Supplementary Information

Insulin Amyloid Polymorphs: Implications for Iatrogenic Cytotoxicity

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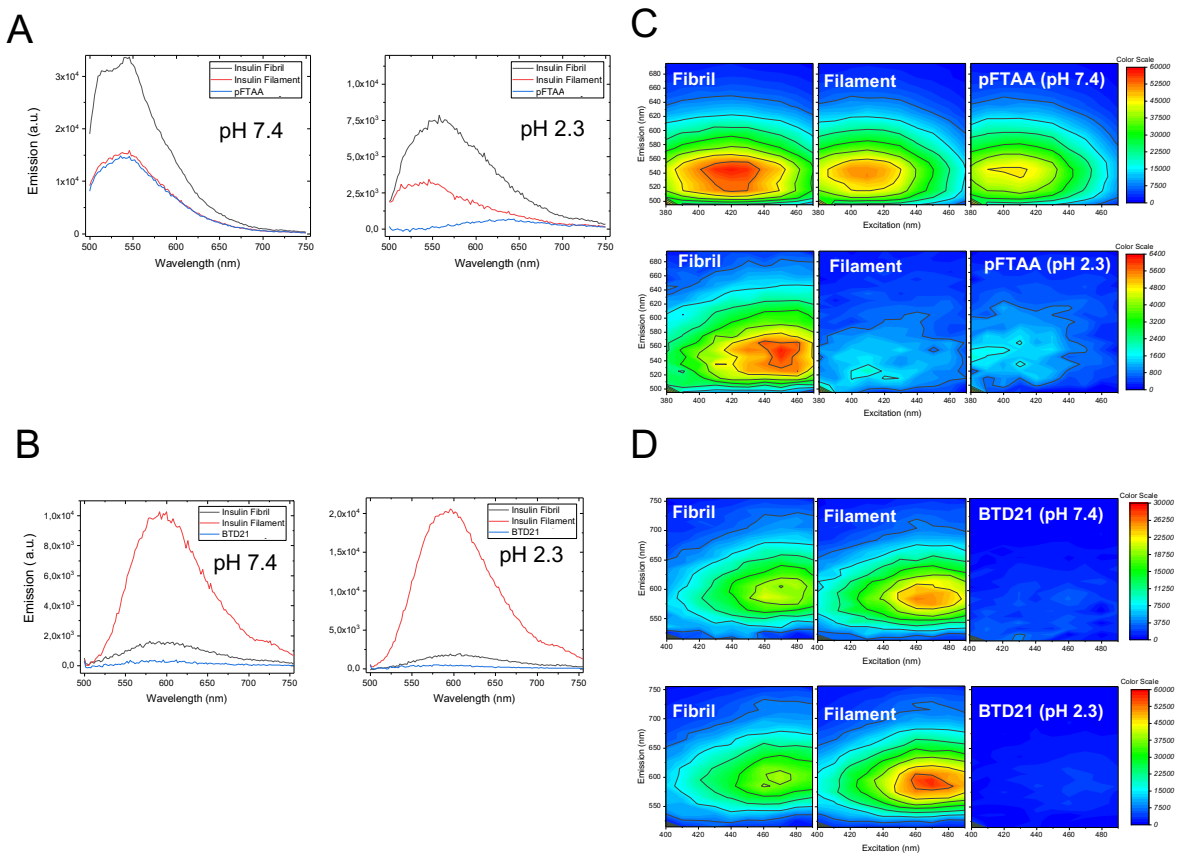

Figure S1 Binding of pFTAA and BTD21 to insulin fibrils and filaments. (A, B) Fluorescence spectra of bovine insulin fibrils and filaments using A) pFTAA and B) BTD21 amyloid probe at pH 7.4 and pH 2.3, respectively. The excitation wavelength was 470 nm (slits 10/5, ex/em). Note that the amplitude scales are different although the settings are the same. (C, D) 2D scan/3D plots of the excitation/emission of C) pFTAA and D) BTD-21 with insulin fibrils and filaments at pH 7.4 (top rows) and pH 2.3 (lower rows). To compare the intensities, the amplitude scales are the same along each row.

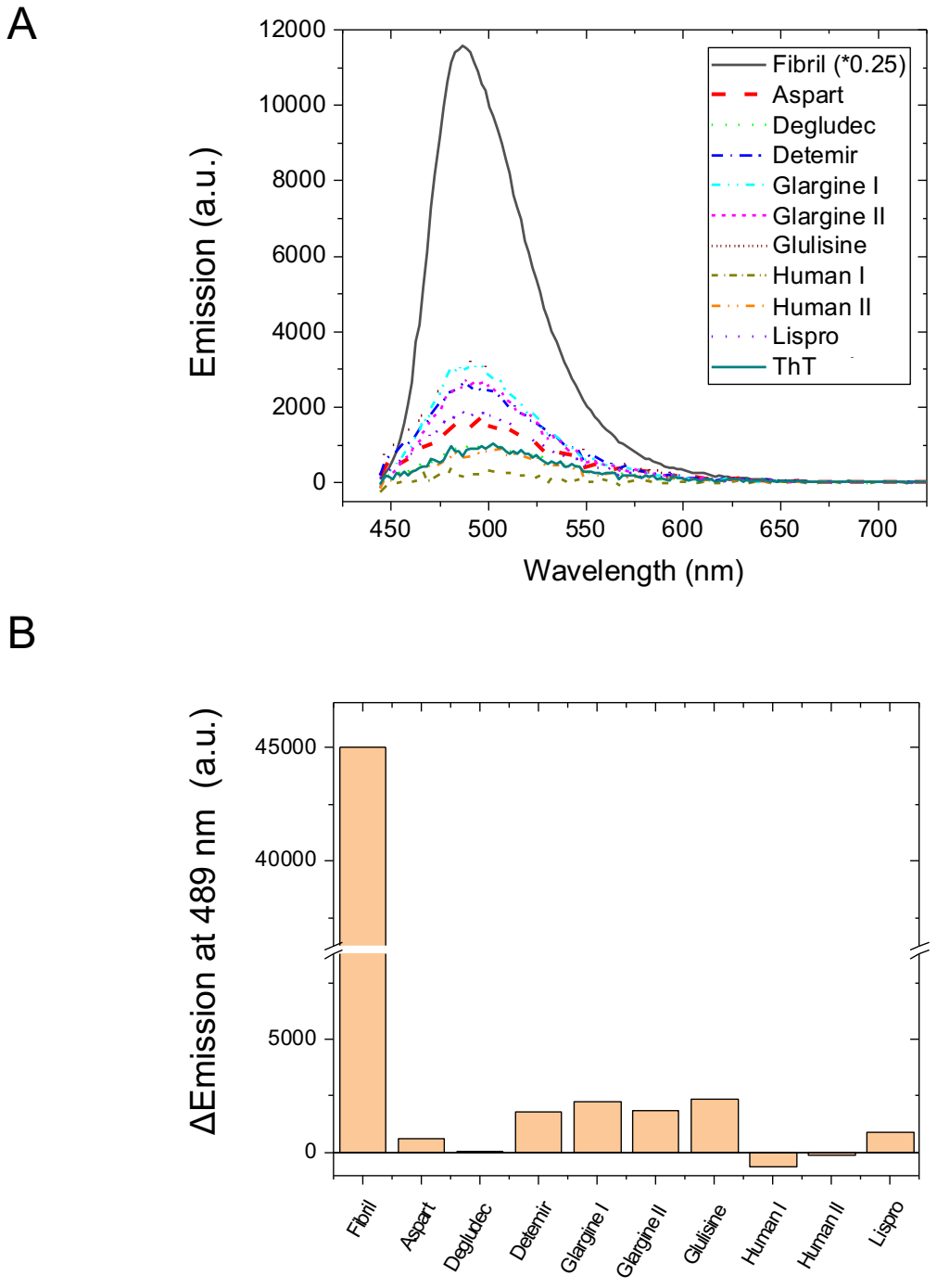

Figure S2 ThT fluorescence of incubated insulin preparations with insulin fibrils shown for comparison. A) Spectral raw data. Note that the spectrum of ThT multiplied with 0.25 was shown for comparison of all spectras. B) The intensity at 489 nm with the intensity of the blank (ThT) sample subtracted.

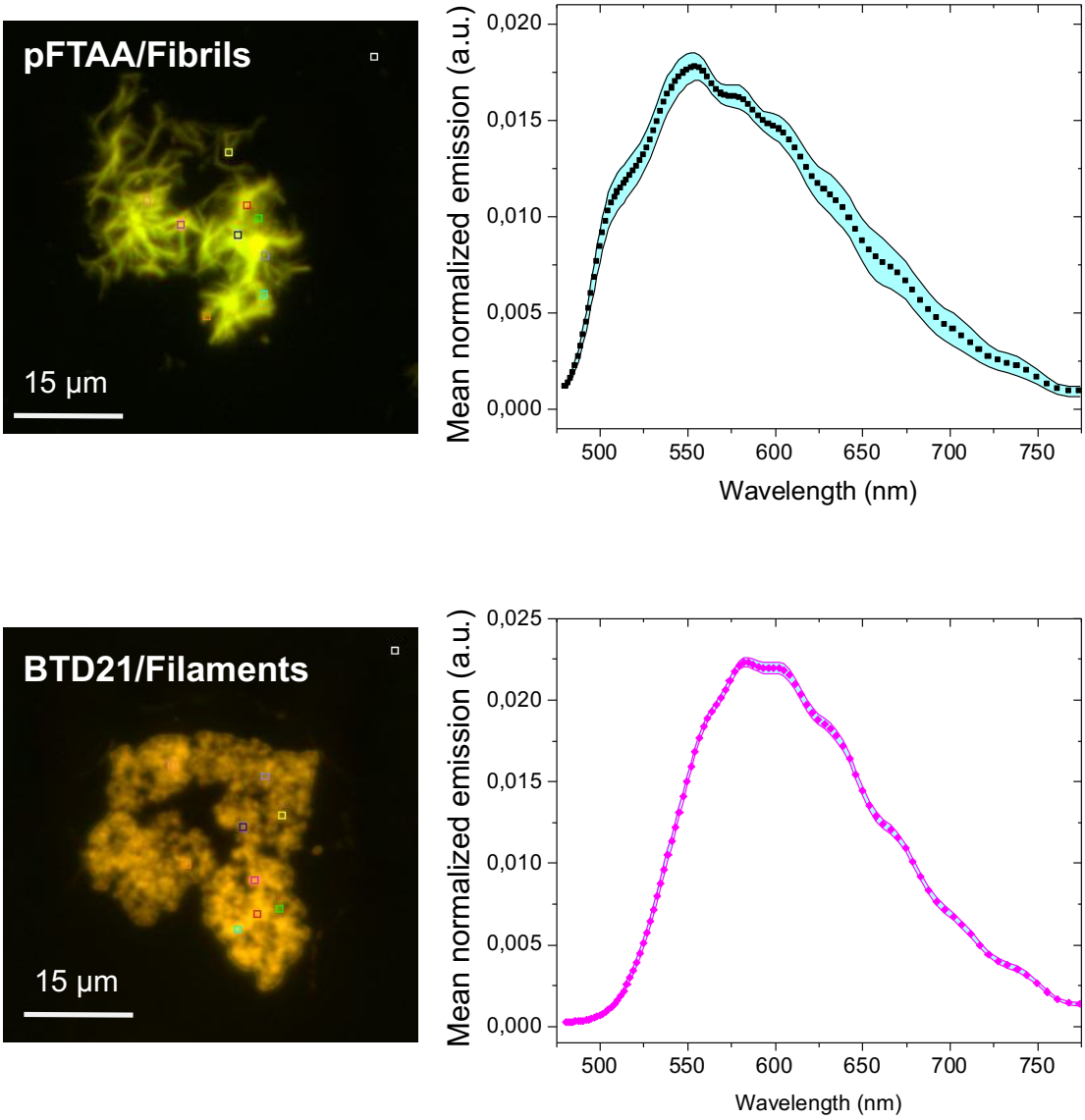

Figure S3 Hyperspectral fluorescence images of stained *in vitro*-formed bovine insulin fibrils and filaments. The top panel shows fibrils stained with pFTAA. The lower panel shows filaments stained with BTD21. The excitation wavelength was 436 nm. To calculate the corresponding spectra (right panels), spectra of nine ROIs were first normalized to the area, then the average spectrum along with the standard deviation for each wavelength was calculated. The symbols represent the average spectrum and the bands surrounding the symbols indicated the variation of the normalized spectra in terms of the calculated standard deviation of the nine normalized components.