

ROLE OF PROBIOTICS IN THE TREATMENT OF IRRITABLE BOWEL SYNDROME: POTENTIAL MECHANISMS AND CURRENT CLINICAL EVIDENCE

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ABSTRACT *Irritable bowel syndrome (IBS) is a highly prevalent functional gastrointestinal disorder affecting 3 to 15 % of the general population. It is characterized by unexplained abdominal pain, discomfort, and bloating in association with altered bowel habits. The pathophysiology of IBS is not well understood, but most likely involves multiple causes. IBS has been associated with abnormal gastrointestinal motor function, visceral hypersensitivity, psychosocial factors, autonomic dysfunction, and inflammation. In a subgroup of patients, IBS develops after an acute bacterial infection of the bowel. Increased numbers of inflammatory cells, such as mast cells and lymphocytes in the colonic mucosa of IBS patients suggest an ongoing state of inflammation in these patients. Probiotics have been shown to have a beneficial effect in acute infectious diarrhea and inflammatory bowel disease and thus could presumably be of potential benefit in postinfectious IBS. Another rationale for using probiotics in IBS is their potential to influence fermentation processes and diminish gas production by changing the colonic flora. Even though evidence from controlled clinical trials supporting a beneficial role of probiotics in the treatment of IBS is still limited, improvement of different IBS symptoms and normalization of inflammatory cytokine levels have been demonstrated. Small sample sizes and the use of different probiotic preparations complicate the interpretation and comparison between different studies. While preliminary results are encouraging, the exact mechanism of action and their clinical efficacy of Probiotics in IBS need to be studied in well designed experiments and larger randomized, controlled trials.*

KEYWORDS: IBS, Immunology, Inflammation, Irritable bowel syndrome, Microbiology, Probiotics, Probiotic bacteria, *Saccharomyces boulardii*

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INTRODUCTION

Irritable bowel syndrome (IBS) is one of the most common functional gastrointestinal disorders with a prevalence of 3 to 15% in the general population (Cremonini and Talley 2005; Drossman, Li et al. 1993). IBS is characterized by abdominal pain and discomfort in association with altered bowel habits, and the symptoms cannot be explained by any structural abnormalities using current standard diagnostic tests (Drossman, Camilleri et al. 2002). In addition to abdominal pain, diarrhea or constipation, typical symptoms include bloating, flatulence, stool urgency or straining and the feeling of incomplete evacuation (Drossman, Camilleri et al. 2002). Characteristic symptom patterns according to the IBS consensus “Rome II” criteria (Table 1) and absence of structural disease allow a positive diagnosis of IBS (Thompson, Longstreth et al. 1999). The pathophysiology of IBS is still not well understood. Several factors such as motor and sensory dysfunction, immune mechanisms and psychological factors are suggested to play a role (Camilleri 2005; Drossman, Camilleri et al. 2002). Currently available IBS therapies are mainly symptom-orientated (e.g. anti-diarrheals for diarrhea, laxatives for constipation or smooth muscle relaxants for pain) and often are of limited efficacy regarding the overall complaints. Other treatment strategies, such as antidepressants for pain relief, are inadequate for long term management due to their side effect profiles. Newer medications targeting the gut nervous system, such as different serotonergic receptor modulators, are efficacious, but their availability is very limited due to restricted drug approval in selected countries, and some agents are suspected to have an unfavorable safety profile.

Probiotics are preparations that contain viable commensal microorganisms, which have potential beneficial effects in the prevention or in the treatment of various gastrointestinal and other disorders (Sartor 2004). They are considered safe and are usually well tolerated.

This review will discuss the potential role of probiotics in the management of IBS focusing on how their currently known benefits influence the pathophysiology of IBS. The evidence from randomized, placebo-controlled trials is reviewed and new frontiers for future research are identified.

Table 1: Rome II Consensus Diagnostic Criteria for Irritable Bowel Syndrome (IBS) in the absence of structural or metabolic abnormalities to explain the symptoms (Thompson, Longstreth et al. 1999)

<p>At least 12 weeks or more, which need not to be consecutive, in the preceding 12 months of abdominal discomfort or pain that has two out of three features:</p> <p>(I) Relieved with defecation; and/or</p> <p>(II) Onset associated with a change in frequency of stool; and/or</p> <p>(III) Onset associated with a change in form (appearance) of stool</p> <p><u>Symptoms that cumulatively support the diagnosis of IBS</u></p> <ul style="list-style-type: none"> • Abnormal stool frequency (for research purposes “abnormal” may be defined as greater than 3 bowel movements per day and less than 3 bowel movements per week). • Abnormal stool form (lumpy/hard or loose/watery stool). • Abnormal stool passage (straining, urgency, or feeling of incomplete evacuation). • Passage of mucus. • Bloating or feeling of abdominal distension
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The potential influence of probiotics on different IBS mechanisms

Immune mechanisms

There is a growing body of clinical and laboratory evidence that IBS has an inflammatory component. Not only do inflammatory bowel disease patients, particularly those with Crohn's disease experience IBS type symptoms following acute flare-ups. These symptoms usually occur in the absence of endoscopically detectable mucosal inflammation. IBS also affects 10-15% of patients after acute infectious enteritis (Spiller 2003). Mucosal biopsies in patients with postinfectious (PI)-IBS indicate a persistent, mild inflammatory state, defined by increased numbers of inflammatory cells and serotonin-releasing entero-endocrine cells in the mucosa (Spiller 2004; Spiller, Jenkins et al. 2000). Since serotonin is one of the most important neurotransmitters of the enteric nervous system, mucosal changes in serotonin levels would affect both sensory and motor functions possibly contributing to IBS symptoms (Borman 2001). Another mucosal immune system-alteration found in IBS patients is an increased number of activated mast-cells in the proximity of colonic nerves in the lamina propria, where mast cell secreted mediators such as tryptase and histamine may contribute to the development of abdominal pain (Barbara, Stanghellini et al. 2004). Further evidence for a pro-inflammatory state in IBS patients comes from a recent study reporting decreased IL10/IL12 ratios in IBS patients (O'Mahony, McCarthy et al. 2005). To date, several studies have demonstrated protecting and anti-inflammatory effects of probiotics in infectious diarrhea (Huang, Bousvaros et al. 2002; Isolauri, Kirjavainen et al. 2002;

Resta-Lenert and Barrett 2003), antibiotic-associated diarrhea (Cremonini, Di Caro et al. 2002) as well as in animal models and human inflammatory bowel diseases (Baumgart and Dignass 2004; Kruijs, Fric et al. 2004; Lammers, Vergopoulos et al. 2005; Madsen, Doyle et al. 1999; Sartor 2004; Schultz, Strauch et al. 2004). Suggested mechanisms include restoration of the microbial milieu by promoting colonization with strains of the commensal flora following infection and shifting the host's immune response towards an anti-inflammatory state (Cremonini, Di Caro et al. 2002) (Sartor 2005). Probiotics may also serve as vehicles to deliver anti-inflammatory cytokines such as a IL-10. The oral administration of IL-10 secreting *Lactococcus lactis* which was originally developed and tested in two murine models of inflammatory bowel disease is currently being evaluated for treatment human inflammatory bowel disease (Baat, Rottiers et al. 2005; Baat, Steidler et al. 2005; Steidler, Hans et al. 2000).

Probiotics may therefore also have beneficial effects on altered colonic immune-states that may be found in IBS patients. In fact, promising evidence is provided by the recent study of O'Mahony et al., in which treatment of IBS with *Bifidobacterium infantis* normalized pro-inflammatory IL10/IL12-ratios towards an anti-inflammatory state, which was associated with significant improvement in IBS symptoms (O'Mahony, McCarthy et al. 2005).

Bacterial milieu and fermentation

The indigenous colonic microbial bacterial flora plays an important physiologic role in the gut. First, the resident flora contributes to enhance the intestinal barrier function, i.e. preventing the adhesion of pathologic bacteria and inhibiting

the invasion of pathogenic agents into the body (Baumgart and Dignass 2002; van der Waaij 1989; van der Waaij 1991). There is evidence that postinfectious IBS patients may experience increased gut permeability (Spiller, Jenkins et al. 2000). While some studies indicated an improvement of barrier functions by probiotics (Lamine, Eutamene et al. 2004; Rosenfeldt, Benfeldt et al. 2004), other studies did not find this beneficial effect (McNaught, Woodcock et al. 2005). However, numerous studies confirmed that probiotics are an effective treatment to re-establish a balanced commensal flora after intestinal infection or antibiotic treatment (Cremonini, Di Caro et al. 2002; Huang, Bousvaros et al. 2002; McFarland, Surawicz et al. 1994; Sartor 2005).

Another important function of colonic bacteria is the metabolism of nutrient substrates that reach the colon. This leads to the formation of gas and the production of short chain fatty acids, SCFA which are an important energy source and trophic regulator of colonic enterocytes (Guarner and Malagelada 2003) and may also induce propulsive contractions (Kamath, Phillips et al. 1990) and accelerate transit or enhance fluid and sodium absorption. Alteration of the colonic flora with administration of probiotics may modify fermentation processes and hence gas production, colonic transit and fluid fluxes. In fact, abdominal bloating, distension or flatulence, which are dominant symptoms in many IBS patients, have been shown to improve significantly by probiotic treatment in several placebo controlled trials (Bausserman and Michail 2005; Kim, Camilleri et al. 2003; Kim, Vazquez Roque et al. 2005; Nobaek, Johansson et al. 2000; O'Mahony, McCarthy et al. 2005).

Motility

Altered bowel habits such as diarrhea or constipation are main characteristics of IBS, and altered gut motility is considered to be an important underlying factor. Recently it was demonstrated that *Lactobacillus casei* significantly decreased muscle dysfunction in a mouse model of PI-IBS (Verdu, Bercik et al. 2004a). This effect was also observed, when live bacteria were administered long after the initial infection (Verdu, Bercik et al. 2004a) or when supernatants of the bacteria culture medium were used (Verdu, Bercik et al. 2004b).

Human studies also indicate a potential influence of probiotics on gut motility: The probiotic cocktail VSL#3 was shown to reduce colonic reflex motor responses to balloon distension (Bazzocchi, Gionchetti et al. 2002). Moreover, in IBS patients with predominantly bloating, VSL#3 significantly slowed down colonic transit compared to placebo without changing bowel habits (Kim, Vazquez Roque et al. 2005). However, this retardation effect on colonic transit was not observed in IBS patients with predominant diarrhea (Kim, Camilleri et al. 2003). Taken together, there is evidence that some probiotics may influence gastrointestinal motility and may improve post-infectious intestinal muscle dysfunction.

Sensory functions

Visceral hypersensitivity has been associated with IBS and is suggested to play a pathogenic role in the symptom generation of abdominal pain and discomfort in IBS (Delgado-Aros and Camilleri 2005; Delvaux 2004). To date, there is limited evidence suggesting potential effects of probiotics on sensory neurotransmission. The probiotic yeast *Saccharomyces boulardii* has been shown to modulate the expression of neuronal markers in the submucous plexus (Kamm, Hoppe et al. 2004). Verdu et al. reported that *Lactobacillus paracasei* NCC2461 normalized sensitivity to colorectal distension and sensory neurotransmitter expression in a mouse model of antibiotic induced visceral hypersensitivity (Verdu, Bercik et al. 2005). Beneficial probiotic effects on sensory mechanisms are furthermore suggested by three clinical trials that demonstrated an improvement of abdominal pain in IBS patients after treatment with different *Lactobacilli* or *Bifidobacteria* (Niedzielin, Kordecki et al. 2001; O'Mahony, McCarthy et al. 2005; Saggioro 2004) (see below). Taken together, there is evidence that probiotics may modulate disturbed visceral perception. Further research is indicated to explore the specific effects of probiotics on gastrointestinal sensory mechanisms.

Bile acid-induced diarrhea

In some IBS patients, diarrhea has been associated with malabsorption of bile acids (Luman, Williams et al. 1995). Bile acid-induced diarrhea is a well known phenomenon in patients who had smaller ileal resections. It is even more pronounced after ileocolonic resections. These surgical procedures compromise bile acid absorption and can result in bile acid-induced secretory diarrhea because less malabsorbed water is present to dilute bile acids below the cathartic threshold. Bile acids have also been demonstrated to induce propulsive contractions in the colon (Bampton, Dinning et al. 2002; Chadwick, Gaginella et al. 1979). Some probiotics, e.g. *Bifidobacteria subspecies* and *Lactobacilli*, have the ability to deconjugate and absorb bile acids. This may reduce the amount of intracolonic bile acids and could therefore have a beneficial effect on diarrhea by decreasing colonic fluid secretion or motility.

Current evidence from randomized placebo-controlled clinical studies

Numerous anecdotal reports and uncontrolled trials suggest beneficial effects of probiotics on IBS symptoms. However, to evaluate the evidence for the therapeutic role of probiotics in IBS, we chose to focus only on data from randomized placebo controlled clinical trials. From 1989 to date, 11 randomized placebo controlled clinical trials were published, 4 of which within 2005. This noticeable increase in clinical trials demonstrates the rising interest in this field. Table 2 summarizes these 11 trials, which evaluated the effects of various probiotics on IBS in general or the relief of specific IBS symptoms.

Table 2: Overview on placebo-controlled clinical trials evaluating the efficacy of probiotics in the treatment of IBS from 1989 to 2005

Study	Probiotic	Patients [n]	Study design (all placebo controlled)	Significant symptomatic improvement compared to placebo
Gade and Thorn (Gade and Thorn 1989)	<i>Streptococcus faecium</i>	Functional lower bowel symptoms [54]	-4 weeks-parallel group	Physicians' overall assessment
Nobaek et al. (Nobaek, Johansson et. al 2000)	<i>Lactobacillus plantarum</i> (DSM 9843)	IBS Rome I[60]	-4 weeks-parallel group-12 month follow up	-Flatulence -Overall GI function after 12 months
O'Sullivan and O'Morain (O'Sullivan and O'Morain 2000)	<i>Lactobacillus casei</i> (GG)	IBS Rome I[24]	(2x) 8 weeks- -crossover	None
Niedzielin et al. (Niedzielin, Kordecki et al. 2001)	<i>Lactobacillus plantarum</i> (LP299V)	IBS Manning criteria [40]	4 weeks parallel group	- Pain - Higher responder rates regarding all symptoms
Sen et al. (Sen, Mullan et al. 2002)	<i>Lactobacillus plantarum</i> (LP299V)	IBS Rome I [12]	-(2x) 4 weeks -crossover	None
Kim et al. (Kim, Camilleri et al. 2003)	VSL#3*	IBS Rome II, diarrhea predominant [25]	8 weeks parallel group	Abdominal bloating
Saggioro et al. (Saggioro 2004)	<i>Lactobacillus plantarum</i> (LP01) + <i>Lactobacillus acidophilus</i> (LA02) <i>or</i> + <i>Bifidobacterium breve</i> (BB03)	IBS Rome II [70]	4 weeks parallel group	-Pain -Overall symptom score
O'Mahony et al. (O'Mahony, McCarthy et al. 2005)	<i>Lactobacillus salivarius</i> UCC4331 <i>or</i> <i>Bifidobacterium infantis</i> 35624	IBS Rome II [75]	8 weeks parallel group 4 weeks washout	Composite and individual symptom scores for: -pain/discomfort -bloating/distension -bowel movement difficulty
Niv et al. (Niv, Naftali et al. 2005)	<i>Lactobacillus reuteri</i> (ATCC 55730)	IBS Rome II[54]	6 months parallel group	None
Bausserman et Michail (Bausserman and Michail 2005)	<i>Lactobacillus GG</i>	Children with IBS Rome II[50]	6 weeks parallel group	Perceived abdominal distension
Kim, Vazquez et al. (Kim, Vazquez Roque et al. 2005)	VSL#3*	IBS Rome II, With bloating [48]	4 or 8 weeks parallel group	Flatulence

* VSL#3 = Probiotic cocktail containing *Bifidobacteria* (*B. longum*, *B. infantis*, *B. breve*), *lactobacilli* (*L. acidophilus*, *L. casei*, *L. bulgaricus*, *L. plantarum*), and *Streptococcus salivarius* subspecies *thermophilus*.

The earliest controlled study by Gade and Thorn (Gade and Thorn 1989) evaluated the efficacy of *Streptococcus faecium* in patients who presented with functional bowel symptoms for at least 6 months. After four weeks of treatment, 81% of the probiotic-treated compared to 41% of the placebo-treated group had symptom improvement according to the physicians' overall assessment.

Several studies tested *Lactobacillus plantarum* with conflicting results. In the study by Niedzielin et al. (Niedzielin, Kordecki et al. 2001), *Lactobacillus plantarum* significantly improved pain and had a responder rate for overall IBS symptom improvement of 95% compared with only 15% in the placebo group. Nobaek et al. (Nobaek, Johansson et al. 2000) observed a significant improvement of flatulence in *Lactobacillus plantarum*-treated IBS patients, while pain was improved in both the probiotic and the placebo-treated group. Moreover, this study incorporated a 12 months follow-up questionnaire in which the probiotic treated patients maintained better overall GI functions than the control patients. This might indicate a possible long-term beneficial alteration in colonic flora. Saggioro (Saggioro 2004), who administered a combination of *Lactobacillus plantarum* with either *Lactobacillus acidophilus* or *Bifidobacterium breve*, also observed a significant improvement of pain and overall IBS symptom score by both probiotic combinations compared to placebo. In contrast, in the crossover-designed study by Sen et al. (Sen, Mullan et al. 2002), *Lactobacillus plantarum* did not show any beneficial effect on colonic fermentation or IBS symptoms.

Similarly, two studies using *Lactobacillus casei* failed to provide convincing evidence for an efficacy of this probiotic in IBS: O'Sullivan and O'Morain (O'Sullivan and O'Morain 2000) observed no significant symptom improvement by *Lactobacillus casei*. However, they reported a trend towards improving stool frequency and consistency in a small subgroup of IBS patients with diarrhea. In children with IBS, *Lactobacillus casei* was not superior to placebo in the treatment of pain and bowel symptoms. This study by Bausserman and Michail (Bausserman and Michail 2005) reported a beneficial effect on perceived abdominal distension, but the observed difference in the treatment groups was solely due to a worsening of this symptom in the placebo group.

The Camilleri group conducted two trials with VSL#3, a probiotic cocktail containing 8 strains of different *Lactobacilli* (*L. acidophilus*, *L. casei*, *L. bulgaricus*, *L. plantarum*), *Bifidobacteria* (*B. longum*, *B. infantis*, *B. breve*) and *Streptococcus salivarius* subspecies *termophilus*. In addition to effects on IBS symptoms, these studies also evaluated the influence of VSL#3 on gastrointestinal transit. The first study (Kim, Camilleri et al. 2003) focused on diarrhea predominant IBS patients and observed a borderline significant improvement of abdominal bloating, but no effects on other IBS symptoms or gastrointestinal transit. Based on these results, the second study (Kim, Vazquez Roque et al. 2005) focused on IBS patients with bloating. This study detected a significant improvement of flatulence in the VSL#3-group, but differences towards greater improvement in bloating, pain

and urgency scores in the VSL#3 group did not reach statistical significance. Moreover, in this study, VSL#3 treatment was associated with a retardation of colonic transit without altering bowel functions. Differences in IBS populations (diarrhea vs. bloating) might partly account for different results between the two studies.

O'Mahony et al. (O'Mahony, McCarthy et al. 2005) compared the effects of two different probiotics, *Lactobacillus salivarius* and *Bifidobacterium infantis*, on IBS symptoms with weekly assessments over 8 weeks of treatment and 4 weeks of washout. Moreover, this study included the assessment of quality of life and of cytokine profiles, i.e. the ratio between IL-10 and IL-12 in peripheral blood mononuclear cells (PBMC), at baseline and after treatment. *Bifidobacterium infantis* was superior to placebo in relieving all individual and composite symptom scores except for stool frequency and stool consistency. Beneficial effects on the weekly symptom scores were significant in all 8 treatment-weeks and in 2 out of 4 washout-weeks. In contrast, *Lactobacillus salivarius* showed a significant symptom improvement over placebo only in the second treatment-week, and was significantly less effective than *Bifidobacterium infantis* in 4 out of 8 treatment weeks and in 3 out of 4 washout weeks. Despite the significant symptom improvement by *Bifidobacterium infantis*, overall quality of life scores were not significantly different between the three treatment groups. However, the most important finding of this study is the observation of a decreased baseline IL-10/IL-12 ratio in IBS patients (compared to reference values of healthy controls) that was normalized after treatment with *Bifidobacterium infantis*. The reduced baseline IL-10/IL-12 ratio reflects a pro-inflammatory state providing further evidence for an immune-mediated pathophysiology of IBS, at least in a subset of IBS patients. Moreover, the probiotic-induced normalization of the IL-10/IL-12-ratio towards an anti-inflammatory state correlates with the beneficial effect of the probiotic on IBS symptoms, which suggests an involvement of immunomodulatory mechanisms in IBS symptom generation.

While most studies have focused on the short-term effect of probiotics, Niv et al. (Niv, Naftali et al. 2005) conducted a six months treatment trial with *Lactobacillus reuteri* ATCC 55730 in patients with IBS. Over the 6 months, the entire study population improved in all symptoms, and there were no significant differences between the probiotic and the placebo group. However, a trend towards better improvement in constipation and gas was observed in the probiotic-treated patients.

This and other probiotic trials in IBS might have suffered from small sample sizes that render a detection of significant treatment effects difficult, especially in the presence of high placebo response rates, which are typical for all IBS trials. Several of the observed symptom improvements across studies have just barely missed the level of statistical significance. Therefore, treatment effects might have been missed because the studies were probably underpowered.

CONCLUSION

In summary, data from basic and clinical research suggest that probiotics may have the potential to influence different IBS disease mechanisms by modulating immune-responses, changing the intraluminal milieu or influence visceral sensory and motor functions. To date, results from placebo controlled clinical trials evaluating the treatment effects of probiotics in IBS vary considerably. However, different study designs, different IBS populations and most important, different probiotic preparations with different strains, different amounts, different mediums and different combinations complicate comparisons between trials. Nevertheless, most studies indicate potential beneficial effects of probiotics in the treatment of IBS and encourage further research in this field.

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