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Complement activation by β-amyloid in Alzheimer disease

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ABSTRACT **Alzheimer disease (AD) is characterized by excessive deposition of the β-amyloid peptide (β-AP) in the central nervous system. Although several lines of evidence suggest that β-AP is neurotoxic, a mechanism for β-AP toxicity in AD brain remains unclear. In this paper we provide both direct *in vitro* evidence that β-AP can bind and activate the classical complement cytolytic pathway in the absence of antibody and indirect *in situ* evidence that such actions occur in the AD brain in association with areas of AD pathology.**

Alzheimer disease (AD) is characterized by excessive central nervous system (CNS) deposition of the β-amyloid peptide (β-AP), a 40- to 42-amino acid peptide derived from a larger amyloid precursor protein (APP) (1–3). Although no specific mechanism of β-AP deposition has yet been formally proven, there are several lines of evidence (4–6) that, once generated, β-AP causes direct or indirect toxicity to CNS neurons. Proposed mechanisms of AD neurotoxicity include membrane changes (7), alterations in Ca^{2+} homeostasis (6, 8), excitotoxicity (5, 6), and disruption of cytoskeletal or axon transport systems (9, 10). However, no single AD pathogenetic mechanism has yet achieved a wide consensus of acceptance.

In addition to studies of β-AP, over the last decade a number of investigators have noted that the AD brain exhibits many of the classical markers of immune-mediated damage. These include elevated numbers of major histocompatibility complex class I- and II-immunoreactive microglia (cells believed to be an endogenous CNS form of the peripheral macrophage) (11–15) and astrocytes expressing interleukin 1 (16) and α_1-antichymotrypsin (17) (both acute phase reactants). Of particular importance, complement proteins of the classical pathway have been immunohistochemically detected in the AD brain (12, 13, 18–20), and we have noted that they most often appear associated with β-AP-containing pathological structures such as senile plaques. Proteins specific to the alternative pathway do not appear to be present (12, 13, 18). The first step in the classical complement pathway entails binding of the C1q component of C1, with subsequent activation of the C1r and C1s components. This is followed by a complex series of autocatalytic reactions, proceeding through C4, C2, and C3, and culminating in formation of the membrane attack complex (MAC), C5b–9. The MAC inserts a lytic plug in adjacent cell membranes, mediating cellular toxicity (21). Although C1q binding to the Fc region of immunoglobulins is the most common mechanism for initiating the classical pathway, several substrates—including viruses, parasites, and mannan-binding protein—have also been demonstrated to activate C1 and to do so in an antibody-independent fashion (22). In this paper we present six converging lines of evidence suggesting that β-AP activates the classical pathway complement cascade without mediation by immunoglobulin. This previously unrecognized mechanism may contribute significantly to the neurotoxicity of β-AP as well as to the pathophysiology of neuronal dysfunction characteristic of AD.

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MATERIALS AND METHODS

Human Brain Samples. Brain materials were obtained at autopsy from volunteer AD patients and nondemented elderly (ND) controls through the Sun Health Research Institute Tissue Donation Program. Postmortem intervals were well matched and did not exceed 3.5 hr in any case. Samples of the superior frontal gyrus, hippocampal complex, amygdala, brain stem, and cerebellum were dissected as approximately 1-cm^3 blocks, postfixed in ice-cold 4% (wt/vol) paraformaldehyde (0.1 M sodium phosphate buffer, pH 7.4) for 16–24 hr, cryoprotected in 2% dimethyl sulfoxide/10% glycerol (vol/vol) followed by 2% dimethyl sulfoxide/20% glycerol (in 0.1 M phosphate buffer, pH 7.4) for 48 hr each, sectioned at 20 or 40 μm on a freezing microtome, and stored at −20°C in a cryopreservation solution composed of 33% glycerol, 33% polyethylene glycol, and 33% 0.1 M phosphate buffer, pH 7.4 (vol/vol).

Immunohistochemistry. Sections were removed from glycol storage and washed six times for 15 min each in TBS (0.01 M Tris·HCl, pH 7.6/0.09 M NaCl). Endogenous peroxidase was blocked by incubation for 5 min with 0.3% H_2O_2/50% methanol (vol/vol) in TBS, followed by three 15-min rinses in TBS/0.05% Triton X-100. We then blocked for 1 hr in 3% bovine serum albumin (BSA) in TBS/0.05% Triton X-100. Although it does not materially affect the results to block with normal serum, we did not do so because of the possibility that normal serum might add exogenous complement proteins, contaminating anti-complement immunohistochemistry. Sections were incubated overnight with primary antibody at 4°C with gentle agitation, washed three times for 15 min each with TBS/0.05% Triton X-100, incubated for 1 hr with 1:50 biotin-conjugated secondary antibody (Vector Laboratories), and allowed to react with diaminobenzidine (DAB) as in standard ABC/DAB immunohistochemistry (Vector Laboratories). Primary and secondary antibodies were diluted in TBS/0.7% λ carrageenan/0.5% Triton X-100/0.2% sodium azide. For double-label immunohistochemistry, sections were next washed three times for 15 min each in TBS, incubated overnight with the second primary antibody at 4°C (gentle agitation), washed three times for 15 min each in TBS/0.05% Triton X-100, incubated for 30 min with second-

Abbreviations: AD, Alzheimer disease; β-AP, β-amyloid peptide; APP, amyloid precursor protein; APP_S 751, C-terminal truncated, secreted form of the 751-residue APP; CNS, central nervous system; MAC, membrane attack complex; ND, nondemented elderly; BSA, bovine serum albumin; TGF, transforming growth factor.

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ary antibody (Vector Laboratories), and allowed to react as in standard ABC-alkaline phosphatase (AP) immunohistochemistry (Vector Laboratories), including a levamisole blocking step. Finally, the sections were counterstained with thioflavin S and mounted. Preabsorption with appropriate antigens (when available), deletion of primary antibody, and absence of staining in ND patients were always employed as negative controls.

Dot Blots for C1q Binding. Samples (200 μl) of 5 μM β-AP fragments β-AP-(1–38) and β-AP-(1–28), the 751-residue APP secreted form (APP_S 751) (11), or various control peptides [here, BSA and the transmembrane domain of transforming growth factor (TGF)] in TBS were blotted onto Westran polyvinylidene difluoride (PVDF) membrane (Schleicher & Schuell). The membrane was rinsed in TBS, blocked for 1 hr in TBS plus 5% nonfat powdered milk, washed, and incubated with human C1q (Quidel) at 10 μg/ml for 2 hr. The membrane was then washed, incubated with rabbit antibodies to human C1q (DAKO, Carpinteria, CA) diluted 1:1000 in TBS plus 5% nonfat powdered milk overnight at 4°C, washed in TBS, incubated in biotinylated goat antibodies to rabbit IgG (Vector Laboratories) at a dilution of 1:50 in TBS plus 5% nonfat powdered milk for 2 hr, washed, and processed according to the ABC/DAB method (Vector Laboratories). All washes were five times for 5 min each, and all incubations and washes were performed with gentle agitation. C1q was deleted on adjacent blots to control for nonspecific immunoreactivity of peptides with the primary and secondary antibodies.

CH_{50} Assay for Complement Activation. The CH_{50} assay is a standard complement activation assay that has been widely used for over a decade. The assay performed here is thoroughly described by Mayer (23). Test solutions containing β-AP-(1–38) added to normal serum at 125, 250, and 500 μg/ml were employed. The results with β-AP-(1–38) were referenced to the normal serum vehicle without β-AP-(1–38).

ELISA for Complement Activation. Samples (100 μl each, all 5.0 μM) of BSA, TGF (transmembrane domain), APP_S 751 (24), β-AP-(1–38), β-AP-(1–28), β-AP-(1–16), β-AP-(17–28), β-AP-(24–35), and β-AP-(10–28) were plated in a 96-well ELISA plate, blocked with 1% BSA/1% powdered milk in 10 mM sodium phosphate, pH 7.4/0.9 mM NaCl (PBS), washed once with PBS/1% BSA, and incubated 40 min at 37°C with 50 μl of fresh normal human serum diluted 1:20 in PBS. Wells were then washed 10 times with PBS/1% BSA, incubated 1 hr at 37°C with 100 μl of a mouse monoclonal anti-C3b antibody (Quidel, San Diego) at 300 ng/ml, washed 10 times with PBS, and incubated 30 min at 37°C with horseradish peroxidase-conjugated goat antibodies to mouse IgG (Chemicon). To each well, 100 μl 2,2′-azinobis(3-ethylbenzthiazolinesulfonic acid) (ABTS)/peroxidase substrate (Kirkegaard and Perry Laboratories, Gaithersburg, MD) was added at room temperature. Optical densities (ODs) of wells at 405 nm were recorded at 2, 6, and 8 min. ODs of wells from which the test peptides had been deleted were subtracted to control for nonspecific background reactivity. Wells from which the primary antibody had been deleted gave only background measures.

RESULTS

C1q Immunoreactivity Colocalizes with β-AP-Containing AD Pathological Structures. Fig. 1 shows typical immunoreactivity for a rabbit antiserum directed against human C1q in AD and ND superior frontal gyrus. In ND patients, no specific staining is observed (Fig. 1*A*), whereas under the same conditions profuse labeling of numerous large roughly spherical structures is revealed in AD samples (Fig. 1*B*). Thioflavin counterstaining (Fig. 1*B Inset*) demonstrates that these C1q immunoreactive structures are senile plaques, a pathological hallmark of AD associated with β-AP deposits. Some thioflavin-positive neurofibrillary tangles also colocalize with C1q, as has been previously reported (12, 13, 20). These results are representative, without exception, of findings in 20 AD and 10 ND patients. Moreover, C1q colocalization with sites of β-AP-containing structures or β-AP immunoreactivity is equally evident in such brain areas as temporal cortex, amygdala, and hippocampus, a result that has been replicated, without exception, in 20 AD patients.

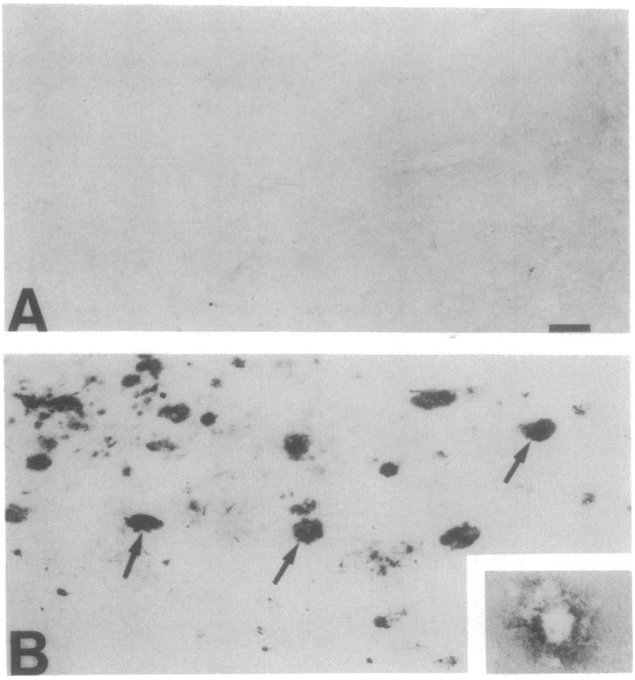

FIG. 1. Typical immunoreactivity in superior frontal gyrus of clinically and neuropathologically confirmed ND (*A*) and AD (*B*) patients when a rabbit antiserum directed against human C1q (1:1000 dilution) was used. In AD, staining is almost exclusively limited to profiles with the morphology and distribution of senile plaques (arrows), as well as some neurofibrillary tangles. Virtually all thioflavin-positive plaques are positive for C1q immunoreaction product (*Inset*) (fluorescence plus dim bright-field optics). (Calibration bar equals 50 μm.)

C1q Immunoreactivity Does Not Colocalize with Immunoglobulin Immunoreactivity. Although immunoglobulin has been reported to occur in AD brain tissue (13, 18, 19, 25), it does not colocalize with C1q immunohistochemically (Fig. 2). Rather, immunoglobulin immunoreactivity occurs on scattered clusters of neurons not associated with senile plaques (Fig. 2*A*), and C1q immunoreactivity occurs in the context of β-AP-containing structures such as senile plaques (Figs. 1*B* and 2*B*). These findings have been repeated in 15 AD patients with seven different anti-immunoglobulin antibodies or Fab fragments (e.g., 1:500 sheep anti-human IgG Fc from Bioproducts for Science, Indianapolis; 1:500 sheep anti-human IgG Fab from Bioproducts for Science; 1:2000 goat $F(ab')_2$ anti-human IgG from Cappel Laboratories). The fact that immunoreactive immunoglobulin can be detected at all in these studies is important, since it shows that the failure to observe immunoglobulin in association with C1q is not the result of inadequate techniques. Much of the immunoglobulin staining in brain has been suggested to be an artifact due to postmortem vascular changes (26). Our studies tend to confirm this hypothesis. Immunoglobulin immunoreactivity similar to that of the representative AD patient in Fig. 2*B* is equally apparent in nondemented young adult patients (S.D.S. and J.R., unpublished data). The staining is patchy throughout all areas of brain, tending to occur diffusely in

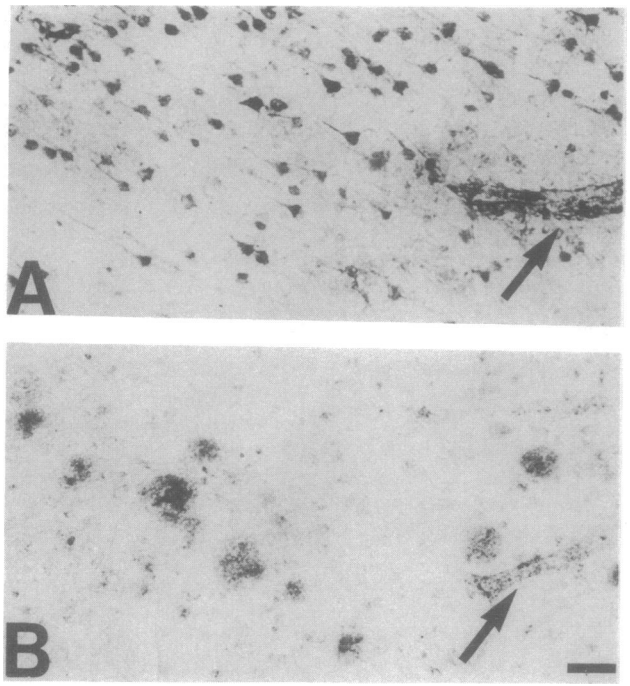

FIG. 2. Serial sections of AD anterior cingulate gyrus that have reacted with a polyclonal antiserum directed against human immunoglobulin (*A*) or a monoclonal antibody directed against C1q (*B*). Immunoglobulin and C1q immunoreactivities do not colocalize here or in other similar sections. For orientation, arrows indicate a blood vessel in the two adjacent sections. (Calibration bar equals 60 μm.)

clusters several hundred μm in diameter that are centered on or near large blood vessels. By contrast, C1q immunoreactivity is evident only in areas of brain that feature significant β-AP deposition, such as association cortex and limbic system, and colocalization is to sites of β-AP deposition or β-AP immunoreactivity, not necessarily to blood vessels. It is therefore most parsimonious to conclude that the localization of C1q to β-AP-containing AD pathological sites is antibody independent.

β-AP Binds C1q *in Vitro*. Dot blots of immobilized β-AP-(1–38), β-AP-(1–28), and APP_S 751 appear to bind physiological concentrations of C1q *in vitro* (Fig. 3). The result with APP_S 751 is interesting since this peptide contains β-AP-(1–15) as its carboxyl terminus. Similar results have been obtained in four different replicate experiments in the laboratory of J.R., as well as in experiments performed in the laboratory of P.L.M.

β-AP Activates the Classical Complement Pathway in CH_{50} Assays. Colocalization of complement proteins with β-AP-containing AD structures does not necessarily prove complement activation nor does it provide a mechanism for such activation. To test for the latter, a standard *in vitro* assay of complement activation, CH_{50}, was employed (23). Referenced to the percent activity of normal serum, the CH_{50} remaining at β-AP-(1–38) concentrations of 125, 250, and 500 μg/ml was 85.0%, 45.8%, and 0.0%, respectively. This antibody-independent complement activation is approximately 10-fold greater than that for equivalent amounts of the oncornaviruses rat leukemia virus, primate Rauscher leukemia virus, Rauscher leukemia virus, and Moloney leukemia virus under similar assay conditions (22). These results have been confirmed in three different replicate experiments.

β-AP Activates the Classical Complement Pathway in ELISAs. The CH_{50} data were confirmed and extended by using an ELISA for measurement of complement activation. This assay is based on detection of newly formed C3b after incubation of test peptides with normal serum. Results are

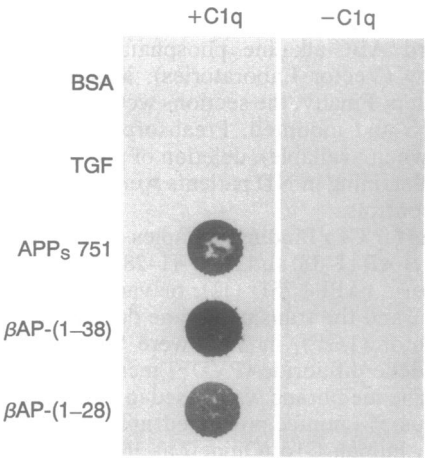

FIG. 3. C1q binding to BSA, the transmembrane domain of TGF, APP_S 751, β-AP-(1–38), and β-AP-(1–28). C1q was deleted from the right lane to control for nonspecific reactivity of the C1q antibody and detection system. Untreated membranes and control peptides—here, BSA and TGF—do not show detectable C1q binding, whereas peptides containing, as a minimum, the β-AP-(1–15) sequence—here, APP_S 751, β-AP-(1–38), and β-AP-(1–28)—do. C1q was obtained from Quidel and BSA (fraction V) was obtained from Sigma. APP_S 751, β-AP fragments, and TGF were the generous gift of Athena Neurosciences (San Francisco).

shown in Fig. 4. Essentially background measures of complement activation were observed with BSA, TGF (transmembrane domain), and APP_S 751, whereas activation by β-AP-(1–38) and β-AP-(1–28) was as much as 4- to 5-fold higher. Interestingly, β-AP fragments containing all or part of the first 16 β-AP amino acid residues [e.g., β-AP-(1–28), -(1–16), -(10–28)] activated complement, whereas β-AP fragments not containing these residues [e.g., β-AP-(17–28), -(24–35)] did not. Interactions with the alternative pathway are unlikely to explain these results: (*i*) alternative pathway activation is weak to absent at the 1:20 serum dilutions used in the present experiments; and (*ii*) activation is abolished when MgEGTA-treated serum is used—a condition wherein alternative pathway activation can occur, but classical pathway activation cannot. These data were replicated over three different assays. Measurement of ODs at time points other than those shown in Fig. 4 gave lower or higher values depending on length of incubation, but the pattern of results remained the same. In summary, these data show that β-AP and β-AP fragments containing all or part of residues 1–16 can functionally activate the classical pathway complement cascade without mediation by immunoglobulin.

β-AP Activates the Full Classical Pathway Complement Cascade *in Vivo*. Like that for C1q, immunoreactivity for other complement proteins occurs in the context of β-AP-containing AD pathological structures (Fig. 5). For example, serial sections through plaques stained by using antibodies directed against C1q and C4d coupled with thioflavin histochemistry reveal that C1q and C4d immunoreactivities colocalize with thioflavin-positive plaques (Fig. 5 *A–C*). Because attachment of C4d requires full activation of the C1 complex (8), these *in situ* data strongly support the *in vitro* observation that β-AP functionally activates complement. Similar results can be shown for the final steps of complement activation, including the MAC, C5b–9 (Fig. 5 *D* and *E*), indicating that β-AP not only activates complement but may lead to complement-mediated pathogenesis at sites of β-AP deposition.

DISCUSSION

The converging evidence presented here strongly supports the hypothesis that β-AP can directly activate complement in

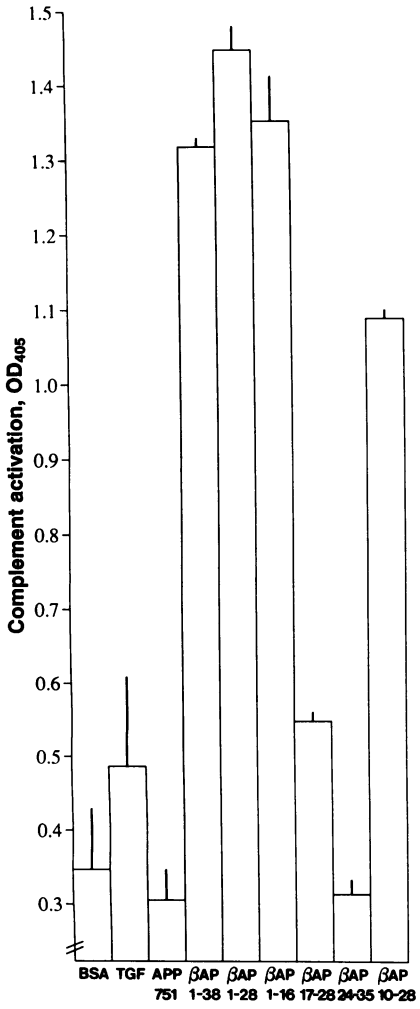

FIG. 4. Complement activation by β-AP and related peptides in an ELISA. Mean ODs (± SEM) at 6 min, reflecting formation of the activated C3 fragment C3b, are shown.

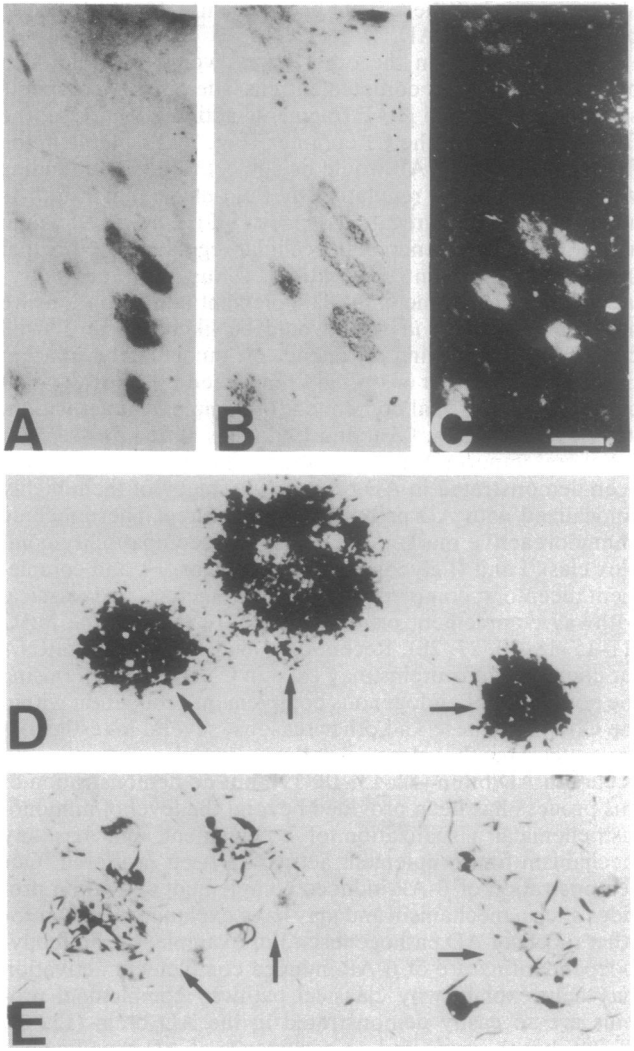

FIG. 5. (*A–C*) Serial 50-μm-thick sections from AD superior frontal gyrus, showing immunohistochemical colocalization of C1q (*A*), C4d (*B*), and thioflavin S-positive senile plaques (*C*). The full range of complement proteins colocalize within the same plaques. *D* and *E*, for example, illustrate C4d and C5b-9 immunoreactivities, respectively, in serial sections from AD hippocampus. Note that although the same plaques are labeled, different elements within the plaques are stained. This is the expected result, since C5b-9 (but not C4d) requires a cell membrane point of attachment. Mouse monoclonal antibodies to human C4d and C5b-9 were obtained from Quidel and used at 1:1000 dilutions. (Calibration bar equals 100 μm in *A–C* and 50 μm in *D* and *E*).

the AD brain without the usual mediation of immunoglobulin. We have demonstrated the mechanism for this process *in vitro*, and we have shown that the process may occur *in situ* in the context of β-AP deposits and β-AP-containing AD pathological structures. Colocalization of C5b–9, the MAC, with β-AP immunoreactivity is particularly important in this regard, since the formation of the MAC is direct evidence that full-blown complement activation has occurred at the site.

Although it remains possible that β-AP-mediated complement activation in AD is simply a response to pathogenesis rather than a cause, several points argue for a more primary role. In the periphery, complement-mediated membrane attack can be a generally beneficial mechanism for removing infected or damaged cells, even if some healthy cells are removed in the process by bystander lysis (21). Nerve cells, however, are postmitotic. For this reason, membrane attack on nervous tissues adjacent to β-AP deposits would almost certainly have deleterious consequences, even as a purely undirected bystander phenomenon. In addition, other laboratories have now demonstrated that complement defense mechanisms such as the membrane inhibitor of reactive lysis (MIRL, CD59) (14), vitronectin (S-protein) (27), and SP40,40 (28) are up-regulated in AD brain. The need for these mechanisms also supports the idea that β-AP-mediated complement activation has pathogenetic significance in AD. Finally, the presence of the MAC, C5b–9, generated as the result of complement activation, implies binding to brain tissue (healthy as well as dystrophic), since the β-AP deposits at the site do not offer an appropriate cell membrane target. This cytolytic process, together with other sequela of complement activation such as opsonization (targeting) of tissue for scavenger cell attack and anaphylotoxin signaling to scavenger cells, is difficult to construe as other than pathogenic.

Antibody-independent complement activation has ample precedent both as a basic immunological phenomenon and as a mechanism of human disease (21, 22). In the fluid phase, C1q binding to naturally occurring nonimmunoglobulin substrates is typically handled by such regulatory factors as C1 inhibitor, so that full-blown complement activation does not occur (21). For this reason, APP_S 751 would be unlikely to activate complement *in vivo* (and does not do so *in vitro*). Likewise, as a cellular or membrane-bound protein (1), APP would still be unlikely to activate complement because cellular defense mechanisms such as decay-accelerating factor, homologous restriction factor (HRF), and membrane cofactor protein (MCP) would come into play (21), and, as noted

above, several of these have been recently demonstrated to be up-regulated in AD.

β-AP deposits, on the other hand, would have no such protection from the complement cascade. Our experiments show that β-AP and β-AP fragments actively bind C1q, the first component of the C1 complex (Fig. 3). Insoluble in the fluid phase (1–3), β-AP would be subject to this C1q binding without subsequent regulation by fluid-phase inhibitors. As an insoluble self-aggregating deposit (1–3), β-AP itself would also not be able to generate the cellular regulatory factors that inhibit complement activation. Thus, the antibody-independent β-AP-mediated complement activation that we have demonstrated *in vitro* would be likely to run its full course *in vivo*, having an impact on immediately adjacent tissues—a point that is strongly reinforced by colocalization of the MAC, the final component of complement activation, with β-AP deposits, C1q, and C4d (Figs. 1 and 5).

Several important markers of immune function have now been demonstrated in AD brain tissue, many of them highly colocalized with AD pathological structures. These include immunoreactive markers for major histocompatibility complex class I and II glycoproteins, cytokines, Fc and complement receptors, complement regulatory factors, and classical pathway complement proteins from C1q through the MAC (11–15, 18–20, 27, 28). Recent data also suggest that mRNA for complement proteins may exist in CNS cells (29), raising the possibility of endogenous complement production within the CNS. For these and other reasons, several investigators have suggested that classical pathway complement activation occurs in AD brain (12, 13, 18, 19), but no demonstration of this process has been provided beyond the level of immunohistochemical visualization of complement, nor has any mechanism for complement activation been proffered. Our demonstration of β-AP-induced complement activation provides such a mechanism and may have explanatory power for other facets of AD pathogenesis. For example, the antibody-independent nature of β-AP-induced complement activation may help explain why classical pathway complement proteins are so easily demonstrated in the AD brain (12, 13, 18–20), but the expected co-occurrence of AD-specific antibodies has been difficult to detect in the great majority of cases (18, 26).

The cause of the dementia of AD is neuronal and neuritic damage, but the cause of AD neuronal and neuritic damage has remained elusive. The fact that we use excessive deposition of β-AP as a unique hallmark and diagnostic criterion may underlie and perhaps justify the experimental attention this peptide has received as a pathogenic factor in AD. However, our laboratory and others (30) have noted extensive β-AP deposition in postmortem brain samples from several elderly patients who were considered to be nondemented throughout life. Likewise, the cerebellum typically exhibits little if any clinical or neural pathology in AD but contains significant numbers of diffuse β-AP deposits (31). These findings suggest that β-AP may be a necessary but not sufficient element in AD pathogenesis, a concept that fits well with the present data and other published reports. For example, full complement activation appears to occur in the context of compacted association cortex β-AP deposits, but it does not occur in the context of diffuse cerebellar β-AP deposits (32, 33). Although both sites contain β-AP (albeit in different aggregation states), it is the association cortex that exhibits concurrent complement activation and neuritic pathology. We also note that AD may well have multiple pathogenic bases and, indeed, this could extend even to the β-AP molecule itself. For example, β-AP might have an innate but limited neurotoxicity (4, 5) that is exacerbated by complement activation. Such hypotheses may be amenable to testing by using neuronal culture and other models.

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1. Selkoe, D. J. (1991) *Neuron* **6**, 487–492.
2. Kang, J., Lemaire, H.-G., Unterbeck, A., Salbaum, J. M., Masters, C. L., Grzeschik, K.-H., Multhaup, G., Beyreuther, K. & Müller-Hill, B. (1987) *Nature (London)* **325**, 733–736.
3. Glenner, G. G. & Wong, C. W. (1984) *Biochem. Biophys. Res. Commun.* **120**, 885–890.
4. Yankner, B. A., Duffy, L. K. & Kirschner, D. A. (1990) *Science* **250**, 279–282.
5. Koh, J. Y., Yang, L. L. & Cotman, C. W. (1990) *Brain Res.* **533**, 315–321.
6. Mattson, M. P., Cheng, B., Davis, D., Bryant, K., Lieberburg, I. & Rydel, R. E. (1992) *J. Neurosci.* **12**, 376–389.
7. Nitsch, R. M., Blusztajn, J. K., Pittas, A. G., Slack, B. E., Growdon, J. H. & Wurtman, R. J. (1992) *Proc. Natl. Acad. Sci. USA* **89**, 1671–1675.
8. Gibson, G. E. & Peterson, C. (1987) *Neurobiol. Aging* **8**, 329–343.
9. Grundke-Iqbal, I., Iqbal, K., Tung, Y. C., Quinlan, M., Wisniewski, H. M. & Binder, L. I. (1986) *Proc. Natl. Acad. Sci. USA* **83**, 4913–4917.
10. Kowall, N. W. & Kosik, K. S. (1987) *Ann. Neurol.* **22**, 639–643.
11. McGeer, P. L., Akiyama, H., Itagaki, S. & McGeer, E. G. (1989) *Neurosci. Lett.* **107**, 341–344.
12. McGeer, P. L., Akiyama, H., Itagaki, S. & McGeer, E. G. (1989) *Can. J. Neurol. Sci.* **16**, 516–524.
13. Rogers, J. & McGeer, P. L. (1992) in *Alzheimer's Disease*, eds. Khachutarian, Z. & Blass, J. (Dekker, New York), pp. 147–163.
14. McGeer, P. L., Walker, D. G. & Akiyama, H. (1991) *Brain Res.* **544**, 315–320.
15. Styren, S. D., Civin, W. H. & Rogers, J. (1990) *Exp. Neurol.* **110**, 93–107.
16. Griffin, W. S. T., Stanley, L. C., Linc, C., White, L., MacLeod, V., Perrot, L. J., White, C. L. & Araoz, C. (1989) *Proc. Natl. Acad. Sci. USA* **86**, 7611–7615.
17. Abraham, C. R., Selkoe, D. J. & Potter, H. (1988) *Cell* **52**, 487–501.
18. Eikelenboom, P. & Stam, F. C. (1982) *Acta Neuropathol.* **57**, 239–244.
19. Ishii, T., Haga, S. & Kametari, F. (1988) in *Immunology and Alzheimer's Disease*, eds. Pouplard-Barthelaix, A., Emile, J. & Christen, Y. (Springer, Berlin), pp. 17–29.
20. Rogers, J., Brachova, L., Webster, S., Lue, L.-F. & Schultz, J., *Res. Immunol.*, in press.
21. Cooper, N. R. (1987) in *Basic and Clinical Immunology*, eds. Stites, D. P. & Terr, A. I. (Appleton & Lange, Norwalk, CT), 6th Ed., pp. 114–127.
22. Cooper, N. R. (1985) *Adv. Immunol.* **37**, 151–171.
23. Mayer, M. M. (1961) in *Experimental Immunochemistry*, eds. Kabat, E. A. & Mayer, M. M. (Thomas, Springfield, IL), 2nd Ed., pp. 133–140.
24. Oltersdorf, T., Ward, P. J., Henriksson, T., Beattie, E. C., Neve, R., Lieberburg, I. & Fritz, L. C. (1990) *J. Biol. Chem.* **265**, 4492–4500.
25. Gaskin, F., Kingsley, B. S. & Fu, S. M. (1987) *J. Exp. Med.* **165**, 245–250.
26. Mori, S., Sternberger, N. H., Herman, M. M. & Sternberger, L. A. (1991) *Lab. Invest.* **64**, 345–351.
27. Akiyama, H., Kawamata, T., Dedhar, S. & McGeer, P. L. (1991) *J. Neuroimmunol.* **32**, 19–28.
28. May, P. C., Lampert-Etchells, M., Johnson, S. A., Poirier, J., Masters, J. N. & Finch, C. E. (1990) *Neuron* **5**, 831–838.
29. Walker, D. G. & McGeer, P. L. (1992) *Mol. Brain Res.* **14**, 109–116.
30. Katzman, R., Terry, R., DeTeresa, R., Brown, T., Davies, P., Fuld, P., Renbing, X. & Peck, A. (1988) *Ann. Neurol.* **23**, 138–144.
31. Yamaguchi, H., Hirai, S., Morimatsu, M., Shoji, M. & Nakazato, Y. (1989) *Acta Neuropathol.* **77**, 314–319.
32. Lue, L. & Rogers, J. (1992) *Neurosci. Abs.* **18**, 733.
33. Rozemuller, J., Stam, F. & Eikelenboom, P. (1990) *Neurosci. Lett.* **119**, 75–78.