

Supporting Information

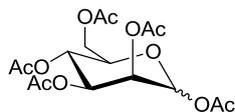
The Synthesis and Lectin-Binding Properties of Novel Mannose-Functionalised Polymers

Hayley Charville, Jianyong Jin*, Clive W. Evans, Margaret A. Brimble and David E. Williams

School of Chemical Sciences, University of Auckland, New Zealand

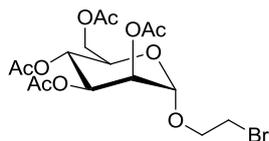
General experimental: Unless stated, all solvents and reagents were used as supplied from commercial sources. hydroxyethyl methacrylate (HEMA) and diethylene glycol dimethacrylate (DEGDMA) were distilled under reduced pressure in order to remove inhibitor prior to use. All moisture sensitive reactions were performed in an inert, dry atmosphere of nitrogen in oven dried glassware. Analytical thin-layer chromatography (TLC) was performed using Kieselgel F254 0.2 mm (Merck) silica plates with visualization by ultraviolet irradiation (254 nm) and/or by staining with vanillin. Flash chromatography was performed using Kieselgel S 63–100 μ m (Riedel-de-Hahn) silica gel. The solvent compositions reported for all chromatographic separations are on a volume/volume (v/v) basis. Infrared (IR) spectra were recorded on a PerkinElmer Spectrum 100 FT-IR Spectrometer. UV-Vis spectra were recorded in a quartz cell (light path 1 cm) on a Jenway 7315 spectrophotometer. UV irradiation was carried out using a UV curing system. The wavelength of illumination ranged from 200 to 400 nm and the irradiation time was 30 min. $^1\text{H-NMR}$ spectra were recorded on a 400 MHz Bruker spectrometer and are reported in parts per million (ppm) on the δ scale relative to CDCl_3 (δ 7.26) and $^{13}\text{C-NMR}$ spectra were recorded on a 100 MHz Bruker spectrometer and are reported in parts per million (ppm) on the δ scale relative to CDCl_3 (δ 77.16). The multiplicities of ^1H signals are designated by the following abbreviations: s = singlet; d = doublet; t = triplet; q = quartet; m = multiplet; br = broad; dd doublet of doublets; All coupling constants J are reported in hertz. Mass spectra were obtained by electrospray ionization in positive ion mode. Dialysis was performed using Spectra/Por dialysis tubing (MWCO = 1000). For polymer molecular weight measurement, the SEC system consisted of a Waters 515 HPLC pump, a Degassex DG-4400 on-line degasser connected to a TSK Gel Super AWM-H SEC column (9 μ m, 6 x 150mm) with guard, and 0.5 μ m in-line filter, a Rheodyne manual injector, and a Waters column oven. The eluent was THF and the flow rate was 0.5 mL/min. Sample concentrations were 5 mg/mL and the injection volume was 200 μ L. All solutions were filtered through 0.45 μ m syringe filters before injection. Data acquisition and processing were performed using the ASTRA 4 software (Wyatt Technologies Corporation). The two detectors used were the Shimadzu RID-10A Differential Refractive Index detector and the DAWN DSP Light scattering detector with 18 angles (Wyatt Technologies Inc.). The columns and RI detector were maintained at 35°C. The dn/dc value of 0.083 ml/g for PMMA in THF was used, obtained from the literature (American Polymer Standards Corporation, <http://www.ampolymer.com/dndc.html>).

Synthesis of 1,2,3,4,6-penta-O-acetyl- α -D-mannopyranoside (2)



D-Mannose (7 g, 38.9 mmol) was added (in five portions) to a stirred solution of iodine (0.4 g, 1.55 mmol) in acetic anhydride (40 mL, 432 mmol) at 0 °C. The reaction mixture was allowed to stir at this temperature for 30 minutes and then allowed to warm up to room temperature. After 1 h at room temperature, methanol (20 mL) was added and the mixture was stirred for a further 30 minutes, followed by evaporation of the solvent *in vacuo*. The residue was dissolved in DCM (50 mL) and washed with Na₂S₂O₅ solution (2 × 50 mL). The layers were separated and the resulting colourless organic phase was washed with NaHCO₃ solution (4 × 50 mL), dried (MgSO₄) and concentrated under reduced pressure to afford a pale yellow oil (12.9 g, 85%) as an anomeric mixture (α : β , 4:1) as determined by ¹H NMR. This was used in the next step without any further purification. δ_{H} (400 MHz, CDCl₃): 1.97 (s, 3H), 2.02 (s, 3H), 2.06 (s, 3H), 2.14(s, 3H), 2.15 (s, 3H), 4.01-4.13 (m, 2H), 4.23-4.29 (m, 1H), 5.22-5.32 (m, 3H), 6.05 (d, J = 1.9). Resonances for minor β anomer: δ_{H} (400 MHz, CDCl₃): 2.00 (s, 3H), 2.07 (s, 3H), 2.18 (s, 3H), 3.77-3.81 (m, 1H), 5.10 (dd, J 6.6, 3.4, 1H), 5.43-5.46 (m, 1H), 5.83 (d, J = 1.2). Data was consistent with that reported in the literature.¹

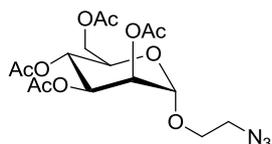
Synthesis of 2-bromoethyl 2,3,4,6-tetra-O-acetyl- α -D-mannopyranoside (3)



BF₃·Et₂O (7.9 mL, 64.05 mmol) was added dropwise to a solution of 1,2,3,4,6-penta-O-acetyl- α -D-mannopyranoside **2** (5 g, 12.81 mmol) and 2-bromoethanol (1.82 mL, 25.62 mmol) in dry DCM (80 mL) at 0 °C. After 1 h the ice bath was removed and the reaction continued at room temperature overnight. The reaction mixture was then slowly added to cold water (80 mL) and the organic layer was separated. The aqueous layer was extracted with DCM (50 mL) and the organic layers combined and washed successively with water (100 mL), sat. NaHCO₃ (2 × 100 mL), water (100 mL). The solution was dried over MgSO₄ and concentrated under reduced pressure to afford a yellow oil. The product was precipitated from diethyl ether to produce a white powdery solid which was filtered, washed with diethyl ether and dried under high vacuum. (4.03 g, 65%). δ_{H} (400 MHz, CDCl₃): 1.99 (s,

3H), 2.05 (s, 3H), 2.11 (s, 3H), 2.16 (s, 3H), 3.50-3.53 (m, 2H), 3.79-3.91 (m, 2H), 4.10-4.15 (m, 2H), 4.24-4.29 (m, 1H), 4.87 (d, $J = 1.7$), 5.26-5.33 (m, 3H). δ_c (100 MHz, CDCl_3): 20.6, 20.7, 20.8, 20.9, 29.6, 62.4, 66.0, 68.5, 68.9, 69.0, 69.4, 97.8, 169.7, 169.8, 170.0, 170.6. Data is consistent with that reported in the literature.²

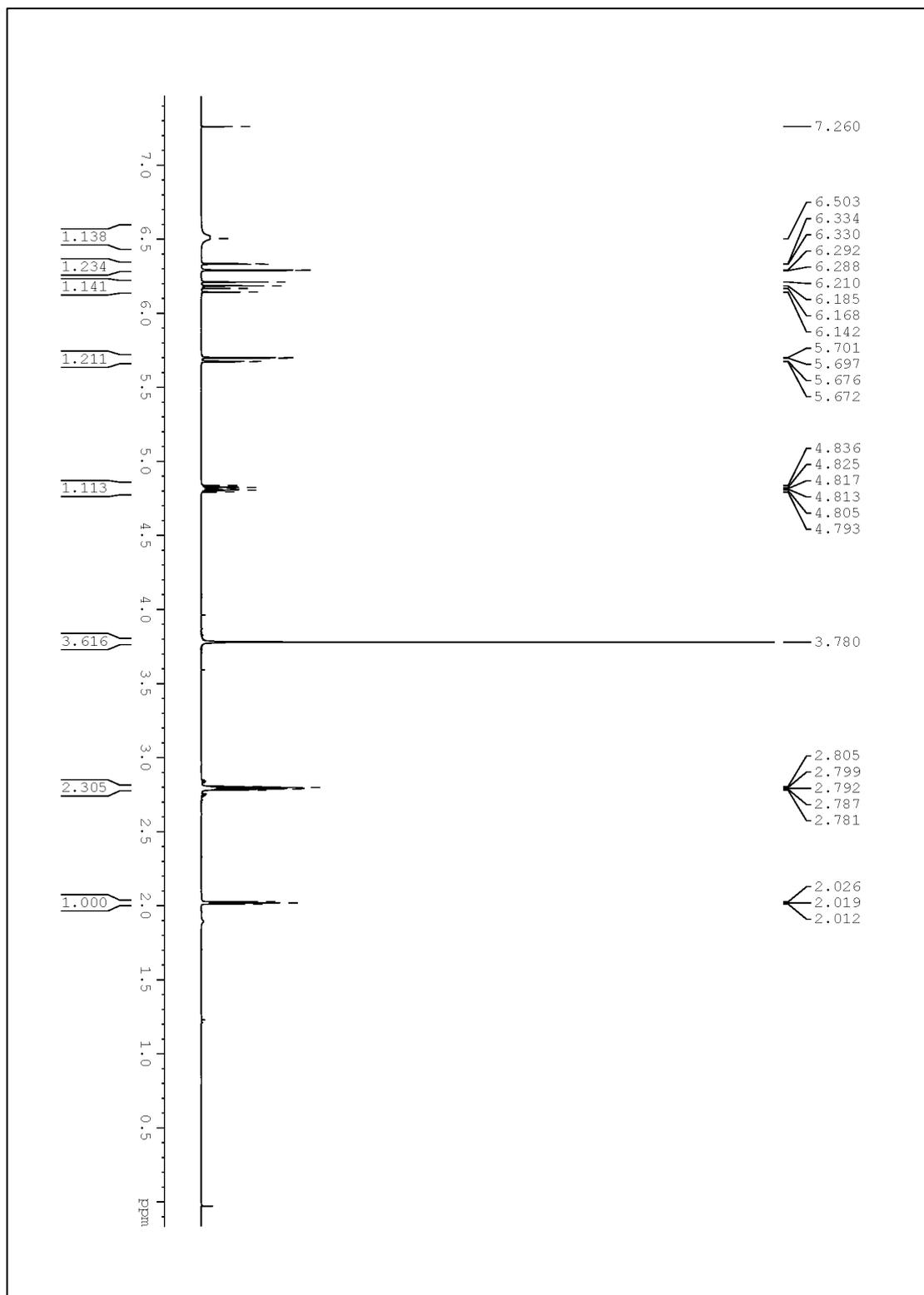
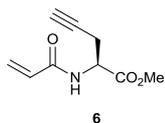
Synthesis of 2-azidoethyl 2,3,4,6-tetra-O-acetyl- α -D-mannopyranoside (**4**)



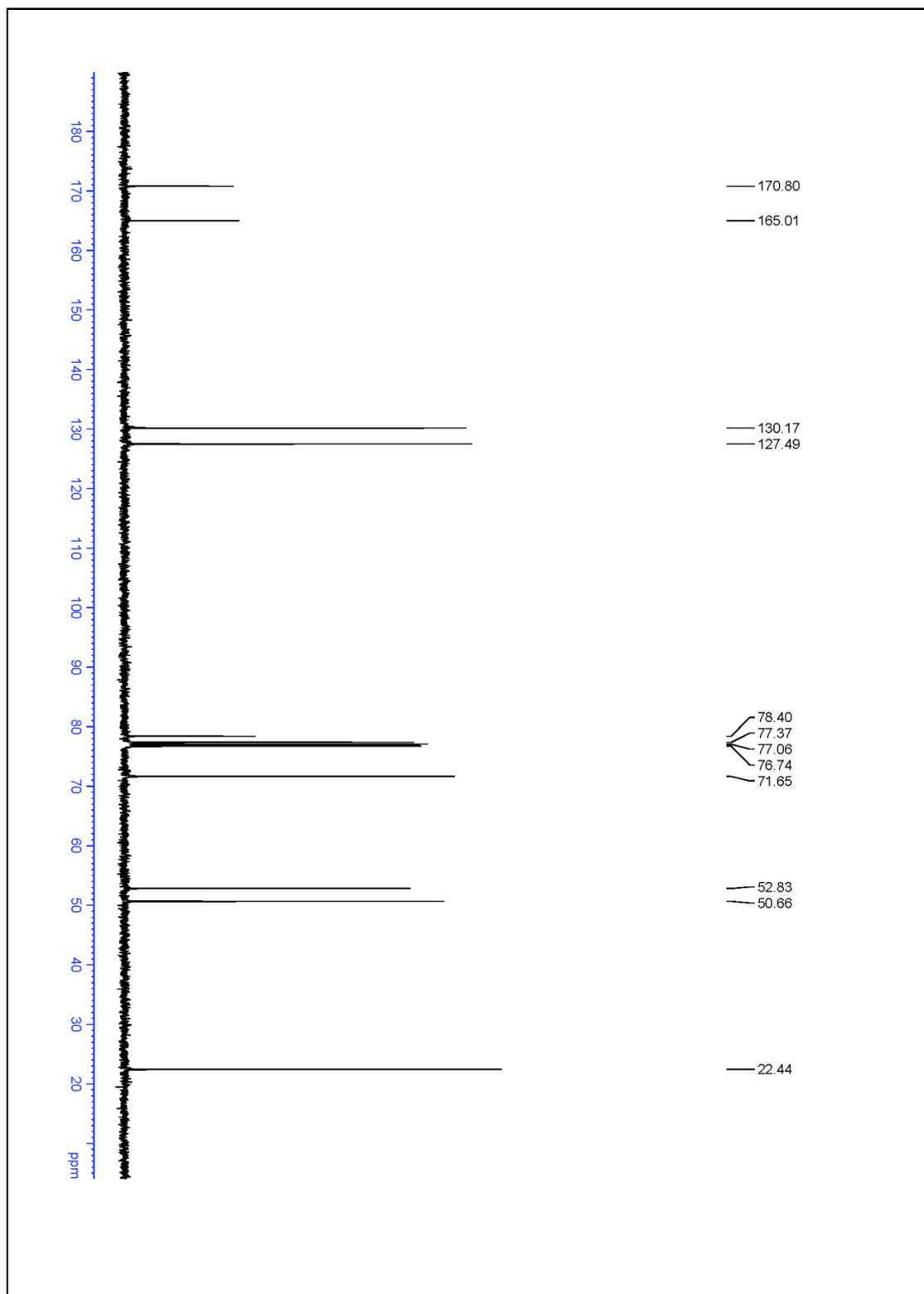
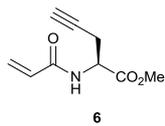
2-bromoethyl 2,3,4,6-tetra-O-acetyl- α -D-mannopyranoside **3** (2 g, 4.4 mmol) and sodium azide (2.28 g, 35.12 mmol) were dissolved in dry DMF (60 mL) and the reaction was stirred under N_2 at 60 °C for 6 h before cooling to room temperature and stirring overnight. The following day the reaction mixture was poured into ethyl acetate (150 mL) and washed with water (3×100 mL) and brine (2×100 mL). The organic layer was dried (MgSO_4), filtered and concentrated under vacuum. The crude product was purified by silica gel column chromatography (ethyl acetate : hexane, 2 : 1) to afford the title compound as white crystals (1.59 g, 87%). δ_H (400 MHz, CDCl_3): 1.99 (s, 3H), 2.05 (s, 3H), 2.12 (s, 3H), 2.16 (s, 3H), 3.46-3.49 (m, 2H), 3.65-3.70 (m, 1H), 3.85-3.89 (m, 1H), 4.02-4.15 (m, 2H), 4.27-4.31 (dd, $J = 5.4$ and 12.1), 4.87 (d, $J = 1.5$), 5.27-5.35 (m, 3H). δ_c (100 MHz, CDCl_3): 20.6, 20.7, 20.7, 20.9, 50.5, 62.5, 66.0, 67.0, 68.9, 69.4, 97.8, 169.8, 169.9, 170.2, 170.8. Data is consistent with that already in the literature.³

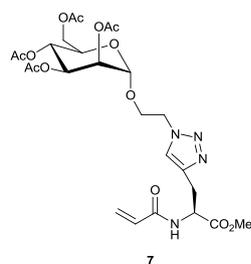
Spectra

Alkyne **6** ^1H NMR (400 MHz, CDCl_3)

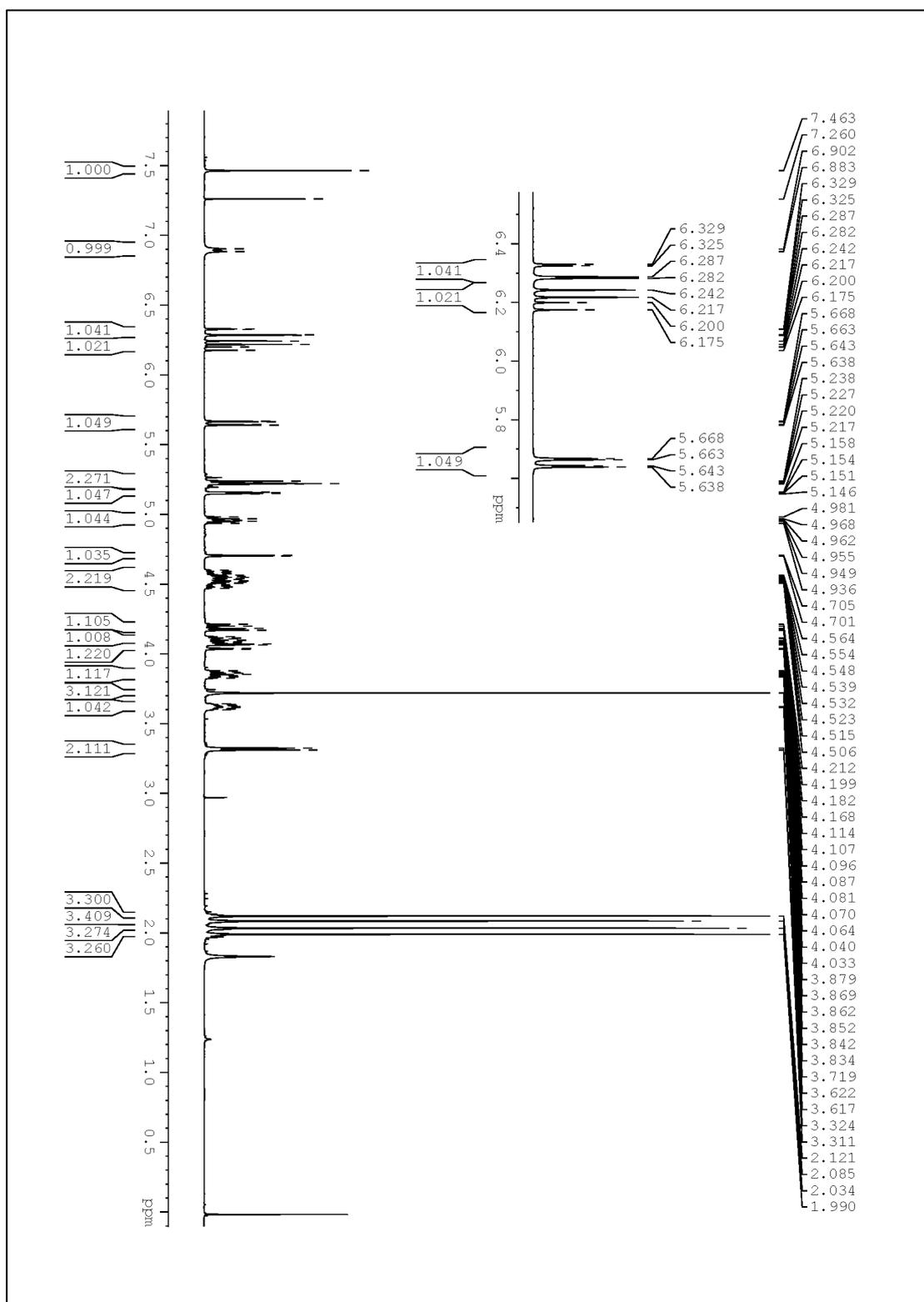


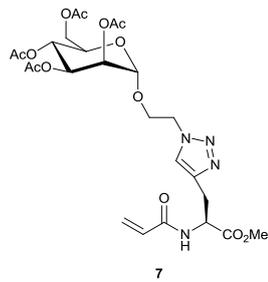
Alkyne **6** ^{13}C NMR (100 MHz, CDCl_3)



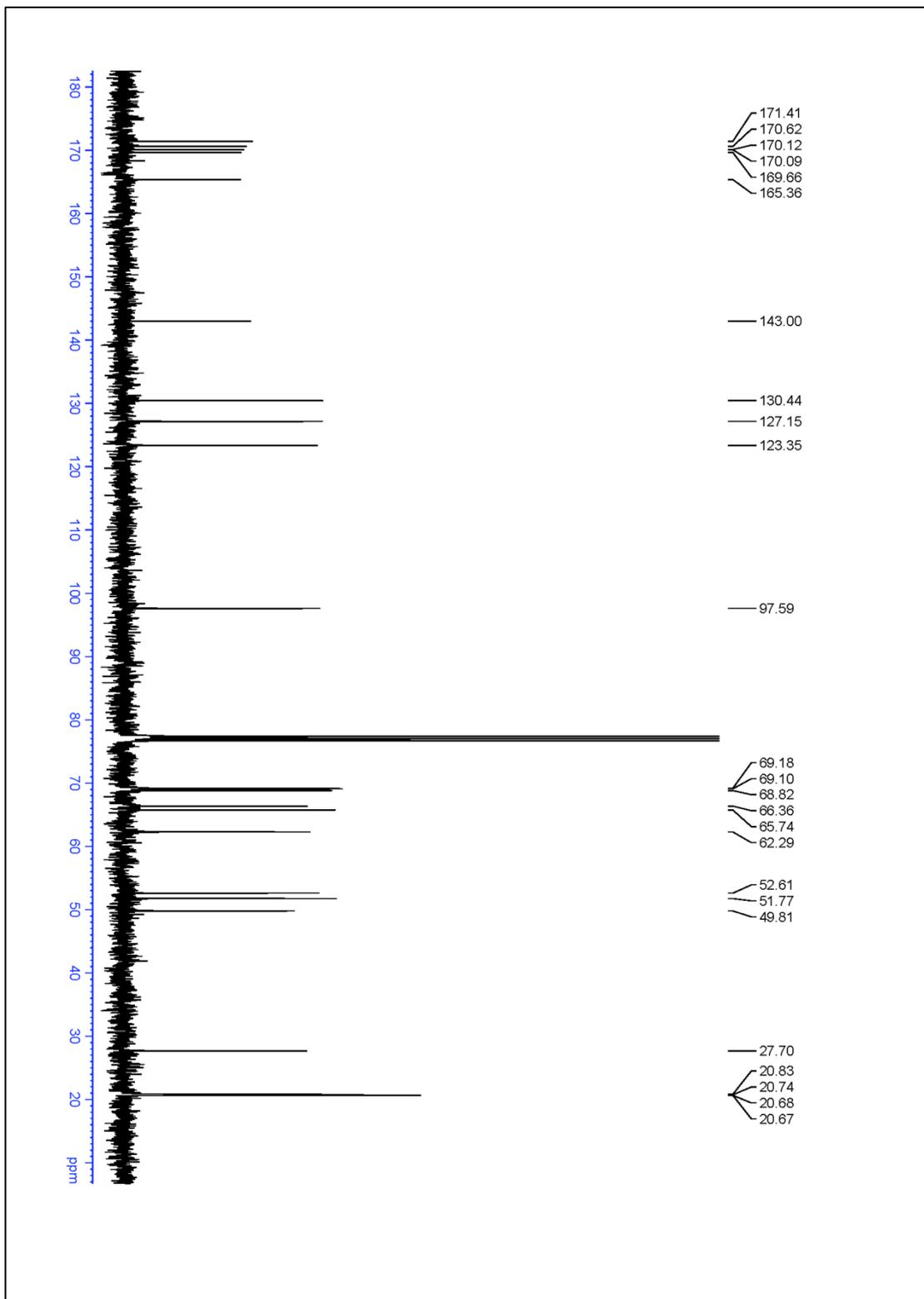


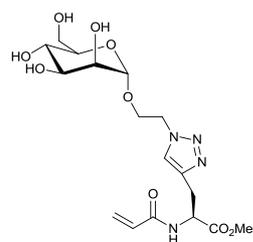
Acrylamide **7** ^1H NMR (400 MHz, CDCl_3)





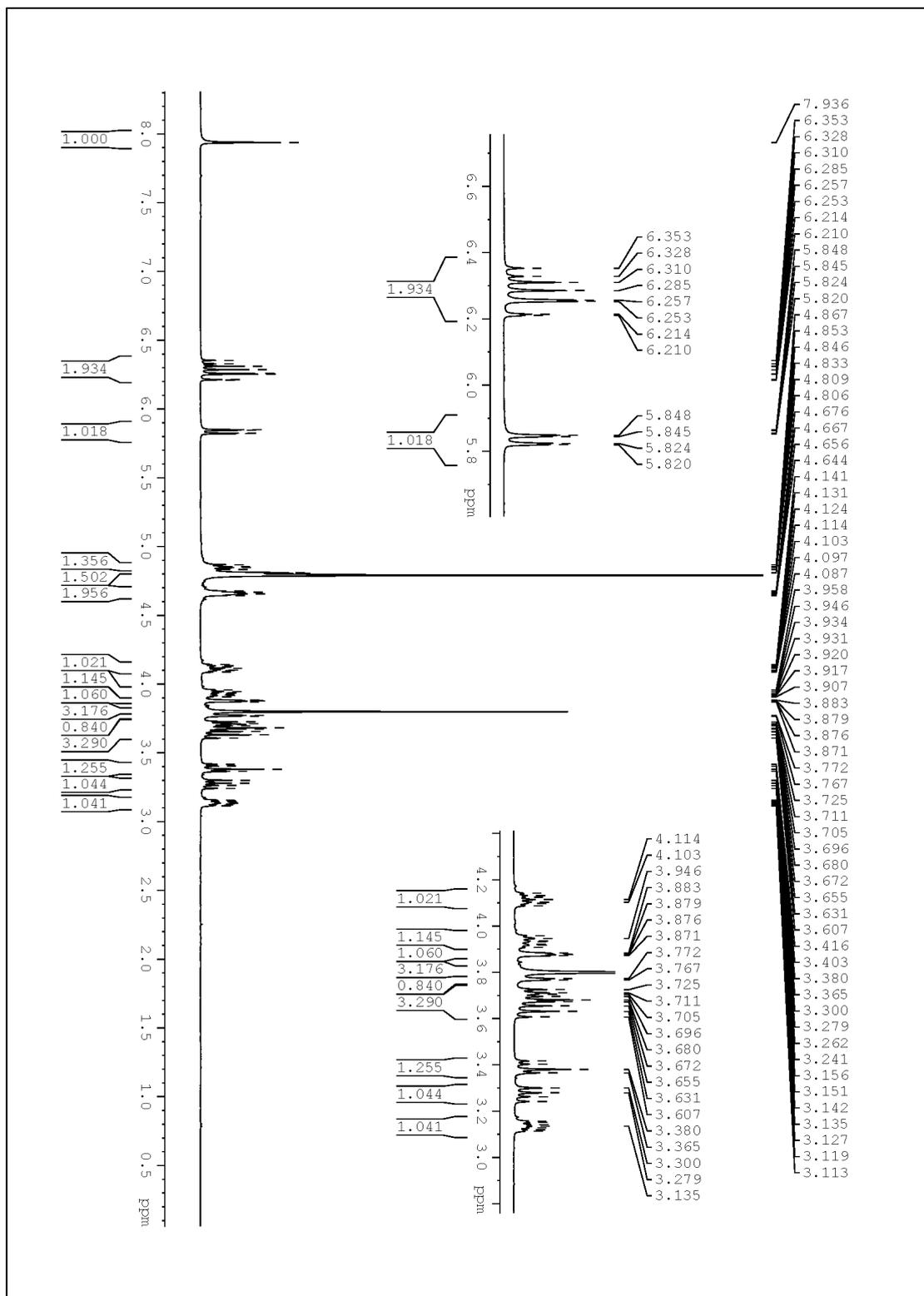
Acrylamide 7 ¹³C NMR (100 MHz, CDCl₃)



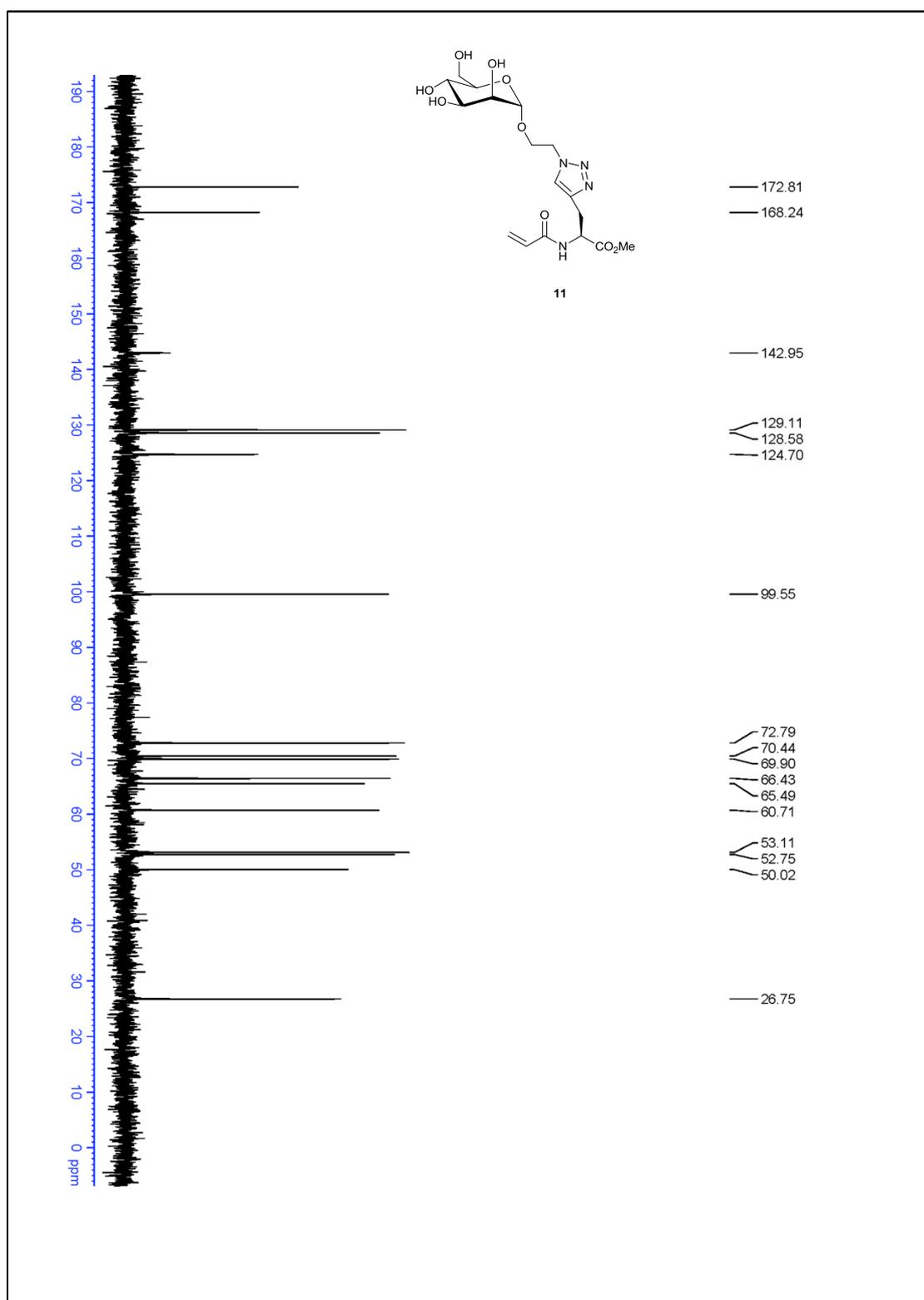


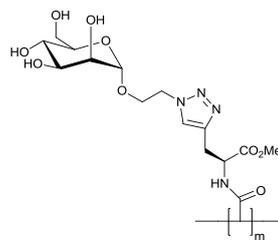
11

Acrylamide **11** ^1H NMR (400 MHz, D_2O)

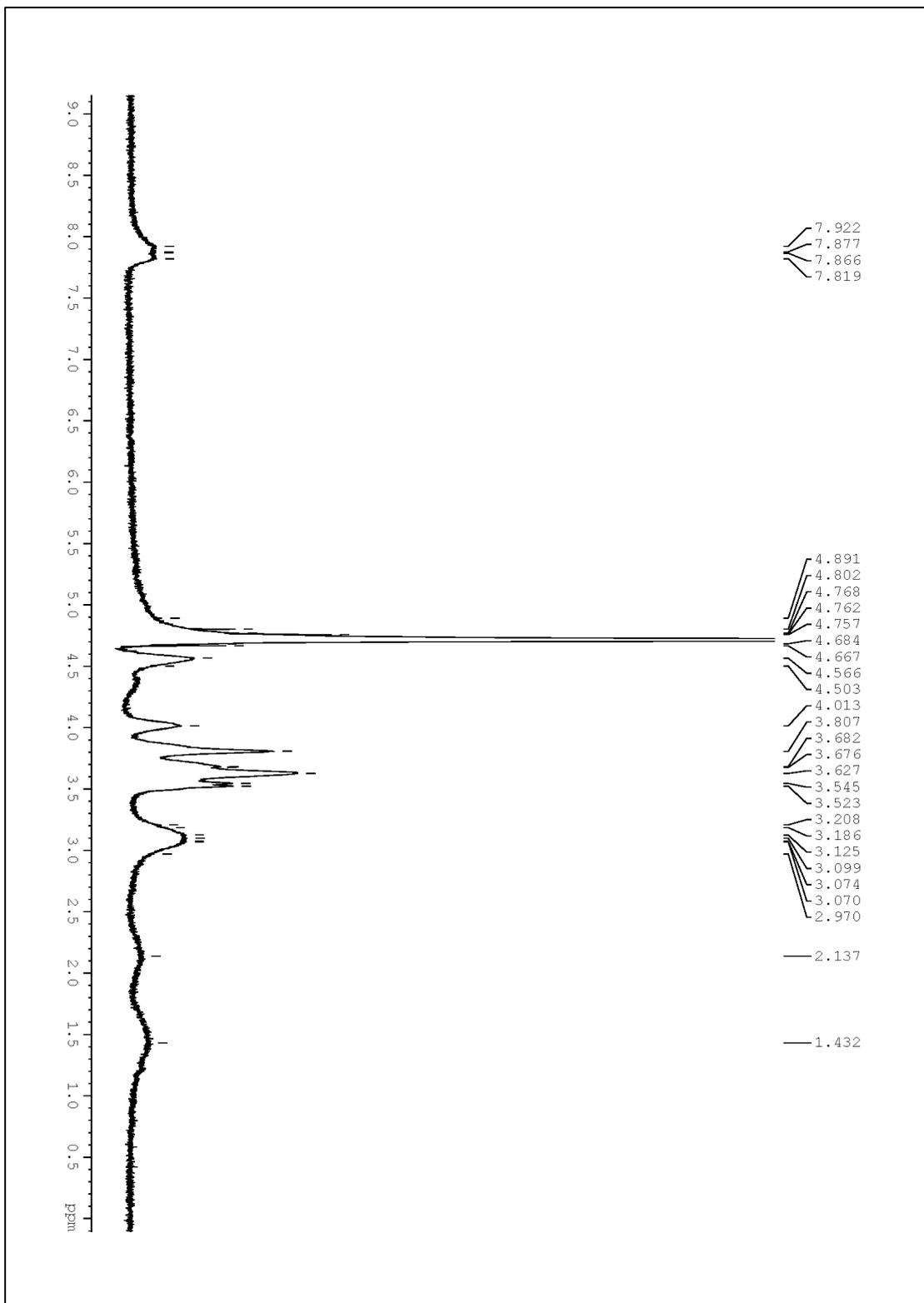


Acrylamide **11** ^{13}C NMR (400 MHz, D_2O)

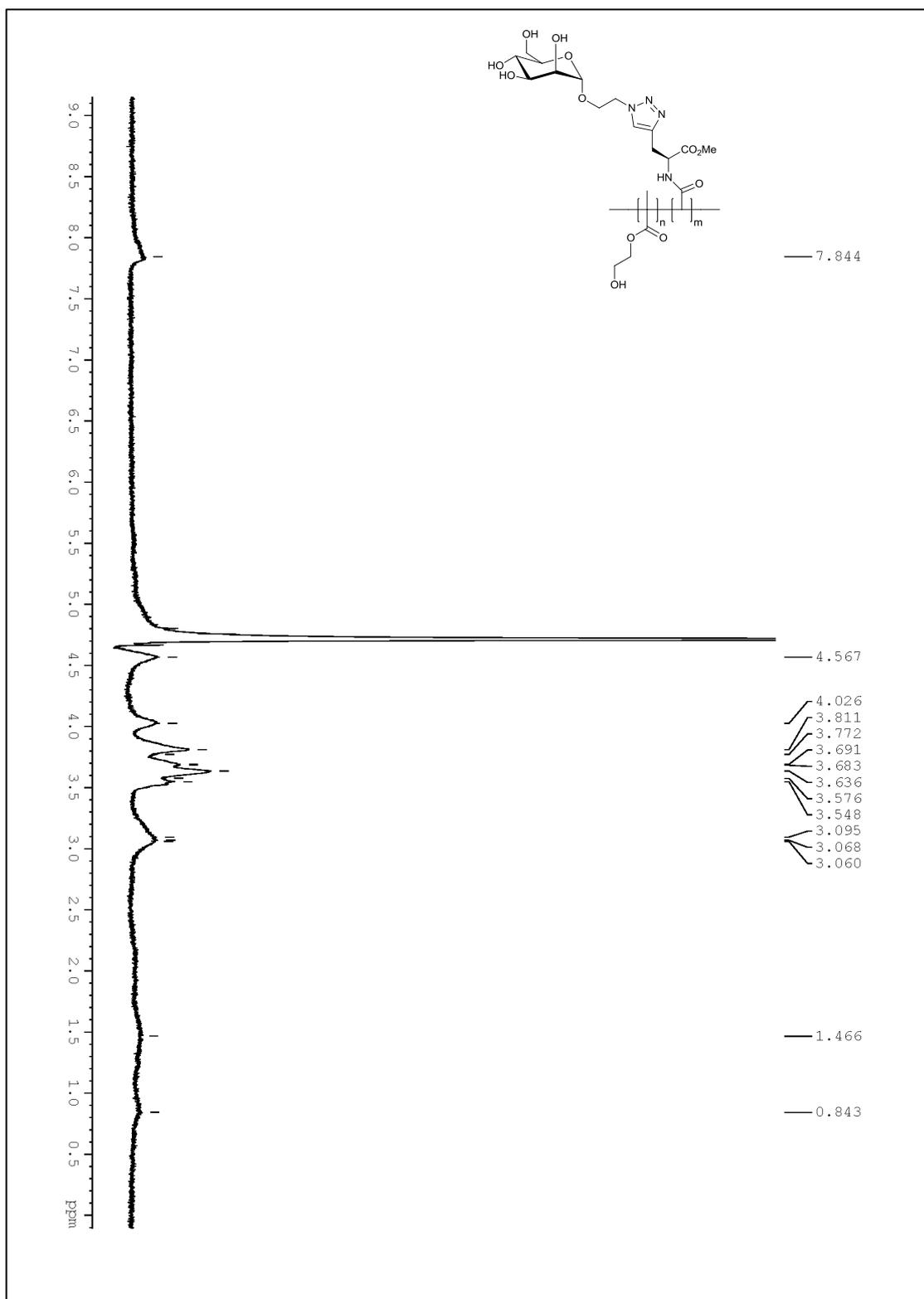




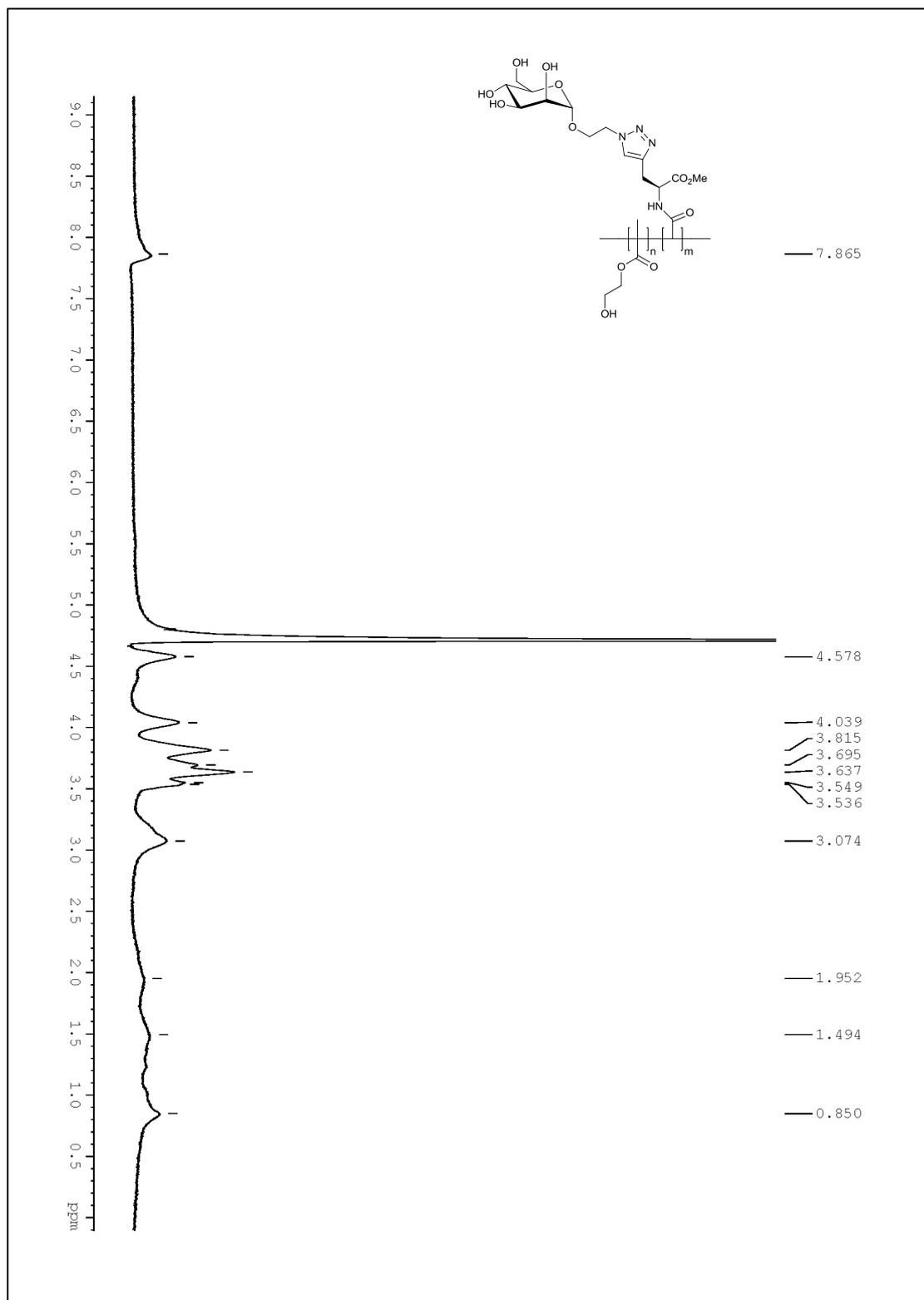
Polymer **8a** (100% **7**) ¹H NMR (400 MHz, D₂O)



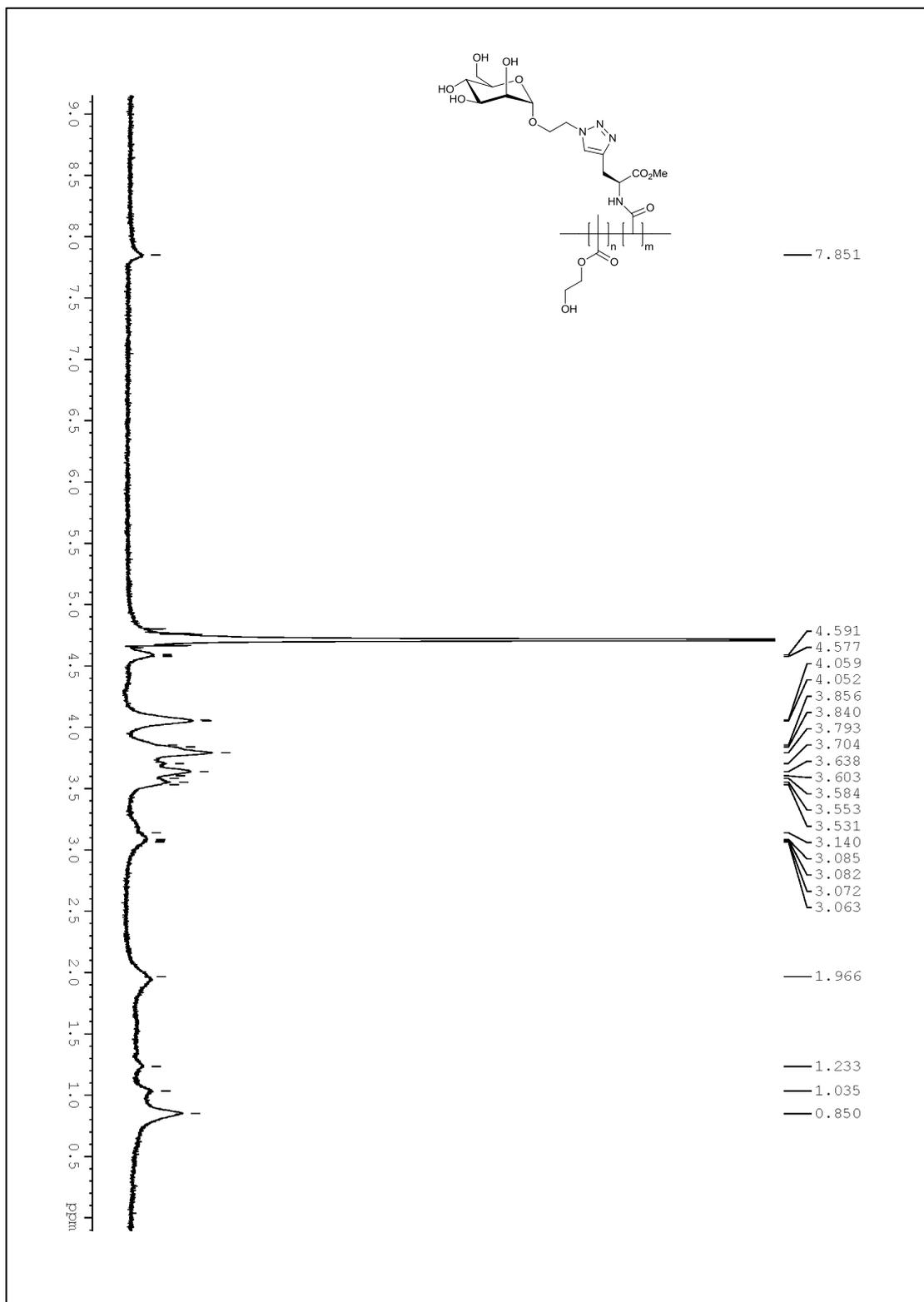
Copolymer **8b** (80% **7** 20 % HEMA) ^1H NMR (400 MHz, D_2O)



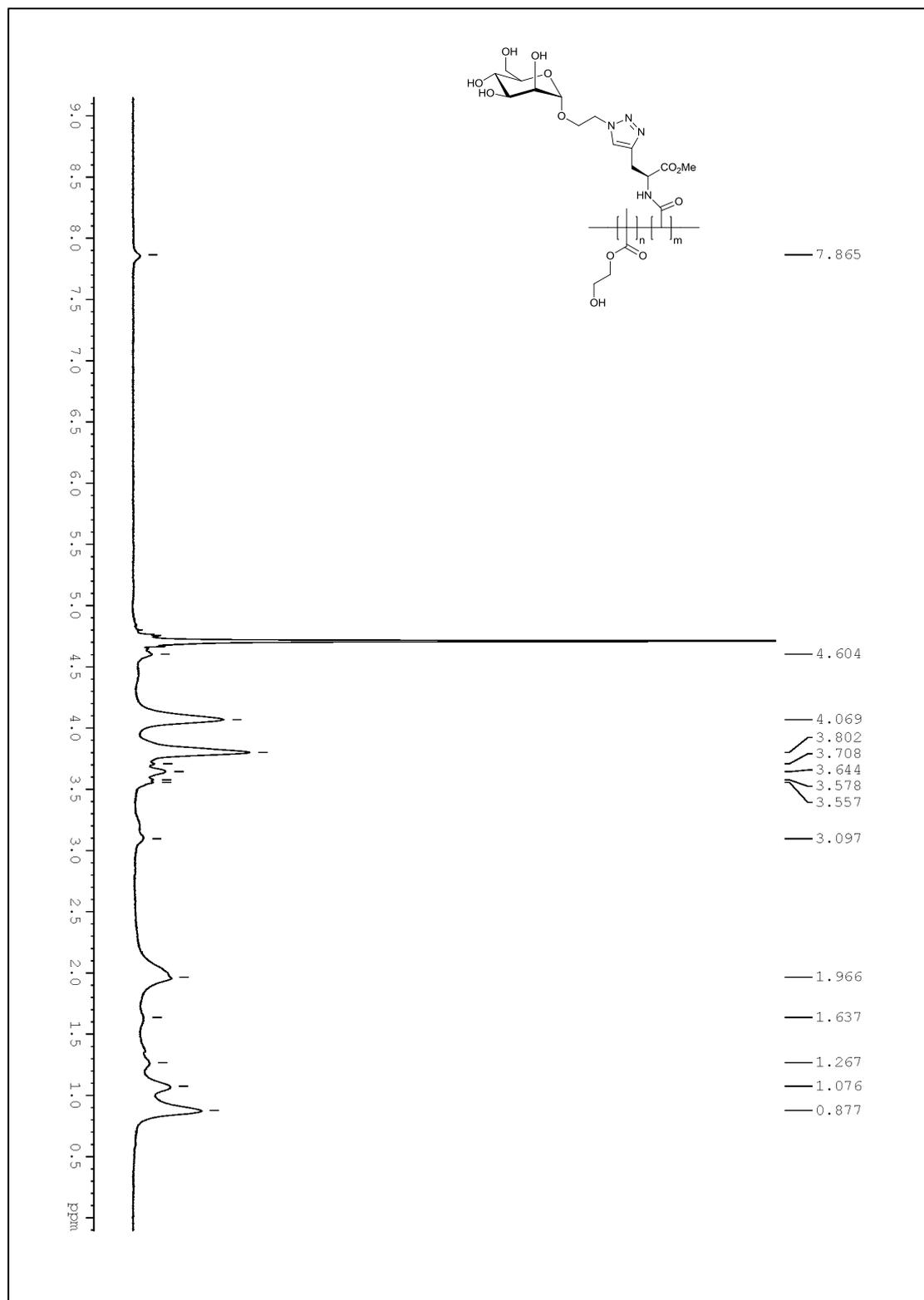
Copolymer **8c** (60% **7** 40% HEMA) ^1H NMR (400 MHz, D_2O)



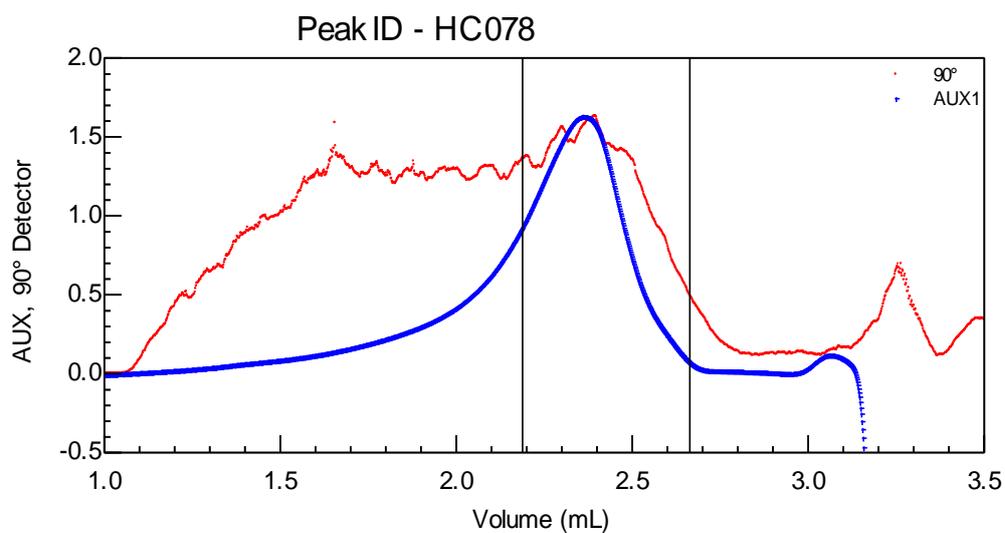
Copolymer **8d** (40% **7** 60% HEMA) ^1H NMR (400 MHz, D_2O)



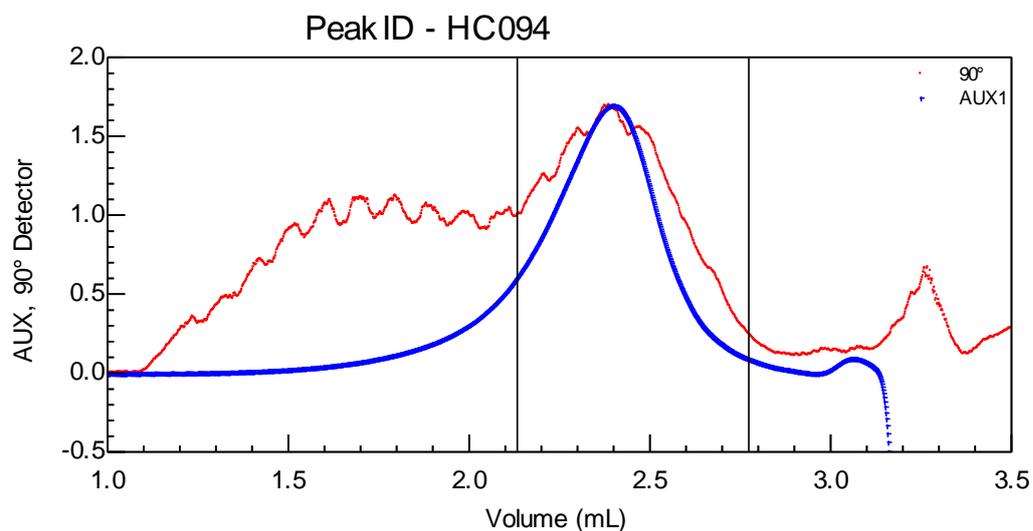
Copolymer **8e** (20% **7** 80% HEMA) ^1H NMR (400 MHz, D_2O)



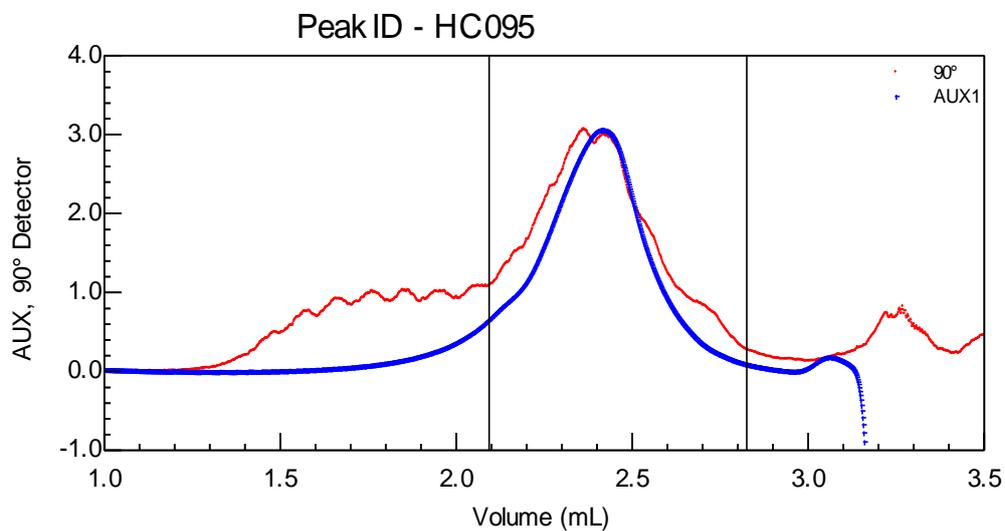
GPC traces of Copolymer **8a** (RI signal - blue line, 90° LS signal red line)



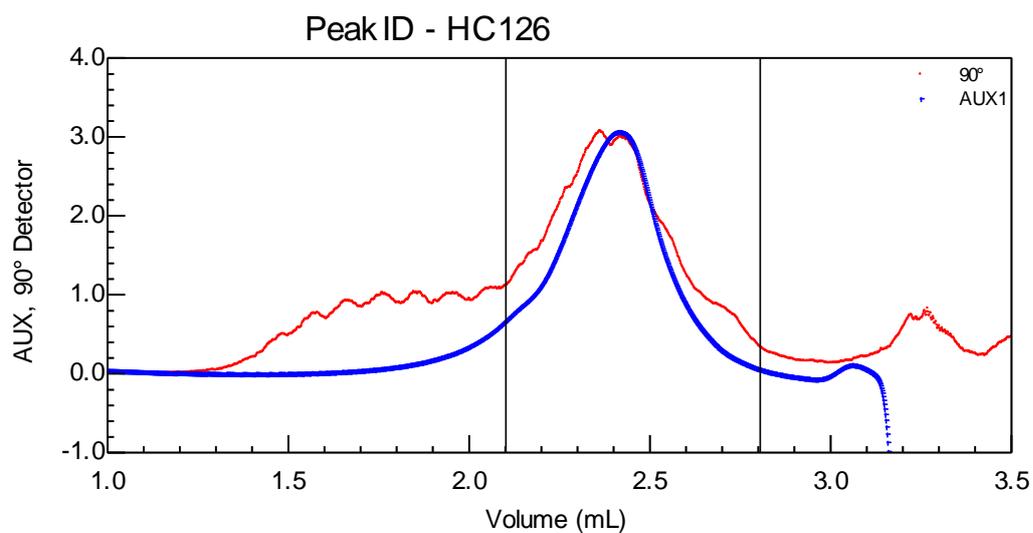
GPC traces of Copolymer **8b** (RI signal - blue line, 90° LS signal red line)



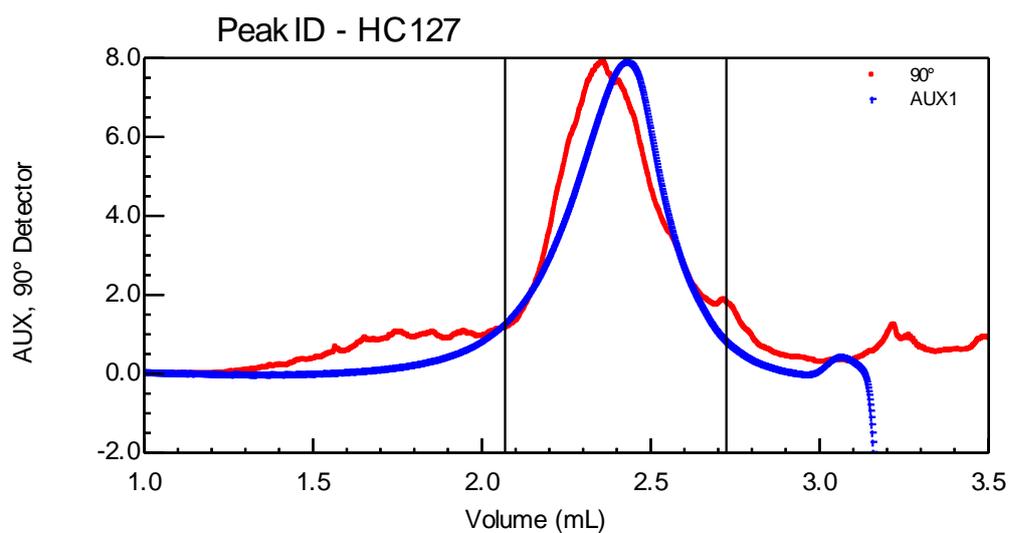
GPC traces of Copolymer **8c** (RI signal - blue line, 90° LS signal red line)



GPC traces of Copolymer **8d** (RI signal - blue line, 90° LS signal red line)



GPC traces of Copolymer **8e** (RI signal - blue line, 90° LS signal red line)



Con A quantitative precipitation assay

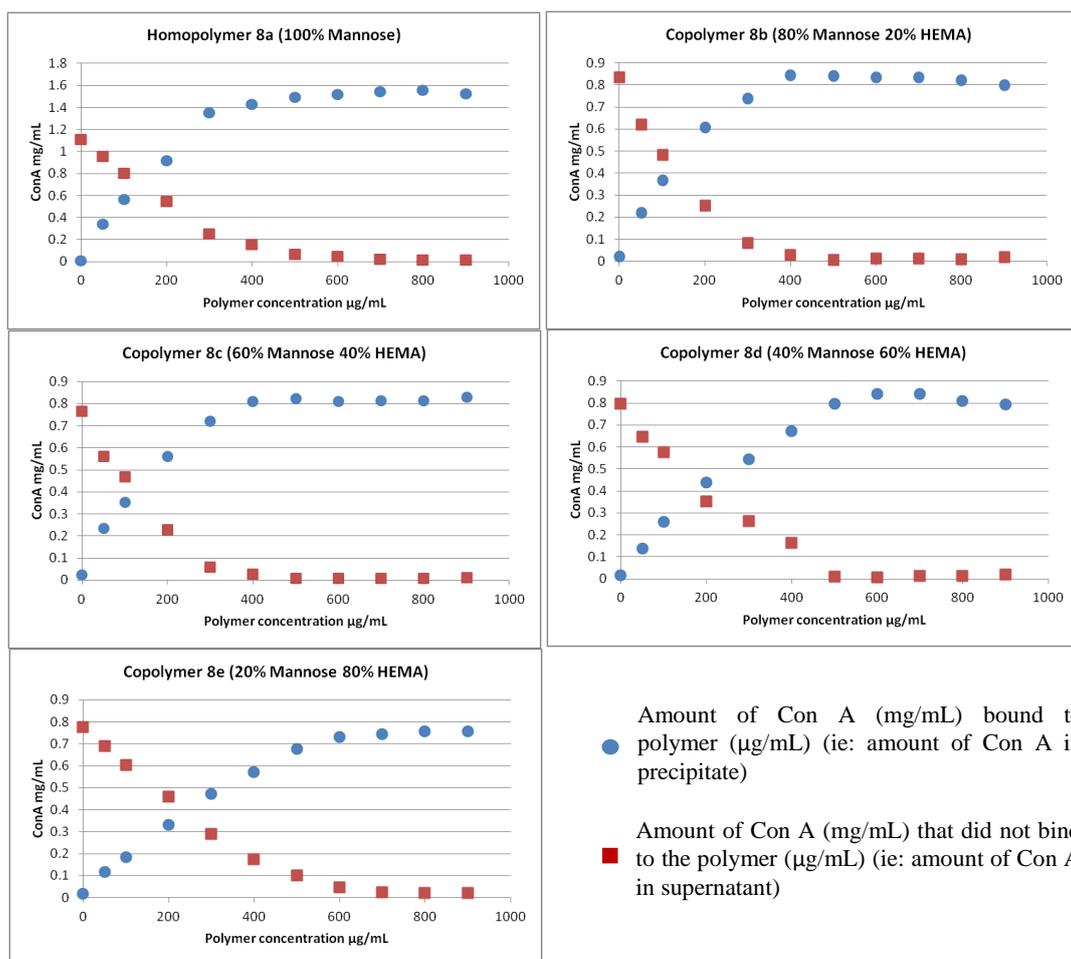


Figure 1S. Results from quantitative precipitation assay

References

- 1 J. Beignet, J. Tiernan, C. H. Woo, B. M. Kariuki, L. R. Cox, *J. Org. Chem.*, 2004, **69**, 6341-6356.
- 2 J. Geng, G. Manotovani, L. Tao, J. Nicolas, G. Chen, R. Wallis, D. A. Mitchell, B. R. G. Johnson, S. D. Evans, D. M. Haddleton, *J. Am. Chem. Soc.*, 2007, **129**, 15156-15163.
- 3 K. Yu, J. N. Kizhakkedathu, *Biomacromol.*, 2010, **11**, 3073-3085.