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Supporting Information

Dendrimersomes with Photodegradable Membranes for Triggered Release of Hydrophilic and Hydrophobic Cargo

Ali Nazemi[†] and Elizabeth R. Gillies*[,†,‡]

[†]Department of Chemistry, The University of Western Ontario, London, ON, Canada, N6G 5B7

[‡]Department of Chemical and Biochemical Engineering, The University of Western Ontario, London, ON, Canada N6A 5B9

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General Procedures and Materials

Compounds **1**[1] and **2**[2] were synthesized according to the previously published procedures. All the other chemicals were purchased from Sigma-Aldrich or Alfa Aesar and were used without further purification unless otherwise noted. Anhydrous *N,N*-dimethylformamide (DMF) was obtained from a solvent purification system using aluminum oxide columns. Dichloromethane was distilled from calcium hydride (CaH_2). Unless otherwise stated, all reactions were performed under a nitrogen atmosphere using flame or oven dried glassware. For the reactions that were stirred in dark, aluminum foil was used to isolate the reaction flasks from light. No special precautions regarding light were taken during purification or isolation of these compounds. For long term storage, the materials were stored in freezer. However no degradation issues were encountered when materials were stored in the dark at ambient temperature. ^{1}H NMR spectra were obtained at 400 MHz, and ^{13}C NMR spectra were obtained at 100 MHz. NMR chemical shifts are reported in ppm and are calibrated against the residual solvent signal of $CDCl_3$ (δ 7.26 and 77.16 ppm). Coupling constants (*J*) are expressed in Hertz (Hz). Infrared (IR) spectra were obtained as films from CH_2Cl_2 on sodium chloride (NaCl) plates using a Bruker Tensor 27 instrument. High-resolution mass spectrometry (HRMS) was performed using a PE-Sciex API 365 electrospray ionization (ESI) mass spectrometer. UV–visible (UV-vis) spectroscopy was performed on a Varian Cary 300 Bio UV–visible spectrophotometer. Dialyses were performed using Spectra/Por regenerated cellulose membranes (Spectrum Labs) with either a 2000 g/mol or 3500 g/mol molecular weight cut-off (MWCO), or 300,000 g/mol MWCO Slide-A-Lyzer cassettes (Pierce). The size exclusion chromatography (SEC) instrument was equipped with a Viscotek GPC Max VE2001 solvent module. Samples were analyzed using the Viscotek VE3580 RI detector operating at 30 °C. The separation technique employed a Polypore guard column (50 x 7.5mm) and two Agilent Polypore (300 x 7.5 mm) columns connected in series. Samples were dissolved in THF (glass distilled grade) at approximately 5 mg/mL concentrations and filtered through 0.22 μm syringe filters. Samples were injected using a 100 μL loop. The THF eluent was filtered and eluted at 1 ml/min for a total of 30 min. A calibration curve was obtained from polystyrene standards with molecular weights ranging from 1,540-1,126,000 g/mol. The light source used for the photochemical reactions was a Hanovia medium pressure mercury lamp (PC 451050/616750, 450 Wage) with an emitting wavelength of 300-400 nm and a power of 25 $mW \cdot cm^{-2}$ at 10 cm from the lamp where the samples were irradiated. DLS data were obtained using a Zetasizer NanoZS instrument from Malvern Instruments.

Fluorescence data were obtained on a QM-4 SE spectrofluorometer equipped with double excitation and emission monochromators from Photon Technologies International. In the self-assembly and photodegradation studies, organic solvents were filtered using Dynagard® polypropylene syringe filters (0.2 μm, surface area 0.8 m^2), and water was filtered using Acrodisc® syringe filters with a 0.1 Supor® membrane (0.1 μm, 25 mm diameter).

Synthesis of Protected G1 Dendrimer 3

Hydrophilic dendritic block **1** (0.55 g, 0.92 mmol, 1.0 equiv.) and monomer **2** (0.49 g, 1.4 mmol, 1.5 equiv.) were dissolved in CH_2Cl_2 (30 mL). EDC·HCl (0.40 g, 2.1 mmol, 2.2 equiv.) and DMAP (0.11 g, 0.92 mmol, 1.0 equiv.) were added in one portion and the resulting solution was stirred at room temperature in dark for 18 h. At this point, CH_2Cl_2 was removed under reduced pressure. The residue was dissolved in DMF and the product was purified by dialysis against DMF using 2000 MWCO membrane for 24 h. The solvent was then removed under reduced pressure to give dendrimer **3** (0.76 g, 0.82 mmol) in 89% yield. ^{1}H NMR ($CDCl_3$) δ: 8.76 (d, J = 4.0, 1H), 8.30 (dd, J_1 = 8.0, J_2 = 4.0, 1H), 7.85 (d, J = 8.0, 1H), 6.67 (s, 2H), 5.66 (s, 2H), 5.27 (s, 2H), 4.28 (d, J = 12.0, 2H), 4.19-4.13 (m, 6H), 3.85 (t, J = 4.0, 4H), 3.73 (t, J = 4.0, 2H), 3.75-3.69 (m, 8H), 3.68-3.62 (m, 12H), 3.56-3.52 (m, 6H), 3.37 (s, 9H), 1.47 (s, 3H), 1.40 (s, 3H), 1.17 (s, 3H). ^{13}C NMR ($CDCl_3$) δ: 173.6, 163.9, 152.7, 147.1, 138.7, 137.1, 134.3, 130.8, 130.4, 128.9, 126.1, 108.3, 98.1, 72.2, 71.8, 71.8, 70.7, 70.6, 70.4, 70.4, 69.6, 68.9, 67.6, 66.1, 63.0, 58.9, 42.3, 26.5, 20.7, 18.2. IR (cm^{-1}, film from CH_2Cl_2): 3097, 2939, 2881, 1731, 1623, 1539, 1351, 1249, 1122. HRMS: calcd $[M+Na]^+$ ($C_{44}H_{67}NO_{20}Na$): 952.4154. Found: (ESI) 952.4114. SEC data: M_n = 1200 g/mol, PDI: 1.01.

Synthesis of Deprotected G1 Dendrimer 4

Dendrimer **3** (0.26 g, 0.28 mmol) was dissolved in methanol (25 mL) and CH_2Cl_2 (5 mL). Concentrated sulfuric acid (0.30 mL) was added and the resulting solution was stirred at room temperature in dark for 2 h. At this point, ^{1}H NMR showed the completion of the reaction. The solution was diluted by the addition of CH_2Cl_2 (50 mL). It was then washed with 1M Na_2CO_3 (2×100 mL) and brine (1×100 mL). The organic layer was dried over $MgSO_4$, filtered, and concentrated under reduced pressure to give dendrimer **4** (0.23 g, 0.26 mmol) in 93% yield. ^{1}H NMR ($CDCl_3$) δ: 8.75 (d, J = 4.0, 1H), 8.31 (dd, J_1 = 8.0, J_2 = 4.0, 1H), 7.83 (d, J = 8.0, 1H), 6.68 (s, 2H), 5.65 (s, 2H), 5.27 (s, 2H), 4.18-4.14 (m, 6H), 3.98 (d,

J = 8.0, 2H), 3.85 (t, J = 4.0, 4H), 3.80-3.77 (m, 4H), 3.73-3.70 (m, 6H), 3.66-3.62 (m, 12H), 3.55-3.52 (m, 6H), 3.36 (s, 9H), 2.81 (br s, 2H), 1.12 (s, 3H). ^{13}C NMR ($CDCl_3$) δ: 174.8, 163.8, 152.6, 147.1, 138.5, 136.8, 134.3, 130.8, 130.4, 129.0, 125.9, 108.2, 72.2, 71.7, 70.6, 70.5, 70.4, 70.3, 69.6, 68.8, 67.6, 66.9, 62.8, 58.8, 49.6, 17.1. IR (cm^{-1}, film from CH_2Cl_2): 3415, 3089, 2906, 2879, 1730, 1623, 1535, 1350, 1253, 1112. HRMS: calcd $[M+Na]^+$ ($C_{41}H_{63}NO_{20}Na$): 912.3841. Found: (ESI) 912.3821. SEC data: M_n = 1200 g/mol, PDI: 1.01.

Synthesis of Protected G2 Dendron 5

Dendrimer **4** (0.21 g, 0.24 mmol, 1.0 equiv.) and monomer **2** (0.26 g, 0.72 mmol, 3.0 equiv.) were dissolved in CH_2Cl_2 (45 mL). EDC· HCl (0.21 g, 1.1 mmol, 4.5 equiv.) and DMAP (59 mg, 0.48 mmol, 2.0 equiv.) were added in one portion and the resulting solution was stirred at room temperature in dark for 18 h. At this point, CH_2Cl_2 was removed under reduced pressure. The residue was dissolved in DMF and product was purified by dialysis against DMF using 3500 MWCO membrane for 24 h. The solvent was then removed under reduced pressure to give dendrimer **5** (0.31 g, 0.20 mmol) in 83% yield. ^{1}H NMR ($CDCl_3$) δ: 8.64-8.58 (m, 3H), 8.23-8.16 (m, 3H), 7.85 (d, J = 8.0, 2H), 7.63 (d, J = 8.0, 1H), 6.68 (s, 2H), 5.65 (s, 4H), 5.60 (s, 2H), 5.26 (s, 2H), 4.67-4.59 (m, 4H), 4.28 (d, J = 12.0, 4H), 4.19-4.13 (m, 6H), 3.85 (t, J = 4.0, 4H), 3.73 (t, J = 4.0, 2H), 3.75-3.69 (m, 10H), 3.67-3.61 (m, 12H), 3.56-3.51 (m, 6H), 3.36 (s, 9H), 1.49 (s, 3H), 1.47 (s, 6H), 1.39 (s, 6H), 1.18 (s, 6H). ^{13}C NMR ($CDCl_3$) δ: 173.6, 171.5, 163.6, 163.4, 152.7, 147.8, 147.1, 138.7, 137.6, 135.0, 134.2, 131.6, 130.4, 130.0, 129.0, 129.0, 126.1, 126.0, 126.0, 108.4, 98.2, 72.3, 71.9, 71.8, 70.7, 70.6, 70.4, 69.6, 68.9, 67.8, 66.4, 66.1, 63.8, 63.0, 58.9, 47.0, 42.4, 26.6, 20.6, 18.2, 17.9. IR (cm^{-1}, film from CH_2Cl_2): 3091, 2918, 2875, 1733, 1623, 1535, 1348, 1249, 1116. HRMS: calcd $[M+Na]^+$ ($C_{73}H_{97}N_3O_{34}Na$): 1582.5851. Found: (ESI) 1582.5857. SEC data: M_n = 1900 g/mol, PDI: 1.01.

Synthesis of Deprotected G2 Dendrimer 6

Dendrimer **5** (0.20 g, 0.13 mmol) was dissolved in methanol (50 mL) and CH_2Cl_2 (10 mL). Concentrated sulfuric acid (0.60 mL) was added and the resulting solution was stirred at room temperature in dark for 2 hrs. At this point, ^{1}H NMR showed the completion of the reaction. The solution was diluted by the addition of CH_2Cl_2 (50 mL). It was then washed with 1M Na_2CO_3 (2×100 mL) and brine (1×100 mL). The organic layer was dried over $MgSO_4$, filtered, and concentrated under reduced pressure to give dendrimer **6** (0.18 g, 0.12

mmol) in 95% yield. ^{1}H NMR ($CDCl_3$) δ: 8.60-8.51 (m, 3H), 8.20- 8.09 (m ,3H), 7.79 (d, *J* = 8.0, 2H), 7.62 (d, *J* = 8.0, 1H), 6.68 (s, 2H), 5.61 (s, 6H), 5.25 (s, 2H), 4.63 (s, 4H), 4.21-4.12 (m, 6H), 4.02-3.94 (m, 4H), 3.85 (t, *J* = 4.0, 4H), 3.83-3.76 (m, 6H), 3.75-3.69 (m, 6H), 3.68-3.61 (m, 12H), 3.56-3.51 (m, 6H), 3.36 (s, 9H), 2.98 (t, *J* = 8.0, 4H), 1.49 (s, 3H), 1.13 (s, 6H). ^{13}C NMR ($CDCl_3$) δ: 175.0, 171.7, 163.8, 163.5, 152.7, 147.8, 147.1, 138.6, 137.4, 135.2, 134.2, 134.2, 131.5, 130.4, 130.2, 130.0, 129.2, 126.1, 125.9, 108.4, 72.3, 71.9 (2), 70.7, 70.6, 70.4, 69.7, 68.9, 67.9, 67.3, 67.3, 66.8, 63.8, 62.9, 59.0, 49.7, 47.0, 18.1, 17.2. IR (cm^{-1}, film from CH_2Cl_2): 3394, 3082, 2927, 2883, 1726, 1625, 1537, 1350, 1255, 1114. HRMS: calcd $[M+Na]^+$ ($C_{67}H_{89}N_3O_{34}Na$): 1502.5225. Found: (ESI) 1502.5205. SEC data: M_n = 1900 g/mol, PDI: 1.02.

Synthesis of Protected G3 Dendrimer 7

Dendrimer **6** (0.14 g, 0.10 mmol, 1.0 equiv.) and monomer **2** (0.21 g, 0.60 mmol, 6.0 equiv.) were dissolved in CH_2Cl_2 (35 mL). EDC· HCl (0.17 g, 0.90 mmol, 9.0 equiv.) and DMAP (49 mg, 0.40 mmol, 4.0 equiv.) were added in one portion and the resulting solution was stirred at room temperature in dark for 18 h. At this point, CH_2Cl_2 was removed under reduced pressure. The residue was dissolved in DMF and product was purified by dialysis against DMF using 3500 MWCO membrane for 24 h. The solvent was then removed under reduced pressure to give dendrimer **7** (0.25 g, 90 μmol) in 90% yield. ^{1}H NMR ($CDCl_3$) δ: 8.61-8.57 (m, 5H), 8.51-8.48 (m ,2H), 8.22-8.16 (m, 5H), 8.12-8.08 (m, 2H), 7.85 (d, *J* = 8.0, 4H), 7.65 (d, *J* = 8.0, 3H), 6.67 (s, 2H), 5.64 (s, 8H), 5.61 (s, 4H), 5.59 (s, 2H), 5.25 (s, 2H), 4.67-4.58 (m, 12H), 4.27 (d, *J* = 12.0, 8H), 4.20-4.12 (m, 6H), 3.85 (t, *J* = 4.0, 4H), 3.73 (t, *J* = 4.0, 2H), 3.75-3.68 (m, 14H), 3.67-3.61 (m, 12H), 3.56-3.51 (m, 6H), 3.36 (s, 9H), 1.50 (s, 6H), 1.48 (s, 3H), 1.46 (s, 12H), 1.38 (s, 12H), 1.17 (s, 12H). ^{13}C NMR ($CDCl_3$) δ: 173.7, 171.6, 171.6, 163.8, 163.6, 163.3, 152.8, 147.9, 147.8, 147.2, 138.9, 137.7, 135.7, 135.2, 134.3, 134.1, 131.7, 130.9, 130.4, 130.3, 130.1, 129.1, 126.2, 126.1, 126.0, 114.8, 108.5, 98.3, 72.4, 72.0 (2), 70.8, 70.7, 70.6, 70.5, 69.8, 69.0, 67.9, 66.6, 66.5, 66.2, 64.0, 63.9, 63.9, 63.1, 59.0, 47.1, 47.0, 42.5, 26.7, 20.7, 18.3, 18.1, 18.0. IR (cm^{-1}, film from CH_2Cl_2): 3084, 2927, 2879, 1731, 1623, 1539, 1346, 1255, 1118. HRMS: calcd $[M+Na]^+$ ($C_{131}H_{157}N_7O_{62}Na$): 2842.9251. Found: (ESI) 2842.9245. SEC data: M_n = 3200 g/mol, PDI: 1.01.

Procedures for the Preparation of Dendrimer Assemblies

Dendrimer assemblies were prepared by first preparing an 8 mg/mL solution of each dendrimer in DMSO. The solution was stirred for 2 h to ensure complete dissolution and was then filtered through a 0.2 μm syringe filter. Next, either 0.1 mL of the DMSO solution was rapidly injected into 0.9 mL of filtered deionized water with rapid stirring or 0.9 mL of filtered, deionized water was added to 0.1 mL of the DMSO solution with rapid stirring over a period of ~ 1 min. The resulting suspension was then dialyzed against deionized water using a 3500 Da MWCO membrane. The same procedure was also performed using THF in place of DMSO.

In the case of dendrimer **7**, an emulsion procedure was also investigated. A solution of dendrimer **7** in CH_2Cl_2 (10 mg/mL, 0.1 mL) was added to water (2 mL) followed by stirring overnight uncapped to evaporate the CH_2Cl_2.

Transmission Electron Microscopy

The suspension of the dendrimer assemblies (5 μL, 0.05 mg/mL) was placed on a carbon formvar grid and was left to dry in air in dark overnight. Imaging was performed using a Phillips CM10 microscope operating at 80 kV with a 40 μm aperture.

General Procedure for Monitoring Dendrimer Degradation by UV-vis Spectroscopy

The sample of **7** was prepared at the concentration of 30 μg/mL in either spectroscopic grade THF or as dendrimersomes (prepared as described above via the procedure involving the addition of water into DMSO) in deionized water. 3 mL of solution was transferred to a quartz cuvette and irradiated with UV light for 30 min. UV-vis spectra were collected every 2 min.

General Procedure for Monitoring Dendrimersome Degradation by DLS

A dendrimersome sample from **7** was prepared as described above via the procedure involving the addition of water into DMSO, and was diluted to a concentration of 0.1 mg/mL. 2 mL of the sample was transferred to a quartz cuvette and irradiated with UV light for 8 h. DLS measurements were performed in the same cuvette at various time points. For each time point three measurements each for 150 s were performed. Mean count rates reported in Figure 2b are the average values obtained for each time point. Parameters including the attenuator and measurement position were kept constant throughout the study.

Procedure for the Encapsulation and Release of Nile Red from Dendrimersomes

To the solution of dendrimer **7** in DMSO (8 mg/mL) was added Nile Red (2.5 mg/mL, 2% w/w). This solution was stirred in dark for 2 h and then filtered through 0.2 μm syringe filter. To 0.1 mL of this solution was added deionized water (0.9 mL filtered through 0.1 μm syringe filter) with rapid stirring during a course of about 1 min. This sample was then stirred in dark for 1 h and then dialyzed against deionized water to remove DMSO using 3500 MWCO membrane for 24 h.

For the triggered release study, to 0.7 mL of this sample was diluted with deionized water (0.8 mL filtered through 0.1 μm syringe filter). The resulting sample was then transferred to a quartz cuvette and irradiated with UV light. Fluorescence emission spectra were collected every minute using an excitation wavelength of 540 nm and emission range of 550 - 750 nm. The maximum emission wavelength of 620 nm was then used to calculate the percent release from dendrimersomes.

For the control experiment, the same procedure was followed without irradiating the sample with UV light.

Procedure for the Encapsulation and Release of Fluorescein from Dendrimersomes

To a solution of **7** in DMSO (0.1 mL, 8 mg/mL) was added a solution of fluorescein in water (0.9 mL, 0.15 mM filtered through 0.1 μm syringe filter) with stirring during a course of about 1 min. This sample was left to equilibrate at room temperature and in dark for 24 h and was then dialyzed against deionized water (1 L with two changes) for 9 h using 300,000 g/mol MWCO Slide-A-Lyzer cassette (Pierce) to remove the non-encapsulated fluorescein. The time required for the removal of free dye using the Slide-A-Lyzer was determined in a control experiment using the same concentration of fluorescein without dendrimersomes.

For the triggered release study, this sample was transferred to a quartz cuvette and irradiated for 10 min. After irradiation, it was placed back in the Slide-A-Lyzer cassette and was dialyzed against deionized water (200 mL). At various time points, 2 mL aliquots of the dialysate were removed for fluorescence measurements and then returned to the dialysate. For fluorescence measurements, the solutions were excited at 494 nm and the emission spectra were recorded in the range of 450-600 nm. The maximum emission wavelength of 510 nm was used for the calculation of percent release. After the emission intensity of the dialysate reached a plateau, to ensure the complete release of the dye, the fluorescent intensity of the sample inside the dialysis cassette was measured and no significant fluorescence was observed. The fluorescence intensity of the dialysate at the final time point

was therefore set as 100% release and the percent release at the other time points was calculated accordingly.

For the control experiment, the same procedure was followed, except the sample was not irradiated.

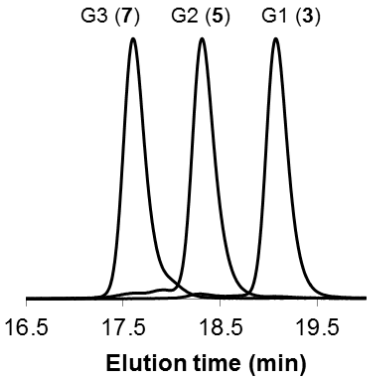

Figure S1. SEC traces for dendrimers **3**, **5**, and **7**.

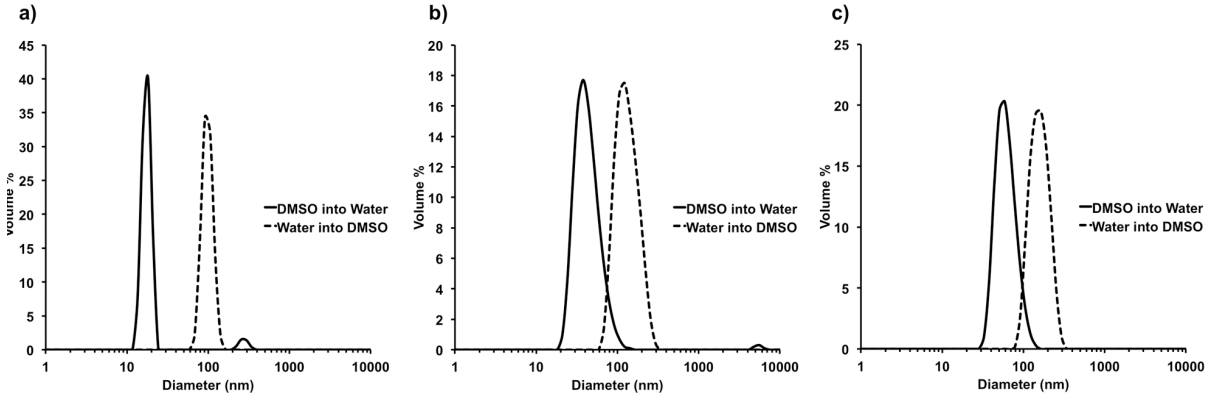

Figure S2. Additional dynamic light scattering data for assemblies formed from: a) G1 dendrimer **3**; b) G2 dendrimer **5**; c) G3 dendrimer **7**.

Table S1. Summary of DLS results for the self-assembly of dendrimers **3**, **5**, and **7**.

	DMSO into H_2O		H_2O into DMSO	
	Z-Average (nm)	PDI	Z-Average (nm)	PDI
G1 (**3**)	-[a]	-	192	0.27
G2 (**5**)	59	0.28	136	0.06
G3 (**7**)	70	0.05	158	0.02

[a]No Z-average could be calculated for this sample due to insufficient data quality due to the presence of two populations of assemblies (Figure S2a).

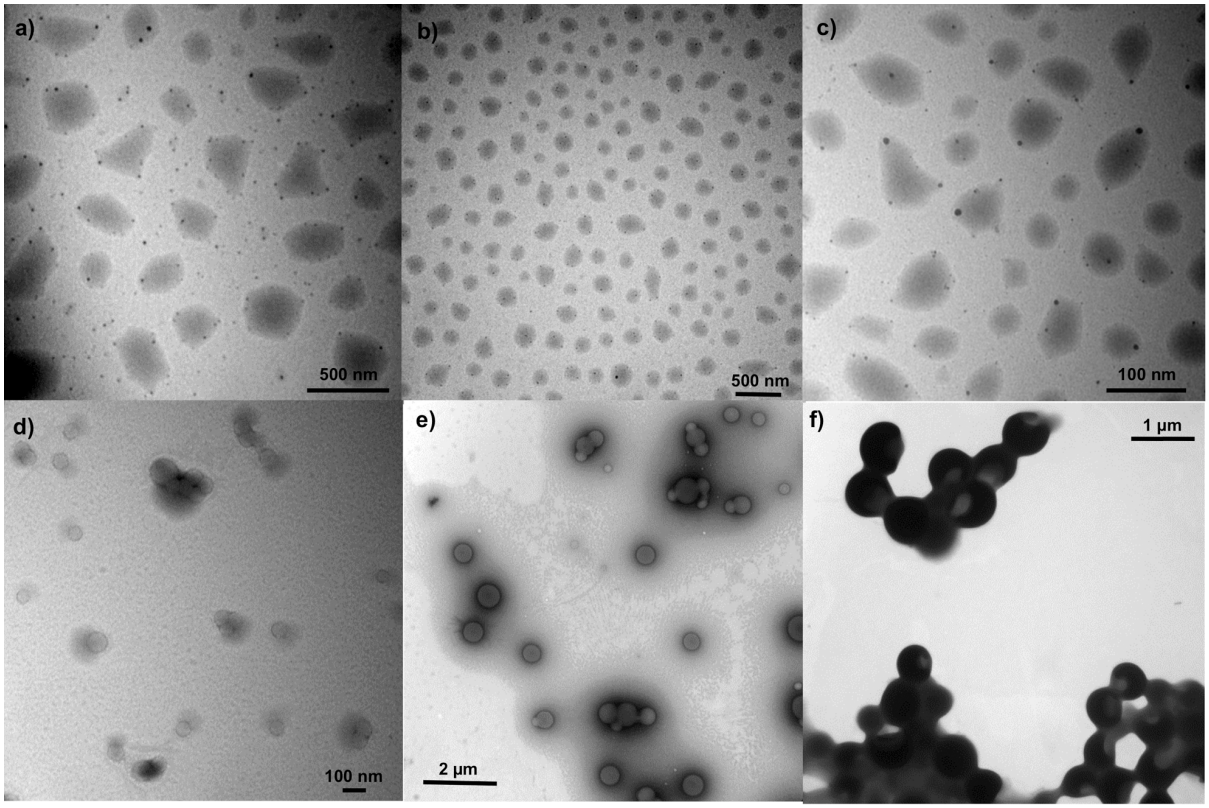

Figure S3. Additional TEM images of assemblies: a) G1 via DMSO into water; b) G1 via water into DMSO; c) G2 via DMSO into water; d) G3 via DMSO into water; e) G3 via water into THF; f) G3 via CH_2Cl_2 in water.

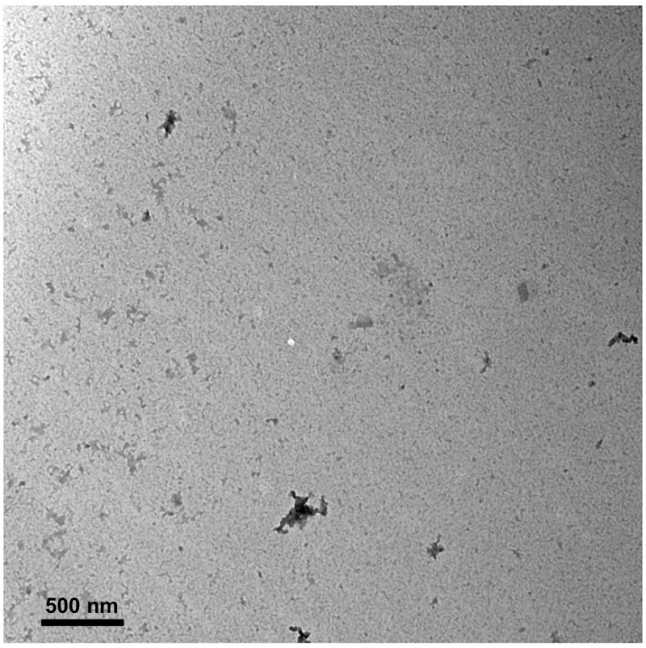

Figure S4. TEM image of dendrimersomes from Figure 1d, following 8 h of UV irradiation, showing that no dendrimersomes were present.

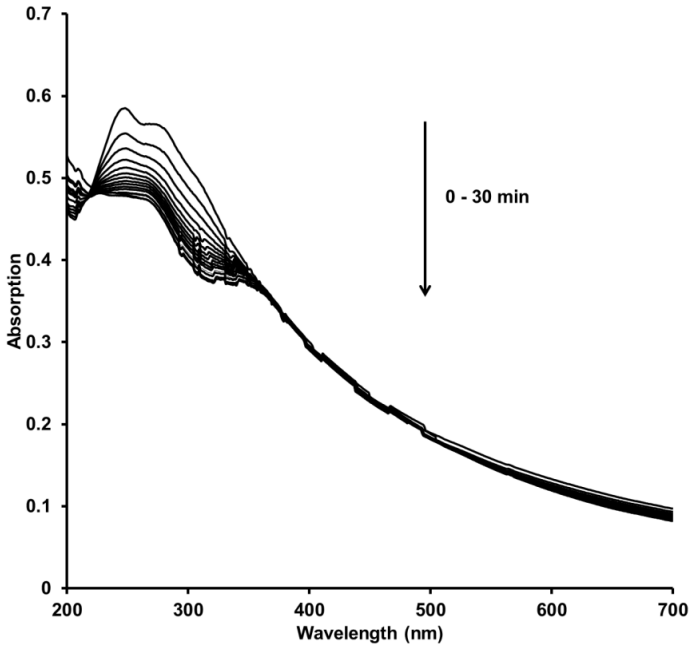

Figure S5. UV-vis spectra for dendrimersomes in water upon irradiation with UV light for 30 min.

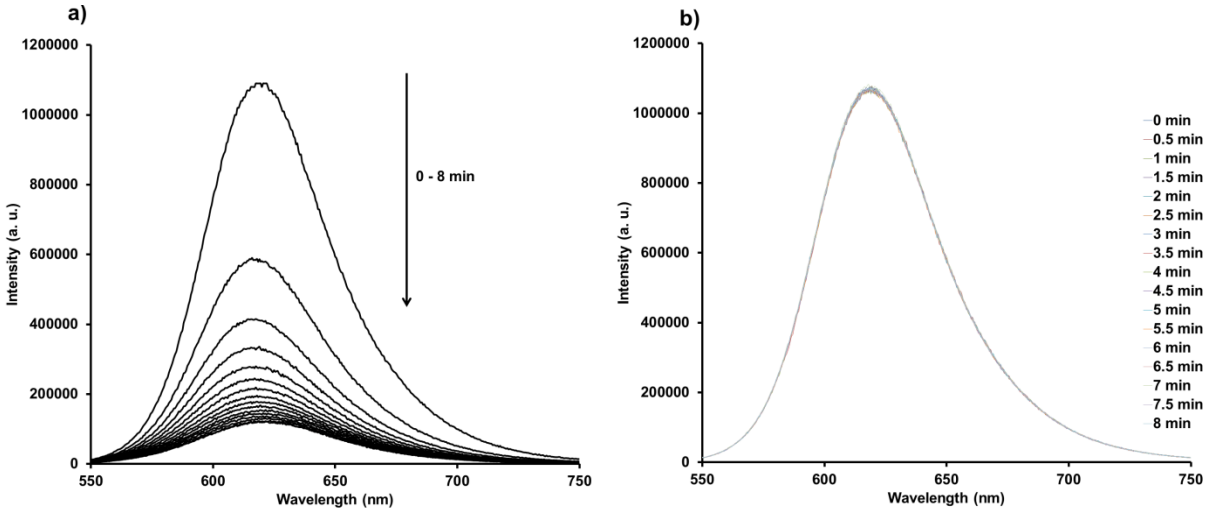

Figure S6. Fluorescence spectra of nile red loaded into the dendrimersome membrane following a) different irradiation periods with UV light and b) kept in dark.

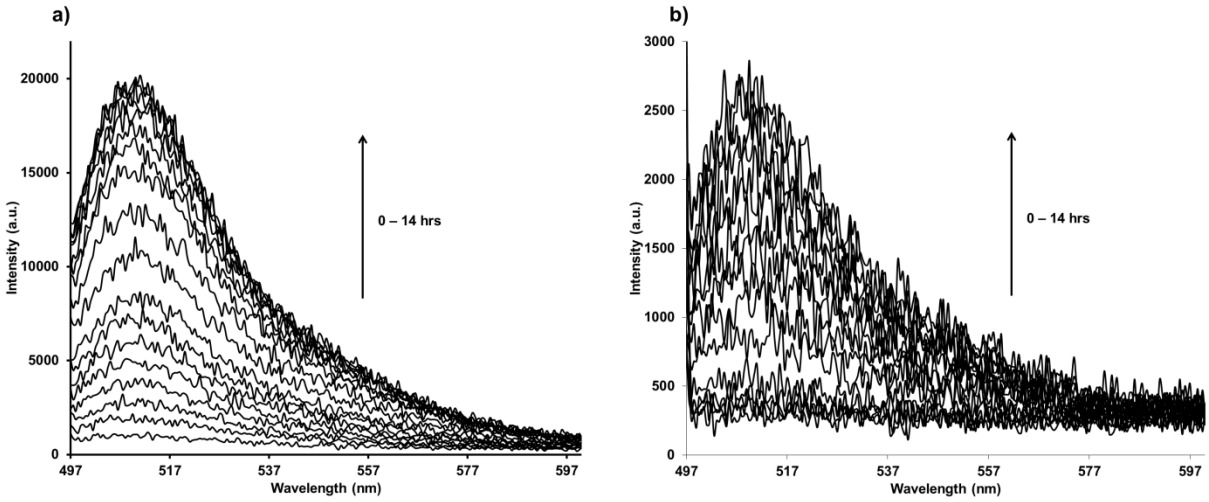

Figure S7. Fluorescence spectra of fluorescein in the dialysate following its encapsulation into dendrimersomes, followed by release (from dendrimersomes in a dialysis cassette) after: a) 10 min irradiation with UV light and b) storage in the dark.

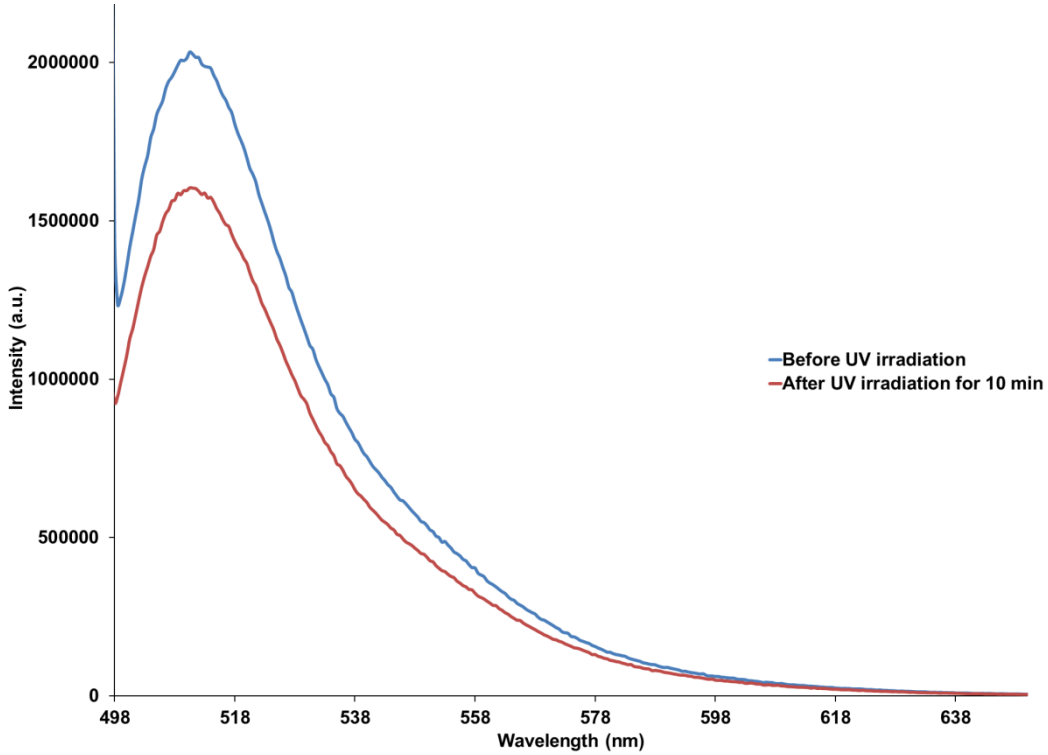

Figure S8. Fluorescent spectra of fluorescein before and after irradiation with UV light for 10 min.

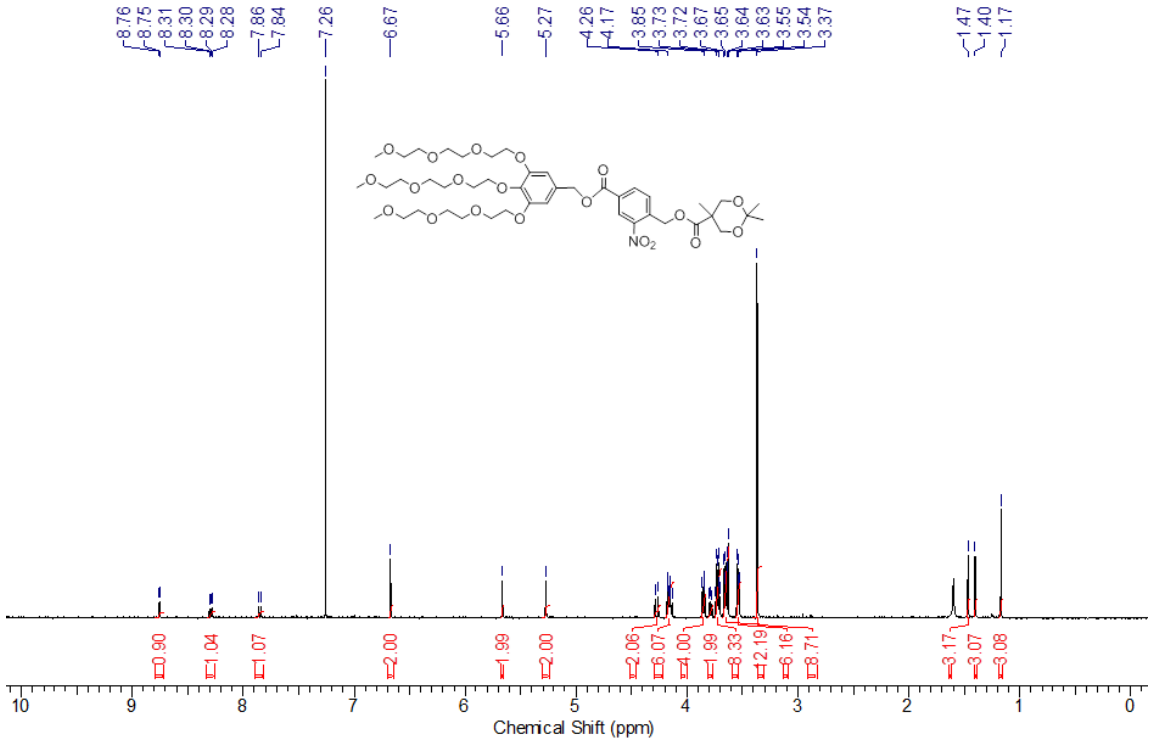

Figure S9. ^{1}H NMR spectrum of compound **3** (400 MHz, $CDCl_3$).

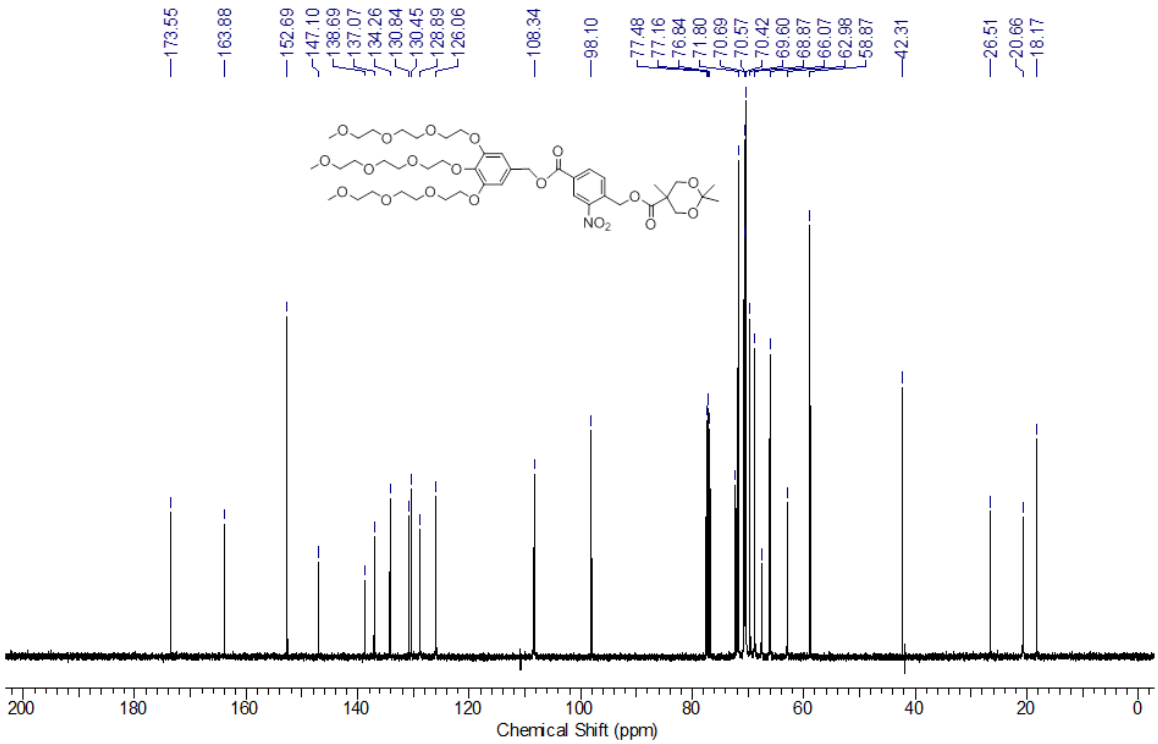

Figure S10. ^{13}C NMR spectrum of compound **3** (100 MHz, $CDCl_3$).

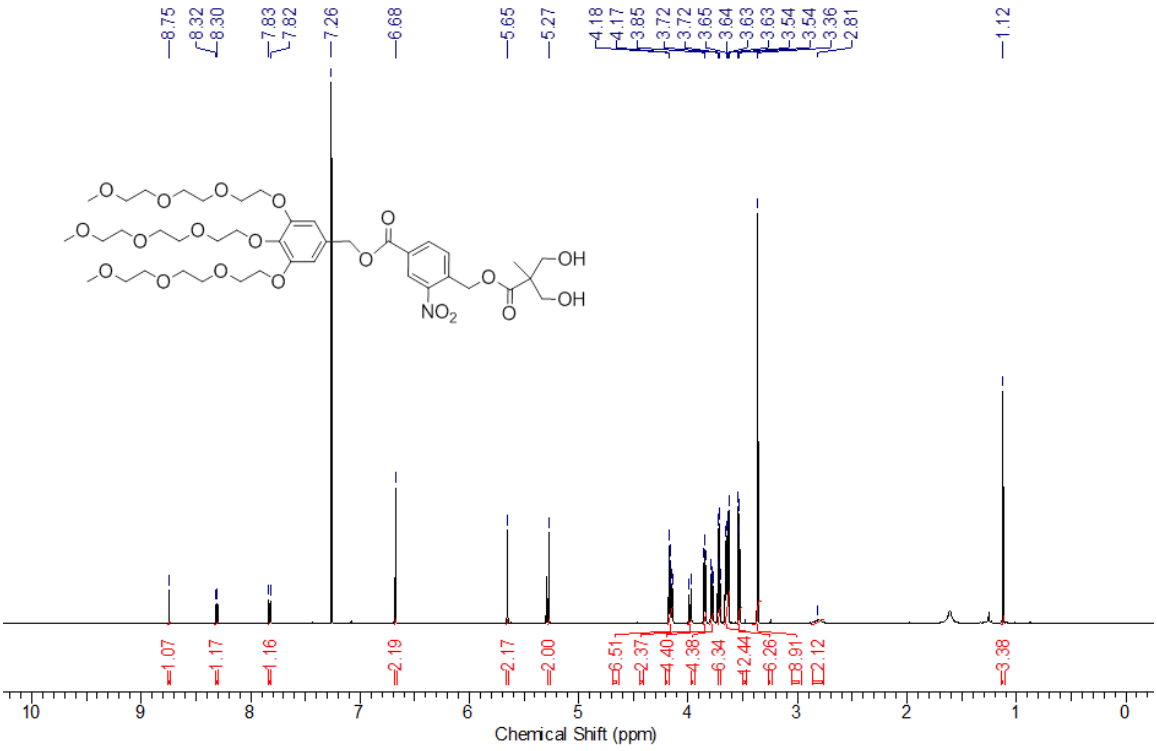

Figure S11. ^{1}H NMR spectrum of compound **4** (400 MHz, $CDCl_3$).

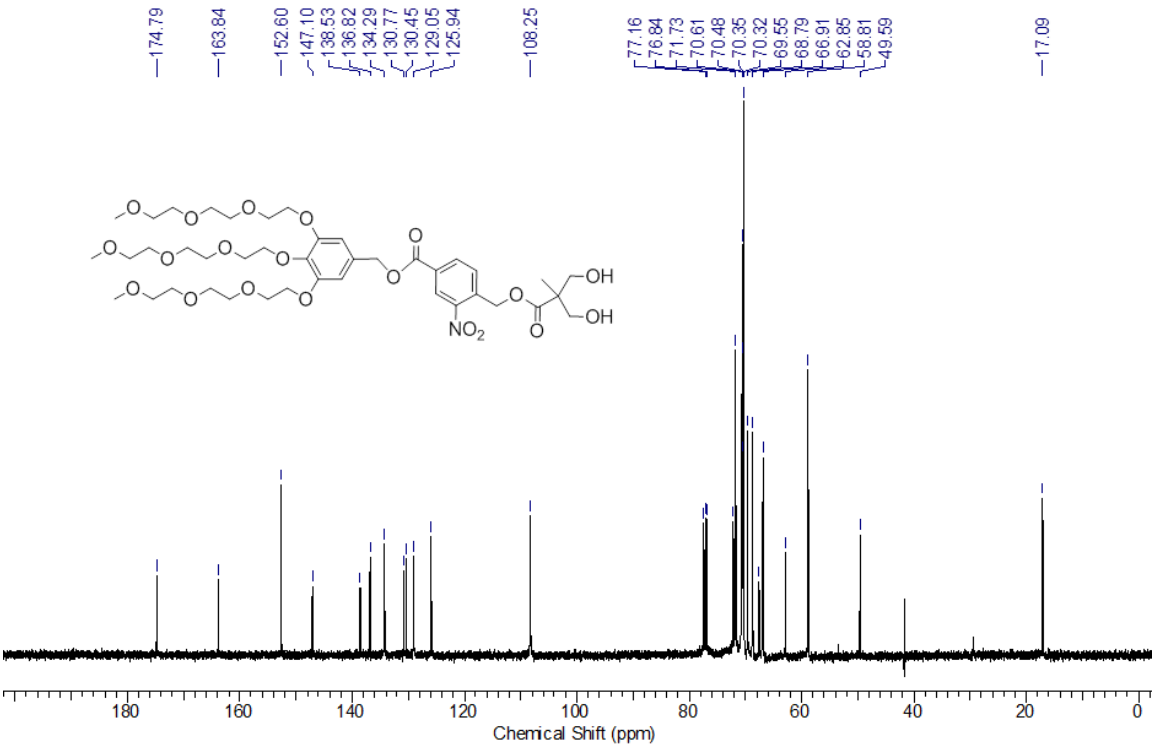

Figure S12. ^{13}C NMR spectrum of compound **4** (100 MHz, $CDCl_3$).

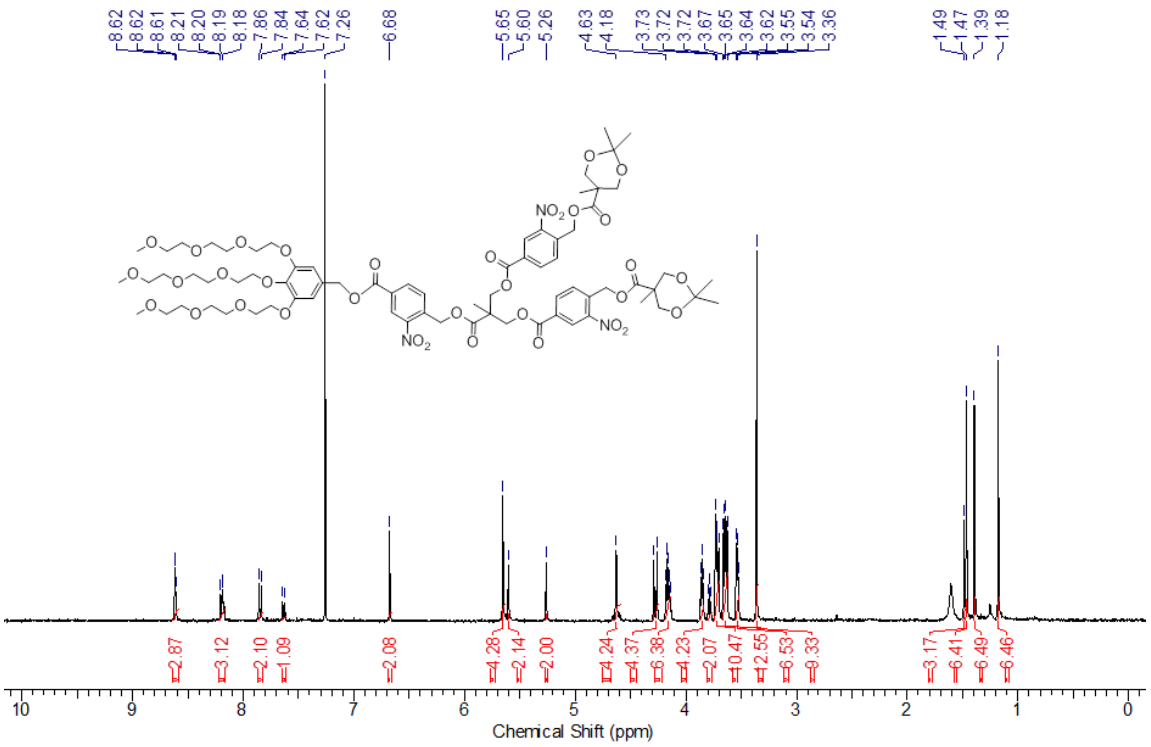

Figure S13. ^{1}H NMR spectrum of compound **5** (400 MHz, $CDCl_3$).

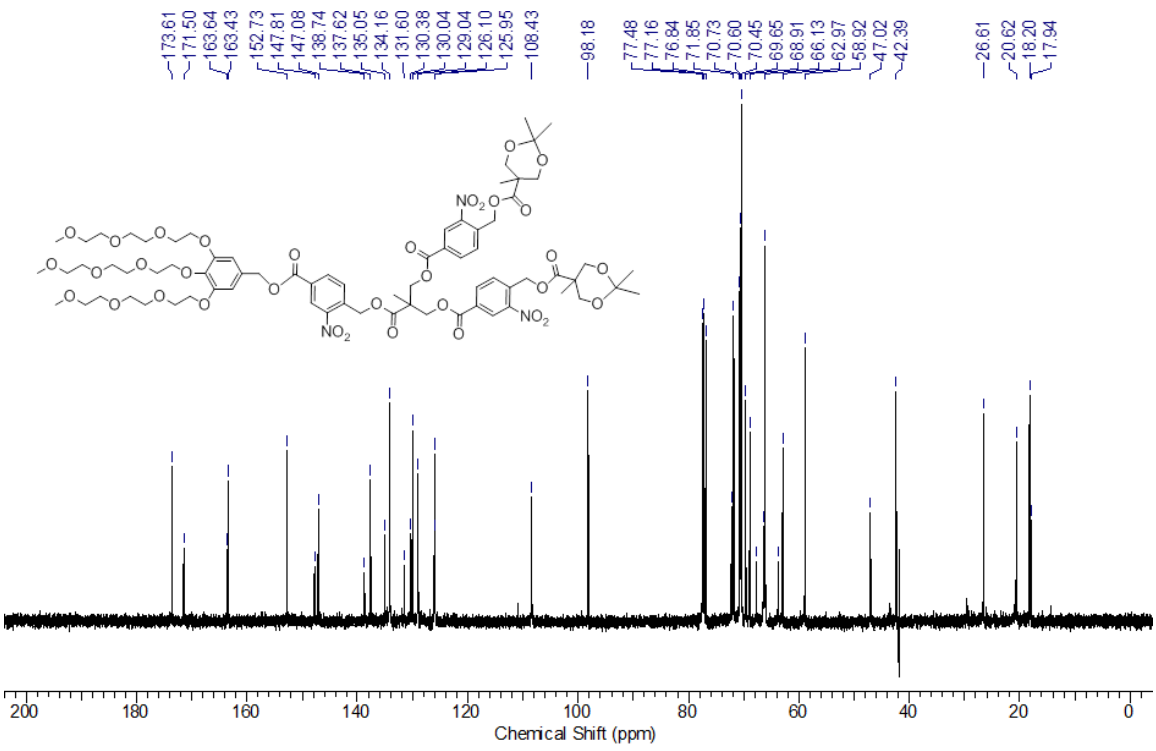

Figure S14. ^{13}C NMR spectrum of compound **5** (100 MHz, $CDCl_3$).

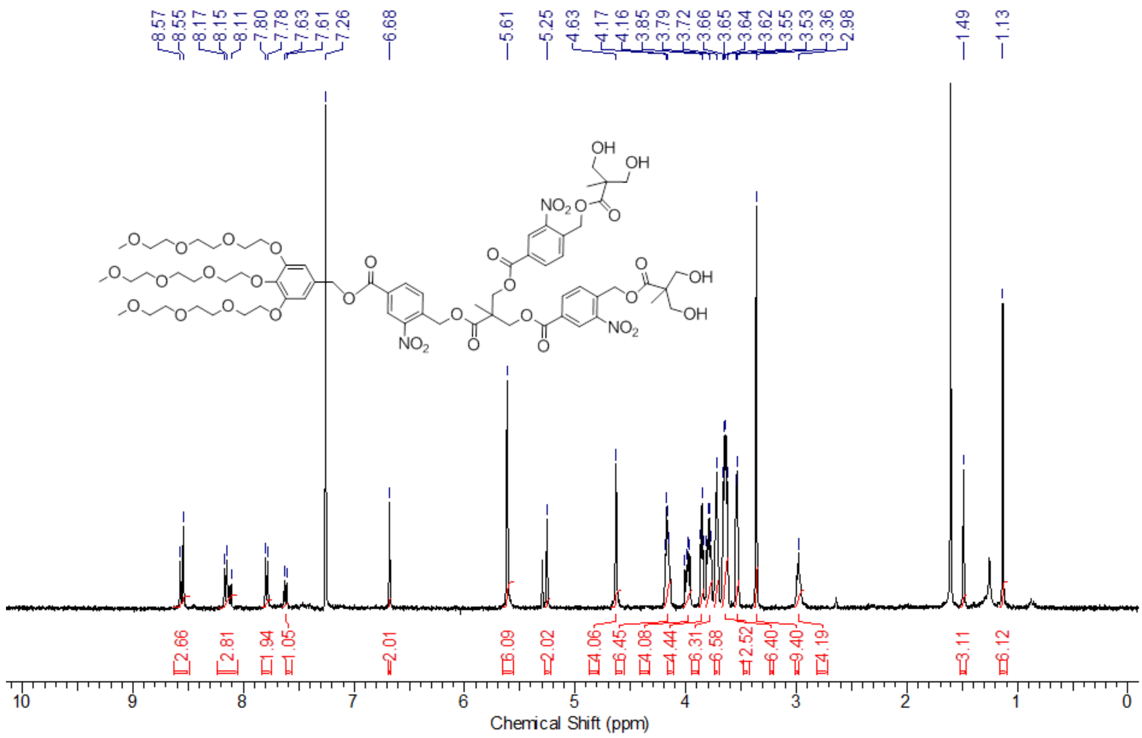

Figure S15. ^{1}H NMR spectrum of compound **6** (400 MHz, $CDCl_3$).

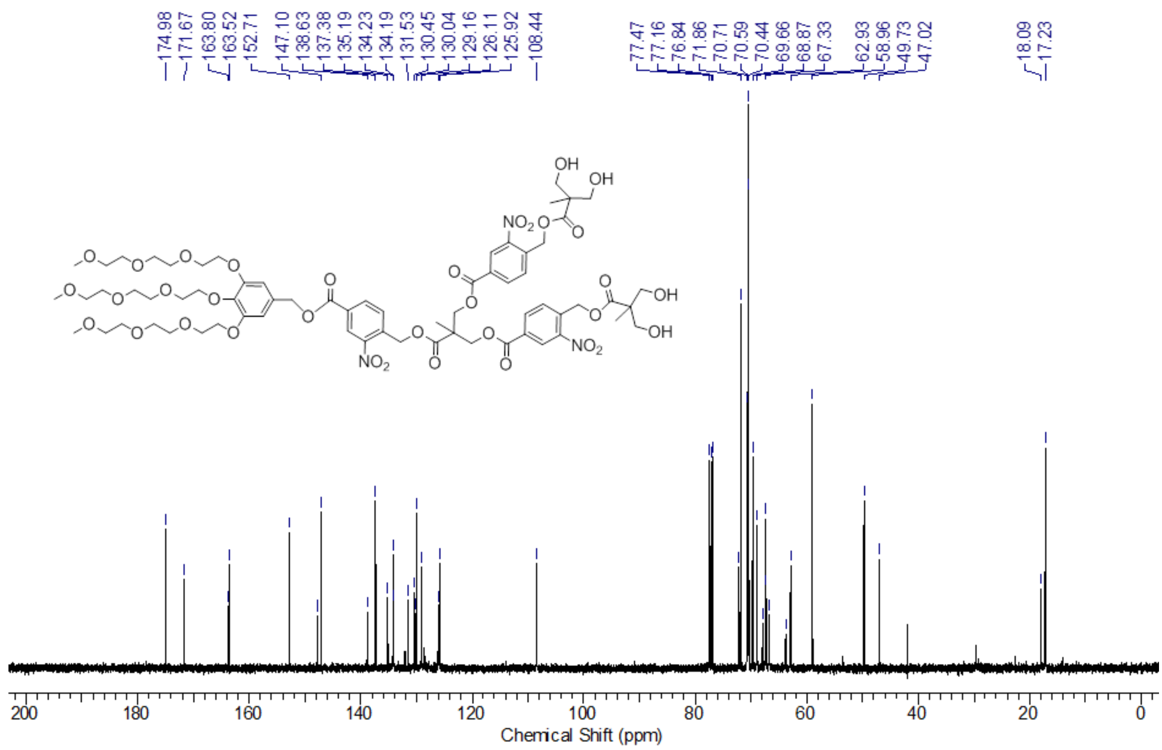

Figure S16. ^{13}C NMR spectrum of compound **6** (100 MHz, $CDCl_3$).

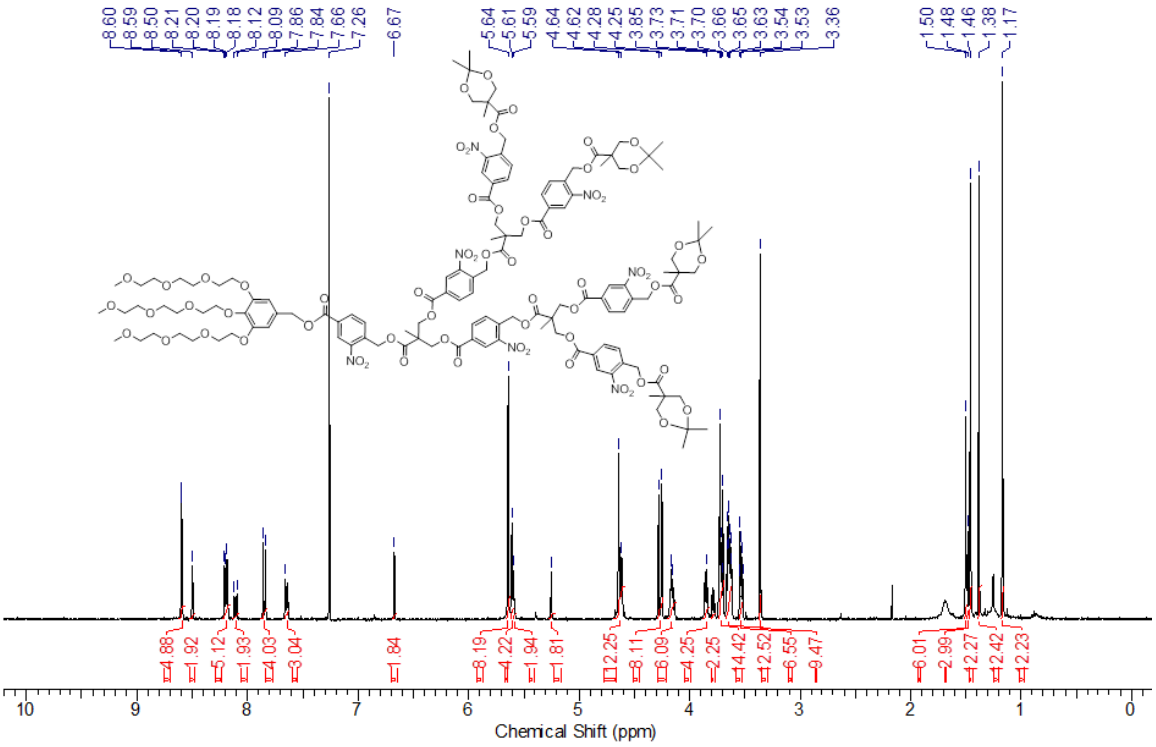

Figure S17. ^{1}H NMR spectrum of compound **7** (400 MHz, $CDCl_3$).

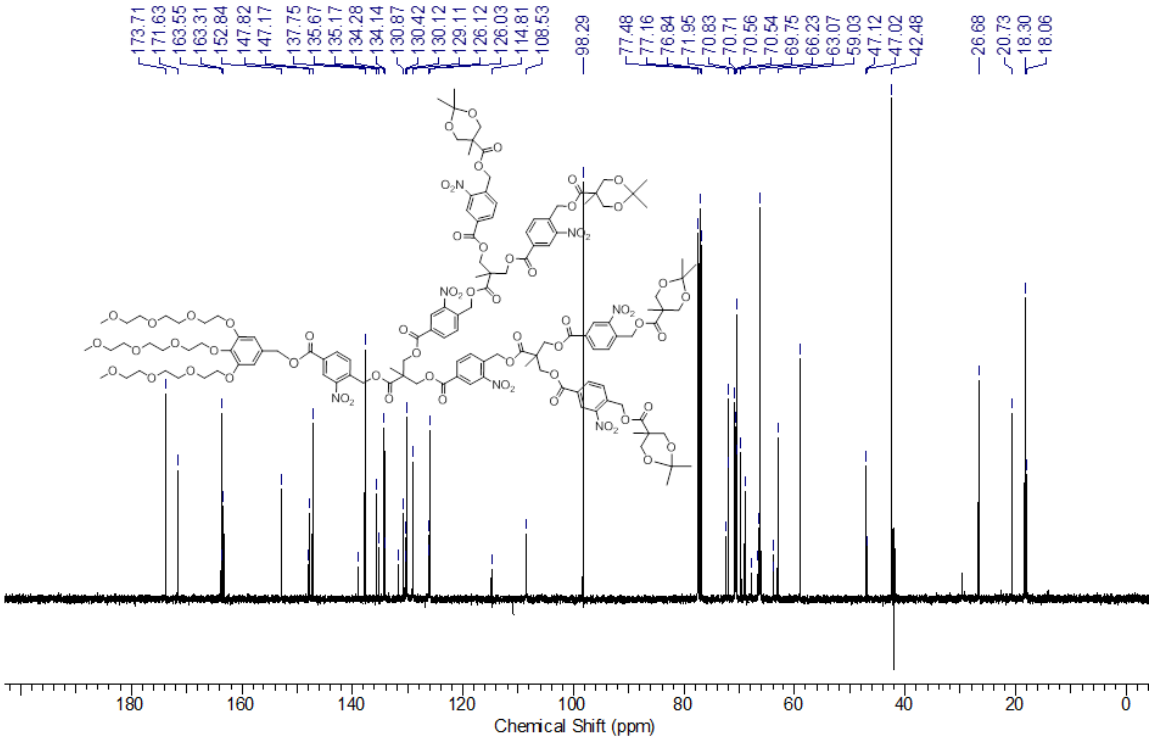

Figure S18. ^{13}C NMR spectrum of compound **7** (100 MHz, $CDCl_3$).

References

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