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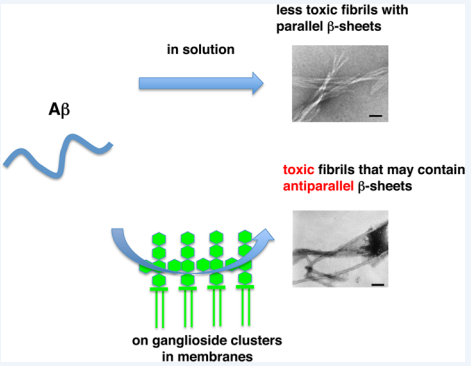
How Do Membranes Initiate Alzheimer's Disease? Formation of Toxic Amyloid Fibrils by the Amyloid β-Protein on Ganglioside Clusters

Katsumi Matsuzaki*

Graduate School of Pharmaceutical Sciences, Kyoto University, 46-29 Yoshida-Shimoadachi-cho, Sakyo-ku, Kyoto 606-8501, Japan

CONSPECTUS: Alzheimer's disease (AD), a severe neurodegenerative disorder, causes more than half of dementia cases. According to the popular "Aβ hypothesis" to explain the mechanism of this disease, amyloid β-peptides (Aβ) of 39−43 amino acid residues aggregate and deposit onto neurons, igniting the neurotoxic cascade of the disease. Therefore, researchers studying AD would like to elucidate the mechanisms by which essentially water-soluble but hydrophobic Aβ aggregates under pathological conditions.



Most researchers have investigated the aggregation of Aβ in aqueous solution, and they concluded that the final aggregation product, the amyloid fibrils, were less toxic than the component peptide oligomers. They consequently shifted their interests to more toxic "soluble oligomers", structures that form as intermediates or off-pathway products during the aggregation process. Some researchers have also investigated artificial oligomers prepared under non-physiological conditions.

In contrast to these "in solution" studies, we have focused on "membrane-mediated" amyloidogenesis. In an earlier study, other researchers identified a specific form of Aβ that was bound to monosialoganglioside GM1, a sugar lipid, in brains of patients who exhibited the early pathological changes associated with AD. This Account summarizes 15 years of our research on this topic. We have found that Aβ specifically binds to GM1 that occurs in clusters, but not when it is uniformly distributed. Clustering is facilitated by cholesterol. Upon binding, Aβ changes its conformation from a random coil to an α-helix-rich structure. A CH−π interaction between the aromatic side chains of Aβ and carbohydrate moieties appended to GM1 appears to be important for binding. In addition, as Aβ accumulates and reaches its first threshold concentration (Aβ/GM1 = ∼0.013), aggregated β-sheets of ∼15 molecules appear and coexist with the helical form.

However, this β-structure is stable and does not form larger aggregates. When the disease progresses further and the Aβ/GM1 ratio exceeds ∼0.044, the β-structure converts to a second β-structure that can seed aggregates. The seed recruits monomers from the aqueous phase to form toxic amyloid fibrils that have larger surface hydrophobicity and can contain antiparallel β-sheets. In contrast, amyloid fibrils formed in aqueous solution are less toxic and have parallel β-sheets. The less polar environments of GM1 clusters play an important role in the formation of these toxic fibrils.

Membranes that contain GM1 clusters not only accelerate the aggregation of Aβ by locally concentrating Aβ molecules but also generate amyloid fibrils with unique structures and significant cytotoxicity. The inhibition of this aggregation cascade could be a promising strategy for the development of AD-modulating therapies.

1. INTRODUCTION

Alzheimer's disease (AD), a severe neurodegenerative disease, is the most common form (more than half) of dementia. A pathological hallmark of AD is the deposition of senile plaques, a major component of which is the fibrillar amyloid β-protein (Aβ). It is widely accepted that Aβ is central to the development of AD.[1,2] Aβ is a small protein or peptide composed of typically 39−43 amino acids residues, but 40 and 42 residue forms are the most common (Figure 1A). Aβ is generated from proteolytic cleavage of the membrane-spanning amyloid precursor protein (APP) by β- and γ-secretases. However, other longer isoforms have been also identified. Aβ-(1−42) is more amyloidogenic than Aβ-(1−40) and the major species found in diffuse plaques, which are considered to be the earliest stage in the deposition of Aβ. In contrast, fibril-rich neuritic plaques contain mixed deposits of Aβ-(1−42) and Aβ-(1−40).[3] Näslund et al. determined the levels of Aβs in various regions of brains using C-terminal specific-antibodies.[4] The average levels of Aβ-(x−42) were higher than those of Aβ-(x−40) even in the subjects without dementia, and a specific role for Aβ-(x−42) early in the disease process was not supported. Both Aβ-(x−42) and Aβ-(x−40) correlated with the degree of dementia. Furthermore, comparing the levels of Aβ-(x−42) and Aβ-(x−40) with the density of neuritic plaques revealed significant correlation.

Aβ is present at a very low concentration (<10^{-8} M) in biological fluids[5] and is essentially soluble with unordered structures. However, under pathological conditions, nontoxic Aβ is converted to aggregated toxic Aβ, which is rich in β-sheet structures and ignites the neurotoxic cascade(s) of Aβ (the so-called "Aβ hypothesis").[6] Therefore, an important theme in

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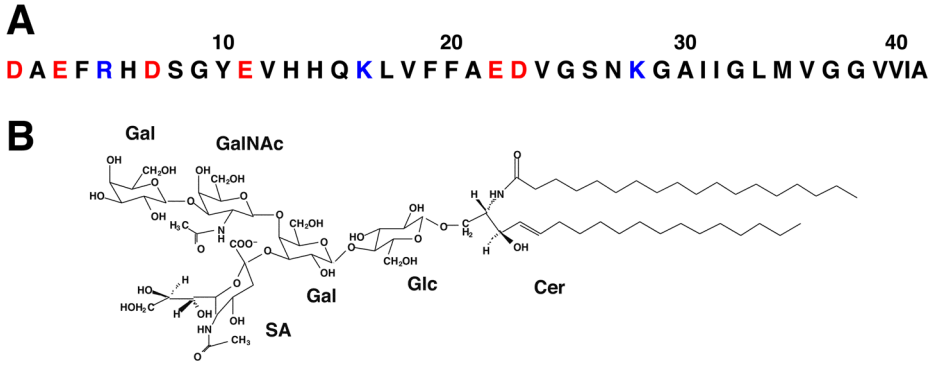

Figure 1. Amino acid sequence of Aβ-(1−42) (A) and chemical structure of GM1 (B). Acidic and basic residues are shown in red and blue, respectively. Cer, ceramide; Glc, glucose; Gal, galactose; GalNAc, *N*-acetylgalactosamine; SA, sialic acid.

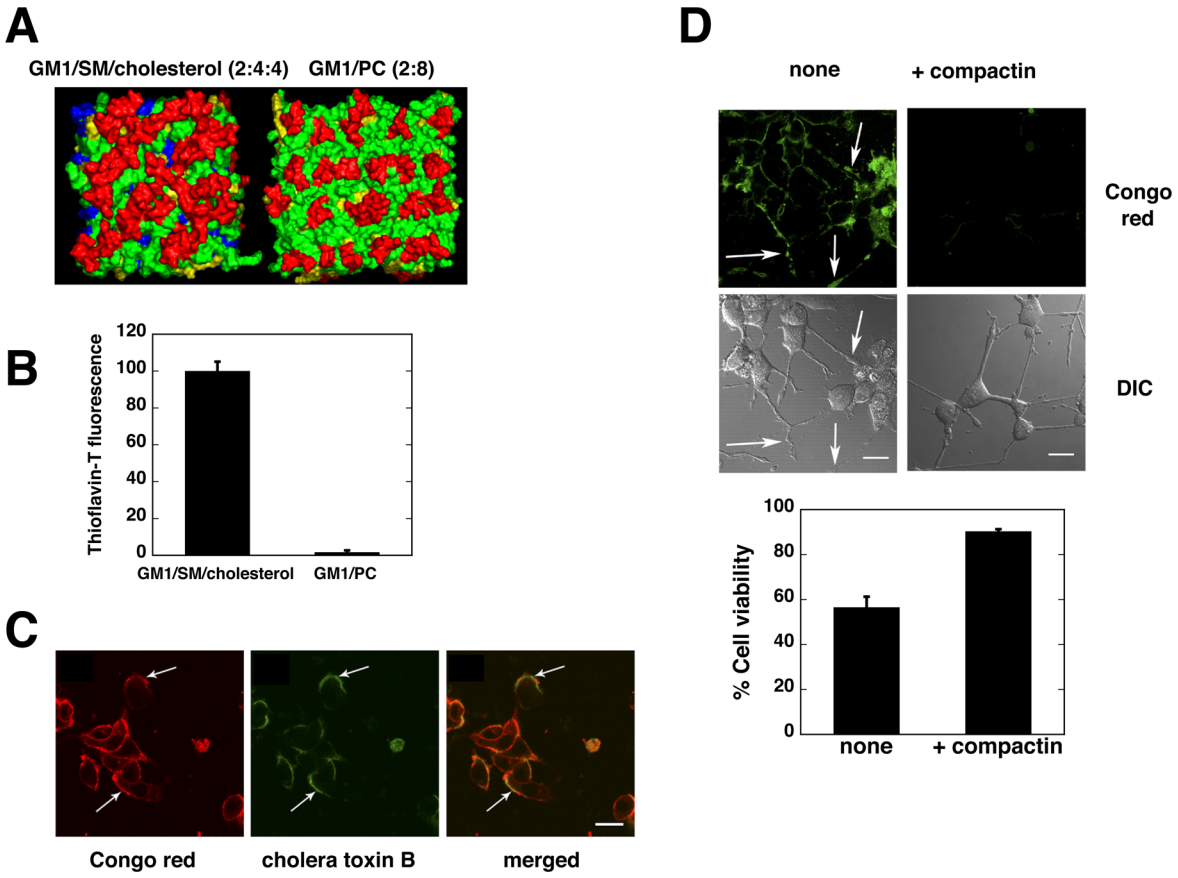

Figure 2. Specific interaction between Aβ and membranes containing GM1 clusters. (A) Top views of the snapshot structures of membranes colored by the lipid type (green, SM or PC; blue, cholesterol; yellow, the ceramide part of GM1; red, the glycan portion of GM1): (left) Aβ-binding GM1/SM/cholesterol (2:4:4); (right) Aβ-nonbinding GM1/PC (2:8). (B) Effects of lipid compositions on fibril formation by Aβ. Aβ-(1−40) (50 μM) in PBS was incubated in the presence of liposomes composed of GM1/SM/cholesterol = 2:4:4 and GM1/PC = 2:8 ([GM1] = 50 μM) at 37 °C for 19 h. Fibril formation was evaluated by the thioflavin-T assay. Data taken from ref 21. (C) Fibril formation on ganglioside-rich domains. PC12 cells were incubated with 10 μM Aβ-(1−42) for 24 h. Amyloid fibrils and cell surface gangliosides were stained with the amyloid-specific dye Congo red (left) and Alexa Fluor 647-conjugated cholera toxin B (middle), respectively. The right panel shows a merged image. Arrows show the colocalization of Congo red fluorescence with ganglioside-rich domains, in which gangliosides were brightly stained with CTX-B. Bar, 20 μm. (D) Cholesterol-dependent amyloid formation. PC12 cells differentiated with neuronal growth factor were incubated with 10 μM Aβ-(1−42) for 24 h with (right) or without (left) the cholesterol synthesis inhibitor compactin. Amyloid fibrils were stained with Congo red (top). The middle row shows differential interference contrast (DIC) images. The depletion of cholesterol rescued cells from amyloid-induced cytotoxicity (bottom). Arrows indicate disrupted neurites. Bar, 20 μm. Adapted with permission from ref 41. Copyright (2007) Elsevier.

current AD research is to elucidate the mechanisms by which Aβ is aggregated.

Aβ-(1−40) spontaneously forms fibrils above ∼1 μM, which is markedly higher than the physiological concentration.[7] Therefore, it is considered that more amyloidogenic Aβ-(1−42) deposits first, possibly as diffuse plaques, and Aβ-(1−40) coassembles with it to form mature plaques.[1] In the case of familial AD, the generation of Aβ is enhanced.[1] However, there is no evidence for accelerated production of Aβ in sporadic AD, a major form of the disease, although a 30−140% increase in

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Aβ production-to-clearance ratio was observed for 7 out of 12 symptomatic AD patients.[8] Therefore, "catalysts" that facilitate the abnormal aggregation of Aβ possibly exist under pathological conditions. Soluble proteins, such as clusterin (Apo J)[9] and metal ions, for example, Zn^{2+} ions[10] were previously shown to facilitate the aggregation of Aβ in aqueous phase. In addition to these soluble factors, accumulating evidence suggests that membranes play an important role in the abnormal aggregation of not only Aβ[11−13] but also other amyloidogenic proteins.[14−16] Yanagisawa et al. identified a specific form of Aβ bound to monosialoganglioside GM1 (GM1) (Figure 1B) in brains exhibiting the early pathological changes associated with AD and also suggested that the GM1-bound form of Aβ may serve as a seed for the formation of Aβ aggregates.[17] Based on this finding, we have been investigating interactions between Aβ and ganglioside-containing model membranes and neuronal cells for 15 years and discovered that clustered, not uniformly distributed, gangliosides specifically mediated the formation of amyloid fibrils by Aβ. The toxicity and physicochemical properties of these fibrils differed from those of the Aβ amyloids formed in solution that most studies have investigated. This review summarizes the Aβ−ganglioside interaction in detail with an emphasis on the latest findings, which were not covered in our previous reviews.[11,18,19]

2. GANGLIOSIDE CLUSTERS AS A PLATFORM FOR THE Aβ−MEMBRANE INTERACTION

Large discrepancies have been reported between the findings obtained by different research groups for the Aβ−membrane interaction (vide infra). One reason for this is the different initial states of Aβ in solution, that is, completely monomeric or already partially aggregated. The amphipathic protein self-aggregates more easily. We focused on the preparation of the aggregate-free Aβ solution. The protein was dissolved in 0.02% ammonia on ice, and any large aggregates were removed by ultracentrifugation at 4 °C. Without agitation, the carefully prepared Aβ-(1−40) solution (50 μM) did not form fibrils at least for 24 h at 37 °C. We examined the binding of Aβ to major membrane lipids including zwitterionic phosphatidylcholine (PC), sphingomyelin (SM), anionic phosphatidylserine (PS), phosphatidylglycerol, and neutral cholesterol. However, none of these lipids exhibited detectable binding to the protein, neither Aβ-(1−40) nor Aβ-(1−42), at physiological pH. From our experiments, the only class of lipids that Aβ bound to was the negatively charged gangliosides, including GM1, even though Aβ is also negatively charged at neutral pH.[20−23] However, note that others have seen the interaction of Aβ-(1−40)[24] and Aβ-(1−42)[25] with anionic PS-containing membranes. Aβ has six acidic amino acids (D^1, E^3, D^7, E^{11}, E^{22}, and D^{23}) with pK_a values of 4.3−4.5 in D_2O, whereas the number of basic amino acids is only three (R^5, K^{16}, and K^{28}), although there are three histidine residues with pK_a values of 6.5−6.6[26,27] (Figure 1A). Interestingly, the existence of gangliosides is not sufficient for binding. Sugar lipids need to be clustered in order to interact with Aβ. For example, Aβ binds to membranes composed of GM1/SM/cholesterol (2:4:4) but not GM1/PC (2:8) despite the same GM1 contents. The former composition resembles that of the so-called lipid raft. Gangliosides, SM, and cholesterol have been proposed to form microdomains, in which the proteins responsible for signal transduction are concentrated.[28] Molecular dynamic (MD) simulations (Figure 2A),[29] as well as excimer fluorescence experiments,[20] suggest that the sugar moiety of GM1 forms a string-like cluster in the former Aβ-binding bilayers, whereas it is uniformly distributed in the latter Aβ-nonbinding membranes. The GM1 cluster has a hydrophobic and negatively charged surface due to the condensation of glycans, especially galactose and sialic acid. Amyloid fibrils are formed on the GM1/SM/cholesterol membranes only, and this was confirmed by an increase in the fluorescence of the amyloid-specific dye thioflavin T (Figure 2B).[30]

The formation of the GM1 cluster was previously shown to be cholesterol-dependent.[20] A decrease in the cholesterol content in GM1/SM/cholesterol mixtures reduces the number of Aβ-binding sites. Based on MD simulations, cholesterol formed many hydrogen bonds with SM and more preferably with GM1 and played important roles in preventing the membrane from interdigitation in the GM1/PC membrane and promoting other lipids to assemble.[29] This cholesterol dependence can explain the roles of two strong risk factors in the pathogenesis of AD, that is, aging and the apolipoprotein E4 allele.[31] Previous studies demonstrated that the amount of cholesterol in the exofacial leaflets of the synaptic plasma membrane was increased in aged[32] as well as apolipoprotein E4-knock-in[33] mice. GM1 clustering was shown to occur at the presynaptic neuritic terminals in mouse brains in an age-dependent manner.[34] Furthermore, human AD brains also show abnormality in lipid metabolism in accordance with our model. Significant increase in GM1 was reported in Aβ-positive nerve terminals from the AD cortex,[35] and lipid rafts from the frontal cortex and the temporal cortex of AD brains contained a higher concentration of GM1 compared with an age-matched control.[36] Diet-induced hypercholesterolemia accelerated the amyloid pathology in a transgenic mouse model.[37] A link between cholesterol, Aβ, and AD has been reported.[38,39]

The GM1 cluster-specific interaction of Aβ has also been observed in neuronal cells. When Aβ was incubated with PC12 cells, amyloid fibrils accumulated selectively on ganglioside-rich regions (Figure 2C), which induced cell death.[40,41] The depletion of cholesterol rescued cells from Aβ accumulation and cytotoxicity (Figure 2D).[41]

3. DRIVING FORCES FOR THE INTERACTION BETWEEN Aβ AND GANGLIOSIDE CLUSTERS

The amino acid sequence of Aβ indicates that the 28 N-terminal residues corresponding to the extracellular part of APP are hydrophilic, whereas the C-terminal remainder corresponding to the membrane-spanning domain is hydrophobic (Figure 1A). However, Aβ only weakly interacts with zwitterionic lipids,[42] which suggests that the contribution of the hydrophobic interaction between the C-terminal hydrophobic residues and hydrocarbon core of the bilayer to the affinity of Aβ for GM1 clusters is minimal. The binding constant of Aβ-(1−28) to raft-like membranes containing GM1 clusters is 500-fold smaller than that of Aβ-(1−40), and this difference has been attributed to a free energy gain upon a coil-to-helix transition.[23] The binding affinity increases with the number of sugar residues in sugar lipids, suggesting the involvement of hydrogen bonding between Aβ and sugar −OH groups or a CH−π interaction between the aromatic side chains of Aβ and sugar carbohydrate moieties.[23] The findings of MD simulations have suggested the importance of the latter interaction.[43] Electrostatic repulsion is know to hamper the binding of net negatively charged Aβ to GM1-containing membranes at physiological pH.[23] However, a cluster of sialic acid residues may trap the side chain of the Lys^{28} residue, inducing a

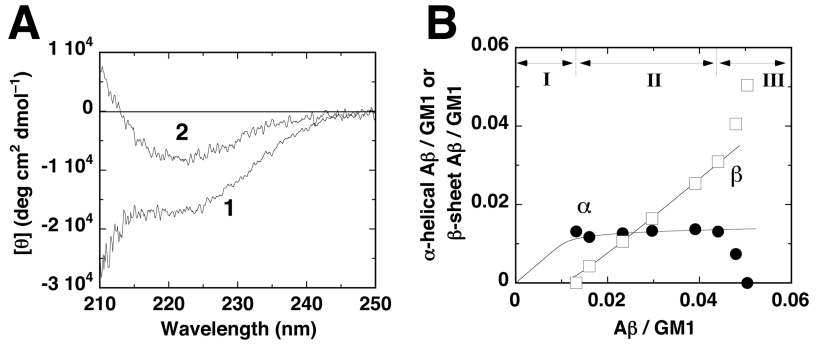

Figure 3. Structural transition of Aβ-(1−40) bound to GM1 clusters. (A) CD spectra of the α-helix-rich (trace 1) and β-sheet-rich (trace 2) forms on GM1/SM/cholesterol (4:3:3) bilayers. Spectral contributions from free Aβ in solution have been subtracted. (B) Concentrations (protein per GM1) of the α-helix-rich (filled circles) and β-sheet-rich (empty squares) species as a function of the total Aβ concentration. CD spectra of membrane-bound Aβ were obtained at various protein-to-bilayer ratios. They were analyzed by a singular value decomposition method to obtain fractions of the α-helix-rich and β-sheet-rich forms. Traces in region II were calculated by assuming that 15 α-helix molecules formed a 15-meric β-sheet with an association constant of 5.8×10^{24}.

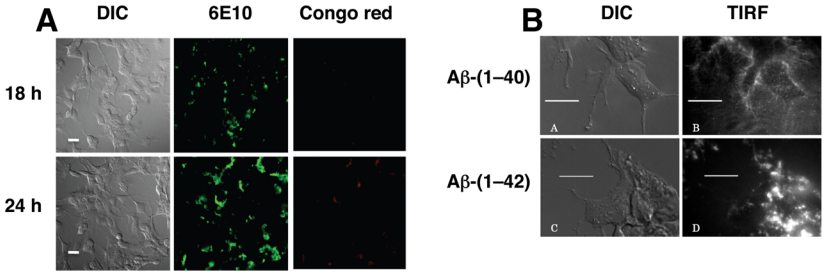

Figure 4. Fibril formation on neuronal cells. (A) Detection of prefibrillar and fibrillar Aβ. SH-SY5Y cells were incubated with 5 μM Aβ-(1−42) for 18 h (upper) and 24 h (lower) at 37 °C, the deposition of Aβ was detected with the anti-Aβ antibody 6E10, and fibrils were stained with Congo red. Bar, 20 μm. (B) Morphology of fibrils. SH-SY5Y cells were incubated with Aβ-(1−42) (5 μM, upper) or Aβ-(1−40) (50 μM, lower) for 48 h at 37 °C, and fibrils stained with Congo red were observed with a total internal reflection fluorescence (TIRF) microscope. Bar, 20 μm. Adapted with permission from ref 22. Copyright 2011 John Wiley and Sons.

conformational change appropriate for the subsequent interaction between Aβ molecules.[43]

4. CONFORMATIONAL TRANSITIONS OF Aβ ON MEMBRANES

Several structural transitions may occur depending on the peptide density in the membrane before Aβ finally forms amyloid fibrils (Figure 3).[20,21,44] Regarding secondary structures, Aβ essentially assumes two conformations, that is, α-helix-rich and β-sheet-rich forms (Figure 3A). Aβ initially assumes the α-helix rich conformation upon binding to GM1 clusters (helicity ∼50%) at lower protein densities (Aβ/GM1 ratio of less than ∼0.013, corresponding to a binding site-occupancy, that is, the amount of bound Aβ divided by the maximum binding capacity, of approximately 1/4, region I in Figure3B). With the accumulation of Aβ, that is, corresponding to the development of the disease, the helical species and aggregated β-sheets (∼15mer) coexist (Aβ/GM1 ratio between ∼0.013 and ∼0.044, region II in Figure3B). However, the β-structure is stable and does not form larger aggregates. At Aβ/GM1 ratios above ∼0.044 (corresponding to a binding site occupancy of approximately 3/4, region III in Figure3B), the β-structure is converted to a second β-structure, which may occur due to rearrangement of β-strands. The second β-structure serves as a seed (nucleus) for amyloid fibrils.

Prefibrillar Aβ and fibrillar Aβ can be visualized on neuronal cells.[22] Aβ was incubated with SH-SY5Y human neuroblastoma cells, the deposition of Aβ was detected with the anti-Aβ antibody 6E10, and fibrils were imaged with Congo red (Figure 4A). The accumulation of Aβ was observed after cells had been incubated for 18 h; however, cells were not Congo red-positive, which indicated that it was prefibrillar Aβ. Fibrillar Aβ was detected at 24 h. More than a 10-fold higher concentration of Aβ-(1−40) was needed to observe a similar extent of fibril deposition with Aβ-(1−42), revealing that the 42-mer aggregates much faster than the 40-mer.

The morphology of the amyloid fibrils that formed on cell membranes can be observed by total internal reflection fluorescence microscopy (TIRFM), which enables the cell surface to be examined (Figure 4B).[22] Relatively long fibrillar structures that originated from membranes and extruded into the aqueous phase were observed for Aβ-(1−40). In contrast, a treatment with Aβ-(1−42) resulted in ambiguous Congo red staining around the cell membrane, suggesting that relatively short fibrils were coassembled.

5. PROPERTIES OF Aβ FIBRILS FORMED ON RAFT-LIKE MEMBRANES

Aβ fibrils that form on raft-like membranes ("membrane fibrils") differ from those that form in aqueous solution ("solution fibrils") in terms of morphology, secondary structure, surface hydrophobicity, and cytotoxicity. Membrane fibrils (width ∼12 nm) are thicker than solution fibrils (∼7 nm) (Figure 5A). Sani et al. also observed similar morphological differences between solution fibrils and fibrils prepared in the presence of various lipids.[45] Furthermore, membrane fibrils are

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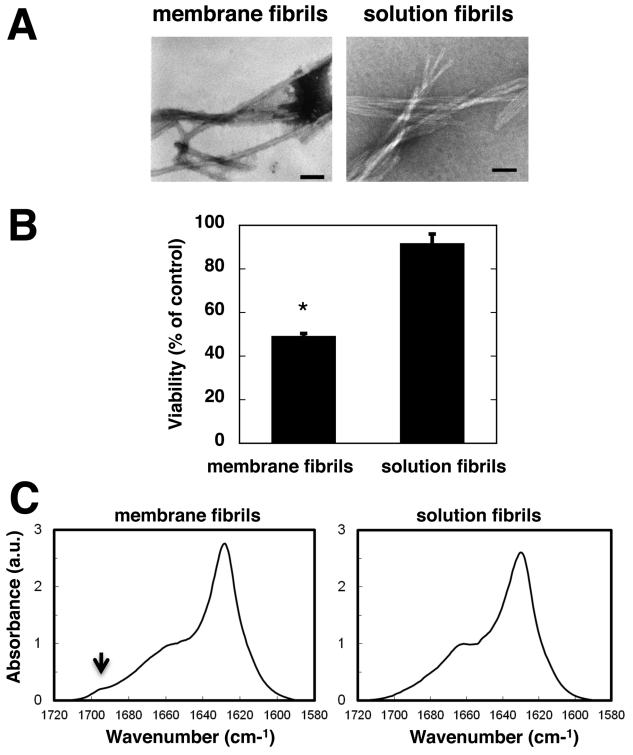

Figure 5. Comparison between Aβ-(1−40) fibrils formed in GM1/SM/cholesterol membranes (membrane fibrils) and those in solution (solution fibrils). (A) Negative-stain transmission electron microscopy images. (B) Cytotoxicity (25 μM) was determined after a 24-h incubation against PC12 cells differentiated with neuronal growth factor with fluorescence intensity of the live cell marker cell-loaded calcein. (mean ± SE; $n = 6$; *$p < 0.001$ against the vehicle treatment). (C) FTIR spectra. Adapted with permission from ref 30. Copyright 2008 Elsevier.

significantly cytotoxic, whereas solution fibrils are less toxic[30,46] (Figure 5B). Differences in surface hydrophobicity and stickiness to cell membranes appear to be related to the different cytotoxicities.

The cytotoxicity of Aβ fibrils has been controversial. It should be noted that the structure and toxicity of an amyloid is highly dependent on preparation.[47] Furthermore, the conventional assay for cytotoxicity using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) is not suitable for determining the toxicity of Aβ, although it has often been used unwittingly for Aβ. Aβ and other amyloidogenic proteins inhibit cellular MTT reduction not because they kill cells but because they dramatically enhance the exocytosis of MTT formazan-containing vesicles.[48−50]

Membrane fibrils formed on GM1 clusters are pathologically relevant. First, GM1-bound Aβ was discovered from AD brains.[17] Second, the antibody 4396C raised against it bound to membrane fibrils and also immunostained the cerebral cortex of AD brains but not control brains.[51] Finally, the peripheral administration of cell penetrating peptide-modified Fab fragments of 4396C into transgenic mice expressing a mutant APP gene substantially suppressed Aβ deposition in the brain.[52]

The structures of solution fibrils have been extensively examined mainly by solid-state NMR and are considered to be composed of in-register parallel β-sheets.[53] We measured FTIR spectra for solution fibrils and membrane fibrils (Figure 5C). Both FTIR spectra exhibited intense bands at approximately 1629 cm^{-1}, indicating that β-sheets are major conformations. A minor band at approximately 1695 cm^{-1} was discernible in the spectrum of membrane fibrils, which suggested the presence of antiparallel β-sheets.[54−56] Detailed structural analysis by isotope-edited FTIR and solid-state NMR is under progress. Toxic oligomeric Aβ has been reported to form antiparallel β-sheets.[57,58] The Iowa (D23N) mutant of Aβ also can form fibrils with an antiparallel β-sheet architecture.[59]

We hypothesized that the low polarity environments provided by GM1 clusters may drive the formation of fibrils with these unique properties. We used a mixture of 1,4-dioxane and phosphate-buffered saline to mimic such microenvironments because (1) the organic solvent is freely miscible with water and therefore the same stock solution of Aβ can be used to prepare both types of fibrils and (2) the presence of CH_2, ether O, and OH groups chemically mimics the sugar group of GM1. We demonstrated that the mixture facilitated the formation of Aβ fibrils with a similar morphology, secondary structure, surface hydrophobicity, and cytotoxicity to those of the membrane fibrils. The formation of in-register parallel β-sheets is unfavorable in low polarity environments because electrostatic repulsion between the charged amino acid side chains in adjacent β-strands manifests in media with a low dielectric constant. Furthermore, the antiparallel orientation of the macrodipoles of helices, precursors of β-sheets (Figure 3),[44] is also preferable in media with a low dielectric constant. It appears to be difficult for membrane fibrils possessing antiparallel β-sheets to completely bury nonpolar amino acids inside the fibrils, resulting in stronger surface hydrophobicity.

6. MECHANISM UNDERLYING THE CYTOTOXICITY OF FIBRILS

Aβ fibrils have been shown to trigger functional disorder in neuronal cells and cell death,[60−64] although soluble Aβ oligomers have also been proposed to play a pivotal role in the onset of AD.[9,64−72] Cell death was detected after the significant accumulation of fibrils, and no antioligomer antibody A11-positive spot was detected when cells were incubated with Aβ,[22] which suggested that fibril-induced physicochemical stress, such as the induction of a negative curvature[73] or membrane deformation upon fibril growth,[74] may lead to cytotoxicity. A11-positive oligomers were also not formed in the fibrillization with GM1-containing liposomes.[30] The mechanism responsible for fibril cytotoxicity is currently being investigated in more detail.

7. SUMMARY AND PERSPECTIVES

Figure 6 depicts our proposed model for the formation of toxic amyloid fibrils by Aβ on GM1 clusters. When GM1 molecules do not form clusters, which corresponds to healthy conditions, Aβ does not interact with the neuronal membranes. However, if they form clusters, Aβ binds to the membrane, changing its conformation from a random coil to an α-helix-rich structure, which initiates the pathology associated with AD, that is, the lateral physicochemical interaction between lipids determining whether one is healthy or diseased. Clustering can be caused by abnormal lipid metabolism such as an increase in cholesterol content or late endocytic dysfunction.[75] As the accumulation of Aβ proceeds and reaches the first threshold concentration (Aβ/GM1 = ∼0.013), aggregated β-sheets (∼15 mer) appear and coexist with the helical form. However, this β-structure is stable and does not form larger aggregates. When the disease

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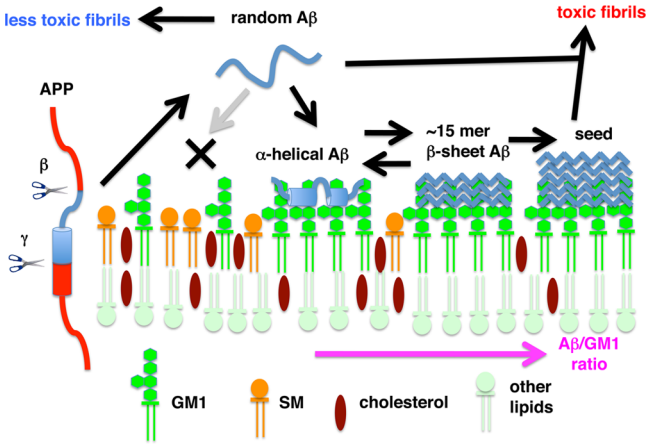

Figure 6. Schematic representation of the proposed formation of toxic amyloid fibrils by Aβ on GM1 clusters. Aβ is generated from the proteolytic cleavage of APP by β- and γ-secretases. When GM1 molecules do not form clusters, Aβ does not interact with neuronal membranes. Aβ specifically binds to a GM1 cluster, changing its conformation from a random coil to an α-helix-rich structure. Helical species and aggregated β-sheets (~15 mer) coexist at Aβ/GM1 ratios between ~0.013 and ~0.044. However, this β-structure is stable and does not form larger aggregates. The β-structure is converted to a second, seed-prone β-structure at Aβ/GM1 values above ~0.044. The seed recruits monomers from the aqueous phase to form toxic amyloid fibrils that may contain antiparallel β-sheets. In contrast, amyloid fibrils formed in aqueous solution are less toxic and have parallel β-sheets.

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An important conclusion of our study is that membranes containing GM1 clusters not only accelerate the aggregation of Aβ but also generate amyloid fibrils with potent cytotoxicity and unique structures. The inhibition of this aggregation cascade could be a promising strategy for the development of AD-modulating drugs. For example, compounds that specifically bind to GM1 clusters can block the cascade at an early stage. The α-helix-to-β-sheet conformational transition can be inhibited by molecules that recognize the α-helical form. Chemicals that bind and break amyloid fibrils could also reduce cytotoxicity. We have already identified several candidate molecules.[76] The driving forces for Aβ binding, the detailed structures of intermediate species and the final amyloid, and the mechanism underlying cytotoxicity will be elucidated in future studies. Furthermore, effects of pH, endosomal lipids, and membrane curvature on the fibril formation by Aβ will be interesting subjects of research because the GM1-bound Aβ has been suggested to form in endosomes.[77]

AUTHOR INFORMATION

Corresponding Author

*E-mail: mkatsumi@pharm.kyoto-u.ac.jp.

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Notes

The authors declare no competing financial interest.

Biography

Katsumi Matsuzaki obtained his Ph D. in 1992 from Kyoto University when he was an Assistant Professor there. He was appointed as an Associate Professor at Kyoto University in 1997 and has been a full Professor of Biophysical Chemistry at Graduate School of Pharmaceutical Sciences, Kyoto University, since 2003.

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