Electronic supporting information for:

Polyglyoxylamide hydrogels for the traceless stimulus-mediated release of covalently-immobilized drugs

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Synthesis of TEG-N_3-30-PGAm. This polymer was synthesized by the same procedure as TEG-N_3-40-PGAm. It was prepared from: PEtG (200 mg, 2.0 mmol of pendent ester, 1.0 equiv.), 2-azidoethylamine (95 mg 1.1 mmol, 0.56 equiv.), dry 1,4-dioxane (5 mL) and TEG-amine (1120 mg, 5.9 mmol, 5.0 equiv.). A tacky solid was obtained. Yield: 92%. ^{1}H NMR ($CDCl_3$, 400 MHz): δ 8.66 – 7.73 (br s, 0.9H), 5.71 (br s, 1.0H), 3.76 – 3.50 (m, 9.4H), 3.43 (s, 1.4H), 3.37 (s, 2.4H). $^{13}C\{^1H\}$ NMR ($CDCl_3$, 400 MHz): δ 167.2, 96.6, 71.9, 70.5, 69.1, 58.9, 39.3. FT-IR: 3516-3148, 2870, 2099, 1674, 1545 cm^{-1}. SEC (DMF, PMMA): M_n = 56 kg/mol, M_w = 90 kg/mol, $Đ$ = 1.61.

Synthesis of *o*NB-TEG-N_3-20-PGAm. The synthetic procedure was same as ***o*NB-TEG-N_3-30-PGAm**. The copolymer was prepared from: **TEG-N_3-30-PGAm** (100 mg, 0.15 mmol of azide, 1.0 equiv.), alkyne-functionalized *o*-nitrobenzyl alcohol (11.9 mg, 0.051 mmol, 0.33 equiv.), sodium ascorbate (10.1 mg, 0.051 mmol, 0.33 equiv.) and copper sulfate (8.14 mg, 0.051 mmol, 0.33 equiv.) A yellow tacky solid was obtained. Yield: 89%. ^{1}H NMR (DMSO-d_6, 400 MHz): δ 8.60 – 7.79 (m, 1.5H), 5.56 (br s, 1.0H), 4.89 (br s, 0.2H), 4.47 (br s, 0.4H), 3.82-3.38 (m, 8.7H), 3.24 (s, 2.2H). FT-IR: 3533-3115, 2902, 2101, 1624, 1524 cm^{-1}.

Synthesis of Phe-*o*NB-TEG-N_3-20-PGAm. The conjugation of Phe methyl ester to the 20% azide-functionalized copolymer followed the same procedure as for the preparation of **Phe-*o*NB-TEG-N_3-30-PGAm** except that *o*NB-TEG-N_3-20-PGAm (115 mg, 0.052 mmol of *o*NB linker, 1.0 equiv.), CDI (84 mg, 0.52 mmol, 10 equiv.) and Phe methyl ester (26 mg, 0.14 mmol, 3.0 equiv.) were used. Yield: 49% ^{1}H NMR (DMSO-d_6, 400 MHz): δ 8.84 – 7.87 (m, 1.7H), 7.46 –7.02 (m, 0.5H), 5.56 (br s, 1H), 4.88 (br s, 0.2H), 4.63 – 4.25 (m, 0.4H), 3.87 – 2.87 (m, 15H).

Table S1. The formulations of hydrogels containing 4-arm-PEG-alkyne and phenylalanine conjugated PGAm copolymer in 300 μL DMF/water (v/v = 4/1).

15% w/v Hydrogels				
	4-arm-PEG-alkyne	**Phe-*o*NB-TEG-N_3-PGAm**	$CuSO_4$	Sodium ascorbate
Mass (mg)				
20% azide	23	22	2.7	3.3
30% azide	27	17	3.5	4.3

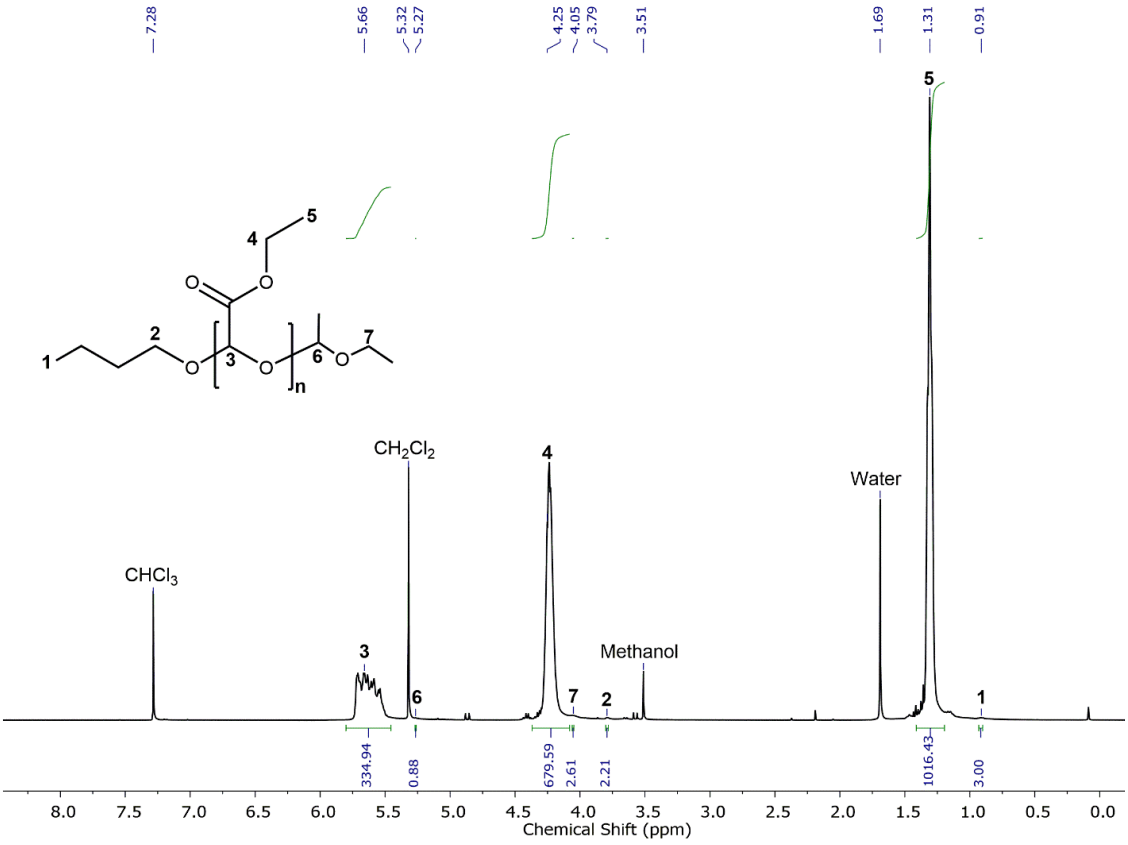

Figure S1. ^{1}H NMR spectrum of 30 kg/mol **PEtG** ($CDCl_3$, 400 MHz). A DP_n of ~335 was determined based on the relative integrals of the methyl CH_3 peak at 0.9 ppm compared to the backbone methine peak at 5.6 ppm.

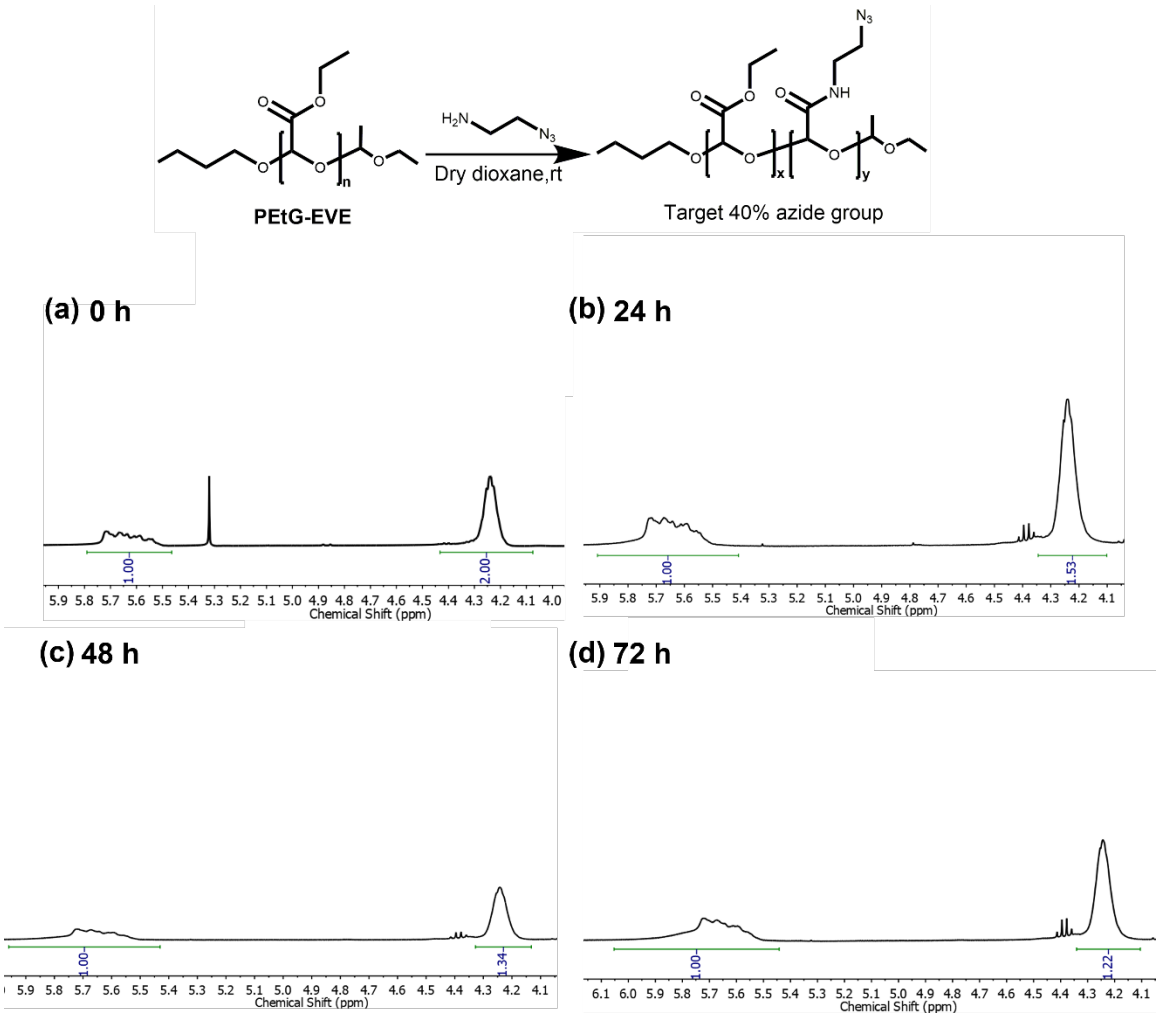

Figure S2. Conversion of 40% of the pendent esters of **PEtG** to pendent 2-azidoethylamides in the synthesis of **TEG-N$_3$-40-PGAm**. The reaction was monitored by ^{1}H NMR spectroscopy. The percent functionalization was determined by comparing the integral of the -CH (methine) peak from the polymer backbone at 5.66 ppm with that of the -CH_2 peak from the pendent ester groups at 4.25 ppm. The reaction was stopped when the integral of -CH_2 peak at 4.25 ppm was ~1.20, indicating ~40% azide functionalization was achieved.

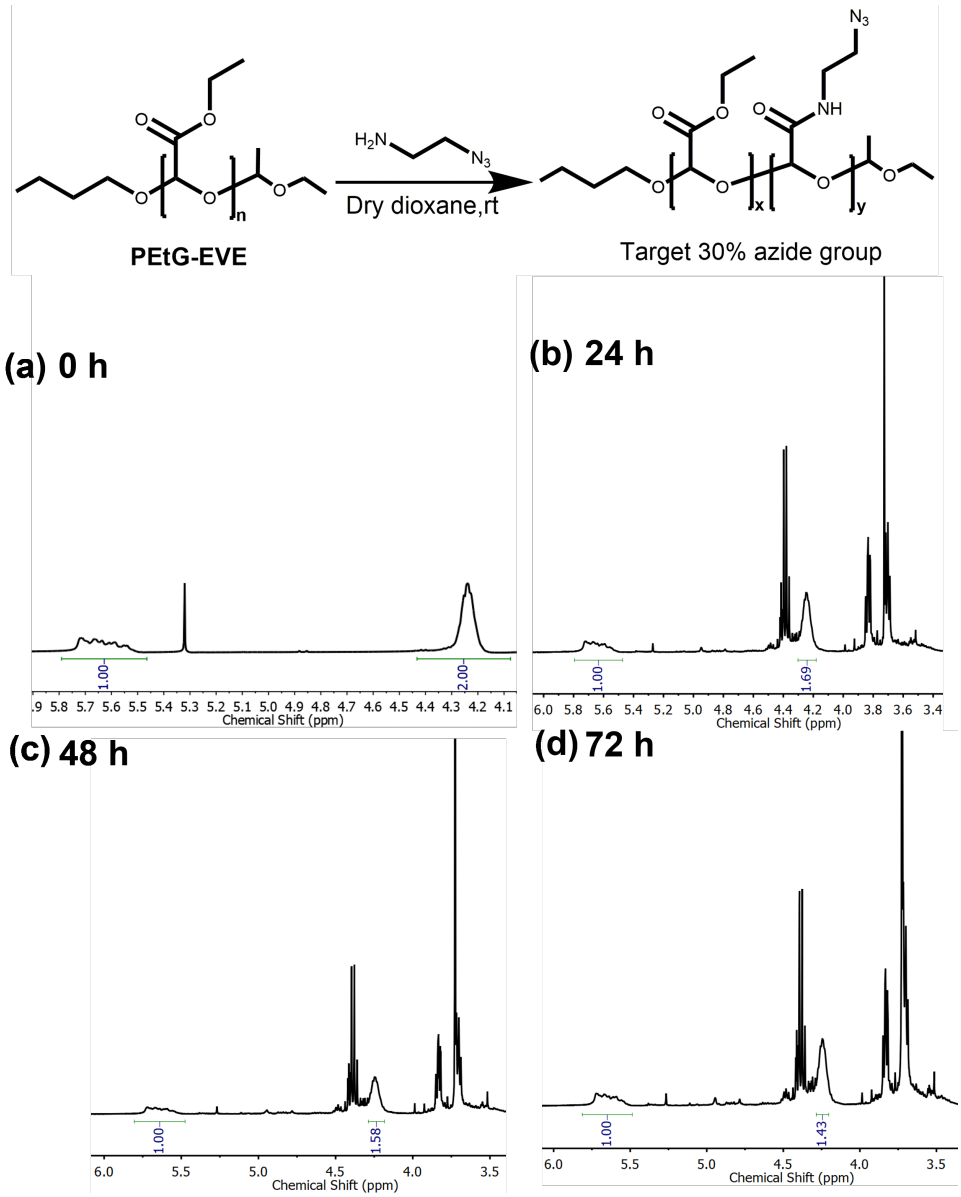

Figure S3. Conversion of 30% of the pendent esters of **PEtG** to pendent 2-azidoethylamides in the synthesis of **TEG-N$_3$-30-PGAm**. The reaction was monitored by ^{1}H NMR spectroscopy. The percent functionalization was determined by comparing the integral of the -CH (methine) peak from the polymer backbone at 5.66 ppm with that of the -CH_2 peak from the pendent ester groups at 4.25 ppm. The reaction was stopped when the integral of -CH_2 peak at 4.25 ppm was ~1.40, indicating ~30% azide functionalization was achieved.

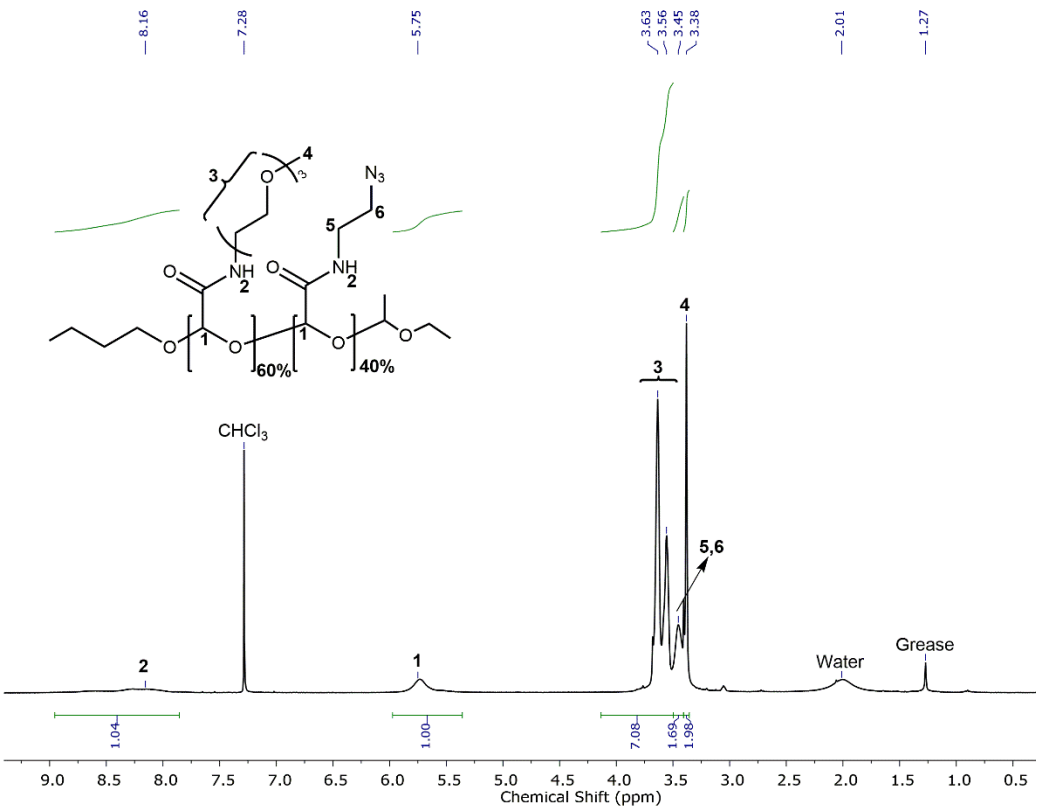

Figure S4. ^{1}H NMR spectrum of **TEG-N_3-40-PGAm** ($CDCl_3$, 400 MHz).

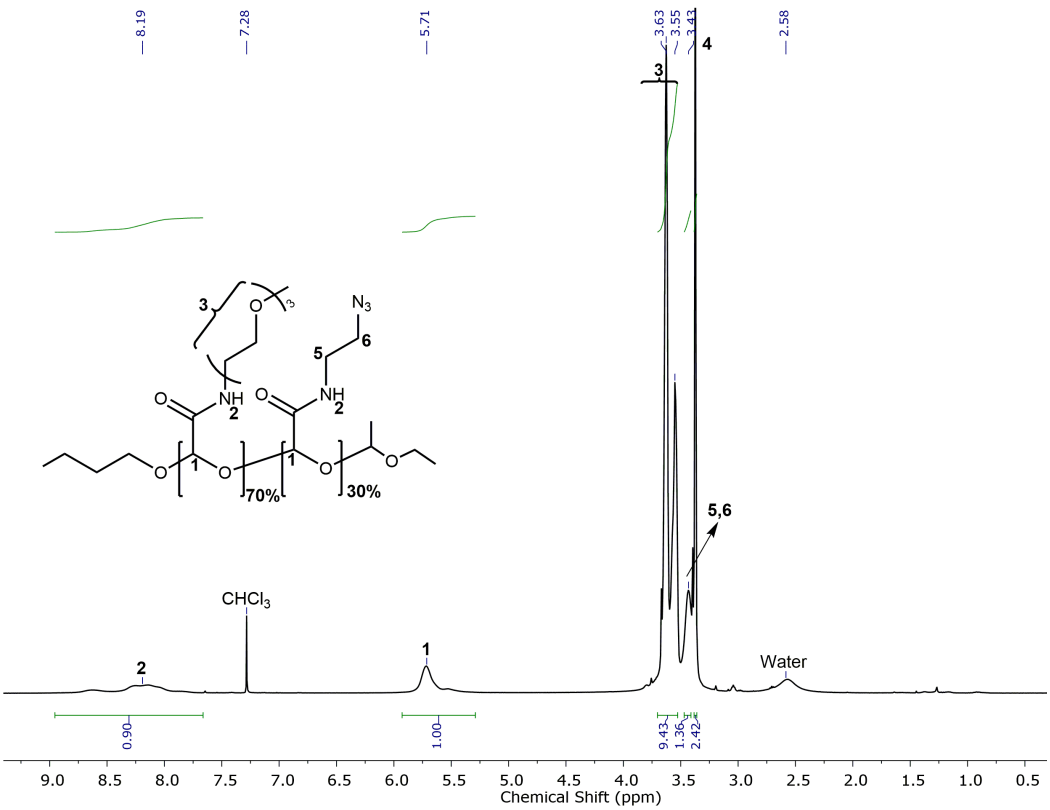

Figure S5. ^{1}H NMR spectrum of **TEG-N_3-30-PGAm** ($CDCl_3$, 400 MHz).

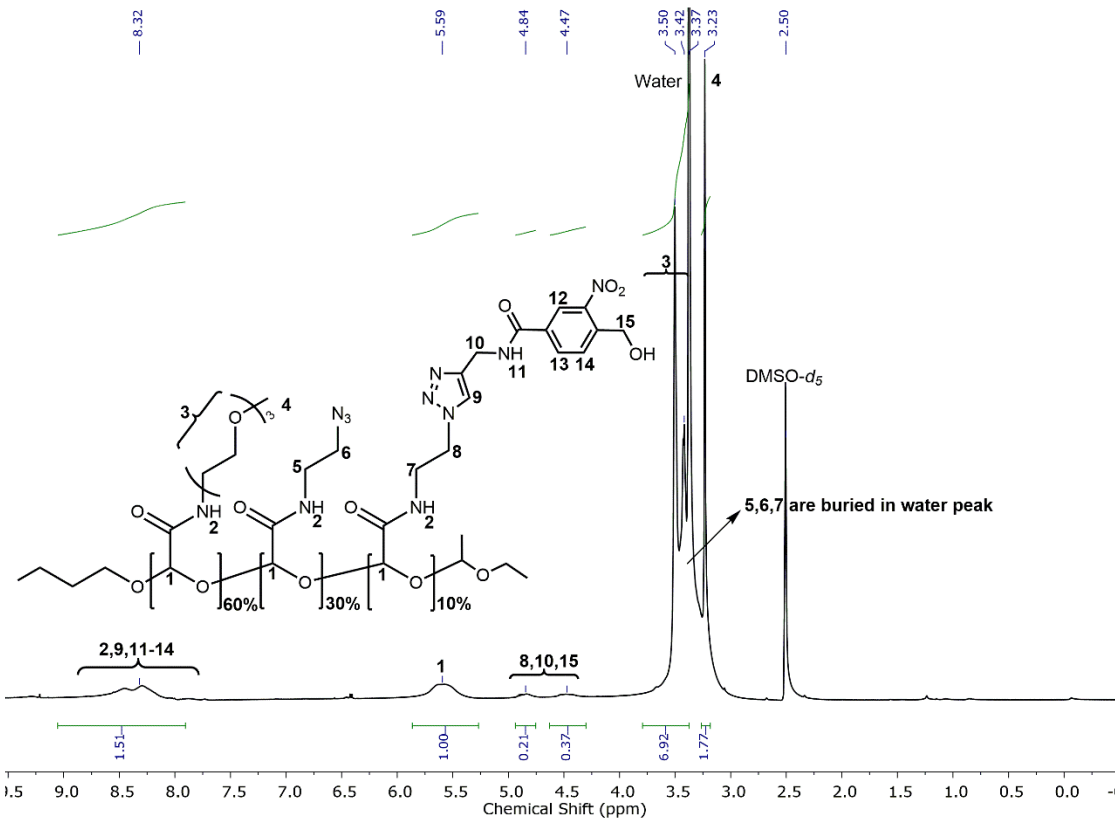

Figure S6. ^{1}H NMR spectrum of ***o*NB-TEG-N$_3$-30-PGAm** (DMSO-d_6, 400 MHz).

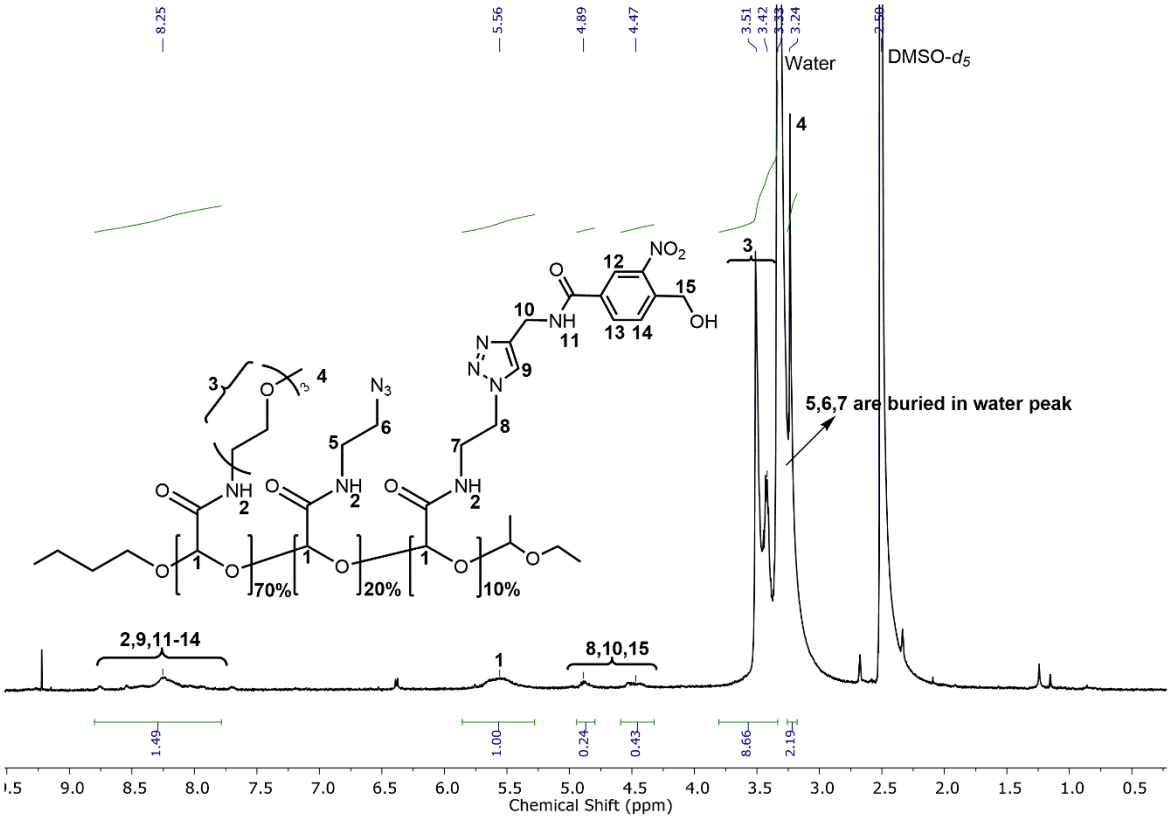

Figure S7. ^{1}H NMR spectrum of ***o*NB-TEG-N$_3$-20-PGAm** (DMSO-d_6, 400 MHz).

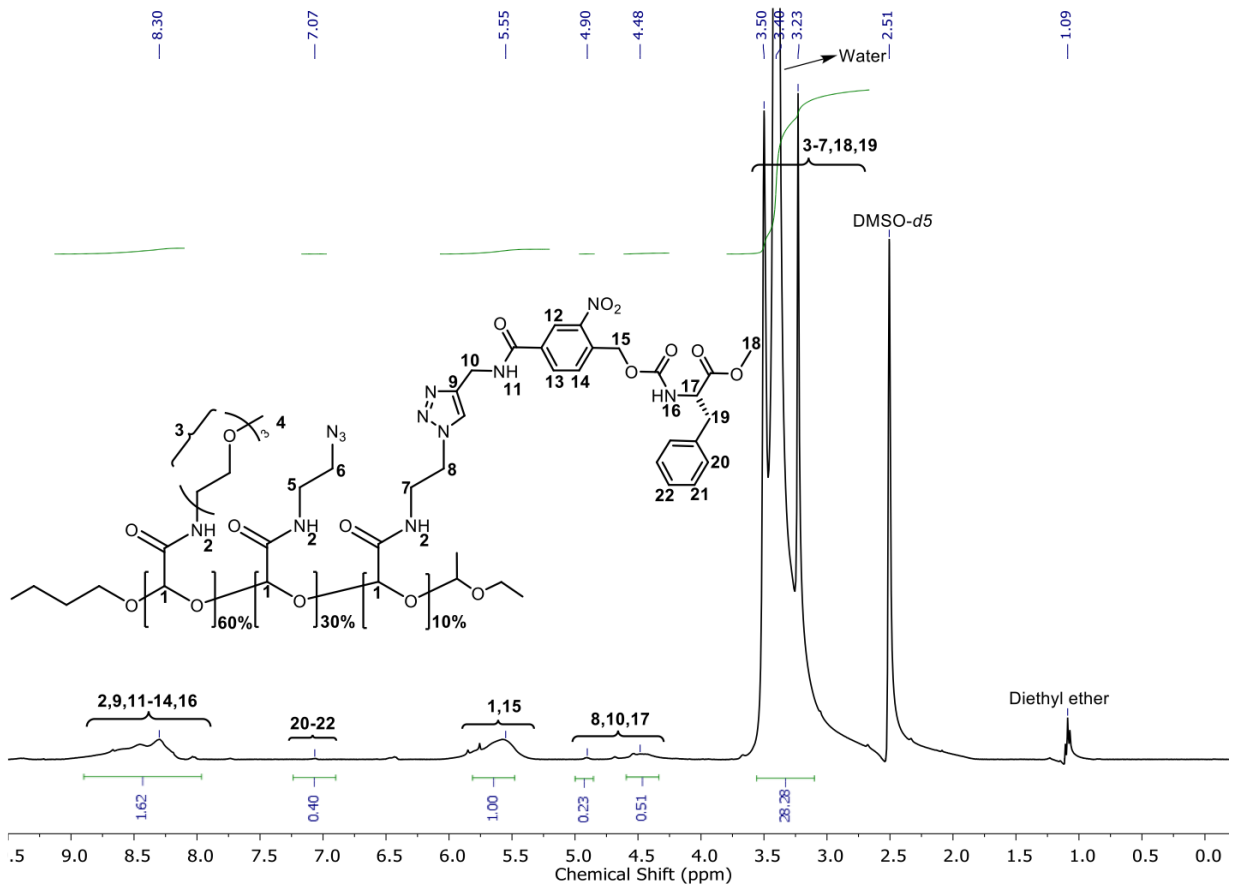

Figure S8. ^{1}H NMR spectrum of **Phe-*o*NB-TEG-N$_3$-30-PGAm** (DMSO-d_6, 400 MHz).

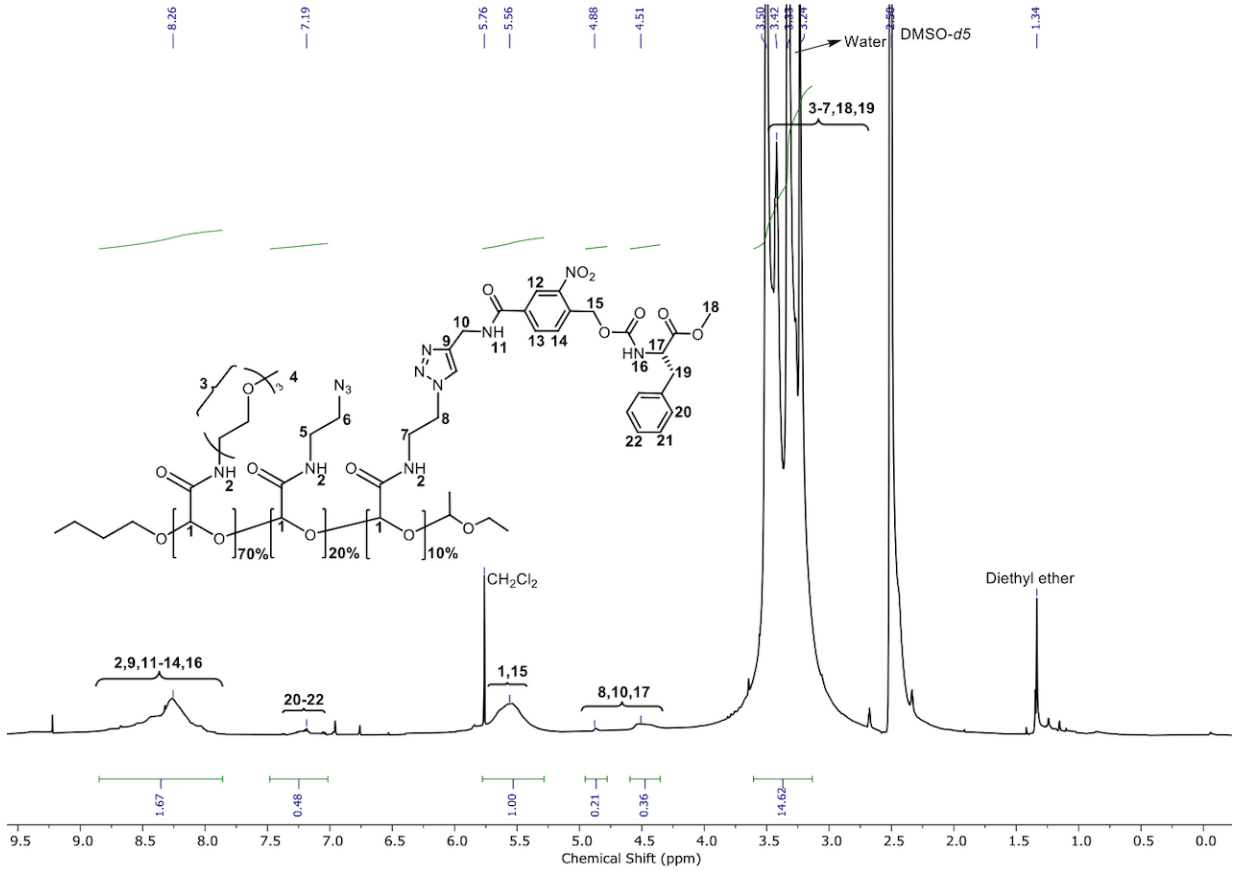

Figure S9. ^{1}H NMR spectrum of **Phe-*o*NB-TEG-N$_3$-20-PGAm** (DMSO-d_6, 400 MHz).

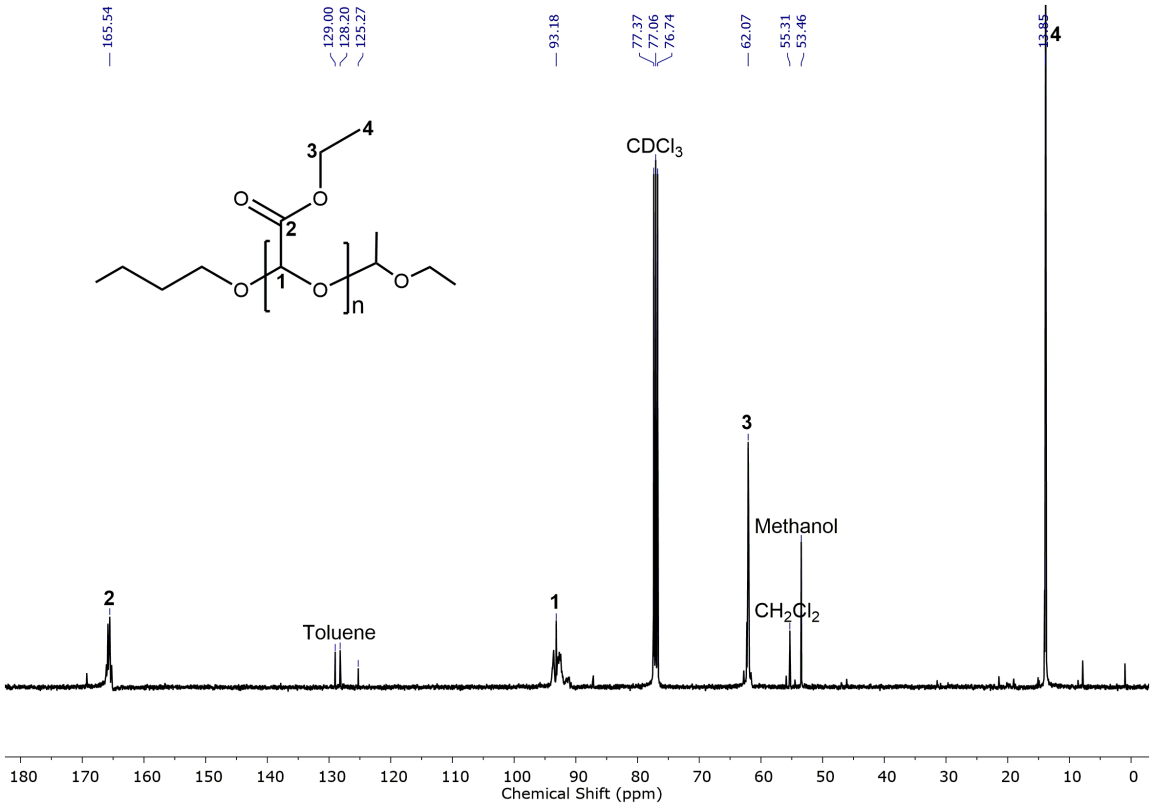

Figure S10. $^{13}C\{^{1}H\}$ NMR spectrum of **PEtG** ($CDCl_3$, 100 MHz). Note that end-cap peaks are not observed in the spectrum due to the high molar mass.

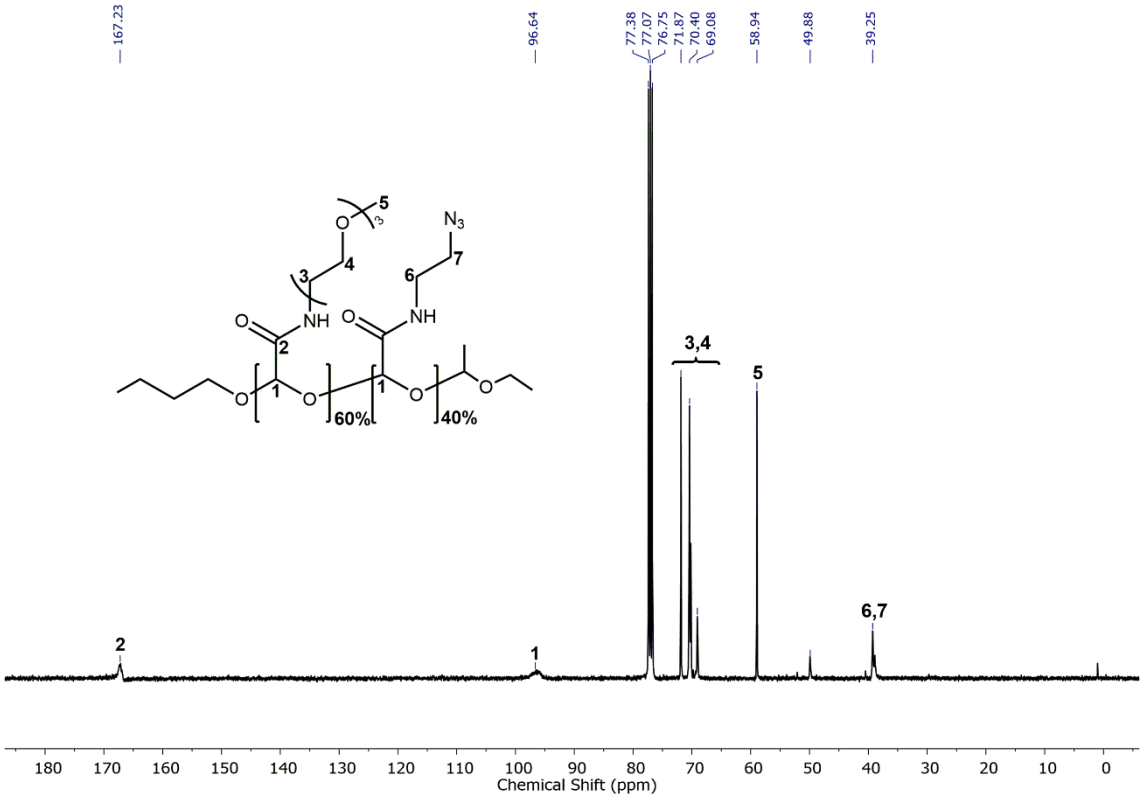

Figure S11. $^{13}C\{^{1}H\}$ NMR spectrum of **TEG-N_3-40-PGAm** ($CDCl_3$, 100 MHz).

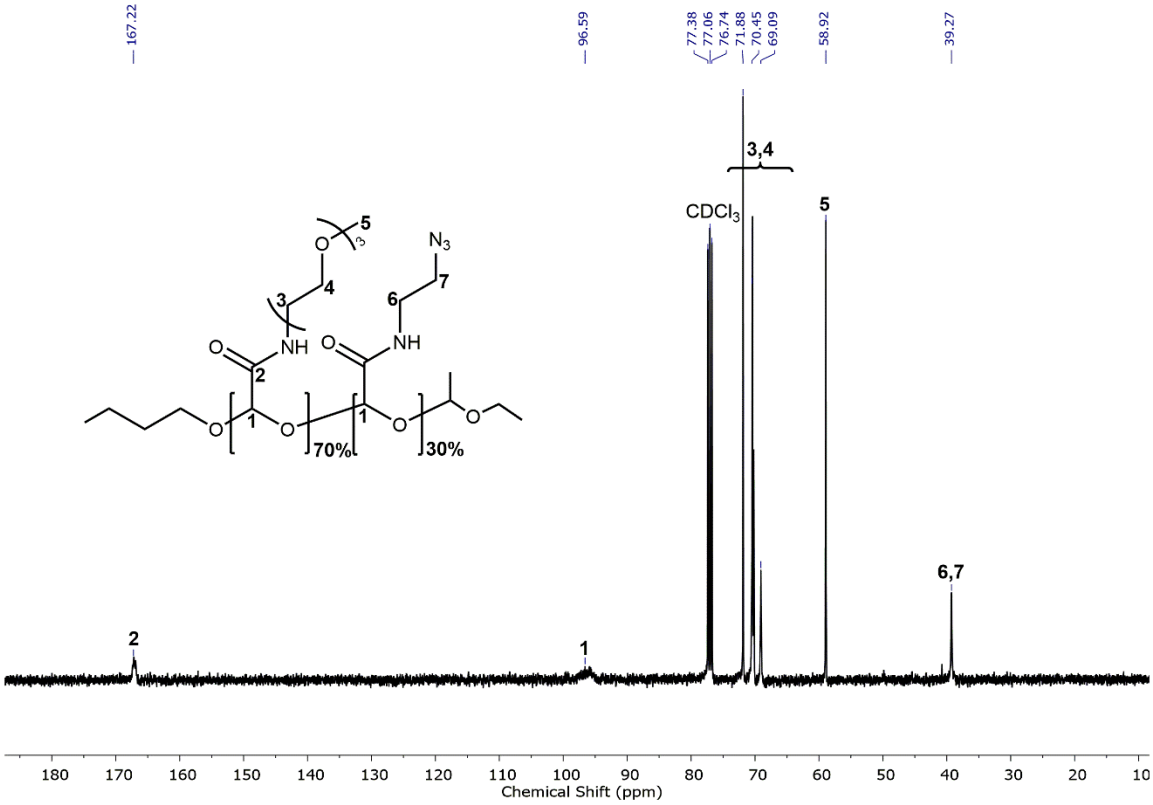

Figure S12. $^{13}C\{^{1}H\}$ NMR spectrum of **TEG-N_3-30-PGAm** ($CDCl_3$, 100 MHz).

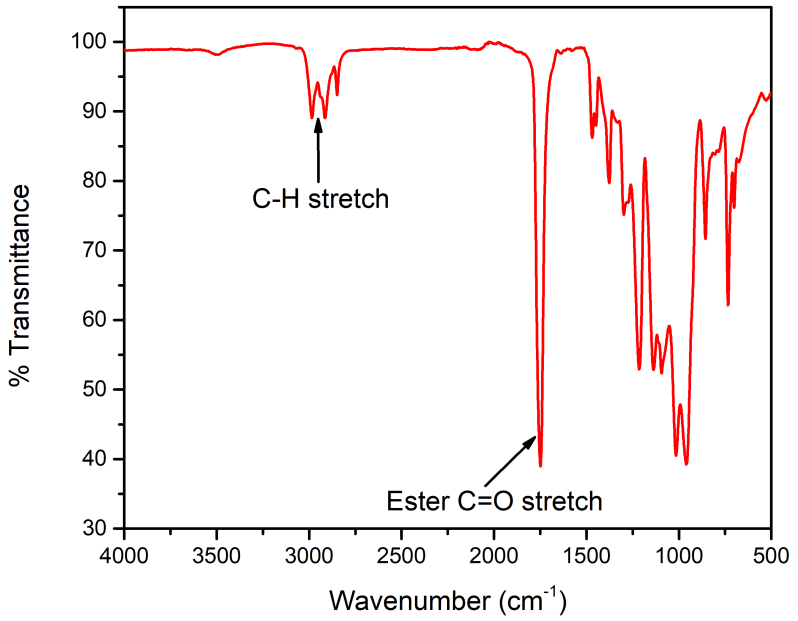

Figure S13. FT-IR spectrum of **PEtG**.

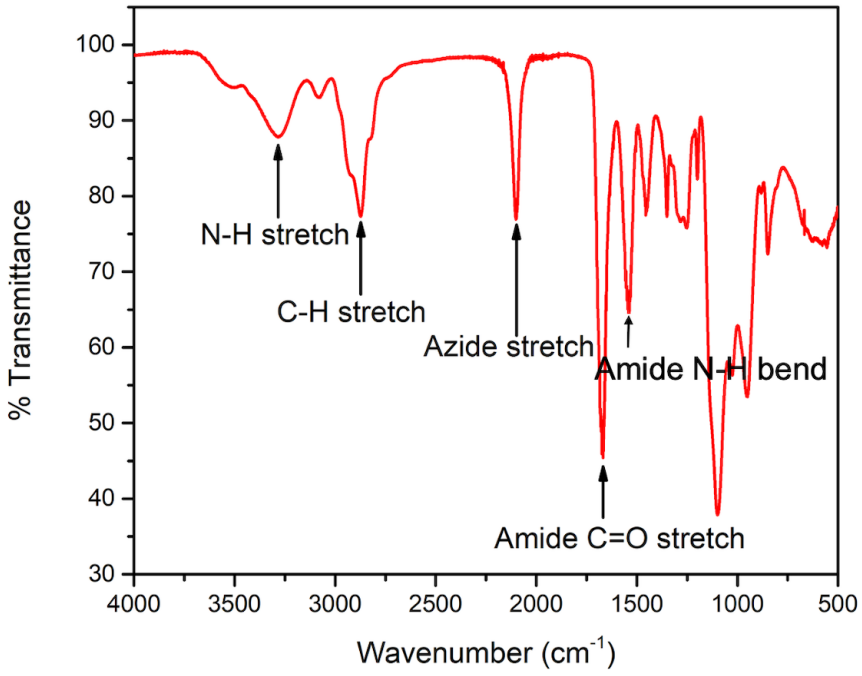

Figure S14. FT-IR spectrum of **TEG-N_3-40-PGAm**.

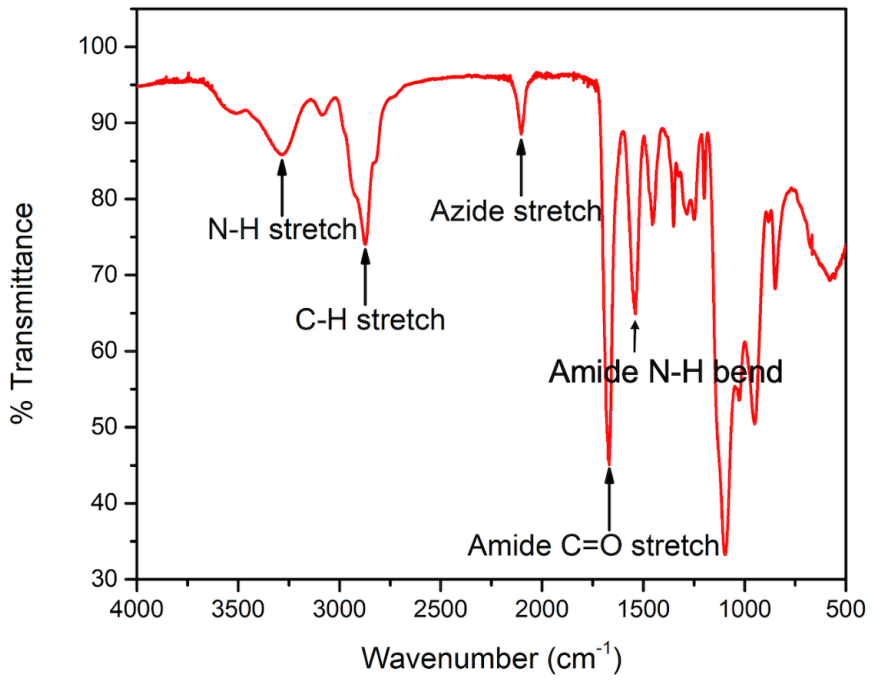

Figure S15. FT-IR spectrum of **TEG-N_3-30-PGAm**.

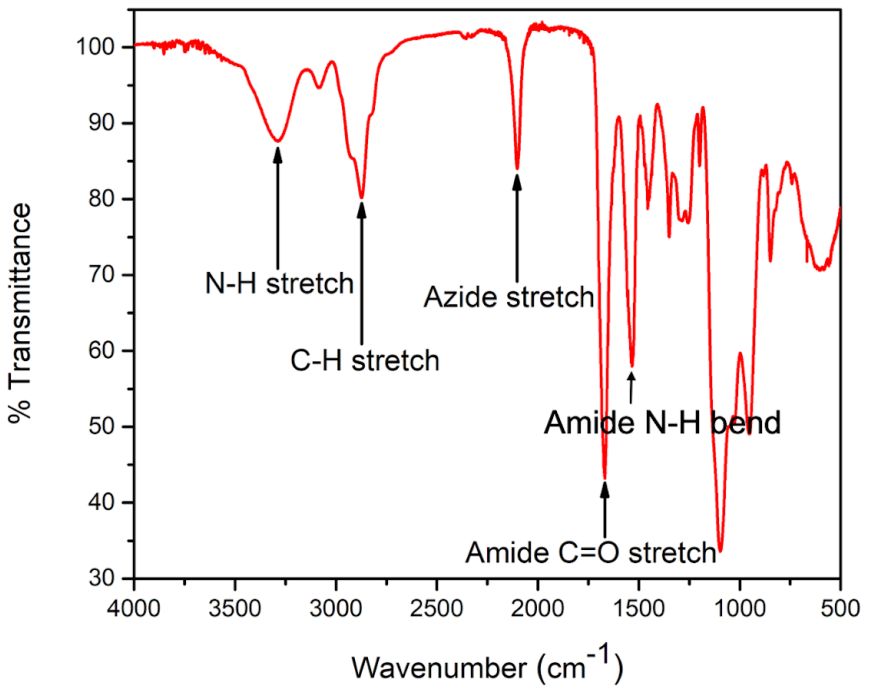

Figure S16. FT-IR spectrum of ***o*NB-TEG-N_3-30-PGAm**.

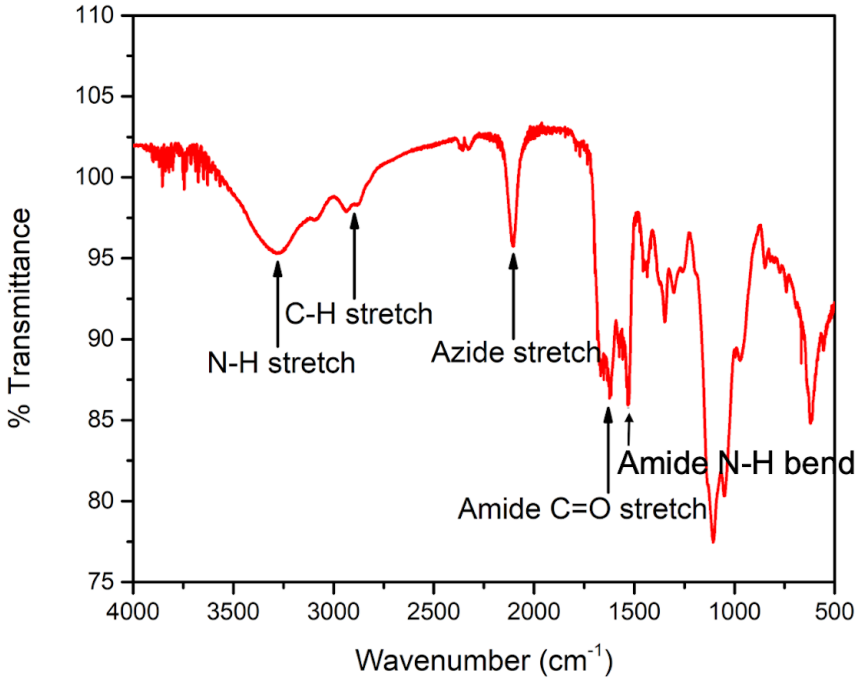

Figure S17. FT-IR spectrum of ***o*NB-TEG-N_3-20-PGAm**.

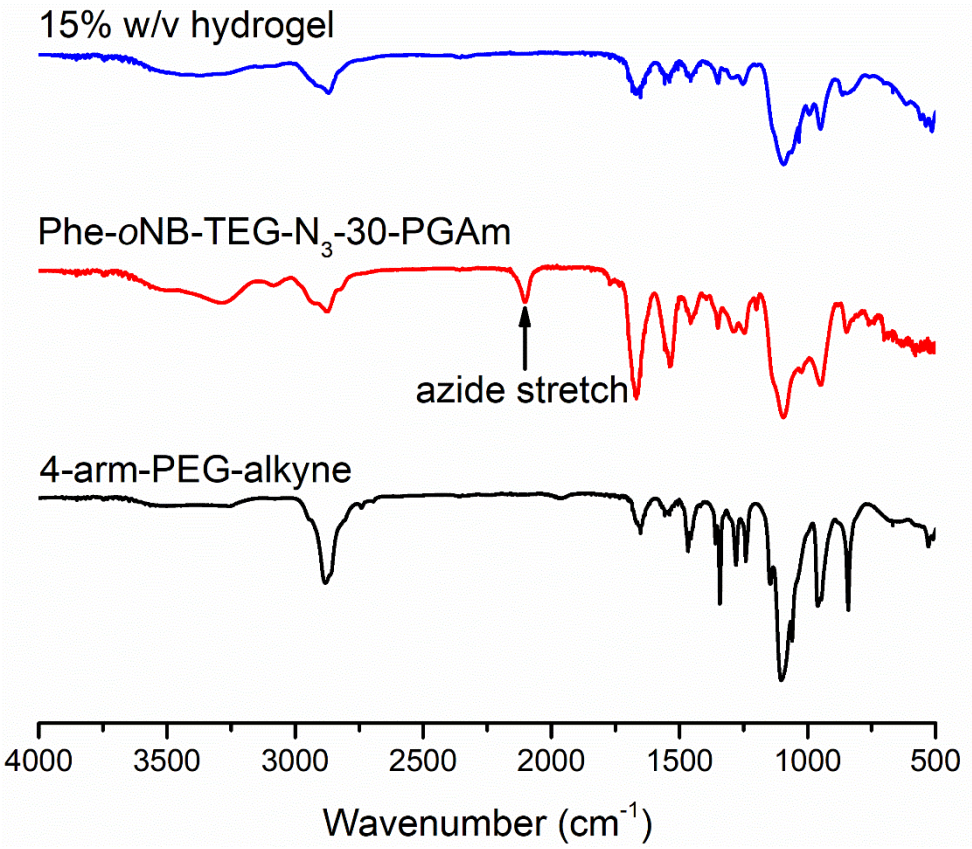

Figure S18. Overlay of the FT-IR spectra of **4-arm-PEG-alkyne**, **Phe-*o*NB-TEG-N_3-30-PGAm** and the corresponding **hydrogel**.

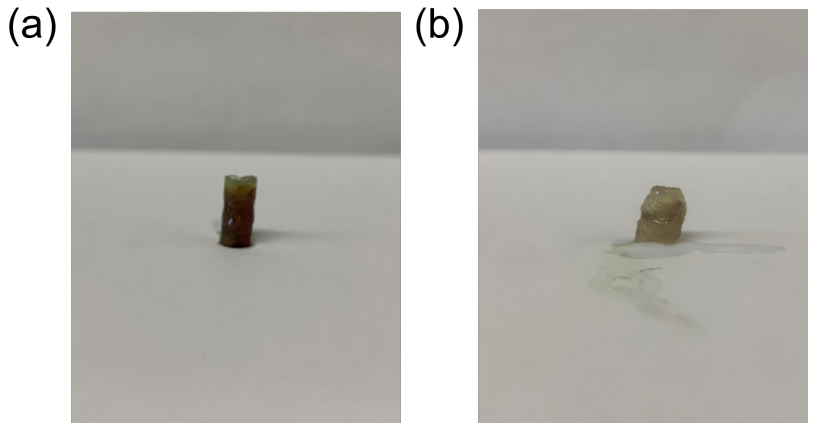

Figure S19. Photos of the 15% w/v **Phe-*o*NB-TEG-N$_3$-30-PGAm** hydrogel a) immediately after gelation and b) after removal of copper and the DMF.

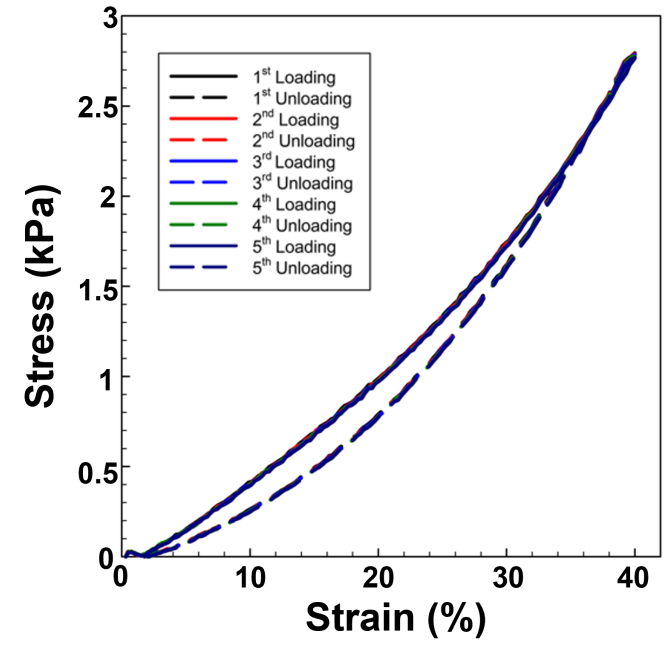

Figure S20. Cyclic loading (compression) and unloading (relaxation) stress-strain curves of a representative **Phe-*o*NB-TEG-N$_3$-30-PGAm** 15% w/v hydrogel, measured in PBS.

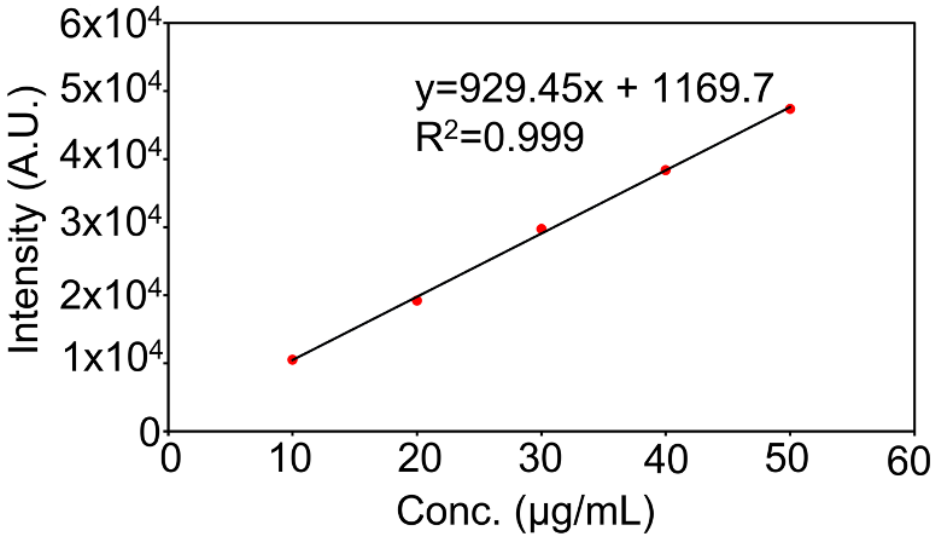

Figure S21. The calibration curve for the fluorescamine assay for Phe methyl ester showing the intensity of fluorescence at 470 nm versus Phe methyl ester concentration.