Gastroentérologie Clinique et Biologique (2008) **32**, 147—152

Disponible en ligne sur www.sciencedirect.com

ScienceDirect

ORIGINAL ARTICLE

A double blind randomized controlled trial of a probiotic combination in 100 patients with irritable bowel syndrome

Étude randomisée en double insu contre placebo sur l'efficacité d'un mélange probiotique chez 100 patients présentant des troubles fonctionnels intestinaux

S. Drouault-Holowacz[a], S. Bieuvelet[a], A. Burckel[a], M. Cazaubiel[b], X. Dray[c], P. Marteau[c,*]

[a] *PiLeJe, 37, quai-de-Grenelle, bâtiment Pollux, 75738 Paris cedex 15, France*
[b] *Biofortis, 42, rue de la Tour-d'Auvergne, 44200 Nantes, France*
[c] *Département de pathologie digestive, université et AP—HP-Paris-7, hôpital Lariboisière, 2, rue Ambroise-Paré, 75010 Paris, France*

Available online 4 March 2008

Summary
Objectives. — The purpose of this study was to evaluate the effects of a probiotic combination on symptoms in patients with irritable bowel syndrome (IBS).
Methods. — We investigated the efficiency of a probiotic dietary supplement, containing four strains of lactic acid bacteria, on symptoms of IBS. One hundred and sixteen patients with IBS fulfilling the Rome II criteria were randomized in a parallel group, double-blind study to receive a placebo or a probiotic combination (1×10^{10} cfu once daily) for four weeks. The symptoms that were monitored weekly included discomfort, abdominal pain, and stool frequency and quality. Quality of life was assessed before and at the end of the treatment using the SF36 and FDD-quality-of-life questionnaires.
Results. — One hundred subjects completed the study (48 probiotic combination, 52 placebo). The probiotic combination was not superior to the placebo in relieving symptoms of IBS (42.6 versus 42.3% improvement). However, the decrease of abdominal pain between the first and the fourth week of treatment was significantly higher in probiotic treated patients (−41.9 versus −24.2%, $P = 0.048$). Interesting findings from the IBS sub-groups were also observed such as a lower pain score at end point in patients with alternating bowel habits ($P = 0.023$) and

* Corresponding author.
E-mail address: philippe.marteau@lrb.aphp.fr (P. Marteau).

0399-8320/$ — see front matter
doi:10.1016/j.gcb.2007.06.001

an increase of stool frequency in the constipated sub-group from the first week of probiotic treatment ($P = 0.043$).
Conclusions. — The probiotic combination was not significantly superior to the placebo in relieving symptoms of IBS. Despite the apparent high placebo response, interesting findings from IBS sub-groups were observed in the field of abdominal pain and stool frequency.

Résumé
Objectifs. — L'objectif de cette étude était d'évaluer l'efficacité d'un mélange probiotique chez des patients présentant des troubles fonctionnels intestinaux (TFI).
Méthodes. — Nous avons évalué l'efficacité d'un complément alimentaire à base de probiotiques (contenant quatre souches de bactéries lactiques) sur les symptômes associés aux TFI. Cent seize patients souffrant de TFI identifiés selon les critères de Rome II ont été inclus dans une étude clinique randomisée en double insu contre placebo. Ils ont reçu pendant quatre semaines un placebo ou un mélange probiotique (1×10^{10} ufc une fois par jour). Les symptômes étudiés ont inclus l'inconfort, la douleur abdominale, ainsi que la fréquence et la qualité des selles. La qualité de vie a été évaluée avant et à la fin du traitement en utilisant les questionnaires SF36 et FDD-*quality-of-life* (QOL).
Résultats. — Cent sujets ont terminé l'étude (48 sous probiotiques, 52 sous placebo). Le mélange probiotique ne s'est pas révélé supérieur au placebo dans le soulagement des symptômes associés aux TFI (42,6 versus 42,3 % d'amélioration). Cependant, la diminution de la douleur abdominale entre la première et la dernière semaine de traitement était significativement plus importante chez les patients ayant consommé le mélange probiotique (−41,9 versus −24,2 %, $p = 0{,}048$). De plus, l'analyse des sous-groupes (TFI avec diarrhée, constipation ou alternance des deux) a permis de mettre en évidence une diminution du score de douleur abdominale chez les patients avec alternance diarrhée—constipation ($p = 0{,}023$) et une augmentation dès le première semaine de traitement de la fréquence des selles chez les patients souffrant de TFI avec constipation prédominante ($p = 0{,}043$).
Conclusions. — Le mélange probiotique n'a pas été supérieur au placebo concernant le soulagement des symptômes associés aux TFI. Cependant, des résultats intéressants ont été obtenus plus spécifiquement dans le domaine de la douleur abdominale et de la fréquence des selles.

Introduction

Irritable bowel syndrome (IBS) is a frequent disorder affecting twice as many women as men. Symptoms include abdominal pain and/or discomfort often associated with abnormal bowel habits [1] which alter the quality of life [2]. There is presently no curative treatment but various therapies may help to alleviate the symptoms [3]. The majority of them target gastrointestinal motility, and/or visceral sensitivity or psychological components of the disease [4]. Recent studies have suggested that intestinal microbiota play a role in the pathogenesis of IBS. IBS occurs more frequently after intestinal infection or antibiotic treatment and exhibits also some signs of minimal intestinal inflammation [5,6]. This provided a rationale to evaluate probiotics to correct the altered microflora and improve symptoms of IBS [7]. Probiotics are live microorganisms which when consumed in adequate amounts confer a health benefit on the host [8]. They can influence immune functions, motility, and the intraluminal milieu [9]. Evidence for the clinical efficacy of some strains or combination of several strains has been obtained for various intestinal disorders including gastroenteritis, antibiotic-associated diarrhea, *Clostridium difficile* infections, pouchitis and prevention of the relapse of ulcerative colitis [10]. The effects of probiotics may differ greatly between two closely related strains (even thus within the same microbial genus and species) and they cannot therefore be extrapolated from one strain to another [11]. Recent trials have shown the potential benefits of some probiotics, especially strains of bifidobacteria or lactobacilli, on symptoms of IBS [12—14] however, only three of these trials included more than 60 subjects [14—16].

The aim of the present study was to examine whether oral administration of a probiotic dietary supplement, for four weeks, would decrease IBS symptoms and improve the quality of life in patients suffering from IBS.

Materials and methods

Study population

One hundred and sixteen patients fulfilling the Rome II criteria for IBS [1] were enrolled in this controlled, double-blind, randomized study. The recruitment and the diagnostic work up were performed by 23 French general practitioners across France between September 2004 and January 2006. The study was approved by the ''CCPPRB HEGP-Broussais'' Ethics Committee on July 20th 2004.

The exclusion criteria were: the presence of any active organic gastrointestinal disease, abdominal surgery in the past (except for appendectomy and cholecystectomy), any concomitant disease susceptible to influence IBS, pregnancy, and an abdominal discomfort/pain score inferior to 1 at the time of randomization. All patients gave written informed consent prior to the study after they had read materials describing the study and been verbally briefed on the double-blind nature of the study, the treatment conditions,

the evaluative method, and the study procedures. Any medications, which could influence IBS, had to be discontinued before entry into the trial and during its entire duration. This included intestinal motility modifiers, antidepressants, opioids, narcotic analgesics and antispasmodic agents. The use of laxatives or loperamide was discouraged and had to be recorded by the patients who needed them transiently. The consumption of probiotic containing drugs, dairy products or food supplements was forbidden during the one week run-in period and during the entire trial.

Study protocol

This was a randomized, double-blind, parallel-group, placebo-controlled, four-week, study in out-patients. At baseline, physicians recorded the patients' age, gender, weight and medical history and classified their predominant disturbances of bowel function as constipation-predominant (C-IBS), diarrhea-predominant (D-IBS) or alternating (A-IBS) according to the Rome II criteria [1].

Patients with a discomfort/pain score superior or equal to 1 at baseline (Week 0 [Wk0]) were randomized (using a randomization table and sealed envelopes). The discomfort/pain score was assessed using a 0—3 likert scale (0: none, 1: tiny; 2: moderate, 3: severe) [18].

Patients were randomized to receive either the probiotic combination (containing 1.10^{10} cfu *B. longum* LA 101 (29%), *Lb. acidophilus* LA 102 (29%), *L. lactis* LA 103 (29%) and *S. thermophilus* LA 104 (13%), 2.3 g glucids including 1.9 g starch, 0.027 g proteins, 0.015 g lipids/sachet) or the placebo (of identical composition except for the bacteria) for four weeks. Each treatment was provided in identical sachets and taken once daily in the fasting state, at least three hours after a meal and 15 min before the next meal. The powder had to be dissolved in water 10 min before its ingestion. At the end of the protocol, patients had to bring back the empty sachets.

Each week, the patients filled in a questionnaire regarding their symptoms. Primary efficacy was assessed using a binary scale based on the patient answers to the following question: ''Did you have satisfactory relief of your overall IBS symptoms during the last week?'' [17]. The patients were also asked each week to answer the following question: ''Compared to the way you usually felt before entering the study, how would you rate your relief of symptoms of abdominal discomfort/pain during the last week? Possible answers were: completely relieved, considerably relieved, somewhat relieved, unchanged or worse'' [18]. Secondary endpoints included weekly assessment of discomfort/pain [18], abdominal pain using a 10 cm visual analogue scale (0: not at all; 10: acute, unimaginable), and stool frequency and consistency measured by subjective evaluation (very hard, hard, mould, soft, liquid). Additionally, patients completed the IBS specific FDD-quality-of-life (QOL) [19] questionnaire and the validated French version of the generalist SF-36 [20] questionnaire at baseline and at the end of the four weeks treatment to assess their quality of life and well-being.

Statistical methods

Week 0 (Wk0) was considered the baseline. As designated by the protocol, the primary comparisons for efficacy were the weekly ''satisfactory relief'' and the pain/discomfort score in the last week of the treatment period (Wk4). Differences between the treatment groups were analyzed applying the two-sided χ^2-test at a significance level of $\alpha = 0.05$, or Fisher exact test, as appropriate. Answers on visual analogue scales were measured in centimetres of the distance separating the point corresponding to the answer given by the subject from the origin of the analogical scale. These results were used for the statistical analysis. The values were compared by a nonparametric test of Wilcoxon's rank-sum test. The percentage of variation were calculated using the formula ([Wk4—Wk0]/Wk0) × 100. The analyses were done for the whole population and for the IBS subtypes (C-IBS, D-IBS and A-IBS).

We arbitrary chose ''satisfactory relief'' according to the Rome II criteria and a hypothesis 20% of positive responses to the primary end point in the patients receiving the placebo [17] and 50% in the probiotic group. It was calculated that 96 patients were required to complete the study in order to detect this difference with a 90% power and $\alpha = 0.05$. In order to compensate for dropouts, it was planed to recruit 110 patients. All data were collected and analyzed independently of the investigators, who did not have access to the data until the study had been completed. Thereafter, investigators had full access to all data.

Table 1 Characteristics of the patients receiving the probiotic combination and those in the control group.
Caractéristiques des patients dans les groupes probiotique et placebo.

	Probiotic	Placebo
Number of patients	48	52
Age	47 ± 14	44 ± 14
Female gender % (*n*)	83.3% (40)	69.2% (36)
Subtype of IBS (%)		
C-IBS	25.0	32.7
D-IBS	29.2	28.8
A-IBS	45.8	36.5
ND	0	1.9
Discomfort/pain score	2.14 ± 0.39	2.10 ± 0.45

ND: not determined.

Results

Subjects

One hundred and sixteen were screened of whom 106 fulfilled the inclusion criteria and were randomized (six declined, four had a pain/discomfort score < 1). Five patients in the probiotic group and one patient in the placebo one were excluded from the study because of low compliance and not having returned the diary and the empty sachets. Thus, the efficacy evaluation included 100 patients, 48 in the probiotic group and 52 in the placebo group. No significant differences were observed between the groups at baseline (Table 1). Patients were predominantly female (76%, $n = 76$), with a mean age of 46 years and a discomfort/pain score of 2.12 (range 1.2—3). Within the total population 29% were classified as constipation-predominant IBS (C-IBS), 29% as diarrhea-predominant IBS (D-IBS), 41% as alternators (A-IBS), and 1% non-classified.

Response to treatment

Primary efficacy variable

The proportion of patients with satisfactory relief of overall IBS (Fig. 1) and of abdominal discomfort/pain increased with time in both treatment groups. At week four, the percentage of patients with satisfactory relief was not significantly different between the two treatment groups (42.6 versus 42.3% for probiotic and placebo respectively).

Secondary efficacy variables

Abdominal pain improved significantly during the study ($P < 0.02$ for all comparisons) in both treatment groups, and in the three IBS sub-groups. The decrease in abdominal pain score between the

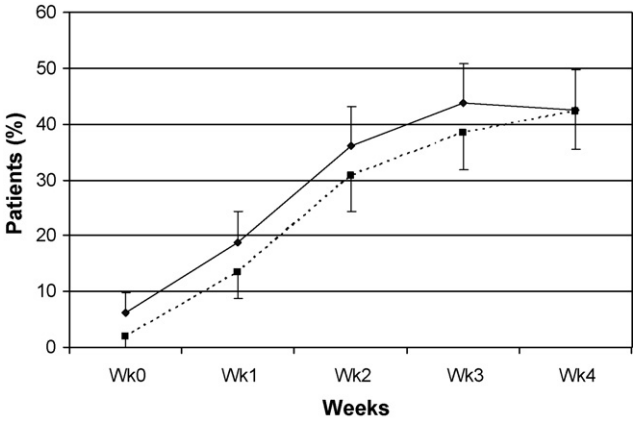

Figure 1 Percentage of patients with satisfactory relief from symptoms of IBS, in the probiotic group (solid line) and in the placebo group (dotted line).
Pourcentages de patients présentant un soulagement de leurs symptômes associés aux TFI, dans le groupe probiotique (trait plein) et dans le groupe placebo (pointillés).

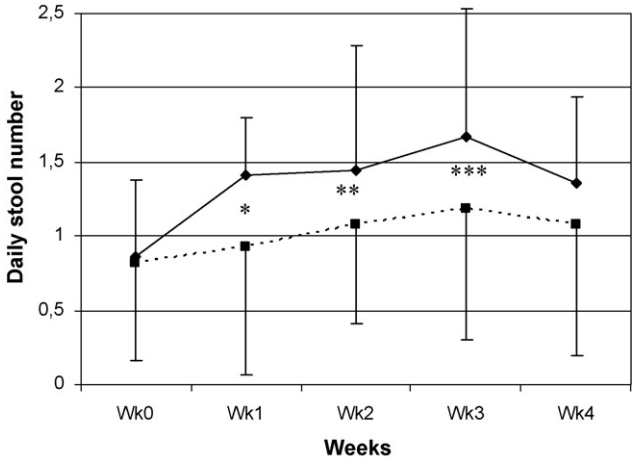

Figure 3 Daily stool number in the constipated-predominant sub-group in the probiotic group (solid line) and in the placebo group (dotted line).
$P = 0.043$, $^{**}P = 0.026$, $^{***}P = 0.049$.
Nombres de selles quotidiens dans le sous-groupe à prédominance diarrhée, dans le groupe probiotique (trait plein) et dans le groupe placebo (pointillés).
$p = 0{,}043$, $^{**}p = 0{,}026$, $^{***}p = 0{,}049$.

first and the fourth week of treatment was significantly higher in the probiotic treated patients ($-41.9\% \pm 44.6$ versus $-24.2\% \pm 51.1$, $P = 0.048$). There was also a trend for a lower abdominal pain score at the end of treatment in the probiotic group when compared to the placebo (2.7 ± 2.1 in the probiotic group versus 3.3 ± 2.2 for the placebo, $P = 0.054$) (Fig. 2A). In the A-IBS group, interesting results were observed with a significant decrease of abdominal pain (2.5 ± 1.7 in the probiotic group versus 3.6 ± 2.1 in the placebo group, $P = 0.023$) (Fig. 2B).

The number and consistency of stools were not significantly different between the probiotic group and the placebo group (not shown). In the constipated population (C-IBS), the number of stools was significantly higher in the probiotic treated group compared to placebo (Fig. 3). This was observed from the first week of treatment ($P = 0.043$ at the end of the first week; $P = 0.026$ at the end of the second week, $P = 0.049$ at the end of the third week).

There was no significant difference in the evolution of quality of life scores (SF-36 and FDD-QOL) between the probiotic and placebo groups. In contrast to placebo treated patients, patients supplemented with the probiotic combination reported a significant improvement regarding flatulence ($P = 0.037$), waking up during the night because of abdominal pain ($P = 0.031$), and needing to loosen their belt or lie down after meal ($P = 0.010$) (Table 2). Bloating improved in both groups: $P = 0.013$ and 0.028 for the probiotic and placebo groups respectively.

Discussion

Several clinical trials recently evaluated the efficacy of probiotics on IBS but many are derived from open studies and there are considerable differences in trial design and the probiotics employed [12,13]. Their primary end points were also often not clearly stated and only three, including the present study, included more than 100 patients [14,21]. Results cannot be extrapolated from one probiotic to another and depend on the dose [14]. The rationale for trying probiotics on IBS relies on their potential effectiveness on intestinal motility, microflora, inflammation and pain [22].

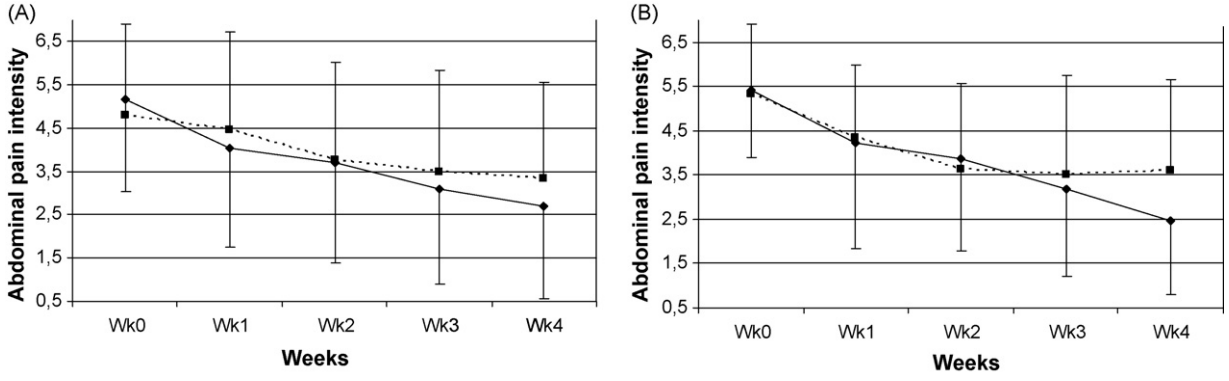

Figure 2 Abdominal pain score in total IBS subjects A; in the alternators sub-group B, in the probiotic group (solid line) and in the placebo group (dotted line).
Scores de douleur abdominale chez la totalité des patients avec IBS A ; dans le groupe avec alternance diarrhée-constipation B, dans le groupe probiotique (trait plein) et dans le groupe placebo (pointillés).

Table 2 Evolution of some quality of life scores in the probiotic and placebo groups (percents of answers).
Évolution de plusieurs scores de qualité de vie dans les groupes probiotique et placebo (pourcentages de réponses).

	Never		Rarely		Occasionnally		Often		Very often	
	Wk0	Wk4	Wk0	Wk4	Wk0	Wk4	Wk0	Wk4	Wk0	Wk4
Have you ever been bothered flatulence problems?										
Probiotic ($P = 0.037$)	2.08	10.42	12.50	33.33	25.00	18.75	39.58	25.00	20.83	12.50
Placebo ($P > 0.05$)	5.88	9.62	15.69	23.08	11.76	28.85	52.94	30.77	13.73	7.69
Have you ever been awakened by digestive troubles or discomfort?										
Probiotic ($P = 0.031$)	14.85	39.58	39.58	25.00	31.25	29.17	14.58	6.25	0.00	0.00
Placebo ($P > 0.05$)	31.37	46.15	21.57	25.00	33.33	23.08	11.76	1.92	1.96	3.85
Have you ever felt the need to loosen your belt, or lie down after a meal?										
Probiotic ($P = 0.009$)	8.33	37.50	25.00	18.75	41.67	27.08	22.92	16.67	2.08	0.00
Placebo ($P > 0.05$)	11.76	30.77	21.57	23.08	33.33	26.92	25.49	15.38	7.84	3.85
Have you ever been bothered by bloating?										
Probiotic ($P = 0.001$)	0	14.58	8.33	20.83	14.58	25.00	54.17	31.25	22.92	8.33
Placebo ($P = 0.003$)	1.96	11.54	17.65	17.31	13.73	25.00	43.14	40.38	23.53	5.77
Do you have difficulties participating in your leisure activities?										
Probiotic ($P = 0.038$)	39.58	68.75	37.50	18.75	14.58	8.33	8.33	4.17	0.00	0.00
Placebo ($P > 0.05$)	39.22	50.98	31.37	27.45	25.49	17.65	1.96	3.92	1.96	0.00

For example, L. paracasei NCC2461 significantly attenuated muscle dysfunction in a murine model of postinfective IBS [23]. The probiotic yeast *Saccharomyces boulardii* modulated the expression of neuronal markers in the submucous plexus of pigs [24]. *L. farciminis* treatment prevented stress induced hypersensitivity, increase in colonic paracellular permeability, and colonocyte myosin light chain phosphorylation in rats [25,26]. Recently, Rousseaux et al., showed that oral administration of specific *Lactobacillus* strains induced, through the NF-κB pathway, MOR1 and CB2 expression and contributed to the modulation and restoration of the normal perception of visceral pain [27]. There also seems to be an inflammatory component and a dysregulation of pro- and anti-inflammatory cytokines in patients with IBS [15]. Most interestingly, B. infantis 35624 was shown to restore the balance of pro- and anti-inflammatory cytokines in the patients [15]. Double blind randomised controlled trials also showed that B. animalis 173-010 shortened the colonic transit time in healthy women [28]. However, the fine mechanisms for these effects are still poorly understood and need further study.

This study was the first trial with this product (combination of four lactic acid bacteria strains) and we had to arbitrarily choose the duration of the product ingestion (four weeks) and the primary end point. We chose to limit the duration of the trial to four weeks as we thought that this was a sufficient period of time for patients to wait for results; however, longer studies may be useful in the future. The evaluation of the effects on IBS is difficult and there is no gold standard. There are no biological markers for IBS nor any obvious markers to use when assessing the severity of IBS symptoms. We arbitrarily decided to use a subjective global assessment of IBS symptoms improvement as the primary endpoint, individual IBS symptoms as secondary endpoints and validated quality-of-life questionnaires as proposed in the guidelines of the Rome Committee on treatment trials; however, other authors have chosen to focus on the improvement of specific symptoms such as bloating.

The results of this study indicate that supplementation with a specific combination of probiotics for four weeks was not superior to the placebo in relieving IBS symptoms. In agreement with many studies on IBS, we observed a high placebo responses rates. In clinical IBS trials, it is not uncommon to have placebo rates as high as 50% [29]. We cannot exclude therefore that the sample size of this study resulted in a lack of power to detect a significant improvement with the probiotic treatment compared with placebo.

Analysis of the IBS sub-groups showed interesting results. A significant reduction in the intensity of the abdominal pain was observed in the alternators subtype, and the mechanism for this effect is not established. Moreover, we observed a significant increase in the number of stools per day in the constipated patients. This strongly suggests that this probiotic supplementation could be effective to alleviate specific IBS symptoms and that these subgroups and specific endpoints warrant further investigation. The probiotic combination used in this study needs also to be evaluated further to determine the optimal regimen in terms of duration and dosage. It would be interesting to consider too the instability between D-IBS and C-IBS, with a tendency to move to A-IBS [30].

Global quality of life scores were not significantly different between the probiotic and placebo groups. The study was not designed to detect significant change in quality of life and it is usually very difficult to show an amelioration of quality of life with a treatment of such a short duration (four weeks) and a small number of subjects. However, our data showed positive and significant evolutions in quality of life scores for the probiotic group from the beginning to the end of the complementation for specific items relative to discomfort and to digestive disorders, mainly flatulence and bloating.

Acknowledgements

The authors thank the investigators of this study: Artus N., Muret; Rodriguez R., Tournefeuille; Mainguy J.-M., Fougères; Jouet B., Echirolles; Peignier M., Nice; Guth E., St-Louis; Viville M., Paris; Lescure P., Urrugne; Courtade J.-P., Brive la Gaillarde; Lasmenes J., Auch; Mauvezin P., Vers pont du Gard; Oliver P., Cannes; Henri J.-J., Saumur; Verdurand O., Millau; Warnet J., Paris; Anthonissen A., Valbonne; Geoffroy D., Thouaré/Loire; Vaillant A.., La flèche; Amendola, M., La Cadière d'Azur; Nejman M., Le Lavandou; Moron M., Chateaubriand; Tournesac P., Paris; Letexier R., St-Malo.

This study was sponsored by PiLeJe, 37, quai-de-Grenelle, bâtiment Pollux, 75738 Paris cedex 15, France.

References

[1] Thompson WG, Longstreth GF, Drossman DA, Heaton KW, Irvine EJ, Muller-Lissner SA. Functional bowel disorders and functional abdominal pain. Gut 1999;45(Suppl. 2):II43—7.

[2] ten Berg MJ, Goettsch WG, van den BG, Smout AJ, Herings RM. Quality of life of patients with irritable bowel syndrome is low compared to others with chronic diseases. Eur J Gastroenterol Hepatol 2006;18:475—81.

[3] Lesbros-Pantoflickova D, Michetti P, Fried M, Beglinger C, Blum AL. Meta-analysis: the treatment of irritable bowel syndrome. Aliment Pharmacol Ther 2004;20:1253—69.

[4] Talley NJ. Pharmacologic therapy for the irritable bowel syndrome. Am J Gastroenterol 2003;98:750—8.

[5] Halvorson HA, Schlett CD, Riddle MS. Postinfectious irritable bowel syndrome — a meta-analysis. Am J Gastroenterol 2006;101:1894—9.

[6] Rhodes DY, Wallace M. Post-infectious irritable bowel syndrome. Curr Gastroenterol Rep 2006;8:327—32.

[7] Camilleri M. Probiotics and irritable bowel syndrome: rationale, putative mechanisms, and evidence of clinical efficacy. J Clin Gastroenterol 2006;40:264—9.

[8] FAO and OMS. Health and nutritional properties of probiotics in food including powder milk with live active lactic acid bacteria. Report of joint FAO/WHO expert consultation on evaluation and health and nutritional properties of probiotics in food including powder milk with live lactic acid bacteria. Cordaba, Argentina, 1—4 october 2001. FAO, OMS, eds.

[9] Borriello SP, Hammes WP, Holzapfel W, Marteau P, Schrezenmeir J, Vaara M, et al. Safety of probiotics that contain lactobacilli or bifidobacteria. Clin Infect Dis 2003;36:775—80.

[10] Bergonzelli GE, Blum S, Brussow H, Corthesy-Theulaz I. Probiotics as a treatment strategy for gastrointestinal diseases? Digestion 2005;72:57—68.

[11] Agence française de sécurité sanitaire des aliments. Effects of probiotics and prebiotics on flora and immunity in adults 2005.

[12] Floch MH. Use of diet and probiotic therapy in the irritable bowel syndrome: analysis of the literature. J Clin Gastroenterol 2005;39:S243—6.

[13] Young P, Cash BD. Probiotic use in irritable bowel syndrome. Curr Gastroenterol Rep 2006;8:321—6.

[14] Whorwell PJ, Altringer L, Morel J, Bond Y, Charbonneau D, O'Mahony L, et al. Efficacy of an encapsulated probiotic Bifidobacterium infantis 35624 in women with irritable bowel syndrome. Am J Gastroenterol 2006;101:1581—90.

[15] O'Mahony L, McCarthy J, Kelly P, Hurley G, Luo F, Chen K, et al. Lactobacillus and bifidobacterium in irritable bowel syndrome: symptom responses and relationship to cytokine profiles. Gastroenterology 2005;128:541—51.

[16] Kajander K, Korpela R. Clinical studies on alleviating the symptoms of irritable bowel syndrome. Asia Pac J Clin Nutr 2006;15:576—80.

[17] Kellow J, Lee OY, Chang FY, Thongsawat S, Mazlam MZ, Yuen H, et al. An Asia-Pacific, double blind, placebo controlled, randomised study to evaluate the efficacy, safety, and tolerability of tegaserod in patients with irritable bowel syndrome. Gut 2003;52:671—6.

[18] Muller-Lissner SA, Fumagalli I, Bardhan KD, Pace F, Pecher E, Nault B, et al. Tegaserod, a 5-HT(4) receptor partial agonist, relieves symptoms in irritable bowel syndrome patients with abdominal pain, bloating and constipation. Aliment Pharmacol Ther 2001;15:1655—66.

[19] Chassany O, Marquis P, Scherrer B, Read NW, Finger T, Bergmann JF, et al. Validation of a specific quality of life questionnaire for functional digestive disorders. Gut 1999;44:527—33.

[20] Leplege A, Ecosse E, Verdier A, Perneger TV. The French SF-36 health survey: translation, cultural adaptation and preliminary psychometric evaluation. J Clin Epidemiol 1998;51:1013—23.

[21] Kajander K, Hatakka K, Poussa T, Farkkila M, Korpela R. A probiotic mixture alleviates symptoms in irritable bowel syndrome patients: a controlled 6-month intervention. Aliment Pharmacol Ther 2005;22:387—94.

[22] Santosa S, Farnworth E, Jones PJ. Probiotics and their potential health claims. Nutr rev 2006;64:265—74.

[23] Verdu EF, Bercik P, Bergonzelli GE, Huang XX, Blennerhasset P, Rochat F, et al. Lactobacillus paracasei normalizes muscle hypercontractility in a murine model of postinfective gut dysfunction. Gastroenterology 2004;127:826—37.

[24] Kamm K, Hoppe S, Breves G, Schroder B, Schemann M. Effects of the probiotic yeast Saccharomyces boulardii on the neurochemistry of myenteric neurones in pig jejunum. Neurogastroenterol Motil 2004;16:53—60.

[25] Lamine F, Fioramonti J, Bueno L, Nepveu F, Cauquil E, Lobysheva I, et al. Nitric oxide released by Lactobacillus farciminis improves TNBS-induced colitis in rats. Scand J Gastroenterol 2004;39:37—45.

[26] it-Belgnaoui A, Han W, Lamine F, Eutamene H, Fioramonti J, Bueno L, et al. Lactobacillus farciminis treatment suppresses stress induced visceral hypersensitivity: a possible action through interaction with epithelial cell cytoskeleton contraction. Gut 2006;55:1090—4.

[27] Rousseaux C, Thuru X, Gelot A, Barnich N, Neut C, Dubuquoy L, et al. Lactobacillus acidophilus modulates intestinal pain and induces opioid and cannabinoid receptors. Nat Med 2007;13:35—7.

[28] Marteau P, Cuillerier E, Meance S, Gerhardt MF, Myara A, Bouvier M, et al. 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.

[29] Pitz M, Cheang M, Bernstein CN. Defining the predictors of the placebo response in irritable bowel syndrome. Clin Gastroenterol Hepatol 2005;3:237—47.

[30] Mearin F, Baro E, Roset M, Badia X, Zarate N, Perez I. Clinical patterns over time in irritable bowel syndrome: symptom instability and severity variability. Am J Gastroenterol 2004;99:113—21.