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Supporting information

A synthetic approach toward pH and sugar responsive diblock copolymer *via* post-polymerization modification

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Materials

4-Acryloylmorphiline (AMP, 97%, Sigma-Aldrich) was purified *via* passing through a basic aluminium oxide column. 2,2'-Azobisisobutyronitrile (AIBN) was recrystallized twice from methanol. 4-Cyano-4-[(dodecylsulfanylthiocarbonyl)sulfanyl] pentanoic acid (CDTPA) as a chain transfer agent (CTA) was prepared following earlier published report.[1] Pentafluorophenyl acrylate was synthesized according to a published procedure.[2] 3-(Aminomethyl)phenylboronic acid hydrochloride (3AMPBA, 96%, Alfa Aesar), D(-)-fructose (Fru, 99%, Merck), Alizarin red S (ARS, Fluka), trimethylamine (Et_3N, 99%), Trioxane (Tri, 99%) and D (-) glucose (Glu, 99%) were purchased from Sigma and used as received.

Characterization

^{1}H NMR spectra were recorded on a Bruker Fourier 300 NMR spectrometer in $CDCl_3$, D_2O/NaOD, and tetramethylsilane (TMS) was used as an internal standard. ^{19}F NMR spectra were obtained on a Varian Gemini 2000BB spectrometer in $CDCl_3$ and MeOD. FT-IR spectra were recorded using the ATR (Smart iTR) unit on a Thermo Scientific Nicolet IS10 FT-IR spectrometer. Gel permeation chromatography (GPC) was used to measure the molecular weight and molecular weight distributions. The system was equipped with an intelligent AI12 pump, an RI 101 detector, two pre-columns MZ-Gel SDplus 50 × 8 mm with 50 Å and 100 Å, respectively, and column MZ-Gel SDplus 300 × 8 mm linear 5 µm. GPC measurements were carried out in DMF with a flow rate of 1.0 mL min^{-1}. Dynamic light scattering (DLS) measurements were recorded using a Malvern Zetasizer Nano-ZS90 apparatus equipped with a He–Ne laser operated at 632 nm and were measured at a scattering angle of 90°. Absorption spectra were measured on a JASCO V-630 UV/Vis-photospectrometer utilising Starna Silica (quartz) cuvette with 10 mm path lengths, two faces polished, while the fluorescence spectra were measured using a Jobin-Yvon-Horiba Fluoromax-4 spectrofluorometer utilising Starna Silica (quartz) cuvette with 10mm path lengths, four faces polished and the excitation wavelength was 470 nm.

Synthesis of (PAMPmacroCTA) as a macromolecular chain transfer agent by RAFT polymerization

The synthesis of macroCTA was as follows: the molar ratio of monomer: CTA: initiator was fixed at 100: 1: 0.1 for preparing the macroCTA. AMP (3.53 g, 25 mmol), CDTPA (0.1039 g, 0.25 mmol) and AIBN as the initiator (0.0049 g, 0.0298 mmol), trioxane as internal standard (0.378 g, 4.19 mmol), and dry 1,4-dioxane were charged into a dried Schleck tube, which was equipped with a magnetic stirring bar and sealed by a rubber septum. Then, the mixture was degassed by three freeze–thaw cycles and then argon was bubbled through the solution for 30 min. Then the tube was subsequently immersed in a preheated oil bath set at 70°C and the polymerization was allowed to proceed for 3 h. Next, the reaction was terminated by cooling down with liquid nitrogen and exposure to air. Subsequently, The crude mixture was purified by precipitation into cold diethyl ether to yield a yellow solid product, which was then dried in a vacuum oven at 40°C for 48 h prior to characterization (Yield = 2.05 g, 65 %)

^{1}H-NMR (300 MHz, $CDCl_3$): 0.88 ppm (t, 3H, –C**H$_3$** from CTA), 1.15 – 1.81 ppm (2H, d, C**H$_2$**-C), 3.24 - 3.80 ppm (8H, m, NC**H$_2$**C**H$_2$**O), 2.46 – 2.66 ppm (1H, t, C**H**-CO).

PAMP ($M_{n,NMR}$ = 10.4 x103 g mol-1) $M_{n,GPC}$ = 14.6 x103 g mol^{-1} PDI = 1.2

Synthesis of poly(4-acrylomorphiline)-*block*-poly(pentaflurorophenyl acrylate) (PAMP$_{70}$-*b*-PPFPA$_{39}$) by RAFT polymerization

The homopolymer PAMP as a macro-CTA was employed for the synthesis of the diblock copolymer (PAMP-*b*-PPFPA). The procedure was as follows: PAMPmacroCTA (0.5 g, 0.05 mmol), PFPA (1.22 g, 4.30 mmol), AIBN (0.0043 g, 0.026 mmol) and dry DMF were transferred into dried Schlenk tube was equipped with a magnetic stirring bar and sealed by a rubber septum. The solution purged with nitrogen for 30 min. Subsequently, the tube was immersed in a pre-heated oil bath set at 70°C and polymer was allowed to proceed for 16 h prior to termination using a combination of cooling and exposure to air. The block copolymer was purified by a series of precipitations in hexane followed by isolation by centrifuge and drying in vacuum oven at 40°C. (Yield: 0.5 g, 32%).

^{1}H-NMR (300 MHz, $CDCl_3$): 1.26 – 1.68 ppm (2H, d, C**H$_2$**-C), 3.29 - 3.41 ppm (8H, m, NC**H$_2$**C**H$_2$**O), 2.46 – 2.57 ppm (1H, t, C**H**-CO) from PAMP backbone, and 2.12 ppm (-C**H**-CH_2), 2.50 – 2.12 ppm (-CH-C**H$_2$**) in the backbone of PPFPA

^{19}F NMR (200 MHz, CDCl3): −152.54 ppm (ortho), −157.57 ppm (para) and −161.82 ppm (meta) from PPFPA.

PAMP-*b*-PFPA ($M_{n,NMR}$ = 21.4 x10^3 g mol^{-1}) $M_{n,GPC}$ = 25 x10^3 g mol^{-1} PDI = 1.4

Synthesis of poly(4-acraylomorphiline)-*block*-poly(3-aminomethylphenylboronic acid) *via* post-polymerization modification

Sugar-responsive moieties in the form of phenyl boronic acid functionalized amine was introduced to **(PAMP$_{70}$-*b*-PFPA$_{39}$)** *via* post-polymerization modification of the PFPA part in the block copolymer. The block copolymer (0.2 g, 1 equiv. of PFPA unit) was dissolved in dry DMF. Separately, 3AMPBA amine (1 equiv.) and TEA (1.5 equiv.) were dissolved in 1 mL DMF and then add to the block copolymer solution and stirred at 40°C for 24 h. The resulting block copolymer was dissolved in basic water (pH 10) and dialyzed against deionized water (DI) for 48 hrs using a 6000 MWCO membrane and the final block copolymer was obtained after lyophilization as a white solid. (Yield: 0,135 g , 67.5%).

^{1}H NMR (300 MHz, D_2O/NaOD): 6.58 - 7.50 ppm (Ar**H**), 4.22 ppm (-O**H**), 2.13-2.26 ppm (-C**H$_2$**) , 2.31ppm (-C**H**) in the backbone of P3AMPBA

p*K*a determination by potentiometric titration.

In order to determine the p*k*a of the block copolymer, pH measurements were carried out (using Mettler Toledo F20 pH meter). Basically pH of block copolymer solution (1 mg/mL) was adjusted to pH 11 by adding NaOH (0.1M). Then the pH value of the solution was measured by titration with HCl (0.001M). After determining the equivalent point, pH corresponding to the half of the equivalence was taken as the apparent pK_a of the block copolymer.

Typical procedure for the self-assembly of block copolymer

Block copolymer **($PAMP_{70}$-*b*-$PFPA_{39}$)** (12 mg) was dissolved in 2 mL basic water (pH ≈10.5). DI water was added dropwise until opalescence appeared. Then the solution was dialyzed (MWCO 6000 Da) against deionized water for 48 hrs. After that, the volume of the solution was increased to 24mL to obtain the solution with a concentration of 0.5 mg/mL. The pH was adjusted to 11, sonicated for 1 hr and filtered with 0.45 µm Millipore membrane filter. DLS measurements were recorded at 25°C and each measurement was performed after equilibration for 2 min. For pH responsive behavior, the pH of the solutions was adjusted with the addition of a freshly prepared aqueous solution of HCl (0.1M) and NaOH (0.1M), respectively. For sugar responsive behavior, a 1.5 mL solution of 200 mM Glucose and Fructose (pH 7.4) was added to the polymer solution (1.5 mL, 0.5 mg/mL) to obtain the micelle solution with a concentration of 0.25 mg/mL with 100 mM fructose at pH 7.4. The micelle solution was filtered through 0.45 µm Millipore membrane filter before DLS measurement. The pH of the solution was adjusted to 9 with an aqueous solution of NaOH (0.1M).

Binding affinity with Glucose and Fructose

ARS dye solution (10^{-4} M) , block copolymer solution (1 mg mL^{-1}), Glucose and Fructose (200 mM) all prepared in deionized water. The pH of the measured samples was adjusted with an aqueous solution of NaOH (0.1 M). The absorption and emssion spectra were performed on the ARS, ARS, and polymer in the absence and prescence of Glucose and Fructose at pH 7.4 and 9.

Results

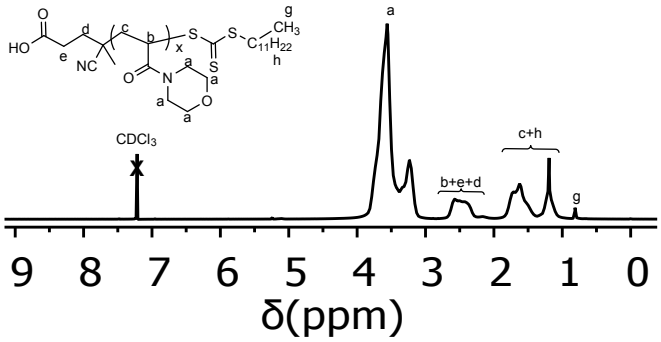

Fig. S1. ^{1}H NMR spectrum of PAMP macroCTA in $CDCl_3$

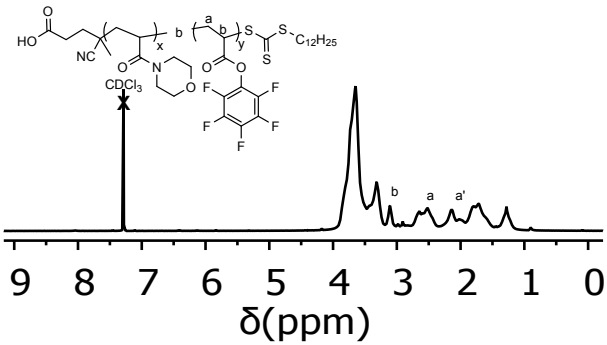

Fig. S2. ^{1}H NMR spectrum of $PAMP_{70}$-*b*-$PPFPA_{39}$ in $CDCl_3$

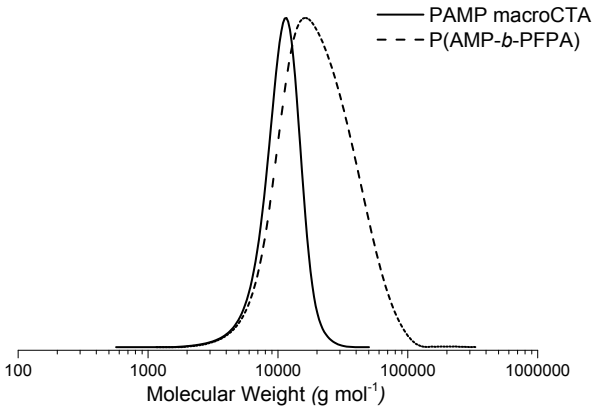

Fig. S3. GPC traces of PAMPmacroCTA (solid line) and $PAMP_{70}$-*b*-$PFPA_{39}$ (dashed line).

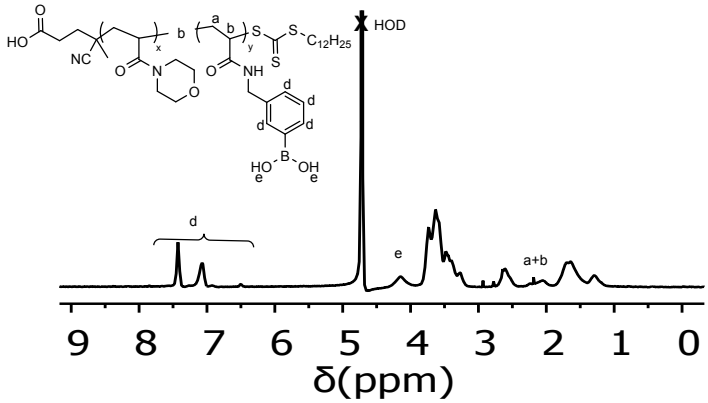

Fig. S4. ^{1}H NMR spectrum of PAMP-*b*-3AMPBA in D_2O/NaOD

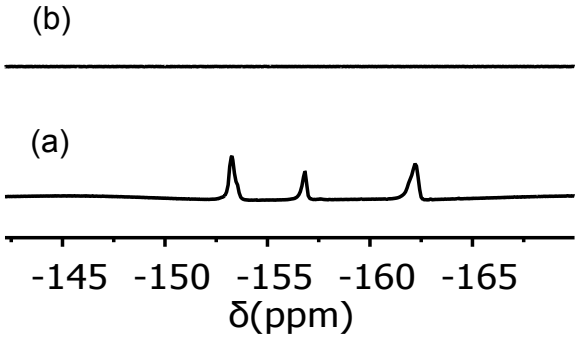

Fig. S5. ^{19}F NMR spectra of (a) $PAMP_{70}$-*b*-$PFPA_{39}$ and (b) PAMP-*b*-3AMPBA in $CDCl_3$

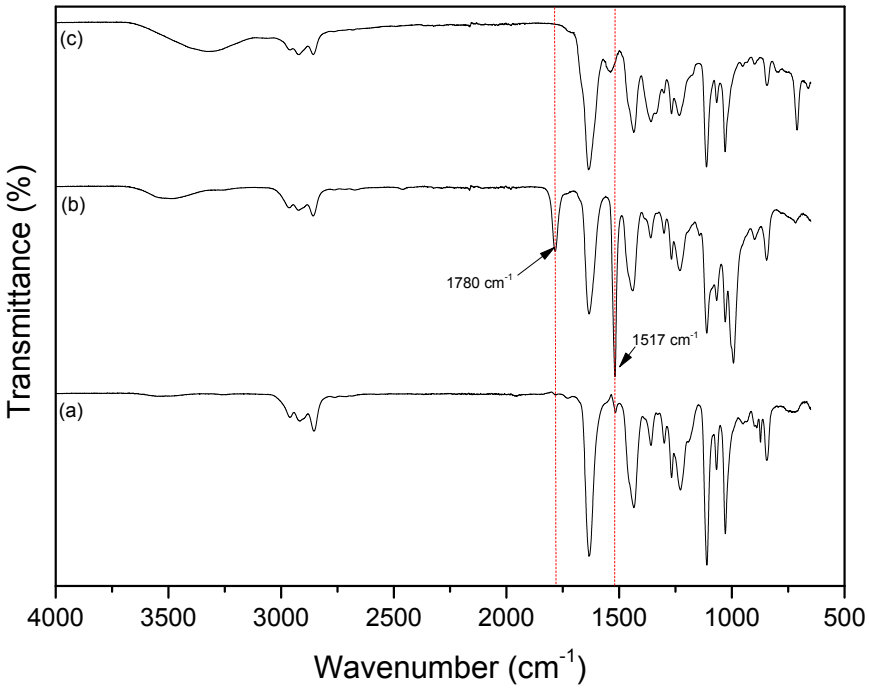

Fig. S6. FT-IR spectra of (a) PAMPmacroCTA, (b) $PAMP_{70}$-*b*-$PFPA_{39}$ and (c) PAMP-*b* - 3AMPBA

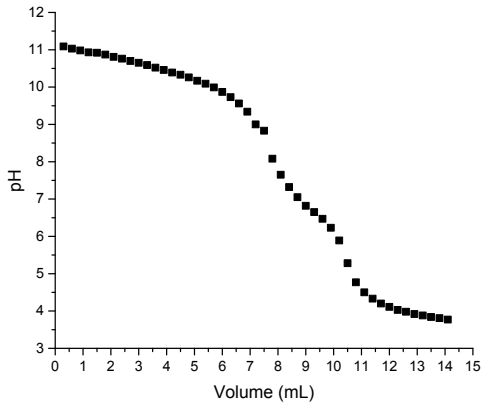

Fig. S7. pK_a determination of diblock copolymer by titration method

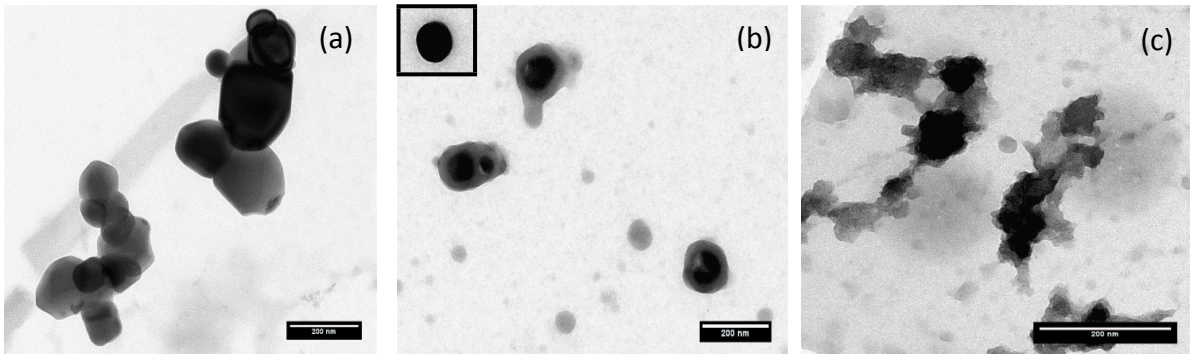

Fig. S8. TEM images of diblock copolymer (a) pH 4, (b) pH 7.4 and (c) pH 9

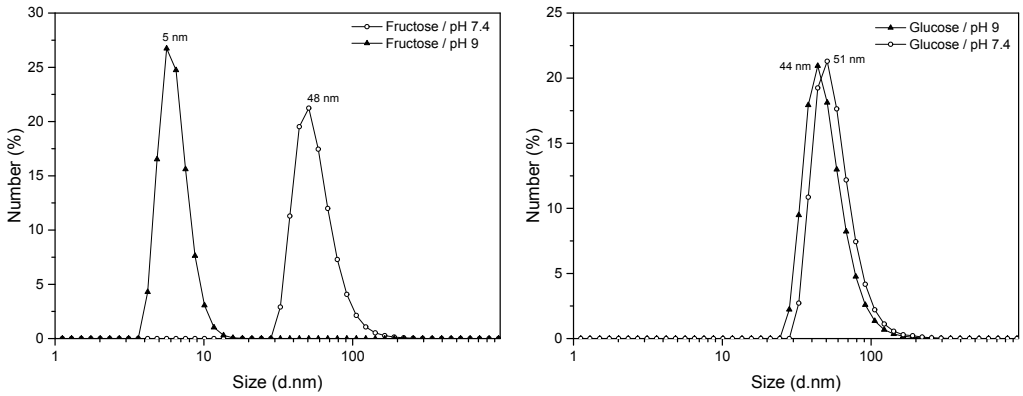

Fig. S9. DLS size distribution in the presence of glucose and fructose (100 mM) at pH 7.4 and pH 9.

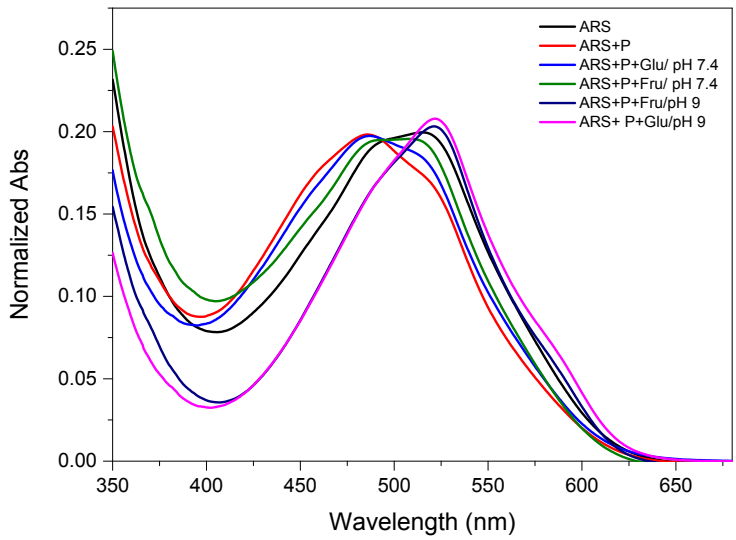

Fig. S10. Absorption spectra of ARS in the presence of PBA block copolymer and after the addition of Glu and Fru (100mM) at pH 7.4 and pH 9.

Fig. S11. Photos of ARS (left), ARS-Polymer complex (middle), ARS-Polymer-Fru at pH 9 (right).

References

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2 M. Eberhardt, R. Mruk, R. Zentel and P. Théato, *Eur. Polym. J.*, 2005, **41**, 1569–1575.