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Nonalcoholic Fatty Liver Disease and the Development of Metabolic Comorbid Conditions in Patients With Human Immunodeficiency Virus Infection

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Background. Cardiovascular and liver disease are main causes of death in people with human immunodeficiency virus (HIV) (PWH). In HIV-uninfected patients, nonalcoholic fatty liver disease (NAFLD) is associated with incident metabolic complications. We investigated the effect of NAFLD on development of metabolic comorbid conditions in PWH.

Methods. We included PWH undergoing a screening program for NAFLD using transient elastography. NAFLD was defined as a controlled attenuation parameter ≥248 dB/m with exclusion of other liver diseases. Incident diabetes, hypertension, dyslipidemia, and chronic kidney disease were investigated using survival analysis and Cox proportional hazards.

Results. The study included 485 HIV-monoinfected patients. During a median follow-up of 40.1 months (interquartile range, 26.5–50.7 months), patients with NAFLD had higher incidences of diabetes (4.74 [95% confidence interval, 3.09–7.27] vs 0.87 [.42–1.83] per 100 person-years) and dyslipidemia (8.16 [5.42–12.27] vs 3.99 [2.67–5.95] per 100 person-years) than those without NAFLD. With multivariable analysis, NAFLD was an independent predictor of diabetes (adjusted hazard ratio, 5.13; 95% confidence interval, 2.14–12.31) and dyslipidemia (2.35; 1.34–4.14) development.

Conclusions. HIV-monoinfected patients with NAFLD are at higher risk of incident diabetes and dyslipidemia. Early referral strategies and timely management of metabolic risk may improve outcomes.

Keywords. diabetes; dyslipidemia; transient elastography; controlled attenuation parameter; liver fibrosis.

Nonalcoholic fatty liver disease (NAFLD), defined as fat accumulation in the liver in the absence of excessive alcohol consumption, is an epidemic entity affecting approximately 25% of the world's population [1]. In the general population, NAFLD is not only associated with liver-related disease and death related to cirrhosis and hepatocellular carcinoma, but it has also been linked with increased mortality rates associated with extrahepatic diseases, particularly cardiovascular disease [2]. Moreover, NAFLD is strongly associated with the metabolic syndrome and has been linked with increased risks of diabetes, hypertension, dyslipidemia, and chronic kidney disease (CKD) [3–5].

NAFLD is expected to increasingly comprise the burden of liver disease in people with HIV (PWH) owing to the widespread availability and effectiveness of direct-acting antivirals for the treatment of chronic hepatitis C. The prevalence of NAFLD in HIV monoinfection is currently estimated to be ≥35% [6]. The prolonged exposure to HIV infection and antiretroviral therapy (ART), as well as increased intestinal dysbiosis and bacterial translocation, are thought to contribute to the development of NAFLD in PWH, in addition to risk factors shared with the general population, including obesity, insulin resistance, and dyslipidemia [7]. Importantly, the classic metabolic risk factors for NAFLD are more frequent in PWH. In HIV-infected men, a 4 times higher prevalence of type 2 diabetes mellitus (T2DM) has been described, compared with that in HIV-uninfected men [8]. Dyslipidemia is also common. owing to both HIV chronic infection and lifelong use of ART [9]. Hypertension and a higher cardiovascular risk have also been consistently reported [10, 11]. HIV-infected patients are also at higher risk for CKD [12].

Given that HIV infection and NAFLD independently increase the risk of metabolic comorbid conditions, the aim of the study was to investigate the effect of NAFLD on development of T2DM, hypertension, dyslipidemia, and CKD in a

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cohort of PWH, by means of transient elastography with associated measurement of controlled attenuation parameter (CAP) [13]. This diagnostic tool for NAFLD and associated liver fibrosis has been validated against liver biopsy results in both HIV-uninfected and HIV-infected patients [14–16].

PATIENTS AND METHODS

Study Design and Population

We conducted a retrospective analysis of the LIVEr in HIV (LIVEHIV), an established prospective cohort of PWH followed up at the McGill University Health Centre. From September 2013 to August 2018, a total of 798 PWH were enrolled at the university-based clinic of the Chronic Viral Illness Service. Patients are regularly followed up by their treating physician for their HIV care every 6 months. Moreover, they are consecutively screened for liver disease; the screening includes hepatitis C virus (HCV) and hepatitis B virus serology, the Alcohol Use Disorders Identification Test (AUDIT-C) questionnaire, and annual transient elastography with CAP measurement using the FibroScan (EchoSens).

For the present study, we included all consecutive patients who met the following criteria: (1) age ≥18 years; (2) HIV infection, as documented by positive enzyme-linked immunosorbent assay with Western blot confirmation; (3) valid transient elastography results with available CAP measurement; and (4) availability of relevant biochemical and physiological parameters. The criteria for exclusion were (1) positivity for HCV antibody or hepatitis B surface antigen; (2) hazardous alcohol intake (AUDIT-C score ≥7); (3) lack of longitudinal follow-up (≥6 months); (4) contraindications (pacemaker or pregnancy), failure of (no successful measurements after 10 attempts), or unreliable transient elastography examination; and (5) decompensated cirrhosis or hepatocellular carcinoma at enrollment.

Ethics

The study was approved by the Research Ethics Board of the Research Institute of the McGill University Health Centre (code 14-182-BMD), in accordance with the declaration of Helsinki. Participants in the study provided written informed consent before enrollment.

Clinical and Biological Parameters

Relevant data were collected during routine follow-up clinic appointments and within 3 months from the transient elastography examination. Demographic and clinical information included age, sex, race/ethnicity, body mass index (BMI), risk factors for HIV infection, time since HIV diagnosis, and exposure to ART. ART drugs were classified as protease inhibitors (PIs), nonnucleoside reverse-transcriptase inhibitors, nucleoside/nucleotide reverse-transcriptase inhibitors (NRTIs), and integrase inhibitors. Laboratory data included aspartate aminotransferase, alanine aminotransferase, platelet count, total cholesterol, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, triglyceride levels, CD4 cell count and nadir, and serum creatinine, bilirubin, and albumin levels. In addition, metabolic comorbid conditions (history of diabetes, hypertension, and dyslipidemia) were documented.

Transient Elastography Examination with CAP Measurement

Transient elastography with CAP measurement was performed in patients who had fasted for ≥3 hours by 2 experienced operators (who had each performed >500 examinations before the study). The M probe was used routinely, with XL probe used if the M probe failed, or the patient had a BMI >30 kg/m^2. Given data published in 2019 on lack of influence of probe type on liver stiffness measurement, the same cutoff value was used for M and XL probes to define suspected significant liver fibrosis [15]. The following criteria were applied to define the result of transient elastography examination as reliable: ≥10 validated measures and an interquartile range ≤30% of the median [17]. Suspected significant liver fibrosis (stages F2–F4) and cirrhosis (stage F4) were defined as liver stiffness measurements of ≥7.1 or ≥13 kPa, respectively [14, 16, 18]. As previously reported in the setting of HIV infection, any-grade steatosis (involving >10% of hepatocytes) and severe steatosis (involving >66% of hepatocytes) were defined as CAP ≥248 and >292 dB/m, respectively [19, 20].

Exposure and Outcome Measures

The main exposure of this study was the diagnosis of NAFLD at baseline (time zero). Any-grade hepatic steatosis (CAP ≥248 dB/m) was used to diagnose NAFLD [19, 20]. As secondary exposure, we also explored suspected significant liver fibrosis [14, 20]. The main study outcomes were the development of metabolic comorbid conditions, as defined below. Hypertension and dyslipidemia were diagnosed according to Canadian Cardiovascular Society guidelines [21, 22]. T2DM was defined as a glycated hemoglobin A_{1c} level ≥6.5%, or as previously diagnosed by an endocrinologist or treating physician [23]. CKD was defined as an estimated glomerular filtration rate <60 mL/min/1.73 m^2 (category G3), as calculated using serum creatinine according to the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) formula [24] or albuminuria category A2 or above obtained on 2 occasions ≥3 months apart, according to the KDIGO (Kidney Disease Improving Global Outcomes) guidelines [25]. Albuminuria was measured using the spot urine microalbumin-creatinine ratio and/or 24-hour protein urine collection. As secondary outcome, we reported the all-cause mortality rate, which was collected by means of dedicated outcome measures form.

Statistical Analysis

Baseline (time zero) corresponded to the first visit after 1 September 2013 when transient elastography examination

was performed. We compared characteristics of participants at baseline by exposure status, using the Student *t* test for continuous variables and Pearson χ^2 or Fisher exact tests for categorical variables. Patients were observed until August 2018 or were censored either when they died or at their last clinic visit. T2DM, hypertension, dyslipidemia, and CKD were recorded as binary outcomes. Participants with the respective outcomes of interest at baseline were excluded from the subgroup calculation of incidence rate, time-to-failure plots, and hazard ratios (HRs). Incidence rates of T2DM, hypertension, dyslipidemia and CKD were estimated by dividing the number of participants developing the outcome by the number of person-years (PY) of follow-up. Poisson count models were used to calculate confidence intervals (CIs) for incidence rates. Kaplan-Meier plots and log-rank tests were used to illustrate time to metabolic outcomes by presence or absence of NAFLD and by the presence or absence of suspected significant liver fibrosis.

Multivariable time-dependent Cox regression models were constructed to assess predictors of T2DM, hypertension, dyslipidemia, and CKD. Results were reported as adjusted HRs (aHRs) with 95% CIs. Robust variance estimation was used in all Cox regression analyses to account for the correlation of data contributed by the same participant at multiple visits. Multivariable models included covariates that were determined a priori to be clinically important, namely, age, sex, and race/ethnicity. Given the known effect of PIs on lipid metabolism, models were also adjusted for current use of PIs, except for the CKD model, which was adjusted for current use of NRTIs, given the known effect of tenofovir on kidney function, and for the T2DM model, given the smaller number of outcomes [12, 26, 27]. Moreover, all models were adjusted for either preexisting hypertension (for the outcomes of T2DM and dyslipidemia) or T2DM (for the outcomes of hypertension and CKD). A complete case analysis was used for the multivariable models, and the percentage of missing data was <10%, unless specified. A 2-tailed α value of .05 was used as a threshold to determine statistical significance. Statistical analyses were performed using SAS (SAS Institute) and Stata 13.1 (StataCorp) software.

RESULTS

After application of exclusion criteria (Figure 1), 485 patients were included in this study. At baseline, the prevalence of NAFLD was 38.1%. Severe hepatic steatosis affected 81 (16.7%) patients. Suspected significant liver fibrosis and cirrhosis affected 72 (14.8%) and 12 (2.5%) of the patients, respectively. Overall, 57 (11.9%) and 102 (21%) patients were exposed to didanosine and stavudine, respectively. Table 1 reports the main demographic, clinical, biochemical and immunovirological characteristics of the study population by NAFLD status at baseline. Patients with NAFLD were older, more likely to be white, and had higher BMIs. Moreover, they had longer time since HIV diagnosis. Finally, they had higher prevalence of hypertension, lower high-density lipoprotein cholesterol, levels, higher triglyceride, glucose, alanine aminotransferase, aspartate aminotransferase, and albumin levels, and higher liver stiffness measurement. At baseline, patients with NAFLD had higher prevalences of hypertension and dyslipidemia (Table 1), and those with suspected significant liver fibrosis had higher prevalence of all metabolic comorbid conditions (Figure 2).

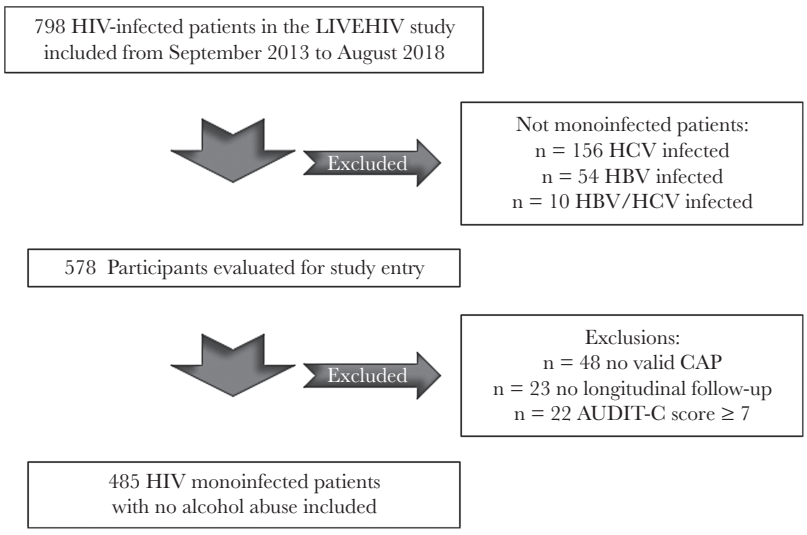

Figure 1. Flow chart displaying the selection of study participants. Liver stiffness measurements with transient elastography were considered reliable if the ratio of the interquartile range to the median of the 10 measures was ≤30%. Abbreviations: AUDIT-C, Alcohol Use Disorders Identification Test; CAP, controlled attenuation parameter; HBV, hepatitis B virus; HCV, hepatitis C virus; HIV, human immunodeficiency virus; LIVEHIV, LIVEr in HIV.

Table 1. Characteristics of Human Immunodeficiency Virus–Monoinfected Patients at Baseline by Nonalcoholic Fatty Liver Disease Status

Characteristic	Whole cohort (n = 485)	NAFLD (n = 185)	No NAFLD (n = 300)	*P* Value[a]
Age, mean (SD), y	49.5 (10.9)	51.0 (9.7)	48.7 (11.6)	.01
Male sex, no. (%)	367 (75.7)	146 (78.9)	221 (73.7)	.19
Ethnicity, no. (%)[b]				
White	226 (47.9)	101 (55.5)	125 (43.1)	.01
Black	165 (35.0)	47 (25.8)	118 (40.7)	
Hispanic	50 (10.6)	17 (9.3)	33 (11.4)	
Asian	16 (3.4)	9 (5.0)	7 (2.4)	
Other	15 (3.1)	8 (4.4)	7 (2.4)	
BMI,[c] mean (SD), kg/m^2	26.5 (5.1)	28.1 (4.5)	25.6 (5.2)	<.001
AUDIT-C score, mean (SD)	1.3 (1.7)	1.3 (1.7)	1.4 (1.7)	.44
Active tobacco smoker, no. (%)	75 (15.5)	29 (15.7)	46 (15.3)	.74
Hypertension, no. (%)	86 (17.7)	43 (23.2)	43 (14.3)	.003
T2DM, no. (%)	68 (14.0)	32 (17.3)	36 (12.0)	.08
MSM, no. (%)	165 (34.8)	66 (37.7)	99 (33.1)	.23
Active IDU, no. (%)	17 (3.6)	7 (4.0)	10 (3.3)	.72
Time since HIV diagnosis, mean (SD), y	13.3 (8.4)	14.7 (8.4)	12.5 (8.3)	.003
CD4 cell count, mean (SD), cells/μL	672.2 (278.5)	668.8 (271.6)	674.3 (282.9)	.91
Undetectable HIV load (≤50 copies/mL), no. (%)	421 (86.8)	157 (84.9)	259 (86.3)	.65
Current ART regimen, no. (%)				
NRTI	411 (84.7)	148 (80.0)	263 (87.7)	.02
NNRTI	170 (35.0)	65 (35.1)	105 (35.0)	.96
PI	173 (35.7)	74 (40.0)	99 (33.0)	.13
Integrase inhibitors	178 (36.7)	65 (35.1)	113 (37.7)	.59
INR, mean (SD)	1.08 (0.37)	1.09 (0.46)	1.08 (0.30)	.82
Platelet count, mean (SD),10^9/L	209.6 (59.7)	215.5 (58.7)	206.1 (60.2)	.10
Laboratory values, mean (SD)				
Total cholesterol, mmol/L	4.74 (1.08)	4.81 (1.05)	4.70 (1.10)	.34
LDL cholesterol, mmol/L	2.78 (0.89)	2.77 (0.93)	2.79 (0.87)	.79
HDL cholesterol, mmol/L	1.23 (0.42)	1.12 (0.34)	1.30 (0.46)	<.001
Triglycerides, mmol/L	1.84 (1.74)	2.37 (2.28)	1.50 (1.17)	<.001
Glucose, mmol/L	5.5 (1.2)	5.7 (1.4)	5.3 (1.0)	.04
Creatinine, μmol/L	85.4 (22.4)	84.8 (20.2)	85.9 (23.6)	.64
ALT, IU/L	31.1 (25.0)	37.7 (34.4)	27.2 (16.1)	<.001
AST, IU/L	26.6 (14.5)	29.3 (20.2)	25.1 (9.4)	<.001
Total bilirubin, μmol/L	13.5 (11.7)	14.7 (13.3)	12.7 (10.6)	.16
Albumin, g/L	42.2 (3.5)	43.0 (3.3)	41.7 (3.6)	<.001
Liver stiffness measurement, mean (SD), kPa	5.60 (3.72)	6.75 (5.45)	4.89 (1.67)	<.001
CAP, mean (SD), dB/m	237.4 (57.9)	293.7 (38.0)	202.1 (36.2)	<.001

Abbreviations; ALT, alanine aminotransferase; ART, antiretroviral therapy; AST, aspartate aminotransferase; AUDIT-C, Alcohol Use Disorders Identification Test; BMI, body mass index; CAP, controlled attenuation parameter; HDL, high-density lipoprotein cholesterol; ; HIV, human immunodeficiency virus; IDU, injection drug use; IU, international units; INR, international normalized ratio; LDL, low-density lipoprotein cholesterol; MSM, men who have sex with men; NAFLD, nonalcoholic fatty liver disease; NNRTI, nonnucleoside reverse-transcriptase inhibitors; NRTI, nucleoside reverse-transcriptase inhibitors; PI, protease inhibitor; SD, standard deviation; T2DM, type 2 diabetes mellitus.

[a]*P* values based on Student *t* or χ^2 tests, for comparison between patients with or without NAFLD.

[b]Data on race/ethnicity were available for 472 patients.

[c]Data on BMI were available for 376 patients.

Incidence and Predictors of Metabolic Comorbid Conditions

Patients were followed up for a median of 40.1 months (interquartile range, 26.5–50.7 months). There were 68, 86, 190, and 84 patients with T2DM, hypertension, dyslipidemia, and CKD, respectively, who were excluded from the longitudinal analysis for having the outcome at baseline. Overall, incidence rates for T2DM, hypertension, dyslipidemia, and CKD were 2.2 (95% CI, 1.6–3.3), 4.2 (3.2–5.5), 5.3 (4.0–7.1), and 2.7 (1.9–3.8) per 100 PY, respectively. Tables 2 and 3 shows incidence rates for metabolic comorbid conditions by NAFLD and suspected significant liver fibrosis status. Patients with NAFLD had a higher incidence rate of T2DM and dyslipidemia than those without NAFLD (Figures 3A and 3C). There was also a tendency for a higher incidence rate of hypertension (Figure 3B), whereas there was no difference for CKD (Figure 3D).

Patients with suspected significant liver fibrosis had a higher incidence rate of T2DM, but no difference was found for hypertension, dyslipidemia, or CKD (Figure 4). Supplementary Table

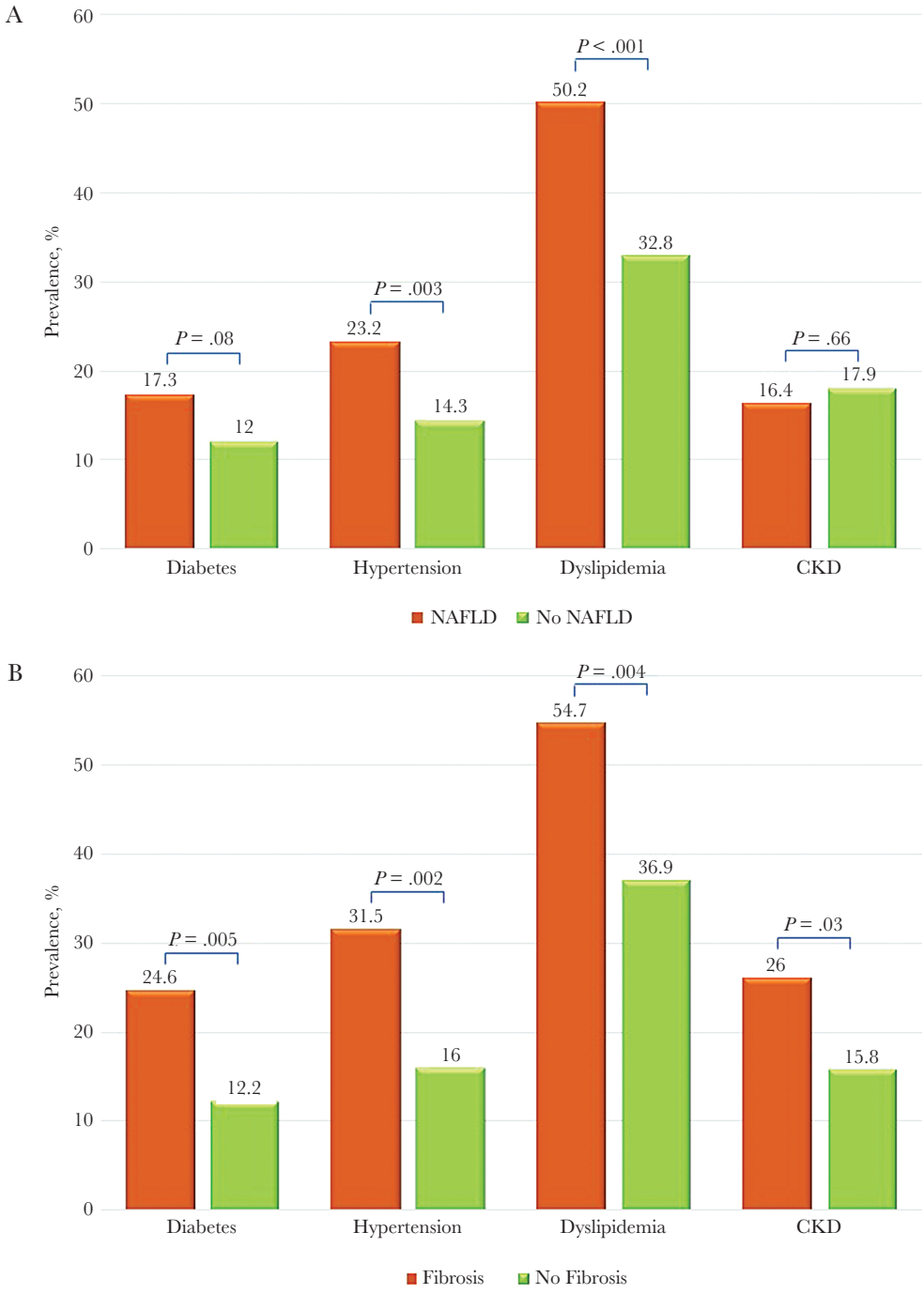

Figure 2. Prevalence of diabetes, dyslipidemia, hypertension and chronic kidney disease (CKD) according to nonalcoholic fatty liver disease (NAFLD) (*A*) or fibrosis (*B*) status at baseline.

1 shows the characteristics of patients who developed metabolic comorbid conditions during the follow-up period. Compared with the whole cohort, patients in whom T2DM developed were more likely to be female and black and had higher BMIs and less likely to be men who have sex with men and to be taking NRTIs. Patients in whom dyslipidemia developed were more likely to be taking NRTIs. Those in whom CKD developed were more likely to be diabetic and to have higher liver stiffness measurement.

During the follow-up period, there were a total of 8 deaths, of which 3 were related to nonliver cancer, 2 to infections, and 2 were of unknown cause. The corresponding incidence rate was 0.4 (95% CI, .2–.9) per 100 PY.

After adjustments, NAFLD and preexisting hypertension were independent predictors of T2DM development (Table 4). Incident hypertension was independently predicted by older age. NAFLD and current use of PIs as ART regimen

Table 2. Incidence Rates of Metabolic Complications During Follow-up by Nonalcoholic Fatty Liver Disease Status[a]

	NAFLD (n = 185)		No NAFLD (n = 300)	
Complication	Patients, No.	Incidence Rate per 100 PY (95% CI)	Patients, No.	Incidence Rate per 100 PY (95% CI)
T2DM	21	4.74 (3.09–7.27)[b]	7	0.87 (0.42–1.83)[b]
Hypertension	22	5.25 (3.45–7.98)	28	3.59 (2.48–5.21)
Dyslipidemia	23	8.16 (5.42–12.27)[c]	24	3.99 (2.67–5.95)[c]
CKD	13	2.82 (1.64–4.86)	20	2.67 (1.72–4.14)

Abbreviations: CI, confidence interval; CKD, chronic kidney disease; NAFLD, nonalcoholic fatty liver disease; PY, person-years; T2DM, type 2 diabetes mellitus.

[a]Rates shown are after exclusion of patients with the outcome of interest at baseline.

[b]$P < .001$ (log-rank test) for comparison between patients with or without NAFLD.

[c]$P < .05$ (log-rank test) for comparison between patients with or without NAFLD.

were independent predictors of development of dyslipidemia. Incident CKD was independently predicted by older age and preexisting T2DM. We also conducted a multivariable analysis including suspected significant liver fibrosis as exposure. After adjustment for age (per 10 years; aHR, 1.14; 95% CI, .78–1.67), male sex (0.84; .35–1.97), and black ethnicity (2.15; .93–4.97), the independent predictors of T2DM development were suspected significant liver fibrosis (2.71; 1.11–6.61; $P = .03$) and preexisting hypertension (4.22; 1.81–9.80; $P = .001$). There was no effect of suspected significant liver fibrosis on development of other metabolic comorbid conditions (results not shown). We also conducted a sensitivity analysis for the subgroup of patients with available BMI. The relative univariable Cox regression analysis showed an HR of 1.20 (95% CI, .99–1.45; $P = .05$) for incident T2DM, 1.18 (.99–1.41; $P = .06$) for incident hypertension, 1.01 (.87–1.17; $P = .91$) for incident dyslipidemia, and 1.24 (.99–1.55; $P = .06$) for incident CKD.

DISCUSSION

The current study, based on a cohort of HIV-infected patients consecutively screened for liver disease, shows that NAFLD predicts the development of important metabolic comorbid conditions, including T2DM and dyslipidemia. Furthermore, NAFLD severity, represented by suspected significant liver fibrosis, predicts development of T2DM. Our findings mirror what has already been reported in the general population and suggest that fatty liver is a central barometer of metabolic health in PWH as well [28]. To the best of our knowledge, ours is the first cohort study linking NAFLD with the development of important metabolic comorbid conditions.

NAFLD is increasingly recognized as the most frequent liver disease in people aging with HIV [13, 29–32]. One meta-analysis situates the prevalence of NAFLD in those with HIV monoinfection at 35% [6], higher than the 25% global prevalence reported for the general population [1]. In the same study, the pooled prevalence of significant liver fibrosis was 22%. One reason for this excess may be that PWH present with particularly high frequency of metabolic conditions. A higher incidence of T2DM in the HIV-infected population has been well described, though the role of the liver in this process has not been investigated. A longitudinal study with a median follow-up of 4 years reported a cumulative incidence of T2DM of 10% in PWH, compared to 3% in uninfected controls [8]. A 2018 meta-analysis reported a pooled incidence rate for T2DM of 13.7 (95% CI, 13–20) per 1000 PY of follow-up [33]. Frequent hypertension and a higher cardiovascular risk have also been consistently reported [10, 11]. Several virological and ART-related factors have been implicated in the pathophysiological mechanism of T2DM and hypertension in HIV infection, including chronic inflammation, immune reconstitution, and lipodystrophy [34]. Dyslipidemia is also common, owing to both HIV chronic infection and lifelong use of ART, particularly PIs [9]. CKD is a frequent complication of HIV infection, occurring in 3.5%–48.5% of the patients, owing to HIV infection itself and as a consequence of ART, such as tenofovir disoproxil fumarate [12, 27].

Table 3. Incidence Rates of Metabolic Complications During Follow-up by Fibrosis Status[a]

	Fibrosis (n = 72)		No Fibrosis (n = 413)	
Complication	Patients, No.	Incidence Rate per 100 PY (95% CI)	Patients, No.	Incidence Rate per 100 PY (95% CI)
T2DM	8	5.01 (2.50–10.01)[b]	20	1.85 (1.19–2.86)[b]
Hypertension	9	5.97 (3.11–11.48)	41	3.92 (2.88–5.32)
Dyslipidemia	5	5.00 (2.08–12.01)	42	5.37 (3.97–7.27)
CKD	7	4.20 (2.00–8.81)	26	2.50 (1.70–3.67)

Abbreviations: CI, confidence interval; CKD, chronic kidney disease; PY, person-years; T2DM, type 2 diabetes mellitus.

[a]Rates shown are after exclusion of patients with the outcome of interest at baseline.

[b]$P < .05$ (log-rank test) for comparison between patients with or without fibrosis.

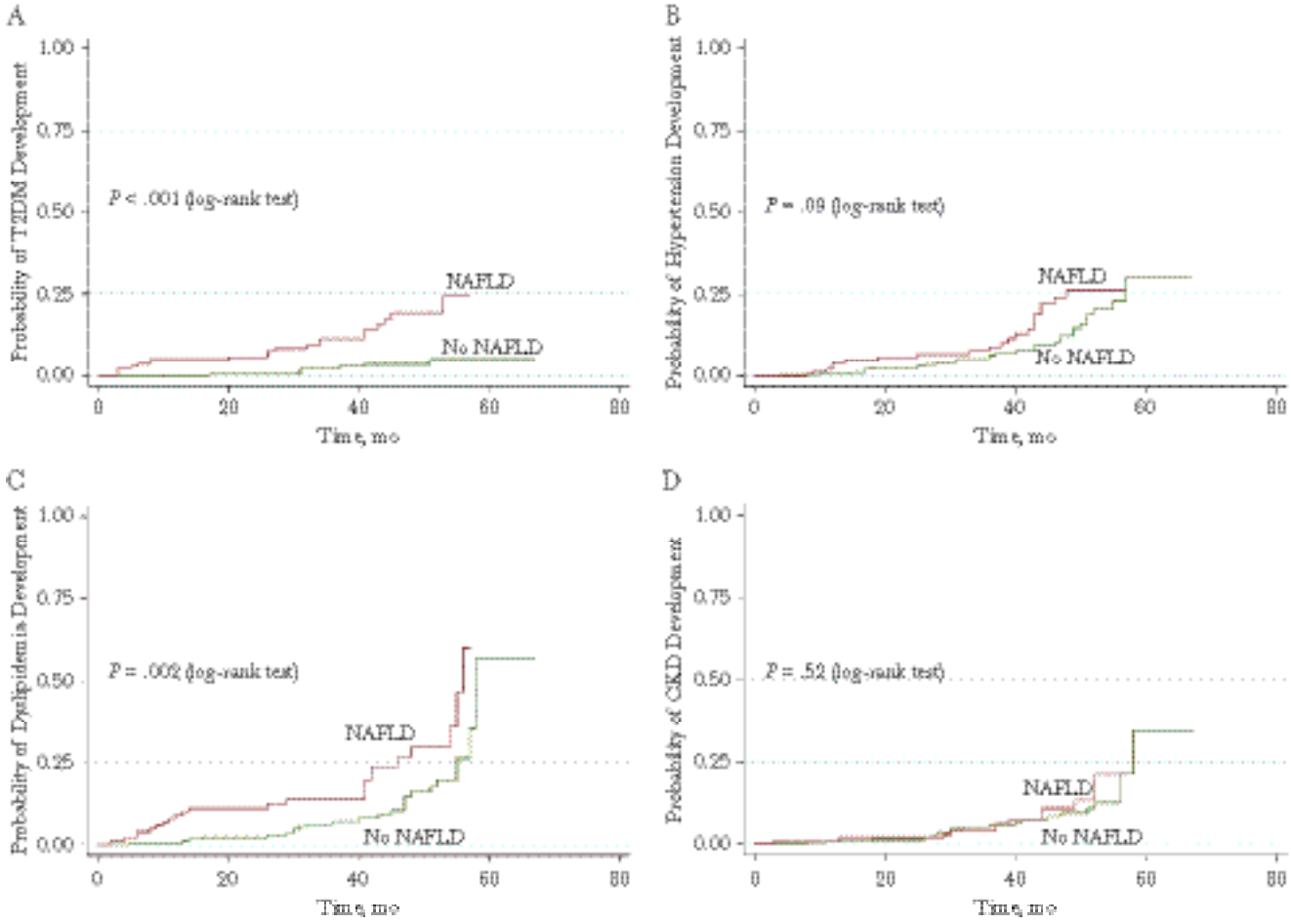

Figure 3. Survival curves for incidence of type 2 diabetes mellitus (T2DM) (*A*), hypertension (*B*), dyslipidemia (*C*), and chronic kidney disease (CKD) (*D*) by nonalcoholic fatty liver disease (NAFLD) status. *P* Value (log-rank test) for comparison between patients with or without NAFLD.

Our study provides novel longitudinal data on the increased risk of T2DM, hypertension and dyslipidemia in PWH with NAFLD. This is consistent with findings in the general population with NAFLD. A large meta-analysis, including 19 observational studies and 296 439 individuals followed up for ≥1 year, reported a 2-fold increased risk of incident T2DM in patients with NAFLD [4]. A community study including 3869 NAFLD subjects and 15 209 controls followed up for a median of 7 years found that a subject with NAFLD and without T2DM, hypertension or dyslipidemia was >2 times more likely (relative risk, 2.62; 95% CI, 2.31-2.96) to develop ≥1 of these comorbid conditions than an age- and sex-matched control [35].

In our study, we found that PWH with NAFLD were more likely to acquire T2DM and dyslipidemia, with aHRs of 5.13 (95% CI, 2.14–12.31) and 2.35 (1.34–4.14), respectively. Moreover, suspected significant liver fibrosis was an independent predictor of T2DM development, with an aHR of 2.71 (95% CI, 1.11–6.61). A recent cross-sectional study of factors associated with liver fibrosis and steatosis in patients with HIV monoinfection demonstrated an association of T2DM with liver fibrosis (odds ratio, 3.78; 95% CI, 1.48–9.68) [36]. Our finding suggests a possible link between the progression of NAFLD-associated liver disease and insulin resistance in the context of HIV infection. Current use of PIs was an independent predictor of dyslipidemia development. This class of ART is associated with a less favorable lipid profile, in particular elevated triglyceride and total cholesterol levels, especially when boosted with ritonavir [37]. We also observed a trend for NAFLD to predict development of hypertension. Conversely, we did not observe any link between NAFLD and incident CKD. In the general population, a cohort study of 41 430 adults in Korea reported an aHR for the development of CKD of 1.22 (95% CI, 1.04–1.43) for sonographically diagnosed NAFLD. We speculate that the pathogenesis of CKD in PWH may be more complex, and HIV-related factors may overcome the effect of NAFLD in this setting [3].

Liver and cardiovascular disease are the primary non–AIDS-related causes of disease and death among PWH [38]. Although HCV coinfection has driven much of the liver-related mortality in the past, the implementation of effective

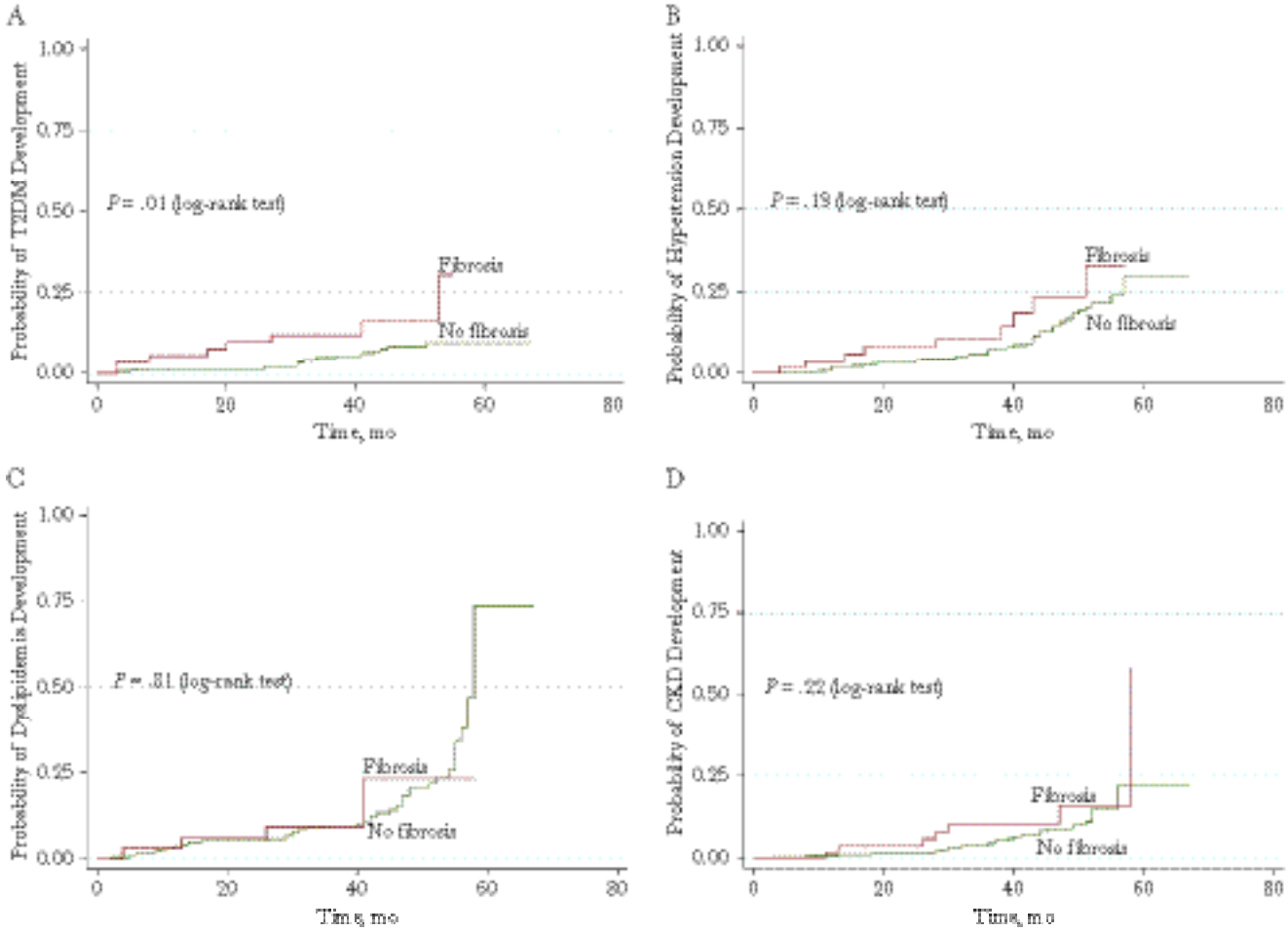

Figure 4. Survival curves for incidence of type 2 diabetes mellitus (T2DM) (*A*), hypertension (*B*), dyslipidemia (*C*), and chronic kidney disease (CKD) (*D*) by fibrosis status.

direct antiviral-agents has inverted this trend. The increasing burden of NAFLD in the HIV-infected population, combined with our findings of its impact on metabolic comorbid conditions, indicate that PWH with NAFLD may be at particularly high risk of cardiovascular-associated disease and death. Indeed, cardiovascular events are the leading cause of death in patients with NAFLD, and PWH have higher cardiovascular risk than the general population [39–41]. This is thought to reflect the effects of systemic inflammation and endothelial dysfunction, as well as the disproportionate presence of the established risk factors of insulin resistance and dyslipidemia. In addition, the effect of NAFLD on long-term outcomes in the HIV-infected population is still not completely understood. Importantly, in HIV-uninfected patients with NAFLD, the severity of liver fibrosis affects not only liver-related, but also all-cause mortality rates [39]. As such, guidelines recommend cardiovascular risk stratification for all patients with NAFLD, particularly those with liver fibrosis [42, 43]. Management of NAFLD in HIV infection may include treating modifiable metabolic risk factors, diet and exercise, and nutritional supplements as well as optimizing HIV-related factors and ART [44–46].

The main strength of our study is the longitudinal design, able to capture dynamics over time and providing novel data regarding the effects of NAFLD on development of metabolic comorbid conditions in PWH. Moreover, we included only consecutive patients in an ongoing screening program for liver disease at a single center.

We also acknowledge several limitations of our study. First, the study was retrospective. Second, we did not have more robust diagnostic tools to diagnose hepatic steatosis, such as liver biopsy or magnetic resonance imaging [14, 32]. However, it would be costly and time consuming to use these approaches in a large cohort. Moreover, transient elastography has not been widely validated against liver biopsy in the context of HIV infection. Although a CAP cutoff of ≥248 dB/m has a good sensitivity for detection of any grade steatosis, higher cutoff values have been proposed and its accuracy for grading steatosis is lower [14]. Third, we could not correlate our findings with actual cardiovascular outcomes. Fourth, given the limited number of

Table 4. Multivariable Analysis of Predictors of Development of Type 2 Diabetes Mellitus, Hypertension, Dyslipidemia, and Chronic Kidney Disease[a]

Variable	aHR (95% CI)	*P* Value
Type 2 diabetes mellitus (n = 417)		
Age (per 10 y)	1.15 (.76–1.72)	.50
Male sex (yes vs no)	0.71 (.30–1.71)	.45
Black race (yes vs no)	1.90 (.83–4.31)	.13
Hypertension (yes vs no)	3.69 (1.56–8.74)	.003
NAFLD (yes vs no)	5.13 (2.14–12.31)	<.001
Hypertension (n = 399)		
Age (per 10 y)	1.62 (1.25–2.10)	<.001
Male sex (yes vs no)	0.93 (.44–1.95)	.85
Black race (yes vs no)	1.62 (.88–2.98)	.12
Current PI use (yes vs no)	0.78 (.45–1.35)	.38
T2DM (yes vs no)	1.53 (.71–3.32)	.28
NAFLD (yes vs no)	1.64 (.93–2.88)	.09
Dyslipidemia (n = 295)		
Age (per 10 y)	1.05 (.79–1.40)	.72
Male sex (yes vs no)	1.19 (.57–2.48)	.64
Black ethnicity (yes vs no)	0.59 (.32–1.10)	.09
Current PI use (yes vs no)	2.08 (1.17–3.70)	.01
Hypertension (yes vs no)	1.02 (.46–2.26)	.96
NAFLD (yes vs no)	2.35 (1.34–4.14)	.003
Chronic kidney disease (n = 401)		
Age (per 10 y)	1.99 (1.46–2.70)	<.001
Male sex (yes vs no)	1.18 (.51–2.80)	.70
Black ethnicity (yes vs no)	1.36 (.64–2.87)	.43
Current NRTI use (yes vs no)	0.54 (.25–1.13)	.10
T2DM (yes vs no)	2.21 (1.04–4.69)	.04
NAFLD (yes vs no)	1.16 (.56–2.38)	.69

Abbreviations: aHR, adjusted hazard ratio; CI, confidence interval; NAFLD, nonalcoholic fatty liver disease; NRTI, nucleoside reverse-transcriptase inhibitors; PI, protease inhibitor; T2DM, type 2 diabetes mellitus.

[a]The aHRs and 95% CIs are presented for all variables analyzed in multivariable Cox regression analysis.

outcomes, we were unable to study the influence of specific ART drugs and to include all metabolic comorbid conditions in the multivariable models. Fifth, BMI was missing for 22% of cases, so we could not account for its effect on multivariable models, although we found a tendency to predict incident T2DM, hypertension and CKD in univariable model. BMI might be an easy predictor to classify patients at risk of metabolic complications in clinical practice. Sixth, the observational design of this study does not allow for conclusions regarding causality. Specifically, our findings do not necessarily implicate NAFLD as a cause of diabetes. Finally, shared risk factors could contribute to NAFLD, T2DM, and dyslipidemia, including medication effects, genetics, type of diet, and other lifestyle factors.

In conclusion, our study suggests that NAFLD is an independent predictor for the development of important metabolic comorbid conditions in PWH. This finding configures NAFLD as a barometer for metabolic health in the setting of HIV infection. Metabolic comorbid conditions might perpetuate the NAFLD spectrum and contribute to increased cardiovascular risk in PWH. In the context of HIV infection, patients with NAFLD should be closely monitored for the development of metabolic outcomes and modifiable risks be managed in a timely fashion. Our findings might be considered a further argument to advocate for screening for NAFLD in PWH, as it is the case for patients with diabetes [47]. Indeed, the European AIDS Clinical Society guidelines recommended screening for NAFLD in PWH with metabolic syndrome, and expansion of these criteria to PWH with any metabolic comorbid condition has been proposed [48, 49]. The implications of our findings on long-term cardiovascular outcomes should be investigated in future studies.

Supplementary Data

Supplementary materials are available at *The Journal of Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

Author contributions. Study conception, study design, and first draft of the manuscript: T. K. and G. S. Data collection and interpretation: T. K., R. S. P., N. K., J. F., G. G., B. L., M. B. K., P. W., M. D, P. G., and G. S. Statistical analysis: T. K., M.M., and G. S. All authors approved the final version of the article.

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References

1. Younossi ZM, Koenig AB, Abdelatif D, Fazel Y, Henry L, Wymer M. Global epidemiology of nonalcoholic fatty liver disease-meta-analytic assessment of prevalence, incidence, and outcomes. Hepatology **2016**; 64:73–84.
2. Adams LA, Anstee QM, Tilg H, Targher G. Non-alcoholic fatty liver disease and its relationship with cardiovascular disease and other extrahepatic diseases. Gut **2017**; 66:1138–53.
3. Sinn DH, Kang D, Jang HR, et al. Development of chronic kidney disease in patients with non-alcoholic fatty liver disease: a cohort study. J Hepatol **2017**; 67:1274–80.
4. Mantovani A, Byrne CD, Bonora E, Targher G. Nonalcoholic fatty liver disease and risk of incident type 2 diabetes: a meta-analysis. Diabetes Care **2018**; 41:372–82.
5. Leake I. PD-1 inhibitors for oesophageal cancer. Nat Rev Gastroenterol Hepatol **2019**; 16:706.
6. Maurice JB, Patel A, Scott AJ, Patel K, Thursz M, Lemoine M. Prevalence and risk factors of nonalcoholic fatty liver disease in HIV-monoinfection. AIDS **2017**; 31:1621–32.
7. Verna EC. Non-alcoholic fatty liver disease and non-alcoholic steatohepatitis in patients with HIV. Lancet Gastroenterol Hepatol **2017**; 2:211–23.
8. Brown TT, Cole SR, Li X, et al. Antiretroviral therapy and the prevalence and incidence of diabetes mellitus in the multicenter AIDS cohort study. Arch Intern Med **2005**; 165:1179–84.
9. Feeney ER, Mallon PW. HIV and HAART-associated dyslipidemia. Open Cardiovasc Med J **2011**; 5:49–63.
10. Hadigan C, Meigs JB, Corcoran C, et al. Metabolic abnormalities and cardiovascular disease risk factors in adults with human immunodeficiency virus infection and lipodystrophy. Clin Infect Dis **2001**; 32:130–9.
11. Triant VA. Cardiovascular disease and HIV infection. Curr HIV/AIDS Rep **2013**; 10:199–206.
12. Woolnough EL, Hoy JF, Cheng AC, et al. Predictors of chronic kidney disease and utility of risk prediction scores in HIV-positive individuals. AIDS **2018**; 32:1829–35.
13. Vuille-Lessard É, Lebouché B, Lennox L, et al. Nonalcoholic fatty liver disease diagnosed by transient elastography with controlled attenuation parameter in unselected HIV monoinfected patients. AIDS **2016**; 30:2635–43.
14. Lemoine M, Assoumou L, De Wit S, et al; ANRS-ECHAM Group. Diagnostic accuracy of noninvasive markers of steatosis, NASH, and liver fibrosis in HIV-monoinfected individuals at risk of nonalcoholic fatty liver disease (NAFLD): results from the ECHAM study. J Acquir Immune Defic Syndr **2019**; 80:e86–94.
15. Eddowes PJ, Sasso M, Allison M, et al. Accuracy of FibroScan controlled attenuation parameter and liver stiffness measurement in assessing steatosis and fibrosis in patients with nonalcoholic fatty liver disease. Gastroenterology **2019**; 156:1717–30.
16. Morse CG, McLaughlin M, Proschan M, et al. Transient elastography for the detection of hepatic fibrosis in HIV-monoinfected adults with elevated aminotransferases on antiretroviral therapy. AIDS **2015**; 29:2297–302.
17. Castera L. Noninvasive methods to assess liver disease in patients with hepatitis B or C. Gastroenterology **2012**; 142:1293–302 e4.
18. Benmassaoud A, Nitulescu R, Pembroke T, et al. Liver-related events in HIV-infected persons with occult cirrhosis. Clin Infect Dis **2019**; 69:1422–30.
19. Karlas T, Petroff D, Sasso M, et al. Individual patient data meta-analysis of controlled attenuation parameter (CAP) technology for assessing steatosis. J Hepatol **2017**; 66:1022–30.
20. Pembroke T, Deschenes M, Lebouché B, et al. Hepatic steatosis progresses faster in HIV mono-infected than HIV/HCV co-infected patients and is associated with liver fibrosis. J Hepatol **2017**; 67:801–8.
21. Nerenberg KA, Zarnke KB, Leung AA, et al; Hypertension Canada. Hypertension Canada's 2018 guidelines for diagnosis, risk assessment, prevention, and treatment of hypertension in adults and children. Can J Cardiol **2018**; 34:506–25.
22. Anderson TJ, Grégoire J, Pearson GJ, et al. 2016 Canadian Cardiovascular Society guidelines for the management of dyslipidemia for the prevention of cardiovascular disease in the adult. Can J Cardiol **2016**; 32:1263–82.
23. Punthakee Z, Goldenberg R, Katz P; Diabetes Canada Clinical Practice Guidelines Expert Committee. Definition, classification and diagnosis of diabetes, prediabetes and metabolic syndrome. Can J Diabetes **2018**; 42(suppl 1):10–5.
24. Lopez-Giacoman S, Madero M. Biomarkers in chronic kidney disease, from kidney function to kidney damage. World J Nephrol **2015**; 4:57–73.
25. Stevens PE, Levin A; Kidney Disease: Improving Global Outcomes Chronic Kidney Disease Guideline Development Work Group Members. Evaluation and management of chronic kidney disease: synopsis of the kidney disease: improving global outcomes 2012 clinical practice guideline. Ann Intern Med **2013**; 158:825–30.

26. Crane HM, Grunfeld C, Willig JH, et al. Impact of NRTIs on lipid levels among a large HIV-infected cohort initiating antiretroviral therapy in clinical care. AIDS **2011**; 25:185–95.
27. Naicker S, Rahmanian S, Kopp JB. HIV and chronic kidney disease. Clin Nephrol **2015**; 83:32–8.
28. Yang KC, Hung HF, Lu CW, Chang HH, Lee LT, Huang KC. Association of non-alcoholic fatty liver disease with metabolic syndrome independently of central obesity and insulin resistance. Sci Rep **2016**; 6:27034.
29. Sterling RK, Smith PG, Brunt EM. Hepatic steatosis in human immunodeficiency virus: a prospective study in patients without viral hepatitis, diabetes, or alcohol abuse. J Clin Gastroenterol **2013**; 47:182–7.
30. Crum-Cianflone N, Dilay A, Collins G, et al. Nonalcoholic fatty liver disease among HIV-infected persons. J Acquir Immune Defic Syndr **2009**; 50:464–73.
31. Ingiliz P, Valantin MA, Duvivier C, et al. Liver damage underlying unexplained transaminase elevation in human immunodeficiency virus-1 mono-infected patients on antiretroviral therapy. Hepatology **2009**; 49:436–42.
32. Price JC, Ma Y, Scherzer R, et al. Human immunodeficiency virus-infected and uninfected adults with non-genotype 3 hepatitis C virus have less hepatic steatosis than adults with neither infection. Hepatology **2017**; 65:853–63.
33. Nansseu JR, Bigna JJ, Kaze AD, Noubiap JJ. Incidence and risk factors for prediabetes and diabetes mellitus among HIV-infected adults on antiretroviral therapy: a systematic review and meta-analysis. Epidemiology **2018**; 29:431–41.
34. Fahme SA, Bloomfield GS, Peck R. Hypertension in HIV-infected adults: novel pathophysiologic mechanisms. Hypertension **2018**; 72:44–55.
35. Allen AM, Therneau TM, Larson JJ, Coward A, Somers VK, Kamath PS. Nonalcoholic fatty liver disease incidence and impact on metabolic burden and death: A 20 year-community study. Hepatology **2018**; 67:1726–36.
36. Perazzo H, Cardoso SW, Yanavich C, et al. Predictive factors associated with liver fibrosis and steatosis by transient elastography in patients with HIV mono-infection under long-term combined antiretroviral therapy. J Int AIDS Soc **2018**; 21:e25201.
37. Guaraldi G, Lonardo A, Maia L, Palella FJ Jr. Metabolic concerns in aging HIV-infected persons: from serum lipid phenotype to fatty liver. AIDS **2017**; 31(suppl 2):147–56.
38. Smith CJ, Ryom L, Weber R, et al. Trends in underlying causes of death in people with HIV from 1999 to 2011 (D:A:D): a multicohort collaboration. Lancet **2014**; 384:241–8.
39. Dulai PS, Singh S, Patel J, et al. Increased risk of mortality by fibrosis stage in nonalcoholic fatty liver disease: systematic review and meta-analysis. Hepatology **2017**; 65:1557–65.
40. Wong CR, Lim JK. The association between nonalcoholic fatty liver disease and cardiovascular disease outcomes. Clin Liver Dis (Hoboken) **2018**; 12:39–44.
41. Shah ASV, Stelzle D, Lee KK, et al. Global burden of atherosclerotic cardiovascular disease in people living with HIV: systematic review and meta-analysis. Circulation **2018**; 138:1100–12.
42. Chalasani N, Younossi Z, Lavine JE, et al. The diagnosis and management of nonalcoholic fatty liver disease: practice guidance from the American Association for the Study of Liver Diseases. Hepatology **2018**; 67:328–57.
43. European Association for the Study of the Liver (EASL); European Association for the Study of Diabetes (EASD); European Association for the Study of Obesity (EASO). EASL-EASD-EASO Clinical Practice Guidelines for the management of non-alcoholic fatty liver disease. J Hepatol **2016**; 64:1388–402.
44. Macías J, Mancebo M, Merino D, et al; Spanish AIDS Research Network-HEP09 Study Group. Changes in liver steatosis after switching from efavirenz to raltegravir among human immunodeficiency virus-infected patients with non-alcoholic fatty liver disease. Clin Infect Dis **2017**; 65:1012–9.
45. Calza L, Colangeli V, Borderi M, et al. Improvement in liver steatosis after the switch from a ritonavir-boosted protease inhibitor to raltegravir in HIV-infected patients with non-alcoholic fatty liver disease. Infect Dis (Lond) **2019**; 51:593–601.
46. Sebastiani G, Saeed S, Lebouche B, et al. Vitamin E is an effective treatment for nonalcoholic steatohepatitis in HIV mono-infected patients. AIDS **2020**; 34: 237–44.
47. Introduction: standards of medical care in diabetes—2019. Diabetes Care **2019**; 42:S1–2.
48. Ryom L, Boesecke C, Bracchi M, et al. Highlights of the 2017 European AIDS Clinical Society (EACS) guidelines for the treatment of adult HIV-positive persons version 9.0. HIV Med **2018**; 19:309–15.
49. Sebastiani G, Cocciolillo S, Mazzola G, et al. Application of guidelines for the management of nonalcoholic fatty liver disease in three prospective cohorts of HIV-monoinfected patients. HIV Med **2020**; 21:96–108.