Poster #728

Carotid Distensibility and Immune Activation in HIV-infected Patients without Coronary Calcium

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ABSTRACT

Background: Coronary artery calcium (CAC) scoring significantly improves cardiovascular risk stratification; however, CAC is a late finding of atherosclerotic vascular disease. Increased vascular stiffness (i.e. decreased distensibility) may be an early marker of vascular disease in HIV-infected patients on antiretroviral therapy (ART) prior to the development of vascular calcification. The relationship of chronic systemic immune dysregulation to these early vascular changes in HIV is poorly understood.

Methodology: High-resolution carotid ultrasound was used to examine common carotid artery intima-media thickness (CCA-IMT) and distensibility among 147 HIV-infected patients on ART. CAC scoring was performed with computed tomography. Spearman correlations were used to examine the relationships between carotid distensibility and biomarkers of inflammation and immune activation among those with and without detectable CAC.

Results: Median (interquartile range) age was 46 (40-53) years; 78% were male and 69% African American. Median CD4 count was 613 (425, 853) cells/µl and 10-year Framingham risk score was 3(1-7)%. Ninety-three subjects (63%) had zero CAC, but 29 (31%) of those with zero CAC had carotid atherosclerosis by ultrasound (CCA-IMT>1mm or discreet carotid plaque). Median carotid distensibility was higher among those without CAC [26(19-33) vs. 23(19-26) 10^{-6}*Newtons^{-1}*m^2, p=0.045, CAC=0 vs. CAC>0, respectively); however, 13 subjects (9% of total population) without any CAC or carotid atherosclerosis had carotid distensibility below the 25th percentile. Distensibility was negatively correlated with CD4+ CD38+HLA-DR+, CD14dimCD16+ (patrolling) monocytes, and IL-6 concentrations among those with CAC=0, but not among those with CAC>0 (see Table). Among subjects with CAC=0, distensibility was significantly or marginally significantly correlated with CCA-IMT (r 0.339, p <0.001) and two measures of endothelial function (r 0.192, p=0.066 for flow-mediated brachial artery dilation; and r 0.374, p<0.001 for hyperemic velocity-time integral).

Conclusions: Among HIV-infected patients on ART who do not have coronary artery calcification, impaired carotid distensibility may be a sign of early vascular disease that is associated with immune activation and inflammation.

METHODS

Design: Cross-sectional

Subjects:

- Baseline examination of 147 subjects enrolled in 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.
- All subjects were ≥18 years of age, on stable ART with LDL-cholesterol (LDL-C) ≤130mg/dL. Additional inclusion criteria included evidence of heightened T-cell activation (CD8+CD38+HLA-DR+ ≥19%) or increased inflammation (high sensitivity C-reactive protein (hsCRP ≥2mg/L) at baseline.

Vascular Ultrasound:

- All carotid and brachial ultrasound studies were performed using a Philips iU22 and L9-3 MHz transducer. Measurements were made by a singler reader (CTL) with semi-automated edge detection software (Medical Imaging Aplications, LLC).
- For ***carotid distensibility***, the diameter of the distal 1cm of the right carotid artery was measured in systole (Ds) and diastole (Dd). Blood pressure was obtained at the time of carotid ultrasound in order to determine the pulse pressure (PP). Carotid distensibility was calculated using the same formula [(2*(Ds-Dd)/Dd)/PP] used in the Women's Interagency Health Study (WIHS)[1] and the Multicenter AIDS Cohort Study (MACS)[2] and is reported in units of 10^{-6}×N^{-1}m^2.
- ***Carotid Atherosclerosis*** was defined as common carotid artery mean-max intima-media thickness (CCA-IMT) >1mm or focal area of plaque >1.5mm or >50% of adjacent vessel in any extra-cranial carotid artery
- Brachial artery endothelial function [***flow-mediated dilation (FMD) and hyperemic velocity-time integral (VTI)***] were measured using standard ACTG protocols as previously described[3].

Coronary Artery Calcium (CAC)

- Measured by ECG gated non-contrast CT scan of the chest (64-slice Simens Somatom Sensation 64) and was calculated using the Agatston method.

Inflammation and Immune Activation:

- Percent CD4+ and CD8+ T-cell activation (CD38, HLA-DR, and PD1 expression)
- Inflammatory (CD14+CD16+) and patrolling (CD14^{dim}CD16+) monocytes.
- Plasma markers of monocyte activation: Soluble CD163 (sCD163) and CD14 (sCD14),
- Plasma biomarkers of inflammation and endothelial activation

Statistics:

- Baseline characteristics were described for CAC=0 and CAC>0 and tested for difference using Wilcoxon signed rank tests or chi-square tests, as appropriate.
- **Spearman correlations** were used to examine the association of biomarkers with carotid stiffness among CAC=0 and CAC>0 groups separately

RESULTS

	Overall n = 147	CAC=0 n= 93	CAC >0 N=54	p value
Demographics				
Age (years)	46 (40-53)	44 (35-49)	50 (44-56)	**<0.0001**
Male	115 (78%)	71 (76%)	44 (81%)	*
African American	101 (69%)	71 (76%)	30 (56%)	**0.04**
HIV Parameters				
Current CD4+ count (cells/µl)	613 (425-853)	647 (438-878)	513 (420-754)	**0.04**
Nadir CD4+ count (cells/µl)	179 (86-298)	197 (105-327)	133 (40-274)	**0.04**
HIV duration (years)	12 (6.2-18)	11 (6.3-17)	13(5.8-19)	*
ART duration (years)	5.3 (3.2-9.8)	5.3 (3.3-9.3)	5.8 (3.2-11)	*
PI duration (years)	3.5 (0.7-7.1)	3.3 (0.8-6.6)	4.3 (0.6-7.4)	*
Undetectable viral load (<48copies/ml)	104 (70%)	68 (73%)	36 (67%)	*
Traditional Cardiovascular Risk Factors				
Systolic Blood Pressure (mmHg)	121 (112-132)	121 (110-132)	122 (114-132)	*
HDL Cholesterol (mg/dL)	46 (37-57)	47 (38-57)	44 (37-57)	*
LDL Cholesterol (mg/dL)	97 (77-113)	92 (74-107)	106 (87-121)	**0.02**
Non-HDL Cholesterol (mg/dL)	123 (102-144)	119 (99-144)	129 (107-148)	*
Creatinine Clearance (ml/min)	112 (86-140)	116 (88-148)	106 (84-128)	*
HOMA-IR	1.8 (1.1-3.3)	1.9 (1.1-3.2)	1.6 (1.0-3.4)	*
Metabolic Syndrome	114 (22%)	74 (20%)	40 (26%)	*
Current smoker	93 (63%)	59 (63%)	34 (63%)	*
Family History of MI	46 (31%)	26 (28%)	20 (37%)	*
10-year Framingham risk score (%)	3.0 (1.0-7.0)	2.0 (1.0-6.0)	6.0 (3.0-10)	**<0.0001**
Reynold's Risk Score	2.1 (0.9-5.1)	1.6 (0.6-3.1)	4.2 (1.6-6.5)	**<0.0001**
Body Mass Index (kg/m^2)	27 (23-30)	27 (24-30)	26 (23-30)	*
Current Medication Use				
Anti-hypertensive medication use	36 (24%)	18 (19%)	18 (33%)	**0.06**
Current PI use	72 (49%)	46 (49%)	26 (48%)	*
Current AZT or D4T use	8 (5%)	4 (4%)	4 (7%)	*
Current abacavir use	7 (5%)	4 (4%)	3 (6%)	*
Ultrasound measures of vascular disease				
Carotid distensibility (10^{-6}×N^{-1}m^2)	24 (19-32)	26 (19-33)	23 (19-26)	**0.05**
Carotid atherosclerosis (CCA-IMT >1mm or focal plaque)	63 (43%)	29 (31%)	34 (63%)	**<0.0001**
Brachial flow mediated dilation (FMD, %))	4.0 (2.0-6.1)	4.2 (2.3-6.5)	3.1 (1.6-5.3)	**0.05**
Brachial hyperemic velocity (VTI, cm)	77 (64-95)	79 (66-94)	75 (62-95)	*

Table 1 (left): Baseline characteristics of study participants by CAC score described as median (IQR) or number (%). 13 subjects (9% of total population) without any CAC or carotid atherosclerosis had carotid distensibility below the 25th percentile. * p >0.1.

Table 2 (below): Carotid distensibility was negatively correlated with CD4+ T-cell activation, patrolling monocytes, IL-6, carotid IMT, and endothelial dysfunction among participants without any evidence of coronary calcium. * p >0.1. **BOLD p <0.05**.

	Spearman Correlation Coefficient	
	CAC=0	CAC>0
Inflammation and Immune Activation		
CD8 38+DR+ T-cells (%)	*	*
CD4 38+DR+ T-cells (%)	**-0.342 (p=0.001)**	*
CD8 PD1+38+DR+ T-cells (%)	*	*
CD4 PD1+38+DR+ T-cells (%)	**-0.300 (p=0.004)**	*
CD14+CD16+ monocytes (%)	*	*
CD14dimCD16+ monocytes (%)	**-0.269 (p=0.011)**	*
Soluble CD14 (ng/ml)	-0.172 (p=0.099)	*
Soluble CD163 (ng/ml)	*	*
Soluble Vascular Cell Adhesion Molecule-1 (ng/ml)	*	**-0.350 (p=0.010)**
Interleukin-6 (pg/ml)	**-0.220 (p=0.034)**	*
Tumor Necrosis Factor-α Receptor-1 (pg/ml)	*	*
High-sensitivity C-reactive Protein (µg/ml)	*	*
Additional ultrasound measures of vascular disease		
Mean-max CCA-IMT (mm)	**-0.339 (p<0.001)**	**-0.386 (p=0.004)**
FMD (%)	0.192 (p=0.066)	*
Hyperemic VTI (cm)	**0.374 (p<0.001)**	*

DISCUSSION

- Coronary artery calcium (CAC) scoring significantly improves cardiovascular risk stratification in the general population. In MESA[4], the area under the receiver operator curve for incident coronary heart disease (CHD) was improved from 0.623 (Framingham risk only) to 0.784 with addition of CAC and net reclassification improvement was 0.659. Still, CAC is a late finding of atherosclerosis, and there is a need to identify vascular disease earlier, especially in the HIV-infected population.
- HIV-infected patients may have higher prevalence of non-calcified vascular disease (including carotid plaque[5] and non-calcified coronary plaque[6]) compared to uninfected controls.
- This study suggests that carotid stiffness is an important marker of vascular disease in HIV-infected patients without detectable CAC.
- Furthermore, CD4+ T-cell activation and monocyte activation may play important roles in early vascular disease prior to the development of calcific atherosclerosis.

CONCLUSION: Carotid distensibility, a marker of vascular stiffness, correlates with lymphocyte and monocyte activation in HIV infected patients on ART who do not have evidence of coronary calcification on CT.

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