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Chronic Hepatitis C Virus Infection Is Associated with Subclinical Coronary Atherosclerosis in the Multicenter AIDS Cohort Study (MACS): a Cross-Sectional Study

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Footnote Page

1) The authors of this manuscript have the following associations that may pose a potential conflict of interest: Mallory Witt: Hepatitis C Advisory Board, Gilead Sciences. Frank Palella: Speaker's Bureau for Gilead Sciences, Janssen Pharmaceuticals, Merck, and Bristol Myers Squibb. Todd Brown: Consultant, Gilead Sciences, Bristol Myers Squibb, Merck, Abbvie, EMD-Serono, ViiV Healthcare.

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Abstract

Background: Hepatitis C virus (HCV) infection may increase the risk of cardiovascular disease (CVD). We evaluated the association of chronic HCV (CHC) and coronary atherosclerosis among participants in the Multicenter AIDS Cohort Study.

Methods: We assessed 994 HIV-infected and –uninfected men (87 with CHC) for coronary plaque using non-contrast coronary CT and CT angiography (n=755/994) and evaluated the associations of CHC and HIV with measures of plaque prevalence, extent, and stenosis.

Results: Adjusted for demographics, HIV serostatus, behaviors, and CVD risk factors, CHC was significantly associated with a higher prevalence of coronary artery calcium (CAC) (prevalence ratio 1.29, 95% CI 1.02-1.63), any plaque (1.26, 95% CI 1.09-1.45), and noncalcified plaque (1.42, 95% CI 1.16-1.75). CHC and HIV were independently associated with the prevalence of any and noncalcified plaque, but there was no evidence of a synergistic effect due to HIV/HCV coinfection. The prevalence of CAC, any, noncalcified, mixed, and calcified plaque were significantly higher among men with HCV RNA $\geq 2x10^6$ IU/mL compared to men without CHC.

Conclusions: Chronic HCV infection is associated with subclinical cardiovascular disease, suggesting that HCV-infected individuals warrant vigilant cardiovascular risk assessments. Future research should determine whether HCV infection duration or HCV treatment impact coronary plaque development.

Introduction

Chronic hepatitis C virus (HCV) and HIV infection contribute to increased immune activation and systemic inflammation,[1, 2] which are associated with the development and progression of atherosclerosis.[3] HIV-infected individuals have an increased prevalence of subclinical atherosclerosis[4] and a higher rate of acute coronary events compared to HIV-uninfected persons.[5] Previously, we demonstrated that HIV-infected men who have sex with men (MSM) in the Multicenter AIDS Cohort Study (MACS) have more subclinical coronary atherosclerosis than HIV-uninfected MSM.[6] HCV infection has been associated with carotid atherosclerosis,[7] heart failure[8] and stroke,[9] but studies evaluating links between HCV infection and cardiac events, specifically myocardial infarction, have yielded conflicting results.[8, 10]

HIV/HCV-coinfection is common due to shared modes of transmission.[11] Among HIV-infected populations, HCV co-infection may be associated with greater carotid atherosclerosis[12] and with cardiovascular and cerebrovascular events,[13-15] though associations between HCV and carotid atherosclerosis have not been demonstrated consistently.[16, 17] It remains unclear, however, whether the effects of HIV and HCV infection on atherosclerosis are independent of each another, or whether the two viruses act synergistically in coinfected individuals.

The purpose of the present study was to determine whether HCV infection was associated with prevalence and extent of subclinical coronary atherosclerosis among HIV-infected and -uninfected MSM in the MACS, and to explore the potential synergistic effect of HIV/HCV-coinfection on atherosclerosis.

Methods

Study Population

The MACS is an ongoing observational cohort study of the natural and treated histories of HIV-1 infection among MSM.[18, 19] Briefly, 6,972 men were enrolled between 1984 and 2003 in four large metropolitan areas of the United States (Baltimore MD/Washington DC, Chicago IL, Pittsburgh PA, and Los Angeles CA) Data were collected at study entry and semi-annual visits via interviewer-administered and computer-assisted questionnaires and physical examinations. Biological specimens were obtained at each visit for laboratory testing and repository storage. The MACS protocol and data collection forms are available at http://statepi.jhsph.edu/macs/macs.html.

Participants in active follow-up during January 2010 were eligible to undergo non-contrast computed tomography (CT) scanning as part of the MACS cardiovascular ancillary study if they were 40-70 years old, did not have a history of cardiac surgery or percutaneous coronary intervention, and weighed less than 300 pounds. Coronary CT angiography (CCTA) was performed on a subset of men without contrast allergy, impaired renal function (estimated glomerular filtration rate <60 ml/min/1.73m^2 within 30 days of scanning), and atrial fibrillation at the time of CT scanning. The study protocol was approved by local Institutional Review Boards at each site. All participants provided written informed consent, and human experimentation guidelines of the United States Department of Health and Human Services and each institution were followed in the conduct of this clinical research.

Cardiac CT Imaging

Subclinical coronary atherosclerosis was assessed using non-contrast cardiac CT and CCTA.[20] Details of CT scanning procedures for this study have been described previously.[6, 21] Briefly, CT images were transferred to the core CT reading center (Los Angeles Biomedical Research Institute at Harbor-UCLA) and analyzed by trained readers blinded to participant characteristics including HIV or HCV serostatus. Coronary artery calcium (CAC) scores were computed using the Agatston method.[22] Using the modified 15-segment model of the American Heart Association,[23] CCTA images were assessed for the presence, size, and composition of coronary plaque and the degree of stenosis in all assessable coronary segments. Plaque size for each segment was graded as 0=no plaque, 1=mild, 2=moderate, or 3=severe. Segment stenosis was defined as 0=no plaque, 1=1-29% (minimal) stenosis, 2=30-49% (mild) stenosis, 3=50-70% (moderate) stenosis, or 4 for >70% (severe) stenosis. The overall extent of coronary plaque (total plaque score) was calculated by summing the plaque size score for all assessable coronary segments. The segment involvement score was calculated as the sum of coronary artery segments with plaque, regardless of the degree of stenosis.

Each coronary segment was then classified as normal or containing noncalcified, mixed (<50% of plaque area occupied by calcium) or calcified plaque. Calcified plaque was defined as any structure with a density >130 Hounsfield units (HU) that was visualized as separate from the intravascular lumen and identified in at least two independent planes. Noncalcified plaque was defined as any discernible structure that could be clearly assignable to the vessel wall with a CT density less than the contrast-enhanced coronary lumen but greater than the surrounding connective tissue, and identified in at least two independent planes. The noncalcified, mixed, and calcified plaque scores for each

participant were calculated by separately summing the scores for noncalcified, mixed, and calcified plaque across all coronary segments.[21] Finally, fatty liver disease was defined as a liver/spleen attenuation ratio of less than 1 HU on non-contrast CT scans.[24]

HCV Testing

HCV testing in the MACS has been described in detail elsewhere.[25] Briefly, the MACS implemented a prospective HCV testing protocol in 2001 through which active MACS participants were screened for HCV antibody (anti-HCV; ADVIA Centaur HCV assay, Siemens Healthcare Diagnostics, Tarrytown, NY, USA) every two years while they remained anti-HCV negative, with additional testing of HCV seroconverters to identify the first anti-HCV positive visit. In anti-HCV positive participants, we quantified HCV RNA first in a blood sample obtained during 2001-03 or at the visit following HCV seroconversion and then in another sample obtained at the last MACS visit from which specimens were available through March 2012 using a quantitative real time PCR assay (COBAS AmpliPrep COBAS TaqMan HCV assay, Roche Molecular Systems, Pleasanton, CA, USA). Participants with HCV RNA detected at both timepoints were classified as having chronic HCV infection during the entire time interval while men with undetectable RNA at both timepoints were classified as having cleared HCV infection. For men who cleared HCV RNA during this interval, we repeatedly tested for HCV RNA at interim visits until the last HCV RNA positive and first HCV RNA negative visits were identified. Using these results, we determined the chronic HCV (CHC) infection status of all participants at the time of CT exam where CHC status was confirmed with at least two positive or negative RNA results at least 6 months apart. Based on studies demonstrating that HCV RNA levels remain relatively constant over time,[26, 27] we

further classified men with CHC according to the most recent HCV RNA level that was obtained prior to the CT scan.

Covariate Measurement

Serological, clinical, and behavioral data were obtained from the MACS research visit a median of 57 (IQR: 23-112) days prior to the date of CT scanning. Education, race/ethnicity, age, medication use (hypertension, diabetes mellitus, and lipid-lowering), cigarette use, alcohol consumption, and injection drug use were self-reported. Hepatitis B infection was defined as a positive test for the hepatitis B surface antigen (HBsAg). Serum glucose and triglyceride levels were measured prospectively from fasting blood samples, while total cholesterol and high-density lipoprotein cholesterol (HDL-C) were measured regardless of fasting status. Low-density lipoprotein cholesterol (LDL-C) was either calculated using the Friedewald equation or measured directly when participants were not fasting or had triglyceride levels >400 mg/dl. An aspartate aminotransferase-to-platelet ratio index (APRI) value ≥1.5 was interpreted as significant liver fibrosis or cirrhosis.

HIV-uninfected men were tested for HIV at each MACS semi-annual visit, and HIV serostatus was established by a positive ELISA test confirmed by Western blot.[18] Measures of HIV disease activity, including $CD4^+$ T-cell counts and plasma HIV RNA levels (Roche ultrasensitive assay with limit of detection of 50 copies/mL; Roche Diagnostics, Nutley, NJ), were measured at each visit for HIV-infected men. Duration of highly active antiretroviral therapy (HAART) was calculated based on the longitudinal participant self-report of antiretroviral medications use at each MACS visit, and history

of clinically-defined AIDS was determined by medical record confirmation of self-reported outcomes.

Statistical Analysis

The distributions of demographic, behavioral, and clinical characteristics of men with and without CHC were compared using the chi-square test for categorical variables, a nonparametric test for trend across ordered groups,[28] and the Kruskal-Wallis tests for continuous variables. The observed prevalence of each plaque type and distribution of plaque scores were described by CHC infection status and compared using chi-square or Wilcoxon rank sum tests.

To adjust for potential confounding factors, we compared plaque prevalence by HIV and CHC status using a Poisson regression model with robust variance.[29] We first fit minimally adjusted models that accounted for age, race, education, study site, and enrollment period (enrolled 1984-1990 vs. 2001-2003) and then fit fully adjusted models to further account for established cardiovascular disease (CVD) risk factors: (body mass index [BMI], cumulative smoking exposure [pack-years], use of hypertension medications, use of diabetes medications, use of lipid-lowering medications, systolic blood pressure [BP], fasting glucose, and total and HDL cholesterol of men not receiving anti-hypertension, diabetes or lipid-lowering medications), history of injecting drugs, and heavy alcohol use (average consumption of more than 21 alcohol-containing drinks per week). Linear regression was used to assess the association of HIV and CHC infection with plaque extent measures among individuals with plaque present (i.e., plaque score >0) adjusted for the demographic and CVD risk factors described above. Plaque scores were natural-log transformed to account for skewed distributions. We then explored the

possibility of a synergistic relationship, defined as a positive interaction, between HIV and HCV infections on coronary plaque outcomes by testing an interaction term in the multivariate model. We also tested for an association between plaque prevalence and HCV RNA level by stratifying men according to the median HCV RNA level ($<2 \times 10^6$ IU/mL vs. $\geq 2 \times 10^6$ IU/mL vs. men without CHC) obtained from the visit closest to the CT scan and then using a Wald test to compare plaque prevalence between groups.

We used multiple imputation to account for missing data in the analysis; no covariate had more than 4% missing data. The imputation model was comprised of all variables that were included in the fully adjusted models. Missing values were imputed 10 times based on the distribution of covariates using the data augmentation Markov chain Monte Carlo method[30] assuming a multivariate normal distribution. Statistical significance was defined as a p-value <0.05. All analyses used Stata 13.1 (StataCorp, College Station, TX).

Results

Among 1,005 HIV-infected and -uninfected men who underwent non-contrast CT scans, we excluded 11 men whose last HCV test had been performed more than 2.5 years before the CT scan. Of the remaining 994 men (87 with CHC), 755 (76%; 56 with CHC) also completed CCTA. Eighty six percent of the men with CHC (n=75) were infected with HCV prior to enrollment in the MACS. The duration of CHC infection is unknown in this subgroup, but their median time between cohort enrollment and CT scanning was 8.2 years. Men who had previously cleared HCV infection comprised less than 5 percent of the study population (n=43 with CT scan and n=26 with CCTA). The most common

reason for not having undergone CCTA was impaired renal function (65%). Only 2 percent of men included in this analysis (n=24) were infected with hepatitis B.

Compared to men without CHC, men with CHC were significantly ($p<0.05$) more likely to be HIV-infected (80.5% and 59.9%, respectively), African-American, participate at the Baltimore MACS site, have lower educational attainment, be current smokers with greater cumulative smoking pack-years, currently abstinent from alcohol, and have injected drugs (Table 1). In addition, men with CHC had lower BMI, were more likely to be taking hypertension or diabetes medications, had lower total and LDL cholesterol levels, and were less likely to be taking lipid lowering medications. The median serum HCV RNA level among men with CHC was $2.0x10^6$ IU/mL (IQR $4.2x10^5$-$5.8x10^6$ IU/mL). Furthermore, among HIV-infected men, those with CHC coinfection had a shorter duration of HAART exposure and were more likely to have detectable plasma HIV RNA levels (>50 copies/mL).

Associations between HCV and Coronary Atherosclerosis

The prevalence and extent of plaque and stenosis among MACS participants stratified by CHC status are presented in Table 2. Men with CHC had a significantly higher unadjusted prevalence of any plaque on CCTA (89.3% vs 75.4%, $p=0.02$) and noncalcified plaque (76.8% vs 57.5%, $p<0.01$) compared to men without CHC. The prevalence did not differ significantly by CHC status for CAC, mixed plaque, calcified plaque, or coronary artery stenosis ≥50%, and CHC status was not significantly associated with any of the plaque scores (plaque extent) among the men with plaque.

Next, we adjusted for participant characteristics and CVD risk factors to better characterize the associations of coronary atherosclerosis with CHC and HIV serostatus (Table 3). Both CHC and HIV infection were independently and positively associated with prevalence of any plaque on CCTA (adjusted prevalence ratio [aPR] 1.26, 95% confidence interval 1.09-1.45, and aPR 1.12 [1.03-1.22], respectively) and noncalcified plaque (aPR 1.42 [1.16-1.75] and aPR 1.27 [1.11-1.45], respectively). CHC, but not HIV infection, was associated with 29% higher CAC prevalence (aPR 1.29 [1.02-1.63]). Among men with plaque, CHC was not associated with extent of any coronary plaque type and did not alter the previously reported [6] association of HIV infection with a higher noncalcified plaque score (mean difference in log score 0.15 [0.01-0.28)]). We also tested for an interaction between HIV infection and CHC in each model and found no evidence of HIV/HCV-coinfection synergy on any plaque outcome (data not shown).

Among the three coronary plaque outcomes significantly associated with CHC infection (CAC, any plaque, and noncalcified plaque), we found a significant relationship between HCV RNA level and presence of CAC (Figure 1) after adjustment for HIV serostatus, demographic, behavioral, and CVD risk factors. The prevalence of CAC>0 was significantly elevated among men with CHC who had HCV RNA $>2x10^6$ IU/mL (aPR 1.53 [1.20-1.97] vs. men without CHC) but not among men with HCV RNA $<2x10^6$ IU/mL (aPR 1.07 [0.76-1.52]), and the difference between these two aPRs was borderline significant (p=0.06). Similarly, the prevalence of any plaque and noncalcified plaque was significantly elevated in men with CHC who had HCV RNA $>2x10^6$ IU/mL compared to men without CHC, and men with CHC and lower HCV RNA levels also had an elevated prevalence of these two outcomes that was borderline significant (p=0.05 and p=0.07, respectively) compared to men without CHC. The prevalence of both mixed plaque and

calcified plaque were also significantly elevated among the men with CHC, but only among those who had HCV RNA >$2x10^6$ IU/mL.

Finally, we performed a series of sensitivity analyses to examine the robustness of the results from the multiple regression analyses. In separate analyses, we found that adjustment for current injection drug use (substituted for ever IDU), the presence of fatty liver disease, fibrosis, CD4 T cell levels, and HIV RNA suppression, as well as the exclusion of men with HBV infection, did not change the inferences regarding the associations of plaque type and extent with HIV infection and CHC (data not shown). Furthermore, stratification of men without CHC into those who had never been infected with HCV and those with cleared HCV revealed that the adjusted prevalence ratios for these two groups were statistically indistinguishable from one another for each plaque outcome (data not shown).

Discussion

This is the largest study to date to demonstrate that chronic HCV infection is associated with subclinical coronary atherosclerosis, an important predictor of future cardiovascular events,[31] in a large, well-characterized group of HIV-infected and HIV- uninfected MSM. In the subgroup of men who also underwent CCTA, CHC was associated with a higher prevalence of any plaque and of noncalcified plaque. While men with CHC in our cohort were more likely to be African-American, less educated, current smokers, and have a history of injection drug use, the association of CHC with plaque remained significant after adjustment for these and other recognized CVD risk factors, and was independent of HIV infection. Our findings are consistent with prior reports linking HCV infection with cardiovascular and cerebrovascular events among HIV-uninfected

persons,[10, 32] and support the hypothesis that chronic HCV infection is independently associated with the development of cardiovascular disease.

Previous studies demonstrated that HIV/HCV-coinfected individuals had a greater prevalence of cardiovascular and cerebrovascular disease events[13, 14] compared to HIV-monoinfected individuals, but conflicting reports existed regarding the association between HIV/HCV coinfection and subclinical carotid atherosclerosis.[12, 16] Our study is the first to show that HIV and CHC are independently associated with increased subclinical coronary atherosclerosis in a large cohort. Although we found no evidence of a synergistic effect for HIV/HCV coinfection, our study had insufficient numbers to formally examine the interaction between infection with these two viruses. Nevertheless, our study adds to the literature by demonstrating that a greater burden of subclinical coronary atherosclerosis exists among men with CHC whether or not they are coinfected with HIV.

The mechanism by which HCV infection may increase CVD remains unclear, but systemic inflammation and immune activation resulting from CHC may provide a mechanistic link. HCV activates the inflammasome leading to IL-18 production,[33] and elevated levels of IL-18 have been demonstrated in patients with acute coronary syndrome[34] and are associated with unstable coronary atherosclerotic plaque.[35] Chattergoon et al. reported that HIV infection leads to IL-18 production through inflammasome activation and postulated that this may be an explanation for the increased risk of CVD associated with HIV infection.[33] In addition, we found that the prevalence of CAC was significantly elevated among men with high HCV RNA levels, which is consistent with a prior study demonstrating an association between higher HCV RNA

levels and greater risk of cerebrovascular death compared to HCV-uninfected controls.[36] We also observed that the prevalence of any, noncalcified, calcified, and mixed plaque outcomes measured by CCTA were each significantly elevated among men with CHC and HCV RNA >$2x10^6$ IU/mL. Collectively, these findings suggest that HCV infection is associated with plaque initiation and that a high HCV RNA level promotes calcification. Further longitudinal studies are needed to improve our understanding of the mechanisms that link chronic HCV infection and CVD.

An important strength of our analysis is that our study population was comprised of a well-characterized cohort of MSM enrolled in a CVD study. All participants underwent a comprehensive CVD evaluation, including CCTA imaging in most, which allowed for detailed characterization of coronary atherosclerosis. This study also benefited from comprehensive HCV testing of all participants. Some limitations of this study include the cross-sectional study design, which limits our ability to establish causal relationships, and the population consisting only of MSM. While the high level of cohort characterization allowed adjustment for possible confounding factors, the possibility of unmeasured confounding remains. Notably, men with renal dysfunction were excluded from participation to minimize the risks associated with intravenous contrast injection, however kidney disease is a known risk factor for CVD among HCV and HIV-infected individuals[37-39] and chronic kidney disease is more prevalent among HCV-infected persons.[37] Our findings, therefore, may represent a more conservative estimate of the excess CVD risk associated with CHC than has been demonstrated in the general HCV-infected population. Another important limitation to our study was the relatively small number of men with CHC, which reduced the power of this study to examine fully the plaque extent outcomes. Lastly, we could not assess CVD clinical events, such as angina

or myocardial infarction, although the presence of coronary atherosclerotic plaque is known to increase the likelihood of these events.[31]

In conclusion, the elevated prevalence of subclinical coronary atherosclerosis among men with chronic HCV infection, especially men with the highest HCV RNA levels, provides further evidence supporting a link between chronic HCV infection and cardiovascular disease. Future research is needed to confirm our novel finding of an association between higher HCV RNA level and atherosclerosis, and to determine whether duration of HCV infection or the use of curative HCV treatment impacts the formation or progression of coronary plaque. Nevertheless, the presence of chronic HCV infection may warrant vigilant cardiovascular risk assessment in these patients.

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Conflict of interest: MW has served on the Hepatitis C Advisory Board for Gilead Sciences. FP is on the Speaker's Bureau for Gilead Sciences, Janssen Pharmaceuticals, Merck, and Bristol Myers Squibb. TB is a consultant for Gilead Sciences, Bristol Myers Squibb, Merck, Abbvie, EMD-Serono, ViiV Healthcare. All other authors report no potential financial or personal conflicts of interest.

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Table 1. Demographic, behavioral, and clinical characteristics of MACS participants assessed with CT scan, by CHC infection status

	Total	**Men Without CHC**	**Men With CHC**	**p-value**
	n=994	n=907	n=87	
Age (years)	53 (48-59)	53 (48-59)	53 (50-58)	0.96
Race (%)				<0.001
Caucasian	58.1	62.4	13.8	
African-American	30.5	26.2	74.7	
Hispanic + Other	11.4	11.4	11.5	
Education (%)				<0.001
High school or less	21.0	18.4	48.3	
At least 1 year college	29.3	28.7	35.6	
Undergraduate degree	22.1	23.7	5.7	
Graduate degree	27.6	29.2	10.3	
Study site (%)				<0.001
Baltimore	27.5	25.6	47.1	

Chicago	25.9	26.4	20.7	
Pittsburgh	20.6	21.2	14.9	
Los Angeles	26.1	26.9	17.2	
Enrollment Period (%)				<0.001
1984-1995	54.8	58.1	20.7	
2001-2003	45.2	41.9	79.3	
Tobacco use (%)				<0.001
Never smoker	25.3	26.5	12.6	
Former smoker	47.4	49.1	29.9	
Current smoker	27.4	24.5	57.5	
Smoking pack-years [a]	11.9 (1.9-29.7)	10.4 (0.9-28.7)	23.9 (10.2-38.9)	<0.001
Alcohol use (%)				<0.001
0 drinks/week	21.0	18.9	43.0	
1-3 drinks/week	51.1	51.7	44.2	
4-21 drinks/week	20.5	22.0	4.7	

> 21 drinks/week	7.4	7.3	8.1	
Fatty liver disease (%)	16.2	17.1	6.8	0.02
Significant liver fibrosis [b] (%)	1.9	0.7	15.3	<0.001
Hepatitis B infection (%)	2.4	2.5	1.1	<0.01
Ever injected drugs (%)	14.2	9.9	58.6	<0.001
Body Mass Index (kg/m^2)	25.9 (23.5-28.9)	26.0 (23.6-29.3)	25.0 (22.0-27.6)	<0.01
Systolic blood pressure (mm Hg)	127 (116-137)	127 (116-137)	129 (113-140)	0.69
Hypertension medications (%)	34.3	33.4	43.7	0.05
Glucose (mg/dL)	98 (90-107)	97 (90-107)	100 (90-114)	0.13
Diabetes medications (%)	8.1	7.5	14.9	0.02
Total cholesterol (mg/dL)	187 (161-213)	190 (163-215)	166 (146-193)	<0.001
HDL cholesterol (mg/dL)	48 (40-58)	48 (40-58)	46 (37-55)	0.13
LDL cholesterol (mg/dL)	108 (85-133)	109 (87-134)	86 (65-113)	<0.001
Triglycerides (mg/dL)	120 (85-183)	120 (85-181)	127 (86-205)	0.36
Lipid-lowering medications (%)	33.2	35.4	10.3	<0.001

HCV RNA viral load (IU/mL)			2,030,000 (421,000-5,780,000)	
HIV infected n (%)	613 (61.7)	543 (59.9)	70 (80.5)	<0.001
Current HIV RNA undetectable [c] (%)	82.1	83.7	69.6	<0.01
Current $CD4^+$ T-cell count (cells/mm^3)	599 (422-766)	609 (431-775)	498 (383-740)	0.05
$CD4^+$ T-cell count nadir (cells/mm^3)	283 (171-397)	283 (171-399)	272 (174-378)	0.62
Currently on HAART (%)	88.3	89.2	80.9	<0.01
HAART duration (years)	9.5 (6.4-12.5)	9.7 (6.6-12.6)	8.0 (5.0-11.3)	0.01
History of AIDS (%)	14.8	14.2	20.0	0.20

Data presented represent median (interquartile range) or percent. Laboratory Glucose and Triglycerides results represent fasting levels. P-values compare men with CHC and men without CHC, by the chi-square test or nonparametric test for trend for categorical variables or Kruskal-Wallis test for continuous variables. [a] Among current and former smokers. [b] Liver fibrosis defined as aspartate aminotransferase-to-platelet ratio index (APRI) >=1.5. [c] HIV RNA undetectable defined as <50 copies/mL.

Table 2. Prevalence and extent of observed coronary artery plaque among MACS participants, by CHC infection status

	Total			**Men Without CHC**			**Men With CHC**			
	n	**(%)**	**median (IQR)**	**n**	**%**	**median (IQR)**	**n**	**%**	**median (IQR)**	**p-value**
Non-contrast CT	**n=994**			**n=907**			**n=87**			
CAC: Agatston Score >0	525	(52.8)		473	(52.2)		52	(59.8)		0.17
CAC Score			73 (22, 209)			70 (20, 211)			103 (30, 146)	0.66
Coronary CT Angiography	**n=755**			**n=699**			**n=56**			
Any Coronary Plaque	**577**	**(76.4)**		**527**	**(75.4)**		**50**	**(89.3)**		**0.02**
Total Plaque Score			4 (2, 7)			4 (2, 7)			3 (2, 5)	0.38
Segment Involvement Score			3 (2, 5)			3 (2, 5)			3 (2, 4)	0.22
Noncalcified Plaque	**445**	**(58.9)**		**402**	**(57.5)**		**43**	**(76.8)**		**<0.01**
Noncalcified Plaque Score			2 (1, 3)			2 (1, 3)			2 (1, 3)	0.38
Mixed Plaque	251	(33.2)		232	(33.2)		19	(33.9)		0.91

Mixed Plaque Score			2	(1, 3)			2	(1, 4)		2	(1, 2)	0.55
Calcified Plaque	279	(37.0)			259	37.1			20	(35.7)		0.84
Calcified Plaque Score			2	(1, 4)			2	(1, 4)		2	(1, 3)	0.88
Coronary Stenosis ≥50%	120	(15.9)			114	(16.3)			6	(10.7)		0.27

Data presented represent median (interquartile range) or percent. Scores are presented among participants with prevalent plaque. The statistical significance of differences by CHC status are assessed by chi-squared or Wilcoxon rank sum tests as appropriate. Bolded results are statistically significant at $p<0.05$.

Table 3. Adjusted prevalence ratio of coronary artery plaque and mean difference of plaque extent, by CHC and HIV serostatus

Plaque Outcome	Predictor	Adjusted Prevalence Ratio	(95% CI)
Coronary artery calcium	CHC	1.29[a]	(1.02,1.63)
	HIV	1.10	(0.97,1.24)
Any plaque	CHC	1.26[b]	(1.09,1.45)
	HIV	1.12[b]	(1.03,1.22)
Noncalcified plaque	CHC	1.42[c]	(1.16,1.75)
	HIV	1.27[c]	(1.11,1.45)
Mixed plaque	CHC	1.07	(0.67,1.71)
	HIV	1.19	(0.95,1.49)
Calcified plaque	CHC	1.41	(0.93,2.14)
	HIV	0.99	(0.81,1.20)
Stenosis ≥50%	CHC	0.58	(0.24,1.38)
	HIV	1.14	(0.79,1.65)
		Mean difference	(95% CI)
Agatston Score (n=525)	CHC	0.34	(-0.25,0.93)
	HIV	-0.03	(-0.35,0.30)
Segment Involvement Score (n=577)	CHC	0.04	(-0.19,0.27)
	HIV	0.10	(-0·02,0.22)
Total Coronary Plaque Score (n=577)	CHC	0.03	(-0.24,0.30)

	HIV	0.12	(-0.03,0.26)
Noncalcified Plaque Score (n=445)	CHC	-0.04	(-0.30,0.21)
	HIV	0.15[a]	(0.01,0.28)
Mixed Plaque Score (n=251)	CHC	-0.33	(-0.77,0.10)
	HIV	0.13	(-0.08,0.34)
Calcified Plaque Score (n=279)	CHC	0.21	(-0.19,0.61)
	HIV	-0.08	(-0.28,0.12)

[a] $p < 0.05$, [b] $p < 0.01$, [c] $p < 0.001$

The CHC and HIV adjusted PRs are from the same models and, thus, represent independent effects. The results for each plaque outcome are adjusted for age, race, education, study site, enrollment period, BMI, cumulative smoking pack-year, use of hypertension medications, use of diabetes medications, use of lipid-lowering medications, systolic blood pressure, fasting glucose, total and HDL cholesterol, history of injecting drugs, and heavy alcohol use (average consumption of more than 21 alcohol-containing drinks per week).

Figure Legend

Figure 1. Association of chronic HCV infection (CHC) with coronary artery calcium, any plaque, noncalcified plaque, mixed plaque, and calcified plaque by HCV RNA level. Regression analyses adjusted for age, race, education, center, enrollment period, injection drug use history, alcohol, HIV serostatus, and cardiovascular disease risk factors. Abbreviations: HCV, hepatitis C virus; CHC, chronic HCV infection; N, number; CT, computed tomography; aPR, adjusted prevalence ratio; ref, reference group. *p<0.05, **p<0.01, ***p<0.001.

CHC Status	HCV RNA Level (IU/mL)	N	Plaque Prevalence	Adjusted Prevalence Ratio (aPR)
Non−contrast CT Scans (N=994)				
Coronary Artery Calcium				
CHC−		907	52%	(ref)
CHC+	< $2x10^6$	43	51%	1.07
CHC+	≥ $2x10^6$	44	68%	1.53 ***
Coronary CT Angiography Scans (N=755)				
Any Plaque				
CHC−		699	75%	(ref)
CHC+	< $2x10^6$	26	85%	1.22
CHC+	≥ $2x10^6$	30	93%	1.29 **
Noncalcified Plaque				
CHC−		699	58%	(ref)
CHC+	< $2x10^6$	26	69%	1.33
CHC+	≥ $2x10^6$	30	83%	1.50 ***
Mixed Plaque				
CHC−		699	33%	(ref)
CHC+	< $2x10^6$	26	15%	0.45
CHC+	≥ $2x10^6$	30	50%	1.72 *
Calcified Plaque				
CHC−		699	37%	(ref)
CHC+	< $2x10^6$	26	27%	1.06
CHC+	≥ $2x10^6$	30	43%	1.72 *