Electronic Supplementary Information

Ratiometric Detection of HSA Using Aggregation Induced Enhanced Emission Conjugated Polyelectrolyte and Prototype Smartphone Device

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EXPERIMENTAL SECTION

Materials and methods:

The reagents 9,10 dibromoanthracene, 4 formyl phenyl boronic acid, 4 bromophenyl acetonitrile, were procured from TCI chemicals and bis-(pinacolato)diborane, dppf-1,1'-bis(diphenylphosphanyl)ferrocene, tetra butyl ammonium iodide (TBAI), 1,6-dibromohexane, 2, 7 dibromo fluorine, Tetrakis(triphenylphosphine)palladium(0) [$(Pd(PPh_3)_4(0$] catalyst.were procured from Sigma-Aldrich. THF, DMSO and all the organic solvents used in the study are of HPLC grade and procured from Merck, Human serum albumin (HSA), Bovine serum albumin (BSA), Immunoglobin and all the interfering analytes used in the selectivity study were purchased from were purchased from Sigma-Aldrich. Milli-Q water has been used for the preparation of the stock solution for all the experiments.

Recording of ^{1}H NMR (500 MHz, 600 MHz) and ^{13}C NMR (125MHz, 150 MHz) spectra were performed on the Varian-AS600 NMR spectrometer. For recording UV-Visible absorption spectra and photoluminescence spectra, Cary 60 UV-Vis Spectrophotometer and Horiba Fluoromax-4 spectrofluorometers have been used with 1 ml quartz cuvettes (path length =1 cm, slit = 3 nm). FESEM images were obtained from the Zeiss Sigma Field Emission Scanning Electron Microscope (FESEM). Lifetime and quantum measurement were done in Edinburgh FLS1000 Photoluminescence Spectrometer Dynamic light scattering and Zeta potential measurements were done DLS spectrometer Malvern Nano ZS90. DFT study is perform for monomer in Gaussian09 and for polymer Gaussian16. Docking study was done in AutoDockTools-1.5.7

Photoluminescence quantum yield (ϕ).

The absolute quantum yield for P1, P2, P3 and PFAN polymers were determined by using Edinburgh FLS 1000 instrument through integrated sphere technique.

Limit of detection (LOD):

The lowest LoD was determined by analyzing the fluorescence spectra of PFAN with varying concentrations of HSA (0-0.25 μg mL^{-1}) and BSA (0-0.25 μg mL^{-1}) in water medium. Using the standard formula, LoD=3.3σ/k, the lowest LoD was calculated from the calibration curve, where the symbols σ and k represent the standard deviation of the blank (only polymer before addition of analyte) and calibration plot's slope respectively.

Cell Viability Assay:

MTT assay was performed to evaluate the cytotoxicity of the polymer PFAN using HEK 293 cell. Cells were seeded into a 96-well plate at the density of 10,000 cells/well and incubated for 24 h. Cells were washed with 1X PBS and followed by the treatment of series of different concentration of polymer PFAN at 0, 10, 20, 30, 40, 50, 60, 50, 60, 70, 80, 90 and 100 μg/mL for 24 h at 37 °C in incubator. Cells were washed with 1X PBS on incubation of 24 h. 10 μL of MTT of stock concentration 5 mg mL^{-1} and 90 μL DMEM medium was added to each well and incubated for 4 hours at 37 °C in incubator. After incubation for 4 h, 75 μL of medium was removed and 50 μL of DMSO was added to each well followed by further incubation of 10 minutes. The multimode microplate reader (Thermofisher Varioskan LUX) was used to record absorbance at wavelength of 490 nm. All the experiments were carried out in triplicate.

Molecular docking:

The purpose of molecular docking was to analyze the stable conformation of the probe while it is attached to a protein, focusing on its geometric and energetic properties. The software AutoDockTools-1.5.7 was utilized to produce a docked arrangement of PFAN monomer with HSA and BSA. This process involves the application of a genetic algorithm (GA) and a Lamarckian genetic algorithm to generate several conformations. The program was utilized to examine the binding free energies and binding locations of PFAN monomer within the active site of HSA/BSA. The crystal structure of HSA and BSA was obtained from RCSB PDB

(4K2C and 3V03) and underwent energy refinement, hydrogen additions, and solvent removal using AutoDockTools-1.5.7 to ensure that all residues achieved a proper and stable conformation. The Lamarckian GA (dpf file) was chosen as the output and utilized in AutoDockTools-1.5.7 to conduct docking simulations. The process of minimizing and optimizing the energy of PFAN monomer was also carried out. Ultimately, the analysis of the docking result of the PFAN monomer was conducted using Discovery Studio Visualizer programmes.[1] The docking structure with the lowest binding energy, as determined by AutoDockTools-1.5.7, was selected as the optimal binding conformation.

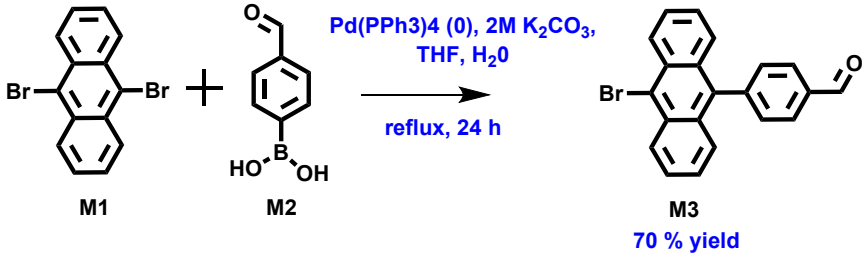
Caution for handling of serum sample:

Blood serum samples must be handled with care during the experiment. Human blood serum, particularly that of patients, can be highly dangerous if exposed; therefore, necessary precautions must be followed when handling them. The serum samples were kept in a freezer at -20 °C. To ensure safety, disposable gloves were worn during all fluorescence studies.

Serum Sample Analysis:

Fresh serum samples were collected from IIT G Hospital and diluted 100 times with Milli Q water to estimate HSA using the standard addition method. The serum samples were then spiked with 0 mg mL^{-1}, 0.5 mg mL^{-1}, 1.5 mg mL^{-1}, and 3.0 mg mL^{-1} of HSA to make four distinct analytical samples. To record fluorescence spectra, 10 µl of each sample was introduced in a 1ml cuvette containing PFAN polymer. Each measurement was taken three times, and the amount of HSA was calculated using the Standard Recovery Technique.

4-(10-bromoanthracen-9-yl)benzaldehyde (M3):

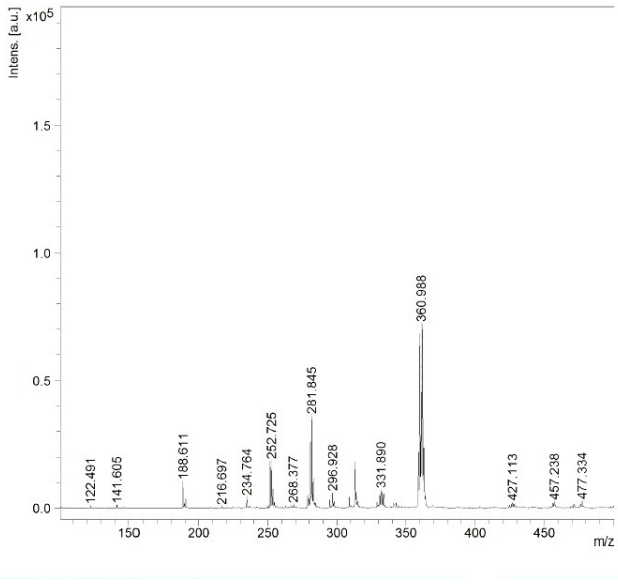


The monomer (M3) was synthesized by Suzuki Coupling reaction. A mixture of monomers 9,10 dibromoanthracene (M1) (500 mg, 1.49 mmol) and 4 formyl phenyl boronic acid (M2) (268.4 mg, 1.79 mmol) were taken in a clean RB flask fitted with water condenser and then made the system inert by degassing followed by purging with argon gas. Thereafter, 5mg of catalyst $Pd(PPh_3)_4$ (0) were added maintaining the inert condition. A mixture of THF (9 ml) and 2M K_2CO_3 (3 ml) were injected into the reaction mixture and were made inert by applying free-thaw degassing technique (3 times). The reaction mixture was stirred at reflux condition under an argon atmosphere and the progress of reaction were monitored by TLC. After 24 h., the reaction was stopped and extracted with $CHCl_3$/water. The organic layer was dried under vacuum and it was purified by column chromatography by 30 % $CHCl_3$/hexane solvent fraction. (yellow color solid, Yield = 70 %, 430 mg).

MALDI_TOF: The mass of monomer M3 (m/z =360.988)

^{1}H NMR (500 MHz, $CDCl_3$, δ (ppm): 10.20 (s, 1H), 8.64 (d, *J* = 8.9 Hz, 2H), 8.12 (d, *J* = 8.0 Hz, 2H), 7.61 (t, *J* = 6.4 Hz, 4H), 7.55 (d, *J* = 8.8 Hz, 2H), 7.40 (dd, *J* = 8.0, 6.9 Hz, 2H).

^{13}C NMR (150 MHz, $CDCl_3$, δ (ppm): 191.95, 145.29, 135.95, 135.86, 132.02, 130.02, 130.57, 130.19, 129.85, 128.09, 127.10, 126.72, 126.08, 123.64.



m/z	S/N	Quality Fac.	Res.	Intens.	Area
101.124	12	71.7	209	72.9	
122.491	43	585	780	242	
141.605	50	731	1145	329	
146.751	17	202	421	161	
188.611	279	224	10395	5587	
208.697	6	674	350	212	
216.697	8	886	547	314	
225.616	3	51.4	220	470	
228.694	6	52.5	409	470	
234.764	45	1410	4244	2299	
241.990	6	794	606	404	
252.725	145	193	14962	17442	
268.377	11	119	1437	1970	
281.845	258	212	36233	38951	
296.928	40	129	5970	6325	
331.890	40	99.9	6992	14958	
360.988	227	147	45648	133385	
427.113	9	102	2240	5537	
457.238	8	298	2423	4641	
471.405	5	208	1685	2521	
477.334	8	361	2753	3499	
505.336	9	118	3902	14256	
534.424	41	162	19735	52273	
550.452	5	212	2626	14285	
562.469	79	255	41187	85022	
585.443	8	69.9	4217	22301	
595.520	12	182	6054	15423	
614.480	59	173	28843	94304	
627.526	5	144	2489	10173	
642.547	54	151	25111	96803	
664.443	5	270	2184	8730	

Figure S1: MALDI-TOF spectra for monomer M3

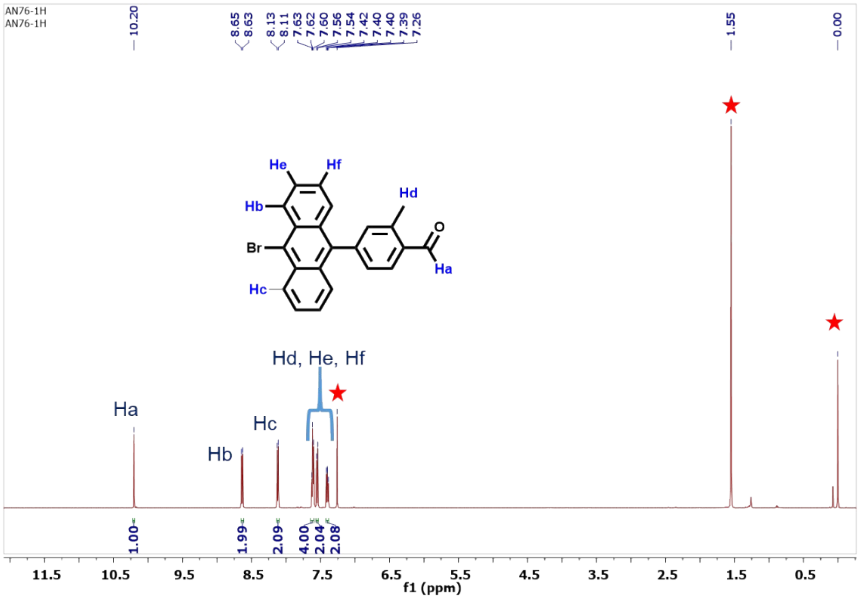


Figure S2: ^{1}HNMR of M3 in $CDCl_3$

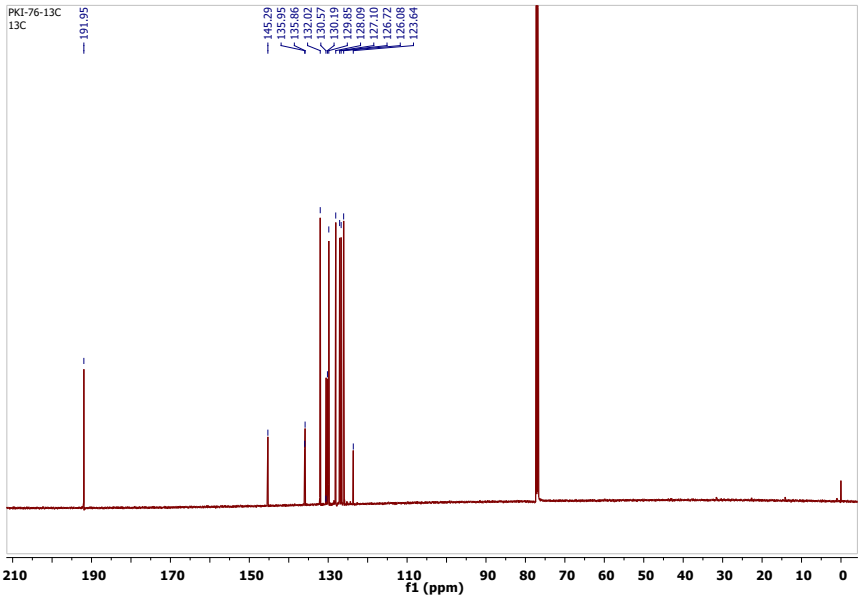
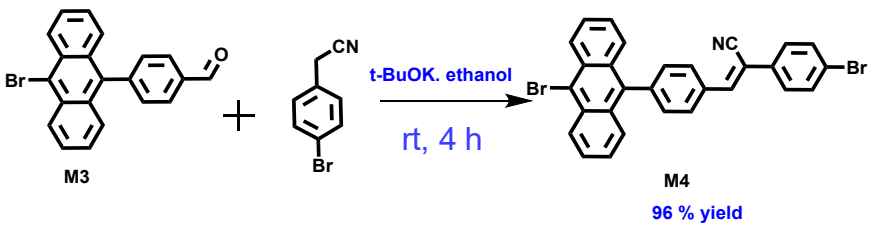


Figure S3: ^{13}C NMR spectra of M3 in $CDCl_3$

(Z)-3-(4-(10-bromoanthracen-9-yl)phenyl)-2-(4-bromophenyl)acrylonitrile (M4):



The monomer M4 was synthesized by Knoevenagel Condensation reaction. The monomer M3 (50 mg, 0.138 mmol) and 4 bromophenyl acetonitrile were solubilised in 20 ml ethanol at 50 °C with continuous stirring. 23 mg of Potassium tertiary butoxide (t-BuOK) was solubilised in 2 ml ethanol and then injected into the reaction mixture dropwise. After the product gets precipitated as yellowish precipitate from the reaction mixture. The progress of the reaction was monitored by TLC and after 4 h the precipitate was filtered through Whatman filter paper and then wash repeatedly by ethanol to get the pure amorphous yellow ppt. (yield= 96 %, 72 mg).

MALDI_TOF: The mass of monomer M4 (m/z =538.909)

^{1}H NMR (600 MHz, $CDCl_3$, δ (ppm): 8.63 (s, 2H), 8.12 (d, J = 8.0 Hz, 2H), 7.69 (s, 1H), 7.64 (d, J = 8.4 Hz, 6H), 7.61 (d, J = 7.9 Hz, 2H), 7.54 (d, J = 8.1 Hz, 2H), 7.43 – 7.40 (m, 2H).

^{13}C NMR (150 MHz, $CDCl_3$, δ (ppm): 142.24, 141.39, 132.98, 130.22, 129.44, 128.08, 127.56, 127.07, 126.94, 125.95, 123.66, 117.68, 111.04

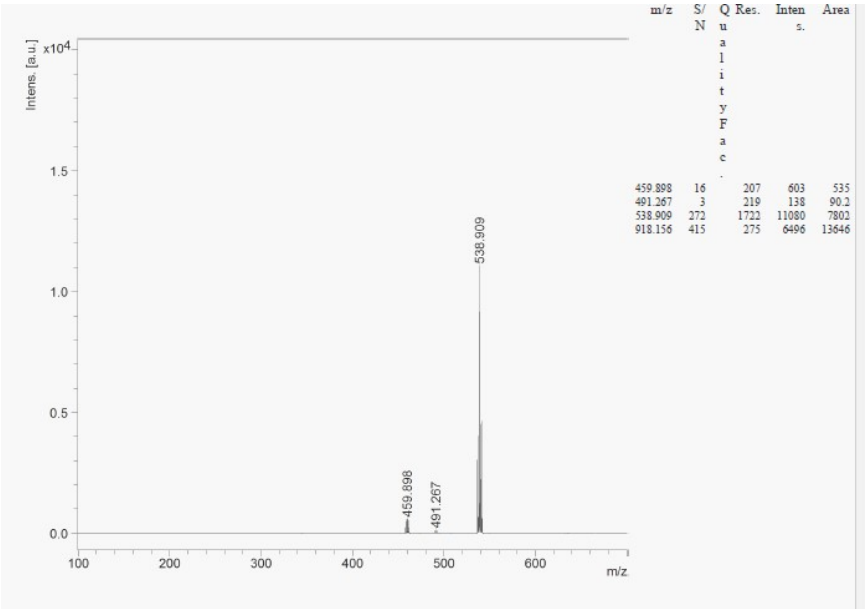


Figure S4: MALDI-TOF spectra for monomer M4.

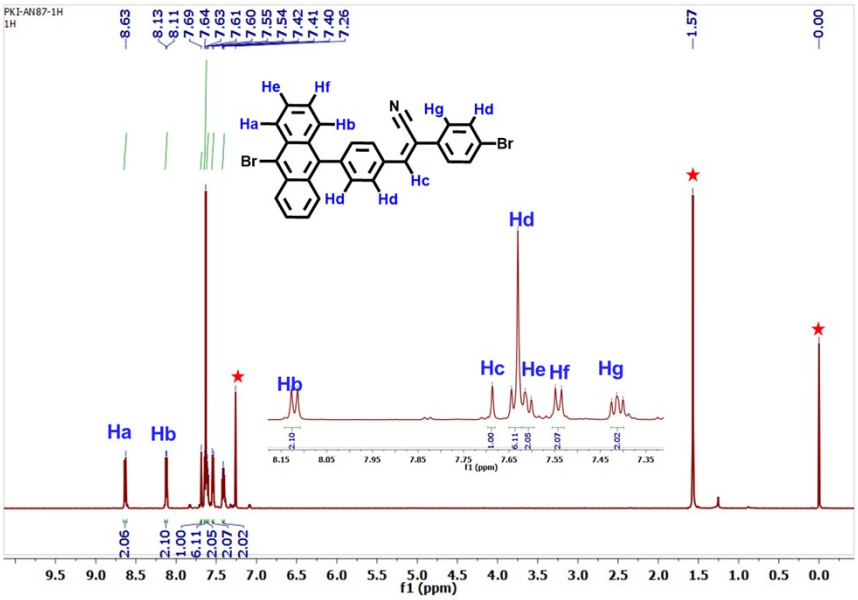


Figure S5: ^{1}H NMR of M4 in $CDCl_3$

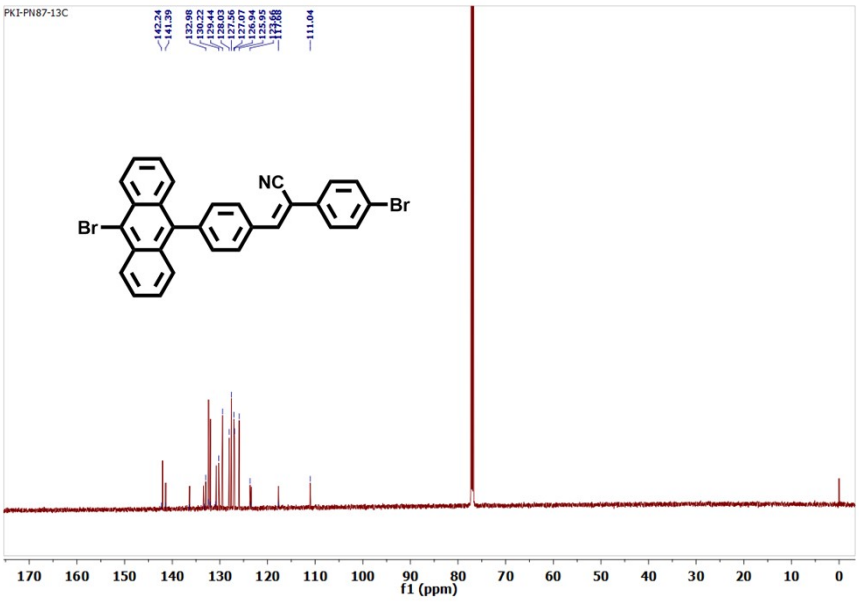
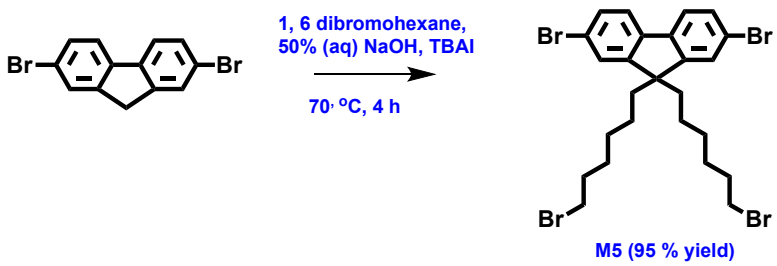


Figure S6: ^{13}C NMR spectra of M4 in $CDCl_3$.

2,7-dibromo-9,9-bis(6-bromohexyl)-9H-fluorene (M5):



The synthesis of the monomers M5 was done by following the already reported procedures.[2] The starting material 2, 7 dibromo fluorene (1g, 3.106 mmol), and tetra butyl ammonium iodide (TBAI) (0.229 g, 0.621 mmol) were taken in a clean RB flask and made inert condition by applying free-thaw and degassing technique (3 times). After that, 1,6-dibromohexane (2.82 mL, 18.636 mmol) followed by 50 % NaOH were injected cautiously through a syringe, maintaining the inert condition. Finally, the reaction was performed at 80 °C with proper stirring for 5 h. TLC in hexane was used to monitor reaction completion, then separated the organic part from water in a separating funnel with water/chloroform mixture. The excess 1,6-dibromohexane from the recovered organic layer dried by Kugelrohr and further purification was done by column chromatography with hexane as eluent to obtain the final product as white crystalline solid. (Yield = 95 %, 1.92 g)

^{1}H NMR (600 MHz, $CDCl_3$, δ(ppm): 7.52-7.54(d, 2H), 7.43-7.48 (m, 4H), 3.28-3.31 (t, 4H), 1.90-1.95 (p, 4H), 1.64-1.71(p, 4H), 1.17-1.24(p, 4H), 1.05-1.12(p, 4H), 0.56-0.63(4H).

^{13}C NMR (150 MHz, $CDCl_3$, δ (ppm): 152.19, 139.09, 130.05, 126.12, 121.58, 121.53, 55.57, 40.03, 32.79, 32.61, 28.95, 27.75, 23.47.

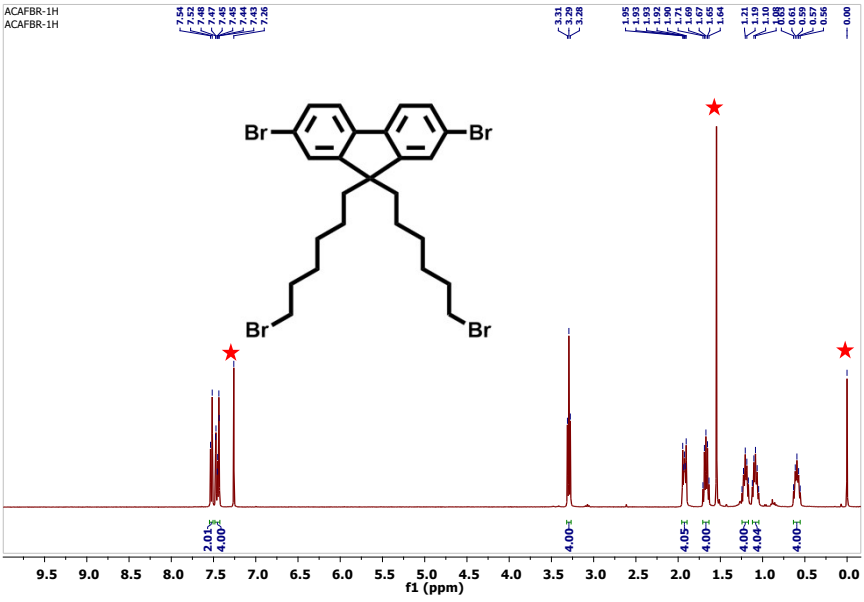


Figure S7: ^{1}H NMR spectra of M5 in $CDCl_3$.

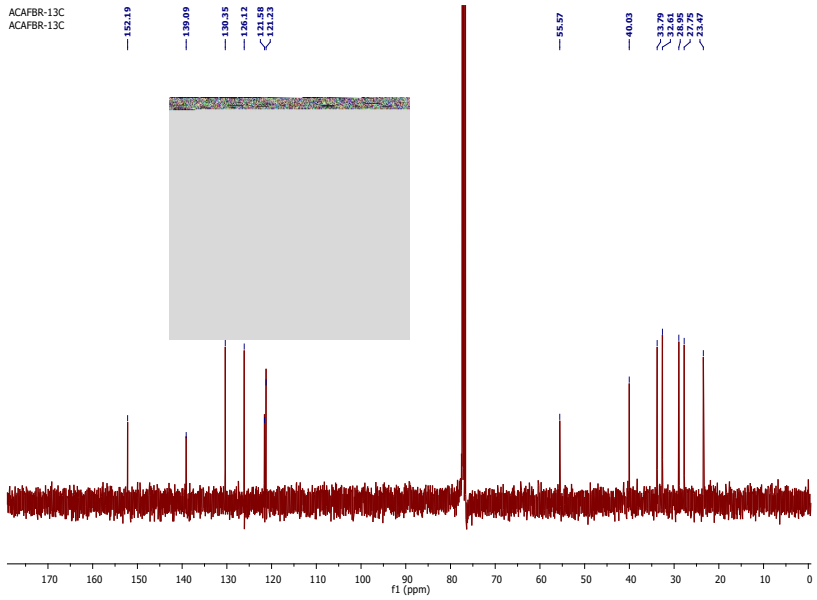
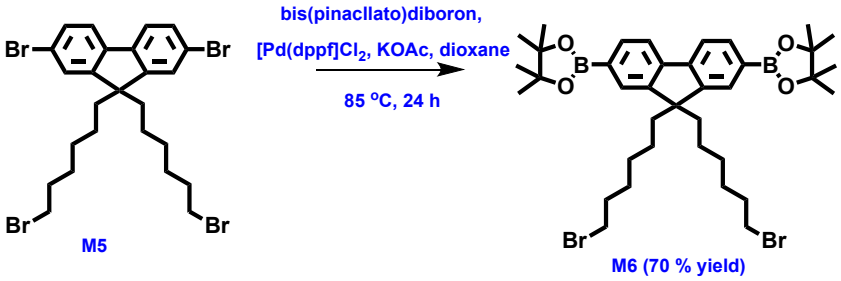


Figure S8: ^{13}C NMR spectra of M5

2,7-bis[9,9′-bis(6″-bromohexyl)fluorenyl]-4,4,5,5-tetramethyl[1,3,2]dioxaborolane (M6):



The mixture monomer M5 (1g, 1.5 mmol), bis-(pinacolato)diborane (0.95 g, 3.75 mmol) and KOAc (1g, 10 mmol were taken in a clean 250 ml RB flask. The mixture is degassed and then 5mg of [Pd(dppf)Cl_2] (dppf-1,1'-bis(diphenylphosphanyl)ferrocene) was added cautiously followed by 12 ml 1, 4 dioxane (dry). The reaction stirred properly at w 85 °C for 24 h. Then the mixture was dried at high vacuum and then extracted the organic soluble part by using ethyl acetate/ water mixture. Further purification was done by flash chromatography using 5 % ethyl acetate hexane mixture as eluent to get. M6 as white crystals (0.63 g, 66 %)

^{1}H NMR (600 MHz, $CDCl_3$, δ(ppm): 7.73-7.75(d, 2H), 7.65-7.66 (m, 4H), 3.23-3.27 (t, 4H), 1.98-2,05 (p, 4H), 1.57-1.67 (p, 4H), 1.39 (s, 24H), 1.11-1.16(p, 4H), 1.00-1.07 (p, 4H), 0.50-0.57(4H).

^{13}C NMR (150 MHz, $CDCl_3$, δ (ppm): 150.17, 143.50, 133.90, 129.12, 119.94, 83.56, 55.13, 40.17, 34.06, 32.49, 29.32, 27.38, 24.50, 23.57

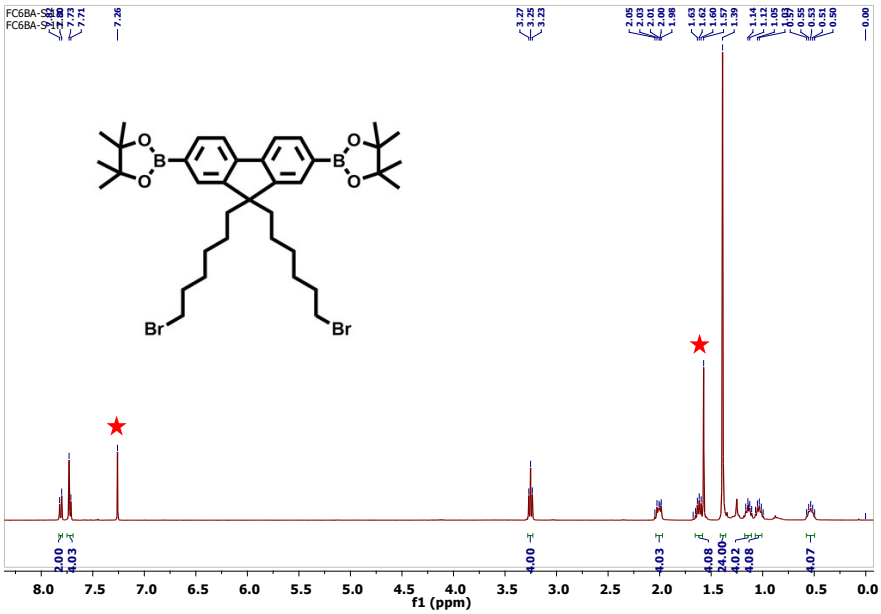


Figure S9: 1H NMR spectra of M6.

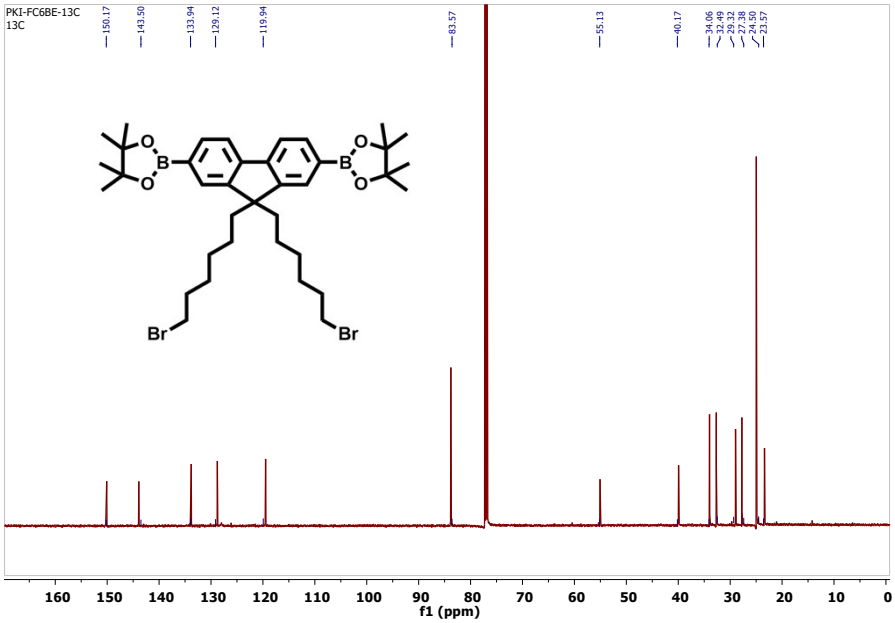


Figure S10: ^{13}C NMR spectra of M6 in $CDCl_3$.

Synthesis of P1, P2 and P3 polymers:

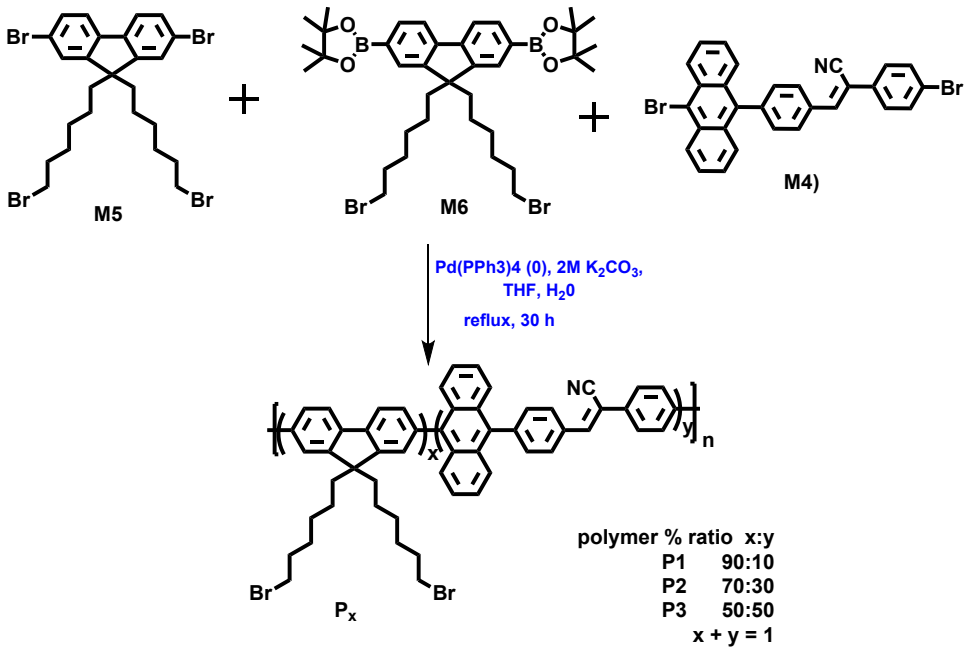


Table S1: The molar ratio of monomers used in polymerization of P1, P2 and P3 along with their yield (MW)

Polymer	Mole fractions of monomers taken			Yield	MW	PDI
	M4 MW 539.27 gmole^{-1}	M5 MW 650.13 gmole^{-1}	M6 MW 744.26 gmole^{-1}	(%)	(gmole^{-1})	
P1	43.47 mg (0.08mmoles)	210 mg (0.322mmoles)	300 mg (0.403mmoles)	470 mg (85 %)	16688	1.60
P2	130.39 mg (0.242 mmole)	104.8 mg (0.161mmoles)	300 mg (0.403mmoles)	430 mg (80 %)	14391	1.54
P3	217.37 mg (0.403 mmoles)	0	300 mg (0.403mmoles)	450 mg (87 %)	15324	1.78

They were synthesized by Suzuki Coupling reaction by taking the required molar ratio of monomers M4, M5 and M6 as given in Table S1. A mixture of monomers M4, M5 and M6 were taken in a clean RB flask fitted with a condenser and then made the system inert by degassing followed by purging with argon gas. Thereafter, 5 mg of catalyst $Pd(PPh_3)_4$ (0) were added maintaining the inert condition. A mixture of THF (9 mL) and 2M K_2CO_3 (3mL) were injected into the reaction mixture and were made inert by applying free-thaw degassing technique (3 times). The reaction mixture was stirred at reflux condition under an argon atmosphere. After 30 h., the reaction the reaction was cool down and extracted with $CHCl_3$/water. The organic layer was dried under vacuum and purified repeated precipitation in methanol and acetone. The molecular weight (MW) of polymers were determined by GPC using polystyrene as a standard.

For P1:

^{1}H NMR (600 MHz, $CDCl_3$, δ(ppm): 7.87 (b), 7.73-7.69 (b), 7.60-7.51 (b), 7.12-6.91 (b) 3.0 (b), 2.19 (b)1.28-1.19 (b), 1.70 (b) 0.84 (b).

For P2:

^{1}H NMR (600 MHz, $CDCl_3$, δ(ppm): 8.11-7.95 (b), 7.75-7.57 (b), 7.37-7.31 (b), 7.06 (b), 7.84 (b), 3.22 (b), 2.05 (b)1.62 (b), 1.09 (b) 0.76 (b).

For P3:

^{1}H NMR (600 MHz, $CDCl_3$, δ(ppm): 8.17-8.01 (b), 7.82-7.77 (b), 7.62 (b), 7.49 (b), 7.36 (b), 3.25 (b), 1.98 (b)1.67 (b), 1.09 (b) 0.82-0.67 (b).

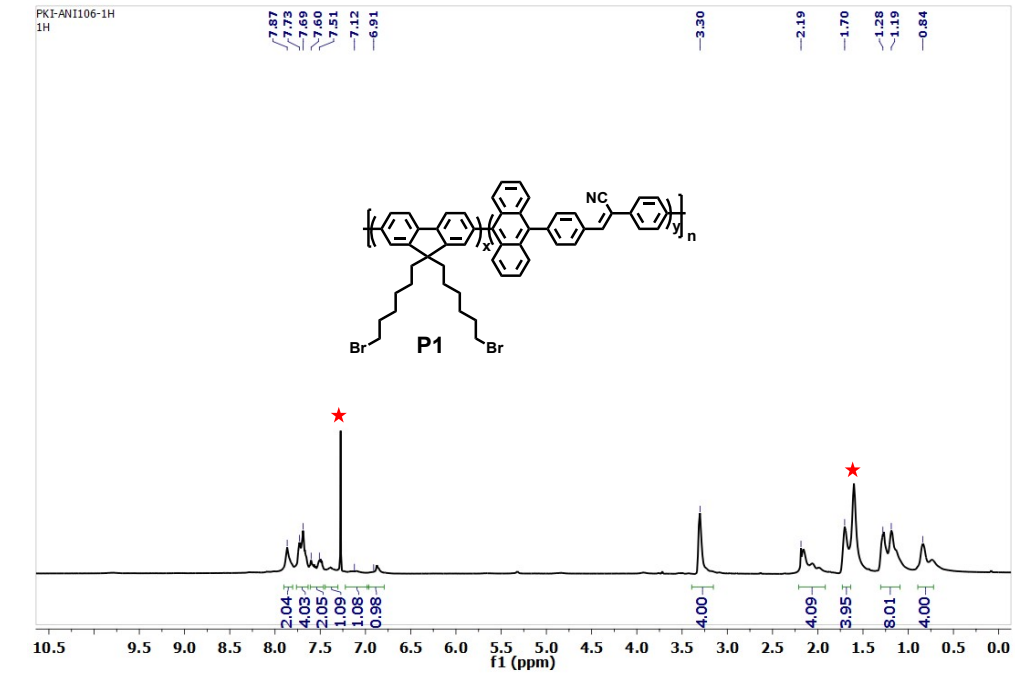


Figure S11: ^{1}H NMR spectra of P1 in $CDCl_3$.

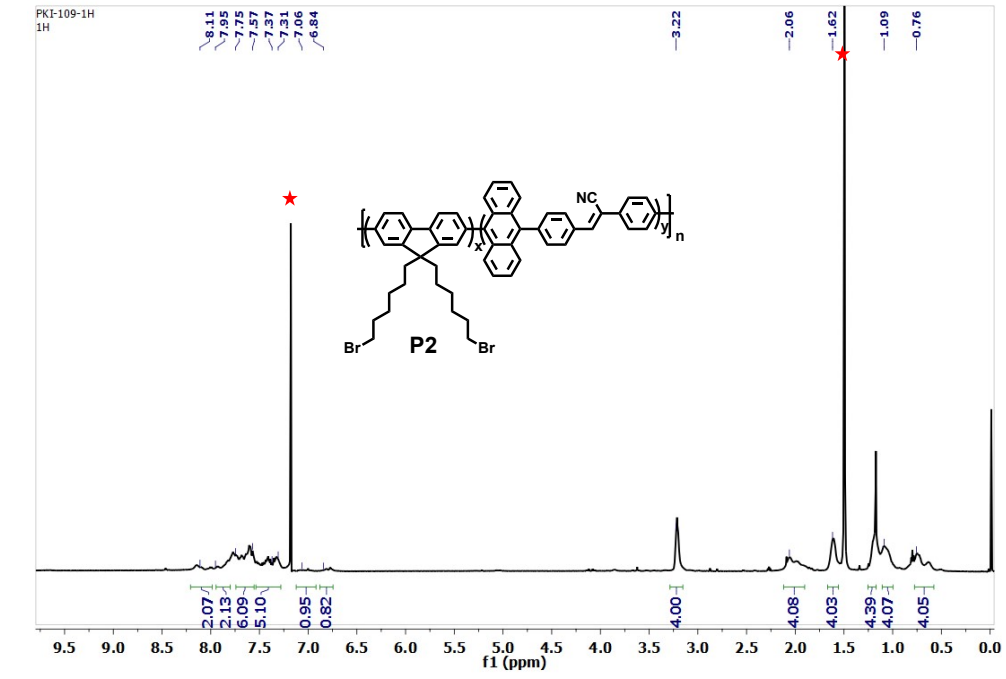


Figure S12: ^{1}H NMR spectra of P2 in $CDCl_3$.

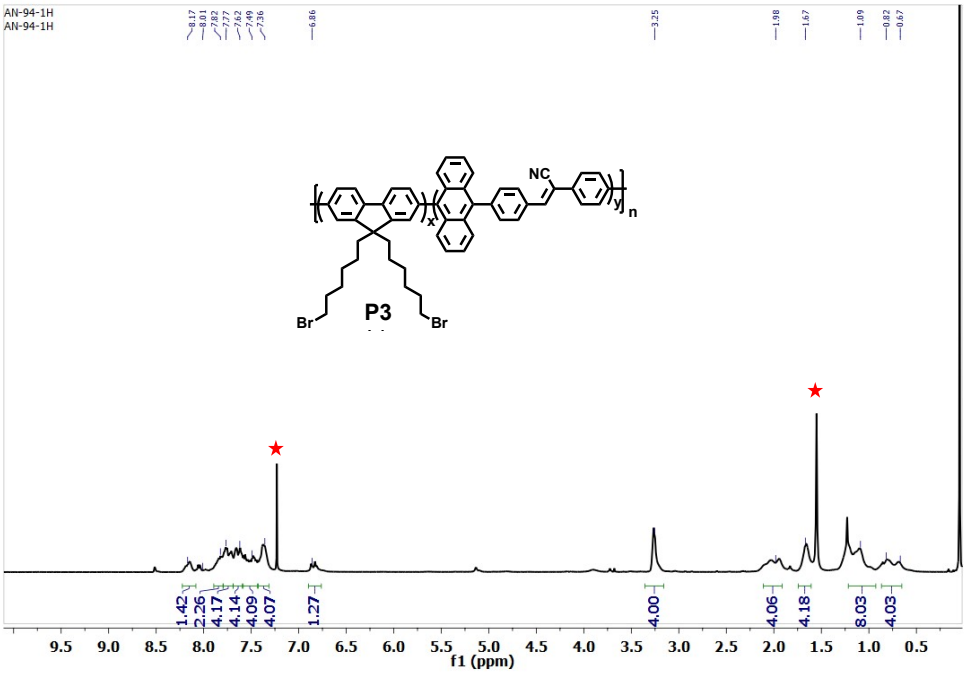
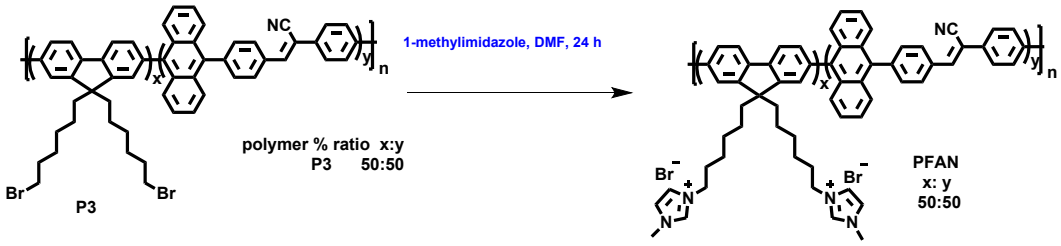


Figure S13: ^{1}H NMR spectra of P3 in $CDCl_3$.

(Z)-3,3'-((2-(10-(4-(2-cyano-2-(p-tolyl)vinyl)phenyl)anthracen-9-yl)-7-methyl-9H-fluorene-9,9-diyl)bis(hexane-6,1-diyl))bis(1-methyl-1H-imidazol-3-ium) (PFAN) :



The final polymer PFAN was synthesized by vigorous stirring of P3 (100 mg), 1-methyl imidazolium (1.5 ml) and K_2CO_3 (100 mg) in 2 mL of dry DMF for 24 h at 110 °C. After the completion of the reaction, DMF and excess 1-methyl imidazolium were removed by precipitation in diethyl ether. Then further reprecipitated from acetone followed by washing with THF. Finally, the precipitate was dried under vacuum to achieve the desired brownish-yellow polymer (PFAN). (Yield = 80 %, 80 mg)

^{1}H NMR (400 MHz, CDCl3, δ (ppm)): 8.99-9.03 (b), 8.28(b), 7.99 (b), 7.63-7.65 (b), 7.36-7.48(b), 3.77-4.01 (b), 2.47 (b), 1.56-1.58 (b), 1.04 (b), 0.58 (b)

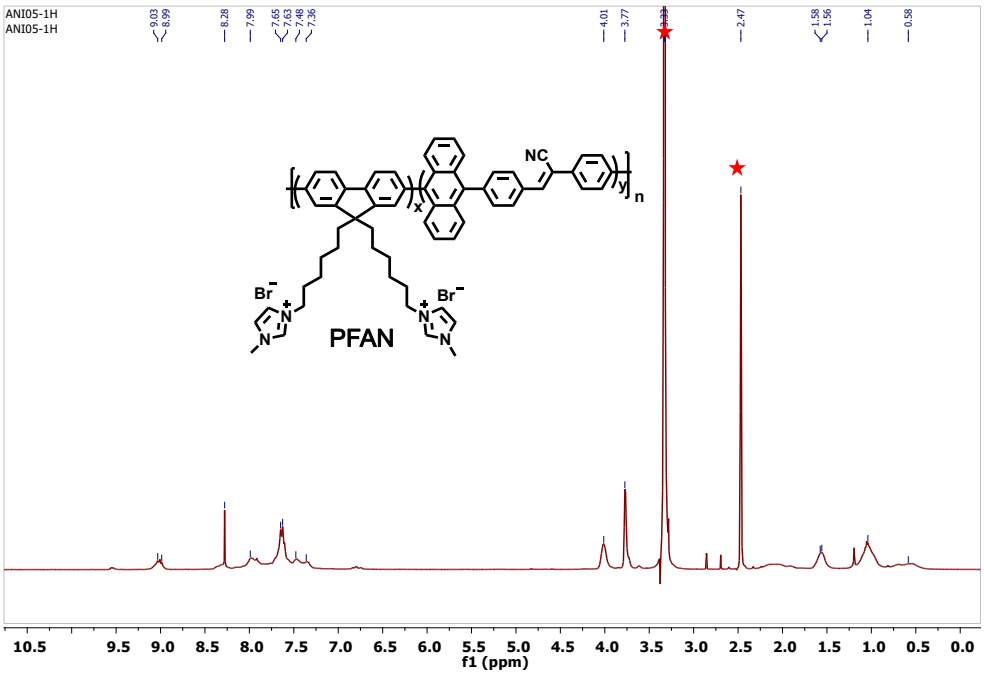


Figure S14: ^{1}H NMR spectra of PFAN in DMSO-d6.

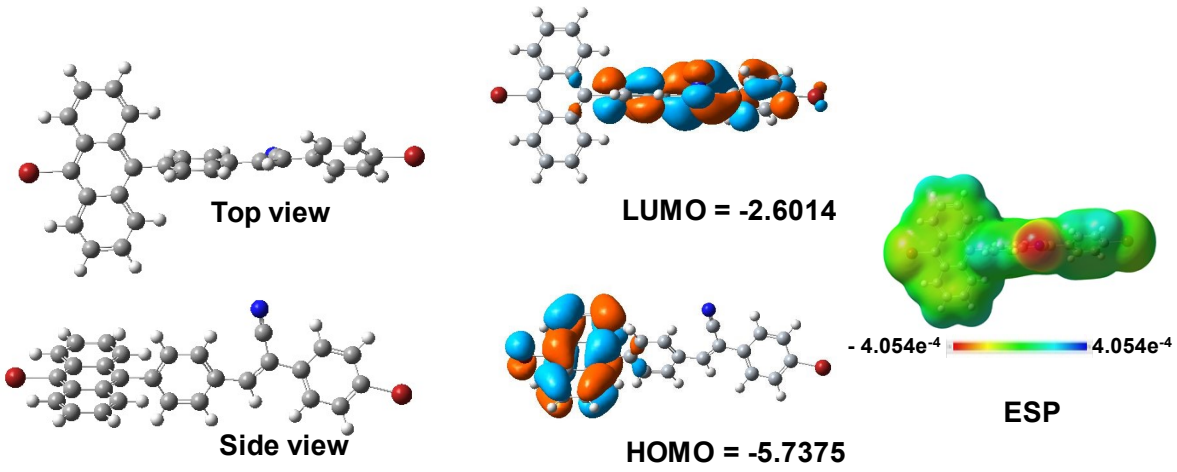


Figure S15: Optimized structure of M4 monomer using B3LYP and 6-31G (d.p) of Gaussian software.[3]

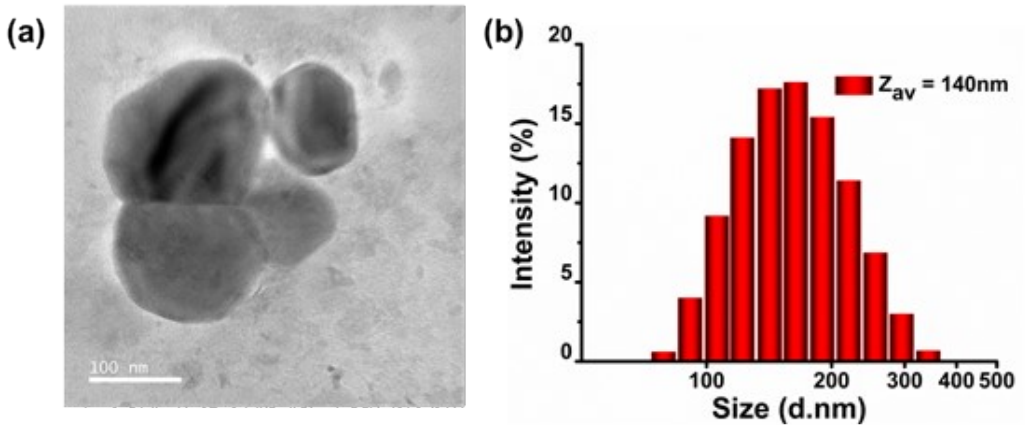


Figure S16: (a) TEM images and (b) DLS measurement of M4 monomer.

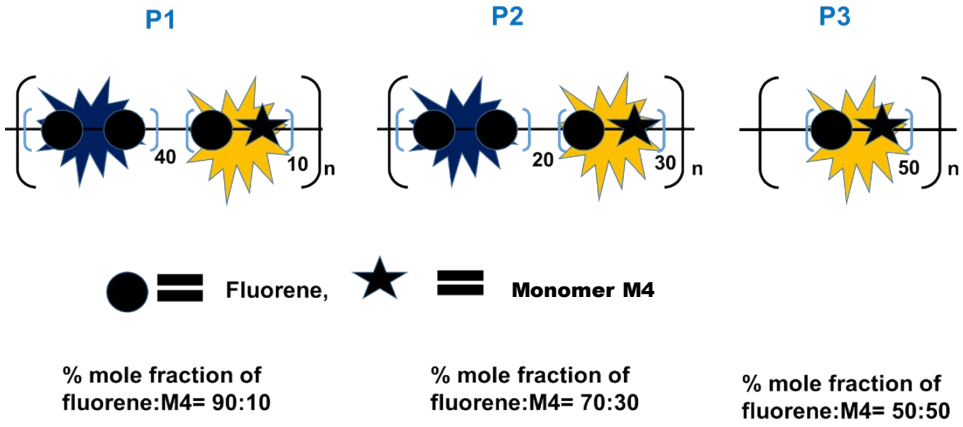


Figure S17: Schematic representation of polymers and the percentage contribution of monomers in the overall emission of polymers.

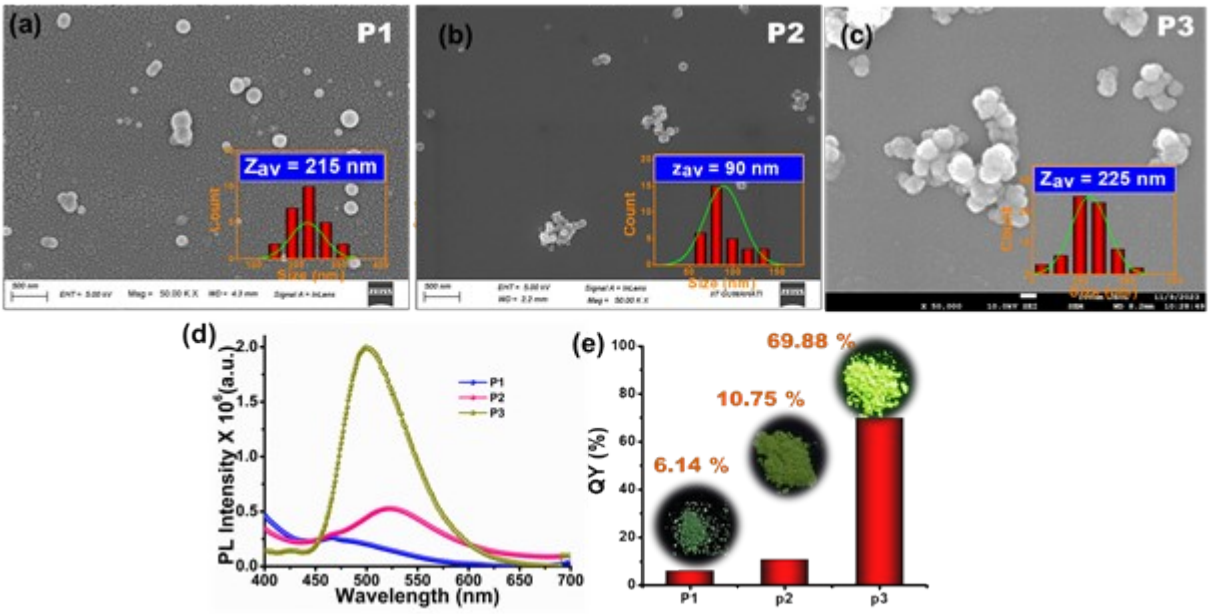


Figure S18: FESEM images of (a) P1, (b) P2, (c) P3 in 99 % f_w; (d) solid state emission spectra and (e) their corresponding Quantum yield and photograph under UV lamp (365 nm).

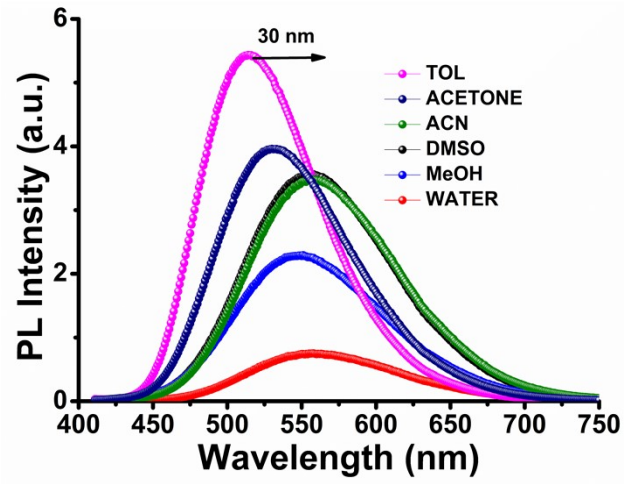


Figure S19: Solvent dependent study of PFAN.

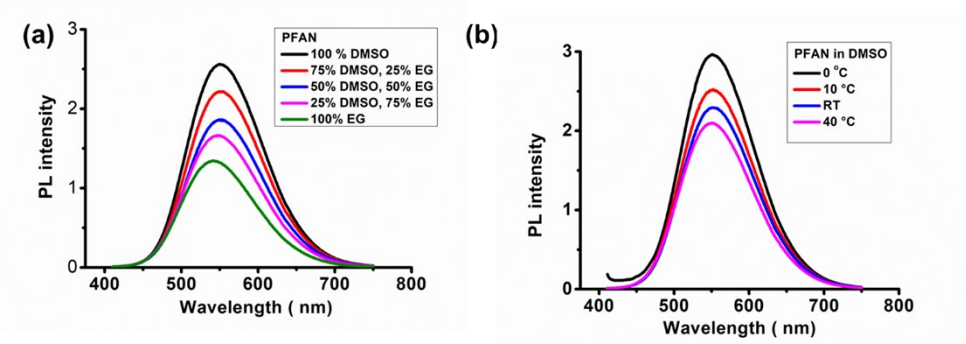


Figure S20: (a) Viscosity and (b) temperature dependent study of PFAN.

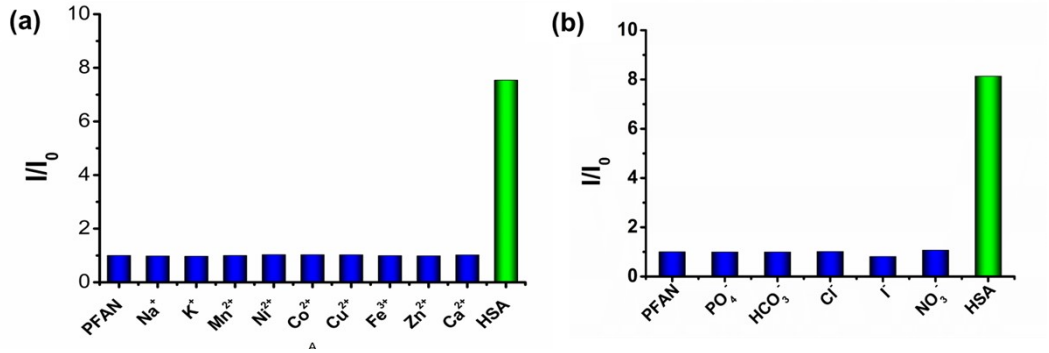


Figure S21: Selectivity study of PFAN with various (a) metal ions and (b) anions

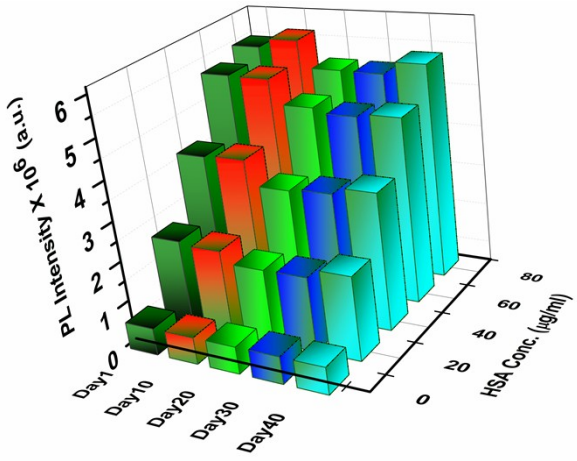


Figure S22: The stability and repeatability of the PFAN polymer under various physiological conditions i.e. HEPES buffer pH 7.4 and temperature 37 ^{o}C.

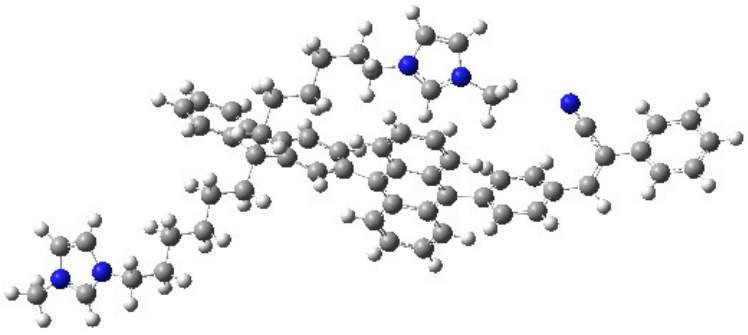


Figure S23: Optimized structure of PFAN monomer using B3LYP function 631G (d, p, charge = +2) basis set of Gaussian Program 16.

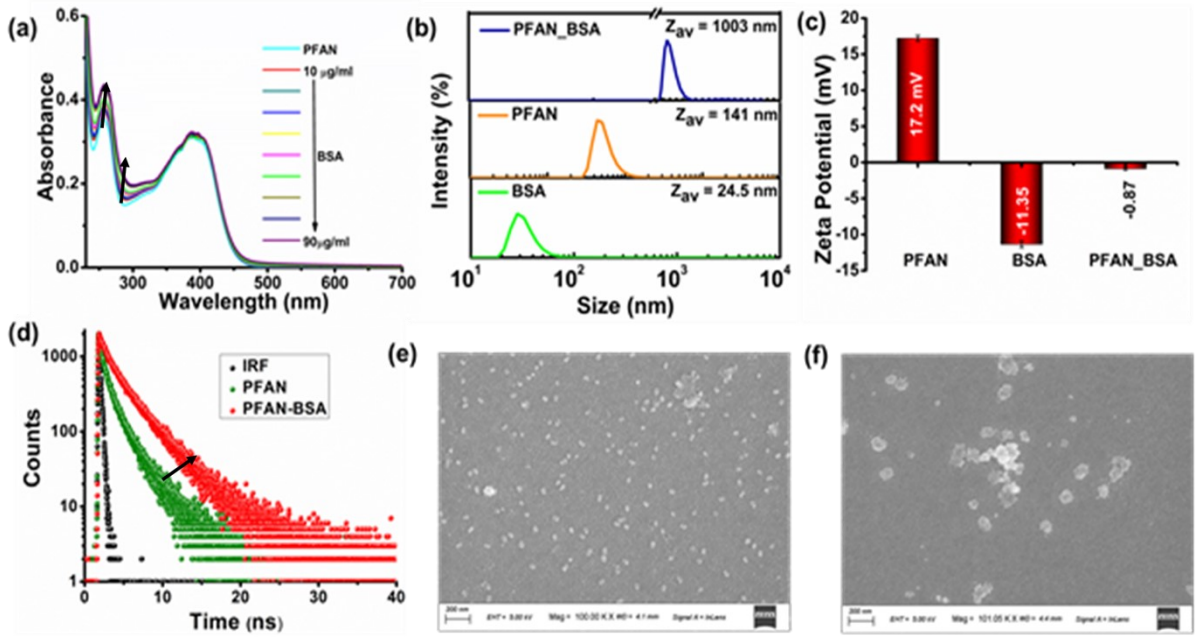


Figure S24: Mechanism of BSA detection: (a) UV-Vis spectra of PFAN by addition of BSA; (b) DLS profile of PFAN, BSA and PFAN-BSA; (c) Zeta potential and (d) Lifetime decay profile of PFAN before and after addition of BSA; FESEM images of PFAN (e) before and (f) after addition of BSA.

Table S2: Lifetime decay data for PFAN in absence and presence of HSA and BSA with an excitation pulse of 405 nm laser

Sample	a_1 (%)	τ_1 (ns)	a_2 (%)	τ_2 (ns)	χ^2	τ_{av} (ns)
PFAN	58.714	0.591	41.286	2.504	1.047	1.38
PFAN-HSA	45.331	1.260	54.669	3.799	1.046	2.65
PFAN-BSA	43.991	1.177	56.009	3.691	1.030	2.58

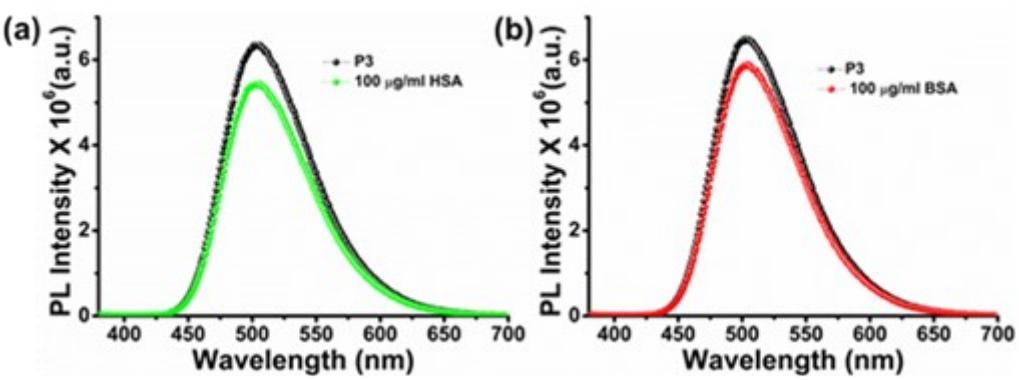


Figure S25: PL spectra of P3 in absence and presence of 100 µg mL^{-1} (a) HSA and (b) BSA.

Table S3: Isoelectric point (IP)[4,5] of various proteins and enzymes at pH 7

Proteins	**IP (net charge at pH 7)**
Cytc	10.5(+)
HRP	9.0(+)
trypsin	10.5(+)
RNase	9.6(+)
pepsin	2.5(−)
Hb	6.8(−)
BSA	4.8(−)
HSA	5.2(−)

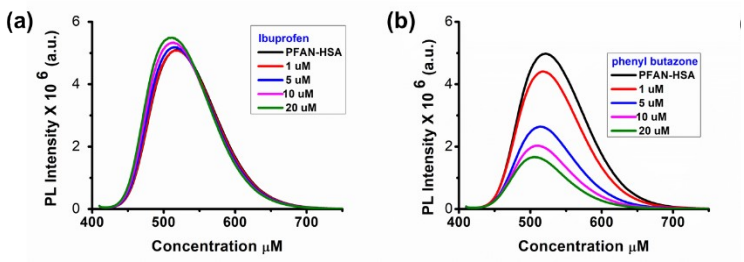


Figure S26: Binding site analysis of PFAN towards HSA using (a) ibuprofen and (b) phenyl butazone

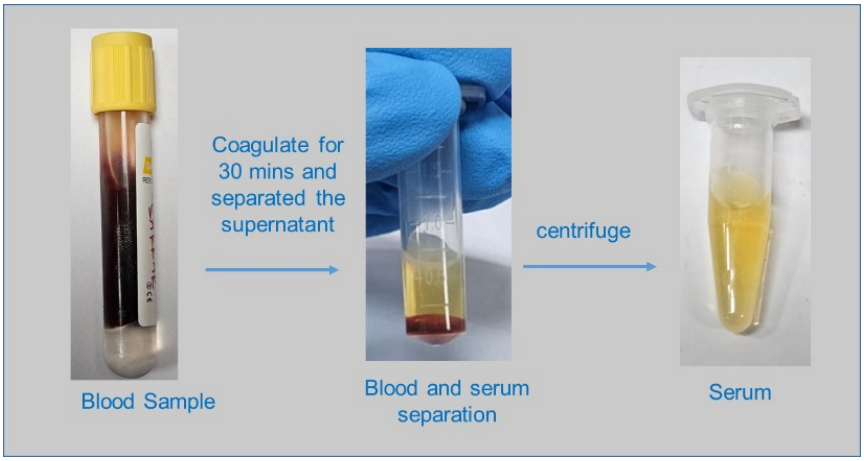


Figure S27: Separation of serum from blood samples.

Table S4: Comparison of our sensing system with the clinical data

Sl no.	Age (years)	HSA detected by our method (g dL^{-1})	HSA detected by clinical method (g dL^{-1})
1	71	3.87±2.13	4.0
2	94	3.93 ± 1.52	3.6
3	72	3.25 ± 2.56	3.4

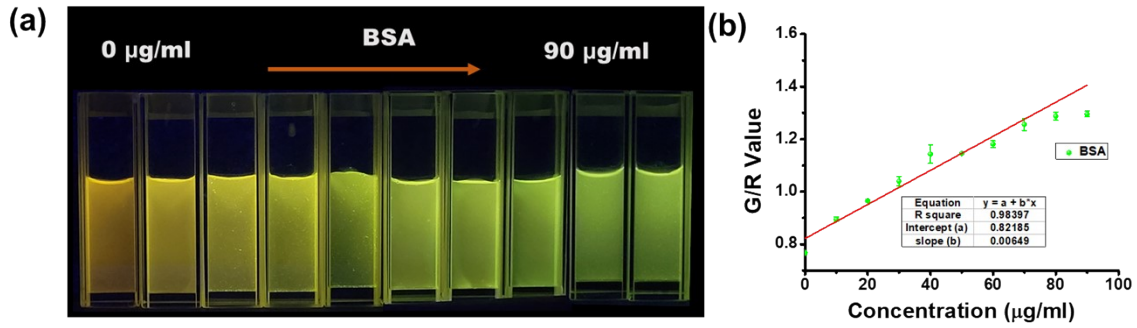


Figure S28: Photograph under UV lamp for (a) BSA (0- 90 µg mL^{-1}) detection by PFAN, (b) corresponding G/R value).

Table S5 Comparison table of HAS and BSA detection with the reported literature

Publications	**Material used**	**Limit of detection (LOD)**	**Selectivity**	**Technique**	**Mechanism**	**PoC Testing**
Present work	AIEE conjugated polyelectrolytes (PFAN)	BSA- 106.7 ng/mL HSA- 50 ng/mL	Selective	Fluorescence	Molecular binding induced AIEE	Yes
Anal.Chem. 2022, **94**,10685−10694	Conjugated polymer surfactant ensembles	BSA 0.50 µg/mL HSA 0.81µg/mL	Selective	Fluorescence	Intermolecular Reverse FRET	Yes
Angew. Chem. Int. Ed. 2020, **59**, 3131	hydroxychalcone derivative	HSA 1.08 µg/ml	Selective	Fluorescence	De-aggregation	Yes
Chem. Commun. 2019, **55**, 13983	Flavanoid derivative	HSA 2.13 µg/mL	Selective	Fluorescnce	AIE	No
Chem. Commun.	Styryl derivative	BSA 3.19	Selective	Fluorecence	TICT Inhibitio	No

2018, **54**, 8383		µg/mL			n	
Chem. Commun. 2018, **54**, 8383	2 Dicyanomethylene-3 cyano-4,5,5-trimethyl-2,5-dihydrofuran derivative	SA 2.5 µg/mL	Selective	Flurescence	AIE	No
ACS Appl. Mater. Interfaces 2017, **9**, 33461	Conjugated polyelectrolyte-Pt(II) complex	Not Reported	Not Selective	Fluorecence	Metal Displacement	No
Anal. Chem. 2017, **89**, 10085	Small organic nanoprobe	HSA 0.14 µM (9.3 mg L^{-1})	Selective	Fluorecence	Disassembly Induced Emission	No
ACS Appl. Mater. Interfaces 2015, **7**, 26094	Pentaaryl substituted pyrrole	BSA 2.18 µg/mL HAS 1.68 µg/mL	Selective	Fluorecence	AIE	No
Macromolec	Conjugated	Not	Not	Fluorescence	Charge	No

ules 2009, **42**, 5933	polyelectrolytes	Reported	Selective		Transfer	
Macromolecules 2008, **41**, 4003	Conjugated polyelectrolyte	Not Reported	Selective	Fluorescence	Interchain FRET	No
Langmuir 2005, **21**, 7985	Conjugated polyelectrolyte	Not Reported	Not Selective	Fluorescence	Aggregation	No
Biochip J. 2017, **11**, 116–120.	Antibody-modified CNT-FET	Not reported	Selective	Electrical conductance	chemical gating effects from HSA bound to mAHSA	No
Top. Catal. 2025, **68**, 677–686	Thin Film of Polydopamine/TiO2 Nanoparticles	10.3 μM (HSA)	Selective	Cyclic voltammetry	Enhanced electron transfer via surface modification and	No

					complexation	
*Anal. Bioanal. Chem.*2024, **414**, 731–741	MIP nanoparticles resulting from solid-phase synthesis	5.3 µg/mL (HSA)	Selective	Quartz crystal microbalance (QCM) sensor	QCMs devices resonance frequency changes when mass deposits on the electrode surface	No
Nat. Commun., 2019, **10**, 3605	synthesized a suite of polycarbodiimide polymers to concomitantly encapsulate carbon nanotubes and interact with specific features	3 mg/L (Albumin)	Selective	Fluorescence	The nanotube emission response behavior, upon interaction with albumin,	No

	of albumin				hypsochromic emission response is caused by the known solvatochromic behavior of nanotube .	
J Clin Pathol. 1987**, 40**, 465-468	immunoturbidimetric method	2.5 mg/L (Albumin)	-	Albumin measured in all samples by automated immunoturbidimetry	-	No
Microchim. Acta. 2020**, 187**, 208	absorbance nanoprobe based on the NCur-VO2+	11nM (Albumin)	Selective	Absorbance	Determination of HSA via aggregati	No

	ensemble was fabricated				on of the NCur by VO2+ and subseque ntly, deaggreg ation of the NCur-VO2+ ensemble in the presence of HSA	
Anal. Chem. 2010**, 82**, 7596–7602	Self-Assembled Protein-Silver Nanoparticle	0.5 µg/mL (BSA)	Not Selective	SERS	Electrost atic interactio n with isolectric point and size is the important parmeter.	NO

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