

Original Article

Perivascular Neuritic Dystrophy Associated with Cerebral Amyloid Angiopathy in Alzheimer's Disease

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Abstract: Cerebral amyloid angiopathy (CAA) affects both leptomeningeal and parenchymal blood vessels and is common in Alzheimer's disease (AD). In some vessels, CAA is accompanied by localized neuritic dystrophy around the affected blood vessel. The aim of this study was to assess the distribution and severity of perivascular neuritic dystrophy in primary visual and visual association cortices. The severity of perivascular neuritic dystrophy and A β deposition was scored in an association cortex (Brodmann area 18) and a primary cortex (Brodmann area 17) with double labeling immunohistochemistry for tau and A β in 31 cases of AD with severe CAA. The perivascular tau neuritic dystrophy score was significantly worse in visual association cortex than in primary visual cortex. On the other hand, there was no difference in the perivascular A β score between the two cortices. There were positive correlations between the severity of perivascular tau and perivascular A β scores for both primary and association cortices. The results suggest that the local neuronal environment determines the severity and nature of the perivascular neuritic pathology more than the severity of the intrinsic vascular disease and suggest a close association between perivascular amyloid deposits, so-called dyschoric angiopathy, and perivascular neuritic dystrophy.

Key Words: Alzheimer's disease, amyloid angiopathy, A β , tau, neuritic dystrophy, visual cortex

Introduction

The neuropathology of Alzheimer's disease (AD) is characterized by neurofibrillary tangles (NFTs), neuropil threads (NTs), senile plaques (SPs) and cerebral amyloid angiopathy (CAA). Both SPs and CAA are composed of extracellular deposits of amyloid beta protein (A β) while NFTs and NTs are composed of intracellular deposits of microtubule associated tau protein. A number of studies have addressed the relationship of parenchymal A β deposits and tau pathology, but there are limited analyses of these relationships of tau pathology to A β vascular deposits. Clinicopathological studies have revealed that extracellular A β deposition in brain tissue is more poorly correlated with intellectual status than tau pathology [2-4]. This is in part due to the fact that some non-demented elderly individuals have many SPs, but minimal or no NFTs, a process that has been referred to as pathological aging [4]. SPs can be classified as neuritic and non-neuritic plaques, with diffuse plaques being a form of

the latter. Neuritic plaques are characterized by amyloid deposits and surrounding tau neuritic dystrophy. Diffuse plaques, which are frequent in pathological aging, are characterized by non-compact amyloid deposits with no tau neuritic pathology. CAA affects both leptomeningeal and intraparenchymal blood vessels and is common in AD. In some vessels, CAA is accompanied by localized neuritic dystrophy around the affected blood vessel [1]. Perivascular tau neuritic dystrophy would not be predicted to occur in CAA in pathological aging given the absence of cortical tau pathology, but this has not been previously studied.

The distribution of CAA is not uniform; some investigators have noted CAA in the frontal lobe, but CAA is more frequent and severe in the occipital lobe [5]. Tau related pathology (i.e. NFTs and NTs) is more severe and occurs earlier in the disease process in association cortices (e.g., visual association cortex - Brodmann areas 18 and 19) compared to

primary cortices (e.g., primary visual cortex - Brodmann area 17) [3, 6, 7]. In this study, we investigated the severity and distribution of tau immunoreactive perivascular neuritic dystrophy associated with CAA in visual and association cortices of AD.

Material and Methods

Selection of Cases

We investigated 31 cases of AD with severe CAA (19 men, 12 women; age of death, 80.0 ± 10.0 years; race 28 Caucasian, 2 African American and 1 Hispanic; brain weight 1052 ± 182 g) from the neuropathology files of Mayo Clinic in Jacksonville, FL. The cases had undergone quantitative neuropathological assessments of SPs and NFTs in multiple cortical and subcortical sections with thioflavine-S fluorescent microscopy to assess Alzheimer-type pathology and to assign a Braak stage [6]. All cases fulfilled the NIA-Reagan criteria for AD [8], and all had many SPs and a Braak NFT stage of IV or greater (average: 5.6 ± 0.6).

To evaluate the severity of perivascular A β deposition, which is also referred to as dyschoric angiopathy, and tau neuritic dystrophy related to CAA, we performed double-labeling immunohistochemistry of tau and A β . Sections from paraffin embedded tissue of occipital cortex, including both visual and association cortices, were cut at a thickness of 5- μ m. The deparaffinized and dehydrated sections were steamed for 30 minutes for antigen retrieval followed by blocking of endogenous peroxidase with 3% H₂O₂. The sections were then treated with 5% normal goat serum for 20 min. Subsequently, the sections were incubated with a phospho-tau monoclonal antibody, CP13 (which recognizes a phospho-epitope on tau at Ser 202 and generously supplied by Peter Davies, Albert Einstein College of Medicine, Bronx, NY) at room temperature for 45 min.

After incubation with the primary antibody, the sections were treated with Envision-Plus, Peroxidase, Mouse (DAKO, Santa Barbara, CA) for 30 min. Peroxidase labeling was visualized with a solution containing 0.02 mg/mL 3, 3'-diaminobenzidine (DAB). After the DAB reaction of the first immunohistochemical cycle, the sections were treated with Doublestain block reagent (DAKO, Santa

Barbara, CA) for 3 min. The second primary antibody, anti-pan A β (33.1.1; Todd Golde, Mayo Clinic) was then applied at room temperature for 45 min. The sections were then treated with alkaline phosphatase labeled polymer (DAKO, Santa Barbara, CA) for 30 min. Alkaline phosphatase labeling was detected with a chromogen solution containing 0.15 mg/mL of 5-bromo-4-chloro-3-indolyl-phosphate/nitro blue tetrazolium (SIGMA, St. Louis, MO). The sections were dehydrated and coverslipped without counterstaining.

In each region (Brodmann areas 18 and 17) of each AD case, five blood vessels with CAA were randomly selected. To evaluate the severity of perivascular neuritic dystrophy and A β deposition related to CAA in the same blood vessels, neuritic tau pathology and A β deposition was scored for each vessel, as follows: perivascular neuritic dystrophy (**Figure 1**, left panel: 0 - absent; 1+ - rare, around vessel; 2+ - partially around vessel; 3+ - entirely around vessel); perivascular A β deposition (**Figure 1**, right panel: 1+ - only vessel, 2+ - vessel and parenchymal deposits partially around vessel; 3+ - vessel and parenchymal entirely around vessel).

A mean score for each measure and region was calculated for each case. Statistical analyses were performed with SigmaStat (Systat Software, Inc., Point Richmond, CA), and the significance level was set at $p < 0.05$. The Mann-Whitney U test was used to assess differences in perivascular A β and neuritic dystrophy scores between visual and association cortices. Spearman's correlation coefficient was used to analyze the correlation between A β and neuritic dystrophy scores in the two cortical regions.

Results

Given that CAA is detected in most AD brains [9, 10], it was not difficult to identify cases suitable for study. All cases had moderate to severe CAA in both parenchymal and leptomeningeal vessels. The severity of CAA differed by region and by individual. Both visual and association cortices were affected in almost every patient. Tau pathology (i.e. NFT and NTs) and perivascular neuritic dystrophy related to CAA was observed in cortical layers II, III and V, and was more severe in the association than visual cortex (**Figure 2**). The severity of perivascular tau neuritic dystrophy

and A β deposition was assessed in the two regions. The perivascular tau neuritic dystrophy score differed significantly between visual and association cortices ($P=0.003$; **Figure 3**). On the other hand, there was no difference in the perivascular A β score

between the two cortices ($P=0.302$; **Figure 4**). There were positive correlations between the severity of perivascular tau and perivascular A β scores for both primary and association cortices (**Table 1**).

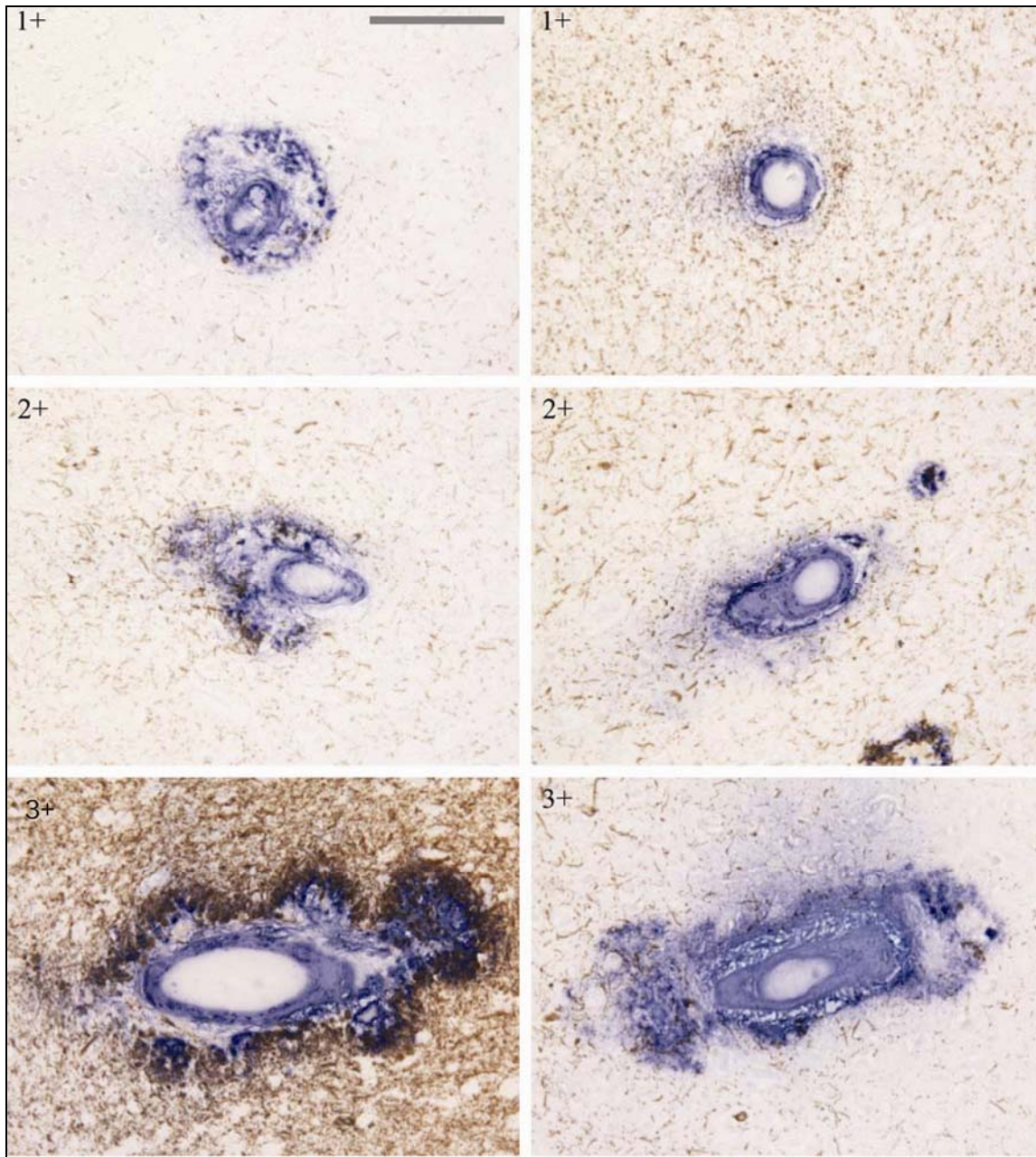


Figure 1 Representative visual fields showing perivascular neuritic tau pathology and amyloid deposition related to CAA with examples for severity score. Double immunostaining for A β (blue) and tau (brown). Left panel shows range of perivascular neuritic dystrophy, with tau (brown) related to CAA. Scores: 1+, 2+ and 3+ (as described in text). Right panel represents perivascular amyloid deposition (blue) related to CAA. Score 1+, 2+ and 3+ (as described in text) (Bar = 38 μ m).

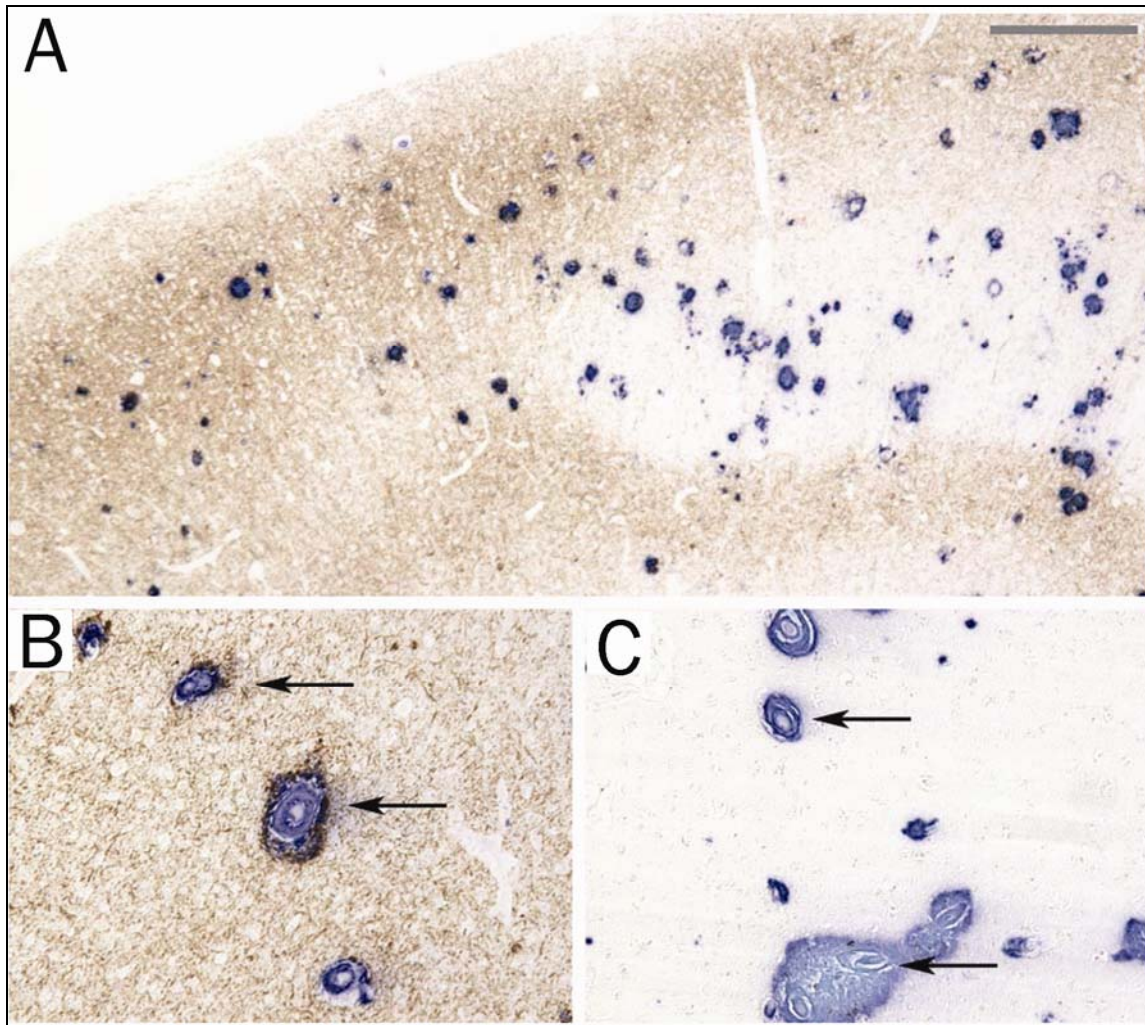


Figure 2 A low power photomicrograph of the border (A) between the primary visual cortex (right half; area 17) and the association cortex (left half; area 18) of a patient with AD and CAA that has been double immunostained for A β (blue) and tau (brown). The left half is rich in tau pathology. (Bar = 188 μ m) B. CAA accompanied by perivascular neuritic dystrophy in association cortex. (Bar = 75 μ m) C. CAA accompanied by minimal to no perivascular neuritic dystrophy in visual cortex. (Bar = 75 μ m).

Discussion

At present, the widely accepted neuropathologic criteria for AD do not include assessment of CAA, let alone perivascular tau neuritic dystrophy associated with CAA [6]. While there have been a few reports addressing the relationship between tau pathology and CAA, there have been no previous studies addressing potential differences in this pathologic feature in primary versus association cortices. In a recent study, McKee and co-workers reported that CAA, SPs and NFTs can be detected in occipital association cortices in cases with

absent or sparse medial temporal lobe Alzheimer type pathology, suggesting that occipital association cortices may be particularly vulnerable in preclinical AD [7] and warranting further investigation of occipital pathology. Delacourte and coworkers reported the morphological characteristics and relationships of CAA to tau pathology in AD [1], and Williams and coworkers described perivascular tau neuritic dystrophy associated with CAA [11]. None of these studies specifically addressed the relationship between perivascular A β deposition and tau neuritic dystrophy associated with CAA with respect to association and visual cortices.

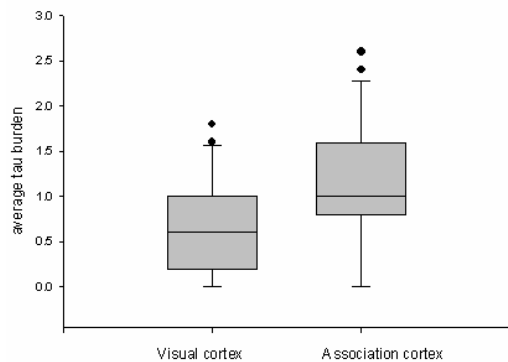


Figure 3 Comparison of perivascular neuritic dystrophy related to CAA in visual and association cortices. The boxes show median and 25th and 75th percentiles with whisker plots showing 10th and 90th percentiles. The outliers are shown as filled circles. The average tau burden score is significantly higher in association cortex than in visual cortex.

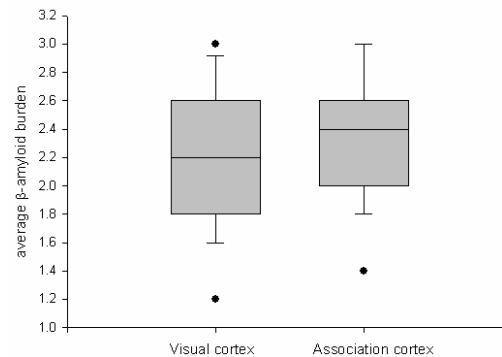


Figure 4 Comparison of perivascular A β deposition related to CAA in visual and association cortices. The boxes show median and 25th and 75th percentiles with whisker plots showing 10th and 90th percentiles. The outliers are shown as filled circles. The average amyloid burden score is not significant between the two cortices.

A result of this study that is worth emphasizing is that perivascular neuritic dystrophy associated with CAA is more severe in association than visual cortices (**Figure 3**). This suggests that the local neuronal environment determines the severity or nature of the perivascular pathology more than the severity of the intrinsic vascular disease. That the relationship had no bearing on severity of CAA or perivascular A β is supported by the fact that there was no difference in the average amount of perivascular A β deposition in the two cortices (**Figure 4**).

In this study, we did not investigate vessels with no A β deposition; however, there is a previous report that A β severity in CAA is not different between the calcarine cortex and visual association area 19 [7]. If all vessels had been examined, the average amount of A β deposition in vessels may not have been different between the two cortices.

The results of the present study suggest that perivascular neuritic dystrophy related to CAA is a neglected feature of AD pathology and that further studies are warranted to determine the clinical significance of this pathology and its usefulness in neuropathologic diagnosis.

The findings support the contention that extracellular A β deposition might trigger local tau neuritic dystrophy [12-14]. In favor of this

hypothesis is the fact that neuritic dystrophy was correlated with severity of perivascular A β deposits. The positive correlation between the severity of perivascular neuritic dystrophy and perivascular A β deposition related to CAA in both cortices (**Table 1**) might support the contention that A β deposition related to CAA induces tau pathology. Arguing against this hypothesis is that the same degree of CAA and perivascular A β was associated with different severity of tau neuritic dystrophy depending upon the local environment. In the association cortex, a region that is vulnerable to tau pathology, it was considerable, while in the primary cortex, which is less vulnerable to tau pathology, there was less perivascular neuritic dystrophy.

On the other hand, extensive vascular pathology associated with severe CAA may lead to leakage of toxic factors from the blood stream through a disrupted blood brain barrier. Changes in glia (perivascular microglia and astrocytes) associated with CAA and perivascular A β deposition may also play a role in perivascular neuritic dystrophy through release of neurotoxic mediators. Further studies looking at these factors may shed light on why dyschoric angiopathy is associated with neuritic dystrophy and why this process varies depending upon the nature of the cortical region in which the dyschoric amyloid angiopathy resides.

Table 1 Correlations between perivascular neuritic dystrophy and A β scores

Primary visual cortex	Visual association cortex
r = 0.245	r = 0.267
p < 0.01	p < 0.01

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