Neuropathologic Heterogeneity Does Not Impair Florbetapir-Positron Emission Tomography Postmortem Correlates

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Abstract

Neuropathologic heterogeneity is often present among Alzheimer disease (AD) patients. We sought to determine whether amyloid imaging measures of AD are affected by concurrent pathologies. Thirty-eight clinically and pathologically defined AD and 17 nondemented patients with quantitative florbetapir F-18 (18F-AV-45) positron emission tomography (PET) imaging during life and postmortem have been analyzed in 6 cortical regions of interest. All AD subgroups had strong histological β-amyloid quantification and neuropathologic examination were assessed. AD patients were divided on the basis of concurrent pathologies, including those with Lewy bodies (LBs) (n = 21), white matter rarefaction (n = 27), severe cerebral amyloid angiopathy (n = 11), argyrophilic grains (n = 5), and TAR DNA binding protein-43 inclusions (n = 18). Many patients exhibited more than 1 type of concurrent pathology. The ratio of cortical to cerebellar amyloid imaging signal (SUVR) and immunohistochemical β-amyloid load were analyzed in 6 cortical regions of interest. All AD subgroups had strong and significant correlations between SUVR and histological β-amyloid measures (p < 0.001). All AD subgroups had significantly greater amyloid measures versus nondemented patients, and mean amyloid measures did not significantly differ between AD subgroups. When comparing AD cases with and without each pathology, AD cases with LBs had significantly lower SUVR measures versus AD cases without LBs (p = 0.002); there were no other paired comparison differences. These findings indicate that florbetapir-PET imaging is not confounded by neuropathological heterogeneity within AD.


INTRODUCTION

Biomarkers are increasingly regarded as essential tools for drug discovery and for monitoring the effects of therapeutic agents for neurodegenerative diseases. In less than 10 years since the first publication on the topic of positron emission tomography (PET) imaging of brain amyloid with Pittsburgh Compound-B (11C-PiB), imaging with this compound has become an important research tool (1). However, the 20-minute half-life of 11C-PiB restricts its usage to specialized research centers in close proximity to a biomedical cyclotron (1). In contrast, labeling with 18F gives a half-life of 110 minutes (2), and numerous ligands are currently under study utilizing this approach (3–10). In an autopsy study of 35 patients that was later extended to 59 patients, one such 18F-labeled compound, florbetapir F-18 (18F-AV-45), has been...
shown to have high sensitivity and specificity for β-amyloid
(8, 11, 12). Detailed neuropathological data on these patients
have not yet been published.

Considerable neuropathologic heterogeneity is often pres-
ent in persons with AD and other forms of dementia, raising the
question as to whether this might have effects on amyloid im-
aging measures. This may be especially crucial because the
diagnostic clinical diagnosis of autopsy-confirmed AD may be incorrect in
20% to 30% of subjects (13); amyloid imaging is expected to
reduce this inaccuracy. AD is pathologically defined by pla-
quases and tangles, but a large majority of AD cases have a variety
of concurrent pathologies, including Lewy bodies (LBs), vas-
cular lesions, argyrophilic grains (Args), and TAR DNA binding
protein-43 (TDP-43) inclusions (14–19).

The purpose of the present study was to test the hypothe-
sis that florbetapir F-18 PET imaging is a reliable method for esti-
mat-ing in vivo cortical amyloid load in AD subjects re-
gardless of neuropathologic heterogeneity. This autopsy series
is the first to describe the full spectrum of neuropathological
findings in AD patients who had received amyloid imaging
during life. We included AD and nondemented cases from the
previously published reports (8, 11), dividing the pathologi-
cally defined AD subjects into subgroups based on some of the
major concurrent pathologies found within AD. These sub-
groups consisted of those with LBs, white matter rarefaction
(WMR), severe cerebral amyloid angiopathy (CAA), Args, and
phosphorylated TDP-43 inclusions. We examined whether in
vivo amyloid imaging measures of AD patients or their corre-
lates with postmortem histological β-amyloid measures varied
due to the presence or absence of these concurrent pathologies.

MATERIALS AND METHODS

Subject Selection

Subjects were derived from those described in 2 prev-
iou-s publications (8, 11). Details of the recruitment, amyloid
imaging, tissue processing, and analytic methodology are
given in the prior publications. Briefly, patients near the end of
their lives were recruited from hospice, long-term care and
community healthcare facilities for florbetapir-PET scanning.
Fifty-nine patients died within 2 years of amyloid imaging, and
they were autopsied and neuropathologically examined. From
these, 55 subjects were selected for inclusion in the present
study, based on their clinicopathological classification as either
AD or nondemented controls. Subjects with AD (n = 38) were
defined as demented subjects meeting Consortium to Estab-
lish a Registry for Alzheimer’s Disease (CERAD) ‘prob-
able’ or ‘definite’ criteria for AD pathology (20). Control
cases (n = 17, Table 1) were defined as those without a final
clinical diagnosis of dementia (regardless of pathology find-
ings) and included clinically normal nondemented individuals
(n = 12) and those with mild cognitive impairment (MCI, n = 5),
but not demented subjects. Three other subjects were excluded
because they were demented but did not meet neuropatho-
logical criteria for AD; these included 1 with Parkinson dis-
case, 1 with dementia with Lewy bodies (DLB), and 1 with
hippocampal sclerosis dementia. One case was excluded due
to methodological deviation.

Florbetapir-PET Imaging Methods

The details of the imaging methods have been previ-
ously described (8, 11). Briefly, each subject underwent a
10-minute PET scan at 50 minutes after receiving an intra-
venous bolus of 370 MBq (10 mCi) florbetapir F-18. Acquired
PET scans were reconstructed either by iterative treatment
with a postreconstruction Gaussian filter or row action maxi-
mum likelihood algorithms to a 128 × 128 matrix with a zoom
of 2.0 to 2.33. Florbetapir F18 PET images were independently
spatially normalized using statistical parametric mapping to
standard atlas coordinates with reference to a florbetapir PET
template. Standard uptake value ratios (SUVR) were expressed
as the average ratio, compared to the whole cerebellar uptake,
of 6 predefined anatomically relevant cortical regions of in-
terest (ROIs): frontal, parietal, temporal, precuneus, posterior-
cingulate and anterior cingulate cortices.

Histological Amyloid Quantification

Upon autopsy, brains were processed with methods as
previously outlined (8, 11), as well as with standard methods
utilized by the Banner Sun Health Research Institute Brain
and Body Donation Program (21). Brains were fixed whole in
10% neutral-buffered formalin for 2 weeks prior to dissection.
One set of tissue blocks was taken from the same ROIs as
were used for imaging (8). These blocks were embedded in
paraffin and stained using immunohistochemistry (IHC) for
β-amyloid using the anti-β antibody 4G8 (Covance, Emeryville, CA) (8). The cortical amyloid burden on each of
these slides, which included all morphologically defined plaque
subtypes, was defined as the percentage of gray matter occu-
pied by stained neuropil exceeding a threshold stain density
(8, 22), using PERMITS™ image processing and analy-
sis software (Biospective, Inc., Montreal, Quebec, Canada).
Amyloid burden estimates were thus obtained for all 6 cortical
ROIs, and a mean cortical amyloid load was determined by
averaging these. Additionally, another set of large (3 × 5 cm)
tissue blocks, from standard levels of the frontal (superior half
of frontal lobe at the coronal level of the genu of the corpus
callosum), parietal (superior half of parietal lobe at the coronal
level of the splenium of the corpus callosum), and temporal
lobes (coronal levels of the amygdala and body of the hippo-
campus), were cryoprotected in ethylene glycol and sectioned
at 80-μm thickness on a sliding freezing microtome (21).
These sections were stained using the Campbell-Switzer stain,
an enhanced, amyloid-selective silver technique, together with
the Thioflavin S stain; semiquantitative estimates of regional
and average (means from all 3 regions) cortical amyloid bur-
den were obtained from these sections. Scores for plaque den-
sity were derived by considering all types of plaques (cored,
necurit, and diffuse) together to obtain a “total” plaque score,
while cored and neuritic plaques were also separately esti-
mated. Plaque density scores were obtained by assigning
values of none, sparse, moderate, and frequent, according to
the published CERAD templates (20, 23). Conversion of
the descriptive terms to numerical values resulted in scores
of 0 to 3 for each area (Fig. 1). Semiquantitative amyloid
scoring was performed blinded to clinical diagnosis by a single
observer (T.G.B.).

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### TABLE 1. Characteristics of Alzheimer Disease Patients Who Had In Vivo Amyloid Imaging

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<td>No</td>
<td>N</td>
<td>Y</td>
<td>4.9</td>
<td>1.1</td>
<td>FTLD-TDP</td>
<td></td>
</tr>
</tbody>
</table>

* WMR score is the sum of the parietal, temporal, frontal, and occipital lobes; each lobe was graded on a 0 to 3 scale (Fig. 1). Cases with a score of 2 or higher in 1 or more lobes were considered to have WMR in the pathology subset.
† The CAA score is the sum of the following cerebral lobes: parietal, temporal, frontal, and occipital, which were graded on a 0 to 3 scale. As nearly our entire AD series had some degree of CAA (87%), we defined CAA in the pathology subset when 1 or more of the cerebral lobes had a severity score of 2 or higher.
‡ Indicates pathology meeting criteria for diagnosis of dementia with Lewy bodies.
§ Indicates pathology met criteria for diagnosis of hippocampal sclerosis.

AD, Alzheimer disease; Arg, argyrophilic grain disease; CAA, cerebral amyloid angiopathy; CERAD, Consortium to Establish a Registry for Alzheimer’s Disease; FTLD, frontotemporal lobar degeneration; LB(s), Lewy body (bodies); NFT, neurofibrillary tangle score; NP, neuritic plaque; TDP, TAR-DNA binding protein-43; WMR, white matter rarefaction; IHC, average percentage of cortical gray matter in 6 cortical regions of interest occupied by β-amyloid immunoreactivity; SUVr, standard uptake value ratio for the average cortical uptake of florbetapir divided by the average cerebellar uptake.
Neuropathological Examination

Neuropathological examination was performed blinded to clinical diagnosis by a single observer (T.G.B.); assignment of a clinicopathological diagnosis was done after unblinding to clinical history. Multiple additional brain regions were dissected for neuropathological assessment and diagnosis according to the standard protocols of the Banner Sun Health Research Institute Brain and Body Donation Program (21). This included a comprehensive set of 22 blocks embedded in paraffin as well as a set of 6 to 8 blocks 3 × 5 cm from standard levels of the frontal, temporal, and parietal lobes as described above, as well as 1 block through the middle of the occipital lobe and 1 parasagittal block through the cerebellum at the level of the dentate nucleus. The temporal and parietal blocks contained standard levels of the thalamus, basal ganglia, and substantia nigra. These cryoprotected blocks were sectioned at 80-μm thickness on a sliding freezing microtome and stained with hematoxylin and eosin, Thioflavin S, and enhanced silver methods (Campbell-Switzer and Gallyas) for amyloid plaques and neurofibrillary tangles. Amyloid plaque and neurofibrillary tangle density and distribution were determined in these thick sections of the frontal, temporal, parietal, and occipital cortex as well as hippocampus and entorhinal cortex, based on the CERAD templates (23) and the aggregate impression from Thioflavin S, Campbell-Switzer, and Gallyas methods. Amyloid plaque distribution was described according to the Thal-Braak system while the distribution of neurofibrillary degeneration was described using the original Braak protocol; for both of these, their development was based on the usage of similarly thick sections (24, 25). As in the originally published studies of this group of subjects, the diagnosis of AD was based on a CERAD “probable” or “definite” classification (23).

Rating of WMR was done using a semiquantitative (0–3) scale on the 80-μm-thick sections stained with hematoxylin and eosin according to the fraction of centrum semiovale affected (Fig. 1) (21). Those cases with a score of 2 or higher in 1 or more lobes were considered to have significant WMR. Cerebral amyloid angiopathy (CAA) was defined semiquantitatively with 3 levels of severity by analogy to the CERAD templates; for this study, significant CAA was defined when 1 or more cerebral lobes had a severity score of 2 or higher. Argyrophilic grains (Args) were defined as typical spindle-shaped structures revealed by the Gallyas silver stain (26, 27) and were recorded as being present or absent.

Formalin-fixed, paraffin-embedded sections from a set of 10 standard brain regions (28) were immunostained with an antibody against phosphorylated α-synuclein peptide (1:10,000; rabbit polyclonal anti-human phosphoserine 129, gift of...
TABLE 2. Comparison of Subject Characteristics of Alzheimer Disease Groups, the Nondemented Group and the Group With Mild Cognitive Impairment

<table>
<thead>
<tr>
<th></th>
<th>Nondemented</th>
<th>MCI</th>
<th>All AD</th>
<th>AD With WMR</th>
<th>AD With LBs</th>
<th>AD With CAA</th>
<th>AD With Arg</th>
<th>AD With TDP-43</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>12</td>
<td>5</td>
<td>38</td>
<td>27</td>
<td>21</td>
<td>11</td>
<td>5</td>
<td>18</td>
</tr>
<tr>
<td>Interval of imaging until death (months), mean ± SD</td>
<td>4.8 ± 5.19</td>
<td>7.8 ± 8.44</td>
<td>7.1 ± 5.96</td>
<td>7.3 ± 6.0</td>
<td>7.0 ± 5.9</td>
<td>5.2 ± 5.5</td>
<td>6.4 ± 6.5</td>
<td>8.8 ± 6.8</td>
</tr>
<tr>
<td>Age at death (years), mean ± SD</td>
<td>79 ± 13.3</td>
<td>71 ± 18.2</td>
<td>82 ± 11.2</td>
<td>83 ± 10.9</td>
<td>82 ± 10.7</td>
<td>78 ± 11.9</td>
<td>90 ± 9.5</td>
<td>83 ± 3.1</td>
</tr>
<tr>
<td>Braak NFT stage, median (range)</td>
<td>III (I-IV)</td>
<td>I (I-IV)</td>
<td>VI (II-VI)*</td>
<td>VI (IV-VI)*</td>
<td>VI (IV-VI)*</td>
<td>VI (V-VI)*</td>
<td>VI (IV-VI)*</td>
<td>V (II-VI)*</td>
</tr>
<tr>
<td>Thal stage, median (range)</td>
<td>1 (0-3)</td>
<td>0 (0-2)</td>
<td>5 (3-5)*</td>
<td>5 (3-5)*</td>
<td>5 (3-5)*</td>
<td>5 (5)*</td>
<td>5 (3-5)*</td>
<td>5 (3-5)*</td>
</tr>
<tr>
<td>Average plaque density, median (range)</td>
<td>0 (0-3)</td>
<td>0 (0-3)</td>
<td>3 (3)*</td>
<td>3 (3)*</td>
<td>3 (3)*</td>
<td>3 (3)*</td>
<td>3 (3)*</td>
<td>3 (3)*</td>
</tr>
<tr>
<td>SUVr, mean (range)</td>
<td>1.0 (0.87-1.1)</td>
<td>1.0 (0.87-1.1)</td>
<td>1.4 (0.98-2.1)*</td>
<td>1.4 (0.98-2.1)*</td>
<td>1.3 (0.98-1.6)*</td>
<td>1.5 (1.2-2.1)*</td>
<td>1.4 (0.98-1.9)*</td>
<td>1.3 (0.98-1.76)*</td>
</tr>
<tr>
<td>β-amyloid IHC, mean (range)</td>
<td>0.2 (0-1.1)</td>
<td>0.4 (0-1.1)</td>
<td>6.4 (0.5-14.2)*</td>
<td>6.4 (0.5-14.2)*</td>
<td>6.0 (0.5-9.9)*</td>
<td>6.8 (1.4-14.2)*</td>
<td>3.6 (0.5-6.1)*</td>
<td>6.0 (0.5-14.2)*</td>
</tr>
</tbody>
</table>

Statistical significance was determined using Kruskal-Wallis test to test for the equality among normal, MCI, and each of the AD groups, respectively. The AD groups were not significantly different with respect to their AD-associated histopathological measures (Braak NFT stage, Thal stage, and average plaque density scores) or with respect to their in vivo (quantitative SUVr method) or postmortem (IHC) average cortical amyloid load estimates. There was considerable overlap among AD subgroups (Table 1).

*Significantly different from the normal control and MCI groups.

AD, Alzheimer disease; Arg, argyrophilic grain disease; CAA, cerebral amyloid angiopathy; LBs, Lewy bodies; MCI, mild cognitive impairment; NFT, neurofibrillary tangle score; NP, neuritic plaque; TDP, TAR-DNA binding protein-43; WMR, white matter rarefaction; IHC, average percentage of cortical gray matter in 6 cortical regions of interest occupied by β-amyloid immunoreactivity; SUVr, standard uptake value ratio for the average cortical uptake of flurbetapir divided by the average cerebellar uptake.

TABLE 3. Unadjusted p Values For Standard Flurbetapir Uptake Value Ratios and β-Amyloid Immunohistochemistry Comparisons Among Alzheimer Disease Groups

<table>
<thead>
<tr>
<th></th>
<th>AD With WMR</th>
<th>AD With LBs</th>
<th>AD With CAA</th>
<th>AD With Arg</th>
<th>AD With TDP-43</th>
</tr>
</thead>
<tbody>
<tr>
<td>AD with WMR</td>
<td>n/a</td>
<td>0.14, 0.636</td>
<td>0.489, 0.770</td>
<td>0.856, 0.060</td>
<td>0.297, 0.683</td>
</tr>
<tr>
<td>AD with LBs</td>
<td>0.14, 0.636</td>
<td>n/a</td>
<td>0.045, 0.499</td>
<td>0.696, 0.064</td>
<td>0.746, 1.000</td>
</tr>
<tr>
<td>AD with CAA</td>
<td>0.489, 0.770</td>
<td>0.045, 0.499</td>
<td>n/a</td>
<td>0.394, 0.082</td>
<td>0.115, 0.573</td>
</tr>
<tr>
<td>AD with Arg</td>
<td>0.856, 0.060</td>
<td>0.696, 0.064</td>
<td>0.394, 0.082</td>
<td>n/a</td>
<td>0.5, 0.139</td>
</tr>
<tr>
<td>AD with TDP-43</td>
<td>0.297, 0.683</td>
<td>0.746, 1.000</td>
<td>0.115, 0.573</td>
<td>0.5, 0.139</td>
<td>n/a</td>
</tr>
</tbody>
</table>

p Values for each comparison are listed in the following order: SUVr, IHC.

AD, Alzheimer disease; Arg, argyrophilic grain disease; CAA, cerebral amyloid angiopathy; LBs, Lewy bodies; n/a, not applicable; TDP, TAR-DNA binding protein-43; WMR, white matter rarefaction; IHC, average percentage of cortical gray matter in 6 cortical regions of interest occupied by β-amyloid immunoreactivity; SUVr, standard uptake value ratio for the average cortical uptake of flurbetapir divided by the average cerebellar uptake.

Dr Haruhiko Akiyama, Tokyo Institute of Psychiatry, Tokyo, Japan) (29); these were used to classify the density and distribution of LBs and related neuropil elements using the Unified Staging System for Lewy Body Disorders (28). Diagnostic criteria for dementia with LBs were those of the third Dementia with Lewy Bodies Consortium (14); the diagnosis was assigned when subjects met “intermediate” or “high” definitions.

Free-floating 80-μm-thick sections of the frontal and temporal lobes were immunostained with an antibody against phosphorylated TDP-43 peptide (1:10,000 rabbit polyclonal antihuman phosphoserine 409/410, gift of Dr Haruhiko Akiyama) (30). Diagnostic criteria for vascular dementia were adapted from those of Roman et al (31).

Statistical Methods

Statistical analyses and graphs were performed with Sigma Plot 12.1 (Systat Software, Inc. San Jose, CA) and Microsoft Excel (Microsoft Corporation, Redmond, WA). The Student t-test and the Mann-Whitney U test were used to compare group means for continuous and discontinuous measures, respectively. Chi-squared and Fisher exact tests were used to determine whether proportional measures were significantly different. Spearman correlations were used to show relationships between semiquantitative measures. For all tests, the type I error rate was set as 0.05. For multiple comparisons, the Benjamini-Hochberg False Discovery Rate Procedure was applied to control the expected proportion of falsely rejected hypotheses.
RESULTS

Table 1 shows the complete list of AD cases and their concurrent pathologies. Of the AD cases, 21 had LB-related α-synuclein pathology, 8 met clinicopathological criteria for dementia with LBs, 27 had significant WMR, 11 had significant CAA, 5 had Args, and 18 had incidental TDP-43 inclusions. Furthermore, 1 AD case had an additional diagnosis of Trisomy 21, and 1 had concomitant frontotemporal lobar degeneration with TDP-43 inclusions. There was considerable overlap, with many cases exhibiting more than 1 type of concurrent pathology. Only 3 AD cases did not have any of the above concurrent diagnoses (“pure” AD). In all groups (AD subsets and nondemented individuals), there were multiple cases with 1 or more infarcts (38% of the nondemented cases and 39% of the AD subjects had 1 or more infarcts). We did not analyze for the effect of infarcts on amyloid measures due to great variability in infarct size, location, and type. Table 2 summarizes the overall group demographics. Groups did not significantly differ from each other with respect to age of death, interval from imaging until death, or gender ratio. All AD subgroups had significantly higher CERAD neuritic plaque densities, as well as higher Braak NFT stages when compared with the MCI and nondemented control group (p < 0.001) but did not significantly differ from each other in these measures. In terms of both in vivo imaging–derived SUVr and postmortem β-amyloid IHC measures, when adjusting for multiple comparisons, all AD subgroups were significantly different from the normal control and MCI groups (p < 0.001), but there were no significant differences among the subgroups. There were no statistically significant differences on any measure between MCI and nondemented individuals. Unadjusted p values generated by comparisons of SUVr and β-amyloid IHC values of each AD subgroup are shown in Table 3. The only significant difference was in SUVr measures between AD with LBs and AD with CAA (p = 0.045).

There were significant correlations between cortical amyloid measures (SUVr and β-amyloid IHC) and both Braak neurofibrillary stage and Thal-Braak amyloid phase. Correlation coefficients (Spearman rho) were 0.76 for AD cases with CAA (p < 0.0001), 0.78 for AD cases with LBs (p < 0.0001), 0.60 for AD cases with Args (p = 0.003), 0.76 for cases with severe CAA (p = 0.0006), and 0.78 for AD cases with TDP-43 inclusions (p = 0.0003). For the entire AD group, the correlation coefficient was 0.71 (p < 0.001). When considering all cases together, using semiquantitative estimates of average cortical amyloid plaque densities derived from the Campbell-Switzer silver stain and Thioflavin S stain, the correlation coefficient (Spearman rho) was 0.76 for the comparison with average cortical β-amyloid IHC (p < 0.0001) and 0.73 for the comparison with SUVr (p < 0.0001). MCI and nondemented control subjects were included in all correlations in order to attain a wider range of measures.

DISCUSSION

There is great interest in validating amyloid imaging methods as they hold great promise for improving AD clinical diagnostic accuracy and for testing of disease-modifying agents. We have recently published results from PET studies using the amyloid ligand florbetapir F-18, demonstrating a strong correlation between in vivo and postmortem amyloid load estimates in the cerebral cortex, utilizing Alzheimer disease subjects as well as nondemented individuals (8, 11). These previous results found florbetapir-PET images rated as positive or negative for amyloid presence agreed, in 55 of 59 individuals, with postmortem histology for the presence or absence of a defined amyloid plaque density (11). The detailed neuropathological findings of these cases have not been published and are of considerable interest because AD is
FIGURE 3. Box plots of median and 25th and 75th percentiles of in vivo amyloid imaging measures for average cortical amyloid load of Alzheimer disease (AD) cases using the standard uptake value ratios (SUVR) in subjects with (gray boxes) or without (white boxes) concurrent pathologies: with CAA, n = 11; without CAA, n = 27; with phosphorylated TAR DNA binding protein-43 (TDP-43) inclusions, n = 18; without TDP-43, n = 20; with LBs, n = 21; without LBs, n = 17; with WMR, n = 27; without WMR, n = 11; with Args, n = 5; without Args = 33. Whiskers above and below the boxes indicate the 90th and 10th percentiles. Using the Mann-Whitney U test, the only significantly different pairwise comparison was that comparing AD subjects with and without WMR. CAA, cerebral amyloid angiopathy; LBs, Lewy bodies; WMR, white matter rarefaction; Args, argyrophilic grains. ** p = 0.002.

neuropathologically heterogeneous. We sought to determine whether amyloid imaging measures differed or had differing in vivo-postmortem correlation strength depending on the type of concurrent pathologies within AD. The 38 AD subjects in this study, which were selected from the original study group of 59 subjects (8), had a high frequency of other pathologies, including LBs, WMR, severe CAA, Args, and TDP-43. There were only 3 AD cases that did not contain at least 1 of these additional abnormalities, supporting the evolving consensus that AD is, more frequently than not, complicated by additional pathology (16–19). Despite this neuropathological heterogeneity, in vivo imaging measures of SUVR remained significantly correlated with average cortical postmortem β-amyloid IHC measures within all AD pathology subgroups. All AD cases except one had mean cortical SUVR values that were above the proposed cutoff “positive” ratio of 1.1 (Fig. 2) (32). The 1 positive case that was below the cutoff ratio based on histology (case #24, Table 1) had multiple concomitant pathologies, including hippocampal sclerosis, TDP-43 inclusions, Args, WMR, and LBs. There are no clear conclusions as to why this case is below the proposed cutoff, and it is possible that it is only random variation. Furthermore, both in vivo and postmortem average cortical amyloid measures for each AD subgroup were significantly greater than those for non-demented individuals, and the average SUVR values of each AD subgroup stay close to the overall mean. Overall, these data suggest that flortbetapir SUVR measures are a reliable predictor of postmortem histopathological β-amyloid load despite the substantial neuropathological variability commonly encountered in AD subjects.

This study contributes new clinicopathological correlative data for subjects that have received amyloid imaging during life. In particular, we are not aware of any reports that have described in vivo amyloid imaging results for AD with concurrent WMR, Args, or aberrant TDP-43 deposition (26, 33). Amyloid imaging results for subjects with clinically diagnosed LB disorders have been published, but these studies have generally lacked postmortem confirmation, and the clinical diagnostic accuracy for DLB has known limitations (34–42). To our knowledge, there has only been 1 small series and 1 case study of amyloid imaging with autopsy confirmation of DLB (36, 37). In the case study, there was a strong association between 11C-PiB retention and postmortem cortical β-amyloid densities in 17 corresponding ROIs (36). In a series of 5 clinically diagnosed Parkinson dementia and DLB cases that had come to autopsy, 3 cases were 11C-PiB-positive, and all had amyloid pathology at autopsy; 1 of these cases also met neuropathological diagnostic criteria for AD. Neither of the 2 11C-PiB-negative cases had significant amyloid pathology at autopsy (37). No amyloid imaging study has compared clinicopathological AD cases with and without LBs. Our results demonstrate that AD cases with LBs had significantly decreased SUVR measures versus AD cases without LBs; additionally, the only significantly different comparison out of all possible comparisons of average cortical SUVR between all AD subgroups was between the AD/CAA subgroup and the AD/LB subgroup, and again, the AD/LB subgroup had a lower average cortical SUVR. These results are consistent with previous reports demonstrating that plaque densities may be lower in the cortex of AD subjects with DLB versus AD subjects without DLB (14, 38, 43, 44). High densities of CAA are likely to be additive to amyloid plaques in affecting cortical SUVR because a high-resolution postmortem study has demonstrated that another amyloid imaging agent, 11C-PiB, binds with high sensitivity to CAA (45). However, we found no significant differences in average cortical SUVR between AD cases with and without higher CAA density.

Our study has some limitations. One limitation is that subjects tended to have either no amyloid or high amyloid loads, with relatively few having sparse to moderate loads. This is most likely due to the clinical trial selection process whereby most subjects were identified by neurologists with an interest in dementia and by the need to select subjects who were near death. Amyloid deposition in AD is hypothesized to be a rapid and mostly preclinical event (46). To enhance the range of amyloid densities, we therefore included the non-demented study individuals. Another limitation is the relatively low number of subjects. Although this is the largest amyloid imaging-to-autopsy study to date, our sample size is still relatively small, particularly when the cases were subdivided into several subgroups. Furthermore, many cases exhibited more than 1 type of concurrent pathology. Nevertheless, this is a real life setting for AD subjects, and most AD patients will have more than 1 additional non-AD pathology on neuropathological examination. The main purpose of this study was to examine whether quantitative cortical amyloid
imaging measures differed as a result of neuropathological heterogeneity within AD. The results show that despite con-
iderable neuropathological diversity, cortical amyloid imaging measures were remarkably uniform. In conclusion, the results of this study indicate that florbetapir PET–derived estimates of AD cortical amyloid load are not significantly altered by several common concomitant pathologies.

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