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Are Probiotics Safe and Effective in Reducing the Incidence of Diarrhea in HIV-positive Individuals?

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ABSTRACT

Objective: The objective of this selective EBM review is to determine whether or not probiotics are safe and effective in reducing the incidence of diarrhea in HIV-positive individuals.

Study Design: Systematic review of three English language primary studies, published in 1998, 2007, and 2008.

Data Sources: Three double-blind, randomized, controlled trials published after 1996, comparing administration of probiotics with placebo, were obtained using EBSCOhost, PubMed, and Cochrane databases.

Outcomes Measured: Frequency and consistency of stools. Daily or weekly questionnaires allowed subjects to report perceived severity of symptoms using scales containing descriptive words and figures with corresponding numeric values. Subjects were evaluated initially at baseline and subsequently at the completion of the studies, and the results were compared.

Results: No intervention-related negative side effects or adverse events were reported in any of the studies. Anukam and colleagues found that those women treated with probiotics had rapid resolution of diarrhea compared to controls, for both the duration of treatment at day fifteen and at follow-up on day ninety. Trois, et al. reports a decrease in loose stools and an increase in normal stools in both groups but no statistically significant decrease in diarrhea in the probiotic group as compared with control.

Conclusion: All three studies showed that probiotics are safe when used in children and adults with HIV/AIDS. Simple comparison of the studies performed by Trois, et al. and Anukam, et al. suggests that probiotic supplements are more efficacious in adult populations than pediatric populations in reducing frequency of loose stools. However, the use of anti-retroviral drugs, geographic location, probiotic bacterial strain, and the cause of diarrhea in these populations may have impacted the outcomes. More studies containing larger patient sample sizes and including those in various other countries should be performed to further determine the efficacy of probiotics in reducing diarrhea and appreciate other health benefits in diverse populations of HIV-positive individuals.

Key words: probiotics, safety, diarrhea, gastrointestinal, children, adults, HIV, AIDS

INTRODUCTION

Diarrhea, in both acute and chronic forms, is one of the most common symptoms experienced by men, women, and children who have been infected with the human immunodeficiency virus (HIV).[1] In its earliest days, HIV/AIDS was described as the "slim disease" due to advanced wasting and persistent diarrhea, which is still a reality in Africa and other disadvantaged parts of the world. Diarrhea also contributes as a major cause of morbidity and mortality for HIV-positive children across the globe.[2] Chronic diarrhea is often defined as three or more loose or liquid stools per day for more than four weeks and/or a daily stool weight greater than 200 g/day. While acute diarrhea is associated with more pain, blood stream invasion, and death for HIV patients, both acute and chronic forms of diarrhea threaten overall health and quality of life.[1]

Despite increased research efforts and education aimed at eradicating HIV-transmission, 2.6 million new infections are reported worldwide yearly, with 50,000 of these occurring in the United States.[3] The care of HIV-positive individuals in the US is of great cost; for example, Farquhar, et al. estimated $18.2 billion in HIV-associated economic losses in 2001 alone.[4] One study in Birmingham, Alabama cites costs between $13,885 and $36,532 per HIV patients per year.[5]   The chief expenses are generated by medication costs (71-84%) and care for those with more advanced disease.

Physician Assistants (PAs) are often the sole care provider for HIV-patients. As of 2005, PAs and Nurse Practitioners (NPs) provided most of the care for 20% of CARE Act Title III HIV clinics. PAs and NPs performed similarly to those physicians trained in infectious disease and superiorly to generalist non-HIV experts in 6 of 8 quality measures.[6] Therefore, PAs play an

important role in the management of HIV in the US and particularly in maintaining quality and specificity of care for these patients.

HIV-related diarrhea is multi-factorial in nature. Infectious and non-infectious causes prevail, yet all physiological aspects are not fully understood. While they are found in the serum, the majority of CD4+ T-lymphocytes reside in the mucosal surfaces, particularly the gut. The intestinal mucosa acts a main target for HIV as well as a "reservoir" for viral replication, especially in early disease, leading to immune dysregulation through increased apoptosis of CD4+ lymphocytes.[1] Bacterial overgrowth, opportunistic infection, and malabsorption of nutrients can also result. Numerous infectious agents are often isolated in stool cultures, including bacterial species (*E. coli*, *Salmonella*, *Shigella, C. difficile, Mycobacterium*, *Campylobacter)*, viruses (Rotavirus, adenovirus, Cytomegalovirus), protozoan parasites (*Giardia lamblia, Entamoeba histolytica, Cryptosporidium parvum,*), and fungi (Candida, Histoplasmosis).[1]

Even in the absence of infection, the HIV virus itself causes "profound changes" in the intestinal mucosa.[1] Increased permeability allows for translocation of microbial products across the gut, which may stimulate innate immune receptors. Villous atrophy and crypt hyperplasia of the jejunum are common findings, termed, "HIV enteropathy." Decreased absorption of sugars, vitamin B12, and other nutrients are linked with chronic diarrhea in HIV patients. Malabsorption is known to directly affect the CD4 count in HIV children adversely.[2] The virus is also associated with inflammatory bowel disease, pancreatic disease (impaired fat absorption), and lymphomas including Hodgkin and Non-Hodgkin B-cell, diffuse large B-cell, primary effusion lymphoma, Burkitt and Burkitt-like lymphomas, and Kaposi sarcoma, all of which have the

potential for gastrointestinal (GI) symptoms.[1] Additionally, diarrhea is one of the most common side effects of most antiretroviral therapies (ARTs).[7]

No specific treatment or drug has been approved for HIV-related diarrhea. The main goals of treatment are to eradicate pathogenic causes, reduce the number of loose stools per day, manage dehydration, decrease abdominal discomfort, and improve quality of life.[7] Antibiotics, anti-virals, and anti-fungals should be given based on the sensitivity of suspected or isolated pathogens. While ARTs have helped to decrease chronic diarrhea in Western populations, practitioners have commonly prescribed antimotility agents, such as loperamide, diphenoxylate, and codeine, or adsorbents like bismuthsabsalicylate, kaolin/pectin, and attapulgite to control symptoms. Cholestyramine (for malabsorption of bile salts) and thalidomide (for HIV-related colitis) have also been employed.   Supplementation with vitamin A and zinc have been shown to reduce gut permeability.[1]

Probiotics are defined by the Food and Agricultural Organization (FAO) as "live micro-organisms which when administered in adequate amounts confer a health benefit on the host"[8]. Supplementation has been shown to enhance mucosal immune defense by supporting macrophages, killer cells, T cells and interferon, and acting against pathogenic colonization and translocation.[2] Their use has been well documented in the alleviation of diarrhea (*Lactobacillus sp)* and other unwanted GI symptoms. Probiotics, *L. reuteri* in particular, have been beneficial in prophylaxis against cryptosporidiosis, a common pathogen in the HIV population.[8] The probiotic organisms do not remain for extended periods of time in the GI tract, so sufficient quantities ($>1 \times 10^7$/day) are necessary to maintain colonization and immune stimulation.[2]

OBJECTIVE

The objective of this selective EBM review is to determine whether or not probiotics are safe and effective in reducing the incidence of diarrhea in HIV-positive individuals.

METHODS

A number of criteria were considered in selection of studies. The population was limited to HIV-positive individuals two years old or greater who experience diarrhea. Only double blind or double-masked, randomized, controlled trials that introduced probiotic supplementation as compared with visually-matched placebo were considered. The following strains were deemed acceptable probiotics: *Lactobacillus rhamnosus, Streptococcus thermophilus, Bifidobacterium bifidum*, and *Lactobacillus reuteri*. Outcomes of interest included tolerance, adverse events, and consistency and frequency of stools.

A detailed search was performed by the author of this review between January 2011 and February 2011 using the Cochrane Library EBM Online Database and PubMed Online, with the key words "HIV," "diarrhea," and "probiotics." All three articles that were selected to be included in this review were written in the English language and published in peer-reviewed journals between 1996 and present. Selection of studies, which was also performed by the author, was based on relevance to the topic of interest, and only studies that reported patient-oriented outcomes (Patient Oriented Evidence that Matters, or POEMs) were accepted. Inclusion criteria simply included HIV-positive status. Exclusion criteria included individuals with other chronic diseases, known food or drug allergies, pregnancy, recent (<1 month) changes in ART or changes during the study. The studies either reported p-values or contained dichotomous data, which could be used to calculate Relative Risk Reduction/Increase (RRR/RRI), Absolute Risk Reduction/Increase (ARR/ARI), and Numbers Needed to Treat/Harm (NNT/NNH).

Demographics of the three final selections are reported and displayed in Table 1.

Table 1. Demographics and characteristics of included studies

Study	Type	# of patients	Age	Inclusion Criteria	Exclusion Criteria	W/D	Intervention
Anukam, 2008[9]	Double-blind, RCT	24	18-44	HIV (+) females age 18-44, CD4>200 cells/mcL	Lactose intolerance, currently taking supplements or ARTs	2	100mL conventional yogurt/day fermented with *Lactobacillus delbruekii* subsp. *Bulgaris* and *Streptococcus thermophilus* supplemented with *L. rhamnosus* GR-1 and *L. reuteri* RC-14 at 10^7/mL
Trois, 2008[2]	Double-blind, RCT	77	2-12	HIV (+) children who acquired the disease perinatally, one child who acquired by blood products	Chronic diseases or allergies; Changes in ART in past 3 months or during the study	1	14g/day of a standard formula containing *Bifidobacterium bifidum* and *Streptococcus thermophilus* at 2.5×10^{10} CFU diluted in 100 mL of milk
Wolf, 1998[10]	Double-blind, RCT	39	18-65	HIV(+) males and non-pregnant females (at least 6 weeks post-partum); CD4 count >400	Known drug or food allergy; Individuals taking any ART other than Zidovudine	4	10^{10} CFU *Lactobacillus reuteri* (strain SD2112)/day in a beverage <37°C [ex: tap water, milk, orange juice from concentrate, apple juice, grape juice, cranberry juice, 7-up]

OUTCOMES

All three studies ascertained POEMs by gathering reports from participants. Wolf, et al. were concerned mainly with safety and tolerance of probiotics. A review of systems was taken at baseline for comparison of the efficacy of implemented therapy. By completing daily questionnaires, subjects reported the severity of diarrhea using the following scale: 0 = absent, 1= mild, 2 = moderate, and 3 = severe; subjects also reported the number of bowel movements daily and the consistency using the following scale: 1 = hard, dry; 2 = hard, formed; 3 = soft, formed; 4 = soft, unformed; 5 = watery.[10] Trois, et al. also used previously approved reference questionnaires, which were completed by the subjects' parents, to determine the frequency and consistency of stools in each 24-hour period. Drawings depicting stools varied from watery to normal and had corresponding numeric scores.[2] Anukam, et al. developed a structured questionnaire given at baseline, fifteen days, thirty days, and three months. Participants rated diarrhea episodes as absent, moderate (daily episodes of watery stools), or severe (persistent watery stools, urgency and frequency of defecation).[9]

RESULTS

Safety. Results of all three studies support the safety of using various probiotics in HIV patients. Wolfe, et al. found no statistically significant difference ($p<0.05$) in the frequency and consistency of stools between the control group and those receiving 10^{10} colony-forming units (CFU) of *L. reuteri* per day during any week of the study. Baseline values were not reported, and results were presented based on percent of subject days. Using this method, most GI tolerance factors were absent for the majority (>80%) of study days (See Table 2). No p-values were reported for these. Both groups reported a greater amount of flatulence, but those receiving probiotics experienced less. A potential side effect, mild or moderate nausea, was reported by

18.26% of the probiotic group, as compared with only 6.72% of the control group. However,

those taking probiotics reported no significant vomiting or reflux. The experimental group also

reported slightly higher, but not statistically significant, incidence of constipation and distention;

these were still absent 93.31% and 86.08% of the time, respectively. All data was continuous,

rather than dichotomous, and no baseline values were reported for comparison or for further

calculations. Results are reported in Table 3. Anukam, et al. saw no adverse effects in the

probiotic group, including any significant change in key lab values—total WBCs, neutrophils,

lymphocytes, RBCs, hemoglobin, hematocrit, or platelet count (see Table 4)—while three in the

control group experienced rashes during the study. Trois et al. did not report any adverse

reactions in the children studied.

Table 2. GI tolerance factors of *L. reuteri* reported by Wolfe, et al., 1998[10]

Parameter	Treatment	Absent	Mild	Moderate	Severe
Flatulence	Placebo	62.89*	23.67	12.89	0.56
	L. reuteri	67.45	19.53	9.4	3.62
Nausea	Placebo	93.14	5.46	1.26	0.14
	L. reuteri	81.56	14.1	4.16	0.18
Vomiting	Placebo	99.58	0.28	0.14	0.0
	L. reuteri	97.47	1.99	0.54	0.0
Reflux	Placebo	95.66	3.78	0.42	0.0
	L. reuteri	94.21	4.88	0.9	0.14
Constipation	Placebo	97.06	1.82	1.12	0.0
	L. reuteri	93.31	3.62	1.27	1.81
Distention	Placebo	91.32	3.92	4.62	0.14
	L. reuteri	86.08	7.96	2.71	3.25

*Results reported in percentage of subject-days

Table 3. Safety and Tolerance of *L. reuteri* reported by Wolfe, et al.,1998[10]

	Week 1	Week 2	Week 3	Week 4	Week 5
Frequency	NS[a]	NS	NS	NS	NS
Consistency[b]	NS	NS	NS	NS	NS

[a]*P*-values represent the level of significance for each week comparing placebo with *L. reuteri*;
NS = not significant ($p > 0.05$).
[b]Fecal consistency score: 1 = hard, dry; 2 = hard, formed; 3 = soft, formed; r = soft, unformed; 5
= watery.

Efficacy. The success of probiotic supplementation in reducing diarrhea varied

significantly between the trials. Anukam, et al. studied adult females living in Nigeria and used

conventional yogurt fermented with *Lactobacillus delbruekii* subsp. *bulgaris* and *Streptococcus*

thermophilus and supplemented with 2.5×10^9 CFU/day of *L. rhamnosus*, GR-1 and *L. reuteri*

RC-14. All participants admitted to moderate diarrhea at baseline. The authors saw a "complete

resolution" of diarrhea in 12/12 women in the experimental group by the second day of the

study, which lasted for the remainder of the treatment period (day 15), as compared with only

one control group participant. At the ninety-day follow-up, 8/12 that received the probiotics still

reported a total absence of diarrhea, while only 2/12 of those in the control group reported its

absence. Risk and treatment statistics are summarized in Table 4.

Table 4. Diarrhea reduction with *L. reuteri* and *L. rhamnosus* reported by Anukam, et al., 2008[9]

	RBI[a]	ABI[a]	NNT [a]	Adverse Events[b]
Day 2 of treatment	[c]	100%	1 subject	NR, NS[d]
Day 15 (full duration of treatment)	10.1	91%	2 subjects	NR, NS
Day 90 (post-treatment follow-up)	2.99	50%	3 subjects	NR, NS

[a]RBI = Relative benefit increase; ABI = Absolute benefit increase; NNT = Numbers needed to
treat
[b]Adverse events in those receiving probiotics; NR = none reported;
[C]Could not be calculated
[d]NS = not significant; pertaining to p-values (all >0.05) comparing laboratory values at baseline
and day 30 for the following hematologic parameters: TWBC, neotrophils, lymphocytes, RBCs,
HB, HCT, MCV, MCH, MCHC, RDW, and platelets

Trois, et al. did not see the same overwhelming efficacy with the children studied in

Brazil. Those in the experimental group received 2.5×10^{10} CFU of *Bifidobacterium bifidum* and

Streptococcus thermophilus. Both groups experienced a statistically significant ($p<0.05$)

decrease in number of liquid stools and an increase number of in normal stools. Notably, no

statistical difference existed between the groups. Results of this study are presented in Table 5.

Children under age 2 or greater than 12 years were excluded from participating in this study.

Only one subject in the probiotics group did not adhere to the treatment regimen yet was still

included in the results due to his or her intention to be treated. One subject in the control group

became hospitalized and was removed entirely from the study. Overall, the adults who received

L. rhamnosus and *L. reuteri* experienced greater resolution of diarrhea than did the children who

received *B. bifidum* and *S. thermophilus.*

Table 5. Changes in stool with *B. bifidum* and *S. thermophilus* reported by Trois, et al., 2007[2]

Outcome measured	*p* from baseline[a]	Significance	*p* between groups	Significance[b]
Decrease in # if liquid stools	0.006	Significant	0.522	NS[c]
Decrease in # of loose-soft stools	0.955	NS	Not reported	
Increase in # of normal stools	0.001	Significant	0.199	NS

[a]p-values from baseline were reported to be statistically the same in both placebo and group
receiving *Bifidobacterium bifidum* and *Streptococcus thermophilus* when compared with
respective baseline values.
[b]Statistical significance of p-value comparison between treatment groups
[c]NS = Not significant; a p-value is deemed significant if <0.05.

DISCUSSION

The author was unable to find another systematic review written to date concerning the

use of probiotics to alleviate HIV-related diarrhea. The study performed by Wolfe, et al. was the

pioneer trial to test and prove the safety of probiotics in HIV-positive patients. The study by

Anukam, *et al*. showed great reduction of diarrhea but may be limited by a sample size of only

twelve subjects in each group, experimental and control, who were not using any ART. The

subjects in Trois, *et al.* were using ART, which is notorious for producing the adverse effect of

diarrhea. This difference could contribute to the discrepancy in results. Anukam, et al. only

provided fifteen days of treatment, while Trois provided 60 days. It has been noted that the

effects of probiotics differ and are unique to each strain.[8] The women in Nigeria were given *L.*

rhamnosus and *L. reuteri*, while the children in Brazil were given *B. bifidum* and *S.*

thermophilus. These differences could also be contributing to such varied results. The nature of

the diarrhea in each subject was not ascertained before beginning the studies, so the exact

mechanisms by which the probiotics were functioning in alleviating or not alleviating diarrhea could not be determined for certain.

Supplementation with probiotics continues to be an area of intense research for HIV and many other conditions. Multiple studies have shown that probiotics also effectively increase the CD4 count in people with HIV/AIDS.[9,11] Strong evidence exists to support benefits against acute diarrhea from rotavirus and in preventing surgical complications like pouchitis. Other areas that have been explored are prevention of necrotizing enterocolitis (NEC), traveler's diarrhea, *H. pylori* infection, the control of irritable bowel syndrome (IBS), inflammatory bowel disease (IBD), reducing the risk of bladder and colon cancer, vulvovaginal candidiasis and other urogenital problems, respiratory infections, atopic conditions, and dental caries.[8,12]

The most common side effects noted across studies have been flatulence and bloating, which are not considered dangerous and are usually mild.[8] Wolfe, et al. noted both of these to be present in subjects of that study.[10] While they have never been proven in a clinical trial, the most serious potential adverse events are bacteremia and fungemia, which have been documented in the past as isolated cases. These are extremely rare, estimated 1 in 1 million and 1 users in 1.69 million users, respectively; yet the severely immunocompromised are considered among those at highest risk.[8] Probiotic use has also been associated with increased mortality in patients with severe acute pancreatitis. *Lactobacillus* preparations are contraindicated for those with a hypersensitivity to lactose or milk, and *S. boulardii* for patients with yeast allergies. Currently, no contraindications for *Bifidobacteria* exist. Probiotics are available as supplements (i.e., tablets, capsules, or powders) or as fermented dairy products (i.e., yogurt and milk) and have not yet been evaluated by the FDA for the creation of specific recommendations or regulations.[8]

CONCLUSION

The articles reviewed demonstrate that probiotic supplementation is safe in HIV patients, but the evidence is conflicting regarding its efficacy in reducing the incidence of diarrhea in this population.   No adverse events were attributed to probiotic use. The evidence provided by Aunkam, et al. is promising and is reason enough to pursue the intervention further. The Food and Agriculture Organization (FAO) and World Health Organization (WHO) published a 2001 Report on Probiotics that recommended "efforts be made to take probiotics to developing countries."[13] Motivated by the work of Anukam, et al., a group called Western Heads East (WHE) has created a community kitchen in Mwanza, Tanzania for producing a probiotic yogurt named Fiti, which uses *L. rhamnosus* GR-1. Trials have been completed but not yet published, which showed fewer episodes of diarrhea in HIV-positive individuals who received Fiti yogurt (p<0.01 in one trial and p=0.05 in another).[13] Future trial designs should include control groups of adults who are using ART to determine the benefits to this segment of the HIV population, as ART is becoming more available. Because probiotics are relatively inexpensive, widely available, and generally safe, trials such as these should continue, especially in children and adults of impoverished areas around the globe, in order to aid these individuals and to expand what is known about the various benefits of probiotic supplementation.

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