

# Supporting Information

## ***IN-SITU* FORMATION OF POLYMER-GOLD COMPOSITE NANOPARTICLES WITH TUNABLE MORPHOLOGIES**

**Richard Bleach,<sup>a</sup> Bunyamin Karagoz,<sup>a,b,c</sup> Shyam M. Prakash,<sup>b</sup> Thomas P. Davis,<sup>\*d,e</sup>  
Cyrille Boyer<sup>\*a,b</sup>**

<sup>a</sup> Centre for Advanced Macromolecular Design (CAMD), School of Chemical Engineering,  
University of New South Wales, Sydney NSW 2052, Australia

<sup>b</sup> Australian Centre for Nanomedicine, The University of New South Wales, Sydney NSW  
2052, Australia,

<sup>c</sup> Istanbul Technical University Department of Chemistry, Maslak 34469 Istanbul-Turkey

<sup>d</sup> ARC Centre for Convergent Bio-Nano Science & Technology, Monash Institute of  
Pharmaceutical Sciences, Monash University, Parkville, VIC 3052, Australia

<sup>e</sup> Department of Chemistry, University of Warwick, UK

Correspondence to: [cboyer@unsw.edu.au](mailto:cboyer@unsw.edu.au) and [Thomas.p.davis@monash.edu](mailto:Thomas.p.davis@monash.edu)

## EXPERIMENTAL PART

### Materials

*N,N*-Dimethylaminoethyl methacrylate (DMAEMA, Aldrich, 98 %), oligo(ethylene glycol) methacrylate (OEGMA) ( $M_n = 300 \text{ g mol}^{-1}$ ) and 2-cyano-2-propyl benzodithioate (CPDB, Aldrich, >97 %) were used as received. Styrene (ST) was de-inhibited by passing through a column of basic alumina. 2,2'-Azobis(isobutyronitrile) (AIBN, Fluka, 98 %) was purified by recrystallization from methanol. All the other reagents were used as purchased unless otherwise specified.

### Instrumentation

$^1\text{H}$ -NMR spectra were recorded using a Bruker 300MHz spectrometer. All chemical shifts were reported in ppm ( $\delta$ ) relative to tetramethylsilane, referenced to the chemical shifts of residual solvent resonances.

The molecular weight and polydispersity (PDI) of the prepared polymers were measured by SEC. DMAc was used as the eluent of SEC (that contained 0.03 % w/v LiBr and 0.05 % w/v 2,6-dibutyl-4-methylphenol (BHT)) at 50 °C (flow rate of  $1 \text{ mL min}^{-1}$ ) with a Shimadzu modular system comprising an SIL-10AD auto-injector, a Polymer Laboratories 5.0 $\mu\text{m}$  bead-size guard column ( $50 \times 7.5 \text{ mm}^2$ ) followed by four linear PL (Styragel) columns (105, 104, 103 and 500 Å) and a RID-10A differential refractive-index detector.

The sizes and morphologies of the triblock polymers were observed using a transmission electron microscopy JEOL1400 TEM at an accelerating voltage of 100 kV. The solution of the triblock polymer in dispersion medium was directly taken and diluted with methanol ( $2 \text{ mg mL}^{-1}$ ) and deposited onto copper grid (ProSciTech) without staining.

DLS measurements were performed using a Malvern ZetasizerNano Series running DTS software and using a 4 mW He-Ne laser operating at a wavelength of 633 nm and an avalanche photodiode (APD) detector.

The scattered light was detected at an angle of 173°. X-ray diffraction (XRD), the crystal characterization of gold nanoparticles tethered on the POEGMA-*b*-DMAEMA-*b*-PST triblock copolymers were performed using a Philips PANalytical Xpert X-ray Multipurpose Diffraction System at 40 mA and 45 kV using monochromated Cu-K $\alpha$  radiation ( $\lambda = 1.541 \text{ Å}$ , step size = 0.01, 0.02 or 0.05, time per step = 10 or 20 s/step).

Thermal gravimetric analysis (TGA) of the POEGMA-*b*-DMAEMA-*b*-PST/gold hybrid materials were performed in a Perkin-Elmer Thermogravimetric Analyzer (Pyris 1 TGA).

Pre-dried samples were heated from room temperature to 600°C at a constant temperature increase of 20°C min<sup>-1</sup> using air as the furnace gas. The weight loss was calculated through the difference between the weights at room temperature and at 600°C.

Fourier transform infrared spectroscopy (FTIR) was performed with a Bruker IFS 66/S instrument under attenuated total reflectance (ATR) and was analyzed on OPUS software version 4.0.

### **Preparation of poly(oligo(ethyleneglycol) methacrylate) (POEGMA) based macro-CTA via RAFT polymerization**

POEGMA based macro-CTA was synthesized via RAFT polymerization using CPDB as a CTA in acetonitrile. In this protocol, OEGMA (12 g,  $4 \times 10^{-2}$  mol), CPDB (0.177 g,  $8 \times 10^{-4}$  mol), AIBN ( $1.64 \times 10^{-2}$  g,  $1 \times 10^{-4}$  mol) and 50 mL acetonitrile were mixed in a 100 mL round bottom flask and sealed with a rubber septum. Polymerization reactions were carried out for 5.5 h at 70 °C in an inert atmosphere. The resulting polymers were purified by precipitation in 100 mL petroleum spirit (boiling range of 40-60°C) and diethyl ether mixture (7:3, v/v). POEGMA was dried in vacuum overnight at 50 °C to yield a red viscous liquid (5g, yield 42%). Dried POEGMA macro-CTA's molecular weight was determined by size exclusion chromatography (SEC).  $M_{n, SEC}$  (DMAc as mobile solvent and PST standards) of 7300 g mol<sup>-1</sup>, PDI= 1.08 and the theoretical number average molecular weight ( $M_{n, NMR}$ ) was 7700 g mol<sup>-1</sup> from <sup>1</sup>H-NMR of the purified polymer sample by comparing the RAFT agent's aromatic protons appearing between 8.0-7.3 ppm to that of methylene protons adjacent to *ester* linkage at 4.1 ppm. The POEGMA was used as macro-CTA for further block copolymer synthesis with *N,N*-dimethylaminoethyl methacrylate.

### **Preparation of POEGMA-*b*-DMAEMA block copolymer via RAFT polymerization**

Block copolymerization was carried out in toluene by RAFT polymerization with a molar feed ratio of DMAEMA: POEGMA: AIBN = 20:1:0.2. For this purpose, POEGMA as a macro-CTA (1.08 g,  $1.40 \times 10^{-4}$  mol), *N,N*-dimethylaminoethyl methacrylate (0.44 g,  $2.8 \times 10^{-3}$  mol) as a functional monomer and azobisisobutyronitrile (AIBN, 4.6×10<sup>-3</sup> g,  $2.8 \times 10^{-5}$  mol) were dissolved in 12 mL toluene and saturated with nitrogen. Then the reaction flask was immersed in an oil bath at 70°C for 6h. The block copolymer was purified in the same way as POEGMA macro-CTA purification. The polymer was dried under vacuum at 50 °C overnight to remove the trace of solvents and to yield red viscous liquid (1.3 g, 90%). From the <sup>1</sup>H-NMR spectra of the reaction mixture before and after polymerization, conversion of

DMAEMA was calculated at 75% by following the integral ratio of the monomer's vinyl protons against to the methyl protons of the POEGMA adjacent to *ether* linkage at 3.3 ppm. Resulting diblock copolymer had a  $M_{n, SEC} = 8500 \text{ g mol}^{-1}$ , PDI=1.09 and 15 repeating units of DMAEMA. In this recipe, 20% deactivation of the active chain end was observed from the repeating unit of the POEGMA between homopolymer and block copolymer  $^1\text{H-NMR}$  spectra by comparing the integral ratio of dithiobenzoate aromatic protons with methyl protons OEGMA adjacent to *ether* linkage at 3.3 ppm. Purified diblock copolymer was then used as macro-CTA for dispersion polymerization with styrene.

### **POEGMA-*b*-DMAEMA-*b*-PST copolymer synthesis in methanol via RAFT dispersion polymerization**

Dispersion polymerization of ST was achieved via RAFT polymerization in methanol by using POEGMA-*b*-DMAEMA as a macro-CTA as explained in the literature<sup>1</sup>. Briefly, POEGMA-*b*-DMAEMA (0.202 g,  $2 \times 10^{-5} \text{ mol}$ ), St (10.4 g, 0.10 mol), AIBN ( $1.1 \times 10^{-3} \text{ g}$ ,  $6.67 \times 10^{-6} \text{ mol}$ ) were mixed in 12.5 mL methanol. Then, the reaction mixture was divided equally into six vials, saturated with nitrogen in an ice-bath and sealed rapidly. The vials were then immersed in an oil bath at 70°C and taken out at specific time points 6, 12, 18, 24, 32 and 40 hours. Resulting triblock copolymer dispersions were purified by dialysis against methanol for 72 hours (methanol was changed every 12h), and then, against MiliQ water for 72 hours (water was changed every 12 h) using a dialysis tube with a molecular weight cut off of 6000 – 8000. Morphologies and number average diameter of the triblock copolymers were investigated after taking the triblock copolymer in aqueous dispersion by diluting the samples with distilled water (2 mg/mL). These purified triblock copolymers with different morphologies in aqueous dispersions were then used as a host material for the formation of gold nanoparticles. The yield of the polymer was reported in **Table S1**.

### **Gold nanoparticle formation onto POEGMA-*b*-DMAEMA-*b*-PST triblock copolymer nano-aggregates with various morphologies**

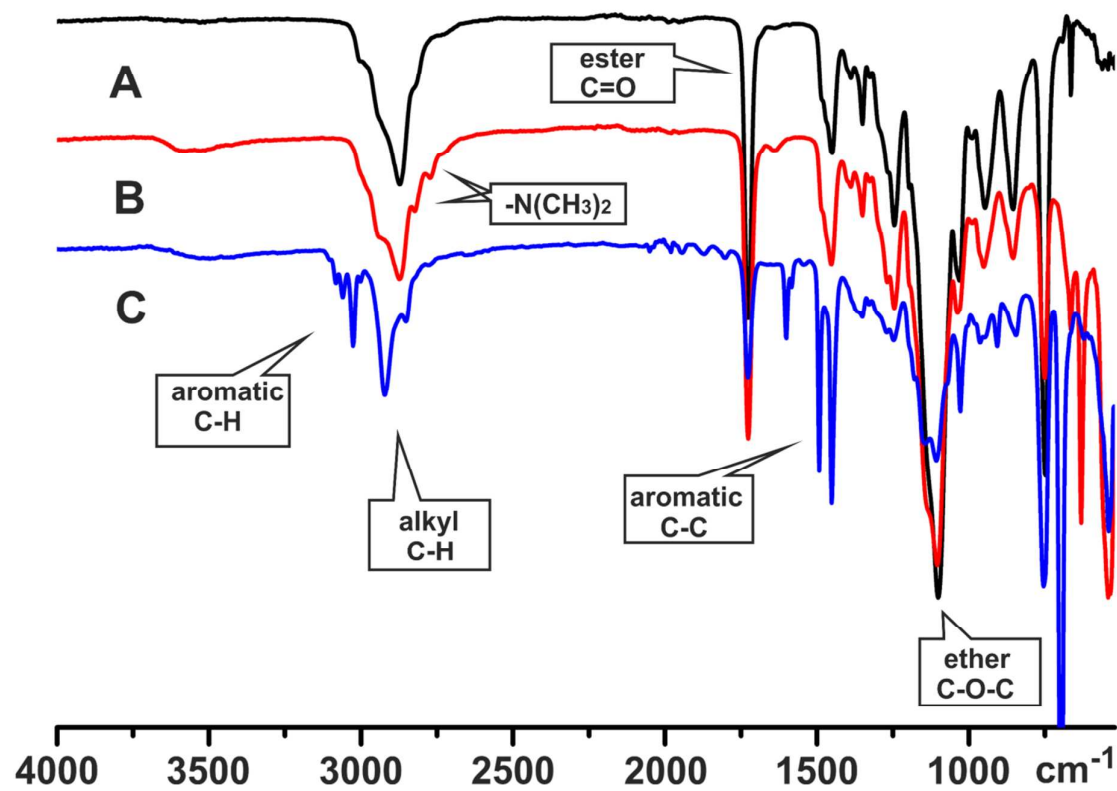
POEGMA-*b*-DMAEMA-*b*-PST triblock copolymer nanoparticles with various morphologies were successfully patterned with gold nanoparticles by interacting the triblock copolymer aggregates in aqueous dispersion with chloroauric acid for one day followed by adding sodium borohydride to achieve reduction. In this procedure, 0.1 mL, 0.02M  $\text{HAu(III)Cl}_4 \cdot 3\text{H}_2\text{O}$  was mixed with 1 mL ( $1.0 \times 10^{-3} \text{ mmol}$  tertiary amine containing) triblock polymer dispersion

and interacted for one day under continuous stirring. The mixture was then reduced by quick addition of 0.1 mL NaBH<sub>4</sub> (0.04 M). The triblock copolymer/Au NP hybrids were purified by dialysis against deionized water for 24 h at room temperature and then characterized by TEM, DLS, XRD and TGA.

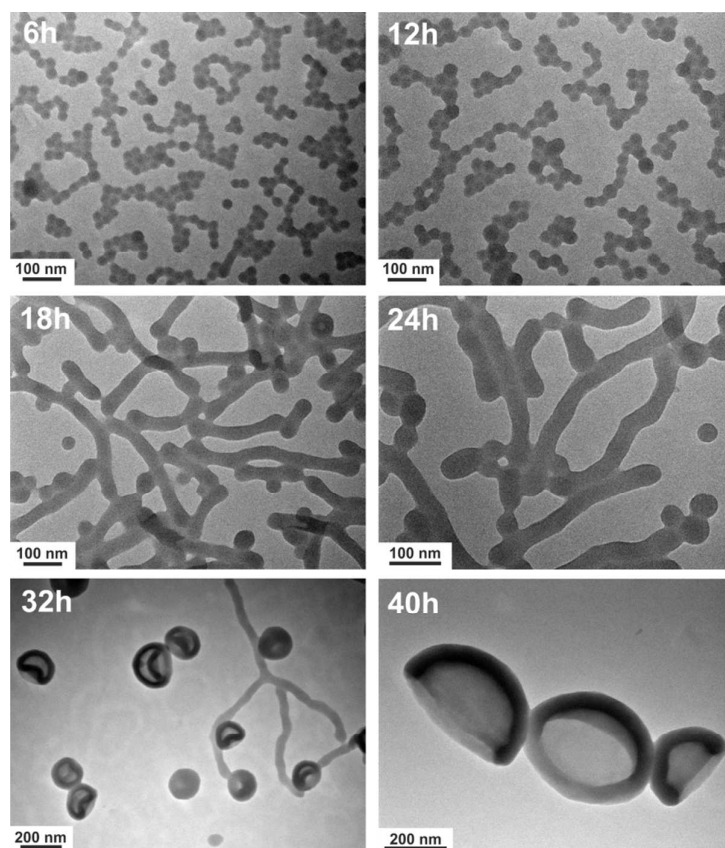
**Table S1.** Summary of gold nanoparticle formation recipe in triblock copolymer dispersion.

Time (h)	Yield (%) <sup>a</sup>	Polymer/water (mg/mL)	Amine (mmol) <sup>b</sup>	Gold (mmol)	NaBH <sub>4</sub> (mmol)	AuNP <sup>c</sup> (%)	AuNP <sup>d</sup> theo. (%)
[Au] : [Amine] = 2 : 1 (mol/mol)							
6	90	3.1	1.0×10 <sup>-3</sup>	2.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	12.0	15.5
12	90	3.6	1.0×10 <sup>-3</sup>	2.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	11.5	13.2
18	85	5.3	1.0×10 <sup>-3</sup>	2.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	11.0	11.3
24	90	5.8	1.0×10 <sup>-3</sup>	2.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	10.0	11.1
32	90	9.5	1.0×10 <sup>-3</sup>	2.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	9.0	8.7
40	90	9.6	1.0×10 <sup>-3</sup>	2.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	9.0	8.5
[Au] : [Amine] = 4 : 1 (mol/mol)							
6	90	3.1	1.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	8.0×10 <sup>-3</sup>	22.0	31.0
12	90	3.6	1.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	8.0×10 <sup>-3</sup>	21.0	26.4
18	85	5.3	1.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	8.0×10 <sup>-3</sup>	20.0	22.6
24	90	5.8	1.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	8.0×10 <sup>-3</sup>	19.0	22.2
32	90	9.5	1.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	8.0×10 <sup>-3</sup>	19.0	19.4
40	90	9.6	1.0×10 <sup>-3</sup>	4.0×10 <sup>-3</sup>	8.0×10 <sup>-3</sup>	18.0	17.0

Note: a-yield the polymer synthesis after purification at different polymerization times, b- Tertiary amine content of the samples were calculated using NMR results; c- AuNP content determined by thermal gravimetric analysis after purification, d- theoretical AuNPs content calculated using the following equation:  $\text{AuNPs (\%)} = \left[ \frac{\text{DP}_n^{\text{PDMAEMA}} \times ([\text{Au}]/[\text{Amine}])}{M_w^{\text{Au}}} \right] \times M_n^{\text{POEGMA-}b\text{-DMAEMA-}b\text{-PST}}$ , with  $\text{DP}_n^{\text{PDMAEMA}}$  and  $M_n^{\text{POEGMA-}b\text{-DMAEMA-}b\text{-PST}}$  correspond to number of PDMAEMA units per chain and molecular weight of triblock copolymers determined by NMR.

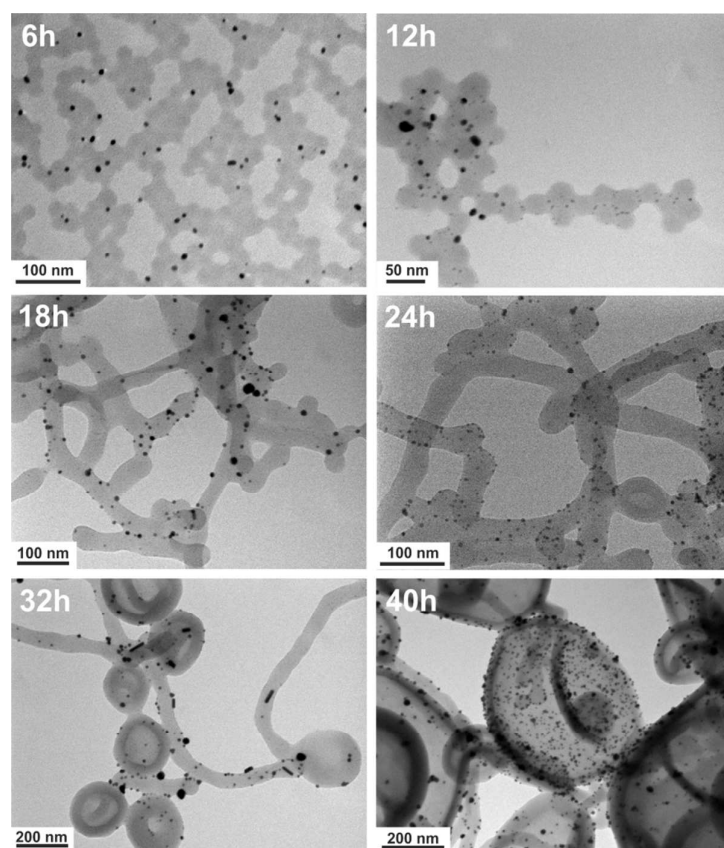


**Figure S1.** FTIR spectra of POEGMA (black line) homopolymer, POEGMA-*b*-PDMAEMA copolymer (red line), and POEGMA-*b*-PDMAEMA-*b*-PST copolymer (blue line).

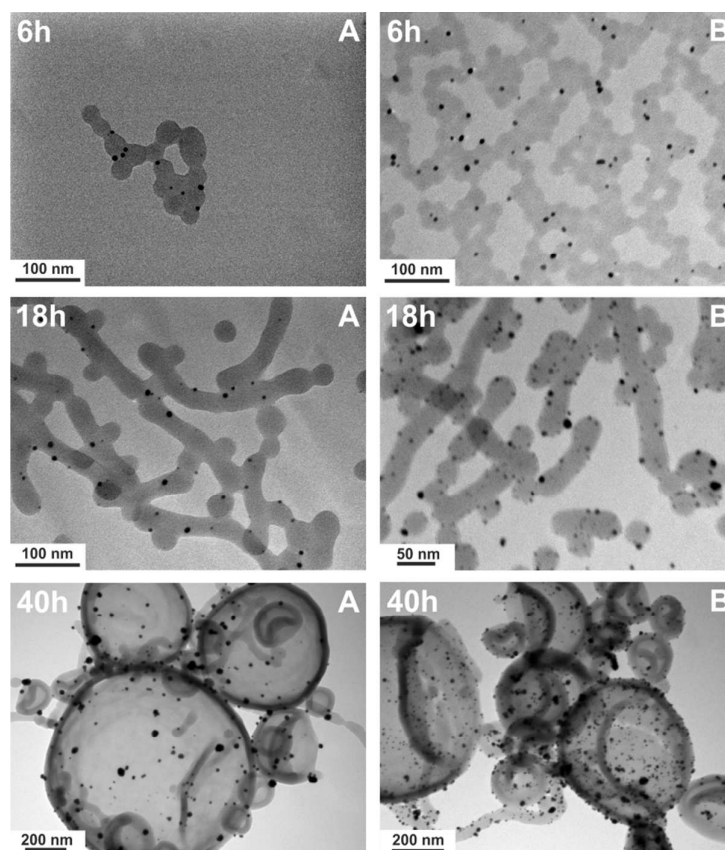


**Figure S2.** Supplementary TEM micrographs of the triblock copolymer nano-aggregates in aqueous dispersion.

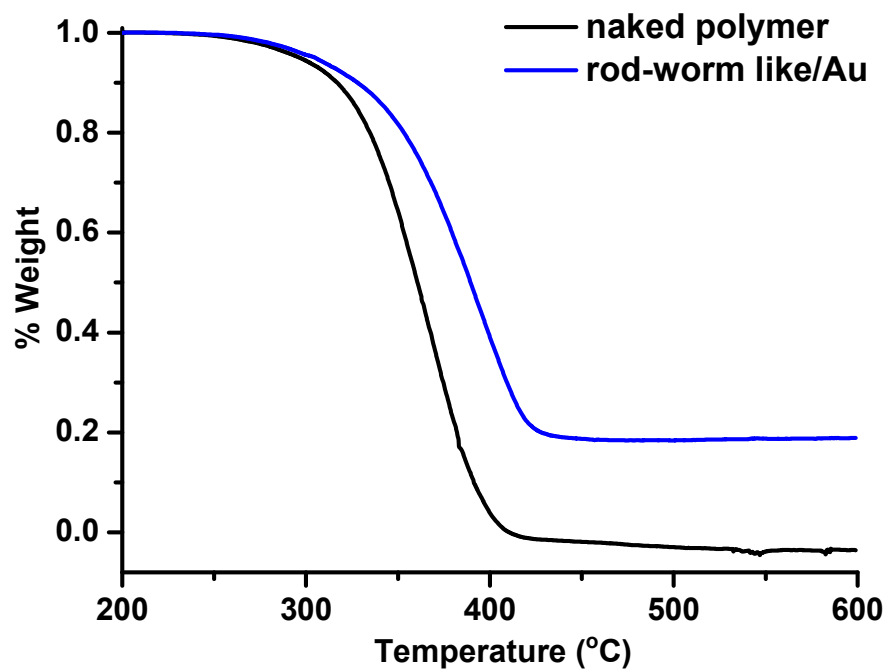




**Figure S3.** Supplementary TEM micrographs of the gold decorated triblock copolymer nanoaggregates in aqueous dispersion obtained using a molar feed ratio, i.e. 4:1 of Au: tertiary amine.



**Figure S4.** Supplementary TEM micrographs of the gold decorated triblock copolymer nanoaggregates in aqueous dispersion obtained using a molar feed ratio (A) 2:1 of Au: tertiary amine and (B) 4:1 of Au: tertiary amine for different morphologies.



**Figure S5.** Typical TGA thermograms of naked POEGMA-*b*-PDMAEMA-*b*-PST triblock copolymer (40h sample) (black) and gold tethered samples with different morphologies