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## Supporting Information for

## Unraveling the Mechanism of Amyloid-β Peptide Oligomerization and Fibrillation at Chiral Interfaces

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Figure S1. (A, B) Semilogarithmic plots of the cumulative surface residence time distributions p(t) of A $\beta$ (1-40) on (A) *R*- and (B) *S*-cysteine modified surfaces with -NH<sub>2</sub> and -COOH groups. The Solid lines represent the best double-exponential fit to the raw data. Error bars represent the standard deviation from replicate experiments. (C, D) AFM height images taken for (C) *R*- and (D) *S*-cysteine modified surfaces with -NH<sub>2</sub> and -COOH groups after they have been incubated in A $\beta$ (1-40) solution (1  $\mu$ M in PBS) at 37 °C for 24 hours. Scale bar: 500 nm. (E, F) Energy-minimized average model of (E) *R*- and (F) *S*-cysteine molecules -NH<sub>2</sub> and -COOH groups interacting with A $\beta$ (1-40) peptide (Protein Data Bank (PDB) ID: 1AML). Peak dotted lines represent for hydrogen bonds.

As shown in the Figs. S1A and S1B, the cumulative residence time function p(t) are well fitted by a double-exponential decay for both *R*- and *S*-cysteine modifications with -NH<sub>2</sub> and -COOH groups with the similar surface residence times,  $\tau_1 \sim 0.1$  s and  $\tau_2 \sim 0.8$  s. The 2D-diffusion of A $\beta$ (1-40) molecules on these two chiral surfaces were constrained to move within locally small regions with the mean diffusion coefficient value of ~0.02 µm<sup>2</sup>s<sup>-1</sup>. AFM images (Figs. S1C and S1D) show elongated fibrils on both two chiral surfaces after 24 h of incubation. These results indicate that the fibrillation processes of A $\beta$ (1-40) are not significantly distinct on both *R*and *S*-cysteine modifications with -NH<sub>2</sub> and -COOH groups via their similar chiral recognition with A $\beta$ (1-40) molecule as illustrated by docking simulations (Figs. S1E and S1F).

**Materials.** *R*-cysteine (*R*-cys), *S*-cysteine (*S*-cys), N-succinimidyl 3-maleimidopropionate (NHS-Mal), trimethoxysilylpropanethiol (MPTMS) were purchased from Sigma-Aldrich. Anhydrous methanol and dimethyl sulfoxide (DMSO) were purchased from Energy-Chemical. Amyloid- $\beta$  peptide A $\beta$ (1-40) and fluorescence TAMRA-labeled A $\beta$ (1-40) were purchased as lyophilized powder from AnaSpec USA. Their purity was confirmed by mass spectroscopy and HPLC. 50mM NaCl phosphate-buffered saline (PBS) was freshly prepared from sodium and potassium salts: NaCl, KCl, Na<sub>2</sub>HPO<sub>4</sub>, and KH<sub>2</sub>PO<sub>4</sub> to give a pH value of 7.4 at 25°C.

**Aβ(1-40)** Solution Preparation. Aβ(1-40) solution was prepared as reported previously.<sup>1,2</sup> First, a fresh DMSO solution of Aβ(1-40) peptide was prepared by dissolving the lyophilized powder in DMSO at a concentration of 1 mg/mL. The DMSO solution was further diluted in a large amount of PBS to a final concentration of 2  $\mu$ M. Then, the solution was filtered through a 20-nm membrane filter to remove the pre-existent aggregates of peptide molecules. The final concentration of peptide was then determined by UV adsorption at 280 nm as ~1  $\mu$ M. Note the 1.0  $\mu$ M concentration used here, more than one order of magnitude lower than the critical micelle concentrations for Aβ(1-40), kept as a constant after filtration through a 20-nm membrane filter. Thus, there is no pre-aggregation or fibril growth in the aqueous solution after filtration processes.

*R*- and *S*-cys modified surfaces. The *R*- and *S*-cys modified surfaces were prepared on freshly cleaned substrates (glass or mica) as follows. First, the substrates were immersed in an anhydrous methanol solution of MPTMS (4 wt%) at 60°C for 10 hours followed by rinsed with anhydrous methanol and dried with nitrogen. Then, the MPTMS modified substrates were immersed in a DMSO solution of NHS-Mal (4 mg/mL) at room temperature for 24 hours followed by rinsed with plenty of ultra-pure water and dried with nitrogen. Finally, the NHS modified substrates were placed into aqueous solution of *R*- or *S*-cys molecules (20 mg/mL) at room temperature for 18 hours, washed with plenty of ultra-pure water and dried with nitrogen. The *R*- or *S*-cys modifications were confirmed by X-ray photoelectron spectroscopy technique.

Atomic force microscope (AFM). The morphologies of  $A\beta(1-40)$  fibers were imaged on an atomic force microscopy (AFM, FM-Nanoview 1000) in magnetic tapping mode. A nitride coated silicon tip on nitride level with 0.7 N/m spring constant and 190 kHz resonance frequency was used. To image the fibrils, we removed each surface from the  $A\beta(1-42)$  solution, rinsed it with purified water, and imaged it under ambient conditions.

Total internal reflection fluorescence microscopy (TIRFM). TIRFM experiments were carried out at 25 °C on a commercial instrument based on a Nikon inverted microscope. A 532 nm laser was shined into a 100x oil immersion objective lens with 1.45 numerical aperture to generate an evanescent wave that illuminates the virus within ~100 nm from the glass-water interface. The fluorescence images were acquired on an Andor EM-CCD. A series of fluorescence images were acquired. We obtained single NPs trajectories (positions vs. time)

from the time-dependent images using an algorithm developed by MATLAB. The timedependent fluorescence images ( $55 \times 55 \ \mu m^2$ ) are shown in movie formats, with time steps per frame of 40 ms on each surface. Particles that existed since the first and the last frames were discarded due to the uncertainty in assigning their residence time and diffusivities. To calculate object positions, the closest objects in sequential frames were linked. The threshold to link objects between sequential frames is set at 2 µm, as the average displacement during 0.04 s in bulk solution is expected to be  $(4 \times 150 \mu m^2 s^{-1} \times 0.04 s)^{1/2} \approx 5 \mu m$ .

The surface residence time ( $\tau$ ) of single objects reflects the binding strength of the molecularsurface interaction. The cumulative surface residence time function  $p(t) = \sum_{i=1}^{t} f_i \exp(-t/\tau_i)$ , representing the probability that a given object has a residence time more than or equal to the time t, is always preferred to analyze heterogeneous behaviors of adsorbed molecules on the surface. The subscript i denotes the population of interacting modes for proteins, each with its own characteristic residence time ( $\tau_i$ ) for the first-order desorption and the fraction of molecules exhibiting a particular  $\tau_i$ . The surface residence time of single objects was calculated by determining the number of frames multiplied by the exposure time of the frame.

The diffusion processes (*D*) of single objects can be accurately analyzed from the cumulative  $p(r^{2}, t) = \sum_{i=1}^{\infty} f_{i} \exp(-r^{2}/4D_{i}t)$ square-displacement distributions described as , where *p* represents the probability that an object will diffuse a distance > *r* in a time interval *t* by a diffusion coefficient  $D_{i}$  with the sum of the multiple modes. To provide more accurate results on the residence time and diffusion coefficient measurements, a large number of 10<sup>4</sup> single molecules were probed to calculate both p(t) and  $p(r^{2}, t)$ .

**Docking studies.** We obtained the structure of  $A\beta(1-40)$  monomer from the PDB (ID: 1AML) as characterized by solution NMR technique. The structures of  $A\beta(1-42)$  monomer, *R*- and *S*-cys molecules were prepared by using AutoDock Tools. The torsion angles of the *R*- and *S*-cys molecules were regarded as flexible. Binding calculations were carried out between  $A\beta(1-40)$  monomer and *R*- or *S*-cys molecules by AutoDock Vina. The starting binding positions of *R*- and *S*-cys were randomly determined, and 50 modes of docking with the resultant low binding energy were obtained during docking processes.

<sup>&</sup>lt;sup>1</sup> Shezad, K.; Zhang, K.; Hussain, M.; Dong, H.; He, C.; Gong, X.; Xie, X.; Zhu, J.; Shen, L. Surface Roughness Modulates Diffusion and Fibrillation of Amyloid-β Peptides. *Langmuir* **2016**, *32*, 8238-8244.

<sup>&</sup>lt;sup>2</sup> Shen, L.; Adachi, T.; Vanden Bout, D.; Zhu, X. Y. A Mobile Precursor Determines Amyloid-β Peptide Fibril Formation at Interfaces. *J. Am. Chem. Soc.* **2012**, *134*, 14172-14178.