Supporting Information

Novel Antifouling Oligopeptides including β-amino acid

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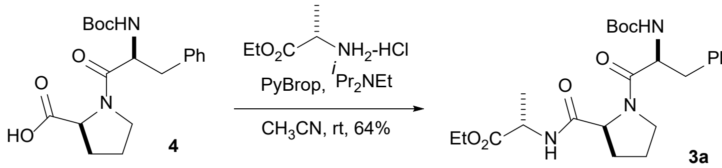
General Methods. The IR spectra were recorded on a JASCO FTIR-4100 Type A spectrometer (JASCO corporation, Tokyo, Japan) using a NaCl cell. ESI-MS were obtained on a JEOL JMS-700TZ (JEOL Ltd., Tokyo, Japan) or BRUKER DALTONICS micro TOF-HS focus spectrometer (Bruker Japan Ltd., Yokohama, Japan). Optical rotations were recorded on a HORIBA SEPA-300 polarimeter (HORIBA Ltd., Kyoto, Japan). The ^{1}H NMR and ^{13}C NMR spectra were recorded using a JNM-EX 400 (400 MHz and 100 MHz) spectrometer (JEOL Ltd., Tokyo, Japan) or JNM-ECZR 500 (500 MHz and 126 MHz) spectrometer (JEOL Ltd., Tokyo, Japan). Chemical shifts were reported in ppm relative to $CHCl_3$ in $CDCl_3$ for ^{1}H NMR (δ = 7.26) and ^{13}C NMR (δ = 77.0). Splitting patterns for ^{1}H NMR were designated as "s, d, t, q, m, dt, dd, and td". These symbols indicate "singlet, doublet, triplet, quartet, multiplet, doublettriplet, doubletdoublet, and tripletdoublet" respectively. All commercially obtained reagents were employed as received. Analytical TLC was carried out using pre-coated silica gel plates (Wako TLC Silicagel 70F_{254}, FUJIFILM Wako Pure Chemical Corporation, Osaka, Japan). Wakogel 60N 63-212 μm was used for column chromatography.

General Procedure for Synthesis of Tripeptide 3: To Boc-β-amino acid ester (1 equivalent) was added TFA/DCM (1:4 v/v, 0.10 M). After 1 h of stirring at room temperature, the mixture was concentrated *in vacuo*. The residue TFA salt was added 0.5 M NaOH aq, extracted with DCM, washed with brine, dried over Na_2SO_4, filtered, and concentrated *in vacuo* to afford crude amine, which was used in the next step without further purification.

To a solution of crude amine in MeCN (0.30 M) was added 4 N HCl in dioxane (1.0 equivalent) under Ar atmosphere. After 10 minutes of stirring at room temperature, the mixture was added **4** (1.0 equivalent) in MeCN (0.30 M), PyBrop (1.3 equivalent) and *i*Pr_2NEt (3.0 equivalent) The mixture was stirred for 24 h, quenched with diluted NaOH, extracted with EtOAc, washed with diluted HCl and brine, dried over Na_2SO_4, filtered, and concentrated *in vacuo*. The crude product was purified using column chromatography (Hexane:AcOEt = 70 : 30) to afford tripeptide **3**:

Characterization and Chemical Yield of Product



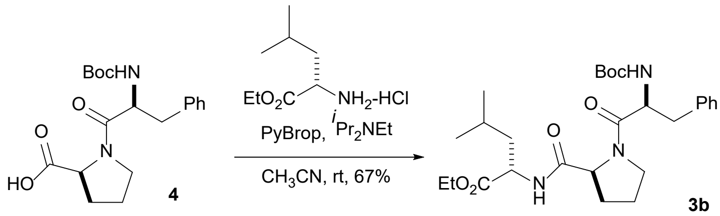
3a: $[\alpha]^{23}_{D}$ –52.2 (*c* 1.37, $CHCl_3$);

IR (neat) 3309, 2979, 2935, 2878, 1739, 1690, 1643, 1530, 1447, 1391, 1366, 1248, 1199, 1167, 1055, 1022, 862, 753, 701, 666 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$, 3:1 mixture of rotamers) δ 1.21 (3H×1/4, t, *J* = 7.1 Hz), 1.28 (3H×3/4, t, *J* = 7.2 Hz), 1.38-1.44 (12H, m), 1.70-1.84 (3H, m), 2.11-2.32 (1H, m), 2.94 (1H, dd, *J* = 12.0, 7.9 Hz), 3.04 (1H, dd, *J* = 12.0, 7.0 Hz), 3.30-3.57 (2H, m), 4.11 (2H×1/4, q, *J* = 7.1 Hz), 4.21 (2H×3/4, q, *J* = 7.1 Hz), 4.38-4.51 (2H, m), 4.54-4.68 (1H, m), 5.11 (1H×1/4, d, *J* = 5.7 Hz), 5.30 (1H×3/4, d, *J* = 8.6 Hz), 7.11-7.31 (5H, m);

^{13}C NMR (101 MHz, $CDCl_3$) δ 14.06, 14.12, 16.8, 18.1, 21.9, 25.0, 27.1, 28.2, 28.3, 30.7, 38.9, 39.4, 46.6, 47.3, 48.4, 48.5, 53.3, 54.2, 59.9, 60.6, 61.1, 61.4, 79.7, 80.1, 126.9, 127.4, 128.4, 128.9, 129.3, 129.4, 136.1, 155.1, 155.5, 170.2, 171.8, 172.3, 172.7;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{24}H_{35}N_3O_6Na$ 484.2418; Found 484.2426.



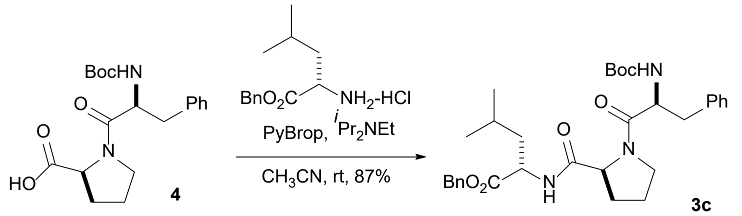
3b: $[\alpha]^{23}_{D}$ –62.9 (*c* 1.63, $CHCl_3$);

IR (neat) 3309, 2960, 2872, 1739, 1690, 1644, 1531, 1446, 1367, 1342, 1249, 1169, 1029, 920, 867, 754, 700, 666cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$, 2:1 mixture of rotamers) δ 0.85-0.98 (6H, m), 1.20 (3H×1/3, t, J = 7.3), 1.30 (3H×2/3, t, J = 7.3 Hz), 1.39 (9H, s), 1.54–2.00 (6H, m), 2.16-2.33 (1H, m), 2.86-3.10 (2H, m), 3.46-3.60 (2H, m), 4.10 (2H×1/3, q, J = 7.1 Hz), 4.19 (2H×/3, q, J = 7.2 Hz), 4.36-4.67 (3H, m), 5.07 (1H×1/3, d, J = 5.3 Hz), 5.28 (1H×2/3, d, J = 8.0 Hz), 7.09-7.31 (5H, m);

^{13}C NMR (101 MHz, $CDCl_3$) δ 14.08, 14.14, 21.2, 21.9, 22.1, 22.7, 23.0, 24.8, 25.0, 27.1, 28.2, 30.6, 38.6, 39.1, 39.5, 41.4, 46.6, 47.3, 51.1, 51.4, 53.3, 54.3, 59.9, 60.7, 60.9, 61.2, 79.7, 80.2, 126.9, 127.5, 128.5, 128.9, 129.3, 135.4, 136.2, 155.1, 155.6, 170.4, 171.0, 171.1, 172.0, 172.3, 172.7;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{27}H_{41}N_3O_6Na$ 526.2888; Found 526.2878.



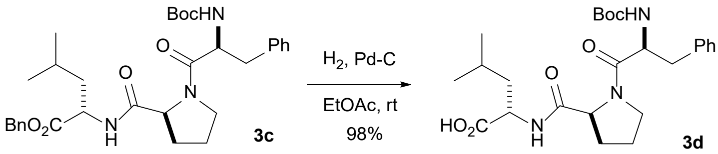
3c: $[\alpha]^{23}_{D}$ –75.1 (c 1.75, $CHCl_3$);

IR (neat) 3308, 2958, 2872, 1741, 1687, 1643, 1523, 1497, 1390, 1366, 1339, 1248, 1168, 1080, 1046, 1020, 868, 752, 699 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$, 2.5:1 mixture of rotamers) δ 0.83-0.94 (6H, m), 1.39 (9H, s), 1.55-1.97 (5H, m), 2.13-2.30 (1H, m), 2.74-3.20 (3H, m), 3.41-3.58 (2H, m), 4.19 (1H×1/3.5, dt, J = 9.6, 5.7 Hz), 4.51-4.59 (2H, m), 4.63 (1H×2.5/3.5, q, J = 7.1 Hz), 4.97-5.31 (3H, m), 7.08 (1H×2.5/3.5, d, J = 5.9 Hz), 7.18 – 7.38 (10H, m), 8.17 (1H×1/3.5, d, J = 8.3 Hz);

^{13}C NMR (101 MHz, $CDCl_3$) δ 21.0, 21.8, 21.9, 22.6, 22.9, 24.7, 25.0, 27.0, 28.2, 30.4, 38.4, 38.8, 39.4, 41.1, 46.4, 47.2, 51.1, 51.4, 53.2, 54.1, 59.8, 60.6, 66.7, 66.9, 77.2, 79.7, 80.2, 126.8, 127.4, 128.18, 128.19, 128.2, 128.4, 128.50, 128.52, 128.8, 129.30, 129.31, 135.1, 135.4, 135.7, 136.1, 155.0, 155.6, 170.4, 171.1, 171.9, 172.4;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{32}H_{43}N_3O_6Na$ 588.3044; Found 588.3041.



3d: To a solution of **3c** (12.5 mg, 0.0221 mmol) in AcOEt (0.5.0 mL) was added Pd-C (5 wt% Pd on carbon, 1.0 mg) at room temperature under Ar atmosphere. The mixture was stirred for 24 h under H_2 atmosphere, filtered through celite pad, and concentrated *in vacuo* to give **3d** (10.9 mg, 0.0216 mmol, 98%).

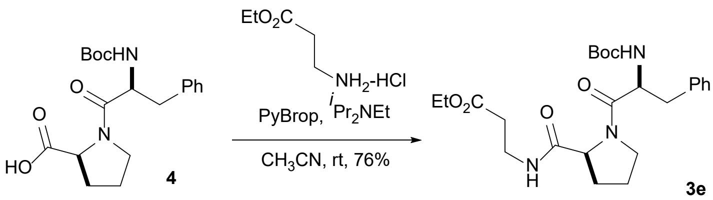
$[\alpha]^{25}_D$ –34.6 (*c* 1.20, $CHCl_3$);

IR (neat) 3309, 2962, 2933, 2872, 1681, 1629 1452, 1392, 1367, 1246, 1166, 1100, 1052, 1002, 875, 754, 701 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$) δ 0.86-0.93 (9H, m), 1.46 (9H, s), 1.62-1.67 (2H, m), 1.81-2.07 (5H, m), 2.16-2.21 (1H, m), 2.45 (1H, dd, J = 13.3, 7.0 Hz), 2.72 (1H, dd, J = 13.3, 7.4 Hz), 3.15-3.19 (1H, m), 3.28-3.34 (1H, m), 4.43-4.59 (3H, m), 5.45 (1H, d, J = 9.3 Hz), 7.17-7.34 (6H, m);

^{13}C NMR (101 MHz, $CDCl_3$) δ 14.18, 14.25, 21.0, 21.8, 22.8, 24.6, 25.1, 27.5, 28.3, 38.4, 40.1, 41.8, 47.1, 54.0, 60.0, 71.1, 79.8, 81.2, 118.1, 126.2, 128.4, 129.3, 140.0, 156.0, 171.1;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{28}H_{43}N_3O_6Na$ 526.2888; Found 526.2892.



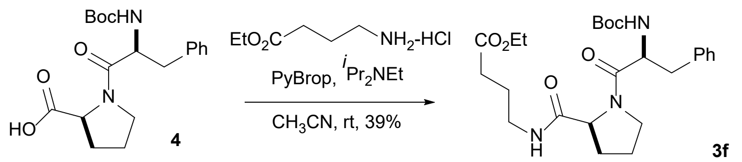
3e: $[\alpha]^{23}_{D}$ –56.6 (*c* 2.29, $CHCl_3$);

IR (neat) 3316, 3062, 2978, 1731, 1710, 1644, 1525, 1445, 1367, 1249, 1172, 1057, 1027, 917, 863, 753, 701, 666 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$, 3:1 mixture of rotamers) δ 1.23 (3H×1/4, t, *J* = 7.2), 1.26 (3H×3/4, t, 7.1 Hz), 1.41 (9H, s), 1.80-1.92 (3H, m), 2.51 (2H, t, *J* = 6.5 Hz), 2.91-3.05 (3H, m), 3.40-3.57 (4H, m), 4.15 (2H, q, *J* = 7.2 Hz), 4.49-4.51 (1H, m), 4.65 (1H, q, *J* = 7.6 Hz), 5.19 (1H×1/4, d, *J* = 7.0 Hz), 5.29 (1H×3/4, d, *J* =8.5 Hz), 7.19-7.30 (5H, m);

^{13}C NMR (101 MHz, $CDCl_3$) δ 14.2, 21.9, 24.9, 27.2, 28.3, 30.8, 33.7, 34.1, 35.0, 35.5, 39.4, 46.5, 47.2, 53.2, 54.1, 59.9, 60.5, 60.6, 60.8, 79.8, 80.2, 127.0, 127.4, 128.4, 128.8, 129.3, 129.3, 136.1, 155.0, 170.5, 170.5, 171.8, 172.0;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{24}H_{35}N_3O_6Na$ 484.2418; Found 484.2412.



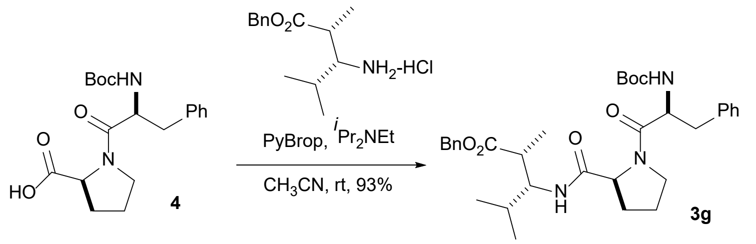
3f: $[\alpha]^{23}_{D}$ –49.7 (*c* 1.83, $CHCl_3$);

IR (neat) 3317, 2977, 2934, 2877, 1731, 1711, 1644, 1530, 1497, 1445, 1367, 1321, 1249, 1171, 1081, 1028, 865, 753, 701, 666 cm^{-1};

^{1}H NMR (400 MHz, $CDCl_3$, 4:1 mixture of rotamers) δ 1.25 (3H, t, *J* = 7.1 Hz), 1.41 (9H, s), 1.79-1.90 (4H, m), 2.27-2.35 (2H, m), 2.91-3.07 (3H, m), 3.13-3.34 (2H, m), 3.43-3.58 (2H, m), 4.11 (2H, q, *J* = 7.1 Hz), 4.50-4.52 (1H, m), 4.67 (1H, q, *J* = 7.8 Hz), 5.14 (1H×1/5, d, *J* = 6.5 Hz), 5.26 (1H×4/5, d, *J* = 8.0), 7.18-7.33 (5H, m);

^{13}C NMR (101 MHz, $CDCl_3$) δ 14.2, 21.9, 24.3, 24.8, 24.9, 27.1, 28.3, 30.6, 31.6, 31.8, 38.7, 39.1, 39.3, 39.4, 46.4, 47.3, 53.2, 54.1, 60.0, 60.38, 60.44, 60.9, 79.9, 80.3, 127.1, 127.5, 128.5, 128.9, 129.3, 135.4, 136.0, 155.0, 155.5, 170.5, 171.2, 171.9, 173.1, 173.2;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{25}H_{37}N_3O_6Na$ 498.2575; Found 498.2582.



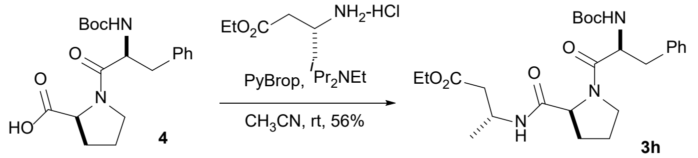
3g: $[\alpha]^{23}_{D}$ –2.0 (*c* 0.99, $CHCl_3$);

IR (neat) 3416, 3316, 2973, 2876, 1714, 1651, 1515, 1455, 1391, 1366, 1247, 1170, 1048, 1026, 868, 752, 699 cm^{-1};

^1H NMR (400 MHz, $CDCl_3$, 5:1 mixture of rotamers) δ 0.80 (6H, m), 1.10 (3H×1/6, d, *J* = 7.1 Hz), 1.22 (3H×5/6, d, *J* = 7.1 Hz), 1.34 (9H, s), 1.60-1.65 (1H, m), 1.97-2.19 (3H, m), 2.80-2.89 (3H, m), 3.15 (1H, dd, *J* = 13.9, 5.0 Hz), 3.41-3.52 (1H, m), 3.67-3.80 (2H, m), 4.53-4.57 (1H, m), 4.69 (1H, 1, *J* = 8.5 Hz), 5.03-5.12 (2H, m), 5.24 (1H×5/6, d, *J* = 8.8 Hz), 5.39 (1H×1/6, d, *J* = 7.4 Hz), 6.80 (1H×1/6, d, *J* = 9.1 Hz), 6.87 (1H×5/6, d, *J* = 9.8 Hz), 7.23-7.30 (10H, m);

^{13}C NMR (101 MHz, $CDCl_3$) δ 15.8, 19.3, 19.8, 21.8, 24.9, 28.2, 28.3, 28.8, 30.9, 31.4, 31.7, 38.8, 40.0, 40.1, 46.1, 47.2, 52.9, 56.9, 57.2, 60.6, 66.3, 79.4, 126.6, 127.1, 128.1, <u>128.21, 128.28,</u> 128.3, <u>128.52, 128.57</u>, <u>129.41, 129.49</u>, 135.5, 136.0, 136.5, 155.1, 171.3, 171.5, 175.7;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{33}H_{45}N_3O_6Na$ 602.3201; Found 602.3195.



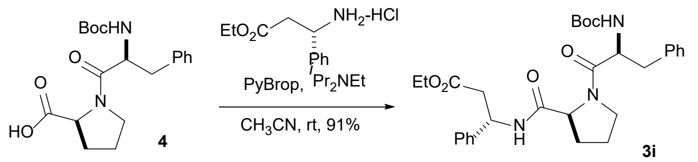
3h: $[\alpha]^{17}_{D}$ –33.9 (*c* 0.53, $CHCl_3$);

IR (neat) 3310, 2977, 2934, 1732, 1709, 1644, 1525, 1498, 1446, 1391, 1367, 1295, 1249, 1170, 1095, 1028, 918, 868, 752, 702 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) δ 1.17 (3H, m), 1.42 (9H, s), 1.85-2.07 (4H, m), 2.26-2.30 (1H, m), 2.39-2.54 (2H, m), 2.86-2.93 (2H, m), 2.99 (1H×1/2, dd, *J* = 12.5, 5.9 Hz), 3.10 (1H×1/2, dd, *J* = 12.9, 6.6 Hz), 3.17-3.22 (1H, m), 3.41-3.59 (2H, m), 4.06-4.15 (2H, m), 4.25-4.34 (1H, m), 4.48-4.50 (1H, m), 4.65 (1H, q, *J* = 6.9 Hz), 5.23 (1H×1/2, d, *J* = 7.1 Hz), 5.27 (1H×1/2, d, *J* = 8.6 Hz), 7.02 (1H, d, *J* = 7.8 Hz), 7.19-7.33 (5H, m);

^{13}C NMR (126 MHz, $CDCl_3$) δ 14.1, 19.7, 19.8, 20.0, 21.7, 25.0, 27.4, 28.2, 28.3, 30.7, 37.3, 39.1, 39.5, 40.3, 40.4, 42.3, 42.7, 46.3, 47.3, 53.2, 60.2, 60.5, 60.5, 60.9, 63.3, 76.7, 77.0, 77.3, 79.7, 80.0, 126.9, 127.4, 128.5, 128.8, 128.9, 129.3, 129.3, 135.4, 135.5, 136.3, 155.1, 155.2, 169.5, 169.9, 171.2, 171.2, 171.3, 171.8, 177.8;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{25}H_{37}N_3O_6Na$ 498.2575; Found 498.2569.



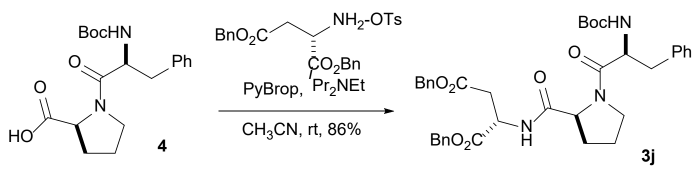
3i: $[\alpha]^{23}_{D}$ −104.4 (*c* 0.80, $CHCl_3$);

IR (neat) 3307, 2978, 2932, 1733, 1684, 1644, 1521, 1497, 1446, 1367, 1249, 1167, 1027, 865, 754, 701 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$, 4:1 mixture of rotamers) δ 1.14 (3H×4/5, d, *J* = 7.0 Hz), 1.17 (3H×1/5, d, *J* = 7.0 Hz), 1.38 (s, 9H), 1.82-2.31 (m, 4H), 2.76-3.63 (m, 6H), 4.01-4.08 (m, 2H), 4.28-4.33 (1H×1/5,m), 4.55 (1H, d, *J* = 7.5 Hz), 4.70 (1H×4/5, q, *J* = 7.3 Hz), 5.24-5.39 (m, 2H), 7.15-7.35 (m, 10H), 7.63 (1H×4/5, d, *J* = 8.0 Hz), 8.11 (1H×1/5, d, *J* = 7.7 Hz);

^{13}C NMR (126 MHz, $CDCl_3$) δ 13.99, 14.04, 21.7, 25.0, 27.2, 28.2, 28.3, 30.2, 38.9, 39.1, 40.2, 40.5, 46.2, 47.3, 49.9, 51.0, 53.2, 54.2, 60.2, 60.5, 60.6, 60.9, 79.7, 80.4, 126.4, 126.6, 126.9, 127.3, 127.4, 127.5, 128.5, 128.5, 128.6, 128.9, 129.3, 129.3, 135.4, 136.3, 140.5, 141.3, 155.1, 155.8, 169.7, 169.9, 170.6, 170.7, 170.8, 172.1;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{30}H_{39}N_3O_6Na$ 560.2731; Found 560.2738.



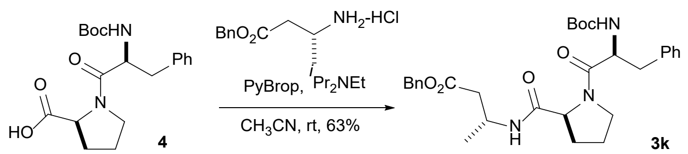
3j: $[\alpha]^{18}{}_{D}$ –21.0 (*c* 0.50, $CHCl_3$);

IR (neat) 3309, 2962, 2933, 2872, 1681, 1629 1452, 1392, 1367, 1246, 1166, 1100, 1052, 1002, 875, 754, 701 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) δ 1.37 (9H, s), 1.84-1.92 (2H, m), 2.17-2.22 (1H, m), 2.82-3.14 (5H, m), 3.30-3.37 (1H, m), 3.51-3.57 (1H, m), 4.48-4.64 (2H, m), 4.86 (1H, dt, *J* = 8.1, 4.7 Hz), 4.95-5.13 (4H, m), 5.23 (1H, d, *J* = 8.8 Hz), 7.09 (1H, d, *J* = 7.5 Hz), 7.18-7.34 (15H, m);

^{13}C NMR (126 MHz, $CDCl_3$) δ 21.9, 24.9, 27.6, 28.3, 30.8, 36.2, 39.2, 39.3, 46.5, 47.2, 48.9, 49.2, 53.2, 60.0, 60.5, 61.1, 61.9, 66.8, 67.5, 79.7, 126.8, 127.4, 128.2, 128.2, 128.3, 128.4, 128.4, 128.5, 128.6, 128.8, 129.3, 129.4, 133.6, 135.1, 135.3, 136.3, 155.1, 170.3, 170.4, 170.6, 170.8, 171.5, 171.7, 177.0;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{37}H_{43}N_3O_8Na$ 680.2942; Found 680.2927.



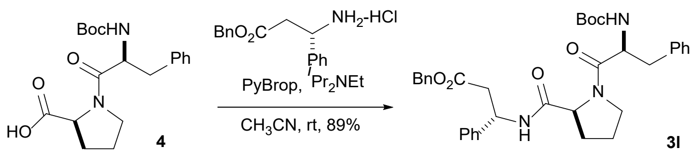
3k: $[\alpha]^{18}{}_{D}$ –28.1 (*c* 0.48, $CHCl_3$);

IR (neat) 3310, 3029, 2975, 2878, 1733, 1707, 1640, 1524, 1498, 1447, 1390, 1365, 1291, 1248, 1171, 1082, 999, 867, 751, 699 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$, 1:1 mixture of rotamers) δ 1.18 (3H×1/2, d, J = 6.7 Hz), 1.22 (3H×1/2, d, J = 6.7 Hz), 1.38 (9H×1/2, s), 1.41 (9H×1/2, s),1.82-2.08 (3H, m), 2.25-2.29 (1H, m), 2.45-2.60 (2H, m), 2.85-3.20 (2H, m), 3.36-3.60 (2H, m), 4.23-4.38 (1H, m), 4.47 (1H, d, J = 7.8 Hz), 4.64 (1H, q, J = 7.0 Hz), 5.07-5.14 (2H, m), 5.23-5.27 (1H, m), 6.99 (1H, m), 7.19-7.33 (10H, m);

^{13}C NMR (126 MHz, $CDCl_3$) δ 19.8, 20.0, 21.7, 24.9, 27.4, 28.2, 28.3, 30.7, 39.0, 39.4, 40.3, 42.3, 42.8, 46.3, 47.2, 53.2, 54.1, 60.2, 60.9, 66.2, 66.3, 76.7, 77.0, 77.3, 79.7, 80.1, 126.9, 127.4, 128.1, 128.2, 128.2, 128.4, 128.5, 128.8, 129.3, 129.3, 135.6, 136.3, 155.1, 169.6, 169.9, 171.0, 171.1, 171.1, 171.8;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{30}H_{39}N_3O_6Na$ 560.2731; Found 560.2749.



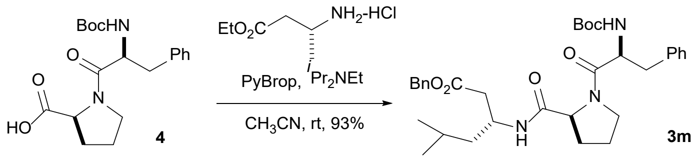
3l: $[\alpha]^{19}_{D}$ –31.7 (c 1.26, $CHCl_3$);

IR (neat) 3308, 3030, 2976, 1683, 1645, 1520, 1507, 1497, 1455, 1365, 1248, 1165, 1027, 752, 699 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$, 4:1 mixture of rotamers) δ 1.40 (9H, s), 1.83-2.31 (4H, m), 2.82-3.61 (6H, m), 4.27-4.32 (1H×1/5, m), 4.53-4.55 (1H×4/5, m), 4.66 (1H×4/5, dd, J = 15.2, 7.2 Hz), 4.99-5.09 (2H, m), 5.20-5.31 (1H+2H×1/5, m), 5.39 (1H×4/5, dd, J = 14.5, 6.6 Hz), 7.14-7.34 (15 H, m), 7.60 (1H×4/5, d, J = 8.2 Hz), 8.08 (1H×1/5, d, J = 7.4 Hz);

^{13}C NMR (126 MHz, $CDCl_3$) δ 21.7, 25.0, 27.2, 28.3, 28.3, 30.2, 39.0, 39.1, 40.1, 40.5, 46.2, 47.3, 50.0, 51.0, 53.2, 54.2, 60.2, 60.9, 66.3, 66.4, 76.6, 79.7, 80.4, 126.4, 126.6, 126.9, 127.4, 127.4, 127.6, 128.0, 128.1, 128.2, 128.5, 128.5, 128.7, 128.9, 129.3, 129.3, 135.4, 135.5, 135.6, 136.3, 140.3, 141.1, 155.1, 155.9, 169.8, 169.9, 170.5, 170.6, 170.8, 172.1;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{35}H_{41}N_3O_6Na$ 622.2888; Found 622.2912.



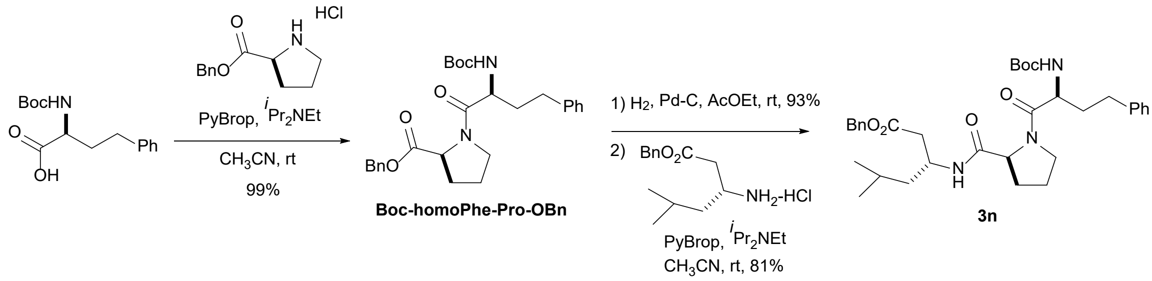
3m: $[\alpha]^{19}{}_D$ –56.3 (*c* 0.64, $CHCl_3$);

IR (neat) 3414, 3315, 3027, 2973, 2876, 1714, 1655, 1516, 1498, 1455, 1391, 1366, 1269, 1248, 1170, 1098, 1047, 1027, 906, 870, 752, 699 cm^{-1};

^1H NMR (500 MHz, $CDCL_3$, 1:1 mixture of rotamers) δ 0.82-0.91 (6H, m), 1.16-1.61 (13H, m), 1.83-2.26 (4H, m), 2.45-2.62 (2H, m), 2.83-2.93 (1H, m), 3.02 (1H×1/2, dd, J = 13.9, 6.1 Hz), 3.14-3.20 (1H×1/2, m), 3.39-3.60 (2H, m), 4.27-4.34 (3H×1/2, m), 4.44-4.46 (1H×1/2, m), 4.64 (1H×1/2, q, J = 7.8 Hz), 5.02-5.17 (2H, m), 5.28 (1H×1/2, d, J = 8.3 Hz), 6.93 (1H×1/2, d, J = 8.8 Hz), 7.17-7.38 (m, 10H), 7.49 (1H×1/2, d, J = 9.3 Hz);

^{13}C NMR (126 MHz, $CDCl_3$) δ 21.6, 21.8, 22.1, 22.8, 23.1, 24.9, 25.0, 27.4, 28.2, 28.3, 30.5, 38.8, 39.0, 39.2, 39.4, 42.7, 43.1, 44.5, 45.1, 46.4, 47.3, 53.2, 54.1, 60.2, 60.9, 66.2, 66.3, 79.7, 80.2, 126.8, 127.4, 128.1, 128.2, 128.2, 128.3, 128.4, 128.5, 128.8, 129.3, 129.3, 135.7, 135.9, 136.3, 155.1, 155.6, 169.8, 170.0, 171.2, 171.3, 171.4, 171.9;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{33}H_{45}N_3O_6Na$ 602.3201; Found 602.3228.



Boc-homoPhe-Pro-OBn

To a solution of Boc-homoPhe-OH (279 mg, 0.999 mmol) and H-Pro-OBn·HCl (290 mg, 1.20 mmol) in MeCN (4.0 mL) were added PyBrop (606 mg, 1.30 mmol) and *i*Pr_2NEt (512 μL3.00 mmol) The mixture was stirred for 3 h, quenched with diluted NaOH, extracted with EtOAc, washed with diluted HCl and brine, dried over Na_2SO_4, filtered, and concentrated *in vacuo*. The crude product was purified using column chromatography (Hexane:AcOEt = 90 : 10 then 80 : 20) to afford Boc-homoPhe-Pro-OBn (460 mg, 0.986 mmol, 99%):

$[\alpha]^{23}_{D}$ –40.2 (*c* 2.30, $CHCl_3$);

IR (neat) 3309, 2975, 1747, 1705, 1647, 1520, 1497, 1456, 1365, 1248, 1168, 1046, 1024, 871, 750, 699 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) δ 1.46 (s, 9H), 1.81-2.23 (6H, m), 2.65-2.75 (2H, m), 3.34-3.38 (1H, m), 3.54-3.58 (1H, m), 4.40-4.47 (1H, m), 4.53-4.62 (1H, m), 5.11 (1H, d, *J* = 12.2 Hz), 5.18 (1H, d, *J* = 12.3 Hz), 5.34 (1H, d, *J* = 8.6 Hz), 7.17-7.36 (10H, m)

^{13}C NMR (126 MHz, $CDCl_3$) δ 14.1, 21.0, 24.8, 28.3, 28.9, 31.2, 34.7, 46.6, 51.1, 58.8, 60.3, 66.8, 79.6, 125.9, 128.1, 128.2, 128.3, 128.4, 128.5, 128.5, 135.5, 141.2, 155.5, 171.0, 171.6;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{27}H_{34}N_2O_5Na$ 489.2360; Found 489.2353.

Boc-homoPhe-Pro-OH

To a solution of Boc-homoPhe-Pro-OBn (300 mg, 0.643 mmol) in AcOEt (5.00 mL) was added Pd-C (5 wt% Pd on carbon, 30.0 mg) at room temperature under Ar atmosphere. The mixture was stirred for 16 h under H_2 atmosphere, filtered through celite pad, and concentrated *in vacuo* to give Boc-homoPhe-Pro-OH (225 mg, 0.598 mmol, 93%), which was employed directly in the next reaction.

$[\alpha]^{23}_{D}$ –45.0 (*c* 2.10, $CHCl_3$);

IR (neat) 3309, 2977, 1714, 1646, 1615, 1497, 1454, 1392, 1367, 1249, 1167, 1048, 1025, 912, 870, 754, 701, 666 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) δ 1.44 (9H, s), 1.90-2.16 (6H, m), 2.67-2.71 (2H, m), 3.30-3.36 (1H, m), 3.57 (1H, q, *J* = 6.3 Hz), 4.44-4.46 (1H, m), 4.55 (1H, t, *J* = 5.2 Hz), 5.51 (1H, d, *J* = 8.7 Hz), 7.15-7.27 (5H, m);

^{13}C NMR (126 MHz, $CDCl_3$) δ 14.1, 21.0, 24.7, 28.2, 28.3, 28.4, 31.2, 34.3, 47.0, 51.1, 59.0, 60.4, 79.8, 126.0, 128.3, 128.4, 128.5, 141.0, 155.6, 171.3, 172.3, 174.4;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{20}H_{28}N_2O_5Na$ 399.1890; Found 399.1885.

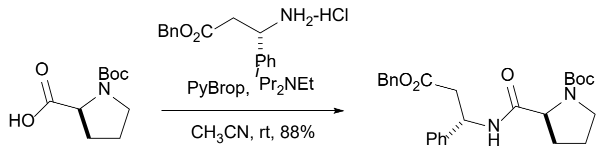
3n: $[\alpha]^{23}_{D}$ –32.7 (*c* 1.94, $CHCl_3$);

IR (neat) 3312, 2956, 1714, 1683, 1644, 1521, 1497, 1455, 1366, 1249, 1169, 1047, 1024, 868, 751, 699 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) δ 0.86 (6H, d, *J* = 6.6 Hz), 1.21-1.27 (1H, m), 1.40-1.47 (1H, m), 1.45 (9H, s), 1.55-1.59 (1H, m), 1.84-2.04 (5H, m), 2.25 (1H, t, *J* = 8.5 Hz), 2.44-2.53 (2H, m), 2.66-2.76 (2H, m), 3.27-3.31 (1H, m), 3.46-3.49 (1H, m), 4.25-4.30 (1H, m), 4.42-4.52 (2H, m), 4.98 (1H, d, *J* = 12.3 Hz), 5.05 (1H, d, *J* = 12.3 Hz), 5.31 (1H, d, *J* = 8.7 Hz), 6.94 (1H, d, *J* = 9.0 Hz), 7.15-7.36 (10H, m);

^{13}C NMR (126 MHz, $CDCl_3$) δ 21.9, 22.9, 24.9, 27.5, 28.3, 31.5, 34.7, 39.1, 43.1, 44.2, 46.9, 51.2, 60.0, 66.2, 79.6, 126.0, 128.2, 128.39, 128.46, 128.47, 128.60, 135.6, 140.9, 155.5, 170.2, 171.3, 172.4;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{34}H_{47}N_3O_6Na$ 616.3357; Found 616.3342.



5a

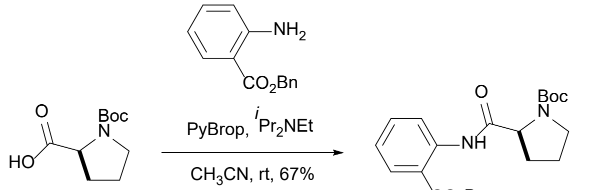
5a: $[\alpha]^{27}_{D}$ –81.1 (*c* 2.55, $CHCl_3$);

IR (neat) 3268, 2975, 2931, 2881, 1696, 1604, 1587, 1522, 1449, 1381, 1297, 1258, 1162, 1142, 1118, 10086, 918, 756, 699 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) see appendix because of rotamer;

^{13}C NMR (126 MHz, $CDCl_3$) see appendix because of rotamer;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{26}H_{32}N_2O_5Na$ 475.2203; Found 475.2205.



5b

5b: $[\alpha]^{29}_{D}$ –63.1 (*c* 1.56, $CHCl_3$);

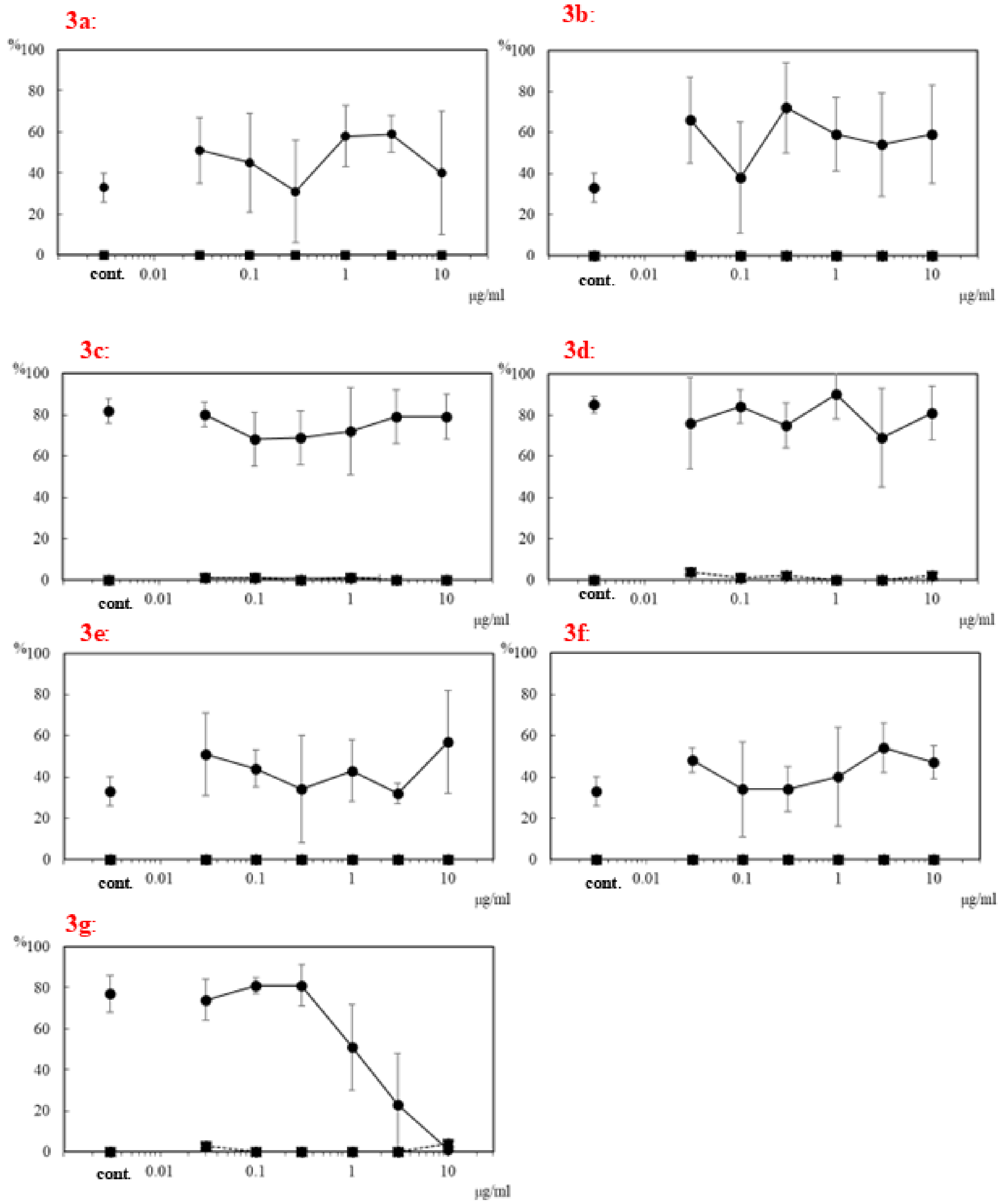
IR (neat) 3308, 3021, 2975, 2878, 1736, 1697, 1522, 1497, 1455, 1391, 1366, 1255, 1162, 1122, 1088, 977, 919, 752, 699 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$, 1.5:1 mixture of rotamers) δ 1.38 (9H×1.5/2.5, s), 1.50 (9H×1/2.5, s), 1.92-2.35 (4H, m), 3.45-3.80 (2H, m), 4.28-4.30 (1H×1.5/2.5, m), 4.58-4.66 (1H×1/2.5, m) 5.33 (2H, s), 7.02-7.11 (1H, m), 7.32-7.44 (5H, m), 7.49-7.61 (1H, m), 8.05-8.12 (1H, m), 8.72-8.80 (1H, m), 11.54 (1H×1.5/2.5, s), 11.60 (1H×1/2.5, s);

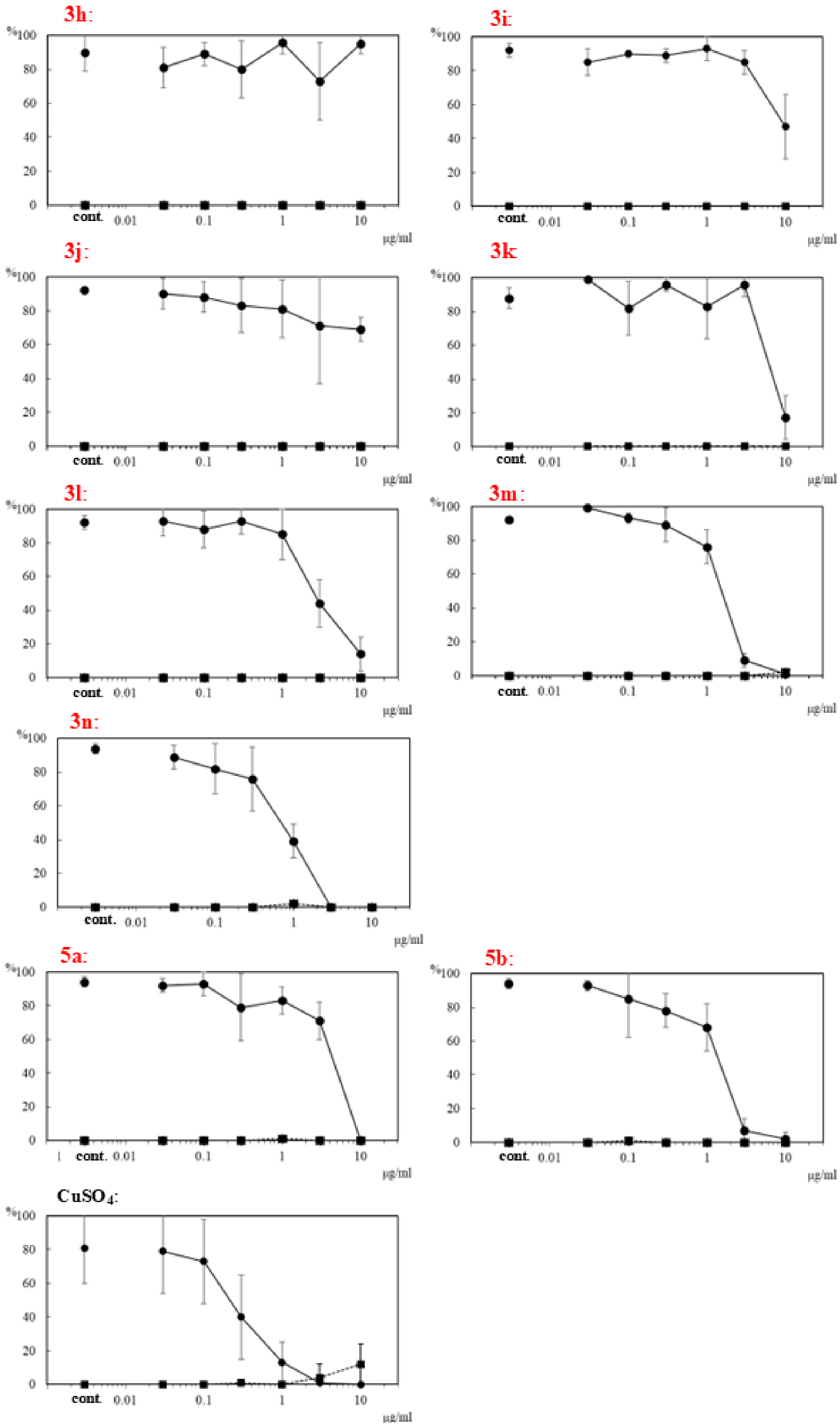
^{13}C NMR (126 MHz, $CDCl_3$) δ 23.8, 24.3, 28.0, 28.2, 28.4, 30.5, 31.5, 46.8, 47.1, 62.1, 62.7, 66.7, 67.0, 80.1, 80.3, 115.2, 120.0, 120.3, 122.5, 122.6, 128.1, 128.3, 128.4, 128.6, 130.7, 130.9, 134.6, 134.7, 135.4, 141.1, 154.2, 167.6, 172.4;

HRMS (ESI) m/z: $[M + Na]^+$ Calcd for $C_{24}H_{28}N_2O_5Na$ 447.1890; Found 447.1901.

Settlement and Mortality Test with Cyprids



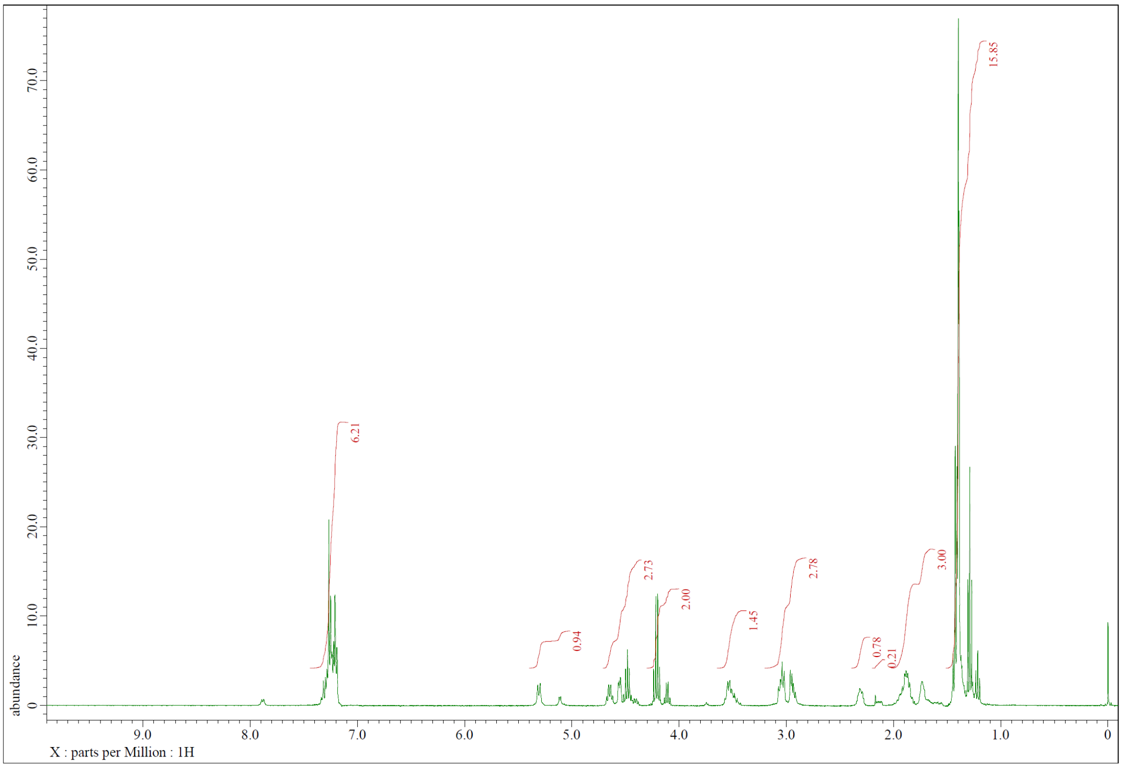
Effects of peptides **3a~3g** on settlement and mortality of cyprids after 48 h exposure. Rate of settlement of cyprids (closed circles) and mortality (closed squares) in different concentrations were plotted. Data plotted are representative means ± SD of 3 replicates. The data points that are not joined to a line indicate the percentage settlement and mortality in filtered seawater diluted to 80 % by deionized water (control).

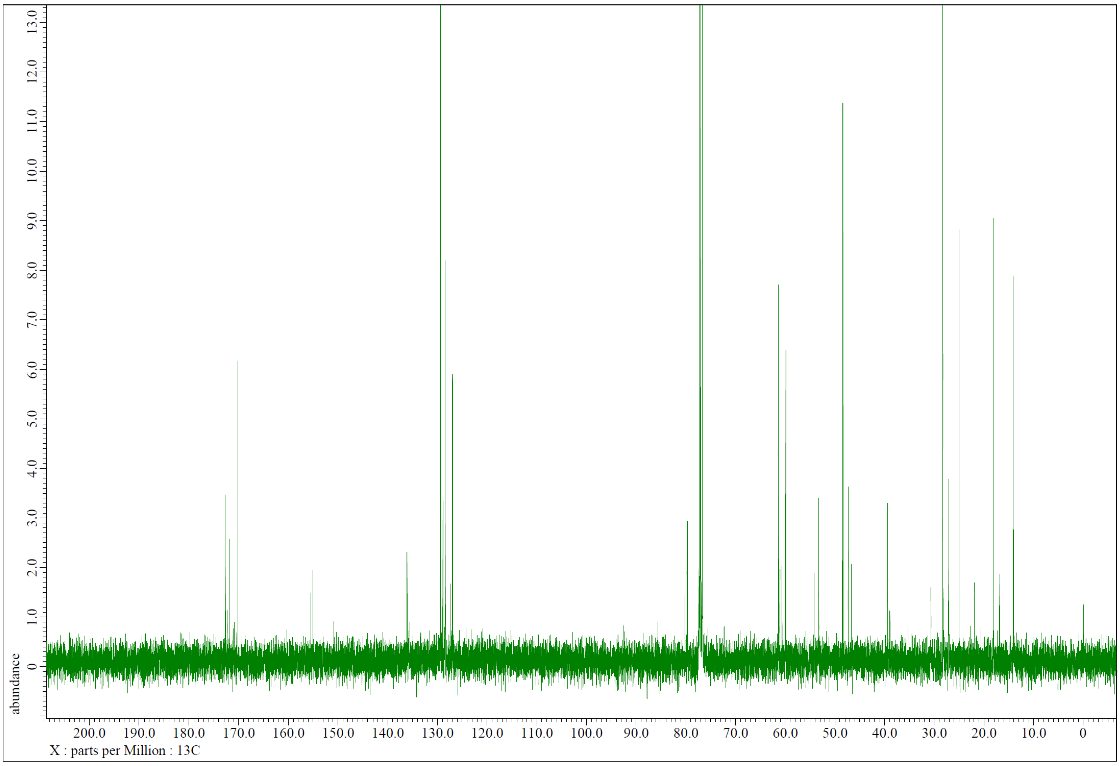


Effects of peptides **3h**~**3n**, **5a**, **5b** and $CuSO_4$ on settlement and mortality of cyprids after 48 h exposure. Rate of settlement of cyprids (closed circles) and mortality (closed squares) in different concentrations were plotted. Data plotted are representative means ± SD of 3 replicates. The data points that are not joined to a line indicate the percentage settlement and mortality in filtered seawater diluted to 80 % by deionized water (cont.).

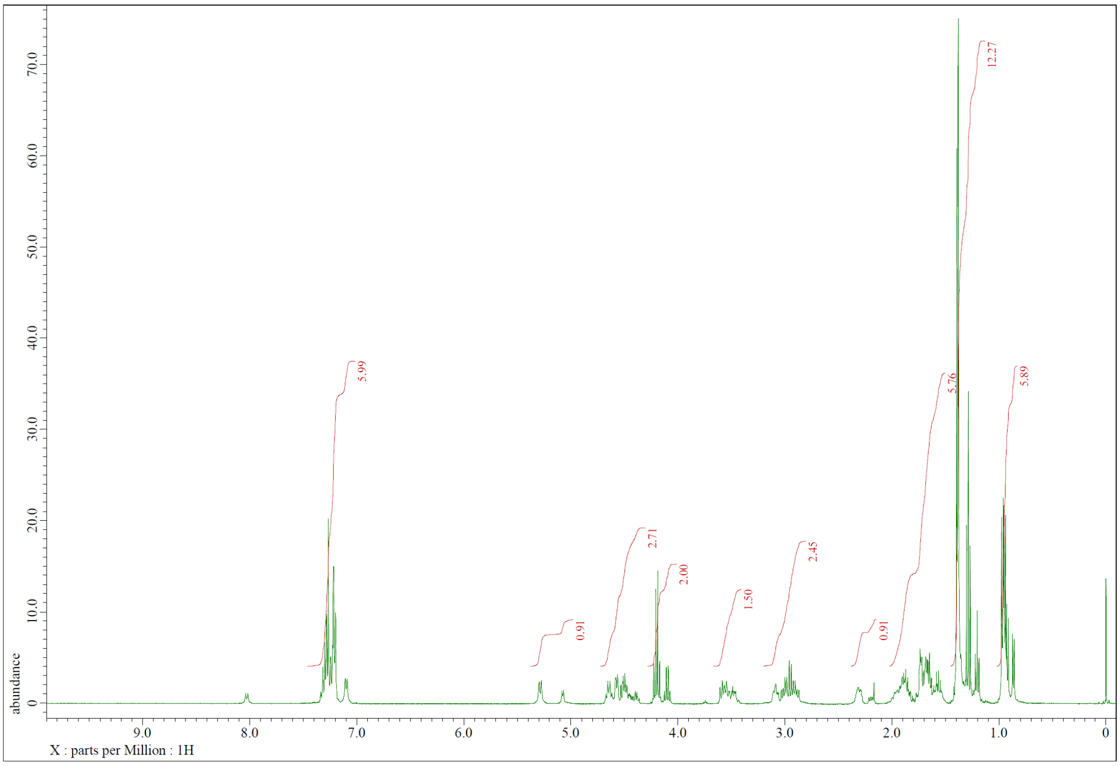
^{1}H and ^{13}C NMR Spectra

3a

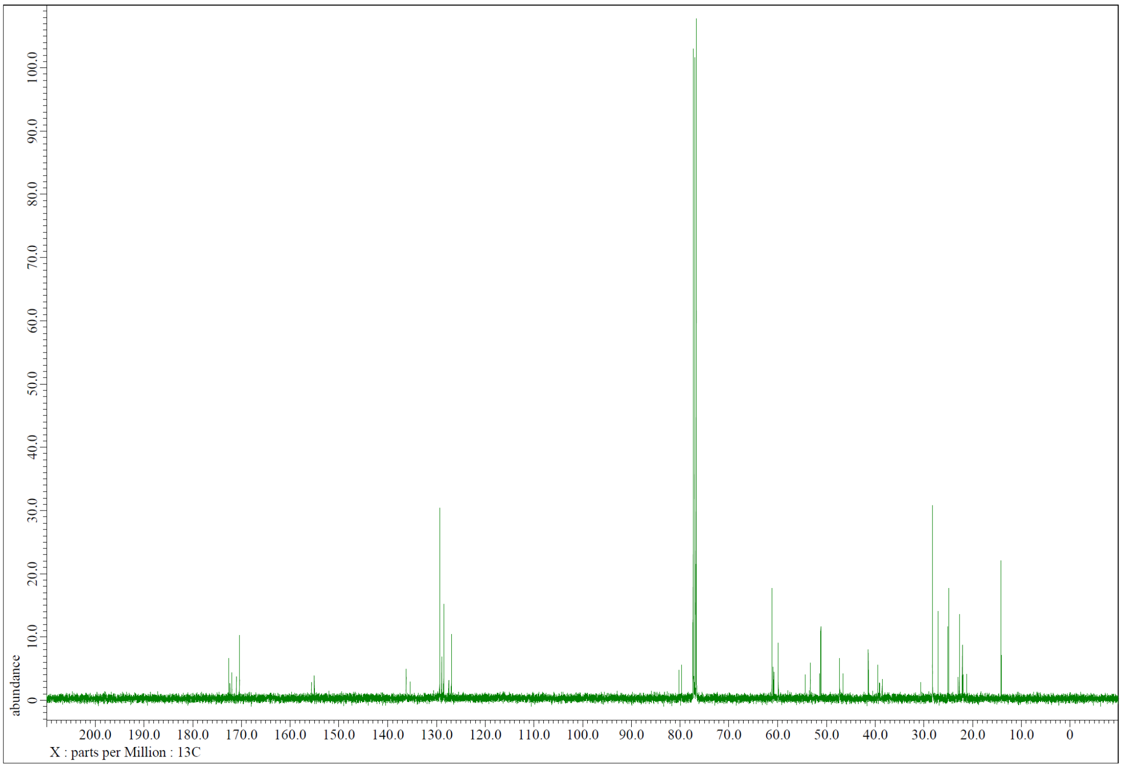




3b

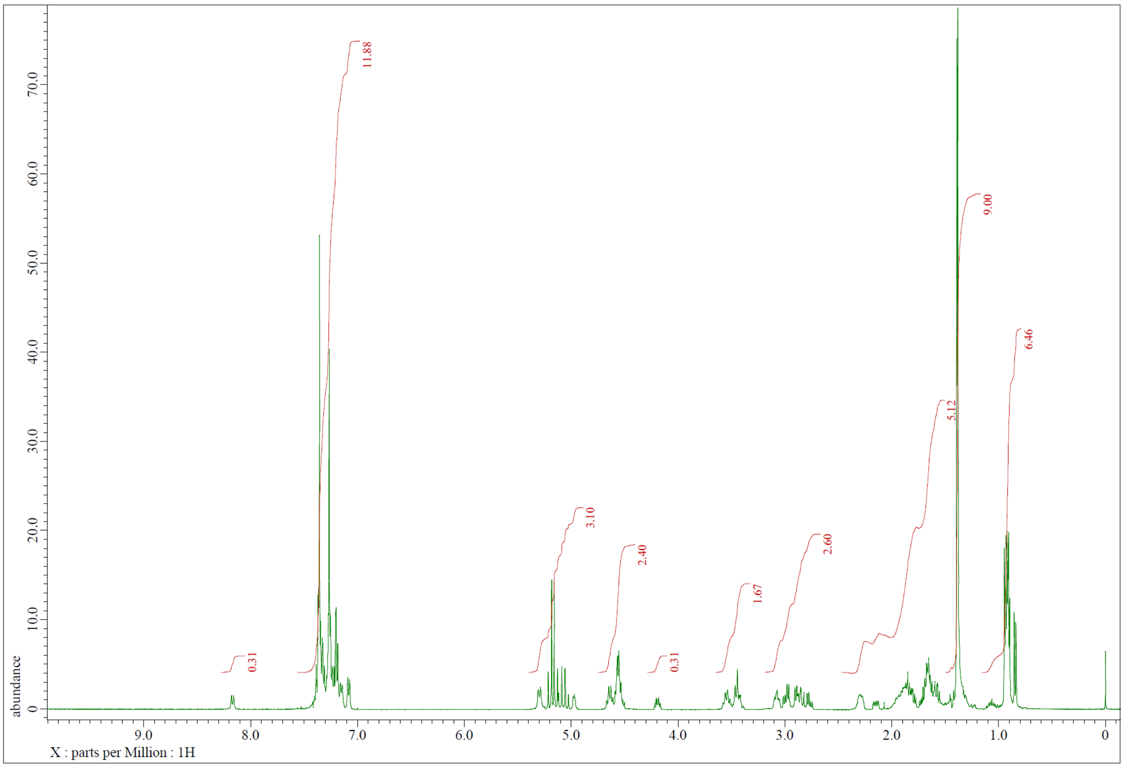


abundance
X : parts per Million : 1H
5.99
0.91
2.71
2.00
1.50
2.45
0.91
5.76
12.27
5.89

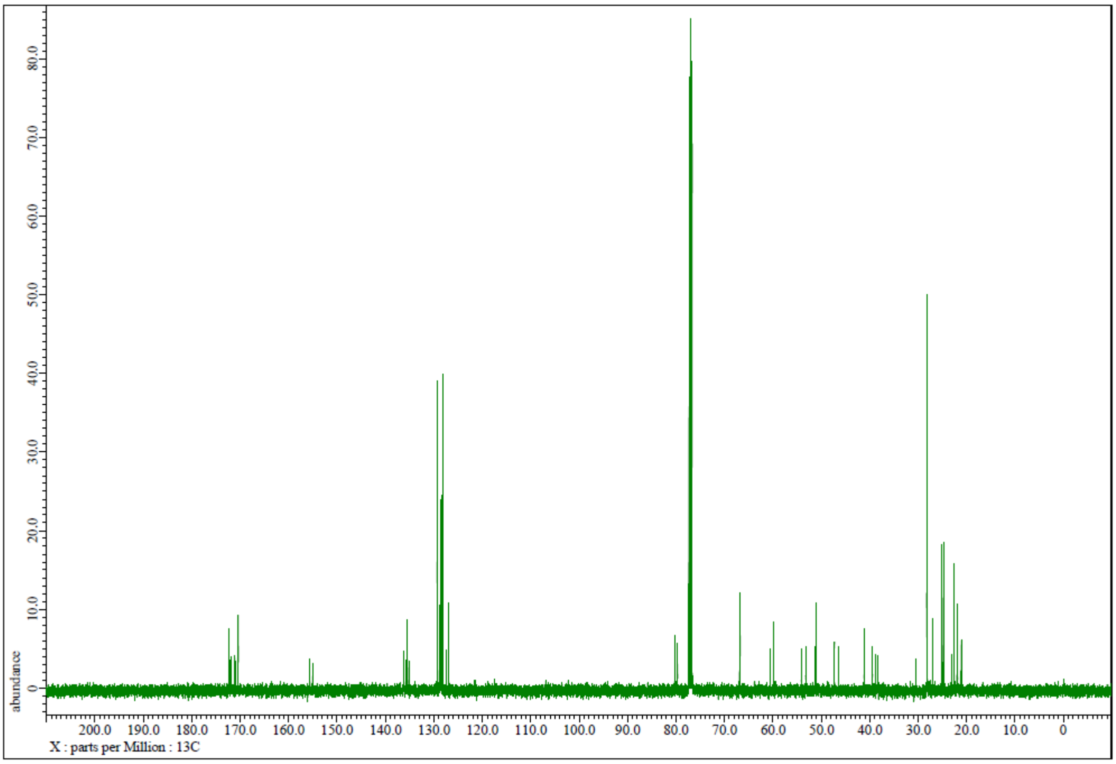


abundance
X : parts per Million : 13C

3c

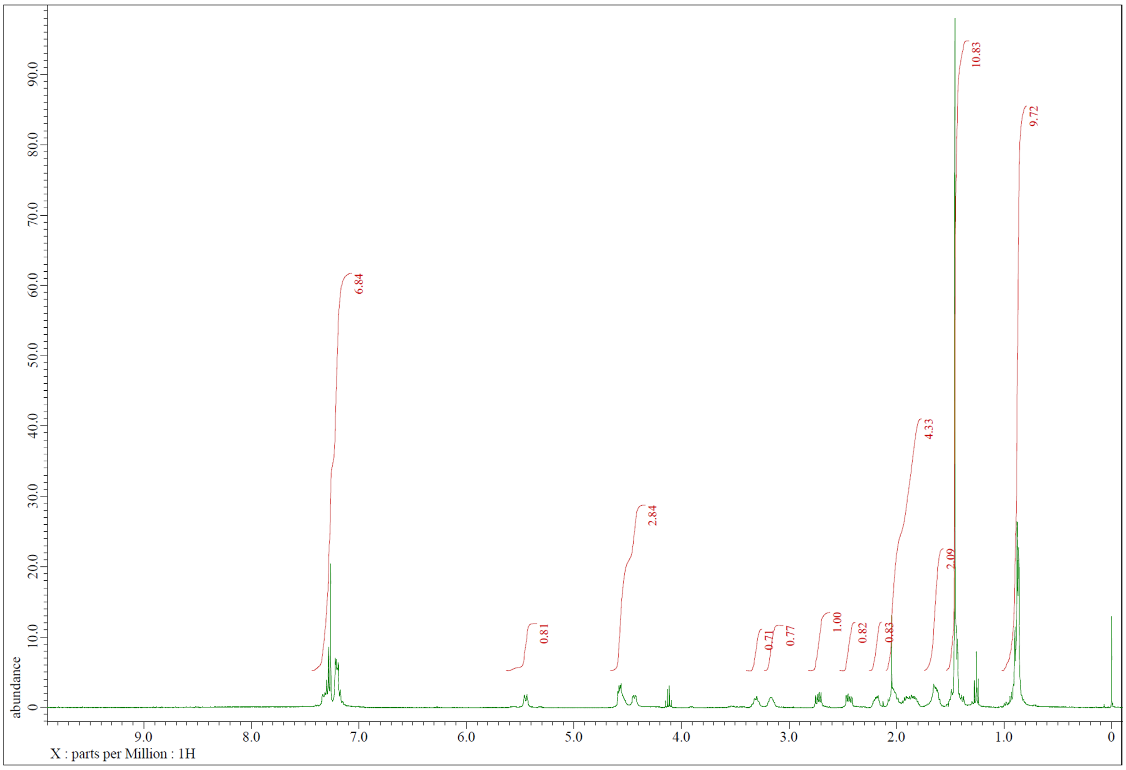


11.88
9.00
6.46
5.12
3.10
2.40
2.60
1.67
0.31
0.31
abundance
X : parts per Million : 1H

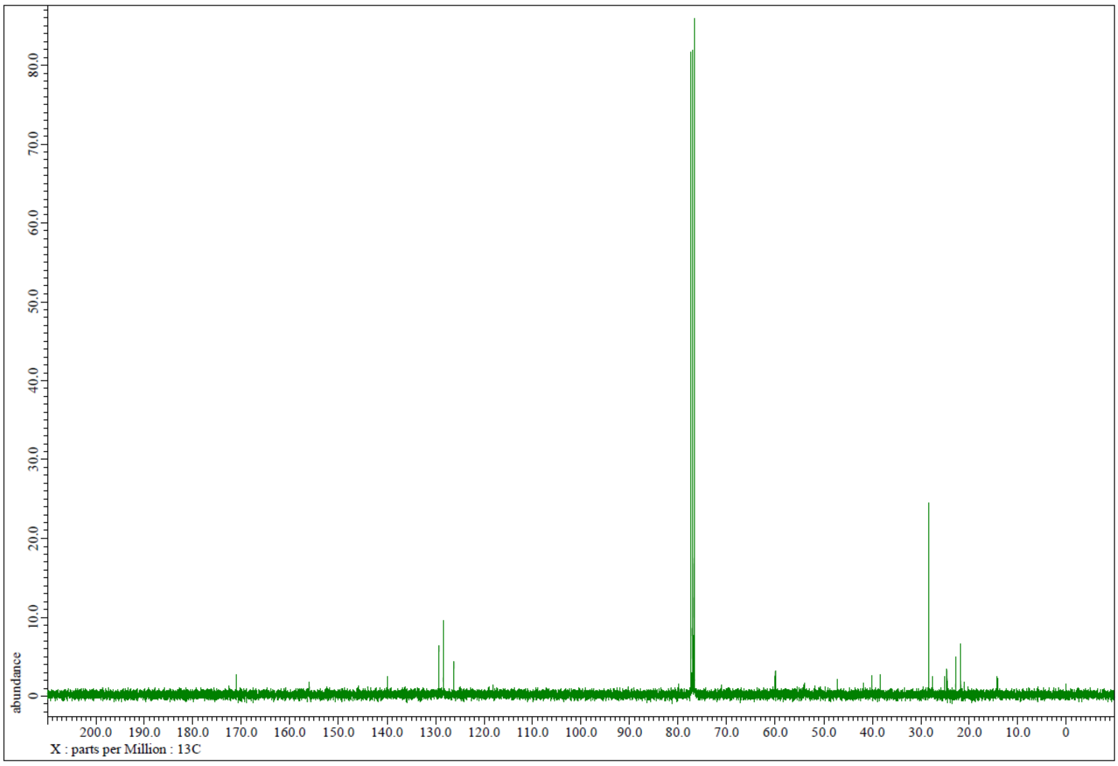


abundance
X : parts per Million : 13C

3d

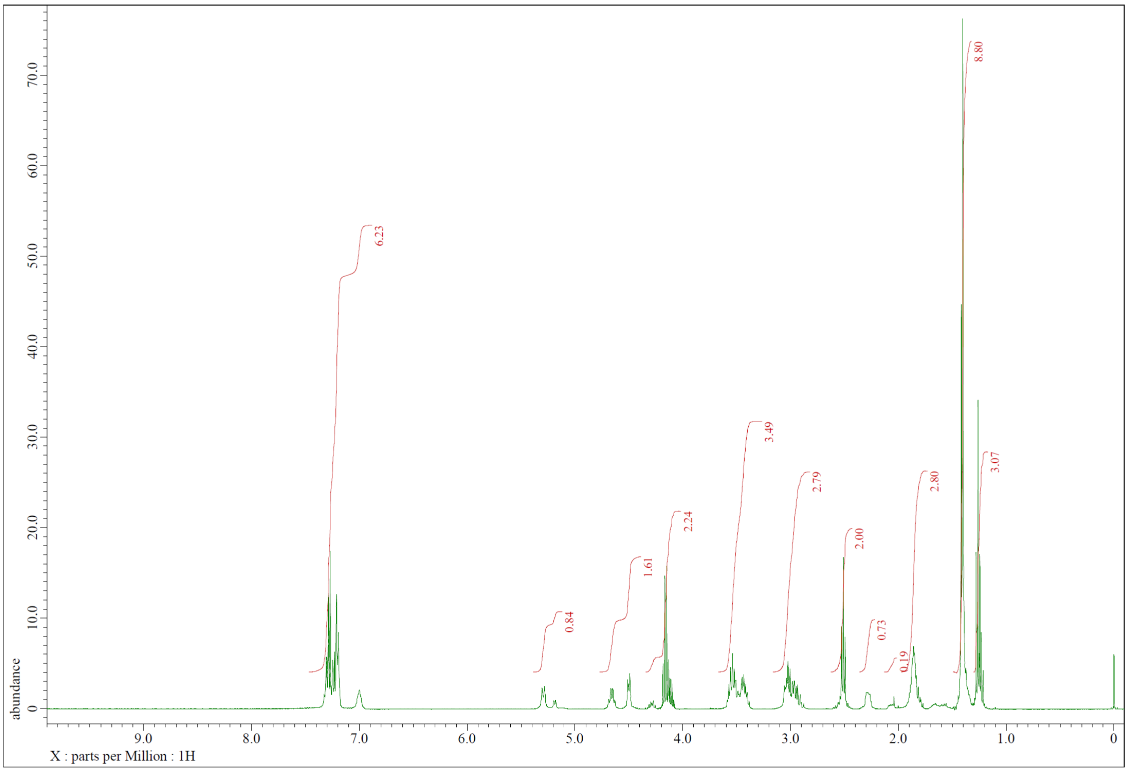


abundance
X : parts per Million : 1H
6.84
0.81
2.84
0.71
0.77
1.00
0.82
0.83
4.33
2.09
10.83
9.72

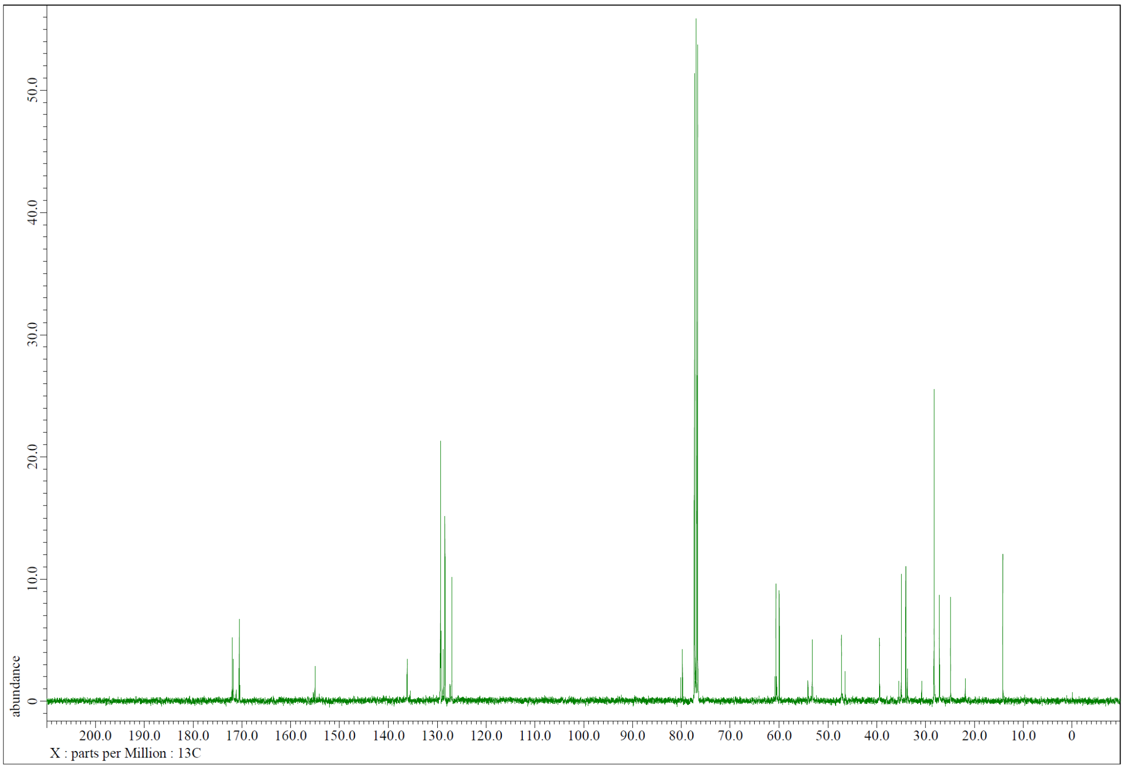


abundance
X : parts per Million : 13C

3e

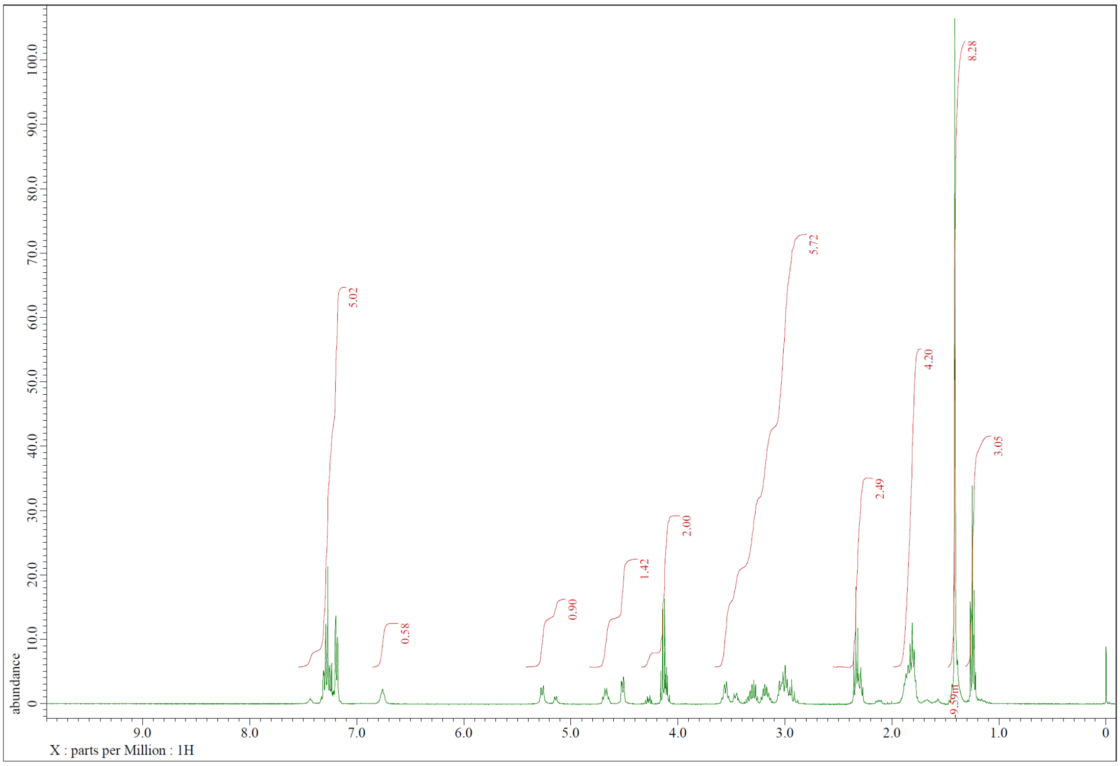


abundance
X : parts per Million : 1H
6.23
0.84
1.61
2.24
3.49
2.79
2.00
0.73
0.19
2.80
8.80
3.07

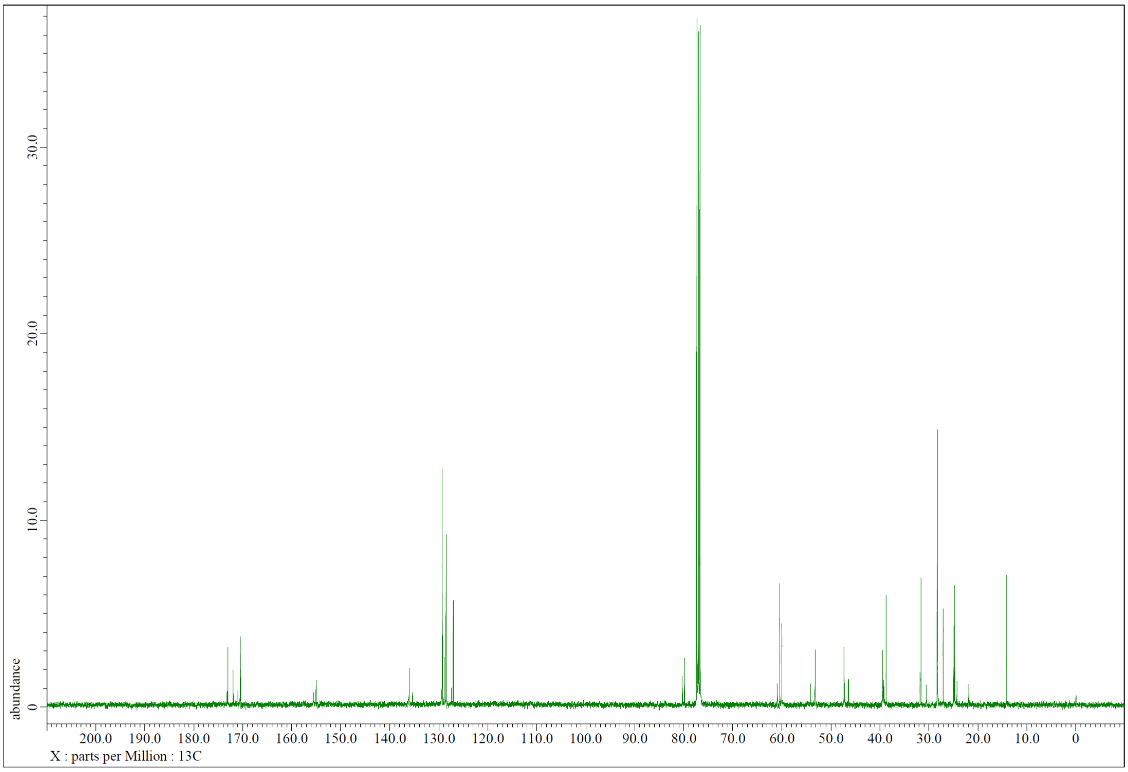


abundance
X : parts per Million : 13C

3f

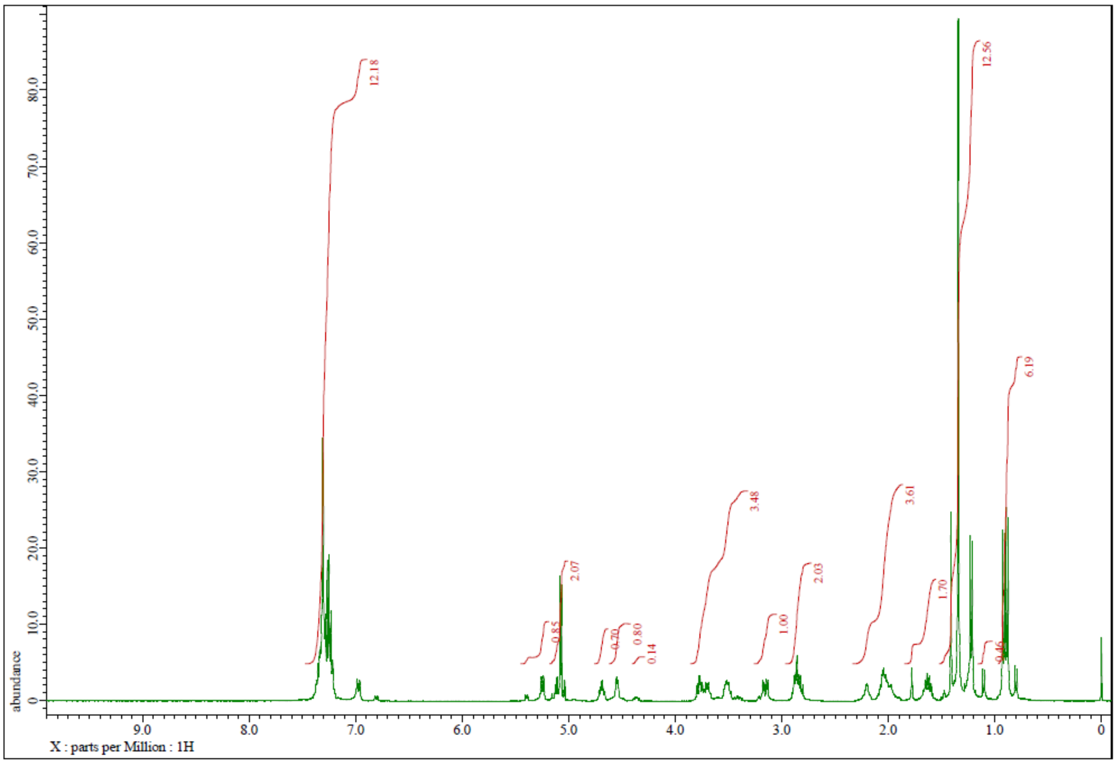


abundance
X : parts per Million : 1H
5.02
0.58
0.90
1.42
2.00
5.72
2.49
4.20
8.28
3.05

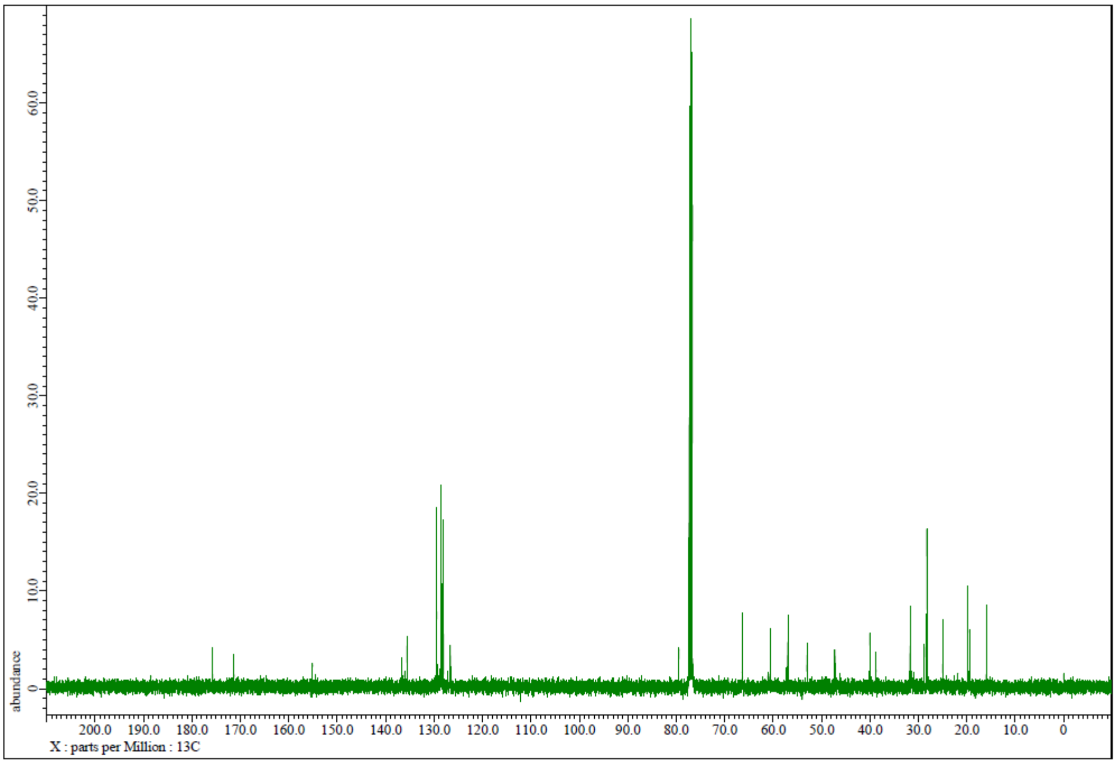


abundance
X : parts per Million : 13C

3g

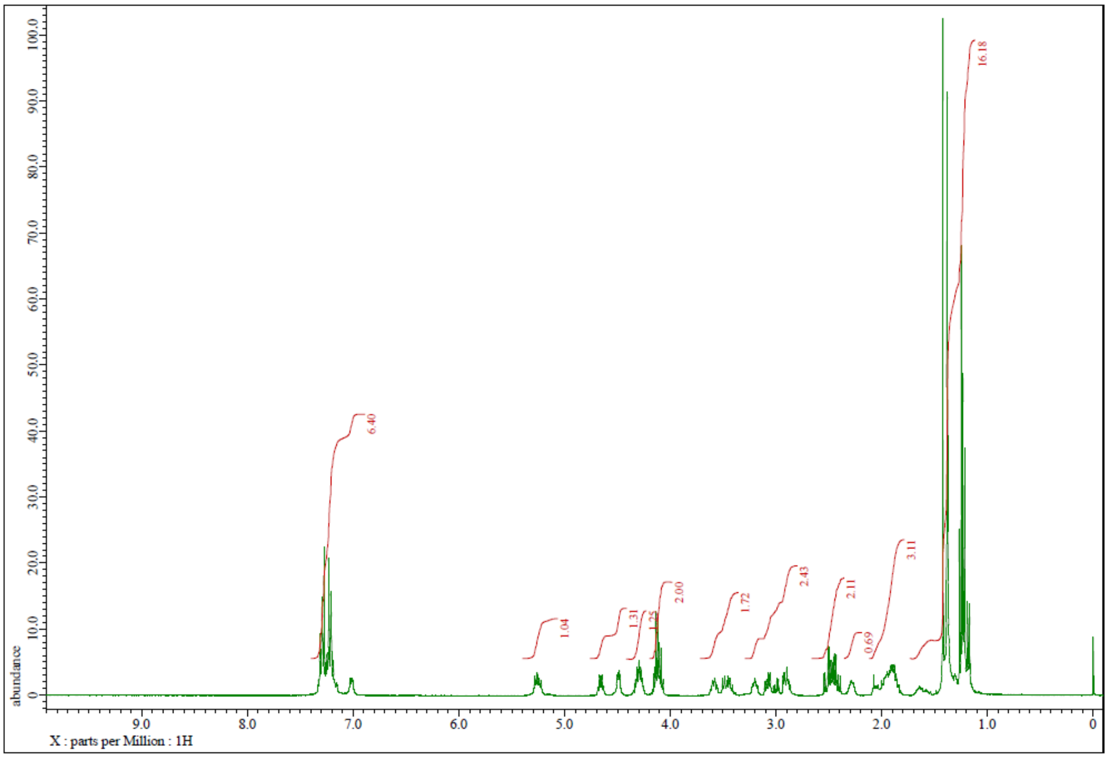


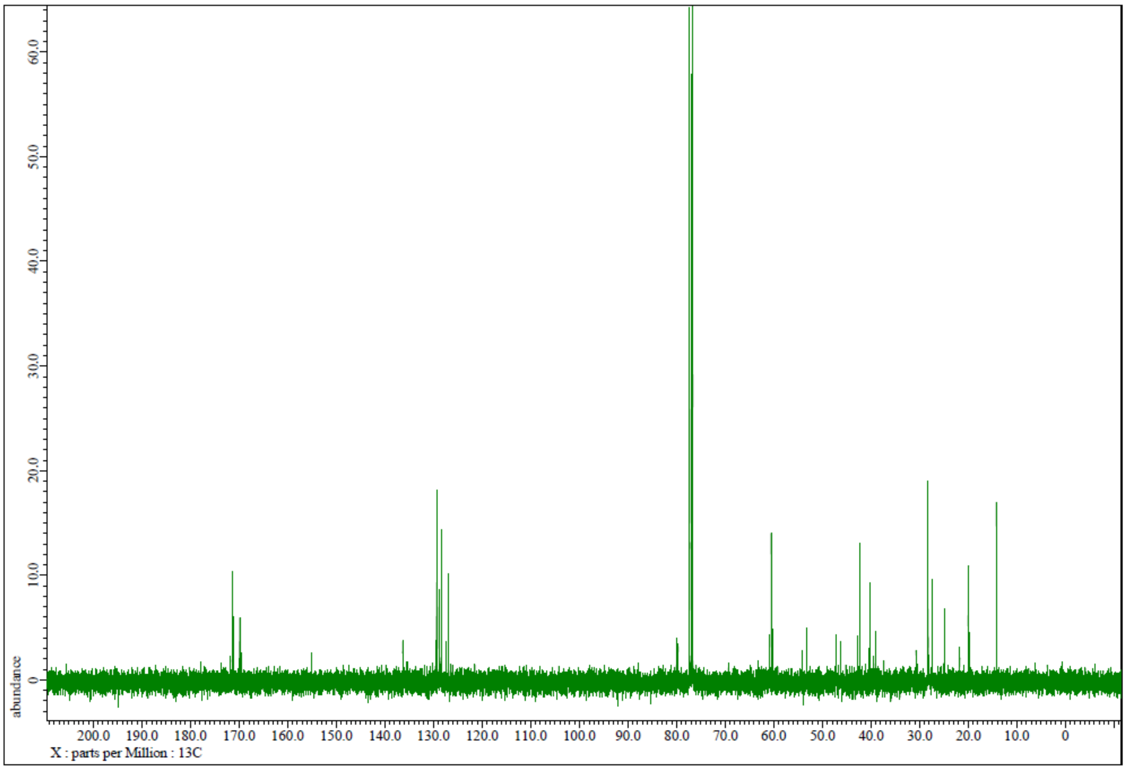
12.18
2.07
0.85
0.70
0.80
0.14
3.48
1.00
2.03
3.61
1.70
12.56
0.46
6.19
abundance
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
X : parts per Million : 1H



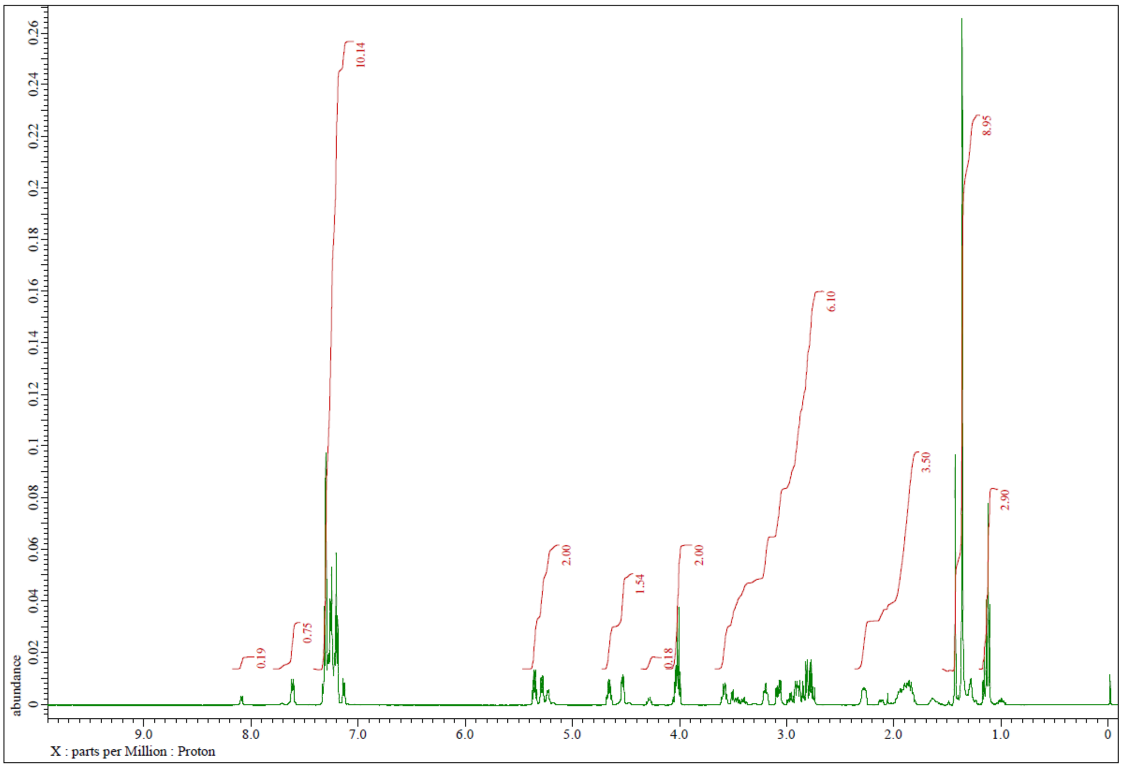
abundance
200.0
190.0
180.0
170.0
160.0
150.0
140.0
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0
X : parts per Million : 13C

3h

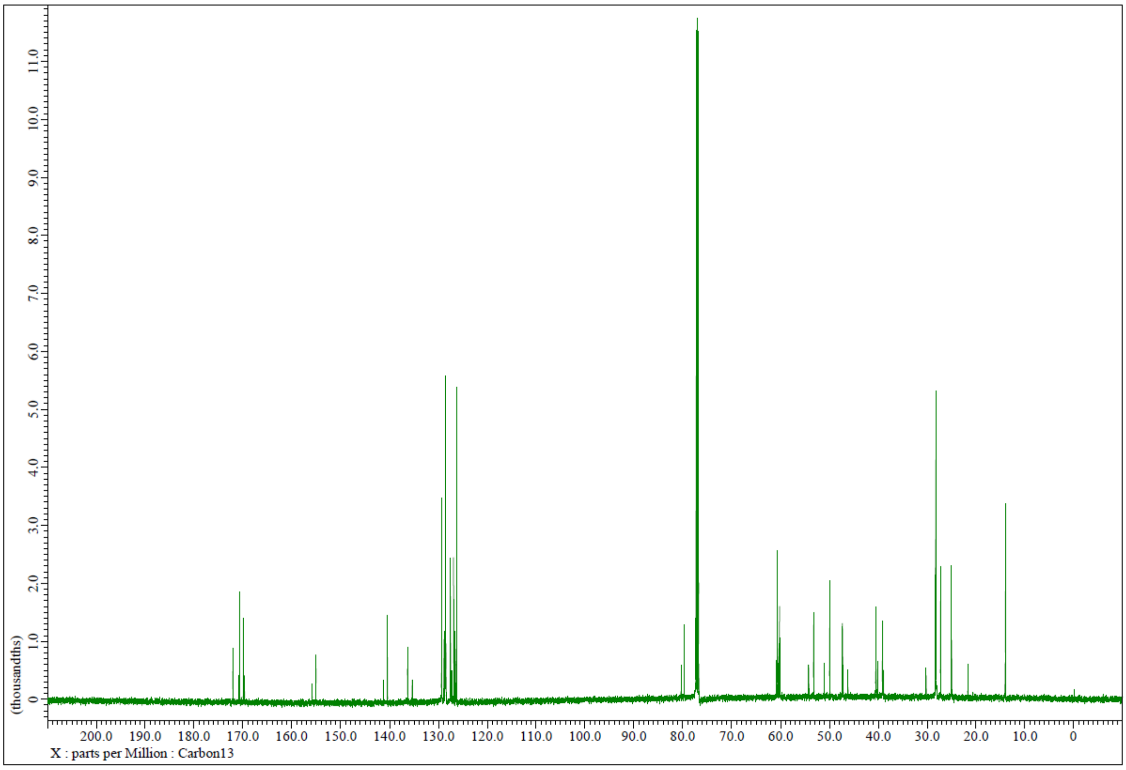

abundance
X : parts per Million : 1H
6.40
1.04
1.31
1.25
2.00
1.72
2.43
2.11
0.69
3.11
16.18


abundance
X : parts per Million : 13C

3i

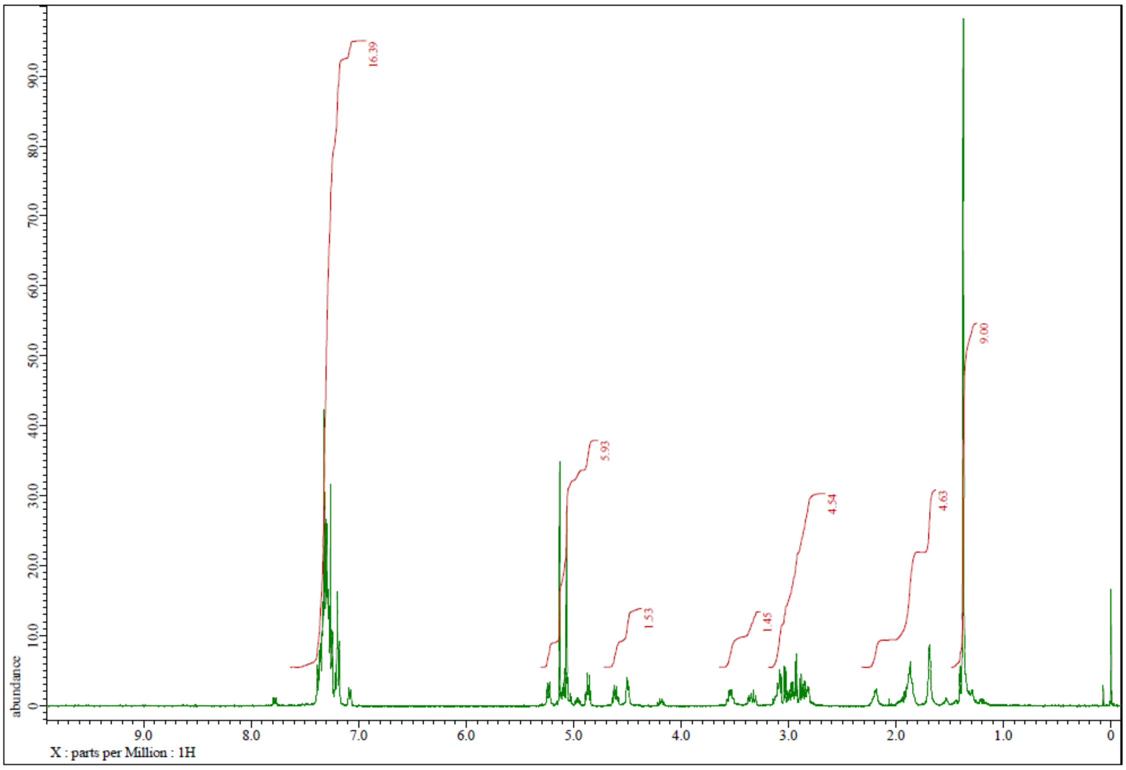


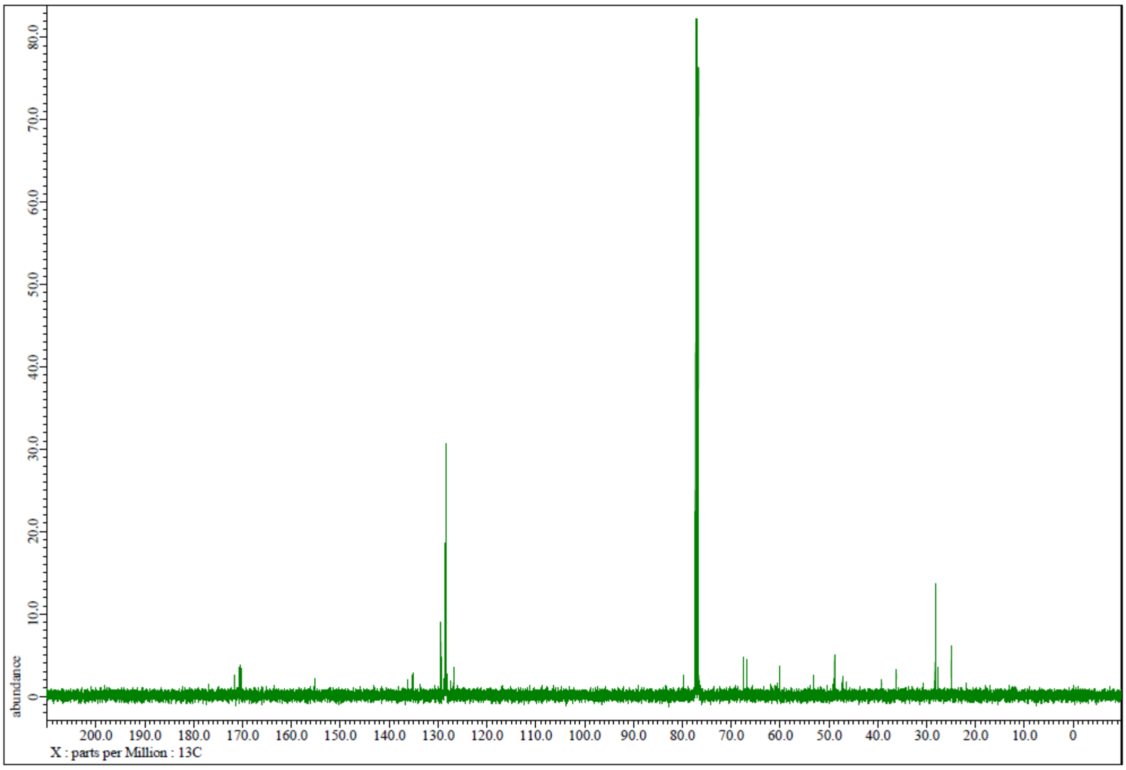
abundance
X : parts per Million : Proton
10.14
8.95
6.10
3.50
2.90
2.00
1.54
2.00
0.19
0.75
0.18



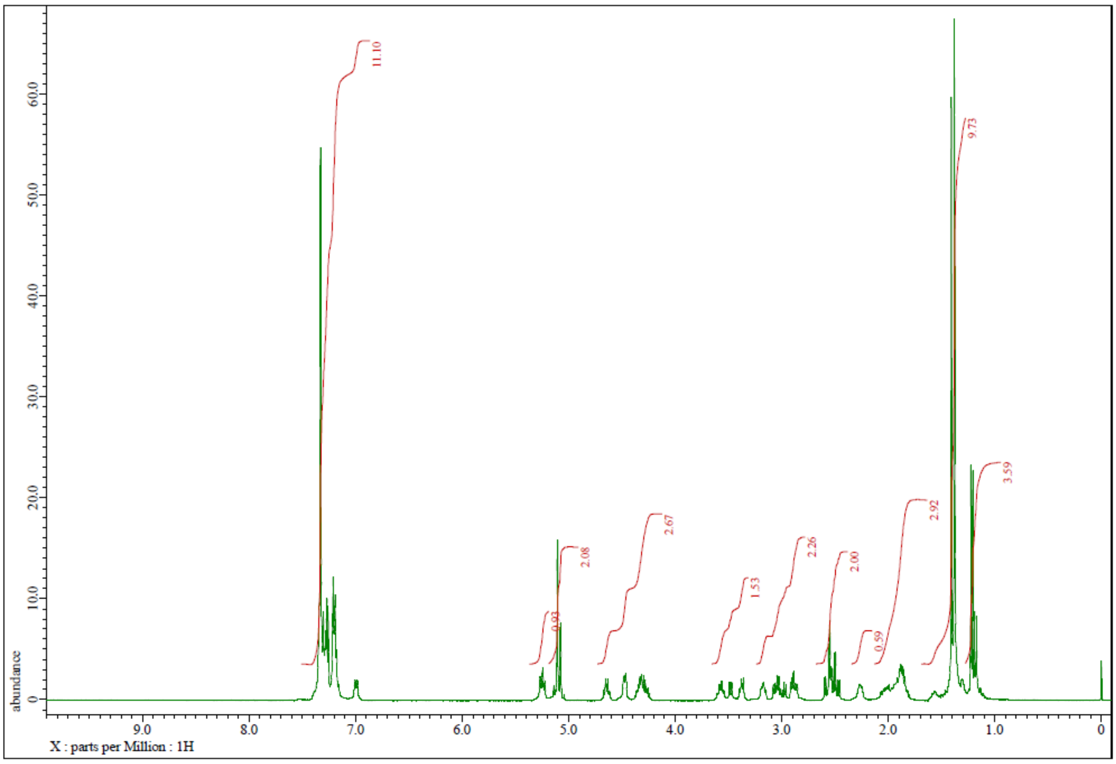
(thousandths)
X : parts per Million : Carbon13

3j

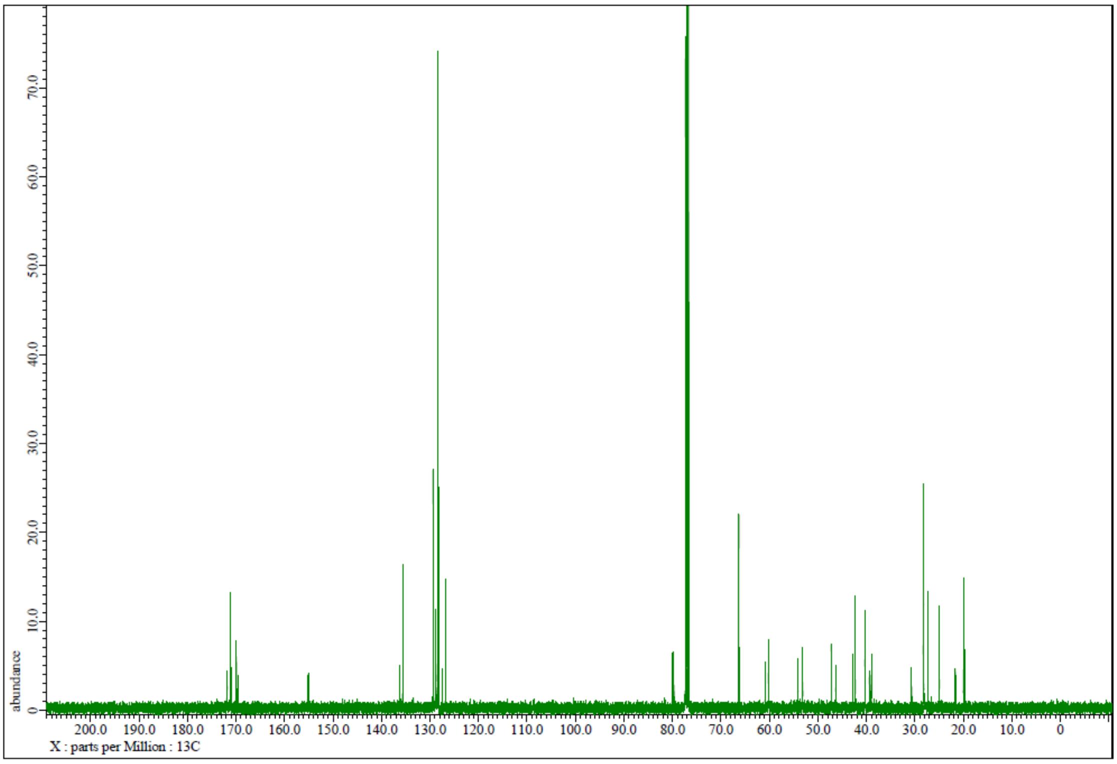

16.39
5.93
1.53
1.45
4.54
4.63
9.00
abundance
X : parts per Million : 1H


abundance
X : parts per Million : 13C

3k

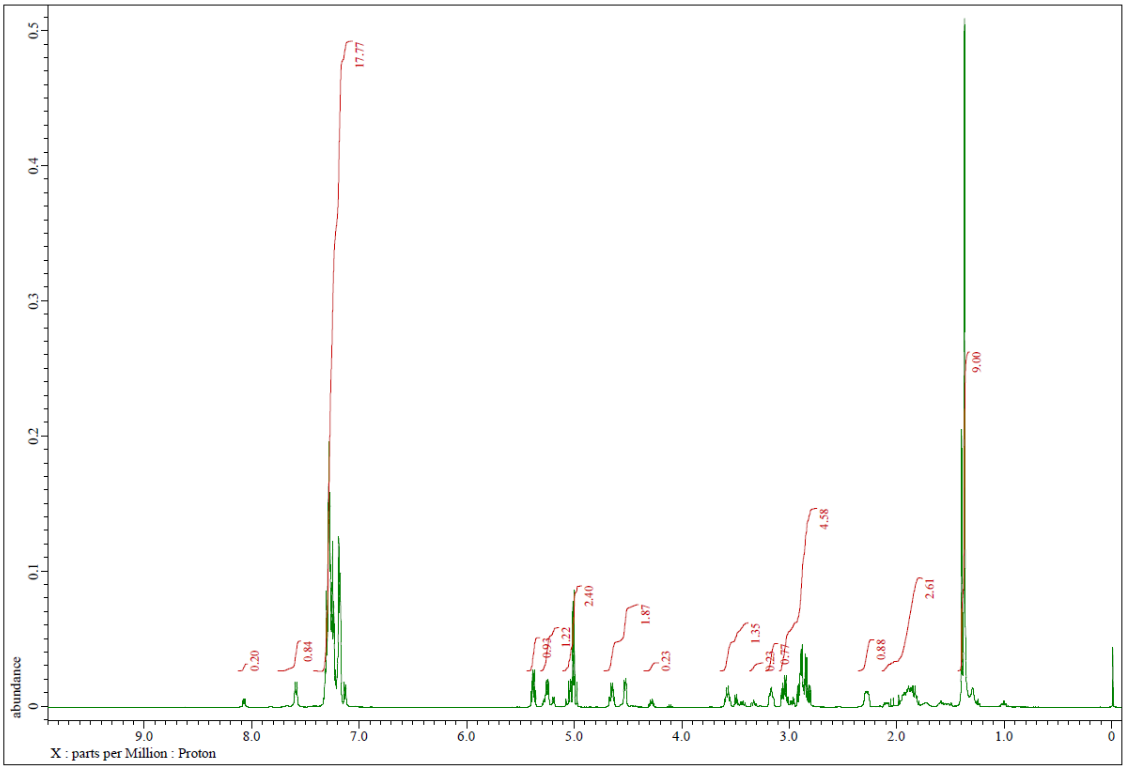


11.10
9.73
3.59
2.92
2.67
2.26
2.08
2.00
1.53
0.93
0.59
abundance
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
X : parts per Million : 1H

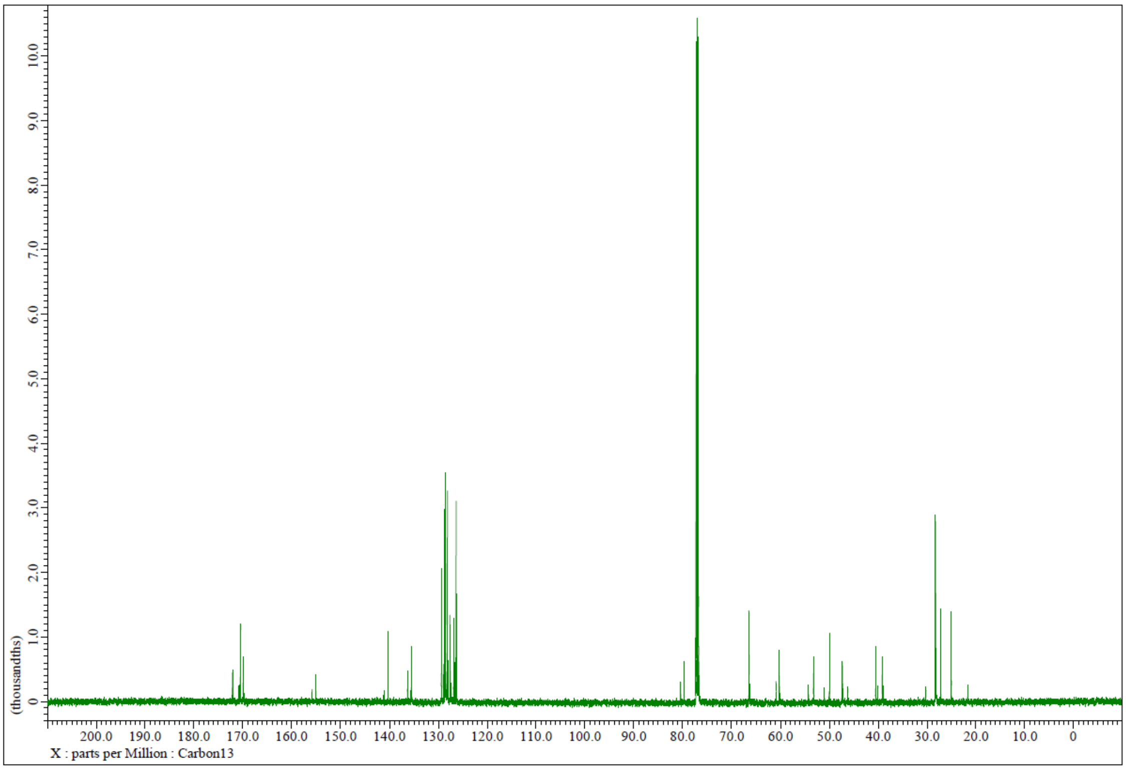


abundance
200.0
190.0
180.0
170.0
160.0
150.0
140.0
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0
X : parts per Million : 13C

3l

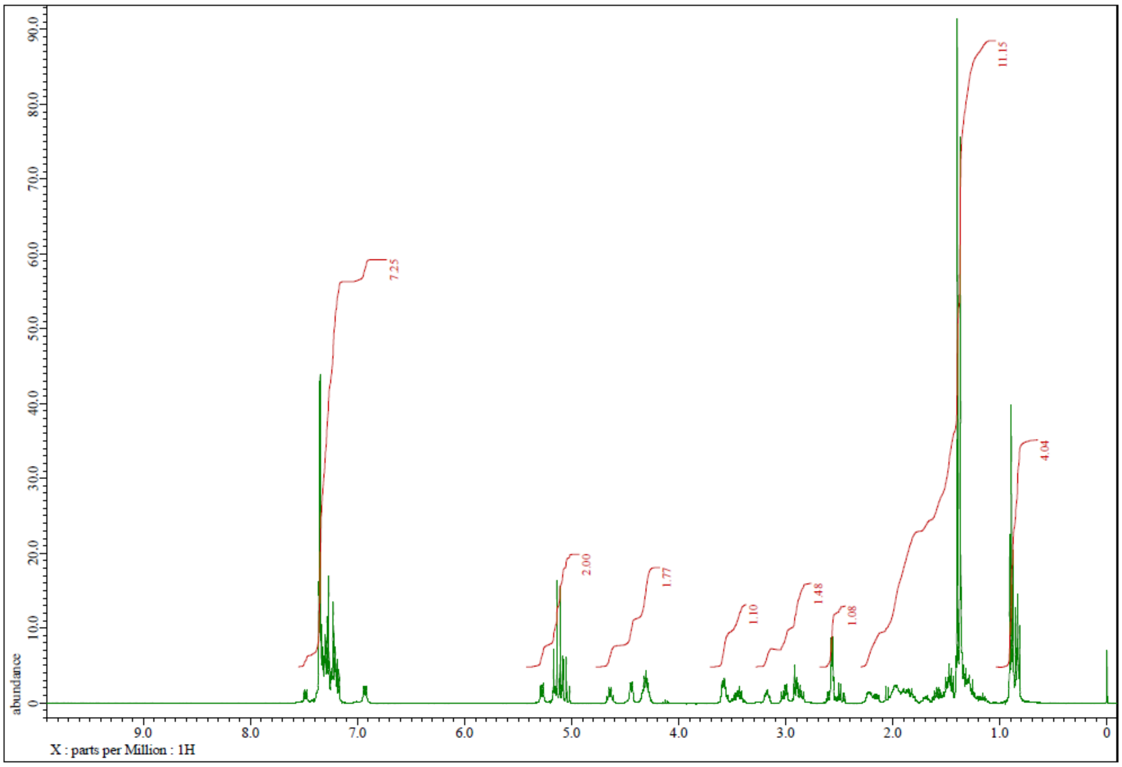


abundance
0.5
0.4
0.3
0.2
0.1
0
17.77
9.00
4.58
2.61
2.40
1.87
1.35
1.22
0.93
0.88
0.84
0.77
0.23
0.23
0.20
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
X : parts per Million : Proton

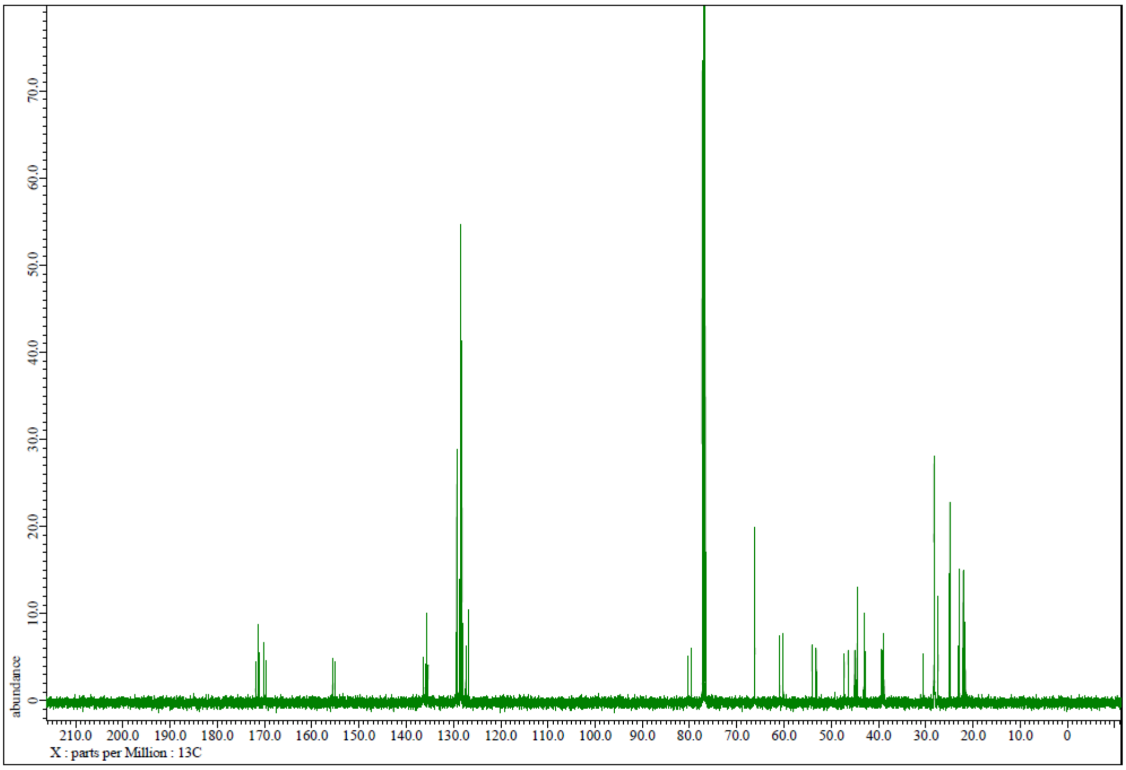


(thousandths)
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0 0
X : parts per Million : Carbon13

3m

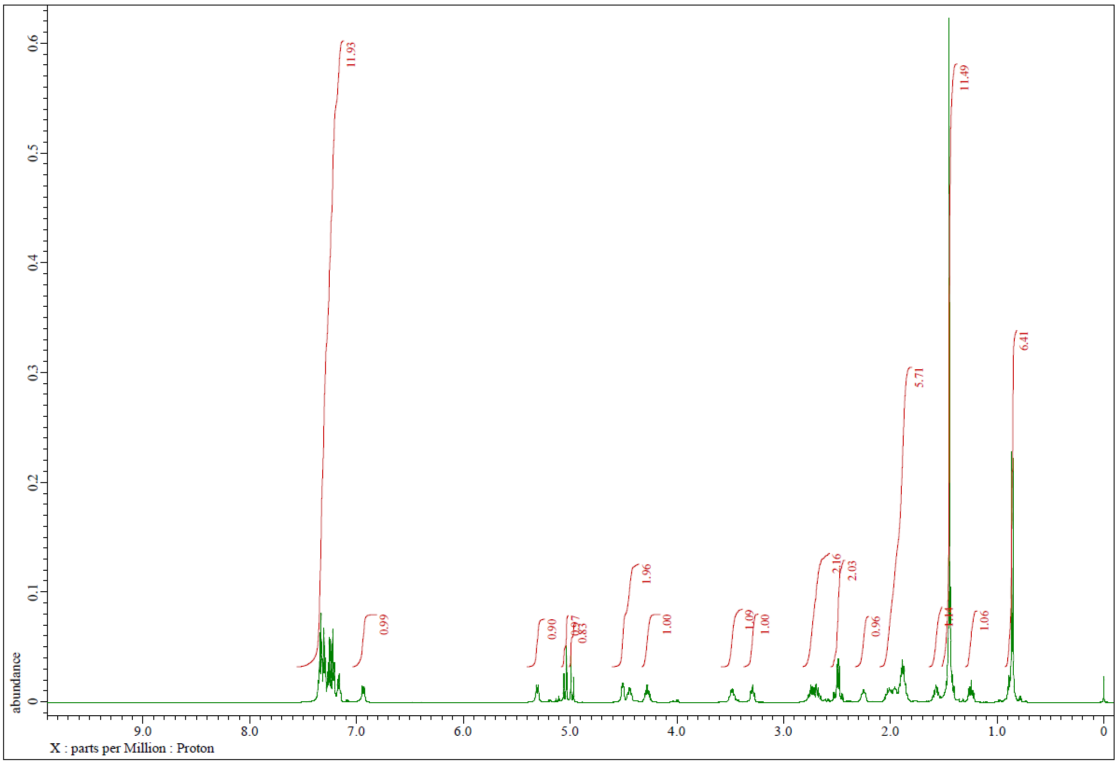


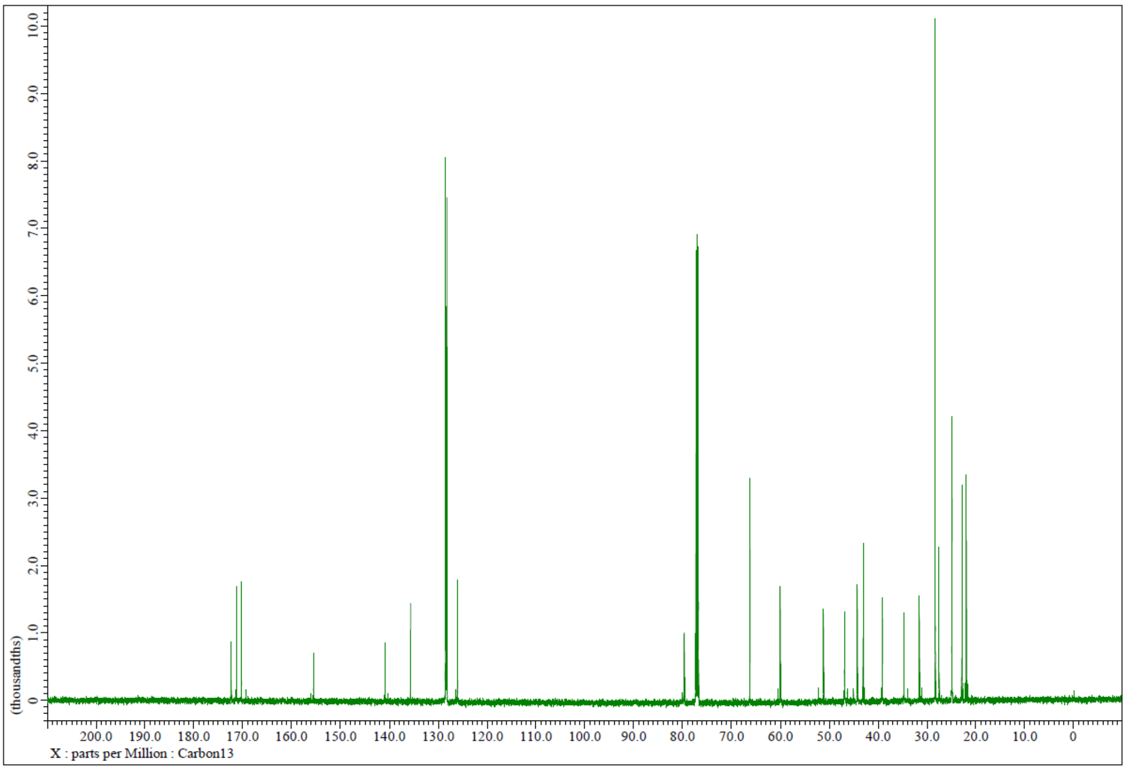
abundance
X : parts per Million : 1H



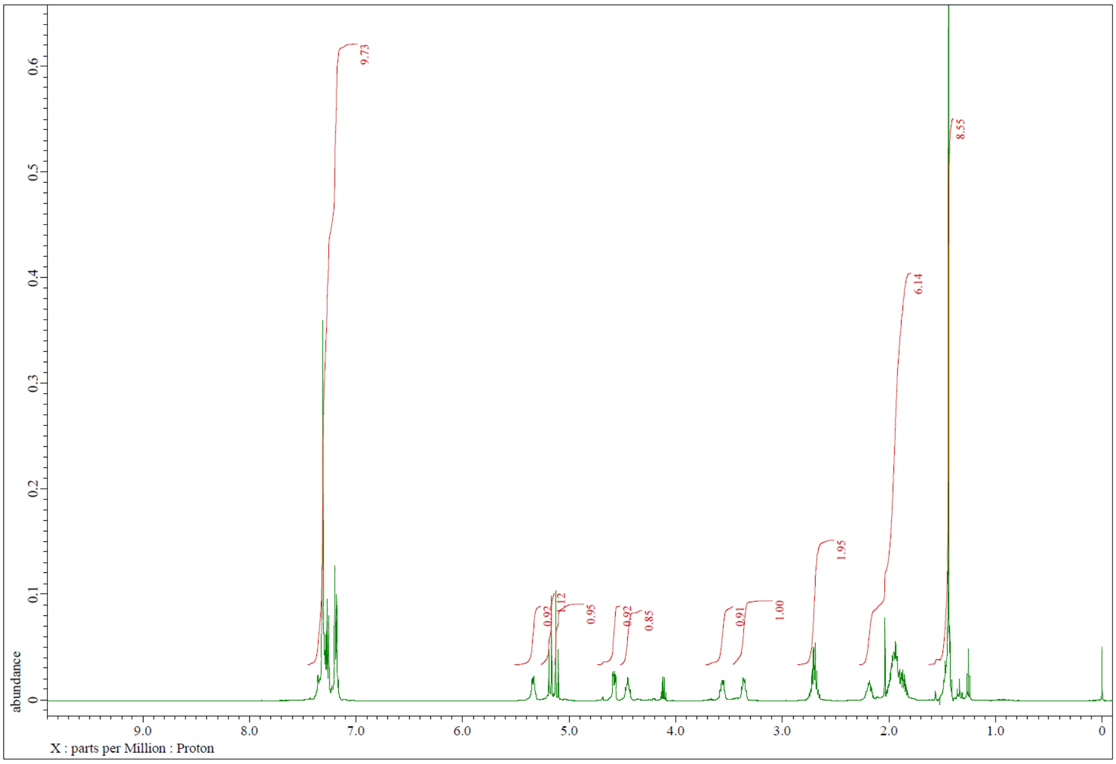
abundance
X : parts per Million : 13C

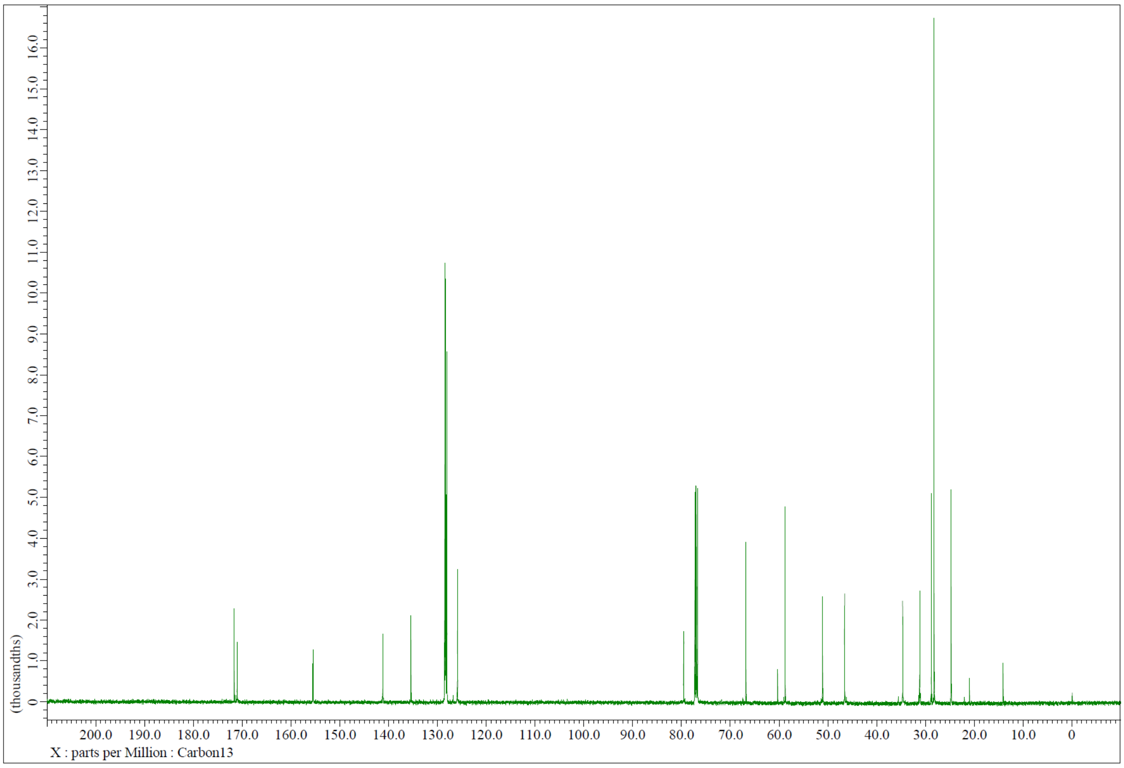
3n


abundance
X : parts per Million : Proton
11.93
0.99
0.90
0.83
1.96
1.00
1.09
1.00
2.16
2.03
0.96
5.71
11.49
1.06
6.41

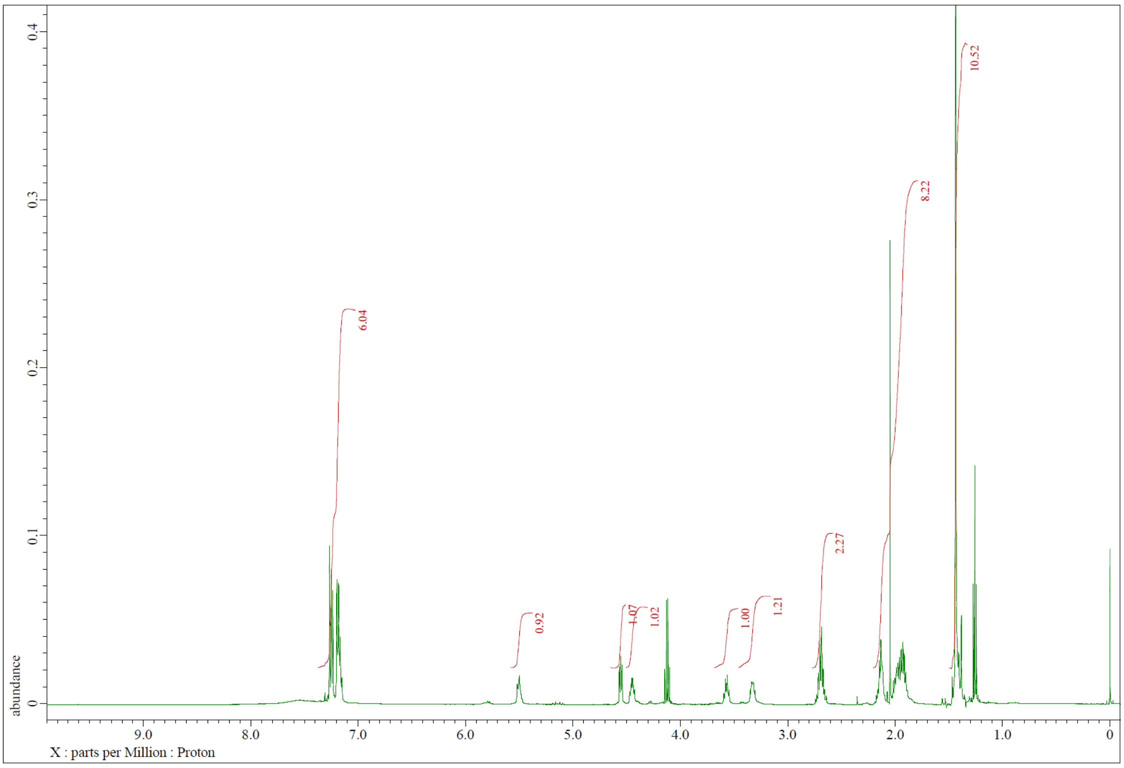

(thousandths)
X : parts per Million : Carbon13

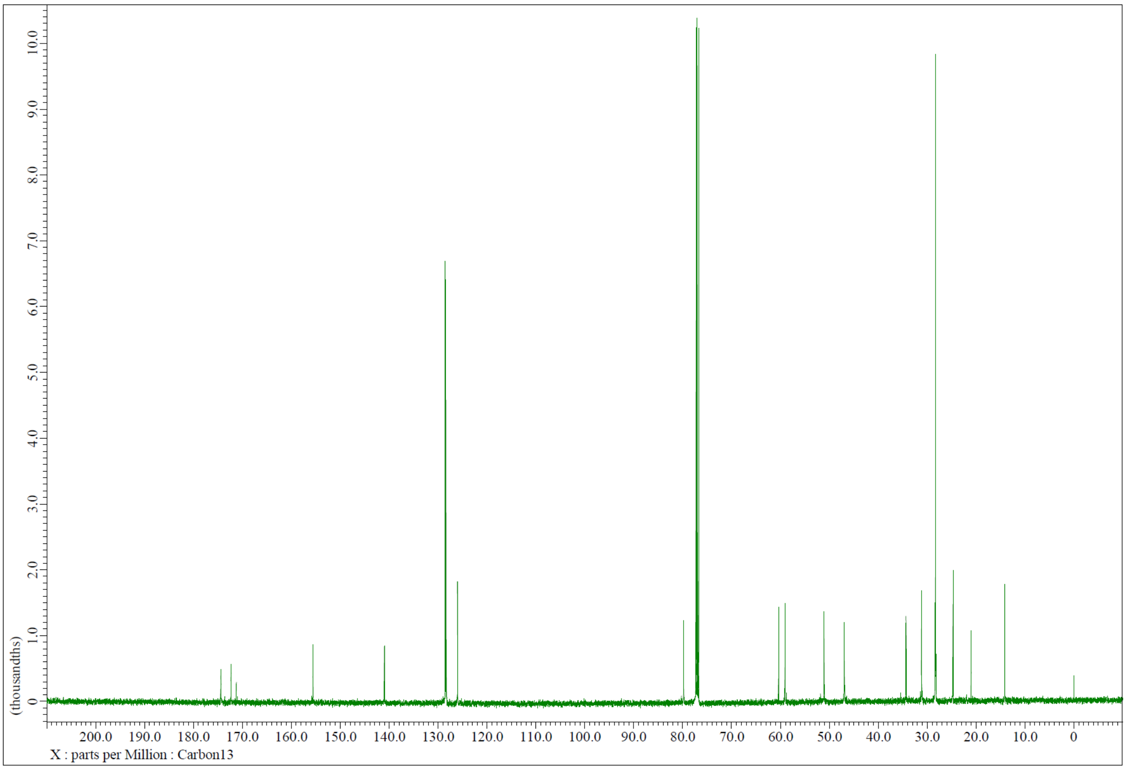
Boc-homoPhe-Pro-OBn



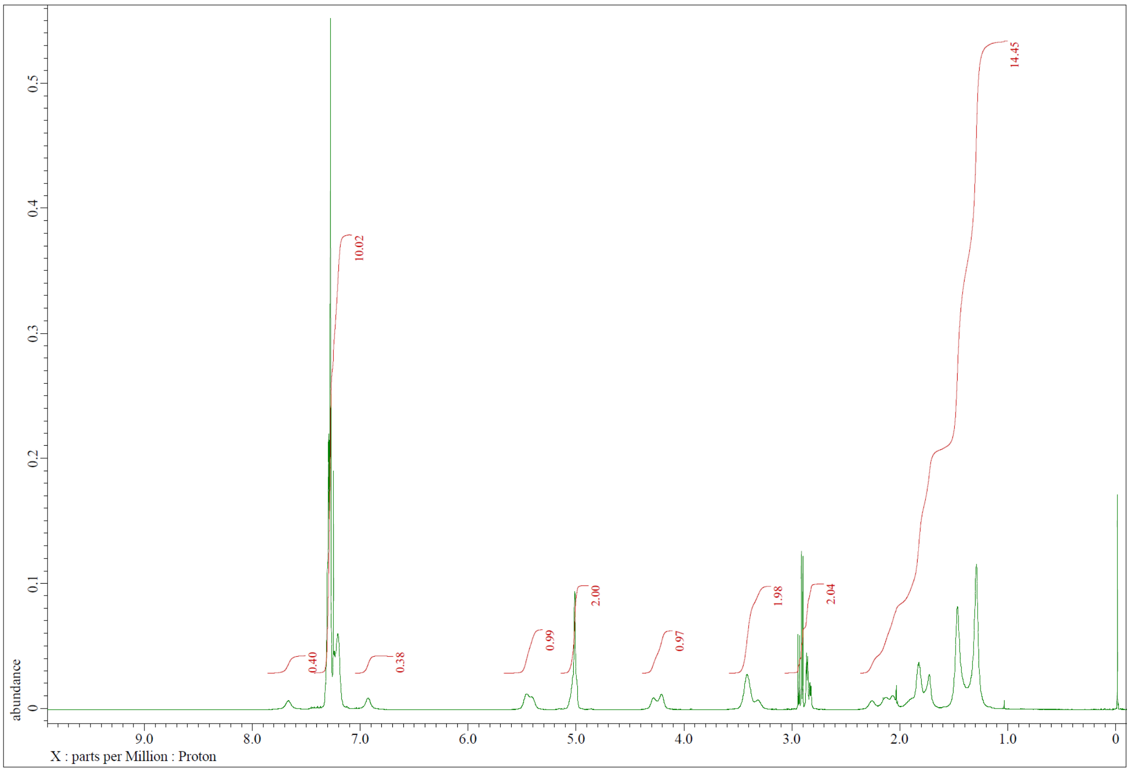


Boc-homoPhe-Pro-OH

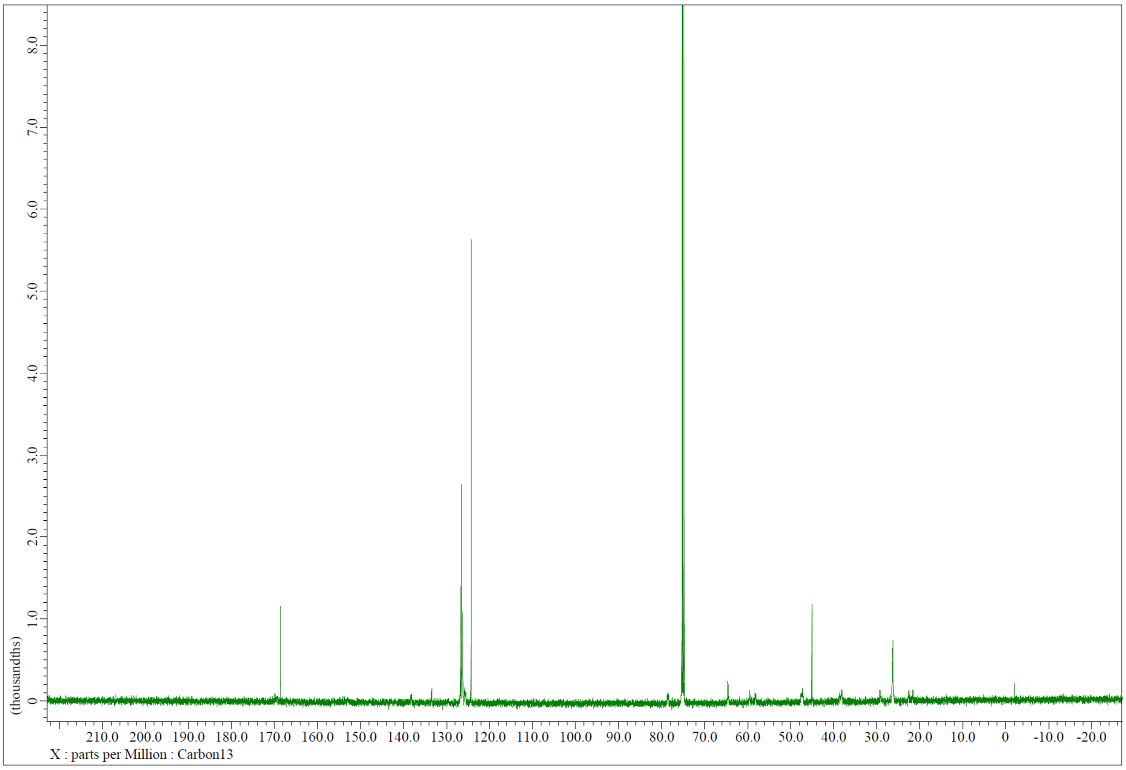




5a

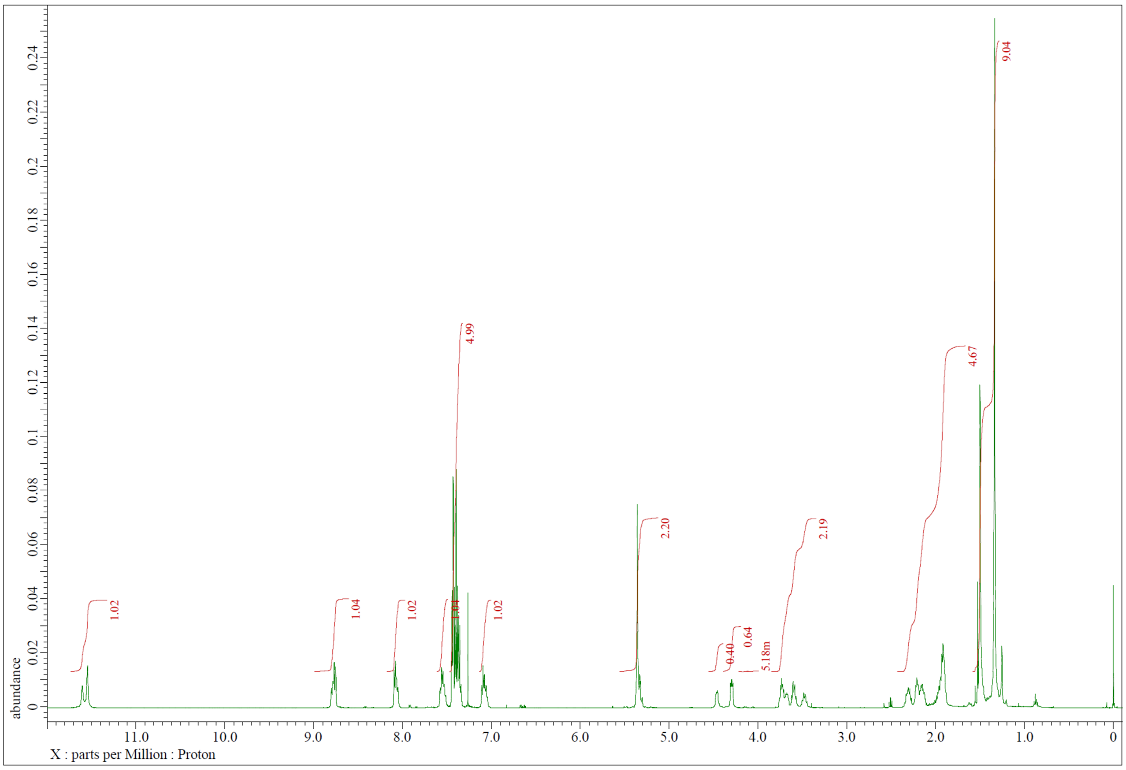


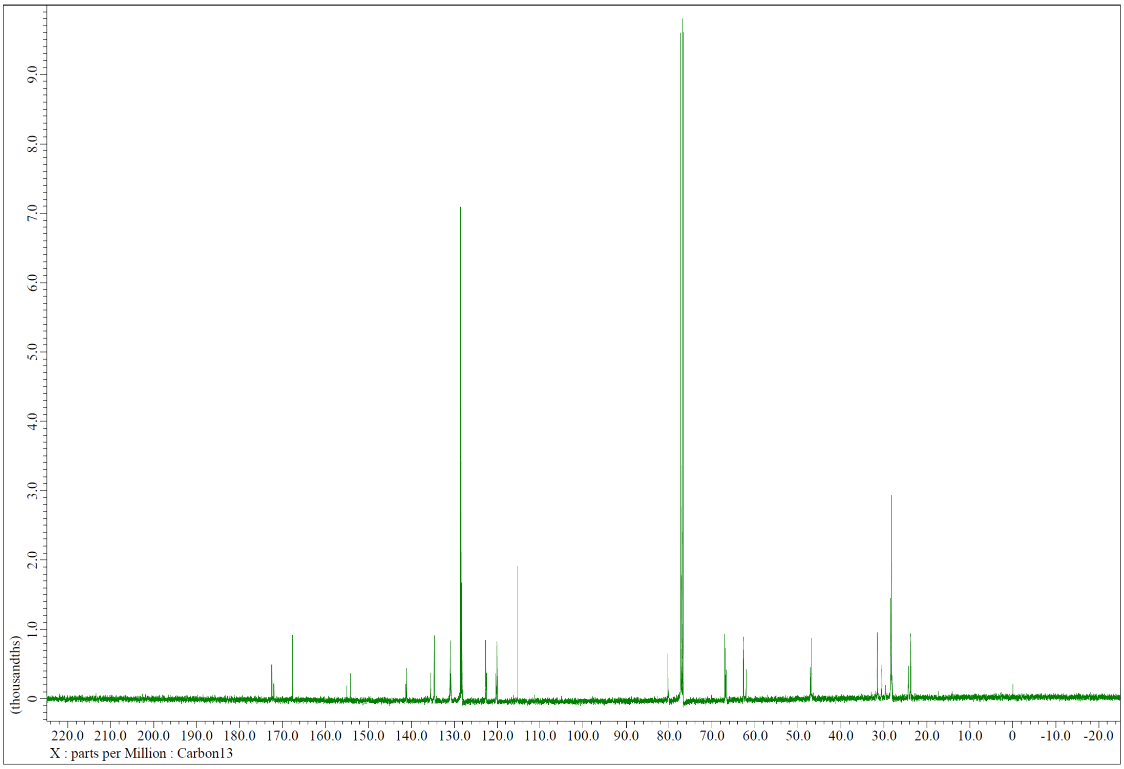
abundance
0
0.1
0.2
0.3
0.4
0.5
0.40
10.02
0.38
0.99
2.00
0.97
1.98
2.04
14.45
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
X : parts per Million : Proton



(thousandths)
0
1.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
210.0
200.0
190.0
180.0
170.0
160.0
150.0
140.0
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0
-10.0
-20.0
X : parts per Million : Carbon13

5b


abundance
X : parts per Million : Proton


(thousandths)
X : parts per Million : Carbon13