Published in final edited form as:
Clin Infect Dis. 2009 October 1; 49(7): 1119–1127. doi:10.1086/605578.

Relationship between Inflammatory Markers, Endothelial Activation Markers, and Carotid Intima-Media Thickness in HIV-Infected Patients Receiving Antiretroviral Therapy

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Abstract

Background—Human immunodeficiency virus (HIV)–infected patients are at increased risk of cardiovascular disease, which may be related to chronic inflammation and endothelial dysfunction despite virological control with antiretroviral therapy. The relationship between carotid intima-media thickness (IMT), a surrogate marker for cardiovascular disease, proinflammatory cytokines, and endothelial activation markers has not been fully explored in HIV-infected patients who are receiving antiretroviral therapy.

Methods—We conducted a prospective, cross-sectional, observational study of treated HIV-infected patients and healthy control subjects to evaluate the relationship between carotid IMT, proinflammatory cytokines, endothelial activation biomarkers, and metabolic parameters in treated HIV-infected patients, compared with healthy control subjects.

Results—We enrolled 73 HIV-infected patients and 21 control subjects. Common carotid artery and internal carotid artery IMT measurements, as well as tumor necrosis factor–α, high-sensitivity C-reactive protein, inter-leukin-6, myeloperoxidase, and soluble vascular cell adhesion molecule-1 levels were higher in the HIV-infected group. High-sensitivity C-reactive protein was the only biomarker that was positively correlated with carotid IMT in both groups. In the HIV-infected group, soluble vascular cell adhesion molecule–1 was positively correlated with all inflammatory cytokine levels. In multiple regression analysis, soluble vascular cell adhesion molecule–1, myeloperoxidase, and tumor necrosis factor–α levels were all associated with internal carotid artery IMT in the HIV-infected group, whereas age was associated with both common carotid artery and internal carotid artery IMT.

Conclusions—Enhanced endothelial activation, inflammation, and increased carotid IMT occur in HIV-infected patients despite antiretroviral therapy. Inflammatory markers are associated with endothelial activation, and both are associated with internal carotid artery IMT, supporting a

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Potential Conflicts of Interest. All other authors: no conflicts.

potential role of inflammation in endothelial activation and cardiovascular disease in HIV infection.

HIV-infected individuals appear to be at higher risk of cardiovascular disease (CVD) than the general population [1, 2]. The etiology of the increased risk remains unclear. Some data suggest that long-term protease inhibitors use contributes to risk [3–5], whereas others cite more traditional risk factors, such as smoking, dyslipidemia, and advanced age [6–8]. Measurement of carotid intima-media thickness (IMT) is a well-accepted, noninvasive method of monitoring subclinical atherosclerotic formation and progression, and increased IMT correlates with an increased risk of CVD events, such as myocardial infarction and stroke, in the general population [9–12]. Although no studies to date show prediction of CVD events on the basis of IMT values in the HIV-infected population, several studies demonstrate increased IMT in HIV-infected individuals, compared with healthy control persons [8, 13–15], and IMT progresses more rapidly over time in HIV-infected patients [8, 14, 16]. Likewise, data show that HIV infection status, when controlled for other factors, is associated with increased carotid IMT [15, 17].

There is mounting evidence to support the role of inflammation and endothelial activation and dysfunction in the development of plaque formation and progression of atherosclerosis in the general population [18–27]. For example, elevated levels of the proinflammatory cytokines interleukin (IL)-6 and C-reactive protein (CRP) are associated with subclinical atherosclerosis [18] and are independently predictive of future cardiovascular events [19, 20]. In addition, tumor necrosis factor (TNF)–α has been implicated in myocardial dysfunction after acute coronary syndromes, and levels of TNF-α have been shown to be higher in patients at baseline who experience recurrent myocardial infarctions or cardiac death [21, 22]. The effects of TNF-α are mediated by 2 receptors, TNFR-I and TNFR-II; these soluble receptors are stable in plasma, easily measured, and reflect activity of TNF-α. Myeloperoxidase, a peroxidase enzyme found in leukocytes and released when leukocytes are activated, has also been established as an independent predictor of early risk of myocardial infarction in patients presenting with chest pain [23], and myeloperoxidase levels are associated with the presence of coronary artery disease and endothelial dysfunction in the general population [24]. Likewise, levels of 2 circulating adhesion molecules, soluble vascular cell adhesion molecule–1 (sVCAM-1) and soluble intercellular adhesion molecule–1 (sICAM-1), as well as levels of von Willebrand factor antigen (vWF), arise from shedding or proteolytic cleavage from activated endothelial cells and are useful markers for increased activation of endothelial cells in atherosclerosis [25–27].

Few studies have examined the relationship between markers of inflammation and endothelial activation and measurements of carotid IMT. In the general population, the Offspring Cohort of the Framingham Heart Study found an association between inflammatory markers and IMT [28], whereas studies examining the role of chronic inflammation as a cause of the increased CVD risk in patients with type-2 diabetes have produced conflicting results [29–31]. Nevertheless, increased levels of inflammatory markers and evidence of endothelial dysfunction are evident in HIV-infected individuals [32–35] and may be correlated with increased risk of CVD. Even in patients who are receiving antiretroviral therapy (ART), inflammatory cytokine levels do not completely normalize to that of the general population [33], suggesting that there is ongoing inflammation despite an undetectable virus level.

To date, no study has comprehensively examined the relationship among inflammatory markers, carotid IMT, and endothelial activation markers in HIV-infected individuals. We hypothesized that inflammatory markers are correlated with both carotid IMT and endothelial activation markers in HIV-infected subjects receiving ART and that both

inflammatory and endothelial activation markers are associated with carotid IMT. In this study, we measured plasma levels of proinflammatory cytokines, TNF-α, sTNFR-I and -II, IL-6, high-sensitivity CRP (hsCRP), and myeloperoxidase, and of 3 endothelial activation markers, sICAM-1, sVCAM-1, and vWF, to explore their relationship to carotid IMT in HIV-infected individuals receiving ART with virologic control. As a secondary objective, we examined carotid IMT, inflammatory markers, and endothelial activation markers in ART-treated, HIV-infected subjects, compared with a small convenience sample of healthy control subjects.

METHODS

Study design and patient population

This is a cross-sectional, multicentered, observational study. Subjects were enrolled prospectively at the John T. Carey Special Immunology Unit of University Hospitals Case Medical Center/Case Western Reserve University and at the Cleveland Clinic Foundation in Cleveland, Ohio. Inclusion criteria included documented HIV infection, age ≥21 years, and stable receipt of ART for ≥24 weeks. Exclusion criteria included known CVD, diabetes, current opportunistic infection, and acute or chronic inflamma-tory condition. Healthy control subjects were selected from a convenience sample that included hospital staff members and relatives of HIV-infected patients and staff and were eligible if they had no recent or current infection and no uncontrolled chronic conditions. Control subjects were matched so that the median age was within 5 years of that for HIV-infected subjects. Exclusion criteria for control subjects were the same as that for the HIV-infected group.

Study evaluations included physical examination; measurement of blood pressure, height, weight, and waist-to-hip ratio (with standardized measurements based on procedure recommendations from the Metabolic Study Group of the AIDS Clinical Trials Group); blood sampling after ≥8 h of fasting; and carotid ultrasound. Homeostasis model assessment of insulin resistance (HOMA-IR) was calculated by dividing the product of the fasting glucose level (mg/dL) and the fasting insulin level (mU/mL) by 22.5 [36]. Extensive chart review was conducted for HIV-infected subjects, including HIV duration, detailed ART history, past and current medical diagnoses, current medications, and nadir CD4 cell count. Demographic characteristics were collected from both groups, including smoking and relevant family history.

Metabolic, inflammatory, and cardiovascular tests

Fasting blood samples were drawn from all subjects for real-time measurements of insulin, glucose, and lipoprotein profile. We also measured plasma levels of TNF-α, sTNFR-I, sTNFR-II, IL-6, hsCRP, myeloperoxidase, and endothelial markers vWF, sICAM-1, and sVCAM-1 with use of an enzyme-labeled immunosorbent sandwich assay (Searchlight; Thermo Fisher Scientific). The median intra-assay coefficients of variation for TNF-a, sTNFR-I, sTNFR-II, IL-6, hsCRP, vWF, sICAM-1, sVCAM-1, and myeloperoxidase measurements were 14.5%, 8.8%, 8.6%, 11.7%, 6.9%, 13.3%, 8.0%, 8.7%, and 7.5%, respectively. The median interassay coefficients of variation for each assay were 9.8%, 10.8%, 6.0%, 10.4%, 4.5%, 9.0%, 4.2%, 13.4%, and 12.6%, respectively. Finally, CD4 cell counts and HIV RNA levels were measured as markers of HIV disease.

Carotid IMT measurements

All carotid ultrasounds were performed by the same experienced sonographer (J.A.), and read by an experienced radiologist (V.D.). Both investigators were blinded to patient characteristics and results. Carotid IMT methods were used as described elsewhere [13]. Briefly, images of the bilateral distal common carotid arteries (CCA), and internal carotid

arteries (ICA) were obtained in longitudinal views separately. Images of the near (proximal) and far wall free of plaques (distal) were acquired with a 7–14 MHz AT 1204 linear array transducer (Toshiba American Medical Systems) operating at 14 MHz with differential harmonics. Three measurements of the IMT were obtained at near and far wall of each CCA and ICA. The mean of 3 measurements at each site (right and left side) was used as final measurement of IMT for that site (for both CCA and ICA, the far and near wall had 3 IMT measurements each, resulting in a total of 12 measurements). Right and left sides were then averaged and reported as a single ICA and CCA measurement. Plaque was defined, measured, and graded as a plaque index according to published protocols from the Cardiovascular Health Study [37].

Statistical methods

Continuous measurements are described as medians and ranges, and nominal variables are described with percentages. Continuous measurements were compared using Wilcoxon rank-sum tests and nominal variables were compared using Fisher's exact test, as appropriate. The level of correlation was estimated using Spearman correlation coefficients.

Two-stage regression analysis was performed within the HIV-infected group for each IMT measure (ICA and CCA) to determine factors associated with IMT. In the first stage, 4 separate models were created for both ICA and CCA, examining different categories expected to affect IMT: (1) clinical characteristics, (2) laboratory values, (3) HIV-related factors, and (4) biomarker values. Variables for inclusion in models were chosen either on the basis of bivariate results or because of clinical significance. All 9 biomarkers were included in the first stage. Departures from normality in these variables were addressed by comparing differences between results when modeling on the raw scale and on $\log_{10}$ scale. It should be noted that the $\log_{10}$ transformations did not achieve normality in most cases, and therefore, models were constructed using the raw scale. Because there is a moderate to high degree of collinearity in the biomarkers, collinearity diagnostics were performed. Any variable from each of the first stage models that was significant at the $P \leq .1$ level, or lacking that, the most significant variable in the model, was included in the second stage analysis. The final model included all variables that remained with $P < .1$. Results are presented as regression coefficients, standard errors of the coefficients, P values, and adjusted R^2 values.

All analyses were performed using SAS, version 9.1 (SAS Institute). The level of significance for all analyses was set at .05.

RESULTS

Study population and between-group comparison of clinical characteristics

We enrolled 73 HIV-infected patients and 21 healthy control subjects for this observational cross-sectional study. Demographic, clinical, and HIV-related characteristics and fasting metabolic parameters are shown in Table 1. Groups were similar with regard to age, race, and body mass index (BMI [calculated as weight in kilograms divided by the square of height in meters]); however, male sex, smoking rates, blood pressure, and waist-to-hip ratio were higher in the HIV-infected group. The HIV-infected group also had significantly higher HOMA-IR scores and lower median high-density lipoprotein (HDL) cholesterol levels, but total cholesterol, triglyceride, and non-HDL median levels were similar between groups. There were, however, more HIV-infected patients receiving taking lipid- and triglyceride-lowering medications at the time of study enrollment. Low-dose aspirin (≤80 mg daily) and antihypertensive use were not statistically significantly different between

groups. Family history of premature CVD was higher among control subjects, but family history of type-2 diabetes was similar.

HIV infection characteristics are described in Table 1. The majority of HIV-infected patients (81%) had HIV RNA levels <50 copies/mL, and the remaining patients had a median HIV RNA level of 292 copies/mL.

Between-group comparison of IMT and plasma biomarkers

The carotid IMT and plasma biomarker results are shown in Table 2. Both CCA and ICA IMT measurements were significantly higher in the HIV-infected group. Also, levels of the inflammatory markers TNF-α, hsCRP, IL-6, and myeloperoxidase were significantly higher in the HIV-infected group; whereas sTNFR-I and sTNFR-II levels were similar between groups. Of the endothelial markers, only sVCAM-1 was significantly higher in the HIV-infected group.

Given the significantly higher number of smokers in the HIV-infected group, biomarker values for smokers and nonsmokers from both groups were compared and found to be similar (data not shown). Likewise, because some subjects were taking medications that can potentially affect inflammatory markers (ie, aspirin, antihypertensives, and hypolipemics), subjects taking any of these medications were compared with the rest of the subjects. No differences in biomarkers were found between those receiving these medications versus those who were not (data not shown).

Correlations among IMT, plasma biomarkers, and metabolic parameters

Significant correlations between IMT measurements, plasma biomarkers and fasting metabolic parameters are shown in Table 3. Age was the only variable that was positively correlated with both IMT measurements for both groups. In the HIV-infected group, non-HDL cholesterol level was positively correlated with both CCA and ICA IMT, but this was not the case for the control group. Triglyceride levels and HOMA-IR score were both positively correlated with one of the IMT measurements in each group. Of the inflammatory markers, only hsCRP was positively correlated with IMT in both groups (ICA for HIV-infected group and CCA for control group). Endothelial markers were not correlated with IMT in the HIV-infected group, and only sVCAM-1 was negatively correlated with ICA in the control group.

Regression Analysis

Only the HIV-infected group was included in regression analysis (Table 4). For the first stage, age, BMI, sex, lipid-lowering medication use, waist-to-hip ratio, and systolic blood pressure were chosen for the first stage model of clinical characteristics on the basis of the inclusion criteria described in the Methods. Likewise, non-HDL, triglyceride, and total cholesterol levels and HOMA-IR score were included in the laboratory values model, and current and nadir absolute CD4 cell counts, HIV duration, and the 3 main ARV classes (nonnucleoside reverse-transcriptase inhibitors [NNRTI], nucleoside reverse-transcriptase inhibitors [NRTI], protease inhibitors) were included in HIV characteristics. Finally, all bio-markers were included in the first stage.

Next, variables within each model that had a probability value of <.1 were included in the second stage of regression analysis. For ICA IMT, variables included age; waist-to-hip ratio; lipid-lowering medication use; duration of NNRTI, NRTI, or protease inhibitor use; and sVCAM-1, myeloperoxidase, vWF, and TNF-α levels. For CCA IMT, variables included age, waist-to-hip ratio, lipid-lowering medication use, and sVCAM-1, myeloperoxidase, vWF, and TNF-α levels. The final model including variables that remained with $P < .1$,

included only age ($P < .001$) and sVCAM-1 (P p .02), myeloperoxidase ($P = .03$), and TNF-α ($P = .03$) levels for ICA. For CCA, the final model included only age ($P < .001$) and waist-to-hip ratio ($P = .02$). It should be noted that none of the models explain a high degree of variability in the outcome measures, the highest R^2 being 0.40 for the final model for CCA. At the first level, again on the basis of comparative R^2 values, the models that include demographic characteristics appear to explain more variability than the other models at this level.

DISCUSSION

With the advent of highly active ART, patients infected with HIV are living decades longer than before. However, increased cardiovascular risk associated with HIV infection presents new challenges and implications for quality of life and life expectancy.

In this study, we examined the relationship between carotid IMT and biomarkers known in the general population to be abnormal in patients with CVD or predictive of CVD-associated events. As presented elsewhere [8, 13, 14], IMT measurements were higher in the HIV-infected group than in the healthy age-matched control group. Likewise, a number of inflammatory and endothelial activation markers were elevated in the HIV-infected group, compared with the control group. These findings are consistent with other studies [14, 35, 38] and underscore the heightened inflammatory state, enhanced endothelial activation, and increased CVD risk among HIV-infected individuals, even those receiving ART.

Not surprisingly, traditional CVD risk factors were positively correlated with IMT in both groups. More interesting is the fact that hsCRP was the only biomarker that was positively correlated with IMT for both groups. hsCRP has been shown consistently to be a predictor of CVD in the general population [19], and a recent study reported a higher hsCRP in HIV-infected subjects who later experience acute myocardial infarction [38]. We also found a strong correlation between sVCAM-1 level and all measured inflammatory markers, corroborating the role of inflammation in the development of endothelial activation and CVD.

In the multiple regression analysis of the HIV-infected subjects, age remained the most significant factor associated with carotid IMT. Importantly, sVCAM-1, myeloperoxidase, and TNF-α levels were also independently associated with IMT, but only ICA IMT. It is not clear why biomarkers would only be associated with ICA and not CCA IMT, because one would expect the development of atherosclerosis in this setting to occur in a diffuse and systemic process. However, similar findings were noted in the Offspring Cohort of the Framingham Study [28], where as a group, inflammatory markers were significantly associated with ICA but not CCA IMT. There are data to suggest that both ICA and CCA are predictive of CVD events in the general population [39]. However, studies that included both measurements in HIV-infected subjects showed increased IMT over control subjects [14, 15], compared with negative studies that only looked at CCA [7, 40], after adjusting for traditional risk factors.

There are limitations to this study. Foremost, some of the IMT increase found in the HIV-infected group may be accounted for by baseline differences between the groups, such as increased smoking rates, blood pressure, waist-to-hip ratio, and HOMA-IR scores and lower HDL, all of which are known to be associated with higher carotid IMT and CVD risk [41–44]. Ideally, this study would have better controlled for these confounders; however, the baseline differences are representative of the HIV-infected population as a whole and have presented challenges in other studies as well [45]. Likewise, the main objective of this study was to evaluate variables, in particular inflammation and endothelial activation, that may

account for increased IMT in the HIV-infected population that we are already assuming is at higher risk of CVD. Ultimately, the inclusion of control subjects was a secondary objective to show that even in a small convenience sample of healthy individuals, IMT measures are worse in HIV-infected patients who therefore are likely at a higher risk of CVD. Moreover, it is unclear how much these differences affected the results, because smoking status did not affect biomarker levels, there was no association between smoking and IMT in bivariate analysis, and lipid profiles (except HDL) were similar between groups.

Another limitation is that, because of the relatively small sample size, it was impossible to comment on the contribution of individual antiretrovirals to carotid IMT values. However, it is reassuring that there were no correlations between antiretroviral classes received and carotid IMT or biomarkers, and antiretroviral classes were not associated with IMT in multiple regression analysis. A significantly larger subject number would be necessary to overcome this limitation. Likewise, because of small sample size, we were unable to show group differences in all of the biomarkers, where there might have been a difference with greater power. It should also be noted that the exaggerated inflammatory response observed in HIV-infected patients is likely multifactorial, and this cross-sectional study cannot prove causality, despite the interesting correlations found. However, this study offers some basic observations that can form the substrate for future research.

To our knowledge, this is the first study specifically designed to investigate the relationship between carotid IMT and inflammation and endothelial biomarkers in HIV-infected adults receiving ART. Our results show a heightened inflammatory state with enhanced endothelial activation in HIV-infected subjects, despite virologic suppression. In addition, significant correlations were found between levels of the inflammation markers, the endothelial activation marker sVCAM-1, and carotid IMT, offering a potential mechanism for the increased CVD risk in this population. Larger, better controlled, longitudinal studies are needed to confirm and further define this relationship. Should these studies show a more definitive role of inflammation and endothelial activation in the increased risk of CVD among HIV-infected individuals, biomarker monitoring, initiation of anti-inflammatory medications, or earlier initiation of highly active ART may be beneficial.

Acknowledgments

Financial support. Partial support provided by Bristol-Myers Squibb.

G.A.M. serves as a consultant and has received research funding from Bristol-Myers Squibb, GlaxoSmithKline, Gilead, and Abbott. A.C.R. has received research funding from Bristol-Myers Squibb.

References

1. Triant VA, Lee H, Hadigan C, Grinspoon SK. Increased acute myocardial infarction rates and cardiovascular risk factors among patients with human immunodeficiency virus disease. J Clin Endocrinol Metab. 2007; 92:2506–12. [PubMed: 17456578]
2. Obel N, Thomsen HF, Kronborg G, et al. Ischemic heart disease in HIV-infected and HIV-uninfected individuals: a population-based cohort study. Clin Infect Dis. 2007; 44(12):1625–31. [PubMed: 17516408]
3. Holmberg SD, Moorman AC, Williamson JM, et al. Protease inhibitors and cardiovascular outcomes in patients with HIV-1. Lancet. 2002; 360:1747–8. [PubMed: 12480430]
4. Friis-Moller N, Sabin CA, Weber R, et al. Combination antiretroviral therapy and the risk of myocardial infarction. N Engl J Med. 2003; 349:1993–2003. [PubMed: 14627784]
5. Barbaro G. Metabolic and cardiovascular complications of highly active antiretroviral therapy for HIV infection. Curr HIV Res. 2006; 4:79–85. [PubMed: 16454713]

6. Mangili A, Jacobson DL, Gerrior J, Polak JF, Gorbach SL, Wanke CA. Metabolic syndrome and subclinical atherosclerosis in patients infected with HIV. Clin Infect Dis. 2007; 44:1368–74. [PubMed: 17443477]
7. Currier JS, Kendall MA, Zackin R, et al. Carotid artery intima-media thickness and HIV infection: traditional risk factors overshadow impact of protease inhibitor exposure. AIDS. 2005; 19:927–33. [PubMed: 15905673]
8. Mercie P, Thiebaut R, Aurillac-Lavignolle V, et al. Carotid intima-media thickness is slightly increased over time in HIV-1–infected patients. HIV Med. 2005; 6:380–7. [PubMed: 16268819]
9. Bots ML, Hoes AW, Hofman A, Witteman JC, Grobbee DE. Cross-sectionally assessed carotid intima-media thickness relates to long-term risk of stroke, coronary heart disease and death as estimated by available risk functions. J Intern Med. 1999; 245:269–76. [PubMed: 10205589]
10. Espeland MA, O'Leary DH, Terry JG, Morgan T, Evans G, Mudra H. Carotid intimal-media thickness as a surrogate for cardiovascular disease events in trials of HMG-CoA reductase inhibitors. Curr Control Trials Cardiovasc Med. 2005; 6:3. [PubMed: 15760471]
11. Ogata T, Yasaka M, Yamagishi M, Seguchi O, Nagatsuka K, Minematsu K. Atherosclerosis found on carotid ultrasonography is associated with atherosclerosis on coronary intravascular ultrasonography. J Ultrasound Med. 2005; 24:469–74. [PubMed: 15784765]
12. O'Leary DH, Polak JF, Kronmal RA, Manolio TA, Burke GL, Wolfson SK Jr. Carotid-artery intima and media thickness as a risk factor for myocardial infarction and stroke in older adults. Cardiovascular Health Study Collaborative Research Group. N Engl J Med. 1999; 340:14–22. [PubMed: 9878640]
13. McComsey GA, O'Riordan M, Hazen SL, et al. Increased carotid intima media thickness and cardiac biomarkers in HIV infected children. AIDS. 2007; 21:921–7. [PubMed: 17457085]
14. Hsue PY, Lo JC, Franklin A, et al. Progression of atherosclerosis as assessed by carotid intima-media thickness in patients with HIV infection. Circulation. 2004; 109:1603–8. [PubMed: 15023877]
15. Lorenz MW, Stephan C, Harmjanz A, et al. Both long-term HIV infection and highly active antiretroviral therapy are independent risk factors for early carotid atherosclerosis. Atherosclerosis. 2008; 196:720–6. [PubMed: 17275008]
16. Thiebaut R, Aurillac-Lavignolle V, Bonnet F, et al. Change in atherosclerosis progression in HIV-infected patients: ANRS Aquitaine Cohort, 1999–2004. AIDS 2005. 19:729–31.
17. Modrich, L.; Scherzer, R.; Zolopa, A., et al. Factors associated with mortality in the study of fat redistribution and metabolic change in HIV infection [abstract 706].. Program and abstracts of the 16th Conference on Retroviruses and Opportunistic Infections; Montreal, Canada. 2009;
18. Amar J, Fauvel J, Drouet L, et al. Interleukin 6 is associated with subclinical atherosclerosis: a link with soluble intercellular adhesion molecule 1. J Hypertens. 2006; 24:1083–8. [PubMed: 16685208]
19. Pai JK, Pischon T, Ma J, et al. Inflammatory markers and the risk of coronary heart disease in men and women. N Engl J Med. 2004; 351:2599–610. [PubMed: 15602020]
20. Ridker PM, Rifai N, Rose L, Buring JE, Cook NR. Comparison of C-reactive protein and low-density lipoprotein cholesterol levels in the prediction of first cardiovascular events. N Engl J Med. 2002; 347:1557–65. [PubMed: 12432042]
21. Irwin MW, Mak S, Mann DL, et al. Tissue expression and immunolocalization of tumor necrosis factor-α in postinfarction dysfunctional myocardium. Circulation. 1999; 99:1492–8. [PubMed: 10086975]
22. Ridker PM, Rifai N, Pfeffer M, Sacks F, Lepage S, Braunwald E. Elevation of tumor necrosis factor-α and increased risk of recurrent coronary events after myocardial infarction. Circulation. 2000; 101:2149–53. [PubMed: 10801754]
23. Brennan ML, Penn MS, Van Lente F, et al. Prognostic value of myeloperoxidase in patients with chest pain. N Engl J Med. 2003; 349:1595–604. [PubMed: 14573731]
24. Zhang R, Brennan ML, Fu X, et al. Association between myeloperoxidase levels and risk of coronary artery disease. JAMA. 2001; 286:2136–42. [PubMed: 11694155]
25. Hwang SJ, Ballantyne CM, Sharrett AR, et al. Circulating adhesion molecules VCAM-1, ICAM-1, and E-selectin in carotid atherosclerosis and incident coronary heart disease cases: the

Atherosclerosis Risk In Communities (ARIC) study. Circulation. 1997; 96:4219–25. [PubMed: 9416885]

26. Chung NA, Lydakis C, Belgore F, Li-Saw-Hee FL, Blann AD, Lip GY. Angiogenesis, thrombogenesis, endothelial dysfunction and angio-graphic severity of coronary artery disease. Heart. 2003; 89:1411–5. [PubMed: 14617549]
27. Davies MJ, Gordon JL, Gearing AJ, et al. The expression of the adhesion molecules ICAM-1, VCAM-1, PECAM, and E-selectin in human atherosclerosis. J Pathol. 1993; 171:223–9. [PubMed: 7506307]
28. Thakore AH, Guo CY, Larson MG, et al. Association of multiple inflammatory markers with carotid intimal medial thickness and stenosis (from the Framingham Heart Study). Am J Cardiol. 2007; 99:1598–602. [PubMed: 17531588]
29. Leinonen ES, Hiukka A, Hurt-Camejo E, et al. Low-grade inflammation, endothelial activation and carotid intima-media thickness in type 2 diabetes. J Intern Med. 2004; 256:119–27. [PubMed: 15257724]
30. Bowden DW, Lange LA, Langefeld CD, et al. The relationship between C-reactive protein and subclinical cardiovascular disease in the Diabetes Heart Study (DHS). Am Heart J. 2005; 150:1032–8. [PubMed: 16290992]
31. Moussavi N, Renier G, Roussin A, Mamputu JC, Buithieu J, Serri O. Lack of concordance between plasma markers of cardiovascular risk and intima-media thickness in patients with type 2 diabetes. Diabetes Obes Metab. 2004; 6:69–77. [PubMed: 14686966]
32. Tabib A, Leroux C, Mornex JF, Loire R. Accelerated coronary atherosclerosis and arteriosclerosis in young human-immunodeficiency-virus–positive patients. Coron Artery Dis. 2000; 11:41–6. [PubMed: 10715805]
33. Aukrust P, Muller F, Lien E, et al. Tumor necrosis factor (TNF) system levels in human immunodeficiency virus–infected patients during highly active antiretroviral therapy: persistent TNF activation is associated with virologic and immunologic treatment failure. J Infect Dis. 1999; 179:74–82. [PubMed: 9841825]
34. Torriani, F.; Parker, R.; Murphy, F.; Stein, J. Antiretroviral therapy improves endothelial function in treatment-naive HIV-infected subjects: a prospective, randomised multicenter trial (A5152S).. Program and abstracts of the 10th European AIDS Conference; Dublin. 2005;
35. Ross AC, Armentrout R, O'Riordan MA, et al. Endothelial activation markers are linked to HIV status and are independent of antiretroviral therapy and lipoatrophy. J Acquir Immune Defic Syndr. 2008; 49:499–506. [PubMed: 18989230]
36. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia. 1985; 28:412–9. [PubMed: 3899825]
37. O'Leary, DH.; Polak, JF.; Wolfson, SK., Jr, et al. Stroke. Vol. 22. The Cardiovascular Health Study. CHS Collaborative Research Group; 1991. Use of sonography to evaluate carotid atherosclerosis in the elderly.; p. 1155-63.
38. Triant, V.; Meigs, J.; Grinspoon, S. Association of C-reactive protein and HIV infection with acute myocardial infarction [abstract O–05].. Program and abstracts of the 10th International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV; London. 2008; p. 17
39. Lorenz MW, von Kegler S, Steinmetz H, Markus HS, Sitzer M. Carotid intima-media thickening indicates a higher vascular risk across a wide age range: prospective data from the Carotid Atherosclerosis Progression Study (CAPS). Stroke. 2006; 37:87–92. [PubMed: 16339465]
40. Kaplan RC, Kingsley LA, Gange SJ, et al. Low $CD4^+$ T-cell count as a major atherosclerosis risk factor in HIV-infected women and men. AIDS. 2008; 22:1615–24. [PubMed: 18670221]
41. Fan AZ, Paul-Labrador M, Merz CN, Iribarren C, Dwyer JH. Smoking status and common carotid artery intima-medial thickness among middle-aged men and women based on ultrasound measurement: a cohort study. BMC Cardiovasc Disord. 2006; 6:42. [PubMed: 17067397]
42. Bongiovanni M, Casana M, Cicconi P, et al. Predictive factors of vascular intima media thickness in HIV-positive subjects. J Antimicrob Chemother. 2008; 61:195–9. [PubMed: 17999980]

43. Kawamoto R, Ohtsuka N, Ninomiya D, Nakamura S. Association of obesity and visceral fat distribution with intima-media thickness of carotid arteries in middle-aged and older persons. Intern Med. 2008; 47:143–9. [PubMed: 18239322]

44. Komulainen P, Kivipelto M, Lakka TA, et al. Carotid intima-media thickness and cognitive function in elderly women: a population-based study. Neuroepidemiology. 2007; 28:207–13. [PubMed: 17851259]

45. Giordano TP, Kramer JR. Does HIV infection independently increase the incidence of lung cancer? Clin Infect Dis. 2005; 40:490–1. [PubMed: 15668878]

Table 1

Clinical Characteristics and Fasting Metabolic Parameters in Human Immunodeficiency Virus (HIV)–Infected Patients and Healthy Control Subjects

Characteristic	HIV-infected patients (*n* = 73)	Control subjects (*n* = 21)	*P*
Age	48 (21–70)	43 (21–65)	.11
Male sex	59 (81)	12 (57)	.03
Race			
African-American	28 (38)	5 (24)	.41
White	33 (45)	15 (71)	
Hispanic	7 (10)	1 (5)	
Other	5 (7)	0 (0)	
BMI, median kg/m^2 (range)	26.0 (19.8–40.8)	27.2 (18.7–34.0)	.82
Systolic blood pressure, median mm Hg (range)	120 (90–160)	113 (90–126)	.003
Diastolic blood pressure, median mm Hg (range)	80 (58–110)	70 (58–90)	.006
Waist...hip ratio, median (range)	0.97 (0.83–1.22)	0.88 (0.69–1.00)	<.001
Total cholesterol, median mg/dL (range)	182 (103–336)	207 (120–286)	.07
HDL cholesterol, median mg/dL (range)	38 (21–93)	53 (28–87)	<.001
LDL cholesterol, median mg/dL (range)	107 (23–228)	131 (54–182)	.08
Non-HDL cholesterol, median mg/dL (range)	144 (61–280)	154 (67–258)	.71
Triglycerides, median mg/dL (range)	169 (42–716)	78 (38–1138)	.30
HOMA-IR, median value (range)	2.8 (0.49–37.13)	1.52 (0.52–3.89)	<.001
Smoking habits			
Current	25 (38)	2 (10)	.04
Past	12 (18)	3 (15)	
Never	29 (38)	15 (75)	
Low-dose aspirin use	11 (15)	0 (0)	.06
Antihypertensive use[a]	14 (19)	3 (14)	.75
Lipid-lowering medication use[b]	20 (28)	0 (0)	.005
Hypertriglyceridemia medication use[c]	18 (25)	0 (0)	.01
Family history of premature CVD	4 (19)	0 (0)	<.001
Family history of type-2 diabetes	4 (19)	13 (18)	.90
HIV duration, median months (range)	162 (37–310)	...	
Nadir CD4 cell count, median cells/mm^3 (range)	162 (0–868)	...	
Current CD4 cell count, median cells/mm^3 (range)	624 (154–1718)	...	
HIV-1 RNA level <50 copies/mL	59 (81)	...	
HIV-1 RNA level in subjects with >50 copies/mL,[d] median cells/mm^3 (range)	292 (68–1606)	...	
Cumulative duration of PI therapy, median months (range)	53 (0–201)	...	
Cumulative duration of NRTI therapy, median months (range)	96 (10–219)	...	
Cumulative duration of NNRTI therapy, median months (range)	17 (0–95)	...	

NOTE. Data are no. (%) of patients, unless otherwise indicated. BMI, body mass index (calculated as weight in kilograms divided by the square of height in meters); CVD, cardiovascular disease; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein; NNRTI, nonnucleoside analogue reverse-transcriptase inhibitor; NRTI, nucleoside analogue reverse-transcriptase inhibitor; PI, protease inhibitor.

[a] Antihypertensives include diuretics, β-blockers, angiotensive-converting inhibitors, angiotensin II receptor blockers, and calcium channel blockers.

[b] Lipid-lowering medications include statins, niacin, bile acid sequestrants, and cholesterol absorption inhibitors.

[c] Hypertriglyceridemia medications includes fish oil and fibrates.

[d] $n = 14$.

Table 2

Carotid Intima-Media Thickness and Biomarkers in Human Immunodeficiency Virus (HIV)–Infected Patients and Healthy Controls Subjects

	Median value (range)		
Measurement	**HIV-infected patients (*n* = 73)**	**Control subjects (*n* = 21)**	***P***
ICA IMT, mm	1.35 (0.7–3.88)	1.10 (0.55–2.98)	.04
CCA IMT, mm	1.25 (0.75–3.23)	1.05 (0.75–1.35)	<.001
TNF-*α*, pg/mL	4.0 (0.4–364)	1.8 (0.4–85.6)	.03
sTNFR-I, pg/mL	702 (161–17, 328)	618 (265–1365)	.09
sTNFR-II, pg/mL	518 (100–3073)	635 (279–1070)	.89
hsCRP, mg/L	5.43 (0.13–84.6)	1.53 (0.05–24.4)	<.001
IL-6, pg/mL	6.0 (0.8–270)	2.8 (1.0–8.2)	.008
MPO, pg/mL	7665 (1022–32,439)	1561 (1124–10,865)	<.001
sICAM-1, ng/mL	401 (73–2818)	403 (195–577)	.25
sVCAM-1, ng/mL	659 (239–17,145)	454 (66–1808)	.002
vWF, U/mL	17.0 (2.0–3121)	11.8 (6.4–200)	.11

NOTE. CCA, common carotid artery; hsCRP high sensitivity C-reactive protein; ICA , internal carotid artery; IL, interleukin; IMT, intima-media thickness; MPO, myeloperoxidase; sICAM-1, soluble intercellular adhesion molecule-1; sTNFR-I , soluble tumor necrosis factor–*α* receptor I; sTNFR-II, soluble tumor necrosis factor–*α* receptor II; sVCAM-1, soluble vascular cell adhesion molecule-1; TNF-*α*, tumor necrosis factor–*α*; vWF, von Willebrand factor.

Table 3

Significant Univariable Correlations between Carotid Intima-Media Thickness and Biomarkers for HIV-Infected Patients and Healthy Control Subjects

Marker	HIV-infected patients (*n* = 73)				Control subjects (*n* = 21)			
	CCA IMT[a]	*P*	ICA IMT[a]	*P*	CCA IMT[a]	*P*	ICA IMT[a]	*P*
Age	0.54	.001	0.47	.001	0.66	.001	0.53	.01
Non-HDL cholesterol	0.27	.02	0.25	.04	...		...	
Triglycerides	0.25	.03	...		0.6	.004	...	
HOMA-IR score	0.24	.04	...		...		0.44	.05
hsCRP	...		0.26	.03	0.47	.03	...	
sVCAM-1	...		...		...		–0.70	<.001

NOTE. No significant correlations were found between IMT and biomarkers not listed in the table (ie, TNF-α, sTNFR-I, sTNFR-II, IL-6, MPO, sICAM-1, and vWF), as well as between IMT and TC, HDL, LDL, or BMI. BMI, body mass index (calculated as weight in kilograms divided by the square of height in meters); CCA, common carotid artery; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; hsCRP, high-sensitivity C-reactive protein; ICA, internal carotid artery; IL, interleukin; IMT, intima-media thickness; LDL, low-density lipoprotein; MPO, myeloperoxidase; sICAM-1, soluble intercellular adhesion molecule-1; sTNFR-I, soluble tumor necrosis factor-α receptor I; sTNFR-II, soluble tumor necrosis factor-α receptor II; sVCAM-1, soluble vascular cell adhesion molecule-1; TC, total cholesterol; TNF-α, tumor necrosis factor-α; vWF, von Willebrand factor.

[a] Spearman correlation coefficient (ρ).

Table 4

Variable Selection of Regression Model for Human Immunodeficiency Virus (HIV)–Infected Patients

	ICA			CCA		
Model stage, variable	**β ± SE**	***P***	**Adjusted R^2**	**β ± SE**	***P***	**Adjusted R^2**
First stage						
Demographics characteristic			0.20			0.35
Age	0.02 ± 0.01	.15		0.02 ± <0.01	.001	
BMI	–0.02 ± 0.02	.39		±0.01 ± 0.01	.95	
Sex	–0.03 ± 0.27	.93		–0.04 ± 0.13	.73	
lipid-lowering drug use	0.36 ± 0.19	.06		–0.07 ± 0.09	.49	
Waist-to-hip ratio	2.66 ± 1.39	.06		1.75 ± 0.69	.01	
Systolic blood pressure	0.01 ± <0.01	.11		±0.01 ± <0.01	.23	
Laboratory value			<0.01			0.02
Non-HDL cholesterol	±0.01 ± <0.01	.45		±0.01 ± <0.01	.25	
Triglycerides	±0.01 ± <0.01	.37		±0.01 ± <0.01	.62	
Total cholesterol	±0.01 ± <0.01	.99		±0.01 ± <0.01	.45	
HOMA-IR	0.01 ± 0.02	.56		±0.01 ± 0.01	.56	
HIV infection measurement			0.05			<0.01
Absolute CD4 cell count	±0.01 ± <0.01	.44		±0.01 ± <0.01	.67	
Nadir CD4 cell count	±0.01 ± <0.01	.86		±0.01 ± <0.01	.78	
HIV duration	0.03 ± 0.02	.25		±0.01 ± 0.01	.70	
NNRTI receipt duration	±0.01 ± <0.01	.05		±0.01 ± <0.01	.19	
NRTI receipt duration	±0.01 ± <0.01	.05		±0.01 ± <0.01	.20	
PI receipt duration	±0.01 ± <0.01	.03		±0.01 ± <0.01	.40	
Biomarker			0.10			0.03
sVCAM	±0.01 ± <0.01	.04		±0.01 ± <0.01	.02	
MPO	±0.01 ± <0.01	.04		±0.01 ± <0.01	.07	
vWF	±0.01 ± <0.01	.02		±0.01 ± <0.01	.08	
TNF-α	±0.01 ± <0.01	.03		±0.01 ± <0.01	.13	
sTNFR-I	±0.01 ± <0.01	.58		±0.01 ± <0.01	.54	
sTNFR-II	±0.01 ± <0.01	.64		±0.01 ± <0.01	.36	
ICAM-1	±0.01 ± <0.01	.98		±0.01 ± <0.01	.55	
hsCRP	±0.01 ± <0.01	.79		±0.01 ± <0.01	.36	
IL-6	±0.01 ± <0.01	.25		±0.01 ± <0.01	.21	
Second stage			0.23			0.40
Age	0.03 ± 0.01	.01		0.03 ± <0.01	<.001	
Waist-to-hip ratio	1.13 ± 1.31	.40		1.39 ± 0.61	.03	
Lipid-lowering drug use	0.34 ± 0.18	.07		–0.07 ± 0.09	.43	
NNRTI receipt duration	±0.01 ± <0.01	.81		...		
NRTI receipt duration	±0.01 ± <0.01	.36		...		
PI receipt duration	±0.01 ± <0.01	.33		...		
sVCAM-1	±0.01 ± <0.01	.02		±0.01 ± <0.01	.34	

	ICA			CCA		
Model stage, variable	**β ± SE**	***P***	**Adjusted R^2**	**β ± SE**	***P***	**Adjusted R^2**
MPO	±0.01 ± <0.01	.06		±0.01 ± <0.01	.04	
vWF	±0.01 ± <0.01	.27		±0.01 ± <0.01	.22	
TNF-α	±0.01 ± <0.01	.05		±0.01 ± <0.01	.16	
Final model			0.27			0.38
Age	0.03 ± <0.01	<.001		0.03 ± <0.01	<.001	
Waist-to-hip ratio	...			1.53 ± 0.59	.01	
Lipid-lowering drug use	0.30 ± 0.16	.07		...		
sVCAM-1	±0.01 ± <0.01	.02		...		
MPO	±0.01 ± <0.01	.03		...		
TNF-α	±0.01 ± <0.01	.03		±0.01 ± <0.01	.11	

NOTE. BMI, body mass index (calculated as weight in kilograms divided by the square of height in meters); CCA, common carotid artery; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; hsCRP high-sensitivity C-reactive protein; ICA, internal carotid artery; IL, interleukin; IMT, intima-media thickness; MPO, myeloperoxidase; NNRTI, nonnucleoside analogue reverse-transcriptase inhibitor; NRTI, nucleoside analogue reverse-transcriptase inhibitor; PI, protease inhibitor; SE, standard error; sICAM-1, soluble intercellular adhesion molecule-1; sTNFR-I, soluble tumor necrosis factor-α receptor I; sTNFR-II, soluble tumor necrosis factor-α receptor II; sVCAM-1, soluble vascular cell adhesion molecule-1; TNF-α, tumor necrosis factor-α; vWF, von Willebrand factor.