Supporting Information

Electrochemical Redox Responsive Polymeric Micelles Formed from Amphiphilic Supramolecular Brushes

Anchao Feng, Qiang Yan*, Huijuan Zhang, Liao Peng and Jinying Yuan*

Key Lab of Organic Optoelectronics & Molecular Engineering of Ministry of Education,
Department of Chemistry, Tsinghua University, Beijing 100084, China.

AUTHOR EMAIL ADDRESS: yanq04@aliyun.com (Q. Yan),

yuanjy@mail.tsinghua.edu.cn (J.Yuan)

1. Materials.

Methoxy poly(ethylene glycol) (mPEG-OH, Mn_{GPC} = 2.00 kg·mol^{-1}) was dried by azeotropic distillation in the presence of toluene. ε-Caprolactone (ε-CL, Acros, 98%) was purified with CaH_2 by vacuum distillation in a nitrogen atmosphere. Glycidyl methacrylate (GMA, Tokyo Chemical Industry Co. Ltd, 95%) was distilled in vacuum before use.Stannous octoate ($Sn(Oct)_2$, Aldrich, 99%) was distilled under reduced pressure before use. β-Cyclodextrin (β-CD) was purchased from Kermel(China) and purified by recrystallization from water before use.2-Dodecylsulfanylthiocarbonylsulfanyl-2-methyl propionic acid (DMP) was synthesized according to reference.[1]Ferrocenemethanol(FcM, Acros, 99%),tosyl chloride (TsCl, Acros, 99%),dicyclohexylcarbodiimide (DCC, Alfa Aesar, 99%), 2, 2-azobis (isobutyronitrile)(AIBN, Alfa Aesar, 98%), 4-dimethaminopyridine (DMAP, Fluka, 99%) were used as received. Methanol, N, N-dimethylformamide(DMF), and dioxane were received from BeijingChemical Reagent Co. Ltd (China) and purified before use. Allother reagents of analytical grade were commercially availableand used without further purification. Distilled water wasutilized throughout the studies.

2. Characterization.

*Fourier Transform Infrared Spectroscopy (FT-IR).*The absorption spectra of all products were recordedon an AVATAR 360 ESP FT-IR spectrometer and the results were collected at 30 scans with a spectralresolution of 1 cm^{-1}.

Nuclear Magnetic Resonance Spectroscopy (NMR).^{1}H NMR spectra for the polymer structural analysiswere obtained from a JEOL JNM-ECA300 (300MHz) and 2D NMR (NOESY) spectra were recorded atJEOL JNM-ECA600 (600MHz) spectrometer.

*Electrospray Ionization Mass Spectrometry (ESI-MS).*The data were collected using aMicroTOF-QII Bruker.

Gel Permeation Chromatography (GPC). GPC analyses of polymers were performed using DMF as the eluent.

The GPC system was a Shimadzu LC-20AD pump system, a MZ-Gel SDplus 10.0 μm guard column(50 × 8.0 mm, 10^2 Å) followed by a MZ -Gel SDplus 5.0 μm bead-size column (50-10^6 Å, linear) and a Shimadzu RID-10A refractive index detector. The system was calibrated with narrow molecular weight distribution polystyrene standards ranging from 200 to 10^6 g·mol^{-1}.

*Transmission electron microscopy (TEM).*The visualized images of the assemblies were obtained from aJEM-2010 microscope with an accelerating voltage of 120 kV and H-7650B microscope with an accelerating voltage of 80 kV.The samples wereprepared by drop-coating the aqueous solution ona carbon-coated copper grid and staining with 0.1%phosphotungstic acid hydrate.

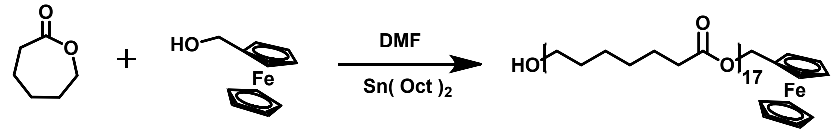
*Dynamic Light Scattering (DLS).*The average radius and size distribution of the aggregates was analyzedby a Malvern 3000HS Zetasizer using a monochromatic coherentHe–Ne laser (633 nm) as the light source and a detector thatdetected the scattered light at an angle of 90º.

*UV-vis Spectroscopy (UV-vis).*The UV-visible spectraof copolymer solutions were acquired on a HITACHI U-3010 spectrophotometer.In addition, the optical transmittance was acquired at a wavelength of 550 nm using acuvette.

*Fluorescence Spectroscopy (FS).*Fluorescence was measured with a Hitachi F-7000spectrofluorometer.

3. Synthetic Procedures.

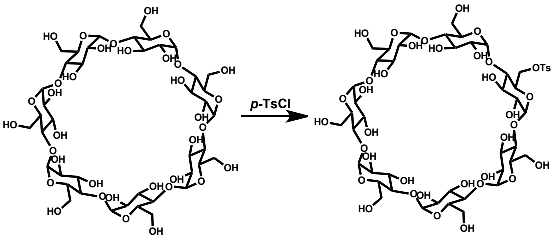
3.1. Synthesis of PCL$_{17}$-Fc homopolymer via ring-opening polymerization (ROP).



A typicalpolymerization procedure was shown as follows: ε-CL (1 mL, 8.8 mmol), FcM (86.4 mg, 0.4 mmol), Sn (Oct)$_2$ (60 μL, 0.06 mmol/L solution) were firstly dissolved in DMF. Then after exhausting-refilling processes repeated three times, the tube was immersed into an oil bath at 120°C under nitrogen atmosphere with vigorous stirring. The reaction proceeded for 24h and then was stopped by diluting with DMF followed by precipitation in methanol. The dissolving-precipitation was repeated three times and the obtained light yellow product was dried in vacuum (yield: ~91%). The DP of PCL block was determined to be 17 by ^{1}H NMR analysis in $CDCl_3$.

Mn_{GPC}= 2600 g/mol, Mw/Mn = 1.26. FT-IR (KBr, cm^{-1}): 2949($\nu_{C\text{-}H}$), 1739 ($\nu_{C=O}$), 1623 ($\nu_{C=C}$ in ferrocene), 1157 ($\nu_{C\text{-}O}$);^{1}H NMR (CDCl3, δ, ppm): 4.89 (s, 2H, =C***H***C(COOH)C***H***=), 4.26 (s, 2H, -C***H***=C***H***-), 4.17 (d, 5H, another cyclopentyl), 4.13–3.98 (m, 34H, $\text{-OC}\boldsymbol{H_2}\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{COO-}$), 2.38–2.24 (m, 35H, $\text{-OCH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{C}\boldsymbol{H_2}\text{COO-}$), 1.78–1.22 (m, 124H, $\text{-OCH}_2\text{C}\boldsymbol{H_2}\text{C}\boldsymbol{H_2}\text{C}\boldsymbol{H_2}\text{CH}_2\text{COO-}$).

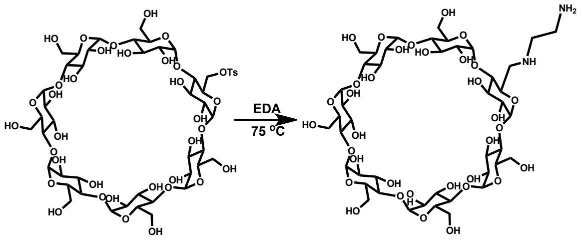
3.2. Synthesisof mono-6-deoxy-6-(p-tolysulfonyl)-β-CD (CD-OTS).



The mono-6-deoxy-6-(p-tolysulfonyl)-β-CD was prepared according to reference.[S2, S3] 20 g of β-CD (17.6mmol) was suspended in 167 mL of water, and 2.19 g of NaOH(54.7 mmol) in 7 mL of water was added dropwise. Thesuspension turned to be homogeneous. The solution wasimmersed into an ice-water bath, and then 5.04 g of TsCl (26.4mmol) in 10 mL of acetonitrile was dripped slowly, causing theformation of white precipitates. After further stirring for 2 h at25°C, the suspension was refrigerated overnight at 4°C. Theresulting precipitate was recovered by suction filtration andrecrystallized from hot water three times. The product was driedunder vacuum at 50°C.

^{1}H NMR (DMSO, δ, ppm): 7.72 (d, 2H, aromatic protons), 7.40(d, 2H, aromatic protons), 5.90–5.47(m, 14H, OH-2,3), 4.80(d,4H, H-1), 4.71 (d, 3H, H-1), 4.49–4.17(d, 6H, OH-6), 3.74–3.48(m, 28H, H-3, 5, 6), 2.35 (s, 3H, –CH_3).

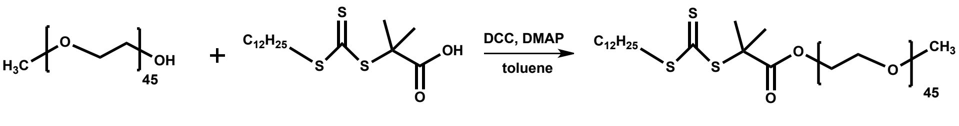
3.3. Synthesis of mono-6-deoxy-6-ethylenediamine-β-CD (CD-EDA).



The mono-6-deoxy-6-ethylenediamine-β-CD (EDA-β-CD) was synthesized according to the previous report.[S4]5.0 g of mono-6-OTs-β-CD was reacted with an excess amount of ethylenediamine (EDA) at 80°C for 4 h. After the reaction was completed, the solution was cooled down and most of the unreacted EDA was removed by rotary evaporation. EDA-β-CD was dissolved in water-methanol mixture and precipitated in acetone three times. The product was dried at 50°C for 24 h in a vacuum oven.

^{1}H NMR (D_2O, δ, ppm): 4.92 (d, 7H, H-1), 3.83-3.67(m, 28H, H-3, 5, 6), 3.51-3.31(m, 14H, H-2, 4), 2.89 (2H, –C$\boldsymbol{H_2}$NH-β-CD), 2.70 (2H, NH_2C$\boldsymbol{H_2}$-CH_2). ESI-MS m/z: 1178.6 [M + H$^+$].

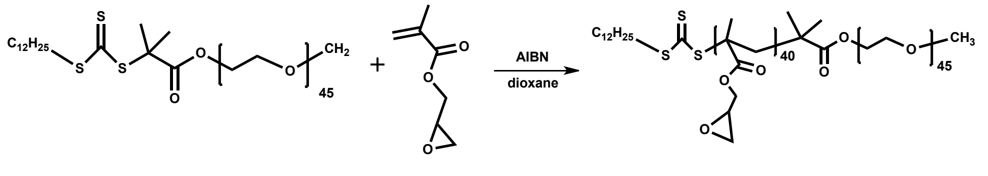
3.4. Synthesis ofmethoxy-DMP-poly(ethylene oxide) (PEG-RAFT)macro-initiator.



A representative reaction of PEG-RAFT was carried out as follow: mPEG-OH was added into a 100 mL two-necked flask equipped with a stirrer. The mPEG-OH was dried by three azeotropic distillations of toluene and finally dissolved in 50 mL dry toluene. DMP (1.1 equiv., 2 g), DCC (1.1 equiv., 1.13 g) and DMAP (1.1 equiv., 0.67 g) were then added, and the flask was heated in an oil bath at 70°C overnight. PEG-RAFT was collected by precipitation in ether at 0°C and then dried at 40°C in vacuum for 24 h(yield: ~87%).

^{1}H NMR (CDCl$_3$, δ, ppm): 3.65 (s, 180H, $-OC\boldsymbol{H_2}C\boldsymbol{H}_2O-$), 3.57-3.53 (m, 2H, $CH_3(CH_2)_8CH_2CH_2C\boldsymbol{H_2}S-$), 3.27 (t, 2H,$CH_3(CH_2)_8CH_2C\boldsymbol{H_2}CH_2S-$), 1.74-1.56 (m, 12H, $-C(C\boldsymbol{H_3})_2COO-$, $CH_3(CH_2)_8C\boldsymbol{H_2}CH_2CH_2S-$), 1.42-1.21 (m, 16H, $CH_3(C\boldsymbol{H_2})_8CH_2CH_2CH_2S-$), 0.88 (t, 3H, $C\boldsymbol{H_3}(CH_2)_8CH_2CH_2CH_2S-$).

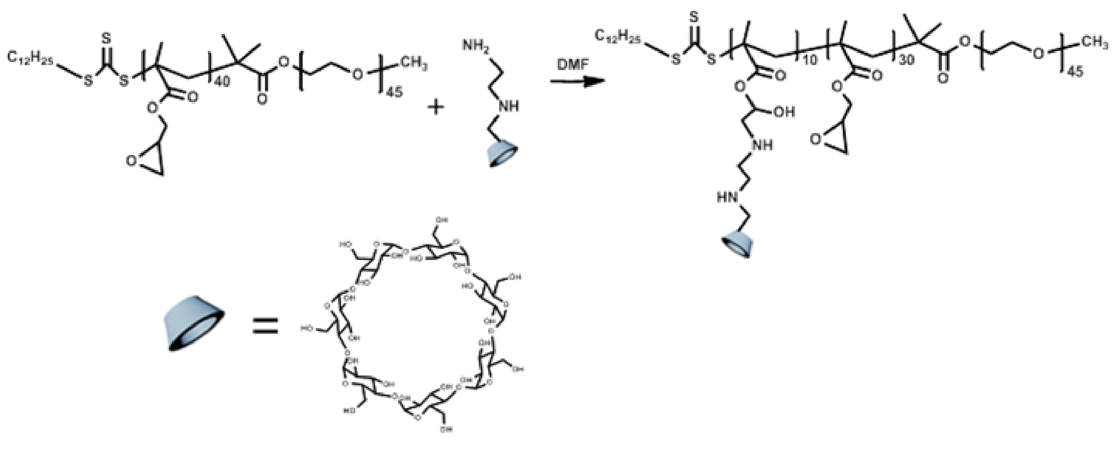
3.5. Synthesis of poly(ethylene oxide)-block-poly(glycidyl methacrylate) (PEG-b-PGMA).



PEG-*b*-PGMA diblock copolymer was synthesized via RAFT polymerization of GMA monomer using PEG macro-initiator. Typically, into a Schlenk flask equipped with a magnetic stirring bar, PEG-RAFT agent (1.17 g, 0.5 mmol), GMA (2.84 g, 20 mmol), AIBN (10 mg, 0.06 mmol), and THF (6 mL) were charged. The flask was carefully degassed by three freeze-pump-thaw cycles and then sealed under vacuum. After thermo-stating at 60 °C for 8 h, the reaction tube was quenched into liquid nitrogen, opened, and diluted with THF. The reaction mixture was precipitated into excess petroleum ether. The above dissolution-precipitation cycle was repeated three times. After drying in a vacuum oven overnight at room temperature, PEG-*b*-PGMA diblock copolymer was obtained as a red solid(yield: ~54%). The DP of PGMA block was determined to be 40 by ^{1}H NMR analysis in CDCl$_3$.

$M\mathrm{n}_{GPC}$ = 7400 g/mol, $M\mathrm{w}/M\mathrm{n}$ = 1.12.^{1}H NMR (CDCl$_3$, δ, ppm): 4.40-4.22 (40H,$-COOC\boldsymbol{H_2}-$ in PGMA), 3.83 (39H, $-COOC\boldsymbol{H_2}-$ in PGMA), 3.60 (180H, $-OC\boldsymbol{H_2}C\boldsymbol{H}_2O-$), 3.26 (41H, $-CH_2C\boldsymbol{H}OCH_2$ in PGMA), 2.80 (39H, $-CH_2CHOC\boldsymbol{H_2}$ in PGMA), 2.64 (s, 39H, $-CH_2CHOC\boldsymbol{H_2}$ in PGMA).

3.6. Synthesis of poly(ethylene oxide)-block-poly(glycidyl methacrylate)-graft-β-CD (PEG-b-PGMA-g-CD).



A typical procedure was shown as follows: A fixed molar ratio of CD-EDA and PEG-*b*-PGMA (80:1) was dissolved in 30 ml DMF, and then a small amount of 1, 4-dihydroxybenzene was added. The reaction lasted 6 h at 60°C. After the reaction was completed, dialyze to remove small molecules of the solvent and unreacted reagents, and freeze-dry to get white powder solid.

Mn_{GPC} = 17800 g/mol, Mw/Mn = 1.19.^{1}H NMR ($CDCl_3$, δ, ppm): 4.40-4.20 (40H, -COOC$\boldsymbol{H_2}$–), 3.86-3.61(H-3, 5, 6 in β-CD), 3.52 (180H, -OC$\boldsymbol{H_2}$C$\boldsymbol{H_2}$O-), 3.30-3.10 (41H, -CH_2C$\boldsymbol{H}$OCH_2 and –OC$\boldsymbol{H}$OHCH_2–), 2.72 (28H, -CH_2CHOC$\boldsymbol{H_2}$–in PGMA), 2.48(21H, NH_2C$\boldsymbol{H_2}$C$\boldsymbol{H_2}$–).

Table S1. Structural parameters of block copolymers

Sample codes	M_n^a (kDa)	M_n^b(kDa)	PDI^b
PEG-*b*-PGMA	7.21	7.40	1.12
PEG-*b*-PGMA-*g*-CD	16.9	17.8	1.19

a) Determined by ^{1}H NMR; b) Determined by GPC using DMF as the eluent.

3.7. The procedure of forming the micelles.

Typically, we used the nanoprecipitation method to prepare the polymer aqueous solution: 30 mg of the pesudocopolymers were firstly dissolved in 1 mL THF, a good solvent to all blocks.The ultrasound method was used to ensure the solid completely dissolved.Then 9 mL deionized water was injected at a slow rate (1.0 mL/h) to yield a translucent colloidal solution and the organic phase was finally removed by dialysis.

4. Supportive Evidence and Results

4.1. FT-IR of PEG-b-PGMA and PEG-b-PGMA-g-CD.

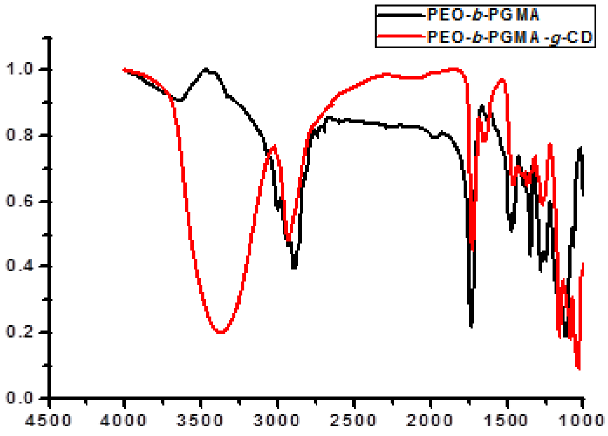


Fig. S1. FT-IR spectra of PEG-*b*-PGMA and PEG-*b*-PGMA-*g*-CD.

The peak at 3416 cm^{-1} is due to the O-H stretching vibrations of hydroxyl group on the β-CD, which could confirm the synthesis of pendent β-CD.

4.2. Critical aggregation concentration (CAC) test of assemblies formed from supramolecular brushes.

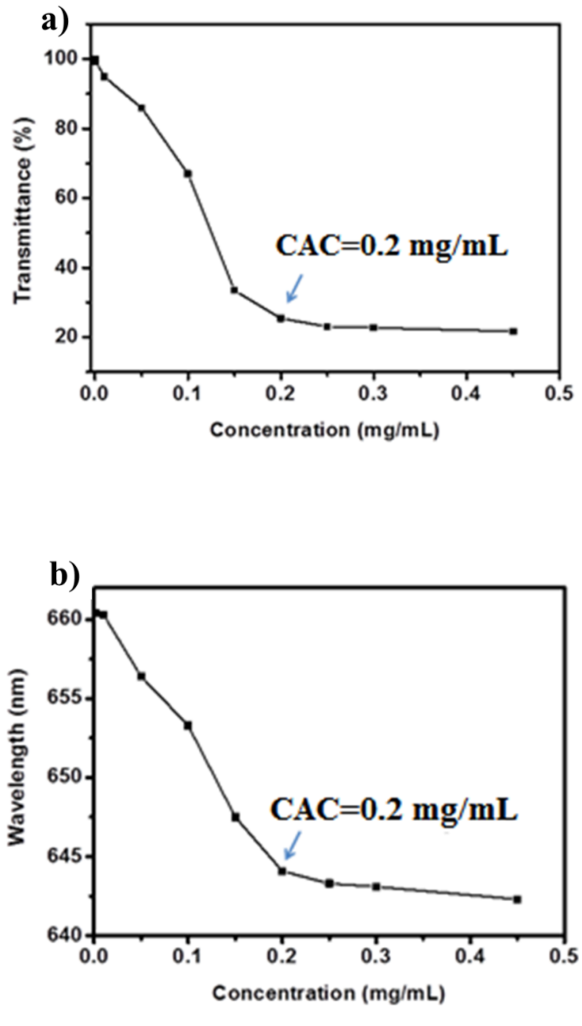


Fig. S2. Determination of CAC for the brush-like supramolecular block copolymers by: a) turbidity tests and b)Nile red (NR) probe.

By controlling the ratio between THF and water, after dialysing to remove THF, aqueous solutions of aggregates of different concentration were obtained. The CAC of the supramolecular block copolymers was firstly determined from the turbidity changes at different copolymer concentrations in aqueous media (a). Then CAC value was reassured by NR probe. A series of solutions with different concentrations were prepared and the CAC was monitored by the maximum fluorescence emission wavelength (b). The CAC of the superamphiphile is 0.2 mg/mL.

4.3. DLS measurements of assemblies formed from supramolecular brushes.

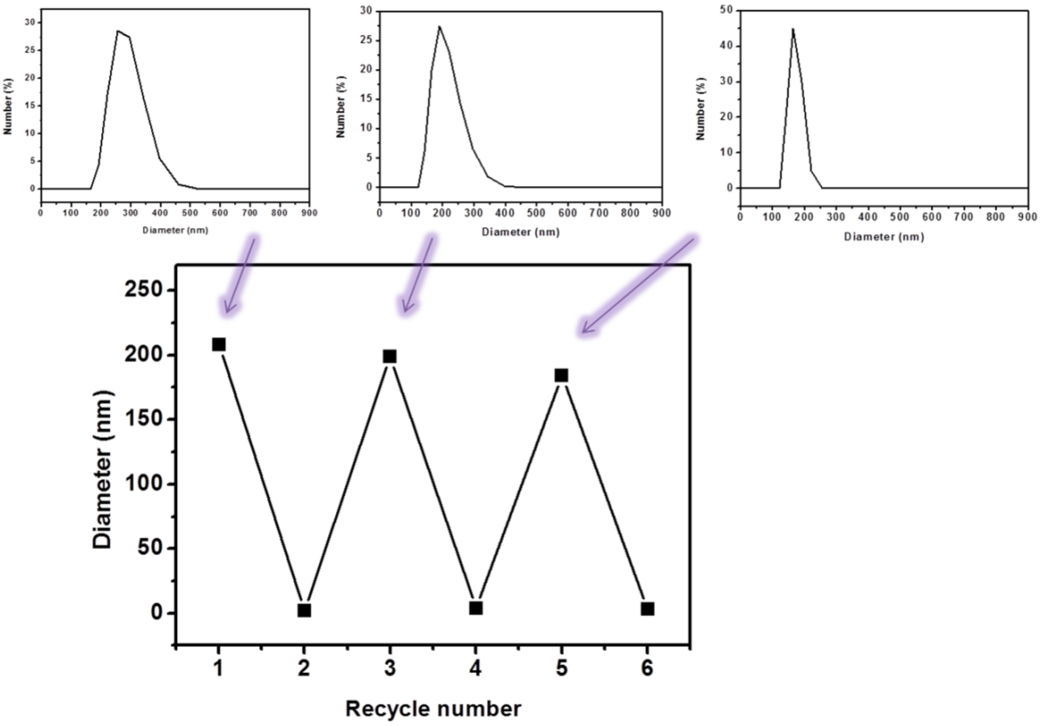


Fig. S3. DLS measurements confirm that the micelles possess voltage-controlled assembly and disassembly behaviors.

To exert an alternating potential stimuli (+1.0 V and −1.0 V) repeatedly to the micelle solution, we can observe a jumped average radius from 105 to 5 nm, demonstrating that the assemblies have a clear disassembly process at an oxidation voltage and have a reversible reassembly process at a reduction potential.

4.4. Characterization of PEG-b-PGMA-g-CD in aqueous solution.

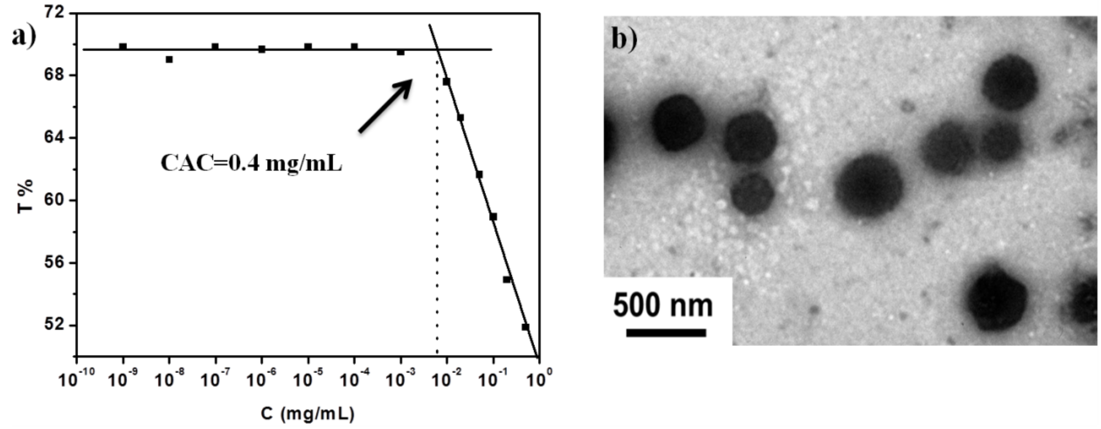


Fig. S4. CAC measurements (a) and TEM images (b) for characterization of PEG-*b*-PGMA-*g*-CD in aqueous solution.

With hydrophobic and hydrophilic parts coexisted in the polymer, PEG-*b*-PGMA-*g*-CD can self-assembly in aqueous solution. To ensure that the reformed micelles came from the supramolecular brushes other than PEG-*b*-PGMA-*g*-CD, we further characterized the properties PEG-*b*-PGMA-*g*-CD possessed in aqueous solution. Through the result of CAC measurements and TEM images (Fig. S4), we found that when the concentration beyond 0.4 mg/mL, it can self-assembly into spherical micelles with diameters of nearly 300 nm. However, the test concentration of electrochemical redox responsive polymeric micelles was around 0.2 mg/mL. In this condition, when Fc species converted to the charged Fc^+ form and the micelles completely disaggregated, the PEG-*b*-PGMA-*g*-CD existed in aqueous solution in the form of dissolved.

Notes and references

S1 J. Lai, D. Filla and R. Shea, *Macromolecules*, 2002, 35, 6754-6756.

S2 R. Petter, J. Salek, C. Sikorski, G. Kumaranel and F. Lin, *J. Am. Chem. Soc.*, 1990, 112, 3860-3868.

S3Y. Kang, J. Yuan, Q. Yan, L. Zheng and L. Zhou, *Polym. Adv. Technol.*, 2012, 23, 255-261.

S4Y. Kang, L. Zhou, X. Li and J. Yuan, *J. Mater. Chem.*, 2011, 21, 3704-3710.