Electronic Supplementary Material (ESI) for Organic & Biomolecular Chemistry.

Supporting Information

Structural and Morphological Diversity of Self-assembled Synthetic γ-Amino Acid Containing Peptides

Maruthi Konda[a], Brice Kauffmann,[b] Dnyaneshwar B. Rasale[a] and Apurba K. Das[*a]

[a]*Department of Chemistry, Indian Institute of Technology Indore, Indore 452017, India*

[b]*Université de Bordeaux, UMS3033, Institut Européen de Chimie et Biologie (IECB), 2 rue Escarpit, 33600 Pessac, France*

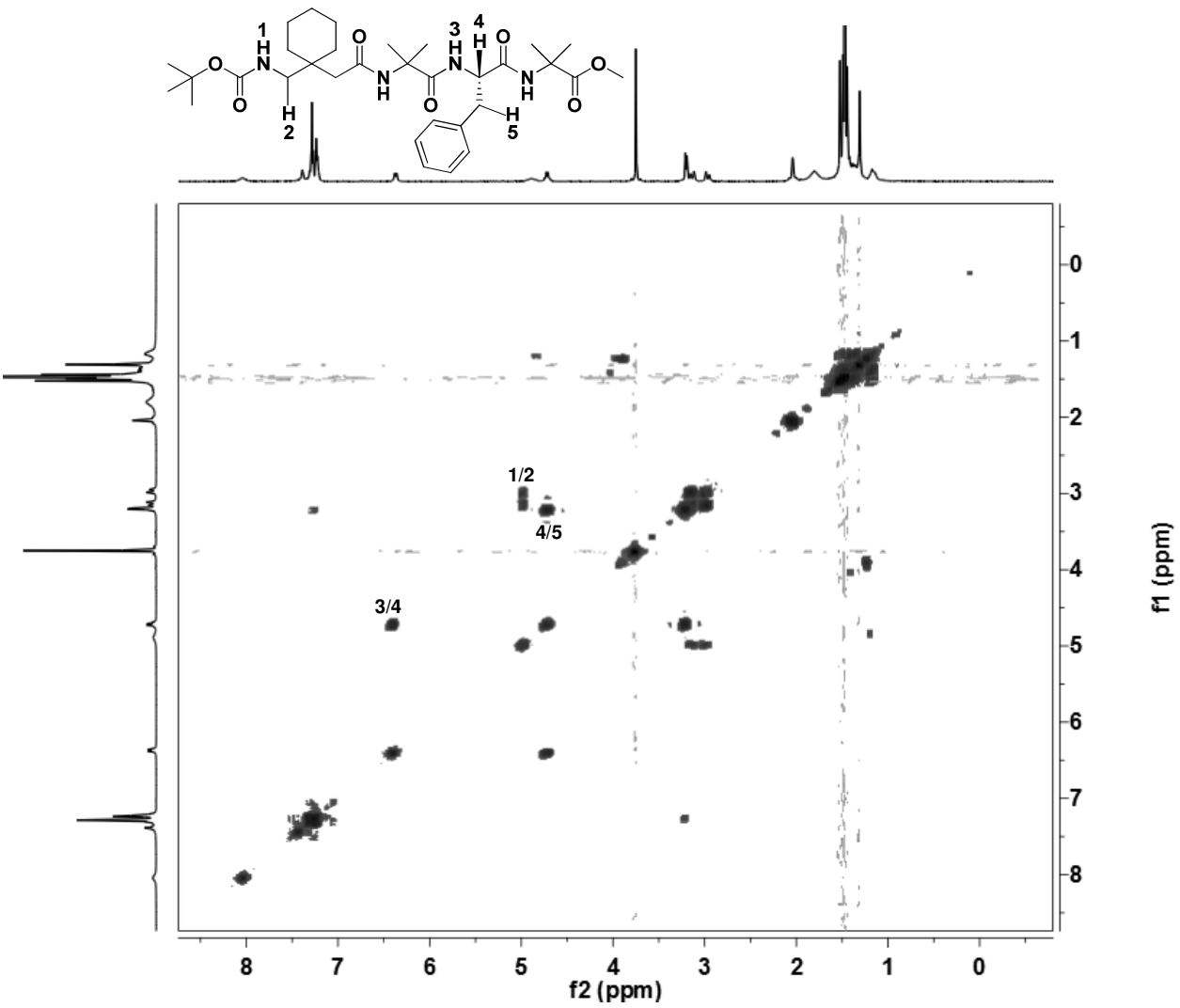


Figure S1. ^{1}H-^{1}H COSY spectrum of peptide P**1** (400 MHz, $CDCl_3$ at 298 K).

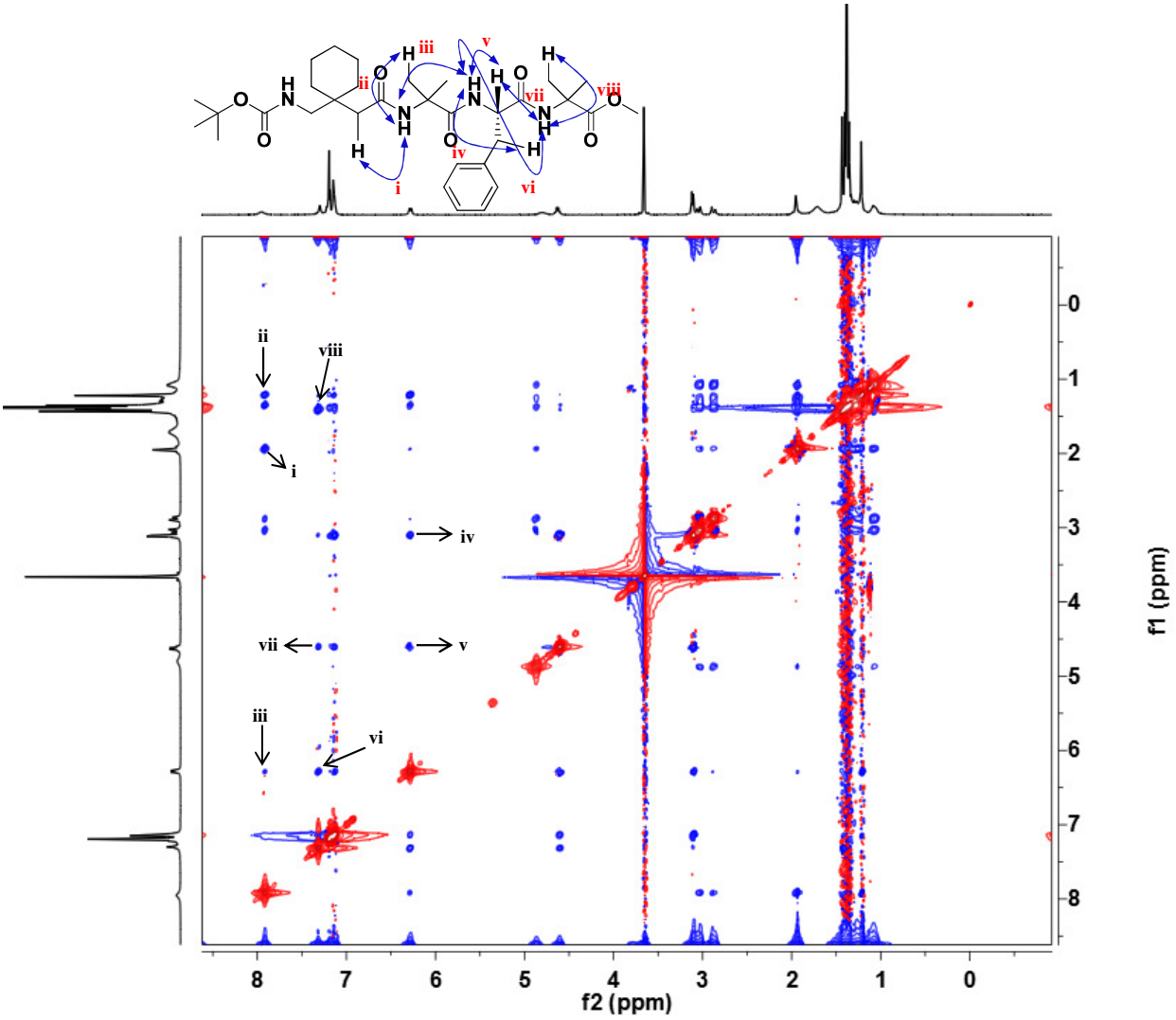


Figure S2. ^{1}H-^{1}H ROESY spectrum of peptide **P1** (400 MHz, $CDCl_3$ at 298 K, mixing time = 200 ms).

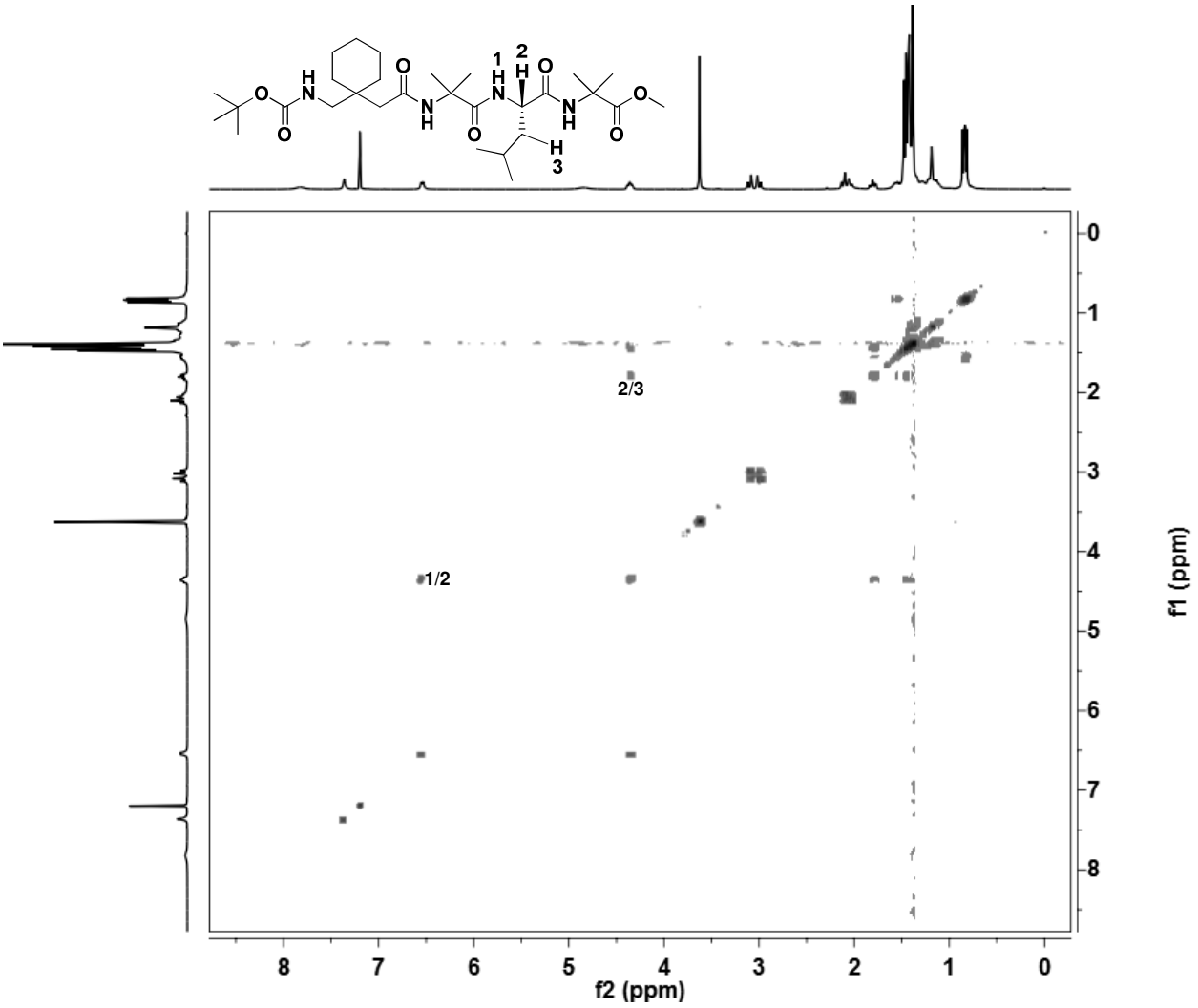


Figure S3. ^{1}H-^{1}H COSY spectrum of peptide P**2** (400 MHz, $CDCl_3$ at 298 K).

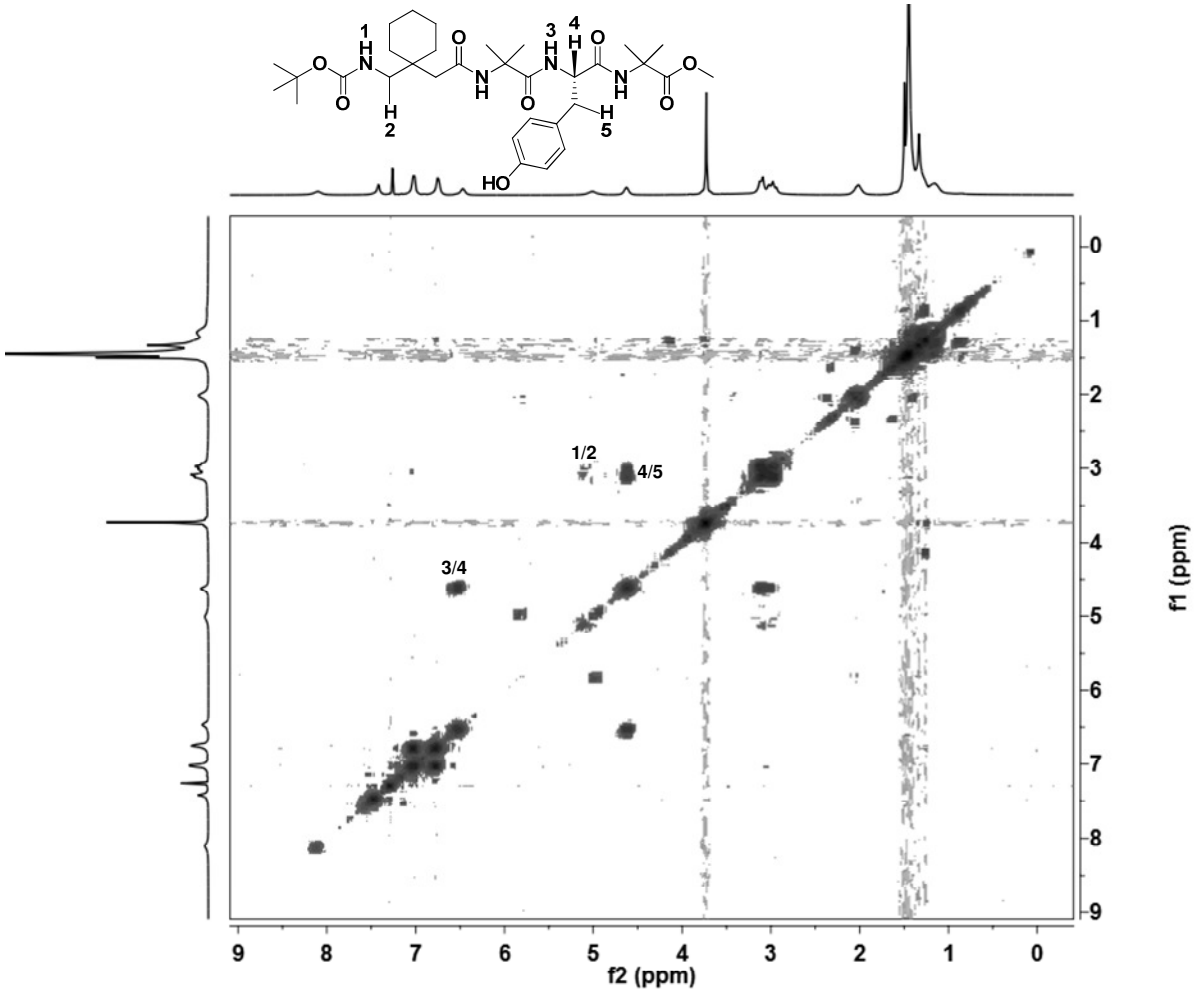


Figure S4. ^{1}H-^{1}H COSY spectrum of peptide P**3** (400 MHz, $CDCl_3$ at 298 K).

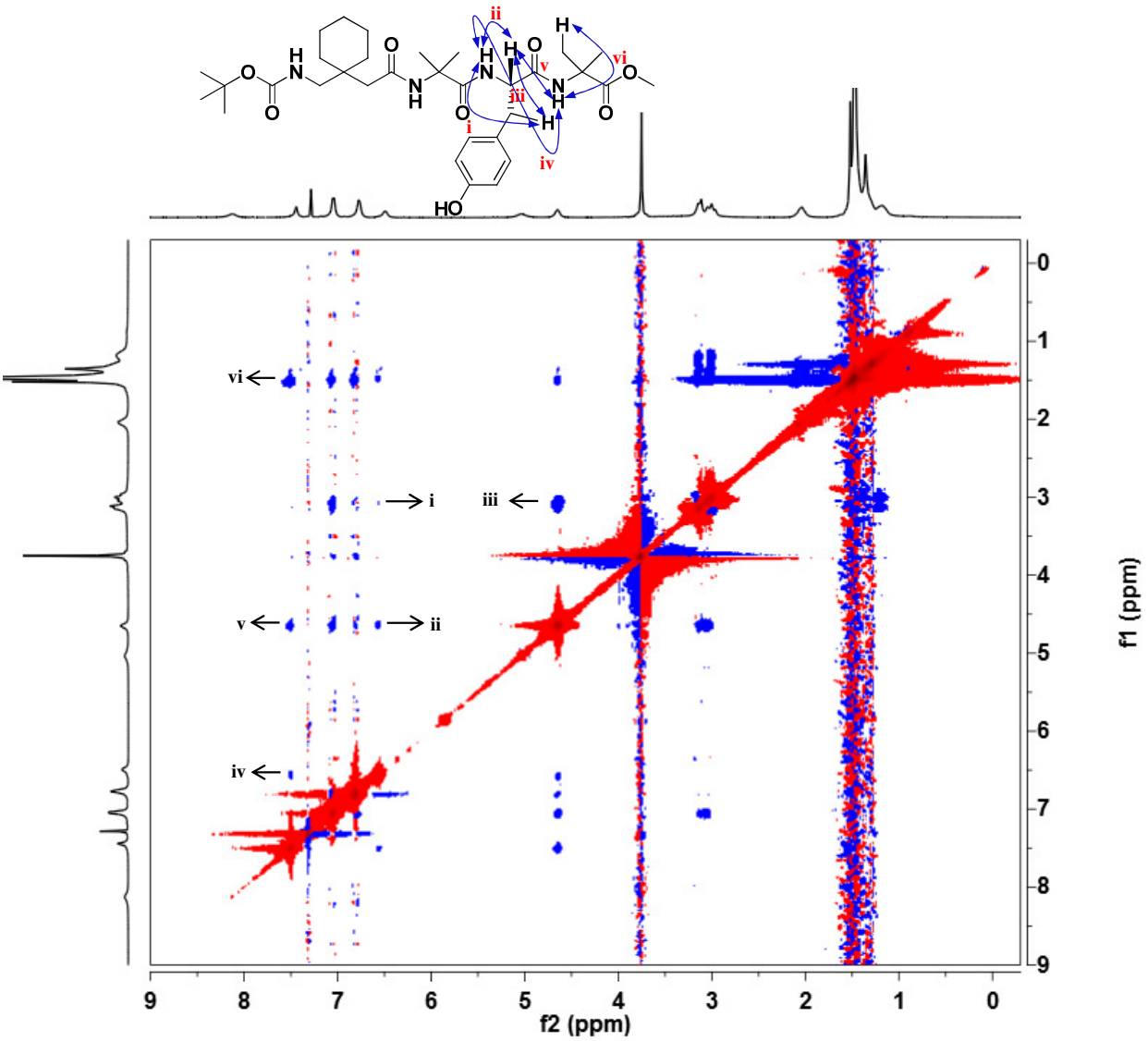


Figure S5. ^{1}H-^{1}H ROESY spectrum of peptide P**3** (400 MHz, $CDCl_3$ at 298 K, mixing time = 200 ms).

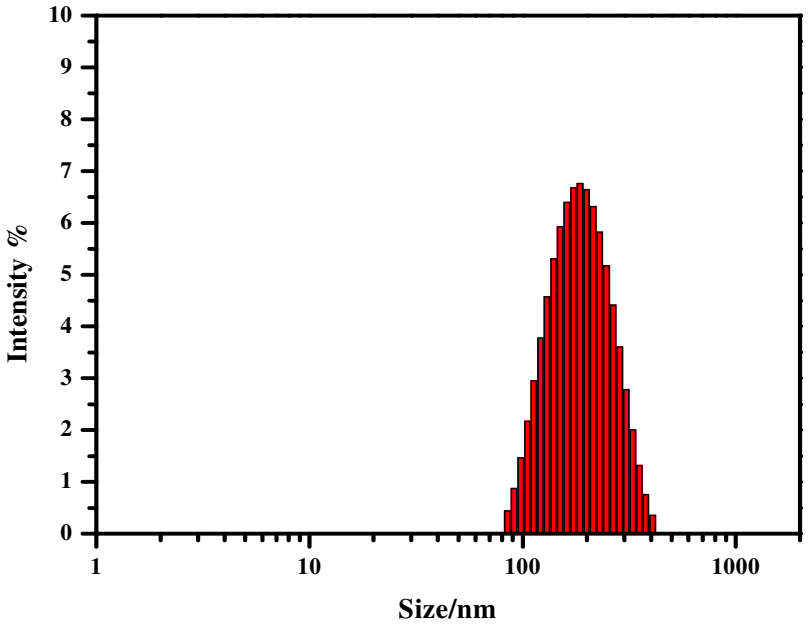


Figure S6. Dynamic light scattering (DLS) studies of self-assembled spherical aggregates of P**1** formed in methanol : water (1 : 1 v/v, *c* = 3.0 mmol/L) at ambient conditions reveals the size distribution of self-assembled spherical aggregates.

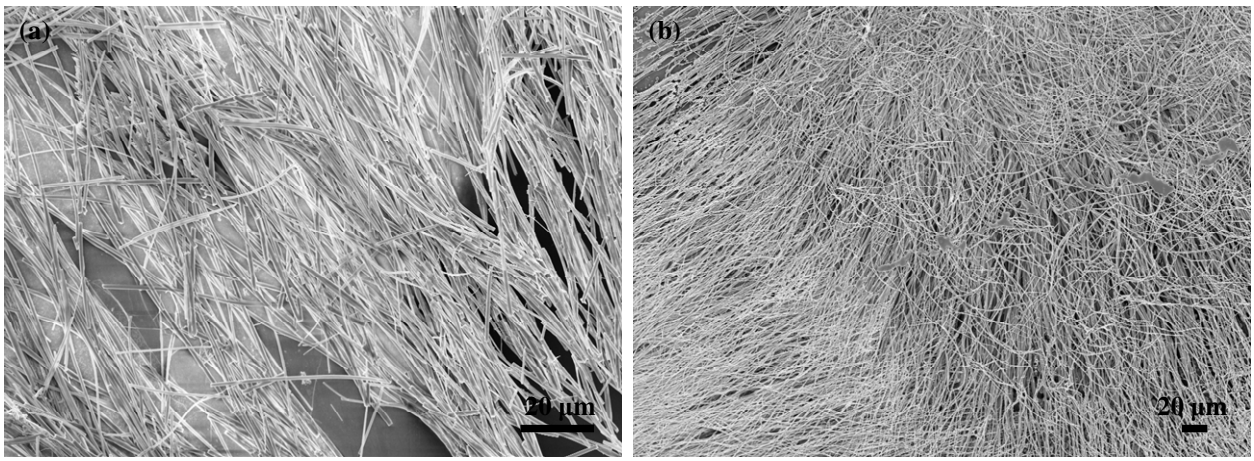


Figure S7. SEM images showing morphological transformations of P**1** by salt-prompted disruption studies in (a) $CaCl_2$ and (b) NaCl respectively. (a) Mineralized aligned nanofibers are formed from disruption of spheres with $CaCl_2$ (c = 3.0 mmol L^{-1} in methanol : water (1 : 1 v/v)) to P**1** in methanol : water (1 : 1 v/v, c = 3.0 mmol/L) at ambient conditions. (b) mineralized aligned fibers are formed from disruption of spheres with NaCl (c = 3.0 mmol L^{-1} in methanol : water (1 : 1 v/v)) to P**1** in methanol : water (1 : 1 v/v, c = 3.0 mmol/L) at ambient conditions. Salt solution mixed to the P**1** 1 : 1 v/v in methanol : water.

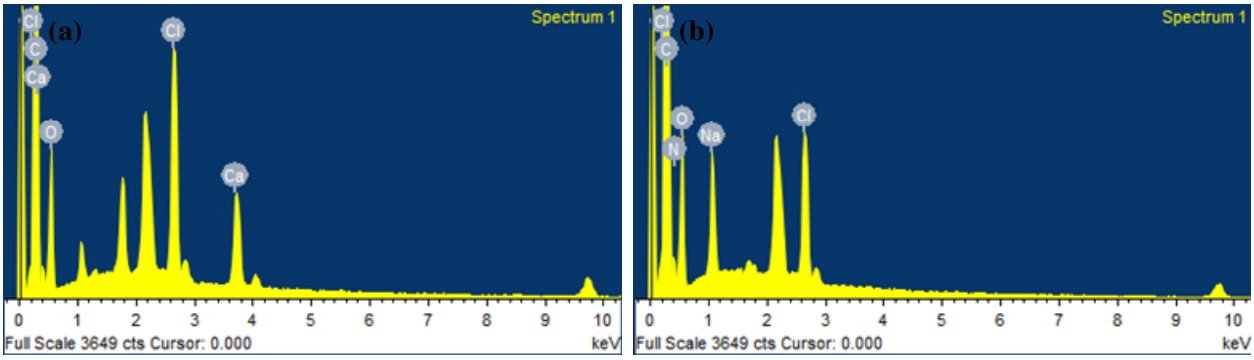


Figure S8. EDX analysis of raptured spherical aggregates of P**1** by salt-prompted disruption studies in (a) $CaCl_2$ and (b) NaCl respectively. (a) shows EDX spectrum of raptured spherical aggregates of P**1** by $CaCl_2$ indicating the presence of C, N, O, Ca and Cl. (b) shows EDX spectrum of raptured spherical aggregates of P**1** by NaCl indicating the presence of C, N, O, Na and Cl.

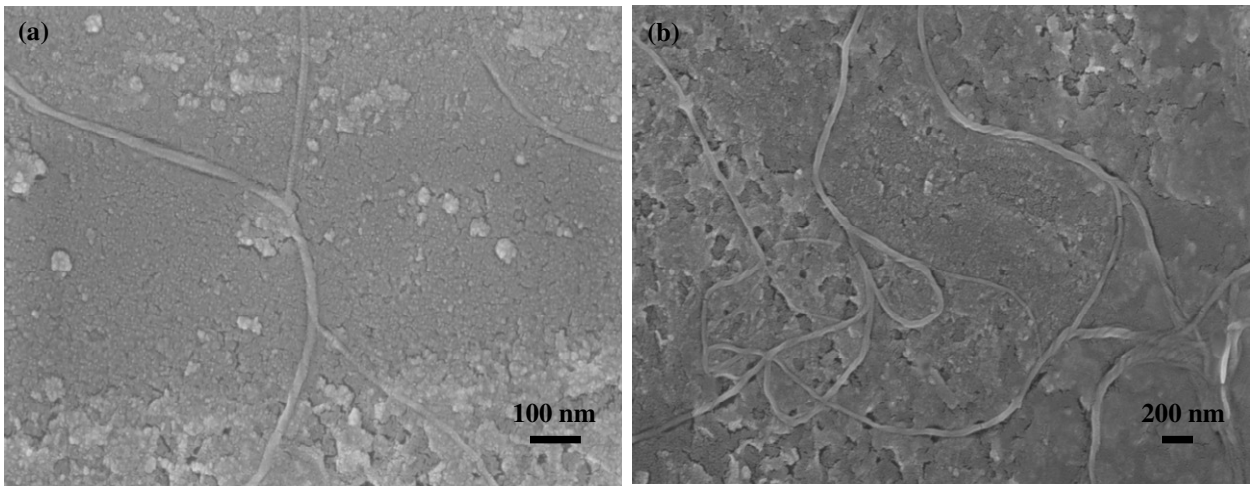


Figure S9. SEM images (a) and (b) show twisted nanofibers of P**2** in methanol : water (1 : 1 v/v, c = 3.0 mmol L^{-1}).

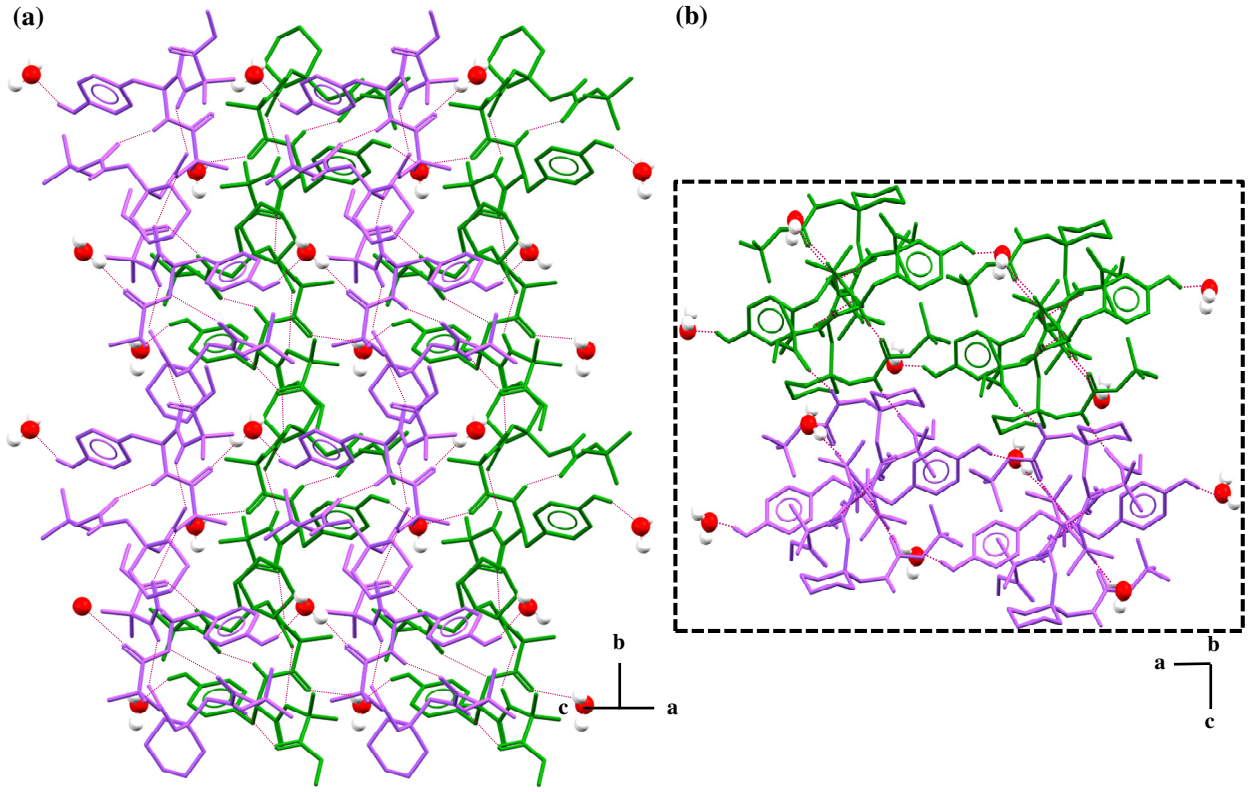


Figure S10. A higher order packing diagram of P**3** showing the role of the water molecules (red ball-stick representation) for formation of the higher ordered supramolecular architecture. (a) Packing diagram of P**3** at supramolecular level illustrating that the water molecules are important for interconnecting the intermolecular hydrogen-bonded supramolecular helices assembly using along the crystallographic *b* axis, supramolecular helices formed by molecule A (purple), supramolecular helix generated by the molecule B (light green) the supramolecular helix-helix are antiparallel self-associate to form higher ordered supramolecular architectures. (b) top view of higher order supramolecular helix demonstrating the important as bridging water molecules in crystal packing. Hydrogen bonds are shown as dotted lines.

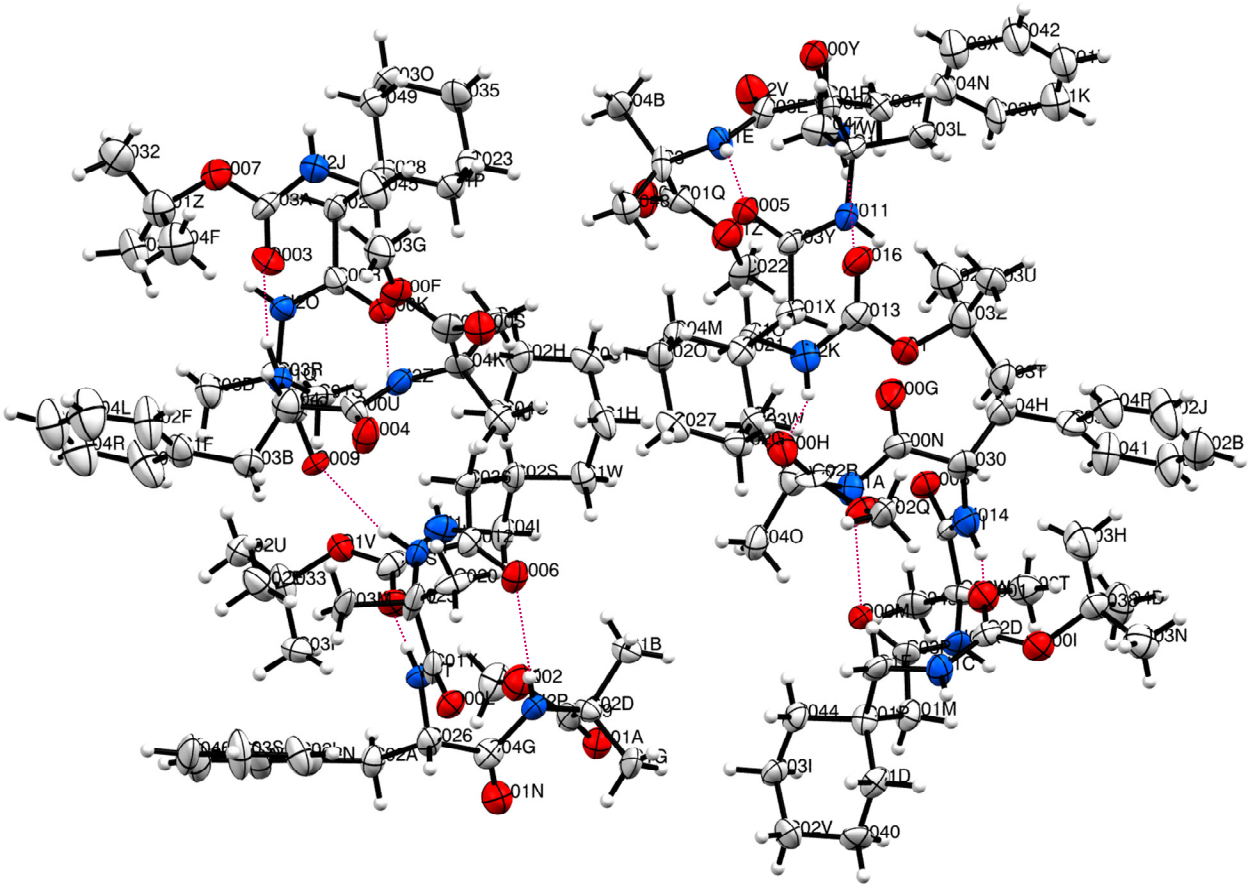


Figure S11. ORTEP diagram of peptide P**1** with atomic numbering. Thermal ellipsoids are shown at 50% probability. Hydrogen atoms are not labelled for clarity. Hydrogen bonds are shown as dotted lines.

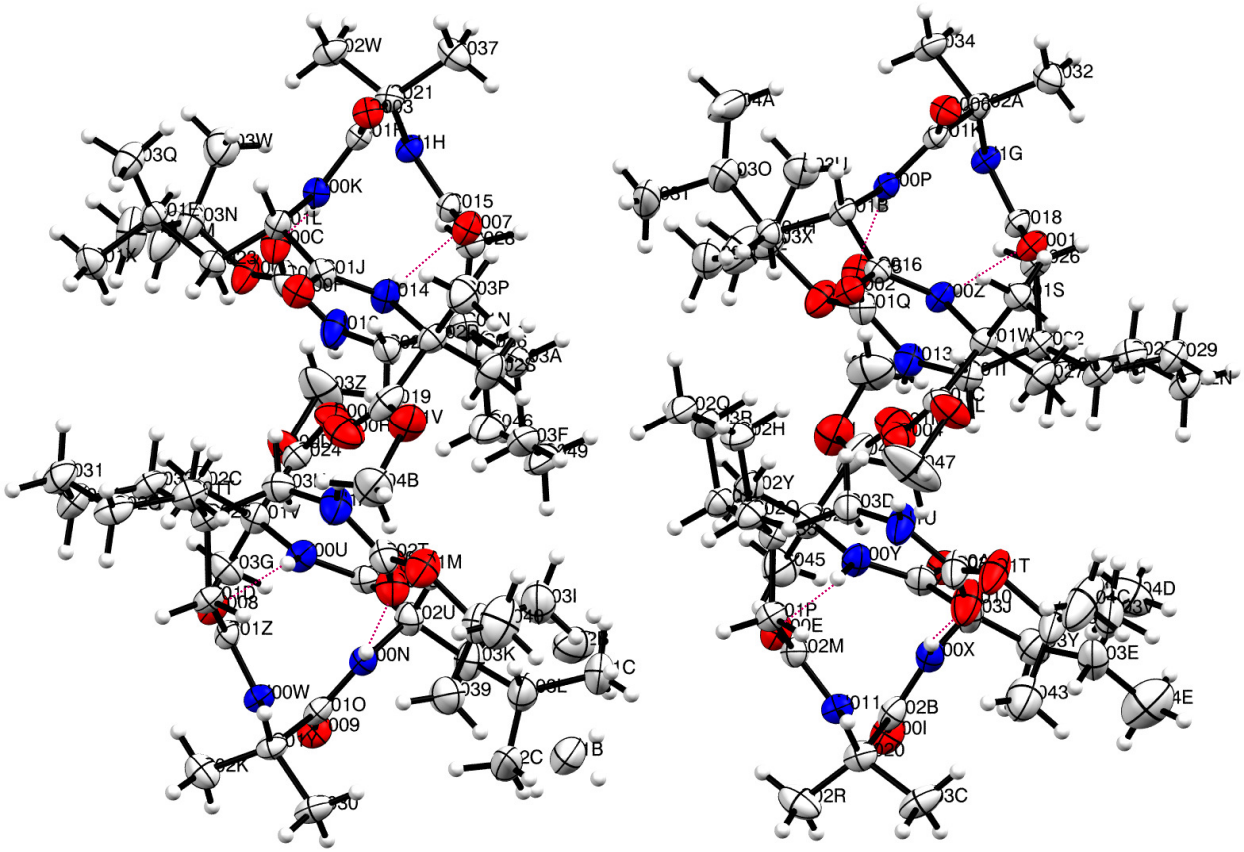


Figure S12. ORTEP diagram of peptide P**2** with atomic numbering. Thermal ellipsoids are shown at 50% probability. Hydrogen atoms are not labeled for clarity. Hydrogen bonds are shown as dotted lines.

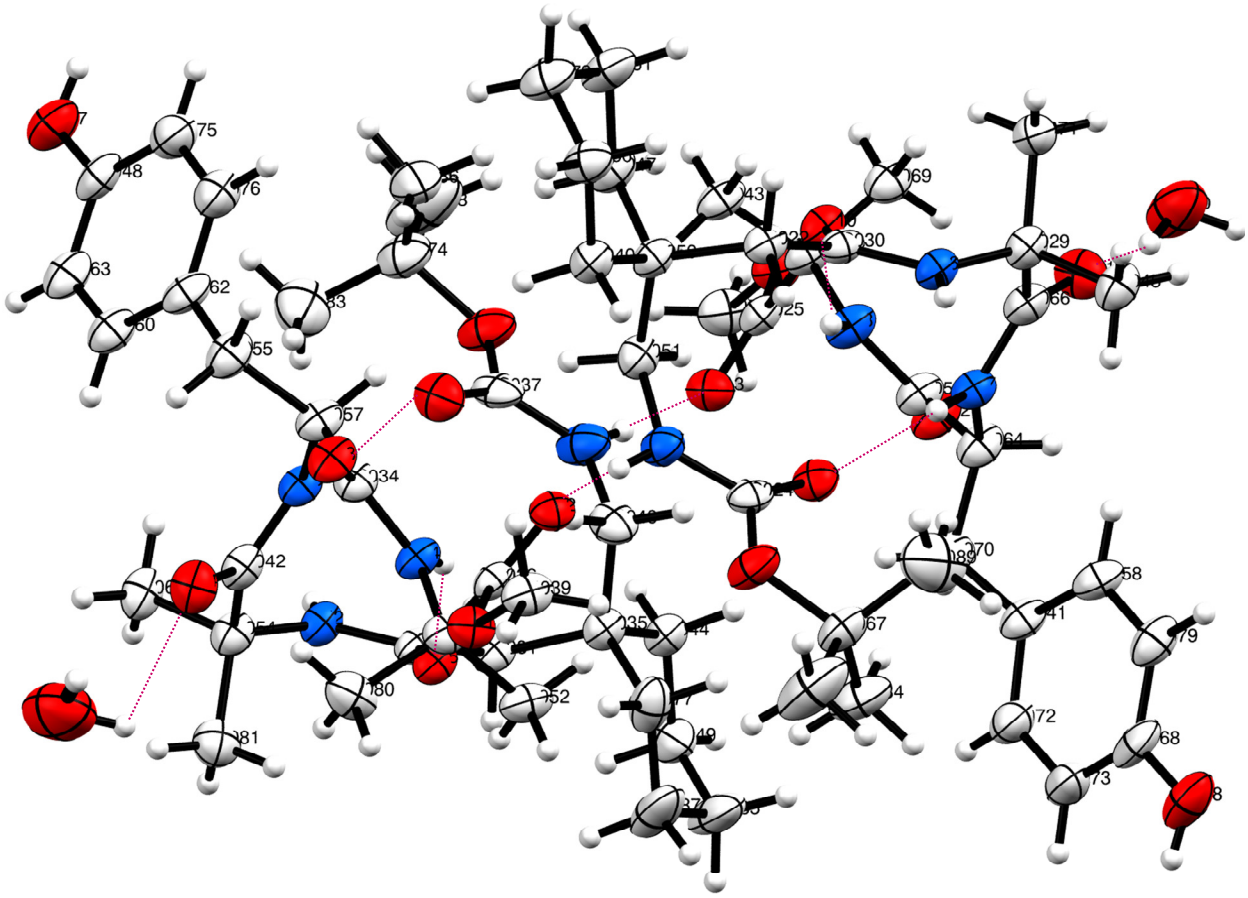
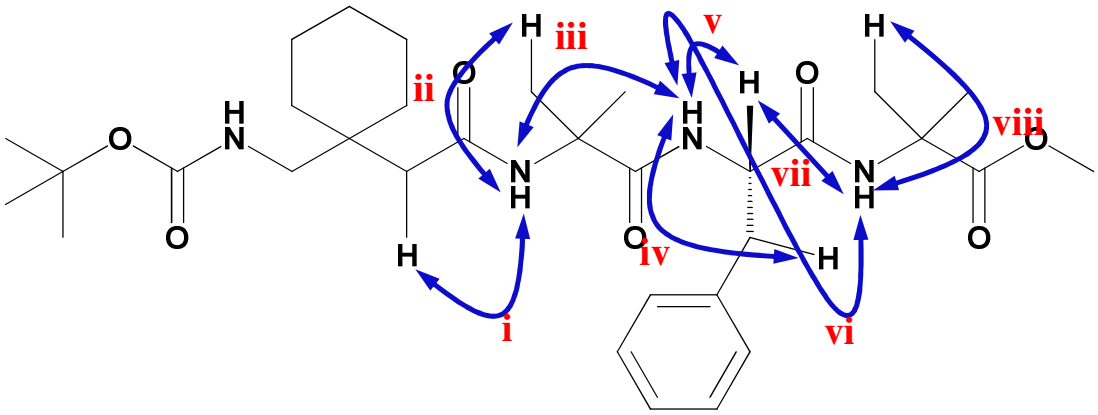


Figure S13. ORTEP diagram of peptide P**3** with atomic numbering along with the two water molecules O20 and O50. Thermal ellipsoids are shown at 50% probability. Hydrogen atoms are not labeled for clarity. Hydrogen bonds are shown as dotted lines.

Table S1. Significant Inter-Residue NOEs observed for P**1** and P**3.**

Peptide P**1**	Peptide P**3**
(i) Aib(2) NH ↔ Gpn(1) $C^{\alpha}H$	(i) Tyr(3) NH ↔ Tyr(3) $C^{\beta}H$
(ii) Aib(2) NH ↔ Aib(2) $C^{\beta}H$	(ii) Tyr(3) NH ↔ Tyr(3) $C^{\alpha}H$
(iii) Aib(2) NH ↔ Phe(3) NH	(iii) Tyr(3) $C^{\alpha}H$ ↔ Tyr(3) $C^{\beta}H$
(iv) Phe(3) NH ↔ Phe(3) $C^{\beta}H$	(iv) Aib(4) NH ↔ Tyr(3) NH
(v) Phe(3) NH ↔ Phe(3) $C^{\alpha}H$	(v) Aib(4) NH ↔ Tyr(3) $C^{\alpha}H$
(vi) Aib(4) NH ↔ Phe(3) NH	(vi) Aib(4) NH ↔ Aib(4) $C^{\beta}H$
(vii) Aib(4) NH ↔ Phe(3) $C^{\alpha}H$	
(viii) Aib(4) NH ↔ Aib(4) $C^{\beta}H$	

NOEs observed in Peptide P**1**



NOEs observed in Peptide P**3**

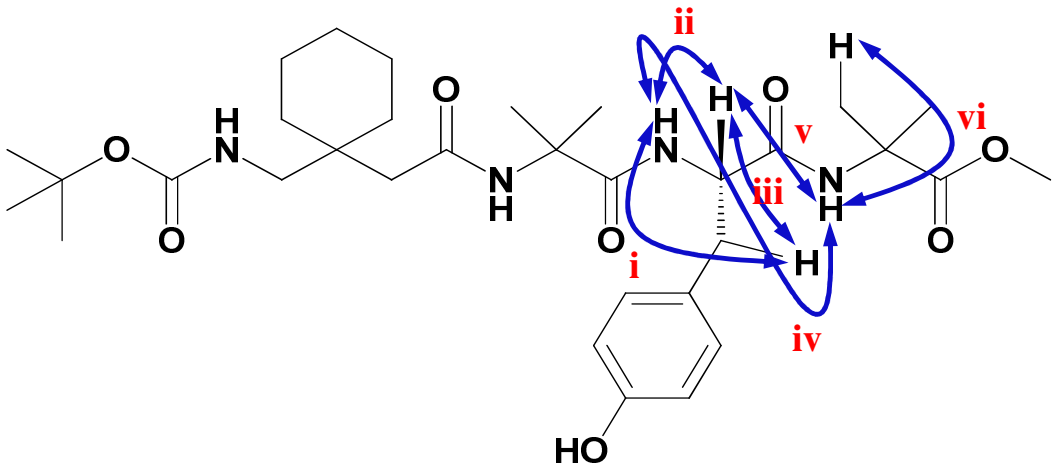
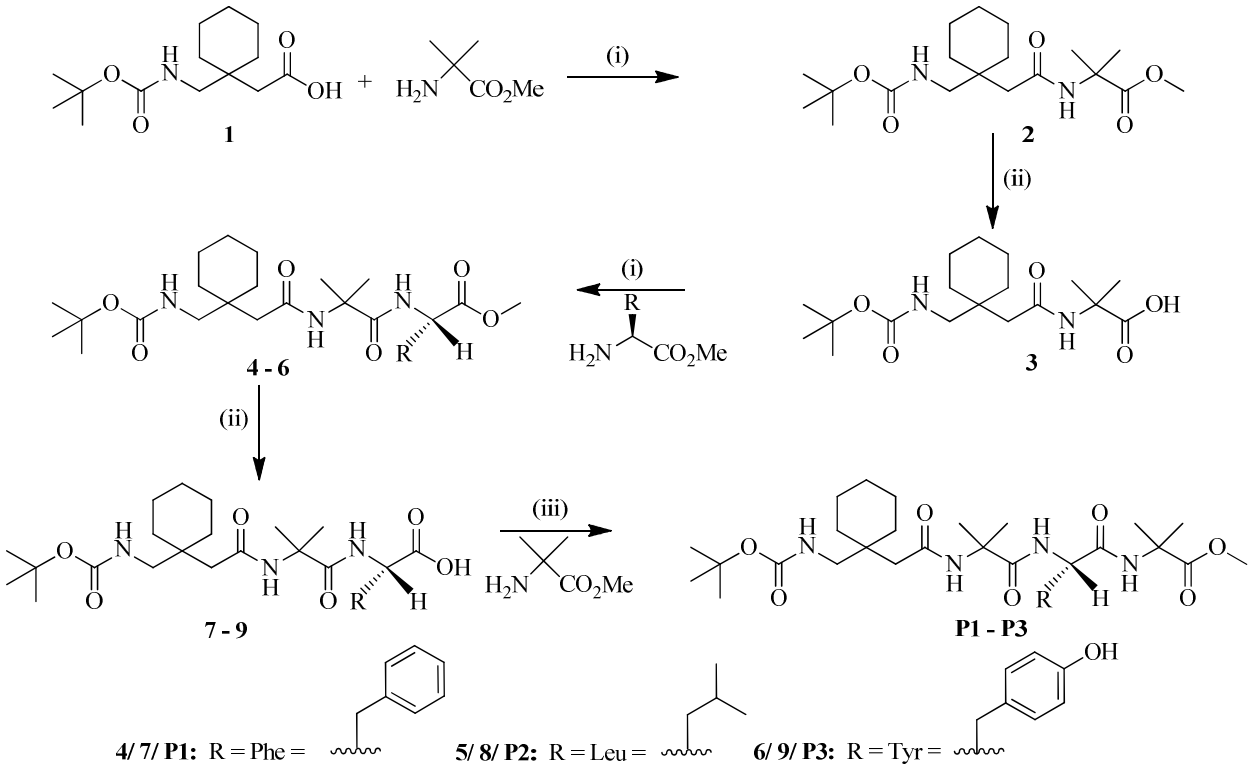


Table S2. Crystal and diffraction parameters of peptides P**1**-P**3.**

	P**1**	P**2**	P**3**
Empirical formula	$C_{32}H_{50}N_4O_7$	$C_{29}H_{52}N_4O_7$	$C_{32}H_{50}N_4O_8.H_2O$
Crystalizing solvent	DMSO	Methanol/Water	Methanol/Water
Space group	*P1*	$P2_1$	$P2_1$
a (Å)	12.332(3)	20.900(4)	11.493(2)
b (Å)	12.412(3)	12.965(3)	17.104(3)
c (Å)	21.736(4)	23.973(5)	18.432(4)
α (deg)	90.58	90	90
β (deg)	95.38	100.04(3)	103.96(3)
γ (deg)	90	90	90
Volume ($Å^3$)	3312.2(12)	6396.45	3516.28
Z	4	4	2
Molecules/asymmetric unit	4	4	2
Co-crystallized solvent	None	None	Water
Molecular weight	602.37	568.75	636.37
Density (g/cm^3) (cal)	1.209	1.181	1.203
F (000)	1304	2480	1376
Radiation	Cu	Cu	Cu
Wavelength (Å)	1.54178	1.54178	1.54178
Temperature (K)	123	120	125
θ Max. (˚)	69.229	68.246	69.210
Measured reflections	90867	37008	25314
Unique reflections	21748	22834	11557
R_{int}	0.0704	0.0647	0.0635
Final R (%)/wR2 (%)	0.1216 / 0.3687	0.0636/0.2265	0.0527/0.1268
Goodness-of-fit (S)	1.506	1.029	0.943
No. of. Parameters / Restraints	1576 / 3	1521/27	828/2
Data-to-parameters ratio	13.8	16.7	14

Table S3. Backbone dihedral angles for peptides P**1**-P**3**.

peptide	Residue	ϕ	θ_1	θ_2	ψ	ω
P**1** **Molecule A**	Gpn(1)	98	-63	-58	158	168
	Aib(2)	58			32	176
	Phe(3)	71			16	158
	Aib(4)	-58			-30	-
P**1** **Molecule B**	Gpn(1)	-96	66	58	-159	-167
	Aib(2)	-53			-32	-171
	Phe(3)	-103			15	-173
	Aib(4)	49			42	-
P**1** **Molecule C**	Gpn(1)	-106	68	57	-159	-170
	Aib(2)	-54			-31	-171
	Phe(3)	-102			15	-172
	Aib(4)	52			37	-
P**1** **Molecule D**	Gpn(1)	105	-65	-58	158	170
	Aib(2)	60			31	175
	Phe(3)	75			08	159
	Aib(4)	-51			-32	-
P**2** **Molecule A**	Gpn(1)	102	-64	-58	151	170
	Aib(2)	-58			28	-177
	Leu(3)	56			35	-180
	Aib(4)	-54			139	-
P**2** **Molecule B**	Gpn(1)	-110	69	54	-135	-171
	Aib(2)	-50			-38	-172
	Leu(3)	-78			-16	175
	Aib(4)	54			-149	-
P**2** **Molecule C**	Gpn(1)	-99	63	58	-157	-167
	Aib(2)	-57			-32	-175
	Leu(3)	-83			-7	167
	Aib(4)	51			-137	-
P**2** **Molecule D**	Gpn(1)	112	-67	-55	136	173
	Aib(2)	52			36	175
	Leu(3)	59			38	172
	Aib(4)	-54			150	-
P**3** **Molecule A**	Gpn(1)	107	-62	-57	152	167
	Aib(2)	60			26	-176
	Tyr(3)	51			38	-170
	Aib(4)	-58			-146	-
P**3** **Molecule B**	Gpn(1)	-100	64	56	-158	-162
	Aib(2)	-62			-24	-176
	Tyr(3)	-75			-17	164
	Aib(4)	56			-141	-



Scheme S1 Chemical structures and synthetic scheme of peptides. Reagents and conditions: (i) DCC, HOBt, DMF; (ii) 2N NaOH, MeOH; (iii) DIPC, HOBt, DMF.

General Methods and Materials:

α-aminoisobutyric acid, L-leucine, L-phenylalanine, L-tyrosine, HOBt (1-hydroxybenzotriazole), DCC (*N*,*N*'-dicyclohexylcarbodiimide) were obtained commercially. For chemical reactions and purification of peptides, methanol, dimethylformamide, ethyl acetate and toluene were dried according to literature. Reactions were monitored by thin-layer chromatography (TLC). Visualization was attained with UV light and potassium permanganate stain followed by charring on hot-plate. All intermediates and final compounds were purified and well characterized by FT-IR, ^{1}H NMR (400 MHz), ^{13}C NMR (100 MHz) and mass spectral studies. The precursor 1-(aminomethyl)cyclohexaneacetic acid hydrochloride salt Gpn.hydrochloride was synthesized according to previously reported method.[1]

Synthesis of Peptides

Boc-Gpn-OH 1. Gabapentin hydrochloride (3.0 g, 14.48 mmol) **1** was added to a mixture of 1,4-dioxane : water (2:1, 30 mL) and neutralized with 1M NaOH (30 mL). This mixture was stirred and cooled in an ice-water bath (the pH should be above 8). After 15 minutes, 3.47 g (15.93 mmol) of di-*tert*-butylpyrocarbonate was added and stirred overnight at room temperature. The solution was concentrated under vacuum to about 10-15 mL and it was cooled for 5 minutes in an ice-water bath. The cooled solution was covered with a layer of ethyl acetate (about 30 mL) and acidified with 1M $KHSO_4$ to pH 2-3. It was extracted with ethyl acetate (3 × 30 mL). The extracted ethyl acetate was dried over anhydrous Na_2SO_4 and evaporated in vacuum to yield **2** as white solid[2] (3.72 g, 95 %). FT-IR (KBr), ῡ (cm^{-1}): 3416 (m), 2928 (m), 1713 (m), 1670 (m), 1535 (m), 1254 (w); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 11.97 (bs, 1H, COOH), 6.58 (m, 1H, NH of Gpn), 3.01 (d, *J* = 6.00 Hz, 2H, C^{γ}Hs of Gpn), 2.13 (s, 2H, C^{α}Hs of Gpn), 1.50-1.20

(m, 19H); ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 173.21, 156.10, 77.51, 46.40, 36.68, 32.39, 28.24, 25.58, 21.07. MS (ESI) *m/z* for $C_{14}H_{25}NO_4Na$ $(M+Na)^+$ calcd.: 294.1681, found 293.9518.

General Procedure for Methyl ester Hydrolysis:

Terminally protected dipeptide or tripeptide in methanol (20 mL/g) was taken in a round bottom flask (R.B) and 2N NaOH was added drop wise. The progress of hydrolysis was monitored by thin layer chromatography (TLC). The reaction was allowed for overnight. After completion of the reaction, 15 mL of distilled water was added to the reaction mixture and methanol was removed under vacuum. The aqueous part was washed with diethyl ether (2 × 30 mL). Then aqueous part was cooled under ice-water bath for 15 minutes and then pH was adjusted to 2-3 by drop wise addition of 1M HCl. It was extracted with ethyl acetate (3 × 50 ml). The extracted ethyl acetate was dried over anhydrous Na_2SO_4 and evaporated under vacuum to yield corresponding carboxylic acid which was used for the next step without purification.

General Procedure for Peptide Coupling:

Boc-protected compound (1.0 equiv) was dissolved in dry-DMF (4 mL/g) and stirred on an ice-water bath. Methyl ester protected amino acid was isolated from its corresponding methyl ester hydrochloride (2.0 equiv) by neutralization and subsequently extracted twice with ethyl acetate (2 × 30 mL). The collected ethyl acetate extracts was dried over anhydrous Na_2SO_4 and concentrated to 6-8 mL. It was then added to the pre-cooled reaction mixture followed by addition of (1.0 equiv) HOBt, (1.1 equiv) dicyclohexylcarbodiimide (DCC) for di and tripeptides synthesis. 1.1 equiv diisopropylcarbodiimide (DIPC) was used as a coupling reagent for tetrapeptides synthesis. The reaction mixture was allowed to come to room temperature and stirred for 2 days for dipeptide or 3 days for tripeptides or 4 days for tetrapeptides synthesis.

After completion of the reaction, ethyl acetate (30 mL) was added to the reaction mixture and dicyclohexylurea or diisopropylurea was filtered off. The organic layer was washed with 1M HCl (3 × 50 mL), brine (2 × 50 mL), 1M sodium carbonate (3 × 50 mL) and brine (2 × 50 mL) and dried over anhydrous Na_2SO_4 and evaporated in a vacuum. The purification was done by using silica gel column (100-200 mesh).

Boc-Gpn(1)-Aib(2)-OMe 2.

2 was obtained as white solid[3] (3.00 g, 88 %). FT-IR (KBr), ῡ (cm^{-1}): 3383 (m), 2928 (s), 2855 (m), 1745 (s), 1692 (s), 1649 (s), 1542 (s), 1279 (m), 1154 (s); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 7.50 (s, 1H, NH of Aib(2)), 5.10-5.00 (m, 1H, NH of Gpn(1)), 3.70 (s, 3H, OCH_3), 3.14 (d, *J* = 7.04 Hz, 2H, C^{γ}Hs of Gpn), 2.07 (s, 2H, C^{α}Hs of Gpn), 1.55-1.46 (m, 11H), 1.45-1.40 (m, 11H), 1.39-1.32 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$, δ ppm): 175.18, 170.93, 157.37, 56.21, 52.50, 37.69, 34.35, 28.53, 26.17, 25.11, 21.64. MS (ESI) *m/z* for $C_{19}H_{34}N_2O_5Na$ $(M+Na)^+$ calcd.: 393.2365, found 393.3706.

Boc-Gpn(1)-Aib(2)-OH 3.

3 was obtained as white solid[3] (1.89 g, 89 %). FT-IR (KBr), ῡ (cm^{-1}): 3354 (m), 3269 (m), 2933 (m), 1727 (ms), 1684 (s), 1623 (m), 1544 (ms), 1395 (w), 1282 (m); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 12.07 (bs, 1H, COOH), 8.16 (s, 1H, NH of Aib(2)), 6.65 (t, *J* = 6 Hz, 1H, NH of Gpn(1)), 2.97 (d, *J* = 6.28 Hz, 2H, C^{γ}Hs of Gpn), 1.97 (s, 2H, C^{α}Hs of Gpn), 1.50-1.35 (m, 14H), 1.34-1.15 (m, 11H); ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 175.53, 170.04, 156.21, 77.67, 54.64, 46.46, 41.68, 37.23, 33.13, 28.24, 25.74, 24.88, 21.10. MS (ESI) *m/z* for $C_{18}H_{32}N_2O_5Na$ $(M+Na)^+$ calcd.: 379.2209, found 379.2342.

Boc-Gpn(1)-Aib(2)-Phe(3)-OMe 4.

Compound **4** was obtained as white solid (0.553 g, 85 %). $[\alpha]_D^{20}$: +0.04 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3422 (ms), 3319 (ms), 2926 (m), 1757 (m), 1714 (ms), 1661 (s), 1503 (s), 1169 (s); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 7.51 (s, 1H, NH of Aib(2)), 7.21-7.17 (m, 1H, NH of Phe (3)), 7.34-7.12 (m, 5H, aromatic protons of Phe), 5.10-4.96 (bs, 1H, NH of Gpn (1)), 4.88-4.77 (m, 1H, $C^{\alpha}H$ of Phe), 3.69 (s, 3H,OCH_3), 3.19-3.11 (m, 2H, $C^{\beta}Hs$ of Phe), 3.11-3.04 (m, 2H, $C^{\gamma}Hs$ of Gpn), 2.08 (s, 2H, $C^{\alpha}Hs$ of Gpn), 1.53-1.46 (m, 11H), 1.43 (s, 9H, CH_3), 1.39-1.34 (m, 3H), 1.28-1.23 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$, δ ppm): 174.25, 172.23, 171.97, 164.82, 129.51, 128.56, 127.07, 80.82, 57.53, 54.45, 53.59, 52.36, 45.82, 38.02, 37.69, 34.31, 28.51, 26.09, 25.31, 25.21, 22.52, 21.60. HRMS (ESI) *m/z* for $C_{28}H_{44}N_3O_6$ $(M+H)^+$ calcd.: 518.3230, found 518.3214.

Boc-Gpn(1)-Aib(2)-Leu(3)-OMe 5.

Compound **5** was obtained as white crystalline solid (0.505 g, 83 %). $[\alpha]_D^{20}$: -1.90 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3432 (w), 3314 (m), 2934 (ms), 1742 (ms), 1689 (s), 1552 (ms), 1364 (w), 1171 (m); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 7.53 (s, 1H, NH of Aib(2)), 7.42 (d, *J* = 7.8 Hz, 1H, NH of Leu (3)), 5.03 (bs, 1H, NH of Gpn (1)), 4.60-4.50 (m, 1H, $C^{\alpha}H$ of Leu), 3.70 (s, 3H,OCH_3), 3.14 (d, *J* = 7.04 Hz, 2H, $C^{\gamma}Hs$ of Gpn), 2.14 (s, 2H, $C^{\alpha}Hs$ of Gpn), 1.70-1.50 (m, 10H), 1.49-1.33 (s, 16H), 1.32-1.20 (m, 2H), 0.92 (d, *J* = 6 Hz, 6H, $C^{\delta}Hs$ of Leu); ^{13}C NMR (100 MHz, $CDCl_3$, δ ppm): 174.66, 173.65, 171.99, 157.47, 80.03, 57.76, 52.28, 51.14, 47.10, 42.94, 41.53, 37.72, 34.32, 28.50, 26.09, 25.45, 25.26, 24.95, 22.98, 22.05, 21.60. HRMS (ESI) *m/z* for $C_{25}H_{46}N_3O_6$ $(M+H)^+$ calcd.: 484.3387, found 484.3472.

Boc-Gpn(1)-Aib(2)-Tyr(3)-OMe 6.

Compound **6** was obtained as white fluffy solid (0.537 g, 80 %). $[\alpha]_D^{20}$: +0.80 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3416 (s), 2928 (s), 1746 (ms), 1659 (s), 1516 (s), 1365 (ms) 1169 (ms); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 7.85 (s, 1H, NH of Aib(2)), 7.11 (bs, 1H, NH of Tyr(3)), 6.97 (d, *J* = 8.04 Hz, 2H, ring protons of Tyr), 6.75 (m, 2H, ring protons of Tyr), 5.10 (bs, 1H, NH of Gpn(1)), 4.83-4.67 (m, 1H, $C^{\alpha}H$ of Tyr), 3.69 (s, 3H, OCH_3), 3.20-3.02 (m, 2H, $C^{\beta}Hs$ of Tyr), 3.01-2.83 (m, 2H, $C^{\alpha}Hs$ of Gpn), 2.09 (s, 2H, $C^{\gamma}Hs$ of Gpn), 1.58-1.46 (m, 8H), 1.44 (s, 9H, CH_3), 1.42-1.34 (m, 5H), 1.29-1.23 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$, δ ppm): 174.32, 172.41, 172.04, 157.58, 155.63, 130.51, 115.77, 80.25, 57.64, 53.79, 52.42, 46.79, 42.93, 37.72, 37.13, 34.22, 29.82, 28.51, 26.03, 25.26, 21.60. HRMS (ESI) *m/z* for $C_{28}H_{43}N_3O_7Na$ $(M+Na)^+$ calcd.: 556.2999, found 556.2991.

Boc-Gpn(1)-Aib(2)-Phe(3)-OH 7.

Compound **7** was obtained as white solid (0.253 g, 86 %). $[\alpha]_D^{20}$: +2.65 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3413 (w), 3308 (w), 2974 (w), 1693 (m), 1660 (m), 1516 (m), 1172 (m); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 12.73 (bs, 1H, COOH), 7.92 (m, 1H, NH of Aib (2)), 7.46 (d, *J* = 7.76 Hz, 1H, NH of Phe(3)), 7.25-7.15 (m, 5H, aromatic protons of Phe), 6.61 (m, 1H, NH of Gpn (1)), 4.38 (m, 1H, $C^{\alpha}H$ of Phe), 3.05-3.00 (m, 2H, $C^{\beta}Hs$ of Phe), 2.98-2.90 (m, 2H, $C^{\gamma}Hs$ of Gpn), 2.00 (s, 2H, $C^{\alpha}Hs$ of Gpn), 1.43-1.34 (m, 13H), 1.32-1.15 (m, 12H); ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 173.64, 172.76, 170.67, 156.12, 137.37, 129.22, 128.01, 126.33, 79.13, 77.64, 55.94, 53.28, 46.39, 42.10, 37.03, 36.60, 33.09, 28.21, 25.66, 24.93, 24.69, 21.04. HRMS (ESI) *m/z* for $C_{27}H_{42}N_3O_6$ $(M+H)^+$ calcd.: 504.3074, found 504.3068.

Boc-Gpn(1)-Aib(2)-Leu(3)-OH 8.

Compound **8** was obtained as white solid (0.297 g, 88 %). $[\alpha]_D^{20}$: -2.10 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3440 (m), 3373 (m), 3304 (m), 2921 (ms), 1716 (ms), 1691 (s), 1654 (s), 1535 (s), 1514 (s), 1175 (m); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 12.49 (bs, 1H, COOH), 7.87 (s, 1H, NH of Aib (2)), 7.55 (d, *J* = 8.0 Hz, 1H, NH of Leu(3)), 6.66 (t, *J* = 6.24 Hz, 1H, NH of Gpn (1)), 4.25-4.15 (m, 1H, C^{α}H of Leu), 2.98 (d, *J* = 6.04 Hz, 2H, C^{γ}Hs of Gpn), 2.02 (s, 2H, C^{α}Hs of Gpn), 1.68-1.55 (m, 2H, C^{β}Hs of Leu), 1.49-1.35 (m, 15H), 1.34-1.20 (m, 11H), 0.87-0.78 (m, 6H); ^{13}C NMR (100 MHz, DMSO-d_6, δ ppm): 174.20, 173.86, 170.68, 156.22, 77.70, 55.94, 50.26, 37.03, 33.38, 33.24, 28.30, 25.77, 25.55, 24.42, 24.05, 23.01, 21.33, 21.13. HRMS (ESI) *m/z* for $C_{24}H_{43}N_3O_6K$ $(M+K)^+$ calcd.: 508.2789, found 508.2753.

Boc-Gpn(1)-Aib(2)-Tyr(3)-OH 9.

Compound **9** was obtained as white foam solid (0.302 g, 82 %). $[\alpha]_D^{20}$: +2.15 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3352 (bs), 2930 (s), 1726 (s), 1656 (s), 1516 (s), 1451 (ms), 1248 (ms), 1170 (ms); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 12.63 (bs, 1H, COOH), 9.20 (bs, 1H, OH of Tyr(3)), 7.96 (s, 1H, NH of Aib(2)) 7.35 (d, *J* = 7.52 Hz, 1H, NH of Tyr), 6.96 (d, *J* = 8.28 Hz, 2H, ring protons of Tyr), 6.66 (m, 1H, NH of Gpn), 6.63 (d, *J* = 8.52 Hz, 2H, ring protons of Tyr), 4.37-4.28 (m, 1H, C^{α}H of Tyr), 2.98 (m, 2H, C^{γ}Hs of Gpn), 2.95-2.80 (m, 2H, C^{β}Hs of Tyr), 2.02 (s, 2H, C^{α}Hs of Gpn), 1.44-1.22 (m, 25H); ^{13}C NMR (100 MHz, DMSO-d_6 δ ppm): *δ* 173.61, 172.92, 170.79, 156.23, 155.95, 130.26, 129.98, 127.31, 114.91, 77.73, 59.82, 56.06, 53.63, 37.15, 36.00, 33.18, 31.02, 28.29, 25.76, 25.02, 24.85, 22.55, 22.12, 21.13. HRMS (ESI) *m/z* for $C_{27}H_{42}N_3O_7$ $(M+H)^+$ calcd.: 520.3023, found 520.3113.

Boc-Gpn(1)-Aib(2)-Phe(3)Aib(4)-OMe P1.

Peptide P**1** was obtained as white solid (0.185 g, 77 %). $[\alpha]_D^{20}$: −3.02 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3406 (ms), 3318 (s), 2923 (m), 1732 (m), 1703 (ms), 1651 (s), 1536 (s), 1279 (ms), 1172 (ms). ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 8.04 (s, 1H, NH of Aib(2)), 7.38 (s, 1H, NH of Aib(4)), 7.32-7.18 (m, 5H, aromatic protons of Phe), 6.37 (d, *J* = 8.2 Hz, 1H, NH of Phe), 4.88 (m, 1H, NH of Gpn(1)), 4.77-4.65 (m,1H, $C^{\alpha}H$ of Phe), 3.74 (s, 3H, OCH_3), 3.20 (d, *J* = 6.5 Hz, 2H, $C^{\gamma}Hs$ of Gpn), 3.17-2.91 (m, 2H, $C^{\beta}Hs$ of Phe), 2.03 (s, 2H, $C^{\alpha}Hs$ of Gpn), 1.54-1.51 (m, 4H), 1.51-1.48 (m, 5H), 1.47 (s, 9H), 1.45-1.40 (m, 6H), 1.33-1.28 (m, 4H), 1.20-1.11 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$ δ ppm): 175.17, 173.94, 171.90, 170.55, 157.57, 137.35, 129.54, 128.64, 126.90, 80.35, 57.05, 56.29, 53.51, 52.46, 37.60, 36.80, 34.42, 34.14, 28.49, 26.05, 25.92, 25.43, 24.62, 24.37, 21.60, 21.46. HRMS (ESI) *m/z* for $C_{32}H_{51}N_4O_7$ $(M+H)^+$ calcd.: 603.3758, found 603.3752.

Boc-Gpn(1)-Aib(2)-Leu(3)Aib(4)-OMe P2.

Peptide P**2** was obtained as white crystalline solid (0.212 g, 79 %). $[\alpha]_D^{20}$: −4.20 (*c* 1 in MeOH); FT-IR (KBr), ῡ (cm^{-1}): 3353 (s), 3307 (s), 2956 (s), 1739 (s), 1698 (s), 1653 (s), 1531 (s), 1385 (m), 1277 (ms), 1161 (ms); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 7.80 (s, 1H, NH of Aib(2)), 7.40 (s, 1H, NH of Aib(4)), 6.59 (d, *J* = 8.28 Hz, 1H, NH of Leu(3)), 4.87 (t, *J* = 6.76 Hz, 1H, NH of Gpn(1)), 4.46-4.38 (m,1H, $C^{\alpha}H$ of Leu), 3.69 (s, 3H, OCH_3), 3.21-3.03 (m, 2H, $C^{\gamma}Hs$ of Gpn), 2.20-2.05 (m, 2H, $C^{\alpha}Hs$ of Gpn), 1.92-1.83 (m, 2H, $C^{\beta}Hs$ of Leu), 1.70-1.15 (m, 32H), 0.97-0.84 (m, 6H, $C^{\delta}Hs$ of Leu); ^{13}C NMR (100 MHz, $CDCl_3$ δ ppm): 175.19, 174.21, 172.11, 171.75, 157.58, 80.41, 57.32, 56.22, 52.42, 51.81, 40.00, 37.84, 34.38, 34.31, 29.84, 28.50,

26.24, 26.06, 25.52, 25.17, 24.61, 24.56, 23.43, 21.50. HRMS (ESI) *m/z* for $C_{29}H_{53}N_4O_7$ $(M+H)^+$ calcd.: 569.3914, found 569.3902.

Boc-Gpn(1)-Aib(2)-Tyr(3)Aib(4)-OMe P3.

Peptide P**3** was obtained as white solid (0.211 g, 71 %). $[\alpha]_D^{20}$: −1.60 (*c* 1 in MeOH); FT-IR (KBr), ϑ (cm^{-1}): 3298 (bs), 2929 (m), 1728 (ms), 1699 (s), 1657 (s), 1540 (ms), 1275 (ms), 1165 (m); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 8.10 (bs, 1H, NH of Aib(2)), 7.41 (s, 1H, NH of Aib(4)), 7.02 (d, *J* = 6.5 Hz, 2H, ring protons of Tyr (3)), 6.74 (d, *J* = 6.5 Hz, 2H, ring protons of Tyr), 6.46 (d, *J* = 7.5 Hz, 1H, NH of Tyr), 5.01(bs, 1H, NH of Gpn (1)), 4.73-4.51 (m, 1H, $C^{\alpha}H$ of Tyr), 3.72 (s, 3H, OCH_3), 3.19-3.07 (m, 2H, $C^{\beta}Hs$ of Tyr), 3.05-2.89 (m, 2H, $C^{\gamma}Hs$ of Gpn), 2.01 (s, 2H, $C^{\alpha}Hs$ of Gpn), 1.55-1.47 (m, 5H), 1.47-1.45 (m, 3H), 1.44 (s, 9H, CH_3), 1.43-1.37 (m, 7H), 1.36-1.27 (m, 5H), 1.21-1.08 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$, δ ppm): 175.43, 174.27, 172.21, 170.88, 157.60, 155.53, 130.61, 128.10, 115.73, 80.26, 57.06, 56.33, 53.74, 52.58, 37.58, 36.00, 34.27, 34.18, 28.51, 26.23, 26.04, 25.46, 24.56, 24.32, 21.61, 21.48. HRMS (ESI) *m/z* for $C_{32}H_{51}N_4O_8$ $(M+H)^+$ calcd.: 619.3707, found 619.3718.

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1. W. Geibel, J. Hartenstein, W. Herrmann, J. Witzke, *US Pat.*, 5091567, 1992.

2. I. O. Lebedyeva, D. A. Ostrov, J. Neubert, P. J. Steel, K. Patel, S. M. Sileno, K. Goncalves, M. A. Ibrahim, K. A. Alamry and A. R. Katritzky, *Bioorg. Med. Chem.*, 2014, **22**, 1479-1486.

3. S. Chatterjee, P. G. Vasudev, S. Raghothama, C. Ramakrishnan, N. Shamala and P. Balaram, *J. Am. Chem. Soc.*, 2009, **131**, 5956-5965.

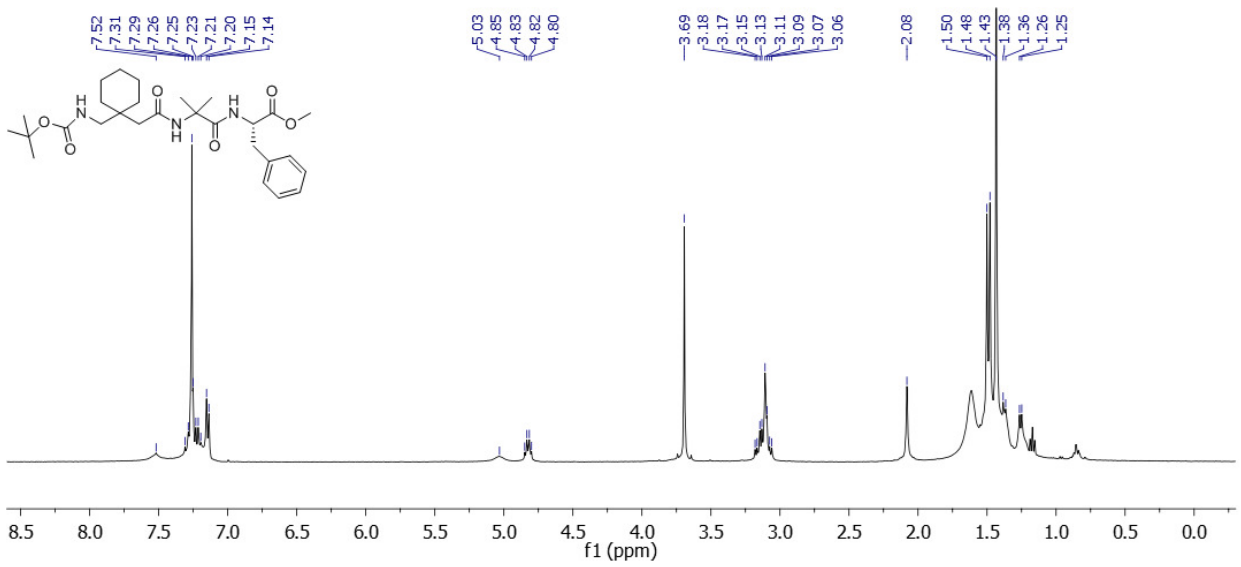

Figure S14. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-OMe **4.**

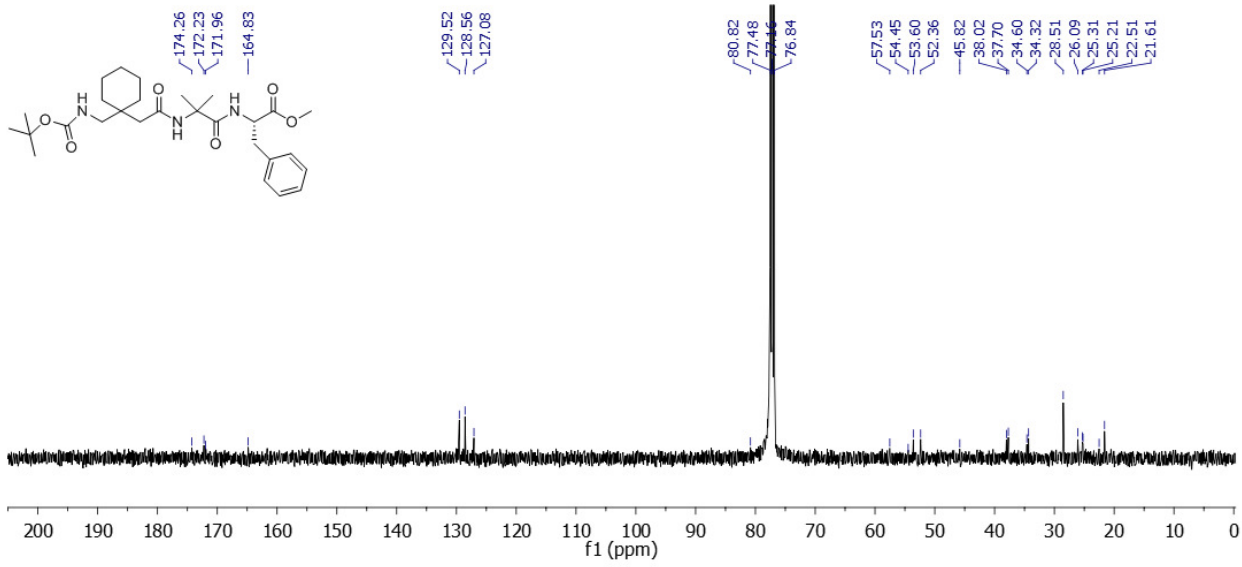

Figure S15. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-OMe **4.**

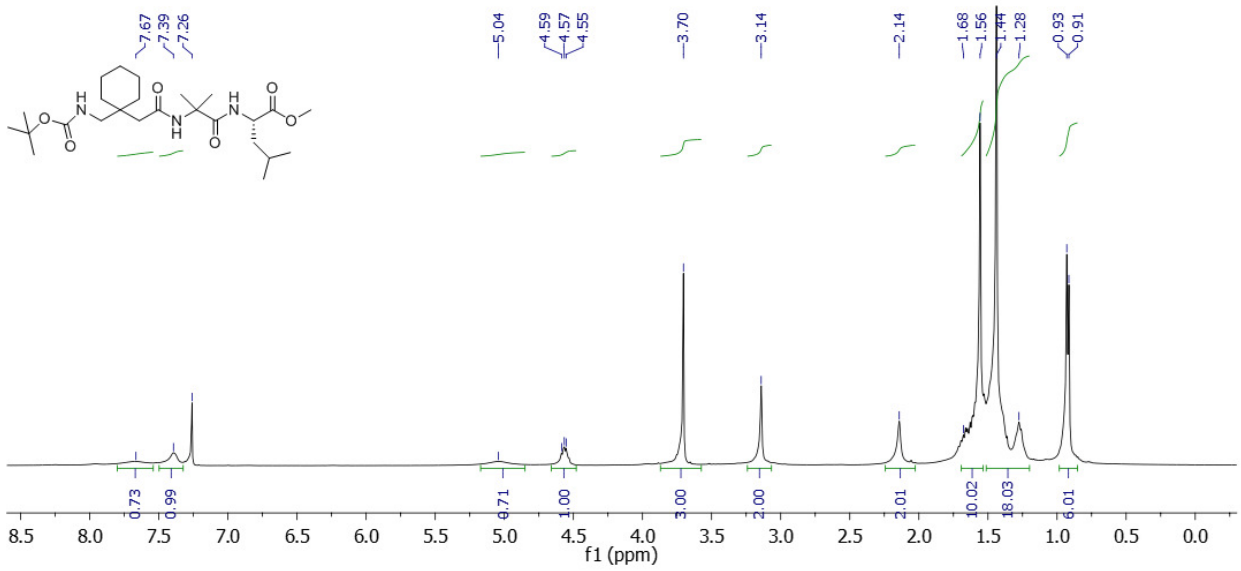

Figure S16. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-OMe **5.**

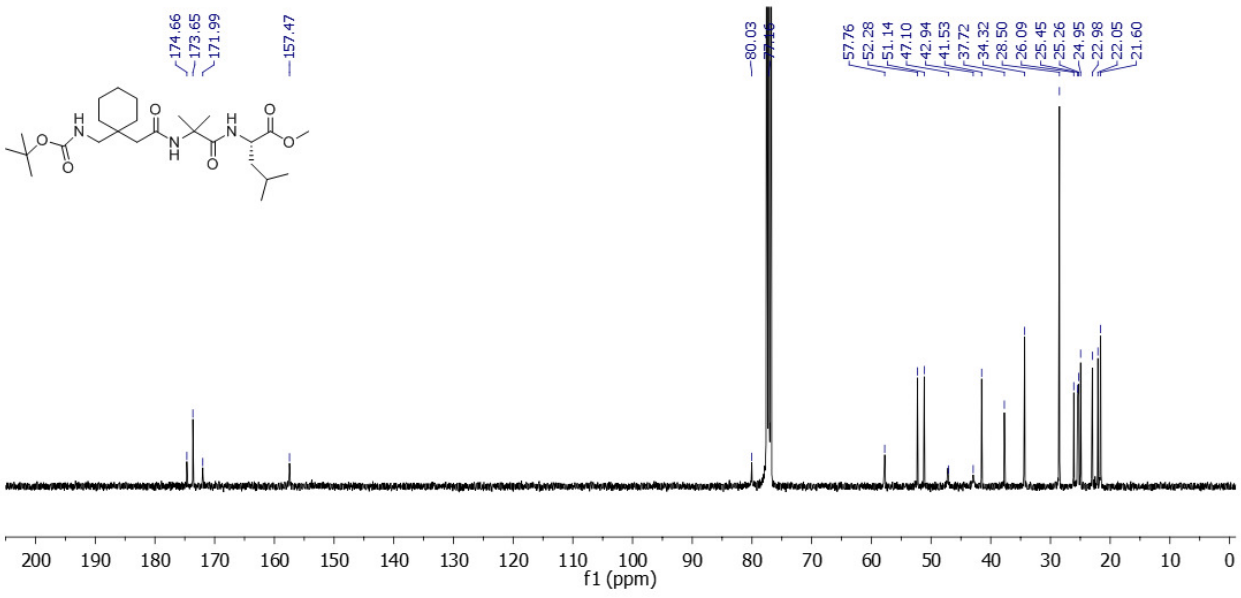

Figure S17. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-OMe **5.**

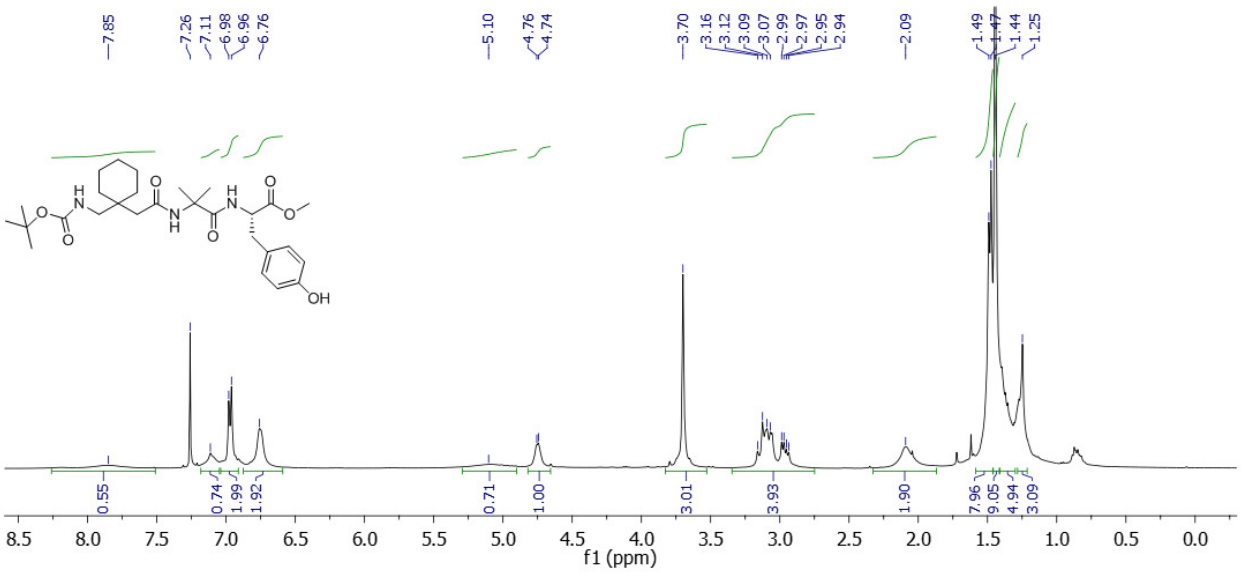

Figure S18. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-OMe **6.**

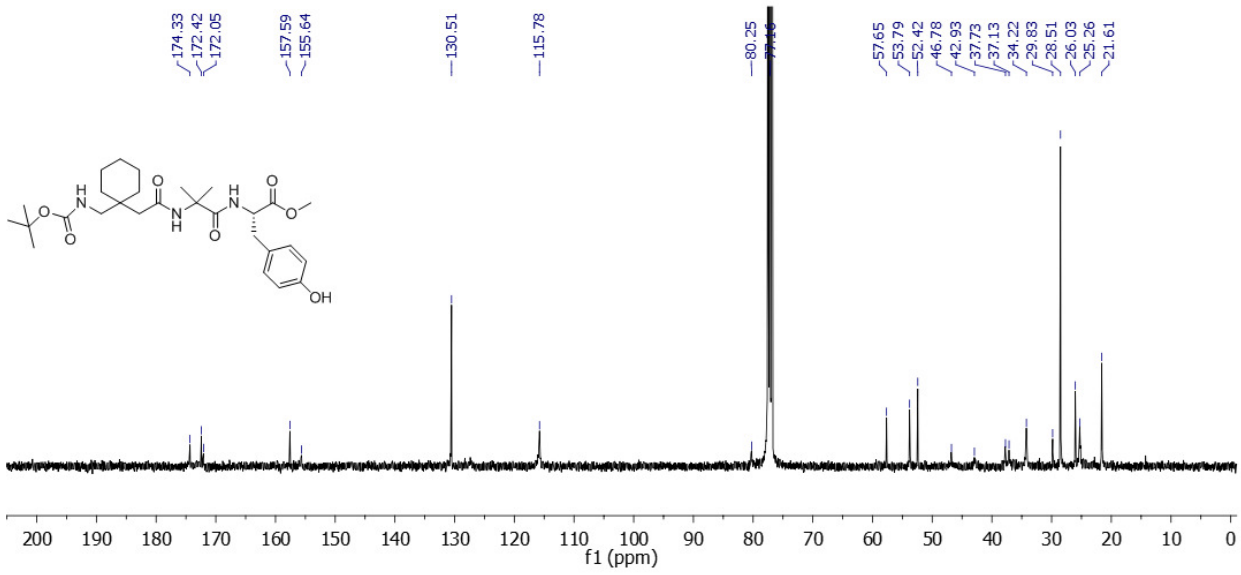

Figure S19. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-OMe **6.**

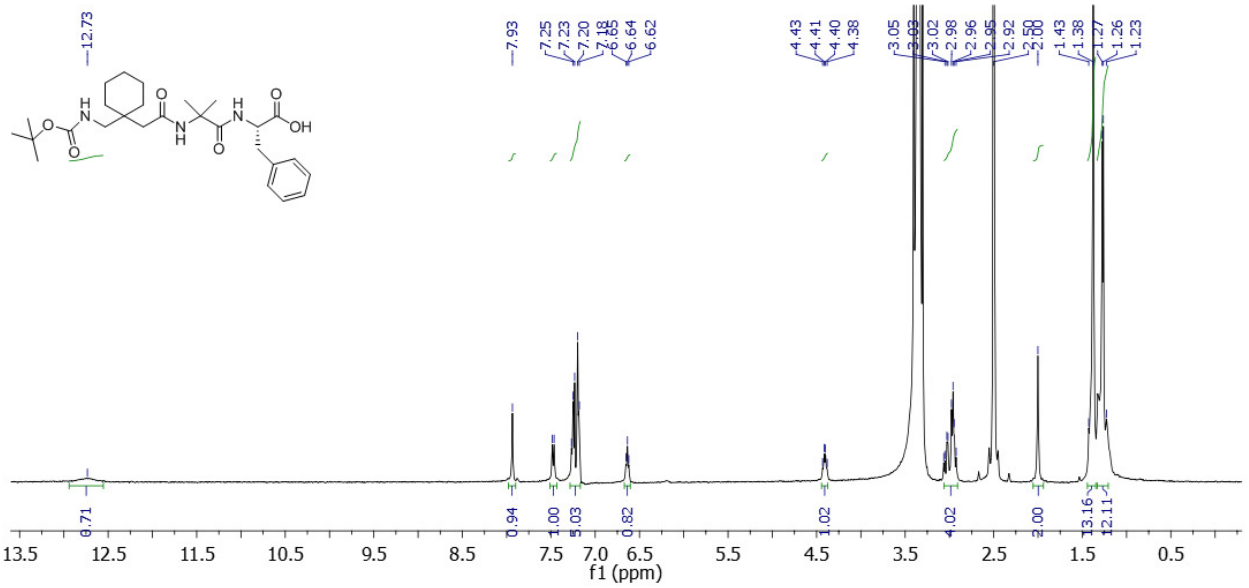

Figure S20. ^{1}H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-OH **7.**

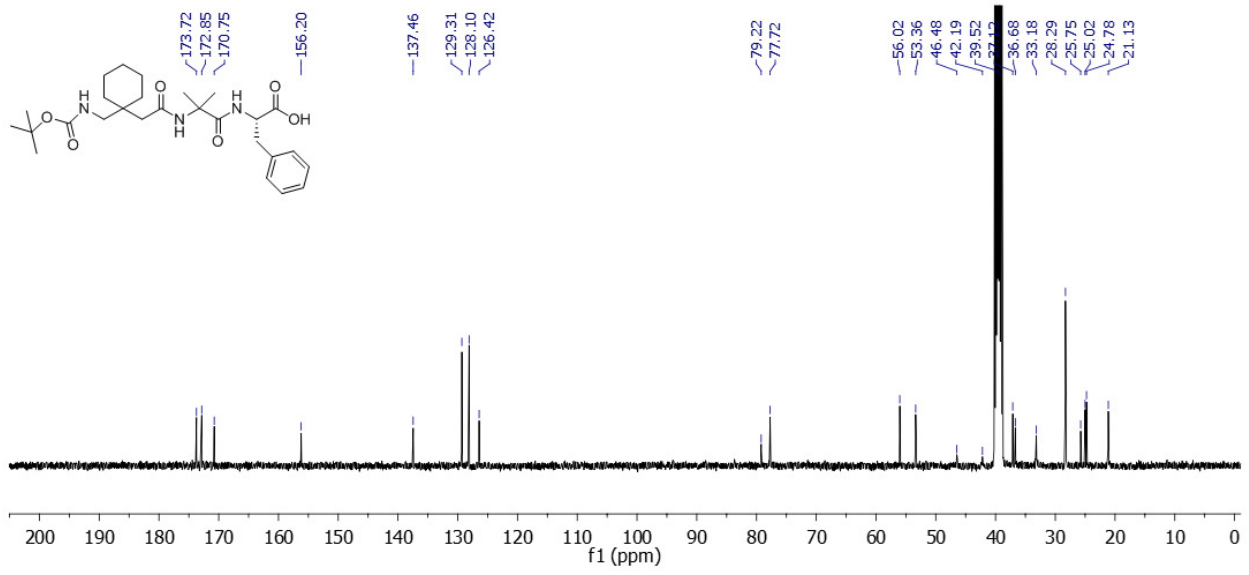

Figure S21. ^{13}C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-OH **7.**

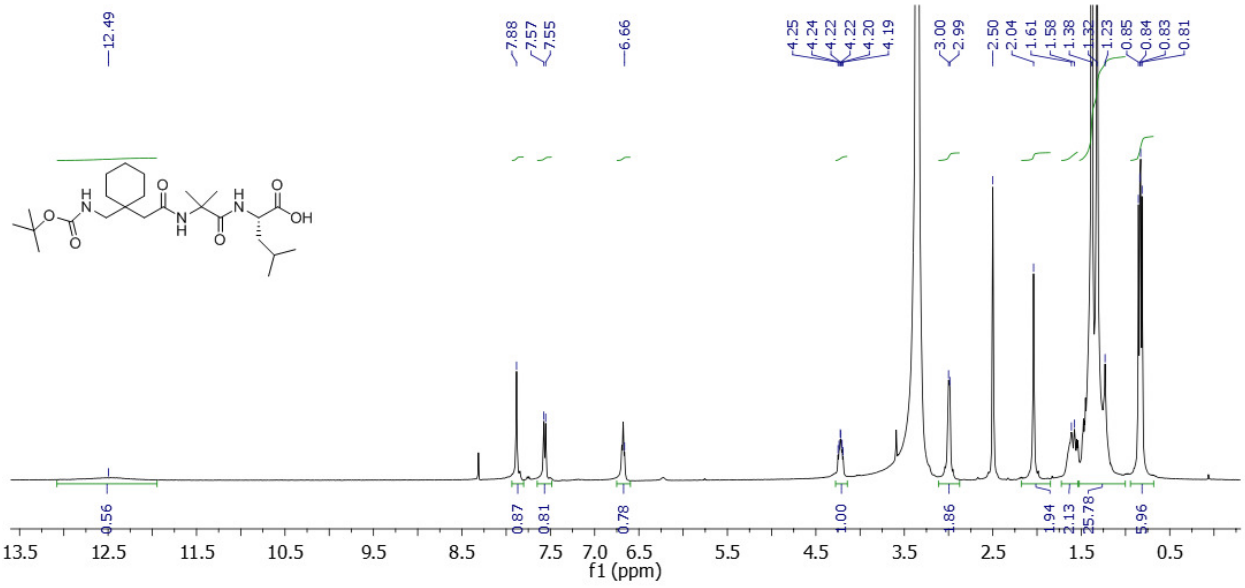

Figure S22. ^{1}H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-OH **8.**

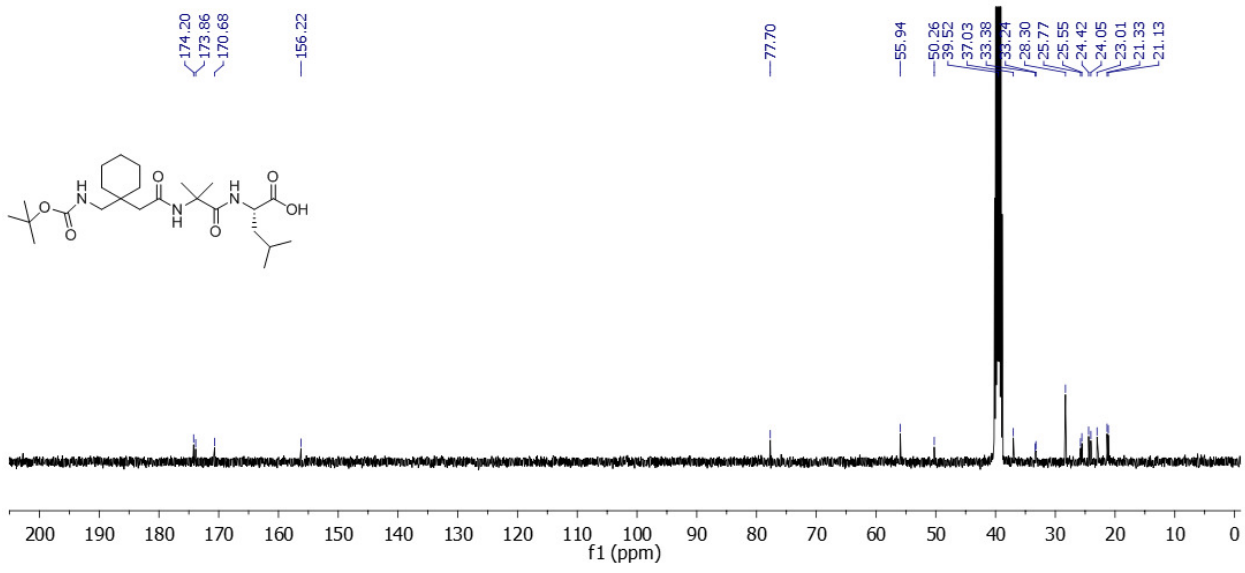

Figure S23. ^{13}C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-OH **8.**

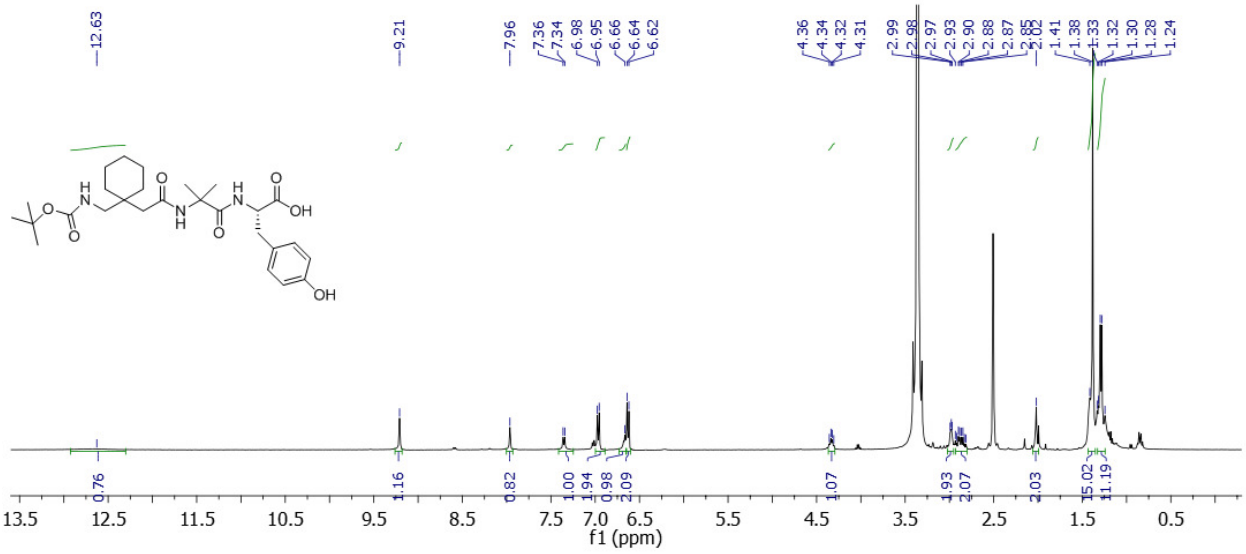

Figure S24. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-OH **9.**

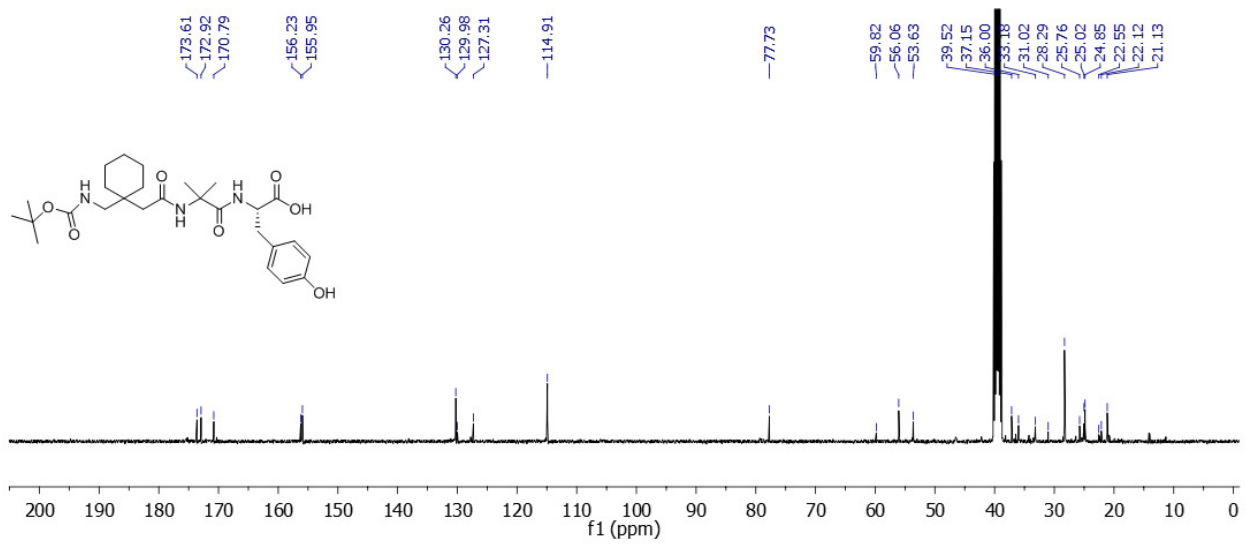

Figure S25. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-OH **9.**

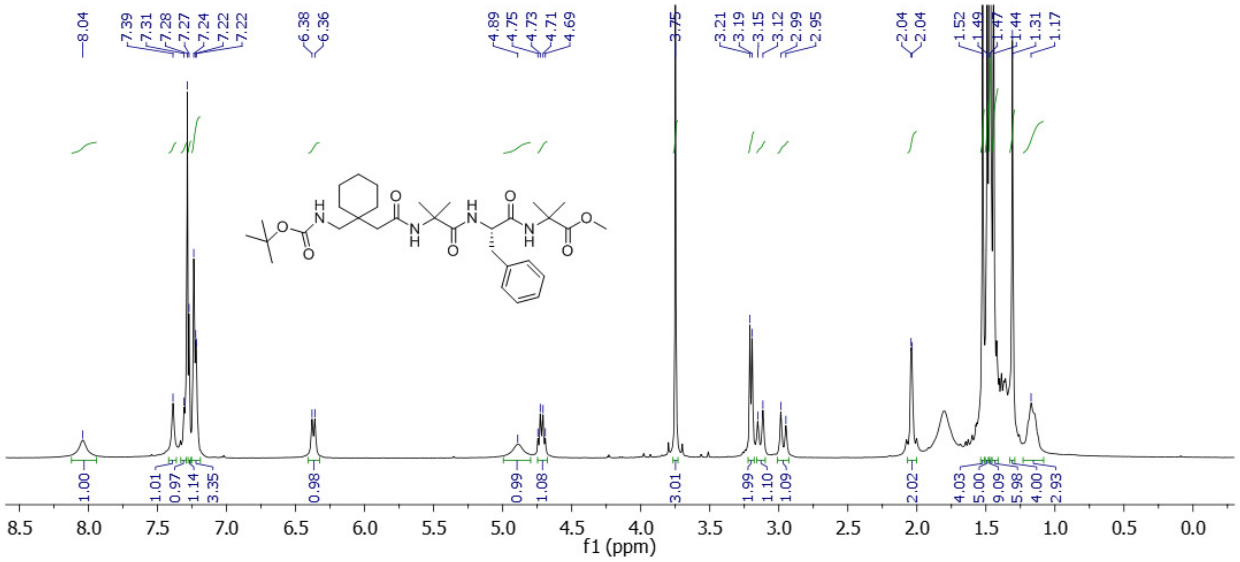

Figure S26. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-Aib(4)-OMe **P1.**

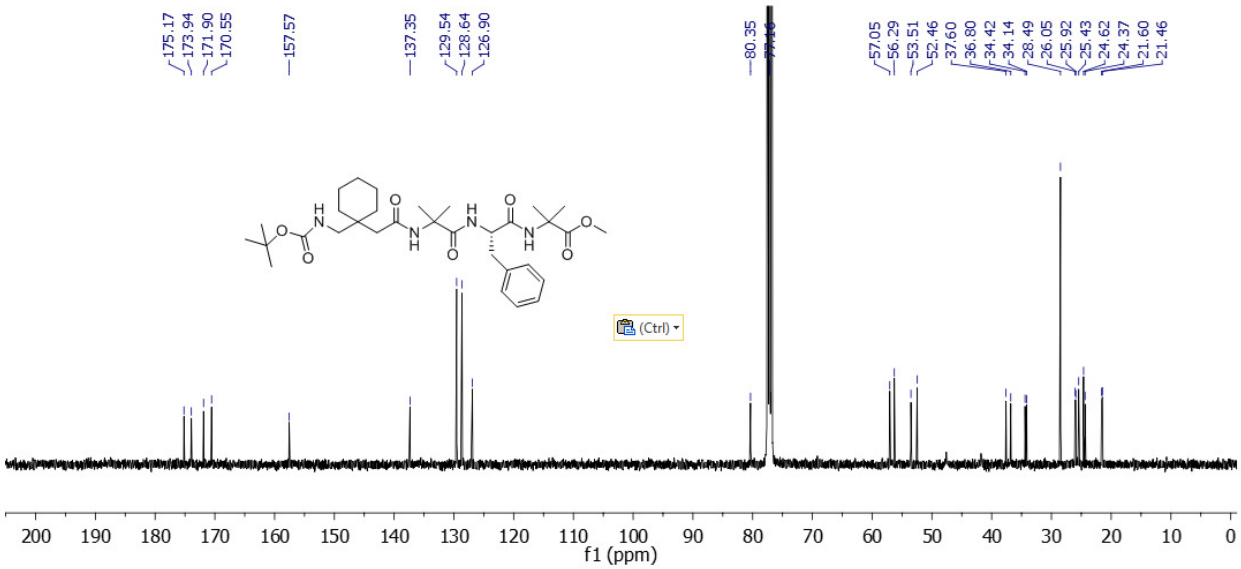

Figure S27. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-Aib(4)-OMe **P1.**

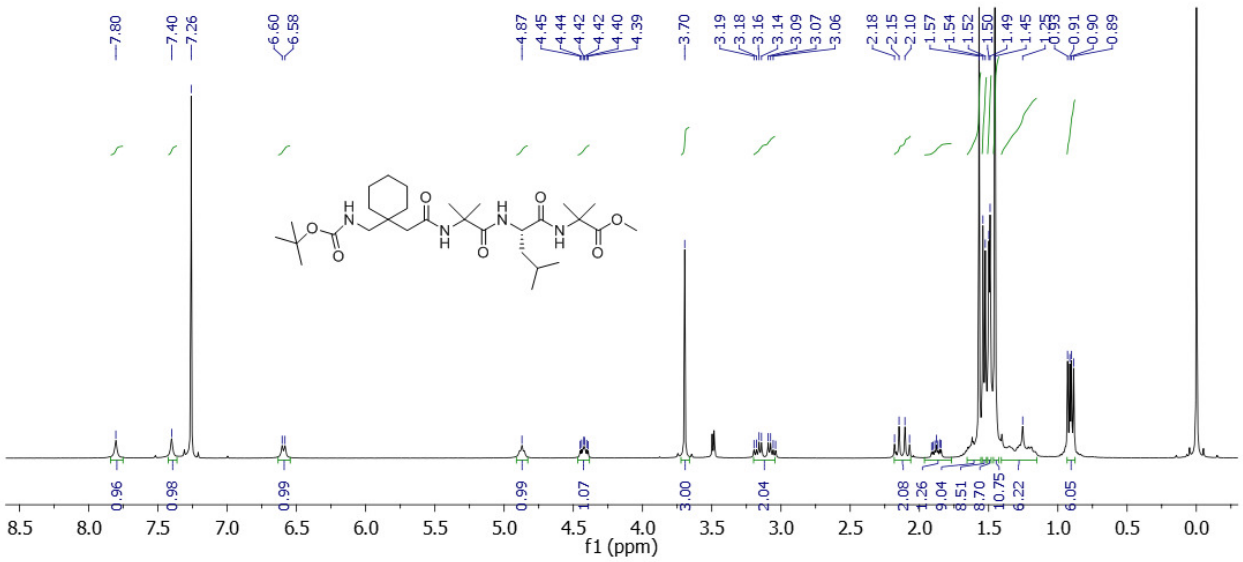

Figure S28. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-Aib(4)-OMe **P2.**

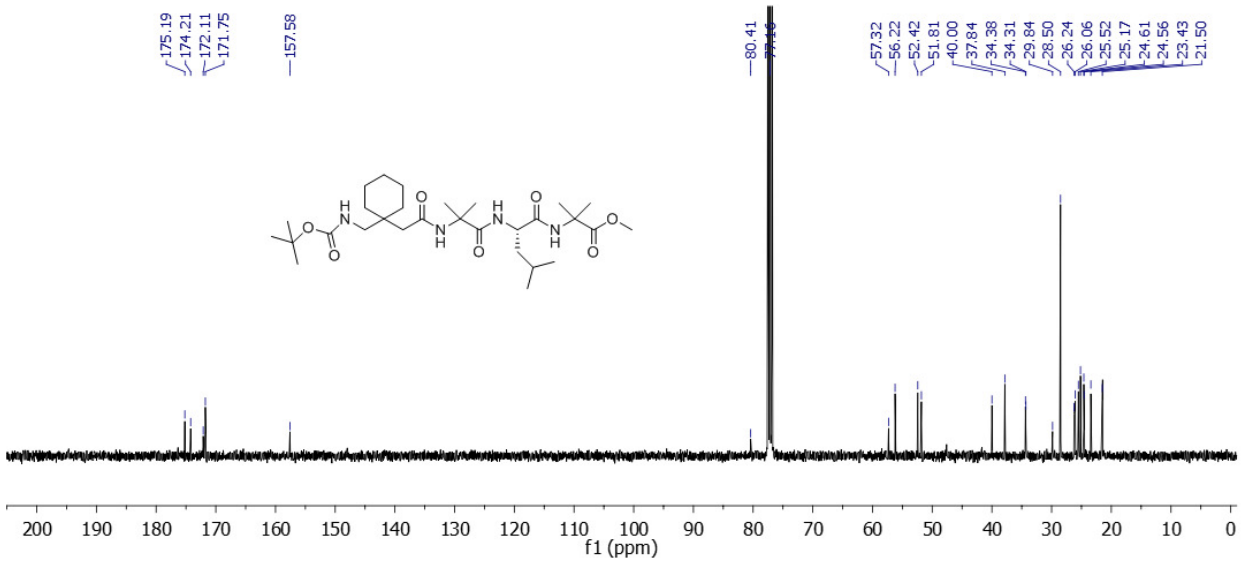

Figure S29. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)- Leu(3)-Aib(4)-OMe **P2.**

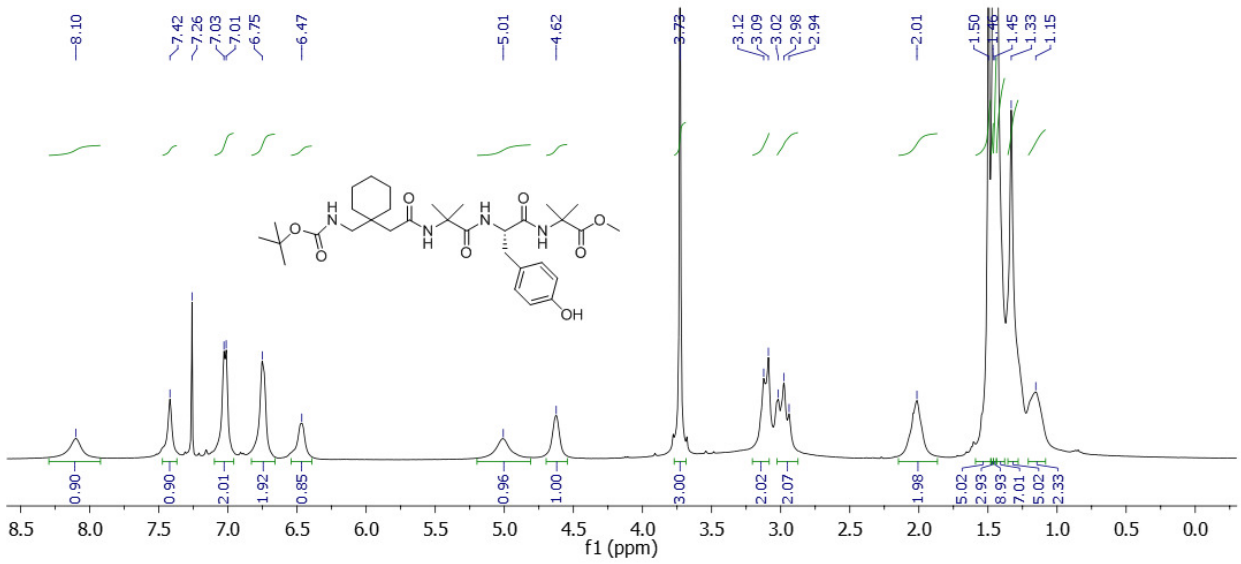

Figure S30. [1]H-NMR spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-Aib(4)-OMe **P3.**

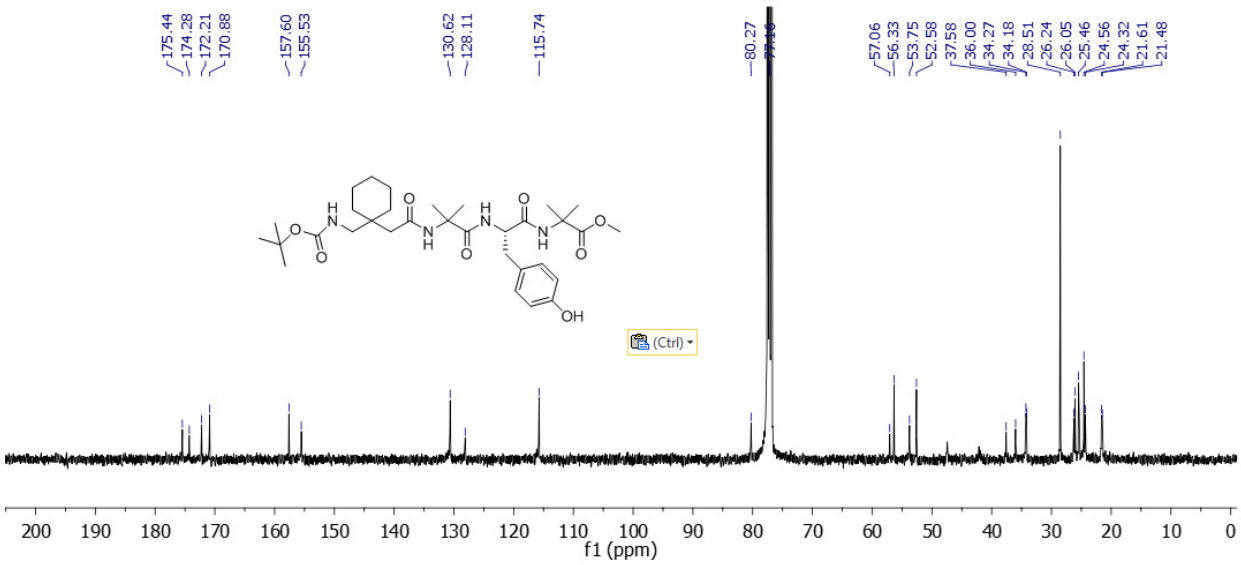

Figure S31. [13]C-NMR spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-Aib(4)-OMe **P3.**

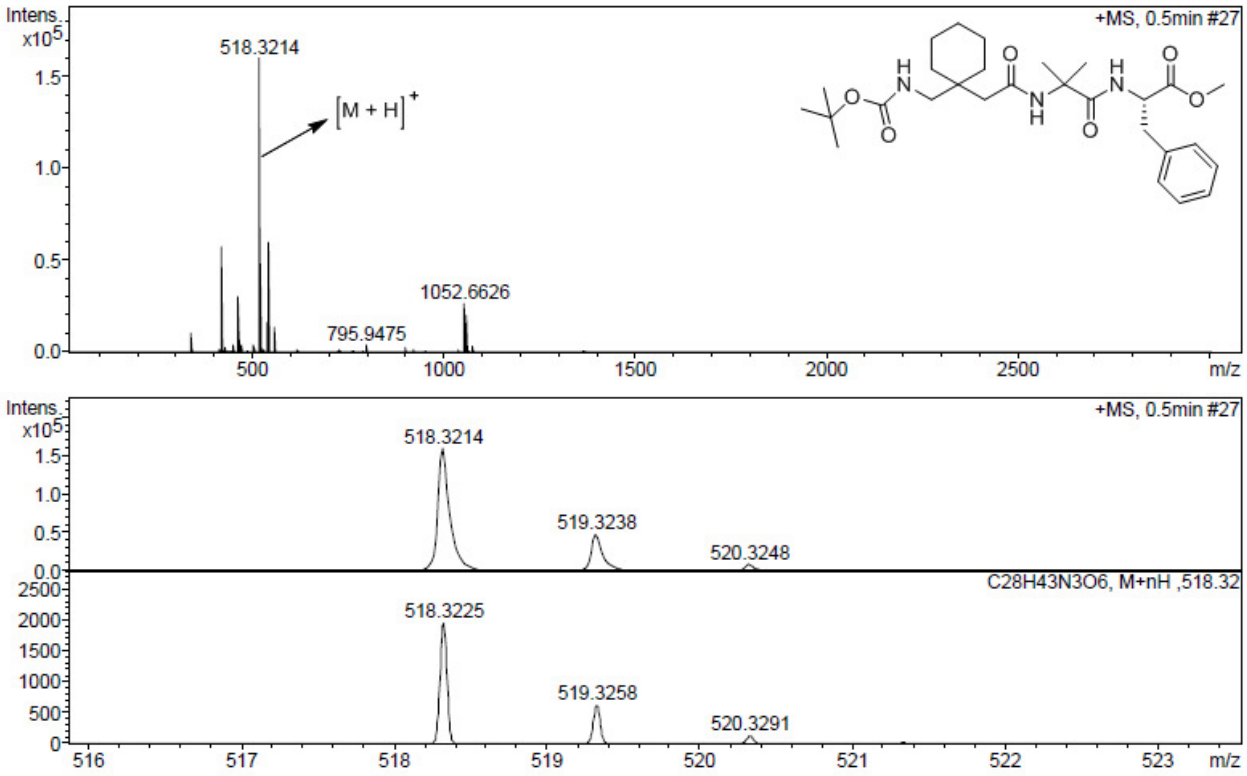

Figure S32. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-OMe **4.**

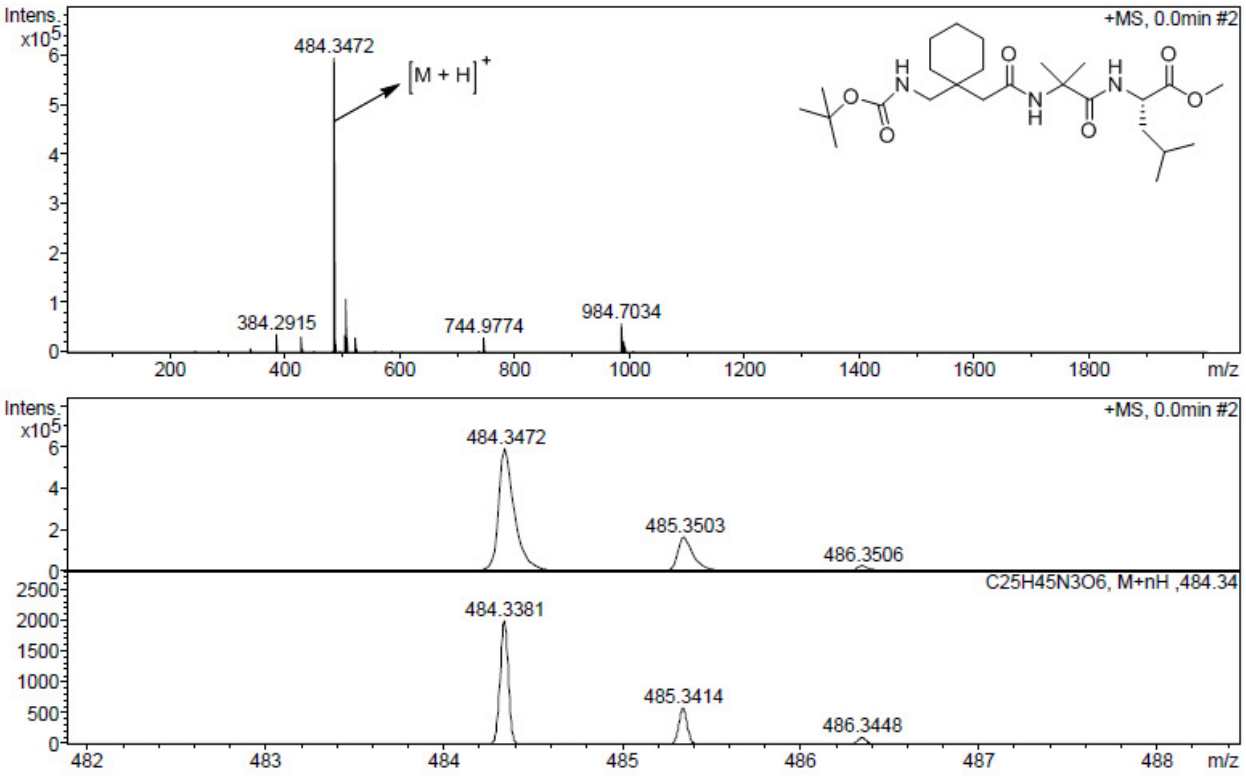

Figure S33. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-OMe **5.**

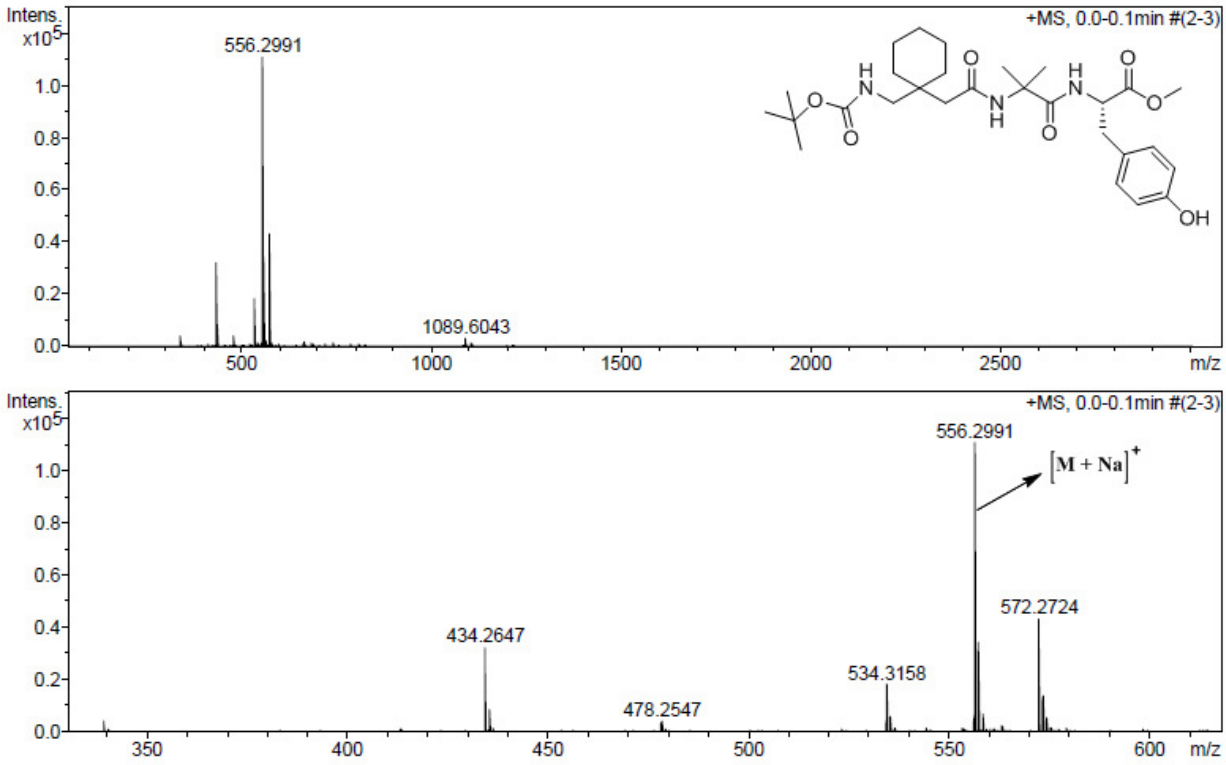

Figure S34. ESI-MS spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-OMe **6.**

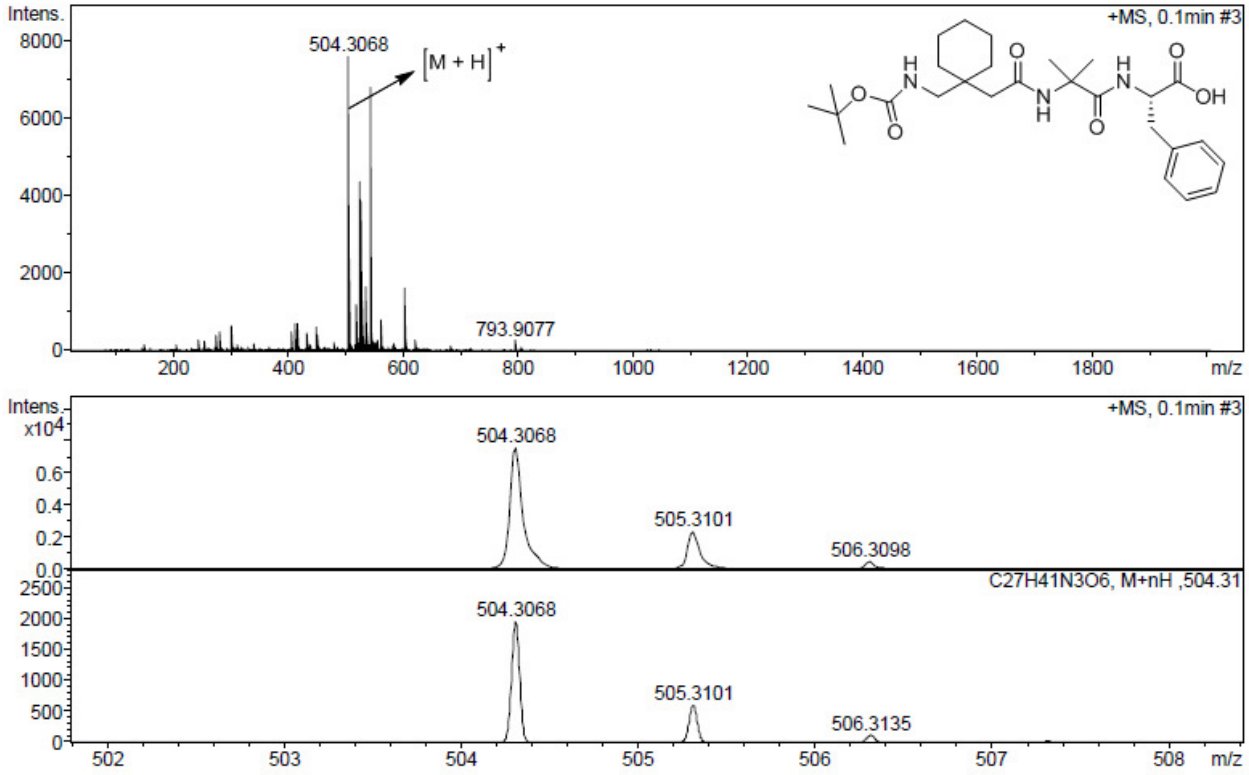

Figure S35. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-OH **7.**

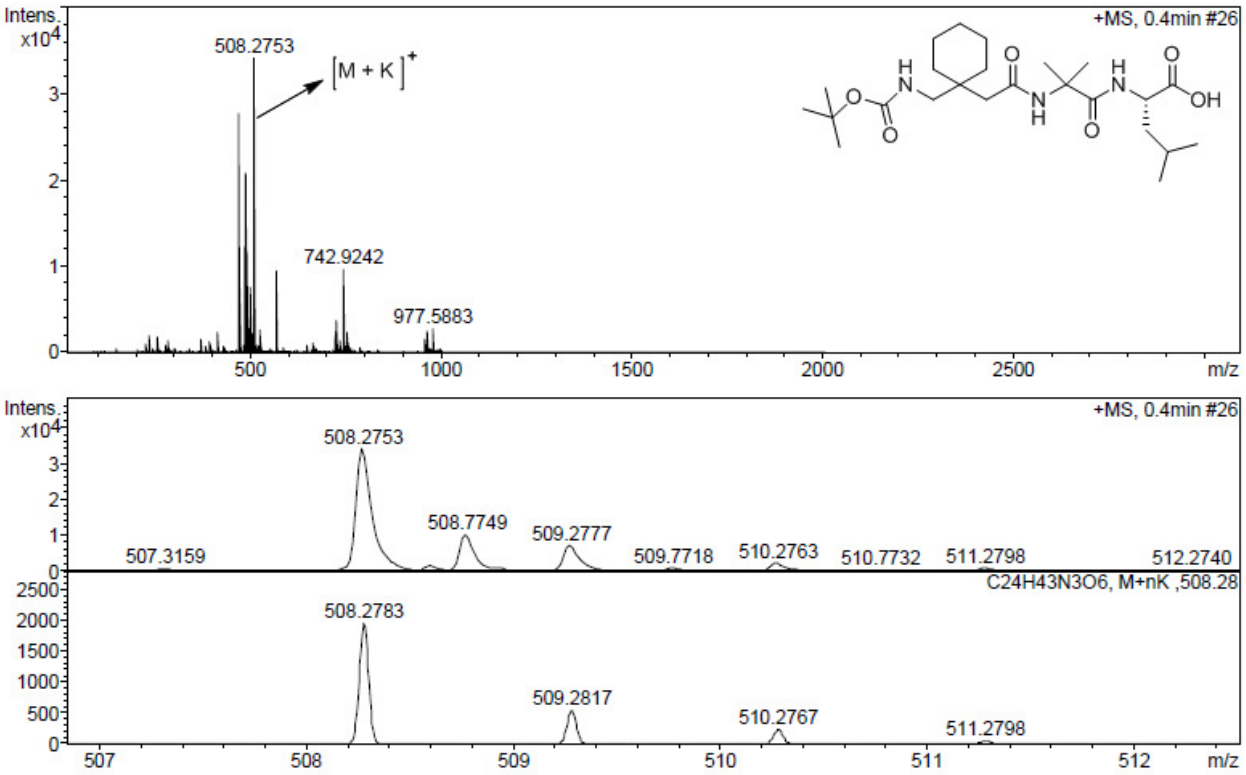

Figure S36. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-OH **8.**

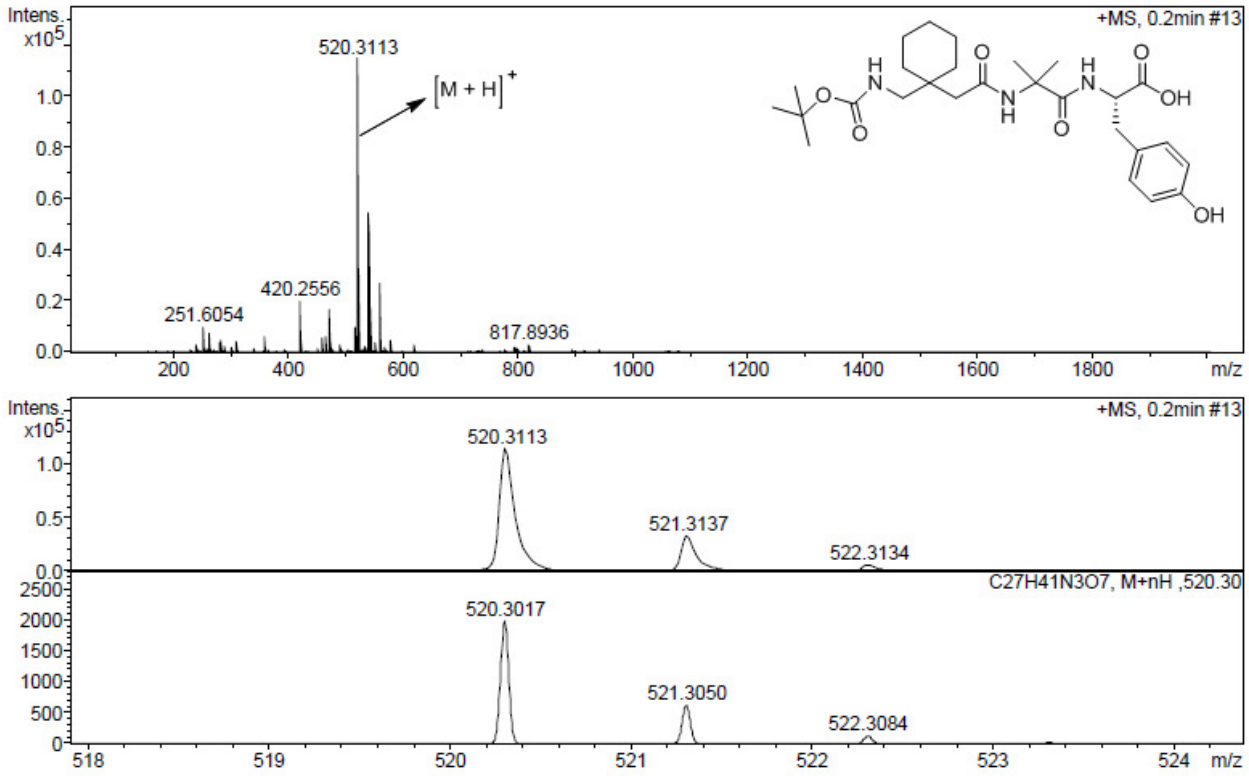

Figure S37. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-OH **9.**

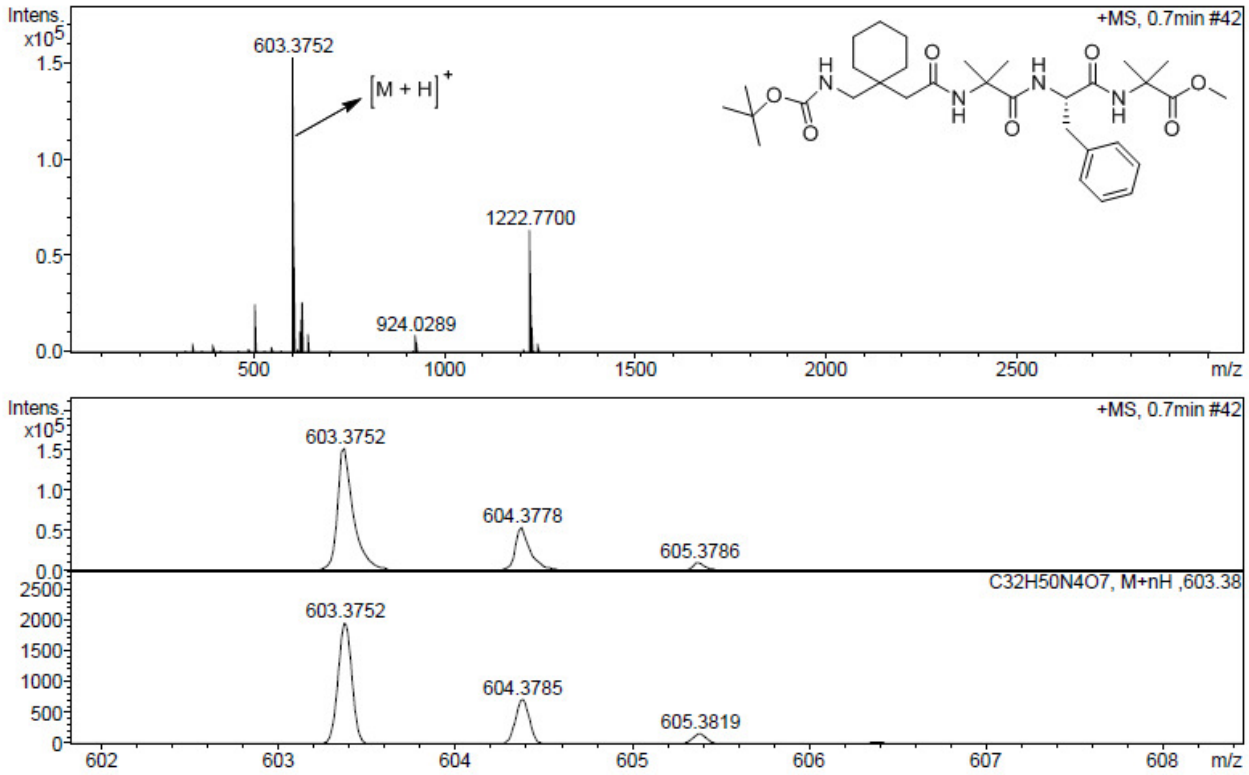

Figure S38. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Phe(3)-Aib(4)-OMe **P1.**

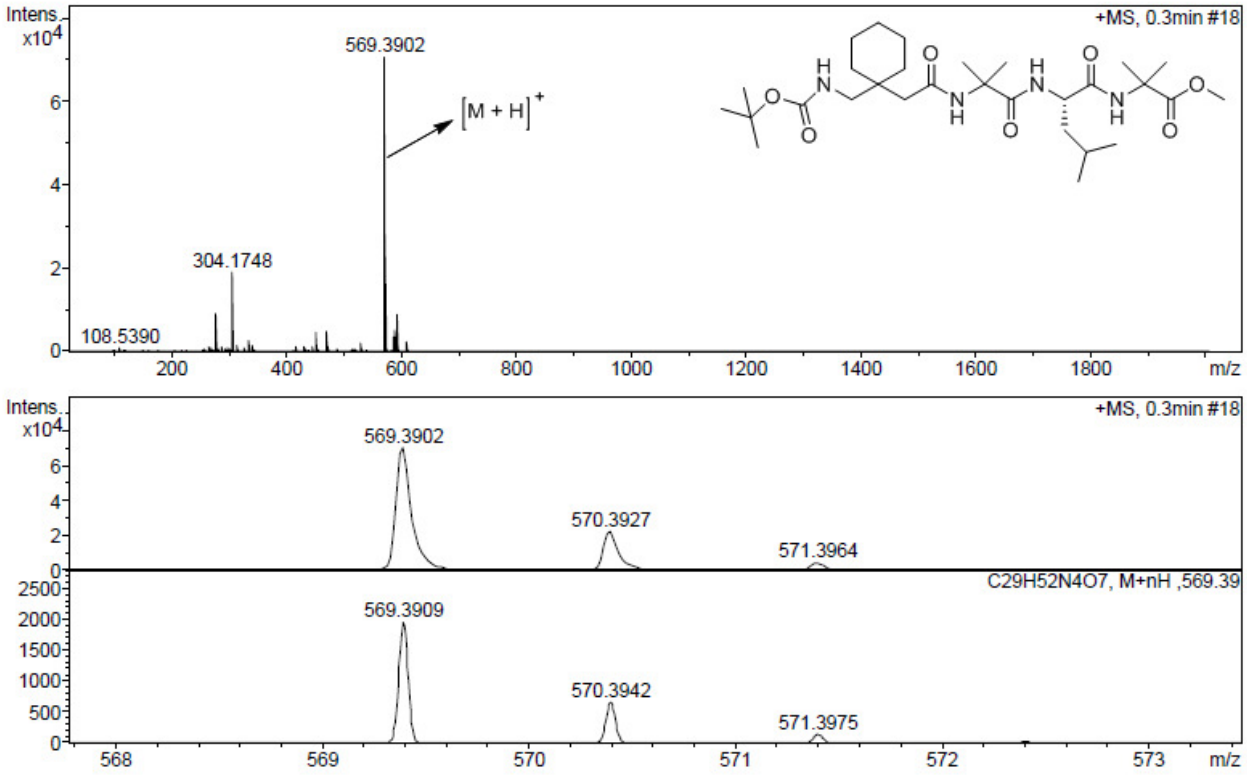

Figure S39. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Leu(3)-Aib(4)-OMe **P2.**

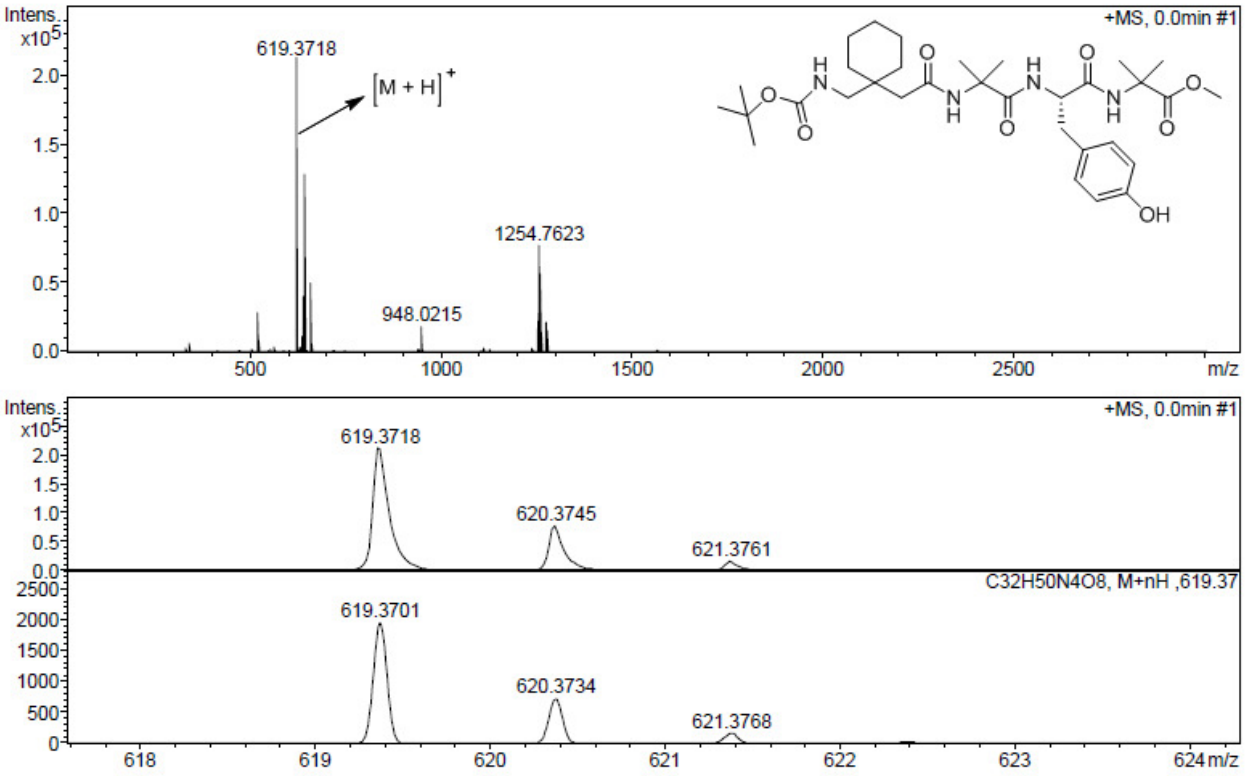

Figure S40. HRMS spectrum of Boc-Gpn(1)-Aib(2)-Tyr(3)-Aib(4)-OMe **P3.**