

Electronic supplementary information (ESI)

Metallo-supramolecular complexes from mPEG/PDPA diblock copolymers and their self-assembled strip nanosheets

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Part 1. Experimental section

1.1 Materials.

Pentaerythritol was from Sinopharm Chemical Reagent Co., Ltd, Shanghai, China. Methoxypolyethylene glycols (mPEG, average molecular weight 4000) and 4-ethylmorpholine (99%) were purchased from TCI, Japan. 2-(Diisopropylamino)ethyl methacrylate (DPA) was from Sigma-Aldrich. 2-Chloropropionyl chloride (CPC, 95%), succinic anhydride (99%), *N*-(3-dimethylaminopropyl)-*N*-ethylcarbodiimide hydrochloride (EDCI, 99%), 4-dimethylaminopyridine (DMAP, 99%), 1,1,4,7,7-pentamethyldiethylenetriamine (PMDETA, 98%), ruthenium(III) chloride hydrate (99.9%) and ammonium hexafluorophosphate (99%) were purchased from J & K Scientific Ltd. 4-([2,2':6',2''-Terpyridin]-4'-yl)benzoic acid (97%) was from ARK. Cuprous chloride ($\geq 97.0\%$, Tianjin Tianli Chemical Reagent Co. Ltd) was purified by stirring it in acetic acid overnight. After filtration, it was washed with anhydrous methanol and then dried in a vacuum oven at 25 °C.

All of the other reagents, including, dichloromethane (DCM), *n*-hexane, ethyl acetate, methanol, tetrahydrofuran (THF), *N,N*-dimethylformamide (DMF), ethanol absolute, ether, sodium bicarbonate and anhydrous sodium sulfate were of analytical grade and made in China.

1.2. Synthesis of 3-hydroxy-2,2-bis(hydroxymethyl)propyl 2-chloropropanoate (@(-OH)₃-Cl)

Pentaerythritol (7 g, 0.051 mol) was first dissolved in dry DMF. Then, a solution of CPC (5.25 mL) in DMF was added stepwise into the pentaerythritol solution at 50°C. The reaction mixture was stirred at 50°C for additional 24 h and the DMF was then removed by a rotary evaporator. The solution was diluted with water and was then neutralized with 10 wt% NaOH solution to ca. pH 9. After the water was removed by a rotary evaporator, DCM and THF were added to extract the resulting product from the mixture. The filtrate was dried over anhydrous sodium sulfate in the presence of sodium bicarbonate overnight, and the organic solvent was then removed by a rotary evaporator. The obtained crude product was purified by a silica gel column using DCM/methanol mixture as the eluent. After drying, 2.8 g of product was obtained. ¹H NMR (DMSO-*d*₆): δ = 4.72 (1H, $-\underline{\text{C}}\text{H}(\text{CH}_3)\text{Cl}$), 4.41(3H, $-\text{CH}_2\underline{\text{O}}\text{H}$), 4.05 (2H, $-\underline{\text{C}}\text{H}_2-\text{O}-\text{C}(=\text{O})-$), 3.38 (6H, $-\underline{\text{C}}\text{H}_2\text{OH}$), 1.58 (3H, $-\text{CH}(\text{Cl})\underline{\text{C}}\text{H}_3$) (**Fig. S1**).

1.3. Synthesis of succinoylate mPEG (mPEG-COOH)

mPEG-COOH were synthesized by the reaction of mPEG with succinic anhydride.^{1,2} Specifically, mPEG (4 g, 1 mmol), succinic anhydride (0.15 g, 1.5 mmol) and DMAP (0.18 g, 1.5 mmol) were successively dissolved in 16 mL of dry DMF. The mixture was stirred at 30 °C for 24 h. After

completion of the reaction, the DMF solution was added to a heated ethyl acetate/*n*-hexane mixture. When cooled, the solids were precipitated out. After the solids collected were dissolved in DCM, it was still added into a heated ethyl acetate/*n*-hexane mixture. When cooled, the solids precipitated were collected. The purification was repeated thrice. Finally, after the product was dissolved in DCM and was precipitated in ether, it was dried under vacuum at 25 °C for 48 h (yield, 3.5 g).

1.4. Synthesis of ATRP macroinitiator mPEG(-OH)₂-Cl

0.97g (4.28 mmol) of @(-OH)₃-Cl, 2.5 g (0.61 mmol) of mPEG-COOH, and 22.4 mg (0.18 mmol) of DMAP was successively dissolved in 15 mL of dry DCM. Under the condition of 0 °C, a solution of EDCI (175.3 mg, 0.91 mmol) in 10 mL of dry DCM was stepwise added. After stirring for 1 h at 0 °C, the reaction mixture was stirred at 20 °C for additional 48 h. Then, it was diluted with DCM, and was washed by 1 wt% HCl aqueous solution and 20 wt% NaCl aqueous solution thrice, respectively. After it was dried over anhydrous sodium sulfate overnight and was concentrated by a rotary evaporator, it was precipitated in *n*-hexane. The obtained polymer was dried under vacuum at room temperature for 48 h (yield, 2.4 g).

1.5. Synthesis of mPEG(-*b*-OH)₂-*b*-PDPA_x

mPEG(-*b*-OH)₂-*b*-PDPA_x were synthesized by using mPEG(-OH)₂-Cl as the ATRP macroinitiator. Typically, PMDETA (86.9 mg, 0.5 mmol), mPEG(-OH)₂-Cl (1.8 g, 0.42 mmol) and DPA (1.78 g, 8.36 mmol) were successively dissolved in 6.3 mL of DMF. After it was bubbled with argon gas for 45 min, CuCl (42 mg, 0.42 mmol) was added. It was bubbled with argon gas for additional 15 min and the reaction system sealed was then stirred magnetically at 70 °C. After 5 h, the reaction mixture was allowed to cool to room temperature, to be exposed to air, and to be diluted with DCM. It was passed through a neutral alumina column for removing the copper complex. The resulting solution was concentrated and it was then subjected to dialysis (molecular weight cutoff: 3500) against methanol for 36 h. After the solvent was removed by a rotary evaporator, the obtained product was dried under vacuum at 40 °C for 48 h.

1.6. Synthesis of mPEG(-*b*-Tpyr)₂-*b*-PDPA_x

mPEG(-*b*-Tpyr)₂-*b*-PDPA_x were synthesized through the esterification of mPEG(-*b*-OH)₂-*b*-PDPA_x with 4-([2,2':6',2''-terpyridin]-4'-yl)benzoic acid in DMF in the presence of EDCI/DMAP. Typically, 4-([2,2':6',2''-terpyridin]-4'-yl)benzoic acid (0.32 g, 0.9 mmol) was firstly dissolved in 30 mL of dry DMF. Then, mPEG(-*b*-OH)₂-*b*-PDPA₃₁ (1.6 g, 0.15 mmol) and DMAP (0.11 g, 0.9 mmol) were added. Afterwards, under the condition of 0 °C, a solution of EDCI (0.35 g, 1.8 mmol) in 10 mL of dry DMF was stepwise added. After the mixture was stirred at 0°C for 1 h, it was continuously

stirred at 25°C for 24 h and at 35 °C for additional 24 h. When completion of the reaction, it was diluted with DCM and was then washed by 20 wt% NaCl aqueous solution several times. After it was dried over anhydrous sodium sulfate overnight and was concentrated by a rotary evaporator, it was subjected to dialysis (molecular weight cutoff: 1000) against DMF for 45 h and against DCM for additional 10 h. After the solvent was removed by a rotary evaporator, the obtained product was dried under vacuum at 40 °C for 48 h.

1.7. Synthesis of mPEG(-*b*-Tpyr)₂-*b*-PDPA_x/Ru(II) complexes

Typically, mPEG(-*b*-Tpyr)₂-*b*-PDPA_x (100 mg) was dissolved in 5 mL of ethanol, and then ethanol solution of ruthenium (III) trichloride (equimolar ratio) was added.^{3,4} After the mixture was refluxed at 75 °C for 12 h, 50 µL of 4-ethylmorpholine was added. The mixture was refluxed for additional 24 h. After ammonium hexafluorophosphate was added, the mixture was stirred at 75°C for additional 2 h. When cooled to room temperature, it was then subjected to dialysis (molecular weight cutoff: 1000) against water for 24 h and against ethanol for additional 10 h. After the ethanol was removed, the obtained product was dissolved in THF for further application.

1.8. Preparation of self-assemblies

Preparation of MSC23 (MSC33 or MSC44) self-assemblies: 12 mL of water was added dropwise into 4 mL of THF solution of MSC23 (MSC33 or MSC44) under magnetic stirring in 1 h. After the system was stirred at room temperature for additional 3 h, the THF was removed by reduced pressure. The obtained solution was diluted with pure water to 1 mg mL⁻¹ of the polymer for further measurements.

Preparation of mPEG(-*b*-Tpyr)₂-*b*-PDPA_x self-assemblies: sample mPEG(-*b*-Tpyr)₂-*b*-PDPA_x was firstly dissolved in THF. The following procedure is similar with that of preparation of MSC23 self-assemblies.

The pHs of the aggregate solutions were adjusted by addition of 1M HCl, or NaOH aqueous solution.

1.9. Preparation of NR-loaded self-assemblies

The THF solutions of Nile red (NR) and MSC23 (MSC33 or MSC44) were mixed. The following procedure is similar with that of section 1.8, but the THF was removed by slow volatilization at room temperature. Note, herein the NR dosage used should not lead to visible precipitate when the THF was removed. The NR-loaded samples were diluted with water to 10 times for fluorescent spectrum measurement (**Fig.S12**).

1.10. Analyses

¹H NMR measurements were conducted on Bruker Ascend 400M spectrometer by using CDCl₃, DMSO-*d*₆, or CD₃CN as solvent. The molecular structure parameters of copolymers were determined on a gel-permeation chromatography (GPC, Waters 1515). HPLC-grade THF was used as the eluent at a flow rate of 1 mL min⁻¹ at 40°C. Calibration was done with polystyrene standards. The Z-average diameters (*D*_{zs}) of samples were characterized by Zetasizer Nano-ZS dynamic light scattering (DLS) (Malvern, Britain). UV/vis spectra were recorded on a spectrophotometer UV-2550 model (Shimadzu, Japan). The morphologies of self-assemblies were observed by transmission electron microscopy (TEM, FEI talos F200X).

Atomic force microscopy (AFM, Bruker, Dimension Icon) studies were conducted using tapping mode under ambient conditions. AFM images were from silicon wafer substrate adsorbed by self-assembled aggregates. The silicon wafer substrate was immersed in the solution of the self-assembled aggregates and washed with pure water and then dried under ambient atmosphere.⁵

Fluorescence was recorded by using a fluorescence spectrophotometer (F-4600, Hitachi, Japan). The excitation wavelength for measurement of NR solution was 485 nm.⁶

References

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Part 2 Figure Section

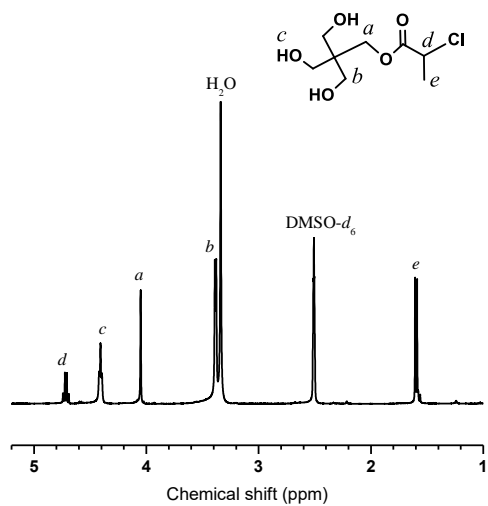


Fig. S1 ^1H NMR spectrum of $(\text{HO})_3\text{C}-\text{Cl}$.

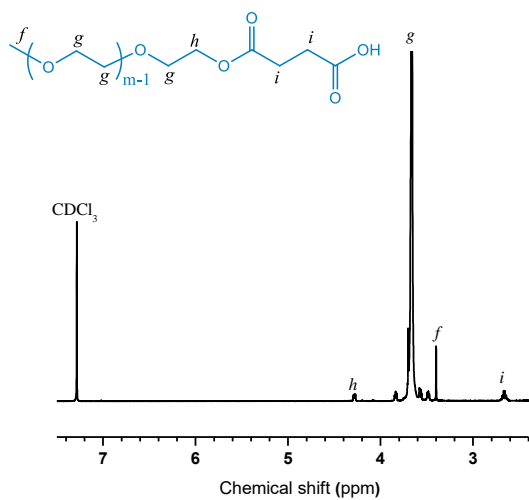


Fig. S2 ^1H NMR spectrum of $m\text{PEG}-\text{COOH}$.

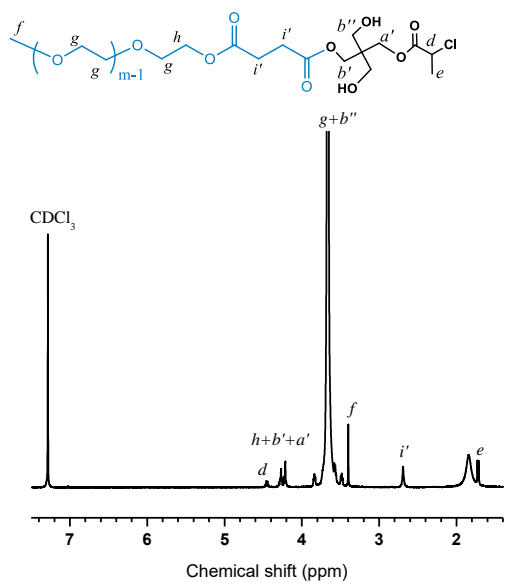


Fig. S3 ¹H NMR spectrum of mPEG(-OH)₂-Cl.

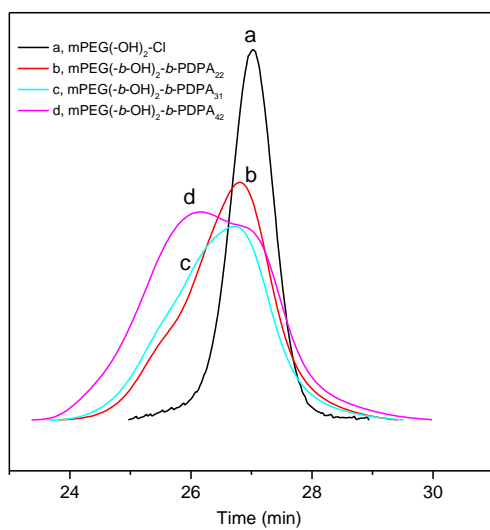


Fig. S4 GPC traces of mPEG(-b-OH)₂-b-PDPA_x and their precursor.

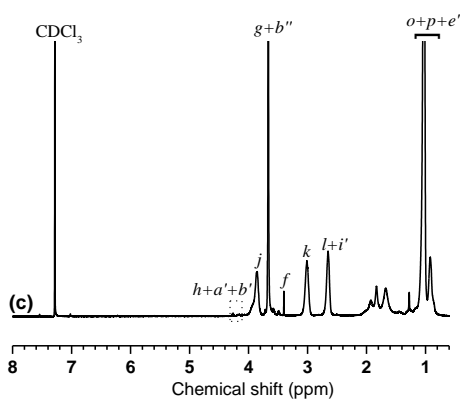
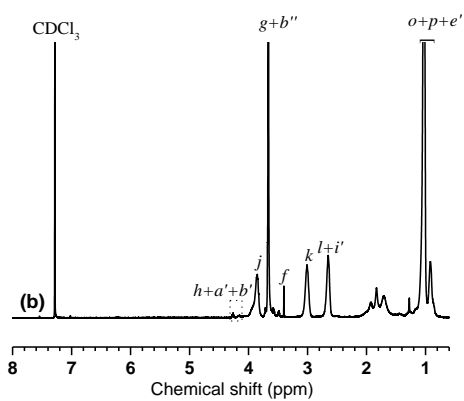
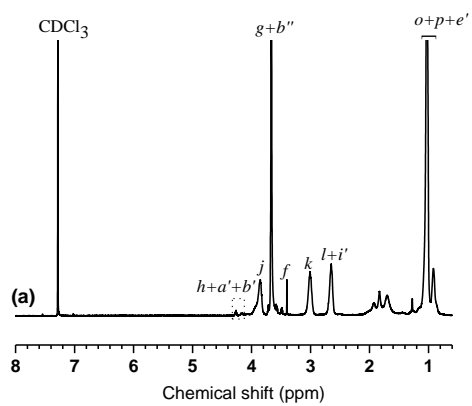
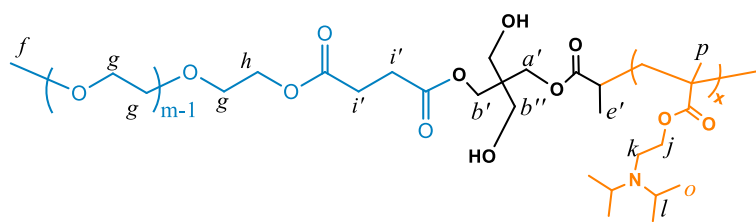


Fig. S5 ^1H NMR spectra of $\text{mPEG}(-b\text{-OH})_2\text{-}b\text{-PDPA}_x$ ($x = 22$ (a), 31(b) and 42(c)).

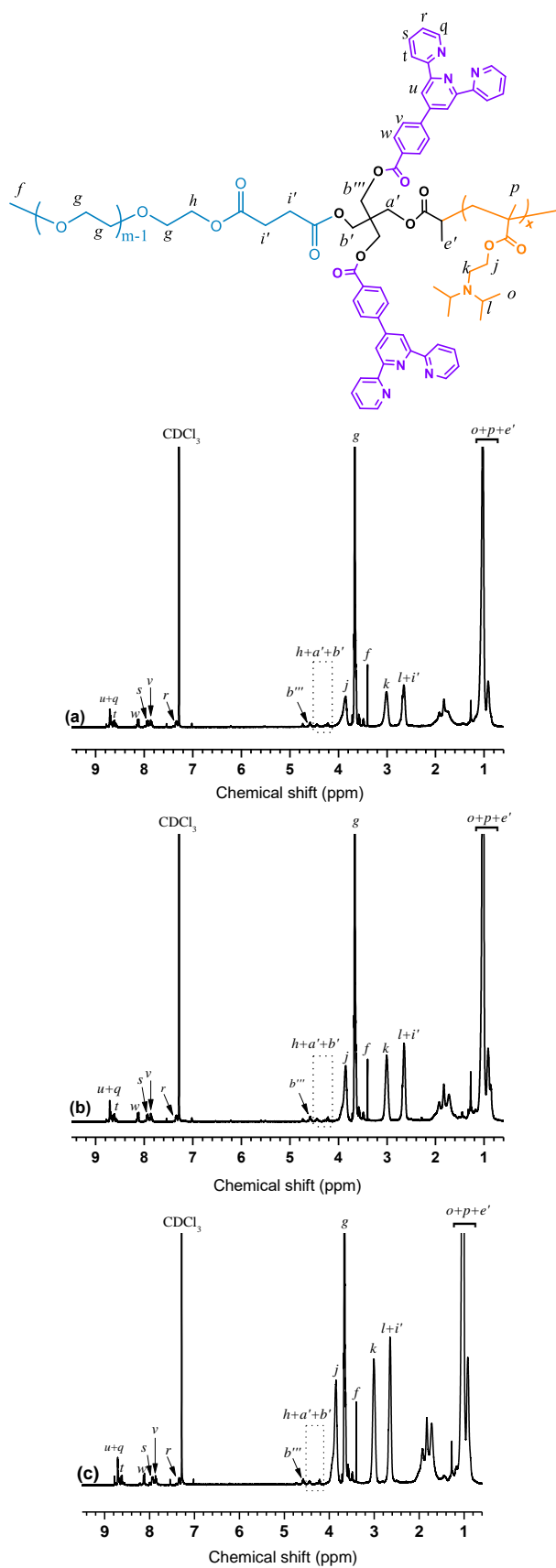


Fig. S6 ^1H NMR spectra of $m\text{PEG}(-b\text{-Tryp})_2-b\text{-PDPA}_x$ ($x = 23$ (a), 33(b) and 44(c)).

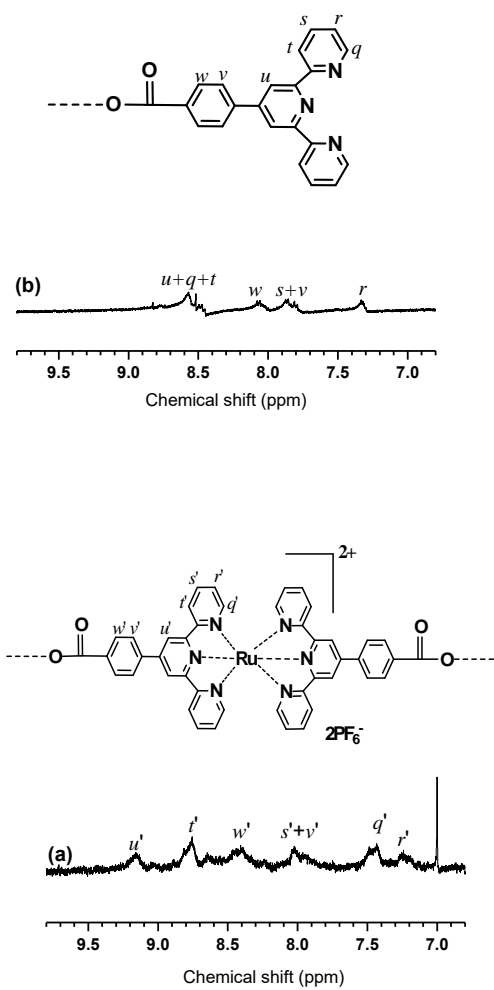


Fig. S7 ^1H NMR spectra of MSC33 (a) and mPEG(-b-Tryp) $_2$ -b-PDPA $_{33}$ (b) in CD_3CN .

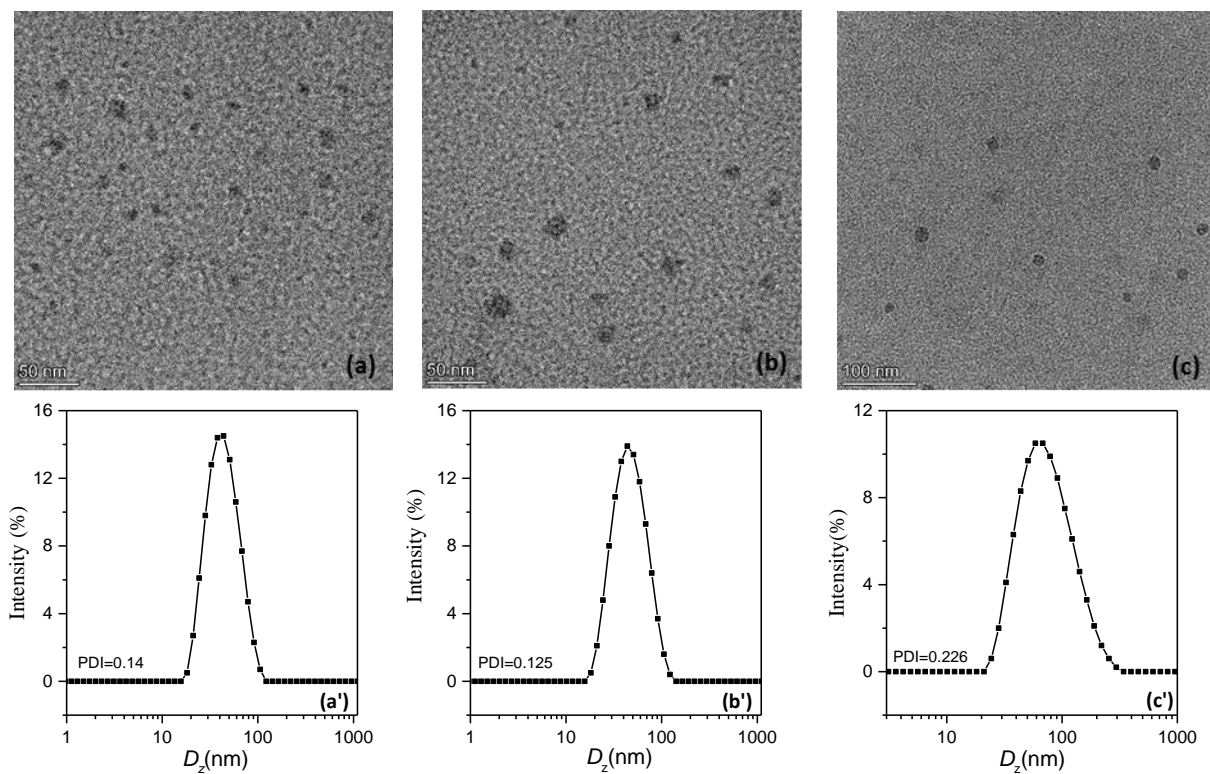


Fig. S8 TEM images (a, b and c) and DLS measurements (a', b' and c') of the self-assembled aggregates from mPEG(-*b*-Tpyp)₂-*b*-PDPA_{*x*} (a and a' for *x* = 23; b and b' for *x* = 33; c and c' for *x* = 44).

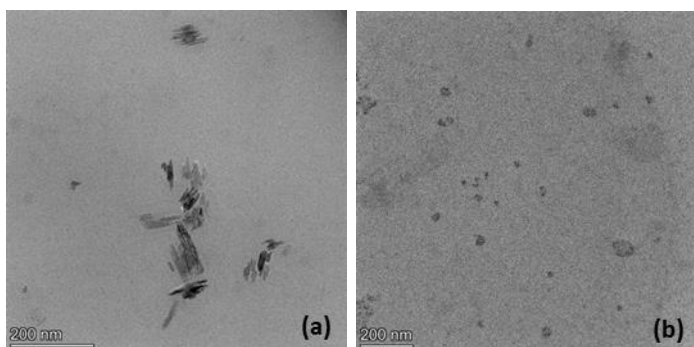


Fig. S9 TEM images of the self-assembled aggregates from MSC23 at pH 8.5 (a) and at pH 3.2 (b).

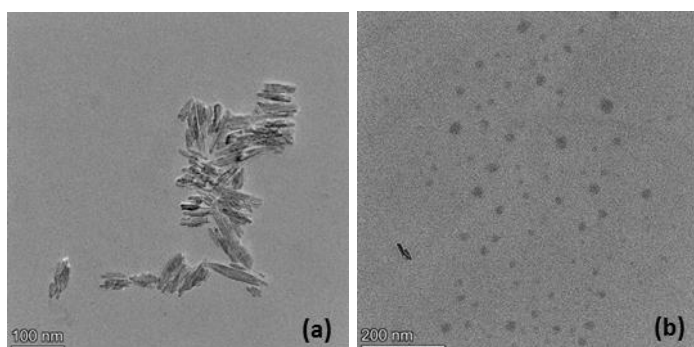


Fig. S10 TEM images of the self-assembled aggregates from MSC33 at pH 8.5 (a) and at pH 3.2 (b).

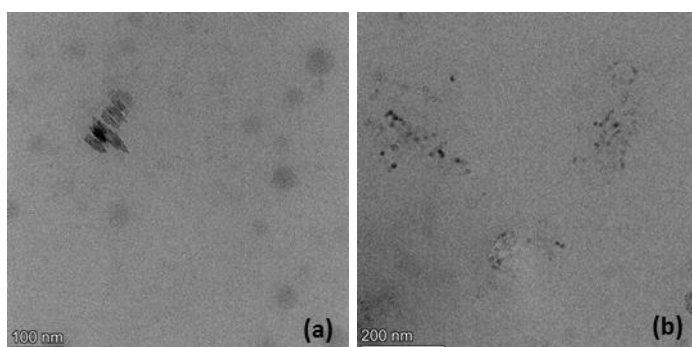


Fig. S11 TEM images of the self-assembled aggregates from MSC44 at pH 8.5 (a) and at pH 3.2 (b).

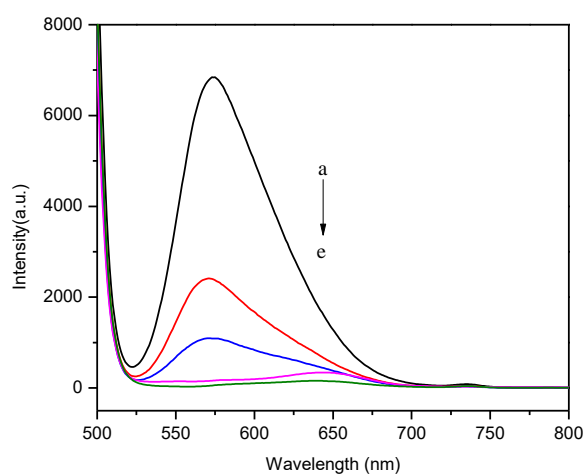


Fig. S12 Fluorescence spectra of the self-assembled samples from NR/MSC44 (1.75/100) (a), NR/MSC33 (0.75/100) (b), NR/MSC23 (1/100) (c) and MSC33 (e); Measurement condition of curve d: after the sample for curve b measurement was incubated at 37 °C for 19.5 h, its pH was adjusted to 3.2 and the fluorescence was determined as curve d after 30 min.

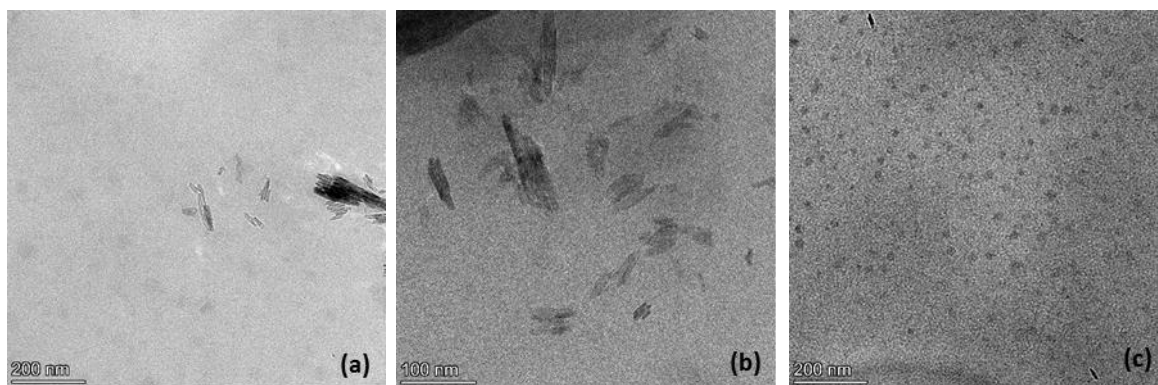


Fig. S13 TEM images of the self-assembled aggregates from NR/MS23 (1/100) (a), NR/MS33 (0.75/100) (b) and NR/MS44 (1.75/100) (c).