Brain aging and cardiovascular factors in HIV: a longitudinal volume and shape MRI study

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Objective: We aimed to examine the relative contributions of HIV infection, age, and cardiovascular risk factors to subcortical brain atrophy in people with HIV (PWH).

Design: Longitudinal observational study.

Methods: Virally suppressed PWH with low neuropsychological confounds ($n = 75$) and demographically matched HIV-negative controls ($n = 31$) completed baseline and 18-month follow-up MRI scans, neuropsychological evaluation, cardiovascular assessments, and HIV laboratory tests. PWH were evaluated for HIV-associated neurocognitive disorder (HAND). Subcortical volumes were extracted with Freesurfer after removal of white matter hyperintensities. Volumetric and shape analyses were conducted using linear mixed-effect models incorporating interactions between age, time, and each of HIV status, HAND status, HIV disease factors, and cardiovascular markers.

Results: Across baseline and follow-up PWH had smaller volumes of most subcortical structures compared with HIV-negative participants. In addition, over time older PWH had a more rapid decline in caudate volumes ($P = 0.041$), predominantly in the more severe HAND subgroups ($P = 0.042$). Higher $CD4^+$ cell counts had a protective effect over time on subcortical structures for older participants with HIV. Increased cardiovascular risk factors were associated with smaller volumes across baseline and follow-up for most structures, although a more rapid decline over time was observed for striatal volumes. There were no significant shape analyses findings.

Conclusion: The study demonstrates a three-hit model of general (as opposed to localized) subcortical injury in PWH: HIV infection associated with smaller volumes of most subcortical structures, HIV infection and aging synergy in the striatum, and cardiovascular-related injury linked to early and more rapid striatal atrophy.

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Introduction

Whether there is altered brain aging in HIV disease is unclear although there are grounds for suspicion. First, the brain may be more vulnerable through loss of reserve related to HIV-associated neurocognitive disorder (HAND) in the past or ongoing damage from current HIV despite viral suppression [1,2]. Second, elevated cardiovascular risk factors [3,4] and stroke [5] are more common in HIV infection and are associated with aging and brain atrophy [6,7]. Recent work has used indirect measures of cardiovascular disease in the form of white matter hyperintensities as a proxy measure of cardiovascular disease and found no evidence of accelerated aging

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in HIV [8]. Nevertheless, direct assessment of cardiovascular risk on brain atrophy in HIV is required.

In longitudinal studies normal aging may interact with confounds to enhance atrophy in several ways. Premature atrophy can occur with a reduction in regional brain volume in older participants at study entry but without any further change over the study period. This is indicated by a significant two-way interaction effect of HIV and age, where older people with HIV (PWH) have smaller volumes than older HIV-negative adults. Alternatively, accelerated aging can occur, defined as atrophy occurring at a faster rate for older participants across the study period, and is demonstrated by a significant three-way interaction of HIV status, age, and time.

The magnitude of age and HIV effects on brain changes in past studies depend on the methods used to quantify atrophy. Premature aging has been demonstrated in both region of interest (ROI) and voxel-based mapping (VBM) studies [9–11], whilst accelerated aging has been shown only in ROI-based studies [12,13]. In addition, shape analysis can localize regional atrophy of subcortical structures which may be insensitive to volumetric methods [14]. However, shape analysis in PWH is limited to samples with low proportions of antiretroviral treatment [15], poor viral control [16], or younger age [17]. Although a cross-sectional shape study has suggested an accelerated aging effect in younger PWH [18], there is a need for longitudinal shape studies in older age groups.

The primary aim of the current study was to quantify longitudinal subcortical volumetric and shape changes in virally suppressed, clinically stable PWH aged over 45 years relative to age-matched HIV-negative controls. The secondary aim was to determine the contributions of aging, HIV status, HAND clinical categories, neuropsychological performance, HIV disease factors, cardiovascular risk factors and their relevant interactions to any longitudinal subcortical volumetric, and shape changes. We hypothesized that premature and accelerated aging would be evident on all subcortical structures in PWH compared with the HIV-negative controls. Further, we hypothesized that older PWH with more severe HAND and greater cardiovascular risk factors would show both volume and shape alterations.

Methods

Participants

Eighty-seven HIV-positive and 39 HIV-negative individuals completed the baseline neuropsychological, MRI, and clinical visit. Ofthose, 75 HIV-positive and 31 HIVnegative participants returned for a follow-up session. Participants were classified as having incidental, contributing, or confounding comorbidities as in the CHARTER study and those with confounding comorbidities were excluded prior to analysis [19]. Description of the baseline cohort characteristics in our HIV and Brain Aging Research Program (including criteria of inclusion/exclusion and comorbidities/treatment data) are published previously [20,21].

The St. Vincent's Hospital, Sydney and The University of New South Wales Human Research Ethics Committees approved our protocol (08/SVH/90). All individuals provided written informed consent before study participation.

MRI, neuropsychological, and clinical assessment

MRI images were acquired at St. Vincent's Hospital, Sydney Medical Imaging Department on a Phillips 3T Achieva TX scanner (Philips Medical Systems, Best, The Netherlands) using an eight-channel head coil. T1-weighted anatomical images were obtained in the coronal plane (TFE: TR/TE: 6.39/2.9 ms, flip angle: 88; FOV 256 mm; 190 slices, 1 mm isotropic). T2 FLAIR images were also acquired in the axial plane (TR/TE: 11 000/110 ms, TI: 2800 ms, flip angle: 908; FOV 250 mm, 38 slices, $512 \times 512 \times 3.5$ mm).

A standard neuropsychological battery for this cohort has been reported previously [20,22]. For this study, we included the HAND clinical categories of asymptomatic neurocognitive impairment (ANI), mild neurocognitive disorder (MND) and HIV-associated dementia (HAD) were classified using Global Deficit Score and functional data. We also included the cognitive domains scores grouped into seven ability domains: mental flexibility, verbal generativity verbal learning verbal delayed memory, attention/working memory, speed of information processing, and motor coordination as in prior work [20,22].

Details of the laboratory visit were also reported in Cysique *et al.* [20] and are reproduced in Supplemental Digital Content 1, http://links.lww.com/QAD/C439. We used standard laboratory markers of HIV in the field and obtained cardiovascular markers to construct risk scores. The 2008 Framingham score [23] of 10-year cardiovascular disease risk was computed for all participants. In the PWH group the 12-month Data Collection on Adverse events of Anti-HIV Drugs (DAD) score was computed to estimate cardiovascular disease risk in the HIV+ group [24]. This score estimates cardiovascular disease outcomes more accurately than the Framingham score in PWH [25].

MRI processing and shape analysis

All images were reviewed by an expert neuroradiologist for extensive abnormalities prior to inclusion in the study. No scans had any such abnormalities. MRI images were processed in Freesurfer 6.0 to generate subcortical

segmentations and were reviewed for accuracy by a trained student (blind to HIV status) and the senior author who has extensive experience in Freesurfer. T2 FLAIR sequences were used to refine the pial surface. The '-big ventricles' flag was used to account for dilated lateral ventricles where default processing failed ($n = 3$). In addition, T2 FLAIR white matter hyperintensities were determined using a fully convoluted neural network, which was the top-ranking method in a recent multi-scanner challenge [26] and manually checked for accuracy. These white matter hyperintensities were subtracted from the Freesurfer subcortical volumes as they are known to artificially inflate volumes [27]. Adjusted volumes were used for all subsequent analyses.

Subcortical segmentations were processed with SPHARM-PDM [14]. Images were minimally processed to fill internal holes and smoothed to ensure a spherical topology, and shapes consisting of 1002 corresponding vertices were composed. For each subcortical structure, a study-specific mean shape was created, and all shapes were aligned to the sample mean using Procrustes alignment.

Statistical analyses

All analyses were performed in R 3.6.2 [28]. Linear mixed models were utilized to account for unbalanced longitudinal data [29] with random intercepts and random slopes. Baseline age was mean centered to facilitate coefficient interpretation [30]. For all analyses, estimated total intracranial volume was considered as a covariate, centered baseline age a fixed variable, and time (baseline and follow-up point) a fixed factor. Additional fixed variables included, in separate models, HIV status, HAND category (ANI, MND, HAD); HIV disease factors (HIV duration, history of AIDS, nadir $CD4^+$ cell count, $CD4^+$ and $CD8^+$ cell count, $CD4^+/CD8^+$ ratio); cognitive domain (mental flexibility, verbal generativity, verbal learning, verbal delayed memory, attention/working memory, speed of information processing, and motor coordination); Framingham score and DAD score. Dependent variables were the mean volume of the bilateral caudate, putamen, pallidum, hippocampus, and thalamus.

Model design was dependent on the independent variables of interest. For models investigating HIV status, HAND category, and HIV disease factors, we included all main effects and both two-way and three-way interactions between baseline age, time, and the variable of interest (i.e. HIV status, HAND category, and HIV disease factors). For cardiovascular measures, models included only two-way interactions between time and the variables of interest (i.e. Framingham and DAD score). Age was not included as we expected multi-collinearity to arise since age is included in the Framingham and DAD equations. To assess for differences in cognitive function between HIV status accounting for volumetric differences, we modeled the effect on subcortical volumes controlling for age and time and examined the main effect of cognitive domains and the interaction between cognitive domain and HIV status.

Shape analysis was also conducted for the caudate, putamen, pallidum, hippocampus, and thalamus. For each vertex in each structure in each hemisphere, the magnitude of displacement along the surface normal (i.e. perpendicular to the surface) between the study mean shape and each participant's shape was calculated. This displacement value was used as the dependent variable in mixed models at each vertex. Negative displacement indicates inward movement, or deflation, compared with an average shape, whereas positive displacement indicates outward movement, or inflation. Mixed models included the same parameters as for volume analysis. Two-stage false discovery rate correction [31] was applied across each surface.

Results

Participant's demographic, and clinical characteristics are presented in Table 1 and HIV disease characteristics are presented in Table 2. The study samples were comparable for all demographics, although with a trend for older participants in the PWH group. Twelve PWH did not return for follow-up, due to claustrophobia (two) or unspecified reasons (10). Eight HIV-negative participants did not have follow-up due to declining a follow-up scan (four), claustrophobia (two), were unavailable (one), or had a new MRI contraindication (one). There were no significant differences in demographics or HIV disease factors between participants who did and did not attend follow-up.

HIV status

Model terms including all main effects and two-way and three-way interactions for age, time, and HIV status are displayed in Table 3. HIV status was associated with smaller mean volumes of the pallidum, hippocampus, and thalamus across the study period. In addition, for the mean caudate volume, there was significant HIV status by time by age interaction (Fig. 1), whereby older PWH had a greater decrease in caudate volume between baseline and follow-up than either younger PWH or the HIVnegative group.

HIV-associated neurocognitive disorder status and cognitive domains

Using the HIV-negative sample as a reference against the HAND clinical categories in the PWH sample (i.e. neuropsychologically normal, ANI, and MND + HAD) there was a significant main effect reduction in the pallidum and hippocampal volumes for the combined MND + HAD group, and reduction in the thalamic volumes for all PWH subgroups (see Supplemental Table

Table 1. Demographic, clinical, and test–retest information at baseline and follow-up.

	HIV−		HIV+		*P*
Baseline					
n	39		87		
	M	SD	M	SD	
Age (years)	53.3	6.7	55.5	7.1	0.138
Age range	45–67		45–74		
Education (years)	15.0	2.7	14.2	2.6	0.172
Predicted VIQ	111.3	6.1	109.1	7.0	0.064
Framingham high risk (%)	18		49		<0.001
DAD high risk (%)			13		
Cardiovascular event (%)	0		13		
Cardiovascular medication (%)	21		32		
Follow-up					
n	31		75		
	M	SD	M	SD	
Age (years)	53.5	6.6	55.1	7.0	0.270
Age range	45–67		45 –74		
Education (years)	14.7	2.6	14.4	2.6	0.582
Predicted VIQ	110.7	5.6	109.5	6.5	0.378
Test–retest interval (months)	24.0	3.7	22.8	3.1	0.102
Test–retest interval range	18.0–30.3		19.3–32.5		

Cardiovascular events had been treated and included atrial fibrillation, myocardial infarction, congestive heart failure, peripheral arteriosclerosis, and carotid/coronary arteriosclerosis. Cardiovascular medications included aspirin, anticoagulants, blood pressure, lipid lowering, and glucose-lowering medications. DAD, Data Collection on Adverse events of Anti-HIV Drugs cardiovascular risk score;VIQ, WAIS-III estimated verbal IQ.

Table 2. Disease characteristics of the HIV+ sample at baseline and follow-up.

Characteristic	Median/%	Range
Baseline		
Median HIV duration (years)	20	(4–31)
Median current cART duration (months)	24	(6–156)
Median nadir $CD4^+$ cell count (cells/ml)	180	(0–490)
Median $CD4^+$ T-cell count (cells/ml)	528	(77–1476)
Median $CD8^+$ T-cell count (cells/ml)	816	(51–2132)
Median $CD4^+/CD8^+$ ratio	1.73	(0.16–8.00)
Historical AIDS	71%	–
Plasma HIV RNA undetectable (<50 copies/ml)	98%	–
CSF HIV RNA undetectable (<50 copies/ml)	100%	–
NP-normal	47%	–
ANI	38%	–
MND	13%	–
HAD	2%	
Follow-up		
Median $CD4^+$ T-cell count (cells/ml)	643	(136–1936)
Median $CD8^+$ T-cell count (cells/ml)	857	(352–2352)
Median $CD4^+/CD8^+$ ratio	1.38	(0.45–4.20)
Plasma HIV RNA undetectable across study	89%	–
Change in cART during study period	14%	–

Thirty-five participants consented to lumbar puncture for cerebrospinal fluid (CSF) analysis. ANI, asymptomatic neurocognitive impairment; cART, combined antiretroviral therapy;HAD, HIV-associated dementia; MND, mild neurocognitive disorder;NP-normal, neuropsychologically normal.

S1 for coefficients, http://links.lww.com/QAD/C439). Significant three-way interaction with age and time was observed for the mean caudate volume for the ANI group (Beta = −8.4, SE = 3.5, $P = 0.019$) and the combined MND + HAD group (Beta = −8.2, SE = 4.0, $P = 0.042$). This suggests that older participants with ANI or MND + HAD had a larger decline in caudate volume over time than younger participants.

Table 3. Main and interaction effects for age, time, and HIV status on mean subcortical volumes (mm^3).

Structure	Effect	Beta	SE	*P*
Caudate				
	Intercept	1489	314	<0.001
	Age	−14	9	0.128
	Time	−22	16	0.165
	HIV	−66	74	0.381
	Age∗Time	0	2	0.923
	Age∗HIV	−1	11	0.902
	Time∗HIV	−32	19	0.097
	Age∗Time∗HIV	**−6**	**3**	**0.041**
Putamen				
	Intercept	2944	428	<0.001
	Age	**−26**	**13**	**0.047**
	Time	**−52**	**22**	**0.020**
	HIV	−128	102	0.215
	Age∗Time	−3	3	0.359
	Age∗HIV	−3	15	0.823
	Time∗HIV	−23	26	0.382
	Age∗Time∗HIV	−4	4	0.333
Pallidum				
	Intercept	835	165	<0.001
	Age	5	5	0.292
	Time	13	12	0.273
	HIV	**−102**	**40**	**0.011**
	Age∗Time	2	2	0.388
	Age∗HIV	−11	6	0.059
	Time∗HIV	4	14	0.783
	Age∗Time∗HIV	2	2	0.357
Hippocampus				
	Intercept	2624	295	<0.001
	Age	**−20**	**9**	**0.026**
	Time	−11	19	0.555
	HIV	**−172**	**71**	**0.017**
	Age∗Time	−1	3	0.654
	Age∗HIV	−6	10	0.588
	Time∗HIV	−3	22	0.903
	Age∗Time∗HIV	−2	3	0.579
Thalamus				
	Intercept	4027	496	<0.001
	Age	−18	16	0.246
	Time	−40	32	0.212
	HIV	**−419**	**123**	**0.001**
	Age∗Time	−7	5	0.175
	Age∗HIV	−19	18	0.291
	Time∗HIV	1	38	0.969
	Age∗Time∗HIV	−3	6	0.564

Age is centered to the mean value (55 years). Effect for estimated total intracranial volume is included in models but not displayed. Values in bold are statistically significant ($P< 0.05$).

For cognitive domains, there were no significant main effects for cognitive tasks on brain structural volumes, controlling for age, time, and HIV status. Only for the motor coordination domain was there a significant interaction between HIV status for caudate volumes (Beta = 57, SE = 29, $P = 0.046$). This indicates that in PWH higher scores on the motor coordination task were associated with higher caudate volumes. There were no other significant interactions. There were no significant shape effects for HAND category or cognitive domains.

HIV disease factors

There was no significant correlation between HIV duration and age ($r = 0.15$, $P = 0.17$). Thus, age and HIV duration were included separately as analysis terms. There was a significant three-way interaction between age, time, and HIV duration for mean caudate (Beta = −0.4, SE = 0.2, $P = 0.044$) and putamen (Beta = −0.7, SE = 0.3, $P = 0.017$) volumes. This indicates that there was a more rapid decline in mean caudate and putamen volumes for older PWH with a longer duration of HIV compared with younger PWH with shorter HIV duration.

There were significant positive three-way interactions between baseline $CD4^+$ cell counts, age, and time for the caudate, putamen, and thalamus volumes ($P < 0.045$; Supplemental Table S2, http://links.lww.com/QAD/C439) and approached significance for hippocampal volume ($P = 0.066$). This suggests that older PWH with lower baseline $CD4^+$ cell counts had faster atrophy in these structures between baseline and follow-up.

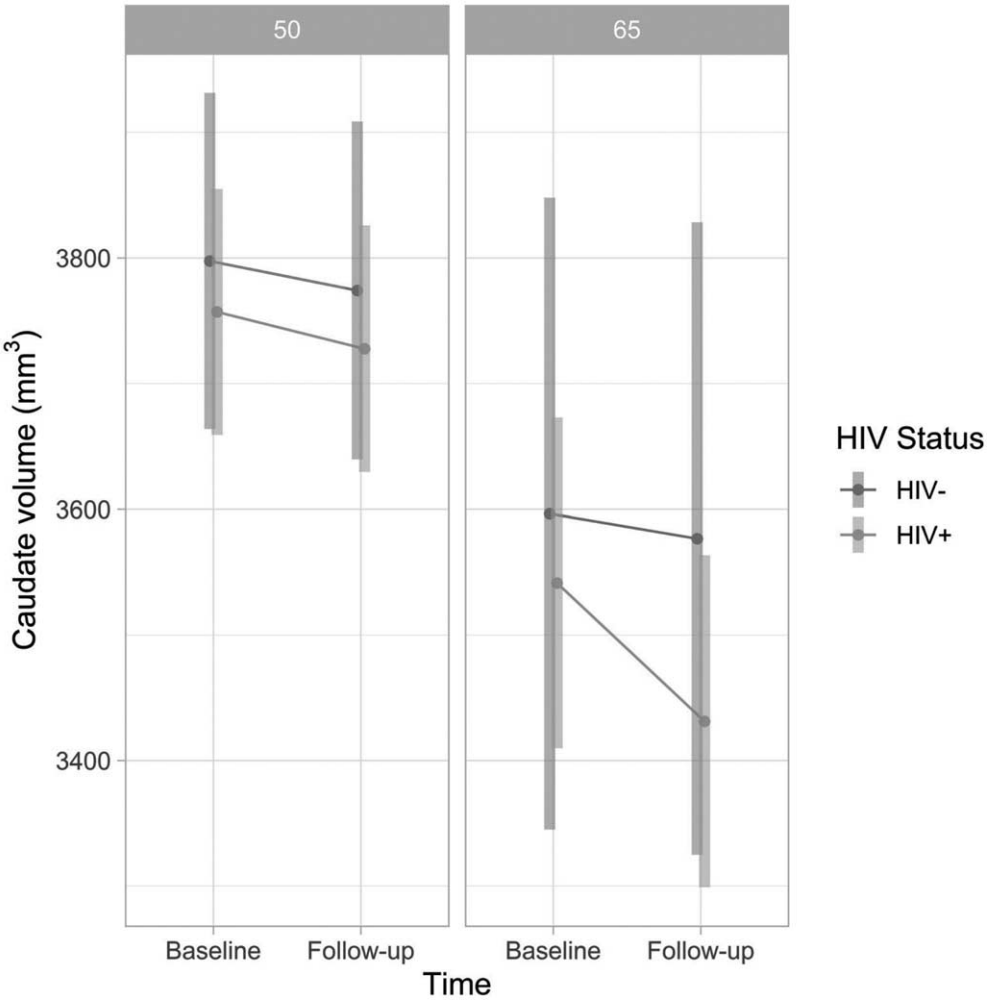

Fig. 1. HIV status by age by time effects in the caudate. Estimated marginal means with 95% confidence intervals shown. Age is dichotomized for representation purposes; in analyses it was included as a continuous variable.

Additional main effects were observed for reductions in mean striatal volumes with lower $CD8^+$ cell counts and $CD4^+/CD8^+$ ratios, and reduced mean caudate volumes with lower nadir $CD4^+$ cell counts and a history of AIDS. There were no significant shape effects for HIV disease factors.

Cardiovascular measures

Both the Framingham and DAD scores were positively skewed and so were reclassified as dichotomous variables. Following established guidelines participants were classified as having no to low cardiovascular risk, or intermediate-to-high cardiovascular risk (less or greater than 10% risk, respectively) [32]. Using Framingham criteria, 44 PWH had no to low cardiovascular risk, and 43 had an intermediate-to-high risk. Using the more selective DAD criteria, 76 were classified as no to low risk and 11 as intermediate-to-high risk. There were no statistically significant differences in demographic or HIV disease factors between cardiovascular risk groups.

There were multiple main effects observed for both the Framingham score and DAD score on subcortical structural volumes in PWH (see Supplemental Table S3 for all values, http://links.lww.com/QAD/C439). Higher Framingham scores were associated with significantly smaller mean volumes of all structures, although there were no statistically significant interactions over time. Similarly, for PWH with higher DAD scores, smaller caudate, putamen, hippocampal, and thalamic volumes were observed (Fig. 2). More importantly, there was a significant interaction with time with higher DAD scores for the caudate (Beta = −88, SE = 35, $P = 0.016$) and putamen (Beta = −105, SE = 49, $P = 0.036$).

Discussion

In this longitudinal study of a virally suppressed PWH cohort with low neuropsychological confounds, we

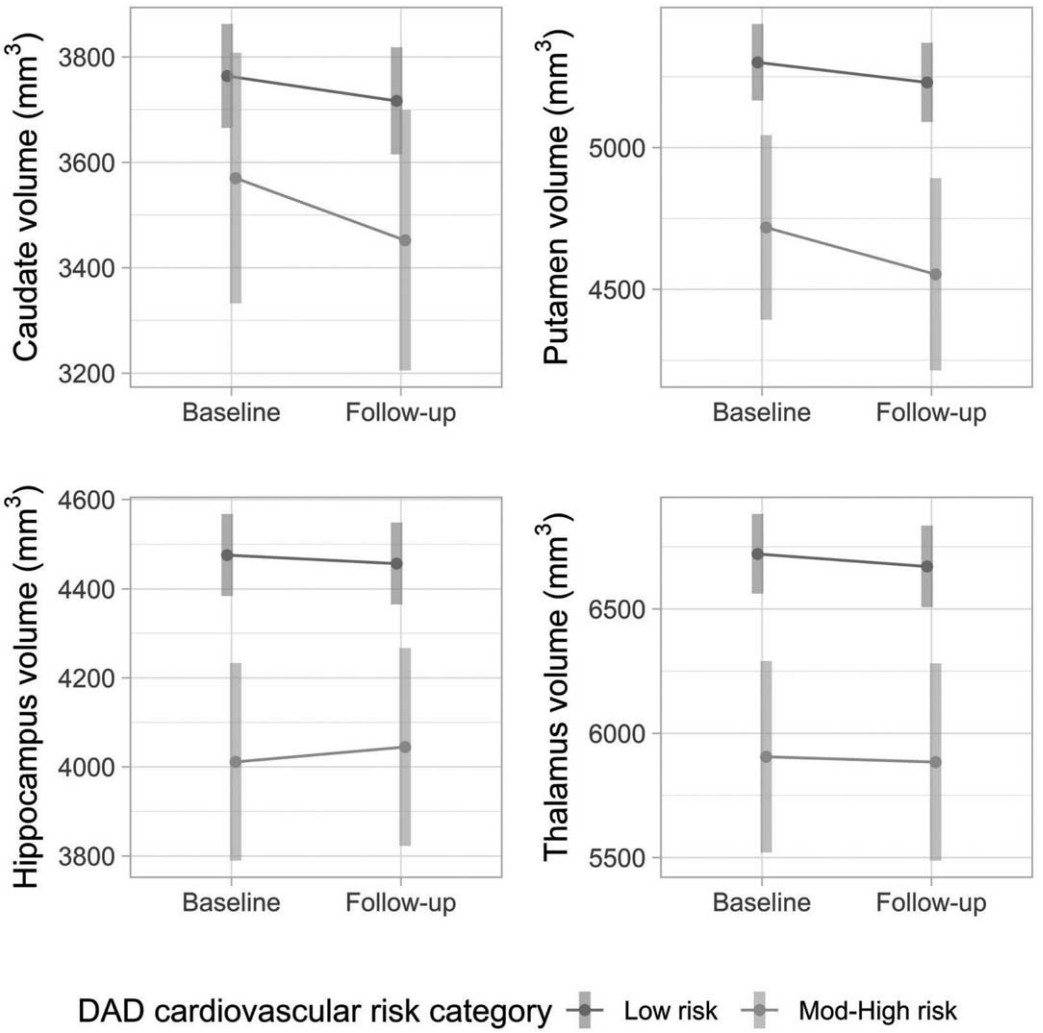

Fig. 2. Data Collection on Adverse events of Anti-HIV Drugs cardiovascular risk score category effects on subcortical volumes. Estimated marginal means with 95% confidence intervals shown. Significant differences between cardiovascular risk categories are present for all structures shown, and additional significant interactions are between cardiovascular risk category and follow-up for the caudate and putamen.

found evidence of a three-hit model of subcortical atrophy: first, HIV-driven atrophy in the hippocampus, thalamus, and pallidum; second, accelerated aging of the caudate in symptomatic HAND and accelerated aging in the striatum in PWH with longer HIV duration and lower $CD4^+$ cell counts; and third, accelerated aging of the striatum in PWH with higher cardiovascular risk scores.

Subcortical volumetric atrophy associated with HIV infection at baseline and follow-up was observed for the pallidum, hippocampus, and thalamus. These differences are more extensive than those reported in the baseline analysis of this dataset [21], probably due to the increase in power from having a longitudinal design and/or due to the adjustment for white matter hyperintensities. There have been multiple studies describing atrophy in almost all subcortical structures in PWH [13,15,16,18]. However, these studies have PWH with a substantial proportion (>20%) with detectable viral loads and participants with greater neuropsychological confounds. In contrast, our study included virally suppressed PWH who had been screened for clinically relevant neuropsychological confounds [20], indicating subcortical atrophy is present and progresses regardless of these confounds.

By contrast, we found no significant shape effects for HIV status or any interactions with age and time, unlike previous work [15,16,18]. The reasons for these differences may be related to our sample being fully virally suppressed, all on combined antiretroviral therapy (cART), and with low neuropsychological confounds. This is distinct from previous cross-sectional shape studies where cART was taken by the majority, but not all,

participants [15,16], or not reported [18], and with 20–30% of the PWH having detectable viral loads [15,16,18]. To the best of our knowledge, the only previous longitudinal study of shape analysis in PWH was in a pediatric cohort which found no longitudinal shape differences by HIV status [17]. The presence ofan overall volumetric effect rather than a shape effect indicates that the entire structure is affected rather than regional areas [33]. This could be associated with our proposed three-hit model leading to a neurotoxic cascade that is deleterious to all part of the neuronal circuitry (efferent and afferent connections) and supportive cells in the subcortical area. There may be a methodological limitation as our methodology only considers shape displacement perpendicular to the surface and other displacements such as stretching, or shearing, are not captured. Alternatively, the disparate findings between volumetric and shape analyses may reflect that atrophy is diffusely distributed over individual structures (reflecting overall volumetric atrophy), and not localized to specific regions as would be expected to be detected by shape analysis.

Accelerated aging was demonstrated in the caudate in conjunction with greater cognitive impairment, suggesting in aging PWH there are parallel axes of pathological insults. Our findings support results from a smaller study [34] and extend it to a larger set of virally controlled PWH with low neuropsychological confounds. In addition, a recent meta-analysis has posited that caudate atrophy may be a biomarker of neurocognitive impairment in HIV [35]. Together, these findings confirm the vulnerability of the caudate to HIV [36,37], show that this effect extends to the modern cART era, and provide a novel finding that aging effects may precipitate damage in this nucleus. The absence of associations between cognitive domains and structural volumes (controlling for age, time point, and HIV status) reflect that we concentrated on subcortical volumes and that most of the cognitive domains tested likely require the involvement of major connections across brain areas rather than subcortical structures per se. Significantly, the association between motor coordination and caudate volumes reflects the known involvement of the caudate in motor planning [38]. Thus, our finding of caudate volumes being positively associated with motor functioning in PWH only (and not HIV-negative) may reflect the structural-cognitive association being enhanced by the pathological effects of HIV on the caudate. Taken together caudate atrophy may therefore serve as a method to distinguish HIV-driven cognitive impairment from other neurodegenerative processes, such as Alzheimer's disease, which can co-occur in older PWH [39].

Accelerated aging was also demonstrated in subcortical structures associated with longer HIV duration and lower baseline $CD4^+$ cell counts. A negative correlation between putamen volume and HIV duration has been demonstrated in a longitudinal ROI study [40], but not in a VBM study [9]. Our previous research in this cohort found that a longer HIV duration was associated with greater neuronal damage via magnetic resonance spectroscopy in the basal ganglia [41], suggesting that the caudate and putamen are vulnerable to combined aging and chronic effects of HIV. We also found that there was a significant positive three-way interaction between $CD4^+$ cell count, age, and time in the caudate, putamen, and thalamus, and approached statistical significance in the hippocampus. The cross-sectional association between subcortical structures and $CD4^+$ cell count has been recently demonstrated in a large meta-analyses [42]. Pathologically, the caudate in particular has been thought to have a high HIV viral load and associated neuropathology due to the proximity to cerebrospinal fluid [37]. Our finding of a significant three-way interaction suggests vulnerability of the striatum to accelerated atrophy in older PWH, which to our knowledge has not been demonstrated previously. The clinical implication is that legacy HIV effects, may contribute to accelerated atrophy in older adults, however maintenance of $CD4^+$ cell count in older PWH may be protective against neurodegeneration of the deep grey matter.

Furthermore, cardiovascular disease is a known complication of HIV infection [4,43]. Literature on brain atrophy due to cardiovascular risk in non-HIV samples has found smaller striatal, hippocampal and thalamic volumes associated with type 2 diabetes [44], and hypertension [45]. Our results also demonstrate atrophy in these volumes in PWH with increased cardiovascular risk factors using the Framingham score, and larger degrees of atrophy using the HIV-specific DAD score. This may represent a metabolic complication of HIV infection or antiretroviral treatment. Since aging itself is a cardiovascular risk factor and is incorporated into both the Framingham and DAD score, longitudinal studies are critical to disentangle the relative contribution of different factors to regional brain atrophy. Our finding of a significant time interaction effect in the caudate and putamen indicates vulnerability of deep subcortical structures to cardiovascular disease above-and-beyond that observed in aging. Moreover, given the observed associations of caudate and putamen accelerated aging in HIV disease, which is also associated with HIV disease risk factors, we posit that striatal structures may be more at risk of synergistic effects of HIV, age, and cardiovascular disease due to metabolic consequences of HIV infection or HIV treatment.

There are limitations to the current study. The effect size of the HIV status on structural volumes is in the order of 5–10% volume reduction, however the interaction effects observed are smaller. Although our study is relatively well powered compared with other longitudinal studies in the field, longer term follow-up studies and meta-analyses are encouraged to ensure stability of the results. Cortical

changes were not considered in this study due to a previous cross-sectional study of this cohort demonstrating minimal cortical changes [21]. Future work is encouraged to consider the synergistic impact of HIV infection and comprehensive assessments of cardiovascular disease on cortical measures. Our sample consisted of well educated males and clinically stable with long-term viral suppressed with relatively few comorbidities. While this is representative of the current epidemic in Australia, we acknowledge that our results may not generalized to other PWH in international locations. Moreover, considering that cardiovascular diseases are actively treated in the HIV population in Australia, our results may underestimate brain atrophy compared with samples with more severe and untreated cardiovascular disease. Further studies examining larger samples is encouraged to replicate and extend our findings.

In conclusion, we have demonstrated subcortical volume atrophy in PWH that is not shape specific. Pathological accelerated aging was observed in the caudate for PWH, potentially driven by symptomatic HAND subgroups. HIV duration and cardiovascular disease are risk factors to basal ganglia atrophy. Conversely, immune function, measured by $CD4^+$ cell counts, has a protective effect in most structures. Further exploration of mediating and moderating effects of HIV disease and its treatment will be important in more diverse samples. More importantly, clinical implications of strict control of immunological and cardiovascular risk factors in healthy brain aging are suggested and will require further studies to evaluate effectiveness in protecting brain volumes.

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All authors contributed to the study design, data interpretation, and provided critical revision of the article. C.D.R., B.J.B., and L.A.C. were involved in data acquisition. D.J. conducted the data and statistical analyses and drafted the article.

Conflicts of interest

The authors declare that they have no conflicts of interest.

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