

Clinical research

Intestinal candidiasis, *L. acidophilus* supplementation and Crook's questionnaire

D.M. BLAIR¹, C.S. HANGEE-BAUER² AND C. CALABRESE³

Twenty-six subjects with positive stool cultures for *Candida albicans* were prospectively studied in a double-blind controlled trial to determine the effect on intestinal *Candida* of supplementation with the strain of *Lactobacillus acidophilus*. Subjective evaluation and follow-up stool cultures demonstrated no significant differences between the treatment and control groups. We did however notice a positive correlation between stool culture results and score average on Crook's questionnaire. The limitations of this study are discussed, with recommendations for future studies. A correlation is observed between stool cultures and the scores from Crook's questionnaire.

INTRODUCTION

FOODS FERMENTED WITH LACTOBACILLI are of great importance to the diets of most of the world's people. Many cultures use some form of fermented food in their diet, ranging from cheese and yogurt to miso and other products. The symbiotic relationship between humankind and this genus of bacteria has a long history of important nutritional and therapeutic benefits for humans. Various lactobacillus species are used as dietary adjuncts, most commonly *Lactobacillus acidophilus*. (1)

Lactobacilli have long been noted for the role they play in the prevention and treatment of diseases, particularly those of the gastrointestinal tract and vagina. As part of the normal flora, they inhibit the growth of other organisms through competition for nutrients, alteration of pH and oxygen tension to levels less favorable to pathogens, prevention of attachment of pathogens by physically covering attachment sites, and production of limiting factors such as antimicrobial factors. Lactobacilli have been shown to produce hydrogen peroxide, lactic acid, acetic acid, and benzoic acid (2), and other inhibitory substances are suspected.(3) Since the environment of the intestinal tract is both aerobic and anaerobic, *L. acidophilus* has been studied under both these conditions. Such studies suggest that *L. acidophilus* may help restrict the growth of organisms in a wide variety of conditions.(3)

¹ D.D.S., N.D. Bastyr College Research Department, Seattle WA
² N.D. Bastyr College Research Department, Seattle WA
³ N.D. Bastyr College Research Department, Seattle WA
Contact address: 144 NE 54th ST., Seattle WA 98105, USA

The presence of *Candida albicans* in the intestinal tract has received much attention in recent years due to its possible role in gastrointestinal and systemic disease.(4,5) Typically, antifungal substances such as *Nystatin*® are used to combat this micro-organism.

Lactobacillus acidophilus has been shown to retard the growth of *Candida albicans* in vitro.(2) *C. albicans* grows more slowly, and the maximal population is slightly less in filtrates of *L. acidophilus*. Clinical studies have suggested that the addition of lactobacilli to the vagina can assist in preventing the recurrence of vaginal candidiasis.(2) These and other studies raise the possibility that lactobacilli may be beneficial in limiting intestinal *Candida*.

This study investigated the *in vivo* effect of oral administration of *Lactobacillus acidophilus* on *Candida albicans*. The effects were measured with both a questionnaire and a stool culture.

MATERIALS AND METHODS

This study was conducted as a prospective, double-blind, placebo-controlled clinical trial. Volunteers were screened for a negative history of gastrointestinal disease and the use of lactobacillus containing products within two months prior to the beginning of this study. Fifty-one volunteers were tested for intestinal *Candida* by a stool culture and 26 with positive cultures were selected for the study.

Twelve subjects were randomly assigned to the treatment group and fourteen to the control (placebo) group. The characteristics of the two groups are shown in *Table 1* including the means of ages Crook questionnaire scores, and stool culture categories. Chi-square analysis revealed no significant differences between the groups.

Cultures were grown on Bacto *Candida* BCG (Bromo Cresol Green) Agar Base at 30° CELSIUS for 7 days. Ten streaks were applied to each plate. *Candida albicans* species were differentially identified based on colony characteristics and the germ tube test. Colony counts were ranked by category: 0 = no colonies, +1 = 1 to 2 colonies, +3 = 7 to 10 colonies, +4 = 12 or more colonies. Fecal specimens were refrigerated prior to plating.

Subjects were additionally asked to complete a CROOK CANDIDA QUESTIONNAIRE at the beginning and end of the study. The Crook questionnaire is commonly used by physicians to screen patients for possible intestinal *Candida* overgrowth and symptomology. There is a strong correlation between the colony count categories and the pre-study questionnaire scores (*Table 1b*). This is consistent with the questionnaire's use for screening purposes. The correlation between Crook scores and the colony count was weaker at the post-study evaluation.

	Number	Age	Initial questionnaire	Initial stool culture
Treatment Group	12	31.92	99.17	2.4167
Control Group	14	32.07	115.36	2.7857

TABLE 1a. Initial questionnaire and stool culture values for treatment and control groups

Culture results	Questionnaire results (Mean values)
0	49.5
+1	67.0
+2	97.9
+3	113.3
+4	117.9

TABLE 1b. Comparison of stool culture results to Crook's Questionnaire

Lactobacillus acidophilus and the placebo were provided by a well-known manufacturer of the dietary supplement. *L. acidophilus* was suspended in a dry whey based at a concentration of at least 10^{10} cells per gram. The placebo consisted of a sterilized whey base. The *L. acidophilus* and placebo were identical in color, odor, size and texture. Both the *L. acidophilus* and the placebo were cultured to confirm these specifications.

The subjects were instructed to take 1 teaspoon twice daily in unchilled water, between meals, for a period of two months. Compliance with the treatment schedule was confirmed by interview. Stool samples were taken initially, monthly during the study, and one month post-study.

Statistical analysis

A chi-square analysis was done to determine the statistical significance of the treatment difference. Results which indicated a degree of significance of less than $p = .05$ were considered not significant.

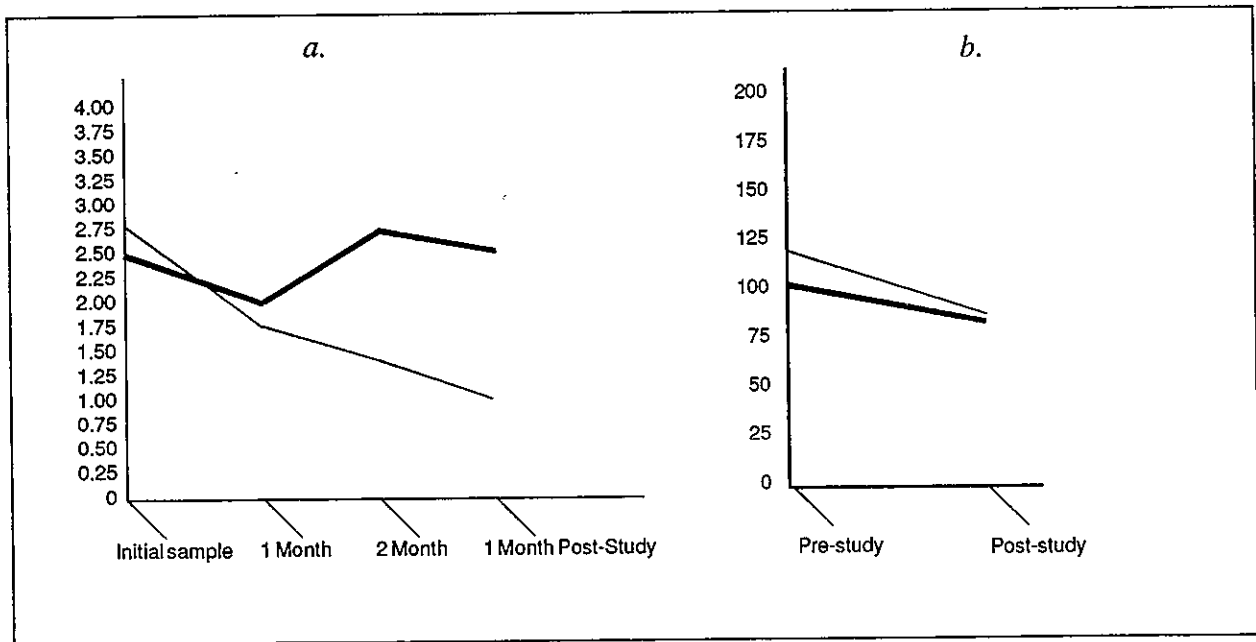


FIGURE 1. (a) Mean stool culture scores for control (light line) and treatment groups (heavy line). (b) Mean questionnaire scores for control (light line) and treatment groups (heavy line).

RESULTS

The results indicate no significant treatment effect (Figure 1 and Table 2). There was a decline in post-treatment questionnaire score that was similar in the treatment and control groups at 28.88 and 32.44 respectively.

DISCUSSION

The treatment protocol has several limitations which may have compromised the results. Future studies should take these limitations into consideration.

1. Treatment with acidophilus is commonly carried out with dietary changes. The subjects in this study were allowed a random diet which may have had an effect on the results. Dietary change in conjunction with *L. acidophilus* may have an effect which either alone does not have.
2. Assigning categorical values to the colony counts rather than using the actual counts may have hidden some differences. Actual counts should be used in future studies.

3. The questionnaire results of both groups improved approximately the same amount. It is well documented that retaking a test results in a learning curve which is reflected in the score. Since both groups' improvement was virtually identical, it could be hypothesized that the improvement is the result of the learning curve and not treatment.
4. Stool culture may be an unreliable method of determining intestinal candidiasis; however, these samples indicate that a stool culture does correlate to some degree with questionnaire scores.

If the mean results on the Crook questionnaire are compared to the stool culture results the following relationship is seen: as the stool culture value increases there is a corresponding increase in questionnaire results, and therefore in symptom reporting. However, the number of patients in each stool culture group is too small to be considered representative of the general population. A larger study could help elucidate this relationship. Studies examining the validity and reliability of the Crook instrument would be of value to physicians treating this syndrome.

Despite the above limitations it appears that the strain of *Lactobacillus acidophilus* used in this test has no effect on intestinal *Candida* as measured by stool cultures for *Candida* and the Crook questionnaire. It cannot be assumed that other strains will yield the same results; only future research can make this determination.

	Treatment	Control
Number of subjects	12	14
Initial stool culture	2.4167	2.7857
1st study culture	1.9167	1.7143
Number of subjects improved	7	8
Number of subjects worse	3	1
Number of subjects unchanged	2	5
2nd study culture	2.6667	1.5714
Number of subjects improved	4	9
Number of subjects worse	6	0
Number of subjects unchanged	2	5
Post study culture	2.5000	1.2143
Initial questionnaire	99.17	115.36
Final questionnaire	70.29	82.92
Difference	28.88	32.44

TABLE 2. Comparison of mean stool culture and questionnaire results of treatment and control groups.

CONCLUSION

The strain of *Lactobacillus acidophilus* employed in this study, when used in a population of subjects with positive stool cultures for *Candida albicans*, has no effect on future stool samples during treatment for a period of 2 months. In addition, though correlating with the stool culture values, the results of the Crook questionnaire did not improve significantly over a placebo group.

REFERENCES

1. Shahani KM, Ayebo AD. The role of dietary Lactobacilli in gastro intestinal microecology. *Am. J. Clin. Nutr.* 1980; 33:2448-2457.
2. Collins EB, Hardt P. Inhibition of *Candida Albicans* by *Lactobacillus Acidophilus*. *J. Dairy Sci.* 1980; 63:830-2.
3. Shahani KM, Friend BA. Nutritional and therapeutic aspects of Lactobacilli. *J. Appl. Nutr.* 1984; 36:125-52.
4. Truss OK. Tissue injury induced by *Candida Albicans*. *J. Ortho. Psych.* 1978; 7(1):17-37.
5. Truss OK. The role of *Candida Albicans* in human illness. *J. Ortho. Psych.* 1981; 10(4):228-238.