Supporting Information

Enzyme-like Catalysis by Single Chain Nanoparticles That Use Transition Metal Cofactors

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1. Experimental

1.1. General experimental procedures

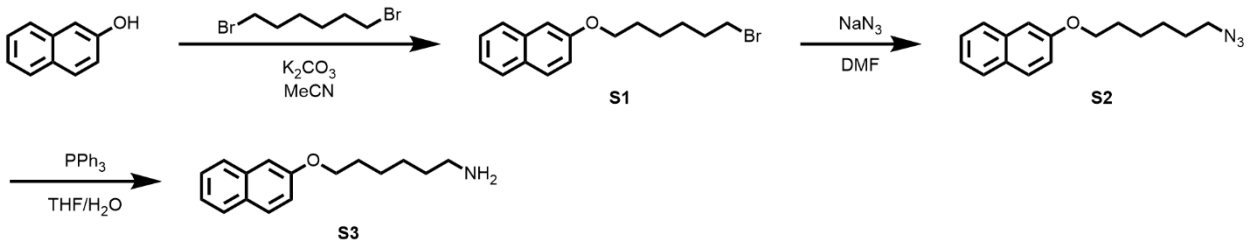
All reagents were purchased from Acros Organics, Cambridge Chemical Technologies, Chem-Impex International, Fisher Scientific, Oakwood Chemicals, Sigma-Aldrich, or TCI America, and used without further purification unless otherwise noted. For the synthetic procedures, 1,4-dioxane, DCM, DIPEA, and DMF were stored over activated 4 Å molecular sieves. NMR spectra were recorded using Varian UI400, U500, VXR500 and VNS750NB or Bruker CB500 and B600 spectrometers in the NMR Laboratory, School of Chemical Science, University of Illinois. NMR spectra were processed using MestReNova software and chemical shifts were in parts per million (ppm). All ^{1}H spectra were referenced to the residual solvent peak. Integration is provided and coupling constants (*J*) are reported in Hertz (Hz). Analytical gel permeation chromatography (GPC) experiments were performed on a Waters system equipped with a Waters 1515 isocratic pump, a Waters 2414 refractive index detector, and a Waters 2998 photodiode array detector. Separations were performed at 50 °C using DMF containing 0.1 M LiBr as the mobile phase. The molecular weights of all polymers were determined using dn/dc values for each sample calculated offline with the internal calibration system processed by the ASTRA 6 software (version 6.1.1, Wyatt Technology CA). Dynamic light scattering (DLS) characterization was performed using a Marvin Instrument Ltd. nanoZS Zetasizer with Zetasizer software. Fluorescence experiments were performed on a Horiba FluoroMax-4 fluorometer with FluorEssence (v3.5) software. Mass spectrometry was performed using a Waters Q-Tof Ultima mass spectrometer with MassLynx

software. The raw data files were processed using OriginPro. BTTAA (**L5**) was purchased from Sigma-Aldrich.

1.2. Synthetic procedures

General procedure for RAFT polymerization. In a 20 mL glass vial equipped with a magnetic stirbar, 46 mg of 2-(dodecylthiocarbonothioylthio)-2-methylpropionic acid and 2.07 mg of AIBN were dissolved in 2.5 mL of 1,4-dioxane. To this mixture was added 2.07 mL of pentafluorophenyl acrylate, and the vial was degassed using three cycles of freeze-pump-thaw. The vial was transferred to an oil bath preheated to 70 °C and stirred for 6.5 h. The solution was diluted with 3 mL of THF and precipitated from 10 mL of MeOH. This process was repeated three times, and the solid was dried under high vac at 40 °C for two days to obtain the polymer.

General procedure for post-polymerization functionalization. To a 20 mL glass vial was added 50-100 mg of poly(pentafluorophenyl acrylate)[1] and appropriate equivalents of hexylamine, 6-(naphthalen-2-yloxy)hexan-1-amine (**S3**), and mono(10-(14-azaneyl)-*N*,*N*,*N*-trimethyldecan-1-aminium)dichloride.[2] The vial was purged with N_2, and 5 mL of DMF and 100-200 eq of DIPEA were added. The vial was transferred to a hot plate preheated to 60 °C and stirred for 24 h. The progress of the reaction was monitored by ^{19}F NMR. The product was precipitated from Et_2O, redissolved in DMSO, and dialyzed (1 kD) against H_2O for two days. The aqueous solution was freeze-dried on a lyophilizer to obtain the functionalized polymer as an off-white solid.

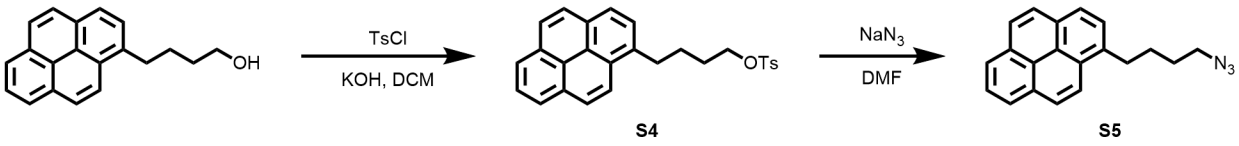


2-((6-Bromohexyl)oxy)naphthalene (S1). To a 3-neck 250 mL round-bottom flask was added 5.00 g 2-naphthol, 7.19 g K_2CO_3, and 50 mL of MeCN. The flask was transferred to an oil bath preheated to 90 °C and stirred for 30 min. To this flask was added a solution of 26.7 mL 1,6 dibromohexane dissolved in 50 mL MeCN, and the mixture was stirred at reflux under N_2 atm for 24 h. The mixture was partitioned between 100 mL H_2O and 100 mL $CHCl_3$. The organic layer was washed with 100 mL saturated aqueous solution of sodium chloride, dried over Na_2SO_4, and concentrated by rotary evaporation. The product was purified twice by silica gel column chromatography using gradient elution (100% hexane to 10% EtOAc/hexane) to obtain a white solid (8.08 g, 76% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.79 – 7.68 (m, 3H), 7.43 (m, 1H), 7.36 – 7.29 (m, 1H), 7.17 – 7.08 (m, 2H), 4.09 (t, J = 6.4, 2H), 3.44 (t, J = 6.8, 2H), 1.90 (m, 4H), 1.55 (p, J = 3.8, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 157.02, 134.60, 129.36, 128.91, 127.66, 126.70, 126.33, 123.53, 118.99, 106.55, 67.74, 33.86, 32.72, 29.10, 27.98, 25.40. High resolution ESI-MS: m/z calculated for $C_{16}H_{19}BrO^+$ ($[M+H]^+$): 307.0692; found 307.0689.

2-((6-Azidohexyl)oxy)naphthalene (S2). To a 250 mL round-bottom flask was added 5.00 g compound **S1**, 1.29 g sodium azide, and 75 mL DMF. The flask was transferred to an oil bath preheated to 60 °C and stirred under N_2 atm overnight. The mixture was diluted with 100 mL Et_2O and washed with H_2O (5 x 50 mL) and 50 mL saturated aqueous solution of sodium chloride. The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation to obtain a

yellow liquid (3.87 g, 88% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.82 – 7.67 (m, 3H), 7.43 (ddd, *J* = 8.2, 6.8, 1.3, 1H), 7.33 (ddd, *J* = 8.1, 6.9, 1.3, 1H), 7.15 (m, 2H), 4.09 (t, *J* = 6.4, 2H), 3.31 (t, *J* = 6.9, 6.3, 2H), 1.87 (p, *J* = 8.1, 6.5, 2H), 1.67 (p, *J* = 12.3, 7.6, 4.4, 2H), 1.62 – 1.41 (m, 4H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 157.02, 134.61, 129.37, 128.92, 127.66, 126.71, 126.34, 123.53, 118.99, 106.55, 67.73, 51.42, 29.15, 28.85, 26.57, 25.79. High resolution ESI-MS: *m*/*z* calculated for $C_{16}H_{20}N_3O^+$ ($[M+H]^+$): 270.1601; found 270.1563.

6-(Naphthalen-2-yloxy)hexan-1-amine (S3). To a 50 mL round-bottom flask was added 1.00 g compound **S2**, 1.17 g PPh_3, and 20 mL 4:1 THF/H_2O solution, and the mixture was stirred for 16 h. The mixture was concentrated by rotary evaporation and resuspended with 20 mL DCM and 20 mL 1 M aqueous HCl. The precipitate was collected by vacuum filtration and partitioned between 25 mL DCM and 25 mL H_2O. To the biphasic mixture was added KOH pellets until pH > 14. The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The solid was recrystallized from hot EtOAc and hexane to obtain a white solid (0.52 g, 58% yield). ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.79 – 7.68 (m, 3H), 7.42 (t, *J* = 7.5, 1H), 7.32 (t, *J* = 7.5, 1H), 7.17 – 7.10 (m, 2H), 4.08 (t, *J* = 6.5, 2H), 2.73 (t, *J* = 7.0, 2H), 1.86 (dt, *J* = 14.5, 6.6, 2H), 1.52 (tt, *J* = 14.0, 7.2, 5H), 1.47 – 1.38 (m, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 157.07, 134.62, 129.33, 128.89, 127.65, 126.70, 126.31, 123.49, 119.02, 106.54, 67.88, 42.12, 33.57, 29.25, 26.70, 26.03. High resolution ESI-MS: *m*/*z* calculated for $C_{16}H_{21}NO^+$ ($[M+H]^+$): 244.1696; found 244.1694.

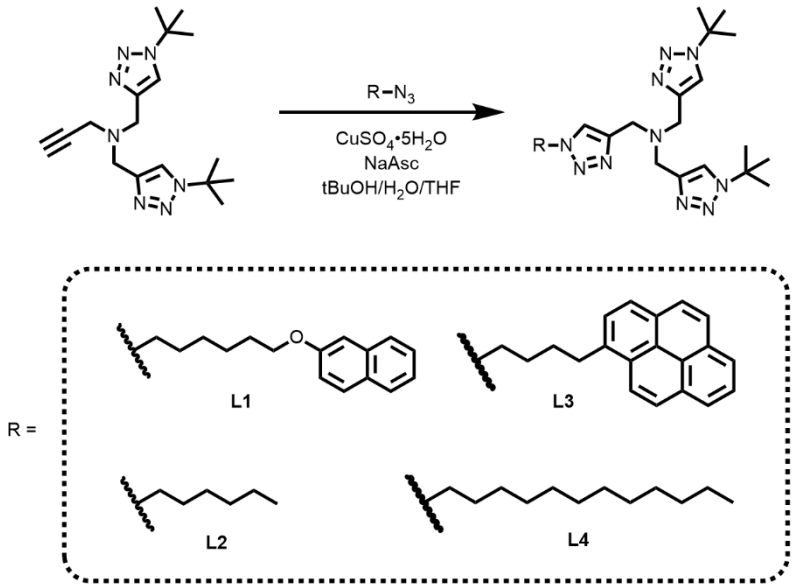


S4 **S5**

4-(1-Pyrenyl)-1-butanol tosylate (S4). To a 25 mL round-bottom flask was added 0.47 g 4-(pyren-1-yl) butan-1-ol in 10 mL DCM. To the flask was added 0.36 g 4-methylbenzenesulfonyl chloride, and the mixture was chilled in an ice bath. To the flask was added 0.38 g KOH over 10 min and the mixture was stirred for 3 h. The mixture was washed with 20 mL H_2O, and the aqueous layer was washed with DCM (2 x 20mL). The combined organic layers were washed with 20 mL saturated aqueous solution of sodium chloride, dried over Na_2SO_4, filtered, and concentrated by rotary evaporation to give the product (0.17 g, 23% yield). ^{1}H NMR (600 MHz, $CDCl_3$) δ 8.23 – 8.13 (m, 3H), 8.13 – 8.06 (m, 2H), 8.03 (s, 2H), 8.00 (t, *J* = 7.6, 1H), 7.79 (d, *J* = 7.7, 1H), 7.75 (d, *J* = 7.9, 2H), 7.25 (d, *J* = 7.8, 2H), 4.08 (t, *J* = 6.3, 2H), 3.31 (t, *J* = 7.6, 2H), 2.36 (s, 3H), 1.89 (p, *J* = 7.5, 2H), 1.80 (p, *J* = 6.4, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 144.68, 135.83, 133.09, 131.43, 130.87, 129.93, 129.80, 128.59, 127.86, 127.49, 127.38, 127.20, 126.73, 125.90, 125.09, 124.99, 124.98, 124.79, 123.19, 70.38, 60.42, 32.71, 28.76, 27.48, 21.57, 14.22. High resolution ESI-MS: *m*/*z* calculated for $C_{27}H_{25}O_3S^+$ ($[M+H]^+$): 429.1519; found 429.1503.

1-(4-Azido-butyl)-pyrene (S5). To a 20 mL glass vial was added 150 mg compound **S4**, 50 mg sodium azide, and 2.5 mL DMF. The vial was transferred to a well plate preheated to 60 °C and stirred under N_2 atm overnight. The mixture was diluted with 12 mL Et_2O and washed with H_2O (5 x 6 mL) and 6 mL saturated aqueous solution of sodium chloride. The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The product was purified by silica gel column chromatography using gradient elution (10% EtOAc/hexane to 20% EtOAc/hexane) to obtain a white solid (40 mg, 25% yield). ^{1}H NMR (600 MHz, $CDCl_3$) δ 8.26 (d, *J* = 9.2, 1H), 8.19 – 8.15 (m, 2H), 8.12 (d, *J* = 8.5, 2H), 8.03 (d, *J* = 2.4, 2H), 8.00 (t, *J* = 7.6, 1H), 7.87 (d, *J* = 7.7, 1H), 3.39 (t, *J* = 7.8, 2H), 3.34 (t, *J* = 6.9, 2H), 1.96 (p, *J* = 7.7, 2H), 1.78 (dt, *J* = 14.5, 7.0,

2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 136.10, 131.45, 130.90, 129.94, 128.62, 127.52, 127.39, 127.23, 126.71, 125.88, 125.13, 125.03, 124.96, 124.84, 124.78, 123.23, 51.41, 33.01, 28.91, 28.85. High resolution ESI-MS: *m*/*z* calculated for $C_{20}H_{18}N_3^+$ ([M+H]$^+$): 300.1501; found: 300.1455.



General procedure for synthesis of bis(*tert*-butyltriazolyl) ligands L1-L4. To a 20 mL glass vial was added 1 eq *N,N*-bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine[3] and 1.15 eq substituted azide dissolved in 1:1:1 *tert*-butanol/H_2O/THF. To this vial was added 0.05 eq $CuSO_4$**.**$5H_2O$ and 0.2 eq NaAsc, and the mixture was stirred overnight at room temperature under N_2 atm. The mixture was concentrated by rotary evaporation, dissolved in 20 mL of EtOAc, and washed with 20 mL of H_2O and 10 mL of saturated aqueous solution of sodium chloride. The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The product was purified by silica gel column chromatography (10% MeOH/DCM).

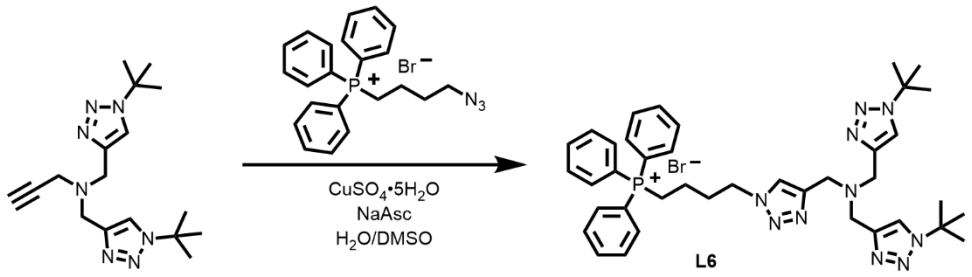
L1. *N,N*-Bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine and compound **S2** were reacted according to the general procedure to obtain an off-white solid (0.32 g, 43% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.86 (m, 3H), 7.78 – 7.68 (m, 3H), 7.42 (t, *J* = 7.7, 1H), 7.32 (t, *J* = 7.4, 1H), 7.16 – 7.09 (m, 2H), 4.39 (t, *J* = 7.0, 2H), 4.06 (t, *J* = 6.4, 2H), 3.77 (m, 6H), 2.00 (p, *J* = 7.8, 2H), 1.85 (p, *J* = 7.5, 2H), 1.69 (s, 18H), 1.45 (p, *J* = 7.4, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 156.98, 134.59, 129.35, 128.90, 127.63, 126.71, 126.31, 123.51, 121.09, 118.97, 106.54, 67.62, 59.26, 50.24, 46.95, 30.31, 30.06, 29.04, 26.36, 25.66. High resolution ESI-MS: *m*/*z* calculated for $C_{33}H_{47}N_{10}O^+$ ($[M+H]^+$): 599.3929; found 599.3947.

L2. *N,N*-Bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine and 1-azidohexane were reacted according to the general procedure to obtain a white solid (0.26 g, 78% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.85 (s, 3H), 4.35 (t, *J* = 7.3, 2H), 3.78 (s, 2H), 3.76 (s, 4H), 1.91 (t, *J* = 7.5, 2H), 1.69 (s, 18H), 1.31 (broad s, 6H), 0.93 – 0.82 (m, 3H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 121.08, 59.26, 50.37, 47.05, 46.94, 31.17, 30.31, 30.06, 26.21, 22.44, 13.96. High resolution ESI-MS: *m*/*z* calculated for $C_{23}H_{41}N_{10}^+$ ($[M+H]^+$): 457.3510; found 457.3510.

L3. *N,N*-Bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine and compound **S5** were reacted according to the general procedure to obtain a viscous oil (24 mg, 29% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.22 (d, *J* = 9.0, 1H), 8.16 (d, *J* = 7.7, 2H), 8.10 (d, *J* = 8.1, 2H), 8.02 (s, 2H), 7.98 (t, *J* = 7.6, 1H), 7.85 (s, 1H), 7.83 (s, 1H), 7.80 (s, 2H), 4.41 (t, *J* = 7.0, 2H), 3.75 (s, 2H), 3.73 (s, 4H), 3.39 (t, *J* = 7.6, 2H), 2.13 – 2.08 (m, 2H), 1.91 (t, *J* = 8.0, 2H), 1.67 (s, 18H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 135.65, 131.39, 130.84, 129.96, 128.56, 127.48, 127.46, 127.24, 126.71, 125.87, 125.07, 124.95, 124.85, 124.79, 123.12, 50.28, 46.66, 42.67, 32.74, 30.15, 29.99,

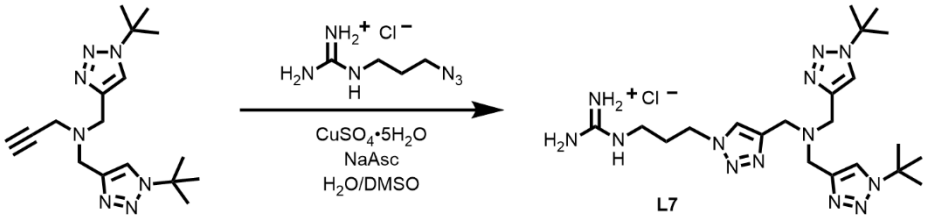
28.51. High resolution ESI-MS: *m*/*z* calculated for $C_{37}H_{45}N_{10}^+$ ([M+H]$^+$): 629.3823; found 629.3829.

L4. *N,N*-Bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine and 1-azidododecane were reacted according to the general procedure to obtain a white solid (90 mg, 57% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.84 (s, 3H), 4.34 (t, *J* = 7.4, 2H), 3.78 (s, 2H), 3.76 (s, 4H), 1.91 (broad s, 2H), 1.69 (s, 18H), 1.28 (m, 18H), 0.87 (t, *J* = 6.9, 3H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 121.08, 59.21, 50.37, 46.93, 31.91, 30.36, 30.06, 29.61, 29.53, 29.41, 29.34, 29.03, 26.55, 22.70, 14.14. High resolution ESI-MS: *m*/*z* calculated for $C_{29}H_{53}N_{10}^+$ ([M+H]$^+$): 541.4449; found 541.4453.



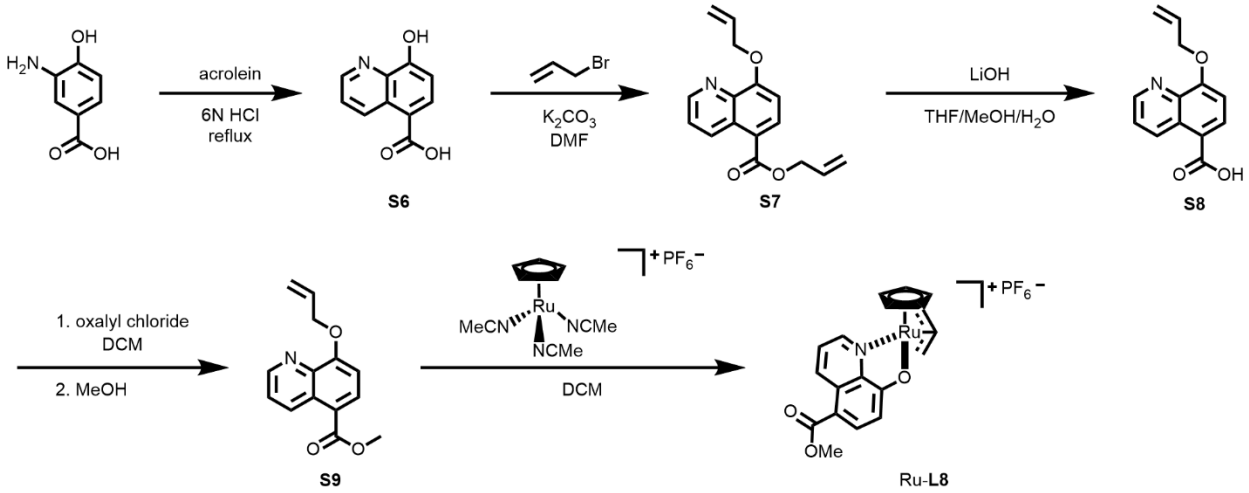
L6. To a 20 mL glass vial was added 0.40 g (4-bromobutyl)triphenylphosphonium azide,[4] 0.33 g *N,N*-bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine,[2] 25 mg $CuSO_4$•$5H_2O$, and 12 mL of 1:1 H_2O/DMSO. To the mixture was added 40 mg NaAsc under N_2 atm. The mixture was stirred at 60 °C for 20 h. To the mixture was added 50 mL of 1 M aqueous EDTA and the mixture was extracted with 50 mL of DCM. The organic layer was washed with 50 mL of H_2O and 50 mL of saturated aqueous solution of sodium chloride, dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The crude product was purified by silica gel column

chromatography using gradient elution (100% DCM to 10% MeOH/DCM) to obtain a yellow solid (0.56 g, 71% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.13 (s, 1H), 7.92 (s, 2H), 7.80 – 7.71 (m, 9H), 7.63 (td, *J* = 7.8, 3.4, 6H), 4.61 (t, *J* = 6.4, 2H), 3.86 (td, *J* = 13.0, 6.6, 2H), 3.72 (s, 2H), 3.67 (s, 4H), 2.38 (p, *J* = 6.7, 2H), 1.66 (s, 18H), 1.63 – 1.53 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 144.67, 143.70, 135.44, 135.41, 133.96, 133.88, 130.83, 130.73, 124.56, 121.37, 118.29, 117.61, 59.52, 48.98, 48.19, 47.56, 30.35, 22.59, 22.18, 19.45. High resolution ESI-MS: *m*/*z* calculated for $C_{39}H_{50}N_{10}P^{+}$: 689.3952; found 689.3951.



L7. To a 20 mL glass vial was added 0.18 g *N*-(3-azidopropyl)-guanidinium chloride,[5] 0.33 g *N,N*-bis((1-(*tert*-butyl)-1*H*-1,2,3-triazol-4-yl)methyl)prop-2-yn-1-amine,[2] 25 mg $CuSO_4$•$5H_2O$, and 12 mL of 1:1 H_2O/DMSO. To the mixture was added 40 mg NaAsc under N_2 atm. The mixture was stirred at 60 °C for 24 h. To the mixture was added 50 mL of 1 M aqueous EDTA and the mixture was extracted with a solution of 2:1 DCM/IPA (3 x 50 mL). The combined organic layers were washed with 50 mL of brine, dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The crude product was purified by silica gel column chromatography using gradient elution (100% DCM to 1% NH_4OH, 15% MeOH in DCM) to obtain a yellow solid (0.11 g, 20% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.40 (s, 1H), 8.16 (s, 1H), 7.93 (s, 2H), 7.78 (broad s, 2H), 4.57 (t, *J* = 7.0, 2H), 3.72 (s, 6H), 3.47 – 3.32 (m, 2H), 3.10 (broad s, 2H), 2.31 – 2.18 (m, 2H), 1.64 (s, 18H). ^{13}C

NMR (126 MHz, $CDCl_3$) δ 158.03, 143.78, 142.98, 125.29, 121.90, 59.90, 47.55, 47.10, 38.81, 30.27, 29.89. High resolution ESI-MS: *m/z* calculated for $C_{21}H_{38}N_{13}^+$: 472.3368; found 472.3370.



8-Hydroxyquinoline-5-carboxylic acid (S6). To a 250 mL round-bottom flask was added 5.5 g 3-amino-4-hydroxybenxoic acid and 100 mL 6 M HCl. The suspension was magnetically stirred at 40 °C in an oil bath. To the suspension was added 3.3 mL acrolein dropwise via an addition funnel over 30 min. The mixture was refluxed at 105 °C for 2 h. The dark brown mixture was vacuum filtered. The pH of the dark brown filtrate was adjusted to pH 9 with 28% w/w aqueous NH_4OH. The basic solution was vacuum filtered. The pH of the dark brown filtrate was adjusted to pH 7 using 10 M HCl and vacuum filtered. The pH of the filtrate was adjusted to pH 6 with 10 M HCl and vacuum filtered. The addition of 10 M HCl was repeated dropwise until solid crashed out and consequently filtered. This process was repeated until an orange solid was obtained (usually pH 4-5). The pH 5 filtrate was extracted with EtOAc and more yellow solid precipitated out. The combined orange solids were washed with DCM twice and dried under vacuum to obtain a bright orange powdery solid (1.02 g, 17% yield). ^{1}H NMR (500 MHz, DMSO-d_6): δ 9.47 (dd, *J*

= 8.8, 1.6, 1H), δ 8.90 (dd, *J* = 4.0, 1.6, 1H), δ 8.24 (d, *J* = 8.2, 1H), δ 7.69 (dd, *J* = 8.8, 4.1, 1H), δ 7.12 (d, *J* = 8.2, 1H). ^{13}C NMR (125 MHz, DMSO-d_6): δ 167.63, 157.75, 148.11, 138.21, 134.51, 133.39, 128.05, 123.21, 116.68, 110.11. High resolution ESI-MS: *m*/*z* calculated for $C_{10}H_8NO_3^+$ ($[M+H]^+$): 190.0499; found 190.0495.

Allyl 8-(allyloxy)quinoline-5-carboxylate (S7). To a 50 mL round-bottom flask was added 1.0 g compound **S6**, 4.4 g potassium carbonate, and 15 mL DMF. The mixture was stirred, and 1.8 mL allyl bromide was added dropwise. The mixture was stirred at 50 °C for 16 h. To the mixture was added 20 mL H_2O and 20 mL EtOAc. The organic layer was collected and the aqueous layer was extracted with EtOAc (2 x 20 mL). The combined organic layers were washed with H_2O (4 x 20 mL) and 20 mL saturated aqueous solution of sodium chloride, dried over Na_2SO_4, and concentrated by rotary evaporation. The product was purified by silica gel column chromatography (30% EtOAc/hexane) to afford a pale yellow solid. The solid was washed with a minimal amount of Et_2O to remove the yellow impurity and obtain a white solid (0.55 g, 39% yield). ^{1}H NMR (500 MHz, $CDCl_3$): δ 9.47 (dd, *J* = 8.8, 1.7, 1H), 8.98 (dd, *J* = 4.1, 1.7, 1H), 8.35 (d, *J* = 8.4, 1H), 7.55 (dd, *J* = 8.8, 4.1, 1H), 7.06 (d, *J* = 8.4, 1H), 6.20 (ddt, *J* = 17.3, 10.7, 5.5, 1H), 6.09 (ddt, *J* = 17.3, 10.5, 5.7, 1H), 5.47 (m, 2H), 5.35 (m, 2H), 4.94 (dt, *J* = 5.6, 1.5, 2H), 4.88 (dt, *J* = 5.6, 1.5, 2H). ^{13}C NMR (126 MHz, $CDCl_3$): δ 165.91, 158.30, 149.43, 140.17, 134.53, 132.57, 132.33, 128.78, 123.05, 118.97, 118.38, 118.05, 107.51, 77.22, 70.07, 65.49. High resolution ESI-MS: *m*/*z* calculated for $C_{16}H_{16}NO_3^+$ ($[M+H]^+$): 270.1125; found 270.1118.

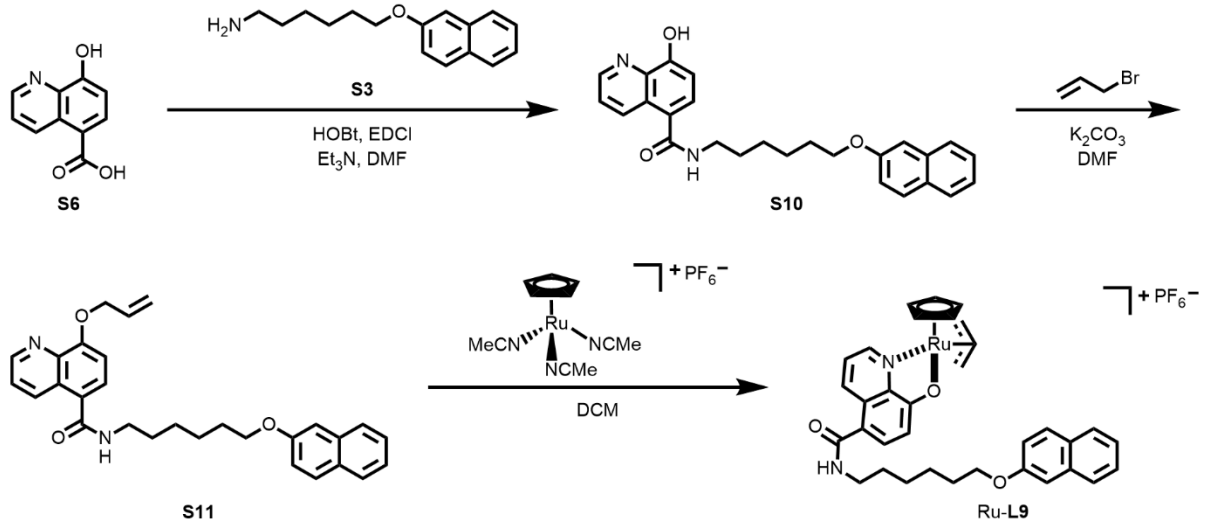
8-(Allyloxy)-7,8-dihydroquinoline-5-carboxylic acid (S8). To a 20 mL glass vial was added 460 mg compound **S7**, 717 mg $LiOH·H_2O$, 2 mL THF, 2 mL methanol, and 1 mL H_2O. The mixture

was stirred at room temperature for 12 h. Volatiles were removed by rotary evaporation and the solid was resuspended in 5 mL H_2O. The pH of the slurry was adjusted to pH 3 with 1 M aqueous HCl and filtered to afford a white solid (374 mg, 95% yield). ^{1}H NMR (500 MHz, DMSO-d_6): δ 12.95 (s, 1H), 9.41 (dd, J = 8.8, 1.7, 1H), 8.91 (dd, J = 4.0, 1.7, 1H), 8.27 (d, J = 8.4, 1H), 7.67 (dd, J = 8.7, 4.0, 1H), 7.27 (d, J = 8.4, 1H), 6.18 (ddt, J = 17.1, 10.7, 5.4, 1H), 5.53 (dq, J = 17.2, 1.6, 1H), 5.35 (dq, J = 10.5, 1.5, 1H), 4.85 (dt, J = 5.4, 1.5, 2H). ^{13}C NMR (126 MHz, DMSO-d_6): δ 167.56, 157.68, 149.02, 139.55, 133.89, 133.10, 132.54, 127.98, 123.06, 118.30, 118.28, 108.13, 69.17. High resolution ESI-MS: m/z calculated for $C_{13}H_{12}NO_3^+$ ($[M+H]^+$): 230.0812; found 230.0808.

Methyl 8-(allyloxy)quinoline-5-carboxylate (S9). To a 25 mL round-bottom flask was added 300 mg compound **S8** and 7.5 mL dry DCM, and 11.5 µL DMF and 285 µL oxalyl chloride were added. The mixture was stirred at room temperature for 1 h. Volatiles were removed by rotary evaporation and the resulting solid was washed with dry THF. The solid was dissolved in 10 mL methanol and stirred at room temperature for 16 h. Volatiles were removed by rotary evaporation and the resulting solid was dissolved in 10 mL EtOAc and washed with 10 mL sat. aqueous $NaHCO_3$. The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The product was purified by silica gel column chromatography (50% EtOAc/hexane) to obtain a pale yellow oil. The oil was washed with hexane to afford a white crystalline solid (164 mg, 52% yield). ^{1}H NMR (500 MHz, acetone-d_6): δ 9.43 (dd, J = 8.8, 1.7, 1H), 8.92 (dd, J = 4.0, 1.7, 1H), 8.32 (d, J = 8.4, 1H), 7.65 (dd, J = 8.8, 4.0, 1H), 7.26 (d, J = 8.4, 1H), 6.23 (ddt, J = 17.3, 10.5, 5.2, 1H), 5.59 (dq, J = 17.2, 1.7, 1H), 5.34 (dq, J = 10.6, 1.5, 1H), 4.90 (dt, J = 5.1, 1.6, 2H), 3.94 (s, 3H). ^{13}C NMR (126 MHz, acetone-d_6): δ 166.28, 158.66, 149.00, 140.33, 133.71, 133.12, 132.47,

128.40, 122.95, 117.91, 117.28, 107.83, 69.41, 51.30. High resolution ESI-MS: *m*/*z* calculated for $C_{14}H_{14}NO_3^+$ ([M+H]$^+$): 244.0968; found 244.0962.

Ru-**L8.** To a 4 mL glass vial was added 27.6 mg compound **S9** and 0.5 mL MeCN. To a different 4 mL glass vial was added 49.3 mg tris(acetonitrile)cyclopentadienylruthenium(II) and 0.5 mL MeCN. The solution of compound **S9** was added to the ruthenium solution. The mixture was stirred at room temperature for 1 h. The mixture was added dropwise to 45 mL Et_2O in a 50 mL centrifuge tube, and a yellow solid was collected by centrifugation. The pellet was washed with Et_2O (2 x 35 mL). The product was collected by centrifugation and dried under vacuum to afford a yellow solid (50 mg, 79% yield). ^{1}H NMR (500 MHz, CD_3CN): δ 9.62 (dd, *J* = 8.8, 1.2, 1H), 8.68 (dd, *J* = 5.1, 1.2, 1H), 8.21 (d, *J* = 8.6, 1H), 7.65 (dd, *J* = 8.8, 5.1, 1H), 6.91 (d, *J* = 8.6, 1H), 5.98 (s, 5H), 4.54 (tt, *J* = 10.7, 6.2, 1H), 4.44 (d, *J* = 11.0, 1H), 4.18 (d, *J* = 10.8, 1H), 4.17 – 4.11 (m, 2H), 3.88 (s, 3H). ^{13}C NMR (126 MHz, CD_3CN): 175.10, 167.04, 164.62, 156.66, 139.61, 136.72, 131.81, 126.14, 115.63, 111.62, 99.98, 96.90, 69.69, 64.43, 52.41. MALDI-TOF: *m*/*z* calculated for $C_{19}H_{18}NO_3Ru^+$ ([M]$^+$): 409.4; found 409.2.



8-Hydroxy-*N*-(6-(naphthalen-2-yloxy)hexyl)-7,8-dihydroquinoline-5-carboxamide (S10). To a 20 mL glass vial was added 400 mg compound **S6**, 490 mg EDC, 390 mg HOBt, 620 mg compound **S3**, and 5 mL DMF. To the mixture was added 0.88 mL triethylamine and the mixture was stirred at room temperature for 16 h. The mixture was diluted with 15 mL H_2O and extracted with EtOAc (3 x 15 mL). The combined organic layers were washed with H_2O (3 x 10 mL). The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The crude product was purified by silica gel column chromatography (5% methanol/DCM) to afford a white solid (230 mg, 26% yield). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.97 (dd, *J* = 8.7, 1.6, 1H), 8.80 (dd, *J* = 4.2, 1.6, 1H), 7.77 – 7.69 (m, 3H), 7.65 (d, *J* = 7.9, 1H), 7.51 (dd, *J* = 8.7, 4.2, 1H), 7.45 – 7.40 (m, 1H), 7.32 (ddd, *J* = 8.0, 6.8, 1.2, 1H), 7.15 – 7.08 (m, 3H), 5.99 (s, 1H), 4.10 (t, *J* = 6.4, 2H), 3.54 (td, *J* = 7.2, 5.8, 2H), 1.94 – 1.84 (m, 2H), 1.72 (p, *J* = 7.3, 2H), 1.66 – 1.58 (m, 4H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 168.32, 157.15, 154.59, 148.43, 138.31, 135.32, 134.73, 129.50, 129.05, 127.79, 127.35, 126.83, 126.76, 126.48, 124.51, 123.67, 123.01, 119.09, 108.39, 106.71, 67.90, 40.14, 29.90, 29.29, 26.94, 26.05. High resolution ESI-MS: m/z calculated for $C_{26}H_{27}N_2O_3^+$ ($[M+H]^+$): 415.2016; found 415.2025.

8-(Allyloxy)-N-(6-(naphthalen-2-yloxy)hexyl)-7,8-dihydroquinoline-5-carboxamide (S11). To a 20 mL glass vial was added 120 mg compound **S10**, 80 mg potassium carbonate, and 3 mL dry DMF. To the mixture was added 31 µL allyl bromide and the mixture was stirred at room temperature for 20 h. The mixture was diluted with 20 mL H_2O and extracted with EtOAc (3 x 15 mL). The combined organic layers were washed with H_2O (3 x 15 mL). The organic layer was dried over Na_2SO_4, filtered, and concentrated by rotary evaporation. The crude product was

purified by silica gel column chromatography (10% methanol/DCM) to afford a white solid (68 mg, 51% yield). ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.97 (dd, *J* = 4.1, 1.7, 1H), 8.84 (dd, *J* = 8.6, 1.7, 1H), 7.78 – 7.69 (m, 3H), 7.58 (d, *J* = 8.1, 1H), 7.48 (dd, *J* = 8.6, 4.1, 1H), 7.43 (ddd, *J* = 8.1, 6.8, 1.3, 1H), 7.33 (ddd, *J* = 8.1, 6.9, 1.2, 1H), 7.16 – 7.10 (m, 2H), 6.93 (d, *J* = 8.1, 1H), 6.18 (ddt, *J* = 17.4, 10.7, 5.5, 1H), 5.97 (s, 1H), 5.46 (dq, *J* = 17.3, 1.5, 1H), 5.35 (dq, *J* = 10.6, 1.4, 1H), 4.86 (dt, *J* = 5.4, 1.6, 2H), 4.10 (t, *J* = 6.4, 2H), 3.55 (td, *J* = 7.1, 5.8, 2H), 1.94 – 1.86 (m, 2H), 1.72 (p, *J* = 7.3, 2H), 1.62 (p, *J* = 6.9, 2H), 1.58 – 1.51 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$): δ 168.50, 157.15, 156.27, 149.93, 140.45, 134.74, 134.47, 132.74, 129.51, 129.05, 127.79, 127.44, 126.83, 126.49, 126.40, 126.12, 123.69, 122.62, 119.10, 118.86, 107.61, 106.71, 70.06, 67.87, 40.13, 29.83, 29.26, 26.87, 26.02. High resolution ESI-MS: m/z calculated for $C_{29}H_{31}N_2O_3^+$ ($[M+H]^+$): 455.2329; found 455.2325.

Ru-**L9.** To a 4 mL glass vial was added 42.7 mg compound **S11**, 40.0 mg tris(acetonitrile)cyclopentadienylruthenium(II) hexafluorophosphate, and 2 mL dry DCM. The mixture was stirred at room temperature for 3 h. The crude product was precipitated by dropwise addition to 12 mL Et_2O and collected by centrifugation (5000 rpm, 5 min). The pellet was resuspended in 1 mL acetone and precipitated by dropwise addition to 13 mL Et_2O. The product was collected by centrifugation (5000 rpm, 5 min) and dried under vacuum to obtain a shimmery brown solid (53 mg, 75% yield). ^{1}H NMR (500 MHz, CD_3CN): δ 9.22 (dd, *J* = 8.7, 1.2, 1H), 8.63 (dd, *J* = 5.1, 1.2, 1H), 7.81 – 7.73 (m, 3H), 7.65 (d, *J* = 8.3, 1H), 7.55 (dd, *J* = 8.7, 5.1, 1H), 7.45 (ddd, *J* = 8.2, 6.8, 1.3, 1H), 7.34 (ddd, *J* = 8.1, 6.8, 1.2, 1H), 7.24 (d, *J* = 2.6, 1H), 7.12 (dd, *J* = 9.0, 2.5, 1H), 6.88 (s, 1H), 6.85 (d, *J* = 8.3, 1H), 5.95 (s, 5H), 4.50 (tt, *J* = 10.6, 6.2, 1H), 4.41 (d, *J* = 10.9, 1H), 4.17 – 4.07 (m, 5H), 3.38 (q, *J* = 6.6, 2H), 1.88 – 1.81 (m, 2H), 1.64 (p, *J* = 7.1, 2H),

1.60 – 1.52 (m, 2H), 1.52 – 1.45 (m, 2H). ^{13}C NMR (126 MHz, CD_3CN): 172.48, 168.00, 158.12, 156.66, 146.80, 139.83, 135.79, 131.43, 130.43, 130.28, 129.88, 128.54, 127.65, 127.43, 125.22, 124.58, 119.83, 119.23, 115.00, 107.70, 99.71, 96.80, 69.69, 68.86, 63.92, 40.33, 30.27, 29.87, 27.46, 26.54. MALDI-TOF: m/z calculated for $C_{34}H_{35}N_2O_3Ru^+$ ($[M]^+$): 620.7; found 620.4.

Allyl (4-methyl-2-oxo-2*H*-chromen-7-yl)carbamate. To a 4 mL glass vial was added 101 mg 7-amino-4-methylcoumarin and 2.0 mL DMF. To the mixture was added 93 µL pyridine and the mixture was chilled in an ice bath. To the mixture was added 72.8 µL allyl chloroformate dropwise and the mixture was allowed to stir at 0 °C for 4 h and at room temperature for 12 h. The mixture was partitioned between 4 mL 5% (v/v) aqueous HCl and 10 mL EtOAc. The aqueous layer was extracted with EtOAc (2 x 5 mL) and the combined organic layers were washed with sat. aqueous $NaHCO_3$ (2 x 5 mL). Volatiles were removed by rotary evaporation and the product was purified by silica gel column chromatography using gradient elution (100% hexane to 20% EtOAc/hexane) to afford a white solid (28 mg, 19% yield). ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.53 (d, J = 8.6, 1H), 7.43 (d, J = 2.1, 1H), 7.37 (dd, J = 8.6, 2.2, 1H), 6.85 (s, 1H), 6.19 (s, 1H), 6.03 – 5.92 (m, 1H), 5.39 (dq, J = 17.1, 1.7, 1H), 5.32 – 5.27 (m, 1H), 4.71 (dd, J = 5.8, 1.6, 2H), 2.41 (t, J = 0.9, 3H). ^{13}C NMR (126 MHz, $CDCl_3$): δ 161.11, 154.68, 152.77, 152.23, 141.43, 132.13, 125.53, 118.94, 115.78, 114.50, 113.45, 106.14, 66.49, 18.70. High resolution ESI-MS: *m*/*z* calculated for $C_{14}H_{14}NO_4^+$ ($[M+H]^+$): 260.0917; found 260.0915.

1.3. Polymer characterization

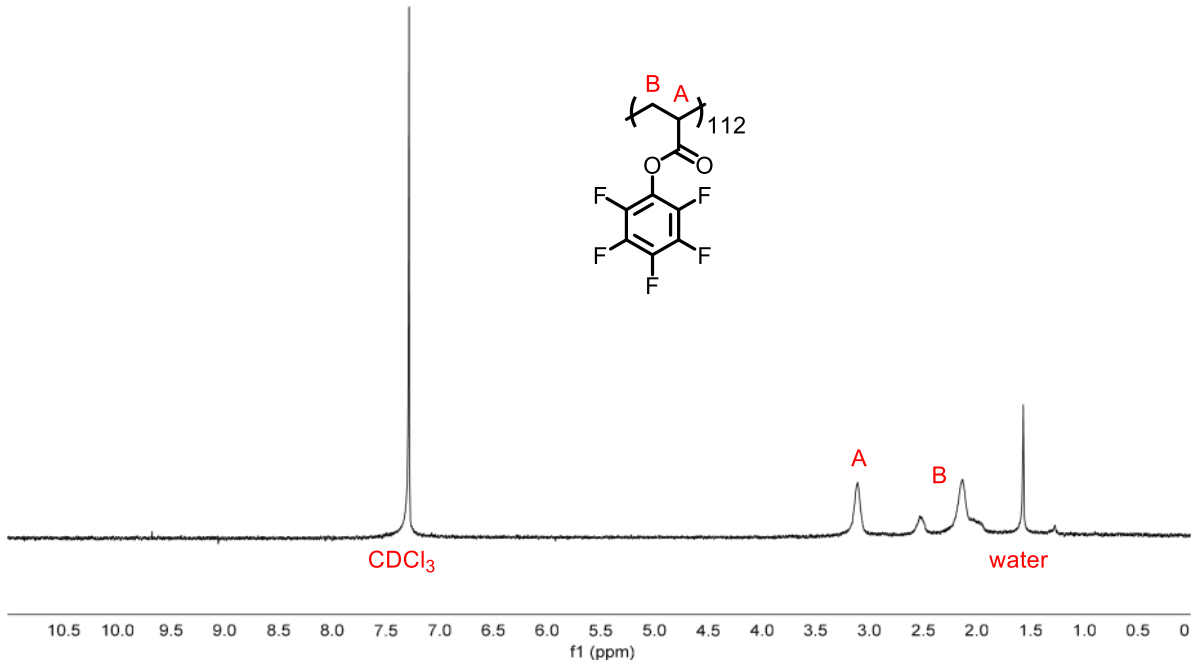


Figure S1. ^{1}H NMR spectrum ($CDCl_3$) of poly(pentafluorophenyl acrylate).

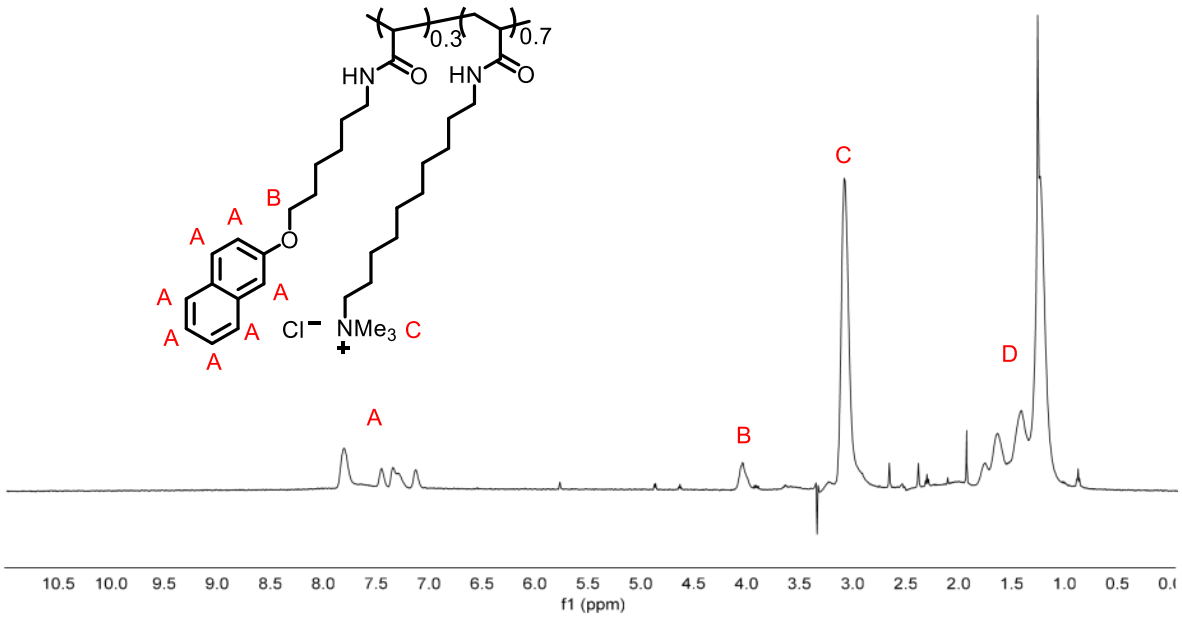


Figure S2. ^{1}H NMR spectrum (DMSO-d_6, H_2O suppression) of polymer **P1**. Overlapping proton peaks labeled "D" correspond to the polymer backbone and alkyl side chains.

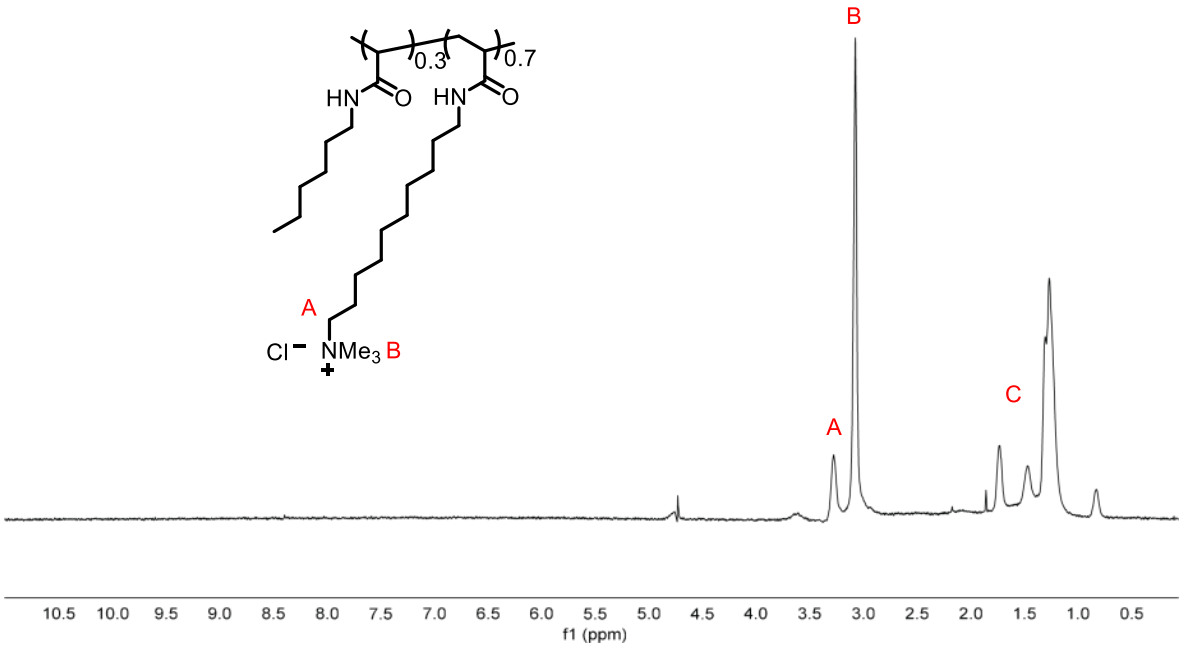


Figure S3. ^{1}H NMR spectrum (DMSO-d_6, H_2O suppression) of polymer **P2**. Overlapping proton peaks labeled "C" correspond to the polymer backbone and alkyl side chains.

2. NMR procedures

Nuclear Overhauser effect spectroscopy (NOESY). To an NMR tube was added polymer **P1**, ligand **L1** or **L2**, and D_2O to obtain final concentrations of 5 μM polymer and 100 μM ligand, and ^1H NMR and NOESY (relaxation delay = 2 s, mixing time = 0.3 s) were conducted.

Diffusion ordered spectroscopy (DOSY) of polymer. To an NMR tube was added polymer **P1** or **P2** and D_2O to obtain a final concentration of 1 μM polymer. DOSY was conducted using bipolar pulse pair stimulated echo (Dbppste) sequence (gradient strength = 2.7 G/cm to 62.31 G/cm, diffusion delay = 150 ms, gradient pulse = 2 ms). **P1** proton peaks at δ 2.96 and δ 1.24 ppm had a diffusion coefficient of 0.52 ± 0.03 and 0.69 ± 0.04 D (D = 10^{-10} m^2/s), respectively. **P2** proton peaks at δ 3.19, δ 2.98, δ 1.66, and δ 1.21 ppm had a diffusion coefficient of 0.71 ± 0.03, 0.64 ± 0.01, 0.66 ± 0.03, 0.62 ± 0.01 D, respectively. The hydrodynamic diameters of **P1** and **P2** were calculated from the Stokes-Einstein relation and the diffusion coefficient at δ 1.2 ppm.

Equation S1. $$D = \frac{kT}{6\pi\eta r}$$

where k = Boltzmann constant, T = absolute temperature (K), η = dynamic viscosity of D_2O at 25 ºC, and r = hydrodynamic radius.

DOSY of polymer and ligand. To an NMR tube was added polymer **P1**, ligand **L1**, and D_2O to obtain final concentrations of 1 μM polymer and 250 μM ligand. DOSY was conducted using bipolar pulse pair stimulated echo (Dbppste) sequence (gradient strength = 2.7 G/cm to 62.31 G/cm,

diffusion delay = 100 ms, gradient pulse = 2 ms). Polymer proton peaks at δ 3.03 and δ 1.03 ppm had a diffusion coefficient of 0.58 ± 0.02 and 0.71 ± 0.02 D, respectively. The ligand proton peak at δ 1.37 ppm had a diffusion coefficient of 1.4 ± 0.04 D.

Saturation-transfer difference (STD) NMR. To an NMR tube was added polymer **P1**, ligand **L1**, and D_2O to obtain final concentrations of 1 μM polymer and 250 μM ligand. STD NMR was conducted using irradiation at 0.4 ppm and saturation times of 1, 2, 3, 4, and 5 s. Ligand proton peaks at 7.66 and 1.41 ppm were integrated and the integrations were normalized to the *off-resonance* spectrum. STD amplification factor (A_{STD}) was calculated using Equation S2, where I_0 is the *off-resonance* intensity, I_{SAT} is the *on-resonance* intensity, $[L]_T$ is the ligand concentration, and [P] is the polymer concentration. A_{STD} was plotted against saturation time. Ligand mapping was achieved by setting the largest A_{STD} to 100% and calculating other A_{STD} with the same saturation time accordingly.

Equation S2. $$A_{STD} = \frac{I_0 - I_{SAT}}{I_0} x \frac{[L]_T}{[P]}$$

3. Fluorescence studies

General procedure for critical micelle concentration study. To 16 1 dram glass vials were added aliquots from stock solutions of Nile red and polymer to obtain final concentrations of 1 μM Nile red and 1 mg/mL (33 μM) to 10 ng/mL (330 pM) polymer. To the vials were added magnetic stirbars and the mixtures were stirred for 24 h. The fluorescence emission was measured on a fluorometer using excitation wavelength = 553 nm, scan emission wavelengths = 561 to 700 nm, and slit width = 3 nm. The fluorescence emission at 619 nm was plotted against polymer concentration. Using OriginPro, the CMC was calculated as the point of intersection between the sigmoidal fitted curve (solid blue) and a line (dotted red) passing through the point of inflection of the linear fitted curves (solid red) and perpendicular to the tangent of the sigmoidal fitted curve.

General procedure for CuAAC. To a 500 μL glass cuvette was added aliquots from stock solutions of polymer, $CuSO_4.5H_2O$ and ligand, azide **1**,[6] alkyne **2**, NaAsc, and PBS buffer or water to obtain final concentrations of 0 – 1 μM polymer, 0 – 5 μM Cu catalyst, 20 μM azide **1**, 40 μM alkyne **2**, and 2 mM NaAsc. The cuvette was capped and the mixture was stirred by pulsing on a vortex machine. CuAAC activity was monitored by measuring the increase in fluorescence on a fluorometer using excitation wavelength = 410 nm, emission wavelength = 480 nm, and slit width = 1.5 nm at room temperature for 30 min or 15 min. The % conversion was calculated based on a standard curve for the coumarin click product **3**. The initial rate was calculated by averaging the second derivative values of the linear segment of the fluorescence emission curves. Experiments were performed in triplicate and standard deviation was calculated for n = 3.

General procedure for Ru-catalyzed cleavage of allylcarbamate groups. To a 500 μL glass cuvette was added aliquots from stock solutions of polymer, Ru catalyst, caged coumarin **4**, GSH, and water to obtain final concentrations of 0 – 1 μM polymer, 0 – 5 μM Ru catalyst, 20 μM caged coumarin **4**, and 300 μM GSH. The cuvette was stirred by pulsing on a vortex machine. Ru-catalyzed cleavage of allylcarbamate groups was monitored by measuring the increase in fluorescence on a fluorometer using excitation wavelength = 375 nm, emission wavelength = 440 nm, and slit width = 2 nm at room temperature for 20 min. The % conversion was calculated based on a standard curve for the decaged coumarin product **5**. The initial rate was calculated by averaging the second derivative values of the linear segment of the fluorescence emission curves. Experiments were performed in triplicate and standard deviation was calculated for n = 3.

4. Additional figures and schemes

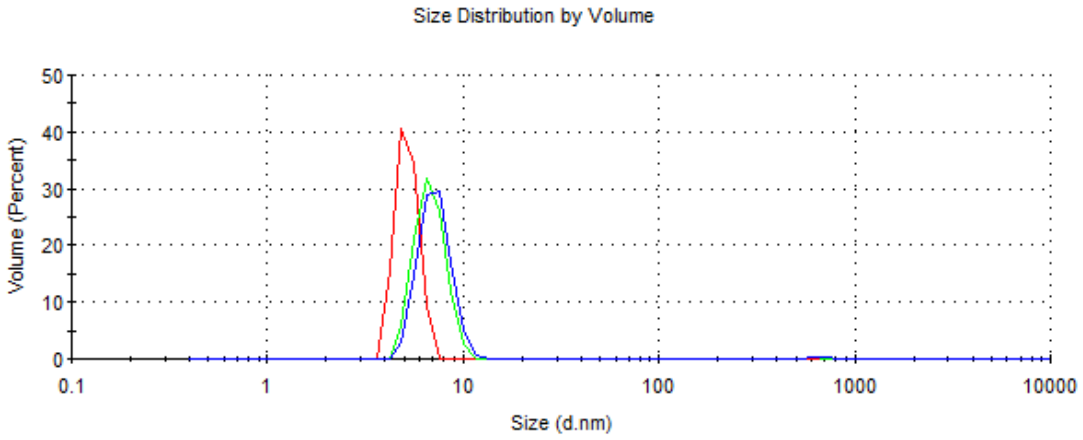


Figure S4. DLS size distribution by volume of 1 μM polymer **P1** in water. Curves are from technical replicates.

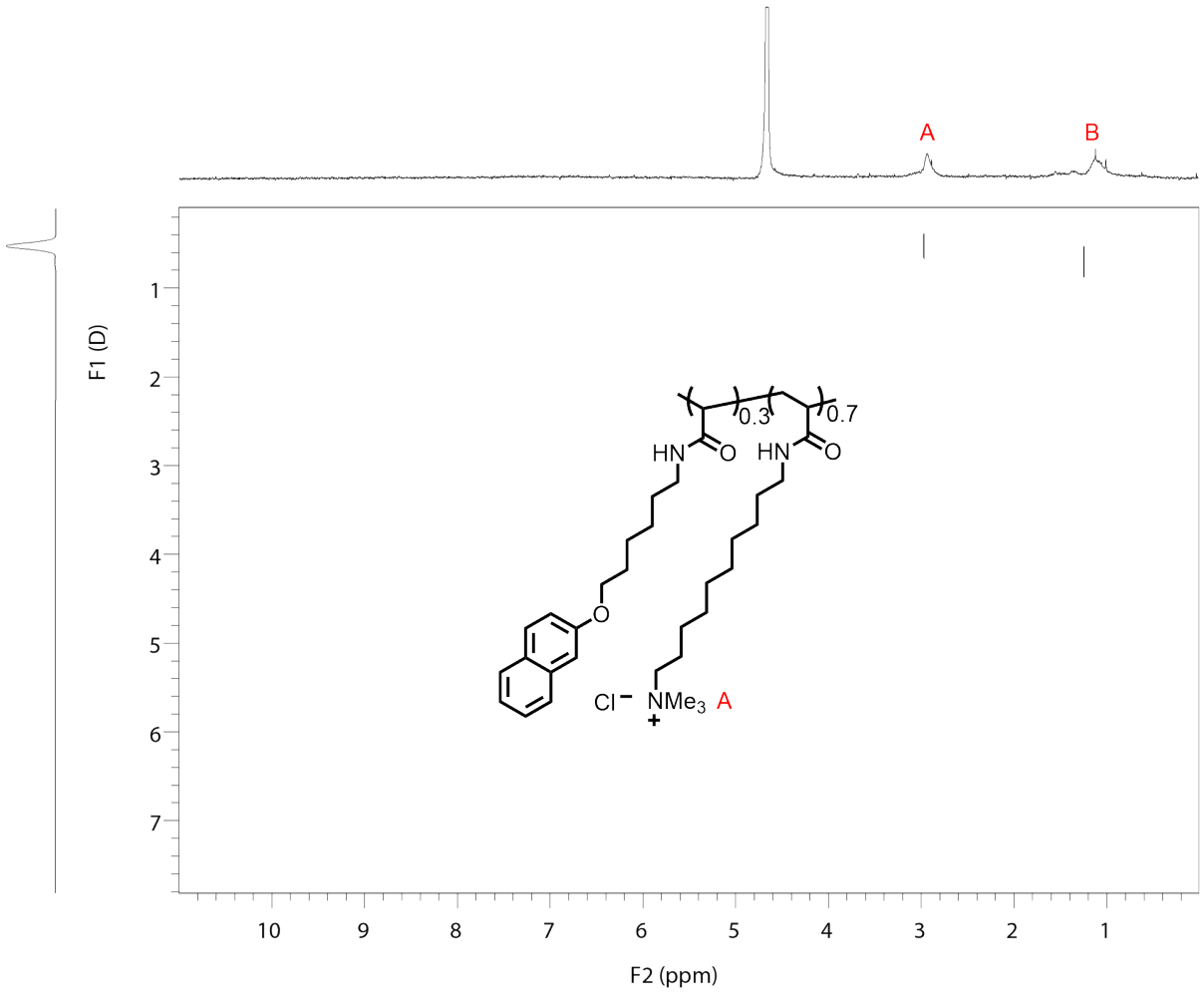


Figure S5. DOSY spectrum (D_2O) of polymer **P1**. Overlapping proton peaks labeled "B" correspond to the polymer backbone and alkyl side chains.

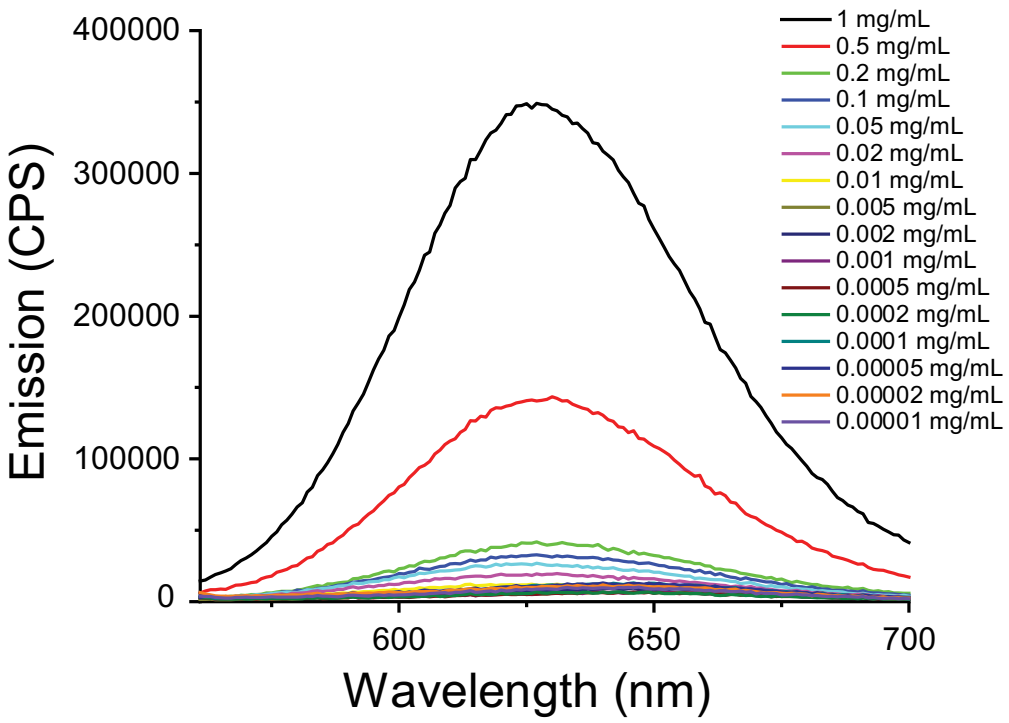


Figure S6. Plot of fluorescence emission vs wavelength for Nile red with 1 mg/mL (33 μM) to 10 ng/mL (330 pM) polymer **P2** in water. [Nile red] = 1 μM.

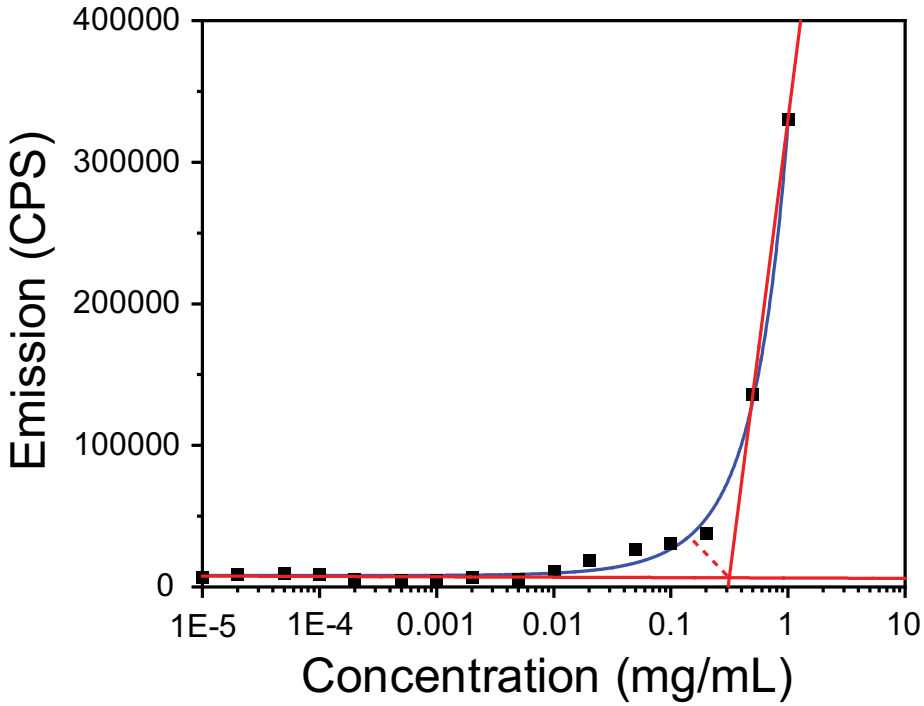


Figure S7. Plot of fluorescence emission (λ_{em} = 619 nm) vs concentration. Critical micelle concentration was calculated as 0.13 mg/mL (4.3 μM) polymer **P2**. [Nile red] = 1 μM.

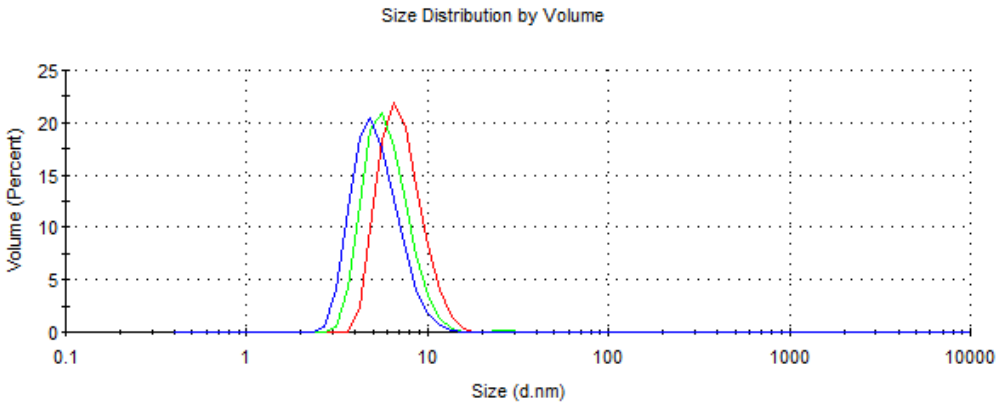


Figure S8. DLS size distribution by volume of 1 μM polymer **P2** in water. Curves are from technical replicates.

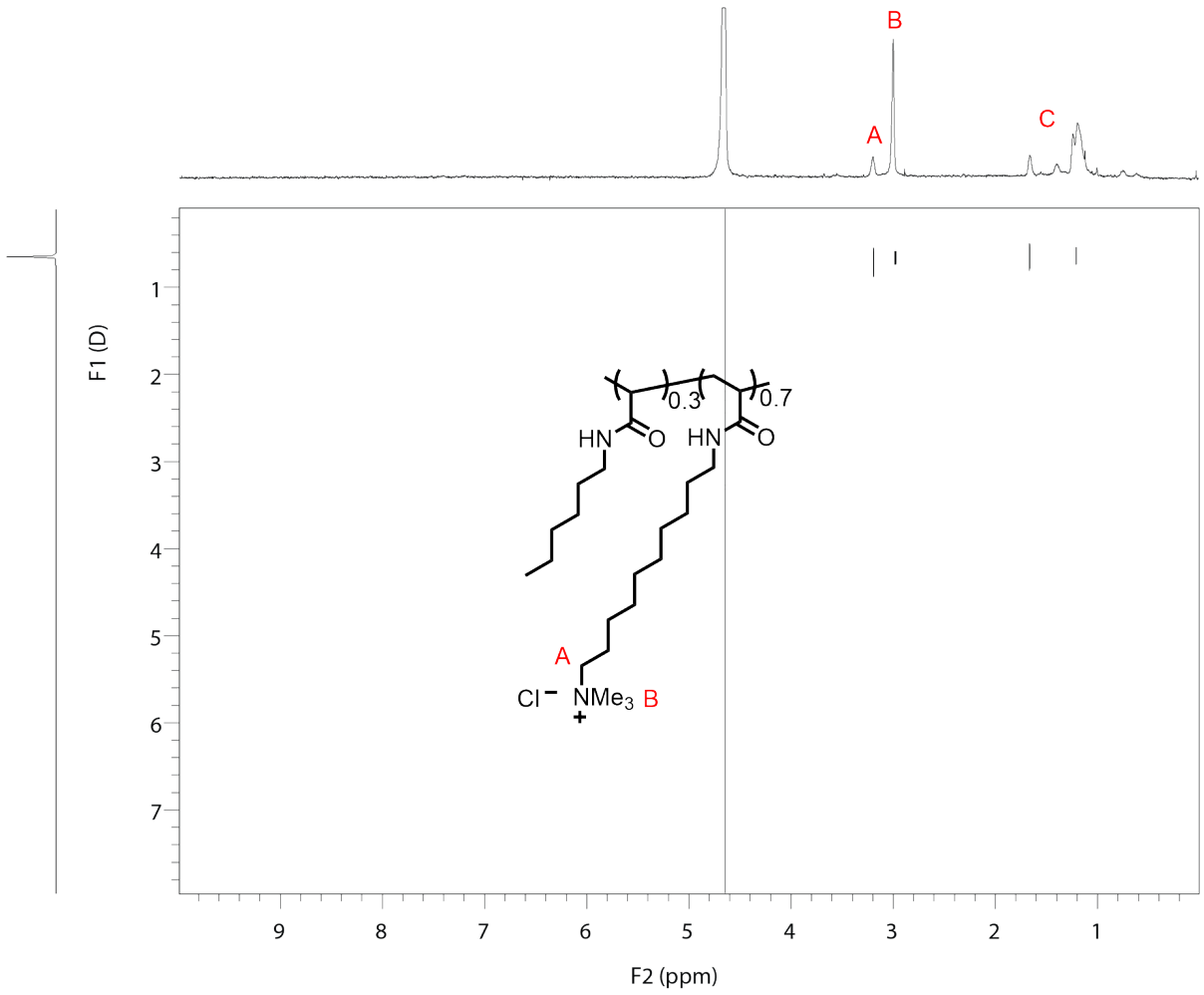


Figure S9. DOSY spectrum (D_2O) of polymer **P2**. Overlapping proton peaks labeled "C" correspond to the polymer backbone and alkyl side chains.

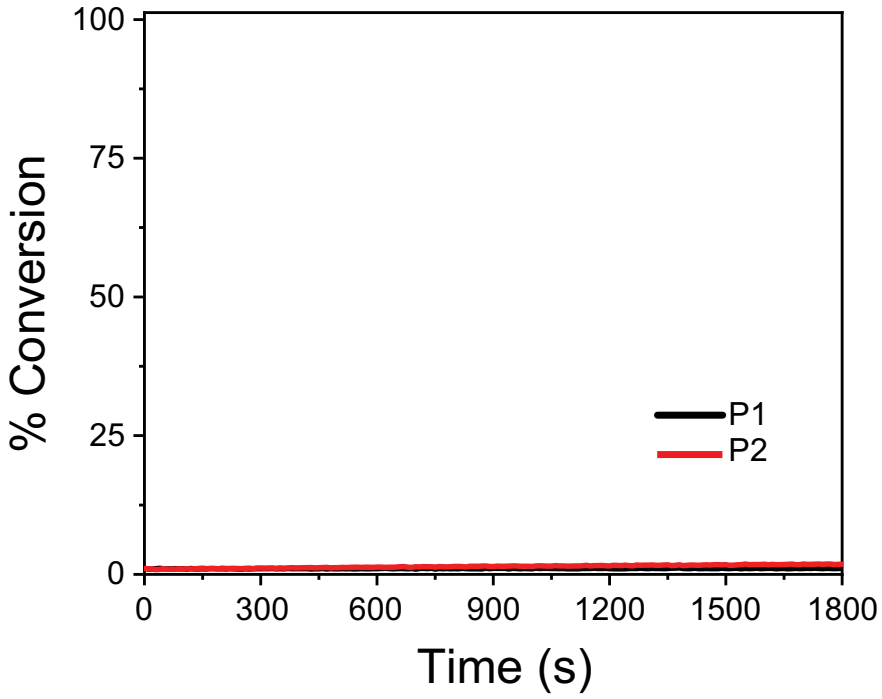


Figure S10. Fluorescence emission curves of CuAAC reaction between 3-azido-7-hydroxycoumarin and 1-ethynyl-4-methoxybenzene in the presence of polymer **P1** and polymer **P2**.

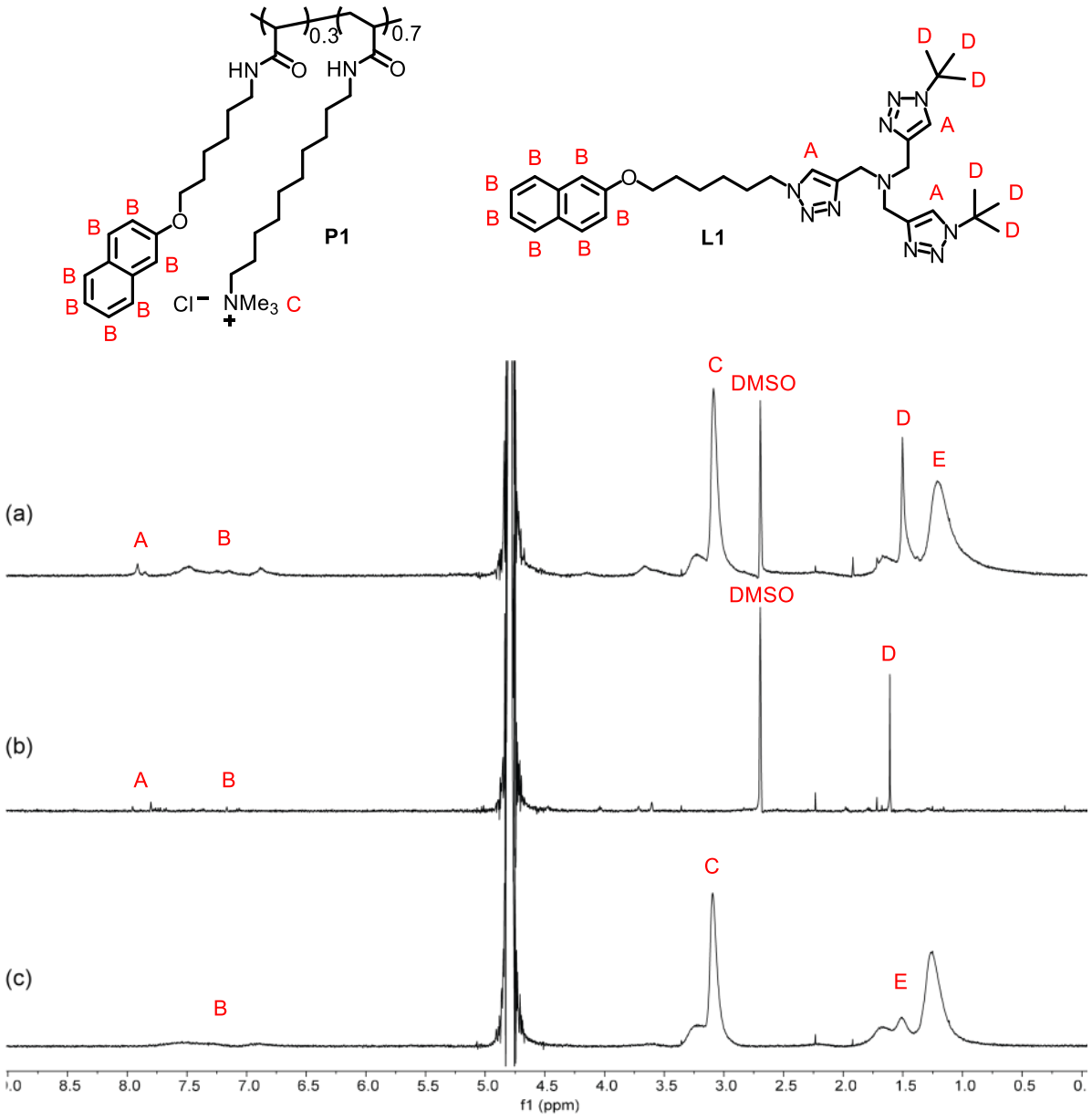


Figure S11. Stacked ^{1}H NMR spectra (D_2O) of (a) polymer **P1** and ligand **L1**, (b) ligand **L1**, and (c) polymer **P1**. Overlapping proton peaks labeled "E" correspond to the polymer backbone and alkyl side chains. Please refer to these proton peak assignments for the following NMR spectra.

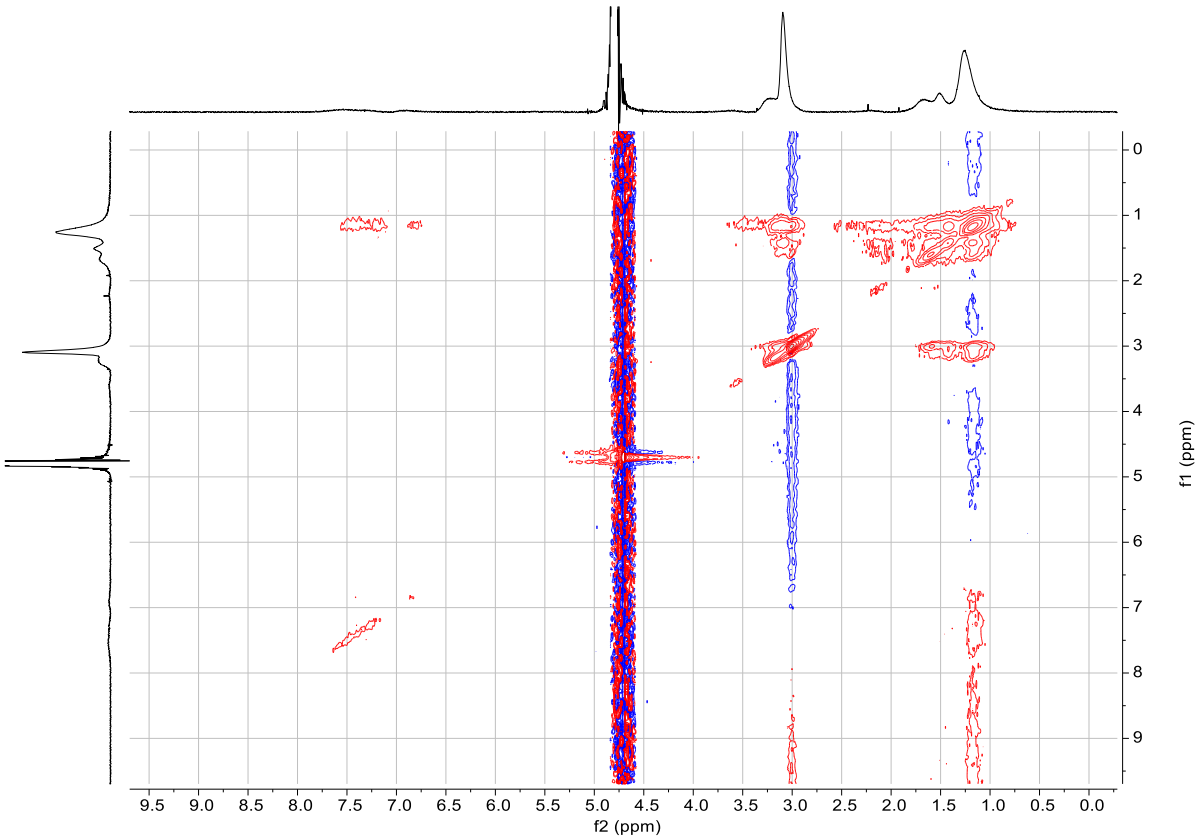


Figure S12. NOESY spectrum (D_2O) of polymer **P1**.

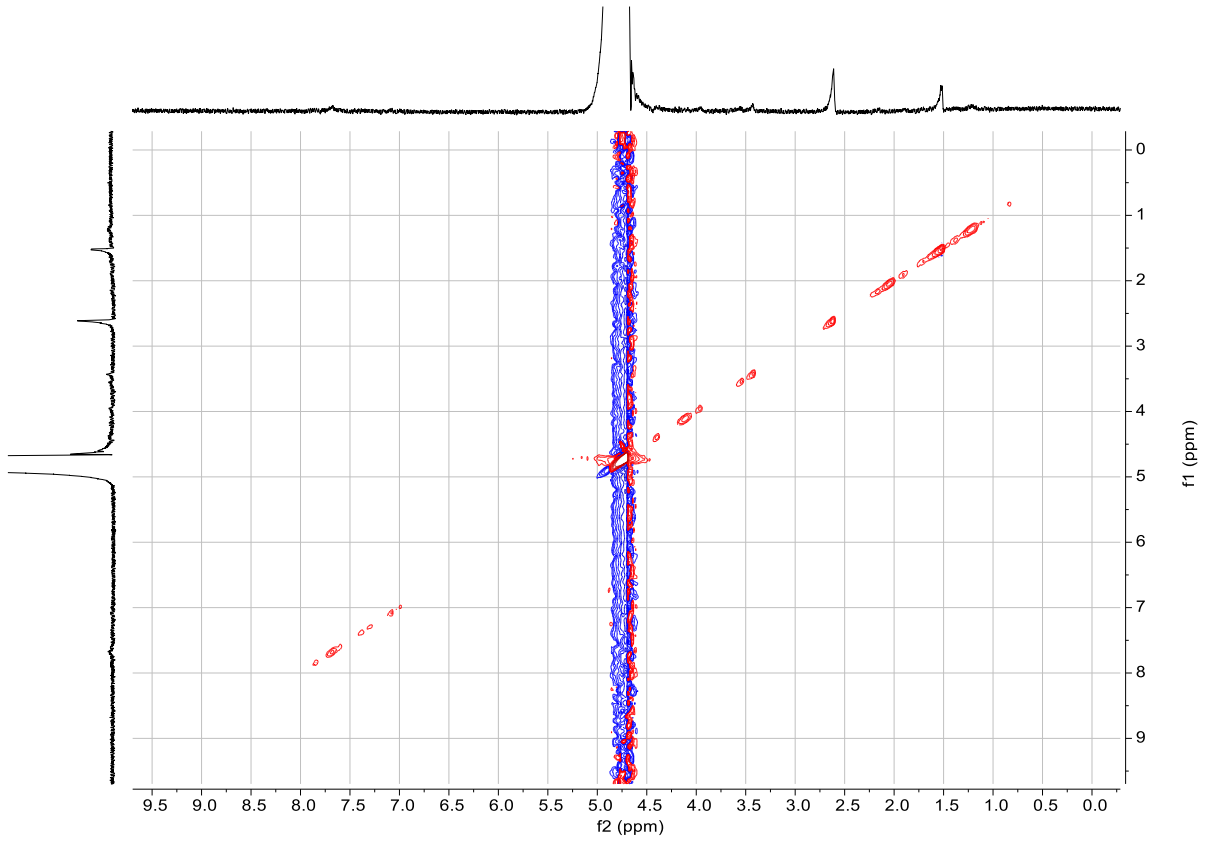


Figure S13. NOESY spectrum (D_2O) of ligand **L1**.

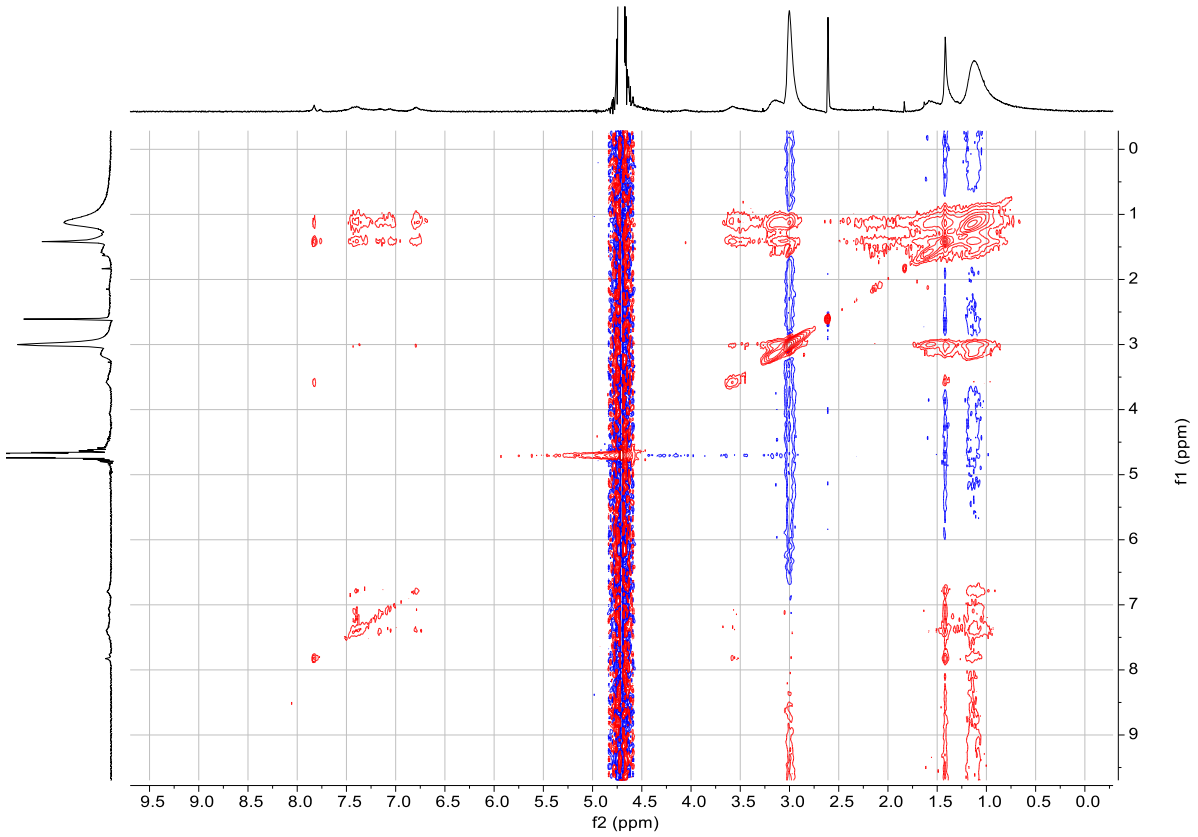


Figure S14. NOESY spectrum (D_2O) of polymer **P1** and ligand **L1**.

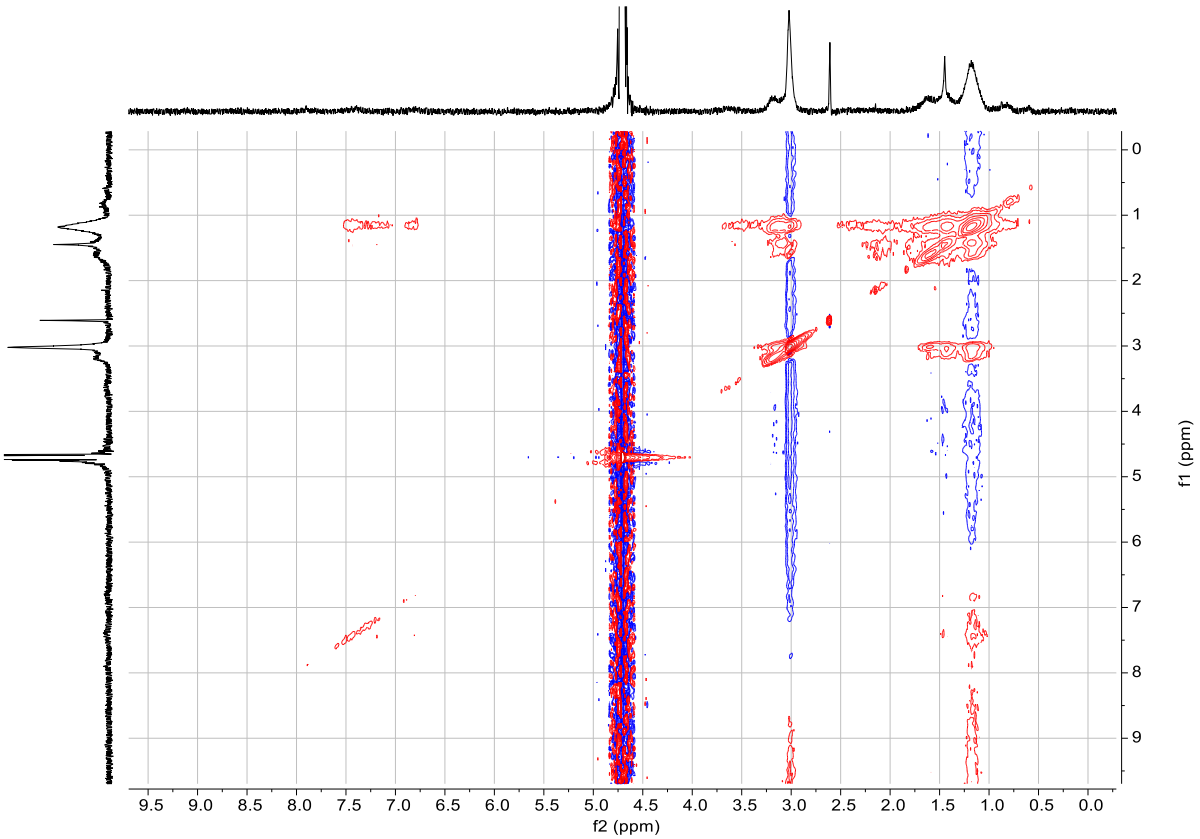


Figure S15. NOESY spectrum (D_2O) of polymer **P1** and ligand **L2**.

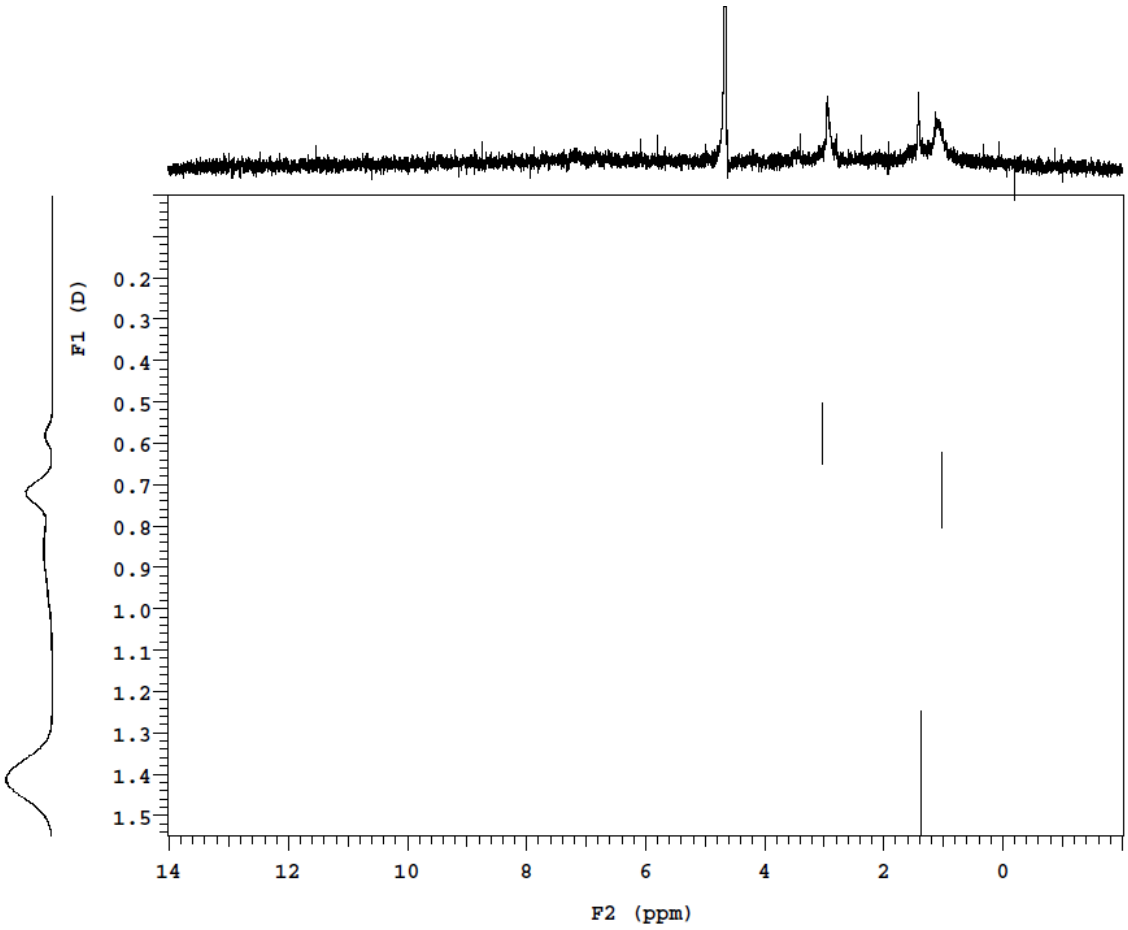


Figure S16. DOSY spectrum (D_2O) of polymer **P1** and ligand **L1**.

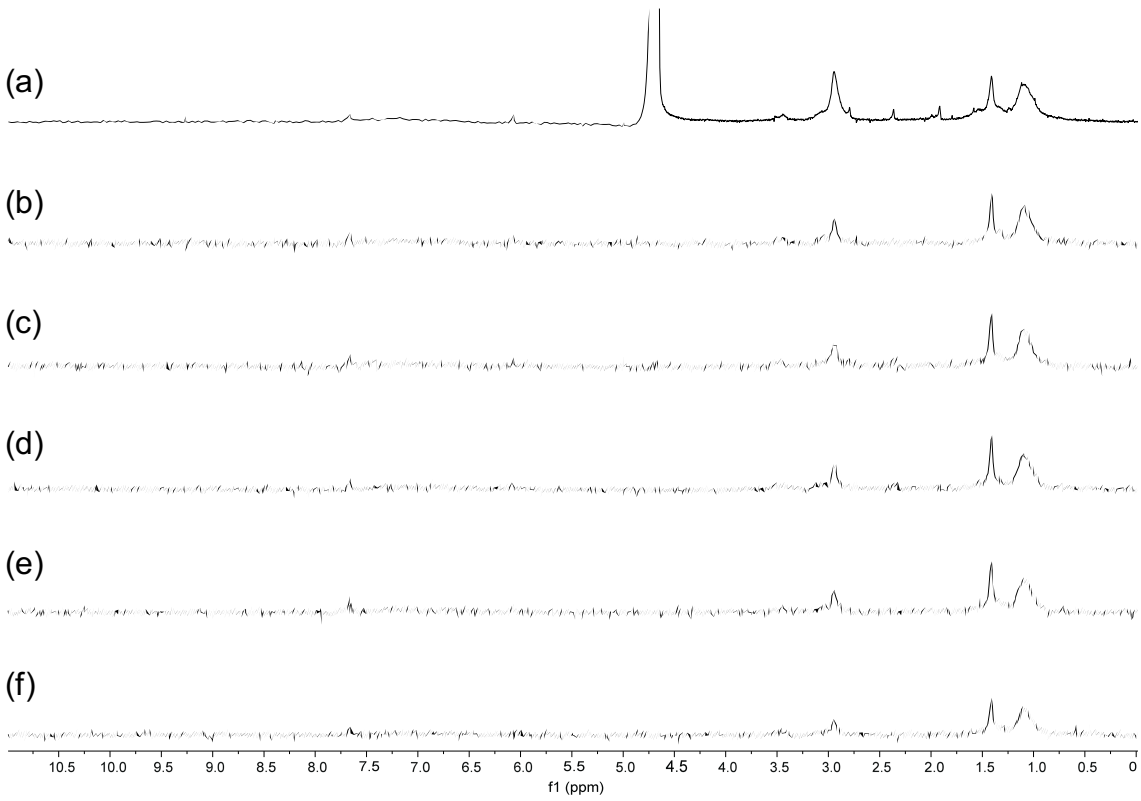


Figure S17. Stacked (a) *off-resonance* spectrum of polymer **P1** and ligand **L1** and STD difference spectra with (b) 5, (c) 4, (d) 3, (e) 2, and (f) 1 s saturation time.

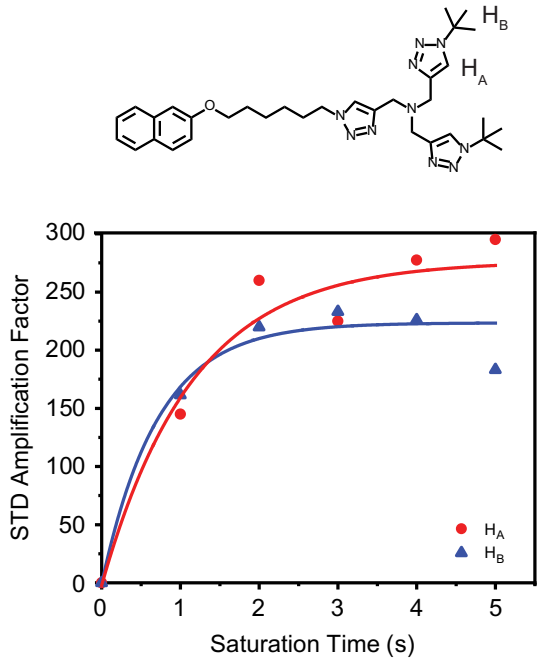


Figure S18. Graph of A_{STD} vs saturation time for triazole and *tert*-Bu proton peaks.

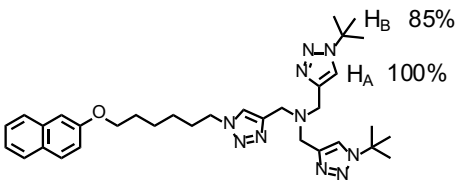


Figure S19. Ligand mapping for ligand **L1** with A_{STD} values at 2 s saturation time.

5. Notes and References

1. A. Das and P. Theato, *Macromolecules*, 2015, **48**, 8695-8707.
2. J. Chen, J. Wang, K. Li, Y. Wang, M. Gruebele, A. L. Ferguson and S. C. Zimmerman. *J. Am. Chem. Soc.*, 2019, **141**, 9693-9700.
3. D. Soriano del Amo, W. Wang, H. Jiang, C. Besanceney, A. C. Yan, M. Levy, Y. Liu, F. L. Marlow and P. Wu, *J. Am. Chem. Soc.*, 2010, **132**, 16893-16899.
4. X. Han, R. Wang, X. Song, F. Yu, C. Lv and L. Chen, *Biomaterials*, 2018, **156**, 134-146.
5. J. Budhathoki-Uprety, L. Peng, C. Melander and B. M. Novak, *ACS Macro Lett.*, 2012, **1**, 370-374.
6. K. Sivakumar, F. Xie, B. M. Cash, S. Long, H. N. Barnhill and Q. Wang, *Org. Lett.*, 2004, **6**, 4603-4606.