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ORIGINAL ARTICLE

Hepatic steatosis in people older and younger than fifty who are living with HIV and HIV-negative controls: A cross-sectional study nested within the POPPY cohort

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Abstract

Background: Hepatic steatosis is a major cause of chronic liver disease associated with several negative health outcomes. We compared the prevalence of and factors associated with steatosis in people living with and without HIV.

Methods: Older (>50 years) and younger (<50 years) people with HIV and older HIV-negative controls (>50 years) underwent liver transient elastography examination with controlled attenuation parameter (steatosis ≥238 dB/m, moderate/severe steatosis ≥280 dB/m, liver fibrosis ≥7.1 kPa). We compared groups using logistic regression/Chi-squared/Fisher's exact/Kruskal–Wallis tests.

Results: In total, 317 participants (109 older people with HIV; 101 younger people with HIV; 107 HIV-negative controls) were predominantly white (86%) and male (76%), and 21% were living with obesity (body mass index ≥30 kg/m^2). Most (97%) people with HIV had undetectable HIV RNA. The prevalence of fibrosis was 8.4%, 3.0%, and 6.5% in the three groups, respectively ($p = 0.26$). Fibrosis was predominately (>65%) mild. The prevalence of steatosis was the same in older people with HIV (66.4%) and controls (66.4%) but lower in younger people with HIV (37.4%; $p < 0.001$). After adjustment, younger people with HIV were less likely to have steatosis (odds ratio [OR] 0.26; 95% confidence interval [CI] 0.14–0.52) than controls, but male sex (OR 2.45; 95% CI 1.20–4.50) and high waist-to-hip ratio (OR 3.04; 95% CI 1.74–5.33) were associated with an increased odds of steatosis. We found no association between steatosis and HIV-related variables.

Conclusions: The prevalence of hepatic steatosis and fibrosis was similar between older participants regardless of HIV status. Age, sex, and abdominal

obesity, but not HIV-related variables, were associated with steatosis. Interventions for controlling obesity should be integrated into routine HIV care.

KEYWORDS
hepatic steatosis, HIV, liver disease, obesity

INTRODUCTION

Hepatic steatosis (HS) is a major cause of morbidity globally and is associated with negative health outcomes, including not only hepatic but also metabolic and cardiovascular complications [1, 2]. Alcohol-induced liver disease significantly contributes to fat infiltration, but non-alcoholic fatty liver disease (NAFLD) represents a more common and complex clinical entity associated with multiple causal factors [3].

HS has also been described in people living with HIV, often at a higher prevalence than in the general population [4]. In addition to factors associated with HS in the general population, such as insulin resistance, type 2 diabetes mellitus (T2DM), or obesity, specific factors such as HIV infection itself or some antiretroviral drugs, might contribute to the development of HS in people with HIV [3, 5]. The prevalence of HS increases with age in the general population [6], but the impact of age on the risk of developing HS in people with HIV is less clear.

The Pharmacokinetic and clinical Observations in PeoPle over fiftY (POPPY) study is a multicentre, prospective, observational study initiated in 2013 to assess some clinical outcomes of people with HIV over the age of 50 years [7] and offers a good opportunity to explore HS in people living with and without HIV. We undertook a cross-sectional sub-study nested within the POPPY cohort to explore the prevalence and severity of and risk factors for HS in people with HIV and suitable controls.

METHODS

Participants and methods

POPPY is an ongoing cohort study conducted in seven sites in the UK and one in Ireland for which participants were recruited as previously described (EudraCT: 2012-003581-40) [8]. In short, the study included three groups: (1) people living with HIV aged >50 years; (2) people living with HIV aged <50 years; and (3) HIV-negative controls aged >50 years at study entry (2013–2016). Between 5 February 2019 and 20 March 2020, participants without known active liver disease from study sites in the UK were invited to join this cross-sectional sub-study, which was approved by the London South-East Research Ethics Committee (reference 18/LO/1669). All participants provided written informed consent. Although the three groups within the main POPPY study were frequency matched for age, gender, sexual orientation, and geographic region, this matching was not necessarily maintained within the sub-study.

At the sub-study visit, we collected data on demographic, anthropometric, clinical, and epidemiological variables, including age, sex at birth, ethnicity, body weight and height, hip and waist circumference, blood pressure, previously diagnosed comorbidities (including self-reported past viral hepatitis B and C), current concomitant medication use, and current and past medication antiretroviral therapy (ART) use. Participants were asked to complete the Alcohol Use Disorders Identification Test (AUDIT) [9] and the Drug Use Disorders Identification Test (DUDIT) [10] questionnaires. Blood samples were collected to measure platelet count and HIV RNA. Plasma and serum samples were stored at −80°C to allow batch testing of aminotransferases, bilirubin, gamma-glutamyl transferase, alkaline phosphatase, lipid profile, blood glucose, insulin, C-reactive protein, interleukin-6, tissue inhibitor of matrix metalloproteinase-1, procollagen III amino-terminal peptide, hyaluronic acid, total plasma proteins, and globulin.

Measurement of steatosis and fibrosis of the liver

Participants underwent liver vibration controlled transient elastography (VCTE) examination with controlled attenuation parameter (CAP) (FibroScan®) conducted by certified and experienced operators. HS was defined as CAP score ≥238 dB/m and severe steatosis as ≥280 dB/m. Liver fibrosis was defined as ≥7.1 kPa and graded as F2 (7.1–8.6 kPa), F3 (8.7–10.2 kPa), F4 (10.3–12.4 kPa), or cirrhosis (≥12.5 kPa) [11]. Severe liver disease was defined as CAP score ≥280 dB/m and liver fibrosis (≥7.1 kPa). Only participants with 10 valid VCTE readings were included in the analysis.

We calculated non-invasive scores for liver fibrosis, including aspartate aminotransferase (AST) to platelet ratio index (APRI) [12], Fibrosis-4 Index [13], and Enhanced Liver Fibrosis (ELF) score [14], and for HS,

including Fatty Liver Index (FLI) [15], NAFLD-Liver Fat Score [16], and Hepatic Steatosis Index (HSI) [17]. We also calculated the FibroScan-AST score for non-alcoholic steatohepatitis (NASH) [18].

Statistical analysis

We summarized key participant characteristics, medical history/comorbidities, medication use (current and past), and laboratory measurements. We made comparisons by study groups using Kruskal–Wallis tests for continuous variables and Chi-squared tests or Fisher's exact tests for categorical variables, as appropriate. We reported missing data for each variable when applicable.

The primary outcome was HS using the VCTE (FibroScan®) scores as defined above. We excluded those with invalid VCTE measurements. We compared the primary outcome by study group using Chi-squared tests for binary outcomes and Cochran–Armitage tests for ordinal outcomes. We further used logistic regression to identify the independent association with study group after adjustment for potential confounders. The list of variables to be considered as confounders was developed following an initial review of the literature and discussion among study investigators. The final selection of variables for inclusion was made pragmatically, based on a combination of clinical and statistical significance, while being mindful of the need to avoid over-fitting models. Therefore, age, sex, race (white/Black African) and categorized AUDIT score (AS; abstainers = 0; low risk = 1–8; risky ≥8) were included in all models, regardless of statistical significance, as these were deemed to be of clinical relevance. Other variables selected for inclusion given their significance in univariate models were dichotomised waist-to-hip ratio (WHR; higher or lower than 0.94 for males or 0.80 for females); and a binary composite measure of insulin resistance (participants were coded as being 'insulin resistant' if they met at least one of (a) diabetes mellitus based on concomitant medications OR medical history; (b) insulin resistance (homeostatic model assessment for insulin resistance >1.4), or (c) blood glucose ≥5.5 mmol/L). To avoid excluding participants with an incalculable AS due to missing AUDIT questionnaire items, we first conducted deductive imputation, where we deduced with certainty the response to a missing item from responses on other items of the AUDIT questionnaire. Subsequently, we conducted hot-deck imputation to impute remaining missing values [19]. However, seven participants (six controls, four of whom had HS, and one older person living with HIV and HS) had missing data on all 10 AUDIT questionnaire items and were excluded from analyses. We did not explore imputation of other variables, and a complete case analysis was conducted for each model. We also explored univariable associations for all variables included in the adjusted models. We conducted sensitivity analyses: (1) a complete case analysis excluding those with missing AUDIT item questionnaire data and (2) excluding participants with risky alcohol consumption (AS ≥8).

We conducted a linear regression analysis of the log-transformed HSI to explore the associations between HSI and HS. We tested for the statistical significance of an interaction between HS and HIV status to assess whether the effect of HS varied by HIV status before and after adjustment for other potential confounders using Wald tests. We limited the linear regression analyses of non-invasive markers/scores to HSI, as this was considered to be the most clinically relevant non-invasive marker/score. For other measured HS and fibrosis markers, we performed unadjusted comparisons by study groups using Kruskal–Wallis tests for continuous variables and Chi-squared tests or Fisher's exact tests for categorical variables, as appropriate.

Finally, we compared those who were and were not missing data on any variables included in the multivariable modelling for HS and HSI (before single imputation of the AUDIT items) and those who were and were not included in the sensitivity analysis based on the exclusion criteria of AS ≥8 using Wilcoxon rank-sum tests for continuous variables and Chi-squared or Fisher's exact tests for categorical variables, as appropriate. These comparisons were made to determine whether these groups differed in any systematic manner to better understand how the results may be biased.

All analyses were performed assuming a two-sided significance level of 0.05, and, where applicable, 95% Wald confidence intervals (CIs) were calculated.

RESULTS

A total of 317 participants (109 older people with HIV; 101 younger people with HIV; 107 HIV-negative controls) were enrolled. Participants were predominantly white (86.4%) and male (76.3%), but there were more female participants in the control group (37.4%) than among the older or younger people with HIV (15.6% and 17.8%, respectively). The median time between main POPPY study enrolment and the sub-study visit was 4.8 years (interquartile range [IQR] 4.3–5.4). Median age at study visit for the older, younger, and control groups was 61 (IQR 57–65), 49 (IQR 45–53), and 63 (IQR 60–67) years, respectively. Overall, 18.4% and 9.5% of the participants reported scores that indicated risky alcohol consumption or problematic drug use, respectively (Table 1). All but one participant with HIV were on ART (99.5%), and 96.5% had undetectable HIV RNA at the study visit. Over one-third of participants on ART (36.2%) were using an integrase strand transfer inhibitor (INSTI)-containing combination (Table 1). Although no study participant

TABLE 1 Baseline characteristics by study group.

Characteristic	Overall ($n = 317$)	Older people with HIV ($n = 109$)	Younger people with HIV ($n = 101$)	HIV-negative controls ($n = 107$)	*p*-value
Age (years)	58 (53–64)	61 (57–65)	49 (45–53)	63 (60–67)	<0.001
Sex at birth					
Male	242 (76.3)	92 (84.4)	83 (82.2)	67 (62.6)	<0.001
Female	75 (23.7)	17 (15.6)	18 (17.8)	40 (37.4)	
Ethnicity					
White	274 (86.4)	93 (85.3)	85 (84.2)	96 (89.7)	0.46
Black African	43 (13.6)	16 (14.7)	16 (15.8)	11 (10.3)	
Smoking status					
Current	47 (14.8)	18 (16.5)	23 (22.8)	6 (5.6)	0.002
Never/ex-smoker	270 (85.2)	91 (83.5)	78 (77.2)	101 (94.4)	
Alcohol consumption					
AS	3 (1–6)	3 (1–6)	3 (2–7)	4 (2–7)	0.28
Risky consumption (AS ≥8)	50 (18.4)	13 (13.3)	18 (21.4)	19 (21.1)	0.26
Missing	45	11	17	17	
Recreational drug use					
DUDIT score	0 (0–2)	0 (0–2)	0 (0–3)	0 (0–0)	<0.001
Problematic use (DUDIT ≥6)	28 (9.5)	14 (13.3)	12 (12.6)	2 (2.1)	0.01
Missing	21	4	6	11	
Comorbidities					
Past HBV	22 (6.9)	16 (14.7)	3 (3.0)	3 (2.8)	<0.001
Past HCV	24 (7.6)	12 (11.0)	11 (10.9)	1 (0.9)	0.006
T2DM	23 (7.3)	11 (10.1)	4 (4.0)	8 (7.5)	0.23
Hypercholesterolaemia	82 (25.9)	38 (34.9)	17 (16.8)	27 (25.2)	0.01
Current medication use for					
Hypertension	56 (17.7)	29 (26.6)	7 (6.9)	20 (18.7)	<0.001
Hyperlipidaemia	91 (28.7)	45 (41.3)	18 (17.8)	28 (26.2)	<0.001
T2DM	15 (4.7)	9 (8.3)	3 (3.0)	3 (2.8)	0.13
BMI (kg/m^2)	26.00 (23.67–29.33)	25.55 (23.70–28.50)	25.40 (23.50–28.80)	26.90 (24.20–30.10)	0.08
Obesity (BMI ≥ 30 kg/m^2)	67 (21.2)	20 (18.5)	18 (17.8)	29 (27.1)	0.18
Missing	1	1	0	0	
Waist-to-hip ratio	0.94 (0.88–0.99)	0.96 (0.91–1.01)	0.93 (0.87–0.98)	0.92 (0.87–0.98)	<0.001
Missing	2	1	1	0	
HIV-specific variables					
HIV RNA ≤50 copies/mL	194 (96.5)	101 (98.1)	93 (94.9)	–	0.27
CD4+ T-cell count (cells/mm^3)[a]	630 (500–802)	625 (520–798)	630 (484–810)	–	0.78
Nadir CD4+ T-cell count (cells/mm^3)[a]	214 (115–333)	200 (104–288)	250 (120–388)	–	0.06
On ART	209 (99.5)	109 (100.0)	100 (99.0)	–	0.49
Any PI-based	58 (27.6)	35 (32.1)	23 (22.8)	–	0.13
Any NNRTI-based	77 (36.7)	35 (32.1)	42 (41.6)	–	0.16
Any INSTI-based	76 (36.2)	43 (39.4)	33 (32.7)	–	0.31
Any TAF-containing	56 (26.7)	37 (33.9)	19 (18.8)	–	0.01
Past exposure to d-drugs	33 (15.7)	21 (19.3)	12 (11.9)	–	0.14

Note: data are presented as *n* (%) or median (interquartile range) unless otherwise indicated.
Abbreviations: ART, antiretroviral therapy; AS, AUDIT score; AUDIT, Alcohol Use Disorders Identification Test; BMI, body mass index; DUDIT, Drug Use Disorders Identification Test; HBV, hepatitis B virus; HCV, hepatitis C virus; INSTI, integrase strand transfer inhibitor; NNRTI, non-nucleoside reverse transcriptase inhibitor; PI, protease inhibitor; TAF, tenofovir alafenamide; T2DM, type 2 diabetes mellitus.
[a]Measured at POPPY baseline visit, not the POPPY Liver sub-study visit.

was taking dideoxynucleosides (d-drugs) at the time of the study visit, 15.7% of participants with HIV had received d-drugs in the past.

History of hepatitis B virus infection was more common in older people with HIV (14.7%) than in the other two groups (3% in both). However, history of prior hepatitis C virus infection was similar in both groups of people with HIV (11%) and much higher than in controls (0.9%). At the time of the sub-study visit, liver function test results mostly fell within the normal range in all study groups.

Metabolic complications such as hyperlipidaemia or T2DM were more common in older participants, regardless of HIV status, than in younger people with HIV (Table 1). However, a larger proportion of older people with HIV was on treatment for hyperlipidaemia (statins, fenofibrate, or omega-3 acid) or T2DM (metformin, gliclazide, sitagliptin, or empagliflozin) (41.3% and 8.3%) than younger people with HIV (17.8% and 3.0%) or controls (26.2% and 2.8%), respectively. Blood glucose, triglycerides and total, high-density lipoprotein, and low-density lipoprotein cholesterol values were similar between the study groups and mostly within the normal range (Table S1). The proportion of people living with obesity (body mass index [BMI] ≥30 kg/m^2) was similar in older and younger people with HIV (18.5% and 17.8%, respectively) but was higher in the control group (27.1%). Median WHR was higher in older people with HIV (0.96 [IQR 0.91–1.01]) than in younger people with HIV (0.93 [IQR 0.87–0.98]) and controls (0.92 [IQR 0.87–0.98]); $p < 0.001$ (Table 1).

Hepatic steatosis

Four participants were excluded from the main analysis because of invalid VCTE scores. The prevalence of HS (CAP scores ≥238 dB/m) was equal in older people with HIV (66.4% [95% CI 57.4%–75.3%]) and controls (66.4% [95% CI 57.4%–75.3%]; $p > 0.99$) and higher than in younger people with HIV (37.4% [95% CI 27.8%–46.9%]; $p < 0.001$) (Table 2). The prevalence of severe steatosis (≥280 dB/m) was also higher in the older groups with and without HIV (29.0% [95% CI 20.4%–37.6%] and 29.9% [95% CI 21.2%–38.6%], respectively) than in the younger group with HIV (21.2% [95% CI 13.2%–29.3%]).

We observed no difference between participants with and without HS on liver function test results, blood glucose levels, or total, low-density, or high-density lipoprotein cholesterol. However, although median triglycerides were within the normal range, they were higher in those with HS (1.3 [IQR 1.0–1.7] vs 1.8 [IQR 1.3–2.5] mmol/L; $p < 0.001$) as was insulin (9.2 [IQR 4.8–15.9] vs 12.8 [IQR 7.5–24.4] mIU/L; $p < 0.001$) than in those without steatosis (Table S2).

The proportion of participants with scores suggestive of HS based on the HSI (>36) was 15.8% (95% CI 8.5%–23.1%), 15.5% (95% CI 7.7%–23.2%), and 27.7% (95% CI 18.6%–36.7%) in older people with HIV, younger people with HIV, and control groups, respectively ($p = 0.06$). We also found no difference between the study groups on the proportion of participants with FLI suggestive of HS (FLI > 60; 42.9% [95% CI 33.4%–52.3%], 40.7% [95% CI 30.6%–50.8%], and 42.6% [95% CI 32.9%–52.2%]; $p = 0.95$) (Table 2).

Liver Fibrosis

The prevalence of liver fibrosis in the study population was low (6.1% [95% CI 3.4%–8.7%]) but was higher in older people with HIV (8.4% [95% CI 3.2%–13.7%]) and controls (6.5% [95% CI 1.9%–11.2%]) than in younger people with HIV (3.0% [95% CI 0.0%–6.4%]; $p = 0.26$). Three younger people with HIV had liver fibrosis, which was mild (F2) in two. Most of the participants with fibrosis in the other groups (>65%) also had mild (F2) disease. Only five participants had scores compatible with F3 or higher (three of the older people with HIV, one of the younger people with HIV, and one in the control group); the single participant in the control group had scores compatible with cirrhosis (Table 2).

Consistent with the VCTE findings, only nine participants (3.0% [95% CI 1.1%–4.9%]) had ELF scores suggestive of liver fibrosis. Six of these participants (5.7% [95% CI 1.3%–10.2%]) were in the group of older people with HIV, one (1.1% [95% CI 0.0%–3.1%]) was a younger person with HIV, and two (1.9% [95% CI 0.0%–4.6%]) were in the control group. Similar results were observed when exploring the APRI scores (Table 2).

Severe liver disease

Eight participants (2.6% [95% CI 0.8%–4.3%]) had severe HS (≥280 dB/m) and liver fibrosis (≥7.1 kPa). Both the group of older people with HIV and the group without HIV had three of these participants each (i.e. 2.8% [95% CI 0.0%–5.9%] for both groups), and two (2.0% [0.0%–4.8%]) were in the younger group of people with HIV. Using the FibroScan-AST score, 6.3% (95% CI 1.4%–11.2%), 8.3% (95% CI 2.4%–14.2%), and 3.2% (95% CI 0.0%–6.7%) of participants had scores suggestive of NASH in the older and younger groups of people with HIV and controls, respectively ($p = 0.34$) (Table 2).

Factors associated with HS

Table 3 shows the factors associated with HS. In univariate analysis, high WHR, as a marker of central fat accumulation, was associated with a higher odds for HS

TABLE 2 Liver fibrosis and hepatic steatosis by elastography and non-invasive markers by study group (participants with valid VCTE scores).

Variable	Overall ($n = 313$)	Older people with HIV ($n = 107$)	Younger people with HIV ($n = 99$)	HIV-negative controls ($n = 107$)	*p*-value
Liver fibrosis					
Elastography					
No liver fibrosis	294 (93.9)	98 (91.6)	96 (97.0)	100 (93.5)	0.26^{CS}; 0.46^{CA}
F2 (7.1–8.6 kPa)	14 (4.5)	6 (5.6)	2 (2.0)	6 (5.6)	
F3 (8.7–10.2 kPa)	2 (0.6)	2 (1.9)	0 (0.0)	0 (0.0)	
F4 (10.3–12.4 kPa)	2 (0.6)	1 (0.9)	1 (1.0)	0 (0.0)	
Cirrhosis (≥12.5 kPa)	1 (0.3)	0 (0.0)	0 (0.0)	1 (0.9)	
APRI (>1)	3 (1.2)	1 (1.2)	1 (1.3)	1 (1.2)	>0.99
Missing	69	22	21	26	
ELF (>10.51)	9 (3.0)	6 (5.7)	1 (1.1)	2 (1.9)	0.16
Missing	9	2	4	3	
HS					
Elastography					
No HS	134 (42.8)	36 (33.6)	62 (62.6)	36 (33.6)	$<0.001^{CS}$; 0.001^{CA}
Mild/moderate (CAP score ≥238 dB/m)	95 (30.4)	40 (37.4)	16 (16.2)	39 (36.4)	
Severe (CAP score >280 dB/m)	84 (26.8)	31 (29.0)	21 (21.2)	32 (29.9)	
FLI >60	125 (42.1)	45 (42.9)	37 (40.7)	43 (42.6)	0.95
Missing	16	2	8	6	
HSI >36	54 (19.8)	15 (15.8)	13 (15.5)	26 (27.7)	0.06
Missing	40	12	15	13	
Severe HS with liver fibrosis					
VCTE: ≥280 dB/m and ≥7.1 kPa	8 (2.6)	3 (2.8)	2 (2.0)	3 (2.8)	>0.99
NASH					
FAST score >0.35	16 (5.9)	6 (6.3)	7 (8.3)	3 (3.2)	0.34
Missing	40	12	15	13	

Note: Data are presented as *n* (%) unless otherwise indicated.

Abbreviations: APRI, aspartate aminotransferase to platelet ratio index; CA, Cochran–Armitage tests or categorical (ordinal) severity version of the variable; CAP, controlled attenuation parameter; CS, Chi-squared tests for binary version of the variable; ELF, enhanced liver fibrosis; FAST, FibroScan-aspartate aminotransferase; FLI, Fatty Liver Index; HS, hepatic steatosis; HSI, Hepatic Steatosis Index; NASH, non-alcoholic steatohepatitis; VCTE, vibration controlled transient elastography.

(OR 2.42 [95% CI 1.50–3.92]; $p<0.001$) but being in the younger group with HIV was associated with a 70% reduction in the odds of having HS compared with older people with HIV (0.30 [95% CI 0.17–0.54]; $p < 0.001$). We found evidence of an association between HS and our composite 'insulin resistance' variable, with a 69% increase in the odds of having HS among those classified as 'insulin resistant' compared with those not (1.69 [95% CI 1.06–2.72]; $p = 0.03$). In the adjusted model, younger people with HIV remained less likely to have HS (0.26 [95% CI 0.14–0.52]; $p < 0.001$), but male sex (2.45 [95% CI 1.20–4.50]; $p = 0.01$) and high WHR (3.04 [95% CI 1.74–5.33]; $p < 0.001$) were independently associated with increased odds of HS. When the analysis was limited to participants living with HIV, we found no association between HIV-related variables (i.e. past exposure to d-drugs, undetectable HIV RNA, or current exposure to any anchor ART drug class or tenofovir alafenamide (Table 3).

Factors associated with the HS index

Table 4 shows the factors associated with the HSI. In the univariate analysis, being classified as having steatosis (estimate [log-transformed HSI as outcome variable in

TABLE 3 Summary of logistic regression results to identify factors associated with hepatic steatosis and liver fibrosis.

Variable	Univariable	*p*-value	Adjusted	*p*-value
POPPY study group				
HIV-negative controls	REF	<0.001[LRT]	REF	<0.001[LRT]
Older people with HIV	0.99 (0.55–1.76)	0.96	0.70 (0.36–1.35)	0.29
Younger people with HIV	0.30 (0.17–0.54)	<0.001	0.26 (0.14–0.52)	<0.001
Sex at birth				
Female	REF		REF	
Male	1.33 (0.78–2.27)	0.30	2.45 (1.20–4.50)	0.01
Race				
White	REF		REF	
Black African	0.73 (0.37–1.44)	0.36	1.04 (0.43–2.54)	0.93
Waist-to-hip ratio				
Low health risk	REF		REF	
Moderate/high health risk (>0.80 female; >0.94 male)	2.42 (1.50–3.92)	<0.001	3.04 (1.74–5.33)	<0.001
Insulin resistance composite binary measure				
No	REF		REF	
Yes[a]	1.69 (1.06–2.72)	0.03	1.36 (0.81–2.28)	0.25
AS				
Abstainers (AS = 0)	REF	0.24 [LRT]	REF	0.07[LRT]
Low risk (1 ≤ AS <8)	0.55 (0.22–1.34)	0.19	0.38 (0.12–1.12)	0.08
Risky (AS ≥8)	0.79 (0.29–2.31)	0.64	0.63 (0.19–2.13)	0.46
Hypertension (based on current treatment)				
No	REF		REF	
Yes	2.36 (1.22–4.56)	0.01	1.32 (0.63–2.80)	0.44
Hypercholesterolaemia				
No	REF		REF	
Yes	1.73 (1.00–2.98)	0.05	1.24 (0.69–2.45)	0.41
HIV-specific variables, considering only those with HIV				
HIV RNA ≤50 copies/mL				
No	REF			
Yes	0.42 (0.08–2.20)	0.30		
History of d-drug exposure[b]				
No	REF			
Yes	1.12 (0.53–2.36)	0.77		
On any Integrase Inhibitor				
No	REF			
Yes	1.33 (0.75–2.36)	0.33		
On any protease inhibitor				
No	REF			
Yes	0.88 (0.48–1.63)	0.69		
On any non-nucleoside reverse transcriptase inhibitor				
No	REF			

(Continues)

TABLE 3 (Continued)

Variable	Univariable	*p*-value	Adjusted	*p*-value
Yes	0.83 (0.47–1.47)	0.53		
On any TAF-containing combination				
No	REF			
Yes	0.98 (0.53–1.81)	0.94		

Note: Data are presented as odds ratio (95% confidence interval) unless otherwise indicated.
Abbreviations: AS, AUDIT score; AUDIT, Alcohol Use Disorders Identification Test; HOMA-IR, homeostatic model assessment for insulin resistance; LRT, *p*-value from a likelihood ratio test to jointly test that all coefficients of a categorical variable are equal to 0, adjusting for all other variables in the model if applicable; TAF, tenofovir alafenamide.
[a]Diabetes mellitus based on concomitant medications or medical history OR insulin resistance (HOMA-IR >1.4) OR blood glucose 5.5–6.9 mmol/L (pre-diabetes range).
[b]Past medication use of didanosine, stavudine, and/or zalcitabine.

linear regression, interpreted as a multiplicative effect relative to reference group for categorical variable or unit change for continuous variable]: 1.14 [95% CI 1.10–1.18]; $p < 0.001$), being Black African (1.16 [95% CI 1.09–1.23]; $p < 0.001$), and having a high WHR (1.15 [95% CI 1.10–1.19]; $p < 0.001$) were all associated with a higher mean HSI. However, living with HIV (0.94 [95% CI 0.90–0.98]; $p = 0.007$) and being male (0.89 [95% CI 0.85–0.93]; $p < 0.001$) were associated with a lower mean HSI. We found no associations between HSI and our composite 'insulin resistance' variable (1.03 [95% CI 0.99–1.07]; $p = 0.12$), age ([per 5 years older] 1.01 [95% CI 1.00–1.02]; $p = 0.26$), and categorical AS ('abstainers' = reference, 'low risk' = 0.96 [0.89–1.03], 'risky' = 0.95 [0.87–1.03]; likelihood-ratio test $p = 0.43$). In the adjusted model, similar associations were observed but with estimates slightly attenuated; and age was statistically significant, with an increase in age associated with a slight reduction in the HSI (per 5 years older): 0.99 (95% CI 0.98–1.00); $p = 0.02$.

Finally, we compared the sub-groups of participants that were excluded from some of the main analyses when missing data on outcomes and/or variables were included in the modelling and found few differences between these groups in baseline characteristics and outcomes, with the exception of race and WHR (Table S7). Conducting analyses among those not missing any AUDIT item data did not modify the findings (Tables S2 and S3). Similarly, excluding participants with an AS ≥8 did not modify the main findings (Tables S4 and S5). We also found few differences between these groups in baseline characteristics and outcomes when comparing sub-groups of participants that were excluded from the sensitivity analysis based on an AS ≥8 (Table S6). Conducting analyses among those with an AS <8 did not modify the findings (Tables S4 and S5), with the exception that our composite 'insulin resistance' variable was no longer statistically significant in the unadjusted analysis (Table S4).

DISCUSSION

In our study, the prevalence of HS was similar between older participants regardless of their HIV status and was higher than in younger people living with HIV. People living with HIV aged <50 years at study entry were 70% less likely to have CAP elastography scores compatible with HS than were older participants.

Abdominal obesity, defined as high WHR, was independently associated with HS. Although central obesity has been reported as a risk factor for HS in the general population [20] and in people living with HIV, such associations were reported in the context of glucose metabolism impairment and/or hyperlipidaemia [21–23]. In our main analysis, we found no evidence of an association between dysglycaemia and HS, and those reporting risky alcohol consumption were not more likely to have HS. When excluding those reporting risky alcohol consumption (AS ≥8), we observed associations, or a lack thereof, similar to those in the main analysis, which is consistent with a smaller study by Michel et al. [24]. HS seems to be driven more by metabolic disorders and is frequently seen in association with clinically relevant hyperlipidaemia and impaired glucose metabolism or diabetes, although it could precede those [25, 26].

In addition to factors associated with HS in the general population, additional elements in people living with HIV, such as past exposure to d-drugs [21, 27] or current use of INSTI, tenofovir alafenamide, or non-nucleoside reverse transcriptase inhibitors [28], may increase the likelihood of fat deposition in the liver [27, 29] and progression to NASH [29]. With the exemption of d-drugs [30], the mechanisms by which contemporary antiretroviral drugs may induce HS are unknown. Body weight gain has been postulated as a mechanism for INSTI-associated HS [29], but significant weight gain remains controversial in those switched to INSTI-based combination treatment [31, 32]. In ART-naïve individuals, dolutegravir has been associated

TABLE 4 Factors associated with (log-transformed) Hepatic Steatosis Index.

Variable	Univariable		Adjusted	
	Estimate (95% CI)	***p*-value**	**Estimate (95% CI)**	***p*-value**
Steatosis (CAP score ≥ 238 dB/m)[a]				
No steatosis	REF		REF	
Steatosis	1.14 (1.10–1.18)	<0.001	1.13 (1.09–1.17)	<0.001
HIV status[a]				
HIV negative	REF			
HIV positive	0.94 (0.90–0.98)	0.007	0.95 (0.91–0.99)	0.02
Age (per 5 years)	1.01 (1.00–1.02)	0.26	0.99 (0.98–1.00)	0.02
Sex at birth				
Female	REF		REF	
Male	0.89 (0.85–0.93)	<0.001	0.93 (0.89–0.98)	0.02
Race				
White	REF		REF	
Black African	1.16 (1.09–1.23)	<0.001	1.02 (1.04–1.17)	0.001
Waist-to-hip ratio				
Low health risk	REF		REF	
Moderate/high health risk (>0.8 female; >0.97 male)	1.15 (1.10–1.19)	<0.001	1.09 (1.05–1.13)	<0.001
Insulin resistance composite binary measure				
No	REF		REF	
Yes[b]	1.03 (0.99–1.08)	0.12	1.03 (0.99–1.06)	0.12
AS				
Abstainers (AS = 0)	REF	0.25^{LRT}	REF	0.94^{LRT}
Low risk (1 ≤ AS <8)	0.94 (0.87–1.01)	0.10	1.01 (0.94–1.07)	0.86
Risky (AS ≥8)	0.93 (0.86–1.02)	0.13	1.00 (0.93–1.08)	0.98
Hypertension (based on current treatment)				
No	REF		REF	
Yes	1.13 (1.07–1.19)	<0.001	1.09 (1.04–1.14)	0.001
Hypercholesterolaemia				
No	REF		REF	
Yes	1.02 (0.97–1.07)	0.44	0.99 (0.96–1.03)	0.76

Abbreviations: AS, AUDIT score; AUDIT, Alcohol Use Disorders Identification Test; HOMA-IR, homeostatic model assessment for insulin resistance; LRT, *p*-value from a likelihood ratio test to jointly test that all coefficients of a categorical variable are equal to 0, adjusting for all other variables in the model if applicable; REF, reference.

[a]Hepatic steatosis and HIV status interaction, before adjustment for other variables ($p = 0.55$, regression estimates: steatosis [1.15; 95% CI 1.08–1.24], HIV status [0.98; 95% CI 0.92–1.04], steatosis: HIV status interaction [0.98; 95% CI 0.90–1.06]); after adjustment for other variables ($p = 0.53$, regression estimates: steatosis [1.15; 95% CI 1.08–1.22], HIV status [0.97; 95% CI 0.91–1.03], steatosis: HIV status interaction [0.98; 95% CI 0.91–1.05]).

[b]Diabetes mellitus based on concomitant medications or medical history OR insulin resistance (HOMA-IR >1.4) OR blood glucose: 5.5–6.9 mmol/L (pre-diabetes range).

with higher weight gain than raltegravir or elvitegravir [33], but it was less likely to be associated with incident steatosis than either of these two in the COCOMO cohort [27]. Similar weight gain has been reported in individuals started on bictegravir- and dolutegravir-containing combinations [34], but the impact of these drugs on visceral fat deposition remain under discussion. We found that body weight but not current exposure to any drug family or previous use of d-drugs was associated with HS. In the POPPY cohort, obesity was also associated with age and gender but not HIV status or ART [8]. Our study cannot inform on hepatic or extrahepatic disease progression but provides an

opportunity to compare the prevalence and severity of steatosis of the liver between people with and without HIV with similar demographic and lifestyle characteristics.

We observed a low prevalence of liver fibrosis in our study population (6.1%), and most participants with fibrosis had VCTE scores compatible with early-stage disease. We did not find any association between fibrosis and plasma-based non-invasive tests, but participants with liver fibrosis had higher ELF scores than those without it, although most did not reach the threshold proposed to identify those with high probability of liver fibrosis. Despite being recommended by the UK National Institute for Health and Care Excellence, ELF seems to have limited ability to perform in populations with a low prevalence of fibrosis [35]. The identification and validation of non-invasive assessment tests for mild/moderate liver fibrosis, particularly in those with steatosis, remains an unmet clinical need [36]. Elastography might provide valuable information in these population groups.

The POPPY study is a relatively large and well-characterized cohort that has benefited from including a control group of HIV-negative individuals. However, our study has some limitations related to its cross-sectional design. Unmeasured residual confounding and channelling bias, particularly in relation to antiretroviral exposure, cannot be excluded. The generalizability of our findings could be limited as women and non-white ethnic groups were underrepresented in our sample. Furthermore, our sample reflects the population of people living with HIV in the UK, most of whom are receiving virally suppressive ART. In addition, we did not collect information on history of lipodystrophy that can promote visceral fat accumulation in people living with HIV [37]. Finally, although the threshold used to define steatosis was based on that used in previous studies, a higher cut-off has been recently proposed for people with HIV [38]. Analyses of severe steatosis (≥280 dB/m) revealed findings consistent with our main analysis, with a higher prevalence in the older groups than in the younger participants.

In summary, we found a similar prevalence of HS in older participants, irrespective of their HIV status, but a higher prevalence than in younger people with HIV. Traditional factors such as age, being male, and abdominal obesity, but not HIV-related variables, were associated with HS. Therefore, lifestyle modification and other interventions for controlling body weight and particularly central adiposity should be integrated into routine HIV care. The impact of contemporary antiretrovirals on HS remains in question.

AUTHOR CONTRIBUTION

AAP and CAS designed the study. AAP, AM, IW, JV, FAP, JA, and AW enrolled participants into the study. AAP, MB, and ND contributed to the coordination and oversight of the study. NB did the statistical analysis. All authors participated in data interpretation. The manuscript was drafted by AAP and NB. All authors provided input into the report and approved the final version of the manuscript.

CONFLICT OF INTEREST STATEMENT

AAP has received grants and/or personal fees from Gilead Sciences, Janssen, and ViiV, including the unrestricted research grant from ViiV that funded this study. CS has received funding from Gilead Sciences, Merck Sharp & Dohme, and ViiV Healthcare for membership of advisory boards and for preparation of educational materials. AW has received speaker fees, advisory board honoraria, or grants via Imperial College London from Gilead Sciences, ViiV Healthcare, MSD, and Janssen. AM has received speaker fees, advisory board honoraria or grants to her institution from Gilead Sciences, ViiV Healthcare, MSD, and Janssen. JA has received personal fees from Gilead Sciences and ViiV, all outside of the work reported here. FAP has received grants and/or personal fees from Gilead Sciences, ViiV, Janssen, and MSD, all outside of the work reported here. JV has received travel, research grants, and personal fees from Merck, Janssen Cilag, Piramal Imaging, ViiV Healthcare, and Gilead sciences, all outside of the work reported here. SLP has received grants from Gilead Sciences, ViiV Healthcare, Janssen-Cilag, EDCTP, Wellcome, and the National Institute for Health and Care Research, all outside of the work reported here. In addition, SLP receives salary support from Medical Research Council core funding (MC_UU_00004/03 and MC_UU_00004/04). NB, IW, MB, AO'B, RG, and ND report no conflicts of interest.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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