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Secondary microvascular degeneration in amyloid angiopathy of patients with hereditary cerebral hemorrhage with amyloidosis, Dutch type (HCHWA-D)

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Abstract Various secondary microvascular degenerative and inflammatory alterations may complicate cerebral amyloid angiopathy (CAA) and contribute to the morbidity of CAA-associated stroke. We have investigated the severity of CAA-associated microangiopathy in a genetically determined Dutch form of CAA (HCHWA-D) that has major similarities to the type of CAA that more commonly occurs with aging or Alzheimer's disease (AD). The presence and extent of the following vascular abnormalities was assessed: (1) hyalinization/fibrosis, (2) microaneurysm formation, (3) chronic (especially lymphocytic) inflammation, (4) perivascular multinucleated giant cells/granulomatous angiitis, (5) macrophages/histiocytes within the vessel wall, (6) vessel wall calcification, (7) fibrinoid necrosis, and (8) mural or occlusive thrombi. (Of these, calcification of CAA-affected vessel walls has, to our knowledge, been described in only a single patient with CAA-associated cerebral hemorrhage.) Some of the changes, such as histiocytes in blood vessel walls and the relationship of vascular hyalinosis to amyloid β/A4 protein deposition, were highlighted by immunohistochemistry. By assessing the numbers of sections in which the changes were present for each case, a 'score' reflective of CAA-associated angiopathy could be obtained. This 'score' was reproducible among several observers. We suggest that it might also be applicable to quantifying severe CAA and related microvascular degenerative changes in patients with AD. β/A4 immunoreactivity was often sparse and adventitial (or almost absent) in severely hyalinized arterioles and microaneurysms. However, macrophages were prominent in the walls of such vessels and may play a role in the pathogenesis and progression of CAA-related microvasculopathy.

Key words Amyloid angiopathy · Dutch (HCHWA-D) · β/A4 protein · Macrophages · Hemorrhage, cerebral

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Introduction

Cerebral amyloid angiopathy (CAA) defines a cortical and leptomeningeal microangiopathy – especially involving arterioles – characterized by the deposition of fibrillar amyloid in the vascular media and/or adventitia (the latter especially prominent among affected meningeal vessels) [5, 34, 35, 41]. With medial involvement, smooth muscle cells undergo degeneration and fragmentation with resultant weakening of the vessel wall [17, 40, 47]. This sometimes leads to rupture of CAA microvessels, resulting in cerebral hemorrhage. CAA is one of the microscopic neuropathological features of Alzheimer's disease (AD) [24, 35, 41]. As with other AD-associated lesions [e.g., senile plaques (SPs)], a significant degree of CAA can occur in non-demented elderly patients [34, 35, 41].

Assessing the severity of CAA in a given brain can be problematic. Vonsattel et al. [42] have suggested a useful scheme by which CAA can be graded in a given cerebral artery/arteriole, utilizing the presence of viable/atrophic smooth muscle (sm) cells, vessel wall fragmentation and expansion, and evidence for leakage of blood (recent or old) from the affected vessel lumen into surrounding brain parenchyma. This system does not take into account the topography of CAA, only the severity of changes in a

given vessel. The tendency for stroke [especially intracerebral hemorrhage (ICH)] associated with CAA may be aggravated by the presence of one or more CAA-associated microangiopathies, which include aneurysmal dilatation of affected vessels, obliterative intimal changes, chronic inflammatory infiltrates in close proximity to the affected vessel, hyaline (nonamyloid) arteriolar degeneration and fibrinoid necrosis [15, 23, 25, 42]. Vonsattel et al. [42] have further suggested that fibrinoid necrosis is commonly seen in the brains of patients with CAA-associated hemorrhage and only in these patients.

To study the pathogenesis and progression of CAA-associated microvasculopathies (CAA-AM) more systematically, an optimal patient population to examine would be one in which (unlike in AD) severe CAA is a consistent and predominant component of the neuropathological picture. Two well-characterized patient populations exist in whom CAA-associated ICH occurs as an inherited autosomal dominant trait. Icelandic CAA or hereditary cystatin C amyloid angiopathy (HCCAA) results from deposition within cerebral arteriolar walls of a mutant form of cystatin C/γ-trace protein [5, 26]. The other arises from a mutation at codon position 693 of the amyloid precursor protein gene (on chromosome 21), resulting in an amino acid substitution at position 22 of the Alzheimer β/A4 protein in Dutch patients (hereditary cerebral hemorrhage with amyloidosis-Dutch, HCHWA-D) [3, 11, 14, 32]. In HCHWA-D, predictably severe CAA with light microscopic and immunohistochemical similarities to the form seen in AD occurs in all patients who have the mutation [3, 20, 45]. Only minimal parenchymal β/A4 deposition is found in the neocortex of HCHWA-D patients [19–21].

Here we report the results of a systematic study of CAA-associated microangiopathies in autopsy brain specimens from HCHWA-D patients. We expand on the concept of CAA-associated microvasculopathy and describe a simple system by which the severity of CAA-associated microvascular change may be assessed and graded. This ‘grading system’ may furthermore be utilized in the assessment of less frequently encountered CAA-associated microangiopathy in Alzheimer’s disease (AD)/senile dementia of Alzheimer type (SDAT) patients, and may facilitate understanding of the role of CAA in stroke and ischemic vascular dementia.

Materials and methods

All available histopathological brain material was reviewed from archival autopsy specimens of 29 HCHWA-D patients. Several previous publications have presented clinical profiles of the patients and relevant neuropathological findings, including various immunohistochemical studies pertinent to this investigation that have reported microglial and macrophage localization in close proximity to CAA-affected microvessel walls [11–13, 18–21, 44, 45]. All autopsy brains have been extensively sampled, yielding (usually) between 15 and 30 separate blocks of neocortex, subcortical white matter, deep central gray matter, brain stem and cerebellum for review. In all cases, routine study of the cases has included staining [in addition to hematoxylin and eosin (H&E)] of serial or sequential sections with silver stains (e.g., Bodian methenamine silver), Congo red, myelin stains (e.g., Klüver-Barrera), stains that highlight connective tissue alterations (e.g., Azan) and (in some cases) immunohistochemistry using various primary antibodies to β/A4 amyloid proteins or components, glial fibrillary acidic protein and synaptophysin [19–21]. For this study, selected blocks were also restained with antibodies to β/A4 protein, CD-68 (a macrophage marker), and MIB-1, which recognizes the nuclei of cells in which DNA synthesis is occurring (β/A4, CD-68 and MIB-1 primary antibodies were purchased from Dako, Copenhagen, Denmark). These immunoreagents were chosen because of our specific interest in the relationship between β/A4 protein, macrophages and proliferating cells, and degenerative changes within vessel walls. Appropriate positive and negative controls were performed for each antibody. In selected cases in which calcified vessels were also noted or suspected, staining was carried out using Perl’s Prussian blue (for iron) and the von Kossa technique (for calcium).

Each available histological section was scored as having (+) or not having (–) the histological feature of interest in relation to CAA, which is present to a severe degree (by definition) in all HCHWA-D brains. Although CAA in HCHWA-D may affect capillaries and venules, the most consistent and severe involvement is of arterioles. Specific histopathological features assessed were: (1) vessel wall fibrosis, hyalinization or collagenosis; (2) microaneurysm formation; (3) chronic (lymphocytic) adventitial (rarely transmural) inflammation of any degree (i.e., with only two to three lymphocytes present); (4) multinucleated/foreign body-type giant cells (granulomatous inflammation) surrounding an affected artery; (5) histiocytes/macrophages within the vessel wall; (6) mural calcification (identifiable on routine stains but highlighted using the von Kossa technique); (7) fibrinoid necrosis; and (8) thrombi that were either occlusive or mural. These CAA-AM are modified slightly from those enunciated by Mandybur [23]. Scoring of cases was performed on available routine stained sections, including those stained with H&E, silver, Klüver-Barrera and Congo red – the availability of special stains on most tissue blocks effectively allowed for evaluation of questionable findings on serial or subserial sections from each study patient. Immunohistochemical and cytochemical stains were carried out to gain insights into the pathogenesis of CAA-AM. The following scores for a given finding were assigned to each case: 1, if an observation was present on one to two sections/case; 2, if present on three to five sections; and 3, if present on six or more sections. Thus, the score for CAA-AM could vary for each case from 0 to 24, i.e., the score could be 0 even when a severe degree of CAA was present. All cases were scored over a short time period by one author (H.V.V.). However, to assess inter-observer reproducibility of case assessments and scores, three cases representing a substantial range of severity of CAA-AM were evaluated and scored independently by three other co-authors (S.G.v.D., R.N., M.L.C.M.) having extensive experience with HCHWA-D.

Statistical correlations between individual CAA-AM features were established using Spearman correlation coefficients.

Results

Of the patients from whom brains were examined, 7 had been known to be clinically hypertensive during life, 15 were not known to be hypertensive, and for 7 patients insufficient clinical information was available to draw a conclusion. The numbers of strokes experienced by the patients varied from one to ten; 6 patients each had either one or two strokes, 2 patients had experienced three strokes, 4 had experienced four strokes each, 3 patients had a history of five or six strokes each, 2 patients had experienced seven, while a single patient had ten episodes. For 2 patients, the clinical number of strokes was given as ‘greater than three’. Apolipoprotein E (ε) genotypes are available for 9 patients and were as follows: 3/3 : 3; 3/4 : 3;

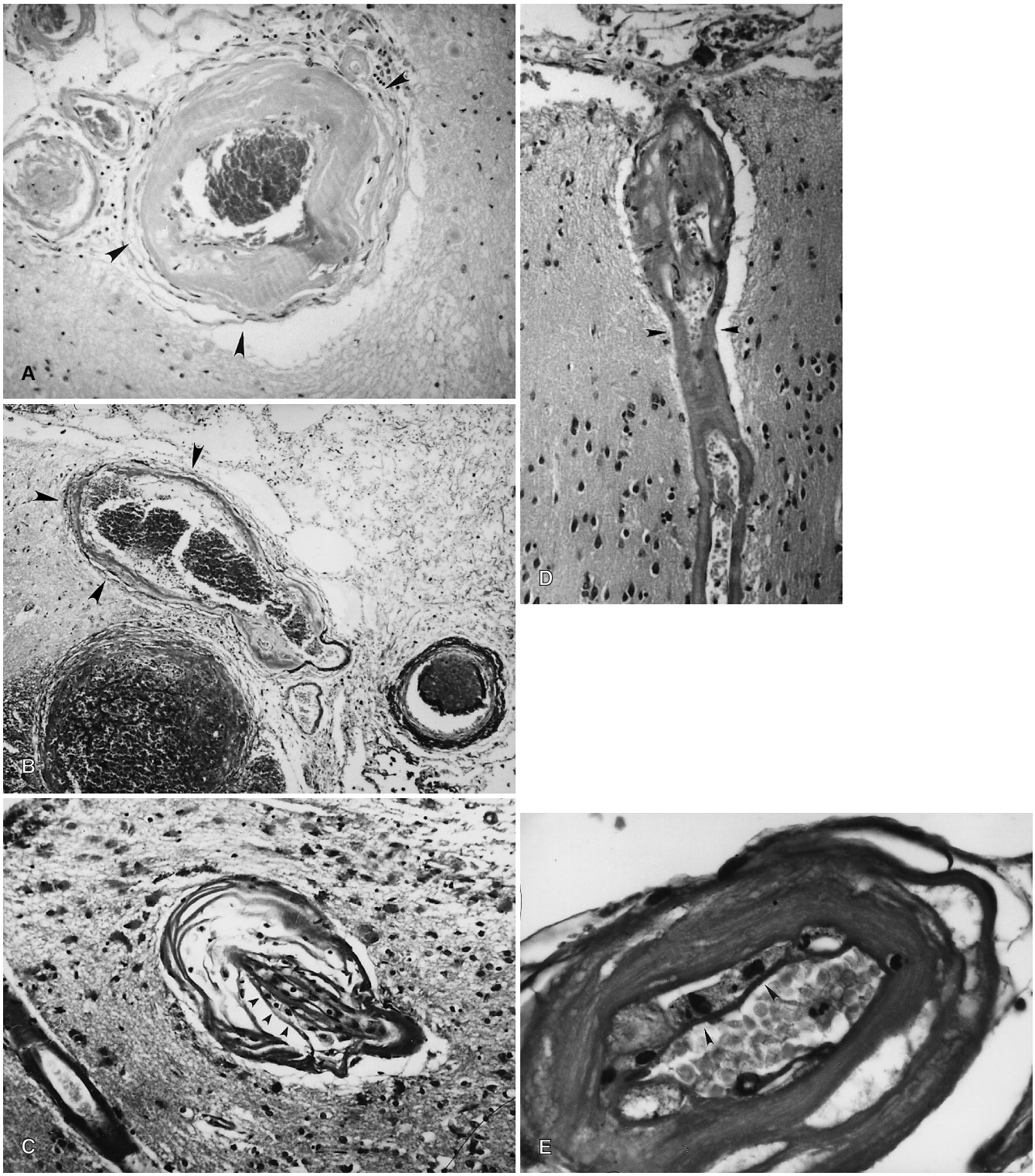

Fig. 1 A–E Examples of CAA-AM in brain specimens from HCHWA-D patients. **A** Markedly hyalinized artery *(arrows)* adjacent to smaller vessels involved by CAA. **B**, **C** Microaneurysm formation in parenchymal arteries (two different patients), with characteristic ectasia and 'flask-like dilatation' *(arrows)* emanating from a small artery in each case. In **C**, note histiocytes *(arrowheads)* in a portion of the aneurysm wall, which may have contributed to narrowing of the lumen. **D** Microaneurysm in the subpial region of segment of an amyloid-laden arteriole; the dilated segment was observed in several subserial sections. *Arrowheads* indicate junction between intact vessel wall and the microaneurysm. **E** Histiocytes *(arrows)* in the wall of a thickened vessel. (*CAA* cerebral amyloid angiopathy, *AM* associated microangiopathy, *HCHWA-D* hereditary cerebral hemorrhage with amyloidosis-Dutch type). **A**, **C–E** H&E; **B** Bodian methenanine silver stain. **A**, **C**, **D** × 124; **B** × 50; **E** × 390

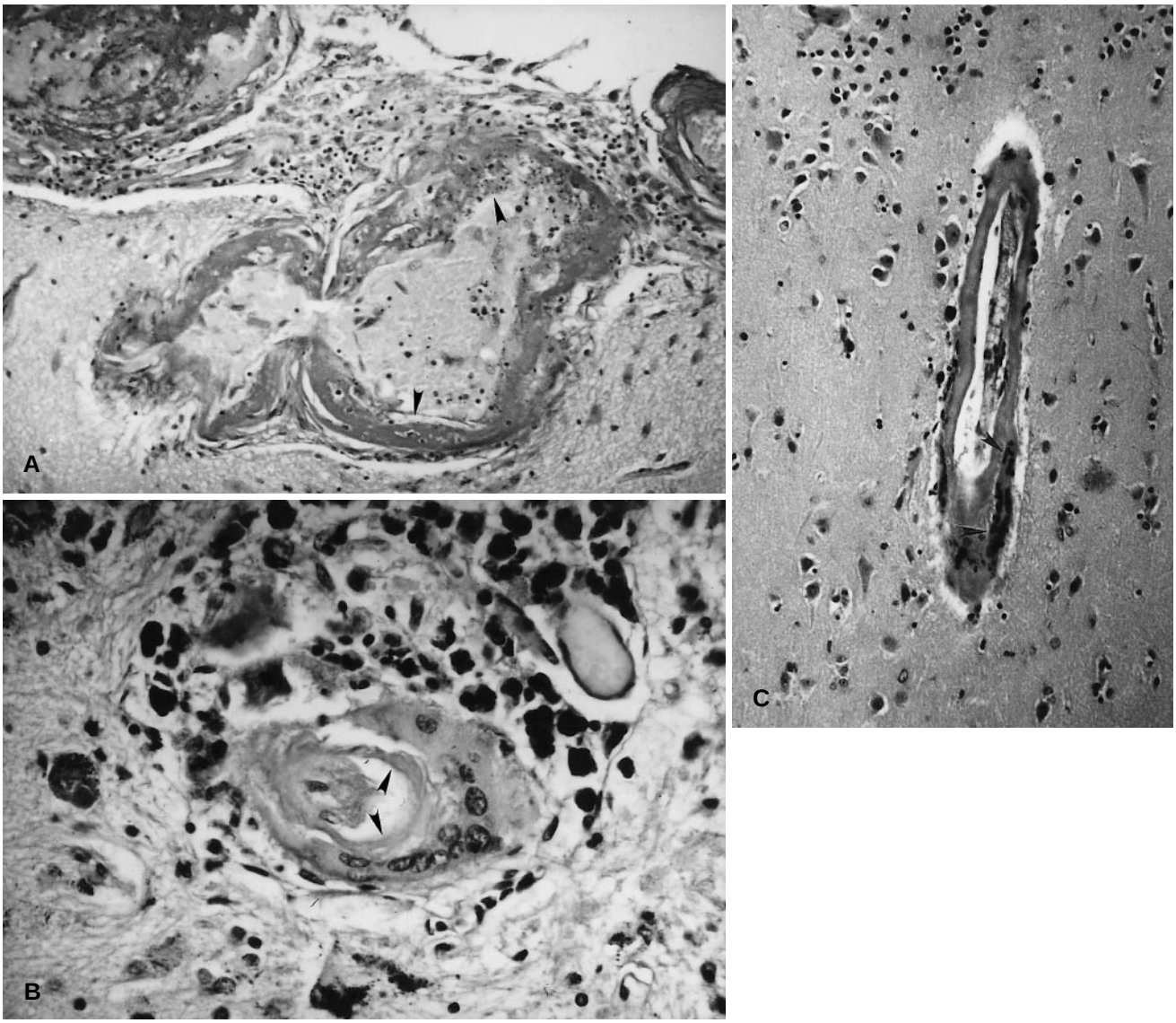

Fig. 2 A–C CAA-AM. **A** Fibrinoid necrosis. Note patchy nuclear fragmentation *(arrowheads)* in arterial wall, which also shows aneurysmal dilatation. **B** MNGC formation adjacent to arteriolar amyloid from an area of subpial cortex in which abundant adjacent astrogliosis and hemosiderin are present. Crescentic MNGC surrounds an amyloid-laden arteriole; vessel wall amyloid indicated by *arrowheads*. **C** Another example of MNGC on one segment of the adventitial aspect of a CAA arteriole in a different patient; *arrowheads* indicate junction between amyloid and the MNGC. (*MNGC* multinucleated giant cell). **A–C** H&E. **A**, **C** × 136; **B** × 350

2/3 : 1; 2/4 : 1; 4/4 : 1. Other work from this group has shown that the apolipoprotein E genotype appears *not* to modulate amyloid-related structural lesions in HCHWA-D [4].

Among the 29 autopsy cases evaluated, CAA-AM scores ranged from 0 to a maximum of 21, with a mean and standard deviation of 5.5 ± 5.2. Selected examples of CAA-AM are illustrated in Figs. 1 and 2. There was good inter-observer consistency of scores and findings (i.e., usually no more than 10–20% standard deviation around a mean) among the four experienced observers who all

Table 1 Frequency of CAA-AM features among HCHWA-D cases. Total of 29 autopsy brains were examined with mean overall score of 5.5 ± 5.2. (*CAA* cerebral amyloid angiopathy, *AM* associated microangiopathy, *HCHWA-D* hereditary cerebral hemorrhage with amyloidosis-Dutch type, *FBGC* foreign body type giant cell)

Histopathological feature	No. of cases with the given CAA-AM score for the stated feature[a]			
Score:	0	1	2	3
	Number of cases			
Hyalinization	9	8	7	5
Microaneurysms	16	6	5	2
Vessel wall histiocytes	15	5	5	4
Perivascular lymphocytes	10	11	4	4
Granulomatous inflammation [FBGC]	24	3	1	1
Vascular thrombi	18	9	1	1
Fibrinoid necrosis	23	1	4	1
Calcification	23	5	1	0

[a] For a discussion of 'scoring' system, see Materials and methods

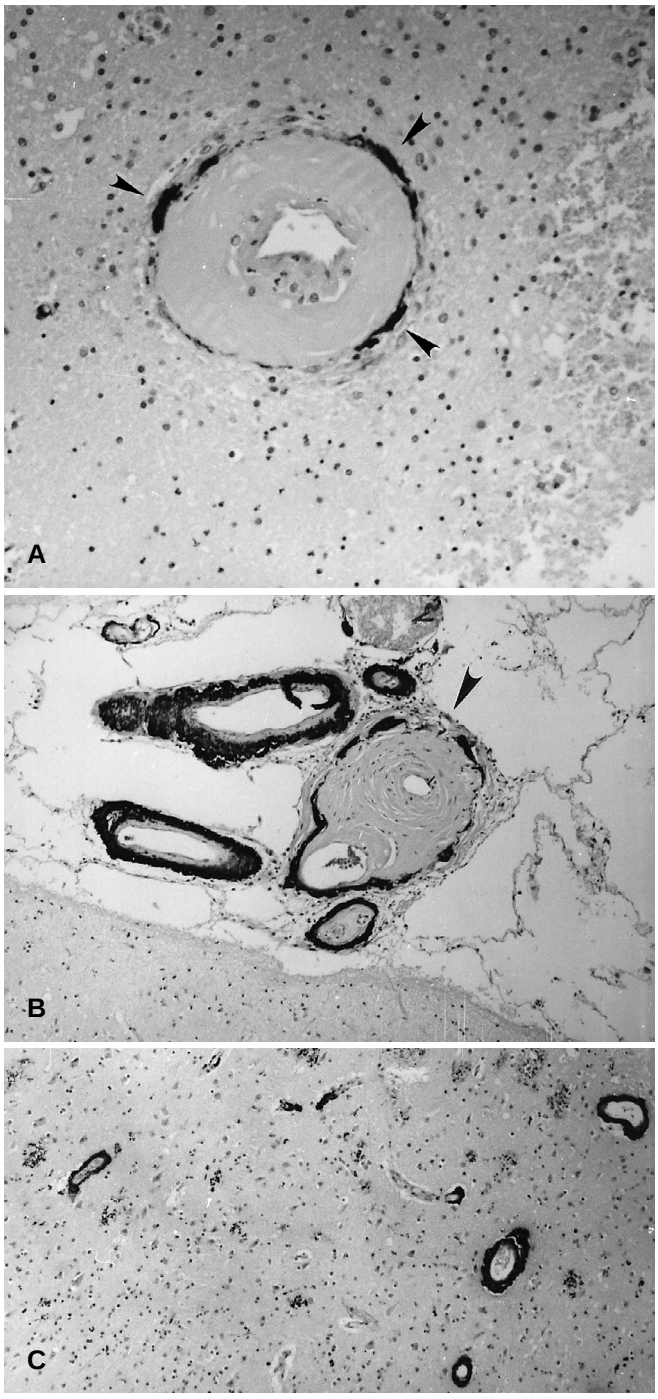

Fig. 3 A–C Relationship of hyaline arterial thickening to β/A4-immunoreactive material in vessel walls. **A** A prominently hyalinized parenchymal arteriole showing only patchy areas of β/A4 immunoreactivity *(arrows)* in vascular adventitia. **B** Meningeal arteries (in another patient), one of which *(arrowhead)* shows prominent hyaline thickening but only moderate adventitial β/A4 immunoreactivity. **C** Cortex underlying the region of meninges illustrated in **B**. Note prominent arteriolar β/A4 immunoreactivity and scattered diffusely stained plaque-like deposits. **A–C** Anti-β/A4 antibody. **A** × 156; **B**, **C** × 62

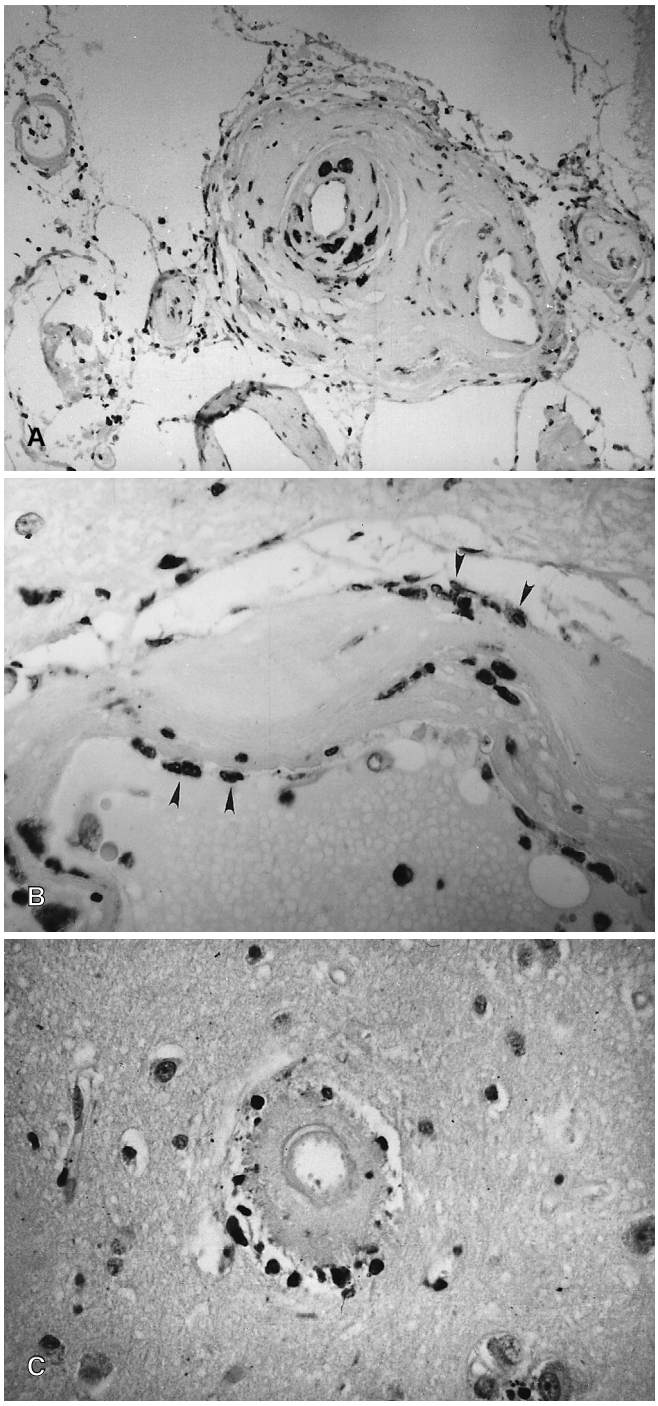

Fig. 4 A–C CD-68-immunoreactive macrophages in and immediately adjacent to markedly hyalinized arterial walls of HCHWA-D patients. **A** Meningeal artery. **B** Parenchymal artery. Note prominent vessel wall macrophages and immunoreactive cells adjacent to thickened wall on both its luminal *(lower arrows)* and adventitial *(upper arrows)* aspects. **C** Smaller cortical arteriole with thick amyloid wall surrounded by CD-68-immunoreactive cells (see also [21]). **A** × 190, **B** × 515, **C** × 505

scored three cases. Furthermore, all observers independently ranked the order of severity of the three 'test cases' identically. The greatest variability in CAA-AM scoring occurred in assessment of the presence of perivascular lymphocytes, hyalinization versus amyloid deposition in vessel walls, and thrombosis versus fibrinoid necrosis, especially since the latter two findings often occurred in

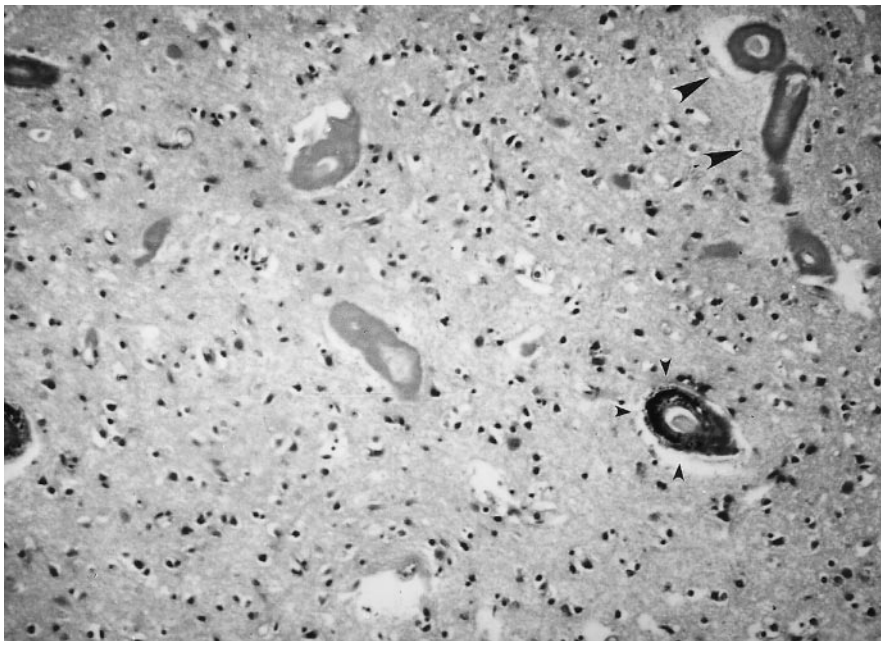

Fig. 5 Calcified arterioles in cortex of a patient with HCHWA-D. Pale vessels *(left of center)* show only walls thickened by amyloid. Vessels at *upper right (large arrowheads)* show moderate staining. Vessel indicated by *small arrowheads* shows dark granular staining in vessel wall, indicative of the presence of calcium. von Kossa stain. × 150

combination. Hyalinization could more easily be differentiated from amyloid deposition with the use of anti-β/A4 immunohistochemistry (see below). Subsequent immunohistochemical characterization of the perivascular lymphocytes found in association with CAA have shown virtually all of them to be labelled as T lymphocytes. A summary of the relative frequency of specific histopathological findings is presented in Table 1.

A prominent finding in many brains was the presence of vascular hyalinization or fibrosis. Immunohistochemical findings in such vessels showed that β/A4-immunoreactive material was present, if at all, in trace amounts in the vessel wall, and was usually localized to its adventitial aspect (Fig. 3). By contrast, many hyalinized vessels showed CD68-immunoreactive macrophages in their walls (Fig. 4). Arterioles infiltrated by amyloid but without significant secondary hyalinization often showed prominently immunolabelled macrophages on their adventitial aspect (Fig. 4C); these macrophages did not usually contain hemosiderin. In five cases, multinucleated foreign body-type giant cells were present around heavily amyloid-laden parenchymal and/or leptomeningeal arteries (Fig. 2B, C); in two patients, granulomatous inflammation around CAA arterioles was widespread and moderately severe.

Variable numbers of calcified CAA-affected arterioles were present in 6 of the 29 cases studied. Such vessels were detected on routine stains as having a basophilic wall. Cytochemical stains for iron and calcium in selected cases showed that these basophilic vessels had an abundance of calcium and less prominent stainable iron (Fig. 5).

Several of the individual histological features correlated quite strongly with each other, e.g., vascular hyalinization correlated with the presence of histiocytes in vessel walls (correlation coefficient $r = 0.80$) and microaneurysm formation ($r = 0.59$; P 0.001 for both). An analysis of clinical correlates and 'predictors' of given CAA-AM is currently underway.

While the aim of performing MIB-1 immunohistochemistry on brain sections with HCHWA-D was to detect proliferating cells within affected arterial walls (e.g., representing sm cell or fibroblast proliferation in relation to mural amyloid deposition), this was not found. However, interestingly several brain sections from the patient with a very severe degree of CAA-AM showed MIB-1 nuclear immunolabelling of scattered microvascular endothelial cells or cells in close apposition to capillary/microvessel walls (Fig. 6), although the affected vessel walls were not ones involved by CAA. In view of prolonged fixation times for many of the brain specimens, we are probably underestimating the true incidence of MIB-1-immunoreactive nuclei.

Discussion

CAA, in its sporadic, AD-associated and familial forms (i.e., HCCAA/HCHWA-I and HCHWA-D) is strongly associated with stroke in the form of primary non-traumatic ICH [11, 18, 20, 26, 34, 35, 41]; its association with ischemic infarcts is less clearly documented [1, 27]. Precise mechanisms of microvascular degeneration or precipitating systemic factors that result in ICH are poorly understood. The HCHWA-D form of CAA could potentially provide a 'window' on such mechanisms because of the predictably severe degree of CAA that results in all patients with the codon 693 amyloid precursor protein mutation, the consistent association of HCHWA-D with ICH (and probably ischemic changes), and the high frequency with which CAA-AM changes develop in the Dutch form of CAA. HCHWA-D is also a relatively 'pure' form of

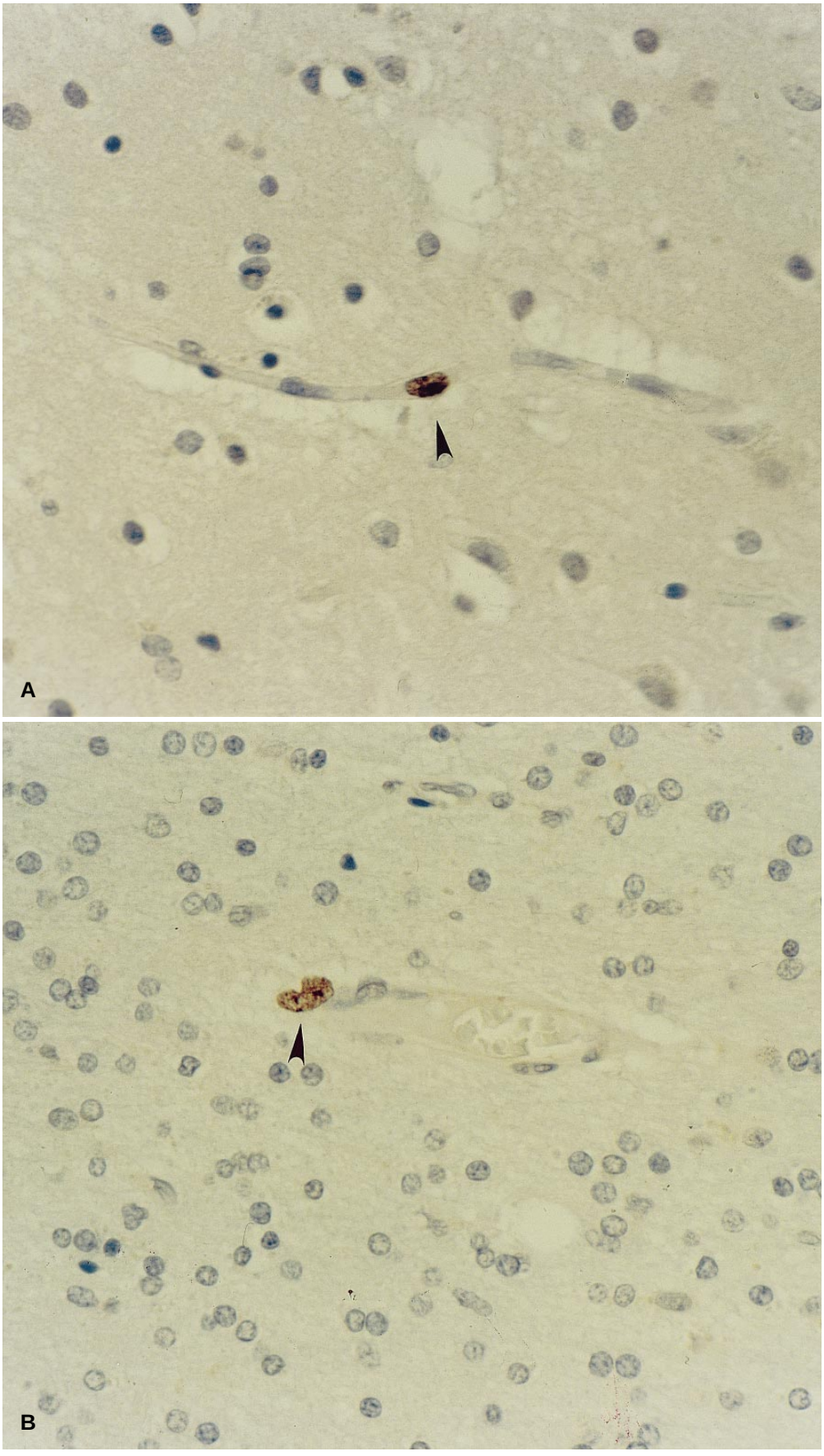

Fig. 6 MIB-1 immunoreactivity of nuclei of cells in or immediately adjacent to capillary (**A**) and venular (**B**) walls (from different brain regions of the same patient), indicated by *arrowheads*. **A**, **B** × 327

CAA in which the vast majority of cortical and leptomeningeal arterioles are usually affected, with less extensive parenchymal deposition of β/A4 (and a relative absence of neurofibrillary tangles) than is usually seen in association with CAA of AD [19–21]. Nevertheless, observations made in the study of HCHWA-D may have important implications for understanding the pathogenesis and complications of sporadic or AD-associated CAA.

Our results show that HCHWA-D is associated with both distinctive neuropathological complications, previously well documented in these patients [3, 11, 12, 20, 44, 45] and an accentuation of CAA-AM seen infrequently with AD-associated CAA. For instance, the calcification of amyloid-infiltrated arterioles in HCHWA-D appears to be almost unique to this form of CAA – we are aware of the phenomenon having been reported only recently in a patient with cerebral hemorrhage-associated CAA [22] but specifically have not observed CAA-associated vascular calcinosis in a large collective experience with AD-associated CAA. The pathogenesis of calcium deposition in amyloid-laden vessel walls appears to merit further investigation. Amyloid β/A4-protein is known to activate calcium-permeable channels in cultured rat hippocampal pyramidal neurons, where it thus has the potential to produce sustained elevation of intracellular calcium concentrations possibly leading to neuronal dysfunction [50]. Conceivably, comparable abnormalities of calcium homeostasis may occur in sm cells of the vessel walls to lead to its accumulation and eventual deposition in this location, or calcium salts may deposit within amyloid after sm cells have disappeared. In addition, the frequent presence of perivascular (and mural) blood breakdown products in CAA microvessels might act as a nidus for calcium deposition.

Vascular fibrosis/hyalinization is the most frequently identified form of CAA-AM. It is often noted in association with macrophage infiltration of vessel walls and microaneurysm formation. Although β/A4 peptide in HCHWA-D-affected vessel walls [29] may initiate vascular fibrosis with eventual aneurysm formation, little immunoreactive β/A4 was usually present in severely hyalinized vessels, with or without associated microaneurysm formation – an observation that has also been noted in cases of AD-related severe CAA [37]. This form of CAA-AM was seen in microvessels that were substantially devoid of β/A4 amyloid, although it is likely that the affected vessel walls contained amyloid *prior* to undergoing fibrosis. Macrophages surrounding CAA microvessels in the brain parenchyma [21], or circulating monocytes that migrate from the lumen into the vessel wall, may contribute to the vascular thickening. In this regard, CAA-related secondary microangiopathies may result from cellular and molecular mechanisms similar to those operative in the pathogenesis of atherosclerosis or even microvascular lipohyalinosis associated with hypertension and primary cerebral hemorrhage [28, 36]. In atherosclerosis, macrophage-derived factors are well known to injure endothelium and contribute to the formation of atherosclerotic fibrous plaques [28]. Mural thrombi, often seen in HCHWA-D, may also become organized to give the appearance of vessel wall fibrosis.

Observations in vitro and those in experimental animals will help to unravel the pathogenesis of CAA and CAA-AM. β/A4-amyloid has vasoactive properties and may mediate vascular endothelial damage [30]. β/A4 peptide derives from degenerating sm cells of arterial walls [17] in AD/SDAT brain and can be immunolocalized in sm and between sm cells in such specimens [9]. Vascular sm cells are also known to accumulate β/A4 peptide and undergo degeneration in aged canines [46]. In culture, wild-type β/A4 1–42 and the Dutch mutant form of β/A4 1–40 induce degeneration when applied to human pericytes and meningeal vascular sm cells [6, 33], and degenerating cells show pronounced increases in levels of cellular β/A4 APP [7, 8]. Cultured vascular sm from aged canines also secrete and accumulate β/A4 protein [10, 48]. A study of interactions between β/A4-immunoreactive cultured cerebrovascular sm and monocytes/macrophages in vitro may provide insights into the pathogenesis of secondary vascular scarring in CAA-AM.

CAA, and by inference CAA-AM, probably results from the interaction of several peptides, including β/A4, γ-trace, ubiquitin, and apolipoprotein E as well as less well characterized 'amyloidogenic' fragments of β/A4 [16, 31, 38, 39]. A final common pathophysiological pathway, however, appears to be sm cell degeneration leading to vessel wall weakening – a phenomenon also observed in HCHWA-I/HCCAA [43]. Determining the roles of various proteins in microvascular degeneration associated with CAA might also be more easily achieved in a tissue culture system or appropriate animal (including transgenic) models. Although fibrinoid necrosis of CAA vessels was identified in 6 of 29 specimens, it did not appear to be as strongly correlated with CAA-related brain hemorrhage as has been suggested in CAA of AD/SDAT [42].

The formation of multinucleated giant cells around amyloid-laden vessels, observed very infrequently in age- or AD/SDAT-associated CAA [2] was noted in 5 of the 29 HCHWA-D autopsy brains. In some cases, multinucleated giant cells were seen in close apposition to vascular amyloid without significant adjacent lymphocytes, whereas in the most severe instances lymphocytes (of T cell phenotype) were present. Studies in non-HCHWA-D cerebral amyloid angiopathies suggest that $CD4^+$ and $CD8^+$ T lymphocytes are a major component of the infiltrate in CAA-associated granulomatous inflammation; amyloid P-component (thought to prevent phagocytic proteolysis of amyloid β-protein) is sometimes absent [49]. Additional studies are being carried out on HCHWA-D-related cases of granulomatous inflammation to characterize its pathogenesis in this context.

In summary, CAA-AM that are infrequently (or unpredictably) found in AD- or age-related CAA are noted with high frequency in autosomal dominant HCHWA-D. We propose a scoring system whereby the severity of CAA-AM is assessed in a semiquantitative way, and suggest that this system may be used to evaluate the contribution of CAA-AM to CAA-related stroke in non-HCHWA-D patients.

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References

1. Anders KH, Secor DL, Vinters HV (1993) Cerebral amyloid angiopathy (CAA) associated with ischemic necrosis (abstract). J Neuropathol Exp Neurol 52 : 275
2. Anders KH, Wang ZZ, Kornfeld M, Gray F, Soontornniyomkij V, Reed LA, Hart MN, Menchine M, Secor DL, Vinters HV (1997) Giant cell arteritis in association with cerebral amyloid angiopathy: immunohistochemical and molecular studies. Hum Pathol 28 : 1237–1246
3. Bornebroek M, Haan J, Maat-Schieman MLC, Van Duinen SG, Roos RAC (1996) Hereditary cerebral hemorrhage with amyloidosis-Dutch type (HCHWA-D). I. A review of clinical, radiologic and genetic aspects. Brain Pathol 6 : 111–114
4. Bornebroek M, Haan J, Van Duinen SG, Maat-Schieman MLC, Van Buchem MA, Bakker E, Van Broeckhoven C, Roos RAC (1997) Dutch hereditary cerebral amyloid angiopathy: structural lesions and apolipoprotein E genotype. Ann Neurol 41 : 695–698
5. Coria F, Rubio I (1996) Cerebral amyloid angiopathies. Neuropathol Appl Neurobiol 22 : 216–227
6. Davis J, Van Nostrand WE (1996) Enhanced pathologic properties of Dutch-type mutant amyloid beta-protein. Proc Natl Acad Sci USA 93 : 2996–3000
7. Davis-Salinas J, Saporito-Irwin SM, Cotman CW, Van Nostrand WE (1995) Amyloid beta-protein induces its own production in cultured degenerating cerebrovascular smooth muscle cells. J Neurochem 65 : 931–934
8. Davis-Salinas J, Van Nostrand WE (1995) Amyloid beta-protein aggregation nullifies its pathologic properties in cultured cerebrovascular smooth muscle cells. J Biol Chem 270 : 20 887–20 890
9. Frackowiak J, Zoltowska A, Wisniewski HM (1994) Non-fibrillar beta-amyloid protein is associated with smooth muscle cells of vessel walls in Alzheimer disease. J Neuropathol Exp Neurol 53 : 637–645
10. Frackowiak J, Mazur-Kolecka B, Wisniewski HM, Potempska A, Carroll RT, Emmerling MR, Kim KS (1995) Secretion and accumulation of Alzheimer's beta-protein by cultured vascular smooth muscle cells from old and young dogs. Brain Res 676 : 225–230
11. Haan J, Roos RAC, Briët PE, et al (1989) Hereditary cerebral hemorrhage with amyloidosis-Dutch type. Clin Neurol Neurosurg 91 : 285–290
12. Haan J, Roos RAC, Algra PR, Lanser JBK, Bots GTAM, Vegter-Van der Vlis M (1990) Hereditary cerebral haemorrhage with amyloidosis-Dutch type. Magnetic resonance imaging findings in 7 cases. Brain 113 : 1251–1267
13. Haan J, Maat-Schieman MLC, Duinen SG van, Jensson O, Thorsteinsson L, Roos RAC (1994) Co-localization of beta/A4 and cystatin C in cortical blood vessels in Dutch, but not in Icelandic hereditary cerebral hemorrhage with amyloidosis. Acta Neurol Scand 89 : 367–371
14. Hendriks L, Broeckhoven C van (1996) The betaA4 amyloid precursor protein gene and Alzheimer's disease. Eur J Biochem 237 : 6–15
15. Jellinger K (1977) Cerebrovascular amyloidosis with cerebral hemorrhage. J Neurol 214 : 195–206
16. Kalaria RN, Premkumar DRD, Pax AB, Cohen DL, Lieberburg I (1996) Production and increased detection of amyloid beta protein and amyloidogenic fragments in brain microvessels, meningeal vessels and choroid plexus in Alzheimer's disease. Mol Brain Res 35 : 58–68
17. Kawai M, Kalaria RN, Cras P, Siedlak SL, Velasco ME, Shelton ER, Chan HW, Greenberg BD, Perry G (1993) Degeneration of vascular muscle cells in cerebral amyloid angiopathy of Alzheimer disease. Brain Res 623 : 142–146
18. Luyendijk W, Bots GTAM, Vegter-van der Vlis M, Went LN, Frangione B (1988) Hereditary cerebral haemorrhage caused by cortical amyloid angiopathy. J Neurol Sci 85 : 267–280
19. Maat-Schieman MLC, Rozemuller AJ, Duinen SG van, Haan J, Eikelenboom P, Roos RAC (1994) Microglia in diffuse plaques in hereditary cerebral hemorrhage with amyloidosis (Dutch). An immunohistochemical study. J Neuropathol Exp Neurol 53 : 483–491
20. Maat-Schieman MLC, Duinen SG van, Bornebroek M, Haan J, Roos RAC (1996) Hereditary cerebral hemorrhage with amyloidosis-Dutch type (HCHWA-D). II. A review of histopathological aspects. Brain Pathol 6 : 115–120
21. Maat-Schieman MLC, Duinen SG van, Rozemuller AJM, Haan J, Roos RAC (1997) Association of vascular amyloid beta and cells of the mononuclear phagocyte system in hereditary cerebral hemorrhage with amyloidosis (Dutch) and Alzheimer disease. J Neuropathol Exp Neurol 56 : 273–284
22. Mackenzie IRA (1997) Cerebral amyloid angiopathy with extensive mineralization. Clin Neuropathol 16 : 209–213
23. Mandybur TI (1986) Cerebral amyloid angiopathy: the vascular pathology and complications. J Neuropathol Exp Neurol 45 : 79–90
24. Maury CPJ (1995) Molecular pathogenesis of beta-amyloidosis in Alzheimer's disease and other cerebral amyloidoses. Lab Invest 72 : 4–16
25. Okazaki H, Reagan TJ, Campbell RJ (1979) Clinicopathologic studies of primary cerebral amyloid angiopathy. Mayo Clin Proc 54 : 22–31
26. Ólafsson Í, Thorsteinsson L, Jensson Ó (1996) The molecular pathology of hereditary cystatin C amyloid angiopathy causing brain hemorrhage. Brain Pathol 6 : 121–126
27. Olichney JM, Hansen LA, Hofstetter CR, Grundman M, Katzman R, Thal LJ (1995) Cerebral infarction in Alzheimer's disease is associated with severe amyloid angiopathy and hypertension. Arch Neurol 52 : 702–708
28. Ross R (1993) The pathogenesis of atherosclerosis: a perspective for the 1990s. Nature 362 : 801–809
29. Rozemuller AJM, Roos RAC, Bots GTAM, Kamphorst W, Eikelenboom P, Van Nostrand WE (1993) Distribution of beta/A4 protein and amyloid precursor protein in hereditary cerebral hemorrhage with amyloidosis-Dutch type and Alzheimer's disease. Am J Pathol 142 : 1449–1457
30. Thomas T, Thomas G, McLendon C, Sutton T, Mullan M (1996) Beta-Amyloid-mediated vasoactivity and vascular endothelial damage. Nature 380 : 168–171
31. Uchida K, Kuroki K, Yoshino T, Yamaguchi R, Tateyama S (1997) Immunohistochemical study of constituents other than beta-protein in canine senile plaques and cerebral amyloid angiopathy. Acta Neuropathol 93 : 277–284
32. Van Broeckhoven C, Haan J, Bakker E, Hardy JA, Van Hul W, Wehnert A, Vegter-Van der Vlis M, Roos RAC (1990) Amyloid beta protein precursor gene and hereditary cerebral hemorrhage with amyloidosis (Dutch). Science 248 : 1120–1122
33. Verbeek MM, Waal RMW de, Schipper JJ, Van Nostrand WE (1997) Rapid degeneration of cultured human brain pericytes by amyloid beta protein. J Neurochem 68 : 1135–1141
34. Vinters HV (1987) Cerebral amyloid angiopathy. A critical review. Stroke 18 : 311–324
35. Vinters HV (1990) Amyloid and the central nervous system: the neurobiology, genetics and immunocytochemistry of a process important in neurodegenerative diseases and stroke. In: Cancilla PA, Vogel FS, Kaufman N (eds) Neuropathology. U.S. and Canadian Academy of Pathology, Monograph Series. Williams & Wilkins, Baltimore, pp 55–85
36. Vinters HV, Mah V (1991) Vascular disease. In: Duckett S (ed) The pathology of the aging human nervous system. Lea & Febiger, Philadelphia, pp 20–76

37. Vinters HV, Pardridge WM, Secor DL, Ishii N (1988) Immunohistochemical study of cerebral amyloid angiopathy. II. Enhancement of immunostaining using formic acid pretreatment of tissue sections. Am J Pathol 133:150–162
38. Vinters HV, Nishimura GS, Secor DL, Pardridge WM (1990) Immunoreactive A4 and gamma-trace peptide colocalization in amyloidotic arteriolar lesions in brains of patients with Alzheimer's disease. Am J Pathol 137:233–240
39. Vinters HV, Secor DL, Pardridge WM, Gray F (1990) Immunohistochemical study of cerebral amyloid angiopathy. III. Widespread Alzheimer A4 peptide in cerebral microvessel walls colocalizes with gamma trace in patients with leukoencephalopathy. Ann Neurol 28:34–42
40. Vinters HV, Secor DL, Read SL, Frazee JG, Tomiyasu U, Stanley TM, Ferreiro JA, Akers M-A (1994) Microvasculature in brain biopsy specimens from patients with Alzheimer's disease: an immunohistochemical and ultrastructural study. Ultrastruct Pathol 18:333–348
41. Vinters HV, Wang ZZ, Secor DL (1996) Brain parenchymal and microvascular amyloid in Alzheimer's disease. Brain Pathol 6:179–195
42. Vonsattel JPG, Myers RH, Hedley-Whyte ET, Ropper AH, Bird ED, Richardson EP Jr (1991) Cerebral amyloid angiopathy without and with cerebral hemorrhages: a comparative histological study. Ann Neurol 30:637–649
43. Wang ZZ, Jensson O, Thorsteinsson L, Vinters HV (1997) Microvascular degeneration in hereditary cystatin C amyloid angiopathy of the brain. APMIS 105:41–47
44. Wattendorff AR, Bots GThAM, Went LN, Endtz LJ (1982) Familial cerebral amyloid angiopathy presenting as recurrent cerebral haemorrhage. J Neurol Sci 55:121–135
45. Wattendorff AR, Frangione B, Luyendijk W, Bots GTAM (1995) Hereditary cerebral haemorrhage with amyloidosis, Dutch type (HCHWA-D): clinicopathological studies. J Neurol Neurosurg Psychiatry 58:699–705
46. Wegiel J, Wisniewski HM, Dziewiatkowski J, Tarnawski M, Nowakowski J, Dziewiatkowska A, Soltysiak Z (1995) The origin of amyloid in cerebral vessels of aged dogs. Brain Res 705:225–234
47. Wisniewski HM, Fraçkowiak J, Zóltowska A, Kim KS (1994) Vascular beta-amyloid in Alzheimer's disease angiopathy is produced by proliferating and degenerating smooth muscle cells. Int J Exp Clin Invest 1:8–16
48. Wisniewski HM, Frackowiak J, Mazur-Kolecka B (1995) In vitro production of beta-amyloid in smooth muscle cells isolated from amyloid angiopathy-affected vessels. Neurosci Lett 183:120–123
49. Yamada M, Itoh Y, Shintaku M, Kawamura J, Jensson Ó, Thorsteinsson L, Suematsu N, Matsushita M, Otomo E (1996) Immune reactions associated with cerebral amyloid angiopathy. Stroke 27:1155–1162
50. Ye C, Ho-Pao CL, Kanazirska M, Quinn S, Rogers K, Seidman CE, Seidman JG, Brown EM, Vassilev PM (1997) Amyloid-beta proteins activate Ca^{2+}-permeable channels through calcium-sensing receptors. J Neurosci Res 47:547–554