The Journal of Nutrition, Health & Aging©
Volume 16, Number 4, 2012

PROBIOTICS IN THE GASTROINTESTINAL DISEASES OF THE ELDERLY

G. MALAGUARNERA[1], F. LEGGIO[2], M. VACANTE[2], M. MOTTA[2], M. GIORDANO[2], A. BIONDI[3], F. BASILE[3], S. MASTROJENI[1], A. MISTRETTA[4], M. MALAGUARNERA[2], M.A. TOSCANO[1], M. SALMERI[1]

1. Department of Microbiology, University of Catania, Italy; 2. Department of Senescence, Urological and Neurological Sciences, University of Catania, Italy; 3. Department of Surgery, University of Catania, Italy; 4. Department of Hygiene and Public Health, University of Catania, Italy. Address Correspondence: to Mariano Malaguarnera, Department of Senescence, Urological and Neurological Sciences, University of Catania, Italy, Phone number +39 95 7262008, Fax number +39 95 7262011, E-mail: malaguar@unict.it

Abstract: Changes of the gut microflora in elderly appear to involve a reduction in numbers of healthy bacteria (lactobacilli and bifidobacteria) and an increase in numbers of potentially pathogenic species. These changes are generally described as gastrointestinal disorders and infections. This review analyses benefits of probiotics in old people, with particular interesting for the latest researches relevant to elderly people, e.g. trials examining enteric infections, antibiotic-associated diarrhea and Clostridium difficile associated diarrhea, functional bowel problems (constipation and irritable bowel syndrome), inflammatory bowel diseases, stimulation of the immune system and prevention of cancer. A growing number of researches indicates that some probiotic strains may help to maintain the health in old people, suggesting both health and cost-saving benefits in offering fermented dairy products. These benefits include: establishment of balanced intestinal microflora; improving colonization resistance and or prevention of diarrhea; reduction of fecal enzymes; reduction of serum cholesterol; reduction of potential mutagenes; reduction of lactose intolerance; synthesis of vitamins; predigestion of proteins.

Key words: Probiotics, elderly, diarrhea, constipation.

Introduction

The human adult microflora is colonized by approximately 10^14 microbial cell, about 10 times more than all tissue cells of the body. This high metabolic rate suggests important regulatory effects on body functions, especially in the colon where the greatest concentration of up to 5x10^11 bacterial cell per g is found (1). The human microbiota is stable at different anatomic locations along the gut, but absolute numbers vary greatly, ranging from 10^11 cells/g content in the ascending colon to 10^7 in the distal ileum and 10^3 in the proximal ileum and jejunum. Anaerobes are more abundant that aerobes in the bacterial community and a majority of the population are representatives of two divisions: the Bacteroidetes and Firmicutes. At birth the gut is sterile and is colonized immediately, although there are marked variations in microbial composition between individuals. More than 400 species are included: Gram-positive, anaerobic genera Bacteroides, Eubacterium, Bifidobacterium, Peptostreptococcum, Streptococcum, Lactobacillus, Fusobacterium, Ruminococcus, Clostridium and Escherichia. Some of these bacteria are potential pathogens and can be a source of infection and sepsis under some circumstances, when the integrity of the bowel barrier is physically or functionally breached. These bacteria maintain the integrity of gut mucosa and the production of short chain fatty acids (SCFA) in a favorite ratio (2, 3). The enormous numbers and diversity of microorganisms in the human intestine contribute to a diverse set of functions, which complement the host for important features such as digestion of complex carbohydrates. The interaction between animal and bacterial cells is very important in the human gastrointestinal tract. The bacterial microbiota has established multiple mechanisms to influence the human host in a beneficial fashion and maintain their stable niche. The human host coevolved with a normal microbiota developed and optimized immune mechanisms. Both the impact of gut microbiota on disease and the impact of disease on gut microbiota need to be investigated to establish a good treatment in the gastrointestinal disease. The gut microflora exerts a considerable influence on host biochemistry including enzymatic activity of intestinal contents, oxidation-reduction potential of luminal contents, short chain fatty acid production in the lumen, host physiology, host immunology, modification of host-synthesized (4). Several studies showed a decrease in Bifidobacteria and an increase in clostridia, lactobacilli, streptococci and Enterobacteriaceae in the gastrointestinal tract of elderly people (5-8). The genera Bifidobacterium are the dominant probiotic bacteria inhabiting the distal jejunum ileum and large intestine of humans and other warm-blooded animals. Bifidobacterium spp has many beneficial effects on human health, including: immunomodulation, reducing serum cholesterol, promoting lactose digestion and protecting against colon cancer. Changes in the microflora could alter the metabolic environment of the colon with important modifications in the concentration of healthy substances that may alter the motor and secretor functions of the bowel. So, changes in the microflora increase susceptibility to gastrointestinal functional disorders, infections, inflammation or cancer. Conversely, probiotics can promote the homeostasis of the colonic microbiota (9). Recent studies have suggested that the gut microbiota may have a role in gastrointestinal diseases through the regulation of energy metabolism by several mechanisms (that is, energy harvest from the diet, regulation of fat storage, modulation of afferent gastrointestinal peptide hormones, induction of metabolic endotoxemia) (10).

Received February 15, 2011
Accepted for publication May 5, 2011

Therapeutic potential and health benefits of probiotics

Probiotics are: 'live microorganisms which when administered in adequate amounts, confer a health benefit on the host" (11). Probiotics work by positively influencing the composition and activity of the micro-organisms in the gut and helping to maintain a beneficial balance of intestinal micro-organisms. Probiotics advantages are strain specific. Good products should contain the correct probiotic strains. The main therapeutic benefits of probiotics are prevention of diarrheal diseases, enhancement of immunity against intestinal infections, improvement of gastrointestinal motility disorders and inflammatory intestinal disorders, immune enhancement, prevention of colon cancer (12). Mechanisms of these effects include a decrease of gut pH, production of antimicrobial substances, agglutination of harmful bacteria (13, 14), maintaining of the gut mucosa, competition for substrates or receptors on the wall of the mucosa, release of intestinal-protective substances such as arginine and short chain fatty acids (SCLA) (15, 16), neutralization of toxic or cancerogenic molecular, modulation of immune system (17), promotion of peristalsis (18). Foods containing probiotics have increased over the years. As effects of probiotics have been shown to be strain specific, it is essentially to report health benefits for single strains or for a mix of strains separately (19-21) (Table 1).

Probiotics and immunomodulation

Older people are more susceptible to illnesses due to weakened immune system. In fact the immune system of elderly people is less able to produce antibodies and mount a cellular immune response (T-cells). Elderly people may have reduced levels of circulating T-cells that can kill viral infected cells, or other activated cells of the adaptive immune response. Moreover the activity or number of Natural Killer (NK), that target virally infected cells and tumors are reduced (22). Modifications in bowel microbiota probably also contribute to the chronic inflammatory status of the elderly and to their increased frailty and immunosenescence (23, 24). Probiotic bacteria and their cell-wall components (peptidoglycanes, lipopolysaccharides), DNA and metabolites are shown to have immunomodulatory properties. One mechanism of probiotic activity is immune modulation; so, it might be expected that probiotics could slow the immune decline and support immune function in older people. Human and in vitro studies with probiotics, explore both the immune benefit and mechanism of activity, using a range of immune markers. These have included immunoglobulins (IgA, IgG: antibodies), Toll-like receptors (antigen recognition sites), mucosal integrity markers, innate immune markers (NK cells, phagocytes, neutrophils, monocytes), cytokines (pro- and anti-inflammatory: the proteins that communicate with and activate other immune cells) and viral response. In a large controlled study involving 360 people over 60 years, the effect of a 3-weeks intervention of a fermented milk containing yogurt cultures and the probiotic L. casei DN-114001 showed that the incidence of winter infections was no different, but the duration of all pathologies was significantly lower in the probiotic group (7.0-3.2 days) compared to the control group (8.7 - 3.7 days) (25). A double-blinded trial involving 24 elderly patients that received enteral nutrition, valued the effects of fermented milk product containing Lactobacillus johnsonii La1 (LC1) for 12 weeks. The probiotic group had significantly less days with infection at the end of the trial, a drop from 15.4% days with infection to 5.7%, and this reduction was larger than that seen for the control group (26). This means that the immune system may be more efficient at warding off infections when probiotics are administered. A variety of effects of probiotic strains on immune system function have been reported. The species Lactobacillus acidophilus, L. casei, L. plantarum, L. delbrueckii, L. rhamnosus, stimulate immune system (25).

Antibiotic-associated diarrhea

Acute diarrhea can have bacterial or viral causes, and is a common problem especially in children in developing countries. Several investigations have shown that the intake of S. boulardii reduced the duration of acute diarrhea (although one study did not find any effect) (27). So, Lactobacillus rhamnosus GG (LGG) was effective in reducing duration of diarrhea. However, a sub-analysis showed that the effect of LGG was only important in children of Western countries, where rotavirus is the main cause of acute diarrhea; it was

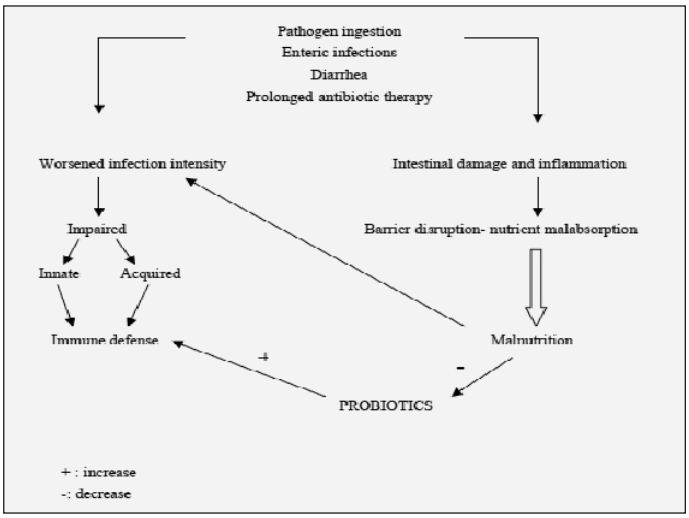
Table 1
Mechanisms of probiotics activity

Modulation of intestinal ecosystem	Modulation of immune resistance	Improved colonization resistance
• beneficial bacteria • short chain fatty acids • gut pH • faecal microbiota mass • stimulation of gut peristalsis • harmful substances	• stimulation of local and systemic systems • enhanced defence response (IgA, CD4+ & CD8+ T cells, NK cell activity, adjuvant effect) • downregulation of inflammatory and allergic response	• competitive exclusion (e.g. for nutrients, adhesion sites) • harmful microbial species • epithelial cell mucin synthesis • enhanced intestinal barrier function

increase; decrease

inefficacy in children in developing countries (27). Studies on the healthy effects of probiotics on traveler's diarrhea showed no good results because there were differences between probiotic strains, the traveled countries, the local microflora, specific (eating) habits of the travelers, and the method of intake of the probiotic. However, some studies described less or shortened episodes of diarrhea in people consuming the probiotic (28-30), others found no advantages (31, 32). Intake of S. boulardii reduced traveler's diarrhea. Data on the probiotic LGG were unclear. In one study the risk of developing traveler's diarrhea was lower in the treatment group compared with the placebo group (28); in other, there were no differences between treatment group and placebo group (29). Also the efficacy of S. boulardii was more important in some areas (32). Antibiotics kill both healthy and pathogenic gut bacteria, alter the intestinal microbiota increasing the risk of opportunistic infections. Antibiotic-associated diarrhea (AAD) can affect 5% to 25% of patients taking antibiotics (33, 34). Intake of some probiotic strains before and during antibiotic treatment in most studies reduced the frequency and/or duration of episodes of antibiotic-associated diarrhea and the severity of symptoms (35, 36). For example, S. boulardii and LGG are the most effective strains for preventing AAD, and lowering the risk of AAD. Actually, mixtures of probiotic strains including L. rhamnosus, L. casei, L. acidophilus, L. bulgaricus and Bifidobacterium Bb-12 have been studied (35-37). Probiotics have been used Clostridium difficile that accounts for 20% to 25% of cases of AAD, causing over 95% of cases of pseudomembranous colitis (34). Although some studies showed that probiotics decrease the risk of Clostridium difficile associated diarrhea (CDAD), in many studies the number of patients was no statistical sufficient (38). A yogurt containing L. casei DN-114 001, S. thermophilus and L. bulgaricus significantly decreased the risk of developing CDAD in hospitalized patients (36). Another study showed a significant difference between S. boulardii and placebo, both in combination with antibiotics, in patients with recurrent Clostridium difficile disease (CDD), but not acute CDD (29). A mix of L. acidophilus and B. bifidum on occurrence of diarrhea and CDAD did not reduce the frequency of episodes both in patients and in the placebo group (40). Various species may be effective in the prevention of diarrhea: in prevention of traveler's diarrhea are saccharomyces bulgarius or mixture of L. acidophilus, B. bifidum, Streptococcus thermophilus, L. bulgarium; in prevention of rotavirus diarrhea are L. rhamnosus, B. bifidum; in prevention of Clostridium difficile diarrhea are L. rhamnosus, Saccharomyces bulgaricus; in prevention of diarrhea are L. Acidophilus, L. rhamnosus, B. bifidum (Figure 1).

Figure 1
Pathophysiology of probiotics in gastrointestinal activity



Functional bowel diseases

Functional bowel diseases in elderly include irritable bowel syndrome (IBS) and constipation. IBS is a chronic disease that is characterized by intermittent abdominal pain, diarrhea and/or constipation and other gut symptoms including flatulence and bloating in the absence of structural alterations in the intestine (41). People with IBS usually describe incomplete evacuation/rectal hypersensitivity, as well as urgency, which are increased in diarrhea-predominant IBS (42). The prevalence of IBS is estimated about 3–25% of the general population (41). There is evidence that bacteria and bacterial components may play a role in aggravating IBD (43). The pathophysiology of IBS is multifactorial (41), with bacterial overgrowth in the small intestine in up to 78% of patients (44). So, probiotics can alleviate symptoms of IBS, and their efficacy has been showed in most studies (45). In a meta-analysis, the results of randomized, controlled, blinded trials were examined to determine the efficacy of LGG in 3 studies and B. infantis in other 2 studies (32). Compared to placebo, probiotics did not reduce significantly abdominal discomfort, bloating and altered bowel habits, but were effective to decrease the risk of abdominal pain. However, insufficient data analyzed individual IBS symptoms or the efficacy of individual probiotic strains. The treatment periods varied between 2 and 24 weeks, in most of the 20 studies was between 4 and 8 weeks (85%) (32). Another study showed that a probiotic mix (B. longum LA 101, L. acidophilus LA 102, Lactococcus lactis LA 103, St. thermophilus LA 104), increased satisfactory relief of overall IBS symptoms and of abdominal discomfort/pain at fourth week, but also similar effects were showed in the placebo group. Abdominal pain improved significantly during the study in both groups, independent of IBS category (constipation-predominant, diarrhea-predominant or alternating). The

difference between the first week and the fourth week was significantly higher in the treated patients. Overall, the frequency and consistency of stools did not differ significantly between the probiotic and placebo groups. In the group of subjects reporting constipation, the frequency of stools was significantly higher in the treated group from the first week of treatment (46). Sinn et al. examined the effect of L. acidophilus strains SDC 2012 and 2013 on IBS symptoms. They observed a significant improvement in the probiotic group at week four compared to the placebo group, based on abdominal pain scores from 0 (good) to 10 (bad) (47). A study showed effects on IBS symptoms of a mixture of probiotic strains consumed as a milk drink. The decrease of distension and abdominal pain score was significantly more pronounced in the probiotics group compared to the placebo group. No differences were found in the percentage of soft stools, hard stools or diarrhea (48). Actually, some evidences show good results for the use of probiotics in the treatment of IBS. However, available studies use different probiotics strains and there is not evidence to make any firm conclusions about individual strains or dose (32, 46-48). In elderly, constipation is a common condition, characterized by a constellation of symptoms proposed by the present of two or more symptoms fixed the "Rome criteria III". These criteria include 12 weeks or more of symptoms in the least year: hard or lumpy stools, painful and strained defecation, a sense of incomplete evacuation, the need to use manual manoeuvres to pass stool or sense of anorectal obstruction with >25% of bowel movements and /or <3 bowel movements/week, no evidence for IBS (49). Studies about probiotics have shown improved colonic transit times, both in a healthy population and in constipated patients (49). Probiotics reduce the pH in the colon by the bacterial production of short chain fatty acids (butyric acid, propionic acid, and lactic acid). A lower pH enhances peristalsis in the colon and, subsequently, might decrease the colonic transit time (50). A study reported a reduction of transit time after 10-day intervention with Bifidobacterium animalis DN-173 010 (51), while other described an increase in stool frequency after a 2-week intervention with Lactobacillus casei Shirota (52). A more recent study has associated the use of a synbiotic combination of lactitol and Lactobacillus acidophilus twice daily by healthy elderly subjects, with increased stool frequency, as well as other indications of positive results in the intestinal microbiota composition and mucosal function (53). Contrary, constipation was not improved, in a trial with Lactobacillus rhamnosus GG (54); thus, it is important to know which probiotic strains have improved bowel functions. Beneficial changes in the microbial metabolism in the colon of people with gut function disorders have also been reported (55). The main metabolic markers of probiotic benefit include short chain fatty acid levels (SCFAs), fecal enzyme activity and certain toxic or carcinogenic compounds (55). SCFAs in the gut, for example, stimulate host receptors for gut motility and the immune response, and regulate mucosal gene expression. The SCFA butyrate, an important energy source for colon cell, is involved in regulation of their growth. Probiotics have also been associated with production of substances that inhibit tumor growth, neutralization of harmful molecular in the gut, and inhibition of species that can produce promoters, mutagens and carcinogens (56). So, phenol, cresol and ammonia are metabolites produced by colonic fermentation. A significant reduction of such substances was associated with consumption of a probiotic containing L. casei Shirota, in double-blind, placebo, controlled trials with healthy subjects (57, 58), and similar results have been reported with other probiotics (59-63). Likewise, data suggest that Bifidobacterium animalis, lactobacillus casei Shirota, L. acidophilus, L. paracasei, L. rhamnosus and Propionibacterium freudenreichii might increase the frequency of defecations and reduce the time of bowel transit (50).

Diverticular disease

Diverticular disease (DD) of the colon is very common in Western countries and its prevalence increases with age and up to an estimated 20% of patients may manifest clinically-relevant illness in their lives (64, 65). Uncomplicated diverticular disease is defined as the presence of diverticula in the absence of complications such as fistula, perforation, obstruction and/or bleeding. The pathophysiology of diverticular disease is complex and relates to abnormal colonic motility, changes in the colonic wall, chronic mucosal low-grade inflammation, imbalance in colonic microflora and visceral hypersensitivity. Moreover, there can be genetic factors involved in the development of colonic diverticula (66, 67). The rationale for the use of probiotics is to promote the production of antimicrobials, competitive metabolic interactions with pro-inflammatory organisms, and inhibition of adherence and translocations of pathogens. They may also enhance mucosal defense at the levels of immune and epithelial function, such as decreasing several pro-inflammatory cytokines, but also alleviating intestinal inflammation and normalizing gut mucosal dysfunction (68). So, probiotics can re-establish the physiological bacterial flora, which in the presence of diverticular disease may be altered by the reduced colonic transit time and stasis of fecal material within the diverticula. There are little data on the treatment of uncomplicated DD with probiotics. First data about the use of probiotics in DD come from a prospective observational trial on the prevention of complications after acute diverticulitis. All patients with a post-diverticular stenosis of the colon were treated sequentially with rifaximin and lactobacilli for a period of 12 months. This treatment proved to be effective in preventing symptom recurrence and complications (69). The effectiveness and safety of mesalazine, with or without Lactobacillus casei, in preventing recurrence of symptomatic diverticular disease of the colon was observed in a multicenter, prospective, randomized, open-label study. The 88.2% of patients were symptom free after the 12th month of treatment,

so the administration of mesalazine, with or without Lactobacillus casei, is effective in preventing recurrence of symptomatic uncomplicated diverticular disease of the colon, and their association is more promising in this field (70). In other study the efficacy of probiotic treatment as a single therapy for uncomplicated DD has been assessed comprising a very small group of patients. A non-pathogenic E. coli strain was given for a mean period of 5 weeks after a course of treatment with an intestinal antimicrobial and absorbent, resulting in a significantly prolonged remission period and important improvement of all abdominal symptoms (71). Prebiotics are dietary substances which stimulate the growth and metabolic activity of beneficial enteric bacteria, especially Lactobacillus and Bifidobacterium species (72). Inulin, fructose oligosaccharides, lactulose, germinated barley extracts and psyllium fibre have been shown to promote the growth of luminal Lactobacillus and Bifidobacterium species and butyrate production (73). Thus, the optimal treatment of uncomplicated DD might be an initial course of antibiotics to normalize the gut flora followed by a combination of a probiotic to prevent relapse, and prebiotic to maintain growth of protective bacteria. In a prospective, randomized, open-label study, Lamika et al. evaluated the effectiveness and tolerability of a symbiotic mixture preventing recurrence of symptomatic diverticular disease of the colon and, in particular, in those patients presenting a constipation-predominant pattern. So, forty-six patients, were enrolled in a 6-month follow-up study. The intake of Lactobacillus acidophilus 145 and Bifidobacterium spp. 420 was regarded as "effective" or "very effective" in more than 78% of the patients altogether ($p<0.01$ vs baseline values) (74). In conclusion, several studies showed the effectiveness of probiotics in preventing recurrence of symptomatic uncomplicated diverticular disease of the colon. However, further studies are needed.

Inflammatory bowel diseases

Bacterial flora has been suggested to play an important role in human inflammatory bowel diseases (IBD) (75). Several species of the microflora may invade the mucosa after induction of colitis causing transmural inflammatory lesions (76) (Table 2).

Table 2
Anti–inflammatory effects of probiotics

a) Compete with and displace pathogenic strains in small intestinal bacterial overgrowth
b) Antibacterial effects mediated by bacteriocins
c) Modify inflammatory pathways induced by bacteria involved in intestinal bacterial overgrowth
d) Ameliorate intestinal barrier function
e) Enhance integrity of the intestinal epithelium
f) Stimulate release of immunoglobulin A
g) Inhibit production of proinflammatory mediators
h) Induce anti –inflammatory responses

Celiac disease

Celiac disease, also known as celiac sprue or gluten-sensitive enteropathy, is a food hypersensitivity disorder caused by an inflammatory response to wheat gluten and similar proteins of barley and rye (77). Pathogenesis of celiac disease involves interactions between genetic, immunological, and environmental/technological factors. Human leukocyte antigen (HLA) DQ2 (or DQ8) molecules of antigen-presenting cells bind and submit gluten peptides to the lamina propria of CD4+ T cells. The latter trigger the T helper 1 (Th1)-based immune response, with the consequent synthesis of gamma interferon (IFN-γ). Among the main dietary proteins, gluten is unique in that it contains about 15% proline and 35% glutamine residues (78). The high concentration of glutamine and, especially, proline prevents the complete degradation by human gastric and pancreatic enzymes and results in the build up of oligopeptides in the small intestine that are resistant to further proteolysis and toxic to genetically predisposed celiac disease patients. Actually, analyses revealed the presence of more than 60 immunogenic oligopeptides from gluten of the Triticum species (79). The length of these oligopeptides may vary from 7/9 to 91 amino acid residues, and the common feature is represented by the high number of glutamine and proline residues. The most important example of such oligopeptides is the 33-mer epitope. The 33-mer epitope was identified as one of the main stimulators of the inflammatory response to gluten. This epitope was shown to be a potent inducer of gut-derived human T-cell lines (80). A strict, lifelong gluten-free diet is the only accepted treatment for celiac disease. Nevertheless, complete exclusion of dietary gluten is difficult due to the ubiquitous nature of this protein, cross-contamination of foods, inadequate food labeling regulations, and social constraints. Furthermore, gluten-free food provides inadequate supply of fibers, minerals, and vitamins and excess calories in the diet and exhibits poor sensory properties (81, 82). Alternative therapeutic options included the proposed oral supplementation with probiota (83). It was demonstrated that Lactobacillus casei enhanced the CD4+ T cell-mediated, Th1-like intestinal sensitivity to gliadin in a model of intestinal gliadin hypersensitivity in DQ8 tg mice characterized by a Th1-like phenotype (84) So, dendrites cells (DCs), that play an important role in immunological responses, have been considered as main target of the modulatory activity of probiotics. Along with antigen uptake and processing, dendritic cells can promote the development of unprimed, naive T cells toward Th1, Th2, or unpolarized T cell responses (85, 86). Three Lactobacillus species and B. lactis exposure altered the immature phenotype of DCs and this finding was independent of which strain was used (83). L. fermentum and L. plantarum were able to stimulate the LPS-inducible IL-12 in mature bone-marrow dentritic cells, a critical Th1-skewing cytokine that elicits IFN-c production by T cells and by NK cells (87). Moreover, IL-10, an anti-inflammatory cytokine that suppresses IL-12 production was found low both in unstimulated and LPS-induced dentritic

cells (88). Incubation of BMDCs with any examined probiotic strains did not elicit its expression. On the other hand, all strains were able to induce high levels of TNFa in immature (B. lactis, L. plantarum, L. paracasei) or in mature (L. fermentum, B. lactis) dendritic cells. The activity of lactobacilli is due to the cytotoxic action of TNF-a and IL-1 produced by macrophages (89,90). So, L. casei enhances the gliadin-specific IFN-c expression both at the systemic and intestinal levels. However, the effect was restricted to this strain (84). In conclusion, data showed the potential of probiotic strains to stimulate the immune system, by acting on cellular components of the innate and/or the adaptive responses in a model of gluten sensitivity. However, these activities are not appropriate for therapeutic application in celiac disease, but only in conditions that require enhancement of this specific immune response, as in cancer therapy.

Crohn's disease

Crohn's disease (CD) is one of the two main types of IBD. CD can affect any part of gut tract, mainly the lower small intestine. It is characterized by patchy, transmural inflammation. CD can be defined by location (terminal ileal, colonic, ileocolic, upper gastrointestinal) or by pattern of disease (inflammatory, fistulating or structuring). The efficacy of probiotics in preventing recurrence of CD after surgery was examined by randomized, double-blind, placebo controlled studies. Two studies used Lactobacillus johnsonii LA1 (91, 92), and one used the strain L. Rhamnosus GG (LGG) (93). In all three studies, there was no significant difference in recurrence rate between the treated group and placebo group (91-93), although Marteau et al. described a lower but non-significant recurrence rate in those treated with LA1 (91). Another study tested Saccharomyces boulardii in addition to standard treatment (mesalazine) in patients with CD who were in remission. After 6 months of treatment, 6 of 16 patients receiving mesalazine relapsed, whereas only 1 of 16 patients also receiving S. Boulardii relapsed (94).

Ulcerative colitis

Ulcerative colitis (UC) is an inflammatory bowel disease limited to the colon and it is characterized by diffuse mucosal inflammation. UC can affect any part of the colon (95). In UC, short-term treatment with an enteric-coated preparation of broad – spectrum antibiotics reduced mucosal release of cytokines and was effective in reduction of inflammatory activity than were intravenous steroids (96). In two studies, the efficacy of Escherichia coli Nissle (EcN) in maintaining remission in patients with UC showed no difference when compared to the gold standard treatment (mesalazine) (97, 98). Another trial studied a combination of different probiotic strains (Bifidobacterium breve strain Yakult, B. bifidum strain Yakult and L. acidophilus) taken over a period of 12 weeks. The clinical activity index (CAI) score of at least three points (overall score maximum 21 = highest activity), was higher in the probiotic group (70%) compared to the placebo group (33%), although statistical power is not achieved, probably due to the small number of patients. The number of patients achieving remission did not differ significantly between the probiotics and placebo group (40% and 33%, respectively). The average CAI score in the probiotics group is significantly lower at week 12 compared to the placebo group (99). One small unblended study showed that patients having daily fermented milk containing B. breve Yakult, B. bifidum Yakult and L. acidophilus YIT 0168 (1010 CFU/day) in addition to standard treatment had a lower rate of exacerbation of UC compared to those on standard treatment only (27% and 90%, respectively). However, the study was small and exacerbation was measured on the basis of self-reported clinical symptoms (100).

Pouchitis

Pouchitis is a complication in patients undergoing j-pouch (ileal pouch-anal anastomosis) surgery for UC (101). Studies demonstrated that the microflora in the pouchities plays a fundamental role; in particulary, people with pouchitis have fewer beneficial bacteria, like lactobacilli and bifidobacteria in gut. So, intake of these beneficial bacteria can be effective in maintaining remission in patients with pouchitis (95). Around 40–60% of patients who undergo ileal pouch surgery for UC suffer from pouchitis that is the most common long-term complication in patients undergoing surgery for UC (95, 101, 102). The causes of pouchitis are unclear, but it is possible that the microflora in the pouch plays a role in the abnormal mucosal immune response (101, 102). The efficacy of VSL#3 (1VSL#3 contains 300 billion viable lyophilized bacteria per gram of four strains of Lactobacilli (L. casei, L. plantarum, L. acidophilus, and L. delbrueckii ssp. bulgaricus), three strains of Bifidobacteria (B. longum, B. breve, and B. infantis) and one strain of St. salivarius ssp. Thermophilus) in the maintenance of remission in patients suffering from chronic relapsing pouchitis, and recurrent or refractory pouchitis, was valued in two studies (103, 104). In the study by Gionchetti et al. all patients receiving placebo (n = 20) relapsed within 4 months, whereas 85% (17/20) of those receiving the probiotic mix were still in remission after 9 months ($P < 0.001$). In the study of Mimura et al. 17 out of 20 (85%) patients in the probiotics group were still in remission after 12 months, whereas only one of 16 (6.3%) in the placebo group remained in remission ($P < 0.001$). As a consequence quality of life significantly deteriorated in the placebo group but showed a slight increase in the VSL#3 group. In another study, Gionchetti et al. examined the efficacy of VSL#3 compared with placebo in the prevention of onset of pouchitis during the first year after surgery. In this study, significantly fewer treated patients developed pouchitis (2/20, 10%) compared with the placebo group (8/20, 40%; $P < 0.05$). The median Pouchitis Disease Activity Index score (PDAI; 0 lowest activity, 18 highest activity) significantly increased in the placebo group but did not significantly increase in the VSL#3 group (105). A randomized,

double-blind, placebo-controlled trial showed no differences between patients undergoing surgery for UC that intake Lactobacillus rhamnosus GG (LGG) and placebo group. After 3 months, no differences were found in the mean pre- or post-treatment PDAI scores between LGG and placebo group. The authors concluded that, as primary therapy, LGG was not effective for the clinical improvement of pouchitis (106).

Prevention of colon cancer

Intestinal bacteria could play an important role in initiation of colon cancer through production of carcinogens, cocarcinogens or procarcinogenes. Probiotic bacteria can have cancer-preventing properties. Probiotics have also been associated with production of compounds that inhibit tumor growth, neutralization of harmful substances in the gut and inhibition of species that can produce promoters, mutagens and carcinogens (56). Probiotic effects have been studied on the colon carcinoma, the most frequent cancer of the gastrointestinal tract in the Western industrial nations (107). Several mechanisms have been studied as a cause of these effects: inhibition of tumor-growth and proliferation of tumor cells by glycopeptides and cytotoxic metabolites of lactobacilli; decrease of (pro)carcinogenic, mutagenic, and genotoxic substances (aflatoxines, nitrosamines) and cancer-promoting enzymes (nitro-azoreductase, glucuronidase) in the bowel due to changes of the gut flora, a reduction in pH, chemical modification, and ad- and absorption by the bacteria; antimutagenic properties of probiotics and probiotic milk products; strengthening of the immune system and stimulation of the production of the tumor-necrosis-factor (TNFα) by macrophages (22). Use of probiotic bacteria with oligosaccharides could promote bacterial growth in the colon and increase great quantities of short chain fatty acids such as butyrate, which has been shown to have antitumor effects at the cell level (108) However, data are insufficient to evidence the health benefits of probiotics, such as prevention of colon cancer (109). Nevertheless, positive effects were also described for other types of cancer. A large Japanese case control study showed that the habitual intake of lactobacilli and especially L. casei Shirota may reduce the risk for bladder cancer in the Japanese population (110). However, only few epidemiological investigations have been performed concerning other probiotic strains and other types of cancer (111). A human study, that compared the composition of the faecal flora of people with different risks of colon cancer, shows highs risk of colon cancer with presence of Bacteroides vulgatus and Bacteroides stercosis and low risk with presence of Lactobacillus acidophilus, Lactobacillus S06 and Eubacterium aerofaciens (112).

Conclusion

In conclusion, age-related changes in the bowel increase susceptibility to infection and gut function disorders. Probiotics are dietary options that have the potential to improve beneficial effects. There is good evidence that strains like LGG and S. boulardii are effective in preventing AAD, but other strains and mixtures of strains seem to be effective as well. There is also good evidence of probiotic intake in C. difficile associated diarrhea, although some studies have been too small. The constant interaction between the host and its microbial guests can infer important to the human host (113). These benefits include: establishment of balanced intestinal microflora; improving colonization resistance and or prevention of diarrhea; reduction of fecal enzymes; reduction of serum cholesterol; reduction of potential mutagens; metabolism of lactose; improved calcium absorption; reduction of lactose intolerance; synthesis of vitamins; predigestion of proteins (4, 25, 114). Evidence is particularly strong for some probiotics for maintenance and improvement of bowel function (e.g. constipation and IBS) (99). Investigations in people with IBD showed probiotic strains to be effective in decreasing the recurrence of UC and occurrence and recurrence of pouchitis, whereas current evidence suggests that probiotics are ineffective in treating patients with CD. Patients with IBS showed an improvement in symptoms when consuming selected probiotic strains; however, high placebo effects have been reported as well. The duration of treatment with probiotics in studies included in this review varied greatly depending on the study outcome (115). General recommendations on how long probiotics should be taken are therefore difficult to make as based on treatment duration in intervention studies. However, further studies are needed to better evaluate the effects of a variety of probiotics supplements on immunity including CRP levels on nutritional status and on colon cancer prevention.

Key message

Probiotic influences

1. Metabolic function
2. Nutritional support
3. Vitamin synthesis
4. Competitive exclusion of pathogens
5. Regulation of both innate and adaptive immune
6. Stimulation of Proliferation
   Angiogenesis
   Epithelial restitution

References

1. Baker HC, Tran DN, Thomas LV. Health benefits of probiotics for the elderly: a review.fJournal of Foodservice 2009; 20: 250–262
2. Hopkins MJ, Macfarlane GT. Changes in predominant bacterial populations in human faeces with age and with Clostridium difficile infection. J Med Microbiol 2002; 51: 448–54
3. Hopkins MJ, Sharp R, Macfarlane GT. Variation in human intestinal microbiota with age. Dig Liver Dis 2002; 34: 12
4. Tannock GW. Probiotic properties of lactic acid bacteria plenty of scope for fundamental R & D. Trends in Biotechnol 1997; 15: 270–274
5. De Simone C, Chiodo F, Delia S, Pastore G, Scalise G, Sorice F, Tonietti G, Zanussi C, Paoloni M, Gargiulo M. Clinical results of a multicenter study with sulbactam/ampicillin for the treatment of patients with lower respiratory and urinary tract infections. J Chemother. 1991; 3: 321-7

6. Elmer GW, Surawicz CM, McFarland LV. Biotherapeutic agents. A neglected modality for the treatment and prevention of selected intestinal and vaginal infections. JAMA 1996; 275: 870-6
7. Orrhage K, Sillerström E, Gustafsson JA, Nord CE, Rafter J. Binding of mutagenic heterocyclic amines by intestinal and lactic acid bacteria. Mutat Res. 1994; 311: 239-48
8. Singh J, Rivenson A, Tomita M, Shimamura S, Ishibashi N, Reddy BS. Bifidobacterium longum, a lactic acid-producing intestinal bacterium inhibits colon cancer and modulates the intermediate biomarkers of colon carcinogenesis. Carcinogenesis.1997; 18: 833-41
9. Holzapfel WH, Schillinger U. Introduction to pre- and probiotics. Food Res Int 2002; 35: 109–116
10. Cani PD, Delzenne NM. The role of the gut microbiota in energy metabolism and metabolic disease. Curr Pharm Des 2009; 15: 1546-58
11. Joint FAO/WHO Working Group Report on Drafting Guidelines for the Evaluation of Probiotics in Food. London, Ontario, Canada, 30 April and 1 May 2002.
12. Kailasapathy K, Chin J. Survival and therapeutic potential of probiotic organisms with reference to Lactobacillus acidophilus and Bifidobacterium. Immunol Cell Biol 2000; 78: 80–88
13. Gill HS, Shu Q, Lin H, Rutherfurd KJ, Cross ML. Protection against translocating Salmonella typhimurium infection in mice by feeding the immuno-enhancing probiotic Lactobacillus rhamnosus strain HN001. Med Microbiol Immunol 2001; 190: 97–104
14. Mangell P, Nejdfors P, Wang M, Ahrne S, Westrom B, Thorlacius H, Jeppsson B. Lactobacillus plantarum 299v inhibits Escherichia coli-induced intestinal permeability. Dig Dis Sci 2002; 47: 511–516
15. Coakley M, Ross RP, Nordgren M, Fitzgerald G, Devery R, Stanton C. Conjugated linoleic acid biosynthesis by human-derived Bifidobacterium species. J Appl Microbiol 2003; 94: 138–145
16. Fonden R, Mogensen G, Tanaka R, Salminen S. Effect of fermented dairy products on intestinal microflora, human nutrition, and health: current knowledge and future perspectives. Bulletin of the International Dairy Federation. 2000; 352
17. Malaguarnera L, Ferlito L, Imbesi RM, Gulizia GS, Di Mauro S, Maugeri D, Malaguarnera M, Messina A. Immunosenescence: a review. Arch Gerontol Geriatr. 2001;32:1-14
18. Walker WA. Role of nutrients and bacterial colonization in the development of intestinal host defense. J Pediatr Gastroenterol Nutr 2000; 30: 2–7
19. Luyer MD, Buurman WA, Hadfoune M. Strain-specific effects of probiotics on gut barrier integrity following hemorrhagic shock. Infect Immun 2005; 73: 3689–92
20. Canani RB, Cirillo P, Terrin G. Probiotics for treatment of acute diarrhoea in children: a randomised clinical trial of five different preparations. BMJ 2007; 335, 340
21. Kekkonen RA, Lummela N, Karjalainen H. Probiotic intervention has strain-specific anti-inflammatory effects in healthy adults. World J Gastroenterol 2008; 14: 2029–36
22. Malaguarnera L, Cristaldi E, Malaguarnera M. The role of immunity in elderly cancer. Crit Rev Oncol Hematol. 2010; 74: 40-60
23. Malaguarnera L, Ferlito L, Di Mauro S, Imbesi RM, Scalia G, Malaguarnera M. Immunosenescence and cancer: a review. Arch Gerontol Geriatr. 2001;32:77-93
24. Candore G, Balistreri CR, Colonna-Romano G, Grimaldi MP, Lio D, Listi' F, Scola L, Vasto S, Caruso C. Immunosenescence and antiimmunosenescence therapies: the case of probiotics. Rejuvenation Res. 2008;11: 425–32
25. Turchet P, Laurenzan M, Auboiron S, Antoine JM. Effect of fermented milk containing the probiotic Lactobacillus casei DN-114001 on winter infections in free-living elderly subjects: a randomised, controlled pilot study. J Nutr Health Aging 2003; 7: 75–7
26. Fukushima Y. Improvement of nutritional status and incidence of infection in hospitalised, enterally fed elderly by feeding of fermented milk containing probiotic Lactobacillus johnsonii La1 (NCC533). Brit J Nutr. 2007; 98: 969–77
27. Szajewska H, Skórka A, Ruszczynski M. Metaanalysis: Lactobacillus GG for treating acute diarrhoea in children. Aliment Pharm Ther 2007; 25: 871–81
28. Black FT, Andersen PL, Orskov J, Orskov F, Gaarslev K, Laulund S. Prophylactic efficacy of lactobacilli on traveller's diarrhea. In: Steffen R (ed) Travel medicine. Conference on international travel medicine 1, Zurich, Switzerland. Springer, Berlin Heidelberg New York, 1989 333–335
29. Hilton E, Kolakowski P, Smith M, Singer C. Efficacy of Lactobacillus GG as a diarrhoea preventive. J Travel Med 1997; 4: 3–7
30. Oksanen PJ, Salminen S, Saxelin M, Hämäläinen P, Ihantola-Vormisto A, Muurasniemi-Isoviita, Nikkari S, Oksanen T, Pörsti I, Salminen E, Siitonen S, Stuckey H, Toppila A, Vapaatalo H. Prevention of travellers' diarrhoea by Lactobacillus GG. Ann Med 1990; 22: 53–56
31. Katelaris PH, Salam I, Farthing MJG. Lactobacilli to prevent traveler's diarrhea? New Engl J Med 1995 ; 333: 1360–1361
32. McFarland LV, Dublin S. Meta-analysis of probiotics for the treatment of irritable bowel syndrome. World J Gastroenterol 2008; 14: 2650–61
33. McFarland LV. Epidemiology risk factors and treatments for antibiotic-associated diarrhoea. Dig Dis 1998; 16: 292–307
34. Gougoulias C, Tuohy KM, Gibson GR. Dietary based gut flora modulation against Clostridium difficile onset. Food Science and Technology Bulletin Functional Foods 2007; 4: 31–41
35. Beausoleil M, Fortier N, Guénette S. Effect of a fermented milk combining Lactobacillus acidophilus Cl1285 and Lactobacillus casei in the prevention of antibiotic-associated diarrhea: a randomized, double-blind, placebo-controlled trial. Can J of Gastroenterol. 2007; 21: 732–6
36. Hickson M, D'Souza AL, Muthu N. Use of probiotic Lactobacillus preparation to prevent diarrhoea associated with antibiotics: randomised double blind placebo controlled trial. BMJ 2007; 335: 80
37. Wenus C, Goll R, Loken EB. Prevention of antibioticassocated diarrhoea by a fermented probiotic milk drink. Eur J Clin Nutr 2008; 62: 299–301
38. Weichselbaum E. Probiotics and health: a review of the evidence British Nutrition Foundation Nutrition Bulletin 2009; 34: 340–373
39. McFarland LV, Surawicz CM, Greenberg RN. A randomized placebo-controlled trial of Saccharomyces boulardii in combination with standard antibiotics for Clostridium difficile disease. JAMA 1994; 271: 1913–8
40. Plummer S, Weaver MA, Harris JC. Clostridium difficile pilot study: effects of probiotic supplementation on the incidence of C. difficile diarrhoea. Int Microbiol 2004; 7: 59–62
41. Cremonini F, Talley NJ. Irritable bowel syndrome: epidemiology, natural history, health care seeking and emerging risk factors. Gastroenterol Clin North Am. 2005; 34: 189–204
42. NICE (National Institute for Health and Clinical Excellence). Irritable bowel syndrome in adults. Diagnosis and management of irritable bowel syndrome in primary care. NICE clinical guideline 61: Developed by the National Collaborating Centre for Nursing and Supportive Care, London, 2008.
43. Seksik P, Sokol H, Lepage P. Review article: the role of bacteria in onset and perpetuation of inflammatory bowel disease. Aliment Pharmacol Ther 2006; 24: 11–18
44. Pimentel M, Chow EJ, Lin HC. Eradication of small intestinal bacterial overgrowth reduces symptoms of irritable bowel syndrome. Am J Gastroenterol 2000; 95: 3503–6
45. Lin HC. Small intestinal bacterial overgrowth: a framework for understanding irritable bowel syndrome. JAMA 2004; 292: 852–8
46. Drouault-Holowacz S, Bieuvelet S, Burckel AA. double blind randomized controlled trial of a probiotic combination in 100 patients with irritable bowel syndrome. Gastroenterol Clin Biol 2008; 32: 147–52
47. Sinn DH, Song JH, Kim HJ. Therapeutic effect of Lactobacillus acidophilus-SDC 2012, 2013 in patients with irritable bowel syndrome. Dig Dis Sci 2008; 53: 2714–18
48. Kajander K, Myllyluoma E, Rajilic-Stojanovic M. Clinical trial: multispecies probiotic supplementation alleviates the symptoms of irritable bowel syndrome and stabilizes intestinal microbiota. Aliment Pharmacol Ther 2008; 27: 48–57
49. Longstreth GF, Thompson WG, Chey WD. Functional bowel disorders. Gastroenterology 2006; 130: 1480–91
50. Chmielewska A, Szajewska H. Systematic review of randomised controlled trials: Probiotics for functional constipation. World J Gastroenterol 2010; 16: 69-75
51. Marteau P. Bifidobacterium animalis strain DN-173 010 shortens the colonic transit time in healthy women: a double-blind, randomized, controlled study. Aliment Pharmacol Ther 2002; 16: 587–93
52. Matsumoto K. The effect of a probiotic milk product containing Lactobacillus casei strain Shirota on the defecation frequency and the intestinal microflora of sub-optimal health state volunteers: a randomized placebo-controlled cross-over study. Bioscience and Microflora 2006; 25: 39–48
53. Ouwehand AC, Tiihonen K, Saarinen M, Putaal H, Rautonen N. Influence of a combination of Lactobacillus acidophilus NCFM and lactitol on healthy elderly: intestinal and immune parameters. Brit J Nutr 2009; 3: 367–75
54. Banaszkiewicz A, Szajewska H. Probiotics in the treatment of constipation: a systematic review of randomized controlled trials. Pediatria Wspolczesna 2005; 7: 9–14
55. Khedkar CD, Ouwehand AC. Modifying the gastrointestinal microbiota with probiotics. In: Gastrointestinal Microbiology (eds Ac Ouwehand & EE Vaughan), 2006 pp 315–333.Taylor & Francies Group, New York, NY
56. Hughes R, Rowland IR. Metabolic activities of the gut microflora in relation to cancer. Microb Ecol Health D 2000; 11: 179–85
57. Spanhaak S, Havenaar R, Schaafsma G. The effect of consumption of milk fermented by Lactobacillus casei strain Shirota on the intestinal microflora and immune parameters in humans. Eur J Clin Nutr 1998; 52: 899–907
58. De Preter V. Effect of dietary intervention with different pre- and probiotics on intestinal bacterial enzyme activities. Eur J Clin Nutr 2008; 62: 225–31
59. Goldin BR, Gorbach SL. The relationship between diet and rat faecal bacterial enzymes implicated in colon cancer. J Natl Cancer I 1976; 57: 371–5
60. Tomoda T, Nakano Y, Kageyama T. Effect of yogurt and yogurt supplemented with Bifidobacterium and/or lactulose in healthy persons: a comparative study. Bifidobacteria Microflora 1991; 10: 123–30
61. Brigidi P, Vitali B, Swennen E, Bazzozzhi G, Matteuzzi D. Effects of probiotic

administration upon the composition and enzymatic activity of human faecal microbiota for patients with irritable bowel syndrome or functional diarrhea. Res Microbiol 2001; 152: 735–41
62. Ouwehand AC, Lagstrom H, Suomalainen T, Salminen S. Effect of probiotics on constipation, fecal azoreductase activity and fecal mucin content in the elderly. Ann Nutr Metabol 2002; 46: 159–62
63. Malaguarnera M, Gargante MP, Malaguarnera G, Salmeri M, Mastrojeni S, Rampello L, Pennisi G, Li Volti G, Galvano F. Bifidobacterium combined with fructo-oligosaccharide versus lactulose in the treatment of patients with hepatic encephalopathy. Eur J Gastroenterol Hepatol.2010; 22: 199-206
64. Sheth AA, Longo W, Floch MH. Diverticular disease and diverticulitis. Am J Gastroenterol 2008; 103: 1550-1556
65. Sandler RS, Everhart JE, Donowitz M. The burden of selected digestive diseases in the United States. Gastroenterology 2002; 122: 1500–11
66. Petruzziello L, Iacopini F, Bulajic M, Shan S, Costamagna G. Review article: uncomplicated diverticular disease of the colon. Aliment Pharmacol Ther 2006; 23: 1379–1391
67. Stollman N, Raskin JB Diverticular disease of the colon. Lancet 2004; 363: 631–9
68. Shanahan F. Turbo probiotics for IBD. Gastroenterology 2001; 120: 1297–8
69. Giaccari S, Tronci S, Falconieri M, Ferrieri A. Long-term treatment with rifaximin and lactobacilli in post-diverticulitic stenoses of the colon. Riv Eur Sci Med Farmacol 1993; 15: 29–34
70. Tursi A, Brandimarte G, Giorgetti GM, Elisei W. Mesalazine and/or Lactobacillus casei in Preventing Recurrence of Symptomatic Uncomplicated Diverticular Disease of the Colon A Prospective, Randomized, Open-label Study. J Clin Gastroenterol 2006; 40: 312–316
71. Fricu P, Zavoral M. The effect of non-pathogenic Escherichia coli in symptomatic uncomplicated diverticular disease of the colon. Eur J Gastroenterol Hepatol 2003; 15: 313–5
72. Malaguarnera M, Greco F, Barone G, Gargante MP, Malaguarnera M, Toscano MA. Bifidobacterium longum with fructo-oligosaccharide (FOS) treatment in minimal hepatic encephalopathy: a randomized, double-blind, placebo-controlled study. Dig Dis Sci. 2007; 52: 3259-65
73. Kleessen B, Hartmann L, Blaut M. Oligofructose and long-chain inulin: influence on the gut microbial ecology of rats associated with a human faecal flora. Br J Nutr 2001; 86: 291–300.
74. Lamika P, Tsuchiya J, Pathak S, Okura R, Solimene U, Jain S, Kawakita S, Marotta F. Probiotics in Diverticular Disease of the Colon: an Open Label Study. J Gastrointestin Liver Dis 2010; 19: 31-36
75. Shanahan F. The host-microbe interface within the gut. Best Pract Res Clin Gastroenterol 2002; 16: 915– 931
76. García-Lafuente A, Antolín M, Guarner F, et al. Incrimination of anaerobic bacteria in the induction of experimental colitis. Am J Physiol 1997; 272: 10–15
77. Maki M, Mustalahti K, Korhonen J, Kulmala P, Haapalahti M, Karttunen T. Prevalence of celiac disease among children in Finland. New Engl J Med. 2003; 348: 2517–2524
78. Stepniak DL, Spaenij-Dekking C, Mitea M, Moester A. de Ru, Baak-Pablo R, van Veelen P, Edens L, Koning F. Highly efficient gluten degradation with a newly identified prolyl endoprotease: implicationsfor celiac disease. Am J Physiol Gastrointest Liver Physiol 2006; 291: 621–629
79. Shan L, Qiao SW, Arentz-Hansen H, Molberg O, Gray GM, Sollid LM, Khosla C. Identification and analysis of multivalent proteolytically resistant peptides from gluten: implications for celiac sprue. J. Proteome Res 2005; 4: 1732–1741
80. Shan L, Molberg O, Parrot I, Hausch F, Filiz F, Gray GM, Sollid LM, Khosla C. Structural basis for gluten intolerance in celiac sprue. Science 2002; 297: 2275–2279
81. Chand N, Mihas AA. Celiac disease: current concepts in diagnosis and treatment. J. Clin. Gastroenterol 2006; 40: 3–14
82. Gobbetti M, Rizzello CG, Di Cagno R, De Angelis M Sourdough lactobacilli and celiac disease. Food Microbiol 2007; 24: 187–196
83. D'Arienzo R, Maurano F, Lavermicocca P, Ricca E, Rossi M. Cytokine 2009; 48: 254–259
84. D'Arienzo R, Maurano F, Luongo D, Mazzarella G, Stefanile R, Troncone R Adjuvant effect of Lactobacillus casei in a mouse model of gluten sensitivity. Immunol Lett 2008; 119: 78–83
85. Mohamadzadeh M, Olson S, Kalina WV, Ruthel G, Demmin GL, Warfield KL. Lactobacilli activate human dendritic cells that skew T cells toward T helper 1 polarization. Proc Natl Acad Sci USA 2005; 102:2880–5
86. Pochard P, Gosset P, Grangette C, Andre C, Tonnel AB, Pestel J. Lactic acid bacteria inhibit TH2 cytokine production by mononuclear cells from allergic patients. J Allergy Clin Immunol 2002; 110: 617–23
87. Heufler C, Koch F, Stanzl U, Topar G, Wysocka M, Trinchieri G. Interleukin-12 is produced by dendritic cells and mediates T helper 1 development as well as interferon-c production by T helper 1 cells. Eur J Immunol 1996; 26: 659–68
88. Asseman C, Mauze S, Leach MW, Coffman RL, Powrie F An essential role for interleukin 10 in the function of regulatory T cells that inhibit intestinal inflammation. J Exp Med 1999; 190: 995–1004
89. Pool-Zobel BL, Neudecker C, Domizla I, Ji S, Schillinger U, Rumney C. Lactobacillus - and bifidobacterium -mediated antigenotoxicity in the colon of rats. Nutr Cancer 1996; 26: 365–80
90. Rangavajhyala N, Shahani KM, Sridevi G, Srikumaran S. Non lipopolysaccharide component(s) of Lactobacillus acidophilus stimulate(s) the production of interleukin-1a and tumor necrosis factor-a by murine macrophages. Nutr Cancer 1997; 28: 130–4
91. Marteau P, Lémann M, Seksik P. Ineffectiveness of Lactobacillus johnsonii LA1 for prophylaxis of postoperative recurrence in Crohn's disease: a randomised, double blind, placebo controlled GETAID trial. Gut 2006; 55: 842–7
92. Van Gossum A, Dewit O, Louis E. Multicenter randomized-controlled clinical trial of probiotics (Lactobacillus johnsonii, LA1) on early endoscopic recurrence of Crohn's disease after ileo-caecal resection. Inflamm Bowel Dis 2007; 13: 135–42
93. Prantera C, Scrobano ML, Falasco G. Ineffectiveness of probiotics in preventing recurrence after curative resection for Crohn's disease: a randomised controlled trial with Lactobacillus GG. Gut 2002; 51: 405-9
94. Guslandi M, Mezzi G, Sorghi M. Saccharomyces boulardii in maintenance treatment of Crohn's disease. Dig Dis Sci 2000; 45: 1462–4
95. Carter MJ, Lobo AJ, Travis SPL. Guidelines for the management of inflammatory bowel disease in adults; on behalf of the IBD Section of the British Society of Gastroenterology. Gut 2004; 53 (Suppl. V): v1–6
96. Borruel N, Casellas F, Antolín M, Llopis M, Carol M, Espíin E, Naval J, Guarner F, Malagelada JR. Effects of nonpathogenic bacteria on cytokine secretion by human intestinal mucosa. Am J Gastroenterol. 2003; 98: 865–870
97. Rembacken BJ, Snelling AM, Hawkey PM. Nonpathogenic Escherichia coli versus mesalazine for the treatment of ulcerative colitis: a randomized trial. Lancet 1999; 35: 635–9
98. Kruis W, Fric P, Pokrotnieks J. Maintaining remission of ulcerative colitis with the probiotic Escherichia coli Nissle 1917 is as effective as with standard mesalazine. Gut 2004; 53: 1617–23
99. Kato K, Mizuno S, Umesaki Y. Randomized placebo controlled trial assessing the effect of bifidobacteria-fermented milk on active ulcerative colitis. Aliment Pharmacol Ther 2004; 20: 1133–41
100. Ishikawa H, Akedo I, Umesaki Y. Tanaka R, Imaoka A, Otani T. Randomized controlled trial of the effect of bifidobacteria-fermented milk on ulcerative colitis. J Am Coll Nutr 2002; 22: 56–63
101. Pardi DS, Sandborn WJ. Systematic review: the management of pouchitis. Aliment Pharmacol Ther 23:1087–96
102. Yu ED, Shao Z, Shen B. Pouchitis. World J Gastroenterol 2006; 13: 5598–604
103. Gionchetti P, Rizzello F, Venturi A. Oral bacteriotherapy as maintenance treatment in patients with chronic pouchitis: a double-blind, placebo-controlled trial. Gastroenterology 2000; 119: 305–9
104. Mimura T, Rizzello F, Helwig U. Once daily high dose probiotic therapy (VSL#3) for maintaining remission in recurrent of refractory pouchitis. Gut 2004; 53: 108–14
105. Gionchetti P, Rizzello F, Helwig U. Prophylaxis of pouchitis onset with probiotic therapy: a double-blind, placebocontrolled trial. Gastroenterology 2003; 124: 1202–9
106. Kuisma J, Mentula S, Jarvinen H. Effect of Lactobacillus rhamnosus GG on ileal pouch inflammation and microbial flora. Aliment Pharmacol Ther 2003; 17: 509–15
107. Calla A, Rao DR, Chawan CB, Shackelford I. Bifidobacterium longum and lactulose suppress azoxymethane-induced colonic aberrant crypt foci in rats. Carcinogenesis 1997; 18: 517–521
108. Young GP. Prevention of colon cancer: Role of short chain fatty acids produced by intestinal flora. Asia Pacific J. Clin. Nutr 1996; 5: 44–7
109. Topping DL. Short chain fatty acids produced by intestinal bacteria. Asia Pacific J. Clin. Nutr 1996; 5: 15–19
110. Ohashi Y, Nakai S, Tsukamoto T, Masumori N, Akaza H, Miyanaga N. Habitual intake of lactic acid bacteria and risk reduction of bladder cancer. Urol Int. 2002; 68: 273–280
111. Rafter JJ. The role of lactic acid bacteria in colon cancer prevention. Scand J Gastroenterol 1995; 30: 497–502
112. Moore WE. Holdeman LV. Human fecal flora: the normal flora of 20 Japanese-Hawaiians. Appl Microbial 1995; 27: 961-79
113. Salminen S, Bouley C, Boutron-Ruault MC, Cummings JH, Franck A, Gibson GR, Isolauri E, Moreau MC, Roberfroid M, Rowland I. Functional food science and gastrointestinal physiology and function. Brit J Nutr 1998; 80: 147–171
114. Watzl B. Natural killer cells, probiotics and cancer. International Journal of Probiotics and Prebiotics 2008; 3: 141–
115. Zaharoni H, Rimon E, Vardi H, Friger M, Bolotin A, Shahar DR. Probiotics improve bowel movements in hospitalized elderly patients- the Proage Study. The Journal of Nutrition, Health & Aging 2011; 15: 215-220