Cationic- and Anionic Conjugated Polyelectrolytes; Aggregation-Mediated Fluorescence Energy Transfer to Dye-Labeled DNA

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All chemicals were purchased from Aldrich Co., and were used without further purification. \(^1\)H- and \(^1^3\)C-NMR spectra were collected on a Varian Unity 400 MHz (or 200 MHz) spectrometer. The UV/vis absorption spectra were recorded on a Shimadzu UV-2401 PC diode array spectrometer. Photoluminescence spectra were obtained on a PTI Quantum Master fluorometer equipped with a Xenon lamp excitation source. Fluorescence quantum yields were measured relative to fluorescein at pH = 11 in water.\(^1,2\) The oligonucleotide (ssDNA-TR: 5′-TR-ATC TTG ACT ATG TGG GTG CT, 20-base) was purchased from Genscript Corp. and the DNA concentration was determined by measuring the absorbance at 260 nm in a 200 µL quartz cuvette.

The anionic polymer, a-PFP, was synthesized according to the procedures reported by Huang et al (Scheme S1).\(^3\) The monomer, 2,7-dibromo-9,9-bis(4-sulfonatobutyl)-fluorene disodium salt, was prepared by alkylation reaction of 2,7-dibromofluorene with 4-butane sultone using NaOH in DMSO at 60 °C. The pure compound was collected by recrystallization from acetone/H\(_2\)O twice in 65.2% yield. The final polymer was prepared by Suzuki polymerization of the monomer with 1,4-phenylenebisboronic acid using Pd(OAc)\(_2\) and sodium carbonate in DMF/water at 80 °C for 2 days. The reaction mixture was precipitated into acetone, re-dissolved in deionized water, and the resulting mixture was purified by dialysis (Mw of 10,000 g/mol cut-off) for 3 days.

**Scheme S1**

The cationic polymer, c-PFB\(_{15}\), was obtained by a series of reactions shown in Scheme S2. The first step consists of deprotonation of 2-bromofluorene with NaOH in a two-phase mixture of water and 1,6-dibromohexane. Another monomer, 2,7-bis[9,9′-bis(6′-bromohexyl)fluorenyl]-4,4,5,5-tetramethyl-
[1.3.2]dioxaborolane, was prepared via lithiation using n-butyl lithium, followed by quenching with 2-isopropoxy-4,4,5,5-tetramethyl-1,3,2-dioxaborolane. Monomer 1 (227 mg, 0.35 mmol), 2 (372 mg, 0.5 mmol), 4,7-dibromo-2,1,3-benzothiadiazole (44.1 mg, 0.15 mmol), Pd(PPh₃)₄ (10 mg), and potassium carbonate (1,660 mg, 12 mmol) were reacted in a solvent mixture of water (6 ml) and toluene (12 ml) at 80 °C for 20 hrs. The resulting polymer was filtered and washed with methanol and acetone, and then dried under vacuum for 24 hrs to afford the neutral precursor. The water-soluble polymer, c-PFB₁₅, was obtained by treatment of the neutral precursor with condensed trimethylamine (4 ml) in a THF/water mixture for 24 hrs. After addition of trimethylamine, a precipitate gradually forms as the reaction proceeds, which is consistent with ionization of the pendant groups. To ensure reaction completion, it is recommended to redissolve the precipitate by adding water (or methanol) and to allow a further reaction with another excess of trimethylamine (4 ml). The final c-PFB₁₅ was dissolved in deionized water, and purified by dialysis (Mw of 10,000 g/mol cut-off) for 3 days. Gel permeation chromatography (GPC) measurements were performed in THF for the neutral precursor polymer ($M_n$=22,500 g/mol, PDI=1.8), relative to a polystyrene standard.

Scheme S2
Figure S1. Fluorescence spectra of the solution containing fluorescein-labeled DNA ($1.4 \times 10^{-8}$ M, 20-bases oligonucleotide) with adding a-PFP ([a-PFP] = $1 \times 10^{-7}$ M ~ $3 \times 10^{-7}$ M). All emission scans were collected by exciting at 380 nm.

Figure S2. Time-resolved PL decay measurements of c-PFP and a-PFP and 1:1 mixture of c-PFP and a-PFP by exciting at $\lambda_{ex} = 380$ nm.
1) a-PFP ($\lambda_{\text{det}} = 405$ nm): PL decay can be fit with a single exponential with time constant of 0.71 ns.

2) c-PFP ($\lambda_{\text{det}} = 410$ nm): PL decay can be fit with a single exponential with time constant of 0.43 ns.

3) 1:1 mixture of a-PFP and c-PFP ($\lambda_{\text{det}} = 410$ nm): PL decay was approximately fit with a double exponential with time constants of 0.36 ns (26%) and the value less than 40 ps (81%). We measured a serious quenching due to aggregation.

**Figure S3.** Time-resolved PL decay measurements of c-PFB$_{15}$ and a-PFP and 1:1 mixture of c-PFB$_{15}$ and a-PFP by exciting at $\lambda_{\text{ex}} = 380$ nm.

1) a-PFP ($\lambda_{\text{det}} = 405$ nm): PL decay can be fit with a single exponential with time constant of 0.71 ns.

2) c-PFP ($\lambda_{\text{det}} = 410$ nm): PL decay can be fit with a single exponential with time constant of 0.6 ns.

3) 1:1 mixture of a-PFP and c-PFP ($\lambda_{\text{det}} = 579$ nm): PL decay was approximately fit with a double exponential with time constants of 2.36 ns (53%) and 0.54 ns (47%). The average lifetime is 1.40 ns.
Figure S4. Fluorescence spectra of ssDNA-TR ([ssDNA-TR] = 1× 10^{-8} M and [– charge] in ssDNA-TR = 2 × 10^{-7} M) with adding c-PFB_{15} ([c-PFB_{15}] = 0 ~ 2.4 × 10^{-7} M) in water. All emission scans were collected by exciting at 380 nm.

Figure S5. Fluorescence spectra of the complex of c-PFB_{15} and ssDNA-TR with adding a-PFP ([c-PFB_{15}] = 3.0× 10^{-7} M, [ssDNA-TR] = 1× 10^{-8} M, and [a-PFP] = 0 M to 3 × 10^{-7} M). All emission scans were collected by exciting at 380 nm.
Figure S6. Fluorescence spectra of (a) ssDNA-TR with adding c-PFB$_{15}$, (b) the complex of c-PFB$_{15}$/ssDNA-TR with adding a-PFP, and (c) ssDNA-TR with adding the complex of c-PFB$_{15}$/a-PFP in water ([ssDNA-TR] = 1 × 10$^{-8}$ M, [c-PFB$_{15}$] = 8 × 10$^{-8}$ M, [a-PFP] = 4 × 10$^{-8}$ M). All emission scans were collected by exciting at 380 nm.

References

