Supporting Information for:

Comparison of diffusion coefficients for matched pairs of macrocyclic and linear molecules over a drug-like molecular weight range

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General Considerations. All reagents and solvents were used as received. THF was distilled from sodium. NMR spectra were recorded on Bruker DRX-600, Bruker DRX-500, or Bruker AMX-400 instruments using residual solvent peak as a reference. Data are reported as s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, br = broad. LC/MS analyses were carried out using an Agilent Technologies HPLC (Agilent Technologies 1100 Series diode array detector, Agilent Technologies 1100 Series column heater, Agilent 1100 Series pump, and Agilent 1100 Series degasser) interfaced with an Agilent Technologies 6110 Quadrupole LC/MS. Column chromatography was performed using a Biotage Horizon automated flash chromatography system equipped with a Biotage Horizon detector, fraction collector and pump where noted.

NMR Diffusion Coefficient Determination. All the NMR experiments were acquired on a Bruker Avance 600 MHz spectrometer, equipped with a 5 mm TCI cryoprobe with z-gradients capable of generating 54 G/cm field strengths. The temperature controller was set to 298K with an air flow of 535 l/h in order to avoid any temperature fluctuations due to sample heating during acquisition and to avoid sample vibrations from a high air-flow. Samples were made up to 10mM solutions in DMSO-d_6 or CDCl3 with some TMS vapour, and 180uL of this solution was added to a 3 mm NMR tube to avoid problems of convection. In the case of samples in 10% DMSO in D_2O, samples were made up at 2.5 mM and 600 uL of this solution was added to a 5mm NMR tube. The lower concentration was required due to the limited solubility in this solvent. To maintain good signal to noise, the use of 5mm NMR tubes is possible for more viscous solvents, as the onset of Rayleigh-Benard convection is ablated at the temperatures used during this investigation.

Diffusion coefficients were determined with a high degree of reproducibility on the NMR system used. Using robust statistical analysis, we have determined that the relative differences in diffusion between the linear and macrocyclic analogues are significant. However, the absolute accuracy of the diffusion coefficient as measured by NMR is limited by the accuracy of the calibration of the gradient and temperature, and often prone to errors introduced during the calculation method. Steps were taken to minimise these errors as detailed, however, it would be difficult to accurately compare diffusion coefficients between different NMR systems. The gradient strength was calibrated using the diffusion coefficient of water in a standard solution of 0.1 mg/ml $GdCl_3$, 0.1% DSS, 1% H_2O in D_2O. The values of the measured diffusion coefficient (D) of water, the known diffusion coefficient of water and the current gradient calibration value (gc(old)) were used to obtain the new gradient calibration value (gc(new)) using the following equation:[1]

$$gc(new) = gc(old) \times sqrt.(D(measured)/ D(known))$$

The temperature was calibrated with a sample of methanol-d4 (99.8 at%), sealed under atmospheric pressure, using details described elsewhere.[2]

All DOSY experiments used the ledbpgp2s sequence (available in standard Bruker pulse sequence library). A gradient duration (d) of 2msec and an eddy current delay of 5 ms was used in all cases. The diffusion time (D) was 100 ms in the case of $CDCl_3$, and 200 ms in the case of d_6-DMSO and 10% d_6-DMSO in D_2O. In each PFG NMR experiment, a series of 16-32 spectra on 16K data points were collected, using a linear gradient ramp from 5-95% of the maximum gradient strength.

After acquisition, the data was zero filled to 32k, Fourier transformed and baseline corrected in f2. The diffusion coefficients were calculated with the T1/T2 relaxation module using mono-

exponential fitting, rather than the 2D processing protocol. This is available in Bruker Topspin v.2. For each sample, several well resolved signals were used to extract individual diffusion coefficients (an example of the raw data and the fitting report is included in this supporting information). These signals have been averaged for each run to give the diffusion coefficient and a standard deviation. Statistical analysis confirms that the variation in diffusion coefficient within each run is the same as the comparison between runs. The analysis also demonstrates that a difference in diffusion coefficients between macrocycles and linear controls of 1% would be statistically significant. The observed difference of 5% is highly statistically significant.

TMS was included in the $CDCl_3$ and DMSO datasets, as a control to check temperature was consistent in all experiments. As can be seen in the table of diffusion data, this is very reproducible and allows confidence in the comparison of data between macrocycles and linear analogues.

LC/MS Analysis. HPLC analyses was performed using a water (formic acid 0.1% w/v / ammonium formate 0.05% w/v) and MeCN (water 5% v/v, formic acid 0.1% v/v, ammonium formate 0.05% w/v) based gradient from 0-100% MeCN over 4 minutes. A Waters XBridge C18 2.5 μm (3.0 x 30 mm) column was used at 80 ˚C with a flow rate of 2.4 mL min^{-1}. Injections were made from diluted reaction mixtures and ionization monitored in positive or negative mode.

Copper Reactor Diskette. The copper tubing used in the Conjure flow reactor comes in the form of a reactor diskette. Copper tubing (0.75 mm inner diameter, 3.0 m length, 1500 μL internal volume) is housed between two metal plates (Figure 1a). Figure 1b shows the fully

assembled reactor diskette (145 mm x 165 mm x 5 mm).

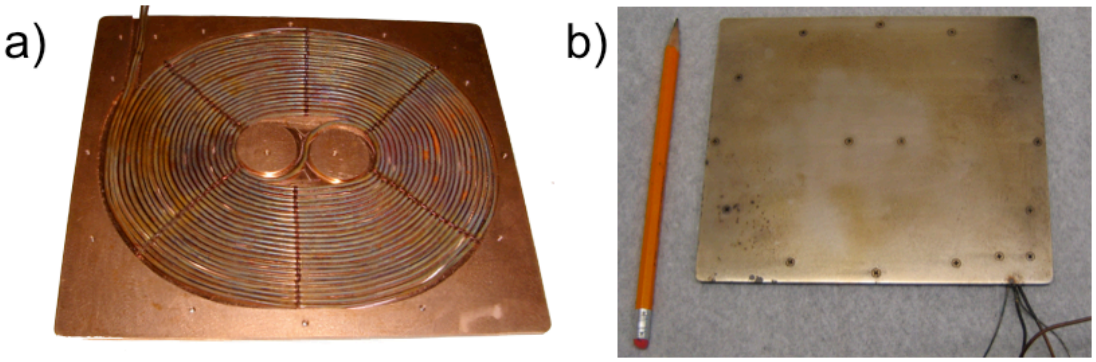
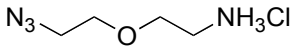


Figure S1. The copper reactor diskette.

Synthesis of Azido Linkers.

4-azidobutyric acid and 6-azidohexanoic acid were synthesized according to literature precedence.[3]

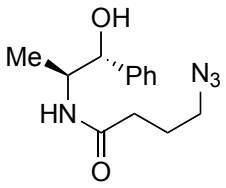


9. CAUTION: Extreme care should be taken with preparing low molecular weight organic azides. Bis(2-bromoethyl) ether (11.5 g, 50.0 mmol, 1.0 eq) and NaN_3 (8.13 g, 125.0 mmol, 2.5 eq) were suspended in DMF at 55 ˚C until the reaction was deemed complete by TLC (~3 hrs). DI H_2O (50 mL) was added to the reaction flask, and the solution was transferred to a separatory funnel. The aqueous phase was extracted with Et_2O (3x 100 mL), the organic extracts were washed with brine, dried over Na_2SO_4 and filtered. The solvent was removed under a stream of

N_2 until the total volume was ~50 mL. Aqueous HCl (2.5 M, 45 mL, 112.5 mmol, 2.25 eq) was added to the organic phase and stirred at 0 ˚C. PPh_3 (12.5 g, 47.6 mmol, 0.95 eq) in EtOAc (50 mL) was added to the ether/HCl mixture drop wise over 2 hours at 0 ˚C, and the reaction was allowed to warm to room temperature and stir overnight.

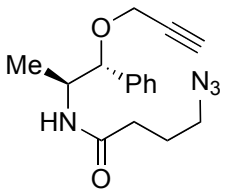
Excess Et_2O (50 mL) was added to the reaction mixture and the phases were allowed to separate. The organic phase was extracted with aqueous HCl (1 M, 50 mL). The combined aqueous extracts were washed with Et_2O (2x 50 mL), concentrated at 60 ˚C under a stream of N_2, and dried under vacuum to yield **9** as a light brown solid (7.6 g, 96% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 3.76 (d, J = 4.8 Hz, 2 H), 3.71 (t, J = 4.2 Hz, 2 H), 3.46 (t, J = 5.4 Hz, 2 H), 3.15 (t, J = 4.8 Hz, 2 H); ^{13}C NMR (150 MHz, CD_3OD): δ 71.0, 67.8, 51.6, 40.6.

Synthesis of Macrocycle Precursors.



10: General Procedure A. (1*R*,2*S*)-(-)-norephedrine (1.0 g, 6.6 mmol, 1.0 eq) and PyBOP (3.43 g, 6.6 mmol, 1.0 eq) were dissolved in dry CH_2Cl_2 (50 mL) under Ar. Diisopropylethylamine (3.43 mL, 19.8 mmol, 3.0 eq) was added to the reaction mixture and the reaction was cooled to 0 ˚C using an ice bath. 4-azidobutyric acid (940.0 mg, 7.3 mmol, 1.1 eq) was dissolved in CH_2Cl_2 (5 mL) and slowly added to the cooled reaction mixture. The reaction was allowed to warm to room temperature and continued to stir overnight.

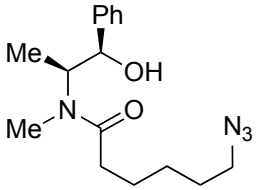
To the reaction mixture was added DI H_2O (50 mL). The mixture was stirred vigorously then transferred to a separatory funnel. The aqueous phase was extracted with CH_2Cl_2 (3x 25 mL) and the combined organic extracts were dried over Na_2SO_4, filtered and concentrated *in vacuo*. The residue was dissolved in Et_2O and the solids (PyBOP impurities) were filtered off. The solution was concentrated *in vacuo* and the residue purified by column chromatography (silica gel, 1:2 hexanes/EtOAc, $R_f = 0.33$) to yield **10** as a yellow oil (1.06 g, 63% yield): ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.28 - 7.40 (m, 5 H), 5.60 (d, J = 7.3 Hz, 1 H), 4.87 (t, J = 3.3 Hz, 1 H), 4.35 (m, 1 H), 3.37 (t, J = 6.4 Hz, 2 H), 3.29 (d, J = 3.3 Hz, 1 H), 2.26 - 2.33 (m, 2 H), 1.88 - 2.01 (m, 2 H), 1.04 (d, J = 7.0 Hz, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 172.3, 140.5, 128.2, 127.7, 126.3, 66.2, 51.0, 50.7, 33.2, 24.8, 14.8. m/z = 263.1 $[M+H]^+$.



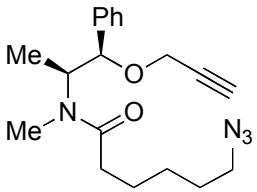
11: General Procedure B. 10 (1.06 g, 4.04 mmol, 1.0 eq) was dissolved in dry THF (40 mL) at room temperature. NaH (dry, 120 mg, 4.8 mmol, 1.2 eq) was added to the reaction mixture and allowed to stir until gas evolution ceased. Propargyl bromide (80% w/w in toluene, 520 µL, 4.8 mmol, 1.2 eq) was added drop wise and allowed to stir overnight.

The reaction was quenched carefully with DI H_2O and concentrated. The resulting residue was purified using column chromatography (silica gel, 1:1 hexanes/EtOAc, $R_f = 0.45$) to yield **11** as a yellow oil (1.00 g, 83% yield): ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.28 - 7.42 (m, 5 H), 5.90 (d, J=7.5 Hz, 1 H), 4.72 (br. s., 1 H), 4.30 (d, J = 15.8 Hz, 1 H), 4.24 (br. s., 1 H), 3.95 (d, J = 16.2 Hz, 1 H), 3.35 (t, J = 6.6 Hz, 2 H), 2.45 (s, 1 H), 2.29 (t, J = 7.2 Hz, 2 H), 1.94 (m, 2

H), 1.00 (d, J = 7.0 Hz, 3 H); ^{13}C NMR (150 MHz, $CDCl_3$): δ 170.8, 137.6, 128.5, 127.9, 126.8, 81.9, 79.7, 74.6, 56.5, 50.7, 49.6, 33.4, 24.8, 13.7. m/z = 301.1 $[M+H]^+$.

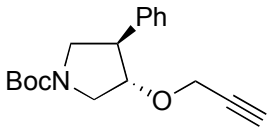


12. Prepared according General Procedure A using (1*R*,2*S*)-(-)-ephedrine (495.0 mg, 3.0 mmol, 1.0 eq), PyBOP (1.56 g, 3.0 mmol, 1.0 eq), diisopropylethylamine (1.6 mL, 9.2 mmol, 3.0 eq) and 6-azidohexanoic acid (520.0 mg, 3.3 mmol, 1.1 eq) in dry CH_2Cl_2 (75 mL). The crude reaction mixture was purified using column chromatography (silica gel, 1:1 hexanes/EtOAc, R_f = 0.31) to yield **12** as a colorless oil (850.0 mg, 93% yield): ^{1}H NMR (4.1:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.28 - 7.34 (m, 5 H), 4.80 (t, 1 H), 4.60* (t, 1 H), 4.49 (m, 1 H), 4.03 (s, 1 H), 3.93* (m, 1 H), 3.24 (t, J = 7.0 Hz, 2 H), 3.17* (t, J = 7.0 Hz, 2 H), 2.79* (s, 3 H), 2.67 (s, 3 H), 1.56 (m, 4 H), 1.34 (m, 2 H), 3.17 (d, J = 7.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 174.5, 172.5*, 142.4*, 142.0, 128.7*, 128.3, 126.5, 126.2*, 77.5, 76.0*, 58.3, 57.9*, 51.4, 34.1, 33.1, 32.7*, 28.9, 26.6, 24.6, 15.2*, 12.4. m/z = 305.2 $[M+H]^+$.



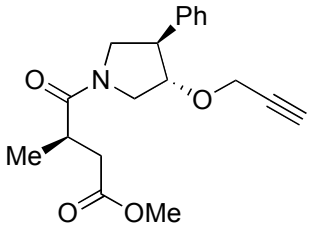
13. Prepared according General Procedure B using **12** (425.0 mg, 1.4 mmol, 1.0 eq), NaH (60% w/w dispersion in mineral oil, 84 mg, 2.1 mmol, 1.5 eq), propargyl bromide (80% w/w in

toluene, 230 µL, 2.1 mmol, 1.5 eq) in dry THF (25 mL). The resulting residue was purified using column chromatography (silica gel, 4:1 hexanes/EtOAc, R_f = 0.20) to yield **13** as a yellow oil (420.0 mg, 88% yield): ^{1}H NMR (2:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.25 - 7.32 (m, 5 H), 4.70 (m, 1 H), 4.65 (d, *J* = 5.5 Hz, 1 H), 4.46* (d, *J* = 8.0 Hz, 1 H), 4.14 (m, 1 H), 4.00* (m, 1 H), 3.82 (m, 1 H), 3.24 (m, 2 H), 2.86 (s, 3 H), 2.81* (s, 3 H), 2.41* (t, *J* = 2.5 Hz, 1 H), 2.36 (t, *J* = 2.5 Hz, 1 H), 2.15 (m, 2 H), 1.47 - 1.58 (m, 4 H), 1.36* (d, *J* = 6.0 Hz, 3 H), 1.29 (m, 2 H), 1.18 (d, *J* = 6.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 172.8, 138.7, 128.9*, 128.5, 128.1, 127.4, 127.2*, 83.2, 82.2*, 80.1, 79.5*, 77.4, 74.9*, 74.3, 57.5*, 56.3, 51.5, 33.9, 32.7, 29.0, 28.8*, 26.7, 24.7*, 24.6, 15.4, 12.1. m/z = 343.2 $[M+H]^+$.



14. Prepared according General Procedure B using tert-butyl 3-hydroxy-4-phenylpyrrolidine-1-carboxylate (racemic, 14.53 g, 55.2 mmol, 1.0 eq), NaH (dry, 1.60 g, 66.7 mmol, 1.2 eq), propargyl bromide (80% w/w in toluene, 8.8 mL, 81.8 mmol, 1.5 eq) in dry THF (400 mL). The resulting residue was purified using column chromatography (silica gel, 33-100% EtOAc in hexanes gradient, R_f = 0.50 in 3:1 hexanes/EtOAc) to yield racemic **14** as a brown oil (16.5 g, 99% yield). The racemic mixture was resolved by preparative chiral SFC using a Chiralpak AD-H column (4.6 x 250 mm) using a 10% MeOH in CO_2 solvent system (3.0 mL/min, 140 bar). Two peaks were collected into separate fractions, yielding **14** and the (3*R*,4*S*) enantiomer in a 1:1 ratio: ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.30 (m, 2 H), 7.22 (m, 3 H), 4.19 (m, 1 H), 4.10 (m, 2 H), 3.63 - 3.82 (m, 2 H), 3.46 - 3.57 (m, 1 H), 3.34 - 3.43 (m, 2 H), 2.40 (s, 1 H), 1.46 (m, 9 H); ^{13}C

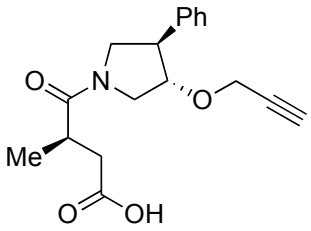
NMR (asterisks denote minor rotamer peaks, 150 MHz, $CDCl_3$): δ 154.5, 140.0, 139.9*, 128.9, 127.35, 127.34*, 127.25, 127.20*, 83.2*, 82.3, 79.7, 79.4, 75.04, 75.00*, 57.1, 57.0*, 50.4, 50.2*, 49.63, 49.61*, 49.5, 48.5*, 28.6. m/z = 245.9 $[M+H, - tBu]^+$.



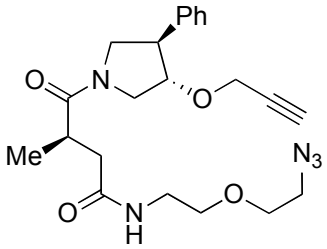
15: General Procedure C. 14 (516.6 mg, 1.7 mmol, 1.0 eq) was dissolved in 4.0 M HCl in 1,4-dioxane (3.0 mL, 12.0 mmol, 7.0 eq) and stirred at room temperature until the deprotection was complete by TLC. The solvent was removed under a stream of N_2 and the residue was dried under vacuum. The residue was dissolved in CH_2Cl_2 (20 mL), and PyBOP (890 mg, 1.71 mmol, 1.0 eq), diisopropylethylamine (1.48 mL, 8.5 mmol, 5.0 eq) and (R)-(+)-3-methylsuccinic acid 1-monomethyl ester (275 mg, 1.88 mmol, 1.1 eq) were added and the reaction allowed to stir overnight at room temperature.

To the reaction mixture was added DI H_2O (50 mL). The mixture was stirred vigorously then transferred to a separatory funnel. The aqueous phase was extracted with CH_2Cl_2 (3x 25 mL) and the combined organic extracts were dried over Na_2SO_4, filtered and concentrated *in vacuo*. The residue was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, R_f = 0.20 in 1:1 hexanes/EtOAc) to yield **15** as a light brown oil (475.1 mg, 81% yield): ^1H NMR (500 MHz, $CDCl_3$): δ 7.19 - 7.31 (m, 5 H), 4.12 - 4.29 (m, 3 H), 3.87 - 3.96 (m, 2 H), 3.78 (m, 1 H), 3.70 (m, 1 H), 3.63 (s, 3 H), 3.55 (m, 1 H), 2.95 - 3.08 (m, 1 H), 2.83 - 2.93 (m, 1 H), 2.40 - 2.42 (m, 1 H), 2.34 (m, 1 H), 1.15 (d, J = 7.0 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks,

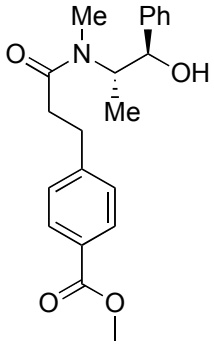
125 MHz, $CDCl_3$): δ 174.3, 174.2*, 173.1, 173.0*, 139.6, 129.1, 129.0*, 127.5*, 127.41, 127.39*, 127.3, 83.8*, 82.0, 79.5*, 79.4, 75.2*, 75.1, 57.2*, 57.1, 51.9*, 51.8, 50.45, 50.42*, 49.8*, 49.6, 49.4*, 47.8, 37.91*, 37.88, 34.3*, 34.1, 17.5, 17.3*. m/z = 330.2 $[M+H]^+$.



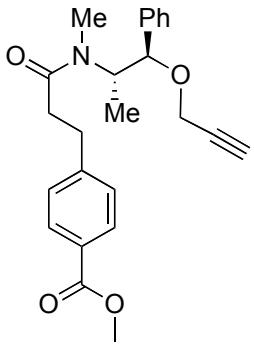
16: General Procedure D. 15 (475.1 mg, 1.44 mmol, 1.0 eq) was dissolved in THF/MeOH (25 mL, 3:2 v/v). LiOH monohydrate (242 mg, 5.8 mg, 4.0 eq) was dissolved in DI H_2O (5 mL) and added drop wise to the solution of **15** in THF/MeOH and allowed to stir overnight at RT. The reaction was concentrated and the resultant residue was acidified using 5 M HCl. The residue was partitioned between DI H_2O and CH_2Cl_2, and the aqueous phase extracted with CH_2Cl_2 (2x 25 mL). The organic phase was dried over Na_2SO_4, concentrated and dried *in vacuo* to yield **16** as a light brown oil (454 mg, >99% yield): ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.29 - 7.32 (m, 2 H), 7.18 - 7.25 (m, 3 H), 4.14 - 4.30 (m, 3 H), 3.88 - 3.97 (m, 2 H), 3.66 - 3.81 (m, 2 H), 3.58 (m, 1 H), 2.95 - 3.09 (m, 1 H), 2.79 - 2.94 (m, 1 H), 2.40 - 2.54 (m, 2 H), 1.20 (d, J = 7.0 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 176.5, 176.5*, 175.1*, 175.0, 139.4, 139.3*, 129.2, 129.1*, 127.6, 127.5*, 127.28, 127.25*, 83.2*, 82.1, 79.4, 79.3*, 75.4*, 75.3, 57.2*, 57.1, 50.6*, 50.5, 49.8*, 49.7, 49.6, 47.6*, 38.13*, 38.06, 34.3*, 34.1, 17.0, 16.8*. m/z = 316.2 $[M+H]^+$.



17. Prepared according General Procedure A using **9** (100.0 mg, 0.60 mmol, 1.1 eq), PyBOP (170 mg, 0.54 mmol, 1.0 eq), diisopropylethylamine (470 µL, 2.69 mmol, 5.0 eq) and **16** (170.0 mg, 0.54 mmol, 1.0 eq) in dry CH_2Cl_2 (30 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 20-100% EtOAc in hexanes gradient, $R_f = 0.41$ in EtOAc) to yield **17** as a light yellow oil (223.2 mg, 97% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.24 - 7.35 (m, 5 H), 4.26 - 4.39 (m, 1 H), 4.15 - 4.25 (m, 2 H), 3.82 - 4.04 (m, 2 H), 3.68 - 3.78 (m, 1 H), 3.51 - 3.68 (m, 5 H), 3.32 - 3.51 (m, 5 H), 3.08 - 3.20 (m, 1 H), 2.88 (m, 1 H), 2.59 - 2.66 (m, 1 H), 2.28 - 2.32 (m, 1 H), 1.20 (d, $J = 4.2$ Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 176.9, 176.8*, 174.14, 174.07*, 140.9*, 140.8, 129.90*, 129.89*, 128.5, 128.3, 128.2*, 84.1*, 83.0, 80.47*, 80.46, 76.34*, 76.30, 70.93*, 70.92, 70.44*, 70.43, 57.76*, 57.75, 51.74*, 51.72, 51.64*, 51.52, 51.0, 50.6*, 50.4, 40.56, 40.55*, 40.36*, 40.33, 35.8*, 35.5, 17.4, 17.2*. m/z = 428.3 $[M+H]^+$.

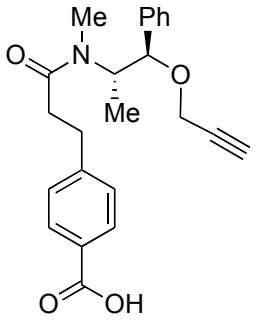


18. Prepared according General Procedure A using (1*R*,2*S*)-(-)-ephedrine (825 mg, 5.0 mmol, 1.0 eq), PyBOP (2.6 g, 5.0 mmol, 1.0 eq), diisopropylethylamine (2.6 mL, 15.0 mmol, 3.0 eq) and 3-(4-Methoxycarbonyl)propionic acid methyl ester (1.14 g, 5.5 mmol, 1.1 eq) in dry CH_2Cl_2 (40 mL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, $R_f = 0.16$ in 1:1 hexanes/EtOAc) to yield **18** as a colorless oil (1.07 g, 60% yield): ^{1}H NMR (3.2:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$) δ 7.93 (d, J = 9.5 Hz, 2 H), 7.87* (d, J = 8.0 Hz), 7.21 - 7.32 (m, 7 H), 7.05* (d, J = 8.0 Hz, 2 H), 4.81 (m, 1 H), 4.58* (m, 1 H), 4.49 (m, 1 H), 3.82 - 3.92 (m, 4 H), 2.94 (t, J = 7.5 Hz, 2 H), 2.79* (s, 3 H), 2.64 (s, 3 H), 2.52 (t, J = 8.5 Hz, 2 H), 1.29* (d, J = 6.0 Hz, 3 H), 1.16 (d, J = 7.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$) δ 173.5, 171.9*, 167.2, 147.2*, 147.0, 142.3*, 142.0, 130.0, 129.9*, 128.7, 128.6*, 128.5, 128.4*, 128.36, 128.32*, 128.1*, 127.7, 126.4, 126.2*, 77.4, 75.9*, 58.3, 58.0*, 52.20, 52.16*, 35.6, 34.2*, 33.1, 31.28*, 31.25, 28.5*, 15.3*, 12.3. m/z = 356.0 $[M+H]^+$.



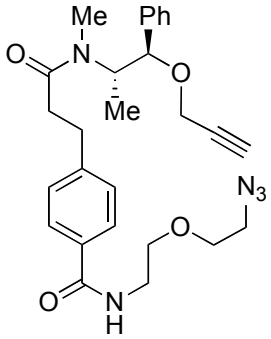
19. Prepared according General Procedure B using **18** (1.07 g, 3.01 mmol, 1.0 eq), NaH (dry, 90 mg, 3.75 mmol, 1.3 eq), propargyl bromide (80% w/w in toluene, 480 μL, 4.5 mmol, 1.5 eq) in dry THF (50 mL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 8-100% EtOAc in hexanes gradient, $R_f = 0.33$

in 2:1 hexanes/EtOAc) to yield **19** as a colorless oil (900 mg, 76% yield): ^{1}H NMR (1.9:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.90 (m, 2 H), 7.19 - 7.32 (m, 7H), 7.08* (d, *J* = 6.5 Hz, 2 H), 4.65 (m, 2 H), 4.30* (d, *J* = 8.5 Hz, 1 H), 4.13 (m, 1 H), 3.94* (m, 1 H), 3.86 (m, 3 H), 3.80 (m, 1 H), 2.88 (m, 2 H), 2.83 (s, 3 H), 2.79* (s, 3 H), 2.40 - 2.51 (m, 2 H), 2.39* (t, *J* = 2.5 Hz, 1 H), 2.33 (t, *J* = 2.5 Hz, 1 H), 1.29* (d, *J* = 7.0 Hz, 3 H), 1.15 (d, *J* = 6.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 171.8, 171.7*, 167.2, 147.3, 147.2*, 138.6, 138.2*, 129.9, 129.8*, 128.9, 128.8*, 128.6, 128.5*, 128.4, 128.2*, 128.08, 128.06*, 127.2, 127*, 83.1, 82.0*, 80.0, 79.4*, 74.9*, 74.3, 57.5, 56.26, 56.22*, 54.6*, 52.13, 52.12*, 35.4, 34.1*, 31.7*, 31.2*, 31.1, 28.4, 15.6, 12.2*. m/z = 394.0 $[M+H]^+$.



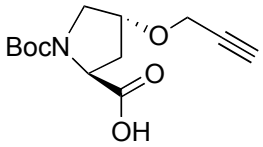
20. Prepared according to General Procedure D using **19** (900 mg, 2.3 mmol, 1.0 eq), LiOH monohydrate (386 mg, 9.2 mg, 4.0 eq) and THF/MeOH/DI H_2O (15 mL, 3:2:1 v/v) to yield **20** as a light brown solid (823 mg, 94% yield): ^{1}H NMR (1.9:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 8.00 (m, 2 H), 7.22 - 7.35 (m, 5 H), 7.15 (m, 2 H), 4.69 (m, 2 H), 4.46* (d, *J* = 7.5 Hz, 1 H), 4.16 (m, 1 H), 3.98* (m, 1 H), 3.82 (m, 1 H), 2.93 (m, 2 H), 2.87 (s, 3 H), 2.84* (s, 3 H), 2.70* (m, 2 H), 2.46 - 2.56 (m, 2 H), 2.42* (t, *J* = 2.5 Hz, 1 H), 2.36 (t, *J* = 2.5 H, 1 H), 2.07 - 2.13* (m, 2 H), 1.32 (d, *J* = 6.5 Hz, 3 H), 1.19 (d, *J* = 7.0 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 172.1. 172.0*, 171.6*, 171.5,

148.0, 138.5, 138.2*, 130.6, 130.5*, 128.92, 128.88*, 128.7, 128.6*, 128.5, 128.4*, 127.7, 127.6*, 127.3, 127.2*, 83.1, 81.9*, 80.0, 79.4*, 74.9*, 74.4, 57.7, 56.3, 56.2*, 35.4, 34.1*, 31.8*, 31.4*, 31.2, 28.6, 15.6*, 12.2. m/z = 379.9 $[M+H]^+$.



21. Prepared according General Procedure A using **9** (164.0 mg, 1.0 mmol, 1.0 eq), PyBOP (513 mg, 1.0 mmol, 1.0 eq), diisopropylethylamine (864 µL, 5.0 mmol, 5.0 eq) and **20** (965.0 mg, 6.6 mmol, 1.1 eq) in dry CH_2Cl_2 (30 mL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, EtOAc, $R_f = 0.41$) to yield **21** as a colorless oil (207.8 mg, 43% yield): ^{1}H NMR (1.9:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.67 (m, 2 H), 7.05 - 7.34 (m, 7 H), 6.59 (br. s., 1 H), 4.69 (m, 1 H), 4.65 (d, J = 5.0 Hz, 1 H), 4.42* (d, J = 7.5 Hz, 1 H), 4.13 (m, 1 H), 3.94* (m, 1 H), 3.81 (m, 1 H), 3.58 - 3.70 (m, 6 H), 3.36 (m, 2 H), 2.74 - 2.90 (m, 5 H), 2.40 - 2.51 (m, 2 H), 2.38* (m, 1 H), 2.33 (m, 1 H), 1.29* (d, J = 6.5 Hz, 3 H), 1.14 (d, J = 6.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 171.9, 171.8*, 167.5*, 167.4, 145.63, 145.58*, 138.6, 138.3*, 132.3, 132.2*, 128.9, 128.8*, 128.7, 128.6*, 128.5, 128.1*,

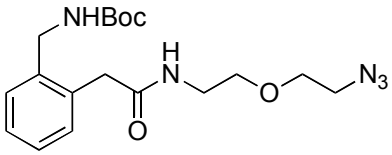
127.34, 127.26*, 127.24, 127.17*, 83.2, 82.0*, 80.0, 79.5*, 74.9*, 74.3, 70.3, 70.2*, 57.5, 56.30, 56.25*, 50.8, 39.8, 35.5, 34.3, 31.1*, 31.0, 28.5, 15.6*, 12.2. m/z = 492.3 $[M+H]^+$.



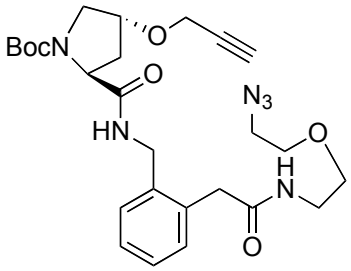
22. To a 1 L-3 necked-RBF equipped with a stir bar and an internal thermometer was added 1-*tert*-butyl 2-methyl (2*S*,4*R*)-4-hydroxypyrrolidine-1,2-dicarboxylate (19.9 g, 81 mmol, 1.0 eq) and THF (300 mL). The clear solution was evacuated and back filled with nitrogen gas and cooled to -78 ˚C. When the internal temperature was at about -50 ˚C, NaH (60 wt % dispersion in mineral oil, 5 g ca., 122 mmol, 1.5 eq) was added in small portions. The reaction flask was quickly capped and charged with nitrogen and stirred for 10 min. Propargyl bromide (80 wt % in toluene, 9.5 mL, 122 mmol, 1.5 eq) was added *via* a syringe over 5 min. During the addition the internal temperature was at about -60 ˚C. Stirring continued and the reaction was allowed to warm to ambient temperature overnight. After 20 h at ambient temperature, LCMS analysis of the reaction mixture showed that starting material still remained. The reaction was cooled again to about -60 ˚C. Additional NaH (60 wt % dispersion in mineral oil, 5 g, 122 mmol, 1.5 mol eq) was added. At about -60 ˚C, propargyl bromide (80 wt % in toluene, 10 mL, 122 mmol, 1.5 mol eq) was added via a syringe over 5 min. Stirring continued and the reaction was allowed to warm to ambient temperature for 4 h. LCMS analysis of the reaction mixture showed major and clean product presence. The reaction mixture was cooled to -65 ˚C and was carefully quenched with water (few drops first, total 20 mL). The reaction mixture was allowed to warm to ambient temperature and was used in the next reaction without further purification.

To the above reaction crude solution in THF-water mixture was added solid lithium hydroxide (3.9 g, 162 mmol, 2 mol eq in 40 mL water). The reaction mixture was stirred at ambient temperature for 1.5 h. LCMS analysis of the reaction mixture showed clean and major product presence. The volatiles were removed *in vacuo*, providing an oil, which was suspended in water. To remove mineral oil from NaH dispersion, the reaction crude mixture was partitioned in heptane (100 mL). The organic layer was separated. The aqueous layer was cooled in an ice bath and was acidified to pH 4 using acetic acid. The aqueous phase was extracted with CH_2Cl_2 (2 x 250 mL). The organic layer was separated. The aqueous phase was then saturated with solid NaCl and extracted with CH_2Cl_2 (2 x 100 mL). The organic layer was separated. The combined organic layer was dried over Na_2SO_4 and evaporated to give an oil (44 g). The above oil was dissolved in ethyl acetate (10 mL). Heptane (100 mL) was slowly added. The mixture was stirred at ambient temperature for 30 min. A light yellow gummy solid came out and was stirred for another 10 min. The light clear top liquid was decanted. The remaining light gummy solid was again dissolved in ethyl acetate (10 mL) and heptane (100 mL) was slowly added with stirring until lightly cloudy. The mixture was stirred for 30 min. The clear liquid was decanted. The remaining oil was evaporated. This oil upon cooling solidified to a light yellow solid, which was dried to give the title compound **22** (17.64 g). ^{1}H NMR analysis showed acetic acid presence (10 mole %). This solid was then dissolved in acetonitrile (30 mL) and water (100 mL). The solution was lyophilized to a residue. This residue was dissolved in ethyl acetate (20 mL). To this solution was added heptane (total 220 mL) with stirring. The mixture was stirred at ambient temperature for 1 h. The clear liquid was decanted. The resulting material was dried *in vacuo* to compound **22** as a light yellow solid (16.61 g, 76% yield): ^{1}H NMR (500 MHz, DMSO-d_6): δ 4.21 (m, 1 H), 4.16 (d, J=2.45 Hz, 2 H), 4.04 - 4.09 (m, 1 H),

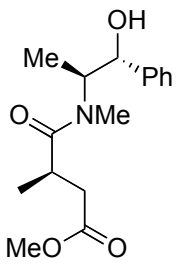
3.38 - 3.49 (m, 3 H), 2.24 - 2.35 (m, 1 H), 1.93 - 2.02 (m, 1 H), 1.39 (s, 3 H) 1.33 (s, 6 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, DMSO-d_6): δ 174.1, 173.6*, 153.5*, 153.1, 80.3, 78.9, 77.2, 76.2*, 75.4, 57.6, 57.4*, 55.7*, 55.6, 51.6*, 51.2, 35.5, 34.7*, 28.1*, 27.9. m/z = 268.1 $[M-H]^+$.



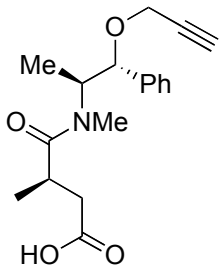
23. Prepared according General Procedure A using **9** (350.0 mg, 2.1 mmol, 1.1 eq), PyBOP (990 mg, 1.9 mmol, 1.0 eq), diisopropylethylamine (1.66 mL, 10.5 mmol, 5.0 eq) and N-Boc-2-aminomethyl-phenylacetic acid (500.0 mg, 1.9 mmol, 1.0 eq) in dry CH_2Cl_2 (50 mL). The residue was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 20-100% EtOAc in hexanes gradient, R_f = 0.51 in EtOAc) to yield **23** as a white solid (596.2 mg, 83% yield): ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.21 - 7.32 (m, 4 H), 6.18 (br. s., 1 H), 5.34 (br. s., 1 H), 4.30 (m, 2 H), 3.58 (m, 4 H), 3.50 (t, J = 5.4 Hz, 2 H), 3.41 (q, J = 5.4 Hz, 2 H), 3.29 (t, J = 4.8 Hz, 2 H), 1.42 (s, 9 H); ^{13}C NMR (150 MHz, $CDCl_3$): δ 171.0, 156.2, 137.7, 133.5, 131.1, 129.8, 128.3, 128.1, 79.6, 77.0, 69.8, 50.8, 42.5, 40.9, 39.5, 28.6. m/z = 278.2 $[M+H, -Boc]^+$.



24. Prepared according to General Procedure C using **23** (596.2 mg, 1.58 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (3.0 mL, 12.0 mmol, 7.6 eq). The peptide coupling was carried out using Boc-deprotected **23** (150.0 mg, 0.48 mmol, 1.1 eq), PyBOP (230 mg, 0.44 mmol, 1.0 eq), diisopropylethylamine (380 µL, 2.2 mmol, 5.0 eq) and **22** (117.0 mg, 0.44 mmol, 1.0 eq) in dry CH_2Cl_2 (30 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 20-100% EtOAc in hexanes gradient, R_f = 0.49 in EtOAc) to yield **24** as a colorless oil (232.6 mg, >99% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.17 - 7.42 (m, 4 H), 4.35 - 4.56 (m, 2 H), 4.15 - 4.34 (m, 4 H), 3.54 - 3.77 (m, 8 H), 3.30 - 3.43 (m, 3 H), 2.86 (m, 1 H), 2.39 (m, 1 H), 2.02 (m, 1 H), 1.27 - 1.48 (m, 9 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 175.0, 174.8*, 173.9*, 173.8, 156.4*, 156.0, 138.3, 137.9*, 135.6, 135.3*, 131.61, 131.57*, 130.7, 130.1*, 139.0, 128.9*, 128.6, 128.49*, 129.46, 81.7, 81.5*, 80.6*, 80.5, 78.0*, 77.2, 76.05, 76.02*, 70.9, 70.4*, 60.9, 60.5*, 57.03*, 56.96, 55.8, 53.5*, 53.0*, 51.8, 43.8, 42.3*, 42.1, 40.8, 40.7*, 40.60, 40.56*, 38.0, 37.2*, 37.0*, 28.7*, 28.5. m/z = 529.3 $[M+H]^+$.



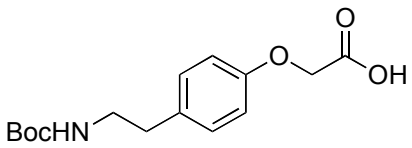
25. Prepared according General Procedure A using (1*R*,2*S*)-(-)-ephedrine (660.0 mg, 4.0 mmol, 1.0 eq), PyBOP (2.1 g, 4.4 mmol, 1.0 eq), diisopropylethylamine (2.1 mL, 12.0 mmol, 3.0 eq) and (R)-(+)-3-methylsuccinic acid 1-monomethyl ester (640 mg, 4.4 mmol, 1.1 eq). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, $R_f = 0.29$ in 1:1 hexanes/EtOAc) to yield **25** as a colorless oil (722 mg, 62% yield): ^{1}H NMR (4.0:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, $CDCl_3$): δ 7.12 - 7.30 (m, 5 H), 4.60 (m, 1 H), 4.06 (m, 1 H), 3.55* (s, 3 H), 3.54 (s, 3 H), 2.91 (m, 1 H), 2.28 (m, 4 H), 1.94 (m, 1 H), 1.24* (d, J = 7.2 Hz, 3 H), 1.15 (d, J = 7.2 Hz, 3 H), 0.93* (d, J = 7.2 Hz, 3 H), 0.74 (d, J = 7.2 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, $CDCl_3$): δ 176.4, 175.6*, 173.5*, 173.1, 141.91*, 141.90, 128.4*, 128.1, 127.7*, 127.6, 126.5, 126.2*, 77.2, 76.5*, 57.8, 55.9*, 51.7*, 51.2, 37.7, 37.5*, 36.7, 32.2*, 31.6*, 29.9, 17.5*, 16.7, 14.5*, 12.9. m/z = 294.2 $[M+H]^+$.



26. 25 (722 mg, 2.46 mmol, 1.0 eq) was dissolved in dry THF (50 mL) at room temperature. NaH (dry, 65 mg, 2.7 mmol, 1.1 eq) and tetrabutylammonium iodide (45 mg, 0.12 mmol, 0.05 eq) were added to the reaction mixture and allowed to stir until gas evolution ceased. Propargyl bromide (80% w/w in toluene, 330 µL, 3.1 mmol, 1.2 eq) was added drop wise and allowed to stir overnight.

The reaction was not complete by TLC after having been stirred overnight, so additional doses of NaH and propargyl bromide were added to the reaction mixture at room temperature and stirred overnight.

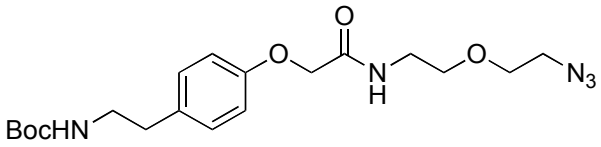
The reaction was deemed complete by TLC and was quenched by the addition of MeOH (15 mL). LiOH monohydrate (450 mg, 10.7 mmol, 4.3 eq) dissolved in 10 mL DI H_2O was added to the reaction and stirred at room temperature until deemed complete by TLC. The reaction was concentrated and the resultant residue was acidified using 5 M HCl. The residue was partitioned between DI H_2O and CH_2Cl_2, and the aqueous phase extracted with CH_2Cl_2 (2x 25 mL). The organic phase was dried over Na_2SO_4, concentrated and dried *in vacuo* to yield **26** as a colorless oil (600 mg, 77% yield): ^{1}H NMR (3.0:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.16 - 7.35 (m, 5 H), 4.76 (br. s., 1 H), 4.54 (m, 1 H), 4.09 (m, 1 H), 4.09* (s, 1 H), 3.77 (m, 1 H), 2.82 - 2.89 (m, 4 H), 2.67 (m, 1 H), 2.40 (m, 1 H), 2.34 (t, J = 2.5 Hz, 1 H), 1.38* (d, J = 7.5 Hz, 3 H), 1.18 (d, J = 7.0 Hz, 3 H), 1.03* (d, J = 7.5 Hz, 3 H), 0.75 (d, J = 7.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 177.1*, 176.4, 175.7, 174.9*, 138.1, 137.8*, 129.04, 129.98*, 128.5, 128.4*, 127.6, 127.0*, 82.9, 82.1*, 79.9, 79.2*, 75.1*, 74.6, 64.6*, 57.9, 56.4*, 56.0, 38.3, 38.2*, 32.7, 32.2*, 31.2*, 29.1, 19.3, 16.8*, 13.9*, 12.8. m/z = 318.2 $[M+H]^+$.



27. *N*-Boc-tyramine (1.0 g, 4.2 mmol, 1.0 eq), K_2CO_3 (1.71 g, 12.4 mmol, 3.0 eq) and methyl bromoacetate (500 µL, 5.3 mmol, 1.3 eq) were stirred in DMF (30 mL) overnight at room temperature. The reaction was concentrated *in vacuo* and the residue dissolved in DI H_2O (50

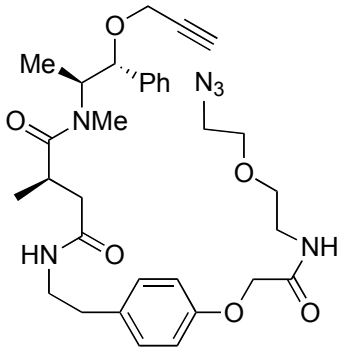
mL) and EtOAc (50 mL). The aqueous layer was extracted with EtOAc (3x 30 mL). The combined organic extracts were dried over Na_2SO_4, filtered, concentrated and dried.

The residue was dissolved in THF/MeOH (25 mL, 3:2 v/v). LiOH monohydrate (530 mg, 12.6 mg, 3.0 eq) was dissolved in DI H_2O (5 mL) and added drop wise to the solution and allowed to stir at RT until deemed complete by TLC. The reaction was concentrated and the pH of the resultant residue was adjusted to pH 5-6 using 1 M HCl. The residue was partitioned between DI H_2O and CH_2Cl_2, and the aqueous phase extracted with CH_2Cl_2 (3x 25 mL). The organic phase was dried over Na_2SO_4, concentrated and dried *in vacuo* to yield **27** as a white solid (1.24 mg, >99% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.13 (d, J = 8.4 Hz, 2 H), 6.86 (d, J = 9.0 Hz, 2 H), 4.62 (s, 2 H), 3.21 (t, J = 7.2 Hz, 2 H), 2.71 (t, J = 7.2 Hz, 2 H), 1.42 (s, 9 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 173.0, 158.6, 158.2, 133.8, 131.0, 115.8, 80.1, 66.1, 43.3, 36.4, 28.9. m/z = 294.1 $[M+H]^+$.

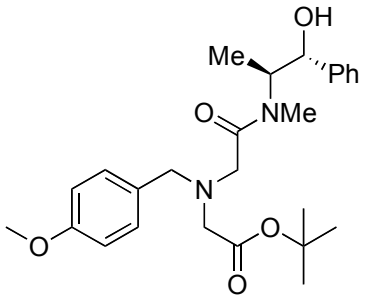


28. Prepared according General Procedure A using **9** (350.0 mg, 2.1 mmol, 1.1 eq), PyBOP (990 mg, 1.9 mmol, 1.0 eq), diisopropylethylamine (1.66 mL, 10.5 mmol, 5.0 eq) and **27** (560.0 mg, 1.9 mmol, 1.0 eq) in dry CH_2Cl_2 (40 mL). The residue was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, R_f = 0.57 in EtOAc) to yield **28** as a viscous oil (685.7 mg, 89% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.15 (d, J = 7.8 Hz, 2 H), 6.91 (d, J = 8.4 Hz, 2 H), 4.48 (s, 2 H), 3.63 (t, J = 4.2 Hz, 2 H), 3.59 (t, J = 6.6 Hz, 2 H), 3.48 (t, J = 4.8 Hz, 2 H), 3.34 (t, J = 4.2 Hz, 2 H), 3.21 (t, J = 7.2 Hz, 2 H), 2.70 (t, J = 7.2 Hz, 2 H), 1.42 (s, 9 H); ^{13}C NMR (150 MHz, CD_3OD): δ 171.5,

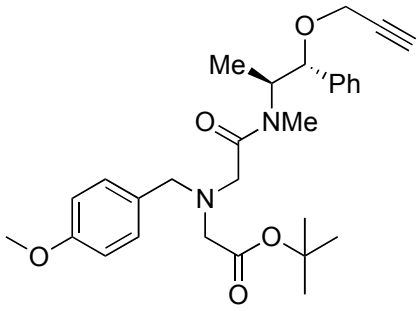
158.6, 157.8, 134.1, 131.1, 116.0, 80.1, 71.2, 70.4, 69.4, 51.9, 43.3, 40.0, 36.5, 28.9. m/z = 308.2 $[M+H, -Boc]^+$.



29. Prepared according to General Procedure C using **28** (685.7 mg, 1.68 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (3.0 mL, 12.0 mmol, 7.1 eq). The peptide coupling was carried out using Boc-deprotected **28** (238.0 mg, 0.69 mmol, 1.1 eq), PyBOP (327 mg, 0.63 mmol, 1.0 eq), diisopropylethylamine (550 µL, 3.14 mmol, 5.0 eq) and **26** (200.0 mg, 0.63 mmol, 1.0 eq) in dry CH_2Cl_2 (40 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 25-100% EtOAc in hexanes gradient, R_f = 0.29 in EtOAc) to yield **29** as a colorless oil (219.0 mg, 59% yield): ^{1}H NMR (4.1:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, CD_3OD): δ 8.31 (m, 1 H), 7.79 (m, 5 H), 7.61 (m, 2 H), 7.36 (m, 2 H), 5.34 (br. s., 1 H), 5.04 (m, 1 H), 4.95 (m, 2 H), 4.64 (m, 1 H), 4.28 (m, 1 H), 4.11 (t, *J* = 6.0 Hz, 2 H), 4.07 (t, *J* = 5.4 Hz, 2 H), 3.97 (t, *J* = 5.4 Hz, 2 H), 3.82 (t, *J* = 5.4 Hz, 2 H), 3.80 (m, 3 H), 3.46 (m, 1 H), 3.41 (s, 3 H), 3.17 (t, *J* = 7.8 Hz, 2 H), 2.59 - 2.87 (m, 1 H), 2.56 (m, 1 H), 1.87* (d, *J* = 6.0 Hz, 3 H), 1.74 (d, *J* = 6.6 Hz, 3 H), 1.44* (d, *J* = 6.0 Hz, 3 H), 1.03 (d, *J* = 6.0 Hz, 3 H); ^{13}C NMR (150 MHz, CD_3OD): δ 177.7, 174.0, 171.2, 157.7, 139.5, 133.8, 130.9, 129.3, 128.8, 128.4, 115.8, 83.9, 80.5, 76.0, 70.9, 70.3, 68.3, 56.5, 51.5, 42.1, 40.5, 39.9, 35.64, 35.62, 33.87, 33.86, 16.9, 13.8. m/z = 607.3 $[M+H]^+$.

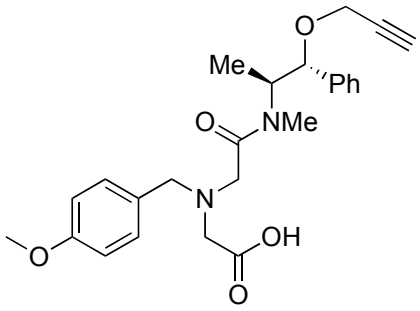


30. Prepared according General Procedure A using (1*R*,2*S*)-(-)-ephedrine (485 mg, 2.93 mmol, 1.0 eq), PyBOP (1.53 g, 2.93 mmol, 1.0 eq), diisopropylethylamine (1.53 mL, 8.8 mmol, 3.0 eq) and *N*-(4-methoxybenzyl)-iminodiacetic acid mono *t*-butyl ester (1.00 g, 3.23 mmol, 1.1 eq) in dry CH_2Cl_2 (40 mL). The crude reaction mixture was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, $R_f = 0.35$ in 1:1 hexanes/EtOAc) to yield **30** as a colorless oil (1.23 g, 91% yield): ^{1}H NMR (2.4:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, $CDCl_3$): δ 7.32 (m, 1 H), 7.25 (m, 2 H), 7.16 - 7.20 (m, 3 H), 6.83* (d, J = 9.0 Hz, 2 H), 6.79 (d, J = 7.2 Hz, 2 Hz), 4.77 (d, J = 4.2 Hz, 1 H), 4.48* (d, J = 7.8 Hz, 1 H), 4.39 (br. s., 1 H), 4.13* (br. s., 1 H), 3.76 (m, 3 H), 3.64 (m, 2 H), 3.34 (m, 2 H), 3.20 (m, 2 H), 2.72 (s, 3 H), 2.60* (s, 3 H), 1.41 (m, 9 H), 1.26 (d, J = 6.6 Hz, 3 H), 1.14* (d, J = 7.2 Hz, 3 H); ^{13}C NMR (150 MHz, $CDCl_3$): δ 171.9, 170.8, 170.5*, 170.4*, 159.2*, 159.0, 141.9, 141.8*, 130.9*, 130.5, 130.4, 129.8*, 128.4, 128.3*, 128.0*, 127.6, 126.5, 126.4*, 113.9*, 113.8, 81.2*, 81.1, 76.9, 75.9*, 58.0*, 57.7, 56.5, 55.8*, 55.4*, 55.34, 55.33*, 55.27, 32.7*, 28.7*, 28.29, 28.28*, 15.2*, 12.1. m/z = 457.3 $[M+H]^+$.

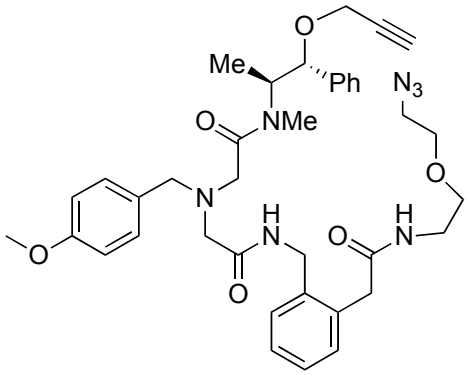


31. 30 (1.23 g, 2.7 mmol, 1.0 eq), propargyl bromide (80% w/w in toluene, 356 µL, 3.3 mmol, 1.2 eq) and tetrabutylammonium iodide (10 mg, 0.03 mmol, 0.01 eq) were dissolved in dry THF (50 mL) at 0 ˚C. NaH (dry, 71 mg, 3.0 mmol, 1.1 eq) was added to the reaction mixture and allowed to stir overnight at room temperature.

The reaction had proceeded to only about 50-60% conversion, but was quenched carefully with DI H_2O and concentrated. The resulting residue dissolved in DI H_2O (50 mL) and EtOAc (50 mL). The organic phase was dried over Na_2SO_4, concentrated and purified using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, R_f = 0.50 in 1:1 hexanes/EtOAc) to yield **31** as a colorless oil (581.2 mg, 43% yield). Residual starting material was also isolated: ^{1}H NMR (1.8:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, $CDCl_3$): δ 7.15 - 7.33 (m, 6 H), 6.97 (m, 1 H), 6.83* (d, *J* = 8.4 Hz, 2 Hz), 6.80 (d, *J* = 8.4 Hz, 2 Hz), 4.71 (br. s., 1 H), 4.63 (d, *J* = 5.4 Hz, 1 H), 4.36* (d, *J* = 7.8 Hz, 1 H), 4.09 (m, 1 H), 3.76 (m, 4 H), 3.60 (m, 2 H), 2.87 - 3.32 (m, 7 H), 2.39* (t, *J* = 2.4 Hz, 1 H), 2.30 (t, *J* = 2.4 Hz, 1 H), 1.42 (m, 9 H), 1.30* (d, *J* = 6.6 Hz, 3 H), 1.15 (d, *J* = 7.2 Hz, 3 H); ^{13}C NMR (150 MHz, $CDCl_3$): δ 171.0, 170.6*, 170.5*, 170.3, 159.1*, 158.9, 138.4, 138.3*, 130.8, 130.6*, 130.5, 130.4*, 128.7, 128.50*, 128.47, 128.2*, 127.4, 127.2*, 113.9*, 113.7, 82.8, 82.0*, 81.0*, 80.9, 79.9, 79.6*, 74.7*, 74.4, 57.9*, 57.3, 56.9*, 56.1, 56.0*, 55.8, 55.5*, 55.48, 55.45*, 55.39, 55.1, 28.4*, 28.33, 28.32, 15.2, 12.4*. m/z = 495.3 $[M+H]^+$.

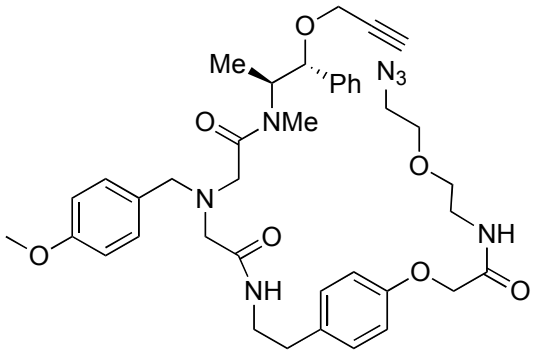


32. **31** (573 mg, 1.16 mmol, 1.0 eq) was dissolved in dry CH_2Cl_2 (5 mL). Trifluoroacetic acid (3 mL, excess) was added at room temperature and allowed to stir for 4 hours. The reaction appeared to stall and an addition dose of trifluoroacetic acid (3 mL) was added and the reaction was allowed to stir overnight. Volatiles were removed under a stream of nitrogen and the residue was allowed to dry under vacuum. **32**, isolated as a light brown oil, was determined to be pure by LC/MS and was used without further purification or characterization.



33. Prepared according to General Procedure C using **23** (685.7 mg, 1.68 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (3.0 mL, 12.0 mmol, 7.1 eq). The peptide coupling was carried out using Boc-deprotected **23** (200.0 mg, 0.64 mmol, 1.1 eq), PyBOP (307 mg, 0.59 mmol, 1.0 eq), diisopropylethylamine (511 µL, 2.93 mmol, 5.0 eq) and **32** (255.8 mg, 0.58 mmol, 1.0 eq) in dry CH_2Cl_2 (30 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 20-100% EtOAc in

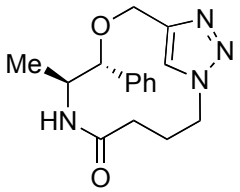
hexanes gradient then 10% MeOH in EtOAc, R_f = 0.41 in EtOAc) to yield **33** as a colorless oil (318.2 mg, 79% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.04 - 7.42 (m, 12 H), 6.94 (br. s., 1 H), 6.70 - 6.83 (m, 2 H), 4.76 (br. s., 1 H), 4.58 (m, 1 H), 4.42 (m, 2 H), 4.09 (m, 2 H), 3.68 - 3.86 (m, 4 H), 3.58 - 3.66 (m, 5 H), 3.54 (m, 3 H), 3.32 - 3.45 (m, 6 H), 3.14 (m, 2 H), 3.06 (br. s., 1 H), 2.79 (m, 2 H), 1.18 (m, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 173.8*, 173.69, 173.65, 173.59*, 172.73, 172.5* 160.7*, 160.6, 139.53, 139.52*, 138.25, 135.20*, 135.18, 131.8*, 131.7, 130.1, 130.0*, 129.73*, 129.69, 129.53,* 129.46, 128.8, 128.7*, 128.55, 128.54*, 128.52, 128.49, 128.4, 127.8, 114.9*, 114.8, 85.5, 82.3*, 80.5, 80.3*, 70.88*, 70.87, 70.35*, 70.32, 58.3, 58.2*, 56.9*, 56.8, 56.1*, 55.8*, 55.73, 55.67, 51.7, 47.38, 47.35*, 45.82*, 43.78, 41.70*, 41.6*, 40.8, 40.57*, 40.56, 37.2, 28.9*, 27.4*, 27.3, 15.6*, 13.2. m/z = 698.4 $[M+H]^+$.



34. Prepared according to General Procedure C using **28** (685.7 mg, 1.68 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (3.0 mL, 12.0 mmol, 7.1 eq). The peptide coupling was carried out using Boc-deprotected **28** (180.0 mg, 0.53 mmol, 1.1 eq), PyBOP (250 mg, 0.48 mmol, 1.0 eq), diisopropylethylamine (420 µL, 2.41 mmol, 5.0 eq) and **32** (210.0 mg, 0.63 mmol, 1.0 eq) in dry CH_2Cl_2 (40 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 25-100% EtOAc in

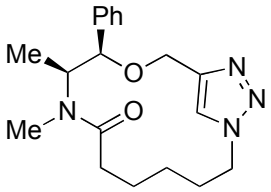
hexanes gradient then 10% MeOH in EtOAc, R_f = 0.29 in EtOAc) to yield **34** as a colorless oil (232.0 mg, 65% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.21 - 7.34 (m, 5 H), 7.11 - 7.17 (m, 3 H), 6.95 - 7.04 (m, 2 H), 6.78 - 6.88 (m, 4 H), 4.70 (br. s., 1 H), 4.61 (m, 1 H), 4.39 (s, 2 H), 4.09 (m, 1 H), 3.75 - 3.80 (m, 4 H), 3.56 - 3.63 (m, 4 H), 3.38 - 3.50 (m, 5 H), 3.33 - 3.36 (m, 4 H), 2.85 - 3.81 (m, 4 H), 2.67 - 2.80 (m, 6 H), 1.23 - 1.35 (m, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 173.7, 173.6*, 172.4*, 172.1, 160.6*, 160.5, 157.64*, 157.63, 139.6, 139.5*, 133.63, 133.58*, 131.62*, 131.55, 131.01, 130.98*, 129.70*, 129.65, 129.62*, 129.4, 128.6, 138.3*, 115.9*, 115.8, 114.9*, 114.8, 83.5, 82.8*, 80.6, 80.3*, 76.2*, 75.9, 71.0, 70.3*, 68.1, 59.6, 59.3*, 68.1, 59.6*, 59.3*, 58.73*, 58.69, 58.1, 56.9*, 56.7, 56.1*, 55.8*, 55.76, 55.70, 51.7, 41.38, 41.36*, 39.9, 35.3, 28.0, 15.6, 13.5*. m/z = 729.3 $[M+H]^+$.

Synthesis of Macrocycles.



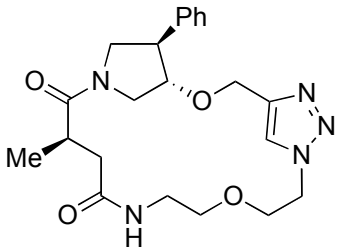
1a: Preparative Scale Flow Macrocyclization, General Procedure E. Azido-alkyne **11** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL) were aspirated from their respective source vials, mixed through a PFA mixing tube (0.2 mm inner diameter), and loaded into an injection loop. The reaction segment was injected into the flow reactor set at 150 °C, passed through the reactor at 300 µL min^{-1} (5 minute residence time). A total of 40

reaction segments prepared in this manner were collected in a round bottom flask. Upon completion, the reaction mixture was concentrated and dried *in vacuo.* The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, EtOAc, $R_f = 0.17$) to yield **1a** as a white solid (103.8 mg, 87% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 8.06 (br. s., 1 H), 7.64 (br. s., 1 H), 7.26 - 7.39 (m, 4 H), 7.19 - 7.25 (m, 1 H), 4.88 (d, $J = 13.2$ Hz, 1 H), 4.73 (br. s., 1 H), 4.39 (br. s., 1 H), 4.21 (d, $J = 13.2$ Hz, 2 H), 3.53 (d, $J = 6.6$ Hz, 1 H), 2.20 - 2.58 (m, 3 H), 1.97 (br. s., 1 H), 0.79 (d, $J = 6.6$ Hz, 3 H); ^{13}C NMR (150 MHz, CD_3OD): δ 173.3, 147.3, 141.0, 129.5, 128.4, 128.3, 127.4, 77.5, 63.3, 52.8, 52.2, 34.2, 26.1, 11.6; HRMS (ESI-TOF): $C_{16}H_{20}N_4O_2$: $[M+H]^+$: calculated 301.1659, found 301.1664.

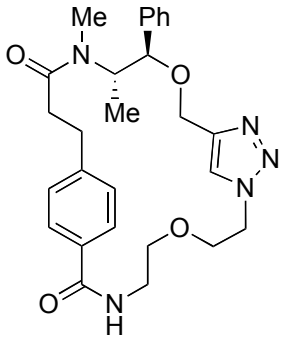


2a. Prepared according to General Procedure E using 40 reaction segments containing azido-alkyne **13** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, EtOAc, $R_f = 0.23$) to yield **2a** as a white solid (109.4 mg, 80% yield): ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.46 (m, 3 H), 7.37 (t, $J = 7.5$ Hz, 2 H), 7.25 - 7.31 (m, 1 H), 4.90 (d, $J = 13.6$ Hz, 1 H), 4.52 - 4.61 (m, 2 H), 4.52 (s, 1 H), 4.22 (d, $J = 13.6$ Hz, 2 H), 2.85 (s, 3 H), 2.22 - 2.32 (m, 1 H), 1.98 - 2.10 (m, 2 H), 1.87 - 1.96 (m, 1 H), 1.76 - 1.87 (m, 1 H), 1.52 - 1.62 (m, 1 H), 1.23 (d, $J = 6.6$ Hz, 1 H), 0.96 (m, 4 H); ^{13}C NMR (150 MHz,

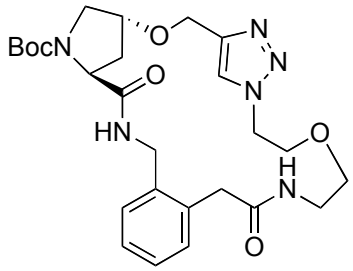
$CDCl_3$): δ 172.1, 143.9, 139.1, 128.3, 127.3, 126.5, 123.7, 80.2, 60.9, 54.3, 50.1, 31.6, 30.9, 28.5, 23.5, 23.4, 9.4; HRMS (ESI-TOF): $C_{19}H_{26}N_4O_2$: $[M+H]^+$: calculated 343.2128, found 343.2127.



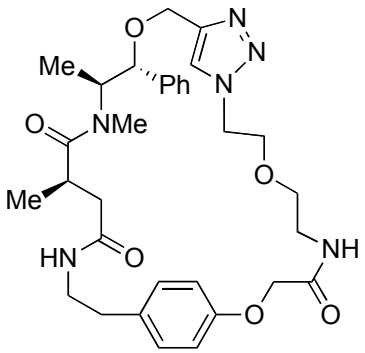
3a. Prepared according to General Procedure E using 40 reaction segments containing azido-alkyne **17** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (KP-NH cartridge, 0-10% MeOH in EtOAc, R_f = 0.44 in 10% MeOH in EtOAc) to yield **3a** as an off-white solid (96.7 mg, 57% yield): ^{1}H NMR (5.0:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, CD_3OD): δ 8.06 (s, 1 H), 8.02* (s, 1 H), 7.20 - 7.35 (m, 5 H), 4.58 (m, 3 H), 4.21 (m, 1 H), 3.76 - 3.90 (m, 3 H), 3.63 (m, 2 H), 3.37 - 3.57 (m, 6 H), 3.07 (m, 1 H), 2.96 (m, 1 H), 2.51 (m, 1 H), 2.15 (m, 1 H), 1.15 (d, J = 6.6 Hz, 3 H), 1.11* (d, J = 6.0 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 176.8, 176.2*, 173.9*, 173.8, 146.6, 144.4*, 141.4, 139.5*, 129.9, 129.8*, 128.8, 128.5*, 128.4, 128.2*, 127.5*, 126.0, 86.5, 80.0*, 71.0, 70.7*, 70.2*, 70.1, 64.2, 63.6*, 52.5, 52.0*, 51.9, 51.5*, 50.9*, 50.8*, 50.6, 49.6, 41.3*, 41.2, 40.5*, 40.4, 36.1, 35.7*, 17.2; HRMS (ESI-TOF): $C_{22}H_{29}N_5O_4$: $[M+H]^+$: calculated 428.2292, found 428.2283.



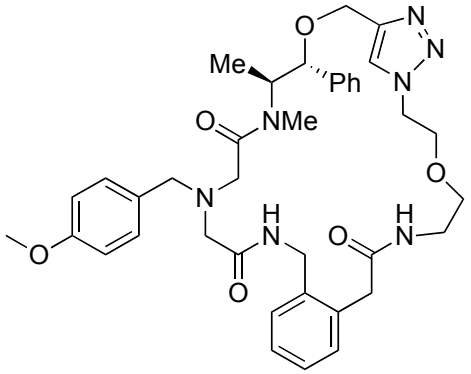
4a. Prepared according to General Procedure E using 30 reaction segments containing azido-alkyne **21** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 2% MeOH in EtOAc, $R_f = 0.14$) to yield **4a** as an off-white solid (105.2 mg, 71% yield): ^{1}H NMR (600 MHz, $CDCl_3$): δ 7.70 (d, *J*=7.9 Hz, 2 H), 7.37 (t, *J*=8.1 Hz, 4 H), 7.34 (s, 1 H), 7.30 (m, 2 H), 7.21 (m, 1 H), 7.04 - 7.10 (m, 1 H), 6.20 - 6.29 (m, 1 H), 4.58 - 4.68 (m, 2 H), 4.49 (t, *J* = 4.6 Hz, 2 H), 4.33 (d, *J* = 11.8 Hz, 1 H), 4.10 (d, *J* = 11.4 Hz, 1 H), 3.81 - 3.91 (m, 2 H), 3.61 - 3.67 (m, 1 H), 3.56 - 3.61 (m, 2 H), 3.50 - 3.55 (m, 1 H), 3.27 - 3.38 (m, 1 H), 3.02 - 3.10 (m, 1 H), 2.96 (m, 1 H), 2.68 (s, 1 H), 2.59 (s, 3 H), 2.53 - 2.60 (m, 1 H), 0.91 (d, *J*=7.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, $CDCl_3$): δ 172.9, 172.7*, 167.4*, 166.9, 145.6, 144.6, 139.2, 132.6*, 132.1, 129.4*, 129.1, 128.4*, 128.3, 127.3*, 127.27, 127.22, 126.4*, 126.2, 123.3, 85.2*, 84.9, 70.0*, 69.4, 69.3*, 68.5, 63.6*, 63.4, 54.6, 50.4*, 50.1, 39.7*, 39.4, 34.5, 32.7, 32.2*, 31.9, 29.7*, 9.7; HRMS (ESI-TOF): $C_{27}H_{33}N_5O_4$: $[M+H]^+$: calculated 492.2605, found 492.2609.



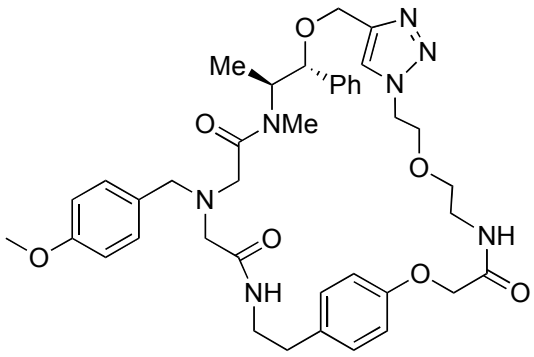
5a. Prepared according to General Procedure E using 30 reaction segments containing azido-alkyne **24** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (KP-NH cartridge, 0-10% MeOH in EtOAc, R_f = 0.41 in 10% MeOH in EtOAc) to yield **5a** as an off-white solid (99.7 mg, 63% yield): ^{1}H NMR (1.6:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, CD_3OD): δ 7.99 (s, 1 H), 7.90* (s, 1 H), 7.17 - 7.30 (m, 4 H), 4.44 - 4.67 (m, 5 H), 4.12 - 4.30 (m, 4 H), 3.85 (m, 2 H), 3.70 (m, 1 H), 3.39 - 3.62 (m, 5 H), 3.21 (m, 2 H), 2.01 (m, 2 H), 1.40* (s, 9 H), 1.29 (s, 9 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 174.3, 174.1*, 173.92*, 173.90, 156.1*, 155.6, 145.5*, 145.4, 137.9, 137.8*, 136.3, 136.1*, 131.8, 131.7, 131.62*, 131.58*, 129.4, 129.2*, 128.61, 128.59*, 126.1, 125.9, 81.6, 81.4*, 75.9*, 75.6, 70.4, 70.2*, 70.1, 63.2, 62.9*, 60.8, 60.4*, 52.7*, 52.5, 51.5, 51.4*, 42.94*, 42.88, 40.8, 40.6, 40.58*, 40.55*, 37.5, 36.6*, 28.6, 26.5 ; HRMS (ESI-TOF): $C_{26}H_{36}N_6O_6$: $[M+H]^+$: calculated 529.2769, found 529.2774.



6a. Prepared according to General Procedure E using 25 reaction segments containing azido-alkyne **29** (0.10 M in EtOH, 100 μL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 μL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 μL, 0.020 mmol, 2.0 eq) and EtOH (200 μL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (KP-NH cartridge, 0-10% MeOH in EtOAc, R_f = 0.33 in 10% MeOH in EtOAc) to yield **6a** as an off-white solid (94.1 mg, 62% yield): ^{1}H NMR (2.6:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, CD_3OD): δ 7.96 (s, 1 H), 7.84* (s, 1 H), 7.62 (br. s., 1 H), 7.17 - 7.39 (m, 7 H), 6.91 (m, 2 H), 4.69 (br. s., 1 H), 4.26 - 4.61 (m, 8 H), 4.00* (m, 1 H), 3.85 (m, 1 H), 3.74* (m, 1 H), 3.55 (m, 4 H), 3.39 (m, 1 H), 3.25 (m, 2 H), 2.62 - 2.86 (m, 6 H), 2.34 (m, 1 H), 1.98 (m, 1 H), 1.22* (d, J = 6.6 Hz, 3 H), 1.01 (d, J = 7.2 Hz, 3 H), 0.96* (d, J = 7.2 Hz, 3 H), 0.41 (d, J = 7.2 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 177.6*, 177.0, 173.9, 173.8*, 171.3, 171.2*, 157.6*, 157.5, 145.9, 145.6*, 140.4*, 140.1*, 134.0*, 133.9, 131.3, 131.2*, 128.6*, 129.3, 129.2*, 128.9, 126.0, 125.8*, 115.9*, 115.7, 85.2, 70.8, 70.2*, 70.1, 69.8*, 68.2*, 68.1, 63.3*, 62.8, 58.6, 51.6, 51.4*, 41.4*, 41.3, 41.0, 40.9, 40.0, 39.8*, 35.4*, 35.0, 34.2*, 34.1, 18.1*, 17.0, 14.4, 13.3*; HRMS (ESI-TOF): $C_{32}H_{42}N_6O_6$: $[M+H]^+$: calculated 607.3238, found 607.3243.

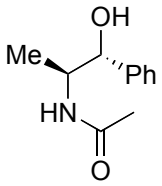


7a. Prepared according to General Procedure E using 30 reaction segments containing azido-alkyne **33** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (KP-NH cartridge, 0-10% MeOH in EtOAc, R_f = 0.33 in 10% MeOH in EtOAc) to yield **7a** as an off-white solid (92.1 mg, 44% yield): ^{1}H NMR (1.5:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, CD_3OD): δ 7.96* (s, 1 H), 7.89 (s, 1 H), 7.14 - 7.42 (m, 9 H), 7.04 (d, *J* = 9.0 Hz, 2 H), 6.99* (d, *J* = 6.0 Hz, 2 H), 6.84* (d, *J* = 9.6 Hz, 2 H), 6.72 (d, *J* = 9.0 Hz, 2 H), 4.42 - 4.61 (m, 6 H), 4.34 (m, 1 H), 3.57 - 3.87 (m, 8 H), 3.34 - 3.55 (m, 5 H), 3.23 (m, 1 H), 3.14 (m, 3 H), 2.88 - 3.09 (m, 1 H), 2.63 - 2.67 (m, 3H), 1.12* (d, *J* = 7.2 Hz, 3 H), 1.03 (d, *J* = 7.2 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 173.5, 173.4, 173.3*, 172.9*, 172.6*, 172.2, 160.8*, 160.5, 146.2*, 145.3, 140.1, 139.8*, 139.0*, 138.7, 135.7, 135.6*, 132.1, 131.84, 131.81*, 131.6*, 131.14*, 131.07, 130.8*, 130.6, 129.5*, 129.4, 129.0, 128.98, 128.93*, 128.69, 128.65*, 128.29, 128.24*, 126.2, 84.5, 83.4*, 70.4, 70.27*, 70.25, 63.3*, 62.3, 60.7*, 60.1, 59.1, 58.8*, 58.3, 57.6*, 57.0*, 55.8*, 55.6, 51.5*, 51.4, 47.4, 47.3*, 41.7, 41.5*, 41.1, 40.9*, 40.7, 40.3*, 29.0, 27.4, 27.3*, 15.8, 12.3*; HRMS (ESI-TOF): $C_{38}H_{47}N_7O_6$: $[M+H]^+$: calculated 698.3660, found 698.3625.

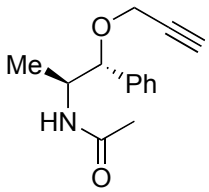


8a. Prepared according to General Procedure E using 20 reaction segments containing azido-alkyne **34** (0.10 M in EtOH, 100 µL, 0.010 mmol, 1.0 eq), TTTA (0.01 M in EtOH, 100 µL, 0.001 mmol, 0.10 eq), DIPEA (0.1 M in EtOH, 200 µL, 0.020 mmol, 2.0 eq) and EtOH (200 µL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (KP-NH cartridge, 0-10% MeOH in EtOAc, R_f = 0.33 in 10% MeOH in EtOAc) to yield **8a** as an off-white solid (86.0 mg, 59% yield): ^{1}H NMR (1.1:1 rotamer ratio, asterisks denote minor rotamer peaks, 600 MHz, CD_3OD): δ 7.97* (s, 1 H), 7.91 (s, 1 H), 7.16 - 7.30 (m, 6 H), 7.06* (d, *J* = 9.0 Hz, 2 H), 6.89 - 6.99 (m, 5 H), 6.79 - 6.87 (m, 2 H), 4.24 - 4.67 (m, 8 H), 3.89 (m, 1 H), 3.81 (m, 1 H), 3.35 - 3.69 (m, 8 H), 3.11 (m, 1 H), 2.70 - 3.00 (m, 5 H), 2.68 (s, 3 H), 2.58* (s, 3H), 1.18 (d, *J* = 7.2 Hz, 3 H), 1.11* (d, *J* = 6.6 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 174.1*, 173.2, 172.0*, 171.9, 171.0*, 170.8, 160.8*, 160.6, 157.7, 145.8, 145.7*, 140.2*, 140.1, 133.9, 133.4*, 131.7*, 131.6, 131.2, 131.1*, 130.8, 129.54*, 129.51, 129.5, 129.3*, 128.5, 128.2*, 125.93, 125.87*, 84.6*, 83.9, 70.8*, 70.6, 70.1, 68.2, 67.9*, 63.2*, 63.0, 59.8, 59.7*, 58.8, 58.24*, 58.18, 56.48, 55.8, 55.7*, 54.8, 51.7*, 51.5, 40.7, 40.5*, 40.1*, 40.0, 34.6, 34.5*, 28.5, 16.0, 13.2*; HRMS (ESI-TOF): $C_{39}H_{49}N_7O_7$: $[M+H]^+$: calculated 728.3766, found 728.3741.

Synthesis of Acyclic Analog Precursors.

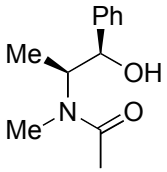


35. (1*R*,2*S*)-(-)-norephedrine (920 mg, 6.1 mmol, 1.0 eq) and Ac_2O (2 mL, 21.2 mmol, 3.5 eq) were heated to 80 ˚C for one hour. The reaction was quenched by the addition of DI H_2O (4 mL) and then concentrated. The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 40-100% EtOAc in hexanes gradient, R_f = 0.29 in EtOAc) to yield **35** as colorless oil (746 mg, 64% yield): ^{1}H NMR (4.8:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.17 - 7.30 (m, 5 H), 6.07 (d, *J* = 7.5 Hz , 1 H), 5.79* (d, *J* = 4.0 Hz , 1 H), 4.79 (d, *J* = 2.5 Hz , 1 H), 4.22 (m, 1 H), 1.91 (s, 3 H), 1.87* (s, 3 H), 1.02* (d, *J* = 7.0 Hz, 3 H), 0.94 (d, *J* = 6.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 171.0, 169.9*, 141.1, 137.0*, 128.6*, 128.28, 128.25*, 127.6, 126.6*, 126.4, 77.6*, 76.4, 51.2, 48.8*, 23.4, 21.4*, 15.3*, 14.3. m/z = 190.0 $[M+H]^+$.

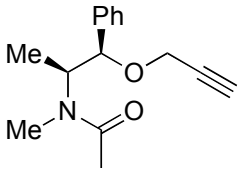


36. Prepared according General Procedure B using **35** (746 mg, 3.86 mmol, 1.0 eq), NaH (dry, 110 mg, 4.6 mmol, 1.2 eq), propargyl bromide (80% w/w in toluene, 620 µL, 5.8 mmol, 1.5 eq) in dry THF (50 mL). The crude reaction mixture was purified using a Biotage Horizon

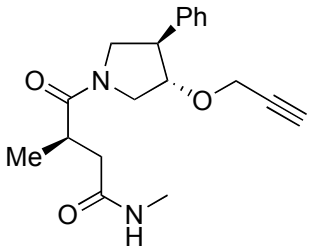
automated flash column chromatography system (silica gel, 15-100% EtOAc in hexanes gradient, R_f = 0.24 in 1:1 hexanes/EtOAc) to yield **36** as a yellow solid (752 mg, 84% yield): ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.23 - 7.34 (m, 5 H), 5.94 (br. s., 1 H), 4.69 (d, *J* = 3.5 Hz , 1 H), 4.25 (dd, *J* = 16, 2.5 Hz , 1 H), 4.16 (m, 1 H), 3.90 (dd, *J* = 16, 2.5 Hz , 1 H), 2.41 (t, *J* = 3.0 Hz, 1 H), 1.96 (s, 3 H), 0.94 (d, *J* = 6.5 Hz, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$): δ 169.4, 138.0, 128.6, 128.0, 126.9, 82.2, 80.0, 74.6, 56.8, 49.9, 23.7, 13.6. m/z = 232.0 $[M+H]^+$.



37. (1*R*,2*S*)-(-)-ephedrine (1.0 g, 6.1 mmol, 1.0 eq) and Ac_2O (2 mL, 21.2 mmol, 3.5 eq) were heated to 80 ˚C for one hour. The reaction was quenched by the addition of DI H_2O (4 mL) and then concentrated. The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 25-100% EtOAc in hexanes gradient, R_f = 0.39 in EtOAc) to yield **37** as colorless oil (860 mg, 69% yield): ^{1}H NMR (2.8:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.19 - 7.34 (m, 5 H), 4.79 (s, 1 H), 4.56* (m, 1 H), 4.42 (m, 1 H), 4.31 (br. s., 1 H), 3.87* (m, 1 H), 3.34* (br. s., 1 H), 2.75* (s, 3 H), 2.68 (s, 3 H), 1.98 (s, 3 H), 1.65* (s, 3 H), 1.32* (d, *J* = 7.0 Hz, 3 H), 1.15 (d, *J* = 7.2 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 172.6, 170.9*, 142.4*, 142.1, 128.6*, 128.3, 128.2*, 127.6, 126.4, 126.2*, 75.9, 59.5, 58.0*, 34.1, 28.2*, 22.7, 21.4*, 14.9*, 12.1. m/z = 208.0 $[M+H]^+$.

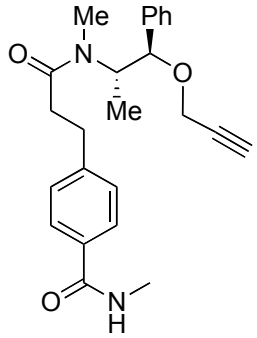


38. Prepared according General Procedure B using **37** (860 mg, 4.15 mmol, 1.0 eq), NaH (dry, 120 mg, 5.0 mmol, 1.2 eq), propargyl bromide (80% w/w in toluene, 660 µL, 6.1 mmol, 1.5 eq) in dry THF (50 mL). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 15-100% EtOAc in hexanes gradient, R_f = 0.29 in 1:1 hexanes/EtOAc) to yield **38** as a colorless oil (945 mg, 93% yield): ^{1}H NMR (1.8:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.22 - 7.32 (m, 5 H), 4.67 (m, 2 H), 4.46* (d, *J* = 8.5 Hz, 1 H), 4.16 (m, 1 H), 3.93* (m, 1 H), 3.80 (m, 1 H), 2.87 (s, 3 H), 2.77* (s, 3 H), 2.40* (t, *J* = 2.5 Hz, 1 H), 2.35* (t, *J* = 2.5 Hz, 1 H), 1.93 (s, 3 H), 1.70* (s, 3 H), 1.35* (d, *J* = 6.5 Hz, 3 H), 1.15 (d, *J* = 6.5 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 170.8, 170.6*, 138.7, 138.2*, 128.9, 128.8*, 128.4, 128.0*, 127.2, 83.1, 82.2*, 80.0, 79.4*, 74.9*, 74.3, 58.6, 56.3, 56.2*, 53.8*, 32.6*, 28.2, 22.6, 21.5*, 15.3*, 12.0. m/z = 246.0 $[M+H]^+$.



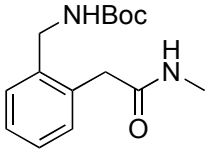
39. Prepared according General Procedure A using methylamine hydrochloride (27 mg, 0.40 mmol, 1.1 eq), PyBOP (187 mg, 0.36 mmol, 1.0 eq), diisopropylethylamine (313 µL, 1.8 mmol, 5.0 eq) and **16** (113.5 mg, 0.36 mmol, 1.0 eq) in dry CH_2Cl_2 (20 mL). The crude reaction

mixture was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 20-100% EtOAc in hexanes gradient, R_f = 0.19 in EtOAc) to yield **39** as a colorless oil (111.2 mg, 94% yield): ^{1}H NMR (1.3:1 rotamer ratio, asterisks denote minor rotamer peaks, 500 MHz, $CDCl_3$): δ 7.12 - 7.32 (m, 5 H), 6.10 - 6.20 (m, 1 H), 4.26* (q, *J* = 5.0 Hz, 1 H), 4.19 (q, *J* = 5.0 Hz, 1 H), 4.11 - 4.16 (m, 2 H), 3.65 - 3.97 (m, 4 H), 3.39 - 3.54 (m, 1 H), 3.04 - 3.20 (m, 1 H), 2.74 (t, *J* = 4.5 Hz, 3 H), 2.62 (m, 1 H), 2.43* (t, *J* = 2.0 Hz, 1 H), 2.41 (t, *J* = 2.5 Hz, 1 H), 2.26 (t, *J* = 4.5 Hz, 1 H), 2.23* (t, *J* = 5.0 Hz, 1 H), 1.15 (d, *J* = 7.0 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 125 MHz, $CDCl_3$): δ 175.0, 174.9*, 172.29, 172.28*, 139.6, 139.2*, 129.11, 129.09*, 127.5*, 127.44*, 127.38, 127.3, 83.3*, 81.9, 79.4, 75.3*, 75.2, 57.18*, 57.15, 50.3, 49.9*, 49.7, 49.4*, 47.8, 40.42, 40.35*, 35.2*, 34.9, 26.43*, 26.40, 17.7, 17.5*. m/z = 329.2 $[M+H]^+$.

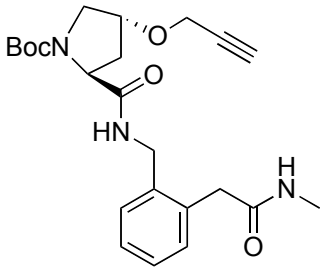


40. Prepared according General Procedure A using methylamine hydrochloride (55 mg, 0.81 mmol, 1.1 eq), PyBOP (385 mg, 0.74 mmol, 1.0 eq), diisopropylethylamine (640 µL, 3.7 mmol, 5.0 eq) and **20** (282 mg, 0.74 mmol, 1.0 eq) in dry CH_2Cl_2 (30 mL). The crude reaction mixture was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, EtOAc, R_f = 0.40 in EtOAc) to yield **40** as a light yellow oil (290.2 mg, >99% yield): ^{1}H NMR (2.0:1 rotamer ratio, asterisks denote minor rotamer

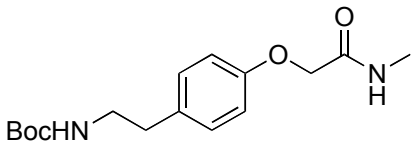
peaks, 600 MHz, $CDCl_3$): δ 7.63 (m, 2 H), 7.03 - 7.33 (m, 7 H), 6.20 (br. s., 1 H), 4.69 (br. s., 1 H), 4.65 (m, 1 H), 4.43* (d, *J* = 7.8 Hz, 1 H), 4.13 (m, 1 H), 3.95* (m, 1 H), 3.81 (m, 1 H), 2.97 (m, 3 H), 2.79 - 2.90 (m, 5 H), 2.30 - 2.51 (m, 3 H), 1.29* (d, *J* = 7.2 Hz, 2 H), 1.15 (d, *J* = 6.6 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, $CDCl_3$): δ 172.0, 171.9*, 168.3, 145.5, 145.4*, 138.6, 138.2*, 132.6*, 132.5, 128.92, 128.87*, 128.8, 128.7*, 128.5, 128.2*, 127.3, 127.2, 127.12*, 127.10*, 83.1, 82.0*, 80.0, 79.5*, 74.9*, 74.4, 64.6*, 57.6*, 56.32, 56.27, 35.6, 34.3*, 31.1, 31.0, 30.8*, 28.5*, 27.0, 15.6, 13.9*. m/z = 393.2 $[M+H]^+$.



41. Prepared according General Procedure A using methylamine hydrochloride (140.0 mg, 2.1 mmol, 1.1 eq), PyBOP (990 mg, 1.9 mmol, 1.0 eq), diisopropylethylamine (1.66 mL, 10.5 mmol, 5.0 eq) and N-Boc-2-aminomethyl-phenylacetic acid (500.0 mg, 1.9 mmol, 1.0 eq) in dry CH_2Cl_2 (50 mL). The residue was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 15-100% EtOAc in hexanes gradient, R_f = 0.43 in EtOAc) to yield **41** as a white solid (220.2 mg, 44% yield): ^1H NMR (600 MHz, $CDCl_3$): δ 7.21 - 7.32 (m, 4 H), 6.05 (br. s., 1 H), 5.25 (br. s., 1 H), 4.28 (m, 2 H), 3.56 (s, 3 H), 2.74 (d, *J* = 5.4 Hz, 3 H), 1.42 (s, 9 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, $CDCl_3$): δ 171.5, 156.2, 137.5, 133.7, 131.2, 129.7, 128.3, 128.0, 79.9, 47.3, 40.8, 28.6, 26.7. m/z = 179.1 [M+H, -Boc]$^+$.

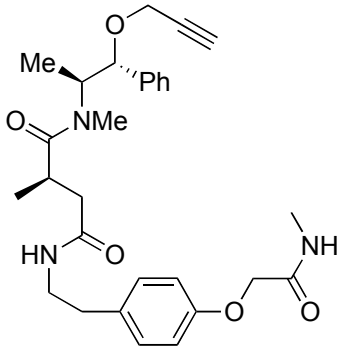


42. Prepared according to General Procedure C using **41** (220.2 mg, 0.79 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (2.0 mL, 8.0 mmol, 10.1 eq). The peptide coupling was carried out using Boc-deprotected **41** (75.0 mg, 0.28 mmol, 1.0 eq), PyBOP (145.7 mg, 0.28 mmol, 1.0 eq), diisopropylethylamine (244 µL, 1.4 mmol, 5.0 eq) and **22** (117.0 mg, 0.44 mmol, 1.0 eq) in dry CH_2Cl_2 (10 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, R_f = 0.32 in EtOAc) to yield **42** as a light yellow oil (120.2 mg, >99% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 8.71 (m, 1H), 7.89 (m, 1 H), 7.24 - 7.34 (m, 4 H), 4.35 - 4.55 (m, 2 H), 4.32 (br. s., 1 H), 4.23 (m, 1 H), 4.12 (d, *J* = 1.8 Hz, 2 H), 3.63 (m, 4 H), 2.88 (s, 1 H), 2.74 (m, 3 H), 2.39 (m, 1 H), 2.02 (m, 1 H), 1.29 - 1.46 (m, 9 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 175.1, 174.9*, 174.3, 174.2*, 156.4*, 156.0, 138.2, 137.8*, 135.6, 135.4*, 131.6, 130.71, 130.71*, 130.3, 129.0, 128.8*, 128.6, 128.5*, 81.7, 81.5*, 80.51*, 80.49, 78.0*, 77.2, 76.08, 76.05*, 60.9, 60.8*, 60.53, 40.49*, 57.0*, 56.9, 55.5*, 53.0, 42.5, 42.4*, 42.2, 42.1*, 37.9, 37.0*, 28.7*, 28.4, 26.9, 26.6*. m/z = 430.2 $[M+H]^+$.



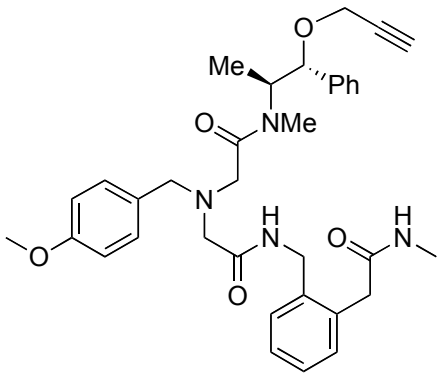
43. Prepared according General Procedure A using methylamine hydrochloride (70.0 mg, 1.0 mmol, 1.1 eq), PyBOP (495 mg, 0.95 mmol, 1.0 eq), diisopropylethylamine (1.66 mL, 10.5

mmol, 5.0 eq) and **27** (280.0 mg, 0.95 mmol, 1.0 eq) in dry CH_2Cl_2 (30 mL). The residue was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 10-100% EtOAc in hexanes gradient, R_f = 0.49 in EtOAc) to yield **43** as a white solid (286.0 mg, 97% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.15 (d, *J* = 9.0 Hz, 2 H), 6.91 (d, *J* = 9.0 Hz, 2 H), 4.46 (s, 2 H), 3.20 (t, *J* = 7.9 Hz, 2 H), 2.81 (s, 3 H), 2.69 (t, *J* = 7.8 Hz, 2 H), 1.42 (s, 9 H); ^{13}C NMR (150 MHz, CD_3OD): δ 171.9, 158.6, 157.8, 134.1, 131.1, 115.9, 80.1, 68.3, 43.4, 36.5, 28.9, 26.2. m/z = 209.1 $[M+H]^+$.



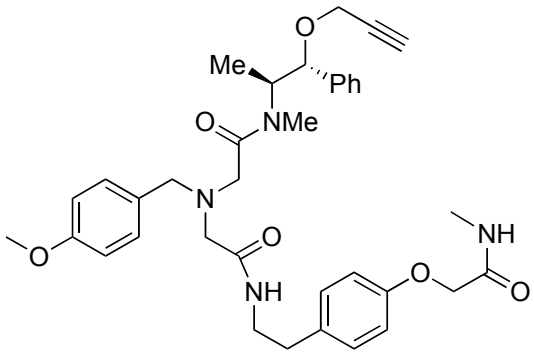
44. Prepared according to General Procedure C using **43** (286.0 mg, 0.93 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (2.0 mL, 8.0 mmol, 8.6 eq). The peptide coupling was carried out using Boc-deprotected **43** (100.0 mg, 0.41 mmol, 1.2 eq), PyBOP (176 mg, 0.34 mmol, 1.0 eq), diisopropylethylamine (300 µL, 1.72 mmol, 5.0 eq) and **26** (108.0 mg, 0.34 mmol, 1.0 eq) in dry CH_2Cl_2 (40 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 25-100% EtOAc in hexanes gradient, R_f = 0.49 in EtOAc) to yield **44** as a colorless oil (149.5 mg, 87% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 8.09 (br. s., 1 H), 7.86 (br. s., 1 H), 7.22 - 7.37 (m, 5 H), 7.13 (d, *J*

= 8.4 Hz, 2 H), 6.90 (d, *J* = 7.8 Hz, 2 H), 4.82 (br. s., 1 H), 4.56 (d, *J* = 2.8 Hz, 1 H), 4.45 (s, 2 H), 4.09 (m, 1 H), 3.72 (dd, *J* = 15.6, 2.4 Hz, 1 H), 2.86 (m, 1 H), 2.92 (s, 3 H), 2.84 (t, *J* = 2.4 Hz, 1 H), 2.81 (m, 5 H), 2.68 (t, *J* = 7.2 Hz, 2 H), 2.39 (m, 1 H), 2.07 (m, 1 H), 1.25 (d, *J* = 7.2 Hz, 3 H), 0.53 (d, *J* = 6.0 Hz, 3 H); ^{13}C NMR (150 MHz, CD_3OD): δ 177.7, 174.1, 171.8, 157.7, 139.5, 133.8, 130.9, 129.4, 129.3, 128.8, 115.8, 83.9, 80.5, 76.0, 68.3, 56.6, 55.8, 42.2, 40.6, 33.9, 26.2, 26.0, 16.9, 13.8, 13.2; m/z = 508.3 $[M+H]^+$.



45. Prepared according to General Procedure C using **41** (220.2 mg, 0.79 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (2.0 mL, 8.0 mmol, 10.1 eq). The peptide coupling was carried out using Boc-deprotected **41** (45.0 mg, 0.21 mmol, 1.1 eq), PyBOP (105 mg, 0.20 mmol, 1.0 eq), diisopropylethylamine (175 µL, 1.0 mmol, 5.0 eq) and **32** (117.0 mg, 0.44 mmol, 1.0 eq) in dry CH_2Cl_2 (10 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 5% MeOH in EtOAc, R_f = 0.43 in EtOAc) to yield **45** as a light yellow oil (88.6 mg, 74% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.07 - 7.38 (m, 11 H), 6.94 (br. s., 1 H), 6.78 (m, 2 H), 4.75 (br. s., 1 H), 4.60 (d, *J* = 7.8 Hz, 1 H), 4.46 (m, 2 H), 4.06 (m, 1 H), 3.76 - 3.83 (m, 4 H), 3.58 - 3.64 (m, 2 H), 3.40 - 3.54 (m, 3 H), 3.11 - 3.26 (m, 2 H), 2.99 - 3.10 (m, 2 H), 2.79 (m, 4 H), 2.71 (m, 3 H), 1.27 (m, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 174.17,

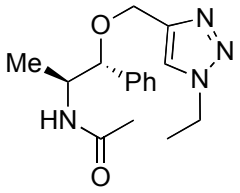
174.14, 173.1, 160.77*, 160.75, 139.52, 139.51*, 138.17*, 138.16, 135.20*, 135.19, 132.1, 131.9*, 131.7, 130.2*, 130.1, 129.8*, 129.7, 129.6*, 129.5, 128.92, 128.89*, 128.7, 128.59, 128.57*, 128.4, 127.8*, 114.9*, 114.8, 83.5, 82.8*, 80.5, 80.2*, 76.2*, 76.0, 65.4, 59.7*, 58.2, 58.1*, 56.9*, 56.7, 56.2*, 55.8*, 55.7, 41.7, 41.6*, 40.7, 26.64, 26.63*, 20.86, 20.80*, 20.15, 15.6*, 14.0, 13.2*; m/z = 599.3 $[M+H]^+$.



46. Prepared according to General Procedure C using **43** (286.0 mg, 0.93 mmol, 1.0 eq) and 4.0 M HCl in 1,4-dioxane (2.0 mL, 8.0 mmol, 8.6 eq). The peptide coupling was carried out using Boc-deprotected **43** (54.0 mg, 0.22 mmol, 1.1 eq), PyBOP (105 mg, 0.20 mmol, 1.0 eq), diisopropylethylamine (175 µL, 1.0 mmol, 5.0 eq) and **32** (88.0 mg, 0.20 mmol, 1.0 eq) in dry CH_2Cl_2 (10 mL). The residue was purified (without filtering off PyBOP impurities) using a Biotage Horizon automated flash column chromatography system (silica gel, 5% MeOH in EtOAc, R_f = 0.43 in EtOAc) to yield **46** as a light yellow oil (101.8 mg, 81% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.06 - 7.32 (m, 8 H), 6.80 - 6.90 (m, 5 H), 4.76 (br. s., 1 H), 4.67 (d, *J* = 7.2 Hz, 1 H), 4.36 (s, 2 H), 4.09 (dd, *J* = 18.6, 2.4 Hz, 1 H), 3.76 - 3.83 (m, 4 H), 3.33 - 3.50 (m, 4 H), 3.14 (m, 2 H), 2.92 - 3.05 (m, 2 H), 2.73 - 2.87 (m, 9 H), 1.26 (d, *J* = 6.0 Hz, 3 H); ^{13}C NMR (asterisks denote minor rotamer peaks, 150 MHz, CD_3OD): δ 173.2, 172.1, 171.5, 160.8, 157.7, 139.5, 139.4*, 133.51, 133.50*, 132.2, 131.9*, 131.01, 130.98*, 129.74*, 129.69, 129.6*,

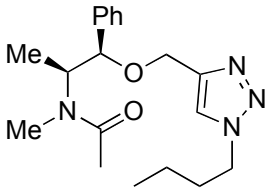
129.5, 128.7, 128.3, 127.3*, 126.6*, 115.88*, 115.87, 115.0*, 114.9, 83.4, 82.7*, 80.5, 80.2*, 76.2*, 76.0, 68.1, 59.5, 58.2, 56.9*, 56.7, 55.8, 55.77, 55.72, 55.70*, 47.36, 47.33*, 41.6, 41.5*, 27.36*, 27.31, 26.01, 15.6*, 13.2; m/z = 629.4 $[M+H]^+$.

Synthesis of Acyclic Analogs.

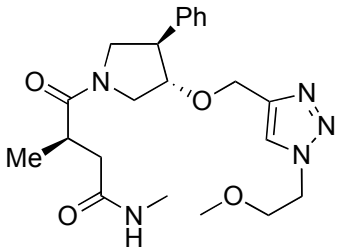


1b: Preparative Scale Flow Intermolecular Click Reaction, General Procedure F. Alkyne **36** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), iodoethane (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq) were aspirated from their respective source vials, mixed through a PFA mixing tube (0.2 mm inner diameter), and loaded into an injection loop. The reaction segment was injected into the flow reactor set at 150 °C, passed through the reactor at 150 µL min^{-1} (10 minute residence time). A total of 10 reaction segments prepared in this manner were collected in a round bottom flask. Upon completion, the reaction mixture was concentrated and dried *in vacuo*. The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 5% MeOH in CH_2Cl_2, R_f = 0.17) followed by PTLC (silica gel, 500 µm plates, 5% MeOH in CH_2Cl_2) to yield **1b** as an off-white solid (78.5 mg, 69% yield): ^1H NMR (600 MHz, $CDCl_3$): δ 7.44 (s, 1 H), 7.16 - 7.35 (m, 5

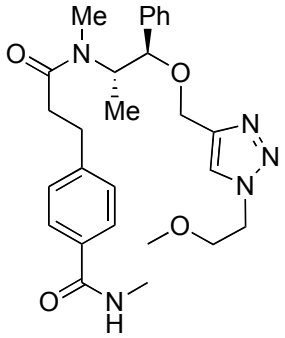
H), 6.56 (d, *J* = 7.2 Hz, 1 H), 4.73 (d, *J* = 13.8 Hz, 1 H), 4.39 (d, *J* = 2.4 Hz, 1 H), 4.33 - 4.43 (m, 3 H), 4.15 (m, 1 H), 1.91 (s, 3 H), 1.52 (t, *J* = 7.2 Hz, 3 H), 0.91 (d, *J* = 6.6 Hz, 3 H); ^{13}C NMR (150 MHz, $CDCl_3$): δ 169.7, 144.9, 138.7, 128.6, 127.8, 126.7, 121.9, 82.8, 62.7, 50.2, 45.5, 23.5, 15.6, 13.5; HRMS (ESI-TOF): $C_{16}H_{22}N_4O_2$: $[M+H]^+$: calculated 303.1815, found 303.1823.



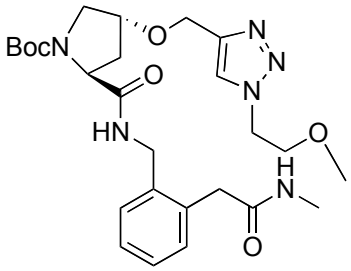
2b. Prepared according to General Procedure F using 15 reaction segments containing alkyne **38** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), iodobutane (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 5% MeOH in CH_2Cl_2, R_f = 0.27) followed by PTLC (silica gel, 500 µm plates, 5% MeOH in CH_2Cl_2) to yield **2b** as yellow oil (127.1 mg, 66% yield): ^{1}H NMR (1.7:1 rotamer ratio, asterisk denotes minor rotamer peak, 600 MHz, $CDCl_3$): δ 7.40* (s, 1 H), 7.33 (s, 1H), 7.16 - 7.33 (m, 5 H), 4.61 (m, 1 H), 4.55 (m, 2 H), 4.35 (m, 1 H), 4.23 - 4.30 (m, 2 H), 3.89* (m, 1 H), 2.75 (s, 3 H), 2.69* (s, 3 H), 1.83 (s, 3 H), 1.80 (m, 2 H), 1.65* (s, 3 H), 1.28 (m, 2 H), 1.26* (d, *J* = 6.6 Hz, 3 H), 1.08 (d, *J* = 7.8 Hz, 3 H), 0.88 (t, *J* = 7.2 Hz, 3 H); ^{13}C NMR (asterisk denotes minor rotamer peak, 150 MHz, $CDCl_3$): δ 170.6, 170,5*, 145.0, 144.7*, 139.2, 138.8*, 128.7, 128.5*, 128.3, 127.8*, 127.1*, 122.3, 83.8, 83.3*, 62.7*, 62.6, 58.6, 50.1*, 50.0, 32.29, 32.38*, 28.1, 22.4, 21.4*, 19.72, 19.72*, 15.2, 13.5, 12.2*; HRMS (ESI-TOF): $C_{19}H_{28}N_4O_2$: $[M+H]^+$: calculated 345.2285, found 345.2280.



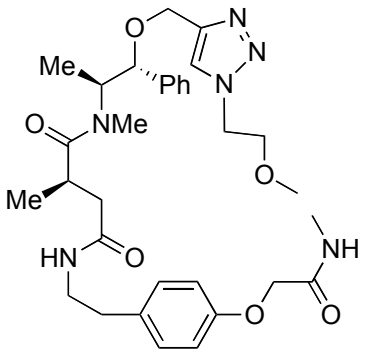
3b. Prepared according to General Procedure F using 7 reaction segments containing alkyne **39** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 0-10% MeOH in EtOAc gradient, R_f = 0.60 in 5% MeOH in EtOAc) followed by PTLC (silica gel, 500 µm plates, 7.5% MeOH in CH_2Cl_2) to yield **3b** as colorless oil (60 mg, 53% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.87 (m, 1 H), 7.19 - 7.38 (m, 5H), 4.65 (m, 2 H), 4.54 (m, 2 H), 4.15 - 4.30 (m, 1 H), 3.85 - 4.07 (m, 2 H), 3.07 - 3.77 (m, 3 H), 3.60 (m, 1 H), 3.44 (m, 1 H), 3.30 (m, 3 H), 3.09 - 3.21 (m, 1 H), 2.70 (m, 3 H), 2.60 (m, 1 H), 2.26 (m, 1 H), 1.13 (m, 3 H); ^{13}C NMR (asterisk denotes minor rotamer peak, 150 MHz, CD_3OD): δ 176.8, 176.3*, 174.5, 174.4*, 145.7*, 145.6, 140.93, 140.86, 129.88*, 129.87, 128.5, 138.3, 128.2*, 125.8*, 125.7, 84.6, 83.6*, 71.7, 63.68*, 63.66, 59.02, 59.01*, 51.8*, 51.6, 51.3, 51.1*, 50.8*, 50.7, 49.2, 49.1, 40.5, 40.4*, 35.7*, 35.4, 26.29*, 26.28, 17.4, 17.2; HRMS (ESI-TOF): $C_{22}H_{31}N_5O_4$: $[M+H]^+$: calculated 430.2449, found 430.2446.



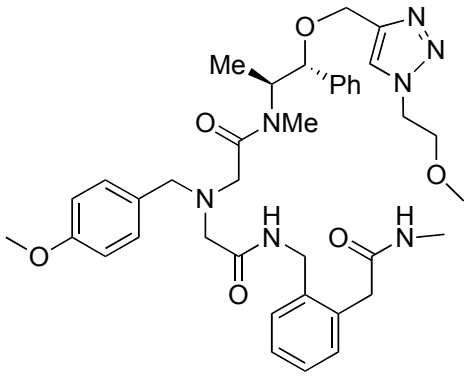
4b. Prepared according to General Procedure F using 10 reaction segments containing alkyne **40** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 5% MeOH in EtOAc, R_f = 0.33) followed to yield **4b** as light green solid (120.7 mg, 65% yield). The solid was dissolved in MeOH (2 mL), stirred overnight with Quadrapure TU resin, filtered and concentrated to give a white solid: ^{1}H NMR (1.7:1 rotamer ratio, asterisk denotes minor rotamer peak, 600 MHz, CD_3OD): δ 8.37 (br. s., 1 H), 7.91* (s, 1 H), 7.89 (s, 1 H), 7.70 (m, 2 H), 7.27 - 7.39 (m, 5 H), 7.21 (d, *J* = 8.4 Hz, 2 H), 7.11 (d, *J* = 8.4 Hz, 2 H), 4.67 (br. s., 1 H), 4.47 - 4.56 (m, 4 H), 4.38 (m, 1 H), 4.06* (m, 1 H), 3.75 (t, *J* = 4.2 Hz, 2 H), 3.32 (m, 3 H), 2.90 (m, 3 H), 2.67 - 2.80 (m, 5 H), 2.37 - 2.54 (m, 2 H), 1.24* (d, *J* = 6.6 Hz, 3 H), 1.17 (d, *J* = 7.2 Hz, 3 H) ; ^{13}C NMR (asterisk denotes minor rotamer peak, 150 MHz, CD_3OD): δ 174.3*, 174.2, 170.59*, 170.55, 146.5, 146.4, 145.7*, 145.6*, 140.31, 140.29*, 133.4, 133.3*, 129.7, 129.57*, 129.55, 129.52*, 129.4, 129.1*, 128.44, 128.39*, 128.35, 128.30*, 125.8, 84.7*, 83.9, 71.70*, 71.68, 63.0*, 62.9, 59.2, 59.02*, 59.01, 51.3*, 51.2, 35.9, 34.8*, 32.0*, 31.8, 29.0, 27.0, 26.9*, 15.7, 13.1*; HRMS (ESI-TOF): $C_{27}H_{35}N_5O_4$: $[M+H]^+$: calculated 494.2762, found 494.2761.



5b. Prepared according to General Procedure F using six reaction segments containing alkyne **42** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). The crude reaction mixture was purified using a Biotage Horizon automated flash column chromatography system (silica gel, 0-5% MeOH in EtOAc, R_f = 0.50) followed by PTLC (silica gel, 500 µm plates, 5% MeOH in CH_2Cl_2) to yield **5b** as colorless oil (73.6 mg, 62% yield): ^{1}H NMR (600 MHz, CD_3OD): δ 7.91 (s, 1 H), 7.20 - 7.35 (m, 4 H), 4.60 (m, 2 H), 4.54 (m, 3 H), 4.41 (m, 1 H), 4.24 (m, 2 H), 3.76 (t, *J* = 4.8 Hz, 2 H), 3.63 (m, 4 H), 3.35 (m, 3 H), 2.71 (s, 3 H), 2.39 (m, 1 H), 2.01 (m, 1 H), 1.45 (s, 3 H), 1.29 (s, 6 H); ^{13}C NMR (asterisk denotes minor rotamer peak, 150 MHz, CD_3OD): δ 175.0, 174.8*, 174.2, 174.1*, 156.4*, 156.0, 145.6, 138.2, 137.8*, 135.5, 135.4*, 131.6, 130.7, 130.2*, 129.0, 128.8*, 128.5, 128.4*, 125.7, 81.6, 81.5*, 78.5*, 77.8, 71.7, 62.8, 60.9*, 60.5*, 59.0, 53.7, 53.1*, 51.3, 49.9, 43.4*, 42.1, 40.7, 40.6*, 38.0, 37.0, 28.7*, 28.5, 26.6; HRMS (ESI-TOF): $C_{26}H_{39}N_6O_6$: $[M+H]^+$: calculated 531.2925, found 531.2933.

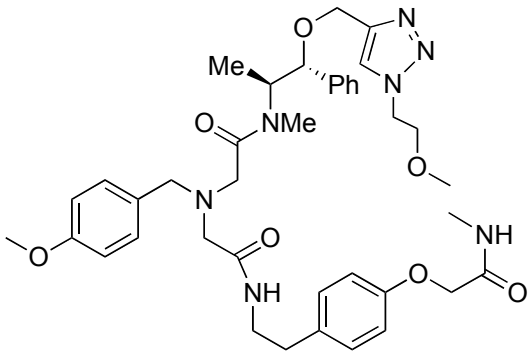


6b. Prepared according to General Procedure F using four reaction segments containing alkyne **44** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). The crude reaction mixture was purified using PTLC (silica gel, 500 µm plates, 10% MeOH in CH_2Cl_2, R_f = 0.33 in 10% MeOH in CH_2Cl_2) to yield **6b** as colorless oil (31.5 mg, 35% yield): ^{1}H NMR (3.6:1 rotamer ratio, asterisk denotes minor rotamer peak, 600 MHz, CD_3OD): δ 7.95* (s, 1 H), 7.89 (s, 1 H), 7.24 - 7.38 (m, 5 H), 7.12 (m, 2 H), 6.89 (m, 2 H), 4.80 (br. s., 1 H), 4.55 (m, 2 H), 4.34 - 4.47 (m, 4 H), 3.77 (m, 2 H), 3.33 (m, 3 H), 3.29 (m, 2 H), 2.84 - 3.00 (m, 4 H), 2.80 (s, 3 H), 2.66 - 2.76 (m, 3 H), 2.37 (m, 1 H), 2.05 (m, 1 H), 1.33* (d, J = 6.6 Hz, 3 H), 1.22 (d, J = 6.6 Hz, 3 H), 0.92* (d, J = 7.2 Hz, 3 H), 0.48 (d, J = 6.6 Hz, 3 H); ^{13}C NMR (150 MHz, CD_3OD): δ 177.7, 174.0, 171.7, 157.7, 145.8, 140.2, 133.8, 130.9, 129.33, 129.26, 128.8, 125.8, 115.8, 85.0, 71.7, 68.3, 62.7, 59.0, 51.2, 42.0, 40.4, 35.6, 33.8, 26.0, 16.8, 13.9; HRMS (ESI-TOF): $C_{32}H_{44}N_6O_6$: $[M+H]^+$: calculated 609.3395, found 609.3402.



7b. Prepared according to General Procedure F using three reaction segments containing alkyne **45** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). Additionally, one segment containing alkyne **45** (0.25 M in DMF, 75 µL, 0.019 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 75 µL, 0.038 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 93.8 µL, 0.028 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 37.5 µL, 0.019 mmol, 1.0 eq) was run. The crude reaction mixture was purified using PTLC (silica gel, 500 µm plates, 10% MeOH in CH_2Cl_2, R_f = 0.33 in 10% MeOH in CH_2Cl_2) to yield **7b** as colorless oil (57.0 mg, 62% yield): ^{1}H NMR (1.6:1 rotamer ratio, asterisk denotes minor rotamer peak, 600 MHz, CD_3OD): δ 7.90* (s, 1 H), 7.88 (s, 1 H), 7.01 - 7.38 (m, 13 H), 6.76 (m, 2 H), 4.57 (br. s., 1 H), 4.51 - 4.57 (m, 2 H), 4.44 - 4.48 (m, 2 H), 4.33 - 4.38 (m, 4 H), 3.75 (m, 6 H), 3.60 (m, 2 H), 3.44 (m, 1 H), 3.33 (m, 2 H), 3.32 (s, 2 H), 3.05 (m, 1 H), 2.98 (m, 1 H), 2.86 (m, 1 H), 2.70 (m, 5 H), 1.28* (d, J = 6.6 Hz, 3 H), 1.19 (d, J = 6.6 Hz, 3 H); ^{13}C NMR (asterisk denotes minor rotamer peak,150 MHz, CD_3OD): δ 174.06*, 174.05, 173.8, 173.6*, 172.6*, 172.3, 160.6*, 160.5, 145.7, 145.6*, 140.3, 140.2*, 138.33, 138.26*, 135.12*, 135.11, 131.66, 131.64*, 131.62*, 131.61, 131.1*, 130.8, 130.08*, 130.07, 129.7, 129.6*, 129.4, 129.3*, 128.78*, 128.76, 128.6, 128.52*, 128.50, 128.4*, 125.74*, 125.72, 114.9*, 114.7, 84.6, 83.8*, 71.71*, 71.68, 63.0*, 62.8, 59.8*, 59.5, 59.04, 59.01, 59.00, 58.7*, 58.5*, 58.3*, 56.5,

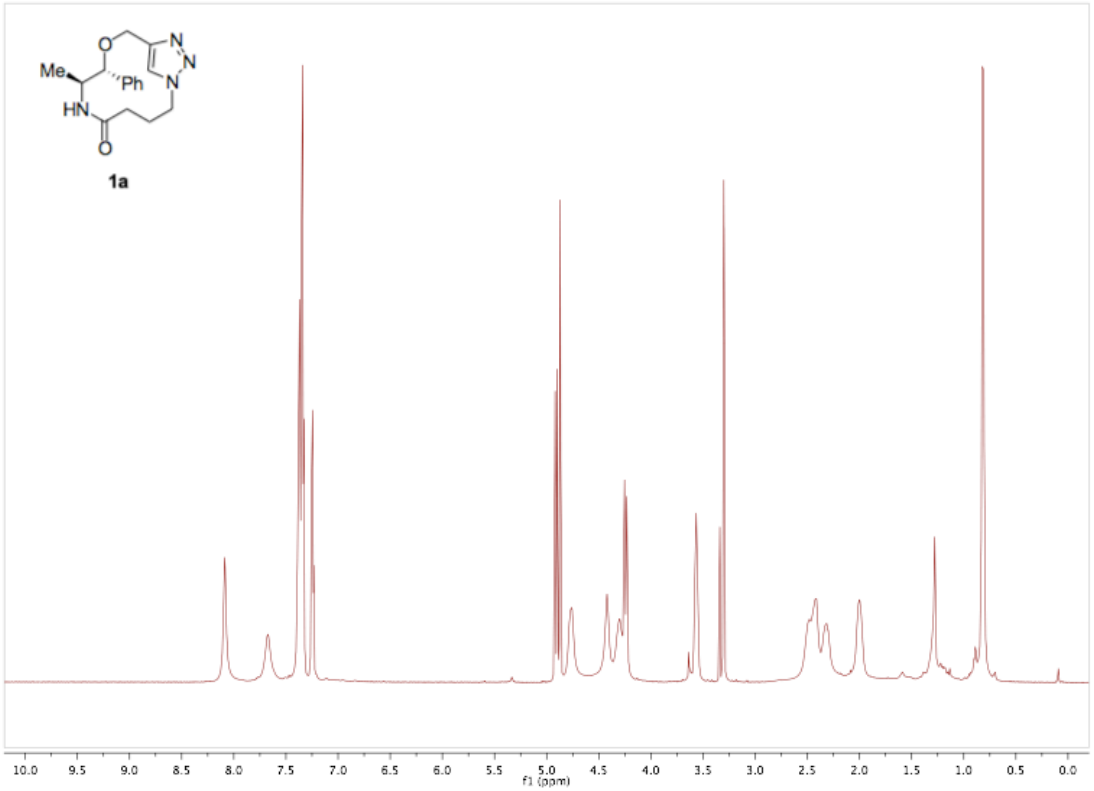
56.3*, 55.70*, 55.65, 55.3*, 51.26*, 51.21, 41.5, 40.73, 40.70*, 33.0, 30.15*, 28.8, 26.62*, 26.61, 15.66, 14.46*, 14.44*, 13.5; HRMS (ESI-TOF): $C_{38}H_{49}N_7O_6$: $[M+H]^+$: calculated 700.3817, found 700.3815.

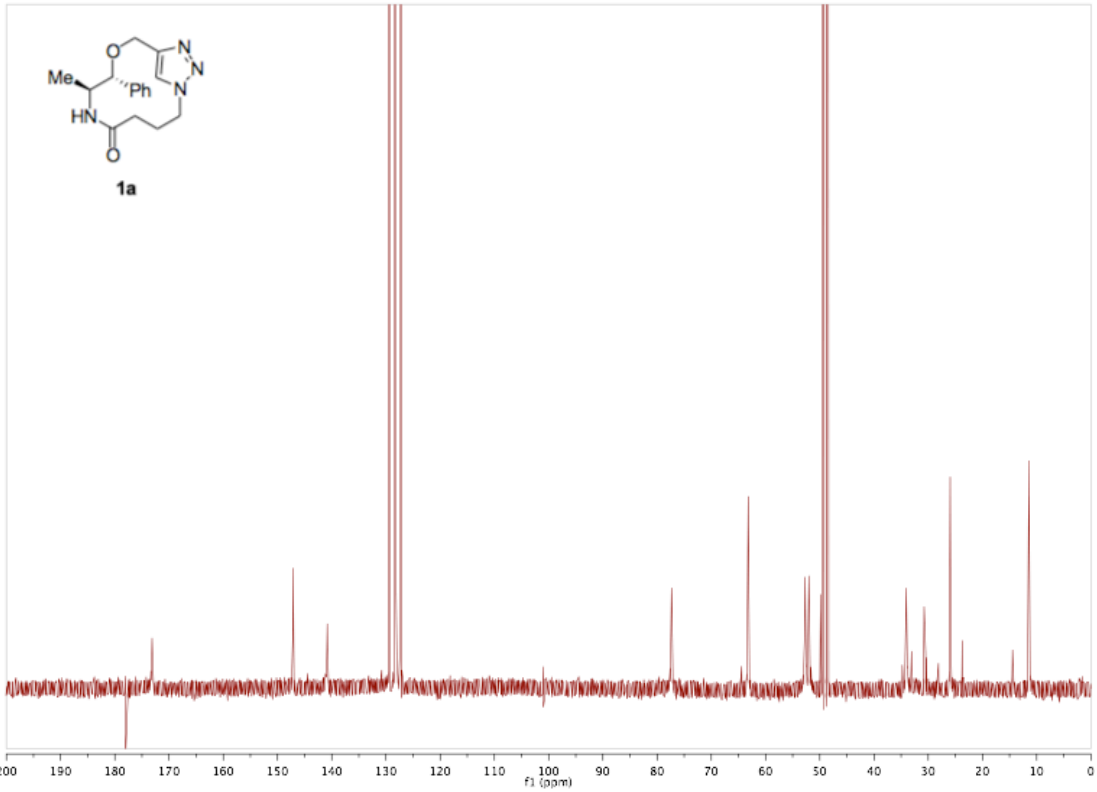


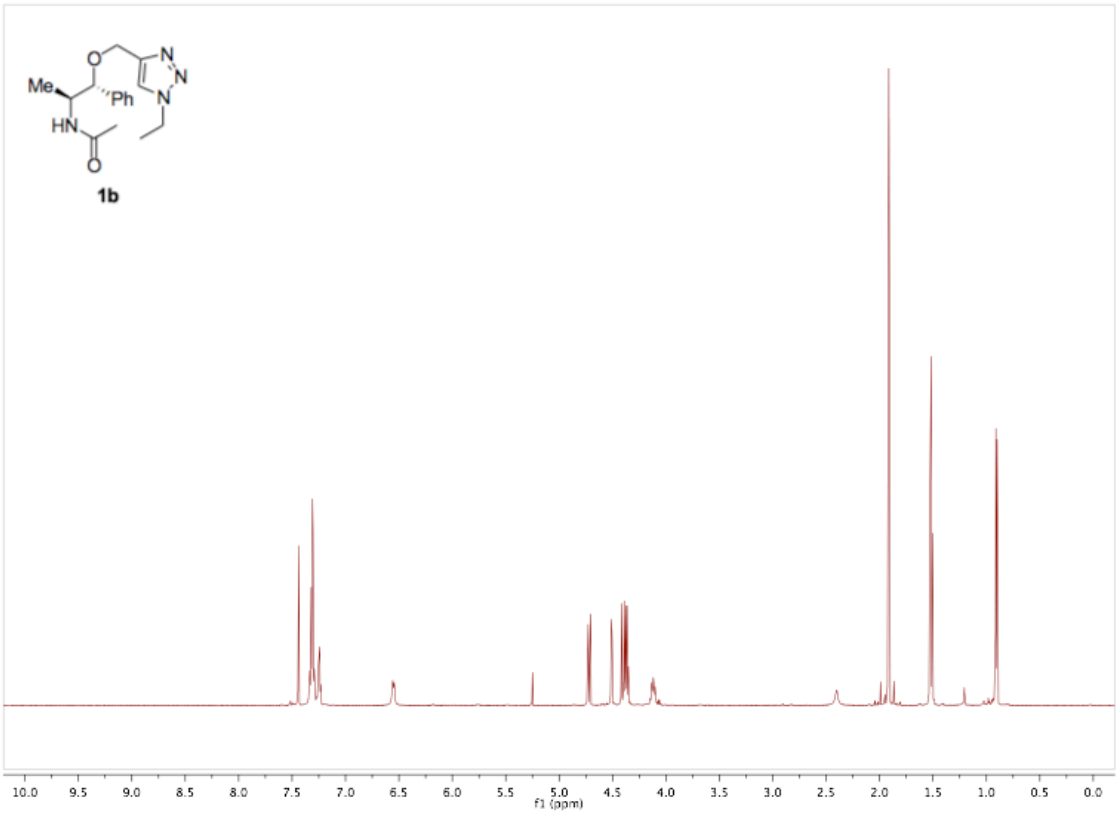
8b. Prepared according to General Procedure F using five reaction segments containing alkyne **46** (0.25 M in DMF, 150 µL, 0.038 mmol, 1.0 eq), 2-bromoethyl methyl ether (0.50 M in DMF, 150 µL, 0.075 mmol, 2.0 eq), NaN_3 (0.30 M in DMF, 187.5 µL, 0.056 mmol, 1.5 eq) and DIPEA (0.5 M in DMF, 75 µL, 0.038 mmol, 1.0 eq). The crude reaction mixture was purified using PTLC (silica gel, 500 µm plates, 10% MeOH in CH_2Cl_2, R_f = 0.33 in 10% MeOH in CH_2Cl_2) to yield **8b** as colorless oil (53.4 mg, 45% yield): ^{1}H NMR (1.7:1 rotamer ratio, asterisk denotes minor rotamer peak, 600 MHz, CD_3OD): δ 7.91* (s, 1 H), 7.88 (s, 1 H), 7.12 - 7.39 (m, 7 H), 6.95 - 7.03 (m, 2 H), 6.78 - 6.88 (m, 4 H), 4.70 (br. s., 1 H), 4.51 - 4.58 (m, 2 H), 4.43 - 4.48 (m, 2 H), 4.33 - 4.38 (m, 3 H), 3.83* (m, 1 H), 3.36 - 3.48 (m, 3 H), 3.31 - 3.34 (m, 4 H), 2.83 - 3.29 (m, 5 H), 2.80 (m, 3 H), 2.75 (m, 2 H), 2.67 (m, 2 H), 1.28* (d, J = 6.6 Hz, 3 H), 1.20 (d, J = 6.6 Hz, 3 H); ^{13}C NMR (asterisk denotes minor rotamer peak,150 MHz, CD_3OD): δ 173.8, 173.6*, 172.5*, 172.1, 171.6, 160.6*, 160.5, 157.70*, 157.69, 145.7, 145.5*, 133.6, 133.5*, 131.6, 131.5*, 131.01, 130.97*, 130.7, 129.64, 129.58*, 128.38, 128.31*, 128.62, 128.32*, 125.74*, 125.7, 115.86*, 115.84, 114.9*, 114.7, 84.6, 83.8* 71.69*, 71.68, 68.13, 68.12*, 62.96*, 62.82,

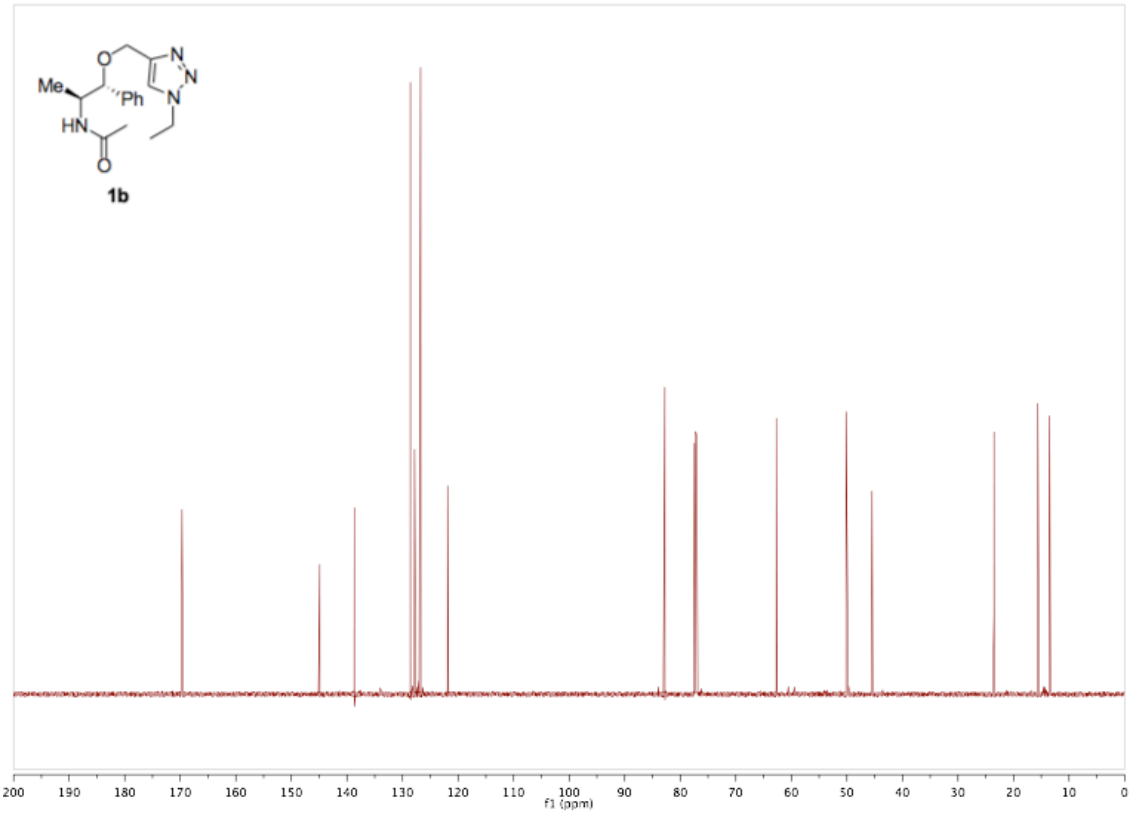
59.7*, 59.3, 59.04*, 59.02, 58.84, 58.74*, 58.27, 56.2*, 55.8*, 55.72, 55.67, 51.3*, 51.2, 47.37, 47.34*, 41.37, 41.35*, 35.34, 35.33*, 33.0, 30.1*, 28.8, 27.4, 27.3*, 26.0, 15.7, 13.6*; HRMS (ESI-TOF): $C_{38}H_{49}N_7O_6$: $[M+H]^+$: calculated 730.3923, found 730.3910.

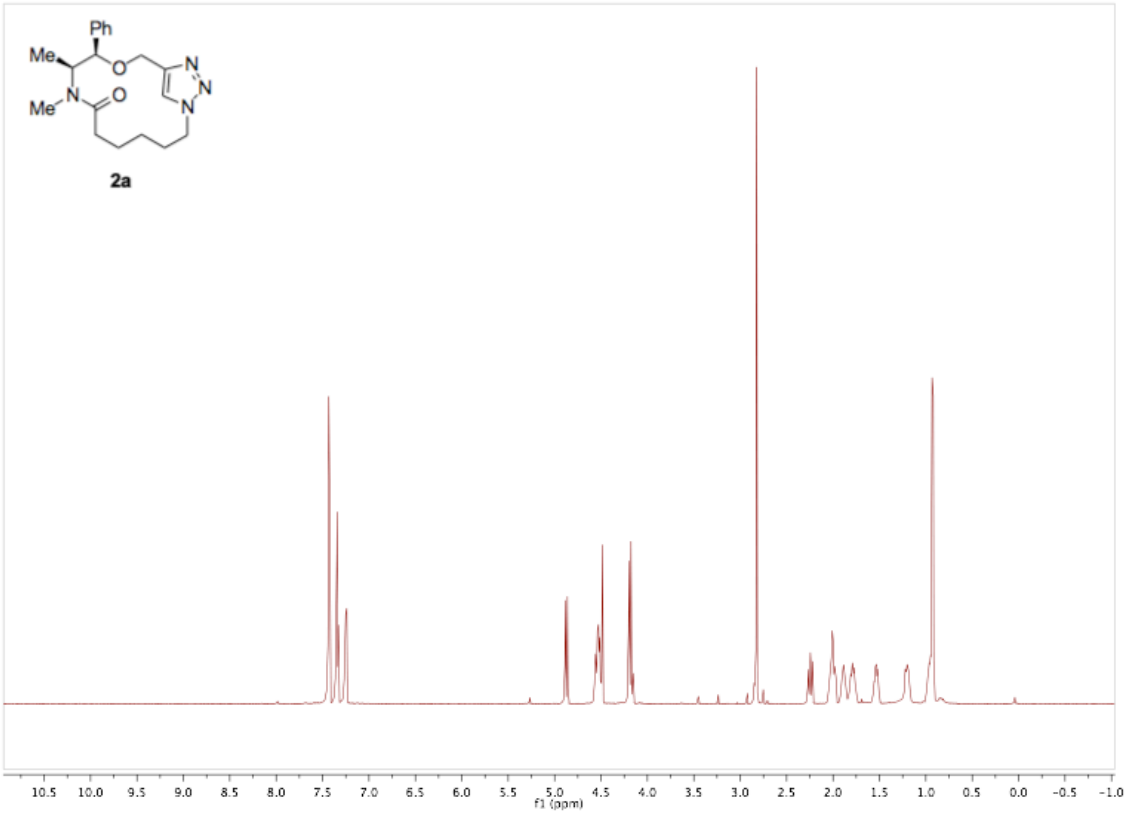
NMR Spectra

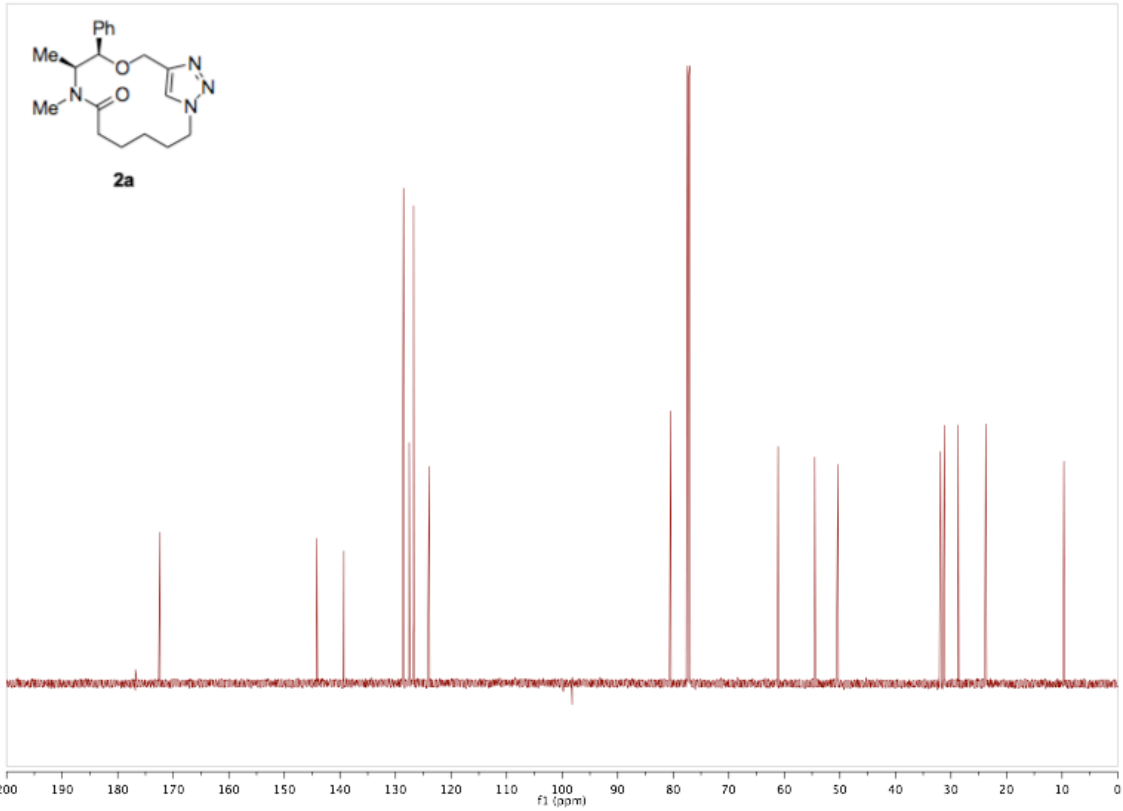


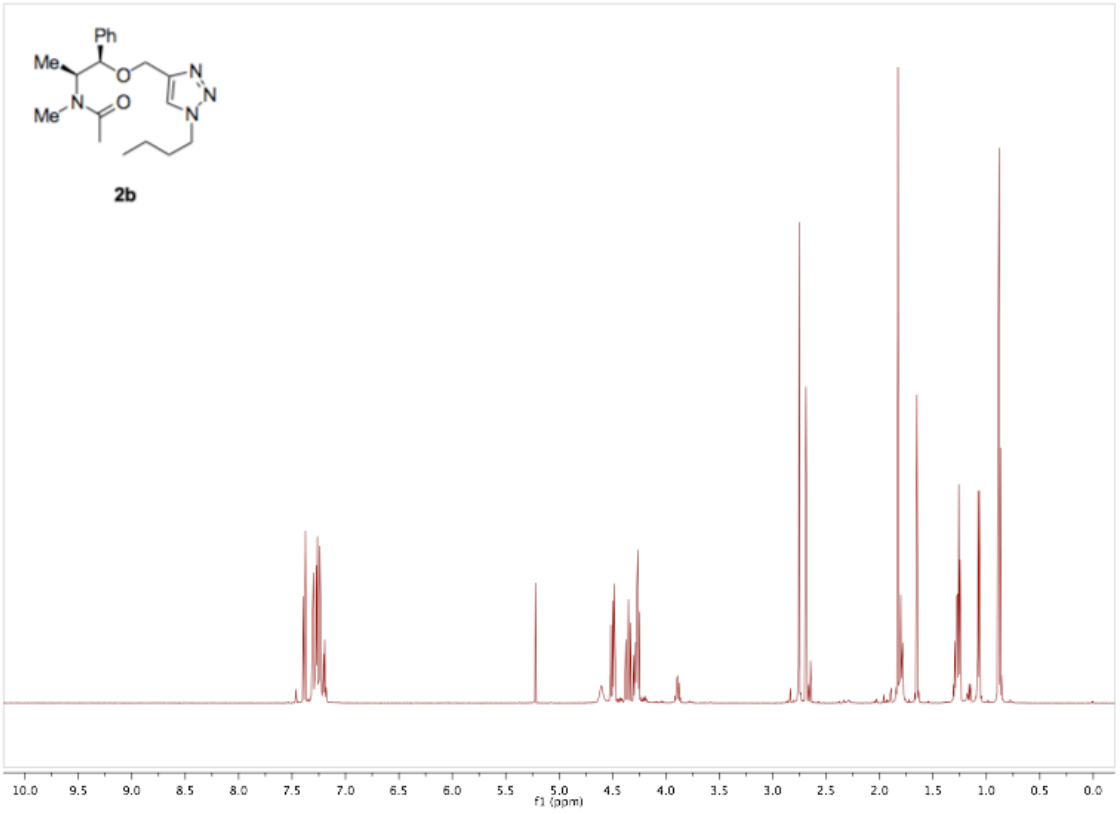


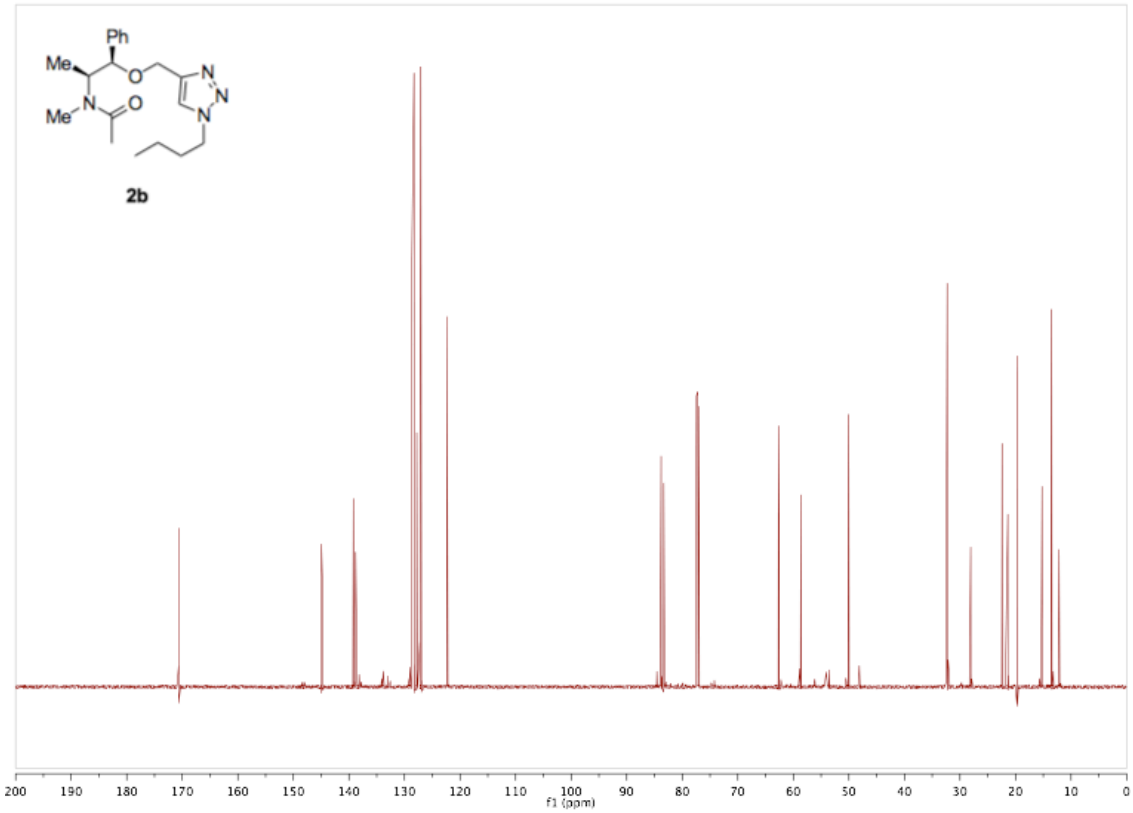


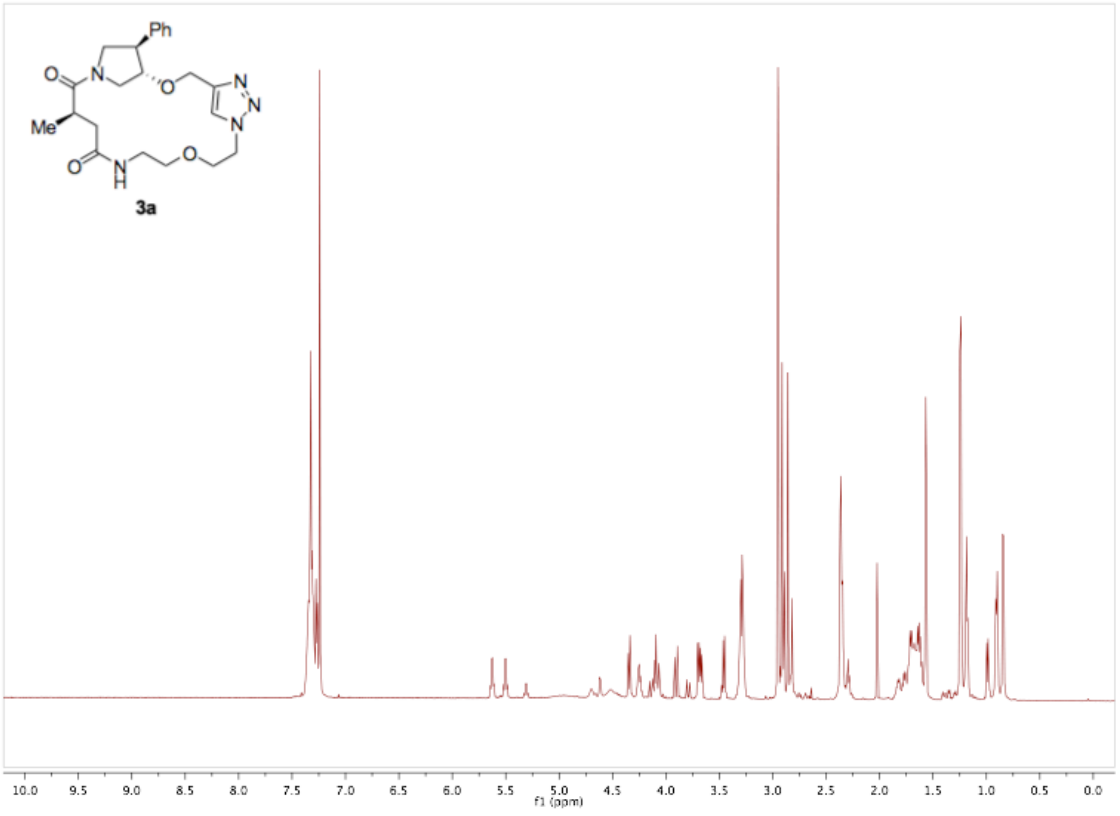


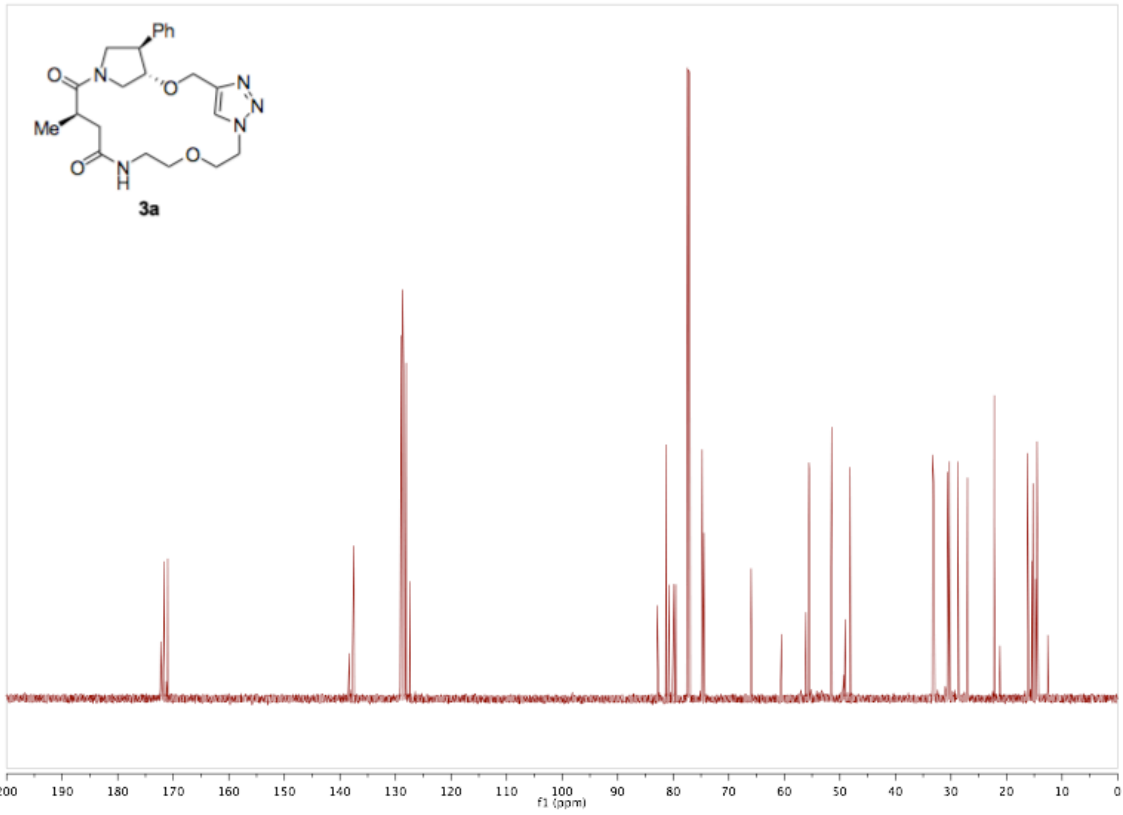


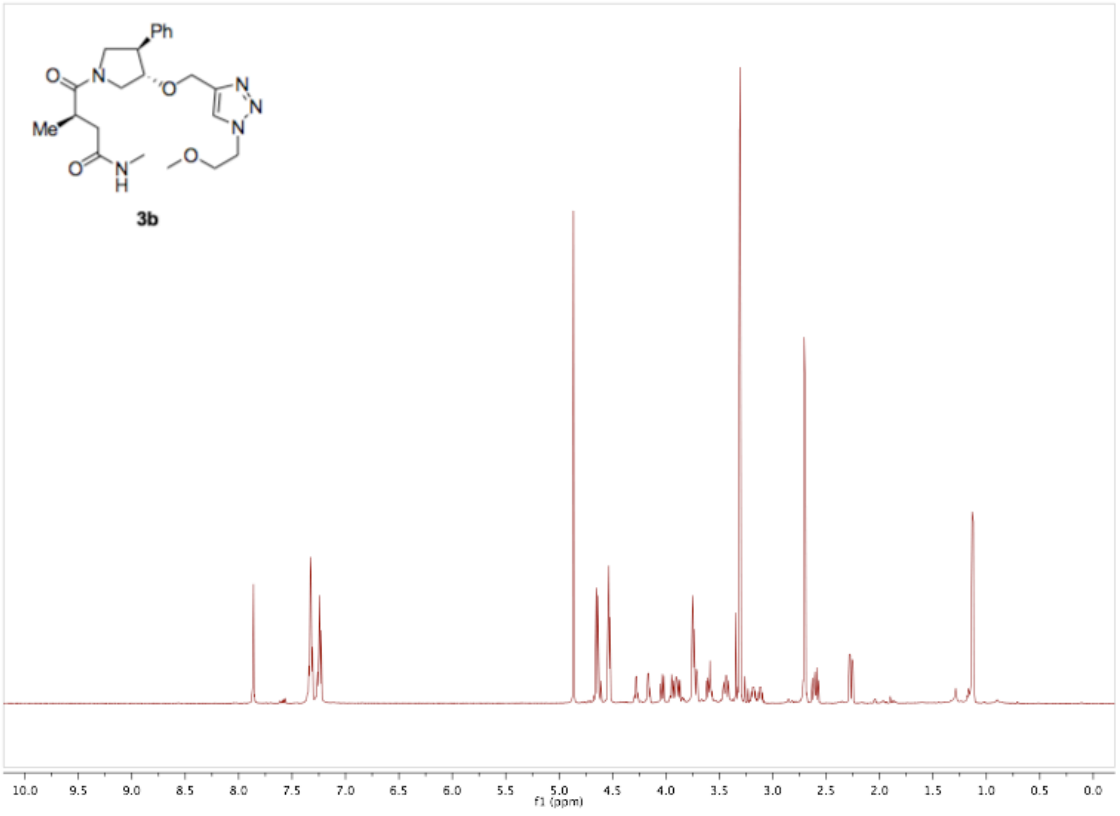


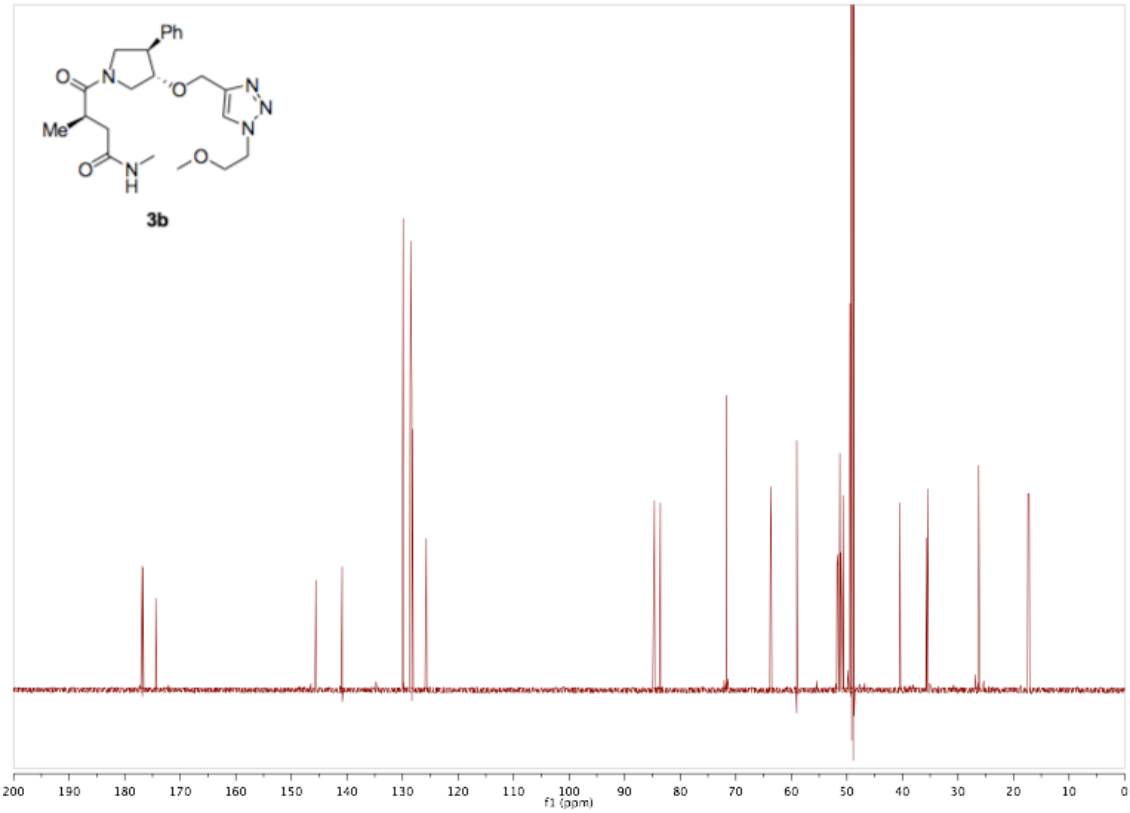


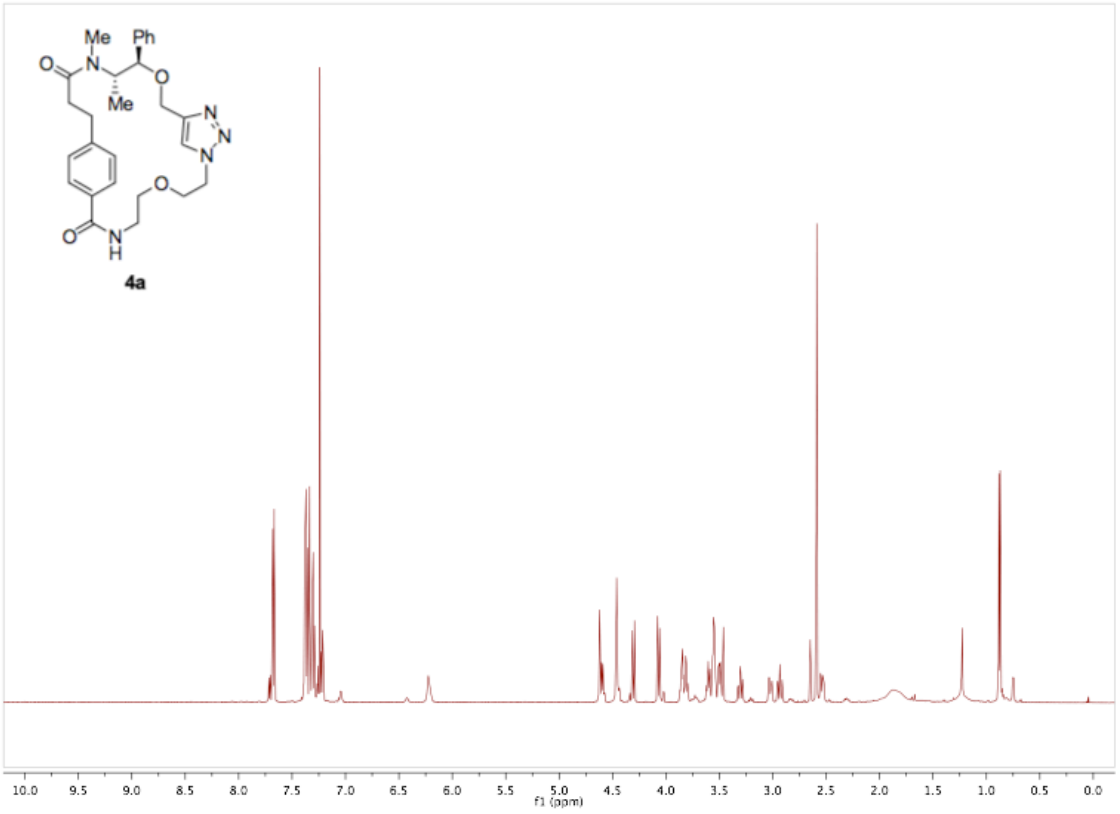


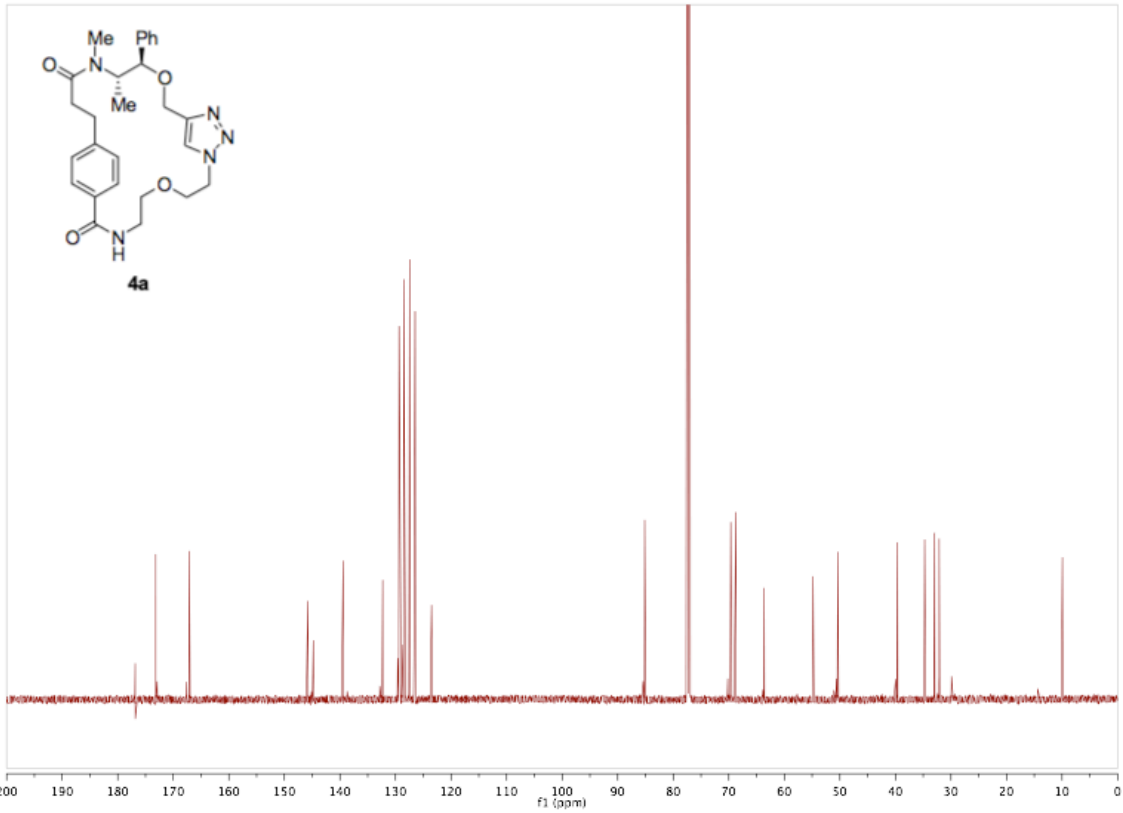


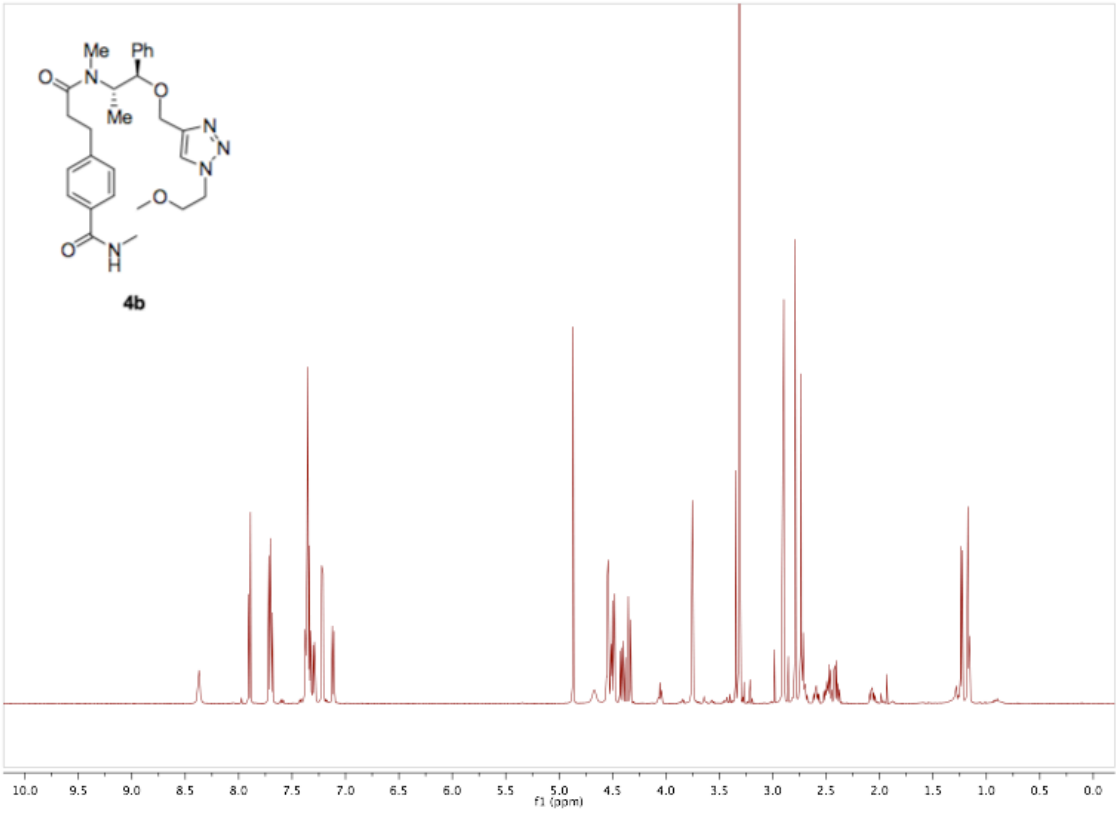


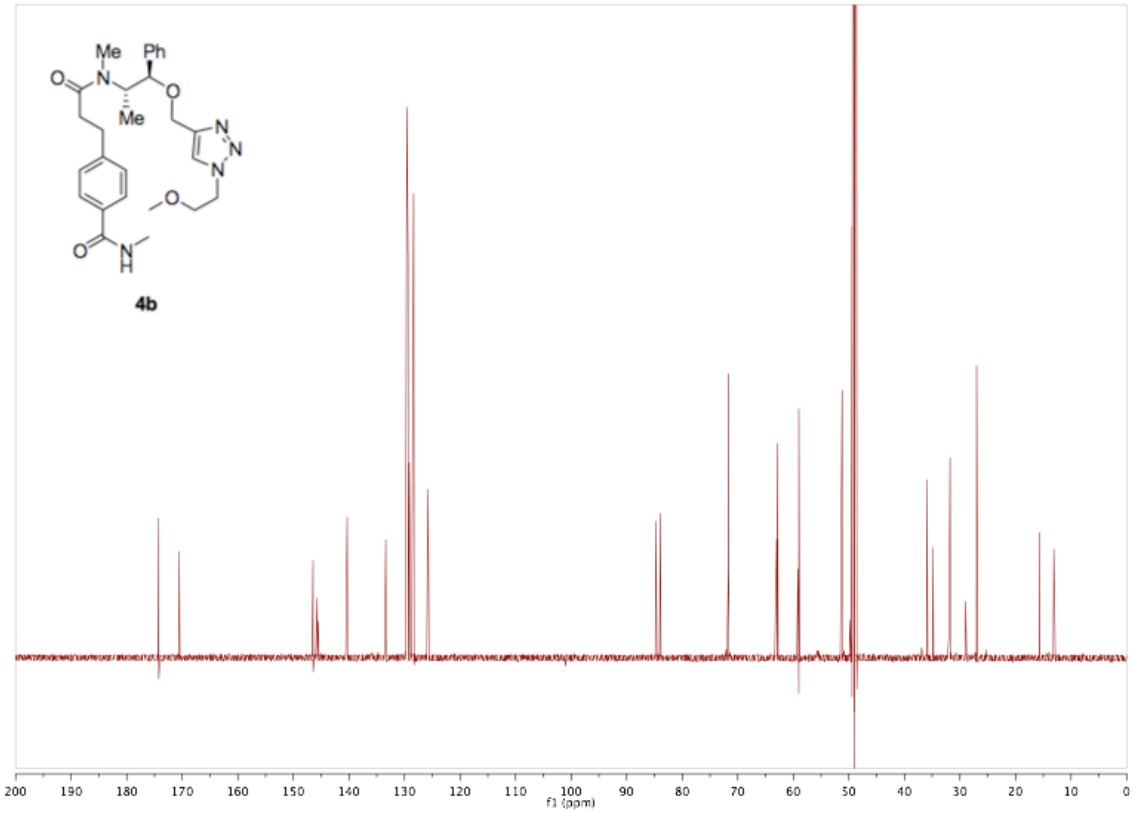


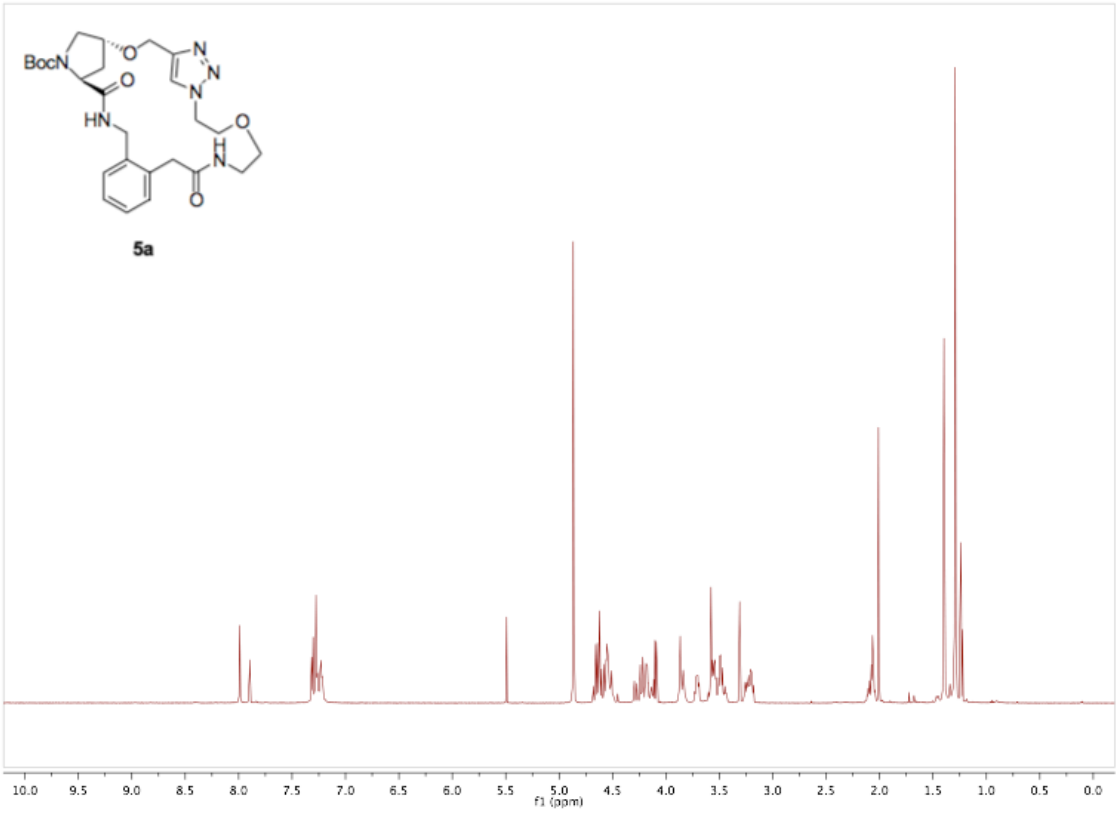


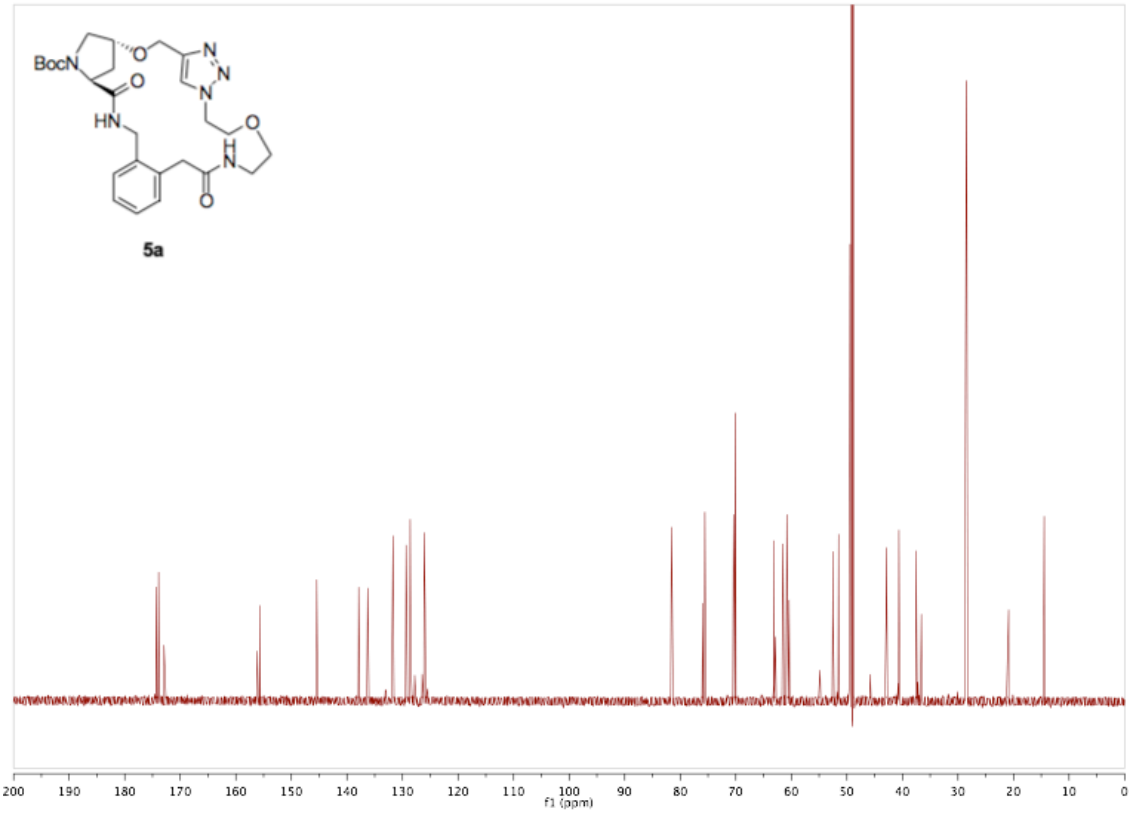


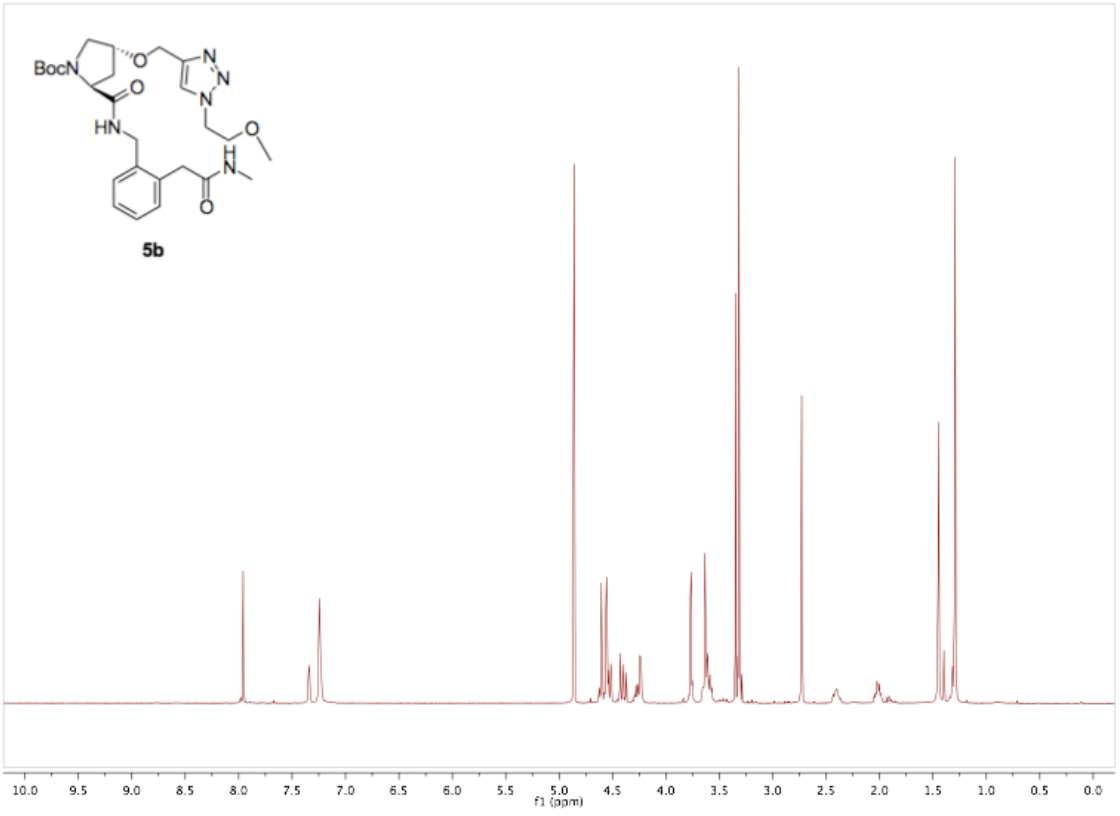


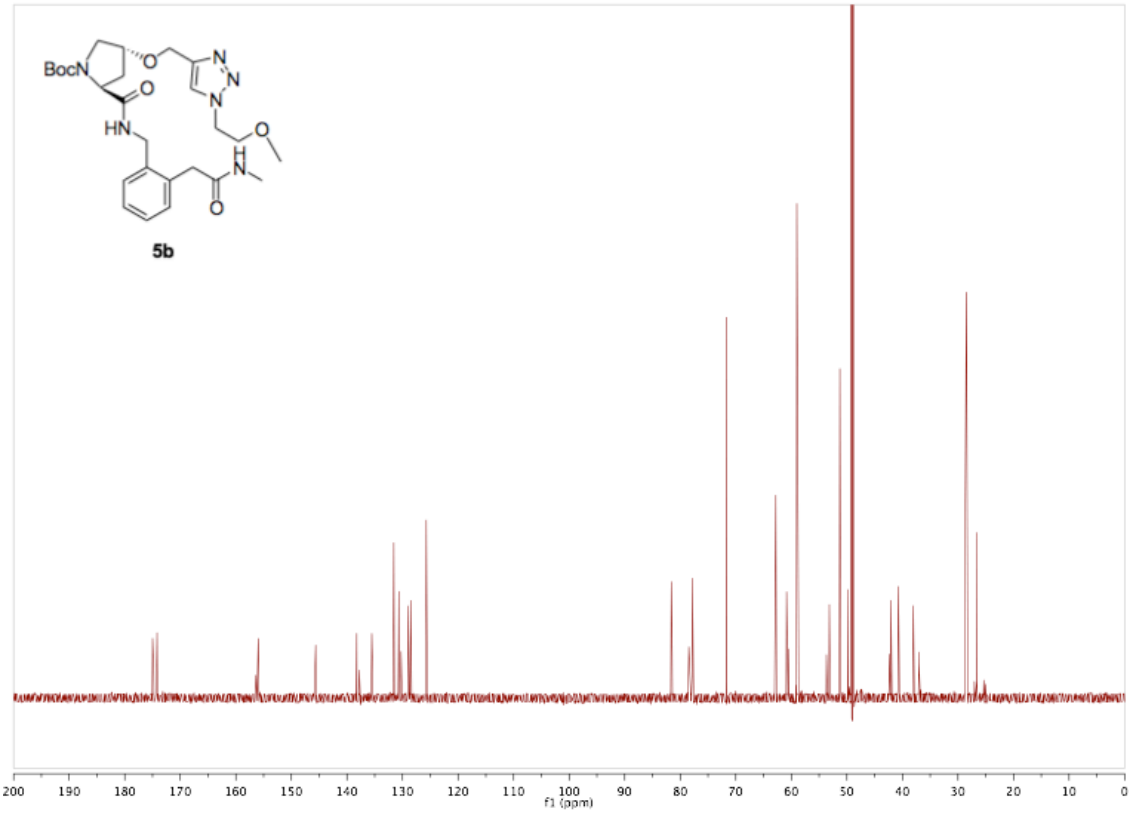


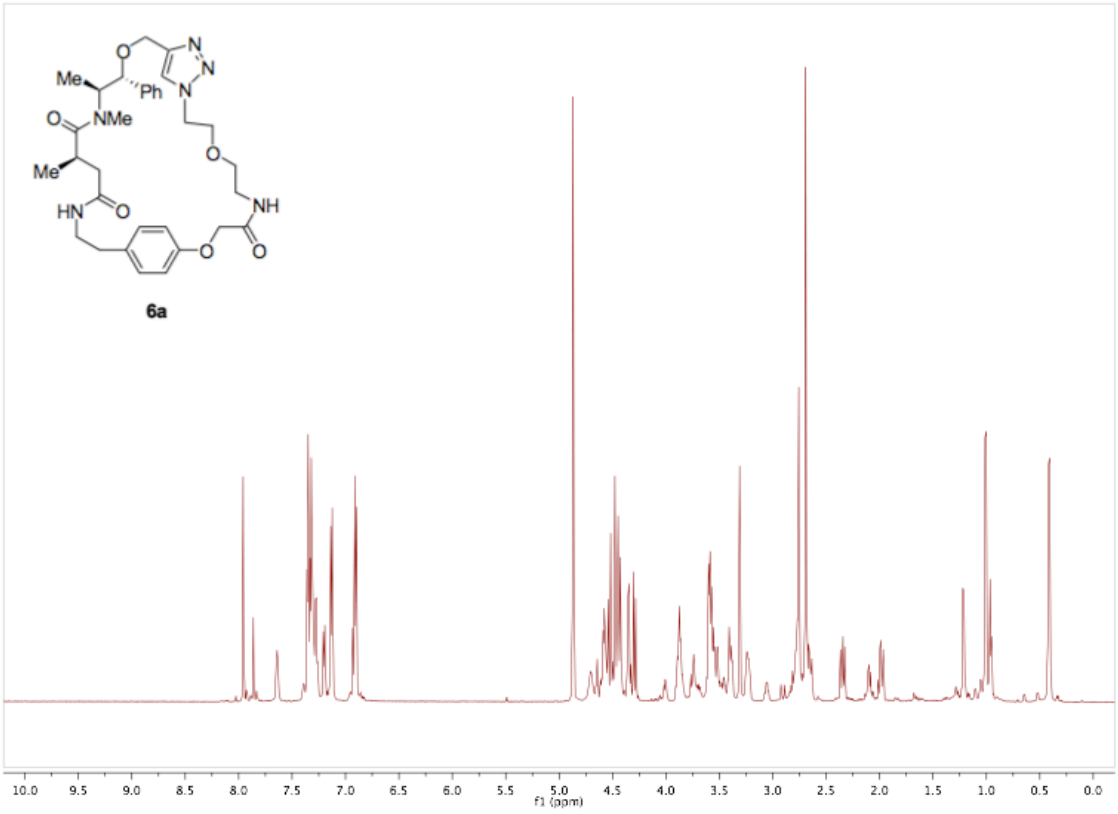


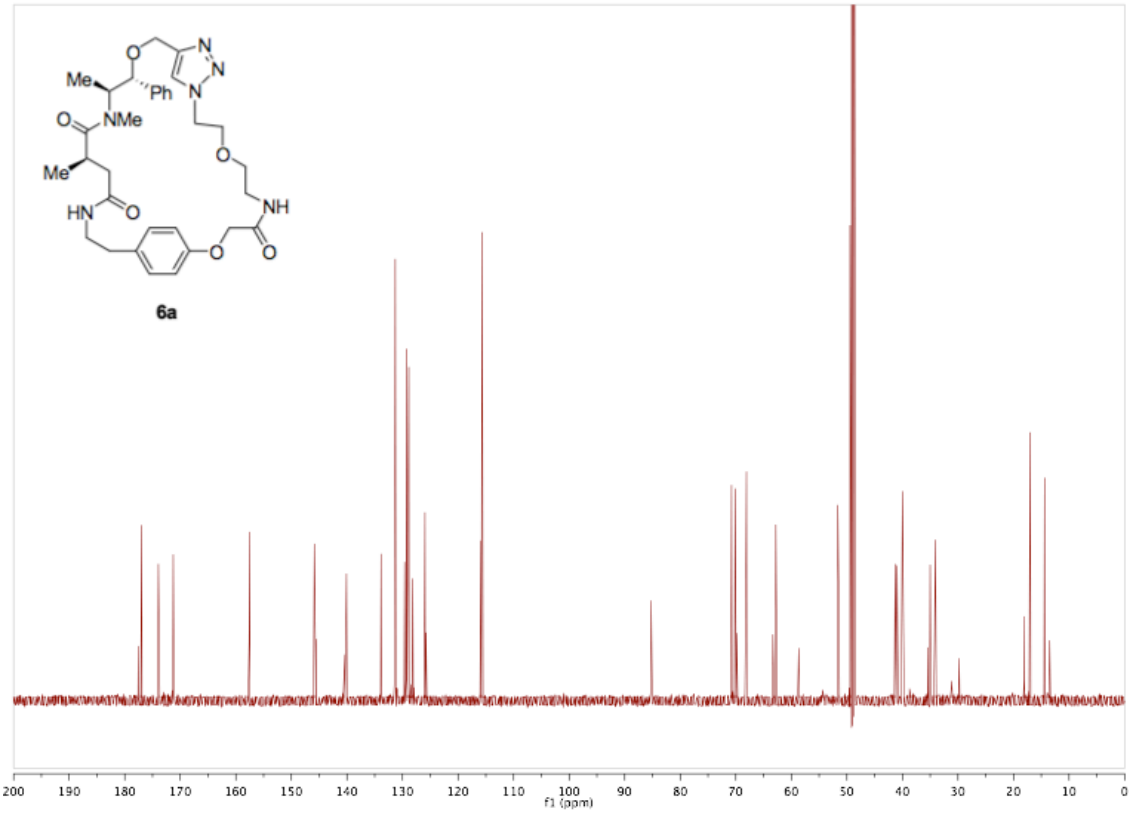


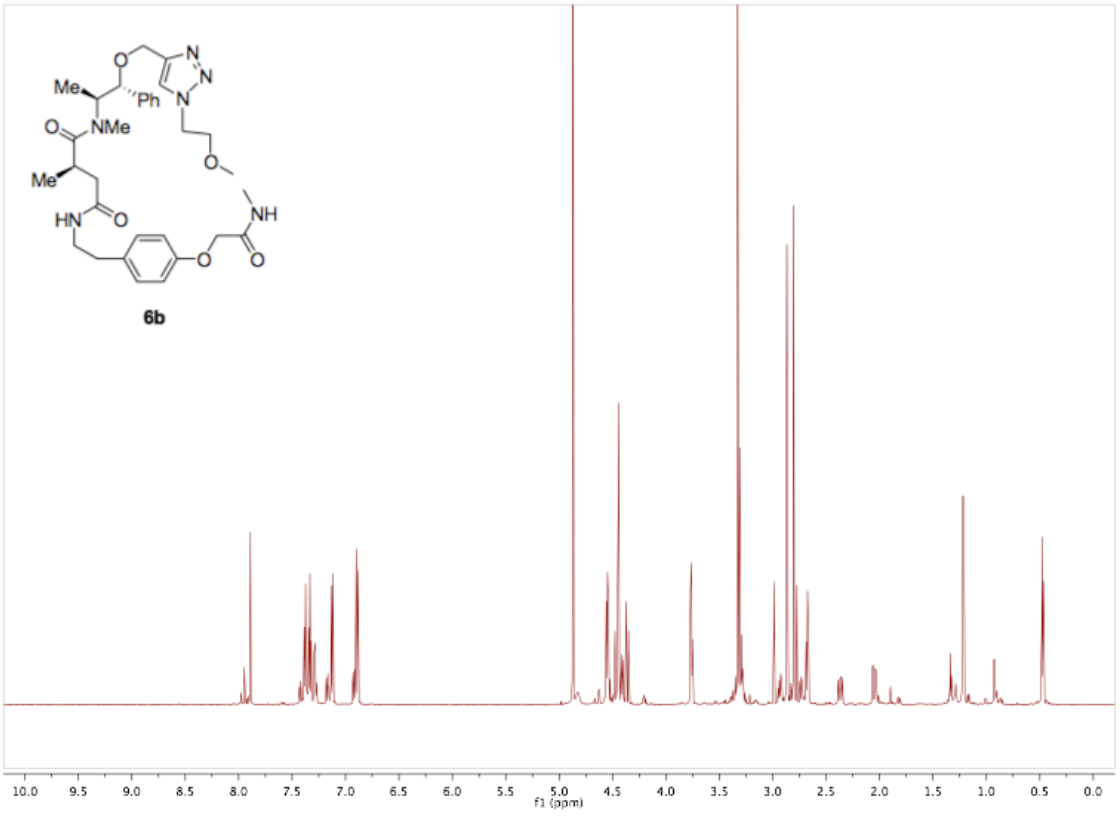




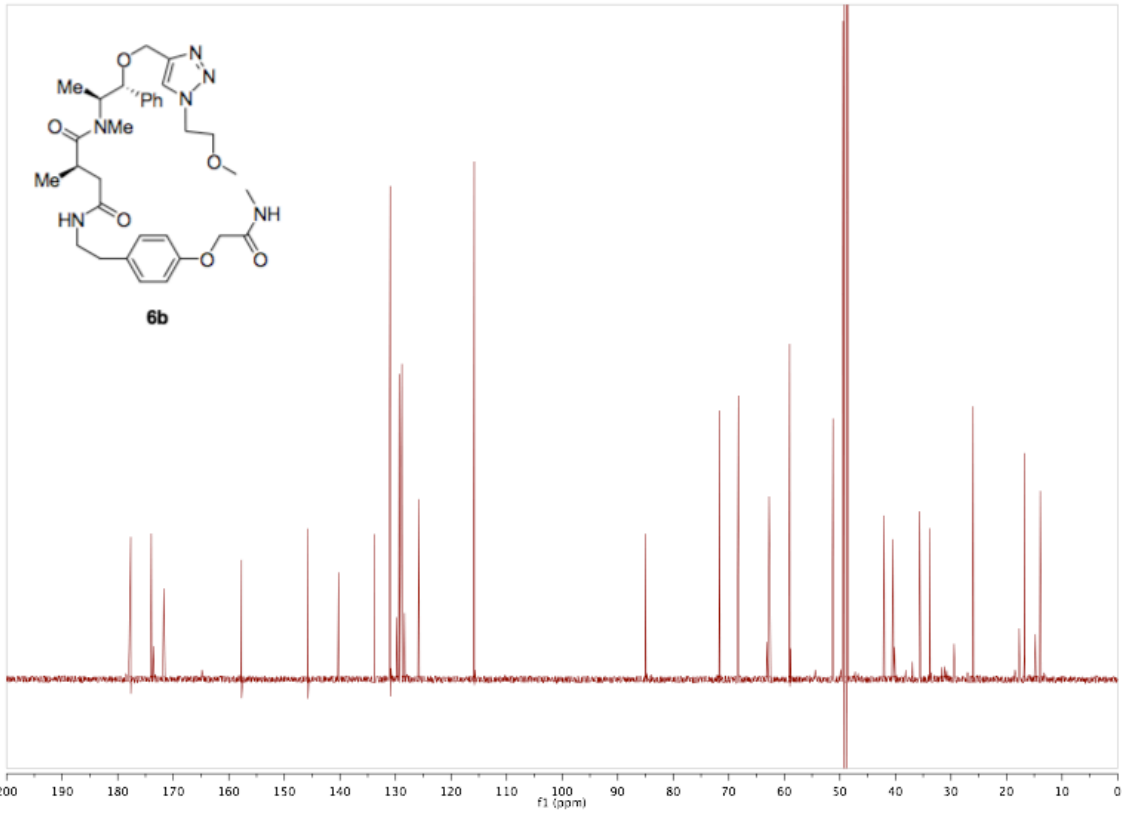




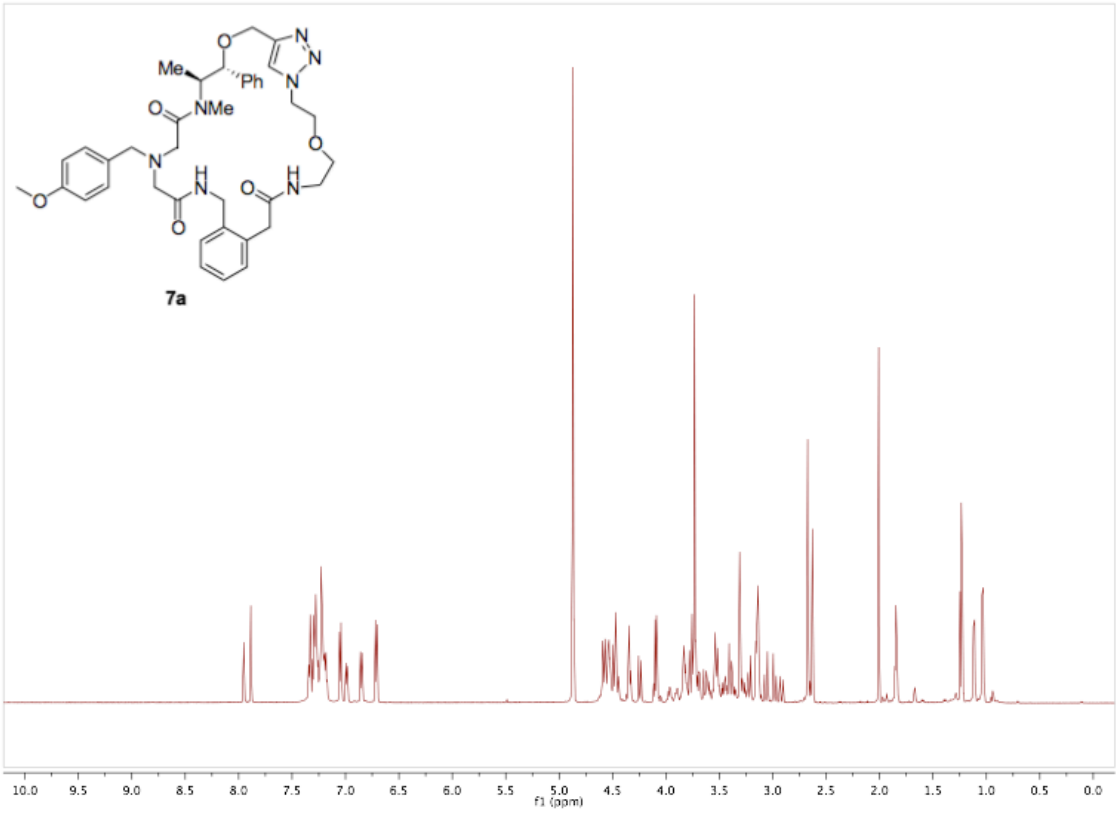


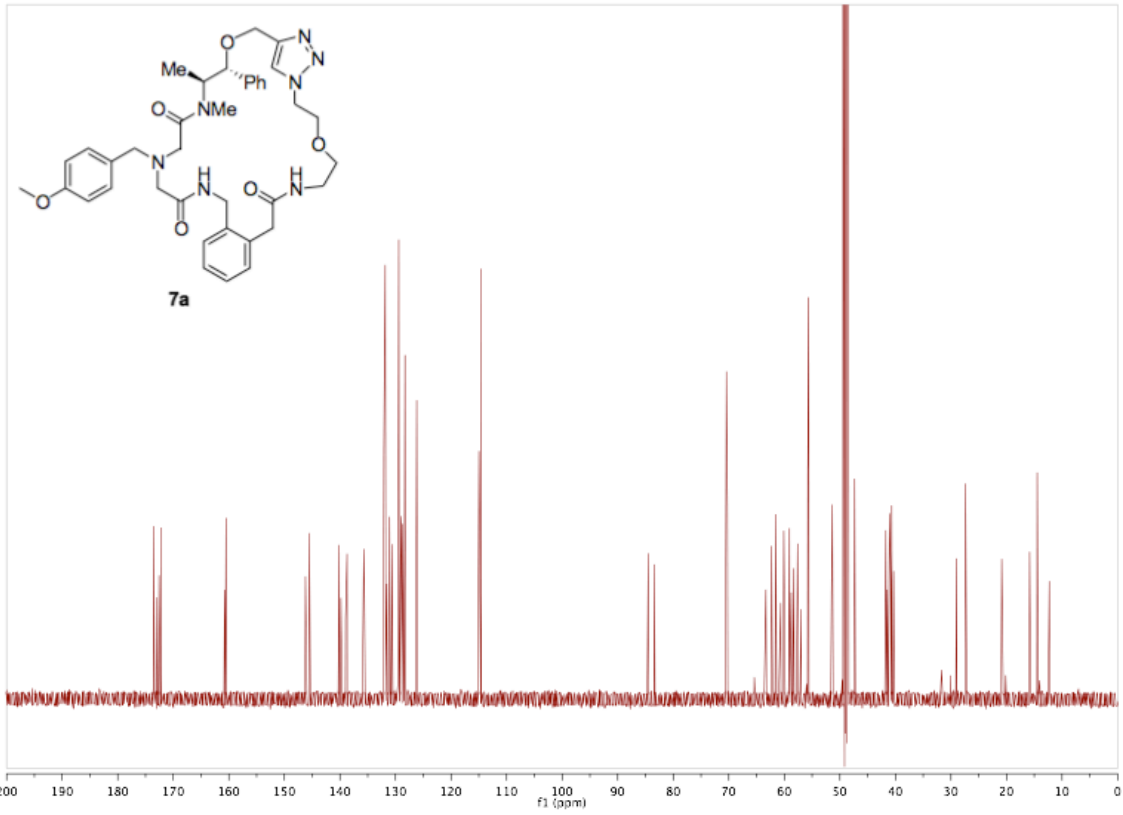


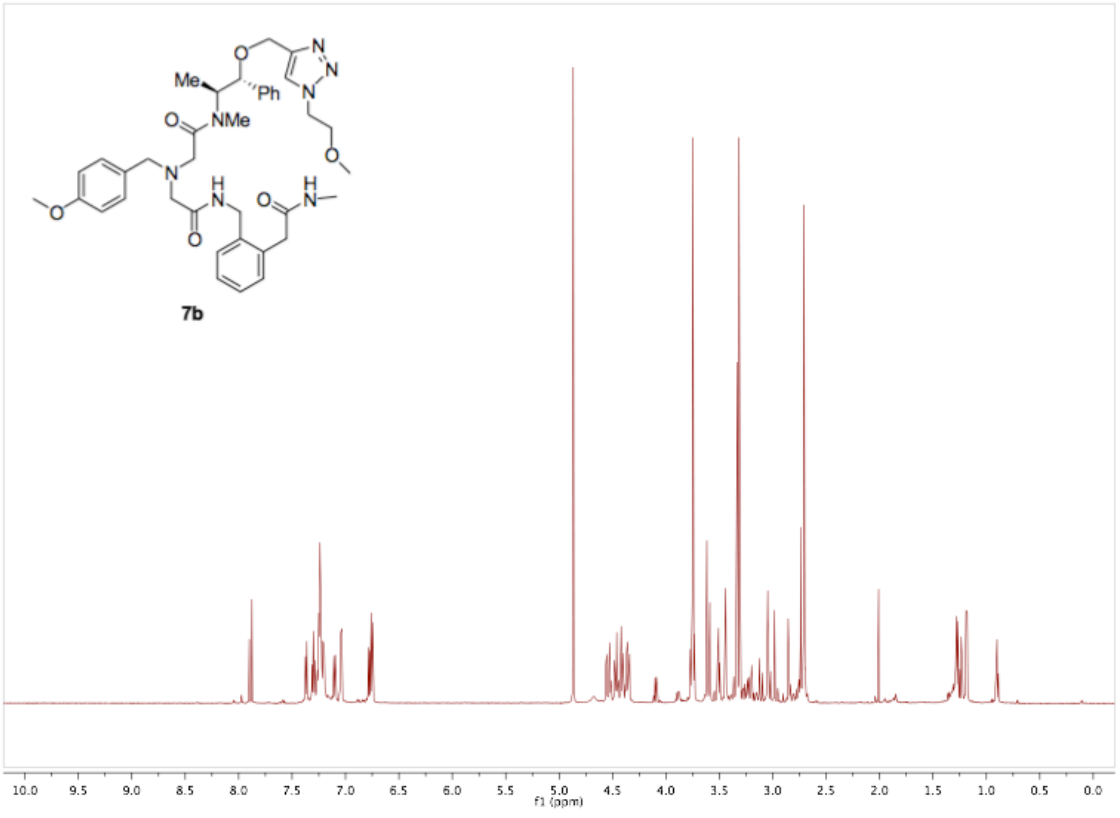
6b

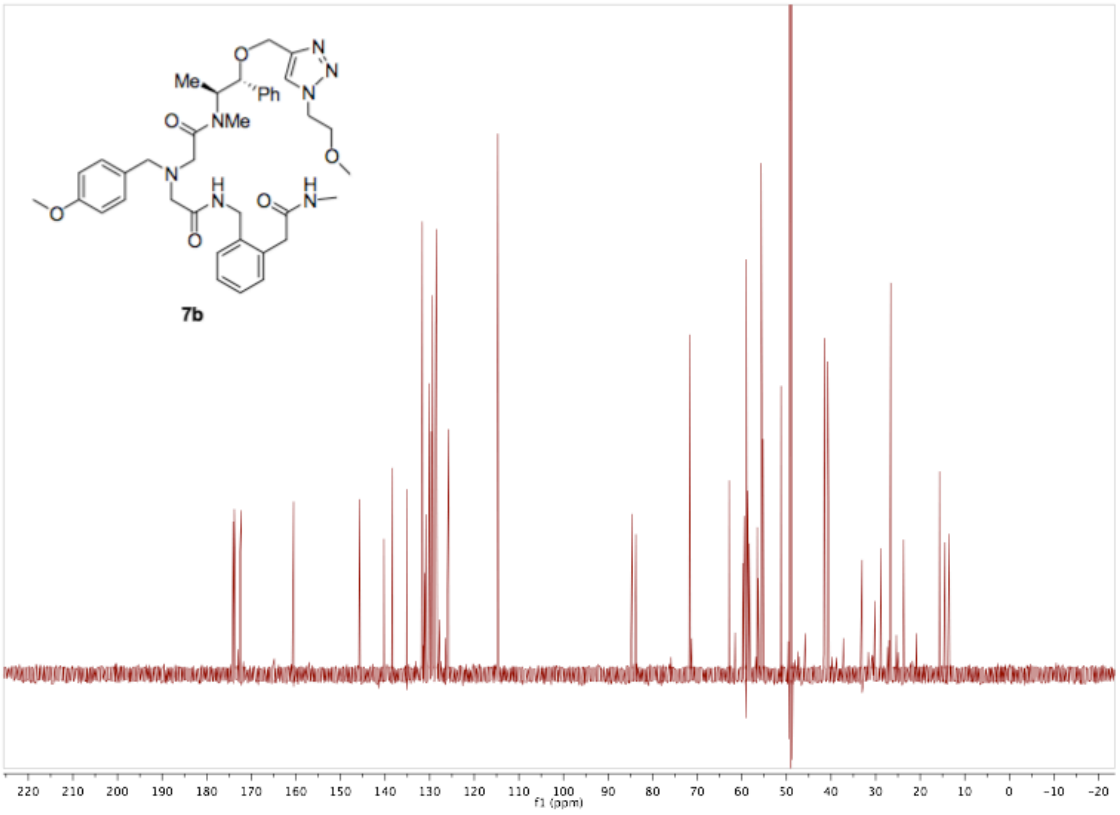


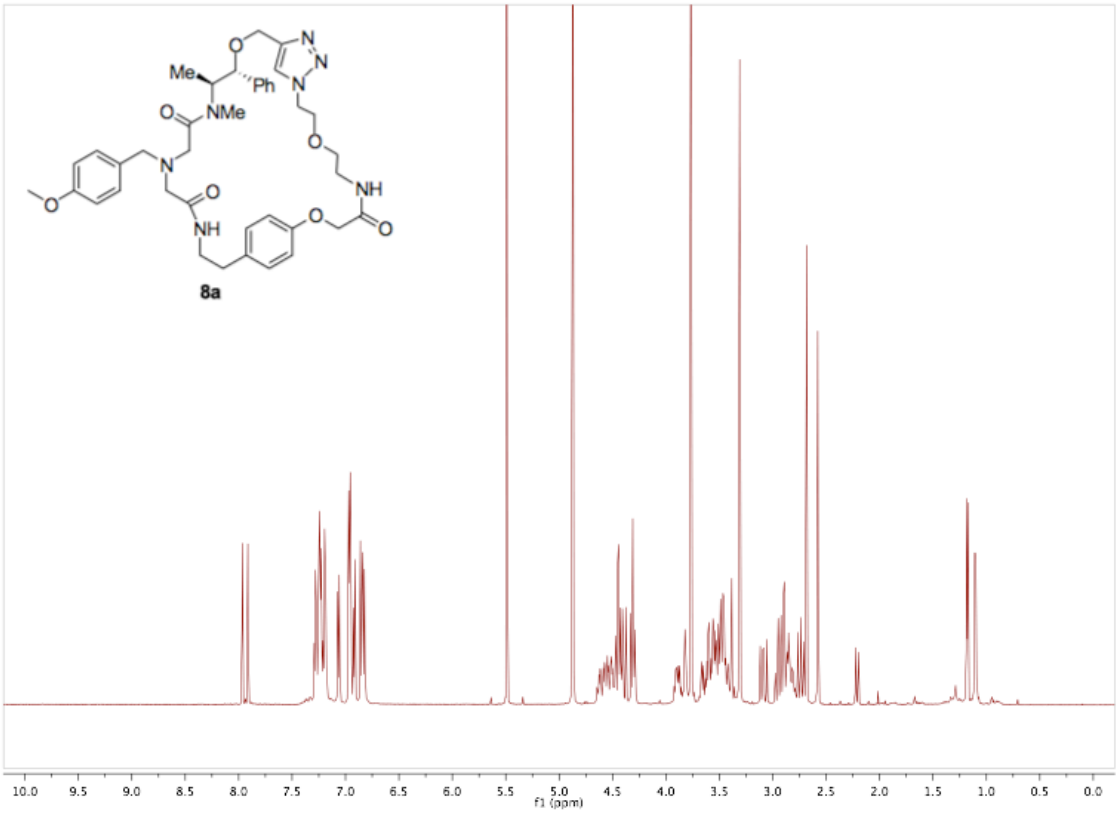
6b

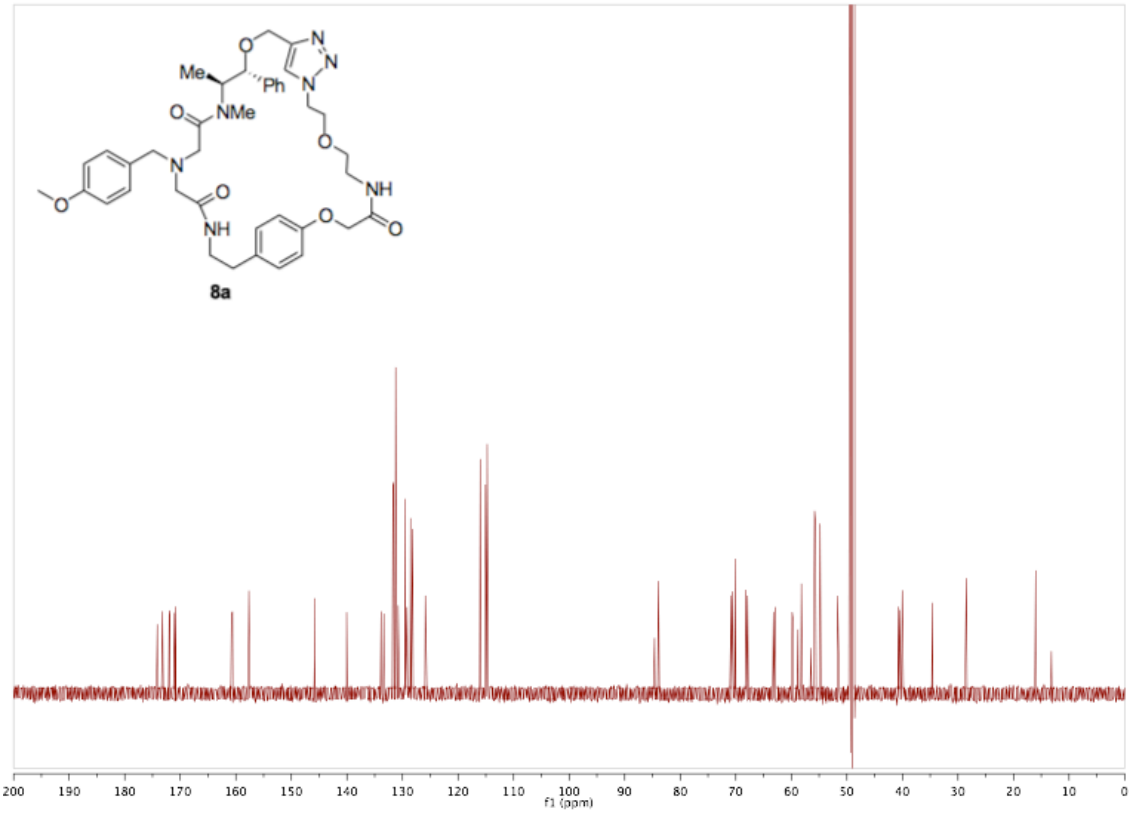


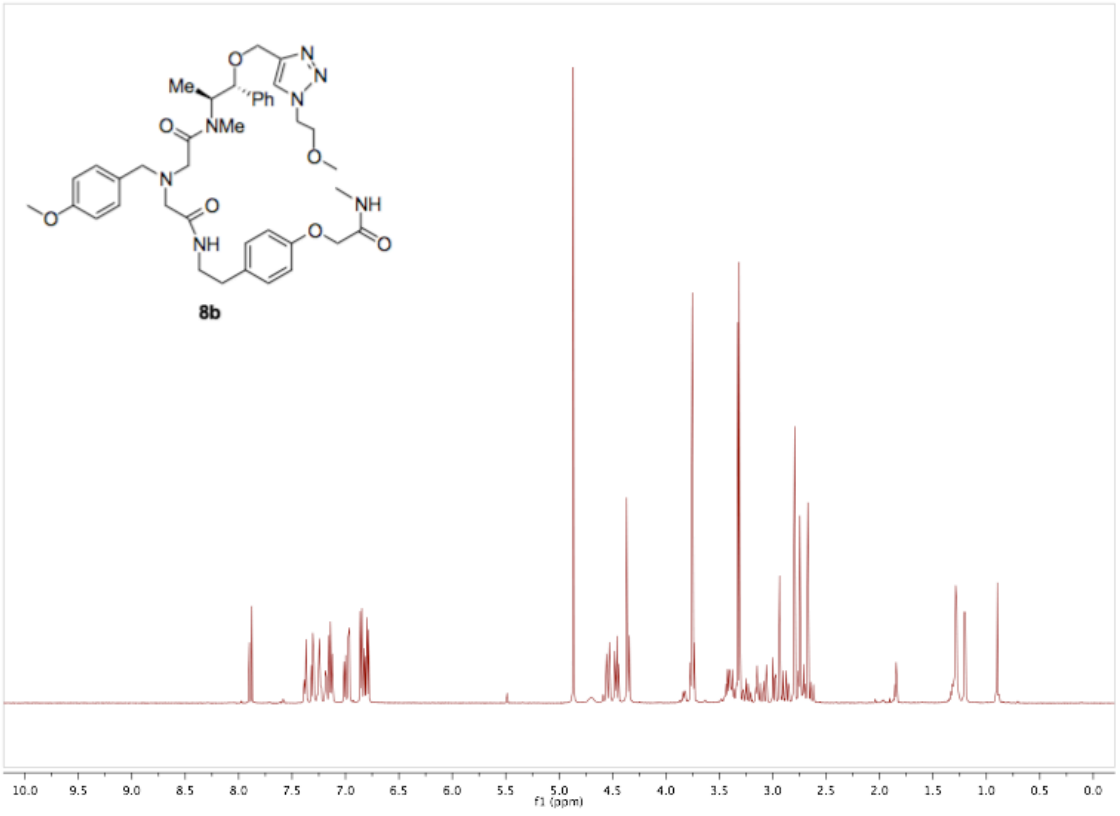


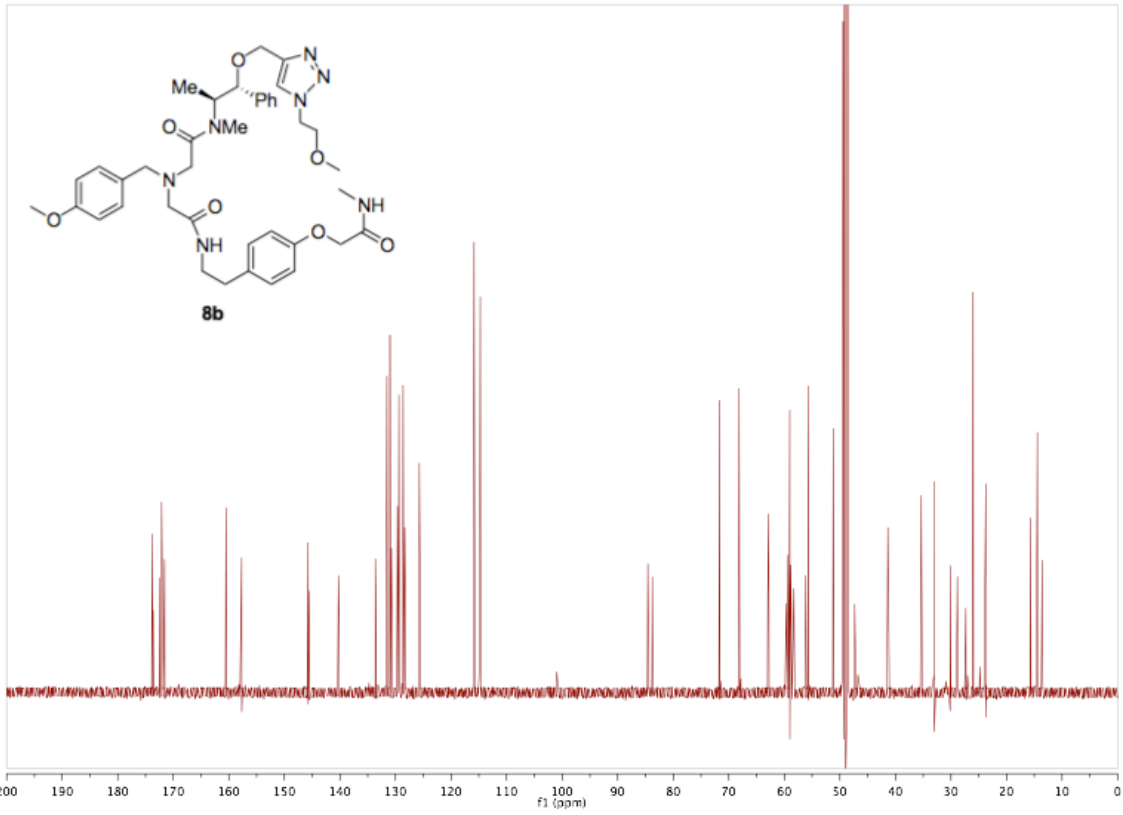




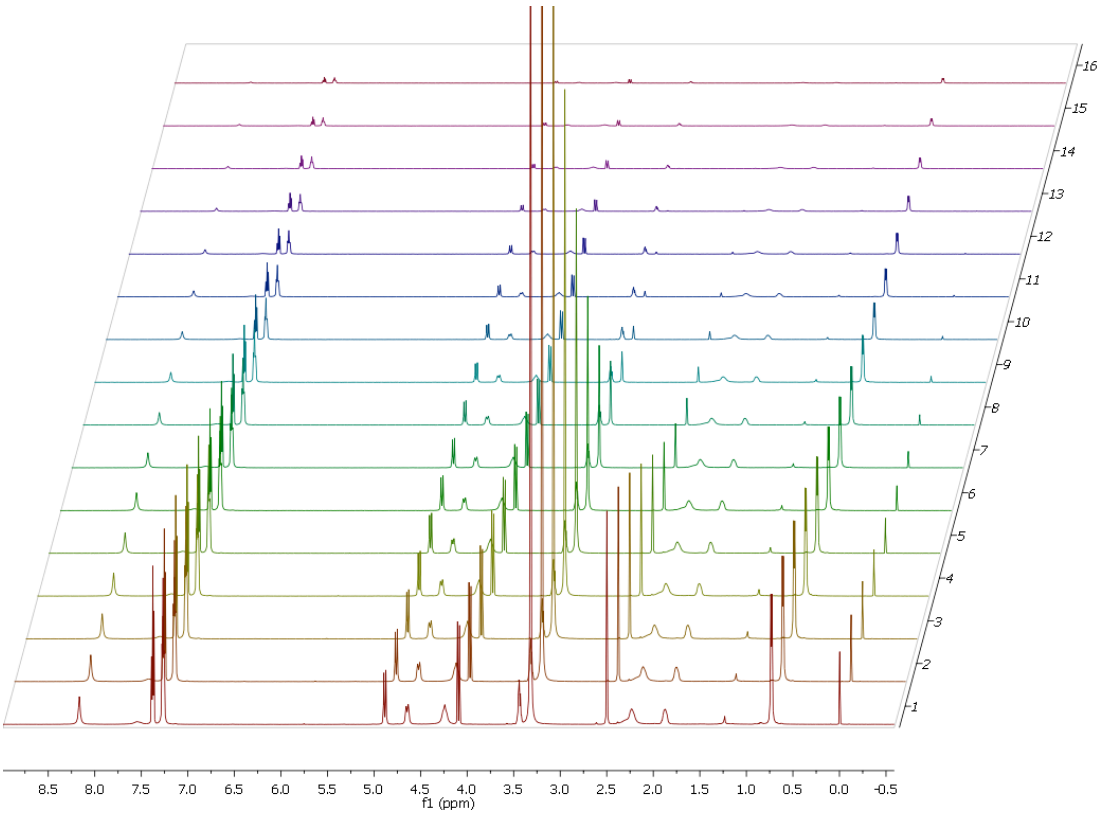








Example of arrayed NMR spectra used to calculate diffusion coefficients using T1/T2 relaxation method on Bruker Topspin, A1, 1st Run in DMSO



Example DOSY Report

SIMFIT RESULTS

==========

Dataset : C:\Bruker\TOPSPIN/data/nichola/nmr/p19562_10mMDMSO/11/pdata/1/ct1t2.txt

INTENSITY fit : Diffusion : Variable Gradient :

I=I[0]*exp(-D*SQR(2*PI*gamma*Gi*LD)*(BD-LD/3)*1e4)

14 points for Peak 1, Peak Point = 8.166 ppm

Converged after 57 iterations!

Results Comp. 1

I[0] = 1.758e-001
Diff Con. = 1.929e-010 m2/s
Gamma = 5.109e+003 Hz/G

Little Delta = 4.000m
Big Delta = 99.900m
RSS = 7.129e-007
SD = 2.257e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	1.745e-001	1.743e-001	-2.159e-004
2	3.746e+000	1.684e-001	1.683e-001	-8.899e-005
3	5.790e+000	1.583e-001	1.583e-001	2.907e-005
4	7.833e+000	1.450e-001	1.451e-001	8.559e-005
5	9.876e+000	1.295e-001	1.295e-001	3.819e-005
6	1.192e+001	1.125e-001	1.126e-001	1.323e-004
7	1.396e+001	9.491e-002	9.544e-002	5.311e-004
8	1.601e+001	7.916e-002	7.877e-002	-3.842e-004
9	1.805e+001	6.310e-002	6.333e-002	2.356e-004
10	2.009e+001	4.971e-002	4.961e-002	-1.014e-004
11	2.214e+001	3.799e-002	3.785e-002	-1.387e-004
12	2.418e+001	2.824e-002	2.813e-002	-1.098e-004
13	2.622e+001	2.069e-002	2.037e-002	-3.129e-004
14	3.031e+001	9.794e-003	9.875e-003	8.096e-005

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16 points for Peak 2, Peak Point = 7.377 ppm

Converged after 52 iterations!

Results Comp. 1

I[0] = 1.008e+000
Diff Con. = 1.936e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 4.360e-006
SD = 5.220e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	1.000e+000	9.990e-001	-9.760e-004
2	3.746e+000	9.653e-001	9.646e-001	-6.425e-004
3	5.790e+000	9.071e-001	9.073e-001	1.482e-004
4	7.833e+000	8.305e-001	8.312e-001	7.438e-004
5	9.876e+000	7.412e-001	7.418e-001	6.160e-004
6	1.192e+001	6.442e-001	6.448e-001	5.612e-004
7	1.396e+001	5.456e-001	5.460e-001	3.697e-004
8	1.601e+001	4.498e-001	4.503e-001	4.854e-004
9	1.805e+001	3.619e-001	3.617e-001	-1.846e-004
10	2.009e+001	2.832e-001	2.831e-001	-1.548e-004
11	2.214e+001	2.162e-001	2.158e-001	-3.946e-004
12	2.418e+001	1.606e-001	1.602e-001	-4.259e-004
13	2.622e+001	1.164e-001	1.159e-001	-4.959e-004
14	2.827e+001	8.180e-002	8.162e-002	-1.795e-004

15	3.031e+001	5.643e-002	5.602e-002	-4.152e-004
16	3.235e+001	3.815e-002	3.744e-002	-7.118e-004

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16 points for Peak 3, Peak Point = 7.266 ppm

Converged after 48 iterations!

Results Comp. 1

I[0] = 9.299e-001
Diff Con. = 1.931e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 4.601e-006
SD = 5.362e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	9.226e-001	9.214e-001	-1.170e-003
2	3.746e+000	8.901e-001	8.898e-001	-3.270e-004
3	5.790e+000	8.368e-001	8.370e-001	2.628e-004
4	7.833e+000	7.668e-001	7.670e-001	1.800e-004
5	9.876e+000	6.839e-001	6.847e-001	7.586e-004
6	1.192e+001	5.946e-001	5.954e-001	7.241e-004
7	1.396e+001	5.038e-001	5.043e-001	5.732e-004
8	1.601e+001	4.161e-001	4.162e-001	7.891e-005
9	1.805e+001	3.343e-001	3.345e-001	2.259e-004
10	2.009e+001	2.621e-001	2.619e-001	-2.024e-004
11	2.214e+001	2.002e-001	1.998e-001	-3.881e-004
12	2.418e+001	1.487e-001	1.485e-001	-2.844e-004
13	2.622e+001	1.081e-001	1.075e-001	-6.693e-004
14	2.827e+001	7.644e-002	7.577e-002	-6.699e-004
15	3.031e+001	5.251e-002	5.205e-002	-4.664e-004
16	3.235e+001	3.521e-002	3.482e-002	-3.886e-004

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16 points for Peak 4, Peak Point = 4.873 ppm

Converged after 54 iterations!

Results Comp. 1

I[0] = 3.462e-001
Diff Con. = 1.929e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 1.124e-006

SD = 2.650e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	3.431e-001	3.430e-001	-8.045e-005
2	3.746e+000	3.319e-001	3.313e-001	-5.973e-004
3	5.790e+000	3.116e-001	3.116e-001	-5.700e-006
4	7.833e+000	2.852e-001	2.856e-001	3.636e-004
5	9.876e+000	2.548e-001	2.550e-001	1.894e-004
6	1.192e+001	2.214e-001	2.217e-001	2.960e-004
7	1.396e+001	1.877e-001	1.879e-001	2.169e-004
8	1.601e+001	1.550e-001	1.551e-001	6.028e-005
9	1.805e+001	1.244e-001	1.247e-001	2.097e-004
10	2.009e+001	9.783e-002	9.764e-002	-1.855e-004
11	2.214e+001	7.478e-002	7.449e-002	-2.858e-004
12	2.418e+001	5.567e-002	5.537e-002	-3.035e-004
13	2.622e+001	3.999e-002	4.009e-002	1.005e-004
14	2.827e+001	2.845e-002	2.828e-002	-1.688e-004
15	3.031e+001	1.955e-002	1.943e-002	-1.159e-004
16	3.235e+001	1.339e-002	1.301e-002	-3.869e-004

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13 points for Peak 5, Peak Point = 4.632 ppm

Converged after 55 iterations!

Results Comp. 1

I[0] = 1.263e-001
Diff Con. = 1.936e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 1.078e-006
SD = 2.880e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	1.251e-001	1.251e-001	3.353e-007
2	3.746e+000	1.213e-001	1.208e-001	-4.894e-004
3	5.790e+000	1.136e-001	1.136e-001	4.851e-005
4	7.833e+000	1.037e-001	1.041e-001	3.848e-004
5	9.876e+000	9.285e-002	9.290e-002	5.040e-005
6	1.192e+001	8.058e-002	8.075e-002	1.771e-004
7	1.396e+001	6.815e-002	6.838e-002	2.319e-004
8	1.601e+001	5.685e-002	5.640e-002	-4.439e-004
9	1.805e+001	4.511e-002	4.531e-002	2.049e-004
10	2.009e+001	3.555e-002	3.546e-002	-8.694e-005
11	2.214e+001	2.698e-002	2.703e-002	4.873e-005
12	2.622e+001	1.457e-002	1.452e-002	-4.766e-005
13	3.235e+001	5.282e-003	4.691e-003	-5.909e-004

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11 points for Peak 6, Peak Point = 4.241 ppm

Converged after 56 iterations!

Results Comp. 1

I[0] = 1.242e-001
Diff Con. = 1.933e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 6.230e-007
SD = 2.380e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	1.234e-001	1.230e-001	-3.484e-004
2	3.746e+000	1.188e-001	1.188e-001	-2.943e-005
3	5.790e+000	1.119e-001	1.117e-001	-1.721e-004
4	7.833e+000	1.018e-001	1.024e-001	5.457e-004
5	9.876e+000	9.124e-002	9.138e-002	1.481e-004
6	1.192e+001	7.949e-002	7.945e-002	-4.179e-005
7	1.396e+001	6.711e-002	6.729e-002	1.792e-004
8	2.009e+001	3.512e-002	3.492e-002	-1.972e-004
9	2.418e+001	2.003e-002	1.978e-002	-2.509e-004
10	3.031e+001	6.858e-003	6.926e-003	6.734e-005
11	3.235e+001	4.737e-003	4.631e-003	-1.058e-004

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16 points for Peak 7, Peak Point = 4.103 ppm

Converged after 41 iterations!

Results Comp. 1

I[0] = 6.525e-001
Diff Con. = 1.948e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 8.784e-007
SD = 2.343e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	6.467e-001	6.465e-001	-1.438e-004
2	3.746e+000	6.240e-001	6.242e-001	1.629e-004
3	5.790e+000	5.869e-001	5.868e-001	-1.176e-004
4	7.833e+000	5.372e-001	5.373e-001	1.639e-004
5	9.876e+000	4.788e-001	4.792e-001	4.445e-004

6	1.192e+001	4.166e-001	4.162e-001	-4.161e-004
7	1.396e+001	3.521e-001	3.520e-001	-1.310e-005
8	1.601e+001	2.902e-001	2.900e-001	-2.181e-004
9	1.805e+001	2.328e-001	2.327e-001	-9.532e-005
10	2.009e+001	1.818e-001	1.818e-001	7.347e-006
11	2.214e+001	1.387e-001	1.383e-001	-3.277e-004
12	2.418e+001	1.023e-001	1.025e-001	2.081e-004
13	2.622e+001	7.372e-002	7.402e-002	3.012e-004
14	2.827e+001	5.181e-002	5.203e-002	2.256e-004
15	3.031e+001	3.536e-002	3.563e-002	2.641e-004
16	3.235e+001	2.377e-002	2.375e-002	-1.387e-005

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16 points for Peak 8, Peak Point = 3.440 ppm

Converged after 53 iterations!

Results Comp. 1

I[0] = 2.816e-001
Diff Con. = 1.950e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 4.046e-006
SD = 5.029e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	2.801e-001	2.790e-001	-1.136e-003
2	3.746e+000	2.694e-001	2.693e-001	-3.721e-005
3	5.790e+000	2.532e-001	2.532e-001	-1.310e-005
4	7.833e+000	2.313e-001	2.318e-001	5.725e-004
5	9.876e+000	2.065e-001	2.067e-001	2.031e-004
6	1.192e+001	1.787e-001	1.795e-001	7.956e-004
7	1.396e+001	1.515e-001	1.518e-001	3.363e-004
8	1.601e+001	1.250e-001	1.250e-001	5.428e-005
9	1.805e+001	9.985e-002	1.003e-001	4.485e-004
10	2.009e+001	7.862e-002	7.835e-002	-2.683e-004
11	2.214e+001	5.977e-002	5.960e-002	-1.702e-004
12	2.418e+001	4.478e-002	4.416e-002	-6.134e-004
13	2.622e+001	3.228e-002	3.187e-002	-4.098e-004
14	2.827e+001	2.264e-002	2.239e-002	-2.512e-004
15	3.031e+001	1.607e-002	1.533e-002	-7.417e-004
16	3.235e+001	1.063e-002	1.021e-002	-4.201e-004

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16 points for Peak 9, Peak Point = 0.737 ppm

Converged after 45 iterations!

Results Comp. 1

I[0] = 8.272e-001
Diff Con. = 1.931e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 3.827e-006
SD = 4.891e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	8.205e-001	8.197e-001	-7.408e-004
2	3.746e+000	7.920e-001	7.916e-001	-4.290e-004
3	5.790e+000	7.448e-001	7.446e-001	-1.824e-004
4	7.833e+000	6.818e-001	6.823e-001	5.275e-004
5	9.876e+000	6.086e-001	6.091e-001	4.582e-004
6	1.192e+001	5.288e-001	5.296e-001	7.556e-004
7	1.396e+001	4.482e-001	4.486e-001	3.923e-004
8	1.601e+001	3.702e-001	3.702e-001	-2.142e-005
9	1.805e+001	2.972e-001	2.975e-001	3.026e-004
10	2.009e+001	2.329e-001	2.330e-001	9.886e-005
11	2.214e+001	1.780e-001	1.777e-001	-3.033e-004
12	2.418e+001	1.322e-001	1.320e-001	-2.186e-004
13	2.622e+001	9.618e-002	9.556e-002	-6.145e-004
14	2.827e+001	6.803e-002	6.737e-002	-6.650e-004
15	3.031e+001	4.684e-002	4.627e-002	-5.619e-004
16	3.235e+001	3.164e-002	3.096e-002	-6.860e-004

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14 points for Peak 10, Peak Point = -0.003 ppm

Converged after 45 iterations!

Results Comp. 1

I[0] = 4.681e-001
Diff Con. = 4.742e-010 m2/s
Gamma = 5.109e+003 Hz/G
Little Delta = 4.000m
Big Delta = 99.900m

RSS = 1.939e-006
SD = 3.721e-004

Point	Gradient	Expt	Calc	Difference
1	1.703e+000	4.570e-001	4.578e-001	8.157e-004
2	3.746e+000	4.203e-001	4.202e-001	-1.778e-004
3	5.790e+000	3.622e-001	3.616e-001	-6.074e-004
4	7.833e+000	2.922e-001	2.918e-001	-4.737e-004
5	9.876e+000	2.210e-001	2.208e-001	-1.913e-004
6	1.192e+001	1.564e-001	1.566e-001	2.330e-004

7	**1.396e+001**	**1.036e-001**	**1.042e-001**	**5.946e-004**
8	**1.601e+001**	**6.515e-002**	**6.502e-002**	**-1.300e-004**
9	**1.805e+001**	**3.775e-002**	**3.802e-002**	**2.725e-004**
10	**2.009e+001**	**2.093e-002**	**2.086e-002**	**-7.532e-005**
11	**2.214e+001**	**1.055e-002**	**1.073e-002**	**1.712e-004**
12	**2.418e+001**	**4.934e-003**	**5.173e-003**	**2.391e-004**
13	**2.622e+001**	**2.196e-003**	**2.340e-003**	**1.440e-004**
14	**2.827e+001**	**9.826e-004**	**9.918e-004**	**9.209e-006**

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