Supporting information

Synthesis of Various Glycopolymers bearing Sialyllactose and the Effect of their Molecular Mobility on Interaction with the Influenza Virus

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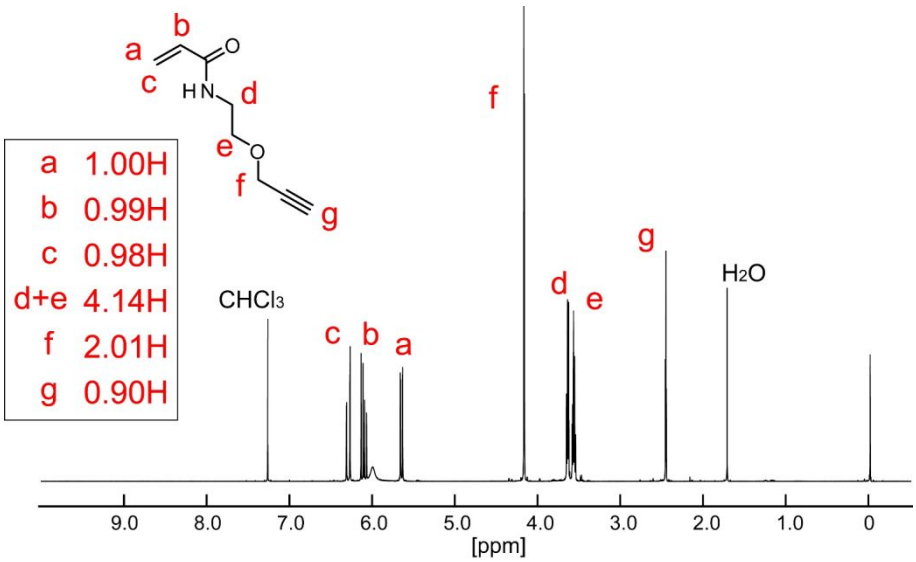

Figure S1-1. ^{1}H NMR spectrum of 2-(2-propynyloxy)ethyl acrylamide (400 MHz, $CDCl_3$).

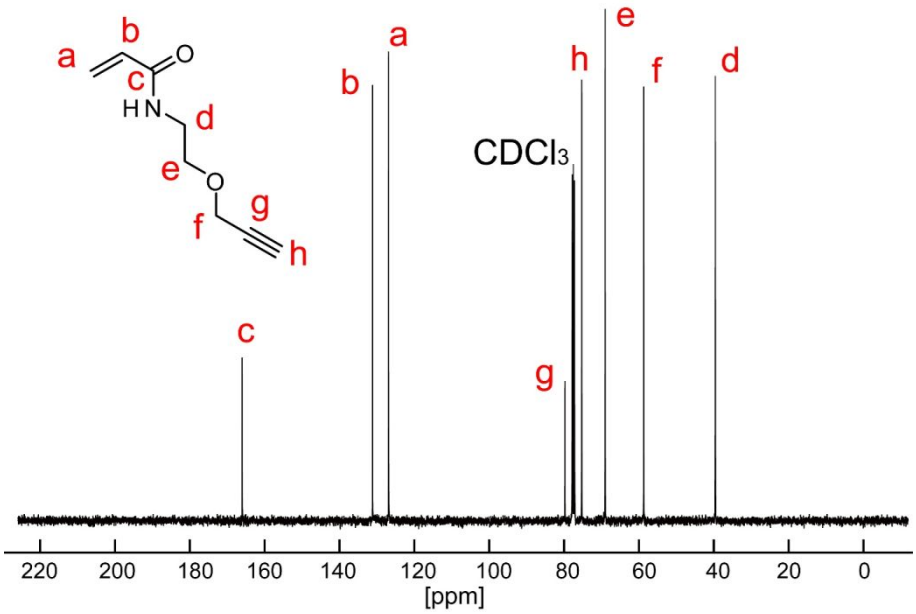

Figure S1-2. ^{13}C NMR spectrum of 2-(2-propynyloxy)ethyl acrylamide (100 MHz, $CDCl_3$).

^{1}H NMR (400 MHz, $CDCl_3$) δ in ppm: 6.29 (dd, 1H, *trans* C**H**$_2$=CH), 6.10 (dd, 1H, CH$_2$=C**H**), 5.99 (br, 1H, amide), 5.65 (dd, 1H, *cis* C**H**$_2$=CH), 4.17 (d, 2H, O–C**H**$_2$–C≡), 3.64 (t, 2H, HN–C**H**$_2$–CH$_2$–O), 3.57 (t, 2H, HN–CH$_2$–C**H**$_2$–O), 2.46 (t, 1H, –C≡C**H**). ^{13}C NMR (100 MHz, $CDCl_3$) δ in ppm: 165.7 (C=O), 130.9 (CH$_2$=**C**H), 126.6 (**C**H$_2$=CH), 79.4 (–**C**≡CH), 75.0 (–C≡**C**H), 68.7 (–C–**C**–O), 58.4 (O–**C**—C≡CH), 39.2 (HN–**C**–C). Mass spectrometry (+ESI-MS) m/z: 329.18 [2M+Na].

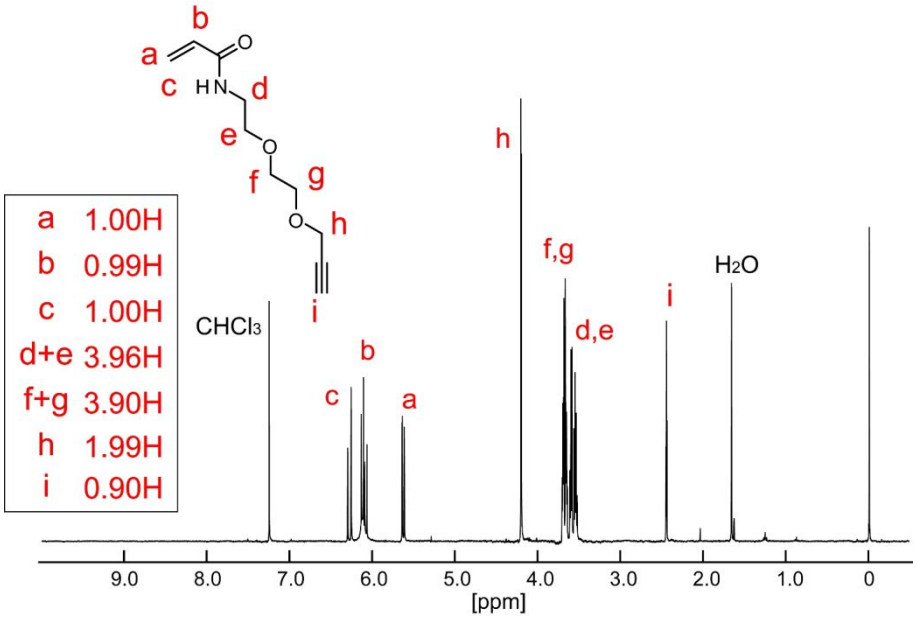

Figure S1-3. ^{1}H NMR spectrum of 2-[2-(2-Propynyloxy)ethoxy]ethyl acrylamide (400 MHz, $CDCl_3$).

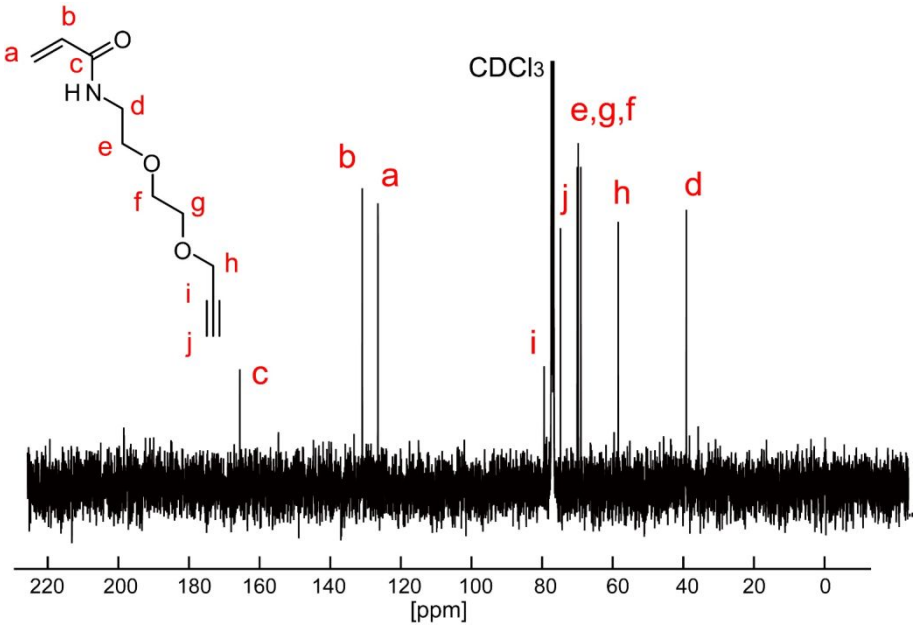

Figure S1-4. ^{13}C NMR spectrum of 2-[2-(2-Propynyloxy)ethoxy]ethyl acrylamide (100 MHz, $CDCl_3$).

^{1}H NMR (400 MHz, $CDCl_3$) δ in ppm: 6.28 (dd, 1H, *trans* C**H**$_2$=CH), 6.10 (dd, 1H, CH$_2$=C**H**), 6.10 (br, 1H, amide), 5.63 (dd, 1H, *cis* C**H**$_2$=CH), 4.20 (d, 2H, O–C**H**$_2$–C≡), 3.69 (m, 2H, –C**H**$_2$–CH$_2$–O–CH$_2$–C≡CH), 3.65 (m, 2H, –CH$_2$–C**H**$_2$–O–CH$_2$–C≡CH), 3.60 (t, 2H, HN–C**H**$_2$–CH$_2$–O), 3.55 (t, 2H, HN–CH$_2$–C**H**$_2$–O), 2.44 (t, 1H, –C≡C**H**). ^{13}C NMR (100 MHz, $CDCl_3$) δ in ppm: 165.6 (C=O), 131.0 (CH$_2$=**C**H), 126.5 (**C**H$_2$=CH), 79.5 (–**C**≡CH), 74.9 (–C≡**C**H), 70.2, 69.9, 69.1 (HN–C–**C**–O–**C**–**C**–O), 58.5 (O–**C**—C≡CH), 39.3 (HN–**C**–C). Mass spectrometry (+ESI-MS) m/z: 220.19 [M+Na].

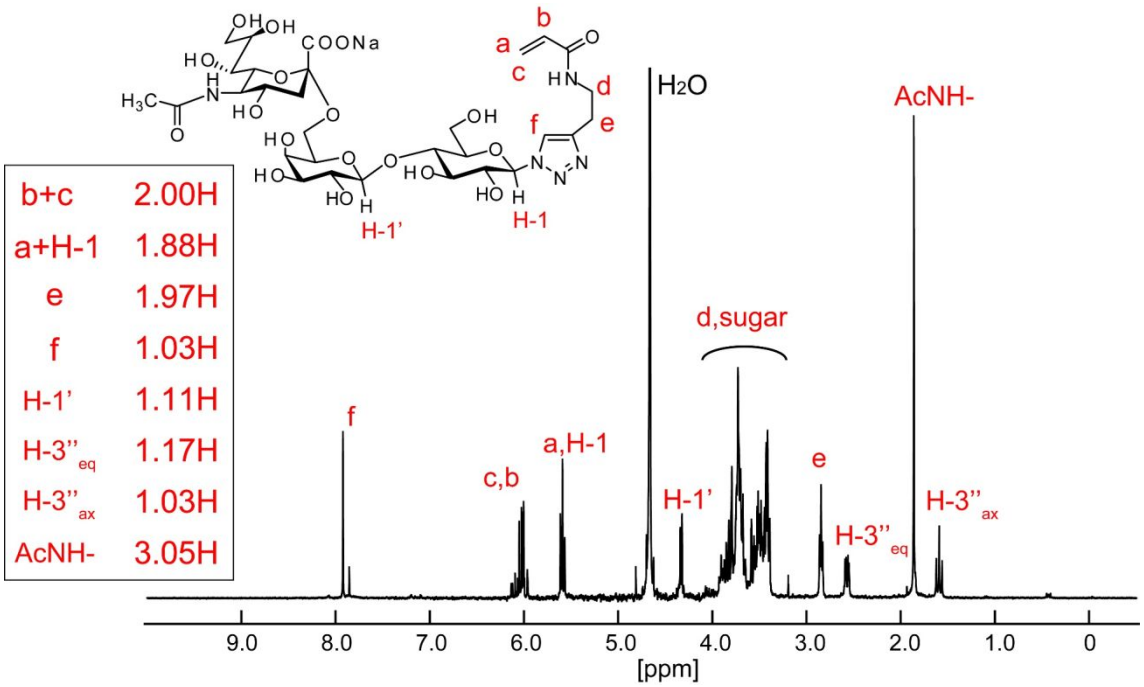

Figure S1-5. ^{1}H NMR spectrum of 6'-SALac AAm (400 MHz, D_2O).

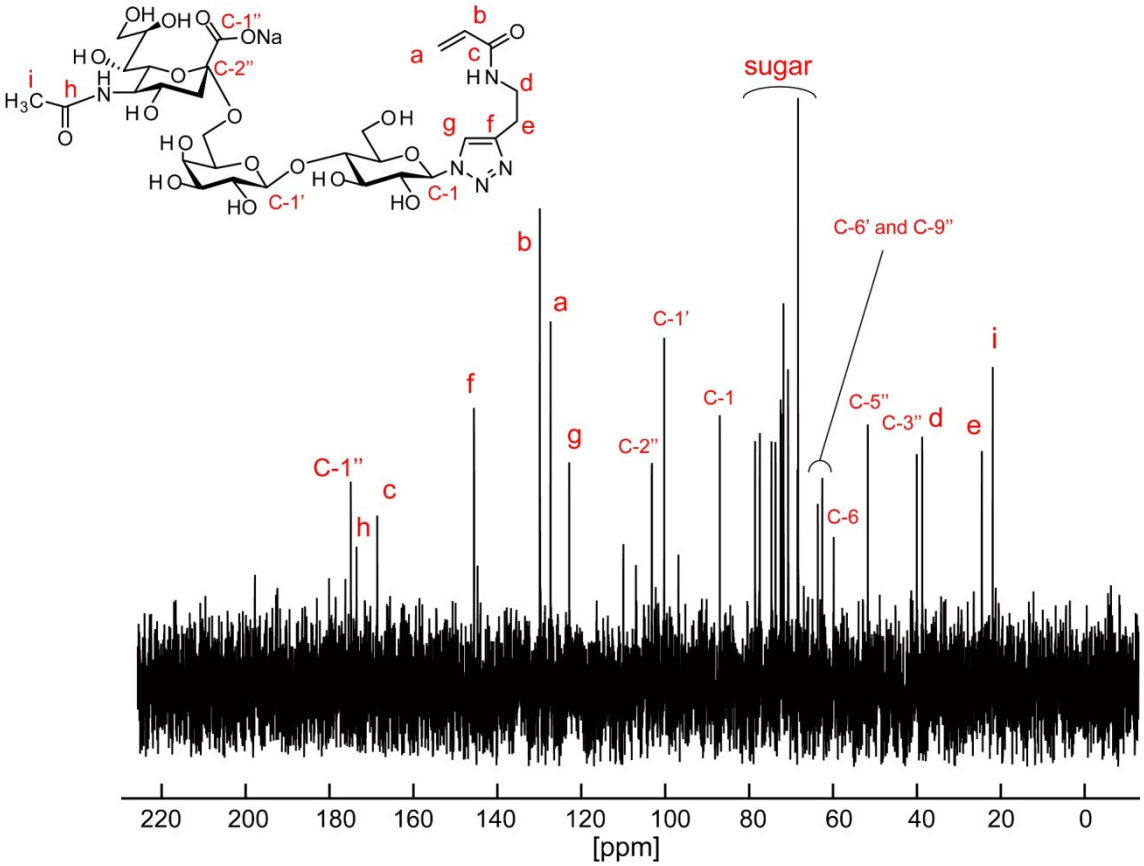

Figure S1-6. ^{13}C NMR spectrum of 6'-SALac AAm (100 MHz, D_2O).

^{1}H NMR (400 MHz, D_2O) δ in ppm: 7.94 (s, 1H, triazole), 6.08 (m, 2H, *trans* C**H**$_2$=CH and CH$_2$=C**H**), 5.62 (m, 2H, *cis* C**H**$_2$=CH and H-1), 4.36 (d, 1H, H-1'), 4.00–3.40 (m, 21H, sugar-H and HN–C**H**$_2$–CH$_2$–), 2.87 (t, 2H, HN–CH$_2$–C**H**$_2$–), 2.60 (dd, 1H, H-3"$_{eq}$), 1.89 (s, 3H, NHCO–C**H**$_3$), 1.62 (t, 1H, H-3"$_{ax}$). ^{13}C NMR (100 MHz, D_2O) δ in ppm: 174.9 (O=**C**–ONa), 173.5 (CH$_3$–**C**ONH), 168.5 (**C**ONH, amide), 145.5 (C=**C**(NH)–CH$_2$–, triazole), 129.9 (CH$_2$=**C**H), 127.3 (**C**H$_2$=CH), 122.9 (sugar–N–**C**=C, triazole), 103.2 (C-2"), 100.3 (C-1'), 87.0 (C-1), 78.6, 77.5, 74.7, 73.8, 72.5, 72.3, 71.9, 70.8, 68.5, 68.4 (sugar-C), 63.7, 62.6 (C-6' and C-9"), 59.9 (C-6), 51.8 (C-5"), 40.1 (C-3"), 38.8 (HN–**C**–C–triazole), 24.6 (HN–C–**C**–triazole), 22.0 (NHCO–**C**H$_3$). Mass spectrometry (+ESI-MS) m/z: 804.3 [M+H], 826.4 [M+Na].

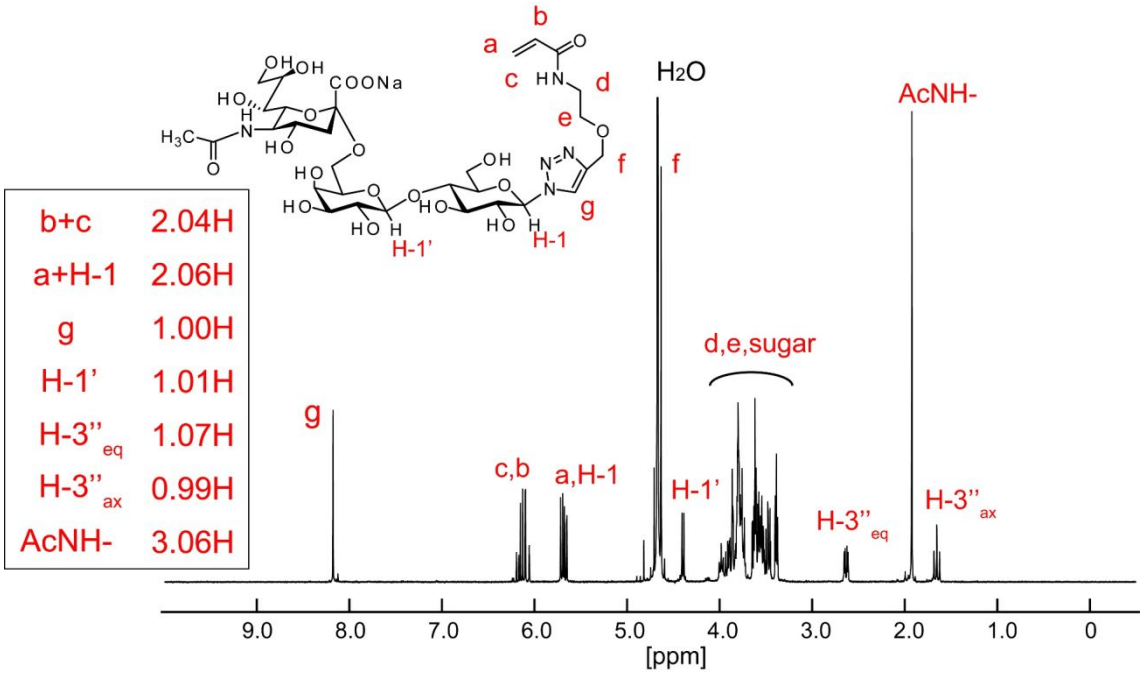

Figure S1-7. ^{1}H NMR spectrum of 6'-SALac-EG-AAm (400 MHz, D_2O).

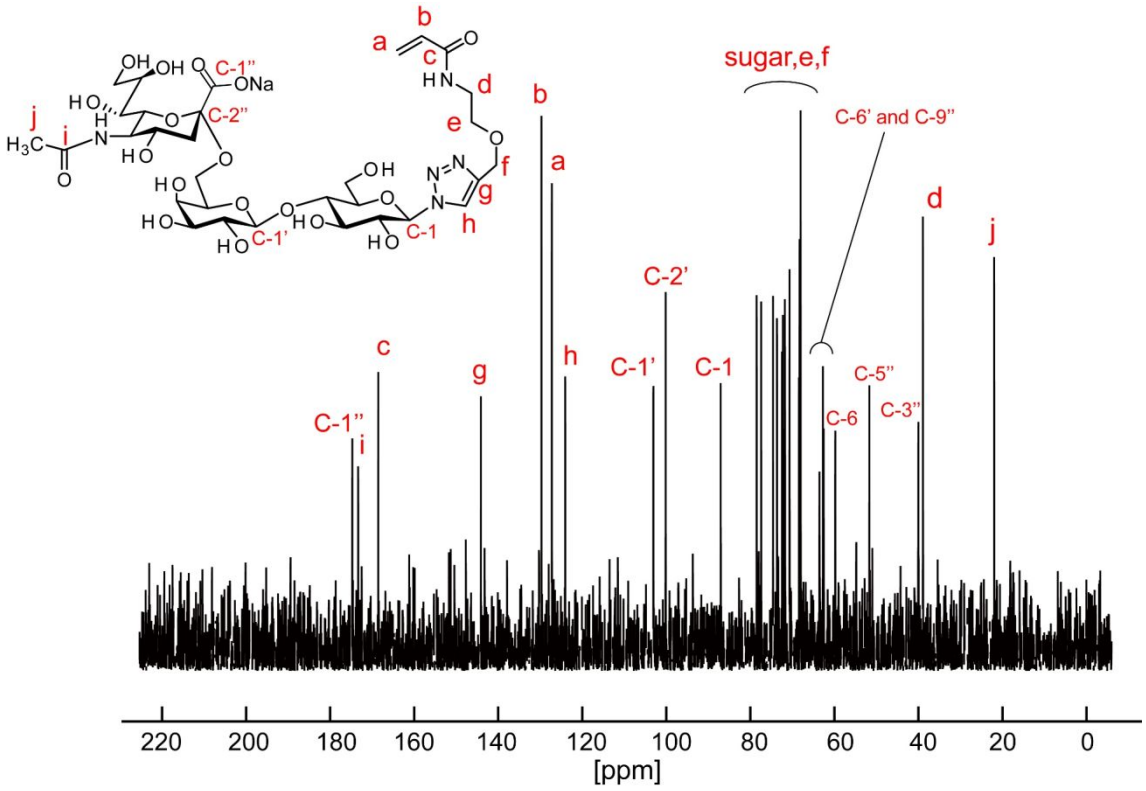

Figure S1-8. ^{13}C NMR spectrum of 6'-SALac-EG-AAm (100 MHz, D_2O).

^{1}H NMR (400 MHz, D_2O) δ in ppm: 8.18 (s, 1H, triazole), 6.15 (dd, 1H, *trans* C**H**$_2$=CH), 6.10 (dd, 1H, CH$_2$=C**H**), 5.70 (m, 2H, *cis* C**H**$_2$=CH and H-1), 4.63 (O–C**H**$_2$–triazole), 4.39 (d, 1H, H-1'), 4.00–3.45 (m, 23H, sugar-H and HN–CH$_2$–C**H**$_2$–O), 3.39 (t, 2H, HN–C**H**$_2$–CH$_2$–O), 2.63 (dd, 1H, H-3"$_{eq}$), 1.92 (s, 3H, NHCO–C**H**$_3$), 1.65 (t, 1H, H-3"$_{ax}$). ^{13}C NMR (100 MHz, D_2O) δ in ppm: 174.9 (O=**C**–ONa), 173.5 (CH$_3$–**C**ONH), 168.8 (**C**ONH, amide), 144.3 (C=**C**(NH)–CH$_2$–O, triazole), 129.9 (CH$_2$=**C**H), 127.4 (**C**H$_2$=CH), 124.2 (sugar–N–**C**=C, triazole), 103.3 (C-1'), 100.3 (C-2'), 87.2 (C-1), 78.7, 77.6, 74.8, 73.8, 72.6, 72.4, 71.9, 70.8, 68.6 (sugar-C), 68.4 (HN–C–**C**–O), 68.2 (–O–**C**–triazole), 63.7, 62.7 (C-6' and C-9"), 60.0 (C-6), 51.8 (C-5"), 40.2 (C-3"), 39.1 (HN–**C**–C–O), 22.1 (NHCO–**C**H$_3$). Mass spectrometry (+ESI-MS) m/z: 834.59 [M+H], 856.60 [M+Na].

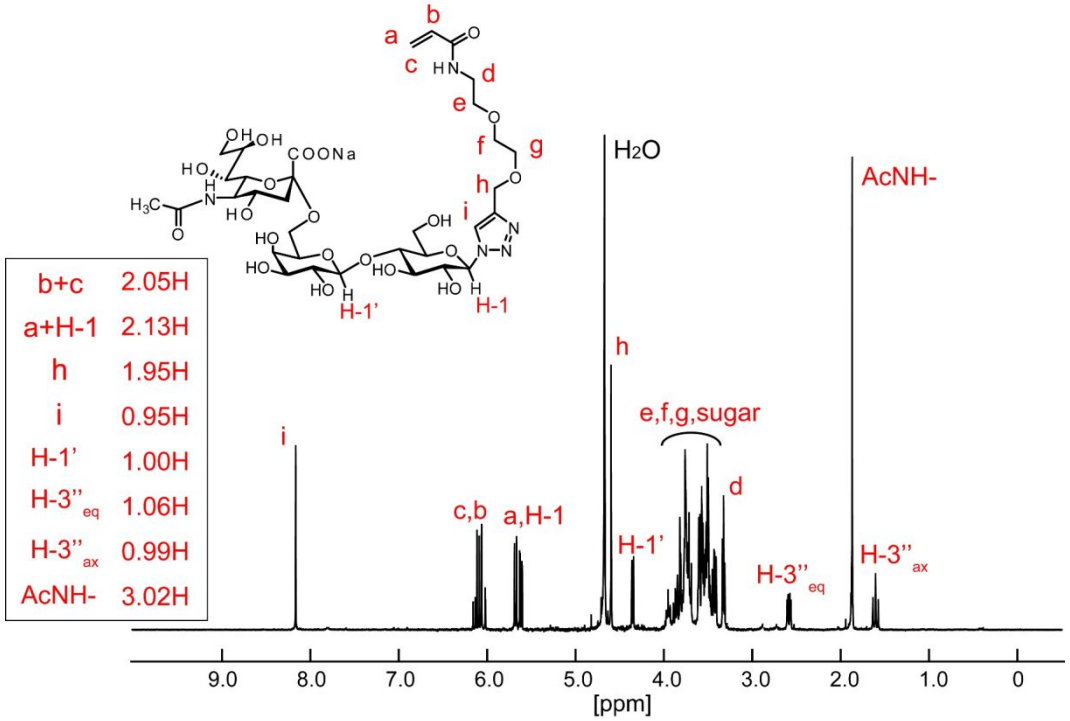

Figure S1-9. ^{1}H NMR spectrum of 6'-SALac-EG$_2$-AAm (400 MHz, D_2O).

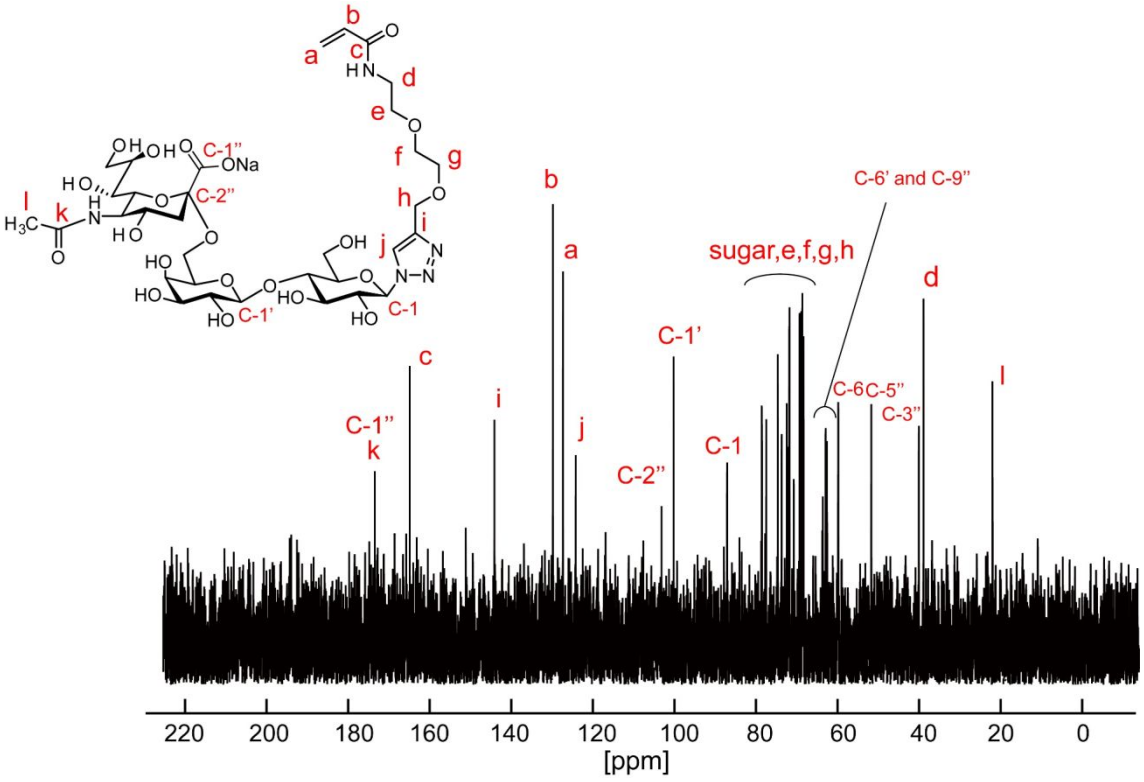

Figure S1-10. ^{13}C NMR spectrum of 6'-SALac-EG$_2$-AAm (100 MHz, D_2O).

^{1}H NMR (400 MHz, D_2O) δ in ppm: 8.15 (s, 1H, triazole), 6.12 (dd, 1H, *trans* C**H**$_2$=CH), 6.04 (dd, 1H, CH$_2$=C**H**), 5.67 (d, 1H, H-1), 5.61 (dd, 1H, *cis* C**H**$_2$=CH), 4.60 (O–C**H**$_2$–triazole), 4.35 (d, 1H, H-1'), 4.00–3.41 (m, 27H, sugar-H, HN–CH$_2$–C**H**$_2$–O, –O–C**H**$_2$–C**H**$_2$–O–), 3.33 (t, 2H, HN–C**H**$_2$–CH$_2$–O), 2.59 (dd, 1H, H-3"$_{eq}$), 1.88 (s, 3H, NHCO–C**H**$_3$), 1.61 (t, 1H, H-3"$_{ax}$). ^{13}C NMR (100 MHz, D_2O) δ in ppm: 173.5 (CH$_3$–**C**ONH), 165.0 (**C**ONH, amide), 144.2 (C=**C**(NH)–CH$_2$–O, triazole), 129.9 (CH$_2$=**C**H), 127.4 (**C**H$_2$=CH), 124.3 (sugar–N–**C**=C, triazole), 103.2 (C-1'), 100.3 (C-2'), 87.2 (C-1), 78.6, 77.6, 74.7, 73.8, 72.5, 72.4, 71.9, 70.8 (sugar-C), 69.4, 69.0 (–O–**C**–**C**–O–), 68.7 (sugar-C), 68.6 (HN–C–**C**–O), 68.4 (–O–**C**–triazole), 63.7, 62.7 (C-6' and C-9"), 59.9 (C-6), 51.8 (C-5"), 40.1 (C-3"), 39.0 (HN–**C**–C–O), 22.0 (NHCO–**C**H$_3$). Mass spectrometry (+ESI-MS) m/z: 878.3 [M+H], 900.4 [M+Na].

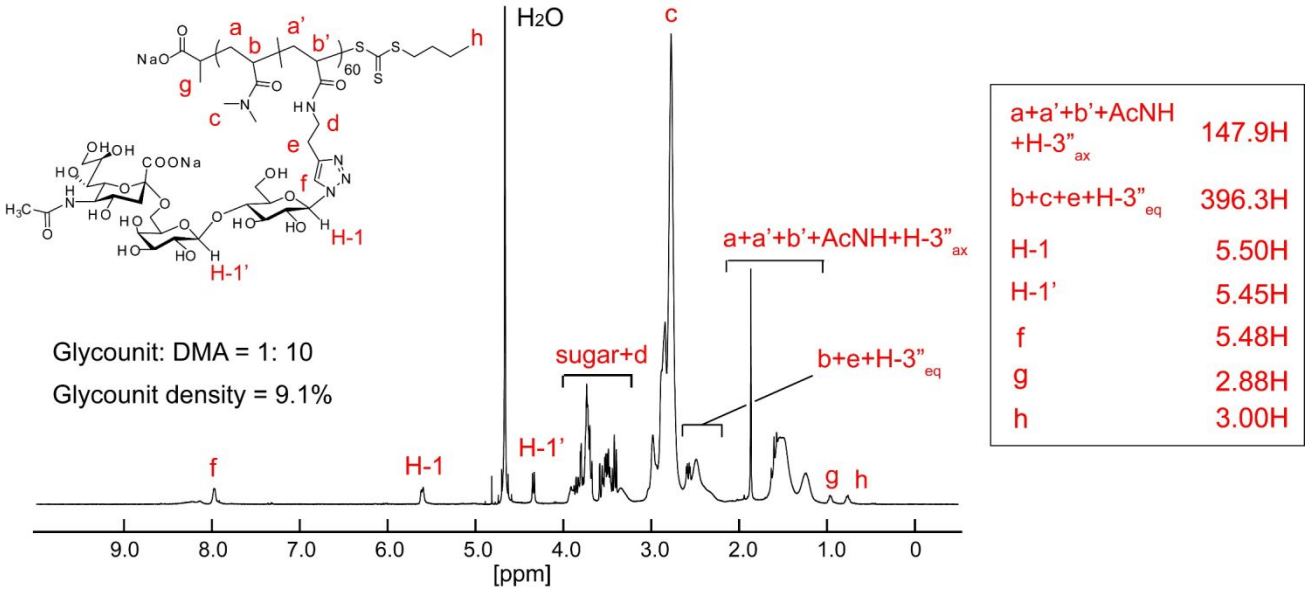

Figure S1-11. ^{1}H NMR spectrum of **S10** (400 MHz, D_2O).

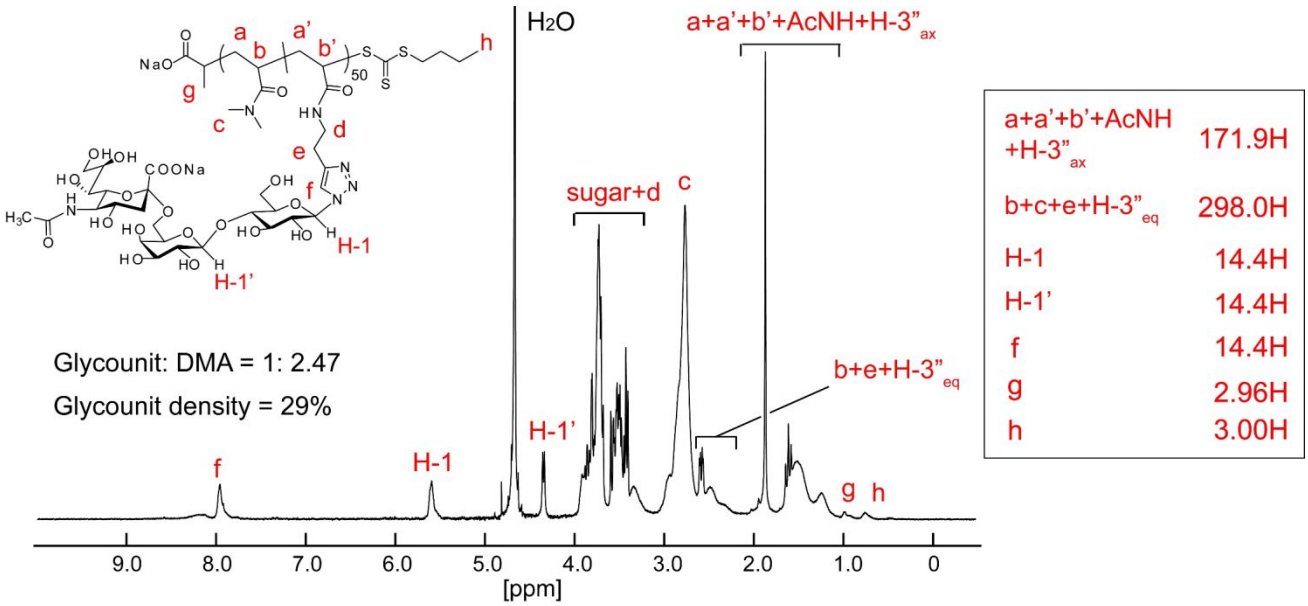

Figure S1-12. ^{1}H NMR spectrum of **S30** (400 MHz, D_2O).

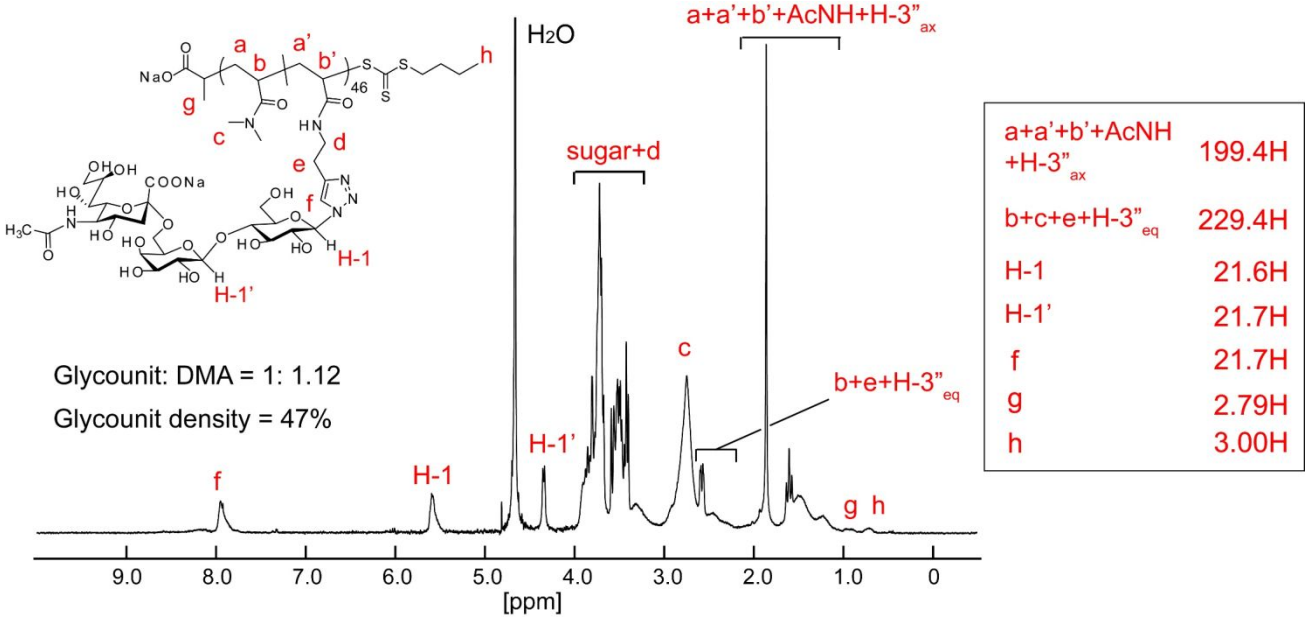

Figure S1-13. ^{1}H NMR spectrum of **S50** (400 MHz, D_2O).

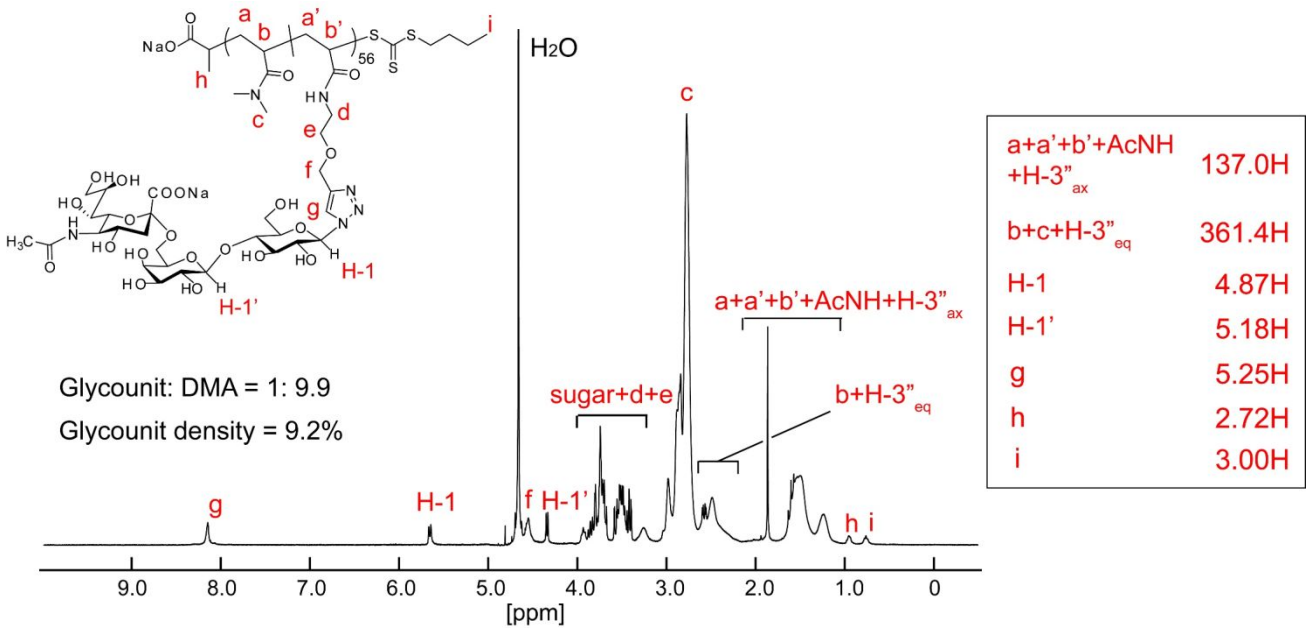

Figure S1-14. ^{1}H NMR spectrum of **M10** (400 MHz, D_2O).

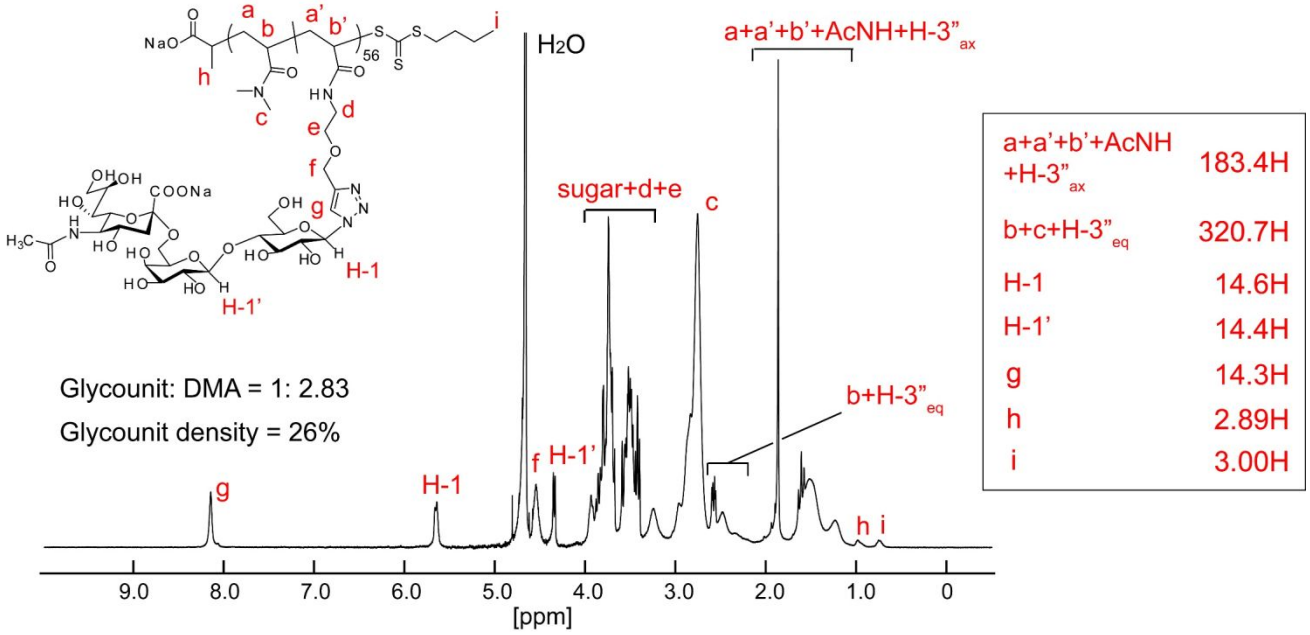

Figure S1-15. ^{1}H NMR spectrum of **M30** (400 MHz, D_2O).

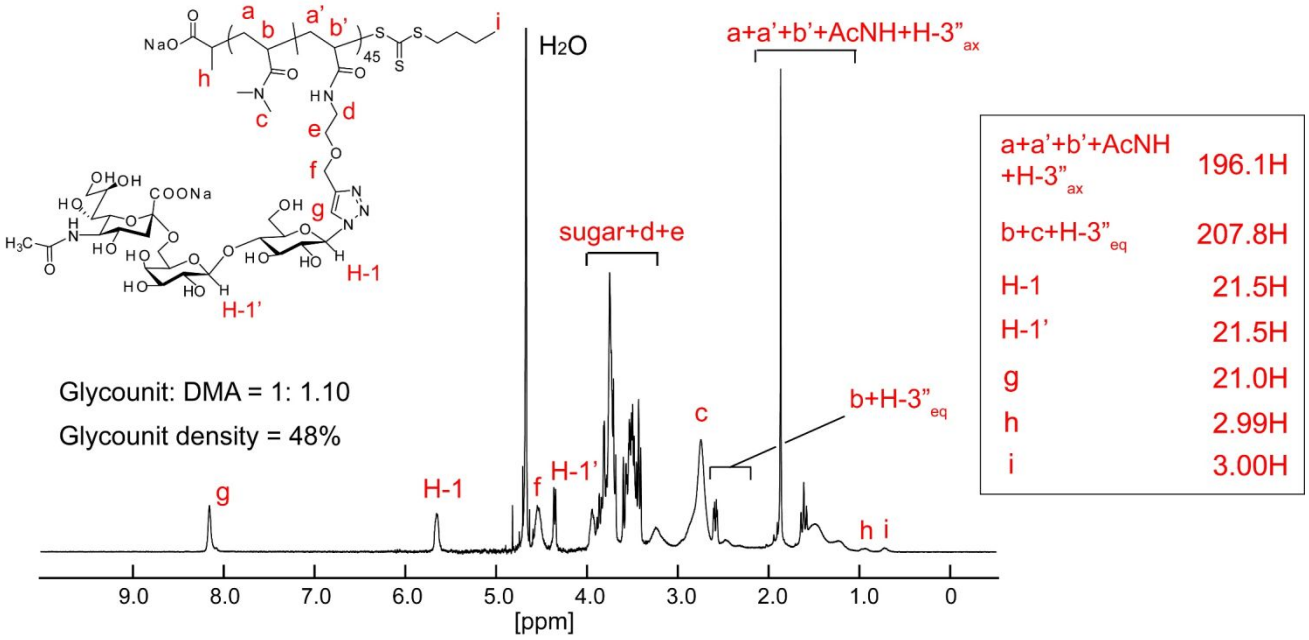

Figure S1-16. ^{1}H NMR spectrum of **M50** (400 MHz, D_2O).

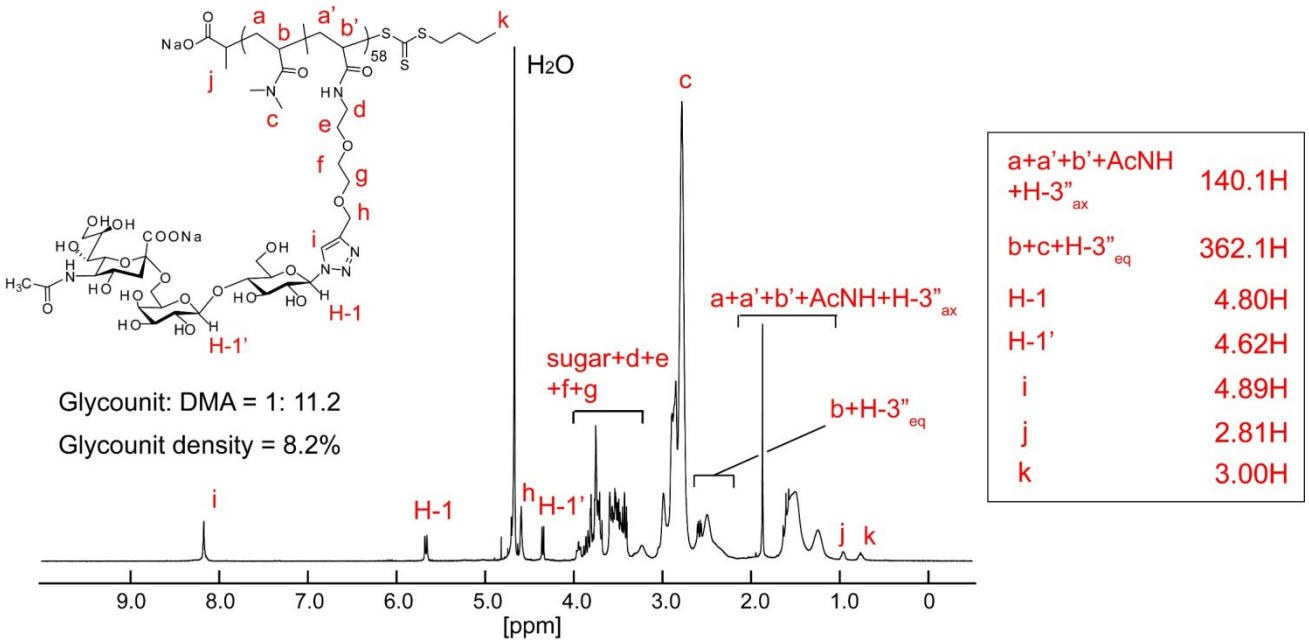

Figure S1-17. ^{1}H NMR spectrum of **L10** (400 MHz, D_2O).

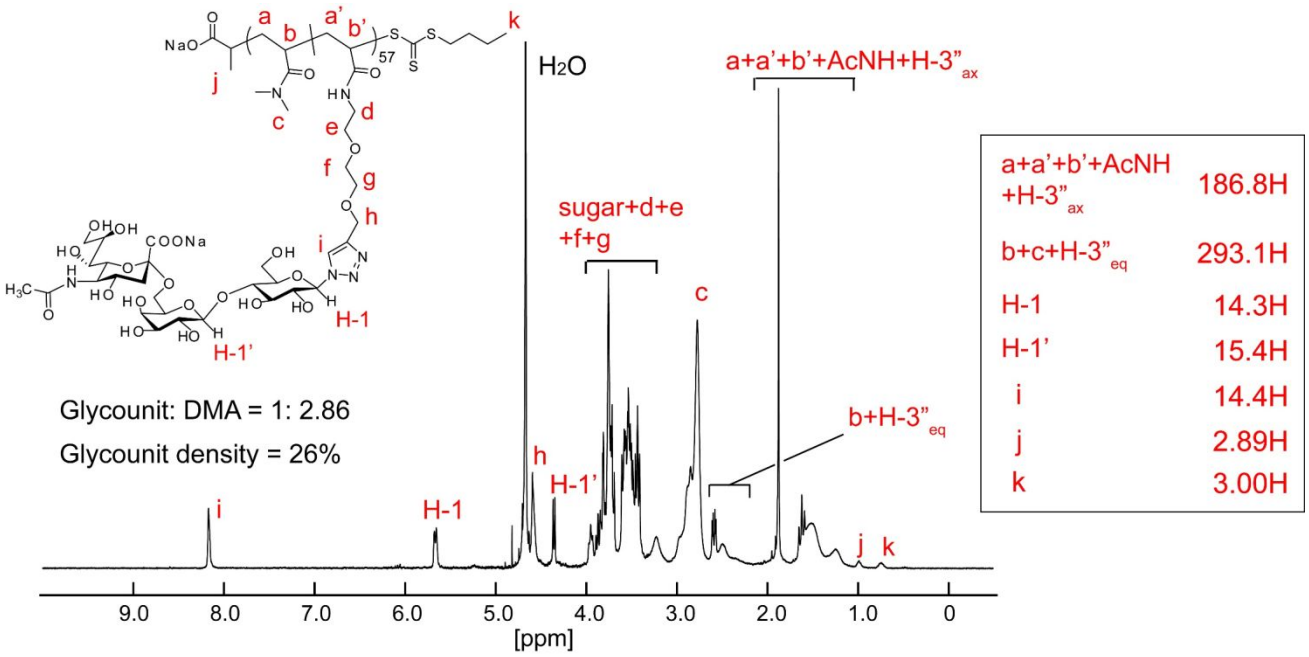

Figure S1-18. ^{1}H NMR spectrum of **L30** (400 MHz, D_2O).

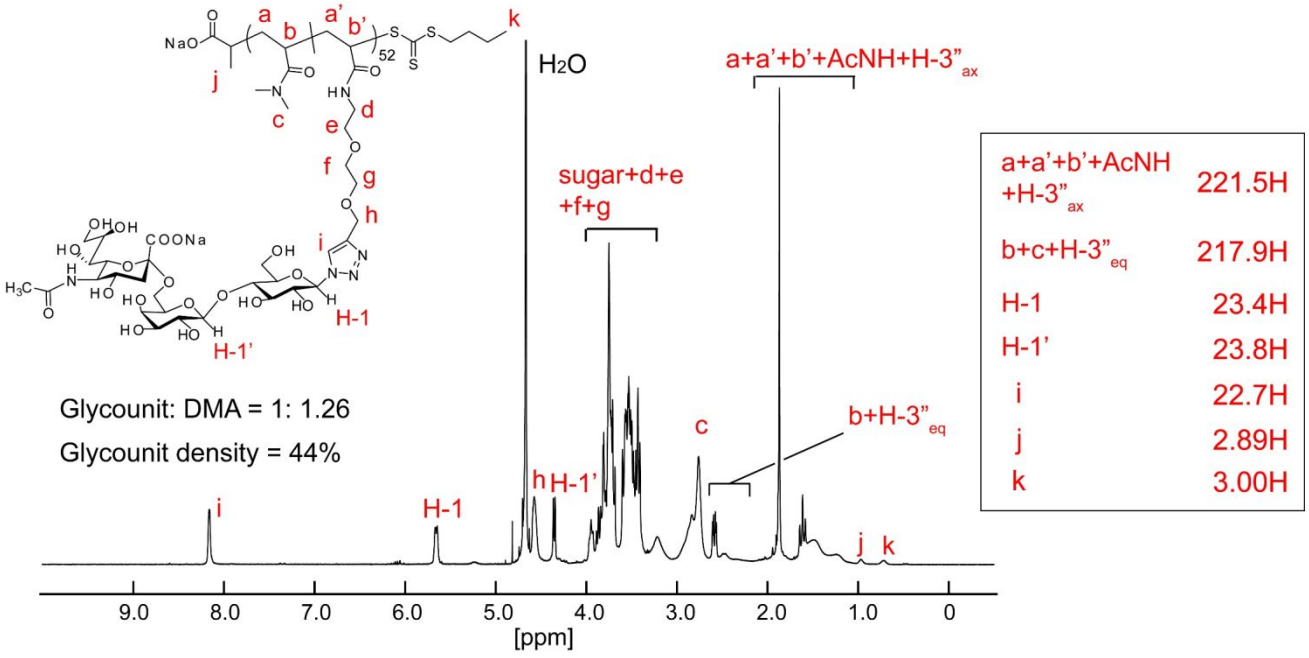

Figure S1-19. ^{1}H NMR spectrum of **L50** (400 MHz, D_2O).

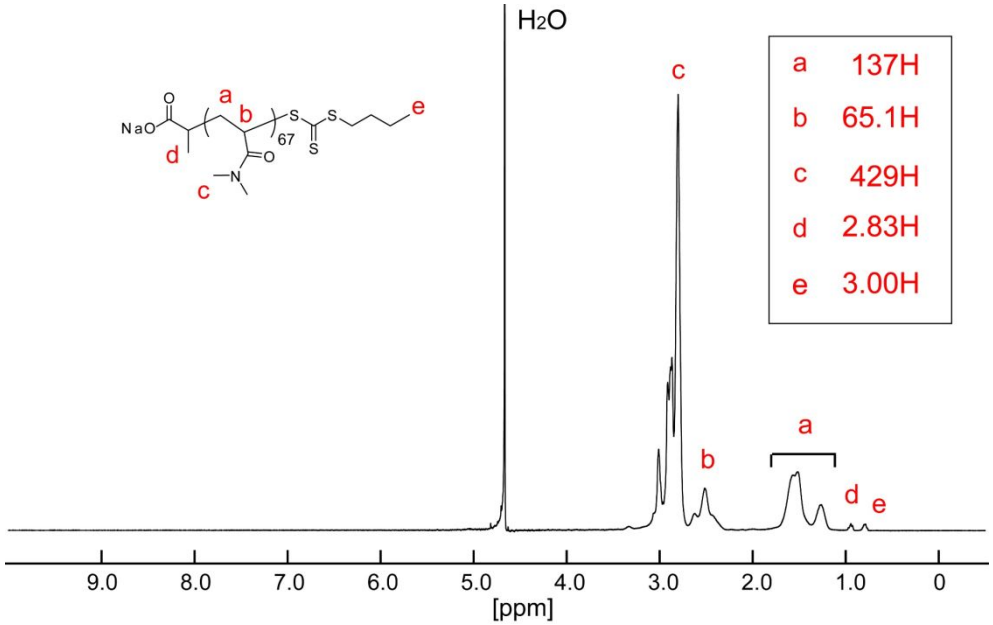

Figure S1-20. ^{1}H NMR spectrum of **PDMA** (400 MHz, D_2O)

^{1}H NMR (400 MHz, D_2O) δ in ppm: 3.05–2.70 (OCN(C**H**$_3$)$_2$), 2.70–2.30 (brs, –CH– main chain), 1.70–1.20 (brd, –CH_2– main chain), 0.96 (ester–CH(C**H**$_3$)–S), 0.78 (SC_3H_6–C**H**$_3$).

Table S1. Detailed condition of the RAFT polymerization for the glycopolymers.

Entry	DMA	6'-SALacAAm (S)	NaBTPA	AIPD	[M]	DMA	H2O	DMF	Time	Conv. (%)		
	(μmol)	(μmol)	(μmol)	(μmol)	(mol/L)	(μL)	(μL)	(μL)	(min)	DMA	Glyco	Total
PS10	270 (27 mg)	30 (24 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	1.0	28	218	54	180	97	88	96
PS30	210 (21 mg)	90 (72 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	0.9	22	249	62	180	94	85	91
PS50	150 (14.9 mg)	150 (120 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	0.8	15	288	72	180	96	73	85

Entry	DMA	6'-SALac-EG-AAm (M)	NaBTPA	AIPD	[M]	DMA	H2O	DMF	Time	Conv. (%)		
	(μmol)	(μmol)	(μmol)	(μmol)	(mol/L)	(μL)	(μL)	(μL)	(min)	DMA	Glyco	Total
PM10	270 (27 mg)	30 (25 mg)	6.00 (1.56 mg)	0.12 (0.039 mg)	1.0	28	218	54	180	>99	>99	>99
PM30	210 (21 mg)	90 (75 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	0.9	22	249	62	180	96	95	95.7
PM50	150 (14.9 mg)	150 (125 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	0.8	15	288	72	180	93	76	84.5

Entry	DMA	6'-SALac-EG2-AAm (L)	NaBTPA	AIPD	[M]	DMA	H2O	DMF	Time	Conv. (%)		
	(μmol)	(μmol)	(μmol)	(μmol)	(mol/L)	(μL)	(μL)	(μL)	(min)	DMA	Glyco	Total
PL10	270 (27 mg)	30 (26.3 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	1.0	28	218	54	180	>99	>99	>99
PL30	210 (21 mg)	90 (79 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	0.9	22	249	62	180	95	84	92
PL50	150 (14.9 mg)	150 (132 mg)	6.0 (1.56 mg)	0.12 (0.039 mg)	0.8	15	288	72	180	92	78	86

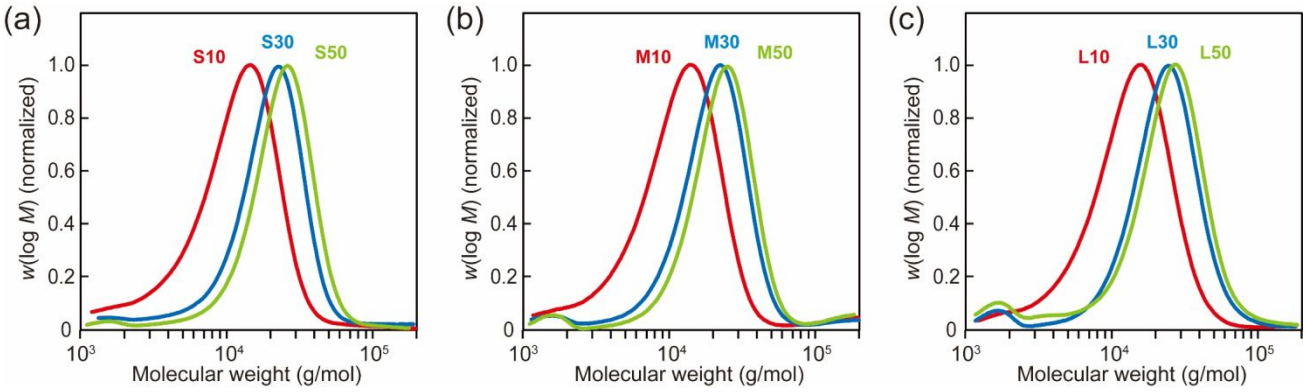

Figure S2. SEC chromatographs of the glycopolymers, calibrated with pullulan standard. The eluent was 100 mM $NaNO_3$ (aq). (a) **PS** polymers, (b) **PM** polymers, and (c) **PL** polymers.

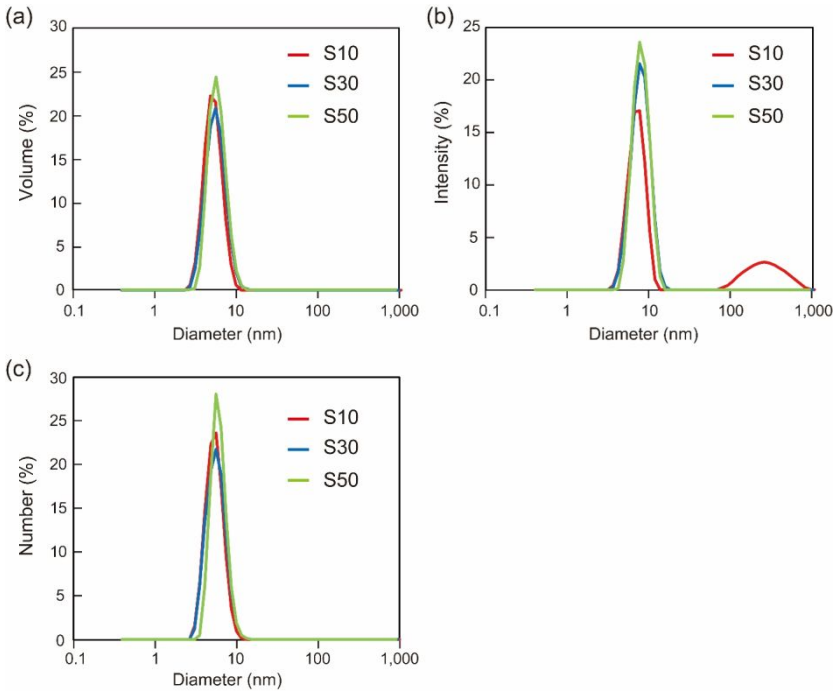

Figure S3-1. Dynamic light scattering distributions of **PS** polymers based on (a) volume, (b) intensity, and (c) number.

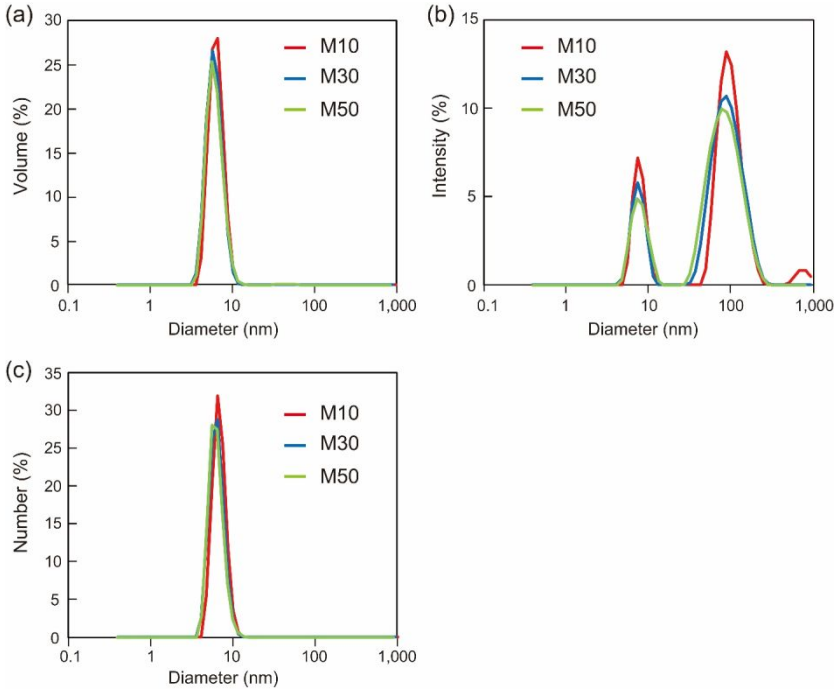

Figure S3-2. Dynamic light scattering distributions of **PM** polymers based on (a) volume, (b) intensity, and (c) number.

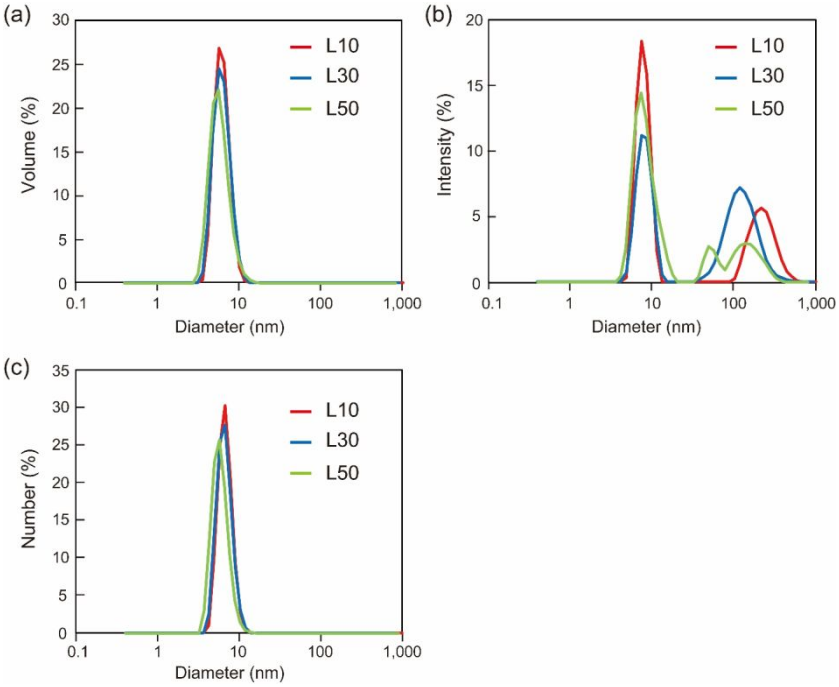

Figure S3-3. Dynamic light scattering distributions of **PL** polymers based on (a) volume, (b) intensity, and (c) number.

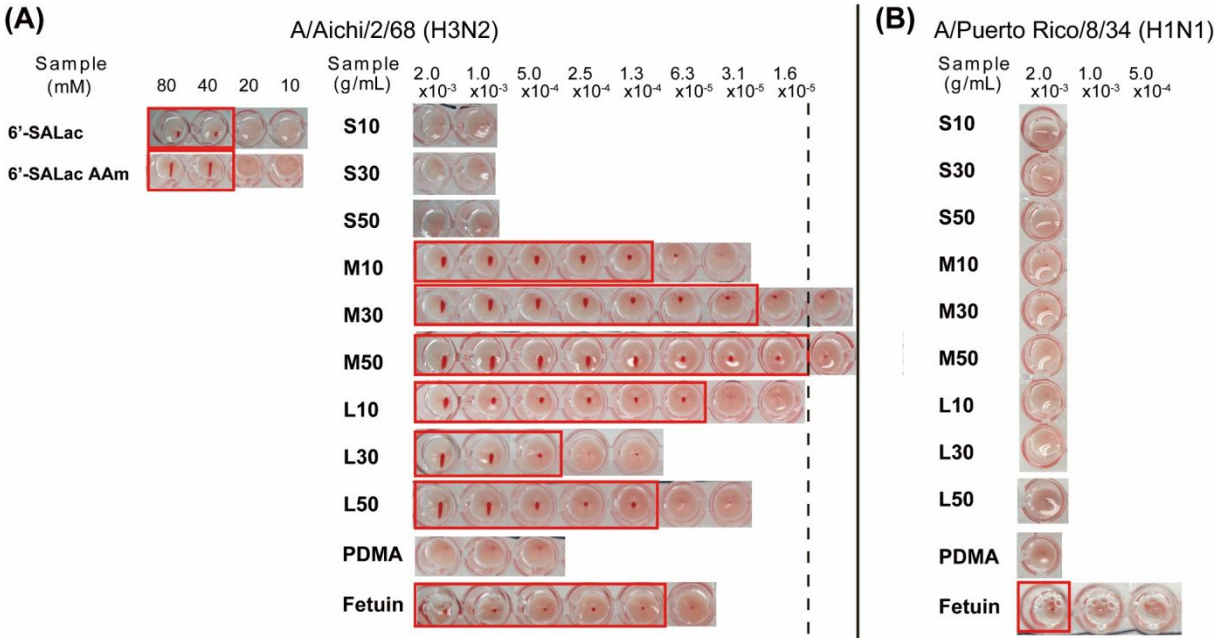

Figure S4. HI assays of the glycopolymers, **PDMA**, 6'-SAlac, glycomonomer (6'-SALac AAm), and fetuin against the influenza viruses [A/Aichi/2/68 (H3N2) (A) and A/Puerto Rico/8/34 (H1N1) (B)]. The images of the 96-well plates after incubation. The red squares show the concentration required for HI activity.