The Dynamic Nature of Amyloid Beta (1-40) Aggregation

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Supporting Information

§1. Compatibility of TAMRA and FITC for the FRET experiment

Figure 1S shows the structure of TAMRA and FITC. Figure S2 shows that 5 µM of FITC has a fluorescence signal between 500 to 640 nm with a peak around 520 nm and represents the green color spectrum. This range overlaps with the absorbance spectrum of TAMRA that has a peak near 550 nm. FITC can therefore act as a donor and TAMRA as an acceptor. When a solution of 5 µM TAMRA was excited at 488 nm, it had a very weak fluorescence signal between 500 to 640 nm with a peak around 570 nm that represents the red area of the spectrum. When the two dye solutions were mixed so that the final concentration of each dye was 5 µM, there was only a little decrease in the fluorescence intensity in the wavelengths at which FITC fluoresces and no enhancement in the fluorescence signal in wavelengths at which the TAMRA emits.

Since FRET is a result of the long range dipole-dipole interactions between the donor and the acceptor and strongly depends on the distance, r, between the donor and the acceptor molecules (~ r^{-6}), a closer spatial arrangement (closer than the mean spacing between the dyes) is required to obtain a FRET signal between FITC and TAMRA at the above concentrations.

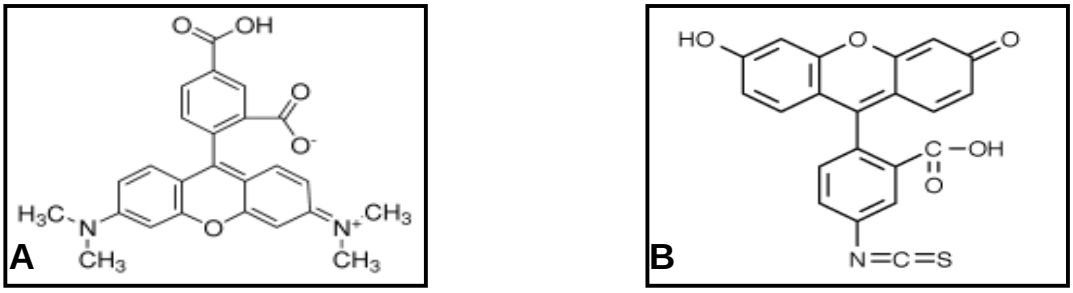

Figure S1. A-TAMRA. B-FITC

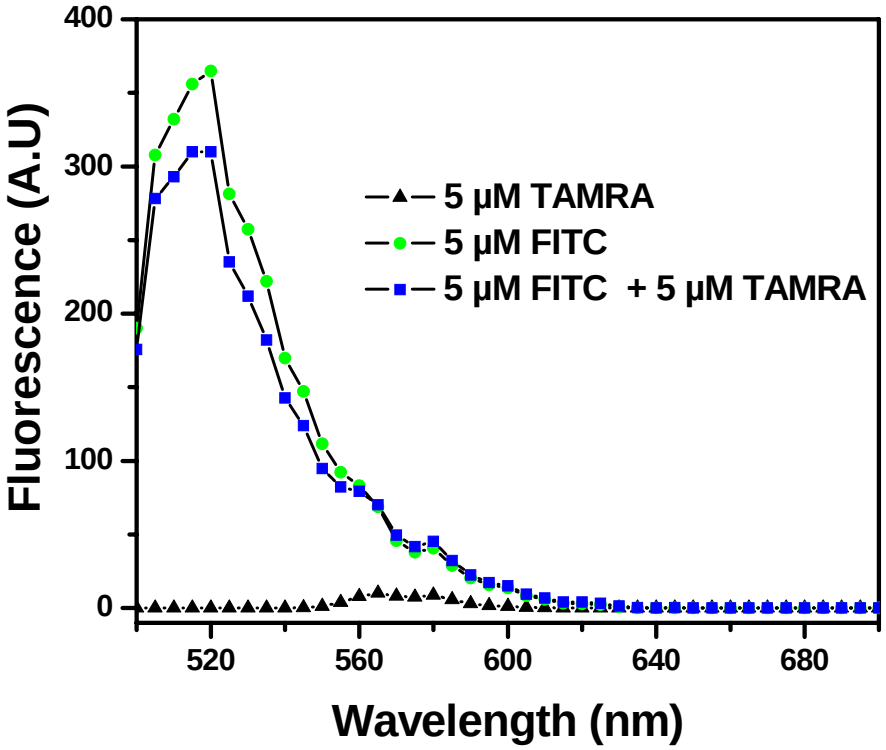

Figure S2. FRET experiments with the dyes alone (without Aβ (1-40)) in 100 mM Tris buffer. Each solution was excited at 488 nm and the fluorescence signal was measured at 500–700 nm. 5 μM TAMRA (solid triangles), 5 μM FITC (solid circles). A mixture that contains 5 μM TAMRA and 5 μM FITC (solid squares) did not show any FRET signal.

§2 Supporting Figures

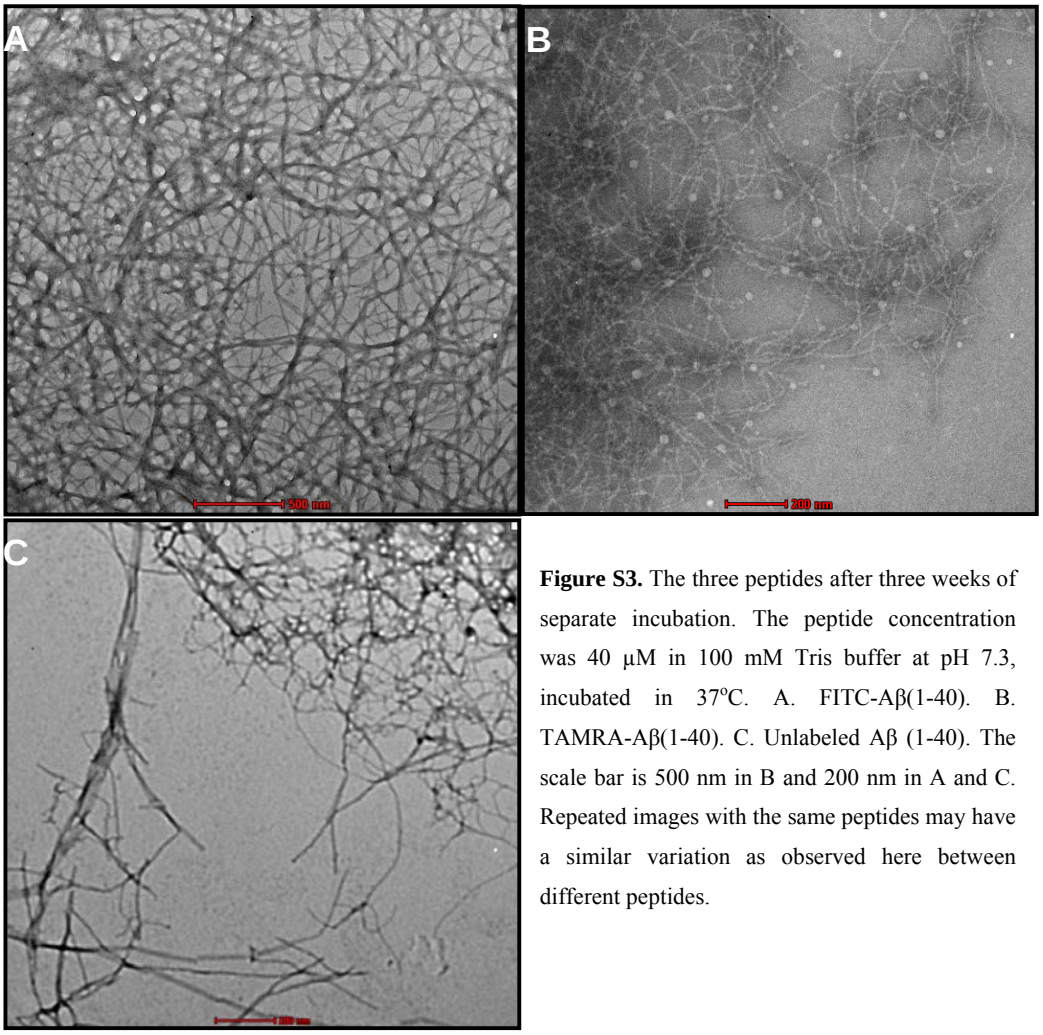

Figure S3. The three peptides after three weeks of separate incubation. The peptide concentration was 40 µM in 100 mM Tris buffer at pH 7.3, incubated in 37°C. A. FITC-Aβ(1-40). B. TAMRA-Aβ(1-40). C. Unlabeled Aβ (1-40). The scale bar is 500 nm in B and 200 nm in A and C. Repeated images with the same peptides may have a similar variation as observed here between different peptides.

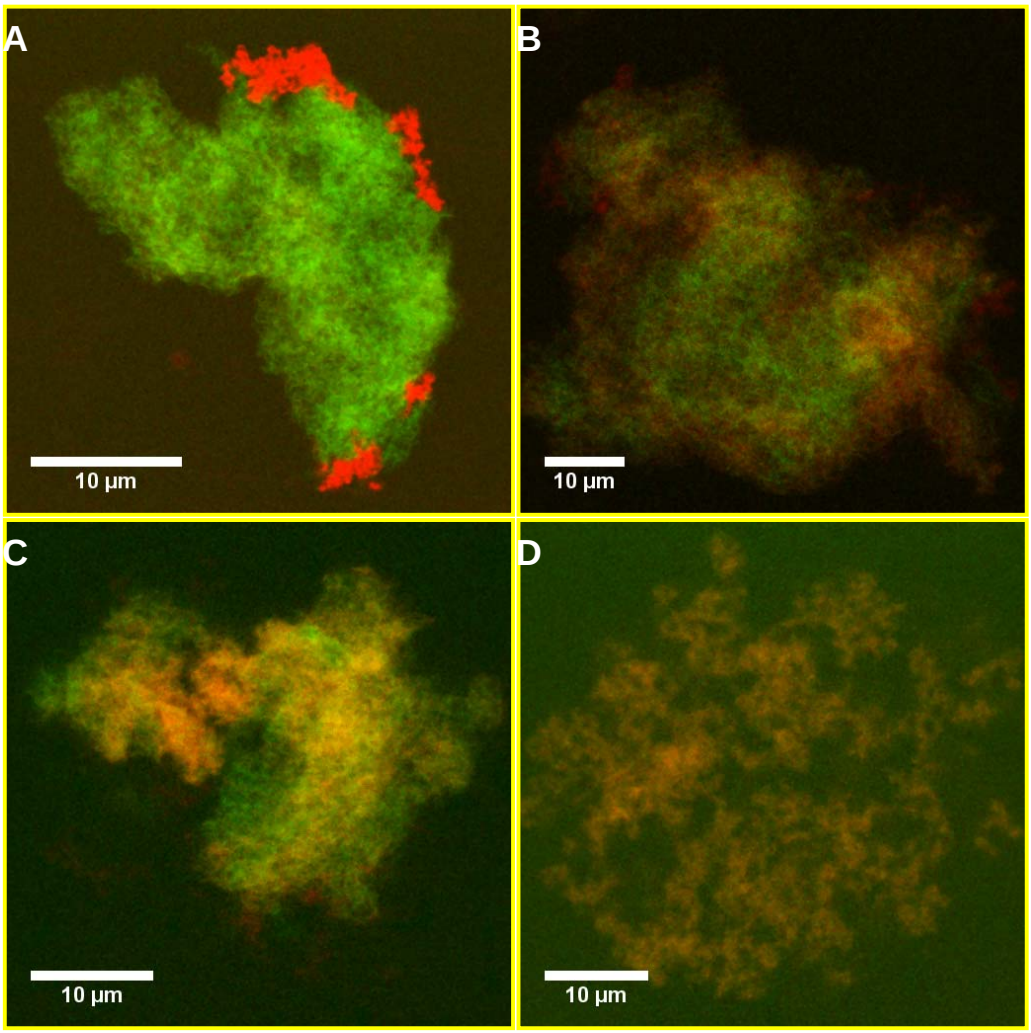

Figure S4. The recycling process over time. The green area represents the donor, the red represents the acceptor and the yellow represents the co-localized area. The concentration of each peptide is 40 µM before mixing and 20 µM after mixing. The samples were mixed after three weeks of separate fibrillization. A. t = 0. B. t = 9 days. C. t = 54 days. D. the control experiment, in which the two labeled monomers were mixed and fibrilized together for three weeks. In this sample we get maximum collocalization and FRET signals. Scale bar is 10 microns in all images.

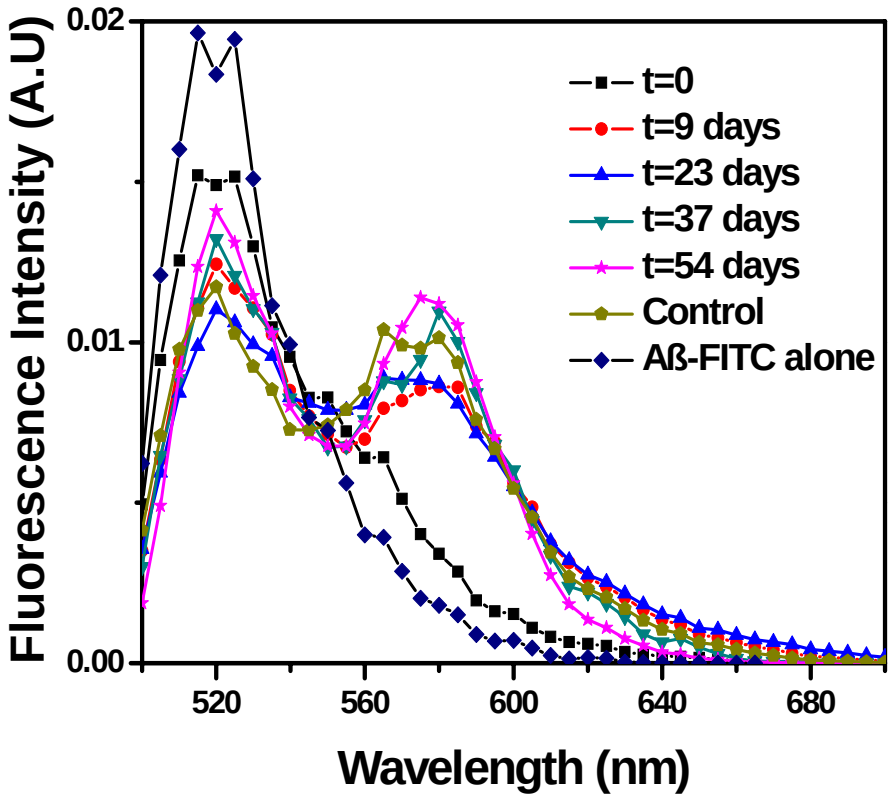

Figure S5. A summary of the 20 μM Aβ (1-40) FRET experiments. The conditions are as in Figure S3. The normalized fluorescence signal was followed as a function of time. Samples were excited at 488 nm and their emission was collected at 500–700 nm.

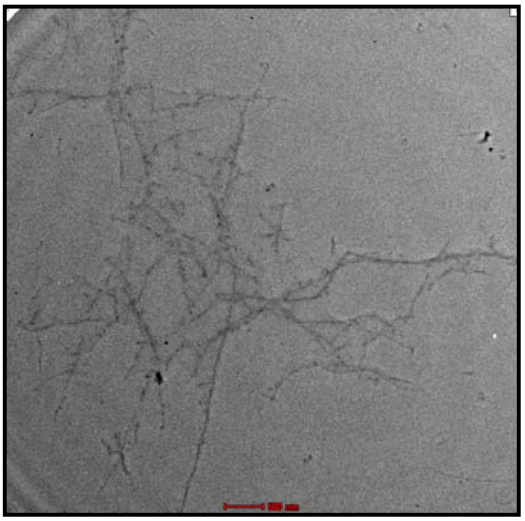

Figure S6. TEM image of mixed fibrils. The concentration of each peptide is 2.5 μM. Scale bar is 500nm.