Carbohydrates as Scaffolds

For Bioactive Agents

by

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ABSTRACT

Carbohydrates as Scaffolds for Bioactive Agents

By

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Carbohydrates are attractive templates for drug design because of their accessibility, highly functionalized structures and rich synthetic chemistry. The goal of this research was to design mimetics of two classes of biologically interesting molecules using carbohydrate scaffolds. These are β-D-galactosylceramide (GalCer) and the tetrahydrofuran (THF) containing AAs.

The emergence of multi-drug resistant (MDR) strains of HIV-1 has created a need for new therapeutic agents. The glycolipid GalCer has been shown to be a cofactor in HIV-1 infection as it mediates the binding of the HIV envelope protein gp120 in $CD4^+$ cells. Mimics of GalCer can serve as potential entry inhibitors of HIV-1. 1,1-Linked galactose-mannose (Gal-Man) and glucose-mannose (Glu-Man) disaccharides with an ester on the Man subunit were found to bind to the V3 loop peptide of gp120 and inhibit HIV infectivity in single round infection assays with the TZM-bl cell line (a derivative of the HeLa cell line that express CD4, CXCR4, and CCR5). IC_{50} values were in the 50 μM range with no toxicity to the cells at concentrations up to 200 μM. These compounds appear to inhibit virus entry at early steps in viral infection since they were inactive if added post viral entry. Although these compounds were found to bind to the V3 loop peptide of gp120, it is not clear that this interaction is responsible for their anti-HIV activity because the binding affinity of closely related analogs did not correlate with their

antiviral behavior. The low cytotoxicity of these 1,1-linked disaccharide fatty acid esters, combined with their easy accessibility to structurally diverse analogs, make these molecules attractive leads for new anti-viral agents.

The THF-containing AAs have drawn much attention because of their potent antitumor activities. Their mode of action involves the inhibition of the NADH: ubiquinone oxidoreductase, Complex I, of the mitochondrial electron transport chain. Their generally high cytotoxicity to both normal and tumor cells has hampered their development as anti-cancer agents. Thus acetogenin analogs that show increased specificity towards cancer cells are of interest as new therapeutic agents. Acetogenin analogs in which the THF core was replaced with either a monosaccharide or disaccharide framework were synthesized and evaluated against various cancer cell lines. The monosaccharide analogs showed antitumor activity in the low micromolar range and were generally more active than their disaccharide counterparts. It is also noteworthy that varying the degree of oxygenation on the monosaccharide ring did not show any significant effect on cytotoxicity. These structure activity observations open up possibilities for the design of tumor selective monosaccharide analogs that target carbohydrate receptors that are overexpressed on tumor cells.

Acknowledgements

The road to a doctoral degree is often paved with uncertainty. However, with the help of the certain individuals you meet along the way, the journey becomes less troublesome. Without these individuals your journey may never be complete. My sincerest thanks go to Professor David. R. Mootoo for his immeasurable patience and excellent guidance throughout this process. I appreciate it more than words can express. I also extend my deepest gratitude to Professor Akira Kawamura and Professor Robert Bittman for serving on my thesis committee.

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TO MY LOVING PARENTS

HARRY and ROSEY BACHAN

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List of Symbols and Abbreviations

AA	annonaceous acetogenins
Ac	acetyl
Ac_2O	acetic anhydride
Bn	benzyl
Brsm	based on recovered starting material
Bs	broad singlet
°C	degree celsius
calcd	calculated
CSA	camphorsulfonic acid
^{13}C NMR	carbon-13 nuclear magnetic resonance spectrometry
CyH	cyclohexane
δ	chemical shift in parts per million
d	doublet
DCC	dicyclohexylcarboiimide
DCM	dichloromethane
DIB	iodobenzene diacetate
DMAP	4-dimethylaminopyridine
DMF	N,N-dimethylformamide
EVE	ethyl vinyl ether

FCC	flash column chromatography
Gal	D-galactose
h	hour
^{1}H NMR	proton nuclear magnetic resonance spectroscopy
HRMS	high resolution mass spectroscopy
Hz	hertz
IC_{50}	half maximal inhibitory concentration
Imid	imidazole
J	coupling constant in Hertz
LAH	lithium aluminum hydride
Man	D-mannose
min	minute
MS	molecular sieves
Ph	phenyl
ppm	parts per million
PPTS	pyridinium *p*-toluenesulfonate
Piv	pivaloyl (trimethylacetyl)
Py	pyridine
q	quartet
rt	room temperature
s	singlet

SAR	structure activity relationship
t	triplet
TBAI	tetrabutylammonium iodide
TBDPS	*tert*-butyldiphenylsilyl
TEA	triethylamine
TES	triethylsilyl
Tf	trifluoromethanesulfonate
TFA	trifluoroacetic acid
THF	tetrahydrofuran
THP	tetrahydropyran
TIPS	triisopropylsilyl
TLC	thin layer chromatography
Tol	toluene
TPP	triphenylphosphine
Trityl	triphenylmethyl
Ts	toluenesulfonyl

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Chapter 1

Carbohydrates in Drug Design

A very active area in drug design is the identification of molecular scaffolds for the attachment of pharmacophoric components in well-defined orientations. [1] Therefore, scaffold design is generally guided by structural information of a specific recognition event, such as, obtained from NMR, binding, and crystallographic studies. When detailed conformational information is available, scaffolds can be designed to mimic the natural binding arrangement of the substituents. Unfortunately, conformational requirements for binding are often not clear, thus making scaffold selection a challenge. In this vein, scaffolds that can be easily obtained and modified to produce structurally diverse compound libraries for combinatorial screening strategies are of much interest.[2]

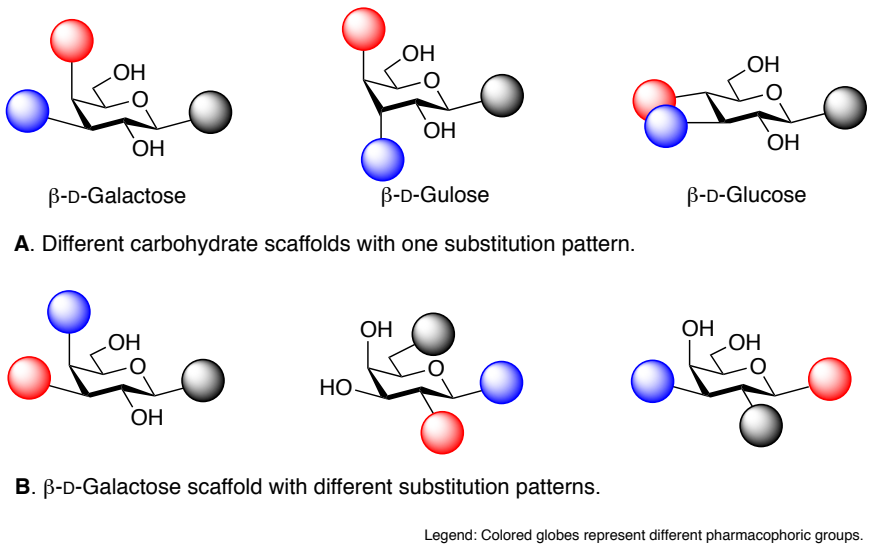


Figure 1: The difference in spatial orientations of the substituents on different carbohydrate scaffolds. (Adapted from ref. 3)

Carbohydrates are unique templates for deriving diverse compound libraries. They are relatively stable to physiological pH and metabolism, once the anomeric (hemiacetal) center has been converted to the glycoside. The availability of naturally occurring stereoisomeric sugars allows for wide stereochemical diversity.[3,4] Their relatively rigid cores comprising several alcohol groups can be selectively modified through established and straightforward synthetic methodologies (Figure 1).

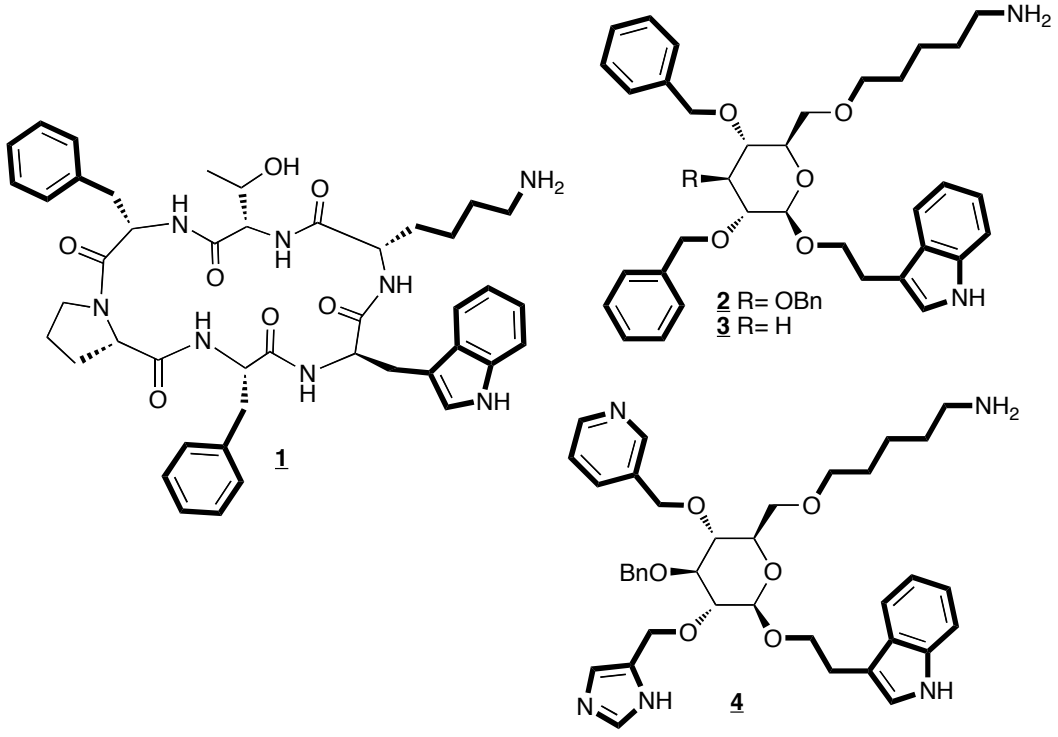


Figure 2: Carbohydrates as SRIF mimics. (Adapted from ref. 5)

Hirschmann and co-workers, in their development of novel somatotropin release inhibiting factor- (SRIF) analogs, illustrated the utilization of carbohydrates as three-dimensional templates in drug design (Figure 2).[5] The cyclic peptide **1** is a potent

somatostatin (SST) agonist with an IC_{50} in the nanomolar (nM) range. The D-glucose derivatives **2** and **3** were the first set of analogs designed to mimic the active conformation of compound **1.** NMR spectroscopy and molecular modeling experiments provided information about the relative positioning of the substituents. Compounds **2** and **3** showed IC_{50} values of 9.5 and 1.3 μM respectively as SST agonist. In a more recent study by the same group the 4-benzyl group was replaced with heterocyclic moieties, such as imidazole, pyrazine, and pyridine. These compounds were tested for their affinity toward the somatostatin receptor. Compound **4** yielded the best result, with an IC_{50} of 53 nM against SSTR4. [6]

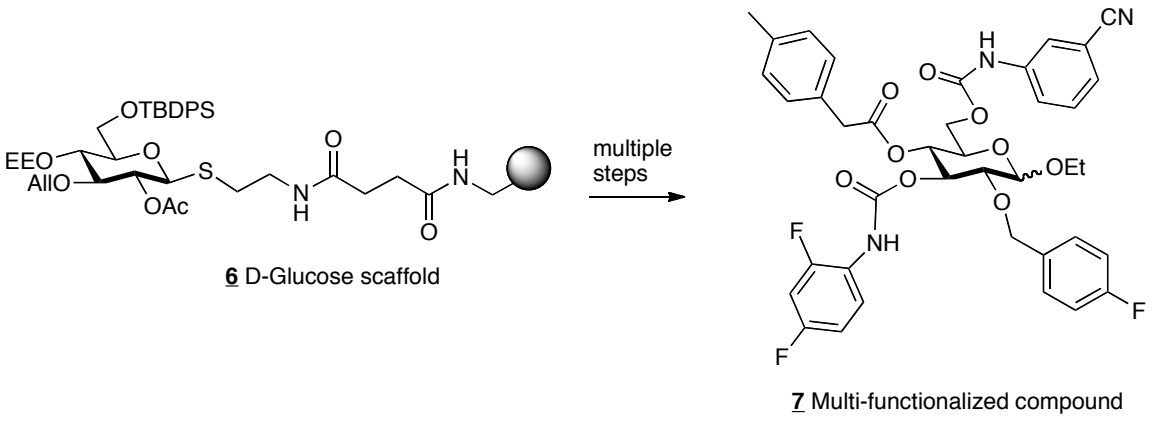


Figure 3: D-Glucose derived template used in combinatorial synthesis, (Adapted form ref. 7).

Combinatorial chemistry has accelerated the identification of active compounds in both pharmaceutical and agro-chemical industries. Kunz and co-workers have utilized monosaccharide scaffolds, namely, D-glucose, and 2,6-diaminoglucose in a novel application of combinatorial synthesis (Figure 3). [7, 8] These monosaccharides were decorated with orthogonal protecting groups, which allowed for selective installation of

desired pharmacophoric residues under mild conditions (*cf.* 6 ➔ 7). The scaffolds were then anchored to a resin for solid-phase synthesis, which increases their utility in combinatorial synthesis.

The goal of this thesis research is to design mimics of two classes of biological molecules, the glycolipid β-D-galactosylceramide (GalCer) and the tetrahydrofuran (THF)-containing acetogenins, using carbohydrate scaffolds. At the end of 2008, it was estimated that thirty-three million people were living with human immunodeficiency virus (HIV) and more are being infected daily. GalCer has been shown to mediate viral entry into host cells, and is a target in the development of new HIV treatments. The THF-containing acetogenins, isolated from the *Annonaceae* family of tropical plants, display potent antitumor activity. The synthesis and biological evaluation of these analogs will be presented in two parts.

Chapter 2

Carbohydrate Based Anti-HIV Agents

2.0 The Human immunodeficiency virus (HIV).

The human immunodeficiency virus type 1 (HIV-1) is known to be the causative agent of the acquired immune deficiency syndrome (AIDS) and has claimed over twenty million lives worldwide, since the first cases were reported in 1981.[9] The continuing rise in the population of people living with HIV-1, approximately thirty-four million, is due to the combined effects of increased rate of new infections, as well as, the improvement in survival rates as a result of more effective antiretroviral therapy. There was a 20% increase in the total number of people living with HIV from the years 2000 to 2008. Also, approximately two million deaths occurred at the end of 2008, as a result of opportunistic, AIDS-related infections. In 2009, there were an estimated 2.6 million [2.3 -2.8 million] people who became newly infected with HIV. This is nearly one-fifth (19%) fewer than the 3.1 million [2.9-3.4 million] people newly infected in 1999, and more than one fifth (21%) fewer than the estimated 3.2 million [3.0-3.5 million] in 1997, the year in which annual new infections peaked.

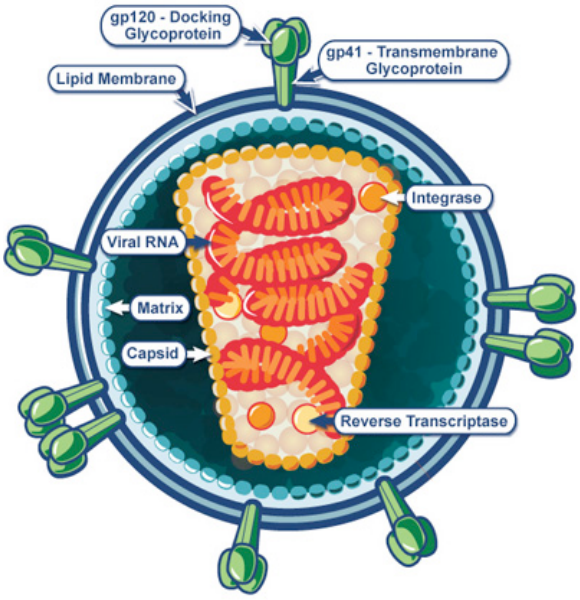


Figure 4: The HIV Virus (from niaid.NIH.gov).

HIV-1 is classified as a lentivirus, which is a subfamily of retroviruses with complex regulation of viral gene expression and replication (Figure 4). Unlike most retroviruses that preserve the host cell, HIV-1 is both cytopathic and cytotoxic with high levels of viral gene expression resulting in death of infected cells. Despite the unusual

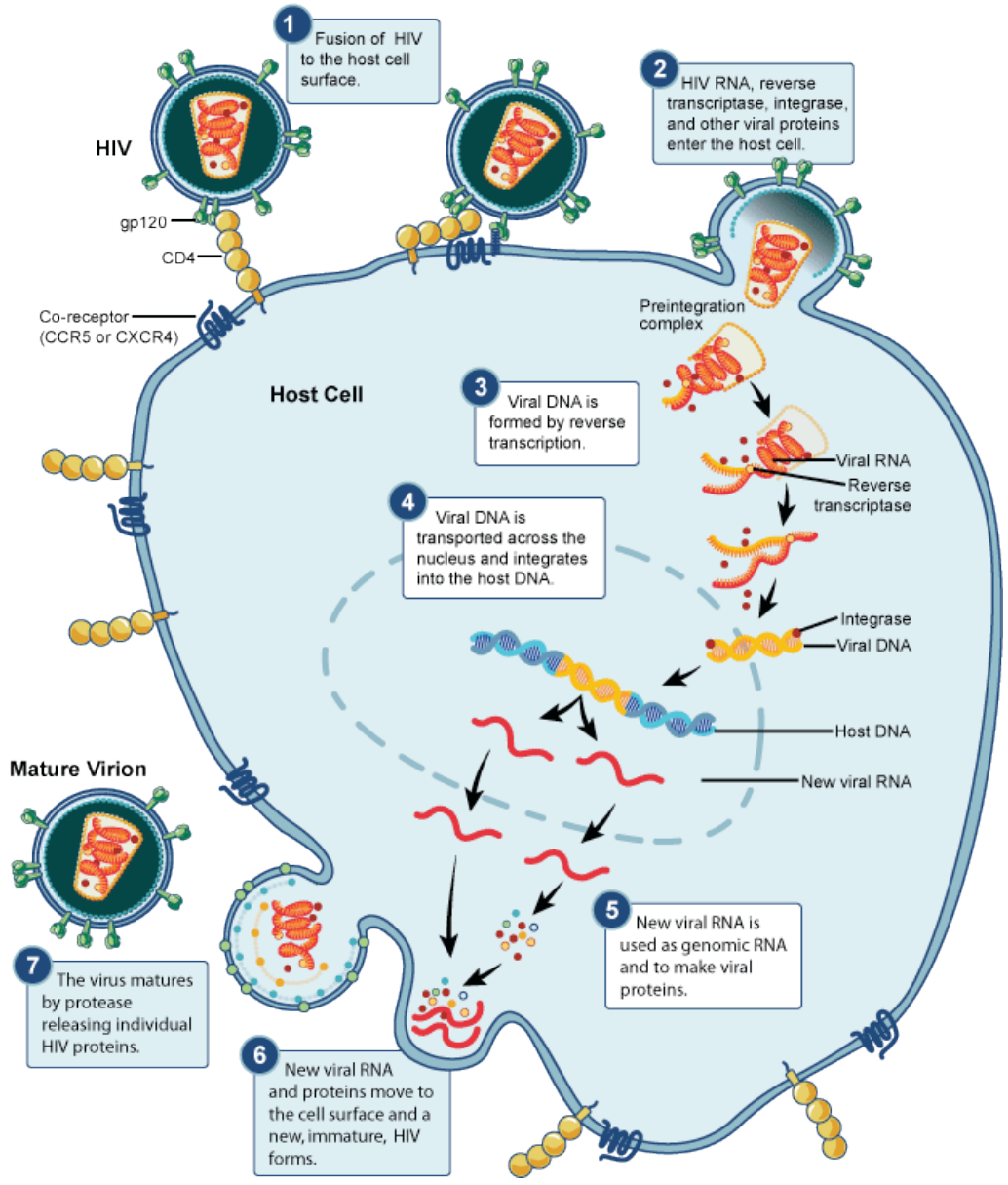


Figure 5: The HIV Replication Cycle (from niaid.nih.gov).

behavior of HIV, it is still a retrovirus, and as such its replication cycle is still comparable to that of most simple retroviruses.

The virion envelope glycoprotein (Env) consists of a complex between gp120 and the membrane-bound gp41 (Figure 4). Infection is initiated by attachment of gp120 to the CD4 receptor of T-helper cells. This results in a series of conformational changes within gp120 that allow for interaction with the chemokine co-receptors CCR5 or CXCR4 of the host cell (Figure 5). These changes in gp120 also trigger conformational changes in gp41, which results in fusion of the viral envelope with the cell membrane.[10,11,12]

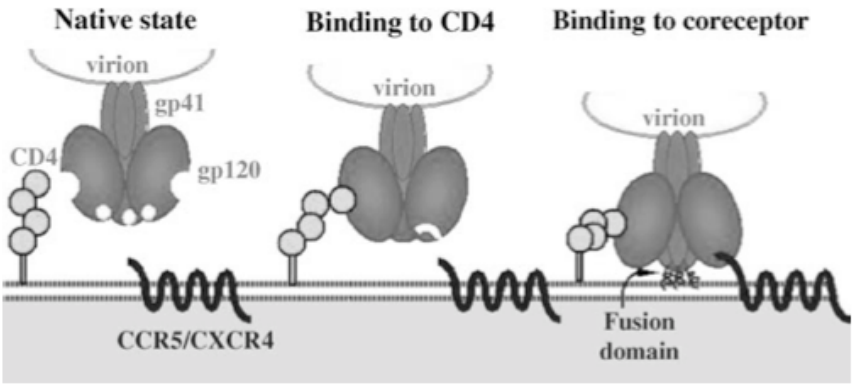


Figure 6: Model for virion attachment and entry (from ref. 13)

At the point of viral-host cell fusion, the enveloped proteins are lost, as the virus uncoats and its RNA is released into the host cell cytoplasm, where it is reverse transcribed into full-length, doubled-stranded DNA by viral reverse transcriptase (RT). The newly synthesized DNA is part of the pre-integration complex that is then transported into the nucleus, where it is inserted into the host chromosome by the action of viral integrase (IN). Once the proviral DNA is integrated, transcription of the viral DNA into mRNA begins. This is followed by translation of the mRNA to viral proteins,

using the host's cellular machinery. After translation, Env proteins migrate and insert into the cell membrane. Gag and Gag-Pol polyproteins also move to the cell membrane and start to assemble. The Gag polyprotein is a precursor that is proteolytically processed into six structural proteins, which rearrange and produce the mature virion. The Gag-Pol polyprotein includes the enzymatic viral proteins protease (PR), IN, and RT. Two copies of single-stranded, viral RNA are incorporated into the virion, which then buds from the cell, taking part of the host cell membrane. Viral protease cleaves the Gag and Gag-Pol polyproteins to produce a mature, functional virion.

2.1 Current HIV-1 treatment.

HIV drugs are classified according to the stage of the viral replication cycle that they target.[13] The Food and Drug Administration (FDA) has approved six classes of HIV drugs. These are nucleoside reverse transcriptase inhibitors (NRTIs), the non-nucleoside reverse transcriptase inhibitors (NNRTIs), protease inhibitors (PIs), fusion or entry inhibitors (FIs), co-receptor inhibitors (CRIs) and integrase inhibitors (INIs). These drugs can be combined in highly active anti-retroviral therapy (HAART) regimens to suppress viral replication and by extension, the progression of HIV infection to AIDS. There are, however, a wide variety of side effects experienced by patients on HAART that range in severity from skin rashes and gastrointestinal intolerance to coronary artery disease, kidney damage, and bone marrow suppression. A further complication from HAART is the emergence multi-drug resistant strains of HIV, which result from mutations that occur in the viral proteins targeted by antiretroviral agents.[14] These multi-drug-resistant viruses

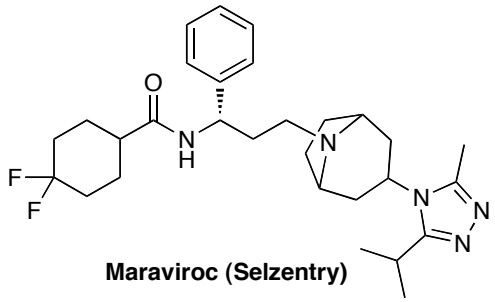
can then be transmitted to other individuals, making it more difficult to treat subsequent generations of HIV-infected individuals. In light of the focus of this thesis, a review on HIV entry inhibitors will be next presented.

2.2 FDA approved HIV entry inhibitors.

Multi-drug-resistant HIV strains provide new and difficult challenges to the treatment of infected individuals. New therapeutic options are needed for individuals as resistance to one treatment occur. HIV drugs based on inhibition of entry or fusion of the virus to the host cells are very appealing as an alternative to the present methods because they involve relatively new targets and therefore, are likely to be effective against HIV strains that are resistant to existing therapies. In addition, entry inhibitors provide distinct advantages over current therapeutic methods, in that they do not require cellular uptake to initiate the desired activity. Also, since this class of drugs act at such an early stage of the viral replication cycle, they can be used both as a preventative and a post-infective course of treatment. Clearly entry inhibitors have great potential as anti-HIV drugs. The FDA to date has approved only two entry/fusion inhibitors, thus leaving this area of research largely underexplored.

Enfuvirtide (Fuzeon) was the first FDA-approved HIV entry inhibitor.[15] As discussed earlier, the HIV replication cycle is initiated by binding of the HIV envelope protein gp120 to CD4 receptor of host cells. This is an ordered, multistep process that leads ultimately to the fusion of viral and cell membranes. Enfuvirtide is a 36-residue synthetic peptide (YTSLIHSKIEESQNQQEKNEQELLELDKWASLWNWF) that

mimics components of the HIV-1 fusion machinery and prevents normal fusion. We know that HIV binds to the host $CD4^+$ cell receptor via the viral protein gp120, resulting in a conformational change in gp41, the viral transmembrane protein, which assists in the fusion of the viral membrane to the host cell membrane. Enfuvirtide binds to gp41 and prevents the creation of an entry pore for the viral capsid. Maraviroc (Selzentry) was approved as an HIV drug in 2007 and is a selective, slowly reversible, small molecule antagonist of the interaction between human CCR5 and HIV-1 gp120 (Figure 7). In binding to the chemokine receptor CCR5, it blocks the HIV gp120 (V3 loop) from associating with the receptor. HIV is then unable to bind and enter human macrophages.[16]

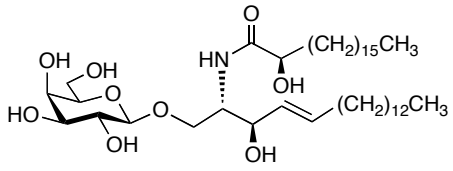


Maraviroc (Selzentry)

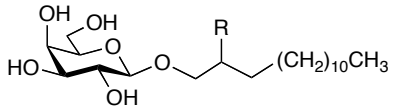
Figure 7: FDA Approved entry inhibitor.

2.3 Analogs of GalCer as HIV entry inhibitors.

Galactose-containing cell surface glycolipids such as GalCer **8** and globotriaosylceramide (Gb_3) are known to facilitate HIV binding to both $CD4^+$ as well as $CD4^-$ cells. More specifically, GalCer has been shown to be an important cofactor in the binding of gp120 to CD4 in a binding event that precedes gp120-CD4 interaction, as determined by kinetic and temperature arrested state experiments.[17,18,19] Thus molecules that inhibit the interaction between gp120 and GalCer are of interest as



8 β-Galactosylceramide



9a R = H
9b R = $(CH_2)_{10}CH_3$

Figure 8: GalCer and Analogs.

entry inhibitors of HIV.[20,21] GalCer analogs containing simple ceramide substitutes were have shown gp120 binding and antiviral activity.[22] Lead compounds, represented by **9** bound gp120 with similar affinity to GalCer and showed significant activity against HIV Env mediated fusion (Figure 8). These studies also suggested that these compounds act at an early step in HIV Env binding that precedes CD4 binding.

The goal of the present study was to design a new class of GalCer analogs that is based on a disaccharide template. These structures are guided by existing SAR data that suggests the central polar head region of the ceramide residue acts as a scaffold, which maintains a specific, relative orientation of the contacts in the sugar ring and the lipid chains.[23] The galactose C4-hydroxyl group is believed to be a key contact in binding of GalCer to the V3 loop of gp120, as glucose derivatives with the C4-hydroxyl in an equatorial position are inactive.[24] It is also possible that the interaction between GalCer and gp120 is not a 1:1 association but involves the binding of gp120 to a multivalent assembly of GalCer monomers, and that simpler analogs of GalCer (with less complex ceramide substitutes) may aggregate and bind gp120 in a similar way.[25]

We argued that β-Gal disaccharides, such as **10**, **11** and **12**, with one or two fatty acid esters in the mannose segment, may act as GalCer mimics (Figure 9). The β-galactose residue is preserved as in GalCer, while the mannose ring serves as a replacement of the polar region of the ceramide in GalCer. It should be noted that galactose-containing disaccharides have been previously suggested as mimics of GalCer. Galβ1-4GlcNAcβ1 has been hypothesized to mimic the gp120 binding of GalCer on T-cells. The 1,1-linked disaccharide template was selected because of its well-defined conformational bias in the intersaccharide linker. Mannose was used because of synthetic

considerations relating to stereoselective construction of the glycosidic linkages and alcohol group differentiation. The β-gluco-disaccharide **13** and the α-Gal-disaccharide **19** were designed to probe the specificity of the substitution pattern on the β-galactose residue, and the monosaccharide analogs **14** and **15** were synthesized to probe the effects of completely deleting the β-galactose residue. The deprotected disaccharide **16** and the alkyl ether derivative **17** are control compounds to determine whether activity is connected to ester hydrolysis. Compound **18**, which has the fatty acid ester at C-6 of the galactose residue, was designed as a control compound to test the specificity with respect to the location of the ester.[26] Since simple fatty acid esters like GML **20** are active against other envelope viruses such as herpes-simplex virus 2 and bacteriophage φ6, GML was selected as a reference to compare the anti-HIV activity of simple fatty acid esters and the new disaccharide esters.[27]

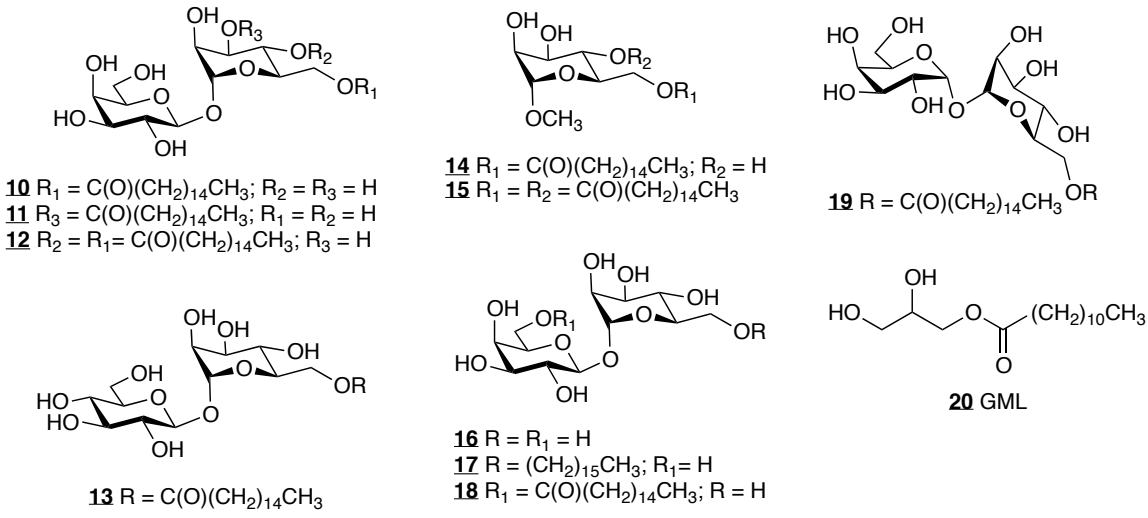
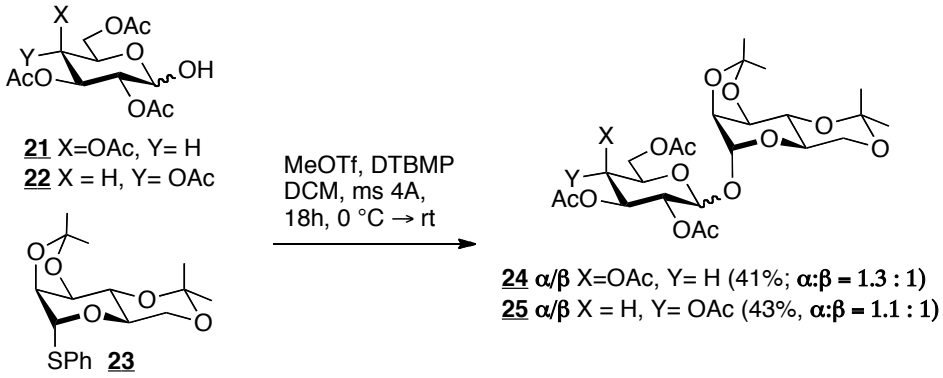


Figure 9: Proposed GalCer mimics and control compounds.

2.4 Synthesis of disaccharide analogs.

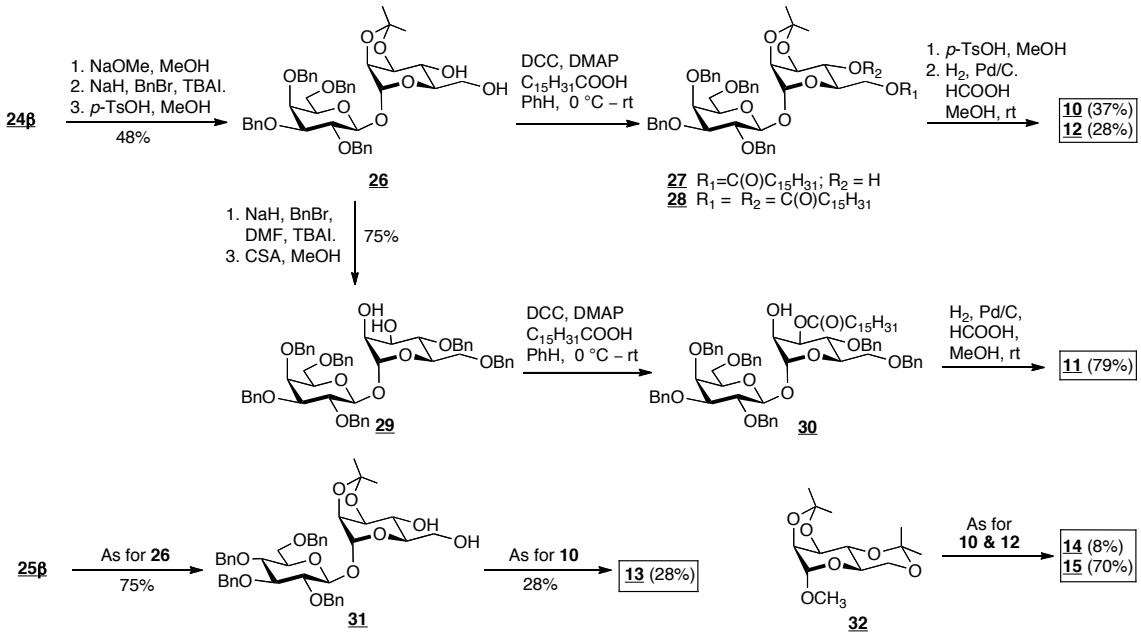
The synthesis of the disaccharide analogs **10-13** was initiated with the construction of the 1,1-linked precursors **24β** and **25β** (Scheme 1). Initial attempts to synthesize this disaccharide scaffold with 2-*O*-acetylated galactosyl donors and mannose



Scheme 1: Disaccharide synthesis.

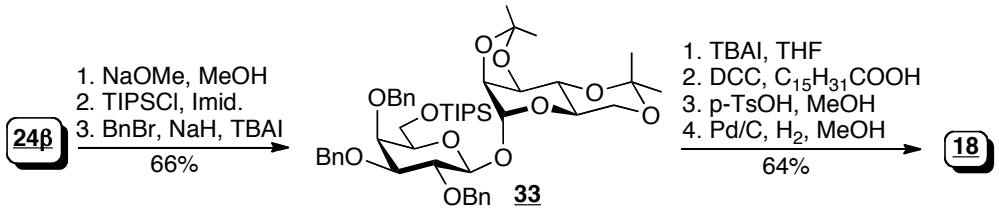
acceptors provided undesired products or very low yields of the desired product. This problem was solved by the methyl triflate (MeOTf) promoted glycosidation of thioglycoside mannosyl donor **23** with galactose and glucose acceptors **21** and **22**, respectively (Scheme 1). Although this strategy yielded anomeric mixtures of the disaccharide products, they were easily separated by column chromatography. The phenylthio glycoside donor **23** was obtained from the tetra-*O*-acetate derivative by using the procedure for the corresponding ethylthio glycoside.[28] The reaction of **23** with acceptors **21** and **22** afforded in each case an approximately 40% yield of α:β galacto disaccharides **24α:β** and **25α:β**, with the desired β-anomer obtained as the minor product (α:β, ca 1.3:1). The stereochemistry at the anomeric position of the *galacto* and *gluco* rings were determined via the vicinal $J_{H,H}$ values [**24α/β**: $J_{1,2}$ (Gal) = 3.5/8.0 Hz; **25α/β**: $J_{1,2}$ (Glu) = 3.8/8.0 Hz]. The α-configuration of the mannose ring was assigned via $J_{C,H}$ values obtained for proton-coupled ^{13}C NMR experiments [**24α/β**: $J_{1,1}$ (Man) = 175/172

Hz; **25α/β**: $J_{1,1}$ (Man) = 175/172 Hz].[29] The tetra-*O*-acetate **24β** was converted to the tetra-*O*-benzyl ether using standard protecting group protocols. The 4,6-*O*-isopropylidene in the product was selectively removed under mildly acidic conditions to provide diol **26** (Scheme 2). DCC-mediated esterification of **26** with one equivalent of palmitic acid gave monoester **27** as the major product. Using excess palmitic acid under similar conditions afforded diester **28**. Acid hydrolysis of **27** and **28** followed by individual hydrogenolysis provided the mono and di-acylated targets **10** and **12**, respectively. Diol **26** was transformed to diol **29** using routine protecting group manipulations. Selective acylation of **29**, followed by hydrogenolysis of the product gave the target compound **11**. Analog **13** was obtained in similar fashion as compound **10**, starting with disaccharide **25β**.



Scheme 2: Esterification and protecting group sequences.

Monosaccharide analogs **14** and **15** were easily obtained from methyl mannopyranoside **32** utilizing similar chemical transformations as in the syntheses of compounds **10** and **12**. The unprotected disaccharide **16** and the ether **17** were prepared from diol **26** by using straightforward alcohol derivatization reactions. Starting from compound **24α**, compound **19** was made using the same methodology as analog **10**. The synthesis of compound **18** started with deacetylation of **24β** and selective protection of the 6-hydroxyl on the galactose ring with TIPS. Standard benzylation was performed to protect the remaining hydroxyl groups. The silyl ether was removed with TBAF to afford the corresponding primary alcohol, which was then esterified using DCC and palmitic acid. Acid hydrolysis followed by hydrogenolysis provided compound **18** in reasonable yield (64%). Known compound **20** (GML) was obtained in two steps from commercially available solketal via esterification with lauric acid.[30]



Scheme 3: Synthesis of compound 18.

2.5 Results and discussion.

2.5.1 Binding to V3 loop peptide of gp120 to the GalCer analogs.

Please note: Peptide binding and critical micelle concentration (CMC) measurements were performed by Jacques Fantini, Professor of Biochemistry, Laboratoire de Biochimie et Physicochimie des Membranes Biologiques, Institut Méditerranéen de Recherche en Nutrition, Marseille, France.

The binding of **10** - **15** to a synthetic V3 loop peptide, RIQRGPGRAFVTIGK, corresponding to the R15K peptide, from gp120 of HIV-1 IIIb isolate was evaluated. The R15K peptide has been previously used as a model for the glycolipid binding domain of gp120. [31] Thus, the change in surface pressure ($\Delta\pi$) at the air-water interface of the glycolipid monolayer on exposure to an aqueous solution of the R15K peptide (10 μM) was measured. An increase in surface pressure is associated with integration of the peptide into the glycolipid monolayer and can be used as a measure of binding affinity. The critical pressure of insertion (CPI), calculated by extrapolation for a null increase in surface pressure, was in the range of 18-49 mN/m. All the compounds tested interacted with the R15K V3 peptide with CPI values in the range of 18 - 49 mN/m (Figure 10). These values were comparable to that measured for GalCer (CPI = 22-25 mN/m). Given the structural similarity of **10** - **12**, and the similarity of their CPI values to GalCer, it is tempting to conclude that these disaccharides may be acting as mimics in their binding to the V3 peptide (i.e. they interact with the GalCer domain of the peptide). However, in the absence of additional data this notion is highly speculative. It is also noteworthy that the monosaccharides **14** and **15** appear to bind appreciably more strongly to the V3 peptide than the disaccharides. This may be a consequence of their simpler structures, which

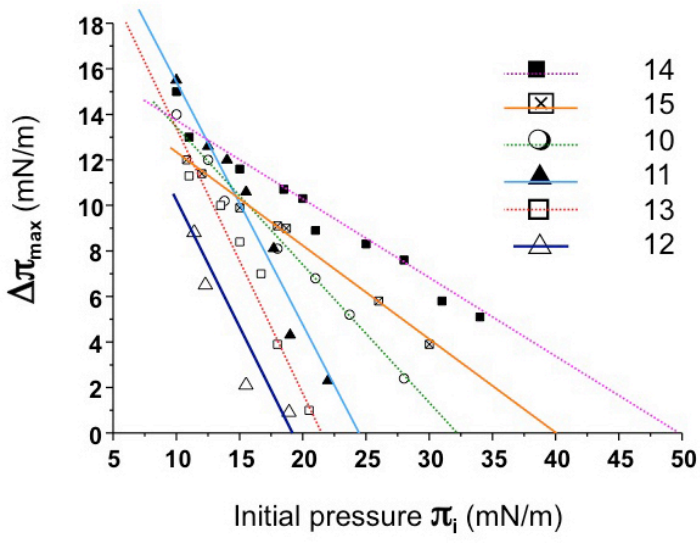


Figure 10: V3 Loop binding of glycolipid analogs.

present less steric demands to binding, although for the same reason, the binding of these structures could be less specific than that of the disaccharides. With the possibility that the test compounds could be interacting with the GalCer domain on the V3 peptide, and in light of the aforementioned hypothesis that GalCer inhibition of HIV is connected to its interaction with this peptide, the next step was to evaluate the anti-HIV activity of the test compounds.

2.5.2 Anti-HIV activity of the GalCer analogs.

Please note: These experiments were performed by Dr. Himanshu Garg, Center for Excellence for Infectious Disease, Texas Technical University Health Sciences Center, El Paso, Texas. Dr. Garg also performed the pre- and post- infection and cytotoxicity assays.

The anti HIV activity of **10** - **20** was next evaluated in a virus infection assay using TZM-bl indicator cell line infected with CXCR4 tropic HIV (NL-Lai).[32] The β-Gal-disaccharide **11**, the β-Glu-disaccharide **13**, and the α-Gal-disaccharide **19** with the ester moiety at *O*-3, *O*-6, and *O*-6 of the mannose subunit, showed appreciable activity with IC_{50} values of 75, 53, and 48 μM, respectively, which are comparable to the activity of **9a** (44 μM) (Table 1). Of special note, the β-Gal-disaccharide **10** with the ester at *O*-6 of the mannose ring, was not active up to a concentration of 100 μM, but its ether derivative **17** showed similar activity as the more active compounds. The β-Gal disaccharide **18** with the ester at *O*-6 of the galactose ring, di-*O*-ester **12**, the monosaccharides **14** and **15**, the free disaccharide **16**, and GML also showed no significant activity up to 100 μM. Since not all the test compounds that bind the V3 peptide show significant anti-HIV activity, V3 loop binding affinity is not a reliable indicator of HIV activity.

This contrasting activity of individual compounds in the two assays is not particularly surprising since it is likely that anti-HIV activity requires binding to a specific domain of the peptide and not simply overall affinity, which is measured in the binding assay. In so far as anti-HIV activity may be due to GalCer mimicry, it may be argued that the anti-HIV disaccharide analogs **11**, **13**, **17**, and **19** interact with the GalCer binding domain on the V3 peptide but the monosaccharides bind non-specifically and more strongly to other domains. However, this argument does not explain why the disaccharide **10** (which is very similar in structure to **11** and **13**), and to a lesser extent **12**, are active in the binding assay but not in the anti-HIV assay. One explanation is that glycolipid presentation in the two assays is different, so that structural factors that control the morphology of the active glycolipid species conflict with those that affect glycolipid-peptide recognition, thereby skewing the data analysis. Of course, the alternative and arguably more obvious explanation for the contrasting behavior of individual test compounds in the two assays is that the mechanism for anti-viral activity is not at all connected to V3 loop binding. Notwithstanding the exact mechanism for anti-viral activity, the data from the anti-HIV assay suggests that a disaccharide framework (vs. monosaccharide) and one ester residue (vs. two), are important for anti-viral activity. That the β-galactoside **11**, the β-glucoside

Compound	IC_{50}
9a	43.5 ± 1.6
10	>100
11	76.0 ± 6.2
12	>100
13	52.5 ± 1.9
14	>100
15	>100
16	>100
17	48.4 ± 1.7
18	>100
19	50.7 ± 4.0
20	>100

Table 1: IC_{50} Values for GalCer analogs.

13, and the α-galactoside **19** were active but the β-galactosides **10** and **12** were not also suggests that the location of the ester is important, and that the optimal position for the ester may vary with the disaccharide framework. That the simple disscharide **16** was not active suggests that the mechanism of anti-HIV activity does not involve ester hydrolysis.

2.5.3 Pre- and post- infection assays of the more active GalCer analogs.

To determine whether the compounds affected the viral entry process or events that occur post-viral entry a simple time of addition experiment was conducted. TZM-bl cells were infected either in the presence of the compounds (100 μM) or added the compounds 4 h post-infection. T20, a inhibitor of gp41 mediated fusion and AMD3100, a CXCR4 antagonist that inhibits HIV Env mediated fusion and viral entry, were used as controls in these experiments.[33,34] All active compounds were able to inhibit virus infection if added at the same time as the virus (Figure 12). However, addition of the compounds 4 h post-infection failed to show significant inhibition. The same was true for the entry inhibitors T20 and AMD3100. The absence of activity when the compounds were added 4 h post-infection also suggest that they did not have a direct effect on cell viability or function and hence the inhibition is at the step of initial virus cell interaction. These results are similar to that previously observed for **9a**.[35] Hence the mechanism of action of these disaccharide lipids may be similar to **9a** and related monosaccharide analogs, but more detailed studies are needed to support this suggestion.

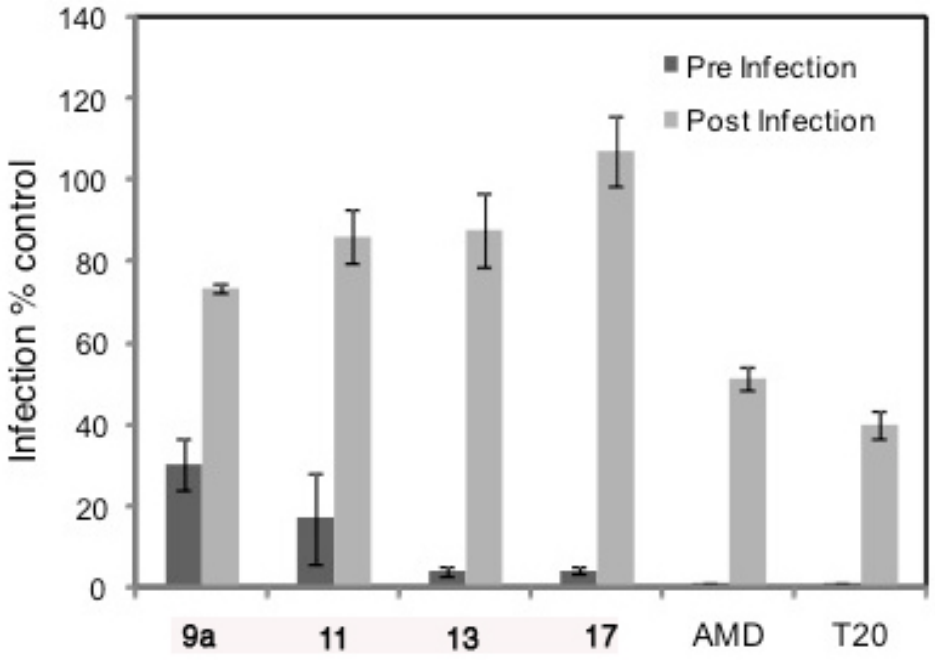


Figure 11: Glycolipid analogues show activity pre-infection only. TZM-bl cells were infected with NL-Lai virus. Compounds (100 μM) were added either prior to infection (pre- infection) or 4 h post-infection. Infectivity was determined 24 h later by measuring luciferase activity. AMD and T20 controls are used at 2 μM. Data are mean ± SD of triplicate observations.

2.5.4 Cytotoxicity assays

Cytotoxicity of the test compounds was determined in TZM-bl cells over a 24 h incubation period using the MTS dye reduction assay.[36] None of the compounds were toxic up to 200 μM dose in a 24 h incubation period.

2.5.5 Critical micelle concentration (CMC) of selected GalCer analogs.

The CMC values of selected analogs were measured in order to determine the physical state of the glycolipids at the concentrations used in the cellular assays. Values were obtained by tensiometer measurements of solutions of glycolipids in water, as previously described.[35] The CMC was taken as the concentration of glycolipid, which did

not induce any further decrease in surface tension. These data were obtained in triplicate: **10** (9.8, 10.0, and 10.5 μM); **11** (12.0, 12.0, and 12.5 μM); **13** (10.0, 10.5, and 12.0 μM); **14** (13.0, 13.5, and 13.7 μM). Thus at their active concentrations analogs **11** and **13** exist as micelles. The CMC values obtained for **10**, **11**, and **13** were in the range observed for related trehalose-type mono-fatty acid esters.[37, 38] The diester **15** was difficult to study; a regular decrease in surface tension from 72.8 mN/m (pure water) to 0 mN/m (glycolipid film collapse) was observed in the concentration range of 0.25 - 5 μM. One interpretation of the results is that **15** form precipitates rather than micelles in water above a concentration of 0.5 μM.

2.5.6 Conclusion.

1,1-Linked mannose derived disaccharides with a long-chain fatty acid ester on the mannose residue were found to inhibit HIV infectivity in the 50 μM range, with no cytotoxicity at concentrations up to 200 μM. These compounds appear to inhibit virus entry at early steps in viral infection since they were inactive if added post-viral entry. Their structural likeness to GalCer and the observation that they bind the V3 loop peptide of HIV-1 and also inhibit HIV infectivity fits with the notion their anti-HIV activity results from their ability to bind the GalCer domain of the V3 peptide. However, in the absence of other data this conclusion is very speculative. That certain closely related disaccharide analogs showed similar binding to the V3 peptide but did not inhibit HIV infectivity suggests that factors other than V3 loop binding may have an impact on anti-HIV activity, or may point to completely different mechanisms. In this context it is noteworthy that the virucidal activity of fatty acid derivatives of related disaccharide

structures and other small molecule amphiphilic lipids has been attributed to surfactant-like, membrane-perturbing properties.[39,40,41,42,43] Notwithstanding the mechanistic basis for their anti-HIV activity, the preliminary structure-activity data obtained from this investigation suggests that a disaccharide-type framework and the position of acylation are important for activity. Although these trehalose-type lipids have similar anti-HIV potency to other small molecule lipids, their low cytoxicity, combined with the easy accessibility to structurally diverse analogs, make them attractive leads for new broad-spectrum, anti-viral agents.[44,45,46]

2.6 Experimental

2.6.1 Surface pressure measurements.

The surface pressure was measured with a fully automated microtensiometer (mTROUGH SX; Kibron, Inc., Helsinki, Finland). The apparatus allowed the real-time recording of the kinetics of interaction of a soluble ligand with the monomolecular film, using a set of specially designed Teflon troughs. All experiments were carried out in a controlled atmosphere at 20°C ± 1°C. Monomolecular films of test compounds were spread on pure water subphases (volume of 800 mL) from hexane-chloroform-ethanol solution as described previously. After spreading of the film, 5 min was allowed for solvent evaporation. To measure the interaction of the V3 peptide with the individual monolayers, the peptide was injected in the subphase with a 10 μL Hamilton syringe, and pressure increases produced were recorded until reaching a stable value (Δp). The experiment was repeated at different values of the initial surface pressure (π) of the

monolayer. The data was analyzed with the Filmware 2.5 program (Kibron, Inc.). The results are expressed as the variations of Δp as a function of pi for the test compounds. The accuracy of the system under the experimental conditions was ± 0.25 mN/m for surface pressure.

2.6.2 Antiviral assays.

Testing of antiviral activity of test compounds was done as previously described.[35] Briefly, virus stocks were prepared by transfection of 293T cells with NL-Lai full-length infectious clone of HIV using Ex gen 500 transfection reagent (Fermentas, Glen Burnie, MD. Virus supernatant was collected 48 h post transfection, cleared of cellular components by centrifugation, aliquoted, stored at -70 ºC. Virus titers were determined in TZM-bl cells prior to use. For determination of antiviral activity TZM cells were seeded in 96 well plates at 2 x 10^4 cells per well and allowed to adhere overnight. The subsequent day cells were infected with HIV virus in the presence of 20 µg/ml DEAE dextran and test compounds at various concentrations. Virus infection was determined 24 h post infection as luciferase activity using Brite Lite Luciferase substrate (Perkin Elmer). Percent infection was calculated based on control wells infected with virus in the absence of any compound. Normalized data was fitted using Sigma Plot software and IC_{50} calculated based on the curve fit. Pooled data from at least three independent experiments was used for IC_{50} determination for each compound.

2.6.3 Cytotoxicity assays.

Cytotoxicity of compounds was determined by a MTS dye reduction assay.[36] Briefly TZM-bl cells were seeded in 96 well plates and incubated overnight. Subsequent

day serial dilutions of compounds starting at 200 μM were added to the cells. The cells were incubated for another 24 h before addition of the MTS reagent (Cell titer Aqueos one solution, Promega). Reduction of the MTS dye was determined by measuring OD at 490 nm and normalized to cells incubated with media only.

2.6.4 Pre and Post Infection assays

To determine the step at which the compounds showed activity a pre post infection time of addition assay was conducted. TZM-bl cells were infected with NL-Lai virus. The compounds (100 μM) were either added prior to infection (pre infection) or 4 h post infection. For the post infection the uninfected virus was removed by washing with PBS followed by addition of fresh media containing the compounds. For controls T20 and AMD3100 were used at 2 μM conc.

2.6.5 CMC Measurements

Stock solutions of glycolipids were prepared in hexane:chloroform:ethanol (11:5:4, v/v/v) and injected in water with a Hamilton microsyringe (dilution 1:1000). The surface tension was continuously recorded with the Kibron microtensiometer. Below the CMC, a drop in surface tension was recorded after each glycolipid injection. Increasing the concentration of the added glycolipid resulted in a linear decrease in surface tension. The CMC was determined as the concentration of glycolipid above which there was no further decrease in surface tension.

2.6.6 Synthesis

General Procedures: Unless otherwise stated, all reactions were carried out under a nitrogen atmosphere in oven-dried glassware using standard syringe and septa techniques. ^{1}H and ^{13}C NMR spectra were obtained on a Bruker 500 MHz spectrometer. Chemical shifts are relative to the deuterated solvent peak and are in parts per million (ppm). Assignments for selected nuclei were determined from ^{1}H COSY experiments. Thin-layer chromatography (TLC) was done on 0.25-mm thick precoated silica gel HF_{254} aluminum sheets. Chromatograms were observed under UV (short and long wavelength) light, and were visualized by heating plates that were dipped in a solution of ammonium(VI)molybdate tetrahydrate (12.5 g) and cerium(IV)sulfate tetrahydrate (5.0 g) in 10% aqueous sulfuric acid (500 mL). Flash column chromatography (FCC) was performed using silica gel 60 (230-400 mesh) and employed a stepwise solvent polarity gradient, correlated with TLC mobility. Solvents were purified by standard procedures or used from commercial sources as appropriate. High resolution mass spectra (HRMS) were obtained on an Ultima Micromass Q-TOF Mass Spectrometer at the Mass Spectrometry facility at Hunter College, CUNY.

2,3,4,6-Tetra-*O*-acetyl-α/β-D-galactopyranosyl-(1→1)-2,3:4,6-di-*O*-isopropylidene-α-D-mannopyranoside 24α/β. A mixture of mannose donor **23** (4.20 g, 11.9 mmol), galactose acceptor **21**[47] (4.10 g, 11.8 mmol), freshly activated, powdered, 4Å molecular sieves, DTBMP (17.3 g, 83.3 mmol), and dry CH_2Cl_2 (50 mL) was stirred for 30 min at rt. The mixture was then cooled to 0 °C and MeOTf (8.1 mL, 77.4 mmol) was added slowly. After the mixture had stirred for an additional 20 h, the reaction was quenched with triethylamine (3 mL). The mixture was diluted with CH_2Cl_2, filtered, and concentrated *in*

vacuo. FCC of the residue afforded **24α** (1.59 g, 23%) and **24β** (1.26 g, 18%) as white amorphous solids.

For **24α**: R_f = 0.32 (30% EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 1.41 (s, 3H), 1.42 (s, 3H), 1.52 (s, 3H), 1.57 (s, 3H), 2.03 (s, 3H), 2.06 (s, 3H), 2.14 (s, 3H), 2.17 (s, 3H), 3.50 (m, 1H), 3.74 (d, 1H, *J* = 8.1 Hz), 3.78 (m, 2H), 4.08 (m, 1H), 4.14 (m, 1H), 4.20 (d, 1H, *J* = 3.4 Hz), 4.29 (t, 1H, *J* = 6.7 Hz), 5.21 (dd, 1H, *J*= 3.7, 10.9 Hz), 5.30 (s, 1H), 5.36 (dd, 1H, *J*= 3.15, 10.9 Hz), 5.49 (apparent d, 2H, *J* = 3.5 Hz, H-1[Gal], H-4[Gal]); ^{13}C NMR ($CDCl_3$) δ 19.3, 21.2 (3 lines), 26.7, 28.6, 29.6, 61.9, 62.2, 62.6, 67.6, 67.8, 67.9, 68.2, 73.0, 75.2, 76.1, 91.8 ($J_{H1, C1}$ [Gal] = 172 Hz), 94.2 ($J_{H1, C1}$ [Man] = 175 Hz), 100.3, 110.4, 170.6, 170.7, 170.8, 170.9; HRMS (ESI) calcd. for $(M+Na)^+$ $C_{26}H_{38}NaO_{15}$ 613.2103, found 613.2110.

For **24β**: R_f = 0.18 (30% EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 1.37 (s, 3H), 1.43 (s, 3H), 1.53 (s, 3H), 1.57 (s, 3H), 2.01 (s, 3H), 2.07 (s, 3H), 2.09 (s, 3H), 2.20 (s, 3H), 3.71 (t, 1H, *J* = 9.81 Hz), 3.76 (d, 1H, *J* = 7.9 Hz), 3.82 (m, 1H), 3.89 (m, 1H), 3.97 (t, 1H, *J* = 6.91 Hz), 4.11 (m, 2H), 4.20 (m, 1H), 4.70 (d, 1H, *J* = 8.0 Hz, H-1 [Gal]), 5.21 (s, 1H), 5.24 (dd, 1H, *J* = 8.1, 10.5 Hz), 5.41 (d, 1H, *J* = 4.4 Hz); ^{13}C NMR ($CDCl_3$) δ 18.9, 20.7, 20.8, 20.9, 21.0, 26.4, 28.3, 29.2, 61.7, 62.0, 62.3, 67.2, 69.2, 71.0, 71.4, 72.5, 75.0, 75.6, 99.8 ($J_{H1, C1}$ [Man] = 172 Hz), 99.9 ($J_{H1, C1}$ [Gal] = 154 Hz), 100.4, 109.9; HRMS (ESI) calcd. for $(M+Na)^+$ $C_{26}H_{38}NaO_{15}$ 613.2103, found 613.2097.

2,3,4,6-Tetra-*O*-acetyl-α/β-D-glucopyranosyl-(1→1)-2,3:4,6-di-*O*-isopropylidene-α-D-mannopyranoside 25α/β. Disaccharides **25α/β** were prepared from mannose donor **23** (415 mg, 1.18 mmol) and gluco-acceptor **22** (373 mg, 1.07 mmol) via the procedure

described for **24α/β**.[48] For or **25α**: (130 mg, 22%); R_f = 0.45 (40% EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 1.42 (s, 3H), 1.43 (s, 3H), 1.58 (s, 6H), 2.05 (s, 3H), 2.06 (s, 3H), 3.52 (m, 1H), 3.76 (m, 3H), 4.08 (m, 1H), 4.13 (m, 2H), 4.24 (m, 2H), 4.30 (dd, 1H, *J* = 4.2, 4.3 Hz), 4.97 (dd, 1H, *J* = 3.9, 10.3 Hz), 5.13 (t, 1H, *J* = 9.0 Hz), 5.30 (s, 1H), 5.47 (d, 1H, *J* = 3.8 Hz, H1[Glu]), 5.50 (t, 1H, *J* = 9.9 Hz); ^{13}C NMR ($CDCl_3$) δ 18.9, 20.7, 20.8, 20.9, 26.4, 28.3, 29.3, 61.8, 61.9, 62.3, 68.3, 68.5, 70.0, 70.3, 72.6, 74.9, 75.7, 91.0 ($J_{H1,\,C1}$ [Glu] = 172 Hz), 93.9 ($J_{H1,\,C1}$ [Man] = 175 Hz), 100.0, 110.1, 169.7, 170.2, 170.4, 170.8; HRMS (ESI) calcd. for $(M+Na)^+$ $C_{26}H_{38}NaO_{15}$ 613.2103, found 613.2095.

For **25β**: (124 mg, 21%); R_f = 0.39 (40% EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 1.37 (s, 3H), 1.43 (s, 3H), 1.52 (s, 3H), 1.57 (s, 3H), 2.03 (s, 3H), 2.05 (s, 3H), 2.08 (s, 3H), 2.12 (s, 3H), 3.74 (m, 4H), 3.89 (dd, 1H, *J* = 5.0, 10.1 Hz), 4.12 (d, 1H, *J* = 5.7 Hz), 4.19 (m, 3H), 4.74 (d, 1H, *J* = 8.0 Hz, H-1[Glu]), 5.01(d, 1H, *J* = 9.6 Hz), 5.05 (t, 1H, *J* = 9.7 Hz), 5.23 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 18.9, 20.8 (two lines), 26.4, 28.4, 29.1, 61.6, 62.4, 62.5, 68.4, 71.6, 72.4, 72.6, 72.9, 75.0, 75.6, 99.7 ($J_{H1,\,C1}$ [Man]) = 172 Hz), 99.8 ($J_{H1,\,C1}$ [Glu] = 152 Hz), 99.9, 109.9, 169.5, 169.6, 170.4, 170.9; HRMS (ESI) calcd. for $(M+Na)^+$ $C_{26}H_{38}NaO_{15}$ 613.2103, found 613.2101.

2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl-(1→1)-2,3-*O*-isopropylidene-α-D-mannopyranoside 26. Disaccharide **24β** (0.514 g, 0.87 mmol) was treated with NaOMe (100 mg, 1.85 mmol) in dry methanol (10 mL) under N_2 for 15 min. The reaction mixture was then neutralized with methanolic HCl and concentrated under reduced pressure. The crude product was dried *in vacuo* and taken up in dry THF (15 mL). TBAI (0.032 g, 0.087 mmol) and NaH (60% dispersion in mineral oil, 0.348 g, 8.7 mmol) were added to

the solution at 0 °C, under nitrogen. After the mixture had stirred for 45 min at this temperature, BnBr was added drop-wise to the mixture and stirring was continued at rt for 15 h. The reaction mixture was then cooled to 0 °C, quenched with water and extracted with ether. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue provided the benzylated disaccharide as a clear oil, (0.498 g, 73%), R_f = 0.53 (30% EtOAc/petroleum ether). To a solution of this material (130 mg, 0.166 mmol) in MeOH (6 mL) and DCM (2 mL) was added *p*-TsOH (50 mg, 0.262 mmol) at rt. The mixture was stirred for 45 min, and then was quenched with saturated aqueous $NaHCO_3$ solution and extracted with DCM. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded diol **26** (118 mg, 96%) as a white solid. R_f = 0.65 (70 % EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 1.37 (s, 3H), 1.53 (s, 3H), 3.39 (m, 1H), 3.56 (dd, 1H, *J* = 2.9, 6.9 Hz), 3.61 (m, 3H), 3.79 (dd, 1H, *J* = 3.1, 8.7 Hz), 3.83 (d, 1H, *J* = 2.8 Hz), 3.87 (t, 1H, *J* = 7.9 Hz), 4.01 (m, 1H), 4.06 (d, 1H, *J* = 5.7 Hz), 4.14 (m, 2H), 4.39 (d, 1H, *J* = 11.8 Hz), 4.48 (d, 1H, *J* = 11.7 Hz), 4.61 (d, 1H, *J* = 6.5 Hz), 4.64 (d, 1H, *J* = 2.6 Hz), 4.75 (d, 2H, *J* = 2.9 Hz), 4.80 (d, 1H, *J* = 11.2 Hz), 4.87 (d, 1H, *J* = 11.2 Hz), 4.94 (d, 1H, *J* = 11.7 Hz), 5.29 (s, 1H), 7.33 (m, 20 H); ^{13}C NMR ($CDCl_3$) δ 26.2, 28.0, 62.4, 69.3, 70.1, 70.6, 73.0, 73.3, 73.5, 73.8, 74.4, 75.3, 75.4, 78.5, 79.4, 82.5, 99.0, 102.2, 109.6, 127.6 (two lines), 127.7, 127.9, 128.0, 128.1, 128.2, 128.3, 128.4 (three lines), 137.4, 138.2, 138.3, 138.5; HRMS (ESI) calcd. for $(M+Na)^+$ $C_{43}H_{50}NaO_{11}$ 765.3245, found 765.3243.

2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl-(1→1)-2,3-*O*-isopropylidene-6-*O*-palmitate-α-D-mannopyranoside 27. To a solution of **26** (118 mg, 0.160 mmol) and palmitic acid (45 mg, 0.176 mmol), in dry benzene (4 mL) were added DCC (36 mg,

0.176 mmol) and DMAP (5 mg, 0.04 mmol) at 0 °C. The reaction mixture was stirred for 18 h at rt and then diluted with ether and filtered through a bed of Celite. The filtrate was concentrated *in vacuo.* FCC of the residue gave **27** (100 mg, 86% brsm). R_f = 0.4 (30% EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 0.90 (t, 3H, *J* = 6.8, 7.0 Hz), 1.27 (bs, 25H), 1.36 (s, 3H), 1.51 (s, 3H), 1.62 (m, 2H), 2.35 (t, 2H, *J* = 7.5 Hz), 3.15 (bs, 1H), 3.46 (m, 2H), 3.55 (m, 2H), 3.60 (t, 1H, *J* = 6.2 Hz), 3.85 (t, 1H, *J* = 7.9 Hz), 3.89 (d, 1H, *J* = 2.4 Hz), 4.00 (m, 1H), 4.06 (m, 1H), 4.15 (t, 1H, *J* = 5.9 Hz), 4.38 (d, 1H, *J* = 11.6 Hz), 4.43 (d, 1H, *J* = 11.6 Hz), 4.57 (m, 2H), 4.63 (d, 1H, *J* = 11.7 Hz), 4.74 (s, 1H), 4.79 (d, 1H, *J* = 11.2 Hz), 4.86 (d, 1H, *J* = 11.2 Hz), 4.95 (d, 1H, *J* = 11.7 Hz), 5.30 (s, 1H), 7.34 (m, 20H); ^{13}C NMR ($CDCl_3$) δ 14.3, 22.9, 25.3, 26.4, 28.3, 29.4, 29.5 (two lines), 29.6, 29.8 (two lines), 29.9, 32.1, 34.4, 62.6, 68.5, 69.3, 69.4, 73.2, 73.4, 73.7, 74.0, 74.7, 75.4, 75.7, 77.8, 79.7, 82.8, 99.6, 102.7, 109.7, 127.7 (three lines), 128.0, 128.1, 128.2, 128.4, 128.6 (three lines), 137.8, 138.4, 138.6, 138.7, 175.4.

2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl-(1→1)-2,3-*O*-isopropylidene-4,6-di-*O*-palmitate-α-D-mannopyranoside 28. Diol **26** (60 mg, 0.081 mmol) was treated as described for synthesis of **27**, with excess palmitic acid (52 mg, 0.2 mmol) to afford **28** as a white solid (68 mg, 70%); R_f = 0.82 (25 % EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 0.92 (s, 6H), δ 1.29 (s, 48H), δ 1.60 (s, 4H), 2.30 (s, 4H), 3.56 (m, 4H), 3.86 (dd, 1H, *J* = 7.8, 9.6 Hz), 3.93 (s, 1H), 4.02 (d, 2H, *J* = 13.7 Hz), 4.19 (d, 2H, *J* = 8.4 Hz), 4.28 (d, 1H, *J* = 12.4 Hz), 4.40 (d, 1H, *J* = 11.7 Hz), 4.48 (d, 1H, *J* = 11.7 Hz), 4.59 (d, 1H, *J* = 9.5 Hz), 4.64 (d, 1H, *J* = 13.4 Hz), 4.74 (s, 1H), 4.79 (d, 1H, *J* = 13.1 Hz), 4.88 (d, 1H, *J* = 13.1 Hz), 4.94 (d, 1H, *J* = 13.3 Hz), 5.18 (t, 1H, *J* = 10.2 Hz), 5.35 (s, 1H), 7.32 (m, 20H); ^{13}C NMR ($CDCl_3$) δ 14.3, 22.9, 25.0, 26.8, 27.9, 29.3, 29.4, 29.5 (two lines), 29.7

(two lines), 29.8, 29.9 (two lines), 32.1, 34.3, 34.4, 61.5, 67.0, 68.5, 68.8, 70.8, 73.2, 73.4, 73.7, 73.9, 74.9, 75.7 (two lines), 76.5, 79.7, 82.9, 99.3, 102.9, 110.3, 127.7, 127.9 (two lines), 128.1, 128.4, 128.6 (two lines), 137.9, 138.4, 138.6, 138.7, 172.5, 173.8; HRMS (ESI) calcd for $(M+Na)^+$ $C_{75}H1_{10}NaO_{13}$ 1241.7389, found 1241.7389.

β-D-Galactopyranosyl-(1→1)-6-*O*-palmitate-α-D-mannopyranoside 10. A solution of compound **27** (100 mg, 0.10 mmol) in MeOH (5 mL) was treated with *p*-TsOH (100 mg, 0.52 mmol). The reaction mixture was stirred for 1 h and then was quenched with saturated aqueous $NaHCO_3$ solution and extracted with DCM. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded the corresponding triol (70 mg, 73%). To a solution of the triol (30 mg, 0.032 mmol) was added 10% Pd/C (40 mg) and HCOOH (0.1 mL). The reaction mixture was then stirred under H_2 atmosphere for 20 h. The mixture was filtered through a bed of Celite and concentrated *in vacuo*. FCC of the residue gave **10** (9 mg, 50%): R_f = 0.36 (20% MeOH/EtOAc); ^{1}H NMR (CD_3OD) δ 0.89 (t, 3H, *J* = 6.8 Hz), 1.28 (m, 28H), 1.61 (m, 2H), 2.36 (t, 2H, *J* = 7.5 Hz), 3.46 (dd, 1H, *J* = 3.0, 9.7 Hz), 3.53 (m, 2H), 3.71 (m, 5H), 3.85 (d, 1H, *J* = 2.7 Hz), 3.95 (s, 1H), 4.05 (m, 1H), 4.25 (dd, 1H, *J* = 4.9, 11.95 Hz), 4.31 (d, 1H, *J* = 11.2 Hz), 4.41 (d, 1H, *J* = 7.7 Hz), 5.02 (s, 1H); ^{13}C NMR (CD_3OD) δ 14.6, 23.9, 26.2, 30.4, 30.6, 30.8, 30.9 (two lines), 33.2, 35.1, 62.2, 64.9, 68.5, 70.1, 71.6, 72.4, 72.7, 72.8, 75.1, 77.0, 103.2, 104.4, 175.9; HRMS (ESI) calcd for $(M+Na)^+$ $C_{28}H_{52}NaO_{12}$ 603.3351, found 603.3352.

β-D-Galactopyranosyl-(1→1)-4,6-di-*O*-palmitate-α-D-mannopyranoside 12. Diester **28** (72 mg, 0.06 mmol) was dissolved in dry MeOH (3 mL) and dry CH_2Cl_2 (1 mL). CSA (25 mg, 0.21 mmol) was then added. The reaction mixture was stirred for 1 h, and then

was neutralized with Et_3N and the solvent was evaporated under reduced pressure. FCC of the residue gave the derived diol (32 mg, 46%): white solid. R_f = 0.54 (35% EtOAc/petroleum ether)**.** This material (32 mg, 0.027 mmol) was subjected to the hydrogenolysis procedure described for **10**. FCC of the crude product afforded **12** (13 mg, 60%) as a white solid; R_f = 0.32 (10% MeOH/EtOAc); ^{1}H NMR (CD_3OD) δ 0.90 (t, 6H, *J* = 6.8 Hz), 1.31 (s, 48H), 1.61 (m, 4H), 2.35 (m, 4H), 3.48 (dd, 1H, *J* = 3.3, 9.7 Hz), 3.54 (m, 2H), 3.72 (d, 2H, *J* = 6.3 Hz), δ 3.85 (d, 1H, *J* = 3.3 Hz), 3.89 (dd, 1H, *J* = 3.3, 9.8 Hz), 4.01 (m, 1H), 4.05 (m, 1H), 4.21 (m, 1H), 4.43 (d, 1H, *J* = 7.7 Hz), 5.07 (s, 1H), 5.24 (t, 1H, *J* = 9.8 Hz); ^{13}C NMR (CD_3OD) δ14.6, 23.9, 26.0, 26.1, 30.3, 30.4, 30.6, 30.7, 30.8 (two lines), 30.9, 31.0, 33.2, 35.0, 35.2, 62.4, 63.8, 70.1, 70.2, 70.6, 71.7, 72.7, 75.1, 77.1, 103.3, 104.7, 174.9, 175.5; HRMS (ESI) calcd for $(M+Na)^+$ $C_{44}H_{82}NaO_{13}$ 841.5648, found 841.5641.

2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl-(1→1)-4.6-*O*-benzyl-α-D-mannopyranoside 29. Diol **26** (61mg, 0.082 mmol) was subjected to the standard benzylation procedure described previously. FCC of the crude product yielded the di-*O*-benzylated deriative (71 mg, 0.08 mmol), which was taken up in dry MeOH (5 mL) and treated with CSA (100 mg, 0.43 mmol). The mixture was stirred for 30 min, quenched with Et_3N, and concentrated *in vacuo*. FCC of the residue afforded **29** (55 mg, 75% two steps) as a clear oil: R_f = 0.55 (60% EtOAc/petroleum ether); ^{1}H NMR ($CDCl_3$) δ 3.56 (m, 5H), 3.73 (dd, 1H, *J* = 3.15, 3.05 Hz), 3.81 (m, 2H), 3.88 (m, 2H), 3.97 (dd, 1H, *J* = 3.40, 3.50 Hz), 4.12 (m, 1H), 4.37 (s, 2H), 4.40 (s, 1H), 4.59 (m, 4H), 4.60 (d, 1H, *J* = 11.1 Hz), 4.73 (s, 2H), 4.78 (d, 1H, *J* = 11.1 Hz), 4.82 (d, 1H, *J* = 11.1 Hz), 4.94 (d, 1H, *J* = 11.7 Hz), 7.24 – 7.39 (m, 30H); ^{13}C NMR ($CDCl_3$) δ 68.7, 68.9, 71.1, 71.5, 71.7, 73.1, 73.6

(two lines), 73.8, 74.6, 74.8, 75.6, 75.8, 79.7, 82.7, 101.5, 103.5, 127.7, 127.8 (three lines), 127.9 (three lines), 128.0, 128.1, 128.4 9 (two lines), 128.5, 128.6 (three lines), 138.1, 138.4, 138.6, 138.7, 138.8.

2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl-(1→1)-4,6-*O*-benzyl-3-*O*-palmitate-α-D-mannopyranoside 30. To a solution of **29** (80 mg, 0.090 mmol) and palmitic acid (26 mg, 0.099 mmol) in dry benzene (3 mL) were added DCC (20 mg, 0.099 mmol) and DMAP (2 mg, 0.018 mmol) at 0 °C. The reaction mixture was stirred for 18 h at rt and then diluted with ether, filtered through a bed of Celite, and concentrated *in vacuo*. FCC of the residue yielded **30** (74 mg, 80% brsm) as a white solid. R_f = 0.3 (30% EtOAc/petroleum ether). ^{1}H NMR ($CDCl_3$) δ 0.90 (t, 3H, *J* = 6.8 Hz), 1.27 (m, 24H), 1.61 (m, 2H), 2.28 (m, 2H), 3.54 (m, 6H), 3.71 (dd, 1H, *J* = 3.5, 11.5), 3.89 (m, 2H), 4.03 (m, 2H), 4.23 (m, 1H), 4.35 (m, 3H), 4.47 (d, 1H, *J* = 11.3 Hz), 4.56 (m, 1H), 4.62 (m, 2H), 4.73 (m, 2H), 4.83 (s, 1H), 4.95 (d, 1H, *J* = 12.0 Hz), 5.11 (d, 1H, *J* = 1.5 Hz), 5.34 (dd, 1H, *J* = 3.15, 9.72), 7.34 (m, 35H); ^{13}C NMR ($CDCl_3$) δ 14.1, 22.7, 24.8, 24.9, 25.5, 29.1, 29.2, 29.3, 29.4, 29.6 (three lines), 29.7, 31.9, 33.7, 34.4, 68.2, 68.7, 69.5, 71.7, 72.7, 72.9, 73.3, 73.6, 73.9, 74.5, 75.5, 79.3, 82.3, 101.4, 103.6, 127.4 (two lines), 127.5 (three lines), 127.6, 127.8 (two lines), 128.0, 128.2 (two lines), 128.3 (three lines), 128.4, 137.9, 138.3, 138.4, 138.5, 138.6, 172.6; HRMS (ESI) calcd for $(M+NH_4)^+$ $C_{70}H_{92}NO_{12}$ 1138.6614, found 1138.6606.

β-D-Galactopyranosyl-(1→1)-3-*O*-palmitate-α-D-mannopyranoside 4. The hexa-*O*-benzyl ether **30** (54 mg, 0.048 mmol) was subjected to the hydrogenolysis procedure described for **10**. FCC of the crude product provided **11** (22 mg, 79%): R_f = 0.58 (20% MeOH/CH_2Cl_2); ^{1}H NMR (CD_3OD) δ 0.90 (t, 3H, *J* = 6.8 Hz), 1.16 (m, 5H), 1.29 (bs,

26H), 1.63 (m, 4H), 1.71 (m, 2H), 1.85 (m, 2H), 2.41 (t, 2H, *J* = 7.45 Hz), 3.46 (m, 2H), 3.56 (m, 1H), 3.65 (m, 3H), 3.78 (m, 3H), 3.88 (d, 1H, *J* = 10.2 Hz), 4.10 (m, 2H), 4.44 (d, 1H, *J* = 7.85 Hz), 5.04 (dd, 1H, *J* = 3.2, 9.8 Hz), 5.06 (s, 1H); ^{13}C NMR (CD_3OD) δ 14.6, 23.9, 26.1, 26.2, 26.9, 30.3, 30.6, 30.8, 30.9 (two lines), 33.2, 34.9, 35.2, 63.0, 63.1, 66.3, 69.7, 70.5, 72.5, 75.0, 75.3, 75.6, 77.4, 102.9, 104.7, 175.5; HRMS (ESI) calcd for $C_{28}H_{52}NaO_{12}$ $(M+Na)^+$ 603.3351, found 603.3351.

2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl-(1→1)-2,3-*O*-isopropylidene-α-D-mannopyranoside 31. Protected disaccharide **25β** was transformed to **31** (58 mg, 75% brsm) by using the three-step procedure described for the conversion of **24β** to **26**. Data for **31**: white solid; R_f = 0.18 (40% EtOAc/Petroleum ether); ^{1}H NMR ($CDCl_3$) δ 1.37 (s, 3H), 1.54 (s, 3H), 2.22 (d, 1H, *J* = 4.35 Hz), 2.55 (dd, 1H, *J* = 5.65, 7.45 Hz), 3.48 (m, 2H), 3.56 (m, 2H), 3.61-3.70 (m, 4H), 3.80 (m, 1H), 3.97 (m, 1H), 4.04 (d, 1H, *J* = 5.8 Hz), 4.14 (m, 1H), 4.51 (m, 3H), 4.67 (d, 1H, *J* = 7.9 Hz), 4.83 (m, 4H), 4.92 (d, 1H, *J* = 10.9 Hz), 5.30 (s, 1H), 7.33 (m, 20H); ^{13}C NMR ($CDCl_3$) δ 26.4, 28.2, 62.6, 69.2, 70.3, 71.1, 73.6, 74.9, 75.2, 75.4, 75.6, 75.9, 78.0, 78.7, 82.4, 85.0, 99.4, 102.5, 109.9, 127.9, 128.0, 128.1 (two lines), 128.2, 128.3, 128.6 (two lines), 128 .7, 137.7, 138.0, 138.4, 138.5; HRMS (ESI) calcd for $(M+Na)^+$ $C_{43}H_{50}NaO_{11}$ 765.3245, found 765.3238.

β-D-Glucopyranosyl-(1→1)-6-*O*-palmitate-α-D-mannopyranoside 13. Diol **31** was transformed to **13** (9 mg, 28% brsm) by using the three-step procedure described for the conversion of **26** to **10**. Data for **13**: R_f = 0.37 (20% MeOH/EtOAc); ^{1}HNMR (CD_3OD) δ 0.92 (t, 3H, *J* = 6.8 Hz), 1.31 (bs, 24H), 1.64 (m, 2H), 2.23 (t, 2H, *J* = 7.4 Hz), 3.23 (t, 1H, *J* = 8.0 Hz), 3.33 (m, 2H), 3.70 (m, 3H), 3.87 (dd, 1H, *J* = 2.2, 11.85 Hz), 3.98 (m, 1H),

4.10 (m, 1H), 4.29 (dd, 1H, *J* = 5.15, 11.9 Hz), 4.36 (dd, 1H, *J* = 2.2, 11.95 Hz), 4.48 (d, 1H, *J* = 8.0 Hz), 5.03 (s, 1H); ^{13}C NMR (CD_3OD) δ 14.6, 23.9, 26.1, 30.4, 30.6, 30.8, 30.9, 33.2, 35.1, 62.7, 64.9, 68.5, 71.3, 71.6, 72.4, 72.8, 75.3, 78.2, 78.4, 103.1, 103.7, 175.9; HRMS (ESI) calcd for $C_{28}H_{52}NaO_{12}$ $(M+Na)^+$ 603.3351, found 603.3344.

Methyl-6-*O*-palmitoyl-α-D-mannopyranoside 14. Compound **14** (8%) was prepared from **32** [49] by using the procedures described for the transformation of **26** to **10**. Data for **14**: R_f = 0.60 (EtOAc); ^{1}H NMR ($CDCl_3$) δ 0.88 (t, 3H, *J* = 6.9 Hz), 1.26 (bs, 24H), 1.65 (m, 2H), 2.28 (bs, 1H), 2.41 (t, 2H, *J* = 7.4 Hz), 3.39 (s, 3H), 3.56 (t, 1H, *J* = 9.6 Hz), 3.69 (m, 1H), 3.83 (dd, 1H, *J* = 3.3, 9.25 Hz), 3.97 (s, 1H), 4.21 (dd, 1H, *J* = 2.0, 12.35 Hz), 4.66 (dd, 1H, *J* = 3.8, 12.35 Hz), 4.77 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 14.3, 22.9, 25.1, 29.3, 29.4, 29.5, 29.6, 29.8 (two lines), 29.9, 32.1, 34.4, 55.3, 63.3, 67.7, 70.4, 70.5, 71.4, 101.0, 175.5; HRMS (ESI) calcd. for $(M+Na)^+$ $C_{23}H_{44}NaO_7$ 455.2979, found 455.2986.

Methyl-4,6-di-*O*-palmitoyl-α-D-mannopyranoside 15. Compound **15** (70%) was prepared from **32** by using the procedures described for the transformation of **26** to **12**. Data for **15**: R_f = 0.6 (80% EtOAc/petroleum ether); ^{1}H NMR δ 0.90 (t, 6H, *J* = 6.8 Hz), 1.27 (bs, 48H), 1.64 (m, 4H), 2.36 (m, 4H), 2.72 (bs, 1H), 3.12 (bs, 1H), 3.41 (s, 3H), 3.90 (m, 2H), 3.96 (s, 1H), 4.19 (dd, 1H, *J* = 2.2, 12.1 Hz), 4.28 (dd, 1H, *J* = 5.5, 12.07 Hz), 4.81 (s, 1H), 5.02 (t, 1H, *J* = 9.8 Hz). ^{13}C NMR δ 14.3, 22.9, 25.0 (two lines), 29.2, 29.3, 29.4, 29.5 (two lines), 29.6, 29.7, 29.8 (three lines), 29.9, 32.1, 34.3, 34.5, 55.3, 62.7, 68.0, 70.1, 70.6, 70.7, 100.7, 173.8, 175.0; HRMS (ESI) calcd for $C_{39}H_{74}NaO_8$ $(M + Na)^+$ 693.5276, found 693.5278.

β-D-Galactopyranosyl-(1→1)-α-D-mannopyranoside 16. Treatment of **26** (20 mg, 0.030 mmol) by using the standard procedures for acetonide hydrolysis and benzyl ether hydrogenolysis as described for **10** provided **16** (6 mg, 65% over 2 steps): R_f = 0.4 (50% MeOH/CH_2Cl_2) ^{1}H NMR (CD_3OD) δ 3.46 (dd, 1H, *J* = 3.3, 7.25 Hz), 3.56 (m, 3H), 3.64 (m, 2H), 3.73 (dd, 1H, *J* = 3.4, 9.5 Hz), 3.77 (dd, 1H, *J* = 7.5, 11.5 Hz), 3.80 (d, 1H, *J* = 2.8 Hz), 3.88 (dd, 1H, *J* = 2.15, 11.5 Hz), 3.95 (m, 1H), 3.97 (m, 1H), 4.43 (d, 1H, *J* = 7.8 Hz), 5.06 (s, 1H). ^{13}C NMR (CD_3OD) δ 63.0, 63.3, 69.0, 70.5, 71.8, 72.6, 75.1, 75.2, 77.4, 102.9, 104.5. HRMS (ESI) calcd for $C_{12}H_{22}NaO_{11}$ $(M+Na)^+$, 365.1048 found 365.1054.

β-D-Galactopyranosyl-(1→1)-6-*O*-hexadecyl-α-D-mannopyranoside 17. To a solution of **26** (44 mg, 0.06 mmol) in CH_2Cl_2 (3 mL) were added dry pyridine (50 μL, 0.6 mmol) and pivaloyl chloride (8 μL, 0.066 mmol). The reaction mixture was stirred for 2 h and then was diluted with EtOAc. The mixture was washed with 1 N HCl (aq), and the organic extract was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. After the crude product was taken up in DCM (1 mL), and ethyl vinyl ether (2 mL, 20.8 mmol) and PPTS (50 mg, 0.20 mmol) were added. This mixture was stirred for 15 min, and then was neutralized with Et_3N and concentrated *in vacuo*. The residue was subjected to FCC and the purified material (50 mg, 0.056 mmol) was dissolved in THF (10 mL). After LAH (50 mg, 1.30 mmol) was added, the mixture was stirred at rt for 2 h. The reaction was cooled to 0 °C and diluted with water (1 mL) and 2 N NaOH (2 mL). The product was extracted with EtOAc and the organic was phase dried (Na_2SO_4), filtered, and concentrated under reduced pressure. FCC of the residue gave the derived primary alcohol. To a solution of this material (40 mg, 0.050 mmol) in THF (6 mL) were added NaH (60% suspension in mineral oil, 10 mg, 0.25 mmol) and 1-bromohexadecane (80 μL,

0.25 mmol). The reaction mixture was stirred for 3 h and then was quenched with water. The product was extracted with ether, and the organic layer was processed as described in the previous step. The crude product was dissolved in MeOH (10 mL) and *p*-TsOH (25 mg, 0.132 mmol) was added. The reaction mixture was stirred for 30 min, and then was neutralized with Et_3N (2 mL) and concentrated *in vacuo*. FCC of the residue gave the derived 6-hexadecyl ether [41 mg, R_f = 0.42 (60% EtOAc/petroleum ether)]. This product was subjected to the standard hydrogenolysis procedure described for **10**, to give **17** (9.4 mg, 37% from **26**): R_f = 0.17 (20% MeOH/EtOAc); ^{1}H NMR (CD_3OD) δ 0.80 (m, 3H), 1.20 (bs, 24H), 1.50 (m, 4H), 3.39 (m, 6H), 3.50 (m, 4H), 3.64 (m, 4H), 3.72 (d, 1H, *J* = 3.0 Hz), 3.83 (m, 1H), 3.91 (m, 1H), 4.31 (d, 1H, *J* = 7.9 Hz), 4.97 (s, 1H). ^{13}C NMR (CD_3OD) δ 14.5, 23.9, 27.2, 30.5, 30.6, 30.8, 30.9 (two lines), 33.2, 62.8, 69.0, 71.8, 71.9, 72.5 (two lines), 73.0, 74.1, 75.0, 77.3, 102.5, 104.6; HRMS (ESI) calcd for $C_{28}H_{54}NaO_{11}$ $(M+Na)^+$, 589.3557 found 589.3558.

α-D-Galactopyranosyl-(1→1)-6-*O*-palmitate-α-D-mannopyranoside 19. Disaccharide **24α** was transformed to **19** (60 mg, 13.2% bsrm) by using the procedures described for the conversion of **24β** to **10**. Data for **19**: R_f = 0.2 (20% MeOH/EtOAc); ^{1}H NMR (CD_3OD) δ 0.81 (t, 3H, *J* = 6.9 Hz), 1.21 (bs, 23H), 1.52 (m, 2H), 2.26 (t, 2H, *J* = 7.5 Hz), 3.21 (m, 1H), 3.61 (m, 4H), 3.74 (m, 4H), 3.82 (d, 1H, *J* = 2.5 Hz), 3.87 (m, 1H), 4.13 (dd, 1H, *J* = 5.9, 11.8 Hz), 4.29 (bd, 1H, *J* = 11.7 Hz), 4.99 (s, 1H), 5.03 (d, 1H, *J* = 3.7 Hz); ^{13}C NMR (CD_3OD) δ 14.5, 23.8, 26.1, 30.4, 30.6 (two lines), 30.7, 30.9 (two lines), 33.2, 35.1, 62.8, 64.8, 68.7, 69.7, 71.1, 71.3, 72.2, 72.5, 73.3, 95.5, 96.7, 175.7; HRMS (ESI) calcd for $C_{28}H_{56}NO_{12}$ $(M+NH_4)^+$, 589.3799 found 589.3797.

2,3,4,-Tri-*O*-benzyl-6-*O*-triisopropylsilyl-β-D-galactopyranosyl-(1→1)-2,3,4,6-di-*O*-isopropylidene-α-D-mannopyranoside 33. To a solution of compound **24β** (0.9 g, 1.5 mmol) in MeOH (20 mL) was added NaOMe (100 mg, 1.85 mmol). The reaction mixture was stirred for 30 min and then was neutralized with 1M HCl in MeOH. The solvent was removed *in vacuo*, and the crude product was taken up into DCM and filtered through a pad of Celite. This afforded the tetraol (635 mg). To a solution of the tetraol (386 mg, 0.91mmol) in dry DCM (40 mL) were added imidazole (618 mg, 9.1 mmol) and TIPSCl (0.2 mL, 0.96 mmol). The reaction mixture was stirred for 18 h at rt and then washed with water. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue provided the 6-Gal silylated compound (500 mg, 94%). To a solution of this compound (275 mg, 0.48 mmol) in DMF (15 mL) were added NaH (60% dispersion in mineral oil, 80 mg, 1.9 mmol), TBAI (18 mg, 0.048 mmol), and BnBr (0.2 mL, 1.7 mmol). The reaction mixture was stirred for 18 h and then was quenched with water. The mixture was extracted with ether and the organic fraction was washed further with water, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave compound **33** (284 mg, 66% over 3 steps). R_f = 0.53 (10% EtOAc/petroleum ether). ^{1}H NMR ($CDCl_3$) δ 1.08 (m, 18H), 1.28 (m, 3H), 1.35 (s, 3H), 1.41 (s, 3H), 1.51 (s, 3H), 1.56 (s, 3H), 3.43 (dd, 1H, *J* = 5.4, 8.3 Hz), 3.57 (dd, 1H, *J* = 2.8, 9.8 Hz), 3.71 (m, 3H), 3.77 (m, 1H), 3.86 (m, 3H), 4.00 (d, 1H, *J* = 2.4 Hz), 4.15 (m, 3H), 4.54 (d, 1H, *J* = 7.8 Hz), 4.59 (s, 1H), 4.74 (m, 3H), 4.80 (d, 1H, *J* = 11.15 Hz), 4.89 (d, 1H, *J* = 11.1 Hz), 4.99 (d, 1H, *J* = 11.35 Hz), 5.30 (s, 1H), 7.35 (m, 15H). ^{13}C NMR ($CDCl_3$) δ 12.0, 18.2 (two lines), 19.0, 26.4, 28.4, 29.2, 61.6, 62.0 (two lines), 72.3, 72.6, 72.9, 73.3, 75.0, 75.1,

75.6, 75.7, 75.9, 79.7, 82.7, 99.5, 99.6, 102.7, 109.7, 127.6, 127.7, 127.8, 127.9, 128.0, 128.2, 128.3 (two lines), 128.6, 138.5, 138.6, 139.0.

6-*O*-Palmitate-β-D-galactopyranosyl-(1→1)-α-D-mannopyranoside 18. To a solution of compound **33** (284 mg, 0.33 mmol) in THF (10 mL) was added TBAF (1M solution in THF, 0.7 mL) at rt. The reaction mixture was stirred for 3 h and then was diluted in EtOAc (30 mL). The organic layer was washed with water, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the primary alcohol in quantitative yield. To a solution of the primary alcohol (100 mg, 0.14 mmol) in dry DCM (10 mL) were added palmitic acid (43 mg, 0.17 mmol), DMAP (10 mg, 0.08 mmol), and DCC (34 mg, 0.17 mmol) at 0 °C. The reaction mixture was stirred for 3 h and then was diluted with ether and filtered through a pad of Celite. The solvents were removed *in vacuo* and FCC of the residue afforded the 6-Gal palmitate (123 mg, 91%). To a solution of the 6-Gal palmitate ester (123 mg, 0.13 mmol) in 2:1 MeOH/DCM (12 mL) was added *p*-TsOH (50 mg, 0.26 mmol) at rt. The reaction mixture was stirred for 2 h and then was quenched with saturated aqueous $NaHCO_3$ solution and extracted with DCM. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the Man-tetraol. To a sample of the Man-tetraol (80 mg, 0.09 mmol) was subjected to standard hydrogenolysis as described for **27** to **10**. FCC of the residue gave compound **18** (40 mg, 64% over 4 steps). R_f = 0.63 (40% MeOH/DCM). ^{1}H NMR (CD_3OD) δ 0.80 (t, 3H, J = 6.8 Hz), 1.19 (s, 24H), 1.50 (m, 2H), 2.21 (t, 2H, J = 7.45 Hz), 3.39 (dd, 1H, J = 3.35, 9.75 Hz), 3.49 (m, 3H), 3.56 (m, 3H), 3.66 (dd, 1H, J = 3.35, 9.5 Hz), 3.70 (dd, 1H, J = 3.35, 11.4 Hz), 3.73 (d, 1H, J = 3.2 Hz), 3.80 (dd, 1H, J = 2.1, 11.6 Hz), 3.87 (m, 1H), 3.91 (m, 1H), 4.35 (d, 1H, J = 7.75 Hz), 4.97 (s, 1H). ^{13}C NMR (CD_3OD) δ 14.6, 23.8,

26.1, 30.3, 30.5, 30.6, 30.7, 30.8 (two lines), 30.9 (two lines), 33.2, 34.9, 52.1, 63.0, 63.3, 69.0, 70.5, 71.8, 72.5, 75.0, 75.1, 77.4, 102.9, 104.5, 176.1.

Glyceryl monolaurate (GML) 20. The synthesis of this compound has been previously reported.[30] Analytical data were in agreement with the reported data.

Chapter 3

Carbohydrate Based Anti-tumor Agents

3.1 Introduction

Cancer is the second most common cause of death in the United States exceeded only by heart disease.[50] The National Cancer Institute estimates that approximately 11.4 million Americans were living with cancer in January 2006. About 1.5 million new cancer cases were expected to be diagnosed and over 0.5 million Americans were expected to die of cancer in 2010 (more than 1500 people a day). Chemotherapeutic agents are a mainstay in the treatment of cancer, and in this context 60% of the presently available anticancer drugs are natural compounds or their analogs. The annonaceous acetogenins (AAs) have drawn considerable attention as potential therapeutic agents because of the potent activity against human tumor cell lines. Over 500 AAs have been isolated from the *Annonaceae* family of tropical plants, which includes both genera *Annona* and *Asimina*.[51,52,53] Studies on the AAs have intensified since the first cytotoxic acetogenin, uvaricin (ii), was isolated in 1982.[54] AAs are C_{35} or C_{37} secondary metabolites that are biosynthesized through a polyketide pathway. The majority of AAs contain one or more tetrahydrofuran (THF) rings and are classified into three major structural subgroups depending on the number and arrangement of the THF rings: the mono-THF acetogenins, the adjacently linked bis-THF acetogenins, and the non-adjacently connected bis-THF acetogenins (Figure 12). A small number of structures in the individual subgroups contain a tetrahydropyran (THP) ring instead of a THF (non-

classical AAs). The THF rings comprise the central polar core of these compounds and are usually flanked by carbinol groups. A methylated γ-lactone ring is connected to one of these carbinol carbons by a polymethylene spacer, which may contain one or more hydroxyl groups. A long hydrophobic side chain is linked to the other carbinol carbon on the THF core.

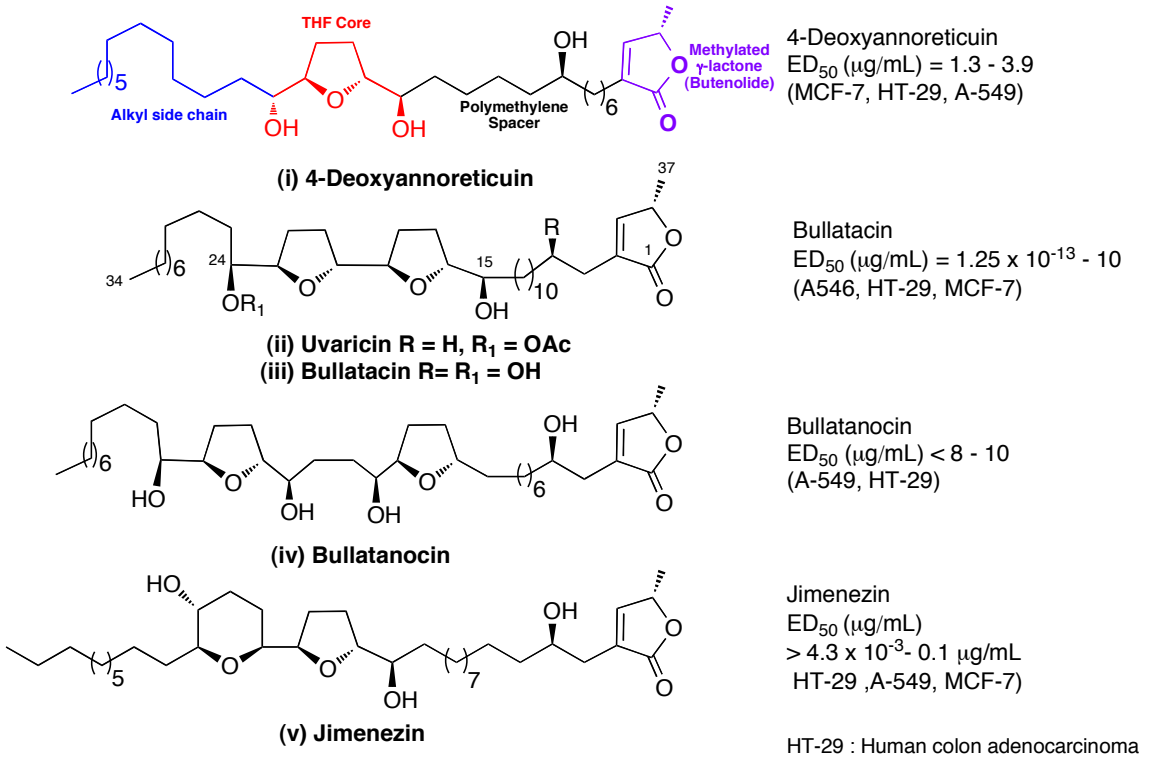


Figure 12: Representative classical and non-classical AAs.

3.2 Mode of action.

THF-containing acetogenins display diverse biological activities, such as antitumor, antimalarial, antimicrobial, and pesticidal, but they are most noted for their potent cytotoxicity against a variety of tumor cell lines.[55, 56] The Upjohn Corporation performed an animal study with asimicin, bullatacin and bullatacinone analogs. The test compounds were all active in mice bearing intraperitoneal implanted L1210 leukemia. Bullatacin effective at only 50 microgram/kg was over 300 times more potent than paclitaxel, and almost equivalent to cisplatin against L1210, with much less weight loss than these standards.[57]

The cellular target is believed to be the reduced nicotinamide adenine dinucleotide (NADH): ubiquinone oxidoreductase (complex I), a membrane-bound protein of the mitochondrial electron-transport system.[58] Bullatacin, one of the most potent AAs, has no structural similarity to known complex I inhibitors such rotenone or piericidin A, even though bullatacin and these compounds both act at the terminal electron transfer step of complex I (Figure 13). Cancer cells may be more sensitive to AAs than normal cells because they have a taxed ATP supply. In addition to triggering necrotic and apoptotic mechanisms of cell death, the disruption of ATP production may

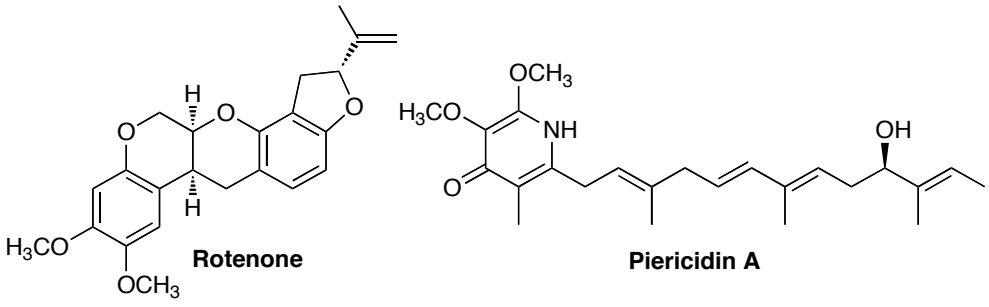


Figure 13: Known complex I inhibitors.

also lead to inhibition of the energy-dependent P-glycoprotein drug efflux pump that is over-expressed in multi-drug resistant (MDR) cells. The latter has been suggested as a reason why certain AAs are effective against MDR tumors.[59,60] There is also evidence that AAs may induce apoptosis (programmed cell death) but the mechanism of this process is not clear.[61]

Based on ^{1}H NMR spectroscopic and calorimetric studies of AAs in liposomal membranes, the McLaughlin group proposed a model for the binding of AAs in the membrane environment.[62,63] The polar cyclic ether region interacts with the hydrophilic region of the membrane and acts as an anchor to the membrane water interface. The simple hydrocarbon chain is buried in the hydrophobic region of the membrane and presents the butenolide segment to a preferred region on the enzyme (Figure 14).[64]

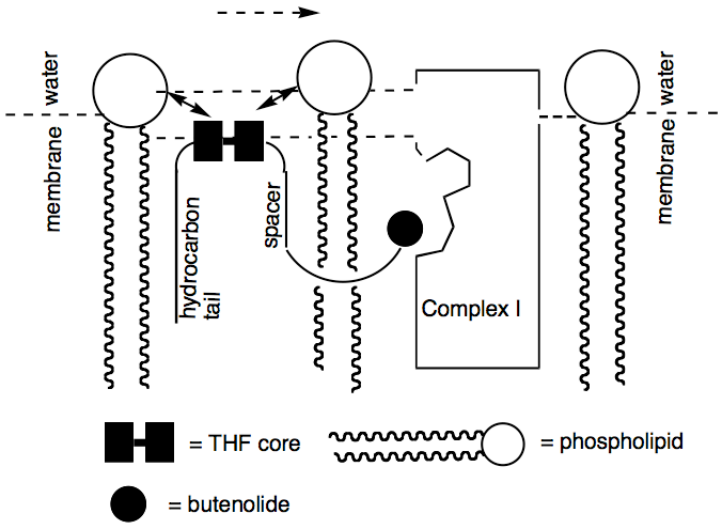


Figure 14: Mode of action of the AAs

3.3 Structure-activity relationship (SAR) studies.

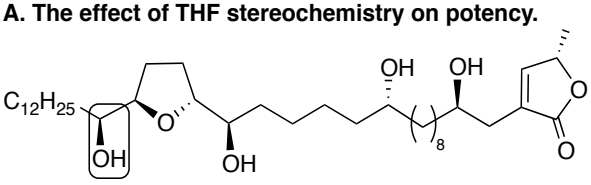
Several reviews on SAR studies for the THF-containing AAs have been published.[65,66,67,68] The structural requirements of the acetogenins do not appear to be particularly

specific. While the bicyclic ether core is integral for activity, different arrangement and numbers of the THF rings can be tolerated. In general, a specific stereochemistry in the THF does not appear to be critical for activity, although certain motifs appear to be more active against individual cell lines.[69] This is illustrated by annoglacin A and B, which differ only in the stereochemistry about the THF ring, but show comparable activity against three different cell lines (Figure 15A). These SARs are consistent with the model of the THF core as an anchor to the mitochondrial membrane, since this picture requires a polar core but does not suggest a high degree of stereochemical specificity.

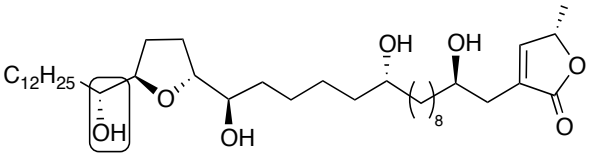
According to the conformational model, the polymethylene spacer controls positioning of the butenolide segment in the ubiquinone domain. The observation of a decrease in activity as the alkyl spacer length was shortened or lengthened (less than seven carbons or more than thirteen) is consistent with this hypothesis. Even a two-carbon decrease in length results in a drastic change in activity (Figure 15B). These results also suggest cooperative binding of the THF core and the butenolide moieties. It has been observed that a high degree of oxygenation on the alkyl spacer, i.e. more than three hydroxyl groups, significantly decreases activity. In the context of the binding model, this may be an indication that too many hydroxyl groups in this spacer region may negatively impact on the presentation of the γ-lactone to the ubiquinone domain. Subtle changes in the lengths of the hydrocarbon chain and the spacer may also have a significant effect on activity. For example, McLaughlin and co-workers evaluated the effects of shifting the hydroxyl-flanked THF ring system along the carbon backbone either toward the γ-lactone ring or toward the alkyl side chain (Figure 15C).[70] Bullatacin (in which the THF ring system is shifted two carbons toward the lactone ring relative to

squamocin) showed a significant increase in potency.

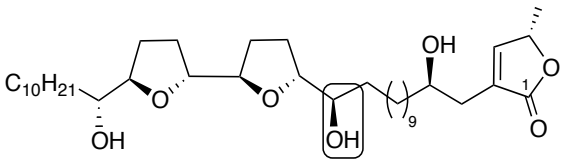
A. The effect of THF stereochemistry on potency.



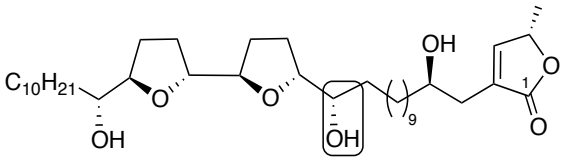
Annoglacin A
ED_{50} (μg/mL)= 9.6 x 10^{-4} (MCF-7),
5.3 x 10^{-3} (HT-29), 5.3 x 10^{-3}(A-549)



Annoglacin B
ED_{50} (μg/mL)= 6.2 x 10^{-4} μg/mL (MCF-7)
5.3 x 10^{-3} (HT-29), 2.8 x 10^{-3}(A-549)

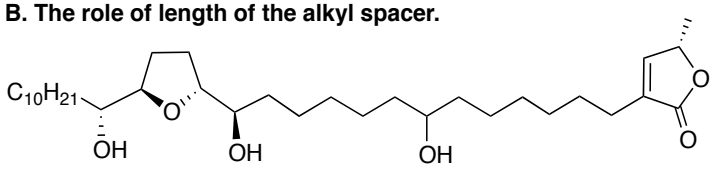


Asimicin
IC_{50} (nM)= 0.51 (HCT)

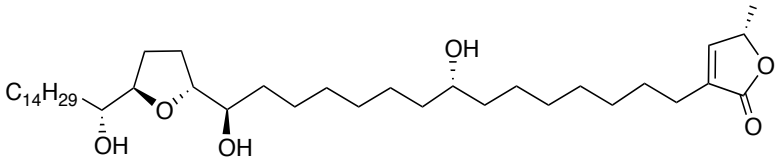


Asimicin analog
IC_{50} (nM)= 27 (HCT)

B. The role of length of the alkyl spacer.

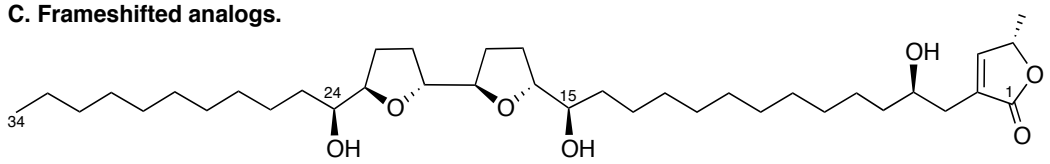


4-Deoxyannoreticuin
ED_{50} (μg/mL)= 2.2 (MCF-7)
3.9 (A-549), 1.7 (HT-29)

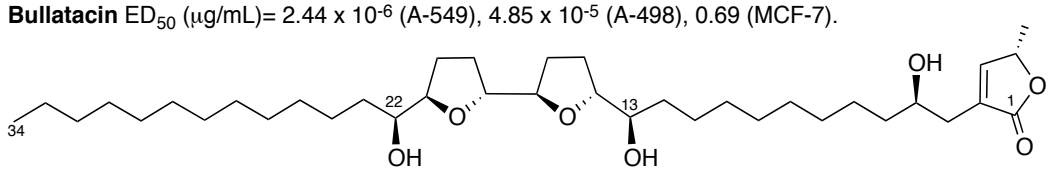


4-Deoxyannomontacin
ED_{50} (μg/mL)= 5.8 x 10^{-3} (MCF-7)
0.14 (HT-29), 6.5 x 10^{-7} (A-549)

C. Frameshifted analogs.



Bullatacin ED_{50} (μg/mL)= 2.44 x 10^{-6} (A-549), 4.85 x 10^{-5} (A-498), 0.69 (MCF-7).



Squamotacin ED_{50} (μg/mL)= 2.77 x 10^{-2} (A-549), >1.0 (A-498), >1.0 (MCF-7).

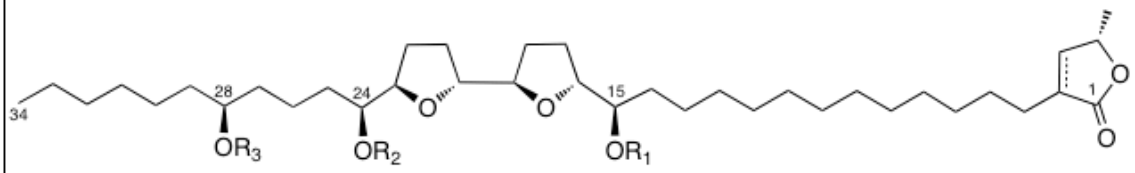
Figure 15: SAR studies.

Although most SAR studies have compared cytotoxicity values, other SAR studies have focused on inhibition of complex I (NADH oxidase activity). Of the compounds screened against both complex 1 inhibition and cytotoxicity, there is a general, direct correlation between NADH inhibition and cytotoxicity. Generally activity against complex I is two to three orders of magnitude higher than inhibition of cellular proliferation.[71]However, it is noteworthy that while bullatacin shows IC_{50} values in the 1.2-1.6 nM range in complex I inhibiton assays, these values are amplified in cytotoxicity assays (Figure 15). This implies that the mechanism of cytotoxicity may be more complex than simply inhibition of complex I.

3.4 Analog Design.

Although there are many cases of high selectivity for certain tumor cell lines over others, the high toxicity of AAs to both tumor and normal cells presents a major drawback to drug development. Given the high potency of AAs with very diverse THF core regions, we hypothesize that a mono- or di-saccharide core may be an effective replacement for the THF or bis-THF core. These analogs are to be distinguished from the glycosylated AAs examined by Hocquemiller and co-workers.[72] In this study, mono and bis- glucoside derivatives of squamocin and dihydrosquamocin were evaluated. One of the mono-glycosylated analogs of dihydrosquamocin showed low nanomolar level activity against KB (human epidermoid carcinoma) and L1210 (mouse lymphocytic leukemia) and was about one or two orders of magnitude less active than dihydrosquamocin. Another mono-glucoside analog was only assayed against L1210 and

was about 10 times less active. In comparison, the bis-glucoside of squamocin showed micromolar level activity, which was was about five orders of magnitude less than that of squamocin. Interestingly, one of the mono-glucosides of dihydrosquamocin was two orders of magnitude less active against a normal monkey cell line, compared to human epidermal carcinoma cells. However, wider screening of this compound is needed to determine if this compound is generally more selective for tumor vs. normal cells. This study also examined carbohydrate analogs with ether and ester groups. Some of these structures showed comparable activity to the parent AAs, but their relative activity compared to the parent AAs may be more dependent on their overall hydrophilicity/hydrophobicity profile than on their glycosylated substructures.



	R_1	R_2	R_3
Squamocin	H	H	H
Dihydro-squamocin	H	H	H
Gly-1D	H	α-glucose	H
Gly-2D	H	β-glucose	H
Gly-4D	β-glucose-Ac	β-glucose-Ac	H
	EC_{50} (μM)		
	KB	VERO	L1210
Squamocin	0.000016	0.016	<0.0004
Dihydro-squamocin	0.00024	0.016	<0.00025
Gly-1D	0.0019	0.6	0.01
Gly-2D			0.08
Gly-4D	1.05	1.05	10
	Legend		
	KB	Human epidermoid carcinoma cells	
	VERO	Monkey kidney epithelial cells	
	L1210	Mouse lymphocytic leukemia cellls	

Figure 16: Glycosylated acetogenins

Glycosylated structures and carbohydrate-like analogs of the type proposed in this thesis have several implications for drug design. First, these carbohydrate motifs may bind to lectins that are overexpressed on certain tumors, which could increase tumor selectivity.[73] Second, the hydrophilicity of the carbohydrate framework may enhance drug efficacy by increasing water solubility. Third, a wide variety of analogs would be possible for SAR studies. The specific goal of this study is to synthesize and evaluate the antitumor activity of a library of AA analogs, in which the cyclic ether core has been replaced with a monosaccharide or disaccharide residue. The synthesis and biological findings are presented herein.

3.5 Synthesis of AA analogs.

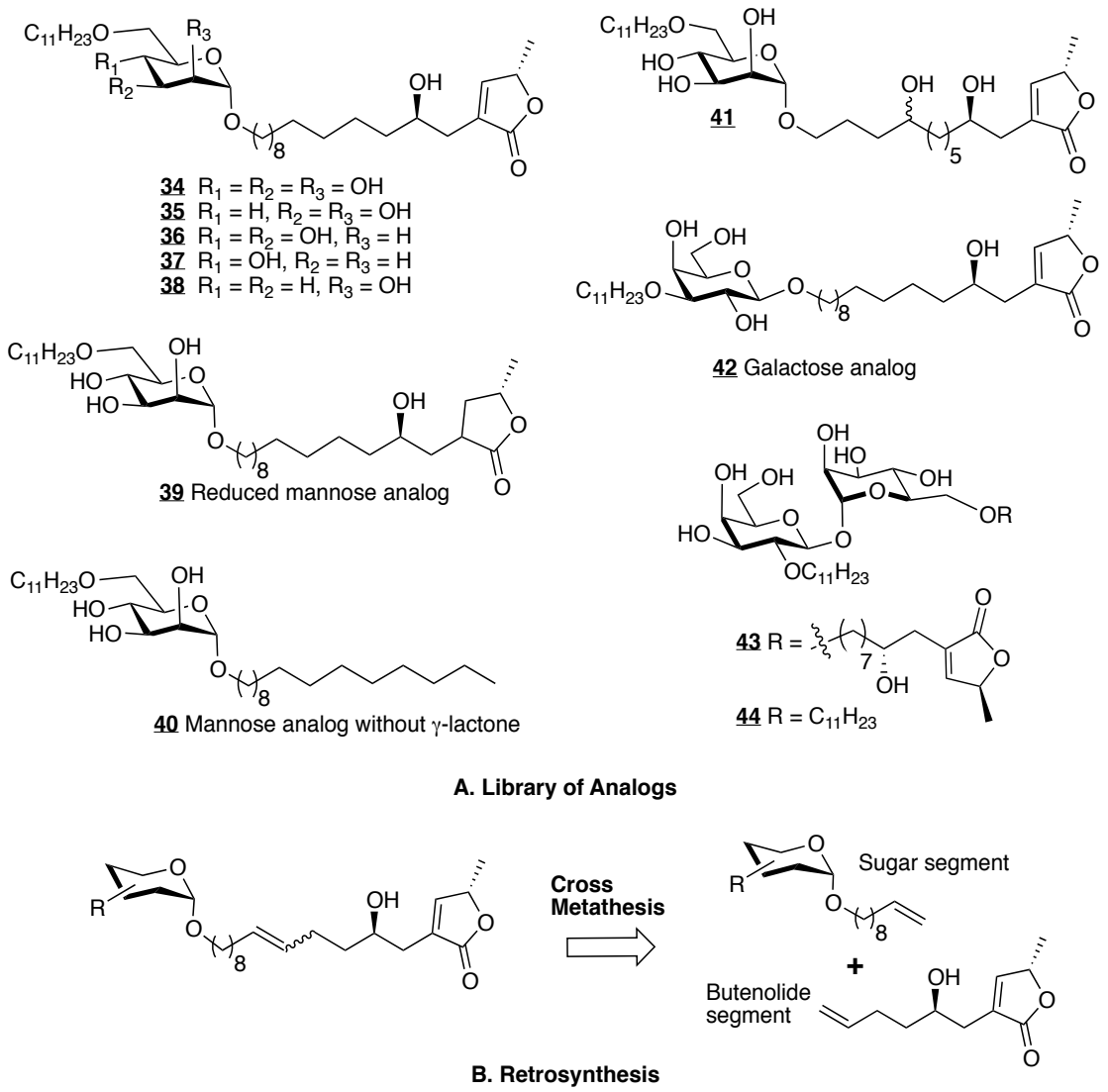


Figure 17: AA analogs and retrosynthesis.

Analogs **34**-**38** contain an α-mannopyrannoside template with varying degrees of hydroxylation (Figure 17A). These structures were designed to probe the importance of individual alcohol groups and changes in hydrophobicity. Analog **41** contains an additional alcohol in the spacer region and is interesting because the number of alcohols in this region of the natural AAs affect activity. Analog **42**, derived from β-D-galactose, was designed to test other monosaccharide frameworks. Analogs **39** and **40** are probes to

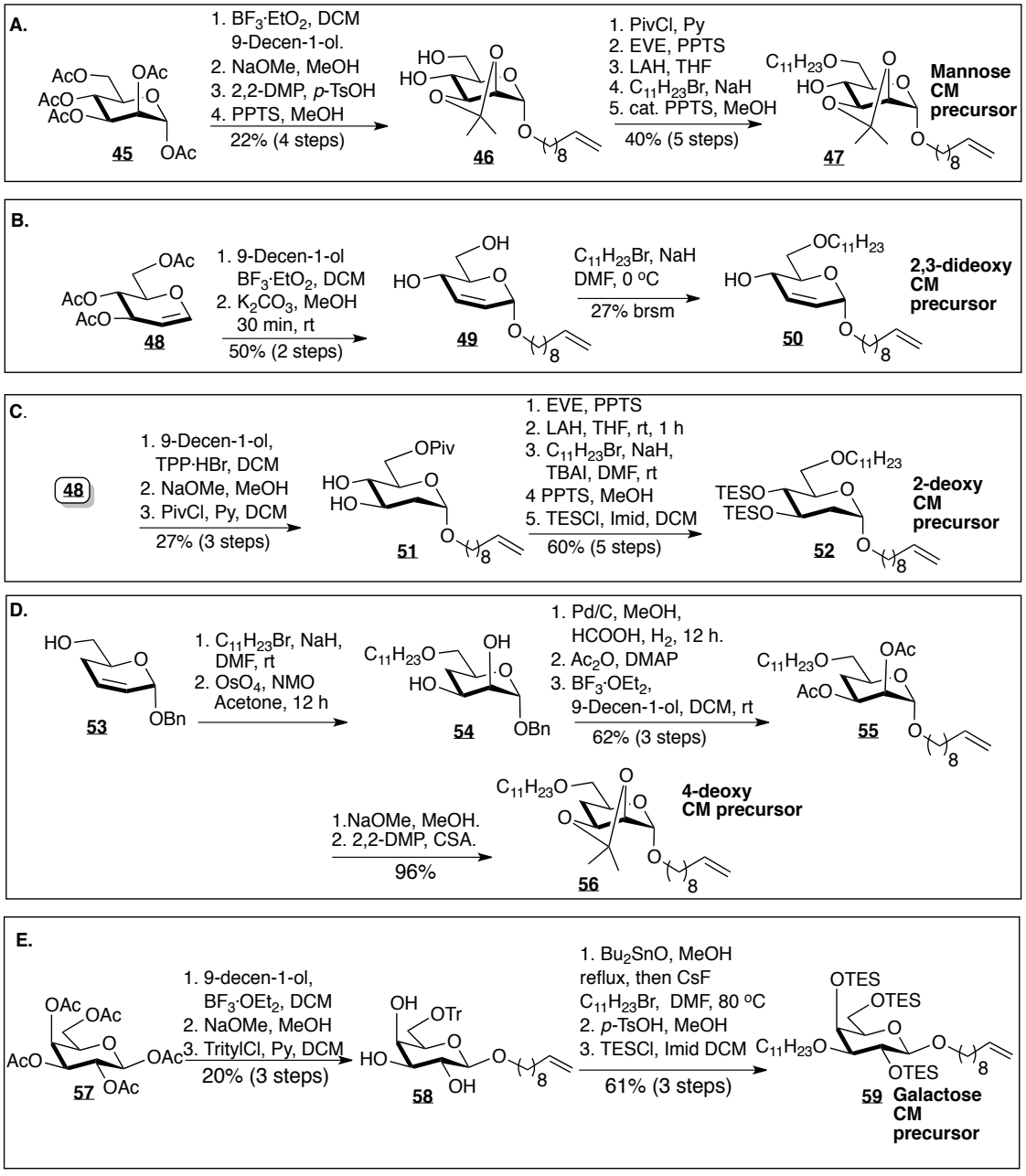
evaluate the requirements with respect to the butenolide moiety. Finally, **43** and **44** were designed as representative disaccharide analogs.. The specific 1,1-linked Gal-Man motif is selected because analogs containing this template bind to selectins, a set of cell-surface lectins.[74] Therefore AAs like **43** and **44** may selectively target tumors that overexpress selectins.[75] A highly convergent strategy in which the sugar and the butenolide segments are separately constructed and then connected via an alkene cross metathesis (CM) reaction was envisaged (Figure 17B).

3.5.1 Sugar Segments.

The fully oxygenated mannose alkene segment was easily obtained from penta-*O*-acetylated mannose **45** in five steps (Scheme 4A). The peracetylated mannose was first treated with 9-decen-1-ol and $BF_3 \cdot OEt_2$ to obtain the 9-decenyl glycoside. Removal of the acetates with NaOMe in methanol yielded the corresponding tetraol, which was converted to the 2,3:4,6-di-*O*-isopropylidene **46**. Selective removal of the 4,6-*O*-isopropylidene group under mildly acidic conditions and selective 6-*O*-alkylation gave undecyl ether **47**.

The 2,3-dideoxy- and 2-deoxy-mannose sugar segments **50** and **52** respectively, were synthesized from a common precursor, tri-*O*-acetyl glucal **48** (Scheme 4B, C). The synthesis of the 2,3-dideoxy CM precursor began with a Ferrier rearrangment performed on glucal **48** with 9-decen-1-ol in the presence of $BF_3 \cdot OEt_2$.[76] The resultant diene, which was obtained as a single α-glycoside, was treated with K_2CO_3 in methanol to give diol **49**. Selective 6-*O*-alkyation of **49** gave undecyl ether **50**.

For the 2-deoxy target, glucal **48** was treated with triphenylphosphine hydrobromide and 9-decen-1-ol to afford the 9-decenyl 2-deoxy-pyranoside derivative as a single α-glycoside (Scheme 4C). [77] Deacetylation of this product followed by selective pivaloylation of the derived triol, afforded **51**. EVE protection on diol **51** and removal of



Scheme 4: Synthesis of sugar alkenes 47, 50, 52, 55 and 59.

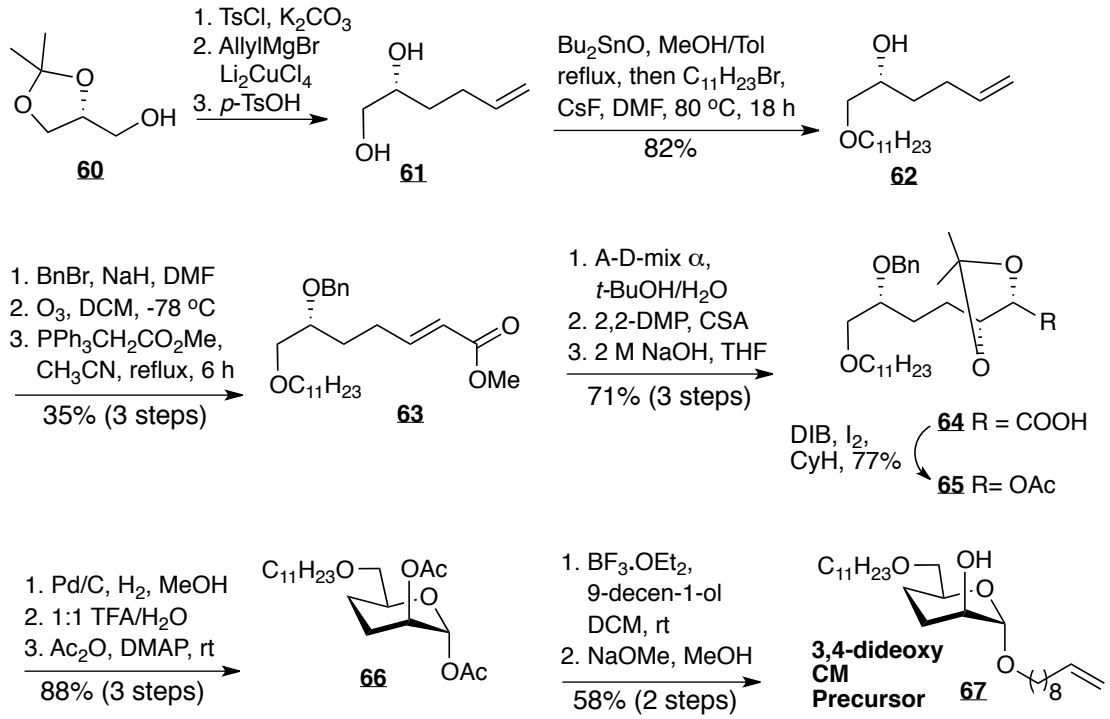
the pivalolate ester in the product, followed by 6-*O*-alkylation of the derived primary alcohol with 1-bromoundecane, produced the corresponding undecyl ether. Replacement of EE with triethylsilyl protecting groups in this material gave **52**, the 2-deoxy-pyranoside alkene for the CM step.

The 4-deoxy-mannose segment **56** was obtained from known pyranoside alkene **53**, which was obtained in five steps from glucal **48**, using a previously established method (Scheme 4D).[78] Conversion of **53** to the undecyl ether followed by OsO_4 dihydroxylation provided benzyl 4-deoxy-mannose glycoside **54**. Hydrogenolysis of **54** afforded the derived pyranose, which was converted to the corresponding tri-*O*-acetate under standard conditions. Treatment of the tri-*O*-acetate with $BF_3 \cdot OEt_2$ and 9-decenol gave the 4-deoxy-α-mannopyranoside **55**. Finally, deacetylation protection of the resulting diol as the *O*-isoproylidene derivative gave **56**, 4-deoxymannose precursor for the CM.

The galactose precursor **59** was obtained in six steps from penta-*O*-acetyl-β-D-galactopyranose **57** (Scheme 4E). Initial $BF_3 \cdot OEt_2$ promoted glycosidation with 9-decen-1-ol followed by deacetylation afforded 9-decenyl-β-D-galactopyranoside. This compound was then treated with trityl chloride and pyridine in DCM to give trityl ether **58**. Selective alkylation at the 3-position of **58** with undecyl bromide was achieved through the established Bu_2SnO protocol.[79] Removal of the trityl ether and global protection with triethylsilyl groups provided the CM alkene substrate **59**.

The 3,4-dideoxy-mannose precursor was prepared from 1,2-*O*-isopropylidene-D-glycerol **60** (Scheme 5). Compound **60** was tosylated and the product was treated with

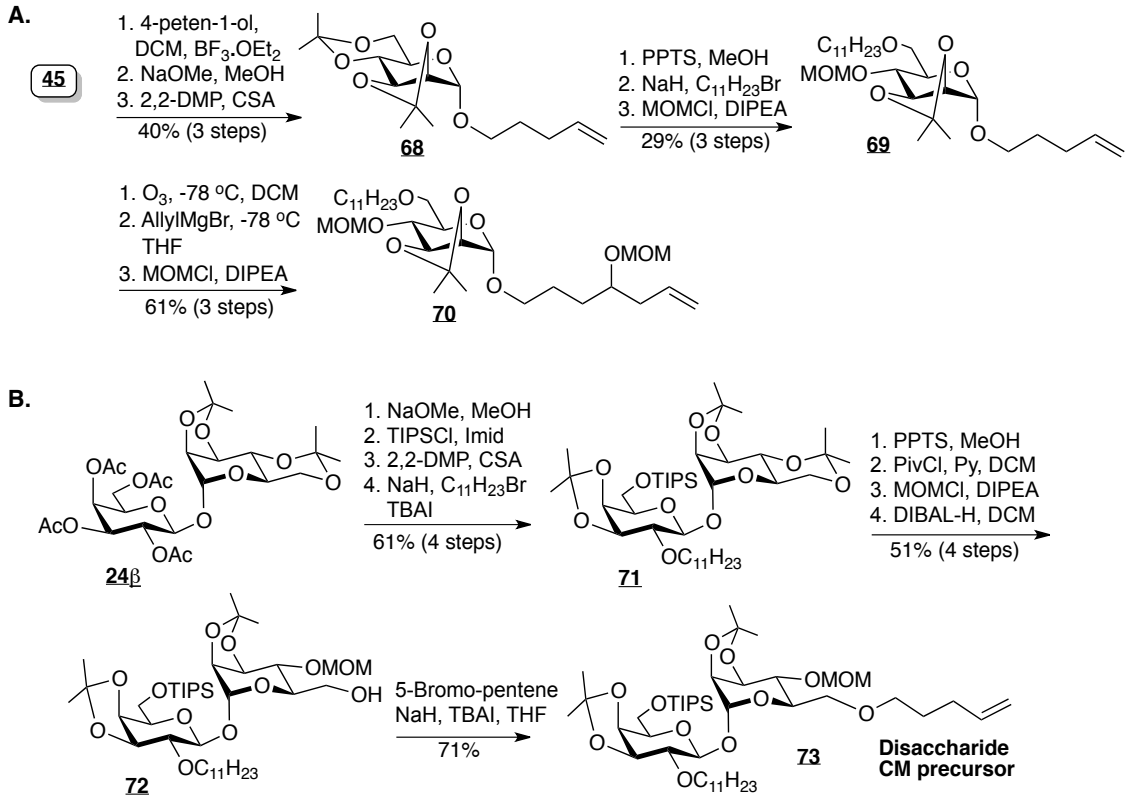
allylMgBr in the presence of catalytic Li_2CuCl_4. Acid hydrolysis on the product from this step provided dihydroxyalkene **61**.[80] Selective Bu_2SnO-mediated *O*-alkylation of the primary alcohol in **61**, with undecylbromide afforded undecyl ether **62**. Protection of the secondary hydroxyl group in **62** followed by ozonolysis of the terminal alkene and reaction of the resulting aldehyde with methoxycarbonylmethyltriphenylphosphonium bromide gave α,β-unsaturated methyl ester **63**. Asymmetric dihydroxylation (AD-mix α) [81] on **63**, followed by sequential isopropylidenation of the resultant diol and hydrolysis of the methyl ester provided carboxylic acid **64**. Suárez fragmentation on **64** afforded acetate **65.** [82] Hydrogenolysis of **65** followed by treatment of the product with aqueous TFA gave the derived lactol, which was converted to the diacetate derivative **66**. A standard glycosidation on **66** with 9-decen-1-ol and $BF_3 \cdot OEt_2$ and deacetylation of the



Scheme 5: Synthesis of the 3,4-dideoxy mannose alkene.

product gave **67**, the 3,4-dideoxy-pyranoside alkene for CM.

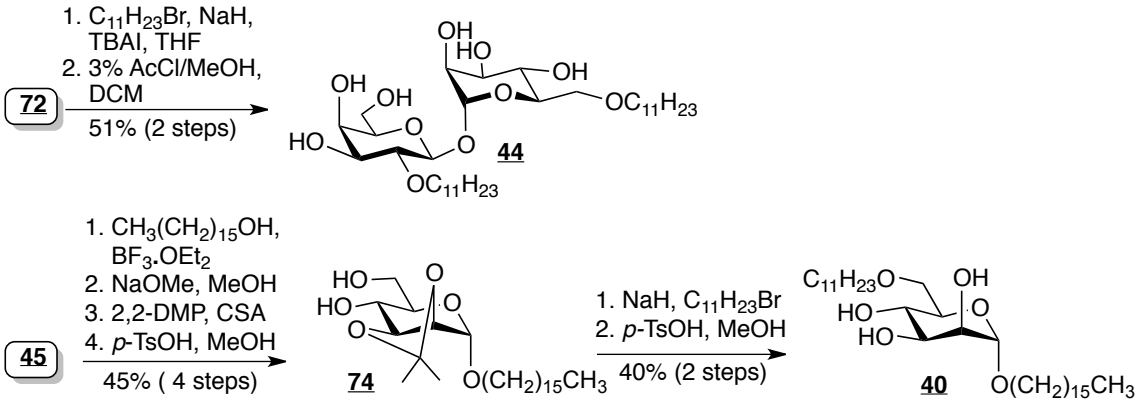
For the sugar alkene 70, penta-*O*-acetylated-α-D-mannopyranoside **45** was converted to the derived α-pentenyl glycoside via the standard glycosidation procedure with 4-pentenol and BF_3•OEt_2 (Scheme 6A). Deacetylation of this material followed by isopropylidenation of the product, gave **68**. Selective cleavage of the 4,6-*O*-isopropylidene in **68**, followed by selective alkylation of the primary alcohol and MOM protection of the derived secondary alcohol, gave undecyl ether **69**. Ozonolysis of the terminal alkene gave the corresponding aldehyde, which was treated with allylMgBr to give a mixture of homoallylic alcohols. This product was converted to **70** for the CM reaction.



Scheme 6: Synthesis of CM precursors 70 and 73.

Synthesis of the disaccharide alkene **73** started from the 1,1-linked disaccharide **24β**, which was described in Chapter 2 (Scheme 1). This material was first converted to the undecyl ether **71** in four steps using standard protecting group transformations (Scheme 6B). Selective removal of the 4,6-*O*-isopropylidene group on the mannose ring generated the 4,6-diol, which was converted to the primary alcohol **72** in three routine protecting group steps. Alkylation of **72** under standard conditions with 5-bromo-1-pentene provided **73**, the disaccharide alkene precursor for the CM reaction.

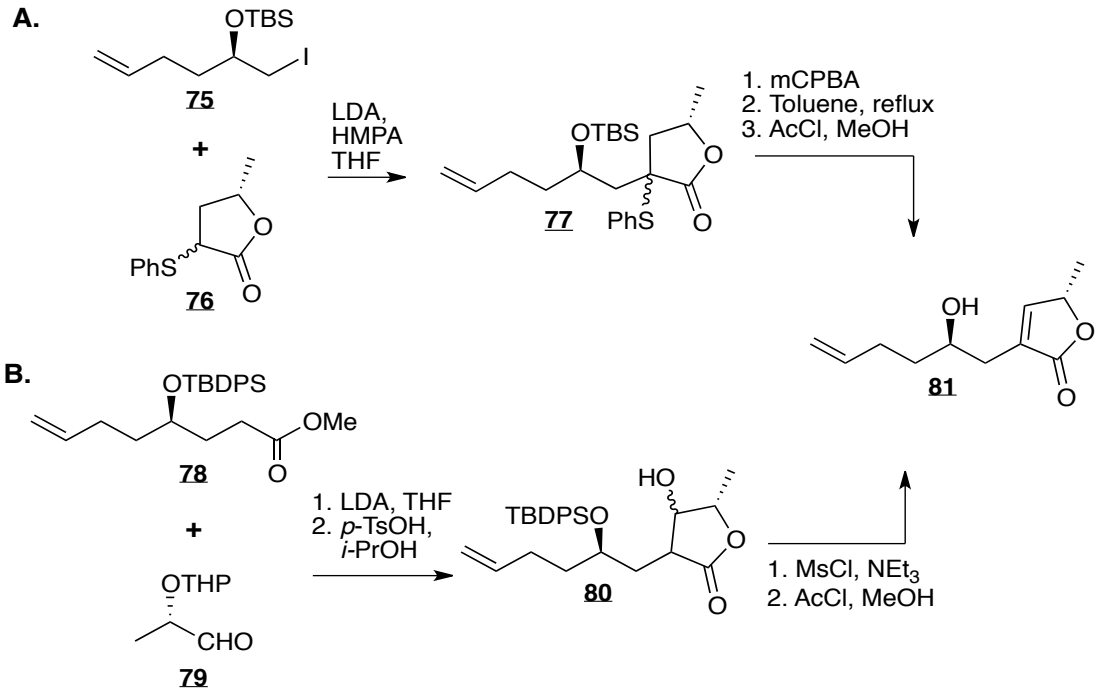
The di-*O*-alkyl analog **44** was prepared by straightforward alkylation and protecting group operations on disaccharide **72** (Scheme 7). The mannose di-alkyl analog **40** was obtained from penta-*O*-acetyl–α-D-mannopyranose **45**. BF_3•OEt_2 promoted glycosidation of **45** with 1-hexadecanol provided the corresponding α-glycoside. This material was converted to diol **74** through standard protecting group transformations. Selective deprotection of the 4,6-isopropylidene group, selective primary alcohol alkylation in the resulting diol, followed by hydrolysis of the 2,3-*O*-isopropliydene, gave the 6-*O*-alkylated mannoside **40**.



Scheme 7: Synthesis of dialkyl AA analogs.

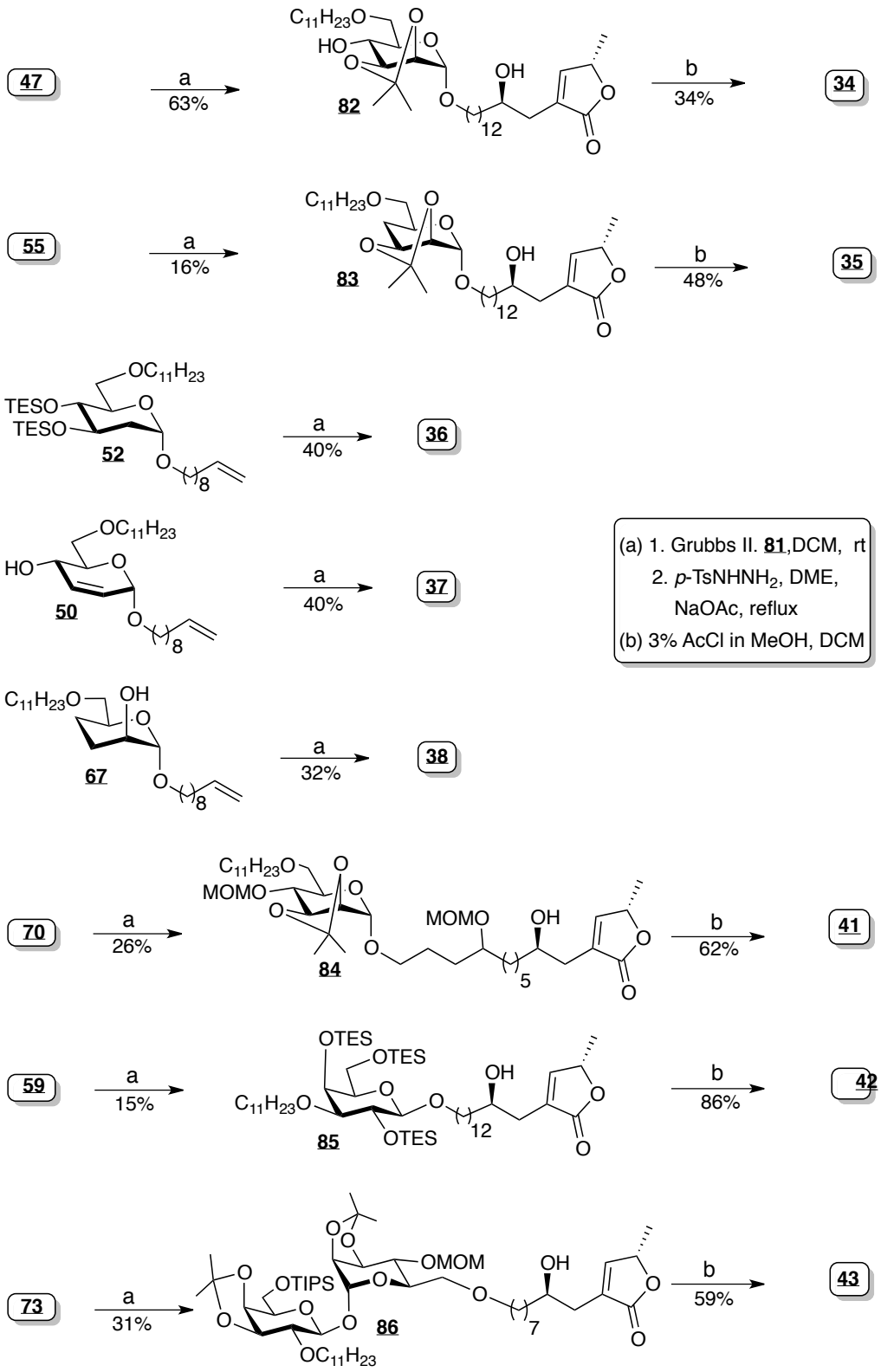
3.5.2 The butenolide segment.

The butenolide segment was synthesized using two previously described methods.[83, 84] The keys steps in these syntheses are summarized in Scheme 8. In Method A, Iodide **75** was obtained from (*R*)-epichlorohydrin in three steps. Known lactone **76** was deprotonated with LDA and treated with **75** to give the alkylated lactone **77**.[85] An oxidation-elimination sequence on thioether **77** and removal of the TBS ether in the product gave the butenolide segment, **81**. In method B, aldehyde **79** was obtained from (*S*)-(-)-methyl lactate in two steps.[86] Reaction of the lithium enolate from ester **78** and **79** followed by treatment of the crude product with acid, led to lactone **80** as a mixture of alcohols. Conversion of **80** to the mesylate and *in situ* elimination of the mesylate, followed by acid promoted removal of the silyl ether in the product, gave **81**.



Scheme 8: Synthesis of butenolide precursors.

3.5.3 Cross metatheses.



Scheme 9: Cross metatheses and final deprotection steps.

The CM reactions of the carbohydrate and butenolide alkene partners were performed with the Grubbs 2nd generation catalyst (Grubbs II). Generally the alkene partners were dissolved in anhydrous DCM and the solution was deoxygenated by purging with N_2 (Scheme 9). Grubbs II was then added and the reaction was monitored by TLC. Upon completion, the solvent was removed and the desired CM product was isolated by FCC. Selective reduction of the bridging alkene in the CM product over the alkene in the butenolide was achieved by diimide reduction using tosylhydrazide and sodium acetate.[87] Thus, via this two-step, CM-reduction protocol, sugar alkenes **47**, **55**, **52**, **50**, **67**, **70**, **59** and **73** were converted to products **82**, **83**, **36**, **37**, **38**, **84**, **85** and **86** respectively in 15 - 63% yields. Three of the products in the desired AA library, **36**, **37** and **38** were produced directly from this sequence. It should be noted that in all cases the alkene formed in the CM reaction was selectively reduced over the alkene in the butenolide. Also in the case of **37**, the alkene within the sugar ring was also selectively reduced over the alkene in the butenolide. Protecting group removal, using 3% acetyl chloride in methanol and DCM, on **82 - 86** provided the remaining AA analogs **34**, **35**, **41 - 43** in 34 - 86% yields. Analog **39** was obtained in quantitative yield from **34** via standard hydrogenation using Pd/C as catalyst.

3.6 Results and discussion

The cytotoxicity of the AA analogs **34 - 44** was evaluated against different cancer cell lines using the CellTiter-Glo® luminescent cell viability assay. This is a homogeneous method to determine the number of viable cells in culture based on quantitation of the ATP present, which signals the presence of metabolically active cells. Test compounds were exposed to four different cell lines, namely human breast cancer,

human cervical cancer, Jurkat (leukemia) cells, and prostate cancer cells. In general, the monosaccharide compounds showed activity in the low micromolar range, with the mannose analog **34** being the most active. This activity was comparable to that of the natural acetogenin, 4-deoxyannorecticiun, which was previously synthesized in this laboratory, and used here as a positive control (Figure 15B). [37] Interestingly, the degree of oxygenation of the monosaccharide ring did not have any significant effect on activity. The activity of the Gal analog **42**, showed noticeably lower activity compared to the other monosaccharide AAs. This result suggests that the relative orientation of the alkyl chain and the butenolide on the pyranoside core may have an impact on activity. AA analogs **39** and **40** where the butenolide was omitted and reduced respectively, showed no significant activity up to 100 μM. A similar effect has been observed for the THF containing AAs, which suggests that the mechanism of action of THF and sugar AAs may be the same.

Compound	Jurkat	HeLa	MDA MB231	PC-3
34	10.11 ±0.84	15.17± 0.98	21.18 ± 3.30	32.56 ±0.76
35	19.66 ± 3.55	23.31 ± 1.07	36.71 ± 4.88	52.83 ±1.21
36	13.29 ± 2.72	16.30 ±1.21	16.81 ±3.51	44.04 ±0.96
37	44.53 ±14.72	82.76 ±14.86	>100	ND
38	10.69 ± 1.02	13.93 ±1.25	21.75 ±2.95	28.73 ±0.80
41	13.09 ± 2.10	11.31 ±1.60	30.69 ±1.79	31.43 ± 1.17
42	78.17 ± 7.68	50.32 ± 9.07	73.55 ± 14.49	ND
4-deoxy-annoreticuin	17.79 ± 1.56	20.99 ± 2.71	34.16 ±1.83	43.05 ±2.40

Table2: IC_{50} values for AA analogs.

Note: 34-38, 41 and 42 (Figure 17); 4-deoxyannorecticuin (Figure 15B)

Finally, the disaccharide analog **43** was not active up to 100 μM. However, with only one disaccharide AA tested, it is not known if this is generally the case for disaccharide AAs.

Future directions in this project include varying the lengths of hydrocarbon chain and spacer to increase potency and evaluating the mannose analogs against tumors that overexpress mannose binding carbohydrates to see whether there is any selectivity for such.[88] In a similar vein, AAs derived from galactose and galactosamine templates would be screened against cells that overexpress receptors for these carbohydrate motifs. [89]

3.8 Experimental

3.8.1 Cytotoxicity assays and IC_{50} calculations.

Please note: These experiments were performed by Dr. Himanshu Garg, Center for Excellence for Infectious Disease, Texas Technical University Health Sciences Center, El Paso, Texas.

Human cervical cancer cell line HeLa, breast cancer cell line MDA-MB231 were cultured in Dulbecco's modified Eagles Medium supplemented with 10% FBS and penicillin streptomycin (5000 U/ml). T cell leukemia cell line Jurkat, were maintained in RPMI media supplemented with 10% FBS and penicillin streptomycin (5000 U/ml). Stock solutions of compounds were made in DMSO at a concentration of 20 mM. Serial dilutions of the test compounds were prepared in media at 37 °C before addition to cells.

Cytotoxicity was determined on various cell lines by incubating with serial dilutions of the test compounds for 48 h. Cell viability was determined using Cell titer Glo (Promega

Corp. Madison, WI) as per the manufacturer's instructions. Cell titer Glo assay is based on the measurement of ATP produced by healthy viable cells. Data were normalized to no compound control and expressed as percent viability. Dose response curves obtained from serial dilution of the compounds were fitted using Sigma plot analysis software. Each experiment was repeated thrice and the average IC_{50} was calculated along with Standard deviation.

3.8.2 Synthesis

General procedures: Unless otherwise stated, all reactions were carried out under a nitrogen atmosphere in oven-dried glassware using standard syringe and septa technique. ^{1}H and ^{13}C NMR spectra were obtained on a Bruker 500 MHz spectrometer, in $CDCl_3$. Chemical shifts are relative to the deuterated solvent peak and are in parts per million (ppm). Assignments for selected nuclei were determined from ^{1}H COSY experiments. Thin layer chromatography (TLC) was done on 0.25 mm thick precoated silica gel HF_{254} aluminum sheets. Chromatograms were observed under UV (short and long wavelength) light, and were visualized by heating plates that were dipped in a solution of ammonium (VI) molybdate tetrahydrate (12.5 g) and cerium (IV) sulfate tetrahydrate (5.0 g) in 10% aqueous sulfuric acid (500 mL). Flash column chromatography (FCC) was performed using silica gel 60 (230-400 mesh) and employed a stepwise solvent polarity gradient, correlated with TLC mobility. Solvents were purified by standard procedures or used from commercial sources as appropriate. High resolution mass spectra (HRMS) were obtained on an Ultima Micromass Q-TOF Mass Spectrometer at the Mass Spectrometry facility at Hunter College, CUNY.

Mannose CM Precursor 47

9-Decenyl 2,3-*O*-isopropyliene-α-D-mannopyranoside 46. To a solution of penta-*O*-acetylated mannose **45** (5.41 g, 13.9 mmol) in DCM (45 mL) were added 9-decen-1-ol (3.5 mL, 21.0 mmol) and $BF_3 \cdot OEt_2$ (5.7 mL, 59.0 mmol). The reaction mixture was stirred for 24 h and then was quenched with saturated aqueous $NaHCO_3$ solution. The mixture was extracted with DCM, dried, and concentrated *in vacuo*. FCC of the residue gave the decenyl glycoside (2.4 g, 35%), R_f = 0.32 (25% EtOAc/petroleum ether). The glycoside was then treated with NaOMe (0.3 g, 5.6 mmol) in MeOH (25 mL). The reaction mixture was stirred for 1 h and then was adjusted to pH 6 with 1 M HCl in MeOH. The mixture was concentrated *in vacuo* to give the crude tetraol, which was dissolved in DCM (25 mL) and treated with 2,2-DMP (10 mL, 82.0 mmol) and *p*-TsOH (100 mg, 0.50 mmol). The reaction mixture was stirred for 1 h and quenched with saturated aqueous $NaHCO_3$ solution, with similar work-up as described previously. FCC of the residue gave the 2,3,4,6-di-*O*-isopropylidene mannopyranoside (1.7 g, 86%), R_f = 0.88 (30% EtOAc/petroleum ether). To a solution of 2,3;4,6-di-*O*-isopropylidene-α-D-mannopyranoside (1.7 g, 4.3 mmol) in MeOH (15 mL) was added PPTS (640 mg, 2.1 mmol). The reaction mixture was stirred for 13 h and then was quenched with Et_3N. The mixture was concentrated *in vacuo*. FCC of the residue gave the 4,6-diol **46** (1.11 g, 73%). R_f = 0.14 (30% EtOAc/petroleum ether) ^{1}H NMR 1.32 (broad, 10H), 1.39 (s, 3H), 1.55 (s, 3H), 2.07 (m, 2H), 2.57 (d, 1H, *J* = 4.9 Hz), 3.46 (m, 1H), 3.73 (m, 4H), 3.76 (m, 3H), 3.86 (m, 3H), 4.95 (m, 1H), 5.02 (s, 2H), 5.83 (m, 1H). ^{13}C NMR δ 28.0, 29.1, 29.2, 29.5, 29.6, 34.0, 62.9, 68.2, 69.7, 70.2, 75.7, 78.2, 97.5, 109.8, 114.4, 139.4. HRMS (ESI) calcd. for $(M+NH_4)^+$ $C_{19}H_{38}NO_6$, 376.2694 found 376.2697.

9-Decenyl 2,3-*O*-isopropyliene-6-undecyl-α-D-mannopyranoside 47. To a sample of diol **46** (1.11g, 3.1 mmol) in DCM (20 mL) was added pyridine (2.5 mL, 31.0 mmol) and PivCl (0.46 mL, 3.1 mmol). The reaction mixture was stirred for 4 h and then was concentrated *in vacuo*. FCC of the residue gave the pivaloate ester of the primary alcohol (1.13g, 82%), R_f= 0.4 (20% EtOAc/petroleum ether). This material was then treated with EVE (9 mL, 94.0 mmol) and PPTS (100 mg, 0.40 mmol) in DCM (3 mL). The reaction mixture was stirred for 1 h and then was quenched with Et_3N. Removal of the volatiles *in vacuo* and FCC of the residue gave the 4-*O*-ethoxy ethyl ether (1.30 g), R_f = 0.85 (20% EtOAc/petroleum ether), in quantitative yield. This product was dissolved in DCM (15 mL) and cooled to −78 °C. A 1 M solution of DIBAL-H in heptane (7.3 mL, 7.3 mmol) was added and the reaction mixture was allowed to warm to rt. After 3 h the reaction was quenched with saturated aqueous Rochelle's salt and extracted with EtOAc. The organic phase was dried (Na_2SO_4), filtered, and concentrated *in vacuo.* FCC of the residue afforded the derived primary alcohol (966 mg, 90%). To a solution of primary alcohol (966 mg, 2.24 mmol) in THF were added NaH (60% dispersion in mineral oil, 180 mg, 4.5 mmol), TBAI (82 mg, 0.20 mmol), and $C_{11}H_{23}Br$ (0.8 mL, 4.5 mmol). The reaction mixture was stirred for 12 h and then was quenched with water and extracted with ether. The organic phase was dried (Na_2SO_4), filtered, and concentrated *in vacuo.* After, the crude residue was treated with PPTS (100 mg, 0.40 mmol) in MeOH (15 mL) for 1 h, and the reaction was quenched with Et_3N. The solvent was removed *in vacuo,* and FCC of the residue gave the mannose CM precursor **47** (828 mg, 52%). R_f = 0.5 (15% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (t, 3H, J = 6.8 Hz), 1.27 (bs, 22H), 1.38 (s, 3H), 1.55 (s, 3H), 1.60 (m, 8H), 2.06 (m, 2H), 2.99 (bs, 1H), 3.44 (m, 1H), 3.50 (m, 2H), 3.65

(m, 1H), 3.73 (m, 4H), 4.17 (m, 2H), 4.96 (m, 3H), 5.84 (m, 1H). ^{13}C NMR δ 14.3, 22.9, 26.3 (two lines), 28.1, 29.1, 29.3, 29.5 (two lines), 29.6, 29.7, 29.8 (three lines), 32.1, 34.0, 68.0 (two lines), 71.6, 71.9, 72.3, 75.5, 78.1, 97.3, 109.6, 114.3, 139.4. HRMS (ESI) calcd. for $(M+NH_4)^+$ $C_{30}H_{60}NO_6$, 530.4415 found 530.4419.

2,3-Dideoxy mannose CM precursor 50.

9-Decenyl 2,3-dideoxy-α-D-hex-2-enopyranose 49. To a solution of tri-*O*-acetyl-D-glucal **48**, (2.5 g, 9.2 mmol) in dry DCM (30 mL) were added 9-decen-1-ol (1.8 mL, 10.1 mmol) and BF_3•OEt_2 (0.1 mL, 0.9 mmol). The reaction mixture was stirred for 1 h and then quenched with Et_3N (2 mL). The solvent was removed *in vacuo* and crude product was taken up in dry MeOH (20 mL), and anhydrous K_2CO_3 (1 g) was added to the mixture. The reaction mixture was stirred for 1 h and then filtered through a bed of Celite. Removal of the volatiles *in vacuo*, and FCC of the residue gave **49** as a white solid (1.3 g, 50%, 2 steps), R_f = 0.5 (50% EtOAc/petroleum ether); ^{1}H NMR δ 1.30 (m, 8H), δ 1.37 (m, 4H), δ 1.61 (m, 2H), δ 2.06 (m, 2H), δ 3.49 (m, 1H), δ 3.75 (m, 2H), δ 3.80 (m, 2H), δ 4.23 (dd, 1H, *J* = 1.4, 10.5 Hz), δ 4.99 (m, 3H), 5.77 (dt, 1H, *J* = 2.3, 10.3 Hz), 5.84 (m, 1H), δ 5.98 (d, 1H, *J* = 10.1 Hz). ^{13}C NMR δ 26.3, 29.0, 29.2, 29.5, 29.6, 29.9, 34.0, 62.9, 64.5, 69.1, 71.5, 94.4, 114.3, 126.6, 133.3, 139.4. HRMS (ESI) calcd for $(M+Na)^+$ $C_{16}H_{28}NaO_4$ 307.1880, found 307.1884.

9-Decenyl 6-undecyl-2,3-dideoxy-α-D-hex-2-enopyranose 50. To a solution of **49** (1.23 g, 4.33 mmol) in dry DMF (20 mL) was added NaH (60% dispersion in mineral oil, 175 mg, 4.38 mmol) and TBAI (80 mg, 0.43 mmol) and $C_{11}H_{23}Br$ (0.97 mL, 4.33 mmol) at 0 °C. The reaction mixture was stirred at this temperature for 2 h, then quenched with

water and extracted with EtOAc. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded the 6-alkylated compound **50** (268 mg, 27% brsm) as a clear oil. R_f = 0.6 (15% EtOAc/petroleum ether). ^{1}H NMR δ 0.91 (m, 3H), 1.25-1.45 (m, 22H), 1.57 (m, 8H), 1.95 (bs, 1H), 2.06 (q, 2H, *J* = 7.1 Hz), 3.46 (m, 2H), 3.65 (m, 1H), 3.79 (m, 3H), 3.88 (dd, 1H, *J* = 2.2, 10.9 Hz), 3.93 (bd, 1H, *J* = 8.9 Hz), 4.98 (m, 3H), 5.77 (dt, 1H, *J* = 2.3, 10.3 Hz), 5.84 (m, 1H), 6.08 (d, 1H, *J* = 10.3 Hz). ^{13}C NMR δ 14.3, 22.9, 26.3, 26.4, 29.1, 29.2, 29.5 (two lines), 29.6 (two lines), 29.8 (two lines), 29.9, 30.2, 32.1, 34.0, 62.9, 68.9, 69.3, 69.6, 71.2, 94.6, 114.3, 126.5, 130.8, 139.4. HRMS (ESI) calcd for $(M+Na)^+$ $C_{27}H_{50}NaO_4$ 461.3601, found 461.3599.

2-Deoxymannose CM precursor 52.

9-Decenyl 6-*O*-pivalate-2-deoxy-α-D-*arabino*-pyranoside 51. To a solution of glucal **48** (2.5 g, 9.20 mmol) in dry DCM (45 mL) were added 9-decen-1-ol (1.8 mL, 10.1 mmol) and triphenylphosphine hydrobromide (472 mg, 1.40 mmol). On complete disappearance of **48** by TLC, the mixture was concentrated *in vacuo*. The residue was taken up in dry MeOH (30 mL) and NaOMe (100 mg, 1.85 mmol) was added to the solution. The reaction mixture was stirred for 1 h and then was neutralized using 1 M HCl in MeOH. The filtrate was concentrated *in vacuo*, and the residue subjected to FCC to provide the derived triol (1.60 g, 59%, 2 steps). R_f = 0.2 (80% EtOAc/petroleum ether). To a solution of the triol (1.6 g, 5.30 mmol) in dry DCM (50 mL) were added pyridine (4.3 mL, 53.0 mmol) and PivCl (0.7 mL, 5.80 mmol). The reaction mixture was stirred for 3 h and then was diluted with EtOAc (150 mL). The organic phase was washed with 1 M HCl, saturated aqueous $NaHCO_3$ solution, dried ($NaSO_4$), filtered, and concentrated *in vacuo*. FCC of the residue gave **51** (910 mg, 45%). R_f = 0.23 (30% EtOAc/petroleum

ether). ^{1}H NMR δ 1.22 (s, 9H), 1.27 (m, 10H), 1.35 (m, 2H), 1.62 (m, 1H), 2.02 (m, 2H), 2.13 (dd, 1H, *J* = 5.1, 12.9 Hz), 2.35 (m, 1H), 3.09 (m, 1H), 3.14 (d, 1H, *J* = 3.7 Hz), 3.32 (m, 1H), 3.58 (m, 1H), 3.66 (d, 1H, *J* = 9.8 Hz), 3.97 (m, 1H), 4.10 (dd, 1H, *J* = 1.9, 12.3 Hz), 4.62 (dd, 1H, *J* = 3.7, 12.9 Hz), 4.88 (d, 1H, *J* = 3.0 Hz), 4.91 (d, 1H, *J* = 10.2 Hz), 4.97 (d, 1H, *J* = 17.2 Hz), 5.78 (m, 1H). ^{13}C NMR δ 26.4, 27.4, 29.1, 29.2, 29.6 (two lines), 29.7, 34.0, 37.3, 63.7, 67.7, 68.8, 70.6, 72.4, 97.6, 114.3, 139.4, 180.3.

9-Decenyl 3,4-di-*O*-triethylsilyl-6-undecyl-2-deoxy-α-D-*arabino*-pyranoside 52. To a solution of **51** (910 mg, 2.30 mmol) in EVE (20 mL) and DCM (10 mL) was added PPTS (200 mg, 0.80 mmol). The reaction mixture was stirred for 30 min, quenched with saturated aqueous $NaHCO_3$ solution, and extracted with ether. The organic fraction were dried (Na_2SO_4), filtered, and concentrated *in vacuo.* The crude residue was with taken up into dry THF (45 mL) and treated with LAH (447 mg, 12.0 mmol). The reaction mixture was stirred for 3 h then quenched at 0 °C with saturated aqueous Rochelle's salt (3 mL) and 2 N NaOH (3 mL). After 2 h of stirring, the mixture was diluted with water and extracted with EtOAc. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo.* FCC of the residue afforded the primary alcohol (1.05 g, 98%) R_f = 0.32 (30% EtOAc/petroleum ether). To a solution of this material (1.05 g, 2.36 mmol) in dry DMF (30 mL) were added NaH (60% dispersion in mineral oil, 283 mg, 7.10 mmol), $C_{11}H_{23}Br$ (1.6 mL, 7.10 mmol), and TBAI (87 mg, 0.24 mmol). The reaction mixture was stirred for 2 h, quenched with water, and extracted with ether. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo.* The crude residue was taken up into MeOH (15 mL) and treated with PPTS (100 mg, 0.40 mmol). The reaction mixture was stirred for 1 h and then was concentrated *in vacuo*. FCC of the residue gave

9-undecenyl 2-deoxy-6-*O*-undecyl-α-D-*arabino*-pyranoside (668 mg, 61%), R_f = 0.24 (30% EtOAc/petroleum ether). To a solution of this material (668 mg, 1.45 mmol) in dry DCM (5 mL) were added triethylsilyl chloride (0.17 mL, 0.98 mmol) and imidazole (112 mg, 1.65 mmol). The reaction mixture was stirred for 2 h and then was quenched with water and extracted with ether. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded **52** (953 mg, 96%). R_f = 0.56 (5% EtOAc/petroleum ether). ^{1}H NMR δ 0.50 (q, 2H, *J* = 8.0 Hz), 0.61 (m, 10H), 0.84-0.96 (m, 18H), 1.23 (m, 23H), 1.35 (m, 3H), 1.57 (m, 8H), 2.01 (m, 3H), 3.27 (m, 1H), 3.42 (m, 3H), 3.56 (m, 4H), 3.91 (m, 1H), 4.81 (d, 1H, *J* = 2.1 Hz), 4.90 (d, 1H, *J* = 10.2 Hz), 4.97 (d, 1H, *J* = 17.1 Hz). ^{13}C NMR δ 5.4, 5.6, 6.6, 7.0, 7.1, 7.2, 14.3, 22.9, 26.4, 29.1, 29.3, 29.5, 29.6 (three lines), 29.8 (three lines), 29.9, 32.1, 34.0, 39.1, 67.3, 70.2, 71.3, 71.9, 72.0, 73.5, 97.2, 114.3, 139.4. HRMS (ESI) calcd for $(M+NH_4)^+$ $C_{39}H_{84}NO_5Si_2$ 702.5883, found 702.5878.

4-Deoxy mannose CM precursor 56.

Benzyl 4-deoxy-6-*O*-undecyl-α-D-*lyxo*-pyranoside 54. To a solution of primary alcohol **53** [78] (1.27 g, 5.80 mmol) in dry DMF (20 mL) were added NaH (60% dispersion in mineral oil, 692 mg, 17.3 mol) and TBAI (212 mg, 0.57 mmol). The reaction mixture was stirred for 15 min and then $C_{11}H_{23}Br$ (2.5 mL, 11.5 mmol) was added at rt. The reaction mixture was stirred for a further 18 h and then was quenched with water and extracted with ether. The organic phase was washed with water, dried (Na_2SO_4), filtered, and then was concentrated *in vacuo*. FCC of the residue gave the derived undecyl ether. To a solution of the undecyl ether (910 mg, 2.43 mmol) in 3:1 acetone/water (20 mL) were added NMO (284 mg, 2.40 mmol) and OsO_4 (62 mg, 0.20 mmol). The reaction

mixture was stirred for 18 h and then was quenched with $NaHSO_3$ (100 mg, 0.96 mmol). The resulting slurry was stirred for 10 min, then filtered through a bed of Celite. The solvents were removed *in vacuo* and the residue was then taken up into EtOAc and washed with water and brine. The organic phase was then dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave **54** (516 mg, 22%, two steps). R_f = 0.42 (70% EtOAc/petroleum ether). ^{1}H NMR δ 0.88 (t, 3H, *J* = 6.8 Hz), 1.26 (m, 14H), 1.60 (m, 4H), 1.80 (m, 1H), 2.12 (d, 1H, *J* = 8.5 Hz), 2.18 (d, 1H, *J* = 7.1 Hz), 3.48 (m, 5H), 3.79 (m, 1H), 3.98 (m, 1H), 4.07 (m, 1H), 4.50 (d, 1H, *J* = 11.9 Hz), 4.73 (d, 1H, *J* = 11.9 Hz), 4.99 (s, 1H), 7.33 (m, 5H). ^{13}C NMR δ 14.3, 22.9, 26.3, 29.5, 29.7, 29.8 (two lines), 31.4, 32.1, 65.8, 67.6, 69.2, 69.4, 72.0, 73.4, 99.5, 128.0, 128.1, 128.6, 137.4. HRMS (ESI) calcd. for $(M+Na)^+$ $C_{24}H_{40}NaO_5$, 431.2768 found 431.2766.

9-Decenyl 2,3-di-*O*-acetyl-6-*O*-undecyl-4-deoxy-α-D-*lyxo*-pyranoside 55. To a solution of diol **54** (516 mg, 1.26 mmol) in MeOH (10 mL) were added HCOOH (0.2 mL) and 10% wt Pd/C catalyst (150 mg). The mixture was then placed under a hydrogen atmosphere (balloon) and stirred for 20 h. The reaction mixture was purged with nitrogen and filtered through a bed of Celite. The filtrate was concentrated *in vacuo*. The crude residue was taken up into EtOAc (25 mL) and treated with Ac_2O (5 mL, 53.0 mmol) and DMAP (50 mg, 0.40 mmol). The mixture was stirred for 30 min and then was quenched with MeOH (10 mL). The mixture was diluted in EtOAc (25 mL) and washed successively with 1 N HCl and saturated aqueous $NaHCO_3$ solution. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the 1,2,3-tri-*O*-acetylated derivative (501 mg, 90%, 2 steps). To a solution of this material (501 mg, 1.10 mmol) in DCM (25 mL) were added 9-decen-1-ol (0.1 mL, 5.60

mmol) and BF_3•OEt_2 (0.45 mL, 3.3 mmol). The reaction mixture was stirred for 6 h, then quenched with saturated aqueous $NaHCO_3$ solution (25 mL), and extracted with DCM. The organic phase was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave **55** (419 mg, 68%). R_f= 0.38 (15% EtOAc/petroleum ether). ^{1}H NMR δ 0.86 (t, 3H, *J* = 6.8 Hz), 1.26-1.39 (m, 28H), 1.57 (m, 2H), 1.81 (m, 2H), 2.02 (s, 3H), 2.1 (m, 1H), 2.13 (s, 3H), 3.39 (m, 1H), 3.45 (m, 3H), 3.52 (m, 1H), 3.68 (m, 1H), 4.01 (m, 1H), 4.82 (s, 1H), 4.94 (m, 2H), 5.07 (s, 1H), 5.28 (m, 1H), 5.81 (m, 1H). ^{13}C NMR δ 14.3, 21.2 (two lines), 22.9, 26.3 (two lines), 28.6, 29.1, 29.3, 29.5 (two lines), 29.6 (two lines), 29.7, 29.8 (two lines), 32.1, 34.0, 67.2, 67.4, 68.1, 68.4, 98.3, 114.3, 139.4, 170.2, 170.5.

9-Decenyl 2,3-*O*-isopropylidene-6-*O*-undecyl-4-deoxy-α-D-*lyxo*-pyranoside 56. To a solution of **55** (419 mg, 0.80 mmol) in MeOH (10 mL) was added NaOMe (50 mg, 0.92 mmol). The reaction mixture was stirred for 30 min and then adjusted to pH ~ 6 with 1 M HCl in MeOH. The mixture was then concentrated *in vacuo* and the crude residue was taken up in DCM (20 mL). 2,2-DMP (5 mL, 40.80 mmol) and *p*-TsOH (50 mg, 0.26 mmol) were then added, and the reaction mixture was stirred for 1 h and then quenched with Et_3N and concentrated *in vacuo*. FCC of the residue afforded **56** (371 mg, 96%). R_f = 0.42 (10% EtOAc/petroleum ether). ^{1}H NMR δ 0.85 (t, 3H, *J* = 6.8 Hz), 1.23 (m, 26H), 1.31 (s, 3H), 1.35 (m, 2H), 1.49 (s, 3H), 1.52 (m, 4H), 1.88 (m, 1H), 2.01 (m, 1H), 3.42 (m, 4H), 3.50 (m, 1H), 3.70 (m, 1H), 3.82 (m, 1H), 3.92 (d, 1H, *J* = 5.6 Hz), 4.33 (m, 1H), 4.91 (m, 2H), 4.99 (s, 1H), 5.77 (m, 1H). ^{13}C NMR δ 14.3, 22.9, 26.3, 26.4 (two lines), 28.3, 29.1, 29.3, 29.5, 29.6 (two lines), 29.8, 29.9, 30.8, 32.1, 34.0, 65.7, 67.7, 71.0, 71.9, 73.5, 73.8, 97.7, 109.0, 114.3, 139.4. HRMS (ESI) calcd. for $(M+NH_4)^+$ $C_{30}H_{60}NO_5$,

514.4466 found 514.4468.

Galactose CM Precursor 59.

9-Decenyl 6-*O*-trityl-β-D-galactopyranoside 58. To a solution of penta-*O*-acetylated galactopyranose **57** (3.0 g, 7.70 mmol) in DCM (30 mL) were added 9-decen-1-ol (5.5 mL, 31.0 mmol) and $BF_3 \cdot OEt_2$ (3.0 mL, 23.0 mmol). The reaction mixture was stirred at rt for 24 h, then quenched with saturated aqueous $NaHCO_3$ solution, and extracted with DCM. The organic phase was dried (Na_2SO_4), filtered, and concentrated *in vacuo.* FCC of the residue gave compound the 9-decenyl tetra-*O*-acetyl-β-D-galactopyranoside (1.31 g, 35%). R_f = 0.4 (30% EtOAc/petroleum ether). To a sample of the product (2.90 g, 6.0 mmol) in dry MeOH (25 mL) was added NaOMe (96 mg, 1.80 mmol). The reaction mixture was a stirred for 30 min and then neutralized with 1 M HCl in MeOH. The solvent was removed *in vacuo.* The corresponding tetraol was obtained in quantitative yield, R_f = 0.23 (10% MeOH/DCM). To a solution of the tetraol (1.90 g, 6.0 mmol) in DCM (30 mL) were added pyridine (1.5 mL, 18.0 mmol), DMAP (73.0 mg, 0.60 mmol), and trityl chloride (1.8 g, 6.60 mmol). The reaction mixture was stirred for 12 h, after which time the solvents were removed *in vacuo*. FCC of the residue gave **58** (1.83 g, 54%). R_f = 0.25 (80% EtOAc/petroleum ether). ^{1}H NMR δ 1.30 (m, 6H), 1.38 (m, 4H), 1.64 (m, 2H), 2.05 (q, 2H, *J* = 7.1 Hz), 2.30 (d, 1H, *J* = 4.4 Hz), 2.41 (s, 1H), 2.59 (d, 1H, *J* = 5.9 Hz), 3.40 (m, 1H), 3.48 (m, 1H), 3.55 (m, 1H), 3.60 (m, 3H), 3.93 (m, 1H), 4.06 (t, 1H, *J* = 3.4 Hz), 4.24 (d, 1H, *J* = 7.1 Hz), 4.96 (m, 2H), 5.81 (m, 1H), 7.27 (m, 2H), 7.33 (m, 7H), 7.48 (d, 6H, *J* = 7.3 Hz). ^{13}C NMR δ 26.2, 29.1, 29.2, 29.5, 29.6, 29.8, 34.0, 62.7, 69.2, 70.2, 72.6, 73.6, 73.7, 87.2, 103.1, 114.3, 127.4, 128.1, 128.8, 139.4, 143.8.

9-Decenyl 2,4,6-tri-*O*-(triethylsilyl)-3-*O*-undecyl-β-D-galactopyranoside 59. To a solution of trityl ether **58** (668 mg, 1.20 mmol) in MeOH (20 mL) was added Bu_2SnO (308 mg, 1.20 mmol). The mixture was heated at reflux for 3 h in MeOH and then at reflux in toluene for 2 h with the azeotropic removal of water for 3 h. The remaining solvent was removed *in vacuo,* and the residue was taken up in dry DMF (10 mL). CsF (606 mg, 4.0 mmol) and $C_{11}H_{23}Br$ (0.7 mL, 4.1 mmol) were introduced and the mixture was stirred at 80 °C for 15 h and then was cooled to rt, diluted with water, and extracted with EtOAc. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo.* FCC of the residue gave the 9-decenyl 3-*O*-undecyl-6-*O*-trityl-β-D-galactopyranoside (560 mg, 66%). R_f = 0.8 (30% EtOAc/petroleum ether). To a solution the undecyl ether (560 mg, 0.78 mmol) in MeOH was added *p*-TsOH (150 mg, 0.80 mmol). The reaction mixture was strirred for 12 h and then quenched with saturated aqueous $NaHCO_3$ solution (30 mL) and extracted with EtOAc. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the derived triol in quantitative yield, R_f = 0.56 (80% EtOAc/petroleum ether). To a solution of this material (225 mg, 0.35 mmol) in DCM (10 mL) was added imidazole (162 mg, 2.4 mmol) and TESCl (0.3 mL, 2.4 mmol). The reaction mixture was stirred for 2 h, then quenched with water (30 mL) and extracted with ether. The organic phase was dried, (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the **59** (360 mg, 80%). R_f = 0.4 (5% EtOAc/petroleum ether). ^{1}H NMR δ 0.54 (q, 2H, *J* = 8.0 Hz), 0.66 (m, 16H), 0.90 (t, 3H, *J* = 7.0 Hz), 0.98 (m, 27H), 1.29 (m, 26H), 1.62 (m, 4H), 2.05 (m, 2H), 2.40 (s, 1H), 3.19 (dd, 1H, *J* = 3.4, 9.0 Hz), 3.44 (m, 2H), 3.50 (m, 1H), 3.61(m, 2H), 3.83 (m, 2H), 3.93 (dd, 1H, *J* = 6.5, 10.3 Hz), 4.05 (s, 1H), 4.18 (d, 1H, *J* = 7.6 Hz), 4.96

(m, 2H), δ 5.84 (m, 1H). ^{13}C NMR δ 1.2, 4.5, 5.2, 6.6, 6.9, 7.0 two lines, 14.3, 22.9, 26.2, 26.3, 29.1, 29.3, 29.5, 29.6 (two lines), 29.7 (two lines), 29.8 (two lines), 30.2, 32.1, 34.0, 62.1, 65.7, 70.0, 70.3, 72.1, 74.7, 82.7, 103.1, 114.3, 139.4. HRMS (ESI) calcd for $(M+Na)^+$ $C_{39}H_{80}NaO_6Si_2$, 723.5386, found 723.5380.

3,4 Dideoxy mannose CM precursor 67.

(*R*)-1-(Undecyloxy)hex-5-en-2-ol 62. Compound **61** was obtained in three steps from **60.**[80] To a solution of diol **61** (1.94 g, 16.7 mmol) in MeOH (30 mL) was added Bu_2SnO (4.32 g, 17.4 mmol). The reaction mixture was heated to reflux for 3 h and then toluene was added (40 mL). The mixture was heated at reflux for 2 h with a Dean Stark trap for removal of the toluene-water azeotrope. The solvent was then removed *in vacuo* and the residue taken up in dry DMF (30 mL). $C_{11}H_{23}Br$ (10 mL, 56.0 mmol) and CsF (8.4 g, 55.0 mmol) were then introduced and the reaction mixture was heated at 80 °C for 15 h. The mixture was then diluted with water and extracted with EtOAc. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave **62** (4.51 g, 82%). R_f = 0.3 (10% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (t, 3H, J = 6.9 Hz), 1.28 (m, 16H), 1.52 (m, 2H), 1.54 (m, 2H), 2.17 (m, 1H), 2.24 (m, 1H), 2.38 (d, 1H, J = 3.0 Hz), 3.27 (dd, 1H, J = 8.0, 9.4 Hz), 3.47(m, 3H), 3.81 (m, 1H), 5.04 (m, 2H), 5.84 (m, 1H). ^{13}C NMR δ 14.3, 22.9, 26.3, 29.5, 29.6, 29.8 (three lines), 29.9, 32.1, 32.4, 69.9, 71.7, 75.1, 115.0, 138.5. HRMS (ESI) calcd. for $(M+H)^+$ $C_{17}H_{35}O_2$, 271.2632 found 271.2633.

(*R,E*)-Methyl-6-(benzyloxy)-7-undecyloxy-hept-2-enoate 63. To a solution of compound **62** (1.95 g, 7.2 mmol) in DMF (20 mL) were added NaH (60% dispersion in

mineral oil, 576 mg, 14.4 mmol), TBAI (265 mg, 0.7 mmol), and BnBr (2.2 mL, 18.0 mmol). The reaction mixture was stirred for 18 h at rt, then quenched with water and extracted with ether. The organic fraction was washed with water, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the derived benzyl ether (2.6 g, 98%). R_f = 0.22 (5% EtOAc/petroleum ether). This material (2.60 g, 7.2 mmol) was dissolved in DCM (25 mL) and then cooled to −78 °C. Ozone (O_3) was bubbled through the solution for 15 min. The mixture was then purged with N_2 and treated with PPh_3 (3.78 g, 14.0 mmol) for 1 h and then concentrated *in vacuo*. FCC of the residue gave the corresponding aldehyde (2.32 g, 88%). R_f = 0.43 (10% EtOAc/petroleum ether). To a solution of the aldehyde (2.32 g, 6.4 mmol) in CH_3CN (30 mL) was added (methoxycarbonylmethyl)-triphenylphosphonium bromide (3.2 g, 9.6 mmol). The mixture was heated at reflux for 15 h, cooled to rt, and then filtered through a bed of Celite. The filtrate was concentrated *in vacuo*. FCC of the residue gave **63** (1.04 g, 39%). R_f = 0.86 (20% EtOAc/petroleum ether). ^{1}H NMR δ 0.85 (t, 3H, *J* = 6.9 Hz), 1.23-1.31 (m, 18H), 1.66 (m, 2H), 2.22 (m, 1H), 2.30 (m, 1H), 3.41 (m, 3H), 3.51 (m, 2H), 3.69 (s, 3H), 4.51 (d, 1H, *J* = 11.7 Hz), 4.68 (d, 1H, *J* = 11.7 Hz), 5.77 (dt, 1H, *J* = 1.5, 15.7 Hz), 6.93 (dt, 1H, *J* = 6.9, 15.7 Hz), 7.25 (m, 1H), 7.31 (m, 4H). ^{13}C NMR δ 14.3, 22.9, 26.3, 28.4, 29.5, 29.7, 29.8, 29.9, 30.7, 32.1, 51.6, 71.9, 72.3, 73.4, 77.2, 121.2, 127.8, 128.0, 128.5, 138.8, 149.4, 167.2. HRMS (ESI) calcd. for $(M+NH_4)^+$ $C_{26}H_{46}NO_4$, 436.3421 found 436.3421.

(4*S*,5*R*)-5-((*R*)-3-(benzyloxy)-4-(undecyloxy)butyl)-2,2-dimethyl-1,3-dioxolane-4-carboxylic acid 64. A mixture of unsaturated ester **63** (1.04 g, 2.5 mmol), AD mix-α (3 g) and 1:1 tBuOH/water (100 mL) was stirred at 0 °C for 16 h. The reaction was

quenched with Na_2SO_3 (1 g, 8.0 mmol), filtered, and the filtrate was extracted with EtOAc. The organic phase was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was dissolved in DCM (10 mL) and treated with 2,2-DMP (10 mL, 82 mmol) and *p*-TsOH (50 mg, 0.30 mmol) for 30 min. Et_3N was added and the mixture was concentrated *in vacuo*. FCC of the residue gave a homogenous product, (875 mg, 72%) R_f = 0.62 (10% EtOAc/petroleum ether). This material was treated with 2M NaOH in THF (10 mL) for 12 h. The reaction mixture was neutralized with 1 M HCl (aq) and then extracted with EtOAc. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave acid **64** (0.92 g, 71% over 3 steps). R_f = 0.6 (15% MeOH/DCM). ^{1}H NMR δ 0.89 (t, 3H, *J* = 6.9 Hz), 1.27 (m, 14H), 1.44 (s, 3H), 1.48 (s, 3H), 1.59 (m, 2H), 1.74 (m, 2H), 1.89 (m, 2H), 3.46 (m, 2H), 3.49 (d, 1H, *J* = 4.6 Hz), 3.55 (dd, 1H, *J* = 5.8, 10.1 Hz), 3.64 (m, 1H), 4.15 (m, 3H), 4.58 (d, 1H, *J* = 11.7 Hz), 4.71 (d, 1H, *J* = 11.7 Hz), 7.29 (m, 1H), 7.36 (m, 4H). ^{13}C NMR δ 14.3, 22.9, 25.8, 26.3, 27.3, 27.9, 29.3, 29.5, 29.7, 29.8, 29.9, 32.1, 71.9, 72.2, 73.6, 78.5, 79.1, 111.4, 127.7, 128.0, 128.5, 138.9, 172.5.

1,2-di-*O*-acetyl-3,4-dideoxy-6-*O*-undecyl-α-D-*threo*-pyranoside 66. To a solution of acid **64** (923 mg, 1.9 mmol) in dry cyclohexane (60 mL) were added DIB (750 mg, 2.3 mmol) and iodine (539 mg, 2.1 mmol). The reaction mixture was stirred at rt for 12 h and then was quenched with saturated aqueous $Na_2S_2O_3$ solution (30 mL). The product was extracted with EtOAc and the organic layer was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave acetate **65** (733 mg, 71%), which was dissolved in MeOH (10 mL) and treated with Pd/C (10% wt, 100 mg) and HCOOH (0.2 mL) over a hydrogen atmosphere. The mixture was stirred for 24 h and then was filtered

through a bed of Celite. The solvent was removed *in vacuo*, and the crude residue was further treated with TFA/H_2O (1:1, 2 mL). The reaction mixture was stirred for 18 h and then quenched with saturated aqueous $NaHCO_3$ solution (10 mL). The product was extracted with EtOAc and the organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The crude residue was acetylated as for compound **56.** FCC of the residue gave diacetate **66** (510 mg, 88%, 3 steps). R_f = 0.73 (20% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (t, 3H, *J* = 6.9 Hz), 1.27 (m, 14H), 1.57 (m, 4H), 1,75 (m, 1H), 1.95 (m, 1H), 2.04 (m, 2H), 2.13 (s, 6H), 3.48 (dd, 1H, *J* = 5.1, 12.7 Hz), 3.47 (m, 2H), 3.52 (m, 1H), 4.02 (m, 1H), 4.75 (m, 1H), 6.03 (s, 1H). ^{13}C NMR δ 14.3, 21.2, 21.3, 22.3, 22.9, 26.2, 29.5, 29.6, 29.8, 32.1, 66.7, 70.3, 72.0, 73.6, 91.2, 169.1, 170.7.

9-Decenyl 3,4-dideoxy-6-*O*-undecyl-α-D-*threo*-pyranoside 67. To a solution of di-*O*-acetate **66** (500 mg, 1.3 mmol) in DCM (10 mL) were added 9-decen-1-ol (1 mL, 5.7 mmol) and $BF_3{\cdot}OEt_2$ (0.1 mL, 0.8 mmol). The reaction mixture was stirred for 3 h and then quenched with saturated aqueous $NaHCO_3$ solution and extracted with EtOAc. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The crude residue was treated taken up into MeOH (5 mL) and treated with NaOMe (50 mg, 0.90 mmol). The reaction mixture was stirred for 30 min and then was neutralized with 1 M HCl in MeOH. The solvent was concentrated *in vacuo*. FCC of the residue gave CM precursor **67** (334 mg, 58% over 2 steps). R_f = 0.4 (20% EtOAc/petroleum ether). ^{1}H NMR δ 0.88 (t, 3H, *J* = 6.9 Hz), 1.26 (m, 30H), 1.58 (m, 2H), 1.67 (m, 1H), 1.75 (m, 1H), 1.99 (m, 2H), 2.04 (m, 2H), 3.41 (dd, 1H, *J* = 6.5, 12.5 Hz), 3.44 (m, 4H), 3.62 (m, 1H), 3.63 (m, 1H), 3.95 (m, 1H), 4.68 (s, 1H), 4.95 (m, 2H), 5.81 (m, 1H). ^{13}C NMR δ 14.3, 21.8, 22.9, 25.3, 26.3, 26.4, 29.1, 29.3, 29.5, 29.6 (two lines), 29.7, 29.8, 29.9, 32.1 34.0,

66.1, 67.7, 67.9, 71.9, 74.0, 99.8, 114.3, 139.4. HRMS (ESI) calcd. for $(M+NH_4)^+$ $C_{37}H_{56}NO_4$, 458.4204 found 458.4204.

Disaccharide CM precursor 73.

6-*O*-Triisopropylsilyl-3,4-*O*-isopropylidene-2-*O*-undecyl-β-D-galactopyranosyl-(1→1)-2,3:4,6-di-*O*-isopropylidene-α-D-mannopyranoside 71. Disaccharide **24β** (1.30 g, 2.2 mmol) was converted to the Gal-tetraol as described in Chapter 2 (p. 28) for compound **26**. The tetraol (904 mg, 2.1 mmol) was taken up into DMF (15 mL) at rt, to which were added imidazole (160 mg, 2.4 mmol) and TIPSCl (0.5 mL, 2.4 mmol). The reaction mixture was stirred for 2 h and then was quenched with water and extracted with EtOAc. The organic fractions were combined, washed with water, dried (Na_2SO_4), filtered and then concentrated *in vacuo*. FCC of the residue gave the Gal 6-TIPS ether triol (1.13 g, 91%), R_f = 0.25 (75% EtOAc/petroleum ether). To a solution of triol (1.13 g, 2.0 mmol) in dry CH_2Cl_2 (10 mL) and 2,2-DMP (10 mL, 82.0 mmol) was added *p*-TsOH (200 mg, 1.0 mmol). The reaction mixture was stirred for 1 h and then was quenched with aqueous saturated $NaHCO_3$ solution and extracted with ether. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. To a solution of the crude product in dry THF (10 mL) were added NaH (60% dispersion in mineral oil, 117 mg, 2.9 mmol) and 1-bromoundecane (1 mL, 5.4 mmol). The reaction mixture was stirred overnight and then was quenched with water and extracted with ether. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded undecyl ether **71** (140 mg, 67% brsm, 2 steps), R_f = 0.75 (20% EtOAc/petroleum ether). ^{1}H NMR δ 0.88 (m, 3H), δ 1.08 (m, 21H), δ 1.27 (m, 16H), 1.35 (s, 3H), δ 1.37 (s, 3H), δ 1.43 (s, 3H), δ 1.53 (s, 3H), δ 1.55 (s, 3H), δ 1.59 (s, 3H), δ 3.30

(t, 1H, *J* = 7.0 Hz), δ 3.65 (m, 1H), δ 3.76 (m, 3H), δ 3.84 (m, 4H), δ 4.02 (t, 1H, *J* = 8.7 Hz), δ 4.10 (m, 1H), δ 4.20 (m, 2H), δ 4.28 (dd, 1H, *J* = 1.8, 5.5 Hz), δ 4.43 (d, 1H, *J* = 8.1 Hz), δ 5.34 (s, 1H). ^{13}C NMR δ 12.1, 14.3, 18.1 (two lines), 19.0, 22.9, 26.4 (three lines), 28.4, 28.5, 29.3, 29.5, 29.7, 29.8, 30.1, 32.1, 62.0, 62.1, 62.3, 72.6 (two lines), 73.8, 75.2, 75.9, 79.3, 81.2, 99.4, 99.7, 101.9, 109.7, 109.8. HRMS (ESI) calcd for $(M+NH_4)^+$ $C_{41}H_{80}NO_{11}Si$, 790.5495 found 790.5488.

6-*O*-Triisopropylsilyl-3,4-*O*-isopropylidene-2-*O*-undecyl-β-D-galactopyranosyl-(1→1)-2,3-*O*-isopropylidene-4-*O*-methoxymethyl-α-D-mannopyranoside 72.

Compound **71** (140 mg, 0.18 mmol) was converted to the Man 4,6-diol as previously described for compound **26** (Chapter 2, p. 28). To a solution of the diol (131 mg, 0.2 mmol) in dry DCM (2 mL) were added pyridine (0.15 mL, 1.8 mmol) and pivaloyl chloride (20 μL, 0.20 mmol) at rt. The reaction mixture was stirred for 3 h, diluted in EtOAc and washed with 1 N HCl and saturated aqueous $NaHCO_3$ solution. The organic phase was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded the Man 6-*O*-pivaloate ester (140 mg, 95%), R_f = 0.17 (10% EtOAc/petroleum ether). To a solution of 6-ester (140 mg, 0.17 mmol) in dry CH_2Cl_2 (3 mL) were added DIPEA (0.2 mL, 1.15 mmol) and MOMCl (50 μL, 0.65 mmol). The reaction mixture was stirred for 2 h and then was quenched with saturated aqueous NH_4Cl solution, and extracted with ether. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue afforded the 4-MOM ether (103 mg, 70%), R_f = 0.43 (10% EtOAc/petroleum ether). To a solution of 4-MOM ether (103 mg, 0.12 mmol) in dry CH_2Cl_2 (3 mL) was added DIBAL-H (1.0 M in hexanes, 0.48 mL, 0.5 mmol) at -78 °C. The reaction mixture was stirred at -78 °C for 1 h and then was

quenched with saturated aqueous Rochelle's salt solution and extracted with ether. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave primary alcohol **72** (88 mg, 78%). R_f = 0.37 (10% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (m, 3H), δ 1.08 (m, 21H), 1.27 (m, 16H), 1.35 (s, 3H), 1.37 (s, 3H), 1.54 (s, 3H), 1.58 (s, 3H), 2.45 (t, 1H, *J* = 6.8 Hz), 3.29 (t, 1H, *J* = 7.0 Hz), 3.43 (s, 3H), 3.66 (m, 1H), 3.75 (m, 4H), 3.86 (m, 4H), 3.96 (m, 2H), 4.11 (t, 1H, *J* = 5.6 Hz), 4.15 (d, 1H, *J* = 5.6 Hz), 4.25 (m, 2H), 4.51 (d, 1H, *J* = 8.0 Hz), 4.69 (d, 1H, *J* = 6.4 Hz), 4.95 (d, 1H, *J* = 6.5 Hz), 5.31 (s, 1H). ^{13}C NMR δ 12.1, 14.3, 18.1 (two lines), 22.9, 26.3, 26.4, 26.6, 28.2, 28.3, 29.5, 29.7, 29.8, 30.3, 32.1, 56.2, 61.8, 62.3, 69.9, 72.6, 73.0, 73.6, 73.8, 75.9, 76.8, 78.5, 79.3, 81.0, 97.1, 99.1, 101.8, 109.7, 109.9.

6-*O*-Triisopropylsilyl-3,4-*O*-isopropylidene-2-*O*-undecyl-β-D-galactopyranosyl-(1→1)-2,3-*O*-isopropylidene-4-*O*-methoxymethyl-6-*O*-pentenyl-α-D-mannopyranoside 73. To a solution of alcohol **72** (60.6 mg, 0.08 mmol) in dry THF (2 mL) were added NaH (60% dispersion in mineral oil, 31 mg, 0.80 mmol), TBAI (6 mg, 0.16 mmol), and C_5H_9Br (0.2 mL, 0.80 mmol). A similar work-up protocol was followed as for **71**. This afforded compound **73** (51 mg, 77%). R_f = 0.76 (10% EtOAc/petroleum ether). ^{1}H NMR δ 0.89 (t, 3H, *J* = 7.0 Hz), 1.08 (m, 21H), 1.27 (m, 14H), 1.35 (s, 3H), 1.37 (s, 3H), 1.55 (s, 3H), 1.56 (s, 3H), 1.73 (m, 2H), 2.12 (m, 2H), 3.28 (t, 1H, *J* = 7.2 Hz), 3.40 (m, 1H), 3.43 (s, 3H), 3.56 (m, 2H), 3.65 (m, 2H), 3.75 (m, 1H), 3.84 (m, 3H), 3.96 (m, 2H), 4.10 (m, 1H), 4.14 (d, 1H, *J* = 6.0 Hz), 4.23 (dd, 1H, *J* = 5.5, 7.1 Hz), 4.28 (dd, 1H, *J* = 1.4, 5.3 Hz), 4.50 (d, 1H, *J* = 8.5 Hz), 4.73 (d, 1H, *J* = 6.5 Hz), 4.85 (d, 1H, *J* = 6.5 Hz), 5.01 (m, 2H), 5.34 (s, 1H), 5.82 (m, 1H). ^{13}C NMR δ 12.0, 14.3, 18.1 (two lines), 22.9, 26.3, 26.4, 26.7, 28.1, 20.4, 28.9, 29.5, 29.7, 29.8, 30.2, 30.5, 32.1, 56.0, 62.0,

68.9, 69.1, 71.3, 72.6 (two lines), 72.8, 73.6, 75.8, 78.5,79.4, 81.2, 96.4, 98.9, 101.3, 109.6, 109.7, 114.8, 138.5.

CM, reduction, and final deprotection steps

Mannose AA analog 34. A solution of **47** (155 mg, 0.30 mmol) and butenolide **81** (20 mg, 0.10 mmol) in DCM (6 mL) was degassed using N_2 for 30 min and then Grubbs II (9 mg, 0.010 mmol) was added. The reaction mixture was stirred for 18 h and then was concentrated *in vacuo*. FCC of the residue gave the CM product. To a refluxing solution of the CM product (44 mg, 0.065 mmol) in DME (12 mL) was added a NaOAc (426 mg, 5.2mmol) solution in water (5 mL) over a 4 h period. The mixture was then cooled, diluted with EtOAc, and washed with 1 M HCl and saturated aqueous $NaHCO_3$ solution. The organic fraction was dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The crude residue was taken up into DCM (5 mL) and treated with 3% AcCl in MeOH (0.5 mL). The reaction mixture was stirred for 1 h and then was quenched with solid $NaHCO_3$. The mixture was filtered, and concentrated *in vacuo*. FCC of the residue afforded mannose analog **34** (15 mg, 22% over 3 steps). R_f = 0.3 (80% EtOAc/petroluem ether). ^{1}H NMR δ 0.88 (m, 3H), 1.26 (m, 34H), 1.44 (d, 3H, *J* = 6.8 Hz), 1.48 (m, 2H), 1.57 (m, 8H), 2.40 (m, 1H), 2.53 (m, 1H), 3.41 (m, 1H), 3.51 (m, 2H), 3.72 (m, 6H), 3.83 (m, 2H), 3.91 (s, 1H), 4.82 (s, 1H), 5.06 (m, 1H), 7.19 (s, 1H). ^{13}C NMR δ 14.3, 19.3, 22.9, 25.7, 26.2, 29.4 (two lines), 29.5, 29.6 (two lines), 29.7 (two lines), 29.8 (two lines), 29.9, 32.1, 33.5, 37.6, 68.1, 69.3, 70.2, 70.7, 70.8, 71.9, 72.4, 78.2, 99.7, 131.4, 152.0, 174.8. HRMS (ESI) calcd for $(M+HCOO)^-$ $C_{37}H_{67}O_{11}$, 687.4689 found 687.4678.

Reduced mannose analog 39. Compound **39** (6 mg, 0.01 mmol) was reduced using standard hydrogenolysis with Pd/C catalyst. The reduced product was obtained in quantitative yield. R_f = 0.3 (80% EtOAc/petroluem ether). ^{1}H NMR δ 0.85 (t, 3H, *J* = 6.9 Hz), 1.23 (m, 40H), 1.41 (d, 3H, *J* = 6.2 Hz), 1.57 (m, 12H), 1.91 (m, 1H), 2.1 (m, 1H), 2.5 (m, 4H), 2.90 (m, 1H), 3.37 (m, 1H), 3.49 (t, 1H, *J* = 9.4 Hz), 3.56 (m, 1H), 3.60 (m, 3H), 3.70 (m, 1H), 3.75 (m, 1H), 3.83 (m, 1H), 3.88 (m, 1H), 3.91 (bs, 1H), [4.37 (m, minor) 4.51 (m, major) 1H], 4.79 (s, 1H). ^{13}C NMR δ 14.3, 21.0, 22.9, 24.4, 25.4, 25.7, 26.2, 26.3, 29.4 (two lines), 29.5 (two lines), 29.6 (two lines), 29.7 (two lines), 29.8 (two lines), 29.9, 30.6, 32.1, 35.6, 36.2, 37.9, 38.2, 38.4, 39.6, 39.9, 40.0, 62.3, 66.8, 68.0, 70.6, 71.3, 71.5, 71.8, 73.5, 76.1, 76.8, 79.9. 99.6, 180.3, 180.4. HRMS (ESI) calcd for $(M+Na)^+$ $C_{35}H_{66}NaO_9$, 653.4599 found 653.4601.

4-Deoxy mannose AA analog 35. Compound **55** (358 mg, 0.72 mmol) and **81** (47 mg, 0.24 mmol) was subjected to the standard CM procedure to give the hetero CM product (31 mg, 19.5 % yield). The standard diimide procedure on this material (31 mg, 0.046 mmol) gave the reduced product (25 mg, 83%). The standard deprotection conditions on this material (25 mg, 0.0037 mmol), from the previous step afforded **35** (11 mg, 46%). R*f* = 0.13 (60% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (m, 3H), 1.27 (m, 33H), 1.41 (d, 3H, *J* = 6.80 Hz), 1.52 (m, 10H), 1.75 (m, 1H), 2.38 (m, 1H), 2.51 (m, 1H), 3.41 (m, 6H), 3.63 (m, 1H), 3.71 (s, 1H), 3.86 (m, 2H), 3.98 (m, 1H), 4.85 (s, 1H), 5.08 (m, 1H), 7.11 (s, 1H). ^{13}C NMR δ 14.3, 19.3, 22.9, 25.8, 26.3 (two lines), 29.4, 29.5 (two lines), 29.6 (two lines), 29.7 (two lines), 29.8, 29.9 31.5, 32.1, 33.5, 37.6, 65.8, 67.2, 67.9, 69.5, 70.2, 72.0, 73.5, 78.2, 100.3, 131.4, 152.0, 174.8. HRMS (ESI) calcd for $(M+NH_4)^+$ $C_{35}H_{68}NO_8$, 630.4939 found 630.4928.

2-Deoxymannose AA analog 36. Compound **52** (426 mg, 0.62 mmol) and butenolide **81** (41 mg, 0.21 mmol) was subjected to the standard CM procedure to give the hetero CM product (43 mg, 24% yield). The standard diimide procedure on this material (43 mg, 0.05 mmol) gave **36** (12.3 mg, 40%). R_f = 0.0.26 (60% EtOAc/petroleum ether). ^{1}H NMR δ 0.85 (m, 3H), 1.28 (m, 34H), 1.41 (d, 3H, *J* = 6.8 Hz), 1.45 (m, 2H), 1.50 (m, 8H), 1.64 (m, 1H), 2.08 (dd, 1H, *J* = 5.1, 12.8 Hz), 2.37 (dd, 1H, *J* = 8.4, 15.2 Hz), 2.49 (d, 1H, *J* = 15.2 Hz), 3.15 (bs, 1H), 3.31 (m, 1H), 3.47 (m, 3H), 3.58 (m, 2H), 3.67 (m, 2H), 3.82 (m, 1H), 3.96 (m, 1H), 4.85 (d, 1H, *J* = 3.2 Hz), 5.04 (q, 1H, *J* = 6.8 Hz), 7.16 (s, 1H). ^{13}C NMR δ 14.3, 19.3, 22.9, 25.7, 26.2, 26.4, 29.5 (two lines), 29.6, 29.7 (three lines), 29.8, 29.9, 32.1, 37.1, 37.6, 67.7, 69.0, 69.2, 70.2, 72.2, 72.3, 75.5, 78.2, 97.6, 131.4, 152.0, 174.8. HRMS (ESI) calcd for $(M+Na)^+$ $C_{36}H_{66}NaO_8$, 649.4650 found 649.4648.

2,3-Dideoxymannose AA analog 37. Compound **50** (101 mg, 0.20 mmol) and butenolide **81** (15 mg, 0.080 mmol) was subjected to the standard CM procedure to give the hetero CM product (31 mg, 67% yield). The standard diimide procedure on a portion of this material (18 mg, 0.03 mmol) gave the final product **37** (10 mg, 59% yield), R_f = 0.14 (35% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (t, 3H, *J* = 6.8 Hz), 1.28 (m, 32H), 1.46 (d, 3H, *J* = 6.8 Hz), 1.49 (m, 3H), 1.62 (m, 7H), 1.70 (m, 3H), 1.86 (m, 1H), 2.02 (m, 2H), 2.45 (m, 3H), 2.57 (bd, 1H, *J* = 15.1 Hz), 3.27 (m, 1H), 3.36 (m, 3H), 3.64 (m, 3H), 3.74 (dd, 1H, *J* = 4.4, 11.3 Hz), 3.87 (m, 1H), 4.78 (s, 1H), 5.07 (q, 1H, *J* = 6.8 Hz), 7.21 (s, 1H). ^{13}C NMR δ 14.1, 19.3, 22.5, 22.9, 24.0, 25.8, 26.3, 26.4, 29.3, 29.5, 29.7 (two lines), 29.8 (two lines), 29.9, 30.2, 31.1, 32.1 33.5, 34.3, 37.6, 63.6, 67.3, 69.0, 70.2, 71.6, 75.0, 78.1, 63.6, 67.3, 69.0, 70.2, 71.6, 75.0, 78.1, 96.4, 131.4, 151.9, 174.8. HRMS (ESI) calcd for $(M+HCOO)^-$ $C_{37}H_{67}O_9$, 655.4791 found 655.4773.

3,4-Dideoxymannose AA analog 38. Compound **67** (27 mg, 0.061 mol) and butenolide **75** (39 mg, 0.20 mmol) was subjected to the standard CM procedure to give the hetero CM product (14 mg, 38% yield). The standard diimide procedure on this material (14 mg, 0.023 mmol) gave the final product **38** (12 mg, 84%), R_f = 0.15 (35% EtOAc/petroleum ether). ^{1}H NMR δ 0.85 (t, 3H, *J* = 6.9 Hz), 1.23 (m, 40H), 1.41 (d, 3H, *J* = 6.8 Hz), 1.46 (m, 3H), 1.57 (m 9H), 1.64 (m, 1H), 1.73 (m, 1H), 1.97 (m, 2H), 2.23 (bs, 1H), 2.38 (m, 1H), 2.52 (m, 1H), 3.36 (dd, 1H, *J* = 4.3, 10.4 Hz), 3.43 (m, 5H), 3.59 (bs, 1H), 3.69 (m, 1H), 3.8 (m, 1H), 3.91 (m, 1H), 4.61 (s, 1H), 5.04 (m, 1H). ^{13}C NMR δ 14.3, 19.3, 21.8, 22.9, 25.2, 25.8, 26.3, 26.4, 29.5, 29.6 (two lines), 29.7 (two lines), 29.8, 29.9, 32.1, 33.5, 37.6, 66.1, 67.7, 67.9, 70.2, 71.9, 73.9, 78.2, 99.8, 131.4, 152.0, 174.8. HRMS (ESI) calcd for $(M+Na)^+$ $C_{35}H_{64}NaO_7$, 619.4544 found 619.4543.

Mannose analog 41 (2 OH groups on alkyl spacer). Compound **70** (35 mg, 0.060 mmol) and butenolide **81** (37 mg, 0.2 mmol) were subjected to the standard CM procedure to give the hetero CM product (14 mg, 31% yield). The standard diimide procedure on a portion of this material (13 mg, 0.017 mmol) gave the reduced product (11 mg, 83%). The standard deprotection conditions on this material (11 mg, 0.015 mmol), from the previous step afforded **41** (6.8 mg, 62%), R_f = 0.45 (10% MeOH/DCM). ^{1}H NMR δ 0.90 (t, 3H, *J* = 6.8 Hz), 1.28 (m, 18H), 1.46 (d, 3H, *J* = 6.8 Hz), 1.51 (m, 3H), 1.59 (m, 4H), 1.67 (m, 4H), 2.43 (m, 2H), 2.56 (m, 2H), 3.46 (m, 1H), 3.53 (m, 1H), 3.63 (m, 3H), 3.77 (m, 3H), 3.88 (m, 3H), 3.92 (s, 1H), 4.85 (s, 1H), 5.09 (q, 1H, *J* = 6.8 Hz), 7.21 (s, 1H). ^{13}C NMR δ 14.3, 19.2/19.3 (1:1), 22.9, 25.5 (two lines), 25.6 (three lines), 25.7, 25.8, 26.2, 29.4 (two lines), 29.5, 29.7 (two lines), 29.8, 29.9, 30.6, 32.1, 33.6, 33.7 (two lines), 34.2 (three lines), 34.4, 37.3 (three lines), 62.2/62.3 (1:1), 68.0/68.0 (1:1),

70.0, 71.2, 71.3, 71.5, 71.7, 71.8 (three lines), 73.5, 73.6, 75.9, 78.2, 99.6/99.7 (1:1), 131.3/131.3 (1:1), 152.2/152.2 (1:1), 174 .9/174.9 (1:1). HRMS (ESI) calcd for $(M+Na)^+$ $C_{33}H_{60}NaO_{10}$, 640.4113 found 640.4105.

β-D-Galactose AA analog 42. Compound **59** (158 mg, 0.20 mmol) and butenolide **81** (14 mg, 0.060 mmol) were subjected to the standard CM procedure to give the hetero CM product (11 mg, 15% yield). The standard diimide procedure on this material (11 mg, 0.011 mmol) gave the reduced product in quantitative yield. The standard deprotection conditions on a portion of the material (11 mg, 0.011 mmol), from the previous step afforded **42** (6 mg, 86%), R_f = 0.15 (60% EtOAc/petroleum ether). ^{1}H NMR δ 0.89 (t, 3H, *J* = 6.4 Hz), 1.31 (m, 30H), 1.44 (d, 3H, *J* = 6.7 Hz), 1.47 (m, 3H), 1.62 (m, 8H), 2.40 (m, 2H), 2.53 (m, 2H), 3.32 (m, 1H), 3.55 (m, 2H), 3.61 (m, 1H), 3.68 (m, 3H), 3.86 (m, 2H), 3.93 (m, 1H), 4.01 (m, 1H), 4.05 (s, 1H), 4.27 (d, 1H, *J* = 7.8 Hz), 5.09 (m, 1H), 7.10 (s, 1H). ^{13}C NMR δ 14.3, 19.3, 22.9, 25.7 (two lines), 26.0, 26.1, 26.2, 29.4, 29.5 (three lines), 29.6 (three lines), 29.7 (three lines), 29.8 (two lines), 29.9, 30.1, 32.1, 33.5, 37.6, 62.8, 66.9, 70.2, 70.3, 70.5, 71.0, 74.5, 78.2, 81.1, 103.2, 131.4, 152.0, 174.8. HRMS (ESI) calcd for $(M+Na)^+$ $C_{35}H_{64}NaO_9$, 665.4599 found 665.4581.

Disaccharide AA analog 43. Compound **73** (51 mg, 0.06 mmol), and butenolide **81** (35 mg, 0.18 mmol) were subjected to the standard CM procedure to give the hetero CM product (18.6 mg, 31% yield). The standard diimide procedure on this material (18.6 mg, 0.018 mmol) gave the reduced product in quantitative yield. The standard deprotection conditions on this material (18.3 mg, 0.0018), from the previous step afforded **43** (7.0 mg, 59%), R_f = 0.46 (10% MeOH/DCM). ^{1}H NMR δ 0.90 (m, 3H, *J* = 6.8 Hz), 1.28 (m,

33H), 1.47 (d, 3H, J = 6.8 Hz), 1.59 (m, 6H), 2.44 (m, 1H), 2.45 (m, 1H), 3.38 (t, 1H, J = 3.0 Hz), 3.53 (m, 2H), 3.62 (m, 6H), 3.79 (m, 3H), 3.99 (s, 2H), 4.17 (bs, 1H), 4.55 (d, 1H, J = 7.7 Hz), 5.09 (m, 1H), 5.16 (s, 1H), 7.23 (s, 1H). ^{13}C NMR δ 14.3, 19.2, 22.9, 25.2, 25.5, 26.4 26.4, 28.9, 29.1, 29.5, 29.8 (two lines), 29.9, 30.5, 32.1, 33.5, 37.1, 37.3, 37.4, 62.7, 69.4, 70.2, 70.6, 71.6 (two lines), 71.7, 72.0, 73.5, 73.7, 75.5, 78.4, 79.8, 101.6, 103.7, 131.3, 152.4, 175.2. HRMS (ESI) calcd for $(M+Na)^+$ $C_{37}H_{66}NaO_{14}$, 757.4345 found 757. 4337.

Dialkyl control compounds 40 and 44.

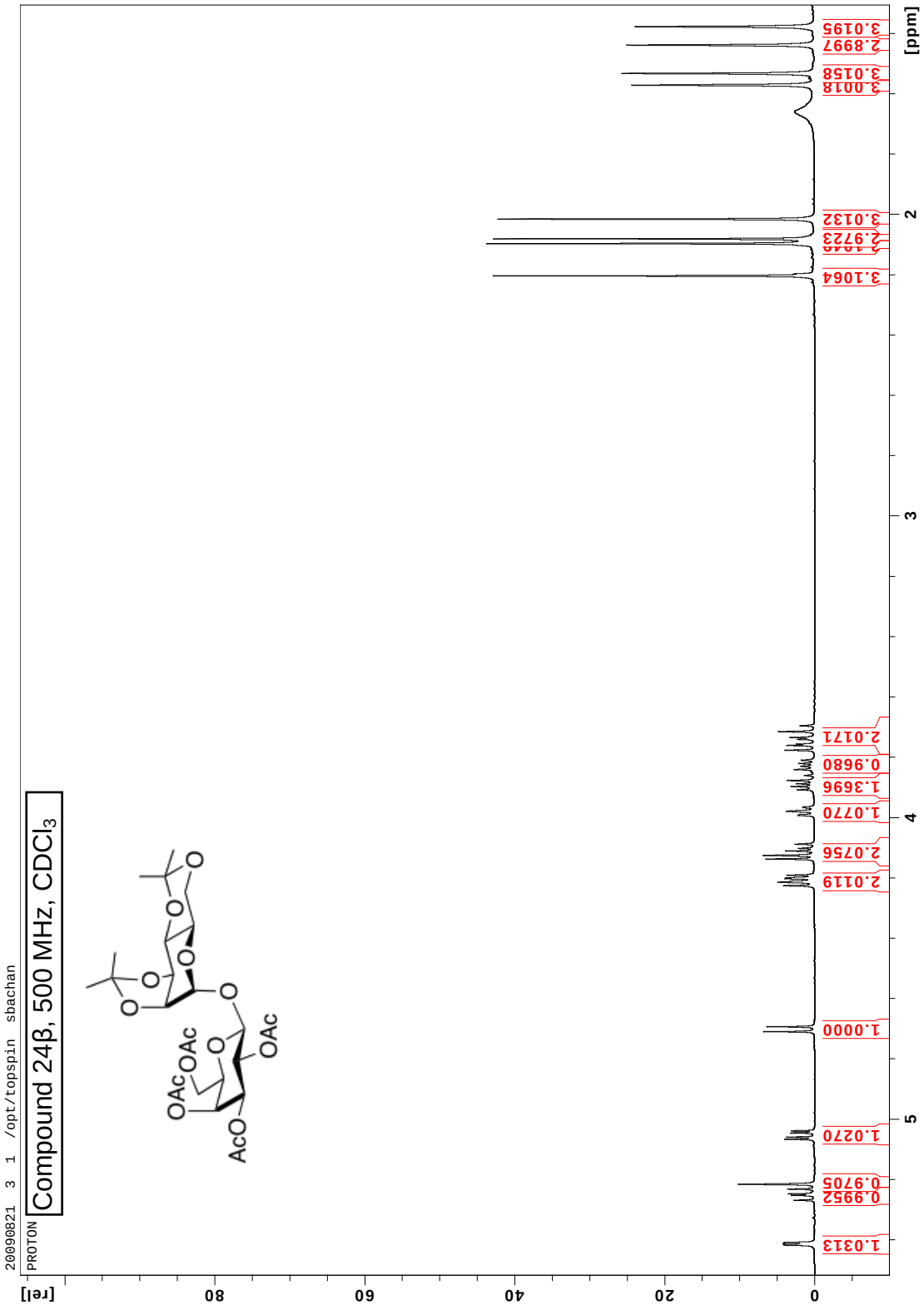
Hexadecyl-2,3-*O*-isopropylidene-α-D-mannopyranoside 74. To a solution of mannose penta-*O*-acetate **45** (1.85 g, 4.7 mmol) in DCM (30 mL) were added $BF_3{\cdot}OEt_2$ (1.8 mL, 14.2 mmol) and 1-hexadecanol (3.4 g, 14.2 mmol). The reaction mixture was stirred at reflux for 20 h, quenched with saturated aqueous $NaHCO_3$ solution and extracted with DCM. The organic fractions were dried (Na_2SO_4), filtered, and concentrated in vacuo. FCC of the residue gave the hexadecyl glycoside (1.6 g, 85%), R_f = 0.43 (10% EtOAc/petroleum ether). The hexadecyl glycoside (1.6 g, 2.8 mmol) was converted to 4,6-diol **74** using similar protection and deprotection protocols as for compound **46**. For compound **74**: (500 mg, 53% over 3 steps). R_f = 0.16 (30% EtOAc/petroleum ether). ^{1}H NMR δ 0.90 (t, 3H, J = 6.9 Hz), 1.28 (m, 28H), 1.38 (s, 3H), 1.55 (s, 3H), 1.60 (m, 2H), 2.14 (t, 1H, J = 6.3 Hz), 2.71 (d, 1H, J = 4.8 Hz), 3.44 (m, 1H), 3.67 (m, 1H), 3.76 (m, 2H), 3.86 (m, 2H), 4.17 (m, 1H), 4.20 (m, 1H), 5.01 (s, 1H). ^{13}C NMR δ 14.3, 22.9, 26.2, 26.3, 28.0, 29.5, 29.6, 29.7, 29.8 (two lines), 29.9 (two lines), 32.1, 62.8, 68.2, 69.7, 70.1, 75.7, 78.2, 97.4, 109.8.

Hexadecyl-6-*O*-undecyl-α-D-mannopyranoside 40. Compound **74** (500 mg, 1.13 mmol) was converted to the 6-undecyl ether using a similar protocol as for compound **46**. The 6-undecyl ether (190 mg, 0.31 mmol) was taken up into 1:1 MeOH/DCM (10 mL) and treated with *p*-TsOH (150 mg, 0.78 mmol). The reaction mixture was stirred for 2 h, quenched with saturated aqueous $NaHCO_3$ solution, and extracted with DCM. The organic fractions were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. FCC of the residue gave the dialkyl mannose compound **40** (171 mg, 40% over 2 steps brsm). R_f = 0.4 (15% EtOAc/petroleum ether). ^{1}H NMR δ 0.85 (t, 6H, *J* = 6.9 Hz), 1.23 (m, 46H), 1.54 (m, 6H), 2.40 (bs, 1H), 2.47 (bs, 1H), 3.35 (m, 1H), 3.52(t, 1H, *J* = 9.4 Hz), 3.55 (m, 1H), 3.60 (m, 2H), 3.68 (m, 1H), 3.75 (dd, 1H, *J* = 3.8, 11.7 Hz), 3.83 (dd, 1H, *J* = 2.4, 11.7 Hz), 3.90 (m, 2H), 4.79 (s, 1H). ^{13}C NMR δ 14.3, 22.9, 26.3 (two lines), 29.5 (two lines), 29.6 (two lines), 29.7 (two lines), 29.8 (three lines), 29.9 (two lines), 30.6, 32.1 (two lines), 62.3, 68.1, 71.3, 71.5, 71.8, 73.5, 76.0, 99.6. HRMS (ESI) calcd for $(M+NH_4)^+$ $C_{33}H_{70}NO_6$, 576.5198 found 576.5198.

2-*O*-Undecyl-β-D-galactopyranosyl-(1→1)-6-*O*-undecyl-α-D-mannopyranoside 44. Compound **73** (28 mg, 0.030 mmol) was converted to the 6-undecyl ether as for compound **71**. Data for the 6-undecyl ether: (23 mg, 67%). R_f = 0.84 (10% EtOAc/petroleum ether). To a solution of dialkyl compound (23 mg, 0.030 mmol) in dry CH_2Cl_2 (1 mL) was added a 0.1 M solution of HCl in MeOH (2 mL). The reaction mixture was stirred for 18 h at rt and then was quenched with solid $NaHCO_3$. The mixture was filtered and concentrated *in vacuo*. FCC of the crude product gave compound **44** (9.8 mg, 61%). R_f = 0.41 (10% MeOH/EtOAc). ^{1}H NMR (CD_3OD) δ 0.90 (t, 6H, *J* = 6.5 Hz), 1.31 (m, 32H), 1.61 (m, 4H), 3.63 (m, 4H), 3.75 (m, 5H), 3.81 (m,

1H), 3.88 (m, 1H), 4.02 (m, 1H), 4.47 (d, 1H, J = 7.8 Hz), 5.03 (s, 1H). ^{13}C NMR (CD_3OD) δ 14.6, 23.9, 27.3, 27.5, 30.5, 30.6, 30.8, 30.9, 31.0, 31.6, 33.2, 2.7, 67.0, 70.4, 71.8, 72.1, 72.5, 72.9, 74.2, 74.6, 74.8, 77.2, 78.3, 78.5, 78.8, 81.8.103.5, 104.8. HRMS (ESI) calcd for $(M+Na)^+$ $C_{34}H_{66}NaO_{11}$, 673.4497 found 673.4496.

APPENDIX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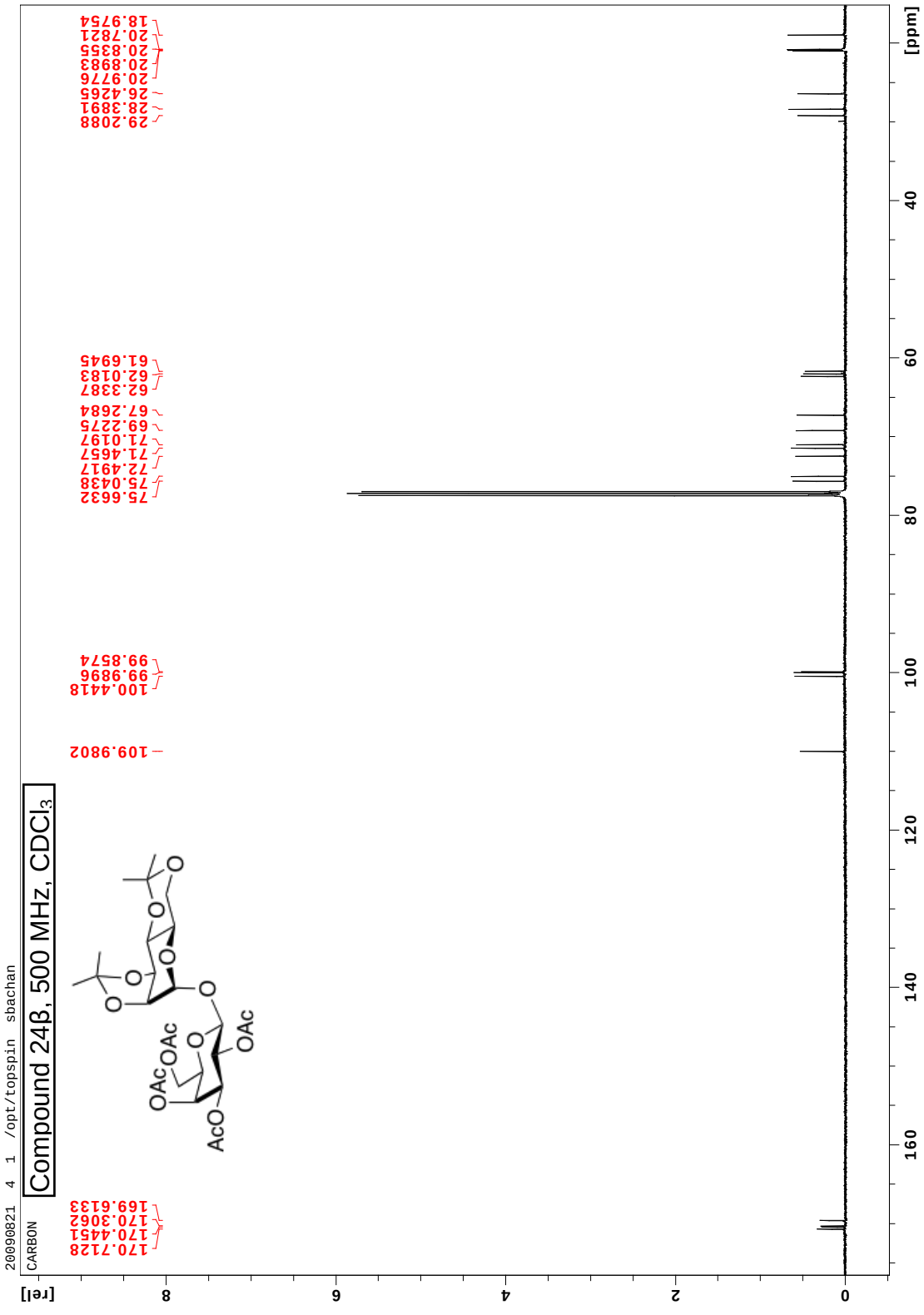
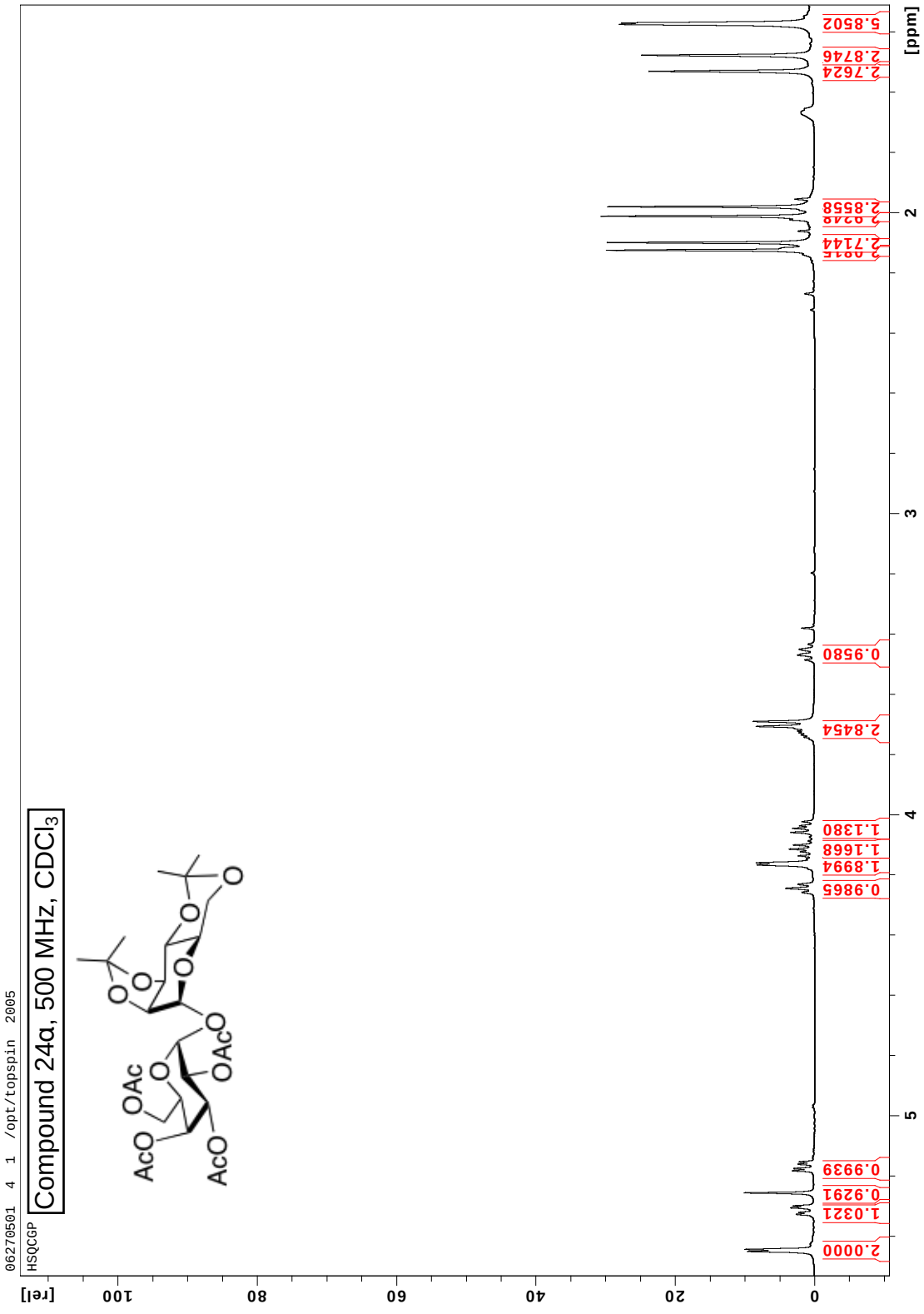
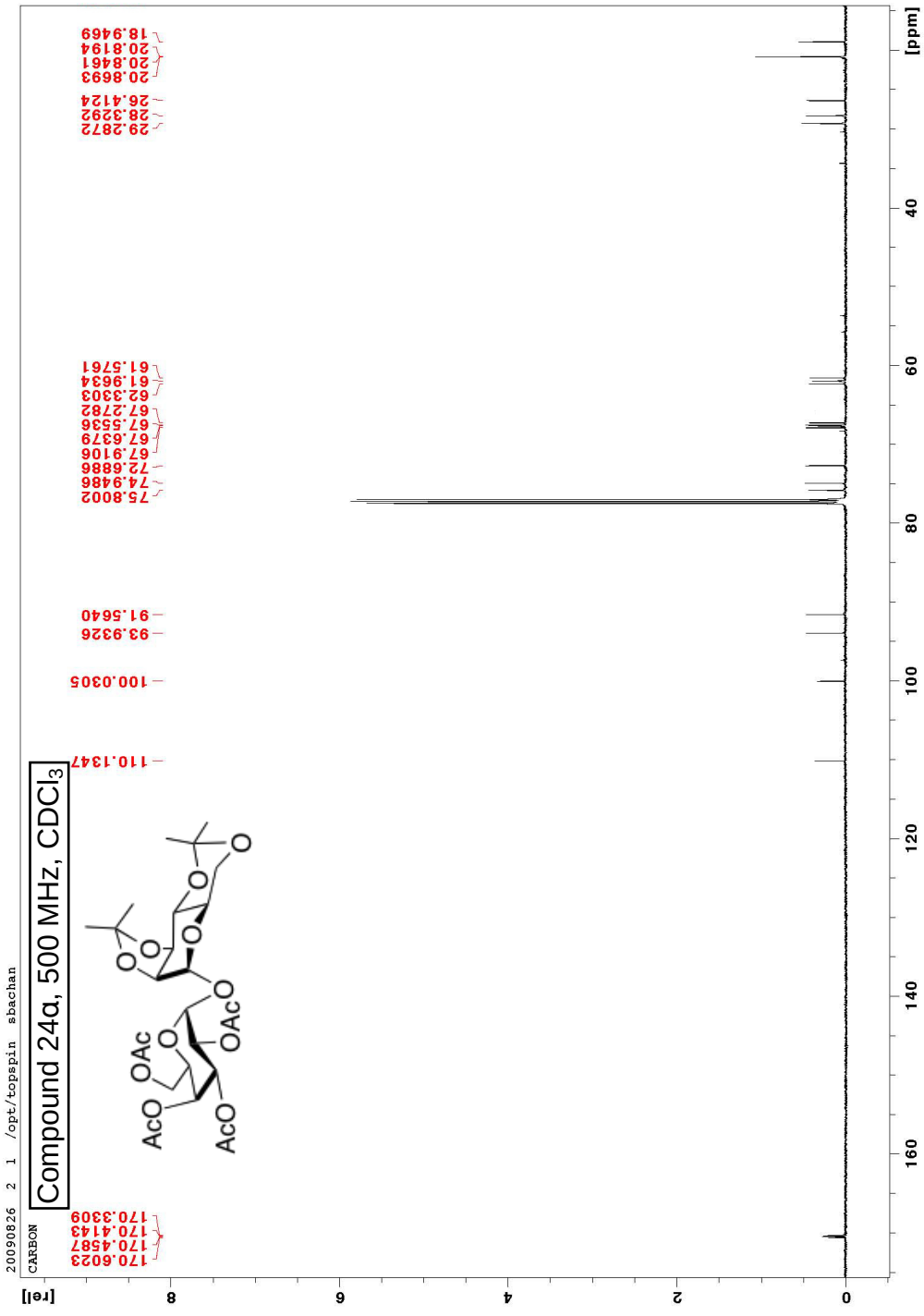
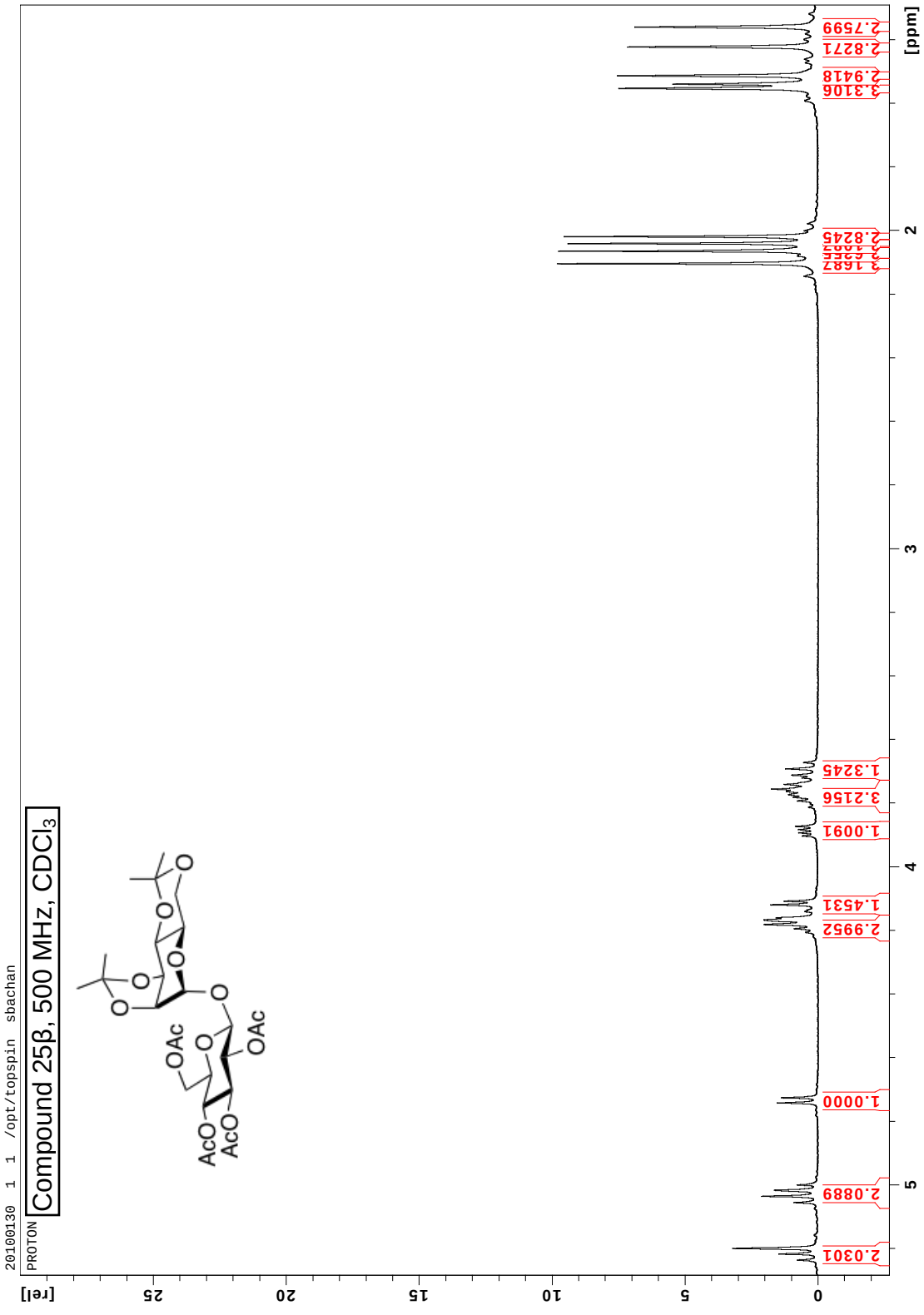
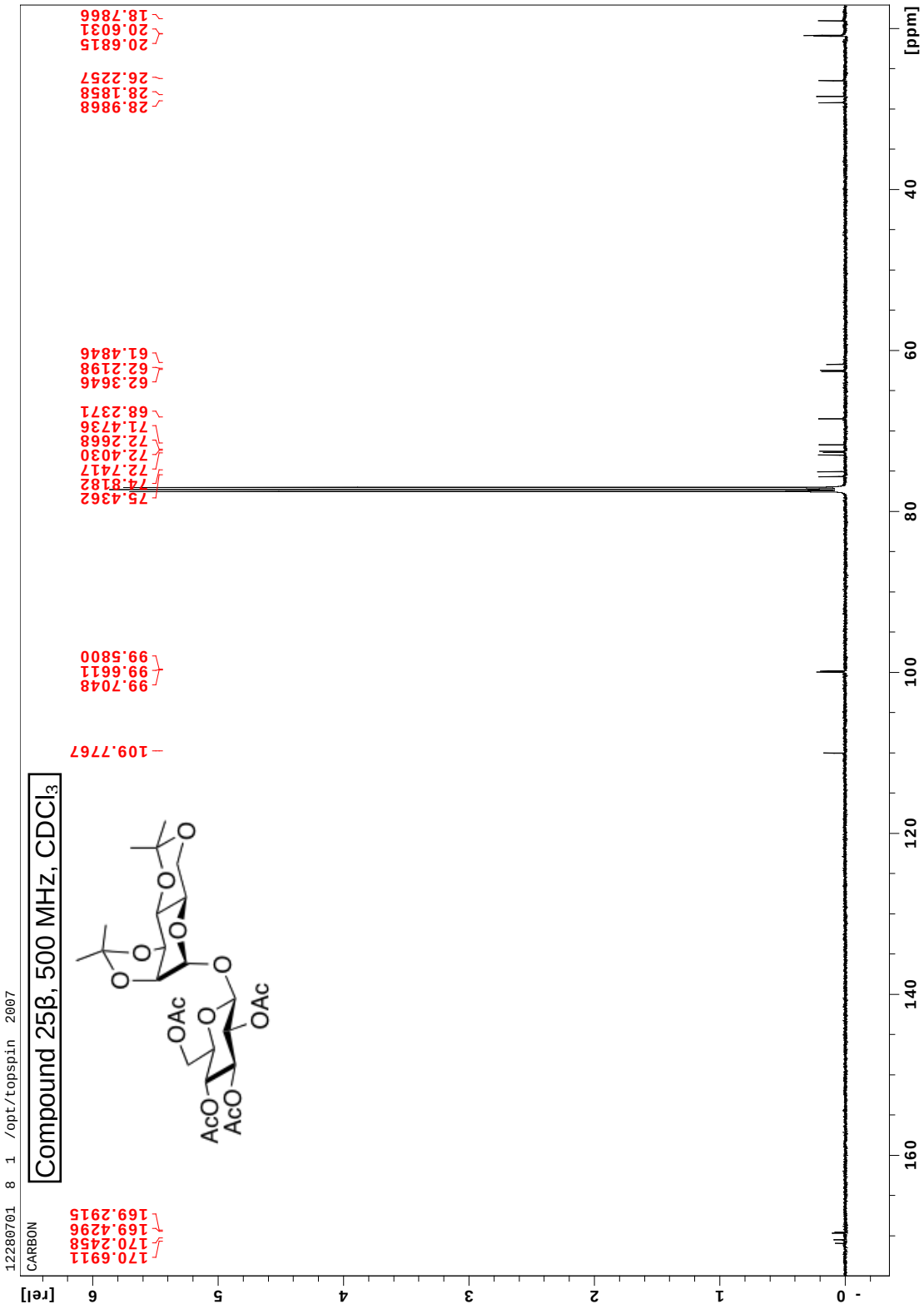
3.0158
2.8997
3.0195
OAc
OAc
OAc
AcO

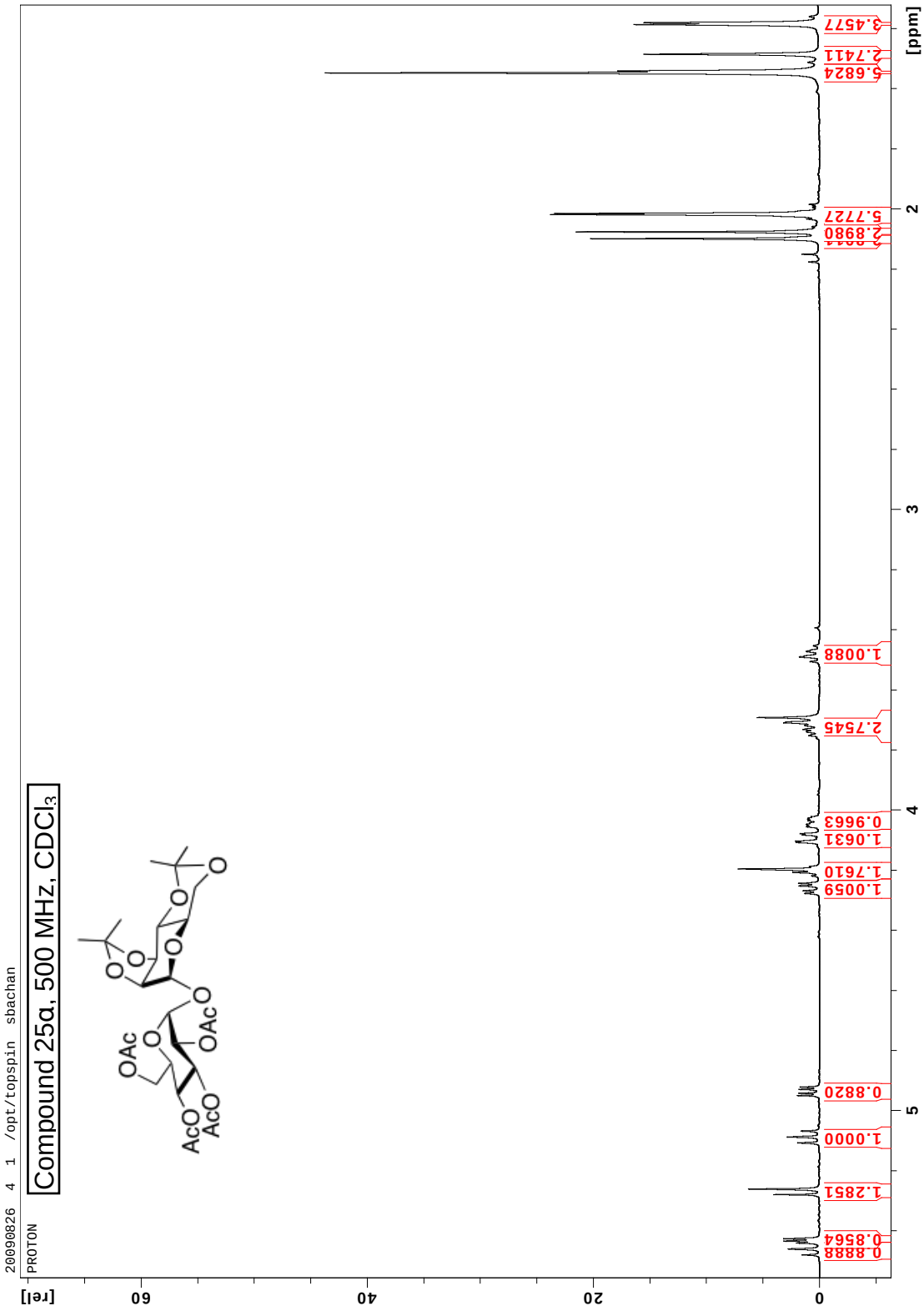

20090821 4 1 /opt/topspin sbachan
CARBON
Compound 24β, 500 MHz, CDCl3
170.7128
170.4451
170.3062
169.6133
109.9802
100.4418
99.9896
99.8574
75.6632
75.0438
72.4917
71.4657
71.0197
69.2275
67.2684
62.3387
62.0183
61.6945
29.2088
28.3891
26.4265
20.9776
20.8983
20.8355
20.7821
18.9754
OAc
OAc
OAc
AcO
O
[rel]
[ppm]

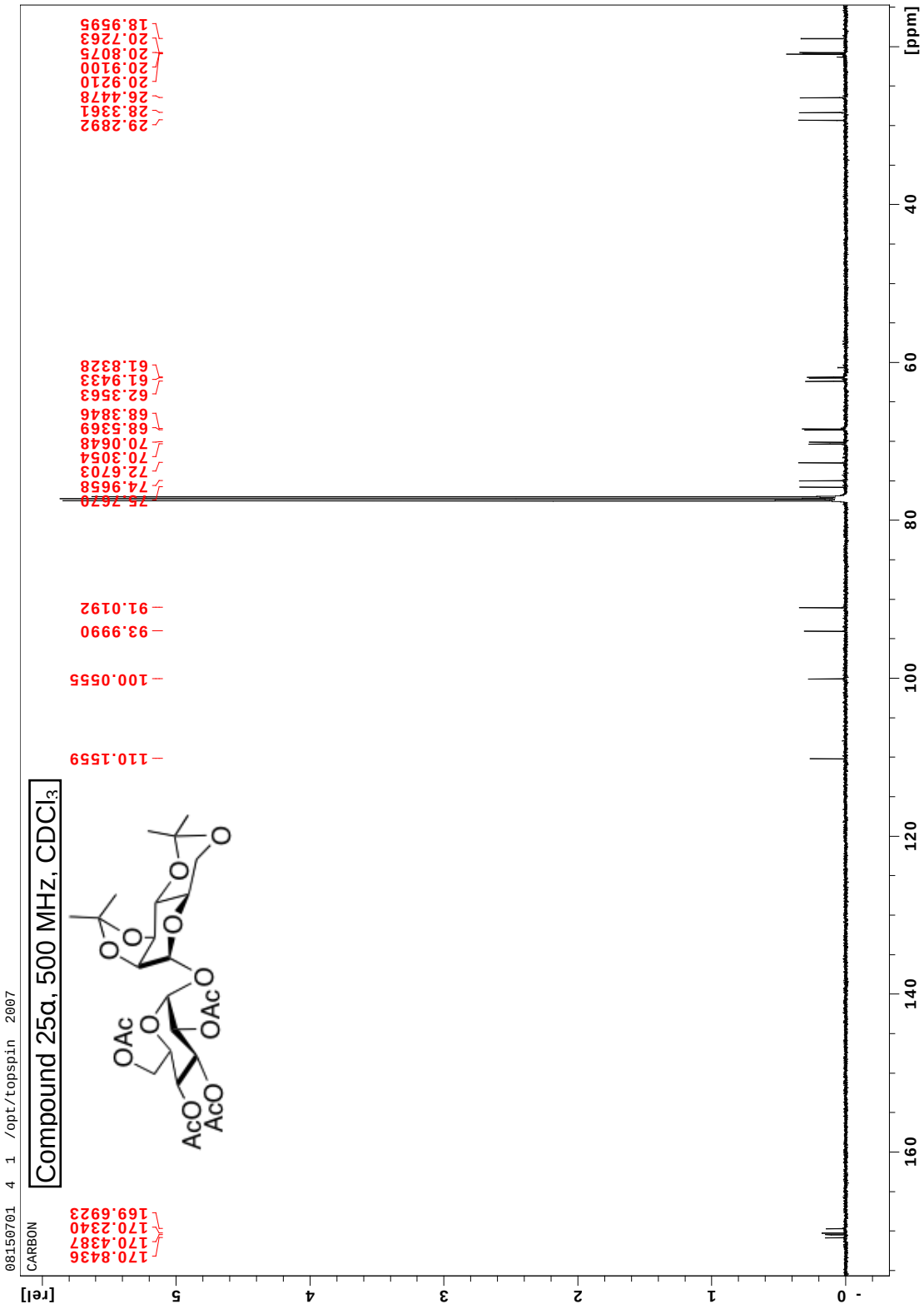

Compound 24a, 500 MHz, CDCl3
HSQCGP
06270501 4 1 /opt/topspin 2005
[rel]
[ppm]
2.0000
1.0321
0.9291
0.9939
0.9865
1.8994
1.1668
1.1380
2.8454
0.9580
2.7144
2.8558
2.7624
2.8746
5.8502
AcO
OAc
OAc
AcO

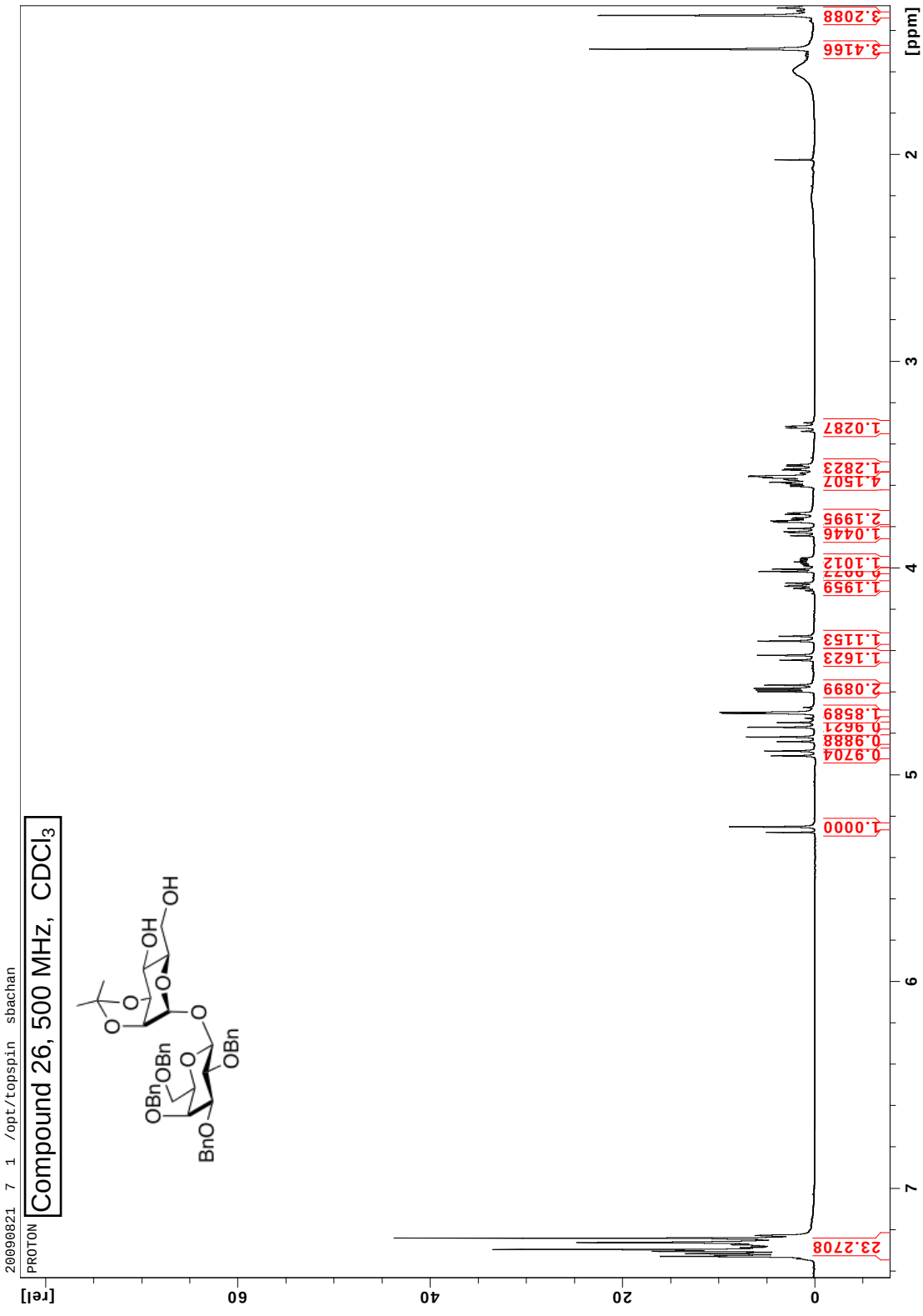

Compound 24a, 500 MHz, CDCl3
20090826 2 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
170.6023
170.4587
170.4143
170.3309
110.1347
100.0305
93.9326
91.5640
75.8002
74.9486
72.6886
67.9106
67.6379
67.5536
67.2782
62.3303
61.9634
61.5761
29.2872
28.3292
26.4124
20.8693
20.8461
20.8194
18.9469
AcO
OAc
OAc
AcO
O
O
O
O
O
O
O

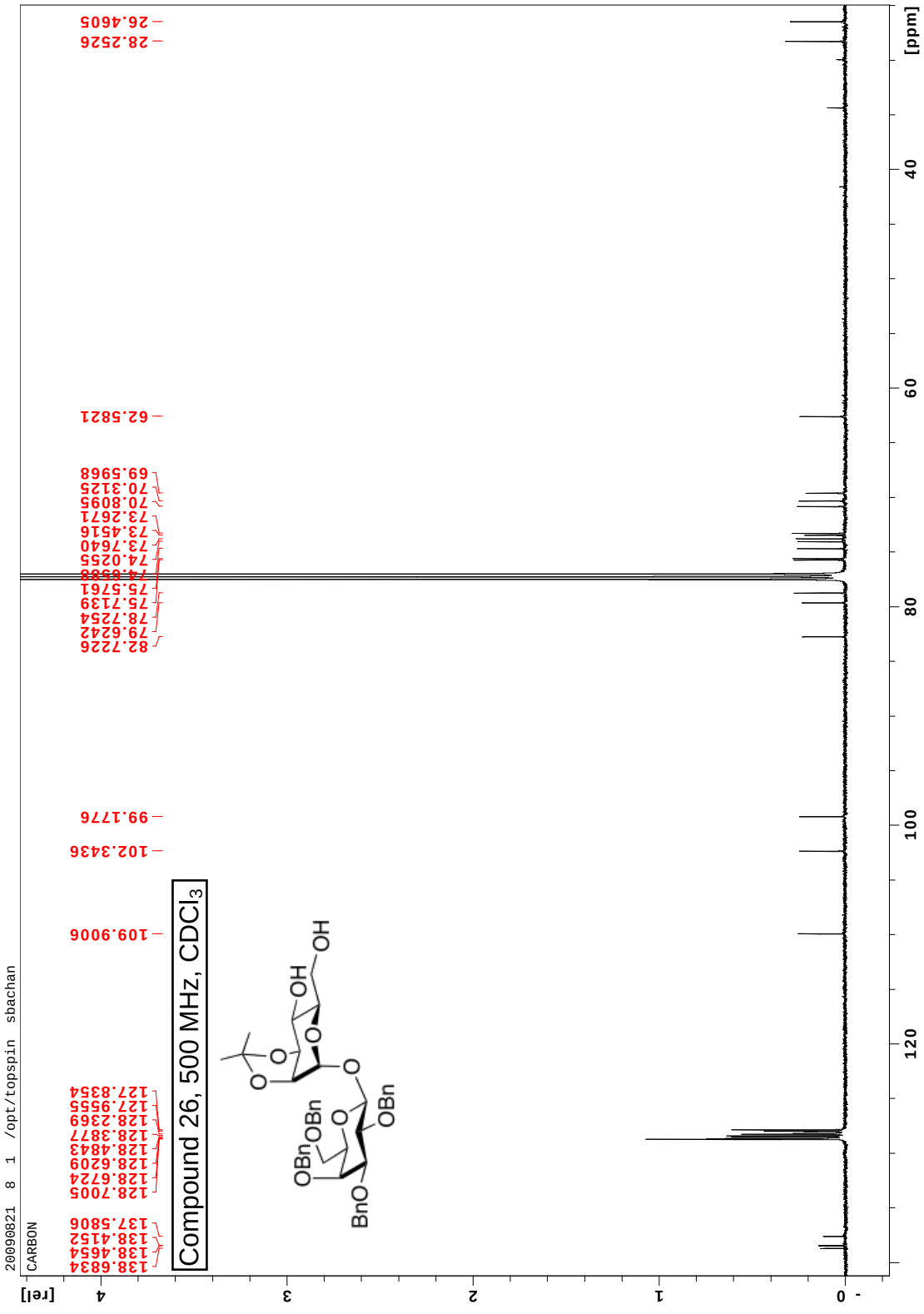

20100130 1 1 /opt/topspin sbachan
PROTON
Compound 25β, 500 MHz, CDCl3
[rel]
[ppm]
2.0301
2.0889
1.0000
2.9952
1.4531
1.0091
3.2156
1.3245
2.8245
2.9418
2.8271
2.7599
OAc
OAc
AcO
AcO

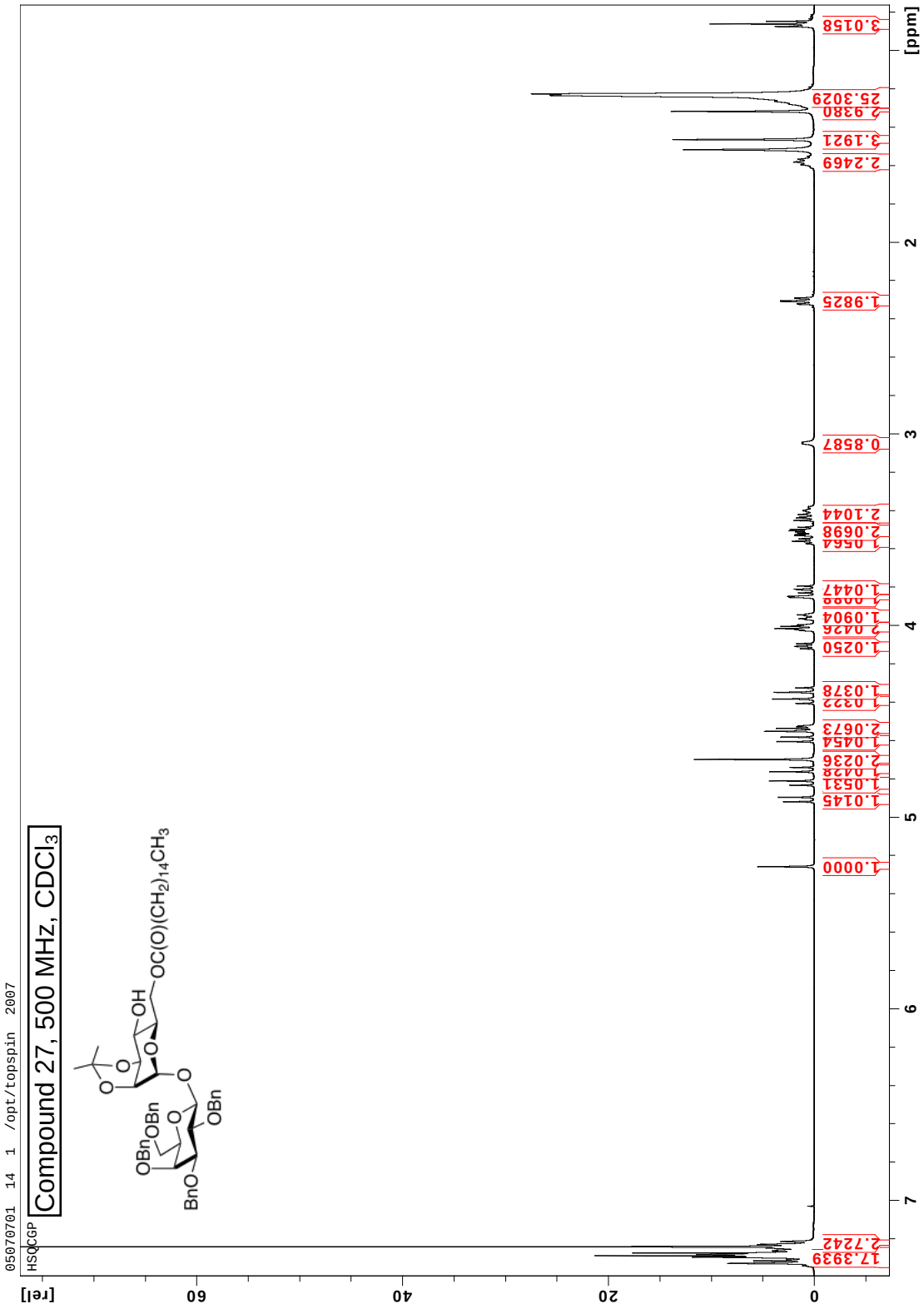

12280701 8 1 /opt/topspin 2007
CARBON
Compound 25β, 500 MHz, CDCl3
170.6911
170.2458
169.4296
169.2915
109.7767
99.7048
99.6611
99.5800
75.4362
74.8182
72.7417
72.4030
72.2668
71.4736
68.2371
62.3646
62.2198
61.4846
28.9868
28.1858
26.2257
20.6815
20.6031
18.7866
OAc
OAc
AcO
AcO
[rel]
[ppm]

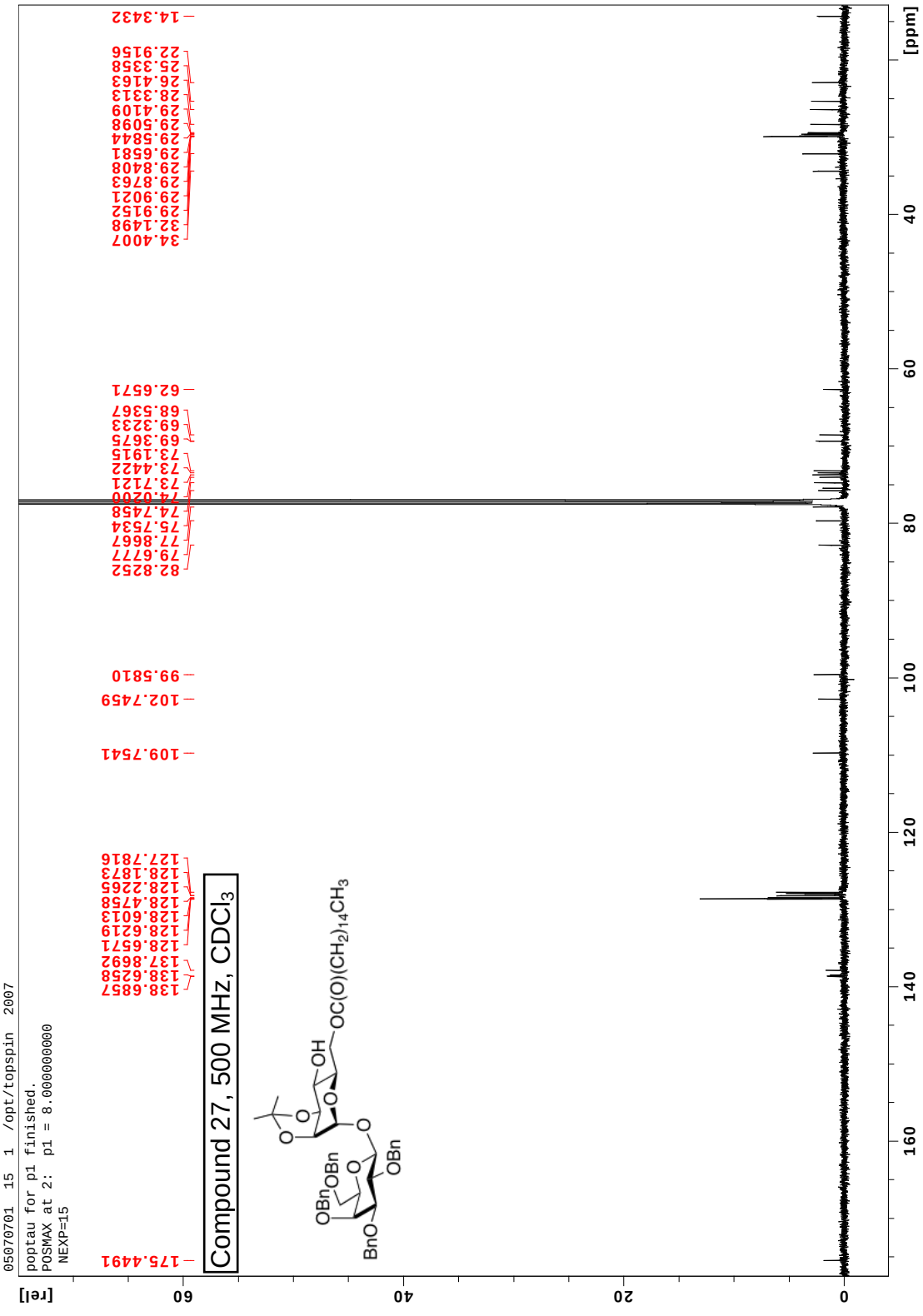

Compound 25a, 500 MHz, CDCl3
20090826 4 1 /opt/topspin sbachan
PROTON
[rel]
60
40
20
0
[ppm]
5
4
3
2
0.8888
0.8564
1.2851
1.0000
0.8820
1.0059
1.7610
1.0631
0.9663
2.7545
1.0088
5.6824
2.7411
3.4577
5.7727
2.8980
OAc
OAc
AcO
AcO
O

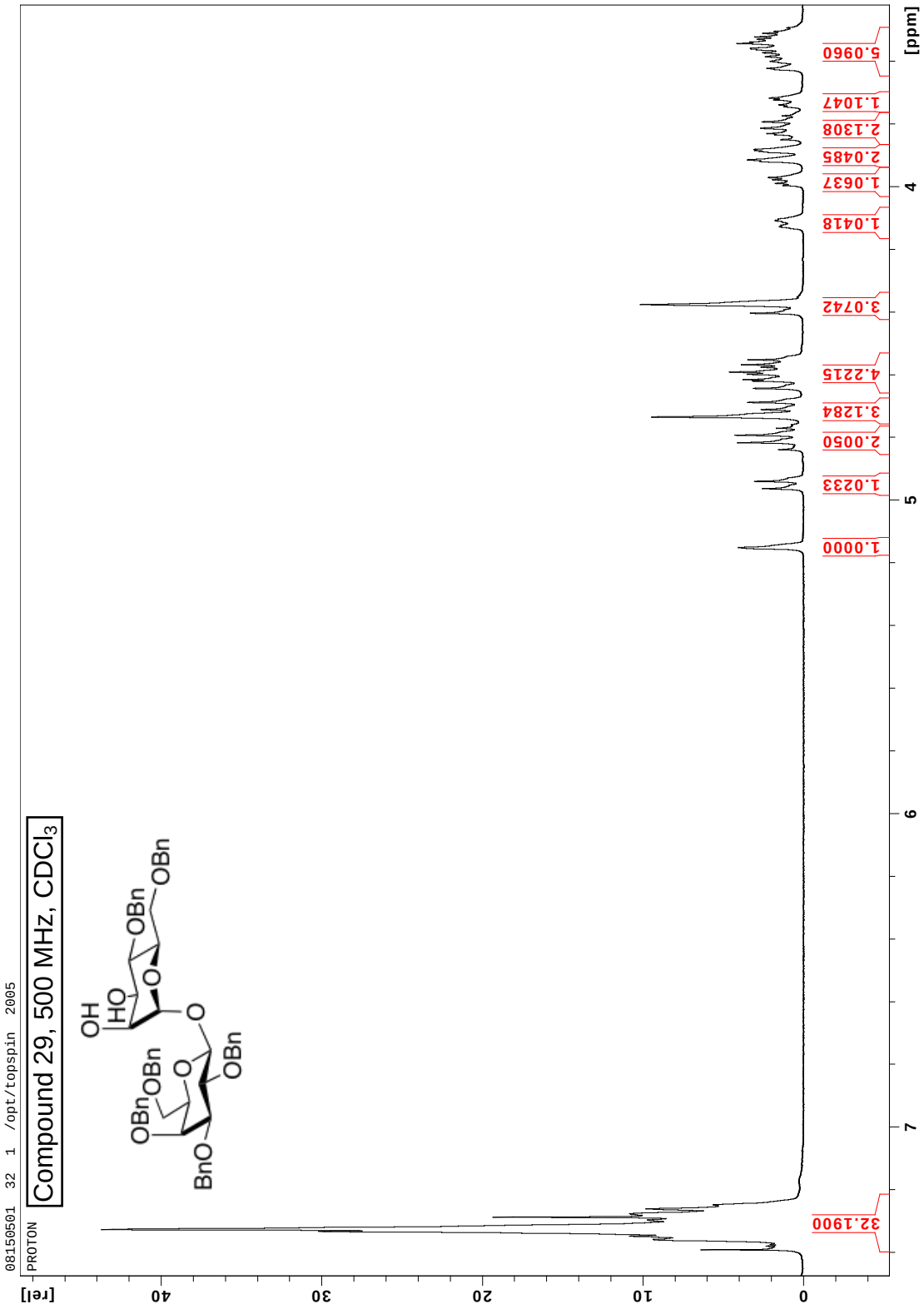

Compound 25a, 500 MHz, CDCl3
CARBON
08150701 4 1 /opt/topspin 2007
[rel]
[ppm]
170.8436
170.4387
170.2340
169.6923
110.1559
100.0555
93.9990
91.0192
74.9658
72.6703
70.3054
70.0648
68.5369
68.3846
62.3563
61.9433
61.8328
29.2892
28.3361
26.4478
20.9210
20.9100
20.8075
20.7263
18.9595
OAc
OAc
AcO
AcO
OAc
O

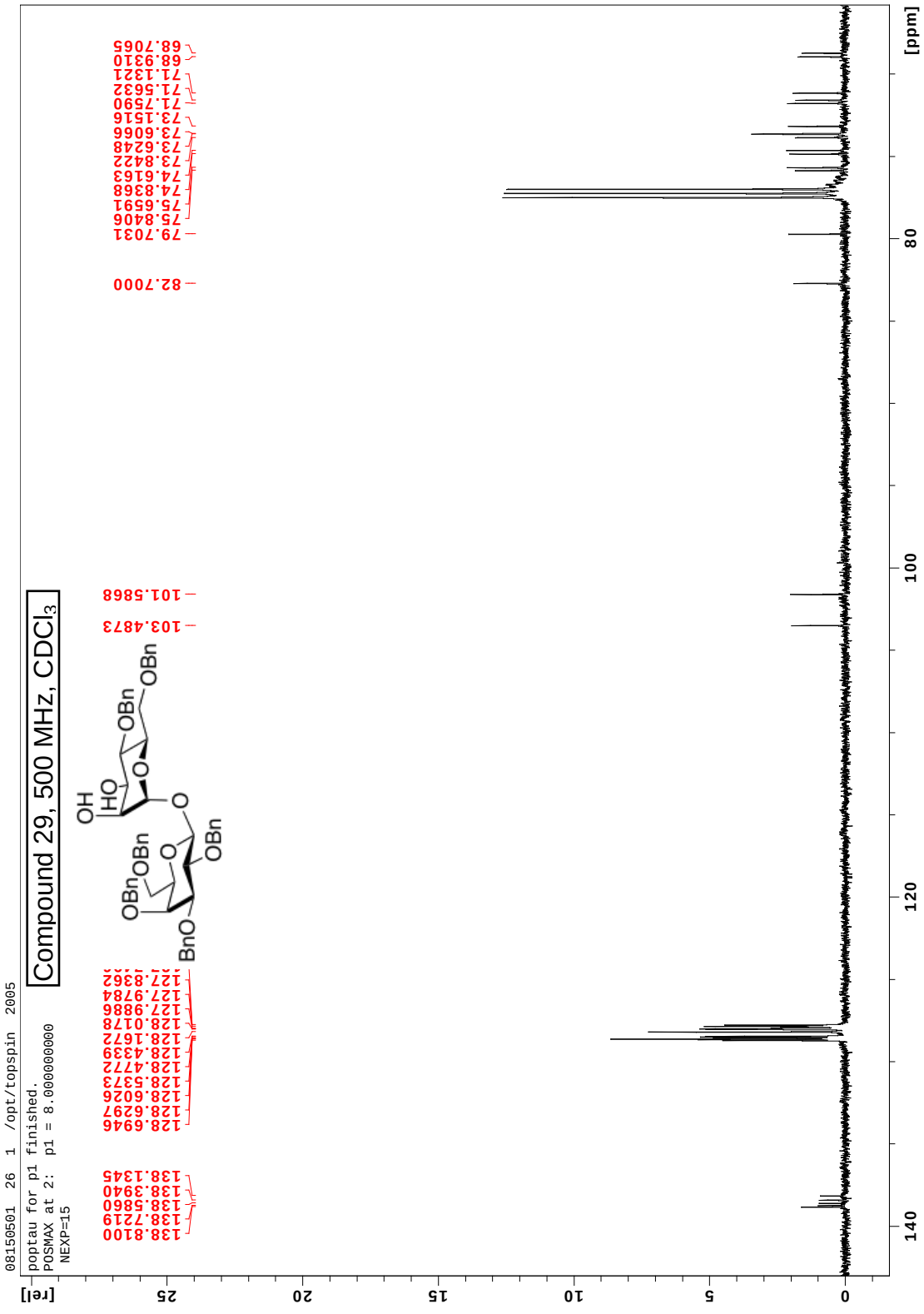

20090821 7 1 /opt/topspin sbachan
PROTON
Compound 26, 500 MHz, CDCl3
[rel]
[ppm]
23.2708
1.0000
0.9704
0.9888
0.9621
1.8589
2.0899
1.1623
1.1153
1.1959
1.1012
1.0446
2.1995
4.1507
1.2823
1.0287
3.4166
3.2088
OH
OH
O
O
O
O
OBn
OBn
OBn
BnO

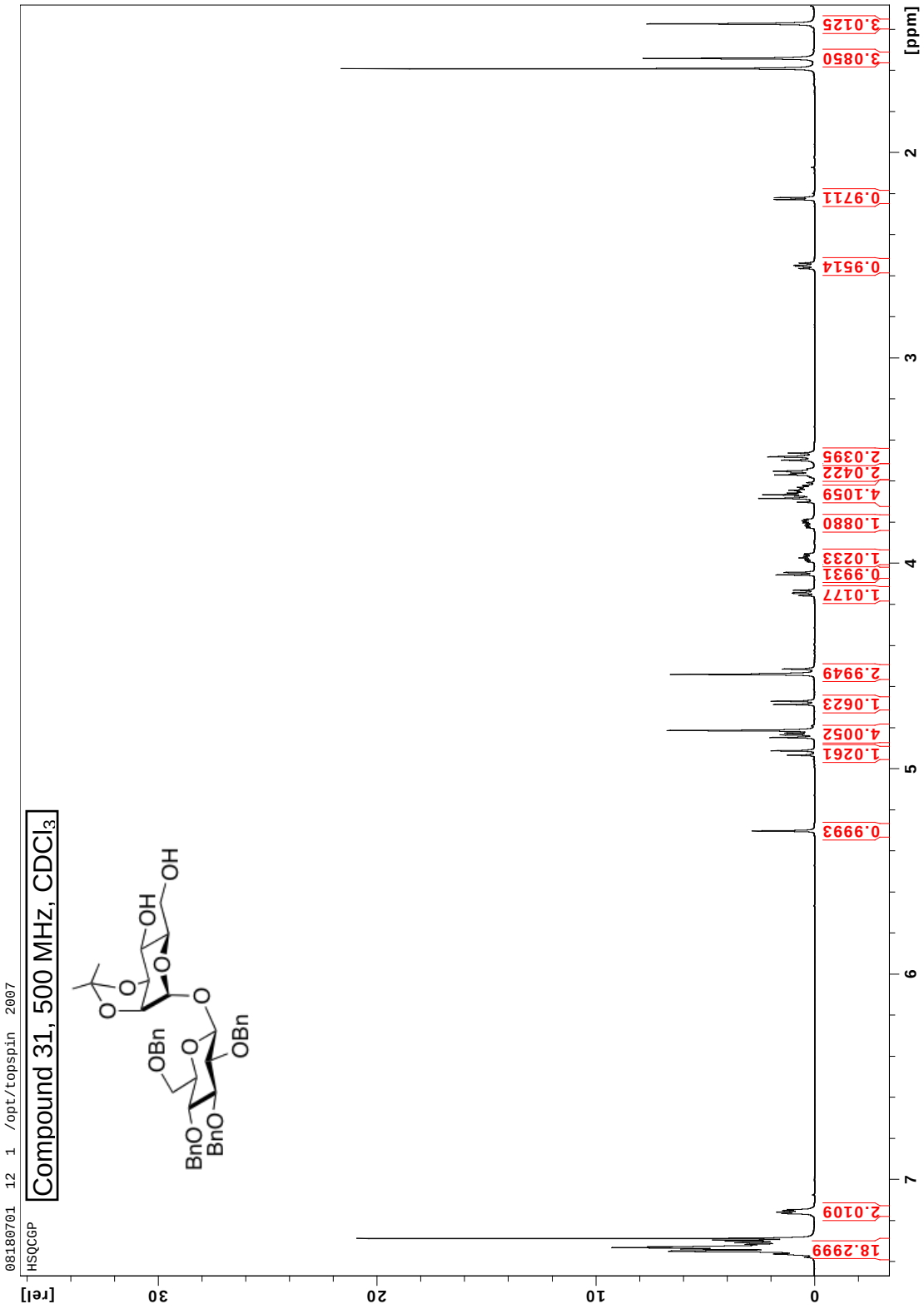

Compound 26, 500 MHz, CDCl3
20090821 8 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
138.6834
138.4654
138.4152
137.5806
128.7005
128.6724
128.6209
128.4843
128.3877
128.2369
127.9555
127.8354
109.9006
102.3436
99.1776
82.7226
79.6242
78.7254
75.7139
75.5761
74.0255
73.7640
73.4516
73.2671
70.8095
70.3125
69.5968
62.5821
28.2526
26.4605
OBn
OBn
OBn
BnO
HO
OH
O

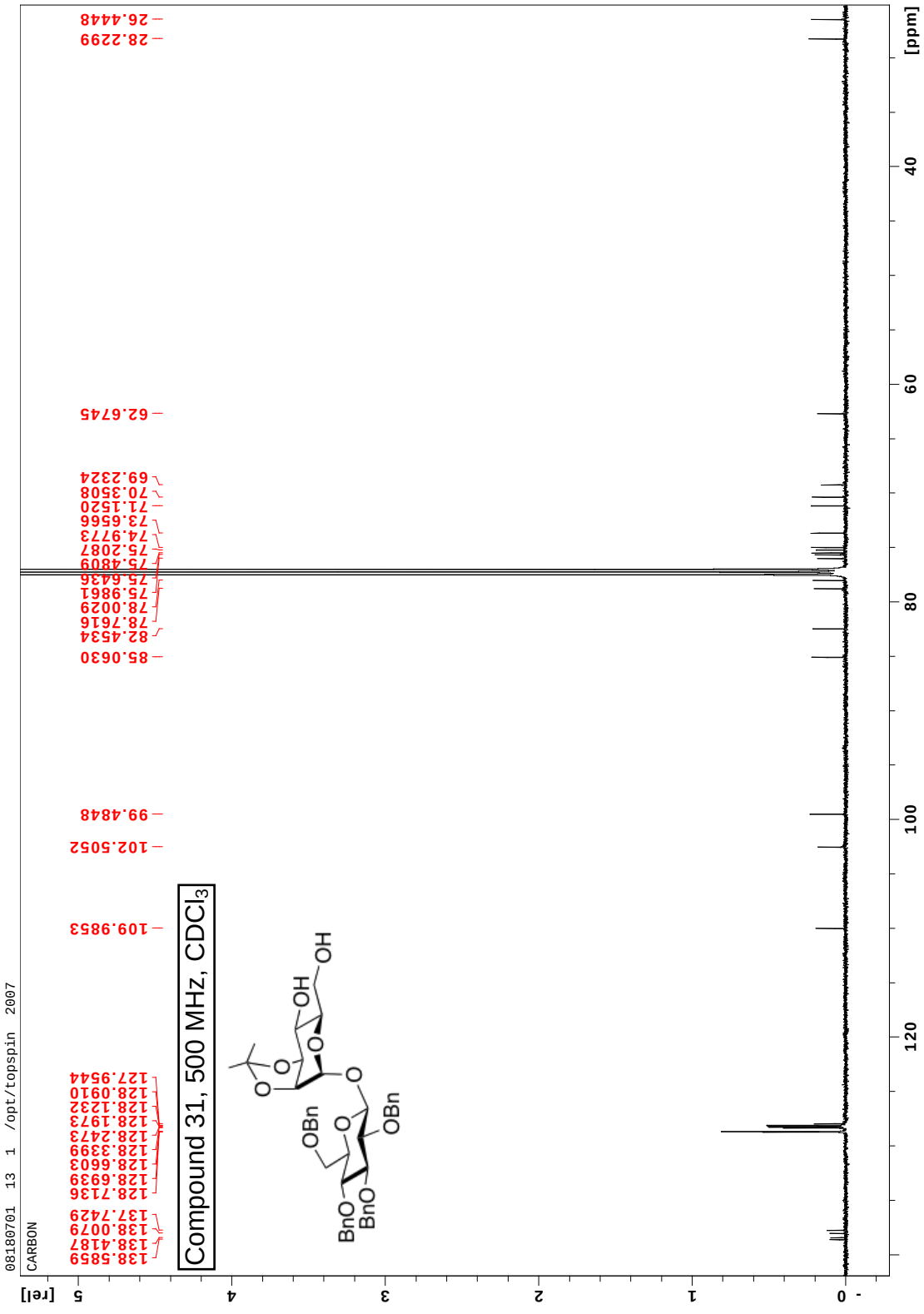

05070701 14 1 /opt/topspin 2007
HSQCGP
Compound 27, 500 MHz, CDCl3
OBn
OBn
OBn
BnO
OH
OC(O)(CH2)14CH3
[rel]
0
20
40
60
[ppm]
7
6
5
4
3
2
17.3939
2.7242
1.0000
1.0145
1.0531
1.0428
2.0236
1.0454
2.0673
1.0322
1.0378
1.0250
2.0425
1.0904
1.0447
1.0564
2.0698
2.1044
0.8587
1.9825
2.2469
3.1921
2.9380
25.3029
3.0158

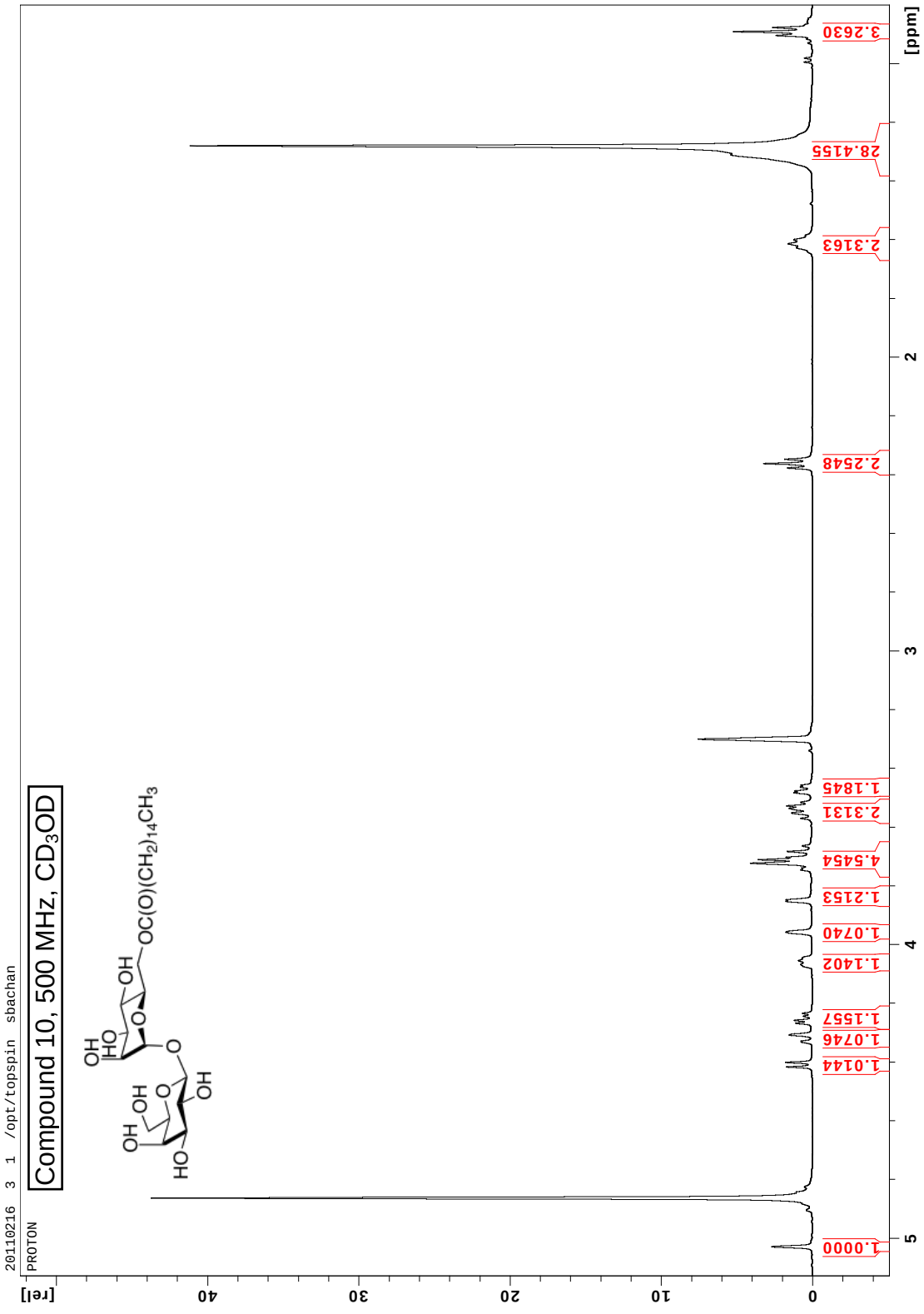

Compound 27, 500 MHz, CDCl3
05070701 15 1 /opt/topspin 2007
poptau for p1 finished.
POSMAX at 2: p1 = 8.000000000
NEXP=15
[rel]
[ppm]
175.4491
138.6857
138.6258
137.8692
128.6571
128.6219
128.6013
128.4758
128.2265
128.1873
127.7816
109.7541
102.7459
99.5810
82.8252
79.6777
77.8667
75.7534
74.7458
74.0200
73.7121
73.4422
73.1915
69.3675
69.3233
68.5367
62.6571
34.4007
32.1498
29.9152
29.9021
29.8763
29.8408
29.6581
29.5844
29.5098
29.4109
28.3313
26.4163
25.3358
22.9156
14.3432
OC(O)(CH2)14CH3
OH
OBn
OBn
OBn
BnO

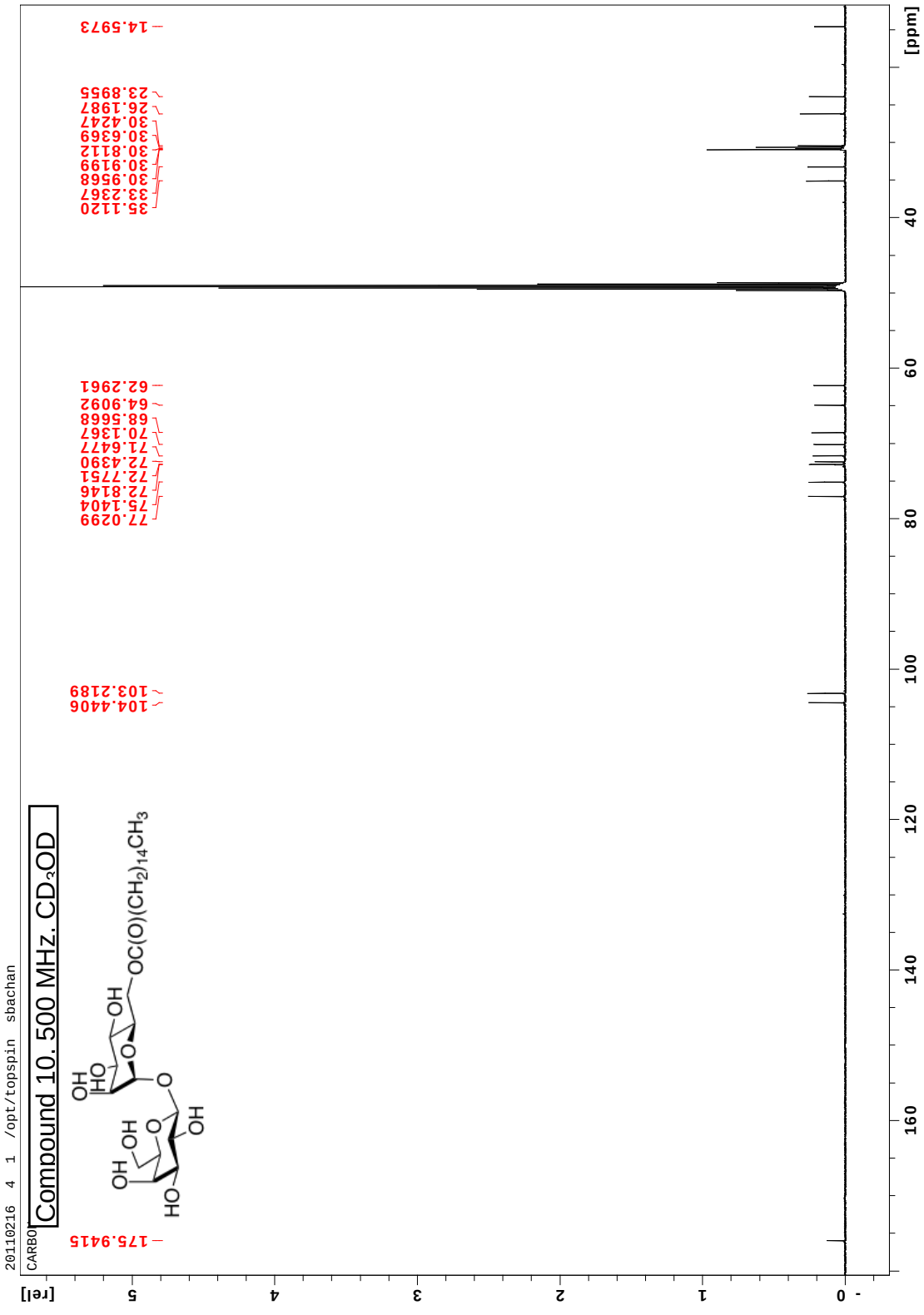

08150501 32 1 /opt/topspin 2005
PROTON
Compound 29, 500 MHz, CDCl3
[rel]
[ppm]
32.1900
1.0000
1.0233
2.0050
3.1284
4.2215
3.0742
1.0418
1.0637
2.0485
2.1308
1.1047
5.0960

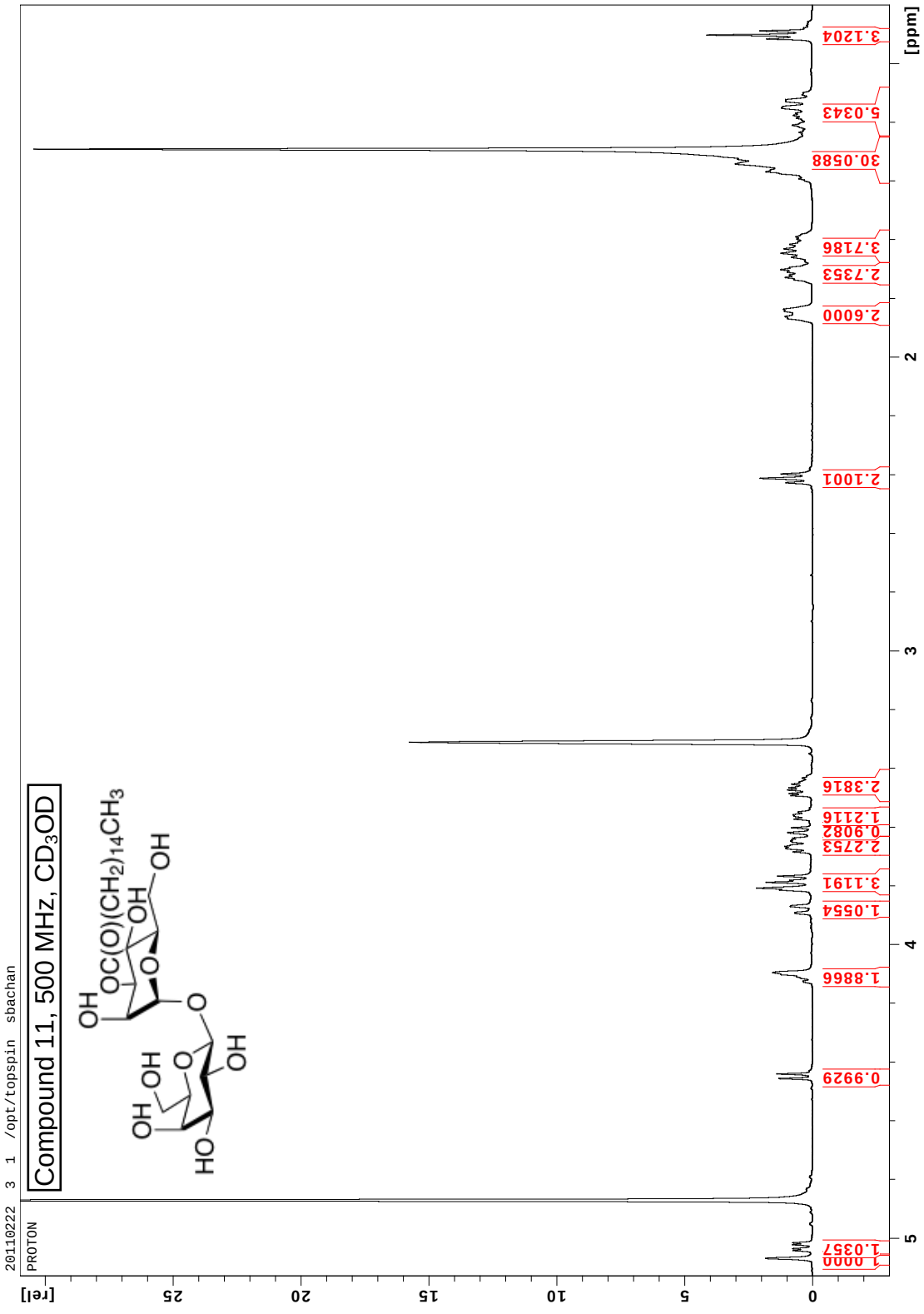

Compound 29, 500 MHz, CDCl3
08150501 26 1 /opt/topspin 2005
poptau for p1 finished.
POSMAX at 2: p1 = 8.000000000
NEXP=15
[rel]
[ppm]
0
5
10
15
20
25
80
100
120
140
138.8100
138.7219
138.5860
138.3940
138.1345
128.6946
128.6297
128.6026
128.5373
128.4772
128.4339
128.1672
128.0178
127.9886
127.9784
127.8362
101.5868
103.4873
82.7000
79.7031
75.8406
75.6591
74.8368
74.6163
73.8422
73.6248
73.6066
73.1516
71.7590
71.5632
71.1321
68.9310
68.7065
OH
HO
O
OBn
OBn
OBn
OBn
OBn
BnO

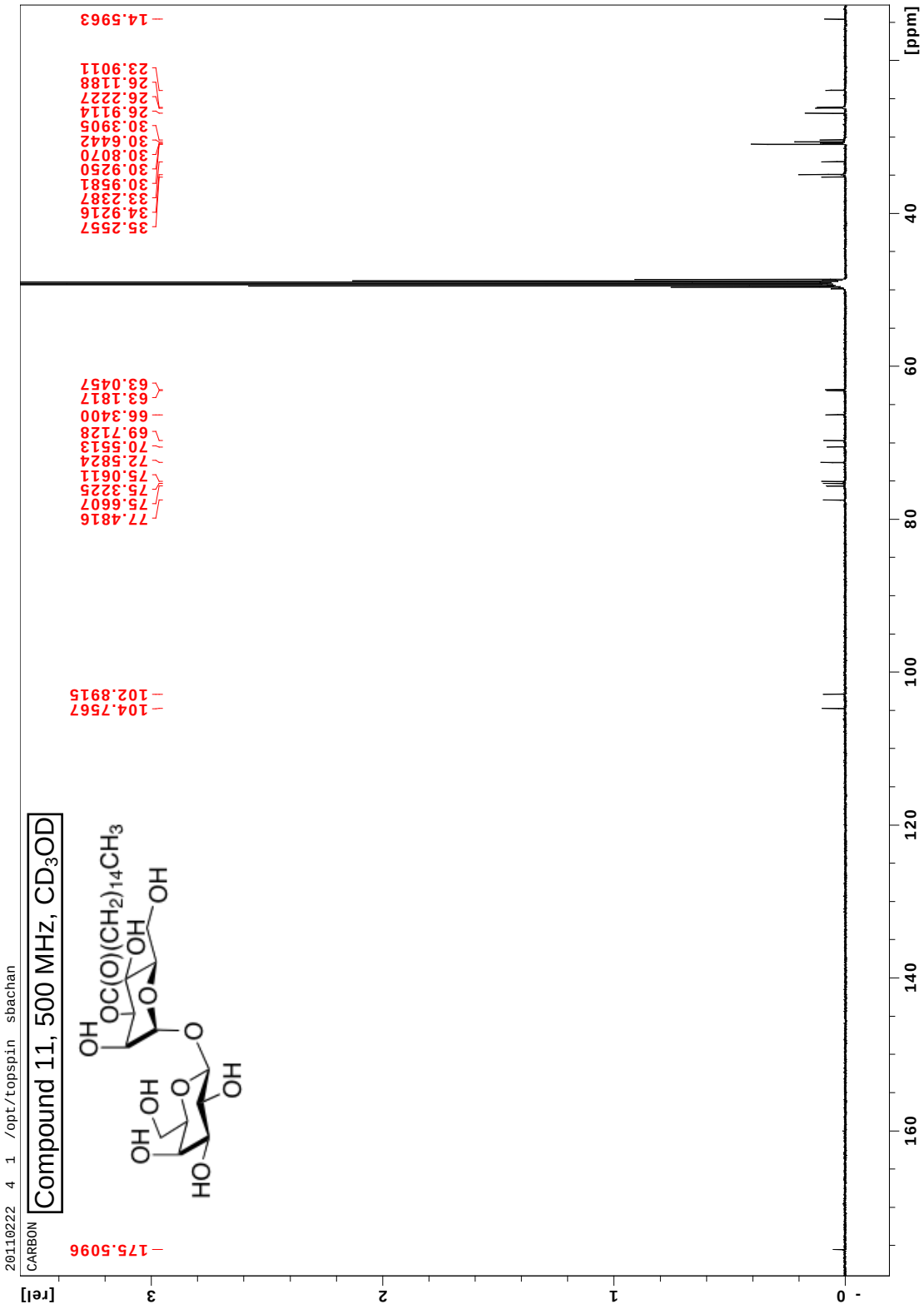

08180701 12 1 /opt/topspin 2007
HSQCGP
Compound 31, 500 MHz, CDCl3
[rel]
[ppm]
18.2999
2.0109
0.9993
1.0261
4.0052
1.0623
2.9949
1.0177
0.9931
1.0233
1.0880
4.1059
2.0422
2.0395
0.9514
0.9711
3.0850
3.0125

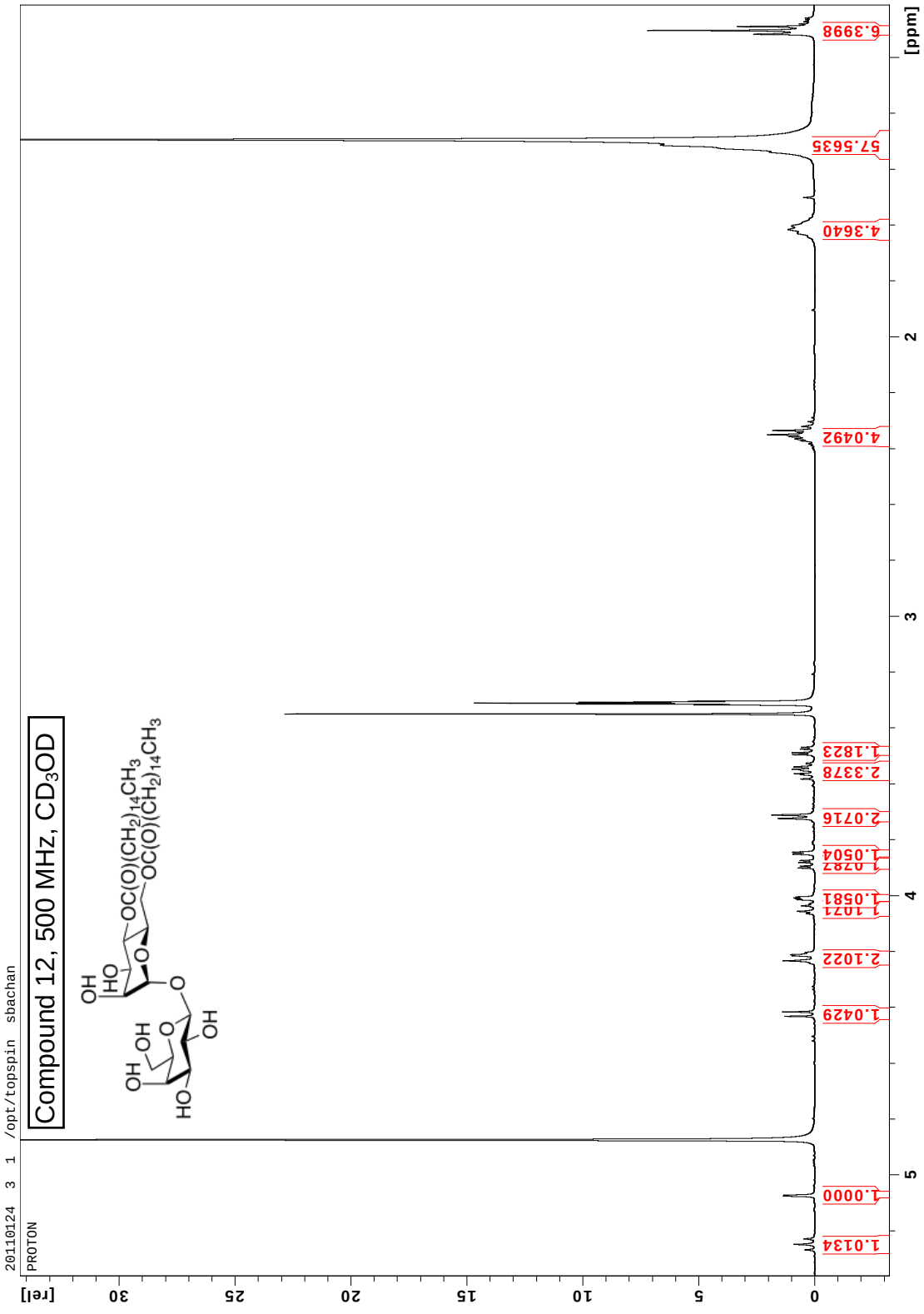

Compound 31, 500 MHz, CDCl3
138.5859
138.4187
138.0079
137.7429
128.7136
128.6939
128.6603
128.3399
128.2473
128.1973
128.1232
128.0910
127.9544
109.9853
102.5052
99.4848
85.0630
82.4534
78.7616
78.0029
75.9861
75.6436
75.4809
75.2087
74.9773
73.6566
71.1520
70.3508
69.2324
62.6745
28.2299
26.4448
CARBON
08180701 13 1 /opt/topspin 2007
[rel]
[ppm]

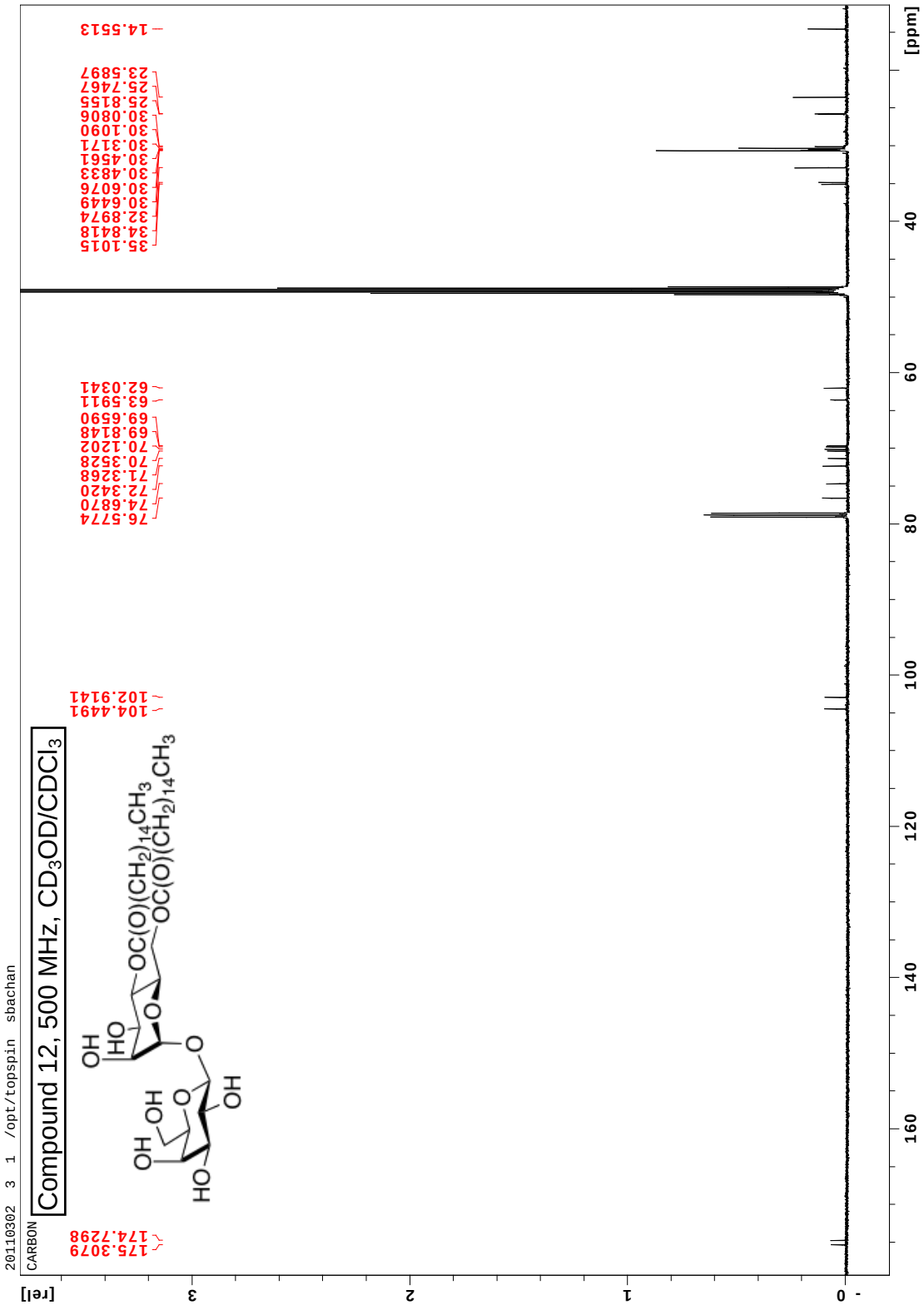

20110216 3 1 /opt/topspin sbachan
PROTON
Compound 10, 500 MHz, CD3OD
[rel]
[ppm]
1.0000
3.2630
28.4155
2.3163
2.2548
1.1845
2.3131
4.5454
1.2153
1.0740
1.1402
1.0144
1.0746
1.1557

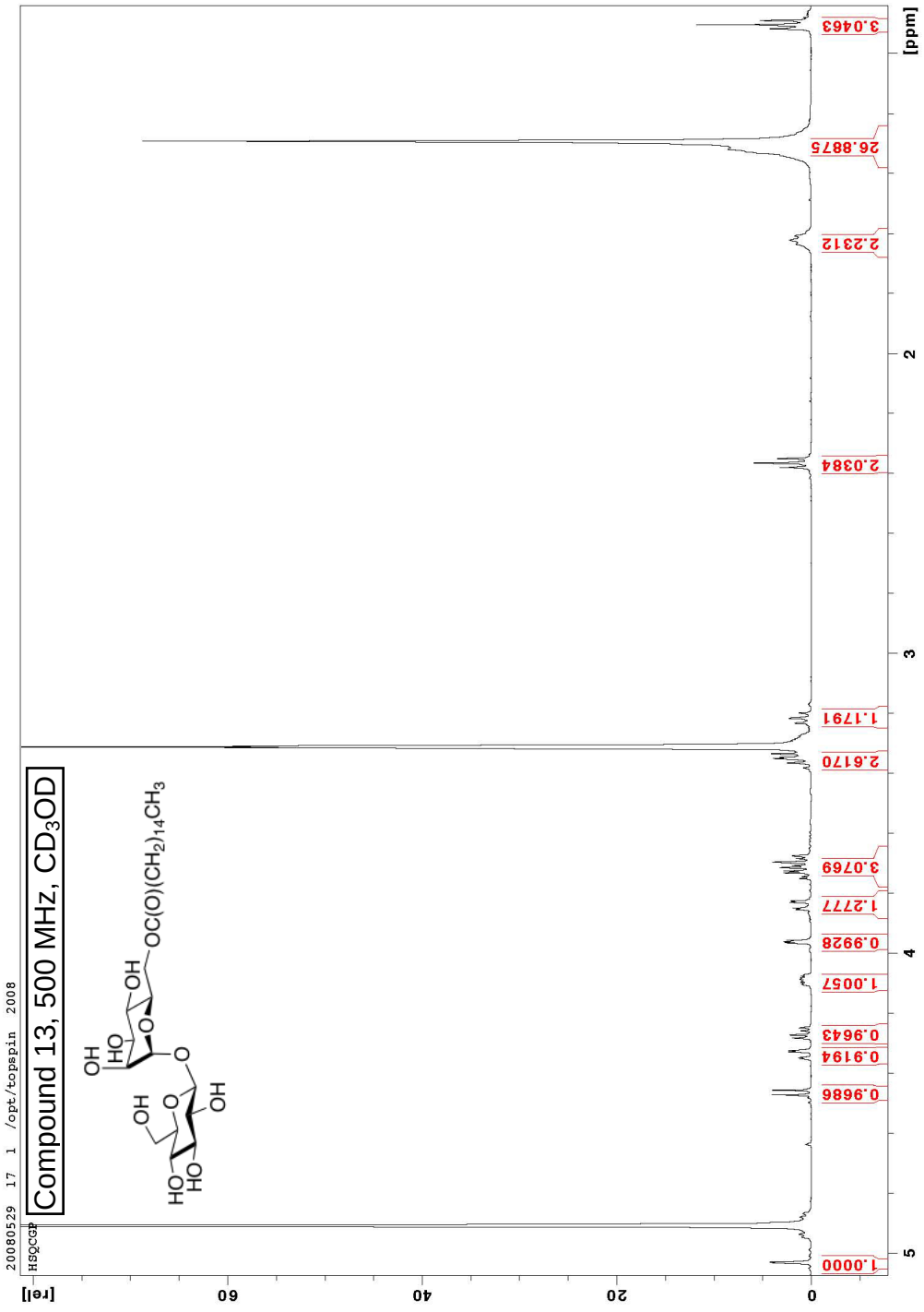

Compound 10. 500 MHz. CD3OD
CARBON
20110216 4 1 /opt/topspin sbachan
175.9415
104.4406
103.2189
77.0299
75.1404
72.8146
72.7751
72.4390
71.6477
70.1367
68.5668
64.9092
62.2961
35.1120
33.2367
30.9568
30.9199
30.8112
30.6369
30.4247
26.1987
23.8955
14.5973
OC(O)(CH2)14CH3
[rel]
[ppm]

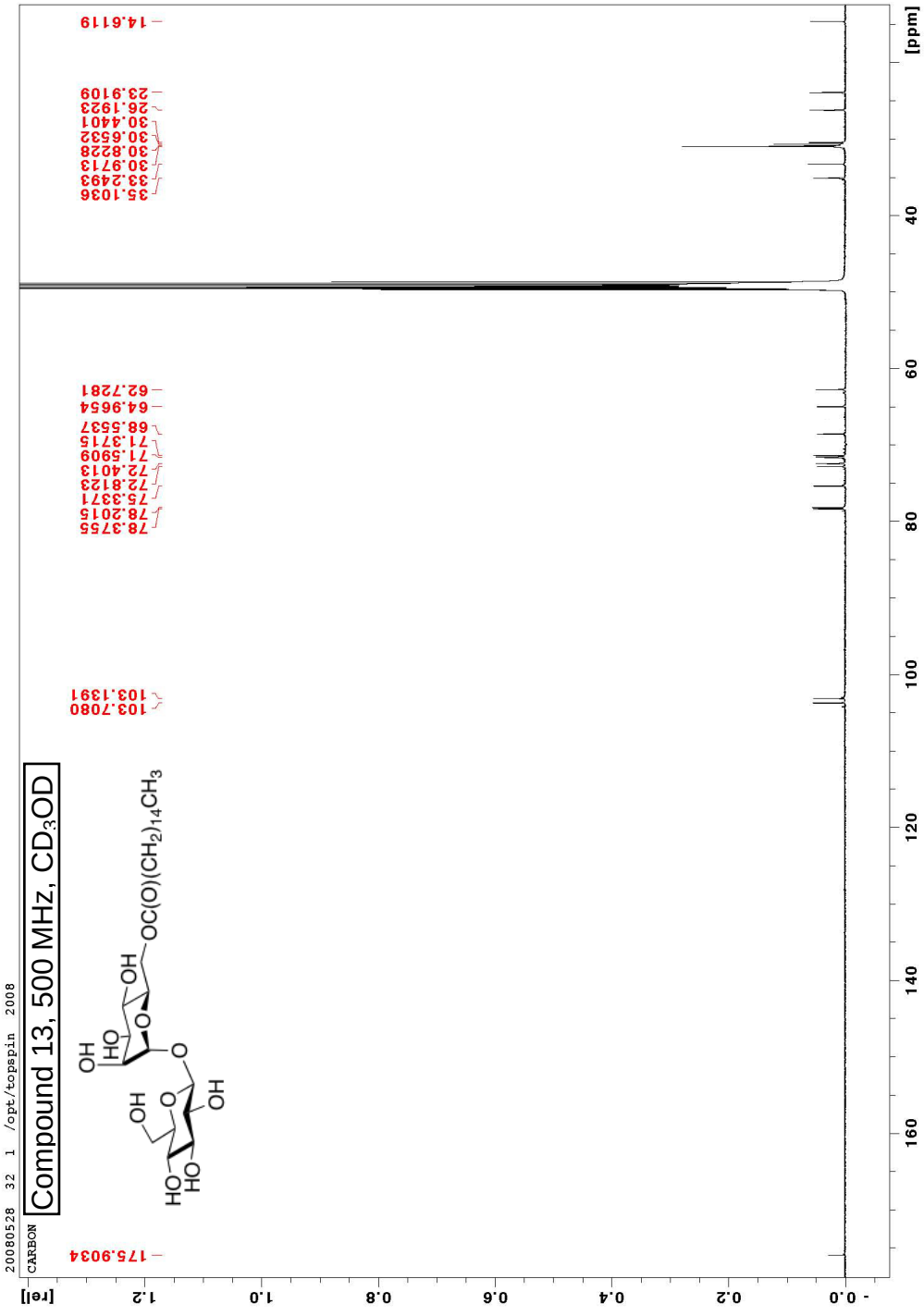

20110222 3 1 /opt/topspin sbachan
PROTON
Compound 11, 500 MHz, CD3OD
OH
OC(O)(CH2)14CH3
OH
O
O
OH
HO
OH
OH
O
HO
[rel]
25
20
15
10
5
0
5
4
3
2
[ppm]
1.0000
1.0357
0.9929
1.8866
1.0554
3.1191
2.2753
0.9082
1.2116
2.3816
2.1001
2.6000
2.7353
3.7186
30.0588
5.0343
3.1204

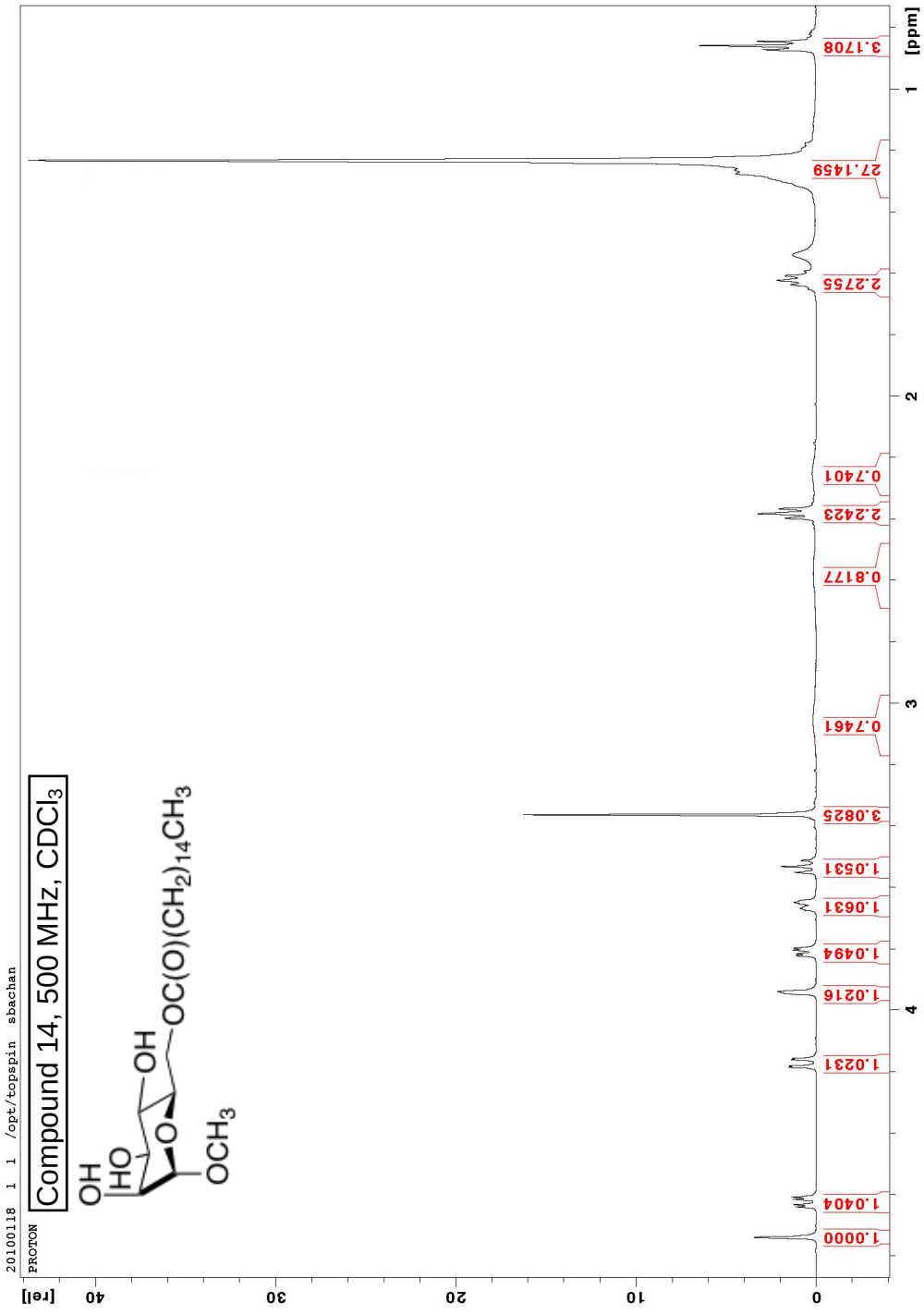

20110222 4 1 /opt/topspin sbachan
CARBON
Compound 11, 500 MHz, CD3OD
175.5096
104.7567
102.8915
77.4816
75.6607
75.3225
75.0611
72.5824
70.5513
69.7128
66.3400
63.1817
63.0457
35.2557
34.9216
33.2387
30.9581
30.9250
30.8070
30.6442
30.3905
26.9114
26.2227
26.1188
23.9011
14.5963
[rel]
[ppm]

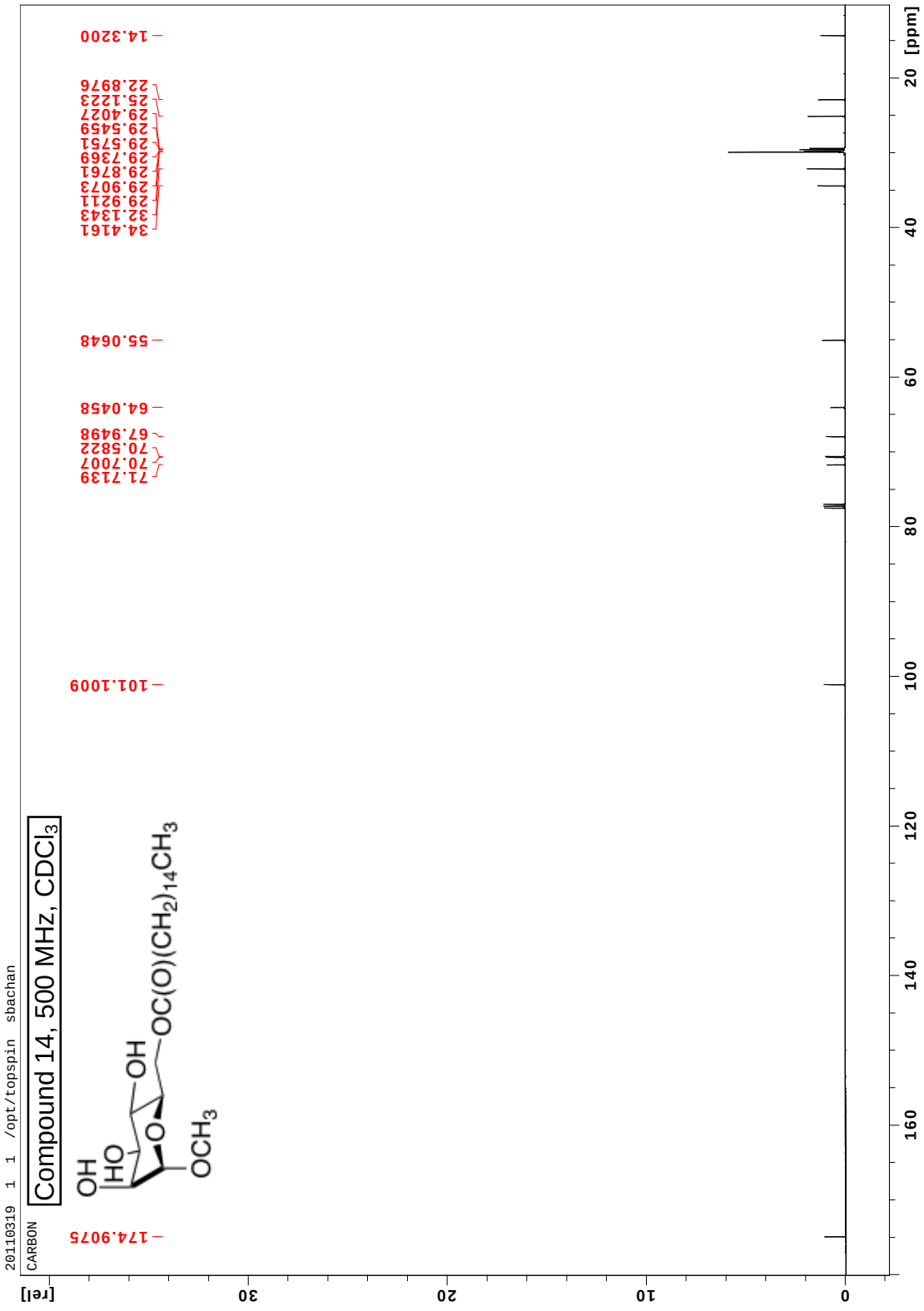

20110124 3 1 /opt/topspin sbachan
PROTON
Compound 12, 500 MHz, CD3OD
[rel]
[ppm]
1.0134
1.0000
1.0429
2.1022
1.1071
1.0581
1.0282
1.0504
2.0716
2.3378
1.1823
4.0492
4.3640
57.5635
6.3998

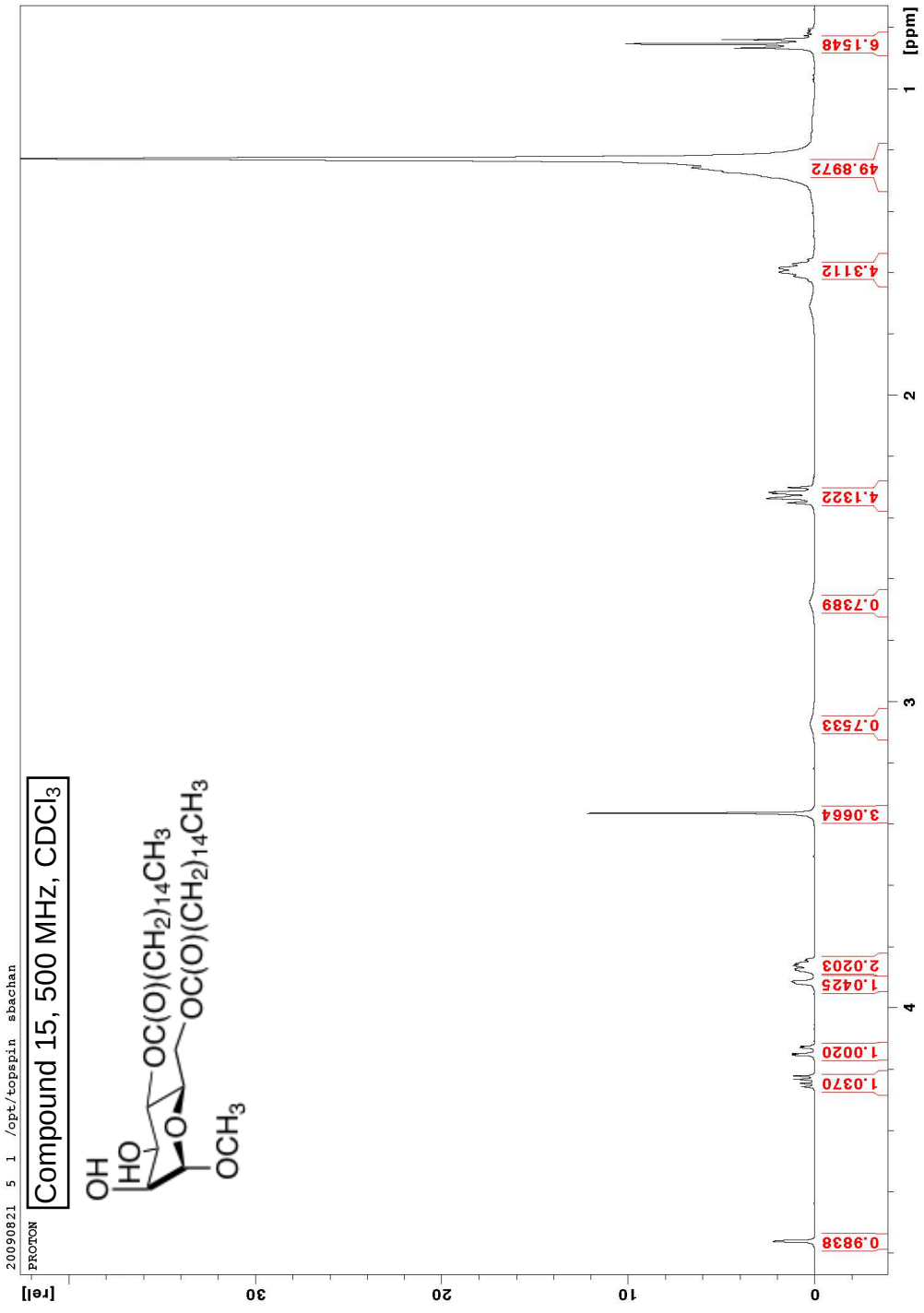

Compound 12, 500 MHz, CD3OD/CDCl3
20110302 3 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
175.3079
174.7298
104.4491
102.9141
76.5774
74.6870
72.3420
71.3268
70.3528
70.1202
69.8148
69.6590
63.5911
62.0341
35.1015
34.8418
32.8974
30.6449
30.6076
30.4833
30.4561
30.3171
30.1090
30.0806
25.8155
25.7467
23.5897
14.5513
OC(O)(CH2)14CH3
OC(O)(CH2)14CH3

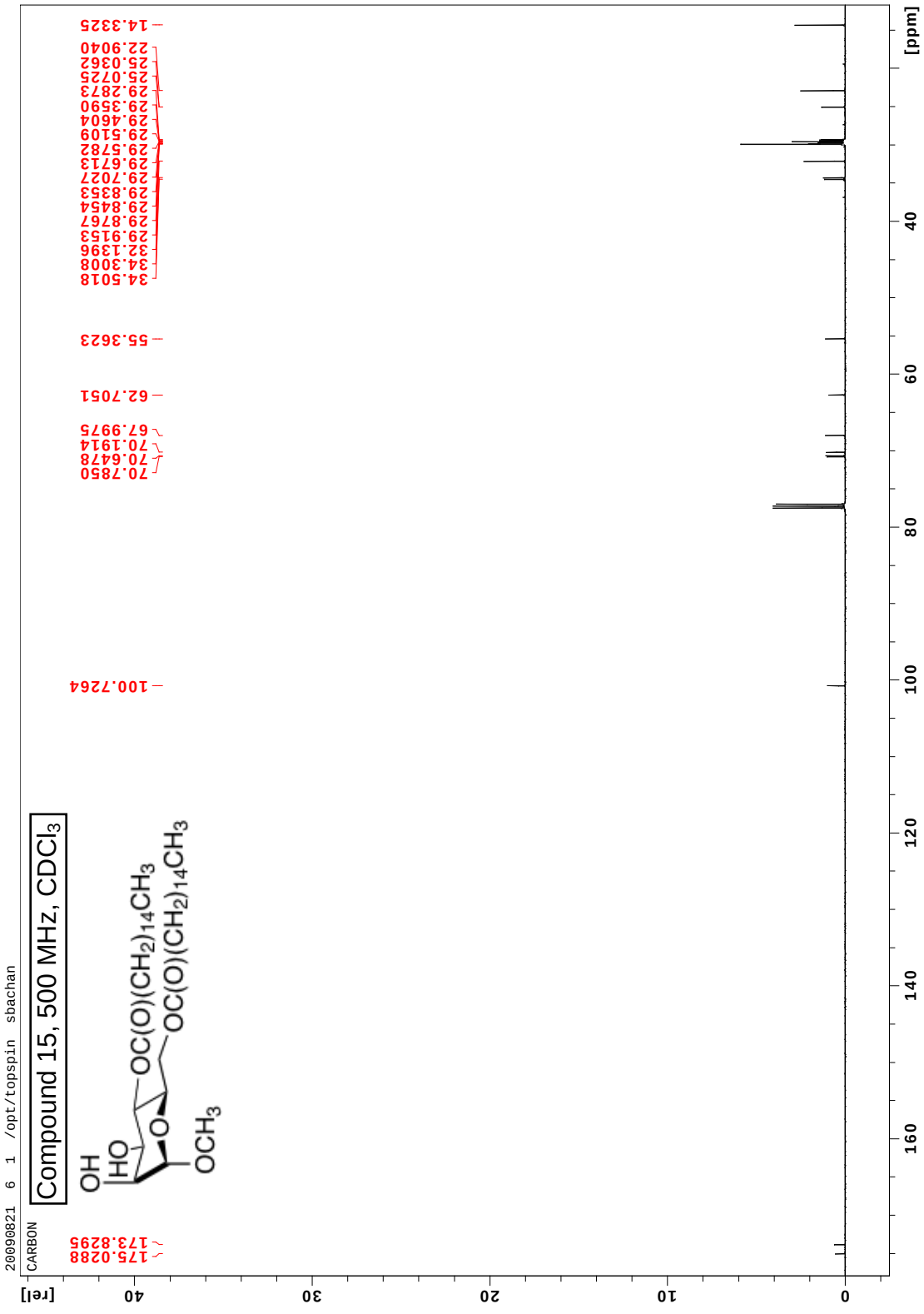

Compound 13, 500 MHz, CD3OD
20080529 17 1 /opt/topspin 2008
HSQCGP
[rel]
[ppm]
1.0000
0.9686
0.9194
0.9643
1.0057
0.9928
1.2777
3.0769
2.6170
1.1791
2.0384
26.8875
2.2312
3.0463

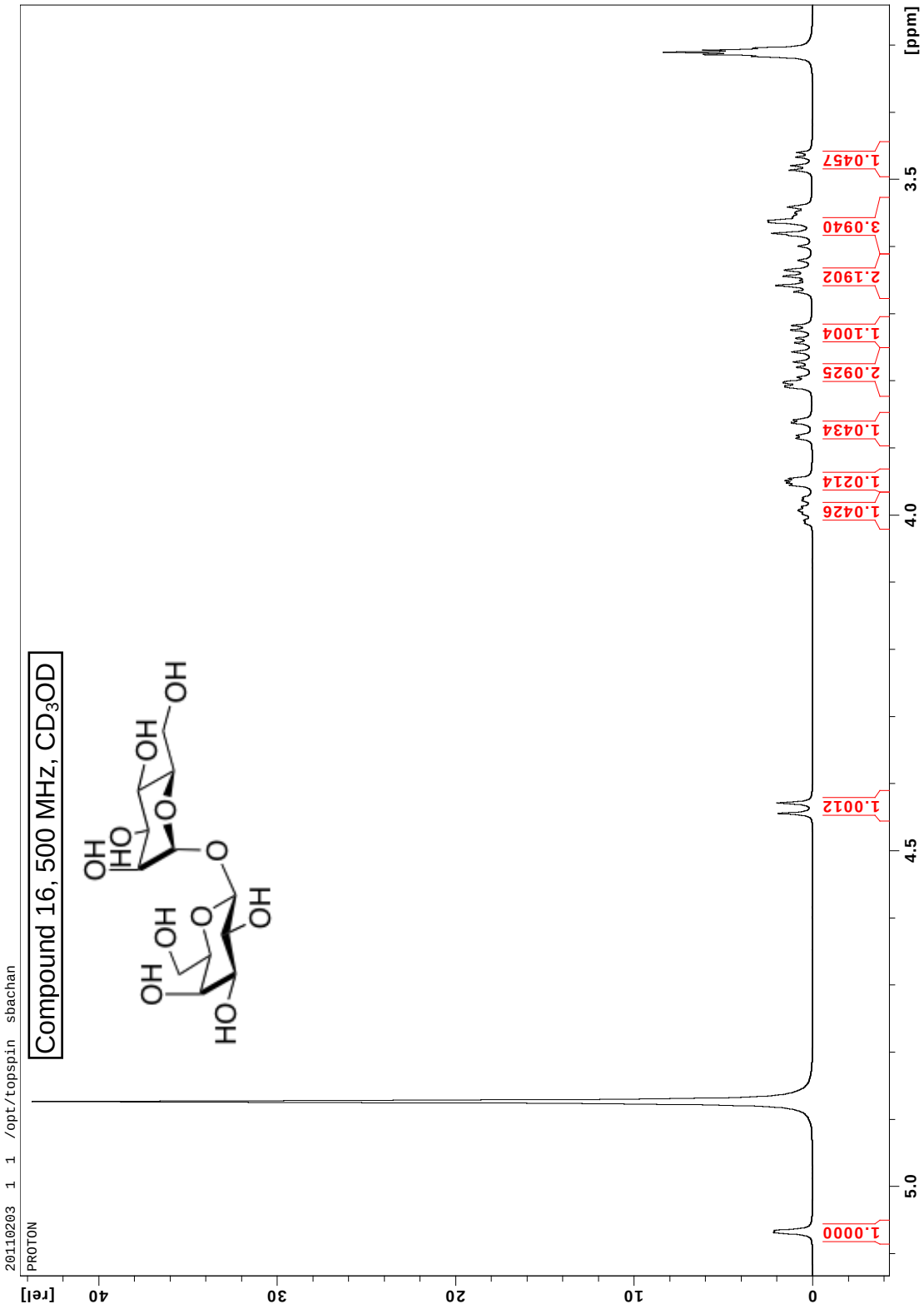

Compound 13, 500 MHz, CD3OD
CARBON
20080528 32 1 /opt/topspin 2008
[rel]
[ppm]
175.9034
103.7080
103.1391
78.3755
78.2015
75.3371
72.8123
72.4013
71.5909
71.3715
68.5537
64.9654
62.7281
35.1036
33.2493
30.9713
30.8228
30.6532
30.4401
26.1923
23.9109
14.6119
OC(O)(CH2)14CH3

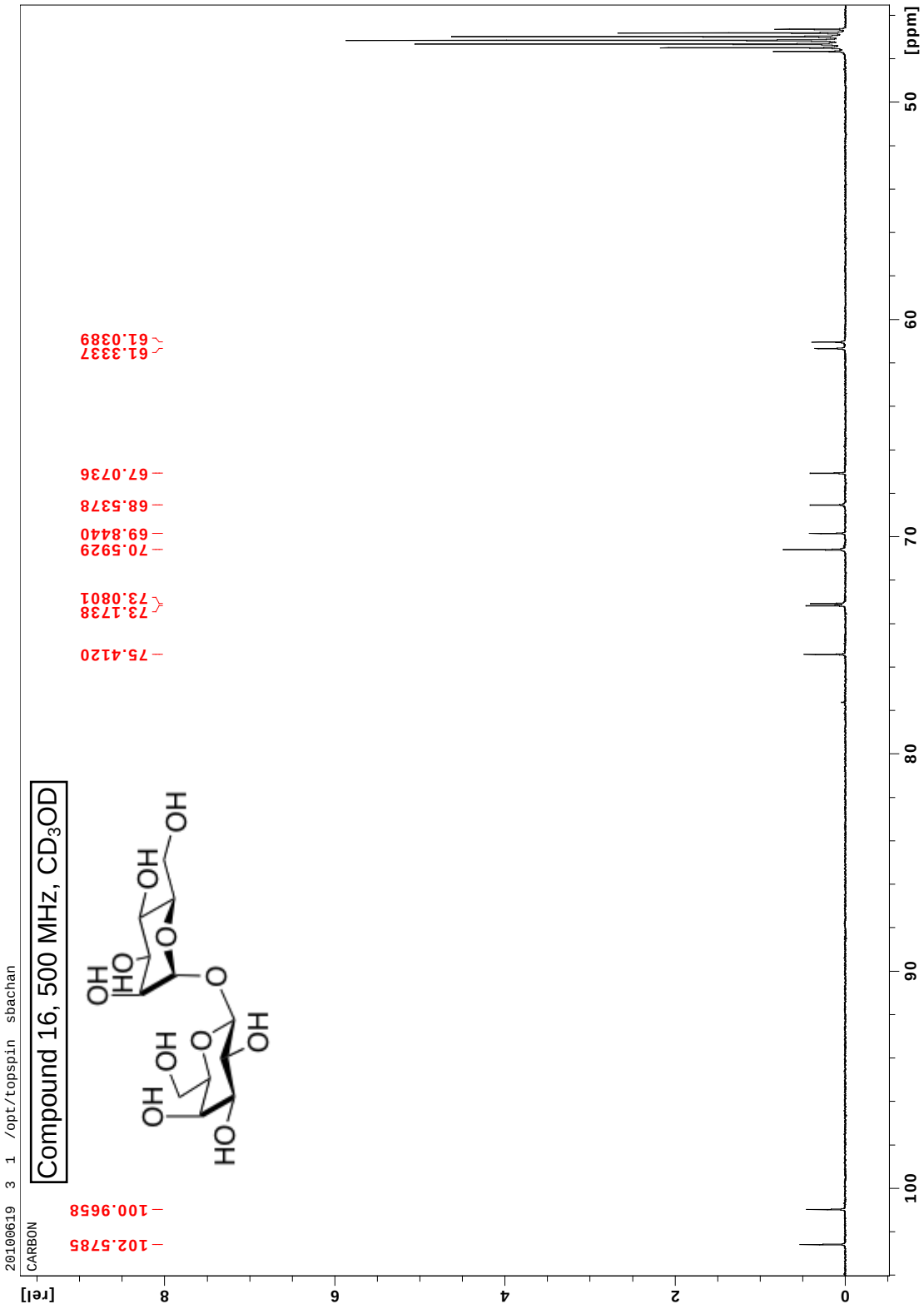

20100118 1 1 /opt/topspin sbachan
PROTON
Compound 14, 500 MHz, CDCl3
OH
HO
OH
O
OC(O)(CH2)14CH3
OCH3
[rel]
40
30
20
10
0
1.0000
1.0404
1.0231
1.0216
1.0494
1.0631
1.0531
3.0825
0.7461
0.8177
2.2423
0.7401
2.2755
27.1459
3.1708
4
3
2
1
[ppm]

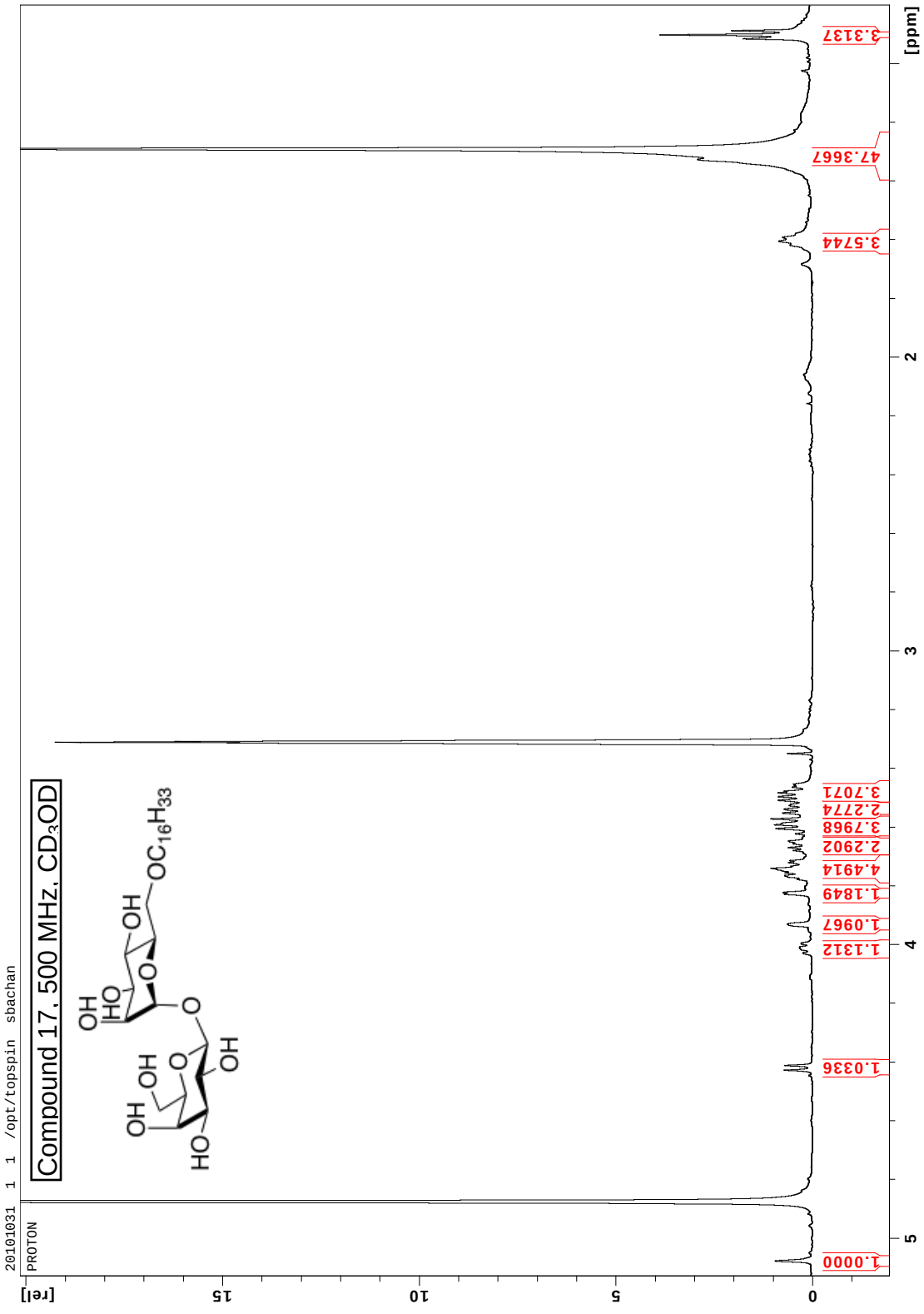

20110319 1 1 /opt/topspin sbachan
CARBON
Compound 14, 500 MHz, CDCl3
OH
HO
OH
O
OC(O)(CH2)14CH3
OCH3
174.9075
101.1009
71.7139
70.7007
70.5822
67.9498
64.0458
55.0648
34.4161
32.1343
29.9211
29.9073
29.8761
29.7369
29.5751
29.5459
29.4027
25.1223
22.8976
14.3200
[rel]
0
10
20
30
160
140
120
100
80
60
40
20 [ppm]

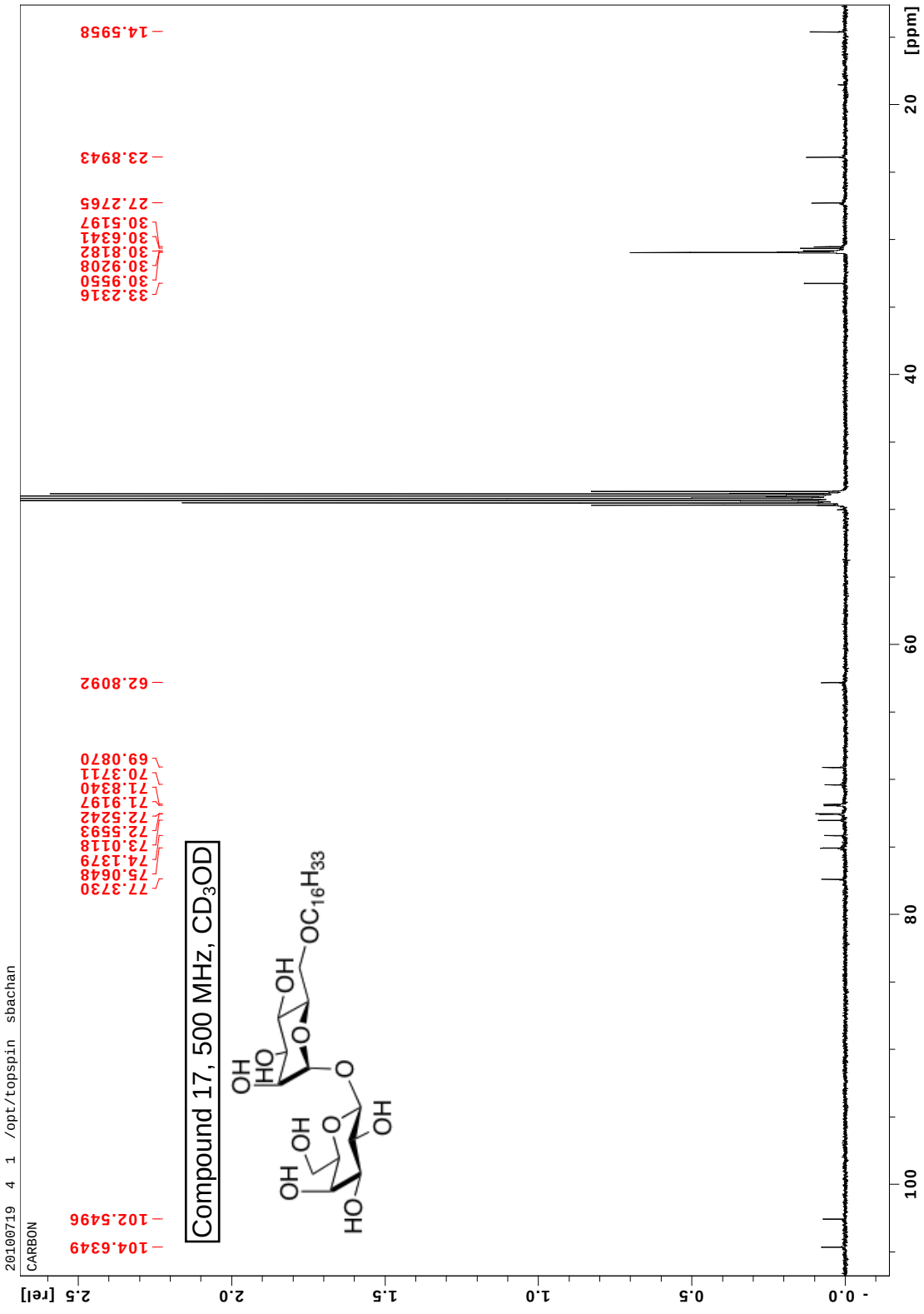

20090821 5 1 /opt/topspin sbachan
PROTON
Compound 15, 500 MHz, CDCl3
OH
HO
O
OC(O)(CH2)14CH3
OC(O)(CH2)14CH3
OCH3
[rel]
30
20
10
0
0.9838
1.0370
1.0020
1.0425
2.0203
3.0664
0.7533
0.7389
4.1322
4.3112
49.8972
6.1548
4
3
2
1
[ppm]

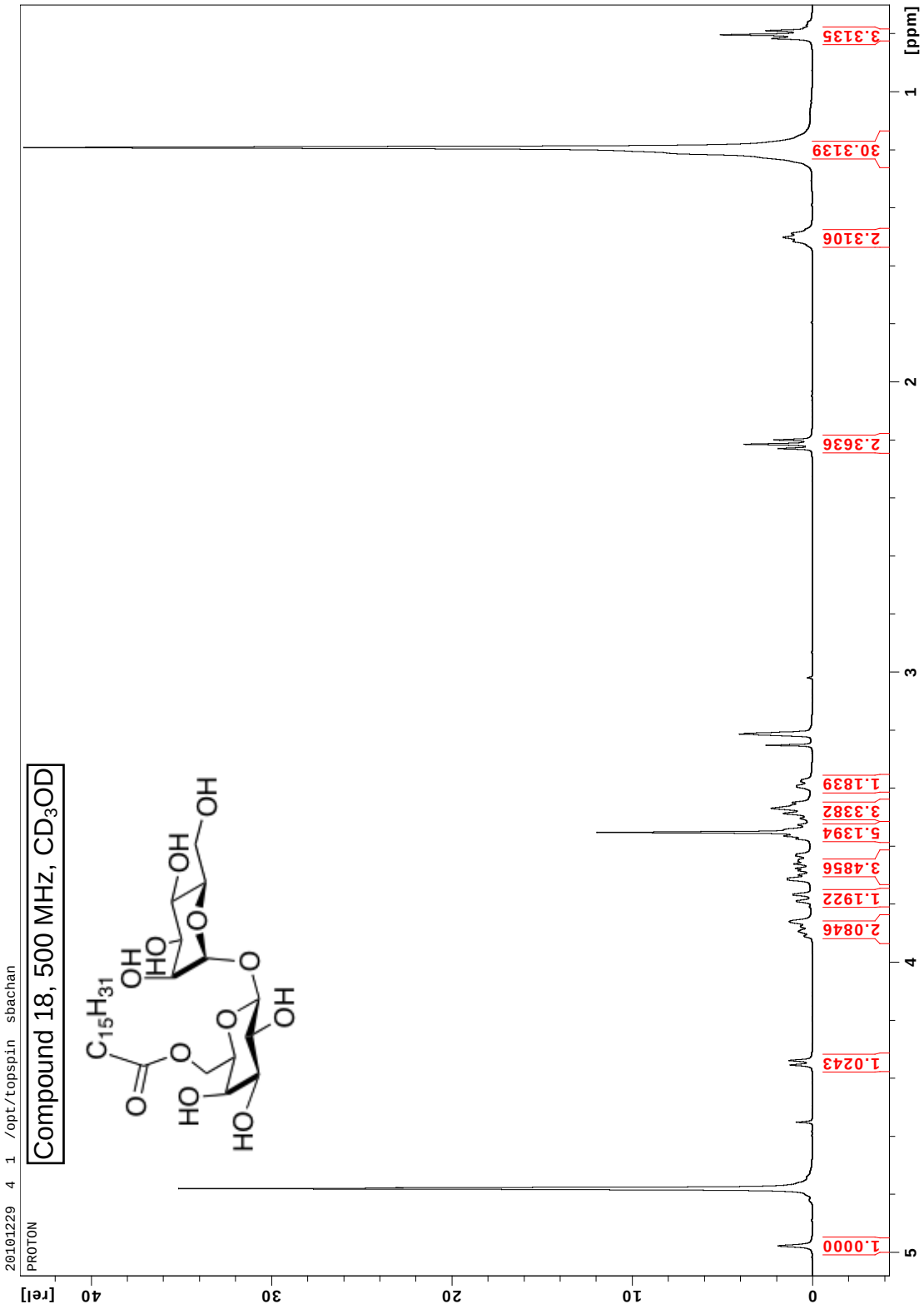

20090821 6 1 /opt/topspin sbachan
CARBON
Compound 15, 500 MHz, CDCl3
OH
HO
O
OC(O)(CH2)14CH3
OC(O)(CH2)14CH3
OCH3
175.0288
173.8295
100.7264
70.7850
70.6478
70.1914
67.9975
62.7051
55.3623
34.5018
34.3008
32.1396
29.9153
29.8767
29.8454
29.8353
29.7027
29.6713
29.5782
29.5109
29.4604
29.3590
29.2873
25.0725
25.0362
22.9040
14.3325
[rel]
40
30
20
10
0
160
140
120
100
80
60
40
[ppm]

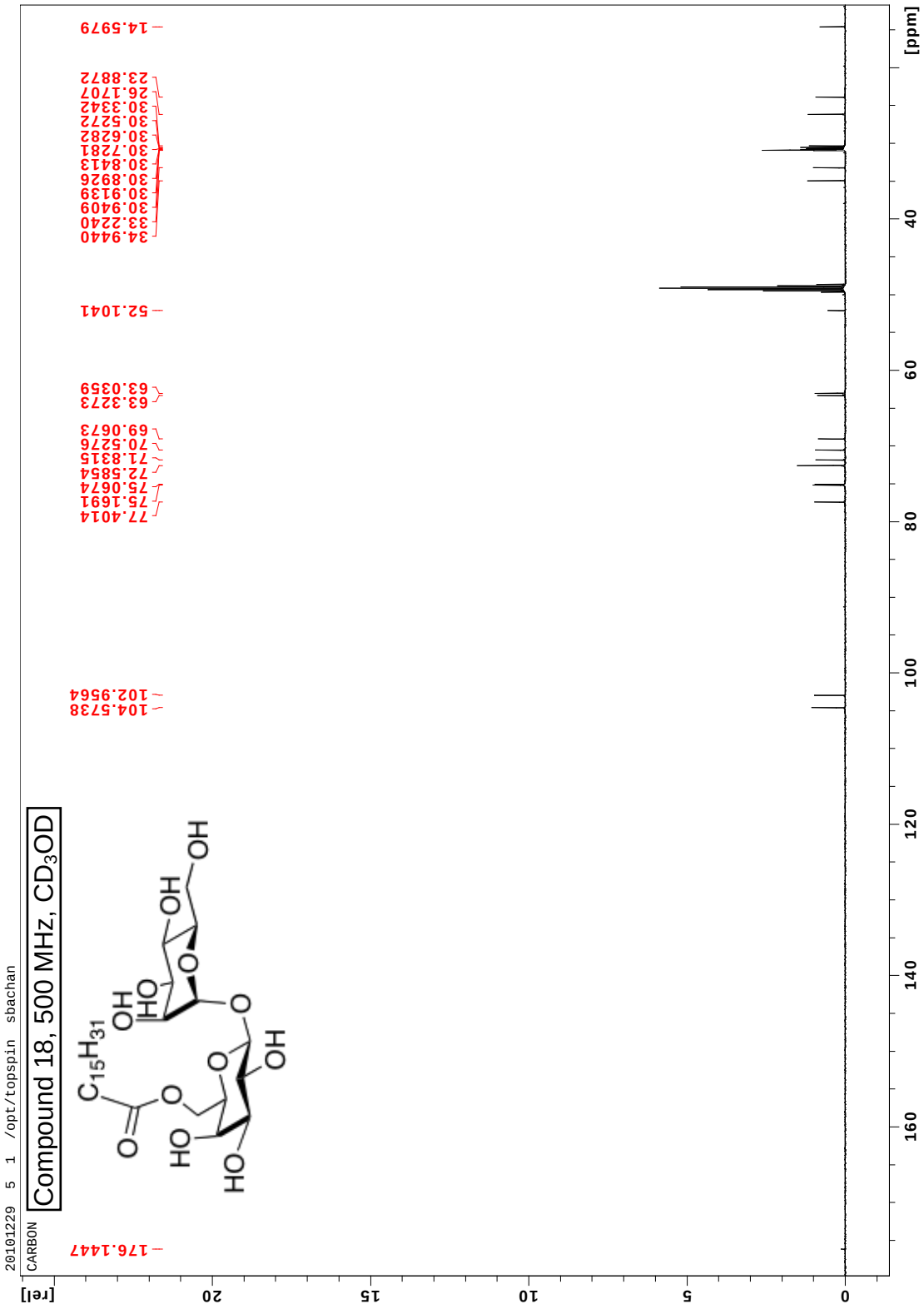

20110203 1 1 /opt/topspin sbachan
PROTON
Compound 16, 500 MHz, CD3OD
[rel]
40
30
20
10
0
5.0
4.5
4.0
3.5
[ppm]
1.0000
1.0012
1.0426
1.0214
1.0434
2.0925
1.1004
2.1902
3.0940
1.0457

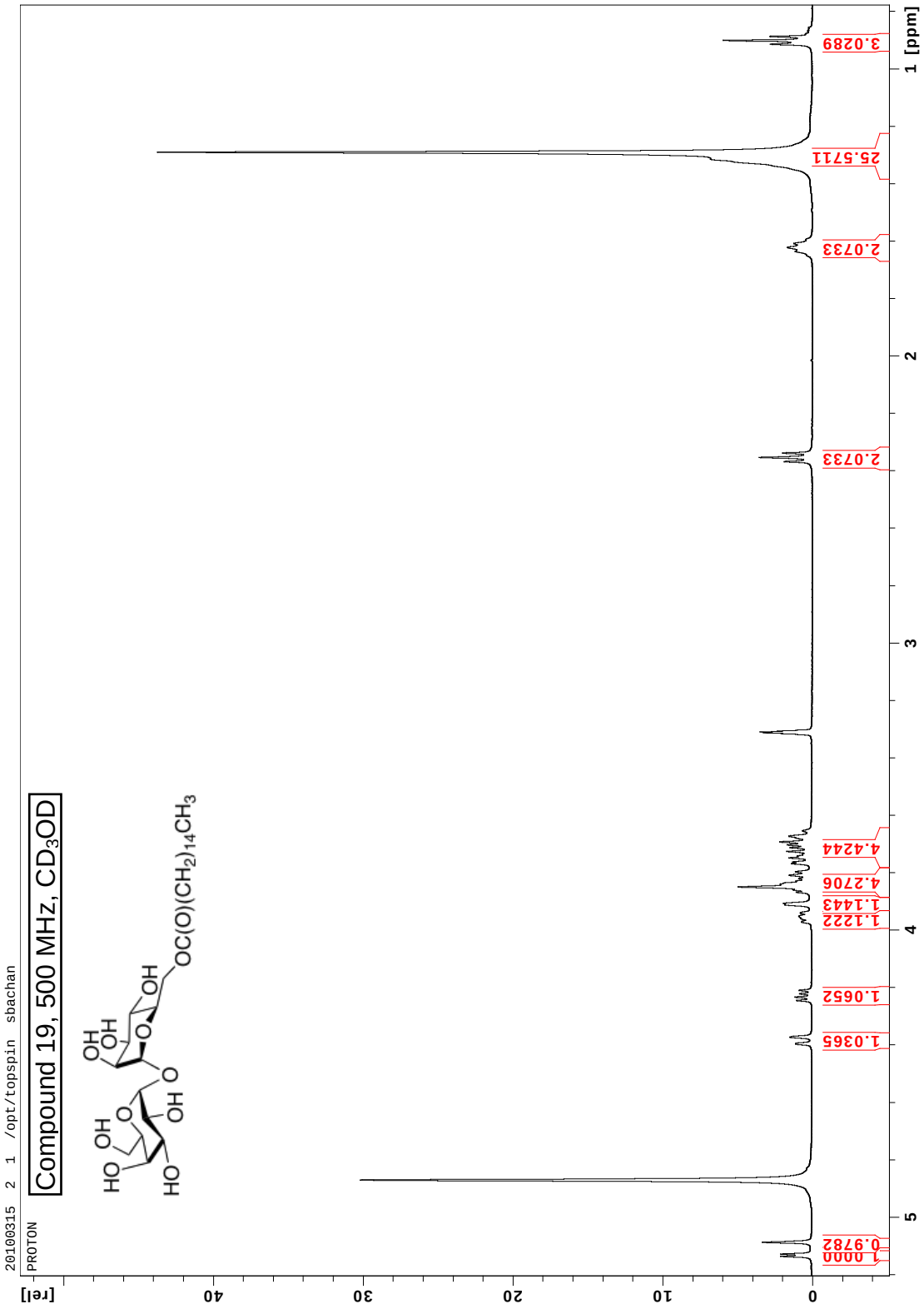

Compound 16, 500 MHz, CD3OD
20100619 3 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
102.5785
100.9658
75.4120
73.1738
73.0801
70.5929
69.8440
68.5378
67.0736
61.3337
61.0389

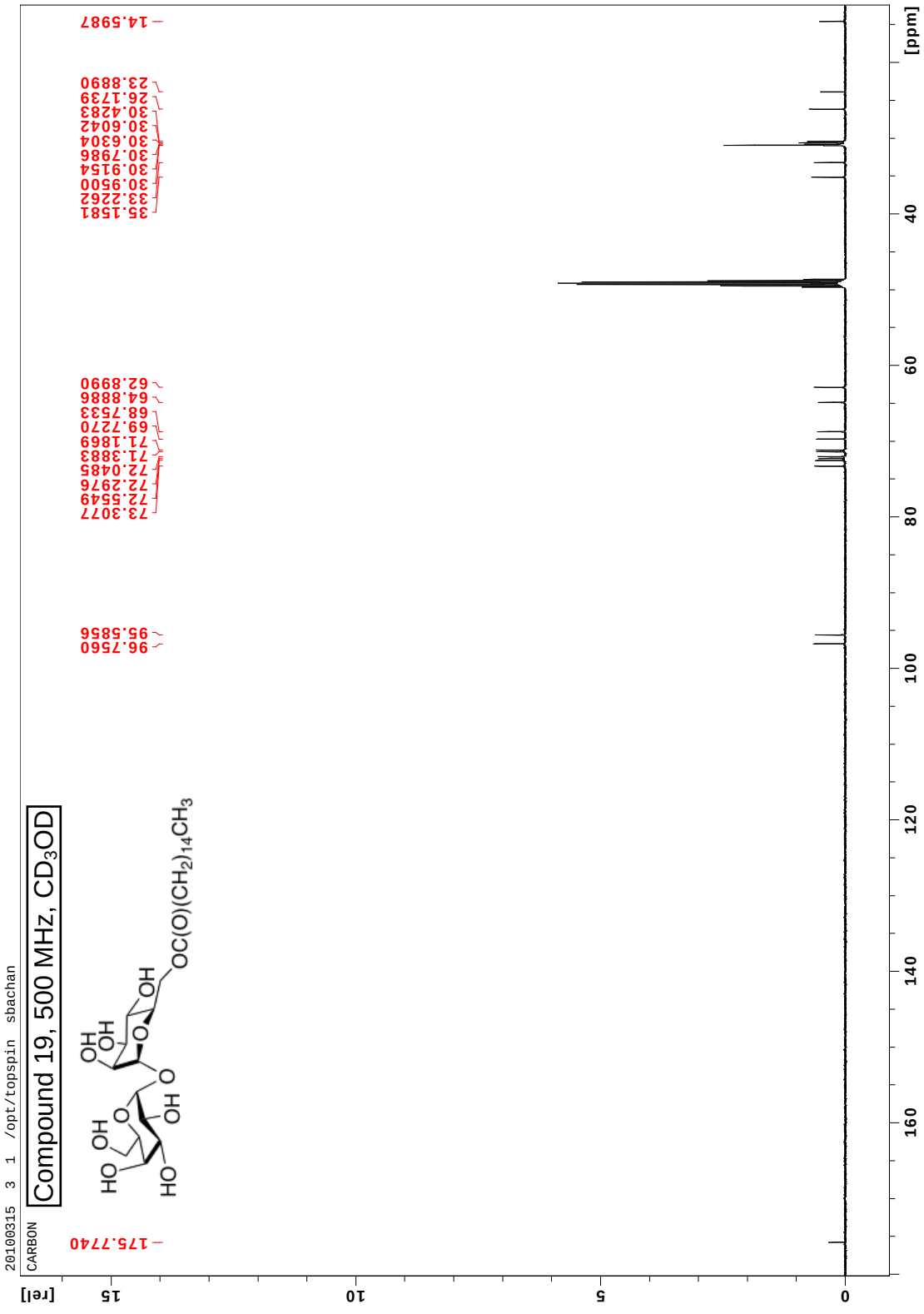

20101031 1 1 /opt/topspin sbachan
PROTON
Compound 17, 500 MHz, CD3OD
[rel]
15
10
5
0
[ppm]
5
4
3
2
1.0000
1.0336
1.1312
1.0967
1.1849
4.4914
2.2902
3.7968
2.2774
3.7071
47.3667
3.5744
3.3137

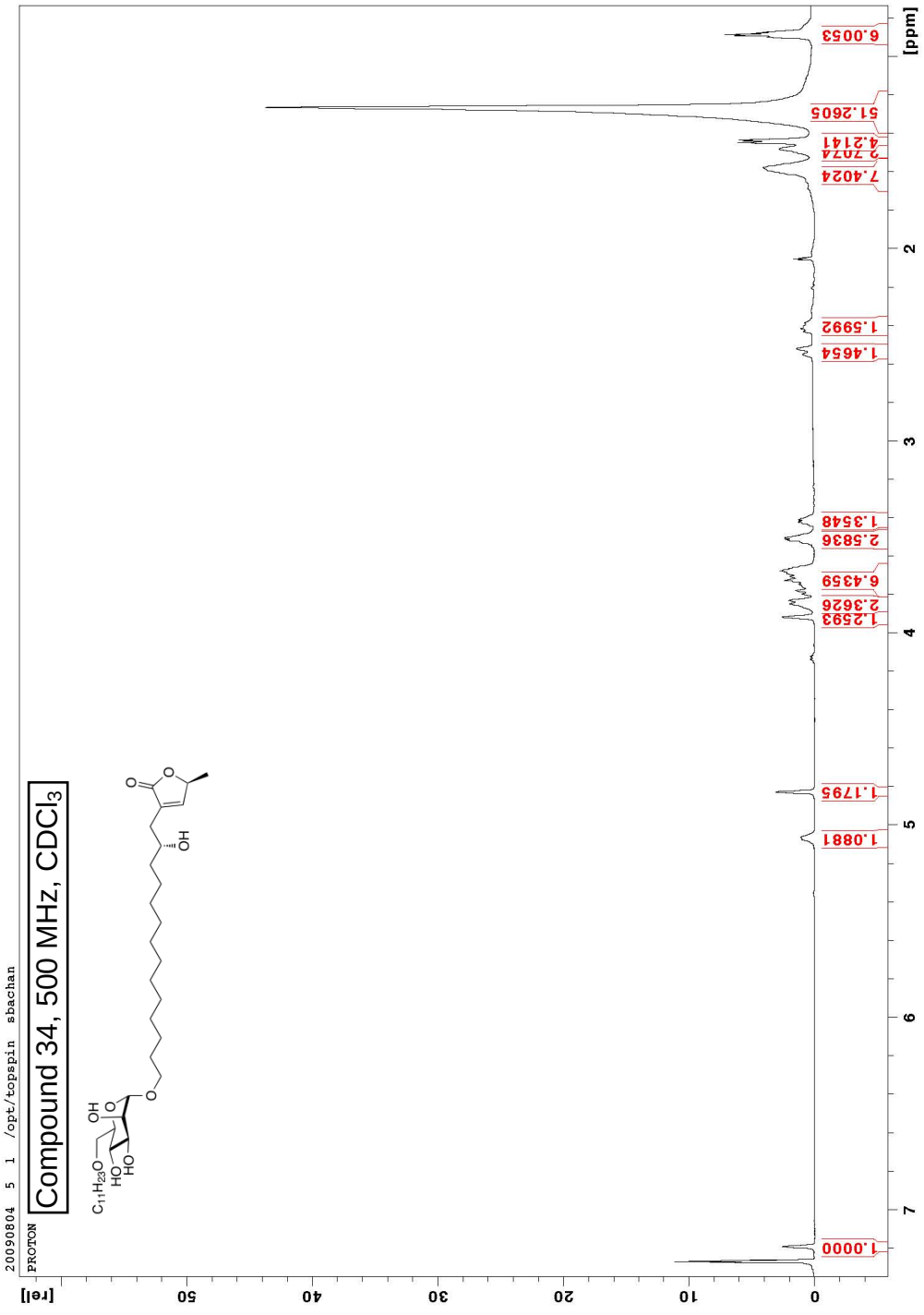

20100719 4 1 /opt/topspin sbachan
CARBON
Compound 17, 500 MHz, CD3OD
104.6349
102.5496
77.3730
75.0648
74.1379
73.0118
72.5593
72.5242
71.9197
71.8340
70.3711
69.0870
62.8092
33.2316
30.9550
30.9208
30.8182
30.6341
30.5197
27.2765
23.8943
14.5958
[rel]
[ppm]

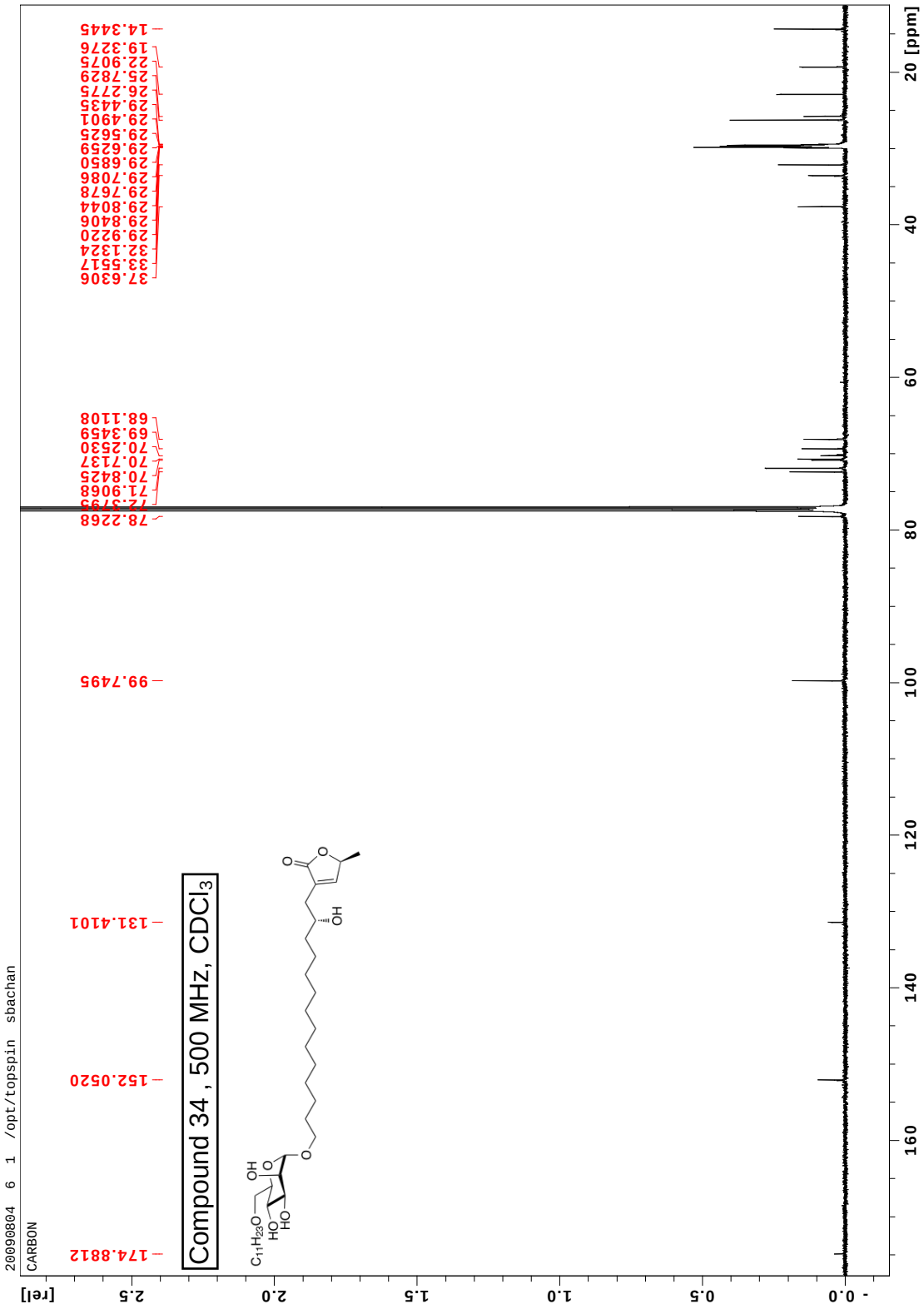

20101229 4 1 /opt/topspin sbachan
PROTON
Compound 18, 500 MHz, CD3OD
C15H31
O
HO
OH
[rel]
40
30
20
10
0
5
4
3
2
1
[ppm]
1.0000
1.0243
2.0846
1.1922
3.4856
5.1394
3.3382
1.1839
2.3636
30.3139
2.3106
3.3135

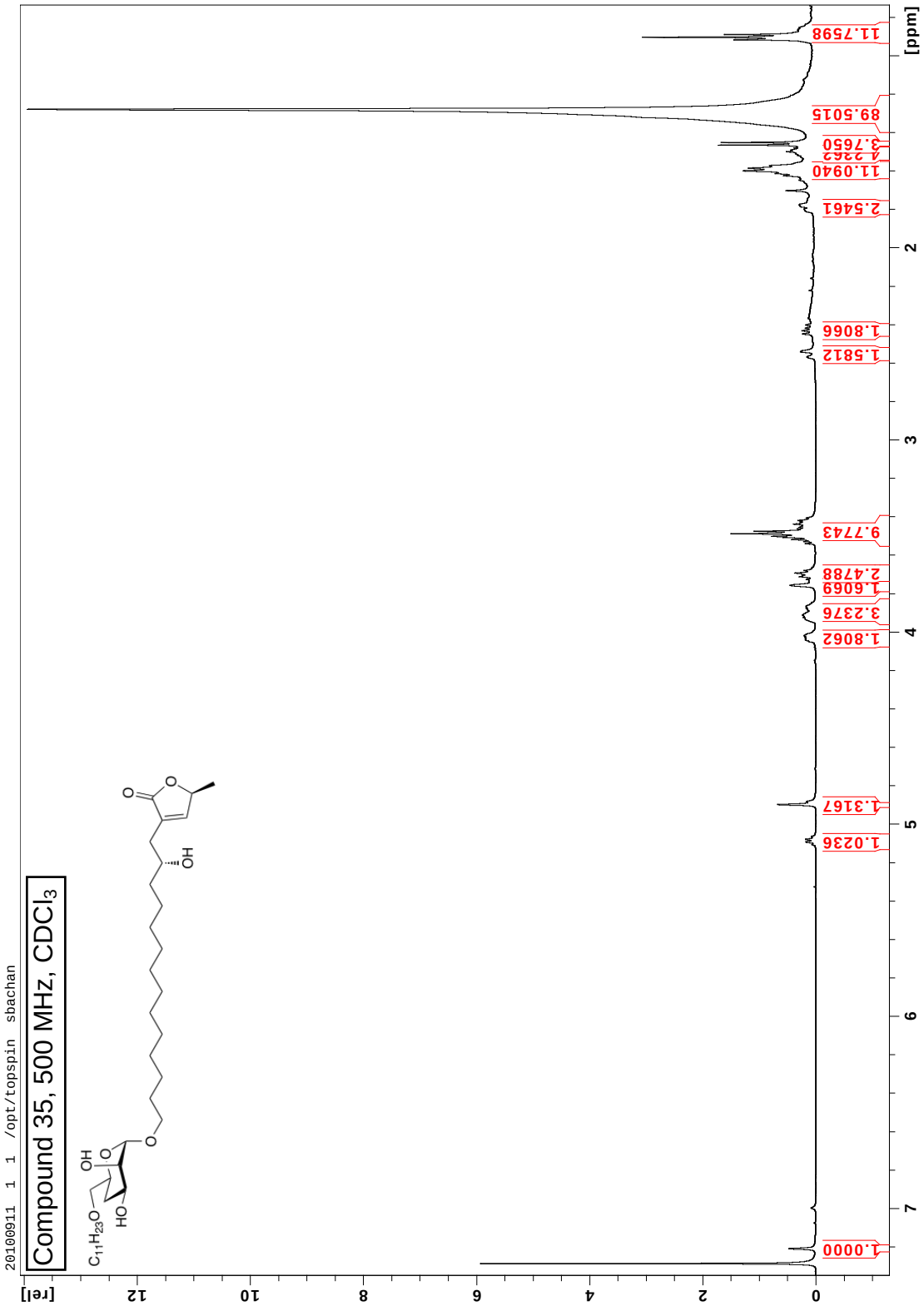

20101229 5 1 /opt/topspin sbachan
CARBON
Compound 18, 500 MHz, CD3OD
C15H31
O
HO
OH
[rel]
[ppm]
176.1447
104.5738
102.9564
77.4014
75.1691
75.0674
72.5854
71.8315
70.5276
69.0673
63.3273
63.0359
52.1041
34.9440
33.2240
30.9409
30.9139
30.8926
30.8413
30.7281
30.6282
30.5272
30.3342
26.1707
23.8872
14.5979

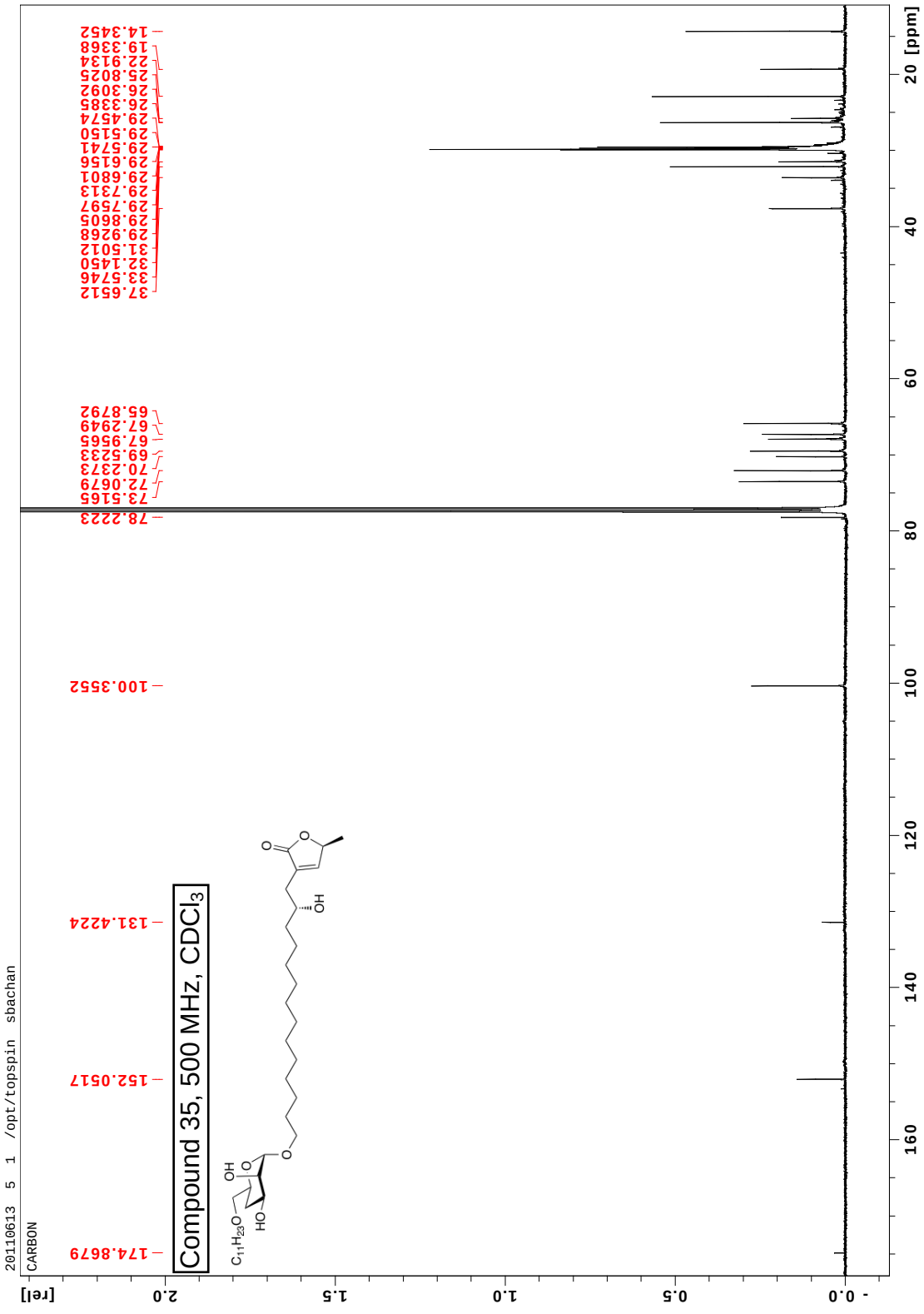

20100315 2 1 /opt/topspin sbachan
PROTON
Compound 19, 500 MHz, CD3OD
OC(O)(CH2)14CH3
[rel]
0
10
20
30
40
1 [ppm]
5
4
3
2
1.0000
0.9782
3.0289
25.5711
2.0733
2.0733
4.4244
4.2706
1.1443
1.1222
1.0652
1.0365

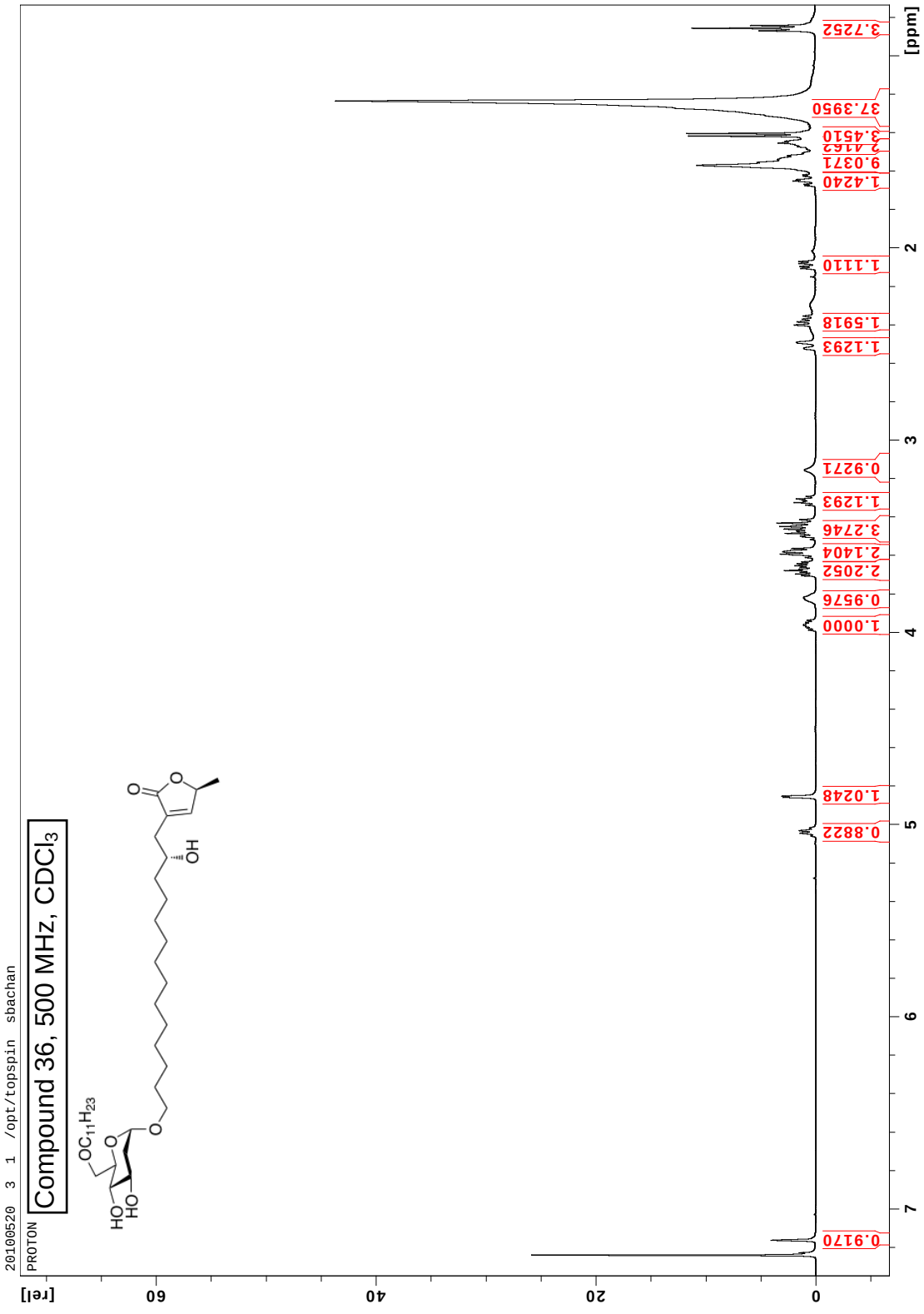

Compound 19, 500 MHz, CD3OD
20100315 3 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
175.7740
96.7560
95.5856
73.3077
72.5549
72.2976
72.0485
71.3883
71.1869
69.7270
68.7533
64.8886
62.8990
35.1581
33.2262
30.9500
30.9154
30.7986
30.6304
30.6042
30.4283
26.1739
23.8890
14.5987
OC(O)(CH2)14CH3

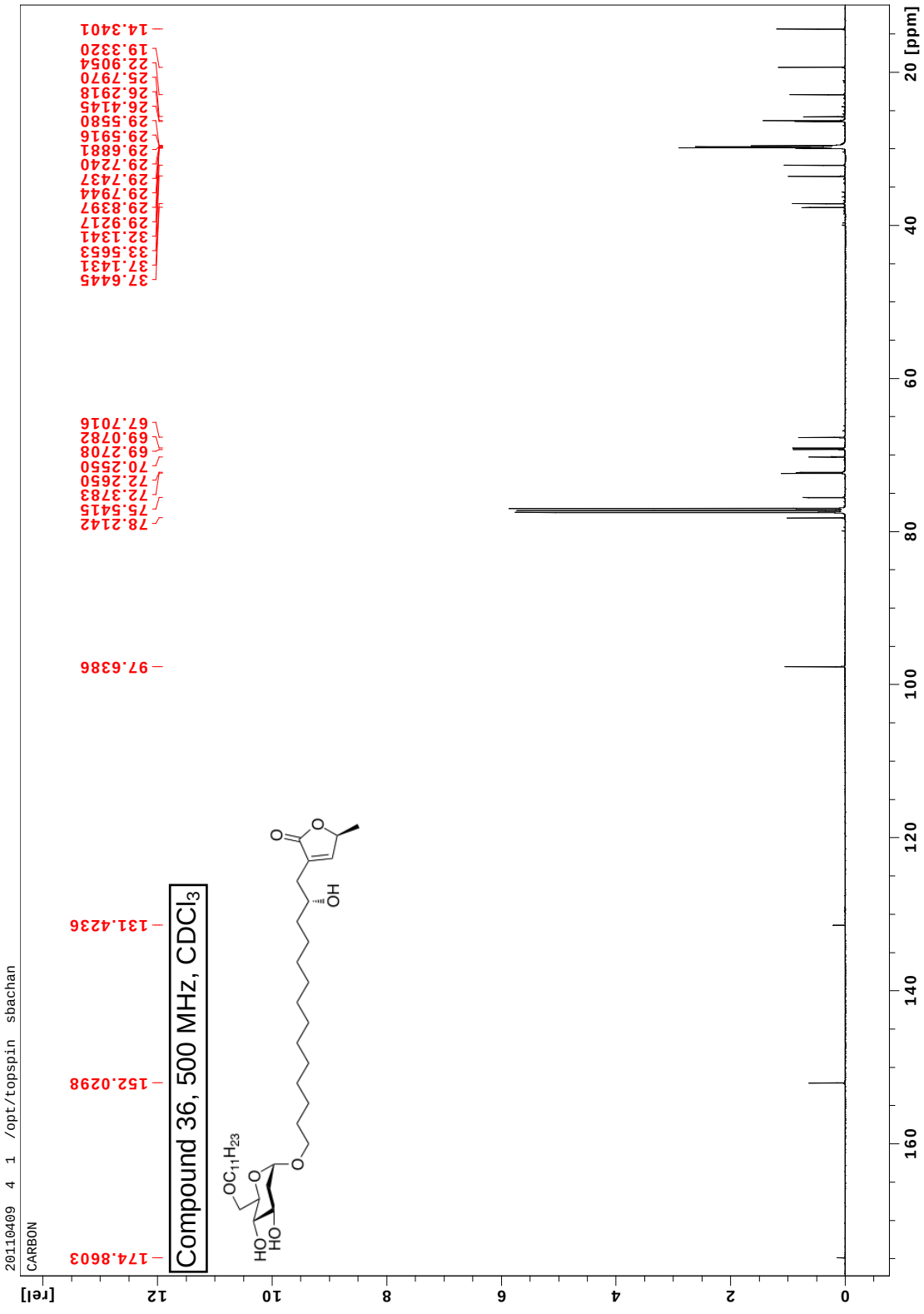

Compound 34, 500 MHz, CDCl3
20090804 5 1 /opt/topspin sbachan
PROTON
[rel]
[ppm]
1.0000
1.0881
1.1795
1.2593
2.3626
6.4359
2.5836
1.3548
1.4654
1.5992
7.4024
2.7074
4.2141
51.2605
6.0053
C11H23O
HO
OH
OH

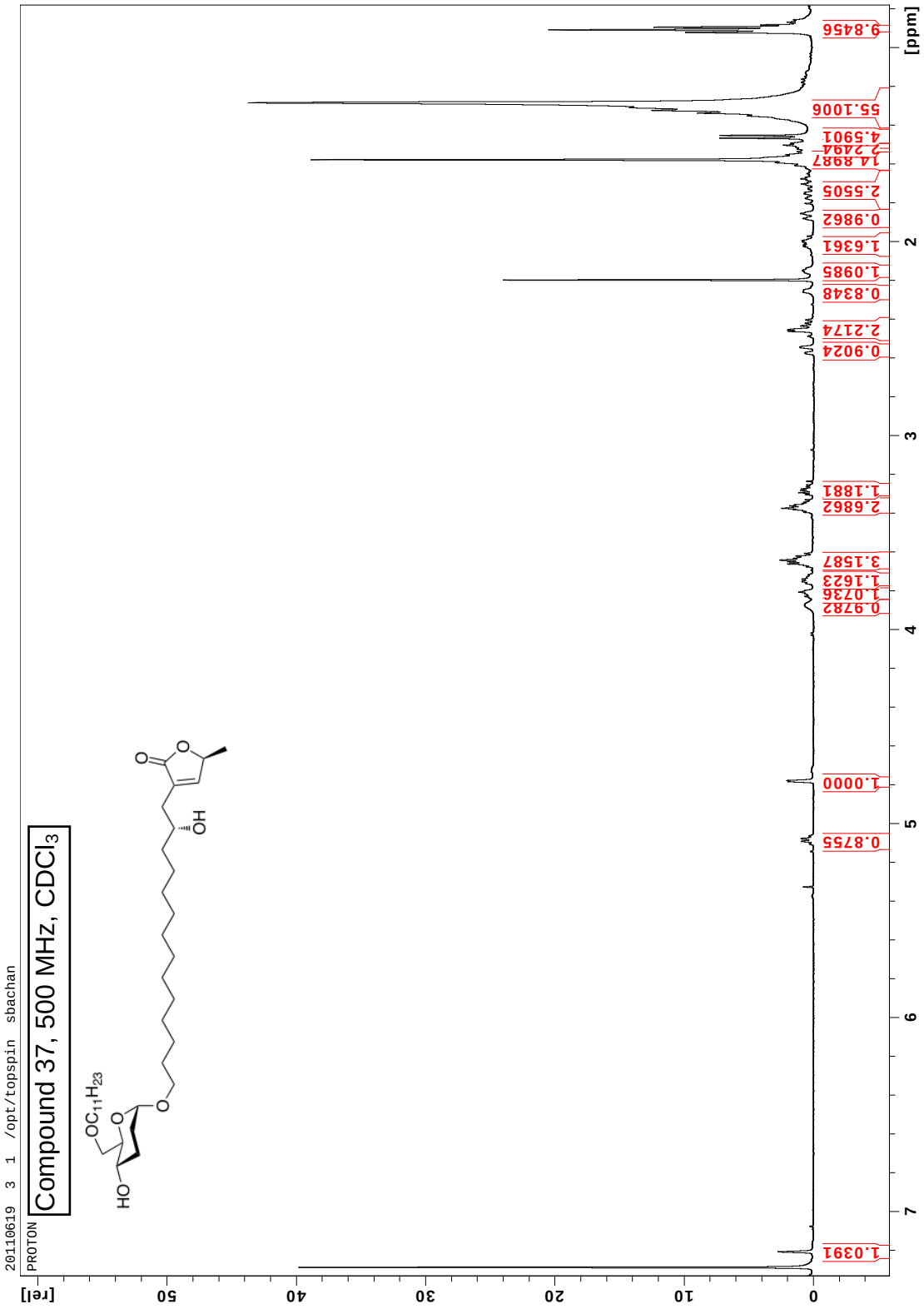

20090804 6 1 /opt/topspin sbachan
CARBON
Compound 34 , 500 MHz, CDCl3
174.8812
152.0520
131.4101
99.7495
78.2268
72.3795
71.9068
70.8425
70.7137
70.2530
69.3459
68.1108
37.6306
33.5517
32.1324
29.9220
29.8406
29.8044
29.7678
29.7086
29.6850
29.6259
29.5625
29.4901
29.4435
26.2775
25.7829
22.9075
19.3276
14.3445
[rel]
20 [ppm]

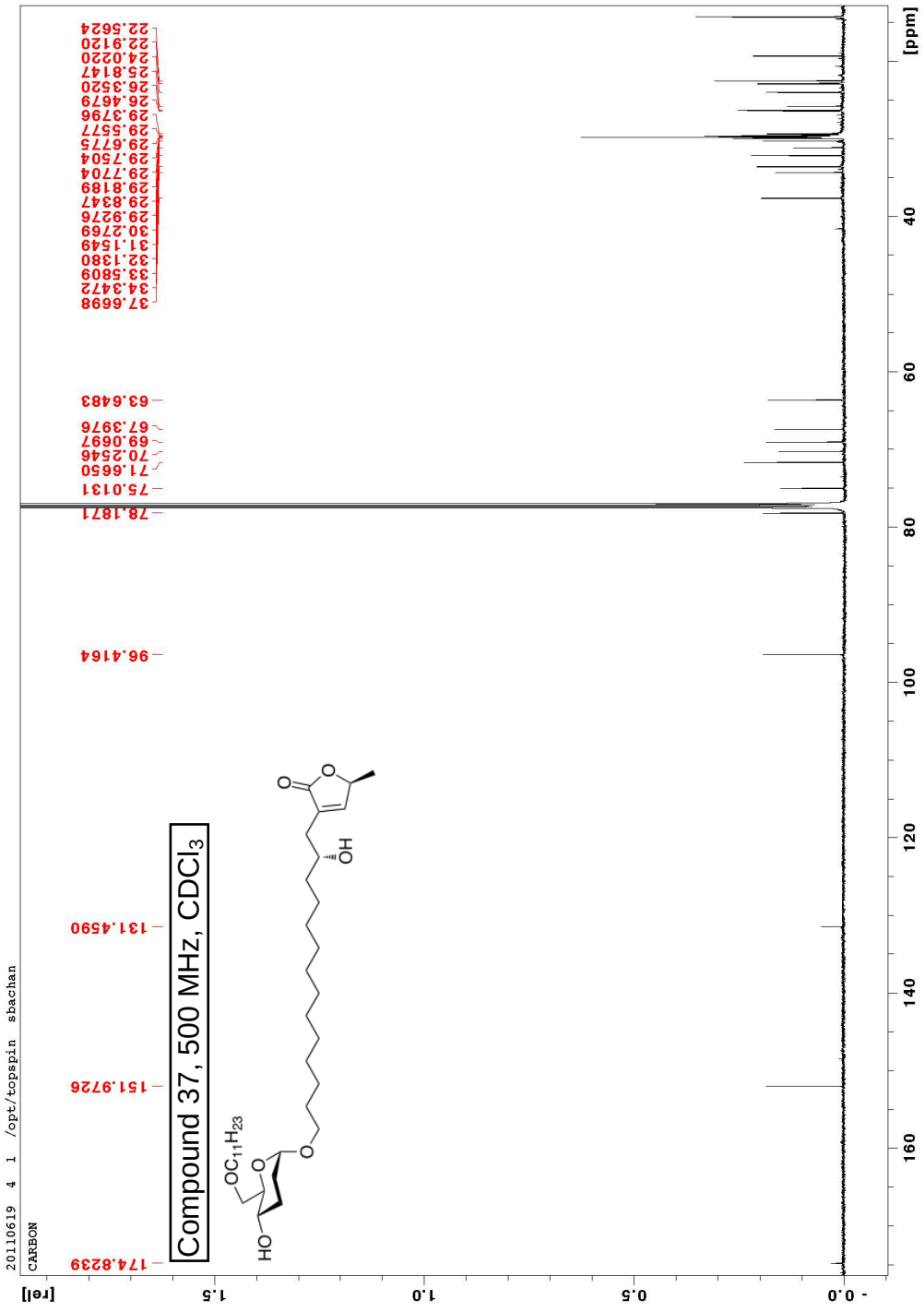

Compound 35, 500 MHz, CDCl3
20100911 1 1 /opt/topspin sbachan
[rel]
[ppm]
1.0000
1.0236
1.3167
1.8062
3.2376
1.6069
2.4788
9.7743
1.8066
1.5812
2.5461
11.0940
4.2262
3.7650
89.5015
11.7598

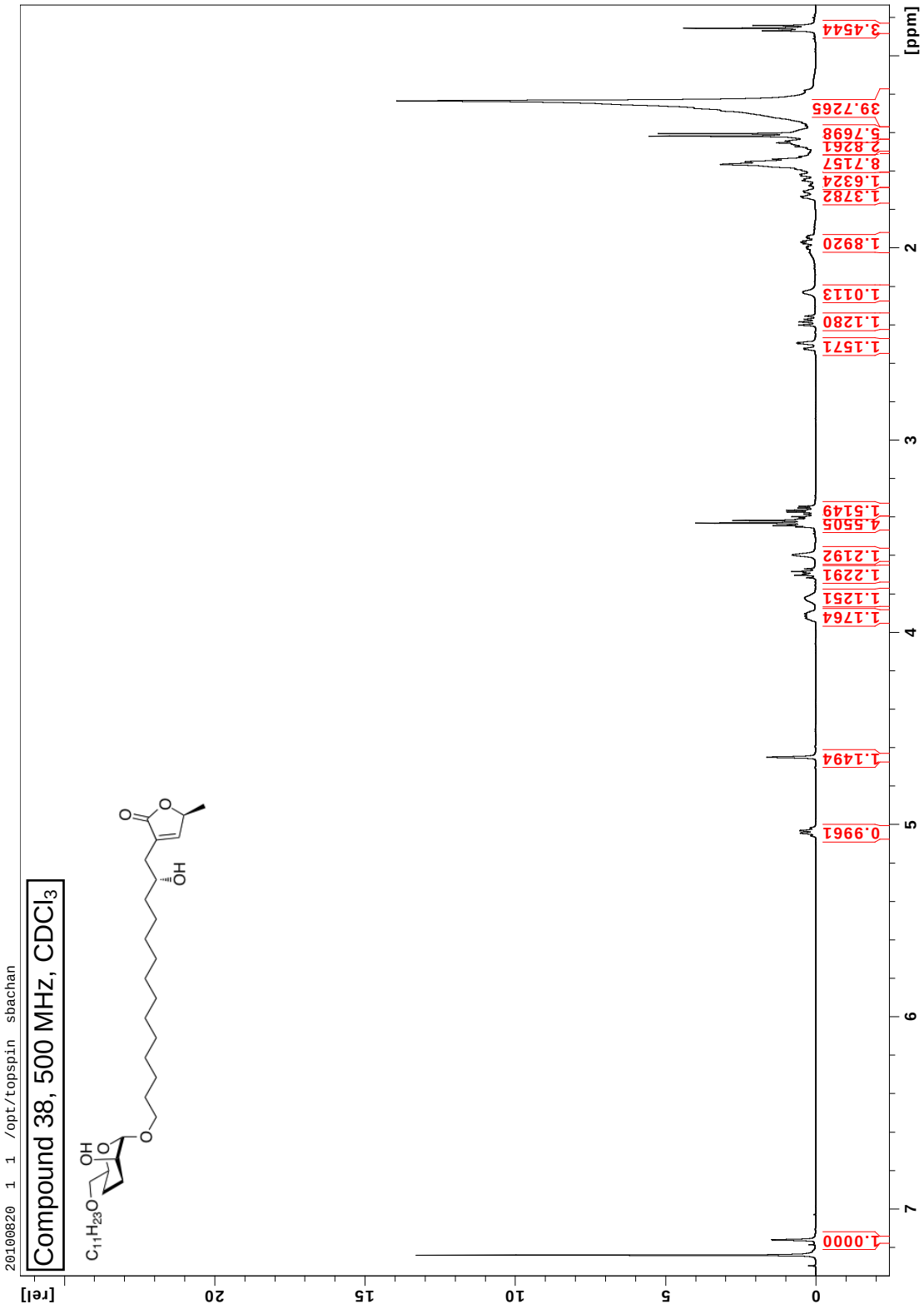

20110613 5 1 /opt/topspin sbachan
CARBON
Compound 35, 500 MHz, CDCl3
174.8679
152.0517
131.4224
100.3552
78.2223
73.5165
72.0679
70.2373
69.5233
67.9565
67.2949
65.8792
37.6512
33.5746
32.1450
31.5012
29.9268
29.8605
29.7597
29.7313
29.6801
29.6156
29.5741
29.5150
29.4574
26.3385
26.3092
25.8025
22.9134
19.3368
14.3452
[rel]
2.0
1.5
1.0
0.5
0.0
20 [ppm]
40
60
80
100
120
140
160

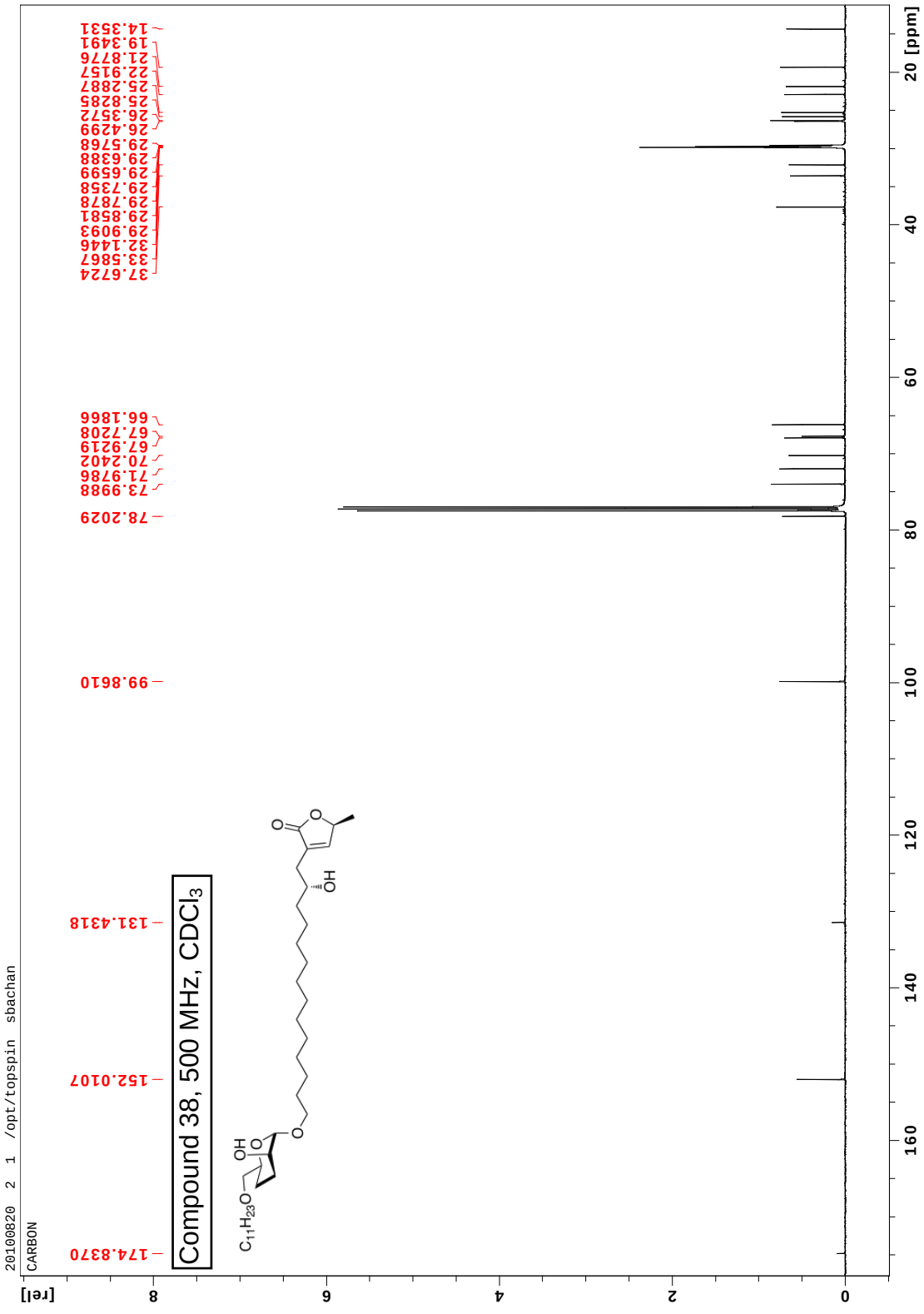

20100520 3 1 /opt/topspin sbachan
PROTON
Compound 36, 500 MHz, CDCl3
[rel]
[ppm]
0.9170
0.8822
1.0248
1.0000
0.9576
2.2052
2.1404
3.2746
1.1293
0.9271
1.1293
1.5918
1.1110
1.4240
9.0371
2.4162
3.4510
37.3950
3.7252

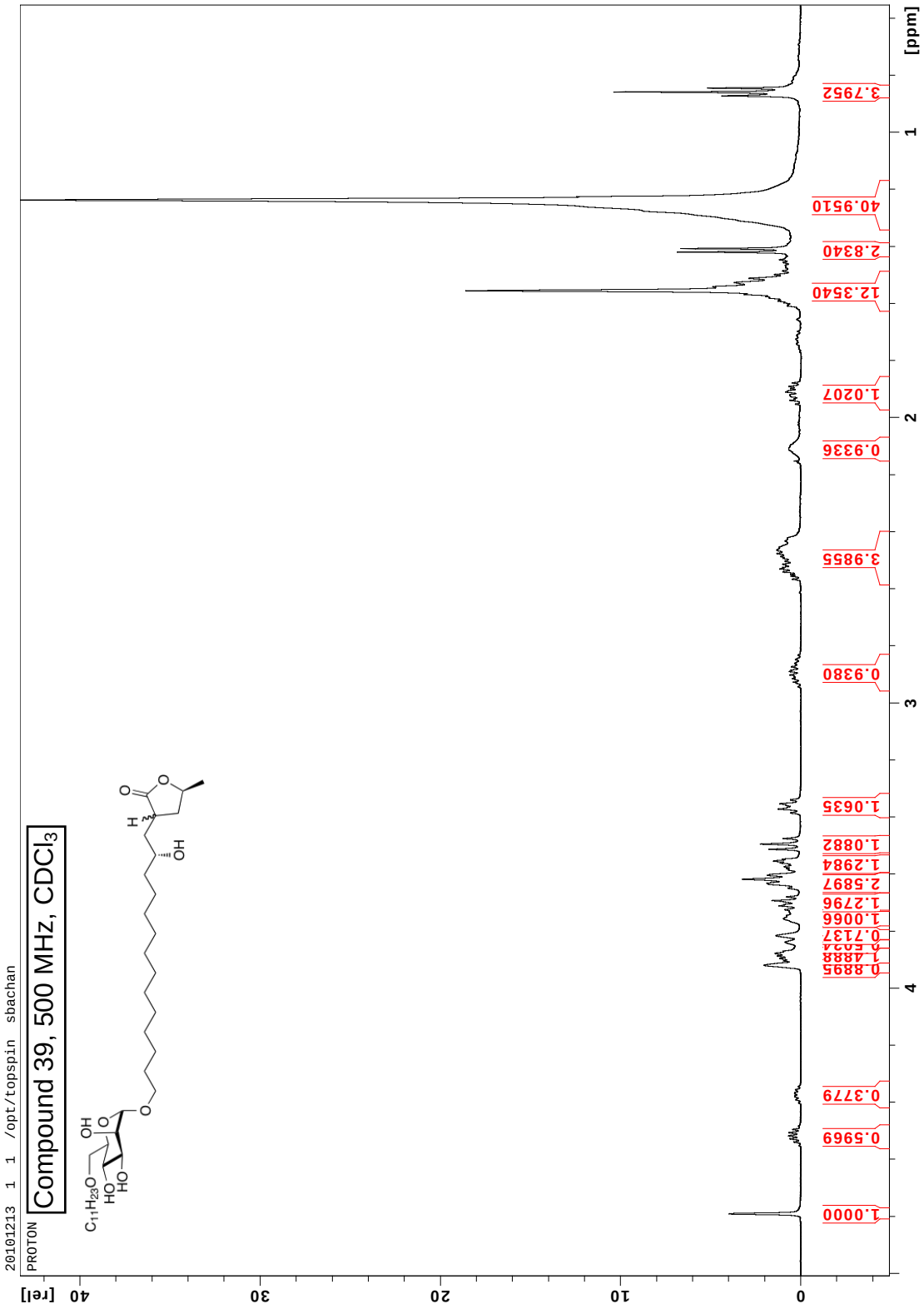

20110409 4 1 /opt/topspin sbachan
CARBON
Compound 36, 500 MHz, CDCl3
174.8603
152.0298
131.4236
97.6386
78.2142
75.5415
72.3783
72.2650
70.2550
69.2708
69.0782
67.7016
37.6445
37.1431
33.5653
32.1341
29.9217
29.8397
29.7944
29.7437
29.7240
29.6881
29.5916
29.5580
26.4145
26.2918
25.7970
22.9054
19.3320
14.3401
OH
HO
HO
OC11H23
[rel]
20 [ppm]

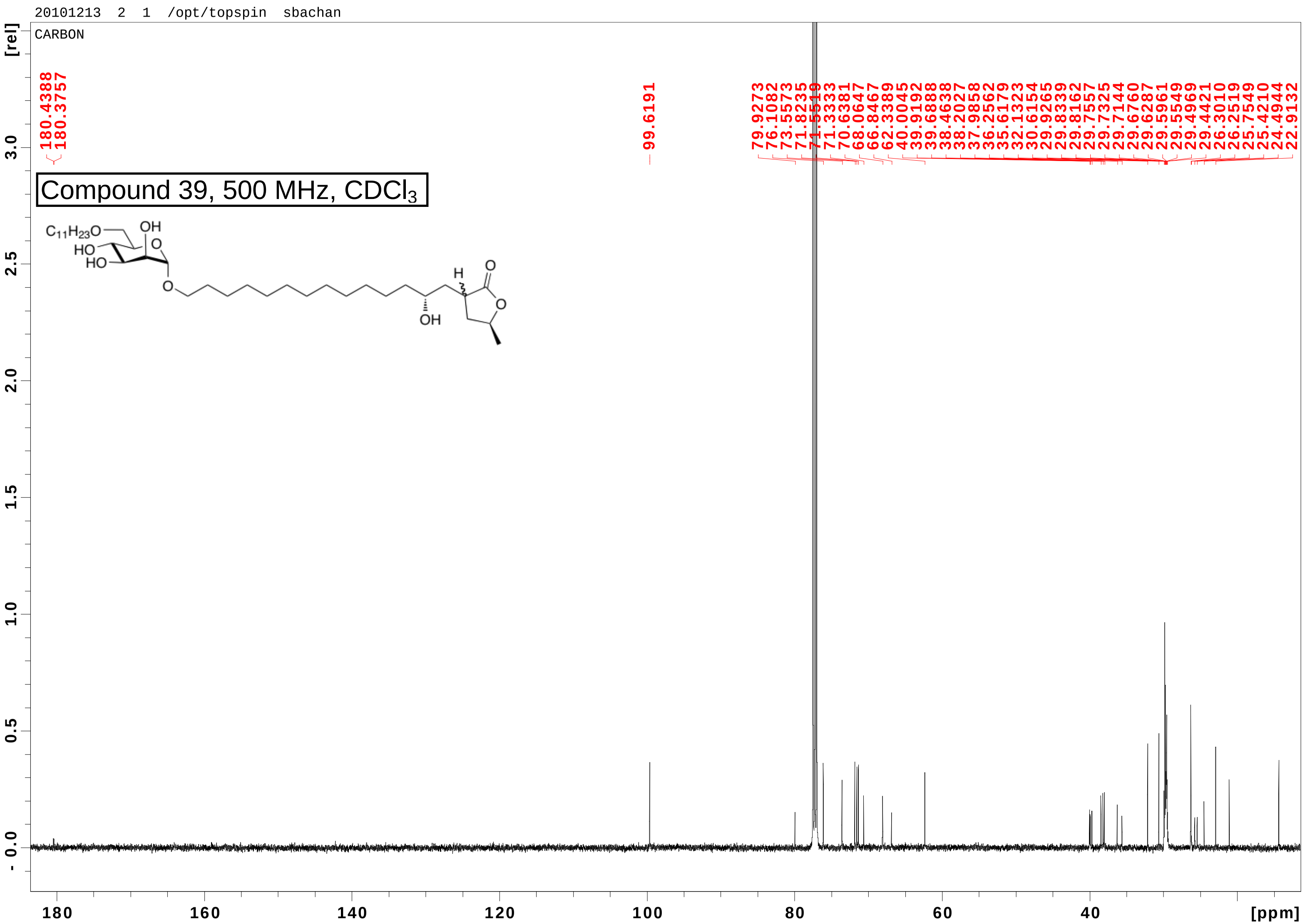

20110619 3 1 /opt/topspin sbachan
PROTON
Compound 37, 500 MHz, CDCl3
[rel]
[ppm]
1.0391
0.8755
1.0000
0.9782
1.0736
1.1623
3.1587
2.6862
1.1881
0.9024
2.2174
0.8348
1.0985
1.6361
0.9862
2.5505
14.8987
2.2494
4.5901
55.1006
9.8456

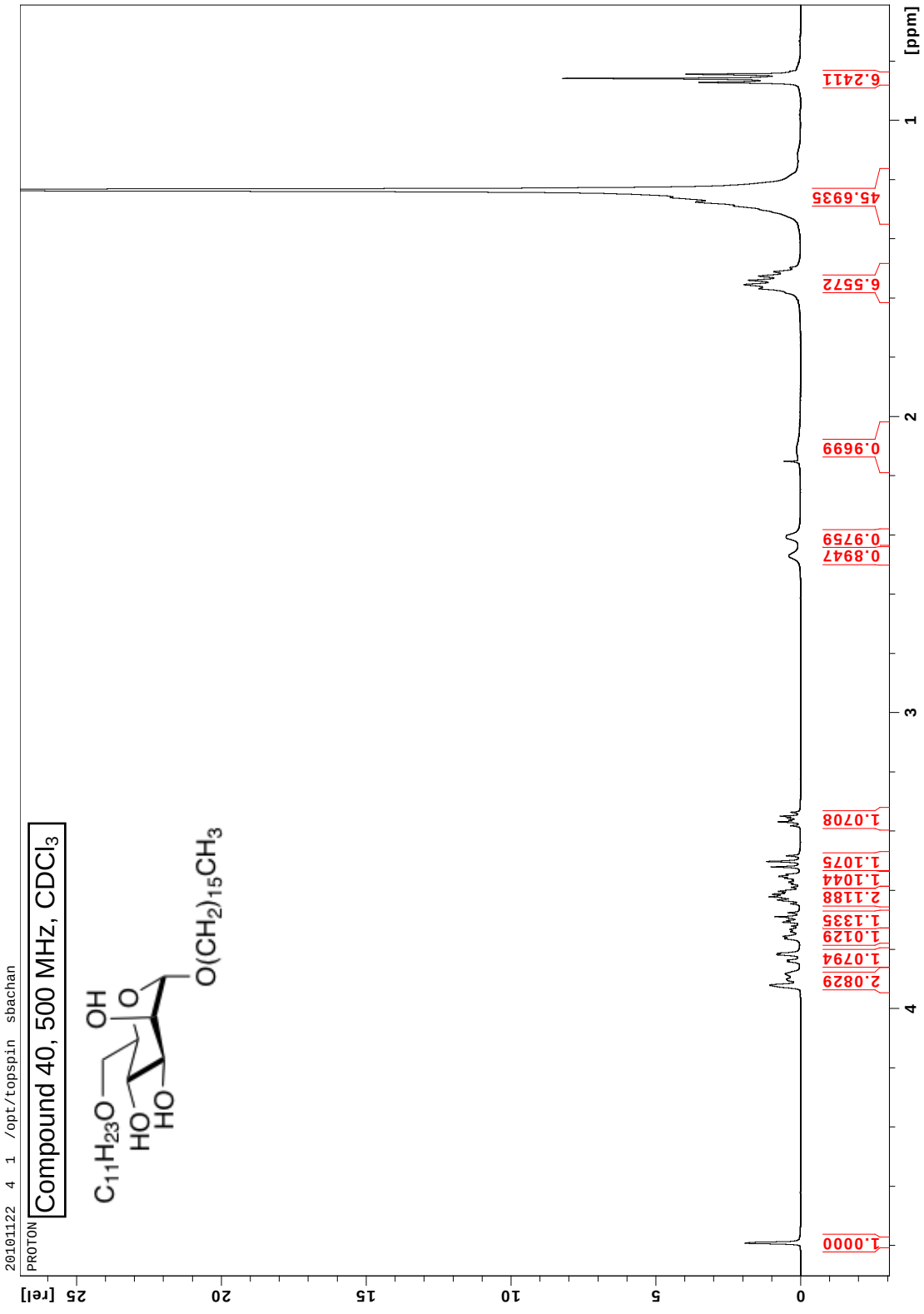

Compound 37, 500 MHz, CDCl3
20110619 4 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
174.8239
151.9726
131.4590
96.4164
78.1871
75.0131
71.6650
70.2546
69.0697
67.3976
63.6483
37.6698
34.3472
33.5809
32.1380
31.1549
30.2769
29.9276
29.8347
29.8189
29.7704
29.7504
29.6775
29.5577
29.3796
26.4679
26.3520
25.8147
24.0220
22.9120
22.5624
HO
OC11H23
O
HO

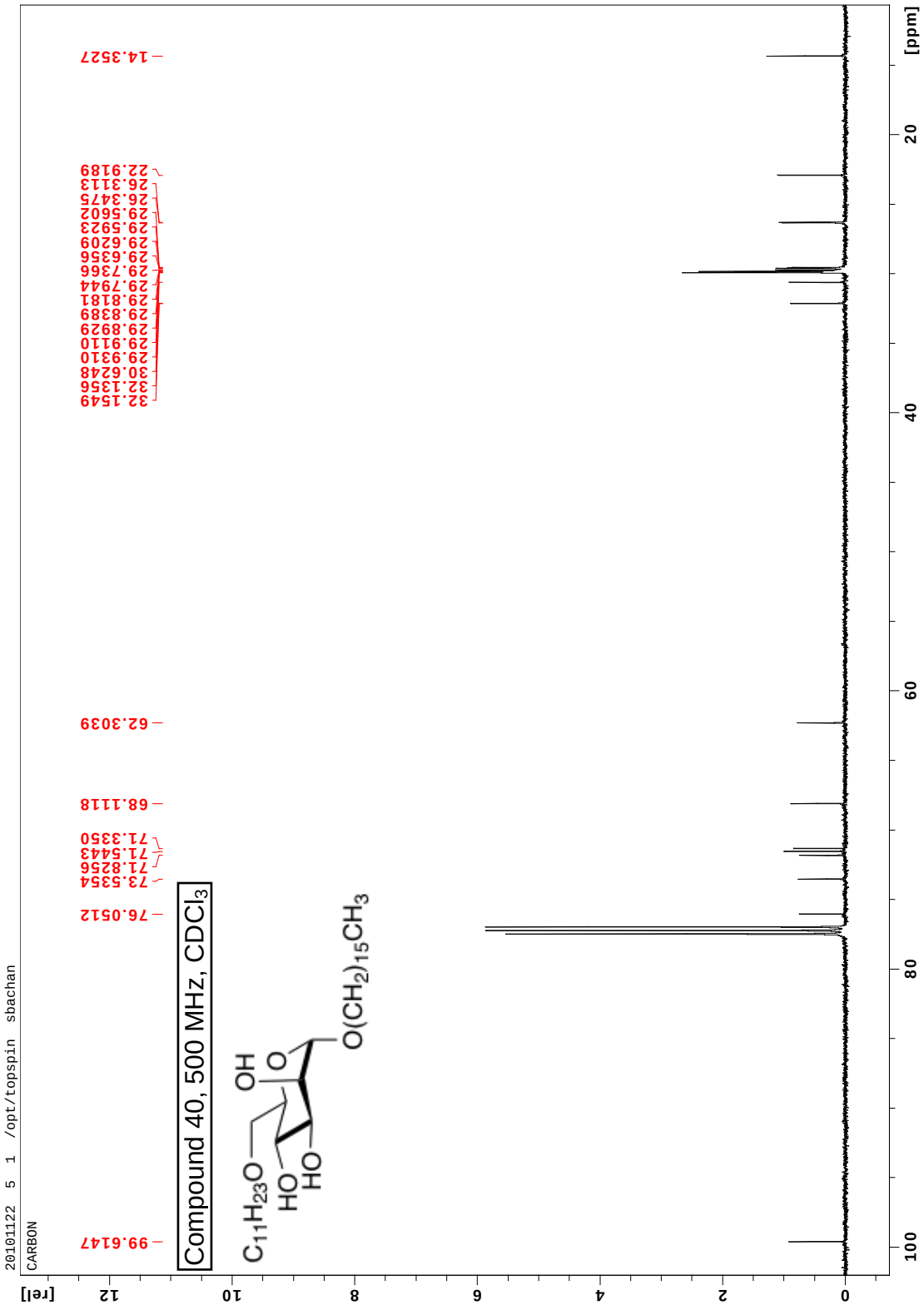

Compound 38, 500 MHz, CDCl3
20100820 1 1 /opt/topspin sbachan
[rel]
[ppm]
1.0000
0.9961
1.1494
1.1764
1.1251
1.2291
1.2192
4.5505
1.5149
1.1571
1.1280
1.0113
1.8920
1.3782
1.6324
8.7157
2.8261
5.7698
39.7265
3.4544

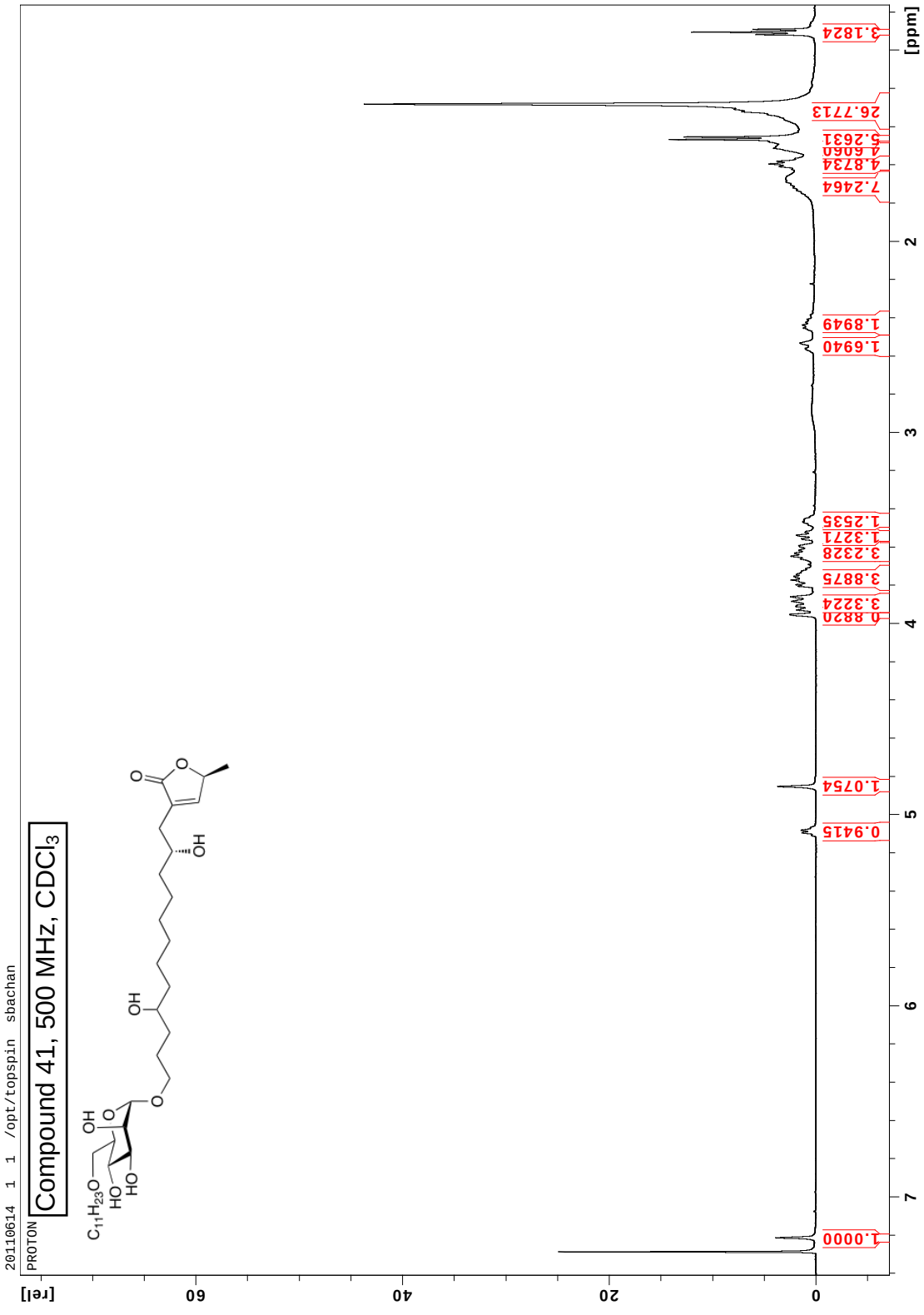

20100820 2 1 /opt/topspin sbachan
CARBON
Compound 38, 500 MHz, CDCl3
C11H23
OH
OH
174.8370
152.0107
131.4318
99.8610
78.2029
73.9988
71.9786
70.2402
67.9219
67.7208
66.1866
37.6724
33.5867
32.1446
29.9093
29.8581
29.7878
29.7358
29.6599
29.6388
29.5768
26.4299
26.3572
25.8285
25.2887
22.9157
21.8776
19.3491
14.3531
[rel]
8
6
4
2
0
20 [ppm]
40
60
80
100
120
140
160

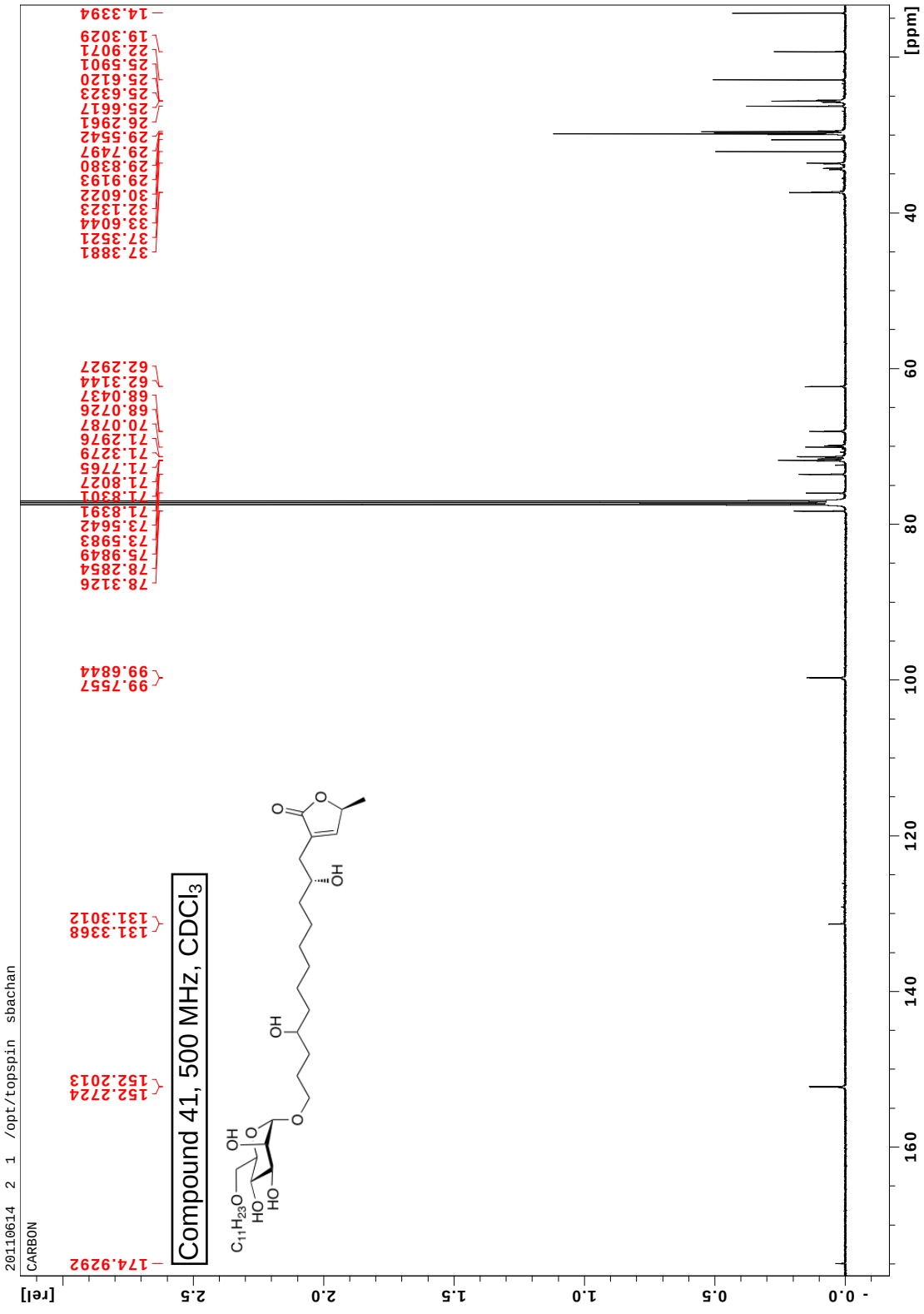

20101213 1 1 /opt/topspin sbachan
PROTON
Compound 39, 500 MHz, CDCl3
[rel]
40
30
20
10
0
[ppm]
1
2
3
4
1.0000
0.5969
0.3779
0.8895
1.4888
0.7137
1.0066
1.2796
2.5897
1.2984
1.0882
1.0635
0.9380
3.9855
0.9336
1.0207
12.3540
2.8340
40.9510
3.7952

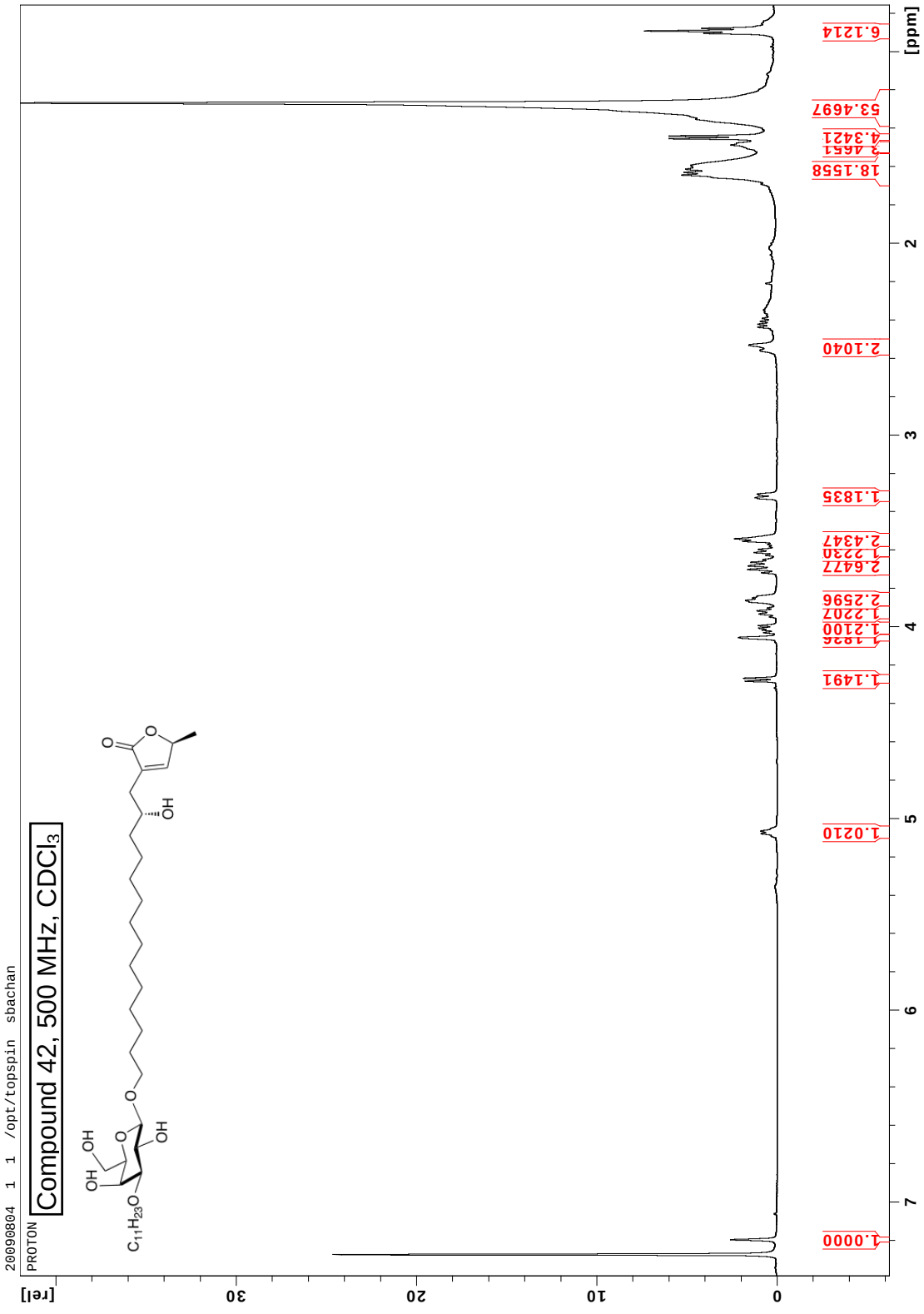

20101213 2 1 /opt/topspin sbachan
CARBON
180.4388
180.3757
99.6191
79.9273
76.1082
73.5573
71.8235
71.5519
71.3333
70.6381
68.0647
66.8467
62.3389
40.0045
39.9192
39.6888
38.4638
38.2027
37.9858
36.2562
35.6179
32.1323
30.6154
29.9265
29.8339
29.8162
29.7557
29.7325
29.7144
29.6760
29.6287
29.5961
29.5549
29.4969
29.4421
26.3010
26.2519
25.7549
25.4210
24.4944
22.9132
Compound 39, 500 MHz, $CDCl_3$
$C_{11}H_{23}O$
OH
O
HO
HO
O
H
O
O
OH
[rel]
3.0
2.5
2.0
1.5
1.0
0.5
0.0
180
160
140
120
100
80
60
40
[ppm]

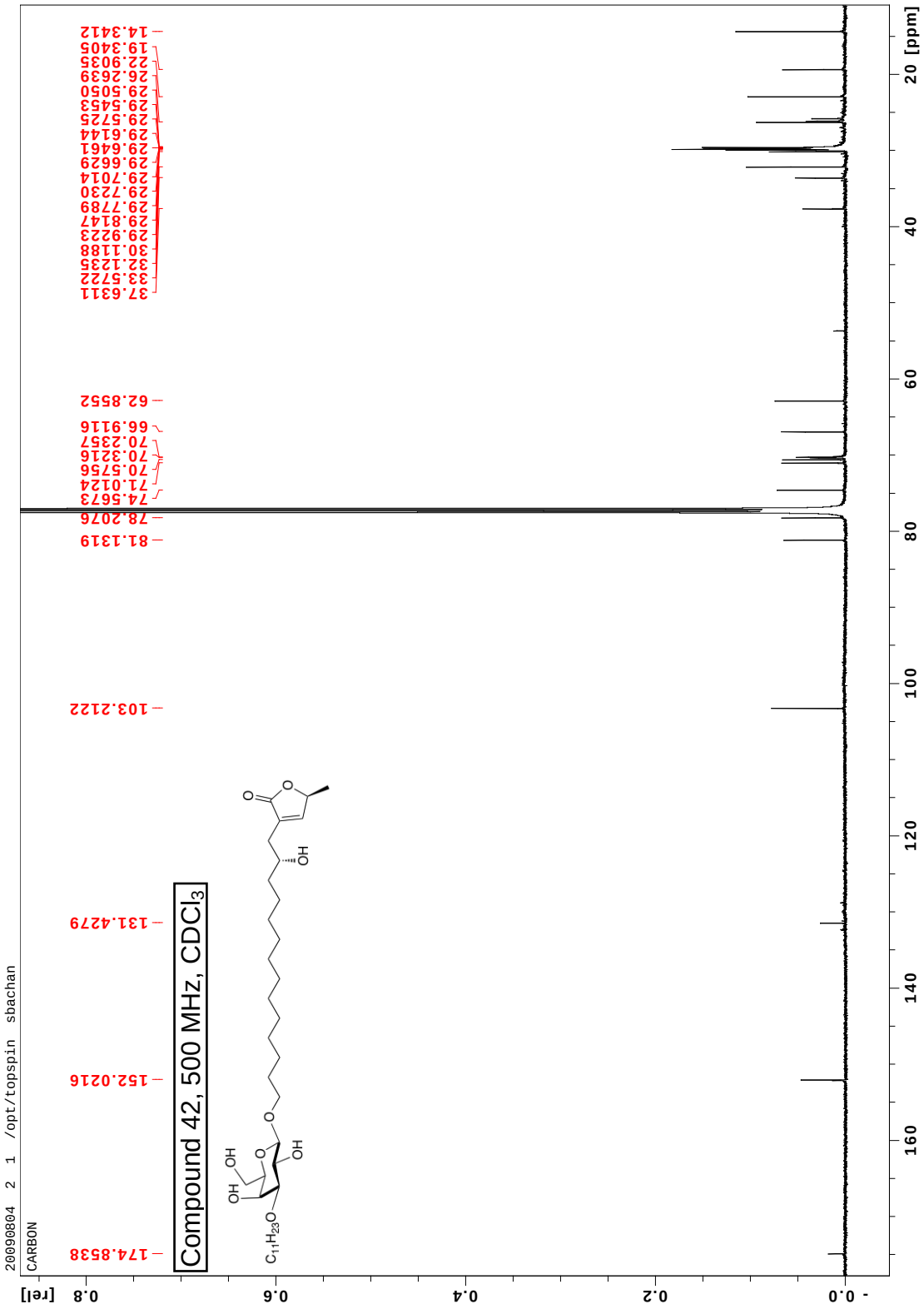

20101122 4 1 /opt/topspin sbachan
PROTON
Compound 40, 500 MHz, CDCl3
C11H23O
HO
HO
OH
O
O(CH2)15CH3
[rel]
25
20
15
10
5
0
[ppm]
4
3
2
1
1.0000
2.0829
1.0794
1.0129
1.1335
2.1188
1.1044
1.1075
1.0708
0.9759
0.8947
0.9699
6.5572
45.6935
6.2411

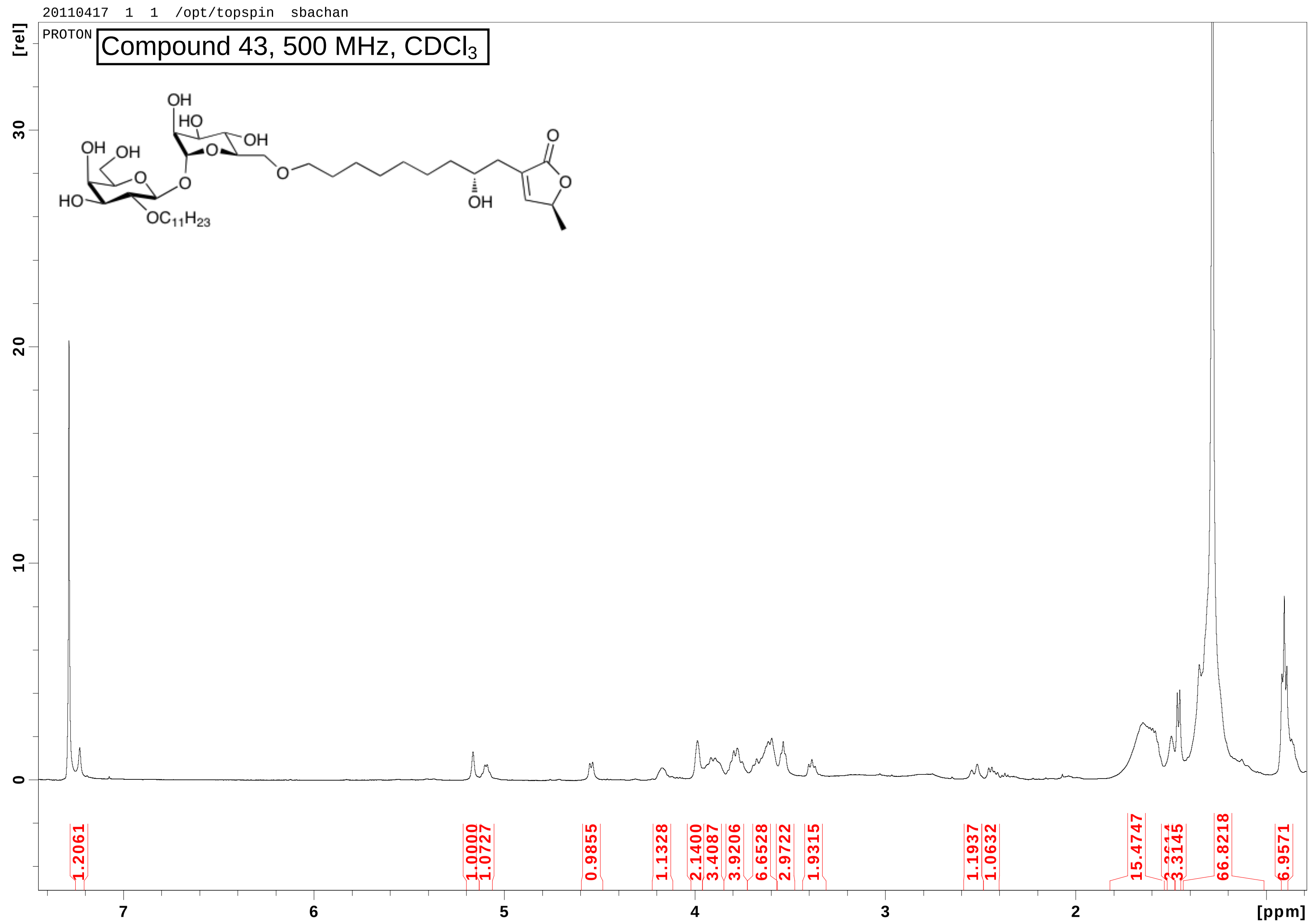

Compound 40, 500 MHz, CDCl3
20101122 5 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
99.6147
76.0512
73.5354
71.8256
71.5443
71.3350
68.1118
62.3039
32.1549
32.1356
30.6248
29.9310
29.9110
29.8929
29.8389
29.8181
29.7944
29.7366
29.6356
29.6209
29.5923
29.5602
26.3475
26.3113
22.9189
14.3527
OH
O
HO
HO
O(CH2)15CH3
C11H23O

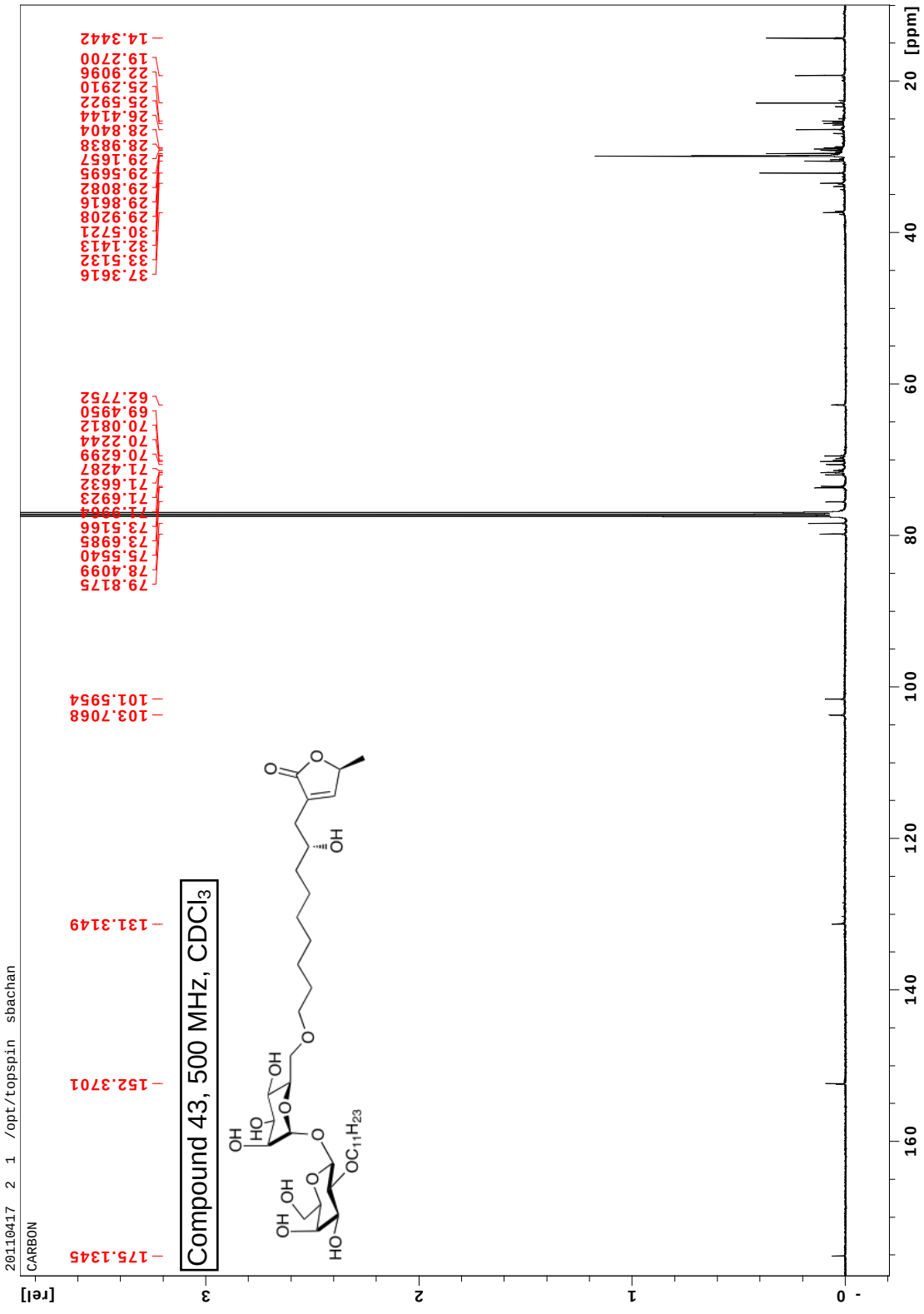

20110614 1 1 /opt/topspin sbachan
PROTON
Compound 41, 500 MHz, CDCl3
C11H23O
OH
HO
HO
OH
OH
O
O
O
[rel]
60
40
20
0
[ppm]
7
6
5
4
3
2
1.0000
0.9415
1.0754
0.8820
3.3224
3.8875
3.2328
1.3271
1.2535
1.6940
1.8949
7.2464
4.8734
4.4080
5.2631
26.7713
3.1824

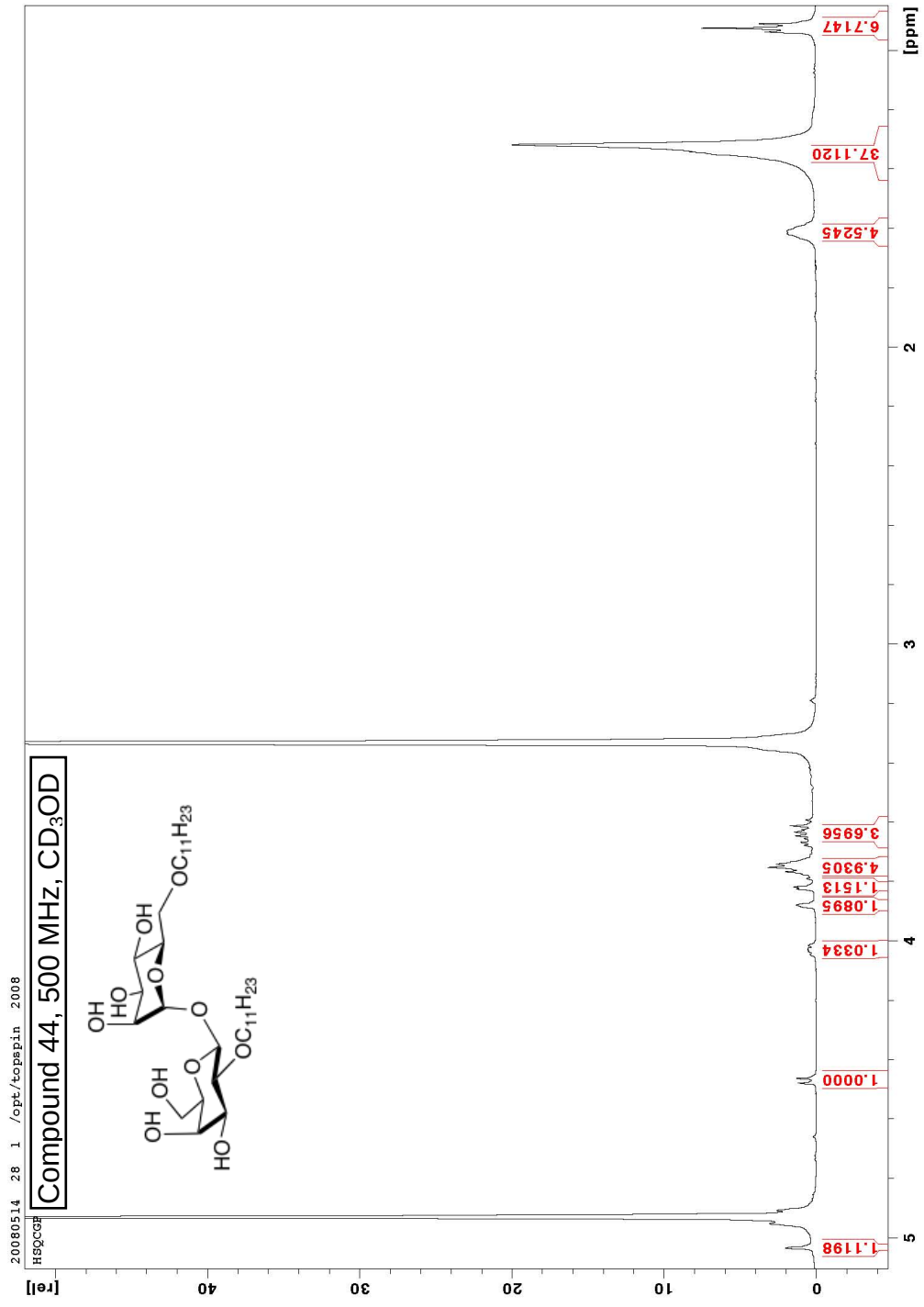

Compound 41, 500 MHz, CDCl3
20110614 2 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
174.9292
152.2724
152.2013
131.3368
131.3012
99.7557
99.6844
78.3126
78.2854
75.9849
73.5983
73.5642
71.8391
71.8301
71.8027
71.7765
71.3279
71.2976
70.0787
68.0726
68.0437
62.3144
62.2927
37.3881
37.3521
33.6044
32.1323
30.6022
29.9193
29.8380
29.7497
29.5542
26.2961
25.6617
25.6323
25.6120
25.5901
22.9071
19.3029
14.3394

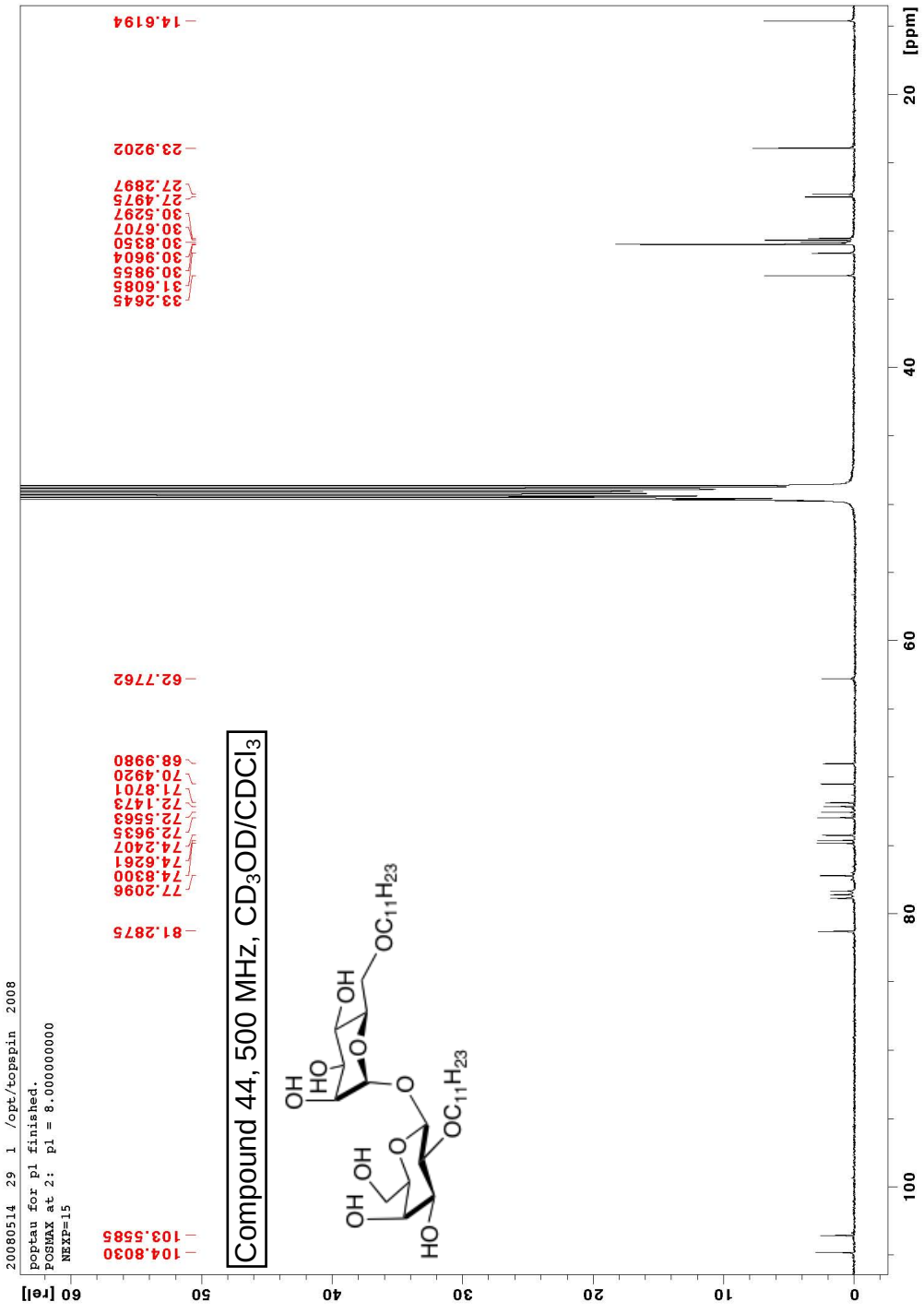

20090804 1 1 /opt/topspin sbachan
PROTON
Compound 42, 500 MHz, CDCl3
C11H23
OH
OH
OH
O
O
O
O
[rel]
0
10
20
30
[ppm]
1
2
3
4
5
6
7
1.0000
1.0210
1.1491
1.1826
1.2100
1.2207
2.2596
2.6477
1.2230
2.4347
1.1835
2.1040
18.1558
2.4651
4.3421
53.4697
6.1214

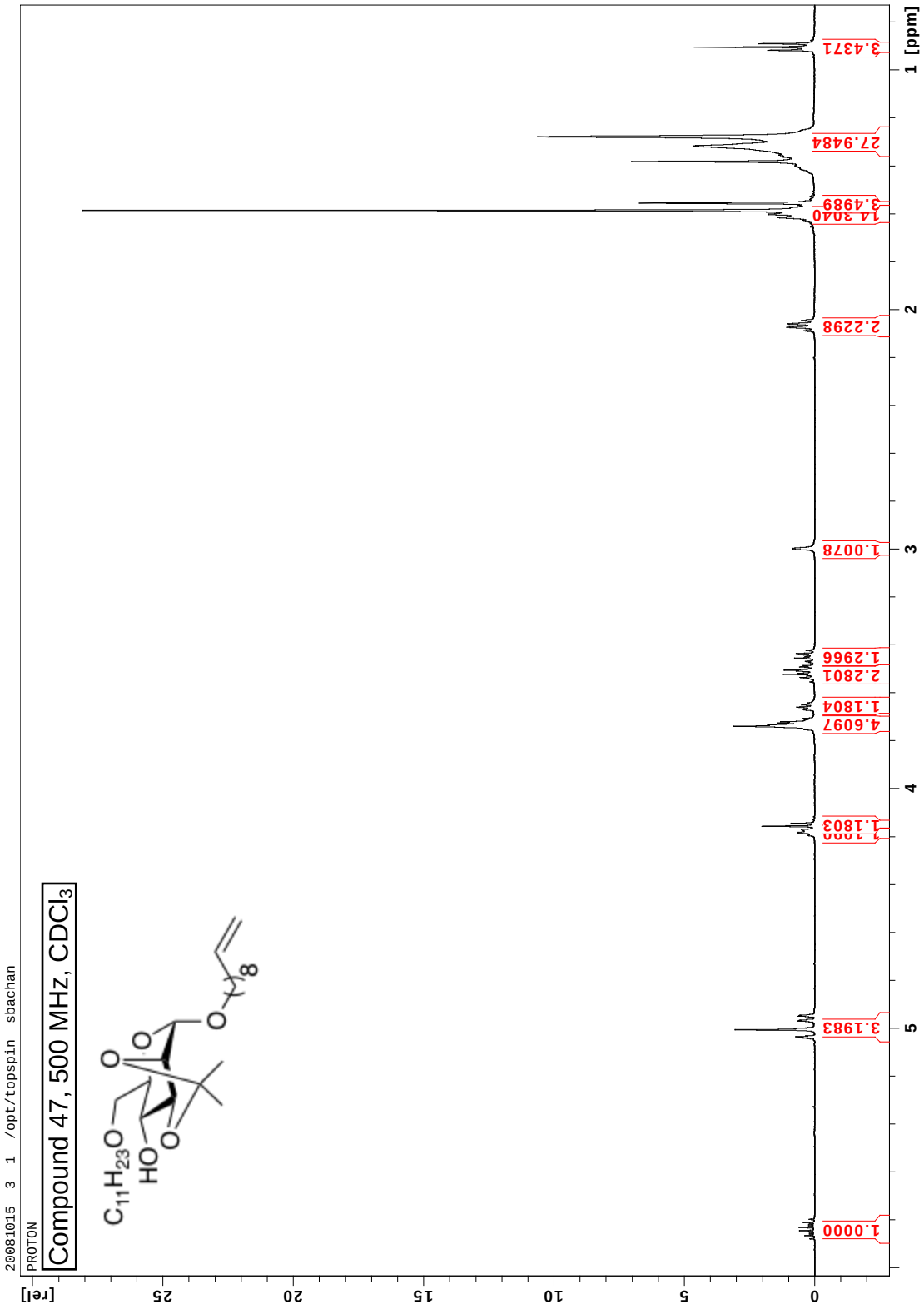

Compound 42, 500 MHz, CDCl3
C11H23O
OH
OH
OH
O
O
O
O
174.8538
152.0216
131.4279
103.2122
81.1319
78.2076
74.5673
71.0124
70.5756
70.3216
70.2357
66.9116
62.8552
37.6311
33.5722
32.1235
30.1188
29.9223
29.8147
29.7789
29.7230
29.7014
29.6629
29.6461
29.6144
29.5725
29.5453
29.5050
26.2639
22.9035
19.3405
14.3412
CARBON
20090804 2 1 /opt/topspin sbachan
[rel]
0.8
0.6
0.4
0.2
0.0
160
140
120
100
80
60
40
20 [ppm]

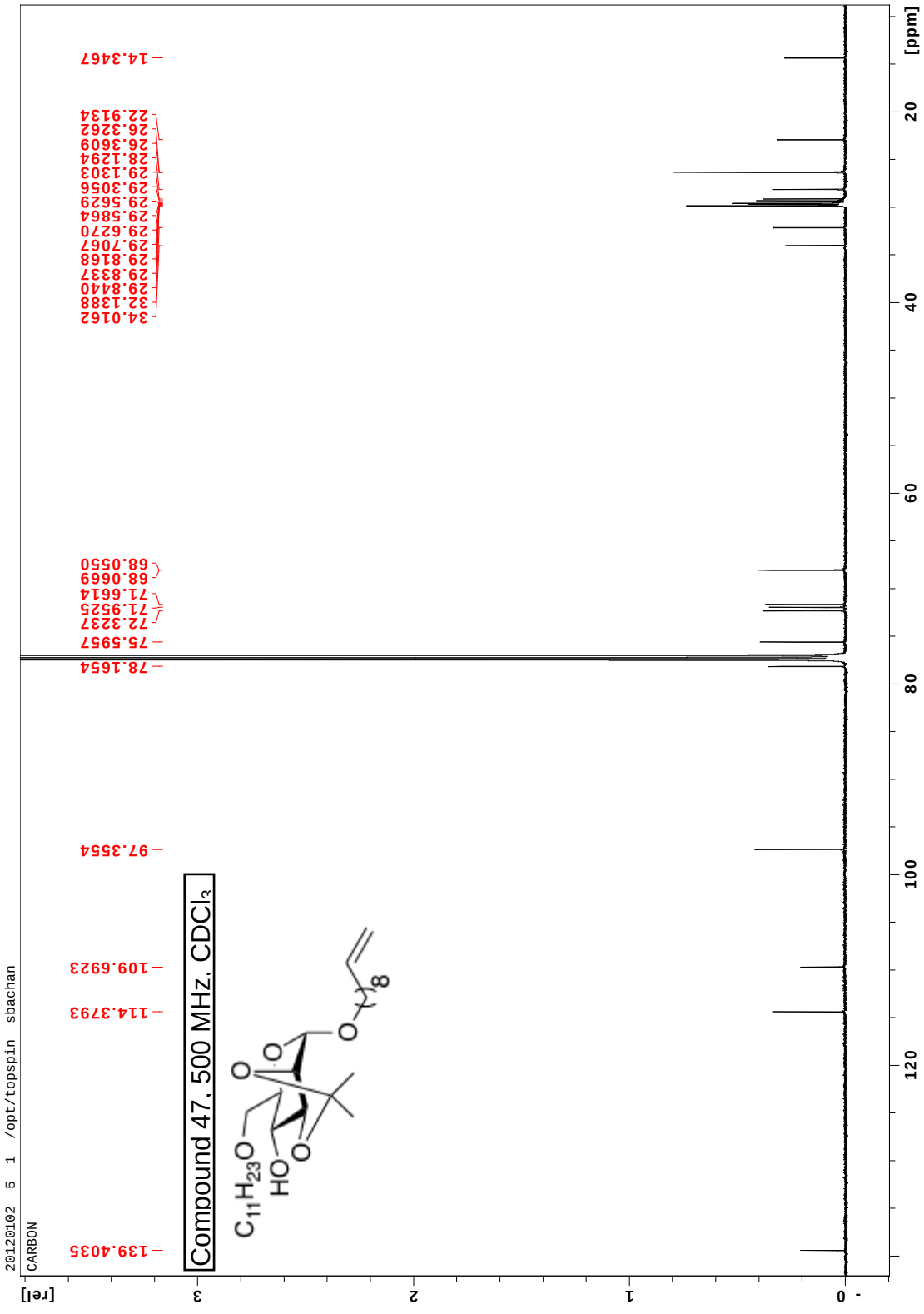

20110417 1 1 /opt/topspin sbachan
PROTON
Compound 43, 500 MHz, CDCl3
[rel]
30
20
10
0
1.2061
1.0000
1.0727
0.9855
1.1328
2.1400
3.4087
3.9206
6.6528
2.9722
1.9315
1.1937
1.0632
15.4747
3.2614
3.3145
66.8218
6.9571
7
6
5
4
3
2
[ppm]

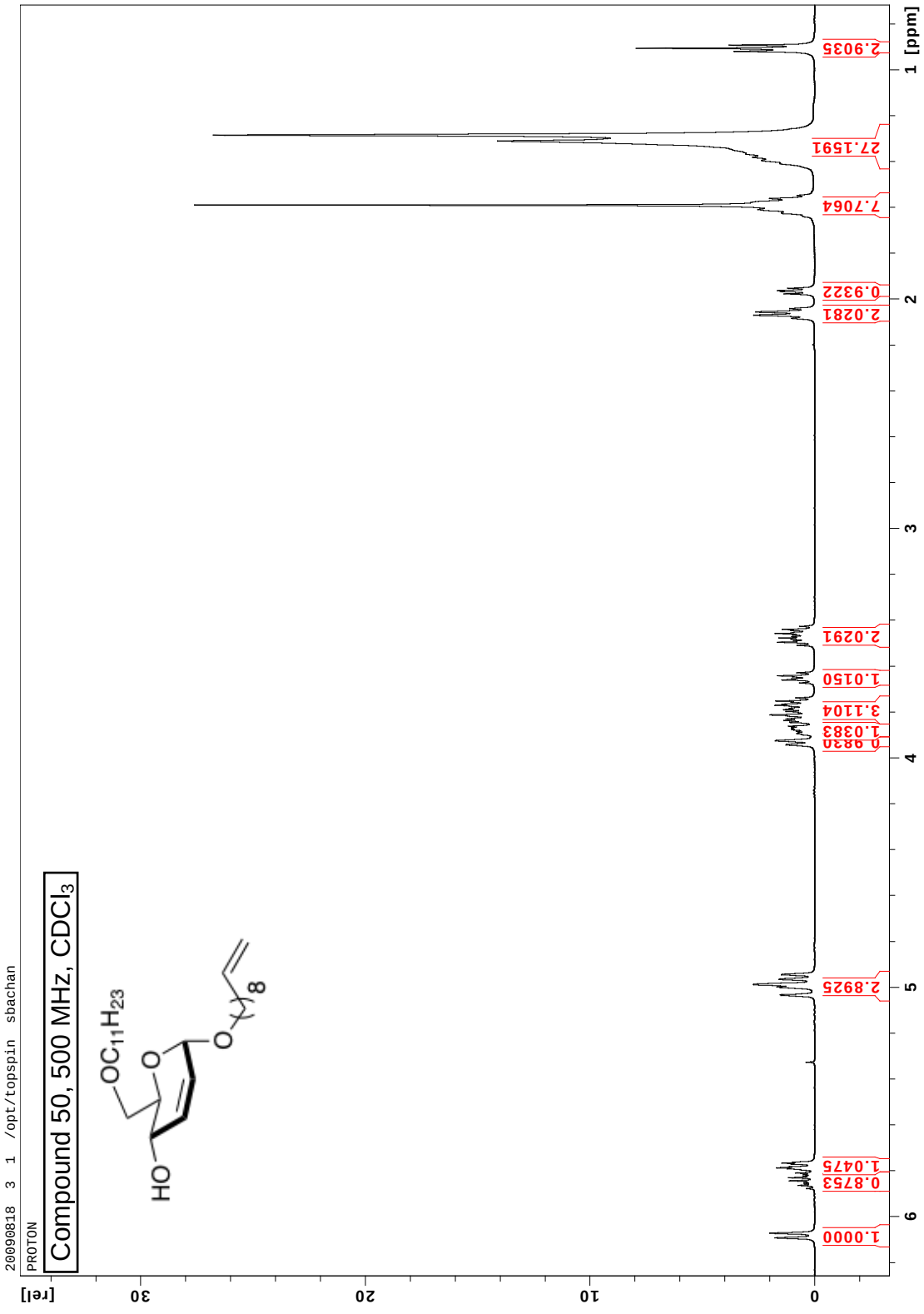

Compound 43, 500 MHz, CDCl3
20110417 2 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
175.1345
152.3701
131.3149
103.7068
101.5954
79.8175
78.4099
75.5540
73.6985
73.5166
71.6632
71.4287
70.6299
70.2244
70.0812
69.4950
62.7752
37.3616
33.5132
32.1413
30.5721
29.9208
29.8082
29.8616
29.5695
29.1657
28.9838
28.8404
26.4144
25.5922
25.2910
22.9096
19.2700
14.3442

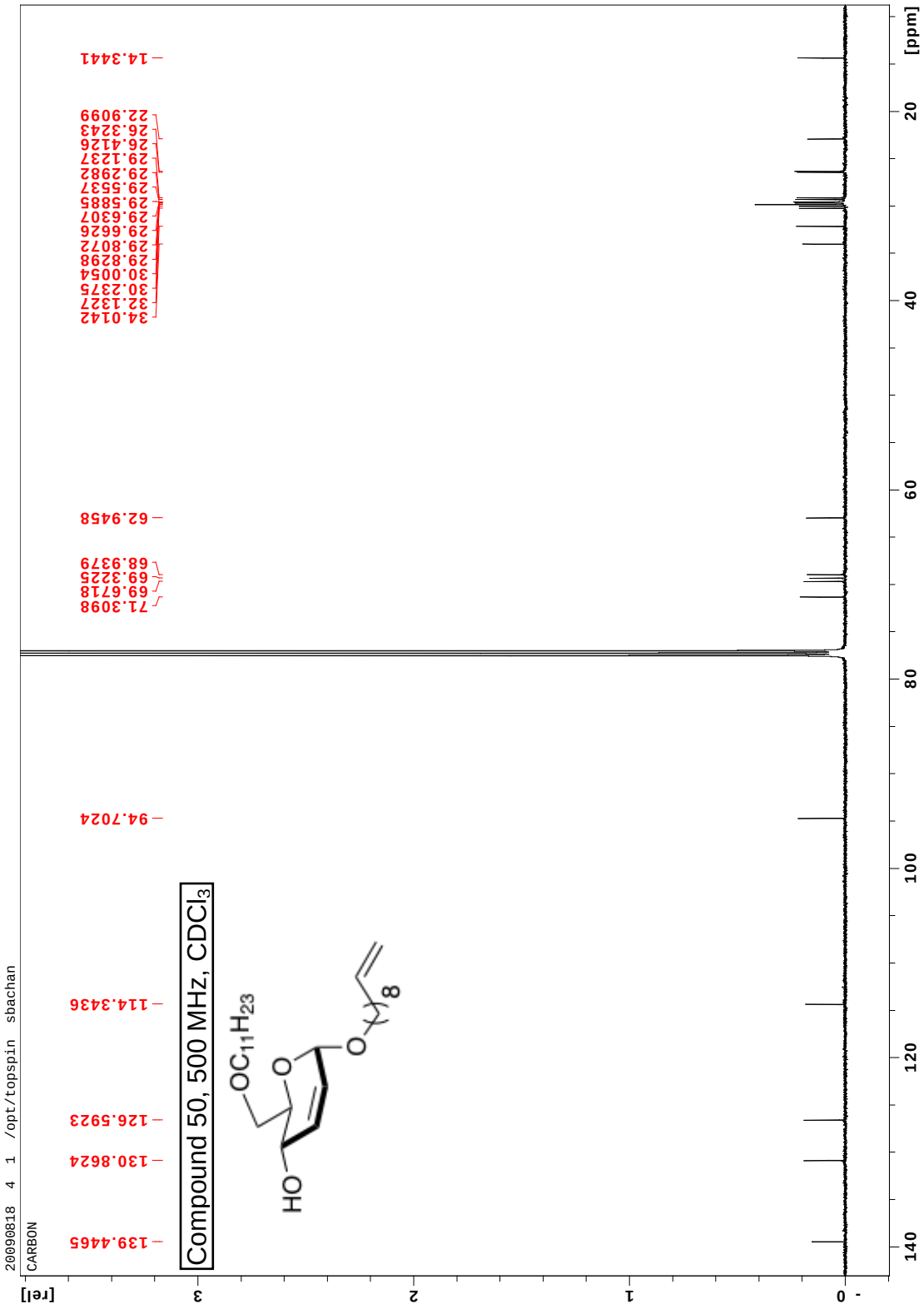

20080514 28 1 /opt/topspin 2008
HSQCGP
Compound 44, 500 MHz, CD3OD
[rel]
[ppm]
1.1198
1.0000
1.0334
1.0895
1.1513
4.9305
3.6956
6.7147
37.1120
4.5245
OH
HO
OH
HO
OH
HO
OC11H23
OC11H23

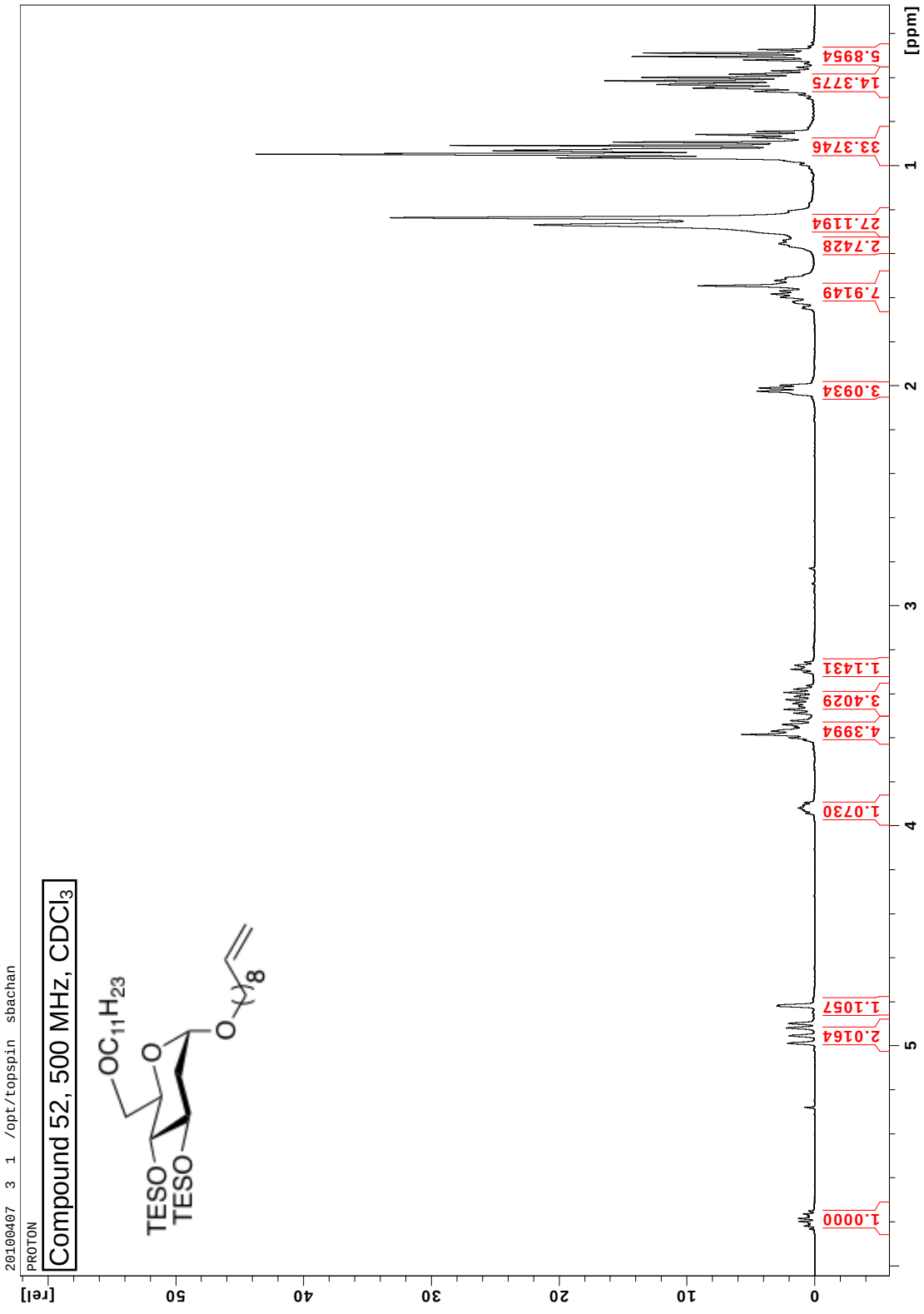

20080514 29 1 /opt/topspin 2008
poptau for pl finished.
POSHAX at 2: pl = 8.000000000
NEXP=15
Compound 44, 500 MHz, CD3OD/CDCl3
104.8030
103.5585
81.2875
77.2096
74.8300
74.6261
74.2407
72.9635
72.5563
72.1473
71.8701
70.4920
68.9980
62.7762
33.2645
31.6085
30.9855
30.9604
30.8350
30.6707
30.5297
27.4975
27.2897
23.9202
14.6194
[rel]
[ppm]

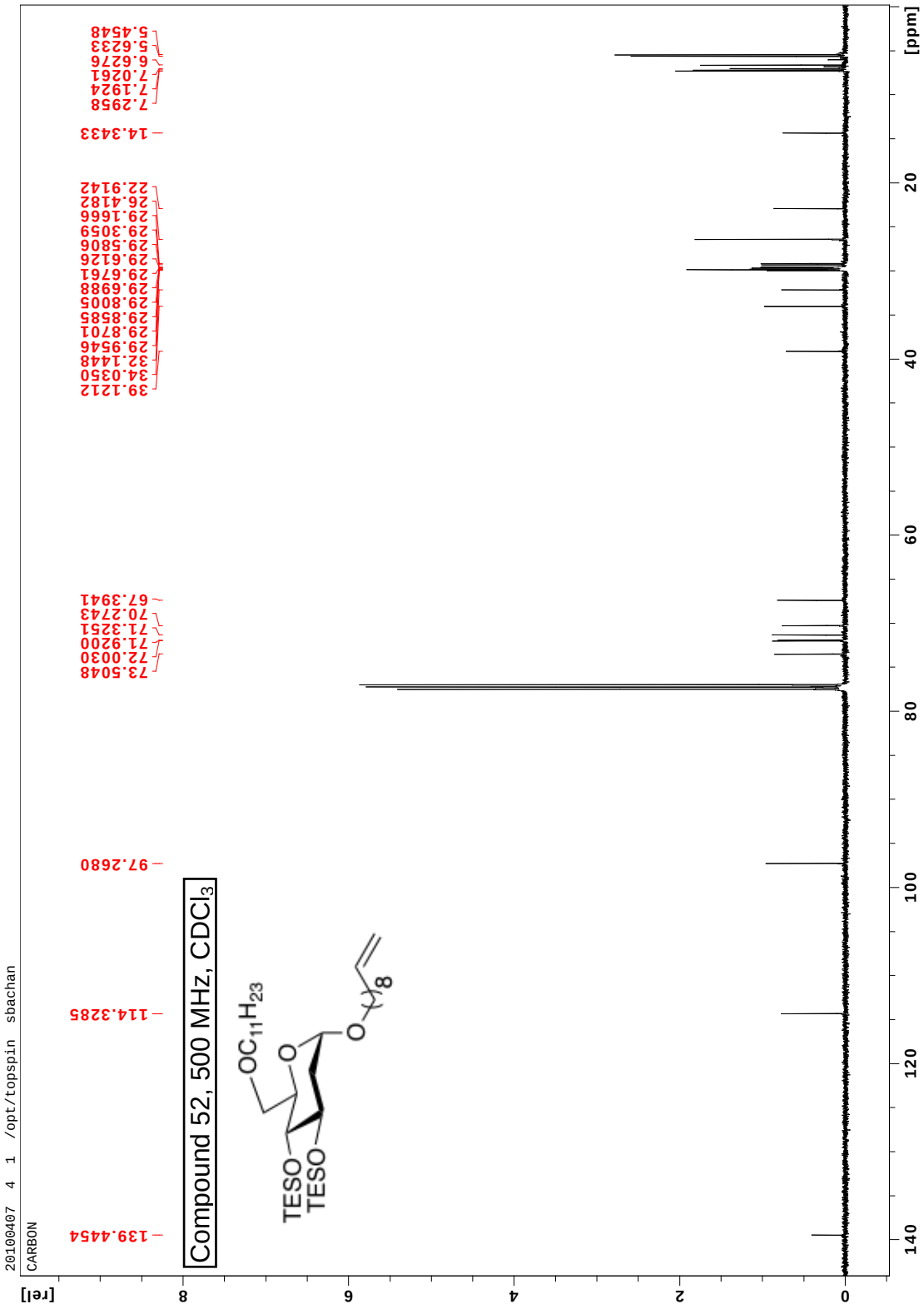

20081015 3 1 /opt/topspin sbachan
PROTON
Compound 47, 500 MHz, CDCl3
[rel]
25
20
15
10
5
0
1 [ppm]
1.0000
3.1983
1.1803
4.6097
1.1804
2.2801
1.2966
1.0078
2.2298
14.3040
3.4989
27.9484
3.4371

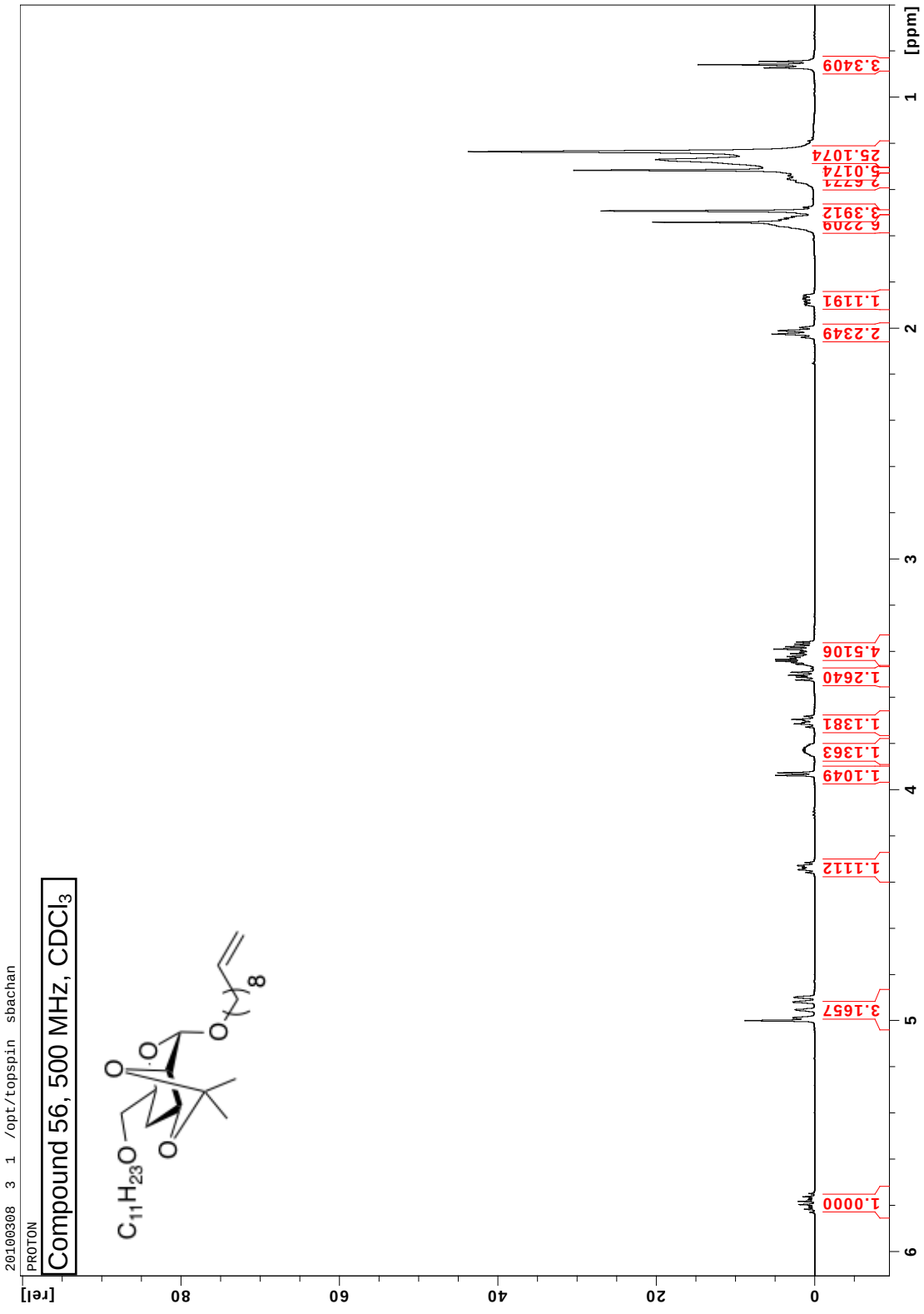

Compound 47, 500 MHz, CDCl3
C11H23O
HO
O
O
O
O
8
139.4035
114.3793
109.6923
97.3554
78.1654
75.5957
72.3237
71.9525
71.6614
68.0669
68.0550
34.0162
32.1388
29.8440
29.8337
29.8168
29.7067
29.6270
29.5864
29.5629
29.3056
29.1303
28.1294
26.3609
26.3262
22.9134
14.3467
CARBON
20120102 5 1 /opt/topspin sbachan
[rel]
[ppm]

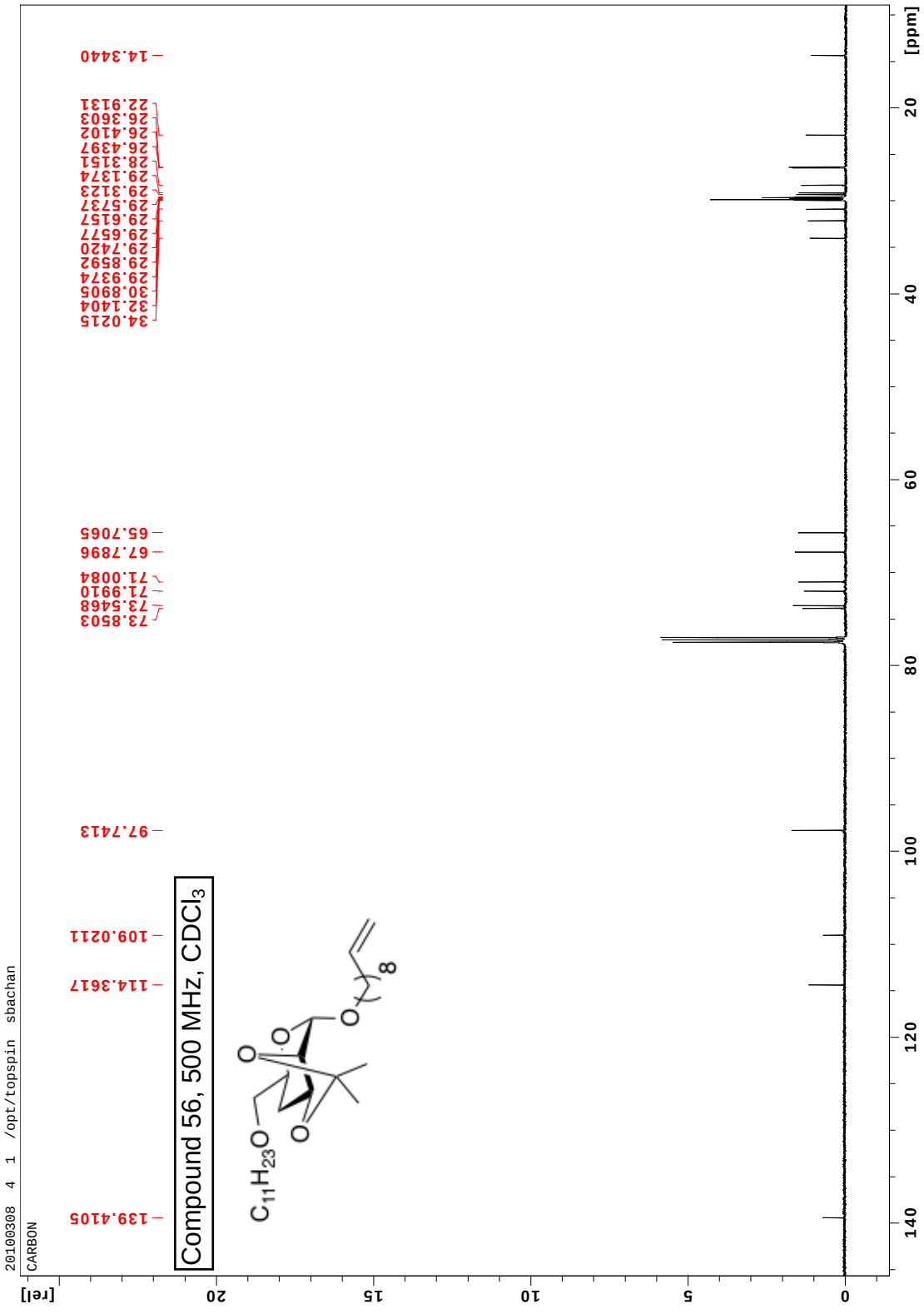

20090818 3 1 /opt/topspin sbachan
PROTON
Compound 50, 500 MHz, CDCl3
[rel]
30
20
10
0
6
5
4
3
2
1 [ppm]
1.0000
0.8753
1.0475
2.8925
0.9830
1.0383
3.1104
1.0150
2.0291
2.0281
0.9322
7.7064
27.1591
2.9035
HO
OC11H23
O
O
8

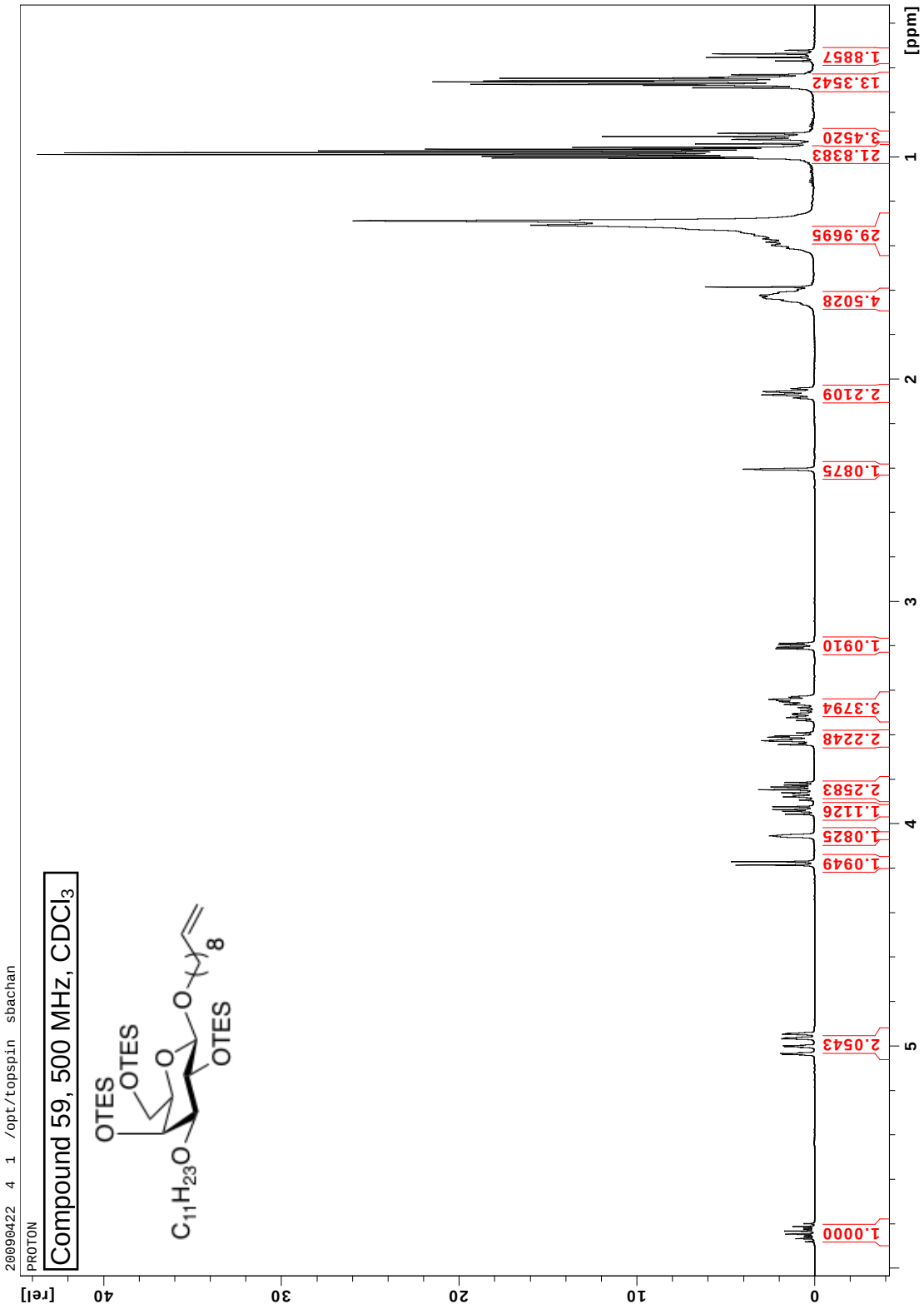

20090818 4 1 /opt/topspin sbachan
CARBON
Compound 50, 500 MHz, CDCl3
OC11H23
HO
O
O
8
139.4465
130.8624
126.5923
114.3436
94.7024
71.3098
69.6718
69.3225
68.9379
62.9458
34.0142
32.1327
30.2375
30.0054
29.8298
29.8072
29.6626
29.6307
29.5885
29.5537
29.2982
29.1237
26.4126
26.3243
22.9099
14.3441
[rel]
[ppm]

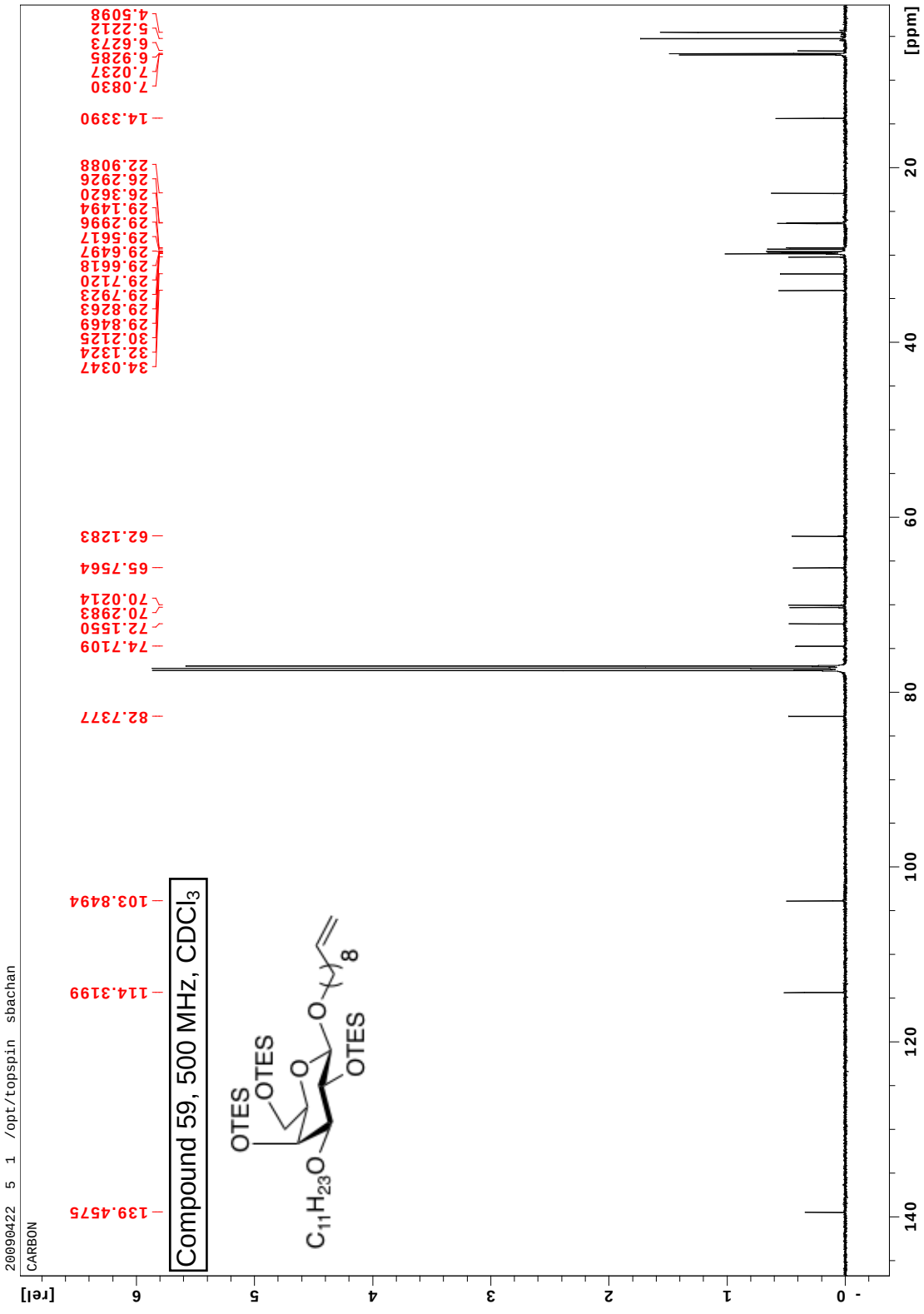

20100407 3 1 /opt/topspin sbachan
PROTON
Compound 52, 500 MHz, CDCl3
TESO
TESO
OC11H23
O
O
8
[rel]
0
10
20
30
40
50
[ppm]
1
2
3
4
5
1.0000
2.0164
1.1057
1.0730
4.3994
3.4029
1.1431
3.0934
7.9149
2.7428
27.1194
33.3746
14.3775
5.8954

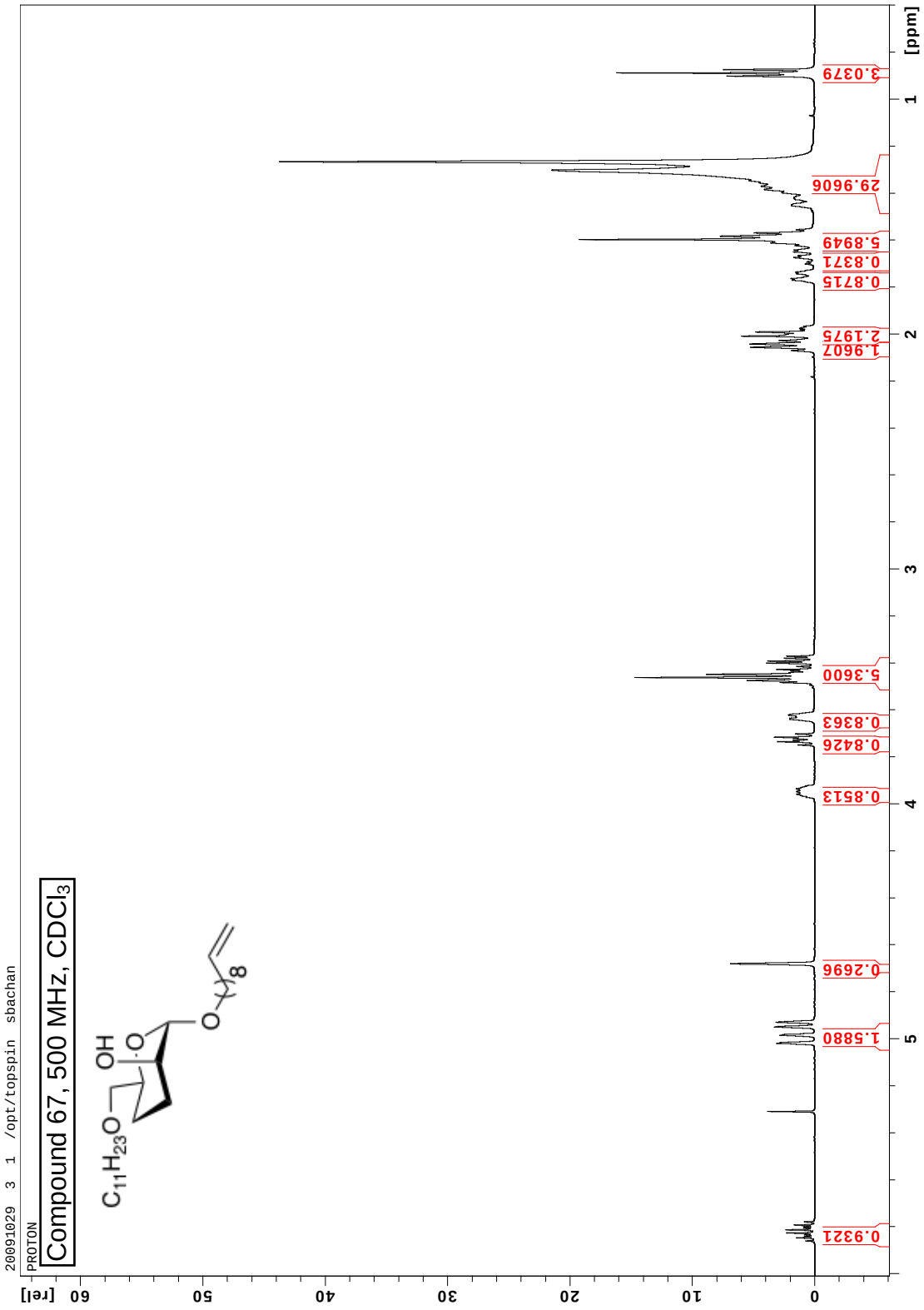

Compound 52, 500 MHz, CDCl3
TESO
TESO
OC11H23
8
20100407 4 1 /opt/topspin sbachan
CARBON
[rel]
[ppm]
139.4454
114.3285
97.2680
73.5048
72.0030
71.9200
71.3251
70.2743
67.3941
39.1212
34.0350
32.1448
29.9546
29.8701
29.8585
29.8005
29.6988
29.6761
29.6126
29.5806
29.3059
29.1666
26.4182
22.9142
14.3433
7.2958
7.1924
7.0261
6.6276
5.6233
5.4548

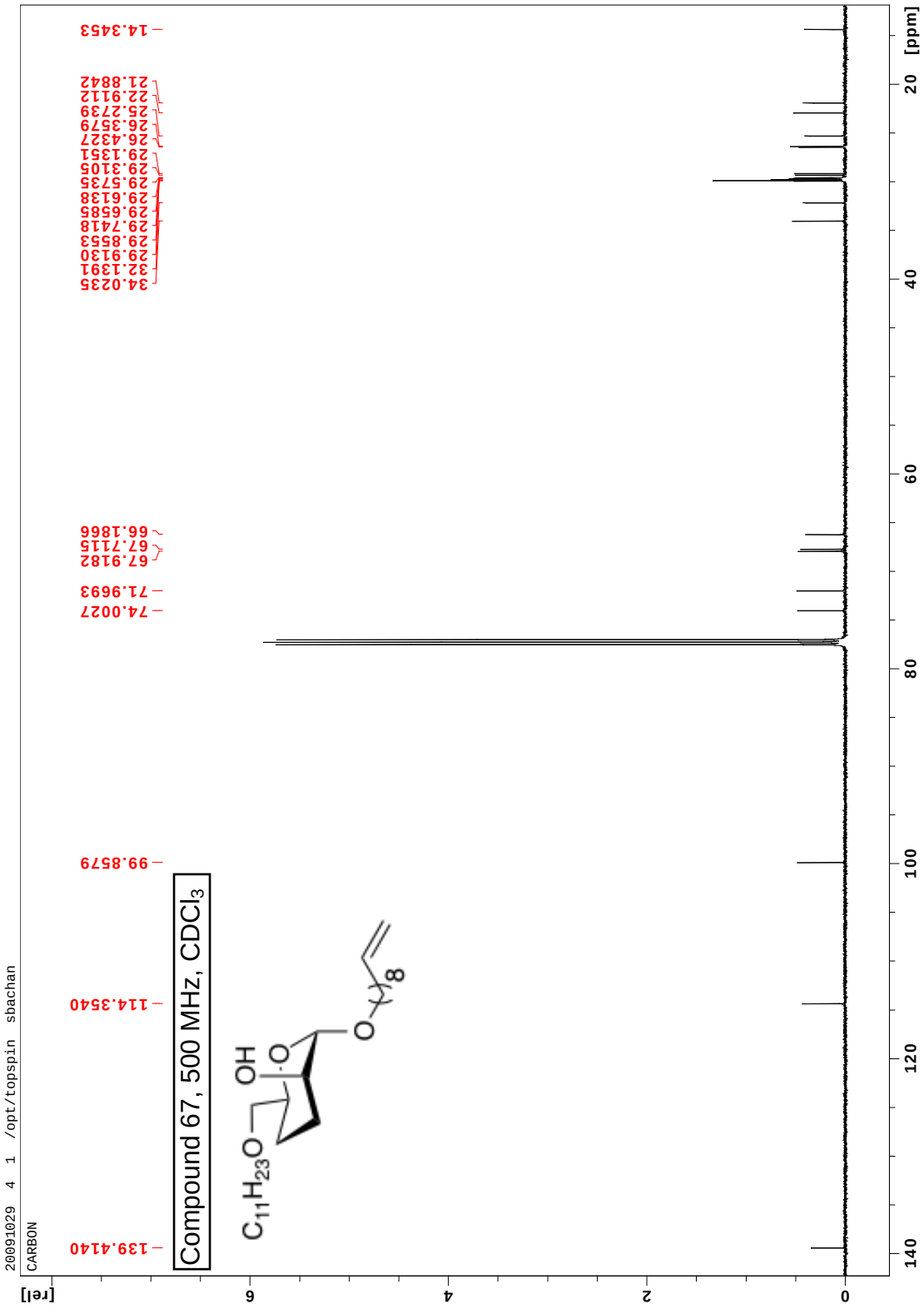

20100308 3 1 /opt/topspin sbachan
PROTON
Compound 56, 500 MHz, CDCl3
C11H23O
O
O
O
O
8
[rel]
0
20
40
60
80
[ppm]
6
5
4
3
2
1
1.0000
3.1657
1.1112
1.1049
1.1363
1.1381
1.2640
4.5106
2.2349
1.1191
6.2209
3.3912
2.6721
5.0174
25.1074
3.3409

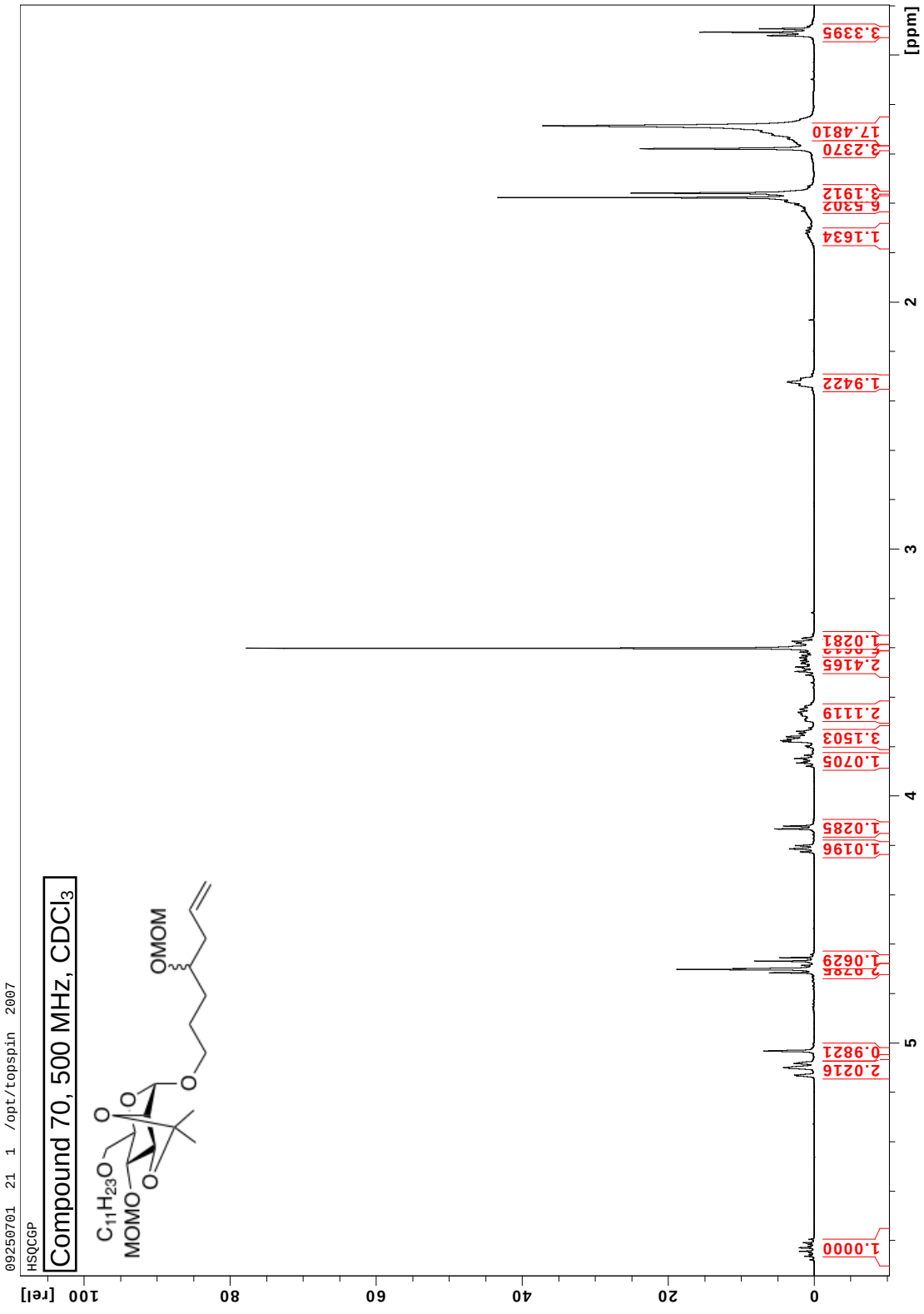

20100308 4 1 /opt/topspin sbachan
CARBON
Compound 56, 500 MHz, CDCl3
C11H23O
O
O
O
O
8
139.4105
114.3617
109.0211
97.7413
73.8503
73.5468
71.9910
71.0084
67.7896
65.7065
34.0215
32.1404
30.8905
29.9374
29.8592
29.7420
29.6577
29.6157
29.5737
29.3123
29.1374
28.3151
26.4397
26.4102
26.3603
22.9131
14.3440
[rel]
20
15
10
5
0
[ppm]
140
120
100
80
60
40
20

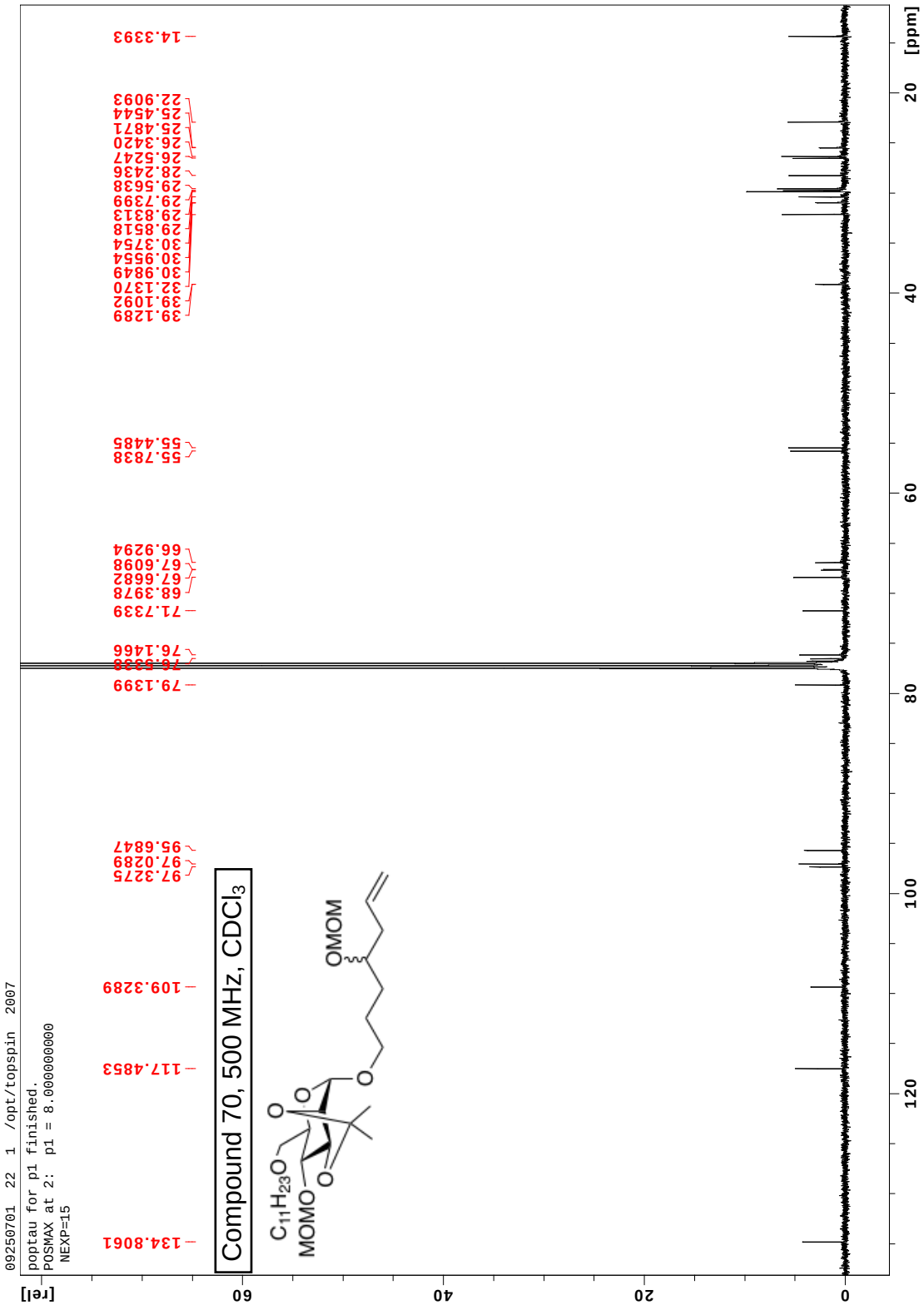

20090422 4 1 /opt/topspin sbachan
PROTON
Compound 59, 500 MHz, CDCl3
OTES
OTES
O
O
OTES
C11H23O
8
[rel]
40
30
20
10
0
[ppm]
1.0000
2.0543
1.0949
1.0825
1.1126
2.2583
2.2248
3.3794
1.0910
1.0875
2.2109
4.5028
29.9695
21.8383
3.4520
13.3542
1.8857

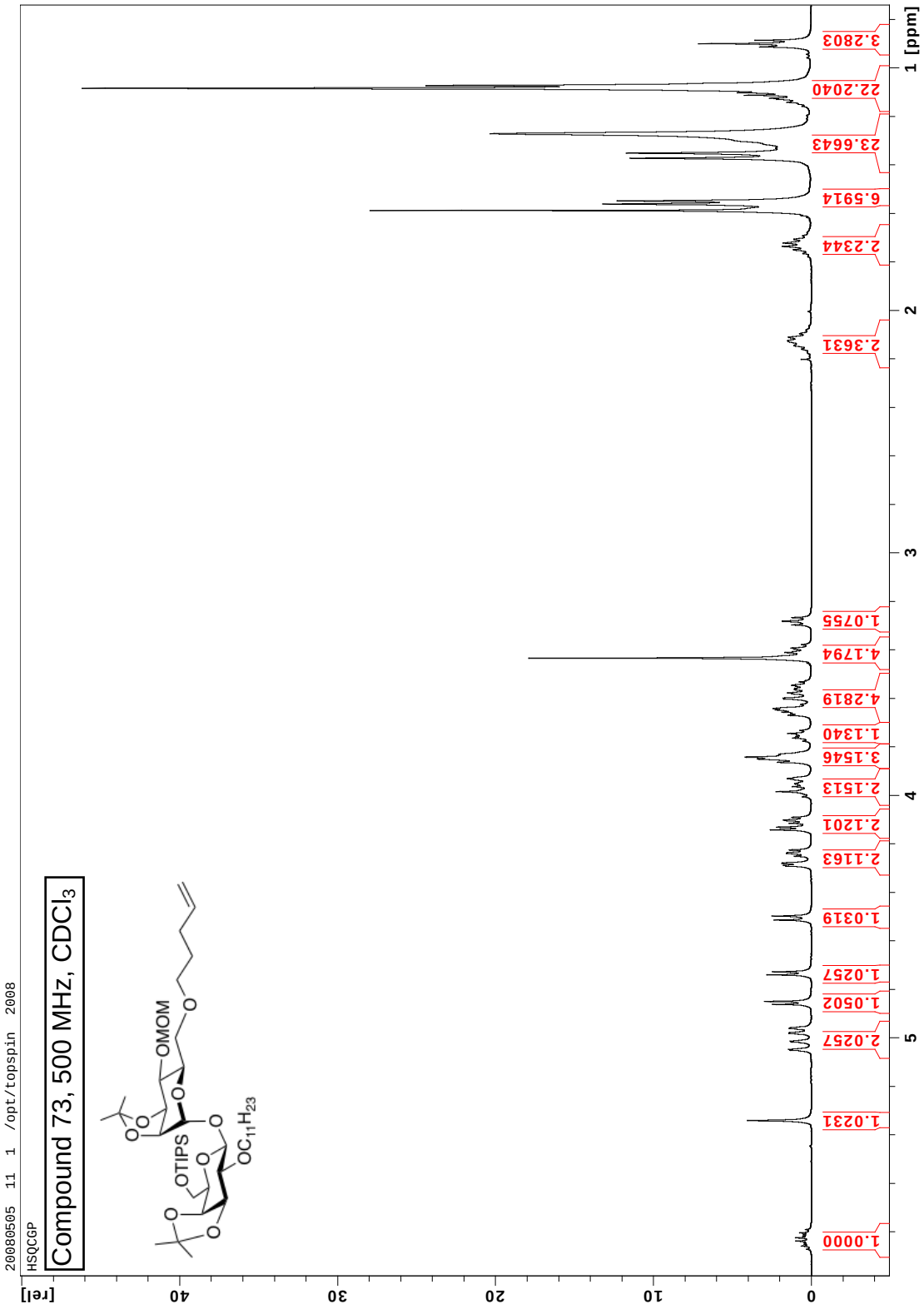

Compound 59, 500 MHz, CDCl3
OTES
OTES
O
O
OTES
C11H23O
8
[rel]
[ppm]
139.4575
114.3199
103.8494
82.7377
74.7109
72.1550
70.2983
70.0214
65.7564
62.1283
34.0347
32.1324
30.2125
29.8469
29.8263
29.7923
29.7120
29.6618
29.6497
29.5617
29.2996
29.1494
26.3620
26.2926
22.9088
14.3390
7.0830
7.0237
6.9285
6.6273
5.2212
4.5098
20090422 5 1 /opt/topspin sbachan
CARBON

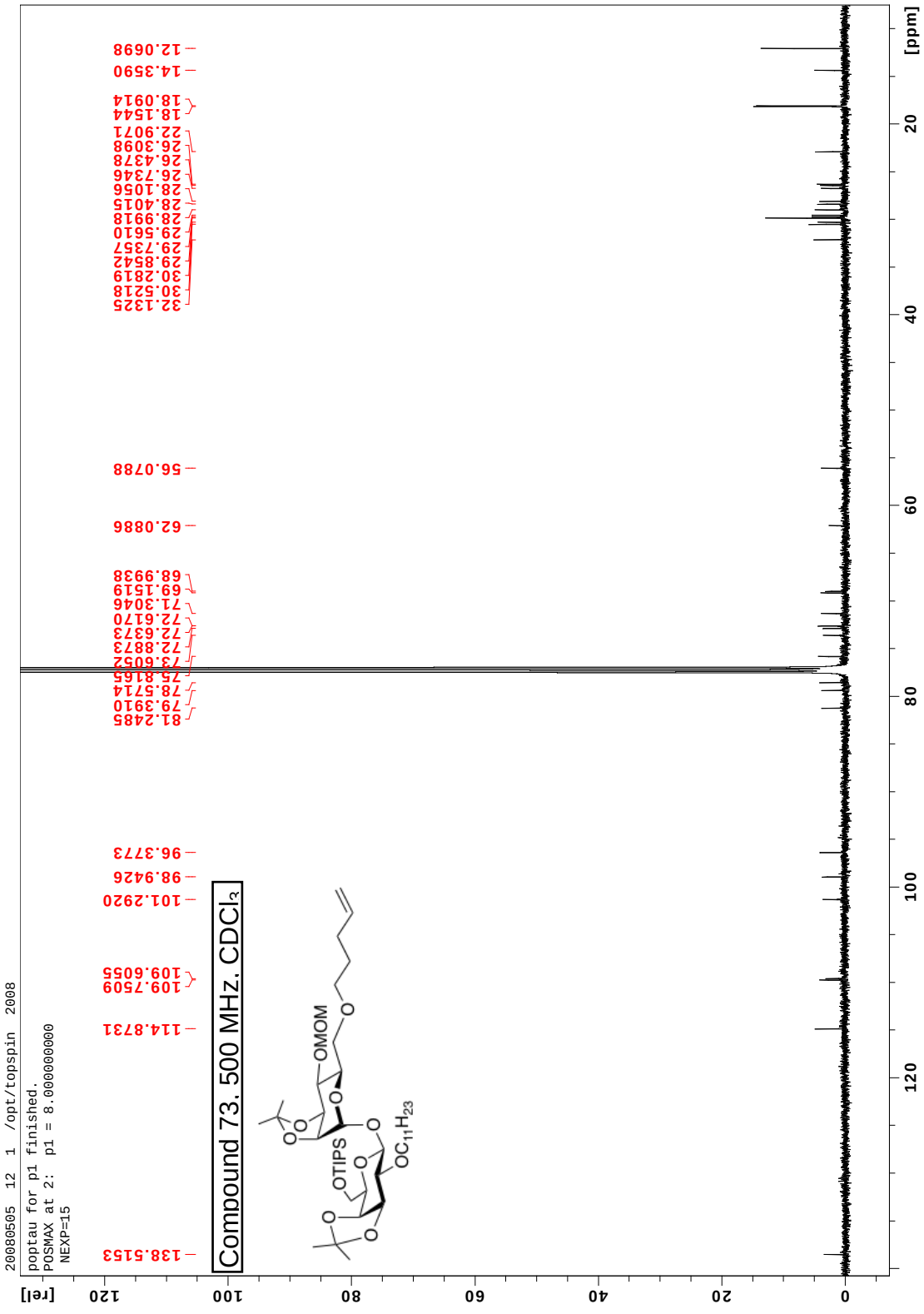

20091029 3 1 /opt/topspin sbachan
PROTON
Compound 67, 500 MHz, CDCl3
C11H23O
OH
O
O
8
[rel]
60
50
40
30
20
10
0
[ppm]
0.9321
1.5880
0.2696
0.8513
0.8426
0.8363
5.3600
1.9607
2.1975
0.8715
0.8371
5.8949
29.9606
3.0379


20091029 4 1 /opt/topspin sbachan
CARBON
Compound 67, 500 MHz, CDCl3
C11H23O
OH
O
O
8
139.4140
114.3540
99.8579
74.0027
71.9693
67.9182
67.7115
66.1866
34.0235
32.1391
29.9130
29.8553
29.7418
29.6585
29.6138
29.5735
29.3105
29.1351
26.4327
26.3579
25.2739
22.9112
21.8842
14.3453
[rel]
[ppm]


09250701 21 1 /opt/topspin 2007
HSQCGP
Compound 70, 500 MHz, CDCl3
C11H23O
MOMO
OMOM
[rel]
[ppm]
1.0000
2.0216
0.9821
1.0629
1.0196
1.0285
1.0705
3.1503
2.1119
2.4165
1.0281
1.9422
1.1634
6.5302
3.1912
3.2370
17.4810
3.3395


Compound 70, 500 MHz, CDCl3
C11H23O
MOMO
OMOM
14.3393
22.9093
25.4544
25.4871
26.3420
26.5247
28.2436
29.5638
29.7399
29.8313
29.8518
30.3754
30.9554
30.9849
32.1370
39.1092
39.1289
55.4485
55.7838
66.9294
67.6098
67.6682
68.3978
71.7339
76.1466
76.5338
79.1399
95.6847
97.0289
97.3275
109.3289
117.4853
134.8061
[rel]
0
20
40
60
[ppm]
120
100
80
60
40
20


20080505 11 1 /opt/topspin 2008
HSQCGP
Compound 73, 500 MHz, CDCl3
OMOM
OTIPS
OC11H23
[rel]
40
30
20
10
0
1 [ppm]
1.0000
1.0231
2.0257
1.0502
1.0257
1.0319
2.1163
2.1201
2.1513
3.1546
1.1340
4.2819
4.1794
1.0755
2.3631
2.2344
6.5914
23.6643
22.2040
3.2803


Compound 73. 500 MHz. CDCl3
OMOM
OTIPS
OC11H23
[rel]
[ppm]
0
20
40
60
80
100
120
12.0698
14.3590
18.0914
18.1544
22.9071
26.3098
26.4378
26.7346
28.1056
28.4015
28.9918
29.5610
29.7357
29.8542
30.2819
30.5218
32.1325
56.0788
62.0886
68.9938
69.1519
71.3046
72.6170
72.6373
72.8873
73.6052
75.8165
78.5714
79.3910
81.2485
96.3773
98.9426
101.2920
109.6055
109.7509
114.8731
138.5153
20080505 12 1 /opt/topspin 2008
poptau for p1 finished.
POSMAX at 2: p1 = 8.000000000
NEXP=15

REFERENCES

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