Supporting Information

pH-Switchable Unimolecular Fluorescent Polymeric Micelles: From Volume Phase Transition to Optical Response

Shusen You,^a Qing Cai,^a Klaus Müllen,^b Wantai Yang,^a Meizhen Yin^{*a}

^a State Key Laboratory of Chemical Resource Engineering, Key Laboratory of Carbon Fiber and Functional Polymers, Beijing University of Chemical Technology, 100029 Beijing, China, Email: <u>yinmz@mail.buct.edu.cn</u>.

^b Max-Planck-Institute for Polymer Research, Ackermannweg 10, 55128 Mainz, Germany.

Materials

2-[(*tert*-Butoxycarbonyl) amino] ethyl methacrylate (Boc-AEMA) was prepared according to previous reports.¹ *tert*-Butyl acrylate (*t*BA, Alfa Aesar, 99%) and 2-butanone (Sinopharm, 99%) were purified by vacuum distillation before use. 4,4'-di-*tert*-Butyl-2,2'-bipyridine (DTB-bipy, Aldrich, 98%) and copper(I) bromide (CuBr, Alfa Aesar, 99.999%) were used without further purification. The buffer solutions were prepared by 0.1 M Tris and 0.1 M HCl. All the salts (AR grade) were purchased from Sinopharm and used without further purification. PDI-labeled macroinitiator bearing eight 2-bromo-2-methylpropionic ester groups (1) was synthesized as described.² Other reagents were purchased from Alfa Aesar without further purification.

Instruments

Nuclear magnetic resonance (NMR) spectra were recorded on Bruker 400 (400 MHz ¹H) spectrometer using CDCl₃, MeOD and D₂O as solvent at room temperature. The gel permeation chromatography (GPC) setup comprised two Polymer Laboratories PL gel 5 μ m Mixed-C columns maintained at 60 °C in series with a Varian 390 LC refractive index detector. The flow rate was 1.0 mL/min, and the mobile phase contained 10 mmol LiBr. A total of 10 near-monodisperse PMMA standards (Mp = 600 to 618000 g/mol) were used for calibration. The pH measurements were carried out using a pH meter (Mettler Toledo S40K), which was calibrated with pH 4 and pH 7 buffers before use. UV-visible spectra were obtained on a spectrometer (Cintra 20, GBC, Australia). The corrected fluorescence spectroscopic studies were performed on a fluorescence spectrophotometer (Horiba Jobin Yvon FluoroMax-4 NIR, NJ, USA) at room temperature.

Small-angle X-ray scattering (SAXS)

SAXS experiments were carried out at a high-flux SAXS instrument (SAXSess, Anton Paar) equipped with a Kratky block-collimation system and an imaging plate (IP) as the detector. According to previous paper, two formulas were performed for calculation:³

$$I(q) = 4\pi \int_0^\infty p(r) \frac{\sin qr}{qr} dr (1),$$

$$p(r) = r^2 \Delta \rho^2(r) (2).$$

Where p(r) is the pair distance distribution function (PDDF) of the particle, and $\Delta \rho^2(r)$ is the convolution square (spatial correlation function) of $\Delta \rho$ (r) averaged over all orientations in space. The functional form of I(q) or p(r) can be used to determine the shape and the internal structure of the scattering object.

Dynamic light scattering (DLS)

DLS experiments was performed with an equipment composed of an ALV SP-86 goniometer, a Spectra Physics 2011-s Kr ion laser (637.2 nm wavelength, 500 mW output power) and an ALV-5000 correlator. Aqueous polymer solutions of various pH values were prepared and passed through a 0.22 μ m filter prior to size determination. Size measurements were performed at a 90° angle at 25 °C. In the experiments, pH was adjusted by using small amounts of 0.1 M NaOH and 0.1 M HCl.

Potentiometric Titration

Aqueous solutions of the polymers were prepared (1 mg/mL) and titrated with 0.001 M NaOH or 0.001M HCl. The pH values were plotted as a function of volume of added NaOH or HCl. There is a buffer zone in every plot. The pKa values were determined in the zone and calculated from the formula (3):⁴

$$pKa = pH + \log \frac{1 - \alpha}{\alpha} \quad (3)$$

Where $\alpha = V_a/V_b$, and V_a is the accumulated volume of NaOH or HCl added to the solution, V_b is the total volume of added NaOH or HCl in the buffer zone. The results are shown in **Table S1**.

Polymer	r.u. ^a	M _{n,NMR} ^b (g/mol)	M _{n,GPC} (g/mol)	M_w/M_n	рКа
PDI-PAA	55	35600	24000	1.13	6.34 ± 0.02
PDI-PAEMA	55	60680	41300	1.10	8.05 ± 0.03

Table S1 Summary of polymer data

^{*a*} repeat units on every arm

^b calculated from ¹H NMR spectra

Synthesis and characterizations

Scheme S1 Synthesis of PDI-PAA and PDI-PAEMA.

Synthesis of PDI-PtBA

A reaction tube was charged with the initiator **1** (50 mg, 0.013 mmol, 1 equiv.), *t*BA (2.65 g, 20.8 mmol, 8×200 equiv.), CuBr (14.9 mg, 0.104 mmol, 8×1 equiv.), and dry 2-butanone (2.0 mL). The tube was degassed by three freeze-pump-thaw cycles, and then DTB-bipy (55.7 mg, 0.208 mmol, 8×2 equiv.) was added to the reaction tube. The solution was stirred for 10 minutes to ensure the catalyst completely form complex, the polymerization was carried out under N₂ atmosphere at 90 °C for 10 h. After that, the solution was poured into excess methanol/water (v/v = 1:1) to precipitate the polymer, and the precipitate was dissolved in dichloromethane and purified using an alumina column to eliminate the used copper salt, then precipitated again in methanol. After drying in a vacuum oven overnight at room temperature, PDI-PtBA was obtained as a red powder in 27.5% yield.

¹H NMR (CDCl₃, 400MHz) δ (ppm): 1.46 (9H, -COOC(C*H*₃)₃), 1.85 (2H, -C*H*₂CH), 2.24 (1H, -CH₂C*H*), 6.85-8.10 (aromatic H).

Hydrolysis of PDI-PtBA

The polymer PDI-P*t*BA was placed in a Schlenk flask equipped with a magnetic stirring bar. Then, the flask was evacuated and backfilled with nitrogen three times. Dichloromethane (20 mL) was added to dissolve the polymer. Then, trifluoroacetic acid (TFA, 10 mL) was added to the solution, and the mixture was stirred at room temperature for 24 h. The solvent was removed from the resulting heterogeneous mixture, and the residual solid was washed with CH_2Cl_2 three times followed by drying under vacuum at room temperature for 10 h to give PDI-PAA as a red solid. ¹H NMR (MeOD, 400MHz) δ (ppm): 1.96 (2H, -CH₂CH), 2.47 (1H, -CH₂CH), 6.78-8.10 (aromatic H).

Synthesis of PDI-P(Boc-AEMA)

A Schlenk tube was charged with **1** (40 mg, 0.01 mmol, 1 equiv.), Boc-AEMA (1.82 g, 8 mmol, 4×200 equiv.), CuBr (11.4 mg, 0.08 mmol, 8×1 equiv.), and dry 2-butanone (2.0 mL). The tube was degassed by three freeze-pump-thaw cycles, and then DTB-bipy (42.9 mg, 0.16 mmol, 8×2 equiv.) was added. After stirred for 10 minutes to ensure the catalyst completely form complex, the polymerization was carried out under N₂ atmosphere at 60 °C for 30 minutes. After that, the solution was cooled down to room temperature, diluted with 10 mL CH₂Cl₂ and passed through an alumina column to eliminate the used copper salts. After concentrated by rotovap, the solution was poured into excess methanol/water (v/v= 1:1) to precipitate the polymer, and the crude product was precipitated in methanol/water (v/v= 1:1) for another three times. After drying in a vacuum oven over night at room temperature, the product was obtained as a red solid in a yield of 27.5%.

¹H NMR (CDCl₃, 400MHz) δ (ppm): 0.80-0.99 (3H, -C(CH₃)CH₂), 1.46 (9H, -NHCOOC(CH₃)₃), 1.84 (2H, -C(CH₃)CH₂), 3.28 (2H, -OCH₂CH₂NH), 3.91 (2H, -OCH₂CH₂NH), 5.52 (1H, -OCH₂CH₂NH), 6.78-8.10 (aromatic H).

Hydrolysis of PDI-P(Boc-AEMA)

The polymer PDI-P(Boc-AEMA) was placed in a Schlenk flask equipped with a magnetic stirring bar. Then, the flask was evacuated and backfilled with nitrogen three times. CH_2Cl_2 (20 mL) was added to dissolve the polymer. Then, phenol (3 M in CH_2Cl_2 , 6 mL) and chlorotrimethyl silane (1M in CH_2Cl_2 , 6 mL) were added to the solution, and the mixture was stirred at room temperature for 2 h. The solvent was removed from the resulting heterogeneous mixture, and the residual solid was washed with diethyl ether three times followed by drying under vacuum at room temperature for 10 h to give PDI-PAEMA as a red solid.

¹H NMR (D₂O, 400MHz) δ (ppm): 0.65-0.82 (3H, -C(CH₃)CH₂), 1.75 (2H, -C(CH₃)CH₂), 3.07 (2H, -OCH₂CH₂NH₂), 3.97 (2H, -OCH₂CH₂NH₂), 6.52-8.10 (aromatic H).



Fig. S1 GPC curves of PDI-PAA and PDI-PAEMA.



Fig.S2 Scattering curves of PDI-PAA (1 mg/mL, red) and PDI-PAEMA (1 mg/mL, black).



Fig. S3 Titration curves of A) PDI-PAA by 0.001 M NaOH; B) PDI-PAEMA by 0.001 M HCl.



Fig. S4 Concentration-dependent absorbance spectra of (A) PDI-PAA and (B) PDI-PAEMA in aqueous solution. Inset: dependence of absorbance intensity (at λ_{max} of 583 nm) as a function of polymer concentration.



Fig. S5 UV-vis spectra of PDI-PAA (0.062 mg/mL) in Tris-HCl buffer (pH range from 3 to 12).



Fig. S6 Fluorescence spectra of PDI-P*t*BA (0.01 mg/mL) in THF with extra (A) water or (B) THF content from 0% to 10% (λ_{ex} = 540 nm) in 1% increments. Inset: The ratio of fluorescence emission intensity after-to-before adding water (I_x/I_0) as a function of water content.

Table	S2	Zeta	potentials	of	PDI-PAA	and	PDI-PAEMA	at	different	pН	values
adjuste	ed by	y NaO	H (0.1 mol	/L)	and HCl (().1 m	ol/L).				

Zeta potential (mV)	pH= 3	pH= 7	pH= 10
PDI-PAA	-0.49±0.1	-14.5±0.5	-17.3±1.2
PDI-PAEMA	16.9±0.9	12.4±0.6	6.8±0.5

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