Supplementary Information

Layered Supramolecular Hydrogels from Thioglycosides

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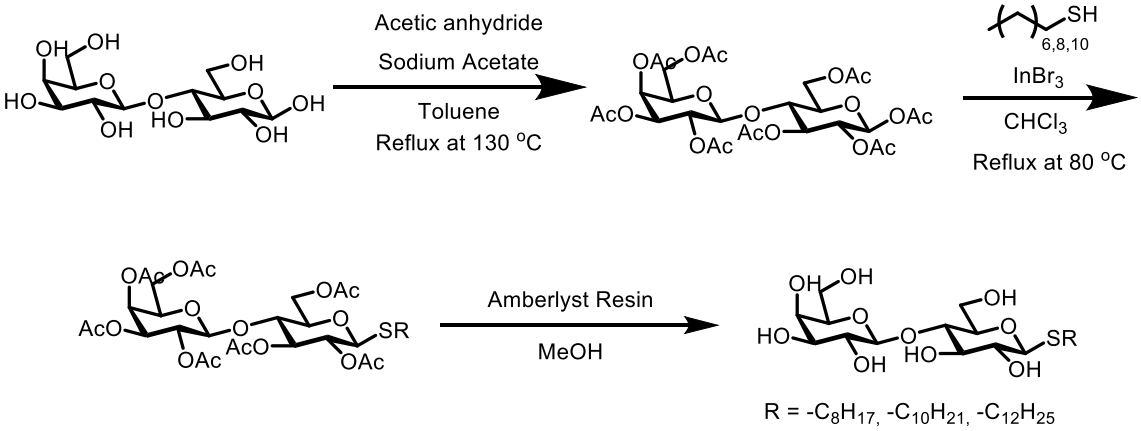
Contents

I. Synthesis of LacSCx and CelSCx

The synthesis of the thiolactosides and thiocellobiosides follows that reported by Szabo et al. as shown in Scheme S1.[1] The general synthetic protocol is as follows.



Scheme S1. General scheme for thioglcolipid synthesis. Chemical structures shown in the scheme are thiolactosides; thiocellobiosides can be synthesized using same procedures.

Acetylation. To an oven-dried round button flask equipped with a stir bar were added sugar (1 eq) and sodium acetate (0.37 eq). The round button flask was evacuated and refilled with dry nitrogen. Anhydrous toluene (20 mL/mmol of sugar) and acetic anhydride (1.7 eq/OH of sugar) were then added via syringe. The contents of the round button flask were stirred and refluxed at 120 °C for 24 h until the reaction was complete as indicated by TLC analysis. The reaction mixture was cooled to room temperature and water (10 mL/mmol of sugar) was added. The contents of the round button flask were stirred for 1 h. Then, diethyl ether (30mL/ mmol of sugar) was added to the re-action. The stirring was continued until white solid precipitated. Suction filtration was then performed to collect the products. Recrystallization by ethanol was then performed to give pure products.

Glycosylation. To an oven-dried round button flask equipped with a stir bar were added sugar octaacetate (1eq) and indium bromide (3 wt%). The round button flask was evacuated and refilled with dry nitrogen. Anhydrous chloroform (5 mL/mmol of sugar octaacetate) and alkyl thiol (2 eq) were then added via syringe. The contents of the round button flask were stirred and refluxed at 60 °C for 24 h until the reaction was complete as indicated by TLC analysis. The reaction mixture was then cooled to room

temperature and extracted with chloroform. The combined organic layers were washed three times with copious amounts of water and then dried over magnesium sulfate. The organic phase was then condensed by evaporation. The resulting residue was purified by column chromatography on silica gel to give the product.

Deacetylation. To an oven-dried round button flask equipped with a stir bar were added alkyl-β-thioperacetyl sugar (1 eq) and Amberlyst A26 hydroxide form (Sigma-Aldrich) resin (35 wt% of alkyl-β-thioperacetyl sugar). Methanol was then added to the mixture. The contents of the round button flask were stirred for 24 h until the reaction was complete as indicated by TLC analysis. Gravity filtration was then performed to separate resin from organic phase. The organic phase was then condensed by evaporation. The chemical structures of final compounds were characterized by ^{1}H NMR, ^{13}C NMR and mass spectrometry.

II. Spectral Data for LacSCx and CelSCx

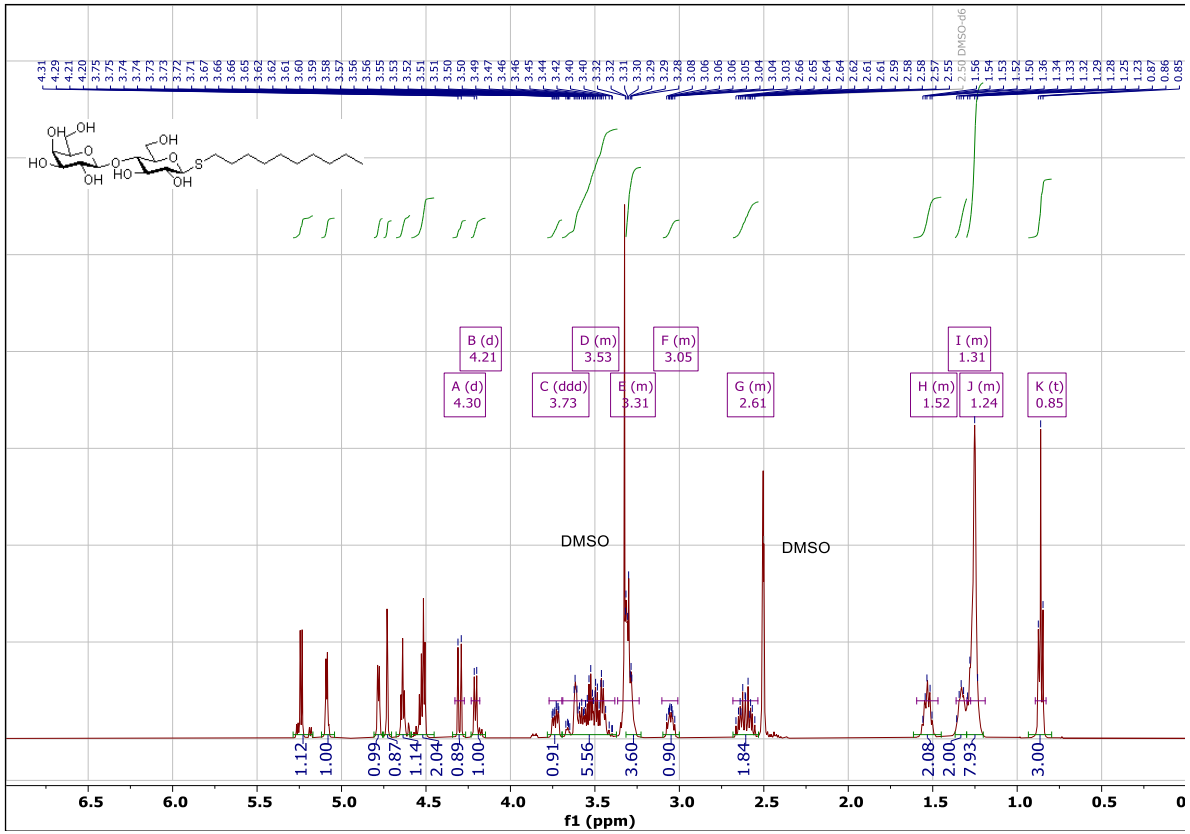


Figure S1. ^{1}H NMR spectrum (500 MHz in $(CD_3)_2SO$) for LacSC8. δ 4.31 (d, J = 9.8 Hz, 1H), 4.21 (d, J = 7.3 Hz, 1H), 3.74 (ddd, J = 11.7, 5.6, 2.1 Hz, 1H), 3.69 – 3.39 (m, 6H), 3.37 – 3.24 (m, 4H), 3.11 – 3.01 (m, 1H), 2.69 – 2.54 (m, 2H), 1.60 – 1.47 (m, 2H), 1.37 – 1.28 (m, 2H), 1.30 – 1.19 (m, 8H), 0.86 (t, J = 6.8 Hz, 3H).

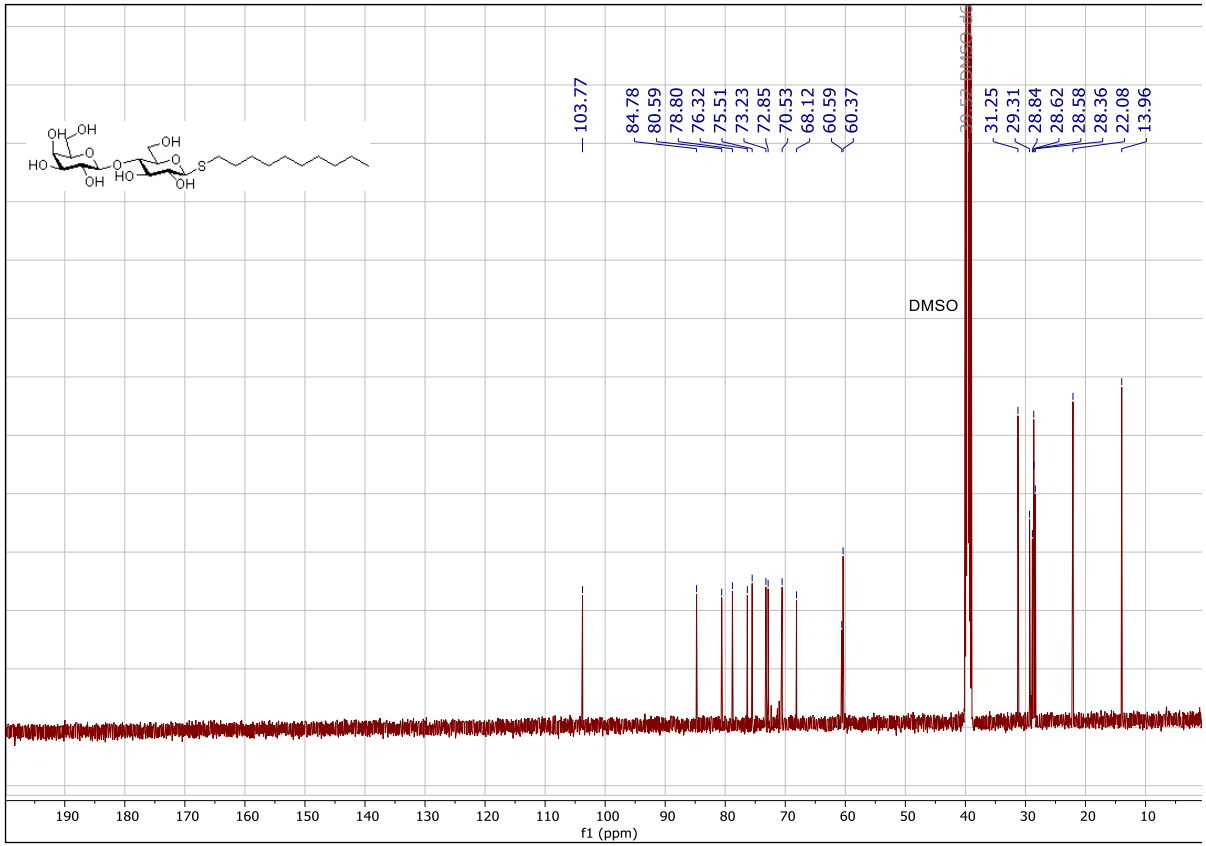


Figure S2. ^{13}C NMR spectrum (126 MHz in $(CD_3)_2SO$) for LacSC8. δ 103.77, 84.78, 80.59, 78.80, 76.32, 75.51, 73.23, 72.85, 70.53, 68.12, 60.59, 60.37, 31.25, 29.31, 28.84, 28.62, 28.58, 28.36, 22.08, 13.96.

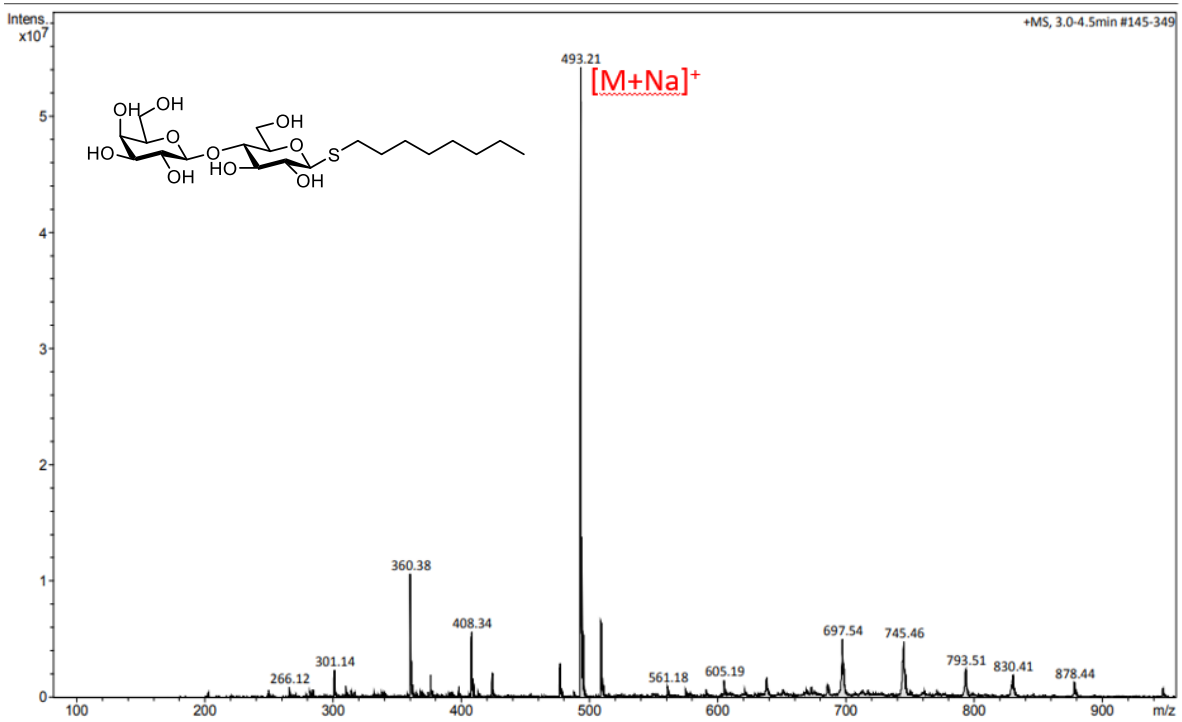


Figure S3. ESI-MS for LacSC8.

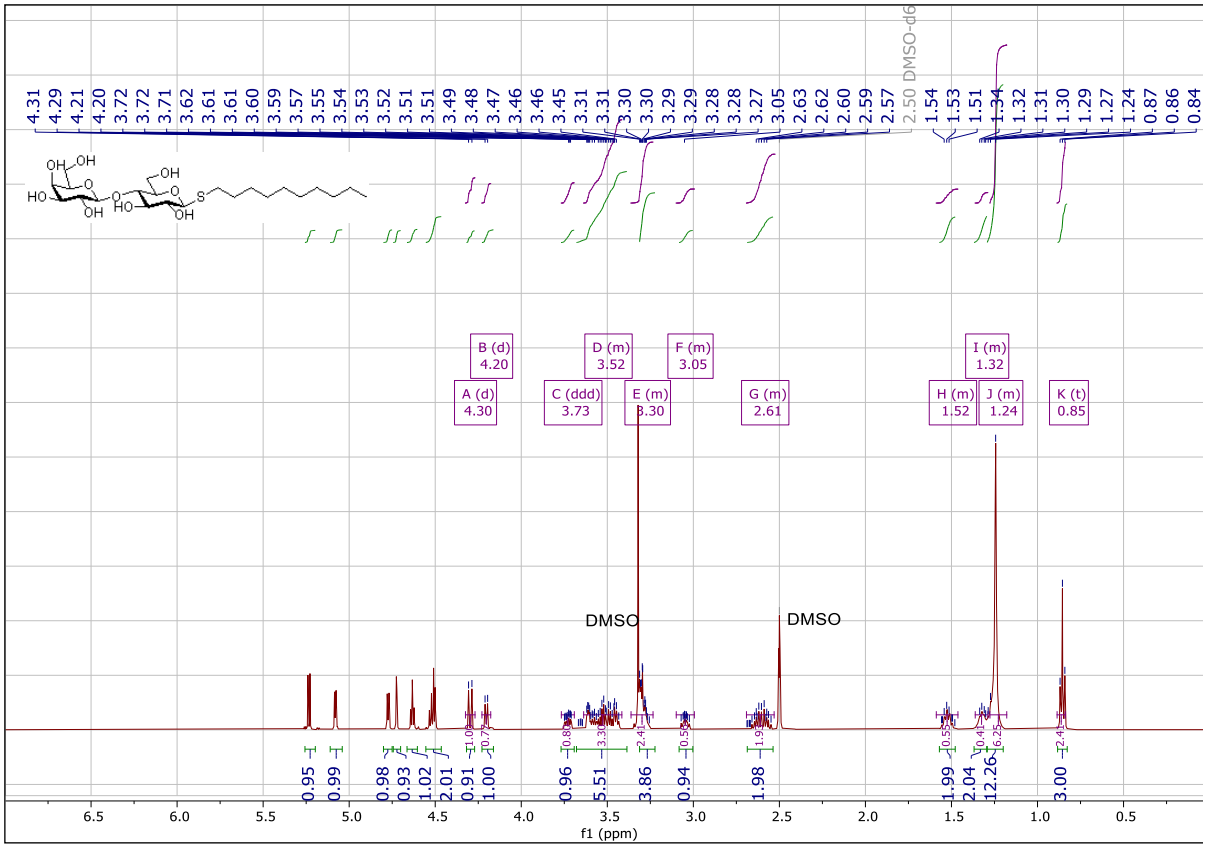


Figure S4. ^{1}H NMR spectrum (500 MHz in $(CD_3)_2SO$) for LacSC10. δ 4.30 (d, J = 9.8 Hz, 1H), 4.20 (d, J = 7.5 Hz, 1H), 3.73 (ddd, J = 11.7, 5.6, 2.1 Hz, 1H), 3.64 – 3.42 (m, 6H), 3.36 – 3.23 (m, 4H), 3.10 – 2.99 (m, 1H), 2.69 – 2.53 (m, 2H), 1.59 – 1.46 (m, 2H), 1.36 – 1.29 (m, 2H), 1.28 – 1.18 (m, 12H), 0.85 (t, J = 6.8 Hz, 3H).

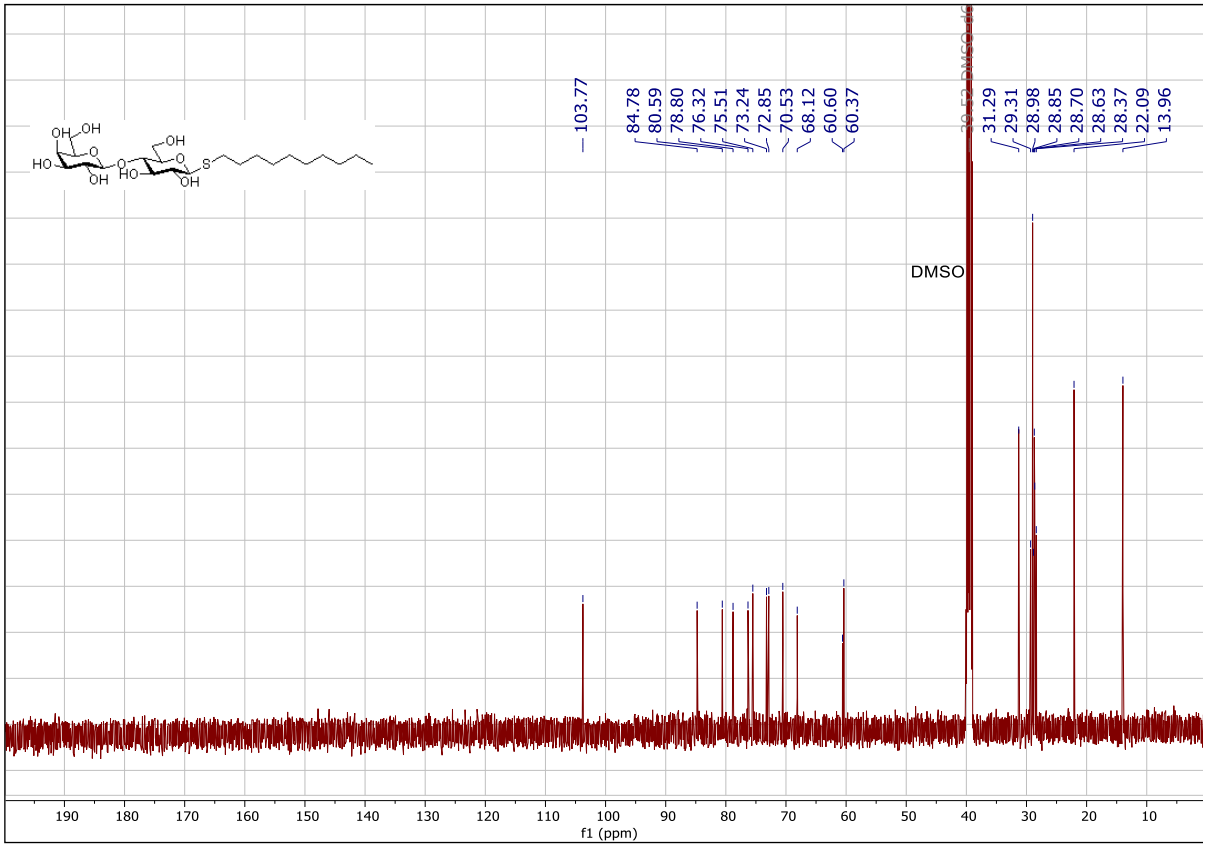


Figure S5. ^{13}C NMR spectrum (126 MHz in $(CD_3)_2SO$) for LacSC10. δ 103.77, 84.78, 80.59, 78.80, 76.32, 75.51, 73.24, 72.85, 70.53, 68.12, 60.60, 60.37, 31.29, 29.31, 28.98, 28.85, 28.70, 28.63, 28.37, 22.09, 13.96.

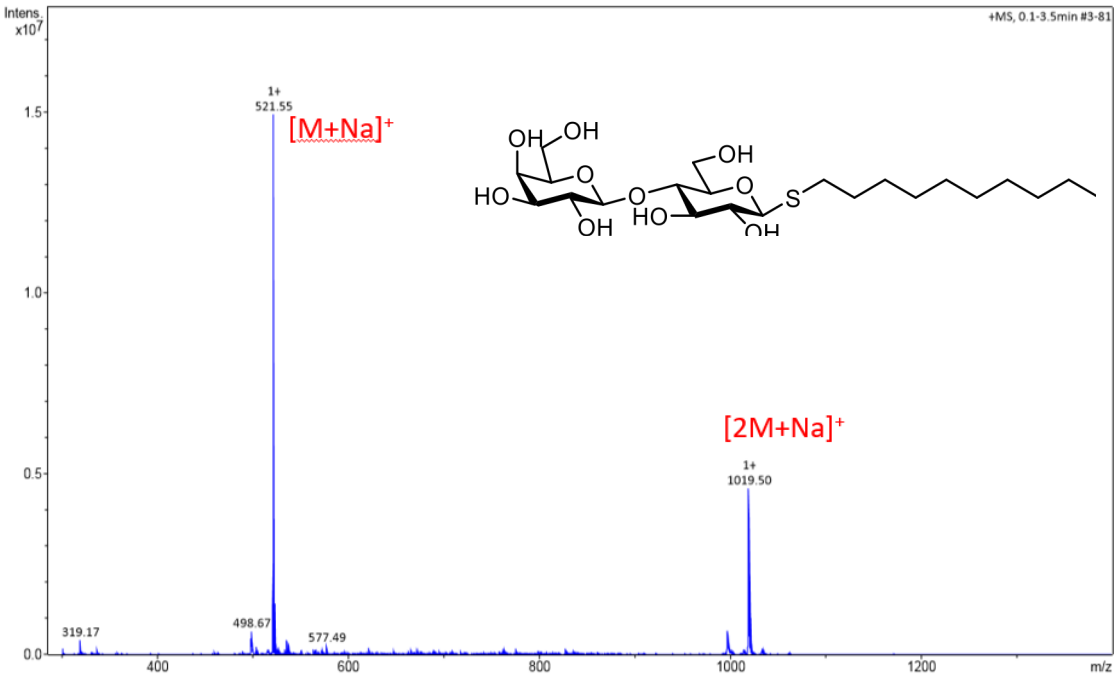


Figure S6. ESI-MS for LacSC10.

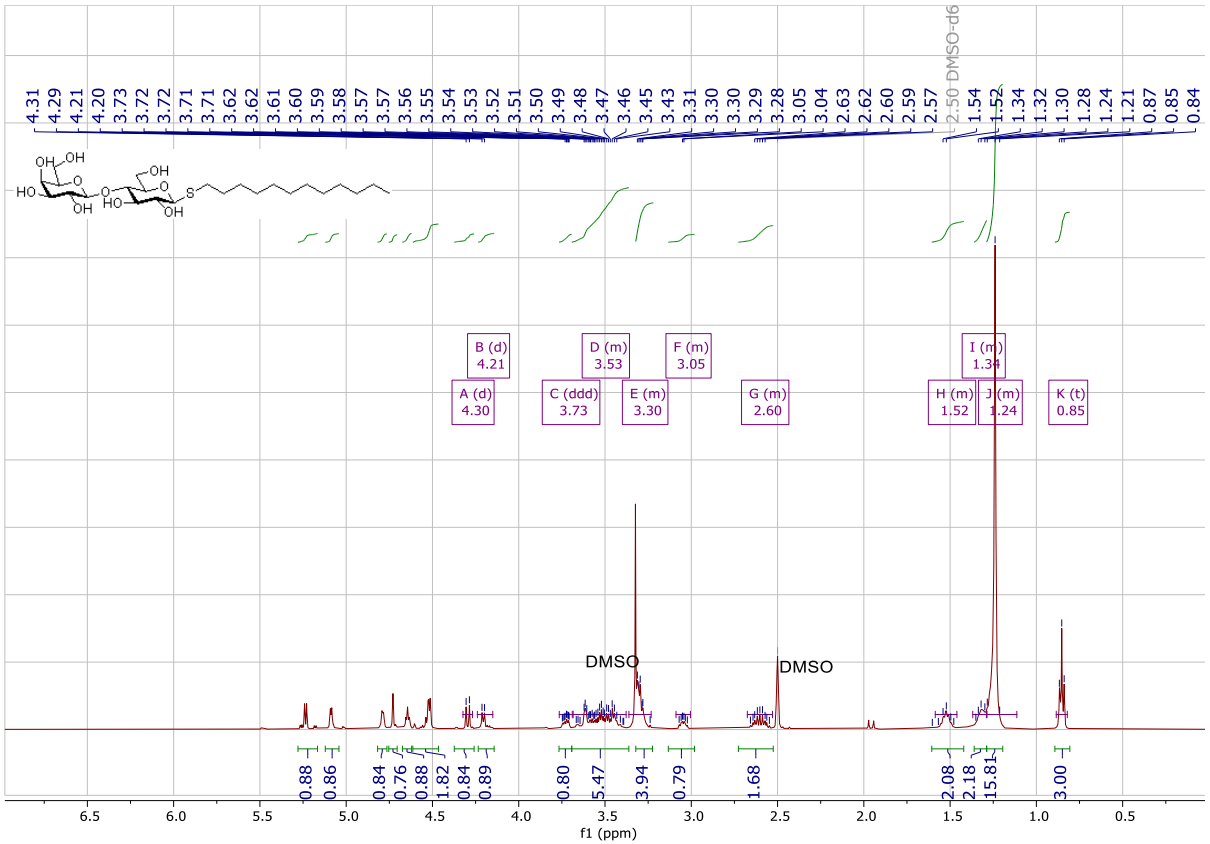


Figure S7. ^{1}H NMR spectrum (500 MHz in $(CD_3)_2SO$) for LacSC12. δ 4.30 (d, J = 9.6 Hz, 1H), 4.21 (d, J = 7.3 Hz, 1H), 3.73 (ddd, J = 11.7, 5.6, 2.1 Hz, 1H), 3.68 – 3.38 (m, 6H), 3.36 – 3.23 (m, 4H), 3.09 – 3.00 (m, 1H), 2.68 – 2.53 (m, 2H), 1.59 – 1.46 (m, 2H), 1.37 – 1.29 (m, 2H), 1.29 – 1.11 (m, 16H), 0.85 (t, J = 6.8 Hz, 3H).

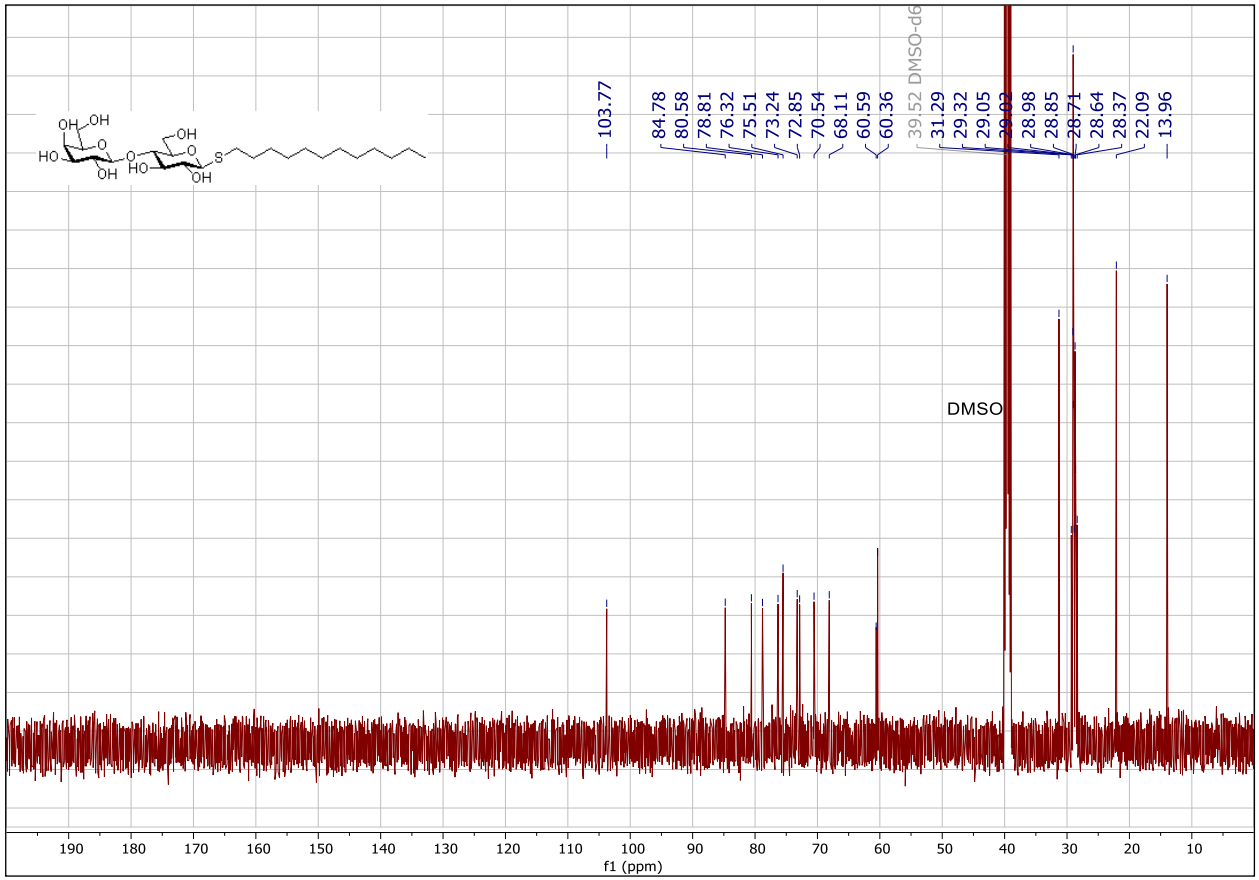


Figure S8. ^{13}C NMR spectrum (126 MHz in $(CD_3)_2SO$) for LacSC12. δ 103.77, 84.78, 80.58, 78.81, 76.32, 75.51, 73.24, 72.85, 70.54, 68.11, 60.59, 60.36, 31.29, 29.32, 29.05, 29.02, 28.98, 28.85, 28.71, 28.64, 28.37, 22.09, 13.96.

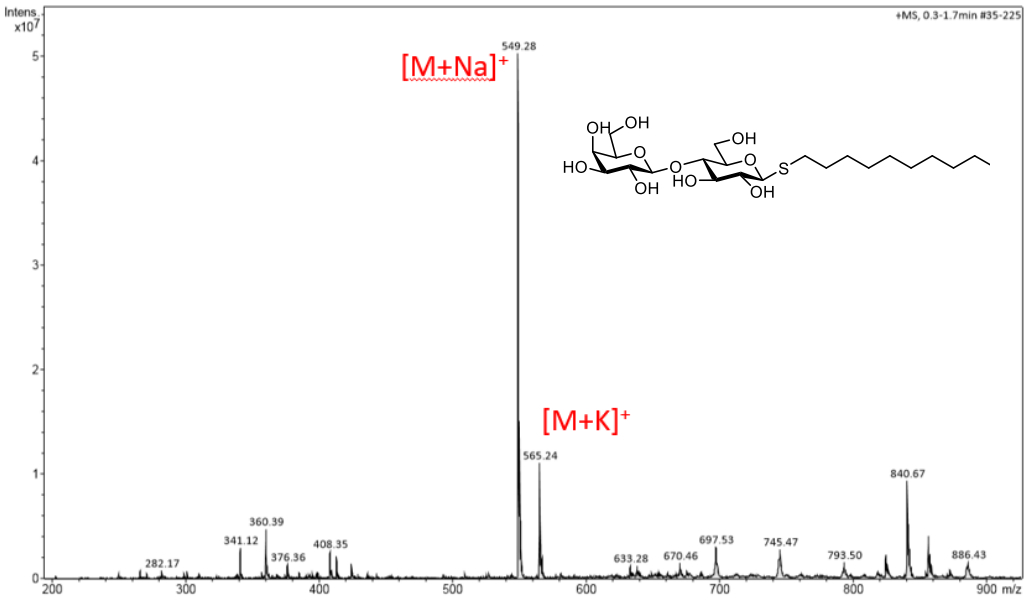


Figure S9. ESI-MS for LacSC12.

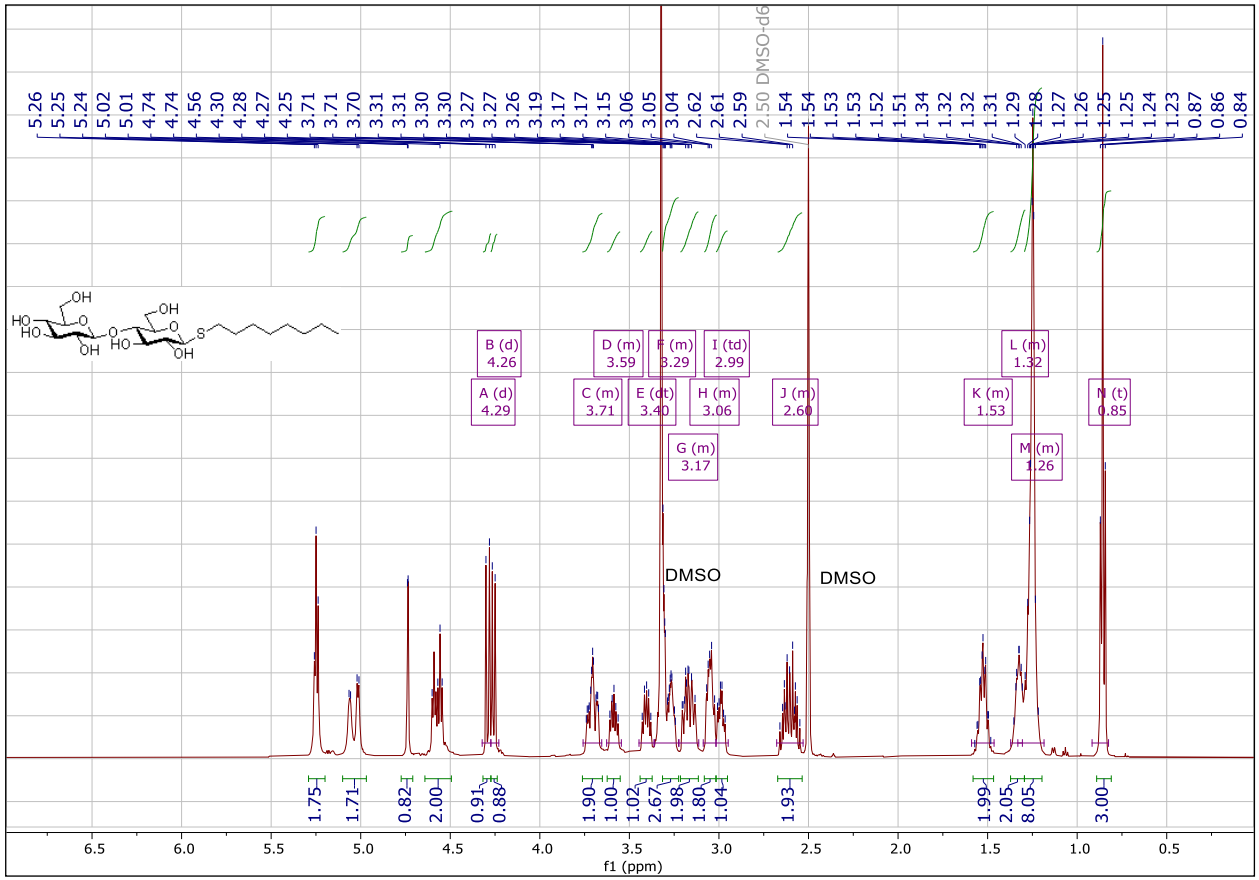


Figure S10. ^{1}H NMR spectrum (500 MHz in $(CD_3)_2SO$) for CelSC8. δ 4.29 (d, J = 9.8 Hz, 1H), 4.26 (d, J = 7.9 Hz, 1H), 3.76 – 3.65 (m, 2H), 3.63 – 3.54 (m, 1H), 3.40 (dt, J = 11.7, 6.1 Hz, 1H), 3.36 – 3.23 (m, 3H), 3.22 – 3.11 (m, 2H), 3.09 – 3.02 (m, 2H), 2.99 (td, J = 8.5, 4.4 Hz, 1H), 2.68 – 2.53 (m, 2H), 1.59 – 1.46 (m, 2H), 1.37 – 1.31 (m, 2H), 1.33 – 1.19 (m, 8H), 0.85 (t, J = 6.8 Hz, 3H).

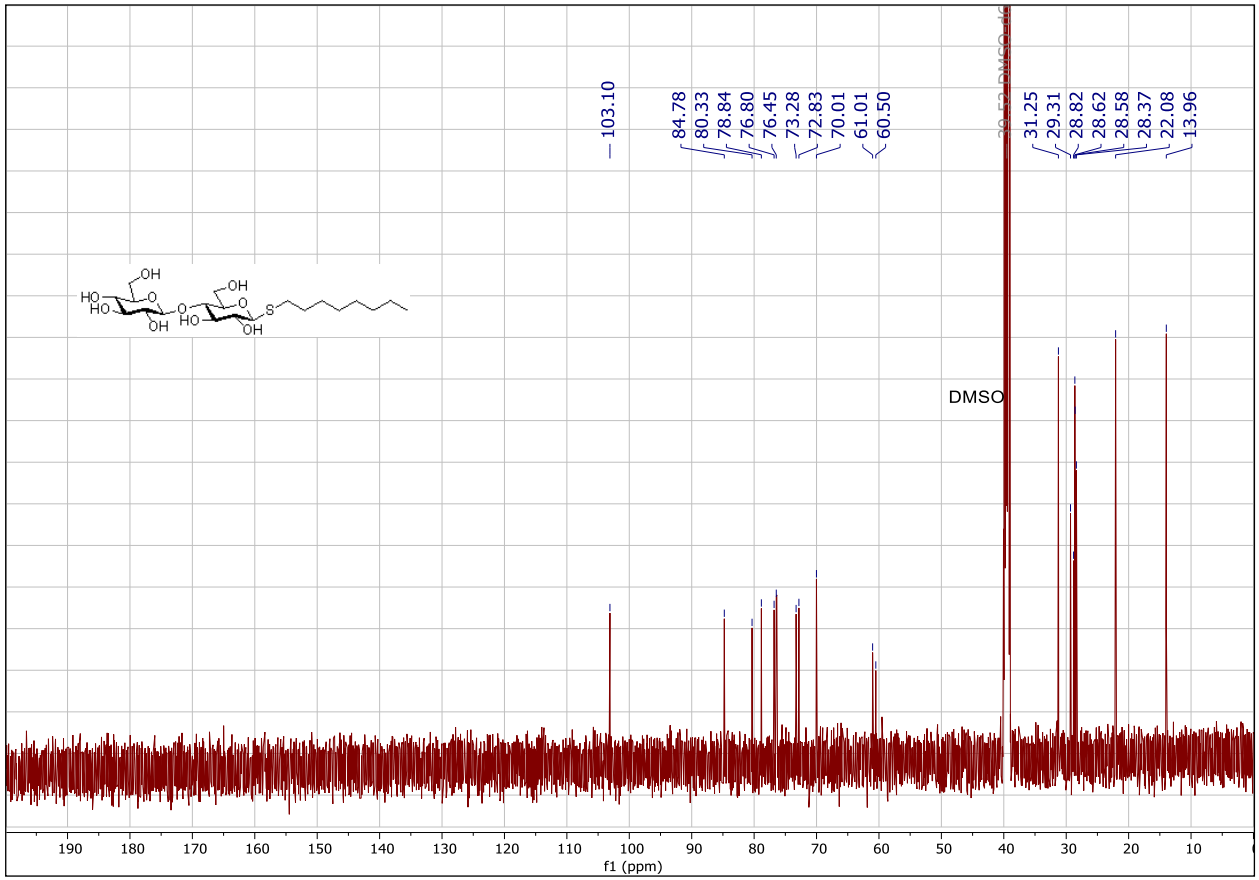


Figure S11. ^{13}C NMR spectrum (126 MHz in $(CD_3)_2SO$) for CelSC8. δ 103.10, 84.78, 80.33, 78.84, 76.80, 76.45, 73.28, 72.83, 70.01, 61.01, 60.50, 31.25, 29.31, 28.82, 28.62, 28.58, 28.37, 22.08, 13.96.

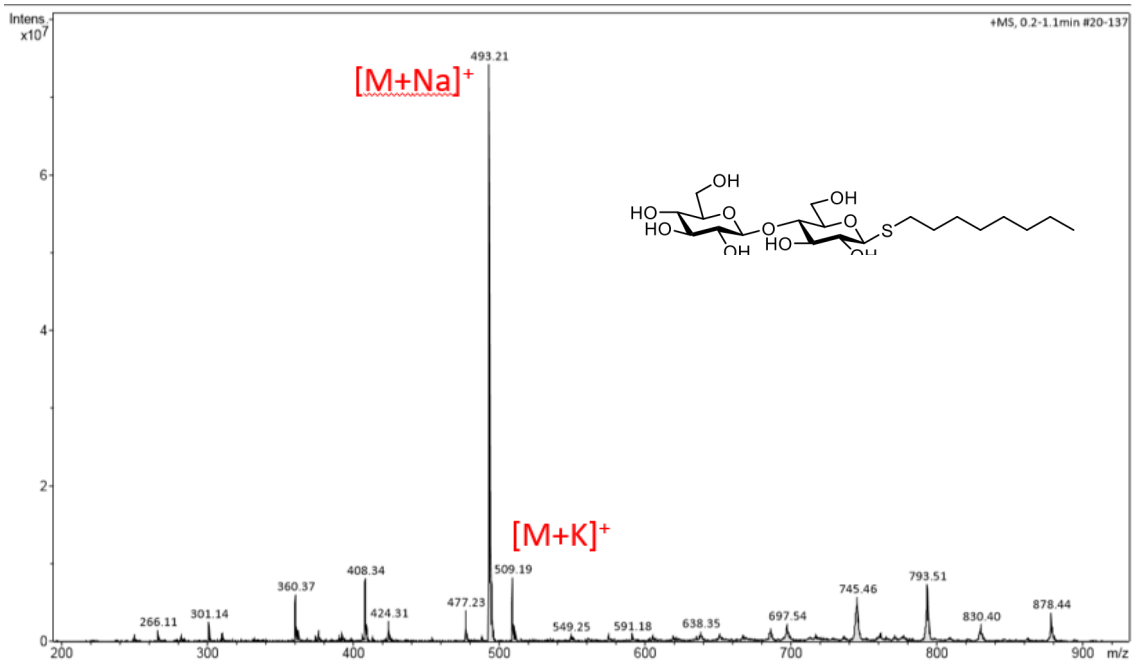


Figure S12. ESI-MS for CelSC8.

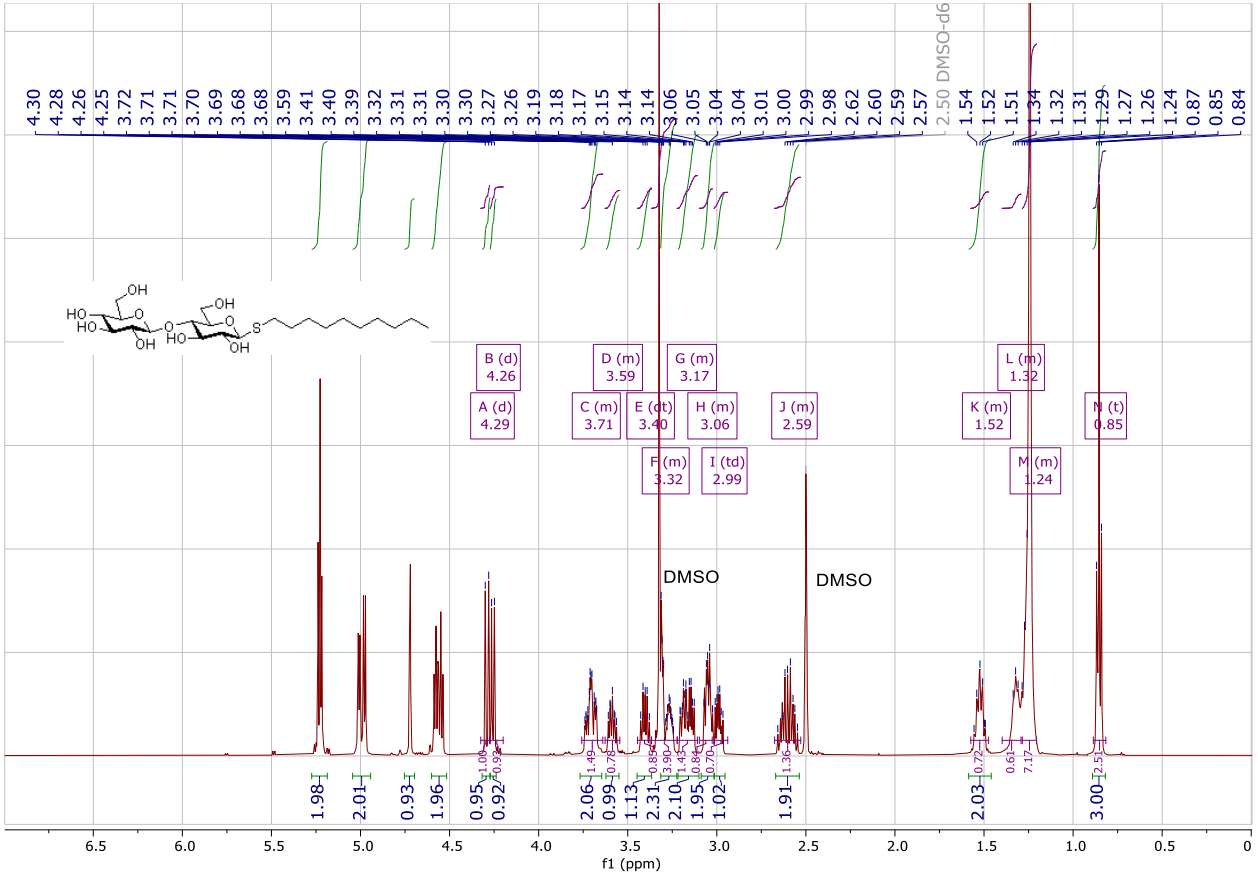


Figure S13. ^{1}H NMR spectrum (500 MHz in $(CD_3)_2SO$) for CelSC10. δ 4.29 (d, J = 9.8 Hz, 1H), 4.26 (d, J = 7.9 Hz, 1H), 3.76 – 3.64 (m, 2H), 3.63 – 3.54 (m, 1H), 3.40 (dt, J = 11.6, 6.3 Hz, 1H), 3.37 – 3.22 (m, 3H), 3.22 – 3.11 (m, 2H), 3.10 – 3.03 (m, 2H), 2.99 (td, J = 9.0, 8.5, 4.9 Hz, 1H), 2.68 – 2.53 (m, 2H), 1.57 – 1.47 (m, 2H), 1.40 – 1.29 (m, 2H), 1.28 – 1.21 (m, 12H), 0.85 (t, J = 6.8 Hz, 3H).

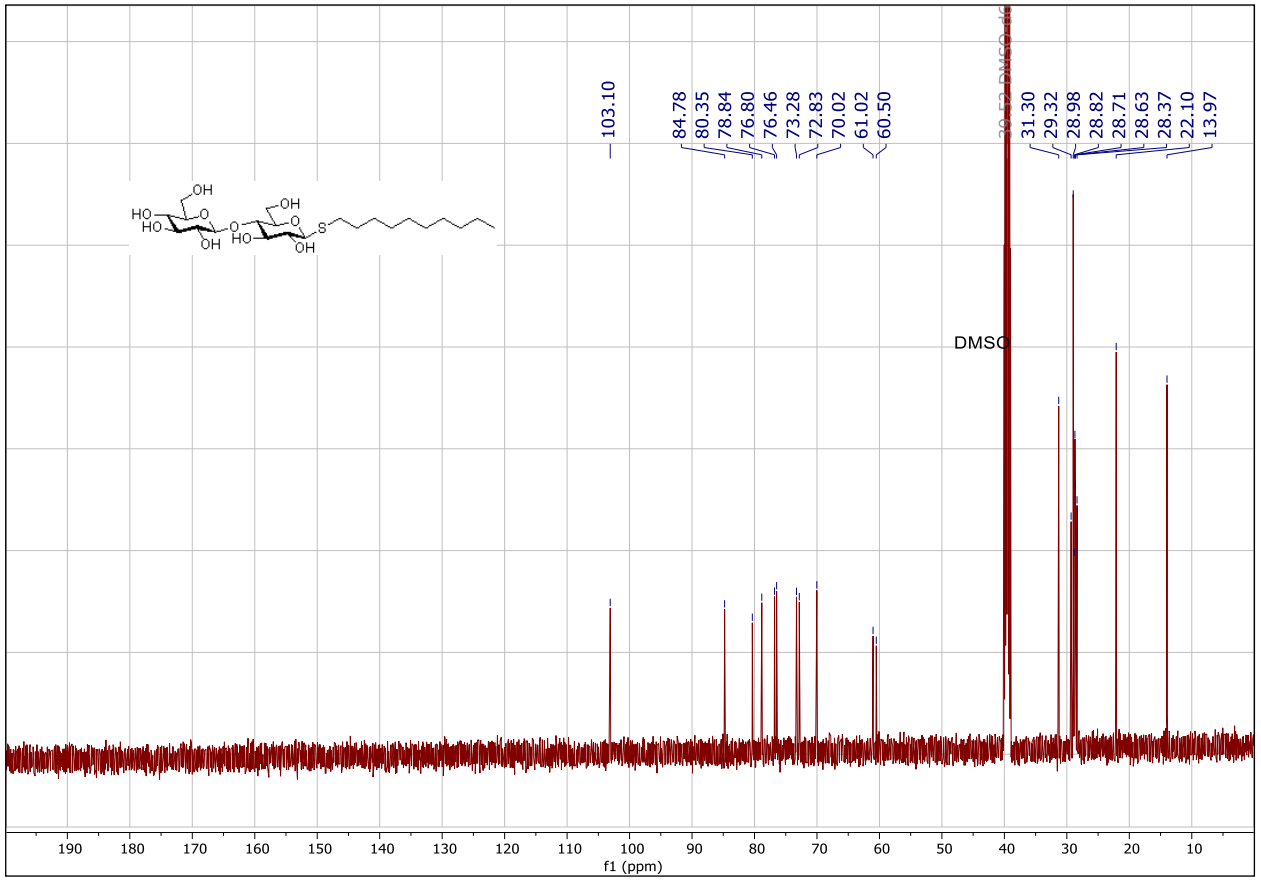


Figure S14. ^{13}C NMR spectrum (126 MHz in $(CD_3)_2SO$) for CelSC10. δ 103.10, 84.78, 80.35, 78.84, 76.80, 76.46, 73.28, 72.83, 70.02, 61.02, 60.50, 31.30, 29.32, 28.98, 28.82, 28.71, 28.63, 28.37, 22.10, 13.97.

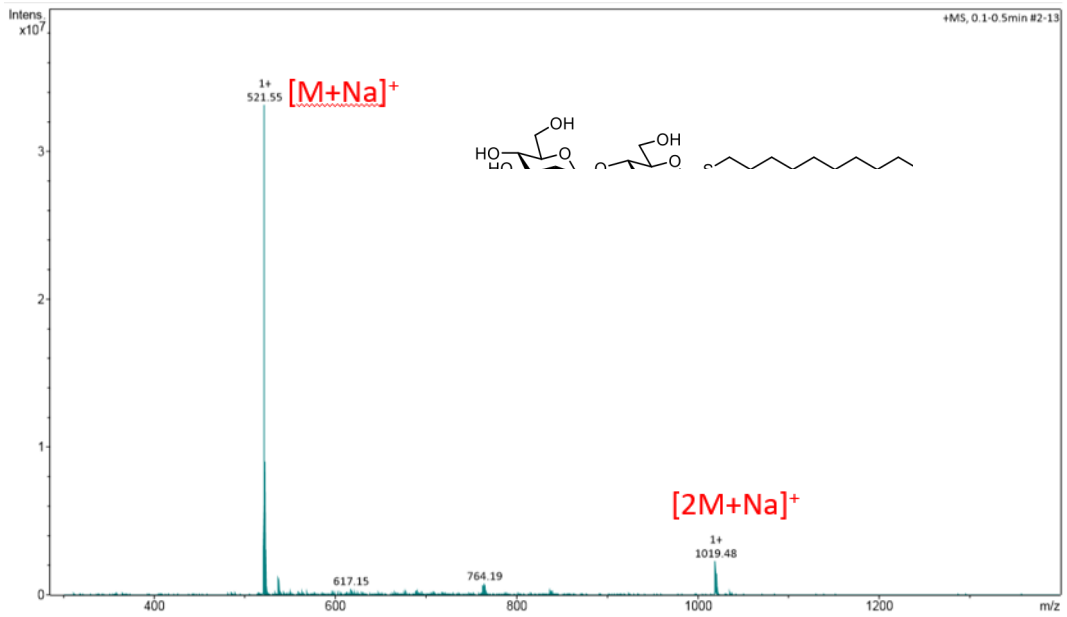


Figure S15. ESI-MS for CelSC10.

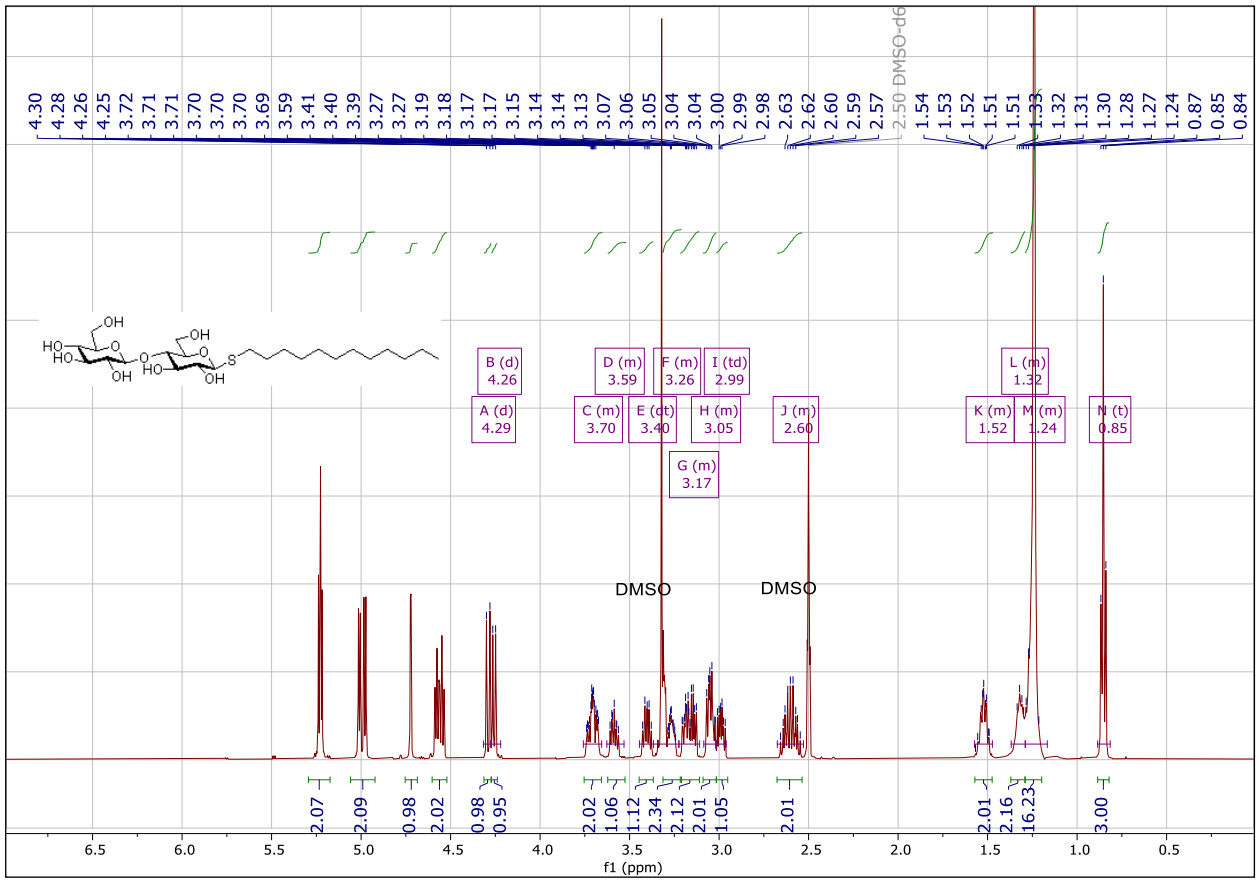


Figure S16. ^{1}H NMR spectrum (500 MHz in $(CD_3)_2SO$) for CelSC12. δ 4.29 (d, J = 9.8 Hz, 1H), 4.26 (d, J = 7.9 Hz, 1H), 3.76 – 3.66 (m, 2H), 3.63 – 3.53 (m, 1H), 3.40 (dt, J = 11.4, 6.2 Hz, 1H), 3.37 – 3.22 (m, 3H), 3.22 – 3.11 (m, 2H), 3.09 – 3.02 (m, 2H), 2.99 (td, J = 9.1, 4.8 Hz, 1H), 2.67 – 2.53 (m, 2H), 1.57 – 1.47 (m, 2H), 1.37 – 1.29 (m, 2H), 1.29 – 1.17 (m, 16H), 0.85 (t, J = 6.8 Hz, 3H).

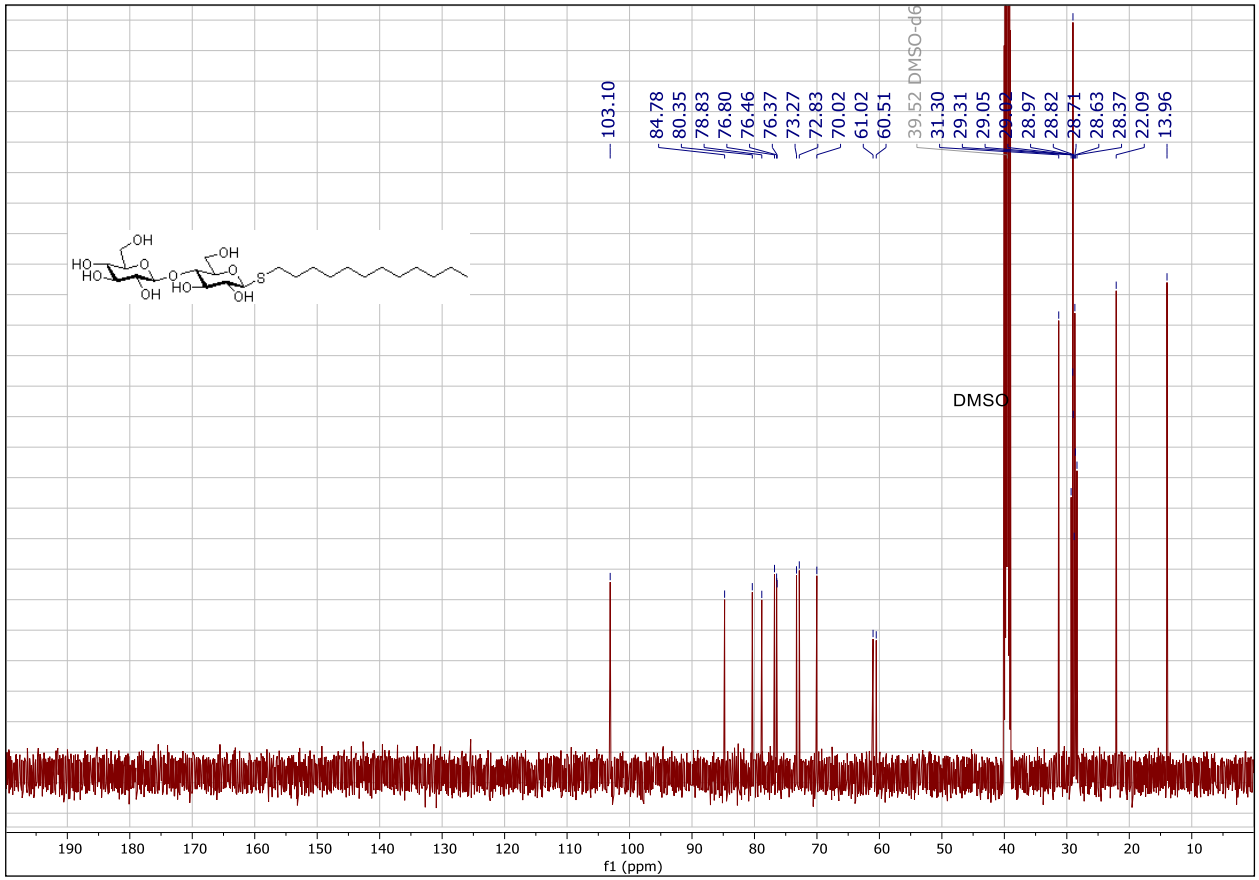


Figure S17. ^{13}C NMR spectrum (126 MHz in $(CD_3)_2SO$) for CelSC12. δ 103.10, 84.78, 80.35, 78.83, 76.80, 76.46, 76.37, 73.27, 72.83, 70.02, 61.02, 60.51, 31.30, 29.31, 29.05, 29.02, 28.97, 28.82, 28.71, 28.63, 28.37, 22.09, 13.96.

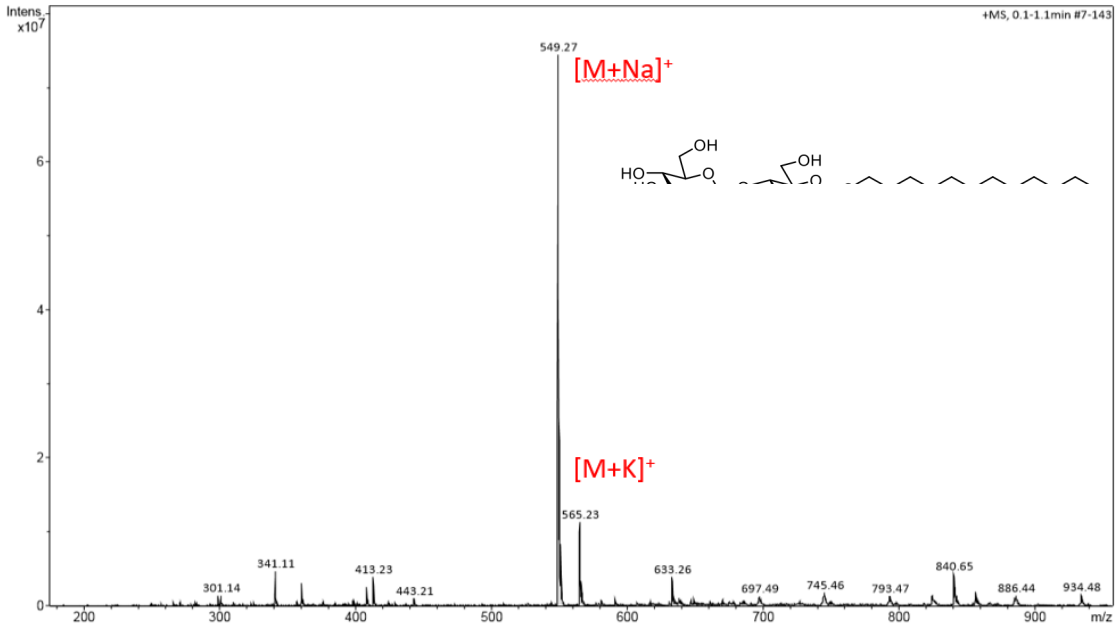


Figure S18. ESI-MS for CelSC10.

III. Hydrogel Preparation

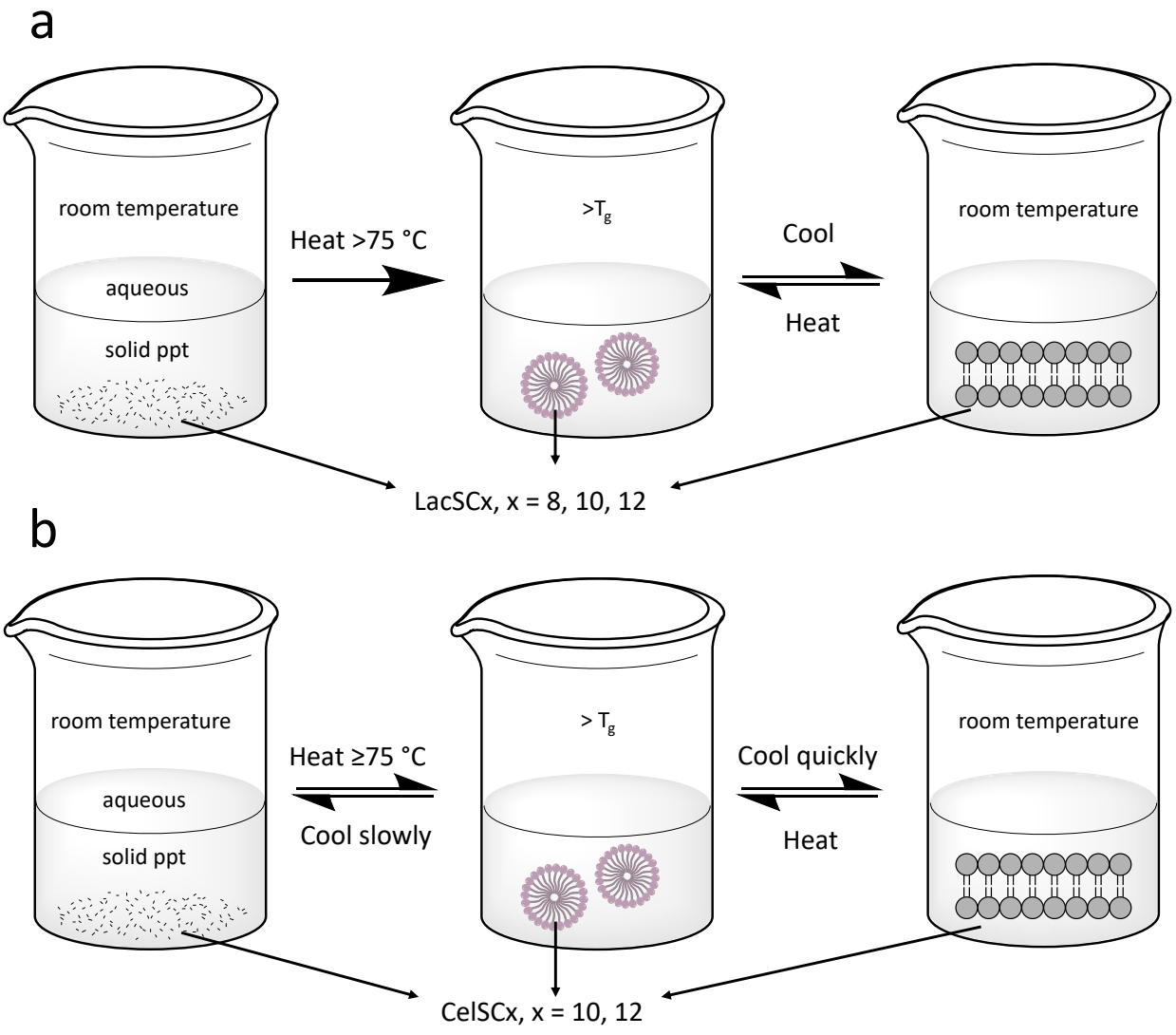


Figure S19. a) Schematic for preparation of LacSCx hydrogels. b) Schematic for preparation of CelSCx hydrogels.

IV. Rheology for LacSCx Systems

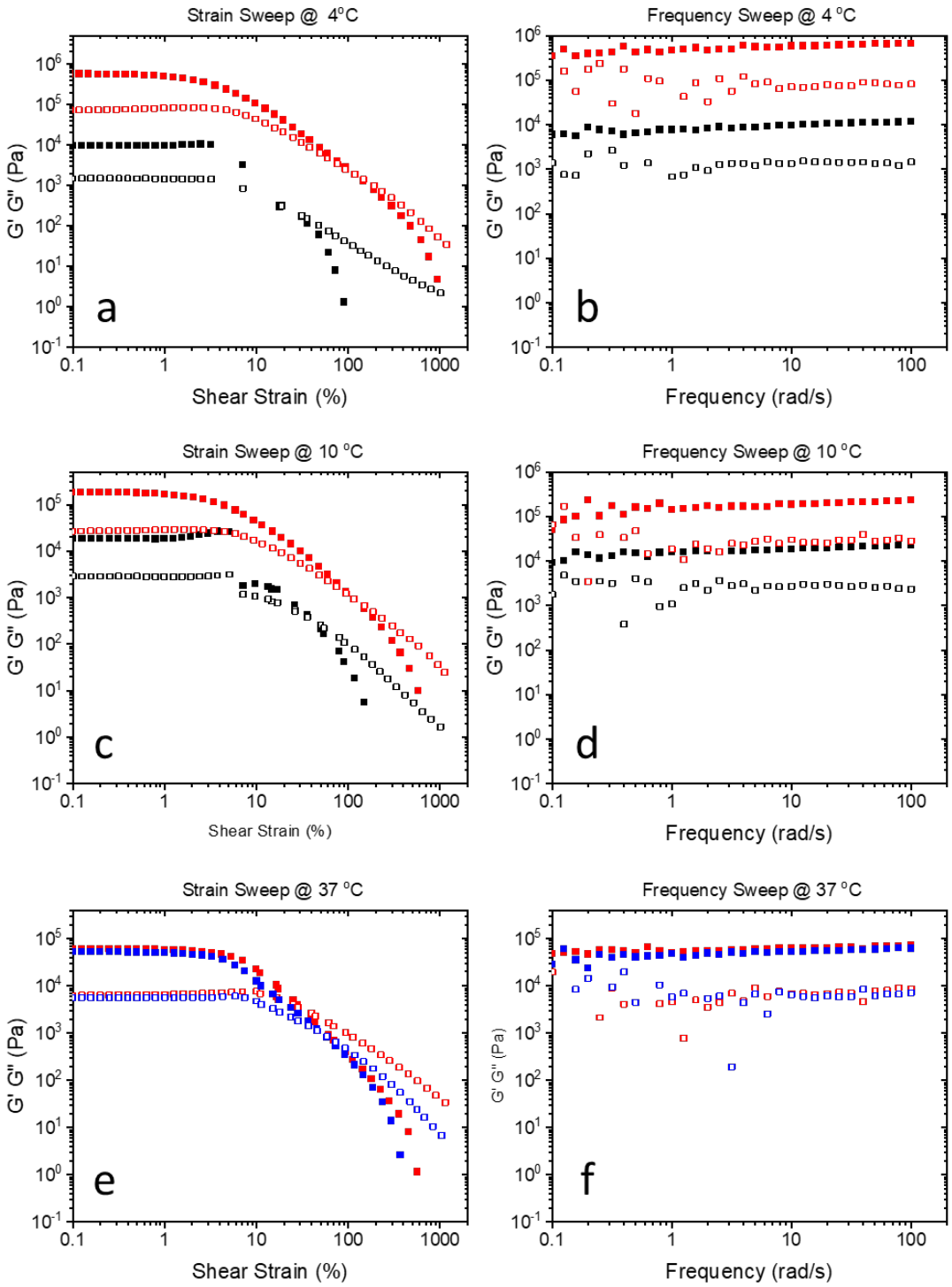


Figure S20. a), c) Strain sweep and b), d) frequency sweep results for 1 wt% LacSC8 and 2 wt% LacSC10 at a), b) 4 °C and c), d) 10 °C. Strain sweep (e) and frequency sweep (f) results for 2 wt% LacSC10 and 1 wt% LacSC12 at 37 °C. Solid symbols indicate G′ values, open symbols represent G″ values for LacSC8 (■, □), LacSC10 (■, □), and LacSC12 (■, □).

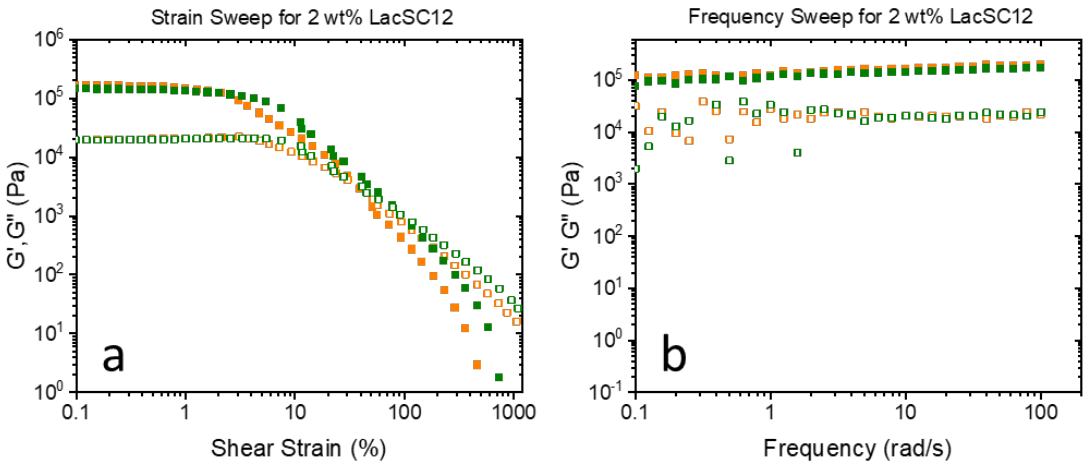


Figure S21. a) Strain sweep and b) frequency sweep results for 2 wt% LacSC12 at 25 °C (■, □) and 37 °C (■, □). Solid symbols indicate G′ values, open symbols represent G″ values.

Table S1. Storage modulus (G′), loss modulus (G″) from rheology measurements on LacSCx and CelSCx hydrogels.

LacSCx (x = 8, 10, 12)								
T (°C)	**1 wt% LacSC8**		**2 wt% LacSC10**		**1 wt% LacSC12**		**2 wt% LacSC12**	
	G′ (Pa)	**G″ (Pa)**	**G′ (Pa)**	**G″ (Pa)**	**G′ (Pa)**	**G″ (Pa)**	**G′ (Pa)**	**G″ (Pa)**
4	1.0×10^4	1.0×10^3	5.7×10^5	9.0×10^4	-	-	-	-
10	1.0×10^4	2.0×10^3	1.8×10^5	2.1×10^4	-	-	-	-
25	1.0×10^4	5.0×10^2	1.3×10^5	2.0×10^4	6.8×10^4	7.0×10^4	1.6×10^5	1.1×10^4
37	-	-	6.0×10^4	7.0×10^3	5.1×10^4	3.9×10^4	1.4×10^5	1.1×10^4

CelSCx (x = 10, 12)								
T (°C)	**1 wt% CelSC10**		**1 wt% CelSC12**					
4	**G′ (Pa)**	**G″ (Pa)**	**G′ (Pa)**	**G″ (Pa)**				
10	6.8×10^4	7.0×10^3	4.0×10^3	2.0×10^2				
25	-	-	-	-				
37	-	-	-	-				
4	-	-	-	-				

V. Fluorescence Spectroscopy for LacSCx Systems

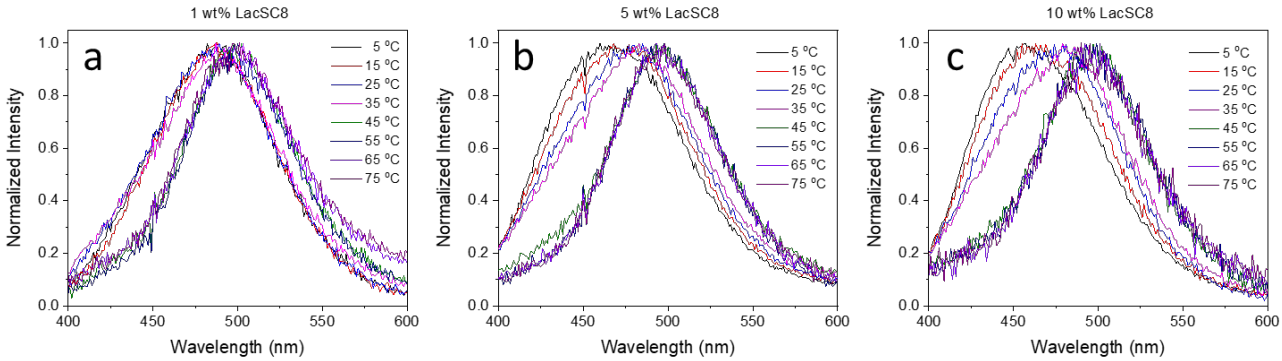


Figure S22. Prodan fluorescence spectra for hydrogels of a) 1 wt%, b) 5 wt%, and c) 10 wt% LacSC8 as a function of temperature.

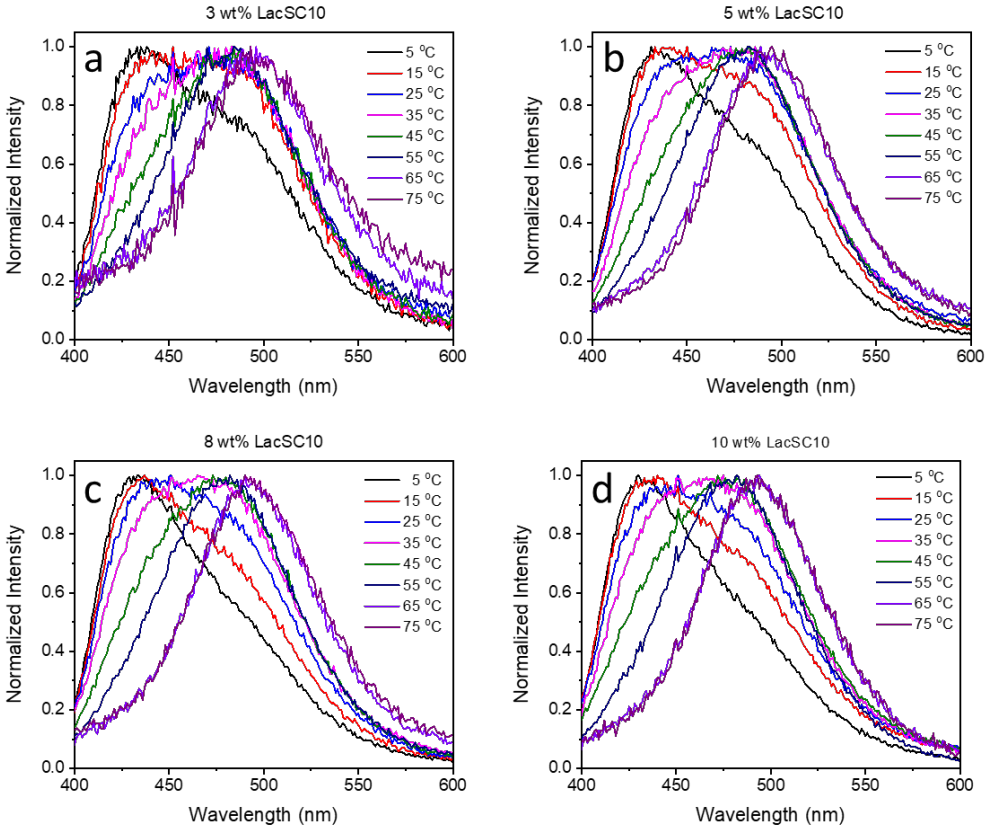


Figure S23. Prodan fluorescence spectra for hydrogels of a) 3 wt%, b) 5 wt%, c) 8 wt%, and d) 10 wt% LacSC10 as a function of temperature.

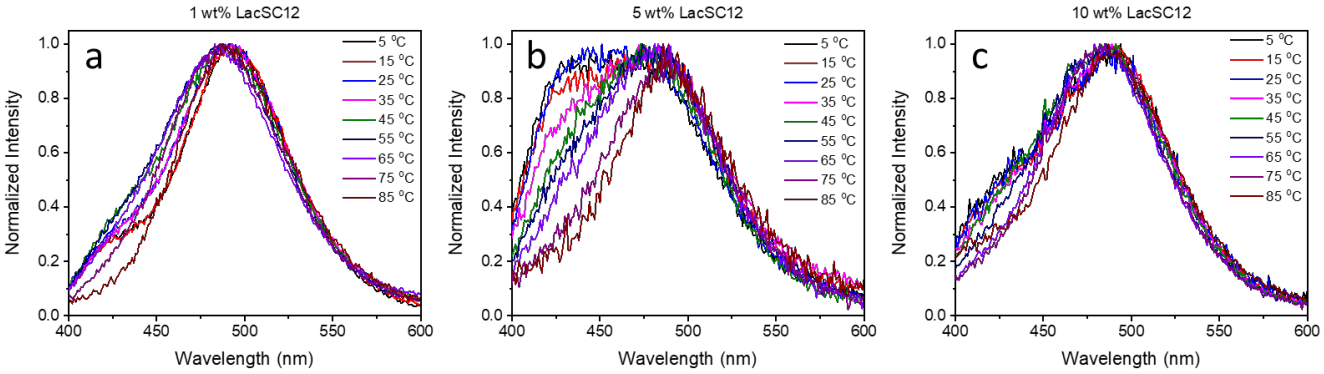


Figure S24. Prodan fluorescence spectra for hydrogels of a) 1 wt%, b) 5 wt%, and c) 10 wt% LacSC12 as a function of temperature.

Table S2. Spectral fitting of prodan fluorescence spectra for LacSCx hydrogels at the lowest concentrations that fully gel.

Thioglycolipid	Gel State (25 °C)		Sol State (75 °C)	
	λ_1 (FWHM) in nm	λ_2 (FWHM) in nm	λ_1 (FWHM) in nm	λ_2 (FWHM) in nm
LacSC8	-	484 (91)	500 (80)	-
LacSC10	428 (38)	478 (90)	493 (78)	-
LacSC12	421 (31)	491(80)	487 (56)*	520 (73)*

*Experiment conducted at 85°C.

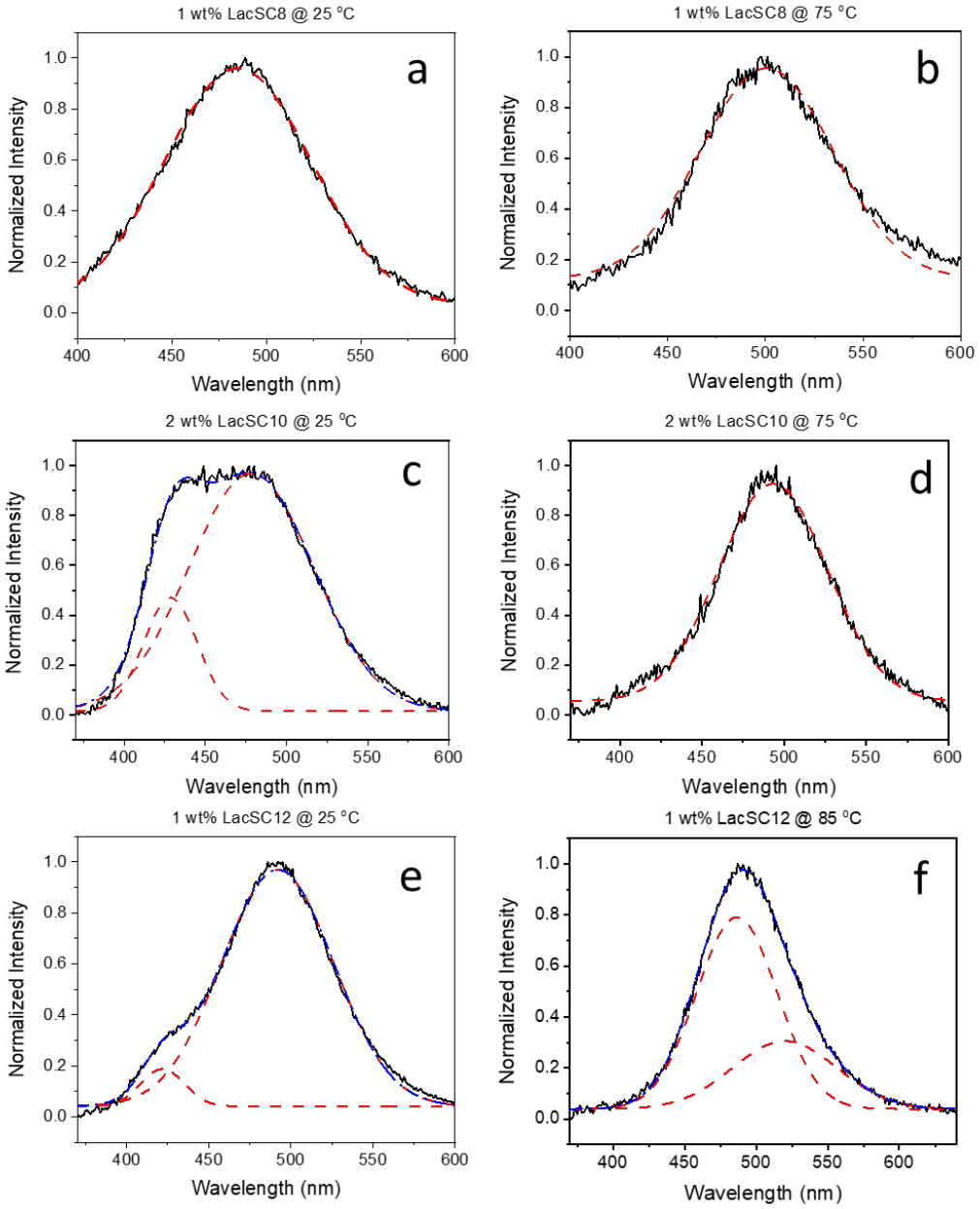


Figure S25. Prodan fluorescence specta and fits to pure Gaussian lineshapes for hydrogels from a) 1 wt% LacSC8 at a) 25°C and b) 75 °C, 2 wt% LacSC10 at c) 25°C and d) 75 °C, and 1 wt% LacSC12 at e) 25°C and f) 85 °C. Raw spectral data are black lines, spectral fits are red dashed lines and blue dot dash lines for systems with multiple bands.

VI. Fluorescence Spectroscopy for CelSCx Systems

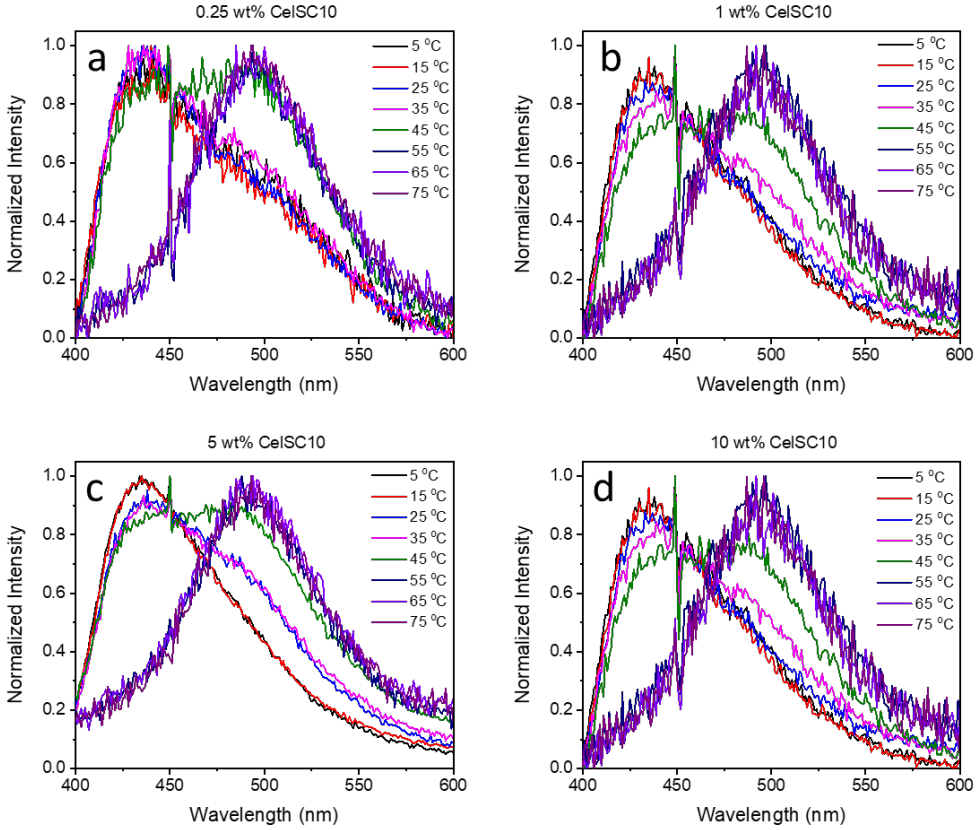


Figure S26. Prodan fluorescence spectra for aqueous solutions of CelSC10 at concentrations of a) 0.25 wt%, b) 1 wt%, c) 5 wt%, and d)10 wt% as a function of temperature.

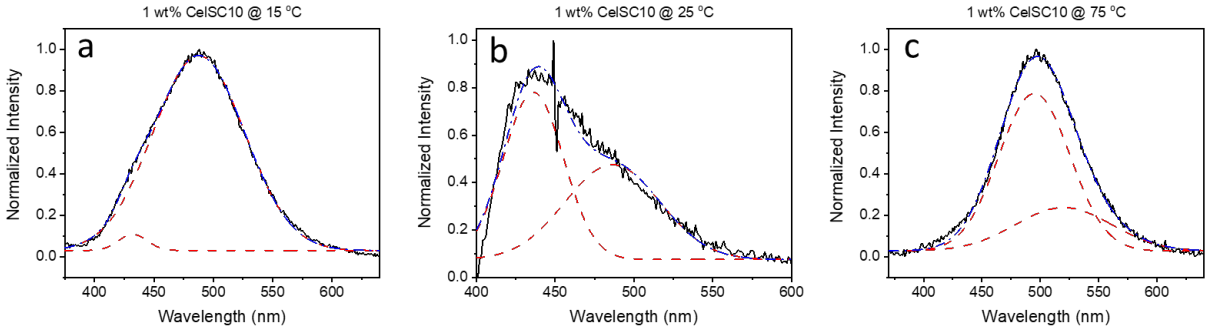


Figure S27. Prodan fluorescence specta and fits to pure Gaussian lineshapes for 1 wt% CelSC10 in a) metastable hydrogel state at 15 °C, b) fibrous state at 25 °C, and c) solution state at 75 °C. Raw spectral data are black lines, spectral fits are red dashed lines and blue dot dash lines for systems with multiple bands.

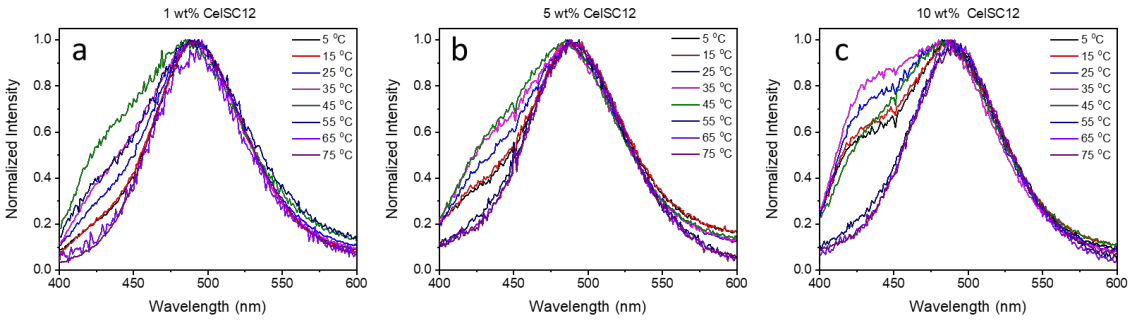


Figure S28. Prodan fluorescence spectra for aqueous solutions of CelSC12 at concentrations of a) 1 wt%, b) 5 wt%, and c) 10 wt% as a function of temperature.

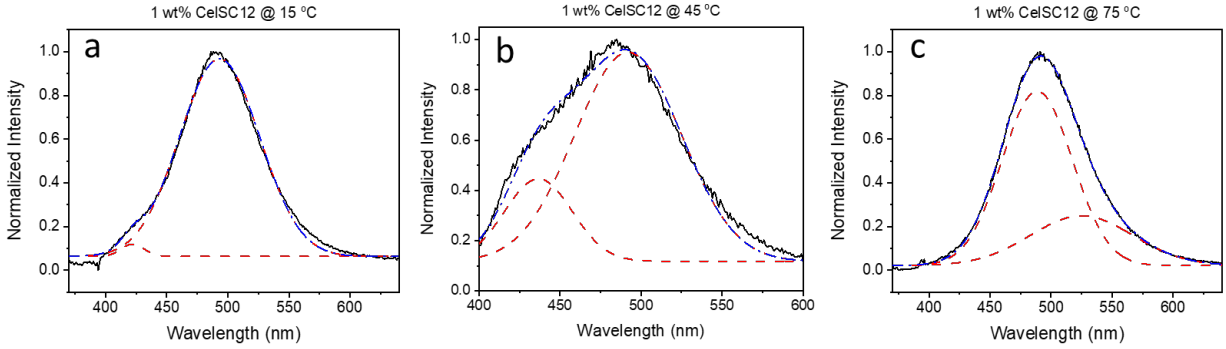


Figure S29. Prodan fluorescence specta and fits to pure Gaussian lineshapes for 1 wt% CelSC12 in a) gel state at 15 °C, b) fibrous state at 45 °C, and c) solution state at 75 °C. Raw spectral data are black lines, spectral fits are red dashed lines and blue dot dash lines for systems with multiple bands.

Table S3. Spectral fitting of prodan fluorescence spectra for 1 wt% CelSCx samples.

Thioglycolipid	Hydrogel State (15 °C)	Fibrous State (25 °C for CelSC10/ 45 °C for CelSC12)		Sol State (75 °C)	
	λ (FWHM) in nm	λ_1 (FWHM) in nm	λ_2 (FWHM) in nm	λ_1 (FWHM) in nm	λ_2 (FWHM) in nm
CelSC10	486 (95)	436 (45)	486 (71)	495 (61)	520 (83)
CelSC12	492 (78)	436 (49)	492 (77)	489 (58)	525 (84)

VII. X-Ray Crystallography for CelSCx Systems

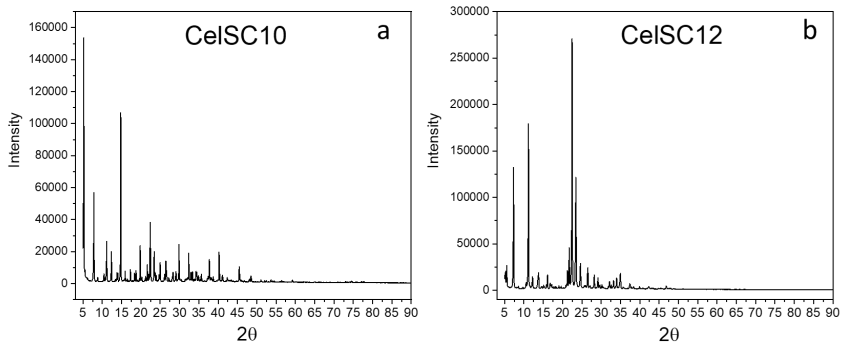


Figure S30. Powder x-ray diffraction from fibers of a) CelSC10 and b) CelSC12.

Single Crystal X-Ray Diffraction

Crystals of CelSC10 and CelSC12 were grown from aqueous solutions. CelSC10 and CelSC12 were first dissolved in water at a concentration of 2 wt% in a hot bath at 75 °C. Once fully dissolved, these CelSCx solutions were place in a 45 °C sand bath for at least 7 days to allow crystal growth.

Crystal Data for CelSC10 ($C_{22}H_{46}O_{12}S$) (M = 534.65 g/mol): monoclinic, space group $P2_1$ (no. 4), a = 16.7335(18) Å, b = 4.8136(5) Å, c = 33.559(4) Å, β = 96.922(4)°, V = 2683.5(5) Å^3, Z = 4, T = 150 K, μ = 0.190 mm^{-1}, D_{calc} = 1.323 g/cm^3, 61061 reflections measured (2.942° ≤ 2Θ ≤ 55.032°), 11006 unique (R_{int} = 0.1814, R_{sigma} = 0.1587) which were used in all calculations. The final R_1 was 0.0856 (I > 2σ(I)) and wR_2 was 0.2350 (all data). (See Figure S30.)

Crystal Data for CelSC12 ($C_{24}H_{50}O_{12}S$) (M = 562.70 g/mol): monoclinic, space group $P2_1$ (no. 4), a = 16.720(2) Å, b = 4.8154(6) Å, c = 35.655(5) Å, β = 94.369(5)°, V = 2862.3(6) Å3, Z = 4, T = 150 K, μ = 0.182 mm-1, D_{calc} = 1.306 g/cm3, 63675 reflections measured (2.846° ≤ 2Θ ≤ 54.918°), 11712 unique (R_{int} = 0.2872, R_{sigma} = 0.2351) which were used in all calculations. The final R_1 was 0.0872 (I > 2σ(I)) and wR_2 was 0.2382 (all data). (See Figure S31.)

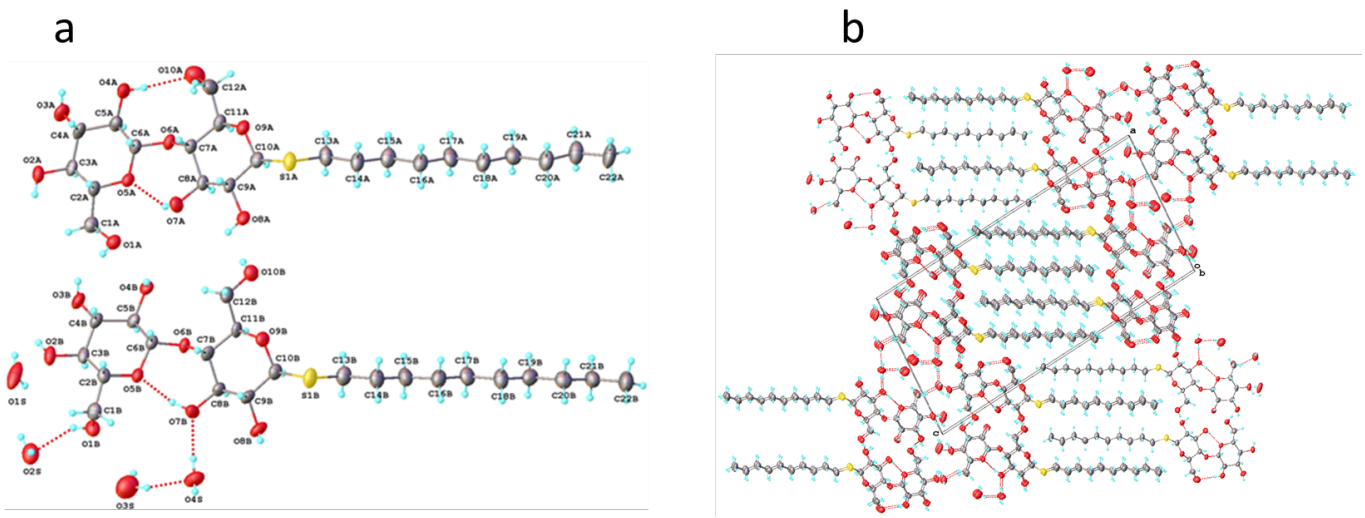


Figure S31. a) Displacement ellipsoid representation of CelSC10, and b) crystal packing in CelSC10.

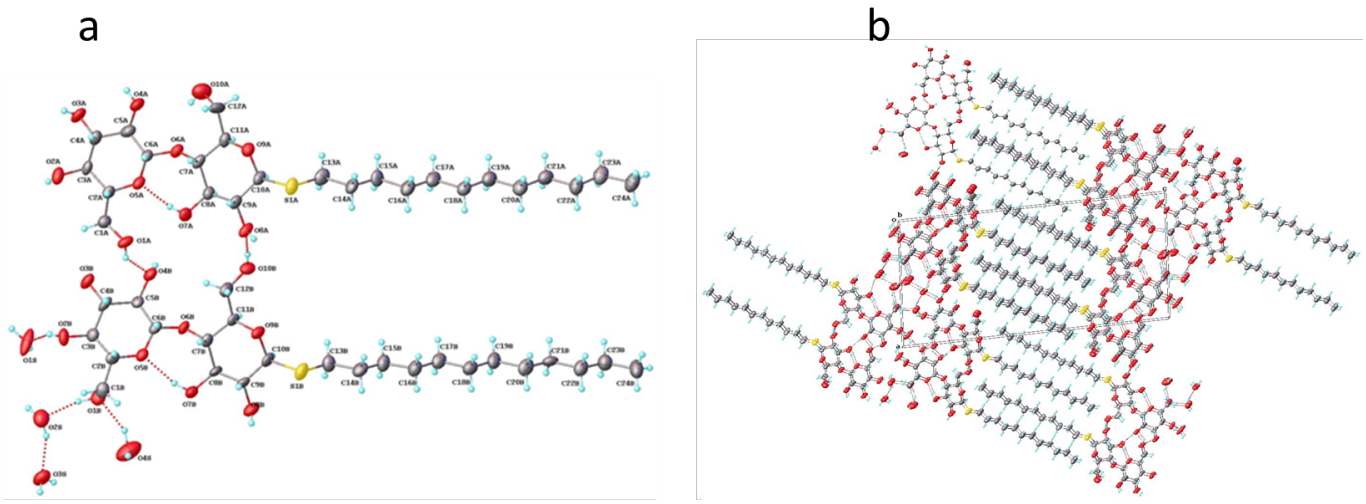


Figure S32. a) Displacement ellipsoid representation of CelSC12, and b) crystal packing in CelSC12.

VIII. References

1. Szabó, L. Z.; Hanrahan, D. J.; Jones, E. M.; Martin, E.; Pemberton, J. E.; Polt, R. Preparation of S-glycoside surfactants and cysteine thioglycosides using minimally competent Lewis acid catalysis. *Carbohydr. Res.* **2016**, *422*, 1-4. DOI: 10.1016/j.carres.2015.12.008