THE EFFECT OF HAART ON LIVER DISEASE PROGRESSION IN HIV INFECTION

by

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OVERALL SUMMARY

Background: Liver disease, primarily due to viral hepatitis coinfection, is a leading cause of death among HIV-infected individuals. The purpose of this research was to determine the influence of highly active antiretroviral therapy (HAART) on liver disease progression using the aspartate aminotransferase (AST)-to-platelet ratio index (APRI), a non-invasive surrogate for hepatic fibrosis.

Methods: First, we performed a cross-sectional study to assess the risk for hepatic fibrosis as measured by APRI among 4 groups of participants in the Multicenter AIDS Cohort Study (MACS): (1) HIV- and viral hepatitis-uninfected; (2) HAART-naive HIV-monoinfected; (3) viral hepatitis-monoinfected; (4) HAART-naïve HIV-viral hepatitis coinfectected. Next, among the HIV-infected participants who initiated HAART during follow-up, we conducted a prospective study of APRI change before and after HAART initiation. Finally, we performed a nested case-control study to evaluate the association of changes in APRI with liver-related mortality within our cohort.

Results: Prior to HAART initiation, the median APRI was highest in the HIV-viral hepatitis-coinfected men (1.0) followed by the viral hepatitis-monoinfected (0.43), HIV-monoinfected (0.42), and uninfected men (0.27). HIV, viral hepatitis, and the interaction between HIV and viral hepatitis infections were independently associated with higher APRI. Among the HIV-infected men, high HIV RNA and low CD4 counts were independently associated with an elevated APRI. Among HAART initiators, APRI increased across the interval 4-years to 1-year before HAART in both the HIV-viral hepatitis-coinfected and HIV-monoinfected men (34% and 17% increase, respectively). However, APRI decreased after HAART for both groups, particularly in the early post-HAART period and in those achieving an undetectable HIV RNA. Finally, among 57
men with viral hepatitis (91% with HIV) who died of liver disease, APRI was consistently higher as compared to matched controls for up to 9 years before death. APRI also rapidly increased in the 3 years prior to death among cases but remained stable among controls.

**Conclusions:** Untreated HIV infection increases the risk of hepatic fibrosis, even in the absence of viral hepatitis infection. Although the biologic basis for liver disease in the setting of HIV is unclear, ongoing HIV replication is likely an important mediator, and effective HAART mitigates liver disease progression.

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CHAPTER 1: INTRODUCTION

The overall aim of this research is to examine the role of highly active antiretroviral therapy (HAART) in liver disease progression among individuals with human immunodeficiency virus (HIV) infection. Since HAART became widely available in the mid-1990’s, there has been a substantial decline in deaths related to acquired immunodeficiency syndrome (AIDS) [1-3]. As patients with HIV are living longer, the relative burden of liver disease within this population has increased; however, the role of HAART on liver disease progression among HIV-infected individuals is unclear [3-5].

Prior to the advent of HAART, the most common causes of liver dysfunction among HIV-infected individuals were opportunistic infections and AIDS-related neoplasms [6,7]. In contrast, in the HAART-era, most liver-related morbidity and mortality is secondary to coinfection with chronic hepatitis C virus (HCV) and/or chronic hepatitis B virus (HBV) [8]. HIV infection alters the natural history of HCV and HBV infections in several ways. When acutely exposed to HCV or HBV, individuals with HIV-infection are more likely to develop chronic viral hepatitis [9, 10]. Once chronically infected with viral hepatitis, they experience an accelerated progression of hepatic fibrosis, an increased risk of developing cirrhosis, and a higher risk of liver decompensation and death compared to viral hepatitis-monoinfected individuals [11-15].

HAART can be hypothesized to have a beneficial effect on the liver, as improvement in CD4 count and suppression of HIV replication may decrease fibrosis progression. In addition, some HAART regimens have dual activity against HBV and therefore also
suppress HBV replication. On the other hand, individuals with viral hepatitis are at higher risk of HAART-related hepatotoxicity, and long-term use of certain antiretroviral medications has been hypothesized to be associated with hepatic steatosis [16, 17]. Individuals with HIV-HBV coinfection are also at risk of HBV reactivation if antiretroviral agents with anti-HBV activity are withdrawn or if resistance develops.

Studies investigating the relationship between HAART and liver disease progression have been inconsistent. In a systematic review of eleven studies involving individuals with HIV-HCV coinfection, three associated HAART with less severe fibrosis, six failed to show a link, one associated protease inhibitors with decompensated liver disease, and one showed varied effects depending on drug class [18].

The liver biopsy is the current gold standard for diagnosing and staging hepatic fibrosis. Several histologic scoring systems exist; the two most frequently used are the Ishak and the METAVIR systems. In the Ishak system, fibrosis is staged from 0 to 6 (0, no fibrosis; 1 or 2, portal fibrotic expansion; 3 or 4, bridging fibrosis; 5 or 6, cirrhosis). The METAVIR scoring system stages fibrosis from 0 to 4 (0, no fibrosis; 1, portal fibrosis; 2, portal fibrosis with few septa; 3, bridging fibrosis; 4, cirrhosis)[19]. Although considered the gold standard for diagnosing and classifying liver disease, the liver biopsy has several limitations. The size of the biopsy specimen, which usually ranges from 1-3 centimeters long, represents only 1/50,000 of the liver [20]. It is vulnerable to sampling errors, and its interpretation is operator-dependent [21, 22]. The procedure is invasive and is associated with morbidity and mortality, and it is costly. It therefore is often unrealistic to obtain liver biopsies in large epidemiologic cohort studies.
A variety of laboratory markers have been investigated as potential surrogates for hepatic fibrosis. One such marker is the AST-to-platelet ratio index (APRI) which is calculated by the following equation: 100*(AST/upper limit of normal)/platelet count (10^9/L). APRI has been validated as a surrogate marker of significant hepatic fibrosis among HCV-infected individuals with and without HIV infection [23-25]. APRI has also been shown to be predictive of liver-related mortality in a cohort of individuals with HCV monoinfection and HIV-HCV coinfection [26].

The objective of this research was to evaluate the impact of HAART on liver disease progression among participants of the Multicenter AIDS Cohort Study (MACS), using APRI as a surrogate of hepatic fibrosis. In Chapter 2, I provide a comprehensive literature review and summary of liver disease among individuals with HIV infection, with an emphasis on common causes of liver disease in this population in the HAART era. In Chapter 3, I report the prevalence of and risk factors for hepatic fibrosis among HAART-naïve HIV-infected men and HIV-uninfected men, with and without viral hepatitis infection, in the MACS using APRI as a surrogate for fibrosis. Next, in Chapter 4, I examine changes in APRI before and after HAART initiation. Finally, to assess whether longitudinal change in APRI is an appropriate surrogate for liver disease progression, in Chapter 5 I examine the association of changes in APRI with liver-related mortality in the MACS.
CHAPTER 2: LIVER DISEASE IN THE HIV-INFECTED INDIVIDUAL

CHAPTER SUMMARY

Since the advent of highly active antiretroviral therapy (HAART) for human immunodeficiency virus-1 (HIV), there has been a substantial decrease in deaths related to acquired immunodeficiency syndrome (AIDS). However, in the HAART-era liver disease is now the most common non-AIDS related cause of death among HIV-infected patients. Just as the burden of non-AIDS morbidity and mortality has changed in the HAART-era, the types of liver disease the clinician is likely to encounter among these patients have changed as well. This chapter will discuss the causes of liver disease in the HIV-infected population in the HAART-era, including chronic hepatitis C virus, chronic hepatitis B virus, medication-related hepatotoxicity, alcohol abuse, nonalcoholic fatty liver disease, and AIDS-related liver diseases.
2.1 INTRODUCTION

Managing liver disease is an increasingly important component to the care of individuals infected with human immunodeficiency virus-1 (HIV). Since the advent of highly active antiretroviral therapy (HAART) for HIV, there has been a substantial decrease in deaths related to acquired immunodeficiency syndrome (AIDS)\textsuperscript{1-3}. However, liver disease has emerged as the most common non-AIDS related cause of death among HIV-infected patients, accounting for 14-18\% of all deaths \textsuperscript{3,4} In some series, nearly half of deaths among hospitalized HIV-infected patients in the ART-era have been attributed to liver disease \textsuperscript{5, 27}.

Just as the burden of non-AIDS morbidity and mortality has changed in the ART-era, the types of liver disease the clinician is likely to encounter among these patients have also changed \textsuperscript{28}. Prior to HAART, the most common causes of liver dysfunction in HIV-infected patients were opportunistic infections, including cytomegalovirus (CMV) and mycobacterium infections, and AIDS-related neoplasms such as lymphoma and Kaposi’s sarcoma \textsuperscript{6, 7}. Since the HAART-era, however, the spectrum of liver disease among HIV-infected individuals has shifted to concomitant infection with chronic hepatitis C virus (HCV), chronic hepatitis B virus (HBV), medication-related hepatotoxicity, alcohol abuse, and nonalcoholic fatty liver disease (NAFLD) (Table 2.1) \textsuperscript{28-30}. This chapter will focus on the major causes of liver disease in the HIV-infected population in the HAART-era and will briefly review liver disease in persons with AIDS.
2.2 VIRAL HEPATITIS

Hepatitis C Virus

Most liver disease among HIV-infected individuals is secondary to coinfection with HCV and/or HBV [8]. Due to shared risk factors, coinfection with HCV and HIV is common. Reported prevalence rates of HIV-HCV coinfection vary depending on the route of HIV transmission, from 10% among those with high-risk sexual behavior to 90% with injection drug use [31]. Overall, approximately 30% of HIV-infected individuals in the United States and Europe are coinfected with HCV [32].

HIV infection alters the natural history of HCV in several ways. HIV-infected patients who are acutely infected with HCV are half as likely as HIV-uninfected individuals to clear HCV viremia [9]. Coinfected individuals also have higher HCV RNA levels, accelerated progression to hepatic fibrosis, an increased risk of developing cirrhosis, and a higher risk of decompensated liver disease once cirrhotic [11-13]. In a meta-analysis of 8 studies, HIV-HCV coinfected subjects had a two-fold increased risk of histologic cirrhosis and five-fold increased risk of decompensated liver disease compared to HCV-monoinfected individuals [33]. Studies of the role of HCV on the natural history of HIV have been conflicting. However, in an analysis of 1428 HIV-HCV coinfected individuals treated for HCV, patients who achieved sustained virologic response (SVR) had lower rates of HIV progression and non-liver mortality compared to those who did not achieve SVR, after adjusting for fibrosis, CDC clinical category, and nadir CD4 count [34].
Given both the high prevalence of HCV among the HIV-infected population and the impact of HIV on HCV-related liver disease progression, all HIV-infected patients should be tested for chronic HCV infection using 3rd generation enzyme immunoassays followed by quantitative HCV RNA testing, if positive. Although 3rd generation immunoassays are highly sensitive, even in the setting of HIV infection (>99%), HCV RNA should be checked in patients with significant risk factors for HCV and advanced immunosuppression or in whom acute infection is suspected [35]. Over the past decade, outbreaks of sexually-transmitted HCV among non-injection-drug-using men who have sex with men (MSM) have been reported in Europe, the United States, and Australia; MSM should therefore be considered at risk for acquiring HCV [36]. Because there is no available vaccine to prevent HCV infection, HIV-infected individuals who test negative for HCV should be counseled to avoid risk factors for HCV infection. For individuals who test positive for HCV, the extent of liver disease should be determined. Aminotransferase levels are not sensitive for fibrosis in the setting of HIV infection; therefore, liver biopsy remains the preferred modality for staging disease among coinfected patients.

Due to the limitations and invasiveness of liver biopsy, non-invasive methods to determine liver disease are being actively investigated and are becoming a viable alternative to liver biopsy. A variety of laboratory markers have been studied as potential surrogates for hepatic fibrosis; most were derived from studies in individuals without HIV infection. A meta-analysis of studies of the markers in the HIV-HCV-coinfected population suggested that they may be useful in excluding cirrhosis if used at their most
sensitive thresholds; however, their diagnostic odds ratios were suboptimal [37].

Transient elastography (TE) employs ultrasound technology to estimate liver stiffness by measuring elastic shear wave velocity through the liver. In a study of 169 HIV-HCV coinfected patients, TE accurately detected significant fibrosis and cirrhosis but was less accurate in discriminating mild from significant fibrosis [38].

The decision to treat HCV in the HIV-infected patient should be made on an individual basis, as the benefits must be weighed against safety and efficacy concerns. HCV treatment should be prioritized in coinfected patients without decompensated cirrhosis who have a liver biopsy revealing portal fibrosis or more advanced disease [39]. Women of child-bearing age may desire treatment prior to becoming pregnant, as pregnancy must be avoided during and six months after anti-HCV therapy due to ribavirin teratogenicity. Because they usually have favorable treatment responses, patients with HCV genotypes 2 or 3 who are motivated and can tolerate treatment should be offered it regardless of liver disease stage. Certain IL28B genotypes respond well to treatment and so may also become an indication to treat without liver disease staging [40]. Early treatment of acute HCV infection has also been associated with improved response rates in HIV-infected individuals [41]. Patients with decompensated cirrhosis should be referred to a liver transplant center with experience in transplantation with HIV infection.

The current FDA-approved treatment for HCV in the setting of HIV-infection is pegylated interferon alfa and ribavirin, which is the standard of care based on four large randomized trials [42-45]. This regimen is less effective in HIV-infected patients, with
SVR rates ranging from 14%-38% among those with HCV genotype 1 infection and 44-73% among genotype 2 and 3 infections. Similar to HCV monoinfected individuals, genotype, baseline HCV RNA, and early response to therapy are predictors of treatment response [42]. In patients receiving HCV treatment, didanosine is contraindicated and zidovudine is not recommended, as ribavirin potentiates the risk of mitochondrial toxicity and anemia, respectively [46]. Stavudine should also be avoided in patients receiving HCV treatment because of the risk of steatosis [47]. Abacavir has been associated with decreased SVR, possibly due to competition with ribavirin as both are guanosine analogues [48-50]. However, this competitive interaction appears to be insignificant when weight-based ribavirin dosing is used [51, 52].

Although HCV-infected patients have a higher incidence of HAART-related liver toxicity, this infrequently leads to HAART discontinuation and the benefits of HAART for HIV treatment are profound; therefore, HAART should not be withheld in the coinfected population. In addition, HAART may have beneficial effects on the progression of liver disease in HIV-HCV coinfection, as improvement in CD4 count may decrease fibrosis progression, although studies investigating this have been inconsistent. A recent systematic review of 11 studies examined the impact of HAART on liver disease in HIV-HCV coinfection: three associated HAART with less severe fibrosis, six failed to show a link, one associated protease inhibitors (PI’s) with decompensated liver disease, and one showed varied effects depending on drug class [18]. In other studies, HIV viral suppression has been linked to slower fibrosis progression, and HAART has been associated with decreased liver-related mortality [53, 54].
Individuals with HCV infection and cirrhosis have an increased risk of developing hepatocellular carcinoma (HCC). The American Association for the Study of Liver Disease (AASLD) recommends screening these patients every six to twelve months with alpha-fetoprotein measurement and imaging [55]. Though separate recommendations for HIV-HCV coinfection do not exist, screening remains important in this population as HCC incidence has been increasing among HIV-infected individuals [56]. Finally, HIV-HCV-coinfected patients without immunity to hepatitis A virus (HAV) should receive vaccination, as HAV can cause a fulminant hepatitis in patients with underlying liver disease.

**Hepatitis B Virus**

Though the prevalence of HIV-HBV coinfection varies by geographic location, approximately 10% of HIV-infected individuals worldwide are also chronically infected with HBV [57]. Like HIV-HCV coinfection, HIV alters the natural history of HBV. Individuals with HIV infection are 3-6 times more likely to develop chronic HBV after an acute exposure than individuals without HIV infection, and hepatitis B surface antibody (anti-HBs) development is improved with higher CD4 cell counts [10, 58]. In addition, HIV-infected patients have a lower rate of spontaneous clearance of HBeAg, increased HBV replication, and a higher rate of loss of anti-HBs and reactivation of HBV [59]. Coinfected individuals also experience an increased progression to cirrhosis and higher liver-related mortality compared to HBV monoinfected individuals [14, 15]. The impact of HBV infection on the natural history of HIV is less clear.
All HIV-infected patients should be screened for HBV with hepatitis B surface antigen (HBsAg), anti-HBs, and hepatitis B core antibody (anti-HBc). Individuals without immunity to HBV should be vaccinated; however, response to vaccination is poor especially in patients whose CD4 cell count is <200 cells/mm$^3$ [60]. Patients should therefore also be counseled to avoid risk factors for HBV transmission. Individuals with persistent HBsAg over six months have chronic HBV and should be evaluated for treatment. Isolated anti-HBc is more common in HIV infection than in the general population; in one study, 42% of HIV-infected patients were only positive for anti-HBc [61]. Occult HBV, defined as positive HBV DNA in the setting of negative HBsAg, has also been described in HIV-infected subjects, though prevalence estimates range widely [62]. The clinical implications of isolated anti-HBc positivity and occult HBV are still unclear, but reactivation of inactive or occult HBV and reverse seroconversion (reappearance of HBsAg and HBV DNA in a patient with evidence of previously resolved infection) have been reported in HIV-infected individuals [63].

Once HIV-HBV coinfection is diagnosed, staging of liver disease is important but challenging. Though serum alanine aminotransferase levels are lower in coinfected patients, this correlates poorly with liver disease [14]. Non-invasive measures of hepatic fibrosis have not been well studied in HIV-HBV coinfection; therefore, liver biopsy remains the gold standard for disease staging.
The decision to initiate HBV treatment depends on whether the patient meets indications to treat either the HIV or HBV. Treatment regimens for either virus must consider both infections, as many anti-viral agents have dual activity, including tenofovir, lamivudine, emtricitabine, entecavir, and adefovir at doses >10mg [64]. Treatment for HBV is indicated in any patient with cirrhosis and detectable HBV DNA. Although a specific HBV DNA threshold for treatment in the absence of cirrhosis has not been determined, treatment should be considered in patients with HBV DNA ≥ 2,000 IU/mL and more than mild liver disease on biopsy [64].

If there is no indication to treat either infection, the patient should be monitored closely. If treatment is indicated for either HIV or HBV, HAART should be initiated and should include the combination of tenofovir and emtricitabine (Truvada) or tenofovir and lamivudine [65]. If tenofovir is contraindicated, entecavir can be used with the HAART regimen, but then lamivudine or emtricitabine should be avoided due to overlapping resistance patterns [59]. For patients requiring treatment for HBV but in whom HAART is not feasible, options are limited by the need to avoid agents with anti-HIV activity to prevent development of drug-resistant HIV. In these patients, pegylated interferon alfa and adefovir 10 mg can be considered. Telbivudine is also a consideration, but some in vivo studies show declines in HIV RNA without emergence of drug-resistant HIV [66]. Elevated ALT and AST during the course of HAART may be due to a variety of potential causes including medications, drug-resistant HBV, HBV reactivation in the setting of medication withdrawal (especially with lamivudine withdrawal due to HIV resistance via
the M184V mutation), loss of HBeAg, or the immune reconstitution inflammatory syndrome (IRIS).

Screening for HCC among individuals with HIV-HBV coinfection should follow AASLD guidelines recommending screening for all cirrhotic HBV carriers and for certain groups of non-cirrhotic carriers [55]. The hepatitis A vaccine should also be provided to individuals without hepatitis A immunity.

2.3 MEDICATION TOXICITY

HAART-related medication toxicity

Liver toxicity is one of the most common serious adverse events associated with HAART [16]. The clinical presentation can range from mild asymptomatic increases in serum transaminases to overt liver failure [67]. In retrospective studies, the incidence of HAART-related severe hepatotoxicity is approximately 10%, and life-threatening events occur at a rate of 2.6 per 100 person years [17, 68].

There are four primary mechanisms by which HAART can lead to liver damage: direct drug toxicity and/or drug metabolism, hypersensitivity reactions, mitochondrial toxicity, and IRIS [17, 69]. IRIS is characterized by the paradoxical worsening of preexisting infectious diseases due to rapid immune restoration in the setting of successful HIV RNA suppression. The syndrome generally manifests within the first two months of HAART initiation and is accompanied by a precipitous decline in HIV RNA and rise in CD4 count. In patients with viral hepatitis, immune restoration can lead to a clinical hepatitis
due to the immune response to the virus. There have been case reports of clinical flares of HBV in the setting of HAART initiation, even with regimens including anti-HBV activity, and of rapidly progressive HCV-related cirrhosis associated with HAART-related immune restoration [70, 71].

Coinfection with HBV or HCV has consistently been associated with increased risk of HAART-related hepatotoxicity [16, 17]. Other risk factors associated with HAART-related liver injury include pre-existing advanced fibrosis, pre-treatment elevated ALT or AST, alcohol abuse, older age, female gender, first exposure to HAART, significant increase in CD4 cell count after HAART initiation, concomitant tuberculosis medications, and cocaine use [17, 69, 72].

While all antiretroviral drugs have some risk of hepatotoxicity, some are more implicated than others, and classes of drugs have characteristic patterns of injury (Table 2.2). The nonnucleoside reverse transcriptase inhibitors (NNRTI’s) typically cause either hypersensitivity reactions or direct drug toxicity and therefore have two peaks of onset: within days to weeks or several months after initiation [17]. Nevirapine (NVP) is the NNRTI most associated with hepatotoxicity, though hypersensitivity reactions resulting in liver failure have been reported with the newer NNRTI etravirine [65]. Efavirenz can also cause hepatotoxicity but does so less frequently than NVP or etravirine.

Hepatotoxicity associated with PI’s generally occurs weeks to months after drug initiation. Full-dose ritonavir (RTV) was strongly associated with hepatotoxicity but is no
longer used. The low-dose RTV used to boost levels of other PI’s does not appear to increase the risk of hepatotoxicity [73]. However, clinical hepatitis and liver failure have been reported with the newer PI tipranavir in combination with RTV boosting [17, 65]. Atazanavir and indinavir both commonly cause an indirect hyperbilirubinemia, which is not associated with liver injury and does not require treatment discontinuation [74].

The nucleoside reverse transcriptase inhibitors (NRTI’s) are associated with mitochondrial toxicity due to their ability to inhibit mitochondrial polymerase γ. Clinically this presents with hepatic steatosis and lactic acidosis from weeks to months after initiation. Stavudine, didanosine (ddI), and zidovudine are the most frequently implicated. Prolonged ddI use has also been associated with cryptogenic liver disease and recently has been linked to noncirrhotic portal hypertension and esophageal varices [75, 76]. Though less associated with mitochondrial toxicity, abacavir may cause hypersensitivity reactions especially in HLA-B*5701 positive patients. Finally, lamivudine, emtricitabine, and tenofovir can lead to HBV reactivation and severe acute hepatitis if withdrawn in an HBV-infected patient or if resistance develops.

The fusion inhibitor enfuvirtide has been rarely associated with hypersensitivity reactions, and the newer drug maraviroc, a CCR5 inhibitor, carries a black box warning for hepatotoxicity due to hypersensitivity.

Given the relatively high incidence of HAART-related hepatotoxicity, all patients should have baseline ALT and AST checked followed by regular monitoring every 3 months.
Patients should be educated regarding symptoms of hepatitis and hypersensitivity reactions. If an adverse liver event occurs, HAART should be discontinued in patients with symptoms, jaundice and elevated direct hyperbilirubinemia, grade 4 hepatotoxicity (ALT/AST >10 times upper limit of normal), or severe lactic acidosis [65]. Mild asymptomatic ALT or AST elevations usually spontaneously resolve without drug discontinuation (Table 2.3).

**Non-HAART-related medication toxicity**

HIV-infected patients are often prescribed a number of non-HAART medications that can have adverse liver effects either alone or in combination (Table 2.4).

### 2.4 ALCOHOLIC LIVER DISEASE

Although alcoholic liver disease is responsible for nearly half of all deaths due to chronic liver disease in the US, the role of alcohol abuse on liver disease in HIV-infected populations has not been well defined. In one study of 2864 HIV-infected adults in the US, 8% of the entire cohort and 15% of current alcohol drinkers were classified as heavy drinkers, which is almost twice as prevalent as in the general population [77].

Active alcohol intake is known to be associated with faster liver disease progression in HCV monoinfection [78]. In one study of HIV-HCV coinfected patients, excessive alcohol use was associated with elevated HCV RNA levels [79]. In another study of 1358 HIV-infected individuals at an urban center, 10% reported hazardous drinking, which was independently associated with an elevated surrogate for hepatic fibrosis [80]. These
results suggest that alcohol abuse is prevalent among HIV-infected individuals and can independently contribute to liver disease progression. As a modifiable risk factor for liver disease, it is important that physicians provide counseling regarding alcohol consumption in this population.

2.5 NONALCOHOLIC FATTY LIVER DISEASE

Nonalcoholic fatty liver disease (NAFLD) refers to fat deposition in hepatocytes, or steatosis, in individuals with little or no alcohol use. When accompanied by inflammation and fibrosis, it is referred to as nonalcoholic steatohepatitis (NASH). The prevalence of NAFLD in the US population ranges from 17-33%, and risk factors include obesity, hyperglycemia, diabetes mellitus, and hypertriglyceridemia [81]. Recently, mounting evidence suggests that the prevalence of hepatic steatosis in HIV-infected patients is high, especially in patients with chronic HCV or on NRTI’s [69]. Most of the prevalence data come from studies in HIV-HCV-coinfected individuals, with rates of steatosis in this population ranging from 40-69% [47, 82]. However, in a recent study of 216 HIV-infected patients without viral hepatitis coinfection, 31% had NAFLD diagnosed, although most were diagnosed with ultrasound rather than liver biopsy [83].

Metabolic abnormalities are extremely common in HIV-infected persons on HAART, especially NRTI-PI combinations. These include insulin resistance, dyslipidemia, hypertriglyceridemia, and lipodystrophy, a disorder of peripheral fat distribution resulting in lipatrophy and visceral adiposity [84]. NRTI’s can also lead to hepatic steatosis via inhibition of mitochondrial DNA replication, resulting in triglyceride accumulation in the
liver [85]. Hypertriglyceridemia, low HDL, and low total cholesterol have also been independently associated with HIV infection and may be mediated by cytokines like interferon-alfa [86]. These metabolic abnormalities have been associated with the development of NASH in HIV-infected patients [87].

The natural history of NAFLD in HIV infection is unknown. In the general population, approximately 10-15% of patients with simple steatosis progress to NASH, and 15-20% of these patients progress to cirrhosis [88]. In general, steatosis alone is not concerning for liver damage, but it may exacerbate underlying chronic liver disease. In HCV-monoinfected patients, steatosis is associated with faster progression of fibrosis and decreased response to treatment [89]. Similarly, in cohorts of HIV-HCV coinfection, hepatic steatosis has been associated with more advanced liver fibrosis [47, 82]. With continued investigation and research into NAFLD, its impact on liver disease progression in HIV-infected individuals will likely be further elucidated.

2.6 NODULAR REGENERATIVE HYPERPLASIA

Nodular regenerative hyperplasia (NRH) is a rare condition characterized by multiple small regenerative nodules in the liver parenchyma. NRH has recently become increasingly recognized in HIV-infected patients with cryptogenic liver disease [90]. Though the etiology is unclear, both ddI use and thrombophilia have been associated with the disease [90, 91]. NRH should be considered in HIV-infected patients with portal hypertension of unclear etiology, especially those on ddI.
2.7 AIDS-RELATED LIVER DISEASE

**AIDS Cholangiopathy**

AIDS cholangiopathy occurs when infection-related strictures in the biliary tract lead to biliary obstruction. It typically presents with right upper quadrant pain (RUQ) and a markedly increased alkaline phosphatase with a less elevated bilirubin and normal or slightly increased transaminases. Patients may also have fever, nausea, vomiting, and diarrhea; jaundice is uncommon [92]. It is usually seen in low CD4 counts (<100/mm³). Consequently, although previously relatively common among HIV-infected patients, it is much less common in the HAART-era. Indeed, in a recent retrospective study of 94 patients diagnosed with AIDS cholangiopathy at an urban hospital between 1983 and 2001, only 13 were diagnosed after 1996 [93].

The most common infection associated with AIDS cholangiopathy is *Cryptosporidium parvum*, followed by CMV. *Microsporidium*, *Cyclospora cayetanensis*, *Mycobacterium avium intracellulare*, and *Histoplasma capsulatum* have all been reported with AIDS cholangiopathy as well [92]. Ultrasound or magnetic resonance cholangiopancreatography may reveal intrahepatic and common bile duct dilation with terminal stenosis. However, endoscopic retrograde cholangiopancreatography remains the gold standard for diagnosis. Biopsies of the papilla and bile duct as well as bile duct brushings may help identify the infectious cause. Sphincterotomy improves the abdominal pain but does not extend survival, and the alkaline phosphatase often remains elevated [93, 94]. The most important aspect to treatment of AIDS cholangiopathy is HAART administration, as survival after diagnosis is poor without HAART [93].
**Acalculous cholecystitis**

Acalculous cholecystitis has been well documented in HIV infection and is usually associated with CMV or *Cryptosporidium*, although other infections including *Isospora* and microsporidia have been implicated [95, 96]. Patients typically present with RUQ abdominal pain and fever with cholestasis; leukocytosis is often not present. Imaging reveals a thickened, distended, acalculous gallbladder, and HIDA scan often shows a nonfunctioning gallbladder [96]. Cholecystectomy is the treatment of choice.

**AIDS-related Neoplasms**

The AIDS-defining malignancies non-Hodgkin lymphoma (NHL) and Kaposi’s sarcoma (KS), involve the liver in 33% and 9% of cases respectively [97, 98]. Hepatic involvement of NHL may present with asymptomatic liver function test abnormalities, although patients may develop abdominal pain or jaundice. Hepatic involvement of KS rarely causes symptoms or mortality [98].

**Opportunistic Infections**

Several opportunistic infections have been associated with hepatic involvement in advanced AIDS (Table 2.5). Of these, *Mycobacterium avium* complex (MAC) is the most common. It is usually characterized histologically by acid-fast bacilli-containing poorly formed granulomas, although mass lesions have been described [98, 99]. Patients often present with nausea, diarrhea, and abdominal pain. Alkaline phosphatase is usually disproportionately increased [100]. Hepatic involvement of *Mycobacterium tuberculosis*,

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including liver abscesses, has been reported in approximately 8% of patients with extrapulmonary tuberculosis and HIV infection [99, 101]. CMV is one of the most common opportunistic infections involving the liver detected on autopsy of patients with advanced AIDS but rarely results in a clinical hepatitis [98, 100]. When CMV presents as hepatitis, patients usually have a mild transaminitis, fever, malaise, weight loss, and hepatomegaly.

Hepatic involvement of fungal infections, including *Cryptococcus neoformans*, *Histoplasma capsulatum*, and *Coccidioides immitis* can be seen in patients with AIDS and is usually detected on liver biopsy or autopsy. Though liver function tests (LFT’s) are often abnormal, the liver involvement is usually asymptomatic [102, 103]. Extrapulmonary *Pneumocystis jirovecii* involving the liver has been described and may be seen in the setting of inhaled pentamidine for prophylaxis of *Pneumocystis jirovecii* pneumonia [104]. Bacillary peliosis hepatitis is a rare disease characterized by multiple blood-filled cavities in the liver parenchyma; it has been reported in patients with AIDS and *Bartonella henselae* infection [105]. Other reported opportunistic infections involving the liver of patients with AIDS include dissemintated herpes simplex virus, human herpesvirus 6, varicella-zoster virus, Epstein-Barr virus, adenovirus, *Candida albicans*, *Aspergillus fumigatus*, *Toxoplasma gondii*, and *Strongyloides stercoralis* [98-100].

*Vanishing Bile Duct Syndrome*
The vanishing bile duct syndrome (VBDS) is an acquired disease resulting in loss of small and medium-sized intrahepatic bile ducts. Multiple causes have been identified, and there have been case reports of VBDS associated with advanced AIDS, with cases attributed to CMV viremia and medication toxicity [106, 107]. The presentation is variable and often related to cholestasis. Diagnosis is based on histology, although the work-up should include imaging to rule out extrahepatic biliary obstruction. The outcome of reported AIDS-associated VBDS cases is very poor with progression to liver failure and death [106, 107].

2.8 CONCLUSION

Liver disease among HIV-infected individuals is a common and important cause of non-AIDS related morbidity and mortality. In the HAART era, the spectrum of liver disease among patients with HIV infection has changed dramatically, shifting from opportunistic infections to sequelae of chronic infections, medication toxicities, alcohol use, and fatty liver. Management of HIV-infected patients requires recognition of these conditions and targeted diagnosis and treatment.

ADDITIONAL NOTES: This chapter was published as a first-author manuscript in Clinical Gastroenterology and Hepatology in 2010 [108].
Table 2.1. Differential diagnosis of liver disease in HIV infection in the HAART-era

<table>
<thead>
<tr>
<th>HEPATIC PARENCHYMAL DISEASE</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Infection</strong></td>
</tr>
<tr>
<td>Viral Hepatitis: HCV, HBV, HDV, HAV, HEV, CMV, EBV, HSV, VZV, HHV6</td>
</tr>
<tr>
<td>Mycobacterium avium complex</td>
</tr>
<tr>
<td>Cryptococcus neoformans</td>
</tr>
<tr>
<td>Microsporidium</td>
</tr>
<tr>
<td>Pneumocystis jirovecii</td>
</tr>
<tr>
<td>Bacillary peliosis hepatitis</td>
</tr>
<tr>
<td>Histoplasma capsulatum</td>
</tr>
<tr>
<td><strong>Nonalcoholic fatty liver disease</strong></td>
</tr>
<tr>
<td><strong>Medication toxicity</strong></td>
</tr>
<tr>
<td><strong>Alcoholic liver disease</strong></td>
</tr>
<tr>
<td><strong>Recreational Drugs</strong></td>
</tr>
<tr>
<td>Cocaine</td>
</tr>
<tr>
<td>MDMA (Ecstasy)</td>
</tr>
<tr>
<td><strong>Neoplasm</strong></td>
</tr>
<tr>
<td>Lymphoma</td>
</tr>
<tr>
<td>Kaposi's sarcoma</td>
</tr>
<tr>
<td>Hepatocellular carcinoma</td>
</tr>
<tr>
<td><strong>Nodular regenerative hyperplasia</strong></td>
</tr>
<tr>
<td><strong>Autoimmune hepatitis</strong></td>
</tr>
<tr>
<td><strong>Hemochromatosis</strong></td>
</tr>
<tr>
<td><strong>Wilson's disease</strong></td>
</tr>
<tr>
<td><strong>Alpha-1 antitrypsin deficiency</strong></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>BILIARY DISEASE</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>AIDS Cholangiopathy</strong></td>
</tr>
<tr>
<td>Cryptosporidium</td>
</tr>
<tr>
<td>CMV</td>
</tr>
<tr>
<td>Microsporidium</td>
</tr>
<tr>
<td>Cyclospora cayetanensis</td>
</tr>
<tr>
<td>Mycobacterium avium intracellulare</td>
</tr>
<tr>
<td>Histoplasma capsulatum</td>
</tr>
<tr>
<td><strong>Acalculous cholecystitis</strong></td>
</tr>
<tr>
<td>Cryptosporidium</td>
</tr>
<tr>
<td>CMV</td>
</tr>
<tr>
<td>Isospora</td>
</tr>
<tr>
<td>Microsporidium</td>
</tr>
<tr>
<td><strong>Neoplasm</strong></td>
</tr>
<tr>
<td>Lymphoma</td>
</tr>
<tr>
<td>Kaposi's sarcoma</td>
</tr>
<tr>
<td><strong>Primary sclerosing cholangitis</strong></td>
</tr>
<tr>
<td><strong>Primary biliary cirrhosis</strong></td>
</tr>
</tbody>
</table>
## Table 2.2. Most common HAART agents associated with liver injury in HIV-infected patients

<table>
<thead>
<tr>
<th>MEDICATION</th>
<th>TYPICAL DOSE</th>
<th>DOSE ADJUSTMENT FOR HEPATIC INSUFFICIENCY</th>
<th>MECHANISM OF LIVER INJURY</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>NNRTI</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nevirapine (NVP)</td>
<td>200 mg po bid</td>
<td>Child-Pugh B or C: Contraindicated</td>
<td>Hypersensitivity reaction, direct drug toxicity/drug metabolism</td>
</tr>
<tr>
<td>Etravirine (ETR)</td>
<td>200 mg po bid</td>
<td>Child-Pugh A or B: No adjustment, Child-Pugh C: Not defined</td>
<td>Hypersensitivity reaction</td>
</tr>
<tr>
<td><strong>PI</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ritonavir (RTV) full-dose</td>
<td>No longer used</td>
<td></td>
<td>Direct drug toxicity/drug metabolism</td>
</tr>
<tr>
<td>Tipranavir (TPV) + RTV low-dose</td>
<td>(TPV 500 mg + RTV 200 mg) po bid</td>
<td>Child-Pugh A: Use with caution, Child-Pugh B or C: Contraindicated</td>
<td>Direct drug toxicity/drug metabolism</td>
</tr>
<tr>
<td>Atazanavir (ATV)</td>
<td>400 mg po once daily</td>
<td>Child-Pugh B: 300 mg po once daily, Child-Pugh C: Contraindicated</td>
<td>Indirect hyperbilirubinemia: does not cause liver injury</td>
</tr>
<tr>
<td>Indinavir (IDV)</td>
<td>800 mg po q8h</td>
<td>Mild to moderate hepatic insufficiency: 600 mg po q8h</td>
<td>Indirect hyperbilirubinemia: does not cause liver injury</td>
</tr>
<tr>
<td><strong>NRTI</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stavudine (D4T)</td>
<td>≥60 kg: 40 mg po bid, &lt;60 kg: 30 mg po bid</td>
<td>Not defined</td>
<td>Mitochondrial toxicity</td>
</tr>
<tr>
<td>Zidovudine (AZT, ZDV)</td>
<td>300 mg po bid</td>
<td>Not defined</td>
<td>Mitochondrial toxicity</td>
</tr>
<tr>
<td>Didanosine (ddl)</td>
<td>Enteric coated: ≥60 kg: 400 mg po once daily, &lt;60 kg: 250 mg po once daily, Oral Solution: ≥60 kg: 200 mg po bid or 400 mg po once daily, &lt;60 kg: 150 mg po bid or 250 mg po once daily</td>
<td>No adjustment</td>
<td>Mitochondrial toxicity, cryptogenic liver disease, noncirrhotic portal hypertension</td>
</tr>
<tr>
<td>Abacavir (ABC)</td>
<td>300 mg po bid</td>
<td>Child-Pugh A: 200 mg po bid (use oral solution), Child-Pugh B or C: Contraindicated</td>
<td>Hypersensitivity reaction, especially in HLA-B*5701 positive patients</td>
</tr>
<tr>
<td>Lamivudine (3TC)</td>
<td>300 mg po once daily or 150 mg po bid</td>
<td>Child-Pugh A or C: Contraindicated, No adjustment</td>
<td>HBV reactivation due to med withdrawal or resistance</td>
</tr>
<tr>
<td>Emtricitabine (FTC)</td>
<td>Oral capsule: 200 mg po once daily, Oral solution: 240 mg po once daily</td>
<td>Not defined</td>
<td>HBV reactivation due to med withdrawal or resistance</td>
</tr>
<tr>
<td>Tenofovir (TDF)</td>
<td>300 mg po once daily</td>
<td>No adjustment</td>
<td>HBV reactivation due to med withdrawal or resistance</td>
</tr>
<tr>
<td><strong>Other</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Enfuvirtide (T20)</td>
<td>90 mg subcutaneous bid</td>
<td>Not defined</td>
<td>Hypersensitivity reaction</td>
</tr>
<tr>
<td>Maraviroc (MVC)</td>
<td>Recommended dose depends on other drugs in regimen</td>
<td>Not defined, caution advised</td>
<td>Hypersensitivity reaction, direct drug toxicity/drug metabolism</td>
</tr>
</tbody>
</table>
Table 2.3. Features associated with presentation, prevention, and management of HAART-related liver injury

<table>
<thead>
<tr>
<th>Hypersensitivity Reaction</th>
<th>Associated Drugs:</th>
<th>Prevention/Monitoring</th>
<th>Management</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NVP, ETR, RTV, T20, MVC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Onset</td>
<td>Greatest risk in first 6 weeks</td>
<td>Check ALT/AST if rash develops</td>
<td>*Discontinue all ART and all other potentially hepatotoxic medications</td>
</tr>
<tr>
<td></td>
<td>Can present through 18 weeks</td>
<td>NVP: •avoid in women with CD4 &gt;250 cells/mm³, men with CD4 &gt;400 cells/mm³</td>
<td>•Rule out other causes of symptoms</td>
</tr>
<tr>
<td></td>
<td></td>
<td>•2-week dose escalation may decrease incidence</td>
<td>•Management is supportive</td>
</tr>
<tr>
<td></td>
<td></td>
<td>•Check ALT/AST q2wks x 1st month then monthly x 2 months, then every 3 months</td>
<td>•Unknown whether other NNRTI’s can be used safely after NVP-associated hepatotoxicity</td>
</tr>
<tr>
<td></td>
<td></td>
<td>ABC: •screen for HLA-B*5701 prior to initiation; do not start ABC if positive</td>
<td>•After ABC-associated hepatotoxicity, switch to another NRTI. ABC contraindicated in future use</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Direct Drug Toxicity/Metabolism</th>
<th>Associated Drugs:</th>
<th>Prevention/Monitoring</th>
<th>Management</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>All NNRTI’s, all PI’s, most NRTI’s, MVC</td>
<td>Monitor LFT’s in NVP as above</td>
<td>*Rule out other causes of hepatotoxicity, including viral hepatitis or HBV reactivation</td>
</tr>
<tr>
<td>Onset</td>
<td>Weeks to months</td>
<td>For other agents, monitor LFT’s every 3 months, more frequently in at-risk patients (HBV or HCV coinfecion, elevated transaminases at baseline, underlying liver disease, alcohol abuse, cocaine use, use of other potentially hepatotoxic drugs, first exposure to ART)</td>
<td>Symptomatic patients:</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>•Discontinue ART and other potentially offending medications</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Once symptoms and LFT abnormalities resolve, resume ART without offending agent(s)</td>
</tr>
<tr>
<td>Clinical Manifestations:</td>
<td>May present with asymptomatic transaminase elevation</td>
<td></td>
<td>Asymptomatic patients:</td>
</tr>
<tr>
<td></td>
<td>Clinical hepatitis may present with anorexia, weight loss, fatigue, jaundice, abdominal pain, nausea, vomiting</td>
<td></td>
<td>•Mild elevations usually resolve without drug discontinuation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>If ALT &gt;5-10x ULN and elevated direct bilirubin, discontinue ART</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>If ALT &gt;10x ULN, discontinue ART</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Once LFT abnormalities resolve, resume ART without offending agent(s)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Mitochondrial Toxicity</th>
<th>Associated Drugs:</th>
<th>Prevention/Monitoring</th>
<th>Management</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NRTI’s: ddI&gt; D4T&gt; AZT/ZDV&gt; 3TC= FTC= ABV= TDF</td>
<td>Check lactate in symptomatic patients or in patients with elevated anion gap or low bicarbonate</td>
<td>Mild Symptoms:</td>
</tr>
<tr>
<td>Onset</td>
<td>Weeks to months</td>
<td></td>
<td>•Change ART regimen to NRTI with lower risk of mitochondrial toxicity or to NRTI-sparing regimen</td>
</tr>
<tr>
<td>Clinical Manifestations</td>
<td>Anorexia, abdominal pain, nausea, vomiting, weight loss, fatigue</td>
<td></td>
<td>•Closely monitor lactate after resuming NRTI</td>
</tr>
<tr>
<td></td>
<td>May progress to tachycardia, tachypnea, jaundice, muscle weakness, altered mental status, multi-organ failure</td>
<td></td>
<td>Severe Symptoms:</td>
</tr>
<tr>
<td></td>
<td>Lab abnormalities include increased lactate, low arterial pH, low bicarbonate, increased anion gap</td>
<td></td>
<td>•Discontinue ART</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>•Supportive care, which may include hemodialysis or hemofiltration, mechanical ventilation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>•IV thiamine and/or riboflavin</td>
</tr>
</tbody>
</table>
Table 2.3 continued

<table>
<thead>
<tr>
<th>IMMUNE RECONSTITUTION INFLAMMATORY SYNDROME</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Associated Drugs:</strong> Any ART</td>
</tr>
<tr>
<td><strong>Onset:</strong> First 2 months</td>
</tr>
<tr>
<td><strong>Clinical Manifestations:</strong> Nonspecific symptoms (fever, night sweats, fatigue, jaundice, nausea) If performed, liver biopsy shows hepatic necrosis with CD8+ T-cell infiltration</td>
</tr>
<tr>
<td><strong>Prevention/Monitoring:</strong> Screen for HCV and HBV prior to ART initiation (should be done in all HIV-positive patients regardless of ART) If performed, liver biopsy shows hepatic necrosis with CD8+ T-cell infiltration</td>
</tr>
<tr>
<td><strong>Management:</strong> Symptomatic patients: Discontinue ART Asymptomatic patients: Discontinue ART if AST/ALT &gt; 10 x ULN Closely monitor patients with less severe increases in AST/ALT</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>HEPATITIS B REACTIVATION</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Associated Drugs:</strong> 3TC, FTC, TDF</td>
</tr>
<tr>
<td><strong>Onset:</strong> After withdrawal of medication with anti-HBV activity or development of HBV resistance (usually months to years of therapy)</td>
</tr>
<tr>
<td><strong>Clinical Manifestations:</strong> Ranges from asymptomatic increase in LFT’s to severe fulminant hepatitis Median onset 12-16 weeks after withdrawal</td>
</tr>
<tr>
<td><strong>Prevention/Monitoring:</strong> In setting HBV, ART regimen should include TDF and FTC (Truvada) or TDF and 3TC If 3TC is withdrawn due to HIV resistance, replace it with an agent with anti-HBV activity</td>
</tr>
<tr>
<td><strong>Management:</strong> If 3TC is withdrawn due to HIV resistance, replace it with an agent with anti-HBV activity Resume anti-HBV therapy with appropriate agent based on resistance profile</td>
</tr>
</tbody>
</table>
Table 2.4. Partial list of potentially hepatotoxic non-HAART medications prescribed to HIV-infected individuals

<table>
<thead>
<tr>
<th>MEDICATION</th>
<th>PATTERN OF LIVER INJURY</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Antifungals</strong></td>
<td></td>
</tr>
<tr>
<td>Ketoconazole, Fluconazole, Amphotericin B</td>
<td>Hepatocellular injury</td>
</tr>
<tr>
<td><strong>Antibiotics</strong></td>
<td></td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>Hepatocellular injury</td>
</tr>
<tr>
<td>Azithromycin, Dapsone</td>
<td>Cholestatic injury</td>
</tr>
<tr>
<td>Trimethoprim-sulfamethoxazole</td>
<td>Mixed hepatocellular-cholestatic injury</td>
</tr>
<tr>
<td><strong>Tuberculosis treatment</strong></td>
<td></td>
</tr>
<tr>
<td>Isoniazid, Rifampin, Pyrazinamide</td>
<td>Hepatocellular injury</td>
</tr>
<tr>
<td>Ethambutol</td>
<td>Cholestatic injury</td>
</tr>
<tr>
<td><strong>Anti-virals</strong></td>
<td></td>
</tr>
<tr>
<td>Ganciclovir, Acyclovir</td>
<td>Hepatocellular injury</td>
</tr>
<tr>
<td><strong>Anabolic/Androgenic steroids</strong></td>
<td></td>
</tr>
<tr>
<td>Testosterone, Nandrolone, Oxandrolone</td>
<td>Cholestatic injury, liver tumors, peliosis hepatis</td>
</tr>
</tbody>
</table>
Table 2.5. Key points regarding infections affecting the liver in HIV-infected individuals

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>Key Points</th>
</tr>
</thead>
</table>
| **Hepatitis C Virus**             | • Screen all HIV-infected patients with HCV antibody  
• Test HCV RNA in patients with positive HCV antibody. If Ab negative, also consider if suspected acute HCV, or significant risk factors and advanced immunosuppression  
• If chronic HCV, immunize against HBV and HAV if not immune |
| **Hepatitis B Virus**             | • All HIV-infected patients should be screened with HBsAg, anti-HBs, and anti-HBc  
• Vaccinate patients without HBV immunity  
• When initiating or adjusting ART, regimen must include adequate anti-HBV coverage  
• Vaccinate against HAV if not immune |
| **Hepatitis D Virus**             | • Requires concomitant HBV infection for replication  
• Acquired during simultaneous infection with HBV or as superinfection in setting of chronic HBV  
• HDV superinfection is associated with fulminant acute hepatitis and severe chronic progressive hepatitis |
| **Hepatitis A Virus**             | • Can cause fulminant hepatitis especially in presence of underlying liver disease |
| **Hepatitis E Virus**             | • Consider in patients with travel to endemic areas; autochthonous cases have also been reported in United Kingdom, France, Germany, and the United States  
• Chronic infection has been reported in HIV-infected patients |
| **Cytomegalovirus**               | • Rarely symptomatic; may present with fever, malaise, weight loss, hepatomegaly  
• Usually mild transaminitis and mild cholestasis  
• May present as mass and mimic neoplasm on CT  
• Liver biopsy with large intranuclear and small cytoplasmic inclusions +/- granulomas |
| **Mycobacterium avium complex**   | • Presents with fever, night sweats, weight loss, abdominal pain, nausea, diarrhea, HSM  
• Marked elevation of AP (>10-20x ULN) is hallmark  
• U/S shows diffusely hyperechoic liver +/- focal lesions  
• Liver biopsy with poorly formed non-caseating granulomas, foamy histiocytes, acid-fast bacilli |
| **Cryptococcus neoformans**       | • May be asymptomatic or present with fever, RUQ pain, hepatomegaly  
• Labs usually show cholestasis with increased AP, variable bilirubin  
• May cause liver abscess  
• Liver biopsy may demonstrate ill-defined cystic areas or granulomas |
| **Mycobacterium tuberculosis**    | • Presents with fever, night sweats, weight loss, LAD, and hepatomegaly  
• AP usually elevated with mildly elevated aminotransferases and bilirubin  
• U/S shows diffusely hyperechoic liver +/- focal lesions; abscesses have also been described  
• Liver biopsy with well-formed granulomas |
| **Microsporidium**                | • May cause increased bilirubin, transaminases, and especially AP |
| **Pneumocystis jirovecii**        | • More common in patients receiving inhaled pentamidine for PCP prophylaxis  
• Usually moderate increases in aminotransferases and AP, but high elevations have been reported  
• CT may show hepatic calcifications  
• Liver biopsy with foamy nodules with pneumocystis cysts on methamine silver staining |
| **Bartonella henselae**           | • Consider in patients with a history of cat contact  
• May present with HSM, liver failure, portal hypertension, fever, anemia, LAD, and skin lesions  
• AP elevated; may develop coagulopathy  
• U/S may show irregular hyperechoic regions  
• CT may reveal multiple hypoattenuating lesions of varying size  
• Liver biopsy with multiple blood-filled cavities of varying size |
| **Histoplasma capsulatum**        | • May be asymptomatic or present with constitutional symptoms, LAD and HSM, multisystem organ failure in fulminant cases  
• Labs usually show cholestasis with increased AP, variable bilirubin  
• Liver biopsy with poorly formed granulomas, rounded yeast with budding on silver staining |

Abbreviations: HSM (hepatosplenomegaly), AP (alkaline phosphatase), RUQ (right upper quadrant) LAD (lymphadenopathy), U/S (ultrasound)
CHAPTER 3: LIVER DISEASE AS MEASURED BY THE AST-TO-PLATELET RATIO INDEX AMONG HAART-NAÏVE MEN IN THE MULTICENTER AIDS COHORT STUDY

CHAPTER SUMMARY

**Background:** Although liver disease commonly causes morbidity and mortality among HIV-infected individuals, data are limited on its prevalence in HIV monoinfection. We used the AST-to-platelet ratio index (APRI) as a surrogate marker of hepatic fibrosis to characterize liver disease in the Multicenter AIDS Cohort Study.

**Methods:** HAART-naïve men were categorized based on their HIV and viral hepatitis status: uninfected (n=1170), HIV-monoinfected (n=509), viral hepatitis-monoinfected (n=74), and HIV-viral hepatitis-coinfected (n=66).

**Results:** The median APRI in the HIV-monoinfected group was similar to that in the hepatitis-monoinfected group (0.42 versus 0.43, p>0.05), higher than in the uninfected group (0.42 versus 0.27, p<0.001), but lower than in the coinfected group (0.42 versus 1.0, p<0.001). On multivariable analysis, HIV infection (1.39-fold increase (FI), p<0.001), viral hepatitis infection (1.52-FI, p<0.001), and the interaction between HIV and viral hepatitis infections were independently associated with a higher APRI (1.57-FI, p<0.001). Among the HIV-infected men, viral hepatitis coinfection (2.34-FI, p<0.001), HIV RNA ≥100,000 cp/mL (1.26-FI, p=0.007), and CD4 count ≤200 cells/mL (1.23-FI, p=0.022) were independently associated with a higher APRI.

**Conclusions:** HIV and viral hepatitis are independently associated with an increased APRI. Further studies are needed to understand the biological basis for the association between HIV and liver disease.
3.1 INTRODUCTION

Chronic liver disease is the most common non-acquired immunodeficiency syndrome (AIDS)-related cause of death among individuals with human immunodeficiency virus-1 (HIV) infection [4]. Coinfection with HIV and hepatitis C virus (HCV) or hepatitis B virus (HBV) is responsible for a large proportion of liver disease among HIV-infected individuals [4, 31, 32]. However, HIV-monoinfected individuals are also at risk for liver disease from a variety of other factors including opportunistic infections, AIDS-related neoplasms, alcohol and other substance abuse, non-alcoholic fatty liver disease, and medication-related hepatotoxicity [22]. Indeed, previous studies have demonstrated that 20-75% of HIV-monoinfected individuals have elevated liver enzymes [109-111]. Recent data suggest that HIV can activate hepatic stellate cells and may be able to infect hepatocytes [112, 113] potentially leading to liver disease. Despite this, there are limited data on the prevalence of and risk factors for liver disease in HIV-infected individuals without viral hepatitis infection [23, 24, 114].

Although liver biopsy is clinically the gold standard for diagnosing and staging liver disease, it is not possible to perform liver biopsies in large cohort studies because of the associated costs and risks to the patient [115]. Noninvasive surrogates of hepatic fibrosis are useful for evaluating liver disease when a liver biopsy is not feasible. The aspartate aminotransferase (AST)-to-platelet ratio index (APRI) is a validated marker of hepatic fibrosis among HCV-infected individuals with and without HIV infection [116-118]. APRI also predicts liver-related mortality among both HCV-monoinfected and HIV-HCV-co-infected individuals [26]. Using an APRI cut-off of >1.5, the sensitivity and
specificity of APRI for biopsy-confirmed significant fibrosis are 41% and 95%, respectively [23].

The objective of this study was to determine the prevalence of and risk factors associated with liver disease as measured by APRI among highly active antiretroviral therapy (HAART)-naïve HIV-monoinfected participants enrolled in the Multicenter AIDS Cohort Study (MACS), which is a cohort of men who have sex with men (MSM) and who either have or are at risk for HIV infection. For comparison, we also assessed APRI among three other groups of MACS participants according to their HIV and hepatitis status.

3.2 PATIENTS AND METHODS

Study population and design

The MACS is an ongoing prospective cohort study established in 1984 that enrolled HIV-infected and -uninfected MSM from four metropolitan areas in the United States (Baltimore, MD/Washington DC; Chicago, IL; Pittsburgh, PA; and Los Angeles, CA) during three recruitment periods: 1984-1985, 1987-1990, and 2001-2003. Details of the MACS participant recruitment, study design, and characteristics of the cohort have been described elsewhere [119, 120]. Participants were followed every 6 months for interview, physical examination, laboratory studies, and collection of biological specimens for repository storage. At entry into the MACS, HIV antibody was tested and, if negative, was tested semiannually in each participant. Specimens within the first two years of study entry into the MACS were tested for HCV antibody and hepatitis B surface antigen (HBsAg). Participants who were anti-HCV positive had HCV RNA testing performed.
This cross-sectional study compared APRI values among four groups of MACS participants: HIV- and viral hepatitis-uninfected, HIV-monoinfected, viral hepatitis-monoinfected, and HIV-viral hepatitis-coinfected. In order to examine liver disease prior to HAART, the visit one year before HAART initiation was selected for HIV-infected subjects who received HAART. For the HIV-uninfected subjects and the HIV-infected non-HAART initiators, we selected the corresponding visit by frequency matching to the calendar year of the visit for the HAART initiators. Men who were taking antiretroviral therapy (ART) that was not HAART were included. All participants provided informed consent, and the Institutional Review Boards at each site approved the study.

**Laboratory Testing and Definitions**

HIV status was determined using enzyme-linked immunosorbent (EIA) assay and confirmed with Western blot, as previously described [120]. HBsAg was performed with EIA (Abbot Laboratories, Abbott Park, Illinois). HCV antibody testing was done with a third generation EIA (ADVIA Centura HCV assay); if positive, HCV RNA testing was performed in the same sample using the COBAS AmpliPrep Taqman HCV assay or the COBAS TaqMan HCV Test (lower limit of detection 50 IU/ml and 43 IU/ml respectively; Roche Diagnostic Systems, Indianapolis, IN). Starting in 2001, AST and alanine aminotransferase (ALT) testing was performed at each visit. For subjects without an AST/ALT result from the selected visit, testing was performed at the Johns Hopkins Hospital clinical laboratory using stored serum or plasma specimens frozen at -70°C until use. Quality control testing demonstrated that AST testing of the thawed specimens was
reliable, but the ALT values were often lower in thawed compared to fresh samples. Thus, our analysis focuses on APRI, which uses AST. Chronic hepatitis B infection was defined as a positive HBsAg at the most recent visit. Chronic hepatitis C infection was defined as HCV antibody and HCV RNA positive. Viral hepatitis was defined as chronic infection with either hepatitis B or hepatitis C.

HAART was defined according to guidelines from the DHHS/Kaiser Panel [121]. The following regimens were considered to be HAART: (a) two or more nucleoside reverse transcriptase inhibitors (NRTI’s) in combination with at least one protease inhibitor (PI) or one nonnucleoside reverse transcriptase inhibitor (NNRTI); (b) one NRTI in combination with at least one PI and at least one NNRTI; (c) ritonavir and saquinavir in combination with one NRTI and no NNRTI’s; and (d) abacavir or tenofovir with three NRTI’s in the absence of both PI’s and NNRTI’s.

Body mass index (BMI) was calculated as body weight(kg)/height(m)$^2$ and was categorized by a modification of the World Health Organization classification system: normal, <25 kg/m$^2$; overweight, 25-30 kg/m$^2$; or obese, ≥30 kg/m$^2$. Age was categorized based upon the age distribution in the cohort: <40 years, 40-50 years, and ≥50 years. Since heavy alcohol use is uncommon in the MACS, we defined heavy alcohol use as a self-reported average of >2 drinks per day over the prior six months. CD4 T-cell count was measured in all participants and HIV RNA was measured in those with HIV infection. To account for a potential cohort effect, the calendar year of the MACS visit used for APRI testing was included in the analysis and was dichotomized into before or
after 2001. Other characteristics included in the analysis were age, race, and self-reported current injection drug use (IDU).

APRI was defined as 100*(AST/upper limit of normal)/platelet count (10^9/L)[116]. The upper limit of normal (ULN) was 30 units/L.

**Statistical Analysis**

Differences in characteristics across groups were compared using Wilcoxon rank-sum test (two-group comparisons) and Kruskal-Wallis rank test (three-or-more-group comparisons). Pearson’s chi-squared test was used to compare categorical variables across groups.

The primary analysis was a comparison of differences in APRI using linear regression. We natural log-transformed the APRI scores for the regression analyses because the empirical APRI distribution was right-skewed. Multivariable regression models were developed using covariates that were statistically significant (p<0.05) on univariate analysis or that were selected *a priori*. Those included in the final model were: HIV status, viral hepatitis status, the interaction between HIV and viral hepatitis infections, age, year of MACS visit, heavy alcohol use, BMI and race. The analysis was performed among the entire cohort and then restricted to the HIV-infected men to determine factors associated with liver disease in the setting of HIV. To facilitate interpretation of the results, coefficients were reported as a fold-change in APRI comparing those with and without the covariate of interest.
A secondary analysis was performed using a proportional odds logistic regression model to determine the relative odds of a higher liver fibrosis category with the following pre-established cut-offs: APRI ≤ 0.5 (mild/no fibrosis), APRI 0.5-1.5 (intermediate fibrosis), and APRI >1.5 (significant fibrosis) [116]. STATA 11.2 (StataCorp, College Station, TX) was used for the analysis.

3.3 RESULTS

Study Population

A total of 1819 men were included in this analysis of whom 1170 were uninfected with HIV or viral hepatitis, 509 were HIV-monoinfected, 74 were hepatitis-monoinfected (56 HCV only, 17 HBV only, 1 HCV-HBV co-infected), and 66 were HIV-viral hepatitis-coinfected (36 HCV, 27 HBV, 3 HCV-HBV).

The median age was 45 years with the hepatitis-monoinfected group being the oldest and the HIV-monoinfected group the youngest (median 48 and 42 years, respectively) (Table 3.1). The majority of participants were Caucasian. However, only 34% of the hepatitis-monoinfected men were Caucasian, most likely because more of these men were enrolled during the 2001-2003 recruitment period, which focused on non-Caucasian populations. The median BMI of the entire cohort was 25.4, and the BMI was highest among the hepatitis-monoinfected men. Although the hepatitis-monoinfected and HIV-hepatitis-coinfected men were more likely to use injection drugs, IDU was low overall (1%). The
median platelet count was normal in all groups but was lowest among the HIV-viral hepatitis-co-infected men (161x10^9/L), who also had the highest median AST (54 IU/mL).

Among HIV-infected individuals, HIV RNA levels were similar between men with and without viral hepatitis. Although median CD4 T-cell counts did not differ significantly between groups, HIV-viral hepatitis-co-infected men were more likely to have a CD4 count \( \leq 200 \) cells/mL than HIV-monoinfected men (29\% versus 18\%, \( p=0.03 \)). Because subject visits were chosen prior to HAART, most HIV-infected subjects (63\%) were not taking any ART. Among those taking ART, the most common class was the NRTI's (38\% of HIV-infected subjects), and within this class, zidovudine was used most frequently (24\%) (Table 3.2). A few individuals had either a PI or an NNRTI in their ART regimen (3.8\%). Among the potentially hepatotoxic non-ART medications, trimethoprim/sulfamethoxazole was used most frequently (19.5\%), followed by fluconazole (8.2\%) and dapsone (3\%) (Table 2).

Four men were on anti-HCV medication (2 HIV-coinfected). Seven HBV-infected men were taking lamivudine (all HIV-HBV-co-infected), and none were taking tenofovir or emtricitabine.

**APRI Values by HIV and Hepatitis Status**

The median APRI for the entire cohort was low at 0.31. The uninfected group had the lowest median APRI at 0.27 (interquartile range (IQR) 0.21-0.36). Notably, the median APRI values in the HIV-monoinfected group (0.42, IQR 0.30-0.59) and hepatitis-
monoinfected group (0.43, IQR 0.28-0.66) were nearly identical and were significantly higher compared to the uninfected group (0.27, p<0.001 respectively compared to the HIV-monoinfected and hepatitis mono-infected). The HIV-hepatitis-coinfected group had the highest median APRI at 1.0 (IQR 0.56-1.6, p<0.001 compared to each group).

According to the APRI values, 80% of the men (n=1,457) had no/mild fibrosis, 17% (n=317) had intermediate fibrosis, and 2.5% (n=45) had significant fibrosis. The prevalence of significant fibrosis was highest in the HIV-hepatitis-co-infected men at 30% but, interestingly, was similar between the HIV-monoinfected and the hepatitis-monoinfected men (3.5% and 4.0%, respectively) (Figure 3.1). In pairwise comparison, each of these was significantly higher than the uninfected men, among whom only 0.3% had significant fibrosis (p for each group comparison versus uninfected <0.001; for HIV-hepatitis -co-infected versus either HIV-monoinfected or hepatitis-monoinfected p<0.001).

Factors Associated with APRI

After establishing that APRI values differed significantly between the various HIV and viral hepatitis groups, we next aimed to determine factors associated with higher APRI. In univariate analysis of the entire cohort, HIV infection (1.66-fold increase, p<0.001) and chronic viral hepatitis infection (2.04-fold increase, p<0.001) were significantly associated with a higher APRI and remained so in multivariable analysis (Table 3.3). Age ≥50 years was also associated with a significantly higher APRI (1.11-fold increase, p=0.004). The possibility of a synergistic effect of infection with both HIV and viral
hepatitis was tested using an HIV-hepatitis interaction term. On multivariable analysis, HIV-hepatitis coinfection was associated with a 1.57-fold higher APRI (p<0.001) than would be expected based on the individual contributions of each infection. The same factors remained statistically significant after adding type of chronic viral hepatitis (HBV or HCV) to the model (results not shown).

After determining that HIV and viral hepatitis infections were independently and synergistically associated with higher APRI values, we next investigated whether this translated into a higher chance of being categorized as having hepatic fibrosis. We therefore performed multivariable proportional odds logistic regression and found that HIV infection (odds ratio (OR) 3.8, p<0.001), chronic viral hepatitis infection (OR 6.6, p<0.001), and age ≥50 (OR 1.6, p=0.013) were each independently associated with increased odds of a higher fibrosis category.

To investigate factors associated with higher APRI in HIV-infected subjects, the next series of analyses were limited to the 575 HIV-infected men. In univariate analysis, viral hepatitis coinfection (2.32-fold increase compared to HIV-monoinfected, p<0.001), HIV RNA≥100,000 copies/ml (1.37-fold increase, p<0.001), CD4 T-cell count ≤200 cells/mL (1.65-fold increase, p<0.001), ART monotherapy (1.24-fold increase compared to no ART, p=0.015), current trimethoprim/sulfamethoxazole use (1.26-fold increase, p=0.002), and current fluconazole use (1.76-fold increase, p<0.001) were all associated with a higher APRI (Table 3.4). After also adjusting for age, BMI, alcohol use, race, and year of MACS visit, the covariates that remained significantly associated with APRI were
viral hepatitis coinfection (2.34-fold increase, p<0.001), CD4 T-cell count ≤200 cells/mL (1.23-fold increase, p=0.022), and HIV RNA≥100,000 copies/mL (1.26-fold increase, p=0.007). Again, the same factors remained statistically significant after adding type of chronic viral hepatitis to the model (results not shown). When use of an NRTI was substituted for ART use in the multivariate model, it was not significantly associated with APRI.

Role of Platelets on APRI Values

Since thrombocytopenia is common among patients with HIV infection[122] and platelet count is used in the APRI calculation, higher APRI values in HIV-infected individuals may be due to HIV infection rather than underlying liver disease. To further explore this, we repeated the analysis after stratifying subjects into low, middle, and high platelet categories based on the distribution of platelets in the cohort: <188 x10^9/L (25th percentile), 188-263 x10^9/L (middle 50%), or >263 x10^9/L (75th percentile). HIV and hepatitis infections were each associated with significantly higher APRI values regardless of stratification by platelet count. The HIV-hepatitis interaction term was only statistically significant in the lowest platelet group. This is likely due to the small number of coinfected individuals in the models after stratification, as there were 43, 19, and 4 coinfected men in the low, middle, and high platelet groups respectively. We also repeated the analyses using ln(AST) as the outcome rather than ln(APRI) since AST itself is correlated with significant fibrosis on liver biopsy among HIV-infected individuals [119]. The inferences were similar in that both viral hepatitis and HIV were independently associated with higher values. Taken together, these analyses suggest that differences in
platelets between the groups were not responsible for the associations with higher APRI values.

3.4 DISCUSSION

This study of 1819 men demonstrates that both HIV and viral hepatitis infections are independently associated with APRI, a noninvasive marker of hepatic fibrosis. In fact, HIV-monoinfected men were much more likely to have significant fibrosis as measured by APRI than HIV-and viral hepatitis-uninfected controls, and surprisingly, their APRI scores were similar to those of the hepatitis-monoinfected men. Among the HIV-infected men, higher HIV RNA and lower CD4 T-cell count were associated with higher APRI suggesting that HIV-associated immunosuppression or opportunistic infections may be associated with liver fibrosis.

Few studies have evaluated the prevalence of liver disease among HIV-monoinfected individuals, and all rely on noninvasive measurements of hepatic fibrosis [23, 24, 109, 114, 123-126]. Our study is unique in that it directly compares HIV-monoinfected men with otherwise similar men who are HIV-uninfected, hepatitis-monoinfected, or HIV-hepatitis-coinfected. We also evaluated APRI values prior to HAART initiation in an effort to exclude the potentially confounding effects of HAART on liver disease.

Four prior studies used APRI as a surrogate for liver disease in HIV monoinfection. The prevalence of significant hepatic fibrosis in these studies ranged from 2.6% to 8.3% [23, 24, 123, 124]. Our finding of a 3.5% prevalence of significant fibrosis among HIV-
monoinfected men is, therefore, consistent with prior findings, although caution must be exercised in comparing APRI across studies as the ULN for AST used in calculations may vary. Unlike our study, the two cohorts with higher prevalence of significant fibrosis (7.4% and 8.3%) were predominantly African American and had a high proportion of IDU [23, 24]. In contrast, a third study of 540 HIV-monoinfected individuals, in which the majority were MSM without IDU, found 4% prevalence of significant hepatic fibrosis, a rate similar to ours [123]. None of these studies included hepatitis-monoinfected individuals as a comparison. It is notable that the prevalence of significant fibrosis as determined by APRI among the HIV-monoinfected men in our study was not only similar to other HIV-monoinfected cohorts, but it was also similar to the hepatitis-monoinfected men within our own cohort. In our cohort, high HIV RNA and low CD4 T-cell count were each independently associated with higher APRI values. This is consistent with prior studies of surrogate markers for hepatic fibrosis in HIV-positive cohorts [23, 24, 114]. However the mechanism by which these factors affect liver disease, especially in the absence of viral hepatitis, deserves further investigation.

While APRI has been validated as a surrogate for hepatic fibrosis in cohorts of HIV-HCV-coinfected individuals, it has not been validated in HIV monoinfection. Such a study is unlikely to be performed, however, as it would be neither feasible nor ethical to obtain liver biopsies in cohorts of HIV-monoinfected individuals without clinical indications for liver biopsy. Although high APRI in HIV may be due to thrombocytopenia, it is unlikely that our findings are primarily due to the effect of HIV on platelets. First, the results were consistent even after stratifying subjects by platelet
count. Second, after substituting AST for APRI in the models, HIV infection was significantly associated with the outcome. This finding is consistent with other studies that have found AST elevations in individuals with HIV infection even in the absence of viral hepatitis coinfection, hepatotoxic medications, or antiviral treatment [111, 127].

The mechanism behind elevated AST in HIV infection is intriguing but poorly understood and is likely multifactorial. It is possible that HIV infects hepatocytes, though evidence of this has been inconclusive [113, 128, 129]. Even without directly infecting hepatocytes, HIV can induce hepatocyte apoptosis [129]. In addition, HIV has been demonstrated to infect and activate human hepatic stellate cells, thereby promoting liver injury and fibrosis. The HIV envelope protein gp120 may also cause fibrosis by increasing hepatic stellate cell chemotaxis and expression of proinflammatory chemokines [130, 131]. Other potential causes of AST elevations include opportunistic infections and medications [22]. Though few men in our cohort were on PI’s or NNRTI’s, a sizable minority were taking NRTI’s which can cause hepatotoxicity due to their ability to inhibit mitochondrial polymerase-γ. However, neither ART nor NRTI use was associated with APRI after adjusting for HIV RNA level and CD4 count. Of the potentially hepatotoxic non-ART medications, trimethoprim/sulfamethoxazole and fluconazole were used most frequently in our cohort but were not associated with higher APRI after adjusting for degree of immunosuppression.

As expected, infection with both HIV and viral hepatitis had a synergistic effect on APRI. This is consistent with prior studies demonstrating the deleterious effect of HIV infection
on viral hepatitis-related liver disease [13, 14, 33]. In vitro experiments have shown that HIV and HCV cooperatively enhance hepatocyte apoptosis [132]. In the setting of viral hepatitis, HIV-associated immune dysregulation and increased gastrointestinal microbial translocation have also been implicated in promoting hepatic fibrosis [133, 134].

The primary limitation of this study is the reliance on an imperfect surrogate marker to assess liver disease. However, a study this size using liver biopsies is unlikely to be performed given its cost, invasiveness, and risk. A second limitation is that our primary analysis treated APRI as a continuous variable, which has not been validated. Our results were supported by our secondary analysis using APRI as a categorical variable. A third limitation is that chronic HCV- and HBV-infected subjects were categorized together in the analysis because of the relatively small number of HBV-monoinfected individuals. Even after adjusting for type of viral hepatitis, however, HIV infection, high HIV RNA and low CD4 T-cell count remained significantly associated with higher APRI. A fourth limitation is that we did not analyze FIB-4, another surrogate marker of liver disease that was derived in an HIV-HCV-coinfected cohort [135]. This marker requires ALT, which we could not obtain reliably in all of our subjects. However, in an analysis of the men with available ALT, the FIB-4 and APRI results were comparable. Lastly, since the MACS only includes men, this study cannot address whether HIV-monoinfected women would also have increased risk for liver disease.

A major strength of this study is the inclusion of otherwise similar HIV- and viral hepatitis-naïve controls. To our knowledge, only one other study has investigated liver
disease among a cohort of HIV-and HCV-positive and negative individuals. However, HBV-infected subjects were included with the uninfected controls, and a head-to-head comparison across viral disease categories was not performed [136]. Other strengths include the large number of subjects, the assessment of other factors that are associated with liver disease, and the lack of confounding from HAART.

In summary, our study demonstrates that HIV and chronic viral hepatitis are each independent risk factors for elevated APRI, there is a synergistic effect of coinfection with HIV and viral hepatitis, and HIV-related advanced immunosuppression is associated with higher APRI. Further studies are needed to understand the biological basis for liver disease in HIV-monoinfected individuals, to understand how immunosuppression affects liver disease, and to determine whether HAART mitigates liver disease.

**ADDITIONAL NOTES:** This chapter was published as a first-author manuscript in Journal of Infectious Diseases in 2012 [137].
Table 3.1. Characteristics of the cohort, by disease status

<table>
<thead>
<tr>
<th></th>
<th>No Infection (N=1170)</th>
<th>HIV Monoinfection (N=509)</th>
<th>Hepatitis Monoinfection (N=74)</th>
<th>Co-infection (N=66)</th>
<th>p-value$^1$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;40, n (%)</td>
<td>302 (26)</td>
<td>199 (39)</td>
<td>5 (6.8)</td>
<td>19 (29)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>40-49</td>
<td>456 (39)</td>
<td>217 (43)</td>
<td>37 (50)</td>
<td>33 (50)</td>
<td></td>
</tr>
<tr>
<td>≥50</td>
<td>412 (35)</td>
<td>93 (18)</td>
<td>32 (43)</td>
<td>14 (21)</td>
<td></td>
</tr>
<tr>
<td><strong>Year of MACS Visit</strong>, n (%)</td>
<td>286 (24)</td>
<td>372 (73)</td>
<td>14 (19)</td>
<td>46 (70)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>&lt;2001</td>
<td>884 (76)</td>
<td>137 (27)</td>
<td>60 (81)</td>
<td>20 (30)</td>
<td></td>
</tr>
<tr>
<td>≥2001</td>
<td>286 (24)</td>
<td>372 (73)</td>
<td>14 (19)</td>
<td>46 (70)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>Caucasian Race</strong>, n (%)</td>
<td>864 (74)</td>
<td>391 (77)</td>
<td>25 (34)</td>
<td>46 (70)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>Heavy alcohol use</strong>, n (%)</td>
<td>98 (8.5)</td>
<td>34 (6.9)</td>
<td>6 (8.3)</td>
<td>6 (9.2)</td>
<td>0.71</td>
</tr>
<tr>
<td><strong>BMI, median (IQR)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;25, n (%)</td>
<td>472 (43)</td>
<td>253 (55)</td>
<td>24 (34)</td>
<td>37 (58)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>25-30, n (%)</td>
<td>426 (38)</td>
<td>162 (35)</td>
<td>29 (41)</td>
<td>20 (31)</td>
<td></td>
</tr>
<tr>
<td>≥30, n (%)</td>
<td>212 (19)</td>
<td>45 (10)</td>
<td>18 (25)</td>
<td>7 (11)</td>
<td></td>
</tr>
<tr>
<td><strong>Current IDU, n (%)</strong></td>
<td>7 (0.6)</td>
<td>6 (1.2)</td>
<td>3 (4.2)</td>
<td>2 (3.2)</td>
<td>0.01</td>
</tr>
<tr>
<td><strong>Log10-HIV VL, median (IQR)</strong></td>
<td>4.27 (3.58, 4.83)</td>
<td></td>
<td>4.16 (3.46, 4.78)</td>
<td>0.29$^2$</td>
<td></td>
</tr>
<tr>
<td><strong>CD4 Count, median (IQR)</strong></td>
<td>394 (250, 531)</td>
<td></td>
<td>393 (160, 524)</td>
<td>0.70$^2$</td>
<td></td>
</tr>
<tr>
<td>≤200, n (%)</td>
<td>93 (18)</td>
<td></td>
<td>19 (29)</td>
<td>0.04$^2$</td>
<td></td>
</tr>
<tr>
<td><strong>Current lamivudine, n (%)</strong></td>
<td>75 (15)</td>
<td></td>
<td>11 (17)</td>
<td>0.70$^2$</td>
<td></td>
</tr>
<tr>
<td><strong>Current ART, n (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No therapy</td>
<td>297 (60)</td>
<td></td>
<td>40 (62)</td>
<td>0.72$^2$</td>
<td></td>
</tr>
<tr>
<td>Monotherapy</td>
<td>80 (16)</td>
<td></td>
<td>8 (12)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Combination therapy</td>
<td>120 (24)</td>
<td></td>
<td>17 (26)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Platelets, 10^9/L, median (IQR)</strong></td>
<td>233 (201, 269)</td>
<td>208 (171, 247)</td>
<td>209 (176, 247)</td>
<td>161 (127, 212)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>AST, IU/mL, median (IQR)</strong></td>
<td>22 (18, 27)</td>
<td>27 (22, 35)</td>
<td>31 (23, 45)</td>
<td>54 (38, 78)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>
Legend for Table 3.1
HIV, human immunodeficiency virus-1; MACS, Multicenter AIDS Cohort Study; BMI, body mass index; IQR, interquartile range; ART, antiretroviral therapy; AST, aspartate aminotransferase

1p-values are for comparison across all groups (unless specified) using Kruskall-Wallis rank test for continuous variables, Pearson chi-squared test for categorical variables

2comparison limited to HIV-infected using Wilcoxon rank-sum test for continuous variables, Pearson chi-squared test for categorical variables
Table 3.2. Current medication use among the 575 HIV-infected subjects.

<table>
<thead>
<tr>
<th>Medication</th>
<th>Current Use # (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ART</strong></td>
<td></td>
</tr>
<tr>
<td>≥1 NRTI</td>
<td>220 (38%)</td>
</tr>
<tr>
<td>Zidovudine</td>
<td>137 (24%)</td>
</tr>
<tr>
<td>Lamivudine</td>
<td>86 (15%)</td>
</tr>
<tr>
<td>Stavudine</td>
<td>58 (10%)</td>
</tr>
<tr>
<td>Didanosine</td>
<td>44 (7.7%)</td>
</tr>
<tr>
<td>Abacavir</td>
<td>1 (0.2%)</td>
</tr>
<tr>
<td>≥1 PI</td>
<td>16 (2.8%)</td>
</tr>
<tr>
<td>Ritonavir</td>
<td>2 (0.3%)</td>
</tr>
<tr>
<td>≥1 NNRTI</td>
<td>6 (1%)</td>
</tr>
<tr>
<td>Nevirapine</td>
<td>2 (0.3%)</td>
</tr>
<tr>
<td><strong>Non-ART</strong></td>
<td></td>
</tr>
<tr>
<td>Trimethoprim/Sulfamethoxazole</td>
<td>112 (19%)</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>47 (8.2%)</td>
</tr>
<tr>
<td>Dapsone</td>
<td>17 (3%)</td>
</tr>
<tr>
<td>Ketoconazole</td>
<td>11 (1.9%)</td>
</tr>
<tr>
<td>Testosterone</td>
<td>5 (0.9%)</td>
</tr>
<tr>
<td>Dehydroepiandrostone</td>
<td>4 (0.7%)</td>
</tr>
<tr>
<td>Nandrolone</td>
<td>2 (0.3%)</td>
</tr>
<tr>
<td>Oxandroline</td>
<td>2 (0.3%)</td>
</tr>
<tr>
<td>Isoniazid</td>
<td>1 (0.2%)</td>
</tr>
</tbody>
</table>

HIV, human immunodeficiency virus-1; ART, antiretroviral therapy; NRTI, nucleoside reverse transcriptase inhibitor; PI, protease inhibitor; NNRTI, non-nucleoside reverse transcriptase inhibitor
Table 3.3. Factors Associated with Difference in ln(APRI), entire cohort

<table>
<thead>
<tr>
<th>HIV Status</th>
<th>Unadjusted Fold Change in APRI (95% CI)</th>
<th>p-value</th>
<th>Adjusted Fold Change in APRI (95% CI)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uninfected</td>
<td>1</td>
<td></td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Infected</td>
<td>1.66 (1.57, 1.76)</td>
<td>&lt;0.001</td>
<td>1.39 (1.31, 1.49)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hepatitis Status</td>
<td></td>
<td></td>
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<tr>
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<tr>
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<td>2.04 (1.85, 2.25)</td>
<td>&lt;0.001</td>
<td>1.52 (1.33, 1.72)</td>
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<tr>
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HIV, human immunodeficiency virus-1; HBV, hepatitis B virus; HCV, hepatitis C virus

Model also adjusts for heavy alcohol use, body mass index, Caucasian race, and year of MACS visit.
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<th>Adjusted</th>
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<td>p-value</td>
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<td><strong>CD4 count ≤200 cells/mL</strong></td>
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<td><strong>Current ART Use</strong></td>
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HIV, human immunodeficiency virus; CI, confidence interval; ART, antiretroviral therapy

Model also adjusts for age, heavy alcohol use, body mass index, and year of MACS visit.
CHAPTER 4: THE INFLUENCE OF HAART ON LIVER DISEASE PROGRESSION AS MEASURED BY CHANGES IN THE AST-TO-PLATELET RATIO INDEX

CHAPTER SUMMARY

Background: Liver disease is a leading cause of death among HIV-infected individuals. Studies on the impact of highly active antiretroviral therapy (HAART) on liver disease progression have been conflicting. The objective of this study was to determine the influence of HAART on liver disease, as assessed by the aspartate aminotransferase to platelet ratio index (APRI), a non-invasive serum marker of hepatic fibrosis.

Methods: We performed a prospective study of APRI changes during up to 5 years before and after HAART initiation among 441 HIV-monoinfected and 53 HIV-viral hepatitis-coinfected HAART initiators in the Multicenter AIDS Cohort Study. Random effects linear regression models were used to examine the change in APRI before and after HAART initiation.

Results: On average, APRI increased in the 494 men during the 3-year interval prior to HAART initiation, and the increase was about twice as high among the HIV-viral hepatitis-coinfected men compared to the HIV-monoinfected men [34% (95% confidence interval (CI) 8% to 67%) versus 17% (95% CI 8% to 25%)], although this difference was not statistically significant (p=0.22). In both groups, APRI decreased with successful HIV RNA suppression in the immediate post-HAART interval (1-year before to 2-years after HAART initiation): 16% for HIV-monoinfected men (p<0.001 compared to pre-HAART interval) and 22% for HIV-viral hepatitis-coinfected men (p=0.01 compared to

52
pre-HAART interval). These changes after HAART initiation persisted in the coinfected men for up to 5 years.

**Conclusions:** Effective HAART improves liver disease in HIV-monoinfected and HIV-viral hepatitis-coinfected men, as measured by a decrease in APRI. The effect of HAART is most pronounced in the early post-HAART period.
4.1 INTRODUCTION

Since the advent of highly active antiretroviral therapy (HAART), liver disease has emerged as one of the most common causes of non-AIDS-related death among people with human immunodeficiency virus (HIV) type 1 infection[138]. Coinfection with the hepatitis C virus (HCV) or hepatitis B virus (HBV) accounts for the majority of liver disease among HIV-infected individuals [8]; however, HIV monoinfection may also increase the risk for hepatic dysfunction[114, 137, 139, 140]. Although the mechanism for this is unknown, elevated HIV RNA levels and advanced immunosuppression have been associated with more advanced liver dysfunction in HIV-infected individuals with and without viral hepatitis coinfection[13, 137, 141, 142].

There is evidence that HAART reduces liver disease progression in HIV-viral hepatitis coinfected individuals[53, 54, 139, 143-145]. However, it is also possible that HAART increases liver disease through direct hepatotoxicity or through other mechanisms such as long-term metabolic complications from steatohepatitis [146]. In support of the latter hypothesis, prolonged HAART exposure and, in particular, dideoxynucleoside analog use has been associated with hepatic fibrosis in HIV-infected individuals [144, 147]. Thus, prospective studies of HIV-infected individuals with and without viral hepatitis coinfection are needed to more clearly elucidate the effects of HAART on liver disease.

The aspartate aminotransferase to platelet ratio index (APRI) is a noninvasive serum marker for hepatic fibrosis that relies on two readily available serum markers, aspartate
aminotransferase (AST) and platelet count. An APRI cut-off of >1.5 is 41% sensitive and 95% specific for significant liver disease on biopsy [116]. APRI has been validated in individuals with HCV with and without HIV coinfection, and it is predictive of liver-related events in HIV-HCV infected cohorts [25, 26, 118, 148].

In a previous cross-sectional study among HAART-naïve men enrolled in the Multicenter AIDS Cohort Study (MACS), we demonstrated that HIV and viral hepatitis were independently associated with an increased APRI, and the viruses exhibited a synergistic effect on APRI [137]. We also found that HIV RNA ≥100,000 copies/mL and CD4 count ≤200 cells/mL were associated with higher APRI values. The objective of the current study was to determine the impact of HAART initiation on liver disease as measured by APRI. To accomplish this objective, we compared changes in APRI in HIV-infected MACS participants before and after HAART initiation.

4.2 METHODS

Study population and design

We nested this prospective study within the MACS, an ongoing cohort study of HIV-infected and -uninfected men who have sex with men (MSM) enrolled from four metropolitan areas in the United States (Baltimore, MD/Washington DC; Chicago, IL; Pittsburgh, PA; and Los Angeles, CA) during 3 recruitment periods: 1984-1985, 1987-1990, and 2001-2003. Details of the MACS participant recruitment, study design, and cohort characteristics have been described elsewhere [119, 120]. Participants were followed every 6 months for interview, physical examination, laboratory studies, and collection of biological specimens for repository storage.
The eligible study population included HIV-infected HAART initiators with and without viral hepatitis infection who had >5 years of follow-up during the HAART era (after 1996) and who had a serum sample available from a visit 1 year before HAART initiation and 2 years after HAART initiation (Figure 4.1). The HAART initiation date was estimated as the midpoint between the last visit at which the subject was not on HAART and the first visit at which the subject was on HAART. Among the 946 MACS participants who initiated HAART during follow-up and prior to 2007 (allowing for follow-up while on HAART), 507 (54%) met the criteria for serum availability. In these men, if serum was available, we also determined APRI at 4 years pre-HAART and at 5 years post-HAART to examine changes over a longer time period (Figure 4.1). Specifically, we evaluated APRI change for the intervals from 4-years to 1-year pre-HAART (interval A), 1-year pre-HAART to 2-years post-HAART (interval B), and 2-years to 5-years post-HAART (interval C). APRI change was calculable during at least one of these intervals for 494 of the HAART initiators. All participants provided informed consent, and the Institutional Review Boards at each site approved the study.

**Laboratory Testing and Definitions**

At entry into the MACS, HIV antibody was tested and, if negative, was tested semiannually in each participant. Testing was performed using enzyme immunoassay (EIA) and confirmed with Western blot, as previously described [120]. Hepatitis B surface antigen (HBsAg) testing was performed with EIA (Abbott Laboratories) using stored specimens from the index visit (1-year pre-HAART) [149]. HBsAg positive
subjects were considered to be HBV infected. HCV status was determined by systematically testing for HCV antibody testing using third-generation EIA (ADVIA Centura HCV assay) and HCV RNA (lower limit of detection 43 IU/mL; Taqman, Roche Diagnostic Systems) as described elsewhere[150]. For this study, HCV infection was defined as having chronic hepatitis C at the 1-year pre-HAART visit, which was determined based on being HCV antibody positive and HCV RNA positive at a minimum of two separate visits prior to or including the 1 year pre-HAART visit [150]. Subjects who were either HBV- or HCV-infected were defined as viral hepatitis-infected.

HAART was defined according to guidelines by the Department of Health and Human Services/Kaiser Panel[151]. The following regimens were considered to be HAART: (1) ≥2 nucleoside reverse transcriptase inhibitors (NRTIs) in combination with ≥1 non-nucleoside reverse transcriptase inhibitor (NNRTI) or ≥1 protease inhibitor (PI); (2) one NNRTI co-administered with 1 ritonavir (RTV)-boosted PI with or without NRTI; (3) an abacavir- or tenofovir-containing regimen of ≥3 NRTIs in the absence of both PIs and NNRTIs; (4) ≥2 RTV-boosted PIs with or without other antiretroviral drugs; and (5) an integrase or entry inhibitor with a combination of 2 other antiretroviral drugs except for 2 unboosted PIs.

Heavy alcohol use is uncommon in the MACS. Therefore, we defined moderate to heavy alcohol use as a self-reported average of ≥2 drinks per day over the prior 6 months. Body mass index (BMI) was calculated as (body weight [kg]/height [m])² and categorized by a modification of the World Health Organization’s classification system: normal, <25
kg/m²; overweight, 25 to <30 kg/m²; or obese, ≥30 kg/m². CD4 T-cell count and HIV RNA were measured at each visit using standard assays [152]. The lower limit of detection of the earliest HIV RNA assay was 500 copies/mL; this cut-off was therefore used to determine undetectable HIV RNA for analysis.

**Statistical Analysis**

Differences in characteristics at the 1-year pre-HAART visit (hereafter referred to as index visit) by hepatitis status were evaluated using Wilcoxon rank-sum test or Kruskal-Wallis rank test for continuous variables and Pearson χ² test for categorical variables. This visit was selected to be the index visit because it is the closest visit to HAART initiation and because all participants were selected to have data at this common time point. APRI was natural log (ln)-transformed because APRI was highly skewed. We examined changes in APRI using random effects linear regression models with maximum-likelihood estimation with ln(APRI) as the outcome. Linear combinations of the estimated coefficients were used to determine and compare annual change in APRI during the study intervals. Coefficients were exponentiated to produce relative differences in APRI change per year. Since HAART initiators are a heterogeneous group in terms of response to HIV therapy, we included HIV RNA in the multivariable model. The final model included the following covariates: viral hepatitis status, time interval, the interaction between viral hepatitis and time interval, HIV RNA (categorized as undetectable, detectable to 75,000 copies/mL, or ≥75,000 copies/mL), the interaction between HIV RNA and viral hepatitis status, pre-HAART CD4 count, race, and age. Analyses were performed using Stata 12.1 (StataCorp).
4.3 RESULTS

Study population

A total of 494 men were included consisting of 441 HIV-monoinfected and 53 HIV-viral hepatitis-coinfected men (24 HIV-HCV, 27 HIV-HBV, 2 HIV-HCV-HBV). The median age at the index visit was 42 years (IQR 38, 47). At the index visit, compared to the HIV-monoinfected men, those with HIV-viral hepatitis coinfection were thinner (median BMI 22.9 vs 24.5 in HIV-monoinfected p=0.01) and had lower CD4 counts (median 303 vs 354 p=0.02) (Table 4.1). They also had lower platelet counts (median 151 vs 210 p<0.01), higher AST levels (median 54 vs 27 p<0.01), and higher APRI values (median 1.11 vs 0.45 p<0.01).

Most of the men were initially placed on a PI-based HAART regimen (71%) and 87% of HAART initiators were placed on regimen containing a dideoxynucleoside analog. Of the 29 men who were HBsAg positive, 28 initiated a HAART regimen that contained lamivudine and/or tenofovir.

Change in APRI

We examined the longitudinal effects of HAART on APRI by determining the APRI change across the following intervals: 4 years to 1 year prior to HAART (interval A, n=390), 1 year prior to 2 years after HAART (interval B, n=474), or 2 years to 5 years after HAART (interval C, n=384) (Figure 4.1). All 494 men had calculable APRI values
across at least 1 of these intervals; for 319 men (65%), including 26 coinfected men, data were available to calculate APRI during all three intervals.

The unadjusted mean APRI value increased prior to HAART (interval A) for both the HIV-monoinfected (from 0.49 to 0.55) and HIV-hepatitis-coinfected (from 1.26 to 1.62) groups (p<0.01 and p=0.02, respectively, compared to the null hypothesis of no change) (Figure 4.2). In contrast, during the initial period after HAART initiation (interval B), the mean APRI declined for both the HIV-monoinfected (from 0.55 to 0.53) and coinfected men (from 1.62 to 1.31) (p=0.01 and p=0.07, respectively, compared to no change). Between 2-5 years after HAART initiation (interval C), mean APRI increased slightly for both groups, but the change was not significantly different from zero (p=0.12 for the HIV-monoinfected men and p=0.65 for the coinfected men).

To determine whether the improvement with HAART was affected by achieving an undetectable HIV RNA, we included HIV RNA at the end of each interval in the multivariable model. Among the HIV-monoinfected men, after adjusting for age, race, and pre-HAART CD4 cell count, APRI increased by 17% (95% CI 8%, 25%) prior to HAART (interval A) (Table 4.2). By comparison, during the period that included HAART initiation (interval B), the HIV-monoinfected men with undetectable HIV RNA at the end of the interval had a 16% decrease in APRI (p<0.001 compared to interval A) and the men with detectable HIV RNA up to 75,000 copies/ml at the end of interval B had a 2% decrease in APRI (p=0.02 compared to interval A). In contrast, the men with HIV RNA ≥75,000 copies/mL at the end of interval B had a 47% increase in APRI
(p=0.07 compared to interval A). In the later period after HAART initiation (interval C), APRI increased slightly while remaining below the pre-HAART level, but the change was similar to interval A irrespective of whether HIV RNA was undetectable at the end of interval C.

Among the HIV-viral hepatitis-coinfected men, after adjusting for age, race, and pre-HAART CD4 cell count, APRI increased by 34% (95% CI 8%, 67%) across interval A (Table 4.2). Across interval B, the coinfected men with undetectable HIV RNA had a 22% decrease in APRI (95% CI -38% to -2%) (p=0.01 compared to interval A) while the coinfected men with a detectable HIV RNA up to 75,000 copies/ml had a 13% decrease in APRI (95% CI -36% to 20%) (p=0.06 compared to interval A) and the coinfected men with HIV RNA ≥75,000 copies/ml at the end of interval B had an 11% decrease in APRI (95% CI -52% to 66%) (p=0.25 compared to interval A). The decline in APRI continued across interval C in the coinfected men with undetectable HIV RNA (mean APRI decrease of 8%), which remained significantly lower than interval A (p=0.03). Although the sample size precluded including type of viral hepatitis in multivariable analysis, the trends in APRI were consistent for both the HIV-HBV-coinfected and HIV-HCV-coinfected men.

**Change in AST**

Since HIV infection is associated with thrombocytopenia even in the absence of liver disease, we performed a sensitivity analysis by repeating the analyses using AST values instead of APRI. The findings were the same as in the APRI analysis (data not shown).
4.4 DISCUSSION

In this longitudinal study examining the effects of HAART on liver disease stage, we demonstrated that in the 494 subjects, APRI values increased prior to HAART initiation and then decreased after HAART initiation, suggesting that HAART has beneficial effects on the liver in both HIV-monoinfected and HIV-viral hepatitis-coinfected men. The HAART effect was particularly pronounced in the early post-HAART period and among the coinfected group, especially in those achieving an undetectable HIV RNA.

Prior studies evaluating the impact of HAART on HIV-viral hepatitis-related liver disease have yielded inconsistent results [18]. Although most of the studies are cross-sectional and rely on calculated fibrosis progression rates based on a single liver biopsy sample, some investigators have analyzed fibrosis progression using paired liver biopsies. Bonnard et al. found rapid fibrosis progression on sequential liver biopsies from 32 HIV-HCV-coinfected patients despite high CD4 counts on antiretroviral therapy [153]. In contrast, Schiavini et al. examined paired liver biopsies of 58 HIV-HCV-coinfected patients and found that although HAART was not associated with liver fibrosis progression, a drop in CD4 count by more than 20% was associated with fibrosis progression [154]. Both of these studies were retrospective, and thus selection bias is possible since there was likely a clinical reason for performing a liver biopsy. These studies are also limited by the small numbers of participants included.
Two other studies prospectively evaluated fibrosis progression rates determined by liver biopsy among 282 and 184 HIV-HCV coinfected individuals, respectively, and neither found a significant association between HAART and fibrosis progression[155, 156]. Importantly, the majority of subjects in both cohorts were on HAART at the time of first liver biopsy (69% and 82%). Thus, an effect of HAART on fibrosis progression may have been missed especially since we found that the largest decline in APRI occurred in the interval shortly after HAART initiation. Finally, Macias et al. analyzed paired liver biopsies among 135 HIV-HCV-coinfected subjects and found that although fibrosis progressed rapidly, effective antiretroviral therapy was associated with a slower rate of progression[157]. Even in these prospective biopsy-based studies, selection bias remains a possibility: among subjects who underwent a first liver biopsy, a second liver biopsy was performed in only 7% in Macias’ cohort[157] and 22% of Sterling’s cohort[156]. More recently, in a French cohort of 313 HIV-HCV-coinfected individuals who underwent repeated liver stiffness (LS) measurements, a non-invasive surrogate for hepatic fibrosis, long-term antiretroviral therapy (>114.5 months) was independently associated with lack of increase in LS over time, again suggesting that HAART ameliorates fibrosis progression in HIV-HCV coinfection[158].

In addition to avoiding liver biopsy selection bias, an advantage of our study design was our ability to capture HAART initiation and thus follow subjects longitudinally for years before and after HAART. Moodie et al. also examined change in APRI over time and found that APRI increased with each year of HAART among the HIV-HCV-coinfected subjects within the cohort[159]. However, that study did not evaluate changes in APRI
prior to HAART initiation. In addition, the majority of the coinfected individuals in that cohort used injection drugs and thus may have had a higher risk of liver disease progression. Supporting our finding of a decrease in APRI and therefore likely decrease in liver disease with HAART, longitudinal studies have found a decrease in clinical outcomes such as hepatic decompensation and liver-related death among HIV-HCV coinfected individuals treated with HAART, particularly when HIV RNA is suppressed and CD4 increases on treatment[54, 143, 145, 160].

Our prior study found that HIV monoinfection was associated with an elevated APRI, and we now demonstrate that APRI decreases in this group after HAART initiation suggesting that HAART decreases HIV-related liver dysfunction. This finding is supported by an analysis of a large cohort of HIV-infected individuals, which demonstrated an increased risk of hepatic dysfunction with low CD4 counts and detectable HIV RNA and a decreased risk of liver-related death with HAART, even after adjusting for viral hepatitis coinfection [139]. Similarly, Mendeni et al. reported that effective suppression of HIV viral replication protected against developing significant liver fibrosis, as measured by the surrogate fibrosis markers FIB-4 and APRI, among 1112 HIV-monoinfected HAART-initiators followed for a mean of 6.2 years[144]. Although our findings are consistent with both of these studies, our study is unique because we evaluated changes in APRI among the same subjects before and after HAART. In particular, we were able to demonstrate that the mean APRI values rose among the HIV-monoinfected subjects prior to HAART and that this trajectory reversed with HAART initiation. It is notable that this reversal was most prominent in men who
achieved undetectable HIV RNA and that it was only seen in the first 2 years after HAART initiation. It is possible that cumulative exposure to antiretroviral medication may slow and may even reverse some of the protective hepatic effects.

The mechanism by which HAART improves liver disease in HIV-infected individuals is not known. Certainly in men with HIV-HBV coinfection, the direct anti-viral activity of some of the nucleoside analogues against HBV can explain some of the improvement. However, this does not explain the improvement in HIV-monoinfected or HIV-HCV-coinfected men. A recent study of HIV-HCV-coinfected individuals with transient increases in HCV RNA and ALT after HAART initiation demonstrated an improvement in histologic necroinflammatory scores and a decline in HCV RNA over time, suggesting that HAART alters HCV replication[161]. HIV activates the immune system and induces cytokine changes that promote fibrosis in the setting of HCV coinfection, and it is possible that HAART ameliorates this HIV-associated immune dysregulation [133].

Additionally, HIV has direct and indirect hepatic effects, which would be mitigated with HAART. HIV might directly infect hepatocytes and hepatic stellate cells, thus promoting fibrogenesis[112, 128]. Signaling via the HIV envelope protein gp120 can also induce hepatocyte apoptosis and hepatic stellate cell activation[129, 162]. HIV infection also leads to depletion of mucosal CD4 T-cells, thereby increasing intestinal microbial translocation; the resultant increase in bacterial endotoxins such as lipopolysaccharide (LPS) is associated with increased liver fibrosis progression in the setting of HIV-HCV coinfection [134]. AIDS-related advanced immunosuppression is also associated with
loss of Kupffer cells, which clear microbial translocation products [163]. Effective HAART leads to an increase in Kupffer cell density and a decrease in circulating LPS [163, 164]. Finally, NRTIs reduce neutrophilic infiltration in the liver by blocking P2X7-mediated inflammasome activation[165]. Our observation that men with undetectable HIV RNA had the largest decreases in APRI and that the effect was reduced in a stepwise fashion with increasing HIV viral load supports the hypothesis that the beneficial hepatic effects of HAART are mediated via suppression of HIV replication. Al-Mohri et al. similarly found that HIV RNA was an independent predictor of APRI in a cohort of HIV-monoinfected and HIV-HCV-coinfected patients on HAART[123].

There are several limitations to this study. A major limitation is the reliance on a noninvasive marker as a surrogate for hepatic fibrosis. However, APRI has been shown to be predictive of all-cause mortality and liver-related events within HIV-infected cohorts[26, 123, 148, 166-168]. Furthermore, increases in APRI over time are associated with higher risk of all-cause and liver-related mortality, including in the MACS cohort[166, 169-171]. Thus, following APRI is a reliable way to assess changes in liver disease in a large cohort where liver biopsies are not possible in most subjects. We believe that the benefits of studying a large population with a non-invasive marker outweigh the limitations of not obtaining liver biopsies. Furthermore, transient elastography to measure LS was not available when the majority of the men in our cohort initiated HAART. A second limitation is that all HIV-viral hepatitis-coinfected subjects were analyzed together due to the relatively small sample size of HIV-HCV and HIV-HBV-coinfected individuals; thus, we were not able to determine whether HCV and HBV
coinfection behaved differently with HAART initiation. The APRI changes that we observed were similar, however, when stratified by type of viral hepatitis infection. Since the vast majority of the cohort was started on a regimen that included a dideoxynucleoside analog, which has been associated with hepatic steatosis, the beneficial effect of HAART in our study may have been reduced[172]. Finally, our cohort was comprised entirely of men, which limits its generalizability.

In summary, we found that APRI increases in HIV-monoinfected and HIV-hepatitis-coinfected men prior to HAART and decreases with HAART initiation. The effect of HAART was largest among coinfect ed men. Nevertheless, HIV-monoinfected men also experienced a decrease in APRI soon after HAART initiation. In both groups, the drop in APRI was inversely proportional to HIV RNA level. Taken together, these findings suggest that effective HAART has an overall beneficial impact on the liver in both HIV-monoinfected and HIV-viral hepatitis-coinfected men. Further studies are needed to determine whether the benefits are extended long-term with newer HAART regimens.
### Table 4.1. Characteristics of the study population one year prior to HAART initiation

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>HIV Monoinfected (N=441)</th>
<th>HIV-Viral Hepatitis-Coinfected (N=53)</th>
<th>p-value</th>
</tr>
</thead>
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<tr>
<td><strong>Age, median (IQR)</strong></td>
<td>43 (38, 47)</td>
<td>43 (38, 46)</td>
<td>0.76</td>
</tr>
<tr>
<td><strong>Caucasian race, n (%)</strong></td>
<td>359 (81)</td>
<td>45 (85)</td>
<td>0.53</td>
</tr>
<tr>
<td><strong>Heavy alcohol use, n (%)</strong></td>
<td>29 (7)</td>
<td>6 (12)</td>
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<td><strong>BMI, median (IQR)</strong></td>
<td>24.5 (22.5, 27.2)</td>
<td>22.9 (21.7, 26.6)</td>
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</tr>
<tr>
<td><strong>Current IDU, n (%)</strong></td>
<td>3 (0.7)</td>
<td>0</td>
<td>0.55</td>
</tr>
<tr>
<td><strong>Log_{10}-HIV VL, median (IQR)</strong></td>
<td>4.34 (3.61, 4.87)</td>
<td>4.35 (3.73, 4.87)</td>
<td>0.80</td>
</tr>
<tr>
<td><strong>CD4 count, median (IQR)</strong></td>
<td>354 (225, 499)</td>
<td>303 (130, 449)</td>
<td>0.02</td>
</tr>
<tr>
<td><strong>CD4 count ≤200, n (%)</strong></td>
<td>92 (21)</td>
<td>19 (37)</td>
<td>0.01</td>
</tr>
<tr>
<td><strong>Platelets, 10^9/L median (IQR)</strong></td>
<td>210 (169, 248)</td>
<td>151 (125, 201)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>AST, IU/mL median (IQR)</strong></td>
<td>27 (22, 35)</td>
<td>54 (37, 78)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>APRI median (IQR)</strong></td>
<td>0.45 (0.33, 0.64)</td>
<td>1.11 (0.66, 2.10)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>APRI &gt;1.5, n (%)</strong></td>
<td>26 (6)</td>
<td>19 (36)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>Initial HAART type</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PI-based, n (%)</td>
<td>306 (70)</td>
<td>38 (72)</td>
<td>0.68</td>
</tr>
<tr>
<td>NNRTI-based, n (%)</td>
<td>101 (23)</td>
<td>13 (25)</td>
<td></td>
</tr>
<tr>
<td>PI + NNRTI or &gt;3 NRTI, n (%)</td>
<td>30 (7)</td>
<td>2 (4)</td>
<td></td>
</tr>
<tr>
<td><strong>Initial dideoxynucleoside analog use</strong></td>
<td>382 (88)</td>
<td>45 (85)</td>
<td>0.58</td>
</tr>
</tbody>
</table>

IDU, injection drug use; HIV, human immunodeficiency virus-1; VL, viral load; IQR, interquartile range; AST, aspartate aminotransferase; APRI, aspartate aminotransferase to platelet ratio index; HAART, highly active antiretroviral therapy

^1 Missing for 5 HIV-monoinfected subjects
Table 4.2. Adjusted percent change in APRI across intervals A, B, and C among HIV-infected HAART initiators

<table>
<thead>
<tr>
<th>Percent Change in APRI (95% CI)</th>
<th>HIV-Monoinfected</th>
<th>HIV/Hepatitis-Coinfected</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>All</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Interval A</td>
<td>17% (8%, 25%)</td>
<td></td>
</tr>
<tr>
<td>Interval B</td>
<td>-16% (-22%, -9%)</td>
<td>-2% (-12%, 9%)</td>
</tr>
<tr>
<td>Interval C</td>
<td>8% (-1.4%, 17%)</td>
<td>7% (-7%, 23%)</td>
</tr>
<tr>
<td><strong>HIV/Hepatitis-Coinfected</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Interval A</td>
<td>34% (8%, 67%)</td>
<td></td>
</tr>
<tr>
<td>Interval B</td>
<td>-22% (-38%, -2%)</td>
<td>-13% (-36%, 20%)</td>
</tr>
<tr>
<td>Interval C</td>
<td>-8% (-28%, 18%)</td>
<td>37% (-15%, 121%)</td>
</tr>
</tbody>
</table>

1 Adjusted for age, race, and pre-HAART CD4 cell count
2 Outcomes are ln-transformed; results are back-transformed to produce estimated relative differences in APRI change/year.
3 Interval A change refers to the change in APRI from the 4-years to 1-year pre-HAART interval, interval B change refers to the change in APRI from the 1-year pre-HAART to 2-year post-HAART interval, and interval C change refers to the change in APRI from 2-years to 5-years post-HAART.
4 Refers to HIV RNA at the end of the interval:
   - HIV-monoinfected interval B HIV RNA: undetectable (n=293), detectable to 75,000 copies/mL (n=125), ≥75,000 copies/mL (n=21)
   - HIV-monoinfected interval C: undetectable (n=253), detectable to 75,000 copies/mL (n=93), ≥75,000 copies/mL (n=11)
   - HIV-viral hepatitis-coinfected interval B HIV RNA: undetectable (n=36), detectable to 75,000 copies/mL (n=14), ≥75,000 copies/mL (n=3)
   - HIV-viral hepatitis-coinfected interval C: undetectable (n=30), detectable to 75,000 copies/mL (n=7), ≥75,000 copies/mL (n=1)
5 p-value <0.05 compared to Interval A
6 Not reported due to small sample size
Figure 4.1. Diagram of study design. APRI was measured 4-years pre-HAART, 1-year pre-HAART (index visit in A), 2-years post-HAART (follow-up visit in A) and 5-years post-HAART.
Figure 4.2. Mean lnAPRI (95% confidence interval) pre- and post-HAART initiation.

*P-value for change across interval is <0.05 compared to the null hypothesis of no change.
CHAPTER 5: THE ASSOCIATION OF CHANGES IN THE AST-TO-PLATELET RATIO INDEX AND LIVER-RELATED DEATH IN THE MULTICENTER AIDS COHORT STUDY

CHAPTER SUMMARY

Background: The AST-to-platelet ratio index (APRI) is a surrogate for hepatic fibrosis. Elevated APRI has been associated with increased all-cause mortality and liver-related events among viral hepatitis-infected individuals, with or without HIV infection. The utility of longitudinal APRI changes in predicting adverse liver outcomes is unclear.

Methods: We performed a nested case-control study in the Multicenter AIDS Cohort Study. Cases were 57 men with viral hepatitis and liver-related death who had serum available within 9 years prior to death. Controls were matched 1:1 to cases by age, HIV and viral hepatitis status, race and calendar year. We measured APRI 9, 6, 3, 2, and 1 years prior to death.

Results: Most men (90%) were HIV-infected, 66% had chronic HBV and 39% had chronic HCV. Cases had significantly higher mean APRI values at each time point. Over 9 years, APRI was relatively stable in controls but increased among cases. After adjusting for CD4 count, average change in APRI of >1 per year was associated with a 9.1-times higher odds of liver death within 3 years (p=0.04).

Conclusions: APRI was consistently elevated in HIV-viral hepatitis co-infected men who died of liver disease up to 9 years before death. A rapid increase in APRI occurred in the 3 years prior to death, whereas APRI was stable in matched controls. Both APRI and change in APRI may predict adverse liver-related outcomes in HIV-viral hepatitis coinfection.
5.1 INTRODUCTION

Liver disease primarily due to coinfection with hepatitis C virus (HCV) or hepatitis B virus (HBV) is a leading cause of non-AIDS mortality among individuals living with HIV[138]. Among patients chronically infected with HCV or HBV, HIV coinfection increases the risk of cirrhosis, liver decompensation and death[108]. Identifying patients at high risk of adverse outcomes in order to treat viral hepatitis and manage cirrhosis is critical to reducing liver-related events in this population.

Although liver biopsy remains the clinical gold-standard for staging liver disease in the setting of viral hepatitis, the procedure is costly and invasive. The aspartate aminotransferase-to-platelet-ratio ratio index (APRI) incorporates two readily available serum markers, aspartate aminotransferase (AST) and platelet count, and has been validated as a surrogate for advanced fibrosis among HCV-infected individuals with and without HIV[116, 118].

Elevated APRI has been associated with increased all-cause mortality among women with HIV/HCV coinfection and with liver-related mortality in a cohort of HCV-monoinfected and HIV/HCV-coinfected adults[26, 166]. The utility of changes in APRI in predicting adverse outcomes in this patient population is unclear. The objective of this study was to determine whether changes in APRI are associated with liver-related death
among men with viral hepatitis infection (HCV or HBV) in the Multicenter AIDS Cohort Study (MACS).

5.2 METHODS

Study population and design

We performed a nested case-control study in the Multicenter AIDS Cohort Study (MACS), which is an ongoing cohort study of HIV-infected and -uninfected men who have sex with men (MSM). Men were enrolled from four metropolitan areas in the United States (Baltimore, MD/Washington DC; Chicago, IL; Pittsburgh, PA; and Los Angeles, CA) during 3 recruitment periods: 1984-1985, 1987-1990, and 2001-2003. Details of the MACS participant recruitment, study design, and cohort characteristics have been described elsewhere [119, 120]. Participants were followed every 6 months for interview, physical examination, laboratory studies, and collection of biological specimens for repository storage.

Men enrolled in the MACS between January 1, 1984 and December 31, 2010 who had evidence of chronic HCV or HBV were eligible for inclusion. Cases were all MACS participants with chronic HCV or HBV who experienced liver-related death during follow-up and who had stored serum available at several time points in the 9 years prior to death. Randomly selected controls who did not die a liver-related death were matched 1:1 to cases by age, HIV and viral hepatitis status, race, and calendar year. APRI was
calculated at 9, 6, 3, 2, and 1 year prior to death. Institutional Review Boards at each site approved the study, and all participants provided informed consent.

**Identification of Liver-Related Deaths**

Causes of death were determined from death certificates and were coded as per the International Classification of Diseases, 9th Revision (ICD-9). Liver-related deaths were those with a listed cause of death, either primary or contributing, with an ICD-9 code for HBV, HCV, viral hepatitis, hepatic failure, chronic hepatitis, alcoholic hepatitis, cirrhosis, liver failure, chronic liver disease, hepatocellular carcinoma, hepatic coma, or hepatorenal syndrome: 70.3, 70.5, 155.0, 155.2, 570.0, 571.0, 571.1, 571.2, 571.3, 571.4, 571.49, 571.5, 571.9, 572.2, 572.8, 573.3, 573.9.

**Laboratory Testing and Definitions**

HCV, HBV, and HIV status were defined at the visit closest to death for cases and the calendar-matched visit for controls. HCV status was determined by systematically testing for HCV antibody testing using third-generation enzyme immunoassay (EIA) (ADVIA Centura HCV assay) and HCV RNA (lower limit of detection 43 IU/mL; Taqman, Roche Diagnostic Systems) as described elsewhere[150]. Subjects with chronic HCV were positive for both anti-HCV and HCV RNA, as previously described[150]. Hepatitis B surface antigen (HBsAg) testing was performed with EIA (Abbott Laboratories) using stored specimens from the visit closest to death [149]. Subjects were considered to be chronically infected with HBV if they had two positive HBsAg tests at least 6 months
apart. HIV antibody testing was performed using EIA and confirmed with Western blot, as previously described [120].

AST was tested at every study visit starting in 2001. For study visits prior to 2001, testing was performed at the Johns Hopkins Hospital clinical laboratory using stored serum or plasma specimens frozen at −70°C until use. APRI was calculated using the formula

\[ \text{APRI} = \frac{(\text{AST}/\text{upper limit of normal AST}) \times 100}{\text{platelet count (}\text{10}^9/\text{L})} \]

and using an upper limit of normal for AST of 30 U/L [116].

Alcohol use was determined by self-report and was categorized as: none; low-moderate, 1-2 drinks per day for the past 12 months or 3-4 drinks per day for no more than 1 month; moderate-heavy, 3-4 drinks per day for more than 1 month or ≥5 drinks per day for less than 1 month; and binge, ≥5 drinks per day for at least 1 month.

**Statistical Analysis**

APRI values were modeled both continuously and categorically based on previously published cut-offs: low, ≤0.5; intermediate, 0.5-1.5; and high, >1.5[116]. We used paired t-test to compare natural-log-transformed (ln) APRI by case status at 9, 6, 3, 2, and 1 year prior to death. One-sample t-test was used to compare the change in APRI per year to the null hypothesis of no change for cases and controls. Finally, we used conditional logistic regression to determine the associations of APRI at each time point and APRI change with odds of death. Stata 12.1 (StataCorp) was use to perform all analyses.
5.3 RESULTS

Study population

Fifty-seven cases who died a liver-related death from 1986 and 2010 were identified and matched to controls by age, HIV and viral hepatitis status, race and calendar year. The majority of cases were HIV-infected (91%) and white (88%), with a median age of 44 years (interquartile range (IQR) 37, 51) (Table 5.1). Chronic HBV was present in 68% of cases and chronic HCV in 40%; 4 cases were HBV/HCV/HIV-coinfected.

Among HIV-infected cases and controls, median CD4 cell counts were 206 (IQR 109, 342) and 431 (IQR 315, 679), respectively (p<0.001). Only 19% of cases and 27% of controls were on highly active antiretroviral therapy (HAART) at the visit closest to death for cases or calendar year-matched visit for controls (p=0.37). There were no differences in HAART use between cases and controls at each time point prior to death. Just under half of liver-related deaths (47%) were prior to 1996, when HAART became widely available in the United States, whereas the majority (53%) occurred in the post-HAART era (Table 5.1). Liver cancer was the cause of death among 7 of the cases, 6 of whom were HIV-infected.

Among the 38 cases and 37 controls with chronic HBV infection, 10 (26%) and 10 (27%), respectively, were ever treated with an anti-HBV medication. However, HBV-related antiviral information was incomplete in our dataset and was missing for 8 cases.
and 5 controls. HCV treatment was rare: only 2 HCV-infected cases and none of the HCV-infected controls were treated with antiviral therapy during follow-up.

APRI at visits 9, 6, 3, 2, and 1 year prior to death

At each visit prior to death, cases had higher APRI values as compared to controls (Figure 5.1). Specifically, mean APRI at the visit 9 years prior to death was 1.57 for cases and 0.75 for controls (p<0.01), 6 years prior to death was 1.78 for cases and 1.06 for controls (p=0.09), 3 years prior to death was 2.24 for cases and 1.08 for controls (p=0.01), 2 years prior to death was 4.11 for cases and 1.09 for controls (p<0.01), and 1 year prior to death was 4.77 for cases and 0.89 for controls (p<0.01) (Table 2).

At each visit prior to death, a higher proportion of cases had an abnormal APRI score as compared to controls (Figure 5.2). Furthermore, the proportion of cases with APRI >1.5 steadily rose over time from 31% to 75% while remaining stable among controls (6% to 9%). Elevated APRI, particularly APRI >1.5, was consistently associated with significantly higher odds of death at each time point tested (Table 5.3).

Change in APRI

Over the course of follow-up, APRI was relatively stable among controls but increased among cases. The rate of increase for the cases was particularly pronounced in the 3 years preceding death (Figure 5.1). We therefore calculated the change in APRI per year in the
“early” interval between 9 and 3 years prior to death (calculable for 37 case pairs) and the “late” interval between 3 and 1 year prior to death (calculable for 38 case pairs). Among cases, on average, APRI increased 0.15 points per year during the early interval and 0.93 points per year during the late interval, although neither of these changes were statistically different from zero change (p=0.07 and 0.08, respectively). Among controls, the mean APRI change during the early interval was 0.05 points per year (p=0.17 compared to the null hypothesis of no change) and was -0.01 points per year during the late interval (p=0.87). The differences in mean APRI change for each interval comparing the cases and controls were not statistically significant.

Since the APRI score incorporates both platelet count and AST level, both of which can be affected by HIV and/or by ART, we also evaluated whether changes in APRI were driven primarily by either of these laboratory values. The mean platelet count was lower in cases compared to controls and declined at each time point prior to death (Figure 5.3); lower platelet count translates to a higher APRI. Conversely, mean AST was higher in cases compared to controls and increased prior to death (Figure 5.4), similar to what we found with APRI.

Using conditional logistic regression, after adjusting for CD4 count, a change in APRI >1 point per year in the late interval was associated with a 9.1-fold higher odds of death [95% confidence interval (CI) 1.2, 72.3; p=0.04]. An APRI change of >1 point per year remained significantly associated with increased odds of death after adjusting for APRI
6-years prior to death (OR 11.7; p=0.04) but not after adjusting for APRI 3-years prior to death (OR 7.2; p=0.10) (Table 5.4).

5.4 DISCUSSION
In this case-control study of 114 men with chronic HCV or HBV, of whom the majority were coinfected with HIV, APRI was consistently elevated in the men who died a liver-related death over the 9 years prior to death. Interestingly, a rapid increase in APRI occurred in the 3 years prior to death, whereas APRI was stable in matched controls. Especially in the 3 years prior to death, APRI > 0.5 increased the risk for liver-related mortality about 20-fold. Thus, both APRI and change in APRI are associated with adverse liver-related outcomes in HIV-viral hepatitis coinfection.

Liver biopsy is the gold standard for assessing hepatic fibrosis stage, which predicts survival in patients with viral hepatitis[173, 174]. However, cost, invasiveness, and risk to the patient limit the feasibility of serial fibrosis assessments using liver biopsy. Furthermore, the biopsy itself is an imperfect gold standard and is subject to sampling error and intra- and inter-observer variability[175, 176]. Noninvasive methods to assess hepatic fibrosis are therefore needed both to estimate liver disease stage and to help predict liver disease progression. Although a variety of noninvasive surrogates for hepatic fibrosis exist, including serum markers and imaging modalities, APRI is calculated using readily available laboratory values which are routinely obtained on HIV-infected patients and therefore is convenient to use in both clinical and research settings.
We found that APRI was associated with liver-related mortality on repeated measurements as far as 9 years prior to death. This is consistent with studies demonstrating that APRI is predictive of mortality in a number of different liver diseases, including chronic HBV, HCV, nonalcoholic fatty liver disease (NAFLD), and alcoholic liver disease[166, 177-182]. APRI has also been associated with all-cause mortality in HIV-infected individuals, with and without viral hepatitis coinfection, as well as in a cohort of HIV-HCV-coinfected women[166-168]. In a cohort of 303 HCV-infected individuals, including 207 coinfected with HIV, APRI predicted 3-year liver mortality with an area under the receiver operator curve (AUROC) of 0.88 (95% CI 0.80-0.93)[26]. In addition, Al-Mohri et al. found that APRI predicted liver complications in a cohort of 133 HIV-HCV-coinfected and 540 HIV-monoinfected patients with a median follow-up of 4.6 years[123].

A unique feature of our study is the assessment of APRI at multiple discrete time points over a 9 year period. We found that APRI was consistently higher in cases compared to controls, despite the fact that platelets and AST can fluctuate in the setting of HIV and viral hepatitis. Although our 9-year study window is longer than other studies evaluating the predictive value of APRI in the setting of HIV and/or viral hepatitis, Angulo et al. and Kim et al. demonstrated that APRI was predictive of mortality in individuals with NAFLD after follow-up of 9 years and 14.5 years, respectively[179, 180].
APRI steadily increased over time among cases but remained relatively constant among controls. This suggests that changes in APRI reflect liver disease progression. Interestingly, APRI increased precipitously in the 3 years prior to death. Although we did not have clinical data to correlate with the laboratory findings, this pattern is similar to the rapid reduction in hepatic function observed in patients with cirrhosis once decompensation occurs. Indeed, median survival after decompensation is only 4-5 years in cirrhotic patients with HCV and is significantly reduced in the setting of HIV coinfection[183, 184]. Two studies have demonstrated that APRI correlates with hepatic venous pressure gradient measurements in patients with cirrhosis[185, 186]. Furthermore, a recent analysis of 903 HIV-HCV-coinfected patients found that both APRI and FIB-4 performed better than liver biopsy in predicting mortality and liver-related events, suggesting that the serum markers more accurately reflect portal hypertension and hepatic dysfunction than fibrosis scoring on liver biopsy[148]. In that study, the AUROC for clinical outcomes was higher for FIB-4 than for APRI. We were unable to evaluate FIB-4 because only 30% of our population had alanine transaminase (ALT) measured on fresh serum and we found that testing of ALT on our frozen samples was unreliable[137].

A few other studies have evaluated longitudinal changes in APRI as predictive of clinical outcomes. Vergniol et al. followed 1,025 patients with chronic HCV and found that both baseline APRI and change in APRI from baseline to 3-year follow-up evaluation were associated with all-cause mortality [169]. Among 153 HIV-HCV-coinfected women in the Women’s Interagency HIV Study who died and had at least 2 calculable APRI values prior to their death, APRI increased over the 5 years prior to death in all women but the
rate of increase was significantly higher in women who died a liver-related death[166]. Although that study did not note a precipitous increase in APRI prior to death, fewer time points were evaluated, and APRI was not tested at common intervals as it was in our study. Finally, a retrospective study of 151 HIV-viral hepatitis-c coinfection patients evaluated APRI at baseline and last clinical follow-up and found a trend towards increased risk of 7-year mortality with a higher APRI change, although this was not statistically significant[170]. Advantages of our study include the prospective design and testing of APRI at discrete follow-up intervals.

Our study has several limitations. Although the nested case-control design was chosen to evaluate APRI at distinct intervals, as a consequence we are unable to directly assess the predictive value of APRI changes. In addition, the relatively small number of cases limited our ability to adjust for other potential factors associated with liver-related mortality. However, cases and controls were matched on age, race, HIV status, and calendar year. Finally, our cohort is comprised only of men, thereby limiting its generalizability.

In summary, this study supports the use of APRI to determine which individuals are at increased risk for death from chronic HBV or HCV, especially in HIV-infected patients. Furthermore, an abrupt increase in APRI is a signal that liver decompensation is likely to occur in the next several years. If our findings are confirmed in other populations, then serial APRI measurements could be used to identify patients at higher risk for liver
disease progression and adverse clinical outcomes to allow targeting for intervention, including prioritization for HCV treatment and evaluation for liver transplantation.
Table 5.1. Characteristics of cases and controls.

<table>
<thead>
<tr>
<th></th>
<th>Cases (N=57)</th>
<th>Controls (N=57)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV status^1</td>
<td>52 (91%)</td>
<td>51 (89%)</td>
</tr>
<tr>
<td>Chronic HBV^2</td>
<td>38 (68%)</td>
<td>37 (65%)</td>
</tr>
<tr>
<td>Chronic HCV</td>
<td>23 (40%)</td>
<td>21 (37%)</td>
</tr>
<tr>
<td>Non-white race</td>
<td>7 (12%)</td>
<td>7 (12%)</td>
</tr>
<tr>
<td>Age^3, median (IQR)</td>
<td>44 (37, 51)</td>
<td>44 (38, 48)</td>
</tr>
<tr>
<td>Alcohol use^3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>None</td>
<td>18 (33%)</td>
<td>11 (20%)</td>
</tr>
<tr>
<td>Low-Moderate</td>
<td>22 (41%)</td>
<td>28 (50%)</td>
</tr>
<tr>
<td>Moderate-Heavy</td>
<td>12 (22%)</td>
<td>14 (25%)</td>
</tr>
<tr>
<td>Binge</td>
<td>2 (4%)</td>
<td>3 (5%)</td>
</tr>
<tr>
<td>CD4, median (IQR)^4</td>
<td>206 (109, 342)</td>
<td>431 (315, 679)</td>
</tr>
<tr>
<td>ART^4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No therapy</td>
<td>19 (44%)</td>
<td>19 (42%)</td>
</tr>
<tr>
<td>Monotherapy</td>
<td>10 (23%)</td>
<td>9 (20%)</td>
</tr>
<tr>
<td>Combination</td>
<td>6 (14%)</td>
<td>5 (11%)</td>
</tr>
<tr>
<td>HAART</td>
<td>8 (19%)</td>
<td>12 (27%)</td>
</tr>
<tr>
<td>Year of liver-related death</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1986-1995</td>
<td>27 (47%)</td>
<td></td>
</tr>
<tr>
<td>1996-2000</td>
<td>13 (23%)</td>
<td></td>
</tr>
<tr>
<td>2001-2010</td>
<td>17 (30%)</td>
<td></td>
</tr>
<tr>
<td>Cause of death</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatocellular carcinoma</td>
<td>7 (12%)</td>
<td></td>
</tr>
<tr>
<td>Complication of cirrhosis</td>
<td>41 (72%)</td>
<td></td>
</tr>
<tr>
<td>Acute/subacute liver necrosis</td>
<td>7 (12%)</td>
<td></td>
</tr>
<tr>
<td>Alcoholic hepatitis</td>
<td>2 (4%)</td>
<td></td>
</tr>
</tbody>
</table>

^1Categorized based on HIV status closest to death
^2Four HBV/HCV/HIV cases, 1 HBV/HCV/HIV control
^3At visit closest to death for cases, calendar year-matched for controls
^4At visit closest to death for cases, calendar year-matched visit for controls, among HIV-infected
Table 5.2. Comparison of APRI between cases and controls at visits prior to death.

<table>
<thead>
<tr>
<th></th>
<th>9 years pre-death pairs=30</th>
<th>6 years pre-death pairs=35</th>
<th>3 years pre-death pairs=32</th>
<th>2 years pre-death pairs=34</th>
<th>1 year pre-death pairs=31</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Case</strong></td>
<td>1.57 (1.52)</td>
<td>1.78 (1.98)</td>
<td>2.24 (1.83)</td>
<td>4.11 (5.54)</td>
<td>4.77 (4.95)</td>
</tr>
<tr>
<td><strong>Control</strong></td>
<td>0.75 (0.51)</td>
<td>1.06 (1.29)</td>
<td>1.08 (1.45)</td>
<td>1.09 (2.01)</td>
<td>0.89 (0.77)</td>
</tr>
<tr>
<td><strong>p-value</strong></td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>0.02</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

*p-values for comparisons of natural log (ln) APRI
Table 5.3. Odds of death with abnormal APRI at visits prior to death.

<table>
<thead>
<tr>
<th>APRI</th>
<th>9 years pre-death pairs=30</th>
<th>6 years pre-death pairs=35</th>
<th>3 years pre-death pairs=32</th>
<th>2 years pre-death pairs=34</th>
<th>1 year pre-death pairs=31</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>OR (95% CI)</td>
<td>p</td>
<td>OR (95% CI)</td>
<td>p</td>
<td>OR (95% CI)</td>
</tr>
<tr>
<td>APRI ≤0.5</td>
<td>Reference</td>
<td></td>
<td>Reference</td>
<td></td>
<td>Reference</td>
</tr>
<tr>
<td>APRI &gt;0.5-1.5</td>
<td>2.7 (0.7, 10.2)</td>
<td>0.15</td>
<td>2.8 (0.9, 9.0)</td>
<td>0.08</td>
<td>4.5 (1.0, 20.6)</td>
</tr>
<tr>
<td>APRI &gt;1.5</td>
<td>16.2 (1.7, 159)</td>
<td>0.02</td>
<td>4.7 (1.2, 19.3)</td>
<td>0.03</td>
<td>16.1 (2.4, 109)</td>
</tr>
</tbody>
</table>
Table 5.4. Odds of death on multivariable regression

<table>
<thead>
<tr>
<th>APRI 6-years prior to death</th>
<th>OR (95% CI)</th>
<th>p-value</th>
<th>APRI 3-years prior to death</th>
<th>OR (95% CI)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>APRI ≤0.5</td>
<td>Reference</td>
<td></td>
<td>APRI ≤0.5</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>APRI 0.5-1.5</td>
<td>3.7 (0.6, 23.7)</td>
<td>0.17</td>
<td>APRI 0.5-1.5</td>
<td>21.8 (0.8, 617)</td>
<td>0.07</td>
</tr>
<tr>
<td>APRI &gt;1.5</td>
<td>14.6 (1.2, 182)</td>
<td>0.04</td>
<td>APRI &gt;1.5</td>
<td>27.5 (0.9, 814)</td>
<td>0.06</td>
</tr>
<tr>
<td>ΔAPRI 3 to 1 year prior to death</td>
<td></td>
<td></td>
<td>ΔAPRI 3 to 1 year prior to death</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ΔAPRI/year ≤1.0</td>
<td>Reference</td>
<td></td>
<td>ΔAPRI/year ≤1.0</td>
<td>Reference</td>
<td></td>
</tr>
<tr>
<td>ΔAPRI/year &gt;1.0</td>
<td>11.7 (1.2, 119)</td>
<td>0.04</td>
<td>ΔAPRI/year &gt;1.0</td>
<td>7.2 (0.7, 73.8)</td>
<td>0.10</td>
</tr>
</tbody>
</table>

1 also adjusted for CD4 count
2 case pairs=25
3 case pairs=28
Figure 5.1. Changes in mean APRI over time by case/control status.
Figure 5.2. Proportion of subjects with APRI (>1.5) across time by case/control status.
Figure 5.3. Changes in mean platelet count over time by case/control status.
Figure 5.4. Changes in mean aspartate aminotransferase (AST) over time by case/control status.
CHAPTER 6: CONCLUSION

Liver disease is a major cause of morbidity and mortality among persons living with HIV infection. Although it is well established that HIV accelerates the natural history of viral hepatitis-related liver disease, whether HAART alters liver disease progression in coinfected individuals has been unclear. Furthermore, few studies have evaluated the role of HIV infection and HAART on liver disease in the absence of viral hepatitis. The objective of this research was to examine the role of HAART on liver disease in HIV-infected individuals. The MACS was an ideal cohort in which to assess this question for several reasons. First, participants were prospectively enrolled and followed prior to and after the availability of HAART. Second, reposed serum was available for testing of non-invasive markers of liver fibrosis. Third, the design of the cohort allowed for the selection of HIV-uninfected controls.

Liver biopsy has traditionally been the gold standard to assess liver disease for clinical and research purposes. However, liver biopsy is infeasible to perform in large epidemiologic studies of liver disease progression. We demonstrated that APRI is significantly elevated in HIV-viral hepatitis-coinfected men nearly a decade prior liver-related death. Furthermore, we established that longitudinal changes in APRI are strongly associated with liver-related mortality in the MACS. Our findings support the use of APRI to assess liver disease status and APRI change to estimate liver disease progression in our cohort.
Prior studies have demonstrated that among viral hepatitis-infected individuals, HIV increases the risk of advanced fibrosis, cirrhosis, hepatic decompensation, and death. Therefore, in our assessment of liver disease in the MACS, we were not surprised to find that the HIV-viral hepatitis-coinfected men had the highest prevalence of fibrosis. We were surprised, however, to demonstrate that HAART-naïve HIV-monoinfected men had a similar risk of liver disease as viral hepatitis-monoinfected men and that HIV infection was an independent risk factor for liver disease. We also found that high HIV RNA levels and low CD4 counts were independently associated with an increased risk of liver disease, regardless of the presence of viral hepatitis coinfection, suggesting that ongoing HIV replication and advanced immunosuppression are deleterious to the liver.

Having established that untreated HIV is associated with an increased risk of liver disease, we next focused our evaluation on the influence of HAART on liver disease progression. Among men whose HAART initiation was captured during their MACS follow-up, we found that on average, APRI increased in the 3-year interval before HAART initiation and decreased in the 3-year interval including HAART initiation. The post-HAART decline in APRI was most pronounced in the men who achieved undetectable HIV RNA on HAART, demonstrating the importance of effective HAART. These findings corroborate our cross-sectional results implicating elevated HIV RNA in liver damage. However, more studies on the precise mechanism by which untreated HIV affects the liver are needed. In addition, our findings of a beneficial HAART effect were primarily seen shortly after HAART initiation. Therefore additional studies evaluating the long-term effects of HAART on the liver are required.
CHAPTER 7: REFERENCES


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ACADEMIC CURRICULUM VITAE

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POSITIONS

2012-present  Assistant Clinical Professor of Medicine, Division of Gastroenterology, University of California, San Francisco

EDUCATION

2008-present  Ph.D. Candidate, Graduate Training Program in Clinical Investigation, Johns Hopkins University Bloomberg School of Public Health; Baltimore, Maryland

2004  M.D., Johns Hopkins University School of Medicine; Baltimore, Maryland

1999  B.A. with Honors and Distinction, Stanford University; Palo Alto, California Major: Human Biology

POSTDOCTORAL TRAINING

2011-present  Fellowship in Transplant Hepatology, Johns Hopkins Hospital

2007-2011  Fellowship in Gastroenterology and Hepatology, Johns Hopkins Hospital

2004-2007  Internal Medicine Internship and Residency, Johns Hopkins Hospital

PROFESSIONAL MEMBERSHIP

Fellow American College of Physicians

American College of Gastroenterology

American Gastroenterological Association

American Association for the Study of Liver Diseases
CERTIFICATION

2012-2022  Board Certification Transplant Hepatology (ABIM 278121)
2011-2021  Board Certification Gastroenterology (ABIM 278121)
2008-2018  Board Certification Internal Medicine (ABIM 278121)

HONORS AND AWARDS

2015  Gilead Research Scholars Program in HIV
2012  ACG Clinical Research Pilot Award
2012  Travel Award AGA/AASLD Academic Skills Workshop
2011  Distinguished Achievement Award for outstanding presentation, North American Conference of Gastroenterology Fellows
2009-2011  Clinical Research Scholars KL2 Award, Johns Hopkins University School of Medicine
2008  Department of Medicine Award for Outstanding Teaching by a Fellow, Johns Hopkins University School of Medicine
2004  *Alpha Omega Alpha*, Johns Hopkins University School of Medicine
2001  Dean’s Award for Research, Johns Hopkins University School of Medicine
1999  *Phi Beta Kappa*, Stanford University
1999  Distinction (awarded to top 10% of graduating class), Stanford University
1999  Human Biology Departmental Honors, Stanford University
1999  Sandy Dornbusch Award for Excellence in Research on Families and Children, Stanford University
1998  Undergraduate Research Opportunities Major Grant, Stanford University
1997  Fairclough Book Prize, Department of Classics, Stanford University
PUBLICATIONS

Peer-reviewed Original Research Publications:


**Other Publications:**


**Book Chapters:**


**INVITED PRESENTATIONS**


November 1, 2014. UCSF Bleeding Disorder Symposium. *New Hepatitis C Virus Treatments*

October 1, 2014. Good Samaritan Hospital Grand Rounds, San Jose, CA. *What’s New in the Treatment of Hepatitis C*


September 11, 2014. UCSF Transplant 2014: Riding the Wave of Change. *Interferon free HCV Treatment in The Transplant Setting- A Game Changer*

May 8, 2014. The American Conference for the Treatment of HIV (ACTHIV). *Hepatitis C: Epidemiology, Transmission, and Screening*


March 21, 2013. The American Conference for the Treatment of HIV (ACTHIV). *Update on HIV-HCV Epidemiology and Natural History*

February 12, 2013. St. Francis Memorial Hospital Grand Rounds, San Francisco, CA. *Chronic Hepatitis B Virus*

November 6, 2012 Johns Hopkins 38th Annual Topics in Gastroenterology and Hepato-Biliary Update Conference. *Hepatitis E Virus in the United States*

November 6, 2012 Johns Hopkins 38th Annual Topics in Gastroenterology and Hepato-Biliary Update Conference. *Primary Biliary Cirrhosis*

February 20, 2012 University of Alabama Gastroenterology Grand Rounds. *HIV and Liver Transplantation*

December, 2010 Clinical Gastroenterology and Hepatology Podcast. *Liver Disease in HIV-Infected Individuals*
OTHER ORAL PRESENTATIONS

April, 2015  UCSF GEMS: Introduction to Human Biology and Medicine Course; *Hepatitis C Virus: The Hepatologist’s Perspective*

October, 2014  UCSF Gastroenterology Grand Rounds; *Liver Program*

September, 2014  UCSF Gastroenterology Fellows Course; *Hepatitis B and D*

August, 2014  AAPNA Online Lecture Series; *Hepatitis C Virus Updates*

March, 2014  CFAR Scientific Council Meeting; *HIV, HCV and Fatty Liver Disease: Host, Virus, or Genes?*

March, 2014  WIHS Science Seminar; *HIV, HCV and Fatty Liver Disease: Host, Virus, or Genes?*

March, 2014  WIHS Science Seminar; *Telomere Shortening and HIV- and HCV related Liver Disease*

March, 2014  UCSF GEMS: Introduction to Human Biology and Medicine Course; *Hepatitis C Virus: The Hepatologist’s Perspective*

January, 2014  UCSF Gastroenterology Grand Rounds; *Liver Program*

June, 2013  UCSF Gastroenterology Grand Rounds; *Liver Program*

January, 2013  UCSF Gastroenterology Fellows Course; *Hepatitis B and D*

February, 2012  Johns Hopkins Gastroenterology Grand Rounds; *HIV and Liver Transplantation*

April, 2011  Johns Hopkins Clinical Research Scholars Research Meeting; *HIV and Liver Disease*

January, 2011  Johns Hopkins Gastroenterology Grand Rounds; *Refractory Helicobacter Pylori*
September, 2010  Johns Hopkins Gastroenterology Grand Rounds; *HCV and Sickle Cell Disease*

January, 2010  Johns Hopkins Gastroenterology Grand Rounds; *AIDS Cholangiopathy*

October, 2009  Johns Hopkins Gastroenterology Grand Rounds; *Hepatitis B Reactivation*

March, 2008  Johns Hopkins Gastroenterology Grand Rounds; *CMV Colonic Pseudo-tumor*

January, 2008  Johns Hopkins Gastroenterology Grand Rounds; *Henoch-Schonlein Purpura*

October, 2007  Johns Hopkins Gastroenterology Grand Rounds; *Endometriosis Masquerading as Crohn’s Disease*

October, 2006  Johns Hopkins Osler Medicine Lunch; “*Got Lactase? The Story of Lactose Intolerance*”

October, 2005  Johns Hopkins Internal Medicine Noon Conference; *Pernicious Anemia*

**RESEARCH SUPPORT**

**ACTIVE**

American College of Gastroenterology Junior Faculty Development Grant (Price)

07/1/15-06/30/18

The objectives of this study are to determine the impact of HIV on NASH prevalence and fibrosis progression among individuals with NAFLD and to identify which HIV-infected
patients with NAFLD are more likely to have NASH or advanced fibrosis on liver biopsy.

Role: Principal Investigator

Gilead Sciences Research Scholars Program in HIV (Price)
04/1/15-03/31/17

The goals of this study are to determine the association of HIV with NASH and to evaluate novel NAFLD fibrosis and NASH risk scores in the setting of HIV infection.

Role: Principal Investigator

P30A027763 (PI-Volberding)
09/30/13-09/29/15

Center for AIDS Research CFAR Administrative Supplement Award

The purpose of this study is to utilize a novel non-invasive modality to assess hepatic fibrosis, magnetic resonance elastography (MRE), to characterize hepatic fibrosis among participants of the Women’s Interagency HIV Study (WIHS) and the Study of Visceral Adiposity, HIV, and HCV: Biologic Mediators of Hepatic Steatosis (VAHH). The goals are to explore the impact of key mediators of fatty liver disease, including hepatic steatosis and systemic immune activation, on hepatic fibrosis among HIV/HCV-coinfected individuals and to evaluate gender differences in hepatic fibrosis.

Role: Principal Investigator of Administrative Supplement

Pilot Award Program in HIV/AIDS (Price)
03/05/14-06/30/15
UCSF RAP Award

HIV, HCV and Fatty Liver Disease: Host, Virus, or Genes? The major goal of this study is to determine the association of host genetics with hepatic steatosis and fibrosis in HIV-infected adults with and without HCV coinfection.

Role: Principal Investigator

Contract (Price)
09/23/13-12/31/16
Gilead Sciences, Inc.
Role: Principal Investigator

Completed:
American College of Gastroenterology Clinical Research Pilot Award. (Price)
07/1/2012-06/30/2013
The goal of this research was to determine the prevalence of and risk factors associated with nonalcoholic fatty liver disease in the Multicenter AIDS Cohort Study using non-contrast CT imaging of the liver and spleen. Risk factors examined included metabolic, genetic, and HIV-specific factors.
Role: Principal Investigator
NIH/Johns Hopkins Institute for Clinical and Translational Research KL2 Award
The major goals of this project were to assess the influence of HAART on liver disease progression in the Multicenter AIDS Cohort Study using noninvasive markers of hepatic fibrosis and to evaluate the association between noninvasive markers of hepatic fibrosis and liver-related mortality in the Multicenter AIDS Cohort Study.
Role: Trainee investigator

Basic Science Research in Digestive Diseases
This divisional training grant provided salary and educational support in the form of full-time coursework at the Johns Hopkins Bloomberg School of Public Health Graduate Training Program in Clinical Investigation.
Role: Trainee investigator