Supporting Information

Facile Synthesis and Properties of Multifunctionalized Polyesters by Passerini Reaction as Thermosensitive, biocompatible and Triggerable Drug Release Carriers

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Materials

Malonic acid and polyethylene glycols (PEG) with molecular weight of 400-4000 were purchased from Kermel Company (Tianjin, China). Succinic acid and glutaric acid were received from Guangfu Fine Chemical Industry Research Institute (Tianjin, China). Adipic acid, sebacic acid, *tert*-butyl isocyanide and *L*-glutathione (GSH) were obtained from J&K Chemicals (Beijing, China). Glutaraldehyde and succinic anhydride was purchased Aladdin Company (Shanghai, China). Glutaraldehyde was purified by distillation prior to use. 3,3'-dithiodipropionic acid was purchased from TCI (Shanghai) Development Co., Ltd. (Shanghai, China). Triethylamine (TEA) was purified by drying and distillation before using. *4*-Dimethylaminopyridine (DMAP) and obtaining from Shanghai Macklin Biochemical Co. Ltd were used directly.

Doxorubicin (DOX) hydrochloride was obtained from Shanghai Macklin Biochemical Co. Ltd (China). Deionized water was used for all aqueous sample preparations. *N, N*-Dimethylformamide (DMF) was obtained from J&K Chemicals (China) with a chromatographic grade. High precision & ready-to-use dialysis bag (molecular weight cut-off (MWCO), 1000 Da) was purchased from Shanghai Green Bird, Shanghai Toscience Biotechnology Co, Ltd, China. Dulbecco's modified Eagle's medium (DMEM) was purchased from Gibco Company. Moreover, HeLa cells were kindly supplied by Peking Union Medical College Hospital. Other reagents and solvents were commercially analytical grade and used without any further purification.

Measurements of contact angle for thermosensitive polyesters

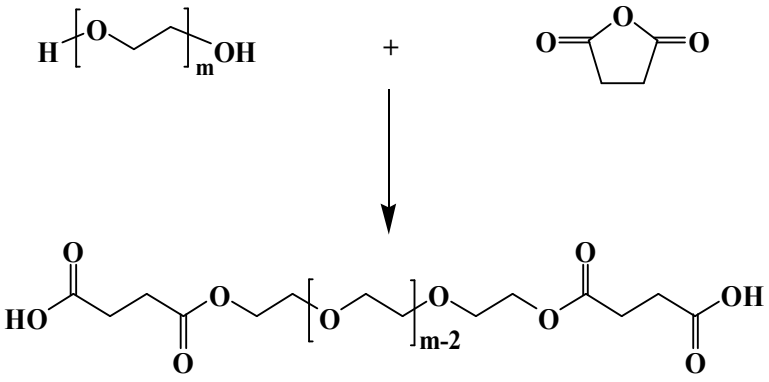
Contact angle measurements of polyesters were performed in air atmosphere by the sessile drop method using a goniometer Data Physics OCA 15EC. Polyester samples were prepared by melting method on a glass plate at about 120 ^{o}C and then placed at room temperature for 2 h for becoming the solid films. The samples were kept at the desired temperature at least 30 min before the measurement. In the contact angle measurement, the angles were recorded after 2 seconds when 2-μL droplet of Milli.Q water was contacted with polyesters samples. The final contact angle for every sample was from the average value of three determined results.

In Vitro release of DOX in the absence or presence of GSH

Firstly, DOX-PSPGBI-800-65 nanocarriers (0.0020 g) were dispersed in PBS (40 mL at pH 7.4). The system was stirred slowly and maintained at 37 ^{o}C. Secondly, the absorbance of DOX released from nanocarriers was measured at 480 nm with a Shimadzu UV-36000 spectrophotometer, approximately every 5 minutes. During the test, the solution volume was maintained constantly by supplementing the fresh 3 mL of PBS after each sampling. The amount of the released DOX was measured by UV-Vis absorbance according to the standard working curve made in advance. In the absence or presence of *L*-glutathione (10 mM) (a chain breaking agent, GSH), the release behaviors of DOX were measured from DOX-PSPGBI-800-65 nanocarriers.

Synthesis of biscarboxylated polyethylene glycol (S-PEG) with different M_n

According to the reported methods by Zi-Chen Li[13], in the representative preparation of biscarboxylated S-PEG-1000, 10.00 g, (10 mmol) PEG with *Mn* 1000 g/mol (PEG-1000), 2.600 g, (26 mmol) succinic anhydride, 2.440 g, (20 mmol) DMAP and 2.021 g, (20 mmol) TEA were dissolved in 125 mL 1,4-dioxane. The solution was stirred for 24 h at 25 °C, next concentrated, and added into 125 mL deionized water. The aqueous solution was then adjusted to pH = 3 with concentrated hydrochloric acid. Finally, the solution was extracted with CH_2Cl_2, the organic phase was collected and precipitated into cold ethyl ether twice. The final products, biscarboxylated polyethylene glycol-1000 (S-PEG-1000) was obtained after vacuum drying. The other biscarboxylated polyethylene glycol (S-PEG-400, S-PEG-800, S-PEG-2000, S-PEG-4000) were prepared to the above procedure.



m=9, S-PEG-400; m=18, S-PEG-800; m=22, S-PEG-1000; m=45, S-PEG-2000; m=90, S-PEG-4000

Scheme S1. Synthesis of Five S-PEGs

Table S1 Effect of the experimental conditions on Passerini-3CP

Samples	Solvents	[a]Ratio	[b]Conc (mol/L)	T ([o]C)	T (h)	[c]Yield (%)	[d]M_n (10^4 g/mol)	PDI
PSGBI	CH_2Cl_2	1:1:1.8	1.0	20	48	36.5	2.06	1.46
PSGBI	CH_2Cl_2	1:1:2.0	1.0	20	48	33.9	2.99	1.70
PSGBI	CH_2Cl_2	1:1:2.2	1.0	20	48	34.1	2.08	1.46
PSGBI	CH_2Cl_2	1:1:2.4	1.0	20	48	32.1	2.08	1.47
PSGBI	CH_2Cl_2	1:1:2.0	0.5	20	48	39.2	1.70	1.38
PSGBI	CH_2Cl_2	1:1:2.0	1.5	20	48	39.3	2.87	1.56
PSGBI	CH_2Cl_2	1:1:2.0	2.0	20	48	47.3	3.03	1.62
PSGBI	$CHCl_3$	1:1:2.0	1.0	20	48	41.6	1.39	1.59
PSGBI	CH_3OH	1:1:2.0	1.0	20	48	28.7	0.44	1.60
PSGBI	THF	1:1:2.0	1.0	20	48	45.3	0.53	1.65
PSGBI	Toluene	1:1:2.0	1.0	20	48	44.7	1.42	1.73
PSGBI	H_2O	1:1:2.0	1.0	20	48	53.6	0.74	2.11
PSGBI	CH_2Cl_2	1:1:2.0	1.0	30	48	50.1	1.54	1.73
PSGBI	CH_2Cl_2	1:1:2.0	1.0	20	72	85.8	1.42	1.58
PSGBI	CH_2Cl_2	1:1:2.0	1.0	20	36	48.4	1.34	1.58
PMGBI	CH_2Cl_2	1:1:2.0	1.0	20	48	22.5	0.31	1.76
PBGBI	CH_2Cl_2	1:1:2.0	1.0	20	48	19.6	---	---
PGGBI	CH_2Cl_2	1:1:2.0	1.0	20	48	45.6	0.12	1.97
PAGBI	CH_2Cl_2	1:1:2.0	1.0	20	48	30.1	1.50	1.70

[a] Molar ratio of dicarboxylic acid to glutaraldehyde to *tert*-butyl isocyanide;

[b] Here shows the concentration of dicarboxylic acids, which is equal to the concentration of glutaraldehyde. The concentration of *tert*-butyl isocyanides is double in molar ratio; t: Time; T: Temperature;

[c] Determined after precipitation in ethyl ether and vacuum drying (30°C, for 12 h);

[d] Measured by GPC in DMF.

Table S2 LCSTs of polyesters from several S-PEG

Samples	M_n (10^4 g/mol)	PDI	LCST (^{o}C)	Solvent
PPGBI-400	1.75	1.68	28.5	H_2O
PPGBI-800	2.02	1.45	69.2	H_2O
PPGBI-1000	2.24	1.83	72.8	H_2O
PPGBI-2000	1.89	1.48	84.2	Normal Saline
PSGBI-4000	2.46	1.67	---	H_2O/Normal Saline
CoP-75-25	1.88	1.56	38.7	H_2O
CoP-50-50	2.13	1.82	42.3	H_2O
CoP-25-75	2.09	1.71	58.0	H_2O
PSPGBI-800-55	2.48	1.41	48.5	H_2O
PSPGBI-800-60	3.01	1.58	40.4	H_2O
PSPGBI-800-65	2.30	1.54	33.4	H_2O

The basic conditions for Passerini-3CP are listed as follow: Molar ratio of dicarboxylic acid to glutaraldehyde to *tert*-butyl isocyanide is controlled as 1:1:2 Solvent for Passerini-3CP is CH_2Cl_2; Polymerization time is 48 hours; Polymerization temperature is room temperature; M_n was measured by GPC in DMF.

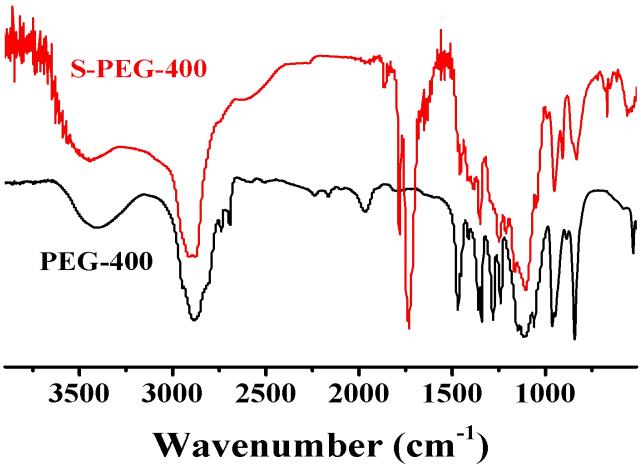


Figure S1. FTIR spectra of PEG-400 and S-PEG-400

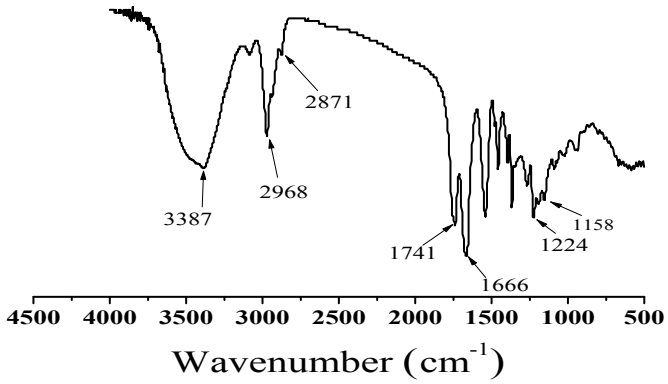


Figure S2. FTIR spectrum of PMGBI

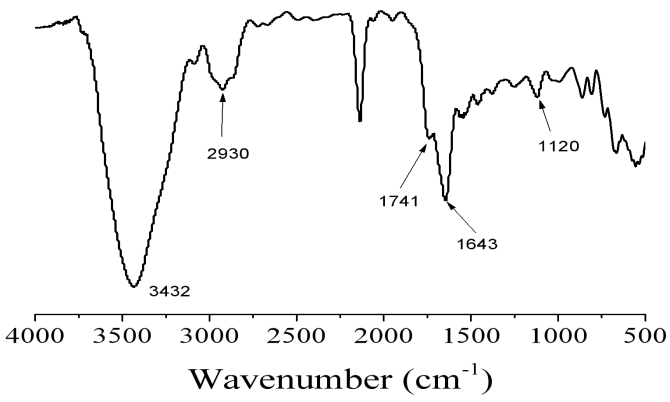


Figure S3. FTIR spectrum of PBGBI

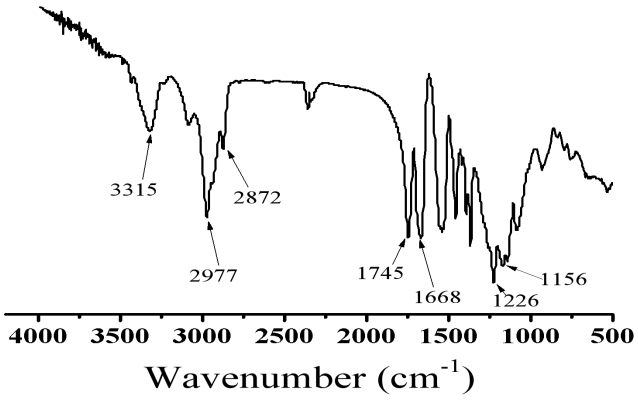


Figure S4. FTIR spectrum of PGGBI

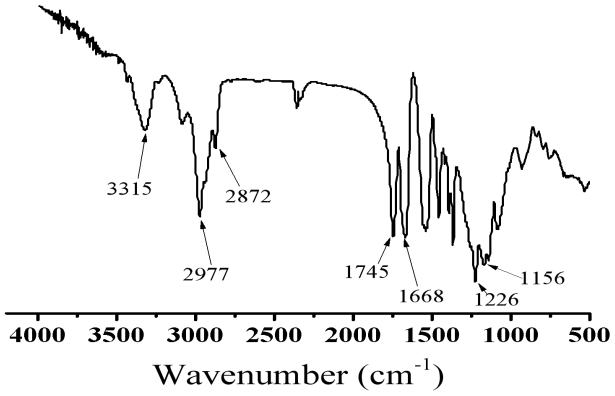


Figure S5. FTIR spectrum of PAGBI

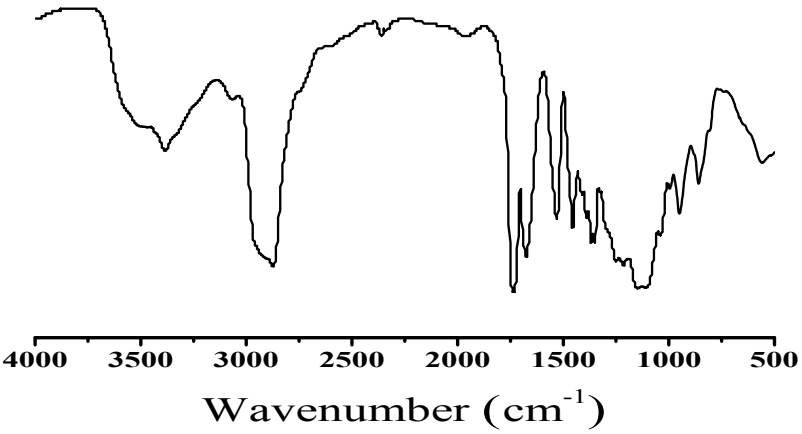


Figure S6. FTIR spectrum of PPGBI-400

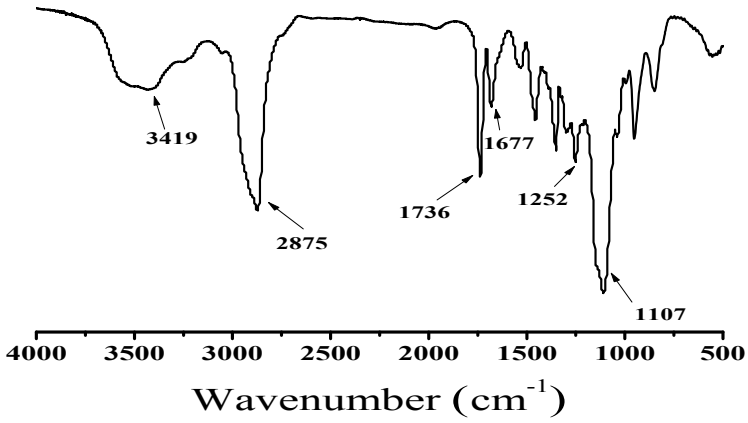


Figure S7. FTIR spectrum of PPGBI-1000

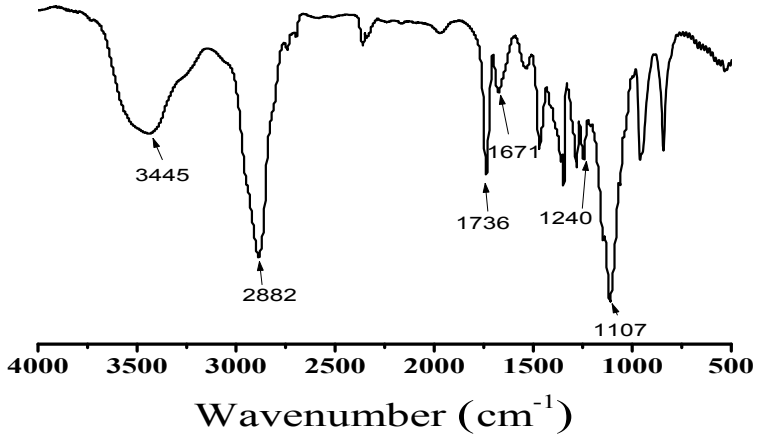


Figure S8. FTIR spectrum of PPGBI-2000

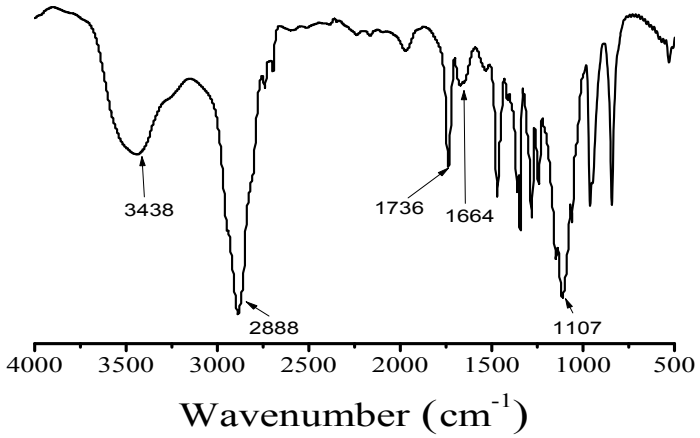


Figure S9. FTIR spectrum of PPGBI-4000

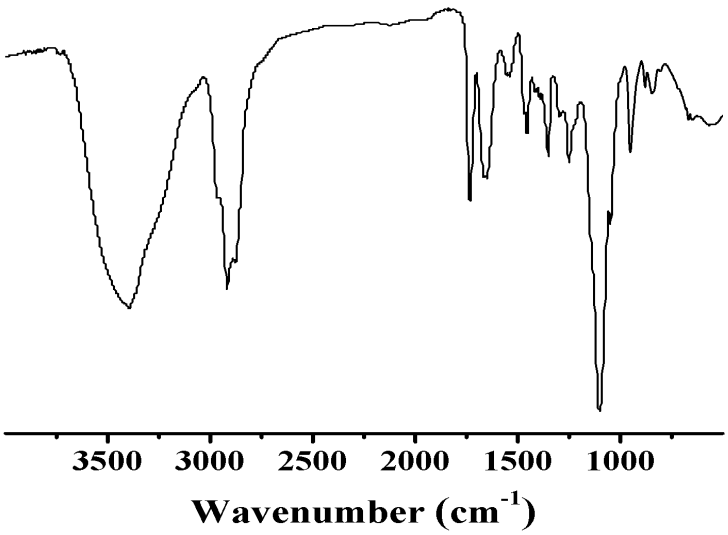


Figure S10. FTIR spectrum of PSPGBI-800-55

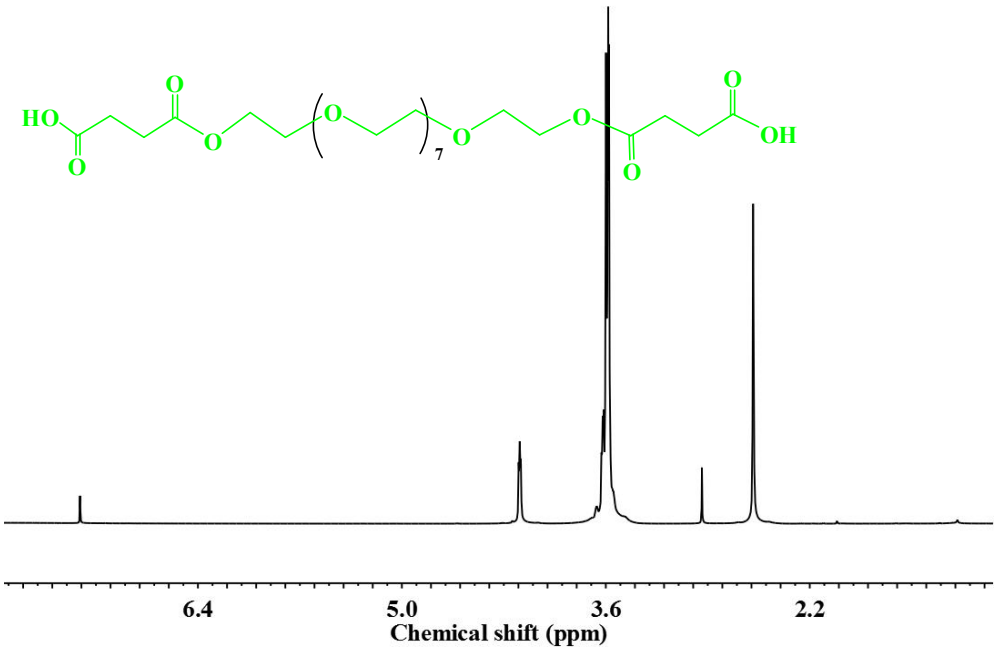


Figure S11. ^{1}H NMR spectrum of S-PEG-400 (in $CDCl_3$)

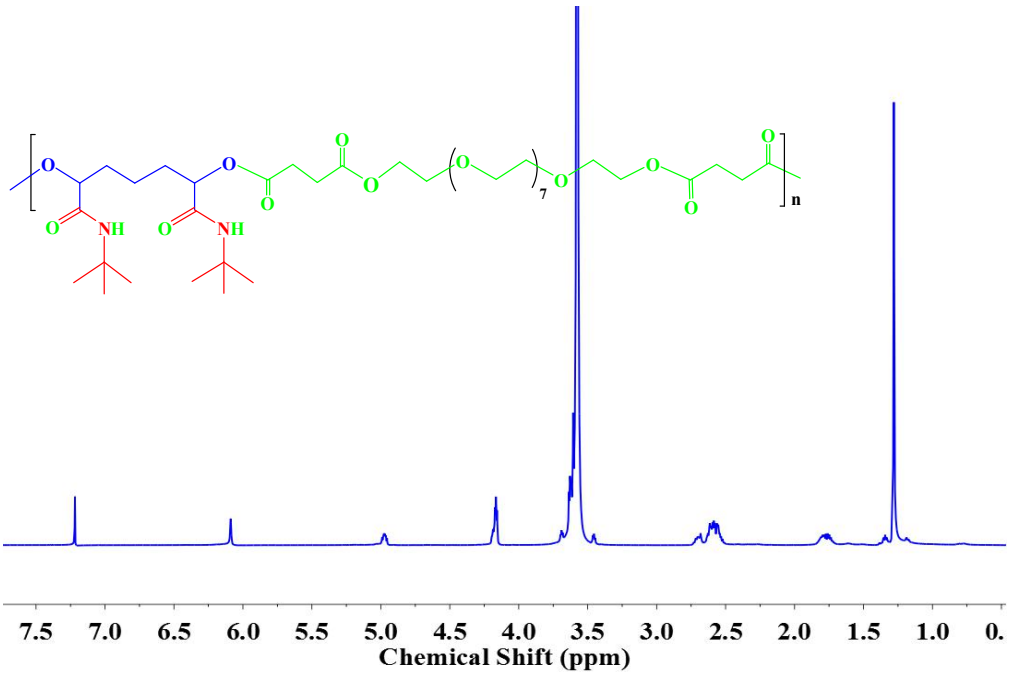


Figure S12. ^{1}H NMR spectrum of PPGBI-400 (in $CDCl_3$)

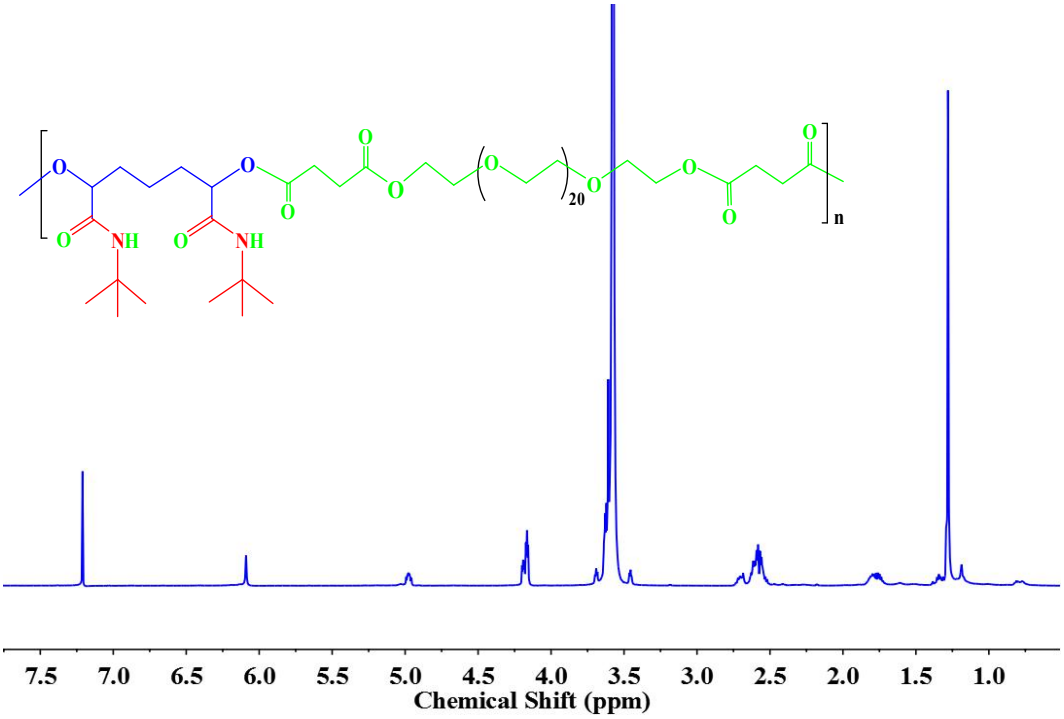


Figure S13. ^{1}H NMR spectrum of PPGBI-1000 (in $CDCl_3$)

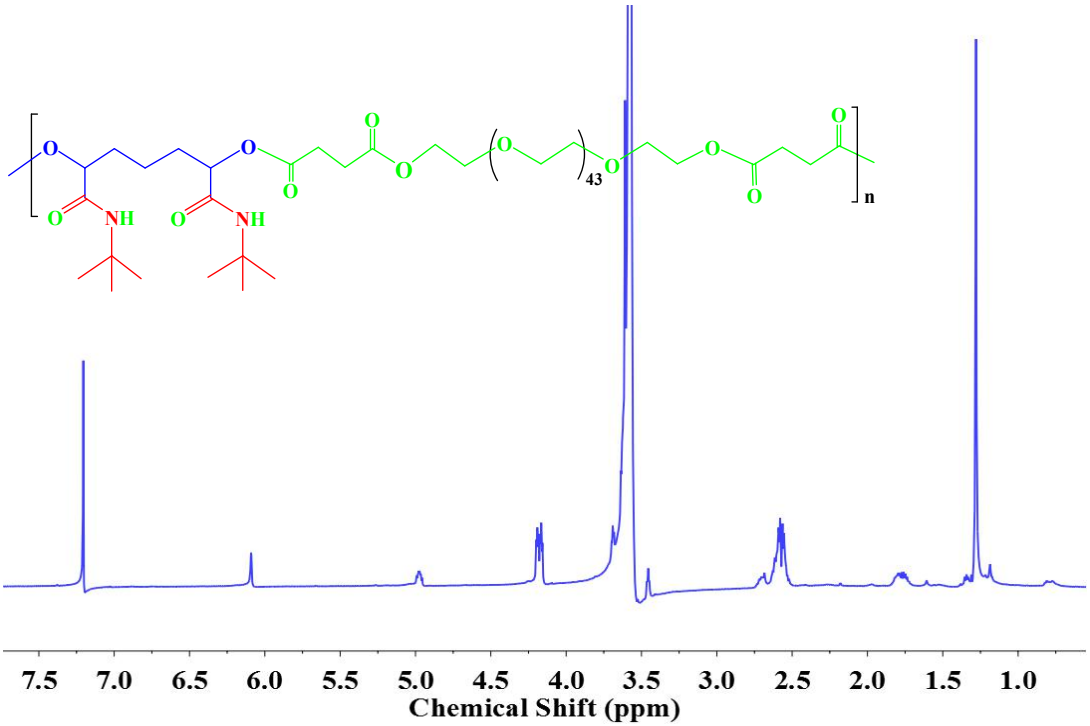


Figure S14. ^{1}H NMR spectrum of PPGBI-2000 (in $CDCl_3$)

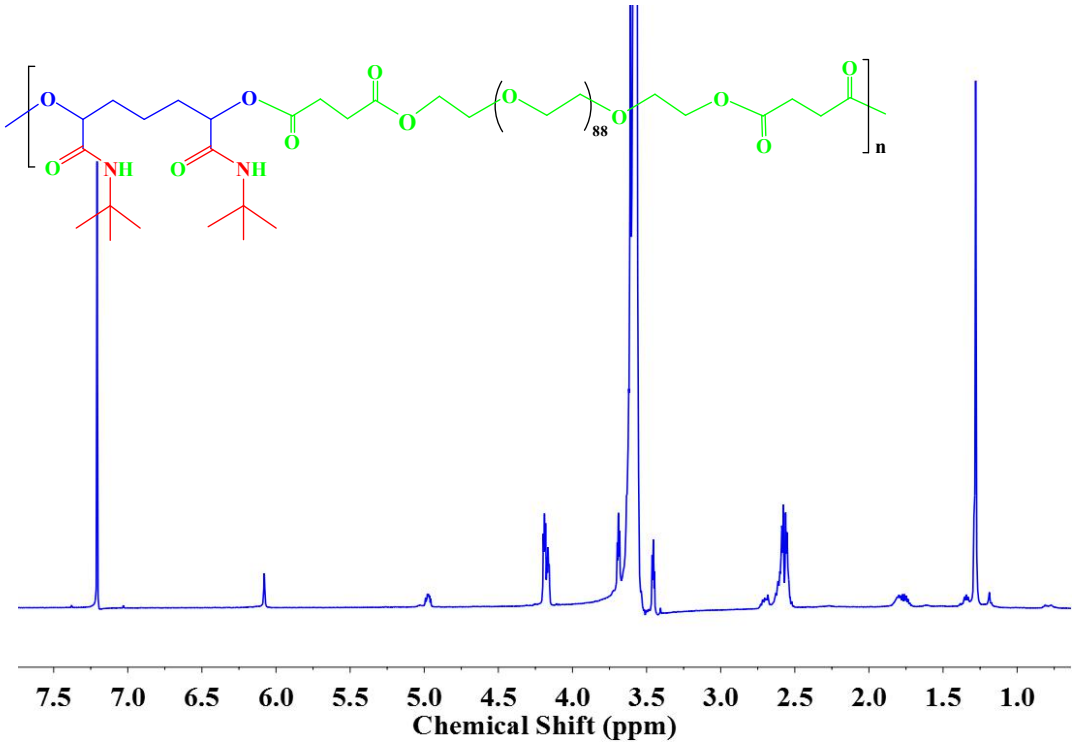


Figure S15. ^{1}H NMR spectrum of PPGBI-4000 (in $CDCl_3$)

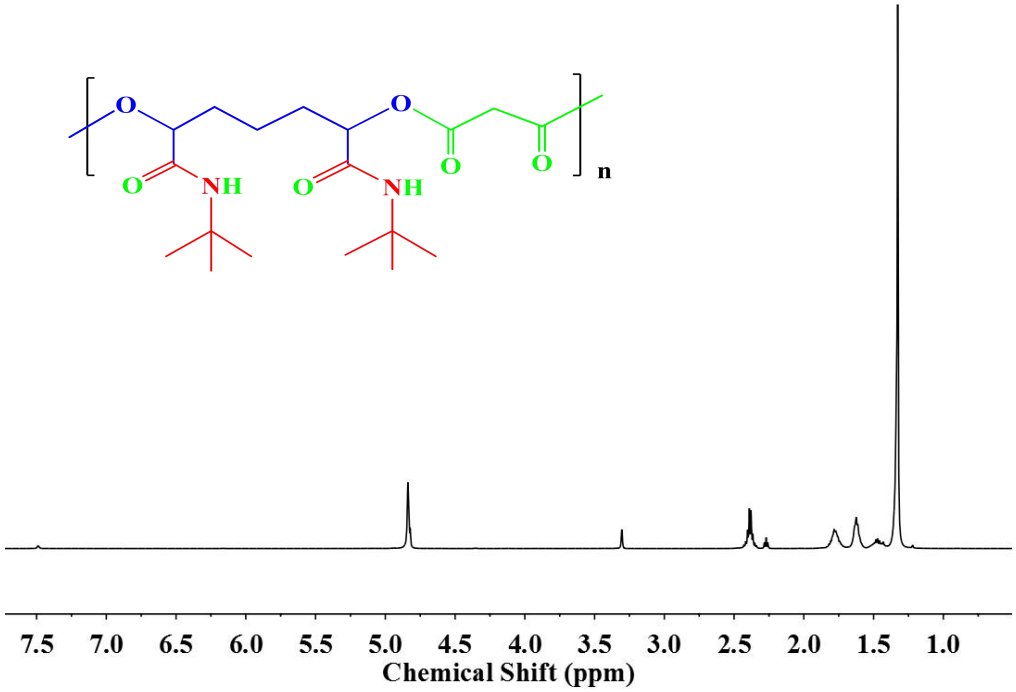


Figure S16. ^{1}H NMR spectrum of PMGBI (in $CDCl_3$)

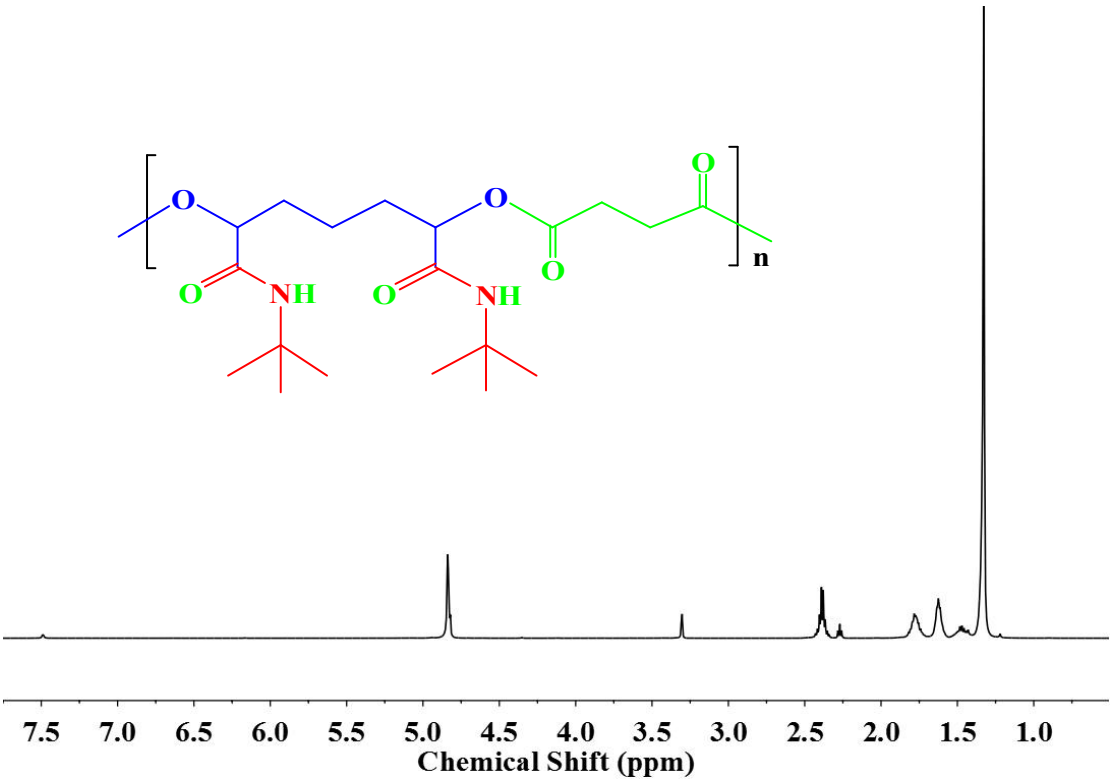


Figure S17. ^{1}H NMR spectrum of PBGBI (in $CDCl_3$)

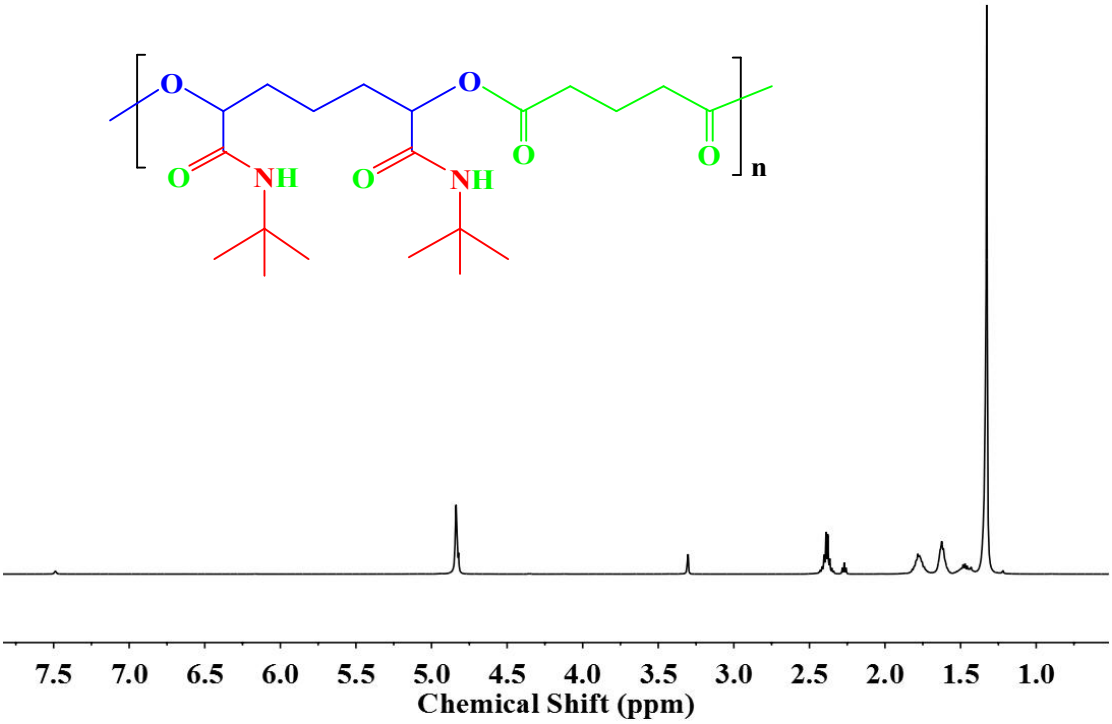


Figure S18. ^{1}H NMR spectrum of PPGBI (in $CDCl_3$)

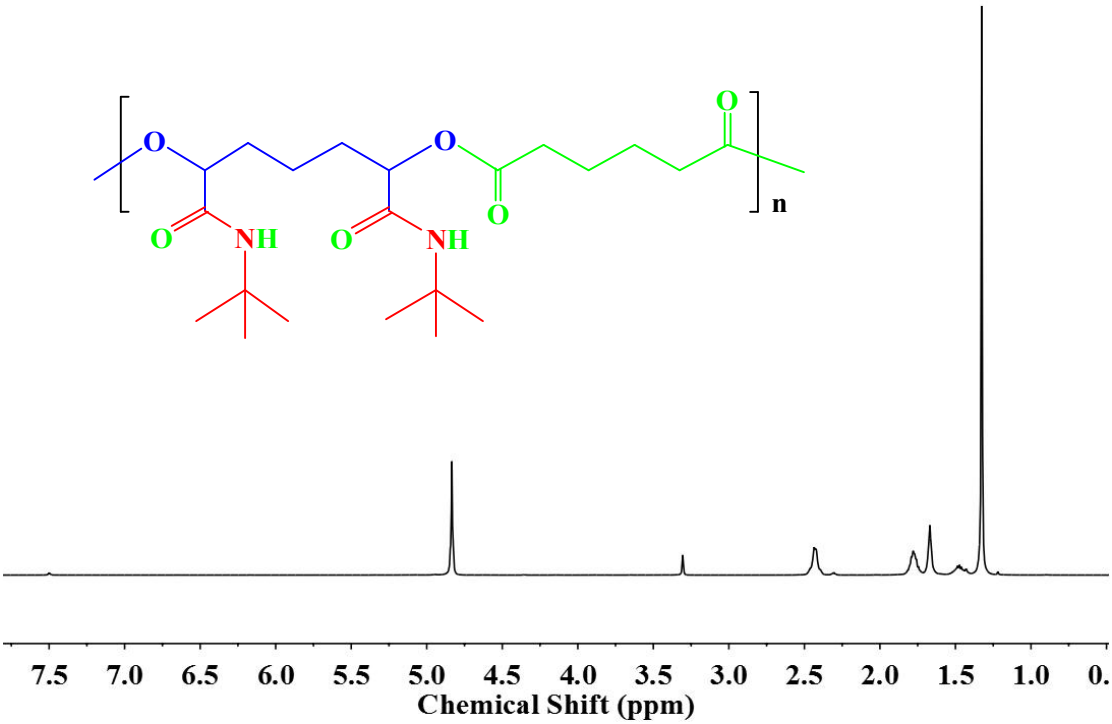


Figure S19. ^{1}H NMR spectrum of PAGBI (in $CDCl_3$)

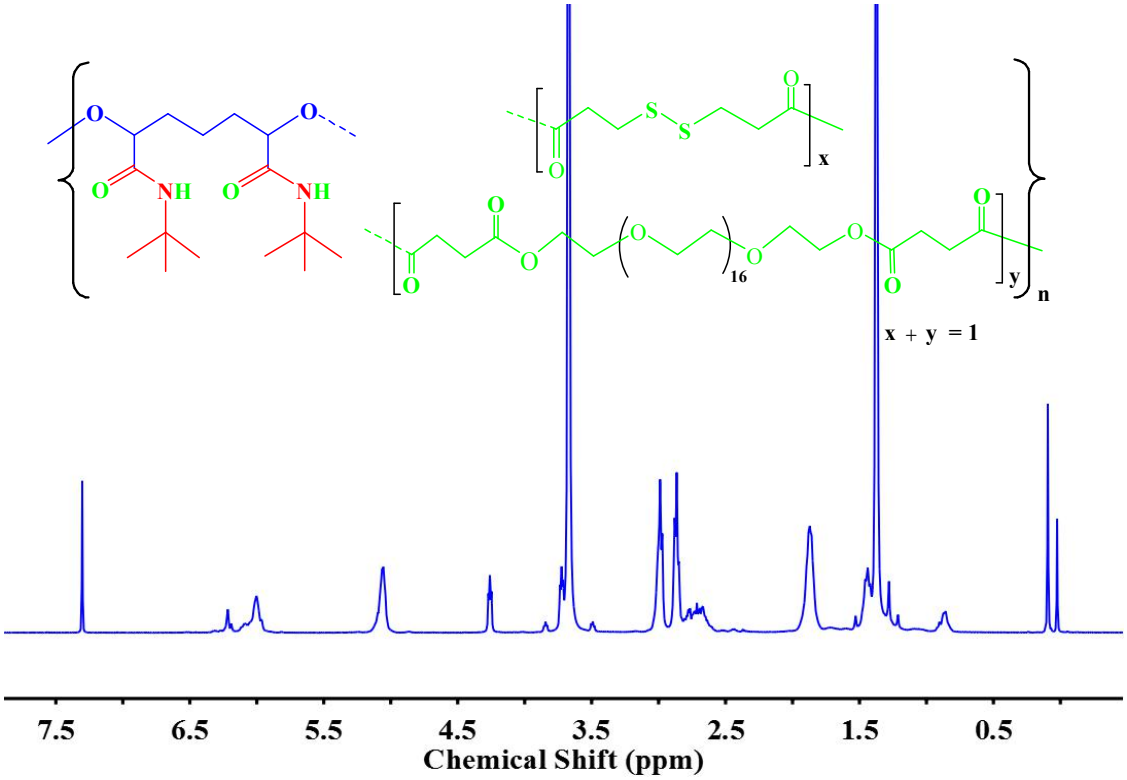


Figure S20. ^{1}H NMR spectrum of PSPGBI-800-55 (in $CDCl_3$)

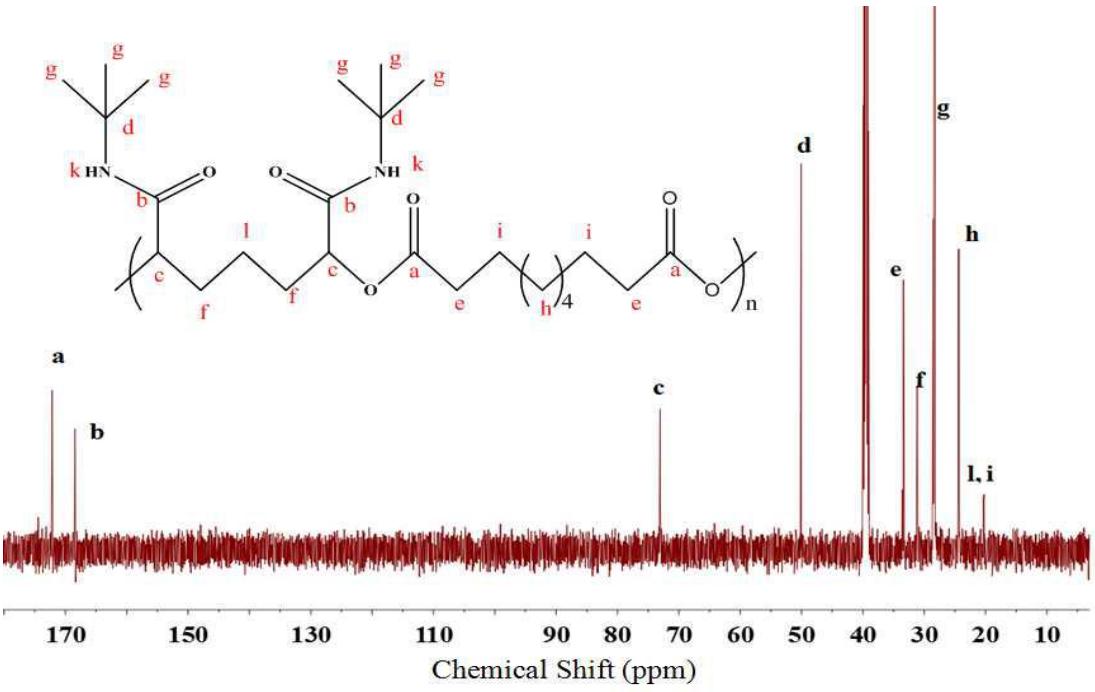


Figure S21. ^{13}C NMR spectrum of PSGBI

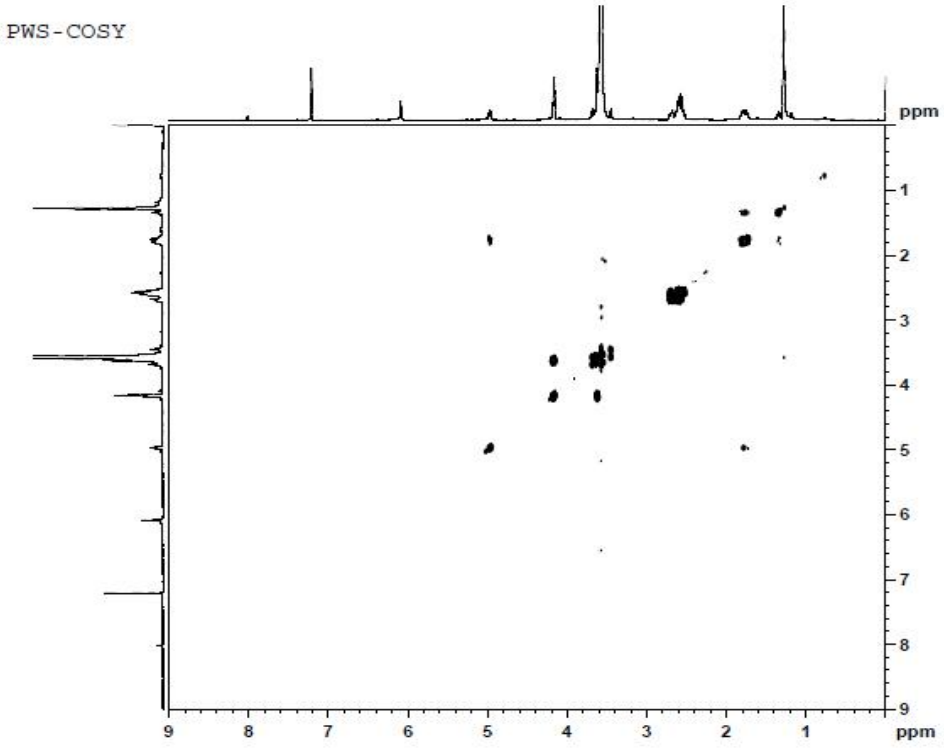


Figure S22. 2D NMR spectrum (COSY) of PPGBI-800

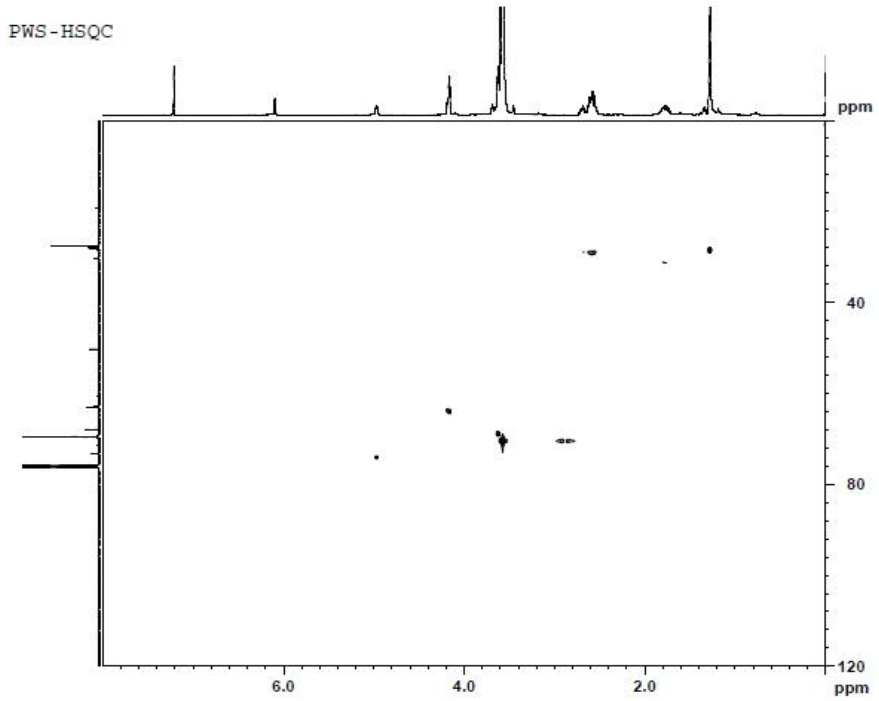


Figure S23. 2D NMR spectrum (HSQC) of PPGBI-800

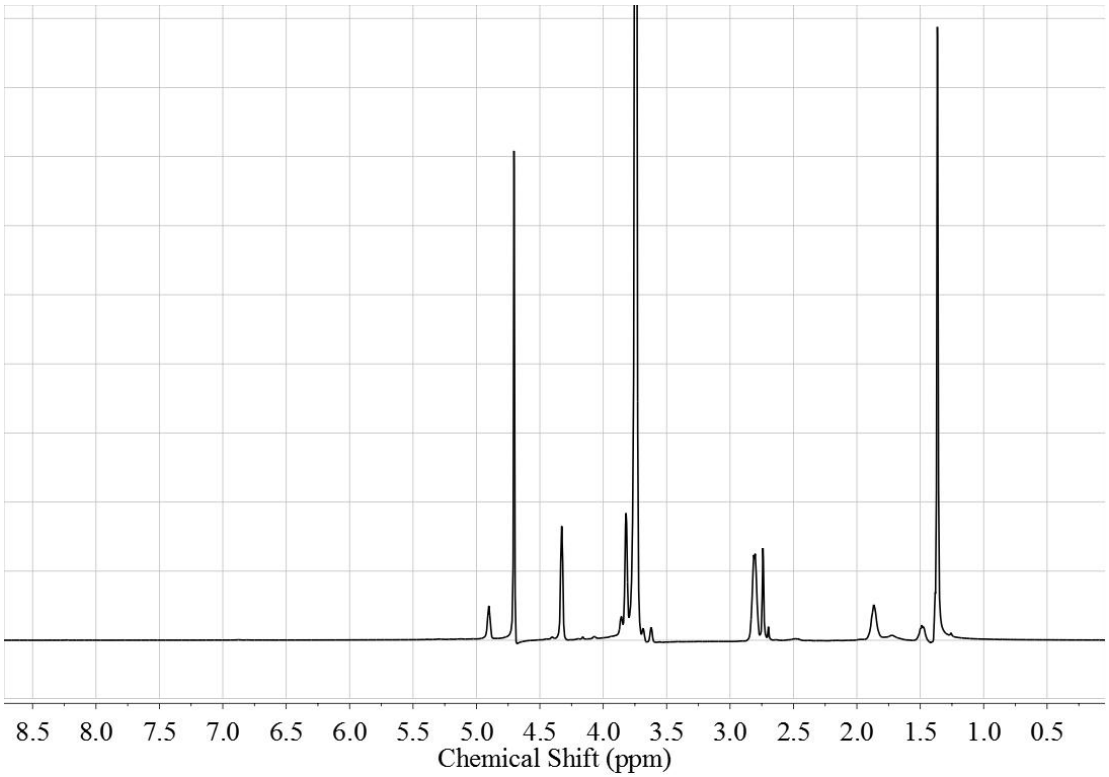


Figure S24. ^{1}H NMR spectrum of PPGBI-800 in D_2O at 35 ^{o}C

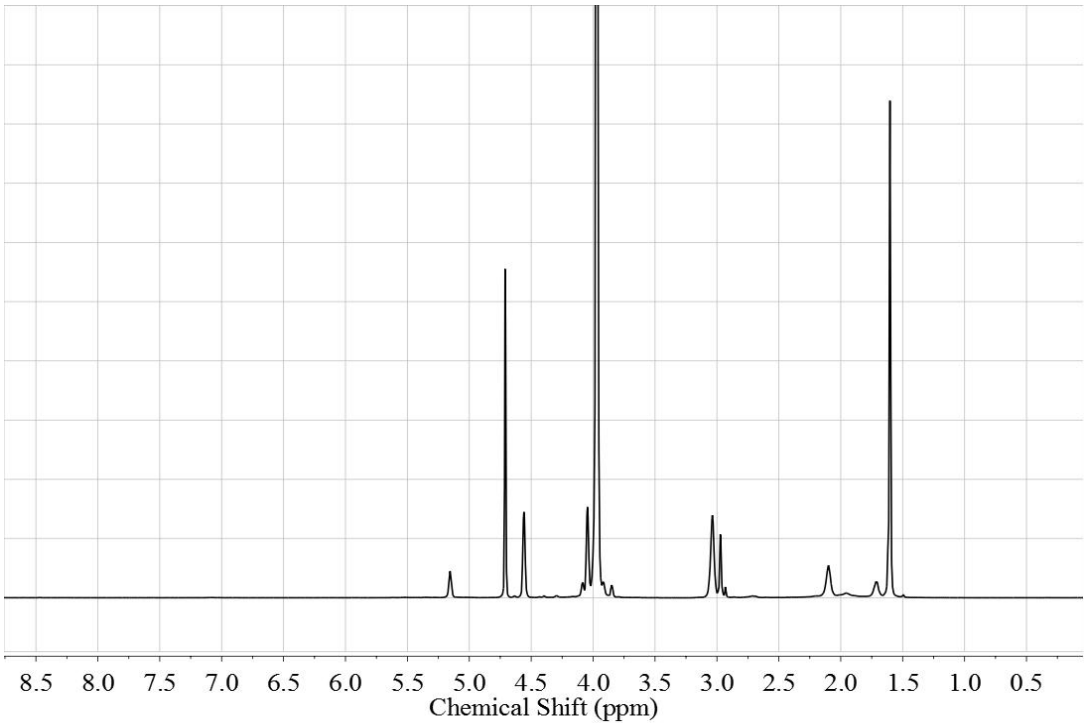


Figure S25. ^{1}H NMR spectrum of PPGBI-800 in D_2O at 55 ^{o}C

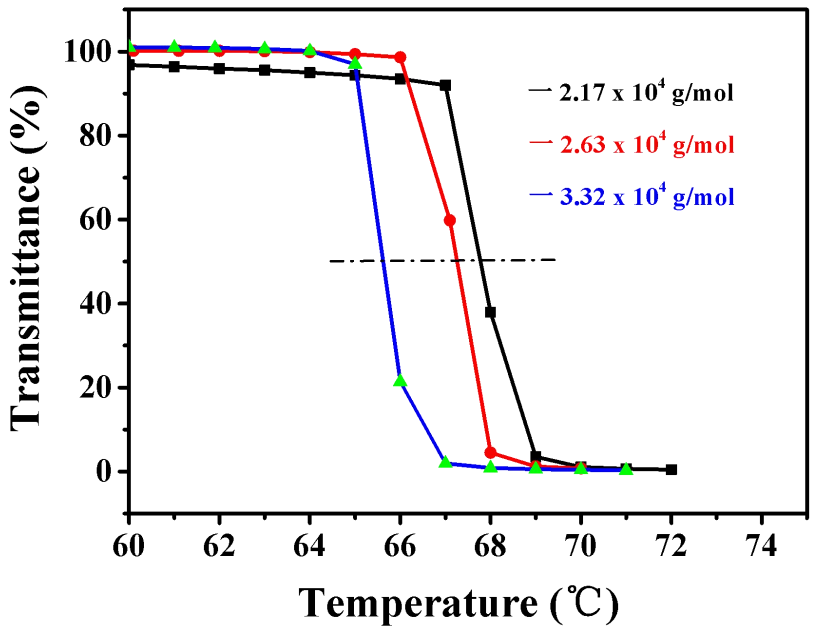


Figure S26. Thermosensitive PPPBI-800 with different M_n

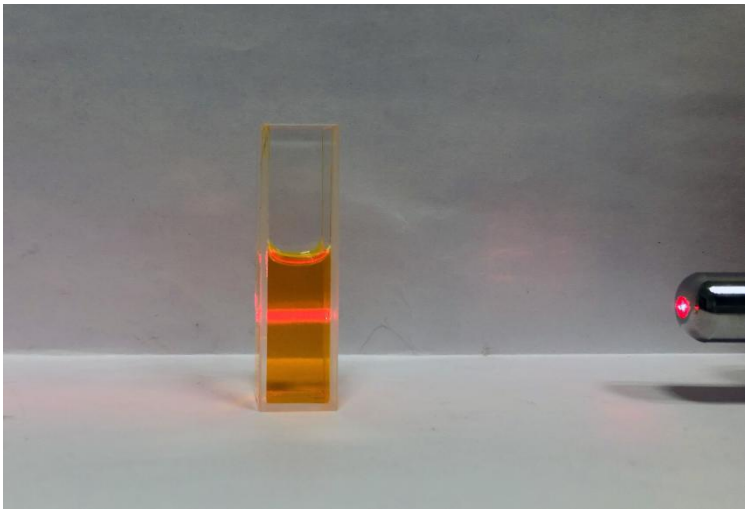


Figure S27. Tyndall effect of DOX-loaded nano-carrier system exposed to a beam of light