Supporting Information

BODIPY Biosensor to Detect and Drive self-assembly of Diphenylalanine

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Measurement and instrument

All starting materials were purchased from Sigma-Aldrich and Fisher Scientific Worldwide Inc, unless otherwise noted. ^{1}H-NMR spectra were recorded on a Bruker NMR 400 DRX Spectrometer at 400 MHz and referenced to the proton resonance resulting from incomplete deuteration of deuterated chloroform (δ 7.26). The mass spectra of samples were recorded using Thermo LTQ-Orbitrap XL ETD ESI-MS. Fluorescence experiments were performed using Perkin ElmerLS-55 Spectrofluorophotometer and Hitachi Fluorescence spectrophotometer F-7000. Nava Nano SEM 230 (FEI) was used for SEM analysis. All tests were in room temperature and under vacuum (2E-6 Torr). 50ul of sample solution was dropped onto silicon wafer, and the sample was left dry in ambient condition for overnight. The samples were coated with a thin Ir (iridium) film (7nm in thickness) with a sputter Coater (208HR High Resolution Sputter Coater, Ted Pella Inc.) The confocal images were obtained using Nikon C2+ laser-scanning confocal microscope (Nikon, Tokyo, Japan) and Olympus FV3000 Confocal Imaging System (Olympus, Tokyo, Japan). AFM Multimode 8 with a top-mounted optical microscope was used to attach 5μm micro-particles on AFM cantilevers to measure intermolecular forces between probe X1 and FF. MLCT and NP-O10 AFM cantilever (Bruker) were used with an average spring constant of 0.01N/m and 0.06N/m, respectively. FF was grown on mica. Force curves were acquired in contact mode with a set-point force about 1nN. Both spring constant and sensitivity of the probe could be calibrated under thermal tuning condition using controlling software. Force curves were interpreted with a data analysis software NanoScope Analysis 1.40 (Bruker).

Synthesis of green fluorescent BODIPYs (compound X1, X2). BODIPYs were synthesized as described in our previous work.[1,2] Briefly, to a mixture of aldehyde compound (10 mmol) and 2,4-dimethypyrrole (20 mmol) in CH_2Cl_2 (500 mL), trifluoroacetic acid (2.47 mmol) was added under nitrogen. The reaction mixture was stirred for 4h at room temperature, then 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ, 10 mmol) in the mixed solvents of CH_2Cl_2 (25 mL) and tetrahydrofuran (25 mL) was added and stirred for 1h. Then Et_3N (12 mL) and BF_3•Et_2O (12 mL) were added dropwise under ice-cold conditions. The mixture was stirred for 30 min before warming up to room temperature, and was stirred for additional 3h at room temperature. The reaction mixture was washed with water (200 mL) for three times, and the organic layers were combined and dried over anhydrous $MgSO_4$. The solvent was evaporated in vacuum, and the residue was purified by column chromatography (Silica gel, eluent: CH_2Cl_2) to obtain powder. The target product was confirmed by NMR and MS. Compound X1: ^{1}H NMR (300 MHz, DMSO-D_6): δ = 1.95 (s, 6H), 2.29 (s, 6H), 5.98 (s, 2H), 7.31-7.37 (m, 5 H); TOF MS EI^+ (nature of the peak) calcd: 324.2. Found: 324.3. Compound X2: ^{1}H NMR (300 MHz, DMSO-D_6): δ = 1.48 (s, 6H), 2.32 (s, 6H), 3.08 (s, 6H), 6.02 (s, 2H), 6.80 (d, J = 8.8 Hz, 2 H), 7.18 (d, J = 8.8 Hz, 2 H); TOF MS EI+ (nature of the peak) calcd: 367.2. Found: 367.9.

Synthesis of near-infrared fluorescent BODIPYs (compound X3). The green fluorescent BODIPY was further modified by aldehyde compound. Specific steps are as follows: green fluorescent BODIPY (0.54 mmol), aldehyde compound (0.54 mmol, 1 equiv or 1.08 mmol, 2 equiv), and p-toluenesulfonamide (PTSA) (0.01 mmol) were dissolved in toluene (25 mL) and piperidine (1 mL) in a round-bottom flask equipped with a Dean−Stark apparatus. The resulting solution was heated at 140 °C until all the solvents were collected by the Dean−Stark apparatus.

Toluene (25 mL) and piperidine (1 mL) were added to the solid reaction media and the drying protocol was repeated four times. The resultant materials were then purified by chromatography on silica gel (1:3 dichloromethane/petroleum ether) to obtain dark solid product. The target product was confirmed by NMR and MS. ^{1}H NMR (300 MHz, DMSO-D_6): δ = 7.54 (d, J = 8.8 Hz, 2H), 7.18 (d, J = 8.8 Hz, 2 H), 6.75-6.79 (m, 5H), 6.02 (s, 1H), 5.65-5.67 (m, 2H), 3.11 (s, 12H), 2.12 (s, 9H); TOF MS EI^+ (nature of the peak): 498.3 (calculated)/499.1 (observed).

Self-assembly of FF and BODIPY-labelled FF. Self-assembly of FF was constructed using the method as described previously.[3] For self-assembly of BODIPY-labelled FF, two methods were used to prepare samples, namely Process I and Process II as detailed below.

Self-assembly of FF. FF (3 mg) was dissolved in 45 μL of 1,1,3,3,6,6-hexafluoro-2-propanol (HFP), and then 105 μL of water was added into the solution, followed by ultrasonic treatment at room temperature for 5 ~ 10 min. As a result, FF was self-assembled into nanofibers. When the HFP was completely evaporated, the FF nanofibers in water was stored for later use.

Process I. Firstly, FF assembly was obtained as described above. Then, 1 mg of BODIPY (in 45 μL HFP) was added into FF nanofibers (in water). As a result, flexible fluorescent labelled nanofibers were obtained. When the HFP was completely evaporated, the BODIPY-labelled FF assembly in water was stored for later use.

Process II. 1 mg of BODIPY and 3 mg of FF were dissolved in 45 μL HFP, and the synthesis procedure was the same as described as self-assembly of FF. As a result, rigid straight nanofibers were obtained. When the HFP was completely evaporated, the BODIPY-labelled FF assembly in water was stored for later use.

Reference

1 L. Quan, W. H. Lin, T. T. Sun, X. G. Guan, M. Zheng, Z. G. Xie, X. B. Jing, *J. Fluoresc.*, 2014, **24**, 841.
2 L. Quan, W. H. Lin, T. T. Sun, Z. G. Xie, Y. B. Huang, X. B. Jing, *Catal. Lett.,* 2014, **144**, 308.
3 M. M. Fu, Q. Li, B. B. Sun, Y. Yang, L. Dai, T. Nylander, J. B. Li, *ACS Nano,* 2017, **11**, 7349.

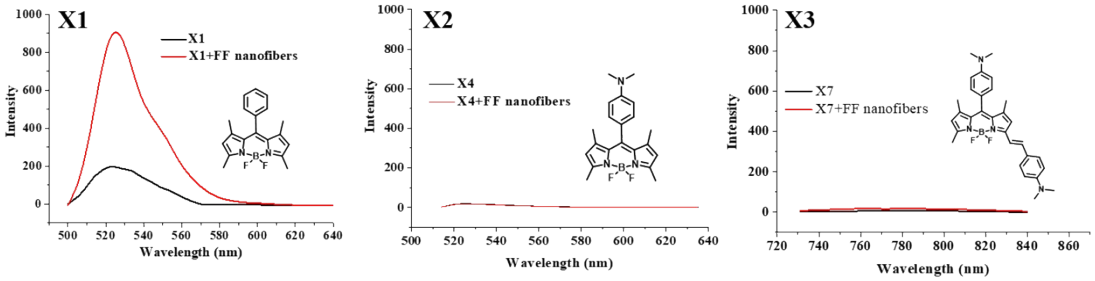

Figure S1. BODIPY fluorescence response to FF nanofibers under the same concentrations of BODIPY probes (0.05mg/ml, 1ml) and FF nanofibers (0.22mg/ml, 10μl).

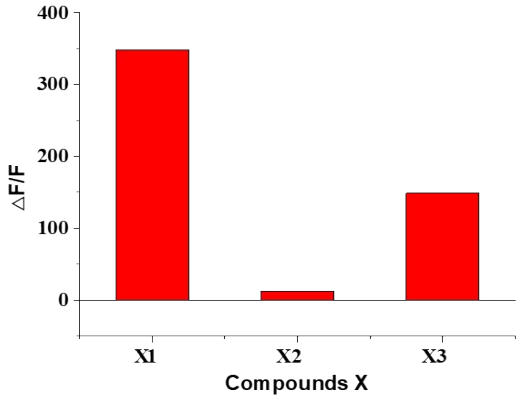

Figure S2. Folds of increased intensity of fluorescence from **Figure 1** when BODIPY binds to FF nanofibers.

Table S1 Detail data from Figure S1.

Sample	λ_{max}	Fluorescence Intensity (a.u.)
X1	523 nm	197
X1+FF nanofibers	526 nm	904

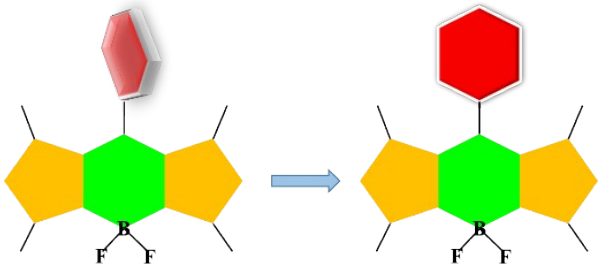

Figure S3. Schematic diagram of molecular structure planarization of BODIPY response to FF monomers/FF nanofibers.

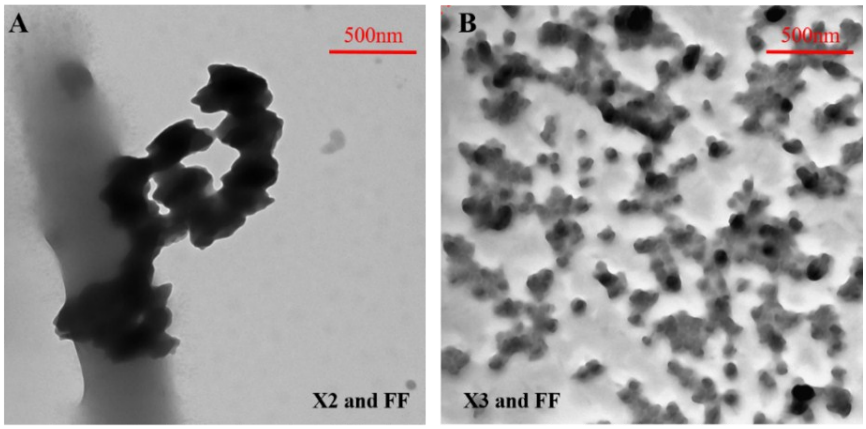

Figure S4. SEM of co-assembly of FF-X2 (A) and FF-X3 (B) using the mixture solution of FF and X2/X3 on silicon wafer.

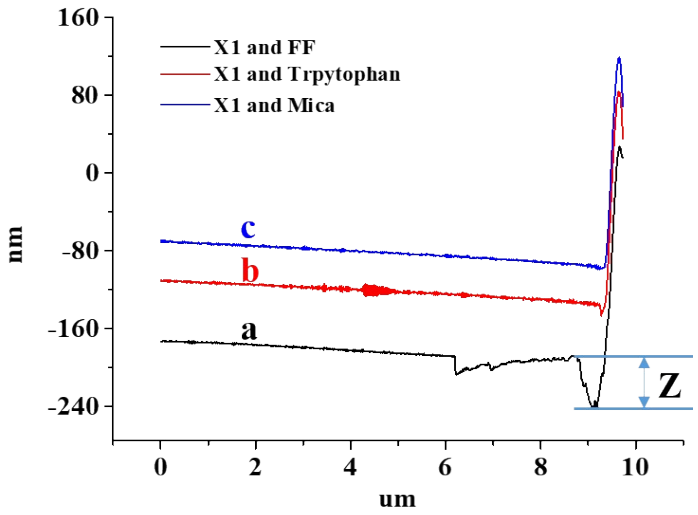

Figure S5. (a-c): Histograms of the adhesion forces between BODIPY and FF (a, 0.53nN) or Trpytophan (b, 0.075nN) or Mica (c, 0.002nN). F = K·Z, F means force; K = 0.01N/M; Z means the distance (between Baseline and peak valley).