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Alcohol and dietary factors associate with gut integrity and inflammation in HIV+ adults

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Abstract

Introduction—HIV+ adults have heightened monocyte activation and inflammation, at least partially due to altered gut integrity. The role of dietary factors on microbial translocation, inflammation and the downstream effect on markers of cardiovascular disease (CVD) has not been explored. Our purpose was to describe the longitudinal dietary patterns of HIV+ adults; and examine the relationship between dietary intake, gut integrity, inflammation and subclinical markers of CVD in HIV+ adults.

Methods—We conducted a secondary analysis of 147 HIV+ participants in a 96-week randomized clinical trial of rosuvastatin as primary CVD prevention. Dietary intake was assessed using a dietary recall; plasma gut integrity, monocyte activation and inflammation markers measured by ELISA; CVD risk assessed by carotid ultrasound and coronary artery calcium score. Linear mixed models were used to analyze longitudinally measured biomarkers.

Results—Median age was 45 years and 78% were male. At baseline participants consumed a mean (SD) of 108 (70) grams of fat daily, 19 (15.6) grams of fiber, 266 (186) grams of carbohydrates, 15.6 (5.9) grams of protein; 45% of the sample consumed alcohol. Over time, alcohol consumption was associated with several markers of gut integrity and inflammation (all p 's < 0.05).

Discussion—HIV+ adults in a contemporary, high-resource setting have poor dietary patterns. Alcohol use was associated with worse gut integrity and increased inflammation, while other aspects of diet (fiber, carbohydrates, fat) were not. These data add to growing evidence illustrating

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the need for a better understanding of the effect of lifestyle factors on comorbidities in HIV+ adults.

Introduction

In this era of effective HIV treatment, HIV+ adults experience marked increases in chronic comorbid conditions such as cardiovascular disease (CVD), kidney disease, and cancers. One of the most prevalent chronic conditions in this population is cardiovascular disease, of which HIV+ adults are at approximately two-fold higher risk for developing. (1, 2) Lifestyle behaviors including diet, exercise and smoking have long been linked to CVD and are increasingly being examined in adults living with HIV. (3–6) Different mechanisms have been proposed linking lifestyle behaviors to chronic diseases including increasing vascular health, decreasing inflammation, among others. (4) The role of lifestyle behaviors, particularly diet, on gut integrity and immune activation and subsequently on cardiovascular health has not yet been explored in this population, and may help define a direct pathway between diet and increased CVD in people living with HIV.

With the rapid increase in the scale-up of antiretroviral therapy (ART) at the end of the 20th century, several investigators examined the role of dietary intake on metabolic abnormalities in HIV+ adults on ART. This work focused on two key dietary components--fat and fiber--and suggested that decreased fat consumption and increased fiber consumption leads to improved insulin resistance and decreased hyperlipidemia in HIV+ adults; (7, 8) and more recent work reported similar results.(9, 10) Yet, the sample sizes in these studies were small, the population homogenous, and these studies had short follow-up. (10) A longer-term study with a diverse sample in the current HIV era will help us to understand the relationship between dietary composition, immune activation and cardiovascular health in this population.

HIV+ individuals also experience significant damage to the gut-associated lymphoid tissues with increases in inflammation leading to decreased TL4 expression and TH17 availability and a loss of enterocytes and tight junctions. (11) Bacterial products, such as lipopolysaccharide (LPS) can more easily translocate into the bloodstream, interact with LPS binding protein (LBP), and induce chronic systemic immune activation in HIV+ adults. (12–14) Metabolic conditions, including diabetes and obesity are also characterized by increased inflammation. A possible link between diet, inflammation and metabolic complications is the gut microbiota. This microbiota has many roles including assisting with immunity and metabolism. Disruption of this microbiota (dysbiosis) has been associated with a number of chronic diseases including irritable bowel syndrome, obesity and type 2 diabetes.(15) Diet can quickly change the gut microbiota with high fat diets raising serum LPS and LBP levels and leading to increased inflammation and weight gain. (16) Further, fiber can improve gut microbiota by increasing production of short-chain fatty acids, leading to immune regulation. (17) Finally, alcohol has also been shown to increase serum LPS levels, increasing inflammation. (18) Given the inflammatory milieu associated with HIV infection, it is possible a poor diet may exacerbate inflammation in HIV+ adults, thereby contributing to the excess cardiovascular disease in this population.

Before exploring this hypothesis, we need to understand the composition of the typical HIV + adult diet intake in the current HIV era. While historically, nutrition counseling for HIV+ patients has focused on preventing HIV-associated wasting, today's dietary recommendations focus on preventing metabolic disease. Though not based on any current or HIV-specific data, low carbohydrate, well balanced diets, with little alcohol consumption are common recommendations. (19–23) Though there are sound data describing the relationship of food security to nutritional status in HIV+ adults, typically in low and middle income settings; (24) we lack longitudinal evidence describing the dietary patterns of HIV+ adults in a high-resource setting and in the contemporary HIV treatment era.

Therefore, the purpose of our analysis is twofold: 1) to describe the longitudinal dietary patterns of HIV+ treated adults in a contemporary, high-resource setting; and 2) to examine the relationship between dietary intake, gut integrity, inflammation and subclinical markers of cardiovascular disease in HIV+ adults. We hypothesized that higher dietary intake of fat would be related to worse gut integrity (increased intestinal fatty acid binding protein (I-FABP) and lipopolysaccharide binding protein (LBP), and decreased zonulin-1), increased monocyte activation (proportion of pro-inflammatory monocytes CD14(Dim)CD16(+) and CD14+CD16+ monocytes; and soluble markers sCD14 and sCD163), inflammation (IL-6, sTNFR-II, VCAM) and worse markers of subclinical CVD (carotid intima-media thickness (CIMT) and coronary calcification (CAC)).

Materials and Methods

Study Design

We conducted an analysis of 147 HIV+ subjects followed for 96 weeks in the SATURN-HIV study, a placebo-controlled randomized clinical trial testing the effect of rosuvastatin on markers of cardiovascular and bone disease, to assess the effects of dietary intake on gut integrity and inflammation.

Study Subjects

All participants were HIV+, 18 years of age, and on stable HIV antiretroviral therapy for at least 3 months. All had HIV RNA <1000 copies/mL, a fasting LDL-cholesterol 130 mg/dL and triglyceride 500 mg/dL. Additionally, participants were had evidence of either heightened T-cell activation, identified as the proportion of activated CD8+ T cells of 19% or levels of high-sensitivity C-reactive protein (hsCRP) 2 mg/L. Participants were excluded if they had a history of coronary disease or diabetes, were pregnant or lactating, or had an active infectious or inflammatory condition. All study procedures were approved by the Institutional Review Board of University Hospitals Case Medical Center, Cleveland, Ohio, USA.

Study Procedures

Potential subjects were recruited via IRB-approved flyer from HIV clinics and AIDS service organizations in northeast Ohio. Interested subjects were scheduled for a screening visit at which study staff explained study procedures and obtained informed, written consent. At this visit, eligibility was confirmed with a fasting blood draw and through medical records

evaluation. If the subject met eligibility criteria, they were randomized 1:1 to rosuvastatin 10 mg daily vs. matching placebo.

Dietary assessment

At study entry, subjects completed a self-reported demographic and substance use questionnaire. At study entry, 24, 48 and 96 weeks post randomization subjects also completed a standardized food interview, the Block Alive Screener, by Nutrition Quest. A research assistant read the questions to all participants, at all time points, to ensure they understood and completed each question. This dietary assessment tool contains 55 questions and asks participants to report both frequency and quantity of food intake based on "eating habits over the past year or so." Portion size is asked for 32 food items and a series of 15 additional questions asked about usual intake of low-fat/trans-fat free or low-carbohydrate/low-sugar versions of various foods. Responses to these 15 questions were used to fine-tune estimates of fat, sugar, and dietary fiber; values for these adjustments were developed from NHANES 1999–2002 dietary recall data. Nutrient estimates are calculated by multiplying frequency, portion size, and nutrient content and summing over all foods. Four-month test-retest reliability (reproducibility) of the dietary questionnaire ranged from 0.66 to 0.78, indicating good reliability. (25, 26) This dietary interview takes approximately 10 minutes to complete. Additionally, alcohol use was assessed with four survey questions. Subjects were asked at each visit whether they currently consumed alcohol. If subjects responded affirmatively, they were prompted to note: (1) how many servings of beer, (2) liquor, or (3) wine they consumed in the past seven days. These responses were summed and analyzed by alcohol category.

Gut Integrity, Immune Activation, and Inflammation

At each of the four time points, subjects completed fasting (> 12 hours) blood draws to obtain measurements of lipid profiles, glucose and insulin levels. Additionally, plasma, serum and peripheral blood mononuclear cells were cryopreserved for measurement of markers of immune activation, systemic inflammation and gut integrity markers as previously described^{27,28}. Soluble markers of monocyte activation (sCD14 and sCD163) and proportion of pro-inflammatory monocytes (CD14+CD16+ and CD14(Dim)CD16(+) monocytes), and systemic inflammation (IL-6, sTNFRii, VCAM) were measured as previously described. (27)

We assessed gut integrity with plasma I-FABP (a systemic marker of gut epithelial cell death), LBP, (an indirect marker of bacterial translocation from the gut to the circulation) (28) and zonulin-1 (a marker of intestinal permeability since zonulin is expressed by viable gut epithelial cells to disassemble tight junctions between cells, increasing permeability and macromolecule absorption) (29). Cryopreserved plasma was assessed by immunoassay for the gut integrity markers intestinal fatty acid binding protein (I-FABP; Human FABP2 DuoSet, R&D Systems), LBP (Hycult Biotech, Plymouth Meeting, PA)), zonulin-1 (ALPCO, Salem, NH).

Markers of subclinical CVD

At entry, week 48 and week 96, mean-mean common carotid artery intima media thickness (CCA-IMT) was measured by high resolution ultrasound. (29,30) Measurements were taken at three separate angles bilaterally and the average of the six measurements was used for analysis. Coronary artery calcium score (CAC) was quantified offline as previously described from gated, non-contrast coronary calcium scans performed on a 64-slice multi-detector CT scanner. (29)

Statistical Analysis

For this exploratory study we performed descriptive analyses of all baseline demographics and study variables. We summarized all continuous and categorical variables using mean $\pm$ SD and percentages of certain categories, respectively. We examined the biomarkers data distribution graphically and by running frequency analysis, and also performed Shapiro-Wilk test for normality. Most of the measured biomarkers distributions were skewed. Therefore, we applied Box-Cox transformations to the biomarkers so that skewness of the biomarkers data was zero.

To examine our hypothesis that higher dietary intake of total daily fat, and lower polyunsaturated fat: saturated fat ratio and total dietary fiber intake led to worse markers of gut integrity which eventually results in increased monocyte activation, inflammation and worse markers of cardiovascular health, we used nonparametric (Spearman) correlation and scatter plots. Based on this correlational analysis, we identified several biomarkers were significantly associated (p -value less than 0.05). We further studied these associations in a regression framework after controlling for several covariates such as age, sex, race, BMI, current CD4+ T cell count, time, and statin treatment assignment (indicator variable). For baseline biomarker analyses we used multiple linear regression models, and for longitudinally measured biomarker analyses we used linear mixed effects (LME) models with random intercept and slope. In the LME model, we assumed that the random intercept and slope follow multivariate normal distribution with exchangeable variance-covariance structure. We also assumed that the missing data followed a missing at random mechanism. A number of subjects withdrew or were lost to follow-up from the study; however, none were due to drug-related adverse events or related to the specific values of our outcome variables that should have been obtained. According to the American Statistical Association statement on p -value, we emphasized on the magnitude of the relationship and effect size (nonzero beta coefficients) rather than (adjusted) p -values.(30) The analyses were carried out using STATA 14 SE (StataCorp, College Station).

RESULTS

Baseline characteristics

All 147 SATURN-HIV participants were included in this analysis, of which 118 completed all longitudinal measures. Demographic and baseline characteristics are displayed in Table 1. Mean age (SD) was 45 (9.9) years, mean BMI was 28.1 (6.5), 78% were male, 68% were African American, and 63% smoked at study entry. Mean current and nadir CD4+ T cell counts were 640 (300) and 200 (146) cells/mm³ respectively. Mean known duration of HIV

infection was 12 years (6.9). All participants were on ART and 85% had HIV-1 RNA <50 copies/mL.

Dietary Characteristics

On average, participants consumed 2395 (± 1547) kcals per day at baseline. Within this diet, participants consumed approximately 108 (± 70) grams of fat of which 36 grams were saturated fat (± 23), 19 (± 15.6) grams of fiber, 266 (± 186) grams of carbohydrates, 246.33 (± 172.44) grams of bad carbohydrates (total carbohydrates minus fiber), and 15.6 (± 5.9) grams of protein. Overall, 47% of participants self-reported consuming alcohol in the past week. Of those, they consumed a mean of 2.87 (± 5.97) beers per work, 2.22 (± 4.87) servings of liquor, and approximately one glass of wine per week. Total kcal, fat, fiber, carbohydrate and protein consumption all significantly declined over the 96 week study period (all p 's < 0.05). Alcohol consumption did not increase over time. Table 2 provided detailed longitudinal dietary characteristics as well as the U.S. Department of Health and Human Services' recommended ranges.

Associations among Diet, Gut Integrity, Inflammation and Cardiovascular Health

We hypothesized that higher dietary intake of total daily fat, higher "bad" carbohydrates (carbohydrate grams – fiber grams), lower polyunsaturated fat: saturated fat ratio, and lower total dietary fiber intake would be associated with worse gut integrity (I-FABP, LBP, zonulin). In contrast to our hypothesis, however, we found that polyunsaturated fat: saturated ratio significantly correlated (in the unexpected direction) with the gut integrity biomarker I-FABP (Spearman $r=0.252$, $p=0.006$) and that liquor consumption in the past week correlated with LBP ($r=0.192$, $p=0.037$) (Table 3).

Furthermore, the association between I-FABP and polyunsaturated: saturated fat ratio remained strong at baseline in a multiple linear regression analysis (beta=1.829, $p=0.014$) and longitudinally in the linear mixed model analysis (beta=1.254, $p=0.009$) after controlling for age, sex, race, BMI, current CD4+ T cell count, time and statin treatment assignment. The gut integrity biomarker LBP and monocyte activation biomarker sCD163 were also statistically significantly correlated $r=0.224$, $p=0.015$). After controlling for age, sex, race, BMI, current CD4+ T cell count, time, and statin treatment as covariates, baseline LBP is significantly associated with sCD163 in the multiple linear regression analysis (beta=0.020, $p=0.026$) and longitudinally measured sCD163 in LME model (beta=0.019, $p=0.002$).

We further hypothesized that alcohol intake would be associated with worse gut integrity and inflammation markers. Indeed, liquor consumption in the past week positively correlated with LBP ($r=0.192$, $p=0.037$); Over time, alcohol use, particularly liquor use was associated with gut integrity, monocyte activation and inflammation. After controlling for age, sex, race, BMI, treatment group, CD4+ T cell count, and time, beer consumption was associated with sCD14 (beta=0.024, $p=0.002$) and liquor consumption was associated with LBP (beta=0.326, $p=0.018$), sCD14 (beta=0.031, $p<0.001$) and IL-6 (beta=0.203, $p=0.004$) (Table 5).

With regard to subclinical vascular disease as measured by CAC score or CIMT, we observed mixed associations. For gut integrity markers, LBP positively correlated with CAC ($r=0.362$, $p=0.014$) but IFAB was negatively correlated ($r=-0.292$, $p=0.049$). The negative association of I-FABP with CAC remained after adjustment in the longitudinal LME model (Table 4). For inflammation markers, sTNFR-II was positively associated with both CCA-IMT and CAC, but VCAM was associated with CCA-IMT only (all $p<0.05$) in cross-sectional analyses. However, the relationship between VCAM and CCA-IMT was not statistically significant after controlling for potential confounders in regression models. In a longitudinal LME models, sTNFR-II was statistically significantly associated with both CCA-IMT (beta=0.004, $p=0.016$) and CAC (beta=0.053, $p=0.032$) scores after controlling for the covariates. Alcohol use was not associated with subclinical CVD in any of our analyses (all $p>0.05$).

Discussion

In our sample of HIV+ adults who were on stable ART in the United States, we found that their dietary intake met caloric needs but was comprised of mostly carbohydrates and fats. This macronutrient composition declined over time with a corresponding decrease in kcals. Alcohol consumption was stable over time. We did not find evidence to support the hypothesis that higher dietary intake of fat would be related to worse gut integrity, increased inflammation, or markers of subclinical CVD.

This is among the first data describing the longitudinal dietary composition of HIV+ adults in a well-resourced country in the contemporary ART era. Diet patterns indicate that on average, our participants consumed within the recommended ranges of calories for their age. The macronutrient composition of their diet, however did not meet recommendations. Specifically, the average carbohydrate consumption was almost twice recommended daily intake, while protein consumption was only a third of the recommended level. This is concerning since substantial evidence in the general population suggests low-carbohydrate and high protein diets favorably impact body mass and improves cardiovascular health. (31, 32) After initiating ART, approximately one third to one half of HIV+ adults are overweight or obese, (33, 34) and that obesity is more prevalent in HIV+ females. (35) Our data, when examined in conjunction with this evidence, suggest that structured, evidence-based interventions designed to lower carbohydrate and increase protein consumption, may help mitigate the rising obesity epidemic in HIV+ adults.

Alcohol use is common among HIV+ adults and our evidence suggests that liquor use is associated with increased microbial translocation and subsequent immune activation and inflammation. Alcohol is known to enhance LPS translocation acutely and over time. (18) This can occur by inducing nitric oxide synthase and NF- κ B signaling which can result in a differential expression of tight junction proteins, impairment of gut integrity indirectly through inflammation, or increasing one's LPS-producing gram negative bacteria microflora. (18) Heavy alcohol use in HIV has been found to quicken HIV disease progression, (36) and one study found an association between unhealthy alcohol intake and higher sCD14 in untreated HIV+ subjects in Uganda, (37) however to the best of our knowledge, no one has previously demonstrated associations of alcohol use with such a

broad range of markers of microbial translocation, immune activation, and inflammation in treated HIV+ adults. This evidence is consistent with the hypothesis that alcohol, in particular liquor, use increases translocation of LPS which interacts with LBP, resulting in increased immune activation and inflammation perhaps resulting in HIV disease progression. In contrast with a recent review by Kelso, et al., (38) we did not find evidence that alcohol use was associated with two common markers of subclinical CVD; however, it is plausible that alcohol use may be related to CVD risk (both positively and negatively) in HIV+ individuals through other pathways (i.e. plaque vulnerability, platelet effects).

Contrary to our hypotheses, other than alcohol we did not find consistent evidence that dietary intake was associated with markers of gut integrity, inflammation and markers of cardiovascular health. This was surprising and may be explained by our measure of dietary intake. Our dietary interview is a widely-used, valid and reliable measure of dietary intake. (26) Yet, like many self-report measures, it is subject to social desirability and memory biases leading participants to overestimate healthy behaviors. To overcome this, nutrition experts recommend collecting dietary intake over the previous 24 hours on multiple, nonconsecutive days to accurately reflect the usual dietary intake of an individual. (39, 40) Our findings may also be explained by the fact that, given the mean CD4+ T cell nadir of 200 cells/ul, our participants may represent a population of HIV+ adults with less exposure to severe immunodeficiency over the course of their HIV disease. (29) Hunt et al reported that the relationship between gut mucosal damage and microbial translocation was a less significant driver of mortality in treated HIV+ adults with historically less immunodeficiency. (29) It is possible that our subjects experienced less gut epithelial cell death over the course of their HIV, attenuating any relationship between diet, gut integrity and inflammation. Additionally, we have also previously shown that rosuvastatin reduces the levels of I-FABP, which may also have attenuated any relationship between diet, gut integrity and inflammation. (27) Taken together, this suggests that different macronutrients may have a differential impact on gut integrity, which may be modified by immunodeficiency and medications, and future research should continue to further examine this relationship.

In addition to finding a high carbohydrate and low protein diet, several other dietary characteristics are noteworthy. There was a trend towards high fat and low fiber consumption indicating that perhaps diet interventions should also target these macronutrients. These dramatic deviations from recommended dietary composition also suggest that an annual dietary assessment may be beneficial in developing targeted, personalized dietary interventions for individual HIV+ adults. Though it may be logistically challenging, our data suggest that improving nutrition is an untapped area in HIV primary care. Additionally, large HIV cohort studies (e.g., NA-ACCORD) should consider adding a regular, valid dietary assessment at their subjects' annual visit. As this population experiences increased chronic comorbidities often linked to diet (e.g., diabetes, bone disease, cardiovascular disease)(41), this rich data may help us better understand the unique aspects of HIV that contribute to this increased prevalence.

Limitations

This study has several limitations. First, the study was conducted at a single U.S. site with a predominately, male, African- American sample that had heightened inflammation. Though this represents the emerging HIV epidemic in the United States, our findings pertaining to dietary consumption may not be generalizable beyond this population. Second, we assessed gut integrity using distal serum markers. To better analyze the relationship between diet, gut integrity, inflammation and cardiovascular health a proximal measure of the gut microbiome (fecal DNA sampling) (42) could be used. Although this measure may be limited by subject acceptability, it is likely to be more direct measure of this pathway. Third, the parent study design required that we enrolled a sample with no history of coronary disease, diabetes, or active infectious or inflammatory conditions. The dietary patterns observed in this, perhaps healthier sample, may not be reflective of the general population with HIV. Finally, it's possible we have unmeasured variables, such as poverty and food security that may help us better understand the dietary patterns in this populations. Future studies investigating the natural dietary habits of HIV+ adults should examine the influence of these variables.

In conclusion, we have presented novel data detailing the generally poor dietary intake of HIV+ adults in a well-resourced setting. Alcohol use was consistently associated with poor gut integrity, immune activation and inflammation suggesting an urgent need to reduce alcohol use in this population. This evidence can be used as a foundation for developing evidence-based, targeted individual and structural interventions to help improve dietary intake and reduce alcohol consumption in this population.

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References

1. Freiberg MS, Chang CC, Kuller LH, Skanderson M, Lowy E, Kraemer KL, et al. HIV Infection and the Risk of Acute Myocardial Infarction. *JAMA Intern Med.* 2013 Mar 4;113:1–9.
2. Paisible A-L, Chang C-CH, So-Armah KA, Butt AA, Leaf DA, Budoff M, et al. HIV Infection, Cardiovascular Disease Risk Factor Profile, and Risk for Acute Myocardial Infarction. *JAIDS Journal of Acquired Immune Deficiency Syndromes.* 2015; 68(2):209–16. [PubMed: 25588033]
3. So-Armah K, Freiberg MS. CURRENT OPINION Cardiovascular disease risk in an aging HIV population: not just a question of biology. *Curr Opin HIV AIDS.* 2014; 9 000-
4. Dirajlal-Fargo S, Webel A, CT L, Kinley B, Labbato D, Sattar A, et al. The effect of physical activity on cardiometabolic health and inflammation in treated HIV infection. *Antiviral Therapy.* 2015 In Press.
5. Webel AR, Barkley J, Longenecker CT, Mittelsteadt A, Gripshover B, Salata RA. A Cross-Sectional Description of Age and Gender Differences in Exercise Patterns in Adults Living With HIV. *J Assoc Nurses AIDS Care.* 2015 Mar-Apr;26(2):176–86. [PubMed: 25249267]
6. Botros D, Somarriba G, Neri D, Miller TL. Interventions to address chronic disease and HIV: strategies to promote exercise and nutrition among HIV-infected individuals. *Curr HIV/AIDS Rep.* 2012 Dec; 9(4):351–63. [PubMed: 22933247]
7. Hadigan C. Dietary Habits and Their Association with Metabolic Abnormalities in Human Immunodeficiency Virus—Related Lipodystrophy. *Clinical Infectious Diseases.* 2003 Sep 1; 37(Supplement 2):S101–S4. 2003. [PubMed: 12942382]

8. Hendricks KM, Dong KR, Tang AM, Ding B, Spiegelman D, Woods MN, et al. High-fiber diet in HIV-positive men is associated with lower risk of developing fat deposition. *The American Journal of Clinical Nutrition*. 2003 Oct 1; 78(4):790–5. 2003. [PubMed: 14522738]
9. Lazzaretti RK, Kuhmmer R, Sprinz E, Polanczyk CA, Ribeiro JP. Dietary Intervention Prevents Dyslipidemia Associated With Highly Active Antiretroviral Therapy in Human Immunodeficiency Virus Type 1–Infected Individuals A Randomized Trial. *Journal of the American College of Cardiology*. 2012; 59(11):979–88. [PubMed: 22402068]
10. Stradling C, Chen YF, Russell T, Connock M, Thomas GN, Taheri S. The effects of dietary intervention on HIV dyslipidaemia: a systematic review and meta-analysis. *PLoS One*. 2012; 7(6):e38121. [PubMed: 22701607]
11. Sandler NG, Douek DC. Microbial translocation in HIV infection: causes, consequences and treatment opportunities. *Nat Rev Micro*. 2012; 10(9):655–66.
12. Brenchley JM, Price DA, Schacker TW, Asher TE, Silvestri G, Rao S, et al. Microbial translocation is a cause of systemic immune activation in chronic HIV infection. *Nat Med*. 2006; 12(12):1365–71. [PubMed: 17115046]
13. Lozupone Catherine A, Li M, Campbell Thomas B, Flores Sonia C, Linderman D, Gebert Matthew J, et al. Alterations in the Gut Microbiota Associated with HIV-1 Infection. *Cell Host & Microbe*. 2013; 14(3):329–39. [PubMed: 24034618]
14. Marchetti G, Tincati C, Silvestri G. Microbial Translocation in the Pathogenesis of HIV Infection and AIDS. *Clinical Microbiology Reviews*. 2013; 26(1):2–18. [PubMed: 23297256]
15. Guinane CM, Cotter PD. Role of the gut microbiota in health and chronic gastrointestinal disease: understanding a hidden metabolic organ. *Therap Adv Gastroenterol*. 2013 Jul; 6(4):295–308.
16. Graham C, Mullen A, Whelan K. Obesity and the gastrointestinal microbiota: a review of associations and mechanisms. *Nutrition Reviews*. 2015 2015-04-06 00:00:00.
17. Maslowski KM, Mackay CR. Diet, gut microbiota and immune responses. *Nat Immunol*. 2011 Jan; 12(1):5–9. [PubMed: 21169997]
18. Wang HJ, Zakhari S, Jung MK. Alcohol, inflammation, and gut-liver-brain interactions in tissue damage and disease development. *World J Gastroenterol*. 2010 Mar 21; 16(11):1304–13. [PubMed: 20238396]
19. Dubé MP, Stein JH, Aberg JA, Fichtenbaum CJ, Gerber JG, Tashima KT, et al. Guidelines for the Evaluation and Management of Dyslipidemia in Human Immunodeficiency Virus (HIV)-Infected Adults Receiving Antiretroviral Therapy: Recommendations of the HIV Medicine Association of the Infectious Disease Society of America and the Adult AIDS Clinical Trials Group. *Clinical Infectious Diseases*. 2003 Sep 1; 37(5):613–27. 2003. [PubMed: 12942391]
20. Gazzard BG. on behalf of the BTGWG. British HIV Association guidelines for the treatment of HIV-1-infected adults with antiretroviral therapy 2008. *HIV Medicine*. 2008; 9(8):563–608. [PubMed: 18826546]
21. Lundgren JD, Battegay M, Behrens G, De Wit S, Guaraldi G, Katlama C, et al. European AIDS Clinical Society (EACS) guidelines on the prevention and management of metabolic diseases in HIV*. *HIV Medicine*. 2008; 9(2):72–81. [PubMed: 18257770]
22. Myerson M, Malvestutto C, Aberg JA. Management of lipid disorders in patients living with HIV. *The Journal of Clinical Pharmacology*. 2015; 55(9):957–74. [PubMed: 25651539]
23. Ballocca F, Gili S, D'Ascenzo F, Marra WG, Cannillo M, Calcagno A, et al. HIV Infection and Primary Prevention of Cardiovascular Disease: Lights and Shadows in the HAART Era. *Progress in Cardiovascular Diseases*.
24. Weiser SD, Young SL, Cohen CR, Kushel MB, Tsai AC, Tien PC, et al. Conceptual framework for understanding the bidirectional links between food insecurity and HIV/AIDS. *The American Journal of Clinical Nutrition*. 2011 Dec 1; 94(6):1729S–39S. 2011. [PubMed: 22089434]
25. Block G, Sternfeld B, Block CH, Block TJ, Norris J, Hopkins D, et al. Development of Alive! (A Lifestyle Intervention Via Email), and Its Effect on Health-related Quality of Life, Presenteeism, and Other Behavioral Outcomes: Randomized Controlled Trial. *J Med Internet Res*. [Original Paper]. 2008; 10(4):e43.

26. Sternfeld B, Block C, Quesenberry CP Jr, Block TJ, Husson G, Norris JC, et al. Improving Diet and Physical Activity with ALIVE. *American Journal of Preventive Medicine*. 36(6):475–83. [PubMed: 19460655]
27. Funderburg N, Boucher M, Sattar A, Kulkarni M, Labbato D, Kinley B, et al. Rosuvastatin decreases Intestinal Fatty Acid Binding Protein (I-FABP), but does not alter zonulin or Lipopolysaccharide Binding Protein (LBP), in HIV-infected subjects on antiretroviral therapy. Under Review. 2016
28. Derikx JP, Luyer MD, Heineman E, Buurman WA. Non-invasive markers of gut wall integrity in health and disease. *World J Gastroenterol*. 2010 Nov 14; 16(42):5272–9. [PubMed: 21072889]
29. Hunt PW, Sinclair E, Rodriguez B, Shive C, Clagett B, Funderburg N, et al. Gut epithelial barrier dysfunction and innate immune activation predict mortality in treated HIV infection. *J Infect Dis*. 2014 Oct; 15(210)(8):1228–38.
30. Wasserstein RL, Lazar NA. The ASA's statement on p-values: context, process, and purpose. *The American Statistician*. 2016
31. Krieger JW, Sitren HS, Daniels MJ, Langkamp-Henken B. Effects of variation in protein and carbohydrate intake on body mass and composition during energy restriction: a meta-regression. *The American Journal of Clinical Nutrition*. 2006 Feb 1; 83(2):260–74. 2006. [PubMed: 16469983]
32. Mateo-Gallego R, Marco-Benedí V, Perez-Calahorra S, Bea AM, Baila-Rueda L, Lamiquiz-Moneo I, et al. Energy-restricted, high-protein diets more effectively impact cardiometabolic profile in overweight and obese women than lower-protein diets. *Clinical Nutrition*. 2016
33. Tate T, Willig AL, Willig JH, Raper JL, Moneyham L, Kempf MC, et al. HIV infection and obesity: where did all the wasting go? *Antivir Ther*. 2012; 17(7):1281–9. [PubMed: 22951353]
34. Koethe JR, Jenkins CA, Lau B, Shepherd BE, Justice AC, Tate JP, et al. Rising Obesity Prevalence and Weight Gain Among Adults Starting Antiretroviral Therapy in the United States and Canada. *AIDS Research and Human Retroviruses*. 2015; 32(1):50–8. 2016/01/01. [PubMed: 26352511]
35. Willig AL, Westfall AO, Overton ET, Mugavero MJ, Burkholder GA, Kim D, et al. Obesity Is Associated with Race/Sex Disparities in Diabetes and Hypertension Prevalence, But Not Cardiovascular Disease, Among HIV-Infected Adults. *AIDS Research and Human Retroviruses*. 2015; 31(9):898–904. 2015/09/01. [PubMed: 26114374]
36. Hahn JA, Samet JH. Alcohol and HIV Disease Progression: Weighing the Evidence. *Current HIV/AIDS Reports*. [journal article]. 2010; 7(4):226–33.
37. Carrico AW, Hunt PW, Emenyonu NI, Muyindike W, Ngabirano C, Cheng DM, et al. Unhealthy Alcohol Use is Associated with Monocyte Activation Prior to Starting Antiretroviral Therapy. *Alcoholism: Clinical and Experimental Research*. 2015; 39(12):2422–6.
38. Kelso NE, Sheps DS, Cook RL. The association between alcohol use and cardiovascular disease among people living with HIV: a systematic review. *The American journal of drug and alcohol abuse*. 2015; 41(6):479–88. [PubMed: 26286352]
39. Thompson FE, Subar AF, Loria CM, Reedy JL, Baranowski T. Need for technological innovation in dietary assessment. *Journal of the American Dietetic Association*. 2010; 110(1):48. [PubMed: 20102826]
40. Forster H, Walsh MC, Gibney MJ, Brennan L, Gibney ER. Personalised nutrition: the role of new dietary assessment methods. *Proceedings of the Nutrition Society*. 2016; 75(01):96–105. [PubMed: 26032731]
41. Schouten J, Wit FW, Stolte IG, Kootstra N, van der Valk M, Geerlings SG, et al. Cross-sectional comparison of the prevalence of age-associated comorbidities and their risk factors between HIV-infected and uninfected individuals: the AGEhIV Cohort Study. *Clinical Infectious Diseases*. 2014 Sep 2. 2014.
42. Le Chatelier E, Nielsen T, Qin J, Prifti E, Hildebrand F, Falony G, et al. Richness of human gut microbiome correlates with metabolic markers. *Nature*. [Article]. 2013; 500(7464):541–6.

Table 1Baseline Characteristics of SATUN-HIV study participants (*n*=147)

Demographic	
	Mean (SD)
Age at baseline (years)	45.4 (9.9)
Male (%)	115 (78)
Race	
African American (%)	100 (68)
Caucasian/Hispanic/Other (%)	47 (32)
Currently Smoking (%)	93 (63%)
Body Mass Index (kg/m ²)	28.07 (6.5)
HIV Disease	
Number of Years Since HIV Diagnosis	12.2 (6.9)
CD4+ T cell count (cells/ul)	639.9 (300)
CD4+ T cell nadir (cells/ul)	200.4 (146.2)
Number of Subjects with Viral Suppression (<75) (%)	123 (83.67)
Gut Integrity	
Intestinal fatty acid binding protein(pg/ml)	4530 (3207)
Lipopolysaccharide binding protein (µg/ml)	20.8 (10.9)
Inflammation	
IL-6 (pg/ml)	4.48 (9.4)
STNFRH (pg/ml)	2337.56 (836.68)
VCAM	722.40 (295.58)
sCD14	2267.40 (1361.38)
CD14+CD16+ monocytes	26.99 (12.74)
Indicators of Subclinical Cardiovascular Disease	
Carotid intima-media thickness (mm)	0.71 (0.14)
Coronary calcification (Agatston units)	31.5 (89.5)
Dietary Intake	
Total Calories	2394.92 (1547.01)

Demographic	
Bad Carbohydrate	246.33 (172.44)

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Table 2

Longitudinal dietary characteristics of HIV+ subjects¹

	Recommend Range ²	Baseline (n=147)	24 weeks (n=135)	48 weeks (n=127)	96 weeks (n=118)	p-value
kcal	1800–2400	2394.9 (1547)	2111.1 (1323)	2061.2 (1374)	1996.4 (1189.4) ³	0.017
Total fat (g)	65–70	108.4 (70.9)	94.3 (61.1)	91.5 (61.9)	88.1 (56.7) ³	0.045
Saturated fat (g)	22–24	36.0 (23.0)	31.59 (20.3)	31.1 (21.2)	29.9 (19.9) ³	0.014
Polyunsaturated fat (g)	<22 g	21.9 (15.0)	19.1 (13.1)	18.0 (12.8)	17.6 (11.6) ³	0.004
Fiber (g)	22.4–33.6	19.2 (15.6)	16.2 (12.5)	16.5 (13.5)	15.8 (10.9) ³	0.042
Carbohydrate (g)	130	265.5 (185.9)	234.2 (162.5)	227.2 (167.2)	224.3 (141.2) ³	0.064
Bad Carbohydrate		246.33 (172.44)	217.50 (151.88)	210.89 (155.67)	208.49 (133.18)	0.067
Protein (g)	46–56	15.6 (5.9)	15.1 (3.2)	15.2 (3.3)	15.9 (8.1) ³	0.012
Consumed alcohol in the past week (%)	n/a	69 (47)	75 (55)	74 (58)	60 (50)	0.779
Average number of beers/week (SD), of those consuming alcohol	n/a	2.87 (5.97)	2.41 (3.36)	2.92 (5.09)	2.81 (4.49)	0.500
Average glasses of wine /week	n/a	1.12 (1.84)	0.77 (1.38)	0.91 (1.47)	0.98 (1.38)	0.771
Average servings of liquor/week	n/a	2.22 (4.87)	1.62 (3.27)	1.99 (3.42)	2.81 (6.01)	0.550

¹ Results reported as means and standard deviations;

² Ranges based on an adult who consumes a daily diet of 1800–2400 kcal. Variations exist by age, sex and activity level. Source: 2015–2020 Dietary Guidelines for Americans 2015–2020. Eighth Edition.

³ Linear mixed models of dietary characteristics over time indicated a significant decline in macronutrient ; however, alcohol consumption did not change over time (using generalized estimating equations).

Table 3

Baseline associations¹ of Dietary Consumption with Markers of Gut Integrity and Subclinical Cardiovascular Disease

	I-FABP	LBP	Zonulin
kcal	−0.125	0.091	0.033
Total fat (g)	−0.104	0.109	0.021
Saturated fat (g)	−0.122	0.065	0.036
Polyunsaturated fat (g)	−0.042	0.108	0.045
Polyunsaturated Fat: Saturated Fat	0.252 ²	0.017	−0.011
Fiber (g)	−0.060	0.151	0.102
Carbohydrate (g)	−0.097	−0.078	−0.086
Bad Carbohydrates	−0.104	0.073	0.083
Protein (g)	−0.131	0.078	0.013
Beer in the last week	−0.001	0.068	−0.037
Wine in the last week	0.024	0.110	−0.004
Liquor in the last week	−0.068	0.192 ²	−0.031
Carotid Intima Media Thickness	−0.075	0.108	−0.158
Coronary Calcification	−0.292 ²	0.362 ²	0.024

¹ Spearman Rho,

² p<0.05

Table 4

Examining the Associations of Gut Integrity Markers with Markers of Inflammation and Subclinical Vascular Disease using Linear Mixed Models

Variable ¹	sCD163			VCAM			Coronary Calcification		
	β (SE)	95% CI	p-value	β (SE)	95% CI	p-value	β (SE)	95% CI	p-value
Intercept	3.193 (0.059)	(3.076, 3.309)	0.000	1.576 (0.003)	(1.571, 1.581)	0.000	5.123 (2.948)	(-0.655, 10.900)	0.082
Age	0.018 (0.006)	(0.005, 0.030)	0.006	0.001 (0.0003)	(0.0002, 0.001)	0.006	1.105 (0.256)	(0.603, 1.606)	0.000
Male	-0.009 (0.018)	(-0.044, 0.026)	0.601	-0.001 (0.001)	(-0.003, 0.0003)	0.116	-0.990 (0.692)	(-2.346, 0.366)	0.153
Race	-0.021 (0.014)	(-0.048, 0.005)	0.119	-0.002 (0.001)	(-0.003, -0.0004)	0.008	-0.703 (0.471)	(-1.626, 0.219)	0.135
BMI	0.002 (0.001)	(-0.00002, 0.004)	0.053	-0.0002 (0.00005)	(-0.0002, -0.0001)	0.001	0.063 (0.043)	(-0.023, 0.148)	0.150
Total Calories³	0.001 (0.004)	(-0.008, 0.009)	0.872	0.0001 (0.0002)	(-0.0004, 0.0005)	0.736	0.185 (0.203)	(-0.212, 0.582)	0.362
Statin	-0.005 (0.013)	(-0.030, 0.019)	0.667	0.001 (0.001)	(-0.0002, 0.002)	0.107	-0.713 (0.479)	(-1.652, 0.226)	0.136
CD4+	0.006 (0.007)	(-0.008, 0.019)	0.419	-0.001 (0.0003)	(-0.001, -0.0002)	0.010	0.003 (0.266)	(-0.518, 0.524)	0.991
Time	-0.003 (0.005)	(-0.013, 0.007)	0.552	0.0005 (0.0002)	(0.0001, 0.0008)	0.015	0.124 (0.205)	(-0.278, 0.526)	0.546
LBP	0.019 (0.006)	(0.007, 0.032)	0.002	0.001 (0.0003)	(0.0003, 0.001)	0.002	-	-	-
IFAB	-	-	-	-	-	-	-0.400 (0.133)	(-0.661, -0.139)	0.003

¹ Age = Age in decades; Race = African American; BMI = Body Mass Index (kg/m²); Statin = Statin Treatment Group; CD4+ = CD4+ T cells (cells/ul),

³ The rescaled variable was used (rescaled variable = original variable/standard deviation of the original variable)

Table 5

Examining the Associations of Alcohol Use with Gut integrity and Inflammation using Linear Mixed Models

Variable ¹	sCD14			Lipopolysaccharide Binding Protein			CD14+CD16+ monocytes			IL-6		
	β(SE)	95% CI	p-value	β(SE)	95% CI	p-value	β(SE)	95% CI	p-value	β(SE)	95% CI	p-value
Intercept	3.892 (0.029)	(3.836, 3.948)	0.000	4.550 (0.474)	(3.621, 5.479)	0.000	3.180 (0.416)	(2.364, 3.996)	0.000	-0.447 (0.231)	(-0.900, 0.005)	0.053
Age	0.008 (0.004)	(0.001, 0.015)	0.018	0.001 (0.059)	(-0.115, 0.116)	0.992	0.142 (0.052)	(0.041, 0.244)	0.006	0.107 (0.029)	(0.050, 0.163)	0.000
Male	0.028 (0.010)	(0.008, 0.048)	0.006	0.273 (0.167)	(-0.054, 0.600)	0.102	0.444 (0.147)	(0.157, 0.731)	0.002	-0.023 (0.082)	(-0.183, 0.138)	0.783
Race	-0.021 (0.008)	(-0.036, -0.005)	0.008	-0.099 (0.126)	(-0.345, 0.147)	0.432	-0.152 (0.114)	(-0.375, 0.071)	0.181	-0.007 (0.063)	(-0.131, 0.117)	0.910
BMI	-0.001 (0.001)	(-0.002, -0.0002)	0.018	-0.020 (0.009)	(-0.038, -0.002)	0.031	0.004 (0.008)	(-0.012, 0.020)	0.592	0.025 (0.005)	(0.016, 0.034)	0.000
Total Calories ³	0.002 (0.003)	(-0.004, 0.008)	0.582	0.062 (0.045)	(-0.025, 0.149)	0.165	0.079 (0.045)	(-0.010, 0.168)	0.082	0.060 (0.024)	(0.012, 0.107)	0.014
Statin	0.005 (0.007)	(-0.009, 0.019)	0.471	0.137 (0.116)	(-0.090, 0.363)	0.237	-0.056 (0.101)	(-0.255, 0.142)	0.579	-0.038 (0.057)	(-0.148, 0.073)	0.507
CD4+	-0.003 (0.004)	(-0.010, 0.005)	0.478	-0.094 (0.063)	(-0.218, 0.029)	0.134	0.083 (0.054)	(-0.022, 0.188)	0.121	0.031 (0.030)	(-0.027, 0.090)	0.291
Time	-0.001 (0.003)	(-0.007, 0.004)	0.644	0.055 (0.056)	(-0.054, 0.164)	0.325	0.005 (0.037)	(-0.068, 0.079)	0.885	-0.019 (0.020)	(-0.059, 0.021)	0.348
Liquor	0.031 (0.009)	(0.014, 0.048)	0.000	0.326 (0.138)	(0.056, 0.595)	0.018	0.408 (0.125)	(0.163, 0.654)	0.001	0.203 (0.070)	(0.066, 0.341)	0.004

¹ Age = Age in decades; Race = African American; BMI = Body Mass Index (kg/m²); Statin = Statin Treatment Group; CD4+ = CD4+ T cells (cells/ul); Liquor = Liquor Use;

³ The rescaled variable was used (rescaled variable = original variable/standard deviation of the original variable)