Antiretroviral Drugs and Risk of Chronic Alanine Aminotransferase Elevation in Human Immunodeficiency Virus (HIV)-Monoinfected Persons: The Data Collection on Adverse Events of Anti-HIV Drugs Study

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Background. Although human immunodeficiency virus (HIV)-positive persons on antiretroviral therapy (ART) frequently have chronic liver enzyme elevation (cLEE), the underlying cause is often unclear.

Methods. Data Collection on Adverse Events of Anti-HIV Drugs (D:A:D) Study participants without chronic viral hepatitis were observed to the earliest of cLEE (elevated aminotransferase ≥6 months), death, last follow-up, or January 2, 2014. Antiretroviral treatment exposure was categorized as follows: no exposure and ongoing short- and long-term exposure (<2 or ≥2 years) after initiation. Association between development of cLEE and ART exposure was investigated using Poisson regression.

Results. Among 21 485 participants observed for 105 413 person-years (PY), 6368 developed cLEE (incidence 6.04/100 PY; 95% confidence interval [CI], 5.89–6.19). Chronic liver enzyme elevation was associated with short-and long-term exposure to didanosine (<2 years rate ratio [RR] = 1.29, 95% CI, 1.11–1.49; >2 years RR = 1.26, 95% CI, 1.13–1.41); stavudine (<2 years RR = 1.51, 95% CI, 1.26–1.81; >2 years RR = 1.17, 95% CI, 1.03–1.32), and tenofovir disoproxil fumarate (<2 years RR = 1.55, 95% CI, 1.40–1.72; >2 years RR = 1.18, 95% CI, 1.05–1.32), but only short-term exposure to nevirapine (<2 years RR = 1.44, 95% CI, 1.29–1.61), efavirenz (<2 years RR = 1.14, 95% CI, 1.03–1.26), emtricitabine (<2 years RR = 1.18, 95% CI, 1.04–1.33), and atazanavir (<2 years RR = 1.20, 95% CI, 1.04–1.38). Chronic liver enzyme elevation was not associated with use of lamivudine, abacavir, and other protease inhibitors. Mortality did not differ between participants with and without cLEE.

Conclusions. Although didanosine, stavudine, nevirapine, and efavirenz have been described to be hepatotoxic, we additionally observed a consistent association between tenofovir and cLEE emerging within the first 2 years after drug initiation. This novel tenofovir-cLEE signal should be further investigated.

Keywords. alanine aminotransferase; antiretroviral therapy; hepatotoxicity; HIV; liver disease.

Human immunodeficiency virus (HIV)-infection has evolved into a chronic disease that requires lifelong antiretroviral therapy (ART). Therefore, drug-related toxicities have emerged as a key issue, including long-term ART-related liver injury [1].

Chronic liver enzyme (alanine aminotransferase [ALT]) elevation, a marker of persistent hepatocyte injury, is frequent among HIV-positive persons, even in the absence of hepatitis C (HCV) and hepatitis B (HBV) coinfection [2–4]. The cause of these elevations, its clinical significance, the need for evaluation, a reasonable diagnostic approach, and its management are often unclear, particularly when the elevation is modest. Most studies on liver disease focused on HCV- or HBV-coinfected persons or on the incidence and prevalence of severely elevated liver enzymes, defined as 3–5 times the upper limit of normal (ULN) or more [5]. Only limited data are available on HIV-monoinfected individuals with ALT values just above normal limits.

In previous studies, metabolic factors, including obesity, dyslipidemia and diabetes mellitus, severe alcohol use, high HIV viral load, and ART were predictive of persistently elevated ALT levels [2–4, 6]. Some antiretrovirals, such as the older stavudine (d4T) and didanosine (ddI), have been linked to liver injury [3, 6–11]. Data on the association of newer antiretrovirals and chronic liver disease are limited.

Study results on the outcome of liver enzyme elevations are controversial. A previous analysis of the Data Collection on Adverse Events of Anti-HIV Drugs (D:A:D) Study suggested that in HIV/HCV-coinfected persons, ALT levels above the normal limit are associated with a higher mortality [12], whereas in another investigation of HIV-monoinfected persons ALT elevation was not predictive of an increased death rate, although the observation period in this study was relatively short [11]. Two liver biopsy series demonstrated a high prevalence of liver fibrosis (FIB) and steatosis among HIV-monoinfected adults with chronic elevated liver enzymes [13, 14]. Such results emphasize that ALT is an important surrogate marker for relevant liver pathology.

Our study aims were (1) to identify risk factors associated with chronic ALT elevation (cLEE), focusing on the individual antiretroviral drugs and (2) to evaluate the outcome of liver enzyme elevation with regard to liver-related and all-cause mortality, within the frame of the D:A:D Study with its large size and long-term prospective observation.

METHODS

Study Design

The D:A:D Study, founded in 1999, is a prospective observational study of 11 previously established cohorts as described in detail [15]. Currently, more than 49 000 HIV-positive participants are followed in Europe, the United States, and Australia. The primary study aim was to investigate the associations between exposure to antiretroviral drugs and risk of myocardial infarction, and, subsequently, other major clinical events. Data, including sociodemographic characteristics, acquired immune deficiency syndrome (AIDS), viral hepatitis, deaths, known risk factors for cardiovascular disease, laboratory markers, and ART, are collected prospectively during routine clinical care. Information on causes of death is captured using the Coding of Causes of Death in HIV (CoDe) form [16]. Clinical events are regularly monitored and centrally adjudicated. The present analyses were limited to the participating cohorts that provided data on ALT levels.

Definitions

Chronic ALT elevation was defined as ALT levels greater than the ULN (males/females >50/>35 U/L) at ≥2 visits spanning at least 6 months within 2 years. We used the date of the first elevated ALT as the event date. A single normal ALT measurement between 2 elevated values was permitted and therefore did not signal the end of a period of cLEE. Hepatitis C virus infection was defined by HCV seropositivity or detectable HCV RNA. Hepatitis B virus infection was defined by a positive HBV surface antigen, HBV e antigen, HBV core antibodies, or detectable HBV DNA. Participants with unknown HBV or HCV status were excluded. Fibrosis-4 and aspartate aminotransferase (AST)-to-platelet index (APRI), 2 noninvasive

biomarkers of liver FIB, were calculated as follows: FIB-4: $(\text{age} \times \text{AST})/[\text{platelet count } (10^9 \text{ cells/L}) \times \text{sqrt(ALT)}]$, and APRI: $[(\text{AST/ULN})/\text{platelet count}] \times 100$. A FIB-4 value >3.25 or an APRI score >1.5 is considered indicative of cirrhosis, whereas with a FIB-4 ≤ 1.45 and an APRI score ≤0.5, respectively, significant FIB is unlikely [17, 18].

Statistical Analyses

Cohort-specific baseline dates were chosen according to the introduction of routine ALT monitoring in the individual cohorts. All D:A:D participants without HBV and HCV infection, with ≥3 ALT measurements, ≥6 months of follow-up, and normal ALT at baseline, were followed from baseline to the earliest of cLEE, death, 6 months prior to a date of a first positive HCV/HBV test, 6 months after last visit, or February 1, 2014. The incidence of cLEE was defined as the number of first events divided by the total person years of follow-up (PYFU).

We used Poisson regression models to assess the incidence of cLEE and its association with ART and other risk factors. Models were manually built using a standard structured approach including variables into a multivariable model in groups. At each stage, factors that were not significantly associated with cLEE were removed before moving to the next set of variables. Fixed covariables were sex, ethnicity, and participating cohort. Time-updated covariables were age, calendar year, hypercholesterolemia, hypertriglyceridemia, use of lipid-lowering drugs, lipodystrophy, body mass index (BMI), arterial hypertension, smoking status, and exposure to the individual antiretrovirals. Antiretroviral therapy exposure was categorized as follows for each individual drug: no exposure, ongoing short- and long-term exposure (<2 or ≥2 years) after initiation; and discontinuation for <2 or ≥2 years. We do not present risk estimates for drugs after discontinuation because interpretation is difficult without considering the subsequent drugs persons were switched to. Use of ritonavir (RTV) includes both full and boosting doses.

We also investigated the association between the total number and duration of cLEE episodes with all-cause mortality using Poisson regression models. For these analyses, individuals were followed to the earliest of death, 6 months prior to a date of a first positive HCV/HBV test, 6 months after last visit, or February 1, 2014, and all periods of cLEE, including any subsequent episodes, were included. Analyses were adjusted for age, sex, ethnicity, mode of HIV acquisition, calendar year, cohort, smoking status, BMI, and in a subsequent model additionally for time-updated CD4 cell counts and HIV viral loads. Note that these models did not include adjustment for the ART drugs received, because it was anticipated that any impact of ART on mortality would be largely driven by changes in CD4 counts and HIV viral loads. Multivariable analyses were not feasible for liver-related deaths because of small numbers. All analyses were performed using SAS version 9.3.

RESULTS

Patient Characteristics

Of 49 711 participants included in D:A:D until the cutoff date for the study of February 1, 2014, 29 291 (58.9%) were HCV and HBV negative and had available ALT results. We further excluded 3986 (8%) with <3 visits with ALT determinations or <6 months of follow-up and 3820 participants (7.7%) because of preexisting elevated ALT levels at time of baseline visit. Excluded participants were similarly compared with those who were included in terms of sex, age, ethnicity, mode of HIV acquisition, previous AIDS, and date of D:A:D enrollment, except that the group with incomplete follow-up contained more intravenous drug users (IDU) and persons of unknown ethnicity, and the group with increased ALT levels at baseline were more often IDUs and participants with previous AIDS. The present study is thus based on 21 485 HIV-positive individuals with normal ALT values at baseline (Figure 1).

The study population consisted of 72.9% men with a median age of 40 years. Of the participants, 51.9% were white, and 51.2% had acquired HIV through sex between men. At study entry, the median CD4 lymphocyte count was 470 cells/μL and 72.1% of participants were on ART. In total, 77.2% had ever been exposed to ART for a median duration of 3.9 years. Aspartate aminotransferase-to-platelet score was >1.5 in 523
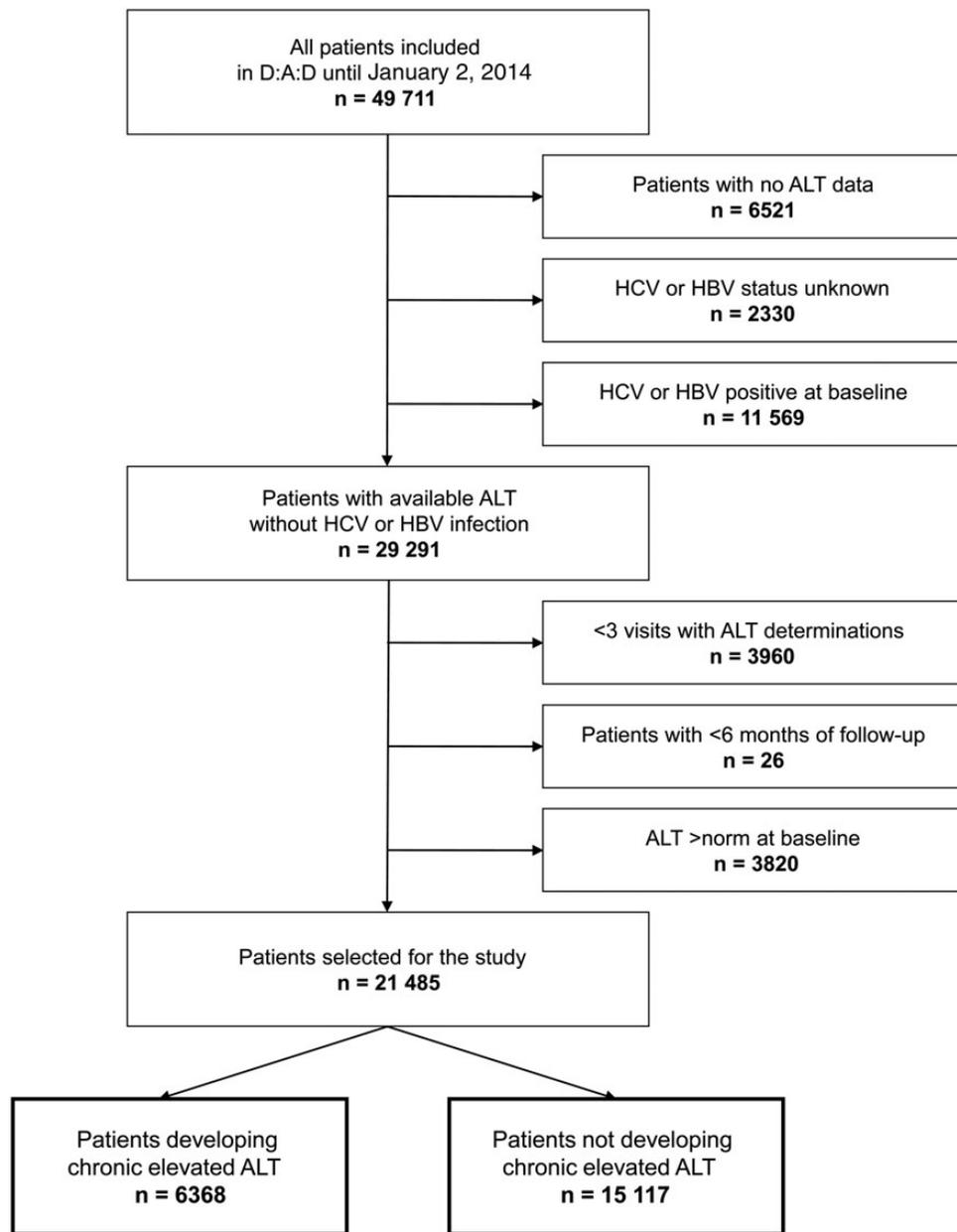
Figure 1. Patient flowchart. ALT, alanine aminotransferase; HBV, hepatitis B virus; HCV, hepatitis C virus.

(2.4%) and FIB-4 score was >3.25 in 647 (3.0%) of 12 857 (60%) persons with available scores (Table 1). Participants contributed a median of 13 (interquartile range [IQR], 6–23) ALT measurements to the analyses. Overall, the median baseline ALT level was 22 (IQR, 16–30) U/L.

Predictors for Chronic Alanine Aminotransferase Elevation

During a median follow-up of 6.6 years (IQR, 2.1–10.8) and 105 413 PYFU, 6368 (29.6%) participants experienced episodes of cLEE, resulting in an incidence of 6.04 per 100 PYFU (95% CI, 5.89–6.19). Increased BMI (>26 kg/m^2), dyslipidemia, use of lipid-lowering drugs, arterial hypertension, high HIV levels (≥5 log$_{10}$ copies/mL), and earlier calendar years were associated with cLEE. Older age (≥50 years), black ethnicity, male gender, and current smoking were inversely correlated (Supplementary Figure 1).

Among the nucleoside reverse-transcriptase inhibitors, cLEE was associated with ongoing exposure to regimens containing ddI, d4T, tenofovir disoproxil fumarate (TDF), and with exposure to emtricitabine (FTC) during the first 2 years. Exposure to lamivudine (3TC) was inversely correlated with cLEE. Among the nonnucleoside reverse-transcriptase inhibitors, the first 2 years of exposure to nevirapine (NVP) and efavirenz (EFV) were only associated with cLEE. The only protease inhibitor (PI) to demonstrate an association with cLEE was atazanavir (ATV) during the first 2 years of exposure. There was no evidence for an association with increased risk for the other PIs. Exposure to darunavir, RTV, and ATV for periods in excess of ≥2 years appeared to be protective for cLEE (Figure 2, Supplementary Table 1).

Because the association of TDF with cLEE was unexpected, we further analyzed commonly used TDF-containing regimens. The association was found to be more pronounced when TDF was used in combination with FTC and/or EFV (Figure 3).

Sensitivity Analyses

To substantiate the positive association between TDF and cLEE, we performed several sensitivity analyses. First, we excluded all participants who acquired HBV/HCV during follow-up, or who ultimately developed end-stage liver disease (ESLD), who might have received TDF for an unreported HBV infection or because of its perceived good liver safety profile. Second, we limited the analyses to ART-naive persons initiating TDF to rule out hepatotoxicity caused by a previous drug exposure. Third, we used a modified LEE definition of a single ALT value of ≥100 U/L. Results of all sensitivity analyses were consistent with our main analyses.

Clinical Outcome

Of the 6368 participants with ≥1 episode of cLEE, 1758 (27.6%) had at least 1 subsequent additional episode of cLEE. In total, the participants experienced a median of 1 (IQR, 1–6) episodes of cLEE with a median total duration of 1.5 (IQR, 0.5–13.7) years.

Overall, 924 persons died over a total of 151 191 PYFU (rate 0.61/100 PYFU; IQR, 0.57–0.65). All-cause mortality was

Table 1. Baseline Characteristics of 21 485 D:A:D Study Participants Without HCV or HBV Coinfection

Total of participants, no. (%)		21 485 (100)
Sex, no (%)	Male	15 661 (72.9)
Age, years	Median (IQR)	40 (33, 49)
Ethnicity, no. (%)	White	11 159 (51.9)
	Black	2110 (9.8)
	Other	580 (2.7)
	Unknown	7636 (35.5)
Mode of HIV acquisition, no. (%)	Heterosexual	8916 (41.5)
	Homosexual	10 990 (51.2)
	Injection drug use	243 (1.1)
	Other/unknown	1336 (6.2)
Duration of D:A:D cohort follow-up	Median (IQR)	6.6 (2.1, 10.8)
Previous clinical AIDS	No. (%)	5090 (23.7)
CD4 cells/µL	Median (IQR)	470 (318, 656)
Ever received antiretroviral therapy	No. (%)	16 578 (77.2)
Cumulative ART exposure (years)	Median (IQR)	3.9 (1.8, 7.0)
Ever received NRTIs	No. (%)	16 360 (76.2)
Cumulative ART exposure (years)	Median (IQR)	3.7 (1.7, 6.7)
Ever received PIs	No. (%)	11 922 (55.9)
Cumulative ART exposure (years)	Median (IQR)	2.5 (1.1, 4.2)
Ever received NNRTIs	No. (%)	10 094 (47.0)
Cumulative ART exposure (years)	Median (IQR)	1.9 (0.7, 4.4)
Body mass index, kg/m^2, no. (%)	<18	545 (2.5)
	≥18, ≤26	14 232 (66.2)
	>26, ≤30	3129 (14.6)
	>30	1154 (5.4)
	Unknown	2425 (11.3)
Diabetes mellitus	No. (%)	704 (3.3)
Total cholesterol, mmol/L	Median (IQR)	5.0 (4.2–5.8)
HDL cholesterol, mmol/L	Median (IQR)	1.2 (1.0–1.5)
Triglycerides, mmol/L	Median (IQR)	1.5 (1.0–2.4)
Use of lipid-lowering drugs	No. (%)	1855 (8.6)
Lipodystrophy	No. (%)	4260 (19.8)
Smoking status, n (%)	Current	7144 (33.3)
	Former	4735 (22.0)
	Never	7459 (34.7)
	Unknown	2147 (10.0)
FIB-4 score	≤1.45	10 352 (48.2)
	>1.45, ≤3.25	1858 (8.7)
	>3.25	647 (3.0)
	Unknown	8628 (40.2)
APRI score	≤0.5	11 385 (53.0)
	>0.5, ≤1.5	949 (4.4)
	>1.5	523 (2.4)

Abbreviations: AIDS, acquired immune deficiency syndrome; ALT, alanine aminotransferase; APRI, aspartate aminotransferase-to-platelet ratio; ART, antiretroviral therapy; D:A:D, Data Collection on Adverse Events of Anti-HIV Drugs; FIB, fibrosis; HBV, hepatitis B virus; HCV, hepatitis C virus; HDL, high-density lipoprotein; HIV, human immunodeficiency virus; IQR, interquartile range; NNRTI, nonnucleoside reverse-transcriptase inhibitor; NRTI, nucleoside reverse-transcriptase inhibitor; PI, protease inhibitor.

slightly higher in those who ever had an episode of cLEE compared with participants without cLEE (0.66/100 PYFU versus 0.60/100 PYFU), but this was not significant, either when unadjusted (rate ratio [RR] = 1.10; IQR, 0.95–1.28; P = .19), adjusted for basic demographic variables (RR = 1.13; IQR, 0.97–1.32; P = .11), or adjusted additionally for the latest CD4 cell count
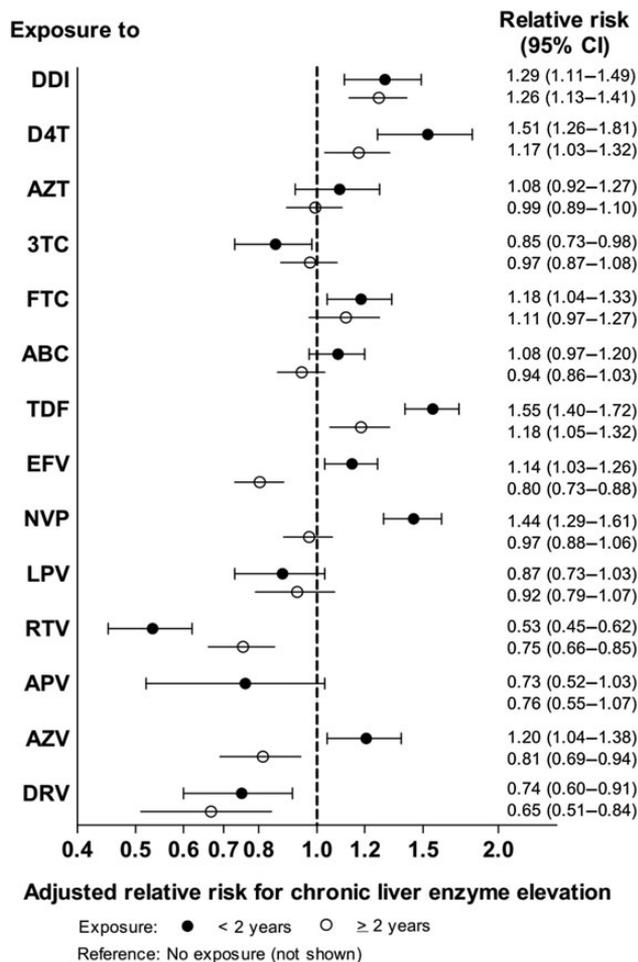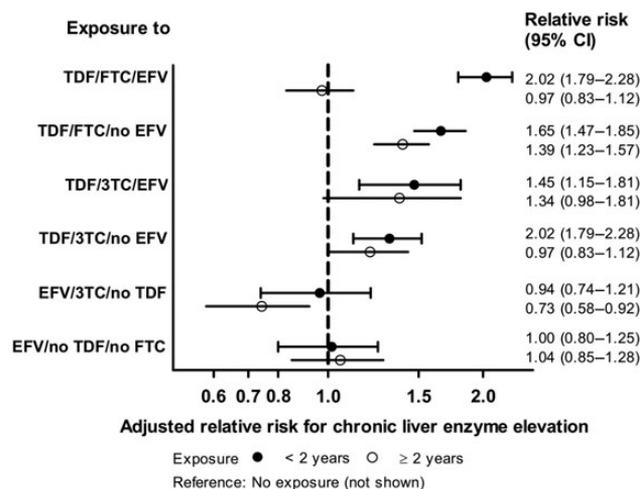
Figure 2. Incidence rate ratios and 95% confidence intervals (CIs) for the development of chronic liver enzyme elevation as a function of duration of antiretroviral drug exposure. Results are from Poisson regression based on 6368 events among 21 485 participants with 105 413 person years of follow-up. Models were adjusted for exposure to the other antiretrovirals, sex, age, ethnicity, body mass index, lipids, use of lipid-lowering drugs, lipodystrophy, arterial hypertension, smoking status, calendar year, and participating cohort, and for each drug discontinued for <2 years and ≥2 years. Abbreviations: ABC, abacavir; APV, amprenavir; AZT, zidovudine; AZV, atazanavir; D4T, stavudine; DDI, didanosine; DRV, darunavir; EFV, efavirenz; FTC, emtricitabine; LPV, lopinavir; NVP, nevirapine; RTV, ritonavir; TDF, tenofovir disoproxil fumarate; 3TC, lamivudine.

Figure 3. Incidence rate ratios and 95% confidence intervals (CIs) for the development of chronic liver enzyme elevation as a function of duration of antiretroviral drug exposure. Analyses are based on 6368 events among 21 485 participants with 105 413 person years of follow-up. Poisson regression models were adjusted for exposure to all other antiretrovirals (excluding lamivudine [3TC], emtricitabine [FTC], efavirenz [EFV], and tenofovir disoproxil fumarate [TDF]), sex, age, ethnicity, body mass index, lipids, use of lipid lowering drugs, lipodystrophy, arterial hypertension, smoking status, calendar year, and participating cohort.

and HIV viral load (RR = 1.15; IQR, 0.99–1.34; P = .08). Neither number of episodes of cLEE nor total duration of cLEE was associated with an increased risk of mortality in unadjusted or adjusted analyses (Table 2). The total number of liver-related deaths was 15 with 5 events among persons with cLEE and 10 events among those without any cLEE.

DISCUSSION

In this large longitudinal cohort analysis, the incidence of cLEE among HIV-positive individuals without viral hepatitis was high. In adjusted analyses, cLEE was associated with ongoing exposure to regimens containing ddI, d4T, and TDF and short-term exposure to NVP, EFV, FTC, and ATV. However, mortality did not appear to be increased in HIV-monoinfected participants with cLEE.

The potential for drug-induced liver toxicity of d4T, ddI, NVP, and EFV has previously been described [19]. In HIV-monoinfected and HCV-coinfected individuals, d4T and ddI have been associated with transaminase elevation, FIB, steatosis, and ESLD, most likely related to mitochondrial toxicity [3, 6–10]. In our study, NVP and EFV, known to cause early acute hepatitis driven by an immune-mediated hypersensitivity reaction, were associated with elevated ALT in the first 2 years of exposure [19, 20].

We were surprised to find a strong relationship between TDF and the development of cLEE emerging within the first 2 years after drug initiation. We confirmed this association using different sensitivity analyses. In another recent D:A:D Study of HIV-monoinfected and HCV/HBV-coinfected participants, TDF was also found to be associated with ESLD/hepatocellular carcinoma, supporting our observation [10]. In addition, our results are consistent with small case studies [21–24]. There are a few reports of liver injury and hepatic failure, including 1 person requiring liver transplantation, developing a few months after starting treatment with TDF/FTC/EFV in persons without preexisting liver disease [22–24]. Lattuada et al [21] reported on 3 individuals with liver enzyme elevation after the addition of TDF to an EFV-containing regimen. The drug registration trials did not identify a link between TDF and hepatotoxicity [25, 26]. The STEAL study, a randomized trial comparing abacavir/3TC

Table 2. Associations between cLEE, number of Episodes of cLEE, Total Duration of cLEE, and All-Cause Mortality[a]

Events	All-Cause Mortality		Unadjusted		Adjusted (1)		Adjusted (2)	
	Deaths/PY	Rate (95% CI)/100 PY	RR (95% CI)	P Value	RR (95% CI)	P Value	RR (95% CI)	P Value
Overall	924/151191	0.61 (.57, .65)						
Any cLEE								
No	682/114389	0.60 (.55, .64)	Ref.		Ref.		Ref.	
Yes	242/36802	0.66 (.57, .74)	1.10 (.95, 1.28)	.19	1.13 (.97, 1.32)	.11	1.15 (.99, 1.34)	.08
No. of episodes of cLEE								
None	682/114389	0.60 (.55, .64)						
1	192/28835	0.67 (.57, .76)						
2	41/6539	0.62 (.43, .82)						
3	9/1168	0.77 (.35, 1.46)						
≥4	0/230	0 (, 1.60)						
Per cLEE		1.05 (.95, 1.17)		.35	1.06 (.96, 1.18)	.25	1.07 (.96, 1.19)	.20
Total duration of cLEE, years								
≤0.5	685/117583	0.58 (.54, .63)						
>0.5–1	90/13227	0.68 (.54, .82)						
>1–1.5	40/5745	0.70 (.48, .91)						
>1.5–2	30/3935	0.76 (.49, 1.04)						
>2–3	51/4566	1.12 (.81, 1.42)						
>3–4	18/2527	0.71 (.42, 1.13)						
>4	10/3609	0.28 (.13, .51)						
Per year		1.02 (.96, 1.07)		.61	1.01 (.95, 1.07)	.76	1.01 (.96, 1.07)	.67

Abbreviations: CI, confidence interval; cLEE, chronic liver enzyme elevation; HIV, human immunodeficiency virus; PY, patient years; Ref., reference; RR, rate ratio.

[a] Adjusted for (1) sex, age, ethnicity, mode of HIV acquisition, body mass index, smoking status, calendar year, and participating cohort, and (2) additionally for the latest CD4 cell count and HIV RNA value.

versus TDF/FTC, showed a small but statistically significant increase in mean ALT levels on TDF/FTC [27], whereas the AIDS Clinical Trials Group A5202, a similar study, showed inconsistent results regarding elevated ALT values [28]. Randomized trials usually enroll participants who are healthier and younger and may therefore not be representative of the general population. Moreover, these trials with viral primary outcomes often lack statistical power to detect rare adverse drug effects due to their limited sample size.

The mechanisms of a possible TDF-related liver injury are unclear. In vitro data showed that TDF is not associated with mitochondrial toxicity [29]. In an animal study, it was found that TDF induced marked oxidative stress in renal and hepatic tissues of Wistar rats [30]. Our finding of the enhanced TDF signal when used in combination with EFV could reflect drug-drug interactions of TDF, which have been described with other antiretrovirals including ddI and ATV [31, 32]. It is interesting to note that in slow EFV metabolizers, such as those with CYP2B6 loss/diminished-function alleles, EFV plasma area-under-the-curve values were highest among patients receiving TDF [33].

In line with other recent reports, most PIs were not associated with liver toxicity [34]. We found that ATV during the first 2 years of exposure was predictive, but with ≥2 years of exposure ATV was protective for cLEE. In a previous study, ATV was associated with grade 3 LEE in HIV/HCV-seropositive individuals [34], whereas other investigations suggested that liver tolerability of ATV was good [35, 36]. Continuing surveillance for possible ATV-related hepatotoxicity is warranted.

Our data confirm that metabolic features, including dyslipidemia and a high BMI, are one of the main causes of liver disease in HIV-positive persons without chronic hepatitis B and C coinfection, similar to the general population. Our finding of black ethnicity as a negative predictor for ALT elevation is in agreement with other investigations, which found an inverse association of black ethnicity and hepatic steatosis in the general population and among HIV-infected patients [37, 38]. A high HIV viral load was associated with an increased risk of hepatopathy compared with individuals on ART with suppressed viral load, supporting the evidence that HIV itself is contributing to liver injury [39].

We found a low number of liver-related deaths among HIV-monoinfected individuals, in accordance with a previous D:A:D Study [40]. All-cause mortality was not significantly increased among persons with cLEE. Because patients are regularly monitored for their ALT levels, this allows the clinician an early warning and the possibility to switch drugs or introduce risk modification (eg, alcohol reduction, diet), reducing the potential impact from ART-related cLEE on mortality. Another possibility for why we did not see a link between cLEE and mortality may be a relatively short follow-up period. In the

large population-based US National Health and Nutrition Examination Survey III study, elevated ALT levels in HCV/HBV-negative persons were associated with a higher risk of liver-related death but not with all-cause death [41]. In contrast, studies including HCV/HBV-positive persons suggested both increased liver-related and all-cause mortality among persons with liver enzyme elevation in the general and HIV-positive population [12, 42]. Recent data indicate that cLEE is a marker of serious liver diseases. A large liver biopsy series of HIV-monoinfected individuals with cLEE revealed in two thirds of patients significant histological abnormalities with nonalcoholic steatohepatitis (NASH) in 55% and bridging FIB in 17% [14]. Given the long lag time between asymptomatic liver enzyme elevation and liver-related death, a study with a longer observation period may find an association between the two.

The strength of this study is its large size and the long-term prospective observation of a multinational population-based cohort collaboration. Such large observational studies are crucial to detect infrequent drug-related toxicities on the population level. To assess an individual drug effect might be challenging because antiretrovirals are always used in combination regimens. The large D:A:D Study, however, with extensive follow-up of patients drawn from heterogeneous settings should allow to disentangle individual drug effects because there is generally enough variability in the way an individual drug is used in combination with other drugs. Our study has several limitations. Information on alcohol use was not collected systematically. However, it is unlikely that the observed associations between antiretrovirals and cLEE are confounded by alcohol because alcohol consumption does not modify a physician's choice of ART. Moreover, by excluding HCV/HBV-coinfected persons and therefore most injection drug users with risk for multiple substance-dependence syndromes, including alcohol, many at-risk individuals were excluded. Furthermore, we did not have information on potentially hepatotoxic non-antiretroviral drugs. Finally, as a significant number of antiretrovirals were included in the analysis, we cannot rule out that findings may be a result of multiple testing.

CONCLUSIONS

In summary, the long-term observation of HIV-monoinfected individuals in this large cohort revealed a high incidence of cLEE. Besides the antiretrovirals ddI, d4T, NVP, and EFV, previously described to be hepatotoxic, FTC and ATV restricted to the first 2 years of treatment, we observed an additional strong association between TDF exposure and cLEE emerging within the first 2 years after drug initiation. Although mortality was not increased in our study, other studies found relevant histological abnormalities in HIV-monoinfected persons, with elevated ALT underscoring its link to significant liver morbidity [13, 14]. In this context, our results emphasize (1) that ddI and d4T should be avoided if alternative treatment is available and (2) that close monitoring of liver enzymes in persons on NVP and EFV is essential. Finally, the observed novel association between TDF and cLEE calls for further investigations to understand the pathophysiological mechanisms and its clinical implications. Ongoing surveillance of drug-related liver injury in large cohort collaborations is of paramount importance.

Supplementary Data

Supplementary material is available online at Open Forum Infectious Diseases online (http://OpenForumInfectiousDiseases.oxfordjournals.org/).

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