Probiotics, prebiotics and the human large bowel

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Summary

Most of the bacteria in the human gastrointestinal tract are found in the large bowel. In the past, these organisms have been viewed as largely harmful and they continue to be implicated in some pathologies. However, the accumulated evidence indicates strongly that some of them promote the normal physiological functions of the viscera through the short chain fatty acids (SCFA) formed by the fermentation of undigested carbohydrates. The presumption that intestinal microflora were deleterious has led to the development of probiotics, principally lactic acid bacteria, which could alter the colonisation favourably. However, the ingestion of probiotics does not seem to produce a sustained change in faecal probiotic numbers or SCFA. This has led to the further development of prebiotics which promote the activities of probiotics and also the beneficial indigenous flora. Prebiotics include oligosaccharides and resistant starch (RS) and possibly gums and uronic acids. While RS is believed to promote a favourable SCFA profile, this needs to be established for other carbohydrates. Lactic acid bacteria are present in relatively greatest number in milk-fed infants. Consideration of the faecal SCFA profile of such shows that the molar proportions of the major acids differ markedly from those in adults while other products (eg ethanol) are also found in quantities not present after weaning. This suggests that, in adults, favourable alterations in SCFA may be achieved best by consumption of prebiotic carbohydrates with or without probiotics.

Introduction

The role of the gut microflora in human health and disease is well accepted and the microbial colonisation of each region of the gastrointestinal (GI) tract makes its own particular contribution to disease risk. For example, colonisation of the stomach by *Helicobacter pylori* is thought to enhance the risk of specific pathologies. H pylori is found adhering to the gastric epithelium and is associated with reflux, gastritis, surface ulceration and cancer (1). However, the numbers of bacteria in the mouth, stomach and small intestine are very low relative to the $\sim 10^{13}$ organisms found in the large bowel of omnivorous animal species including humans (2). This population is very diverse and comprises over 50 genera and 400 species. This large population makes the large bowel a prime focus for any health-related interaction between the gut bacteria and the host although, historically, this interaction has been viewed rather unfavourably. Metchnikoff (3) propounded the idea that the luminal generation of waste products by the microflora had adverse health consequences ('intestinal autointoxication'). Consequently, large bowel bacteria were viewed largely as potential pathogens and they continue to be implicated in the aetiology of several important human large bowel pathologies including colorectal cancer, irritable bowel syndrome (IBS), and inflammatory bowel disease (IBD) (for an overall review see: 4). Putrefactive species could contribute to the initiation and promotion of colo-rectal carcinogenesis through the metabolism of luminal substances to potential toxins, mutagens and carcinogens. *Streptococcus bovis* has been implicated specifically in cancer of the large bowel but the connection has not been confirmed. *Inter alia* there is little evidence supporting a direct role for

this or other bacterial agents in initiating or promoting colorectal cancer. The same appears to be true for constipation and diverticular disease both of which relate principally to a sub-optimal fibre intake and can be prevented and managed by increasing fibre consumption (5). IBS is characterised by disordered gut motility and pain with bloating, diarrhoea and/or constipation and while it can affect the whole gut, its primary focus appears to be the large bowel. The condition appears to be to be unrelated to fibre intake while the role of the microflora remains largely undefined. IBD occurs in two main forms – ulcerative colitis (UC) and Crohn's disease. UC occurs mostly in the distal colon and is characterised by surface ulceration and bleeding while Crohn's disease may be found throughout the GI tract but particularly at the ileo-caecal junction. Again, the microflora have been implicated in both conditions but without conclusive proof (4,5).

The large bowel microflora, short chain fatty acids and colonic function

The negative perceptions of large bowel microflora have tended to obscure the fact that these organisms (and their metabolic products) are pivotal to the maintenance of normal human colonic physiology. In adults, these products include short chain fatty acids (SCFA), principally acetate, propionate and butyrate (6). Collectively, SCFA appear to promote colon function by enhancing the absorption of fluid and electrolytes (including Na^+ , K^+ and Ca^{2+}) and increasing colonic blood flow and muscular activity (7). Butyrate seems to be a preferred substrate for colonocytes; it and, to a lesser extent, propionate appear to promote a normal phenotype in these cells. SCFA are formed by the fermentation of dietary carbohydrates not absorbed in the small intestine and include non-starch polysaccharides (NSP, major components of dietary fibre), oligosaccharides (OS), some disaccharides (eg lactose) and starch which has escaped small intestinal digestion (resistant starch, RS) (6). There also appears to be a potentially important role for undigested protein (resistant protein, RP) in fermentation (8). While fibre was thought to be the dominant fermentative substrate it has become apparent that RS can make an equal or greater contribution. Of interest is the fact that RS fermentation may favour butyrate production and a recent report suggests that consumption of a particular RS by African natives led to a substantial rise in butyrate (9). The population studied is one at low risk of colo-rectal cancer and the food (stale maize porridge) is traditional in that group.

Modulation of the large bowel microflora – probiotics and prebiotics

The view that the normal colonisation of the gut was potentially harmful implies that a more favourable one is preferable. This might be achieved through the consumption of live organisms, specifically the lactic acid bacteria (LAB). These organisms are found in fermented milk products where their primary role is food preservation and the prevention of spoilage. However, within the gastrointestinal tract they could promote health through their metabolic activities and products. This is the central concept underpinning probiotics ie a specific colonisation of the human (or animal) gastrointestinal tract by one or several exogenous live bacterial species which act to improve health. Probiotic organisms commonly used in commercial foods include (among others) strains of *Lactobacillus* and *Bifidobacterium*. There have been clear demonstrations of effects of the ingestion of probiotics eg immune system stimulation, taxonomic modulation of the enteric microflora and decreased faecal bacterial enzyme activities (6). While it has been suggested that they are useful in the prevention and/or treatment of intestinal and systemic disorders including infantile and travellers' diarrhoea, lactose maldigestion,

hypercholesterolemia, constipation, and colon and other cancers, these actions remain to be established (10).

Effective use of probiotics in humans is limited by the apparent refractoriness of the human gut microflora to exogenous bacteria, resulting in only transient colonisation which could reflect loss of viability on passage through the stomach and small intestine. This drawback has led to the development of the concept of prebiotics which are defined as "non-digestible food ingredients that beneficially affect the host by selectively stimulating the growth and (or) activity of one or a limited number of bacterial species already resident in the colon, and, thus, attempt to improve host health" (2). Perhaps one of the best known of these agents is fructo-oligosaccharide (FOS) which has been shown to enhance faecal excretion of probiotics in humans. FOS appear to act by providing metabolic substrates for LAB on transit through the intestine. Starch and NSP have been regarded as unlikely prebiotics, largely because many of the colonic bacterial species are incapable of metabolising these carbohydrates. However, studies in experimental animals and in vitro suggest otherwise. Alginates (11) and acacia gums (12), which are used widely in the food industry, appear to be prebiotics while RS also has prebiotic potential. During studies with a high amylose RS it was noted that the granules developed a particular etching pattern on passage through the upper gut of either pigs or humans (4). This resulted in surface pitting and it was thought that this could enhance the viability of probiotics by providing a surface for physical adhesion. This adhesion has been confirmed *in vitro* and animal trials showed greater faecal bifidobacteria excretion when they were fed live *Bifidobacterium longum* with this RS. There was no effect of the probiotic on SCFA levels but the RS increased SCFA and butyrate excretion. When FOS and RS were fed separately, both stimulated faecal bifidobacteria excretion and when fed together their actions were additive. Only RS raised faecal SCFA excretion. There is some evidence from rats to suggest that the effects of FOS on faecal excretion of LAB may be relatively transient while changes in SCFA are of longer duration (13). It seems that the duration of the feeding period may become an important issue in human trials.

Lactic Acid Bacteria, SCFA and prebiotics

Probiotics do not seem to effect sustained modification of faecal SCFA. LAB are found in the human gastrointestinal system in greatest relative abundance in milk-fed infants (2). These babies have a profile of products which is quite unlike that in adults. Acetate is the predominant SCFA (as in adults) but propionate is present at much lower concentrations while butyrate is virtually absent. Other fermentative products such as ethanol, formate, succinate and lactate appear in infant faeces (but not in quantity in adults). It has been suggested (4) that consumption of LAB *per se* need not alter propionate and butyrate but can be achieved quite effectively through changing the supply of fermentable carbohydrate ie consuming prebiotics. Favourable changes including enhancement of Ca^{2+} absorption (14) and accelerated recovery from infectious diarrhoea (15) have been shown following the ingestion of these carbohydrates without any additional change. This indicates the potential value of these compounds but it is expected that concomitant ingestion of probiotics will confer the separate benefits associated with these organisms.

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