4. Amyloid B/A4 Peptide Associated with Alzheimer’s Disease and Cerebral Amyloid Angiopathy

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Key words: amyloid B protein, Alzheimer’s disease, cerebral amyloid angiopathy, senile plaque

We reviewed the recent progress in cerebral amyloid research, especially on the amyloid B/A4 (Aβ) peptide from results in our laboratory and those reported in the literature. Cerebral amyloid deposition is an extracellular deposition in the brain and brain blood vessels of insoluble aggregates of the Aβ peptide. Isolation and sequencing of the peptide from senile plaques and cerebrovascular amyloid deposits has led to the elucidation of the etiology and pathogenesis of Alzheimer’s disease (1, 2). The Aβ peptide, about 40 amino acids in length (3), is a breakdown product of a much larger protein, the amyloid B/A4 protein precursor (APP) (2). APP exists mainly in three isoforms that result from alternative splicing. APP is a glycosylated transmembrane protein with a large extracellular domain, a single transmembrane domain and a small cytoplasmic domain; the Aβ fragment composes a part of the transmembrane domain and a short stub of the extracellular domain.

First, cerebral amyloid was examined by immunohistochemical staining using an antiserum to synthetic Aβ peptide (1-28). The Aβ peptide was closely associated with cerebrovascular and senile plaque amyloid deposition (Fig. 1); also, another type of “diffuse plaque”, in addition to “classic”, “primitive”, and “compact (burned-out)” plaques, was demonstrated (4). Diffuse (very primitive) plaques of the cerebral cortex and cerebellum were seen as ill-defined areas of fine fibrillar materials labeled by Aβ peptide immunostaining and silver impregnation. Electron and immunogold electron microscopic investigation revealed that the diffuse plaques consist of accumulations of small bundles of amyloid fibrils or non-fibrillar materials (preamyloid) scattered among the normal-appearing neurites, including a few altered neurites (5). The Aβ peptide deposits are also seen in the subpial area, subcortical white matter and in some eosinophilic tangles which represent the end stage of neurofibrillary tangles (6).

Although Aβ peptide has been considered to be deposited only in the brain, Aβ peptide deposits have been reported in the skin tissue and intestines of Alzheimer’s disease patients and normal elderly humans (7). There are multiple initiating mechanisms (Alzheimer’s disease, aging, Down’s syndrome, mutation in APP, and trauma) in the production of excess amyloid deposition.

Various antisera against synthetic peptides to APP and against recombinant APP (Boehringer) were used to study the immunohistochemical distribution of APP. In normal mice, rats and humans, the immunoreactivities are recognized in almost all neurons and some glia cells of both central and peripheral nervous systems (8). Here, accumulations of APP-positive swollen neurites were seen in the classic and primitive plaques.

Fig. 1. A variety of senile cerebral amyloid deposits (senile plaques, subpial deposits and cerebral amyloid angiopathy) are positive for antiserum to a synthetic Aβ peptide (1-28) (×100).

Fig. 2. Swollen neurites in senile plaques and neurons are positive for antiserum to amyloid precursor protein (Boelinger, monoclonal, ×250).

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Moreover, APP-positive swollen neurites were seen in aged or lesioned rats and in/or near the lesions of cerebral infarction. The APP was not found in the amyloid core or cerebral amyloid angiopathy (CAA). The accumulation of APP in the swollen neurites may be caused by the reduced axonal transport. APP has been suggested to be related to growth regulation, neurite outgrowth, cell-to-cell and cell-matrix interactions. The Aβ peptide has been demonstrated to have trophic and toxic effects on neurons in vitro (9, 10). However, the exact physiological functions of APP and the Aβ peptide have not yet been disclosed.

Recently, it was reported that the cell normally produces and releases a soluble 4-kilodalton (kD) Aβ peptide (11, 12). Shoji et al (12) demonstrated that human neuroblastoma (M17) cells transfected with constructs expressing full-length APP and M17 cells which express only endogenous APP, also release soluble 4-kD Aβ peptide. Similar fragments were also detected in the cerebrospinal fluid (CSF) from individuals with Alzheimer’s disease and normal subjects, but, there was no difference in the Aβ peptide contents in the CSF between Alzheimer’s disease patients and control subjects. There seems to be two major pathways for APP metabolism as shown in Fig. 3. The first, “secretase” pathway (13), is cut off within the Aβ portion of APP which occurs on the cell surface. The second, the “endosome/lysosome” pathway, is potentially amyloidogenic (11, 12).

Analysis of a large number of familial Alzheimer’s disease cases has shown them to be genetically heterogenous (14, 15). However, recent DNA sequence analyses of the APP have revealed six distinct missense mutations within or just outside the Aβ peptide regions in several families exhibiting autosomal dominant transmission of Alzheimer’s disease (Fig. 4). These findings strongly suggest that some cases of familial Alzheimer’s disease are caused by a mutation in the APP gene (14, 15).

α1-antichymotrypsin (ACT), a serum glycoprotein which is regarded to be an acute-phase protein, has been shown to be one of the components of senile plaque amyloid (16). Almost all senile plaques, labeled by Aβ peptide immunostaining, were immunoreactive with anti-ACT antiserum (17). Serum and CSF levels of ACT have been demonstrated to be significantly and specifically higher in Alzheimer’s disease patients than in other subjects (16).

Cerebral amyloid angiopathy (CAA) is most commonly associated with normal aging, Alzheimer’s disease, Down’s syndrome and Dutch-type of cerebral hemorrhage with amyloidosis (HCHWA-D). HCHWA-D was shown to have a point mutation in the APP gene (Fig. 4). Amyloid fibrils in the Iceland-type of hereditary cerebral hemorrhage with amyloidosis (HCHWA-I) are composed of a variant of cystatin C. In a case of CAA showing dual immunohistochemical reactivity with antibodies to both Aβ peptide and cystatin C, it was demonstrated that cystatin C is not an intrinsic component of amyloid fibrils in this type of CAA (18). CAA associated with granulomatous angitis of the central nervous system (GANS) has rarely been reported. The relationship between CAA and GANS is not clear. However, in the present case, some giant cells showed inclusions which were positive for the Aβ peptide and/or APP by immunostaining (19).

References


5. New Type of Amyloidosis

a) β2-Microglobulin and Hemodialysis

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Key words: β2-microglobulin, hemodialysis, 131I-β2-M scintigraphy, adsorption column

Introduction

Dialysis amyloidosis is a serious complication encountered currently in chronic hemodialysis. In 1985 we determined that the principal amyloid protein is β2-microglobulin (β2-M) (1). Later β2-M associated amyloidosis has been known to invade mainly the synovial membrane of chronic hemodialysis patients and to cause a unique osteoarthropathy such as carpal tunnel syndrome (CTS), trigger finger, bone cyst and destruutive spondyloarthropathy (2, 3). Recently, β2-M amyloid has been noticed in the tongue, gastrointestinal tract, liver, heart, lung, prostate and adrenal glands, showing that this amyloidosis is a systemic one.

This amyloidosis is conventionally diagnosed by examining tissue specimens obtained at biopsy and by a positive response to Congo-red stain or direct observation of amyloid fibrils on electron microscopy is confirmatory. The non-invasive detection of the amyloid deposits has recently become possible using scintigraphy with radiolabelled amyloid precursors protein (131I-β2-M) (4).

It has now been clearly established that β2-M is amyloidogenic and the elimination of its causal substance is the most important problem in the therapy. It is well known that a large amount of β2-M is accumulated and the serum β2-M concentration reaches a 50-fold or higher levels in patients on chronic hemodialysis, compared with normal subjects (5). While recent attempts include the use of high-flux membrane dialyzers in performing HD (hemodialysis) or HDF (hemodiafiltration) to eliminate β2-M, there is inevitably a limit to the β2-M extraction capacity. In this situation, our group conducted a clinical application of a β2-M adsorption column (BM-01).

We report the results of our studies on the clinical features, 131I-β2-M scintigraphy and β2-M adsorption column of dialysis-related amyloidosis.

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