Supporting Information: Aggregate Size Dependence of Amyloid Adsorption onto Charged Interfaces

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QCM-D Data Analysis.

For a thin, homogeneous film that does not extend into the bulk solution and has a low degree of acoustic coupling to the surrounding medium, the temporal variation in frequency, \( \Delta F \), is proportional to the mass, \( \Delta m \), adsorbed on the surface of a quartz crystal resonator according to the Sauerbrey relation:¹

\[
\Delta m = c \frac{|\Delta F|}{n}
\]  

(S1)

where \( c \approx 17.7 \text{ ng cm}^{-2} \text{ Hz}^{-1} \) for a 5 MHz crystal, \( n \) is the overtone number and \( \Delta F \) is the frequency shift measured at different harmonics.

Figure S1 shows the QCM-D data from six experiments discussed in this work. In the
deposition of lipid bilayers, Equation S1 is a good approximation\textsuperscript{1,2} as indicated in Figure S2 by the relatively low $\Delta D$ and by the similar response in $\Delta F/n$ for different overtones. The deposition of 3:1 POPC:DOTAP bilayers resulted in a $|\Delta F|/n$ of 25-27 Hz corresponding to an adsorbed layer mass of 443-478 ng cm\textsuperscript{-2}. The estimated average area per lipid is 0.52-0.56 nm\textsuperscript{2} which is consistent with previous QCM-D measurements.\textsuperscript{3,4}

In the case of viscoelastic films, Equation S1 is not valid and, in this study, the QCM-D response is analyzed with the viscoelastic Voigt-based model.\textsuperscript{1,5} The model consists of one or several homogeneous layers at the sensor surface characterized by four parameters (density, viscosity, shear modulus and thickness). The outer layer is in contact with a semi-infinite solution modelled as a Newtonian fluid of certain density and viscosity. The fitting of the experimental $\Delta F$ and $\Delta D$ signals to the viscoelastic model yields a measure of the adsorbed mass and viscoelastic properties of the layer. Data treatment and modeling were conducted using Qtools version 3.0 (Biolin scientific AB, Gothenburg, Sweden).

The QCM-D data obtained upon injection of fibrillar solutions at low ionic strength, $c_s$, are fitted by a one-layer extended viscoelastic Voigt-based model\textsuperscript{6} using overtone numbers 3, 5, 7 and 9. The viscoelastic layer represents the adsorbed fibrils, while the supported lipid bilayer is considered to be unaffected by fibril adsorption. The extended model accounts for a linear frequency dependence of the viscosity, $\eta$, and shear modulus, $\mu$, of the fibril layer,\textsuperscript{5} expressed in terms of the frequency factors $\alpha_\eta$ and $\alpha_\mu$, for $\eta$ and $\mu$, respectively. The density and viscosity of the bulk fluid are fixed to 1 kg l\textsuperscript{-1} and 1 g m\textsuperscript{-1} s\textsuperscript{-1}, respectively. The density of the fibril layer is also set to 1 kg l\textsuperscript{-1}, while $\Delta m$, $\eta$, $\mu$, $\alpha_\eta$ and $\alpha_\mu$ are free fitting parameters. The results are summarized in Table S1, where $\Delta m$, $\eta$, $\mu$ are averaged over the long-time plateau region. As the adsorbed mass increases, both the viscosity and the shear modulus of the fibril layer increase, in agreement with the formation of a viscoelastic network.\textsuperscript{7} In order to determine the confidence interval, the fitting is repeated with fixed $\alpha_\eta$ and $\alpha_\mu$ deviating by 10% from the values yielding a minimum of the $\chi^2$-parameter. The main finding from our experiments is that adsorption on the positively charged lipid
Figure S1: Frequency (blue lines) and dissipation changes (red lines) for overtone-numbers 3, 5, 7, 9 after the injection of the vesicle dispersion during the QCM-D experiments reported in Figure 1 in the main text. The zero values of $\Delta F/n$ and $\Delta D$ correspond to the lipid bilayer equilibrated with the buffer solution. Injections of buffer and fibrils is indicated by the green and black arrows, respectively.

The length of $\text{A}_\beta_{1-40}$ sonicated fibrils was characterized using a XE 100 atomic force microscope (Park) and XEI software. Non-contact atomic force microscopy (AFM) was performed with NANOSENSORS™ PPP-NCHR (PointProbe® Plus Non-Contact / tapping mode -

Atomic Force Microscopy Measurements.

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Figure S2: Frequency (blue lines) and dissipation changes (red lines) for overtone-numbers 3, 5, 7, 9, 11 after injection of 0.5 mg/ml dispersion of POPC:DOTAP 3:1 vesicles in 200 mM NaCl aqueous solution.

Table S1: Parameters from the best-fit of the QCM-D data to the one-layer extended viscoelastic Voigt-based model. Averages and standard deviations of \( \Delta m \), \( \eta \) and \( \mu \) are calculated over the long-time plateau regions. Frequency factors are scalar quantities and time-independent.

|       | \( \Delta m \) (10\(^2\) ng cm\(^{-2}\)) | \( \eta \) (10\(^{-3}\) s N m\(^{-2}\)) | \( \mu \) (10\(^4\) N m\(^{-2}\)) | \( \alpha_\eta \) (10\(^{-3}\)) | \( \alpha_\mu \) (10\(^{-2}\)) |
|-------|------------------------------------|----------------------------------|----------------------------------|------------------|------------------|
| Batch 1 | 6.6 ± 0.3 | 1.16 ± 0.01 | 3.1 ± 0.2 | -7 ± 1 | 9 ± 1 |
| Batch 2 | 6.7 ± 0.2 | 1.25 ± 0.01 | 4.8 ± 0.2 | -5 ± 1 | 8 ± 1 |

High resonance frequency - Reflex coating) probes. Samples of peptide concentration 10 \( \mu \)M of non-sonicated and sonicated fibrils were dried on freshly cleaved mica surfaces. Figure S4 shows amplitude and topography frames of size 5\( \times \)5 \( \mu \)m or 3\( \times \)3 \( \mu \)m (256\( \times \)256 pixels). The length of non-sonicated fibrils at \( c_s = 50 \) mM is 1–2 \( \mu \)m, while sonicated fibrils at \( c_s = 50 \) mM and \( c_s = 200 \) mM have lengths between 40 nm and 130 nm.

Molecular Dynamics Simulations.

The all-atom model of A\( \beta \)\(_{17–42}\) fibrils is constructed from a pair of peptides\(^8\) forming a cross-section of the fibril. The pair structure is replicated 80 times yielding a roughly 40 nm long fibril. The obtained structure is annealed twice from 380 K to 300 K during 2 ns and subsequently equilibrated at 300 K for 10 ns to a more realistic structure, which includes
Figure S3: Analysis of QCM-D data for two different batches of sonicated fibrils at $c_s = 50$ mM. Top panels: frequency (blue lines) and dissipation changes (red lines) for overtone-numbers 3, 5, 7, 9 after injection of 4 $\mu$M A$_{\beta 1-40}$ fibrils in 20 mM phosphate pH 7.4 buffer and fit to the Voigt-based model (black lines). Bottom panels: changes in adsorbed mass per area (blue lines) obtained from the fit of the data shown in the corresponding upper panels. Shaded areas represent confidence intervals of the estimated quantities. The zero values of $\Delta F/n$ and $\Delta D$ correspond to the POPC:DOTAP 3:1 bilayer equilibrated with the buffer solution.

The fibril twist. We use implicit solvent with the Generalized Born formalism$^9$ and solvent relative permittivity of 80. Simulations are performed with 2 fs time step, 1.2 nm cutoff for both van der Waals and real-space Coulombic interactions, and the Berendsen thermostat$^{10}$ with 1 ps coupling. The all-atom structure is modelled with the Amber99SB force field$^{11}$ and simulated using the GROMACS package,$^{12}$ version 4.6.6.

The all-atom models of A$_{\beta 1-42}$ and A$_{\beta 1-40}$ fibrils were constructed by replicating PDB entries 5KK3 and 2M4J, respectively, using the open source package Packmol.$^{13}$ Simulations were performed using OpenMM, version 7.0.1.$^{14}$ The energy of the system is minimized using an implementation of the L-BFGS optimization algorithm.$^{15}$ The obtained structure was simulated for 10 ns in the NVT ensemble. In order to minimize the energy of the structures, simulated annealing was carried out using a Langevin integrator,$^{16}$ with friction coefficient 1 ps$^{-1}$ and time step 2 fs, sweeping the temperature twice, from 380 K to 300 K. We used the Onufriev-Bashford-Case GBSA model using the GB$^{OBC}$II parameters$^{17}$ and the Amber99SB force field.$^{11}$ Simulations are performed with 2 fs time step, 1.5 nm cutoff for the nonbonded
Figure S4: AFM amplitude (A, B, C) and topography (D, E, F) images for samples of Aβ₁–₄₀ nonsonicated fibrils at $c_s = 50$ mM (A, D) and sonicated fibrils at $c_s = 50$ mM (B, E) and $c_s = 200$ mM (C, F). Samples have peptide concentration 10 $\mu$M.

interactions, and 0.15 M implicit salt concentration.

**Aggregates of Aβ₁⁷–₄₂ and Aβ₁–₄₀ Peptides.**

Figure S5 and S6 are analogous to Figure 3A in the main text, showing surface–aggregate free energy profiles for aggregates of Aβ₁–₄₀ and Aβ₁⁷–₄₂, respectively. Compared to the case of Aβ₁–₄₂, the attraction to the surface is of similar strength for the 1-40 peptide, and weaker for the 17-42 fragment. As a consequence, the oscillating trend of $K_{HA}$ is observed at salt concentrations, $c_s$, larger than 0.15 M for Aβ₁⁷–₄₂, and at $c_s > 0.35$ M for Aβ₁–₄₂ and Aβ₁–₄₀ aggregates (Figure 4 in the main text). The parameters used for the line segment models of Aβ₁–₄₂, Aβ₁–₄₀, and Aβ₁⁷–₄₂ assemblies are reported in Table S2.
Figure S5: Angularly averaged interaction free energy, $w(r)$, as a function of surface–aggregate separation for $A\beta_{1-40}$ assemblies of various size, $N$, and 0.4 M salt concentration, $c_s$.

Figure S6: Angularly averaged interaction free energy, $w(r)$, as a function of surface–aggregate separation for $A\beta_{17-42}$ assemblies of various size, $N$, and 0.2 M salt concentration, $c_s$. 
Table S2: Parameters for the line segment models of Aβ1–42, Aβ1–40, and Aβ17–42 assemblies.

|        | $z_m^*$ | $z_m$ | $l_m$ (Å) | $s_0$ (Å) | $s_1$ (Å) | $\nu$ |
|--------|---------|-------|-----------|-----------|-----------|-------|
| Aβ1–42 | -1.55   | -3    | 2.8       | 17.0      | 24.2      | 91.8  |
| Aβ1–40 | -1.35   | -3    | 2.2       | 19.3      | 72.7      | 1556  |
| Aβ17–42| -1.00   | -1    | 2.6       | 20.3      | 27.8      | 208   |

**Preferential Aggregate Orientation.**

MC simulations are performed using the converged free energy profiles shown in Figure 3, S5, and S6 as an additional term of the Hamiltonian. In the biased simulations, all surface–aggregate separations are equally probable and we thoroughly sample the probability of the orientation of the aggregate with respect to the surface. The orientation is defined by the angle $\theta$ between the principal axis of the aggregate and the surface. The probabilities are normalized by the average value at large separations, where all orientations are equally probable, and converted into free energy values using the Boltzmann formula. Figure S7 shows free energy values of aggregate orientations for equally probable surface–aggregate separations. The distance from the surface is expressed as fractions of the length of the aggregate along its principal axis. The white regions indicate the decrease of the number of available rotational states as the aggregate approaches the surface. For $r/L = 0.5$, Figure S7 shows that the larger aggregates are preferentially oriented perpendicularly to the surface, due to end-point electrostatic attraction.
Figure S7: Angularly resolved free energy for equally probable surface–aggregate separations for $\text{A}\beta_{1-42}$, $\text{A}\beta_{1-40}$, and $\text{A}\beta_{17-42}$ assemblies of various size, $N$, and ionic strength, $c_s$. 
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