Materials and Methods
All chemicals and reagents were purchased from commercial sources and were used as received, unless otherwise mentioned. $^1$H NMR spectra were recorded on a 400 MHz Bruker NMR spectrometer using the residual proton resonance of the solvent as the internal standard. Chemical shifts are reported in parts per million (ppm). $^{13}$C NMR spectra were proton decoupled and recorded on a 100 MHz NMR spectrometer using the carbon signal of the deuterated solvent as the internal standard. Molecular weights of the polymers were estimated by gel permeation chromatography (GPC) using PMMA standard with a refractive index detector. Fluorescence spectra were obtained from a JASCO FP-6500 spectrofluorimeter. UV-visible absorption spectra were collected using a Cary 100 spectrophotometer.

Synthesis
Synthetic scheme of probes:

Structure of surfactants and amphiphilic polymer used to form micelles:
Synthesis of *N*-Boc-hexamethylenediamine (3):

Di-tert-butyl dicarbonate (4.0 g, 18.4 mmol) was dissolved in chloroform and added drop-wise to a solution of hexamethylenediamine (10.6 g, 91.6 mmol) in chloroform at 0 °C. The mixture was allowed to warm to room temperature. After stirring for 12 hours, the reaction crude was filtered and washed with chloroform. The filtrates were collected and solvent was evaporated. The residue was re-dissolved in ethyl acetate and washed with water and then brine. The organic solution was dried over anhydrous Na₂SO₄, filtered and concentrated under reduced pressure to afford 1.68 g (42%) *N*-Boc-hexamethylenediamine. ¹H NMR (400MHz, CDCl₃) δ: 4.52 (bs, 1H), 3.10 (q, J = 6.6 Hz, 2H), 2.68 (t, J = 7.0 Hz, 2H), 1.49-1.30 (m, 17H), 1.25 (t, J = 7.2 Hz, 2H); ¹³C NMR (100 MHz, CDCl₃) δ: 156.1, 79.1, 42.2, 40.5, 33.8, 30.2, 28.4, 26.7, 26.6.

Synthesis of compound 4:

To a solution of 1-pyrenebutyric acid (1.0 g, 3.5 mmol) in dry tetrahydrofuran was added N-(3-dimethylaminopropyl)-N’-ethylcarbodiimide hydrochloride (EDC) (0.80 g, 4.2 mmol) at ice bath temperature. The mixture was allowed to stir for 30 min, then *N*-Boc-hexamethylenediamine (0.90 g, 4.2 mmol) was added at the same temperature. The reaction mixture was stirred for 12 hours at room temperature. Tetrahydrofuran was evaporated and the residue was re-dissolved in ethyl acetate and washed with 1M HCl aqueous solution and then brine. The organic layer was dried over anhydrous Na₂SO₄ and evaporated to dryness. The crude product was purified by silica gel column chromatography using mixture of dichloromethane/methanol as eluent to yield 0.35 g (82%) of probe 1. ¹H NMR (400MHz, CDCl₃) δ: 8.35 (m, 2H), 8.12 (m, 2H), 6.91 (bs, 2H), 2.98 (m, 8H), 1.49-1.30 (m, 17H), 1.25 (t, J = 7.2 Hz, 2H); ¹³C NMR (100 MHz, CDCl₃) δ: 170.4, 161.9, 150.6, 131.7, 129.0, 127.7, 26.3.

Synthesis of 4-sulfamoylbenzoic acid-NHS (5):

To a solution of 4-sulfamoylbenzoic acid (1.0 g, 5.0 mmol) in dry tetrahydrofuran was added N-hydroxysuccinimide (0.69 g, 6.0 mmol) and cooled to ice bath temperature. Then EDC (1.15 g, 6.0 mmol) was added at the same temperature. The reaction mixture was stirred for 12 hours at room temperature. Tetrahydrofuran was evaporated and the residue was re-dissolved in ethyl acetate and washed with saturated NaHCO₃ aqueous solution and then brine. The organic solution was dried over anhydrous Na₂SO₄, filtered and concentrated in vacuo to yield 1.0 g (67%) of 4-sulfamoylbenzoic acid-NHS. ¹H NMR (400MHz, Acetone-δ₆) δ: 8.32-8.29 (m, 2H), 8.15-8.12 (m, 2H), 6.91 (bs, 2H), 2.99-2.98 (m, 4H); ¹³C NMR (100 MHz, Acetone-δ₆) δ: 170.4, 161.9, 150.6, 131.7, 129.0, 127.7, 26.3.

Synthesis of probe 1:

To a solution of compound 4 (0.31 g, 0.75 mmol) in dry tetrahydrofuran was added 4-sulfamoylbenzoic acid-NHS (0.25 g, 0.83 mmol) and triethylamine (0.23 g, 2.3 mmol). The reaction mixture was heated for 6 h at 50 °C. Products precipitated out during the course of reaction. The reaction mixture was concentrated in vacuo and the crude product was purified by silica gel column chromatography using mixture of dichloromethane/methanol as eluent to yield 0.35 g (82%) of probe 1. ¹H NMR (400MHz, CDCl₃) δ: 8.35 (m, 2H), 8.12 (m, 2H), 6.91 (bs, 2H), 2.98 (m, 8H), 1.49-1.30 (m, 17H), 1.25 (t, J = 7.2 Hz, 2H); ¹³C NMR (100 MHz, CDCl₃) δ: 170.4, 161.9, 150.6, 131.7, 129.0, 127.7, 26.3.
incubated for 3 hours at room temperature before the emission spectrum of pyrene was recorded ($\lambda_{ex}$ was added 10 $\mu$L (0.098 g, 0.51 mmol) and stirred at RT for 1h. To this mixture was added solution of compound 4 (0.17 g, 0.34 mmol) and triethylamine (0.069 g, 0.68 mmol) in dry dimethylformamide and stirred for 12h. The crude product was precipitated by pouring the reaction mixture into a large volume of diethyl ether. The precipitates were collected and purified by silica gel column chromatography using mixture of dichloromethane/methanol as eluent to yield 0.15 g (72%) of probe 2. $^1$H NMR (400MHz, DMSO-$d_6$) $\delta$: 8.39-7.93 (m, 9H), 7.81 (t, $J = 5.6$ Hz, 1H), 7.72 (t, $J = 5.7$ Hz, 1H), 6.41 (s, 1H), 6.35 (s, 1H), 4.28-4.25 (m, 1H), 4.09-4.07 (m, 1H), 3.31 (t, $J = 8.3$ Hz, 2H), 3.07-2.97 (m, 5H), 2.78 (dd, $J_1 = 5.1$ Hz, $J_2 = 12.4$ Hz, 1H), 2.56 (d, $J = 12.4$ Hz, 1H), 2.22 (t, $J = 7.1$ Hz, 2H), 2.04-1.97 (m, 4H), 1.62-1.23 (m, 14H); $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$: 172.1, 171.9, 162.9, 159.7, 159.5, 154.2, 151.4, 148.9, 148.2, 142.9, 142.3, 123.6, 61.2, 59.3, 55.6, 55.1, 38.5, 38.4, 35.3, 35.2, 32.4, 29.2, 29.2, 28.3, 28.1, 27.7, 26.3, 26.3, 25.5; HRMS (ESI) calculated for C$_{33}$H$_{53}$N$_3$O$_7$S 569.23, found 592.23 (M+Na).

Synthesis of probe 2:

Biotin (0.083 g, 0.34 mmol) was dissolved in dry dimethylformamide heated at 50 °C and then allowed to cool to RT. To the above-obtained biotin solution was added EDC (0.098 g, 0.51 mmol) and triethylamine (0.069 g, 0.68 mmol) in dry dimethylformamide and stirred for 12h. The crude product was precipitated by pouring the reaction mixture into a large volume of diethyl ether. The precipitates were collected and purified by silica gel column chromatography using mixture of dichloromethane/methanol as eluent to yield 0.15 g (72%) of probe 2. $^1$H NMR (400MHz, DMSO-$d_6$) $\delta$: 8.39-7.93 (m, 9H), 7.81 (t, $J = 5.6$ Hz, 1H), 7.72 (t, $J = 5.7$ Hz, 1H), 6.41 (s, 1H), 6.35 (s, 1H), 4.28-4.25 (m, 1H), 4.09-4.07 (m, 1H), 3.31 (t, $J = 8.3$ Hz, 2H), 3.07-2.97 (m, 5H), 2.78 (dd, $J_1 = 5.1$ Hz, $J_2 = 12.4$ Hz, 1H), 2.56 (d, $J = 12.4$ Hz, 1H), 2.22 (t, $J = 7.1$ Hz, 2H), 2.04-1.97 (m, 4H), 1.62-1.23 (m, 14H); $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$: 172.1, 171.9, 162.9, 159.7, 159.5, 154.2, 151.4, 148.9, 148.2, 142.9, 142.3, 123.6, 61.2, 59.3, 55.6, 55.1, 38.5, 38.4, 35.3, 35.2, 32.4, 29.2, 29.2, 28.3, 28.1, 27.7, 26.3, 26.3, 25.5; HRMS (ESI) calculated for C$_{36}$H$_{54}$N$_3$O$_7$S 612.31, found 613.31 (M+H).

Synthesis of PEG-dodecyl random copolymer:

A mixture of cyanomethyl dodecyl thiocarbonate (6.6 mg, 0.021 mmol), lauryl acrylate (200 mg, 0.83 mmol), poly(ethylene glycol) methyl ether acrylate ($M_n = 480$) (600 mg, 1.25 mmol) and AIBN (0.68 mg, 0.0042 mmol) was dissolved in toluene (1.5 mL) and degassed by performing three freeze-pump-thaw cycles. The reaction mixture was sealed and then heated with a pre-heated oil bath at 65 °C for 48 h. The resultant mixture was precipitated twice in diethyl ether to remove unreacted monomers and initiators. $^1$H NMR (400MHz, CDCl$_3$) $\delta$: 4.27-4.07, 4.07-3.85, 3.62-3.55, 3.37, 2.45-2.01, 1.94-1.78, 1.70-1.52, 1.34-1.18; GPC (THF) $M_n$: 42800 Da. PDI: 1.3.

Encapsulation of probe/quencher in micellar solutions:

To a 970 $\mu$L solution of surfactants or polymer in Tris buffer (25 mM, pH = 7.4) was slowly added 10 $\mu$L (100 $\mu$M) solution of probe in dimethyl sulfoxide (DMSO), followed by 10 $\mu$L (500 $\mu$M) solution of benzophenone (BP) in DMSO under vigorously stirring. The mixture was allowed to stir for 30 min at room temperature and then filtered through a filter with a pore size of 0.22 $\mu$m to remove excess BP that was not encapsulated. Excess BP was needed to minimize the background signal.

Probe dissociation with different concentrations of the target protein and other proteins:

To a 990 $\mu$L solution of probe/BP or pyrene/BP encapsulated micelles in Tris buffer (25 mM, pH = 7.4) was added 10 $\mu$L (0 – 500 $\mu$M) solution of proteins in Tris buffer (25 mM, pH = 7.4). The mixture was incubated for 3 hours at room temperature before the emission spectrum of pyrene was recorded ($\lambda_{ex}$ =
345 nm, the excitation and emission band widths were set to 3 and 3 nm respectively and the scanning speed was 500 nm/min).

Figure S1. Absorption spectra of probes (1 µM) in Tris buffer solutions with or without Brij 35 (1 mM): (a) probe 1; (b) probe 2.

Figure S2. Fluorescence spectra of probes (1 µM) and Brij 35 (1 mM) in Tris buffer with or without BP (5 mM): (a) probe 1; (b) probe 2. ($\lambda_{ex} = 345$ nm, the excitation and emission band width were 1 and 3 nm respectively and the scanning speed was 500 nm/min).

Figure S3. Fluorescence spectra of pyrene (1 µM), BP (5 mM), and Brij 35 (1 mM) in Tris buffer (25 mM, pH 7.4) treated with HCA (5 µM) or avidin (1 µM) ($\lambda_{ex} = 335$ nm for pyrene).
**Figure S4.** Fluorescence spectra of probe 1 (1 µM), BP (5 mM), and Brij 35 (1 mM) treated with or without HCA (1 µM): (a) in Tris buffer solution; (b) in 50% fetal bovine serum in Tris buffer solution.

**Figure S5.** Fluorescence spectra of probe 2 (1 µM), BP (5 mM), and Brij 35 (1 mM) in Tris buffer treated with different proteins (1 µM).
Figure S6. Fluorescence spectra of probe 1 (1 µM) and BP (5 mM) without or with 5 µM HCA in different concentrations of surfactants or amphiphilic polymer.