Supporting Information

Biologics through Chemistry: Total Synthesis of a Proposed Dual Acting Vaccine Targeting Ovarian Cancer by Orchestration of Oligosaccharide and Polypeptide Domains

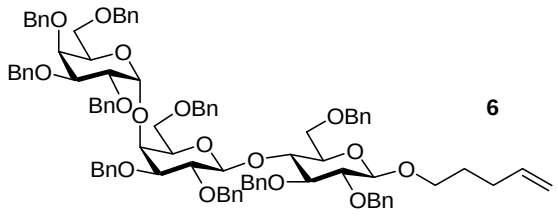
Jianglong Zhu,[1] Qian Wan,[1] Govind Ragupathi,[2] Constantine M. George,[2] Philip O. Livingston,[2] and Samuel J. Danishefsky[,1,3]*

[1]Laboratory for Bioorganic Chemistry, [2]Laboratory of Tumor Vaccinology, Clinical Immunology Service, Department of Medicine, Sloan-Kettering Institute for Cancer Research, 1275 York Avenue, New York, New York 10065, and [3]Department of Chemistry, Columbia University, Havemeyer Hall, 3000 Broadway, New York, New York 10027

s-danishefsky@ski.mskcc.org

Materials and Methods

All commercial materials (Aldrich, Fluka) were used without further purification. All solvents were reagent grade or HPLC grade (Fisher). Anhydrous THF, diethyl ether, CH_2Cl_2, toluene, and benzene were obtained from a dry solvent system (passed through column of alumina) and used without further drying. All reactions were performed under an atmosphere of pre-purified dry Ar(g). ^{1}H NMR spectra and ^{13}C NMR spectra were recorded on a Bruker Advance DRX-500 MHz at ambient temperature unless otherwise stated. Chemical shifts are reported in parts per million relative to residual solvent $CDCl_3$ (^{1}H, δ 7.24; ^{13}C, δ 77.0), CD_3OD (^{1}H, δ 3.31; ^{13}C, δ 49.15), D_2O (^{1}H, δ 4.80). Data for ^{1}H NMR are reported as follows: chemical shift, integration, multiplicity (app = apparent, par obsc = partially obscure, ovrlp = overlapping, s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet) and coupling constants. All ^{13}C NMR spectra were recorded with complete proton decoupling. Low-resolution mass spectral analyses were performed with a JOEL JMS-DX-303-HF mass spectrometer or Waters Micromass ZQ mass spectrometer. All reactions were carried out in oven-dried glassware under an argon atmosphere unless otherwise noted. Analytical TLC was performed on E. Merck silica gel 60 F254 plates and flash column chromatography was performed on E. Merck silica gel 60 (40–63 mm). Yields refer to chromatographically and spectroscopically pure compounds.

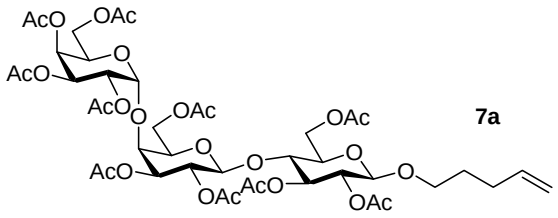


6

$C_{93}H_{100}O_{16}$
Exact Mass: 1472.7011
Mol. Wt.: 1473.7795

Perbenzylated trisaccharide 6. Prepared based on our previously reported glycosylation protocol with slight modification.[S1] A mixture of fluoro-donor **4** (618 mg, 1.14 mmol, 2.0 equiv) and lactoside acceptor **5** (540 mg, 0.57 mmol) was azeotroped with anhydrous benzene (3 X 10 mL) and further dried on high vacuum for 3 h. The above mixture was dissolved in toluene (5.0 mL) and THF (0.5 mL), and transferred *via* cannula to a flask containing 2,6-di-tert-butyl-4-methylpyridine (175 mg, 0.85 mmol) and freshly prepared 4 Å molecular sieves (900 mg) under argon. The flask was then cooled to -20°C and $Cp_2Zr(OTf)_2$ (336 mg, 0.57 mmol, 1.0 equiv) was quickly added to the reaction mixture. The reaction was slowly warmed and stirred for 72 h at 7°C under dark. The reaction mixture was diluted with EtOAc (15 mL) and filtered through a pad of anhydrous $MgSO_4$ with EtOAc (3 X 15mL). The filtrate was washed with saturated $NaHCO_3$ (2 X 15 mL), dried over Na_2SO_4, and concentrated to dryness. Flash column chromatography (Hexane : EtOAc = 12:1 to 7:1) gave the desired α-product **6** (653 mg, 78%). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.44~7.12 (50H, m), 5.82 (1H, m), 5.11~5.08 (2H, ovrlp), 5.05~4.97 (2H, m), 4.91~4.87 (3H, ovrlp), 4.81~4.69 (6H, ovrlp), 4.56~4.46 (6H, ovrlp), 4.40~4.37 (3H, ovrlp), 4.28 (2H, dd, *J* = 20.0 Hz, 12.0 Hz), 4.20 (1H, t, *J* = 9.0 Hz), 4.14~3.92 (8H, ovrlp), 3.84 (1H, dd, *J* = 11.0 Hz, 4.3 Hz), 3.75 (1H, d, *J* = 9.7 Hz), 3.66 (1H, dd, *J* = 9.8 Hz, 7.9 Hz), 3.61~3.50 (4H, ovrlp), 3.42~3.30 (4H, ovrlp), 3.20 (1H, dd, *J* = 8.2 Hz, 4.6 Hz), 2.16 (2H, m), 1.76 (2H, m); ^{13}C NMR (125.0 MHz, $CDCl_3$) δ 138.36, 138.04, 137.96, 128.39, 128.29, 128.20, 128.19, 128.16, 128.14, 128.12, 128.06, 128.05, 128.03, 128.00, 127.75, 127.59, 127.53, 127.51, 127.47, 127.41, 127.34, 127.31, 127.27, 127.23, 127.09, 114.80, 103.50, 102.79, 100.67, 82.60, 81.63, 81.60, 79.35, 77.19, 76.54, 75.16, 75.03, 74.95, 74.92, 74.81, 74.74, 73.64, 73.20, 73.10, 72.97, 72.94, 72.37, 72.01, 69.36, 69.17, 68.26, 67.75, 67.63, 30.16, 28.88; IR (thin film) 3030, 2921, 2865, 1495, 1452, 1364, 1093, 740 cm^{-1}; ESI-MS *m/z* 1496.0, $[M+Na]^+$. $[\alpha]_D^{22} = +26^o$ (c = 1.0, $CHCl_3$).

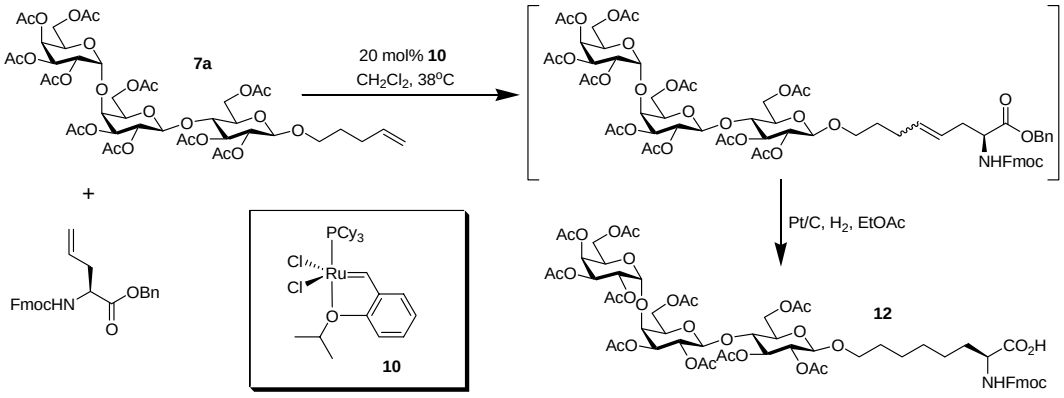
[S1] Allen, J. R.; Allen, J. G.; Zhang, X.-F.; Williams, L. J.; Zatorski, A.; Ragupathi, G.; Livingston, P. O.; Danishefsky, S. J. *Chem. Eur. J.* **2000**, *6*, 1366-1375.



7a

$C_{43}H_{60}O_{26}$
Exact Mass: 992.3373
Mol. Wt.: 992.9209

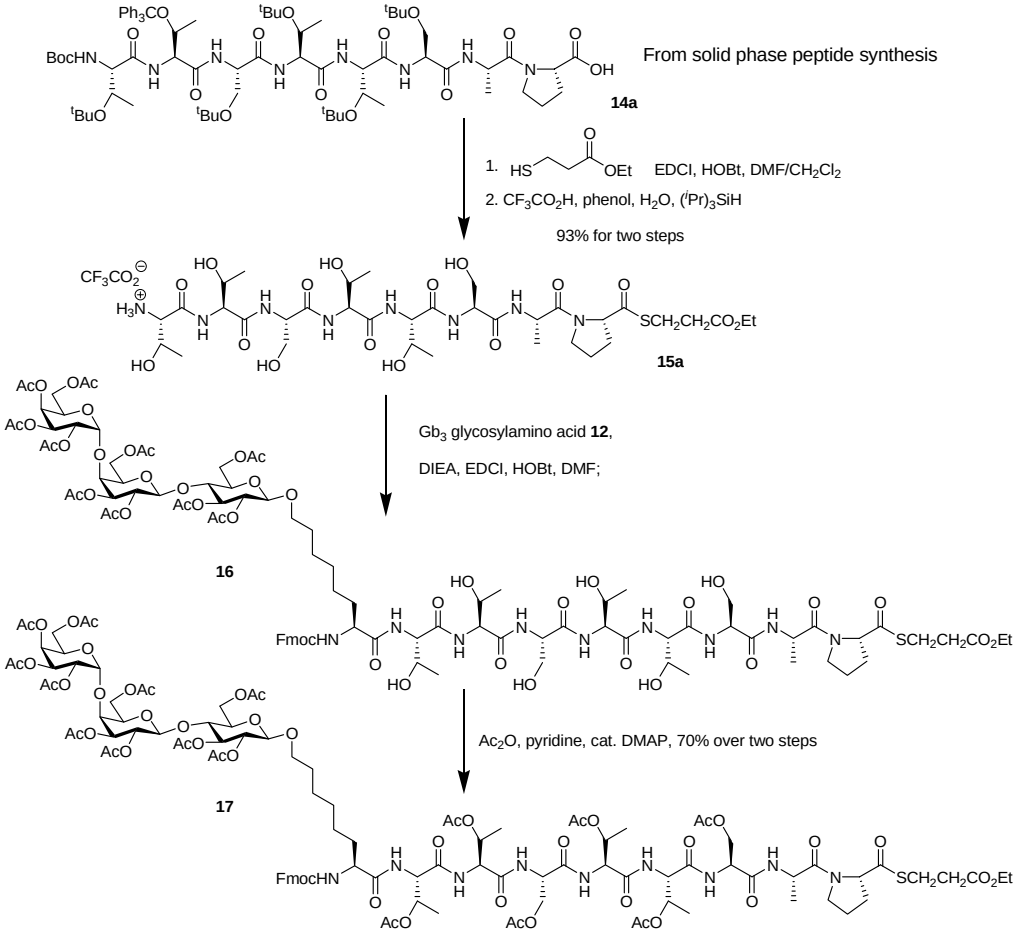
Peracetate of globo-H pentenyl glycoside 7a: To condensed liquid NH_3 (30 mL) cooled at -78°C was added sodium (676 mg, 29.4 mmol) under positive argon pressure, and then the resulting blue solution was stirred at -78°C for 20 min. Anhydrous THF (2.0 mL) was added to the blue solution, 10 minutes later perbenzylated trisaccharide **6** (722 mg, 0.49 mmol) in 4.0 mL THF was added. The resulting blue solution was stirred at -78°C for 2 hours. The reaction was quenched with solid ammonium chloride (1.54 g) and anhydrous MeOH (3.0 mL), concentrated under a stream of dry N_2. To the residue was added 5.0 mL acetic anhydride, 10.0 mL pyridine and a crystal of 4-dimethylaminopyridine (DMAP), and then the reaction stirred at RT overnight. Concentration followed by purification by flash column chromatography (toluene/EtOAc = 1/1 to 1/1.5) gave 446 mg desired product **7a** as a white solid (92% yield). All data were consistent with our previously reported ^1H, ^{13}C NMR, IR, and CIHRMS data of **7a** [S2].



Gb_3 glycosylamino acid **12**: To peracetate of globo-H pentenyl glycoside **7a** (446 mg, 0.45 mmol), allylglycine benzylester (1.54 g, 3.6 mmol) and Hoveyda-Grubbs 1st generation catalyst **10** (67 mg, 0.11 mmol) was added degassed CH_2Cl_2 (6 mL), and then the mixture was heated at 37-38 °C with water cooling for 48 hours. The reaction mixture was purified on column chromatography (Hexanes/EtOAc = 3/1 to 1/1 to 1/2) to afford 560 mg slightly impure cross-linked product. To this cross-linked compound (560 mg) was added 5% platinum on carbon (156 mg) and EtOAc (6.0 mL). The reaction mixture was

[S2] Wan, Q.; Cho, Y. S.; Lambert, T. H.; Danishefsky, S. J. *J. Carbohydr. Chem.* **2005**, *24*, 425-440.

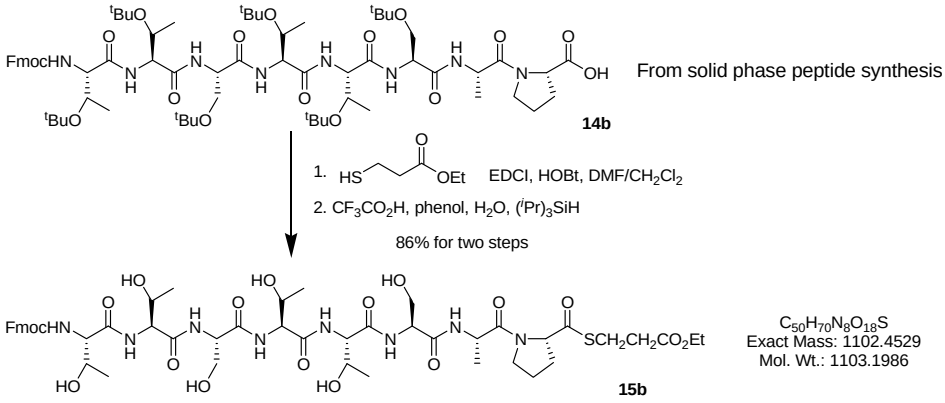
stirred under H_2 atmosphere until full disappearance of starting material. Concentration followed by purification by flash column chromatography (CH_2Cl_2 to CH_2Cl_2/EtOAc = 1.5/1 to CH_2Cl_2/MeOH = 30/1 with 0.2% HOAc) gave 384 mg desired Gb_3 glycosylamino acid **12** (66% over two steps). All data were consistent with our previously reported ^1H, ^{13}C NMR, IR, and CIHRMS data of **12**.[S2]



To a mixture of peptide **14a** (prepared by solid phase peptide synthesis, 500 mg, 0.36 mmol), EDCI (414 mg, 2.16 mmol), HOBt (292 mg, 2.16 mmol) in DMF/CH_2Cl_2 (2.0/2.0 mL) was added ethyl 3-mercaptopropionate (456 μL, 3.6 mmol), the reaction was stirred at room temperature overnight. Nitrogen flow was applied to remove all of the volatiles and the residue was purified on flash column chromatography (CH_2Cl_2 to CH_2Cl_2/MeOH = 40/1) to afford the desired thioester (504 mg, 93% yield). To this thioester (504 mg, 0.335 mmol) was added phenol (100 mg, 1.1 mmol), triisopropylsilane (251 μL, 1.23 mmol), water (335 μL, 18.6 mmol) and 5.0 mL trifluoroacetic acid, the reaction was stirred at room temperature for 3.5 hours before nitrogen flow was applied to remove all of the volatiles. Diethyl ether was added to the residue, the resulting heterogeneous mixture was shaken for 5 minutes and the

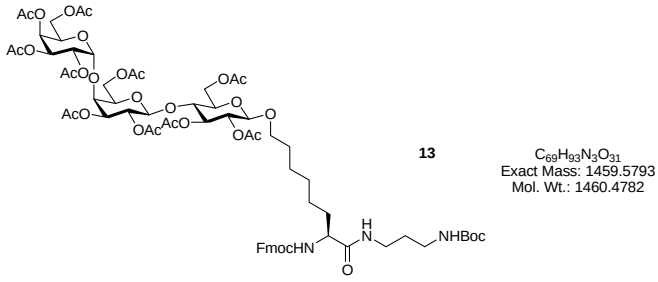
ether layer was removed by centrifugalization. This process was repeated three times to ensure complete removal of the impurities. The desired salt **15a** (332 mg, quantitative yield) was obtained as white solid. ESI-MS *m/z* 881.6 $[M+H]^+$, 903.4 $[M+Na]^+$, 977.5 $[M+CF_3CO_2H-H_2O+H]^+$, 999.5 $[M+CF_3CO_2H-H_2O+Na]^+$,1073.4 $[M+2CF_3CO_2H-2H_2O+H]^+$, 1095.6 $[M+2CF_3CO_2H-2H_2O+Na]^+$.

To salt **15a** (292 mg, 0.293 mmol), Gb_3 glycosylamino acid **12** (180 mg, 0.138 mmol), EDCI (69 mg, 0.360 mmol), HOBt (65 mg, 0.48 mmol) was added DMF (3.0 mL) and *N*, *N*-diisopropylethylamine (48 μL), the reaction mixture was stirred at room temperature for 38 hours. Nitrogen flow was applied to remove all of the volatiles to afford crude **16** (ESI-MS *m/z* 2189.0 $[M+Na]^+$, 1106.1 $[M+2Na]^{2+}$). Compound **16** was next treated with Ac_2O (2.0 mL), pyridine (3.0 mL) and 4-dimethylaminopyridine (10 mg). The mixture was stirred at room temperature overnight before nitrogen flow was applied to remove all of the volatiles. The residue was first purified on column chromatography (CH_2Cl_2 to CH_2Cl_2/MeOH = 10/1); then preparative TLC (CH_2Cl_2/MeOH = 15/1); and further by preparative HPLC (C18 column, 45-95% CH_3CN in H_2O, 16 mL/min, 30 min) to afford 234 mg desired product Gb_3-MUC5AC cassettes **17** (70% yield for two steps). ESI-MS *m/z* 2440.8 $[M+Na]^+$, 1232.2 $[M+2Na]^{2+}$.

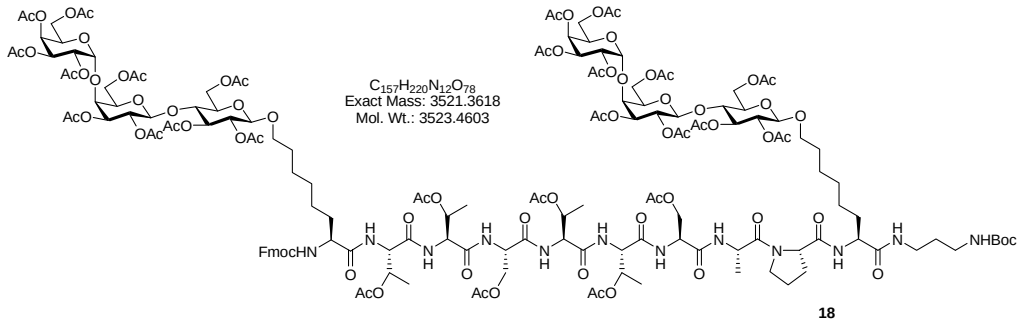


To a mixture of peptide **14b** (prepared by solid phase peptide synthesis, 264 mg, 0.2 mmol), EDCI (384 mg, 2.0 mmol), HOBt (270 mg, 2.0 mmol) in DMF/CH_2Cl_2 (1.5/1.5 mL) was added ethyl 3-mercaptopropionate (506 μL, 4.0 mmol), the reaction was stirred at room temperature overnight. Nitrogen flow was applied to remove all of the volatiles and the residue was purified on flash column chromatography (CH_2Cl_2 to CH_2Cl_2/MeOH = 25/1) to afford the desired thioester (280 mg, 97% yield). To this thioester (260 mg, 0.18 mmol) was added phenol (108 mg, 1.15 mmol), triisopropylsilane (300 μL, 1.47 mmol), water (360 μL, 20 mmol) and 5.4 mL trifluoroacetic acid, the reaction was stirred at room temperature for 3.5 hours before nitrogen flow was applied to remove all of the volatiles. The

residue was purified on column chromatography (CH_2Cl_2/MeOH = 20/1 to 15/1 to 10/1 to 6/1) to yield 177 mg desired product **15b** (89% yield). ESI-MS *m/z* 1125.5 $[M+Na]^+$.

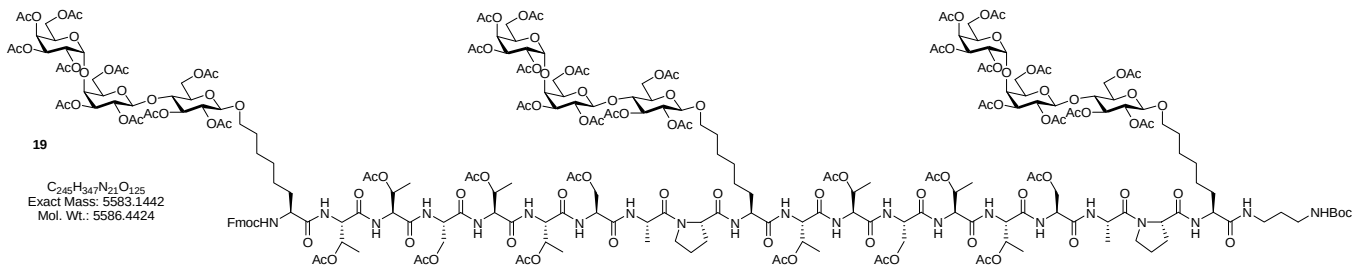


To an oven-dried 5 ml round-bottomed flask was added Gb_3-glycosylamino acid **12** (112 mg, 86 μmol), 1-Hydroxybenzotriazole (17.5 mg, 129 μmol), and *N*-(3-Dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (EDCI, 25 mg, 129 μmol). Anhydrous DMF (0.6 mL) and 16.5 mg of *N*-Boc-1,3-propanediamine in anhydrous CH_2Cl_2 (0.6 mL) was then added. The mixture was stirred at RT under argon for 30 minutes. The reaction mixture was diluted with EtOAc, sequentially washed with 1M citric acid, saturated aqueous $NaHCO_3$ solution, saturated aqueous NaCl solution. The organic layer was separated, dried over Na_2SO_4 anhydrous, filtered, and concentrated. The residue was purified on column chromatography (CH_2Cl_2: MeOH = 15:1) to yield 119 mg desired product **13** (95% yield). ESI-MS *m/z* 1482.9 $[M+Na]^+$.

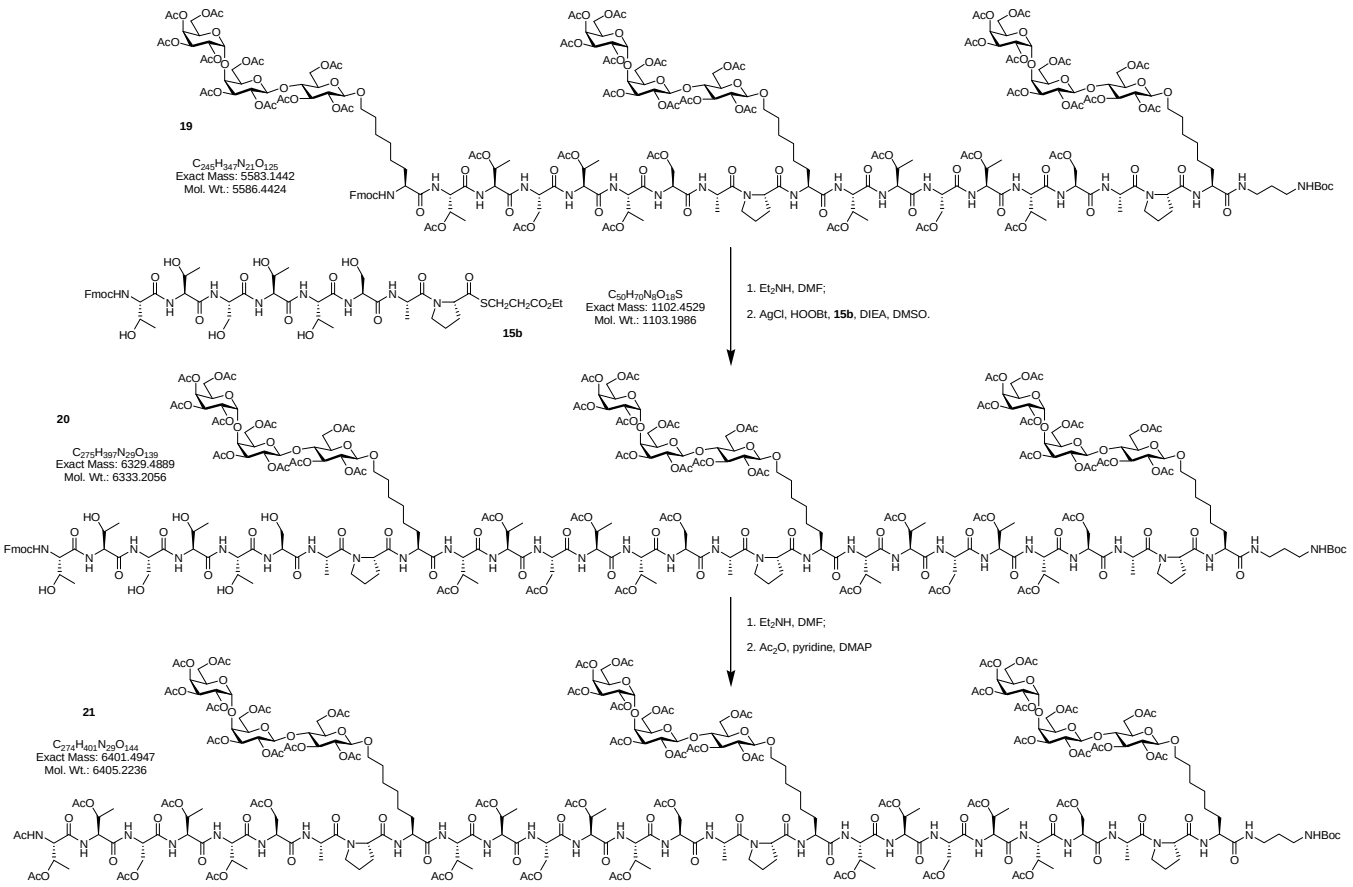


To construct **13** (67 mg, 46 μmol) was added 1.0 mL DMF and 50 μL diethylamine. The reaction mixture was stirred for 2 h before N_2 flow was applied to remove excess diethylamine and DMF. The residue was redissolved in CH_2Cl_2, and then N_2 flow was applied again applied to remove all volatiles. This process was repeated three times and further dried under high vacuum overnight to make sure excess diethylamine was completelty removed. To this residue was sequentially added Gb_3-MUC5AC

thioester **17** (97 mg, 40 μmol), AgCl (17.2 mg, 120 μmol), HOOBt (98 mg, 0.6 mmol), DMSO (2.0 mL) and *N*, *N*-diisopropylethylamine (70 μL, 0.4 mmol), and then this reaction mixture was stirred under dark for 48 h. After N_2 flow was applied to remove DMSO and *N*, *N*-diisopropylethylamine, the residue was first purified on column chromatography (CH_2Cl_2 to CH_2Cl_2/MeOH = 10/1); then preparative TLC (CH_2Cl_2/MeOH = 16/1); and further by preparative HPLC (C18 column, 45-95% CH_3CN in H_2O, 16 mL/min, 30 min) to afford 98.5 mg desired product bis-Gb_3-mono-MUC5AC **18** (70% yield for two steps). ESI-MS *m/z* 1783.9 $[M+2Na]^{2+}$, 1197.3 $[M+3Na]^{3+}$.



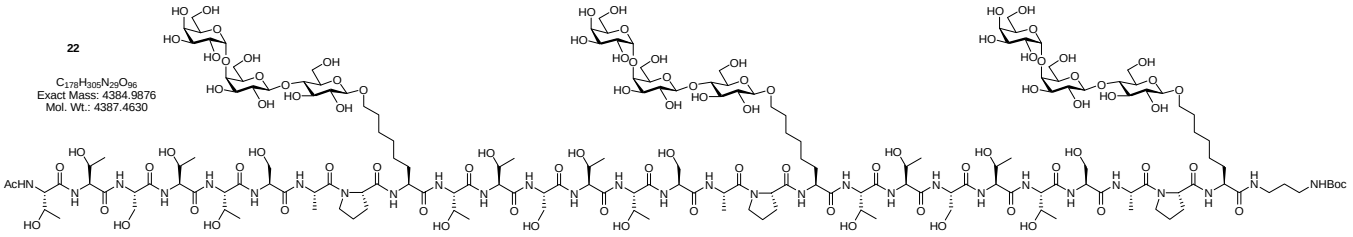
To bis-Gb_3-mono-MUC5AC **18** (42.5 mg, 12 μmol) was added 0.8 mL DMF and 40 μL diethylamine. The reaction mixture was stirred for 2 h before N_2 flow was applied to remove excess diethylamine and DMF. The residue was redissolved in CH_2Cl_2, and then N_2 flow was applied again applied to remove all volatiles. This process was repeated three times and further dried under high vacuum overnight to make sure excess diethylamine was completelty removed. To this residue was sequentially added Gb_3-MUC5AC thioester **17** (32 mg, 13.2 μmol), AgCl (5.7 mg, 40 μmol), HOOBt (31 mg, 192 μmol), DMSO (0.8 mL) and *N*, *N*-diisopropylethylamine (23 μL, 132 μmol), and then this reaction mixture was stirred under dark for 36 h. After N_2 flow was applied to remove DMSO and *N*, *N*-diisopropylethylamine, the residue was purified on column chromatography (CH_2Cl_2/MeOH = 30/1 to 15/1) and further by preparative HPLC (C18 column, 45-95% CH_3CN in H_2O, 16 mL/min, 30 min) to afford 48.5 mg desired product tris-Gb_3-bis-MUC5AC **19** (72% yield for two steps). ESI-MS *m/z* 2814.4 $[M+2Na]^{2+}$, 1884.5 $[M+3Na]^{3+}$, 1418.9 $[M+4Na]^{4+}$.



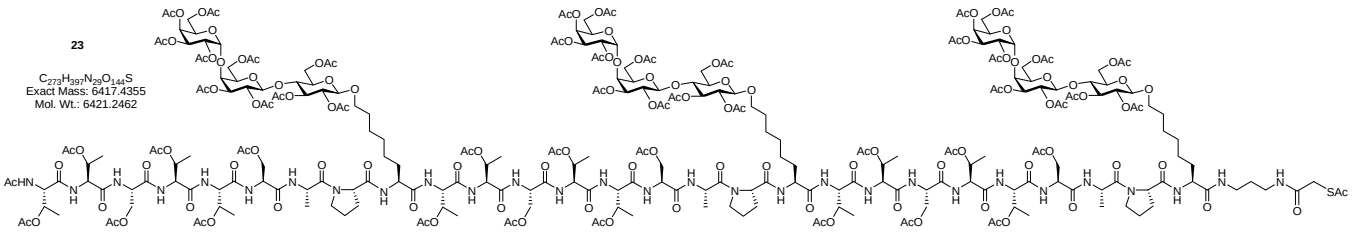
To tris-Gb_3-bis-MUC5AC **19** (36.0 mg, 6.44 μmol) was added 0.6 mL DMF and 30 μL diethylamine. The reaction mixture was stirred for 2 h before N_2 flow was applied to remove excess diethylamine and DMF. The residue was redissolved in CH_2Cl_2, and then N_2 flow was applied again applied to remove all volatiles. This process was repeated three times and further dried under high vacuum overnight to make sure excess diethylamine was completelty removed. To this residue was sequentially added MUC5AC thioester **15b** (35.5 mg, 32.2 μmol), AgCl (13.8 mg, 96.6 μmol), HOOBt (31.5 mg, 193 μmol), DMSO (1.5 mL) and *N*, *N*-diisopropylethylamine (22.5 μL, 129 μmol), and then this reaction mixture was stirred under dark for 48 h. After N_2 flow was applied to remove DMSO and *N*, *N*-diisopropylethylamine, the residue was purified on preparative TLC (CH_2Cl_2/MeOH = 9/1) afforded the desired product **20** which was directly used in the next step. ESI-MS *m/z* 2133.7 $[M+3Na]^{3+}$, 1606.2 $[M+4Na]^{4+}$, 1289.5 $[M+5Na]^{5+}$.

To the above construct **20** was added 0.6 mL DMF and 30 μL diethylamine. The reaction mixture was stirred for 2 h before N_2 flow was applied to remove excess diethylamine and DMF. The residue was redissolved in CH_2Cl_2, and then N_2 flow was applied again applied to remove all volatiles. This process was repeated three times and further dried under high vacuum overnight to make sure excess

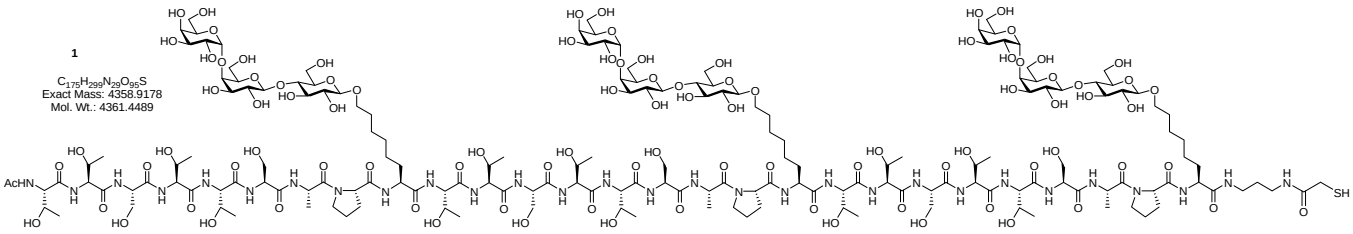
diethylamine was completelty removed. To this residue was added acetic anhydride (0.6 mL), pyridine (0.9 mL) and a small crystal of 4-dimethylaminopyridine, and then this reaction mixture was stirred overnight. N_2 flow was applied again applied to remove all volatiles and the residue was purified by column chromatography (CH_2Cl_2/MeOH = 30/1 to 10/1) to afford 25.4 mg desired tris-Gb_3-tris-MUC5AC **21** (62% over four steps). ESI-MS *m/z* 2156.7 $[M+3Na]^{3+}$.



To peracetate **21** (6.0 mg, 0.94 μmol) was added degassed NH_2NH_2/MeOH (500 μL, 1/4, v/v) at room temperature under argon. The reaction was stirred at room temperature for 36 hours before N_2 was applied to remove excess NH_2NH_2 and MeOH. The residue was dissolved in minimum amount of water and purified by bio-gel P4 column (water as eluant). All fractions containing the desired compound were combined and lyophilized to afford 3.7 mg pure construct **22** (90%). ESI-MS *m/z* 2215.5 $[M+2Na]^{2+}$, 1484.9 $[M+3Na]^{3+}$, 1119.2 $[M+4Na]^{4+}$.



To peracetate **21** (33 mg, 5.15 μmol) in 1.2 mL dichloromethane was added 0.3 mL trifluoroacetic acid, the reaction mixture was stirred at room temperature for 3 hours before nitrogen flow was applied to remove the volatiles. The residue was dried under high vacuum for 2 hours to afford corresponding crude amine salt. To this amine salt was added 0.8 mL pyridine and S-acetylthioglycolic acid pentafluorophenyl ester (SAMA-OPfp, 12.4 mg, 41 μmol) at room temperature. After stirring for 24 hours, nitrogen flow was applied to remove the volatiles and residue was purified on silica column (CH_2Cl_2/MeOH = 15:1 to 10:1) to afford 22 mg desired product **23** as white solid (66% for two steps). ESI-MS *m/z* 2162.2 $[M+3Na]^{3+}$.



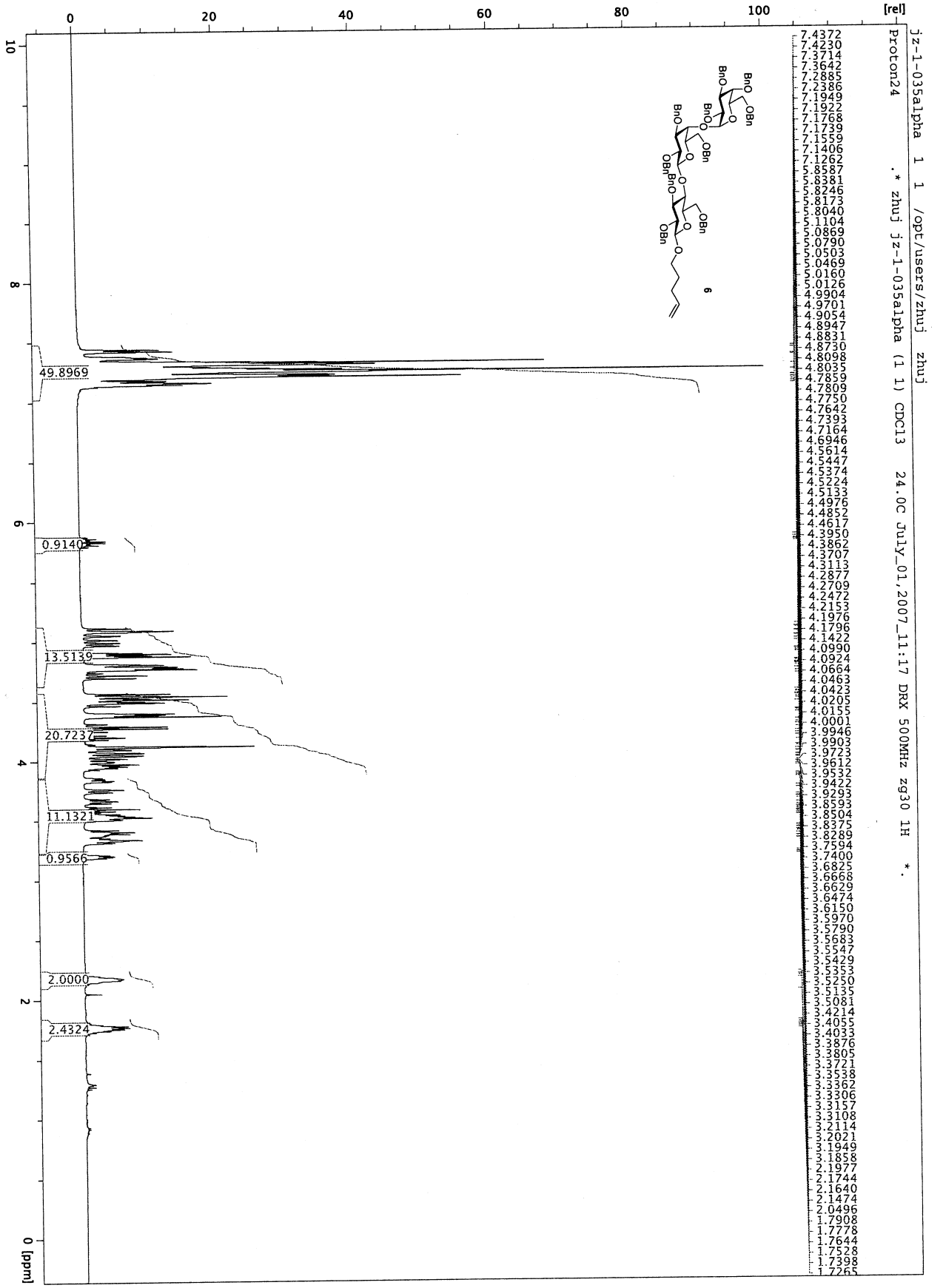
To peracetate **23** (21.0 mg, 3.27 μmol) was added degassed NH_2NH_2/MeOH (5.0 mL, 1/4, v/v) at 0 ^oC under argon. The reaction mixture was slowly warmed and stirred at room temperature for 36 hours before being concentrated to remove excess NH_2NH_2 and MeOH. To the residue was added 1.0 mL degassed water and tris(2-carboxyethyl)phosphine (TCEP, 150 μL, 0.5 M in neutral buffer) under argon pressure, and the reaction mixture was stirred for 1 hour at room temperature. This aqueous mixture was directly purified by bio-gel P4 column (***degassed water*** as eluant). All fractions containing the desired compound were combined and lyophilized to afford 12.3 mg pure construct **1** (86%). ESI-MS *m/z* 1476.2 $[M+3Na]^{3+}$, 1112.9 $[M+4Na]^{4+}$.

Conjugation of Gb_3-MUC5AC vaccine construct to KLH:

Gb_3-MUC5AC glycopeptide **1** was covalently attached to KLH using the heterobifunctional crosslinker Sulfo-MBS (*m*-maleimidobenzoyl-*N*-hydroxysulfosuccinimide ester, Pierce Co., Rockford, IL) which couples the terminal free sulfhydryl functional to amino group on KLH as described earlier.[S3,4] First KLH (9.0 mg, Sigma, molecular weight 8.6×10^6) was treated with Sulfo-MBS (3.0 mg), then the unconjugated Sulfo-MBS was eliminated by passage over a G25 Sephadex column. Maleimide activated KLH is then added to the freshly de-protected Gb_3-MUC5AC glycopeptide **1** (3.8 mg). The mixture was incubated at room temperature for 3 h, following incubation, unreacted glycopeptide was removed using a 30,000 molecular cut-off Centriprep filter. Finally the Gb_3-MUC5AC-KLH conjugate was obtained in a 6.0 mL buffer solution. The Gb_3-MUC5AC concentration in Gb_3-MUC5AC-KLH conjugate was

[3] Ragupathi, G.; Cappello, S.; Yi, S. S.; Spassova, M.; Bornmann, W.; Danishefsky, S. J.; Livingston, P. O. *Vaccine* **2002** *20,* 1030-1038.
[4] Ragupathi, G.; Coltart, D. M.; Williams, L. J.; Koide, F.; Kagan, E.; Allen, J.; Harris, C.; Glunz, P. W.; Livingston, P. O. Danishefsky, S. J. *Proc. Natl. Acad. Sci. USA* **2002**, *99*, 13699-13704.

determined using ion exchange chromatography with pulsed amperometric detection by measuring carbohydrate content of Gb_3-MUC5AC and KLH by a dye binding method (BioRad, Dye reagent). The epitope ratio of Gb_3-MUC5AC/KLH in the conjugate was **698/1** assuming a KLH molecular weight of 8.6 million.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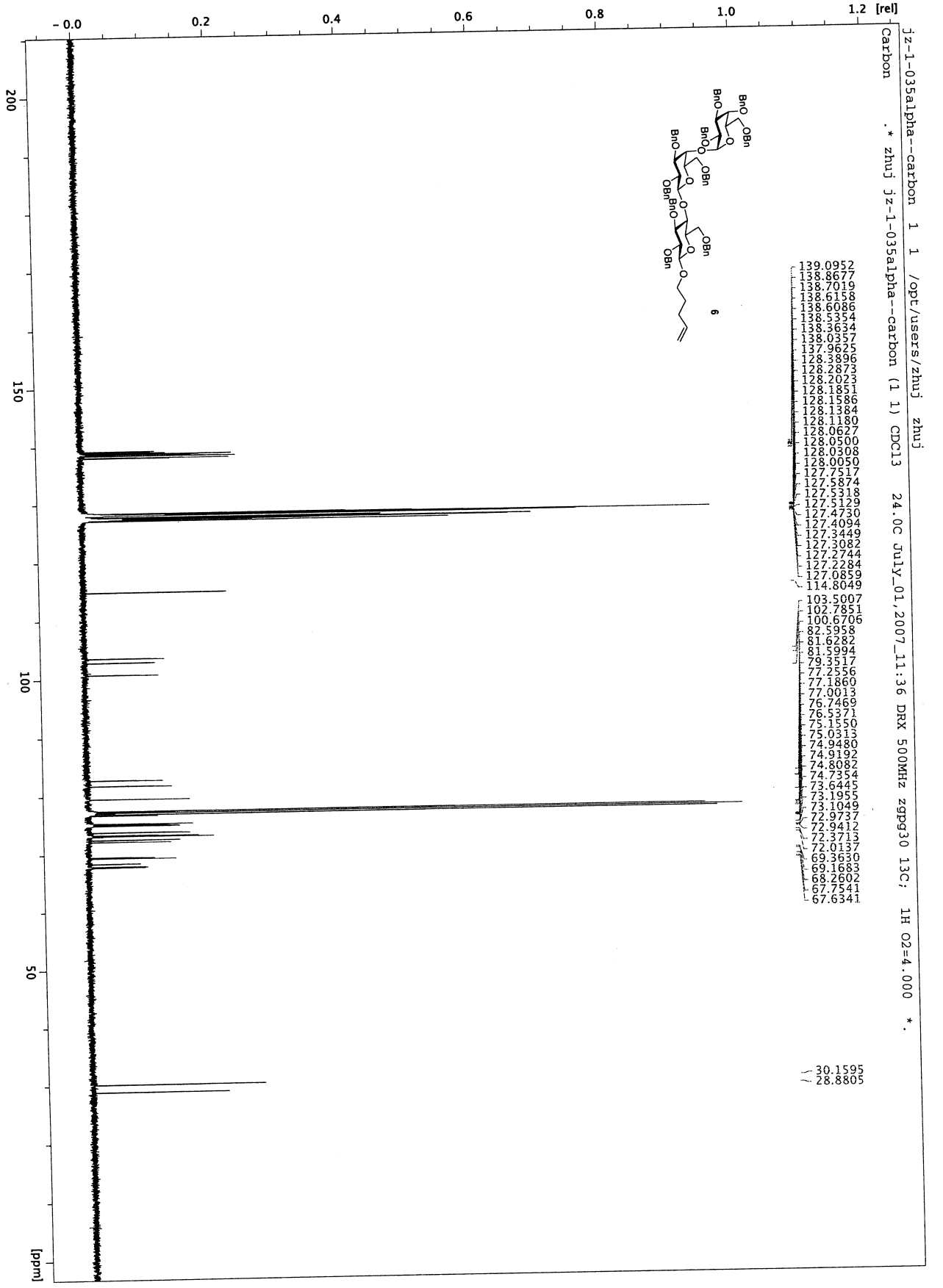
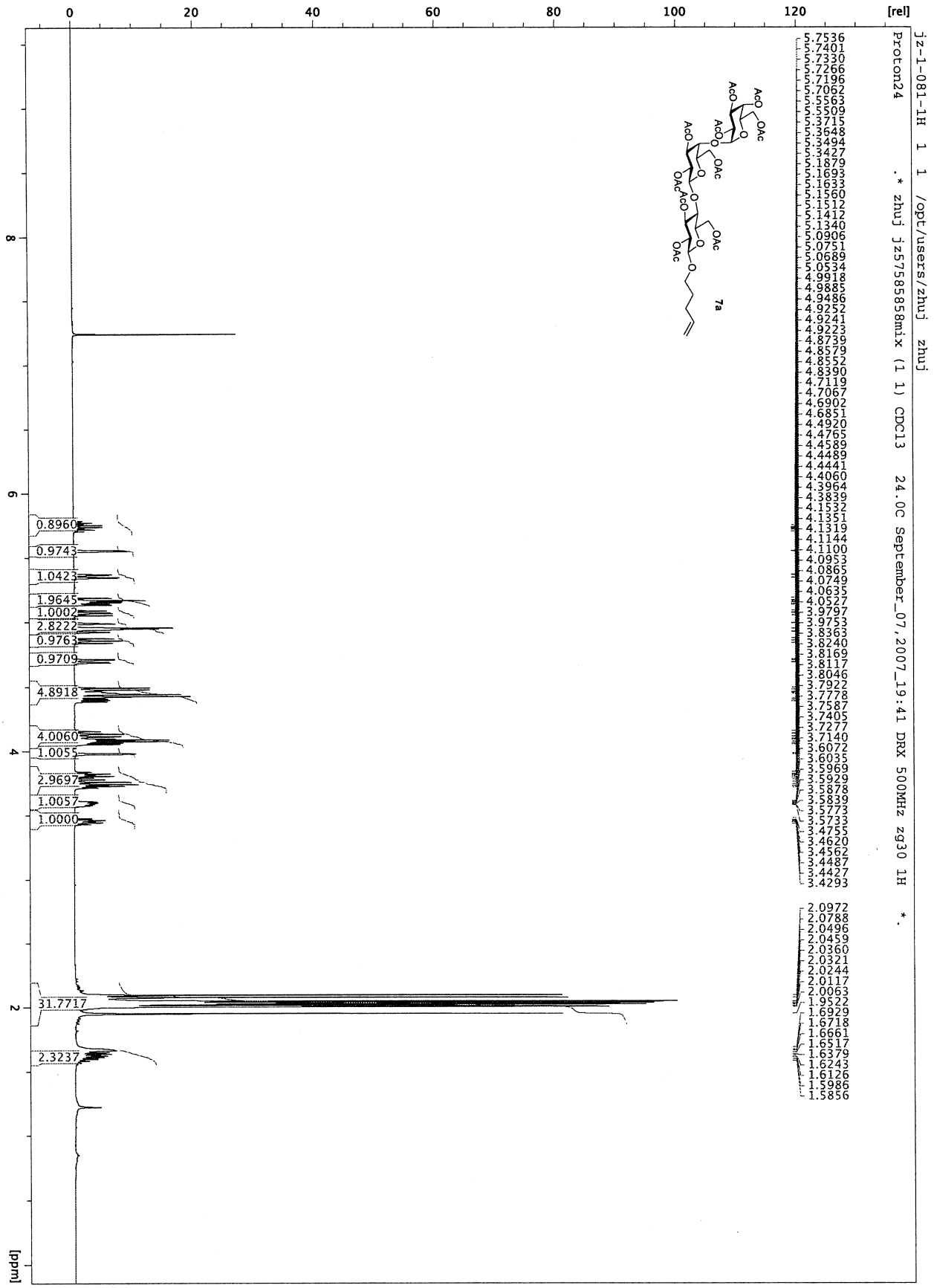
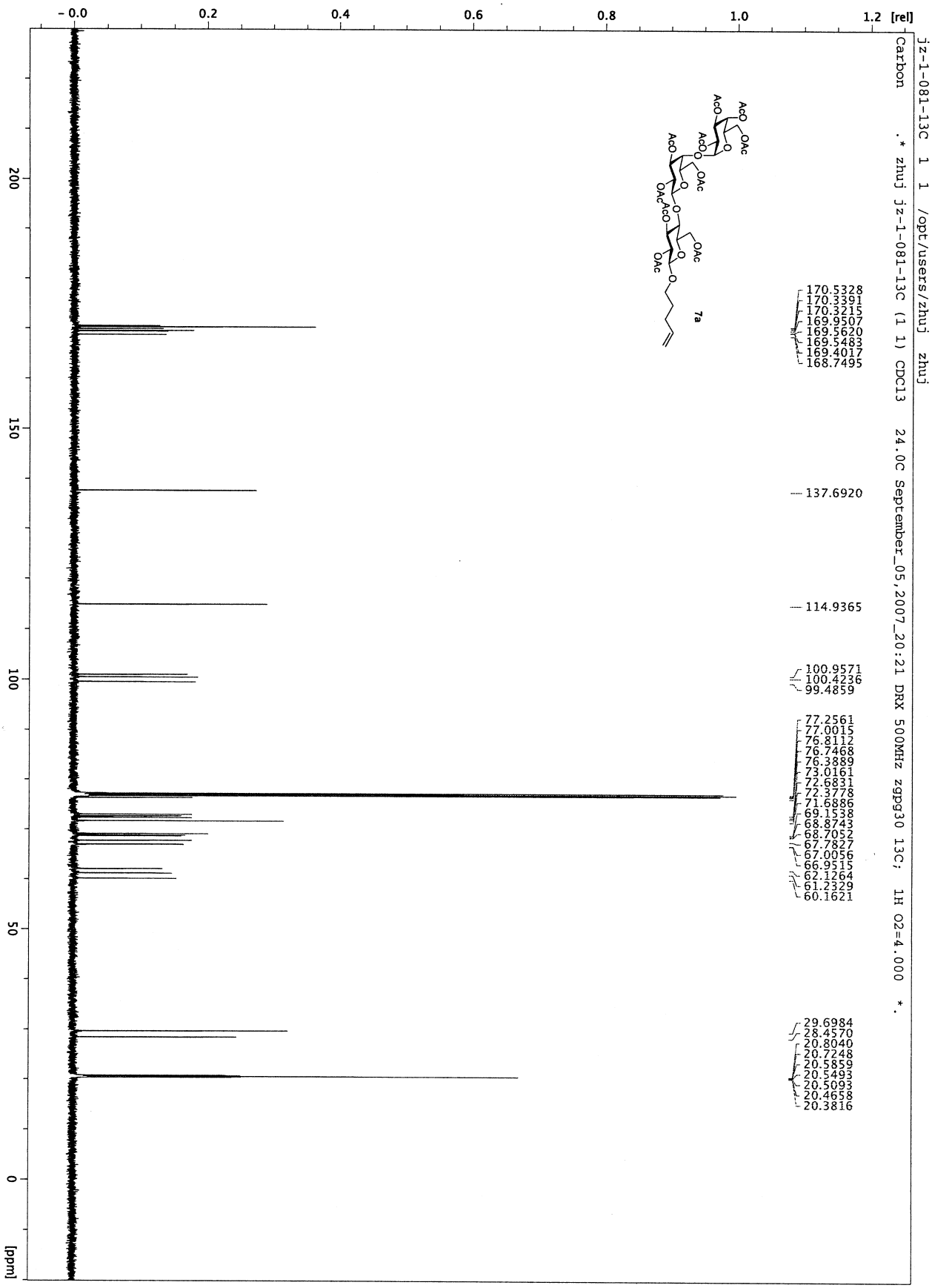
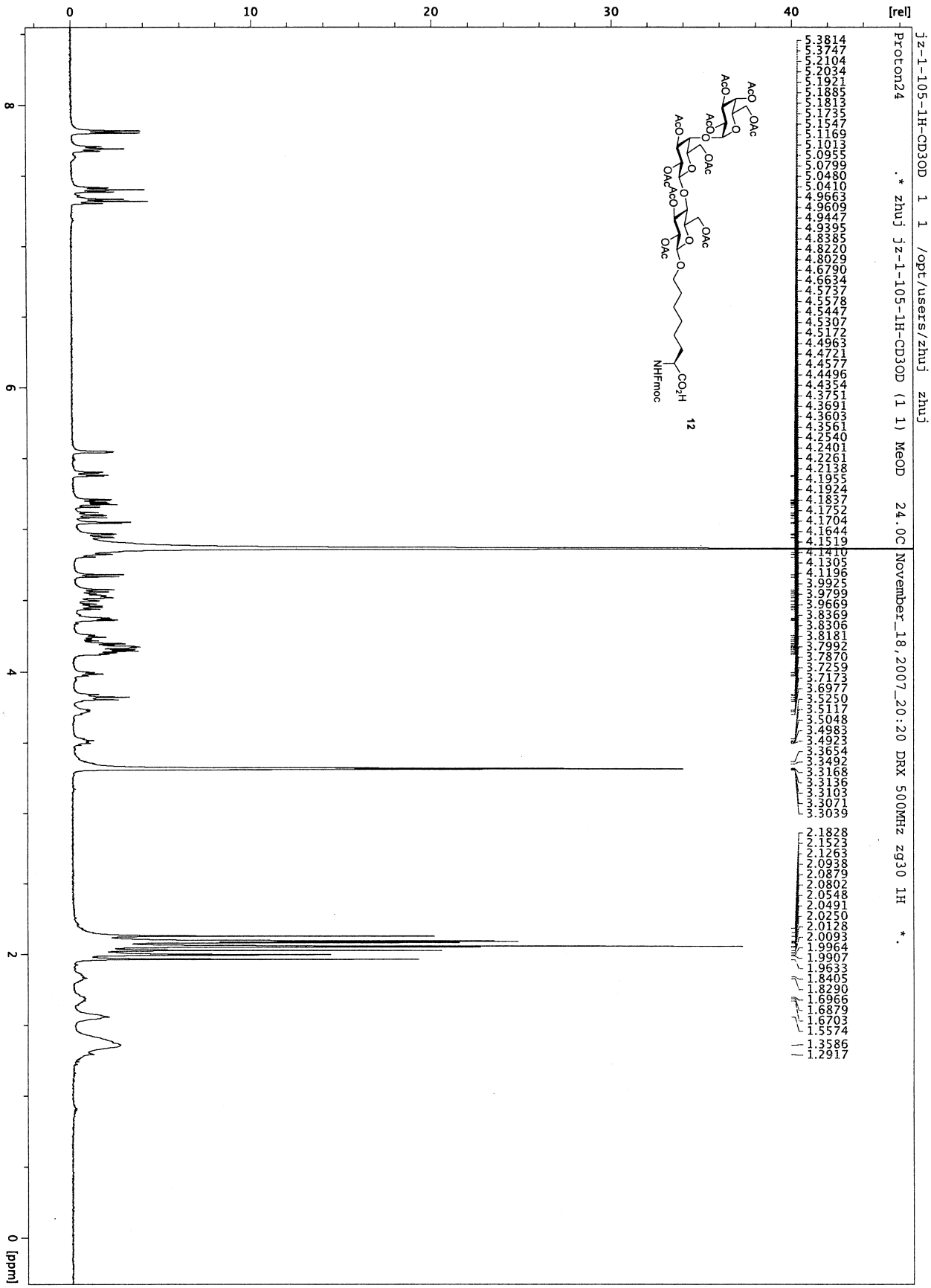
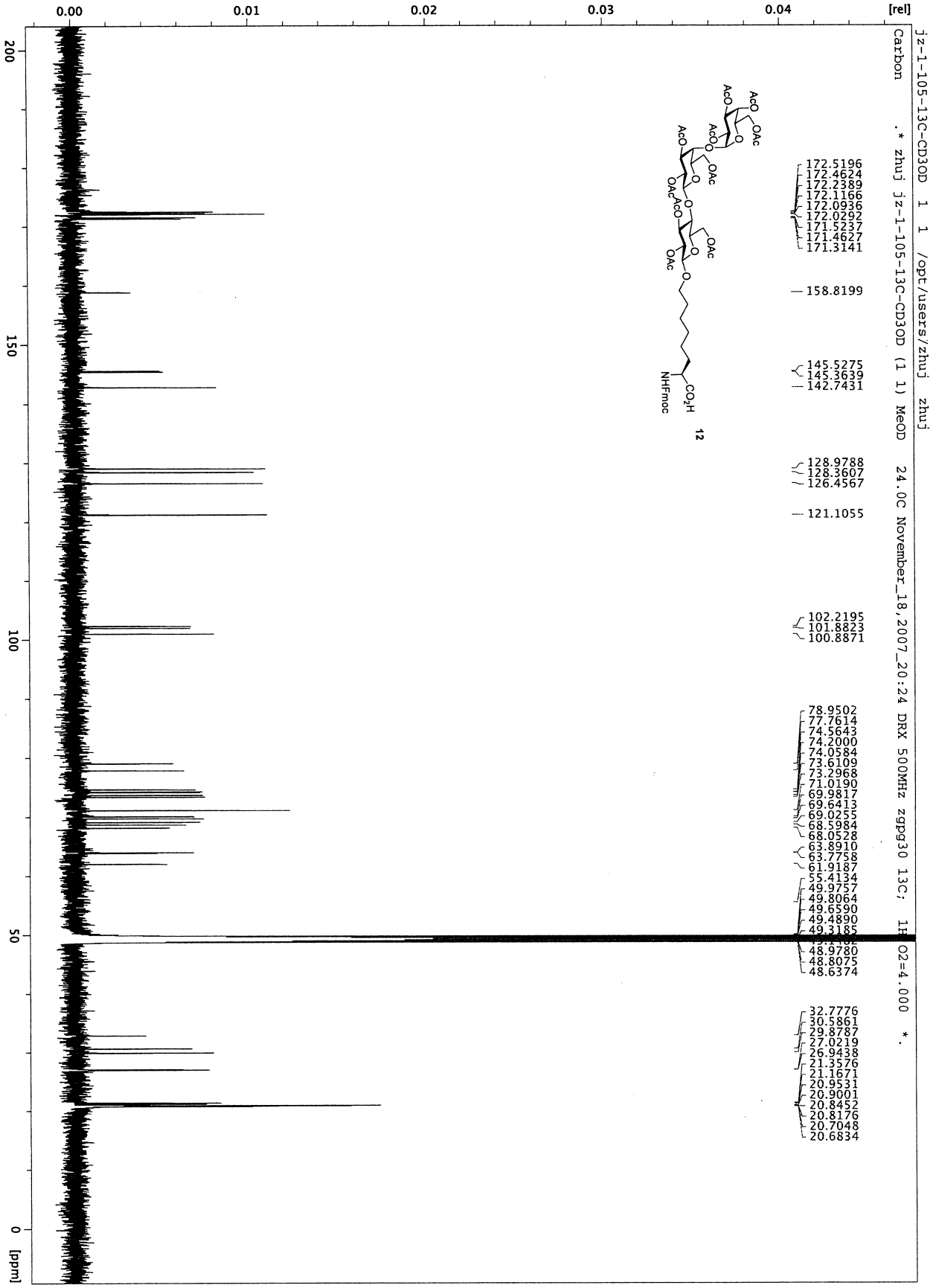
20.7237
11.1321
0.9566
2.0000
2.4324
6

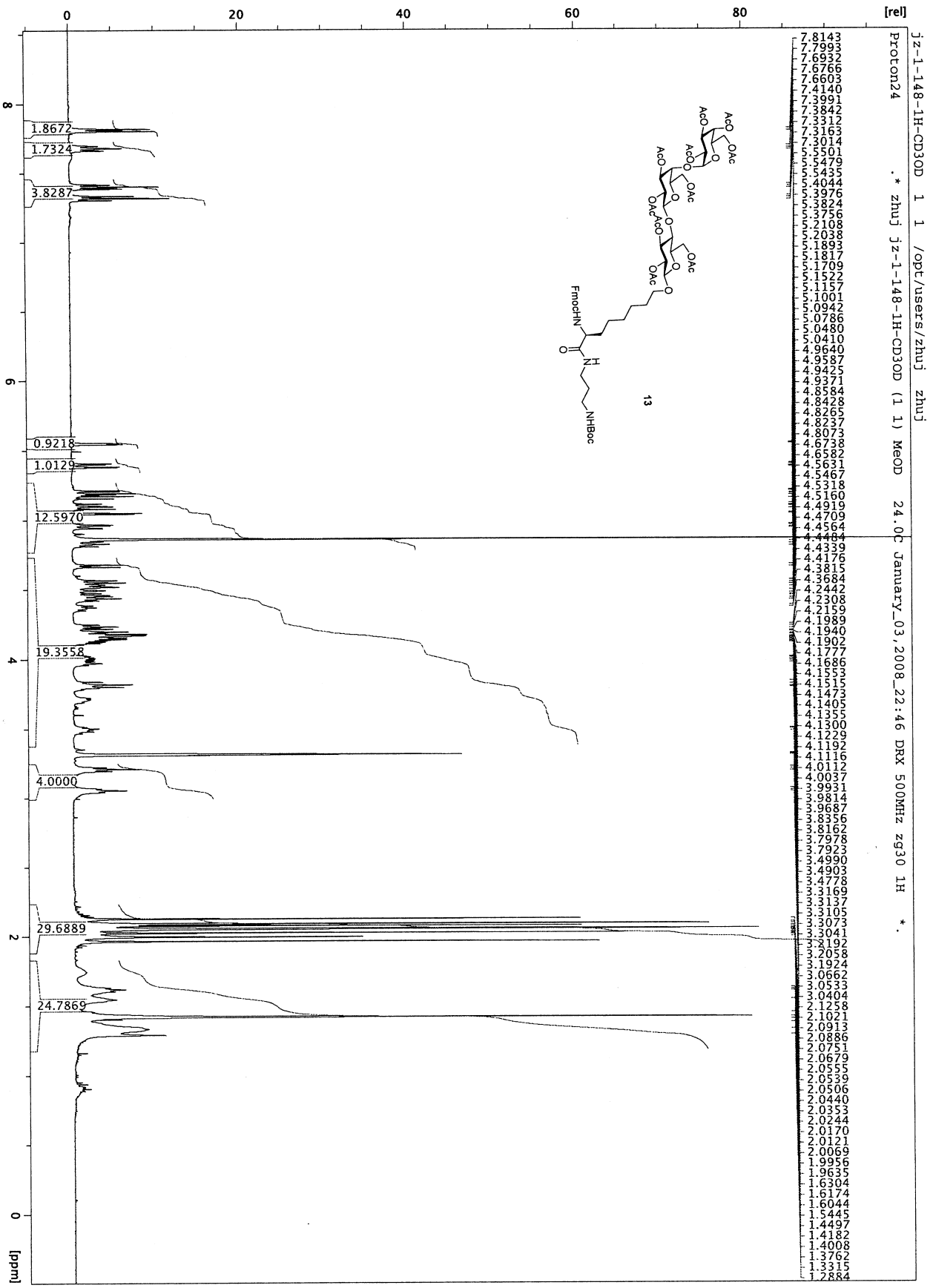

jz-1-035alpha--carbon 1 1 /opt/users/zhuj zhuj
Carbon .* zhuj jz-1-035alpha--carbon (1 1) CDCl3 24.0C July_01,2007_11:36 DRX 500MHz zgpg30 13C; 1H O2=4.000 *.
[rel]
[ppm]
6
139.0952
138.8677
138.7019
138.6158
138.6086
138.5354
138.3634
138.0357
137.9625
128.3896
128.2873
128.2023
128.1851
128.1586
128.1384
128.1180
128.0627
128.0500
128.0308
128.0050
127.7517
127.5874
127.5318
127.5129
127.4730
127.4094
127.3449
127.3082
127.2744
127.2284
127.0859
114.8049
103.5007
102.7851
100.6706
82.5958
81.6282
81.5994
79.3517
77.2556
77.1860
77.0013
76.7469
76.5371
75.1550
75.0313
74.9480
74.9192
74.8082
74.7354
73.6445
73.1955
73.1049
72.9737
72.9412
72.3713
72.0137
69.3630
69.1683
68.2602
67.7541
67.6341
30.1595
28.8805

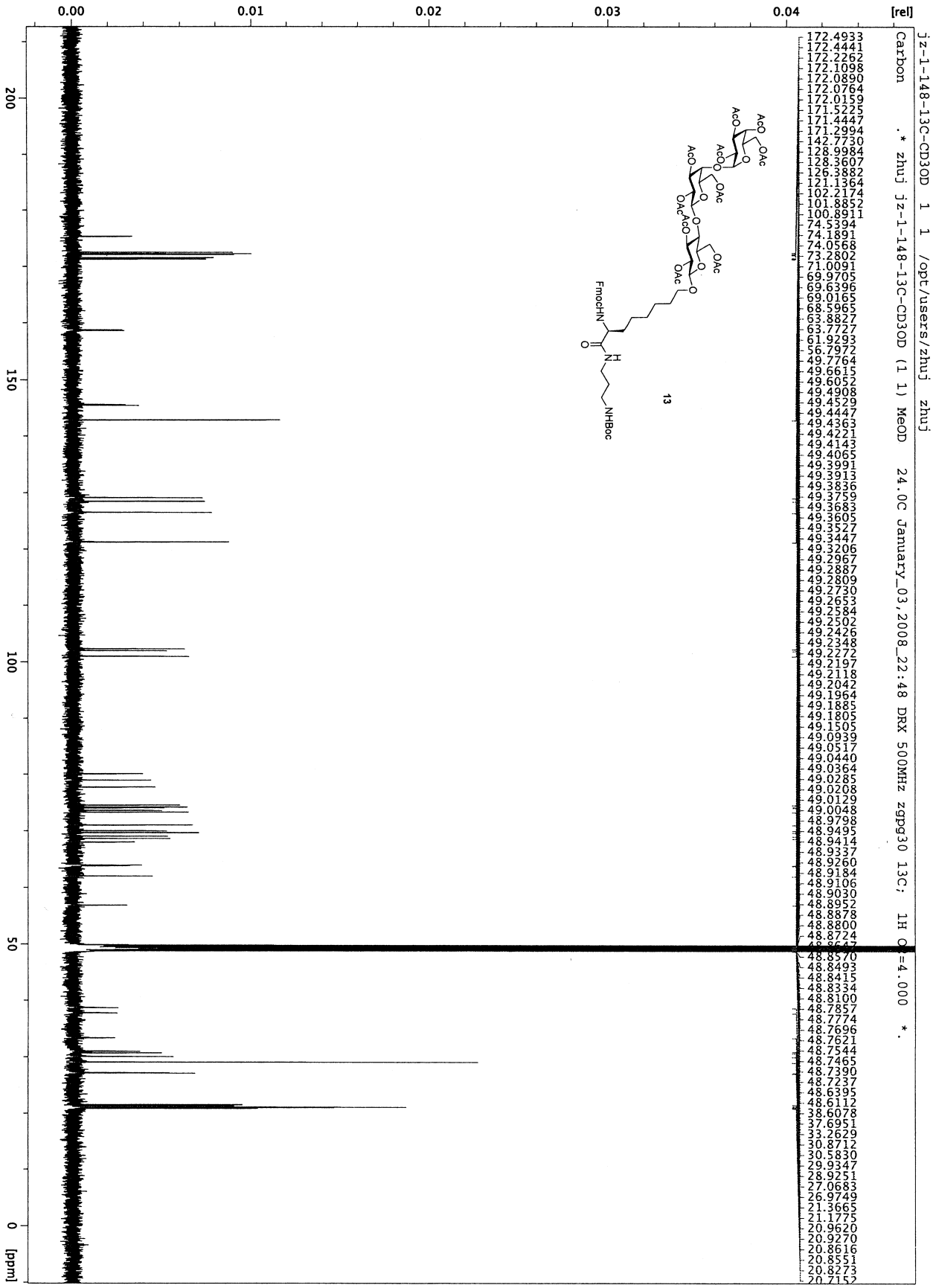

jz-1-081-1H 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz57585858mix (1 1) CDCl3 24.0C September_07,2007_19:41 DRX 500MHz zg30 1H *.
[rel]
[ppm]
7a
0.8960
0.9743
1.0423
1.9645
1.0002
2.8222
0.9763
0.9709
4.8918
4.0060
1.0055
2.9697
1.0057
1.0000
31.7717
2.3237

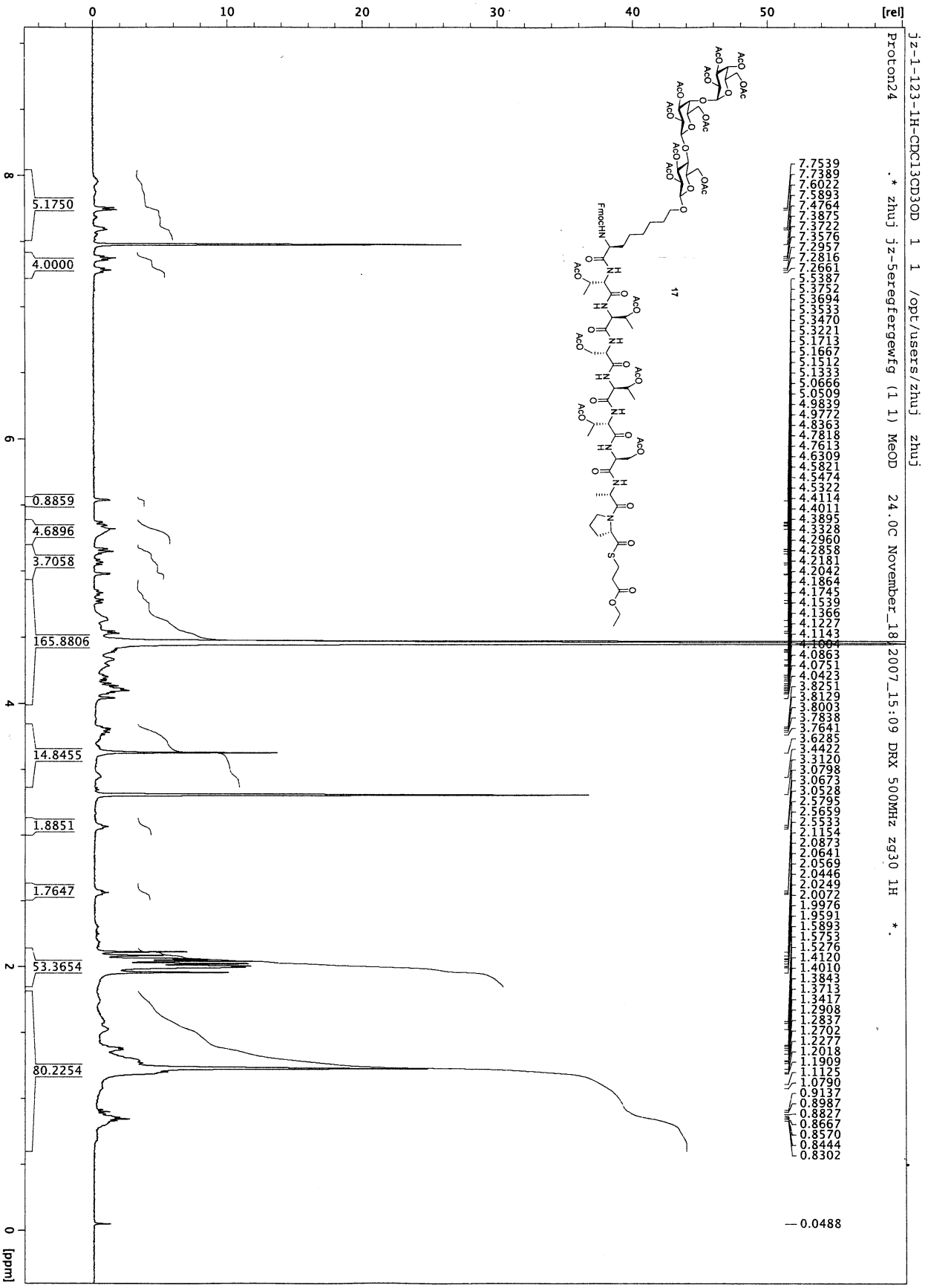

jz-1-081-13C 1 1 /opt/users/zhuj zhuj
Carbon .* zhuj jz-1-081-13C (1 1) CDCl3 24.0C September_05,2007_20:21 DRX 500MHz zgpg30 13C; 1H O2=4.000 *.
7a
170.5328
170.3391
170.3215
169.9507
169.5620
169.5483
169.4017
168.7495
137.6920
114.9365
100.9571
100.4236
99.4859
77.2561
77.0015
76.8112
76.7468
76.3889
73.0161
72.6831
72.3778
71.6886
69.1538
68.8743
68.7052
67.7827
67.0056
66.9515
62.1264
61.2329
60.1621
29.6984
28.4570
20.8040
20.7248
20.5859
20.5493
20.5093
20.4658
20.3816
[ppm]
[rel]

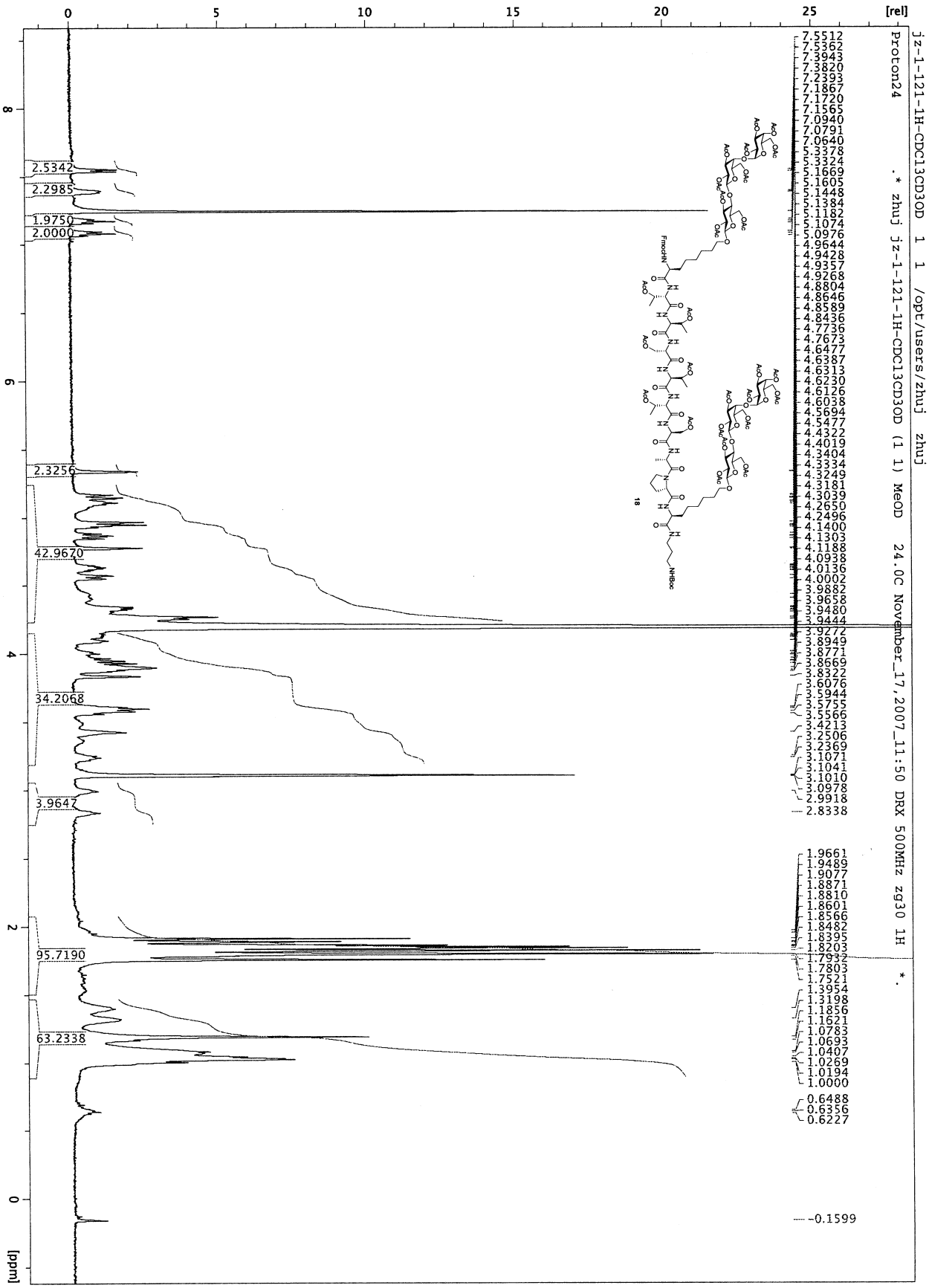

jz-1-105-1H-CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz-1-105-1H-CD3OD (1 1) MeOD 24.0C November_18,2007_20:20 DRX 500MHz zg30 1H *.
[rel]
[ppm]
12

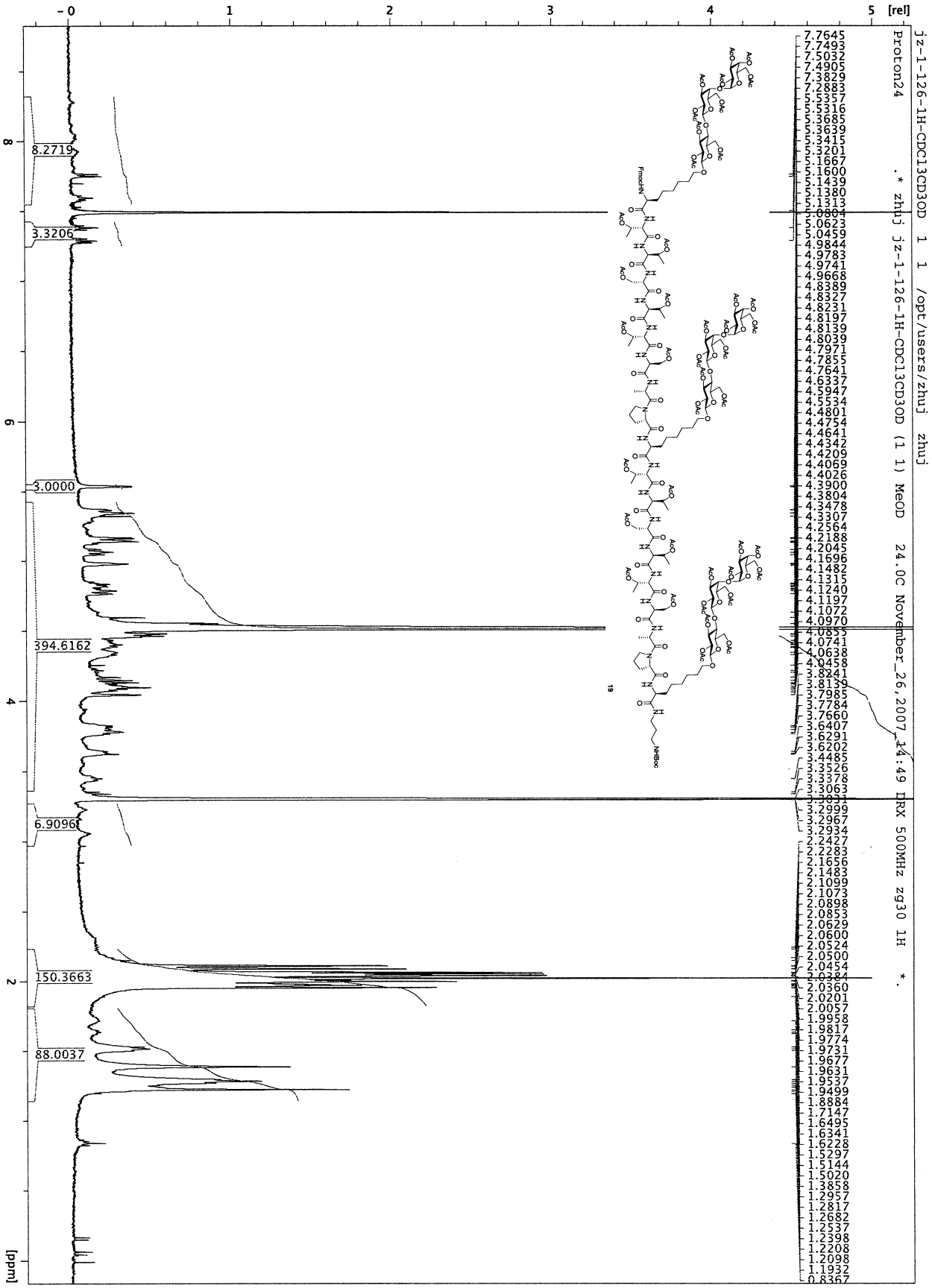

jz-1-105-13C-CD3OD 1 1 /opt/users/zhuj zhuj
Carbon .* zhuj jz-1-105-13C-CD3OD (1 1) MeOD 24.0C November_18,2007_20:24 DRX 500MHz zgpg30 13C; 1H O2=4.000 *.
0.00
0.01
0.02
0.03
0.04
[rel]
200
150
100
50
0
[ppm]
12
172.5196
172.4624
172.2389
172.1166
172.0936
172.0292
171.5237
171.4627
171.3141
158.8199
145.5275
145.3639
142.7431
128.9788
128.3607
126.4567
121.1055
102.2195
101.8823
100.8871
78.9502
77.7614
74.5643
74.2000
74.0584
73.6109
73.2968
71.0190
69.9817
69.6413
69.0255
68.5984
68.0528
63.8910
63.7758
61.9187
55.4134
49.9757
49.8064
49.6590
49.4890
49.3185
48.9780
48.8075
48.6374
32.7776
30.5861
29.8787
27.0219
26.9438
21.3576
21.1671
20.9531
20.9001
20.8452
20.8176
20.7048
20.6834

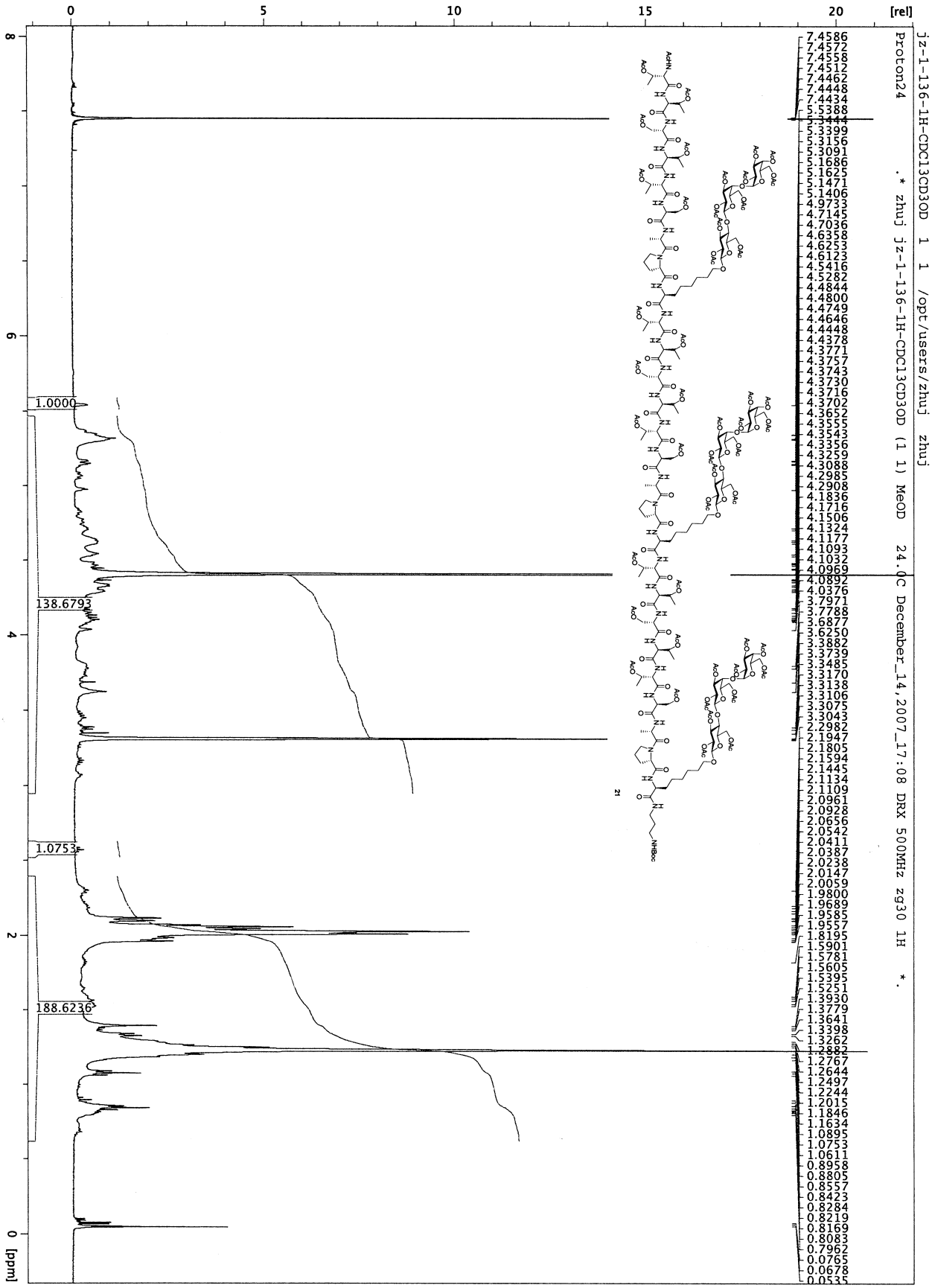

jz-1-148-1H-CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz-1-148-1H-CD3OD (1 1) MeOD 24.0C January_03,2008_22:46 DRX 500MHz zg30 1H *.
[rel]
[ppm]
1.8672
1.7324
3.8287
0.9218
1.0129
12.5970
19.3558
4.0000
29.6889
24.7869
FmocHN
NHBoc
13

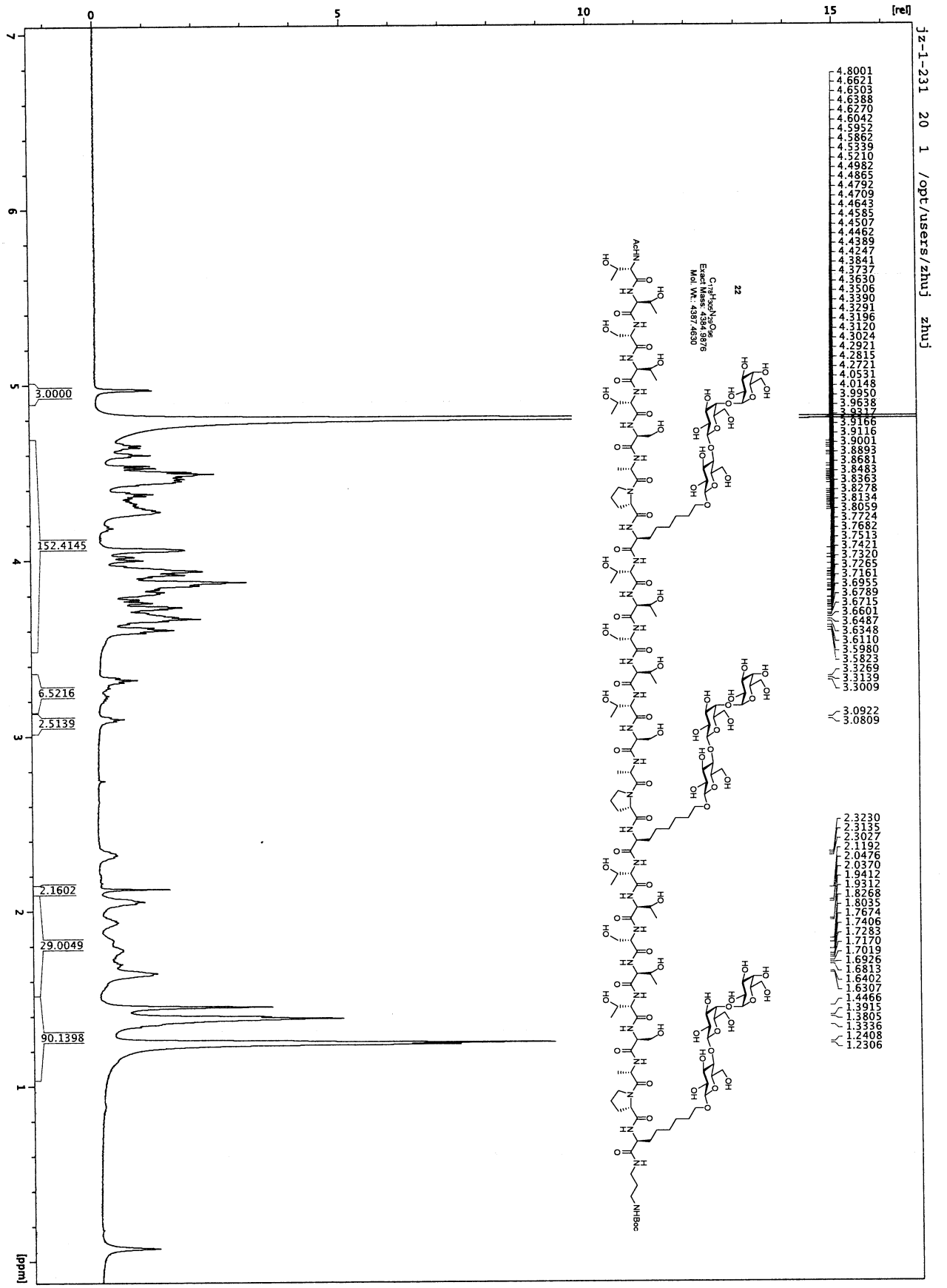

jz-1-148-13C-CD3OD 1 1 /opt/users/zhuj zhuj
Carbon .* zhuj jz-1-148-13C-CD3OD (1 1) MeOD 24.0C January_03,2008_22:48 DRX 500MHz zgpg30 13C; 1H O=4.000 *.
[rel]
0.00
0.01
0.02
0.03
0.04
200
150
100
50
0
[ppm]
FmocHN
NHBoc
AcO
OAc
13

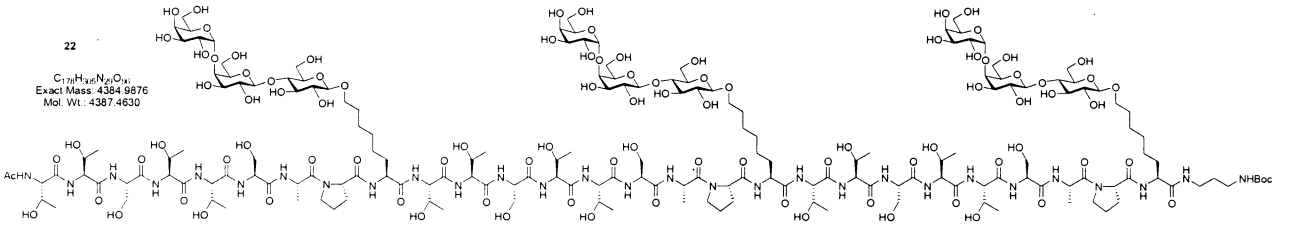

jz-1-123-1H-CDCl3CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz-5eregfergewfg (1 1) MeOD 24.0C November_18 2007_15:09 DRX 500MHz zg30 1H *.
17
5.1750
4.0000
0.8859
4.6896
3.7058
165.8806
14.8455
1.8851
1.7647
53.3654
80.2254
0.0488
[rel]
[ppm]

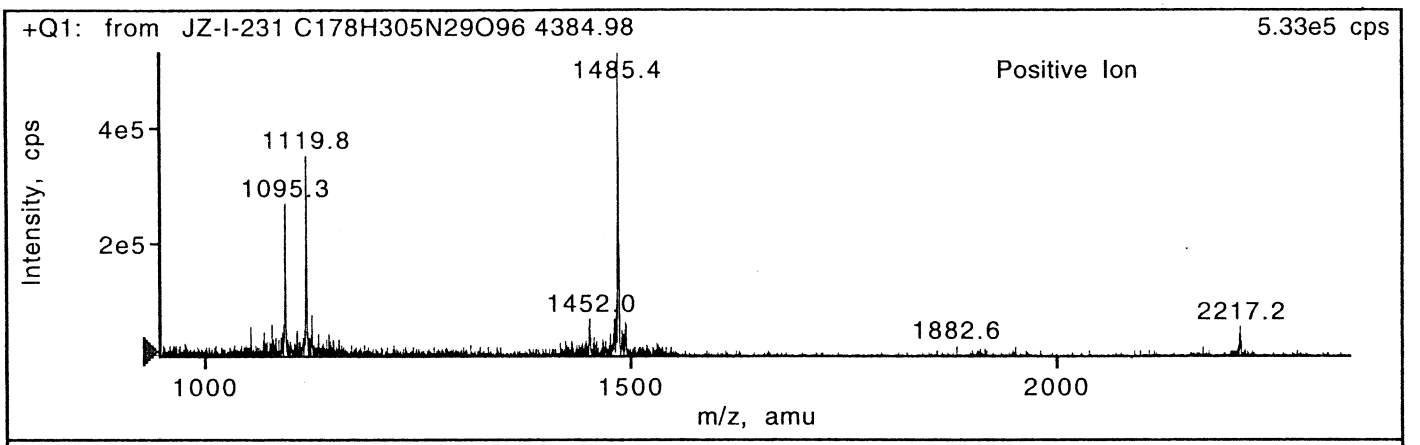

jz-1-121-1H-CDCl3CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 . * zhuj jz-1-121-1H-CDCl3CD3OD (1 1) MeOD 24.0C November_17,2007_11:50 DRX 500MHz zg30 1H
[rel]
[ppm]
2.5342
2.2985
1.9750
2.0000
2.3256
42.9620
34.2068
3.9647
95.7190
63.2338
-0.1599

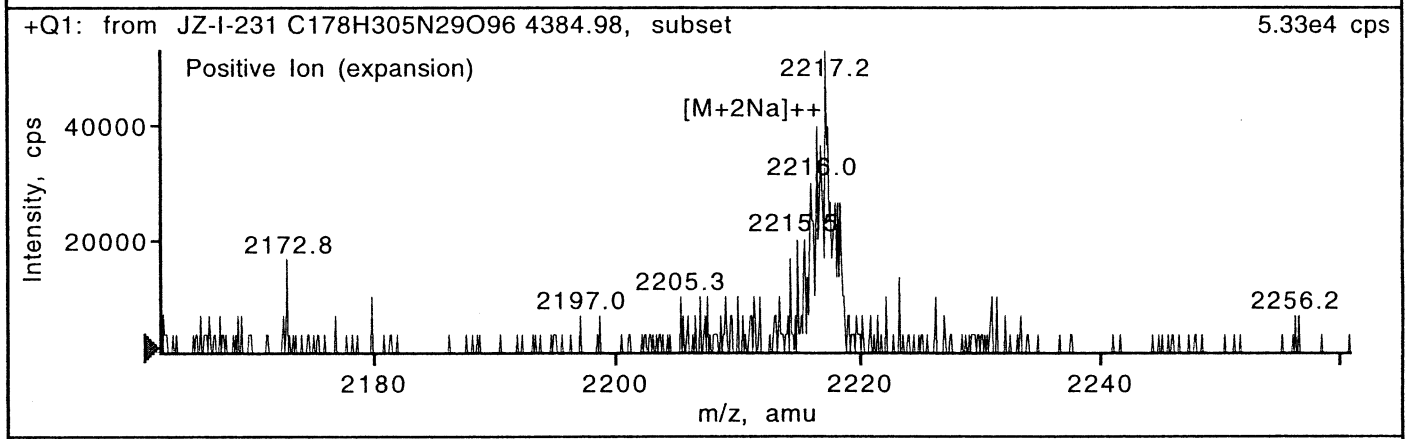

jz-1-126-1H-CDCl3CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz-1-126-1H-CDCl3CD3OD (1 1) MeOD 24.0C November_26,2007_14:49 DRX 500MHz zg30 1H *.
8.2719
3.3206
3.0000
394.6162
6.9096
150.3663
88.0037
[ppm]
[rel]

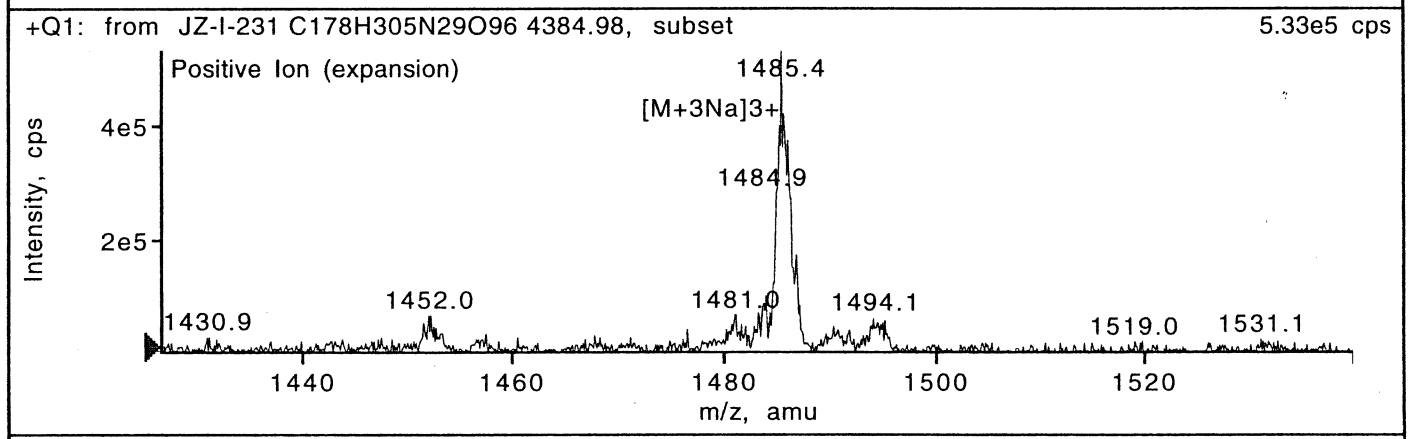

jz-1-136-1H-CDCl3CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz-1-136-1H-CDCl3CD3OD (1 1) MeOD 24.0C December_14,2007_17:08 DRX 500MHz zg30 1H *.
[rel]
[ppm]
1.0000
138.6793
1.0753
188.6236

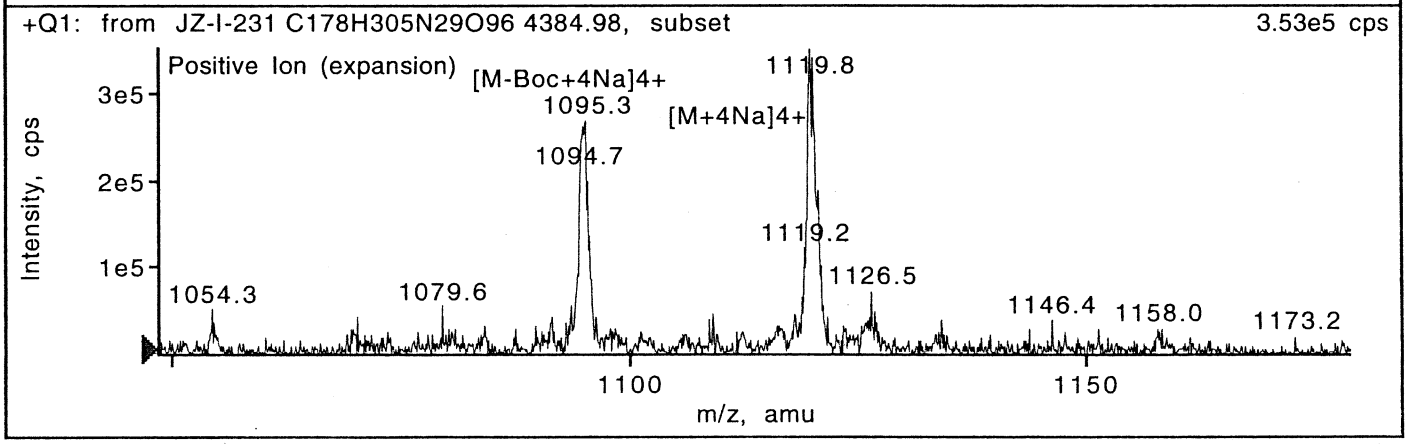

jz-1-231 20 1 /opt/users/zhuj zhuj
[rel]
[ppm]
3.0000
152.4145
6.5216
2.5139
2.1602
29.0049
90.1398
22
AcHN
NHBoc
4.8001
4.6621
4.6503
4.6388
4.6270
4.6042
4.5952
4.5862
4.5339
4.5210
4.4982
4.4865
4.4792
4.4709
4.4643
4.4585
4.4507
4.4462
4.4389
4.4247
4.3841
4.3737
4.3630
4.3506
4.3390
4.3291
4.3196
4.3120
4.3024
4.2921
4.2815
4.2721
4.0531
4.0148
3.9950
3.9638
3.9317
3.9166
3.9116
3.9001
3.8893
3.8681
3.8483
3.8363
3.8278
3.8134
3.8059
3.7724
3.7682
3.7513
3.7421
3.7320
3.7265
3.7161
3.6955
3.6789
3.6715
3.6601
3.6487
3.6348
3.6110
3.5980
3.5823
3.3269
3.3139
3.3009
3.0922
3.0809
2.3230
2.3135
2.3027
2.1192
2.0476
2.0370
1.9412
1.9312
1.8268
1.8035
1.7674
1.7406
1.7283
1.7170
1.7019
1.6926
1.6813
1.6402
1.6307
1.4466
1.3915
1.3805
1.3336
1.2408
1.2306

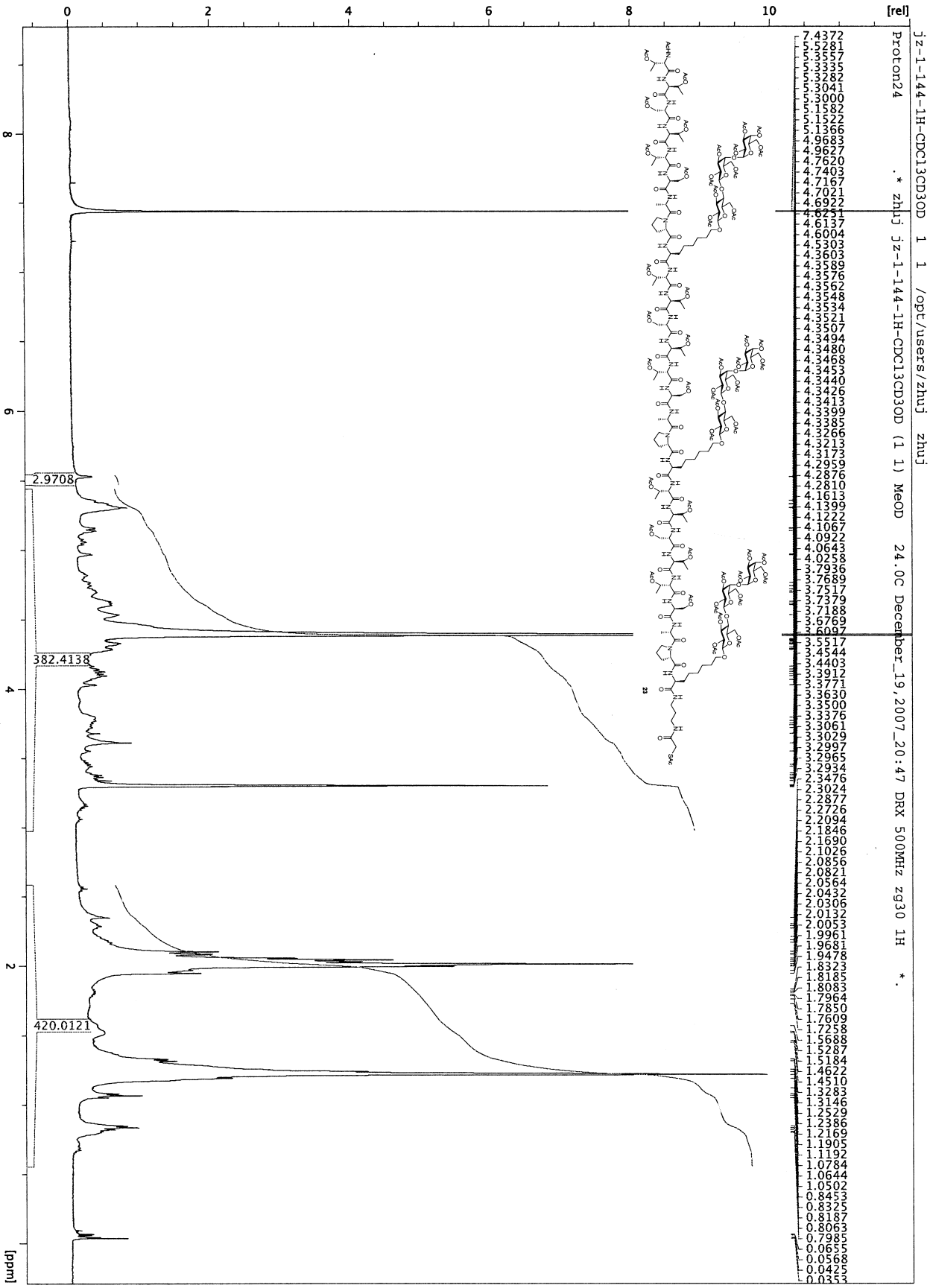

22
C178H305N29O96
Exact Mass: 4384.9876
Mol. Wt.: 4387.4630
AcHN
NHBoc

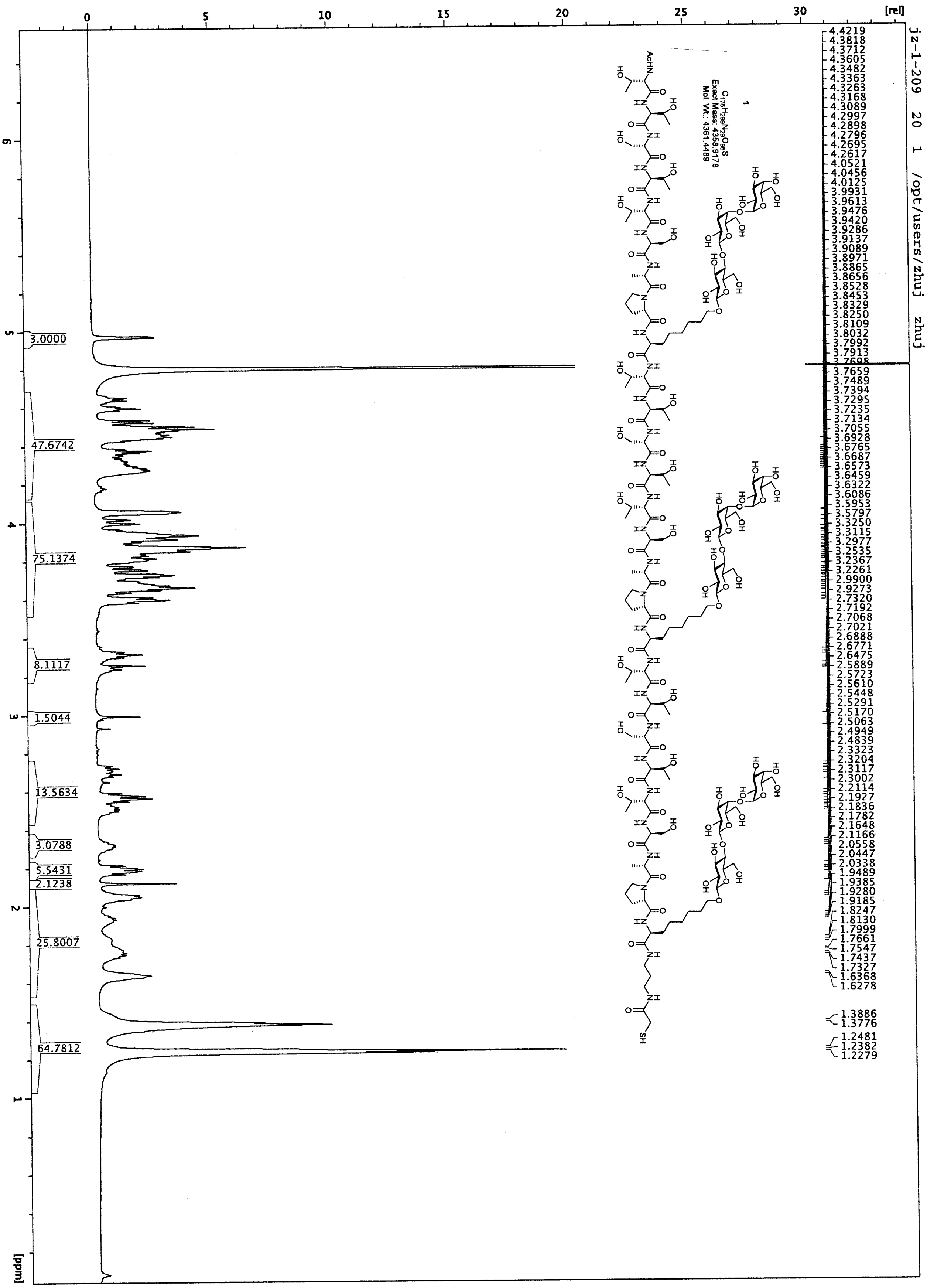

+Q1: from JZ-I-231 C178H305N29O96 4384.98
5.33e5 cps
Positive Ion
1485.4
1119.8
1095.3
1452.0
1882.6
2217.2
Intensity, cps
4e5
2e5
1000
1500
2000
m/z, amu


+Q1: from JZ-I-231 C178H305N29O96 4384.98, subset
5.33e4 cps
Positive Ion (expansion)
2217.2
[M+2Na]++
2216.0
2215.5
2172.8
2205.3
2197.0
2256.2
Intensity, cps
40000
20000
2180
2200
2220
2240
m/z, amu

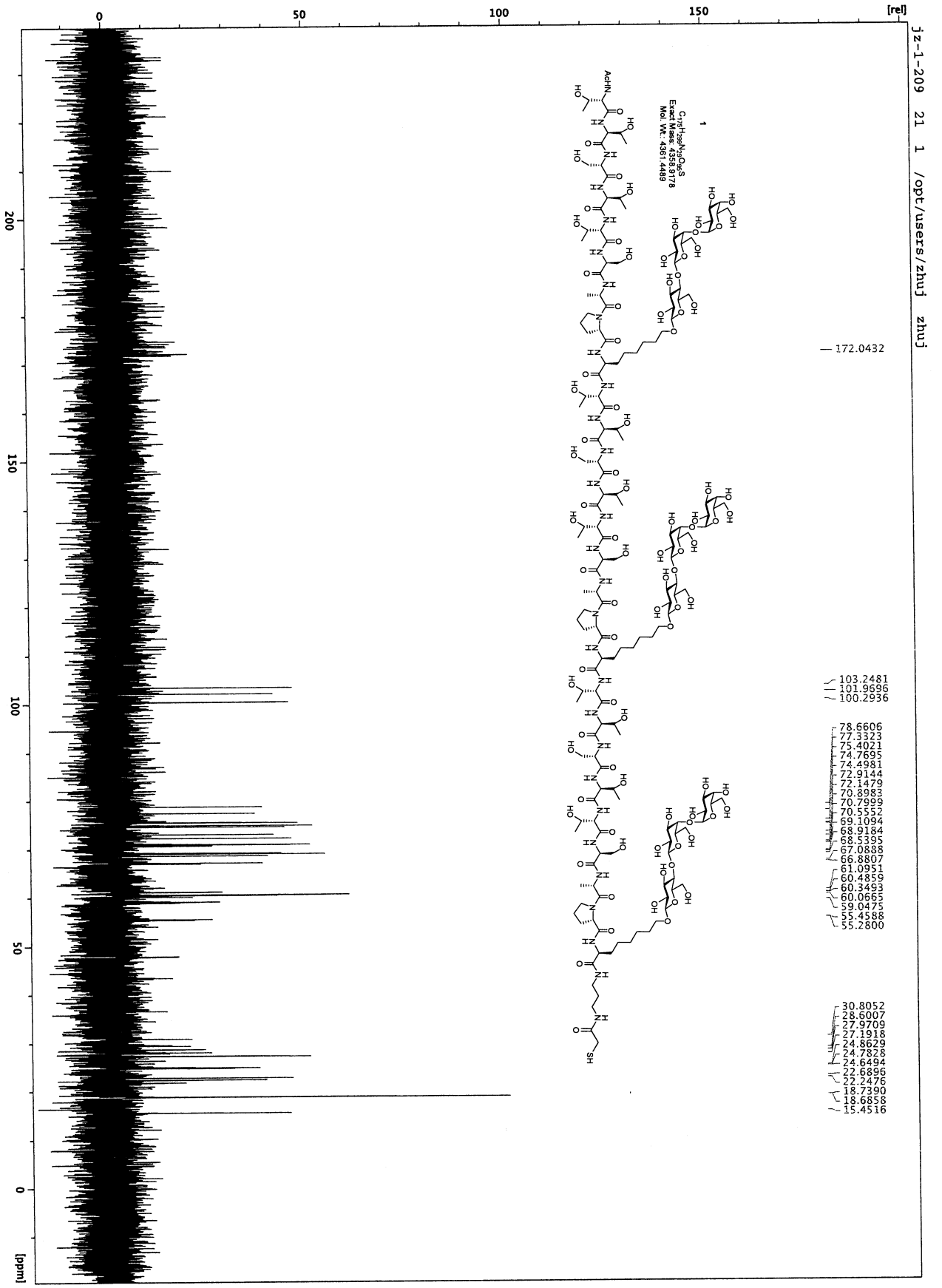

+Q1: from JZ-I-231 C178H305N29O96 4384.98, subset
5.33e5 cps
Positive Ion (expansion)
1485.4
[M+3Na]3+
1484.9
1452.0
1481.0
1494.1
1430.9
1519.0
1531.1
Intensity, cps
4e5
2e5
1440
1460
1480
1500
1520
m/z, amu

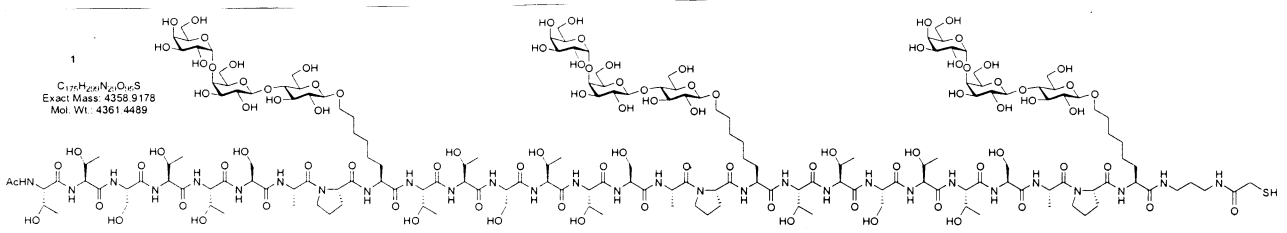

+Q1: from JZ-I-231 C178H305N29O96 4384.98, subset
3.53e5 cps
Positive Ion (expansion)
[M-Boc+4Na]4+
1095.3
1094.7
1119.8
[M+4Na]4+
1119.2
1126.5
1054.3
1079.6
1146.4
1158.0
1173.2
Intensity, cps
3e5
2e5
1e5
1100
1150
m/z, amu

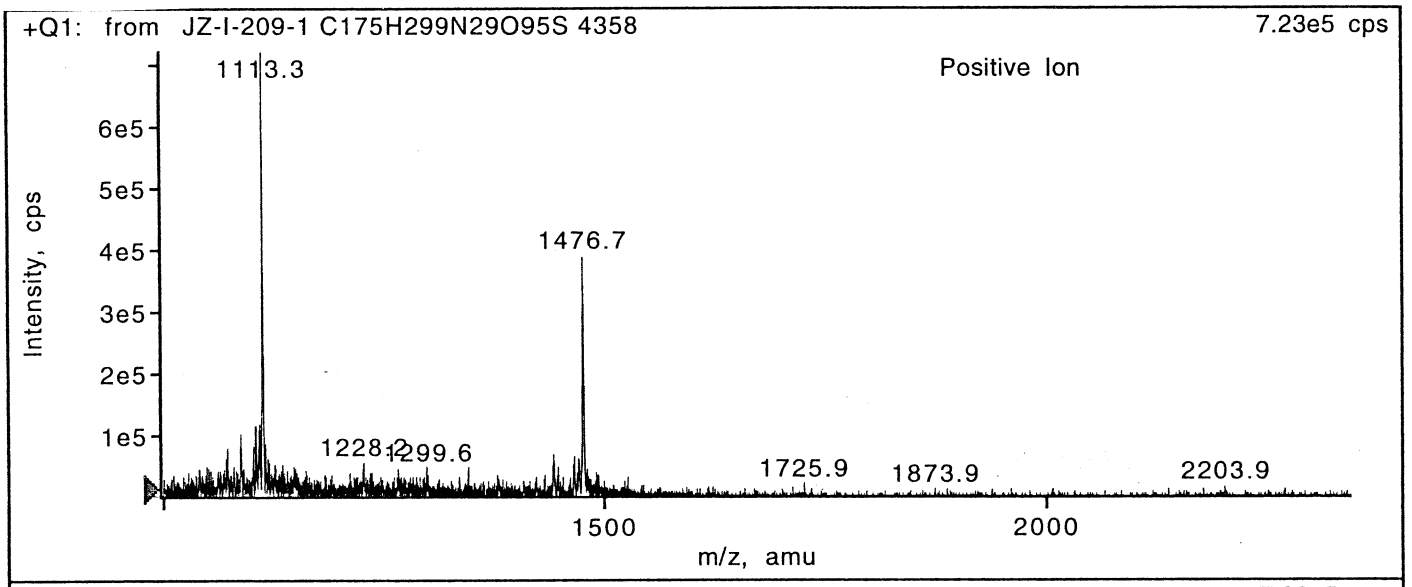

jz-1-144-1H-CDCl3CD3OD 1 1 /opt/users/zhuj zhuj
Proton24 .* zhuj jz-1-144-1H-CDCl3CD3OD (1 1) MeOD 24.0C December_19,2007_20:47 DRX 500MHz zg30 1H *.
[rel]
[ppm]
2.9708
382.4138
420.0121

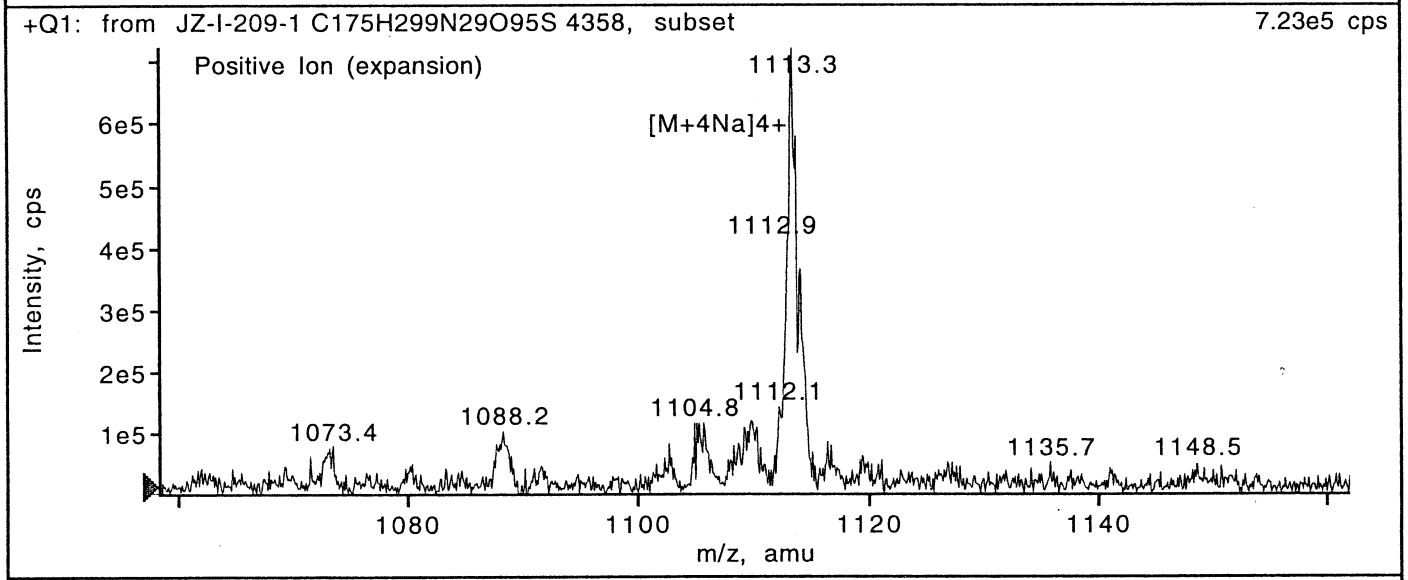


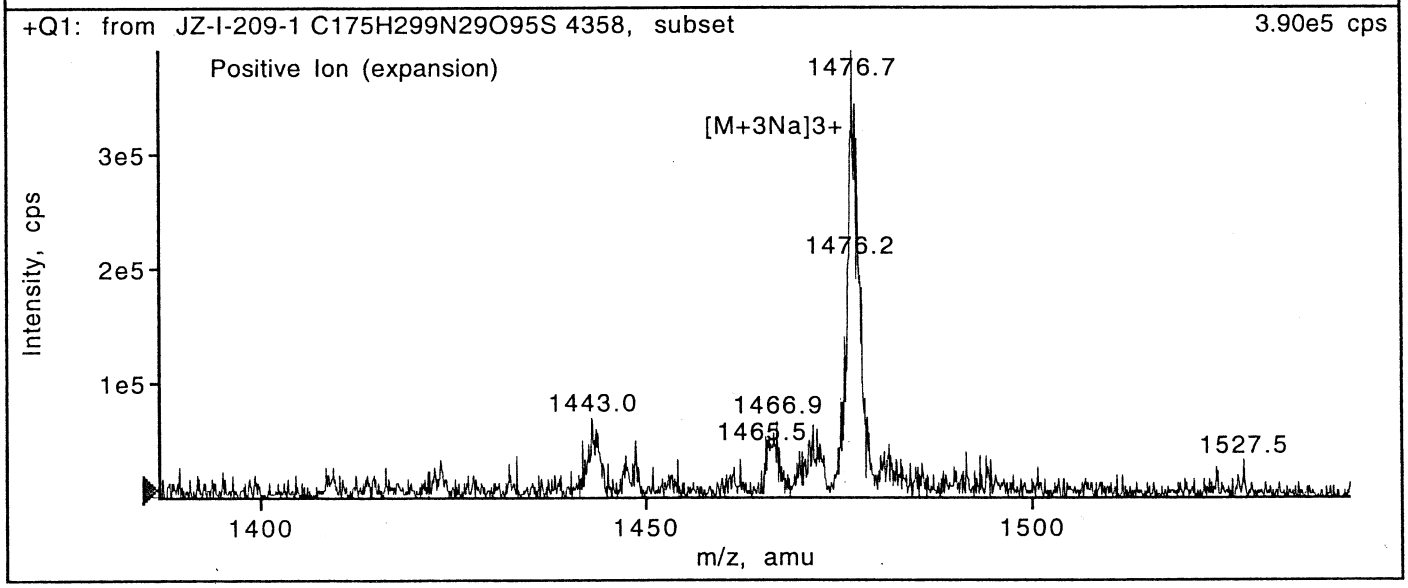
jz-1-209 20 1 /opt/users/zhuj zhuj
1
$C_{175}H_{299}N_{29}O_{85}S$
Exact Mass: 4358.9178
Mol. Wt.: 4361.4489
3.0000
47.6742
75.1374
8.1117
1.5044
13.5634
3.0788
5.5431
2.1238
25.8007
64.7812
4.4219 4.3818 4.3712 4.3605 4.3482 4.3363 4.3263 4.3168 4.3089 4.2997 4.2898 4.2796 4.2695 4.2617 4.0521 4.0456 4.0125 3.9931 3.9613 3.9476 3.9420 3.9286 3.9137 3.9089 3.8971 3.8865 3.8656 3.8528 3.8453 3.8329 3.8250 3.8109 3.8032 3.7992 3.7913 3.7698 3.7659 3.7489 3.7394 3.7295 3.7235 3.7134 3.7055 3.6928 3.6765 3.6687 3.6573 3.6459 3.6322 3.6086 3.5953 3.5797 3.3250 3.3115 3.2977 3.2535 3.2367 3.2261 2.9900 2.9273 2.7320 2.7192 2.7068 2.7021 2.6888 2.6771 2.6475 2.5889 2.5723 2.5610 2.5448 2.5291 2.5170 2.5063 2.4949 2.4839 2.3323 2.3204 2.3117 2.3002 2.2114 2.1927 2.1836 2.1782 2.1648 2.1166 2.0558 2.0447 2.0338 1.9489 1.9385 1.9280 1.9185 1.8247 1.8130 1.7999 1.7661 1.7547 1.7437 1.7327 1.6368 1.6278 1.3886 1.3776 1.2481 1.2382 1.2279
[ppm]
[rel]


jz-1-209 21 1 /opt/users/zhuj zhuj
[rel]
[ppm]
1
C176H288N36O96S
Exact Mass: 4358.8178
Mol. Wt.: 4361.4489
172.0432
103.2481
101.9696
100.2936
78.6606
77.3323
75.4021
74.7695
74.4981
72.9144
72.1479
70.8983
70.7999
70.5552
69.1094
68.9184
68.5395
67.0888
66.8807
61.0951
60.4859
60.3493
60.0665
59.0475
55.4588
55.2800
30.8052
28.6007
27.9709
27.1918
24.8629
24.7828
24.6494
22.6896
22.2476
18.7390
18.6858
15.4516


1
C175H299N29O95S
Exact Mass: 4358.9178
Mol. Wt.: 4361.4489
AcHN
SH


+Q1: from JZ-I-209-1 C175H299N29O95S 4358
7.23e5 cps
Positive Ion
1113.3
1476.7
1228.2
1299.6
1725.9
1873.9
2203.9
Intensity, cps
6e5
5e5
4e5
3e5
2e5
1e5
1500
2000
m/z, amu


+Q1: from JZ-I-209-1 C175H299N29O95S 4358, subset
7.23e5 cps
Positive Ion (expansion)
1113.3
[M+4Na]4+
1112.9
1112.1
1104.8
1088.2
1073.4
1135.7
1148.5
Intensity, cps
6e5
5e5
4e5
3e5
2e5
1e5
1080
1100
1120
1140
m/z, amu


+Q1: from JZ-I-209-1 C175H299N29O95S 4358, subset
3.90e5 cps
Positive Ion (expansion)
1476.7
[M+3Na]3+
1476.2
1443.0
1466.9
1465.5
1527.5
Intensity, cps
3e5
2e5
1e5
1400
1450
1500
m/z, amu