The Immunomodulatory Nutritional Intervention NR100157 Reduced CD4$^+$ T-Cell Decline and Immune Activation: A 1-Year Multicenter Randomized Controlled Double-Blind Trial in HIV-Infected Persons Not Receiving Antiretroviral Therapy (The BITE Study)

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Background. The immunomodulatory nutritional product NR100157 was developed for human immunodeficiency virus (HIV)–infected individuals. We hypothesized that targeting the compromised gastrointestinal tract of HIV-infected individuals would result in systemic immunological benefits.

Methods. In a multicenter, randomized, controlled, double-blind trial, 340 HIV-1–positive adults not on antiretroviral therapy, with CD4$^+$ T-cell counts <800/μL, were given either NR100157 or an isocaloric and isonitrogenous control for 52 weeks. Primary outcome was CD4$^+$ T-cell count. Secondary outcomes included plasma viral load (pVL), safety, and tolerability. In a pilot study (n = 20), levels of CD4$^+$CD25$^+$ and CD8$^+$CD38$^+$ activation were measured (n = 20). The trial is registered at the Dutch Trial Register (NTR886) and ISRCTN81868024.

Results. At 52 weeks, CD4$^+$ T-cell decline showed a 40-cell/μL difference ($P = .03$) in the intention-to-treat population in favor of the immunomodulatory NR100157 (control vs active, −68 ± 15 vs −28 ± 16 cells/μL/year). The change in pVL from baseline was similar between groups ($P = .81$). In the pilot study, the percentage of CD4$^+$CD25$^+$ was lower in the active group ($P < .05$) and correlated with changes in CD4$^+$ T-cell count ($r = -0.55$, $P < .05$). The percentage of CD8$^+$CD38$^+$ levels was unaffected.

Conclusions. The specific immunonutritional product NR100157 significantly reduces CD4$^+$ decline in HIV-1–infected individuals, and this is associated with decreased levels of CD4$^+$CD25$^+$. (This nutritional intervention is likely to affect local gut integrity and gut-associated lymphoid tissue homeostasis, which in turn translates positively to systemic effects.)

Clinical Trials Registration. ISRCTN81868024.

Keywords. immunonutrition; NR100157; immune activation; CD4 decline.

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Loss of plasma $CD4^+$ T cells [1] is a hallmark of human immunodeficiency virus (HIV) infection. Residual immune activation in relation to $CD4^+$ T-cell depletion in HIV-infected persons is increasingly recognized [2, 3]. HIV infection is associated with a rapid loss of gut-associated $CD4^+$ T cells, impaired function of epithelial cells, and alterations in the composition of the gut microbiota, leading to a loss of gastrointestinal (GI) homeostasis and barrier function [4, 5]. Residual immune activation persists even in patients on stable antiretroviral (ARV) treatment, and this is increasingly linked to non-AIDS events such as cardiovascular disease and liver and kidney toxicities [6]. Moreover, the efficacy of ARV treatment in the GI tract appears less effective, as shown by the incomplete restoration of $CD4^+$ T-cell counts [7, 8] and incomplete viral suppression, leading the GI tract to function as a viral reservoir with low levels of ongoing viral replication [7, 9]. As a consequence of a compromised gut, bacteria or bacterial products can translocate and enter the circulation, and these have been suggested to contribute to immune activation. In HIV-1–infected individuals, increased markers of bacterial translocation have been reported and have been correlated with parameters of immune activation and disease progression [10]. In patients on highly active ARV therapy (HAART), markers of bacterial translocation also correlate with immune activation and lower levels of $CD4^+$ T-cell restoration [11]. These data support the hypothesis that the GI tract plays a major role in HIV-induced chronic immune activation [12]. Immune activation in HIV-infected patients has been reported to be a stronger predictor of disease progression than viral load or $CD4^+$ count [13].

Disturbance of intestinal microbiota composition occurs early after HIV infection [4], and intervention with prebiotic oligosaccharides can help normalize microbiota. The components in the immunomodulatory nutritional product NR100157 are listed in Table 1. The specific oligosaccharides in NR100157 have been shown to enhance T-helper 1–dependent immune responses and increase resistance to infection in a murine influenza vaccination model and in infants [14, 15] and to reduce markers of immune activation in HIV-1–infected patients [16]. A higher inflammatory state of the gut mucosa has been found in many infected with HIV, with evidence that intestinal inflammation is a direct consequence of HIV infection [17, 18]. Long-chain n-3 and n-6 polyunsaturated fatty acids induce anti-inflammatory effects by reducing the production of arachidonic acid–derived inflammatory mediators [19, 20]. Substantial data demonstrate that intestinal integrity is compromised in HIV-1 infection, and bovine colostrum has reportedly aided in decreasing permeability [21]. Glutathione depletion is observed in HIV-infected individuals and is associated with impaired survival [22] as well as with perturbations of immune responses [23].

Taking this evidence into account, together with the fact that HIV may initiate a complex dysregulation of metabolism associated with changes in nutritional status, lipid metabolism, and immune function, NR100157 was designed to target the gut. It was hypothesized that NR100157 could beneficially affect the health status of HIV-infected individuals by supporting gut integrity and immune function because beneficial effects in the gut would translate to preservation of plasma $CD4^+$ count. The target population was HIV-1–infected patients not on ARV treatment.

Table 1. Product Composition: Intake Per Day (2 Doses)

Ingredient	Active	Control
Energy	314 kcal	314 kcal
Total lipids	7.4 g	7.4 g
EPA	1.24 g	. . .
DHA	0.65 g	. . .
GLA	0.49 g	. . .
Total protein	16.4 g	16.4 g
Bovine colostrum protein	12.5 g	. . .
Cysteine from (1.8 g) NAC	1.3 g	. . .
Cysteine (total)	1.6 g	0.1 g
Total carbohydrates	40.2 g	45.8 g
Lactose	6.9 g	6.9 g
Glucose	0.1 g	0.3 g
Oligosaccharides	15.0 g	. . .
Short-chain GOS	6.75 g	. . .
Long-chain FOS (inulin)	0.75 g	. . .
Pectin-derived AOS	7.50 g	. . .

Abbreviations: AOS, acidic oligosaccharides; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; FOS, fructo oligosaccharides; GLA, γ-linolenic acid; GOS, galacto oligosaccharides; NAC, N-acetyl cysteine.

METHODS

Study Design

A multicenter, multinational, randomized, controlled, double-blind trial (ISRCTN81868024; BITE), with 52 weeks of intervention, was conducted between March 2007 and July 2009. Eligible participants were HIV-1–infected adults (aged >18 years) not on ARV therapy who were enrolled from 30 sites in 8 countries worldwide: Italy (3 sites), the Netherlands (4 sites), the United Kingdom (3 sites), Thailand (1 site), United States (6 sites), Brazil (4 sites), Argentina (4 sites), and Australia (5 sites). The protocol included an interim analysis after 340 randomized participants completed the 52-week supplementation period. Protocol and consent forms were approved by the ethics review committee at each site. A pilot study of 20 patients was performed at 1 site to study $CD4^+$ and $CD8^+$ T-cell activation. Written informed consent was obtained from all individuals before study participation.

The time to ART initiation for the participants who dropped out of the study due to commencement of therapy was plotted retrospectively.

Study Outcomes

The primary outcome of the trial was the change from baseline in plasma $CD4^+$ T-cell count. Plasma concentrations of HIV-1 RNA, $CD8^+$ cell count, % $CD4^+$, % $CD8^+$, and the $CD4^+/CD8^+$ ratio were assessed as secondary parameters at baseline and at weeks 13, 26, 39, and 52. All laboratory and clinical assessments were performed blinded at the participating centers. $CD4^+$ and $CD8^+$ T-cell counts were performed using flow cytometry. Plasma HIV-1 RNA levels were quantified using the Amplicor HIV-1 monitor test, NucliSens HIV-1 QT, or Quantiplex HIV-1 RNA. Adherence was assessed using patient diaries that were checked against the returned sachets. GI tolerance was assessed using a questionnaire and scoring the severity of GI symptoms within a 4-point scale.

Statistical Analyses

The power calculation was based on published longitudinal $CD4^+$ T-cell data, predicting a decline of 50–70 cells/μL per year [1]. Combined with an anticipated dropout rate of 25%, a power of 80%, and an alpha of 0.05, this led to an initial sample size calculation of 800 patients. A protocol-planned interim analysis was performed when full data would become available for 340 participants. An independent data and safety monitoring board (DSMB) reviewed the results of the blinded interim analysis. In accordance with predefined stopping rules, the trial was stopped and arms unblinded due to high rates of loss to follow-up. All data presented in this paper, including graphs and tables, and all statistical analyses were performed on the intention-to-treat (ITT) population, unless stated otherwise. Logarithmic transformation was used to normalize the distribution of $CD4^+$ T-cell count, viral load, % $CD4^+$ T-cell count, aspartate aminotransferase (AST), alanine aminotransferase (ALT), creatinine, prothrombin time, hemoglobin, white blood cell count, lymphocytes, and neutrophils.

To analyze efficacy and safety, differences between the 2 study arms were compared using repeated measures mixed models. The SPSS statement REPEATED was used to model the covariance among the repeated measures obtained on the same participants with the structure compound symmetry. Study site was entered as a random effect (statement RANDOM) with a covariance structure identity. In these models, a response profile analysis was used to describe the mean response over time, allowing a study arm comparison at each measurement occasion. In addition, the mean response over time was modeled with a linear trend (first-order polynomial). The significance of the study arm by time coefficient indicates the overall intervention effect that can be interpreted as a difference in slopes.

GI tolerability questionnaires were analyzed using cross table statistics, comparing the 2 groups. Values are reported as estimated marginal mean ± SE, unless stated otherwise. All analyses were performed using SPSS software, version 15.0.1 or higher, and statistical significance was reached when $P < .05$.

Role of the Funding Source

Nutricia Advanced Medical Nutrition designed the study and collected and analyzed the data. Local investigators were involved in sample collection. The study report was written by the study sponsor. The publication coauthors made the decision to submit the paper for publication. The corresponding author confirms that he had full access to all data in the study and had final responsibility for the decision to submit for publication.

RESULTS

Study Population

Of the 409 participants who were screened in the study, 69 were not eligible and 340 underwent randomization at 1 of the 30 sites. A total of 143 participants completed the full 1-year study on treatment, 83 in the control and 60 in the active arm (Figure 1). The baseline characteristics were not significantly different between groups (Table 2). The average compliance to the product was 85% and similar between active and control groups (85% and 86%, respectively).

$CD4^+$ T-Cell Count Decline

A significant decline in $CD4^+$ T-cell count was observed in the NR100157 intervention group: the baseline-corrected decline in $CD4^+$ T-cell count was 28 ± 16 cells/μL in the active group compared with 68 ± 15 cells/μL in the control group at 52 weeks (Figure 2; $P = .030$), showing a difference of 40 cells/μL in favor of the NR100157 group. A similar difference was observed when the slopes of $CD4^+$ T-cell decline over 52 weeks were compared between the study groups (steeper slope of decline in the control group compared with the active group, $P = .016$) or when the baseline-corrected decline in $CD4^+$ T-cell count over 52 weeks was analyzed in the group of participants who completed the study (−18 ± 20 cells/μL in the active group, −68 ± 17 cells/μL the control group; a difference of 50 cells/μL, $P = .014$).

HIV-1 RNA and $CD4^+/CD8^+$ Ratio and Percentages

Secondary outcomes did not differ significantly between the study groups (see Supplementary Table 1). HIV-1 RNA levels remained stable during the study; the change from baseline

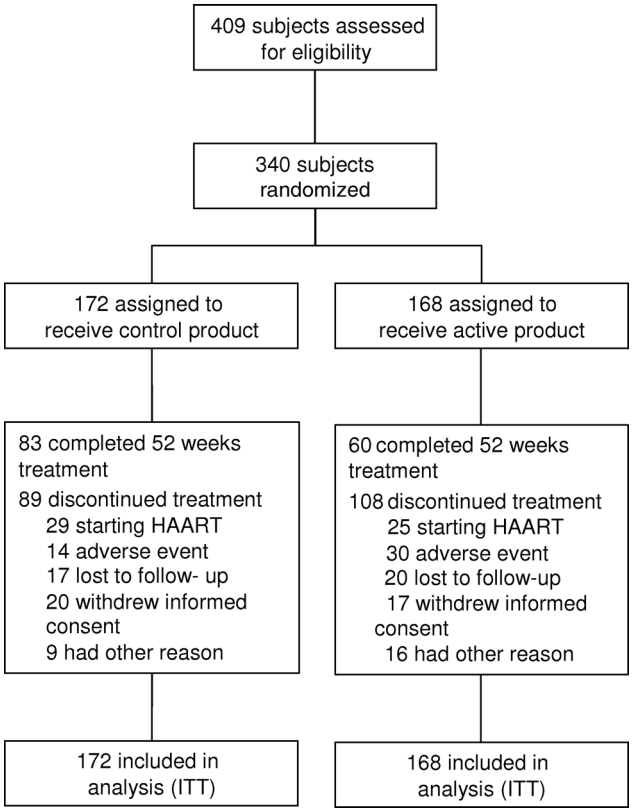

Figure 1. Trial profile, screening, randomization, and study completion. Abbreviations: HAART, highly active antiretroviral therapy; ITT, intention-to-treat.

was -0.01 ± 0.07 $\log_{10}$ RNA copies/mL in the active group compared with -0.03 ± 0.07 $\log_{10}$ RNA copies/mL in the control group at 52 weeks ($P = .811$; see Supplementary Figure 1). The baseline-corrected $CD8^+$ cell count was higher in the active group compared with the control group at 26 weeks ($P = .050$) but not at 39 weeks ($P = .097$) or at 52 weeks ($P = .477$). Furthermore, % $CD4^+$, % $CD8^+$, and % $CD4^+$/% $CD8^+$ ratio did not differ between study groups at the different timepoints (see Supplementary Table 1).

$CD4^+$ and $CD8^+$ T-Cell Activation

A total of 20 individuals (n = 10 per group) were analyzed in a pilot study for the expression of T-cell activation markers. Baseline characteristics did not differ between the groups and were comparable to the characteristics of the total study population (see Supplementary Table 2). Three participants dropped out of the study due to the start of ARV treatment, 1 in the control (>24 weeks) and 2 in the active group (>39 weeks). The effect of the intervention showed trends that were similar to those seen in the total study population: the decline in $CD4^+$ T-cell count tended to be less in the active group

Table 2. Patient Baseline Characteristics of Study Groups (Mean ± SD or n [%]); Intention-to-Treat

Variable (Unit)	Control Group (n = 172)	Active Group (n = 168)
Sex (n [%]) Male	140 (81%)	139 (83%)
Female	32 (19%)	29 (17%)
Age (y)	40.1 ± 9.7	39.2 ± 9.3
Body mass index (kg/m^2)	24.6 ± 3.4	24.4 ± 4.0
Duration of HIV-1 infection from diagnosis (d)	424 ± 122	413 ± 110
Race: White	142	128
Asian	13	17
Hispanic	9	9
African	3	5
Other	5	9
$CD4^+$ T-cell count (cells/μL)	413 ± 136	425 ± 154
$CD8^+$ T-cell count (cells/μL)	1028 ± 413	1022 ± 419
% $CD4^+$ T cells	23.2 ± 6.8	23.4 ± 7.3
% $CD8^+$ T cells	54.2 ± 9.0	54.1 ± 10.9
Ratio % $CD4^+$/% $CD8^+$	0.45 ± 0.20	0.47 ± 0.22
Plasma HIV-1 RNA ($\log_{10}$ copies/mL)	4.53 ± 0.64	4.47 ± 0.60
Medical history (n [%])		
HBV	10 (6)	8 (5)
HCV	6 (3)	5 (3)
HBV/HCV status unknown	1 (1)	0 (0)
CDC classification (n [%])		
Class A (asymptomatic)	151 (88)	149 (89)
A1 (CD4 ≥ 500 cells/mm^3)	43 (25)	54 (32)
A2 (CD4 200–499 cells/mm^3)	105 (61)	89 (53)
A3 (CD4 < 200 cells/mm^3)	3 (2)	6 (4)
Class B (symptomatic, not A or C)	19 (11)	18 (11)
B1 (CD4 ≥ 500 cells/mm^3)	1 (1)	3 (2)
B2 (CD4 200–499 cells/mm^3)	18 (11)	14 (8)
B3 (CD4 < 200 cells/mm^3)	0	1 (1)
Class C (AIDS indicator)	2 (1)	1 (1)
C2 (CD4 200–499 cells/mm^3)	2 (1)	1 (1)

Abbreviations: CDC, Centers for Disease Control and Prevention; HBV, hepatitis B virus; HCV, hepatitis C virus; HIV-1, human immunodeficiency virus type 1.

(difference at 52 weeks of 77 cells/μL; $P = .107$), and the plasma viral load did not differ between groups (see Supplementary Table 2).

$CD4^+$ T-cell activation, measured as baseline-corrected expression of CD25 on $CD3^+CD4^+CD45^+$ cells, was significantly higher in the control group than in the active group at week 26 ($P = .036$; Figure 3). When $CD4^+$ T-cell activation was analyzed in the participants who completed the study on treatment, this difference was significant at week 26 and week 52 ($P = .006$ and $P = .048$, respectively; data not shown). Moreover, $CD4^+$ T-cell activation correlated with $CD4^+$ T-cell count at 52 weeks

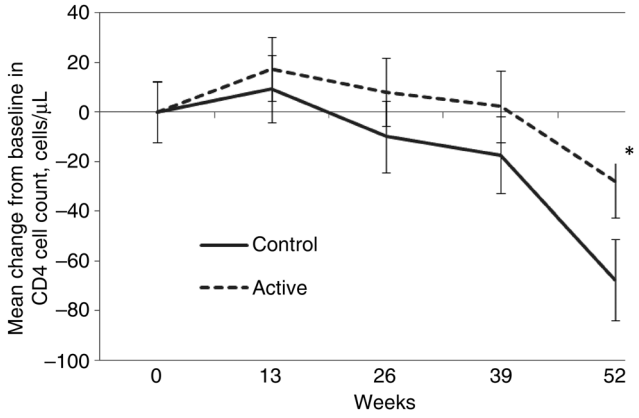

Figure 2. Change from baseline in CD4$^+$ T-cell count over 52 weeks of intervention (intention-to-treat population; *P = .03, change from baseline). Data are estimated marginal mean ± SE.

(r = 0.552, P = .027). Activation of CD8$^+$ T-cells, as analyzed by the expression of CD38 on CD3$^+$CD8$^+$CD45$^+$CD45RO$^+$ cells, was not significantly affected by the intervention (see Supplementary Table 2).

No difference was observed in FoxP3 expression between groups (data not shown).

Reasons for Study Discontinuation

In the active group, 83 patients (49%) dropped out of the study for reasons other than ARV treatment initiation, compared with 60 (35%) in the control arm (Figure 1). Loss to follow-up and withdrawal of informed consent occurred similarly in both groups. The number of patients with adverse events as the reason for discontinuation was 30 (18%) in the active group and 14 (8%) in the control group. Adverse events as the reason for dropout were GI-related events in 74% of the cases in the active group and in 57% of the cases in the control group.

ARV treatment initiation was a protocol-defined endpoint of the study. Although the number of individuals initiating ARV treatment during the study was not significantly different (n = 25 active vs n = 29 control groups), the average time to ARV treatment initiation was significantly longer in the active treatment group (230 ± 15 days, n = 25; P = .018) than in the ARV treatment dropout population receiving the control treatment (167 ± 13 days, n = 29; Figure 4). Baseline CD4$^+$ T-cell counts were analyzed as a potential confounding factor for the treatment effect on time to ARV treatment initiation. However, neither a significant difference between the 2 groups nor a significant correlation between both parameters were observed (data not shown).

Safety

Ten significant adverse events (SAEs) occurred in 9 participants during the study; 9 SAEs in 8 individuals in the control group and 1 SAE in 1 individual in the active group. The investigator considered 2 SAEs in the control group, diarrhea and non-Hodgkin lymphoma, to be possibly related to the use of the study product. After consultation with several HIV experts and the study's principal investigator, the DSMB classified non-Hodgkin lymphoma as being not related (see Supplementary Table 3 listing of SAEs per study group). There were 513 SAEs in the active group and 489 in the control group. Of

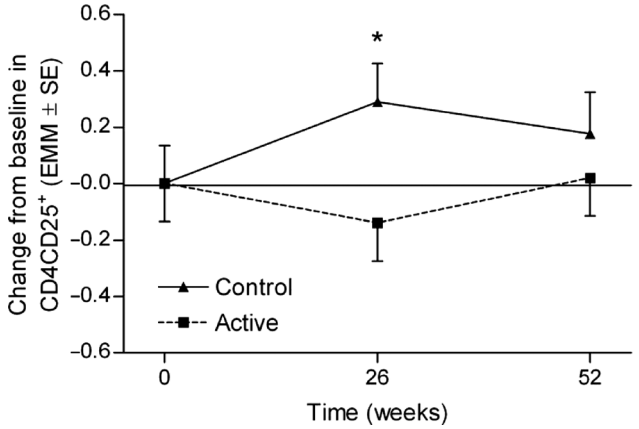

Figure 3. Changes from baseline in activated CD4$^+$ T cells (% CD4$^+$CD25$^+$) during the 52-week intervention period in the intention-to-treat population from a BITE pilot study (*P = .036, change from baseline).

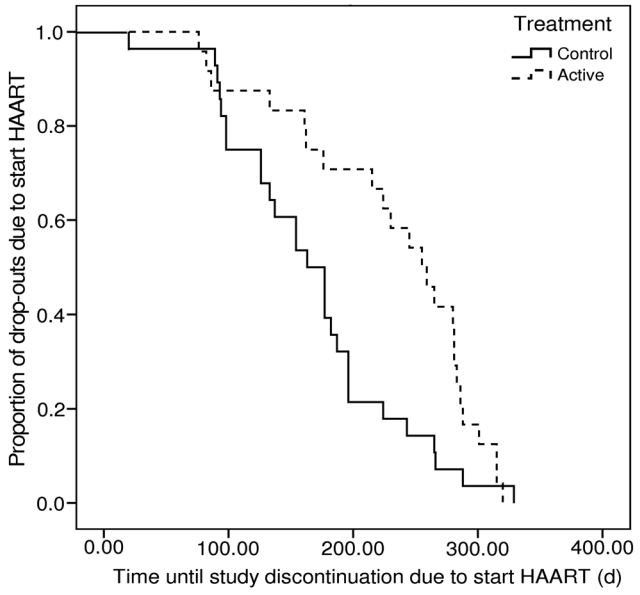

Figure 4. Time until participant study discontinuation due to highly active antiretroviral therapy therapy (HAART) initiation (population HAART dropouts; n = 25 for active treatment and n = 29 for control treatment).

these, 42% and 31% were product related in the active and control group, respectively, showing a higher number of GI symptoms in the active group (192 vs 140 in control group; see also Supplementary Table 4). Temporary mild to moderate GI complaints were noted more frequently in the active group: the abdominal distention score was significantly higher at week 5 ($P = .010$) and the flatulence score was higher until week 13 ($P < .001$). Blood safety parameter means were all within reference ranges, and no grade 3 or 4 abnormalities were reported. The mean baseline-corrected AST and ALT concentrations increased over time in the active group and reached statistical significance at week 52 compared with the control group (32.3 ± 18.8 [active] vs 27.9 ± 13.6 U/L [control], $P \leq .022$ and 38.9 ± 26.5 [active] vs 31.7 ± 23.0 U/L [control], $P \leq .001$, respectively). However, both AST and ALT concentrations remained within normal reference ranges.

DISCUSSION

$CD4^+$ T-cell decline in HIV-1–infected participants not on ARV treatment was significantly reduced with NR100157, a nutritional product specifically designed to support the GI tract of HIV-infected patients. In the control group, the rate of $CD4^+$ T-cell decline (68 cells/μL) at 52 weeks was within the expected range [1, 24]. However, the difference in decline between both groups was larger (40 cells/μL difference) than anticipated in the initial sample size calculation.

The reduction of $CD4^+$ T-cell count in the active group was not accompanied by any effects on viral load, emphasizing that disease progression is not an exclusive function of HIV replication. In a small pilot study, a significant reduction of $CD4^+$ $CD25^+$ T cells was observed. The significant correlation between $CD4^+$ T-cell activation and $CD4^+$ T-cell decline supports the potential relevance of this mechanism. A recent 12-week interventional study with 15 g oligosaccharides per day, the same as is present in NR100157, showed a significant reduction of $CD4^+CD25^+$ T cells accompanied by a significant reduction of soluble CD14 [16]. No clinically relevant safety concerns were found based on the reported adverse events and blood chemistry and hematology parameters. Liver transaminases were within reference values (ALT <45 U/L, AST <40 U/L).

Study limitations included the small sample size of the pilot study and the high loss to follow-up due to GI events, which resulted in interim termination of the trial. Adverse events accounted for 44 participants dropping out of the complete study. The most common adverse events were GI complaints, which were generally increased after intake of both NR100157 and the control product in those who dropped out (data not shown). Both the control and active products contained lactose. Even though lactose intolerance was an exclusion criterion for this study, it is known that the prevalence of lactose intolerance is higher in HIV-1–infected individuals than in healthy volunteers [25]. Undiagnosed lactose intolerance within the study population could also have contributed to the GI complaints experienced by those in both groups. Moreover, temporary GI complaints such as flatulence and abdominal distention can be expected after any diet change. Temporary GI complaints were reported in the early weeks of the intervention but disappeared over time in both the ITT population and the population of those who completed the study while on treatment. A variety of other factors led to study discontinuation, including initiation of ARV treatment, which has been a protocol-defined endpoint, and loss to follow-up. The current efficacy results could provide the incentive for higher compliance in a follow-up study.

The target population in this study was patients who were not yet receiving ARV treatment. However, current ARV treatment guidelines, whether for the developed or developing world are moving toward earlier initiation of therapy [26, 27], which is likely to reduce the size of the target population described in this study. In the recent randomized controlled CORAL trial, hyperimmune bovine colostrum was tested in HIV-infected patients with suboptimal $CD4^+$ T-cell response [28]. In that trial, hyperimmune bovine colostrum, similar to raltegravir, failed to reconstitute $CD4^+$ cell count when used for intensification in virally suppressive ARV therapy. Initial data from a recent study with probiotics in combination with ARV treatment in simian immunodeficiency virus–infected pigtail macaques showed enhanced mucosal immunity by reconstitution of $CD4^+$ T cells in the colon, decrease of $CD4^+$ activation, and increase in overall functionality of mucosal $CD4^+$ T cells [29]. Further studies are required to establish whether NR100157 can be more effective in combination with ARV treatment than ARV treatment alone in targeting and restoring the gut-associated lymphoid tissue, reducing persistent local immune activation, and thereby improving long-term prognosis.

Supplementary Data

Supplementary materials are available at *Clinical Infectious Diseases* online (http://cid.oxfordjournals.org/). Supplementary materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyedited. The contents of all supplementary data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

Notes

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The BITE study group included the following:

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All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Mellors JW, Munoz A, Giorgi JV, et al. Plasma viral load and CD4+ lymphocytes as prognostic markers of HIV-1 infection. Ann Intern Med **1997**; 126:946–54.
2. Hunt PW, Brenchley J, Sinclair E, et al. Relationship between T cell activation and CD4(+) T cell count in HIV-seropositive individuals with undetectable plasma HIV RNA levels in the absence of therapy. J Infect Dis **2008**; 197:126–33.
3. Sousa AE, Carneiro J, Meier-Schellersheim M, Grossman Z, Victorino RMM. CD4 T cell depletion is linked directly to immune activation in the pathogenesis of HIV-1 and HIV-2 but only indirectly to the viral load. J Immunol **2002**; 169:3400–6.
4. Gori A, Tincati C, Rizzardini G, et al. Early impairment of gut function and gut flora supporting a role for alteration of gastrointestinal mucosa in human immunodeficiency virus pathogenesis. J Clin Microbiol **2008**; 46:757–8.
5. Hummelen R, Vos AP, van't Land B, van Norren K, Reid G. Altered host-microbe interaction in HIV: a target for intervention with pro- and prebiotics. Int Rev Immunol **2010**; 29:485–513.
6. Deeks SG, Phillips AN. HIV infection, antiretroviral treatment, ageing, and non-AIDS related morbidity. BMJ **2009**; 338:a3172.
7. Mehandru S, Poles MA, Tenner-Racz K, et al. Lack of mucosal immune reconstitution during prolonged treatment of acute and early HIV-1 infection. PLoS Med **2006**; 3:e484.
8. Guadalupe M, Reay E, Sankaran S, et al. Severe CD4+ T-cell depletion in gut lymphoid tissue during primary human immunodeficiency virus type 1 infection and substantial delay in restoration following highly active antiretroviral therapy. J Virol **2003**; 77:11708–17.
9. Belmonte L, Olmos M, Fanin A, et al. The intestinal mucosa as a reservoir of HIV-1 infection after successful HAART. Aids **2007**; 21:2106–8.
10. Brenchley JM, Price DA, Schacker TW, et al. Microbial translocation is a cause of systemic immune activation in chronic HIV infection. Nat Med **2006**; 12:1365–71.
11. Jiang W, Lederman MM, Hunt P, et al. Plasma levels of bacterial DNA correlate with immune activation and the magnitude of immune restoration in persons with antiretroviral-treated HIV infection. J Infect Dis **2009**; 199:1177–85.
12. Brenchley JM, Douek DC. HIV infection and the gastrointestinal immune system. Mucosal Immunology **2008**; 1:23–30.
13. Giorgi JV, Hultin LE, McKeating JA, et al. Shorter survival in advanced human immunodeficiency virus type 1 infection is more closely associated with T lymphocyte activation than with plasma virus burden or virus chemokine coreceptor usage. J Infect Dis **1999**; 179:859–70.
14. Arslanoglu S, Moro GE, Schmitt J, Tandoi L, Rizzardi S, Boehm G. Early dietary intervention with a mixture of prebiotic oligosaccharides reduces the incidence of allergic manifestations and infections during the first two years of life. J Nutr **2008**; 138:1091–5.
15. Vos AP, Haarman M, van Ginkel JWH, et al. Dietary supplementation of neutral and acidic oligosaccharides enhances Th1-dependent vaccination responses in mice. Pediatr Allergy Immunol **2007**; 18:304–12.
16. Gori A, Rizzardini G, Van't Land B, et al. Specific prebiotics modulate gut microbiota and immune activation in HAART-naive HIV-infected adults: results of the "COPA" pilot randomized trial. Mucosal Immunol **2011**; 4:554–63.
17. Kotler DP, Gaetz HP, Lange M, Klein EB, Holt PR. Enteropathy associated with the acquired immunodeficiency syndrome. Ann Intern Med **1984**; 101:421–8.
18. Ullrich R, Zeitz M, Heise W, L'Age M, Hoffken G, Riecken EO. Small intestinal structure and function in patients infected with human immunodeficiency virus (HIV): evidence for HIV-induced enteropathy. Ann Intern Med **1989**; 111:15–21.
19. Barham JB, Edens MB, Fonteh AN, Johnson MM, Easter L, Chilton FH. Addition of eicosapentaenoic acid to gamma-linolenic acid-supplemented diets prevents serum arachidonic acid accumulation in humans. J Nutr **2000**; 130:1925–31.
20. Calder PC. The relationship between the fatty acid composition of immune cells and their function. Prostaglandins Leukot Essent Fatty Acids **2008**; 79:101–8.
21. Playford RJ, MacDonald CE, Calnan DP, et al. Co-administration of the health food supplement, bovine colostrum, reduces the acute

non-steroidal anti-inflammatory drug-induced increase in intestinal permeability. Clin Sci (Lond) **2001**; 100:627–33.
22. Herzenberg LA, De Rosa SC, Dubs JG, et al. Glutathione deficiency is associated with impaired survival in HIV disease. Proc Natl Acad Sci U S A **1997**; 94:1967–72.
23. Peterson JD, Herzenberg LA, Vasquez K, Waltenbaugh C. Glutathione levels in antigen-presenting cells modulate Th1 versus Th2 response patterns. Proc Natl Acad Sci U S A **1998**; 95:3071–6.
24. Madec Y, Boufassa F, Porter K, Meyer L, Collaboration C. Spontaneous control of viral load and CD4 cell count progression among HIV-1 seroconverters. AIDS **2005**; 19:2001–7.
25. Corazza GR, Ginaldi L, Furia N, Marani-Toro G, Di Giammartino D, Quaglino D. The impact of HIV infection on lactose absorptive capacity. J Infect **1997**; 35:31–5.
26. Thompson MA, Aberg JA, Cahn P, et al. Antiretroviral treatment of adult HIV infection: 2010 recommendations of the International AIDS Society-USA panel. JAMA **2010**; 304:321–33.
27. WHO. Antiretroviral therapy for HIV infection in adults and adolescents: recommendations for a public health approach: 2010 revision. Geneva: WHO, **2010**.
28. Byakwaga H, Kelly M, Purcell DF, et al. Intensification of antiretroviral therapy with raltegravir or addition of hyperimmune bovine colostrum in HIV-infected patients with suboptimal CD4+ T-cell response: a randomized controlled trial. J Infect Dis **2011**; 204:1532–40.
29. Klatt N, Canary L, Sun X, et al. Probiotic supplementation of ARV treatment during SIV infection of pigtail macaques results in enhanced GI tract CD4+ T cell frequency and immunological function. In: Conference on Retroviruses and Opportunistic Infections. Seattle, WA, **2012**.