Hepatitis B and C Co-Infection Are Independent Predictors of Progressive Kidney Disease in HIV-Positive, Antiretroviral-Treated Adults

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Abstract

Chronic kidney disease (CKD) is an important cause of morbidity and mortality in HIV-positive individuals. Hepatitis C (HCV) co-infection has been associated with increased risk of CKD, but prior studies lack information on potential mechanisms. We evaluated the association between HCV or hepatitis B (HBV) co-infection and progressive CKD among 3,441 antiretroviral-treated clinical trial participants. Progressive CKD was defined as the composite of end-stage renal disease, renal death, or significant glomerular filtration rate (eGFR) decline (25% decline to eGFR <60 mL/min/1.73 m² or 25% decline with a baseline <60). Generalized Estimating Equations were used to model the odds of progressive CKD. At baseline, 13.8% and 3.3% of participants were co-infected with HCV and HBV, respectively. Median eGFR was 111, and 3.7% developed progressive CKD. After adjustment, the odds of progressive CKD were increased in participants with HCV (OR 1.72, 95% CI 1.07–2.76) or HBV (OR 2.26, 95% CI 1.15–4.44). Participants with undetectable or low HCV-RNA had similar odds of progressive CKD as HCV seronegative participants, while participants with HCV-RNA >800,000 IU/ml had increased odds (OR 3.07; 95% CI 1.60–5.90). Interleukin-6, hyaluronic acid, and the FIB-4 hepatic fibrosis index were higher among participants who developed progressive CKD, but were no longer associated with progressive CKD after adjustment. Future studies should validate the relationship between HCV viremia and CKD.

Trial Registration: ClinicalTrials.gov NCT00027352; NCT00004978.

Introduction

In 2006, ten years after the widespread introduction of effective combination antiretroviral therapy (cART) for the treatment of human immunodeficiency virus (HIV) infection, the randomized Strategies for Management of Antiretroviral Therapy trial (SMART) established uninterrupted cART with the goal of continuous viral suppression as the standard of care. [1] Although the primary outcome of SMART focused on mortality and opportunistic illness associated with acquired immunodeficiency syndrome (AIDS), the results also highlighted the growing burden of comorbid disease in the cART era. During an average follow-up of 16 months, serious cardiac, liver, and kidney events were more common than serious AIDS-defining events, regardless of treatment assignment. [1]

Chronic kidney disease (CKD) has been associated with increased morbidity and mortality in HIV-positive individuals receiving cART. [2–3] In addition to traditional CKD risk factors such as diabetes and hypertension, co-infection with hepatitis C virus (HCV) has been suggested as a possible risk factor for CKD in HIV-positive individuals. [4] Although there are conflicting data on the relationship between HCV infection and CKD in the general population, meta-analysis of published studies in HIV-positive populations supports an association between HIV-HCV co-infection and increased risk of CKD. [5] The relationship between hepatitis B virus (HBV) infection and CKD has not been as extensively studied, although cross-sectional studies have not...
demonstrated an association between HBV mono-infection and prevalent CKD. [6–7].

Both HBV and HCV have been implicated in the pathogenesis of specific immune complex kidney diseases in the general population and in HIV-positive individuals,[8–11] and clinically silent immune complex kidney disease has been observed in HCV mono-infected patients with end-stage liver disease. [12] End-stage liver disease has also been associated with increased risk of overt CKD in HCV-infected individuals, although data on the relative contribution of immune complex disease and hepatorenal syndrome were not available. [13] Previous studies have not investigated other potential mediators of the relationship between viral hepatitis and CKD, including earlier stages of hepatic fibrosis and liver dysfunction, increased levels of systemic inflammation, or nephrotoxic effects of antiviral therapy for HBV or HCV. In addition, the majority of prior studies defined viral hepatitis co-infection by serology alone, and did not report data on HBV DNA, HCV RNA, or HCV genotype as potential mediators or effectors of the relationship between viral hepatitis and CKD.

We evaluated the association between viral hepatitis co-infection and progressive CKD among 3,441 cART-treated participants enrolled in two large international HIV treatment trials, with the goal of identifying potential mediators of the relationship.

Methods

Study Population

The study designs of SMART and the Evaluation of Subcutaneous Proleukin in a Randomized International Trial (ESPRIT) have been described previously. [1,14] Briefly, SMART randomized 5,472 HIV-positive adults with CD4 cell count (CD4) >500 cells/mm$^3$ to receive uninterrupted cART with the goal of viral suppression versus episodic cART guided by CD4. SMART was stopped early because of a safety risk in the episodic therapy arm. [1] ESPRIT randomized 4,111 HIV-positive adults with CD4 >500 cells/mm$^3$ to receive cART alone or in combination with subcutaneous interleukin-2. ESPRIT failed to demonstrate a clinical benefit of interleukin-2 despite an increase in CD4. For consistency with standards of care, the current analysis included only participants randomized to the viral suppression arm of SMART and the control arm of ESPRIT. Baseline was defined as the date of randomization into SMART or ESPRIT. Eligible participants with plasma specimens available for centralization measurement of creatinine at baseline and at least one subsequent study visit were included in this analysis.

Definition of Study Endpoints and Covariates

Plasma specimens were collected in EDTA tubes, aliquoted, and shipped frozen to a central repository. Available specimens from the randomization (“baseline”), 12-month, and subsequent annual visits were retrieved for centralized creatinine testing using an isotope dilution mass spectrometry (IDMS)-traceable enzymatic assay (Roche Creatinine Plus in SMART and Diazyme Liquid Reagents Creatinine Assay in ESPRIT). The eGFR was calculated from centralized creatinine values using the Chronic Kidney Disease Epidemiology Consortium (CKD-EPI) equation. [15] In a sensitivity analysis, we explored the impact of adjusting eGFR in Asian participants using a correction coefficient of 0.813, as suggested for Japanese individuals, [16] or of 1.052, which was shown to improve the bias of the CKD-EPI equation among Chinese individuals. [17]

For the purposes of this analysis, progressive CKD was defined as the composite of end-stage renal disease (ESRD), renal death, or significant decline in eGFR (25% decline in eGFR to a level ≤60 mL/min/1.73 m$^2$) in participants with a baseline eGFR >60 mL/min/1.73 m$^2$ or a 25% decline in eGFR for those with a baseline eGFR <60 mL/min/1.73 m$^2$. To address the potential misclassification of acute kidney injury in participants with a 25% decline in eGFR based on a single follow-up creatinine value, we performed a sensitivity analysis requiring a confirmed 25% decline in eGFR based on at least 2 consecutive values.

Hepatitis co-infection was defined serologically at baseline, based on the detection of HBV surface antigen (HBsAg) or anti-HCV antibody. Additional testing was performed in participants with serologic evidence of HBV or HCV co-infection, including HBV-DNA or HCV-RNA and HCV genotype, respectively. Relevant comorbid conditions were defined at baseline. Hypertension, diabetes, and hyperlipidemia were defined by the self-reported use of medications to treat these conditions. Cardiovascular disease was defined by self-reported medical treatment or a history of coronary revascularization, myocardial infarction, or cerebrovascular accident prior to baseline. Plasma hyaluronic acid was measured as a circulating marker of hepatic fibrosis in co-infected participants, [18] and the FIB-4 and APRI fibrosis indices were calculated for all participants with available data at baseline.[19–20] The systemic inflammatory markers IL-6 and hsCRP were also measured at baseline in approximately 70% of participants, independent of hepatitis status.

Statistical Methods

Descriptive statistics were used to compare baseline characteristics between participants enrolled in SMART and ESPRIT, and between participants who did or did not develop progressive CKD. A descriptive analysis was also performed to compare the proportion of participants with progressive CKD at yearly intervals. The odds of progressive CKD were investigated using Generalised Estimating Equations, using binomial regression and adjusting for repeated measurements per person. Participants were included in analyses until the development of progressive CKD or the last eGFR measurement. In addition to HBV or HCV co-infection, other potential explanatory variables included age, sex, race, HIV exposure category, history of AIDS-defining illness, presence of other relevant comorbid conditions, HIV-RNA, CD4, CD4 nadir, body mass index (BMI), eGFR, and the use of cART both prior to and at randomization into the parent trial. Any exploratory variables with p < 0.1 in univariate analyses were included in multivariate analyses. Excluded variables were tested in the final model to determine if their inclusion improved the model fit. Separate sensitivity analyses were performed including time-updated (“on-treatment”) variables for selected antiretroviral agents, and including only participants with progressive CKD as defined by a clinical CKD event or a confirmed decline in eGFR based on two consecutive measures.

Further analyses focused on the role of baseline HBV or HCV viremia and HCV genotype as explanatory variables; HBV and HCV viremia were also explored as time-updated variables. Exploratory analyses using the same methods investigated the role of baseline hyaluronic acid, APRI, FIB-4, IL6 and hsCRP. Separate multivariate models were run for each of these markers to minimize the impact of missing data; as a result, the presented results were not mutually adjusted for the other markers. Multivariable models were adjusted for the same factors that were found to be of importance in the main analysis.

Results

A total of 4,792 participants were enrolled in the standard therapy arms of SMART and ESPRIT. [1,14] After excluding 971
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In multivariate analysis adjusting for baseline characteristics, the odds of developing progressive CKD were significantly increased in participants with HBV (adjusted OR 2.26, 95% CI 1.15–4.44) or HCV co-infection (adjusted OR 1.72, 95% CI 1.07–2.76). Other factors associated with increased odds of progressive CKD included black or Asian race, older age, and self-reported use of antihypertensive medications. Higher baseline eGFR and higher CD4 nadir were associated with lower odds of developing progressive CKD, as was a later date of randomization into the parent trial.

Sensitivity Analyses

In sensitivity analysis, the inclusion of time-updated “on-treatment” variables for potentially nephrotoxic antiretroviral agents (atazanavir, indinavir, boosted lopinavir, and tenofovir) and agents with dual activity against HIV and HBV (lamivudine and emtricitabine) did not significantly affect the relationship between progressive CKD and HBV (adjusted OR 2.20, 95% CI 1.13–2.89) or HCV co-infection (adjusted OR 1.77, 95% CI 1.08–2.89). In a separate sensitivity analysis including 2,659 participants with at least 2 centralized creatinine values during follow-up, 30 participants (1.1%) developed progressive CKD as defined by a clinical event or a decline in eGFR confirmed on two consecutive measures. In adjusted analysis, the odds ratios associated with hepatitis virus co-infection were similar to the primary analysis but no longer reached statistical significance (HBV co-infection 1.94, 95% CI 0.46–8.18; HCV co-infection 1.73, 95% CI 0.61–4.89).

Other relationships were consistent with the results of the primary analysis (data not shown). In a final sensitivity analysis, we explored the impact of adjusting eGFR in Asian participants using recommended correction coefficients for Japanese and Chinese populations.[16–17] While these adjustments impacted the number of CKD events in Asian participants, the relationships between HBV, HCV, Asian race, and progressive CKD were similar to the primary analysis.

Planned Subgroup Analysis of Participants with HBV or HCV Co-infection

Participants with serologic evidence of HBV co-infection were further stratified by the presence or absence of detectable HBV DNA at baseline, with a cutoff of <357 IU/ml (Figure 2). Among 70 participants with detectable HBV DNA, the median HBV DNA was 7.3 (IQR 5.3–7.3) log_{10} copies/mL. After adjustment for the variables in Table 2, participants with serologic evidence of HBV co-infection had similar odds of developing progressive CKD regardless of whether HBV DNA was undetectable (adjusted OR 2.16; 95% CI 0.84–5.55) or detectable at baseline (adjusted OR 2.33; 95% CI 0.97–5.50), but this failed to reach statistical significance in these smaller subgroups. Because of the small number of participants with HBV co-infection, it was not possible to further stratify viremic participants or to explore the role of HBV rebound during follow-up.

Participants with serologic evidence of HCV co-infection were further stratified into those with undetectable HCV RNA, low HCV RNA (≤800,000 IU/mL), and high HCV RNA (>800,000 IU/mL). After adjustment, participants with undetectable HCV RNA (adjusted OR 0.91, 95% CI 0.30–2.79) or with low HCV RNA (adjusted OR 1.41; 95% CI 0.71–2.79) had similar odds of developing progressive CKD as those with a negative HCV antibody result, while participants with high HCV RNA had significantly increased odds (adjusted OR 3.07; 95% CI 1.60–5.90). Among those with HCV co-infection, the test for trend moving from those with undetectable HCV RNA to those with high HCV RNA was marginally statistically significant (p = 0.057).
Similar results were seen when RNA levels were stratified by the median value (585,000 IU/ml) among viremic participants (data not shown). Further stratification of HCV RNA levels by quartiles suggested a gradual increase in risk of progressive CKD associated with increasing HCV RNA levels. The test for trend was again marginally statistically significant (p = 0.066), and the small number of events in each stratum meant that the confidence intervals for individual strata were wide (Figure 3). Among 353 participants with known HCV genotype, there were no differences in the odds of developing progressive CKD in those with genotype 1 versus other genotypes (Figure 2).

Circulating levels of hyaluronic acid, as well as the calculated FIB-4 index of hepatic fibrosis, were significantly higher at baseline among participants who subsequently developed progressive CKD, while there was no significant difference in the aspartate aminotransferase platelet ratio index (APRI). None of the markers of hepatic fibrosis remained significantly associated with progressive CKD after adjustment for other important covariates.

### Table 1. Baseline characteristics of study participants, stratified by the development of progressive chronic kidney disease (CKD).

|                           | All (n = 3,441) | No progressive CKD (n = 3,314) | Progressive CKD (n = 127) | p     |
|---------------------------|-----------------|-------------------------------|---------------------------|-------|
| Median age Years          | 43              | 42                            | 36.39                     | 48    | 39.55 | <0.0001 |
| Male sex                  | 2638            | 2544                          | 76.8                      | 94    | 74.0  | 0.47    |
| Race                      |                 |                               |                           |       |       |         |
| Black                     | 693             | 663                           | 20.0                      | 30    | 23.6  | <0.0001 |
| Asian                     | 206             | 185                           | 5.6                       | 21    | 16.5  | –       |
| White                     | 2280            | 2211                          | 66.7                      | 69    | 54.4  | –       |
| Other                     | 262             | 255                           | 7.7                       | 7     | 5.5   | –       |
| Exposure category         |                 |                               |                           |       |       |         |
| Intraavenous drug use     | 331             | 315                           | 9.5                       | 17    | 13.4  | 0.14    |
| Heterosexual contact      | 1387            | 1331                          | 40.2                      | 56    | 44.1  | 0.38    |
| Same sex contact          | 1812            | 1745                          | 53.0                      | 56    | 44.1  | 0.049   |
| Median CD4 cells/mm³      | 524             | 526                           | 415,072                   | 457   | 390,570 | <0.0001 |
| Median nadir CD4 cells/mm³ | 223           | 226                           | 117,333                   | 153   | 87,262 | <0.0001 |
| HIV RNA <500 copies/mL    | 2626            | 2533                          | 76.6                      | 93    | 73.2  | 0.38    |
| History of AIDS           | 906             | 862                           | 26.0                      | 44    | 34.7  | 0.030   |
| Antiretroviral naïve      | 786             | 765                           | 23.1                      | 21    | 16.5  | 0.085   |
| Specific antiretrovirals  |                 |                               |                           |       |       |         |
| Atazanavir use            | 128             | 128                           | 3.9                       | 0     | 0     | 0.024   |
| Prior history             | 29              | 28                            | 0.8                       | 1     | 0.8   | 0.94    |
| Indinavir use             | 304             | 283                           | 8.5                       | 21    | 16.5  | 0.0018  |
| Prior history             | 980             | 942                           | 28.4                      | 38    | 29.9  | 0.71    |
| Ritonavir use             | 413             | 395                           | 11.9                      | 18    | 14.2  | 0.44    |
| Prior history             | 603             | 578                           | 17.4                      | 25    | 19.7  | 0.51    |
| Tenofovir use             | 419             | 411                           | 12.4                      | 8     | 6.3   | 0.039   |
| Prior history             | 84              | 82                            | 2.5                       | 2     | 1.6   | 0.52    |
| Hepatitis C virus         | 473             | 448                           | 13.5                      | 25    | 19.7  | 0.049   |
| Antibody positive³        | 363             | 342                           | 76.7                      | 21    | 84.0  | 0.40    |
| RNA positive³             | 151             | 139                           | 31.1                      | 12    | 48.0  | 0.079   |
| RNA >800,000             | 258             | 240                           | 72.3                      | 18    | 85.7  | 0.18    |
| Genotype 1³               | 114             | 100                           | 3.0                       | 14    | 11.0  | <0.0001 |
| Hepatitis B virus         | 70              | 64                            | 62.0                      | 8     | 57.1  | 0.73    |
| Surface antigen positive¹ | 111             | 112                           | 101,121                   | 91    | 79,106 | <0.0001 |
| HIV RNA ml/min/1.73 m²    | 24              | 27                            | 24,27                     | 24    | 22,27 | 0.19    |
| Median BMI kg/m²          | 177             | 159                           | 4.8                       | 18    | 14.2  | <0.0001 |
| Diabetes mellitus         | 447             | 415                           | 12.5                      | 32    | 25.2  | <0.0001 |
| Anti hypertensive therapy | 501             | 475                           | 14.3                      | 26    | 20.5  | 0.054   |
| Lipid-lowering therapy    | 114             | 109                           | 3.3                       | 5     | 3.9   | 0.69    |

Baseline was defined at randomization into the parent trial (SMART or ESPRIT). Categorical variables are presented as N (%) and continuous variables presented as median (interquartile range).

1Data on HIV RNA and Hepatitis B surface antigen were available for 3435 participants (99.8%).

2Data on Hepatitis C antibody status were available for 3436 participants (99.8%).

3Among 473 participants with positive Hepatitis C antibody, RNA viral load was available for 471 (99.6%) and genotype was available for 353 (74.6%).

eGFR, estimated GFR calculated using the CKD-EPI formula; BMI, body mass index.

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variables (Table 3). The small number of participants with significant elevations in these markers precluded consideration of clinically relevant cutoff values. As previously reported, the systemic inflammatory markers high sensitivity C-reactive protein (hsCRP) and interleukin-6 (IL-6) were not significantly associated with progressive CKD in this population after adjustment for other

![Figure 1. Cumulative proportion of participants with progressive kidney disease.](doi:10.1371/journal.pone.0040245.g001)

**Table 2.** Baseline characteristics associated with progressive CKD in univariate and multivariate analysis.

|                        | Univariate |          |          | Multivariate |          |          |
|------------------------|------------|----------|----------|--------------|----------|----------|
|                        | Odds Ratio | 95% CI   | Odds Ratio | 95% CI       | Odds Ratio | 95% CI   |
| Hepatitis B surface antigen positive | 3.28       | 1.90–5.67 | 2.26       | 1.15–4.44    |
| Hepatitis C antibody positive        | 1.58       | 1.02–2.44 | 1.72       | 1.07–2.76    |
| Age, per 10 years            | 1.69       | 1.41–2.02 | 1.36       | 1.10–1.68    |
| Race                      |            |          |            |              |          |          |
| Black                     | 1.59       | 1.04–2.45 | 2.07       | 1.23–3.49    |
| Asian                     | 2.43       | 1.49–3.97 | 2.33       | 1.19–4.55    |
| Other                     | 0.97       | 0.45–2.11 | 1.27       | 0.57–2.83    |
| Same sex exposure         | 0.70       | 0.49–0.99 | 0.76       | 0.51–1.13    |
| CD4, per 100 cells/mm³    | 0.88       | 0.80–0.97 | 1.00       | 0.90–1.11    |
| Nadir CD4, per 100 cells/mm³| 0.79       | 0.70–0.89 | 0.84       | 0.72–0.98    |
| History of AIDS           | 1.51       | 1.05–2.17 | 1.13       | 0.74–1.72    |
| Antiretroviral naive      | 0.65       | 0.41–1.04 | 0.80       | 0.49–1.30    |
| Current or previous indinavir use | 1.68       | 1.05–2.70 | 0.76       | 0.43–1.34    |
| Antihypertensive therapy  | 2.62       | 1.75–3.90 | 1.83       | 1.14–2.94    |
| Lipid-lowering therapy    | 1.61       | 1.04–2.47 | 1.28       | 0.80–2.05    |
| eGFR, per 5 ml/min/1.73 m² | 0.82       | 0.80–0.85 | 0.85       | 0.82–0.87    |
| Enrolled in ESPRIT        | 1.77       | 1.22–2.56 | 1.77       | 0.90–3.49    |
| Date of randomization, per year | 0.80       | 0.71–0.90 | 0.81       | 0.67–0.98    |
variables (Abstract # O-271, Conference on Retroviruses and Opportunistic Infections 2012). The relationships between HBV and HCV co-infection and progressive CKD were not significantly affected by inclusion of any individual marker of hepatic fibrosis or systemic inflammation in separate multivariate models (data not shown).

Post-hoc Subgroup Analyses

Because of the unexpected relationship between HBV co-infection and progressive CKD, this relationship was further investigated after stratification by race and by clinical trial. After adjustment, HBV co-infection was associated with a greater than 6-fold increased odds of CKD in SMART and a 1.5-fold increased odds in ESPRIT, but there was no evidence that the adjusted OR was significantly different between the trials (p = 0.12 for interaction). When stratified by race, the odds of progressive CKD associated with HBV co-infection were highest in black participants (OR 14.54, 95% CI 4.16–50.85) and lowest in Asian participants (adjusted OR 1.26, 95% CI 0.36–4.47), although this difference was not statistically significant (p = 0.67 for interaction).

We also considered the use of antiviral agents with dual activity against HIV and HBV, as well as the use of antiviral agents for the treatment of HBV. Among 114 participants with HBV co-infection, 12 (10.5%) were taking tenofovir, 76 (66.7%) were taking lamivudine, and 31 reported prior use of lamivudine. There were no differences between participants with and without HBV co-infection in the use of these agents at or prior to baseline (p > 0.05 all comparisons). As described above, the inclusion of “on-treatment” variables for these agents did not affect the relationship between HBV and progressive CKD in sensitivity analysis. Among 74 ESPRIT participants with HBV, none reported the use of adefovir at baseline; data on the use of adefovir were not routinely collected in SMART. The anti-HBV agents entecavir and telbivudine were not approved for clinical use during enrollment in the parent trials.

Discussion

In this analysis of data and specimens from two large randomized HIV treatment trials, co-infection with either HCV or HBV was independently associated with progressive CKD among HIV-positive adults receiving cART. After adjusting for other important characteristics, the relationship between viral hepatitis co-infection and CKD did not appear to be mediated by mild hepatic fibrosis or by systemic inflammation. Increasing plasma HCV RNA, but not HBV DNA, was an independent predictor of progressive CKD. These results support current guidelines that consider HCV co-infection a risk factor for CKD. [4] The observed relationship between HCV viremia and CKD is also consistent with a recent report from a large European HIV cohort. [21] If confirmed in future studies, the relationship between HCV RNA and CKD may provide an additional impetus
for antiviral therapy as newer agents become available for the treatment of HCV co-infection.

Although we observed an unexpected relationship between serologic evidence of HBV co-infection and progressive CKD, this relationship did not appear to require active HBV replication at baseline. More refined stratification of baseline HBV DNA and consideration of viral rebound during follow-up were limited by the small number of patients with active HBV. In addition, HBV e antigen (HBeAg) and quantitative HBsAg were not measured, and HBV DNA was only measured in participants with serologic evidence of HBV infection. Data on the use of adefovir for the treatment of HBV were not collected in SMART, so it was not possible to exclude an effect of this potentially nephrotoxic antiviral agent. Adjustment for the use of tenofovir, which may have been used preferentially in the setting of HBV co-infection, did not change our findings.

Suppression of HBV replication with interferon or lamivudine has been associated with remission of kidney disease in some, but not all, cases of HBV-related immune complex kidney disease.[22–23] The pathogenesis of HBV-related kidney disease is hypothesized to involve the deposition of HBeAg in glomerular capillaries. [8] Although we were unable to explore this hypothesis in the current study, it is possible that circulating HBV antigens may continue to deposit in the kidney, or that previously deposited antigens may continue to trigger an immune response in the kidney, even in the absence of active HBV replication. This effect

![Figure 3. Association of Hepatitis C viremia with progressive kidney disease.](image)

Table 3. Markers of hepatic fibrosis as proposed mediators of progressive chronic kidney disease (CKD).

| Marker          | No progressive CKD | Progressive CKD | Univariate OR 95% CI | Multivariate OR 95% CI |
|-----------------|---------------------|-----------------|----------------------|------------------------|
| Hyaluronic acid | 25 (15, 44)         | 36 (17, 72)     | Per 20 higher        | 1.05, 1.02, 1.08       |
| APRI            | 0.27 (0.19, 0.40)   | 0.30 (0.21, 0.49)| Per 1 higher         | 1.02, 0.95, 1.11       |
| FIB4            | 0.86 (0.63, 1.19)   | 1.08 (0.74, 1.51)| Per 1 higher         | 1.02, 0.99, 1.06       |

Data available for $^1n=827$,
$^2n=1421$,
$^3n=1268$.
The number of participants who developed progressive kidney disease was $^1n=42$,
$^2n=73$,
$^3n=63$.
APRI, aspartate aminotransferase platelet ratio index.

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may be magnified in patients with HIV co-infection, who are less likely to clear HBV antigens even when HBV DNA is suppressed.

[24] Future studies should collect data on proteinuria, hematuria, and circulating markers of immune complex disease [8–11] in order to evaluate this hypothesis.

In addition to the novel relationships observed in this study, progressive CKD was also associated with traditional CKD risk factors. Other characteristics that were independently associated with progressive CKD included older age, black race, and hypertension, as well as lower baseline eGFR and lower nadir CD4. These findings are consistent with expert guidelines that recommend increased frequency of CKD screening in individuals with these risk factors. [4] Diabetes was rare in our population, and did not remain independently associated with progressive CKD in multivariate analysis. Female sex and body mass index (BMI) at baseline were not associated with progressive CKD in our population, in contrast to some prior studies in HIV-positive populations.[25–26] Of note, very low BMI was rare and women made up less than a quarter of our study population. While Asian race has not been associated with CKD in the setting of HIV infection, Asian nations report some of the highest incidence rates of ESRD in the general population. [27].

Strengths of the current analysis include a large patient population treated according to the standard of care for HIV infection, centralized measurement of serum creatinine, adjustment for markers of HIV disease severity, and inclusion of data on HBV and HCV viremia. Despite these strengths, several limitations should be considered when interpreting the results of this study. Most importantly, this was a secondary analysis of data from randomized clinical trials designed to evaluate non-renal outcomes. Nonetheless, all clinical events were reviewed centrally, and the majority of clinically relevant CKD events were captured based on eGFR decline. It is more likely that we misclassified acute events using the eGFR criteria; however, we obtained similar results in a sensitivity analysis that required confirmation of eGFR decline on two consecutive measures. Second, we had incomplete data on markers of hepatic fibrosis and systemic inflammation. Although we did not observe a relationship between these markers and progressive CKD, clinically significant hepatic fibrosis was rare in this population of clinical trial participants. Because of the small number of participants with elevated markers of hepatic fibrosis, we were also unable to dichotomize these markers at clinically relevant cutoffs. [13] Third, we were unable to fully adjust for cumulative exposure to tenofovir and other potentially nephrotoxic antiretroviral agents, [28] although the inclusion of exposure history was included in our adjusted analyses. A risk factor for HIV exposure in fewer than 10% of participants, and exposure history was included in our adjusted analyses. Fourth, we were unable to adjust for proteinuria and blood pressure, as these data were not collected in SMART and ESPRIT. Proteinuria is a strong predictor of CKD progression in HIV-positive individuals, [29] and HCV mono-infection has been associated with increased prevalence of proteinuria. [30] Hypertension, as defined by the use of antihypertensive medication at baseline, was associated with progressive CKD in our population. Unfortunately, data on the use of specific antihypertensive agents were not rigorously collected in these HIV treatment trials, and we were unable to consider potential risks or benefits associated with specific agents or classes. [4].

In summary, in this large cohort of HIV-positive clinical trial participants, co-infection with either HBV or HCV was independently associated with progressive CKD. The observed relationship did not appear to be mediated by early hepatic fibrosis or by increased systemic inflammation in co-infected individuals, although we were unable to exclude a role for more advanced liver disease in this relatively healthy population. Active HCV replication was an independent predictor of progressive CKD, and future trials of direct acting antivirals for HCV should consider the impact of successful antiviral treatment on the risk of CKD in co-infected individuals. Future studies are needed to confirm the observed relationship between HBV co-infection and progressive CKD.

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Author Contributions

Conceived and designed the experiments: AM MJR JN LP DG LR CMW JL. Analyzed the data: AM JN CMW. Contributed reagents/materials/analysis tools: JN LP MB JK AS JL. Wrote the paper: AM JN LP LR MB DG JK AS JL MJR CMW.

References

1. El-Sadr WM, Lundgren JD, Neaton JD, Gordin F, Abrams D, et al. (2006) CD4+ count-guided interruption of antiretroviral treatment. N Engl J Med 355: 2293–2303.
2. Choi AK, Li Y, Deeks SG, Grulke C, Volberding PA, et al. (2010) Association between kidney function and albuminuria with cardiovascular events in HIV-infected persons. Circulation 121: 651–658.
3. Wyatt CM, Hoover DR, Shi Q, Tien PC, Karim R, et al. (2011) Pre-existing albuminuria predicts AIDS and non-AIDS mortality in women initiating antiretroviral therapy. Antivir Ther 16: 591–596.
4. Gupta SK, Eustace JA, Winston JA, Boydstun II, Ahuja TS, et al. (2005) Guidelines for the management of chronic kidney disease in HIV-infected patients: recommendations of the HIV Medicine Association of the Infectious Diseases Society of America. Clin Infect Dis 40: 1559–1565.
5. Wyatt CM, Malvestutto C, Coca SG, Klotman PE, Parikh CR (2008) The impact of hepatitis C virus coinfection on HIV-related kidney disease: a systematic review and meta-analysis. AIDS 22: 1799–1807.
6. Huang JF, Chiang WL, Dai CY, Ho CK, Hwang SJ, et al. (2006) Viral hepatitis and proteinuria in an area endemic for hepatitis B and C infections: another chain of link? J Intern Med 260: 255–262.
7. Lee JJ, Lin MY, Yang YH, Lu SN, Chen HC, et al. (2010) Association of hepatitis C and B virus infection with CKD in an endemic area in Taiwan: a cross-sectional study. Am J Kidney Dis 56: 23–31.
8. Lai KN, Li PK, Lui SF, Au TC, Tam JS, et al. (1991) Membranous nephropathy related to hepatitis B virus in adults. N Engl J Med 324: 1457–1463.
9. Cheng JT, Anderson HL, Jr., Markowitz GS, Appel GB, Pogue VA, et al. (1999) Hepatitis C virus-associated glomerular disease in patients with human immunodeficiency virus infection. J Am Soc Nephrol 10: 1566–1574.
10. George F, Nadkarni GN, Estrella MM, Lucas GM, Sperati CJ, et al. (2011) The Impact of Hepatitis C Coinfection on Kidney Disease Related to Human Immunodeficiency Virus (HIV): A Biopsy Study. Medicine (Baltimore) 90: 279–295.
11. Szczeczk LA, Gupta SK, Habash R, Guasch A, Kalayjian R, et al. (2004) The clinical epidemiology and course of the spectrum of renal diseases associated with HIV infection. Kidney Int 66: 1145–1152.
12. McGuire BM, Julian BA, Byren JSJ, Cook WJ, King SJ, et al. (2006) Brief communication: Glomerulonephritis in patients with hepatitis C cirrhosis undergoing liver transplantation. Ann Intern Med 144: 735–741.
13. Butt AA, Wang X, Fried LF (2011) HCV infection and the incidence of CKD. Am J Kidney Dis 57: 396–402.
14. Abrams D, Levy Y, Losso MH, Babiker A, Collins G, et al. (2009) Interleukin-2 therapy in patients with HIV infection. N Engl J Med 361: 1548–1559.
15. Levey AS, Stevens LA, Schmid CH, Zhang YL, Castro AF, et al. (2009) A new equation to estimate glomerular filtration rate. Ann Intern Med 150: 694–712.
16. Horio M, Indaii E, Yasuda Y, Watanabe T, Matsu S (2010) Modification of the_ckd epidemiology collaboration (CKD-EPI) equation for Japanese: accuracy and use for population estimates. Am J Kidney Dis 56: 32–38.
17. Stevens LA, Claybon MA, Schmid CH, Chen J, Horio M, et al. (2011) Evaluation of the Chronic Kidney Disease Epidemiology Collaboration equation for estimating the glomerular filtration rate in multiple ethnicities. Kidney Int 79: 555–562.
18. Oberti F, Vallesia E, Planeta C, Rousset MC, Bedossa P, et al. (1997) Noninvasive diagnosis of hepatic fibrosis or cirrhosis. Gastroenterology 113: 1609–1616.
19. Valler-Pichard A, Mallet V, Naipas B, Verkarre V, Naipas A, et al. (2007) FIB-4: an inexpensive and accurate marker of fibrosis in HCV infection. comparison with liver biopsy and fibrotest. Hepatology 46: 32–36.
20. Wai CT, Greenson JK, Fontana RJ, Kaufman JD, Marnero J, et al. (2003) A simple noninvasive index can predict both significant fibrosis and cirrhosis in patients with chronic hepatitis C. Hepatology 38: 519–526.
21. Peters I, Grunt D, Lundgren JD, Rockstroh JK, Soriano V, et al. (2012) HCV viremia increases the incidence of chronic kidney disease in HIV-infected patients. AIDS, in press.