Background and Purpose—The purpose of this study was to determine if postmortem intracranial arteries from donors with HIV without stroke have thinner media layers compared with patients without HIV and without stroke.

Methods—Cross-sectional cuts from intracranial arteries were stained with van Gieson and hematoxylin and eosin. Arteries were examined for thickness of each arterial layer. Univariable and multivariable models were used for statistical analyses with probability values $<0.05$ considered significant.

Results—A total of 18 brains were analyzed, 5 with HIV and 13 without. Fifty-five arteries were collected, 15 from HIV-infected brains and 40 from unaffected controls. In univariable analysis, in arteries from HIV-infected brains, the media to wall thickness ratio was smaller than in donors without HIV (0.496 versus 0.563, $P=0.017$). In multivariable analysis, HIV infection was the only independent predictor of smaller media ratios compared with the same-aged control subjects ($P=0.049$) but not with aged control subjects ($P=0.081$).

Conclusions—In patients with HIV without clinical stroke, the media arterial layer is thinner than in patients without HIV. This suggests that a thinner media layer might be a preclinical stage in the development of HIV-related vasculopathy.

Key Words: HIV ■ vascular remodeling ■ vasculopathy ■ vascular remodeling

HIV vasculopathy is characterized by multifocal fusiform aneurysms affecting all ages. Pathological studies on arteries affected by HIV vasculopathy have demonstrated significant arterial media (ie, muscularis) layer disruption characterized by atrophy, fibrosis, and fragmentation of the internal elastic lamina (IEL). The HIV infection of the smooth muscle cells may underlie the subsequent deterioration of the media. Little is known about arterial wall changes that could precede the development of clinical cerebrovascular events, particularly those related to HIV vasculopathy.

We hypothesized that brain arteries of subjects with HIV without clinical or pathological presence of stroke have a thinner media layer compared with noninfected control subjects.

Methods

Postmortem tissues were obtained from the University of Miami Brain Endowment Bank from brains without clinical or pathological evidence of stroke. The clinical data were obtained from the donor registry and medical records. HIV status was evaluated by enzyme-linked immunosorbent assay. Consents were obtained from our live donors before death and from the next of kin after death.

The arteries were embedded in paraffin and thin cross-sections were stained. Hematoxylin and eosin stains were used to determine the presence of adventitial lymphocytes. The van Gieson stains were read to score IEL disruption, IEL duplication, intimal hyperplasia, and the measurements of the arterial wall (Figures 1 and 2). The media to wall thickness ratio (MWR) was calculated to account for normal variation in the arterial dimensions depending on anatomic location. The reproducibility measures were obtained from each rater blinded to the HIV status (J.G. and M.G.). Uneven thickness of the wall was captured as cross-sectional bias (Figure 1) and when present, we looked for the more uniform part of the vessel for measurements (Figure 2).

Statistical Analysis

Chi-squared tests were used for categories and 1-way analysis of variance for continuous variables. Statistically significant variables in univariate comparisons were used for a multilevel multivariate MIXED model. Reproducibility was measured with $\kappa$ value and intraclass correlation coefficient. The analysis was carried out using SAS software, Version 9.2 (Cary, NC).

Results

We analyzed arteries obtained from 5 HIV-affected individuals and 13 unaffected control subjects (Table). The group with HIV was younger than the non-HIV group (44.7±4.8 versus 58.9±24.4 years, respectively; $P=0.031$) and included more blacks than whites (80% versus 15.3%; $P=0.022$). The
groups were similar by gender, prevalence of diabetes mel-litus, hypertension, dyslipidemia, atherosclerosis, drug abuse, body weight, height, and brain weight. The proportion of posterior versus anterior arteries was similar in both groups (50% versus 66%; \( P = 0.269 \)). Both groups (HIV versus non-HIV) had similar prevalence of adventitial lymphocytes, IEL disruption, IEL duplication, and cross-sectional bias. HIV was associated with intima hyperplasia (OR, 4.64; 95% CI, 1.16–18.82; \( P = 0.033 \)), thicker adventitia (93.7 versus 72.5 \( \mu \text{m}; P = 0.009 \)), and smaller MWR (0.496 versus 0.563; \( P = 0.017 \)). A smaller MWR was also observed in men (\( P = 0.036 \)), in arteries from the anterior circulation (\( P = 0.055 \)) and in the absence of hypertension (\( P = 0.026 \)) or dyslipidemia (\( P = 0.010 \)). Advanced age was associated with larger media ratios (\( \beta = 0.001; P = 0.027 \)), whereas smaller brain weights were associated with smaller media ratios (\( \beta = -0.0001; P = 0.049 \)).

Gender, age, HIV status, dyslipidemia, hypertension, artery location, and brain weight were used for multivariate analysis. In the complete sample, brain weight and HIV status showed a trend for smaller MWR (\( P = 0.068 \) and \( P = 0.081 \), respectively). Among cases <50 years, HIV became the only independent predictor of smaller MWR (\( P = 0.0495 \), but the MWRs were similar comparing the HIV group with aged control subjects (\( P = 0.366 \)). The intra- and interreader intra-class correlation coefficient was >0.8 for all the arterial measurements. The intrareader \( k \) values were >0.8 for all categorical variables except for intimal hyperplasia (0.64). The interreader agreement was >0.8 for IEL disruption and between 0.6 and 0.8 for the rest of the studied variables.

**Discussion**

In our sample, patients with HIV infection have a thinner MWR than unaffected control subjects within the same age group but not compared with older control subjects. Because none of the affected cases had a stroke, this suggests that the thinning of the media could be a preclinical stage in the occurrence of clinical cerebrovascular disease in patients with HIV. A thinner media has been reported in entities associated with arterial dilatation in patients with and without HIV.1,2,6,7

The lymphocytes in the adventitia were seen in equal proportions among groups, making vasculitis a less likely explanation to our results. Disruption of the IEL has been reported in HIV vasculopathy,1,2,4 but we failed to reproduce this finding. Some authors suggest that IEL disruption is usually encountered in flow-mediated dilation.8 The smooth muscle cells that form the media layer can be directly infected with HIV with progressive damage of the media independent of classical atherosclerosis and inflammation,5 although some endothelial role in the expression of metalloproteinases has also been suggested.9

Lack of information about the administration of antiretro-viral therapy, duration of the HIV infection, degree of immunosuppression, nutritional status, and viral coinfection is a major limitation. The lack of extracranial arteries precludes further inferences. The HIV brains are hospital-based, which limits the generalizability of the findings.
The results suggest that in patients with HIV without clinical stroke, there is thinning of the media arterial layer. Thinning of the media could be a preclinical stage in the development of HIV vasculopathy.

Sources of Funding
This study was funded with an internal grant from the University of Miami/Jackson Health System Neurology Residency Program (Resident–Clinician Researcher Track). The Brain Endowment Bank is funded by a gift from the Frances and Morris McGowan Trust. The brain specimens were provided from a biospecimen resource funded by PHS grant DA06227.

Disclosures
None.

References
Thinning of the Arterial Media Layer as a Possible Preclinical Stage in HIV Vasculopathy: A Pilot Study
Jose Gutierrez, Melanie Glenn, Richard S. Isaacson, Angelica Dawn Marr, Deborah Mash and Carol Petito

*Stroke*. 2012;43:1156-1158; originally published online December 22, 2011;
doi: 10.1161/STROKEAHA.111.643387

*Stroke* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2011 American Heart Association, Inc. All rights reserved.
Print ISSN: 0039-2499. Online ISSN: 1524-4628

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://stroke.ahajournals.org/content/43/4/1156

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in *Stroke* can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to *Stroke* is online at:
http://stroke.ahajournals.org//subscriptions/