Prebiotics In Foods And Their Beneficial Effects

International Life Sciences Institute-India

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FOREWORD

There has been an immense awareness and knowledge about the Gut Microbiome and its composition as well as health benefits. Gut bacterial population is largely related to the diets consumed and vary from region to region, person to person and even from time to time. The presence of a healthy population of gut bacteria depends on the substrate i.e. the non-digestible components of food. These are broadly classified as **Prebiotics.** They are complex carbohydrates that cannot be broken down by the digestive enzymes in the gut. If the appropriate prebiotics are consumed through foods in adequate quantities then the gut flora will be in a healthy and beneficial combination and promote gut as well as overall health and well-being.

The **Indian Food Composition Tables** are recent version of the erstwhile **Nutritive Value of Indian Foods** published by the ICMR-NIN provide a data base of the nutrient and non nutrient components of commonly consumed foods in India. The authors have culled out the prebiotic content of these foods to help consumers choose foods with significant prebiotic content. This may also help the food formulators to come up with good combinations of pre and probiotics for short-term needs of those with impaired gut health and help researchers in the area of gut microbiome research.

We sincerely hope that this data compilation will serve the needs of those who intend having a healthy gut microbiome through their daily food choices..

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PREBIOTICS IN FOODS AND THEIR BENEFICIAL EFFECTS

1. What Are Prebiotics ?

The term "prebiotics" was coined by Gibson and Roberfroid in 1995 and is defined as 'a nondigestible food ingredient that is known to selectively stimulate the growth and activity of one or more beneficial bacteria in the colon and which in turn improves the host health status'. To perform as prebiotics, the components of functional food or supplement must be able to withstand the digestive processes, before they reach the colon and preferably persist throughout the large intestine such that their benefits to be conferred upon the essential microbiota (Gibson et al., 2004).The bacteria which belong to the family of *bifidobacteriaceae* and *lactobacillaceae*, have been receiving a considerable attention from the ancient days, as essential gut microbes which perform the role of probiotics to improve the health status of the host (Gibson and Roberfroid., 1995).

The recent studies in this area highly acknowledge the role of oligosaccharides as essential prebiotics, as these are low molecular weight carbohydrates and represent major sources of carbon for the colonic bacteria (Gupta and Kaur., 2000). In addition, oligosaccharides act as dietary bulking agent and act as available substrate for resident colonic bacteria, inducing fermentation, thereby resulting in a pH decrease, and initiating the production of short chain fatty acids which are known to antagonize the growth and multiplication of pathogenic microorganisms (Morisse et al., 1993).

Oligosaccharides are relatively short-chain carbohydrates, present abundantly in plant kingdom with relatively smaller quantities in human milk and in the colostrum of various animals (Bucke and Rastall, 1990). Earlier they were considered as reserve sugars, available in seeds and tubers, assisting in growth and germination. However, their promising impact on gastrointestinal physiology has highlighted a renewed significance to human health, in recent years (Van Loo et al., 1999). Naturally, this component is present in whole food and even incorporated as supplements in the Western diets to improve the quality of food, wherein it demonstrated an increase in the colonization of good bacteria over harmful bacteria and thereby improving the health status of individuals who consume it, particularly those with metabolic syndrome (Roberfroid et al., 1993).

Various types of oligosaccharides are evident in natural foods and most of them are found in fruits, vegetables, seeds, onion, garlic, milk, honey and Japanese traditional foods like sake and sweet sake etc (Nakuki., 2005; Longvah et al., 2017). The major food sources containing some of the common oligosaccharides obtained from plant sources are tabulated in Table 1. The monosaccharide unit of lactose, and xylose are the major sources of oligosaccharides, which are either naturally or industrially processed, as hot water or alcoholic extracts, and are synthesized enzymatically after hydrolysis. Obviously, the market for prebiotics is heralding a rapid global expansion in recent years (Machuca et L., 2015) and is highly focused in the areas of dairy and baking industries, resulting in development of end products which are enriched with prebiotics as functional foods (Machuca et L., 2015). The major types of oligosaccharides as prebiotics, available either naturally or industrially are tabulated in Table 2.

2. Health Benefits of Oligosaccharide

Gastrointestinal Health

The recent scientific studies provide a strong correlation between intestinal microbiota and development of various metabolic syndrome in humans (Clemente et al., 2012). Hence, the knowledge on intestinal microbiota has picked up momentum in recent years. Prebiotics promote good microbial colonies and prevent colonization of pathogens, resisting them and interacts intensively with the host by modulating both the innate and adaptive immune responses, and by producing metabolites which affect the body (Clemente et al., 2012). Diet composition and intake of drugs (antibiotics) play a major decisive role as an environmental factor, along with the host's genetic factors to determine the microbiota pattern of intestine. The oligosaccharides as prebiotics selectively alter both the composition and activity of the gastrointestinal microbiota, thereby directly impacting the health status of the host (Rochfort et al., 2007).

Both *in-vitro* and *in-vivo* studies provide a strong evidence about the increase in the growth of bifidobacteria, when the subjects were fed with inulin enriched foods (Gibson et al., 1995). These bacteria dominate the intestinal flora in breastfed infants and constitute one of the major organisms in the colonic flora of healthy children and adults (Gibson et al., 1995). Interestingly, the counts for this strain appear to be declining or all together disappearing in unhealthy individuals (Mitsuoka 1987) and, further a study by Gibson and Wang (1994) elucidated that bifidobacteria have a potential to secrete bioactive compounds which have antimicrobial properties, that prevent colonization of pathogenic bacteria in humans and experimental animals. Further studies have also established that oligosaccharides cure constipation by providing bulk to the digested food material and by enhancing bowel movements (Gibson et al., 1995). Oligosaccharides have also shown promising results in combating acute gastroenteritis, E.coli gastroenteritis, Salmonella and Helicobacter Pyroli–associated gastroenteritis and irritable and inflammatory bowel conditions and inflammatory bowel disorders etc (Macfarlane et al., 2006).

Immune Stimulation

The microflora influenced by prebiotics demonstrates an inherent tolerance to commensal flora and also determine the extent and type of proinflammatory factors activated by the infective commensal microflora species (Cross, 2002). Further, the helpful bacteria enhance the secretion of IL-10, which confers protection against proinflammation (Roitt et al., 1998), and its mediated hypersensitivity, allergy and inflammatory bowel disorders (Saarela et al., 2002). A study by Sullivan and Nord, (2002) focused on the increased circulatory IL10 levels in IL-10 deficit mice, which were fed with lactobacilli, showed a decline in the onset of colitis. Further, in another study, a faster clearance of bacterial and viral agents from the respiratory tract was noted and it was accompanied by an increase in non-specific phagocytic activity of alveolar macrophages and pathogen-specific serum IgG levels, in probiotic administered animal models (Cross., 2002).

Cholesterol Reduction and Cardiovascular Disease Risk

Many *in-vivo* studies have demonstrated the potential of normalizing lipid profiles in obese individuals, who were administered prebiotics along with lactobacilli and this was also characterized by a decline in LDL, and an enhancement of HDL cholesterol levels in these subjects (Naruszewicz et al., 2002; Pereira et al., 2003) and this function was attributable to enhanced bile acid production, which in turn degrades the excess cholesterol by bacterial activity. Wang et al., 2015, successfully demonstrated from a rodent study, that feeding of 5-15g of oligosaccharides for a period of two-three months lowered the blood lipid profiles and glucose levels along with an inhibition of lipogenic enzymes and an enhanced production of short-chain fatty acids. However, on the other hand, in many human studies no significant effects were observed in normal individuals, while reduced blood triglyceride levels were noticed in hypercholesterolemic men who consumed 20 g inulin/day, for a period of three months (Mahendra and Sheth., 2013).

Cancer

Cancer represents a major inducer of mortality amongst non-communicable disorders of which large bowel cancer is a prime reason for death in the western world. Both genetic and environmental factors including diet play an important role in these aliments (Rao, 1999). In several cases cooking of meat stimulates the production of cyclohexanes, which are mutagenic in nature and the presence of prebiotics with essential bacteria, prevents such components mediated mutagenic induction (Cummings et al., 1979). Further, it is deduced from a study on Sprague Dawley female rats that prebiotics mediated protection against methylnitroso-urea-induced mammary carcinogenesis by lowering the % of mammary gland tumors in experimental animals (Taper and Roberfroid et al., 2005).

Effect on Mineral Metabolism

Oligosaccharides have a direct impact on calcium absorption and so promote bone and teeth mineral density. An extract from chicory roots when administered, increases gut absorption of calcium, which results in enhanced bone mineral density and there by postponing osteoporosis in aged individuals (Gibson., 1995). Studies in experimental animals demonstrated enhanced calcium and magnesium absorption in rats administered with inulin as compared to controls (Delzenne et al 1995). This increased calcium absorption could be due to its increased availability, because of its transfer from the small intestine to the large bowel and the osmotic effect of inulin and oligofructose, which increases the solubility of calcium by advancing the bacterial phytase activity and which in turn decreases the pH of ileum, resulting in increased concentration of ionized minerals leading to an enhanced passive absorption (Lopez et al 2000).

3. Oligosaccharides as Prebiotics and its Components in Indian Foods

Among oligosaccharides, fructo oligosaccharides are a class of well researched prebiotics, and are known to improve colonization of bifidobacteria and lactobacilli in the colon of human subjects (Murphy., 2000). The Indian food sources are rich in fructo oligosaccharides (Inulin and oligofructose) and their % per wet weight from various sources is mentioned in Table 3. Different plant species contain either singly or in combination, short (wheat, onion, and banana), medium (dahlia tubers, garlic and Jerusalem artichoke) and long chains (globe artichoke and chicory) of inulin (Van Loo et al., 1995). It is well established that chicory root is the best source for inulin and contains about 70-80% of its dry weight as reserve carbohydrate (Gupta et al 1985).

In the Indian scenario, the cereals, millets and pulses are a rich source of dietary fiber and as per the Indian Food Composition Table (IFCT -2017), the fiber content ranges between 5-12% of total weight, amongst which millets and pulses boast a higher proportion than other food ingredients (Longvah et al., 2017). Interestingly, many studies show that germination reduces the prebiotic content in many pulses (Martínez-Villaluenga et al., 2008). However, an enhanced formation of bifidobacteria colony was recorded for fermented food products than for natural ones, in high-fat diet models (Wang et al., 2015).

Total oligosaccharides in different pulses range from 70.7-114.9 mg/g (dry weight basis) from lentils to chickpeas (Han and Baik 2006). The beans contain sucrose, stachyose, and verbascose as predominant oligosaccharides, while the majority of pulses contain agalactosides which are characterized by (1 - 6) links between the galactose moieties (Han and Baik 2006). These are the second most abundant soluble carbohydrates after sucrose and are classified as raffinose (trisaccharide), stachyose (tetrasaccharide) and verbascose (pentasaccharide) based on their structure (Hoveer et al., 2010). A study by Moussou et al. (2016) showed that lentils have the lowest content of raffinose (37.5 mg/g dry weight), while peas have a higher range (66.3mg/g dry weight). They also reported that cotyledons had a higher content of these oligosaccharides than that of their seed coats. These agalactosides are not hydrolyzed by the human digestive system and hence are fermented by colonic bacteria, thereby helping normalize bowel function by affecting an increase in the lactobacilli and bifidobacteria content, while simultaneously decreasing the enterobacteria content of the colon. The common agricultural crops containing the prebiotic active principles in Indian foods are listed in Table 4.

Further, the Inulin and oligofructose of oligosaccharides are officially recognized as natural food ingredients in most European countries and are Generally Regarded As Safe (GRAS) in the United States (Schaafsma et al., 2015). 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. The average daily consumption of inulin and oligofructose has been found to be nearly 1–4 and 3-11 g in the United States and Europe respectively (Van Loo et al 1995). However, such a data is not available with respect to the Indian scenario, and this paucity is attributable mainly to diversity in food source, culture etc. However, deducing from the data published by National Nutrition Monitoring Bureau, 2012 and Indian Food Composition Tables 2017, it is clear that amongst the adults, the intake of prebiotics through oligosaccharide derived from various sources is well within the western population consumption pattern, though a bit on the lower side (Western population 1-4g/day vs Indian population 1.5-2g/day). The main reason for higher levels of prebiotics in Indian adults is because of intake which consists of cereals and millets, pulses and legumes and oils and oil seeds (Table. 5). On the other hand, there is no significant contribution of prebiotics in the diet from vegetables, fruits and tubers. In fact, the overall reason for the little diminished value of prebiotics for Indian adults in comparison to western adults is due to their poor intake of millets over cereals in recent years.

Beneficial Effects of Oligosaccharides

Many studies have depicted the beneficial effects of inulin on digestive process, where it enhances the length and weight of the small intestine (Yusrizal and Chen, 2003) along with regulation of villus structure, digestion of disaccharides, and absorption of glucose, leucine, proline and glycyl-sarcosine, in experimental animals (Buddington et al., 1999). In addition, it propels the production of gastrointestinal (GI) hormonal peptides and, consequently hormonal regulation of gastrointestinal motility as well as systemic metabolic processes (Roberfroid., 1996). Further inulin enhances bacterial biomass in proximal colon and increases the production of short chain fatty acids such as acetate, propionate, lactate, butyrate etc (Rycroft et al., 2001), which result in selective stimulation of growth of bifidobacteria and reduction in the number of Clostridium spp etc. Besides its bifidogenic effect, it also probably increases the counts of Clostridium coccoides – Eubacterium rectale, the clusters that contains most of the nonpathogenic butyrate-producing microorganisms, which might stimulate the production of short chain fatty acids. This fact has received acknowledgment by the administration of inulin supplemented foods in experimental animals which had depicted an antagonizing properties against various harmful bacteria (Roberfroid., 1998).

In human studies, fermentation of fructans leads to the selective stimulation of growth of the bifidobacteria in populations (Gibson and Wang 1994; Gibson et al 1995; Roberfroid et al 1995). The study by Gibson et al (1995) demonstrated a significant modification in gut microbiota of humans who consumed inulin rich diet for 15 days, which was characterized by enhanced bifidobacteria colonization, with reduced count of bacteroids, fusobacteria and clostridia in individuals (Table 6). Similar results were evident from adult human studies conducted in Europe, Japan and North America, where different daily doses of inulin were administered (Kleessen et al 1997). It has been postulated that the beneficial effect of inulin is attributable to the ability of bifidobacteria to change the colonic environment by inhibiting detrimental bacteria via the formation of bacteriocins, by blocking the successful competition for substrates or adhesion sites on the gut epithelium, and also by stimulating the immune system (Miller et al., 1989; Gibson et al 1995).

4. Conclusion

From the above discussion, it is clear that Indian diet compositions are rich in prebiotics. However, presently due to consumption of the western diet which are rich in refined carbohydrate and fats, there is a momentum towards the development of various unhealthy metabolic syndromes. Since the refined foods are known to alter gut microflora, they might be one of the major factors responsible for the sudden increase in the prevalence of metabolic diseases in the urban areas, especially targeting the adolescents. Since oligosaccharides as prebiotics are known to increase bifidobacterial colony in the intestine, which in turn can have a great positive impact on gut health by regulating the lipid metabolism and immune modulation (Fig.1). Formulation of food by including natural or synthetic oligosaccharides can be considered as one of the major guidelines for preventing the unhealthy metabolic diseases in India.

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Fig 1: Schematic Representation of Beneficial Effect of Prebiotics on Health and Nutrition Absorption

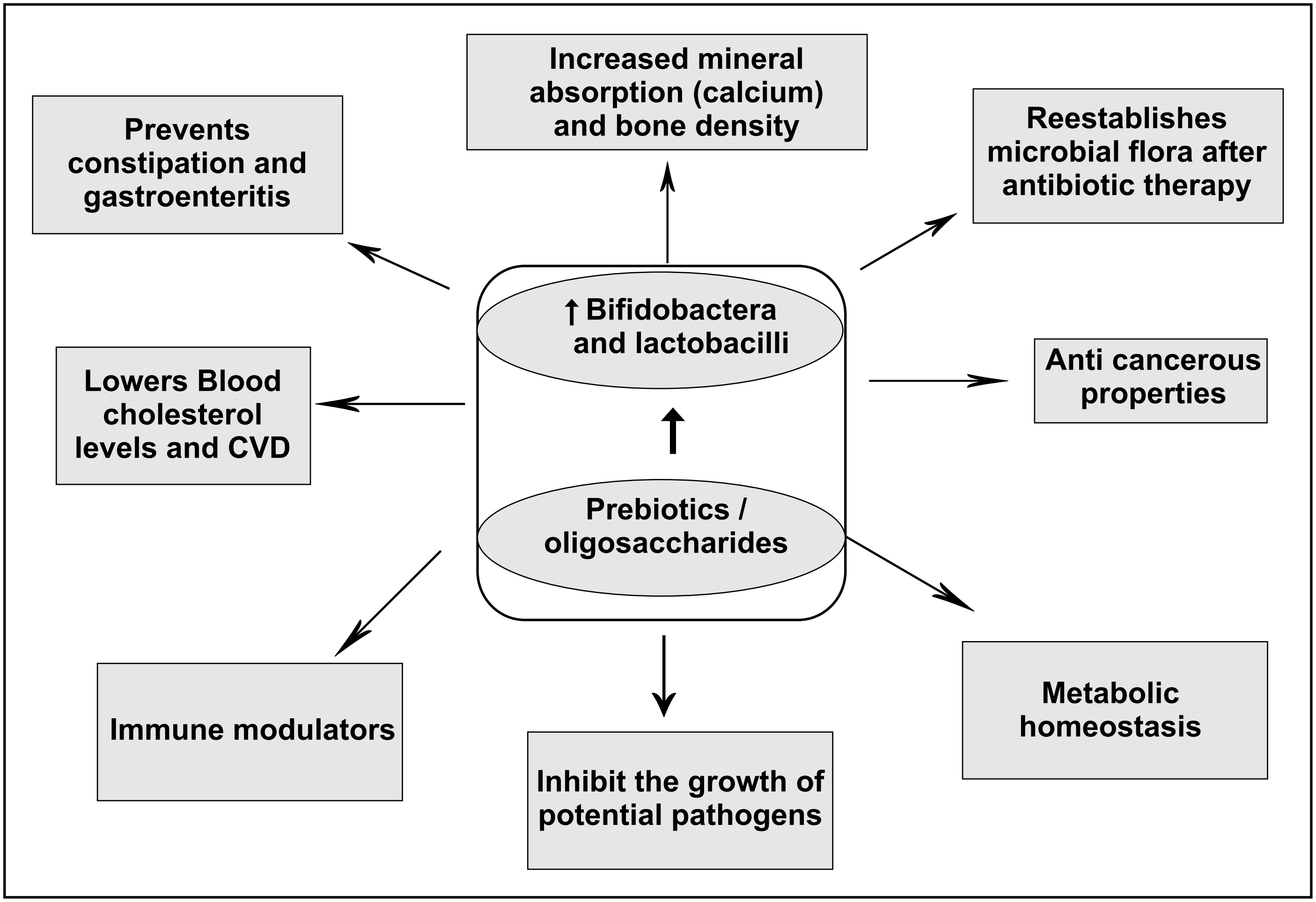

Table 1: The Natural Oligosaccharides/Prebiotics from Different Food Sources

Class	Source
Vegetables	Jerusalem artichokes (116mg/g)$, chicory (70% dry weight), garlic (32mg/g), onion (38mg/g), leek (30mg/g), shallots, spring onion, asparagus, beetroot (10mg/g), fennel bulb, green peas (7mg/g), snow peas, sweet corn (3mg/g) and Savoy cabbage(6mg/g).
Legumes	Chickpeas (12mg/g), lentils (11.5mg/g), red kidney beans (15mg/g), baked beans (1mg/g), soybeans (23mg/g).
Fruit	Custard apples (1.5mg/g), nectarines, white peaches (2mg/g), persimmon, tamarillo, watermelon (1mg/g), rambutan, grapefruit, pomegranate. dry fruits (eg. dates, figs)
Bread/Cereals / Snacks	Barley (7mg/g), wheat bran (5.3mg/g), wheat bread Oats (5mg/g)
Nuts and Seeds	Cashews (28mg/g), pistachio nuts (13mg/g)
Other	Human Breast milk*

Source: Longvah et al., 2017; Jantscher-Krenn E and Bode et al., 2012.Slimestad et al., 2010$*

Table 2: Types of Oligosaccharides as Prebiotics Naturally Available or Synthesized Commercially

Type of Oligosaccharide	Naturally Occurring	Synthesized Commercially
Fructo- oligosaccharides	Fruits and vegetables, onions, banana, garlic etc.	Synthesized from saccharose
Gakacto- oligosaccharides	Human milk	Enzymatic synthesis from lactose
Lactulose	—	Synthesized from lactose
Lactosucrose/glycosylsucrose	—	Synthesized from saccharose and lactose
Malto- oligosaccharide transfer from starch	—	Hydrolyzed or by glycosyl
Xylo-oligosaccharide	—	Hydrolyzed from polyxylons
Stacchyose, Raffinose	Soya bean	
Palatinose- oligosaccharide	—	Synthesis from starch
Gentio-oligosaccharide	—	Enzymatic synthesis from glucose
Cyclodextrins	—	Synthesis from starch

Source: Murphy., 2001.

Table 3: Percent of Fructooligosaccharides Content on Wet Weight basis from some Important Food Sources

Food Source	% content on wet weight basis
Banana	0.3–0.7
Rye	0.5–1.0
Garlic	15-20
Wheat	0.8–4.0
Chicory roots	15-20
Asparagus shoot	15-20
Onion	1.1–7.5
Dandelion	12.0–15.0
Dahlia	13.0
Burdock	3.6
Jerusalem artichoke	15-20
Salisfy	15-20

Source: Samanta et al., 2011 and Schaafsma et al., 2015

Table 4: Agricultural Crops Containing Prebiotics in Indian Foods

Crops	Fraction containing prebiotics action/precursor
Cereals: rice (0.8mg/g), Hulled Barley (0.7mg/g), Bajra (0.9mg/g), Jowar (0.3mg/g), wheat (5mg/g) and ragi (2mg/g).	Raffinose and arabinoxylan
Pulses and legumes : Green gram (12.9mg/g), Field pea, Black gram (13.8mg/g), Bengal gram (8mg/g), cow pea (1.2mg/g), lentils (11.5mg/g) and soya beans (21.7mg/g).	Raffinose and a-Galactosidase
Roots and Tubers: Beet root (1mg/g), carrot (1mg/g) ,Tapioca (2mg/g), Onion (38mg/g) and garlic (32mg/g).	Inulin and arabinoxylan
Nuts and seeds: Fenugreek (22.9mg/g), Almonds (1.5mg/g), cashews (28.4mg/g), mustard (9.1mg/g) etc	Raffinose and arabinoxylan

Source : Longvah et al., 2017; Samanta et al., 2007

Table 5: Approximate Average Daily Consumption of Prebiotics (gm/day) from Different Age Group of People from Rural and Urban Parts of India derived from National Nutrition Monitoring Bureau and Indian Food Composition Table Data

Consumption (based on Age group) (Yrs)	Cereals and millets	Pulses and legumes	Green leafy vegetables	Fruits and other vegetables	Roots and tubers	Oils and seeds	Others	Total
1-3	0.267	0.219	0.002	0.019	0.028	0.033	0.036	0.604
4-6	0.426	0.292	0.003	0.025	0.049	0.149	0.034	0.978
7-9	0.534	0.350	0.003	0.028	0.061	0.182	0.030	1.188
10-12 boys	0.614	0.379	0.004	0.034	0.071	0.265	0.031	1.398
10-12 Girls	0.587	0.365	0.005	0.030	0.067	0.232	0.027	1.313
13-15 boys	0.707	0.423	0.005	0.039	0.075	0.251	0.027	1.527
13-15 Girls	0.660	0.394	0.005	0.039	0.076	0.265	0.035	1.474
16-17 boys	0.787	0.423	0.005	0.042	0.079	0.347	0.024	1.707
16-17 Girls	0.705	0.423	0.005	0.040	0.075	0.298	0.031	1.577
Adult Men	0.775	0.467	0.006	0.048	0.093	0.430	0.030	1.849
Adult Women (NPNL)	0.797	0.452	0.005	0.044	0.057	0.331	0.031	1.717
Pregnant Women	0.722	0.496	0.006	0.055	0.080	0.414	0.029	1.802
Lactating Women	0.805	0.496	0.006	0.044	0.093	0.380	0.030	1.824
Average Rural Consumption	0.764	0.452	0.006	0.046	0.084	0.248	0.036	1.636
Average Urban Consumption	0.656	0.610	0.007	0.021	0.126	0.528	0.028	1.976

Table 6. Effect of Feeding 15 g/day of Sucrose, Oligofructose and Inulin on Percent Composition of Iintestinal Microflora in Humans.

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

Source: Gibson et al 1995

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