

Supplementary information

Materials

Poly(ethylene glycol) methyl ether acrylate (PEGA, average $M_n = 480 \text{ g mol}^{-1}$), *n*-butyl acrylate (*n*-BA, >99%), acrylic acid (AA, >98%), bromo-propionic acid (>99%), 1-butanethiol (99%), propargyl amine, propargyl alcohol, 3-(trimethylsilyl)propargyl alcohol, *N,N'*-dicyclohexylcarbodiimide (DCC), 4-dimethylaminopyridine (DMAP), benzotriazol-1-yl-oxytripyrrolidinophosphonium hexafluorophosphate (PyBOP), *N,N'*-diisopropylethylamine (DIPEA), CuSO₄ and ascorbic acid, carbon disulphide (>99%) and methoxypolyethylene glycol azide (PEG-2k-N₃, $M_n = 2000 \text{ g mol}^{-1}$) were obtained from Sigma-Aldrich. All monomers above were passed through basic aluminium oxide to remove inhibitor before use. Deuterium oxide (99.9% D atom) and chloroform-*d*₃ (99.8% D atom), were obtained from Sigma Aldrich and used for ¹H NMR spectroscopy. Thermal initiator 4,4'-azobis(4-cyanovaleric acid) (ACVA, >98%, Aldrich). RAFT agent, 2-(((butylthio)carbonothioyl)thio)propanoic acid (PABTC) was synthesised as previously described.⁴⁵ Macro-RAFT agent COOH-(P(PEGA)₈-*b*-P(*n*-BA)₈) (P[(PEGA)₈-*b*-(*n*-BA)₈]) was synthesised as previously described.³⁸ Solvents were acquired from commercial sources.

Instrumentation and Analysis

NMR spectroscopy

¹H NMR and ¹³C NMR spectra were recorded on a Bruker DPX-250, DPX-300, or DPX-400 spectrometer using deuterated solvent (Materials section).

Size exclusion chromatography

An Agilent Infinity II MDS instrument equipped with differential refractive index (DRI), viscometry (VS), dual angle light scatter (LS) and multiple wavelength UV detectors was used for SEC analysis. The system was fitted with 2 x PLgel Mixed C columns (300 x 7.5 mm) and a PLgel 5 μm guard column. The eluent used was THF with 2 % TEA (triethylamine), and 0.01 % BHT (butylated hydroxytoluene) additives. Samples were run at 1 ml min⁻¹ at 30°C. Poly(methyl methacrylate) and polystyrene standards (Agilent EasyVials) were used for calibration. Analyte samples were filtered through a GVHP membrane with 0.22 μm pore size before injection. Respectively, experimental molar mass ($M_{n,SEC}$) and dispersity (\mathcal{D}) values of synthesized polymers were determined by conventional calibration using Agilent GPC/SEC software.

Theoretical molar mass calculation

$$M_{n,th} = \frac{[M]_0 p M_M}{[CTA]_0} + M_{CTA}$$

Equation 1 Calculation of theoretical number average molar mass ($M_{n,th}$) where $[M]_0$ and $[CTA]_0$ are the initial concentrations (in mol dm⁻³) of monomer and chain transfer agent respectively. p is the monomer conversion as determined by ¹H NMR spectroscopy. M_M and M_{CTA} are the molar masses (g mol⁻¹) of the monomer and chain transfer agent respectively.

Dynamic light scattering and ζ -potential

Size and ζ -potential measurements were carried out using a Malvern Zetasizer Nano-ZS at 25°C with a 4 mW He-Ne 633 nm laser at a scattering angle of 173° (back scattering). Measurements were taken assuming the refractive index of: polyethylene glycol for macro-RAFT agents, and the refractive index of the core material (e.g poly(*n*-butyl acrylate)) for latex particles. DLS samples of latex particles were prepared by diluting by 1000 with 1 mL of water and measured unfiltered in 1.5 mL polystyrene cuvettes for measuring size and a Malvern DTS-1070 zeta cuvette for ζ -potential. Samples were incubated for 60 seconds at 25°C prior to measurement. Measurements were repeated three times with automatic attenuation selection and measurement position. Results were analysed using Malvern DTS 6.20 software. PDI values were calculated using the following equation. Measurements of ζ -potential were modelled with the Smoluchowski theory.

$$PDI = \frac{\sigma^2}{d_h^2}$$

Equation 2 Calculation of nanoparticle polydispersity (PDI) from standard deviation (σ), and diameter (d).

Synthesis procedures

Synthesis of Alkyne-PAmBTC⁴⁶

A stirred solution of PyBOP (1.64 g, 3.15 mmol, 1 eq) and DIPEA (0.97 g, 6.74 mmol, 2.4 eq) in DCM (5 mL) was added to a separate solution of PABTC (0.75 g, 3.15 mmol, 1 eq; dissolved in 5 mL DCM) in a 25 mL round bottomed flask fitted with a magnetic stirrer, turning the solution deep red. Propargyl amine (173 mg, 3.15 mmol, 1 eq) was added subsequently dropwise to the PABTC solution. After all of the amine had been added, the solution returned to its original yellow colour and was left to stir at room temperature overnight. The reaction was monitored with TLC using a 50/50 hexane/ethyl acetate eluent. The reaction mixture was then concentrated under reduced pressure, and purified *via* automatic column chromatography using a gradient eluent (10% -50% ethyl acetate in hexane, over 30 min) yielding **Alkyne-PAmBTC** (589.6 mg, 68% yield) as a pale yellow solid. ¹H NMR (500 MHz, CDCl₃) δ 6.57 (br, J = 45.1 Hz, 1H, NH), 4.77 (q, J = 7.4 Hz, 1H, CH₃-CH), 4.05 (qdd, J = 17.6, 5.3, 2.5 Hz, 2H, NH-CH₂), 3.41 (t, 2H, S-CH₂), 2.25 (t, J = 2.4 Hz, 1H, C \equiv CH), 1.72 (p, 2H, S-CH₂-CH₂), 1.61 (d, J = 7.3 Hz, 3H, CH-CH₃), 1.46 (h, 2H, CH₂-CH₃), 0.97 (t, J = 7.4 Hz, 3H, CH₂-CH₃). ¹³C NMR (126 MHz, CDCl₃) δ 224.04 (C=S), 170.16 (C=O), 79.05 (C \equiv CH), 71.82 (C \equiv CH), 47.48 (S-CH), 37.35 (S-CH₂), 29.86 (NH-CH₂), 29.55 (S-CH₂-CH₂), 22.06 (CH₃-CH₂), 16.06 (CH-CH₃), 13.59 (CH₂-CH₃). FTIR ν cm⁻¹: 3281(s, C-H alkyne), 2957 + 2927 (m, N-H amide), 2868 (m, C-H alkane), 1656 (s, C=O amide). ESI-MS m/z : 276.0 [M+H]⁺.

Synthesis of TMS-Alkyne-PEsBTC⁴⁷

DCC (0.714 g, 3.46 mmol, 1.1 eq) and DMAP (38 mg, 0.315 mmol, 0.1 eq) was added to a stirred solution of PABTC (0.750 g, 3.15 mmol, 1 eq) in DCM (20 mL), in a 50 mL round bottomed flask turning the solution deep red. 3-(Trimethylsilyl)propargyl alcohol (0.444 g, 3.46 mmol, 1.1 eq) was added to the activated PABTC solution and stirred overnight at room temperature. The mixture was washed with 50 mL water (3x) and 1 M HCl (3 x),

the organic phase dried over sodium sulfate, and purified *via* flash column chromatography using a gradient eluent (0% - 20% ethyl acetate in hexane over 30 min), yielding **TMS-Alkyne-PEsBTC** as a bright yellow oil (0.53 g, 48% yield). ¹H NMR (500 MHz, CDCl₃) δ 4.84 (q, *J* = 7.4 Hz, 1H, S-CH), 4.73 (d, *J* = 3.1 Hz, 2H, O-CH₂), 3.36 (t, *J* = 7.4 Hz, 2H, S-CH₂), 1.68 (quint, 2H, S-CH₂-CH₂), 1.62 (d, *J* = 7.4 Hz, 3H, CH-CH₃), 1.43 (sext, 2H, CH₃-CH₂) 0.93 (t, *J* = 7.4 Hz, 3H, CH₂-CH₃), 0.18 (s, 9H, Si-CH₃). ¹³C NMR (126 MHz, CDCl₃) δ 222.07 (C=S), 170.81 (C=O), 98.64 (C≡C-Si), 93.10 (C≡C-Si), 54.29 (O-CH₂), 48.10 (S-CH), 37.30 (S-CH₂), 30.24 (S-CH₂-CH₂), 22.39 (CH₃-CH₂), 17.04 (CH-CH₃), 13.91 (CH₂-CH₃), -0.01 (Si-CH₃). FTIR ν cm⁻¹: 2958 + 2931 (m, N-H amide), 2872 (m, C-H alkane), 2186 (w, C≡C alkyne), 1741 (s, C=O ester). ESI-MS *m/z*: 349.6 [M+H]⁺.

Synthesis of Alkyne-PEsBTC⁴³

EDC (3.86 g, 20.1 mmol, 1.2 eq) and DMAP (0.240 g, 2.1 mmol, 0.12 eq) was added to a stirred solution of PABTC (4 g, 16.7 mmol, 1 eq) in DCM (20 mL), in a 50 mL round bottomed flask turning the solution deep red. Propargyl alcohol (1.13 g, 20.1 mmol, 1.2 eq) was added to the activated PABTC solution and stirred overnight at room temperature. The mixture was washed with 50 mL water (3x) and 1 M HCl (3 x), the organic phase dried over sodium sulfate, and purified *via* flash column chromatography using a gradient eluent (0% - 50% ethyl acetate in hexane over 30 min), yielding **Alkyne-PEsBTC** (4.20 g, 91 % yield) as an orange oil. ¹H NMR (500 MHz, CDCl₃) δ 4.85 (q, *J* = 7.4 Hz, 1H, S-CH), 4.73 (t, *J* = 2.8 Hz, 2H, O-CH₂), 3.36 (t, *J* = 7.4 Hz, 2H, S-CH₂), 2.50 (t, *J* = 2.4 Hz, 1H, C≡C-H), 1.72 – 1.65 (m, 2H, S-CH₂-CH₂), 1.62 (d, *J* = 7.4 Hz, 3H, S-CH-CH₃), 1.47 – 1.38 (m, 2H, CH₃-CH₂), 0.93 (t, *J* = 7.4 Hz, 3H, CH₂-CH₃). ¹³C NMR (126 MHz, CDCl₃) δ 221.75 (C=S), 170.51 (C=O), 75.43 (C≡CH), 53.18 (O-CH₂), 47.62 (S-CH), 37.03 (S-CH₂), 29.91 (S-CH₂-CH₂), 22.07 (CH₃-CH₂), 16.70 (CH-CH₃), 13.60 (CH₂-CH₃). FTIR ν cm⁻¹: 3290(s, C-H alkyne) 2957 + 2930 (m, N-H amide), 2871 (m, C-H alkane), 2131 (w, C≡C alkyne), 1738 (s, C=O ester). ESI-MS *m/z*: 277.0 [M+H]⁺.

Preparation of macro-RAFT agents

Macro-RAFT agents **TMS-Alkyne-O-P[(PEGA)₁₂-*b*-(*n*-BA)₁₂]**, **TMS-Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-(*n*-BA)₁₅]** and **Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-(*n*-BA)₁₅]** were prepared using the following RAFT polymerisation procedure. As an example, synthesis of **Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-(*n*-BA)₁₅]** is described: **Alkyne-PEsBTC** (16 mg, 57.8 μmol), PEGA (0.416 g, 0.867 mmol), acrylic acid (0.0124 g, 0.173 mmol), ACVA (0.81 mg, 2.9 μmol) and 1,4-dioxane (542 μL) were added to a 7.5 mL vial fitted with a stirrer bar and sealed with a rubber septum. The solution was deoxygenated with dinitrogen and was immersed in an oil bath preheated to 70 °C for 5 h. The resulting polymer **Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-(*n*-BA)₁₅]** was isolated *via* precipitation in diethyl ether/hexane (20/80, v/v).

Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-(*n*-BA)₁₅] (0.5g, 84.5 μmol) was dissolved in 1,4-dioxane (3.18 mL) and *n*-BA (0.195 g, 1.5 mmol), ACVA (1.18 mg, 4.2 μmol) was added and heated to 70 °C for 5 h. **Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-(*n*-BA)₁₅]** was purified by evaporating 1,4-dioxane and residual *n*-BA under reduced pressure at 40 °C. Full details for polymerisation conditions can be found in the supplementary information (**Table S4**).

Preparation of micelle blends⁴⁸

Micelle blends of **TMS-Alkyne-O-P[(PEGA)₁₂-*b*-(*n*-BA)₁₂]** and **COOH-[P(PEGA)₈-*b*-P(*n*-BA)₈]**, were prepared by dissolving both di-block macro-RAFT agents in THF (10 mg mL⁻¹), which were mixed in separate test tubes in 5 molar ratios of TMS/COOH (100/0, 50/50, 25/75, 10/90, 0/100). The mixtures were then dried thoroughly at 70°C, were rehydrated in 1 mL of water, and were left to stir overnight. RAFT emulsion polymerisations were then performed using these micelle blends using conditions described below.

RAFT emulsion polymerisation

RAFT emulsion polymerisations were performed using conditions described previously.³⁸ As an example, **NP7**, was prepared as follows. **Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃]-P(*n*-BA)₁₅** (49 mg, 7.125 μmol) was dissolved in 1.93 mL of water in a 7.5 mL vial fitted with a stirrer bar. ACVA pre-dissolved in water (neutralised with 2 eq NaOH; 5 mg mL⁻¹, 0.363 mL) was added to the vial, and sealed with a rubber septum. The solution was deoxygenated with dinitrogen, then deoxygenated *n*-BA (204 μL, 1.425 mmol) was added *via* syringe. The vial was immersed in an oil bath heated to 70°C for 3 h. Monomer conversion was determined using gravimetric techniques.

Deprotection of TMS protected nanoparticles

The TMS protected alkyne functional nanoparticles (1 eq alkyne), were treated with KF (10 eq) and stirred overnight. The nanoparticle suspension was then dialysed three times in D₂O (30 mL) using a 30,000 Da MWCO centrifugation dialysis tube. The nanoparticle suspension was then analysed directly with ¹H NMR spectroscopy.

Deprotection of TMS protecting group using TBAF or KF

TMS-Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃] (20 mg, 3.38 μmol, 1 eq) was dissolved in water (1 mL) and treated with either TBAF (1M in THF; 33.8 μmol, 10 eq) or KF (33.8 μmol, 10 eq). The mixtures were stirred overnight, then dialysed (3500 Da MWCO) against pure water for 24 h. Both solutions were then freeze-dried and the product extracted with CDCl₃, then analysed with ¹H NMR spectroscopy directly.

Synthesis of Fluorescein-N₃

N-methyl morpholine (NMM; 19 mg, 0.19 mmol, 3 eq) was added to solution of **Fluorescein-COOH** (30 mg, 63.4 μmol, 1 eq) dissolved in 1 mL DMSO and stirred for 5 min. Azidopropanamine (6.3 mg, 63.4 μmol, 1.0 eq) was added directly and stirred overnight. The reaction mixture was analysed *via* HPLC showing complete conversion into **Fluorescein-N₃**. This was then precipitated into methyltertbutyl ether (5 mL), collected *via* centrifugation, and dried under reduced pressure yielding **Fluorescein-N₃** (5 mg, 10.5 mmol, 16%) as an orange powder. ESI-MS *m/z*: 457.1 [M-H]⁻.

CuAAC reactions on P[(PEGA)₁₂-*co*-(AA)₃]

Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃] (25 mg, 4.67 μmol, 1 eq), CuSO₄ (1.75 mg, 7.00 μmol, 1.5 eq) and either **Fluorescein-N₃** (2.13 mg, 4.7 μmol, 1. eq) or **PEG-2k-N₃** (11.2 mg, 5.6 μmol, 1.2 eq) were dissolved in 1 mL water, and purged with dinitrogen for 5 min. A 0.5 mL solution of ascorbic acid (3.29 mg, 18.7 μmol, 4 eq) was

then transferred *via* syringe and stirred overnight. The reaction mixture was then lyophilised, the organic components extracted in CHCl₃ and analysed with THF-SEC and ¹H NMR spectroscopy.

CuAAC reactions on alkyne functional nanoparticles

The alkyne functional nanoparticles (100 μL, 0.285 μmol alkyne, 1 eq), CuSO₄ (0.128 mg, 0.513 μmol, 1.8 eq) and **PEG-2k-N₃** (0.855 mg, 0.427 μmol, 1.5 eq) were dissolved in 1 mL water, and purged with dinitrogen for 5 min. A 0.5 mL solution of ascorbic acid (0.2 mg, 1.14 μmol, 4 eq) was then transferred *via* syringe and stirred overnight. CuAAC reactions were performed on the alkyne functional nanoparticles with **Fluorescein-N₃** with identical conditions.

Supplementary data

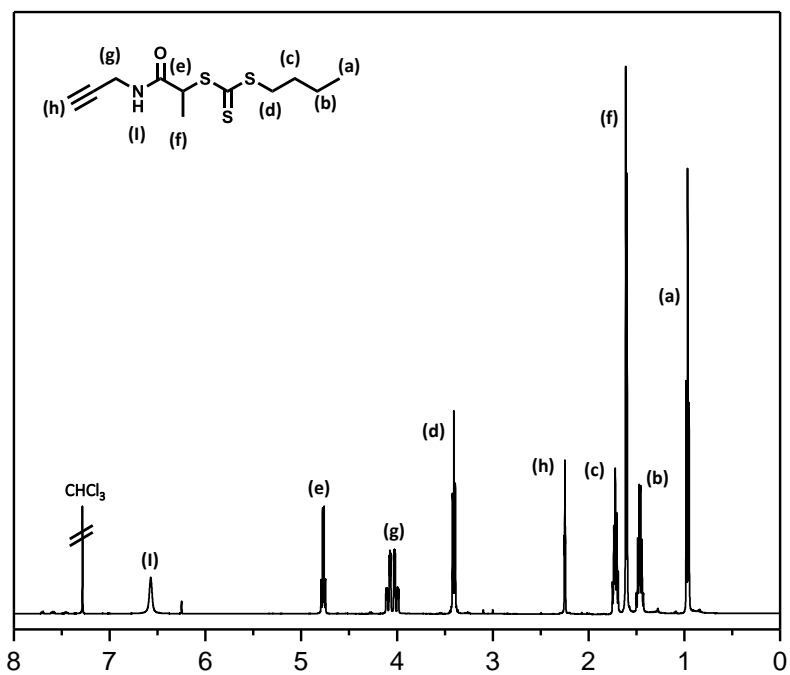


Figure S1 ¹H NMR spectrum of Alkyne-PAmBTC measured at 500 MHz in CDCl₃.

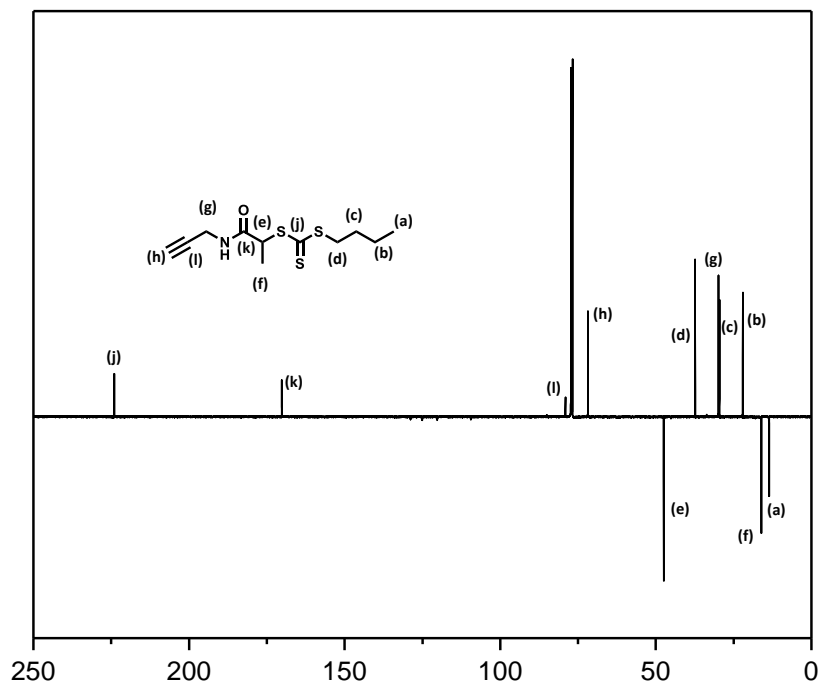


Figure S2 DEPT-135 ¹³C NMR spectrum of Alkyne-PAmBTC measured at 126 MHz in CDCl₃.

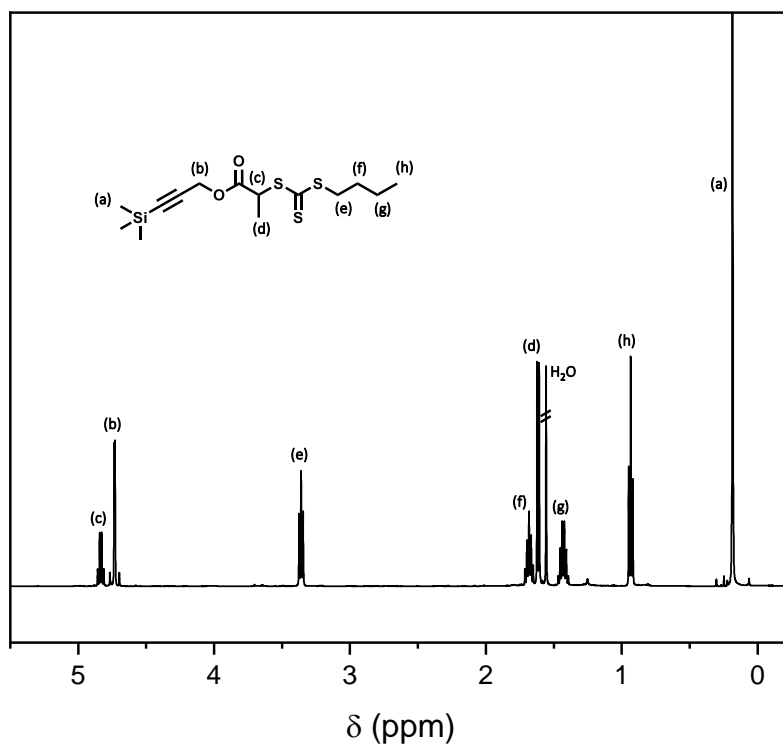


Figure S3 ^1H NMR spectrum of TMS-Alkyne-PEsBTC measured at 500 MHz in CDCl_3 .

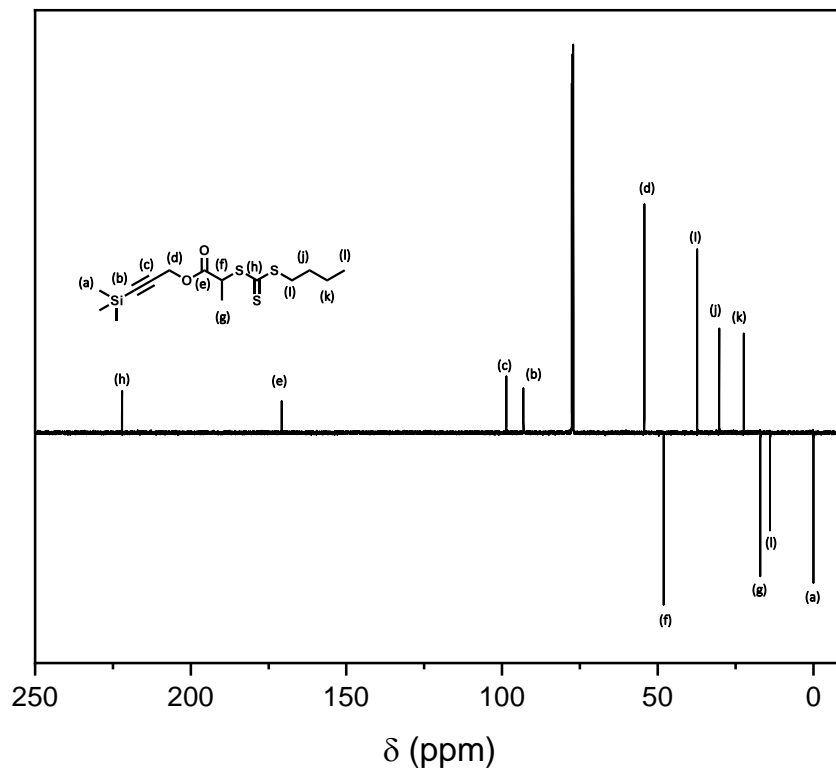


Figure S4 DEPT-135 ^{13}C NMR spectrum of TMS-Alkyne-PEsBTC measured at 126 MHz in CDCl_3 .

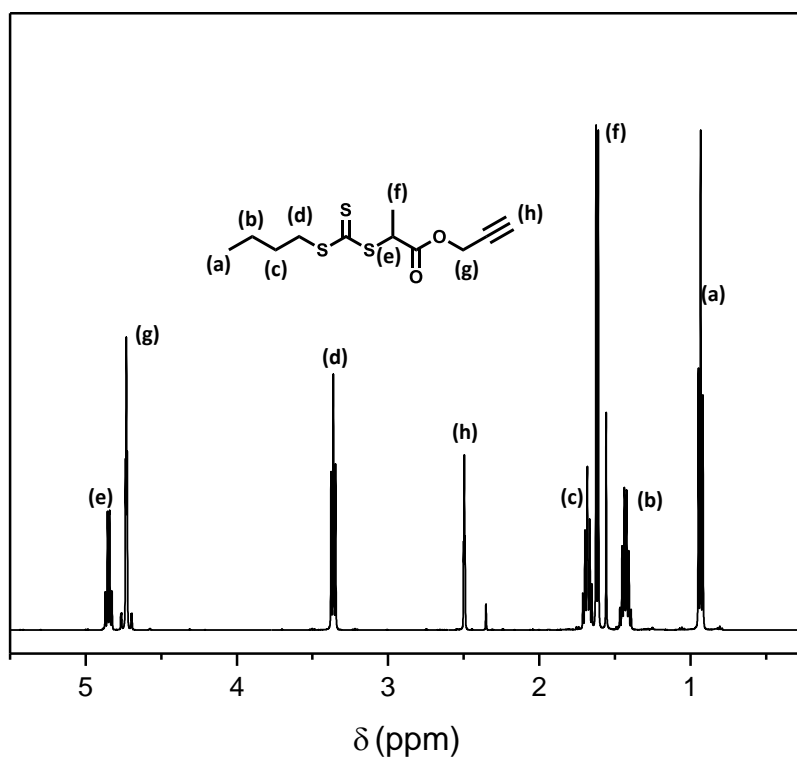


Figure S5 ^1H NMR spectrum of Alkyne-PEsBTC measured at 500 MHz in CDCl_3 .

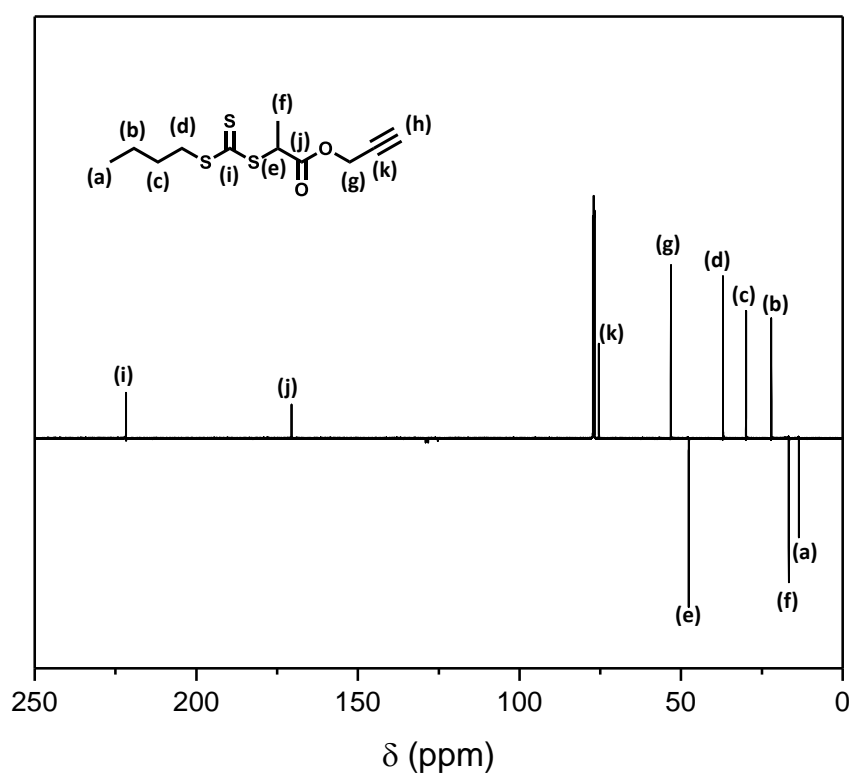


Figure S6 DEPT-135 ^{13}C NMR spectrum of Alkyne-PEsBTC measured at 126 MHz in CDCl_3 .

Table S1 Alkyne-PAmBTC as the CTA.

Time (h)	% conv ^a	$M_{n,th}$ (g mol ⁻¹) ^b	$M_{n,SEC}$ (g mol ⁻¹) ^c	$M_{w,SEC}$ (g mol ⁻¹) ^c	\mathcal{D}^c
0	0	275	-	-	-
0.25	12	1400	4750	5350	1.13
0.5	33	3400	6000	6900	1.15
1	52	5200	7500	8600	1.15
1.5	66	6600	8500	9700	1.15
2	73	7200	9000	10400	1.16
3	82	8100	9500	11100	1.17
4	87	8600	9700	11400	1.17
5	89	8800	9800	11550	1.18

^aDetermined by ¹H NMR spectroscopy. ^bTheoretical molar masses calculated with **Equation 1**. ^cDetermined by THF-SEC and analysed against PMMA standards.

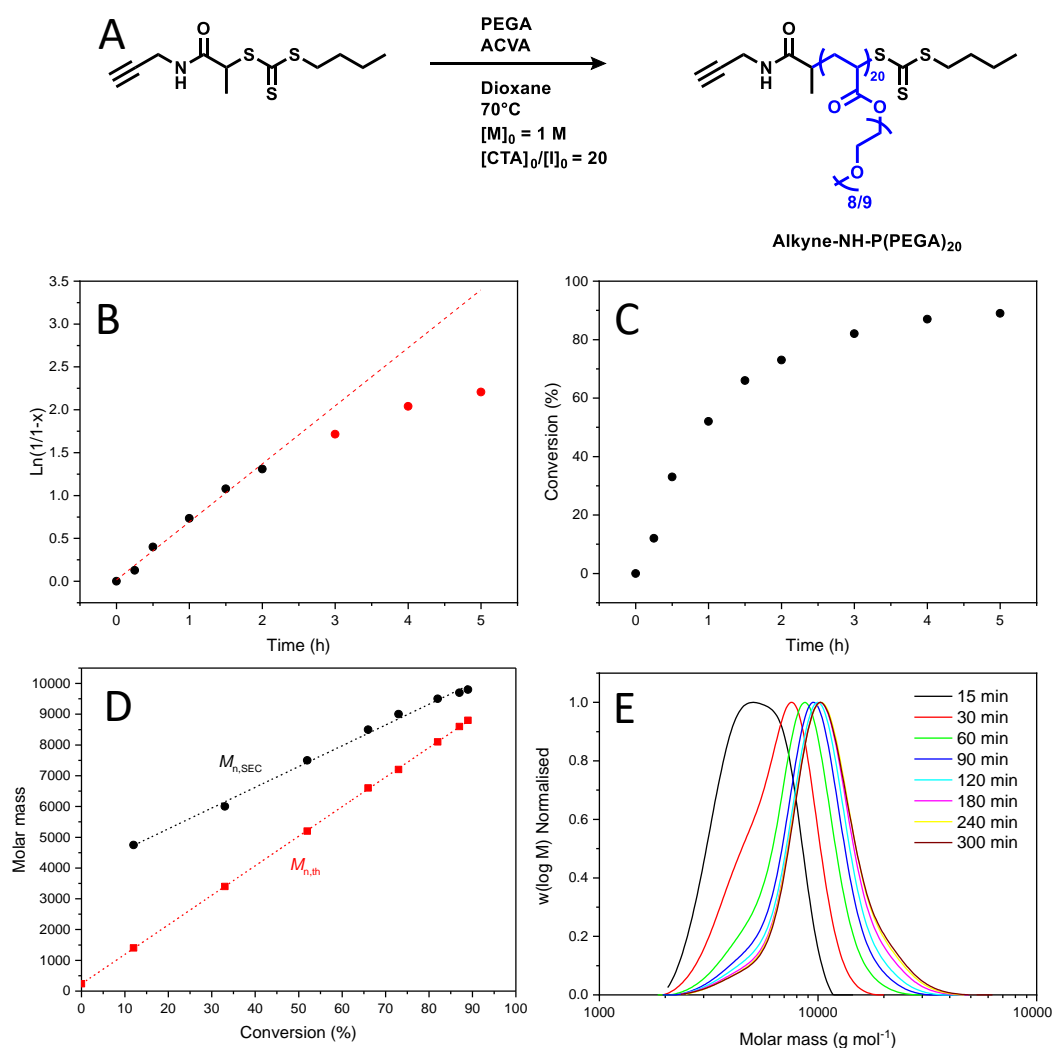


Figure S7 (A) Polymerisation of PEGA mediated by Alkyne-PAmBTC, (B) pseudo first order plot, red points indicate data masked from the linear fit (red dashed line), (C) time vs conversion, (D) conversion vs $M_{n,SEC}$ and linear fit (dashed black line), (E) evolution of SEC chromatograms over time.

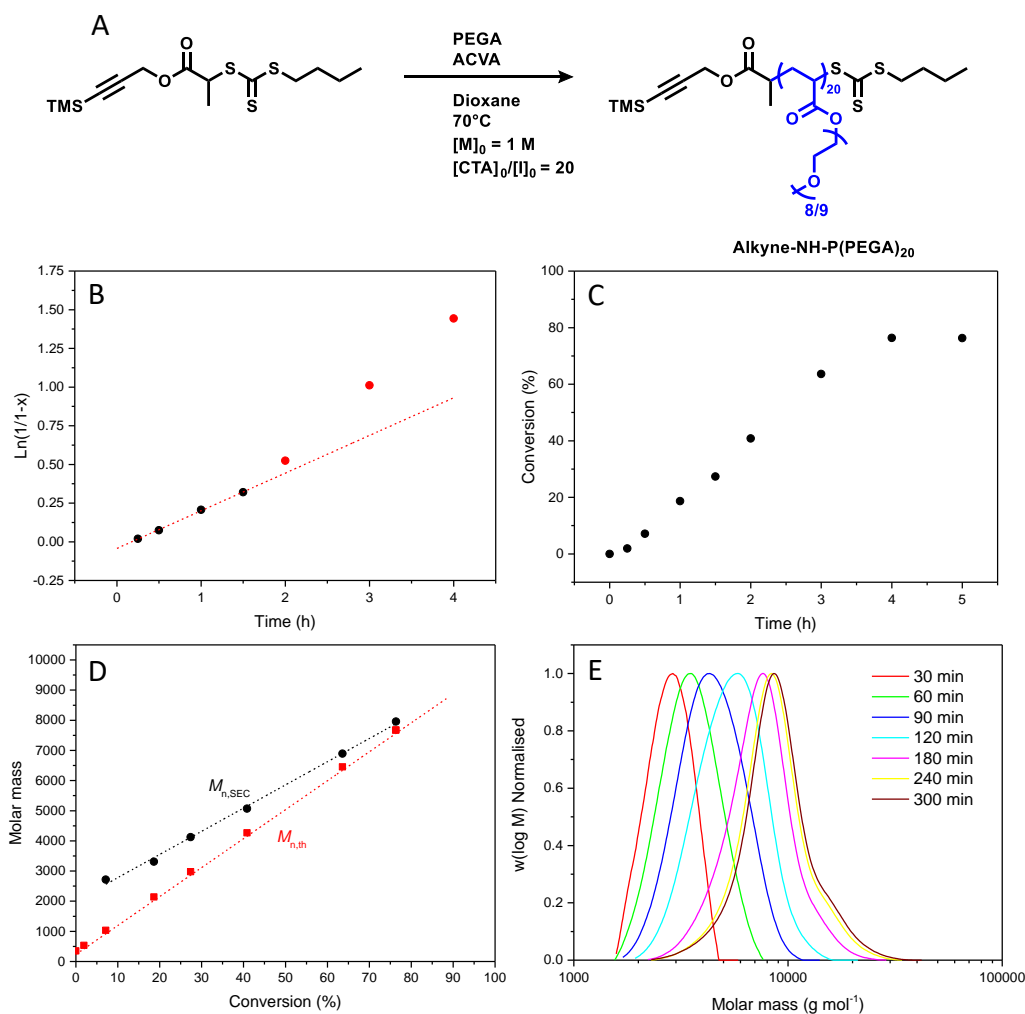


Figure S8 (A) Polymerisation of PEGA mediated with **TMS-Alkyne-PEsBTC**, (B) pseudo first order plot, red points indicate data masked from the linear fit (red dashed line), (C) time vs conversion, (D) conversion vs $M_{n,SEC}$ and linear fit (dashed black line), (E) evolution of SEC chromatograms over time.

Table S2 Data for the polymerisation kinetics of PEGA using with **TMS-Alkyne-PEsBTC** as the CTA.

Time (h)	% conv ^a	$M_{n,th}$ (g mol ⁻¹) ^b	$M_{n,SEC}$ (g mol ⁻¹) ^c	$M_{w,SEC}$ (g mol ⁻¹) ^c	\mathcal{D}^c
0	0	348	-	-	
0.25	2	500	-	-	
0.5	7	1000	2700	2900	1.05
1	18	2100	3300	3600	1.1
1.5	27	3000	4100	4600	1.12
2	41	4300	5100	5700	1.13
3	64	6500	6900	7900	1.14
4	76	7700	8000	9100	1.14
5	76	7700	8300	9500	1.15

^aDetermined by ¹H NMR spectroscopy, ^bTheoretical molar masses calculated with **Equation 1**, ^cDetermined by THF-SEC and analysed against PMMA standards.

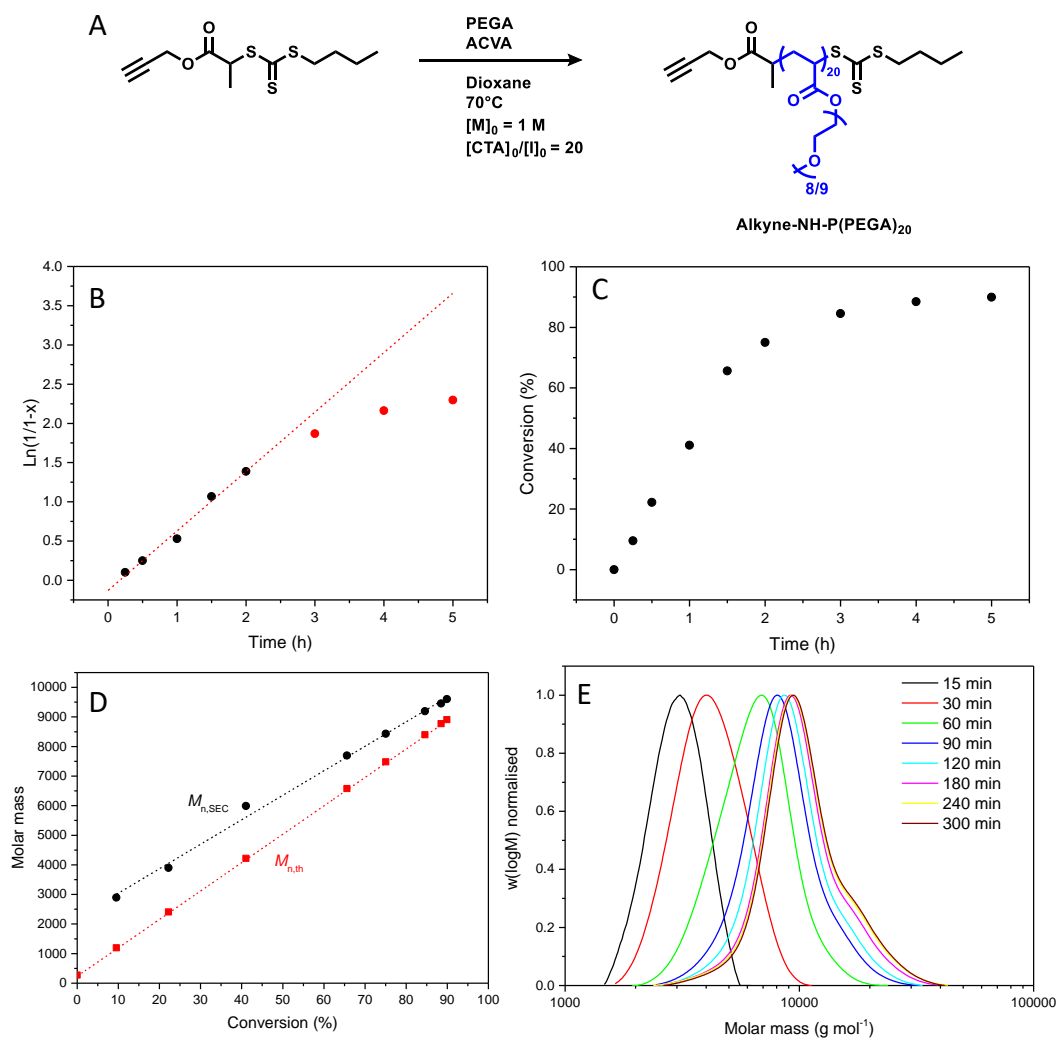


Figure S9 (A) Polymerisation of PEGA mediated with Alkyne-PEsBTC, (B) pseudo first order plot, red points indicate data masked from the linear fit (red dashed line), (C) time vs conversion, (D) conversion vs $M_{n,SEC}$ and linear fit (dashed black line), (E) evolution of SEC chromatograms over time.

Table S3 Data for the polymerisation kinetics of PEGA using with Alkyne-PEsBTC as the CTA.

Time (h)	% conv ^a	$M_{n,th}$ (g mol ⁻¹) ^b	$M_{n,SEC}$ (g mol ⁻¹) ^c	$M_{w,SEC}$ (g mol ⁻¹) ^c	\mathcal{D}^c
0	0	276	-	-	
0.25	10	1200	2900	3100	1.13
0.5	22	2400	3900	4300	1.15
1	41	4200	6000	6800	1.15
1.5	66	6600	7700	8800	1.15
2	75	7500	8400	9700	1.16
3	85	8400	9200	10700	1.17
4	88	8800	9500	11100	1.17
5	89	8900	9600	11200	1.18

^aDetermined by ¹H NMR spectroscopy, ^bTheoretical molar masses calculated with Equation 1, ^cDetermined by THF-SEC and analysed against PMMA standards.

	Monomer	RAFT agent	$\frac{[M]_0}{[RAFT]_0}$	Time (h)	$\frac{[CTA]_0}{[I]_0}$
TMS-Alkyne-O-P[(PEGA) ₁₂]	PEGA	TMS-Alkyne-PEsBTC	15	5	20
TMS-Alkyne-O-P[(PEGA) ₁₂ - <i>b</i> -(<i>n</i> -BA) ₁₂]	<i>n</i> -BA	TMS-Alkyne-O-P[(PEGA) ₁₂]	14	5	20
TMS-Alkyne-O-P[(PEGA) ₁₂ - <i>co</i> -(AA) ₃]	PEGA/AA	TMS-Alkyne-PEsBTC	15+3	5	20
TMS-Alkyne-O-P[(PEGA) ₁₂ - <i>co</i> -(AA) ₃]-(<i>n</i> -BA) ₁₅	<i>n</i> -BA	TMS-Alkyne-O-P[(PEGA) ₁₂ - <i>co</i> -(AA) ₃]	15	5	10
Alkyne-O-P[(PEGA) ₁₂ - <i>co</i> -(AA) ₃]	PEGA/AA	Alkyne-PEsBTC	15+3	5	20
Alkyne-O-P[(PEGA) ₁₂ - <i>co</i> -(AA) ₃]-(<i>n</i> -BA) ₁₅	<i>n</i> -BA	Alkyne-O-P[(PEGA) ₁₂ - <i>co</i> -(AA) ₃]	15	5	20

Table S4 Polymerisation conditions for macro-RAFT agents synthesised
 All reactions were performed at $[M] = 1M$, at $70^\circ C$ using ACVA as thermal initiator and 1,4-dioxane as the solvent.

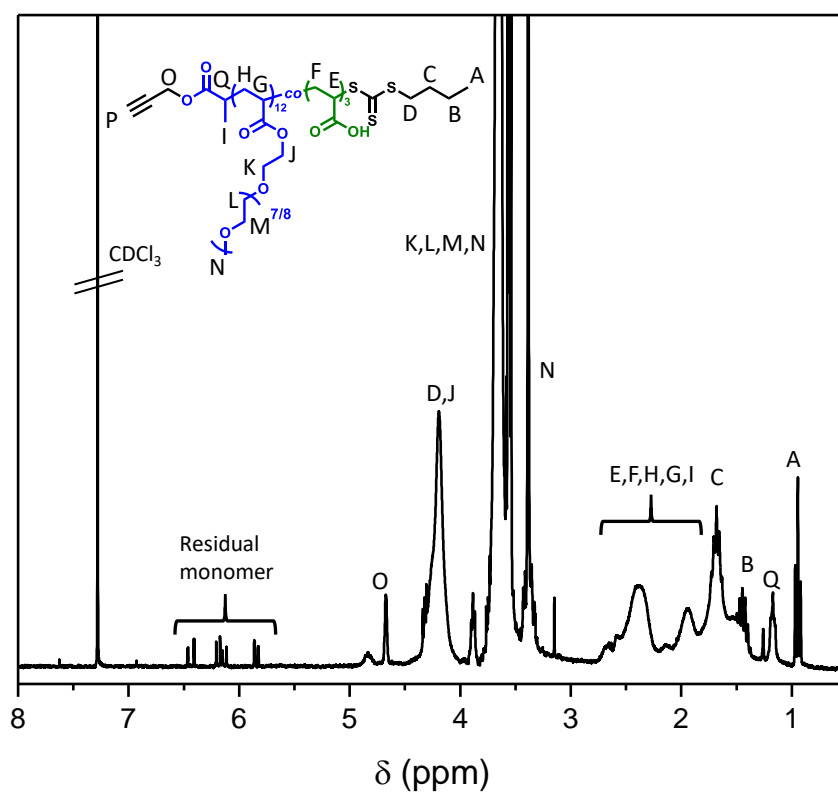


Figure S10 ¹H NMR spectrum of Alkyne-O-P[(PEGA)₁₂-*co*-(AA)₃] in CDCl₃.

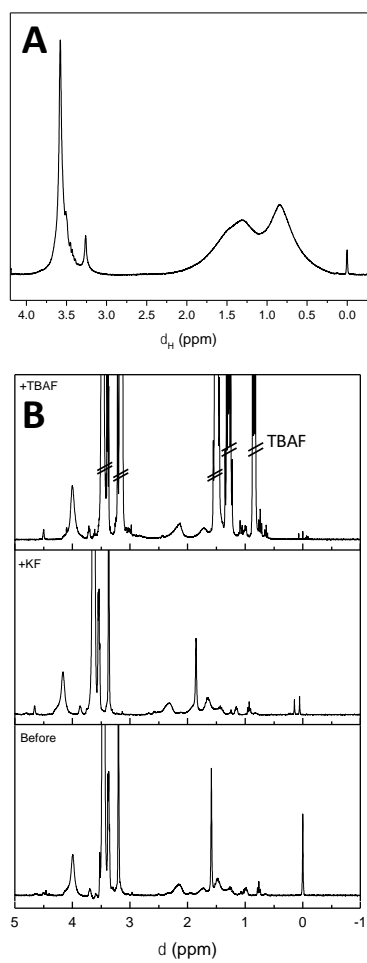


Figure S11 ^1H NMR spectra of (A) TMS protected nanoparticles after treatment with 10 eq KF, and (B) TMS-Alkyne-O-P[(PEGA)₁₂-co-(AA)₃] before (bottom), and after treatment with KF (middle) and TBAF (top).

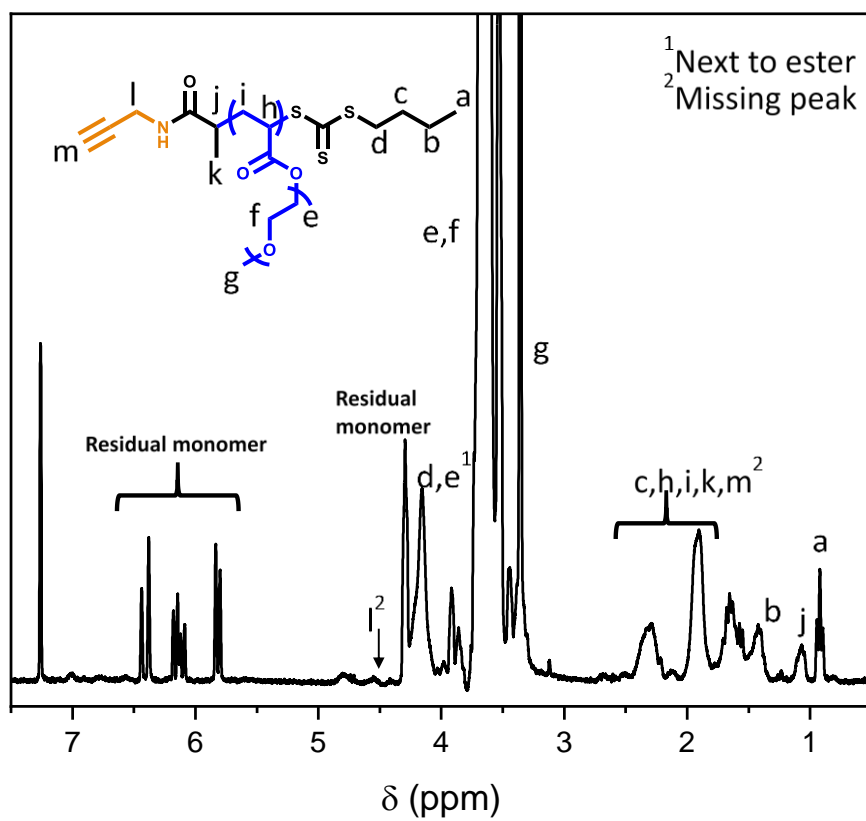


Figure S12 ¹H NMR spectrum of Alkyne-NH-P[(PEGA)12] in CDCl₃. Superscript denotations can be seen in figure.