# Supporting Information

# Fluorescence "turn-on" Lectin Sensors Fabricated by Ligand-assisted Labeling Probes for Detecting Protein-Glycoprotein Interactions

Pei-Jhen Li,[†] Mohammed Tarigue Anwar,[†] Chen-Yo Fan,[†] Duane S. Juang,[†] Hsin-Yi Lin,[‡] Tsung-Che Chang,[†] Sachin Kisan Kawade,[†] Hsiang-Jung Chen,[†] Yu-Ju Chen,[‡] Kui-Thong Tan,[†] Chun-Cheng Lin[†,§,*]

[†]Department of Chemistry, National Tsing Hua University, Hsinchu, Taiwan

[‡]Institute of Chemistry, Academia Sinica, Taipei, Taiwan

[§]Department of Medicinal and Applied Chemistry, Kaohsiung Medical University, Kaohsiung, Taiwan

**E-mail:** cclin66@mx.nthu.edu.tw

**TEL**: +886 3 573 1287

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## Materials and Methods

All solvents were dried and distilled by standard techniques. Dichloromethane (DCM), toluene, and acetonitrile (ACN) were distilled from calcium hydride under $N_2$. Tetrahydrofuran (THF) was distilled from sodium under $N_2$ prior to use. The chemicals for the synthesis were all obtained from Acros Organics, Merck, Fluka, or Sigma-Aldrich and used without further purification unless otherwise noted. Bovine serum albumin (BSA, A9418), *Ricinus communis* agglutinin 120 (RCA120, L7886), ovalbumin (OVA, A5378), and monoclonal anti-biotin antibody conjugated with HRP (A0185) were purchased from Sigma-Aldrich. Protein Deglycosylation Mix II (P6044S) was purchased from New England Biolabs. Neuraminidase from *Arthrobacter ureafaciens* was purchased from Nacalai Tesque. LysoTracker™ Green DND-26 was purchased from Thermo Fisher Scientific.

All reactions were carried out in oven-dried glassware (104.0 °C) and performed under anhydrous conditions with $N_2$ unless indicated otherwise. The reactions were monitored by analytical thin-layer chromatography (TLC) on Merck silica gel 60 $F_{254}$ plates (0.25 mm). Detection was accomplished by examination under UV light (254 nm) and by staining with *p*-anisaldehyde, ninhydrin, cerium molybdate, or potassium permanganate staining solution. Silica gel column chromatography was performed using a forced flow of the indicated solvent on silica gel 60 (Merck). C18 reverse-phase silica column chromatography cartridges were purchased from Waters (SepPak Vac C18 cartridge 35c.c./10 g, 55-105 μm). $^{1}H$ and $^{13}C$ NMR spectra were recorded by Bruker AV-400, AV-600, Varian MR-400 or VNMRS-700 NMR spectrometers. Chemical shifts are expressed in ppm using residual $CDCl_3$ (7.24 ppm), $CD_3OD$ (3.31 ppm) as internal standard in $^{1}H$-NMR spectra. $^{13}C$-NMR spectra were recorded in either

$CDCl_3$, $CD_3OD$ or $D_2O$ at a 100 MHz, using the central resonances of $CDCl_3$ (77.0 ppm) and $CD_3OD$ (49.0 ppm) as the internal references. Multiplicities are reported using the following abbreviations: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, br = broad. *J* = coupling constant values are expressed in Hertz. High resolution mass spectra (HRMS) were recorded on Varian 901-FTMS. Fluorescence spectra were recorded using a TECAN Infinite M200Pro multimode plate reader. Fluorescence imaging was carried out using a laser scanning confocal microscope (LSM 700, Zeiss, Germany). For the Cy5 channel, images were taken using a 555 nm excitation laser with emission from 590 to 700 nm. For the Hoechst channel, a 405 nm laser with a SP490 emission filter. For LysoTracker™ Green DND-26, images were taken using a 488 nm laser with a BP490-555 emission filter.

**Cloning and Overexpression of Enzymes**

**Bacterial strains, plasmids, and materials:** Chemical competent *E. coli* BL21(DE3) were purchased from Yeastern Biotechnology (Taipei, Taiwan). Vector plasmid pTXB1 and chitin bead were purchased from NEB (Ipswich, MA). Bicinchoninic acid (BCA) protein assay kit was obtained from Thermo (Waltham, MA).

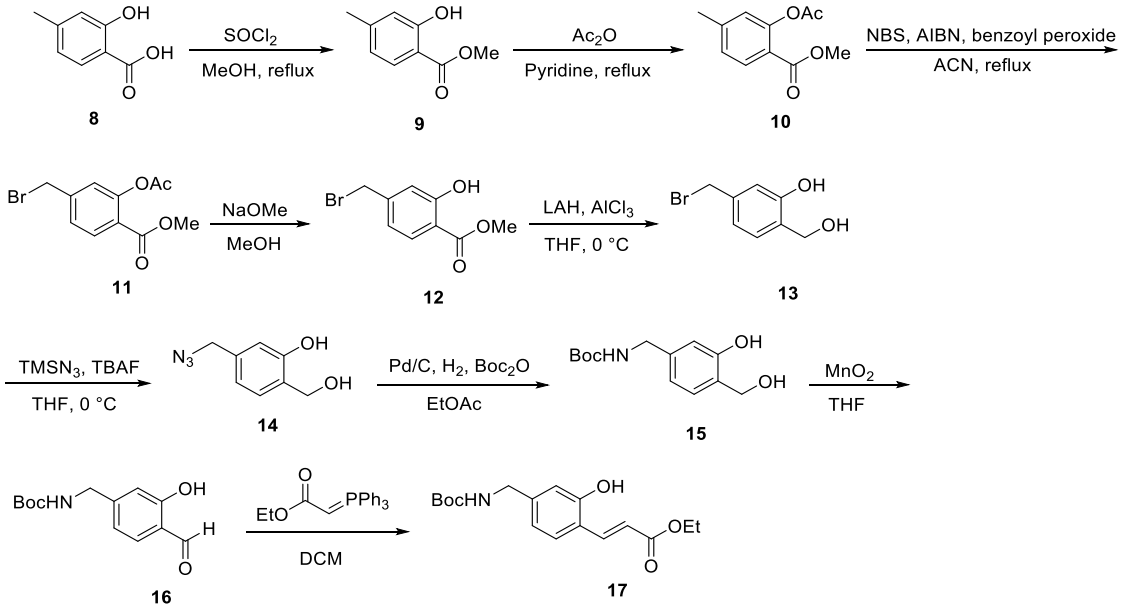
Pd2,6ST was cloned and overexpressed as previously reported procedures.[1] Gene encoded Pd2,6ST was cloned in pTXB1 and expressed in *E. coli* as a C-terminal intein fusion protein and purified by using the IMPACT™ system according to the instruction manual provided by the manufacturer (NEB).

**Protein Overexpression for Pd2,6ST**: The protein overexpression procedure is similarly as previous report.[1] Positive plasmid was selected and subsequently transformed into *E. coli* BL21(DE3) chemical competent cells. The *E. coli* BL21 (DE3)

harboring the recombinant plasmid was grown in LB rich medium containing ampicillin (100 $\mu$g/mL) at 37 °C until the $OD_{600}$ reached 0.5-0.8. Protein expression was then induced by adding 0.5 mM of IPTG (isopropyl-1-thio-$\beta$-D-galactopyranoside) followed by incubation at 16 °C for 16-20 h with vigorous shaking at 200 rpm in a shaking incubator (Firstek S300R).

**Protein Purification for Pd2,6ST**: The protein purification procedure is similarly as previous report.[1] Intein-fusioned target protein was purified from cell lysate using chitin bead. The cell lysate was collected and applied to a 6-mL chitin bead column, then washed with column buffer. The resin was then quickly washed with 1 column volume of the same buffer containing 80 mM DTT, and the effluent was reloaded. The column was clamped at both ends, and the intein tag was cleaved on-column from the fusion protein by incubating the column at 4 °C for 16 h. The purified protein was eluted using column buffer without DTT. The effluent was concentrated using a centrifugal filter device (Amicon Ultra, Millipore), added with glycerol as final concentration in 10%, divided into aliquots, and stored at -20 °C.

**Synthesis of Ligand-assisted imprinting probe**




**Scheme S1**. Synthesis of compound **17**.

Compound **9**

To a solution of 2-hydroxy-4-methylbenzoic acid **8** (2.00 g, 13.15 mmol) in dry MeOH (26.0 mL) was added thionyl chloride (1.4 mL, 19.73 mmol) at 0 ˚C. After being stirred at 0 ˚C for 10 min, the reaction mixture was heated to reflux for 8 hours then cooled to room temperature. MeOH was removed under reduced pressure, and the resulting residue was diluted with ethyl acetate, washed with water (×3) and brine, and dried over $MgSO_4$. The organic solvent was removed *in vacuo* and the resulting crude brown oil was purified via silica gel column chromatography to yield compound **9** (2.01 g, 11.84 mmol, 90% yield) as a yellow oil. TLC (EtOAc:Hexanes, 1:3 v/v): $R_f$ = 0.75; $^1$H NMR (400 MHz, $CDCl_3$) δ10.68 (s, 1H), 7.69 (d, $J$ = 8.1 Hz, 1H), 6.77 (s,1H), 6.67 (d, $J$ = 8.1 Hz, 1H), 3.91 (s, 3H), 2.32 (s, 3H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ170.4, 161.4, 146.8, 129.5, 120.2, 117.5, 109.6, 51.9, 21.6; HRMS (ESI-APCI, *m/z*) calculated for $C_9H_{11}O_3$ $[M+H]^+$:167.0708, found: 167.0704.

Compound **10**

To a solution of compound **19** (1.00 g, 6.02 mmol) in pyridine (4.8 mL) was added acetic anhydride (3.10 g, 30.11 mmol). The reaction mixture was refluxed for 1 hour. The solvent was removed *in vacuo* and the resulting residue was diluted with ethyl acetate, then washed with 1N $HCl_{(aq)}$ (×3) and brine, dried over $MgSO_4$, filtered, and concentrated *in vacuo* to give the crude product. The crude product was purified by silica gel column chromatography to give compound **10** (1.26 g, 6.00 mmol, 99.7% yield) as a yellow oil. TLC (EtOAc:Hexanes, 1:3 v/v): $R_f$ = 0.38; $^1$H NMR (400 MHz, $CDCl_3$) δ 7.89 (d, *J* = 8.0 Hz, 1H), 7.09 (d, *J* = 8.0 Hz, 1H), 6.89 (s,1H), 3.83 (s, 3H), 2.37(s, 3H), 2.32 (s, 3H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 169.6, 164.6, 150.5, 145.0, 131.5, 126.6, 124.1, 119.8, 51.8, 21.1, 20.7; HRMS (ESI-TOF, *m/z*) calculated for $C_{11}H_{12}NaO_4$ $[M+Na]^+$: 231.0633, found: 231.0637.

Compound **11**

To a stirred mixture of compound **10** (300.0 mg, 1.44 mmol) and NBS (310.0 mg, 1.73 mmol) in dry ACN (10.0 mL), AIBN (1.0 mg, 6 μmol), and benzoyl peroxide (12.0 mg, 5 μmol) were added and the reaction mixture was refluxed for 3 hours. ACN was removed under reduced pressure and the resulting residue was diluted with ethyl acetate. The organic layer was washed with sat. $Na_2S_2O_{3(aq)}$ (×3) and brine, dried over $MgSO_4$, and filtered. After concentration of the filtrate, the resulting crude product was purified by silica gel column chromatography to afford compound **11** (330.0 mg, 1.15 mmol, 80% yield) as a white solid. TLC (EtOAc:Hexanes, 1:5 v/v): $R_f$ = 0.35; $^1$H NMR (400 MHz, $CDCl_3$) δ 7.95 (d, *J* = 8.1 Hz, 1H), 7.29 (dd, *J* = 8.1, 1.7 Hz, 1H), 7.11 (d, *J* = 1.7 Hz, 1H), 4.41 (s, 2H), 3.83 (s, 3H), 2.32 (s, 3H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ169.0, 163.9, 150.4, 143.6, 131.7, 126.1, 124.0, 122.5, 51.8, 30.9, 20.5; HRMS (ESI-TOF, *m/z*) calculated for $C_{11}H_{11}BrNaO_4$ $[M+Na]^+$: 308.9738, found: 308.9739.

Compound **12**

To a solution of compound **11** (1.69 g, 5.91 mmol) in dry MeOH (29.0 mL) was added sodium methoxide (0.48 g, 8.89 mmol) at 0 ˚C. After being stirred at 0 ˚C for 10 min, the reaction mixture was warmed to room temperature and stirred for 3 hours. 1N $HCl_{(aq)}$ was added to neutralize the reaction mixture at 0 ˚C. The solvent was removed under reduced pressure and the resulting residue was diluted with ethyl acetate. The organic layer was washed with water (×3) and brine, dried over $MgSO_4$, and filtered. After concentration of the filtrate, the resulting crude product was purified by silica gel column chromatography to afford compound **12** (1.30 mg, 5.31 mmol, 90% yield) as a white solid. TLC (EtOAc:Hexanes, 1:5 v/v): $R_f$ = 0.44; $^1$H NMR (400 MHz, $CDCl_3$) δ 10.77 (s, 1H), 7.79 (d, *J* = 8.2 Hz, 1H), 6.98 (d, *J* = 1.6 Hz, 1H), 6.98 (dd, *J* = 8.2, 1.6 Hz, 1H), 4.38 (s, 2H), 3.93 (s, 3H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ170.1, 161.5, 145.5, 130.4, 119.8, 117.8, 112.1, 52.4, 32.0; HRMS (ESI-TOF, *m/z*) calculated for $C_9H_8BrO_3$ $[M-H]^-$: 242.9657, found: 242.9652.

Compound **13**

$AlH_3$ was generated *in situ* by the addition of LAH (1.65 g, 12.29 mmol) (divided into five portions) slowly to the solution of $AlCl_3$ (11.58 g, 24.59 mmol) in dry THF (48.0 mL) at 0 ˚C. Compound **12** (3.53 g, 14.47 mmol) was dissolved in dry THF (24.0 mL) at 0 ˚C and then was transferred to the activated $AlH_3$ solution slowly at 0 ˚C. After being stirred at 0 ˚C for 10 min, the reaction mixture was warmed to room temperature and stirred for 2 hours. The reaction was quenched by addition of silica gel at 0 ˚C and water was added until the bubble generation ceases. The solvent was removed under reduced pressure and the resulting residue was purified by silica gel column chromatography to afford compound **13** (2.94 g, 13.60 mmol, 94% yield) as a white solid. TLC (EtOAc:Hexanes, 1:3 v/v): $R_f$ = 0.12; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.23 (d, *J* = 7.7 Hz, 1H), 6.87 (dd, *J* = 7.7, 1.6 Hz, 1H), 6.83 (d, *J* = 1.6 Hz, 1H), 4.63 (s 2H), 4.47 (s, 2H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 156.2, 139.7, 129.4, 129.0, 121.1, 116.4,

60.6, 34.1; HRMS (ESI-TOF, *m/z*) calculated for $C_8H_8BrO_2$ [M-H]$^-$: 214.9708, found: 214.9707.

Compound **14**

To a solution of compound **13** (560.0 mg, 2.59 mmol) in dry THF (8.4 mL) was added azidotrimethylsilane (450.0 mg, 3.91 mmol) and TBAF (1M in THF, 3.9 mL, 3.89 mmol) at room temperature. After being stirred for 2 hours, the reaction mixture was worked up by water at 0 ˚C. The solvent was removed under reduced pressure and the resulting residue was diluted with ethyl acetate. The organic layer was washed with water and brine, dried over $MgSO_4$, and filtered. After concentration of the filtrate, the resulting crude product was purified by silica gel column chromatography to afford compound **14** (0.45 g, 2.51 mmol, 97% yield) as a white solid. TLC (EtOAc:Hexanes: 1:2 v/v): $R_f$ = 0.26; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.28 (d, *J* = 7.7 Hz, 1H), 6.80 (dd, *J* = 1.6, 7.7 Hz, 1H), 6.77 (d, *J* = 1.6 Hz, 1H), 4.65 (s, 2H), 4.25 (s, 2H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 156.1, 136.7, 128.3, 124.9, 119.7, 116.1, 63.8, 54.3; HRMS (ESI-TOF, *m/z*) calculated for $C_8H_8N_3O_2$ [M-H]$^-$: 178.0617, found: 178.0611.

Compound **15**

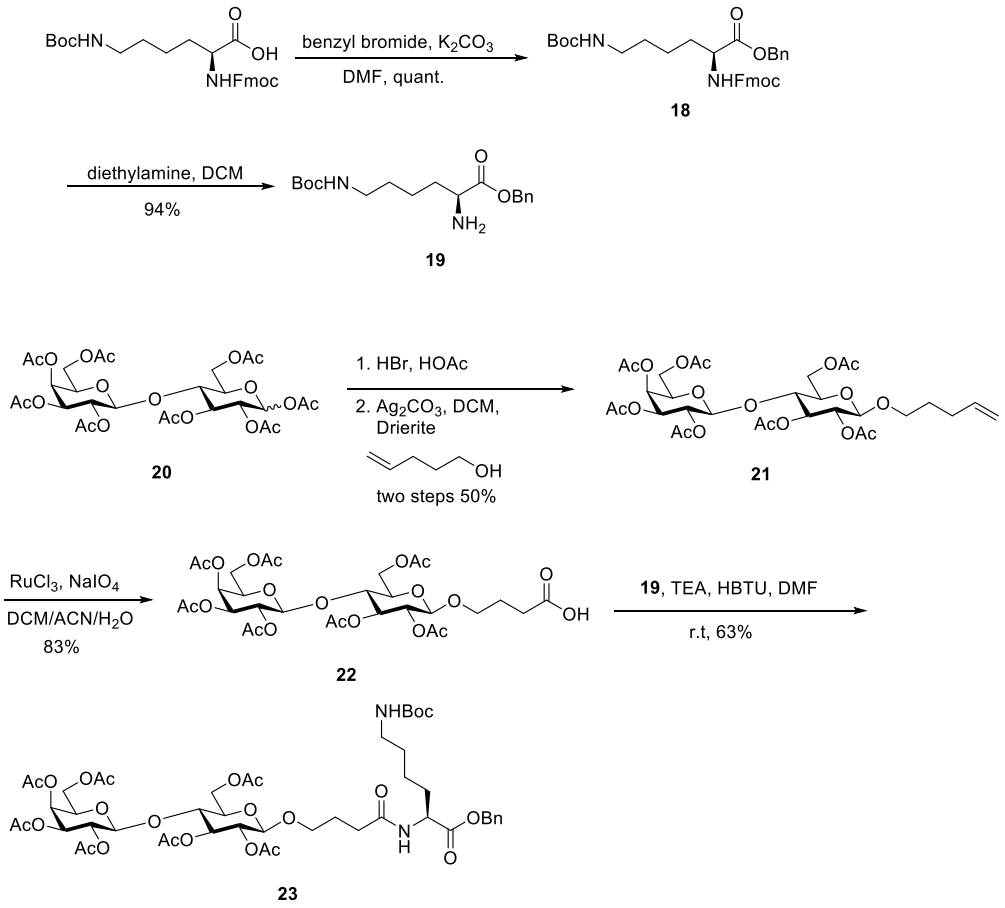
To a solution of compound **14** (620.0 mg, 3.46 mmol) in EtOAc (70.0 mL) was added 10% Pd/C (124 mg, 1.17 mmol) and di-*tert*-butyl dicarbonate (450.0 mg, 3.91 mmol) under $N_{2(g)}$ at room temperature. The reaction mixture was purged with $H_{2(g)}$ for 8 hr. The solvent was removed under reduced pressure and the resulting residue was purified by silica gel column chromatography to give compound **15** (780.0 mg, 3.08 mmol, 89% yield) as a white solid. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f$ = 0.36; $^1$H NMR (400 MHz, $CDCl_3$) δ 6.96 (d, *J* = 7.6 Hz, 1H), 6.74 (s, 1H), 6.70 (d, *J* = 7.6 Hz, 1H), 4.86 (br, 1H), 4.78 (s, 2H), 4.18 (d, *J* = 5.6 Hz, 2H), 1.44 (s, 1H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 158.4, 156.1, 141.2, 129.3, 127.1, 119.0, 114.6, 80.1, 60.9, 44.7, 28.8 (3C); HRMS (ESI-TOF, *m/z*) calculated for $C_{13}H_{18}NO_4$ [M-H]$^-$: 252.1236, found: 252.1235.

Compound **16**

To a solution of compound **15** (100.0 mg, 0.40 mmol) in dry THF (2.0 mL) was added manganese dioxide (515.0 mg, 5.92 mmol) under $N_{2(g)}$ at room temperature. After being stirred at room temperature for 2 hours, the solvent was removed under reduced pressure. The resulting residue was purified by silica gel column chromatography to yield compound **16** (100.0 mg, 0.39 mmol, 98% yield) as a white solid. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f$ = 0.62; $^1$H NMR (400 MHz, $CDCl_3$) δ 11.03, 9.83, 7.48 (d, *J* = 7.9 Hz, 1H), 6.90 (d, *J* = 7.9 Hz, 1H), 6.9 (s, 1H), 4.96 (s, 1H), 4.31 (d, *J* = 5.6 Hz, 2H), 1.44 (s, 9H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 195.9, 161.8, 155.8, 149.4, 133.9, 119.6, 118.5, 115.5, 79.8, 44.2, 20.3 (3C); HRMS (ESI-TOF, *m/z*) calculated for $C_{13}H_{16}NO_4$ $[M-H]^-$: 250.1079, found: 250.1073.

Compound **17**

To a solution of compound **16** (410.0 mg, 1.63 mmol) in dry $CH_2Cl_2$ (8.2 mL) was added (carbethoxymethylene)triphenylphosphorane (852.6 mg, 2.45 mmol) under $N_{2(g)}$ at room temperature. After being stirred for 3 hours, the solvent was removed under reduced pressure. The resulting residue was purified by silica gel column chromatography to afford compound **17** (520.0 mg, 1.62 mmol, 99% yield) as a white solid. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f$ = 0.40; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.92 (d, *J* = 16.2 Hz, 1H), 7.44 (d, *J* = 7.9 Hz, 1H), 6.78 (s, 1H), 6.78-6.75 (m, 2H), 4.22 (q, *J* = 7.1 Hz, 2H), 4.17 (s, 2H), 2.31 (t, *J* = 7.4 Hz, 2H), 1.89 (q, *J* = 7.1 Hz, 2H), 1.46 (s, 9H), 1.32 (t, *J* = 7.1 Hz, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 169.6, 158.5, 158.4, 145.2, 141.9, 130.2, 121.3, 119.4, 118.0, 115.4, 80.3, 61.4, 44.7, 28.8 (3C), 14.6; HRMS (ESI-TOF, *m/z*) calculated for $C_{17}H_{22}NO_5$ $[M-H]^-$: 320.1498, found: 320.1491.




**Scheme S2.** Synthesis of ligand intermediate compound **23**.

Compound **18**

To a solution of Fmoc-Lys(Boc)-OH (1.00 g, 2.13 mmol) in dry DMF (14.0 mL) was added potassium carbonate (1.40 g, 4.30 mmol) and benzyl bromide (0.51 mL, 4.27 mmol) under $N_{2(g)}$ at room temperature at room temperature. After being stirred for 1 hour, the solid residue of potassium bicarbonate was filtered and the solvent was removed under reduced pressure. The resulting residue was purified by silica gel column chromatography to give compound **18** (1.18 g, 2.11 mmol, 99% yield) as a semi-solid. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f$ = 0.50; $^1$H NMR (400 MHz, $CDCl_3$) δ7.75 (d, *J* = 7.4 Hz, 2H), 7.59 (d, *J* = 7.1 Hz, 2H), 7.40-7.27 (m, 9H), 5.47 (s, 1H), 5.26-5.12 (m, 2H), 4.54 (s, 1H), 4.43-4.36 (m, 3H), 4.20 (t, *J* = 6.6 Hz 1H), 3.05 (s, 2H), 1.85-1.43 (m, 15H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 172.2, 156.0, 155.9, 143.8, 143.6,

141.2 (2C), 135.2, 128.5 (2C), 128.4 (2C), 128.2 (2C), 127.6 (2C), 127.0 (2C), 125.0 (2C), 119.86 (2C), 67.03, 66.92, 53.7, 47.1, 40.0, 32.0, 29.5, 28.3 (3C), 22.2; HRMS (ESI-TOF, *m/z*) calculated for $C_{33}H_{38}N_2O_6Na$ $[M+Na]^+$: 581.2628, found 581.2634

Compound **21**

To a flask with lactose octaacetate **20** (2.00 g, 2.95 mmol) was added 33% HBr in AcOH (10.0 mL). The mixture was stirred at room temperature for 1 h. The solution was poured into ice water and then extracted with dichloromethane (DCM). The organic layer was washed with saturated $NaHCO_{3(aq)}$, dried over $MgSO_4$, filtered, and concentrated *in vacuo* to give the lactosyl bromide. To a mixture of pent-4-en-1-ol (0.6 ml, 8.35 mmol), $Ag_2CO_3$ (1.63 g, 5.92 mmol), and 4 ÅMS in DCM (15 mL) was added a solution of lactosyl bromide in DCM (15 mL) under $N_{2\,(g)}$ at room temperature. After being stirred for 8 h, the reaction mixture was filtered through celite. The filtrate was concentrated and the resulting residue was purified by silica gel column chromatography to give compound **21** (1.20 g, 1.71 mmol, 58% yield) as a white solid. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f = 0.26$; $^1$H NMR (400 MHz, $CDCl_3$) δ5.56-5.51 (m, 1H), 5.12 (dd, $J$ = 3.3, 0.8 Hz, 1H), 4.97 (dd, $J$ = 9.4 Hz, 1H), 4.87 (dd, $J$ = 10.4, 7.8 Hz, 1H), 4.89-4.73 (m, 3H), 4.66 (dd, $J$ = 9.5, 8.0 Hz, 1H), 4.36 (d, $J$ = 7.8 Hz, 1H), 4.29-4.24 (m, 2H), 3.93-3.86 (m, 3H), 3.76 (dd, $J$ = 7.0 Hz, 1H), 3.61 (t, $J$ = 8.9 Hz, 2H), 3.45-3.41 (m, 1H), 3.31-3.25 (m, 1H), 1.93 (s, 3H), 1.90 (s, 3H), 1.84-1.82 (m, 12H), 1.73 (s, 3H), 1.46-1.40 (m, 2H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 169.84, 169.76, 169.6, 169.4, 169.3, 169.0, 168.6, 137.3, 114.6, 100.4, 100.0, 75.9, 72.4, 72.1, 71.2, 70.5, 70.1, 68.7 (2C), 66.3, 61.7, 60.5, 29.3, 28.1, 20.3, 20.2, 20.1, 20.07, 20.05, 20.04, 19.4; HRMS (ESI-TOF, *m/z*) calculated for $C_{31}H_{44}O_{18}Na$ $[M+Na]^+$: 727.2425, found: 727.2415

Compound **22**

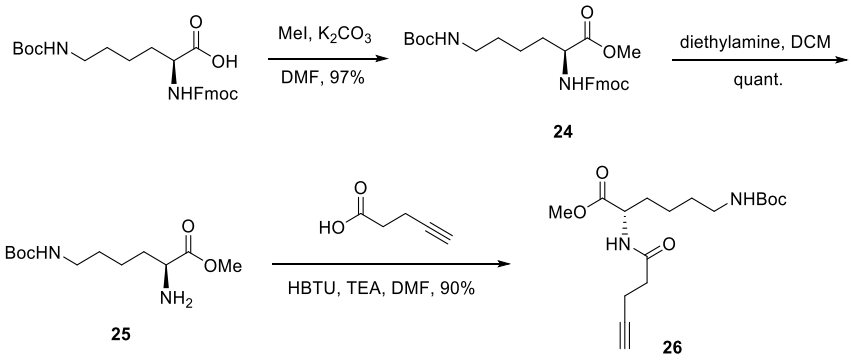
To a solution of compound **21** (1.25 g, 1.85 mmol) in DCM (5.3 mL) / acetonitrile (5.3

mL) / $H_2O$ (8.0 mL) was added $RuCl_3$ (76.7 mg, 0.37 mmol) and $NaIO_4$ (1.06 g, 5.00 mmol) at room temperature. After being vigorously stirred at room temperature for 2 h, more $NaIO_4$ (0.53 g, 2.49 mmol) was added and the mixture was stirred for another 2 h at room temperature. The reaction mixture was concentrated to dryness *in vacuo*, and then extracted with ethyl acetate. The organic phase was dried over $MgSO_4$, filtered, and then concentrated *in vacuo* to give crude product. The residue was purified by silica gel column chromatography to give product **22** (0.93 g, 1.30 mmol, 70% yield) as a white solid TLC (EtOAc): $R_f$ = 0.40; $^1$H NMR (400 MHz, $CDCl_3$) δ 5.30 (dd, *J* = 3.4, 0.7 Hz, 1H), 5.14 (dd, *J* = 9.2 Hz, 1H), 5.05 (dd, *J* = 10.4, 7.9 Hz, 1H), 4.92 (dd, *J* = 10.4, 3.4 Hz, 1H), 4.84 (dd, *J* = 9.5, 8.0 Hz, 1H), 4.45-4.40 (m, 3H), 4.11-4.00 (m, 3H), 3.83 (t, *J* = 6.9 Hz, 2H), 3.75 (dd, *J* = 9.5 Hz, 1H), 3.57-3.53 (m, 1H), 3.51-3.47 (m, 1H), 2.36 (t, *J* = 7.2 Hz, 2H), 2.10 (s, 3H), 2.07 (s, 3H), 2.02 (s, 3H), 2.00 (s, 9H), 1.92 (s, 3H), 1.84 (quintet, 3H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 177.8, 170.4, 170.3, 170.1, 170.0, 169.8, 169.7, 169.1, 100.9, 100.4, 76.1, 72.7, 72.5, 71.6, 70.9, 70.6, 69.1, 68.5, 66.6, 61.9, 60.8, 30.0, 24.4, 20.75, 20.71, 20.6, 20.05 (2C), 20.04, 14.1; HRMS (ESI-TOF, *m/z*) calculated for $C_{30}H_{42}O_{20}Na$ $[M+Na]^+$: 745.2167, found: 745.2159

Compound **23**

To a stirred solution of compound **18** (300.0 mg, 0.54 mmol) in DCM (5.3 mL) was added diethylamine (5.3 mL). After being stirred at room temperature for 2 hours, the reaction mixture was purified by silica gel column chromatography to afford compound **19** (170.0 mg, 0.51 mmol) as a yellow syrup. TLC (MeOH:DCM, 1:5 v/v): $R_f$ = 0.40; HRMS (ESI-TOF) calculated for $C_{18}H_{29}N_2O_4$ $[M+H]^+$ = 337.2127, found 337.2129. To a solution of the above compound **19** (140.0 mg), compound **22** (300 mg, 0.42 mmol), and HBTU (262.5 mg, 0.69 mmol) in dry DMF (3.3 mL) at room temperature under a nitrogen atmosphere. After being stirred at room temperature for 5 h, the mixture was concentrated to dryness *in vacuo*, and ethyl acetate was added. The organic layer was

washed with brine, dried over $MgSO_4$, filtered, and then concentrated under reduced pressure to give the crude product. The residue was purified by column chromatography on silica gel to afford compound **23** (350.0 mg, 0.34 mmol, 63% yield) as a white syrup. TLC (EtOAc): $R_f$ = 0.55; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.35-7.31 (m, 5H), 6.22 (d, *J* = 7.5 Hz, 1H), 5.32 (dd, *J* = 2.6, 0.8 Hz, 1H), 5.19-5.06 (m, 4H), 4.94 (dd, *J* = 10.4, 3.4 Hz, 1H), 4.84 (dd, *J* = 9.6, 8.0 Hz, 1H), 4.61-4.56 (m, 1H), 4.53-4.45 (m, 3H), 4.13-4.03 (m, 3H), 3.85 (dd, *J* = 7.2 Hz, 1H), 3.77 (t, *J* = 9.6 Hz, 2H), 3.59-3.54 (m, 2H), 3.02 (d, *J* = 5.5 Hz, 2H), 2.27-2.22 (m, 2H), 2.13 (s, 3H), 2.10 (s, 3H), 2.04-2.01 (m, 12H), 1.9 (s, 3H), 1.86 (t, *J* = 6.8 Hz, 2H), 1.67-1.64 (m, 4H), 1.43-1.38 (m, 12H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 172.4, 172.3, 170.4, 170.3, 170.04, 170.00, 169.71, 169.66, 169.0, 156.0, 135.3, 128.5 (2C), 128.4, 128.2 (2C), 100.9, 100.5, 76.1, 72.7, 72.6, 71.7, 70.9, 70.6, 69.08, 69.06, 67.0, 66.6, 61.7, 60.7, 51.9, 40.0, 38.5, 32.3, 31.8, 29.4, 28.3 (3C), 25.4, 22.3, 20.8, 20.7, 20.6, 20.5 (3C), 20.4, 14.1; HRMS (ESI-TOF, *m/z*) calculated for $C_{48}H_{68}N_2O_{23}Na$ $[M+Na]^+$= 1063.4110, found 1063.4102.




**Scheme S3.** Synthesis of compound **26**.

Compound **24**

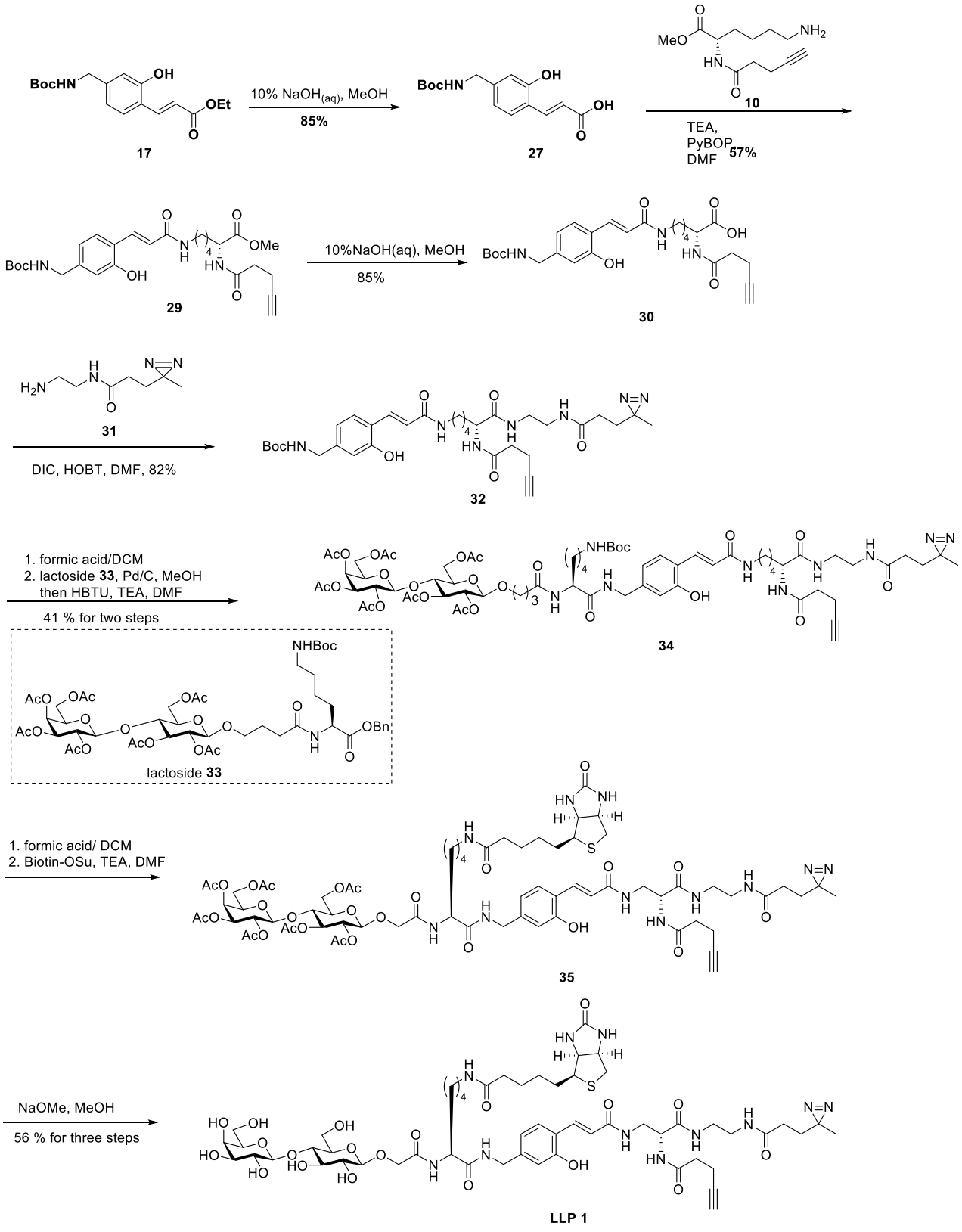
To a solution of Fmoc-Lys(Boc)-OH (1.00 g, 2.13 mmol) in dry DMF (17.8 mL) was added potassium carbonate (1.40 g, 4.30 mmol) and iodomethane (0.2 mL, 3.20 mmol) at room temperature. After being stirred for 1 hour, the solid residue of potassium

bicarbonate was filtered and the solvent was removed under reduced pressure. The resulting residue was purified by silica gel column chromatography to provide compound **24** (1.01 g, 2.09 mmol, 98% yield) as a semi-solid. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f$ = 0.45; $^1$H NMR (400 MHz, $CDCl_3$) δ 7.74 (d, *J* = 7.6 Hz, 2H), 7.58 (d, *J* = 7.0 Hz, 2H), 7.38 (dd, *J* = 7.4 Hz, 2H), 7.29 (ddd, *J* = 7.4, 1.1 Hz, 2H), 5.39 (s, 1H), 4.56 (s, 1H), 4.43-4.32 (m, 1H), 4.20 (t, *J* = 7.0 Hz, 2H), 3.73 (s, 3H), 3.83 (t, *J* = 5.7 Hz, 2H), 1.87-1.80 (m, 1H), 1.74-1.67 (m, 1H), 1.48-1.36 (m, 13H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 172.8, 156.0, 155.9, 143.7, 143.6, 141.1, 127.5, 126.9, 124.9, 119.8, 66.8, 53.6, 52.2, 47.0, 39.8, 31.8, 29.4, 28.2 (3C), 22.12; HRMS (ESI-TOF, *m/z*) calculated for $C_{27}H_{34}N_2NaO_6$ $[M+Na]^+$: 505.2315, found: 505.2315

Compound **26**

To a stirred solution of compound **24** (1.8 g, 3.73 mmol) in DCM (37 mL) was added diethylamine (3.7 mL). After being stirred at room temperature for 3 hours, the reaction solvent was removed and the resulting residue was purified by silica gel column chromatography to afford compound **25** (949.8 mg, 3.65 mmol) as yellow syrup. $R_f$ = 0.60 (MeOH/DCM = 1/5); HRMS (ESI-TOF, *m/z*) calculated for $C_{12}H_{25}N_2O_4$ $[M+H]^+$: 261.1814, found: 261.1816. To a solution of the above compound **25** (460.0 mg, 1.77 mmol), pent-4-ynoic acid (144.5mg, 1.47 mmol), and HBTU (1.1 g, 2.94 mmol) in dry DCM (29.4 mL) was stirred at room temperature under a nitrogen atmosphere. After being stirred at room temperature for 2 hours, the mixture was concentrated to dryness *in vacuo* and then ethyl acetate was added. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated under reduced pressure to give crude product. The residue was purified by silica gel column chromatography to afford compound **26** (480 mg, 1.41 mmol, 96% yield) as a colorless liquid. TLC (EtOAc:Hexanes, 2:1 v/v): $R_f$ = 0.41; $^1$H NMR (400 MHz, $CD_3OD$) δ; 4.39 (dd, *J* = 9.0, 5.0 Hz, 1H), 3.71 (s, 3H), 3.30 (t, *J* = 6.6 Hz, 2H), 2.50-2.43 (m, 4H), 2.28 (s, 1H),

1.87-1.79 (m, 1H), 1.73-1.64 (m, 1H), 1.48-1.38 (m, 13H); $^{13}$C NMR (100 MHz, $CDCl_3$) δ 172.7, 170.9, 156.0, 82.6, 78.7, 52.0, 51.7, 39.7, 34.6, 31.4, 29.1, 28.1 (3C), 22.1, 14.5; HRMS (ESI-TOF, *m/z*) calculated for $C_{17}H_{27}N_2O_5$ $[M-H]^-$: 339.1920, found: 339.1915.




**Scheme S4.** Synthesis of **LLP 1**.

Compound **29**

To a stirred solution of compound **17** (85.0 mg, 0.27 mmol) in EtOH (0.5 mL) was added 10% $NaOH_{(aq)}$ (1.0 mL). After being stirred at room temperature for 1.5 hours, the reaction mixture was neutralized by 1N $HCl_{(aq)}$ to pH = 2 at 0 ˚C. The EtOH was removed under reduced pressure and the resulting residue was extracted by EtOAc (20 mL × 3) and the collecting organic solvent was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated *in vacuo* to give compound **27** (68.3 mg, 0.23 mmol) as a colorless syrup. TLC (MeOH:DCM, 1:5 v/v): $R_f$ = 0.33; HRMS (ESI-TOF) calculated for $C_{15}H_{18}NO_5$ $[M-H]^-$: 292.1185, found: 292.1187. To a stirred solution of compound **26** (210 mg, 0.62 mmol) in DCM (1.2 mL) was added formic acid (3.5 mL). After being stirred at room temperature for 1.5 h, the residue was neutralized with Dowex resin 550 ($OH^-$), filtered, and concentrated *in vacuo* to give product **28** (140.0 mg, 0.58 mmol) as yellow syrup; TLC (MeOH:DCM, 1:5 v/v): $R_f$ = 0.15. To a solution of the above compound **27** (40.0 mg, 0.14 mmol), amine **28** (65.5 mg, 0.27 mmol), and PyBOP (142.0 mg, 0.27 mmol) in dry DMF (2.6 mL) was added triethylamine (0.1 mL, 68.22 mmol) at room temperature under a nitrogen atmosphere. After being stirred at room temperature for overnight, the mixture was concentrated to dryness *in vacuo* and ethyl acetate was added. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and then concentrated under reduced pressure to give the crude product. The residue was purified by silica gel column chromatography to afford compound **29** (40.0 mg, 0.077 mmol, 57% yield) as a white syrup. TLC (EtOAc:Hexanes, 2:1 v/v): $R_f$ = 0.40; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.78 (d, *J* = 15.9 Hz, 1H), 7.39 (d, *J* = 8.0 Hz, 1H), 6.77 (d, *J* = 1.4 Hz, 1H), 6.74 (dd, *J* = 1.4, 8.0Hz, 1H), 6.68 (d, *J* = 15.9 Hz, 1H), 4.40 (dd, *J* = 5.0, 9.1, 1H), 4.16 (s, 2H), 3.70 (s, 3H), 3.29-3.27 (m, 1H), 2.48-2.43 (m, 1H), 2.26 (dd, *J* = 2.5, 1H), 1.88-1.40 (m, 16H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 174.1 (x2), 169.6, 158.5,158.0, 144.2, 137.5, 130.0, 121.9, 121.2, 119.3, 115.3, 83.5, 80.2, 70.4, 53.6, 52.7, 44.7,40.1, 35.6, 32.1, 30.0, 28.8 (x3), 24.1, 15.6; HRMS (ESI-TOF,

*m/z*) calculated for $C_{27}H_{37}N_3NaO_7$ $[M+Na]^+$: 538.2529, found: 538.2526

Compound **32**

To a stirred solution of compound **29** (160.0 mg, 0.31 mmol) in MeOH (2.3 mL) was added 10% $NaOH_{(aq)}$ (4.6 mL). After being stirred at room temperature for 2 hours, the reaction mixture was neutralized by 1N $HCl_{(aq)}$ to pH = 2 at 0 ˚C. The EtOH was removed under reduced pressure and the resulting residue was extracted by EtOAc (20mL × 3). The collected organic solvent was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated *in vacuo* to give compound **30** (155.1 mg, 0.31 mmol) as colorless syrup. TLC (MeOH:DCM, 1:3 v/v): $R_f$ = 0.49; HRMS (ESI-TOF, *m/z*) calculated for $C_{26}H_{35}N_3NaO_7$ $[M+Na]^+$: 524.2327, found: 524.2377. To a solution of above compound (140.0 mg, 0.27 mmol), amine **31** (78.0 mg, 0.46 mmol), DIC (70.0 mg, 0.55 mmol), and HOBt (57.0 mg, 0.42 mmol) in dry DMF (5.6 mL) was stirred at room temperature under a nitrogen atmosphere for 2.5 hours. The mixture was concentrated to dryness *in vacuo* and ethyl acetate was added. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and then concentrated under reduced pressure to give the crude product. The residue was purified by silica gel column chromatography to afford compound **32** (140.0 mg, 0.21 mmol, 82% yield) as a semi-solid. TLC (EtOAc:Hexanes:MeOH, 2:1:0.3 v/v/v): $R_f$ = 0.23; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.79 (d, *J* = 15.9 Hz, 1H), 7.39 (d, *J* = 8.0 Hz, 1H), 6.78 (s, 1H), 6.75 (dd, *J* = 1.2, 8.0Hz, 1H), 6.68 (d, *J* = 15.9 Hz, 1H), 4.23 (dd, *J* = 5.0, 8.8, 1H), 4.16 (s, 2H), 3.32-3.28 (m, 5H), 2.49-2.47 (m, 4H), 2.29 (t, *J* = 2.24, 1H), 2.07 (dd, *J* = 9.20, 8.16, 2H), 1.84-1.45 (m, 18H), 1.00 (s, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 174.9, 174.8, 174.3, 169.7, 158.6, 158.0, 144.2, 137.6, 130.0, 121.9, 121.2, 119.3, 115.4, 83.7, 80.3, 70.4, 55.1, 44.8, 44.7,40.1, 40.0 (x2), 35.8, 32.5, 31.3, 30.1, 28.8 (x3), 26.3, 24.2, 19.7, 15.6; HRMS (ESI-TOF, *m/z*) calculated for $C_{33}H_{47}N_7NaO7$ $[M+Na]^+$: 676.3435, found: 676.3441

Compound **34**

To a stirred solution of compound **32** (70.0 mg, 0.11 mmol) in DCM (1.2 mL) was added formic acid (2.4 mL). After being stirred at room temperature for 2 hours, the residue was neutralized with Dowex resin 550 ($OH^-$), filtered, and concentrated *in vacuo* to give amine intermediate (155.1 mg, 0.31 mmol) as a yellow syrup. TLC (EtOAc:Hexanes:MeOH, 2:1:0.6 v/v/v): $R_f$ = 0.14; HRMS (ESI-TOF, *m/z*) calculated for $C_{28}H_{40}N_7O_5$ $[M+H]^+$: 554.3099, found: 554.3091.

To a solution of **23** (115 mg, 0.11 mmol) was added MeOH (2.2 mL) containing 10% Pd/C (23 mg). $H_{2(g)}$ was bubbled through the solution and the resulting mixture was stirred at room temperature for 2 hours. The mixture was then filtered and the residue was washed with MeOH twice. The filtrate was evaporated under vacuum to afford compound **33** as a semi-solid (80 mg, 0.08 mmol). TLC (EtOAc:Hexanes, 3:1 v/v): $R_f$ = 0.51; HRMS (ESI-TOF, *m/z*) calculated for $C_{41}H_{62}N_2O_{23}$ $[M-H]^-$ : 949.3665, found: 949.3653.

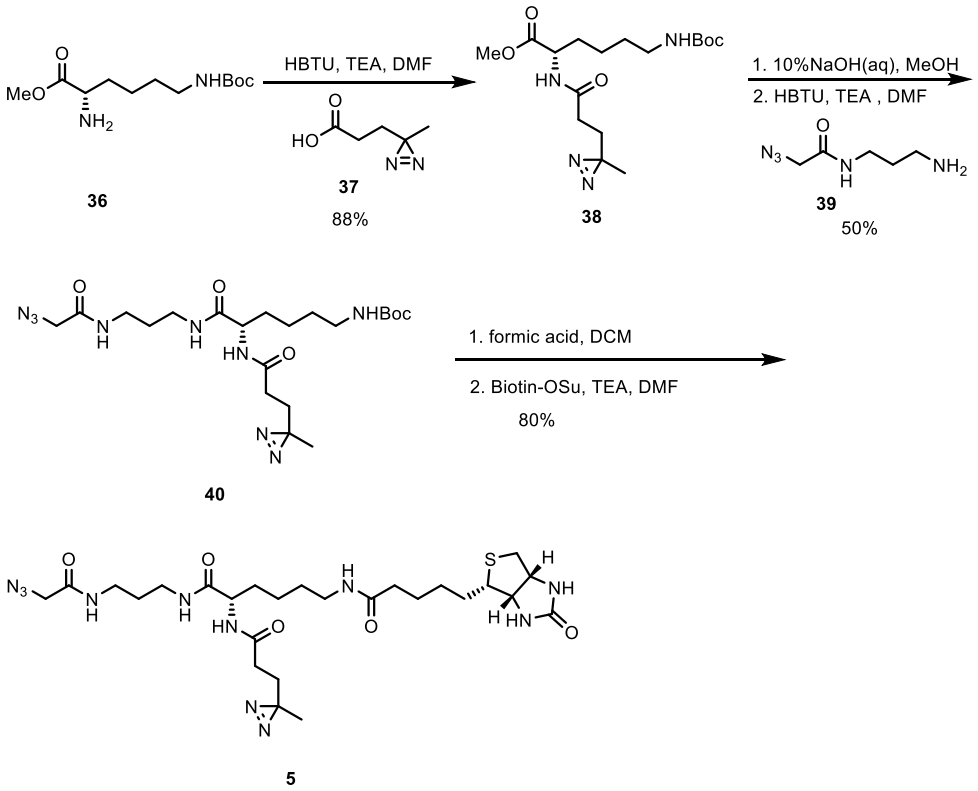
To a solution of the above amine intermediate (16.0 mg, 0.04 mmol), lactoside **33** (45.0 mg, 0.05 mmol), and HBTU (22.0 mg, 0.06 mmol) in dry DMF (0.3 mL) was added triethylamine (0.1 mL, 68.22 mmol) at room temperature under a nitrogen atmosphere. After being stirred at room temperature for overnight, the mixture was concentrated to dryness *in vacuo* and ethyl acetate was added. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and then concentrated under reduced pressure to give the crude product. The residue was purified by silica gel column chromatography to afford compound **34** (23 mg, 0.02 mmol, 41% yield for two steps) as a white syrup. TLC (EtOAc:Hexanes:MeOH, 3:1:0.4 v/v/v): $R_f$ = 0.24; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.78 (d, *J* = 15.8 Hz, 1H), 7.41 (d, *J* = 7.8 Hz, 1H), 6.77-6.75 (m, 2H), 6.70(d, *J* = 15.8 Hz, 1H), 5.35 (d, *J* = 3.4 Hz, 1H), 5.17 (dt, *J* = 4.3, 9.3 Hz, 1H), 5.11 (dd, *J* = 3.5, 10.4, 1H), 5.01 (dd, *J* = 7.8, 10.4, 1H), 4.79 (dd, *J* = 2.4, 9.6, 1H), 4.69 (dd, *J* = 1.9, 7.8, 1H), 4.60

(dd, $J$ = 5.9, 8.0, 1H), 4.51 (ddd, $J$ = 2.0, 5.9, 8.0, 1H), 4.33-4.27 (m, 3H), 4.23 (dd, $J$ = 5.1, 9.0, 1H), 4.16-4.11 (m, 4H), 3.86-3.78 (m, 2H), 3.74-3.69 (m, 1H), 3.29-3.28 (m, 4H), 3.06-3.01 (m, 2H), 2.49-2.48 (m, 4H), 2.33-2.29 (m, 3H), 2.13 (s, 3H), 2.10 (s, 3H), 2.08-2.02 (m, 14H), 1.93 (s, 3H), 1.88-1.78 (m, 4H), 1.69-1.63 (m, 4H), 1.60-1.57 (m, 2H), 1.51-1.39 (m, 17H), 1.00 (s, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 175.8, 174.9, 174.8, 174.6, 174.3, 172.5, 172.1, 172.0, 171.8, 171.5 (2C), 171.2, 169.6, 158.1, 157.8, 143.0, 137.5, 130.1, 122.1, 121.4, 120.0, 115.6, 102.0, 101.6, 83.7, 77.6, 74.5, 73.9, 73.2, 72.5, 71.7, 70.7, 70.5, 70.2, 68.6, 63.6, 62.3, 55.1, 43.6, 41.1,40.1, 40.0 (2C), 35.8, 33.2, 33.0, 32.7, 32.4, 31.4 (2C), 31.1, 30.7, 30.6, 30.1, 28.8 (3C), 26.9, 26.4, 24.3, 24.2, 21.1, 20.8 (2C), 20.7, 20.6, 20.5, 19.7, 15.6; HRMS (ESI-TOF, *m/z*) calculated for $C_{69}H_{98}N_9O_{27}$ $[M-H]^-$: 652.3435, found: 652.3459.

Compound **LLP 1**

To a stirred solution of compound **34** (13 mg, 0.01 mmol) in DCM (0.3 mL) was added formic acid (150 μL). After being stirred at room temperature for 3 hours, the mixture was neutralized with Dowex resin 550 ($OH^-$) at 4°C, and then filtered and concentrated *in vacuo* to give a crude product, which was used in the next reaction without purification; $R_f$ = 0.18 (DCM/MeOH = 1/5). The above material (12.0 mg, 0.01 mmol) and NHS-activated biotin (biotin-Osu) (14.8 mg, 0.04 mmol) were dissolved in dry DMF (300 μL) followed by addition of triethylamine (10 μL, 0.07 mmol). The reaction was stirred at room temperature under a nitrogen atmosphere for 4 h. After the reaction was complete, the mixture was concentrated to dryness *in vacuo* and the resulting residue was purified by silica gel column chromatography to afford compound **35** (16 mg, 0.01 mmol) as a white solid. TLC (MeOH:DCM, 1:5 v/v): $R_f$ = 0.74; HRMS (ESI-TOF, *m/z*) calculated for $C_{74}H_{106}N_{11}O_{27}S$ $[M+H]^+$: 1612.6980, found: 1612.6974. The above residue was dissolved in MeOH (300 μL) and stirred with NaOMe (0.5 mg, 0.02 mmol) at room temperature for 3 h. The reaction mixture was neutralized with

Amberlite IR-120 ($H^+$) resin, filtered, and concentrated *in vacuo* to give the crude product. The residue was purified by C18 reverse phase column to afford compound **LLP 1** (6.5 mg, 0.005 mmol, 56% yield for three steps) as a semi-solid. TLC (DCM:MeOH, 1:1 v/v): $R_f$ = 0.27; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.78 (d, *J* = 15.9 Hz, 1H), 7.41 (d, *J* = 7.9 Hz, 1H), 6.77-6.74 (m, 2H), 6.69 (d, *J* = 15.8 Hz, 1H), 4.49 (dd, *J* = 5.0, 7.5, 1H), 4.40-4.27 (m, 6H), 4.25-4.21 (m, 2H), 3.90-3.76 (m, 6H), 3.71 (dd, *J* = 4.6, 11.4, 1H), 3.61-3.58 (m, 2H), 3.55-3.48 (m, 2H), 3.24 (t, *J* = 8.12, 2H), 3.20-3.17 (m, 3H), 2.92 (dd, *J* = 5.0, 13.0 Hz, 1H), 2.70 (d, *J* = 12.7 Hz, 1H), 2.48 (m, 4H), 2.45-2.34 (m, 2H), 2.30 (m, 1H), 2.20 (t, *J* = 7.2 Hz, 2H), 2.08 (t, *J* = 7.4 Hz, 2H), 1.94-1.89 (m, 2H), 1.85-1.77 (m, 2H), 1.74-1.52 (m, 12H), 1.46-1.37 (m, 6H), 1.00 (s, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 176.2, 176.1, 174.9, 174.8, 174.6, 174.3, 169.6, 166.1, 158.1, 143.1, 137.5, 130.0, 122.1, 121.4, 119.7, 115.6, 105.1, 104.2, 83.7, 80.7, 77.1, 76.4 (2C), 74.8, 72.6, 70.5, 70.3, 69.9, 69.7, 63.4, 62.5, 61.9, 61.6, 57.0, 55.1, 54.8, 43.6, 41.0, 40.1, 40.0 (2C), 36.8, 35.8, 33.4, 33.1, 32.6, 32.5, 31.4, 30.7, 30.1, 29.7, 29.5, 27.0, 26.9, 26.8, 26.4, 24.3, 24.2, 19.7, 15.6; HRMS (ESI-TOF, *m/z*) calculated for $C_{60}H_{92}N_{11}O_{20}S$ $[M+H]^+$: 1318.6241, found: 1318.6235.




**Scheme S5.** Synthesis of compound **5**.

Compound **38**

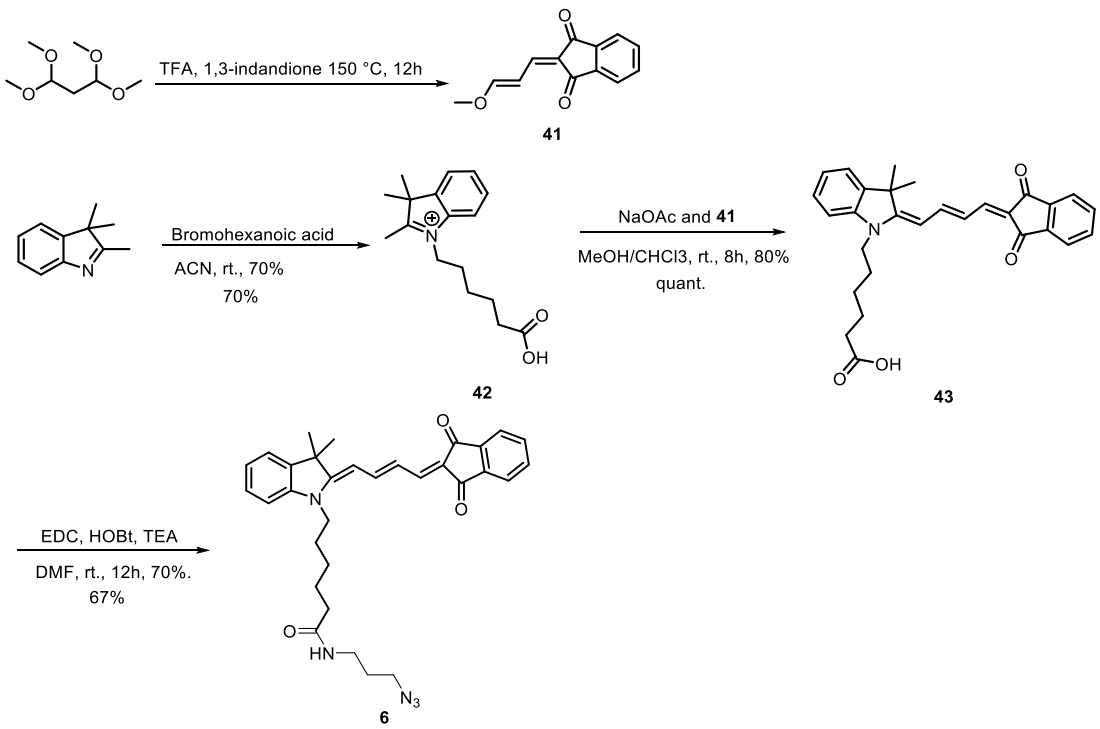
To a solution of compound **36** (41.0 mg, 0.16 mmol), compound **37** (10.0 mg, 0.08 mmol), HBTU (59.2 mg, 0.16 mmol), and triethylamine (30.0 μL, 0.24 mmol) in dry DMF (800.0 μL) was stirred at room temperature under a nitrogen atmosphere for 2 h. The mixture was concentrated to dryness *in vacuo* and ethyl acetate was added. The organic layer was washed with 1 N $HCl_{(aq)}$, brine, dried over $MgSO_4$, filtered, and then concentrated under reduced pressure to give the crude product. The residue was purified by silica gel column chromatography to afford compound **38** (27.6 mg, 0.07 mmol, 88% yield) as a white syrup. TLC (EtOAc:Hexanes, 1:1 v/v): $R_f = 0.27$; $^1$H NMR (400 MHz, $CDCl_3$) δ 6.16 (br s, 1H), 4.59-4.53 (m, 2H), 3.71 (s, 3H), 3.07 (q, $J$ = 9.8, 6.6 Hz, 2H), 2.06-2.00 (m, 2H), 1.82-1.66 (m, 6H), 1.47-1.39 (m, 11H), 1.00 (s, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 174.74, 174.66, 174.2, 53.8, 52.6, 41.0, 32.2, 31.4, 31.0,

30.9, 30.5, 28.8 (3C), 26.3, 24.1, 19.7. HRMS (ESI-TOF, *m/z*) calculated for $C_{17}H_{31}N_4O_5$ $[M+H]^+$: 371.2294, found: 371.2293.

Compound **5**

To a stirred solution of compound **38** (15.0 mg, 0.04 mmol) in MeOH (0.4 mL) was added 10% $NaOH_{(aq)}$ (0.8 mL). After being stirred at room temperature for 2 hours, the reaction mixture was neutralized by 1N $HCl_{(aq)}$ to pH = 2 at 0 ˚C. The MeOH was removed under reduced pressure and the resulting residue was extracted by EtOAc (20 mL × 3). The collected organic solvent was washed with brine, dried over $MgSO_4$, and filtered. The filtrate was concentrated *in vacuo* to give carboxylic acid product (14.4 mg, 0.04 mmol) as a white syrup; TLC (MeOH:DCM, 1:3 v/v): $R_f$ = 0.81. To a solution of the above compound (14.4 mg, 0.04 mmol), amine **39** (9.4 mg, 0.06 mmol), and HBTU (41.0 mg, 0.08 mmol) in dry DMF (1.0 mL) was added triethylamine (21.0 μL, 0.12 mmol) at room temperature under a nitrogen atmosphere. After being stirred at room temperature for 3 hours, the mixture was concentrated to dryness *in vacuo* and ethyl acetate was added. The organic layer was washed with 1N $HCl_{(aq)}$ and brine, dried over $MgSO_4$, filtered, and then concentrated under reduced pressure to give the crude product. The residue was purified by silica gel column chromatography to afford compound **40** (23.3 mg, 0.02 mmol, 50% yield) as a white syrup; TLC (EtOAc:Hexanes:MeOH, 1:1:0.2 v/v/v): $R_f$ = 0.30. To a stirred solution of compound **40** (21.0 mg, 0.02 mmol) in DCM (0.3 mL) was added formic acid (0.9 mL). After being stirred for 1 hours at room temperature, the mixture was concentrated to dryness *in vacuo* to give a crude product, which was used in the next reaction without purification. To a solution of above material (5.5 mg, 0.01 mmol) and biotin-OSu (4.0 mg, 0.01 mmol) in dry DMF (0.2 mL) was added triethylamine (40.0 μL, 0.3 mmol). The reaction was stirred at room temperature for 2 h under a nitrogen atmosphere. After the reaction was complete, the mixture was concentrated to dryness *in vacuo* and the

resulting residue was purified by silica gel column chromatography to afford compound **5** (5.0 mg, 0.008 mmol, 80% yeild) as a white solid. TLC (MeOH:DCM, 1:7 v/v): $R_f$ = 0.42; $^1$H NMR (400 MHz, $CD_3OD$) δ 4.50 (dd, *J* = 7.9, 4.8 Hz,1H), 4.31 (dd, *J* = 7.8, 4.4 Hz,1H), 4.22 (dd, *J* = 8.7, 5.5 Hz,1H), 3.9 (s, 2H), 3.25-3.26 (m, 7H), 2.93 (dd, *J* = 12.8, 5.0 Hz, 1H), 2.71 (d, *J* = 12.7 Hz, 1H), 2.22-2.13 (m, 4H), 1.83-1.61 (m, 10H), 1.55-1.41 (m, 6H), 1.01 (s, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 176.0, 174.7, 174.6, 170.3, 63.4, 61.6, 57.0, 55.1, 53.0, 41.0, 40.0, 37.7, 37.5, 36.8, 32.6, 31.2, 31.0, 30.8, 30.12, 30.06, 29.7, 29.5, 26.9, 26.4, 24.4, 19.8; HRMS (ESI-TOF, *m/z*) calculated for $C_{26}H_{42}N_{11}O_5S$ [M-H]$^-$: 620.3091, found: 620.3105.



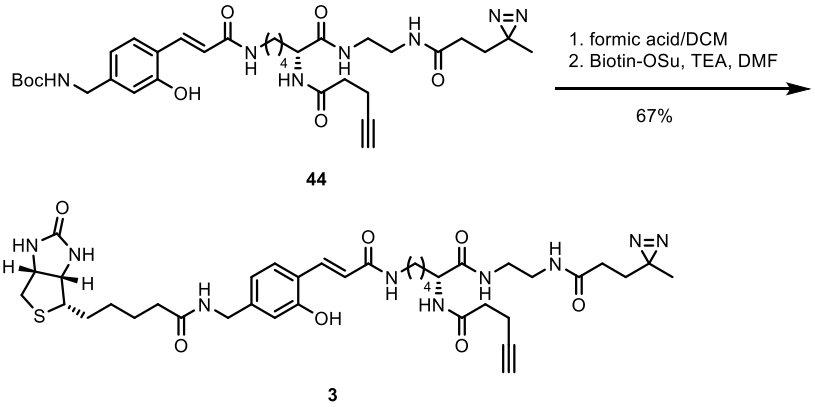

**Scheme S6**. Synthesis of compound **6**.

The syntheses of compounds **41**, **42** and **43** were followed by reported procedures.[2]

Compound **6**

To a reaction flask containing compound **43** (15.0 mg, 0.03 mmol), HOBt (6.0 mg, 0.04

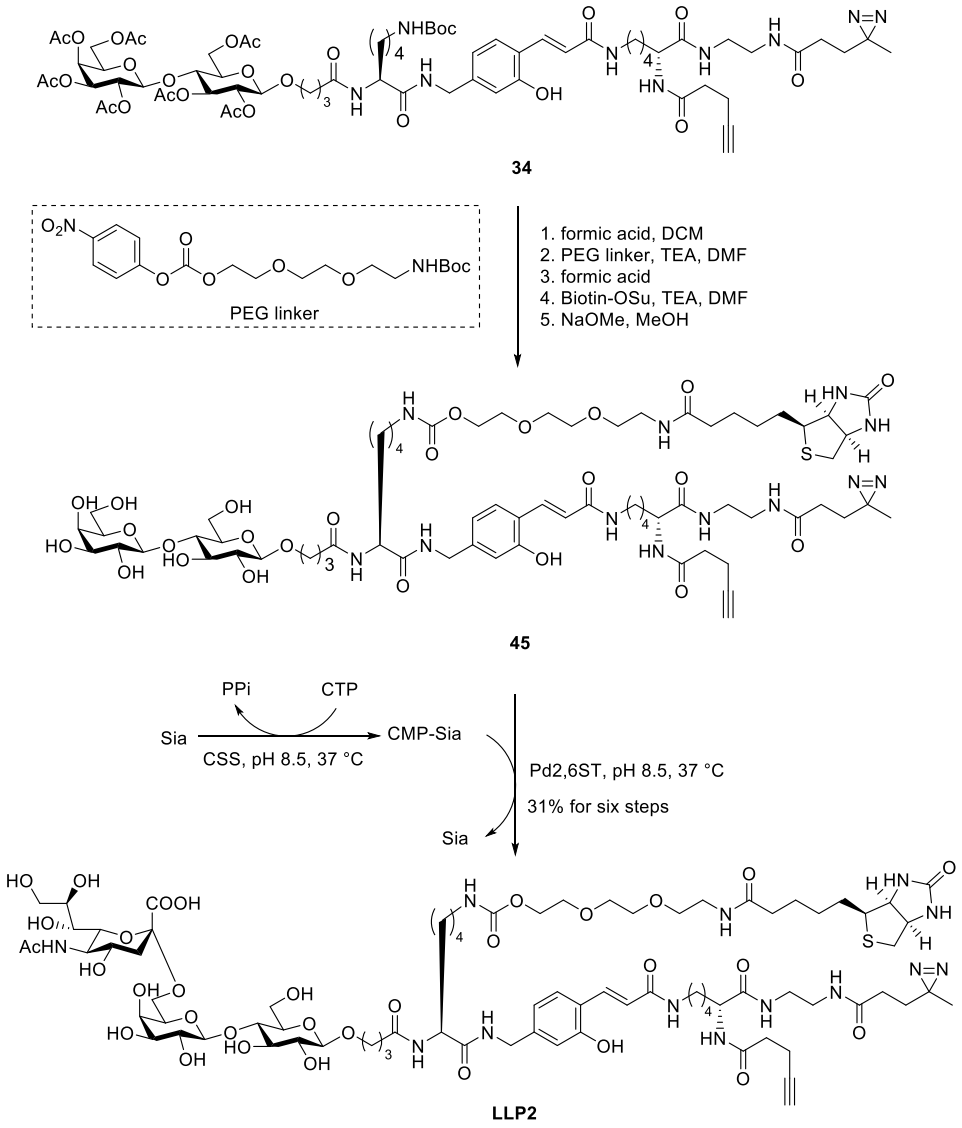
mmol), and *N*-(3-dimethylaminopropyl)-*N*'-ethylcarbodiimide hydrochloride (8.0 mg, 0.04 mmol) was added DMF (2 mL) at room temperature. After 10 minutes, triethylamine (20.0 μL) and 3-azido-1-propanamine (5.6 mg, 0.06 mmol) were added respectively and the resulting solution was stirred at room temperature overnight. The solvent was removed and the crude mixture was purified by flash column chromatography to give product **6** as a purple solid (12.0 mg, 0.02 mmol, 67%). TLC (EtOAc:Hexanes, 1:1 v/v): $R_f$ = 0.5. $^1$H NMR (500 MHz, DMSO-$d_6$): δ 8.92 (dd, *J* = 13.1 Hz, 1H), 8.62 (dd, *J* = 5.6 Hz, 1H), 8.55 (d, *J* = 13.1 Hz, 1H), 8.54-8.48 (m, 3H), 8.34 (d, *J* = 13.1 Hz, 1H), 8.31 (d, *J* = 7.2 Hz, 1H), 8.13 (dd, *J* = 7.5 Hz, 1H), 8.02 (d, *J* = 7.9 Hz, 1H), 7.94 (dd, *J* = 7.5 Hz, 1H), 7.01 (d, *J* = 13.1 Hz, 1H), 4.80 (dd, *J* = 7.1 Hz, 2H), 4.11 (t, *J*= 6.8 Hz, 2H), 3.87 (q, *J* = 6.7 Hz, 1H), 2.86 (t, *J* = 7.3 Hz, 2H), 2.48-2.34 (m, 12H), 2.19-2.12 (m, 2H); $^{13}$C NMR (125 MHz, DMSO-$d_6$): δ 190.4, 190.2, 171.9, 169.9, 155.1, 146.4, 142.5, 141.1, 140.5, 139.8, 133.7, 133.5, 128.2, 123.3, 122.2, 121.2, 120.9, 119.4, 116.8, 109.9, 100.5, 48.4, 48.0, 42.6, 35.7, 35.1, 28.4, 27.4, 26.5, 25.7, 25.0; HRMS (ESI, *m/z*) calculated for $C_{32}H_{36}N_5O_3$ $[M+H]^+$: 538.2818, found: 538.2815.




**Scheme S7**. Synthesis of compound **3**.

Compound **3**

To a stirred solution of compound **44** (22.0 mg, 0.03 mmol) in DCM (0.7 mL) was added formic acid (1.5 mL). After being stirred at room temperature for 2 hours, the residue was concentrated *in vacuo* to give amine product (18.6 mg, 0.03 mmol). To a solution of above amine (18.6 mg, 0.03 mmol) in dry DMF (0.2 mL) was added biotin-OSu (34.2 mg, 0.09 mmol) and triethylamine (21.0 μL, 0.15 mmol) at room temperature. After being stirred for 3.5 hours, the solvent was removed under reduced pressure and the resulting residue was purified by silica gel column chromatography to afford compound **3** (16.1 mg, 0.02 mmol, 67%) as a white solid. TLC (MeOH:DCM, 1:3): $R_f$ = 0.46; $^1$H NMR (400 MHz, $CD_3OD$) δ 7.78 (d, *J* = 15.9 Hz, 1H), 7.41 (d, *J* = 8.4 Hz, 1H), 6.78-6.76 (m, *2*H), 6.69 (d, *J* = 15.9 Hz, 1H), 4.47 (dd, *J* = 7.9, 4.8 Hz, 2H), 4.29 (brs, 2H), 4.27-4.21 (m, 4H), 3.28 (brs, 4H), 3.16 (ddd, *J* = 8.8, 5.7, 4.4 Hz, 1H), 2.91 (dd, *J* = 12.8, 5.0 Hz, 1H), 2.70 (d, *J* = 12.8 Hz, 1H), 2.48 (brs, 4H), 2.29 (s, 1H), 2.26 (t, *J* = 7.4 Hz, 2H), 2.08 (d, *J* = 7.6 Hz, 1H), 2.06 (d, *J* = 6.4 Hz, 1H), 1.84-1.55 (m, 12H), 1.46-1.39 (m, 4H), 1.00 (s, 3H); $^{13}$C NMR (100 MHz, $CD_3OD$) δ 175.9, 174.9, 174.8, 174.3, 169.6, 166.1, 158.1, 143.3, 137.5, 130.1, 122.1, 121.4, 119.8, 116.0, 83.7, 70.4, 63.3, 61.6, 57.0, 55.1, 43.8, 41.0, 40.1, 40.04, 39.92, 36.7, 35.8, 32.5, 31.4, 31.3, 30.2, 29.7, 29.4, 26.9, 26.4, 24.2, 19.7, 15.6; HRMS (ESI, *m/z*) calculated for $C_{38}H_{53}N_9NaO_7S$ $[M+Na]^+$: 802.3686, found: 802.3687.




**Scheme S8.** Synthesis of compound **LLP 2**

Compound **LLP 2**

To a stirred solution of compound **34** (13 mg, 0.01 mmol) in DCM (0.3 mL) was added formic acid (150 μL). After being stirred at room temperature for 2 hours, the mixture was neutralized with Dowex resin 550 ($OH^-$) at 4°C, and then filtered and concentrated *in vacuo* to give a crude product, which was used in the next reaction without purification; $R_f$ = 0.18 (DCM/MeOH = 1/5). The above material (12.0 mg, 0.01 mmol) and PEG linker (10.8 mg, 0.03 mmol) were dissolved in dry DMF (300 μL) followed by addition of triethylamine (10 μL, 0.07 mmol). The reaction was stirred at room

temperature under a nitrogen atmosphere for 1 h. After the reaction was complete, the mixture was concentrated to dryness *in vacuo* and the resulting residue was purified by silica gel column chromatography as a semi-solid. TLC (EtOAc:Hexanes:MeOH, 4:2:1.2 v/v/v): $R_f$ = 0.33; HRMS (ESI-TOF, *m/z*) calculated for $C_{76}H_{112}N_{10}NaO_{31}$ $[M+Na]^+$: 1683.7392, found: 1683.7428. The above material (12.0 mg, 0.008 mmol) in DCM (0.3 mL) was added formic acid (150 μL). After being stirred at room temperature for 2 hours, the mixture was neutralized with Dowex resin 550 ($OH^-$) at 4°C, and then filtered and concentrated *in vacuo* to give a crude product, which was used in the next reaction without purification; The above material (10.8 mg, 0.008 mmol) and biotin-OSu (7.4 mg, 0.02 mmol) were dissolved in dry DMF (300 μL) followed by addition of triethylamine (10 μL, 0.07 mmol). The reaction was stirred at room temperature under a nitrogen atmosphere for 2 h. After the reaction was complete, the mixture was concentrated to dryness *in vacuo* and the resulting residue was purified by silica gel column chromatography as a semi-solid. TLC (MeOH:DCM, 1:5 v/v): $R_f$ = 0.55; HRMS (ESI-TOF, *m/z*) calculated for $C_{81}H_{118}N_{12}NaO_{31}S$ $[M+Na]^+$:1809.7644, found: 1809.7595. The above residue (12.6 mg, 0.008 mmol) was dissolved in MeOH (300 μL) and stirred with NaOMe (0.5 mg, 0.02 mmol) at room temperature for 3 h. The reaction mixture was neutralized with Amberlite IR-120 ($H^+$) resin, filtered, and concentrated *in vacuo* to give the crude product. The residue was purified by C18 reverse phase column to afford compound **45** (6.5 mg, 0.004 mmol) as a semi-solid. TLC (*n*-Propanol:$H_2O$:AcOH, 6:2:1 v/v): $R_f$ = 0.83; HRMS (ESI-TOF, *m/z*) calculated for $C_{67}H_{105}N_{12}O_{24}S$ $[M+H]^+$: 1493.6775, found: 1493.70853.

The reaction was carried out in a 15 mL centrifuge tube with 54 μL of Tris·HCl buffer (100 mM, pH 8.5) containing 50 mM of sialic acid (7 mg, 23 μmol), 55 mM of CTP (11 mg, 21 μmol), and 20 mM of $MgCl_2$. The pH of reaction mixture was adjusted to 8.5 by adding 2N NaOH. Then, 0.5 mg/mL of CSS was added to the above solution.

The resulting mixture was incubated at 37 °C with agitation at 600 rpm for 3 h and the formation of CMP-Sia was monitored by TLC analysis (*n*-PrOH/ $H_2O$/ AcOH = 6: 2: 1 (v/ v/ v), $R_f$ = 0.15). After the completion of the reaction as indicated by the disappear of sialic acid on TLC, 2.5 mM of compound **45** (3 mg, 1.8 $\mu$mol) was added and the pH of the solution was adjusted to pH 8.5 by addition of 2N NaOH. Then, 0.3 mg/mL of Pd2,6ST was added and the solution was incubated at 37 °C. More enzymes were added if necessary. After be shaked for 2 h, the reaction was quenched by adding the same reaction volume of EtOH. The reaction solution was centrifuged (10,000 x *g*, 10 min), filtered (0.45-μm PVDF filter; Millipore), and then concentrated. The resulting residue was purified by a C18 reverse-phase silica column to afford compound **LLP 2** (3.2 mg, 1.6 $\mu$ mol, 31% yield for six steps) as a semi-solid. TLC (*n*-Propanol:$H_2O$:AcOH, 6:2:1 v/v/v): $R_f$ = 0.62; $^1$H NMR (400 MHz, $D_2O$) δ 7.73 (dd, *J* = 16.0, 2.6 Hz, 1H), 7.56 (d, *J* = 7.9 Hz, 1H), 6.89-6.86 (m, *2*H), 6.73 (dd, *J* = 16.0, 5.2 Hz, 1H), 4.65-4.55 (m, 2H), 4.42-4.22 (m, 8H), 3.94-3.61 (m, 27H), 3.37-3.29 (m, 8H), 3.10 (t, *J* =6.0 Hz, 2H), 3.00 (dd, *J* = 13.0, 5.0 Hz, 1H), 2.95 (dd, *J* = 13.0, 5.0 Hz, 1H), 2.75 (d, *J* = 13.0 Hz, 2H), 2.50-2.48 (m, 4H), 2.35 (t, *J* = 7.5 Hz, 2H), 2.28-2.18 (m, 4H), 2.12-2.04 (m, 7H), 1.80-1.38 (m, 22H), 1.00 (s, 3H); $^{13}$C NMR (175 MHz, $D_2O$) δ 176.6, 175.2, 175.0, 174.7, 174.6, 174.2, 174.0, 173.3, 173.2, 168.7, 165.1, 158.2, 155.5, 141.6, 135.8, 129.1, 120.6, 120.2, 119.1, 114.7, 103.4, 101.8, 100.4, 99.7, 83.2, 82.7, 79.5, 74.9, 74.4, 73.6, 72.5, 72.4, 72.2, 71.64, 71.56, 70.8, 70.0, 69.6, 69.3, 68.7, 68.4, 68.2, 68.1, 66.0, 64.8, 63.9, 62.5, 62.3, 61.9, 55.2, 53.8, 51.6, 42.3, 40.0, 39.5, 38.8, 38.4, 35.3, 34.0, 31.6, 30.5, 30.0, 29.6, 28.2, 27.8, 27.7, 27.5, 26.0, 25.0, 22.1, 21.91, 21.85, 20.7, 20.0, 18.4, 14.3; HRMS (ESI-TOF, *m/z*) calculated for $C_{78}H_{121}N_{13}NaO_{32}S$ $[M+Na]^+$: 1806.7858, found: 1806.7901.

**Photo-labeling of $RCA_{120}$ by LLP 1**

To evaluate the imprinting efficiency, $RCA_{120}$ (6 μM, 60 μL in PBS buffer, pH 7.4) was incubated with various concentrations of compound **LLP 1** (50, 150, 500, 1000, and 1500 μM, respectively) The mixture was briefly agitated at 4 °C for 60 min to ensure proper mixing and then irradiated with a UV lamp (365 nm, 18.7 $mW/cm^2$ at 4 cm) (Blak-Ray® B-100AP High Intensity UV lamp) at 4 °C for 30 min. Excess photoaffinity probe was removed from the reaction via spin concentration (Microcon centrifugal filter 10,000 MWCO, Millipore, MA). An aliquot of this partially purified mixture was treated with SDS-PAGE loading buffer (0.3 M Tris-HCl, 10% SDS, 30% glycerol, 9.3% DTT, pH 6.8) containing 100 mM DTT for 10 min under room temperature followed by electrophoresis. Samples were visualized by Instantblue staining and photolabeling was verified by Western blot analysis using an HRP-conjugated anti-biotin antibody. Negative control experiments were performed by using compound **3**. A saturated biotin signal was observed when **LLP 1** was used at a concentration of 1000 μM (Fig. S1, lane 4). The labeling specificity of **LLP 1** was further confirmed using control probe **3** (lacking the carbohydrate ligand, Fig. 1b) under the same imprinting conditions (lane 8), resulting in no detectable biotin signal.

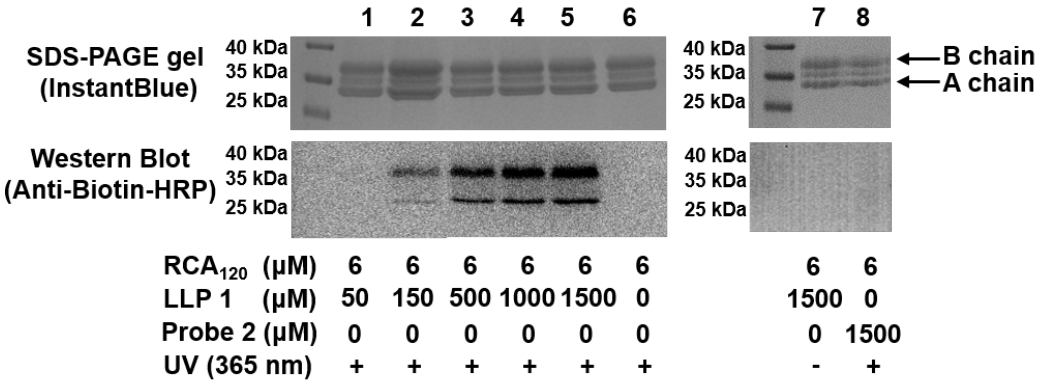


**Fig. S1**. $RCA_{120}$ was modified by **LLP 1**. (a) Photolabeling of $RCA_{120}$ with various concentrations of **LLP 1** (lanes 1-5). Total protein staining by InstantBlue (*top*) and Western blot analysis using an anti-biotin antibody (*bottom*), showing that both chains of $RCA_{120}$ were labeled. Lane 6: **LLP 1**-$RCA_{120}$ crosslinked adducts formed by a PAL reaction in the absence of **LLP 1**. Lane 7: $RCA_{120}$ without UV irradiation and in the presence of probe **LLP 1**. Lane 8: **3**-$RCA_{120}$ crosslinked adducts formed by a PAL reaction with probe **3**, which lacked lactose, as a negative control.

**Purification of LLP 1-$RCA_{120}$**

As shown in Fig. S2, an aliquot of above partially purified mixture (200 μL) was incubated with monomeric avidin-MNP (BcMag™ Monomeric Avidin Magnetic beads, MAMB) at room temperature for 60 min with vortexing. The $RCA_{120}$-MAMB complex was isolated by applying a magnet, and the resulting MNPs were washed three times with 50 μL washing buffer (PBS with 0.05% Tween 20, pH 7.4). The samples were then treated with SDS-PAGE loading buffer (0.3 M Tris-HCl, 10% SDS, 30% glycerol, 9.3% DTT, pH 6.8) containing with 100 mM DTT for 10 min under room temperature followed by electrophoresis. Samples were visualized by Instantblue staining and verified by Western blotting.

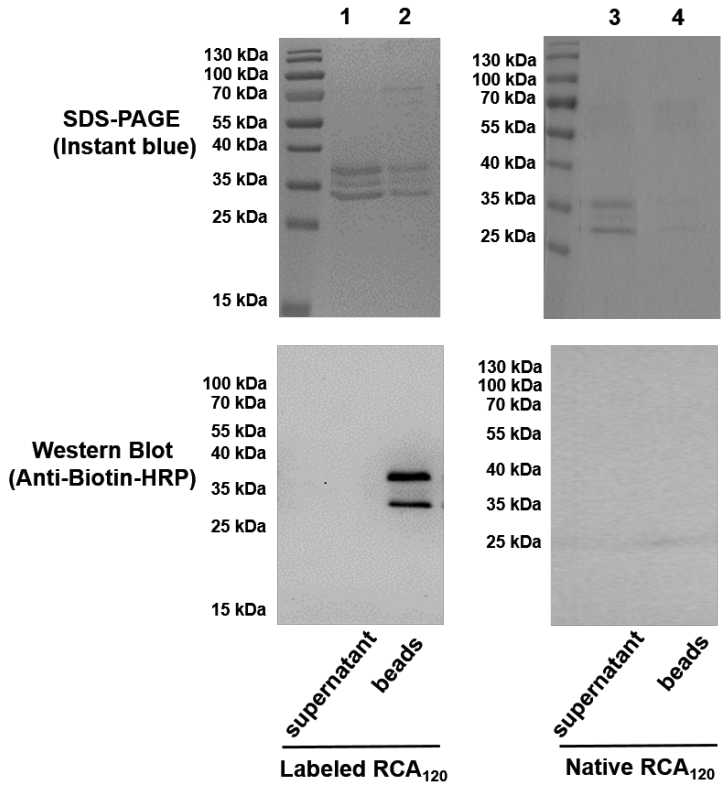



**Fig. S2**. Purification of **LLP 1**-$RCA_{120}$ by monomeric avidin magnetic beads. SDS-PAGE: lane 1: supernatant and lane 2: beads.

## Preparation of alkynated $RCA_{120}$

The pH of $RCA_{120}$-MAMB complex solution was adjusted to pH = 3 by adding citric acid–$Na_2HPO_4$ buffer solution and the resulting solution was incubated for 1 hour at room temperature. Equivalent amounts of all samples were treated with SDS-PAGE loading buffer (0.3 M Tris-HCl, 10% SDS, 30% glycerol, 9.3% DTT, pH 6.8) containing 100 mM DTT for 10 min under room temperature followed by electrophoresis. Samples were visualized by Instantblue staining and verified by Western blotting.

**Determination of alkynylated $RCA_{120}$ labeled site**

**Sample preparation for mass spectrometry.**

*Protein and peptide Processing.* Protein extracts were subsequently reduced with 10 mM dithiothreitol (Sigma, D9779) for 30 min at 29°C and alkylated with 50 mM iodoacetamide (Sigma, I1149) for 45 min. Samples were diluted 2-fold with 50 mM TEABC prior to digestion with LysC (Wako, 125-02543) for 3hr at 29°C and 8-fold to trypsin (Promega, V51110) overnight, the enzyme-to-protein ratio for LysC was 1:100 and trypsin was 1:50. Digested peptides were acidified with 10% trifluoroacetic acid (TFA, Wako, 208-02741) to a final concentration of 0.5% (pH ~2 to 3) and desalted on the homemade stage-tip by SDB-XC Empore wafers (3M). Peptide samples were dried by vacuum centrifugation and saved for LC-MS/MS analysis at -80°C.

**Liquid chromatography-tandem mass spectrometry (LC-MS/MS)**

LC-MS/MS analysis was performed on a Q Exactive Plus mass spectrometer (Thermo Fisher Scientific, Bremen, Germany) equipped with a nanospray interface (Proxeon, Odense, Denmark). Peptides were separated on a nanoAcquity system (Waters, Milford, MA) which was connected to mass spectrometry The spin-vacuum dried peptide samples were re-dissolved in 0.1% formic acid (FA) and loaded onto a 25 cm commercial analytical column with 3 μm reverse phase C18 beads (75 μm inner diameter, Thermo Fisher Scientific), the column temperature was maintained at 35°C. The peptides were separated with a binary buffer system of 0.1% FA (buffer A) and ACN plus 0.1% FA (buffer B), at a flow rate of 3 μL/min with 120 min gradient. The instruments were operated in data-dependent acquisition mode where the top-15 peaks were performed fragmentation. The full MS scans were acquired with a resolution of 70,000, an AGC target of 3e6 and a mass range from 350 to 1700 *m/z*. MS/MS scans

were triggered with a resolution of 17,500, an AGC target of 2e5, an isolation window 2.0 m/z and a normalized collision energy of 27%. The dynamical exclusion was 30 sec.

**Raw MS data processing**

Raw mass spectrometry data were processed with Proteome Discoverer 2.1.1 platform (Thermo Scientific, Bremen, Germany) and searched using Mascot and SequestHT against Swiss-Prot Homo sapiens protein database, allowing for up to two missed cleavages. The variable modifications included oxidized methionine (M, +15.995 Da), acetylation (protein N-term, +42.011 Da), deamidation (N/Q, +0.984 Da) and probe ($C_{18}H_{31}N_4O_3$, +351.239616 Da). Carbamidomethylation (C, +57.0214 Da) was set as a fixed modification. Precursor mass tolerance was set to 10 ppm and 0.1 Da for fragment mass. The false-discovery rate (FDR) filtration performed at 0.01 was both done on peptide and protein level. For label-free quantification, the average values of each sample were calculated and normalized with total summary of intensity.

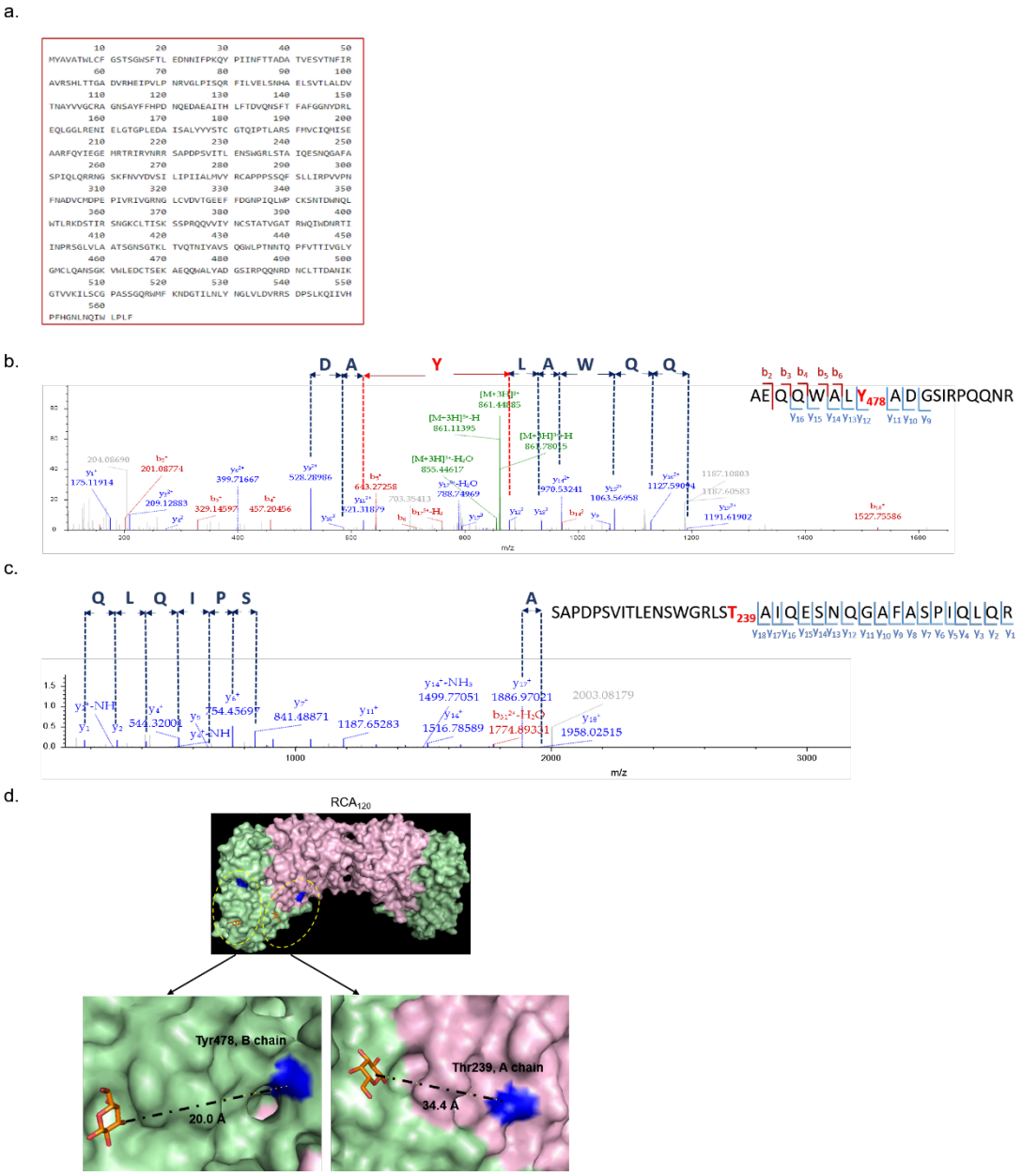



**Fig. S3.** Identification of labeled sites by LC-MS/MS analysis after Trypsin digestion of alkynylated $RCA_{120}$. (a) Primary sequence of $RCA_{120}$. (b) and (c) ESI-TRAP MS/MS analysis of alkynated-$RCA_{120}$. (d) The crystal structure of $RCA_{120}$ (PDB: 1RZO). The amino acids, Tyr478 of B chain and Thr239 of A chain, labeled by **1** are highlighted.

## Modification of alkynylated $RCA_{120}$ by CuAAC

Stock solutions: $CuSO_4$: 60 mM (in water), THPTA: 60 mM (in water), sodium ascorbate: 200 mM (fresh prepared), azido-molecule: 50 mM and buffer: PBS buffer

pH 7.4.

CuAAC procedure

Reagents were added into a 1.5 mL eppendorf tube by the following order:
10 μL of mixed 60 mM $CuSO_4$ (final concentration: 1.71 mM) and 50 μL of 60 mM THPTA (final concentration: 8.55 mM) were added to the alkynylated-$RCA_{120}$ solution (final concentration: 2.4 μM) followed by addition of 20 μL of 200 mM sodium ascorbate (final concentration: 11.43 mM) and 1.5 μL of 50 mM probe **5** (final concentration: 0.21 mM). After incubation for 12 hours, product was purified by PD Minitrap™ G-25 column following the protocol suggested by manufacturer.

**Photo-crosslink of 5-$RCA_{120}$ with OVA**

**5**-$RCA_{120}$ (final concentration 0.85 μM) was incubated with OVA (final concentration 85 μM) in PBS buffer (pH 7.4) at 4 °C for 60 min and then irradiated with a UV lamp (365 nm, 18.7 mW/$cm^2$ at 4 cm) (Blak-Ray® B-100AP High Intensity UV lamp) at 4 °C for 30 min.

**Environmental sensitive detection of interaction between 6-$RCA_{120}$ and OVA**

(1) Direct interaction. **6**-$RCA_{120}$ (final concentration 0.1 μM) was incubated with OVA (final concentration 12.5 μM) in PBS buffer (pH 7.4) at room temperature for 2 hours. The formation of **6**-$RCA_{120}$-OVA complex was investigated by measuring the fluorescent emission spectrum with excitation wavelength at 570 nm and emission wavelength from 590 to 750 nm. For non-specific binding, OVA was replaced with BSA (Fig. S4).

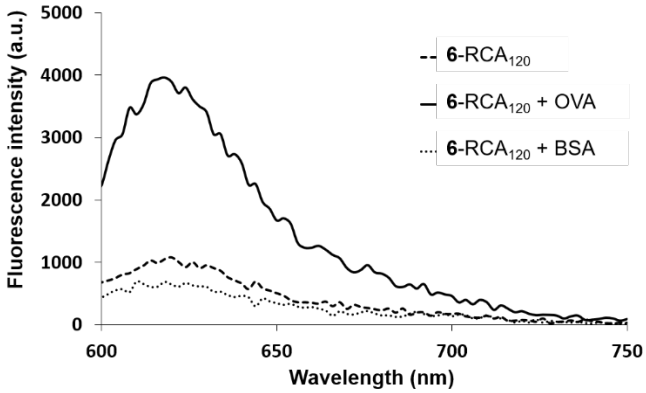



**Fig. S4**. Fluorescent emission spectra of **6**-$RCA_{120}$ and with OVA and BSA.

(2) Lactose competition. High concentration of lactose (125 mM) was added into the solution of **6**-$RCA_{120}$-OVA complex, obtained by above incubation. The mixture was incubated for 6 hours. The result was investigated by measuring the fluorescent emission spectrum with excitation wavelength at 570 nm and emission wavelength from 590 to 750 nm.

(3) Negative control experiment. 10 μL of protein deglycosylation mix II (NEB) enzymes was mixed with OVA (12.5 μM) in PBS buffer (pH 7.4) at room temperature for 2 hours. Then, the **6**-$RCA_{120}$ (0.1 μM) was added to the above protein solution and the resulting solution was incubated for 2 hours. The formation of **6**-$RCA_{120}$-OVA complex was investigated by measuring the fluorescent emission spectrum with excitation wavelength at 570 nm and emission wavelength from 590 to 750 nm.

**Random modification of $RCA_{120}$ by compound 7**

To a solution of compound **43** (40.0 mg, 0.09 mmol), *N*-hydroxysuccinimide (10.1 mg,

0.09 mmol) and *N*-(3-dimethylaminopropyl)-*N*'-ethylcarbodiimide hydrochloride (16.8 mg, 0.09 mmol) in dry DCM (1.8 mL) was stirred at room temperature under a nitrogen atmosphere for 10 hours. The solvent was removed under reduced pressure and the resulting residue **7** was diluted with DMSO to a 50 mM stock solution. HRMS (ESI-TOF, *m/z*) calculated for $C_{33}H_{33}N_2O_6$ $[M+H]^+$ 553.2339, found: 553.2338.

$RCA_{120}$ (final concentration 2 μM, 200 μL in PBS buffer, pH 7.4) was incubated with NHS-activated compound **7** (final concentration 200 μM). After incubation for 12 hours at room temperature, the protein was purified using a PD Minitrap™ G-25 column. The above randomly modified **7**-$RCA_{120}$ was incubated with OVA or BSA separately in PBS buffer (pH 7.4) at room temperature for 2 hours. The formation of protein complex was investigated by measuring the fluorescent emission spectrum with excitation wavelength at 570 nm and emission wavelength from 590 to 750 nm (Fig. S5).

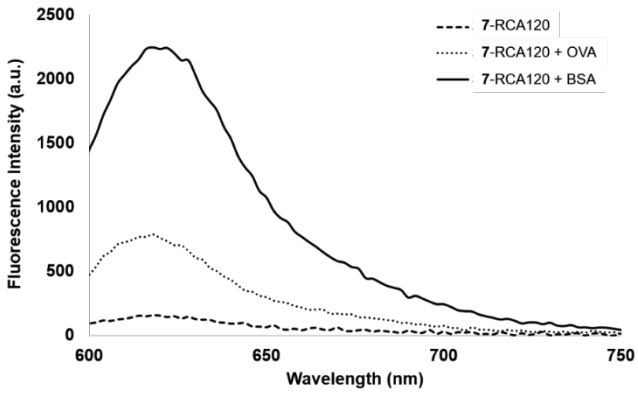
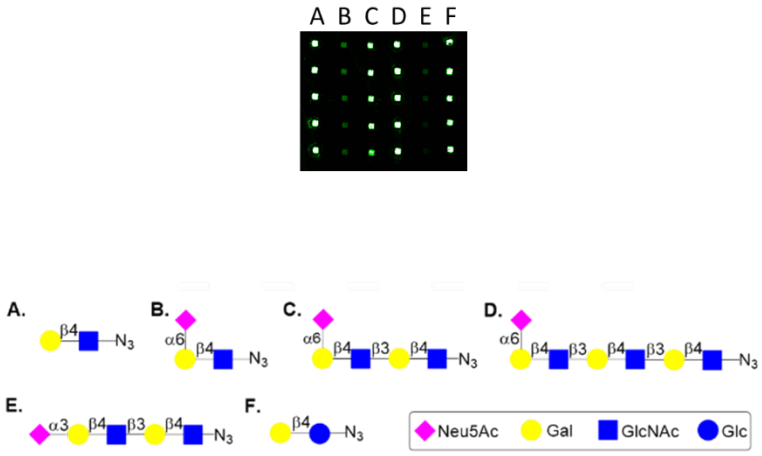



**Fig. S5**. Environment-sensitive fluorescent intensity between random modified **7**-$RCA_{120}$ with OVA and BSA, respectively.

(a)

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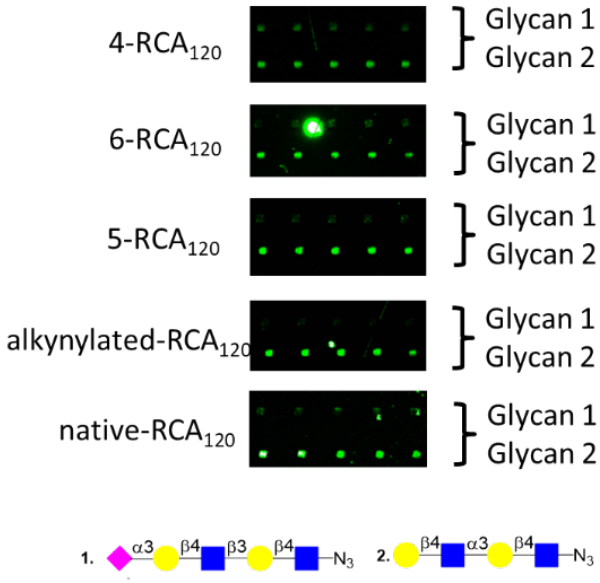



**Fig. S6**. Microarray fluorescence images for $RCA_{120}$ and modified $RCA_{120}$ probes binding assays. (a) glycan ligands binding affinities toward native $RCA_{120}$. (b) binding affinities of $RCA_{120}$ and modified $RCA_{120}$ toward di-LacNAc.

**Cell culture**

HeLa cells were cultured in DMEM medium supplemented with 10% fetal bovine serum and incubated at 37 °C under a humidified atmosphere of 95% air and 5% $CO_2$.

**No-wash fluorescence imaging of surface interaction molecules on HeLa cell by**

**6-RCA$_{120}$**

About 1 × $10^3$ cells were maintained in a culture medium supplemented with 10% FBS and seeded in 8-well chamber slides and cultured for 24 h at 37 °C in air with 5% $CO_2$. As shown in the Fig. S7, cells were treated with 40 nM **6**-RCA$_{120}$ (20 μL) and the resulting mixture was incubated for 90 min under 37 °C in air with 5% $CO_2$, followed by acquisition of live-cell images without any washing steps. For control experiments, cells were treated with 10 μL of Protein Deglycosylation Mix II (NEB) or 0.01 U of Neuraminidase from *Arthrobacter ureafaciens* [EC 3.2.1.18] (Nacalai Tesque) under cell culture conditions for 2 hours. Then, 40 nM **6**-RCA$_{120}$ (20 μL) was added and the resulting mixture were incubated for 90 min under the same conditions. Cell images were taken without removing the excess probe using a laser scanning confocal microscope (LSM 700, Zeiss, Germany). The images were taken using a 555 nm laser for excitation and emission from 590 to 700 nm.

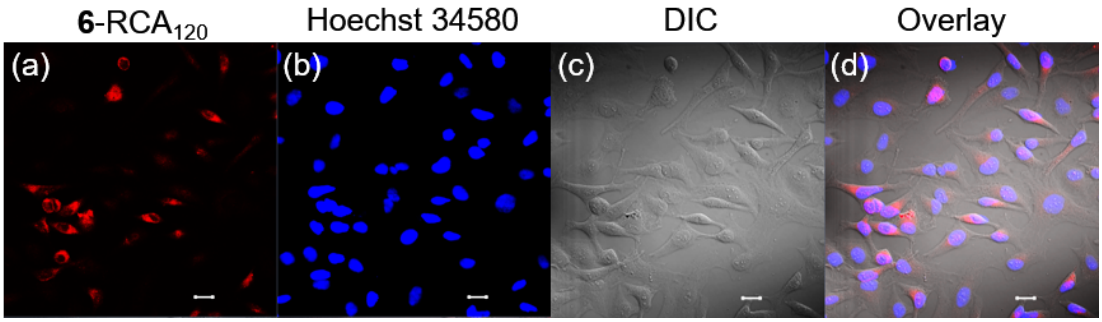



**Fig. S7**. Confocal fluorescence microscopy images showing the glycan-mediated HeLa cell imaging using an environment-sensitive **6**-RCA$_{120}$ probe. (a) The location of **6**-RCA$_{120}$ is indicated by red fluorescence. (b) Cell nuclei are stained with Hoechst 34580, indicated by blue fluorescence. (c) The HeLa cell morphology was showed by

differential interference contrast microscopy. (d) An overlay of the images shown in (a–c). Scale bar: 20 μm.

**Dynamic observation of 6-RCA$_{120}$ and glycoprotein interactions.**

About $1 \times 10^3$ cells were seeded in 8-well chamber slides and cultured at 37 °C in air with 5% $CO_2$ for 24 h. At the first part, cells were treated with 10 μL Protein Deglycosylation Mix II (NEB) and incubated under cell culture conditions for 1.5 hours. Then, the medium was replaced with the fresh medium containing 40 nM **6**-RCA$_{120}$ and 0.01 U 10 μL neuraminidase. Cell images were taken without removing the excess probe using a laser scanning confocal microscope for 40 min. Then the second part, the medium containing excess probe was replaced by the fresh medium and images were taken for an additional 2 hours. The videos (Movie S1 and S2) show the first part (before media replacement) and the second part (after media replacement) respectively. In addition, **4**-RCA$_{120}$ was used as the probe for control experiments to dynamically observe probe-glycoprotein interactions on the cell surface, as shown in Movie S3.

To confirm the location of the uptake of **6**-RCA$_{120}$ via lectin-glycoprotein interaction, $1.5 \times 10^3$ cells were seeded in 8-well chamber slides and cultured at 37 °C in air with 5% $CO_2$ for 15 hr. Then, cells were treated with 0.01 U of neuraminidase from *Arthrobacter ureafaciens* [EC 3.2.1.18] (Nacalai Tesque) and 1 μM LysoTracker™ Green DND-26 under cell culture conditions. After 2 hours incubation, cells were cooled down to 4 ˚C for 10 min then 40 nM of **6**-RCA$_{120}$ was added and the resulting mixture was incubated at 4 ˚C for 10 min, then at 25 ˚C for 30 min. Cell images were taken without removing the excess probe using a laser scanning confocal microscope (Fig. S8).

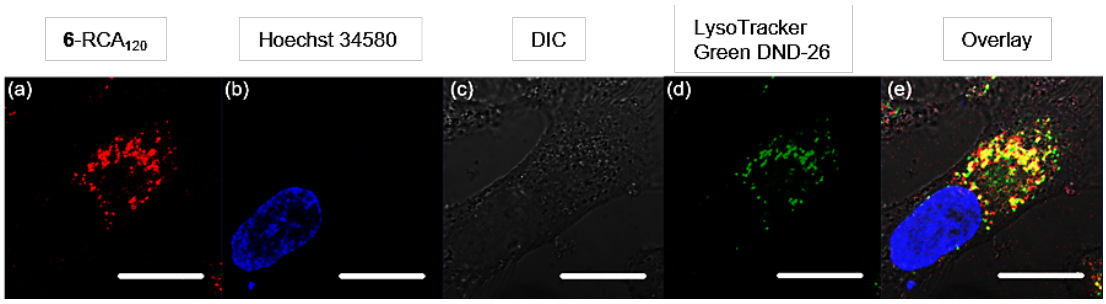



**Fig. S8**. Confocal fluorescence microscopy images showing the localization of **6**-$RCA_{120}$ within late endosomes and/or lysosomes of HeLa cells. (a) The location of **6**-$RCA_{120}$ is indicated by red fluorescence. (b) Cell nuclei are stained with Hoechst 34580, indicated by blue fluorescence. (c) HeLa cell morphology shown by differential interference contrast microscopy. (d) The green fluorescence is labeled by LysoTracker Green DND-26, indicative of the location of endosomes and/or lysosomes. (e) An overlay of the images shown in (a–d). The yellow fluorescence indicates that **6**-$RCA_{120}$ co-localized with LysoTracker Green DND-26 in late endosomes and/or lysosomes. Scale bar: 20 μm.

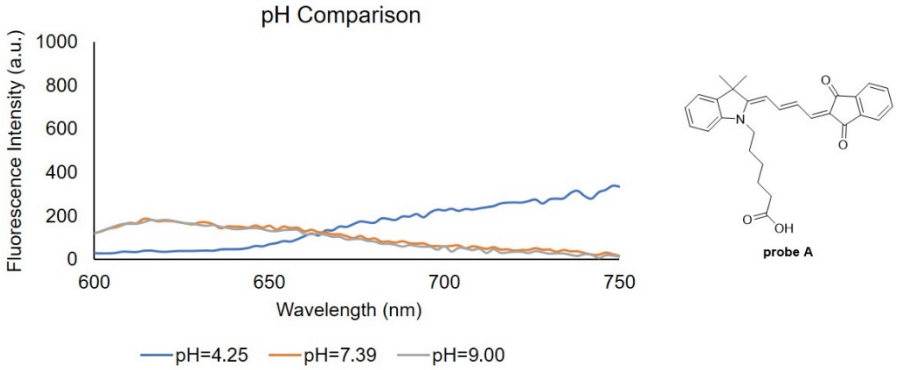
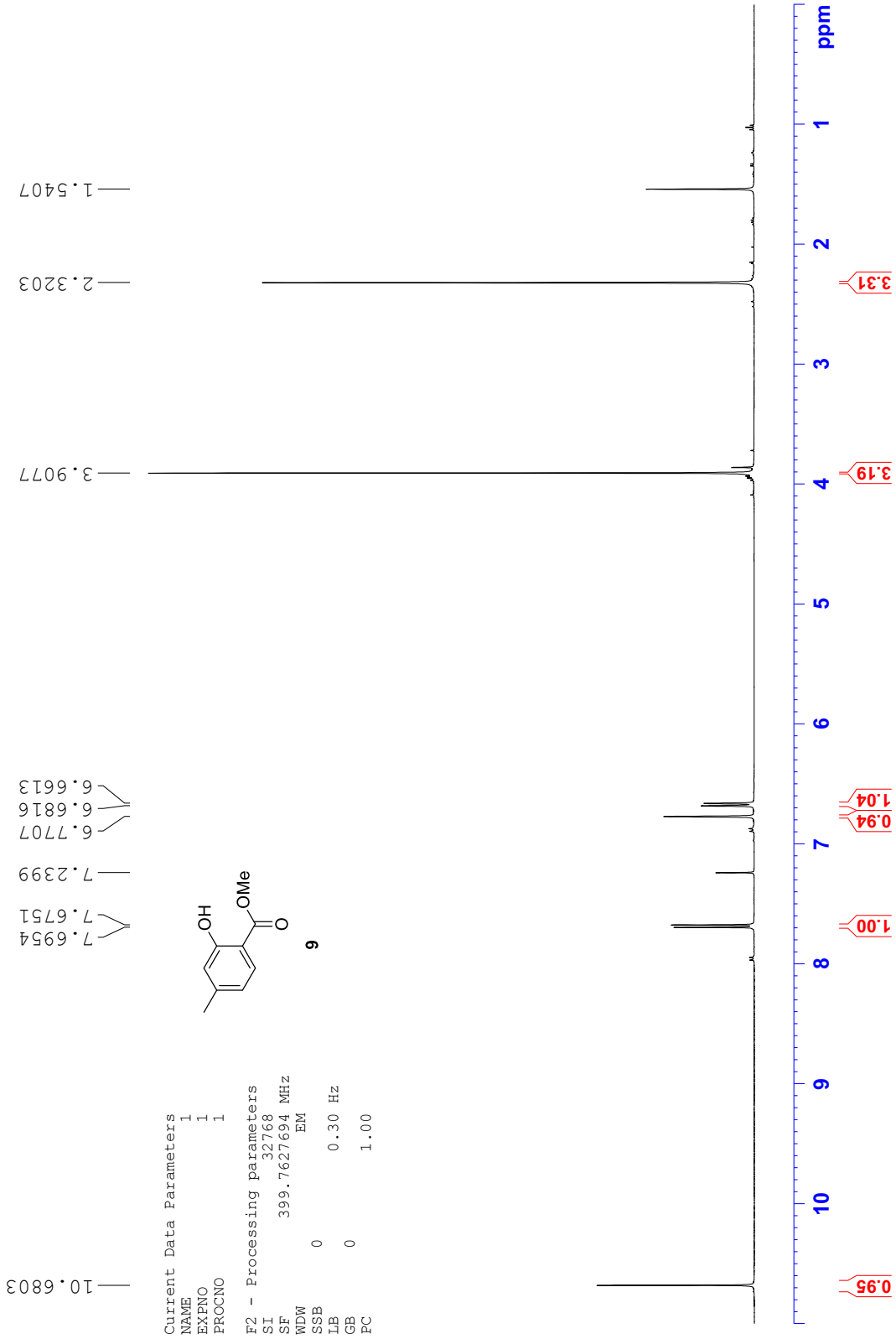



**Fig. S9**. Evaluating the fluorescence of merocyanine dye (acid form of **6**) under different pH conditions.

## Reference


1. W.-T. Chien, C.-F. Liang, C.-C. Yu, C.-H. Lin, S.-P. Li, I. Primadona, Y.-J. Chen, K. K. T. Mong, C.-C. Lin, Sequential one-pot enzymatic synthesis of oligo-*N*-acetyllactosamine and its multi-sialylated extensions. *Chem. Commun.* **50,** 5786-5789 (2014).
2. H.-J. Chen, C. Y. Chew, E.-H. Chang, Y.-W. Tu, L.-Y. Wei, B.-H. Wu, C.-H. Chen, Y.-T. Yang, S.-C. Huang, J.-K. Chen, I. C. Chen, K.-T. Tan, *S*-*Cis* Diene Conformation: A New Bathochromic Shift Strategy for Near-Infrared Fluorescence Switchable Dye and the Imaging Applications. *J. Am. Chem. Soc.* **140,** 5224-5234 (2018).

## NMR spectra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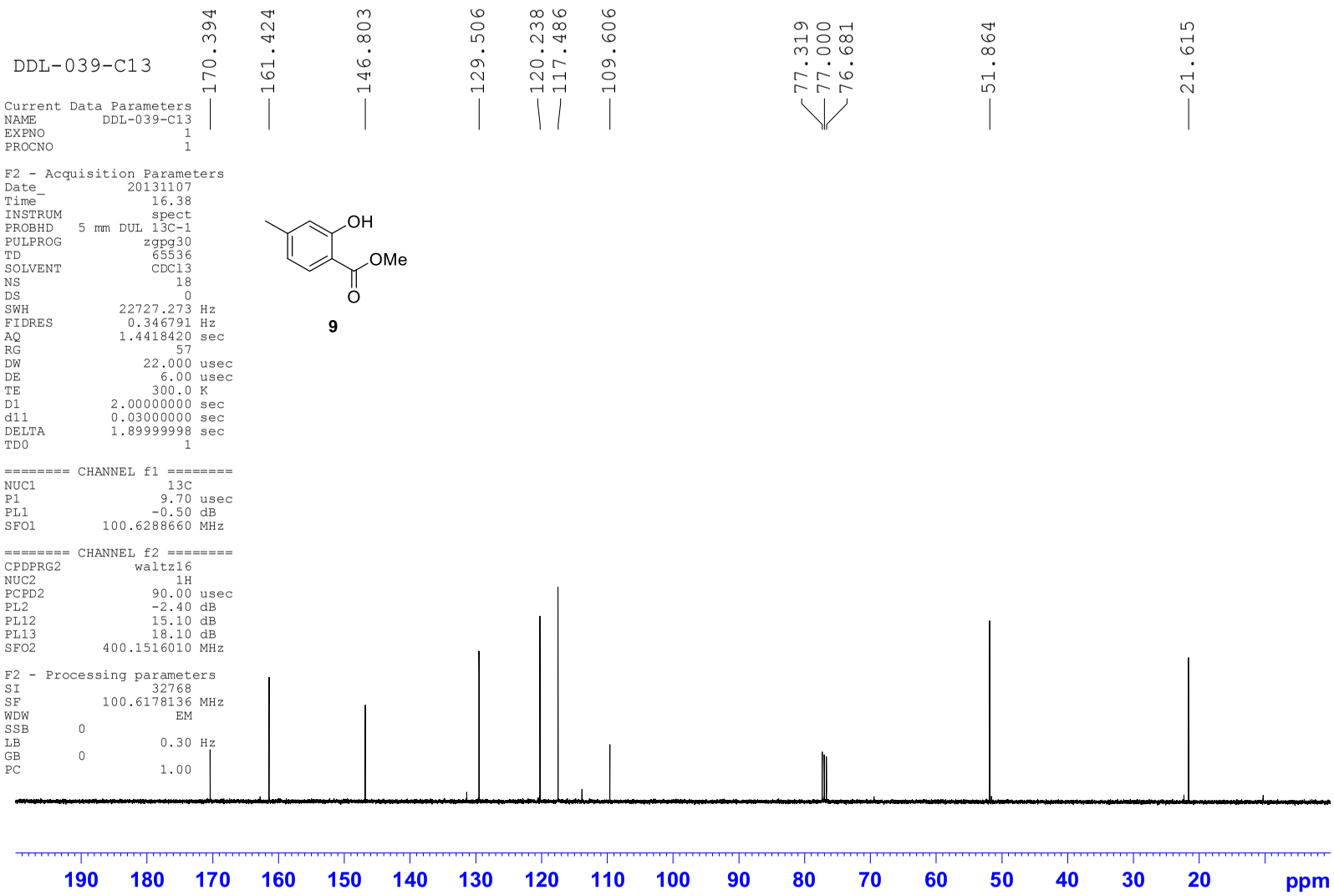
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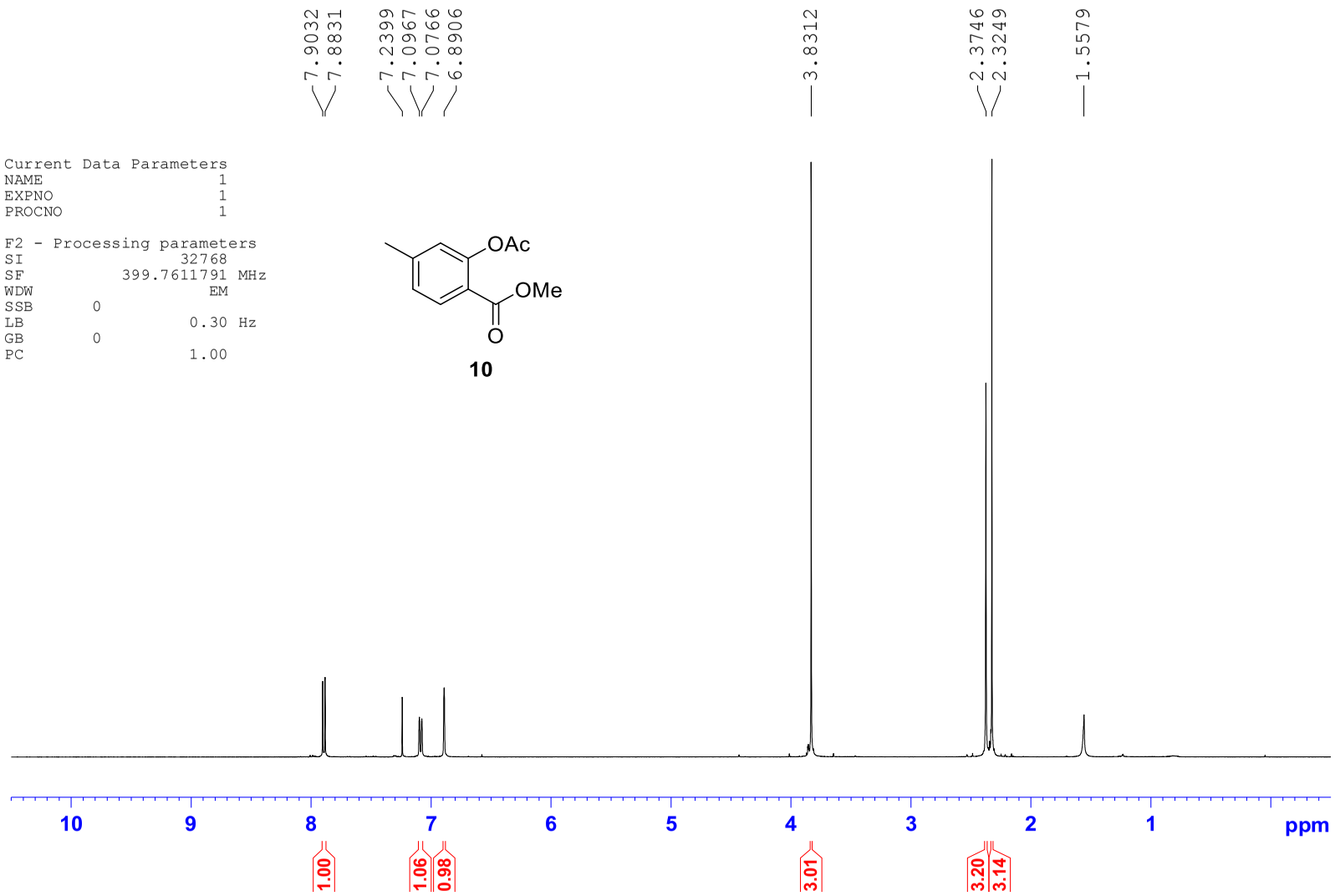
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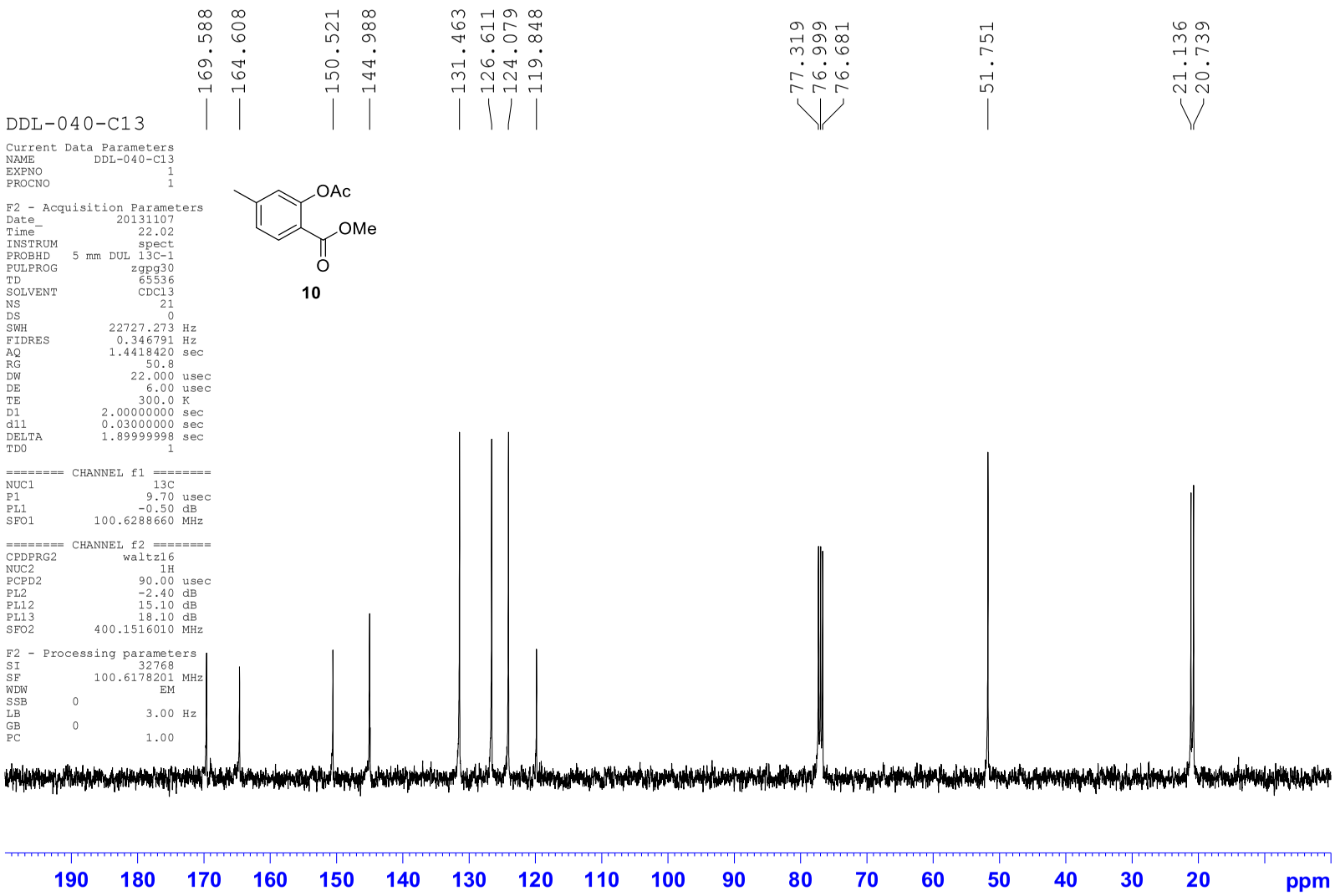
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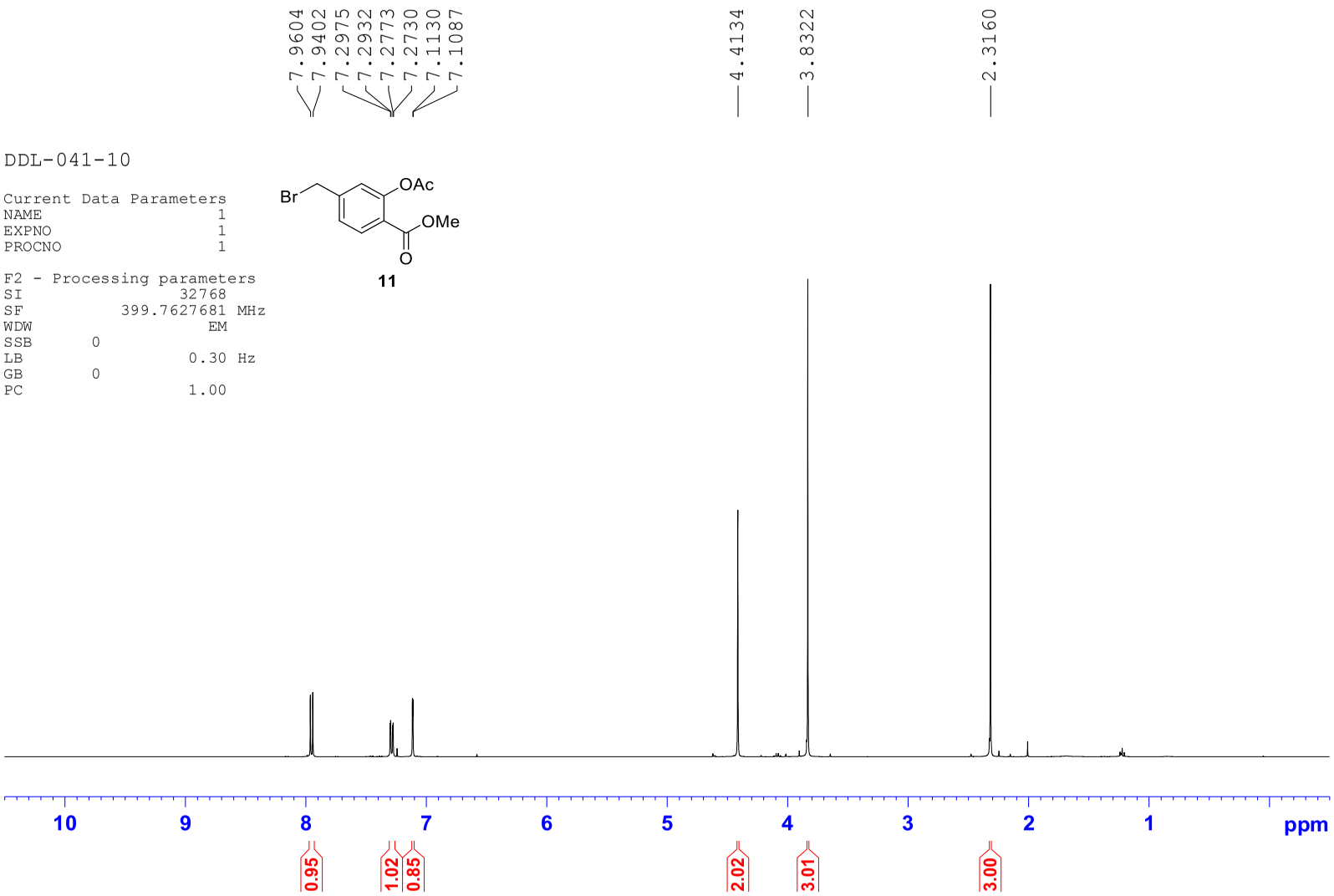
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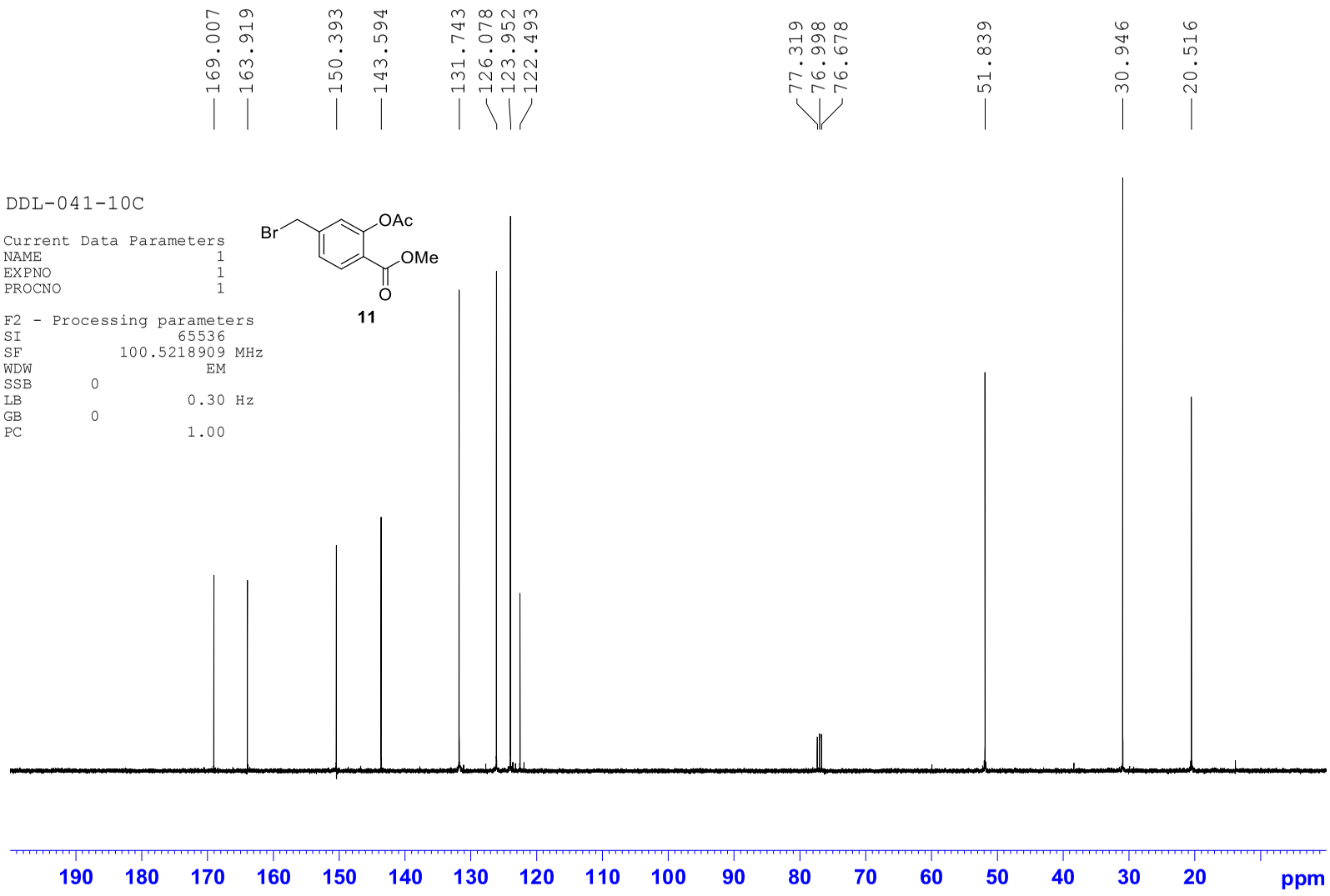
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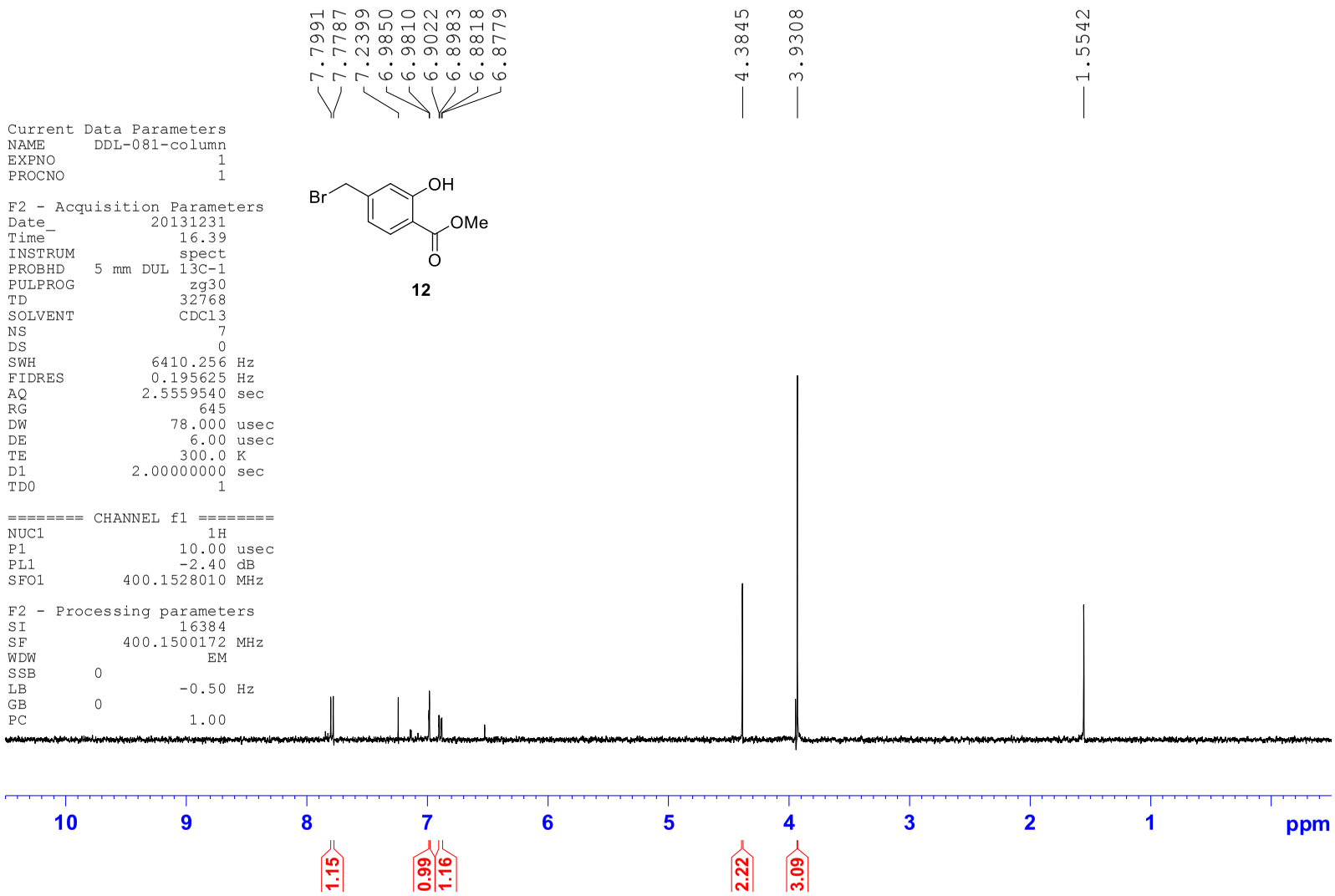
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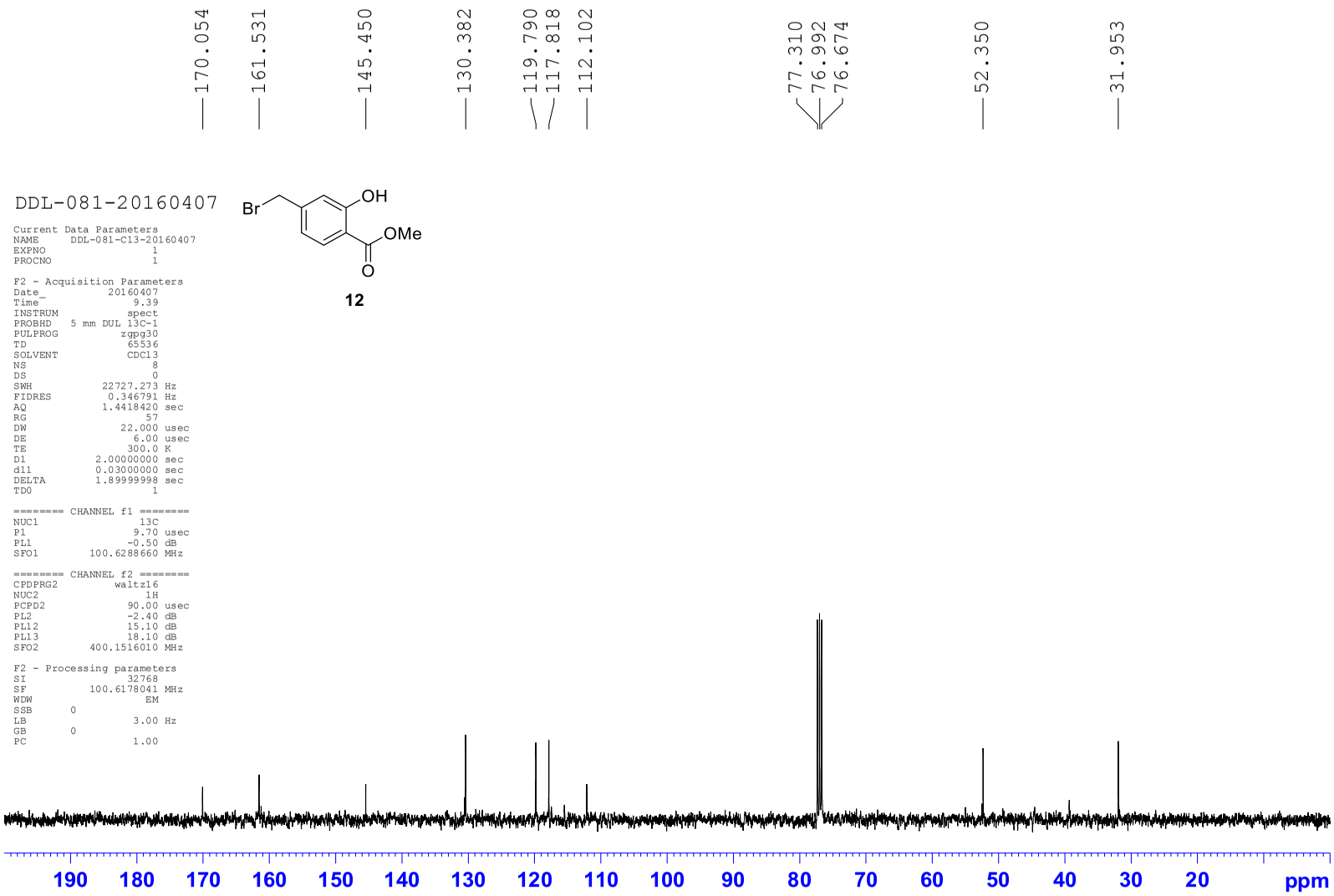
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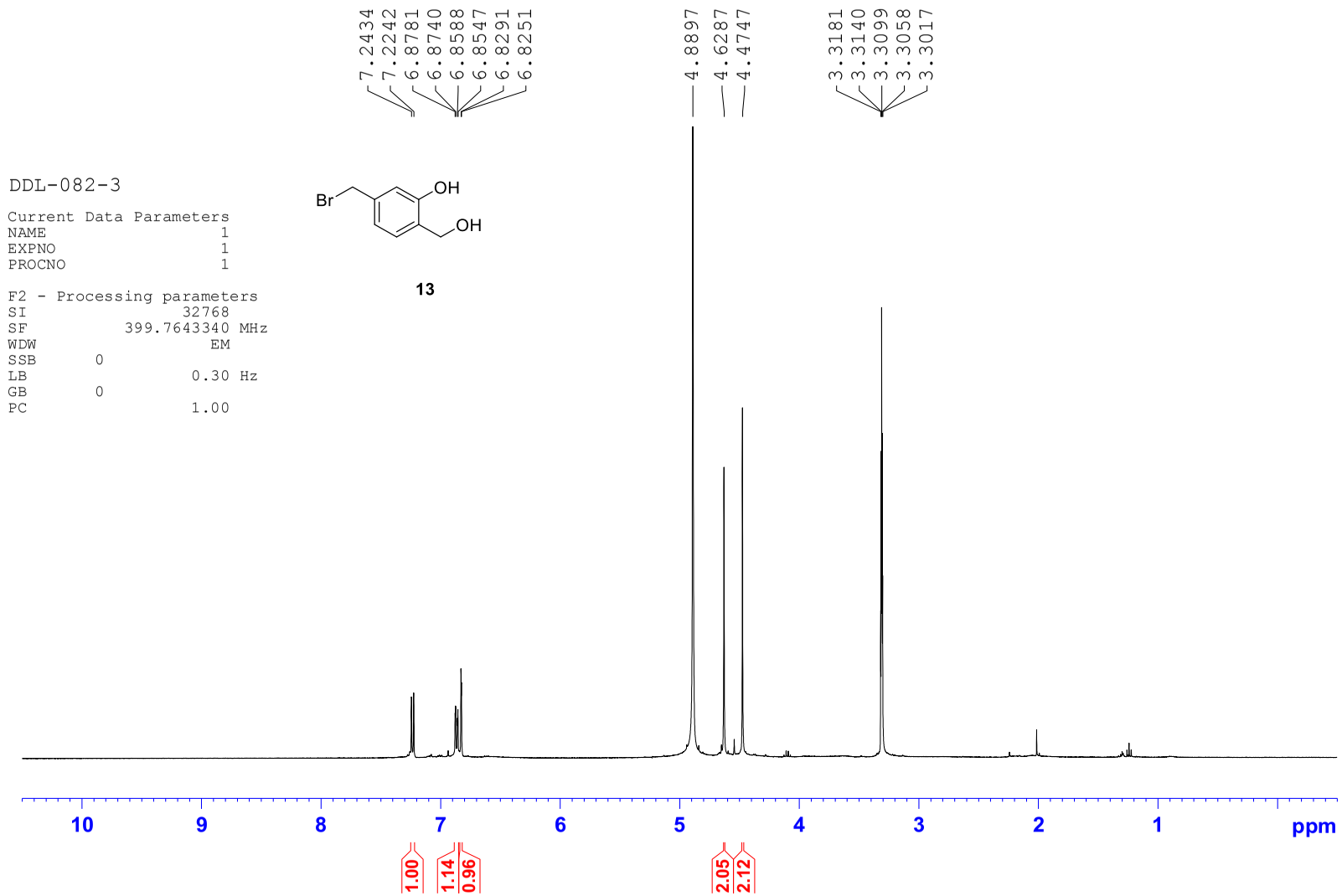
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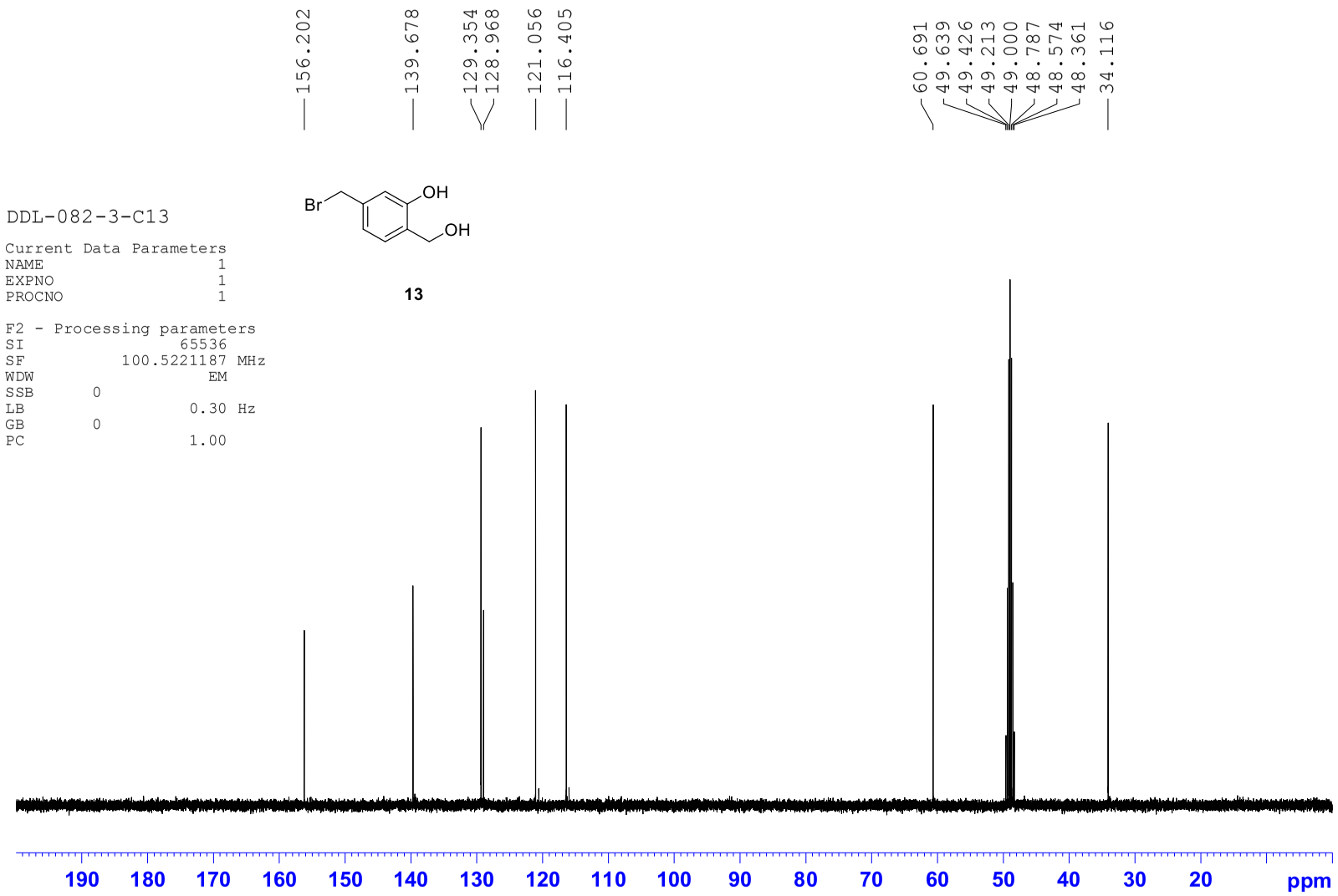
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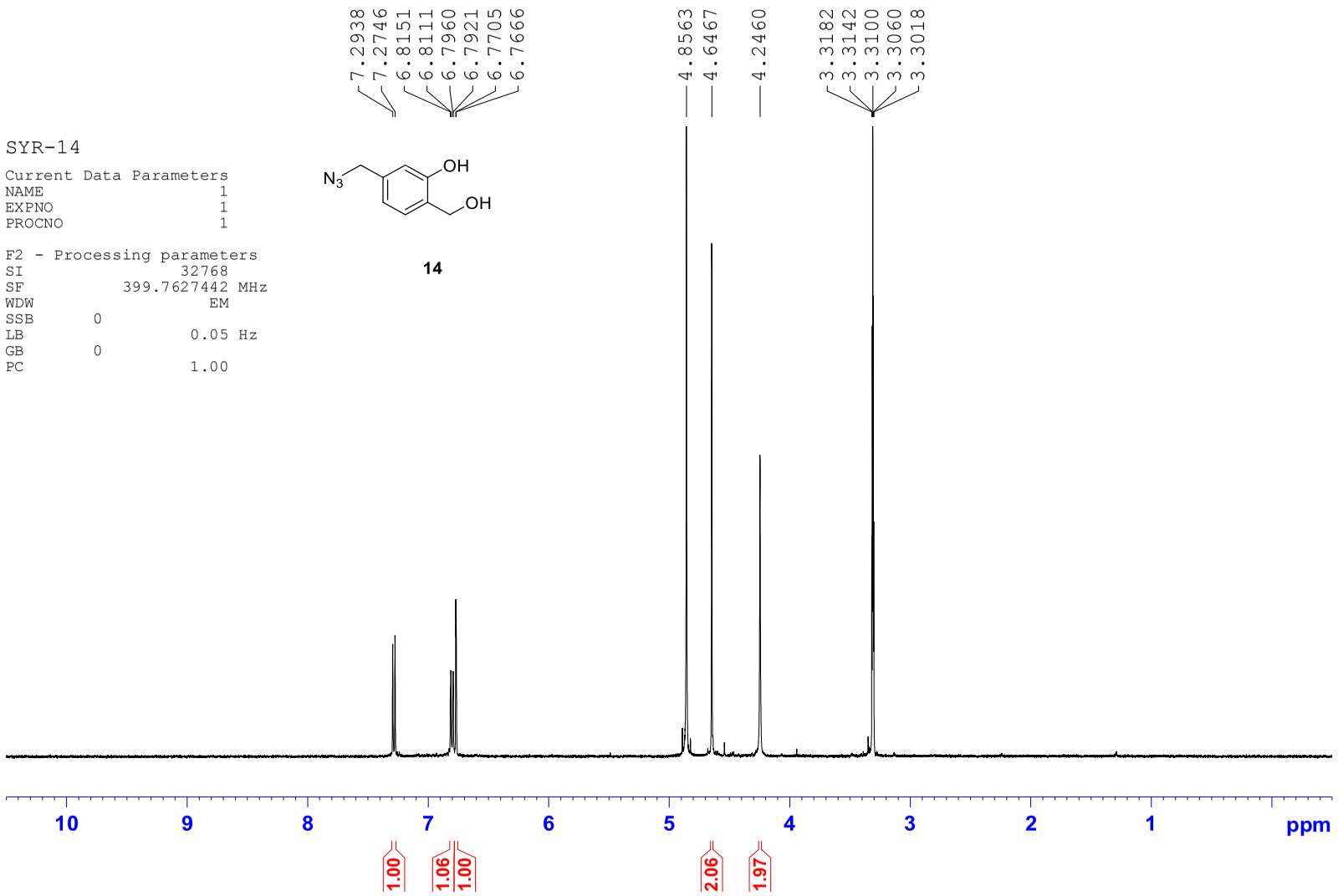
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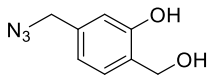
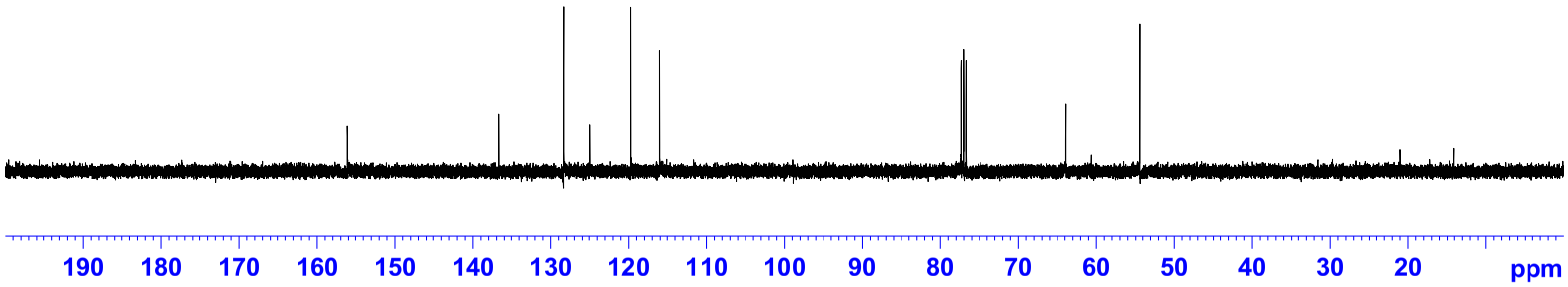
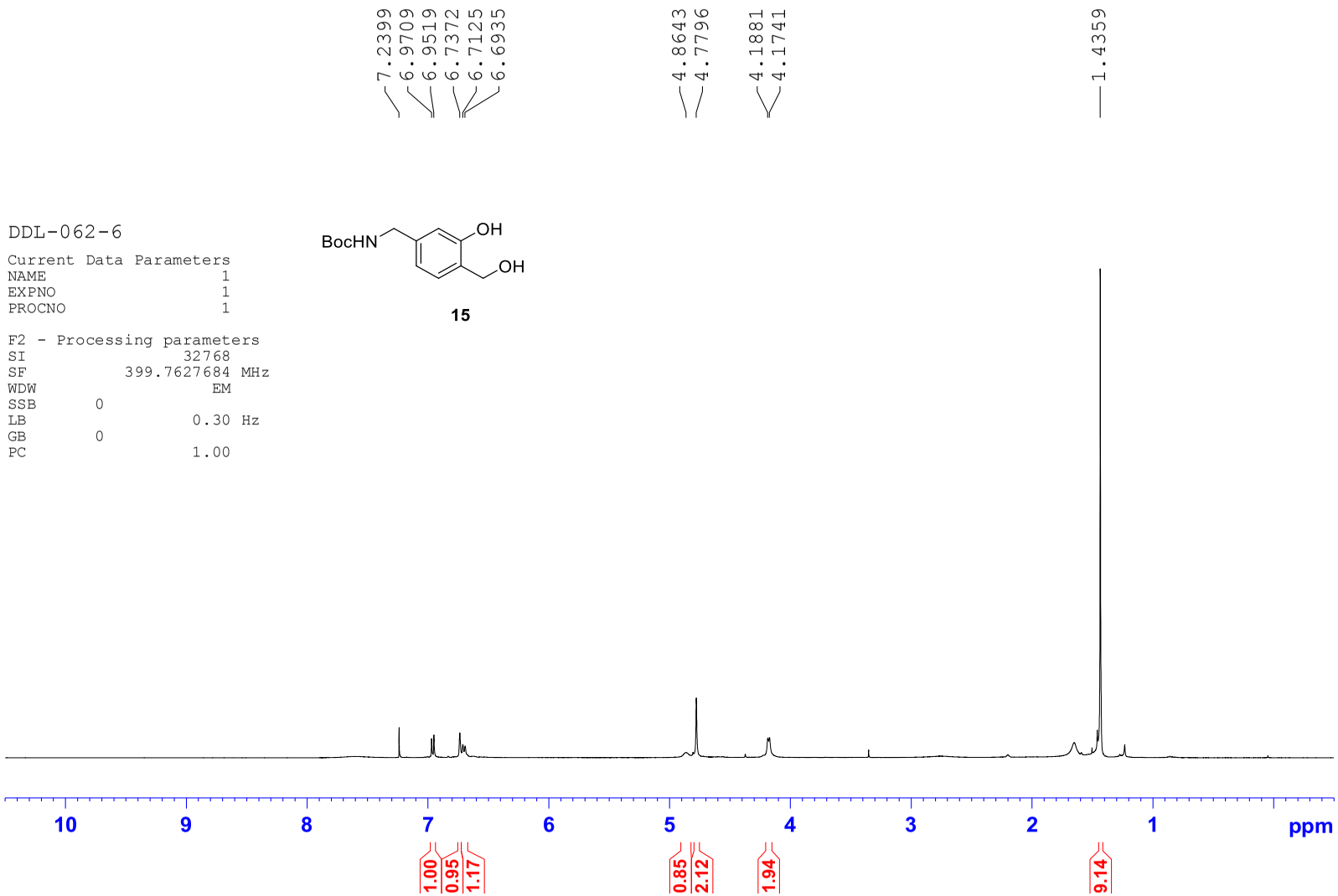



**14**

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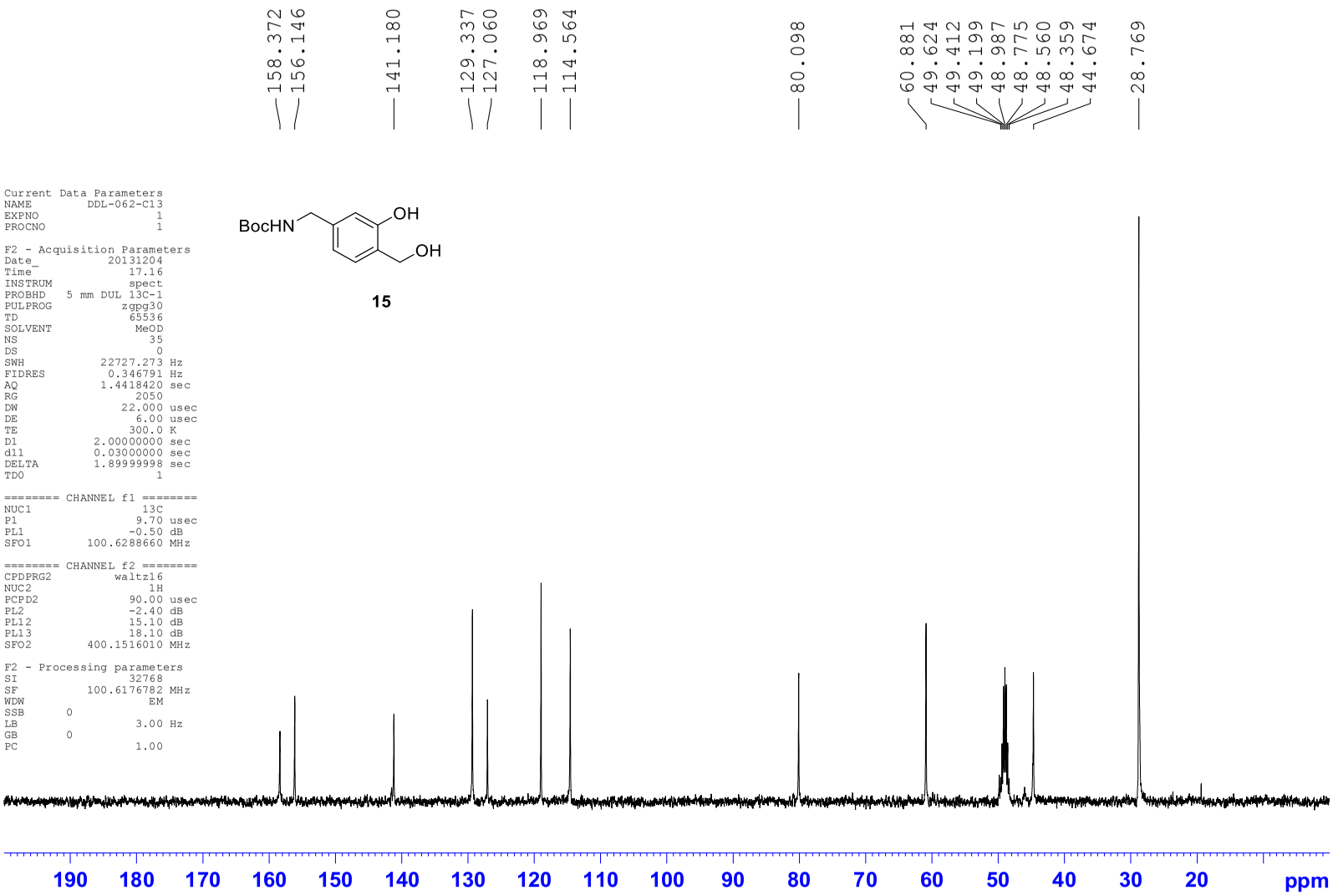
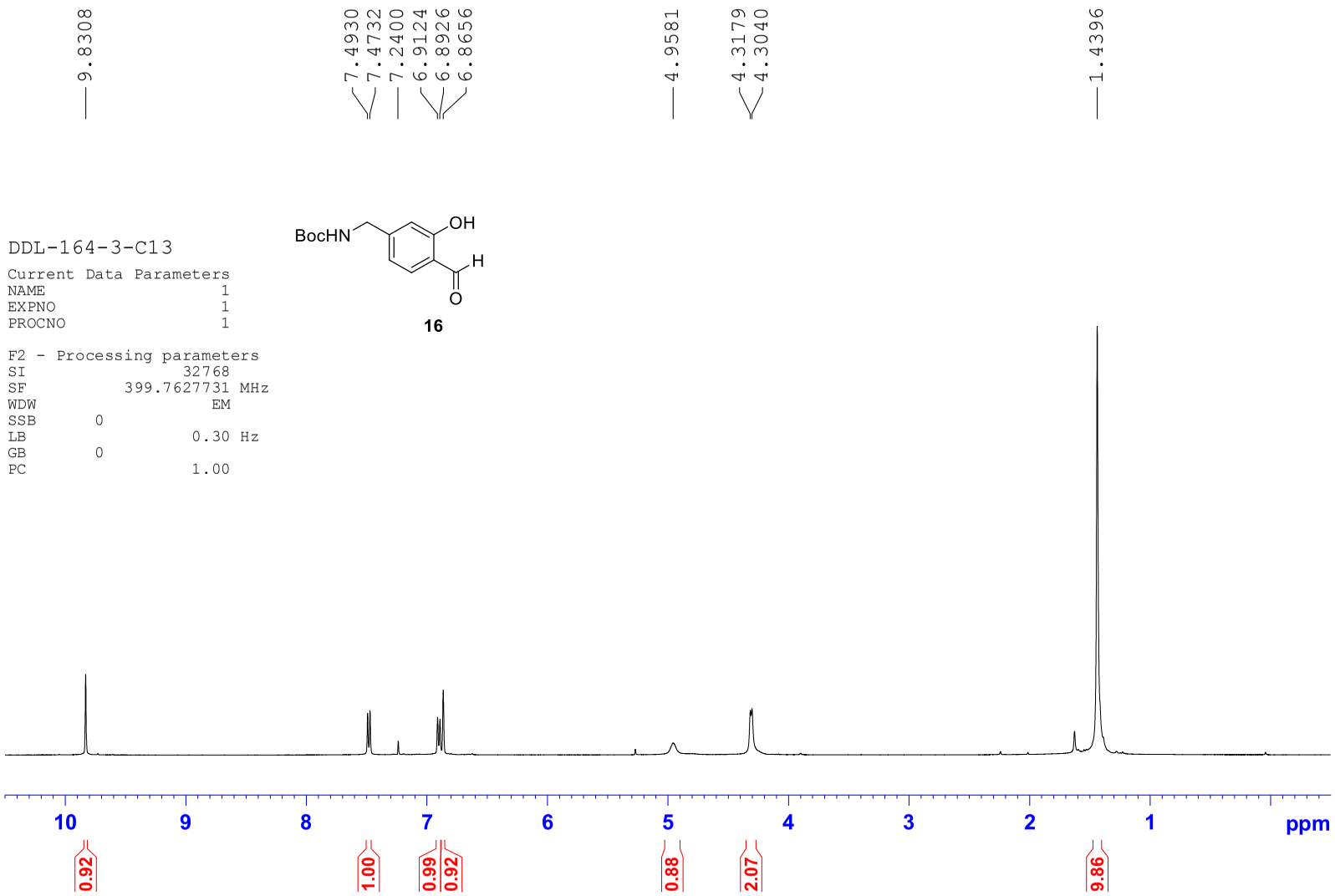
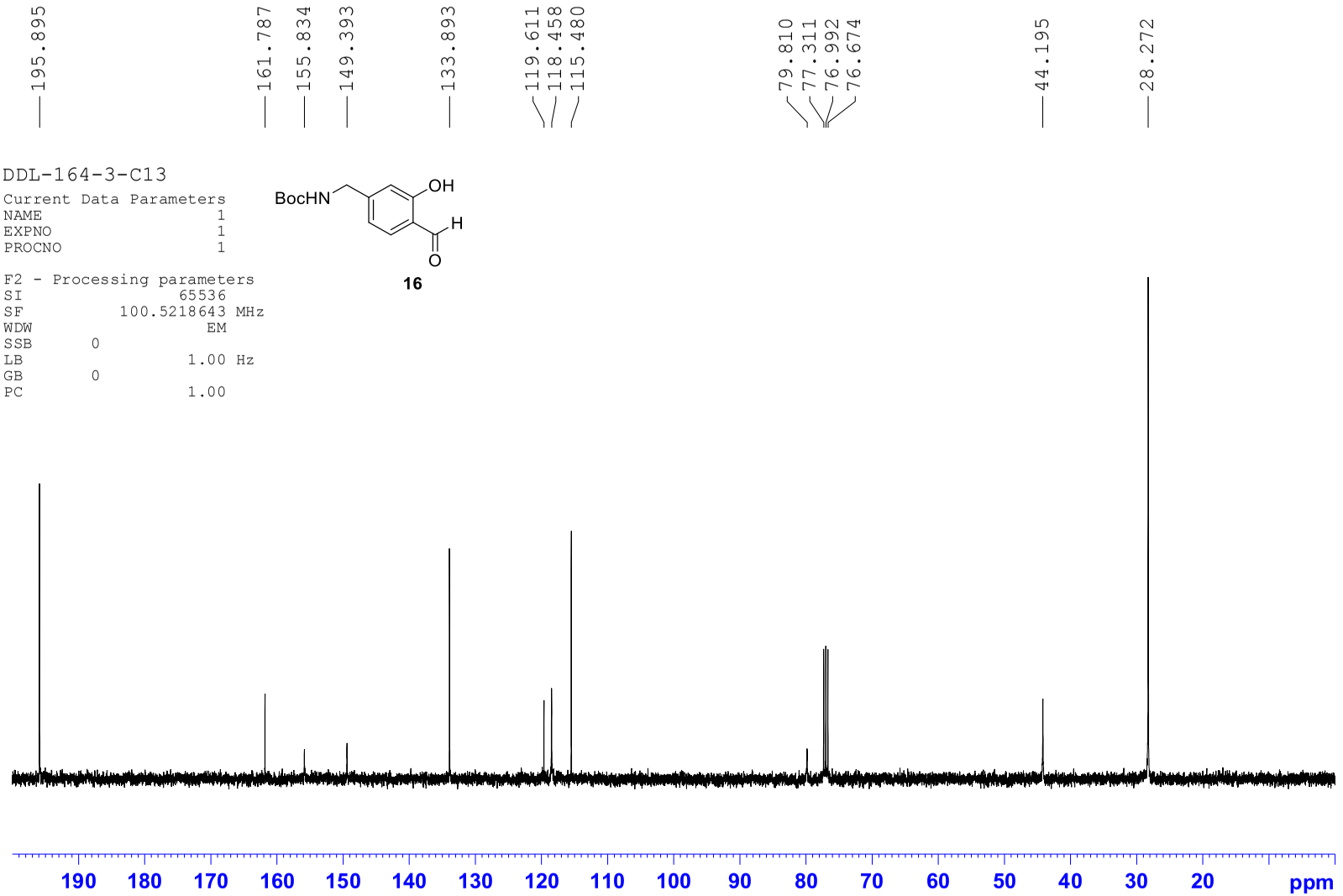
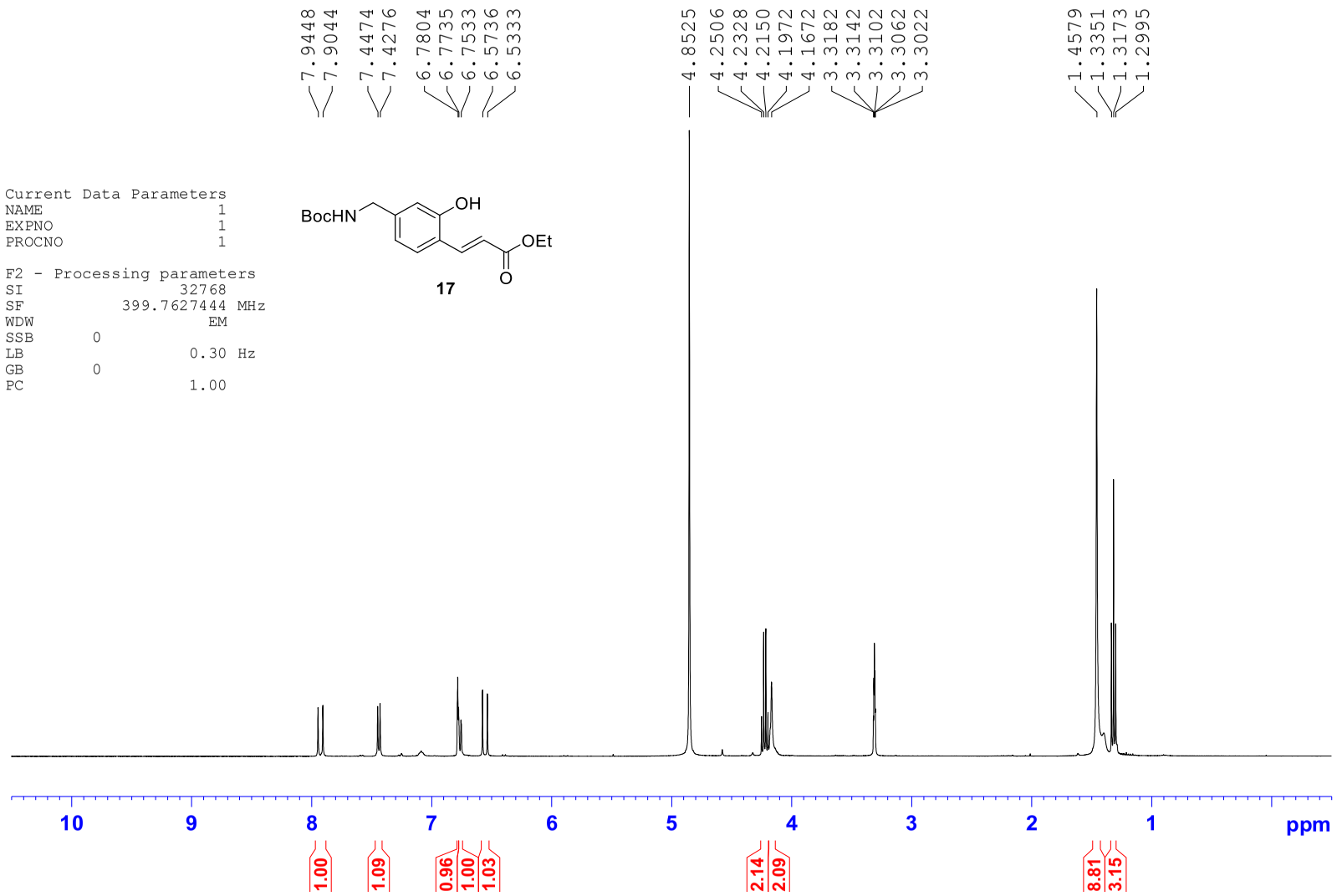
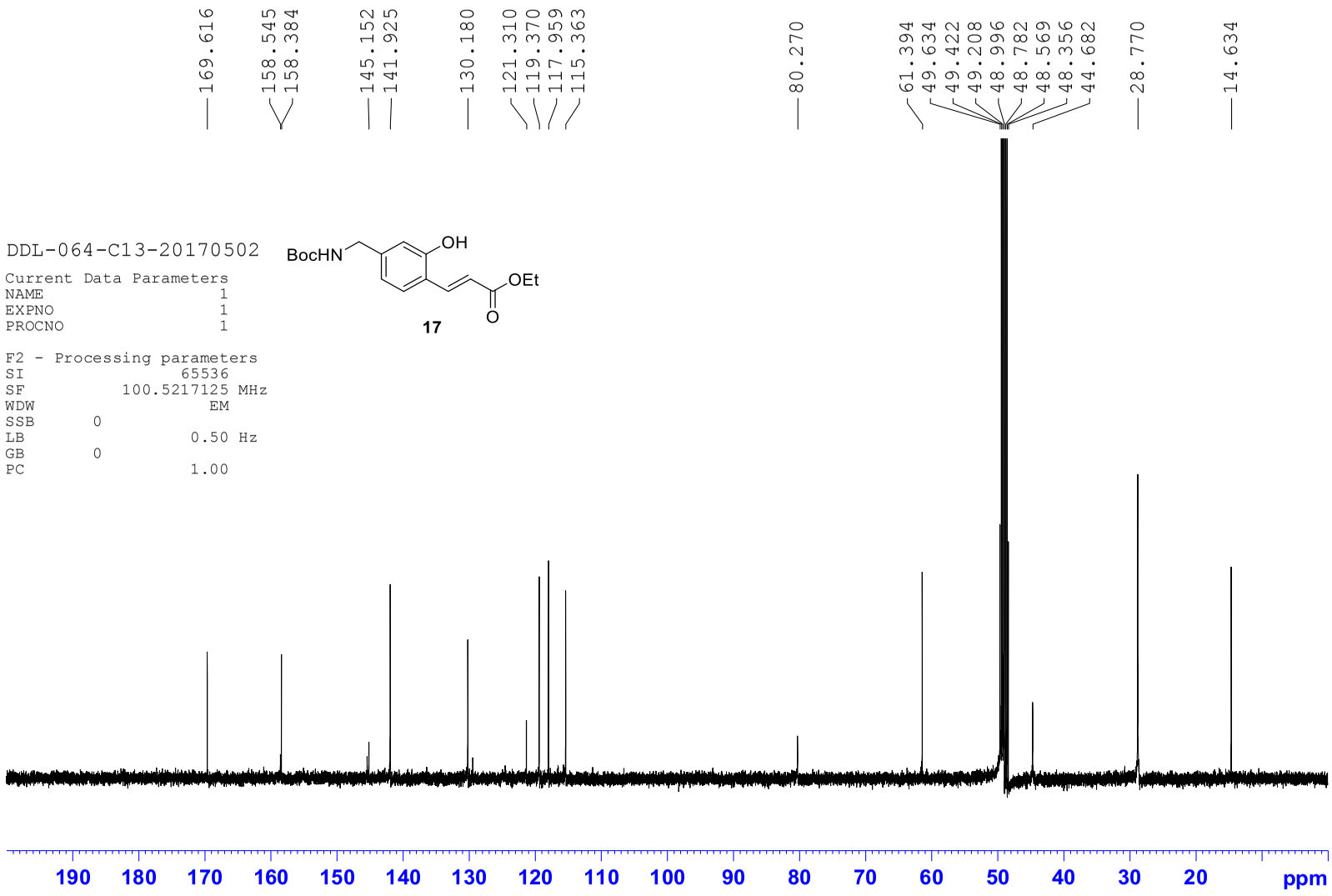
1.94
9.14

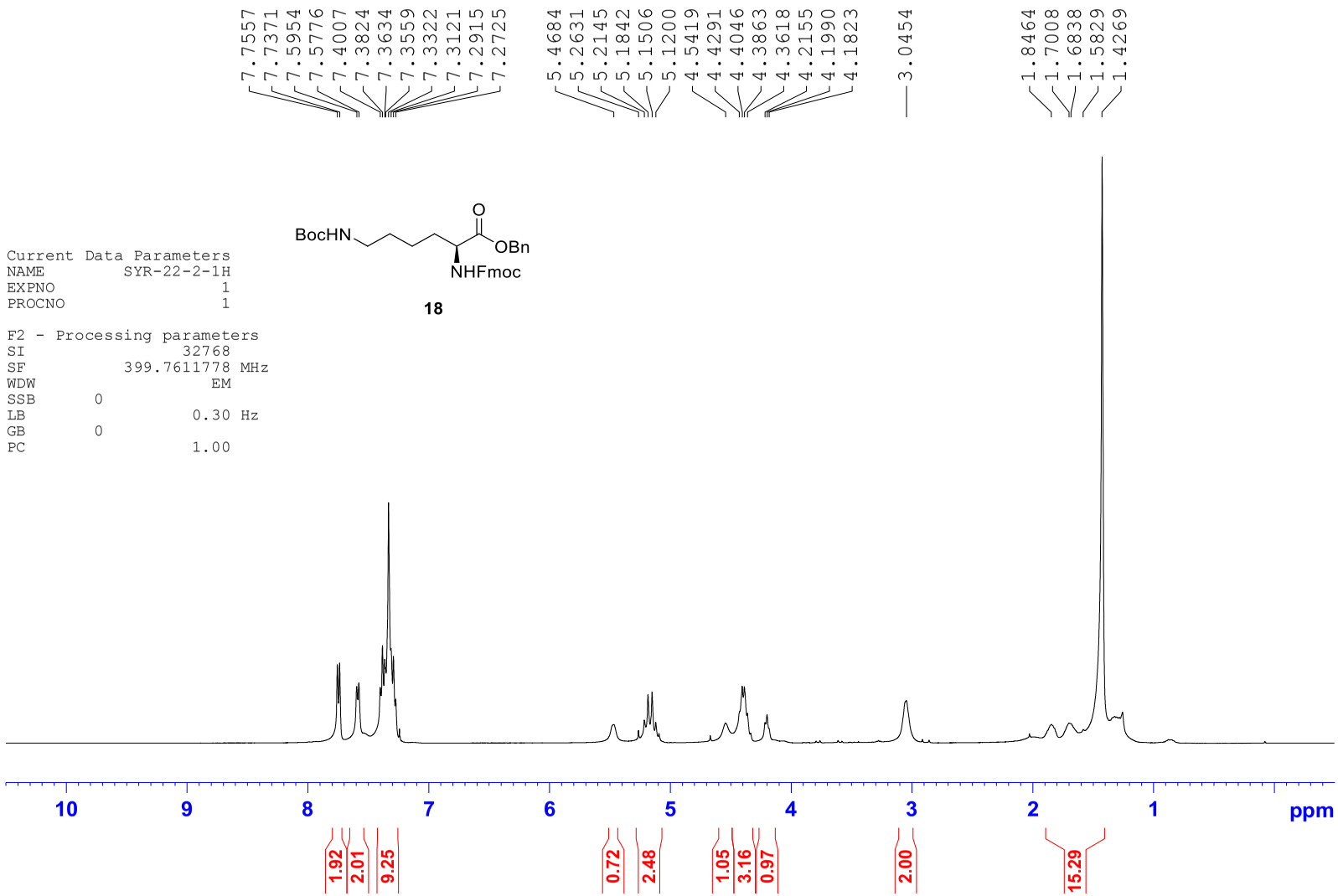

158.372
156.146
141.180
129.337
127.060
118.969
114.564
80.098
60.881
49.624
49.412
49.199
48.987
48.775
48.560
48.359
44.674
28.769
Current Data Parameters
NAME DDL-062-C13
EXPNO 1
PROCNO 1
F2 - Acquisition Parameters
Date_ 20131204
Time 17.16
INSTRUM spect
PROBHD 5 mm DUL 13C-1
PULPROG zgpg30
TD 65536
SOLVENT MeOD
NS 35
DS 0
SWH 22727.273 Hz
FIDRES 0.346791 Hz
AQ 1.4418420 sec
RG 2050
DW 22.000 usec
DE 6.00 usec
TE 300.0 K
D1 2.00000000 sec
d11 0.03000000 sec
DELTA 1.89999998 sec
TD0 1
======== CHANNEL f1 ========
NUC1 13C
P1 9.70 usec
PL1 -0.50 dB
SFO1 100.6288660 MHz
======== CHANNEL f2 ========
CPDPRG2 waltz16
NUC2 1H
PCPD2 90.00 usec
PL2 -2.40 dB
PL12 15.10 dB
PL13 18.10 dB
SFO2 400.1516010 MHz
F2 - Processing parameters
SI 32768
SF 100.6176782 MHz
WDW EM
SSB 0
LB 3.00 Hz
GB 0
PC 1.00
BocHN
OH
OH
15
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

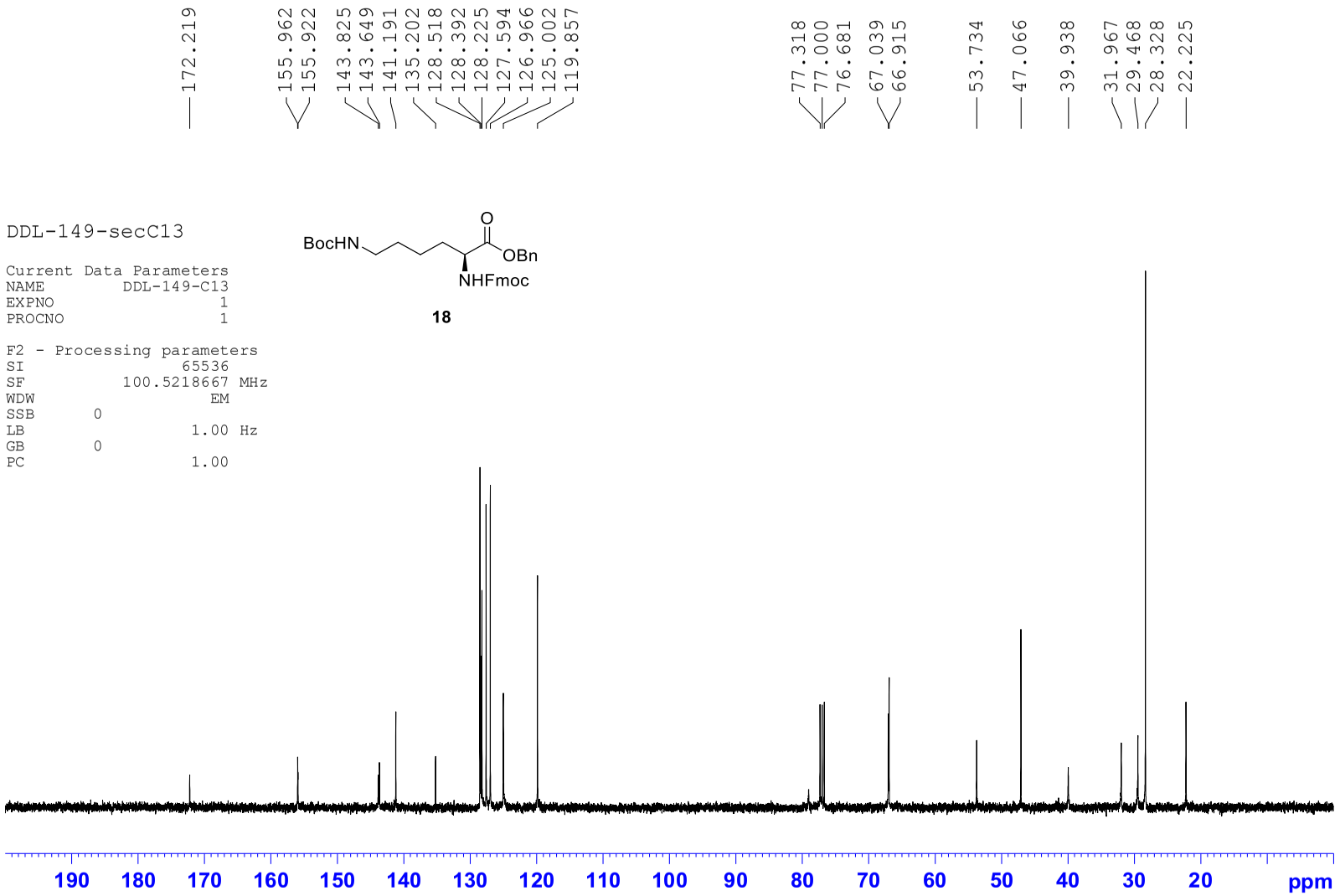

9.8308
7.4930
7.4732
7.2400
6.9124
6.8926
6.8656
4.9581
4.3179
4.3040
1.4396
DDL-164-3-C13
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627731 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
BocHN
OH
H
O
16
10
9
8
7
6
5
4
3
2
1
ppm
0.92
1.00
0.99
0.92
0.88
2.07
9.86

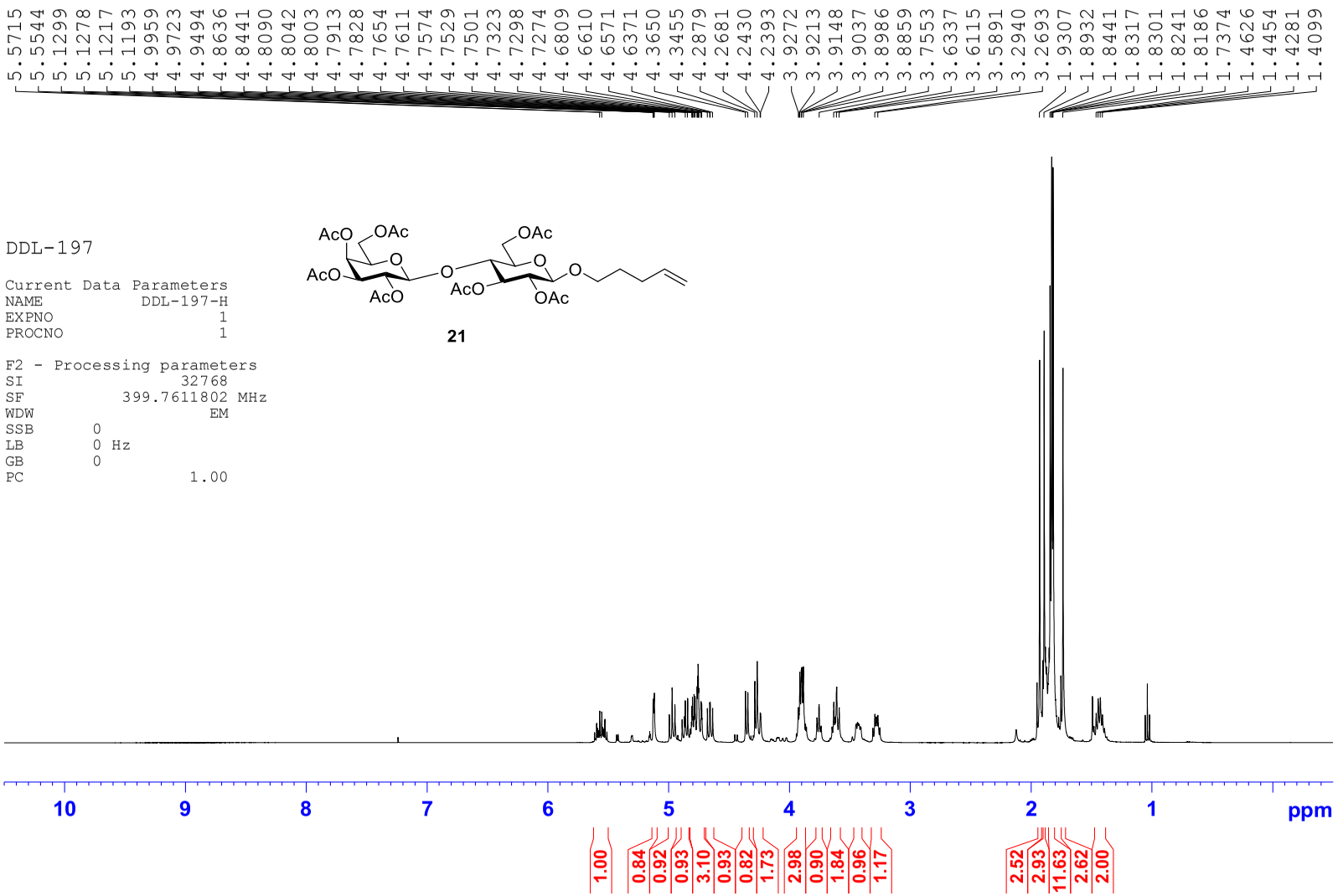

195.895
161.787
155.834
149.393
133.893
119.611
118.458
115.480
79.810
77.311
76.992
76.674
44.195
28.272
DDL-164-3-C13
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5218643 MHz
WDW EM
SSB 0
LB 1.00 Hz
GB 0
PC 1.00
BocHN
OH
H
O
16
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 ppm


7.9448
7.9044
7.4474
7.4276
6.7804
6.7735
6.7533
6.5736
6.5333
4.8525
4.2506
4.2328
4.2150
4.1972
4.1672
3.3182
3.3142
3.3102
3.3062
3.3022
1.4579
1.3351
1.3173
1.2995
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627444 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
BocHN
OH
OEt
O
17
10
9
8
7
6
5
4
3
2
1
ppm
1.00
1.09
0.96
1.00
1.03
2.14
2.09
8.81
3.15


169.616
158.545
158.384
145.152
141.925
130.180
121.310
119.370
117.959
115.363
80.270
61.394
49.634
49.422
49.208
48.996
48.782
48.569
48.356
44.682
28.770
14.634
DDL-064-C13-20170502
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217125 MHz
WDW EM
SSB 0
LB 0.50 Hz
GB 0
PC 1.00
BocHN
OH
OEt
O
17
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 ppm

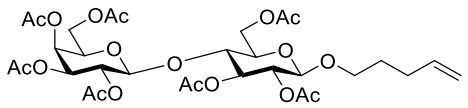

7.7557
7.7371
7.5954
7.5776
7.4007
7.3824
7.3634
7.3559
7.3322
7.3121
7.2915
7.2725
5.4684
5.2631
5.2145
5.1842
5.1506
5.1200
4.5419
4.4291
4.4046
4.3863
4.3618
4.2155
4.1990
4.1823
3.0454
1.8464
1.7008
1.6838
1.5829
1.4269
BocHN
O
OBn
NHFmoc
18
Current Data Parameters
NAME SYR-22-2-1H
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7611778 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
10
9
8
7
6
5
4
3
2
1
ppm
1.92
2.01
9.25
0.72
2.48
1.05
3.16
0.97
2.00
15.29


172.219
155.962
155.922
143.825
143.649
141.191
135.202
128.518
128.392
128.225
127.594
126.966
125.002
119.857
77.318
77.000
76.681
67.039
66.915
53.734
47.066
39.938
31.967
29.468
28.328
22.225
DDL-149-secC13
Current Data Parameters
NAME DDL-149-C13
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5218667 MHz
WDW EM
SSB 0
LB 1.00 Hz
GB 0
PC 1.00
BocHN
O
OBn
NHFmoc
18
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

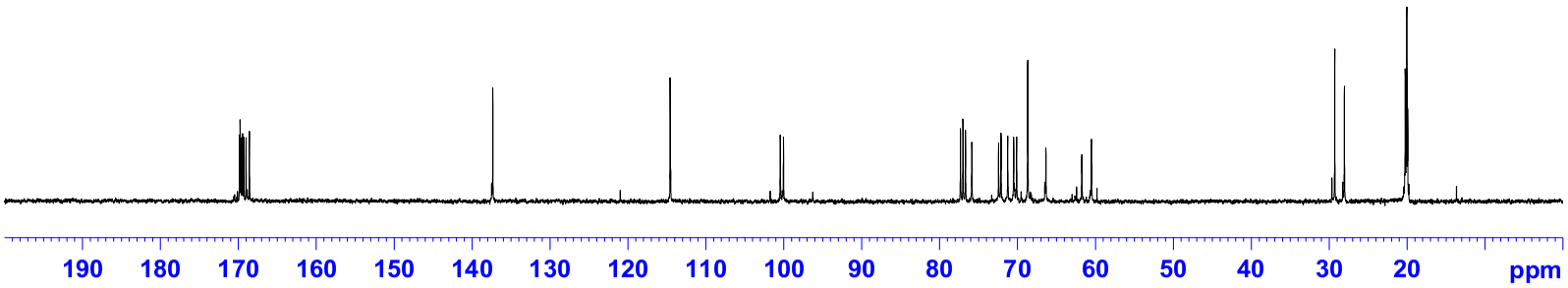

5.5715
5.5544
5.1299
5.1278
5.1217
5.1193
4.9959
4.9723
4.9494
4.8636
4.8441
4.8090
4.8042
4.8003
4.7913
4.7828
4.7654
4.7611
4.7574
4.7529
4.7501
4.7323
4.7298
4.7274
4.6809
4.6610
4.6571
4.6371
4.3650
4.3455
4.2879
4.2681
4.2430
4.2393
3.9272
3.9213
3.9148
3.9037
3.8986
3.8859
3.7553
3.6337
3.6115
3.5891
3.2940
3.2693
1.9307
1.8932
1.8441
1.8317
1.8301
1.8241
1.8186
1.7374
1.4626
1.4454
1.4281
1.4099
DDL-197
Current Data Parameters
NAME DDL-197-H
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7611802 MHz
WDW EM
SSB 0
LB 0 Hz
GB 0
PC 1.00
AcO
OAc
AcO
AcO
OAc
AcO
OAc
21
10
9
8
7
6
5
4
3
2
1
ppm
1.00
0.84
0.92
0.93
3.10
0.93
0.82
1.73
2.98
0.90
1.84
0.96
1.17
2.52
2.93
11.63
2.62
2.00

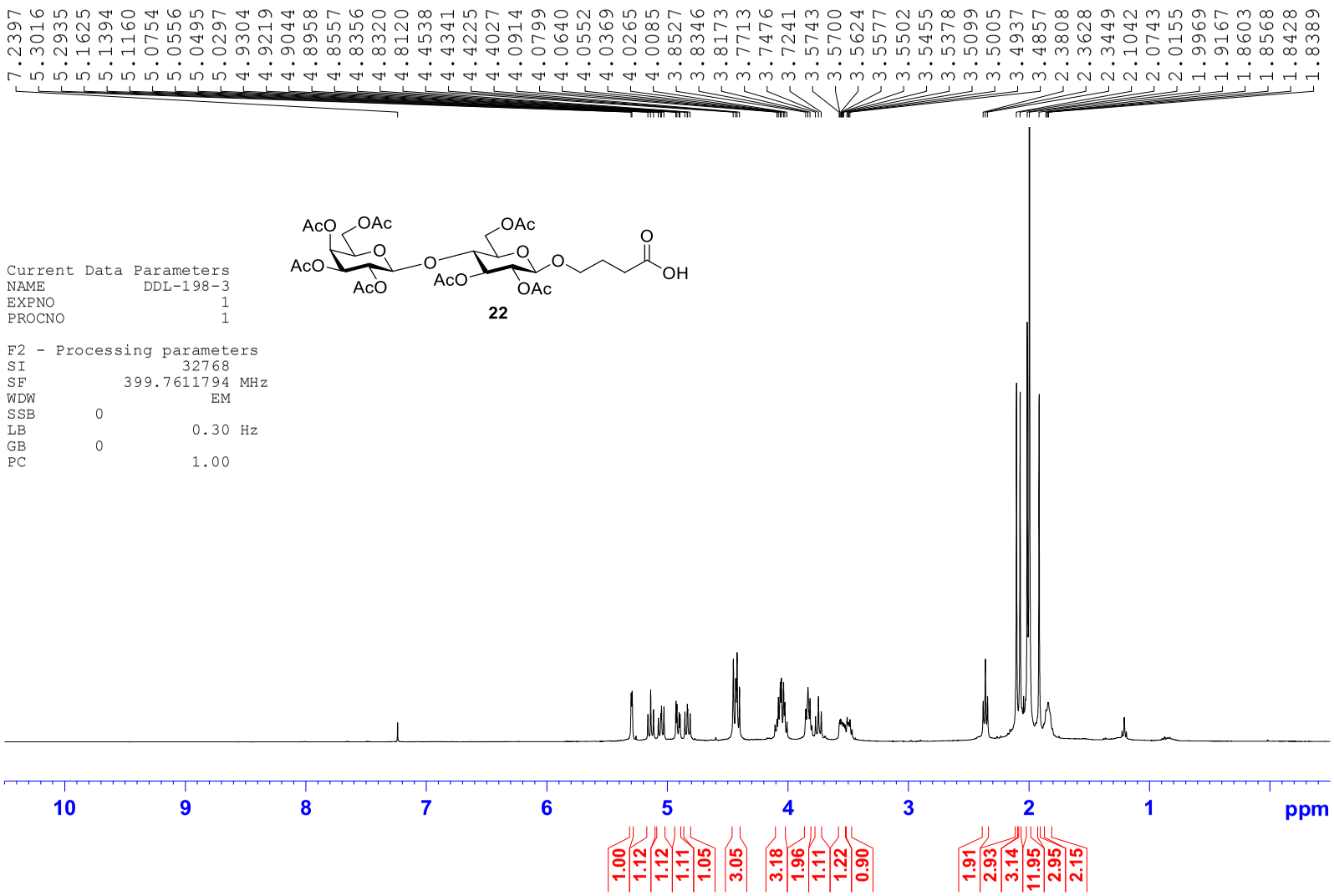
169.839
169.763
169.635
169.446
169.273
168.996
168.574
137.328
114.571
100.436
100.021
77.315
76.995
76.673
75.853
72.416
72.104
71.244
70.482
70.102
68.678
66.342
61.737
60.497
29.308
28.072
20.267
20.240
20.120
20.070
20.055
20.040
19.943

DDL-197




**21**




7.2397
5.3016
5.2935
5.1625
5.1394
5.1160
5.0754
5.0556
5.0495
5.0297
4.9304
4.9219
4.9044
4.8958
4.8557
4.8356
4.8320
4.8120
4.4538
4.4341
4.4225
4.4027
4.0914
4.0799
4.0640
4.0552
4.0369
4.0265
4.0085
3.8527
3.8346
3.8173
3.7713
3.7476
3.7241
3.5743
3.5700
3.5624
3.5577
3.5502
3.5455
3.5378
3.5099
3.5005
3.4937
3.4857
2.3808
2.3628
2.3449
2.1042
2.0743
2.0155
1.9969
1.9167
1.8603
1.8568
1.8428
1.8389
AcO
OAc
AcO
AcO
OAc
AcO
OAc
OAc
O
OH
22
Current Data Parameters
NAME DDL-198-3
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7611794 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
10
9
8
7
6
5
4
3
2
1
ppm
1.00
1.12
1.12
1.11
1.05
3.05
3.18
1.96
1.11
1.22
0.90
1.91
2.93
3.14
11.95
2.95
2.15

177.761
170.448
170.326
170.118
170.025
169.770
169.687
169.053

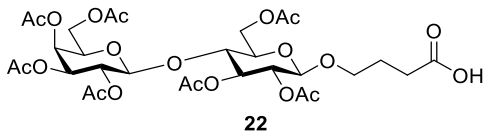
100.945
100.387

77.316
76.997
76.679
76.139
72.720
72.526
71.558
70.901
70.583
69.055
68.498
66.572
61.895
60.758

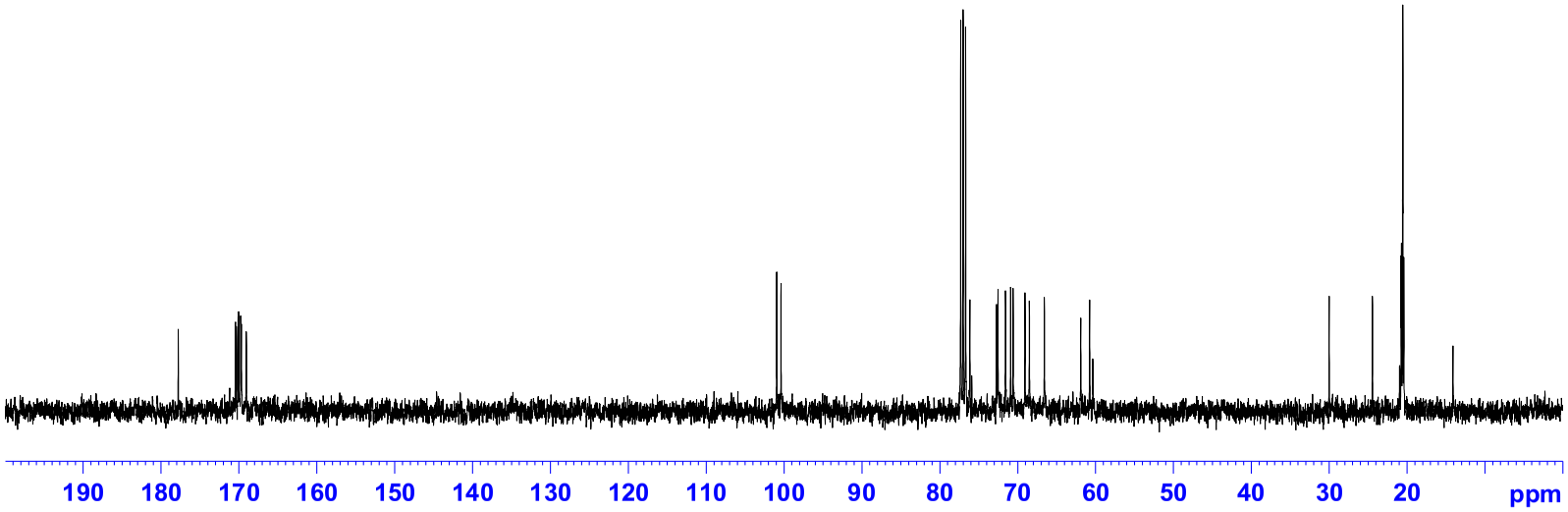
29.996
24.439
20.747
20.705
20.567
20.535
20.409
14.092

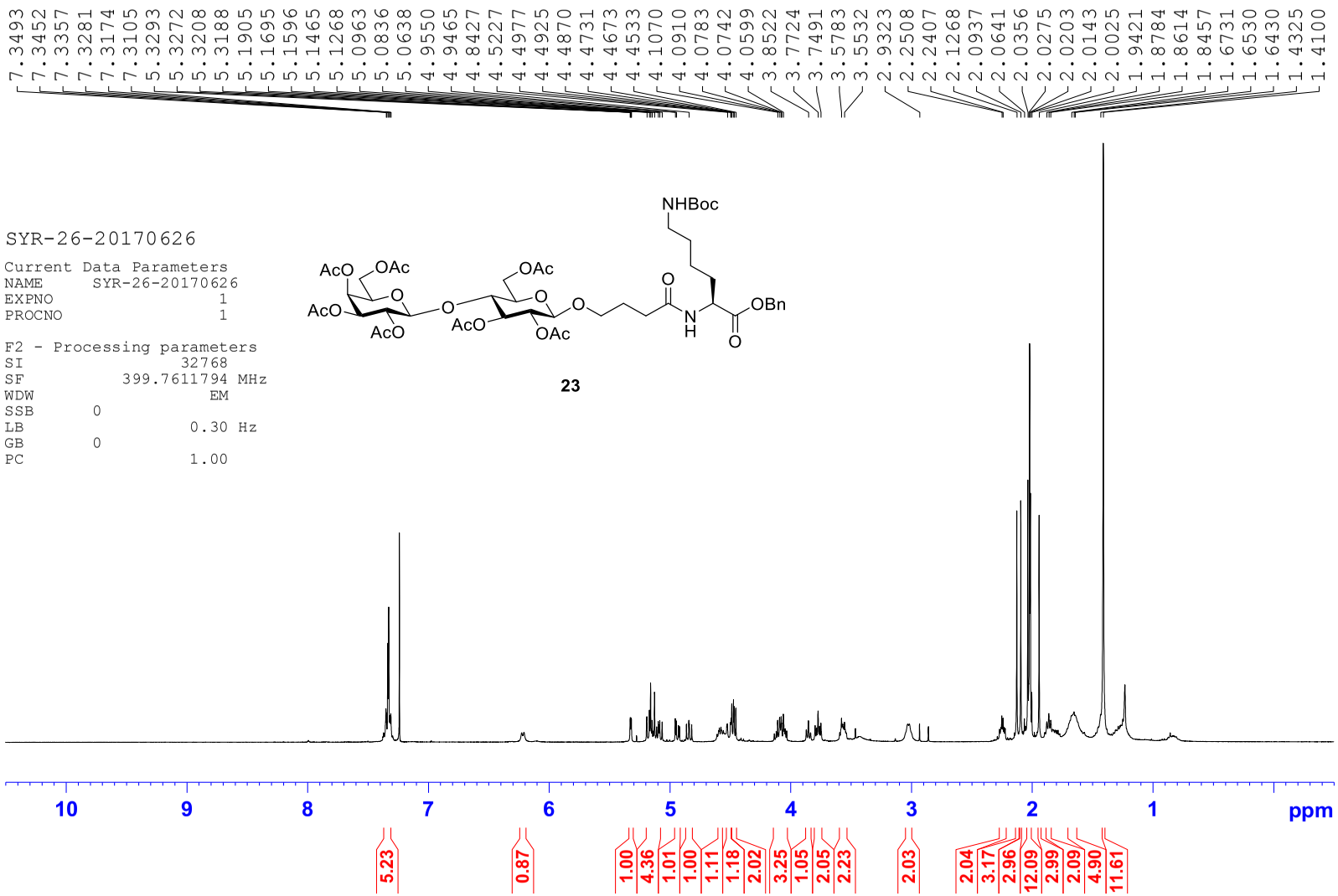
Current Data Parameters
NAME DDL-198-2-C-2
EXPNO 1
PROCNO 1

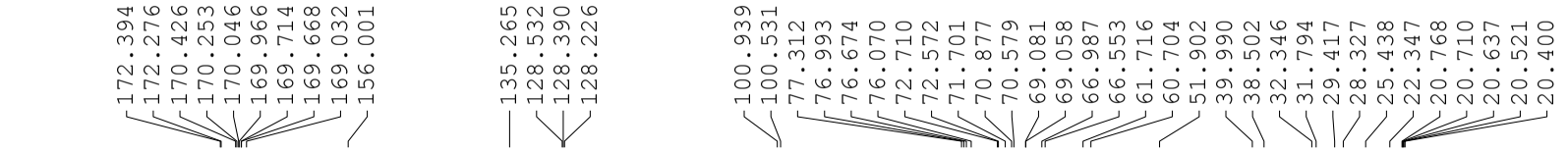
F2 - Processing parameters
SI 65536
SF 100.5214612 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00

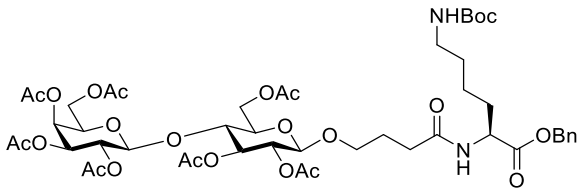



22




7.3493
7.3452
7.3357
7.3281
7.3174
7.3105
5.3293
5.3272
5.3208
5.3188
5.1905
5.1695
5.1596
5.1465
5.1268
5.0963
5.0836
5.0638
4.9550
4.9465
4.8427
4.5227
4.4977
4.4925
4.4870
4.4731
4.4673
4.4533
4.1070
4.0910
4.0783
4.0742
4.0599
3.8522
3.7724
3.7491
3.5783
3.5532
2.9323
2.2508
2.2407
2.1268
2.0937
2.0641
2.0356
2.0275
2.0203
2.0143
2.0025
1.9421
1.8784
1.8614
1.8457
1.6731
1.6530
1.6430
1.4325
1.4100
SYR-26-20170626
Current Data Parameters
NAME SYR-26-20170626
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7611794 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
NHBoc
AcO
OAc
OAc
AcO
AcO
AcO
OAc
OAc
O
N
H
OBn
23
10
9
8
7
6
5
4
3
2
1
ppm
5.23
0.87
1.00
4.36
1.01
1.00
1.11
1.18
2.02
3.25
1.05
2.05
2.23
2.03
2.04
3.17
2.96
12.09
2.99
2.09
4.90
11.61

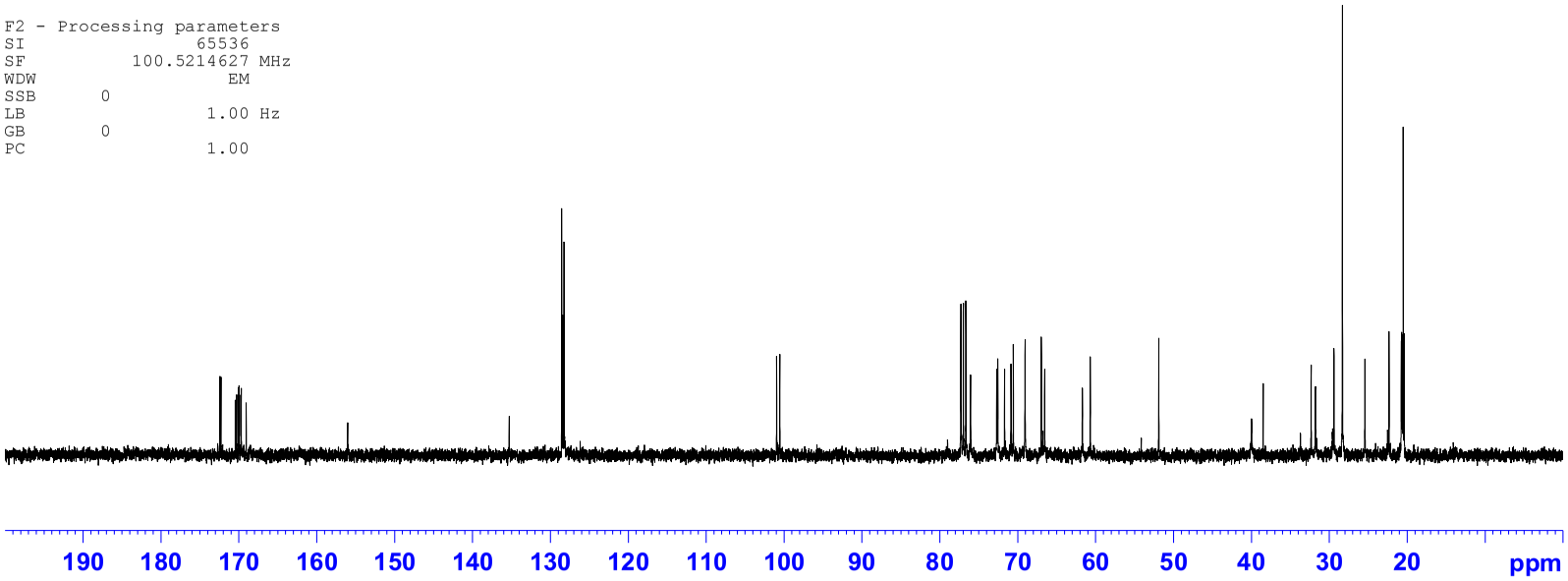


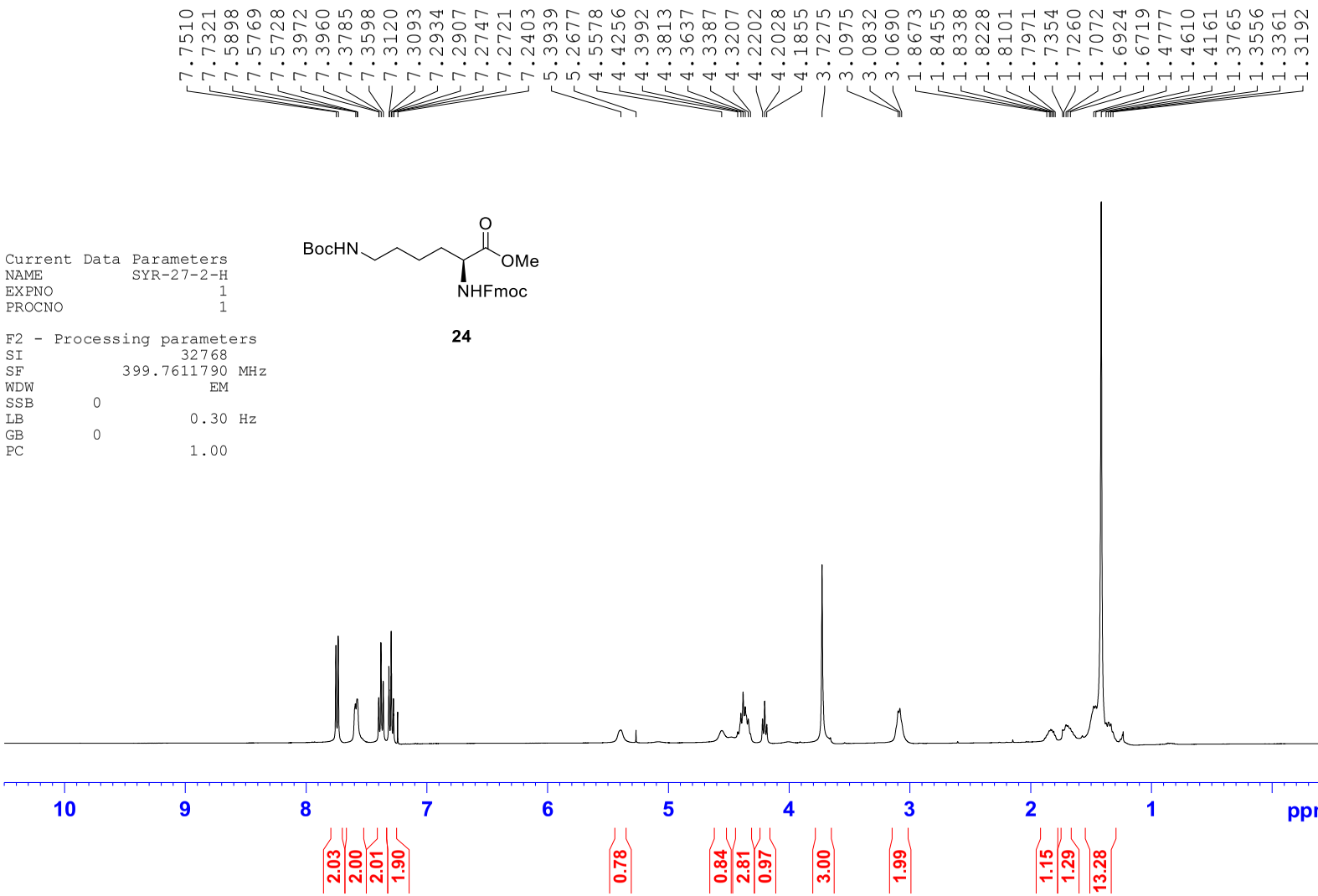




**23**

DDL-202-20160902-C13

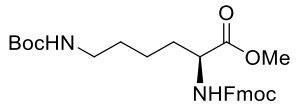
```
Current Data Parameters
NAME      DDL-202-20160902-C13
EXPNO                       1
PROCNO                      1

F2 - Processing parameters
SI                      65536
SF           100.5214627 MHz
WDW                        EM
SSB       0
LB                   1.00 Hz
GB        0
PC                   1.00
```




7.7510
7.7321
7.5898
7.5769
7.5728
7.3972
7.3960
7.3785
7.3598
7.3120
7.3093
7.2934
7.2907
7.2747
7.2721
7.2403
5.3939
5.2677
4.5578
4.4256
4.3992
4.3813
4.3637
4.3387
4.3207
4.2202
4.2028
4.1855
3.7275
3.0975
3.0832
3.0690
1.8673
1.8455
1.8338
1.8228
1.8101
1.7971
1.7354
1.7260
1.7072
1.6924
1.6719
1.4777
1.4610
1.4161
1.3765
1.3556
1.3361
1.3192
Current Data Parameters
NAME SYR-27-2-H
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7611790 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
BocHN
O
OMe
NHFmoc
24
10
9
8
7
6
5
4
3
2
1
ppm
2.03
2.00
2.01
1.90
0.78
0.84
2.81
0.97
3.00
1.99
1.15
1.29
13.28

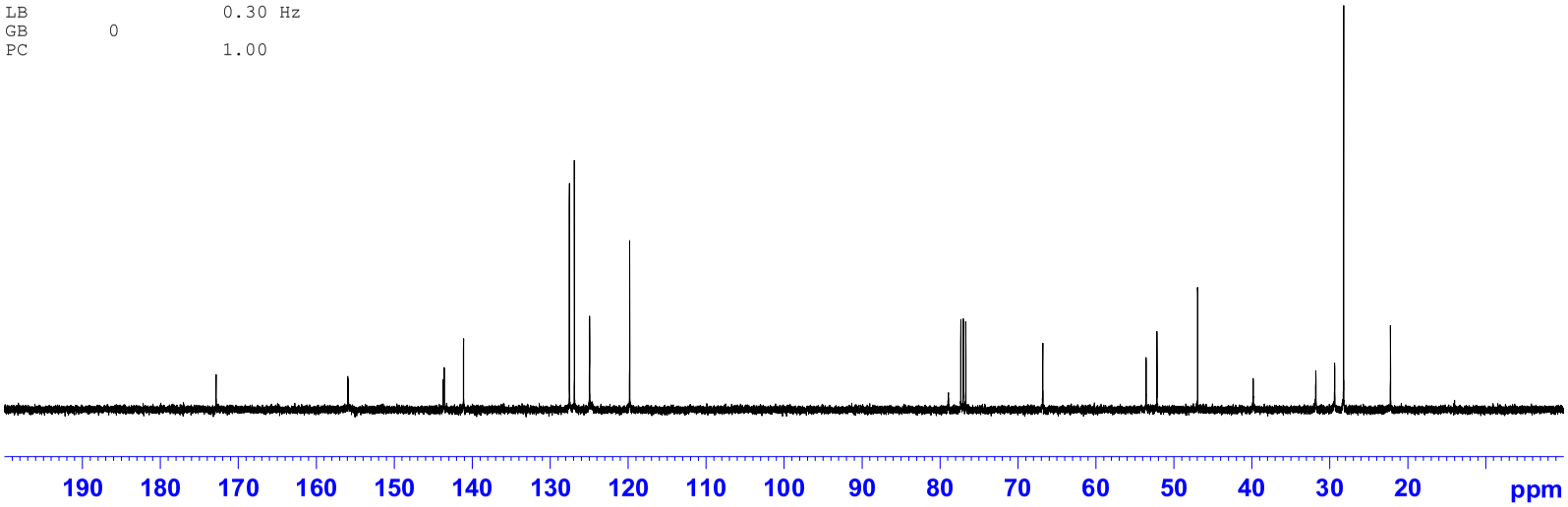
172.834
155.951
155.897
143.737
143.582
141.103
127.524
126.888
124.935
119.793
77.315
76.996
76.676
66.795
53.585
52.193
46.978
39.846
31.817
29.398
28.246
22.243

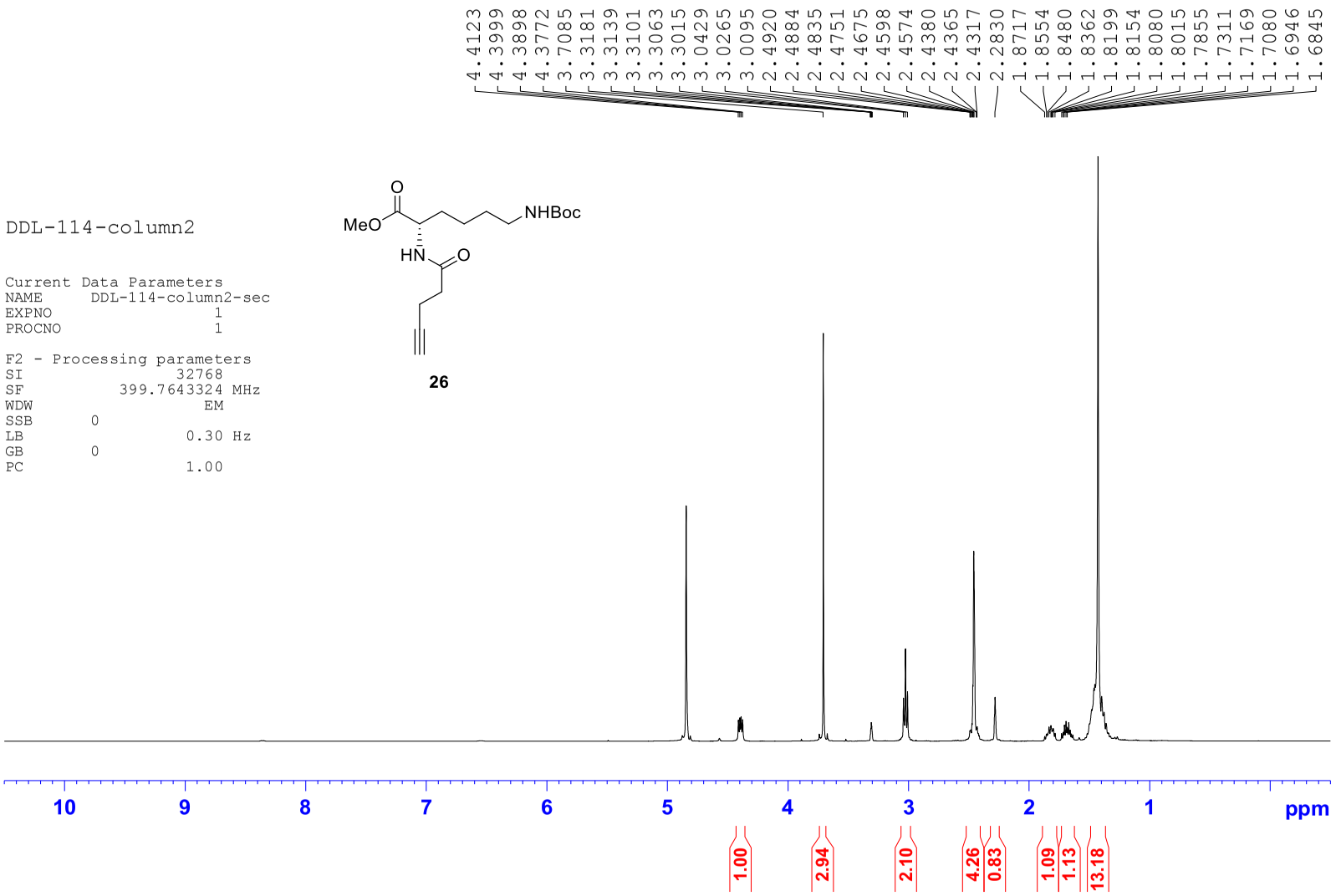
DDL-112-C13

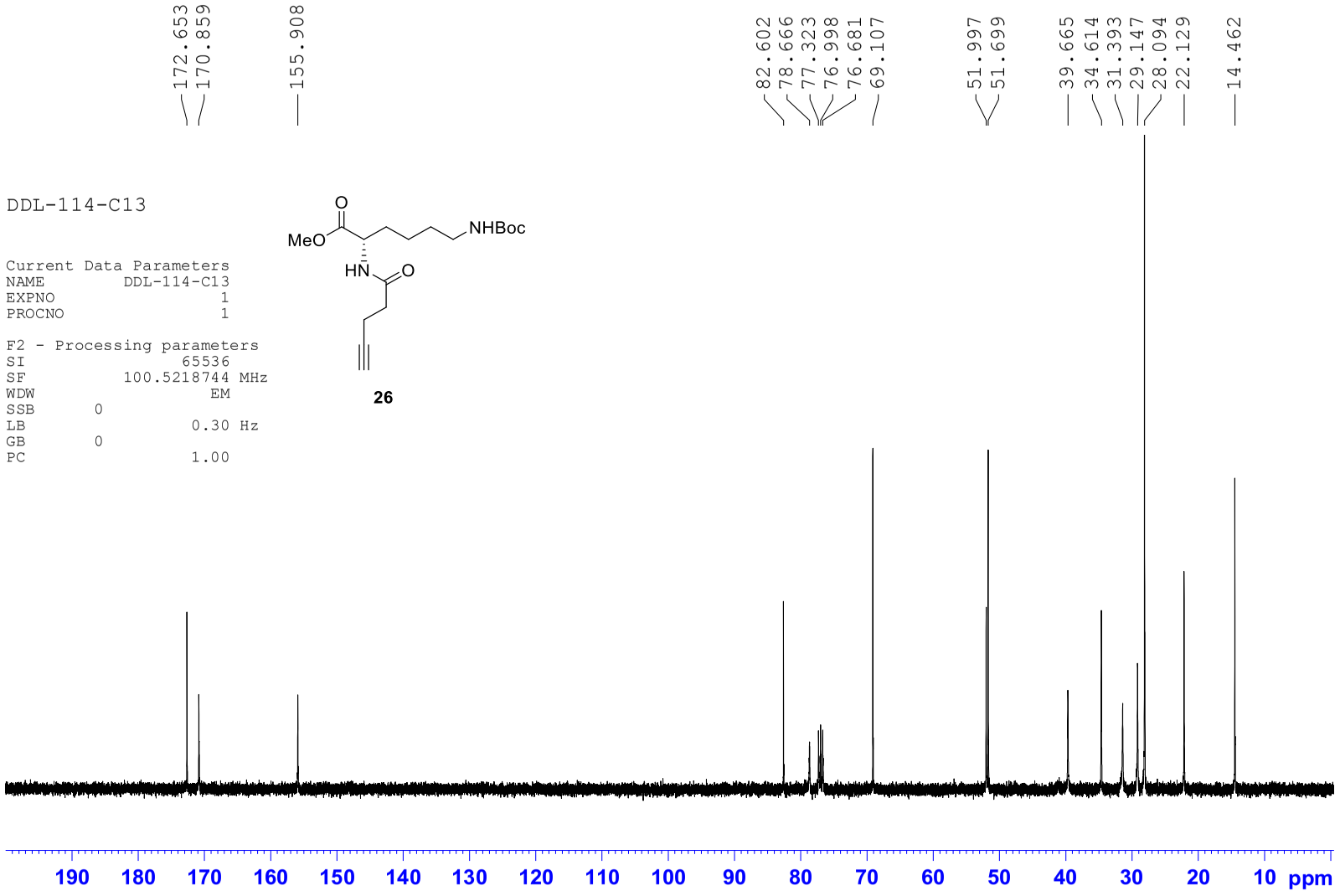


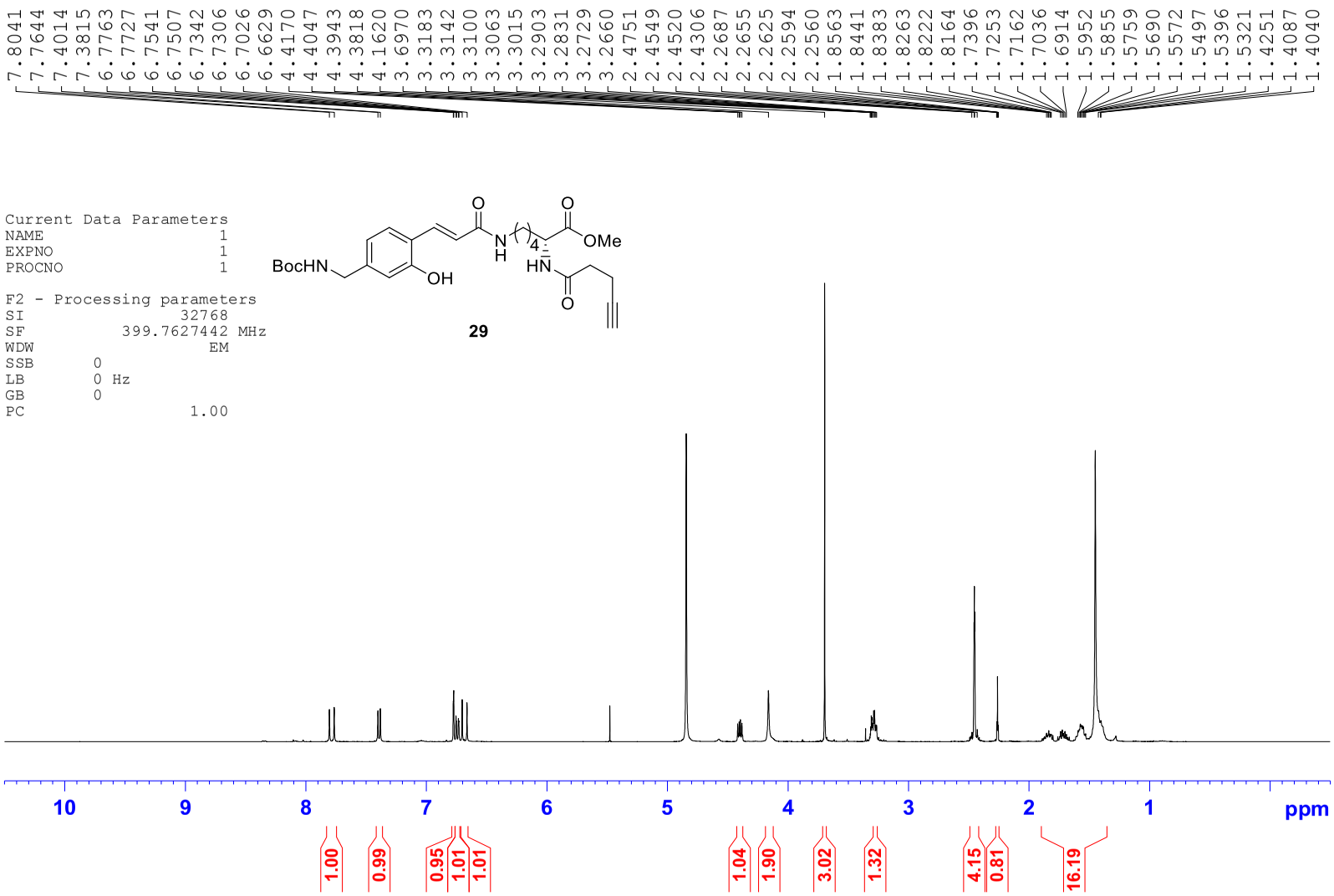

**24**

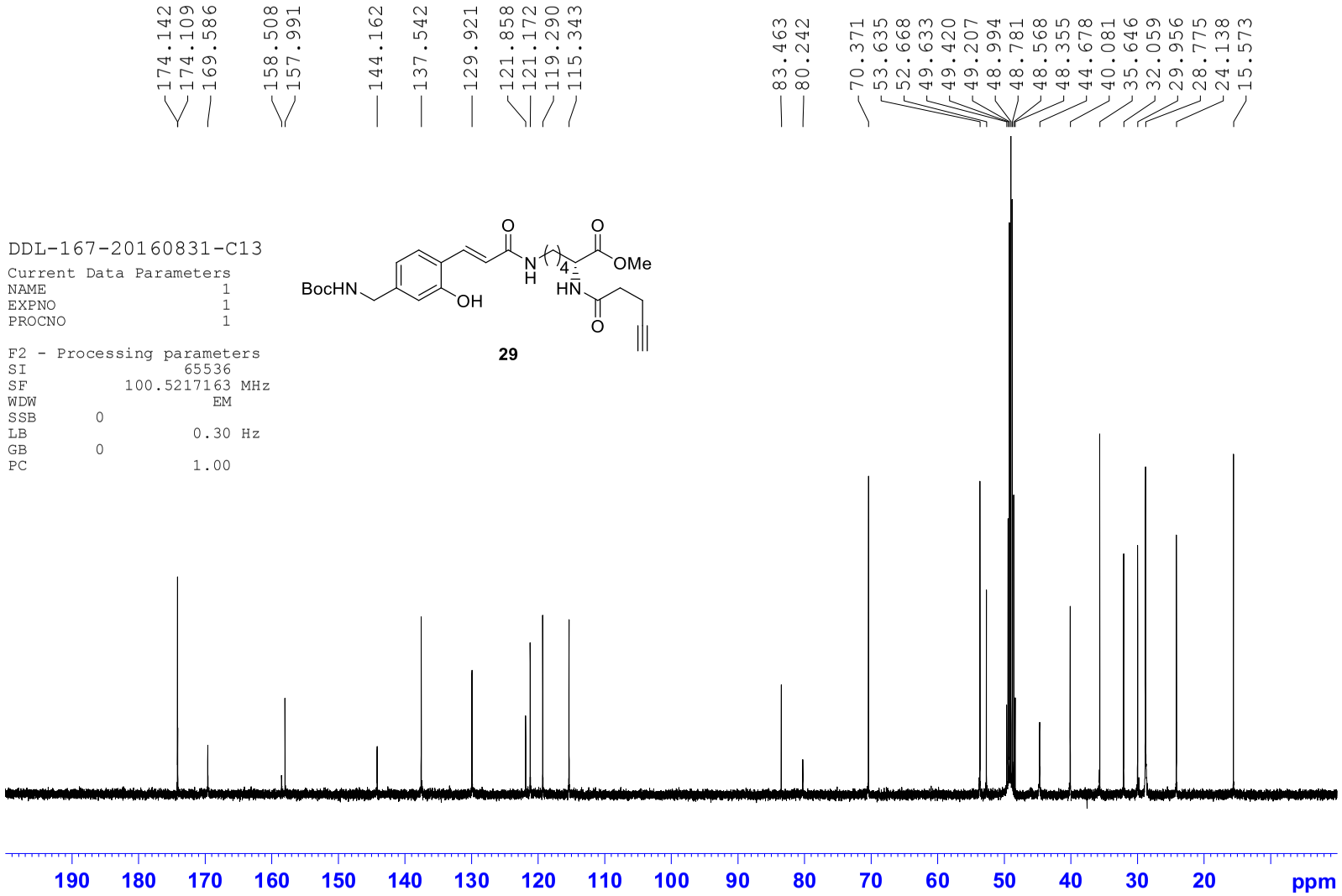
```
Current Data Parameters
NAME       DDL-112-2-C13
EXPNO                  1
PROCNO                 1

F2 - Processing parameters
SI                 65536
SF          100.5218706 MHz
WDW                   EM
SSB       0
LB                 0.30 Hz
GB        0
PC                 1.00
```

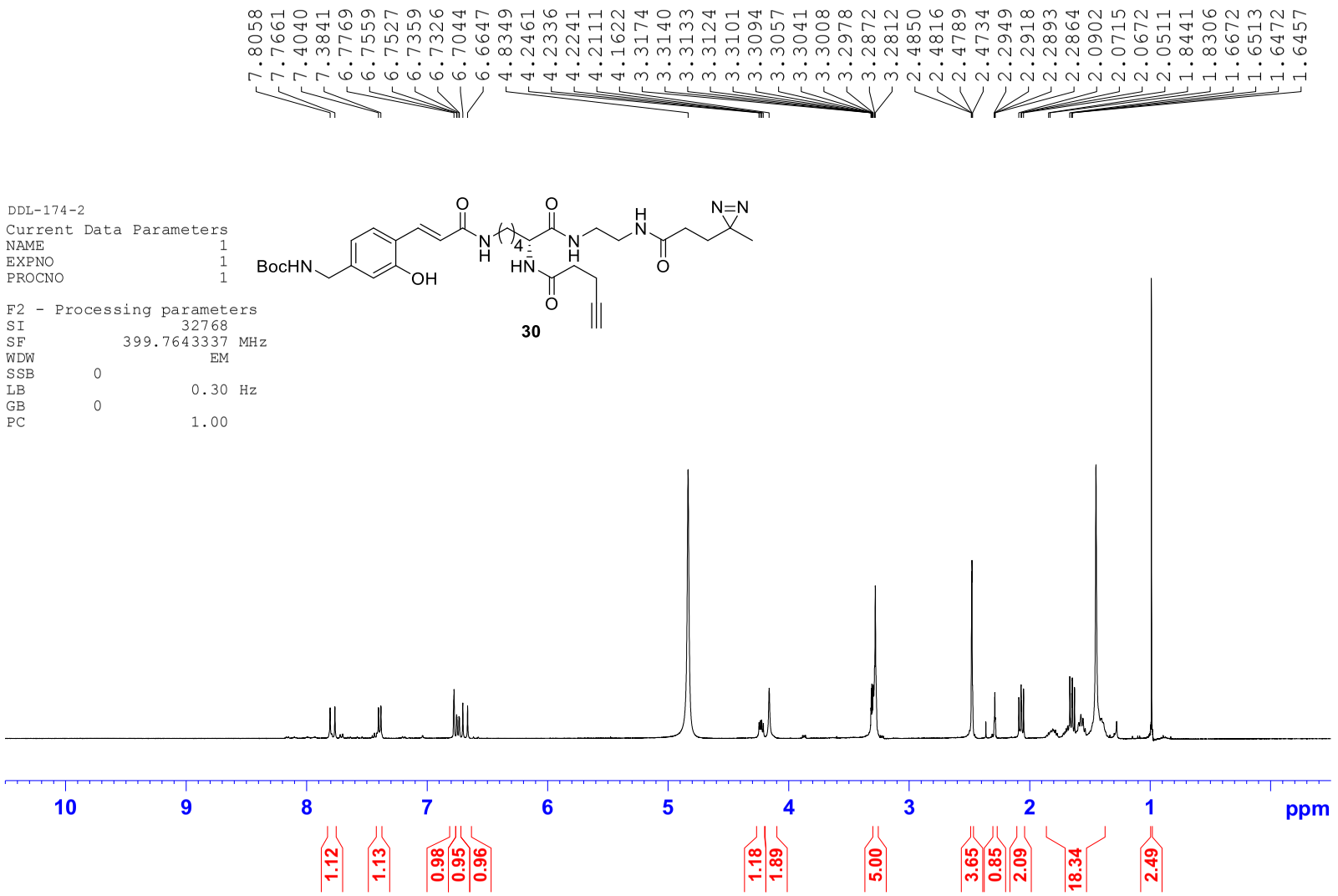


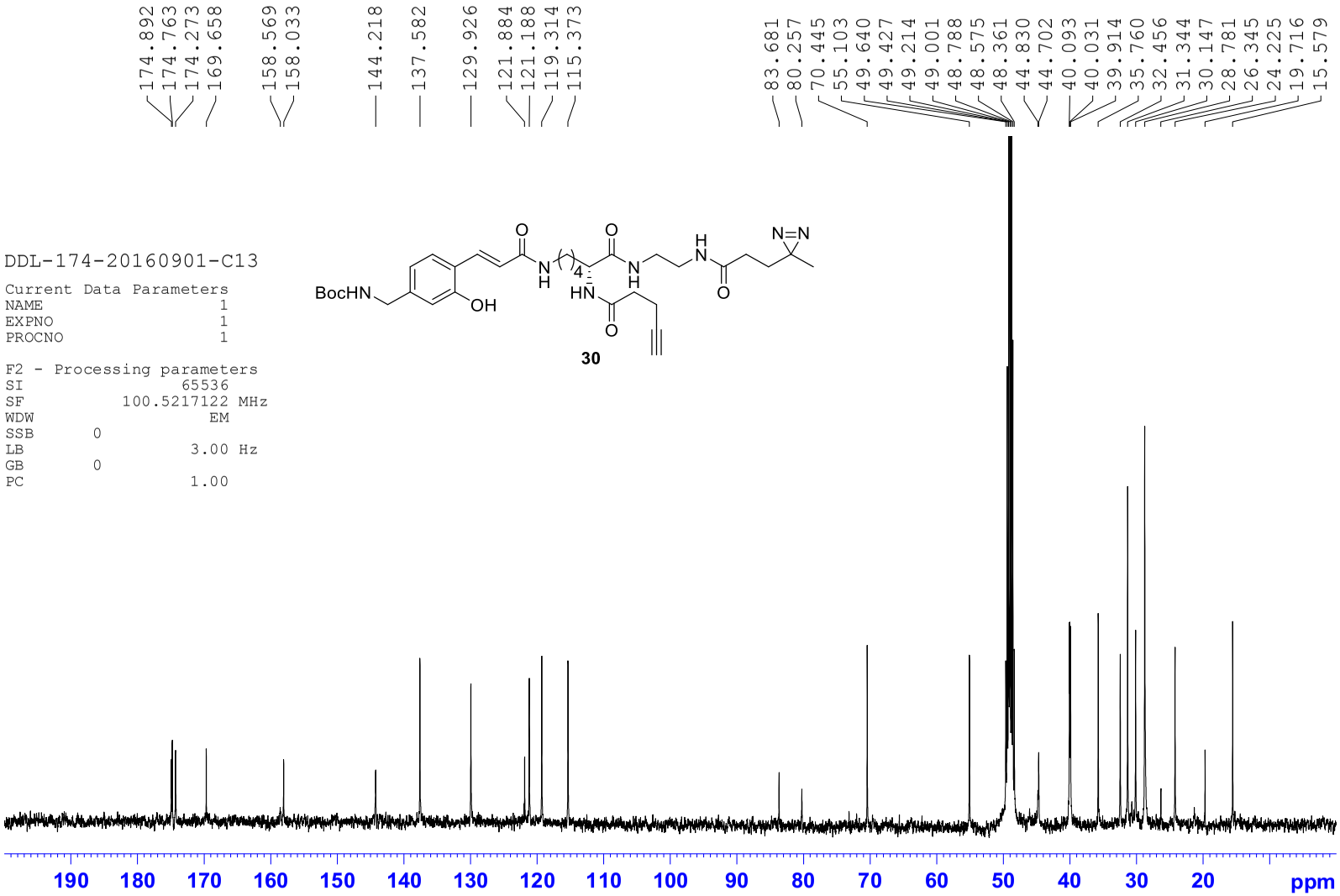

4.4123
4.3999
4.3898
4.3772
3.7085
3.3181
3.3139
3.3101
3.3063
3.3015
3.0429
3.0265
3.0095
2.4920
2.4884
2.4835
2.4751
2.4675
2.4598
2.4574
2.4380
2.4365
2.4317
2.2830
1.8717
1.8554
1.8480
1.8362
1.8199
1.8154
1.8080
1.8015
1.7855
1.7311
1.7169
1.7080
1.6946
1.6845
DDL-114-column2
Current Data Parameters
NAME DDL-114-column2-sec
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7643324 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
MeO
HN
O
NHBoc
26
10
9
8
7
6
5
4
3
2
1
ppm
1.00
2.94
2.10
4.26
0.83
1.09
1.13
13.18

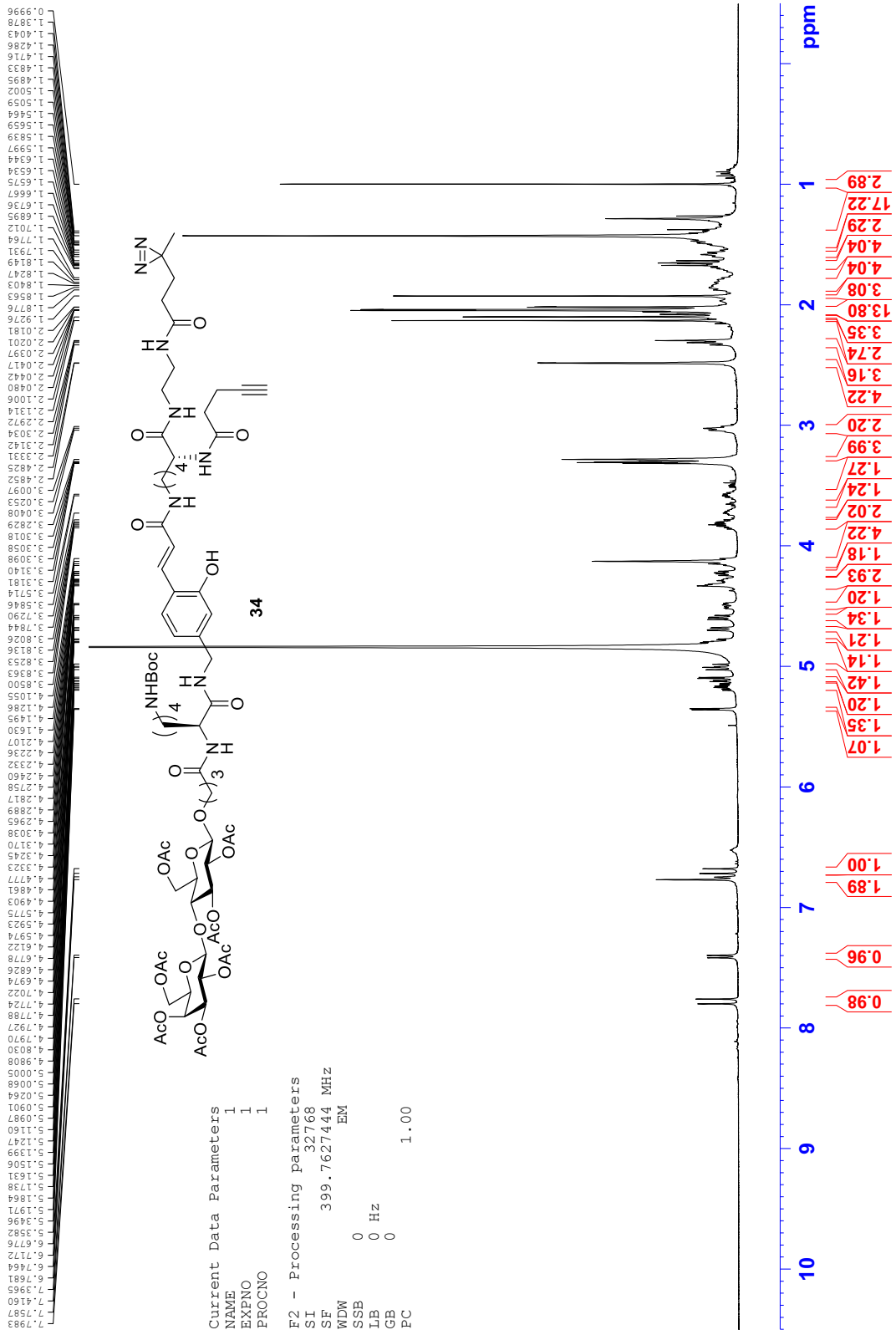

172.653
170.859
155.908
82.602
78.666
77.323
76.998
76.681
69.107
51.997
51.699
39.665
34.614
31.393
29.147
28.094
22.129
14.462
DDL-114-C13
Current Data Parameters
NAME DDL-114-C13
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5218744 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
MeO
NHBoc
HN
O
26
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 ppm

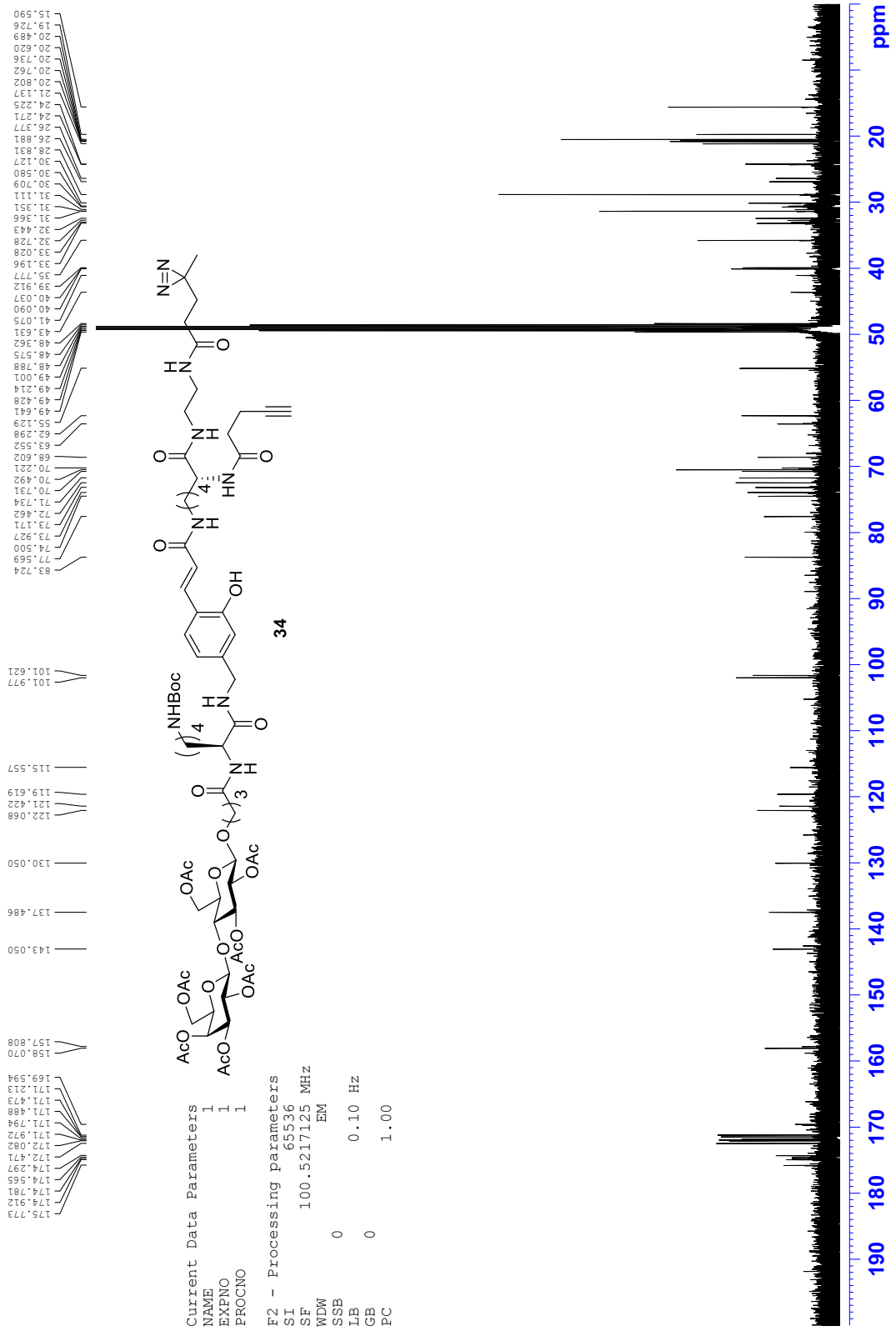

7.8041
7.7644
7.4014
7.3815
6.7763
6.7727
6.7541
6.7507
6.7342
6.7306
6.7026
6.6629
4.4170
4.4047
4.3943
4.3818
4.1620
3.6970
3.3183
3.3142
3.3100
3.3063
3.3015
3.2903
3.2831
3.2729
3.2660
2.4751
2.4549
2.4520
2.4306
2.2687
2.2655
2.2625
2.2594
2.2560
1.8563
1.8441
1.8383
1.8263
1.8222
1.8164
1.7396
1.7253
1.7162
1.7036
1.6914
1.5952
1.5855
1.5759
1.5690
1.5572
1.5497
1.5396
1.5321
1.4251
1.4087
1.4040
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627442 MHz
WDW EM
SSB 0
LB 0 Hz
GB 0
PC 1.00
BocHN
OH
N
H
HN
OMe
O
29
10
9
8
7
6
5
4
3
2
1
ppm
1.00
0.99
0.95
1.01
1.01
1.04
1.90
3.02
1.32
4.15
0.81
16.19

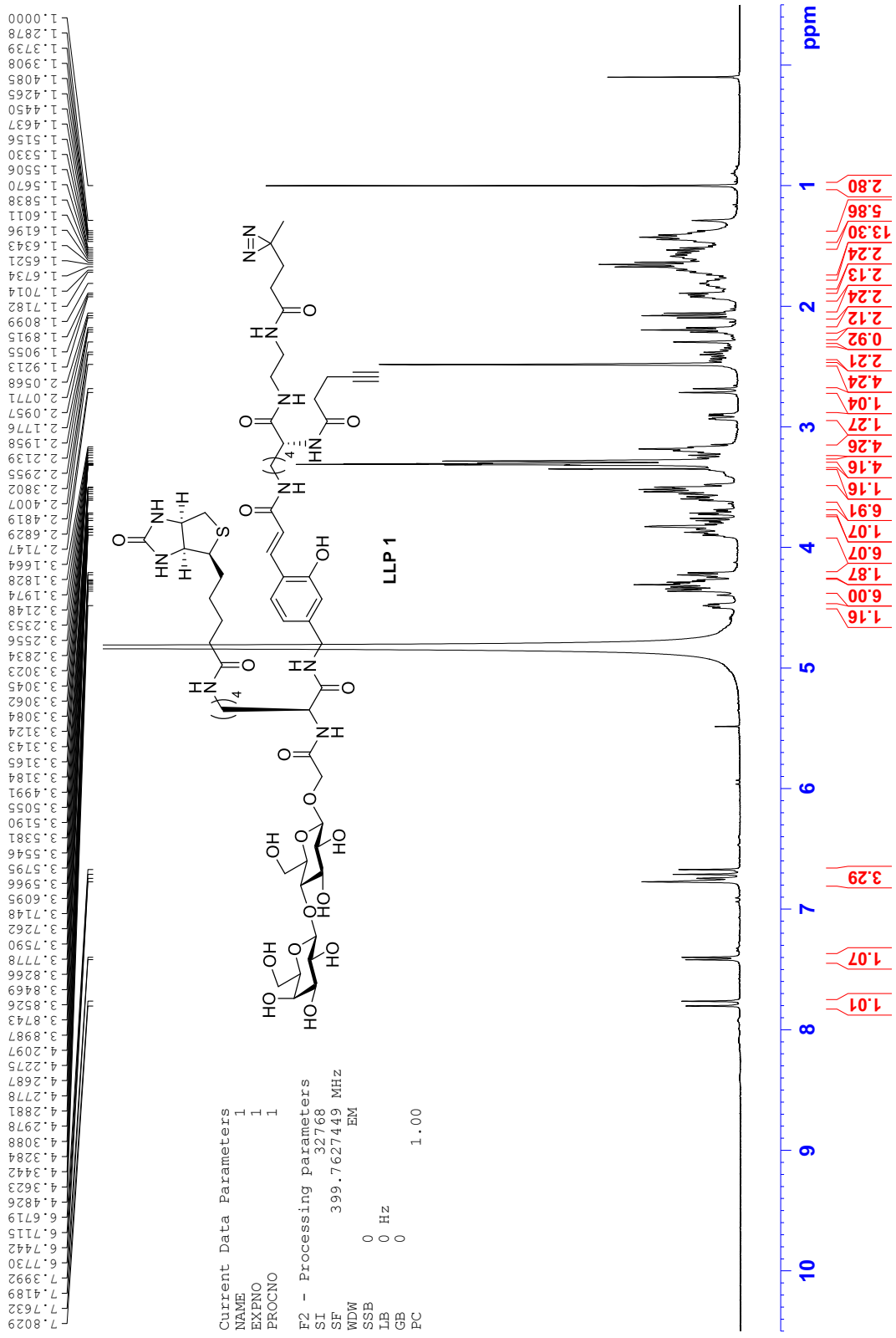

174.142
174.109
169.586
158.508
157.991
144.162
137.542
129.921
121.858
121.172
119.290
115.343
83.463
80.242
70.371
53.635
52.668
49.633
49.420
49.207
48.994
48.781
48.568
48.355
44.678
40.081
35.646
32.059
29.956
28.775
24.138
15.573
DDL-167-20160831-C13
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217163 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
BocHN
OH
N
H
OMe
HN
O
29
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

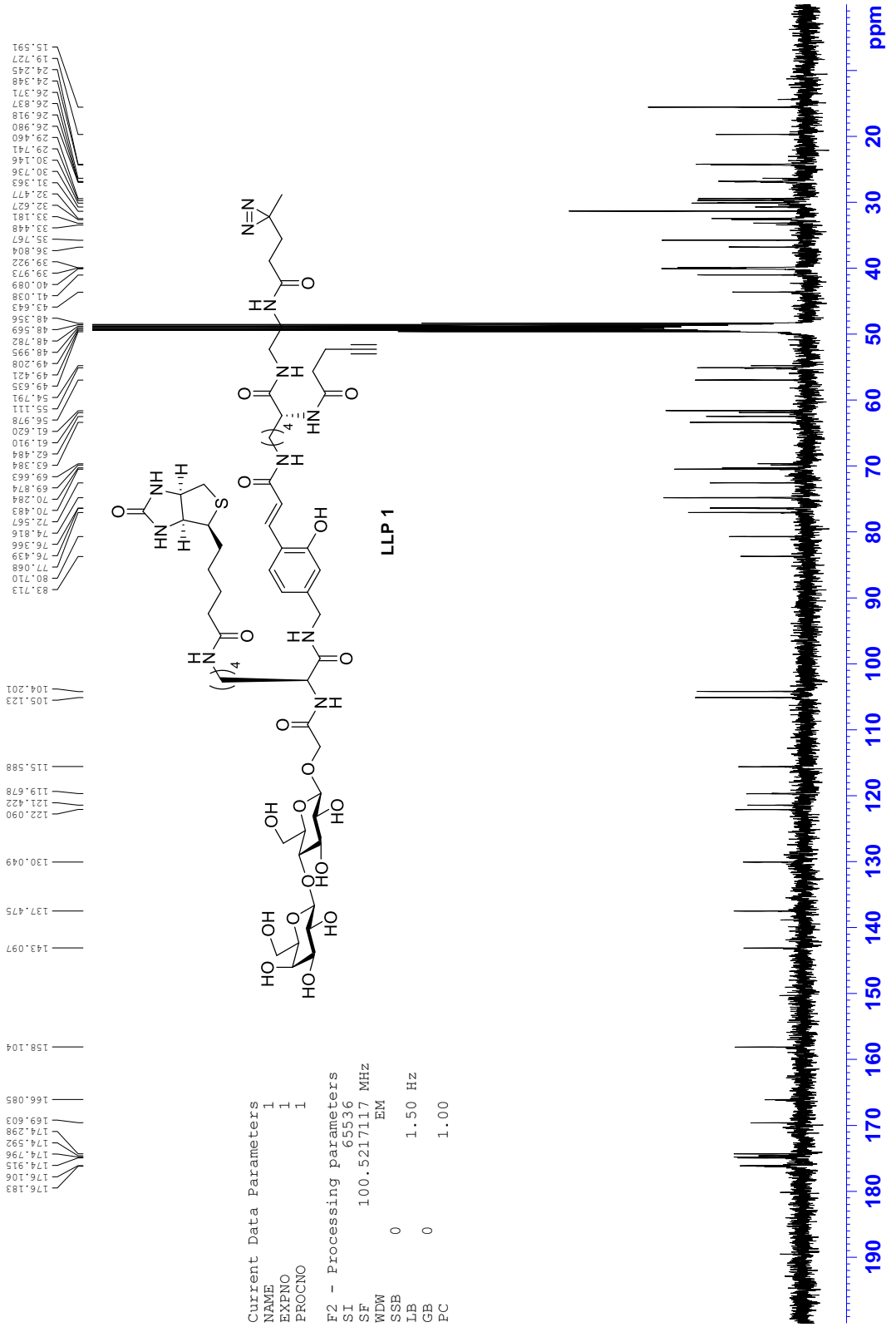

7.8058
7.7661
7.4040
7.3841
6.7769
6.7559
6.7527
6.7359
6.7326
6.7044
6.6647
4.8349
4.2461
4.2336
4.2241
4.2111
4.1622
3.3174
3.3140
3.3133
3.3124
3.3101
3.3094
3.3057
3.3041
3.3008
3.2978
3.2872
3.2812
2.4850
2.4816
2.4789
2.4734
2.2949
2.2918
2.2893
2.2864
2.0902
2.0715
2.0672
2.0511
1.8441
1.8306
1.6672
1.6513
1.6472
1.6457
DDL-174-2
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7643337 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
BocHN
OH
HN
N=N
30
10
9
8
7
6
5
4
3
2
1
ppm
1.12
1.13
0.98
0.95
0.96
1.18
1.89
5.00
3.65
0.85
2.09
18.34
2.49

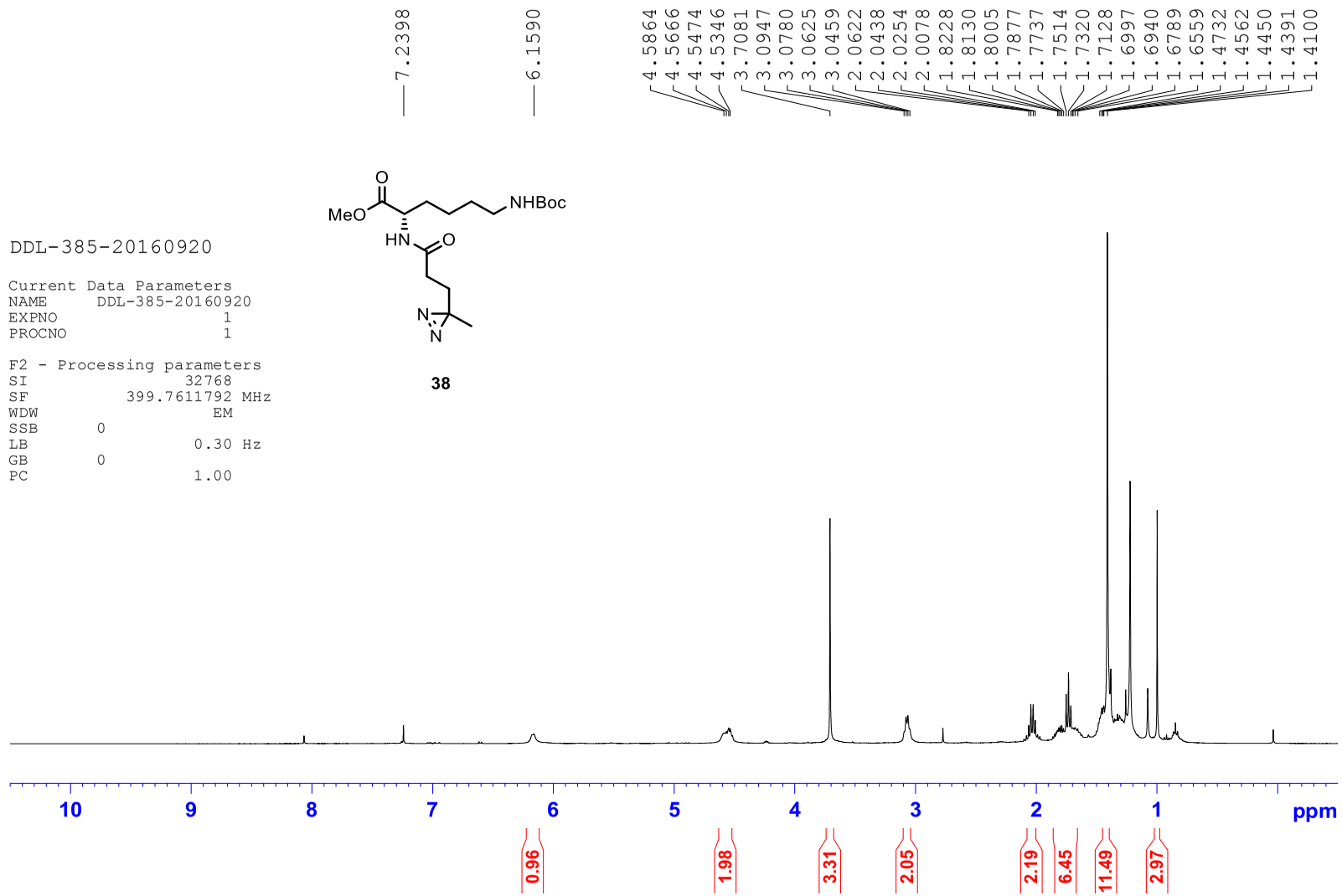

174.892
174.763
174.273
169.658
158.569
158.033
144.218
137.582
129.926
121.884
121.188
119.314
115.373
83.681
80.257
70.445
55.103
49.640
49.427
49.214
49.001
48.788
48.575
48.361
44.830
44.702
40.093
40.031
39.914
35.760
32.456
31.344
30.147
28.781
26.345
24.225
19.716
15.579
DDL-174-20160901-C13
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217122 MHz
WDW EM
SSB 0
LB 3.00 Hz
GB 0
PC 1.00
BocHN
OH
HN
N=N
30
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

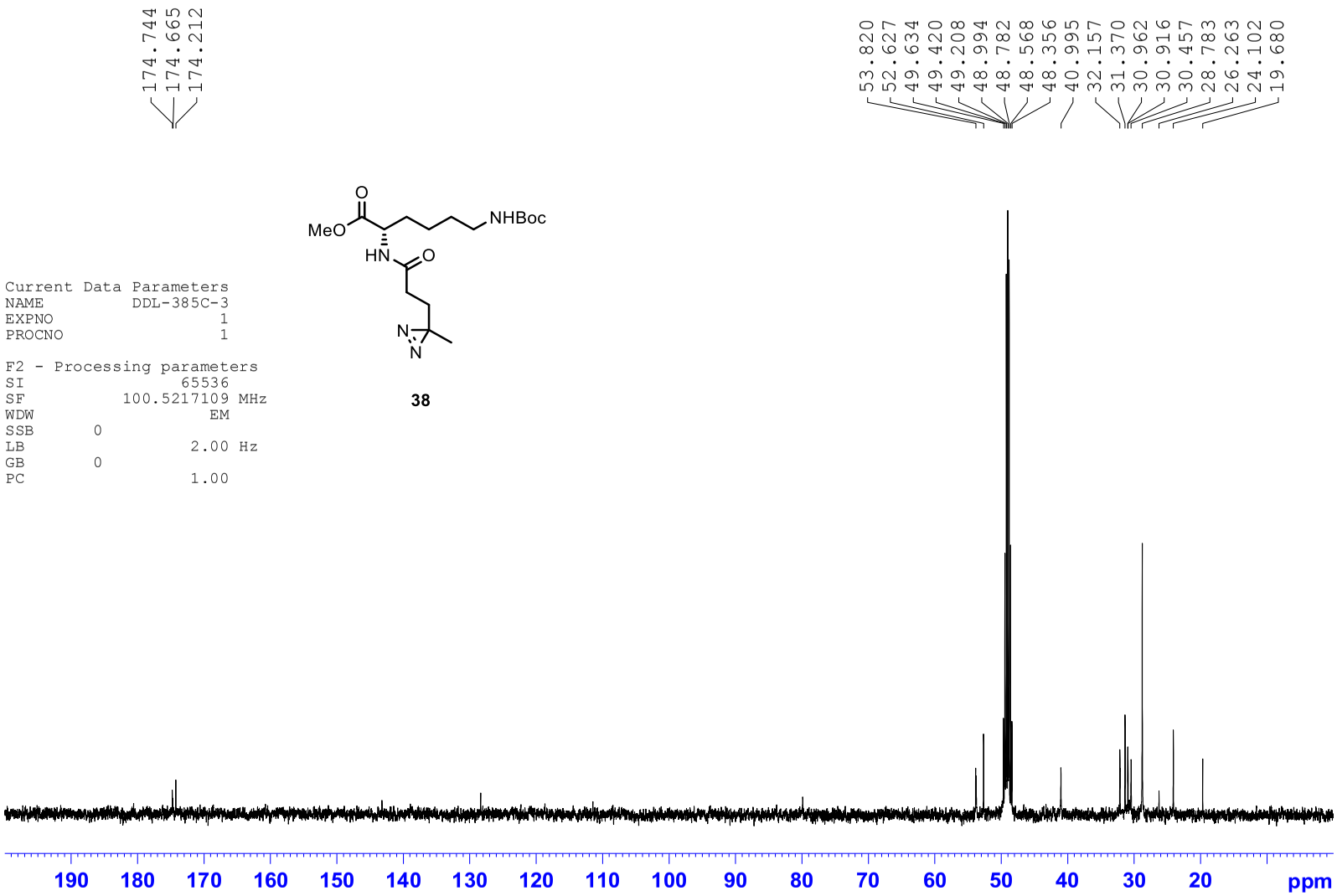

34
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627444 MHz
WDW EM
SSB 0
LB 0 Hz
GB 0
PC 1.00
ppm
1.07
1.35
1.20
1.42
1.14
1.21
1.34
1.20
2.93
1.18
4.22
2.02
1.24
1.27
3.99
2.20
4.22
3.16
2.74
3.35
13.80
3.08
4.04
4.04
2.29
17.22
2.89
1.89
1.00
0.96
0.98

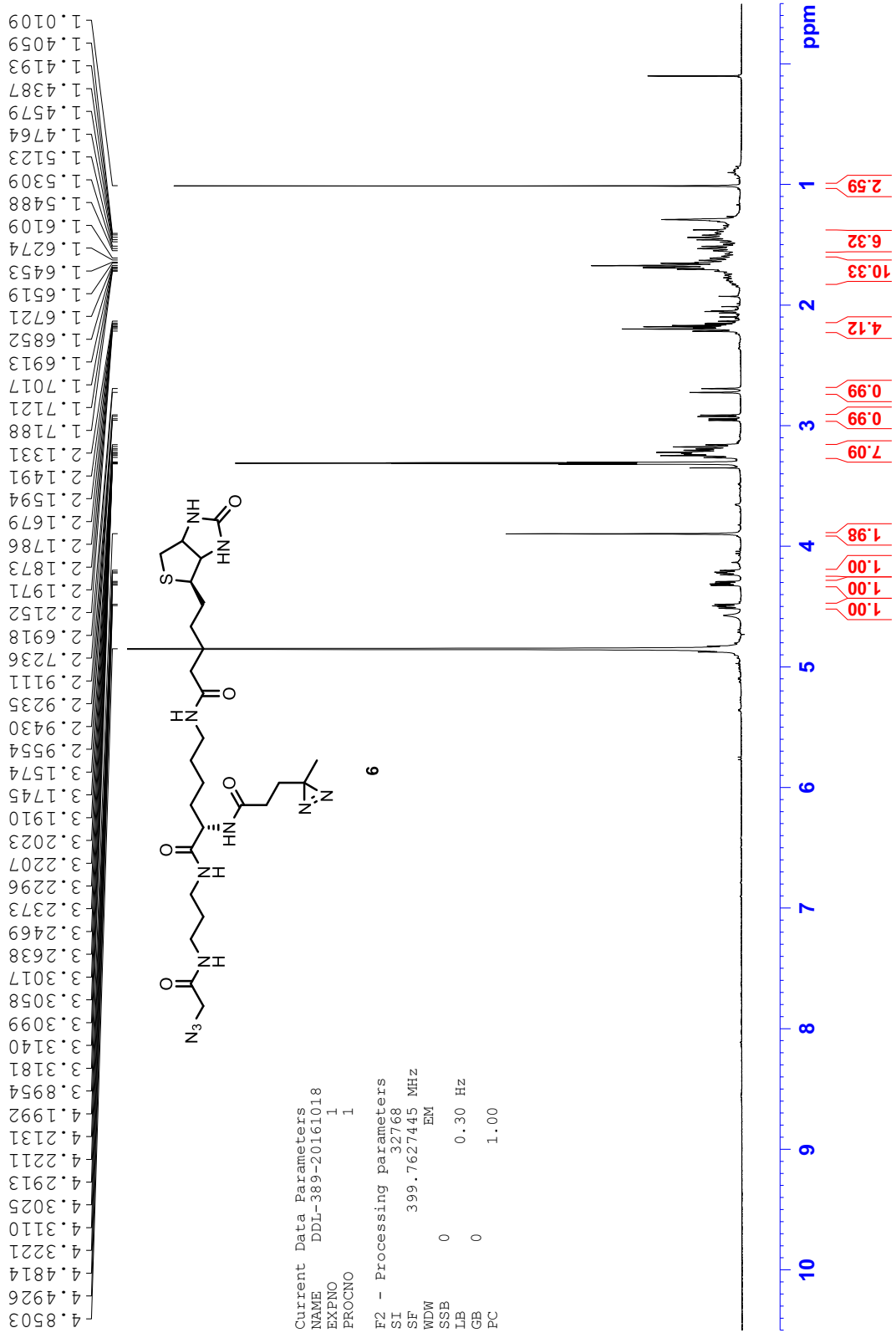

Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217125 MHz
WDW EM
SSB 0
LB 0.10 Hz
GB 0
PC 1.00
34
ppm

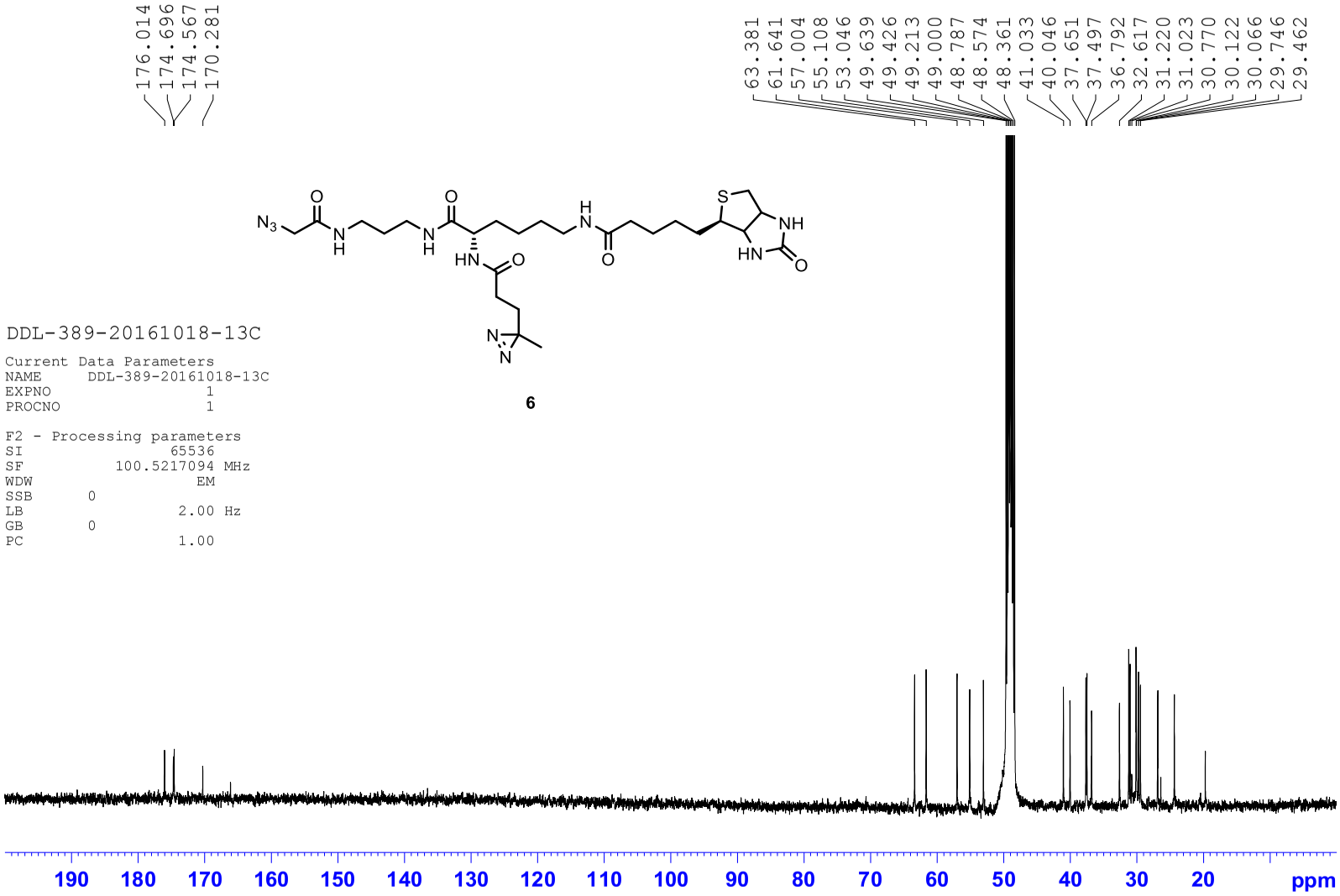

LLP 1
ppm
1 2 3 4 5 6 7 8 9 10
2.80
5.86
13.30
2.24
2.13
2.24
2.12
0.92
2.21
4.24
1.04
1.27
4.26
4.16
1.16
6.91
1.07
6.07
1.87
6.00
1.16
3.29
1.07
1.01
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627449 MHz
WDW EM
SSB 0
LB 0 Hz
GB 0
PC 1.00

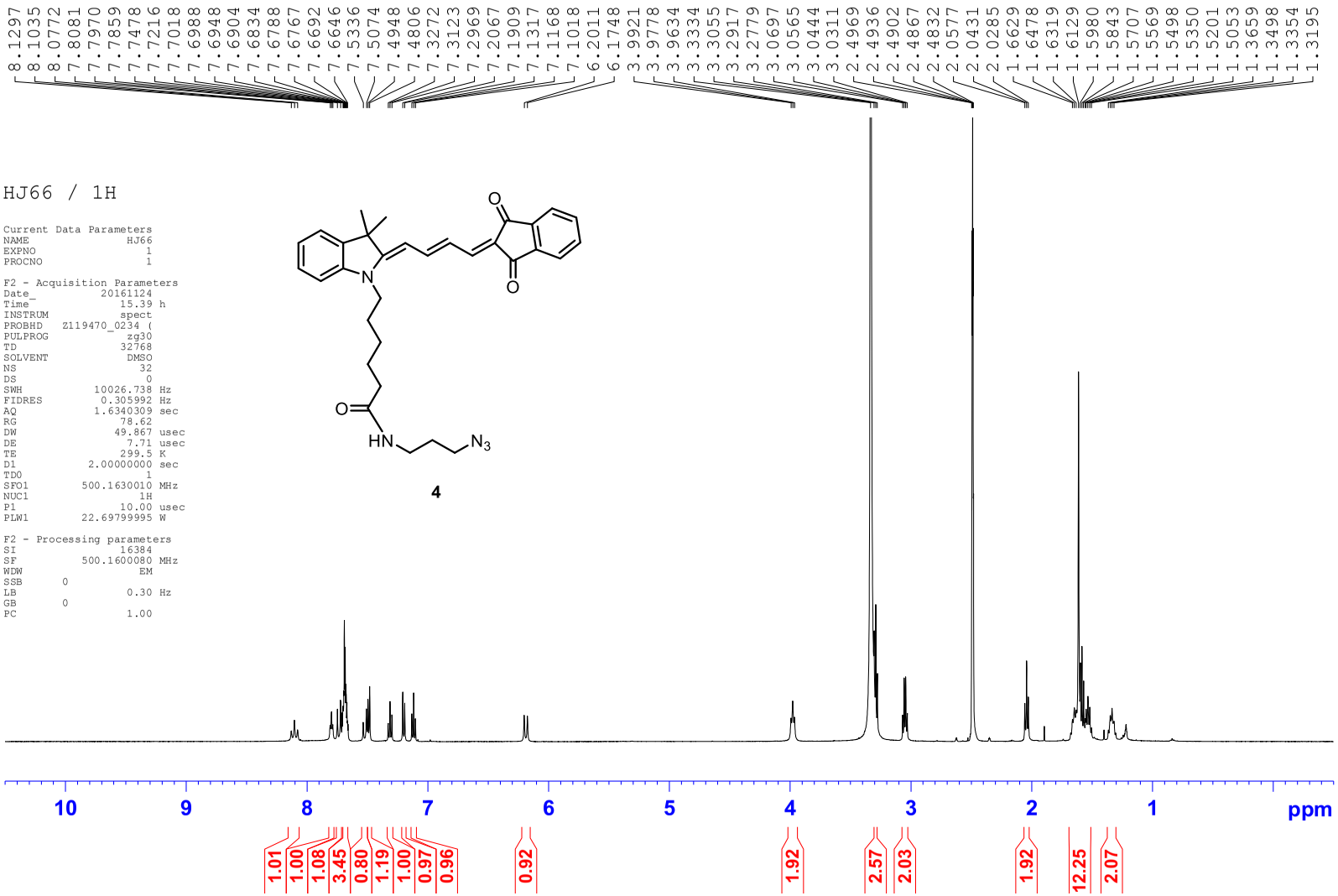

LLP 1
Current Data Parameters
NAME 1
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217117 MHz
WDW EM
SSB 0
LB 1.50 Hz
GB 0
PC 1.00
ppm

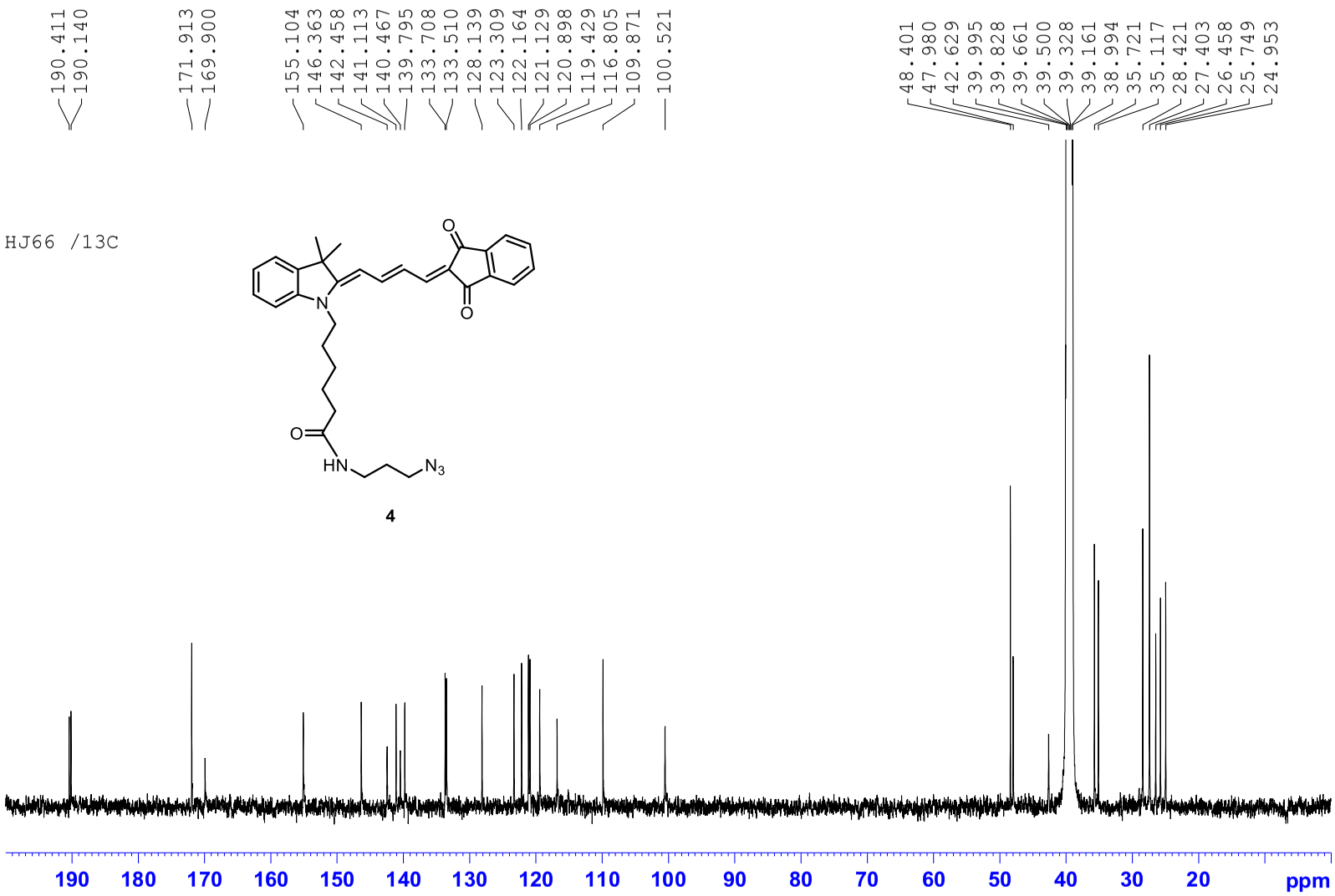

7.2398
6.1590
4.5864
4.5666
4.5474
4.5346
3.7081
3.0947
3.0780
3.0625
3.0459
2.0622
2.0438
2.0254
2.0078
1.8228
1.8130
1.8005
1.7877
1.7737
1.7514
1.7320
1.7128
1.6997
1.6940
1.6789
1.6559
1.4732
1.4562
1.4450
1.4391
1.4100
MeO
O
HN
O
NHBoc
N
N
38
DDL-385-20160920
Current Data Parameters
NAME DDL-385-20160920
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7611792 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
10
9
8
7
6
5
4
3
2
1
ppm
0.96
1.98
3.31
2.05
2.19
6.45
11.49
2.97

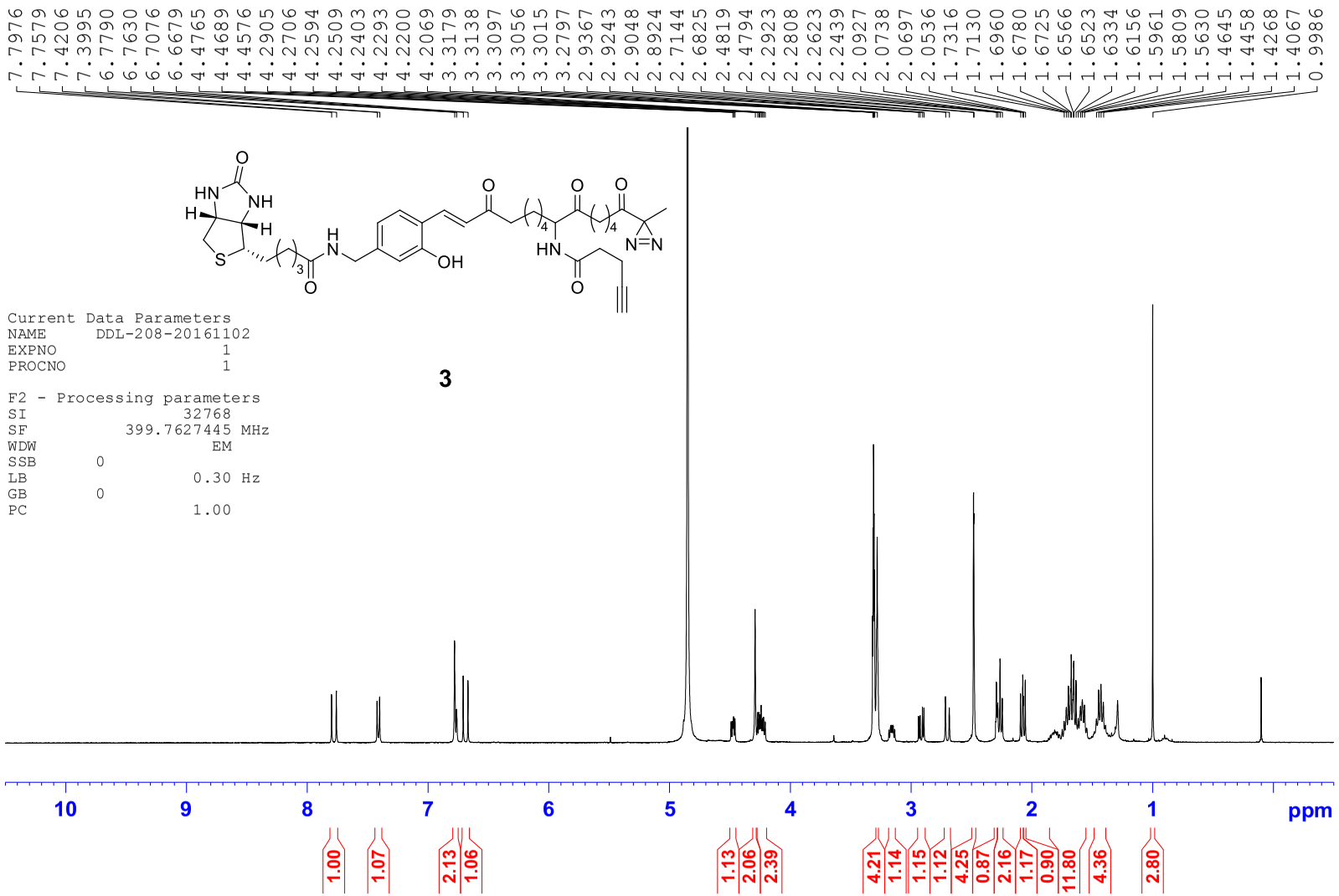

174.744
174.665
174.212
53.820
52.627
49.634
49.420
49.208
48.994
48.782
48.568
48.356
40.995
32.157
31.370
30.962
30.916
30.457
28.783
26.263
24.102
19.680
MeO
O
HN
O
NHBoc
N
N
38
Current Data Parameters
NAME DDL-385C-3
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217109 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 ppm

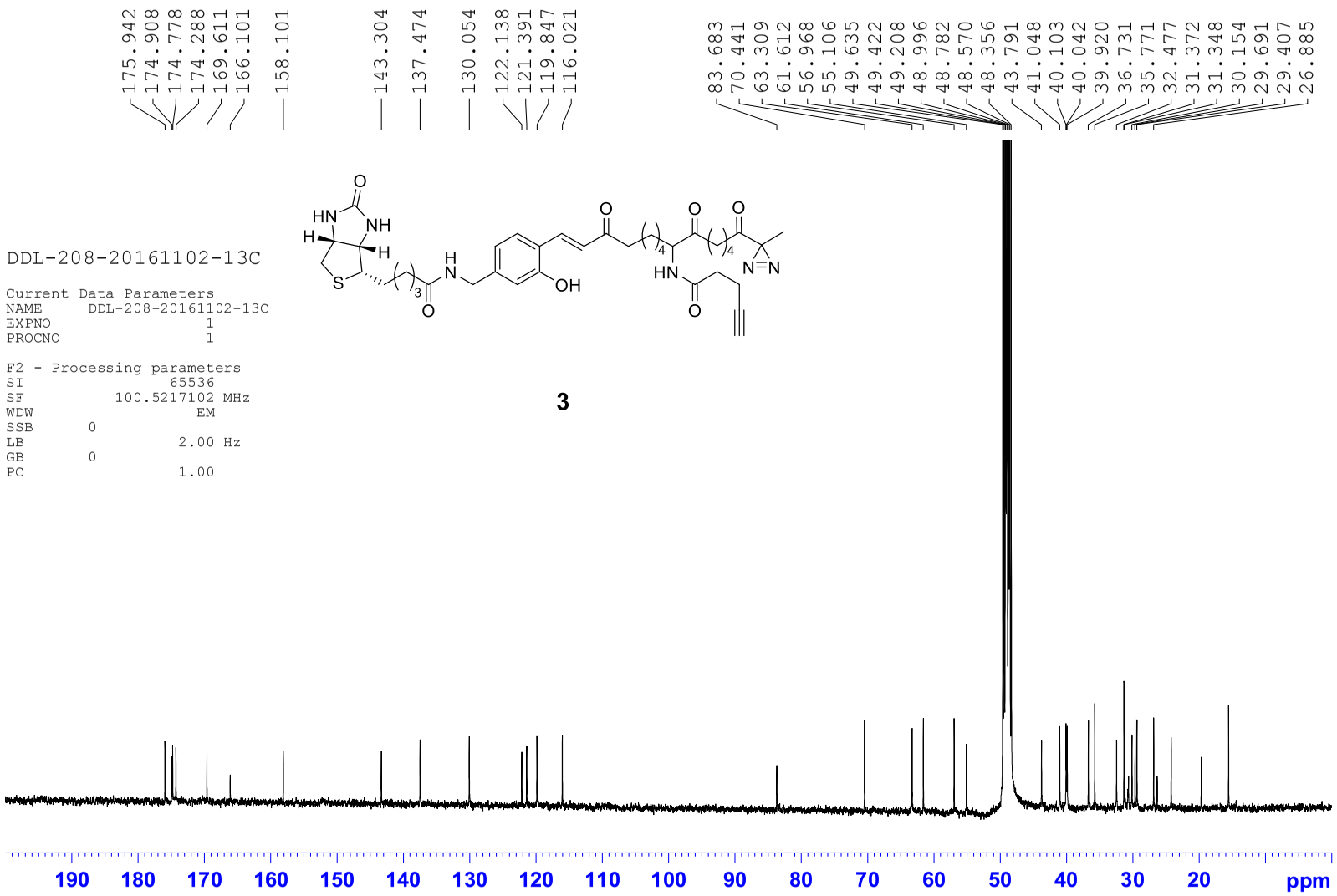

Current Data Parameters
NAME DDL-389-20161018
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627445 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
6
ppm
2.59
6.32
10.33
4.12
0.99
0.99
7.09
1.98
1.00
1.00
1.00

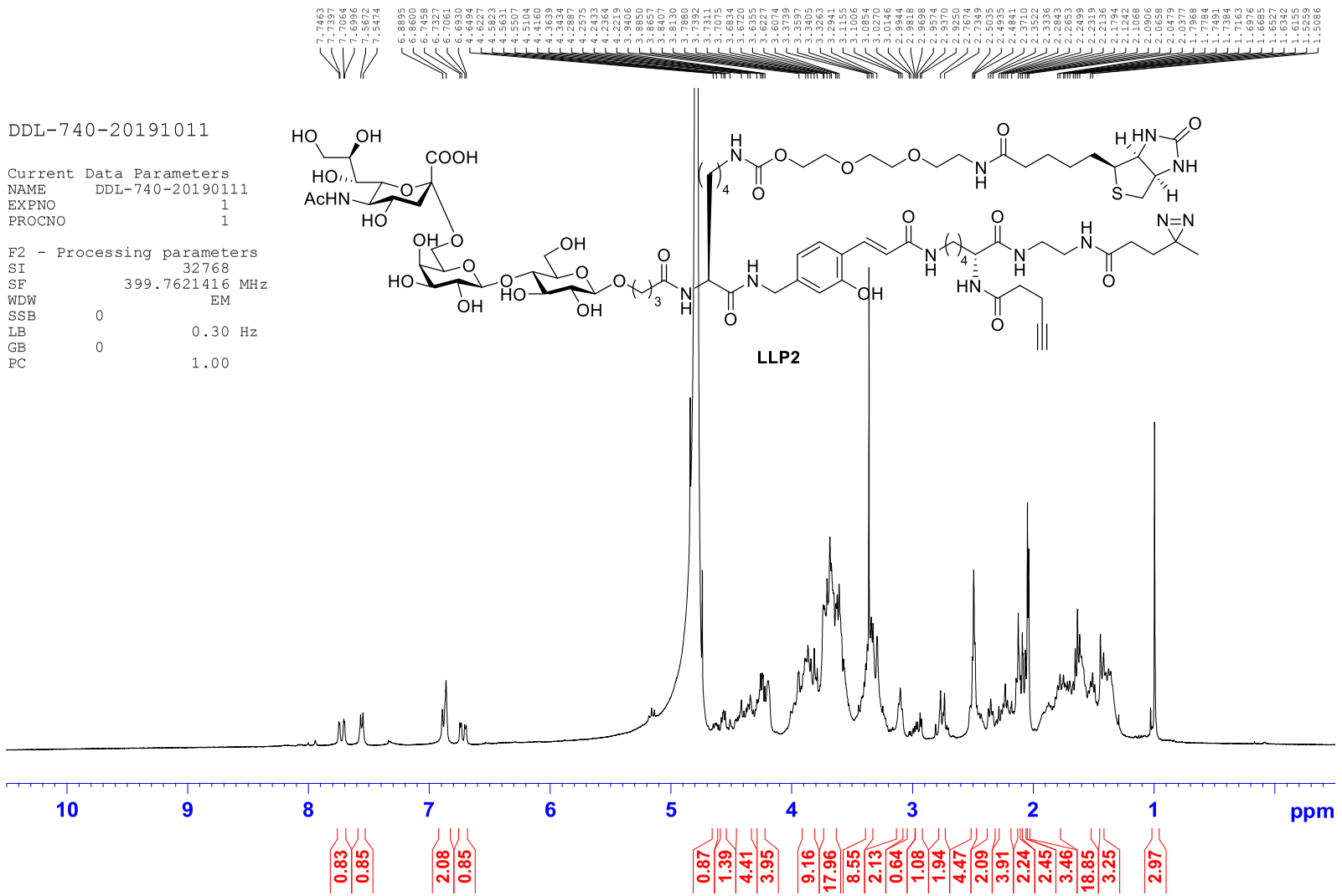

176.014
174.696
174.567
170.281
63.381
61.641
57.004
55.108
53.046
49.639
49.426
49.213
49.000
48.787
48.574
48.361
41.033
40.046
37.651
37.497
36.792
32.617
31.220
31.023
30.770
30.122
30.066
29.746
29.462
DDL-389-20161018-13C
Current Data Parameters
NAME DDL-389-20161018-13C
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217094 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00
6
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

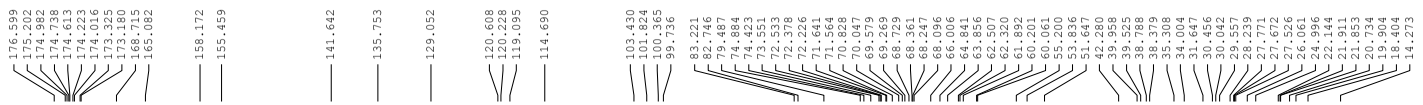

8.1297
8.1035
8.0772
7.8081
7.7970
7.7859
7.7478
7.7216
7.7018
7.6988
7.6948
7.6904
7.6834
7.6788
7.6767
7.6692
7.6646
7.5336
7.5074
7.4948
7.4806
7.3272
7.3123
7.2969
7.2067
7.1909
7.1317
7.1168
7.1018
6.2011
6.1748
3.9921
3.9778
3.9634
3.3334
3.3055
3.2917
3.2779
3.0697
3.0565
3.0444
3.0311
2.4969
2.4936
2.4902
2.4867
2.4832
2.0577
2.0431
2.0285
1.6629
1.6478
1.6319
1.6129
1.5980
1.5843
1.5707
1.5569
1.5498
1.5350
1.5201
1.5053
1.3659
1.3498
1.3354
1.3195
HJ66 / 1H
Current Data Parameters
NAME HJ66
EXPNO 1
PROCNO 1
F2 - Acquisition Parameters
Date_ 20161124
Time 15.39 h
INSTRUM spect
PROBHD Z119470_0234 (
PULPROG zg30
TD 32768
SOLVENT DMSO
NS 32
DS 0
SWH 10026.738 Hz
FIDRES 0.305992 Hz
AQ 1.6340309 sec
RG 78.62
DW 49.867 usec
DE 7.71 usec
TE 299.5 K
D1 2.00000000 sec
TD0 1
SFO1 500.1630010 MHz
NUC1 1H
P1 10.00 usec
PLW1 22.69799995 W
F2 - Processing parameters
SI 16384
SF 500.1600080 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
O
O
N
O
HN
N3
4
10
9
8
7
6
5
4
3
2
1
ppm
1.01
1.00
1.08
3.45
0.80
1.19
1.00
0.97
0.96
0.92
1.92
2.57
2.03
1.92
12.25
2.07


190.411
190.140
171.913
169.900
155.104
146.363
142.458
141.113
140.467
139.795
133.708
133.510
128.139
123.309
122.164
121.129
120.898
119.429
116.805
109.871
100.521
48.401
47.980
42.629
39.995
39.828
39.661
39.500
39.328
39.161
38.994
35.721
35.117
28.421
27.403
26.458
25.749
24.953
HJ66 /13C
O
O
N
O
HN
N3
4
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm


7.7976
7.7579
7.4206
7.3995
6.7790
6.7630
6.7076
6.6679
4.4765
4.4689
4.4576
4.2905
4.2706
4.2594
4.2509
4.2403
4.2293
4.2200
4.2069
3.3179
3.3138
3.3097
3.3056
3.3015
3.2797
2.9367
2.9243
2.9048
2.8924
2.7144
2.6825
2.4819
2.4794
2.2923
2.2808
2.2623
2.2439
2.0927
2.0738
2.0697
2.0536
1.7316
1.7130
1.6960
1.6780
1.6725
1.6566
1.6523
1.6334
1.6156
1.5961
1.5809
1.5630
1.4645
1.4458
1.4268
1.4067
0.9986
Current Data Parameters
NAME DDL-208-20161102
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7627445 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
3
10
9
8
7
6
5
4
3
2
1
ppm
1.00
1.07
2.13
1.06
1.13
2.06
2.39
4.21
1.14
1.15
1.12
4.25
0.87
2.16
1.17
0.90
11.80
4.36
2.80


175.942
174.908
174.778
174.288
169.611
166.101
158.101
143.304
137.474
130.054
122.138
121.391
119.847
116.021
83.683
70.441
63.309
61.612
56.968
55.106
49.635
49.422
49.208
48.996
48.782
48.570
48.356
43.791
41.048
40.103
40.042
39.920
36.731
35.771
32.477
31.372
31.348
30.154
29.691
29.407
26.885
DDL-208-20161102-13C
Current Data Parameters
NAME DDL-208-20161102-13C
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 65536
SF 100.5217102 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00
3
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

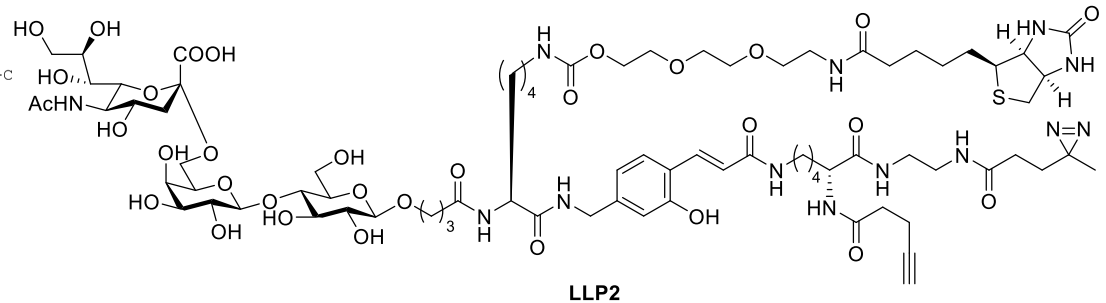

DDL-740-20191011
Current Data Parameters
NAME DDL-740-20190111
EXPNO 1
PROCNO 1
F2 - Processing parameters
SI 32768
SF 399.7621416 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00
LLP2
10
9
8
7
6
5
4
3
2
1
ppm
0.83
0.85
2.08
0.85
0.87
1.39
4.41
3.95
9.16
17.96
8.55
2.13
0.64
1.08
1.94
4.47
2.09
3.91
2.24
2.45
3.46
18.85
3.25
2.97




DDL-740-190109

Current Data Parameters
NAME CCLin-DDL-740-190109-C
EXPNO 1
PROCNO 1

F2 - Processing parameters
SI 131072
SF 175.9536856 MHz
WDW EM
SSB 0
LB 3.00 Hz
GB 0
PC 1.00



**LLP2**