Pediatric functional constipation treatment with *Bifidobacterium*-containing yogurt: A crossover, double-blind, controlled trial

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**Abstract**

**AIM:** To evaluate the treatment of pediatric functional chronic intestinal constipation (FCIC) with a probiotic goat yogurt.

**METHODS:** A crossover double-blind formula-controlled trial was carried out on 59 students (age range: 5-15 years) of a public school in Belo Horizonte, MG, Brazil, presenting a FCIC diagnostic, according to Roma III criteria. The students were randomized in two groups to receive a goat yogurt supplemented with 10⁹ colony forming unit/mL *Bifidobacterium longum* (*B. longum*) (probiotic) daily or only the yogurt for a period of 5 wk (formula). Afterwards, the groups were intercrossed for another 5 wk. Defecation frequency, stool consistency and abdominal and defecation pain were assessed.

**RESULTS:** Both treatment groups demonstrated improvement in defecation frequency compared to baseline. However, the group treated with probiotic showed most significant improvement in the first phase of the study. An inversion was observed after crossing over, resulting in a reduction in stool frequency when this group was treated by formula. Probiotic and formula improved stool consistency in the first phase of treatment, but the improvement obtained with probiotic was significantly higher (*P* = 0.03). In the second phase of treatment, the group initially treated with probiotic showed worsening stool consistency when using formula. However, the difference was not significant. A significant improvement in abdominal pain and defecation pain was observed with both probiotic and formula in the first phase of treatment, but again the improvement was more significant for the group treated with *B. longum* during phase I (*P* < 0.05). When all data of the crossover study were analyzed, significant differences were observed between probiotic yogurt and yogurt only for defecation frequency (*P* = 0.012), defecation pain (*P* = 0.046) and abdominal pain (*P* = 0.015).

**CONCLUSION:** An improvement in defecation frequency and abdominal pain was observed using both supplemented and non-supplemented yogurt, but an additional improvement with *B. longum* supplementation was obtained.

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**Key words:** Functional chronic constipation; Probiotic; *Bifidobacterium longum*; Yogurt; Adolescents; Children
INTRODUCTION

The worldwide prevalence of childhood constipation in the general population ranges from 0.7% to 29.6%[1], and the wide range indicates differences in definition and selection of patients. This functional defecation disorder is characterized by infrequent defecation less than three times per week, frequent episodes of fecal incontinence, the periodic passage of large and painful stools which clog the toilet, and retentive posturing. Upon physical examination a palpable fecal mass is often found in the abdomen and the rectum. Accompanying symptoms may include irritability, decreased appetite, and/or early satiety. In the vast majority of cases (90% to 95%), no underlying organic cause is found and functional constipation is diagnosed[2].

The standard treatment consists of disimpaction and the administration of laxatives to achieve a normal bowel habit of passing a soft stool without pain. Even though the traditional treatment is well established and safe, for many patients it does not provide a satisfying improvement, prompting interest in other therapeutic strategies[3,4].

Probiotics are increasingly being used as an alternative in the management of constipation. Probiotics are defined as live microorganisms which when administered in adequate amounts confer a benefit on the host health[5]. In a recent review, the efficacy and safety of probiotic supplementation for the treatment of constipation was evaluated[6]. Studying 5 randomized controlled trials, with a total of 377 subjects (three trials with adults and two trials with children), the data suggests a favorable effect of some strains of Lactobacillus, Bifidobacterium and Escherichia coli (E. coli). Only one of the randomized controlled trials described the ineffectiveness of Lactobacillus rhamnosus Goldin and Gorbach as an adjunct to lactulose for the treatment of constipation in children[7].

The authors of the review concluded that until more data are available, the use of probiotics for the treatment of constipation should be considered investigational. More recently, Lactobacillus reuteri administered in infants with chronic constipation had a positive effect on frequency of bowel movements, but not on stool consistency[8] and the intake of mixed probiotic strains [Lactobacillus plantarum, Bifidobacterium breve, Bifidobacterium animalis subsp. lactis (B. animalis var. lactis)] was able to relieve evacuation disorders and hard stools in healthy adults[9].

In the present study, the ingestion of goat yogurt containing a Bifidobacterium longum (B. longum) strain was evaluated for the treatment of functional chronic intestinal constipation (FCIC) in children and adolescents.

MATERIALS AND METHODS

Subjects and eligibility criteria

Children aged 5-15 years and with FCIC, referred to a public school in the central area of the city of Belo Horizonte, Minas Gerais, Brazil, were eligible for study entry. Constipation was characterized according to Rome III criteria as presenting at least two out of six of the following symptoms for two or more months: two or fewer defecations per week; at least one episode of fecal incontinence per week; history of retentive posturing or excessive volitional stool retention; history of painful or hard bowel movements; presence of a large fecal mass in the rectum; history of wide diameter stools that may obstruct the toilet[10]. Exclusion criteria were the use of any oral laxative < 4 wk before intake, metabolic disease, a history of gastrointestinal surgery and fecal incontinence. Patients with fecal incontinence were excluded in order to make the sample more homogeneous in relation to disease severity. The follow-up protocol included defecation frequency, stool consistency, and abdominal and defecation pains recorded daily by the adolescents or parents. All children older than 12 years and/or parents gave informed consent. The study was approved by the Ethical Committee in Research of the Universidade Federal de Minas Gerais (COEP/UFMG, number ETIC0506/08).

Yogurt and bacterium

The B. longum strain used in the trial was isolated from the feces of a healthy child and identified by Multiplex Polymerase Chain Reaction. This strain was selected as a candidate for probiotic use based on technological (aerotolerance and high growth rate) and beneficial (wide antagonistic spectrum against pathogenic indicators, few antimicrobial resistance) criteria. The bacterium was grown in de Man, Rogosa and Sharp broth (Difco, Sparks, United States) containing N\textsubscript{2} 10% and CO\textsubscript{2} 5%. After growth, the culture was concentrated by centrifugation and resuspended in peptone sterile water. An aliquot of 1 mL of the concentrated bacterium suspension was added to 9 mL of a commercial goat yogurt (Capril Jacomé, Contagem, Brazil) to obtain a final concentration of 10^8 colony forming unit (CFU)/mL. The control formula was prepared by addition of 1 mL of peptoned water to 9 mL of goat yogurt. The goat yogurt contained the two classical yogurt starters, Lactobacillus delbrueckii subsp. bulgaricus and Streptococcus thermophilus from the YF-L812 commercial culture (DVS - Christian Hansen Laboratory, Horsholm, Denmark). Both yogurts were maintained at 4°C until use and for a maximum of one week. During this period, the Bifidobacterium cells remained viable at 10^6 CFU/mL levels.
Probiotic (Lactobacillus straw 12, B. longum B2023) daily or the same dose of goat yogurt daily for 5 wk (randomization list were computer-generated using the Epi Info during the study. The allocation sequence and randomization was identical in weight, color, smell, taste and package. All products, goat yogurt with or without probiotic were consumed other fermented dairy products or yogurts during the study. Children were instructed to maintain their ordinary dietary habits, but were asked to avoid consuming other fermented dairy products during the study. The allocation sequence and randomization list were computer-generated using the Epi Info Program.

Inclusion and exclusion criteria

- Age: 5-15 years
- Previous treatment for intestinal constipation
- Defecation frequency: ≤ 3 times/wk
- Stool consistency: Bristol scale B1-7
- Abdominal and defecation pain: No

RESULTS

The participant flow diagram (Figure 1) shows that among 286 students interviewed, 67 (23.4%) were diagnosed with FCIC following the Roma III criteria. Seven of them were excluded based on the exclusion criteria, and the remaining students were randomized to receive the probiotic or formula treatment. After the beginning of the trial only one parental withdrawal occurred in the formula group. There was no adverse effect due to the interventions in the present study protocol.

Table 1 summarizes the subjects’ baseline demographic and clinical characteristics. The two groups were comparable in regard to age, sex, and baseline features of constipation. More female subjects than male were present in the two groups and at a similar frequency (79.3% and 80.0% in formula and probiotic groups, respectively).

Figure 2A shows and compares the evolution of the two groups for hard stool consistency (Bristol scale < 4) during the trial. An improvement was observed with both
significant improvement in defecation frequency was also noted for both groups when compared to baseline with a tendency to a slight additional improvement at the end of the first intervention when \( B.\ longum \) was supplemented, and an inversion after crossing over.

Figure 2C shows and compares the evolution of defecation pain in the two groups during the trial. An improvement was observed for both treatments in relation to the baseline, but with a better evolution for the probiotic group. However, a significant difference \( (P = 0.009) \) between formula and probiotic was observed only for phase B1, and contrarily to Figures 1 and 2 an inversion was not observed after crossing over.

Figure 2D shows and compares the evolution of abdominal pain in the two groups during the trial. When the symptomatology was compared before and after the intervention, a significant improvement was noted for both groups as compared to baseline, but again with better results for the probiotic group. However, at the end of the second intervention after crossing over, the symptomatology was similar for the two groups.

When all data of the crossover study were analyzed, significant differences were observed between probiotic yogurt and yogurt only for defecation frequency \( (P = 0.012) \), defecation pain \( (P = 0.046) \) and abdominal pain \( (P = 0.015) \).

**DISCUSSION**

The prevalence of FCIC observed in the present study \( (23.4\%) \) was similar to the data cited in the literature\(^1\). The predominance of FCIC in female subjects \( (about 80\%) \) was also described in the literature\(^2\).

Within the first week of intervention, a significant improvement in all constipation symptoms was observed in both treatment groups \( (yogurt or yogurt plus B.\ longum) \) when compared to the baseline. However, when the yogurt was supplemented with the probiotic, further improvement was obtained when compared to the yogurt only. Yogurt is generally considered to alleviate gastrointestinal conditions such as constipation and diarrhea\(^3\). However, regarding the effect of yogurt alone on constipation, few reports are available in the literature, and the results reported are contradictory. Additionally, in most of the clinical trials comparing the effect of probiotic yogurt with control yogurt, the starter lactic acid bacteria are heat-killed in the second situation, which does not correspond to the reality. In the few studies where viability of the starter strains was maintained in the control yogurt, improvement of constipation symptoms was observed in both probiotic and control groups with an increment in the first one\(^4\).

There are several hypotheses to explain how probiotics might have therapeutic potential for the treatment of constipation. Firstly, quite old and well known observations showed that the absence of gut microbiota in germ-free animals result in abnormal characteristics of the intestinal morphology and function such as increased transit time of contents, altered myenteric neurons, impaired intestinal muscle function and decreased intestinal...
mass\(^{13,14}\). Interestingly, the mono-association of germ-free animals with *Lactobacillus acidophilus* or *Bifidobacterium bifidum* reduced the migrating myoelectric complex period and accelerated the small intestinal transit. Inversely, some *E. coli* strains presented an inverse effect when mono-associated in gnotobiotic animals\(^{15}\). Short-chain fatty acids (SCFAs), main metabolic products derived from the fermentative activity of the gut microbiota, have a direct influence on intestinal motility through the Gpr41 receptor\(^{16}\). In colonized Gpr41 knockout mice, an increased intestinal transit rate was associated with a reduced expression of peptide YY, an enteroeendocrine cell-derived hormone that normally inhibits gut motility\(^{17}\). Secondly, there are some data suggesting differences in the intestinal microbiota of healthy individuals and patients with chronic constipation\(^{17,18}\). The main features were an increased number of clostridia and enterobacteria, and a decrease in bifidobacteria and lactobacilli. These differences have an influence on the metabolic profile of the gut environment, and particularly on SCFA pattern\(^{19}\). However, a key question is if this dysbiosis is a secondary manifestation of constipation, or is a factor contributing to constipation. Another set of data favoring the microbiota influence describes the higher defecation frequency and softer stool consistency in breast-fed than in formula-fed infants in the first four months of life, which can be due to the higher fecal levels of bifidobacteria in breast-fed infants\(^{20}\). Thirdly, studies involving the administration of *Bifidobacterium animalis* subspecies *lactis* DN-173010 have shown improved colonic transit times, both in a healthy population\(^{21}\) and in constipated patients\(^{22}\). Another study showed that the intake of probiotic (*Lactobacillus helveticus* and *B. longum*) can modify the gut microbial ecology and metabolic profiles\(^{23}\). Finally, in a study using a guinea-pig isolated tissue model, results showed that cytoplasmatic fraction of probiotic bacteria (*Lactobacillus, Bifidobacterium*) stimulated the contraction of the ileum segment and induced proximal colon relaxation\(^{24}\).

In conclusion, an improvement in constipation symptoms was observed using both supplemented and non-supplemented yogurt. An additional improvement with *B. longum* supplementation was suggested in the present intercrossed double-blind formula-controlled study.

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