Supporting Information

Aziridine electrophiles in the functionalisation of peptide chains with amine nucleophiles

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1. General Methods

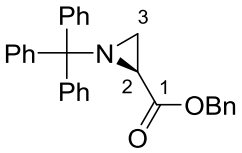
Compounds **8**[S1], **13**[S2] and **14**[S3] as well as benzyl azide[S4], 3-azido-7-hydroxycoumarine[S5] and L-alanine dibenzylamide[S6] are literature known (see references). Compound **13** and L-alanine dibenzylamide are also commercially available from Aurora Building Blocks (US) and Aldlab Chemicals Building Blocks (US). Benzyl azide is also commercially available but potentially explosive and was therefore only prepared in very small quantities.[S4] All other chemicals were purchased from standard suppliers. All solvents and reagents were used as commercially supplied without further purification unless otherwise stated. In all aziridine ring-opening reactions only free amines have been used. In those cases in which only the corresponding hydrochlorides were commercially available, the desired free amines were obtained by dissolving the corresponding hydrochlorides in AcOEt, DCM or $CHCl_3$ and washing with sat. aqueous $NaHCO_3$ solution. The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. All anhydrous reactions were carried out in flame-dried glassware and under an inert atmosphere of argon. Anhydrous solvents were dried by filtration through an activated alumina purification column or directly purchased as anhydrous solvents in sealed bottles. Petrol (PE) refers to petroleum ether in the boiling range 30-40 °C.

Flash column chromatography (FCC) was performed using Merck Kieselgel 60 (40-63 μm). Thin layer chromatography (TLC) analyses were performed on aluminum plates precoated with 0.25 mm silica gel 60 F_{254} (VWR). Visualization of the spots was carried out using UV light (254 nm) and/or staining under heating (Vanillin-H_2SO_4 staining solution: 4 g vanillin, 25 mL conc. H_2SO_4, 80 mL AcOH and 680 mL MeOH).

Melting points T_{mp} were obtained using a Leica VMTG heated-stage microscope and are uncorrected. Specific optical rotation values $[\alpha]_D^{20}$ are quoted in $°cm^3g^{-1}dm^{-1}$ and were recorded on a Perkin-Elmer polarimeter with a Na source using a 10 cm cell [concentrations c are quoted in $g(100\ mL)^{-1}$]. ^1H nuclear magnetic resonance spectra (NMR) were recorded on a Bruker AV400 (400 MHz) or Bruker AVII500 (500 MHz). ^{13}C NMR spectra were recorded on a Bruker AV400 (101 MHz) or AVII500 (126 MHz) as stated. Chemical shifts are reported relative to residual solvent peaks. Coupling constants *J* are quoted to the nearest 0.1 Hz for ^1H NMR. Chemical shifts δ are quoted in ppm (parts per million) to the nearest 0.01 ppm (^1H NMR) or 0.1 ppm (^{13}C NMR) with signal splittings recorded as singlet (s), doublet (d), triplet (t), quartet (q), multiplet (m). All NMR spectra were recorded at room temperature. Assignments were based upon DEPT, COSY, HSQC and HMBC experiments. Atoms have been numbered according to a self-consistent system used for clarity of assignment of the NMR data which does not reflect the IUPAC rules in naming compounds. Fourier transform infrared spectra (FTIR) were recorded neat on a Bruker Tensor 27 FT-IR spectrometer equipped with Attenuated Total Reflectance (ATR) sampling accessories. The nine most intense absorption maxima are quoted in wavenumbers ν $[cm^{-1}]$. Mass spectra (MS) under the conditions of electrospray ionization (ESI) were recorded on a Fisons Platform II and on a Bruker MicroTof (resolution = 10000 FWHM). Calibration was *via* the lock-mass of tetraoctyl ammonium bromide for positive ions and sodium dodecyl sulfate for negative ions.

2. Preparation of unknown compounds and key building block 9

Preparation of aziridine building block **9**



The existing protocols for the preparation of **9** from **8**[S1] were significantly improved by employing $(MeSO_2)_2O$ instead of $MeSO_2Cl$ or SO_2Cl_2 and thereby avoiding the formation of *N*-Trityl-β-chloro-L-alanine benzyl ester as side-product: Serine derivative **8**[S1] (14.0 g, 32.0 mmol) was dissolved in abs. THF (100 mL) and was cooled to 0 °C. NEt_3 (13.4 mL, 96.0 mmol) and a solution of Ms_2O (8.36 g, 48.0 mmol) in abs. THF (25 mL) were added at 0 °C. The reaction mixture was stirred for 20 min at 0 °C, for 30 min at rt and for 60 h at 60 °C. The solvent was evaporated *in vacuo*. The residue was dissolved in Et_2O (300 mL) and washed with water (200 mL), 10 wt% aqueous citric acid solution (2 x 150 mL) and sat. aqueous $NaHCO_3$ solution (2 x 150 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (250 g, 5.0 x 21 cm, PE:DCM, 50:50 → 40:60) to give 12.7 g (30.3 mmol, 95%) of the title compound as a colourless solid.

TLC: R_f = 0.39 (PE:DCM, 30:70).

Melting point: T_{mp} = 109 °C [Lit.[S1]: T_{mp} = 106-118 °C].

Specific rotation: $[\alpha]_D^{20}$ = -98.9 (c = 1.08, THF) [Lit.[S1]: $[\alpha]_D^{20}$ = -95.5-(-98.3) (c = 0.92-1.0, THF)].

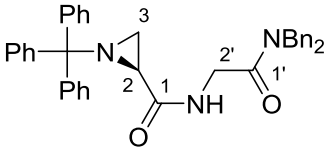
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.51-7.19 (m, 20 H, 20 x *H*C_{Ar}), 5.26 (d, *J* = 12.2 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.21 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H*$_b$), 2.30 (dd, *J* = 2.8, 1.7 Hz, 1 H, 3-*H*$_a$), 1.95 (dd, *J* = 6.2, 2.8 Hz, 1 H, 2-*H*), 1.43 (dd, *J* = 6.2, 1.7 Hz, 1 H, 3-*H*$_b$).

^{13}C NMR (101 MHz, $CDCl_3$): δ = 171.6 (*C*-1), 143.7, 136.0 (4 x *C*$_{Ar}$), 129.5, 128.7, 128.5, 128.5, 127.8, 127.1 (20 x H*C*$_{Ar}$), 74.5 (Ph_3C), 66.8 (Bn-*C*H$_2$), 31.9 (*C*-2), 29.0 (*C*-3).

IR (ATR): ν = 1729, 1447, 1234, 1171, 1015, 908, 746, 699, 631.

MS (ESI^+): m/z = 861.4 $[2M+Na]^+$, calculated: 442.1778 $[M+Na]^+$,
$C_{29}H_{25}NO_2$ (419.51 g(mol)$^{-1}$), found: 442.1774 [-0.7 ppm] (ESI^+-HRMS).

Preparation of dipeptide **11**



To aziridine building block **9** (4.00 g, 9.53 mmol) in degassed EtOH (50 mL) degassed NEt_3 (3.99 mL, 28.6 mmol) and 10 wt% palladium on charcoal (250 mg, 0.235 mmol) were added. The resulting suspension was stirred under a hydrogen atmosphere (1 bar, balloon) for 1.5 h and then filtered through a syringe filter. The syringe filter was washed with EtOH (3 x 5 mL) and the solvent of the combined filtrates evaporated *in vacuo*. After co-evaporation with toluene:THF 1:1 (2 x 30 mL) the resulting colourless solid was dried *in vacuo*. With respect to its poor stability the unprotected carboxylate was always prepared freshly and used instantly in the subsequent transformation without further purification.

HOBt (709 mg, 5.25 mmol) was added to 1/2 of the crude product (only 1/2 of the initially prepared benzyl-deprotected aziridine building block, *vide supra*, was used in the 2nd step,

calculated maximal amount of substance: 4.77 mmol) in abs. DMF (18 mL). EDAC (1.01 g, 5.25 mmol) was added after cooling the solution to 0 °C. After stirring for 10 min at 0 °C NEt_3 (0.74 mL, 5.3 mmol) was added. Glycine derivative **13**[S2] (1.34 g, 5.25 mmol) in abs. DCM (4 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 12 h and concomitantly slowly warming to rt the reaction mixture was diluted with Et_2O (300 mL) and washed with 10 wt% aqueous citric acid solution (2 x 200 mL) and sat. aqueous $NaHCO_3$ solution (2 x 200 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (150 g, 5.0 x 15 cm, PE:AcOEt, 80:20 → 70:30) to give 2.46 g (4.35 mmol, 91%) of the title compound as a colourless solid.

TLC: R_f = 0.25 (PE:AcOEt, 70:30).

Melting point: T_{mp} = 78 °C.

Specific rotation: $[\alpha]_D^{20}$ = -73.2 (c = 0.98, $CHCl_3$)

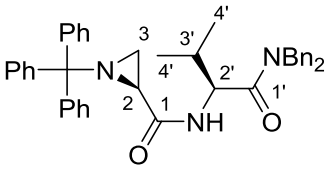
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.79 (dd, *J* = 4.5, 4.4 Hz, 1 H, 2'-N*H*), 7.54-7.20 (m, 25 H, 25 x *H*C_{Ar}), 4.78 (d, *J* = 14.8 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.64 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.51 (d, *J* = 17.4 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.46 (d, *J* = 17.4 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.35 (dd, *J* = 17.4, 4.4 Hz, 1 H, 2'-*H*$_a$), 4.26 (dd, *J* = 17.4, 4.5 Hz, 1 H, 2'-*H*$_b$), 2.16 (dd, *J* = 2.7, 0.7 Hz, 1 H, 3-*H*$_a$), 2.05 (dd, *J* = 6.6, 2.7 Hz, 1 H, 2-*H*), 1.52 (dd, *J* = 6.6, 0.7 Hz, 1 H, 3-*H*$_b$).

^{13}C NMR (101 MHz, $CDCl_3$): δ = 171.3, 168.8 (*C*-1, *C*-1'), 143.4, 136.6, 135.5 (5 x *C*$_{Ar}$), 129.6, 129.3, 128.9, 128.4, 128.1, 127.9, 127.2, 126.6 (25 x H*C*$_{Ar}$), 74.8 (Ph$_3$*C*), 49.1, 48.7 (2 x Bn-*C*H$_2$), 41.0 (*C*-2'), 34.2 (*C*-2), 29.9 (*C*-3).

IR (ATR): ν = 1648, 1494, 1448, 1221, 1010, 909, 731, 699, 632.

MS (ESI^+): m/z = 566.3 $[M+H]^+$, calculated: 588.2621 $[M+Na]^+$,
$C_{38}H_{35}N_3O_2$ (565.70 g(mol)$^{-1}$), found: 588.2593 [+4.9 ppm] (ESI^+-HRMS).

Preparation of dipeptide **12**



To aziridine building block **9** (4.00 g, 9.53 mmol) in degassed EtOH (50 mL) degassed NEt_3 (3.99 mL, 28.6 mmol) and 10 wt% palladium on charcoal (250 mg, 0.235 mmol) were added. The resulting suspension was stirred under a hydrogen atmosphere (1 bar, balloon) for 1.5 h and then filtered through a syringe filter. The syringe filter was washed with EtOH (3 x 5 mL) and the solvent of the combined filtrates evaporated *in vacuo*. After co-evaporation with toluene:THF 1:1 (2 x 30 mL) the resulting colourless solid was dried *in vacuo*. With respect to its poor stability the unprotected carboxylate was always prepared freshly and used instantly in the subsequent transformation without further purification.

HOBt (709 mg, 5.25 mmol) was added to 1/2 of the crude product (only 1/2 of the initially prepared benzyl-deprotected aziridine building block, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 4.77 mmol) in abs. DMF (18 mL). EDAC (1.01 g, 5.25 mmol) was added after cooling the solution to 0 °C. After stirring for 10 min at 0 °C NEt_3 (0.74 mL, 5.3 mmol) was added. Valine derivative **14**[S3] (1.56 g, 5.25 mmol) in abs. DCM (4 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 12 h and

concomitantly slowly warming to rt the reaction mixture was diluted with Et_2O (300 mL) and washed with 10 wt% aqueous citric acid solution (2 x 200 mL) and sat. aqueous $NaHCO_3$ solution (2 x 200 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (150 g, 5.0 x 15 cm, PE:AcOEt, 85:15 → 80:20) to give 2.61 g (4.29 mmol, 90%) of the title compound as a colourless solid.

TLC: R_f = 0.29 (PE:AcOEt, 80:20).

Melting point: T_{mp} = 74 °C.

Specific rotation: $[\alpha]_D^{20}$ = -109.3 (c = 1.03, $CHCl_3$)

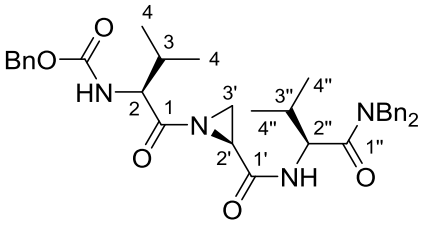
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.62 (d, *J* = 9.2 Hz, 1 H, 2'-N*H*), 7.53-7.21 (m, 25 H, 25 x *H*C_{Ar}), 5.04 (dd, *J* = 9.2, 6.2 Hz, 1 H, 2'-*H*), 4.98 (d, *J* = 14.6 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.73 (d, *J* = 16.5 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.49 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.34 (d, *J* = 14.6 Hz, 1 H, Bn-CH$_a$*H*$_b$), 2.20 (dqq, *J* = 6.7, 6.7, 6.2 Hz, 1 H, 3'-*H*), 2.10 (d, *J* = 2.4 Hz, 1 H, 3-*H*$_a$), 2.04 (dd, *J* = 6.5, 2.4 Hz, 1 H, 2-*H*), 1.50 (d, *J* = 6.5 Hz, 1 H, 3-*H*$_b$), 1.02 (d, *J* = 6.7 Hz, 3 H, 4'-*H*$_a$), 0.97 (d, *J* = 6.7 Hz, 3 H, 4'-*H*$_b$).

^{13}C NMR (101 MHz, $CDCl_3$): δ = 172.2, 170.8 (*C*-1, *C*-1'), 143.5, 137.1, 136 (5 x *C*$_{Ar}$), 129.6, 129.1, 128.9, 128.5, 128.0, 127.9, 127.7, 127.3, 127.2 (25 x H*C*$_{Ar}$), 74.8 (Ph$_3$*C*), 53.2 (C-2'), 50.0, 47.9 (2 x Bn-*C*H$_2$), 33.9 (*C*-2), 32.2 (*C*-3'), 30.3 (*C*-3), 19.9, 17.6 (2 x *C*-4').

IR (ATR): ν = 1641, 1495, 1447, 1215, 1011, 909, 732, 705, 633.

MS (ESI$^+$): m/z = 608.3 $[M+H]^+$, calculated: 630.3091 $[M+Na]^+$,
$C_{41}H_{41}N_3O_2$ (607.78 g(mol)$^{-1}$), found: 630.3068 [+3.7 ppm] (ESI$^+$-HRMS).

Preparation of tripeptide **4**



To a solution of dipeptide **12** (2.30 g, 3.78 mmol) in abs. MeOH (10 mL) and abs. $CHCl_3$ (10 mL) precooled to 0 °C, TFA (2.0 mL) was added dropwise. The solution was stirred for 3.5 h at 0 °C, diluted with AcOEt (150 mL) at 0 °C and washed with a preformed mixture of 1 M aqueous NaOH solution (100 mL) and sat. aqueous NaCl solution (100 mL), which was also cooled to 0 °C beforehand. The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (100 g, 4.5 x 14 cm, DCM:MeOH, 96:4 [R_f = 0.21 (DCM:MeOH, 95:5)]). With respect to its poor stability the trityl-deprotected dipeptide was always prepared freshly and used in the subsequent transformation without any delay.

HOBt (92 mg, 0.68 mmol) was added to *N*-Carbobenzoxy-L-valine (172 mg, 0.684 mmol) in abs. DMF (2.5 mL). EDAC (131 mg, 0.684 mmol) was added after cooling the solution to 0 °C. After stirring for 10 min at 0 °C NEt_3 (95 µL, 0.68 mmol) was added. 1/6 of the crude product (only 1/6 of the initially prepared trityl-deprotected dipeptide, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 0.630 mmol) in abs. DMF (1.5 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 16 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (150 mL) and washed with water (100 mL), 10 wt% aqueous citric acid solution (100 mL) and sat. aqueous $NaHCO_3$

solution (100 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (50 g, 4.5 x 7.0 cm, PE:AcOEt, 75:25 → 65:35) to give 256 mg (0.428 mmol, 68%) of the title compound as a colourless solid.

TLC: R_f = 0.24 (PE:AcOEt, 60:40).

Melting point: T_{mp} = 47 °C.

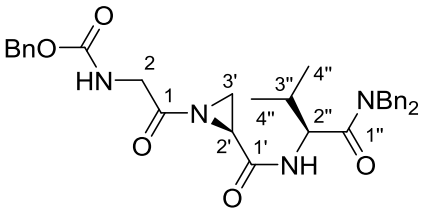
Specific rotation: $[\alpha]_D^{20}$ = -99.0 (c = 1.23, $CHCl_3$)

^{1}H NMR (400 MHz, D_6-DMSO): δ = 8.88 (d, *J* = 8.8 Hz, 1 H, 2"-N*H*), 7.45 (d, *J* = 8.5 Hz, 1 H, 2-N*H*), 7.37-7.16 (m, 15 H, 15 x *H*C_{Ar}), 5.02 (d, *J* = 16.9 Hz, 1 H, Bn-C$H_a$$H_b$), 4.99 (d, *J* = 16.9 Hz, 1 H, Bn-C$H_a$$H_b$), 4.69-4.55 (m, 4 H, 2"-*H*, Bn-C$H_a$$H_b$, Bn-C$H_2$), 4.33 (d, *J* = 15.0 Hz, 1 H, Bn-C$H_a$$H_b$), 4.02 (dd, *J* = 8.5, 5.3 Hz, 1 H, 2-*H*), 3.34-3.30 (m, 1 H, 2'-*H*), 2.55 (dd, *J* = 5.4, 2.2 Hz, 1 H, 3'-H_a), 2.28 (dd, *J* = 2.6, 2.2 Hz, 1 H, 3'-H_b), 2.18-2.04 (m, 2 H, 3-*H*, 3"-*H*), 0.90-0.88 (m, 9 H, 4-*H*, 4"-H_a), 0.76 (d, *J* = 6.7 Hz, 3 H, 4"-H_b).

^{13}C NMR (101 MHz, D_6-DMSO): δ = 181.7 (*C*-1), 171.4 (*C*-1"), 166.3 (*C*-1'), 156.2 (N*C*(=O)O), 137.3, 137.1, 137.0 (3 x C_{Ar}), 128.6, 128.5, 128.3, 127.8, 127.6, 127.5, 127.4, 127.1 (15 x HC_{Ar}), 65.4 (Bn-*C*H_2), 60.9 (*C*-2), 54.2 (*C*-2"), 49.9, 48.0 (2 x Bn-*C*H_2), 35.1 (*C*-2'), 30.1 (*C*-3, *C*-3"), 29.6 (*C*-3'), 19.3, 19.3, 17.9, 17.6 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 1698, 1629, 1497, 1451, 1220, 1027, 909, 729, 698.

MS (ESI^+): m/z = 599.3 $[M+H]^+$, calculated: 621.3047 $[M+Na]^+$,
$C_{35}H_{42}N_4O_5$ (598.73 g(mol)$^{-1}$), found: 621.3056 [-1.3 ppm] (ESI^+-HRMS).



Preparation of tripeptide **5**

To a solution of dipeptide **12** (2.30 g, 3.78 mmol) in abs. MeOH (10 mL) and abs. $CHCl_3$ (10 mL) precooled to 0 °C, TFA (2.0 mL) was added dropwise. The solution was stirred for 3.5 h at 0 °C, diluted with AcOEt (150 mL) at 0 °C and washed with a preformed mixture of 1 M aqueous NaOH solution (100 mL) and sat. aqueous NaCl solution (100 mL), which was also cooled to 0 °C beforehand. The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (100 g, 4.5 x 14 cm, DCM:MeOH, 96:4 [R_f = 0.21 (DCM:MeOH, 95:5)]). With respect to its poor stability the trityl-deprotected dipeptide was always prepared freshly and used in the subsequent transformation without any delay.

HOBt (92 mg, 0.68 mmol) was added to *N*-Carbobenzoxy-glycine (143 mg, 0.684 mmol) in abs. DMF (2.5 mL). EDAC (131 mg, 0.684 mmol) was added after cooling the solution to 0 °C. After stirring for 10 min at 0 °C NEt_3 (95 µL, 0.68 mmol) was added. 1/6 of the crude product (only 1/6 of the initially prepared trityl-deprotected dipeptide, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 0.630 mmol) in abs. DMF (1.5 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 16 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (150 mL) and washed with water (100 mL), 10 wt% aqueous citric acid solution (100 mL) and sat. aqueous $NaHCO_3$ solution (100 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the

filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (50 g, 4.5 x 7.0 cm, PE:AcOEt, 45:55 → 40:60) to give 223 mg (0.401 mmol, 64%) of the title compound as a colourless solid.

TLC: R_f = 0.26 (PE:AcOEt, 40:60).

Melting point: T_{mp} = 48 °C.

Specific rotation: $[\alpha]_D^{20}$ = -95.7 (c = 1.09, $CHCl_3$)

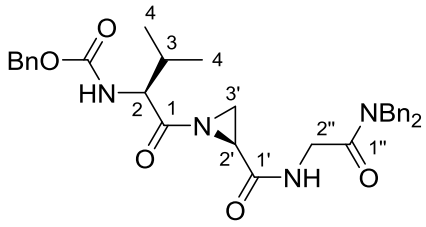
^{1}H NMR (400 MHz, D_6-DMSO): δ = 8.96 (d, *J* = 9.0 Hz, 1 H, 2"-N*H*), 7.57 (dd, *J* = 6.1, 6.0 Hz, 1 H, 2-N*H*), 7.38-7.17 (m, 15 H, 15 x *H*C_{Ar}), 5.03 (s, 2 H, Bn-C*H*$_2$), 4.70-4.62 (m, 3 H, 2"-*H*, 2 x Bn-C*H*$_a$H$_b$), 4.55 (d, *J* = 16.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.33 (d, *J* = 15.0 Hz, 1 H, Bn-CH$_a$*H*$_b$), 3.84 (dd, *J* = 17.4, 6.1 Hz, 1 H, 2-*H*$_a$), 3.70 (dd, *J* = 17.4, 6.0 Hz, 1 H, 2-*H*$_b$), 3.35-3.33 (m, 1 H, 2'-*H*), 2.44 (dd, *J* = 5.3, 1.9 Hz, 1 H, 3'-*H*$_a$), 2.23 (dd, *J* = 2.8, 1.9 Hz, 1 H, 3'-*H*$_b$), 2.09 (dqq, *J* = 7.5, 6.8, 6.7 Hz, 1 H, 3"-*H*), 0.87 (d, *J* = 6.8 Hz, 3 H, 4"-*H*$_a$), 0.76 (d, *J* = 6.7 Hz, 3 H, 4"-*H*$_b$).

^{13}C NMR (101 MHz, D_6-DMSO): δ = 179.8 (*C*-1), 171.3 (*C*-1"), 166.3 (*C*-1'), 156.4 (N*C*(=O)O), 137.3, 137.1, 137.0 (3 x *C*$_{Ar}$), 128.6, 128.5, 128.4, 127.8, 127.7, 127.5, 127.4, 127.2, 127.0 (15 x H*C*$_{Ar}$), 65.5 (Bn-*C*H$_2$), 54.2 (*C*-2"), 49.9, 48.0 (2 x Bn-*C*H$_2$), 44.5 (*C*-2), 34.8 (*C*-2'), 30.2 (*C*-3"), 28.9 (*C*-3'), 19.3, 17.9 (2 x *C*-4").

IR (ATR): ν = 1707, 1632, 1524, 1451, 1249, 1167, 1049, 732, 699.

MS (ESI^+): m/z = 579.3 $[M+Na]^+$, calculated: 579.2578 $[M+Na]^+$,
$C_{32}H_{36}N_4O_5$ (556.65 g(mol)$^{-1}$), found: 579.2583 [-0.8 ppm] (ESI^+-HRMS).

Preparation of tripeptide **6**



To a solution of dipeptide **11** (2.25 g, 3.98 mmol) in abs. MeOH (10 mL) and abs. $CHCl_3$ (10 mL) precooled to 0 °C, TFA (2.0 mL) was added dropwise. The solution was stirred for 3.5 h at 0 °C, diluted with AcOEt (200 mL) at 0 °C and washed with a preformed mixture of 1 M aqueous NaOH solution (100 mL) and sat. aqueous NaCl solution (100 mL), which was also cooled to 0 °C beforehand. The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (100 g, 5.0 x 9.0 cm, DCM:MeOH, 95:5 [R_f = 0.14 (DCM:MeOH, 94:6)]). With respect to its poor stability the trityl-deprotected dipeptide was always prepared freshly and used in the subsequent transformation without any delay.

HOBt (104 mg, 0.773 mmol) was added to *N*-Carbobenzoxy-L-valine (194 mg, 0.773 mmol) in abs. DMF (3 mL). EDAC (148 mg, 0.773 mmol) was added after cooling the solution to 0 °C. After stirring for 10 min at 0 °C NEt_3 (0.11 mL, 0.77 mmol) was added. 1/6 of the crude product (only 1/6 of the initially prepared trityl-deprotected dipeptide, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 0.663 mmol) in abs. DMF (1.5 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 16 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (150 mL) and washed with water (100 mL), 10 wt% aqueous citric acid solution (100 mL) and sat. aqueous $NaHCO_3$ solution (100 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the

filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (50 g, 4.5 x 7.0 cm, PE:AcOEt, 50:50) to give 261 mg (0.469 mmol, 71%) of the title compound as a colourless solid.

TLC: R_f = 0.33 (PE:AcOEt, 40:60).

Melting point: T_{mp} = 51 °C.

Specific rotation: $[\alpha]_D^{20}$ = -62.1 (c = 1.01, $CHCl_3$)

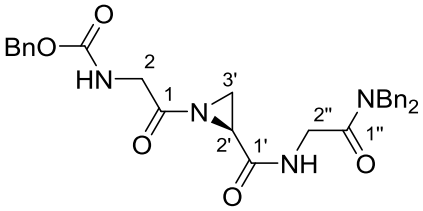
^{1}H NMR (400 MHz, D_6-DMSO): δ = 8.55 (t, *J* = 5.5 Hz, 1 H, 2''-N*H*), 7.54 (d, *J* = 8.3 Hz, 1 H, 2-N*H*), 7.39-7.21 (m, 15 H, 15 x *H*C_{Ar}), 5.02 (d, *J* = 12.6 Hz, 1 H, Bn-C$H_a$$H_b$), 4.97 (d, *J* = 12.6 Hz, 1 H, Bn-C$H_a$$H_b$), 4.54-4.45 (s, 4 H, 2 x Bn-CH_2), 4.09 (d, *J* = 5.5 Hz, 2 H, 2''-*H*), 4.03 (dd, *J* = 8.3, 5.9 Hz, 1 H, 2-*H*), 3.23 (dd, *J* = 5.7, 3.1 Hz, 1 H, 2'-*H*), 2.62 (dd, *J* = 5.7, 2.0 Hz, 1 H, 3'-H_a), 2.38 (dd, *J* = 3.1, 2.0 Hz, 1 H, 3'-H_b), 2.15 (dqq, *J* = 6.8, 6.8, 5.9 Hz, 1 H, 3-*H*), 0.92 (d, *J* = 6.8 Hz, 3 H, 4-H_a), 0.89 (d, *J* = 6.8 Hz, 3 H, 4-H_b).

^{13}C NMR (101 MHz, D_6-DMSO): δ = 182.5 (*C*-1), 168.6 (*C*-1''), 166.8 (*C*-1'), 156.4 (N*C*(=O)O), 137.3, 137.0, 136.7 (3 x C_{Ar}), 128.8, 128.4, 128.3, 127.8, 127.7, 127.7, 127.4, 127.1, 126.6 (15 x HC_{Ar}), 65.5 (Bn-*C*H_2), 61.0 (*C*-2), 49.2, 48.5 (2 x Bn-*C*H_2), 40.7 (*C*-2''), 35.3 (*C*-2'), 29.9 (*C*-3, *C*-3'), 19.4, 17.8 (2 x *C*-4).

IR (ATR): ν = 1700, 1647, 1525, 1453, 1224, 1081, 1028, 732, 699.

MS (ESI^+): m/z = 579.3 $[M+Na]^+$, calculated: 579.2578 $[M+Na]^+$,
$C_{32}H_{36}N_4O_5$ (556.65 g(mol)$^{-1}$), found: 579.2572 [+1.1 ppm] (ESI^+-HRMS).

Preparation of tripeptide **7**



To a solution of dipeptide **11** (2.25 g, 3.98 mmol) in abs. MeOH (10 mL) and abs. $CHCl_3$ (10 mL) precooled to 0 °C, TFA (2.0 mL) was added dropwise. The solution was stirred for 3.5 h at 0 °C, diluted with AcOEt (200 mL) at 0 °C and washed with a preformed mixture of 1 M aqueous NaOH solution (100 mL) and sat. aqueous NaCl solution (100 mL), which was also cooled to 0 °C beforehand. The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (100 g, 5.0 x 9.0 cm, DCM:MeOH, 95:5 [R_f = 0.14 (DCM:MeOH, 94:6)]). With respect to its poor stability the trityl-deprotected dipeptide was always prepared freshly and used in the subsequent transformation without any delay.

HOBt (104 mg, 0.773 mmol) was added to *N*-Carbobenzoxy-glycine (162 mg, 0.773 mmol) in abs. DMF (3 mL). EDAC (148 mg, 0.773 mmol) was added after cooling the solution to 0 °C. After stirring for 10 min at 0 °C NEt_3 (0.11 mL, 0.77 mmol) was added. 1/6 of the crude product (only 1/6 of the initially prepared trityl-deprotected dipeptide, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 0.663 mmol) in abs. DMF (1.5 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 16 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (150 mL) and washed with water (100 mL), 10 wt% aqueous citric acid solution (100 mL) and sat. aqueous $NaHCO_3$ solution (100 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (50 g,

4.5 x 7.0 cm, PE:AcOEt, 40:60 → 25:75) to give 217 mg (0.422 mmol, 63%) of the title compound as a colourless solid.

TLC: R_f = 0.13 (PE:AcOEt, 30:70).

Melting point: T_{mp} = 62 °C.

Specific rotation: $[\alpha]_D^{20}$ = -56.9 (c = 0.81, $CHCl_3$)

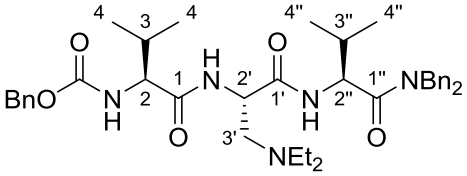
^{1}H NMR (400 MHz, D_6-DMSO): δ = 8.72 (t, *J* = 5.5 Hz, 1 H, 2''-N*H*), 7.61 (t, *J* = 6.1 Hz, 1 H, 2-N*H*), 7.39-7.22 (m, 15 H, 15 x *H*C_{Ar}), 5.03 (s, 2 H, Bn-C*H*$_2$), 4.53 (s, 2 H, Bn-C*H*$_2$), 4.50 (s, 2 H, Bn-C*H*$_2$), 4.10 (d, *J* = 5.5 Hz, 2 H, 2''-*H*), 3.78 (d, *J* = 6.1 Hz, 2 H, 2-*H*), 3.31 (dd, *J* = 5.5, 3.0 Hz, 1 H, 2'-*H*), 2.47 (dd, *J* = 5.5, 1.8 Hz, 1 H, 3'-H_a), 2.30 (dd, *J* = 3.0, 1.8 Hz, 1 H, 3'-H_b).

^{13}C NMR (101 MHz, D_6-DMSO): δ = 180.4 (*C*-1), 168.7 (*C*-1''), 166.9 (*C*-1'), 156.4 (N*C*(=O)O), 137.3, 137.0, 136.7 (3 x C_{Ar}), 128.8, 128.4, 128.3, 127.8, 127.7, 127.4, 127.1, 126.6 (15 x HC_{Ar}), 65.5, 49.2, 48.6 (3 x Bn-*C*H_2), 44.7 (*C*-2), 40.8 (*C*-2''), 35.2 (*C*-2'), 28.9 (*C*-3').

IR (ATR): ν = 3317, 1647, 1527, 1452, 1251, 1168, 1048, 733, 698.

MS (ESI^+): m/z = 537.3 $[M+Na]^+$, calculated: 537.2108 $[M+Na]^+$,
$C_{29}H_{30}N_4O_5$ (514.57 g(mol)$^{-1}$), found: 537.2086 [+4.1 ppm] (ESI^+-HRMS).

Preparation of tripeptide **15**



To a solution of tripeptide **4** (15 mg, 0.025 mmol) in $CHCl_3$ (1.5 mL), diethylamine (16 µL, 0.15 mmol) was added. After stirring for 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 60:40:0 → 98:0:2) to give 16 mg (0.024 mmol, 96%) of the title compound as a colourless solid.

TLC: R_f = 0.14 (DCM:MeOH, 96:4).

Melting point: T_{mp} = 106 °C.

Specific rotation: $[\alpha]_D^{20}$ = -3.5 (c = 1.34, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 9.17 (d, *J* = 8.7 Hz, 1 H, 2''-N*H*), 7.31-7.09 (m, 15 H, 15 x *H*C_{Ar}), 6.77 (d, *J* = 4.9 Hz, 1 H, 2'-N*H*), 5.34 (d, *J* = 8.6 Hz, 1 H, 2-N*H*), 5.06 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 5.02 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 4.79-4.76 (m, 2 H, 2''-*H*, Bn-C$H_a$$H_b$), 4.55 (d, *J* = 16.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.42 (d, *J* = 16.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.33-4.28 (m, 1 H, 2'-*H*), 4.22 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.02 (dd, *J* = 8.6, 5.8 Hz, 1 H, 2-*H*), 2.81-2.74 (m, 3 H, 3'-H_a, 2 x C$H_a$$H_b$$CH_3$), 2.65-2.58 (m, 2 H, 2 x C$H_a$$H_b$$CH_3$), 2.46 (dd, *J* = 11.7, 11.7 Hz, 1 H, 3'-H_b), 2.09-1.98 (m, 2 H, 3-*H*, 3''-*H*), 1.00 (dd, *J* = 7.2, 7.1 Hz, 6 H, 2 x CH_2CH_3), [0.90 (d, *J* = 6.8 Hz, 3 H), 0.89 (d, *J* = 6.9 Hz, 3 H), 0.86 (d, *J* = 6.8 Hz, 3 H), 0.83 (d, *J* = 6.8 Hz, 3 H) (4-H_a, 4-H_b, 4''-H_a, 4''-H_b)].

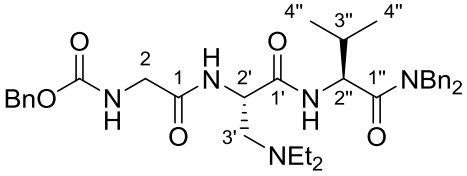
^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.0, 171.3, 171.2 (*C*-1, *C*-1', *C*-1''), 156.4 (N*C*(=O)O), 137.2, 136.5, 136.4 (3 x C_{Ar}), 129.0, 128.7, 128.7, 128.4, 128.3, 128.2, 128.0, 127.6, 127.4 (15 x HC_{Ar}), 67.1 (Bn-*C*H_2), 60.3 (*C*-2), 55.5 (*C*-3'), 54.7 (*C*-2''), 50.1 (Bn-*C*H_2), 49.7 (*C*-2'),

48.0 (Bn-*C*H_2), 46.1 (2 x *C*H_2CH$_3$), 31.7, 31.1 (*C*-3, *C*-3''), 20.1, 19.3, 17.8, 17.5 (2 x *C*-4, 2 x *C*-4''), 11.2 (2 x CH$_2$*C*H$_3$).

IR (ATR): ν = 3292, 2964, 1633, 1534, 1448, 1233, 1028, 733, 697.

MS (ESI$^+$): m/z = 672.5 $[M+H]^+$, calculated: 672.4120 $[M+H]^+$,
$C_{39}H_{53}N_5O_5$ (671.40 g(mol)$^{-1}$), found: 672.4102 [-2.7 ppm] (ESI$^+$-HRMS).

Preparation of tripeptide **16**



To a solution of tripeptide **5** (15 mg, 0.027 mmol) in $CHCl_3$ (1.5 mL), diethylamine (17 µL, 0.16 mmol) was added. After stirring for 20 h at rt and for 24 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:MeOH, 93:7) to give 16 mg (0.025 mmol, 94%) of the title compound as a colourless solid.

TLC: R_f = 0.17 (DCM:MeOH, 95:5).

Melting point: T_{mp} = 159 °C.

Specific rotation: $[\alpha]_D^{20}$ = -6.8 (c = 1.21, $CHCl_3$)

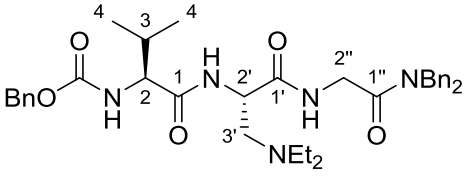
^{1}H NMR (500 MHz, $CDCl_3$): δ = 9.17 (s, 1 H, 2''-N*H*), 7.31-7.09 (m, 15 H, 15 x *H*C$_{Ar}$), 6.83 (s, 1 H, 2'-N*H*), 5.38 (s, 1 H, 2-N*H*), 5.08 (d, *J* = 12.5 Hz, 1 H, Bn-C*H$_a$*H$_b$), 5.05 (d, *J* = 12.5 Hz, 1 H, Bn-CH$_a$*H$_b$*), 4.81 (d, *J* = 14.8 Hz, 1 H, Bn-C*H$_a$*H$_b$), 4.77 (dd, *J* = 8.9, 5.8 Hz, 1 H, 2''-*H*), 4.56 (d, *J* = 16.5 Hz, 1 H, Bn-C*H$_a$*H$_b$), 4.40-4.32 (m, 2 H, 2'-*H*, Bn-CH$_a$*H$_b$*), 4.18 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H$_b$*), 3.92-3.79 (m, 2 H, 2-*H*), 2.81-2.74 (m, 3 H, 3'-*H$_a$*, 2 x C*H$_a$*H$_b$CH$_3$), 2.67-2.59 (m, 2 H, 2 x CH$_a$*H$_b$*CH$_3$), 2.54-2.41 (m, 1 H, 3'-*H$_b$*), 2.02 (dqq, *J* = 6.7, 6.7, 5.8 Hz, 1 H, 3''-*H*), 1.01 (dd, *J* = 7.0, 7.0 Hz, 6 H, 2 x CH$_2$C*H$_3$*), 0.89 (d, *J* = 6.7 Hz, 3 H, 4''-*H$_a$*), 0.83 (d, *J* = 6.7 Hz, 3 H, 4''-*H$_b$*).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.0, 171.3, 168.9 (*C*-1, *C*-1', *C*-1''), 156.5 (N*C*(=O)O), 137.1, 136.4, 136.4 (3 x *C$_{Ar}$*), 129.0, 128.8, 128.7, 128.4, 128.3, 128.2, 128.0, 127.6, 127.4 (15 x H*C$_{Ar}$*), 67.3 (Bn-*C*H$_2$), 55.4 (*C*-3'), 54.8 (*C*-2''), 50.1 (Bn-*C*H$_2$), 49.7 (*C*-2'), 48.0 (Bn-*C*H$_2$), 46.1 (2 x *C*H$_2$CH$_3$), 44.5 (*C*-2), 31.1 (*C*-3''), 20.1, 17.5 (2 x *C*-4''), 11.1 (2 x CH$_2$*C*H$_3$).

IR (ATR): ν = 3277, 1714, 1637, 1539, 1446, 1246, 1044, 738, 696.

MS (ESI$^+$): m/z = 630.4 $[M+H]^+$, calculated: 630.3650 $[M+H]^+$,
$C_{36}H_{47}N_5O_5$ (629.80 g(mol)$^{-1}$), found: 630.3628 [-3.5 ppm] (ESI$^+$-HRMS).

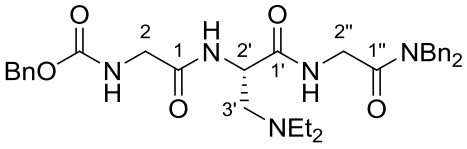
Preparation of tripeptide **17**



To a solution of tripeptide **6** (15 mg, 0.027 mmol) in $CHCl_3$ (1.5 mL), diethylamine (17 µL, 0.16 mmol) was added. After stirring for 20 h at rt and for 24 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:MeOH, 97:3) to give 16 mg (0.025 mmol, 94%) of the title compound as a colourless solid.

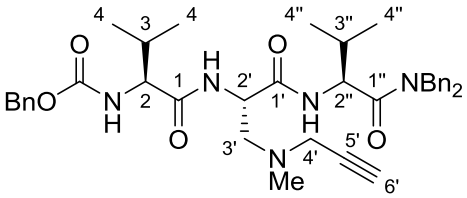
TLC: R_f = 0.25 (DCM:MeOH, 95:5).
Melting point: T_{mp} = 105 °C.
Specific rotation: $[\alpha]_D^{20}$ = +19.4 (c = 1.27, $CHCl_3$)
^{1}H NMR (500 MHz, $CDCl_3$): δ = 9.07 (s, 1 H, 2''-N*H*), 7.40-7.14 (m, 15 H, 15 x *H*C_{Ar}), 7.01 (s, 1 H, 2'-N*H*), 5.46 (d, *J* = 8.1 Hz, 1 H, 2-N*H*), 5.13 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 5.09 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 4.69 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.62 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.46-4.38 (m, 3 H, 2'-*H*, Bn-CH_2), 4.28-4.19 (m, 2 H, 2''-*H*), 4.14 (dd, *J* = 8.1, 6.2 Hz, 1 H, 2-*H*), 2.85-2.81 (m, 3 H, 3'-H_a, 2 x C$H_a$$H_b$$CH_3$), 2.65-2.56 (m, 3 H, 3'-H_b, 2 x C$H_a$$H_b$$CH_3$), 2.46 (dqq, *J* = 6.7, 6.6, 6.2 Hz, 1 H, 3-*H*), 1.12 (dd, *J* = 7.1, 7.0 Hz, 6 H, 2 x CH_2CH_3), 0.99 (d, *J* = 6.6 Hz, 3 H, 4-H_a), 0.95 (d, *J* = 6.7 Hz, 3 H, 4-H_b).
^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.2, 168.4 (*C*-1, *C*-1', *C*-1''), 156.4 (N*C*(=O)O), 136.7, 136.5, 135.6 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.4, 128.2, 128.1, 128.0, 127.8, 126.5 (15 x HC_{Ar}), 67.0 (Bn-*C*H_2), 60.2 (*C*-2), 55.2 (*C*-3'), 50.3 (*C*-2'), 49.1, 48.5 (2 x Bn-*C*H_2), 46.9 (2 x *C*$H_2$$CH_3$), 41.8 (*C*-2''), 31.7 (*C*-3), 19.3, 17.8 (2 x *C*-4), 11.7 (2 x CH_2*C*H_3).
IR (ATR): ν = 3287, 1719, 1630, 1523, 1222, 1028, 733, 697, 646.
MS (ESI^+): m/z = 630.4 $[M+H]^+$, calculated: 630.3650 $[M+H]^+$,
$C_{36}H_{47}N_5O_5$ (629.80 g(mol)$^{-1}$), found: 630.3630 [-3.2 ppm] (ESI^+-HRMS).

Preparation of tripeptide **18**



To a solution of tripeptide **7** (14 mg, 0.027 mmol) in $CHCl_3$ (1.5 mL), diethylamine (17 µL, 0.16 mmol) was added. After stirring for 18 h at rt and for 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:MeOH, 96:4) to give 14 mg (0.026 mmol, 96%) of the title compound as a colourless solid.
TLC: R_f = 0.18 (DCM:MeOH, 95:5).
Melting point: T_{mp} = 78 °C.
Specific rotation: $[\alpha]_D^{20}$ = +16.6 (c = 1.18, $CHCl_3$)
^{1}H NMR (500 MHz, $CDCl_3$): δ = 9.00 (s, 1 H, 2''-N*H*), 7.40-7.07 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.59 (s, 1 H, 2-N*H*), 5.15 (d, *J* = 12.5 Hz, 1 H, Bn-C$H_a$$H_b$), 5.12 (d, *J* = 12.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.66 (d, *J* = 15.2 Hz, 1 H, Bn-C$H_a$$H_b$), 4.63 (d, *J* = 15.2 Hz, 1 H, Bn-C$H_a$$H_b$), 4.47-4.39 (m, 3 H, 2'-*H*, Bn-CH_2), 4.25-4.16 (m, 2 H, 2''-*H*), 3.99 (dd, *J* = 16.9, 5.4 Hz, 1 H, 2-H_a), 3.91 (dd, *J* = 16.9, 5.4 Hz, 1 H, 2-H_b), 2.88-2.78 (m, 3 H, 3'-H_a, 2 x C$H_a$$H_b$$CH_3$), 2.67-2.58 (m, 3 H, 3'-H_b, 2 x C$H_a$$H_b$$CH_3$), 1.11 (dd, *J* = 7.1, 7.1 Hz, 6 H, 2 x CH_2CH_3).
^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.2, 169.0, 168.4 (*C*-1, *C*-1', *C*-1''), 156.6 (N*C*(=O)O), 136.6, 136.4, 135.6 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.4, 128.2, 128.2, 128.0, 127.8, 126.5 (15 x HC_{Ar}), 67.2 (Bn-*C*H_2), 55.0 (*C*-3'), 50.2 (*C*-2'), 49.1, 48.5 (2 x Bn-*C*H_2), 46.9 (2 x *C*$H_2$$CH_3$), 44.5 (*C*-2), 41.7 (*C*-2''), 11.5 (2 x CH_2*C*H_3).
IR (ATR): ν = 3278, 1717, 1633, 1523, 1452, 1223, 1046, 733, 697.
MS (ESI^+): m/z = 588.3 $[M+H]^+$, calculated: 588.3181 $[M+H]^+$,
$C_{33}H_{41}N_5O_5$ (587.72 g(mol)$^{-1}$), found: 588.3167 [-2.3 ppm] (ESI^+-HRMS).

Preparation of tripeptide **19**



To a solution of tripeptide **4** (72 mg, 0.12 mmol) in $CHCl_3$ (2.5 mL), *N*-methylpropargylamine (61 µL, 0.72 mmol) was added. After stirring for 8 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 80:20 → 70:30) to give 74 mg (0.11 mmol, 92%) of the title compound as a colourless solid.

TLC: R_f = 0.15 (DCM:AcOEt, 80:20).

Melting point: T_{mp} = 123 °C.

Specific rotation: $[\alpha]_D^{20}$ = -13.8 (c = 1.84, $CHCl_3$)

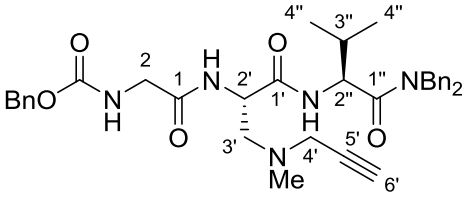
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.52 (d, *J* = 8.9 Hz, 1 H, 2"-N*H*), 7.32-7.08 (m, 15 H, 15 x *H*C_{Ar}), 6.80 (d, *J* = 5.3 Hz, 1 H, 2'-N*H*), 5.39 (d, *J* = 8.7 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.2 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.01 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.83-4.80 (m, 2 H, 2"-*H*, Bn-C*H*$_a$H$_b$), 4.56 (d, *J* = 16.4 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.35-4.30 (m, 2 H, 2'-*H*, Bn-CH$_a$*H*$_b$), 4.14 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.04 (dd, *J* = 8.7, 5.8 Hz, 1 H, 2-*H*), 3.52 (dd, *J* = 17.2, 2.3 Hz, 1 H, 4'-*H*$_a$), 3.37 (dd, *J* = 17.2, 2.3 Hz, 1 H, 4'-*H*$_b$), 2.76 (dd, *J* = 12.2, 4.1 Hz, 1 H, 3'-*H*$_a$), 2.50 (dd, *J* = 12.2, 10.9 Hz, 1 H, 3'-*H*$_b$), 2.44 (s, 3 H, NC*H*$_3$), 2.16 (dd, *J* = 2.3, 2.3 Hz, 1 H, 6'-*H*), 2.08-1.98 (m, 2 H, 3-*H*, 3"-*H*), [0.90 (d, *J* = 6.9 Hz, 3 H), 0.88 (d, *J* = 6.8 Hz, 3 H), 0.85 (d, *J* = 6.8 Hz, 3 H), 0.84 (d, *J* = 6.8 Hz, 3 H) (4-*H*$_a$, 4-*H*$_b$, 4"-*H*$_a$, 4"-*H*$_b$)].

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.1, 171.3, 171.1 (*C*-1, *C*-1', *C*-1"), 156.4 (N*C*(=O)O), 137.1, 136.5, 136.2 (3 x *C*$_{Ar}$), 129.1, 128.8, 128.6, 128.3, 128.2, 128.1, 128.0, 127.6, 127.4 (15 x H*C*$_{Ar}$), 78.3 (*C*-5'), 73.7 (*C*-6'), 67.1 (Bn-*C*H$_2$), 60.2 (*C*-2), 57.3 (*C*-3'), 54.7 (*C*-2"), 50.0 (Bn-*C*H$_2$), 49.9 (*C*-2'), 47.9 (Bn-*C*H$_2$), 46.3 (*C*-4'), 41.0 (N*C*H$_3$), 31.7, 31.1 (*C*-3, *C*-3"), 20.1, 19.3, 17.8, 17.3 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 3289, 2962, 1630, 1531, 1449, 1232, 1028, 733, 697.

MS (ESI$^+$): m/z = 668.4 $[M+H]^+$, calculated: 668.3807 $[M+H]^+$,
$C_{39}H_{49}N_5O_5$ (667.85 g(mol)$^{-1}$), found: 668.3788 [-2.8 ppm] (ESI$^+$-HRMS).

Preparation of tripeptide **20**



To a solution of tripeptide **5** (25 mg, 0.045 mmol) in $CHCl_3$ (1.5 mL), *N*-methylpropargylamine (23 µL, 0.27 mmol) was added. After stirring for 20 h at rt and 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 50:50 → 40:60) to give 22 mg (0.035 mmol, 78%) of the title compound as a colourless solid.

TLC: R_f = 0.17 (DCM:AcOEt, 40:60).

Melting point: T_{mp} = 118 °C.

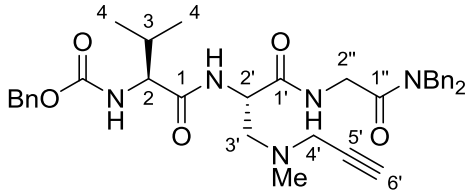
Specific rotation: $[\alpha]_D^{20}$ = -14.1 (c = 1.53, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.57 (d, *J* = 8.9 Hz, 1 H, 2''-N*H*), 7.32-7.08 (m, 15 H, 15 x *H*C_{Ar}), 6.84 (d, *J* = 5.3 Hz, 1 H, 2'-N*H*), 5.42 (dd, *J* = 5.3, 5.2 Hz, 1 H, 2-N*H*), 5.07 (d, *J* = 12.5 Hz, 1 H, Bn-C$H_a$$H_b$), 5.04 (d, *J* = 12.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.84 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.80 (dd, *J* = 8.9, 5.3 Hz, 1 H, 2''-*H*), 4.57 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.36-4.29 (m, 2 H, 2'-*H*, Bn-C$H_a$$H_b$), 4.10 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 3.89 (dd, *J* = 16.9, 5.3 Hz, 1 H, 2-H_a), 3.81 (dd, *J* = 16.9, 5.9 Hz, 1 H, 2-H_b), 3.50 (dd, *J* = 17.2, 2.2 Hz, 1 H, 4'-H_a), 3.38 (dd, *J* = 17.2, 2.2 Hz, 1 H, 4'-H_b), 2.76 (dd, *J* = 11.9, 3.4 Hz, 1 H, 3'-H_a), 2.50 (dd, *J* = 11.9, 11.4 Hz, 1 H, 3'-H_b), 2.44 (s, 3 H, NCH_3), 2.16 (dd, *J* = 2.2, 2.2 Hz, 1 H, 6'-*H*), 2.01 (dqq, *J* = 6.8, 6.3, 5.3 Hz, 1 H, 3''-*H*), 0.87 (d, *J* = 6.3 Hz, 3 H, 4''-H_a), 0.84 (d, *J* = 6.8 Hz, 3 H, 4''-H_b).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.0, 171.2, 168.9 (*C*-1, *C*-1', *C*-1''), 156.5 (N*C*(=O)O), 137.0, 136.4, 136.1 (3 x C_{Ar}), 129.1, 128.8, 128.7, 128.3, 128.3, 128.2, 128.1, 127.7, 127.4 (15 x HC_{Ar}), 78.2 (*C*-5'), 73.7 (*C*-6'), 67.2 (Bn-*C*H_2), 57.4 (*C*-3'), 54.7 (*C*-2''), 50.0 (Bn-*C*H_2), 49.8 (*C*-2'), 47.9 (Bn-*C*H_2), 46.3 (*C*-4'), 44.4 (*C*-2), 41.1 (N*C*H_3), 31.0 (*C*-3''), 20.1, 17.2 (2 x *C*-4'').

IR (ATR): ν = 3275, 1714, 1631, 1514, 1445, 1219, 1043, 730, 697.

MS (ESI^+): m/z = 626.4 $[M+H]^+$, calculated: 626.3337 $[M+H]^+$,
$C_{36}H_{43}N_5O_5$ (625.77 g(mol)$^{-1}$), found: 626.3331 [-0.9 ppm] (ESI^+-HRMS).



Preparation of tripeptide **21**

To a solution of tripeptide **6** (25 mg, 0.045 mmol) in $CHCl_3$ (1.5 mL), *N*-methylpropargylamine (23 µL, 0.27 mmol) was added. After stirring for 20 h at rt and 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 80:20 → 60:40) to give 26 mg (0.042 mmol, 93%) of the title compound as a colourless solid.

TLC: R_f = 0.20 (DCM:AcOEt, 60:40).

Melting point: T_{mp} = 87 °C.

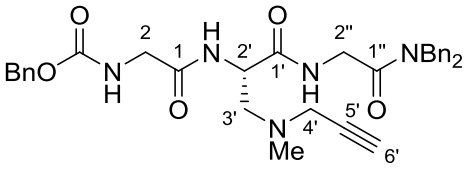
Specific rotation: $[\alpha]_D^{20}$ = +16.1 (c = 1.87, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.41 (dd, *J* = 4.5, 4.2 Hz, 1 H, 2''-N*H*), 7.30-7.05 (m, 15 H, 15 x *H*C_{Ar}), 6.85 (d, *J* = 5.5 Hz, 1 H, 2'-N*H*), 5.37 (d, *J* = 8.6 Hz, 1 H, 2-N*H*), 5.03 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 4.99 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 4.57 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.52 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.35-4.28 (m, 3 H, 2'-*H*, Bn-CH_2), 4.23 (dd, *J* = 17.4, 4.5 Hz, 1 H, 2''-H_a), 4.06-4.02 (m, 2 H, 2-*H*, 2''-H_b), 3.44 (dd, *J* = 17.2, 2.2 Hz, 1 H, 4'-H_a), 3.37 (dd, *J* = 17.2, 2.2 Hz, 1 H, 4'-H_b), 2.77 (dd, *J* = 12.4, 4.5 Hz, 1 H, 3'-H_a), 2.60 (dd, *J* = 12.4, 9.7 Hz, 1 H, 3'-H_b), 2.40 (s, 3 H, NCH_3), 2.15 (dd, *J* = 2.2, 2.2 Hz, 1 H, 6'-*H*), 2.01 (dqq, *J* = 6.8, 6.7, 6.0 Hz, 1 H, 3-*H*), 0.90 (d, *J* = 6.7 Hz, 3 H, 4-H_a), 0.85 (d, *J* = 6.8 Hz, 3 H, 4-H_b).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.4, 170.9, 168.5 (*C*-1, *C*-1', *C*-1''), 156.4 (N*C*(=O)O), 136.6, 136.4, 135.5 (3 x C_{Ar}), 129.2, 128.9, 128.6, 128.4, 128.2, 128.2, 128.1, 127.8, 126.6 (15 x HC_{Ar}), 78.3 (*C*-5'), 73.7 (*C*-6'), 67.1 (Bn-*C*H_2), 60.2 (*C*-2), 57.1 (*C*-3'), 50.3 (*C*-2'), 49.1, 48.6 (2 x Bn-*C*H_2), 46.5 (*C*-4'), 41.7 (*C*-2''), 41.3 (N*C*H_3), 31.7 (*C*-3), 19.3, 17.8 (2 x *C*-4).
IR (ATR): ν = 3289, 1632, 1522, 1452, 1220, 1028, 735, 697, 632.

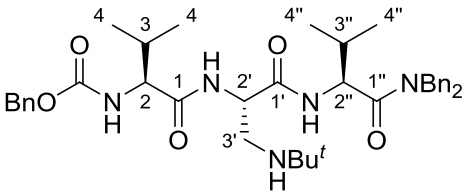
MS (ESI^+): m/z = 626.3 $[M+H]^+$, calculated: 626.3337 $[M+H]^+$,
$C_{36}H_{43}N_5O_5$ (625.77 g(mol)$^{-1}$), found: 626.3333 [-0.7 ppm] (ESI^+-HRMS).

Preparation of tripeptide **22**



To a solution of tripeptide **7** (23 mg, 0.045 mmol) in $CHCl_3$ (1.5 mL), *N*-methylpropargylamine (23 µL, 0.27 mmol) was added. After stirring for 20 h at rt and 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 40:60 → 20:80) to give 19 mg (0.033 mmol, 73%) of the title compound as a colourless solid.
TLC: R_f = 0.3 (DCM:AcOEt, 20:80).
Melting point: T_{mp} = 83 °C.
Specific rotation: $[\alpha]_D^{20}$ = +12.3 (c = 1.21, $CHCl_3$)
^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.35 (dd, *J* = 4.5, 4.0 Hz, 1 H, 2''-N*H*), 7.31-7.05 (m, 15 H, 15 x *H*C_{Ar}), 6.89 (d, *J* = 5.2 Hz, 1 H, 2'-N*H*), 5.45 (s, 1 H, 2-N*H*), 5.04 (s, 2 H, Bn-CH_2), 4.54 (s, 2 H, Bn-CH_2), 4.37-4.32 (m, 3 H, 2'-*H*, Bn-CH_2), 4.20 (dd, *J* = 17.3, 4.5 Hz, 1 H, 2''-H_a), 4.02 (dd, *J* = 17.3, 4.0 Hz, 1 H, 2''-H_b), 3.91-3.76 (m, 2 H, 2-*H*), 3.43 (dd, *J* = 17.1, 2.1 Hz, 1 H, 4'-H_a), 3.35 (dd, *J* = 17.1, 2.1 Hz, 1 H, 4'-H_b), 2.79-2.73 (m, 1 H, 3'-H_a), 2.60 (dd, *J* = 11.6, 8.8 Hz, 1 H, 3'-H_b), 2.39 (s, 3 H, NCH_3), 2.15 (dd, *J* = 2.1, 2.1 Hz, 1 H, 6'-*H*).
^{13}C NMR (101 MHz, $CDCl_3$): δ = 171.0, 169.1, 168.6 (*C*-1, *C*-1', *C*-1''), 156.6 (N*C*(=O)O), 136.6, 136.4, 135.5 (3 x C_{Ar}), 129.2, 128.9, 128.7, 128.4, 128.3, 128.2, 128.1, 127.8, 126.6 (15 x HC_{Ar}), 78.3 (*C*-5'), 73.7 (*C*-6'), 67.3 (Bn-*C*H_2), 57.0 (*C*-3'), 50.4 (*C*-2'), 49.1, 48.7 (2 x Bn-*C*H_2), 46.4 (*C*-4'), 44.5 (*C*-2), 41.7 (*C*-2''), 41.4 (N*C*H_3).
IR (ATR): ν = 3293, 1714, 1630, 1522, 1452, 1236, 1029, 732, 696.
MS (ESI^+): m/z = 584.3 $[M+H]^+$, calculated: 584.2868 $[M+H]^+$,
$C_{33}H_{37}N_5O_5$ (583.69 g(mol)$^{-1}$), found: 584.2863 [-0.7 ppm] (ESI^+-HRMS).

Preparation of tripeptide **23**



To a solution of tripeptide **4** (30 mg, 0.050 mmol) in $CHCl_3$ (2.0 mL), *tert*-butyl amine (32 µL, 0.30 mmol) was added. After stirring for 22 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt:MeOH, 60:40:0 → 96:0:4) to give 31 mg (0.046 mmol, 92%) of the title compound as a colourless solid.
TLC: R_f = 0.18 (DCM:MeOH, 95:5).

Melting point: T_{mp} = 117 °C.

Specific rotation: $[\alpha]_D^{20}$ = +20.3 (c = 2.56, $CHCl_3$)

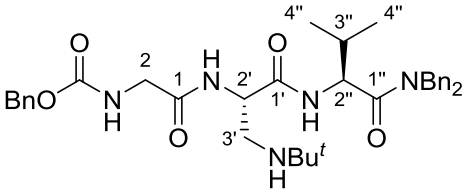
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.67 (d, *J* = 6.2 Hz, 1 H, 2''-N*H*), 7.31-7.03 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.41 (d, *J* = 8.5 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 5.01 (d, *J* = 12.3 Hz, 1 H, Bn-$CH_a$$H_b$), 4.83 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.80 (dd, *J* = 8.0, 6.2 Hz, 1 H, 2''-*H*), 4.58 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.50-4.22 (m, 2 H, 2'-*H*, Bn-$CH_a$$H_b$), 4.11 (d, *J* = 14.8 Hz, 1 H, Bn-$CH_a$$H_b$), 4.03 (dd, *J* = 8.5, 5.9 Hz, 1 H, 2-*H*), 3.16-3.10 (m, 1 H, 3'-H_a), 2.60-2.52 (m, 1 H, 3'-H_b), 2.09-2.00 (m, 2 H, 3-*H*, 3''-*H*), 1.11 (s, 9 H, C(CH_3)$_3$), 0.93-0.85 (m, 12 H, 4-*H*, 4''-*H*). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.2, 171.4, 171.0 (*C*-1, *C*-1', *C*-1''), 156.5 (N*C*(=O)O), 137.0, 136.4, 136.1 (3 x C_{Ar}), 129.0, 128.8, 128.6, 128.3, 128.2, 128.2, 128.0, 127.6, 127.4 (15 x HC_{Ar}), 67.1 (Bn-*C*H_2), 60.3 (*C*-2), 55.2 (*C*-2''), 52.4 (*C*-2'), 50.1, 47.9 (2 x Bn-*C*H_2), 43.8 (*C*-3'), 31.6, 30.9 (*C*-3, *C*-3''), 28.6 (C(*C*H_3)$_3$), 20.0, 19.4, 17.8, 17.5 (2 x *C*-4, 2 x *C*-4''). Despite several attempts, the ^{13}C NMR signal attributed to quaternary carbon atom *C*$(CH_3)_3$ was not observed, probably due to very pronounced line-broadening. However, the presence of the Bu^t group was unambiguously confirmed by appearance of signals for the corresponding three CH_3 groups both in the ^{1}H NMR and ^{13}C NMR spectra, *vide supra*.

IR (ATR): ν = 3287, 2962, 1630, 1529, 1448, 1217, 1028, 733, 697.

MS (ESI^+): m/z = 672.4 $[M+H]^+$, calculated: 672.4120 $[M+H]^+$,
$C_{39}H_{53}N_5O_5$ (671.40 g(mol)$^{-1}$), found: 672.4095 [-3.6 ppm] (ESI^+-HRMS).

Preparation of tripeptide **24**



To a solution of tripeptide **5** (15 mg, 0.027 mmol) in $CHCl_3$ (1.5 mL), *tert*-butyl amine (17 μL, 0.16 mmol) was added. After stirring for 20 h at rt and for 24 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:MeOH, 96:4) to give 16 mg (0.025 mmol, 94%) of the title compound as a colourless solid.

TLC: R_f = 0.11 (DCM:MeOH, 95:5).

Melting point: T_{mp} = 96 °C.

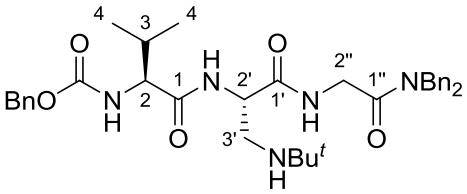
Specific rotation: $[\alpha]_D^{20}$ = -36.3 (c = 0.84, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.69 (d, *J* = 6.2 Hz, 1 H, 2''-N*H*), 7.32-7.08 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.51 (s, 1 H, 2-N*H*), 5.08 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 5.04 (d, *J* = 12.3 Hz, 1 H, Bn-$CH_a$$H_b$), 4.86 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.77 (dd, *J* = 8.0, 6.2 Hz, 1 H, 2''-*H*), 4.58 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.51-4.42 (m, 1 H, 2'-*H*), 4.32 (d, *J* = 16.4 Hz, 1 H, Bn-$CH_a$$H_b$), 4.08 (d, *J* = 14.8 Hz, 1 H, Bn-$CH_a$$H_b$), 3.89 (dd, *J* = 16.9, 5.5 Hz, 1 H, 2-H_a), 3.83 (dd, *J* = 16.9, 5.5 Hz, 1 H, 2-H_b), 3.16 (dd, *J* = 11.1, 2.4 Hz, 1 H, 3'-H_a), 2.59 (dd, *J* = 11.1, 9.7 Hz, 1 H, 3'-H_b), 2.05 (dqq, *J* = 6.7, 6.7, 5.9 Hz, 1 H, 3''-*H*), 1.13 (s, 9 H, C(CH_3)$_3$), 0.92 (d, *J* = 6.7 Hz, 3 H, 4''-H_a), 0.86 (d, *J* = 6.7 Hz, 3 H, 4''-H_b). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.2, 170.9, 169.1 (*C*-1, *C*-1', *C*-1''), 156.6 (N*C*(=O)O), 137.0, 136.3, 136.0 (3 x C_{Ar}), 129.1, 128.8, 128.7, 128.4, 128.3, 128.2, 128.1, 127.7, 127.4 (15 x HC_{Ar}), 67.3 (Bn-*C*H_2), 55.3 (*C*-2''), 52.1 (*C*-2'), 50.1, 48.0 (2 x Bn-*C*H_2), 44.6 (*C*-2), 43.7 (*C*-3'), 30.9 (*C*-3''), 28.4 (C(*C*H_3)$_3$), 20.0, 17.5 (2 x *C*-4''). Despite several attempts, the ^{13}C NMR signal attributed to quaternary carbon atom *C*$(CH_3)_3$ was not observed, probably due to very pronounced line-broadening. However, the presence of the Bu^t group was unambiguously confirmed by appearance of signals for the corresponding three CH_3 groups both in the ^{1}H NMR and ^{13}C NMR spectra, *vide supra*.

IR (ATR): ν = 3286, 2963, 1630, 1526, 1449, 1216, 1046, 734, 697.

MS (ESI^+): m/z = 630.4 $[M+H]^+$, calculated: 630.3650 $[M+H]^+$,
$C_{36}H_{47}N_5O_5$ (629.80 g(mol)$^{-1}$), found: 630.3631 [-3.0 ppm] (ESI^+-HRMS).



Preparation of tripeptide **25**

To a solution of tripeptide **6** (15 mg, 0.027 mmol) in $CHCl_3$ (1.5 mL), *tert*-butyl amine (17 μL, 0.16 mmol) was added. After stirring for 20 h at rt and for 2 d at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:MeOH, 95:5) to give 16 mg (0.025 mmol, 94%) of the title compound as a colourless solid.

TLC: R_f = 0.11 (DCM:MeOH, 95:5).

Melting point: T_{mp} = 81 °C.

Specific rotation: $[\alpha]_D^{20}$ = +4.6 (c = 1.21, $CHCl_3$)

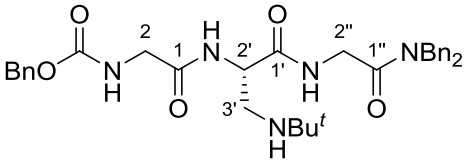
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.59 (s, 1 H, 2''-N*H*), 7.32-7.08 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.49 (s, 1 H, 2-N*H*), 5.05 (d, *J* = 12.5 Hz, 1 H, Bn-CH_aH$_b$), 5.02 (d, *J* = 12.5 Hz, 1 H, Bn-CH$_a$$H_b$), 4.66-4.46 (m, 3 H, 2'-*H*, Bn-CH_2), 4.34 (d, *J* = 17.4 Hz, 1 H, Bn-CH_aH$_b$), 4.30 (d, *J* = 17.4 Hz, 1 H, Bn-CH$_a$$H_b$), 4.16-3.99 (m, 3 H, 2-*H*, 2''-*H*), 3.33-3.24 (m, 1 H, 3'-H_a), 2.84-2.74 (m, 1 H, 3'-H_b), 2.10 (dqq, *J* = 6.8, 6.7, 6.2 Hz, 1 H, 3-*H*), 1.20 (s, 9 H, C(CH_3)$_3$), 0.92 (d, *J* = 6.7 Hz, 3 H, 4-H_a), 0.87 (d, *J* = 6.8 Hz, 3 H, 4-H_b). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.9, 170.6, 169.1 (*C*-1, *C*-1', *C*-1''), 156.9 (N*C*(=O)O), 136.4, 135.2 (3 x C_{Ar}), 129.3, 128.9, 128.7, 128.4, 128.2, 128.2, 128.2, 127.9, 127.6 (15 x HC_{Ar}), 67.2 (Bn-*C*H_2), 60.9 (*C*-2), 52.0 (*C*-2'), 49.4, 49.0 (2 x Bn-*C*H_2), 43.8 (*C*-3'), 41.9 (*C*-2''), 31.2 (*C*-3), 27.5 (C(*C*H_3)$_3$), 19.4, 17.9 (2 x *C*-4). Despite several attempts, the ^{13}C NMR signal attributed to quaternary carbon atom *C*$(CH_3)_3$ was not observed, probably due to very pronounced line-broadening. However, the presence of the Bu^t group was unambiguously confirmed by appearance of signals for the corresponding three CH_3 groups both in the ^{1}H NMR and ^{13}C NMR spectra, *vide supra*.

IR (ATR): ν = 2962, 1639, 1496, 1452, 1219, 1080, 1027, 735, 698.

MS (ESI^+): m/z = 630.4 $[M+H]^+$, calculated: 630.3650 $[M+H]^+$,
$C_{36}H_{47}N_5O_5$ (629.80 g(mol)$^{-1}$), found: 630.3633 [-2.7 ppm] (ESI^+-HRMS).

Preparation of tripeptide **26**



To a solution of tripeptide **7** (23 mg, 0.045 mmol) in $CHCl_3$ (2.0 mL), *tert*-butyl amine (28 µL, 0.27 mmol) was added. After stirring for 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt:MeOH, 30:70:0 → 92:0:8) to give 24 mg (0.041 mmol, 91%) of the title compound as a colourless solid.

TLC: R_f = 0.15 (DCM:MeOH, 93:7).

Melting point: T_{mp} = 63 °C.

Specific rotation: $[\alpha]_D^{20}$ = -5.1 (c = 1.83, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.47 (s, 1 H, 2''-N*H*), 7.77 (s, 1 H, 2'-N*H*), 7.31-7.07 (m, 15 H, 15 x *H*C_{Ar}), 5.66 (s, 1 H, 2-N*H*), 5.05 (d, *J* = 12.6 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.02 (d, *J* = 12.6 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.55-4.47 (m, 3 H, 2'-*H*, Bn-C*H*$_2$), 4.31 (s, 2 H, Bn-C*H*$_2$), 4.13 (d, *J* = 17.0 Hz, 1 H, 2''-*H*$_a$), 4.01 (d, *J* = 17.0 Hz, 1 H, 2''-*H*$_b$), 3.91 (dd, *J* = 16.9, 5.7 Hz, 1 H, 2-*H*$_a$), 3.82 (dd, *J* = 16.9, 5.6 Hz, 1 H, 2-*H*$_b$), 3.32 (dd, *J* = 11.5, 2.9 Hz, 1 H, 3'-*H*$_a$), 2.72 (dd, *J* = 11.5, 6.9 Hz, 1 H, 3'-*H*$_b$), 1.13 (s, 9 H, C(C*H*$_3$)$_3$). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

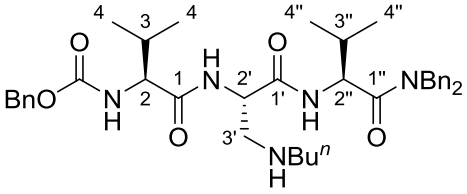
^{13}C NMR (126 MHz, $CDCl_3$): δ = 170.8, 169.7, 169.0 (*C*-1, *C*-1', *C*-1''), 156.9 (N*C*(=O)O), 136.5, 136.3, 135.3 (3 x *C*$_{Ar}$), 129.3, 128.9, 128.6, 128.4, 128.3, 128.2, 128.1, 127.9, 127.6 (15 x H*C*$_{Ar}$), 67.3 (Bn-*C*H$_2$), 53.0 (*C*(CH$_3$)$_3$), 52.3 (*C*-2'), 49.3, 48.9 (2 x Bn-*C*H$_2$), 44.7 (*C*-2), 43.6 (*C*-3'), 41.8 (*C*-2''), 28.0 (C(*C*H$_3$)$_3$).

IR (ATR): ν = 2962, 1647, 1496, 1451, 1218, 1047, 735, 698, 613.

MS (ESI$^+$): m/z = 588.3 [M+H]$^+$, calculated: 588.3181 [M+H]$^+$,

$C_{33}H_{41}N_5O_5$ (587.72 g(mol)$^{-1}$), found: 588.3164 [-2.7 ppm] (ESI$^+$-HRMS).

Preparation of tripeptide **27**



To a solution of tripeptide **4** (30 mg, 0.050 mmol) in $CHCl_3$ (2.0 mL), *n*-butyl amine (30 µL, 0.30 mmol) was added. After stirring for 22 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt:MeOH, 60:40:0 → 96:0:4) to give 33 mg (0.049 mmol, 98%) of the title compound as a colourless solid.

TLC: R_f = 0.18 (DCM:MeOH, 95:5).

Melting point: T_{mp} = 115 °C.

Specific rotation: $[\alpha]_D^{20}$ = -23.6 (c = 2.77, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.63 (d, *J* = 8.5 Hz, 1 H, 2''-N*H*), 7.31-7.08 (m, 15 H, 15 x *H*C_{Ar}), 6.98 (d, *J* = 5.7 Hz, 1 H, 2'-N*H*), 5.44 (d, *J* = 8.6 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.3 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.01 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.81-4.78 (m, 2 H, 2''-*H*, Bn-C*H*$_a$H$_b$), 4.55 (d, *J* = 16.4 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.36-4.33 (m, 2 H, 2'-*H*, Bn-CH$_a$*H*$_b$), 4.15 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.03 (dd, *J* = 8.6, 5.9 Hz, 1 H, 2-*H*), 3.05 (dd, *J* = 12.0, 3.4 Hz, 1 H, 3'-*H*$_a$), 2.68-2.58 (m, 3 H, 3'-*H*$_b$, C*H*$_2$CH$_2$CH$_2$CH$_3$), 2.08-2.00 (m, 2 H, 3-*H*,

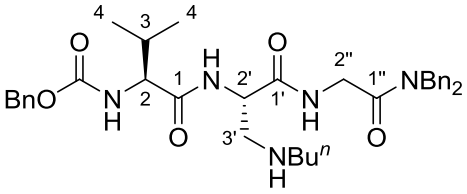
3''-*H*), 1.45-1.39 (m, 2 H, $CH_2CH_2CH_2CH_3$), 1.33-1.25 (m, 2 H, $CH_2CH_2CH_2CH_3$), 0.91-0.83 (m, 15 H, 4-*H*, 4''-*H*, $CH_2CH_2CH_2CH_3$). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.1, 171.4, 171.2 (*C*-1, *C*-1', *C*-1''), 156.5 (N*C*(=O)O), 137.1, 136.5, 136.2 (3 x C_{Ar}), 129.0, 128.8, 128.6, 128.3, 128.2, 128.1, 128.0, 127.6, 127.3 (15 x HC_{Ar}), 67.1 (Bn-CH_2), 60.3 (*C*-2), 54.8 (*C*-2''), 51.6 (*C*-2'), 50.7 (*C*-3'), 50.0 (Bn-CH_2), 49.1 ($CH_2CH_2CH_2CH_3$), 47.9 (Bn-CH_2), 32.1, 31.6, 31.0 (*C*-3, *C*-3'', $CH_2CH_2CH_2CH_3$), 20.4, 20.0, 19.4, 17.8, 17.4 (2 x *C*-4, 2 x *C*-4'', $CH_2CH_2CH_2CH_3$), 14.1 ($CH_2CH_2CH_2CH_3$).

IR (ATR): ν = 3286, 2960, 1631, 1531, 1450, 1219, 1028, 733, 696.

MS (ESI^+): m/z = 672.4 $[M+H]^+$, calculated: 672.4120 $[M+H]^+$,
$C_{39}H_{53}N_5O_5$ (671.40 g(mol)$^{-1}$), found: 672.4098 [-3.2 ppm] (ESI^+-HRMS).

Preparation of tripeptide **28**



To a solution of tripeptide **6** (25 mg, 0.045 mmol) in $CHCl_3$ (2.0 mL), *n*-butyl amine (27 µL, 0.27 mmol) was added. After stirring for 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt:MeOH, 50:50:0 → 96:0:4) to give 24 mg (0.038 mmol, 85%) of the title compound as a colourless solid.

TLC: R_f = 0.05 (DCM:MeOH, 96:4).

Melting point: T_{mp} = 121 °C.

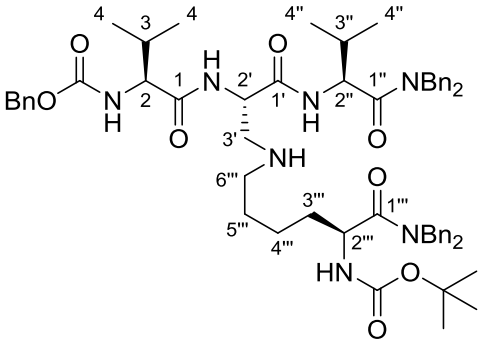
Specific rotation: $[\alpha]_D^{20}$ = +3.6 (c = 2.03, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.42 (s, 1 H, 2''-N*H*), 7.37-7.07 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.45 (d, *J* = 7.7 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.2 Hz, 1 H, Bn-CH_aH$_b$), 5.01 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H$_b$*), 4.55-4.45 (m, 3 H, 2'-*H*, Bn-C*H*$_2$), 4.32 (s, 2 H, Bn-C*H*$_2$), 4.11 (s, 2 H, 2''-*H*), 4.02 (dd, *J* = 7.7, 6.1 Hz, 1 H, 2-*H*), 3.18 (dd, *J* = 8.7, 3.3 Hz, 1 H, 3'-*H$_a$*), 2.73-2.59 (m, 3 H, 3'-*H$_b$*, C*H*$_2$CH$_2$CH$_2$CH$_3$), 2.09 (dqq, *J* = 6.7, 6.6, 6.1 Hz, 1 H, 3-*H*), 1.50-1.44 (m, 2 H, $CH_2CH_2CH_2CH_3$), 1.32-1.25 (m, 2 H, $CH_2CH_2CH_2CH_3$), 0.92-0.82 (m, 9 H, 4-*H*, $CH_2CH_2CH_2CH_3$). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.6, 171.0, 168.8 (*C*-1, *C*-1', *C*-1''), 156.7 (N*C*(=O)O), 136.5, 136.4, 135.4 (3 x C_{Ar}), 129.3, 128.8, 128.6, 128.4, 128.3, 128.2, 128.1, 127.8, 126.6 (15 x HC_{Ar}), 67.2 (Bn-CH_2), 60.6 (*C*-2), 51.9 (*C*-2'), 50.5 (*C*-3'), 49.3 ($CH_2CH_2CH_2CH_3$), 49.2, 48.8 (2 x Bn-CH_2), 41.7 (*C*-2''), 31.4 ($CH_2CH_2CH_2CH_3$), 31.3 (*C*-3), 20.4 ($CH_2CH_2CH_2CH_3$), 19.4, 17.8 (2 x *C*-4), 14.0 ($CH_2CH_2CH_2CH_3$).

IR (ATR): ν = 3287, 2960, 1630, 1523, 1452, 1219, 1028, 736, 697.

MS (ESI^+): m/z = 630.4 $[M+H]^+$, calculated: 630.3650 $[M+H]^+$,
$C_{36}H_{47}N_5O_5$ (629.80 g(mol)$^{-1}$), found: 630.3634 [-2.5 ppm] (ESI^+-HRMS).



Preparation of lysine tripeptide conjugate **29**

To lysine derivative **S1** (100 mg, 0.179 mmol) in degassed EtOH (4 mL), 10 wt% palladium on charcoal (10 mg, 9.4 μmol) was added. The resulting suspension was stirred under a hydrogen atmosphere (1 bar, balloon) for 3 h and then filtered through a syringe filter. The syringe filter was washed with EtOH (3 x 3 mL) and the solvent of the combined filtrates evaporated *in vacuo*. The resulting colourless solid was dried *in vacuo*. The resulting free amine was always prepared freshly and used instantly in the subsequent transformation without further purification.

To a solution of tripeptide **4** (24 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), 1/2 of the crude product (only 1/2 of the initially prepared Cbz-deprotected lysine derivative, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 0.080 mmol) was added. After stirring for 24 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 50:50:0 → 95:0:5) to give 35 mg (0.034 mmol, 85%) of the title compound as a colourless solid.

TLC: R_f = 0.36 (DCM:MeOH, 93:7).

Melting point: T_{mp} = 142 °C.

Specific rotation: $[\alpha]_D^{20}$ = -30.9 (c = 2.79, $CHCl_3$)

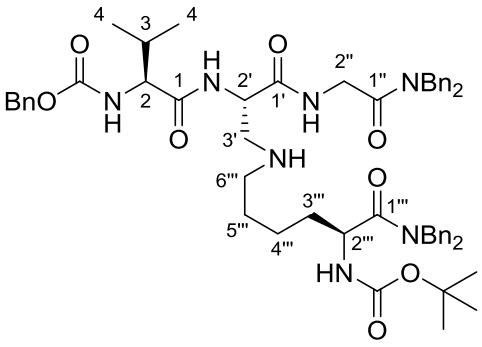
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.54 (d, *J* = 7.6 Hz, 1 H, 2"-N*H*), 7.39-7.16 (m, 25 H, 25 x *H*C_{Ar}), 7.03 (s, 1 H, 2'-N*H*), 5.59 (d, *J* = 7.7 Hz, 1 H, 2-N*H*), 5.46 (d, *J* = 8.4 Hz, 1 H, 2'"-N*H*), 5.10 (s, 2 H, Bn-C*H*$_2$), 4.90-4.85 (m, 2 H, 2"-*H*, Bn-C*H*$_a$H$_b$), 4.73-4.52 (m, 6 H, 2'"-*H*, Bn-C*H*$_a$H$_b$, 2 x Bn-C*H*$_2$), 4.43 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.36 (s, 1 H, 2'-*H*), 4.24 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.11 (dd, *J* = 7.7, 6.3 Hz, 1 H, 2-*H*), 3.10 (dd, *J* = 12.1, 3.6 Hz, 1 H, 3'-*H*$_a$), 2.67-2.53 (m, 3 H, 3'-*H*$_b$, 6'"-*H*), 2.17-2.08 (m, 2 H, 3-*H*, 3"-*H*), 1.64-1.57 (m, 2 H, 3'"-*H*), 1.44-1.27 (m, 13 H, 4'"-*H*, 5'"-*H*, C(C*H*$_3$)$_3$), 0.99-0.90 (m, 12 H, 4-*H*, 4"-*H*). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 173.5, 172.0, 171.4, 171.2 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.5, 155.7 (2 x N*C*(=O)O), 137.1, 136.9, 136.5, 136.4, 136.2 (5 x *C*$_{Ar}$), 129.0, 128.8, 128.8, 128.6, 128.3, 128.2, 128.2, 128.0, 127.9, 127.6, 127.3 127.0 (25 x H*C*$_{Ar}$), 79.7 (*C*(CH$_3$)$_3$), 67.1 (Bn-*C*H$_2$), 60.4 (*C*-2), 54.7 (*C*-2"), 51.7 (*C*-2'), 50.5 (*C*-3'), 50.5 (*C*-2'"), 50.1, 50.0 (2 x Bn-*C*H$_2$), 49.2 (*C*-6'"), 48.5, 47.9 (2 x Bn-*C*H$_2$), 33.3 (*C*-3'"), 31.4 (*C*-3), 31.0 (*C*-3"), 29.7 (*C*-5'"), 28.5 (C(*C*H$_3$)$_3$), 23.1 (*C*-4'"), 20.0, 19.4, 17.8, 17.5 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 3286, 1678, 1637, 1527, 1450, 1218, 1167, 1028, 695.

MS (ESI$^+$): m/z = 1024.5 $[M+H]^+$, calculated: 1024.5906 $[M+H]^+$,

$C_{60}H_{77}N_7O_8$ (1024.32 g(mol)$^{-1}$), found: 1024.5907 [+0.1 ppm] (ESI$^+$-HRMS).



Preparation of lysine tripeptide conjugate **30**

To lysine derivative **S1** (100 mg, 0.179 mmol) in degassed EtOH (4 mL), 10 wt% palladium on charcoal (10 mg, 9.4 μmol) was added. The resulting suspension was stirred under a hydrogen atmosphere (1 bar, balloon) for 3 h and then filtered through a syringe filter. The syringe filter was washed with EtOH (3 x 3 mL) and the solvent of the combined filtrates evaporated *in vacuo*. The resulting colourless solid was dried *in vacuo*. The resulting free amine was always prepared freshly and used instantly in the subsequent transformation without further purification.

To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), 1/2 of the crude product (only 1/2 of the initially prepared Cbz-deprotected lysine derivative, *vide supra*, was used in the 2nd step, calculated maximal amount of substance: 0.080 mmol) was added. After stirring for 24 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 30:70:0 → 95:0:5) to give 37 mg (0.038 mmol, 95%) of the title compound as a colourless solid.

TLC: R_f = 0.25 (DCM:MeOH, 93:7).

Melting point: T_{mp} = 83 °C.

Specific rotation: $[\alpha]_D^{20}$ = -11.9 (c = 2.78, $CHCl_3$)

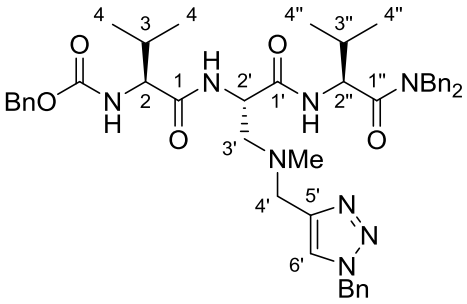
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.34 (s, 1 H, 2''-N*H*), 7.38-7.14 (m, 26 H, 2'-N*H*, 25 x *H*C_{Ar}), 5.73 (d, *J* = 7.2 Hz, 1 H, 2-N*H*), 5.55 (d, *J* = 8.2 Hz, 1 H, 2'''-N*H*), 5.13 (s, 2 H, Bn-CH_2), 4.70-4.39 (m, 10 H, 2'-*H*, 2'''-*H*, 4 x Bn-CH_2), 4.18 (s, 2 H, 2''-*H*), 4.09 (dd, *J* = 7.2, 6.4 Hz, 1 H, 2-*H*), 3.19 (dd, *J* = 12.1, 3.5 Hz, 1 H, 3'-H_a), 2.71-2.51 (m, 3 H, 3'-H_b, 6'''-*H*), 2.17 (dqq, *J* = 6.6, 6.6, 6.4 Hz, 1 H, 3-*H*), 1.69-1.56 (m, 2 H, 3'''-*H*), 1.47-1.27 (m, 13 H, 4'''-*H*, 5'''-*H*, C(CH_3)$_3$), 0.99 (d, *J* = 6.6 Hz, 3 H, 4-H_a), 0.93 (d, *J* = 6.6 Hz, 3 H, 4-H_b). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 173.5, 171.5, 171.1, 168.7 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.7, 155.7 (2 x N*C*(=O)O), 136.9, 136.6, 136.5, 136.4, 135.5 (5 x C_{Ar}), 129.2, 129.0, 128.8, 128.8, 128.6, 128.4, 128.2, 128.1, 128.0, 127.9, 127.8, 127.6, 127.1, 126.6 (25 x HC_{Ar}), 79.7 (*C*(CH_3)$_3$), 67.1 (Bn-*C*H_2), 60.7 (*C*-2), 52.0 (*C*-2'), 50.5 (*C*-3'), 50.5 (*C*-2'''), 50.1 (Bn-*C*H_2), 49.3 (*C*-6'''), 49.1, 48.7, 48.5 (3 x Bn-*C*H_2), 41.7 (*C*-2''), 33.1 (*C*-3'''), 31.2 (*C*-3), 29.3 (*C*-5'''), 28.4 (C(*C*H_3)$_3$), 23.0 (*C*-4'''), 19.4, 17.9 (2 x *C*-4).

IR (ATR): ν = 3312, 1640, 1524, 1451, 1220, 1166, 1028, 732, 697.

MS (ESI$^+$): m/z = 1004.5 $[M+Na]^+$, calculated: 982.5437 $[M+H]^+$,

$C_{57}H_{71}N_7O_8$ (982.24 g(mol)$^{-1}$), found: 982.5429 [-0.8 ppm] (ESI$^+$-HRMS).



Preparation of 1,2,3-triazole **31**

To a thoroughly degassed solution of alkyne **19** (24 mg, 0.036 mmol) in abs. $CHCl_3$ (1.5 mL), benzyl azide[S4] (29 mg, 0.22 mmol), CuI (10 mg, 0.054 mmol) and NEt_3 (60 µL, 0.43 mmol) were added. After stirring for 3 h the reaction mixture was diluted with DCM (50 mL) and washed with water (50 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 60:40 → 40:60) to give 19 g (0.024 mmol, 67%) of the title compound as a pale yellow solid.

TLC: R_f = 0.13 (DCM:AcOEt, 40:60).

Melting point: T_{mp} = 74 °C.

Specific rotation: $[\alpha]_D^{20}$ = +2.3 (c = 1.12, $CHCl_3$)

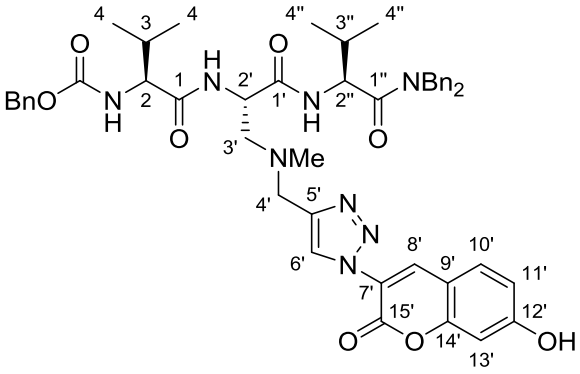
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.75 (d, *J* = 8.7 Hz, 1 H, 2"-N*H*), 7.77 (s, 1 H, 6'-*H*), 7.31-7.06 (m, 20 H, 20 x *H*C_{Ar}), 6.85 (d, *J* = 4.2 Hz, 1 H, 2'-N*H*), 5.44 (d, *J* = 14.9 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.39 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$*H*$_b$), 5.35 (d, *J* = 8.4 Hz, 1 H, 2-N*H*), 5.06 (d, *J* = 12.2 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.02 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.80 (dd, *J* = 8.7, 6.0 Hz, 1 H, 2"-*H*), 4.76 (d, *J* = 14.9 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.56 (d, *J* = 16.5 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.39 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.37-4.33 (m, 1 H, 2'-*H*), 4.16 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.02 (dd, *J* = 8.4, 6.1 Hz, 1 H, 2-*H*), 3.85 (d, *J* = 13.6 Hz, 1 H, 4'-*H*$_a$), 3.71 (d, *J* = 13.6 Hz, 1 H, 4'-*H*$_b$), 2.71 (dd, *J* = 11.9, 3.0 Hz, 1 H, 3'-*H*$_a$), 2.53 (dd, *J* = 11.9, 11.0 Hz, 1 H, 3'-*H*$_b$), 2.32 (s, 3 H, NC*H*$_3$), 2.05 (dqq, *J* = 6.7, 6.6, 6.1 Hz, 1 H, 3-*H*), 1.96 (dqq, *J* = 6.7, 6.7, 6.0 Hz, 1 H, 3"-*H*), [0.90 (d, *J* = 6.6 Hz, 3 H), 0.84 (d, *J* = 6.7 Hz, 3 H), 0.80 (d, *J* = 6.7 Hz, 3 H), 0.78 (d, *J* = 6.7 Hz, 3 H) (4-*H*$_a$, 4-*H*$_b$, 4"-*H*$_a$, 4"-*H*$_b$)].

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.0, 171.3, 170.8 (*C*-1, *C*-1', *C*-1"), 156.3 (N*C*(=O)O), 144.3 (*C*-5'), 137.0, 136.4, 136.1, 134.9 (4 x *C*$_{Ar}$), 129.0, 129.0, 128.7, 128.6, 128.6, 128.2, 128.1, 128.1, 127.9, 127.5, 127.2 (20 x H*C*$_{Ar}$), 123.5 (*C*-6'), 67.0 (Bn-*C*H$_2$), 60.1 (*C*-2), 57.9 (*C*-3'), 54.5 (*C*-2"), 54.1 (Bn-*C*H$_2$), 51.7 (*C*-4'), 50.0 (Bn-*C*H$_2$), 49.6 (*C*-2'), 47.9 (Bn-*C*H$_2$), 41.6 (N*C*H$_3$), 31.5, 31.1 (*C*-3, *C*-3"), 19.9, 19.2, 17.7, 17.3 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 3288, 2961, 1632, 1531, 1496, 1452, 1219, 1029, 697.

MS (ESI^+): m/z = 801.4 $[M+H]^+$, calculated: 801.4446 $[M+H]^+$,

$C_{46}H_{56}N_8O_5$ (801.01 g(mol)$^{-1}$), found: 801.4423 [-2.9 ppm] (ESI^+-HRMS).



Preparation of 1,2,3-triazole **32**

To a thoroughly degassed solution of alkyne **19** (24 mg, 0.036 mmol) in abs. $CHCl_3$ (1.0 mL) and abs. EtOH (1.0 mL), 3-azido-7-hydroxycoumarine[S5] (15 mg, 0.072 mmol), CuI (3.4 mg, 0.018 mmol) and NEt_3 (15 µL, 0.11 mmol) were added. After stirring for 3 h the reaction mixture was diluted with DCM (30 mL) and washed with water (30 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 50:50 → 30:70) to give 16 mg (0.018 mmol, 51%) of the title compound as a yellow solid.

TLC: R_f = 0.18 (DCM:AcOEt, 40:60).

Melting point: T_{mp} = 108 °C.

Specific rotation: $[\alpha]_D^{20}$ = -10.7 (c = 0.70, $CHCl_3$)

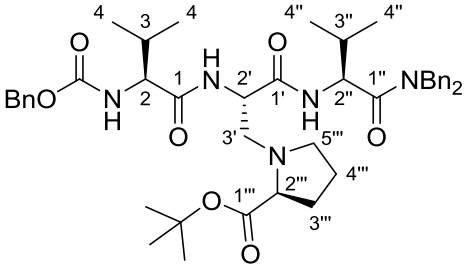
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.75 (d, *J* = 8.5 Hz, 1 H, 2''-N*H*), 8.59 (s, 1 H, 6'-*H*), 8.20 (s, 1 H, 8'-*H*), 7.42-7.18 (m, 16 H, 10'-*H*, 15 x *H*C_{Ar}), 7.13 (d, *J* = 4.9 Hz, 1 H, 2'-N*H*), 6.76-6.72 (m, 2 H, 11'-*H*, 13'-*H*), 5.53 (d, *J* = 8.0 Hz, 1 H, 2-N*H*), 5.15 (d, *J* = 12.3 Hz, 1 H, Bn-CH_aH$_b$), 5.11 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$$H_b$), 4.93 (dd, *J* = 8.5, 6.7 Hz, 1 H, 2''-*H*), 4.83 (d, *J* = 14.9 Hz, 1 H, Bn-CH_aH$_b$), 4.70 (d, *J* = 16.5 Hz, 1 H, Bn-CH_aH$_b$), 4.63-4.57 (m, 2 H, 2'-*H*, Bn-CH$_a$$H_b$), 4.39 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$$H_b$), 4.16 (dd, *J* = 8.0, 6.6 Hz, 1 H, 2-*H*), 4.06 (d, *J* = 14.2 Hz, 1 H, 4'-H_a), 3.85 (d, *J* = 14.2 Hz, 1 H, 4'-H_b), 2.89 (dd, *J* = 9.9, 4.0 Hz, 1 H, 3'-H_a), 2.72 (dd, *J* = 10.7, 9.9 Hz, 1 H, 3'-H_b), 2.47 (s, 3 H, NCH_3), 2.25-2.17 (m, 2 H, 3-*H*, 3''-*H*), [1.04 (d, *J* = 6.6 Hz, 3 H), 1.03 (d, *J* = 6.5 Hz, 3 H), 0.98 (d, *J* = 5.7 Hz, 3 H), 0.97 (d, *J* = 6.5 Hz, 3 H) (4-H_a, 4-H_b, 4''-H_a, 4''-H_b)]. The signal attributed to the hydroxy O*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.7, 171.9, 171.0 (*C*-1, *C*-1', *C*-1''), 162.0, 156.6, 156.1, 154.6 (*C*-12', *C*-14', *C*-15', N*C*(=O)O), 143.8 (*C*-5'), 136.7, 136.4, 136.0 (3 x C_{Ar}), 133.5 (*C*-8'), 130.1 (*C*-10'), 129.2, 128.9, 128.7, 128.3, 128.2, 128.1, 128.1, 127.8, 127.3 (15 x HC_{Ar}), 124.1 (*C*-6'), 119.4, 114.7, 110.7, 103.1 (*C*-7', *C*-9', *C*-11', *C*-13'), 67.2 (Bn-*C*H$_2$), 60.5 (*C*-2), 57.7 (*C*-3'), 55.0 (*C*-2''), 51.6 (*C*-4'), 50.5 (Bn-*C*H$_2$), 50.3 (*C*-2'), 48.5 (Bn-*C*H$_2$), 42.2 (N*C*H$_3$), 31.5, 31.3 (*C*-3, *C*-3''), 20.0, 19.4, 17.9, 17.8 (2 x *C*-4, 2 x *C*-4'').

IR (ATR): ν = 2961, 1609, 1515, 1452, 1231, 1118, 1029, 734, 697.

MS (ESI$^+$): m/z = 871.4 $[M+H]^+$, calculated: 871.4137 $[M+H]^+$,

$C_{48}H_{54}N_8O_8$ (871.01 g(mol)$^{-1}$), found: 871.4113 [-2.8 ppm] (ESI$^+$-HRMS).



Preparation of proline tripeptide conjugate **33**

To a solution of tripeptide **4** (24 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), L-proline *tert*-butyl ester (14 mg, 0.080 mmol) was added. After stirring for 24 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 90:10 → 80:20) to give 30 mg (0.039 mmol, 98%) of the title compound as a colourless solid.

TLC: R_f = 0.21 (DCM:AcOEt, 80:20).

Melting point: T_{mp} = 81 °C.

Specific rotation: $[\alpha]_D^{20}$ = -44.4 (c = 2.83, $CHCl_3$)

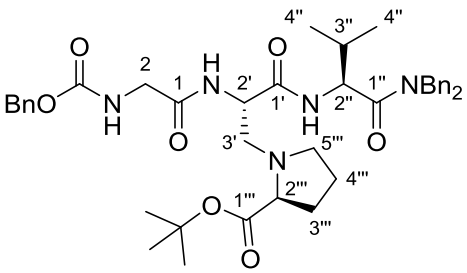
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.30 (d, *J* = 8.5 Hz, 1 H, 2"-N*H*), 7.48 (d, *J* = 4.1 Hz, 1 H, 2'-N*H*), 7.38-7.18 (m, 15 H, 15 x *H*C_{Ar}), 5.59 (d, *J* = 8.8 Hz, 1 H, 2-N*H*), 5.14 (d, *J* = 12.2 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.09 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.86-4.83 (m, 2 H, 2"-*H*, Bn-C*H*$_a$H$_b$), 4.62 (d, *J* = 16.6 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.52 (d, *J* = 16.6 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.32-4.22 (m, 3 H, 2-*H*, 2'-*H*, Bn-CH$_a$*H*$_b$), 3.40 (dd, *J* = 9.0, 4.4 Hz, 1 H, 2'"-*H*), 3.23-3.19 (m, 1 H, 5'"-*H*$_a$), 2.99 (dd, *J* = 12.7, 7.5 Hz, 1 H, 3'-*H*$_a$), 2.78 (dd, *J* = 12.7, 8.1 Hz, 1 H, 3'-*H*$_b$), 2.56-2.51 (m, 1 H, 5'"-*H*$_b$), 2.23-2.12 (m, 3 H, 3-*H*, 3"-*H*, 3'"-*H*$_a$), 1.94-1.78 (m, 3 H, 3'"-*H*$_b$, 4'"-*H*), 1.48 (s, 9 H, C(C*H*$_3$)$_3$), 1.00-0.90 (m, 12 H, 4-*H*, 4"-*H*).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 175.1, 172.1, 171.8, 170.6 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.4 (N*C*(=O)O), 137.2, 136.6, 136.5 (3 x *C*$_{Ar}$), 129.0, 128.7, 128.6, 128.4, 128.1, 127.9, 127.5, 127.4 (15 x H*C*$_{Ar}$), 81.4 (*C*(CH$_3$)$_3$), 67.0 (Bn-*C*H$_2$), 66.2 (*C*-2'"), 60.1 (*C*-2), 55.9 (*C*-3'), 54.7 (*C*-2"), 53.9 (*C*-5'"), 51.9 (*C*-2'), 50.0, 48.0 (2 x Bn-*C*H$_2$), 31.8 (*C*-3), 31.2 (*C*-3"), 30.4 (*C*-3'"), 28.2 (C(*C*H$_3$)$_3$), 24.2 (*C*-4'"), 19.9, 19.3, 17.4, 17.4 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 1720, 1632, 1532, 1449, 1367, 1218, 1149, 1028, 697.

MS (ESI^+): m/z = 770.4 $[M+H]^+$, calculated: 770.4487 $[M+H]^+$,

$C_{44}H_{59}N_5O_7$ (769.98 g(mol)$^{-1}$), found: 770.4458 [-3.8 ppm] (ESI^+-HRMS).



Preparation of proline tripeptide conjugate **34**

To a solution of tripeptide **5** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), L-proline *tert*-butyl ester (14 mg, 0.080 mmol) was added. After stirring for 24 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 70:30 → 60:40) to give 27 mg (0.037 mmol, 93%) of the title compound as a

colourless solid.

TLC: R_f = 0.11 (DCM:AcOEt, 70:30).

Melting point: T_{mp} = 128 °C.

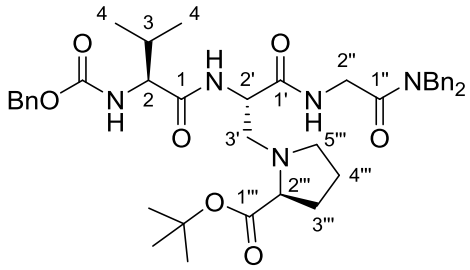
Specific rotation: $[\alpha]_D^{20}$ = -38.4 (c = 2.58, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.45 (d, *J* = 8.5 Hz, 1 H, 2"-N*H*), 7.39-7.18 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.54 (dd, *J* = 5.0, 5.0 Hz, 1 H, 2-N*H*), 5.13 (s, 2 H, Bn-C*H*$_2$), 4.85-4.82 (m, 2 H, 2"-*H*, Bn-C*H*$_a$H$_b$), 4.62 (d, *J* = 16.5 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.51 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.32-4.22 (m, 2 H, 2'-*H*, Bn-CH$_a$*H*$_b$), 3.97 (d, *J* = 5.0 Hz, 2 H, 2-*H*), 3.41 (dd, *J* = 9.1, 4.2 Hz, 1 H, 2'"-*H*), 3.25-3.20 (m, 1 H, 5'"-*H*$_a$), 2.97 (dd, *J* = 12.4, 6.5 Hz, 1 H, 3'-*H*$_a$), 2.78 (dd, *J* = 12.4, 9.1 Hz, 1 H, 3'-*H*$_b$), 2.59-2.54 (m, 1 H, 5'"-*H*$_b$), 2.21-2.11 (m, 2 H, 3"-*H*, 3'"-*H*$_a$), 1.95-1.76 (m, 3 H, 3'"-*H*$_b$, 4'"-*H*), 1.46 (s, 9 H, C(C*H*$_3$)$_3$), 0.99 (d, *J* = 6.8 Hz, 3 H, 4"-*H*$_a$), 0.93 (d, *J* = 6.8 Hz, 3 H, 4"-*H*$_b$).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 175.0, 172.1, 170.6, 169.0 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.5 (N*C*(=O)O), 137.2, 136.5, 136.4 (3 x *C*$_{Ar}$), 129.0, 128.7, 128.6, 128.3, 128.2, 128.2, 127.9, 127.5, 127.4 (15 x H*C*$_{Ar}$), 81.3 (*C*(CH$_3$)$_3$), 67.1 (Bn-*C*H$_2$), 66.4 (*C*-2'"), 56.4 (*C*-3'), 54.9 (*C*-2"), 54.2 (*C*-5'"), 51.7 (*C*-2'), 50.1, 48.0 (2 x Bn-*C*H$_2$), 44.4 (*C*-2), 31.1 (*C*-3"), 30.5 (*C*-3'"), 28.2 (C(*C*H$_3$)$_3$), 24.2 (*C*-4'"), 19.9, 17.3 (2 x *C*-4").

IR (ATR): ν = 1711, 1629, 1519, 1445, 1366, 1218, 1149, 1041, 698.

MS (ESI^+): m/z = 728.3 $[M+H]^+$, calculated: 728.4018 $[M+H]^+$,
$C_{41}H_{53}N_5O_7$ (727.90 g(mol)$^{-1}$), found: 728.3986 [-4.3 ppm] (ESI^+-HRMS).



Preparation of proline tripeptide conjugate **35**

To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), L-proline *tert*-butyl ester (14 mg, 0.080 mmol) was added. After stirring for 24 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 80:20 → 75:25) to give 27 mg (0.037 mmol, 93%) of the title compound as a colourless solid.

TLC: R_f = 0.19 (DCM:AcOEt, 75:25).

Melting point: T_{mp} = 75 °C.

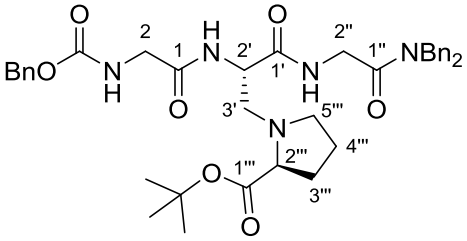
Specific rotation: $[\alpha]_D^{20}$ = -17.5 (c = 1.78, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.90 (dd, *J* = 4.2, 4.0 Hz, 1 H, 2"-N*H*), 7.62 (d, *J* = 4.4 Hz, 1 H, 2'-N*H*), 7.30-7.06 (m, 15 H, 15 x *H*C_{Ar}), 5.58 (d, *J* = 8.9 Hz, 1 H, 2-N*H*), 5.08 (d, *J* = 12.3 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.08 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.57 (d, *J* = 14.8 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.50 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.32 (s, 2 H, Bn-C*H*$_2$), 4.28-4.15 (m, 3 H, 2-*H*, 2'-*H*, 2"-*H*$_a$), 4.03 (dd, *J* = 17.4, 4.0 Hz, 1 H, 2"-*H*$_b$), 3.27 (dd, *J* = 8.9, 5.0 Hz, 1 H, 2'"-*H*), 3.11-3.06 (m, 1 H, 5'"-*H*$_a$), 2.90 (dd, *J* = 13.0, 9.2 Hz, 1 H, 3'-*H*$_a$), 2.83 (dd, *J* = 13.0, 6.5 Hz, 1 H, 3'-*H*$_b$), 2.48-2.43 (m, 1 H, 5'"-*H*$_b$), 2.23-2.05 (m, 2 H, 3-*H*, 3'"-*H*$_a$), 1.82-1.73 (m, 3 H, 3'"-*H*$_b$, 4'"-*H*), 1.35 (s, 9 H, C(C*H*$_3$)$_3$), 0.93 (d, *J* = 6.8 Hz, 3 H, 4-*H*$_a$), 0.84 (d, *J* = 6.9 Hz, 3 H, 4-*H*$_b$).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 175.0, 172.4, 170.8, 168.5 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.6 (N*C*(=O)O), 136.7, 136.6, 135.6 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.5, 128.2, 128.1, 128.0, 127.7, 127.6 (15 x HC_{Ar}), 81.6 (*C*(CH_3)$_3$), 67.0 (Bn-*C*H_2), 66.1 (*C*-2'''), 60.1 (*C*-2), 55.4 (*C*-3'), 53.4 (*C*-5'''), 52.8 (*C*-2'), 49.0, 48.5 (2 x Bn-*C*H_2), 41.7 (*C*-2''), 31.8 (*C*-3), 30.1 (*C*-3'''), 28.2 (C(*C*H_3)$_3$), 24.0 (*C*-4'''), 19.4, 17.3 (2 x *C*-4).

IR (ATR): ν = 3297, 1713, 1634, 1515, 1219, 1151, 1027, 735, 697.

MS (ESI^+): m/z = 728.4 $[M+H]^+$, calculated: 728.4018 $[M+H]^+$,
$C_{41}H_{53}N_5O_7$ (727.90 g(mol)$^{-1}$), found: 728.3989 [-4.0 ppm] (ESI^+-HRMS).



Preparation of proline tripeptide conjugate **36**

To a solution of tripeptide **7** (21 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), L-proline *tert*-butyl ester (14 mg, 0.080 mmol) was added. After stirring for 24 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 40:60 → 30:70) to give 26 mg (0.038 mmol, 95%) of the title compound as a colourless solid.

TLC: R_f = 0.14 (DCM:AcOEt, 40:60).

Melting point: T_{mp} = 42 °C.

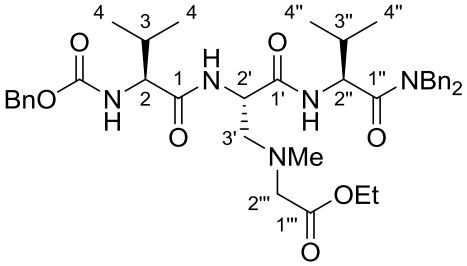
Specific rotation: $[\alpha]_D^{20}$ = -19.3 (c = 2.42, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.06 (dd, *J* = 4.5, 3.7 Hz, 1 H, 2''-N*H*), 7.64 (d, *J* = 4.5 Hz, 1 H, 2'-N*H*), 7.38-7.14 (m, 15 H, 15 x *H*C_{Ar}), 5.61 (s, 1 H, 2-N*H*), 5.13 (s, 2 H, Bn-CH_2), 4.63 (d, *J* = 14.9 Hz, 1 H, Bn-CH_aH$_b$), 4.59 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$$H_b$), 4.40-4.35 (m, 3 H, 2'-*H*, Bn-CH_2), 4.25 (dd, *J* = 17.1, 4.5 Hz, 1 H, 2''-H_a), 4.12 (dd, *J* = 17.1, 3.7 Hz, 1 H, 2''-H_b), 4.04-3.97 (m, 2 H, 2-*H*), 3.35 (dd, *J* = 8.5, 4.4 Hz, 1 H, 2'''-*H*), 3.20-3.15 (m, 1 H, 5'''-H_a), 2.99 (dd, *J* = 12.4, 8.4 Hz, 1 H, 3'-H_a), 2.89 (dd, *J* = 12.4, 6.6 Hz, 1 H, 3'-H_b), 2.59-2.51 (m, 1 H, 5'''-H_b), 2.21-2.13 (m, 1 H, 3'''-H_a), 1.91-1.80 (m, 3 H, 3'''-H_b, 4'''-*H*), 1.45 (s, 9 H, C(CH_3)$_3$).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 174.8, 170.8, 169.8, 168.5 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.5 (N*C*(=O)O), 136.7, 136.5, 135.5 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.4, 128.2, 128.0, 127.8, 126.7 (15 x HC_{Ar}), 81.5 (*C*(CH_3)$_3$), 67.1 (Bn-*C*H_2), 66.3 (*C*-2'''), 55.6 (*C*-3'), 53.6 (*C*-5'''), 52.6 (*C*-2'), 49.1, 48.6 (2 x Bn-*C*H_2), 44.4 (*C*-2), 41.6 (*C*-2''), 30.1 (*C*-3'''), 28.2 (C(*C*H_3)$_3$), 23.9 (*C*-4''').

IR (ATR): ν = 1721, 1645, 1496, 1452, 1367, 1218, 1151, 732, 697.

MS (ESI^+): m/z = 686.3 $[M+H]^+$, calculated: 686.3548 $[M+H]^+$,
$C_{38}H_{47}N_5O_7$ (685.82 g(mol)$^{-1}$), found: 686.3520 [-4.1 ppm] (ESI^+-HRMS).



Preparation of sarcosine tripeptide conjugate **37**

To a solution of tripeptide **4** (24 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), sarcosine ethyl ester (9.4 mg, 0.080 mmol) was added. After stirring for 40 h at 65 °C the solvent was evaporated *in vacuo*. The resultant

crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 90:10:0 → 99:0:1) to give 28 mg (0.039 mmol, 98%) of the title compound as a colourless solid.

TLC: R_f = 0.25 (DCM:AcOEt, 80:20).

Melting point: T_{mp} = 104 °C.

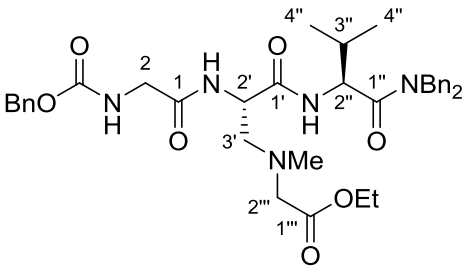
Specific rotation: $[\alpha]_D^{20}$ = -16.3 (c = 2.46, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.32 (d, *J* = 8.8 Hz, 1 H, 2''-N*H*), 7.31-7.08 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.40 (d, *J* = 8.8 Hz, 1 H, 2-N*H*), 5.06 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 5.02 (d, *J* = 12.3 Hz, 1 H, Bn-C$H_a$$H_b$), 4.81-4.76 (m, 2 H, 2''-*H*, Bn-C$H_a$$H_b$), 4.56 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.38 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.27-4.23 (m, 1 H, 2'-*H*), 4.17 (d, *J* = 14.9 Hz, 1 H, Bn-C$H_a$$H_b$), 4.13-4.06 (m, 3 H, 2-*H*, Et-CH_2), 3.50 (d, *J* = 17.3 Hz, 1 H, 2'''-H_a), 3.25 (d, *J* = 17.3 Hz, 1 H, 2'''-H_b), 2.88 (dd, *J* = 12.6, 4.5 Hz, 1 H, 3'-H_a), 2.56 (dd, *J* = 12.6, 9.3 Hz, 1 H, 3'-H_b), 2.43 (s, 3 H, NCH_3), 2.12-2.02 (m, 2 H, 3-*H*, 3''-*H*), 1.19 (dd, *J* = 7.2, 7.1 Hz, 3 H, Et-CH_3), 0.92-0.84 (m, 12 H, 4-*H*, 4''-*H*).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.1, 171.5, 171.3, 171.0 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.4 (N*C*(=O)O), 137.2, 136.5, 136.3 (3 x C_{Ar}), 129.0, 128.7, 128.6, 128.3, 128.2, 128.2, 128.0, 127.6, 127.4 (15 x HC_{Ar}), 67.1 (Bn-*C*H_2), 60.8 (Et-*C*H_2), 60.2 (*C*-2), 58.3, 58.2 (*C*-2''', *C*-3'), 54.9 (*C*-2''), 50.6 (*C*-2'), 50.0, 47.9 (2 x Bn-*C*H_2), 42.7 (N*C*H_3), 31.7, 30.9 (*C*-3, *C*-3''), 20.0, 19.3, 17.7, 17.3 (2 x *C*-4, 2 x *C*-4''), 14.4 (Et-*C*H_3).

IR (ATR): ν = 3288, 2963, 1720, 1630, 1532, 1448, 1235, 1027, 697.

MS (ESI^+): m/z = 716.4 $[M+H]^+$, calculated: 716.4018 $[M+H]^+$,
$C_{40}H_{53}N_5O_7$ (715.89 g(mol)$^{-1}$), found: 716.3996 [-3.1 ppm] (ESI^+-HRMS).



Preparation of sarcosine tripeptide conjugate **38**

To a solution of tripeptide **5** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), sarcosine ethyl ester (9.4 mg, 0.080 mmol) was added. After stirring for 40 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 70:30:0 → 97:0:3) to give 25 mg (0.037 mmol, 93%) of the title compound as a colourless solid.

TLC: R_f = 0.31 (DCM:AcOEt, 50:50).

Melting point: T_{mp} = 106 °C.

Specific rotation: $[\alpha]_D^{20}$ = -17.2 (c = 2.19, $CHCl_3$)

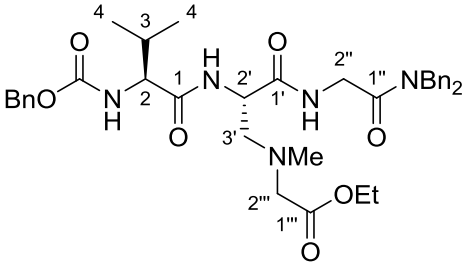
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.30 (d, *J* = 8.8 Hz, 1 H, 2''-N*H*), 7.31-7.08 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.45 (s, 1 H, 2-N*H*), 5.05 (s, 2 H, Bn-CH_2), 4.81 (d, *J* = 15.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.78 (dd, *J* = 8.8, 5.9 Hz, 1 H, 2''-*H*), 4.56 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.35 (d, *J* = 16.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.28 (ddd, *J* = 9.3, 5.0, 4.8 Hz, 1 H, 2'-*H*), 4.14 (d, *J* = 15.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.12-4.07 (m, 2 H, Et-CH_2), 3.92-3.82 (m, 2 H, 2-*H*), 3.49 (d, *J* = 17.3 Hz, 1 H, 2'''-H_a), 3.25 (d, *J* = 17.3 Hz, 1 H, 2'''-H_b), 2.88 (dd, *J* = 12.6, 4.8 Hz, 1 H, 3'-H_a), 2.57 (dd, *J* = 12.6, 9.3 Hz, 1 H, 3'-H_b), 2.43 (s, 3 H, NCH_3), 2.05 (dqq, *J* = 6.7, 6.7,

5.9 Hz, 1 H, 3"-*H*), 1.19 (dd, *J* = 7.2, 7.2 Hz, 3 H, Et-C*H*$_3$), 0.90 (d, *J* = 6.7 Hz, 3 H, 4"-*H*$_a$), 0.84 (d, *J* = 6.7 Hz, 3 H, 4"-*H*$_b$).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.0, 171.3, 171.1, 169.1 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.5 (N*C*(=O)O), 137.1, 136.4, 136.3 (3 x *C*$_{Ar}$), 129.0, 128.8, 128.6, 128.3, 128.3, 128.2, 128.0, 127.6, 127.4 (15 x H*C*$_{Ar}$), 67.2 (Bn-*C*H$_2$), 60.9 (Et-*C*H$_2$), 58.4, 58.2 (*C*-2'", *C*-3'), 54.9 (*C*-2"), 50.7 (*C*-2'), 50.0, 47.9 (2 x Bn-*C*H$_2$), 44.4 (*C*-2), 42.7 (N*C*H$_3$), 30.9 (*C*-3"), 20.0, 17.2 (2 x *C*-4"), 14.4 (Et-*C*H$_3$).

IR (ATR): ν = 3270, 1720, 1631, 1507, 1443, 1220, 1040, 733, 701.

MS (ESI$^+$): m/z = 674.4 [M+H]$^+$, calculated: 674.3548 [M+H]$^+$,
$C_{37}H_{47}N_5O_7$ (673.81 g(mol)$^{-1}$), found: 674.3525 [-3.5 ppm] (ESI$^+$-HRMS).



Preparation of sarcosine tripeptide conjugate **39**

To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), sarcosine ethyl ester (9.4 mg, 0.080 mmol) was added. After stirring for 40 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 80:20:0 → 98:0:2) to give 26 mg (0.039 mmol, 98%) of the title compound as a colourless solid.

TLC: R_f = 0.37 (DCM:AcOEt, 70:30).

Melting point: T_{mp} = 59 °C.

Specific rotation: $[\alpha]_D^{20}$ = +13.6 (c = 2.09, $CHCl_3$)

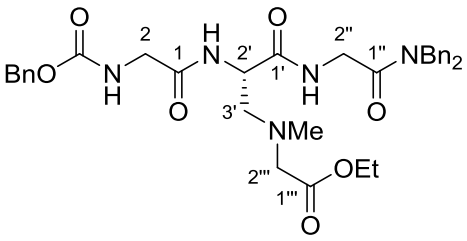
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.30 (dd, *J* = 4.7, 4.4 Hz, 1 H, 2"-N*H*), 7.30-7.07 (m, 16 H, 2'-N*H*, 15 x *H*C$_{Ar}$), 5.41 (d, *J* = 8.7 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.3 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.02 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.54 (d, *J* = 14.9 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.50 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.32-4.21 (m, 4 H, 2'-*H*, 2"-*H*$_a$, Bn-C*H*$_2$), 4.12-4.06 (m, 3 H, 2-*H*, Et-C*H*$_2$), 4.02 (dd, *J* = 17.2, 4.4 Hz, 1 H, 2"-*H*$_b$), 3.36 (d, *J* = 17.3 Hz, 1 H, 2'"-*H*$_a$), 3.31 (d, *J* = 17.3 Hz, 1 H, 2'"-*H*$_b$), 2.89 (dd, *J* = 12.7, 5.3 Hz, 1 H, 3'-*H*$_a$), 2.65 (dd, *J* = 12.7, 8.6 Hz, 1 H, 3'-*H*$_b$), 2.41 (s, 3 H, NC*H*$_3$), 2.12 (dqq, *J* = 6.7, 6.6, 5.9 Hz, 1 H, 3-*H*), 1.18 (dd, *J* = 7.2, 7.2 Hz, 3 H, Et-C*H*$_3$), 0.92 (d, *J* = 6.6 Hz, 3 H, 4-*H*$_a$), 0.85 (d, *J* = 6.7 Hz, 3 H, 4-*H*$_b$).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.7, 171.7, 171.1, 168.6 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.5 (N*C*(=O)O), 136.7, 136.5, 135.6 (3 x *C*$_{Ar}$), 129.0, 128.8, 128.6, 128.4, 128.2, 128.2, 128.0, 127.7, 126.6 (15 x H*C*$_{Ar}$), 67.1 (Bn-*C*H$_2$), 60.9 (Et-*C*H$_2$), 60.2 (*C*-2), 58.9 (*C*-2'"), 58.3 (*C*-3'), 51.1 (*C*-2'), 49.0, 48.5 (2 x Bn-*C*H$_2$), 42.5 (N*C*H$_3$), 41.7 (*C*-2"), 31.6 (*C*-3), 19.3, 17.6 (2 x *C*-4), 14.3 (Et-*C*H$_3$).

IR (ATR): ν = 3289, 1719, 1631, 1520, 1452, 1222, 1027, 734, 697.

MS (ESI$^+$): m/z = 674.4 [M+H]$^+$, calculated: 674.3548 [M+H]$^+$,
$C_{37}H_{47}N_5O_7$ (673.81 g(mol)$^{-1}$), found: 674.3524 [-3.6 ppm] (ESI$^+$-HRMS).

Preparation of sarcosine tripeptide conjugate **40**



To a solution of tripeptide **7** (21 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), sarcosine ethyl ester (9.4 mg, 0.080 mmol) was added. After stirring for 40 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt:MeOH, 70:30:0 → 97:0:3) to give 24 mg (0.038 mmol, 95%) of the title compound as a colourless solid.

TLC: R_f = 0.12 (DCM:AcOEt, 50:50).

Melting point: T_{mp} = 62 °C.

Specific rotation: $[\alpha]_D^{20}$ = +5.7 (c = 1.86, $CHCl_3$)

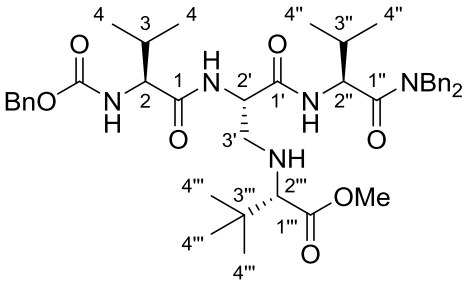
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.24 (dd, *J* = 5.0, 4.3 Hz, 1 H, 2"-N*H*), 7.35 (d, *J* = 5.2 Hz, 1 H, 2'-N*H*), 7.31-7.07 (m, 15 H, 15 x *H*C_{Ar}), 5.49 (dd, *J* = 5.7, 5.2 Hz, 1 H, 2-N*H*), 5.05 (s, 2 H, Bn-C*H*$_2$), 4.54 (d, *J* = 14.9 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.50 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.34-4.28 (m, 3 H, 2'-*H*, Bn-C*H*$_2$), 4.20 (dd, *J* = 17.2, 5.0 Hz, 1 H, 2"-*H*$_a$), 4.10-4.05 (m, 2 H, Et-C*H*$_2$), 4.02 (dd, *J* = 17.2, 4.3 Hz, 1 H, 2"-*H*$_b$), 3.91-3.87 (m, 2 H, 2-*H*), 3.34 (d, *J* = 17.4 Hz, 1 H, 2'''-*H*$_a$), 3.30 (d, *J* = 17.4 Hz, 1 H, 2'''-*H*$_b$), 2.89 (dd, *J* = 12.7, 5.9 Hz, 1 H, 3'-*H*$_a$), 2.66 (dd, *J* = 12.7, 8.4 Hz, 1 H, 3'-*H*$_b$), 2.39 (s, 3 H, NC*H*$_3$), 1.18 (dd, *J* = 7.2, 7.2 Hz, 3 H, Et-C*H*$_3$).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 171.8, 171.1, 169.4, 168.6 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.6 (N*C*(=O)O), 136.6, 136.4, 135.5 (3 x *C*$_{Ar}$), 129.2, 128.8, 128.6, 128.4, 128.2, 128.2, 128.0, 127.8, 126.6 (15 x H*C*$_{Ar}$), 67.2 (Bn-*C*H$_2$), 61.0 (Et-*C*H$_2$), 58.9 (*C*-2'''), 58.3 (*C*-3'), 51.2 (*C*-2'), 49.1, 48.6 (2 x Bn-*C*H$_2$), 44.5 (*C*-2), 42.6 (N*C*H$_3$), 41.6 (*C*-2"), 14.3 (Et-*C*H$_3$).

IR (ATR): ν = 3290, 1717, 1630, 1521, 1452, 1217, 1028, 735, 696.

MS (ESI$^+$): m/z = 632.3 $[M+H]^+$, calculated: 632.3079 $[M+H]^+$,
$C_{34}H_{41}N_5O_7$ (631.73 g(mol)$^{-1}$), found: 632.3058 [-3.3 ppm] (ESI$^+$-HRMS).

Preparation of *tert*-leucine tripeptide conjugate **41**



To a solution of tripeptide **4** (24 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), L-*tert*-leucine methyl ester (12 mg, 0.080 mmol) was added. After stirring for 20 h at 40 °C and for 70 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 80:20) to give 23 mg (0.031 mmol, 78%) of the title compound as a colourless solid.

TLC: R_f = 0.32 (DCM:AcOEt, 80:20).

Melting point: T_{mp} = 118 °C.

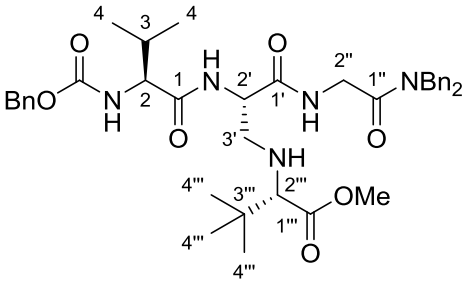
Specific rotation: $[\alpha]_D^{20}$ = -37.0 (c = 1.82, $CHCl_3$)

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.56 (d, *J* = 9.0 Hz, 1 H, 2"-N*H*), 7.31-7.08 (m, 15 H, 15 x *H*C_{Ar}), 6.90 (d, *J* = 6.6 Hz, 1 H, 2'-N*H*), 5.42 (d, *J* = 7.4 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.2 Hz, 1 H, Bn-C$H_a$$H_b$), 5.00 (d, *J* = 12.2 Hz, 1 H, Bn-C$H_a$$H_b$), 4.81 (dd, *J* = 9.0, 6.6 Hz, 1 H, 2"-*H*), 4.75 (d, *J* = 14.7 Hz, 1 H, Bn-C$H_a$$H_b$), 4.53 (d, *J* = 16.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.38 (d, *J* = 16.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.36-4.30 (m, 1 H, 2'-*H*), 4.20 (d, *J* = 14.7 Hz, 1 H, Bn-C$H_a$$H_b$), 4.00 (dd, *J* = 7.4, 6.2 Hz, 1 H, 2-*H*), 3.63 (s, 3 H, OCH_3), 2.98 (dd, *J* = 11.9, 2.7 Hz, 1 H, 3'-H_a), 2.86 (s, 1 H, 2'"-*H*), 2.45 (dd, *J* = 11.9, 7.0 Hz, 1 H, 3'-H_b), 2.13-2.01 (m, 2 H, 3-*H*, 3"-*H*), 0.91-0.81 (m, 21 H, 4-*H*, 4"-*H*, 4'"-*H*). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (101 MHz, $CDCl_3$): δ = 175.2, 171.9, 171.2, 170.5 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.5 (N*C*(=O)O), 137.2, 136.4, 136.3 (3 x C_{Ar}), 129.0, 128.8, 128.6, 128.4, 128.3, 128.2, 127.9, 127.6, 127.3 (15 x HC_{Ar}), 71.2 (*C*-2'"), 67.2 (Bn-*C*H_2), 60.5 (*C*-2), 54.6 (*C*-2"), 52.7 (*C*-2'), 51.5 (O*C*H_3), 50.2 (*C*-3'), 50.0, 48.0 (2 x Bn-*C*H_2), 34.3 (*C*-3'"), 31.4 (*C*-3, *C*-3"), 27.0 (3 x *C*-4'"), 19.9, 19.3, 17.8, 17.7 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 2962, 1731, 1630, 1528, 1448, 1217, 1155, 1028, 697.

MS (ESI^+): m/z = 744.5 $[M+H]^+$, calculated: 766.4150 $[M+Na]^+$,
$C_{42}H_{57}N_5O_7$ (743.95 g(mol)$^{-1}$), found: 766.4134 [-2.1 ppm] (ESI^+-HRMS).



Preparation of *tert*-leucine tripeptide conjugate **42**

To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), L-*tert*-leucine methyl ester (12 mg, 0.080 mmol) was added. After stirring for 20 h at 40 °C and for 70 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 70:30) to give 21 mg (0.030 mmol, 75%) of the title compound as a colourless solid.

TLC: R_f = 0.31 (DCM:AcOEt, 70:30).

Melting point: T_{mp} = 156 °C.

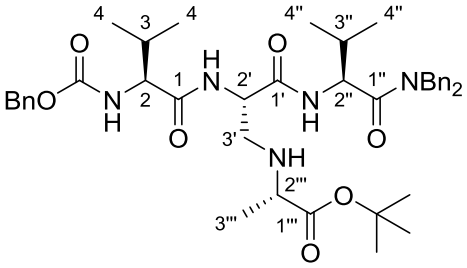
Specific rotation: $[\alpha]_D^{20}$ = -5.8 (c = 1.65, $CHCl_3$)

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.71 (dd, *J* = 4.4, 4.3 Hz, 1 H, 2"-N*H*), 7.30-7.05 (m, 15 H, 15 x *H*C_{Ar}), 6.98 (d, *J* = 6.7 Hz, 1 H, 2'-N*H*), 5.36 (d, *J* = 7.8 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.2 Hz, 1 H, Bn-C$H_a$$H_b$), 5.01 (d, *J* = 12.2 Hz, 1 H, Bn-C$H_a$$H_b$), 4.52 (s, 2 H, Bn-C$H_2$), 4.41-4.35 (m, 1 H, 2'-*H*), 4.31 (s, 2 H, Bn-CH_2), 4.11-4.00 (m, 3 H, 2-*H*, 2"-*H*), 3.63 (s, 3 H, OCH_3), 3.05 (dd, *J* = 11.8, 3.2 Hz, 1 H, 3'-H_a), 2.84 (s, 1 H, 2'"-*H*), 2.48 (dd, *J* = 11.8, 6.9 Hz, 1 H, 3'-H_b), 2.11 (dqq, *J* = 6.8, 6.7, 5.8 Hz, 1 H, 3-*H*), 0.92-0.85 (m, 15 H, 4-*H*, 4'"-*H*). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (101 MHz, $CDCl_3$): δ = 174.9, 171.3, 170.8, 168.4 (*C*-1, *C*-1', *C*-1", *C*-1'"), 156.6 (N*C*(=O)O), 136.7, 136.4, 135.6 (3 x C_{Ar}), 129.2, 128.8, 128.7, 128.5, 128.3, 128.2, 128.0, 127.8, 126.6 (15 x HC_{Ar}), 71.3 (*C*-2'"), 67.2 (Bn-*C*H_2), 60.5 (*C*-2), 52.6 (*C*-2'), 51.5 (O*C*H_3), 50.1 (*C*-3'), 49.1, 48.7 (2 x Bn-*C*H_2), 41.6 (*C*-2"), 34.3 (*C*-3'"), 31.3 (*C*-3), 26.9 (3 x *C*-4'"), 19.3, 17.8 (2 x *C*-4).

IR (ATR): ν = 3298, 1715, 1631, 1520, 1218, 1155, 1028, 752, 698.
MS (ESI$^+$): m/z = 724.3 $[M+Na]^+$, calculated: 724.3681 $[M+Na]^+$,
$C_{39}H_{51}N_5O_7$ (701.87 g(mol)$^{-1}$), found: 724.3667 [-1.9 ppm] (ESI$^+$-HRMS).

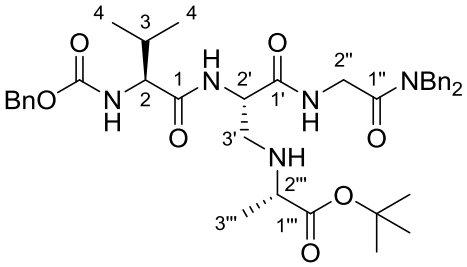
Preparation of alanine tripeptide conjugate **43**



To a solution of tripeptide **4** (24 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), alanine *tert*-butyl ester (12 mg, 0.080 mmol) was added. After stirring for 48 h at 40 °C and 20 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 70:30 → 60:40) to give 28 mg (0.038 mmol, 95%) of the title compound as a colourless solid.

TLC: R_f = 0.20 (DCM:AcOEt, 70:30).
Melting point: T_{mp} = 66 °C.
Specific rotation: $[\alpha]_D^{20}$ = -29.6 (c = 2.33, $CHCl_3$)
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.28 (d, *J* = 8.9 Hz, 1 H, 2''-N*H*), 7.31-7.08 (m, 15 H, 15 x *H*C_{Ar}), 6.96 (d, *J* = 6.2 Hz, 1 H, 2'-N*H*), 5.40 (d, *J* = 8.6 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.2 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.01 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.83-4.78 (m, 2 H, 2''-*H*, Bn-C*H*$_a$H$_b$), 4.55 (d, *J* = 16.5 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.35 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.30-4.25 (m, 1 H, 2'-*H*), 4.17 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.04 (dd, *J* = 8.6, 5.5 Hz, 1 H, 2-*H*), 3.20 (q, *J* = 7.0 Hz, 1 H, 2'''-*H*), 3.11 (dd, *J* = 12.0, 3.6 Hz, 1 H, 3'-*H*$_a$), 2.48 (dd, *J* = 12.0, 9.0 Hz, 1 H, 3'-*H*$_b$), 2.12-1.97 (m, 2 H, 3-*H*, 3''-*H*), 1.39 (s, 9 H, C(C*H*$_3$)$_3$), 1.24 (d, *J* = 7.0 Hz, 3 H, 3'''-*H*), 0.91-0.83 (m, 12 H, 4-*H*, 4''-*H*). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.
^{13}C NMR (126 MHz, $CDCl_3$): δ = 175.2, 172.1, 171.2, 170.9 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.5 (N*C*(=O)O), 137.1, 136.5, 136.2 (3 x *C*$_{Ar}$), 129.0, 128.8, 128.6, 128.4, 128.2, 128.2, 128.0, 127.6, 127.3 (15 x H*C*$_{Ar}$), 81.4 (*C*(CH$_3$)$_3$), 67.1 (Bn-*C*H$_2$), 60.3 (*C*-2), 57.7 (*C*-2'''), 54.7 (*C*-2''), 52.3 (*C*-2'), 50.0 (Bn-*C*H$_2$), 49.5 (*C*-3'), 47.9 (Bn-*C*H$_2$), 31.6, 31.2 (*C*-3, *C*-3''), 28.2 (C(*C*H$_3$)$_3$), 20.0, 19.4, 19.4, 17.7, 17.4 (*C*-3''', 2 x *C*-4, 2 x *C*-4'').
IR (ATR): ν = 3291, 1726, 1632, 1530, 1449, 1216, 1149, 1028, 698.
MS (ESI$^+$): m/z = 744.4 $[M+H]^+$, calculated: 744.4331 $[M+H]^+$,
$C_{42}H_{57}N_5O_7$ (743.95 g(mol)$^{-1}$), found: 744.4324 [-0.9 ppm] (ESI$^+$-HRMS).

Preparation of alanine tripeptide conjugate **44**



To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), alanine *tert*-butyl ester (12 mg, 0.080 mmol) was added. After stirring for 48 h at 40 °C and 20 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 70:30 → 60:40) to give 21 mg (0.030 mmol, 75%) of the title compound as a

colourless solid.

TLC: R_f = 0.18 (DCM:AcOEt, 60:40).

Melting point: T_{mp} = 57 °C.

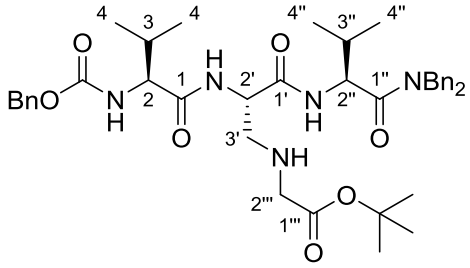
Specific rotation: $[\alpha]_D^{20}$ = -3.8 (c = 1.58, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.07 (s, 1 H, 2''-N*H*), 7.31-7.06 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.40 (d, *J* = 8.5 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.2 Hz, 1 H, Bn-C$H_a$$H_b$), 5.01 (d, *J* = 12.2 Hz, 1 H, Bn-C$H_a$$H_b$), 4.56 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.51 (d, *J* = 14.8 Hz, 1 H, Bn-C$H_a$$H_b$), 4.36-4.30 (m, 3 H, 2'-*H*, Bn-CH_2), 4.16 (dd, *J* = 17.4, 4.7 Hz, 1 H, 2''-H_a), 4.09-4.04 (m, 2 H, 2-*H*, 2''-H_b), 3.19-3.14 (m, 2 H, 2'''-*H*, 3'-H_a), 2.53 (dd, *J* = 12.0, 7.4 Hz, 1 H, 3'-H_b), 2.11 (dqq, *J* = 6.7, 6.7, 6.1 Hz, 1 H, 3-*H*), 1.38 (s, 9 H, C(CH_3)$_3$), 1.24 (d, *J* = 7.0 Hz, 3 H, 3'''-*H*), 0.92 (d, *J* = 6.7 Hz, 3 H, 4-H_a), 0.86 (d, *J* = 6.7 Hz, 3 H, 4-H_b). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 174.9, 171.4, 170.8, 168.4 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.6 (N*C*(=O)O), 136.7, 136.4, 135.5 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.5, 128.3, 128.2, 128.1, 127.8, 126.6 (15 x HC_{Ar}), 81.4 (*C*(CH_3)$_3$), 67.2 (Bn-*C*H_2), 60.4 (*C*-2), 57.7 (*C*-2'''), 52.6 (*C*-2'), 49.1 (Bn-*C*H_2), 49.0 (*C*-3'), 48.6 (Bn-*C*H_2), 41.7 (*C*-2''), 31.5 (*C*-3), 28.2 (C(*C*H_3)$_3$), 19.4, 19.2 (*C*-3''', C_a-4), 17.7 (C_b-4).

IR (ATR): ν = 3317, 1638, 1519, 1453, 1240, 1151, 1043, 744, 697.

MS (ESI^+): m/z = 702.4 $[M+H]^+$, calculated: 702.3861 $[M+H]^+$,
$C_{39}H_{51}N_5O_7$ (701.87 g(mol)$^{-1}$), found: 702.3855 [-0.9 ppm] (ESI^+-HRMS).



Preparation of glycine tripeptide conjugate **45**

To a solution of tripeptide **4** (24 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), glycine *tert*-butyl ester (10 mg, 0.080 mmol) was added. After stirring for 40 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 80:20 → 50:50) to give 26 mg (0.036 mmol, 90%) of the title compound as a colourless solid.

TLC: R_f = 0.27 (DCM:AcOEt, 60:40).

Melting point: T_{mp} = 84 °C.

Specific rotation: $[\alpha]_D^{20}$ = -37.2 (c = 1.89, $CHCl_3$)

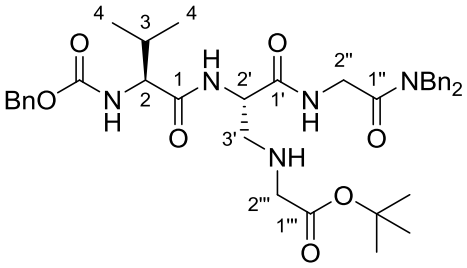
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.14 (d, *J* = 9.0 Hz, 1 H, 2''-N*H*), 7.31-7.09 (m, 16 H, 2'-N*H*, 15 x *H*C_{Ar}), 5.46 (d, *J* = 8.8 Hz, 1 H, 2-N*H*), 5.05 (d, *J* = 12.4 Hz, 1 H, Bn-C$H_a$$H_b$), 5.02 (d, *J* = 12.4 Hz, 1 H, Bn-C$H_a$$H_b$), 4.84-4.71 (m, 2 H, 2''-*H*, Bn-C$H_a$$H_b$), 4.55 (d, *J* = 17.5 Hz, 1 H, Bn-C$H_a$$H_b$), 4.34-4.28 (m, 2 H, 2'-*H*, Bn-C$H_a$$H_b$), 4.13 (d, *J* = 14.9 Hz, 1 H, Bn-C$H_a$$H_b$), 4.10 (dd, *J* = 9.0, 5.7 Hz, 1 H, 2-*H*), 3.37 (d, *J* = 17.7 Hz, 1 H, 2'''-H_a), 3.27 (d, *J* = 17.7 Hz, 1 H, 2'''-H_b), 3.13 (dd, *J* = 12.5, 3.3 Hz, 1 H, 3'-H_a), 2.55 (dd, *J* = 12.5, 7.9 Hz, 1 H, 3'-H_b), 2.14-2.01 (m, 2 H, 3-*H*, 3''-*H*), 1.39 (s, 9 H, C(CH_3)$_3$), [0.92 (d, *J* = 6.8 Hz, 3 H), 0.87 (d, *J* = 6.8 Hz, 3 H), 0.85 (d, *J* = 6.8 Hz, 3 H), 0.84 (d, *J* = 6.7 Hz, 3 H) (4-H_a, 4-H_b,

4''-H_a, 4''-H_b)]. The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.5, 172.0, 171.5, 171.0 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.5 (N*C*(=O)O), 137.1, 136.5, 136.2 (3 x C_{Ar}), 129.1, 128.8, 128.6, 128.4, 128.2, 128.1, 128.0, 127.6, 127.3 (15 x HC_{Ar}), 81.7 (*C*(CH_3)$_3$), 67.0 (Bn-*C*H_2), 60.2 (*C*-2), 54.7 (*C*-2''), 52.5 (*C*-2'), 51.3 (*C*-2'''), 50.8 (*C*-3'), 49.9, 47.9 (2 x Bn-*C*H_2), 31.6, 31.1 (*C*-3, *C*-3''), 28.2 (C(*C*H_3)$_3$), 20.0, 19.4, 17.6, 17.3 (2 x *C*-4, 2 x *C*-4'').

IR (ATR): ν = 3285, 1630, 1533, 1452, 1368, 1223, 1152, 1029, 697.

MS (ESI^+): m/z = 730.4 $[M+H]^+$, calculated: 752.3994 $[M+Na]^+$,
$C_{41}H_{55}N_5O_7$ (729.92 g(mol)$^{-1}$), found: 752.3965 [-3.8 ppm] (ESI^+-HRMS).



Preparation of glycine tripeptide conjugate **46**

To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), glycine *tert*-butyl ester (10 mg, 0.080 mmol) was added. After stirring for 40 h at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 6.0 cm, DCM:AcOEt, 70:30 → 40:60) to give 22 mg (0.032 mmol, 80%) of the title compound as a colourless solid.

TLC: R_f = 0.22 (DCM:AcOEt, 50:50).

Melting point: T_{mp} = 79 °C.

Specific rotation: $[\alpha]_D^{20}$ = -8.0 (c = 1.55, $CHCl_3$)

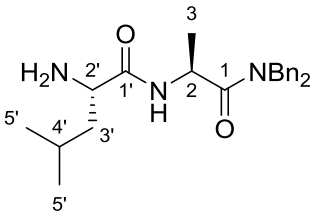
^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.93 (dd, *J* = 4.3, 4.2 Hz, 1 H, 2''-N*H*), 7.57 (d, *J* = 6.3 Hz, 1 H, 2'-N*H*), 7.31-7.07 (m, 15 H, 15 x *H*C_{Ar}), 5.50 (d, *J* = 8.6 Hz, 1 H, 2-N*H*), 5.04 (s, 2 H, Bn-C*H*$_2$), 4.55 (d, *J* = 14.8 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.50 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.36-4.28 (m, 3 H, 2'-*H*, Bn-C*H*$_2$), 4.14-4.10 (m, 3 H, 2-*H*, 2''-*H*), 3.29 (s, 2 H, 2'''-*H*), 3.24 (dd, *J* = 12.3, 2.7 Hz, 1 H, 3'-*H*$_a$), 2.57 (dd, *J* = 12.3, 6.6 Hz, 1 H, 3'-*H*$_b$), 2.17 (dqq, *J* = 6.7, 6.6, 5.9 Hz, 1 H, 3-*H*), 1.37 (s, 9 H, C(C*H*$_3$)$_3$), 0.93 (d, *J* = 6.6 Hz, 3 H, 4-*H*$_a$), 0.86 (d, *J* = 6.7 Hz, 3 H, 4-*H*$_b$). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.3, 171.7, 171.1, 168.6 (*C*-1, *C*-1', *C*-1'', *C*-1'''), 156.7 (N*C*(=O)O), 136.7, 136.5, 135.5 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.5, 128.2, 128.0, 127.8, 126.6 (15 x HC_{Ar}), 81.7 (*C*(CH_3)$_3$), 67.1 (Bn-*C*H_2), 60.4 (*C*-2), 52.8 (*C*-2'), 51.1 (*C*-2'''), 50.5 (*C*-3'), 49.1, 48.6 (2 x Bn-*C*H_2), 41.7 (*C*-2''), 31.4 (*C*-3), 28.2 (C(*C*H_3)$_3$), 19.5, 17.5 (2 x *C*-4).

IR (ATR): ν = 3290, 1714, 1630, 1521, 1219, 1151, 1028, 734, 697.

MS (ESI^+): m/z = 688.4 $[M+H]^+$, calculated: 688.3705 $[M+H]^+$,
$C_{38}H_{49}N_5O_7$ (687.84 g(mol)$^{-1}$), found: 688.3678 [-3.9 ppm] (ESI^+-HRMS).

Preparation of dipeptide **47**



To a solution of *N*-(*tert*-Butoxycarbonyl)-L-leucine (1.29 g, 5.58 mmol) in abs. DMF (10 mL), HOBt (754 mg, 5.58 mmol) was

added. EDAC (1.07 g, 5.58 mmol) was added after cooling the solution to 0 °C. After stirring for 5 min at 0 °C NEt_3 (0.78 mL, 5.6 mmol) was added. L-Alanine dibenzylamide[S6] (1.50 g, 5.58 mmol) in abs. DMF (5 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 15 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (200 mL) and washed with water (2 x 200 mL), 10 wt% aqueous citric acid solution (200 mL) and sat. aqueous $NaHCO_3$ solution (200 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resulting dipeptide was used in the subsequent transformation without further purification.

The crude product (calculated maximal amount of substance: 5.58 mmol) was dissolved in AcOEt (30 mL) and cooled to 0 °C. MeOH (4.53 mL, 112 mmol) and AcCl (3.98 mL, 55.8 mmol) were added at 0 °C. After stirring for 1 h at 0 °C and 4 h at rt the reaction mixture was diluted with AcOEt (150 mL) and washed with 1 M aqueous NaOH solution (150 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (150 g, 4.5 x 18 cm, DCM:MeOH, 95:5) to give 2.08 g (5.45 mmol, 98%) of the title compound as a colourless oil.

TLC: R_f = 0.22 (DCM:MeOH, 93:7).

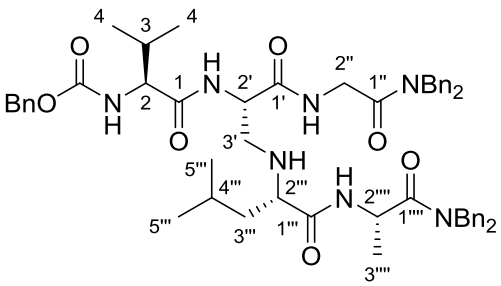
Specific rotation: $[\alpha]_D^{20}$ = -74.0 (c = 2.51, $CHCl_3$)

^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.02 (d, *J* = 8.0 Hz, 1 H, 2-N*H*), 7.41-7.19 (m, 10 H, 10 x *H*C_{Ar}), 5.04 (dq, *J* = 8.0, 6.8 Hz, 1 H, 2-*H*), 4.87 (d, *J* = 14.8 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.61 (d, *J* = 16.7 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.50 (d, *J* = 16.7 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.34 (d, *J* = 14.8 Hz, 1 H, Bn-CH$_a$*H*$_b$), 3.36 (dd, *J* = 9.8, 4.0 Hz, 1 H, 2'-*H*), 1.79-1.66 (m, 2 H, 3'-*H*$_a$, 4'-*H*), 1.43-1.36 (m, 4 H, 3-*H*, 3'-*H*$_b$), 0.99 (d, *J* = 6.4 Hz, 3 H, 5'-*H*$_a$), 0.95 (d, *J* = 6.3 Hz, 3 H, 5'-*H*$_b$). The signal attributed to the primary amine N*H*$_2$ protons was not observed in the ^{1}H NMR.

^{13}C NMR (101 MHz, $CDCl_3$): δ = 175.2, 173.4 (*C*-1, *C*-1'), 136.9, 136.0 (2 x *C*$_{Ar}$), 129.1, 128.8, 128.2, 127.9, 127.6, 127.0 (10 x H*C*$_{Ar}$), 53.6 (*C*-2'), 49.8, 48.1 (2 x Bn-*C*H$_2$), 45.1 (*C*-2), 44.1 (*C*-3'), 24.9 (*C*-4'), 23.6, 21.5 (2 x *C*-5'), 19.2 (*C*-3).

IR (ATR): ν = 2954, 1637, 1495, 1451, 1365, 1220, 1079, 732, 698.

MS (ESI$^+$): m/z = 382.3 $[M+H]^+$, calculated: 382.2489 $[M+H]^+$,
$C_{23}H_{31}N_3O_2$ (381.52 g(mol)$^{-1}$), found: 382.2489 [-0.1 ppm] (ESI$^+$-HRMS).



Preparation of dipeptide tripeptide conjugate **48**

To a solution of tripeptide **5** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), dipeptide **47** (23 mg, 0.060 mmol) was added. After stirring for 4 d at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 70:30 → 50:50) to give 28 mg (0.030 mmol, 75%) of the title compound as a colourless solid.

TLC: R_f = 0.20 (DCM:AcOEt, 50:50).

Melting point: T_{mp} = 65 °C.

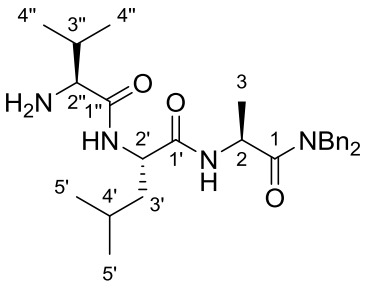
Specific rotation: $[\alpha]_D^{20}$ = -38.3 (c = 2.10, $CHCl_3$)

^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.09 (d, *J* = 8.6 Hz, 1 H, 2''''-N*H*), 7.98 (d, *J* = 7.4 Hz, 1 H, 2'-N*H*), 7.49 (dd, *J* = 4.3, 4.0 Hz, 1 H, 2''-N*H*), 7.34-7.02 (m, 25 H, 25 x *H*C_{Ar}), 5.80 (d, *J* = 9.0 Hz, 1 H, 2-N*H*), 5.06 (dd, *J* = 8.6, 6.9 Hz, 1 H, 2''''-*H*), 5.03 (d, *J* = 12.3 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.96 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.78 (d, *J* = 14.9 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.58 (ddd, *J* = 10.2, 7.4, 3.4 Hz, 1 H, 2'-*H*), 4.54 (d, *J* = 16.5 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.52 (d, *J* = 14.7 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.44 (d, *J* = 14.7 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.38 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.28 (s, 2 H, Bn-C*H*$_2$), 4.24 (dd, *J* = 9.0, 6.1 Hz, 1 H, 2-*H*), 4.13 (dd, *J* = 17.3, 4.3 Hz, 1 H, 2''-*H*$_a$), 4.12 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$*H*$_b$), 3.91 (dd, *J* = 17.3, 4.0 Hz, 1 H, 2''-*H*$_b$), 3.14-3.10 (m, 2 H, 2'''-*H*, 3'-*H*$_a$), 2.64 (dd, *J* = 11.8, 10.2 Hz, 1 H, 3'-*H*$_b$), 2.17 (dqq, *J* = 6.7, 6.7, 6.1 Hz, 1 H, 3-*H*), 1.62 (ddqq, *J* = 7.9, 6.6, 6.5, 6.4 Hz, 1 H, 4'''-*H*), 1.49 (ddd, *J* = 14.0, 7.9, 5.3 Hz, 1 H, 3'''-*H*$_a$), 1.38 (ddd, *J* = 14.0, 8.7, 6.4 Hz, 1 H, 3'''-*H*$_b$), 1.26 (d, *J* = 6.9 Hz, 3 H, 3''''-*H*), [0.92 (d, *J* = 6.7 Hz, 3 H), 0.87 (d, *J* = 6.5 Hz, 3 H), 0.86 (d, *J* = 6.7 Hz, 3 H), 0.84 (d, *J* = 6.6 Hz, 3 H) (4-*H*$_a$, 4-*H*$_b$, 5'''-*H*$_a$, 5'''-*H*$_b$)]. The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 174.8, 174.7, 172.9, 170.2, 168.5 (*C*-1, *C*-1', *C*-1'', *C*-1''', *C*-1''''), 156.7 (N*C*(=O)O), 136.6, 136.5, 135.6, 135.4 (5 x *C*$_{Ar}$), 129.2, 129.2, 128.9, 128.8, 128.6, 128.4, 128.2, 128.2, 128.1, 128.0, 127.8, 127.7, 127.4, 126.7 (25 x H*C*$_{Ar}$), 67.0 (Bn-*C*H$_2$), 62.0 (*C*-2'''), 60.5 (*C*-2), 54.8 (*C*-2'), 50.1 (Bn-*C*H$_2$), 49.4 (*C*-3'), 49.1, 48.6, 48.2 (3 x Bn-*C*H$_2$), 44.6 (*C*-2''''), 42.9 (*C*-3'''), 41.6 (*C*-2''), 31.4 (*C*-3), 25.2 (*C*-4'''), 23.3, 22.1, 19.8, 18.8, 17.9 (*C*-3'''', 2 x *C*-4, 2 x *C*-5''').

IR (ATR): ν = 3292, 2956, 1633, 1496, 1451, 1218, 1028, 732, 697.

MS (ESI^+): m/z = 938.5 $[M+H]^+$, calculated: 960.4994 $[M+Na]^+$,
$C_{55}H_{67}N_7O_7$ (938.18 g(mol)$^{-1}$), found: 960.4992 [-0.2 ppm] (ESI^+-HRMS).



Preparation of tripeptide **49**

To a solution of *N*-(*tert*-Butoxycarbonyl)-L-valine (256 mg, 1.18 mmol) in abs. DMF (5 mL), HOBt (159 mg, 1.18 mmol) was added. EDAC (226 mg, 1.18 mmol) was added after cooling the solution to 0 °C. After stirring for 5 min at 0 °C NEt_3 (0.17 mL, 1.2 mmol) was added. Dipeptide **47** (450 mg, 1.18 mmol) in abs. DMF (3 mL) was added after additional 15 min of stirring at 0 °C. After stirring for 15 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (150 mL) and washed with water (2 x 100 mL), 10 wt% aqueous citric acid solution (200 mL) and sat. aqueous $NaHCO_3$ solution (200 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resulting tripeptide was used in the subsequent transformation without further purification.

AcOEt (6 mL) was added to the crude product (calculated maximal amount of substance: 1.18 mmol) and the resulting suspension was cooled to 0 °C. MeOH (0.955 mL, 23.6 mmol) and AcCl (0.842 mL, 11.8 mmol) were added at 0 °C. After stirring for 1 h at 0 °C and 3 h at rt the reaction mixture was diluted with AcOEt (150 mL) and washed with 1 M aqueous

NaOH solution (100 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (100 g, 4.5 x 12 cm, DCM:MeOH, 95:5 → 90:10) to give 360 mg (0.749 mmol, 63%) of the title compound as a colourless solid.

TLC: R_f = 0.22 (DCM:MeOH, 93:7).

Melting point: T_{mp} = 157 °C.

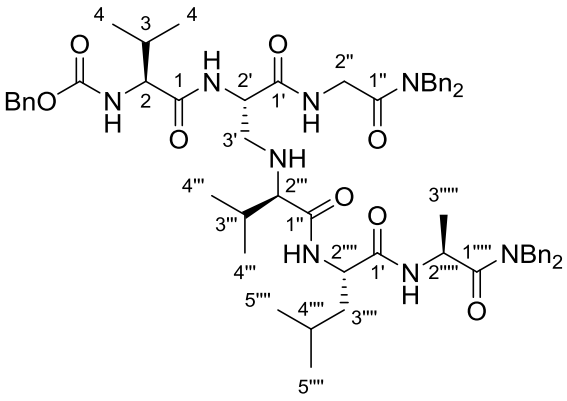
Specific rotation: $[\alpha]_D^{20}$ = -83.3 (c = 3.12, $CHCl_3$)

^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.65 (d, *J* = 8.5 Hz, 1 H, 2'-N*H*), 7.30-7.06 (m, 11 H, 2-N*H*, 10 x *H*C_{Ar}), 4.89 (dq, *J* = 7.2, 6.9 Hz, 1 H, 2-*H*), 4.68 (d, *J* = 14.9 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.47-4.38 (m, 3 H, 2'-*H*, Bn-C*H*$_2$), 4.31 (d, *J* = 14.9 Hz, 1 H, Bn-CH$_a$*H*$_b$), 3.21 (d, *J* = 3.8 Hz, 1 H, 2''-*H*), 2.27-2.20 (m, 1 H, 3''-*H*), 1.28-1.26 (m, 3 H, 3'-*H*, 4'-*H*), 1.27 (d, *J* = 6.9 Hz, 3 H, 3-*H*), 0.91 (d, *J* = 7.0 Hz, 3 H, 4''-*H*$_a$), 0.88 (d, *J* = 6.1 Hz, 3 H, 5'-*H*$_a$), 0.85 (d, *J* = 6.1 Hz, 3 H, 5'-*H*$_b$), 0.75 (d, *J* = 6.9 Hz, 3 H, 4''-*H*$_b$). The signal attributed to the primary amine N*H*$_2$ protons was not observed in the ^{1}H NMR.

^{13}C NMR (101 MHz, $CDCl_3$): δ = 174.6, 173.0, 171.5 (*C*-1, *C*-1', *C*-1''), 136.7, 135.9 (2 x *C*$_{Ar}$), 129.1, 128.8, 128.1, 128.0, 127.6, 126.9 (10 x H*C*$_{Ar}$), 60.2 (*C*-2''), 51.4 (*C*-2'), 49.7, 48.1 (2 x Bn-*C*H$_2$), 45.6 (*C*-2), 41.3 (*C*-3'), 30.8 (*C*-3''), 24.9 (*C*-4'), 23.2 (*C*$_a$-5'), 21.9 (*C*$_b$-5'), 19.8 (*C*$_a$-4''), 19.1 (*C*-3), 16.2 (*C*$_b$-4'').

IR (ATR): ν = 3267, 1637, 1540, 1428, 1221, 1078, 753, 719, 695.

MS (ESI$^+$): m/z = 481.3 $[M+H]^+$, calculated: 481.3173 $[M+H]^+$,
$C_{28}H_{40}N_4O_3$ (480.65 g(mol)$^{-1}$), found: 481.3171 [-0.5 ppm] (ESI$^+$-HRMS).



Preparation of tripeptide tripeptide conjugate **50**

To a solution of tripeptide **5** (33 mg, 0.060 mmol) in $CHCl_3$ (2.0 mL), tripeptide **49** (43 mg, 0.090 mmol) was added. After stirring for 4 d at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 50:50 → 30:70) to give 51 mg (0.049 mmol, 82%) of the title compound as a colourless solid.

TLC: R_f = 0.20 (DCM:AcOEt, 30:70).

Melting point: T_{mp} = 163 °C.

Specific rotation: $[\alpha]_D^{20}$ = -29.4 (c = 2.52, $CHCl_3$)

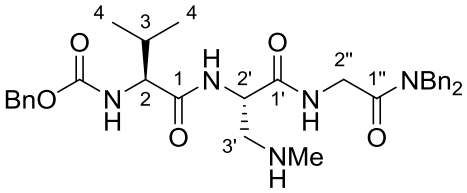
^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.84-7.81 (m, 2 H, 2'-N*H*, 2''-N*H*), 7.75 (d, *J* = 9.5 Hz, 1 H, 2''''-N*H*), 7.23-7.01 (m, 26 H, 2''''-N*H*, 25 x *H*C_{Ar}), 5.55 (d, *J* = 8.7 Hz, 1 H, 2-N*H*), 5.02-5.00 (m, 2 H, 2''''-*H*, Bn-C*H*$_a$H$_b$), 4.95 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.70-4.59 (m, 3 H, 2'-*H*, 2''''-*H*, Bn-C*H*$_a$H$_b$), 4.48-4.35 (m, 4 H, 2 x Bn-C*H*$_2$), 4.31-4.23 (m, 4 H, 2''-*H*$_a$, Bn-CH$_a$*H*$_b$, Bn-C*H*$_2$), 4.07 (dd, *J* = 8.7, 6.6 Hz, 1 H, 2-*H*), 3.75 (dd, *J* = 17.2, 4.0 Hz, 1 H, 2''-*H*$_b$), 2.98 (dd, *J* = 11.8, 3.4 Hz, 1 H, 3'-*H*$_a$), 2.82 (d, *J* = 6.3 Hz, 1 H, 2'''-*H*), 2.72 (dd, *J* = 11.8, 8.3 Hz, 1 H, 3'-*H*$_b$), 2.02 (dqq, *J* = 6.7, 6.7, 6.6 Hz, 1 H, 3-*H*), 1.83 (dqq, *J* = 6.7, 6.7, 6.3 Hz, 1 H,

3'''-*H*), 1.67-1.44 (m, 3 H, 3''''-*H*, 4''''-*H*), 1.25 (d, *J* = 6.7 Hz, 3 H, 3'''''-*H*), 0.88-0.76 (m, 18 H, 4-*H*, 4'''-*H*, 5''''-*H*). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (126 MHz, $CDCl_3$): δ = 173.9, 173.3, 173.0, 172.3 170.5, 168.9 (*C*-1, *C*-1', *C*-1'', *C*-1''', *C*-1'''', *C*-1'''''), 156.5 (N*C*(=O)O), 136.5, 136.4, 136.3, 135.7, 135.4 (5 x C_{Ar}), 129.2, 129.1, 128.9, 128.8, 128.6, 128.2, 128.2, 128.1, 128.0, 127.8, 127.7, 127.0, 126.6 (25 x HC_{Ar}), 70.2 (*C*-2'''), 67.0 (Bn-*C*H$_2$), 60.7 (*C*-2), 53.5 (*C*-2'), 51.5 (*C*-2''''), 50.4 (*C*-3'), 50.0, 49.1, 48.5, 48.2 (4 x Bn-*C*H$_2$), 45.6 (*C*-2'''''), 41.6 (*C*-3''''), 41.4 (*C*-2''), 31.7 (*C*-3'''), 31.2 (*C*-3), 25.0 (*C*-4''''), 23.2, 21.5, 19.6, 19.4, 19.2, 19.0, 18.3 (*C*-3''''', 2 x *C*-4, 2 x *C*-4''', 2 x *C*-5'''').

IR (ATR): ν = 3271, 2958, 1636, 1537, 1452, 1225, 1028, 732, 697.

MS (ESI$^+$): m/z = 1037.6 $[M+H]^+$, calculated: 1037.5859 $[M+H]^+$,
$C_{60}H_{76}N_8O_8$ (1037.32 g(mol)$^{-1}$), found: 1037.5860 [+0.1 ppm] (ESI$^+$-HRMS).



Preparation of tripeptide **51**

To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), $MeNH_2$ (2 M in THF, 0.12 mL, 0.24 mmol) was added. After stirring for 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:MeOH, 96:4 → 94:6) to give 21 mg (0.036 mmol, 90%) of the title compound as a colourless solid.

TLC: R_f = 0.14 (DCM:MeOH, 93:7).

Melting point: T_{mp} = 98 °C.

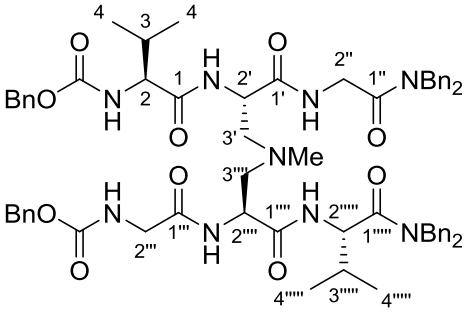
Specific rotation: $[\alpha]_D^{20}$ = +7.3 (c = 1.25, $CHCl_3$)

^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.37 (s, 1 H, 2''-N*H*), 7.30-7.05 (m, 16 H, 2'-N*H*, 15 x *H*C$_{Ar}$), 5.52 (d, *J* = 8.4 Hz, 1 H, 2-N*H*), 5.03 (d, *J* = 12.3 Hz, 1 H, Bn-CH_aH$_b$), 4.99 (d, *J* = 12.3 Hz, 1 H, Bn-CH$_a$$H_b$), 4.57-4.46 (m, 3 H, 2'-*H*, Bn-CH_2), 4.31 (s, 2 H, Bn-CH_2), 4.17-4.03 (m, 3 H, 2-*H*, 2''-*H*), 3.07 (dd, *J* = 11.9, 3.6 Hz, 1 H, 3'-H_a), 2.67 (dd, *J* = 11.9, 7.8 Hz, 1 H, 3'-H_b), 2.41 (s, 3 H, NCH_3), 2.06 (dqq, *J* = 6.7, 6.7, 6.2 Hz, 1 H, 3-*H*), 0.90 (d, *J* = 6.7 Hz, 3 H, 4-H_a), 0.85 (d, *J* = 6.7 Hz, 3 H, 4-H_b). The signal attributed to the secondary amine N*H* proton was not observed in the ^{1}H NMR.

^{13}C NMR (101 MHz, $CDCl_3$): δ = 171.6, 171.0, 168.7 (*C*-1, *C*-1', *C*-1''), 156.7 (N*C*(=O)O), 136.5, 136.4, 135.4 (3 x C_{Ar}), 129.2, 128.8, 128.6, 128.4, 128.2, 128.2, 128.1, 127.8, 126.6 (15 x HC_{Ar}), 67.1 (Bn-*C*H$_2$), 60.5 (*C*-2), 52.7 (*C*-3'), 51.8 (*C*-2'), 49.1, 48.7 (2 x Bn-*C*H$_2$), 41.7 (*C*-2''), 36.1 (N*C*H$_3$), 31.4 (*C*-3), 19.4, 17.8 (2 x *C*-4).

IR (ATR): ν = 3284, 1710, 1629, 1526, 1244, 1096, 1026, 739, 697.

MS (ESI$^+$): m/z = 588.3 $[M+H]^+$, calculated: 588.3181 $[M+H]^+$,
$C_{33}H_{41}N_5O_5$ (587.72 g(mol)$^{-1}$), found: 588.3181 [+0.1 ppm] (ESI$^+$-HRMS).



Preparation of tripeptide tripeptide conjugate **52**

*From isolated and purified tripeptide **51**:* To a solution of tripeptide **5** (19 mg, 0.035 mmol) in $CHCl_3$

(1.5 mL), tripeptide **51** (31 mg, 0.053 mmol) was added. After stirring for 2 d at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt 40:60 → 20:80) to give 38 mg (0.033 mmol, 95%) of the title compound as a colourless solid.

*One-pot protocol without isolation and purification of tripeptide **51**:* To a solution of tripeptide **6** (22 mg, 0.040 mmol) in $CHCl_3$ (1.5 mL), $MeNH_2$ (2 M in THF, 0.12 mL, 0.24 mmol) was added. After stirring for 20 h at 40 °C the solvent and any excess of $MeNH_2$ were evaporated *in vacuo*. The remaining colourless solid was dissolved in $CHCl_3$ (1.5 mL) and tripeptide **5** (15 mg, 0.027 mmol) was added. After stirring for 2 d at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (4 g, 1.0 x 8.0 cm, DCM:AcOEt, 40:60 → 20:80) to give 28 mg (0.024 mmol, 91%) of the title compound as a colourless solid.

TLC: R_f = 0.09 (DCM:AcOEt, 40:60).

Melting point: T_{mp} = 77 °C.

Specific rotation: $[\alpha]_D^{20}$ = -11.4 (c = 2.59, $CHCl_3$)

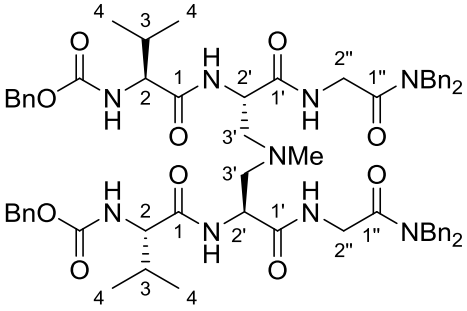
^{1}H NMR (500 MHz, $CDCl_3$): δ = 8.03-7.89 (m, 3 H, 2''-N*H*, 2''''-N*H*, 2''''''-N*H*), 7.40-7.10 (m, 31 H, 2'-N*H*, 30 x *H*C_{Ar}), 5.78 (s, 1 H, 2'''-N*H*), 5.65 (d, *J* = 9.0 Hz, 1 H, 2-N*H*), 5.15-5.05 (m, 4 H, 2 x Bn-C*H*$_2$), 4.97 (dd, *J* = 8.7, 7.0 Hz, 1 H, 2''''-*H*), 4.83 (d, *J* = 14.8 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.70-4.51 (m, 6 H, 2'-*H*, 2''''-*H*, 2 x Bn-C*H*$_2$), 4.40-4.33 (m, 3 H, Bn-CH$_a$*H*$_b$, Bn-C*H*$_2$), 4.28-4.25 (m, 2 H, 2-*H*, 2''-*H*$_a$), 4.01 (dd, *J* = 17.5, 2.3 Hz, 1 H, 2''-*H*$_b$), 3.95-3.87 (m, 2 H, 2'''-*H*), 2.82-2.75 (m, 3 H, 3'-*H*$_a$, 3''''-*H*), 2.60 (dd, *J* = 12.3, 8.1 Hz, 1 H, 3'-*H*$_b$), 2.32 (s, 3 H, NC*H*$_3$), 2.20-2.04 (m, 2 H, 3-*H*, 3''''''-*H*), 1.04-0.88 (m, 12 H, 4-*H*, 4''''''-*H*).

^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.7, 172.5, 170.7, 169.3, 168.5 (*C*-1, *C*-1', *C*-1'', *C*-1''', *C*-1'''', *C*-1''''''), 156.6, 156.5 (2 x N*C*(=O)O), 136.8, 136.5, 136.5, 136.5, 136.0, 135.3 (6 x *C*$_{Ar}$), 129.2, 129.1, 128.8, 128.8, 128.6, 128.4, 128.3, 128.2, 128.2, 128.1, 128.1, 128.1, 127.8, 127.7, 127.3, 126.6 (30 x H*C*$_{Ar}$), 67.1, 67.0 (2 x Bn-*C*H$_2$), 60.6 (*C*-3'), 60.3 (*C*-2), 59.1 (*C*-3''''), 54.5 (*C*-2''''), 51.1, 51.0 (*C*-2', *C*-2''''), 50.3, 49.1, 48.6, 48.3 (4 x Bn-*C*H$_2$), 44.4 (*C*-2'''), 41.6 (*C*-2''), 41.1 (N*C*H$_3$), 31.8, 31.5 (*C*-3, *C*-3''''''), 19.7, 19.5, 18.1, 17.9 (2 x *C*-4, 2 x *C*-4''''''').

IR (ATR): ν = 3296, 2961, 1635, 1496, 1452, 1217, 1028, 734, 697.

MS (ESI$^+$): m/z = 1144.5 $[M+H]^+$, calculated: 1144.5866 $[M+H]^+$,
$C_{65}H_{77}N_9O_{10}$ (1144.38 g(mol)$^{-1}$), found: 1144.5858 [+2.5 ppm] (ESI$^+$-HRMS).

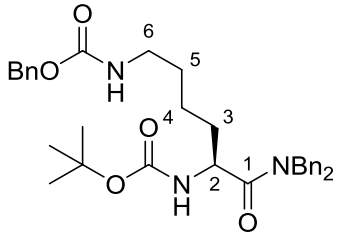
Preparation of tripeptide tripeptide conjugate **53**



To a solution of tripeptide **6** (42 mg, 0.075 mmol) in $CHCl_3$ (1.5 mL), $MeNH_2$ (2 M in THF, 0.13 mL, 0.25 mmol) was added. After stirring for 24 h at 40 °C and 2 d at 65 °C the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (3 g, 1.0 x 8.0 cm, DCM:AcOEt, 70:30 → 40:60) to give 26 mg (0.023 mmol, 92%) of the title compound as a colourless solid.

TLC: R_f = 0.14 (DCM:AcOEt, 50:50).
Melting point: T_{mp} = 86 °C.
Specific rotation: $[\alpha]_D^{20}$ = +6.0 (c = 1.85, $CHCl_3$)
^{1}H NMR (500 MHz, $CDCl_3$): δ = 7.79 (s, 2 H, 2 x 2"-N*H*), 7.61 (d, *J* = 6.0 Hz, 2 H, 2 x 2'-N*H*), 7.28-7.02 (m, 30 H, 30 x *H*C$_{Ar}$), 5.59 (d, *J* = 8.9 Hz, 2 H, 2 x 2-N*H*), 5.01 (d, *J* = 12.3 Hz, 2 H, 2 x Bn-C*H$_a$*H$_b$), 4.95 (d, *J* = 12.3 Hz, 2 H, 2 x Bn-CH$_a$*H$_b$*), 4.62 (d, *J* = 14.8 Hz, 2 H, 2 x Bn-C*H$_a$*H$_b$), 4.55 (ddd, *J* = 7.4, 6.8, 6.0 Hz, 2 H, 2 x 2'-*H*), 4.38 (d, *J* = 14.8 Hz, 2 H, 2 x Bn-CH$_a$*H$_b$*), 4.35-4.24 (m, 6 H, 2 x 2"-*H$_a$*, 2 x Bn-C*H$_2$*), 4.04-3.98 (m, 4 H, 2 x 2-*H*, 2 x 2"-*H$_b$*), 2.75 (dd, *J* = 12.6, 7.4 Hz, 2 H, 2 x 3'-*H$_a$*), 2.64 (dd, *J* = 12.6, 6.8 Hz, 2 H, 2 x 3'-*H$_b$*), 2.15 (s, 3 H, NC*H$_3$*), 2.03 (dqq, *J* = 6.8, 6.7, 6.7 Hz, 2 H, 2 x 3-*H*), 0.90 (d, *J* = 6.7 Hz, 6 H, 2 x 4-*H$_a$*), 0.86 (d, *J* = 6.7 Hz, 6 H, 2 x 4-*H$_b$*).
^{13}C NMR (126 MHz, $CDCl_3$): δ = 172.3, 170.8, 168.9 (2 x *C*-1, 2 x *C*-1', 2 x *C*-1"), 156.6 (2 x N*C*(=O)O), 136.5, 136.5, 135.3 (6 x *C$_{Ar}$*), 129.2, 128.8, 128.6, 128.4, 128.2, 128.1, 128.1, 127.8, 126.7 (30 x H*C$_{Ar}$*), 67.1 (2 x Bn-*C*H$_2$), 60.5 (2 x *C*-2), 59.5 (2 x *C*-3'), 51.1 (2 x *C*-2'), 49.1, 48.5 (4 x Bn-*C*H$_2$), 41.6 (2 x *C*-2"), 41.0 (N*C*H$_3$), 31.2 (2 x *C*-3), 19.5, 18.3 (4 x *C*-4).
IR (ATR): ν = 3288, 2961, 1636, 1528, 1452, 1218, 1026, 734, 697.
MS (ESI$^+$): m/z = 1166.5 $[M+Na]^+$, calculated: 1144.5866 $[M+H]^+$,
$C_{65}H_{77}N_9O_{10}$ (1144.38 g(mol)$^{-1}$), found: 1144.5863 [-0.3 ppm] (ESI$^+$-HRMS).

Preparation of lysine derivative **S1**

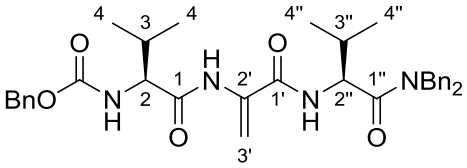


To a solution of N^{α}-(*tert*-Butoxycarbonyl)-N^{ε}-carbobenzoxy-L-lysine (500 mg, 1.31 mmol) in abs. DMF (12 mL), HOBt (177 mg, 1.31 mmol) was added. EDAC (251 mg, 1.31 mmol) was added after cooling the solution to 0 °C. After stirring for 5 min at 0 °C NEt_3 (0.36 mL, 2.6 mmol) was added. Dibenzyl amine (0.25 mL, 1.3 mmol) was added after additional 15 min of stirring at 0 °C. After stirring for 15 h and concomitantly slowly warming to rt the reaction mixture was diluted with AcOEt (200 mL) and washed with water (200 mL), 10 wt% aqueous citric acid solution (200 mL) and sat. aqueous $NaHCO_3$ solution (200 mL). The organic layer was dried over Na_2SO_4, filtered and the solvent of the filtrate evaporated *in vacuo*. The resultant crude product was purified by FCC (50 g, 4.5 x 7.0 cm, PE:AcOEt, 85:15 → 60:40) to give 682 mg (1.22 mmol, 94%) of the title compound as a colourless solid.
TLC: R_f = 0.29 (PE:AcOEt, 60:40).
Melting point: T_{mp} = 68 °C.
Specific rotation: $[\alpha]_D^{20}$ = -24.5 (c = 2.28, $CHCl_3$)
^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.28-7.09 (m, 15 H, 15 x *H*C$_{Ar}$), 5.32 (d, *J* = 8.7 Hz, 1 H, 2-N*H*), 5.00 (s, 2 H, Bn-C*H$_2$*), 4.74 (s, 1 H, 6-N*H*), 4.63-4.56 (m, 2 H, 2-*H*, Bn-C*H$_a$*H$_b$), 4.51-4.43 (m, 3 H, Bn-CH$_a$*H$_b$*, Bn-C*H$_2$*), 3.02 (ddd, *J* = 6.2, 6.1, 5.6 Hz, 2 H, 6-*H*), 1.57-1.48 (m, 2 H, 3-*H*), 1.35 (s, 9 H, C(C*H$_3$*)$_3$), 1.31-1.17 (m, 4 H, 4-*H*, 5-*H*).
^{13}C NMR (101 MHz, $CDCl_3$): δ = 173.4 (*C*-1), 156.5, 155.8 (2 x N*C*(=O)O), 136.9, 136.8, 136.3 (3 x *C$_{Ar}$*), 129.1, 128.8, 128.6, 128.3, 128.2, 128.0, 127.7, 127.0 (15 x H*C$_{Ar}$*), 79.9

(*C*(CH_3)$_3$), 66.7 (Bn-*C*H_2), 50.2 (*C*-2), 50.1, 48.6 (2 x Bn-*C*H_2), 40.8 (*C*-6), 33.2 (*C*-3), 29.3 (*C*-5), 28.4 (C(*C*H_3)$_3$), 22.5 (*C*-4).

IR (ATR): ν = 3332, 1696, 1645, 1527, 1429, 1253, 1168, 1026, 696.

MS (ESI^+): m/z = 560.3 $[M+H]^+$, calculated: 582.2938 $[M+Na]^+$,
$C_{33}H_{41}N_3O_5$ (559.71 g(mol)$^{-1}$), found: 582.2933 [-1.0 ppm] (ESI^+-HRMS).



Preparation of tripeptide **S2**

To a solution of tripeptide **4** (180 mg, 0.30 mmol) in abs. $CHCl_3$ (4 mL), TMEDA (90 µL, 0.60 mmol) was added. After stirring for 40 h the solvent was evaporated *in vacuo*. The resultant crude product was purified by FCC (18 g, 2.5 x 7.0 cm, PE:AcOEt, 75:25) to give 162 mg (0.271 mmol, 90%) of the title compound as a colourless solid.

TLC: R_f = 0.42 (PE:AcOEt, 60:40).

Melting point: T_{mp} = 62 °C.

Specific rotation: $[\alpha]_D^{20}$ = -23.2 (c = 2.03, $CHCl_3$)

^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.31 (s, 1 H, 2'-N*H*), 7.29-7.08 (m, 16 H, 2"-N*H*, 15 x *H*C_{Ar}), 6.44 (d, *J* = 1.8 Hz, 1 H, 3'-*H*$_a$), 5.39-5.37 (m, 2 H, 2-N*H*, 3'-*H*$_b$), 5.06 (d, *J* = 12.2 Hz, 1 H, Bn-C*H*$_a$H$_b$), 5.01 (d, *J* = 12.2 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.95 (dd, *J* = 8.6, 5.5 Hz, 1 H, 2"-*H*), 4.90 (d, *J* = 14.6 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.60 (d, *J* = 16.5 Hz, 1 H, Bn-C*H*$_a$H$_b$), 4.30 (d, *J* = 16.5 Hz, 1 H, Bn-CH$_a$*H*$_b$), 4.09-4.06 (m, 2 H, 2-*H*, Bn-CH$_a$*H*$_b$), 2.12-2.03 (m, 2 H, 3-*H* ,3"-*H*), 0.91-0.84 (m, 12 H, 4-*H*, 4"-*H*).

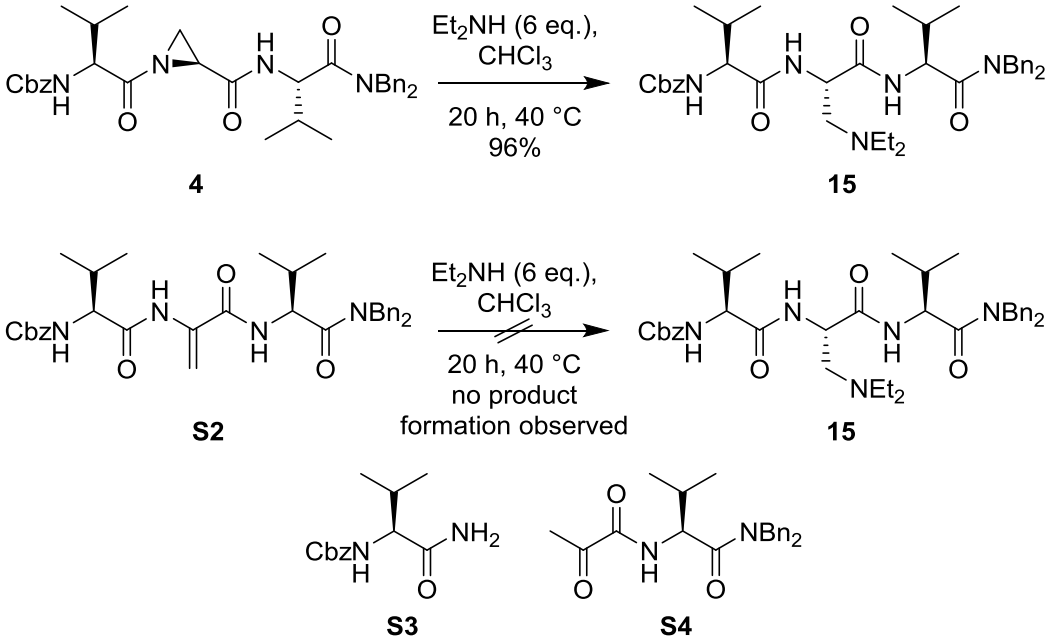
^{13}C NMR (101 MHz, $CDCl_3$): δ = 171.8 (*C*-1"), 170.5 (*C*-1), 163.5 (*C*-1'), 156.5 (N*C*(=O)O), 136.7, 136.4, 135.7 (3 x *C*$_{Ar}$), 133.8 (*C*-2'), 129.1, 128.8, 128.6, 128.4, 128.2, 128.1, 127.7, 127.1 (15 x H*C*$_{Ar}$), 102.8 (*C*-3'), 67.1 (Bn-*C*H_2), 60.9 (*C*-2), 54.5 (*C*-2"), 50.0, 48.0 (2 x Bn-*C*H_2), 32.0, 31.5 (*C*-3, *C*-3"), 19.8, 19.3, 17.7, 17.3 (2 x *C*-4, 2 x *C*-4").

IR (ATR): ν = 3301, 1720, 1627, 1503, 1448, 1215, 1032, 749, 699.

MS (ESI^+): m/z = 621.3 $[M+Na]^+$, calculated: 621.3047 $[M+Na]^+$,
$C_{35}H_{42}N_4O_5$ (598.74 g(mol)$^{-1}$), found: 621.3053 [-0.9 ppm] (ESI^+-HRMS).

3. Mechanistic control experiment

*In agreement with the preparation of tripeptide **15** from aziridine containing tripeptide **4**, didehydroalanine derivative **S2** was subjected to identical reaction conditions*:

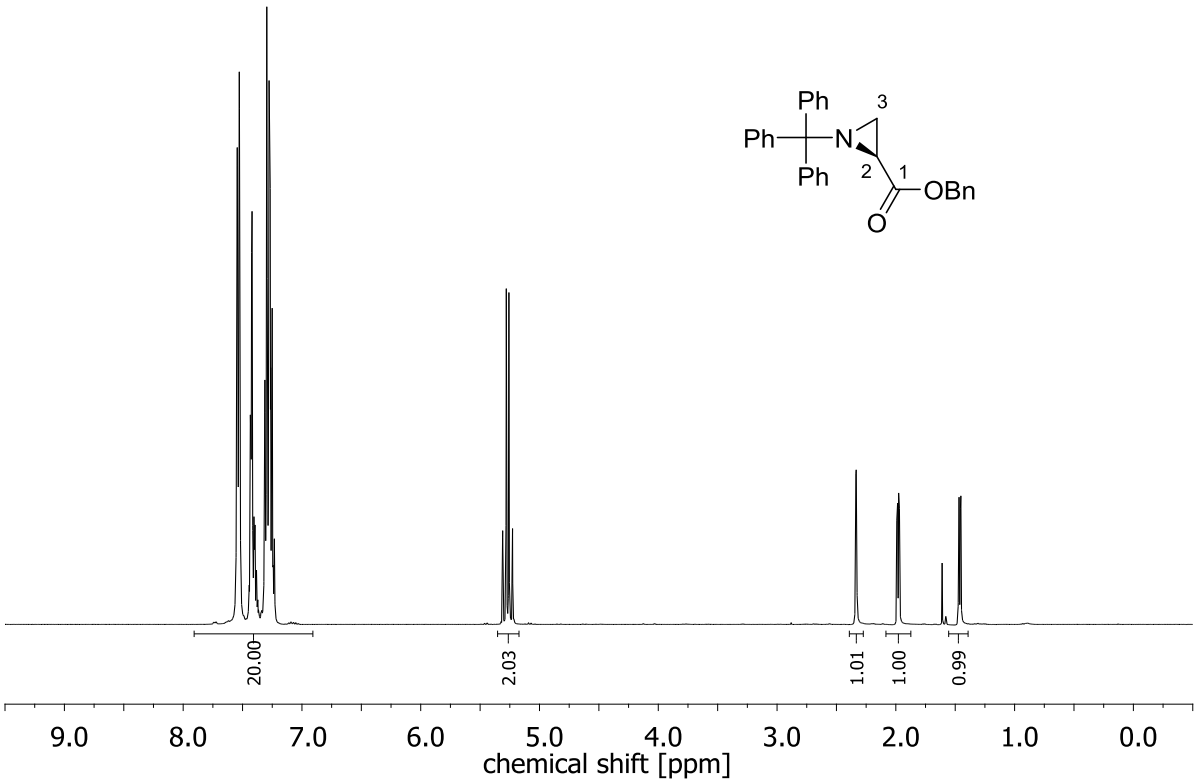


Attempted preparation of tripeptide **15** from didehydroalanine derivative **S2**

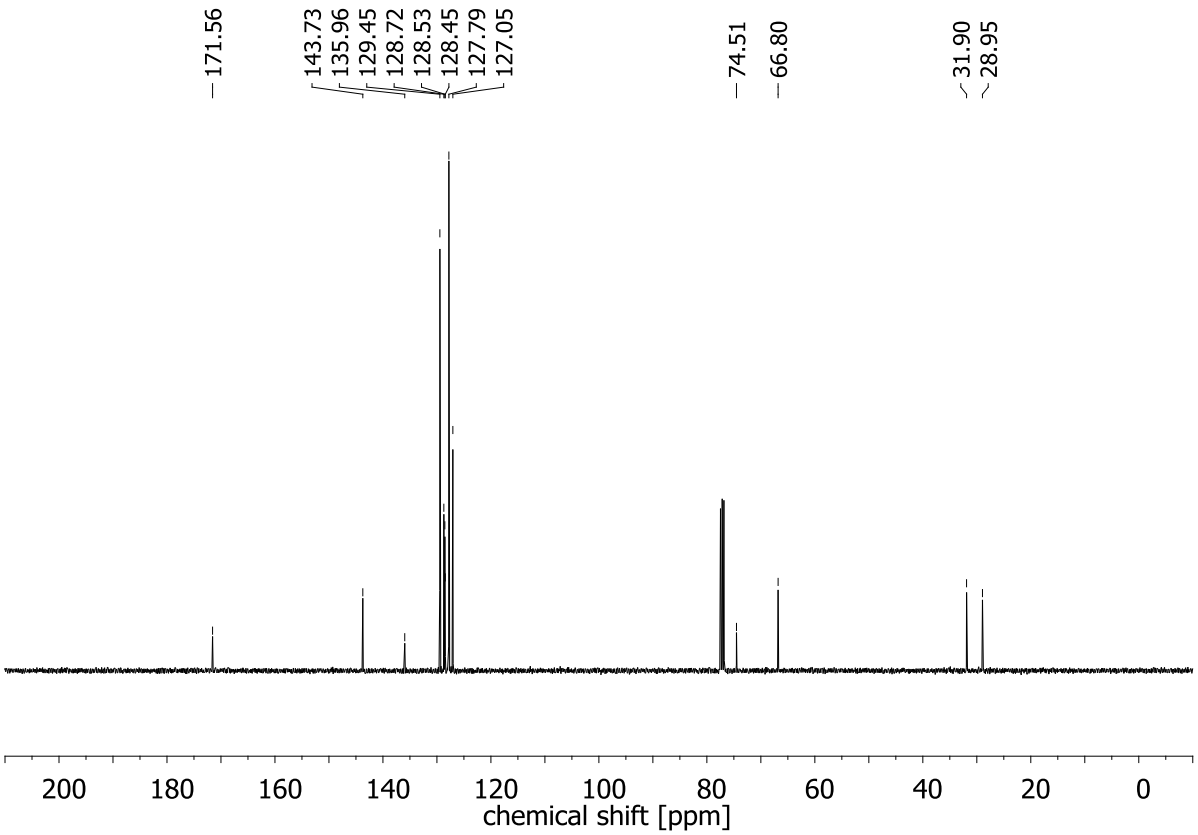
To a solution of didehydroalanine containing tripeptide **S2** (15 mg, 0.025 mmol) in $CHCl_3$ (1.5 mL), diethylamine (16 µL, 0.15 mmol) was added. After stirring for 20 h at 40 °C the solvent was evaporated *in vacuo*. The resultant crude product was filtered through silica (2 g, 1.0 x 4.0 cm, DCM:MeOH, 95:5) and the solvent of the filtrate evaporated *in vacuo* to give 13 mg of a colourless solid.

Based on rigorous TLC, MS and NMR analysis of the isolated material it was unambiguously proven that all starting material **S2** was consumed while no indication for any formation of the expected product **15** was found. Furtheremore, the NMR and MS analysis strongly suggested that the isolated material consisted of a mixture of compounds **S3** and **S4**. Since it was not possible to fully purify and to separate **S3** and **S4** the identity of **S3** and **S4** was not unambiguously proven. Nevertheless, it was shown, that the reaction of **S2** with diethylamine does not yield the ring-opening product **15** or any isomer, under the aziridine ring opening conditions.

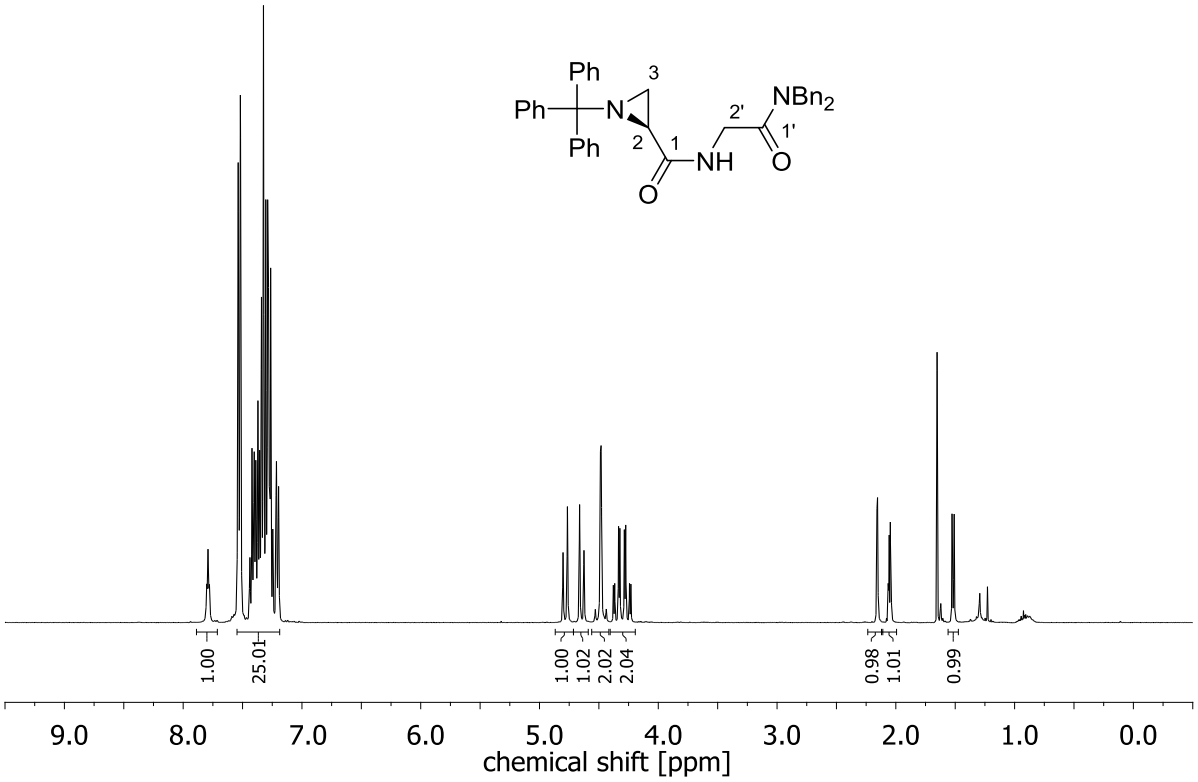
4. NMR spectra of unknown compounds and key building block 9



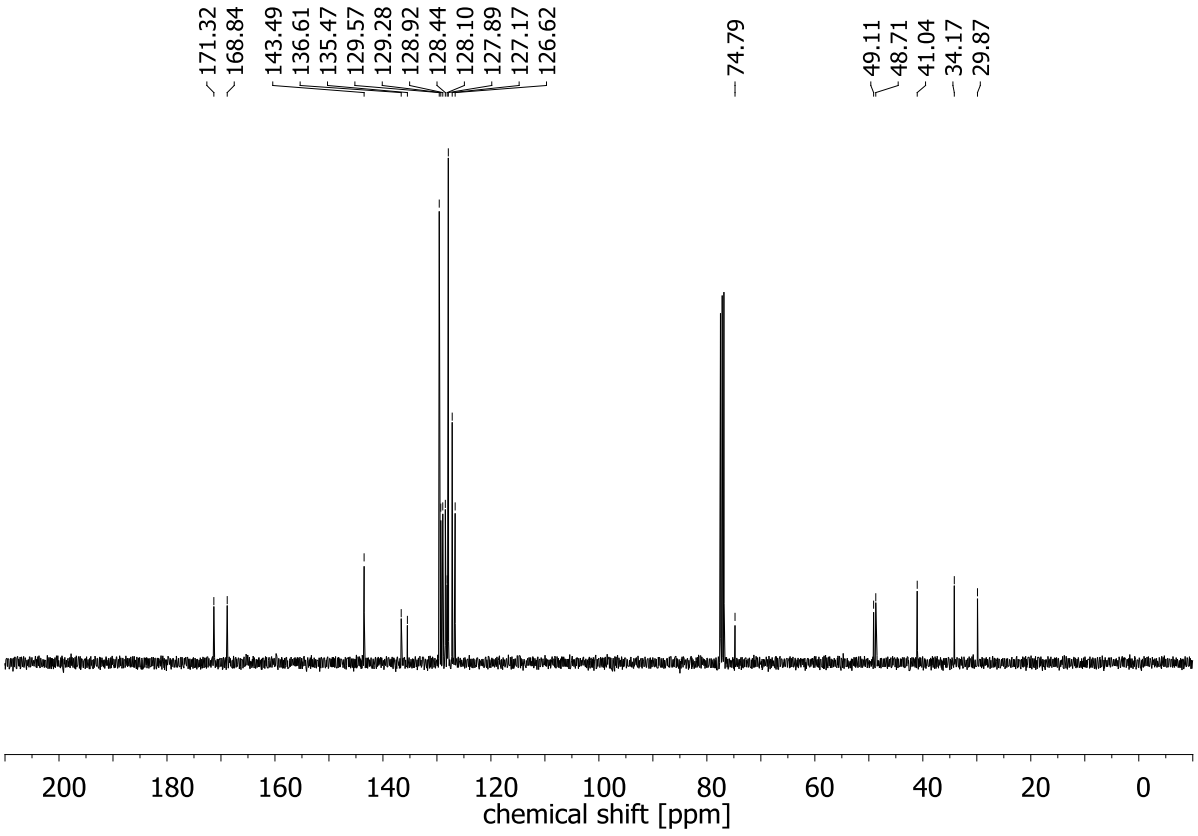
^{1}H NMR spectrum of **9** (400 MHz, $CDCl_3$)



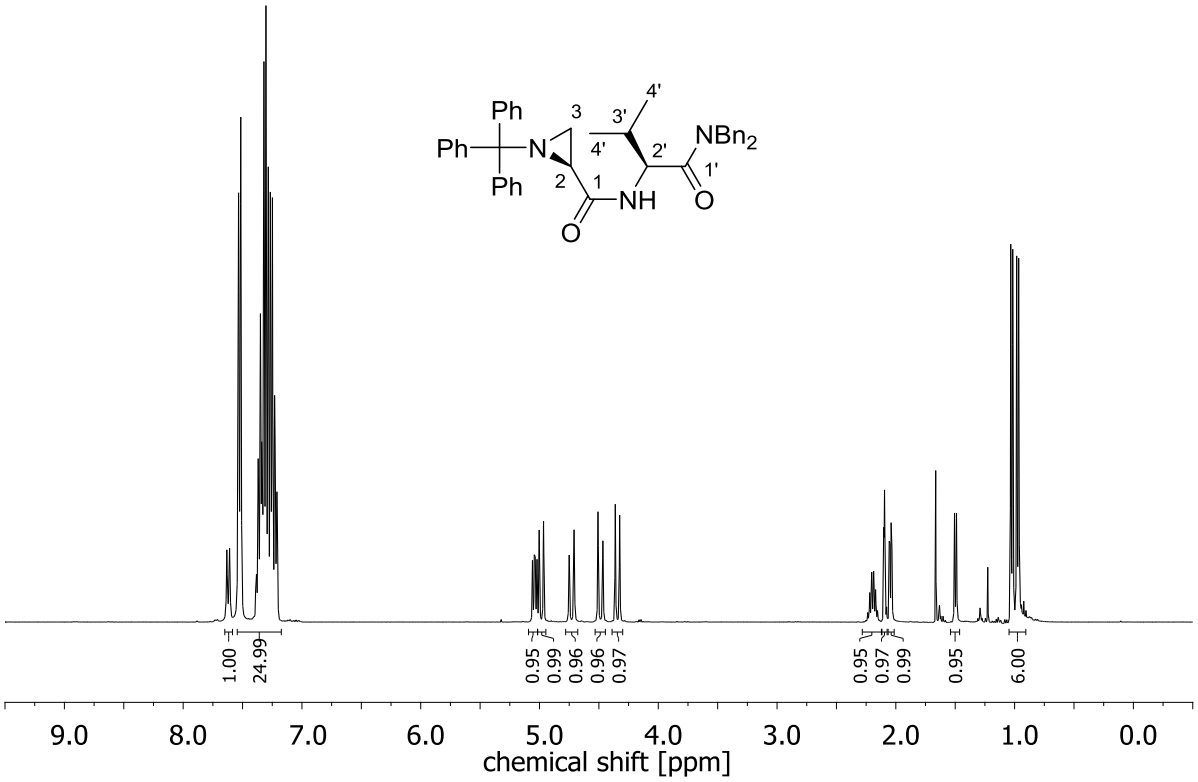
^{13}C NMR spectrum of **9** (101 MHz, $CDCl_3$)



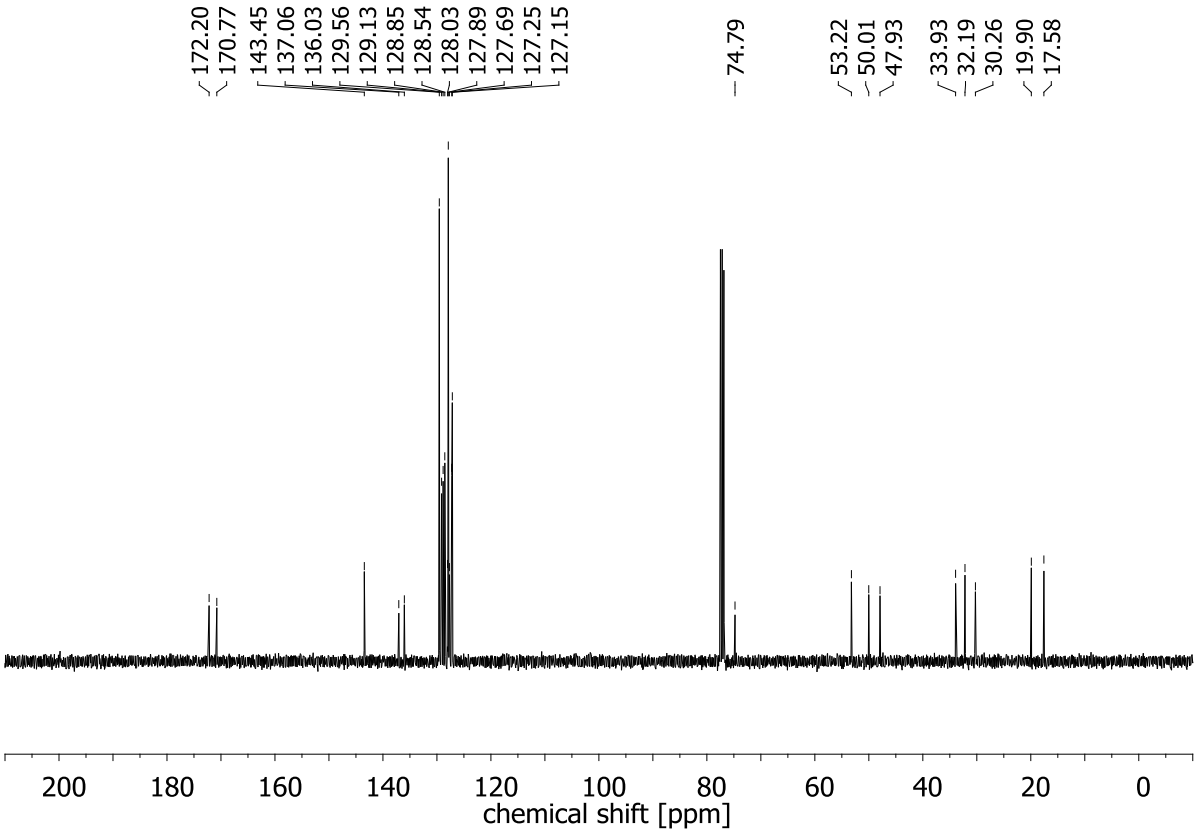
^{1}H NMR spectrum of **11** (400 MHz, $CDCl_3$)



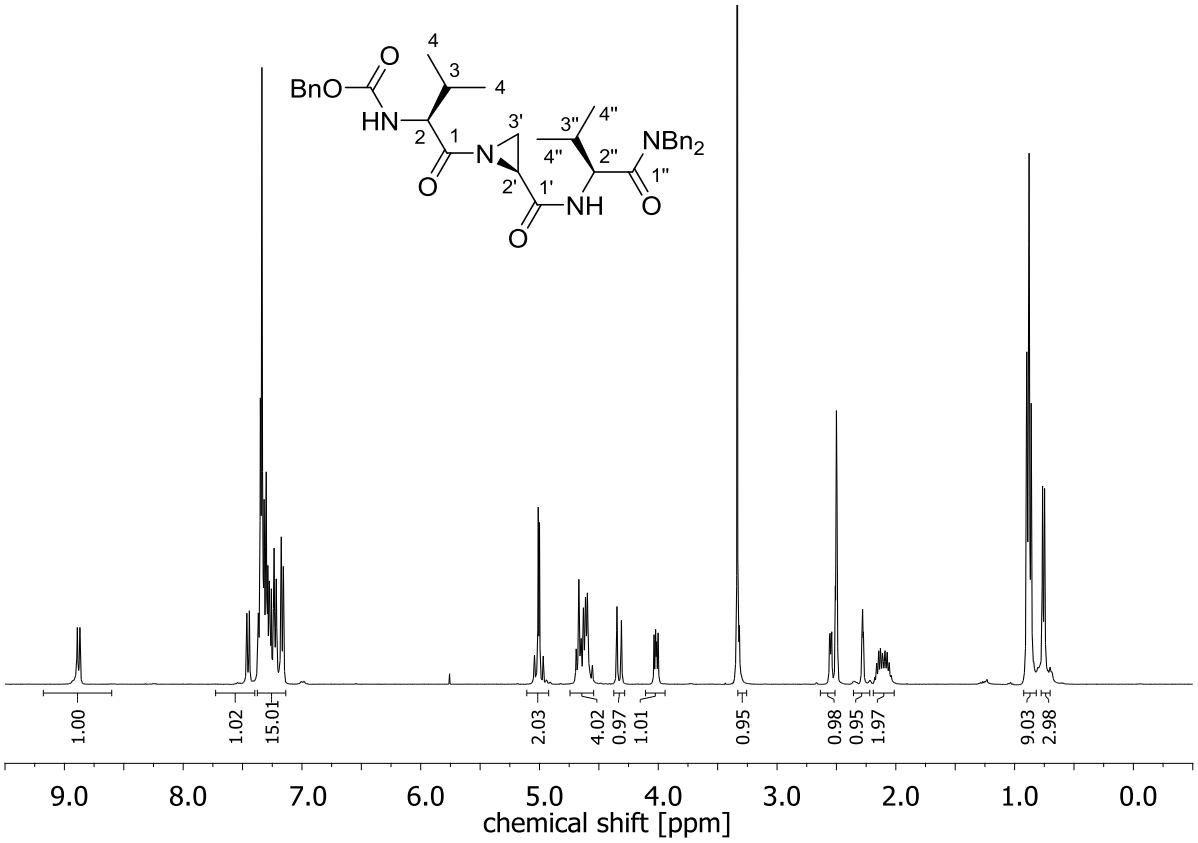
^{13}C NMR spectrum of **11** (101 MHz, $CDCl_3$)



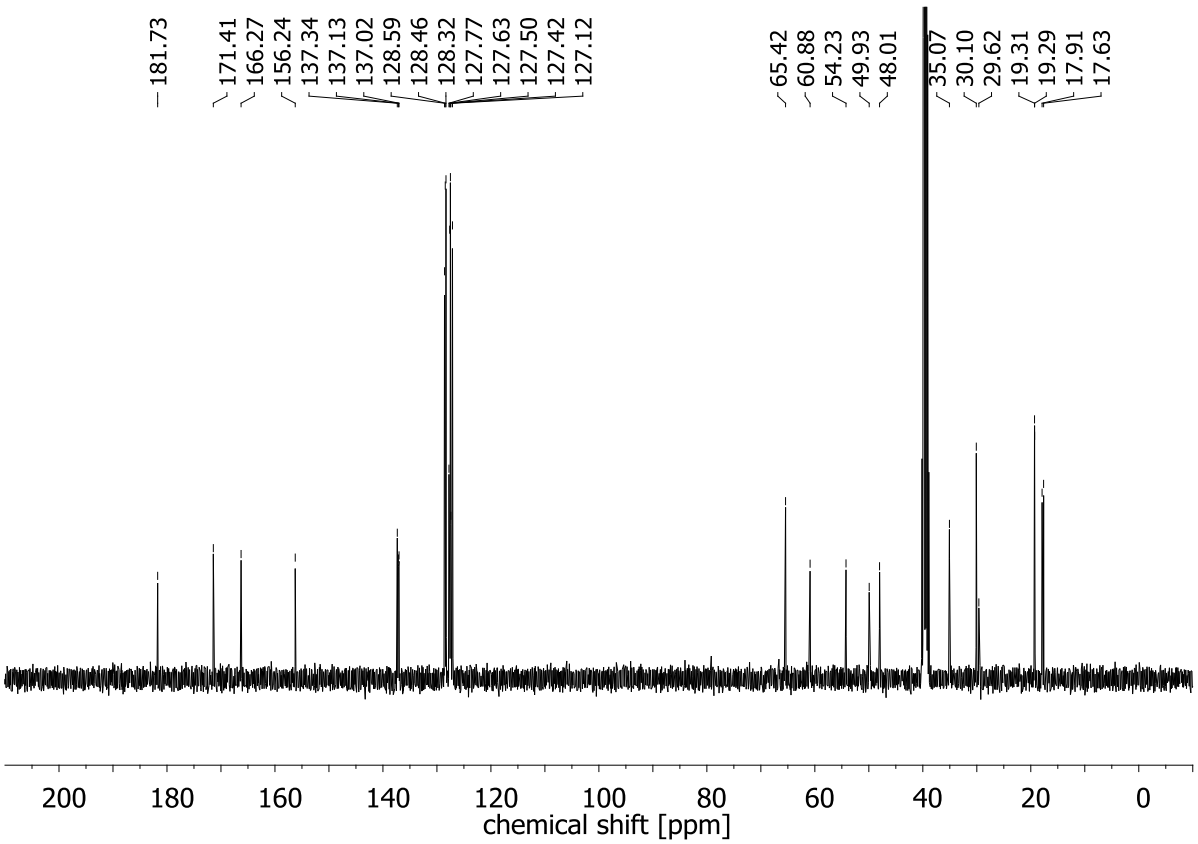
^{1}H NMR spectrum of **12** (400 MHz, $CDCl_3$)



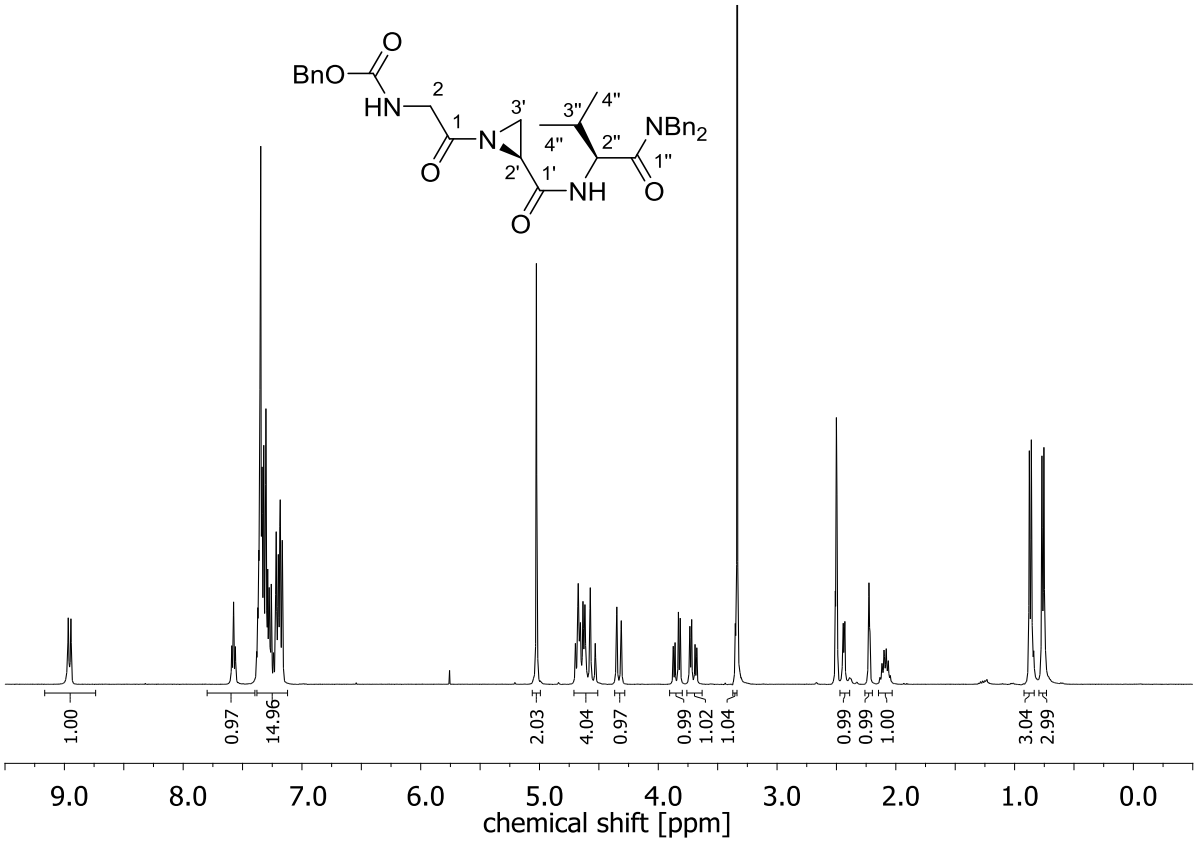
^{13}C NMR spectrum of **12** (101 MHz, $CDCl_3$)



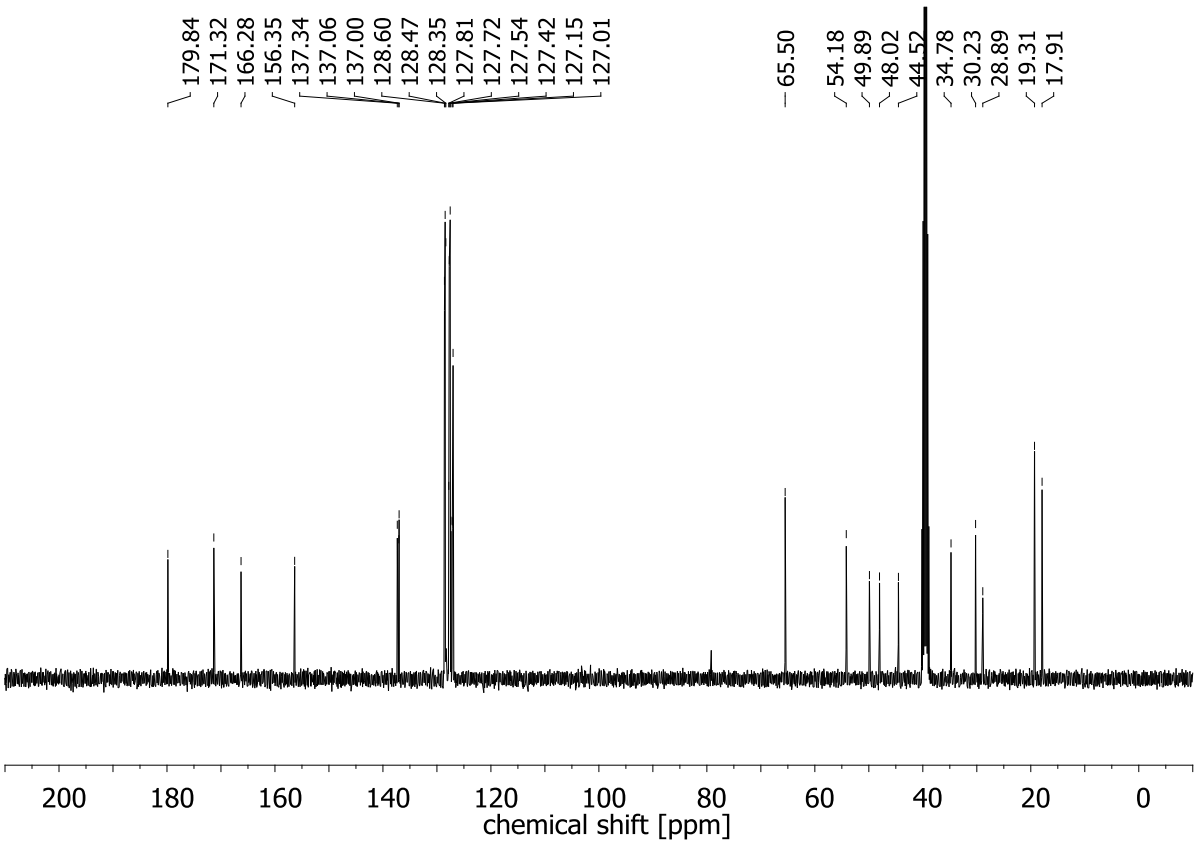
^{1}H NMR spectrum of **4** (400 MHz, D_6-DMSO)



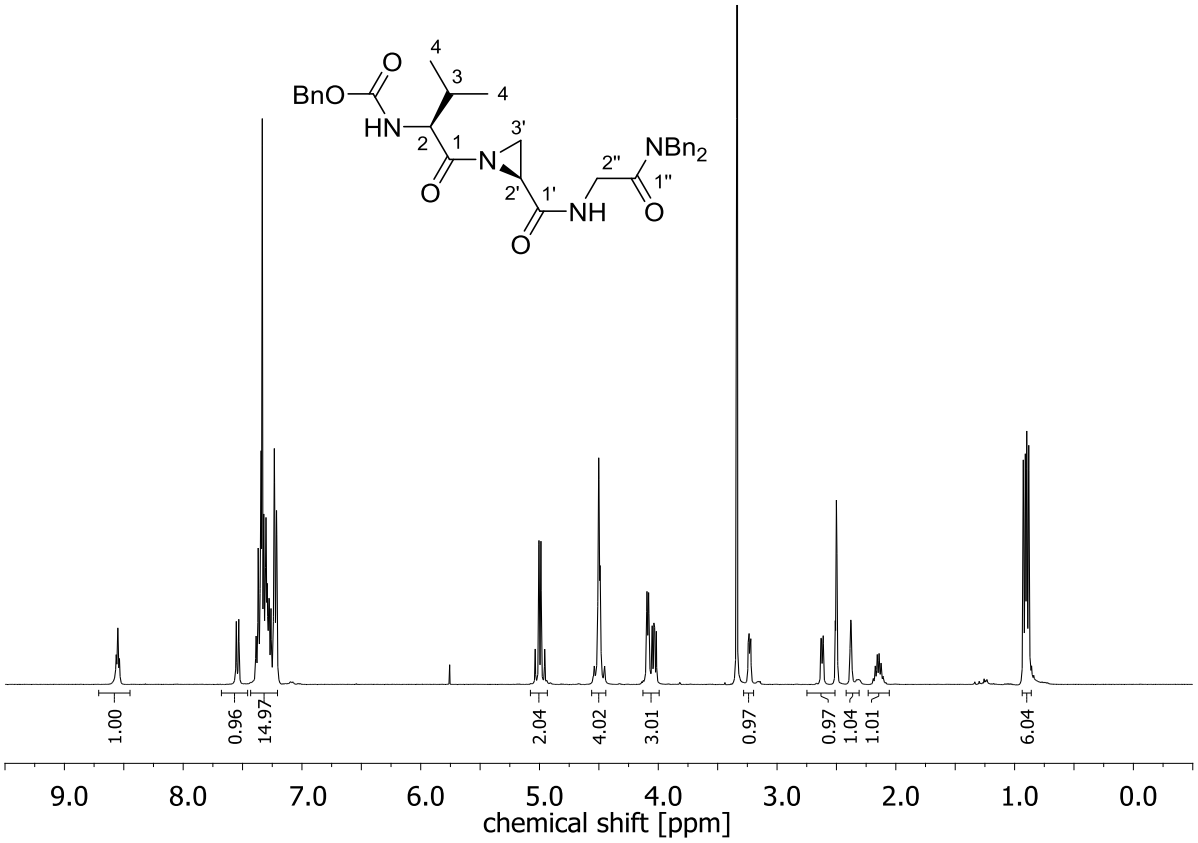
^{13}C NMR spectrum of **4** (101 MHz, D_6-DMSO)



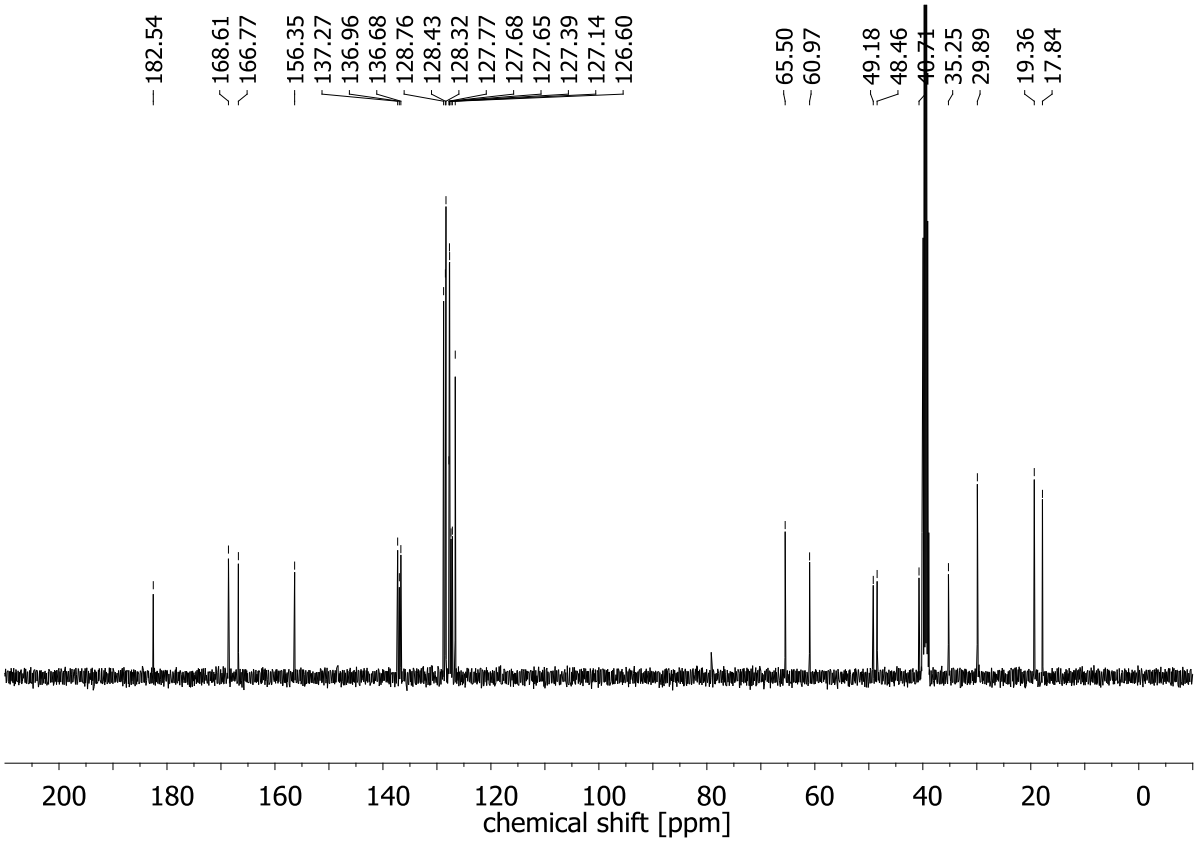
^{1}H NMR spectrum of **5** (400 MHz, D_6-DMSO)



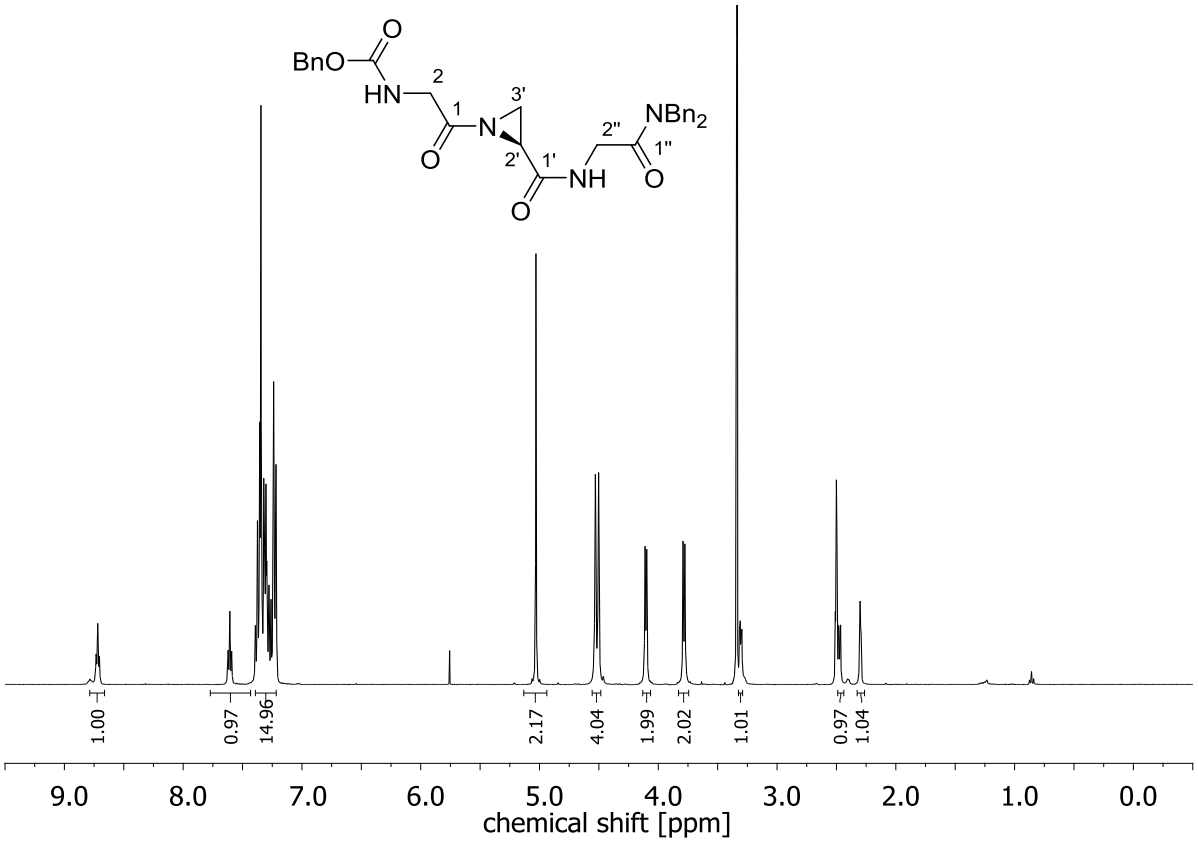
^{13}C NMR spectrum of **5** (101 MHz, D_6-DMSO)



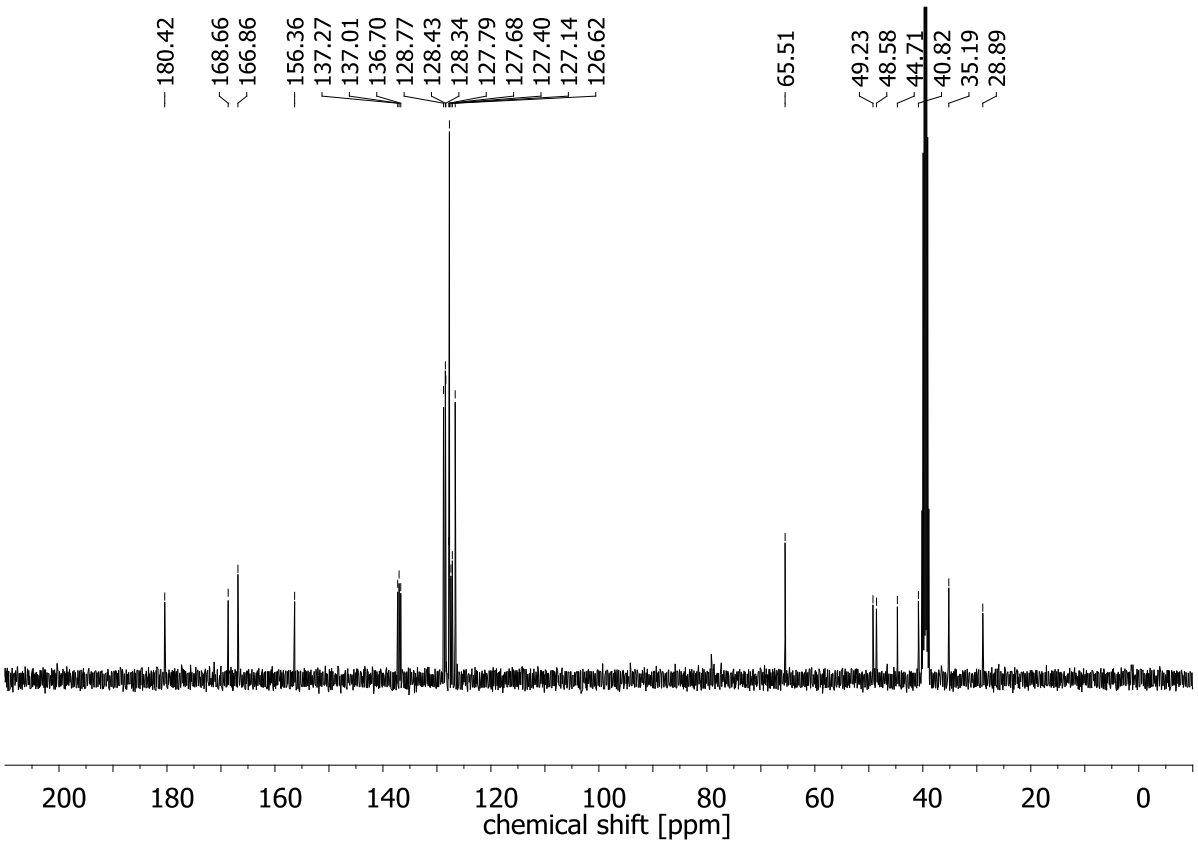
^{1}H NMR spectrum of **6** (400 MHz, D_6-DMSO)



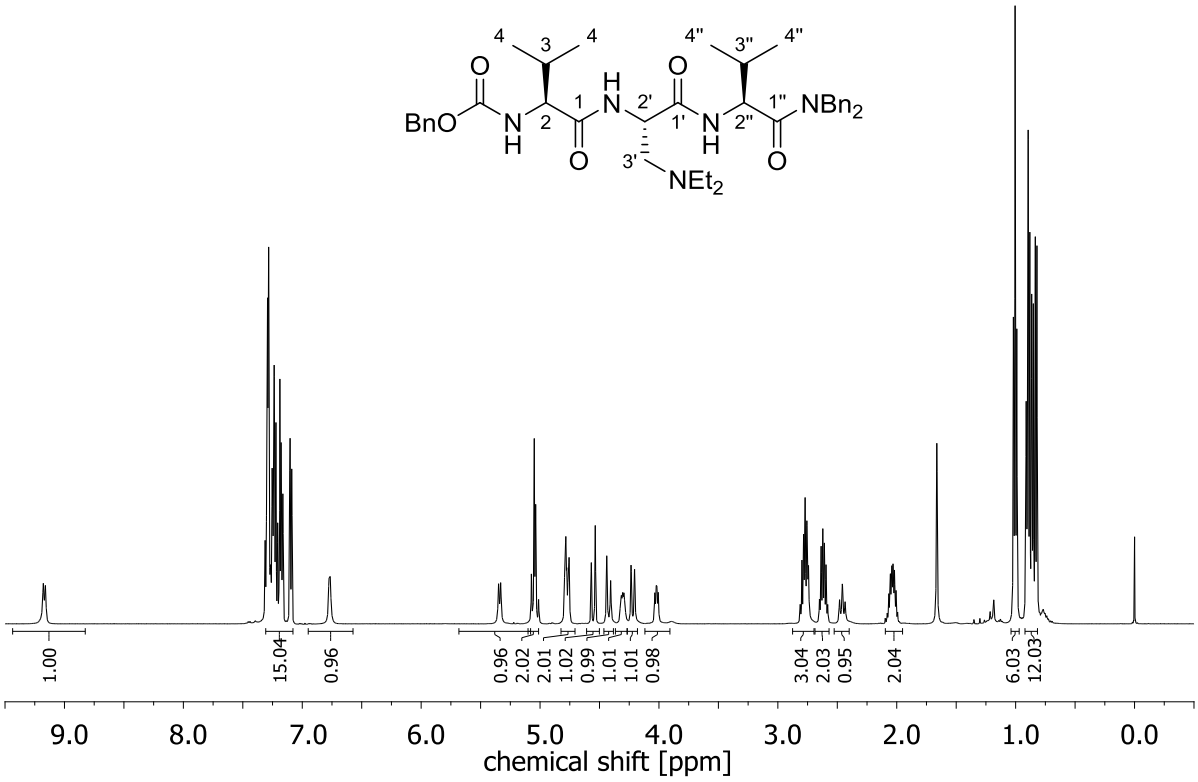
^{13}C NMR spectrum of **6** (101 MHz, D_6-DMSO)



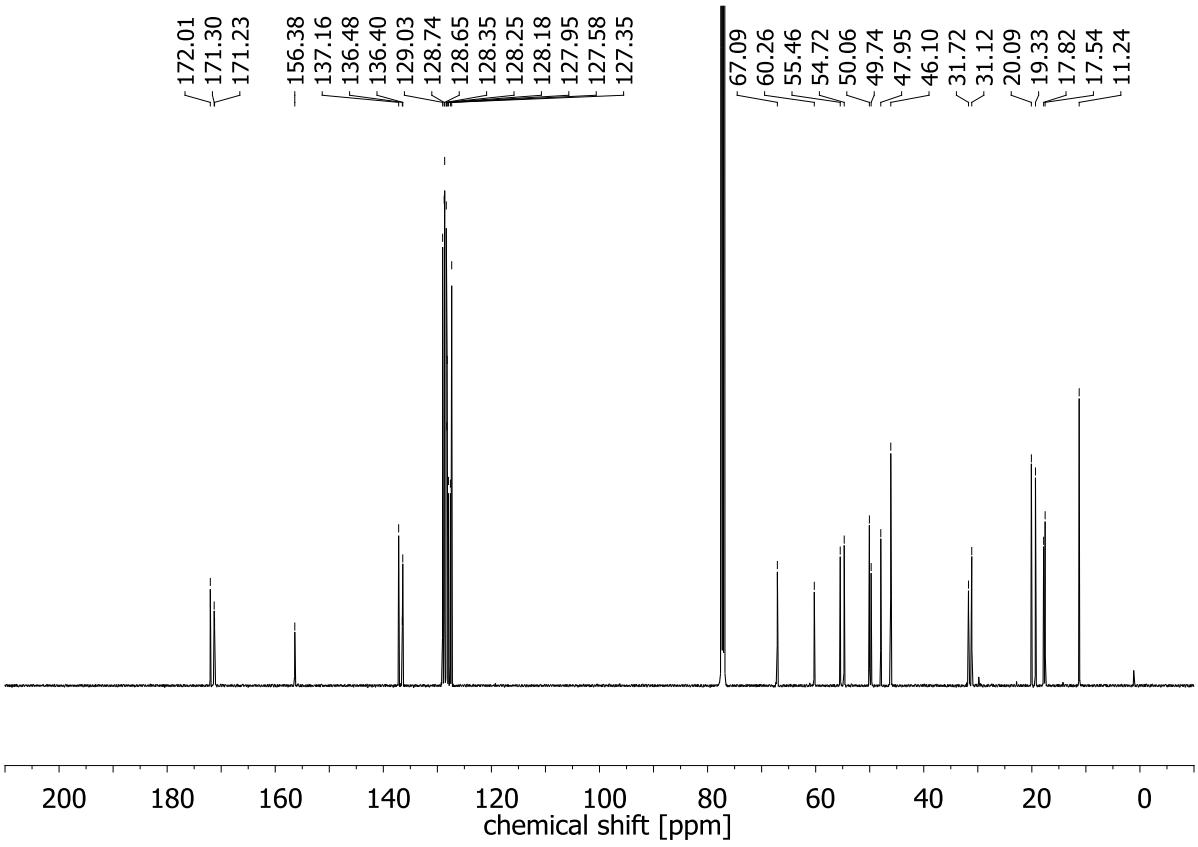
^{1}H NMR spectrum of **7** (400 MHz, D_6-DMSO)



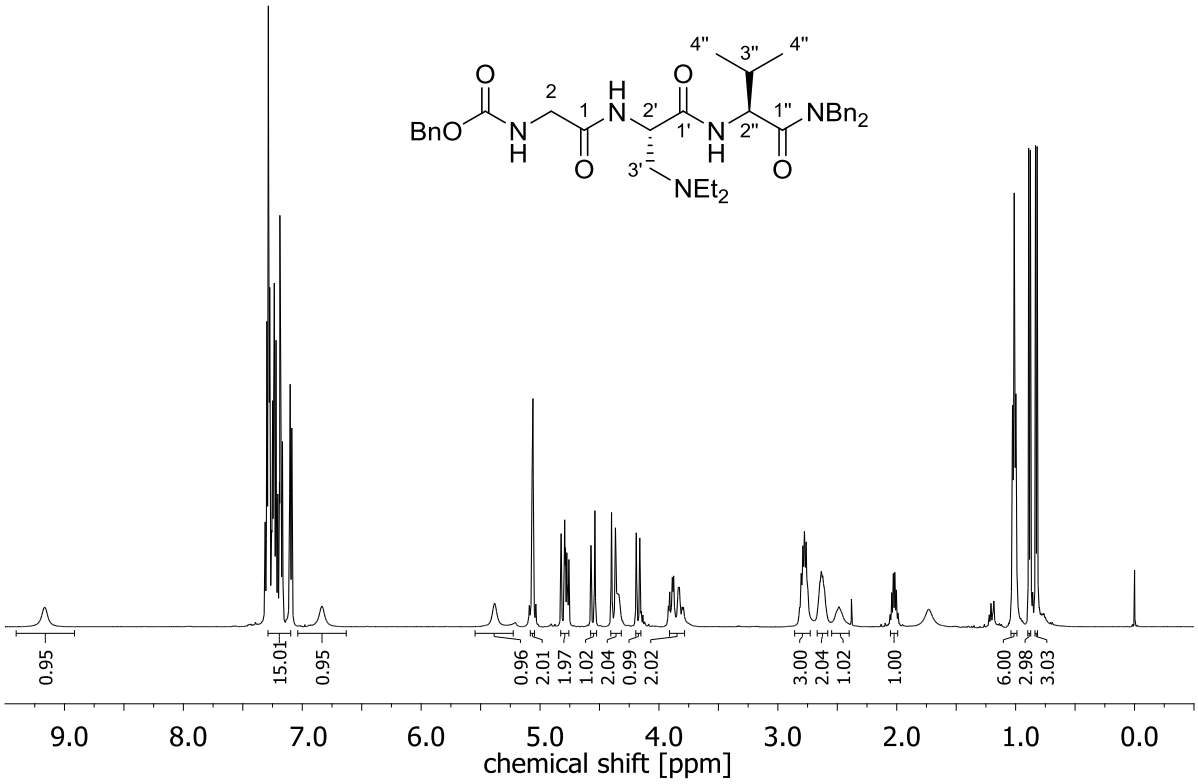
^{13}C NMR spectrum of **7** (101 MHz, D_6-DMSO)



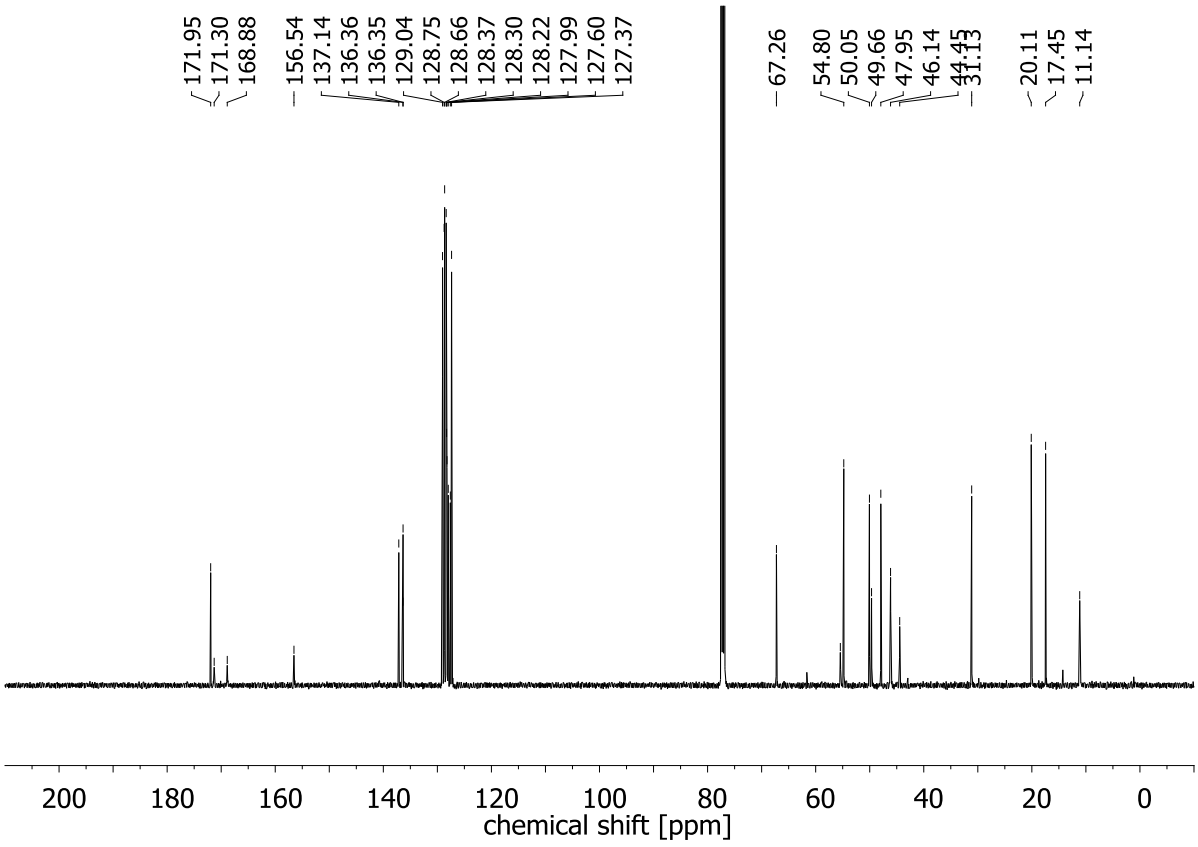
^{1}H NMR spectrum of **15** (500 MHz, $CDCl_3$)



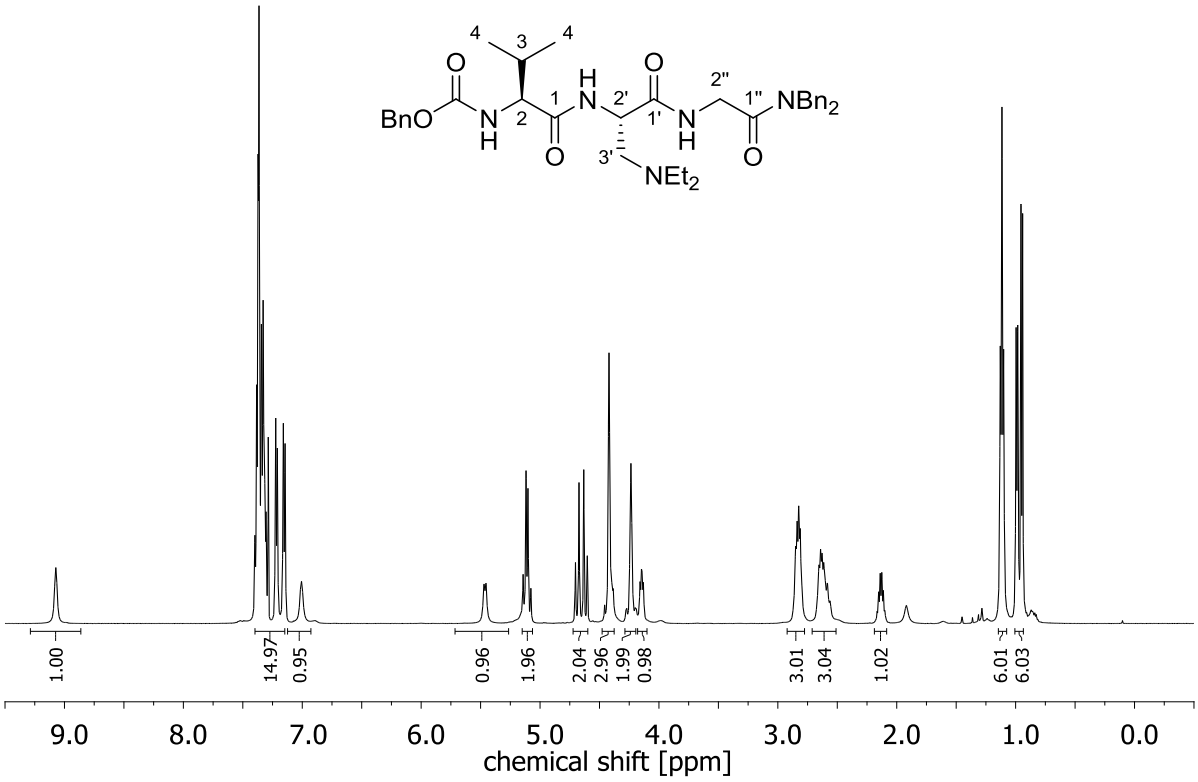
^{13}C NMR spectrum of **15** (126 MHz, $CDCl_3$)



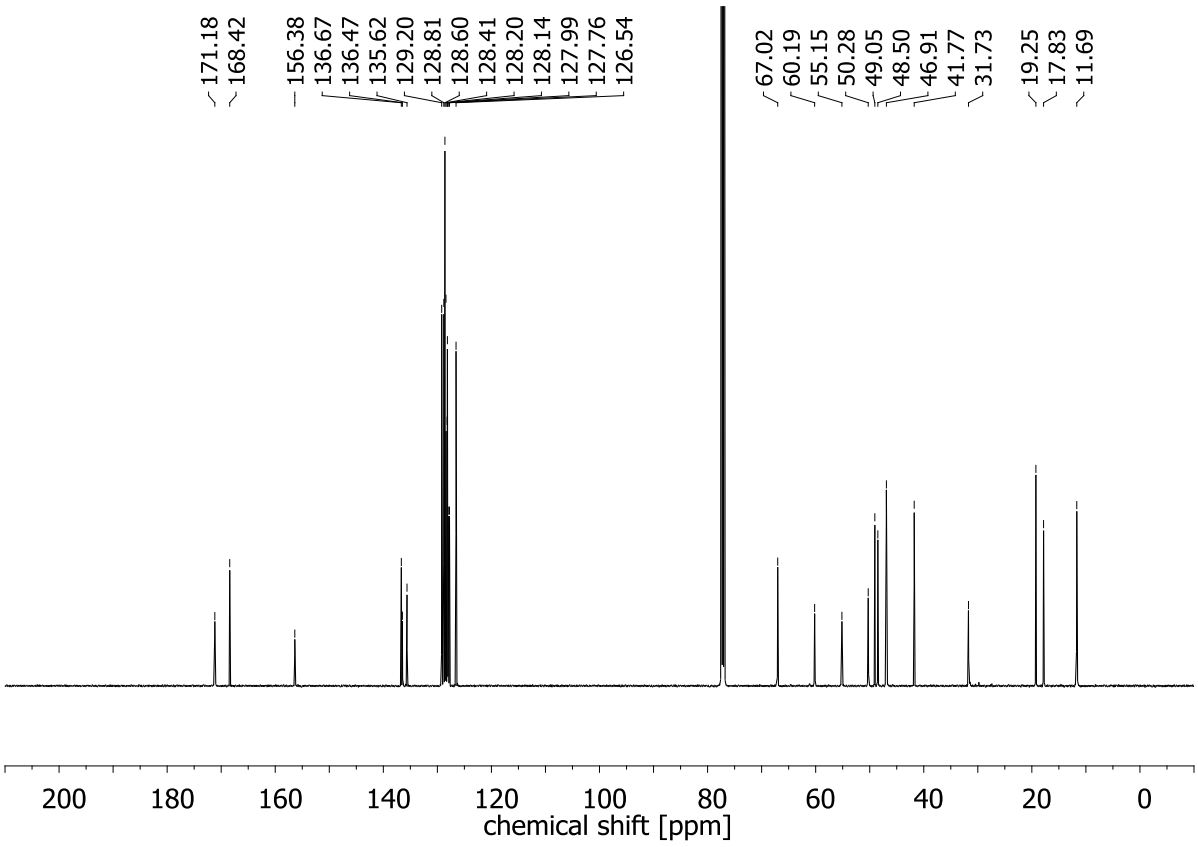
^{1}H NMR spectrum of **16** (500 MHz, $CDCl_3$)



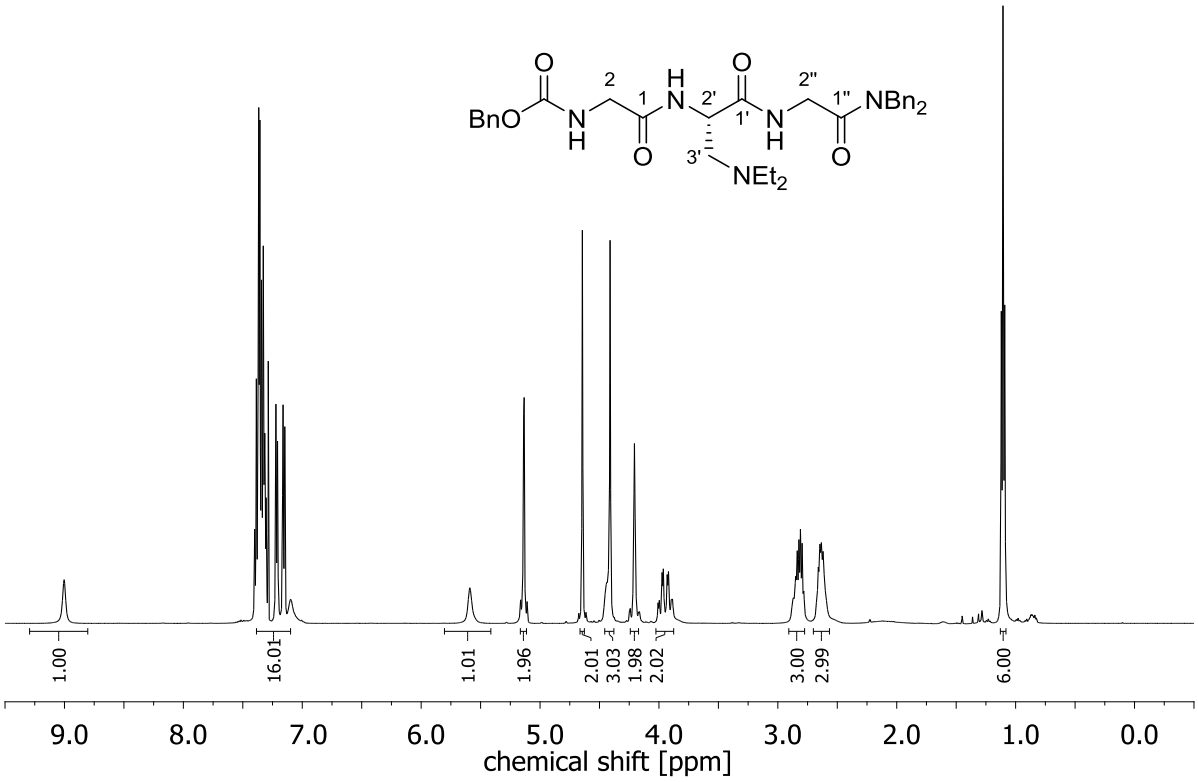
^{13}C NMR spectrum of **16** (126 MHz, $CDCl_3$)



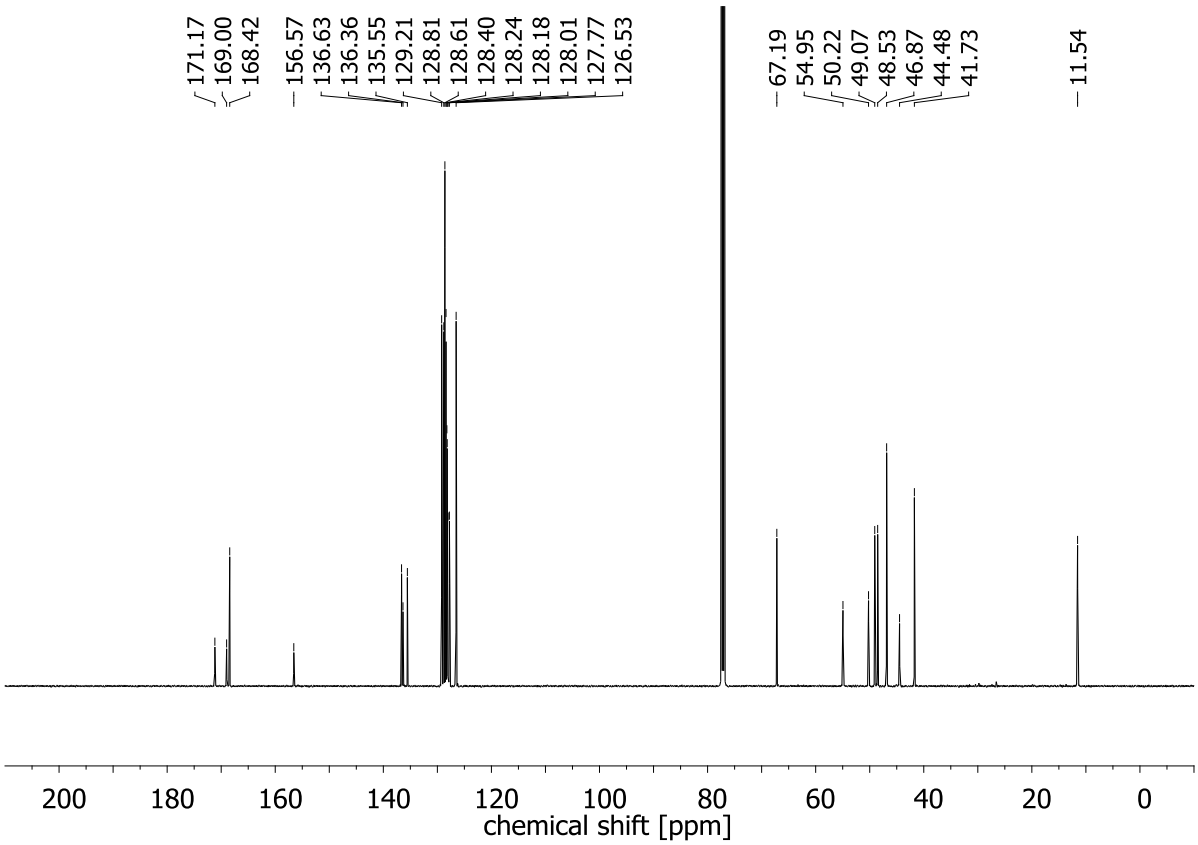
^{1}H NMR spectrum of **17** (500 MHz, $CDCl_3$)



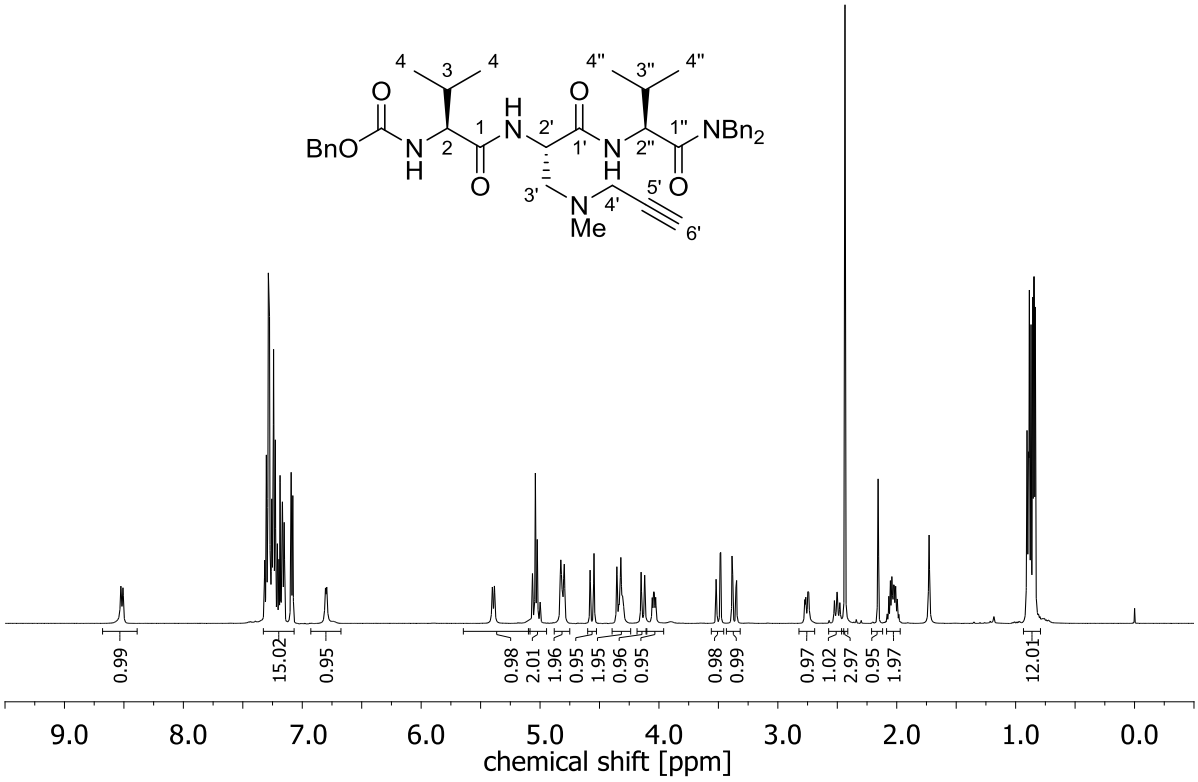
^{13}C NMR spectrum of **17** (126 MHz, $CDCl_3$)



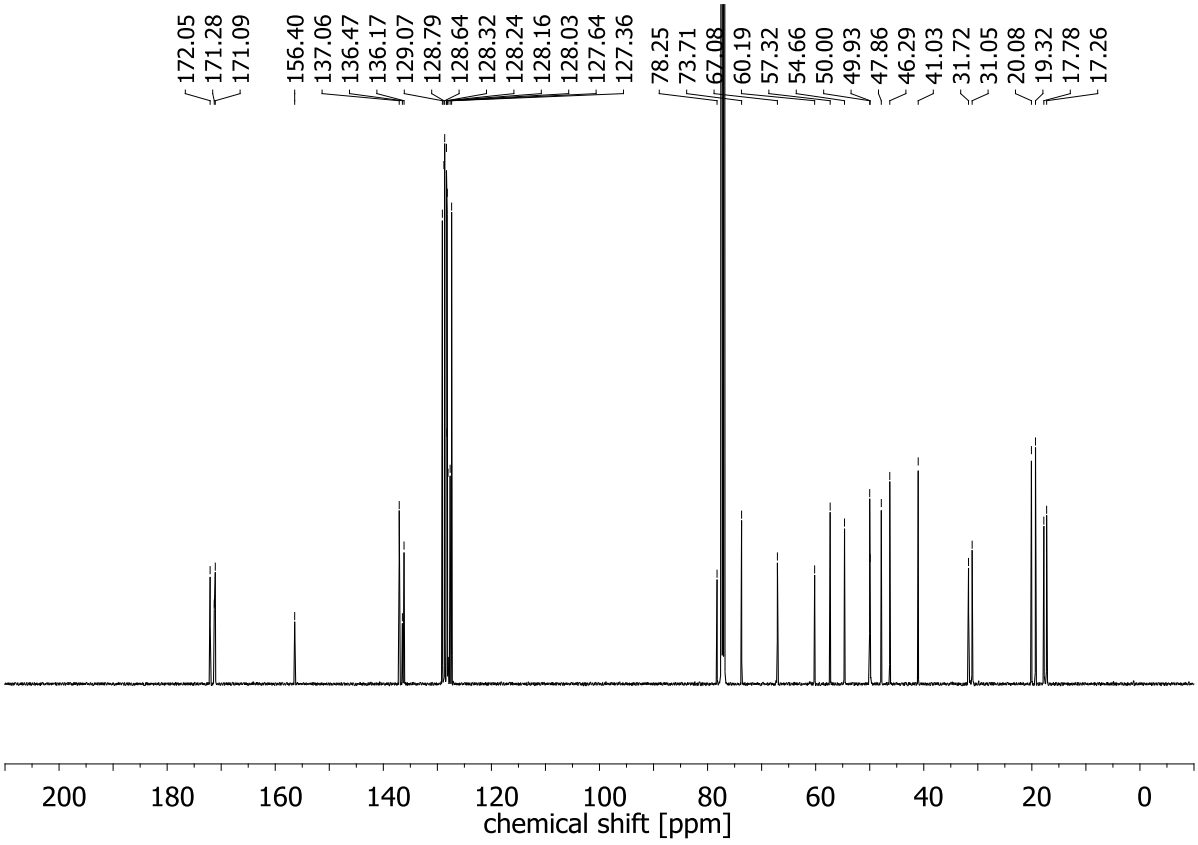
^{1}H NMR spectrum of **18** (500 MHz, $CDCl_3$)



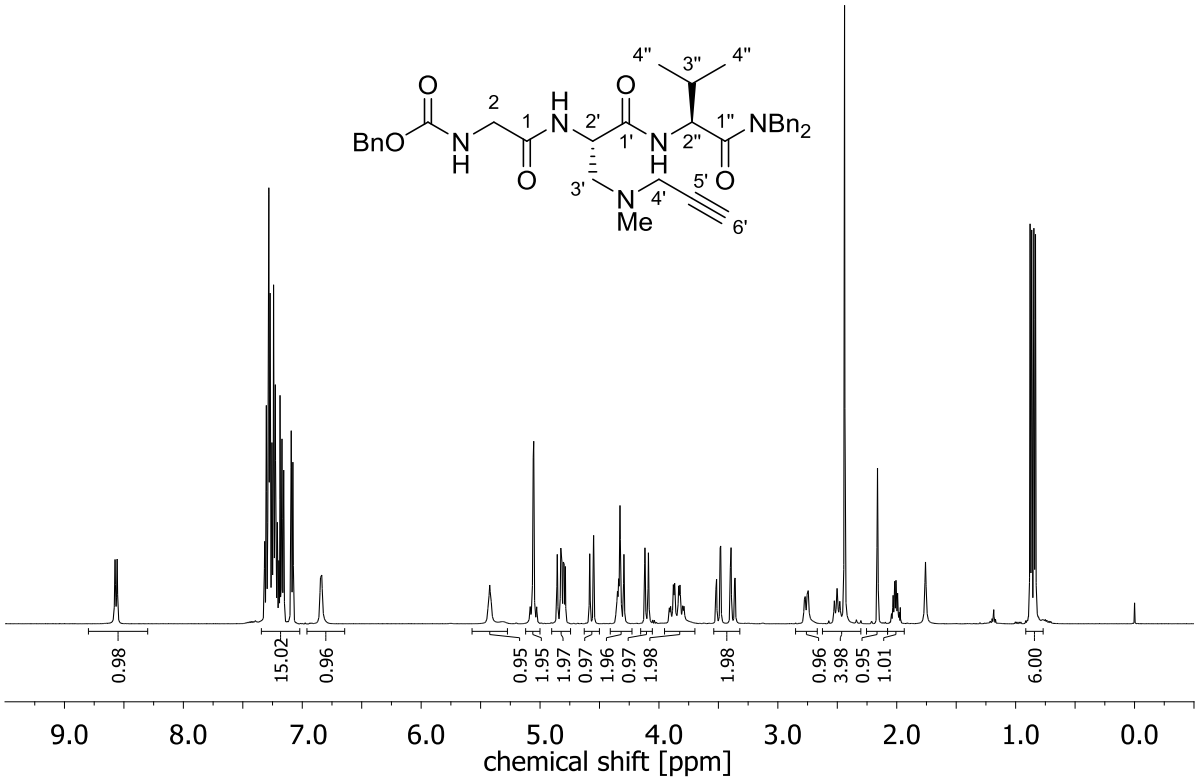
^{13}C NMR spectrum of **18** (126 MHz, $CDCl_3$)



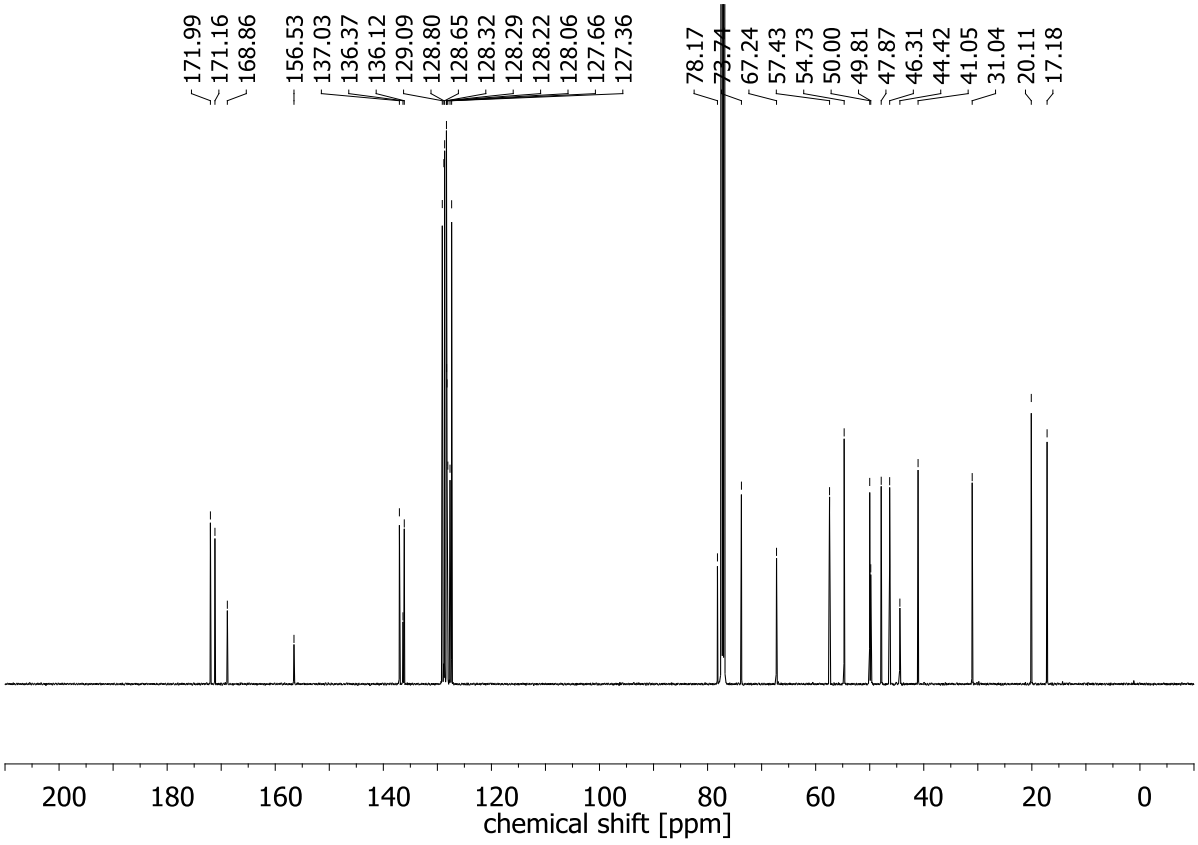
^{1}H NMR spectrum of **19** (500 MHz, $CDCl_3$)



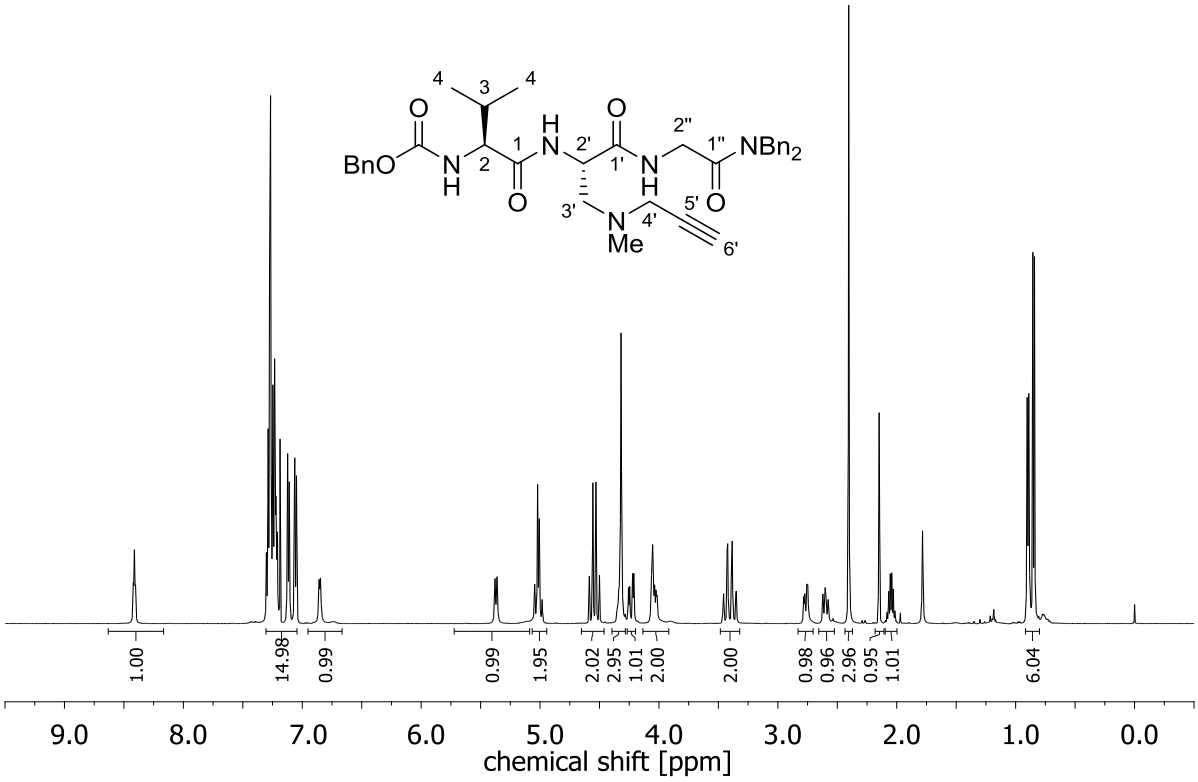
^{13}C NMR spectrum of **19** (126 MHz, $CDCl_3$)



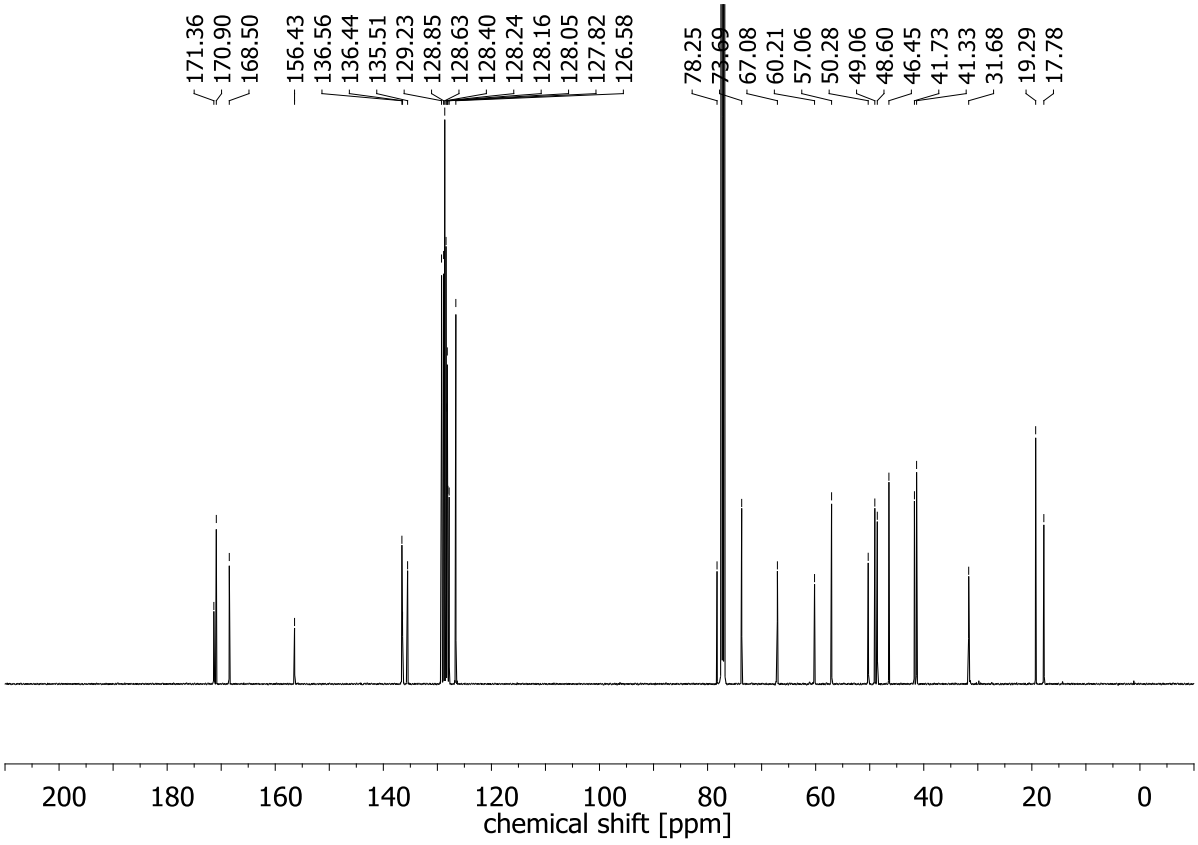
^{1}H NMR spectrum of **20** (500 MHz, $CDCl_3$)



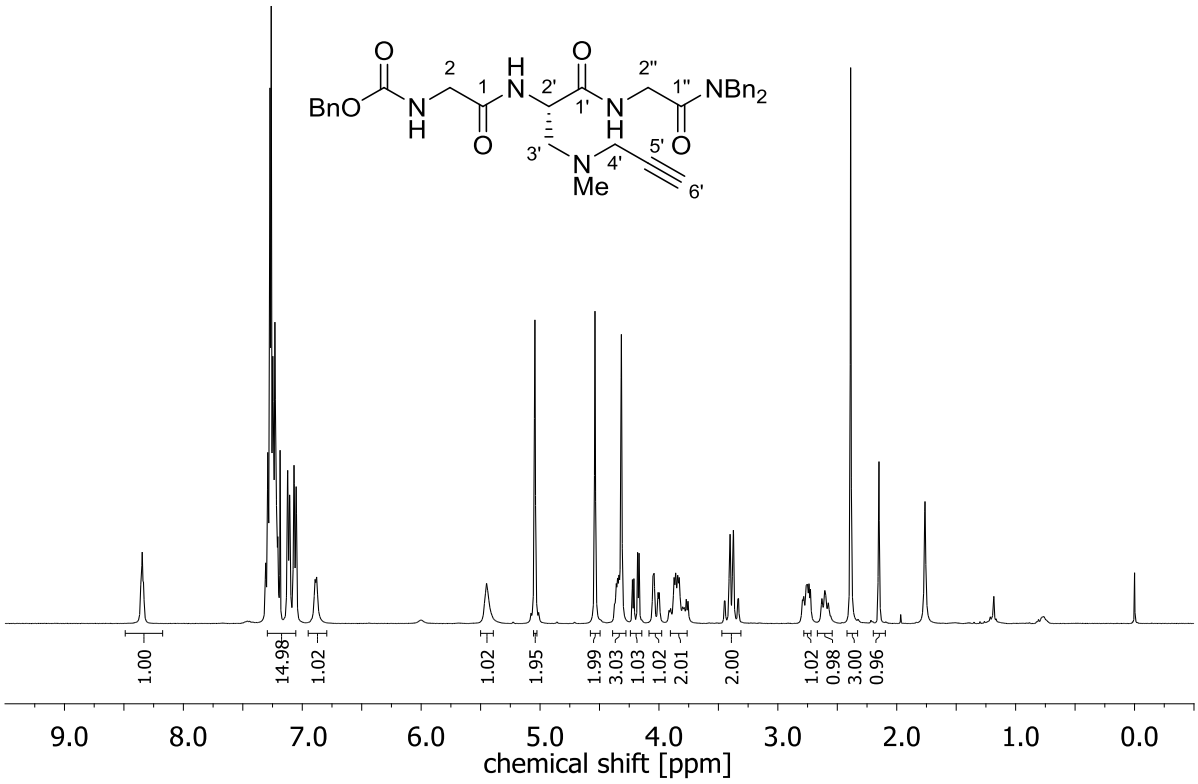
^{13}C NMR spectrum of **20** (126 MHz, $CDCl_3$)



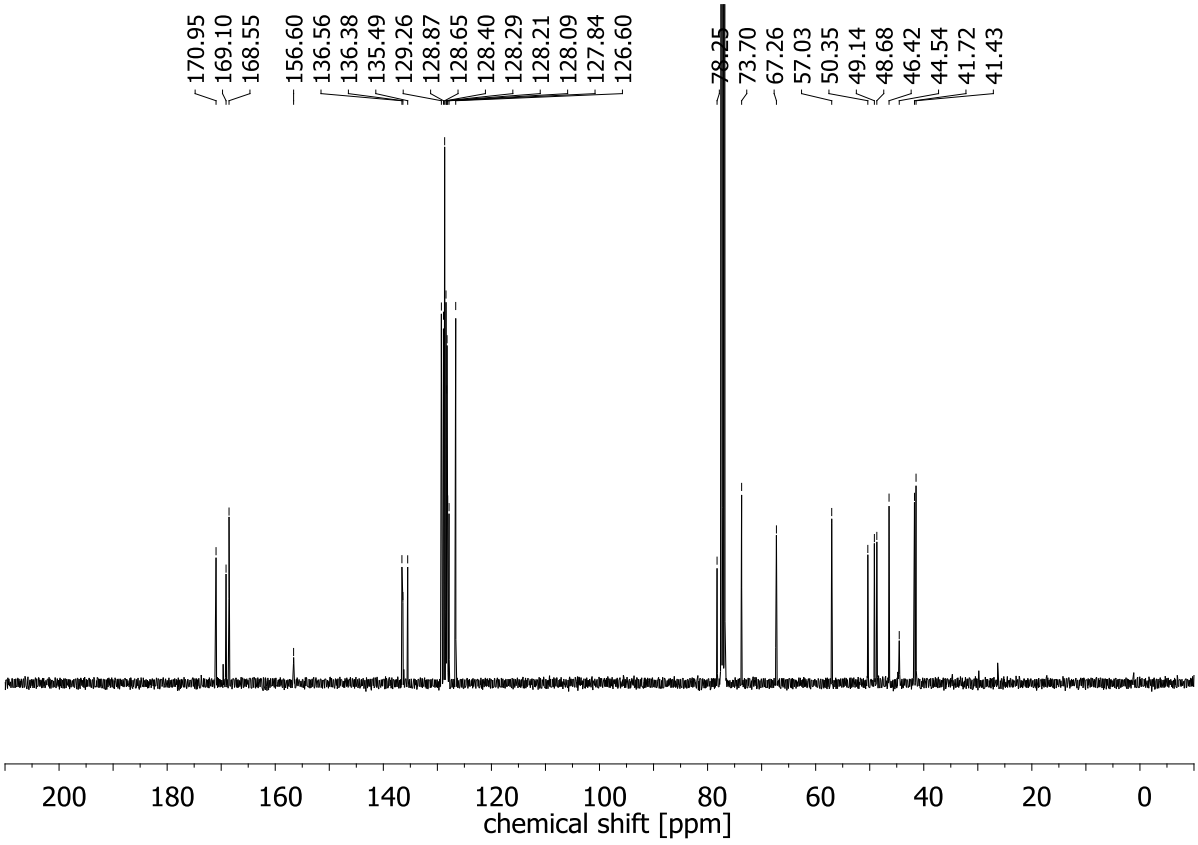
^{1}H NMR spectrum of **21** (500 MHz, $CDCl_3$)



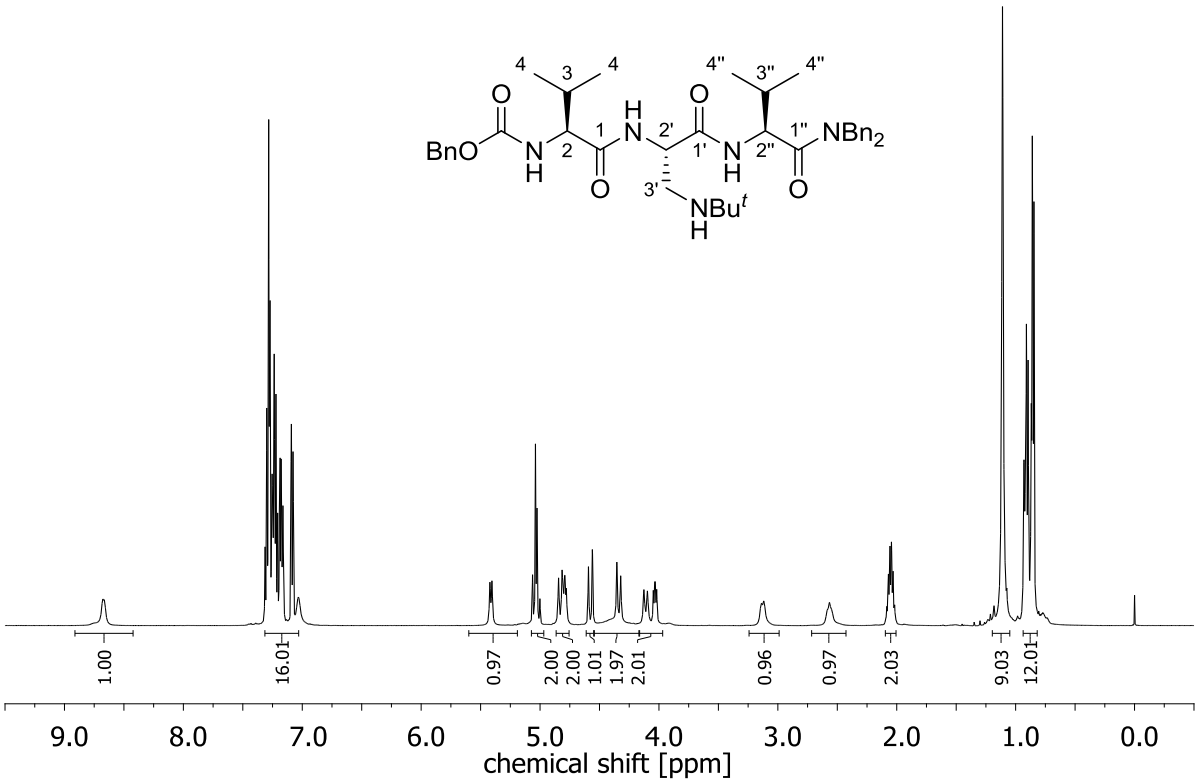
^{13}C NMR spectrum of **21** (126 MHz, $CDCl_3$)



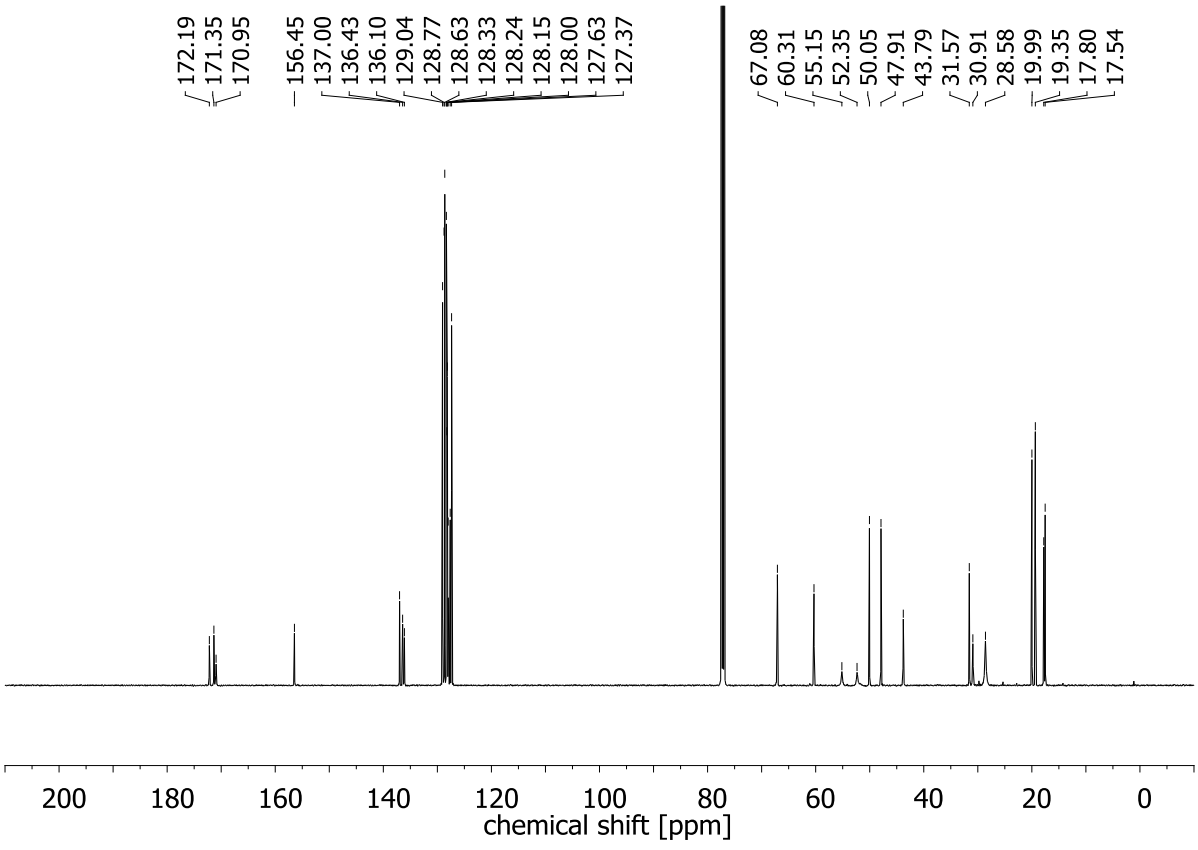
^{1}H NMR spectrum of **22** (400 MHz, $CDCl_3$)



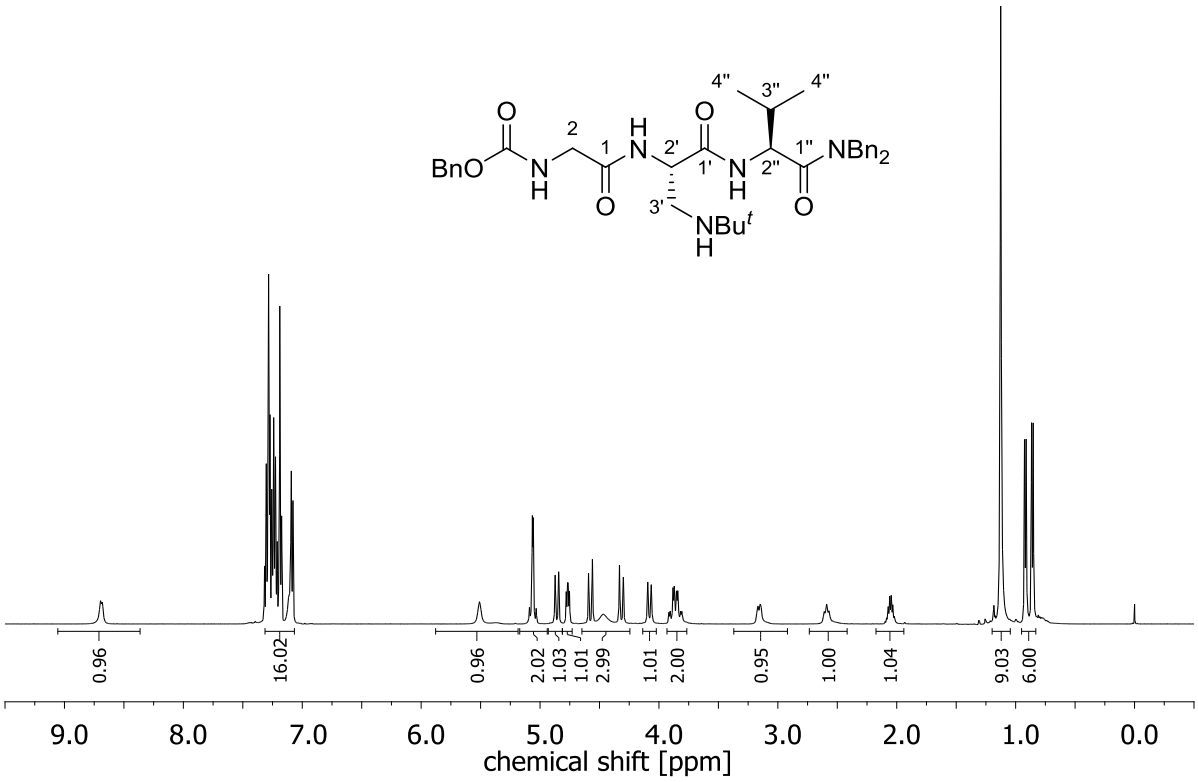
^{13}C NMR spectrum of **22** (101 MHz, $CDCl_3$)



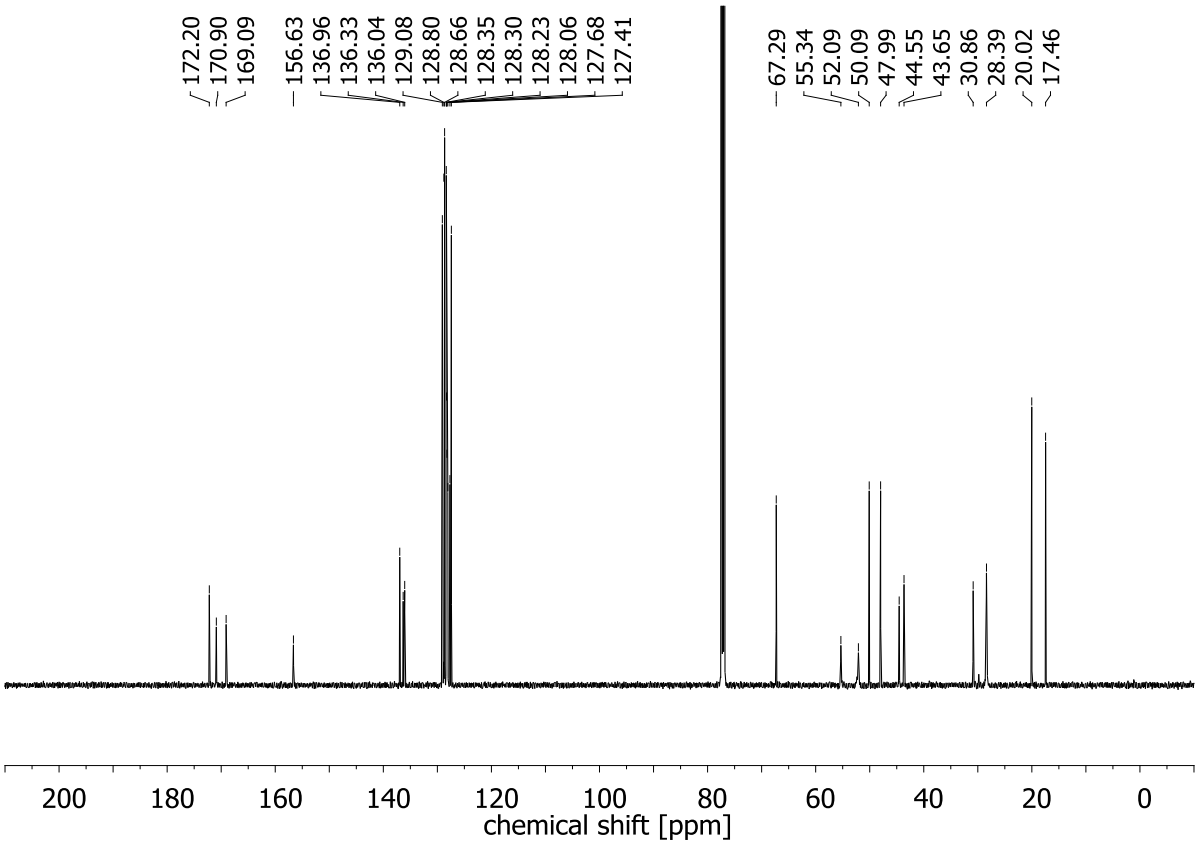
^{1}H NMR spectrum of **23** (500 MHz, $CDCl_3$)



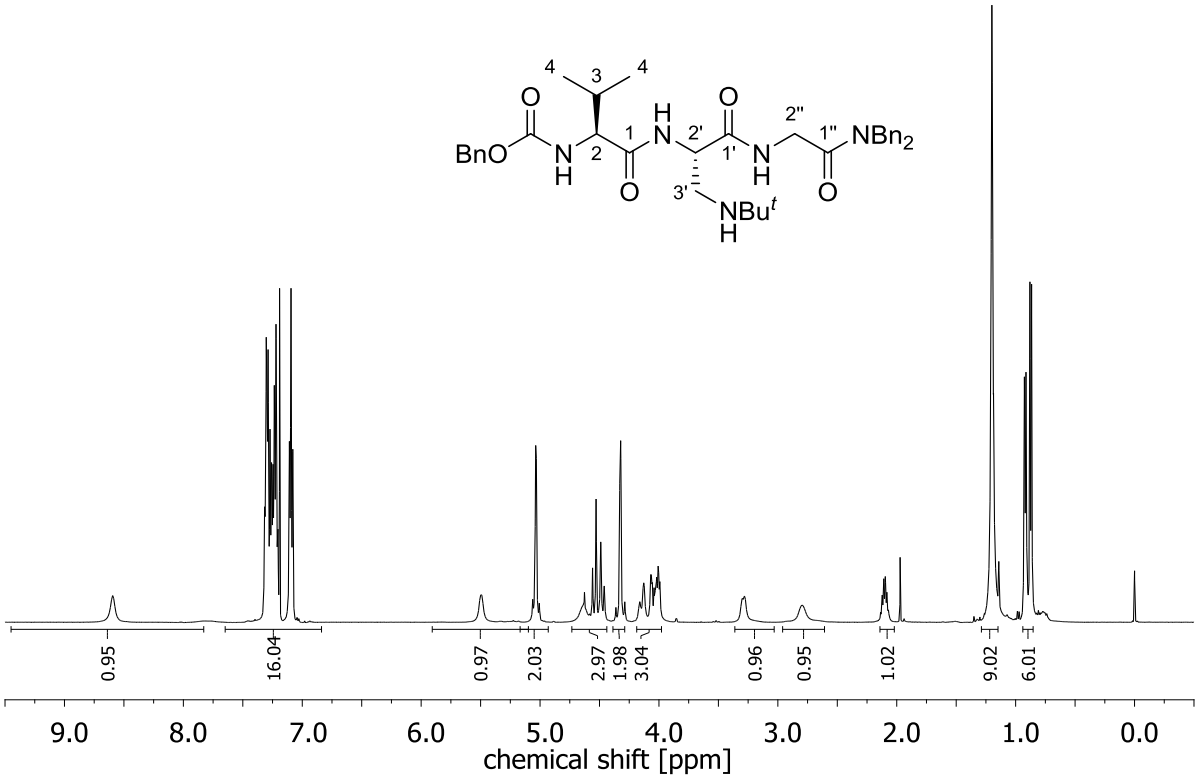
^{13}C NMR spectrum of **23** (126 MHz, $CDCl_3$)



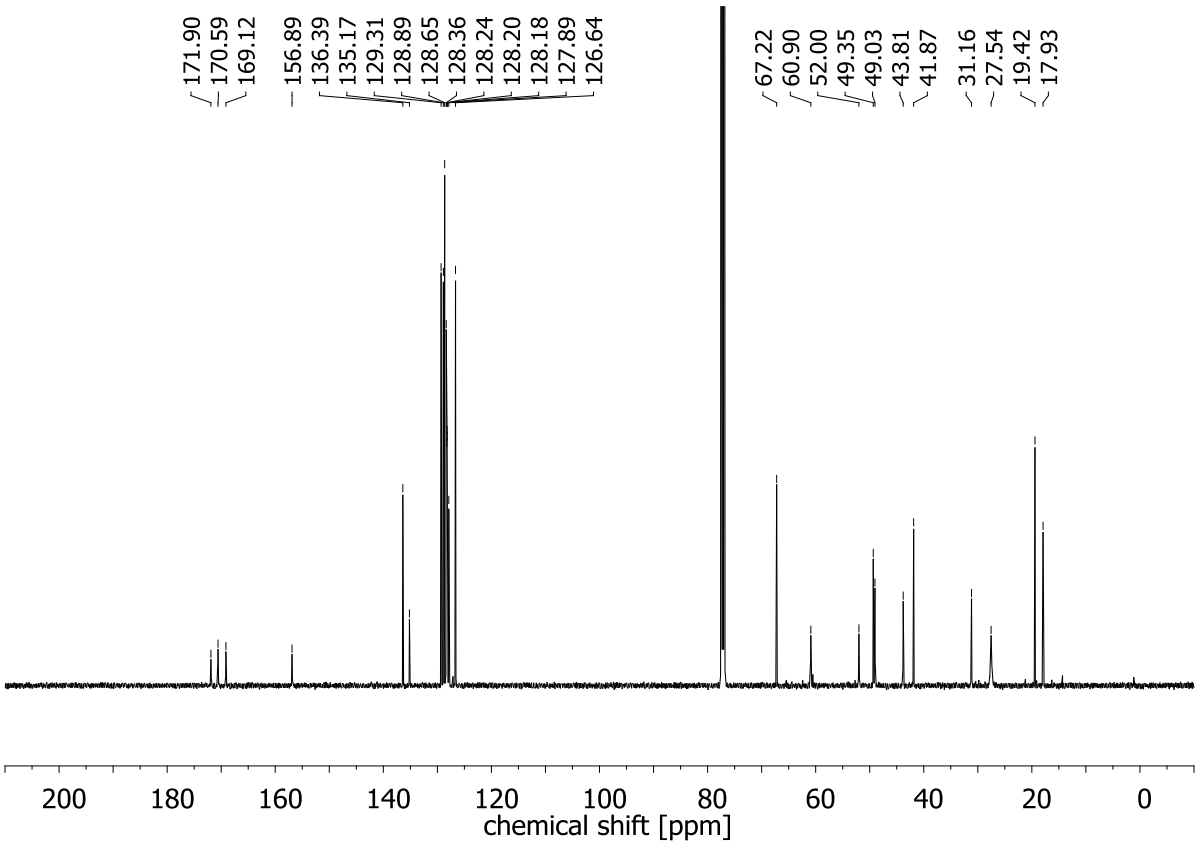
^{1}H NMR spectrum of **24** (500 MHz, $CDCl_3$)



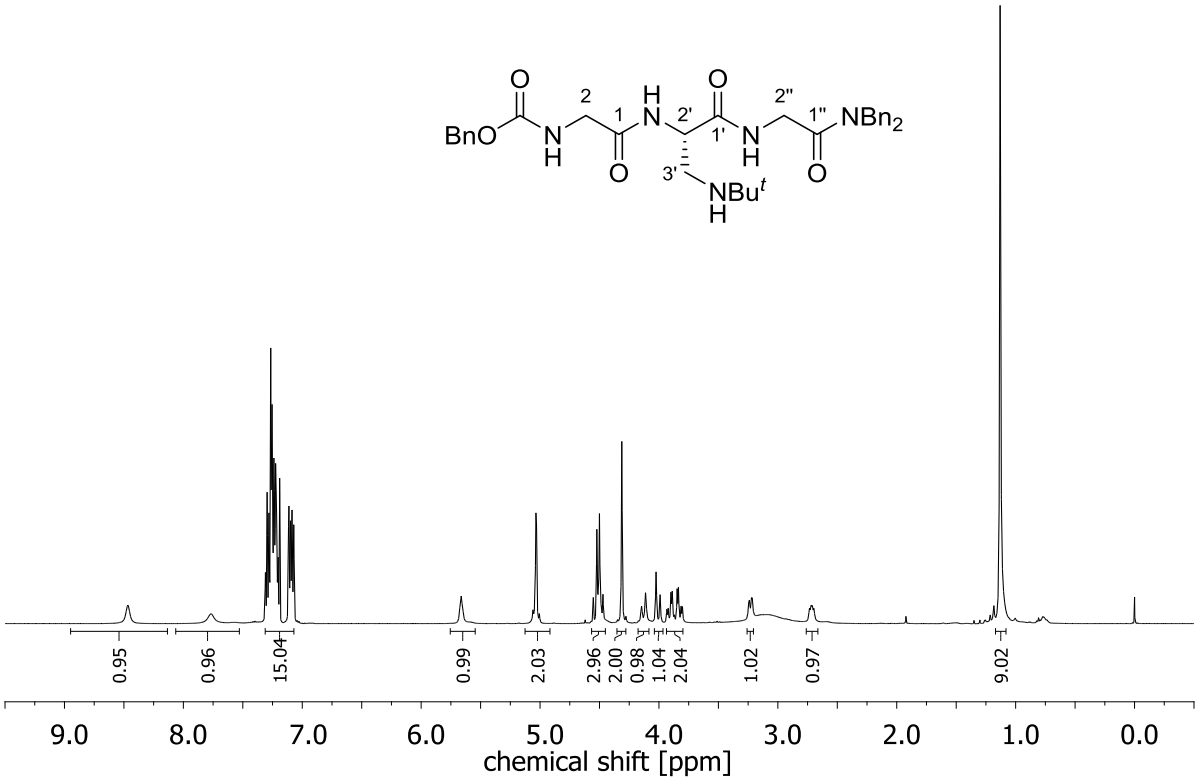
^{13}C NMR spectrum of **24** (126 MHz, $CDCl_3$)



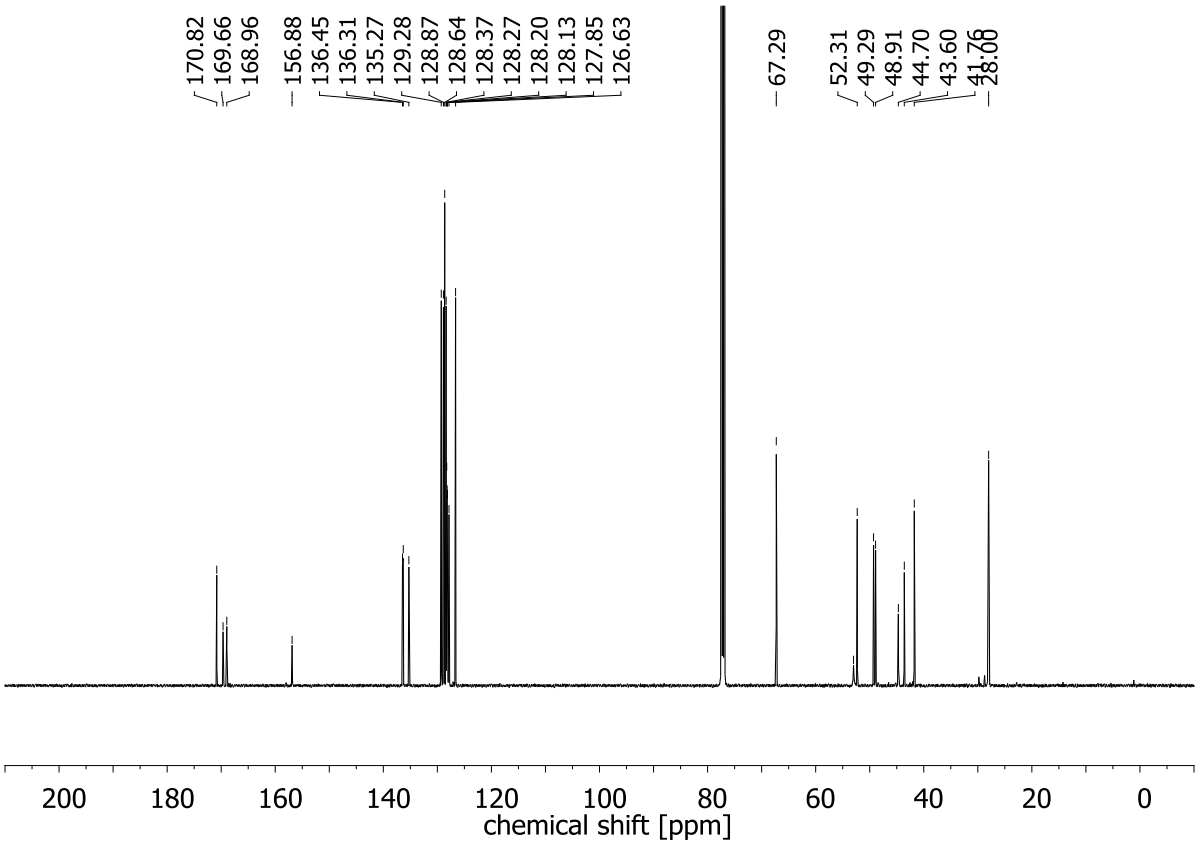
^{1}H NMR spectrum of **25** (500 MHz, $CDCl_3$)



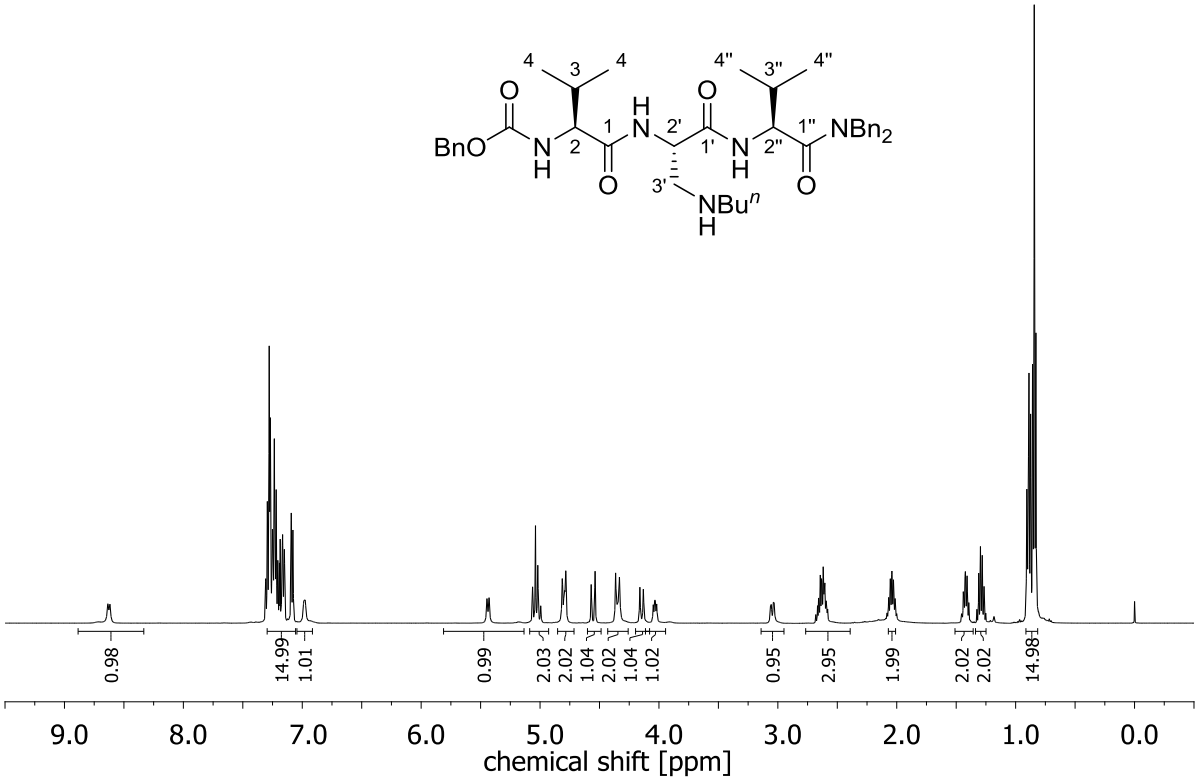
^{13}C NMR spectrum of **25** (126 MHz, $CDCl_3$)



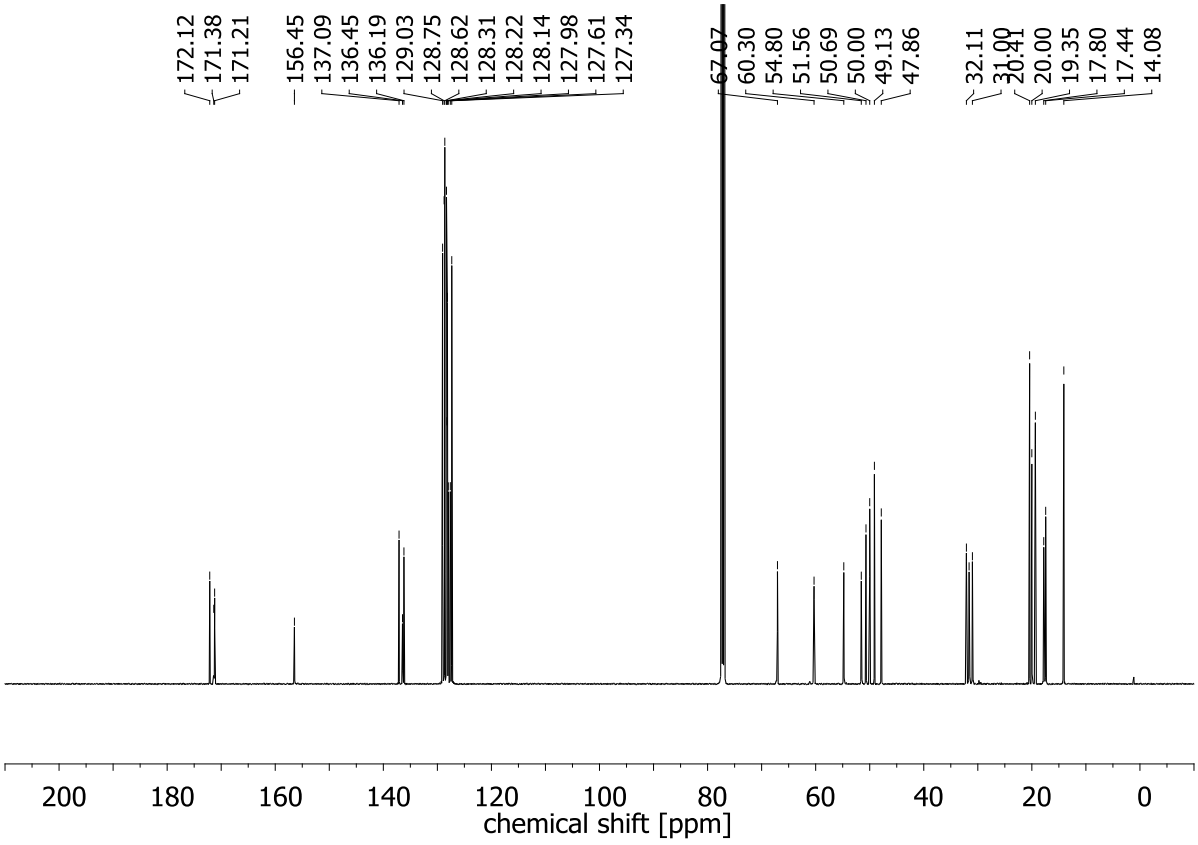
^{1}H NMR spectrum of **26** (500 MHz, $CDCl_3$)



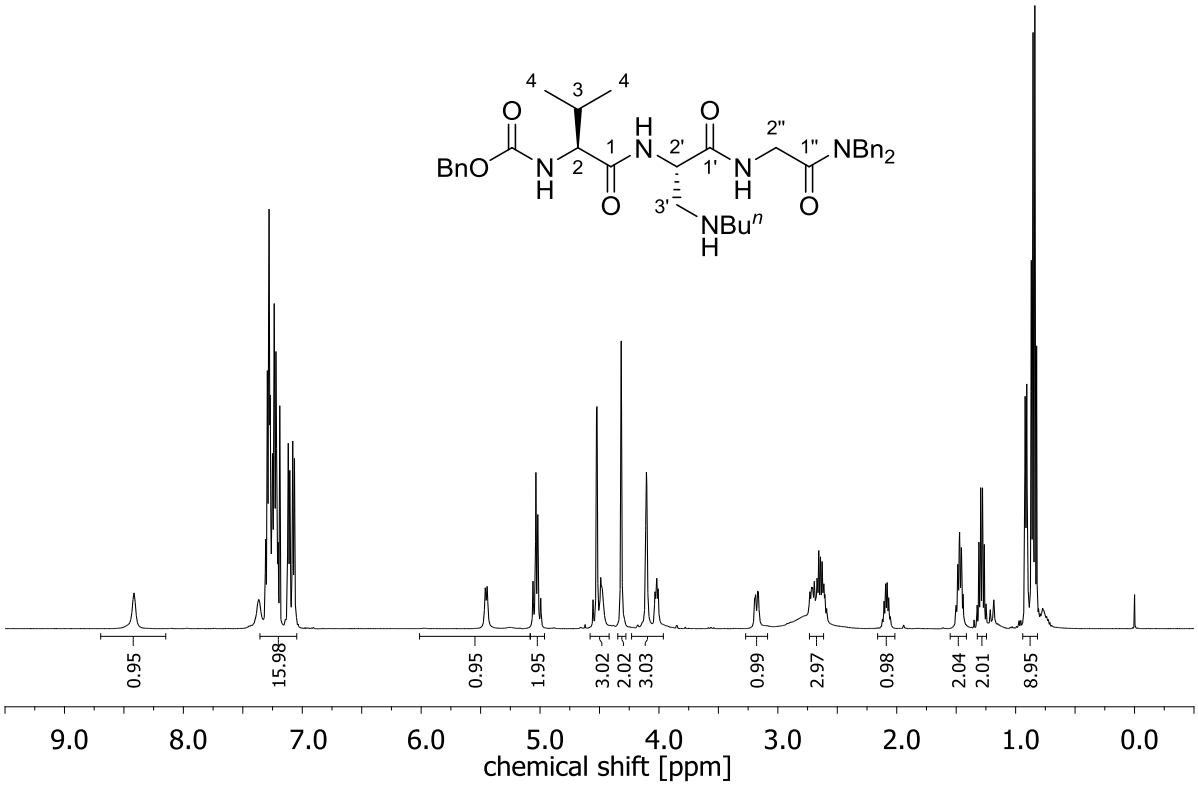
^{13}C NMR spectrum of **26** (126 MHz, $CDCl_3$)



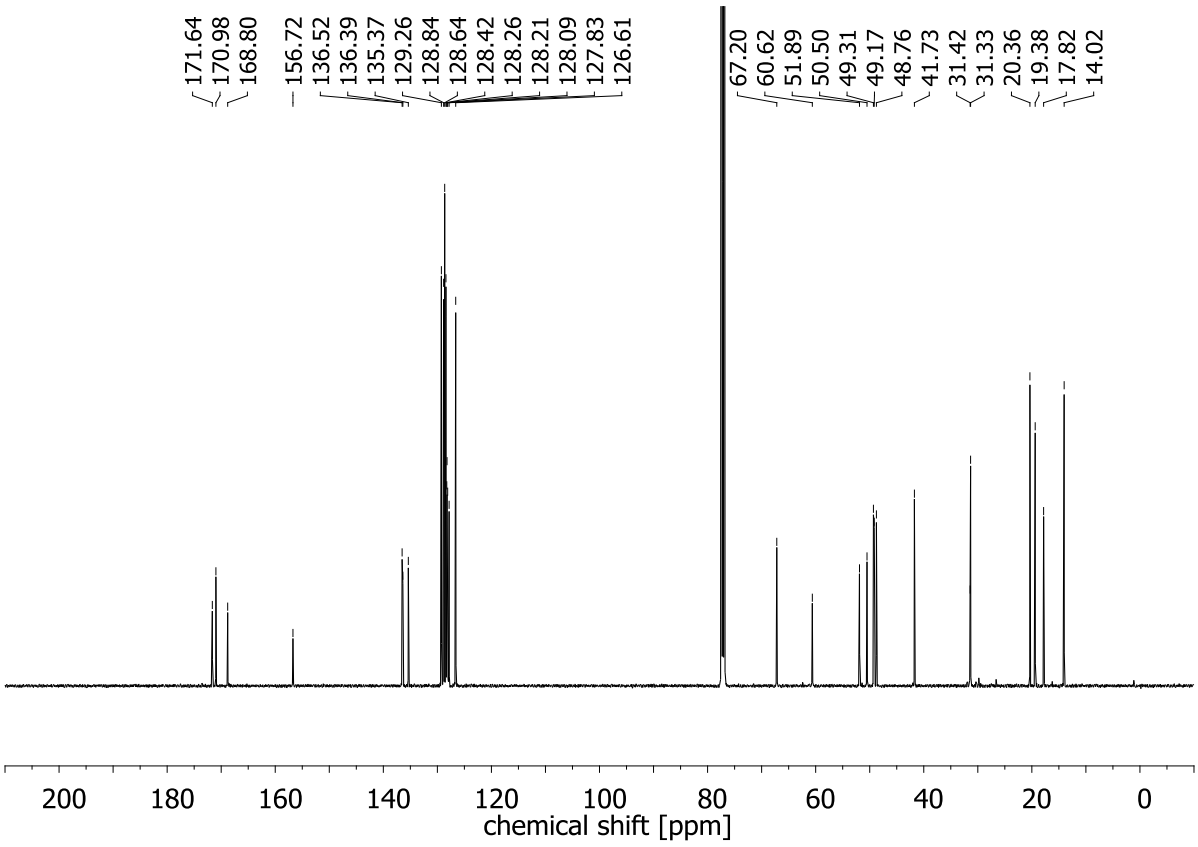
^{1}H NMR spectrum of **27** (500 MHz, $CDCl_3$)



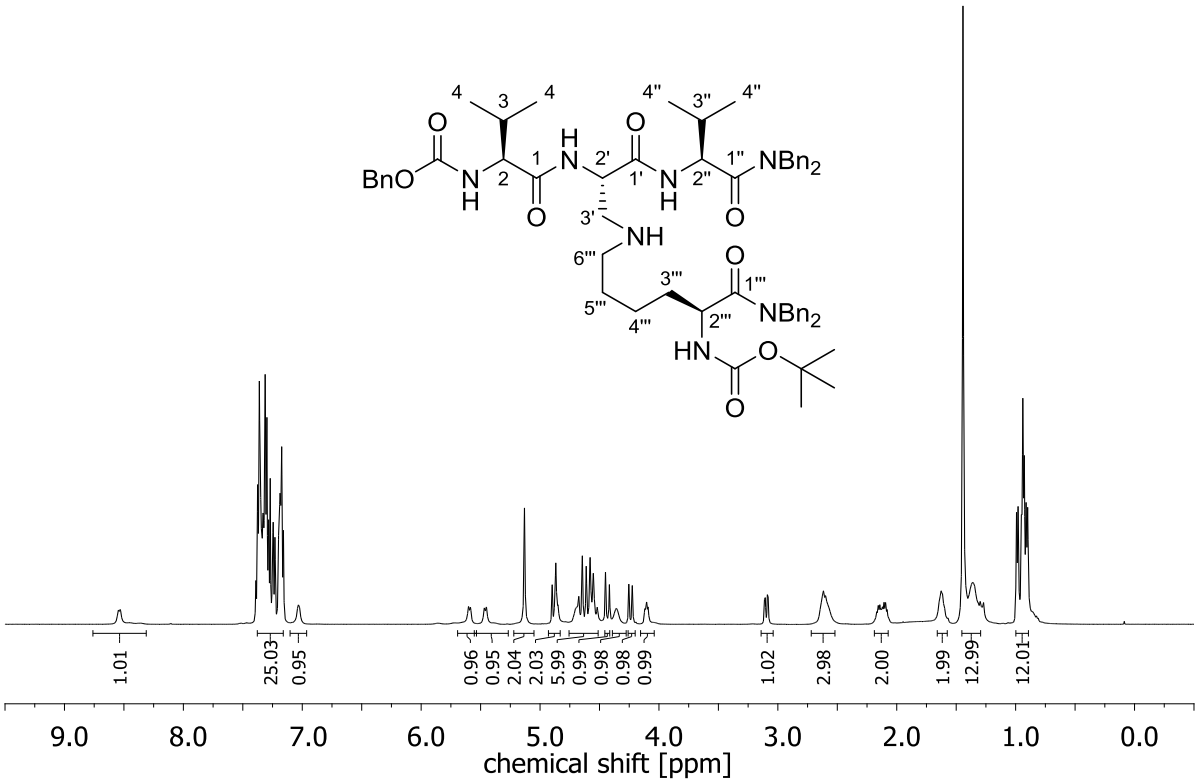
^{13}C NMR spectrum of **27** (126 MHz, $CDCl_3$)



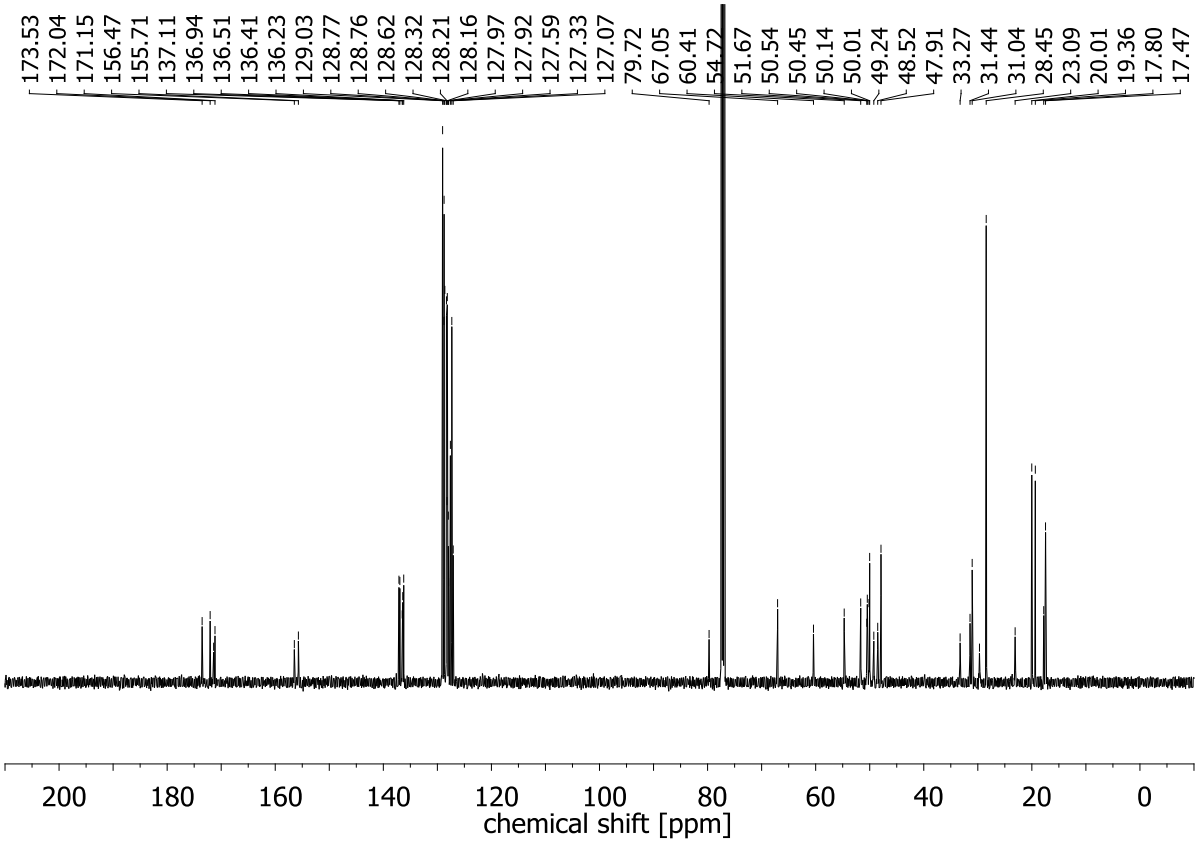
^{1}H NMR spectrum of **28** (500 MHz, $CDCl_3$)



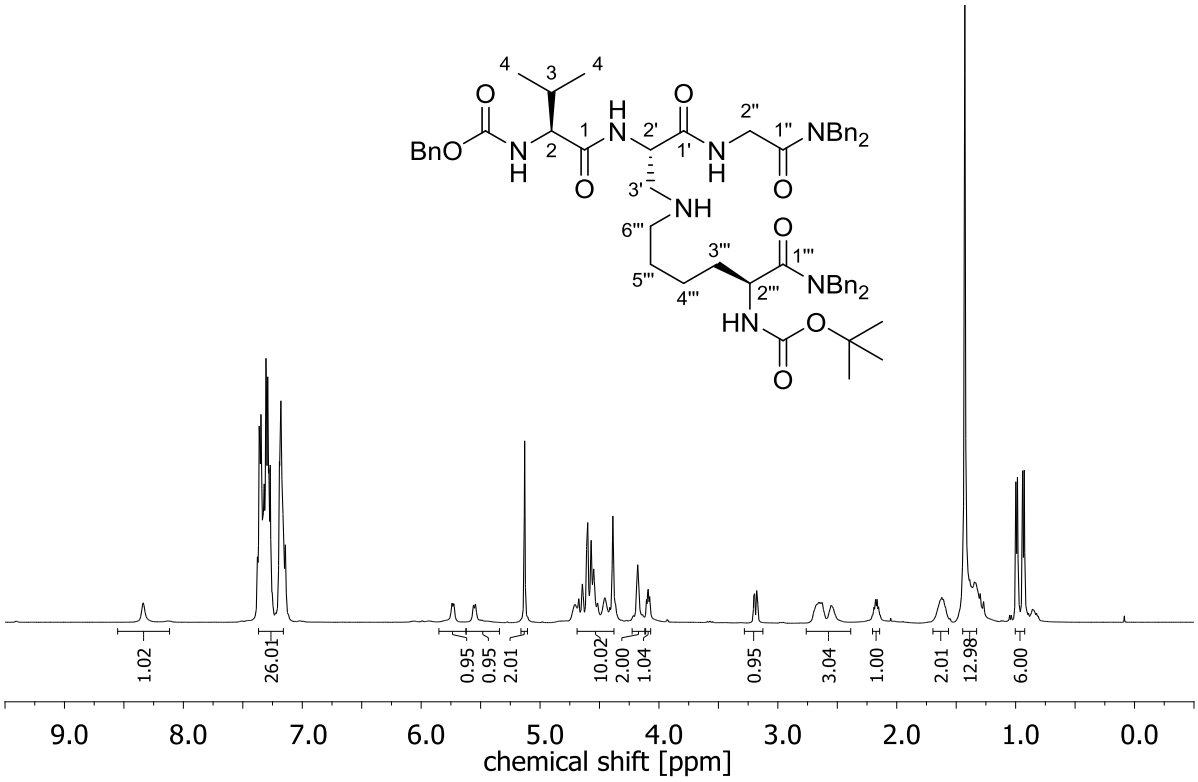
^{13}C NMR spectrum of **28** (126 MHz, $CDCl_3$)



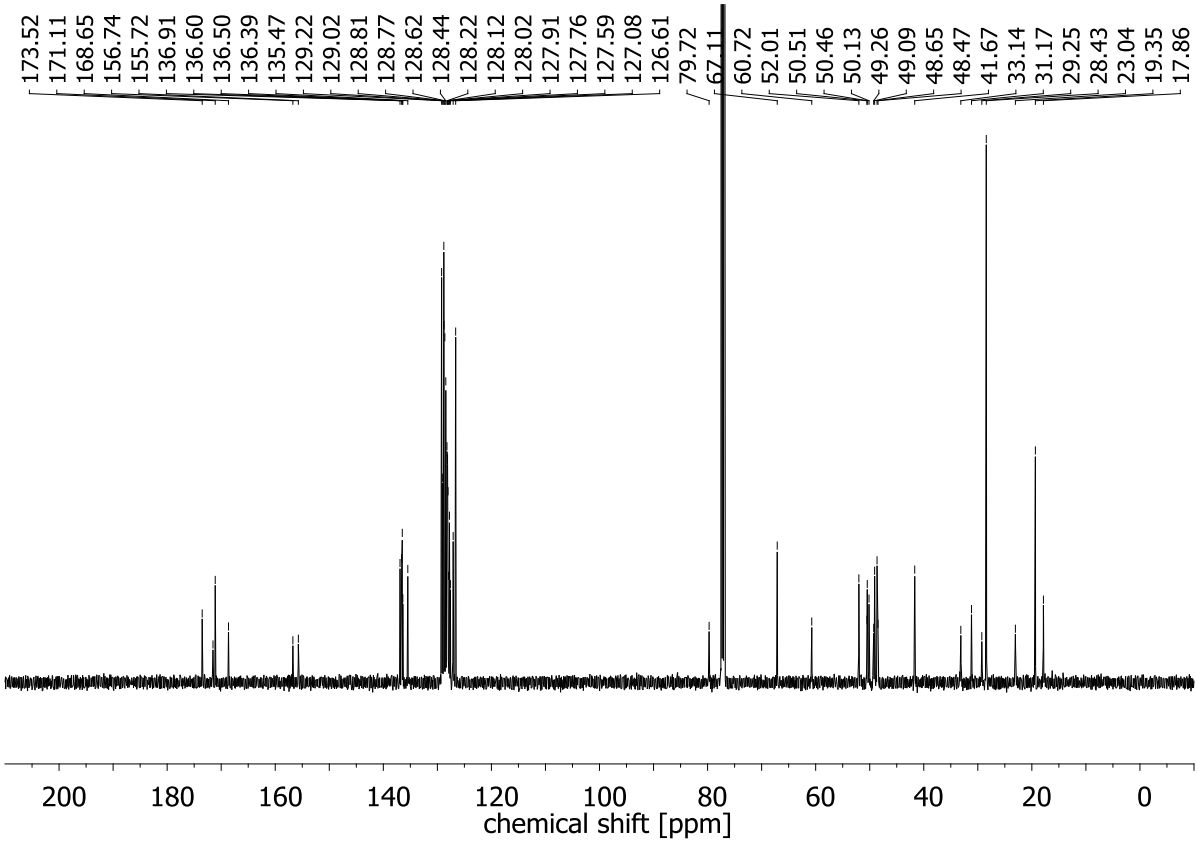
^{1}H NMR spectrum of **29** (500 MHz, $CDCl_3$)



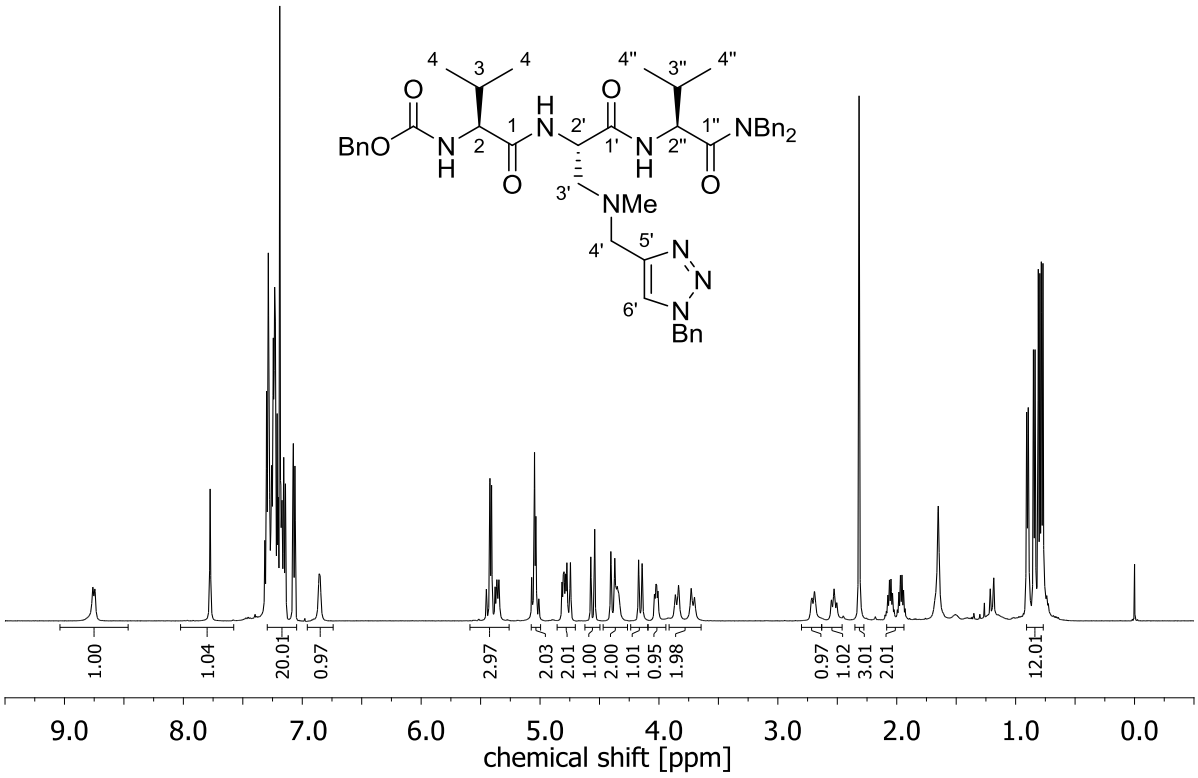
^{13}C NMR spectrum of **29** (126 MHz, $CDCl_3$)



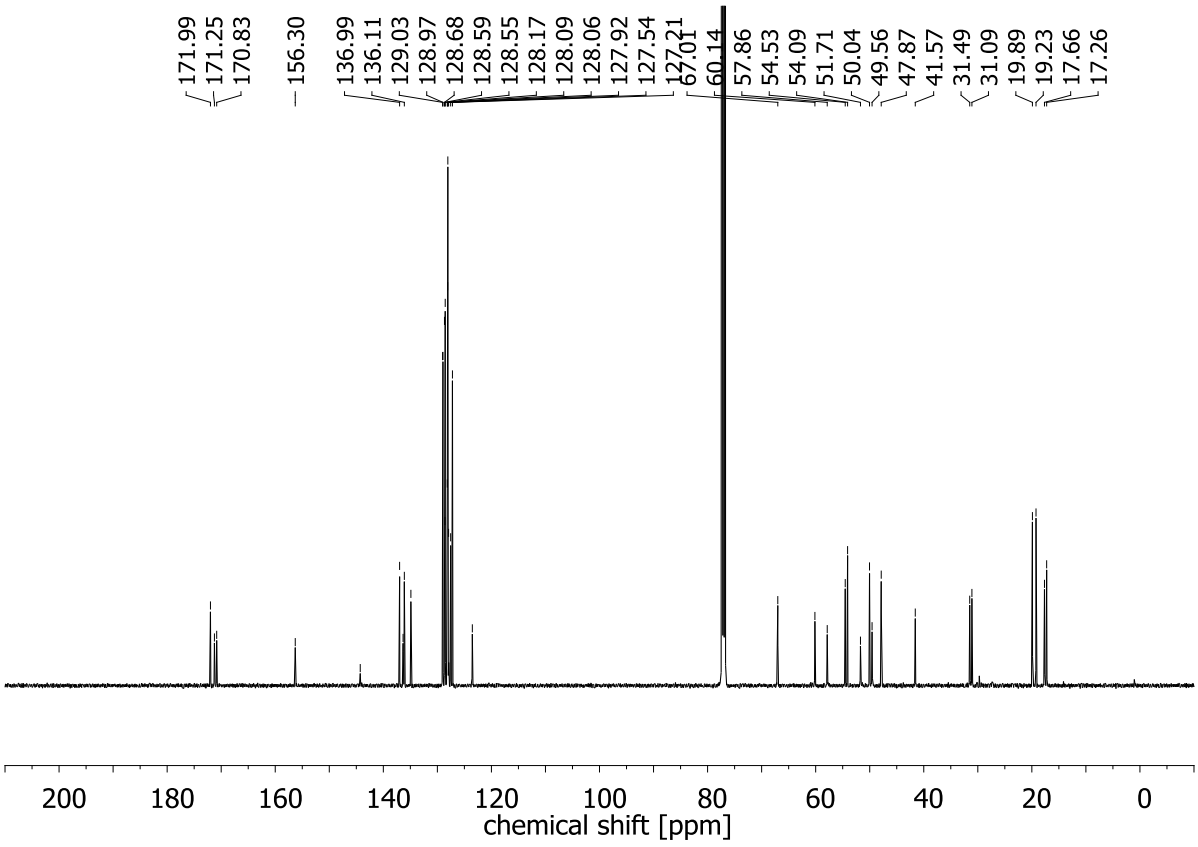
^{1}H NMR spectrum of **30** (500 MHz, $CDCl_3$)



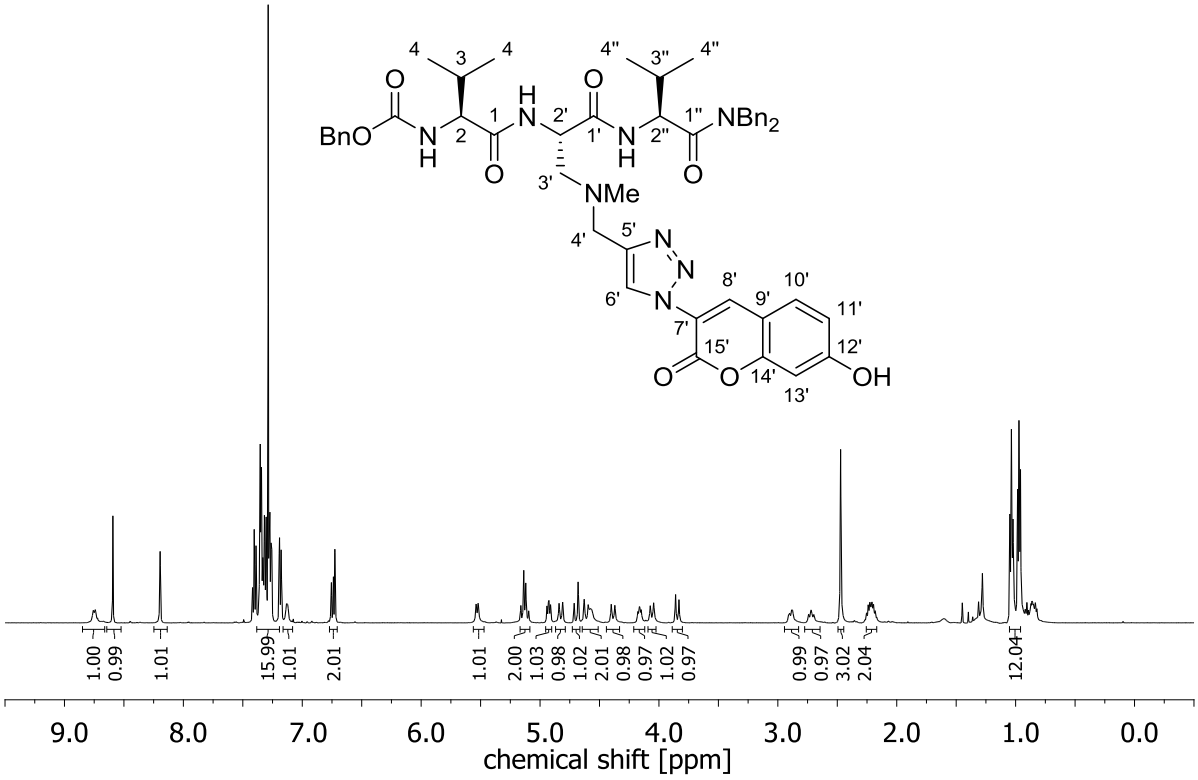
^{13}C NMR spectrum of **30** (126 MHz, $CDCl_3$)



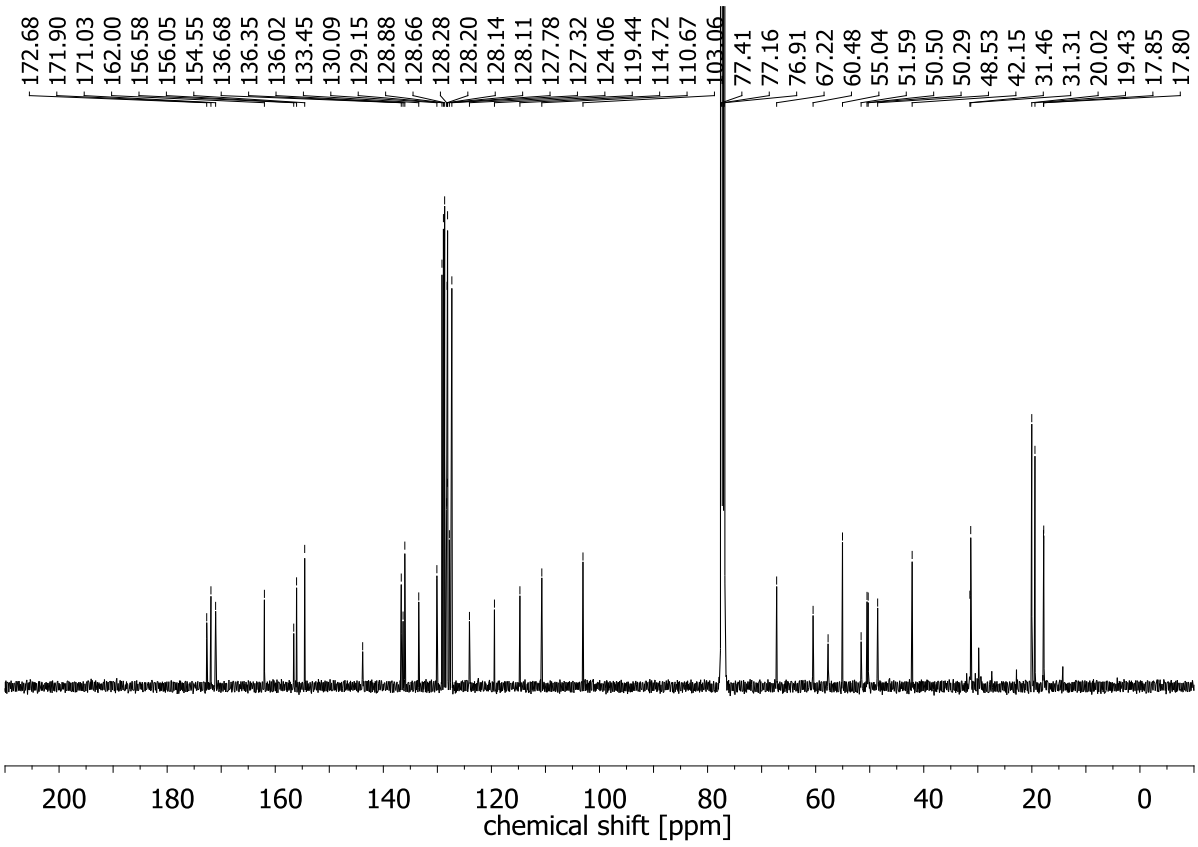
^{1}H NMR spectrum of **31** (500 MHz, $CDCl_3$)



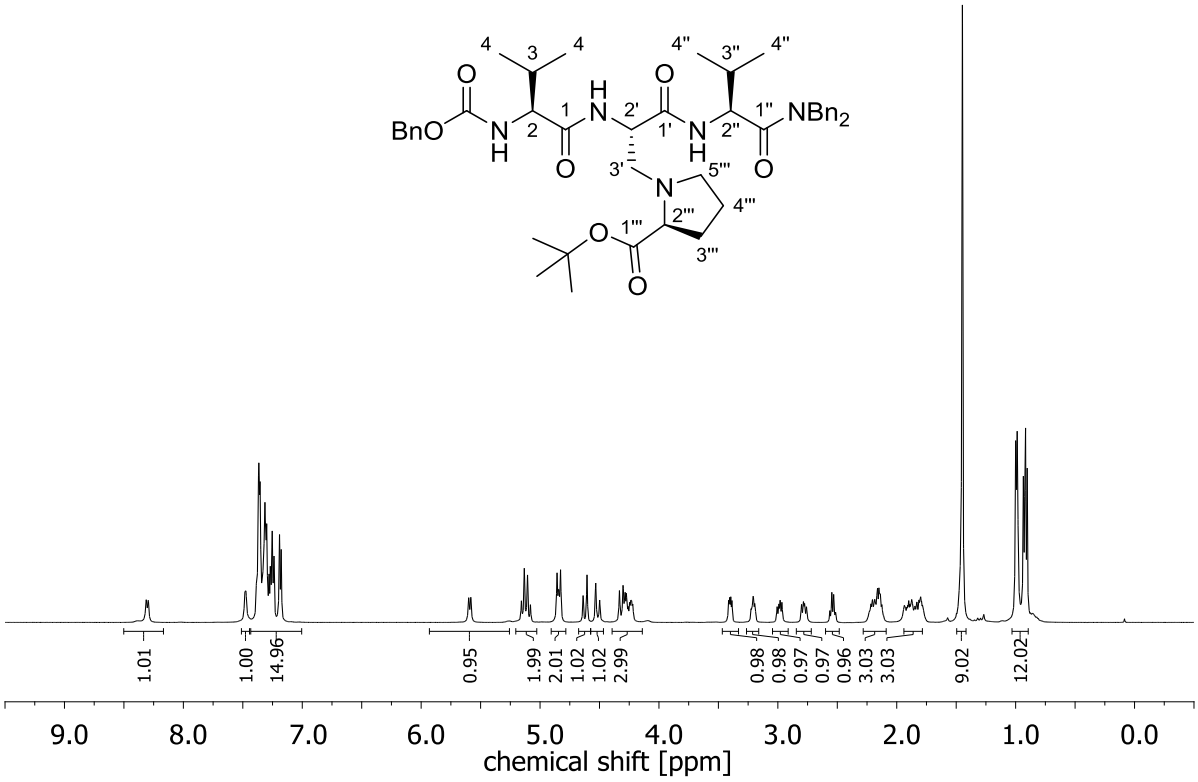
^{13}C NMR spectrum of **31** (126 MHz, $CDCl_3$)



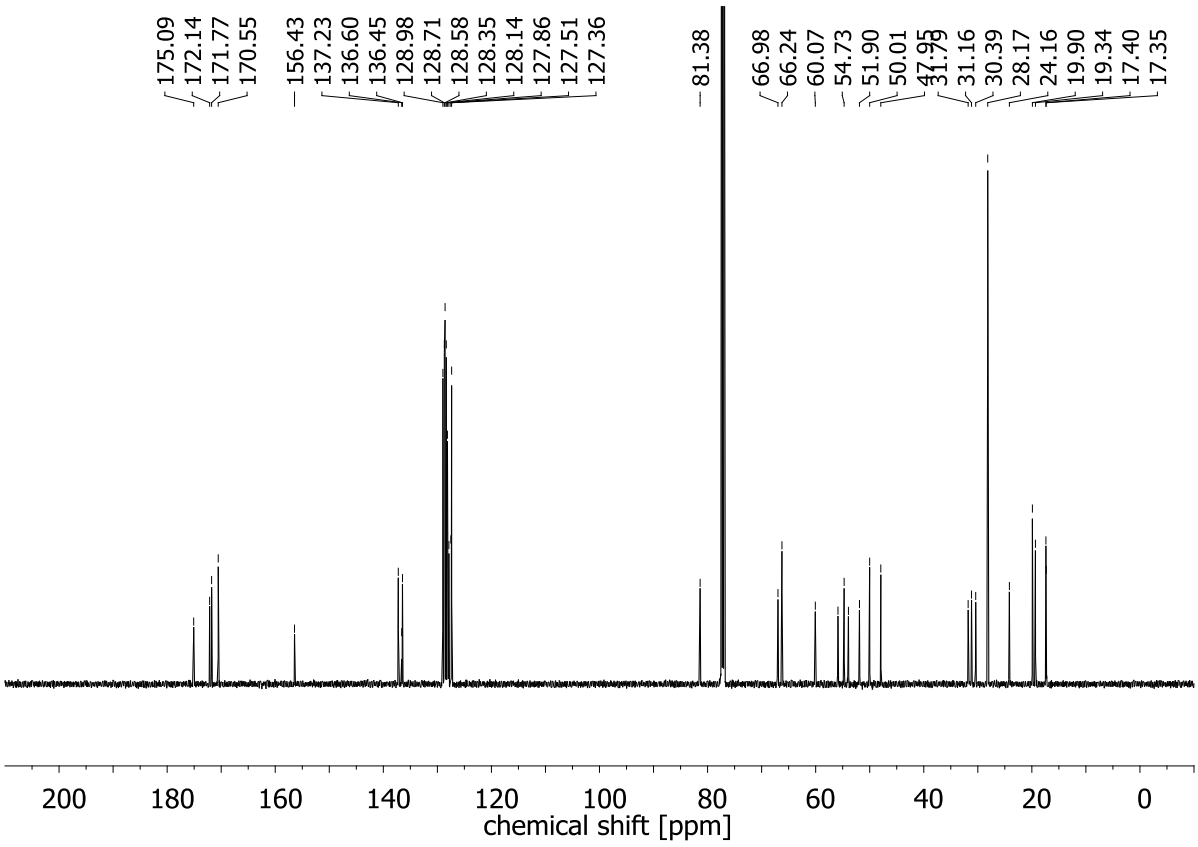
^{1}H NMR spectrum of **32** (500 MHz, $CDCl_3$)



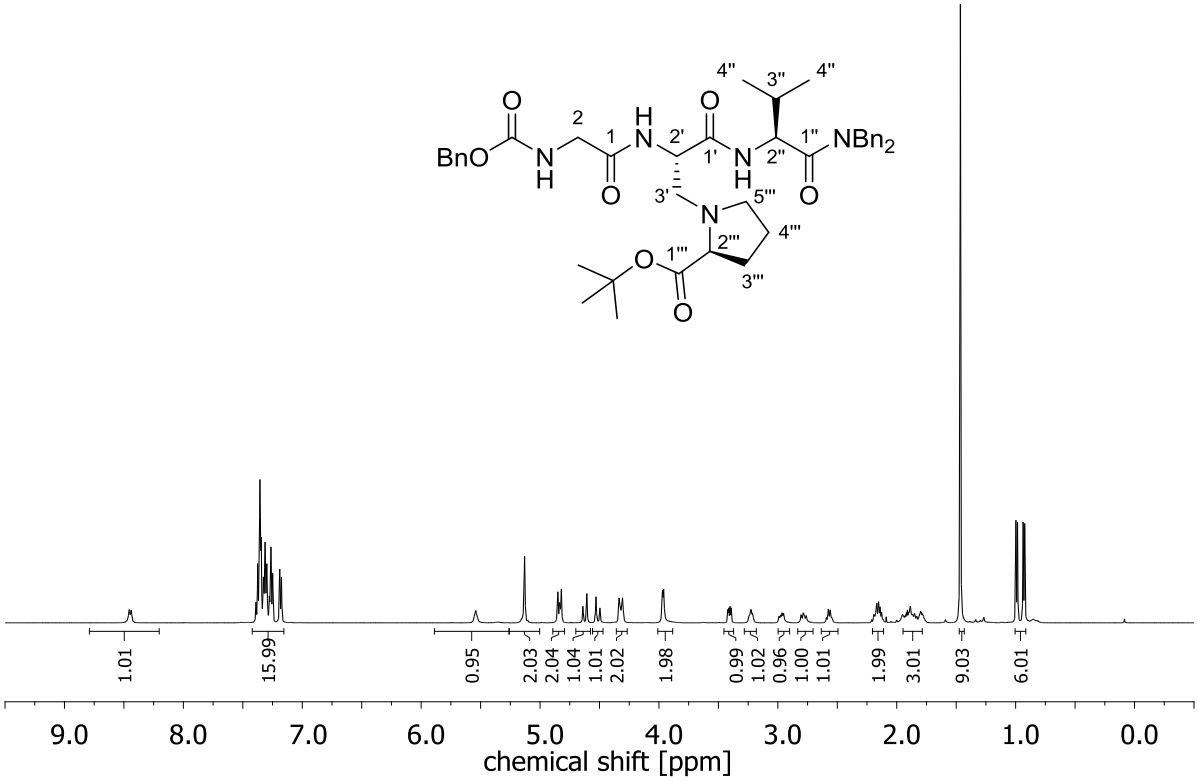
^{13}C NMR spectrum of **32** (126 MHz, $CDCl_3$)



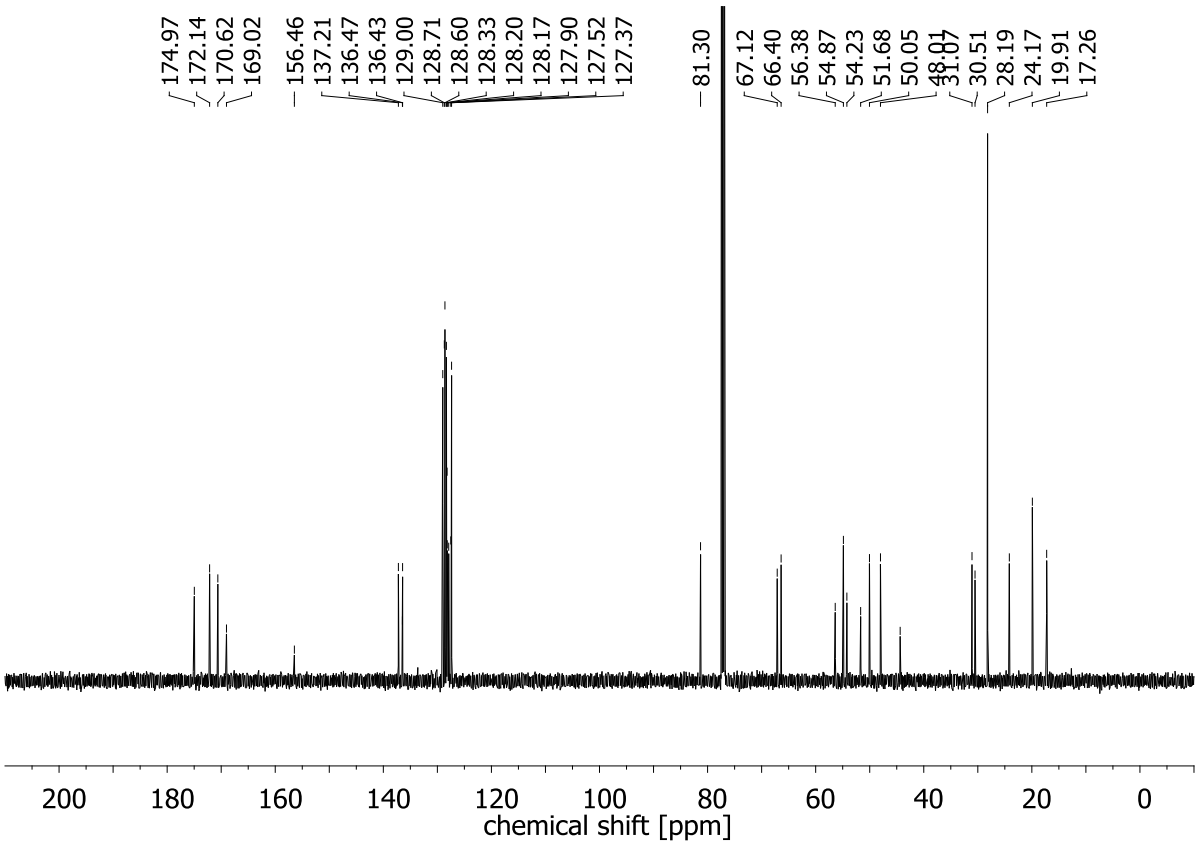
^{1}H NMR spectrum of **33** (500 MHz, $CDCl_3$)



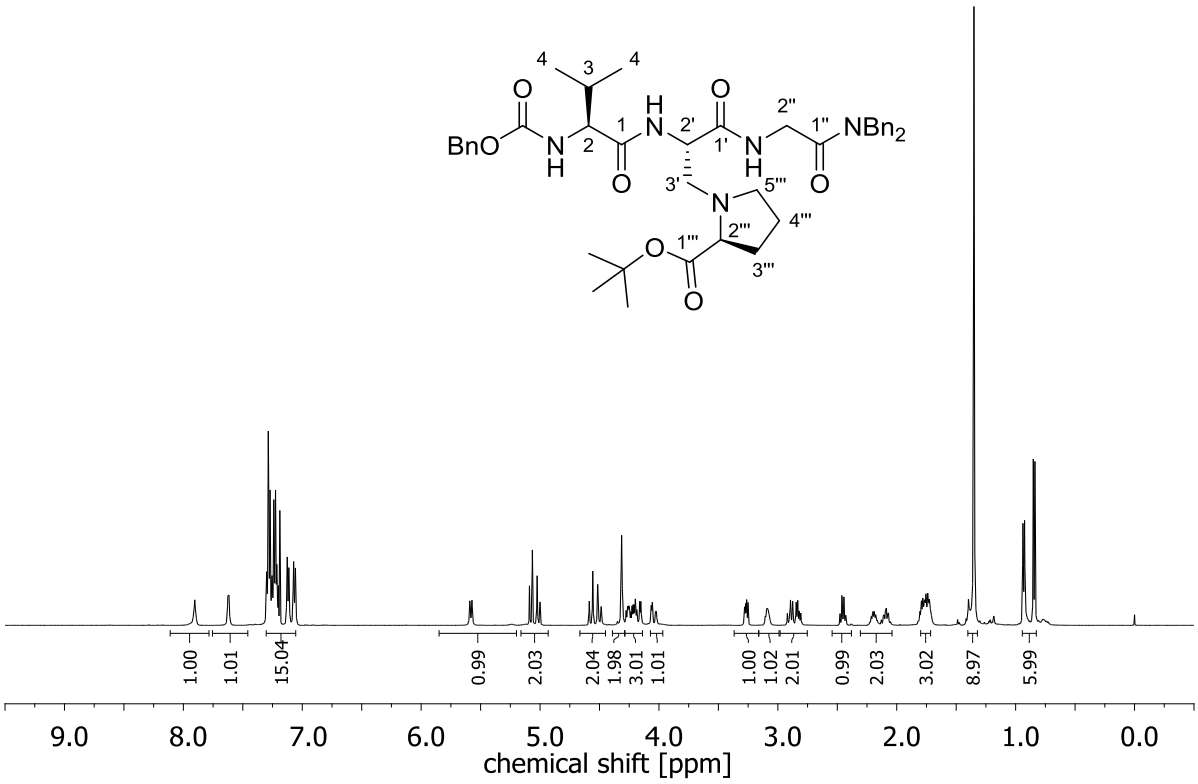
^{13}C NMR spectrum of **33** (126 MHz, $CDCl_3$)



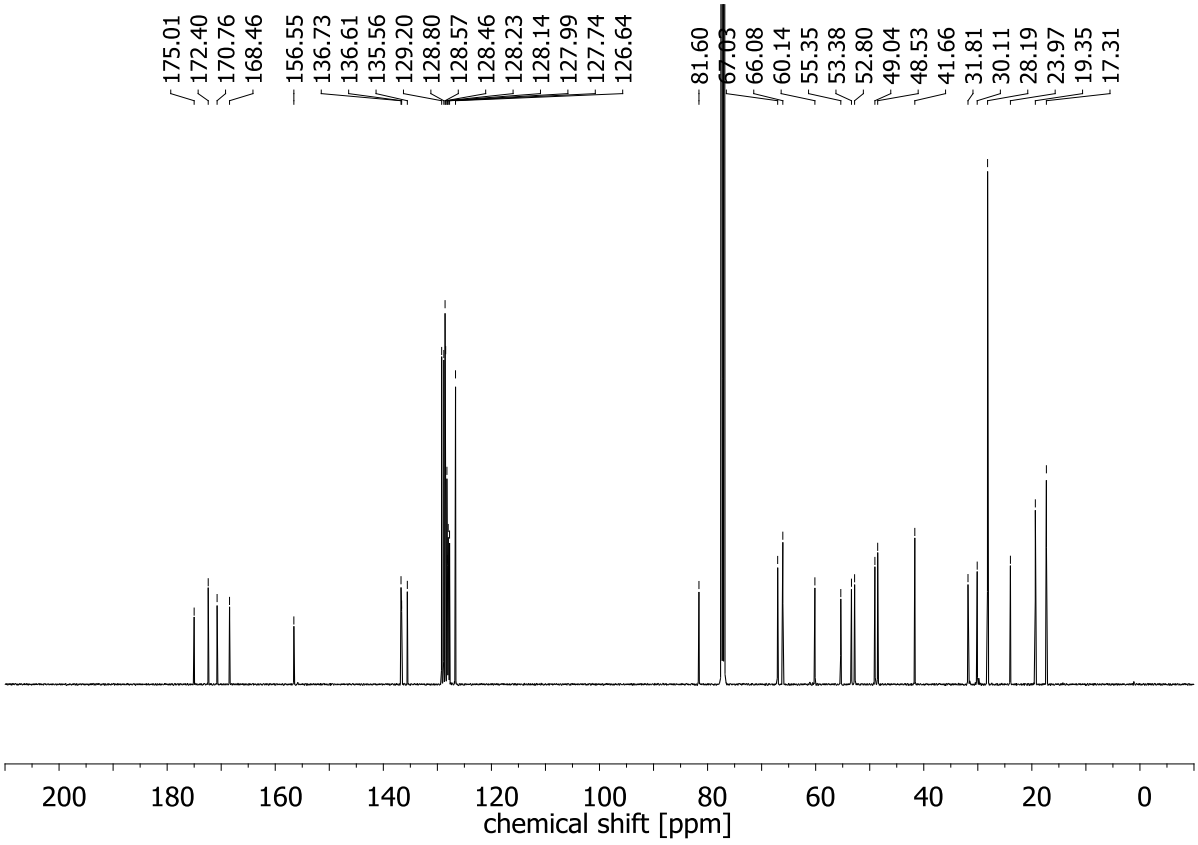
^{1}H NMR spectrum of **34** (500 MHz, $CDCl_3$)



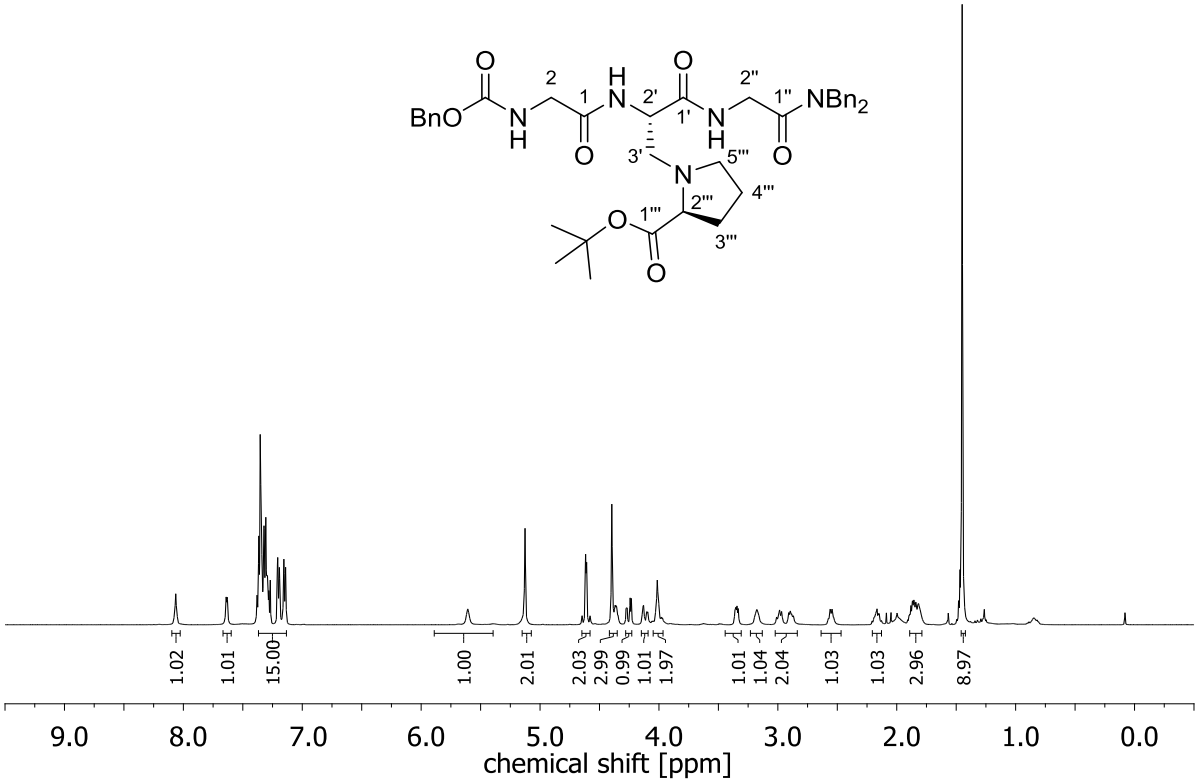
^{13}C NMR spectrum of **34** (126 MHz, $CDCl_3$)



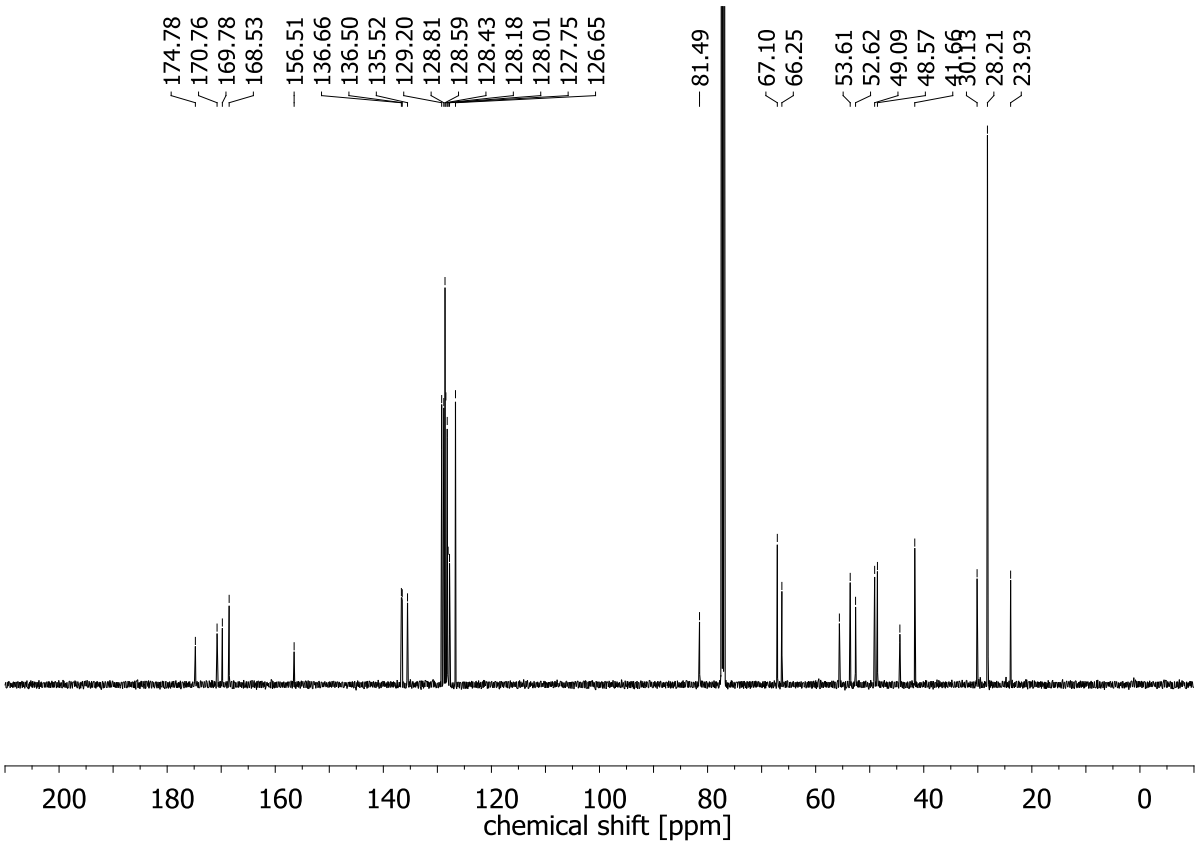
^{1}H NMR spectrum of **35** (500 MHz, $CDCl_3$)



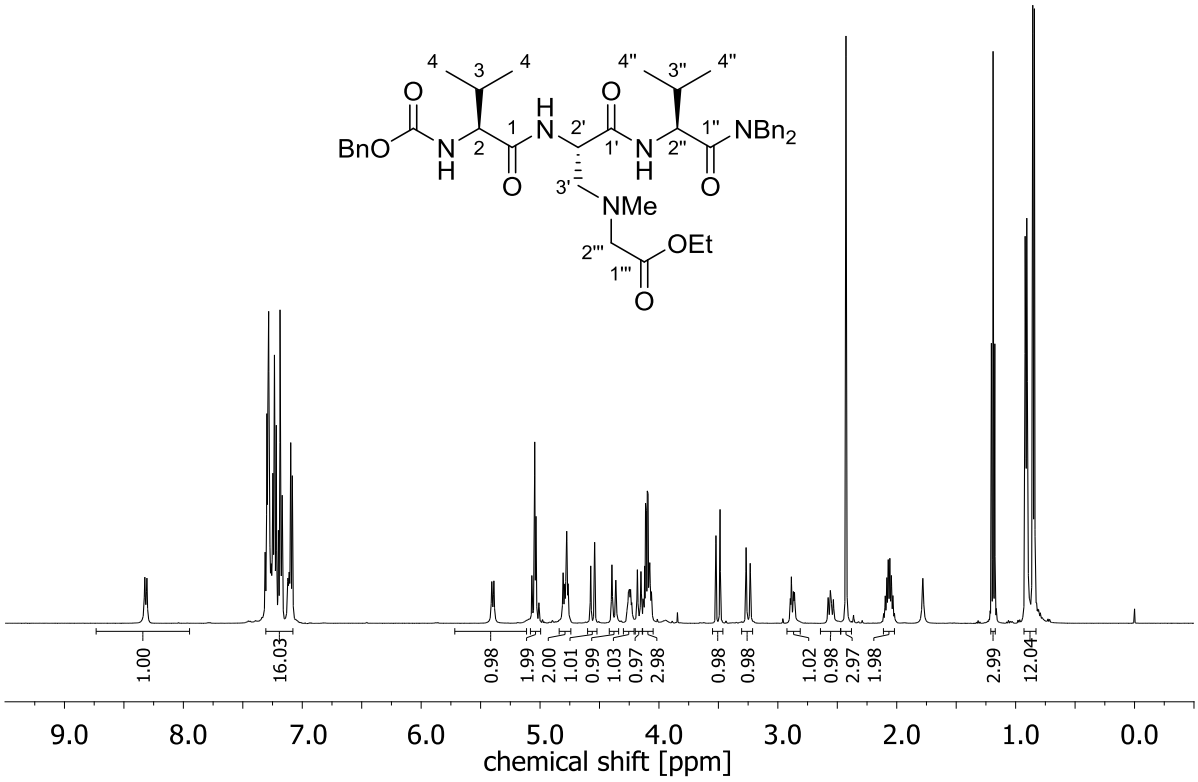
^{13}C NMR spectrum of **35** (126 MHz, $CDCl_3$)



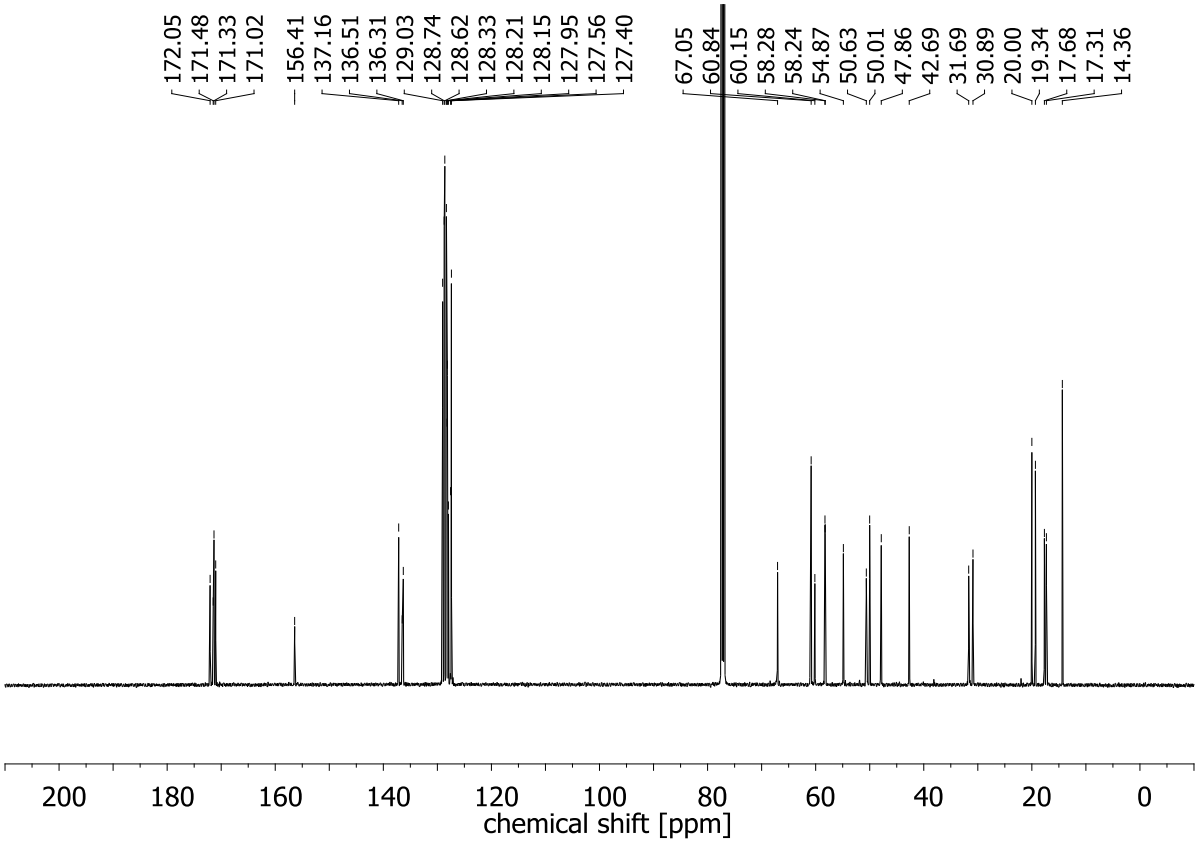
^{1}H NMR spectrum of **36** (500 MHz, $CDCl_3$)



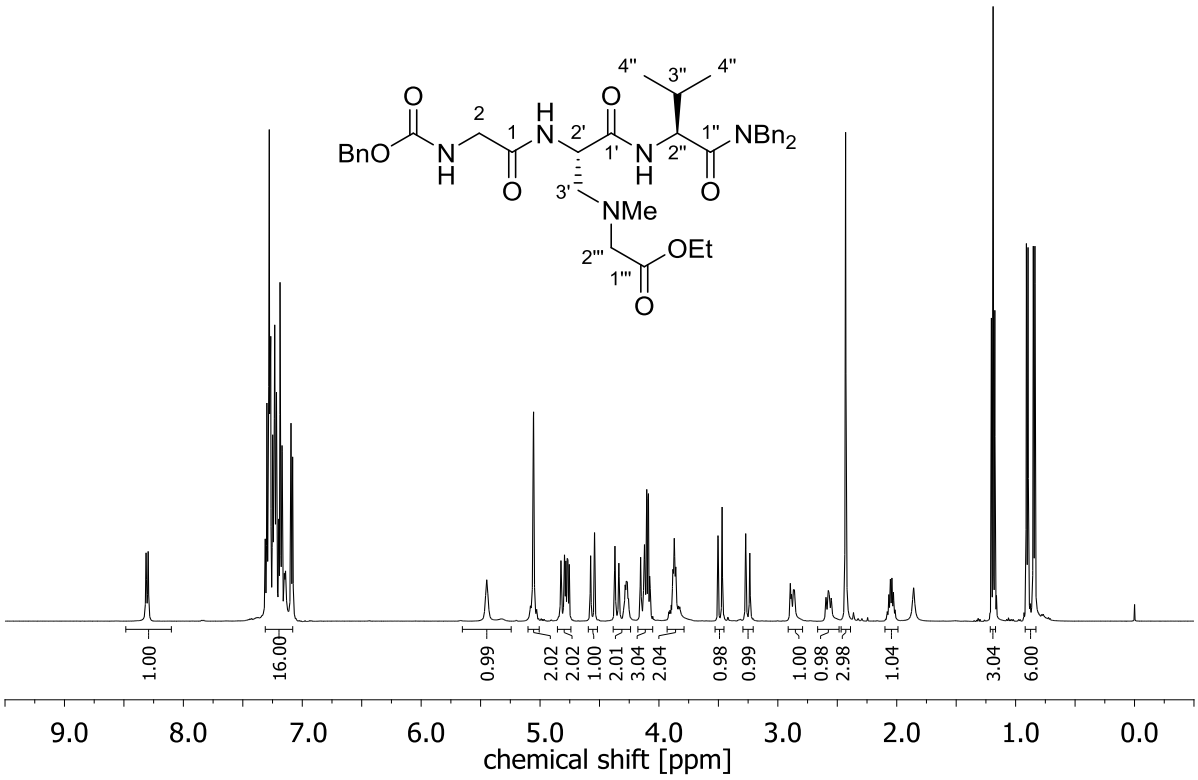
^{13}C NMR spectrum of **36** (126 MHz, $CDCl_3$)



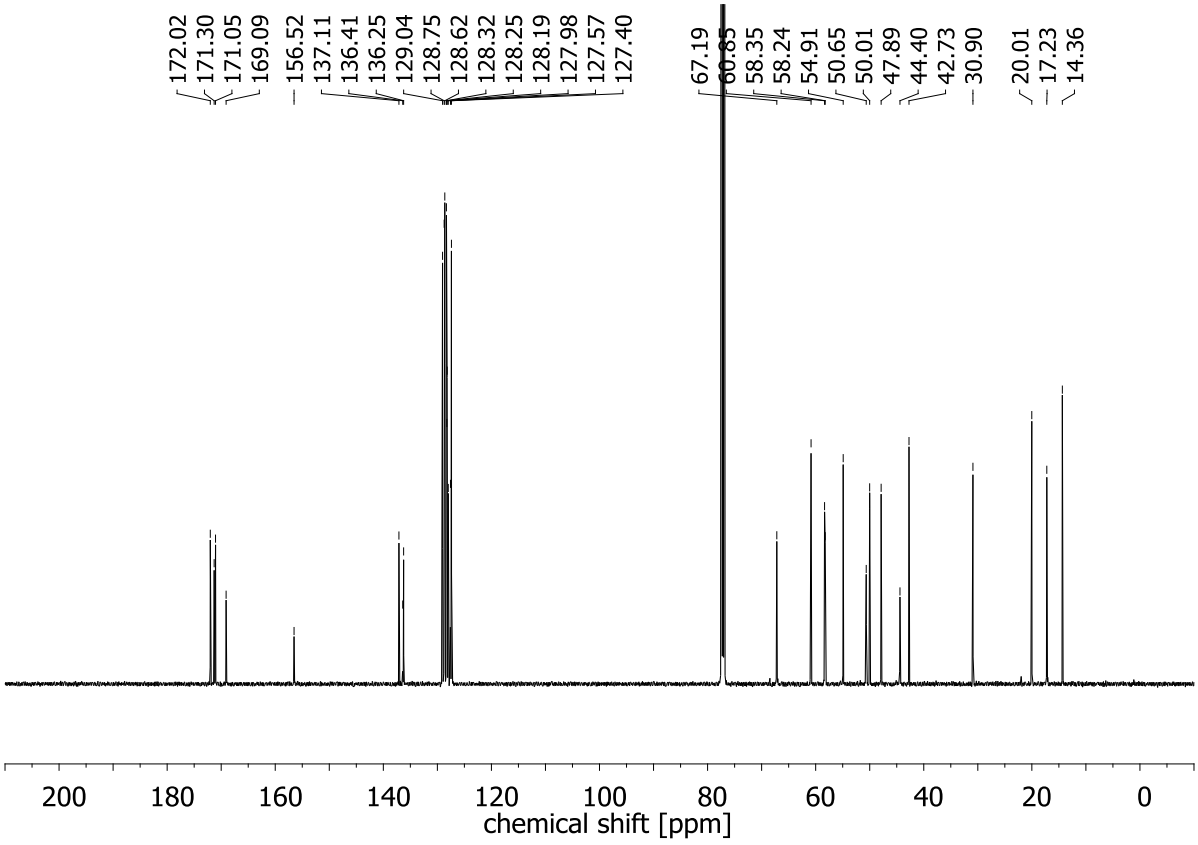
^{1}H NMR spectrum of **37** (500 MHz, $CDCl_3$)



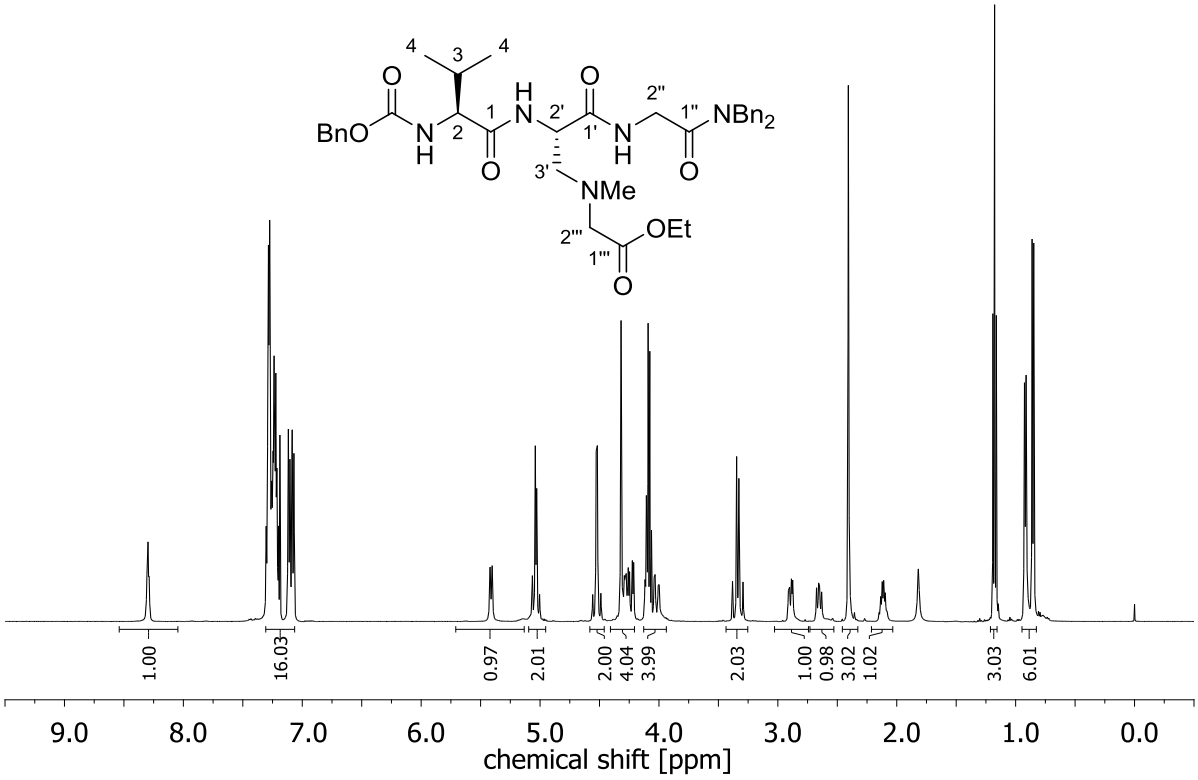
^{13}C NMR spectrum of **37** (126 MHz, $CDCl_3$)



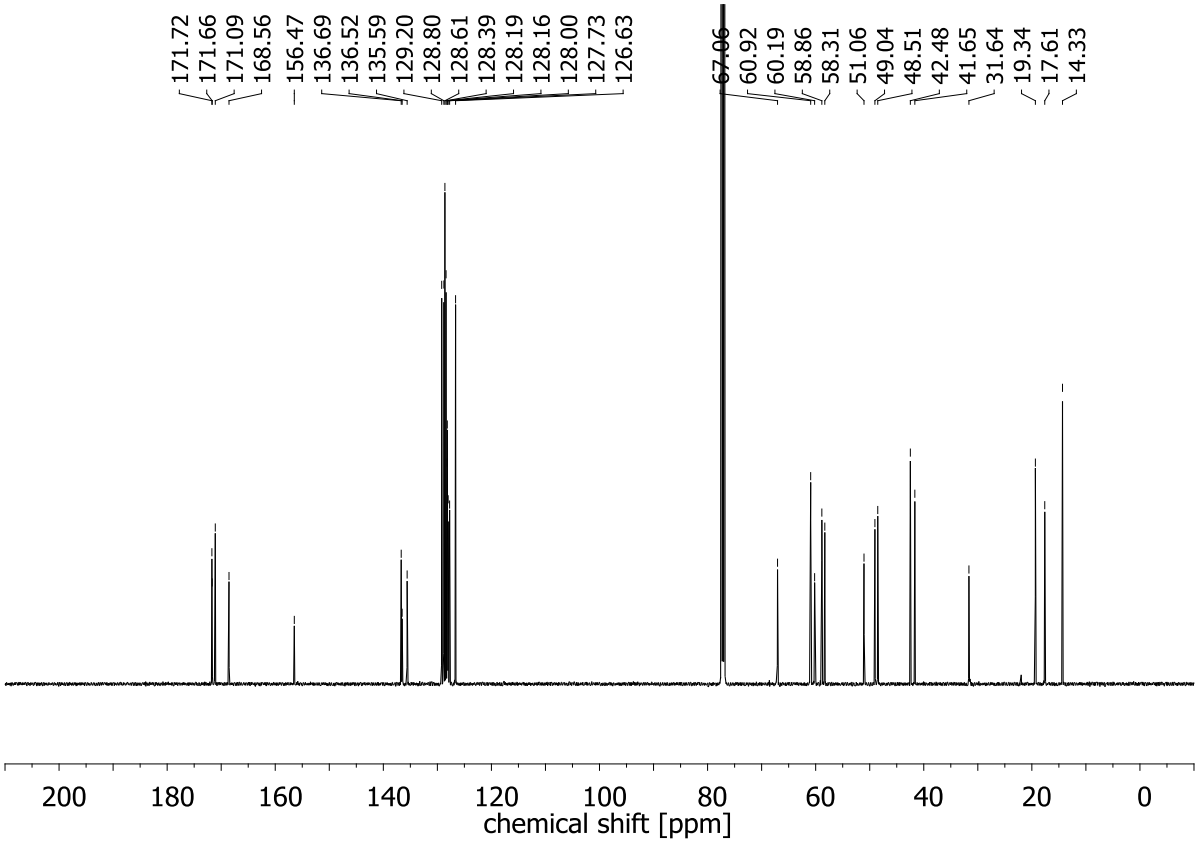
^{1}H NMR spectrum of **38** (500 MHz, $CDCl_3$)



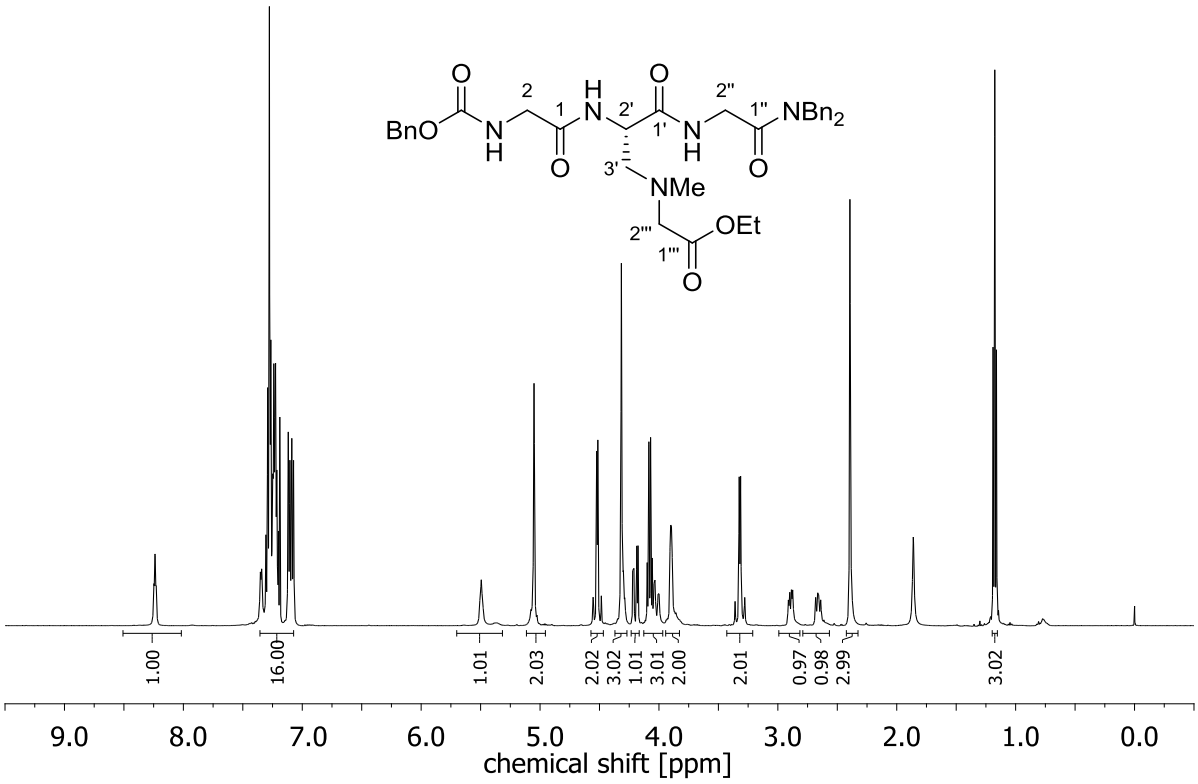
^{13}C NMR spectrum of **38** (126 MHz, $CDCl_3$)



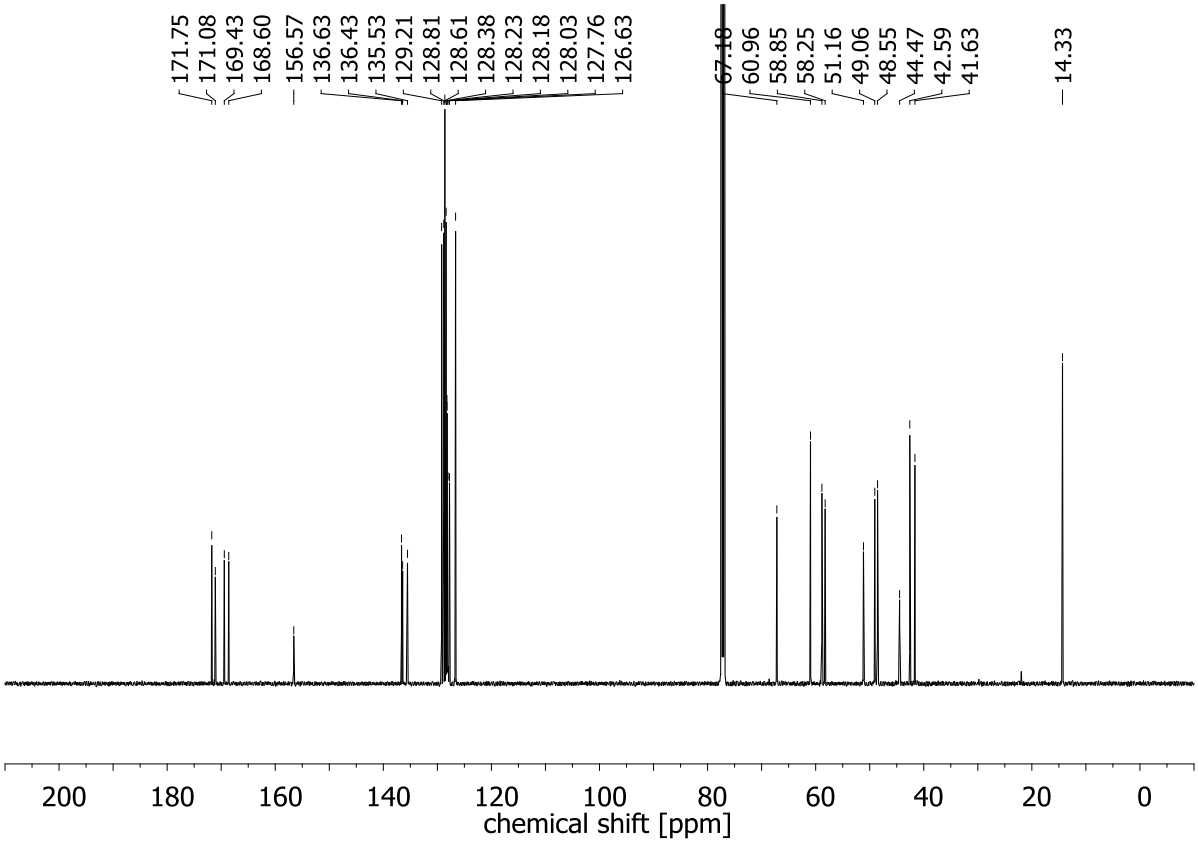
^{1}H NMR spectrum of **39** (500 MHz, $CDCl_3$)



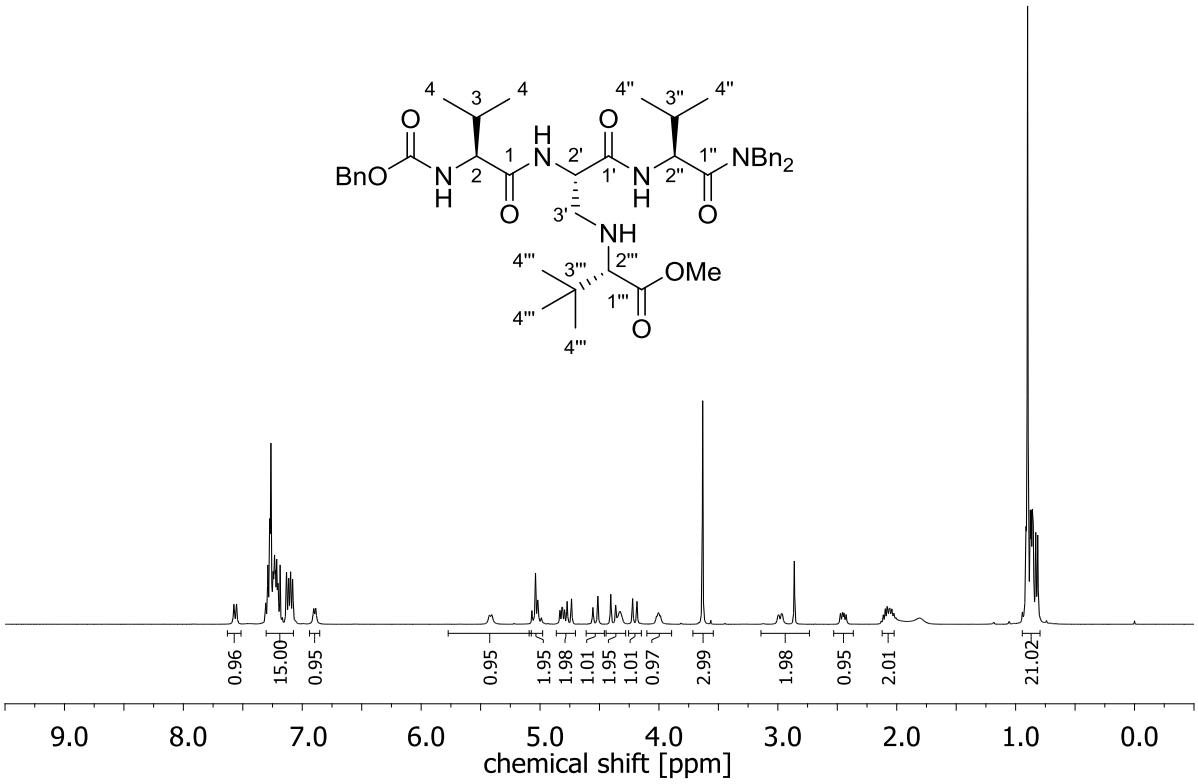
^{13}C NMR spectrum of **39** (126 MHz, $CDCl_3$)



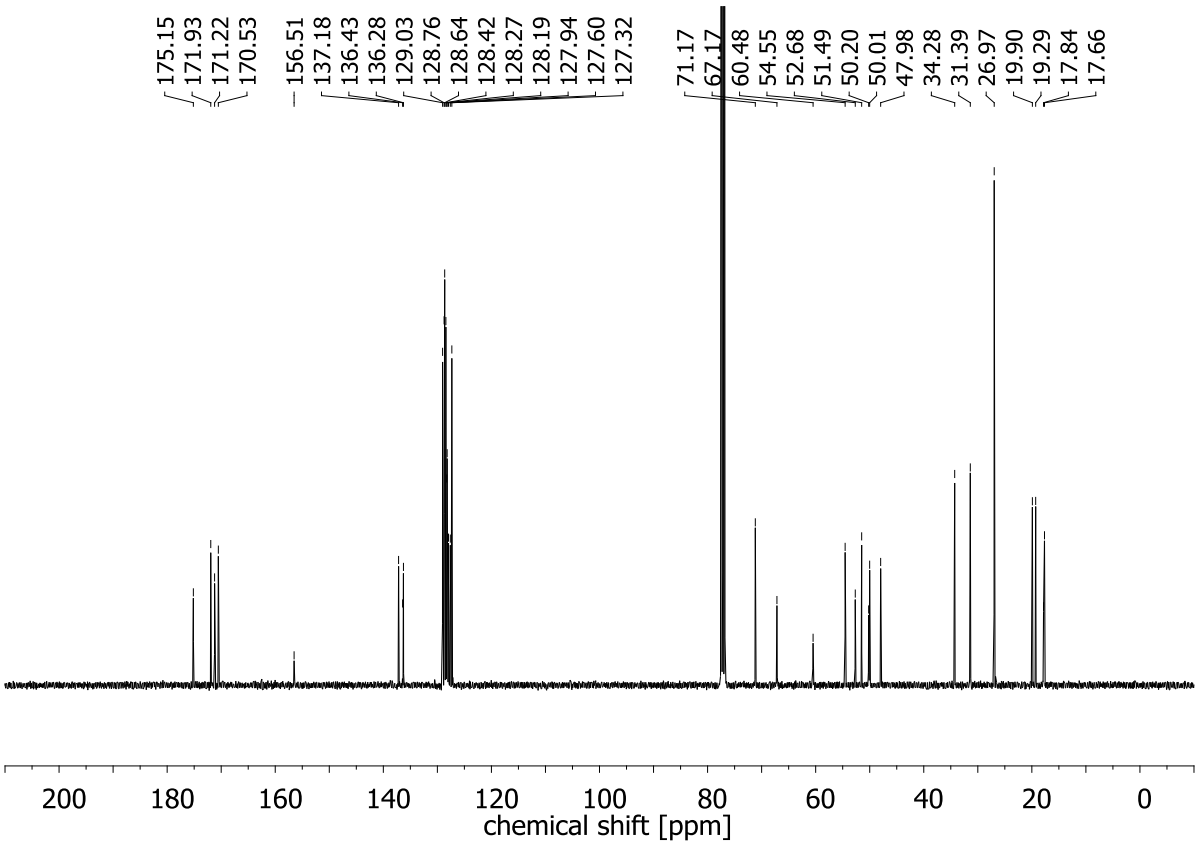
^{1}H NMR spectrum of **40** (500 MHz, $CDCl_3$)



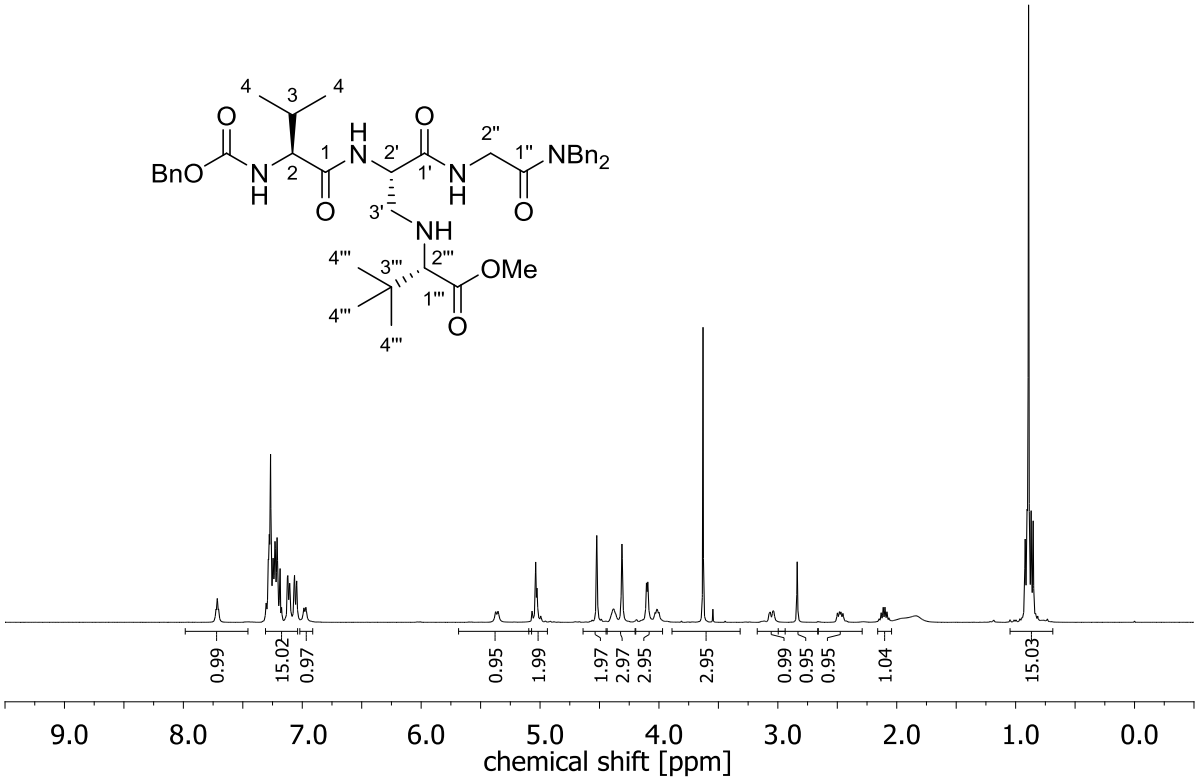
^{13}C NMR spectrum of **40** (126 MHz, $CDCl_3$)



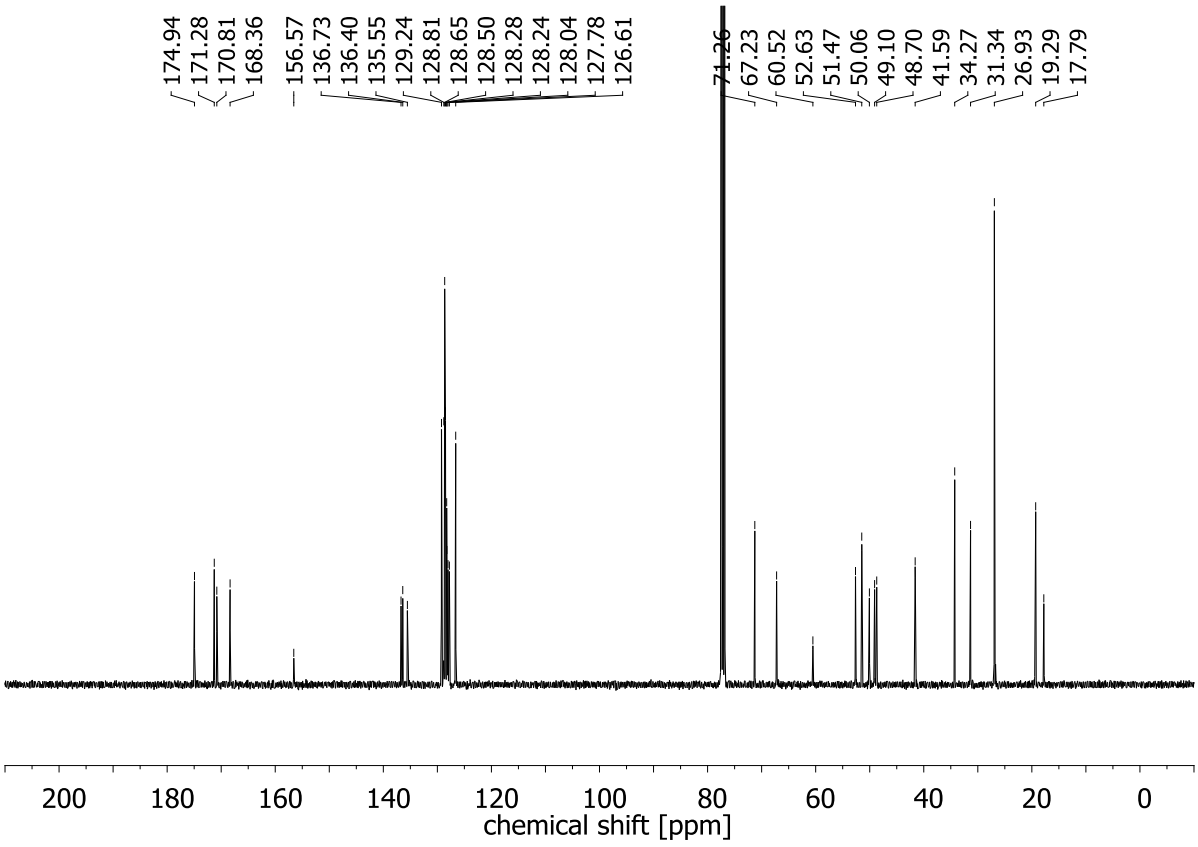
^{1}H NMR spectrum of **41** (400 MHz, $CDCl_3$)



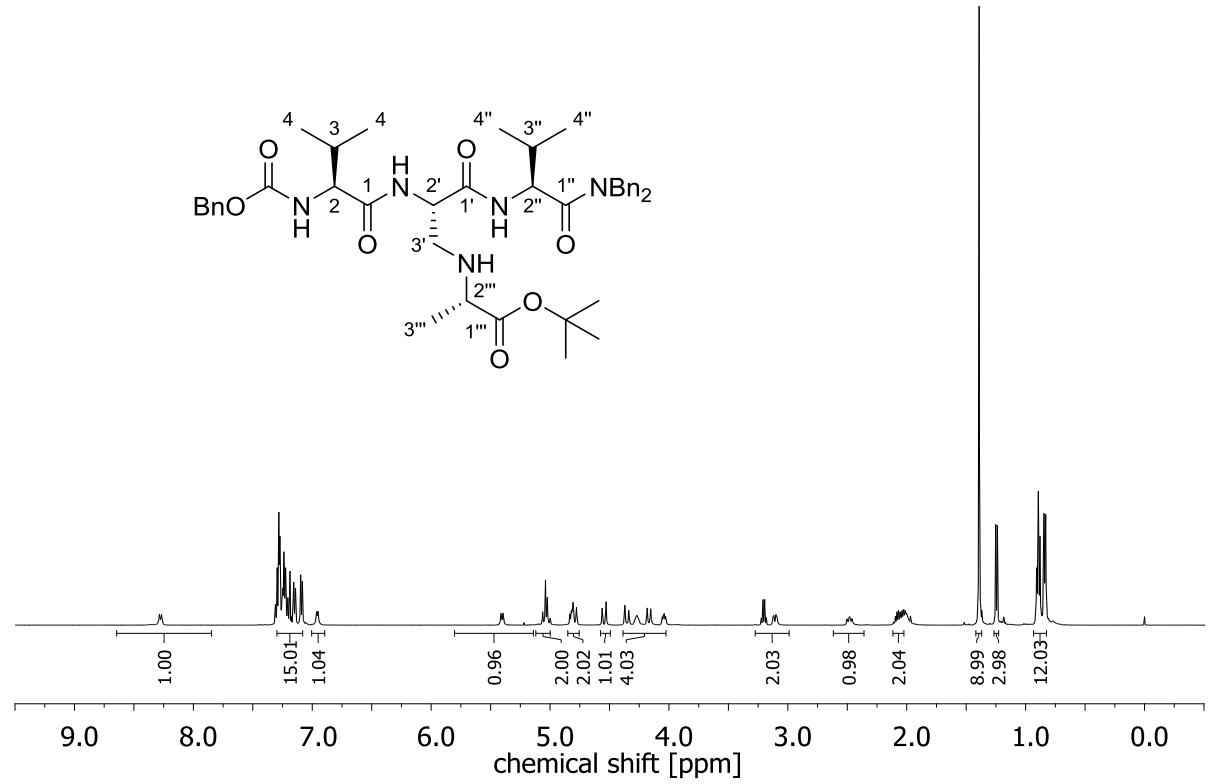
^{13}C NMR spectrum of **41** (101 MHz, $CDCl_3$)



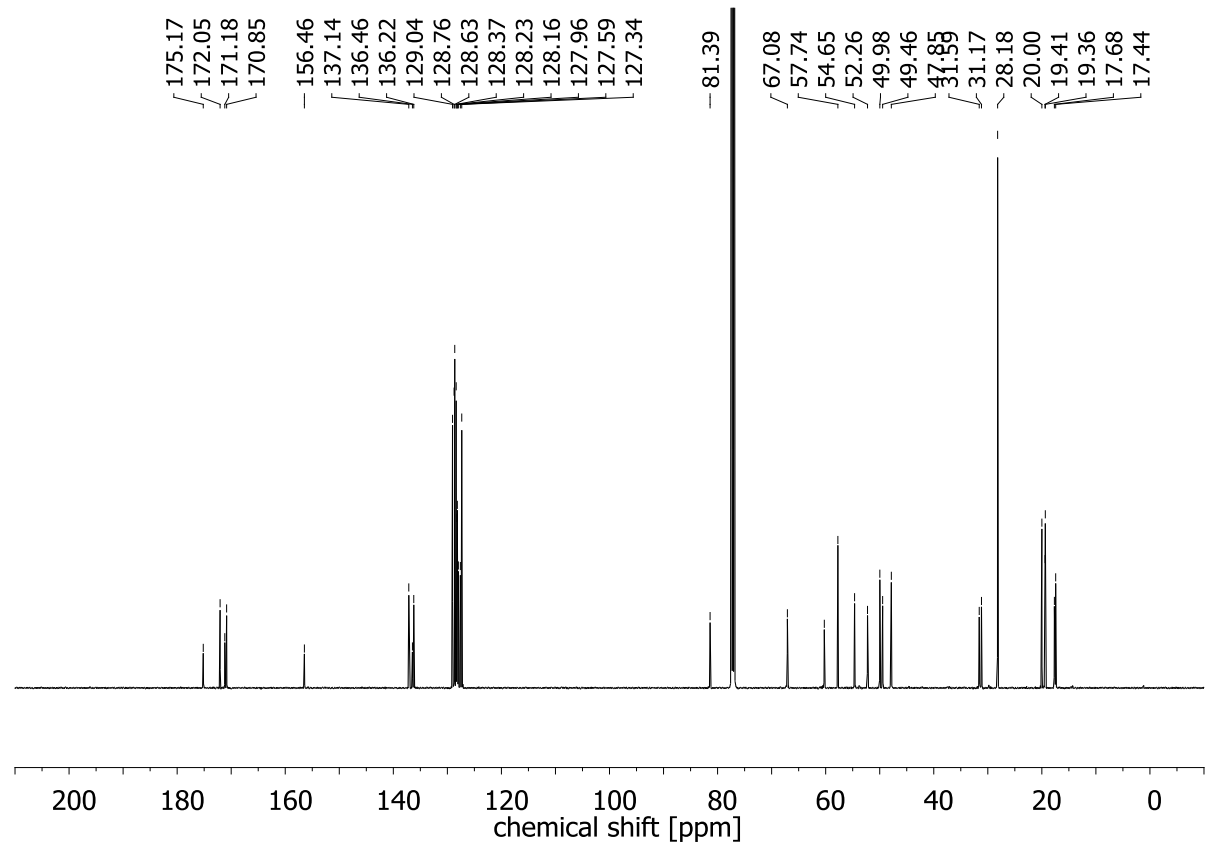
^{1}H NMR spectrum of **42** (400 MHz, $CDCl_3$)



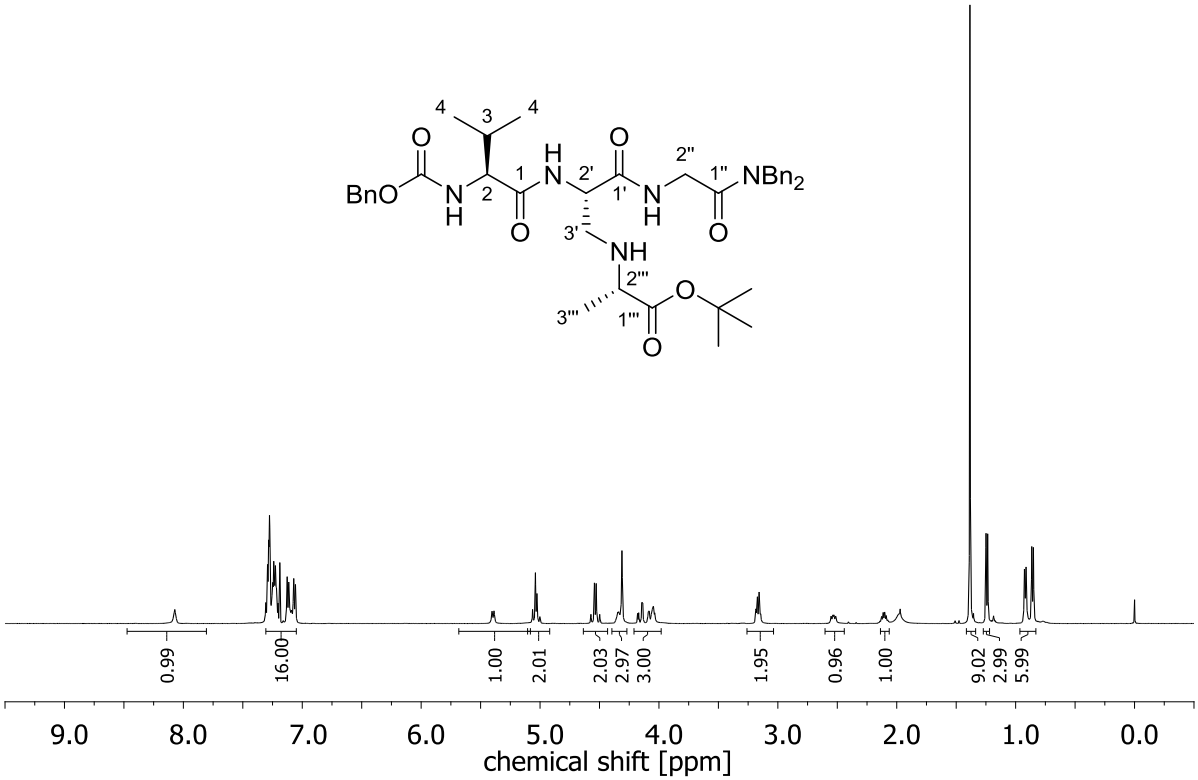
^{13}C NMR spectrum of **42** (101 MHz, $CDCl_3$)



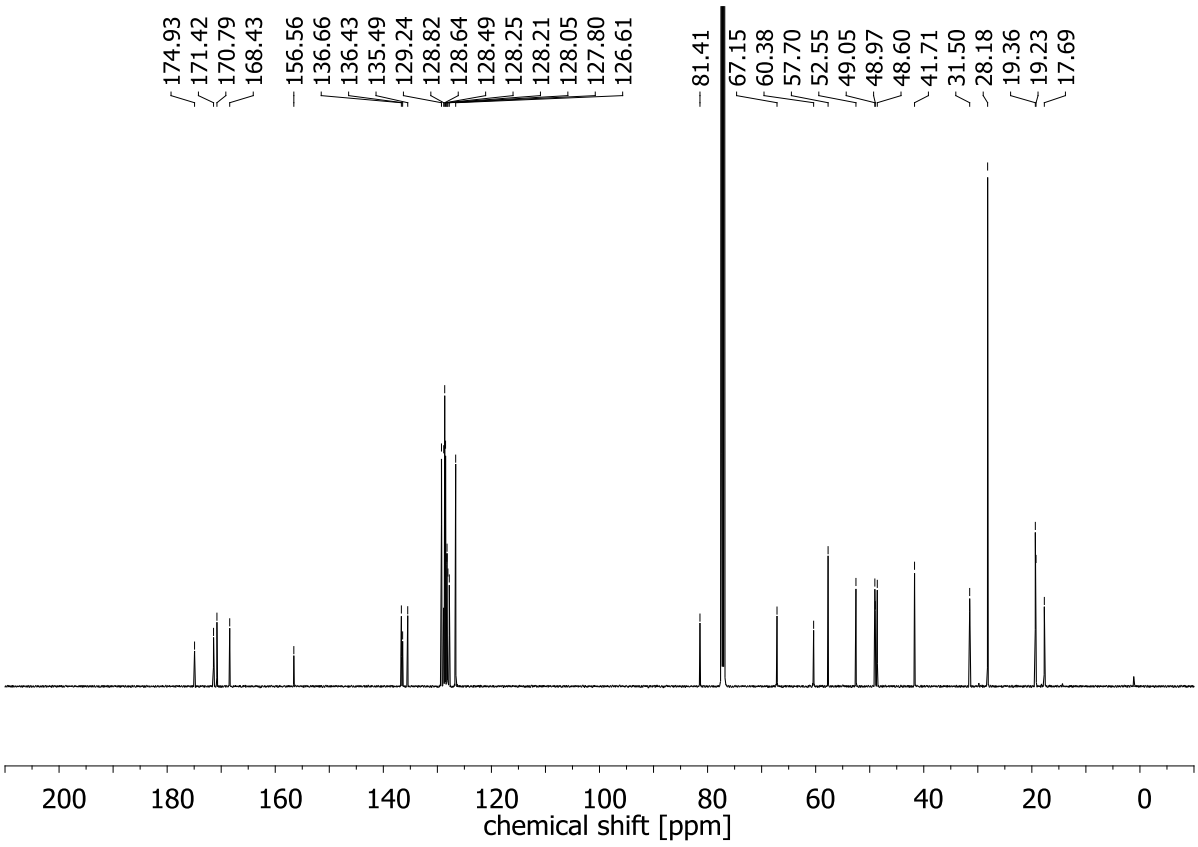
^{1}H NMR spectrum of **43** (500 MHz, $CDCl_3$)



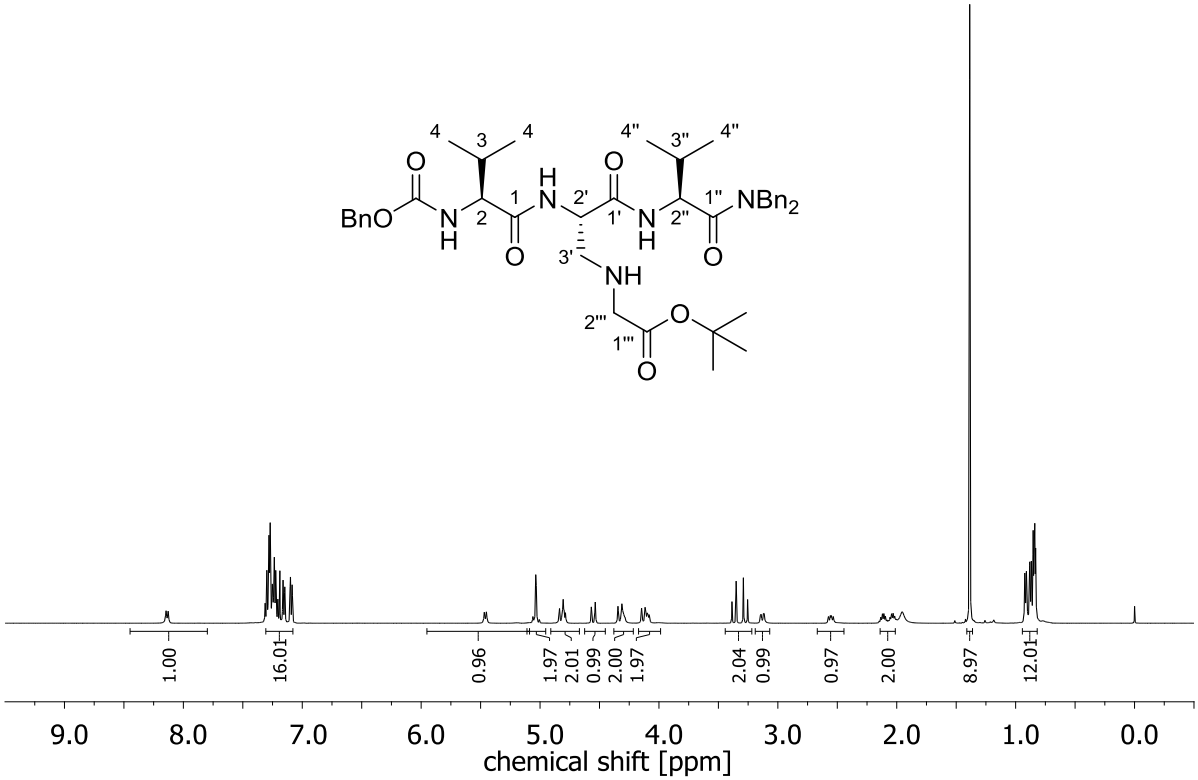
^{13}C NMR spectrum of **43** (126 MHz, $CDCl_3$)



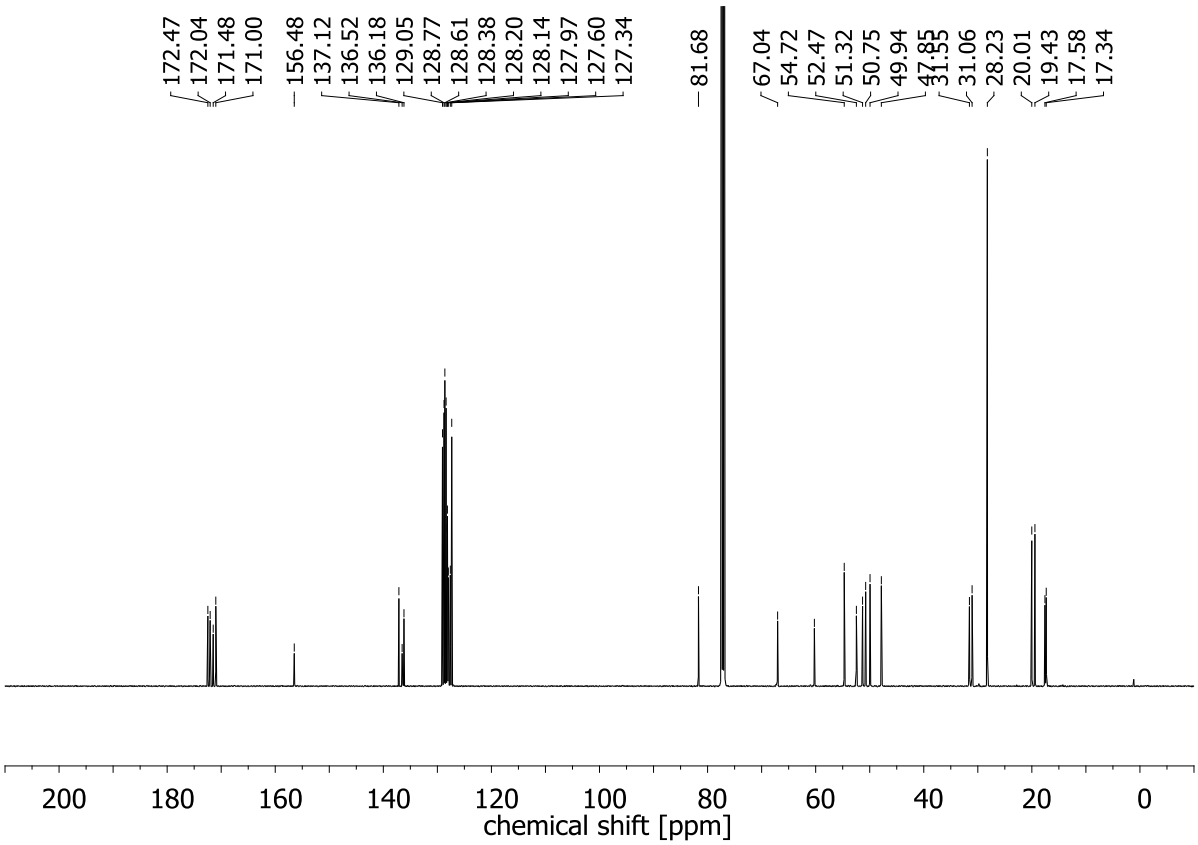
^{1}H NMR spectrum of **44** (500 MHz, $CDCl_3$)



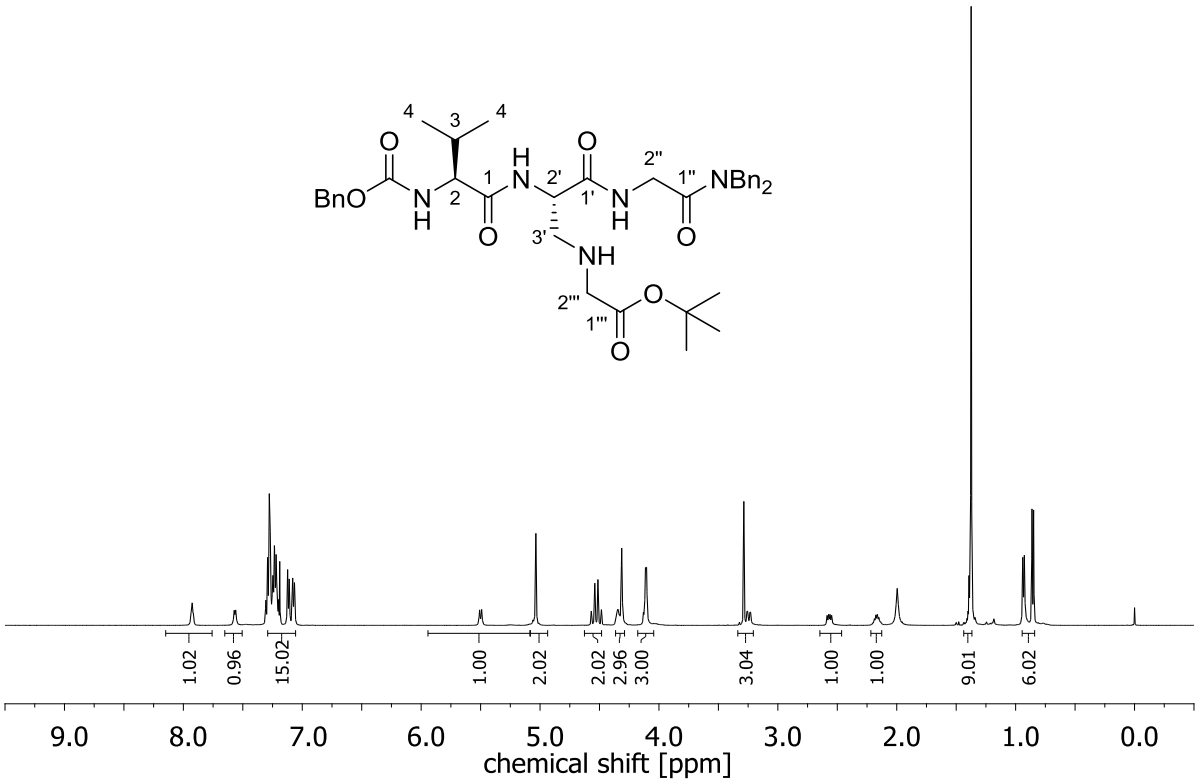
^{13}C NMR spectrum of **44** (126 MHz, $CDCl_3$)



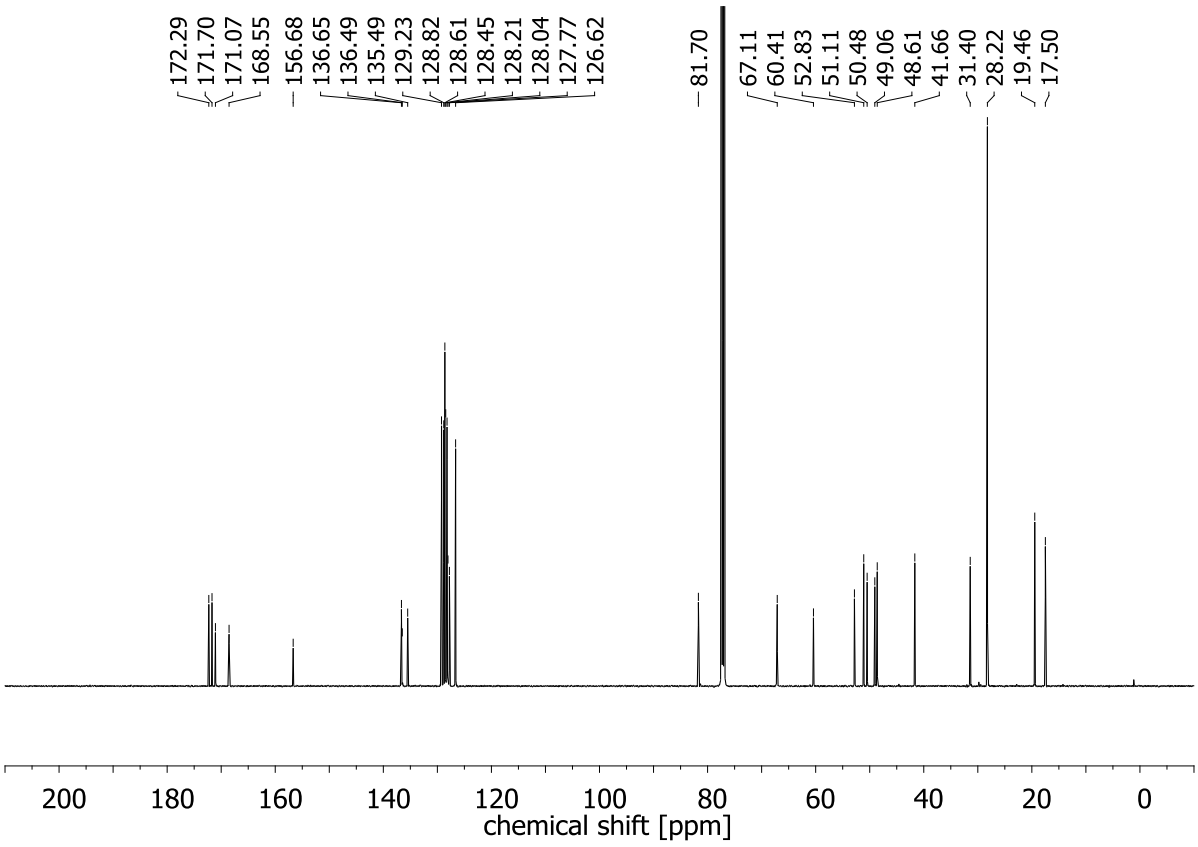
^{1}H NMR spectrum of **45** (500 MHz, $CDCl_3$)



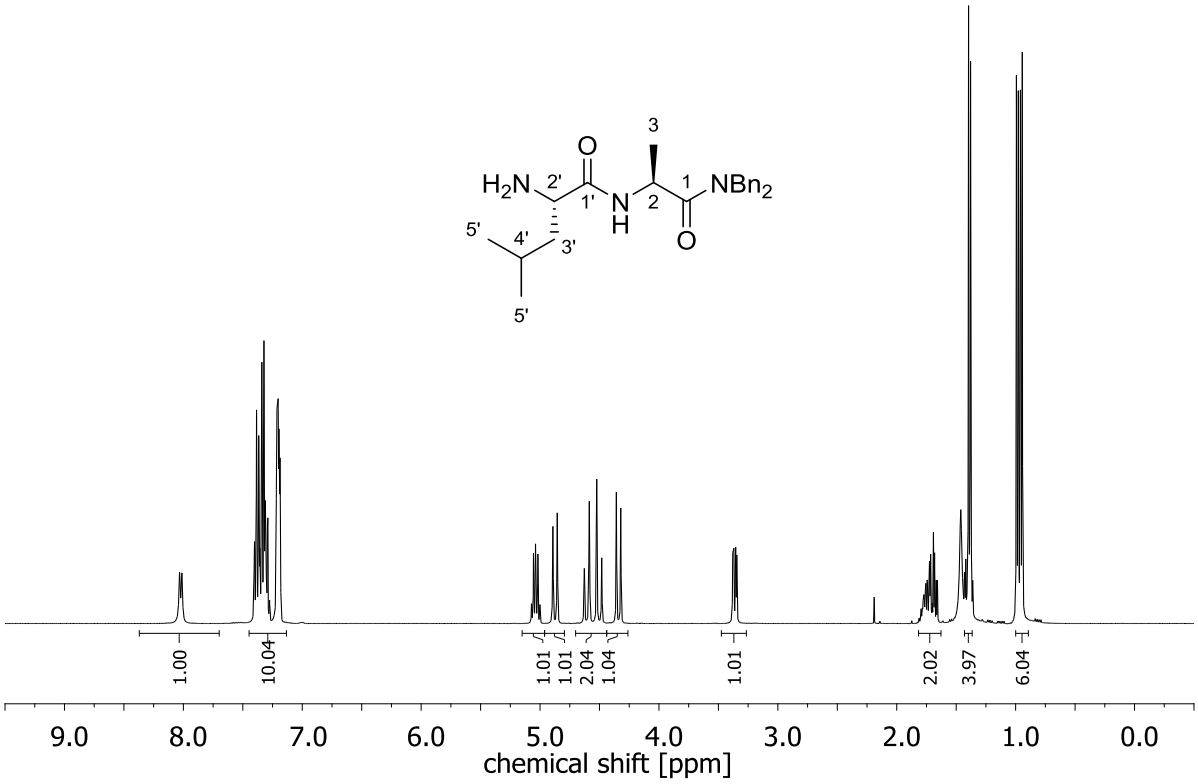
^{13}C NMR spectrum of **45** (126 MHz, $CDCl_3$)



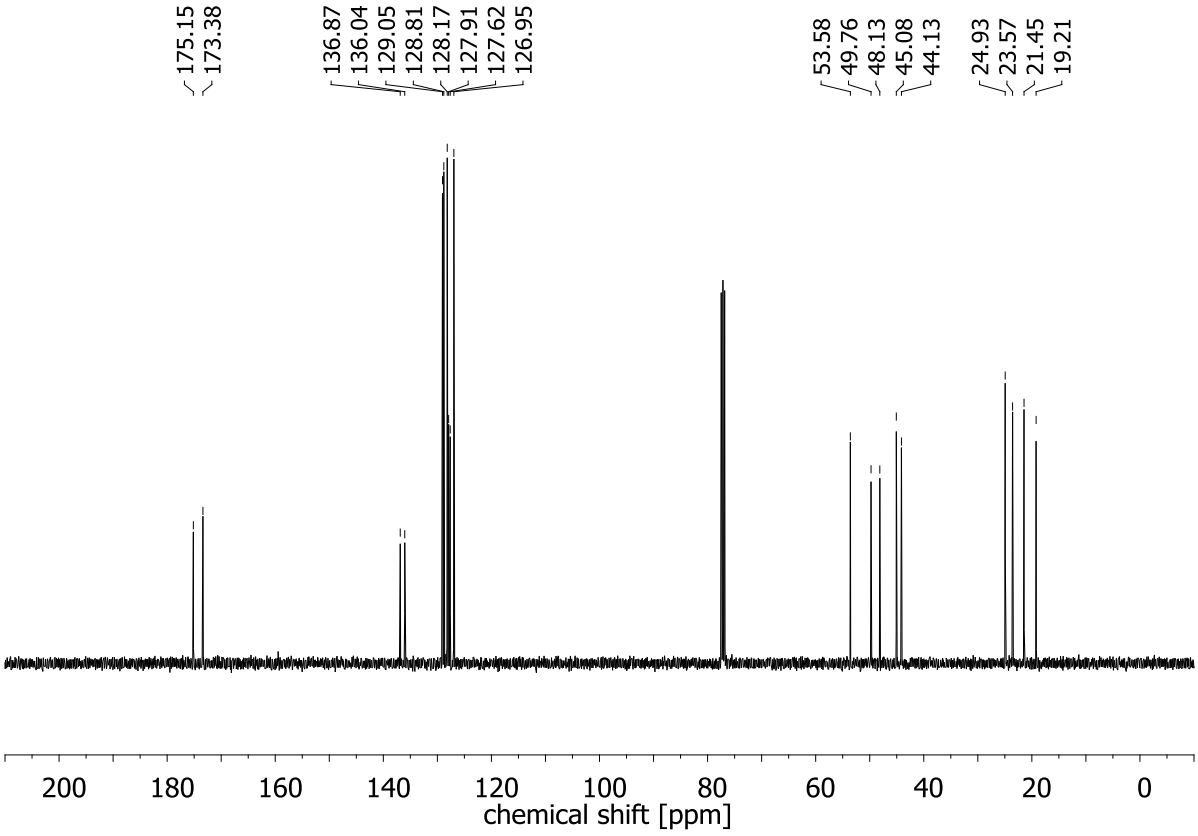
^{1}H NMR spectrum of **46** (500 MHz, $CDCl_3$)



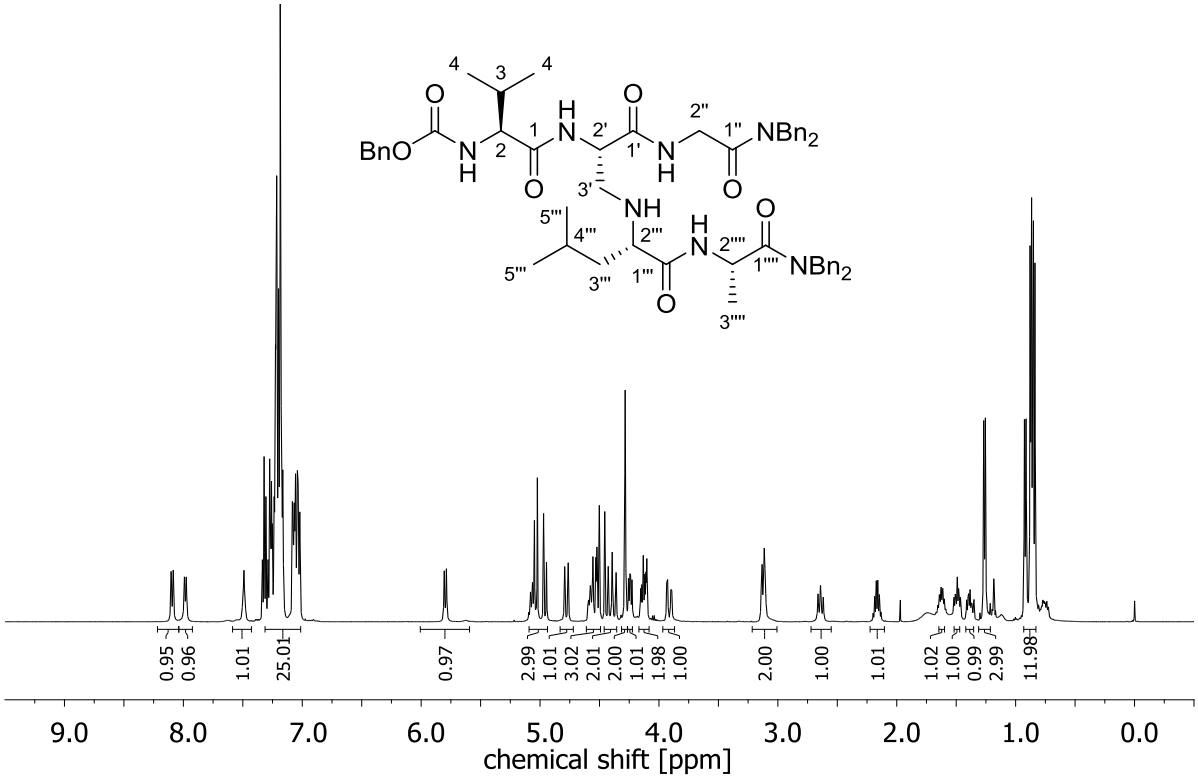
^{13}C NMR spectrum of **46** (126 MHz, $CDCl_3$)



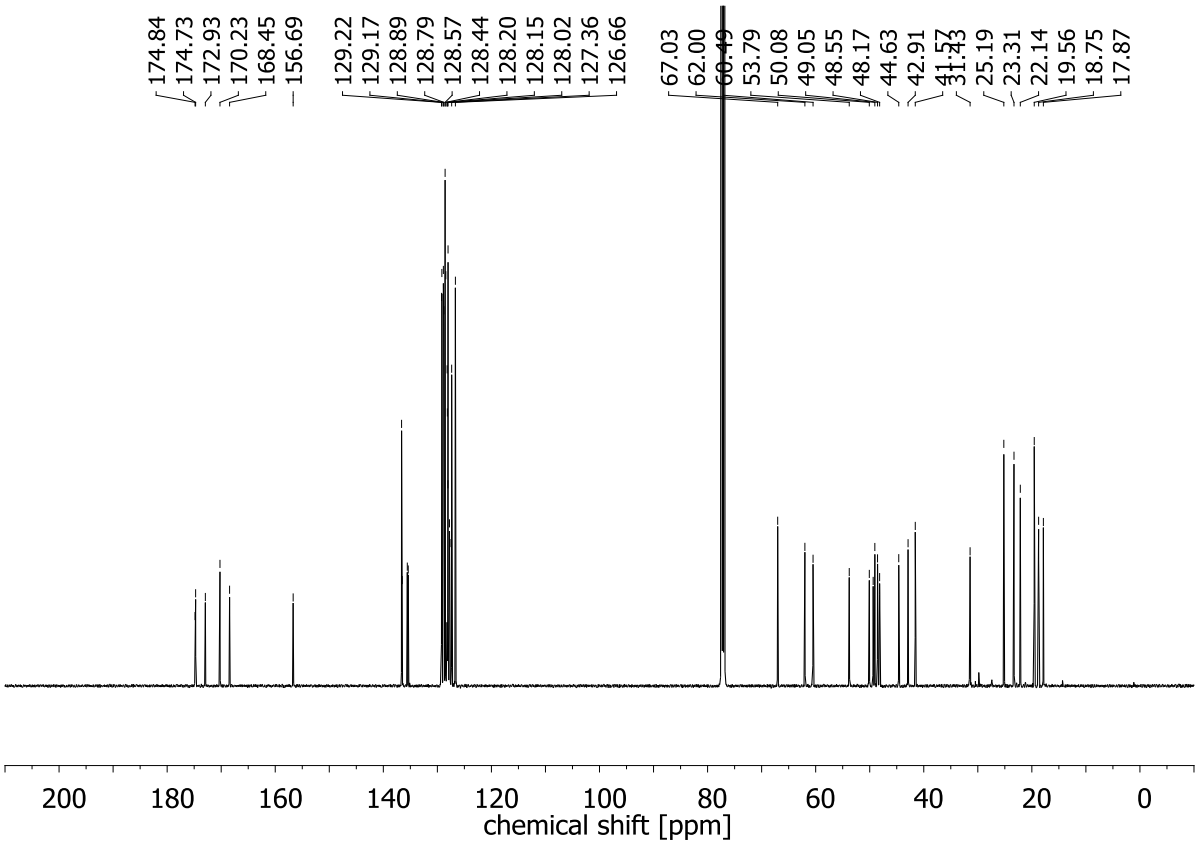
^{1}H NMR spectrum of **47** (400 MHz, $CDCl_3$)



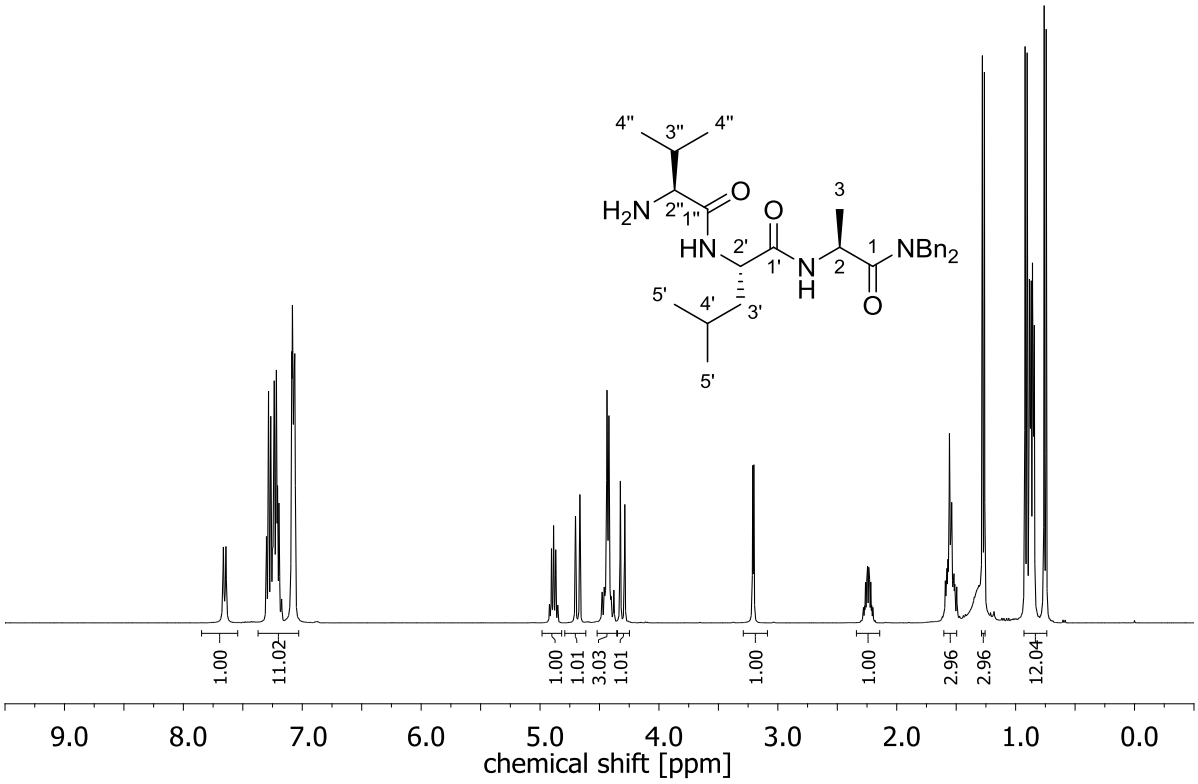
^{13}C NMR spectrum of **47** (101 MHz, $CDCl_3$)



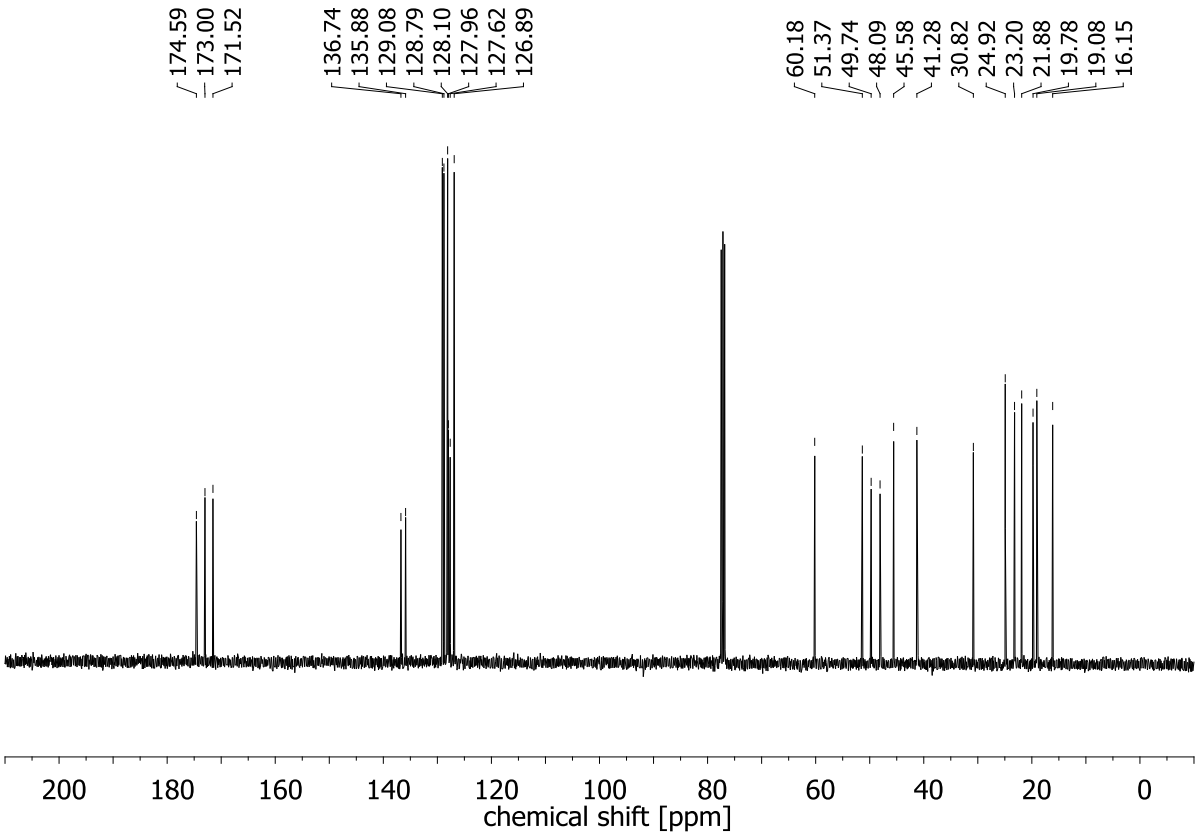
^{1}H NMR spectrum of **48** (500 MHz, $CDCl_3$)



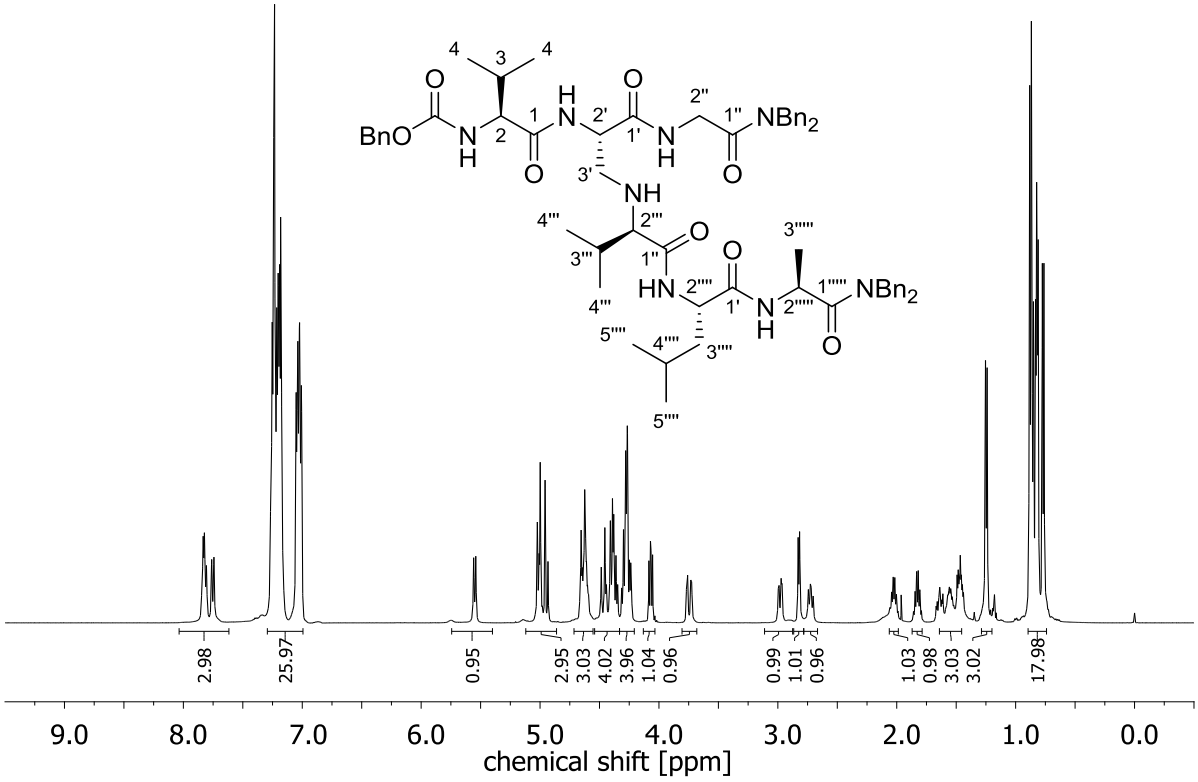
^{13}C NMR spectrum of **48** (126 MHz, $CDCl_3$)



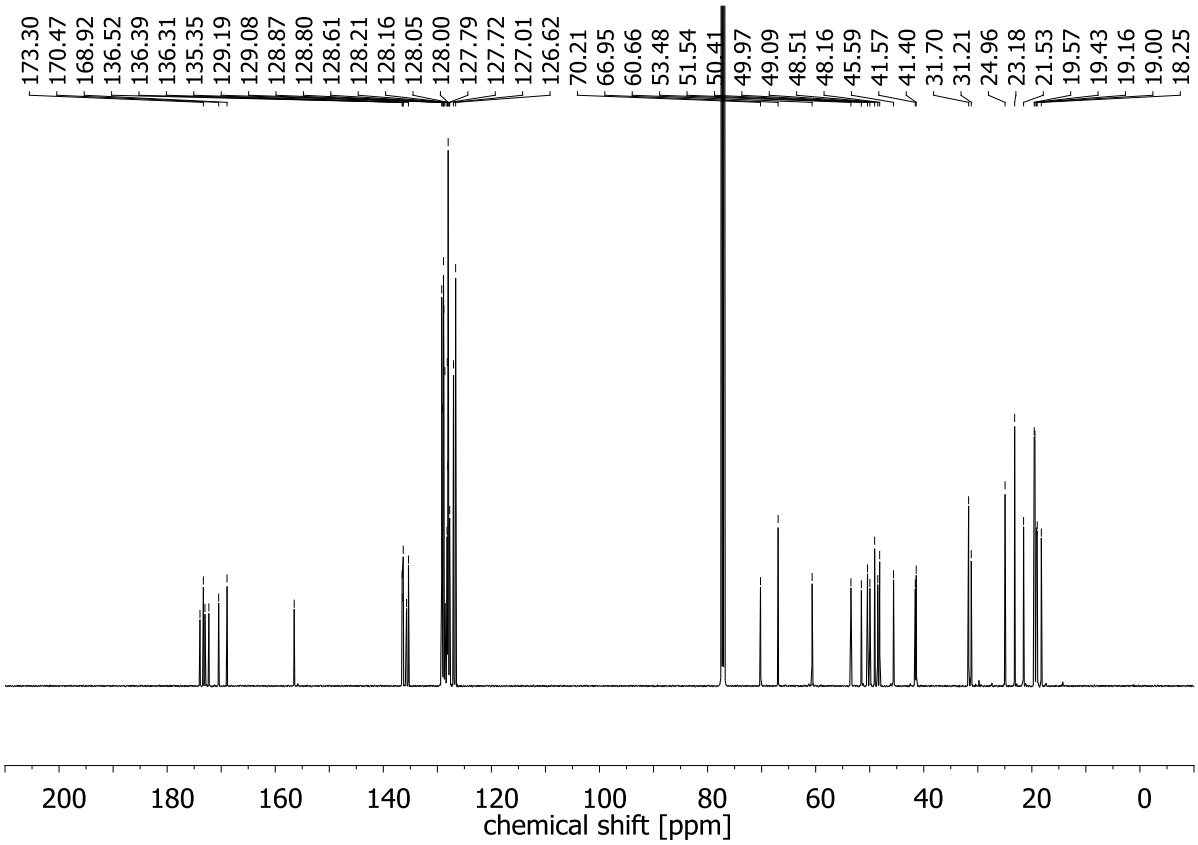
^{1}H NMR spectrum of **49** (400 MHz, $CDCl_3$)



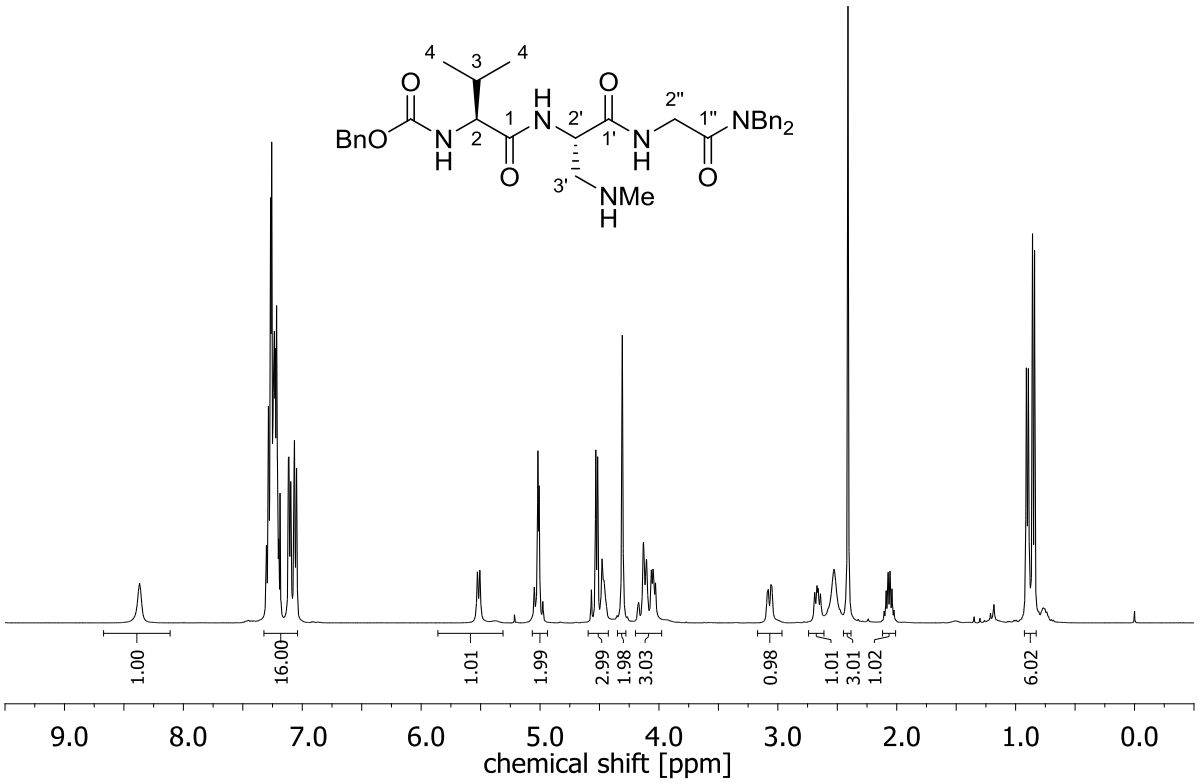
^{13}C NMR spectrum of **49** (101 MHz, $CDCl_3$)



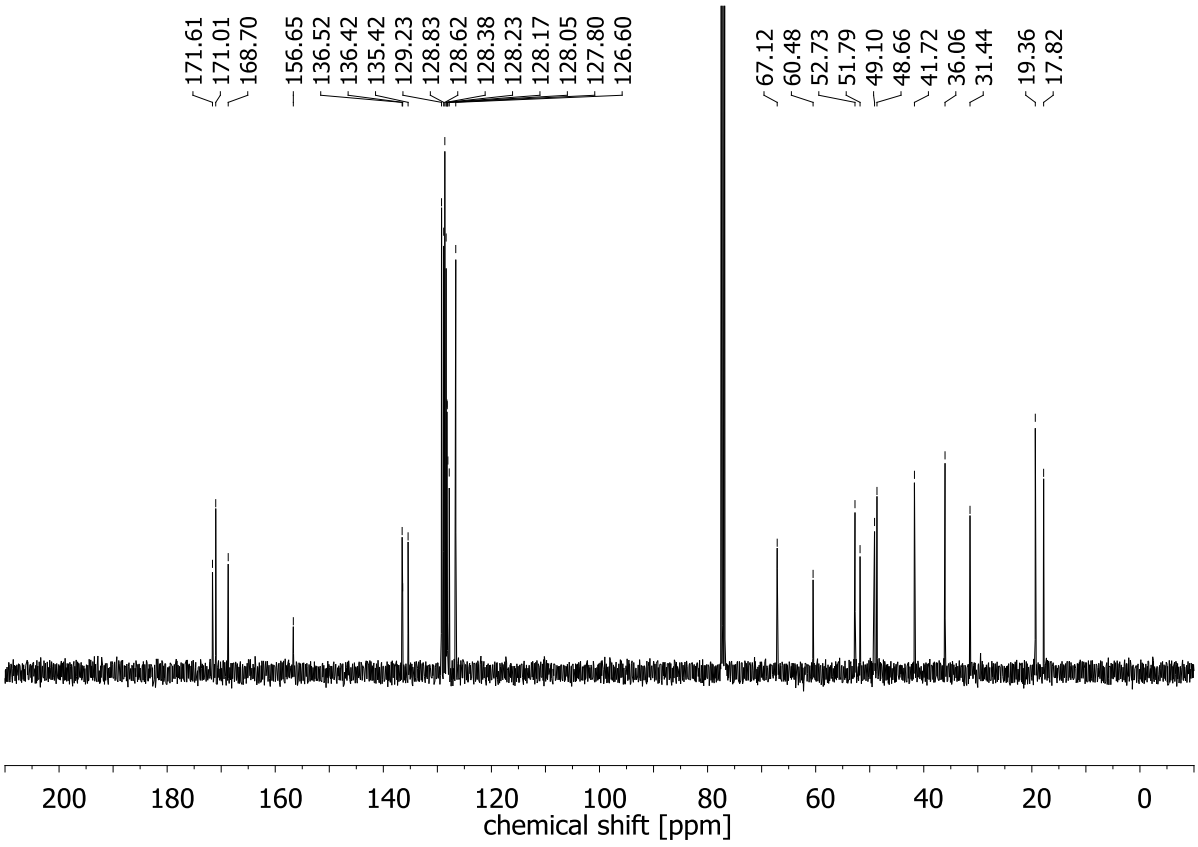
^{1}H NMR spectrum of **50** (500 MHz, $CDCl_3$)



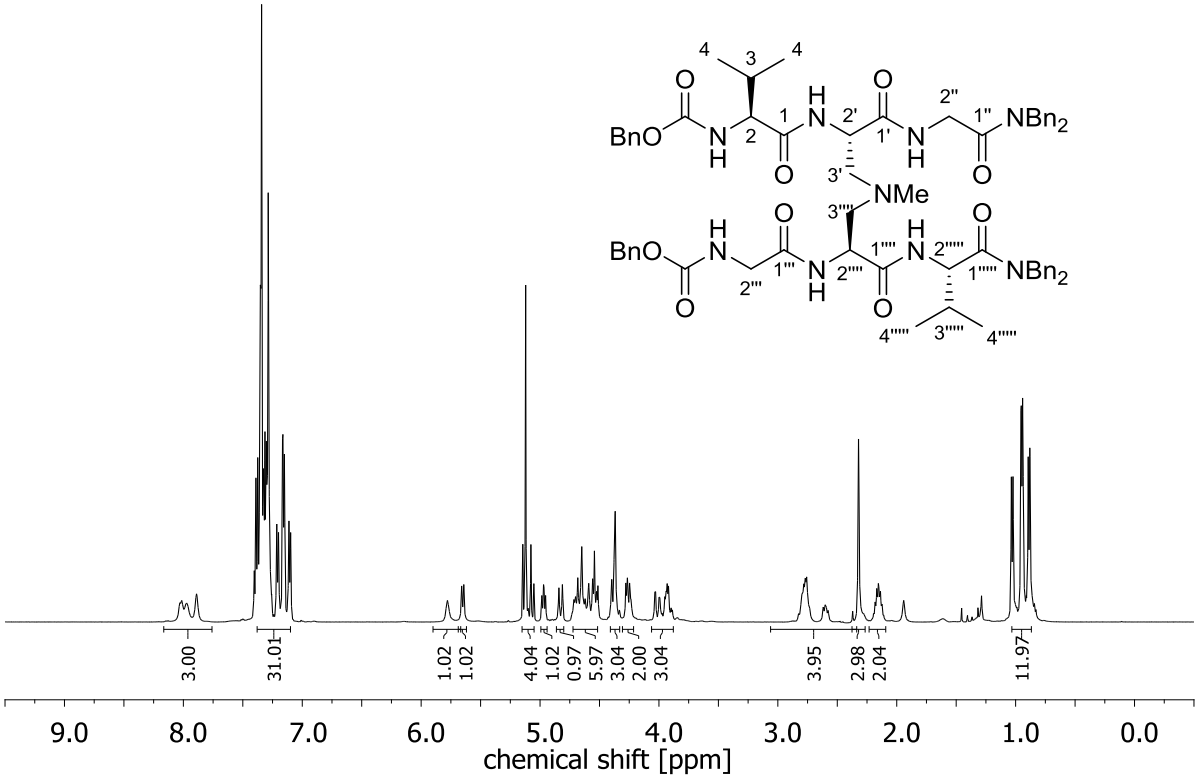
^{13}C NMR spectrum of **50** (126 MHz, $CDCl_3$)



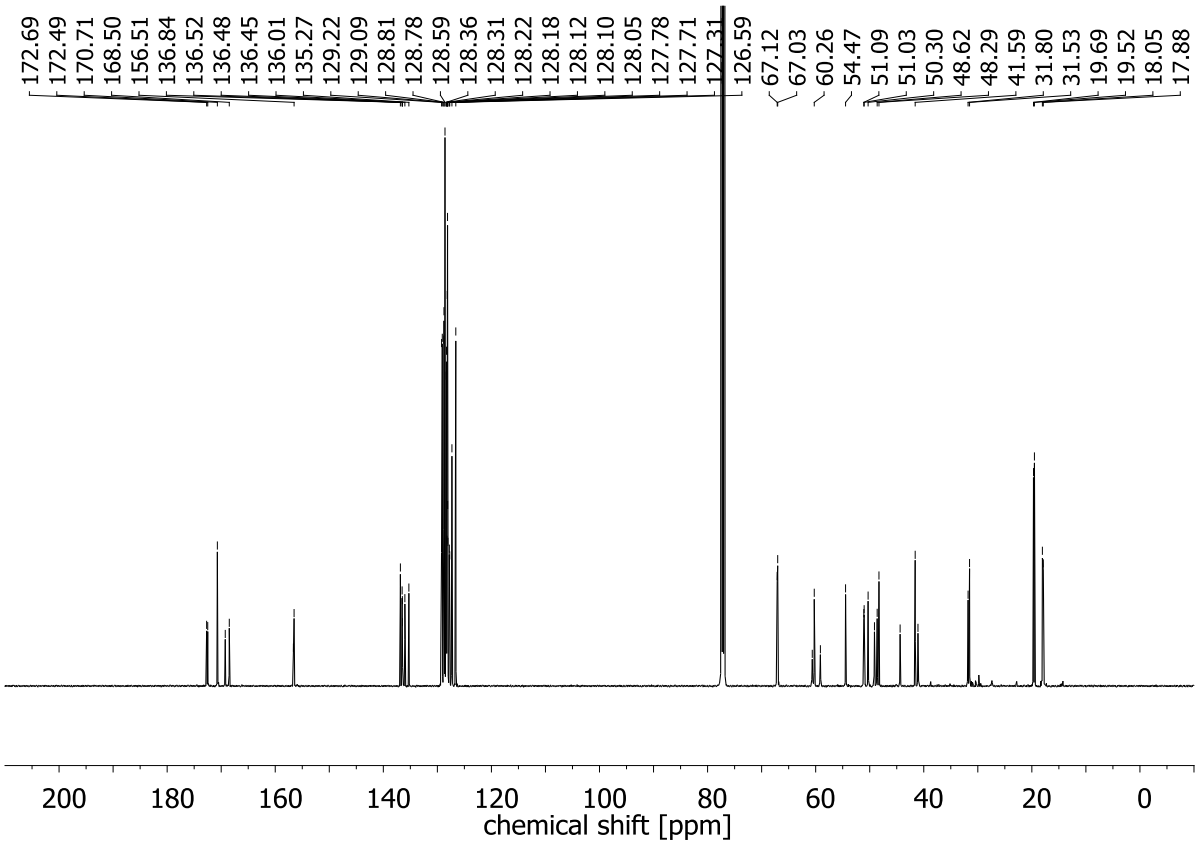
^{1}H NMR spectrum of **51** (400 MHz, $CDCl_3$)



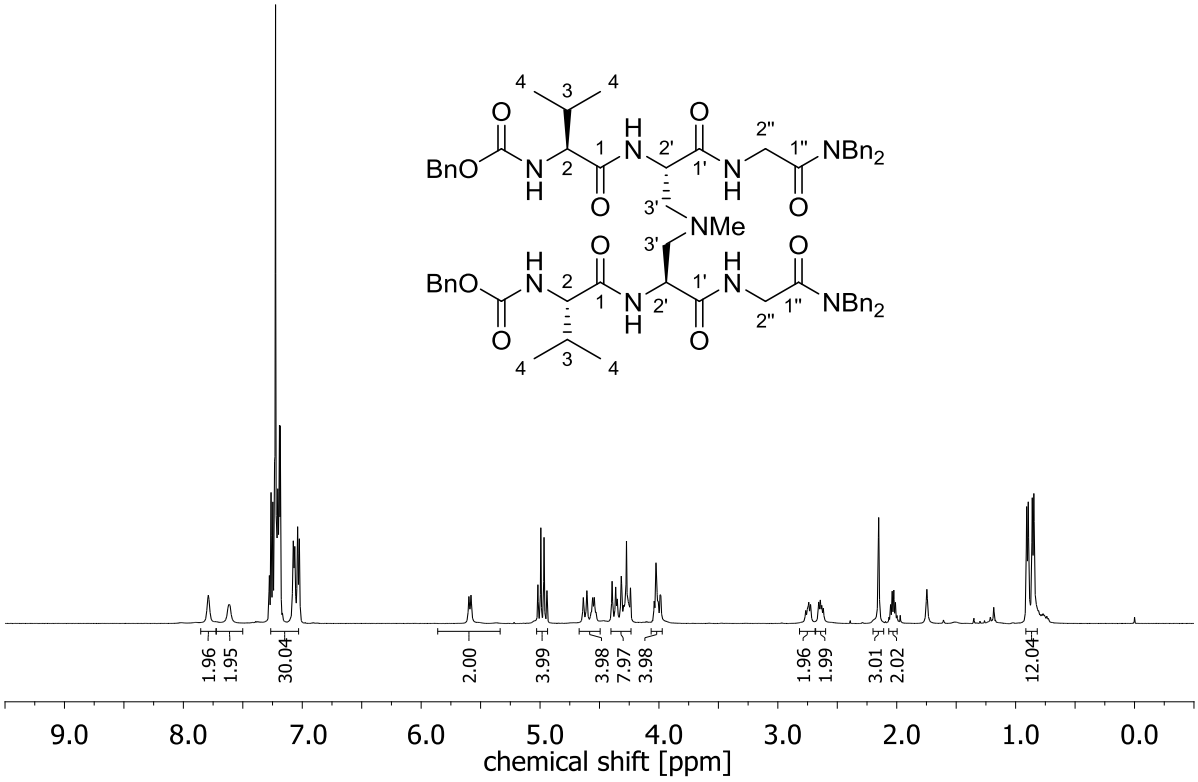
^{13}C NMR spectrum of **51** (101 MHz, $CDCl_3$)



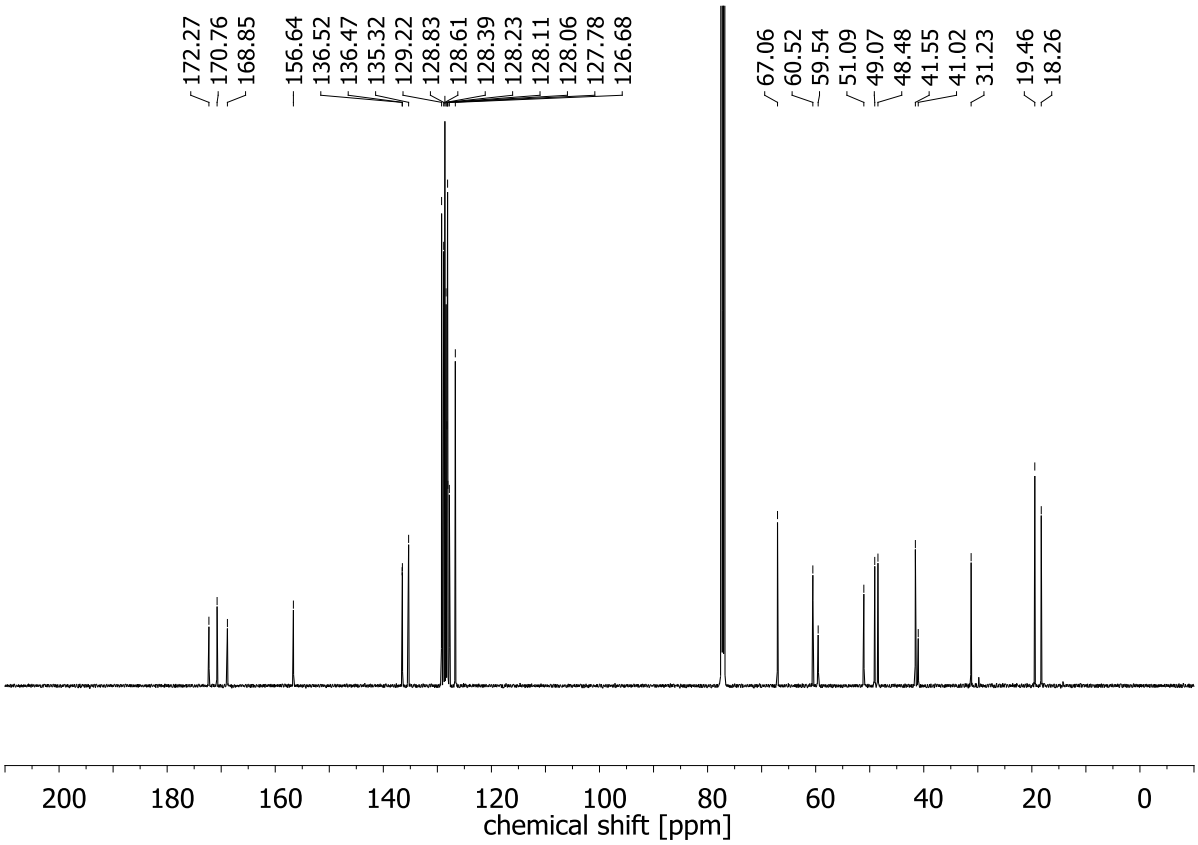
^{1}H NMR spectrum of **52** (500 MHz, $CDCl_3$)



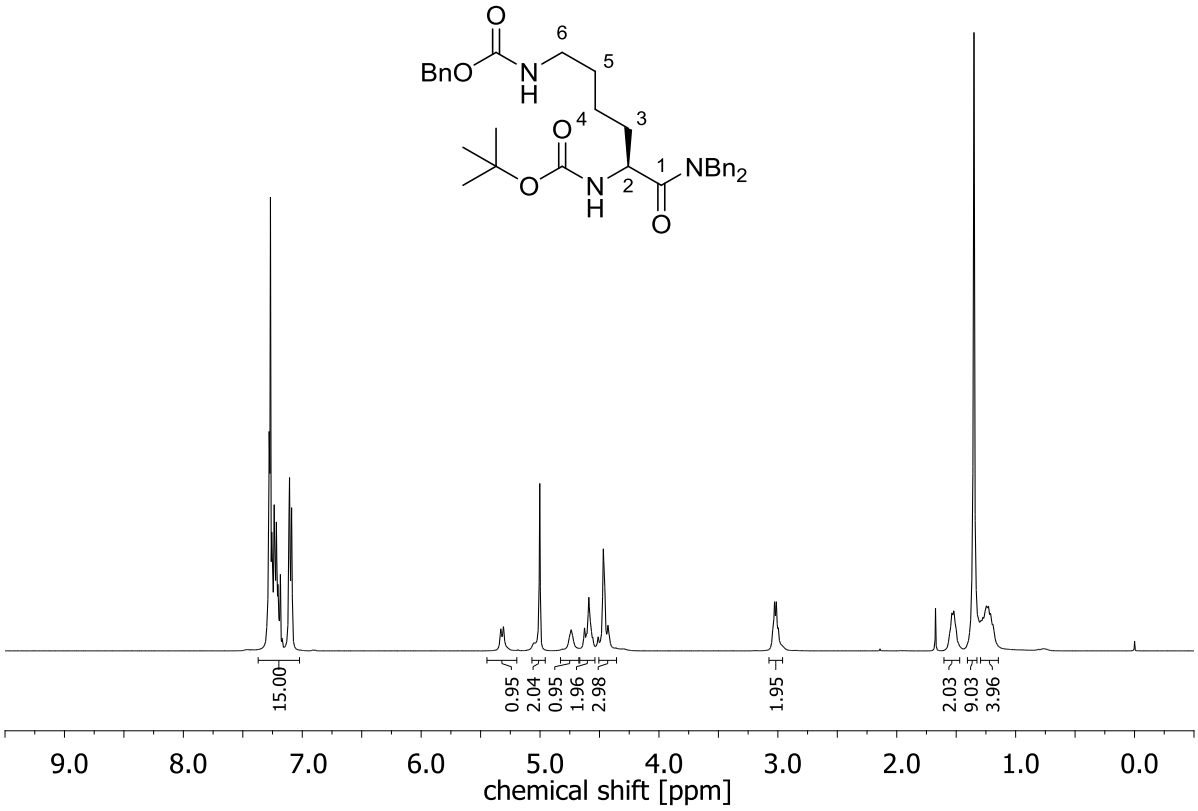
^{13}C NMR spectrum of **52** (126 MHz, $CDCl_3$)



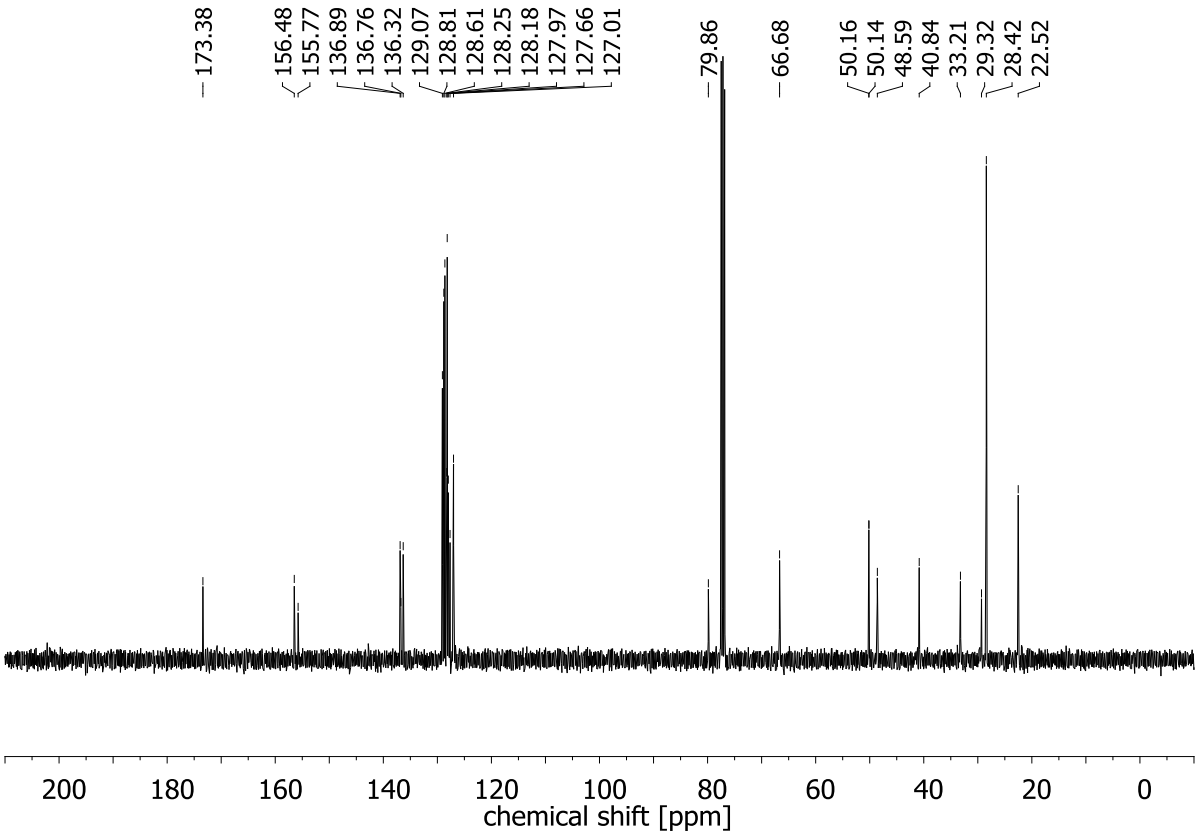
^{1}H NMR spectrum of **53** (500 MHz, $CDCl_3$)



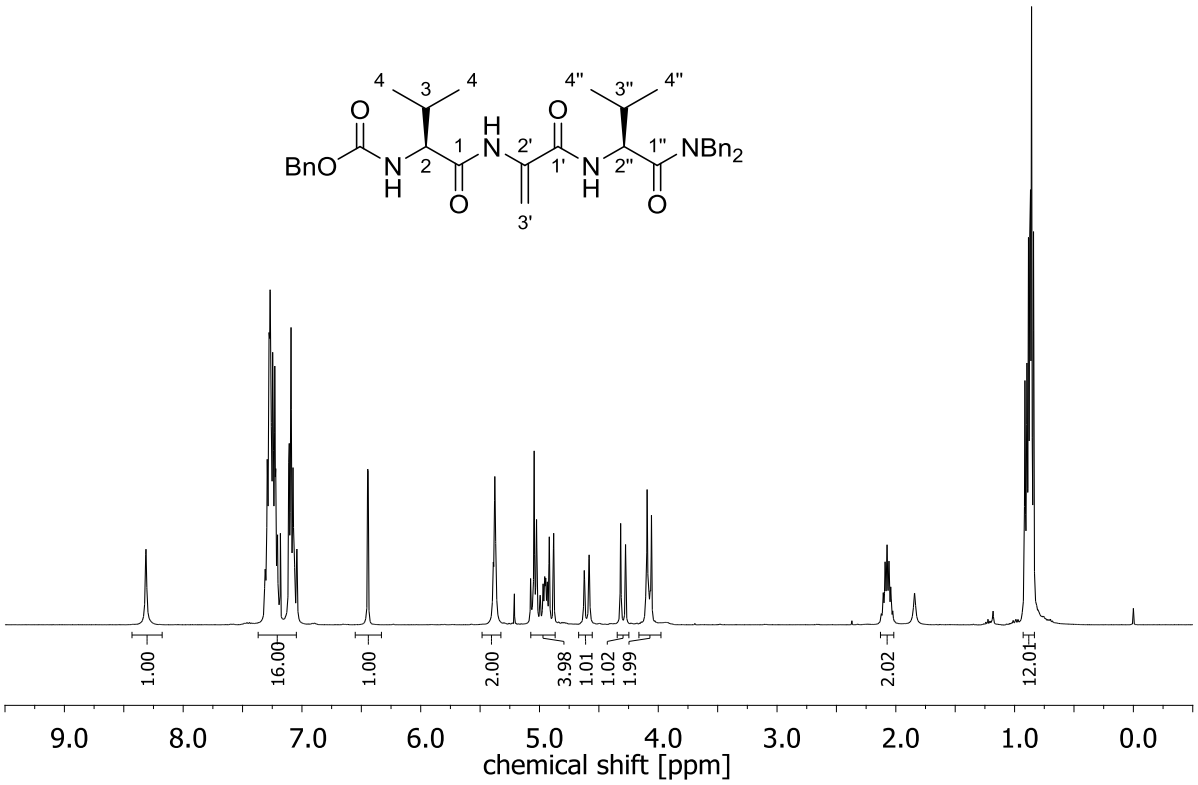
^{13}C NMR spectrum of **53** (126 MHz, $CDCl_3$)



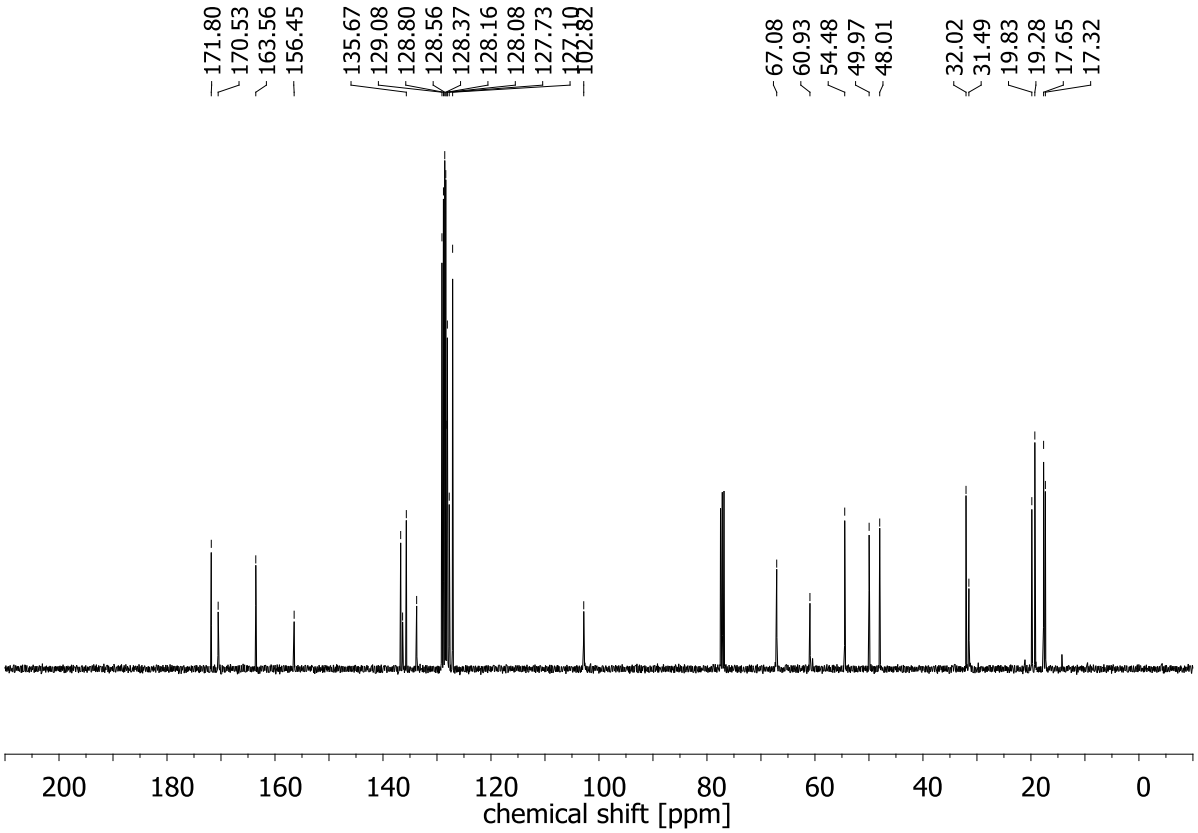
^{1}H NMR spectrum of **S1** (400 MHz, $CDCl_3$)



^{13}C NMR spectrum of **S1** (101 MHz, $CDCl_3$)



^{1}H NMR spectrum of **S2** (400 MHz, $CDCl_3$)



^{13}C NMR spectrum of **S2** (101 MHz, $CDCl_3$)

5. References

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