

EFFECTS OF *SACCHAROMYCES BOULARDII* IN CHILDREN WITH CHRONIC DIARRHEA, ESPECIALLY CASES DUE TO GIARDIASIS

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SUMMARY

40 babies aged 6 to 36 months were studied, suffering from chronic diarrhea. 35 of them had *Giardia lamblia* parasites and had been treated with tinidazole, 4 had been infected with *Shigella* and had been treated with trimetopim + sulfametoxazole, and there was 1 case of diarrhea from an undetermined cause. Despite the specific treatment and elimination of the underlying causes the patients continued to have diarrhea, which constituted the criterion for inclusion in this clinical study. The double-blind mode and random assignment to groups were used. Group A received *Saccharomyces boulardii* (S.b.) in granulated form in two daily doses of 250 mg for a month, while group B was given a placebo with the same characteristics but without the main active ingredient, in the same dosage and for the same time. The two groups were compared by the chi-squared test for two populations at significance level $\alpha=0.01$. A 65% reduction in stools was achieved in group A (3-6 at the beginning and 1-3 at the end) and one of only 15% in group B, significantly in favour of the group treated with S.b. ($p=0.0006$). As far as the status of the jejunal mucosa, established by biopsy before and after treatment is concerned, in the group that received S.b. recovery was predominant among the patients with partial atrophy of the villi, while in the control group no change was observed in the jejunum histology. The overall evaluation of the clinical and histological response to the treatment in both groups shows that 70% of the patients improved, 25% remained unchanged, and only 5% got worse, while in the control group 50% showed no change in condition, 40% got worse, and only 10% improved, this difference being highly significant ($p=0.0007$). The study confirms the beneficial effect of *Saccharomyces boulardii* (Biocodex Floratil®) in chronic diarrhea caused by a postgastroenteritis syndrome, especially when it is due to giardiasis.

Keywords: *Saccharomyces boulardii*, chronic diarrhea in infants, postgastroenteritis syndrome, giardiasis.

INTRODUCTION

There are many causes of chronic diarrhea in infants. Infections with intestinal parasites are of major importance in this respect, one of the most frequent conditions being giardiasis [6]. On the other hand, a postgastroenteritis syndrome determined by acute infectious diarrhea of viral, bacterial, or parasitic origin is also a common cause of chronic diarrhea [21], with the consequences of reduced disaccharidase activity, a disturbance of the intestinal ecosystem, and dysbacteriosis.

In the treatment of chronic diarrhea in infants, consideration must be given not only to the specific cause of the problem, but also, where necessary, to substitution therapy with milk products in the presence of disaccharidase deficiency, particularly lactase, and to the maintenance of adequate nutritional state. However, there still remains much to be done to prevent the disequilibrium of the intestinal ecosystem.

The use of *Saccharomyces boulardii* (S.b.) [22] alongside oral rehydration represents a new therapy for acute diarrhea. It is extremely well tolerated, and has made it possible to achieve a significant reduction in the duration of diarrhea and in its complications [1, 3, 9, 12, 13, 23, 24, 26, 28, 29, 30].

S.b. is a non-pathogenic, non-toxic, and non-transmissible yeast strain whose specific action mechanism is such that it has a bioregulatory effect on the intestinal flora and the disaccharidase enzymes. It is a tropical yeast found in a permanent unicellular state, a eukaryote of the order Endomycetales. The strain was isolated by Boulard from certain tropical fruits in Indochina, after he had observed that their skins were eaten as an antidiarrheal by the local population. It was subsequently used in France for the treatment of diarrhea [13, 22].

This yeast, now supplied in freeze-dried form (Biocodex Ultra-Levure, Merck Floratil), shows optimal growth at temperatures above 30°C. In terms of temperature, therefore, the intestine is entirely suitable. The product is resistant to antibiotics, sulfonamides, and other microorganisms. The freeze-dried cells can be activated in any appropriate medium, e.g. the intestinal contents, under both aerobic and anaerobic conditions.

S.b. exerts a protective action in the human intestine by its mechanisms of action — antagonism of bacteria and *Candida albicans*, inhibition of the effects of bacterial toxins, reinforcement of the non-specific defence against infection, and an increase in the activity of intestinal disaccharidases [7, 8, 11, 14, 15, 17, 22, 33]. The trophic effect of *Saccharomyces boulardii* in the intestine can be explained by the release of polyamines in the intraluminal space during intestinal transit of the yeast S.b. [10]. The present study was carried out to assess the clinical and histomorphological response to treatment with *Saccharomyces boulardii* in chronic diarrhea in children under 3 years of age.

MATERIAL AND METHOD

Method:

The double-blind mode was used, patients being randomly assigned to treatment with the active product or with placebo on the basis of a table of random numbers [16].

Patients:

40 children divided into two groups of 20 were used in the study, all of whom had been suffering from diarrhea for 3 or 4 weeks. The following tests were performed in all cases prior to the start of the clinical study: routine haematological investigations (blood picture, erythrocyte sedimentation rate, reticulocyte count, serum iron), parasitological, bacteriological, and mycological investigations of the faeces to determine the pathogen causing the diarrhea, and, in addition, a jejunal biopsy.

35 of the patients had been diagnosed as infected with *Giardia lamblia*, 4 had *Shigella*, and, in 1 case the cause of the diarrhea remained undetermined.

The patients with giardiasis had been given specific treatment with tinidazole (25 mg/kg body weight per day for 5 days) and the patients with shigellosis had been treated with trimethoprim and sulfamethoxazole (60 mg/kg body weight per day for 7 days). The patient with diarrhea of undetermined cause was not treated with any antimicrobial or antiparasitic agent.

Despite the specific treatment and the disappearance of the infectious agents detected in the parasitological and bacteriological investigations of the faeces, the patients continued to suffer from diarrhea. This constituted the inclusion criterion for the clinical study with S.b.

Treatment:

Group A was treated with *Saccharomyces boulardii* as the active ingredient in the form of a granulate of 50 mg based on S.b.; group B was given placebo with the same characteristics, but without any active ingredient, at the same dose. The duration of treatment was in each case 1 month.

The products were presented in sachets of similar appearance, and the dose in each case was 2 x 250 mg/day, diluted with water or juice in accordance with the manufacturer's instructions (Biocodex, France).

Evaluation criteria:

1) Number of bowel movements:

The number of bowel movements was determined on a daily basis and assigned to categories of 1-3, 4-6, 7-10, and more than 10 per day. The determinations were begun 15 days before the start of the clinical-therapeutic study and were continued from then on, throughout the month of treatment, until 7 days after the end of the treatment.

The evaluation criterion was the difference between the numbers of bowel movements on the day before the start of this treatment (day 0) and on the last day of the treatment (day 30).

2) Degree of reversal of histological changes:

The jejunal biopsy was repeated in all patients within 7 days after the end of the 30-day treatment period, to establish the state of the jejunal mucosa before and after the therapeutic study.

For the histological investigation sections of a fragment of jejunal mucosa were stained with haematoxylin-eosin to permit identification of any structural changes in accordance with the classification system of Metayer and Laumunier [25], which distinguishes different degrees of intestinal atrophy (partial/subtotal/total), but which was here modified so as to subdivide partial atrophy into slight, moderate, and severe.

3) Clinicohistological response criterion:

On the basis of the results for the two criteria described above, three response categories were established: better, unchanged, and worse. "Better" meant a reduction in the number of bowel movements per day and in the histomorphological damage, "unchanged" meant that there was no change in these parameters or that only one of them had changed, and "worse" meant an increase in the number of bowel movements or in the severity of lesions of the jejunal mucosa.

Statistical analysis:

The chi-squared (χ^2) test for two populations was carried out to compare the numbers of bowel movements per day and the state of the jejunal mucosa in the two groups of patients at baseline with the same parameters after treatment with *Saccharomyces boulardii* or with the placebo. The significance level used was $\alpha = 0.05$.

RESULTS

40 patients divided into two groups of 20 and suffering from chronic diarrhea were assessed. The patients were between 6 and 36 months old, the majority (60%) being less than 1 year of age, and the sex distribution 1:1.

The cause of the chronic diarrhea was in most cases parasitization by *Giardia lamblia*. The patients with a diagnosis of giardiasis were more or less equally distributed between the two groups (making up 85% of group A, which was treated with *Saccharomyces boulardii*, and 90% of group B, which was given the placebo) (Table 1).

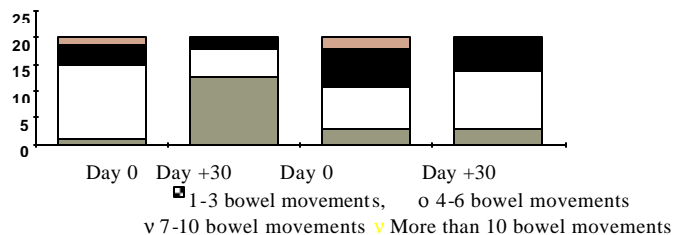
Table 1: Causes of diarrhea

FINAL DIAGNOSIS	GROUP A		GROUP B	
	n	%	n	%
DIARRHEA DUE TO PARASITIC INFECTION <i>Giardia lamblia</i>	17	(85)	18	(90)
DIARRHEA DUE TO BACTERIAL INFECTION <i>Shigella</i>	2	(10)	2	(10)
DIARRHEA OF UNDETERMINED CAUSE	1	(5)	0	(0)

Comparison of the numbers of bowel movements and the biopsy results prior to the clinical study does not reveal any significant differences between the two groups ($p = 0.44$ for bowel movements and $p = 0.95$ for the state of the jejunal mucosa).

The evaluation of bowel movements prior to treatment shows that the average number of bowel movements per day was 4-6 (70% of the patients in group A and 40% of the patients in group B). Comparison with the values after the end of the study shows a decrease in the number of bowel movements — the frequency was now 1-3 per day in 65% of the patients in group A as opposed to only 15% in group B. There was a highly significant difference in the number of bowel movements between the group given *S.b.* (group A) and the controls (group B) ($p = 0.006$), with a marked reduction in bowel movements in the group treated with *S.b.* (Fig. 1).

Fig. 1: Numbers of bowel movements before and after 1 month of treatment with *saccharomyces boulardii* in patients with chronic diarrhea

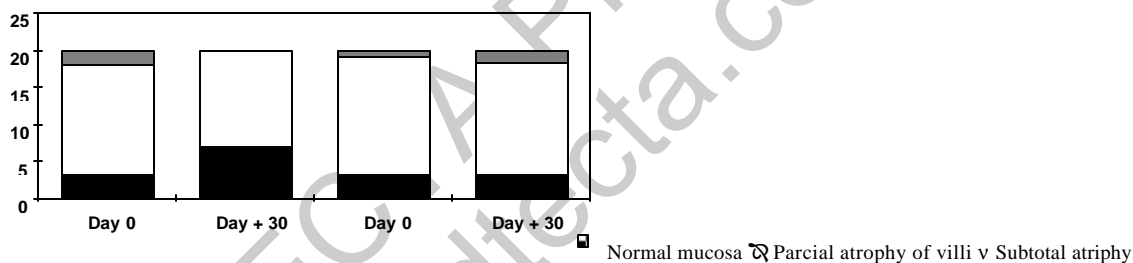


In the control group there was a small increase in the number of bowel movements; in most cases the daily number of bowel movements was still 4-6 (55%).

The histomorphological findings showed that prior to the treatment most of the 40 patients in the study had partial atrophy of the villi (PAV) (75% in group A and 80% in group B). Jejunal biopsies performed at the end of the clinical-therapeutic study still showed a majority with PAV, but in group A the proportion of patients with a normal mucosa rose from 15 to 35%, reducing the percentage with PAV and eliminating altogether the 2 cases with subtotal atrophy of the intestinal villi (who initially represented 10% of the group). It should be pointed out that there was no histological deterioration in any of the babies in this group, which demonstrates the favourable effect of treatment with S.b. (Fig. 2).

fig. 2:

Histological status before and after 1 month of treatment with *Saccharomyces boulardii* in patients with chronic diarrhea



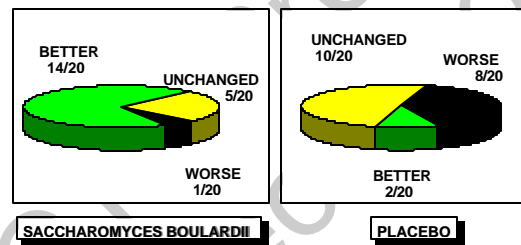
Prior to the treatment with placebo the majority (80%) of the children in group B had PAV, only 3 (15%) had a normal histological status (the same percentage as in group A), and 1 (5%) showed subtotal atrophy of the villi (SAV). After the treatment none of the patients who did not have a normal mucosa at the onset showed a normalization of their histological status; there was no significant change in the percentage with PAV (75%), and the percentage of cases with SAV increased slightly (to 10%) as a second patient now came into this category. An examination of the before/after assessments in this group shows us that there was no change compared with the baseline histological findings — in contrast to the situation in group A (Fig. 2).

When we combine the clinical criterion ("number of bowel movements") with the histological criterion ("state of the jejunal mucosa") at the end of the therapeutic study to evaluate the effect of *Saccharomyces boulardii* versus placebo, we find that the majority (70%) of the patients in the active treatment group (group A) showed an improvement, 25% remained unchanged, and only 5% (1 case) deteriorated.

In the group which was given the placebo (group B), half of the patients remained unchanged (50%), 8 (40%) deteriorated, and only 2 (10%) improved. Statistically, the difference between the two groups was highly significant ($p = 0.0007$), the situation in the active treatment group being much better than that in the controls (Fig. 3).

No adverse reactions to the administration of *Saccharomyces boulardii* were reported during the clinical study, the product being well tolerated.

fig. 3: Clinicohistological results of treatment with saccharomyces boulardii in patients with chronic diarrhea



DISCUSSION

The use of *Saccharomyces boulardii* in addition to specific treatment can provide a solution to the effects of parasitic damage of the intestinal mucosa in diarrhea caused by *Giardia lamblia* — whether acute, persistent, or chronic — and to the other phenomena which can occur in the course of chronic diarrhea caused by a postgastroenteritis syndrome and especially giardiasis, as has been demonstrated by our study, which brought to light clear evidence of clinical and histological recovery.

This is why — taking as a basis the experimental study in rats carried out by Rigother et al. [27], who reported that *S.b.* reduces the number and severity of the lesions caused by *E. histolytica* — we carried out the present clinical study in children with chronic diarrhea, particularly cases of giardiasis. The overall assessment of the clinical response and of the state of the jejunal mucosa showed an improvement in the active treatment group compared with the placebo, confirming the beneficial effects of *S.b.* in chronic diarrhea.

The clinical use of *S.b.* has been documented in various types of diarrhea. Among the most notable indications in this connection are the acute diarrheas in children and adults [3, 12, 13, 22, 26], the prevention of antibiotic-associated diarrhea [24, 30], the treatment of diarrhea caused by *C. difficile* [9, 11, 23], the treatment of chronic diarrhea in patients infected with HIV [29], and the treatment of children with a dysfunctional short small intestine aggravated by bacterial proliferation [28].

Many factors are involved in the breakdown of equilibrium of the intestinal ecosystem in diarrhea, including an immunological deficit, gross-physiological alterations, superinfections resulting from the destruction of microorganisms which maintain the microbial barrier, and suppression of the local immune response.

S.b. can play an important part in combating the harmful effects of giardiasis, as the parasite responsible for the condition is capable of upsetting the balance of the intestinal ecosystem. One must emphasize here the effect of bacterial proliferation, which is actively involved in persistence of the diarrheal symptoms in such patients in spite of specific treatment. The diarrhea does not disappear when the parasite is eliminated, as the symptoms are caused by the resistance of certain bacterial strains [32]. The value of *S.b.*, both in terms of its antimicrobial action and in the recovery of intestinal flora is thus demonstrated.

The mechanisms by which the trophozoite of *Giardia lamblia* impairs absorption have not been clarified. Damage to the intestinal mucosa with or without its invasion [2, 18, 19, 20], bacterial proliferation associated with the presence of the parasite [31, 32], and deconjugation of bile salts by the bacteria and parasites have been considered in this connection [31]. The morphological alterations of the jejunal mucosa range from an absence of lesions to severe atrophy of the intestinal villi, with changes in their height and configuration and an increased depth of the crypts. Other modifications of intestinal structure have also been reported, including acute and chronic inflammatory infiltration with destruction of the epithelial cells [5, 19].

Partial atrophy of the jejunal mucosa is the commonest finding in giardiasis-determined chronic diarrhea [5, 18] — which is in line with what we found in our patients. Repair of the damage to the intestinal mucosa is usually slow, which is why 1 month after the specific treatment there is no appreciable improvement in mucosal histology, and this can exert an influence on the persistence of the clinical symptoms.

A finding of major interest in the present study was the rapid recovery of the mucosa, irrespective of the extent to which it had been affected, in children who had received S.b. for 30 days in addition to the specific treatment. In contrast, there were no changes in the alterations of the jejunal mucosa in the group given a placebo — here the degree of partial atrophy after the 30 days of treatment was either unchanged or worse than it had been earlier.

Our results suggest that S.b. should be used in combination with specific treatment of giardiasis for a period of 1-4 weeks, depending on the clinical manifestations of the diarrhea, i.e. whether it is acute or persistent or chronic, the latter form sometimes being associated with intestinal malabsorption, whose adverse effects can be alleviated or reversed more quickly by the beneficial action of *Saccharomyces boulardii*.

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