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# Perivascular Fat, Inflammation, and Cardiovascular Risk in HIV-infected Patients on Antiretroviral Therapy

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## Abstract

**Background—**HIV-infection is characterized by chronic immune activation that persists despite effective antiretroviral therapy (ART) and is associated with elevated cardiovascular risk. Whether specific perivascular fat depots are associated with inflammation in HIV is unknown.

**Methods—**In a cross-sectional study, epicardial (EAT) and thoracic periaortic (TAT) adipose tissue volume were measured by computed tomography in 100 HIV-infected adults, on stable ART, with LDL-cholesterol ≤130mg/dL and evidence of heightened T-cell activation (CD8+CD38+HLA−DR+ ≥19%) or increased inflammation (high sensitivity C-reactive protein ≥2mg/L).

**Results—**Overall, 77% were male and 70% African American. Mean (standard deviation) age and body mass index were 47 (10) years and 28 (6.4) kg/m$^2$, respectively. All subjects had HIV-1 RNA <1,000 copies/mL with mean (standard deviation) CD4+ T cell count of 665 (280) cells/μL; 50% were on a protease inhibitor. EAT and TAT were correlated with each other (r=0.766, p<0.0001). Both were associated with metabolic syndrome, atherogenic lipid profile, insulin resistance, total and central body fat, serum biomarkers of inflammation, and soluble CD163, but not with cellular immune activation markers. In multivariable models that adjusted for age, sex, and other measures of adiposity, both perivascular fat depots were independently associated with the presence of coronary calcium.

**Conclusions—**Perivascular fat is associated with soluble CD163, biomarkers of inflammation, insulin resistance, and subclinical atherosclerosis in this population of virologically suppressed



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HIV-infected patients on ART. The association of perivascular fat with coronary artery calcification appears to be independent of other measures of adiposity.

### Keywords

Adipose tissue; Atherosclerosis; HIV; Inflammation; Macrophages

## Introduction

Although the incidence of peripheral lipoatrophy appears to have decreased after the introduction of more metabolically favorable antiretroviral therapy (ART) regimens [1-3], visceral lipohypertrophy remains a common and important complication of HIV infection. This phenotype of adiposopathy (i.e. “sick fat”) is linked to chronic inflammation and immune activation[4], elevated cardiovascular risk[5], and mortality[6] in patients on ART.

Epicardial adipose tissue is a perivascular fat depot of particular interest because of dense inflammatory cell infiltration and higher production of NF-κB-dependent inflammatory cytokines such as tumor-necrosis factor-α and interleukin-6 (IL-6) compared to subcutaneous adipose tissue[7-9]. Since epicardial fat envelopes the coronary vessel adventitia without fascial separation, pathologic inflammation in the fat may promote the growth of atherosclerotic plaque in an ‘outside-in’ fashion[10]. Epicardial fat is quantitatively increased in HIV compared to un-infected controls[11] and is associated with the presence of coronary artery calcification[12] and prevalent cardiovascular disease[13].

Similarly, periaortic adipose tissue may play an important role in mediating aortic disease, although comparatively fewer studies have examined this. Periaortic fat has been associated with coronary and abdominal aortic calcification[14] as well as lower extremity peripheral arterial disease[15] in the Framingham Heart Study. HIV-infected patients appear to have increased aortic wall inflammation by 18fluorine-2-deoxy-D-glucose positron emission tomography (FDG-PET) compared to uninfected controls[16]; however, to our knowledge, no study has described periaortic fat in HIV infection.

Whether perivascular fat depots contribute to the elevated systemic inflammation seen in HIV independent of other types of abnormal fat distribution is unknown. In this study, we therefore aimed to comprehensively examine the association of epicardial and periaortic fat depots with clinical and radiologic measures of adiposity, subclinical atherosclerosis, and systemic biomarkers of inflammation and immune activation in HIV-infected patients on ART.

## Methods

### Study design

We performed a cross-sectional analysis of 100 subjects who underwent initial comprehensive metabolic and cardiovascular assessments at the time of enrollment into the **S**topping **A**therosclerosis and **T**reating **U**nhealthy bone with **R**osuvastati**N** in HIV (SATURN-HIV) trial. SATURN-HIV is a randomized double-blind placebo-controlled trial designed to measure the effect of rosuvastatin 10mg daily on the progression of subclinical vascular disease. Enrollment spanned from March 2011 to January 2012. All subjects were ≥18 years of age, on stable ART with viral load <1,000 copies/mL, without known coronary disease or diabetes, and with a fasting LDL-cholesterol (LDL-C) ≤130mg/dL. Additional entry criteria included evidence of heightened T-cell activation (CD8+CD38+HLA−DR+ ≥19%) and/or increased inflammation (high sensitivity C-reactive protein (hs-CRP≥2mg/L). The study was approved by the Institutional Review Board of University Hospitals Case

Medical Center (Cleveland, OH) and written informed consent was obtained from each study subject. The authors of this manuscript have certified that they comply with the Principles of Ethical Publishing in the International Journal of Cardiology[17].

### Study evaluations

At the initial screening visit, self-reported demographics and medical history were obtained along with a targeted physical exam including height, weight, waist, and hip measurements. Blood was drawn after at least a 12-hour fast for glucose, insulin, and lipoproteins. If enrollment criteria were met, subjects returned within 30 days for entry evaluations. HIV-1 RNA level and CD4+ cell count were obtained as part of routine clinical care.

### Inflammation and Coagulation Markers

Several serum biomarkers of inflammation were measured: hs-CRP, IL-6, soluble tumor necrosis factor-α receptor I (sTNFR-I), and two adhesion molecules that are considered markers of endothelial activation—soluble vascular cell adhesion molecule-1 (sVCAM-1) and soluble intercellular adhesion molecule-1 (sICAM-1). D-dimer and fibrinogen were measured as serum markers of coagulation.

IL-6, sTNFR-I, sVCAM-1, and sICAM-1 were determined by quantitative sandwich ELISAs (R&D Systems, Minneapolis, MN). Inter-assay variability ranged from 2.02%-15.36%, 3.66%-5.77%, 4.76%-8.77%, and 3.43%-7.37%, respectively. Hs-CRP and fibrinogen were determined by particle enhanced immunonepholometric assays on a BNII nephelometer (Siemens). Inter-assay variability ranged from 3.01%-6.46% and 3.42%-7.59%, respectively. D-dimer was determined by immuno-turbidometric assay on a STA-R Coagulation Analyzer (DiagnosticaStago). Inter-assay variability ranged from 1.54%-9.03%.

### Immune Activation Markers

The serum levels of two monocyte activation markers, soluble CD14 and soluble CD163, were measured by ELISA (R&D Systems, Minneapolis, MN). Inter-assay variability ranged from 0.4-8.6% for soluble CD14 and 0.7-18.3% for sCD163.

Monocytes and CD8+ T-cells were identified by size, granularity, and by expression of CD14 or CD3 and CD8, respectively. Monocyte phenotype was monitored by staining cells with the following fluorochrome-labeled antibodies: anti-Tissue Factor fluorescein isothiocyanate (FITC) (American Diagnostica, Stamford, CT), anti-CD14 Pacific Blue, anti-CD16 phycoerythrin (PE), (BD Pharmingen, San Diego, CA). In order to assure that monocyte populations were not contaminated by lymphocytes, preliminary experiments were performed that excluded CD3, CD20, and CD56 expressing cells.

T-cell activation was measured using anti-CD38 PE, anti-HLA-DR FITC, anti-CD3 Peridinin-chlorophyll-protein Complex (PerCP), anti-CD8 PerCP (BD Biosciences), and appropriate isotype control monoclonal antibodies.

Whole blood samples were incubated for 15 minutes on ice with FACS Lyse buffer (BD Biosciences) and then washed in wash buffer (phosphate buffered saline with 1% bovine serum albumin and 0.1% sodium azide). Cells were then stained for 30 minutes in the dark on ice and then washed in wash buffer, fixed in 1% formaldehyde. Monocytes were analyzed using a Miltenyi MACS Quant flow cytometer (MiltenyiBiotec, BergischGladbach, Germany) and MACS Quant software (version 2.21031.1, MiltenyiBiotec). T-lymphocytes were analyzed using an LSR II flow cytometer (Becton-

Dickinson, San Jose, CA) and FACSDiva software version 6.1.1 (BD Biosciences, San Diego, CA).

### Dual-Energy X-ray Absorptiometry

Evaluations included whole-body dual-energy absorptiometry (DEXA) scans. Fat distribution was measured by DEXA in the anteroposterior view using Lunar Prodigy Advance (GE Healthcare). Whole body fat volumes as well as peripheral fat depots (limb and arm fat) and central fat depots (trunk fat) were used for analysis. Technicians used the same machine on the same subject throughout the study. All DEXA scans were read at University Hospitals Case Medical Center by an experienced radiologist blinded to study information.

### Coronary Calcium, Epicardial Fat, and Periaortic Fat

All subjects had a baseline computed tomography (CT) scan of the chest to quantify perivascular adipose tissue volume and coronary artery calcium score. A 64-slice multidetector CT scanner (Somatom Sensation 64, Siemens Medical Solutions USA) was used with 30 × 0.6mm collimation, 330ms rotation time, and 120kV tube voltage. Three-millimeter slices were obtained from the carina to the diaphragm with prospective ECG gating at 60% of the R-R interval.

Measurements were performed offline (Aquarius iNtuition Cloud, Terarecon, San Mateo, CA USA) by a single reader (CTL) on a single workstation using methods described previously[18]. Briefly, calcified coronary lesions were defined as areas of ≥6 pixels with density >130 Hounsfield units (HU). Total coronary calcium score was quantified using the Agatston method. Epicardial adipose tissue volumes were quantified using a semi-automatic segmentation technique in which the region of interest was defined by manually tracing the pericardial borders. Fat tissue was defined as pixels within a window of −195 to −45 HU, and epicardial fat was then selected as adipose tissue within the pericardial sac. Previous studies have used the term “pericardial” adipose tissue to describe this depot; however, because most of the fat within the pericardial sac is true epicardial fat, and to be consistent across studies in HIV, we opted to use the term “epicardial”. Similarly, thoracic periaortic adipose tissue was defined as all adipose tissue surrounding the thoracic aorta extending 69mm caudally from the level of the bifurcation of the pulmonary arteries. This method has been validated in the Framingham Heart Study and other populations[14, 18].

### Carotid Artery Ultrasound

A high resolution B-mode ultrasound scan of the carotid arteries was performed at the entry visit using a Philips iU22 ultrasound system with a L9-3 MHz linear array transducer according to the consensus protocol of the American Society of Echocardiography[19]. Complete scans of the bilateral common carotid arteries (CCA), internal carotid arteries (ICA), and external carotid arteries (ECA) were used to identify plaque, defined as IMT >1.5cm or > 50% thicker than the adjacent vessel. R-wave gated still frame images of the distal 1cm of the CCA far wall were obtained at 3 separate angles bilaterally (anterior, lateral, and posterior). CCA-IMT was measured offline by a single reader (CTL) using semi-automated edge detection software (Medical Imaging Applications LLC, Coralville, IA). The mean-mean and mean-max CCA-IMT was used for analysis.

### Statistical Analysis

Variables were examined for departures from normality of their distributions and log transformed when appropriate. Continuous variables were summarized using medians and inter-quartiles ranges and categorical variables with frequencies and percentages. The

correlation between each of the perivascular fat depots (EAT and TAT) with the measures of interest was estimated using Spearman's correlation coefficient. In parallel analyses, multiple linear regression expressed the relationship of log EAT and separately of log TAT with selected covariates after adjusting for the contribution of traditional risk factors and clinical measures of adiposity. To further sharpen potential associations with presence of CAC, the highest and lowest quartiles of the distribution of EAT, and separately of TAT, were used as outcomes in binary logistic regressions, also adjusting for traditional risk factors. All statistical tests were two-sided with a 0.05 significance level. Nominal p-values are presented throughout. Analyses were performed with SAS version 9.2 (SAS Institute, Cary, NC).

## Results

### Baseline characteristics

Baseline characteristics of study participants are displayed in Table 1. Approximately three-quarters were male and three-quarters were African American. HIV disease was well-controlled (mean CD4+ count was 665 cells/μL and 81% had HIV-1 RNA < 50 copies/mL), although participants were pre-selected to have higher levels of either CD8+ T-cell activation or hs-CRP. Half of the subjects were on a protease inhibitor-based regimen; however, only 4% were currently taking a thymidine analog nucleoside reverse transcriptase inhibitor and 12% were currently taking abacavir. One quarter of participants had metabolic syndrome as defined by Adult Treatment Panel III criteria[20]. Despite a low LDL-C (all ≤130mg/dL), over a third of patients had evidence of non-obstructive carotid plaque and a third of patients had detectable coronary artery calcium (CAC).

### Perivascular fat and traditional risk factors

Log-transformed epicardial and thoracic periaortic fat volumes were strongly correlated with each other; although, periaortic fat was more strongly correlated with age (Figure 1). Epicardial fat did not differ by gender [median(IQR) 69(52-95) vs. 68(45-91) $cm^3$, male vs. female, p= 0.475]; but periaortic fat volume was higher in men [9.5(7.3-15.2) vs. 7.1(5.2-9.6) $cm^3$, p = 0.003]. Statistically significant correlations were observed between epicardial and periaortic fat and several clinical and radiologic measures of adiposity, including positive associations with both trunk fat and leg fat (Table 2). Additionally, both were significantly correlated with the metabolic syndrome and its individual components (hypertension, lower HDL, higher triglycerides, insulin resistance, and waist circumference; Figure 2). A strong and statistically significant relationship between epicardial fat and insulin resistance persisted in multivariable models (Table 3) after adjustment for age, sex, and clinical measures of adiposity (BMI and waist:hip ratio).

### Perivascular fat, HIV-related factors, and Inflammation

Higher epicardial fat volume was associated with higher current CD4+ count and lower nadir CD4+ count (Table 2); whereas, the association between nadir CD4+ count and periaortic fat was borderline statistically significant (p=0.053). Both fat depots were associated with total duration of antiretroviral therapy but not with total duration of protease inhibitor use. Several serum biomarkers of inflammation were correlated with epicardial and periaortic fat (Table 2); however, biomarkers of endothelial activation and coagulation were not associated. Notably, one marker of monocyte activation—soluble CD163—was significantly correlated with both fat depots, whereas sCD14 and percent circulating CD14+CD16+ monocytes were not.

Compared to those in the lowest quartile (<51.3$cm^3$), participants in the highest quartile of epicardial fat volume (>94$cm^3$) had significantly higher levels of inflammatory biomarkers

including 3.8-fold higher mean IL-6 (p=0.022), 1.4-fold higher TNFαRI (p = 0.006), and 1.5-fold higher sCD163 (p=0.004). Similar results were seen when the highest (>13.5cm$^3$) and lowest (<6.53cm$^3$) quartiles of periaortic fat volumes were compared (p=0.087, p=0.002, p=0.017 for IL-6, TNFαRI, and sCD163 respectively).

In multivariable linear regression models (Table 3), adjustment for age, sex, and clinical measures of adiposity partially removed associations of epicardial and periaortic fat with markers of inflammation and soluble CD163. Adjustment for trunk fat by DEXA reduced the estimated effect even further.

### Perivascular fat and subclinical atherosclerosis

Epicardial and periaortic fat were both correlated with subclinical coronary atherosclerosis as defined by the presence of detectable CAC score >0 (Table 2). An independent association between both fat depots and presence of CAC generally persisted in models that adjusted for age, sex, measures of adiposity, and traditional cardiovascular risk factors (Table 4). Participants in the highest quartile of epicardial fat had 2.2-fold higher odds of having any detectable CAC compared to those in the lowest quartile (p =0.04). Periaortic fat was significantly correlated with mean and max CCA-IMT (Table 2); however, adjustment for age and sex completely removed this association (Table 4).

## Discussion

The aim of this study was to investigate the relationship between perivascular fat depots, systemic biomarkers of inflammation, insulin resistance and subclinical atherosclerosis in HIV-infected patients on antiretroviral therapy (ART). For the first time, periaortic adipose tissue is described in this population. Our data are consistent with a model of excessive epicardial and periaortic fat as a marker of pathologic adipose tissue hypertrophy and dysfunction that contributes to chronic inflammation, insulin resistance, and subclinical atherosclerosis observed in HIV even with more metabolically favorable ART regimens.

Despite considerable variability in methods, prior studies of echocardiography-derived epicardial fat thickness and CT-derived volumes in HIV-infected patients have described similar correlations with visceral adiposity measured by MRI[21] or CT[11, 12]. Our study also confirms the associations of CT epicardial fat volume with insulin resistance[11], metabolic syndrome and its individual components[21], and coronary artery calcium[12]. Our study further demonstrates that the remarkably strong association with insulin resistance persists even after adjustment for BMI, visceral adiposity, and traditional cardiovascular disease risk factors.

HIV-infected subjects are prone to aortic aneurysms[22, 23] and have elevated levels of aortic wall inflammation[16] and atherosclerosis[24] compared to uninfected controls. Prior studies in the uninfected population have shown that periaortic fat volume increases with age and male sex, two significant risk factors for aortic aneurysms in the general population[25]. Periaortic fat has also been associated with both aortic and coronary calcification[14, 18] and peripheral arterial disease[14]. In HIV-infected patients, we demonstrate that periaortic adipose tissue volume appears to be more strongly associated with age and male sex than epicardial adipose tissue. Similarly to uninfected populations, periaortic fat was independently associated with coronary artery calcification in our study after adjustment for multiple potential confounders. Periaortic fat was also correlated with carotid IMT, a surrogate measure of abnormal arterial remodeling; although, adjustment for age and sex completely removed this association.

As in other studies of lipodystrophy and surrogate measures of cardiovascular risk in HIV[26-29], perivascular fat depots were associated with lower nadir CD4 count and longer cumulative duration of ART; although this association does not appear to be independent of age, sex, measures of adiposity, and traditional risk factors. Interestingly, neither current nor cumulative duration of protease inhibitor use was associated with perivascular fat depots in our study.

One of the principle strengths of our study was the ability to comprehensively evaluate the association of fat depots with multiple biomarkers of inflammation and immune activation. In contrast to Lo et al.[11], we found significant positive associations between perivascular adipose tissue volume and a variety of serum markers of inflammation including sCD163, a marker of monocyte activation. The pro-inflammatory role of adipose tissue is well-described[30], and the qualitative function of fat may be more important than fat quantity in mediating disease[31]. To what extent pro-inflammatory cytokines act locally in a paracrine fashion vs. systemically in an endocrine fashion is not clear; although, community based cohort studies outside of HIV have also been able to demonstrate relationships between circulating biomarkers of inflammation and quantities of visceral[32], subcutaneous[32], and perivascular fat[33].

Both macrophages[34, 35] and T-cells[36] appear to mediate inflammation in visceral and epicardial fat. In particular, sCD163, a marker of monocyte/macrophage activation, is elevated in obesity[37] and is associated with insulin resistance in the obese state[38]. While previous studies have associated sCD163 with non-calcified coronary plaque by CT angiography[39] and aortic inflammation by positron emission tomography[16], neither study adjusted for any measure of adiposity. Our findings suggest that visceral and perivascular adipose tissue volumes are associated with systemic levels of sCD163 and may partly explain the relationship between sCD163 and cardiovascular disease. On the other hand, we did not observe any relationship between perivascular fat depots and sCD14, circulating inflammatory CD14+CD16+ monocytes, or activated CD8+CD38+HLA−DR+ T-cells. Future longitudinal studies should further examine the relationship between inflamed perivascular adipose tissue and vascular disease in HIV. Future studies should also examine the relative utility of CT-derived adipose tissue volumes versus echocardiography-derived fat pad thickness in longitudinal studies of cardiovascular risk in this population.

### Study Limitations

Because the study population was pre-selected to have higher levels of inflammation or CD8+ T-cell activation, our results may not be generalizeable to the entire HIV-infected population. This may also be considered a strength, however, because it allows us to characterize a subset of HIV-infected individuals that may be at high risk of cardiovascular disease despite low LDL-C. Our study also cannot prove causality or rule out the possibility of residual confounding, although this is a limitation of all cross-sectional studies. Our study was adequately powered to detect significant relationships with several serum markers of inflammation, but may have lacked power to detect more subtle relationships with other markers of immune activation. Finally, we did not measure anti-inflammatory soluble biomarkers of inflammation; although, the classic non-inflammatory monocyte population (CD14+CD16–) was not associated with either fat depot in our study.

## Conclusions

Two important perivascular fat depots, epicardial and thoracic periaortic adipose tissue, are associated with biomarkers of inflammation, monocyte/macrophage activation, and insulin resistance in this population of virologically suppressed HIV-infected patients on ART, although many of these relationships appear dependent on other measures of adiposity.

Furthermore, both perivascular fat depots are independently associated with subclinical atherosclerosis as defined by the presence of coronary artery calcium. The effect of statins on perivascular fat volumes and inflammation will be tested in the ongoing SATURN-HIV trial.

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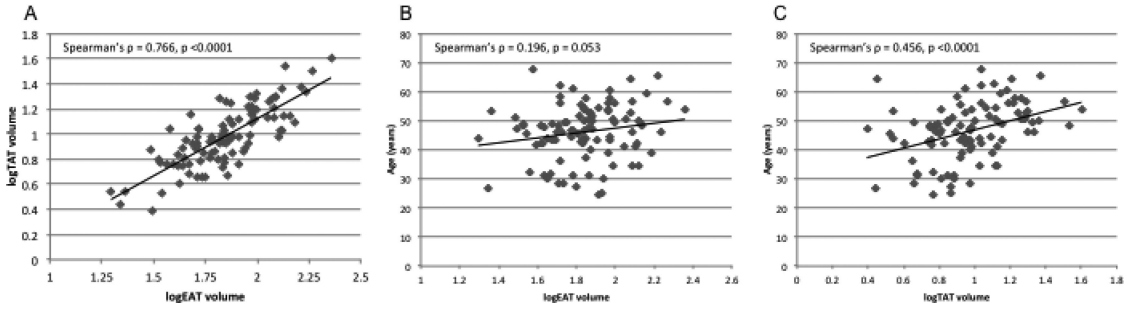


**Figure 1.**

**A)** Log transformed epicardial and thoracic periaortic adipose tissue volumes are linearly correlated to each other. **B and C)** Thoracic periaortic adipose tissue is more strongly correlated to age than epicardial adipose tissue. EAT = epicardial adipose tissue, TAT = thoracic periaortic adipose tissue.

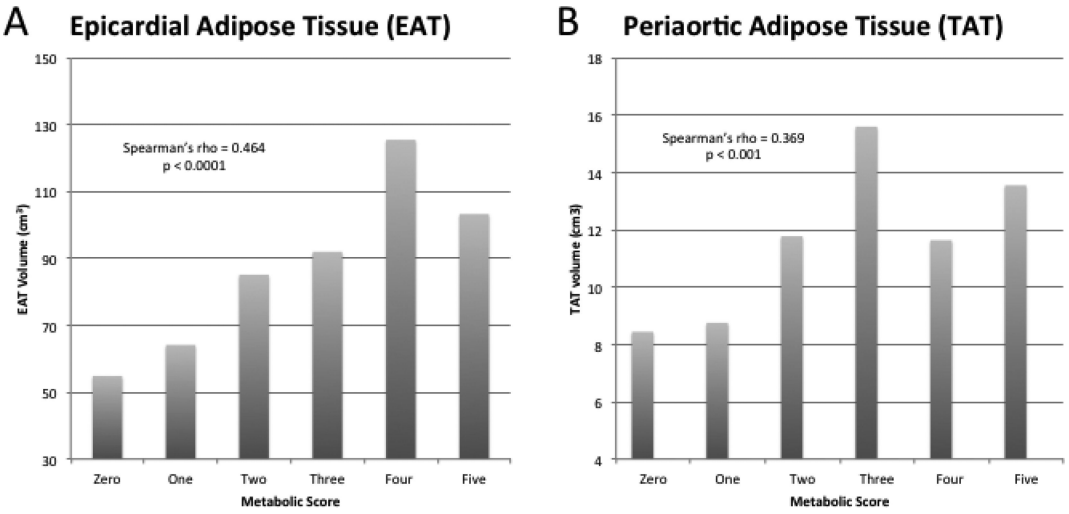


**Figure 2.**

Higher metabolic score is associated with larger mean volumes of epicardial (**A**) and thoracic periaortic (**B**) adipose tissue. Metabolic score is defined as the number of criteria for metabolic syndrome (out of 5) as defined by the Adult Treatment Panel III guidelines[20]. The five components are 1) waist circumference >102cm (men), >88cm (women); 2) Triglycerides ≥150mg/dL; 3) high density lipoprotein <40mg/dL (men), <50mg/dL (women); 4) blood pressure ≥130/≥85mmHg or on anti-hypertensive treatment; 5) fasting glucose ≥110mg/dL.

## Table 1

Baseline characteristics of study participants

| **Demographics** | **n = 100** |
|---|---|
| ***Age (years)*** | 47 (10)* |
| ***Sex*** | |
| Male | 77% |
| Female | 23% |
| ***Race*** | |
| White | 29% |
| African American | 70% |
| **HIV Parameters** | |
| ***Current CD4+ count (cells/μl)*** | 665 (280) |
| ***Nadir CD4+ count (cells/μl)*** | 199 (99-299) |
| ***HIV duration (years)*** | 13 (6.6) |
| ***Antiretroviral therapy duration (years)*** | 6.3 (3.3-9.9) |
| ***Undetectable viral load (<48copies/ml)*** | 81% |
| **Metabolic Parameters** | |
| ***Systolic Blood Pressure (mmHg)*** | 121 (15) |
| ***HDL Cholesterol (mg/dL)*** | 48 (14) |
| ***LDL Cholesterol (mg/dL)*** | 95 (25) |
| ***Homeostatic Model Assessment of Insulin Resistance*** | 1.8 (1.1-3.2) |
| ***Metabolic Syndrome*** | 25% |
| **Body Composition** | |
| ***Body Mass Index (kg/m$^2$)*** | 28 (6.4) |
| ***Total Lean Body Mass (kg)*** | 56 (11) |
| ***Total Body Fat (kg)*** | 26 (16) |
| ***Total Leg Fat (kg)*** | 8.2 (5.6) |
| ***Total Arm Fat (kg)*** | 2.3 (2.0) |
| ***Total Trunk Fat (kg)*** | 14 (8.2) |
| **Other Cardiovascular Risk Factors** | |
| ***Smoking*** | |
| Current | 62% |
| Past | 17% |
| Never | 21% |
| ***Family history of myocardial infarction*** | 32% |
| ***10-year Framingham risk score (%)*** | 4.0 (1.0-7.3) |
| **Baseline Inflammation and Immune Activation** | |
| ***High sensitivity C-reactive protein (mg/L)*** | 2.3 (0.9-4.5) |
| ***CD8+CD38+HLA-DR+ (%)*** | 27 (22-39) |

| Demographics | n = 100 |
|---|---|
| **Current Medication Use** | |
| ***Anti-hypertensive Drug*** | 22% |
| ***Fibrate*** | 5% |
| ***Fish Oil*** | 10% |
| ***Protease Inhibitor*** | 47% |
| ***AZT or D4T*** | 4% |
| ***Abacavir*** | 12% |
| **Measures of Subclinical Vascular Disease** | |
| ***Mean-mean common carotid artery IMT (μm)*** | 662 (627-803) |
| ***Mean-max common carotid artery IMT (μm)*** | 838 (773-984) |
| ***Carotid plaque (IMT > 1.5mm)*** | 38% |
| ***Coronary Artery Calcium*** | |
| 0 | 67% |
| 1-100 | 23% |
| >100 | 10% |

HDL = high-density lipoprotein, LDL = low-density lipoprotein, IMT = intima media thickness

*Data presented as median (interquartile range) for non-normally distributed continuous variables, mean (standard deviation) for normally distributed continuous variables, and percent for categorical variables

## Table 2

Relationship of epicardial and periaortic adipose tissue to HIV parameters, metabolic parameters, body composition, biomarkers of inflammation and immune activation, and sublinical atherosclerosis

| | Epicardial Adipose Tissue[§] | | Periaortic Adipose Tissue[§] | |
|---|---|---|---|---|
| **HIV Parameters** | **Spearman's ρ** | **p value** | **Spearman's ρ** | **p value** |
| ***Current CD4+ count*** | **0.207** | **0.041** | 0.092 | 0.365 |
| ***Nadir CD4+ count***[§] | **−0.206** | **0.045** | −0.197 | 0.053 |
| ***HIV duration*** | 0.076 | 0.456 | 0.168 | 0.095 |
| ***Antiretroviral therapy duration***[§] | **0.232** | **0.036** | **0.304** | **0.005** |
| ***Duration of Protease Inhibitor Use*** | 0.014 | 0.910 | 0.122 | 0.311 |
| ***Duration of NRTI Use*** | 0.141 | 0.187 | **0.254** | **0.015** |
| **Metabolic Parameters** | | | | |
| ***Systolic Blood Pressure*** | **0.230** | **0.023** | **0.227** | **0.023** |
| ***Use of Antihypertensive Medication*** | **0.215** | **0.034** | **0.258** | **0.010** |
| ***HDL Cholesterol*** | **−0.340** | **<0.001** | **−0.345** | **<0.001** |
| ***LDL Cholesterol*** | 0.044 | 0.666 | 0.051 | 0.613 |
| ***Triglycerides*** | **0.395** | **<0.0001** | **0.301** | **0.002** |
| ***HOMA-IR***[§] | **0.454** | **<0.0001** | **0.347** | **<0.001** |
| ***Metabolic Syndrome*** | **0.347** | **<0.001** | **0.321** | **0.001** |
| **Body Composition** | | | | |
| ***Body Mass Index*** | **0.529** | **<0.0001** | **0.378** | **0.0001** |
| ***Waist circumference*** | **0.650** | **<0.0001** | **0.541** | **<0.0001** |
| ***Waist:Hip Ratio*** | **0.576** | **<0.0001** | **0.648** | **<0.0001** |
| ***Total Lean Body Mass*** | 0.122 | 0.232 | **0.215** | **0.03** |
| ***Total Body Fat*** | **0.561** | **<0.0001** | **0.381** | **<0.0001** |
| ***Total Leg Fat*** | **0.361** | **<0.001** | 0.168 | 0.094 |
| ***Total Arm Fat*** | **0.571** | **<0.0001** | **0.371** | **<0.001** |
| ***Total Trunk Fat*** | **0.643** | **<0.0001** | **0.487** | **<0.0001** |
| **Biomarkers of Inflammation, Coagulation, and Immune Activation** | | | | |
| ***High sensitivity C-reactive protein*** | **0.307** | **0.002** | 0.186 | 0.064 |
| ***Interleukin-6***[§] | **0.245** | **0.016** | 0.195 | 0.054 |
| ***Soluble Tumor Necrosis Factor-α Receptor 1***[§] | **0.295** | **0.003** | **0.336** | **<0.001** |
| ***Soluble Intracellular Adhesion Molecule*** | 0.071 | 0.490 | −0.051 | 0.615 |
| ***Soluble Vascular Cell Adhesion Molecule*** | −0.014 | 0.892 | 0.082 | 0.418 |
| ***Fibrinogen*** | 0.181 | 0.075 | 0.109 | 0.279 |
| ***D-dimer*** | −0.017 | 0.868 | −0.028 | 0.785 |
| ***Percent CD8+CD38+HLA-DR+ T-cells*** | 0.032 | 0.775 | 0.063 | 0.572 |
| ***Percent CD14+CD16—Monocytes*** | −0.004 | 0.972 | 0.021 | 0.843 |
| ***Percent CD14+CD16+ Monocytes*** | 0.030 | 0.778 | 0.017 | 0.869 |

| HIV Parameters | Epicardial Adipose Tissue[§] | | Periaortic Adipose Tissue[§] | |
|---|---|---|---|---|
| | Spearman's ρ | p value | Spearman's ρ | p value |
| ***Soluble CD163***[§] | **0.282** | **0.005** | **0.274** | **0.006** |
| ***Soluble CD14*** | 0.034 | 0.738 | −0.049 | 0.627 |
| **Measures of Subclinical Atherosclerosis** | | | | |
| ***Mean-mean common carotid artery IMT***[§] | 0.087 | 0.401 | **0.275** | **0.006** |
| ***Mean-max common carotid artery IMT***[§] | 0.149 | 0.147 | **0.345** | **<0.001** |
| ***Carotid plaque (IMT > 1.5mm)*** | 0.034 | 0.742 | 0.172 | 0.087 |
| ***Coronary Artery Calcium >0*** | **0.228** | **0.024** | **0.244** | **0.014** |

NRTI = nucleoside reverse transcriptase inhibitor, HDL = high-density lipoprotein, LDL = low-density lipoprotein, HOMA-IR = Homeostatic Model Assessment of Insulin Resistance, IMT = intima media thickness

[§]Variable was log transformed to achieve a normal distribution

## Table 3

Relationship of epicardial and periaortic adipose tissue with selected biomarkers of inflammation, monocyte activation, HIV parameters, insulin resistance, and carotid intima media thickness after adjustment for other measures of adiposity and traditional risk factors.

| | age, sex | | age, sex, BMI | | age, sex, BMI, waist:hipratio | | age, sex, trunk fat | | Traditional risk factors* | |
|---|---|---|---|---|---|---|---|---|---|---|
| | **Estimate** | **P-value** | **Estimate** | **P-value** | **Estimate** | **P-value** | **Estimate** | **P-value** | **Estimate** | **P-value** |
| **Epicardial adipose tissue§** | | | | | | | | | | |
| IL-6§ | **0.371** | **<0.01** | 0.293 | 0.07 | 0.33 | 0.07 | 0.298 | 0.12 | 0.317 | 0.06 |
| sTNFR-I§ | **0.234** | **0.01** | **0.234** | **0.04** | 0.123 | 0.34 | 0.134 | 0.31 | **0.263** | **0.03** |
| sCD163§ | **0.188** | **0.02** | 0.189 | 0.06 | 0.169 | 0.13 | 0.068 | 0.53 | 0.176 | 0.10 |
| ART duration§ | 0.356 | 0.06 | 0.256 | 0.26 | 0.319 | 0.22 | 0.240 | 0.37 | 0.218 | 0.37 |
| Nadir CD4+§ | −0.416 | 0.06 | −0.305 | 0.25 | −0.410 | 0.17 | −0.192 | 0.53 | −0.359 | 0.20 |
| HOMA-IR§ | **0.873** | **<0.0001** | **0.666** | **<0.01** | **0.637** | **0.01** | **0.641** | **0.01** | **0.580** | **<0.01** |
| Mean-mean CCA-IMT§ | −0.043 | 0.20 | −0.056 | 0.18 | −0.78 | 0.10 | −0.033 | 0.50 | −0.048 | 0.24 |
| Mean-max CCA-IMT§ | −0.024 | 0.47 | −0.033 | 0.43 | −0.070 | 0.14 | −0.012 | 0.81 | −0.03 | 0.45 |
| **Periaortic adipose tissue§** | | | | | | | | | | |
| IL-6§ | **0.309** | **0.01** | 0.197 | 0.20 | 0.221 | 0.20 | 0.16 | 0.37 | 0.225 | 0.15 |
| sTNFR-I§ | **0.270** | **<0.01** | **0.292** | **<0.01** | 0.199 | 0.11 | 0.216 | 0.09 | **0.317** | **<0.01** |
| sCD163§ | 0.150 | 0.06 | 0.135 | 0.19 | 0.104 | 0.36 | 0.151 | 0.22 | 0.093 | 0.39 |
| ART duration§ | 0.231 | 0.196 | 0.092 | 0.67 | 0.099 | 0.68 | 0.016 | 0.95 | 0.033 | 0.88 |
| Nadir CD4+§ | −0.359 | 0.09 | −0.234 | 0.36 | −0.329 | 0.26 | −0.116 | 0.69 | −0.128 | 0.31 |
| HOMA-IR§ | **0.734** | **<0.0001** | **0.489** | **0.02** | 0.421 | 0.08 | 0.407 | 0.10 | 0.377 | 0.07 |
| Mean-mean CCA-IMT§ | −0.008 | 0.78 | −0.004 | 0.93 | −0.011 | 0.80 | 0.037 | 0.41 | 0.02 | 0.58 |
| Mean-max CCA-IMT§ | 0.017 | 0.60 | 0.027 | 0.50 | 0.006 | 0.89 | 0.069 | 0.13 | 0.046 | 0.24 |

BMI = body mass index, IL-6 = interleukin 6, sTNFR-I = soluble tumor necrosis factor alpha receptor 1, sCD163, ART = antiretroviral therapy, HOMA-IR = homeostasis model of insulin resistance, CCA-IMT = common carotid artery intima-media thickness

* Adjusts for age, sex, BMI, smoking status, systolic blood pressure, HDL, non-HDL, and family history of myocardial infarction

§ Variable was log transformed to achieve a normal distribution

**Table 4**

Relationship of epicardial and periaortic adipose tissue with the presence of coronary artery calcium after adjustment for other measures of adiposity and traditional risk factors.

| | age, sex | | age, sex, BMI | | age, sex, BMI, waist:hipratio | | age, sex, trunk fat | | Traditional risk factors* | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Odds Ratio | P-value | Odds Ratio | P-value | Odds Ratio | P-value | Odds Ratio | P-value | Odds Ratio | P-value |
| **Epicardial adipose tissue** | **1.015** | **0.028** | **1.032** | **0.004** | **1.029** | **0.016** | **1.042** | **0.001** | **1.032** | **0.004** |
| **Periaortic adipose tissue** | **1.083** | **0.047** | **1.148** | **0.016** | 1.114 | 0.094 | **1.181** | **0.012** | **1.147** | **0.008** |

Odds ratio per $1cm^3$ increase in adipose tissue volumes

*Adjusts for age, sex, body mass index, smoking status, systolic blood pressure, high density lipoprotein (HDL), non-HDL, and family history of myocardial infarction infarction