum (Tetens *et al* 1996; Younes *et al* 1995). Besides, propionate, an important end product of bacterial fermentation of inulin type fructans, also inhibits ureagenesis in the liver in the presence of ammonia and amino acids. But whether such results (decreased uremia, shift of nitrogen excretion toward the colon) can be extrapolated to humans is questionable because of the difference in digestive tract structure and colonic microflora. However, in humans, consumption of nondigestible carbohydrates had resulted in a higher fecal excretion of nitrogen (Mortensen 1992). In addition to increasing total nitrogen transfer to colon, it is important to limit the formation of ammonia and various end products of protein catabolism, which have been proposed as causative risk factors for colonic carcinogenesis in the distal part of the large bowel (Lupton and Marchand 1989). Chicory root extract has also been reported to inhibit xanthine oxidase, an enzyme involved in the synthesis of uric acid from purines, and thus can be useful in relief from gout.

### 14. Effect of inulin, oligofructose and *Bifidobacterium longum* on colon carcinoma

Cancer of the colon is one of the leading causes of cancer morbidity and mortality among men and women. Aberrant crypts (ACP) are putative precursor lesions from which adenomas and carcinomas may develop in the colon. Administration of oligofructose or inulin in the diet significantly suppressed the total number of aberrant crypt foci (ACF) per colon compared with the control diet; the degree of inhibition was more pronounced in

rats fed inulin than in those fed oligofructose. The role played by inulin and oligofructose in reducing ACF formation, an early preneoplastic marker of malignant potential in the process of colon carcinogenesis, suggests that they may suppress colon tumourigenesis (Wargovich *et al* 1996). Kulkarni and Reddy (1994) indicated that the dietary administration of lyophilized cultures of *B. longum* (probiotic) at 1.5 and 3% levels significantly inhibited the total ACF formation and crypt multiplicity. Dietary administration of 2% lyophilized cultures of *B. longum* significantly inhibited the incidence of colon adenocarcinoma and colon tumour multiplicity in terms of tumours per animal, and in tumours per tumour bearing animal induced by subcutaneous administration of azoxymethane (Singh *et al* 1997).

2-Amino-3-methyl imidazo (4,5-F) quinoline (IQ) a heterocyclic aromatic amine has a multi target organo-specificity with specific cancer induction in the zymbal gland, skin, colon, oral cavity and mammary gland of rodents (Sugimura *et al* 1991). Cultures of *B. longum* (0.5%) significantly inhibited the IQ induced incidence of colon tumours and multiplicity of colon tumours (tumours per animal) in male rats. In female rats also, these cultures suppressed the mammary carcinogenesis to 50% of that observed in animals fed with control diets (Reddy and Rivenson 1993).

#### 14.1 Possible mechanisms of colon cancer inhibition by prebiotics

It appears that oligofructose and inulin inhibit preneoplastic lesions, probably by changing the composition of microflora (Wang and Gibson 1993; Gibson *et al* 1995; Gibson and Roberfroid 1995). The bifidobacteria, colonizing at the expense of enteropathogens, may bind the ultimate carcinogen by physically removing it via feces. The colonizing cells of bifidobacteria also produce lactic acid and thereby lower the intestinal pH to create a bacteriocidal environment for putative enteropathogens such as *Escherichia coli* and *Clostridium pufingens*. The developing of a favourable microenvironment may also involve the modulation of bacterial enzymes such as *b*-glucuronosidase that can convert procarcinogens to proximate carcinogens (Kulkarni and Reddy 1994). Buddington *et al* (2002) reported that resistance against pathogenic bacteria could also be increased in mice by feeding them with diets rich in inulin and oligofructose, which selectively encourage the proliferation of lactic acid producing bacteria already residing in gastro intestinal tract. Increasing the lactic acid producing bacteria by feeding inulin and oligofructose reduced the activities of enzymes implicated in carcinogenesis (Rowland *et al* 1998) and decreased the incidence of tumours after exposure to known carcinogens (Kato 2000).

#### 14.2 Possible mechanism of colon cancer inhibition by *B. longum*

Increased ornithine decarboxylase (ODC, EC 4.1.1.17) levels have been reported in neoplastic human colons vs normal appearing colon mucosa (Porter *et al* 1987; Singh *et al* 1992), in dysplastic vs non-dysplastic polyps (Luk and Baylin 1984) and non involved mucosa from polyposis patients vs non involved mucosa from normal individuals (Luk *et al* 1989). Studies conducted by Reddy (1999) demonstrated that colon tumour inhibitory effect by *B. longum* was associated with inhibition of colonic mucosal cell proliferation and suppression of ODC activity in the colonic mucosa. Elevated levels of ODC were also reported in colon tumours and uninvolved colonic mucosa of Azoxymethane (AOM)-treated animals and the activity was significantly decreased in colon tumours of AOM-treated animals administered with lyophilized cultures of *B. longum* (Singh *et al* 1997).

Furthermore, Mukhopadhyay *et al* (1991) demonstrated that dietary *B. longum* cultures significantly suppressed the expression of total and mutated ras-p-21 in the colonic mucosa of tumours, compared with the control diet. This inhibitory effect of *B. longum* cultures on ras-p-21 expression was again correlated with colon tumour outcome. Possibly bifidobacterial cells, as biological response modifiers, modulate the induction of the methyl guanine repair protein, O<sup>6</sup>-methylguanine DNA methyl transferase, which acts as a suicidal enzyme that stoichiometrically accepts a methyl group onto itself, restoring the original guanine in DNA by *in situ* demethylation (Pegg and Dolan 1989). It is clear from these results that *B. longum*-augmented suppression of AOM induced ras-activity may interfere with the progression of events leading to colon tumour development (Reddy 1999).

An additional mechanism for tumour suppression may involve a role for *B. longum* as an immunomodulator and biological response modifier (Okawa *et al* 1993; Sekine *et al* 1995). Kohwi *et al* (1978), demonstrated that repeated intralesional injections of *Bifidobacterium* inhibited

the growth of Meth-A tumour cells transplanted subcutaneously into syngeneic BALB/c mice. Furthermore Sekine *et al* (1995) and Okawa *et al* (1993) demonstrated that a water soluble cell fraction of bifidobacteria induces an antitumour effect and plays an important role as an immunomodulator in the intestines of humans and animals.

### 15. Breast cancer

Taper and Roberfroid (1999) studied the influence of inulin and oligofructose on breast cancer and tumour growth. In a preliminary study on methylnitroso-urea induced mammary carcinogenesis in Sprague-Dawley female rats, 15% oligofructose added to the basal diet lowered the number of tumour bearing rats and also decreased the total number of mammary tumours. The functional effects of inulin and oligofructose on humans and animals have been summarized in tables 3 and 4, respectively.

### 16. Application of inulin and oligofructose in the food industry

Different functional attributes of inulin and oligofructose are due to the difference in their chain lengths. Due to its longer chain length inulin is less soluble than oligofructose and has the ability to form inulin microcrystals when sheered in water or milk. These crystals are not discretely perceptible in the mouth, but they interact to form a smooth creamy texture and provide a fat like mouth feel. Inulin has therefore been used successfully to replace fat in table spreads, baked goods, fillings, dairy products, frozen desserts and dressings. Furthermore, fructans are non cariogenic as they are not used by *Streptococcus mutans* to form acid and glucans which are responsible for dental caries.

Oligofructose is composed of shorter chain oligomers and possesses functional qualities similar to sugar or glucose syrups. It is actually more soluble than sucrose and provides about 30 to 50% of the sweetness of table sugar.

**Table 3.** Effect of inulin/oligofructose in diet on various health parameters in humans.

Effect	Reference
Fermentation of fructans leads to selective stimulation of growth of bifidobacteria population in large intestine	Roberfroid <i>et al</i> 1995; Gibson <i>et al</i> 1995
Oligofructose and inulin relieved constipation	Hidaka <i>et al</i> 1991; Kleesen <i>et al</i> 1997
Inulin lowers the blood glucose level	Rumessen <i>et al</i> 1990; Yamashita <i>et al</i> 1984
Inulin improves the absorption of calcium	Coudray <i>et al</i> 1997; Van den Heuvel <i>et al</i> 1999
Reduce fasting triglyceride and LDL cholesterol	Williams 1999
Fructan lowers serum total and LDL cholesterol in non-insulin dependent diabetic patients but not in healthy subjects	Yamashita <i>et al</i> 1984; Luo <i>et al</i> 1996



**Table 4.** Effect of feeding inulin/oligofructose on various parameters in experimental animals.

Effect	Reference
Fermentation of fructans leads to selective stimulation of growth of bifidobacteria population	Campbell <i>et al</i> 1997
Inulin/oligofructose feeding increased the absorption of calcium and magnesium	Delzenne <i>et al</i> 1995; Ohta <i>et al</i> 1994
Oligofructose reduces the post prandial glycemia and insulinemia in rats	Oku <i>et al</i> 1984
Lowers triglyceride content of blood and liver in rats and hamsters	Kaur <i>et al</i> 1988; Delzenne and Kok 1999; Trautwein <i>et al</i> 1998
Feeding of inulin decreased total cholesterol level in serum of rats	Fiordaliso <i>et al</i> 1995
Inulin/oligofructose decreased uremia in normal and nephrectomized rats	Delzenne <i>et al</i> 1995; Younes <i>et al</i> 1997
Addition of inulin and oligofructose significantly inhibited the growth of various kinds of cancerous tumours in rats	Taper <i>et al</i> 1998; Taper and Roberfroid 1999

Oligofructose contributes body to dairy products and humectancy to soft baked goods, depresses the freezing point in frozen desserts, provides crispness to low fat cookies and acts as a binder in nutritional or granola bars in much the same way as the sugar, but with the added benefits of fewer calories, fibre enrichment and other nutritional properties. Oligofructose is often used in combination with high intensity sweeteners to replace sugars and provide a well balanced sweetener profile, and mask the after taste of aspartame or acesulfamek (Weidmann and Jager 1997). So with the oligofructose as an additive, healthy dairy drinks improved with added fibre content and sugar reduced baked goods and products fit for the diabetics can be developed.

## 17. Conclusions

Inulin and oligofructose have many interesting functional attributes that are useful in formulating the food of today and tomorrow. The consumer of today is health conscious and demands foods which is both tasty as well as low in fat and calories, with additional health benefits. In present day society, the leading health concerns are heart disease, cancer, high cholesterol, weight control, osteoporosis and diabetes. Inulin and oligofructose are widely used in functional foods throughout the world for their health promoting properties. Use of garlic, ginger and onion in traditional Indian foods is well known. Fructan (inulin and oligofructose) are the ingredients that will meet the needs of the food industry for healthy foods in the future.

## References

Arbeeny C A, Meysers D S, Bergquist K E and Gregg R E 1992 Inhibition of fatty acid synthesis decreases very-low density lipoprotein secretion in the hamster; *J. Lipid Res.* **33** 843–851

Baird G D, Lomax M A, Symonds H W and Shaw D R 1980 Net hepatic and splanchnic metabolism of lactate, pyruvate

and propionate in dairy cows *in vivo* in relation to lactation and nutrient supply; *Biochem. J.* **186** 47–57

Boillot J, Alamowitch C, Berger A M, Luo J and Bruzzo F 1995 Effect of dietary propionate on hepatic glucose production, whole body glucose utilization, carbohydrate and lipid metabolism in normal rats; *Br. J. Nutr.* **73** 241–255

Buddington K K, Donahoo J B and Buddington R K 2002 Dietary oligofructose and inulin protect mice from enteric and systemic pathogens and tumor inducers; *J. Nutr.* **132** 472–477

Buddington R K, Williams C H, Chen S C and Witherly S A 1996 Dietary supplementation of neosugar alters the fecal flora and decreases activities of some reductive enzymes in human subjects; *Am. J. Clin. Nutr.* **63** 709–716

Campbell J M, Fahey G C and Wolf B W 1997 Selected indigestible oligosaccharides affect large bowel mass, cecal and fecal short-chain fatty acids, pH and microflora in rats; *J. Nutr.* **127** 130–136

Carabin I G and Flamm W G 1999 Evaluation of safety of inulin and oligofructose as dietary fiber; *Regul. Toxicol. Pharmacol.* **30** 268–282

Coudray C, Bellanger J, Castiglia-Delavaud C, Remesy C, Vermorel M and Rayssiguier Y 1997 Effects of soluble or partly soluble dietary fibres supplementation on absorption and balance of calcium, magnesium, iron and zinc in healthy young men; *Eur. J. Clin. Nutr.* **51** 375–380

Crittenden R G and Playne M J 1996 Production, properties and applications of food grade oligosaccharides; *Trends Food Sci. Technol.* **7** 353–361

Cummings J H, Macfarlane G T and Englyst H N 2001 Prebiotic digestion and fermentation; *Am. J. Clin. Nutr. (Suppl.)* **73** 415–420

Daubioul C A, Taper H S, De Wispelaere L D and Delzenne N M 2000 Dietary oligofructose lessens hepatic steatosis, but does not prevent hypertriglyceridemia in obese rats; *J. Nutr.* **130** 1314–1319

Davignon J and Cohn J S 1996 Triglycerides: a risk factor for coronary heart disease; *Atherosclerosis* **124** 57–64

Debruyne A, Alvarez A P, Sandra P and De Leenheer L 1992 Isolation and identification of *b*-D-fructofuranosyl-(2,1)-D-fructose, a product of the enzymatic hydrolysis of the inulin from *Cichorium intybus*; *Carbohydr. Res.* **235** 303–308

De Leenheer 1996 Production and use of inulin: industrial reality with a promising future; in *Carbohydrates as organic raw materials* (eds) H Vanbekkum, H Roper and F Varagen (New York: VCH) Vol. 3, pp 67–92

- Delzenne N and Kok N N 1998 Effect of non-digestible fermentable carbohydrates on hepatic fatty acid metabolism; *Biochem. Soc. Trans.* **26** 228–230
- Delzenne N and Kok N N 1999 Dietary fructooligosaccharides modify lipid metabolism in the rat; *J. Nutr.* **129** 1467S–1469S
- Delzenne N, Aertssens J, Verplaetse H, Roccaro M and Roberfroid M 1995 Effect of fermentation fructo oligosaccharides on mineral, nitrogen and energy digestive balance in the rat; *Life Sci.* **57** 1579–1587
- Demigne C, Morand C, Levrat M A, Besson C, Moundras C and Remesy C 1995 Effect of propionate on fatty acid and cholesterol synthesis and on acetate metabolism in isolated rat hepatocytes; *Br. J. Nutr.* **74** 209–219
- Ellegard L, Andersson H and Boseus L 1997 Inulin and oligofructose do not influence the absorption of cholesterol or the excretion of cholesterol, Ca, Mg, Zn, Fe or bile acids but increase energy excretion in ileostomy subjects; *Eur. J. Clin. Nutr.* **51** 1–5
- Fiordaliso M, Kok N, Desager J P, Goethals F, Deboyser D, Roberfroid M and Delzenne N 1995 Dietary oligofructose lowers serum and VLDL concentrations of triglycerides, phospholipids and cholesterol in rats; *Lipids* **30** 163–167
- Fishbein L, Kaplan H and Gough M 1988 Fructooligosaccharides: a review; *Toxicology* **30** 104–107
- Gibbons G F 1990 Assembly and secretion of hepatic very-low-density lipoprotein; *Biochem. J.* **268** 1–13
- Gibson G R, Beatty E R, Wang X and Cummings J H 1995 Selective stimulation of bifidobacteria in the human colon by oligofructose and inulin; *Gastroenterology* **108** 975–982
- Gibson G R and Roberfroid M B 1995 Dietary modulation of the human colonic microbiota: introducing the concept of prebiotics; *J. Nutr.* **125** 1401–1412
- Gibson G R and Wang X 1994 Enrichment of bifidobacteria from human gut contents by oligofructose using continuous culture; *FEMS Microbiol. Lett.* **118** 121–128
- Gupta A K and Bhatia I S 1980 Glucofructosan biosynthesis in *Fusarium oxysporum*; *Phytochemistry* **19** 2557–2563
- Gupta A K and Kaur N 1997 Fructan storing plants – A potential source of high fructose syrups; *J. Sci. Ind. Res.* **56** 447–452
- Gupta A K and Kaur N (eds) 2000 Fructan metabolism in Jerusalem artichoke and chicory; in *Carbohydrate reserves in plants – synthesis and regulation* (The Netherlands: Elsevier Science) pp 223–248
- Gupta A K, Kaur N and Nath S 1986 Hypocholesterolemic effect of *Cichorium intybus* roots in plasma and liver of rats; *Med. Sci. Res.* **14** 212
- Gupta A K, Mamta and Bhatia I S 1985 Glucofructosan metabolism in *Cichorium intybus* roots; *Phytochemistry* **24** 1423–1427
- Hendry G 1993 Evolutionary origins and natural functions of fructans. A climatological, biogeographic and mechanistic appraisal; *New Phytol.* **123** 3–14
- Hidaka H, Eida T, Takiwaza T, Tokunga T and Tashiro Y 1986 Effects of fructooligosaccharides on intestinal flora and human health; *Bifidobac. Microflora* **5** 37–50
- Hidaka H, Tashiro Y and Eida T 1991 Proliferation of bifidobacteria by oligosaccharides and their useful effect on human health; *Bifidobac. Microflora* **10** 65–79
- Hillgartner F D, Salate L M and Goodridge A G 1995 Physiological and molecular mechanisms involved in nutritional regulation of fatty acid synthesis; *Physiol. Rev.* **75** 47–76
- Hond E D, Geypens B and Ghooys Y 2000 Effect of high performance chicory inulin on constipation; *Nutr. Res.* **20** 731–736
- Hussein H S, Flickinger E A and Fahey G C 1999 Pet food applications of inulin and oligofructose; *J. Nutr. (Suppl.)* **129** 1454–1456
- Illman R J, Toppoong D L, McIntosh G H, Trimble R P and Storer G B 1988 Hypercholesterolemic effects of dietary propionate: studies in whole animals and perfused rat liver; *Ann. Nutr. Metab.* **32** 95–107
- Jackson K G, Taylor G R J, Clohessy A M and Williams C M 1999 The effect of daily intake of inulin on fasting lipid, insulin and glucose concentration in middle-aged men and women; *Br. J. Nutr.* **82** 23–30
- Jenkins D J A, Wolever T M S and Jenkins A 1991 Specific types of colonic fermentation may raise low-density-lipoprotein-cholesterol concentrations; *Am. J. Clin. Nutr.* **54** 141–147
- Kato I 2000 Antitumour activity of the lactic acid bacteria; in *Probiotics* (eds) R Fuller and G Perdigon (Dordrecht: Kluwer Academic Publishers) pp 115–138
- Kaur N, Gupta A K and Saijpal S 1988 Hypotriglyceridaemic effect of *Cichorium intybus* roots in ethanol injected and saturated fat-fed rats; *Med. Sci. Res.* **16** 91–92
- Kaur N, Gupta A K, Saijpal S, Indu and Gupta P P 1989 Triglyceride and cholesterol lowering effect of chicory roots in the liver of dexamethasone-injected rat; *Med. Sci. Res.* **17** 1009–1010
- Kaur N, Gupta A K and Uberoi S K 1991 Cholesterol lowering effect of chicory (*Cichorium intybus*) root in caffeine-fed rats; *Med. Sci. Res.* **19** 643
- Kaur N, Jain H, Mann P, Gupta A K and Singh R 1992 A comparison of properties of invertases and inulinase from chicory; *Plant Physiol. Biochem.* **30** 445–450
- Kleessen B, Sykura B, Zunft H J and Blaut M 1997 Effects of inulin and lactose on faecal microflora, microbial activity and bowel habit in elderly constipated persons; *Am. J. Clin. Nutr.* **65** 1397–1402
- Kohwi Y, Imai K, Tamura Z and Hashimoto Y 1978 Antitumour effect of *Bifidobacterium infantis* in mice; *Gann* **69** 163–168
- Kok N N, Roberfroid M, Robert A and Delzenne N 1996 Involvement of lipogenesis in the lower VLDL secretion induced by oligofructose in rats; *Br. J. Nutr.* **76** 881–890
- Kulkarni N and Reddy B S 1994 Inhibitory effect of *Bifidobacterium longum* cultures on the azoxymethane-induced aberrant crypt loci formation and fecal bacterial *b*-glucuronidase; *Proc. Soc. Exp. Biol. Med.* **207** 278–283
- Lee K U, Park J I, Kim C H, Hong S K and Suh K I 1996 Effect of decreasing plasma free fatty acids by acipimox on hepatic glucose metabolism; *Metabolism* **45** 1408–1414
- Levrat M A, Remesy C and Demigne C 1991 High propionate acid fermentation and mineral accumulation in the caecum adapted to different levels of inulin; *J. Nutr.* **121** 1730–1737
- Levrat M A, Remesy C and Demigne C 1993 Influence of inulin on urea and ammonia in the rat caecum: consequences on nitrogen excretion; *J. Nutr. Biochem.* **4** 351–356
- Lopez H W, Coudray C, Levrat M A, Coudray C F, Demigne C and Remesy C 2000 Fructo oligosaccharides enhance mineral apparent absorption and counteract the deleterious effects of phytic acid on mineral homeostasis in rats; *J. Nutr. Biochem.* **11** 500–508
- Luk G D and Baylin S B 1984 Ornithine decarboxylase as biological marker in familial colonic polyps; *New Engl. J. Med.* **311** 80–83
- Luk G D, Zhang S Z and Hamilton S R 1989 Effects of timing of administration and dose of difluoro methyl ornithine on rat colonic carcinogenesis; *J. Natl. Cancer Inst.* **81** 421–427

- Luo J, Rizkalla S W, Alamovitch C, Boussairi A and Blayo A 1996 Chronic consumption of short chain fructooligosaccharides by healthy subjects decreased basal hepatic glucose production but had no effect on insulin-stimulated glucose metabolism; *Am. J. Clin. Nutr.* **63** 639–645
- Lupton J R and Marchand L J 1989 Independent effects of fibre and pectin on colonic luminal ammonia concentration; *J. Nutr.* **119** 235–241
- MacGarey J D, Leatherman G F and Foster D W 1978 Carnitine palmitoyl transferase 1. The site of inhibition of hepatic fatty acid oxidation by malonyl CoA; *J. Biol. Chem.* **253** 4128–4136
- Menne E, Guggenbuhl N and Roberfroid M 2000 Fn-type chitory inulin hydrolysate has a prebiotic effect in humans; *J. Nutr.* **130** 1197–1199
- Miller-Catchpole R 1989 Bifidobacteria in clinical microbiology and medicine; in *Biochemistry and physiology of bifidobacteria* (eds) A Bezkorovainy and R Miller-Catchpole (Boca Raton: CRC Press) pp 177–200
- Morgan L M 1996 The metabolic role of G1P: Physiology and pathology; *Biochem. Soc. Trans.* **24** 585–591
- Mortensen P B 1992 Effect of oral administered lactulose on colonic nitrogen metabolism and excretion; *Hepatology* **16** 1350–1356
- Mukhopadhyay T, Tainsky M, Cavander A C and Roth A C 1991 Specific inhibition of K-ras expression and tumorigenicity of long cancer cells by antisense RNA; *Cancer Res.* **51** 5270–5274
- Ohta A, Ohtsuki M, Takizawa T, Inaba H, Adachi T and Kimura S 1994 Effects of fructooligosaccharides on the absorption of magnesium and calcium by cecectomized rats; *Int. J. Vitam. Nutr. Res.* **64** 316–323
- Okawa T, Niibe H, Arai T, Sekiba K, Noda K, Takeuchi S, Hashimoto S and Ogawa N 1993 Effect of LC 9018 combined with radiation therapy on carcinoma of the uterine cervix; *Cancer* **72** 1949–1954
- Oku T, Tokunaga T and Hosoya H 1984 Non-digestibility of a new sweetener, “Neosugars” in the rat; *J. Nutr.* **114** 1574–1581
- Parker S L, Tong T, Bolden S and Wingo C A 1997 Cancer statistics; *Cancer J. Clin.* **47** 5–27
- Pazola Z and Cieslak J 1979 Changes in carbohydrates during the production of coffee substitute extracts especially in the roasting processes; *Food Chem.* **4** 41–44
- Pegg A E and Dolan M E 1989 Investigation of sequence specificity in DNA alkylation and repair using oligodeoxy ribonucleotide substrate; in *DNA repair mechanisms and their biological implications in mammalian cells* (eds) M W Lambart and J Laval (New York: Plenum Press) pp 45–59
- Porter C W, Herrera-ornelas L, Clark J, Pera P, Petrelli N J and Mittleman A 1987 Polyamine biosynthetic activity in normal and neoplastic human colorectal tissue; *Cancer* **60** 1275–1281
- Reddy B S 1999 Possible mechanisms by which pro- and prebiotics influence colon carcinogenesis and tumour; *J. Nutr. (Suppl.)* **129** 1478–1482
- Reddy B S and Rivenson A 1993 Inhibitory effect of *Bifidobacterium longum* on colon, mammary and liver carcinogenesis induced by 2-amino-3-methyl imidazol [4,5-f] quinoline, a food mutagen; *Cancer Res.* **53** 3914–3918
- Remesy C, Behr S R, Levrat M A and Demigne C 1992 Fibre fermentability in the rat caecum and its physiological consequence; *Nutr. Rev.* **12** 1235–1244
- Remesy C, Levrat M A, Gamet L and Demigne C 1993 Faecal fermentation in rats fed oligosaccharides (inulin) are modulated by dietary calcium level; *Am. J. Physiol.* **264** 855–862
- Roberfroid M B 1993 Dietary fibre, inulin and oligofructose: a review comparing their physiological effects; *Crit. Rev. Food Sci. Technol.* **33** 103–148
- Roberfroid M B 2001 Prebiotics: preferential substrates for specific germs?; *Am. J. Clin. Nutr. (Suppl.)* **73** 406–409
- Roberfroid M B, Bornet F, Bouley Ch and Cummings J H 1995 Colonic microflora : nutrition and health; *Nutr. Rev.* **53** 127–130
- Rodwell V W, Nordstorn J L and Mitshelen J L 1976 Regulation of HMG CoA reductase; *Adv. Lipid Res.* **14** 1–74
- Roland N, Nugon-Baudon L, Raiband P and Szilic O 1993 Comparative study of the fermentative characteristics of inulin and different types of fibre in rats inoculated with a human whole faecal flora; *Br. J. Nutr.* **74** 239–249
- Rowland I R, Rumney C J, Coutts J T and Lievens L C 1998 Effect of *Bifidobacterium longum* and inulin on gut bacterial metabolism and carcinogen-induced aberrant crypt foci in rats; *Carcinogenesis* **19** 281–285
- Rumessen J J, Bode S, Hamberg O and Gudman-Hoyer E 1990 Fructans of jerusalem artichokes: intestinal transport, absorption, fermentation and influence; *Am. J. Clin Nutr.* **52** 675–781
- Sekine K, Ohta J, Onishi M, Tatsuki T, Shimokawa Y, Toida T, Kawashima T and Hashimoto Y 1995 Analysis of antitumour properties of effector cells stimulated with a cell wall preparation of *Bifidobacterium* infants; *Biol. Pharm. Bull.* **18** 148–153
- Singh J, Kelloff G and Reddy B S 1992 Effect of chemopreventive agents on intermediate biomarkers during different stages of azoxymethane-induced colon carcinogenesis; *Cancer Epidemiol. Biomark. Prev.* **1** 405–411
- Singh J, Rivenson A, Tomita M, Shimamura S, Ishibashi N and Reddy B S 1997 *Bifidobacterium longum*, a lactic acid-producing intestinal microflora inhibit colon cancer and modulate the intermediate biomarkers of colon carcinogenesis; *Carcinogenesis* **18** 1371–1377
- Smeekens S, Angenent G, Ebskamp M and Weisbeek P 1991 Molecular biology of fructan accumulation in plants; *Biochem. Soc. Trans.* **19** 565–569
- Stals H K, Top W and Declercq P E 1994 Regulation of triacylglycerol synthesis in permeabilized rat hepatocytes. Role of fatty acid concentration and diacylglycerol acyl transferase; *FEBS Lett.* **343** 99–109
- Sugimura T, Wakabayashi K, Ohgaki H, Takayama S, Nagao M G and Esumi H 1991 Heterocyclic amines produced in cooked food: unavailable xenobiotics; in *Xenobiotics and cancer* (ed.) L Ernster (Tokyo: Japan Scientific Society Press) pp 279–288
- Taguchi A, Ohta A, Abe M, Baba S, Ohtsuki M, Takizawa T, Yuda Y and Adachi T 1994 The influence of fructooligosaccharides on the bone of model rats with ovariectomized osteoporosis; *Sci. Rep. Meiji Seika Kaisha* **33** 37–44
- Taper H S, Lemort C and Roberfroid M 1998 Inhibition effect of dietary inulin and oligofructose on growth of transplantable mouse tumour; *Anticancer Res.* **18** 4123–4126
- Taper H S and Roberfroid M 1999 Influence of inulin and oligofructose on breast cancer and tumor growth; *J. Nutr. (Suppl.)* **129** 1488–1491
- Tetens I G, Livesey G and Eggum B O 1996 Effect of type and level of dietary fibre supplements on nitrogen retention and excretion patterns; *Br. J. Nutr.* **75** 461–469

- Tokunaga T, Oku T and Hosoya N 1986 Influence of chronic intake of a new sweetener fructooligosaccharide (Neosugar) on growth and gastro intestinal function in the rat; *J. Nutr. Sci. Vitam.* **32** 111–121
- Trautwein E A, Rieckhoff D and Erbersdobler H F 1998 Dietary inulin lowers plasma cholesterol and triacylglycerol and alters biliary bile acid profile in hamsters; *J. Nutr.* **128** 1937–1943
- Van den Heuvel E G H M, Muys T, Van Dokkum W and Schaafsma G 1999 Oligofructose stimulates calcium absorption in adolescents; *Am. J. Clin. Nutr.* **69** 544–548
- Van Loo J, Coussement P, De Leenheer L, Hoebregs H and Smits G 1995 On the presence of inulin and oligofructose as natural ingredients in western diet; *CRC Crit. Rev. Food Sci. Nutr.* **35** 525–552
- Vijin I and Smeekens S 1999 Fructan : more than a reserve carbohydrate?; *Plant Physiol.* **120** 351–359
- Wang X and Gibson G R 1993 Effects of the *in vitro* fermentation of oligofructose and inulin by bacteria growing in the human large intestine; *J. Appl. Bacteriol.* **75** 373–380
- Wargovich M H, Chen D D, Jimenez A, Steele V E, Velasco M, Stephens C, Price R, Gray K and Kelloff G J 1996 Aberrant crypts as a biomarker for colon cancer, evaluation of potential chemopreventive agents in the rat; *Cancer Epidemiol. Biomark. Prev.* **5** 355–360
- Weidmann M and Jager M 1997 Synergistic sweeteners; *Food Ingredients Int.* (November–December) 51–56
- Williams C M 1999 Effects of inulin on lipid parameters in humans; *J. Nutr.* **129** 1471S–1473S
- Yamashita K, Kawai K and Itakura K 1984 Effect of fructooligosaccharides on blood glucose and serum lipids in diabetic subjects; *Nutr. Res.* **4** 961–966
- Younes H, Demigne C and Remesy C 1996 Acidic fermentation in the caecum increases absorption of calcium and magnesium in the large intestine of the rats; *Br. J. Nutr.* **75** 301–314
- Younes H, Garleb K, Behr S, Remesy C and Demigne C 1995 Fermentable fibres or oligosaccharides reduce urinary nitrogen excretion by increasing urea disposal in the rat cecum; *J. Nutr.* **125** 1010–1016
- Younes H, Remesy C, Behr S and Demigne C 1997 Fermentable carbohydrate exerts an urea lowering effect in normal and nephrectomised rats; *Am. J. Physiol.* **35** 515–521

*MS received 12 June 2002; accepted 27 August 2002*

Corresponding editor: MAN MOHAN JOHRI