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Amyloid Aggregation on Lipid Bilayers and Its Impact on Membrane Permeability

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Fibrillar protein aggregates (amyloids) are involved in several common pathologies, e.g., Alzheimer's disease and type II diabetes. Accumulating evidence suggests that toxicity in amyloid-related diseases originates from the deposition of protein aggregates on the cell membrane, which results in bilayer disruption and cell leakage. The molecular mechanism of damage to the membrane, however, is still obscure. To shed light on it we have performed coarse-grained molecular dynamics simulations of fibril-forming amphipathic peptides in the presence of lipid vesicles. The simulation results show that highly amyloidogenic peptides fibrillate on the surface of the vesicle, damaging the bilayer and promoting leakage. In contrast, the ordered aggregation of peptides with low amyloidogenicity is hindered by the vesicles. Remarkably, leakage from the vesicle is caused by growing aggregates, but not mature fibrils. The simulation results provide a basis for understanding the range of aggregation behavior that is observed in experiments with fibril-forming (poly)peptides.

Keywords: protein aggregation; Alzheimer's disease; diabetes type II; membrane leakage; coarse-grained; molecular dynamics

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Introduction

Amyloid-related diseases such as Alzheimer's, Parkinson's and type II diabetes are characterized by the presence of protein aggregates in one or more tissues. These protein aggregates are assembled into well-ordered filamentous, β-sheet-rich structures[1–6] termed amyloid fibrils. Compelling evidence suggests that oligomeric or other prefibrillar precursors of amyloid fibrils are the main toxic species,[7–9] but the mechanism of cell and tissue damage in amyloid-related diseases is poorly understood. The interactions of amyloidogenic polypeptides with the cell membrane can accelerate fibril formation and are reported to be involved in the toxicity of oligomers or protofibrils.[10,11] Furthermore, amyloid peptides not only aggregate on the lipid surface, but also penetrate into membranes.[12,13] The accumulation of amyloid peptides destabilizes lipid bilayers and alters their permeability,[14,15] which may contribute to cell damage and toxicity. Accordingly, recent biophysical studies have targeted the interactions of amyloid aggregates with lipid vesicles.[16–18] These studies have provided evidence that the formation of fibrils on a membrane damages the bilayer's integrity.

The fibrillation process is dynamic and transient, making it difficult to explain the mechanism of aggregation based on experimental evidence alone. Many efforts have therefore been made to study protein and peptide aggregation by computational methods and in particular molecular dynamics simulations, as reviewed in Refs. 19 and 20. Atomistic simulations of proteins are limited to the time scale of 10–100 ns, which is not enough to simulate aggregation or even oligomerization. Accordingly, there is a need to reduce the complexity, e.g., by investigation of oligopeptide systems[21–27] or by use of simplified models. Recently, a coarse-grained model of amphipathic peptides has enabled the simulation of amyloid fibril formation and the analysis of aggregation pathways and kinetics, which were observed to depend on the intrinsic amyloidogenic tendency of the peptide.[28,29] The peptide monomer has a single degree of freedom. Its free-energy profile has two minima corresponding to the amyloid-competent and amyloid-protected states. The peptides can fibrillate only in the former

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Abbreviations used: LJ, Lennard-Jones.

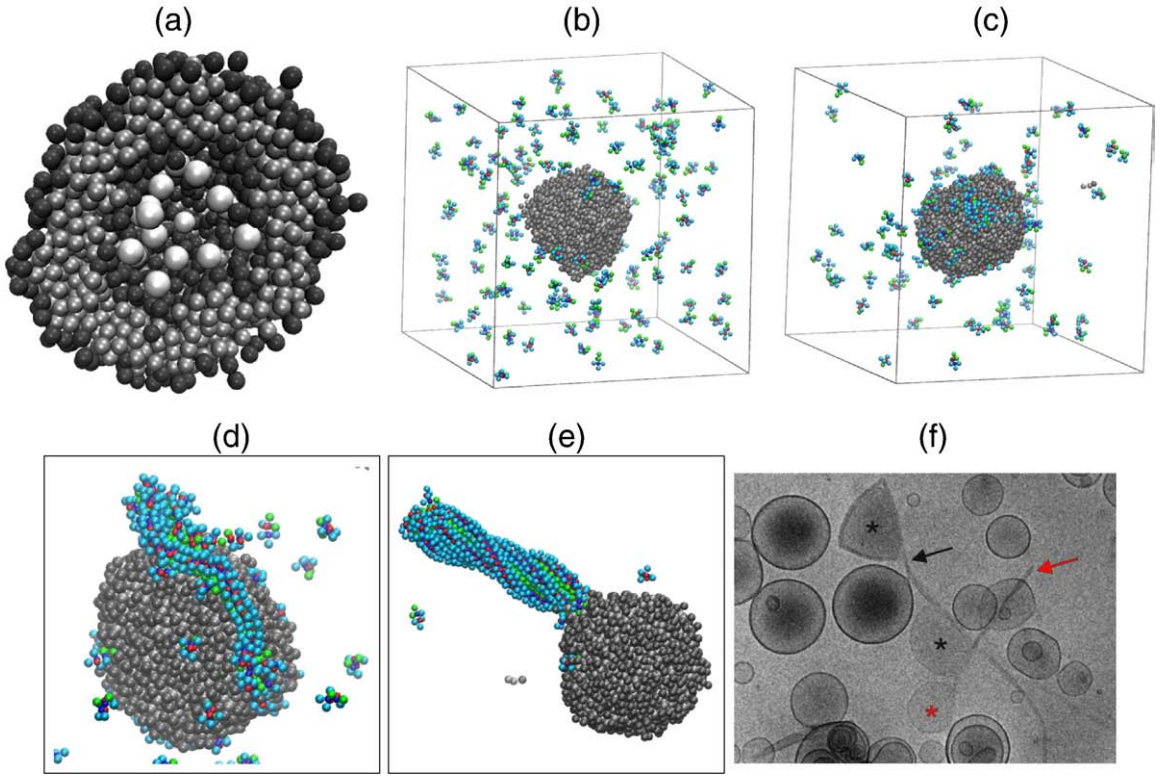

Fig. 1. Aggregation on the surface of a vesicle. (a) A cut through a lipid vesicle showing probe spheres in the inner space. The two-bead hydrophobic tail and one-bead hydrophilic head of the lipids are colored gray and black, respectively; probe spheres are shown in white. (b–e) Snapshots from a simulation of peptides with intermediate amyloidogenicity (see subsection "Coarse-grained model of amphipathic peptide") at times 0 (b), 10 (c), 125 (d), and 700 ns (e). Peptide beads are color coded according to their type: hydrophobic in green, hydrophilic in cyan and dipoles in red and blue. Details about the peptide model are given in Refs. 28 and 29. (f) Cryotransmission electron microscopy of the interaction between human islet amyloid polypeptide and unilamellar phospholipid vesicles (reproduced with permission from Ref. 18). Few vesicles (asterisks) are in contact with fibrils (arrows). Note the similar relative orientation of the fibril and the vesicle in the simulations (e) and electron microscopy image (f), in particular the vesicle and fibril marked in red.

conformation. Hence, the amyloidogenic potential corresponds to the free-energy difference between the two states. Here, we use a novel coarse-grained model of a bilayer-forming lipid (the lipid molecules spontaneously assemble into spherical, unilamellar bilayer vesicles; see Fig. 1a), together with the previously developed amphipathic peptide model, to study the amyloid aggregation on lipid vesicles. The goal of the simulation study is twofold: to analyze the influence of the vesicles on the aggregation mechanism and to analyze the effect of aggregation on the structure and permeability of the lipid bilayer.

Results and Discussion

Multiple independent simulations were performed for each of four types of amphipathic peptides having high, intermediate, low or very low amyloidogenic potential (see Table 1). Note that the interaction energy between the monomeric peptide and the lipids is similar for the amyloid-prone and amyloid-protected conformations.

Fibrillation on the vesicles

Four snapshots from a simulation of peptides with intermediate amyloidogenic potential are depicted in Fig. 1b–e. Upon starting the simulation, about half of the peptides quickly adsorb to the vesicle surface. The initial adsorption does not depend on the amyloidogenicity because of the aforementioned similar hydrophobic interactions between the lipids and amyloid-prone or amyloid-protected peptide conformations. Peptide adsorption is followed by transient formation of small oligomeric species (2–10 peptides) within tens of nanoseconds (Figs. 1c and 2). Peptide monomers and oligomers subsequently form larger aggregates with filament characteristics, i.e., a file of ordered peptides (Fig. 1d), before their maturation into a fibrillar structure (Fig. 1e, note the similarity to electron microscopy of amyloid fibrils attached to the surface of vesicles; Fig. 1f; Ref. 18).

Highly amyloidogenic peptides fibrillate more rapidly in the presence of lipid vesicles than in their absence, while the opposite is observed for peptides of low amyloidogenicity (half-times of fibril formation are given in Table 1). The faster