Nonalcoholic Fatty Liver Disease in HIV-Infected Patients Referred to a Metabolic Clinic: Prevalence, Characteristics, and Predictors

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Background. The prevalence and predictors of nonalcoholic fatty liver disease (NAFLD) in human immunodeficiency virus (HIV)–infected highly active antiretroviral therapy–experienced patients and the association of NAFLD with risk of cardiovascular disease and subclinical atherosclerosis are unknown.

Methods. We performed a cross-sectional observational study. NAFLD was defined by liver-spleen attenuation values of <1.1 on computed tomography in persons who had neither evidence of chronic viral hepatitis nor a significant history of alcohol consumption.

Results. We enrolled 225 patients; 163 (72.4%) were men. Mean (±SD) HIV infection duration was 147 ± 60 months, and mean (±SD) body mass index (calculated as weight in kilograms divided by the square of height in meters) was 23.75 ± 3.59. NAFLD was diagnosed in 83 patients (36.9% of the total cohort). The following variables were significantly associated with NAFLD in univariate analyses: sex, waist circumference, body mass index, cumulative exposure to nucleoside reverse-transcriptase inhibitors, visceral adipose tissue, homeostasis model assessment of insulin resistance index, serum alanine and aspartate aminotransferase levels, and ratios of total serum cholesterol to high-density lipoprotein cholesterol. Coronary artery calcium scores and a diagnosis of diabetes were not associated with NAFLD. In multivariable logistic regression analyses, factors associated ($P < .001$) with NAFLD were higher serum alanine to aspartate ratio (odds ratio, 4.59; 95% confidence interval, 2.09–10.08), male sex (odds ratio, 2.49; 95% confidence interval, 1.07–5.81), greater waist circumference (odds ratio, 1.07; 95% confidence interval, 1.03–1.11), and longer nucleoside reverse-transcriptase inhibitor exposure (odds ratio, 1.12 per year of exposure; 95% confidence interval, 1.03–1.22).

Conclusions. NAFLD is common among HIV-infected persons who have the traditional risk factors for NAFLD (elevations in serum alanine level, male sex, and increased waist circumference) apparent. Exposure to nucleoside reverse-transcriptase inhibitors was an independent risk factor for NAFLD, with an 11% increase in the odds ratio for each year of use.

Nonalcoholic fatty liver disease (NAFLD) is a clinical-pathological syndrome that includes a range of disorders associated with fatty liver and that occurs in the absence of chronic infection with viral hepatitis or a patient history of significant alcohol consumption [1]. The clinical consequences of NAFLD not only include progression to liver fibrosis and hepatocellular insufficiency but also include the possibility of developing metabolic alterations that can lead to atherosclerosis [2]. Currently, a relationship between NAFLD and metabolic syndrome [1] is being increasingly recognized in the context of multiple classic and nonclassic risk factors for cardiovascular disease (CVD) [2]. NAFLD might be not only a marker but also an early mediator of CVD. This hypothesis underscores the importance of assessing overall CVD risk in patients with NAFLD [3].

The benchmark for NAFLD diagnosis is histological assessment of liver fat content through use of biopsy

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[1]. Noninvasive imaging evaluations to diagnose steatosis include ultrasound, CT, and MRI. With use of non–contrast-enhanced CT, fatty liver can be determined because normal liver tissue attenuation is greater than that of the spleen. When this relationship is reversed, with a difference in liver-spleen attenuation of >10 Houndsfield units, hepatic steatosis is suspected [4]. Among living liver donors, it has been shown that a liver-spleen attenuation ratio <1.1 can predict >30% of cases of hepatic steatosis [5]. Liver-spleen ratios can discriminate between none-to-mild and moderate-to-severe hepatic steatosis, with sensitivity and specificity of 0.833 and 0.815, respectively [5]. Although NAFLD is the most common cause of elevated serum hepatic transaminases in Western countries, elevations in serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are not uniformly seen and cannot be used reliably to confirm the diagnosis [6].

NAFLD prevalence in general population–based surveys varies from 14% to 31% [7]. NAFLD is found in more than two-thirds of obese individuals, regardless of whether diabetes mellitus is a comorbid condition [8]. The risk of NAFLD increases when body mass index (BMI, calculated as weight in kilograms divided by the square of height in meters) is >25 or when waist circumference is >102 cm for men [9–12].

The few studies analyzing NAFLD prevalence in HIV-infected populations [13–15] suggest that NAFLD may affect 30%–40% of patients with HIV infection [16]. Sutinen et al. [15] demonstrated that the level of liver fat was significantly higher among 25 HIV-infected patients with lipodystrophy (LD) who received HAART than it was among HIV-seronegative subjects. Moreno-Torres et al. [14] found intrahepatic triglyceride deposits in 17 of 29 HAART recipients, of whom 4 (13.8%) had liver fat contents compatible with a diagnosis of liver steatosis. More recently, Hadigan et al. [17] identified hepatic steatosis in 42% of their subjects. All of these studies involved the use of proton spectroscopy to measure liver fat content, which is capable of identifying liver fat content >5%. Mohammed et al. [13] evaluated diverse patients with NAFLD and found that affected HIV-infected persons had lower BMIs than did affected HIV-seronegative patients, and they suggested that NAFLD may be associated with factors other than those known to occur as a consequence of obesity, including HIV infection and antiretroviral therapy.

Lemoine et al. [18], using liver biopsy, were able to detect nonalcoholic steatohepatitis (NASH) in 6 of 9 insulin-resistant lipodystrophic patients and in 2 of 5 non–insulin-resistant patients with HIV infection; evidence of liver fibrosis was apparent in patients with NAFLD.

A potential etiologic role for antiretroviral agents in NAFLD is yet to be determined, and NAFLD may represent a newly recognized long-term toxicity that should be looked for in HAART-exposed patients. The objective of this study was to assess the prevalence of and risk factors for NAFLD in HIV-infected, HAART-experienced patients and its association with cardiovascular risk and subclinical atherosclerosis.

METHODS

We conducted a cross-sectional observational study that included all consecutive HIV-infected patients seen at the metabolic clinic of University of Modena and Reggio Emilia School of Medicine (Modena, Italy) during January 2006 through June 2007 who had received antiretroviral therapy for at least 2 years.

The multidisciplinary team includes infectious diseases physicians, cardiologists, endocrinologists, radiologists, nutritionists, personal trainers, psychologists, and plastic surgeons [19]. Inclusion criteria were serologically documented HIV infection, age >18 years, at least 2 years of HAART exposure, and, among persons with hyperlipidemia or hyperglycemia that required treatment, receipt of stable lipid-lowering and diabetic medication for at least 6 months. Exclusion criteria were serological evidence of hepatitis B or C infection documented with hepatitis B surface antigen and hepatitis C virus antibody, autoimmune hepatitis, diagnosis of any inborn error of metabolism, evidence of illicit drug use, or alcohol consumption >20 g of ethanol per day.

Demographic characteristics and clinical data, including duration of HIV infection, prior opportunistic diseases (Centers for Disease Control and Prevention classification), antiretroviral therapy history, and lifestyle were obtained from medical files. Smoking status, alcohol consumption, and physical activity were assessed at study entry, with the following criteria: smoking status, if applicable, was classified as being heavy or low, with a use cutoff of 10 cigarettes per day; alcohol consumption was defined as heavy when >20 g of ethanol per day was consumed; physical activity was defined as mild or intensive when activity of <4 h or ⩾4 h per week, respectively, was reported.

Insulin resistance (IR) was calculated using the homeostasis model assessment (HOMA) equation—HOMA-IR = fasting insulin (mU/mL) × fasting glucose mmol/L)/22.5. Information about therapy for hyperglycemia—including insulin and oral hypoglycemic agents, omega-3 fatty acids, fibrates, and statins—was collected from medical records.

CD4 cell counts (most recent value and nadir), plasma HIV RNA levels, and cumulative exposure to nonnucleoside and nucleoside reverse-transcriptase inhibitors, protease inhibitors, fusion inhibitors, and specific antiretrovirals within each class were recorded. Serum hepatic transaminase, total cholesterol, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, triglyceride, apolipoprotein A and B, glucose, and insulin levels were assessed at study entry after an overnight fast.

Table 1. Demographic characteristics, medical history, serum biochemistry results, liver function test results, and anthropometric measurements, by nonalcoholic fatty liver disease (NAFLD) status.

Variable	Patients with NAFLD	Patients without NAFLD	Measure of association (95% CI)	P^a
Demographic characteristics				
Sex			OR, 3.22 (1.59–6.49)	**<.001**
Male	71 (85.54)	92 (64.79)		
Female	12 (14.46)	50 (35.21)		
Age, mean years ± SD	48.43 ± 8.16	48.04 ± 8.84	MD, −0.39 (−2.74 to −1.95)	.740
Alcohol consumption			OR, 0.63 (0.36–1.12)	.112
None	55 (67.07)	80 (56.34)		
Not heavy (⩽20 g per day)	27 (32.93)	62 (43.66)		
Smoking status, by no. of cigarettes per day				.592
None	53 (64.63)	82 (57.75)		
<10	10 (12.20)	20 (14.08)	OR, 0.77 (0.34–1.78)	
⩾10	19 (23.17)	40 (28.17)	OR, 0.73 (0.39–1.40)	
Physical activity				
None	50 (60.98)	83 (58.45)		.625
Mild	21 (25.61)	44 (30.99)	OR, 0.79 (0.42–1.48)	
Intensive	11 (13.41)	15 (10.56)	OR, 1.22 (0.52–2.86)	
Medical history				
Blood pressure, mean ± SD mm Hg				
Diastolic	80.33 ± 13.01	77.64 ± 13.46	MD, −2.69 (−6.32 to 0.95)	.146
Systolic	121.06 ± 15.59	119.58 ± 16.26	MD, −1.48 (−5.86 to 2.90)	.505
Diabetes			OR, 1.76 (0.82–3.79)	.148
Absent	67 (81.71)	126 (88.73)		
Present	15 (18.29)	16 (11.27)		
Exogenous insulin use			OR, 1.74 (0.11–28.21)	.698
No	81 (98.78)	141 (99.30)		
Yes	1 (1.22)	1 (0.70)		
Oral hypoglycemic drug use			OR, 1.65 (0.67–4.08)	.278
No	72 (87.80)	131 (92.25)		
Yes	10 (12.20)	11 (7.75)		
Omega-3 fatty acid use			OR, 1.32 (0.53–3.27)	.555
No	74 (89.16)	130 (91.55)		
Yes	9 (10.84)	12 (8.45)		
Fibrate use			OR, 4.64 (1.92–11.23)	**<.001**
No	65 (78.31)	134 (94.37)		
Yes	18 (21.69)	8 (5.63)		
Statin (HMG CoA reductase inhibitor) use			OR, 1.58 (0.58–4.26)	.373
No	75 (90.36)	133 (93.66)		
Yes	8 (9.64)	9 (6.34)		
Biochemistry				
Serum lipid				
Triglycerides, mean mmol/L ±SD	2.19 ± 1.21	2.21 ± 1.75	MD, 0.03 (−0.41 to 0.46)	.908
Total cholesterol, mean mmol/L ±SD	5.04 ± 1.23	5.16 ± 1.09	MD, 0.12 (−0.19 to 0.44)	.451
HDL, mean mmol/L ±SD	1.11 ± 0.36	1.21 ± 0.35	MD, 0.1 (0–0.2)	**.047**
LDL, mean mmol/L ±SD	3.3 ± 1	3.3 ± 0.9	MD, −0.01 (−0.27 to 0.25)	.966
Total cholesterol to HDL ratio, mean value ±SD	4.91 ± 1.78	4.49 ± 1.23	MD, −0.42 (−0.82 to −0.02)	**.040**
Apolipoprotein A, mean mg/dL ±SD	138.13 ± 29.35	141.83 ± 22.50	MD, 3.70 (−3.29 to 10.70)	.298
Apolipoprotein B, mean mg/dL ±SD	109.25 ± 28.03	106.90 ± 26.42	MD, −2.35 (−9.86 to 5.15)	.537
Glucose metabolism and insulin resistance				
Glucose, mean mmol/L ±SD	5.76 ± 2.07	5.35 ± 1.5	MD, −0.41 (−0.9 to 0.06)	.090
HOMA-IR, mean value ±SD	5.11 ± 4.55	4.09 ± 9.33	MD, −1.02 (−3.24 to 1.20)	.368
HOMA-IR in nondiabetic patients, mean value ±SD	4.17 ± 2.81	2.49 ± 1.37	MD, −1.68 (−2.28 to −1.07)	**<.001**
Liver function test results				
AST level, mean U/L ±SD	33.20 ± 22.24	25.08 ± 11.80	MD, −8.12 (−12.68 to −3.57)	**<.001**
ALT level, mean U/L ±SD	51.18 ± 52.75	28.44 ± 16.25	MD, −22.74 (−32.28 to −13.19)	**<.001**

(continued)

Table 1. ***(Continued.)***

Variable	Patients with NAFLD	Patients without NAFLD	Measure of association (95% CI)	P[a]
FIB-4 index				
Mean value ± SD	1.57 ± 0.87	1.29 ± 0.47	MD, −0.27 (−0.69 to 0.14)	.190
FIB-4 score <1.45	10 (55.56)	18 (72)	OR, 0.48 (0.13–1.74)	.266
Anthropometric characteristics				
BMI, mean value ± SD	25.12 ± 3.63	22.96 ± 3.33	MD, −2.16 (−3.10 to −1.21)	**<.001**
Waist circumference, mean cm ± SD	90.26 ± 9.24	83.88 ± 9.27	MD, −6.38 (−8.92 to −3.85)	**<.001**
Waist-to-hip ratio, mean value ± SD	0.99 ± 0.07	0.95 ± 0.06	MD, −0.04 (−0.06 to −0.03)	**<.001**
Total fat cell mass, mean g ± SD	11459.45 ± 6012.90	10358.35 ± 6200.57	MD, −1101.1 (−2795.27 to 593.07)	.202
Total lean cell mass, mean g ± SD	55473.21 ± 8564.50	49337.76 ± 9426.15	MD, −6135.46 (−8655.57 to −3615.34)	**<.001**
VAT, mean cm^3 ± SD	167.23 ± 75.49	126.38 ± 100.60	MD, −40.85 (−65.97 to −15.73)	**.002**
SAT, mean cm^3 ± SD	148.32 ± 100.74	129.62 ± 84.54	MD, −40.85 (−65.97 to −15.73)	.239
VAT to BMI ratio, mean value ± SD	6.63 ± 2.80	5.38 ± 4.15	MD, −1.25 (−2.26 to −0.23)	**.017**
VAT to TAT ratio, mean value ± SD	0.55 ± 0.17	0.51 (0.18)	MD, −0.04 (−0.09 to 0.01)	.088
VAT to SAT ratio, mean value ± SD	1.65 ± 1.31	1.59 (2.21)	MD, −0.06 (−0.58 to 0.47)	.826
No lipodystrophy	10 (12.20)	18 (13.14)	…	
Lipoatrophy	23 (28.05)	66 (48.18)	OR, 0.63 (0.25–1.55)	**.006**
Fat accumulation or mixed forms	49 (59.76)	53 (38.69)	OR, 1.66 (0.70–3.95)	

NOTE. Data are no. (%) of patients, unless otherwise indicated. Percentages are based on available data. ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index (calculated as weight in kilograms divided by the square of height in meters); HDL, high-density lipoprotein; HMG CoA, 3-hydroxy-3-methylglutaryl coenzyme A; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein; MD, mean difference; SAT, subcutaneous adipose tissue; TAT, total adipose tissue; VAT, visceral adipose tissue.

[a] Significant values are shown in bold.

The FIB-4 index was calculated as the putative predictor of liver fibrosis in a subgroup of patients in our cohort [20]. An FIB-4 index <1.45 was used to exclude any extensive fibrosis (F3–F4) and an FIB-4 index >3.25 was used to confirm the existence of significant fibrosis (F3–F4) [21]. LD was defined using the Multicenter AIDS Cohort Study definition, with anthropometric categorizations of lipoatrophy, lipohypertrophy, and mixed form [22].

The following anthropometric measurements were taken on the same day that samples were obtained to perform serum chemistries:

1. Waist circumference and hip circumference;
2. BMI;
3. Total body fat mass and total lean cell mass, determined using whole-body dual energy X-ray absorptiometry (DEXA); and
4. Abdominal visceral adipose tissue volume (VAT), subcutaneous adipose tissue volume (SAT), and total adipose tissue volume (TAT), as well as VAT:TAT, VAT:SAT, and VAT:BMI ratio calculations, with use of single-slice abdominal CT at the level of the L4 vertebra, as per standard protocols.

The outcome variable was NAFLD expressed as a ratio of mean liver-spleen (L:S) attenuation values at CT.

Calculation of the L:S ratio. All CT examinations were performed with a 64-multislice CT (LightSpeed VTC; General Electric Medical System). Hepatic and splenic attenuation values were measured using noncontrast CT by using circular region of interest cursors in the liver and in the spleen. All measurements were manually obtained in regions of uniform parenchyma attenuation, with care taken to avoid vessels and other areas that might give spuriously increased or decreased measurements. Measurements from each point of the liver were averaged. The L:S ratio was calculated as follows: L:S ratio = average attenuation value of liver (4 points)/attenuation value of spleen. NAFLD was defined by a liver-to-spleen ratio <1.1.

Cardiovascular risk assessment. We undertook cardiovascular risk assessments for each patient, using the Framingham equation [23] and the PROCAM score (based on the Prospective Cardiovascular Münster study) (for female patients, who have a 4-fold less risk than men, as suggested by the authors of the International Task Force for the Prevention of Coronary Heart Disease) [24]. Metabolic syndrome was defined according to National Cholesterol Education Program criteria [25]. Coronary artery calcium was quantified by means of a volume score, as described and validated elsewhere [26, 27], with use of the same multislice CT that was used for the L:S assessment. Coronary artery calcium score was both used as a continuous variable and transformed to a categorical variable, adjusted for sex and age, as suggested by Hoff et al. [28].

Statistical analysis. Comparisons between continuous variables were performed using the Student's t test, whereas the χ^2 test was used for qualitative variables. Logarithmic trans-

formation was performed on data that were not normally distributed. A P value <.05 was considered to be statistically significant.

Univariate and multivariate logistic regression analyses were performed to identify factors associated with NAFLD. ORs and 95% CIs were reported with P values. All analyses were done using the statistical software package Stata, version 9.2 for Windows (STATA).

RESULTS

Three hundred forty-one HIV-infected patients, each with at least 2 years of HAART experience, underwent an initial screening for NAFLD by CT. Per protocol, patients were excluded from analysis as follows: 10 participants who admitted alcohol abuse or who drank >20 g/day, 91 who had positive results of tests for hepatitis C virus serum antibody, 15 who were positive for serum hepatitis B surface antigen, 8 who had serological test results positive for both hepatitis B virus and hepatitis C virus, and 2 who had serological test results positive for both hepatitis B virus and hepatitis D virus. Of the 225 patients eligible for our study, 163 (72.44%) were men, and 62 (27.56%) were women.

Mean patient age was 48 years (range, 19–74 years). Mean BMI (±SD) was 23.75 ± 3.6. Eleven patients (4.9%) were obese (BMI, ⩾30). Diabetes was diagnosed in 31 patients (13.78%). Hypertransaminasemia, defined as an AST level >38 U/L or an ALT level >40 U/L, was present in 63 participants (28.0%).

A liver-spleen ratio, determined by CT, of <1.1 was diagnosed in 83 patients. Overall, NAFLD prevalence in the study population was 36.89%, with a prevalence of 43.56% in men and 19.35% in women ($P < .001$).

Baseline characteristics of study participants and HIV clinical histories, grouped according to NAFLD status, are presented in table 1. Mean HIV infection duration (±SD) was 157.45 ± 60.15 months. FIB-4 values <1.45 were identified in 10 patients with NAFLD and in 18 control subjects, FIB-4 values of 1.45–3.25 were evident in 7 patients with NAFLD and in 7 control subjects, and an FIB-4 value >3.25 was found in 1 patient with NAFLD.

Table 2 depicts participant HIV clinical histories and antiretroviral drug class exposures by NAFLD status. Table 3 describes cardiovascular event histories and prevalences of classic and nonclassic risk factors for CVD by NAFLD status.

The following variables, all of which were significantly associated with NAFLD in univariate analyses, were evaluated in multivariable analyses: sex, waist circumference, BMI, cumulative exposure to NRTIs, VAT, VAT:BMI, HOMA-IR, ALT:AST, a ratio of total cholesterol to high-density lipoprotein cholesterol, and total lean body mass.

In stepwise multivariable logistic regression analyses (table 4), the following were independently associated with a diagnosis of NAFLD: elevated serum ALT:AST (OR, 4.59; 95% CI, 2.09–10.08), male sex (OR, 2.49; 95% CI, 1.07–5.81), elevated waist circumference (OR, 1.07; 95% CI, 1.03–1.11), and NRTI exposure (OR, 1.12; 95% CI, 1.03–1.22). The risk of NAFLD increased 1.12 times for each year of NRTI exposure.

DISCUSSION

In this large cohort of very well-characterized HIV-infected patients, we found a prevalence of NAFLD that was higher than anticipated (nearly 40%) and significant associations between the presence of NAFLD and other patient features. We observed a positive association between NAFLD and cumulative exposure to NRTIs (with an increased risk associated with each additional year of NRTI exposure), male sex, and biochemical evidence of hepatocellular inflammation (as evidenced by elevations in serum ALT and/or AST levels).

We found a positive association between NAFLD and waist size. NAFLD was associated with the presence of LD (when considered as a categorical variable) but not with specific morphological phenotypes characteristic of LD. Although we did not find a significant association between NAFLD and predictive algorithms for CVD, there was a trend toward statistical significance for the association of NAFLD with PROCAM scores ($P = .06$) or coronary artery calcium scores ($P = .08$). The high prevalence of NAFLD seen in our cohort is comparable to recently reported findings from another cohort study in which magnetic resonance spectroscopy was used for diagnosis [17].

To our knowledge, this is the largest study to date that has evaluated NAFLD among HIV-infected persons with other known HIV-related metabolic complications but without chronic hepatitis C virus coinfection. The benchmark for NAFLD diagnosis is liver biopsy, but there are known limitations and risks associated with this procedure. A single percutaneous biopsy specimen reveals the degree of hepatic steatosis only within the anatomic liver area from which it was taken; multiple needle aspirations would be necessary to accurately evaluate steatotic changes in the whole liver [5]. Noninvasive diagnostic tools have been recommended for liver fat evaluation in HIV-infected persons, particularly for individuals without chronic viral hepatitis coinfection. Magnetic resonance spectroscopy is a very sensitive diagnostic tool [4], capable of detecting a liver fat content of 5%, yet it is seldom used outside of research settings. CT has been validated in HIV-seronegative liver donors, and its advantages include its wide availability, as well as its sensitivity and specificity [5] (similar to that seen with ultrasound but with the advantage that its diagnostic value is not as operator dependent).

We anticipated a positive association between NAFLD and increasing BMI. Characteristic features of our cohort include

Table 2. Clinical features and antiretroviral drug class exposure of HIV-infected patients, by nonalcoholic fatty liver disease (NAFLD) status.

Variable	Patients with NAFLD	Patients without NAFLD	Measure of association (95% CI)	*P*
CDC group C, no. (%) of patients				
Without AIDS	56 (72.73)	99 (70.21)		.695
With AIDS	21 (27.27)	42 (29.79)	OR, 0.88 (0.48–1.64)	
Duration of HIV infection, months	161.11 ± 60.58	155.29 ± 60.01	MD, −5.82 (−22.26 to 10.62)	.486
CD4 cell count nadir, cells/μL	199.7 ± 146.13	172.18 ± 133.13	MD, −27.52 (−65.86 to 10.83)	.159
CD4 cell count, cells/μL	585.85 ± 271.26	509.11 ± 284.87	MD, −76.73 (−160.04 to 6.58)	.071
HIV load, copies/mL	6681.30 ± 29767.26	8043.96 ± 29685.97	MD, 132.66 (−7676.45 to 10,401.77)	.766
HIV load, $\log_{10}$, copies/mL	2.15 ± 0.98	2.23 ± 1.03	MD, 0.08 (−0.23 to 0.39)	.602
Cumulative exposure to antiretroviral drug classes, months				
NRTI	124.16 ± 44.87	109.85 ± 49.16	MD, −14.31 (−27.48 to −1.15)	.033[a]
NNRTI	41.95 ± 30.43	35.54 ± 30.13	MD, −6.40 (−15.18 to 2.37)	.152
PI	61.34 ± 36.87	60 ± 39.73	MD, −1.34 (−12.62 to 9.93)	.815
Fusion inhibitor	8.4 ± 2.97	12.33 ± 15.72	MD, 3.93 (−11.81 to 19.67)	.596

NOTE. Data are mean ± SD, unless otherwise indicated. CDC, Centers for Disease Control and Prevention; MD, mean difference; NNRTI, nonnucleoside reverse-transcriptase inhibitor; NRTI, nucleoside reverse-transcriptase inhibitor; PI, protease inhibitor.

[a] Statistically significant.

the ability to use BMI values within normal weight ranges to predict metabolic abnormalities, as well as the overall high prevalence of LD. In the general population, the prevalence of obesity is 32.9% (National Health and Nutrition Examination Survey 2003–2004); in the NAFLD cohort, obesity prevalence had a range of 30%–100% [1]. In our total cohort, obesity prevalence was only 4.8%. Furthermore, the high prevalence of both lipoatrophy (found in 40.64% of the total cohort) and the "mixed form"—that is, lipoatrophy with lipoaccumulation (found in 37.90% of the total cohort)—were closely related to extensive antiretroviral exposure. In our cohort, NAFLD was an entity most commonly seen among HIV-infected, nonobese lipoatrophic men. We found associations between NAFLD and high-density lipoprotein cholesterol, HOMA-IR, waist circumference, and VAT. These associations led us to believe that there are etiologic links between NAFLD and HIV-associated body fat redistribution syndrome (i.e., LD) and other HIV-associated metabolic abnormalities, particularly serum lipid abnormalities and decreased insulin sensitivity, as evidenced by HOMA. We found a positive association between NAFLD and IR only after excluding persons with overt evidence of diabetes (by using HOMA-IR calculations). Overall serum triglyceride levels were not associated with NAFLD, but severe hypertriglyceridemia, defined as triglyceride level >4.56 mmol/L or fibrate use, was associated with NAFLD ($P = .006$; data not shown).

The association between NAFLD and serum ALT elevations is logical, given the histopathological findings characteristic of this disorder, including hepatocyte ballooning and necrosis [1]. Serum alkaline phosphatase elevation has been proposed as a marker of hepatic steatosis in HIV-infected patients without chronic hepatitis C virus and hepatitis B virus coinfection [29]. We could not undertake such an evaluation in this cohort, because alkaline phosphatase isoenzyme measurements were not available and the patient prevalence of osteopenia/osteoporosis was high (data not shown).

Cumulative NRTI exposure was an independent predictor of NAFLD, with each year of NRTI exposure increasing the risk of NAFLD by 11%. Most investigators believe that IR and the oxidative stress associated with lipid oxidative metabolism are probably significant contributors to NAFLD [30, 31]. We also know that HIV medications can promote IR and have been associated with fatty liver infiltration [13]. NRTI use (thymidine analogues in particular) are especially associated with the emergence of IR [32]. No associations between cumulative exposure to single antiretroviral drugs, single NRTIs, or commonly prescribed dual NRTI combinations (zidovudine + lamivudine or stavudine + didanosine) and NAFLD were found (data not shown; $P > .05$).

No associations were found between NAFLD and either standardized algorithms that predict cardiovascular events or nonclassic risk factors for CVD. Our interpretation is that NAFLD may represent a liver correlate of metabolic syndrome [33] and may itself be an indicator of metabolic abnormalities in HIV-infected people. Nevertheless, such metabolic alterations may not be sufficient to significantly increase cardiovascular risk in an otherwise healthy and relatively young population in which genetic factors and comorbidities (e.g., hypertension and smoking) are needed to produce clinical events. It is not surprising that there was no apparent association between NAFLD and coronary artery calcium, because NAFLD may represent a pro-

Table 3. Prior cardiovascular events and classic and nonclassic risk factors for cardiovascular disease, by nonalcoholic fatty liver disease (NAFLD) status.

Variable	Patients with NAFLD	Patients without NAFLD	Measure of association (95% CI)	*P*
Myocardial infarction			OR, 0.85 (0.21–3.49)	.820
Absent	80 (96.39)	136 (95.77)		
Present	3 (3.61)	6 (4.23)		
Framingham score, mean value ± SD	7.98 ± 6.32	7.06 ± 6.62	MD, −0.92 (−2.70 to 0.86)	.340
PROCAM score, mean value ± SD	7.23 ± 8.13	5.38 ± 6.58	MD, −1.85 (−3.81 to 0.11)	.065
Metabolic syndrome				
Absent	58 (71.60)	118 (83.10)		.046[a]
Present	23 (28.40)	24 (16.90)	OR, 1.95 (1.02–3.74)	
Coronary calcium score, mean value ± SD	44.18 ± 151.56	19.29 ± 52.25	MD, −24.89 (−52.77 to 3.00)	.080
Stratified coronary calcium score			OR, 1.83 (0.80–4.16)	.153
Normal	69 (84.15)	126 (90.65)		
Abnormal	13 (15.85)	13 (9.35)		

NOTE. Data are no. (%) of patients, unless otherwise indicated. MD, mean difference.

[a] Statistically significant.

cess that antedates by years the calcific atherosclerotic plaques detected by coronary CT and is a much earlier predictor of cardiovascular risk.

In addition to antiretroviral effects, HIV infection [34, 35] and dietary factors [36] may contribute to abnormal serum lipid profiles and IR. IR, in particular, may be the driving force for both metabolic syndrome and NAFLD. Both lean and lipoatrophic individuals have an increased amount of fat in the form of triglycerides in insulin-sensitive tissue, especially the liver and skeletal muscle [37].

Our study has several limitations. First, because of its cross-sectional and noncomparative nature, we cannot comment confidently on an observed sequence of events that led to NAFLD. Second, the diagnosis of NAFLD was based on a measured cutoff that did not allow us to describe liver fat accumulation between 5% and 30%. Also, histologic confirmation of NAFLD diagnoses by liver biopsy was not available to us, and because of CT's inability to discern between NAFLD and NASH, we were unable to identify individuals with the latter diagnosis, which can involve more severe and more progressive liver damage. Only 1 patient had an FIB-4 index >3.25, which suggests the presence of significant hepatic fibrosis (F3–F4). Our results could be interpreted to support either the assertion that few people with NAFLD have NASH or the assertion that the use of FIB-4 scores needs to undergo further validation in HIV-infected populations without chronic viral hepatitis coinfection.

Also, the lack of a control group disallowed potential identification of novel predictors of NAFLD. Caution is needed to generalize these results, given the high prevalence of LD in our cohort and the exclusion of patients with hepatitis coinfection.

It is clear that moderate-to-severe NAFLD among HIV-infected persons is an emergent clinical entity of significant prevalence that is likely to be multifactorial in etiology but is linked to risk factors known to be associated with other HIV-related metabolic and morphologic abnormalities (most notably, the prolonged use of NRTIs). We believe that this report supports the evolving assertion that the clinical identification of NAFLD is the hepatic equivalent of metabolic syndrome and that cumulative NRTI exposure is a chronic risk that can contribute to this syndrome, independent of NRTI effects on other anthropometric and metabolic parameters. Although NAFLD alone may not be sufficient to alter CVD burden, it may contribute to CVD risk in association with other classic or nonclassic CVD risk factors.

Table 4. Multivariate logistic regression analysis of independent variables associated with nonalcoholic fatty liver disease.

Variable	OR (95% CI)	*P*
ALT to AST ratio	4.59 (2.09–10.08)	<.001
Male sex	2.49 (1.07–5.81)	<.001
Cumulative NRTI	1.12 (1.03–1.22)	<.001
Waist circumference	1.07 (1.03–1.11)	<.001

NOTE. ALT, alanine aminotransferase; AST, aspartate aminotransferase; NRTI, nucleoside reverse-transcriptase inhibitor.

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