β-Amyloid Aggregation in Human Brains With Cerebrovascular Lesions

Leena Aho; Jukka Jolkkonen, PhD; Irina Alafuzoff, MD, PhD

Background and Purpose—The present study assessed β-amyloid (Aβ) protein aggregates in postmortem human brains in subjects who had experienced stroke to examine the proposed association between ischemic stress and the accumulation of Aβ reported in rodents.

Methods—A sample of 484 postmortem brains from nondemented subjects, lacking isocortical neurodegenerative pathology with verified cerebrovascular lesions, and 57 age-matched controls were assessed with respect to Aβ, Aβ40, and Aβ42 aggregates in the cortex and thalamus by immunohistochemical techniques.

Results—The load of Aβ aggregates did not display a significant association with cerebrovascular lesions. The load of Aβ, Aβ40, and Aβ42 aggregates increased with age, and there was a tendency toward higher odds ratios for Aβ aggregates, though not statistically significant, in subjects with acute cerebrovascular lesions. In the oldest subjects with cerebrovascular lesions and with both thalamic and cortical Aβ aggregates, the load of thalamic Aβ42 was significantly higher than the load of Aβ40.

Conclusions—Our findings indicate that cerebrovascular disease does not influence the load of Aβ, whereas a shift of aggregation from the Aβ40 to the Aβ42 residue is noted in the thalamus but only in aged subjects. It is impossible, however, to state whether this result is attributable to increased Aβ production, its insufficient elimination, or other susceptibility factors. (Stroke. 2006;37:2940-2945.)

Key Words: β-amyloid ■ cerebrovascular lesions ■ human brain ■ immunohistochemistry ■ postmortem

Recent publications suggest that focal cerebral ischemia in rats leads to the deposition of β-amyloid peptide (Aβ) in thalamic nuclei. The accumulation of Aβ was attributed to the disruption of the fast anterograde axonal transport caused by the ischemia, whereas the insufficient clearance of accumulated protein from thalamic nuclei was related to their location in the vascular end artery area. It has been reported that in humans also, ischemic brain damage leads to the accumulation of Aβ, at least in cortical arterial boundary zones.

Aβ is a hydrophobic, self-aggregating peptide consisting of 40 to 42 residues, and it is derived from the Aβ precursor transmembrane protein. Normally, cultured cells produce and release soluble Aβ peptide, and it has been shown in rats that Aβ is rapidly cleared from the brain. Recently, it was reported that in humans, the protein is eliminated from the extracellular space along the perivascular route. These findings seem to indicate that both the release and clearance of Aβ are possible mechanisms that could be involved in Aβ aggregation.

Deposition of Aβ in the brain is common in aged subjects and specifically in patients with Alzheimer disease (AD). Some recent studies have indicated that cerebrovascular disease (CVD) might play a role in the pathogenesis of AD, whereas other studies have not found any association between CVD and the presence of Aβ. The aim of the present study was to assess the accumulation of Aβ in the cortex and thalamus in humans as a possible secondary phenomenon to a verified stroke episode.

Materials and Methods

Figure 1 describes the scheme involved in the selection of the material.

Neuropathological Assessment

According to a standardized dissection protocol, all 1506 brains were routinely weighed, evaluated for grossly detectable lesions, fixed in 4% buffered formaldehyde for at least 1 week, and cut into coronal slices. Before the brains were sliced, the severity of atherosclerosis of the arterial circle of Willis was graded on a 4-step scale as none, mild, moderate, and severe. Grossly notable lesions were assessed on all fixed coronal slices (thickness, 1 cm) and noted as present or absent. During sampling, brain tissue sections were routinely taken from the nonaffected side, and additional sections were taken from affected regions. For all cases, the brain specimens were systematically taken from the frontal, temporal, parietal, precentral, and occipital cortices; the gyri cinguli; striatum; the basal forebrain including the amygdala; thalamus; hippocampus; the midbrain including the substantia nigra; the pons including the locus ceruleus; medulla; vermis; and cerebellar cortex.

Received June 30, 2006; accepted July 20, 2006.
From the Department of Clinical Medicine, Neurology (L.A., J.J.), and the Department of Clinical Medicine Neurology and Pathology (I.A.), Kuopio University, Finland.
Correspondence to Irina Alafuzoff, Kuopio University, P-O-B 1647, Harjulantie 1, Kuopio, Finland 70211. E-mail irina.alafuzoff@uku.fi
© 2006 American Heart Association, Inc.
Stroke is available at http://www.strokeaha.org

DOI: 10.1161/01.STR.0000248777.44128.93
Cerebrovascular microscopically detectable lesions were estimated on all 15 hematoxylin/eosin-stained sections and graded according to histopathological features as shown in Figure 2. Lesions were classified as intra- or extra-axial and ischemic or hemorrhagic. They were additionally classified by their predominant age, ie, acute, subacute, or chronic/old; however, subjects with lesions of age type were grouped as other (Figure 2). The volume of lesions was graded on a 2-step scale, with 1 only microscopically noted lesions and 2 both microscopically and macroscopically noted lesions. Selected sections were stained with the modified Bielschowsky silver impregnation technique, and AD-related lesions were classified into stages as proposed by the guidelines established by Braak and Braak.14

Case Selection
Of the original group, 484 subjects (mean±SE age, 68±1 years at death; range, 23 to 98 years) with cerebrovascular lesions (CVLs) and 57 age-matched control subjects (mean±SE age, 71±2 years at death; range, 42 to 91 years) were studied. The clinical data were collected retrospectively from medical records. None of these subjects had a diagnosis of dementia based on NINCDS-ADRDA15 and DSM-III-R criteria. It should be noted, however, that some of the cases might have displayed mild cognitive impairment that had not been recognized during life. The brain sections used in this study were from the end arterial zone, ie, thalamus at the level of the lateral geniculate body and from the watershed area, ie, parietal cortex. All samples were taken from the nonaffected side lacking acute, subacute, or chronic infarcts. The demographics of the study group are given in Table 1.

Immunohistochemistry
Aβ was visualized by immunohistochemical methods. Seven-micron-thick serial sections were cut from formalin-fixed, paraffin-embedded tissue from the parietal cortex and thalamus. Moreover, in sections from all subjects that showed Aβ positivity in the thalamic region, consecutive sections were immunostained with antibodies specific for Aβ40 and Aβ42 residues. Details regarding the staining procedures are given in Table 2. For detection, the Histostain SAP kit (Zemed) was used, together with Vector Red or diaminobenzidine chromogen.

Semiquantitative Assessment
The Aβ, Aβ40, and Aβ42 labelings were estimated in the most affected area that was chosen by inspection under magnification (×100). The number of stained aggregates was counted in the total
microscopic field (×100 magnification) and semiquantitatively assessed on a scale from 0 to 3. Score 0 represented no aggregates; 1 was equivalent to 1 to 5 aggregates; 2 was equivalent to 6 to 20 aggregates; and 3 represented ≥21 aggregates. The final Aβ score, ranging from 0 to 3 for each subject, was obtained by calculating the average of scores obtained in the 2 most affected areas.

Statistical Analysis
Data were analyzed with the SPSS 11.5 for Windows program. The relations between various clinical and pathological parameters were assessed by adjusted logistics regression and adjusted general linear univariate analyses. The strength of association was estimated by the odds ratio (OR), and this is presented with the 95% CI. Furthermore, nonparametric Mann-Whitney tests were used.

Results
The mean±SE age at death of subjects with CVLs (CVL subjects) was 68±1 years, with a range of 23 to 98 years, this being comparable to that of the 57 control subjects without CVLs (non-CVL subjects; mean±SE age at death, 71±2 years; range, 42 to 91 years). The sex distribution, postmortem delay, brain weight, and the age at death (Table 1) did not significantly differ between the CVL and non-CVL subjects.

The mean±SE age at death of the CVL subjects with Aβ aggregates was 73±1 years (108 women, 82 men), and that of non-CVL subjects with Aβ aggregates was 72±3 years (9 women, 11 men). The frequency of Aβ-positive subjects was associated with age at death (P<0.001), whereas no association was noted with sex (Table 1). Overall, Aβ aggregates were seen in 190 (39%) of CVL and in 20 (35%) of non-CVL subjects. There was no significant difference in the frequency of Aβ positivity in the comparison of CVL with non-CVL subjects. In general, the parietal cortex was affected with Aβ aggregates in 168 of 484 (34%) CVL and in 14 of 57 (25%) non-CVL subjects, whereas the thalamus was affected in 120 of 484 (25%) CVL and in 12 of 57 (21%) non-CVL subjects. No correlation was noted between the severity of arteriosclerosis of the arterial circle of Willis and the frequency of Aβ positivity. The results did not change after adjusting for age and Braak stage.

All cases, both non-CVL and CVL, were grouped on the basis of the Aβ distribution: group A included those without any aggregates; group B, those with only cortical/parietal aggregates; group C, those with only central/thalamic aggregates; and group D, those with both parietal and thalamic Aβ aggregates. Detailed results regarding these groups are given in Tables 3 and 4.

Aβ labeling (Aβ positivity) in non-CVL and CVL subjects is given in Table 3. Comparing non-CVL with CVL subjects, neither age at death nor Aβ load differed significantly in group B (n=78) and group C (n=28). In group C, the load of thalamic Aβ40 was rather equal, whereas the load of Aβ42

![Figure 2. Features of CVLs assessed in a hematoxylin-eosin-stained section at ×100 (insert at ×400). Ischemic lesions in acute (ie, a few days; A), pale areas (right lower corner) and vacuolization at the border between the lesion and the intact brain tissue (left upper corner); in subacute (ie, weeks; B) foamy macrophages; and in chronic (ie, months; C) cystic space surrounded by gliosis. Hemorrhagic lesions (D) extravasated red blood cells (right upper corner). E, Numerous hemosiderin-laden macrophages and F, gliosis around scattered hemosiderin-laden macrophages.](http://stroke.ahajournals.org/lookup/suppl/doi:10.1161/01.STR.0000213394.86931.6E/-/DC1/figure2)
was higher, but not statistically significantly, in CVL compared with non-CVL subjects. In group D (n=104), the age at death and the parietal and thalamic Aβ load between CVL and non-CVL subjects did not differ significantly. However, in group D, the load of thalamic Aβ42 was significantly higher in the CVL compared with non-CVL subjects and significantly higher than the corresponding burden of Aβ40. Furthermore, the load of thalamic Aβ and Aβ42 was significantly higher in CVL subjects in group D (n=98) when compared with CVL subjects in group C (n=22) and the load of parietal Aβ when compared with CVL subjects in group B (n=70). Contrary to this, the load of Aβ40 was significantly higher in CVL subjects in group C (n=22) when compared with CVL subjects in group D (n=98). The mean age was lowest in groups A and C (n=359). In these subjects, no Aβ was seen in the cortex, whereas surprisingly, in 28 subjects (group C) of 359 (8%), thalamic aggregates were noted. The results were not influenced by sex.

The influence of age and type of CVL lesion on Aβ labeling is given in Table 4. The type or age of the vascular lesions did not significantly influence the frequency of detecting Aβ positivity in any of the Aβ positivity distribution groups, i.e., B, C, or D. Even after adjusting for age and Braak stage, there was still no significant association between the type and age of the lesion and the presence of Aβ aggregates. However, a tendency toward a higher but non-significant OR for Aβ positivity was noted for subjects with an acute lesion when compared with subjects with any other type of lesion.

In CVL subjects, Braak stage increased with the age at death. When adjusted for age, there was a significant association between Braak stage and the numbers of Aβ+ subjects (P=0.001). Aβ aggregates were almost twice as more likely (OR, 1.8) to be seen in Braak stages I and II and 7 times as more likely (OR, 7.0) to be seen in Braak stage III and IV when compared with the reference group composed of subjects with Braak stage 0. In subjects with neurofibrillary pathology (Braak stages I through IV), Aβ aggregates were more often seen in both the parietal cortex and thalamus when compared with those subjects with no neurofibrillary pathology (Braak stage 0). In contrast to CVL subjects, in non-CVL individuals there was no association between the frequency of Aβ pathology and Braak stage.

### Discussion

In the normal human brain, the production and elimination of Aβ are considered to be in balance, and thus, no Aβ aggregates should be formed. It has been suggested that ischemic brain damage can induce deposition of Aβ in the human brain, and this was further supported by animal studies wherein Aβ aggregation was seen after occlusion of the middle cerebral artery in rats. One major discrepancy between these 2 studies is that in contrast to the situation in rodents, the human group included subjects with severe, concomitant, AD-related pathology; therefore, comparison of these 2 experiments might not be justified. To assess the relation between CVLs and Aβ aggregation, we included only nondemented subjects with no cortical AD-related pathology. The clinical data in our study were, however, collected retrospectively, and thus, some of our subjects assessed as being cognitively unimpaired might have displayed mild cognitive impairment during life.

### TABLE 2. Immunohistochemistry

<table>
<thead>
<tr>
<th>Antibody</th>
<th>Source, Code</th>
<th>Clone</th>
<th>Pretreatment</th>
<th>Dilution and Incubation*</th>
<th>Chromogen</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperphosphorylated τ</td>
<td>Innogenetics, BR-03</td>
<td>AT8</td>
<td>None</td>
<td>1:500 overnight + 4°C</td>
<td>4C° Diaminobenzidine</td>
</tr>
<tr>
<td>Aβ</td>
<td>Dako, M0872</td>
<td>6F/3D</td>
<td>80% Formic acid, 6 h</td>
<td>1:100 overnight + 4°C</td>
<td>Vector Red</td>
</tr>
<tr>
<td>Aβ40</td>
<td>BioSource, 44–348</td>
<td>Polyclonal</td>
<td>80% Formic acid, 6 h</td>
<td>1:1000 overnight + 4°C</td>
<td>Vector Red</td>
</tr>
<tr>
<td>Aβ42</td>
<td>BioSource, 44–344</td>
<td>Polyclonal</td>
<td>80% Formic acid, 6 h</td>
<td>1:1000 overnight + 4°C</td>
<td>Vector Red</td>
</tr>
</tbody>
</table>
TABLE 3.  

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Sex, F/M</th>
<th>Age at Death, Mean±SE, y</th>
<th>Cortical Aβ, Mean±SE</th>
<th>Thalamic Aβ, Mean±SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>n=484 subjects with CVD:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group A thalamic Aβ-negative</td>
<td>331</td>
<td>Non-CVL</td>
<td>13/24</td>
<td>70±2</td>
<td></td>
</tr>
<tr>
<td>Cortical Aβ-negative</td>
<td>294</td>
<td>CVL</td>
<td>127/167</td>
<td>65±1</td>
<td></td>
</tr>
<tr>
<td>Group B thalamic Aβ-negative</td>
<td>78</td>
<td>Non-CVL</td>
<td>8</td>
<td>72±4</td>
<td>1.3±0.3</td>
</tr>
<tr>
<td>Cortical Aβ-negative</td>
<td>70</td>
<td>CVL</td>
<td>34/36</td>
<td>72±1</td>
<td>1.3±0.1*</td>
</tr>
<tr>
<td>n=57 subjects without CVD:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group C thalamic Aβ-positive</td>
<td>28</td>
<td>Non-CVL</td>
<td>6</td>
<td>67±7</td>
<td>1.2±0.2* 0.8±0.3 0.3±0.2</td>
</tr>
<tr>
<td>Cortical Aβ-negative</td>
<td>22</td>
<td>CVL</td>
<td>11/11</td>
<td>64±3</td>
<td>1.3±0.1* 0.9±0.1* 0.8±0.1*</td>
</tr>
<tr>
<td>Group D thalamic Aβ-positive</td>
<td>104</td>
<td>Non-CVL</td>
<td>6</td>
<td>77±2</td>
<td>1.3±0.2 1.8±0.3 0±0 0.2±0.2*</td>
</tr>
<tr>
<td>Cortical Aβ-positive</td>
<td>98</td>
<td>CVL</td>
<td>63/35</td>
<td>76±1</td>
<td>1.7±0.1* 2.0±0.1* 0.5±0.12* 1.7±0.1*</td>
</tr>
</tbody>
</table>

For statistics, the nonparametric Mann-Whitney test was used (significant difference: *P<0.05).

In contrast to the results obtained in rodents, in our study no obvious association could be detected between Aβ aggregates and CVLs of various location, severity, and age. Similar results were recently obtained by Mastaglia and colleagues,13 when they evaluated 100 subjects not known to be demented or to have had a neurological disorder. CVD or stroke is considered a significant risk factor for dementia.9,10 Recently, circulating Aβ levels have been shown to be elevated in patients with acute ischemic stroke,11 suggesting a causal association between stroke and AD. According to this theory, one would have expected that when we assessed 484 subjects with CVLs either acute, subacute, or chronic in nature, some association would have been found between Aβ aggregates and CVLs. In agreement with our negative results, Honig et al12 reported in their study including 1054 subjects that after adjustment for age and sex, no significant association between the number of neuritic plaques and either clinical stroke or neuropathological infarcts could be seen. However, they found an association between neuritic plaque counts and atherosclerosis of large vessels, concluding that it is atherosclerosis rather than ischemic tissue damage that might be associated with AD. In our study, wherein we examined the extent of atherosclerosis in the arterial circle of Willis, no association was found between the severity of vessel pathology and Aβ aggregation. When comparing our study with previous investigations,11,12 one can note major differences in study design. In our study, the goal was to assess the ischemic/hypoxic stress–induced deposition of Aβ in subjects lacking concomitant isocortical AD pathology. In our opinion, this made it possible for us to assess Aβ pathology as a secondary phenomenon to CVLs alone. With this design, however we were unable to confirm any significant association between CVLs and Aβ aggregation.

A tendency toward a higher OR for Aβ aggregates, though not statistically significant, was noted in our study for

**TABLE 4.  Aβ Labeling in Subjects With CVLs in Relation to the Type and Age of the CVL**

<table>
<thead>
<tr>
<th></th>
<th>n=484 subjects with CVD</th>
<th>Sex, n/n, F/M=235/249</th>
<th>Age of Lesion</th>
<th>n</th>
<th>Age at Death, Mean±SE</th>
<th>Type of Lesion</th>
<th>Cortical Aβ, Mean±SE</th>
<th>Thalamic Aβ, Mean±SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thalamic Aβ-negative,</td>
<td>127/167</td>
<td>Acute</td>
<td></td>
<td>130</td>
<td>60±1</td>
<td>IAI</td>
<td>40</td>
<td>59</td>
</tr>
<tr>
<td>Cortical Aβ negative</td>
<td></td>
<td>Subacute</td>
<td></td>
<td>13</td>
<td>64±4</td>
<td>IAH</td>
<td>9</td>
<td>4</td>
</tr>
<tr>
<td>Old</td>
<td>67</td>
<td>71±2</td>
<td>52</td>
<td>13</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>84</td>
<td>69±1</td>
<td>52</td>
<td>27</td>
<td>5</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group B:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thalamic Aβ-negative,</td>
<td>34/36</td>
<td>Acute</td>
<td></td>
<td>27</td>
<td>68±2</td>
<td>IAI</td>
<td>9</td>
<td>12</td>
</tr>
<tr>
<td>Cortical Aβ-positive</td>
<td></td>
<td>Subacute</td>
<td></td>
<td>1</td>
<td>65</td>
<td>IAH</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Old</td>
<td>23</td>
<td>77±2</td>
<td>18</td>
<td>4</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>19</td>
<td>72±2</td>
<td>12</td>
<td>5</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group C:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thalamic Aβ-positive,</td>
<td>11/11</td>
<td>Acute</td>
<td></td>
<td>13</td>
<td>59±4</td>
<td>IAI</td>
<td>3</td>
<td>8</td>
</tr>
<tr>
<td>Cortical Aβ-negative</td>
<td></td>
<td>Subacute</td>
<td></td>
<td>1</td>
<td>81</td>
<td>IAH</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Old</td>
<td>4</td>
<td>70±1</td>
<td>3</td>
<td>1</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>4</td>
<td>71±3</td>
<td>0</td>
<td>3</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group D:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thalamic Aβ-positive,</td>
<td>63/35</td>
<td>Acute</td>
<td></td>
<td>41</td>
<td>73±1</td>
<td>IAI</td>
<td>12</td>
<td>20</td>
</tr>
<tr>
<td>Cortical Aβ-positive</td>
<td></td>
<td>Subacute</td>
<td></td>
<td>5</td>
<td>71±2</td>
<td>IAH</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Old</td>
<td>32</td>
<td>80±1</td>
<td>27</td>
<td>5</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>20</td>
<td>76±2</td>
<td>12</td>
<td>6</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

IAI indicates intra-axial ischemia; IAH, intra-axial haemorrhage; and EAH, extra-axial haemorrhage. For statistical differences, logistic-regression analysis was used, and no statistical differences were noted.
subjects with acute rather than chronic lesions. This would support the hypothesis that Aβ represents an acute-phase protein produced in a stress situation (present after acute lesion) that is sufficiently eliminated with time (not present in chronic lesions). This is in line with Aβ labeling seen in rodents in the surrounding areas of acute infarcts, which vanishes within a few weeks.1

There are several different mechanisms and pathways to eliminate Aβ from the brain. Aβ is removed from the brain by peptides7,18 such as neprilysin and insulin-degrading enzyme, as well as by microglia and astrocytes.19 Aβ is also absorbed into the blood via LDL receptor–related-protein-1 in younger animals and probably in young humans as well.20 All of these mechanisms appear to fail with age, at least in animals, and possibly in humans.21

Thus, the most important mechanism for the elimination of Aβ from the brain in the aged is the secretion of interstitial fluid along the perivascular drainage pathways. There is evidence that CVLs may cause functional changes in cerebral blood vessels and may thereby influence the perivascular elimination route7 and consequently lead to accumulation of Aβ. The loads of Aβ, Aβ40, and Aβ42 increased with age in our study; however, this phenomenon was not influenced by CVLs or sex. Our result might indicate that it is diffuse age-related changes in the walls of blood vessels rather than cerebral ischemia and CVLs that might be responsible for the insufficient elimination of Aβ peptide.

In the youngest groups, A and C, no Aβ was seen in the cortex, which is evidence of sufficient elimination. Compared with group D, which represent the oldest subjects, Aβ aggregates were noted in both the cortex and thalamus. Interestingly, the load of thalamic Aβ42 aggregates was significantly higher than the corresponding burden of Aβ40 in CVL subjects in group D. These findings indicate that CVD might indeed influence the accumulation of Aβ in the thalamus, particularly in shifting the accumulation from the Aβ40 to the Aβ42 residue. However, with our study design, it was impossible to state whether these results were caused by increased Aβ42 production, its insufficient elimination, or both processes. It should also be noted that when we used linear-regression analysis to assess the influence of other factors such as age and sex, no significant association between CVLs and Aβ, Aβ40, and Aβ42 residues was noted.

When assessing material from human subjects, a major limitation is the extensive variability of the material. Our CVL subjects varied regarding not only the type and age of the CVLs but also the etiopathogenesis of the lesion, age, sex, cause of death, concomitant systemic diseases, medications, education, lifestyle, and genetic predispositions. This should also be remembered when comparing human studies with those carried out on experimental animals.

In conclusion, in our postmortem human study, no obvious association could be detected between Aβ aggregates and CVLs, but a strong association was detected between Aβ aggregates, age at death and Braak stage. Further studies need to be completed that focus on both the vessel wall and issues related to the etiopathogenesis of CVLs before we can understand the functional deficiencies leading to the accumulation of pathological proteins.

Disclosures
None.

References
β-Amyloid Aggregation in Human Brains With Cerebrovascular Lesions
Leena Aho, Jukka Jolkkonen and Irina Alafuzoff

*Stroke*. 2006;37:2940-2945; originally published online November 9, 2006;
doi: 10.1161/01.STR.0000248777.44128.93
*Stroke* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2006 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/37/12/2940

**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/