THE DISCOVERY OF NOVEL MACROLIDE ANTIBIOTICS THAT ADDRESS BACTERIAL RESISTANCE

A Dissertation

Submitted to the

Temple University Graduate Board

In Part Fulfillment of the Requirements

For the Degree of

DOCTOR OF PHILOSOPHY

By

Miseon Lee

May, 2017

Examining Committee Members

Dr. Rodrigo B. Andrade, Research Advisor, Department of Chemistry
Dr. Scott McN. Sieburth, Committee Chair, Department of Chemistry
Dr. Franklin A. Davis, Committee Member, Department of Chemistry
Dr. Kevin C. Cannon, External Committee Member, Pennsylvania State University

---

ABSTRACT

Bacterial resistance is a formidable 21st-century global public health threat. If left unaddressed, we risk moving toward a "post-antibiotic era." While resistance is a natural consequence of antibiotic use, the rate at which pathogenic bacteria have evaded multiple classes of drugs has markedly outpaced the introduction of new ones. New antibiotics are desperately needed to fill this void. Macrolides are one of the safest and most effective drug classes in medicine; however, resistance has compromised efficacy. To date, three generations have been developed with only the lattermost targeting bacterial resistance. Single next-generation macrolides will not keep pace with the inevitable onset of resistance; thus, there is a critical need to greatly accelerate the procurement of multiple future-generation antibiotics to tackle both current and future resistance mechanisms. My research is to meet this need by designing, synthesizing, and evaluating a novel, future-generation macrolide antibiotics that will serve as an armamentarium to be individually deployed on demand.

In the previous research in Andrade group, we synthesized and evaluated various desmethyl ketolide analogs. The fact that 4-desmethyl telithromycin was *fourfold* less potent than telithromycin against A2058G mutants indicated replacing the 4-Me with hydrogen (i.e., desmethylation) to avoid a steric clash with the 2-amino group of G2058 was insufficient in rescuing bioactivity. Guided by MD simulation, we concluded a logical, superior alternative strategy was the replacement of the 4-Me group with one possessing a smaller vdW radius and capable of establishing favorable interactions with both wild-type and A2058G mutant ribosomes. Specifically, we reasoned that 4-fluoro solithromycin

would be ideal candidate. The hypothesis was that the 4-fluoro moiety would engage in dipole-dipole interactions (C-F···H) with the exocyclic 2-amino group of guanine, which is based on accumulated evidence that strategic placement of organofluorine can strongly impact potency, selectivity, and physicochemical properties. In addition, the axially disposed of 4-fluorine would provide conformational stabilization from a gauche effect with the vicinal O5 group.

The novel synthetic routes to unexplored desosamine analogs at the C3'-amino substituent to the macrolide antibiotic would play a role in bioactivity and resistance. Hofmann reaction was employed to execute the same 2,3-epoxide ring opening method without removing desosamine and re-glycosylating. This markedly reduces the steps, time, and cost involved in preparing novel desosamine-modified analogs. Significantly, this route enables the first synthesis of N,N'-disubstituted desosamine analogs from an epoxide, which was utilized to prepare novel analogs of clarithromycin.

The application of *in situ* click chemistry toward the discovery of novel macrolide antibiotics first required the synthesis of suitable azide and aryl alkyne reactants. Alkyne partners were procured by commercial vendors or chemical synthesis. We targeted two logical, validated positions to tether the side chains, specifically N11 on the macrolactone and N3' of desosamine. The first (N11) has been the most utilized. Moreover, extensive structure-activity relationships have revealed a four-carbon tether is ideal. Based on the solithromycin−*E.coli* X-ray structure, I designed, synthesized, and evaluated dehydro solithromycin, which possesses an (***E***)-alkene in the side-chain. The use of an unsaturated side chain would conformationally preorganize the bi-aryl side chain in order to pay the entropic penalty and thus favorably contribute to the overall binding. An insightful

observation made from MD simulationed ribosomes bound with to solithromycin revealed that the interaction of the side-chain includes H-binding as well as π-stacking. The hypothesis was that employing tethered side-chains bearing motifs that maximize H-bonding and π-stacking would be superior antibiotics for treating resistant bacterial strains bearing *erm*-mediated N6 methyl and dimethylated ribosomes. To test this hypothesis, we developed various analogs with different alkynes by introducing different functional groups at the 3 and 5 positions on the aromatic ring. Another desosamine sugar modification is bis-azide. To date, the use of a two side chain strategy has not been reported. To access the requisite bis-azides, we employed a tactic the oxidative demethylation and alkylation of desosamine to afford bis-click solithromycin analogs.

DEDICATION

This dissertation is dedicated to my parents, family, and friends

who have been a constant source of support and encouragement

during the challenges of graduate school and life

ACKNOWLEDGEMENTS

I would first like to thank my advisor, Dr. Rodrigo A. Andrade, for giving me a home in his lab and support over the years. I am grateful for his guidance and the opportunities he has afforded me. I would not have been able to come this far without his expertise, knowledge, care, and patience. Words are not enough to express my special thanks for what he has done for me as well as how he Has truly cared for me as his student. I have learned greatly from him through his excellent teaching and outstanding advising.

I would like to thank my thesis committee members, Prof. Franklin A. Davis and Prof. Scott McN. Sieburth, for their valuable scientific opinions and assistance. I am truly honored to have both on my committee. I would like to express my gratitude to Prof. Kevin C. Cannon for serving as my external committee member of my defense. I would like to give thanks to Dr. Charles DeBrosse for his time for my research. I would like to thank Dr. Mike Lawlor and Mary-Anne Beasten who made my teaching life much more enjoyable.

I would like to give special thanks to my second family, the past and present Andrade group members. I would love to thank formal macrolide team seniors who gave me great memory and help. I would like to thank Senzhi who was a great colleague, mentor, and friend. I would love to give special thanks to Praveen and Vijay. They are not the only great colleagues but also great friends who have been going through extreme ups and downs of graduate school life together from day one. I would like to thank Christiana who is my first female lab mate and brings energy and joy to the lab. I would like to thank Sam and Xiao who are the last members of macrolide team. I never forget what we did together. Finally, thanks to Manish and Po Cheng to be there to listen.

I would like to thank my parents who have supported me to pursue this dream with confidence. Their unconditional love and care are the source of this climax in my education as well as the reason I am who I am today. I respect your wisdom and patience in providing me with more than what I asked for and needed in my life. Thank you to my sisters for their encouragement and care. They are always there to offer support, love, and inspiration.

Finally, I would like to thank my friends for their continued support and encouragement. My lifelong friends back in Korea, you are always missed. I really appreciate your generous support anytime. I also send my special thanks to Jaeeun who gave courage. I would also like to thank Dr.Sieburth's group, Dr. Sharfmeister's group, Dr.Wuest's group and Dr. Wengryniuk's group for their help. I would also like to thank every person I have had the pleasure to interact with at Temple.

TABLE OF CONTENTS

LIST OF SCHEMES

LIST OF FIGURES

LIST OF TABLES

LIST OF ABBREVIATIONS

Ac – Acetyl

ACN – Acetonitrile

Ac_2O – Acetic anhydride

AcOH – Acetic acid

aq – Aqueous

Å – Angstrom

AIBN – Azobisisobutyronitrile

AgOTf - Silver Triflate

Ar Aryl

Bn Benzyl

BF_3•Et_2O – Boron trifluoride diethyl etherate

Boc – *tert*-Butoxylcarbonyl

Boc_2O – Di-*tert*-butyl decarbonate

BnOH - Benzyl alcohol

BRSM - Based on recovered starting materials

Bu_2BOTf - Dibutylboron triflate

Bu_3SnH - Tributyltin hydride

CDI - Carbonyldiimidazole

CBr_4 - Tetrabromomethane

$CeCl_3$ -Cerium (III) chloride

CH_2Cl_2 -Dichloromethane

$Cl_3C_6H_2COCl$ -2,4,6-Trichlorobenzoyl chloride

$(COCl)_2$ - Oxalyl chloride

$CrCl_2$ - Chromium (II) Chloride

CSA - Camphor sulphonic acid

CSD - Cambridge Structural Database

DAST - Diethylaminosulfur trifluoride

DBU - 1,8 - Diazabicyclo[5.4.0]undec-7-ene

DCM - Dichloromethane

DDQ - 2,3-Dichloro-5,6-dicyano-1,4-benzoquinone

DEAD - Diethylazodicarboxylate

DIBAL – Diisobutylaluminium hydride

DIPEA - Diisopropylethylamine (Hunig's base)

DIPT - Diisopropyl tartrate

DMAP – Dimethylaminopyridine

DMF – Dimethylformamide

DMSO – Dimethylsulfoxide

Deoxofluor - Bis(2-methoxyethyl)-amino sulfur trifluoride

d.r. – Diastereomeric ratio

DTBMP - 2,6-di-*tert*-butyl-3-methyl pyridine

Et – Ethyl

Equiv. – Equivalent

Et_2O – Diethyl ether

Et_3N – Triethylamine

EtOAc – Ethyl acetate

ESI – Electrospray Ionization

FAB – Fast atomic bombardment

FLUOLEAD - 4-*tert*-2,6-Dimethylphenyl sulfur trifluoride

H_2 - Hydrogen

H_2O - Water

H_2SO_4 - Sulfuric acid

HF – Hydrofluoric acid

HG – Hoveyda-Grubbs

HATU - 1-[Bis(dimethylamino)methylene]-1H-1,2,3-triazolo[4,5-b] pyridinium 3-oxid hexafluorophosphate

HCl - Hydrochloric acid

HMDS - 1,1,1,3,3,3-Hexamethyldisilazane

HRMS – High resolution mass spectrometry

I_2- Iodine

iPr_2NEt – *N*,*N*-diisopropylethylamine

h – Hour

IR - Infrared spectroscopy

KHMDS - Potassium bis(trimethylsilyl)amide

KOH - Potassium hydroxide

LAH/$LiAlH_4$ - Lithium aluminum hydride

LiOH - Lithium hydroxide

LCMS - Liquid Chromatography Mass Spectrometer

LDA – lithium diisopropylamide

LHMDS – Lithium bis(trimethylsilyl)amide

M – Molar

Me - Methyl

MeCN - Acetonitrile

MeOH – Methanol

Me_2S - Dimethyl sulfide

MeOTf - Methyl triflate

mg - Milligram(s)

MHz - Mega hertz

mL - Milliliter(s)

mmol - Millimole(s)

$NaBH_4$ Sodium borohydride

$MgSO_4$ – Magnesium sulfate

MHz – Mega hertz

MsCl – Methanesulfonyl chloride

$NaBH_4$ – Sodium borohydride

NaH – Sodium hydride

NaOMe – Sodium methoxide

NFOBS - *N*-Fluoro-O-benzenedisulfonimide

NFSI - *N*-Fluorobenzenesulfonimide

NHK - Nozaki-Hiyama-Kishi

NMR – nuclear magnetic resonance

$NaHCO_3$ - Sodium bicarbonate

$Na_2S_2O_3$ - Sodium thiosulfate

Na_2SO_4 - Sodium sulfate

$NH_2OH{\cdot}HCl$ - Hydroxylamine hydrochloride

PDB - Protein Data Base

Ph – Phenyl

Pt_2O – Platnium dioxide

ppm – Parts per million

PPTS – Pyridinium p-toluenesulfonate

Pd/C Palladium on carbon

Ph - Phenyl

PhMe Toluene

Phth Phthalimide

PPh_3 Triphenylphosphine

Pr – Propyl

pyr - Pyridine

rt – Room temperature

RCM - ring-closing metathesis

RaNi - Raney Nickel

SAD - Sharpless asymmetric dihydroxylation

Selectfluor - *N*-Fluoro-*N*'-(chloromethyl)triethylenediamine bis(tetrafluoroborate)

SM – Starting material

$SnCl_2$ -Tin(II) chloride

TAS-F - Tris(dimethylamino)sulfonium difluoromethylsilicate

TBAF- *n*-tetrabutylammonium fluoride

TBAT - tetrabutylammonium difluorotriphenyltin

TBS – *tert*-butyldimethylsilyl

t-BuOK – Potassium *t*-butoxide

TEA – Triethylamine

TES – Triethylsilyl

TFA – Trifluoroacetic acid

THF – Tetrahydrofuran

$Ti(OEt)_4$ – Titanium (IV) ethoxide

$TiCl_4$ - Titanium tetrachloride

TLC - Thin layer chromatography

TBSCl - *t*-Butyldimethylsilyl chloride

TBSOTf - *t*-Butyldimethylsilyl triflate

t-BuOK - Potassium *t*-butoxide

t-BuOOH - *t*-butylhydroperoxide

TESCl - Triethylsilyl chloride

TESOTf - Triethylsilyl trifluoromethanesulfonate

TMSOTf - Trimethylsilyl triflate

TLC – Thin layer chromatography

tert – Tertiary

UV – Ultraviolet

CHAPTER 1

MACROLIDE ANTIBIOTICS AND THE DESMETYHL APPROACH TO ADDRESS ANTIBIOTIC RESISTANCE

1.1 Introduction

The macrolides are a class of antibiotics that have in common a macrocyclic lactone ring composed of 12 to 16 atoms to which one or more deoxysugars are attached via glycosidic bonds.[1] The term “macrolide” was originally proposed by R. B. Woodward in 1957. Etymologically, the "macro-" prefix comes from Greek meaning ‘large’ and refers to the prefix in “macrolide.” As this class of antibiotics all possess one or several carbohydrates or “glycosides,” the,"-olide" suffix takes this into account. Thus, the term “macrolide” is an abbreviated (i.e., contracted) form the descriptor “macrolactone glycoside.” Macrolides are typically isolated from the soil-dwelling actinomycete family of bacteria and have found widespread use in medicine.[2]

A number of new macrolides have been discovered over the past several decades. Proactinomycin was isolated from *Streptomyces gardneri* by Gardner and Chain in 1942.[3] Although this was the first isolated member of the macrolide family, the macrolide era was not considered to begin until the report of pikromycin. Pikromycin **1.1**, the first natural macrolide antibiotic, was isolated from *Streptomyces felleus* by Brockmann and Henkel in 1950.[4] A few years later, erythromycin **1.2** was isolated from the *Streptomyces erythreus* (*Saccharopolyspora erythraea*) by McGuire et al in 1952 (Figure 1.1). Thus, the actual macrolide era began with the human use of erythromycin dating back to 1952.

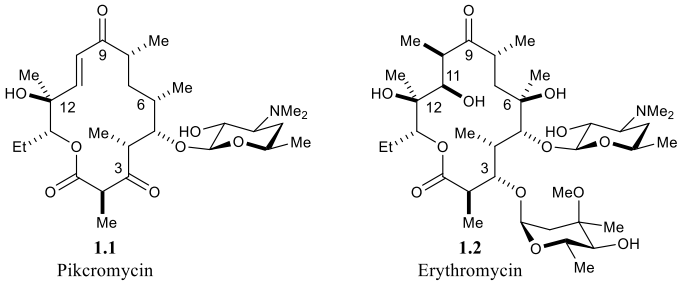


Figure 1.1 Chemical structures of pikromycin (**1.1**) and erythromycin (**1.2**)

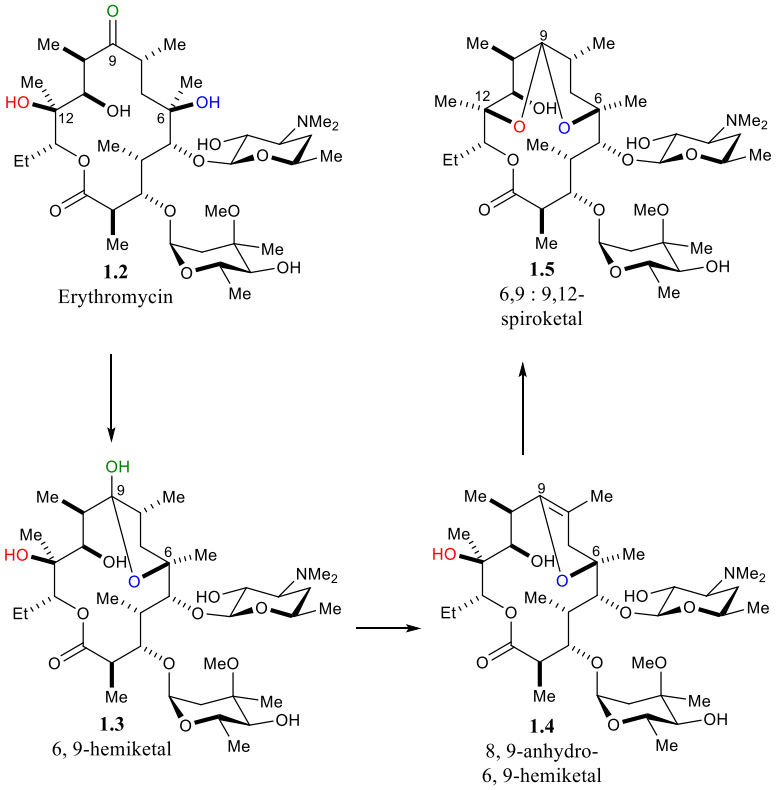
1.2 Erythromycin: The First-Generation Macrolide Antibiotic

The first 14-membered macrolide, erythromycin A (**1.2**) was isolated from the actinomycete *Streptomyces erythreus*, which was later renamed *Saccharopolyspora erythraea*. Due to the slight difference in the structure produced by *Streptomyces erythreus*, there are other minor congeners (erythromycin B-F). However, erythromycin refers to erythromycin A, which is the most abundant and the most potent. Erythromycin consists of a 14–member macrocyclic lactone ring containing two pendant sugars, 3-*O*-L-cladinose attached via an α-glycosidic linkage and and 5-*O*-D-desosamine attached via a β-glycosidic linkage.

Erythromycin has an antimicrobial spectrum similar to that of penicillin and was widely used for patients allergic to penicillin. It has been in clinical use since 1952 and is one of the most effective antibiotics against Gram-positive bacteria. Erythromycin has been used in the treatment of lower and upper respiratory tract infections, as well as skin and soft tissue infections. While effective as an antibacterial agent, it shows negative side effects such as low bioavailability, stomach pain, a short *in vivo* half-life, and most importantly, it was found to be unstable under acidic conditions (i.e., the pH of the gut).

Studies carried out by scientists at Abbott on the chemical stability of erythromycin under acidic conditions revealed it decomposes by sequential intramolecular cyclization reactions.[5] At low pH, erythromycin is first converted to a hemiketal by the attack of the C6 hydroxy group onto the protonated C9 ketone, leading to the formation of 6,9-hemiketal **1.3**. Loss of water leads to 8,9-anhydro-6,9-hemiketal **1.4** and 6,9-9,12-spiroketal **1.5** derivatives. To overcome instability to gastric acid, an enteric-coated tablet used which helped to improve, however, innovative chemical solution was much desired.

Scheme 1.1 Acid-catalyzed decomposition of erythromycin



1.3 Second-Generation Analogs of Erythromycin

The second-generation analogs of erythromycin [i.e., clarithromycin (**1.6**) and azithromycin (**1.7**)] were prepared semi-synthetically from erythromycin in the 1980s, and were developed to address the physicochemical shortcomings of their first-generation predecessor (Figure 1.2). Inspired by the work of Abbott scientists, two different strategies were undertaken to address the issue of acid-mediated ketalization shown in Scheme 1.1.

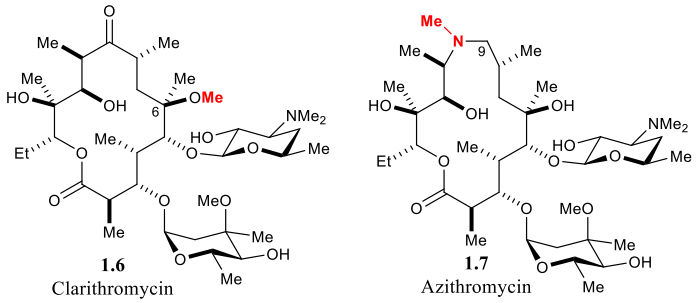


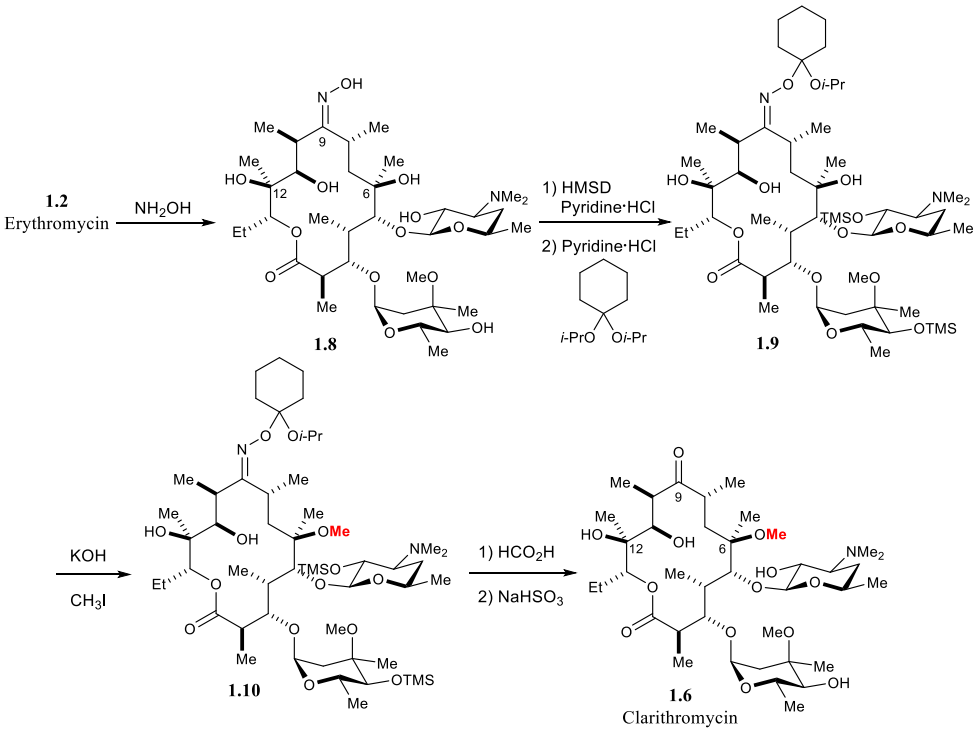
Figure 1.2 Structures of second-generation macrolide antibiotics clarithromycin (**1.6**) and azithromycin (**1.7**)

1.3.1 Clarithromycin

Clarithromycin (6-*O*-methyl erythromycin, **1.6**) was developed by Taisho Pharmaceuticals who were interested in the role of the hydroxyl groups of erythromycin.[6] Strategically, they sought to preclude the initial cyclization event at the C6 hydroxyl by blocking nucleophilic attack onto the ketone. Tactically, this was realized by alkylating the alcohol to prepare an ether derivative. Systematic screening of various 6-*O*-ether analogs indicated that 6-*O*-methyl erythromycin (**1.6**) was superior to other *O*-alkylated analogs. Because of the poor chemoselectivity of methylating the 6-OH in the presence of the many

hydroxyl groups on the erythromycin scaffold (including the 3'-dimethylamino functionality on desosamine), direct synthesis of clarithromycin was problematic. Later, it was found that introduction of a 9-oxime ether reversed the chemoselectivity of hydroxyl group methylation from 11-OH to 6-OH (Scheme 1.2).

Scheme 1.2 Semi-synthesis of clarithromycin from erythromycin



Reaction of the C9 ketone in erythromycin with hydroxylamine gave oxime **1.8**. Protection of the free alcohols on the sugar residues as their TMS ethers with 1,1,1,3,3,3-hexamethyldisilazane (HMDS) followed by treatment with the diisopropyl ketal of cyclohexanone and Py•HCl furnished **1.9**. Treatment of **1.9** with KOH and MeI allowed for the chemoselective methylation of the C6-OH. Removal of the protecting groups in

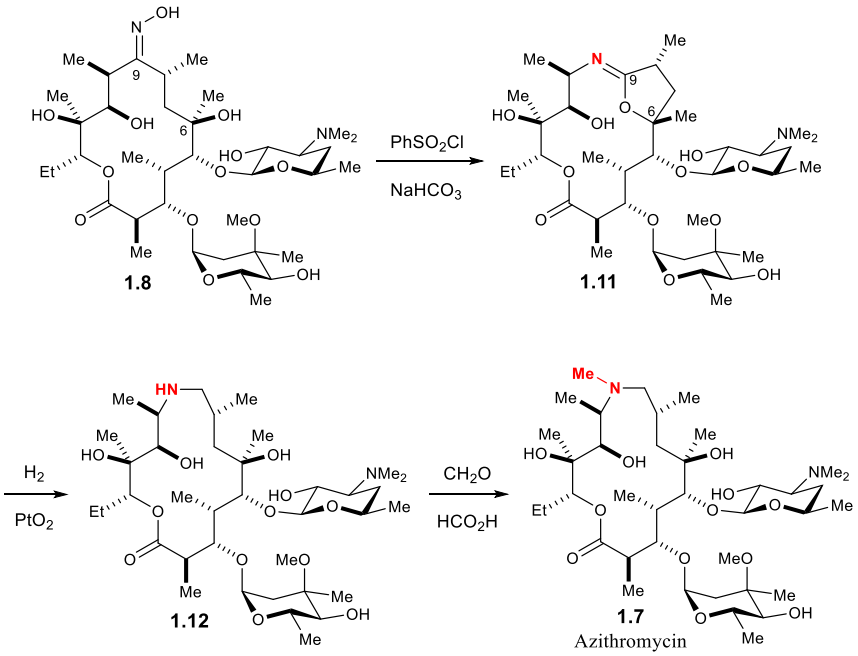
1.10 under acidic conditions gave clarithromycin (**1.6**). The addition of the methyl group in **1.6** endowed it with a slightly expanded spectrum of activity relative to erythromycin (**1.2**) by blocking the formation of the 6,9-hemiacetal. Clarithromycin exhibited antibacterial activities against both Gram-positive and some Gram-negative bacteria. The antibacterial activities of clarithromycin were equal to or two-fold better than erythromycin. It was found to be acid-stable and orally active. The structural difference between clarithromycin and erythromycin is only the 6-*O*-methyl group, but the biological properties of clarithromycin were improved considerably.

1.3.2 Azithromycin

Another innovative semi-synthetic solution from erythromycin was azithromycin (**1.7**), which was developed by Pliva in 1980.[7] Strategically, the Pliva team chose to address the ketalization issue by altering the electrophile as opposed to the nucleophile, as was the case with Taisho. Tactically, the C9 ketone was replaced with a markedly less electrophilic amide. To accomplish this goal, erythromycin was subjected to the following reaction sequence: (1) oxime formation, (2) ring-expansion by means of a Beckmann rearrangement, (3) hydrogenolysis of the resulting imino ether intermediate, and (4) *N*-methylation (Scheme 1.3). This modification in the macrolide ring structure led to the development of a new subgroup of macrolides, the azalides. Azithromycin was the first azalide on the market, and it displays the best antibacterial activity among other azalides. It exhibited reduce activity against Gram-positive bacteria but showed better activity against Gram-negative bacteria such as *E. coli* and *H. influenza*, which extended its

antimicrobial spectrum to Gram-negative bacteria. In comparison with erythromycin, it has improved acid stability, increased oral bioavailability, longer half-life, higher intracellular concentration, and broader antibacterial activity. The conformation of the macrolactone ring in **1.7** proved to be very similar to **1.2** by NMR.[8] This implies that the ring-expansion and the introduction of an amine moiety does not significantly alter the ring conformation between **1.7** and **1.2**, but the ring expansion has a pronounced effect on the antibacterial spectrum.

Scheme 1.3 Semi-synthesis of azithromycin (**1.7**)



Despite improvements in pharmacokinetic properties, however, this generation failed to address the emerging resistance to antibiotics. The discoveries of second-generation macrolide antibiotics clarithromycin (**1.6**) and azithromycin (**1.7**) are excellent

examples of mechanism-based drug development by medicinal chemists that also show creativity and innovation in solving problems that have significant consequences.

1.4 Mechanism of Action

Macrolides inhibit bacterial protein synthesis by binding to the 50S ribosomal subunit and ultimately inhibiting microbial growth. The ribosome is composed of two subunits 30S (i.e., small, 16S rRNA and 20 proteins) and 50S (i.e., large, 5S, 23S rRNA and 34 proteins) built with RNA and proteins, which assemble to produce a functional macromolecule capable of carrying out protein synthesis in every living cell. Each part undertakes a specific function. The small subunit 30S decodes messenger RNA (mRNA). In the large 50S part, the protein is formed by the polymerization of amino acids according to the genetic code. Transfer RNA (tRNA) molecules carry the amino acids. Ribosomes possess three tRNA binding sites: A (acceptor), where the aminoacyl tRNA binds; P (peptidyl), where the peptidyl-tRNA is bound before the formation of the peptide bond; and, E (exit), where the tRNA exits. Each elongation cycle involves the advancement of the mRNA together with A → P → E site passage of the tRNA molecule driven by GTPase activity.[9]

The 50S subunit is formed in part by 23S rRNA which is organized into six domains. The domain V loop called peptidyl transferase center (PTC) contains the active site of peptide bond formation.[10] This PTC loop is positioned at the bottom of a cavity located at the interface of the two subunits, adjacent to the entrance of the peptide tunnel. This tunnel crosses the 50S subunit and emerges on the back of the ribosome. The bacterial

ribosome is the target of macrolides which exert their antimicrobial effects by blocking protein synthesis.

Macrolides of the erythromycin class (e.g., erythromycin, clarithromycin, and azithromycin) do not block peptidyl transferase activity. Extensive structural studies of complexes of these macrolides with the 50S subunit have shown that they block the exit tunnel that channels the growing peptide away from the peptidyl-transferase center. [11]

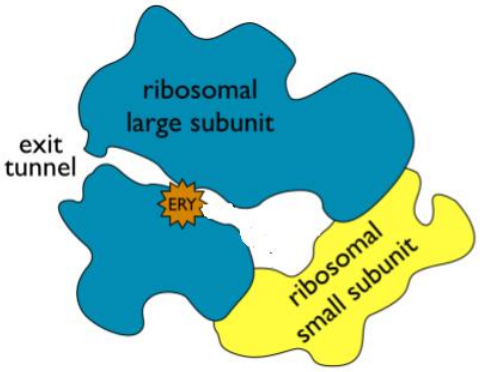


Figure 1.3 The macrolide binds to the exit tunnel

1.5 Macrolide Antibiotic Resistance

Resistance to macrolides was detected when erythromycin was released for clinical use in the early 1950s. The most common mechanisms for macrolide resistance include ribosomal mutation, ribosomal methylation, and efflux mechanism.[12]

Studies with the mutants have revealed that several structures participating in the binding of macrolides, domains V and II of 23S rRNA and proteins L4 and L22, can display mutations responsible for macrolide resistance. The resistance phenotype conferred by alterations in the ribosomal target varies according to the nature of the mutated structure.

Without direct contact with the drug, their mutation effectively alters the macrolide binding site. The most common mechanism of antibiotic resistance is through the modification of the target – the bacterial ribosome. A base substitution in the 23S rRNA where macrolides make several contacts with the ribosome is the most common, and the residue most often associated with this mutation is Adenine at position 2058 (*E. coli* numbering or A2058). Resistance by methylation of an adenine residue in domain V of the 23S rRNA is mediated by the erythromycin ribosome methylase (erm) genes. Methylation prevents binding of the macrolides to domain V and results in high-levels macrolide resistance.[13]

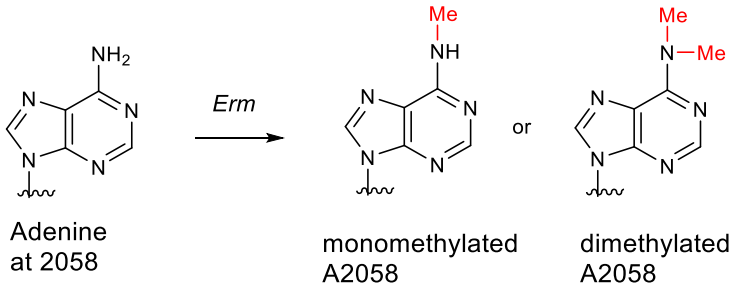


Figure 1.4 Ribosomal Methylation

The overlapping binding sites of macrolides, lincosamides, and streptogramins B in 23S rRNA account for cross-resistance to the three classes of drugs, which gave its name to the MLS_B resistance phenotype. A wide range of microorganisms that are targets for macrolides and lincosamides express Erm methylases. The third mechanism of resistance to macrolides is through the production of efflux proteins that pump the antibiotics out of the cell, of which there are two types. One set of proteins, coded by the macrolide efflux (mef) gene, is found in *Streptococcus spp*. The second set of proteins, coded by the msr

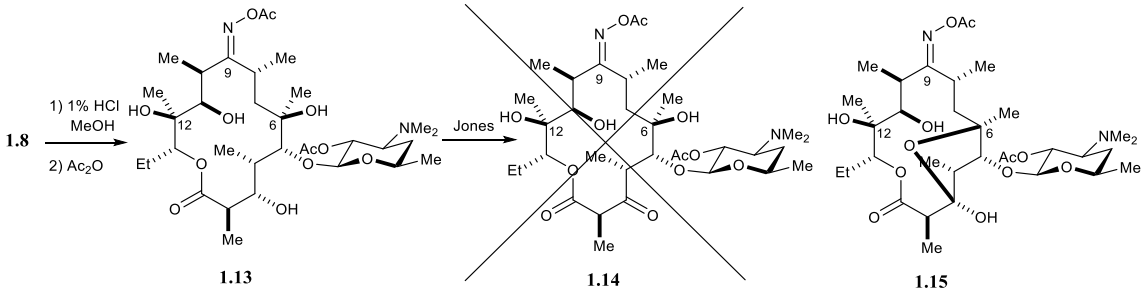
gene, belongs to the ABC transporter family but has been isolated so far only from *Staphylococcus spp*.

1.6 Thrid-Generation Analogs of Erythromycin

Despite the fact that second-generation of erythromycin (i.e., clarithromycin and azithromycin) showed improvements in pharmacokinetic properties, this generation failed to address the emerging resistance to the macrolide class of antibiotics. Thus, significant efforts were directed in the 1990s toward the development of macrolides that addressed the aforementioned mechanisms of resistance, ushering in the third-generation of analogs based on erythromycin.

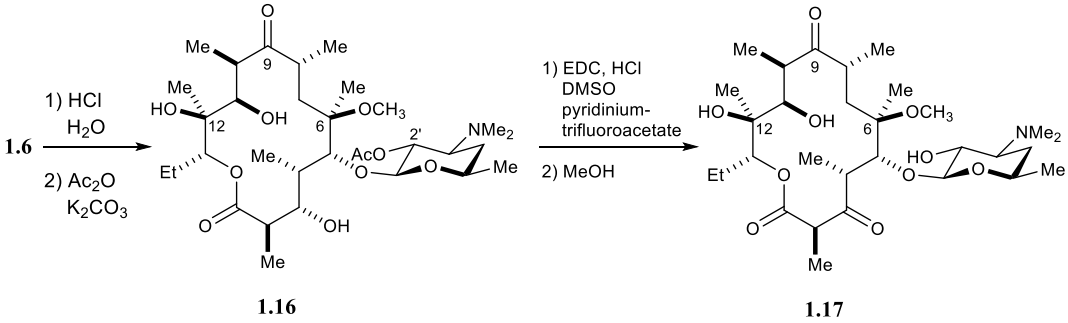
It was long believed that the cladinose sugar was, in fact, crucial for the antibacterial activity, as well as the resistance-inducibility properties of erythromycin. However, Allen showed that macrolides such as pikromycin (**1.1**), which do not contain cladinose, failed to induce macrolide resistance yet possessed antibacterial properties.[14] These studies showed that cladinose was, in fact, not essential for antibacterial activity. This finding ultimately led to the investigation and development of ketolides by Hoechst-Marion-Roussel (later Aventis). In 1998, they disclosed a series of 6-*O*-Methyl-3-oxoerythromycin derivatives), and this novel class of macrolides were termed "ketolides" based on the presence of the C3 ketone functionality.[15] Originally, 3-keto erythromycin **1.14** was prepared from oxime derivative **1.13**; however, **1.14** was not isolable to the spontaneous formation of C3,6-hemiacetal **1.15**.[16]

Scheme 1.4 Attempted synthesis of 3-keto erythromycin from **1.8**



To avoid ketalization at C-3, 6-*O*-methyl erythromycin (clarithromycin) was used as a starting material. Hydrolysis of the cladinose followed by acetylation of the 2′-OH desosamine gave the 3-hydroxy intermediate **1.16**. Oxidation of the C3 hydroxyl with a modified Pfitzner-Moffat procedure (1-(3-(dimethylamino)propyl)-3-ethylcarbodiimide hydrochloride (EDC, HCl)-DMSO, pyridinium trifluoroacetate) instead of Jones's reagent, which gave a complex mixture of inseparable products, smoothly introduced the requisite 3-keto functionality.

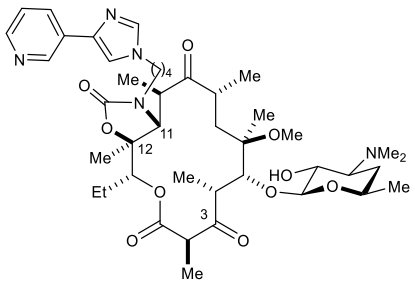
Scheme 1.5 Synthesis of 3-Keto erythromycin from **1.6**



A number of ketolides were prepared from **1.17** with a variety of substituents such as 9-keto, 9-oxime, 6-*O*-alkyl, C11,12-cyclic carbonate, C11,12-cyclic carbamate, C11,12-

cyclic carbazate, and their combinations. Overall, the ketolides showed improved acid stability, antibacterial activity, and pharmacokinetics.

The third-generation derivatives [e.g., telithromycin (**1.18**) and cethromycin (**1.19**)] were developed to address resistant pathogens. Several distinct modifications were introduced. The C-3 cladinose was removed then oxidized to a ketone. A biaryl side chain, which was designed to make additional contacts with the macrolide-binding site within the ribosome, was installed via an oxazolidinone at C11,12, and C6 alcohol was methylated as it was derived from clarithromycin.



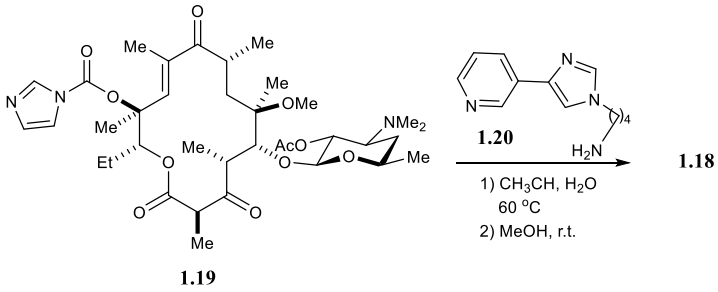
1.18

Figure 1.5 Third-generation macrolide antibiotic telithromycin **(1.18**)

1.6.1 Telithromycin

Telithromycin (HMR-3647) was synthesized by the French pharmaceutical company Hoechst-Marion-Roussel in 1988. The C11,12 oxazolidinone-containing ketolides bearing heterocyclic butyl substituents at N11 were synthesized from 10,11-anhydro-12-*O*-imidazolyl carbonyl ketolide precursor **1.19** (Scheme 1.6).[17]

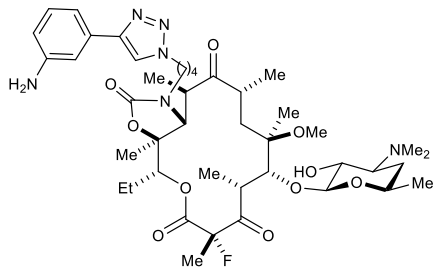
Scheme 1.6 Final steps in the synthesis of telithromycin (**1.18**)



The side chain at the C11,12 position of the ketolides interacts with domain II, at A752, of the 23S RNA and provides a second binding site of telithromycin, thus confering activity against macrolide-resistant strains.[18] Simultaneous interaction both with domain V and domain II strengthens binding of the drug to resistant ribosomes, making telithromycin a potent drug against macrolide-resistant pneumococci. Telithromycin binds to the bacterial ribosome with high affinity and kills bacteria, while older macrolides stop bacterial reproduction. In 2004, telithromycin received FDA marketing authorization. When first approved by the FDA, telithromycin was indicated for use in treating numerous bacterial infections. However, due to safety issues with the drug, the FDA removed its approval to treat some infections and issued a black box warning.[19] Telithromycin is the first ketolide antibiotic to enter clinical use. Despite effectiveness against many pathogens that eluded previous erythromycin derivatives, the third-generation is far from perfect. Adverse effects, notably hepatotoxicity, have been reported for telithromycin, and susceptible pathogens eventually develop resistance.

1.7 Fourth-Generation Analogs of Erythromycin

Solithromycin (**1.20**) is a novel fluoroketolide which contains a novel 5-membered 1,2,3-triazole ring that was introduced via copper-catalyzed Click chemistry and marketed by Cempra Pharmaceuticals.[20] Its activity against macrolide-resistant organisms is enhanced by the addition of a side chain aniline moiety and a 1,2,3-triazole, which provides multiple ribosomal interactions. This allows tighter binding to the ribosome, thus overcoming the resistance caused by methylation of the 23S rRNA in the 50S ribosomal subunit. In addition, the fluorine at the C2 position of the 14-membered macrocyclic lactone allows for tighter binding, resulting in lower MICs. Additional details regarding solithromycin (**1.2**) is found in Chapter 4.



1.20
Solithromycin

Figure1.6 Fourth-generation macrolide antibiotic solithromycin (**1.20**)

1.8 Desmethyl Telithromycin – An Approach to Address Antibiotic Resistance

This work was inspired from Steitz and co-workers, who demonstrated that ribosomal mutation of adenine to guanine at position 2058 resulted in a steric clash between the C4 methyl group of the macrolide/ketolide and the exocyclic amino group of guanine (Figure 1.7-A,B).[21] The distance between C4 methyl and N2 residue G2058 is 2.8Å, which

could sterically crash disrupting the binding interaction. Accordingly, the Andrade laboratory hypothesized that 'mutating' the C4 methyl group into hydrogen (i.e., desmethyl analogs) would rescue binding and restore bioactivity against such resistant strains. Based on structure-based drug design, the Andrade laboratory targeted four desmethyl analogs including desmethyl analogs at the C8 and C10 positions (Figure 1.7-C).

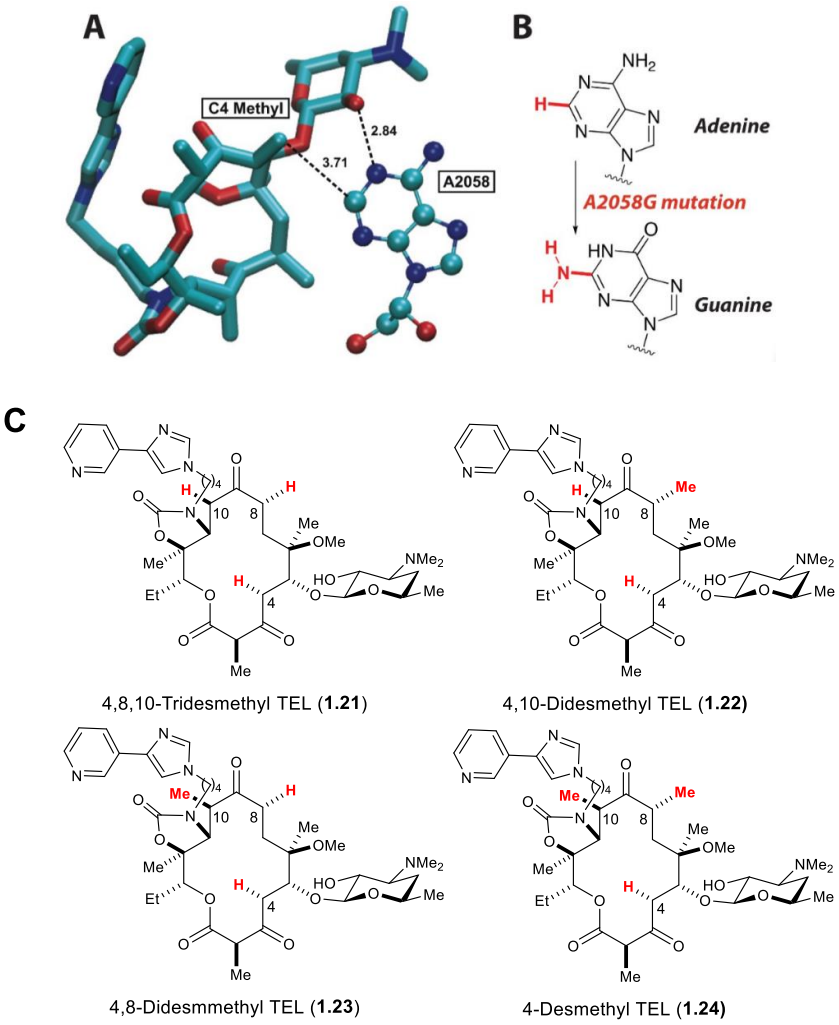


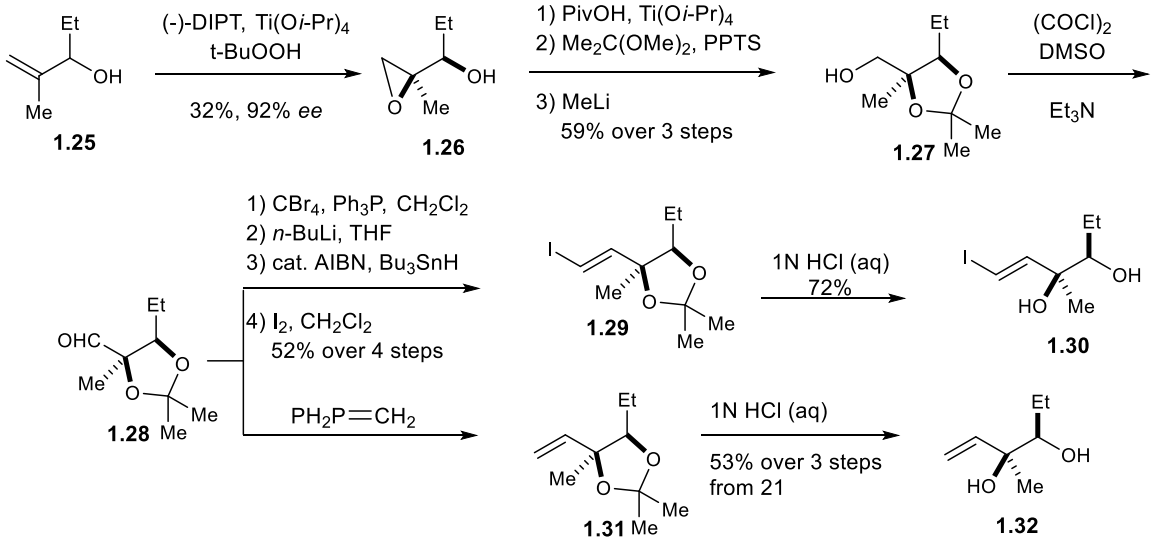
Figure 1.7 (A) Telithromycin (**1.18**) and A2058 interactions in *H. marismortui* with select distances in Angstroms; (B) consequences of A2058G mutation; (C) **8** Structures of four desmethyl analogs of telithromycin

1.8.1 Total Synthesis of (–)-4,8,10-Tridesmethyl Telithromycin

Dr. Venkata Velvadapu competed for the total synthesis of (–)-4,8,10-tridesmethyl telithromycin (**1.21**) in 2011. A total of 12.1 mg of analog **1.21** was prepared in 42 steps (31 steps in the longest linear sequence), which was active against several wild types and resistant bacterial strains.[22]

The synthesis began with preparing **1.30** and **1.32**. The preparation of fragments **1.30** and **1.32** was accomplished from aldehyde **1.28**, which was synthesized from **1.25** (Scheme 1.7). Kinetic resolution of racemic **1.25** via the Sharpless asymmetric epoxidation protocol provided enantioenriched alcohol **1.26** in 32% yield (92% ee). Regioselective ring-opening, acetonide formation, and treatment with MeLi afforded alcohol **1.27** (59% over three steps). Swern oxidation of **1.27** to aldehyde **1.28** proceeded without incident. At this point, **1.28** was subjected to the Corey-Fuchs alkynylation protocol flowed by

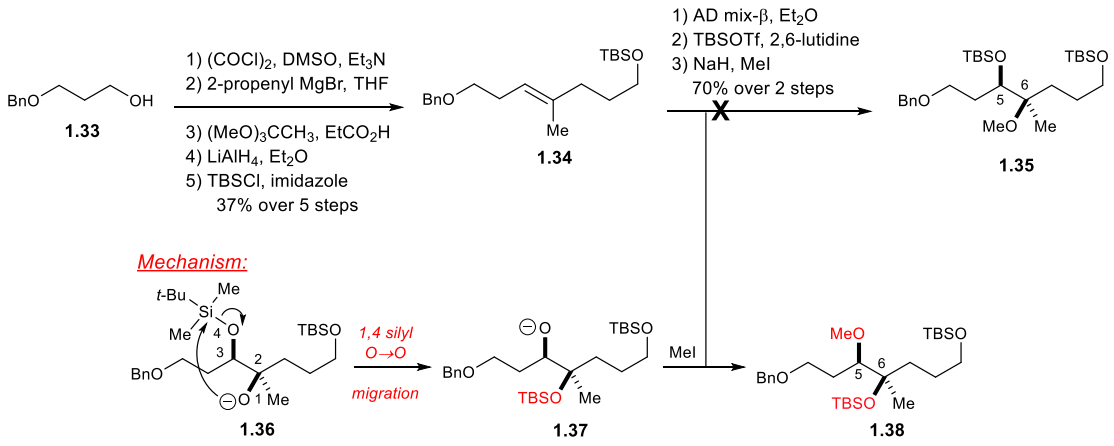
Scheme 1.7 Synthesis of fragments **1.30** and **1.32** from aldehyde **1.25**



hydrostannation of the intermediary alkyne with Bu_3SnH and catalytic AIBN to afford an (*E*)-vinyl stannane. The stannane was converted to vinyl iodide **1.29** with I_2 in 52% over four steps. Removal of the acetonide in **1.29** with 1 M HCl (aq) furnished fragment **1.30**. Access to fragment **1.32** was accomplished by Wittig methylenation and acetonide removal under acidic condition.

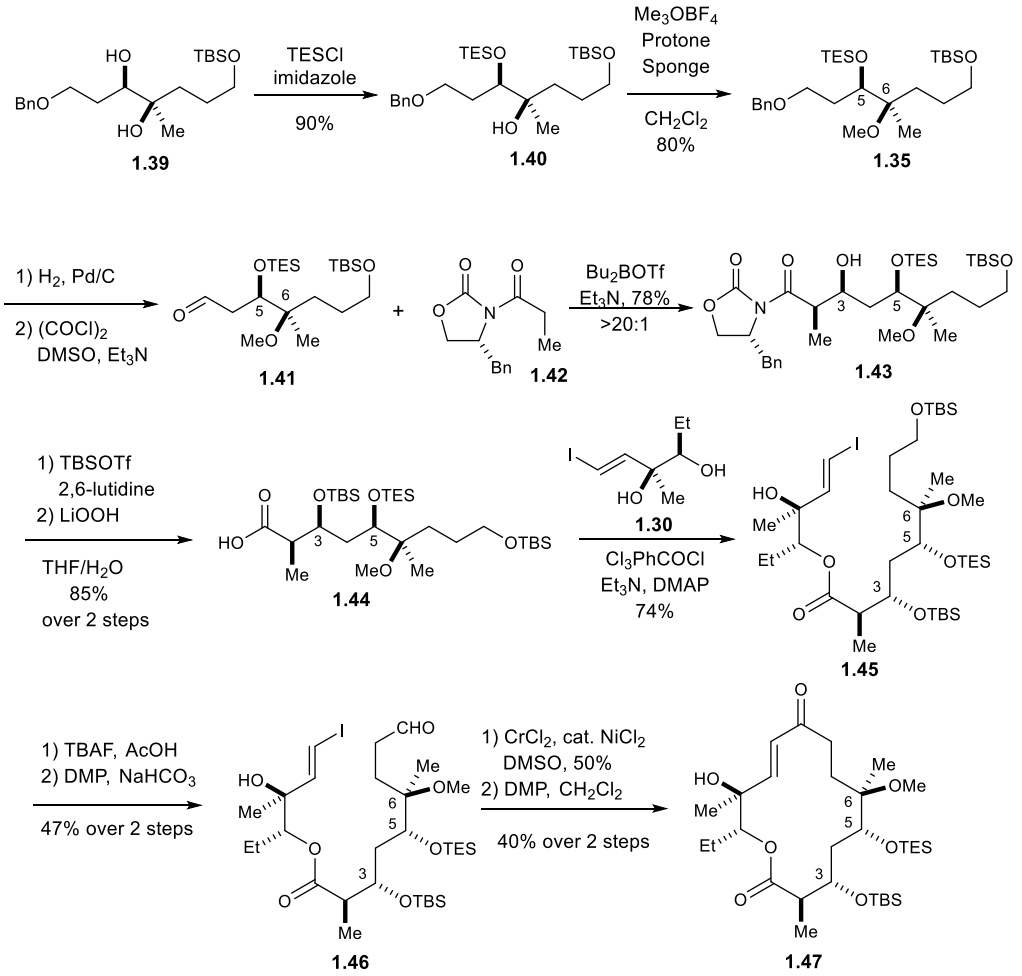
Right-handed fragment started from commercially available 3-benzyloxy-propanol, which was oxidized using the Swern protocol (Scheme 1.8). The addition of 2-propenyl MgBr and subsequent Johnson-Claisen orthoester rearrangement afforded enoate, which was reduced and protected as its *tert*-butyldimethylsilyl (TBS) ether to provide **1.34**. Sharpless dihydroxylation (AD mix-β) established the stereochemistry of the hydroxyls at C5 and C6. Selective protection of the secondary C5 alcohol with TBS group followed by treatment with NaH and MeI resulted in the formation of **1.38** as opposed to the desired regioisomer **1.35** via a 1,4-*O*→*O* silyl migration.

Scheme 1.8 Unexpected synthesis of regioisomeric fragment **1.38**



To resolve this issue, the more reactive methylating agent (e.g., Meerwein's salt) and Proton Sponge was employed to afford desired regioisomer (Scheme 1.9). In addition, the TES protecting group established at C5 was deprotected selectively in the presence of C3 TBS group. Removal of the benzyl ether afforded an alcohol that was subjected to the Swern protocol to furnish aldehyde **1.41.** The Evans aldol reaction furnished **1.43** bearing stereocenters at C2 and C3. Protection of the C3 alcohol with TBSOTf and removal of

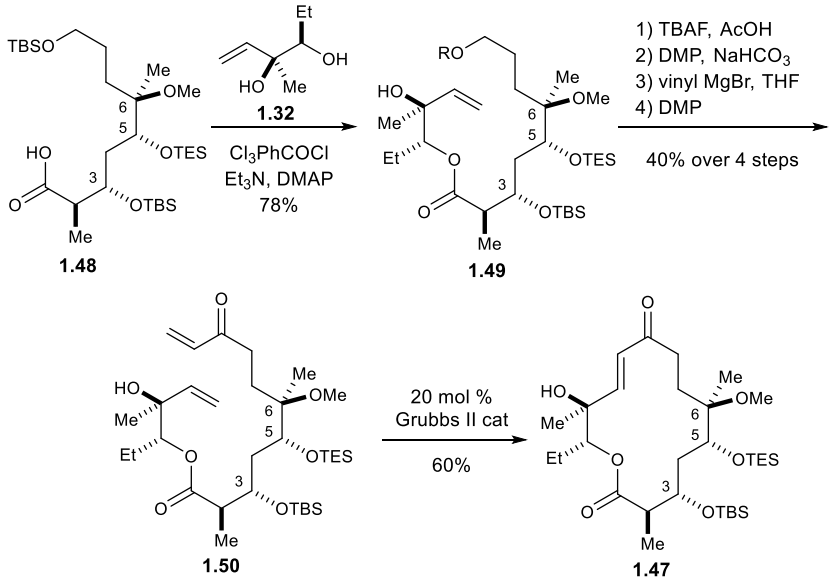
Scheme 1.9. Synthesis of the desired regioisomer and Intramolecular Nozaki-Hiyama-Kishi (NHK) Route to macroketolactone **1.47**



the auxiliary with LiOOH gave acid **1.44** in 85% yield over two steps. Yamaguchi esterification of **1.44** and **1.30** delivered ester **1.45** in 74% yield. Removal of the primary TBS group and subsequent DMP oxidation set the stage for intramolecular Nozaki-Hiyama-Kishi (NHK) coupling, which yielded C9 alcohol. Oxidation of the ~1:1 diastereomeric mixture of allylic alcohols at C9 furnished desired macroketolactone **1.47**.

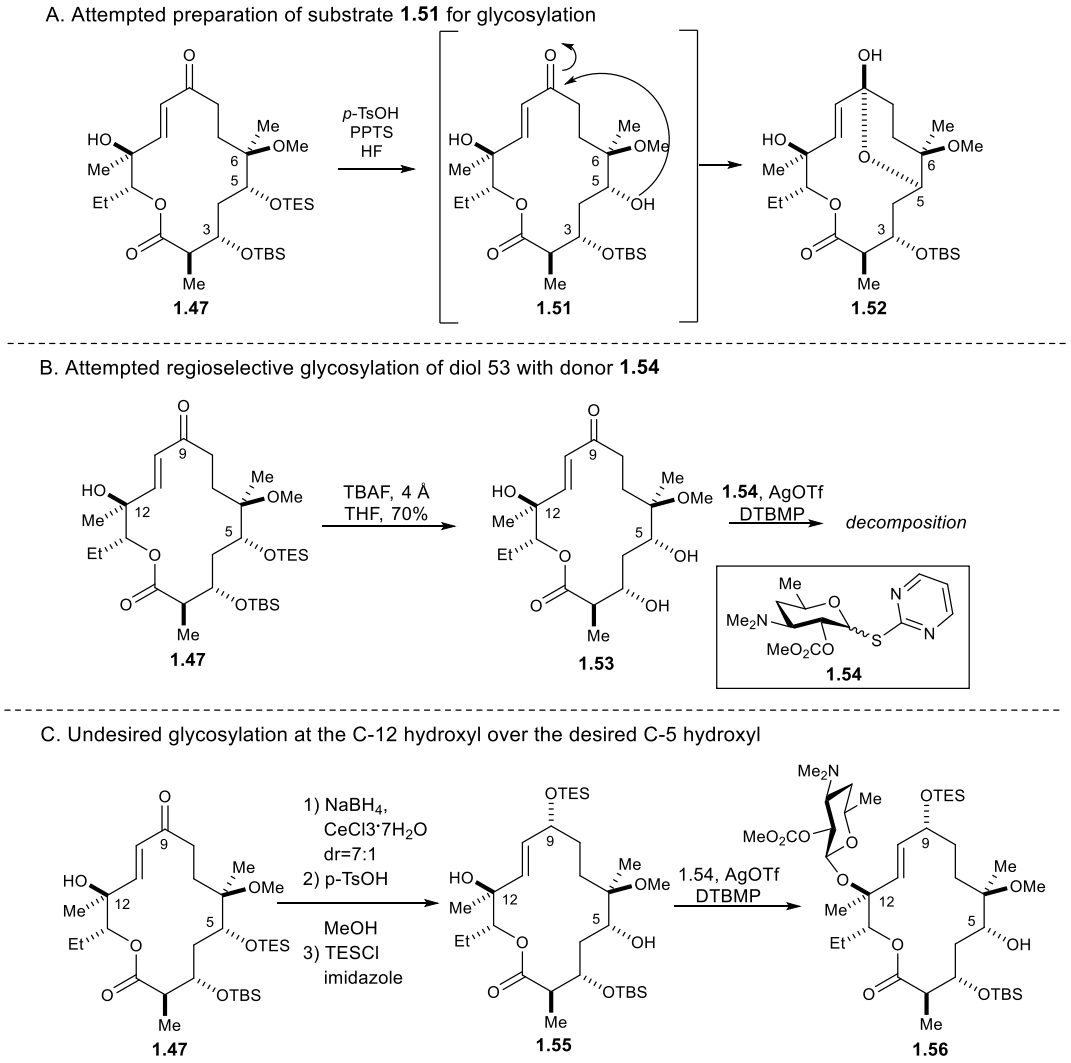
In parallel, a scalable ring-closing metathesis (RCM) route was developed that was inspired by Kang's synthesis of narbonolide (Scheme 1.10).[23] Chemoselective Yamaguchi esterification of acid **1.48** and diol **1.32** accessed ester **1.49**. Selective removal of the primary TBS ether, DMP oxidation, treatment with vinyl MgBr, and subsequent re-oxidation with DMP furnished vinyl ketone and RCM substrate **1.50** in 40% over four steps. The RCM reaction proceeded with 20 mol% Grubbs second-generation catalyst to provide **1.47** in 60% yield.

Scheme 1.10 Alternate ring-closing metathesis (RCM) route to macroketolactone **1.47**



Next, attention was directed at the installation of the desosamine moiety onto the C5 hydroxyl. Attempts to remove C5 TES ether under various conditions (e.g., *p*-TsOH,

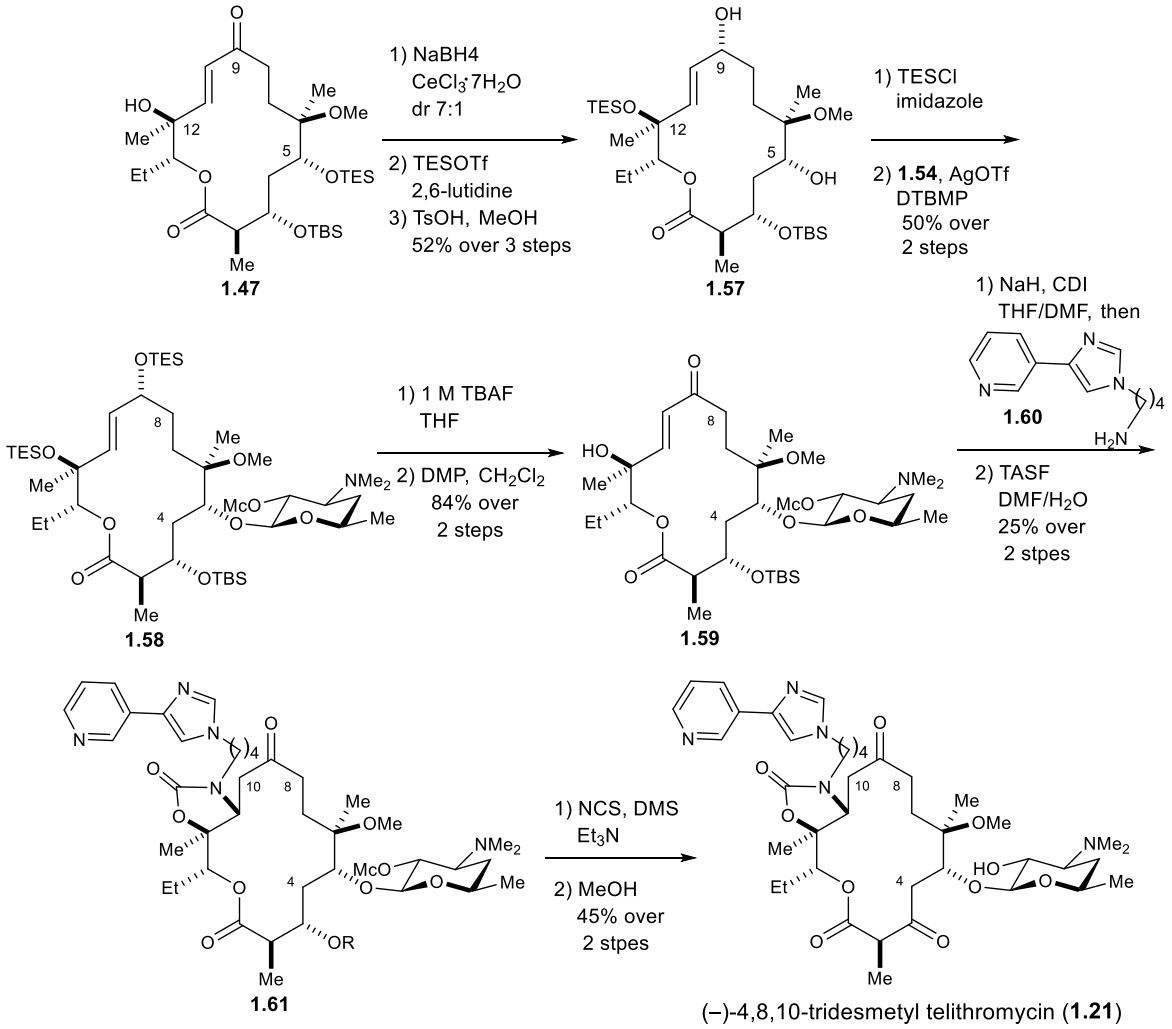
Scheme 1.11 Attempted preparation of substrate **1.51** for glycosylation and attempted glycosylation



PPTS, HF) unfortunately led to ketalization at the C9 position under these acidic conditions (Scheme 1.11-A). Treatment of **1.47** under basic conditions resulted in the removal of both silyl groups, affording triol **1.53** in 70% yield (Scheme 1.11-B). Reported regioselective glycosylation conditions were employed on **1.53** at C5 with donor **1.54**. In the event, AgOTf and DTBMP led to the decomposition of starting material. Recourse at this point was made to a more conservative strategy, which was an installment of desosamine with the C3 position blocked. This would be feasible because the secondary C5 hydroxyl would be more reactive than the hindered tertiary C12 hydroxyl. To test this, the C9 ketone of **1.47** was first subjected to a Luche reduction, followed by treatment with *p*-TsOH in methanol selectively remove the C5 TES ether in the presence of the C3 TBS ether to afford triol. Regioselective TES protection of the more reactive allylic C9 hydroxyl furnished diol **1.55**. Glycosylation of **1.55** resulted in the desosamine at the C12 position, as opposed to the desired C5 and was confirmed by 2D NMR experiments.

The unexpected glycosylation at the more hindered position necessitated protection at this position as well (Scheme 1.12). Macroketolactone **1.47** was first reduced under Luche conditions followed by silylation of both C9 and C12 hydroxyls with the TES group. Treatment with *p*-TsOH selectively removed the C9 and C5 ethers to afford **1.57**. Regioselective silylation at the allylic C9 alcohol and subsequent glycosylation at the C5 position ultimately proved successful. Fluoride-mediated cleavage of silyl ethers at C9 and C12 followed by DMP oxidation at C9 afforded glycosylated macroketolactone **1.59**.

Scheme 1.12 Synthesis of macroketolactone **1.59** and Endgame for **1.21**



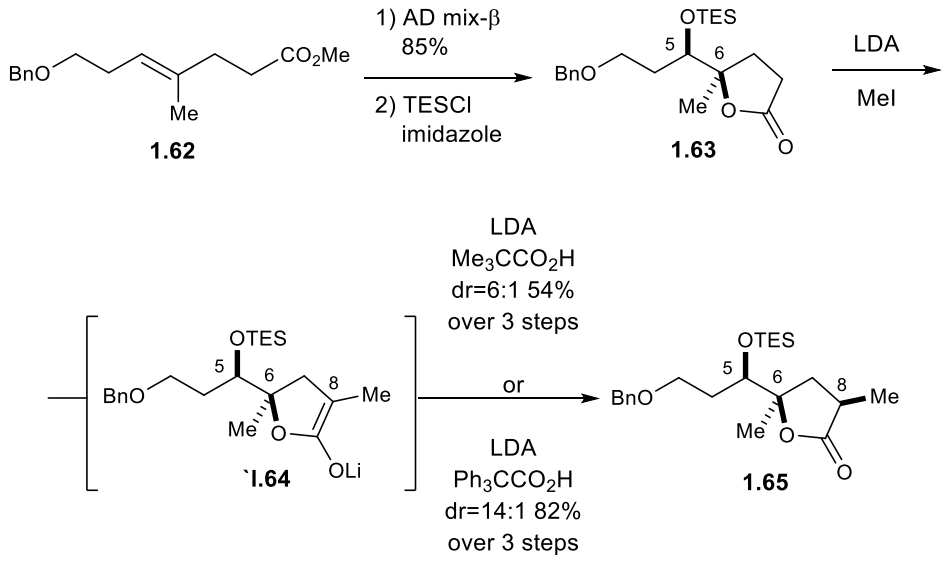
Activation of the C12 alcohol with NaH and carbonyldiimidazole (CDI) followed by treatment with butylamine **1.60** effected a tandem carbamoylation/intramolecular aza-Michael sequence, which was developed by Baker, and afforded oxazolidinone **1.61** selectively. Removal of the C3 TBS ether with TAS-F followed by Corey-Kim oxidation furnished the C3 ketone. Finally, methanolysis removed the methyl carbonate on the C2' hydroxyl of desosamine to deliver (–)-4,8,10-tridesmethyl telithromycin (**1.21**).

1.8.2 Total Synthesis of 4,10-Didesmethyl Telithromycin

Dr. Venkata Velvadapu competed for total synthesis of (–)-4,10-didesmethyl telithromycin (**1.22**) in 2011. It was prepared in 44 steps overall (32 steps in the longest linear sequence).[24] The addition of an extra methyl group at C8 resulted in a 4-fold increase in activity as compared to (–)-4,8,10-tridesmethyl analog **1.21**, demonstrating that methyl groups play an important role in antibiotic function.

Sharpless dihydroxylation (AD mix-β) with **1.62** furnished γ-lactone by *in situ* lactonization. The newly formed C6 hydroxyl was protected with TESCl. Enolate generation with lithium diisopropylamide (LDA) followed by alkylation with MeI afforded an (*S*)-C8 methyl intermediate. Inversion of stereochemistry at C8 to obtain the (*R*) configuration was accomplished by re-enolization and quenching with trimethylacetic acid (dr = 6:1, 54% over 3 steps). Alternatively, quenching the enolate with a bulkier acid source, triphenylacetic acid, improved the selectivity of epimerization (dr = 14:1, 82% over 3 steps). The stereochemistry was confirmed by X-ray analysis. This was the first project that I was involved as a graduate student. Tedious synthesizing circle to generate material was always the motivation to optimize reaction condition. This reaction was based on the kinetic protonation that was introduced in 1954.[25] The face selectivity of a proton approach to the α-carbon is controlled by steric hindrance. The effect is enhanced with large proton donors. When a bulkier proton donor, triphenylacetic acid, was subjected to the reaction, it approached from the bottom face to avoid steric interaction with other functional groups on the top face. The α-carbon was protonated from the less hindered side. This method delivered our desired stereochemistry outcome along with excellent yield.

Scheme 1.13 Installation of (*R*)-C8 Methyl Group and Stereochemical Confirmation



Subsequent steps for synthesizing 4,10-didesmethyl TEL (**1.22**) were guided by the previously reported synthesis of (–)-4,8,10-tridesmethyl analog (**1.21**) using an RCM approach.

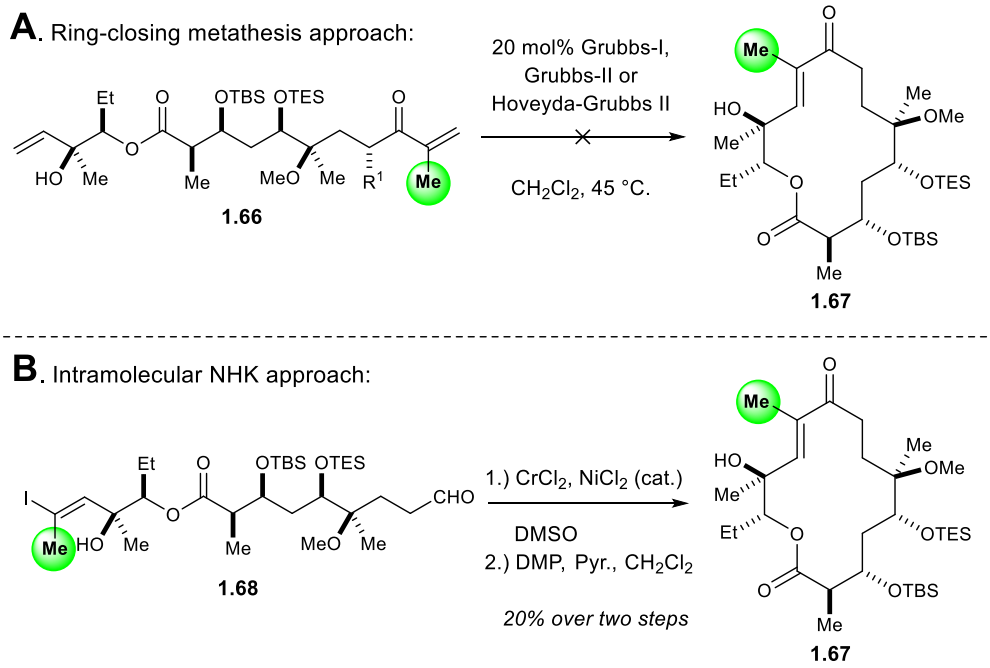
1.8.3 Total Synthesis of 4,8-Didesmethyl Telithromycin

Dr. Bharat Wagh synthesized 4,8-didesmethyl telithromycin (**1.23**) in 2012. It was prepared via an intramolecular NHK approach in 36 steps (26 steps in the longest linear sequence).[26] The addition of an extra methyl group at C10 resulted in an 8-fold increase in activity as compared to the tridesmethyl analog, again underscoring the role that methyl groups play in antibiotic activity.

Based on the previous synthesis of desmethyl analogs, the ring-closing metathesis (RCM) strategy was applied for preparing 14-membered macroketolactone. Despite multiple attempts with different catalysts such as Grubbs' first-generation catalyst and

Hoveyda−Grubbs second-generation catalyst, the reaction was not successful. It was reasoned that the steric congestion about the trisubstituted enone, which was absent in previous analogs, scuttled efforts. An alternate macrocyclization strategy, the intramolecular Nozaki−Hiyama−Kishi (NHK) reaction (Scheme 1.14), was pursued. In the synthesis of (–)-4,8,10-tridesmethyl analog **1.21**, NHK/DMP oxidation furnished macrolactone in 41% overall yield; however, the presence of the C10 methyl group in **1.68** negatively impacted this reaction, and an unsustainable 20% yield was obtained after oxidation.

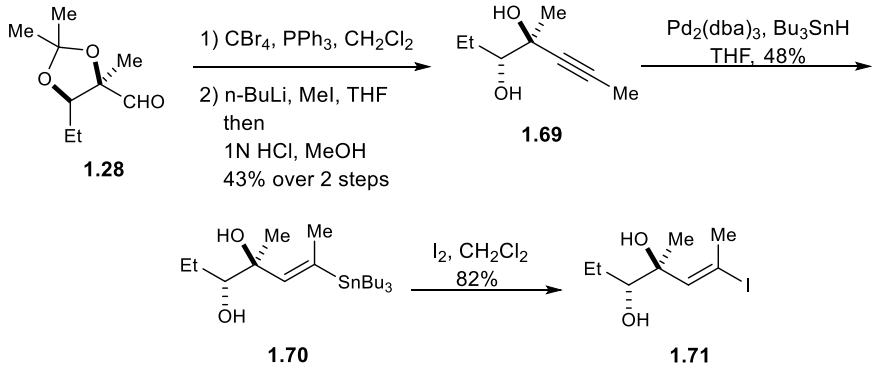
Scheme 1.14 Macrolactonization approaches



Inspired by Martin's abiotic strategy for the synthesis of erythromycin, the decision made to conduct the macrocyclization reaction on a substrate bearing carbohydrate residues. The synthesis began with the preparation of the requisite vinyl iodide **1.71**

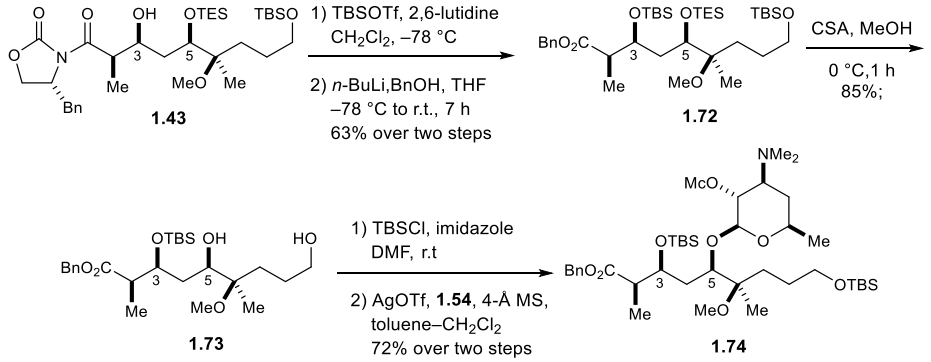
(Scheme 1.15). Corey−Fuchs alkynylation of **1.28**, subsequent trapping with methyl iodide, and removal of the acetonide afforded **1.69**. Palladium-catalyzed hydrostannylation followed by treatment with I_2 furnished vinyl iodide **1.71**.

Scheme 1.15 Synthesis of C10–C13 fragment **1.71** bearing the C10 methyl group

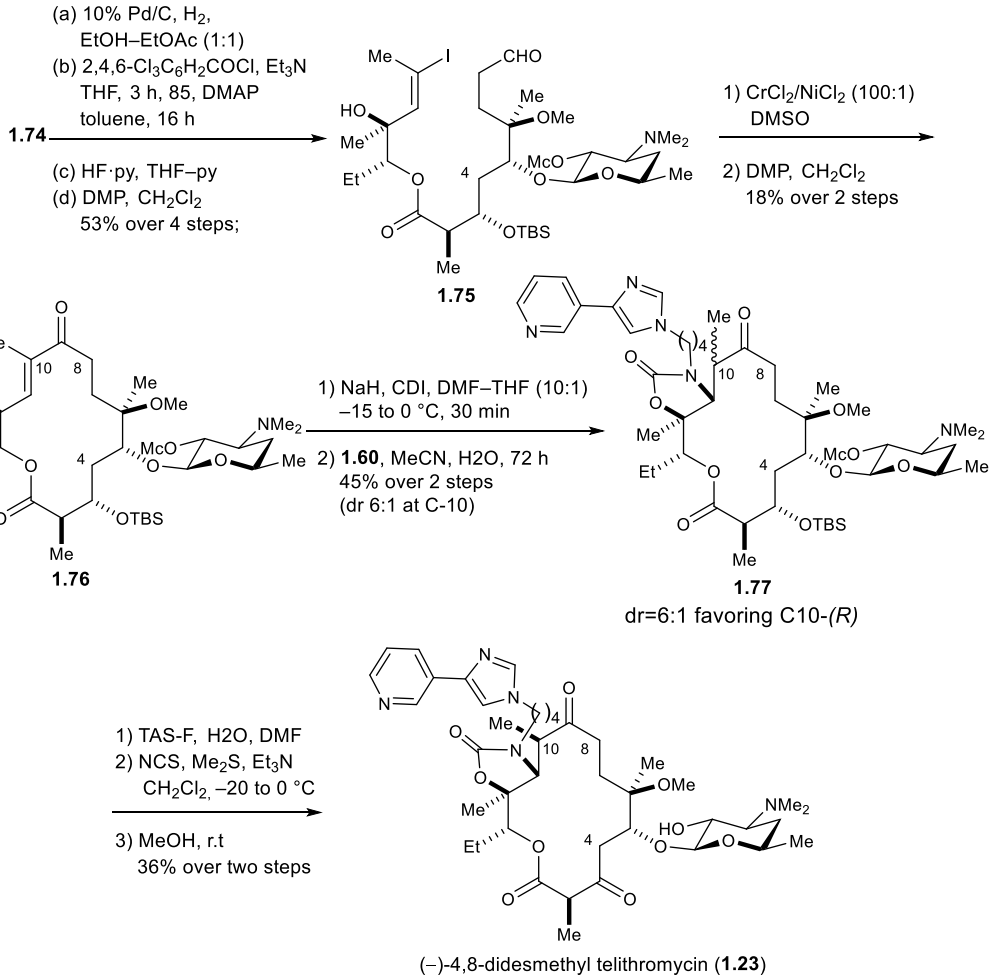


The C1−C9 aldol fragment was prepared by the same manner of tridesmethyl congener **1.21** (Scheme 1.16). Aldol **1.43** was protected as its *tert*-butyldimethylsilyl (TBS) ether, and removal of the Evans auxiliary with LiOBn furnished ester **1.72**. The chemo- and stereoselective installation of the D-desosamine residue required protecting group manipulation. Both C5-OTES and C9-OTBS ethers were removed with camphorsulfonic acid (CSA). Chemoselective reprotection of the primary C9 hydroxyl with TBS group gave the C5 alcohol. Glycosylation with Woodward's desosamine donor **1.54** under the agency of AgOTf furnished **1.74**. Hydrogenation of the benzyl ester in **1.74** afforded the intermediary acid (Scheme 1.17). Chemoselective Yamaguchi esterification with diol **1.71** afforded an ester intermediate. Removal of the primary C9-OTBS group followed by oxidation to the aldehyde furnished NHK substeate **1.75**. Treatment of **1.75** with $CrCl_2$ and

Scheme 1.16 Synthesis of the glycosylated C1–C9 fragment



Scheme 1.17 Endgame of (−)-4,8-tridesmethyl telithromycin (**1.23**)



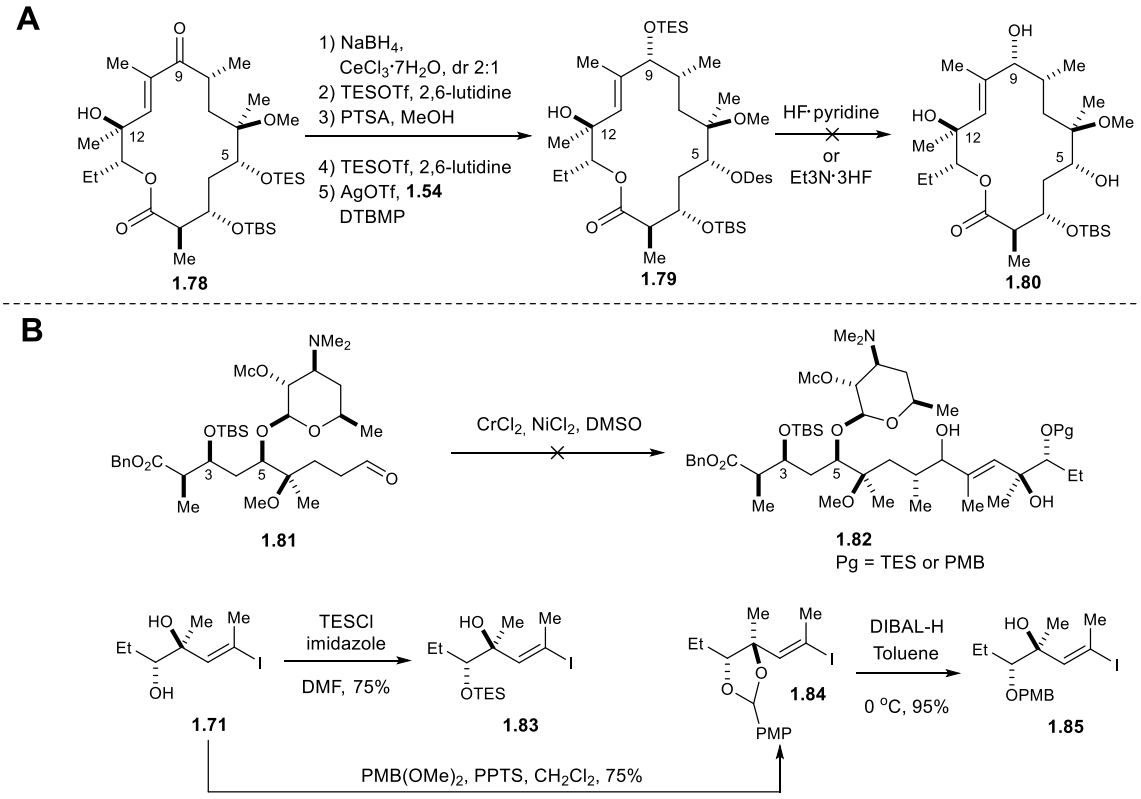
$NiCl_2$ (100:1) in degassed DMSO at rt and DMP-mediated oxidation of the diastereomeric mixture of C-9 alcohols gave macroketolactone **1.76** in 18% yield over two steps. Endgame for the synthesis of **1.23** required installation of the biaryl side chain. One-pot amidation/intramolecular aza-Michael reaction furnished oxazolidinone **1.77** as an inseparable 6:1 mixture of diastereomers at C10 with the major isomer being the desired C10-(*R*) isomer. Removal of the C3-OTBS ether, followed by Corey−Kim oxidation, furnished the C3 ketone. Finally, methanolysis removed the methyl carbonate to deliver (–)-4,8-tridesmethyl telithromycin (**1.23**).

1.8.4 Total Synthesis of 4-Desmethyl Telithromycin

In 2013, Dr. Ian Glassford synthesized (–)-4-desmethyl telithromycin (**1.24**). It was prepared using an intermolecular NHK/Yamaguchi macrolactonization approach in 36 steps (26 steps in the longest linear sequence).[27] Evaluation of the biological activity of **1.24** via MIC assays revealed it was equipotent with comparator telithromycin against an *E. coli* WT strain but was 4-fold less potent against the A2058G mutant. MD simulations were employed to help rationalize these results and revealed **1.24** adopts a slightly different conformation once bound to the A2058G ribosome, thus impacting noncovalent interactions reflected in a lower MIC value.

Glycosylation of the macroketolactone was inspired by the 4,10-didesmethyl telithromycin (**1.22**) protocol. Luche reduction provided a 2:1 mixture of diastereomers. Protecting group manipulation followed by glycosylation furnished **1.79**. Unfortunately,

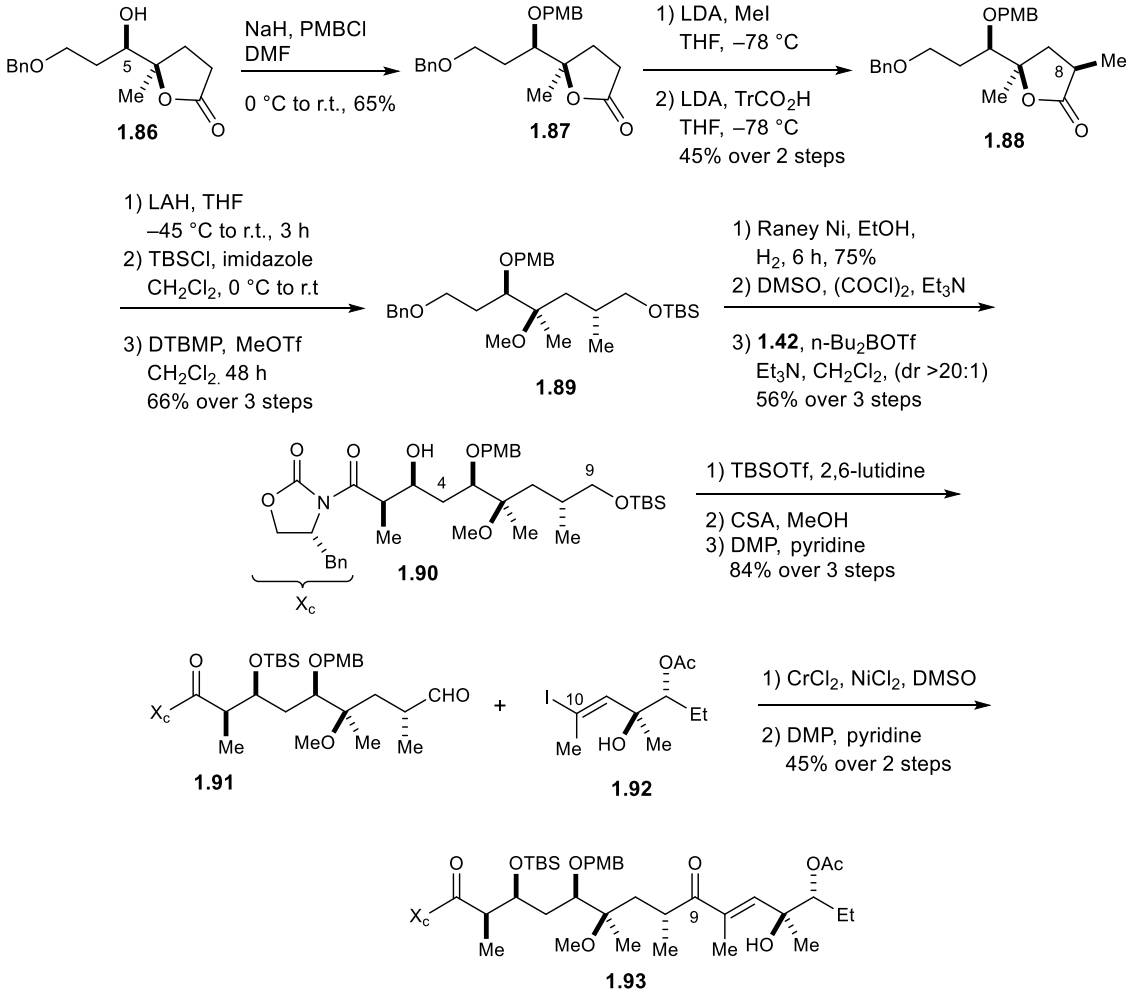
Scheme 1.18 Attempted reactions prior to the third generation



removal of the TES groups with HF was unsuccessful (Scheme 1.18-A). The next approach taken was the installment of desosamine prior to NHK coupling, which paralleled the route to 4,8-didesmethyl telithromycin **1.23**. To carry out the NHK reaction, various protecting groups for the vinyl iodide that would not require saponification to remove were surveyed. Vinyl iodide coupling partners **1.83** and **1.85** were synthesized to introduce different protecting groups, specifically TES and PMB. Unfortunately, none of the coupling partners with aldehyde **1.81** was suitable to do NHK reaction to provide **1.81**.

To access the target, a new C5 protecting group was required. Protection of the C5

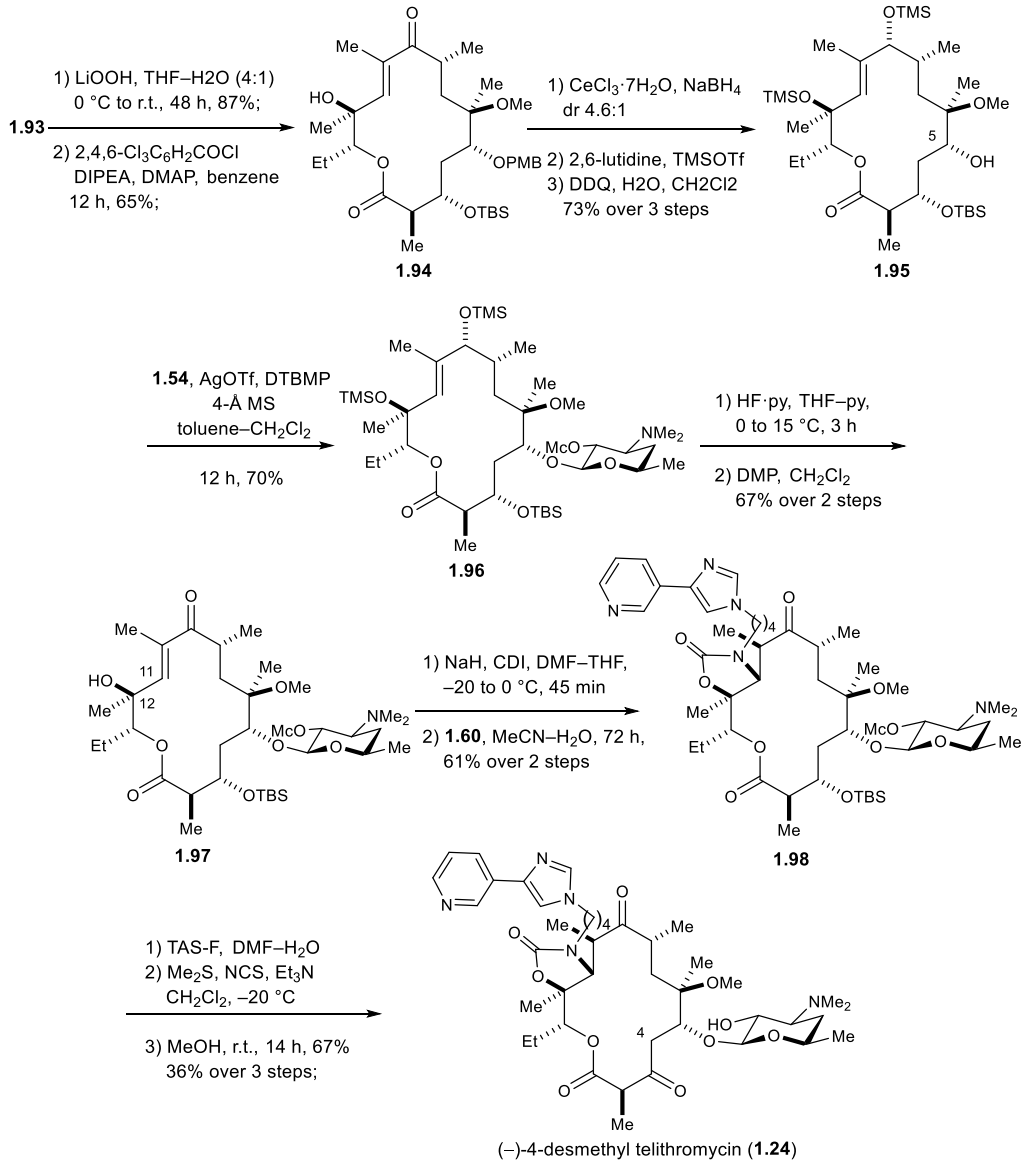
Scheme 1.19 Intermolecular NHK approach to the C1–C13 framework



hydroxyl group with PMB group followed the stereoselective installation of the C8-methyl group via kinetic protonation was realized. With PMB as a protecting group instead of TES group, a dr of 6:1 was obtained. Lactone reduction with $LiAlH_4$, protection of the primary alcohol as its TBS ether, and methylation of the tertiary C5 carbinol delivered **1.89**. Chemoselective reduction of the C3-*O*-benzyl ether over the C5-O-PMB ether was accomplished with Raney Ni. Swern oxidation to the aldehyde followed by the Evans aldol reaction furnished **1.90**. Protection of the C3-hydroxyl group as its TBS ether proceeded

smoothly. Chemoselective removal of the primary C9 TBS ether with CSA afforded alcohol that was oxidized to aldehyde **1.91**. NHK/oxidation delivered enone **1.93**. It is important to note that the acetate moiety in **1.92** was critical in order to execute the NHK coupling reaction.

Scheme 1.20 Synthesis of (–)-4-desmethyl telithromycin (**1.24**)



The next goal in the synthesis of **1.24** was preparing the requisite macroketolactone **1.94** (Scheme 1.20). To accomplish this, removal of the Evans auxiliary yielded acid which was subjected to Yamaguchi's protocol delivered a macroketolactone **1.94**. Reduction of the C9 ketone under Luche condition and protected both the C9 and C12 hydroxyls as TMS ethers. Employing a TES ether at the C9 hydroxyl, the additional methyl at C10 made both protection and removal of the TES group more intractable. Thus, the smaller TMS ether that ultimately suited our purposes. Removal of PMB ether set the stage for the stereoselective glycosylation with donor **1.54**. Glycosylation furnished product **1.96** as a single stereoisomer by featuring of the C2′-O-methoxycarbonyl (Mc) protecting group. With the macrolactone framework fully assembled, the oxidation state at C9 was adjusted by removing TMS ethers at C9 and C12 and oxidizing C9 with DMP. Baker cyclization formed oxazolidinone side chain. Endgame for 4-desmethyl TEL (**1.24**) began adjustment of the C3 oxidation state. Removal of the TBS protecting group followed by oxidation furnished the C3 ketone. Methanolysis to remove Mc protecting group on the 2′-hydroxyl position of desosamine delivered target compound **1.24** in 67% yield.

1.9 Conclusion

Based on knowledge regarding macrolide antibiotics, it was hypothesized that replacing the 4-methyl group of ketolide drugs such as telithromycin with hydrogen (i.e., desmethylation) would avoid a steric clash with the 2-amino group of guanine at 2058 (Figure 1.7). The total synthesis of all four proposed desmethyl analogs of telithromycin were successfully accomplished. In addition to replacing C4 methyl, the C8 and C10

positions were targeted for desmethylation to expedite the synthesis and probe the roles of those methyls in determining bioactivity. Significantly, the synthetic route to each analog was unique. The proper sequencing of three key coupling tactics ring-closing metathesis, NHK reaction, and Yamaguchi esterification was effective. The antibacterial activity of the analogs was evaluated using minimum inhibitory concentration (MIC) assays against a panel of wild-type and resistant *E. coli* and *S. aureus* strains. Overall, these data do not support the hypothesis that strategic desmethylation of ketolide antibiotics can directly address resistance arising from the pathogenically relevant A2058G mutation. The sequential addition of methyl residues corresponded with an increase in bioactivity, thus revealing the critical nature of these in biasing macrolactone conformation as well as participating in VDW interactions with the macrolide binding site in the 50S subunit of the bacterial ribosome.

1.10 References

1. Woodward, R. B. *Angew. Chem.* **1957**, *69*, 50–58.
2. Katz, L.; Ashley, G. W. *Chem. Rev.* **2005**, *105*, 499–527.
3. Gardner, A. D.; Chain, E. *Br J Exp Path.* **1942**, *23*, 123–127.
4. Brockmann, H.; Henkel, W. *Naturwissenschaften.* **1951**, *37*, 284–288.
5. Kurath, P.; Jones, P. H.; Egan, R. S.; Perun, T. J. *Experientia.* **1971**, *27*, 362.
6. Morimoto, S.; Takahashi, Y.; Watanabe, Y.; Omura, S. *J. Antibiot.* **1984**, *37*, 187–189.
7. DjokiC, S.; Kobrehel, G.; Lazaravski, G.; Lopotar, N.; Z. T. *J. Chem. Soc. Perkin. Trans. I.* **1986**, 1881–1886.
8. Barber, J. *Magn. Reson. Chem.* **1991**, *29*, 740–743.
9. Agmon, I.; Amit, M.; Auerbach, T.; Bashan, A.; Baram, D.; Bartels, H.; Berisio, R.; Greenberg, I.; Harms, J.; Hansen, H. A. S.; Kessler, M.; Pyetan, E.; Schluenzen, F.; Sittner, A.; Yonath, A.; Zarivach, R. *FEBS Lett.* **2004**, *567*, 20–26.
10. Bashan, A.; Agmon, I.; Zarivach, R.; Schluenzen, F.; Harms, J.; Berisio, R.; Bartels, H.; Franceschi, F.; Auerbach, T.; Hansen, H. A. S.; Kossoy, E.; Kessler, M.; Yonath, A. *Mol. Cell.* **2003**, *11*, 91–102.
11. Schlünzen, F.; Zarivach, R.; Harms, J.; Bashan, A.; Tocilj, A.; Albrecht, R.; Yonath, A.; Franceschi, F. *Nature* **2001**, *413*, 814–821.
12. Blair, J. M. A.; Webber, M. A.; Baylay, A. J.; Ogbolu, D. O.; Piddock, L. J. V. *Nat. Rev. Microbiol.* **2015**, *13*, 42–51.
13. Weisblum, B. *Antimicrob. Agents Chemother.* **1995**, *39*, 577–585.

14. Allen, N. E. *Antimicrob. Agents Chemother.* **1977**, *11*, 669–674.

15. Agouridas, C.; Denis, A.; Auger, J. M.; Benedetti, Y.; Bonnefoy, A.; Bretin, F.; Chantot, J. F.; Dussarat, A.; Fromentin, C.; D'Ambrières, S. G.; Lachaud, S.; Laurin, P.; Le Martret, O.; Loyau, V.; Tessot, N. *J. Med. Chem.* **1998**, *41*, 4080–4100.

16. LeMahieu, R. A.; Carson, M.; Kierstead, R. W.; Fern, L. M.; Grunberg, E. *J. Med. Chem.* **1974**, *17*, 953–956.

17. Baker, W. R.; Clark, J. D.; Stephens, R. L.; Kim, K. H. *J. Org. Chem.* **1988**, *53*, 2340–2345.

18. Shain, C. S.; Amsden, G. W. *Ann. Pharmacother.* **2002**, *36*, 452–464.

19. Ross, D. B. *N. Engl. J. Med.* **2007**, *356*, 1601–1604.

20. Fernandes, P.; Martens, E.; Bertrand, D.; Pereira, D. *Bioorg. Med. Chem.* **2016**, *24*, 6420–6428.

21. Tu, D.; Blaha, G.; Moore, P. B.; Steitz, T. A. *Cell*. **2005**, *121*, 257–270.

22. Velvadapu, V.; Paul, T.; Wagh, B.; Glassford, I.; Debrosse, C.; Andrade, R. B. *J. Org. Chem.* **2011**, *76*, 7516–7527.

23. Xuan, R.; Oh, H. S.; Lee, Y.; Kang, H. Y. *J. Org. Chem.* **2008**, *73*, 1456–1461.

24. Velvadapu, V.; Glassford, I.; Lee, M.; Paul, T.; Debrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2012**, *3*, 211–215.

25. Zimmerman, H. E. *Acc. Chem. Res* **1987**, *20*, 263–268.

26. Wagh, B.; Paul, T.; Glassford, I.; Debrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2012**, *3*, 1013–1018.

27. Glassford, I.; Lee, M.; Wagh, B.; Velvadapu, V.; Paul, T.; Sandelin, G.; DeBrosse,

C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2014**, *5*, 1021–1026.

CHAPTER 2

PROGRESS TOWARD THE SYNTHESIS OF (*R*)-4-FLUORO-4-DESMETHYL-SOLITHROMYCIN

2.1 Introduction

The biological evaluation of (–)-4-desmethyl telithromycin was insightful and offered an opportunity to reformulate strategy. The fact that 4-desmethyl telithromycin was fourfold *less* potent than comparator telithromycin against the A2058G mutant strain indicated that replacing the 4-methyl with hydrogen to avoid a steric clash with the 2-amino group of G2058 was insufficient to rescue bioactivity.[1] Guided by MD simulations of G2058 mutant ribosomes (Figure 2.1), it was reasoned that an alternative, logical strategy would be the replacement of the 4-methyl group with one possessing a smaller vdW radius yet capable of establishing favorable interactions with both wild-type and A2058G mutant ribosomes. The candidate that was put forward was a fluorine atom possessing the same configuration as the natural 4-methyl group to arrive at (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1a**).

In 2005, solithromycin (**2.1a)** was developed by Optimer Pharmaceuticals.[2] Inspired by the structure of telithromycin, which possesses an imidazole-pyridyl biaryl side chain, chemists at Optimer modified the imidazole nucleus into a triazole so that they could prepare a library of analogs by employing the Cu-catalyzed azide-alkyne (Click) reaction[3]. The addition of a fluoro group at C2, initially reported by Hoechst-Marion-Roussel,

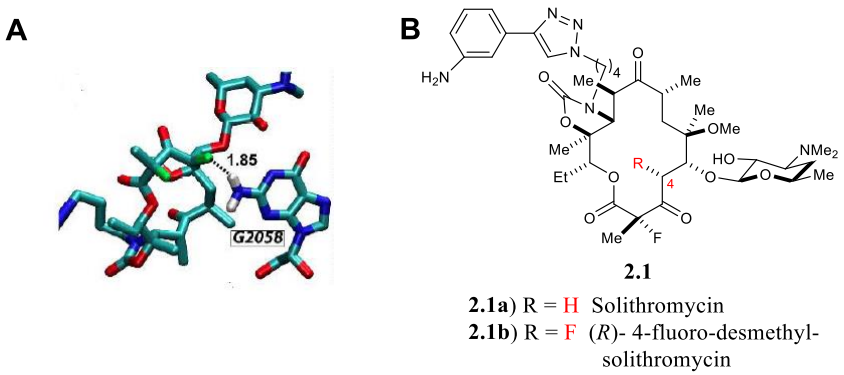


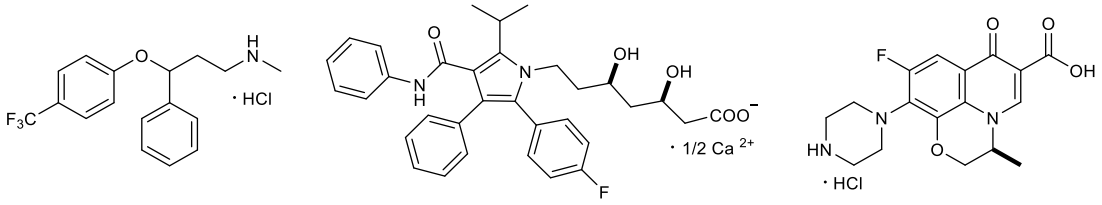
Figure 2.1 (A) (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1**) bounds to A2058G mutant ribosome; (B) Structure of solithromycin (**2.1a**) and 4-fluoro-4-desmethyl congener (**2.1b**)

resulted in a fourfold improvement in potency. Co-crystal structures of **2.1a** and *E. coli* ribosomes reveal the 2-fluoro group makes favorable hydrophobic interactions with C2611.[4] Solithromycin is currently in Phase III clinical trials and is the best-in-class ketolide developed to date.

2.2 Background

The field of fluorine chemistry was born when Henry Moissan first isolated elemental fluorine in 1886.[5] He made a solution of hydrofluoric (HF) acid with potassium hydrogen fluoride (KHF_2) and passed an electric current through it. In the event, a gas evolved at one end of the apparatus, which was the new element. Moissan gave the name "fluorine" since it was isolated from the mineral fluorite.[6] Since then, fluorine has received considerable attention from the scientific community due to its physical and chemical properties (e.g., small size, strong electronegativity, and low polarizability). The

introduction of fluorine into molecules often results in significant changes in their chemical, physical, and biological properties without substantial modification because of its size. Fluorine's atomic radius is intermediate between hydrogen and oxygen (Table 2.2). Nowadays, molecules containing at least one fluorine atom are widespread in various fields, particularly agrochemicals, electronics, and medicine. In medicinal chemistry, fluorine has become a prodigious tool in drug discovery. Accordingly, it is no surprise that nearly one fifth of all drugs on the market today contain at least one fluorine atom. For example, three of the top blockbuster drugs contain fluorine atoms, including the antidepressant fluoxetine (Prozac) **2.2**, the cholesterol-lowering drug atorvastatin (Lipitor) **2.3**, and the potent antibiotic ciprofloxacin (Ciprobay, **2.4**), which has an aromatic fluorine substituent (Figure 2-2).[7]



Prozac **2.2** Lipitor **2.3** Ciprobay **2.4**

Figure 2.2 Examples of drugs containing a fluorine atom

2.3 Effects of Fluorine

2.3.1 Electronic Effect of Fluorine

Even though fluorine is a very common element in the earth's crust (13th most abundant), fluorine is difficult to prepare because of its high reactivity. Fluorine is the most electronegative element in the periodic table (4.0 on the Pauling scale), which results from the strong attraction between valence electrons of the outer 2p shell and the positive charge in the nucleus. Consequently, it possesses low polarizability and a high ionization potential. The C-F bond is among the strongest in organic chemistry.[8] This can be rationalized in terms of the high electronegativity of the fluorine atom, which strongly polarizes the C-F. As such, there is a charge attraction between fluorine and carbon atoms where the fluorine atom bears a partial negative charge and the carbon atom bears a partial positive charge. The C-F bond possesses significant ionic character; it is a very short and strong bond.

Table 2.1 Bond dissociation energies of various C-X bonds[4]

Bond	Dissociation energy (kcal/mol)
C-F	105.4
C-H	98.8
C-O	84.0
C-C	83.1
C-Cl	78.5
C-N	69.7

2.3.2 Hydrogen Bonding to Fluorine

One important feature of molecules in biological systems is the presence of hydrogen bonds. A hydrogen bond is defined as the electrostatic interaction between a

hydrogen atom that is bound to a highly electronegative element with another electronegative element with a distance smaller than the sum of the Van der Waals radii. Therefore, in the case of fluorine, this C-F···H-X distance should be shorter than 2.35 Å, where X is an electronegative atom such as O or N.[8] The high electronegativity of fluorine, as well as the fact that the C-F bond is highly polarized, suggests that C-F should be a good hydrogen bond acceptor. However, studies evaluating structures deposited in the Cambridge Structural Database (CSD) shows that true H···F contacts are rare.[9] Moreover, fluoroorganic compounds form only weak hydrogen bonds. Calculations give 2.0–3.2 kcal/mol for a C-F···H-O interaction as compared to 5.0–10.0 kcal/mol for a C-O···H-O interaction. Thus, it is less than half the strength of a typical hydrogen bond.[9–11] According to Dunitz, "organic fluorine hardly ever accepts hydrogen bonds, that is, it does so only in the absence of a better acceptor."[11] This reluctance to engage in hydrogen bonding can be explained by the high electronegativity of the fluorine atom and the strong electrostatic character of the C-F bond which compresses the lone pairs around the fluorine atom, reducing dramatically the capacity of organic fluorine to act as a hydrogen bonding acceptor. However, even if interactions are weaker than C=O···H-X (X= N or O), they do play a role in crystal packing.[12] Several studies of crystal structures of small molecules from CSD and protein-ligand complexes from the Protein Data Base (PDB) have shown evidence of dipole-dipole interactions between C-F (aliphatic and aromatic) and polarized functional groups such as carbonyl, carbonyl derivatives, nitriles and even the nitro group.[13–15]

2.3.3 Steric and Geometric Effects of Fluorine

Fluorine is a small atom with a Van der Waals radii (Å) between that of hydrogen and oxygen (Table 2.2).[16] Due to its modest size, fluorine can be introduced into organic molecules as a replacement for either a hydrogen atom or a hydroxyl group without a significant change in the steric environment. The close isosteric relationship between fluorine and oxygen is evidenced by its Van der Waals radius, which is closer to oxygen than hydrogen. Despite the fact that fluorine is larger than hydrogen, solid state X-ray structures show fluorine and hydrogen often interchange.[17] Consistent with this, replacing hydrogen by fluorine causes little change to the steric footprint and binding of analogs to their cognate target proteins. Despite its size, fluorine has emerged as a reasonable hydrogen isostere (i.e., mimic).

Table 2.2 Van der Waals radii of various atoms[16]

Atom	Van der Waals radii (Å)
H	1.20
F	1.47
O	1.52
N	1.55
Cl	1.75

1,2-Difluoroethane has been the subject of extensive conformational analysis (Figure 2.3).[18] There are two possible conformations wherein the fluorine atoms are either *gauche* or *anti*. Surprisingly, NMR and molecular modeling results have shown that the *gauche*

conformer possesses lower energy. Despite the steric repulsion between the fluorines in the *gauche* conformation, it is 0.08 kcal/mol more stable than the *anti*- conformation. The conformations of fluorinated compounds depend on their steric requirements and their electronic properties, which is related to the explanation the *gauche effect* of fluorine. In the *gauche* conformer of 1,2-difluoroethane, antibonding orbital of C-F bonds ($\sigma^*_{C\text{-}F}$) are aligned with adjacents orbital of C–H bonds ($\sigma_{C\text{-}H}$), which can donate electron density into the $\sigma^*_{C\text{-}F}$ orbital in a process known as hyperconjugation.[4]

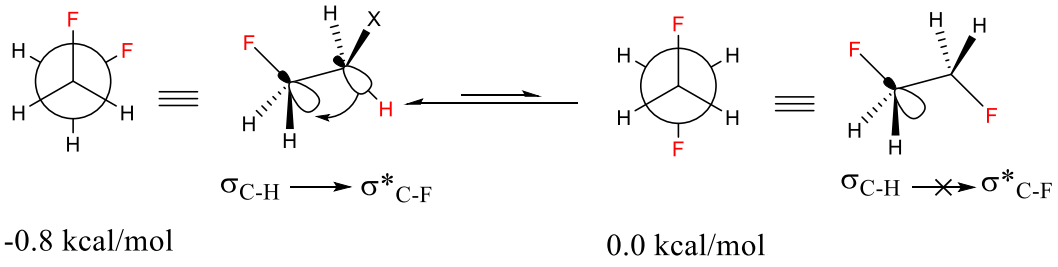


Figure 2.3 Conformational effect associated with C–F bond[19]

The delocalization of electron density into antibonding orbitals corresponds to partial bond-breaking, so that hyperconjugation results in the elongation of C–F bond and slightly diminished covalent character. Overall, the *gauche* conformer will have lower energy under the stabilizing effect of hyperconjugation. In contrast, each antibonding orbital of C-F in the *anti*- conformer is aligned with an adjacent s orbital of C–F bond, which is highly polarized and less donating than the s orbital of C–H bond and hence hyperconjugation does not occur. The high electronegativity of fluorine polarizes the C-F

bond, and hyperconjugative electron donation from a vicinal C–H bonding s orbital to antibonding orbital of the C–F bond stabilizes the *gauche* conformation. Altogether, the *gauche* effect has proven to be a general conformational phenomenon in fluorinated organic molecules.[18]

2.3.4 Lipophilicity of Fluorine

Lipophilicity is an important property, particular in medicinal chemistry. To cross lipid membranes, a drug needs to be sufficiently lipophilic such that it can pass into the lipid core but not become trapped in it. Lipophilicity is expressed as a logarithmic coefficient of a compound's partition between octanol and water at a given pH and abbreviated as logP. The most lipophilic compounds will be partitioned in the octanol layer whereas the least lipophilic compounds will be partitioned in the water layer. In order to obtain good binding with or affinity for a molecular target, lipophilic (i.e., Van der Waals) interactions are important. However, a drug must not be too lipophilic as this would reduce its water solubility and hence bioavailability. Selective fluorination emerges as a good method in which to tune the lipophilicity of a molecule, as the introduction of one or more fluorine atoms can increase the lipophilicity in an incremental manner. However, it can be difficult to predict precisely what the effect of the introduction of a fluorine atom will be, but some general rules have been established. In the case of aromatic molecules, the presence of a fluorine atom will usually increase the lipophilicity. On the other hand, with aliphatic molecules, the situation is a bit more complicated and it is important to go back to the

definition of lipophilicity. Usually when the lipophilicity increases, the hydrophobicity increases, and vice versa. However, with the introduction of fluorine, lipophilicity can decrease but the hydrophobicity can increase at the same time. As the solubility of the fluorinated molecule decreases more in water than in octanol, there is an apparent overall lipophilicity increase.[20]

2.4 Fluorine in Organic Chemistry

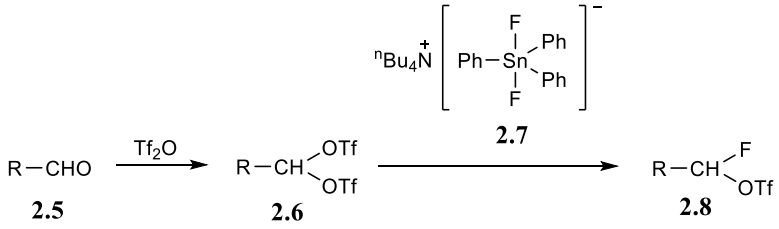
Since there are few naturally occurring fluorine containing compounds,[21] it is necessary to fluorinate organic compounds at a certain stage of the syntheses. Though fluorine gas and hydrogen fluoride are used as fluorine sources, they are very toxic and corrosive and require special equipment and techniques for handling them. Therefore, alternate fluorinating agents have been developed,[22] which are used in laboratories to easily introduce a fluorine atom at selected positions of the compounds. Fluorinating agents are classified into one of two categories: nucleophilic or electrophilic. Nucleophilic fluorinating agents are those where the fluoride anion serves as the reactive species. Electrophilic fluorinating agents are those where an electron-deficient fluorine atom serves as the species.

2.4.1. Nucleophilic Fluorinating Agents

The most common nucleophilic fluorinating agent is hydrogen fluoride, which is extensively employed in the industrial production of fluorinated compounds. Hydrogen fluoride, however, is scarcely used in laboratories due to its toxic and corrosive properties

and its low reactivity resulting from high H-F bond energy. Nucleophilic fluorinating agents such as KF, CsF, and $Bu_4N{\cdot}F$ (TBAF) are readily available. These fluorinating agents are hygroscopic, and unfortunately, the moisture in these substances forms strong hydrogen bonds with the fluoride anion and this leads to decrease in reactivity. "Naked" fluoride anions, where the fluoride anion is completely free of hydrogen bonds, have been developed. For example, tetrabutylammonium difluorotriphenyltin **2.7** (TBAT), which was reported by Gingrasas and alternative reagent to $Bu_4N{\cdot}F$, is soluble in organic solvents.[23] Martinese *et al.*, synthesized gem-fluoro triflate **2.8** from aldehyde using **2.7**.[24,25]

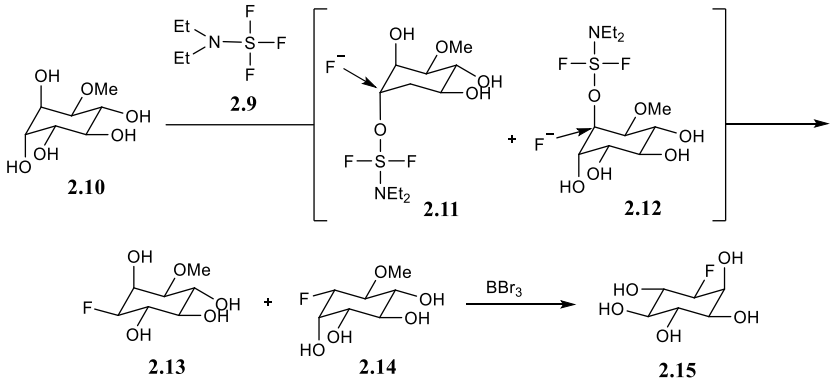
Scheme 2.1. Reaction of **2.7** with aldehyde



Diethylaminosulfur trifluoride (DAST, **2.9**), is used in a wide variety of synthetic processes for stereospecifically substituting a fluorine atom for a hydroxyl group.[26–29] Kozikowski *et al.*, have developed a method for producing (–)-1-deoxy-1-fluoro-*myo*-inositol (**2.15**) from quebrachitol (**2.10**) in two stages.[30] The authors reported that DAST reacts with either one of two axial hydroxyl groups of **2.10** to form a DAST-functionalized intermediate, which greatly increases the leaving ability of the hydroxyl group. This enables an easy nucleophilic displacement of the hydroxyl group by the fluoride anion

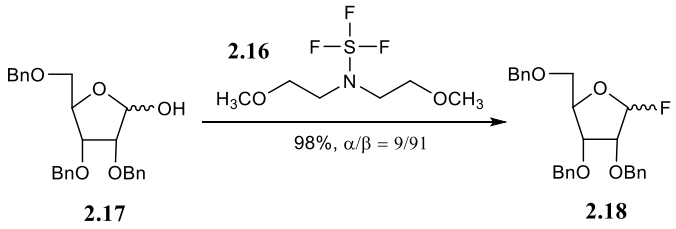
accompanied with steric inversion. DAST is unstable to heat and decomposes upon heating. In general, reactions employing DAST are completed with short reaction times and are conducted at or below room temperature.

Scheme 2.2. Synthesis of (–)-1-deoxy-1-fluoro-*myo*-inositol (**2.15**) with DAST



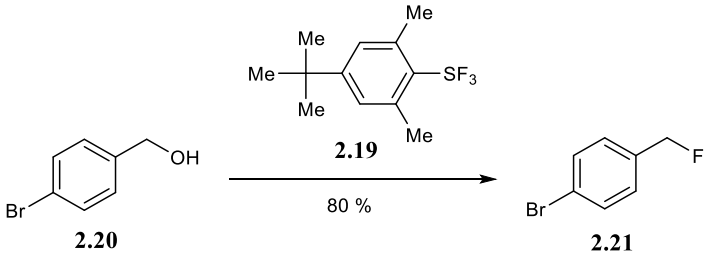
Lal *et al*. have reported that a DAST analog, Bis(2-methoxyethyl)-amino sulfur trifluoride (Deoxofluor) **2.16**, was more thermally stable than **2.9**.[31–34] The reactivity of Deoxofluor (**2.16**) is comparable to DAST (**2.9**) and in some substrates superior. As such, it has received more attention as a nucleophilic fluorinating agent.

Scheme 2.3 Reaction of **2.16** with D-ribofuranose to prepare glycosyl fluoride **2.18**



4-*tert*-2,6-Dimethylphenyl sulfur trifluoride (FLUOLEAD, **2.19**), which was first reported by Umemoto, is a novel nucleophilic fluorinating agent.[35] **2.19** is a white crystalline solid with high thermal stability (232 °C thermal decomposition temperature by DSC analysis). Unlike other existing fluorinating agents such as DAST, FLUOLEAD (**2.19**) does not appreciably fume upon handling. The reactivity of **2.19** with water is slow, which makes it easier to handle in the open air. Fluorinations of hydroxyl or carbonyl groups with **2.19** afforded the corresponding fluorinated compounds in good yields under a wide range of conditions (0 to 100 °C).

Scheme 2.4. Reaction of **2.19** with 4-bromo benzyl alcohol

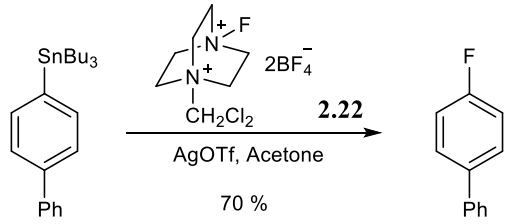


2.4.2. Electrophilic Fluorinating Agents

The simplest and most straighfordward reagent amongst the electrophilic fluorinating agents is fluorine gas; however, it is not suitable for partial fluorination due to its vigorous reactivity and strong toxicity. *N*-Fluoro-*N*'-(chloromethyl)triethylenediamine bis(tetrafluoroborate) (Selectfluor, **2.22**) is an easily handled and a versatile electrophilic fluorinating agent for enols, silyl enol ethers, alkenes, stabilized carbanions, aromatic

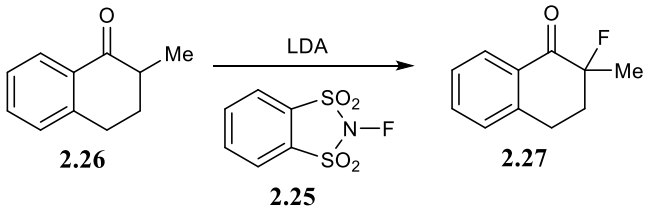
compounds, organosulfur compounds, and Grignard agents among other nucleophilic groups.[22,36]

Scheme 2.5. Reaction of aryl stannane **2.23** with Selectfluor (**2.22**)



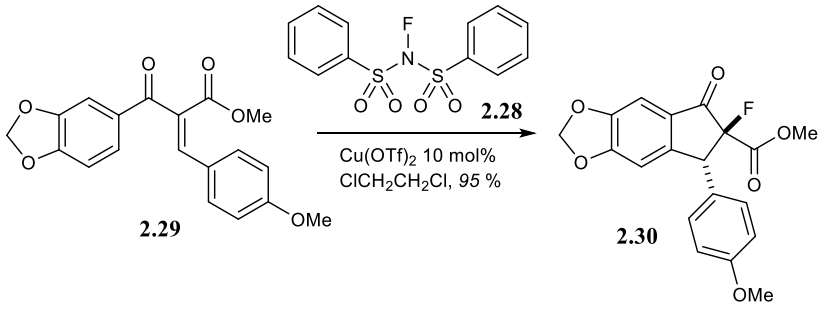
N-Fluoro-O-benzenedisulfonimide (NFOBS, **2.25**) was developed by Davis in 1991.[37] It is a stable, easily prepared, highly efficient source of electrophilic fluorine which fluorinates enolates, azaenolates and carbanions in good to excellent yields. Ketone enolates were monofluorinated in excellent yield.[38]

Scheme 2.6. Fluorination of enolate with NFOBS



N-Fluorobenzenesulfonimide (NFSI, **2.28**) is another easily handled and stable crystalline electrophilic fluorinating agent for enolates, silyl enol ethers, aromatic compounds, organolithium compounds, and alkyl phosphate carbanions among others.[39]

Scheme 2.7. Catalytic tandem Nazarov cyclization/sequential fluorination trapping using NSFI (**2.28**)

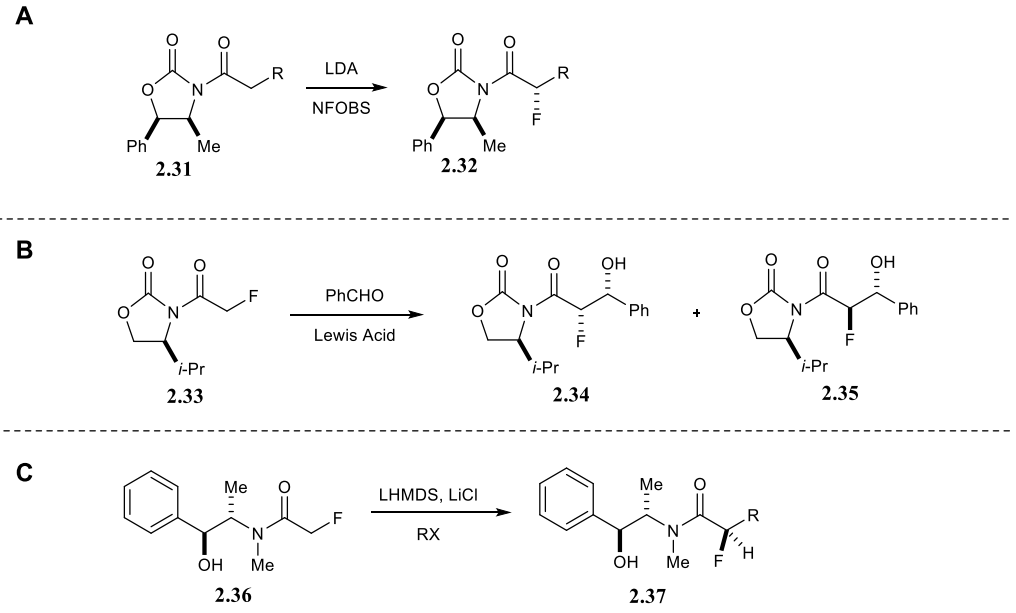


2.4.3 Chiral Auxiliary as a Fluorine Source

Direct enantioselective fluorinations can involve the use of chiral auxiliaries. Davis and co-workers have studied oxazolidinones as potential fluorinating chiral auxiliaries in 1992.[40] While covalent attachment of the auxiliary to the carbonyl restricts the substrate compatibility, this auxiliary has generated α-fluoro carboximides in excellent diastereomeric excesses and yields (Scheme 2.8A).[41] In the context of asymmetric aldol reactions, Pridgen *et al.* have advanced the most impressive methodology with fluoroacetyl oxazolidinone **2.33**, which was inspired by the work of Evans.[42] Various combinations of base or Lewis acid were used to generate a chiral α-fluoroenolate, which reacted with aromatic aldehydes to generate α-fluoro-β-hydroxy compounds. The reaction generated

predominantly the *syn*-aldol **2.34** product via non-coordinated transition states when boron, titanium or tin(II) were employed as Lewis acids. *Anti*-aldol **2.35** was the major isomer via a coordinated transition state in the cases of tin(IV) and zinc (Scheme 2.8B). Notably, the 2-oxazolidinone moiety employed in this method is highly versatile in that the resulting fluorinated products are readily converted into esters, acids, amides, alcohols, or diols.[43–45] In 1997, Myers introduced asymmetric alkylation with pseudoephedrine-functionalized α-fluoroacetamide **2.36** and reactive alkyl halides which was efficient and with highly diastereoselective (Scheme 2.8C).[46] The alkylation products could be hydrolyzed under mildly basic conditions to form highly enantiomerically enriched α-fluoro carboxylic acids. Using this methodology, the Myers group successfully synthesized a novel series of HIV protease inhibitors which contained monofluoro hydroxyethlene dipeptide isosteres.[47]

Scheme 2.8 (A) Oxazolidinone directed fluorination, (B) Aldol reaction with the Evans auxiliary and (C) Alkylation using Myers' pseudoephedrine auxiliary



2.5 Present Study

2.5.1 Rationale

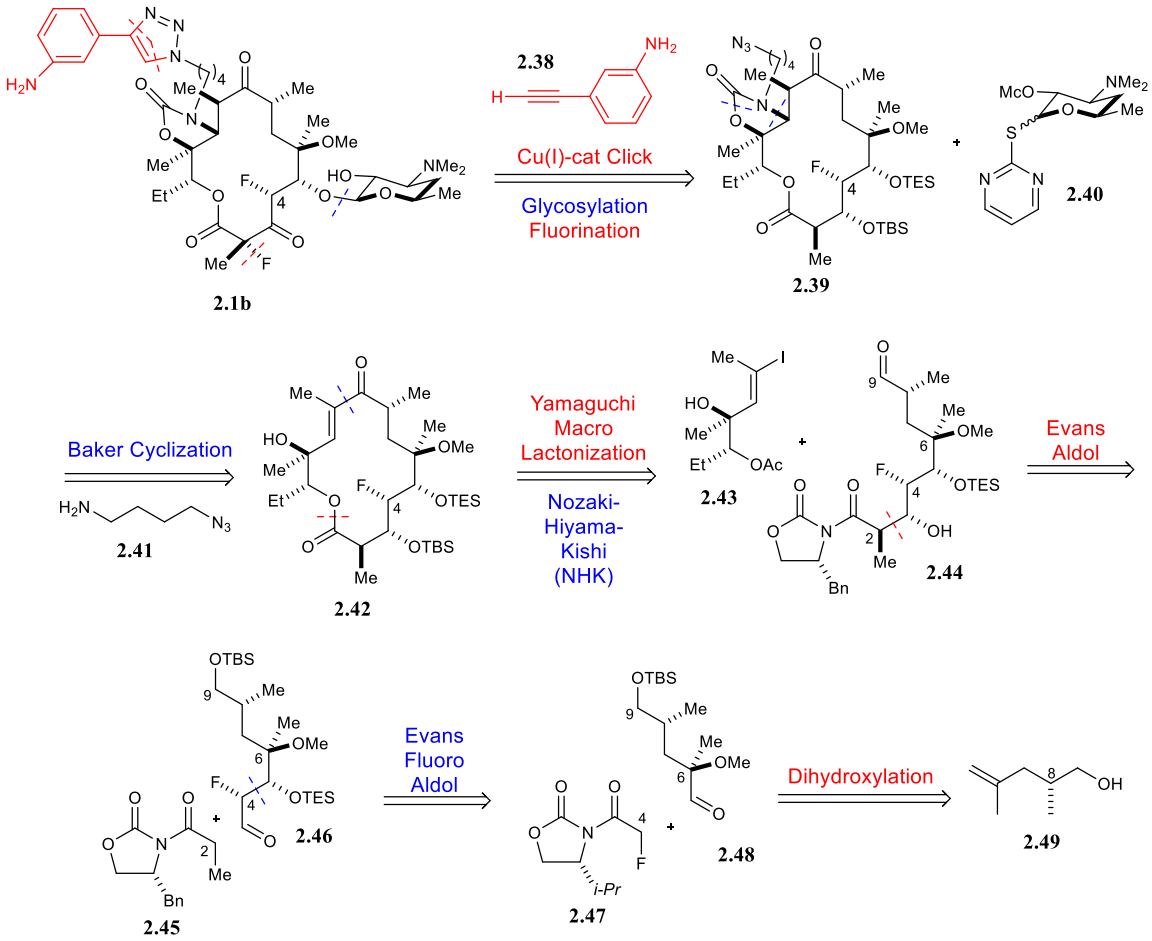
Specifically, we reasoned that (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1b**) would be an ideal candidate. The hypothesis was that the 4-fluoro moiety would engage in dipole-dipole interactions (C-F·H) with the exocyclic 2-amino group of guanine (Figure 2.1), which is based on accumulated evidence that strategic placement of organofluorine can strongly impact potency, selectivity, and physicochemical properties.[48] In addition, the axially disposed fluoro group in (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1b**) will provide conformational stabilization from a gauche effect with the vicinal O5 group.[18] The 4-methyl group in all erythromycin congeners acts to stabilize the diamond-lattice (Perum-Celmer) conformation by avoiding *syn*-pentane interactions with methyl groups at C2 and C6.[49]

2.5.2 Retrosynthetic Analysis

Scheme 2.9 details the retrosynthetic analysis of (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1b**). The aniline-triazole biaryl side chain would be prepared from 3-ethynyl aniline (**2.38**) and azide using the Cu(I)-catalyzed variant of the Huisgen dipolar cycloaddition, which was first introduced in 1961.[50] The Cu-catalyzed variant (i.e., Click reaction) would smoothly lead to the desired 1,2,3-triazole, as was done by Optimer in their synthesis of solithromycin (**2.1a**).[51] Enolate formation of **2.39** with KO-*t*Bu and reaction with nucleophilic fluorinating agent *N*-fluorobenzene sulfonimide (NFSI) would install the

C2 fluorinated substituent. Stereoselective glycosylation of the C5 hydroxyl group would be installed with Woodward's thiopyrimidine desosamine donor **2.40**. Installation of the oxazolidinone in **2.42** would be accomplished using the protocol first reported by Baker and co-workers at Abbott and require enone **2.42** and primary amine **2.41**.[52] The 14-membered macrolactone in **2.42** would be accessed via intramolecular Yamaguchi-

Scheme 2.9 Retrosynthetic analysis of (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1b)**



macrolactonization.[53] Further disconnection followed by intermolecular Nozaki-Hiyama-Kishi (NHK) coupling[54] between aldehyde **2.44** and vinyl iodide **2.43**, which were successfully coupled in the synthesis of previous desmethyl analogs (see Chapter 1). The C1-C9 fragment **2.44** would be derived from the union of **2.45** and **2.46** via the venerable Evans aldol reaction.[55] Finally, intermediate **2.46** would be prepared using fluoroacetate aldol methodologies with **2.47**[42] and the aldehyde **2.48** derived from known C5-C9 building block **2.49**.[56]

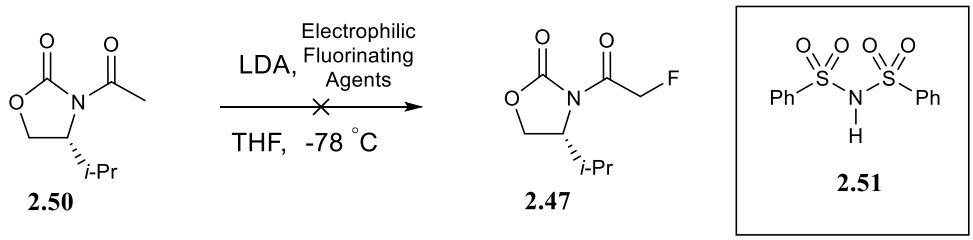
2.5.3 Synthesis of Right-Hand Fragment 2.79

2.5.3.1 Synthesis of Fluoroacetyl Oxazolidinone (2.47)

To expedite the synthesis, the chiral auxiliary **2.47** bearing fluorine was targeted (Scheme 2.10). Rapid enhancements in the understanding of organofluorine chemistry allowed fluorinated compounds to be synthesized more easily and safely than ever before using electrophilic or nucleophilic fluorine agents.

The first attempt to prepare **2.47** was direct alkylation of the enolate of *N*-acyl oxazolidinone (**2.50**) with previously discussed electrophilic fluorinating agents NFOBS (**2.25)** or NFSI (**2.28**). Treatment of **2.50** with LDA followed by fluorinating with NFSI provided (*R*)-3-(2-fluoroacetyl)-4-isopropyl-2-oxazolidinone (**2.47**). However, attempts to separate **2.47** from the residual NFSI were hampered by tedious chromatographic separations and significant product decomposition

Scheme 2.10 Electrophilic fluorination

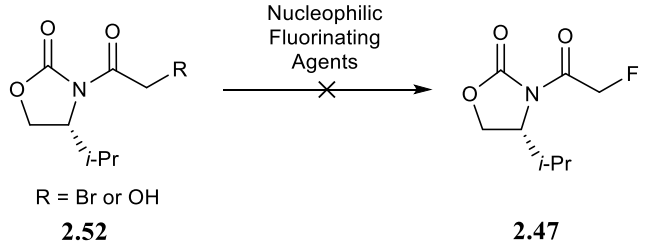


2.50 **2.47** **2.51**

Test reactions with **2.47** in the Evans fluoroaldol reaction were unsuccessful due the presence of NFSI and its acidic byproduct, dibenzenesulfonimide (DBSI) **2.51**. Purification was accomplished only with significant difficulty and results were not reproducible. Interestingly, a report by Umemoto and coworkers in 1986 showed that *N*-fluoropyridinium triflates are able to α-fluorinate sulfides at room temperature.[57] Based on this report, we speculated that if NFSI was capable of reacting with sulfides, in the same manner, a volatile sulfide such as dimethylsulfide (DMS) could be used to transform any remaining fluorinating agent into the DBSI byproduct and the resultant fluorosulfite could simply be removed by evaporation. When DMS was added to a suspension of NFSI at 4 °C, an instantaneous and exothermic reaction was observed wherein the NFSI was completely consumed. However, while excess NSFI was consumed, the DBSI side product was inseparable from the auxiliary. As DBSI is acidic, attempts at washing the reaction mixture with aqueous solutions of $NaHCO_3$, NaOH, Na_2CO_3, and K_2CO_3 could remove it without decomposing the product. Unfortunately, washing with stronger bases and also led to product decomposition. Based on difficulties associated with preparing **2.47**, recourse was made to alternative routes.

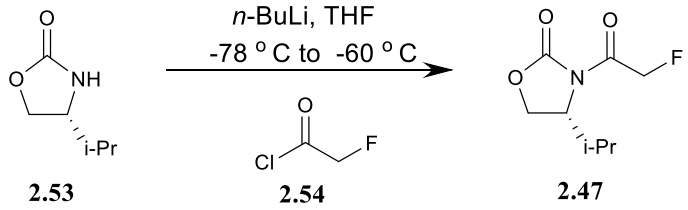
Since electrophilic fluorination was not suitable to access **2.47** with suitable purity, nucleophilic fluorination was investigated. To this end, many different fluorine nucleophiles such as KF, CsF, TBAF and DAST were used to prepare **2.47**. Unfortunately, the desired outcome was never realized under these conditions.

Scheme 2.11 Nucleophilic fluorination



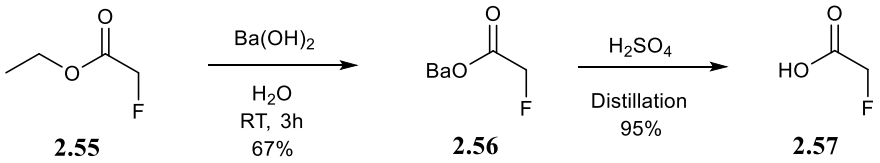
Our next approach would introduce fluorine from the fluoroacetyl chloride **2.54** which was employed by Pridgen *et al* (Scheme 2.12).[42]

Scheme 2.12 Pridgen's approach



Fluoroacetyl chloride **2.54** was commercially available but with the impractical price (1g $580 to $770). Then, the plan was set to make fluroacetic acid **2.57** to mimic fluoroacetyl chloride by activation of acid to make mixed anhydride. General ester hydrolysis reaction conditions, LiOH in the presence of THF/H_2O, did not generate acid as a product. In 1948, Stacey made fluoroacetic acid from methyl fluoroacetate with barium hydroxide and sulfuric acid.[58] This efficient alkaline hydrolysis of ester was applied to our system, ethyl fluoroacetate, to isolate barium fluoroacetate **2.56** as an intermediate. Slow addition of barium fluoroacetate to sulfuric acid on distillation under high vacuum, the fluoroacetic acid came over and crystallized immediately. The fluoroacetic acid is toxic, careful handling would be required.

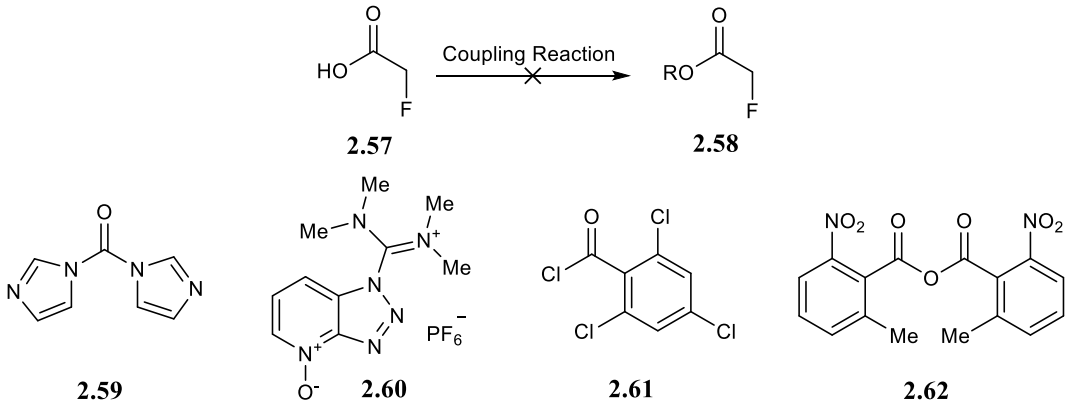
Scheme 2.13 Synthesis of fluoroacetic acid



With fluoroacetic acid in hand, the construction of the requisite fluoroacetate oxazolidinone **2.47** was pursued. Generating fluoroacetic anhydride or mixed anhydride *in situ* from **2.57** with 1,1'-carbonyldiimidazole (CDI) **2.59**, *N*-[(dimethylamino)-1*H*-1,2,3-triazolo-[4,5-*b*]pyridin-1-ylmethylene]-*N*-methylmethanaminium hexafluorophosphate *N*-oxide (HATU, **2.60**), 2,4,6-trichlorobenzoyl chloride (Yamaguchi Reagent, **2.61**), or 2-

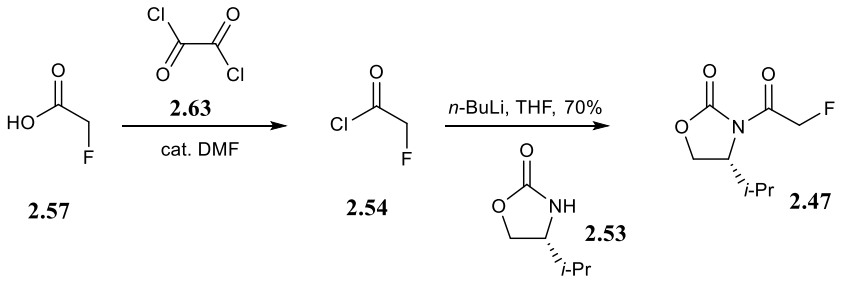
methyl-6-nitrobenzoic anhydride (Shiina reagent, **2.62**) was unsuccessful to furnish **2.47**.[53,59,60]

Scheme 2.14 Coupling reaction



Fortunately, oxalyl chloride (**2.63**) proved effective in generating chloroacetyl fluoride (**2.54**), which upon reaction with lithiated oxazolidinone **2.53** furnished the desired reagent **2.47**.[61]

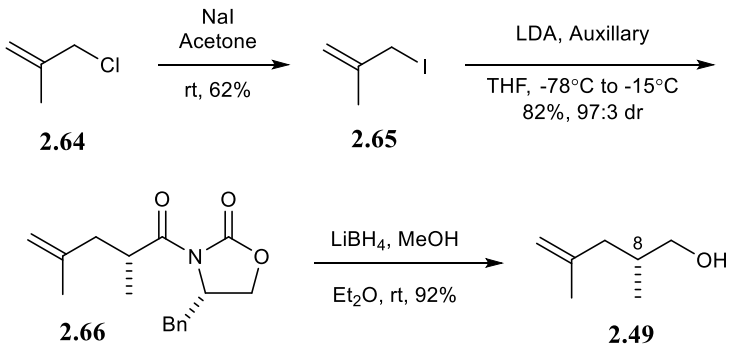
Scheme 2.15 Synthesis of **2.47**



2.5.3.2 Forward Synthesis

The construction of **2.49** began via Finkelstein conversion of commercially available methallyl chloride **2.64** to the more reactive iodide, followed in turn by a highly diastereoselective Evans alkylation. Reductive removal of the chiral auxiliary with $LiBH_4$ furnished alcohol **2.49** with the first chiral center C-8 successfully installed.[56]

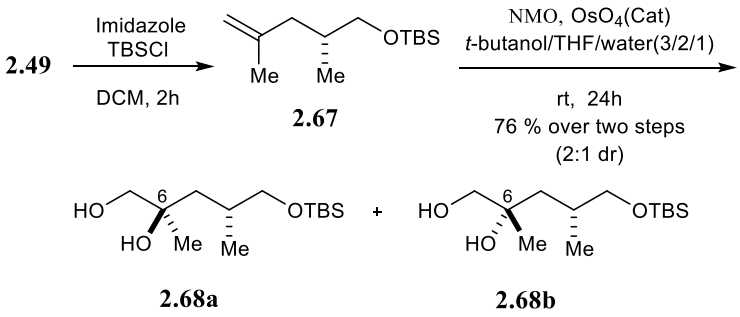
Scheme 2.16 Synthesis of known alcohol **2.49**



To introduce the C6 stereogenic center, **2.49** was first protected as its TBS ether, Upjohn dihydroxylation of the 1,1-disubstituted alkene in **2.49** would be used to set the stereochemistry at C6 (scheme 2.17).[62] In the event, diastereomeric diols **2.68** and **2.69** were isolated in 76% yield over two steps (dr=2:1). The major product **2.68** was later confirmed as the desired diastereomer (*vide infra*). To optimize the reaction, many conditions were screened. Changing the reaction temperature and/or solvent resulted in eroded diastereoselectivity. At this stage, the Sharpless asymmetric dihydroxylation (SAD) reaction was conducted. Since terminal alkenes do not follow the empirical model

(mnemonic device) for the SAD, this condition was not expected to have good selectivity.[63] Dihydroxylation conducted in the presence of the chiral auxiliary in **2.66** also did not give good selectively.

Scheme 2.17 Synthesis of Diol **2.68a**

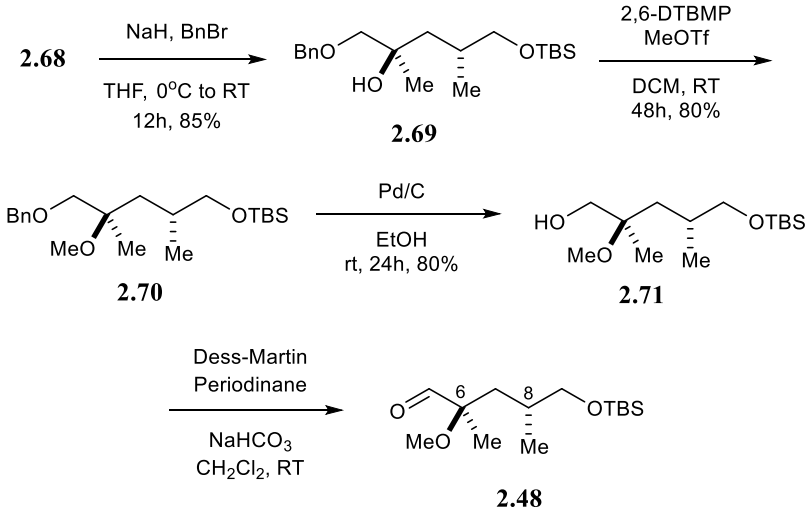


The resultant primary alcohol in **2.68** was chemoselectively protected as its benzyl ether with BnBr (1eq) and NaH (2eq) to provide tertiary alcohol **2.70**. Utilizing methylation conditions optimized in the previous desmethyl telithromycin syntheses, MeOTf and 2,6-di-*tert*-butyl-4-methylpyridine were enlisted to obtain methyl ether **2.71**.[64] Hydrogenolysis of the benzyl ether with 10% Pd/C was accomplished in 80% yield.

At this stage, the TBS group in **2.71** was removed with TBAF and the resultant diol was treated with *p*-nitrobenzoyl chloride and DMAP with the hope of preparing a crystalline bis-benzoate derivative. Unfortunately, preparing crystalline derivate was not successful and an alternative route to confirming the stereochemical configuration at C6

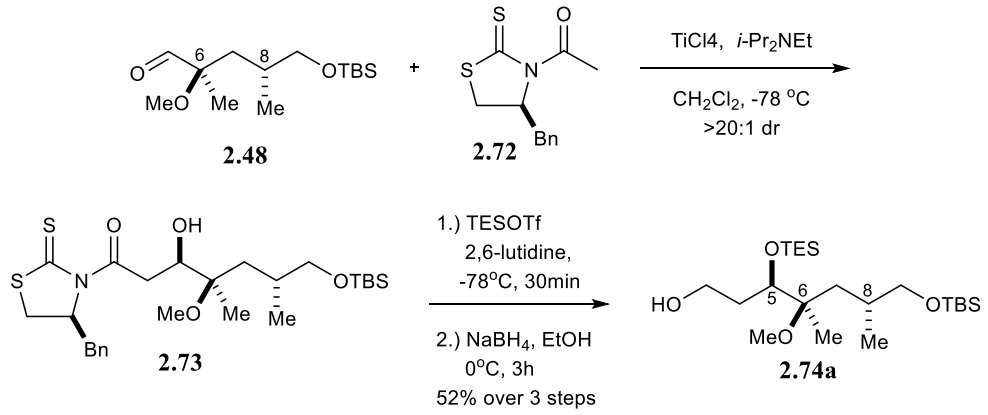
was undertaken. Thus, Dess-Martin oxidation[65] of **2.71** gave aldehyde **2.48**, which was taken directly to the next step without purification.

Scheme 2.18 Synthesis of Aldol Precursor **2.48**



Ultimately, the stereochemical configuration of **2.68** at C6 was determined by chemical correlation with a fragment previously used in the synthesis of 4,10-didesmethyl telithromycin.[64] Secondary alcohol **2.73** is available via the asymmetric aldol reaction of aldehyde **2.48** with a chiral acetyl thiazolidinethione enolate generated from **2.72** using Vilarrasa's condition.[66] The titanium enolate generated with $TiCl_4$ and Hunig's base gave predominantly the desired isomer **2.70** in excellent yield. With minor diol product **2.69**, the same reaction steps were conducted separately to correlate with the known compound **2.74a**.

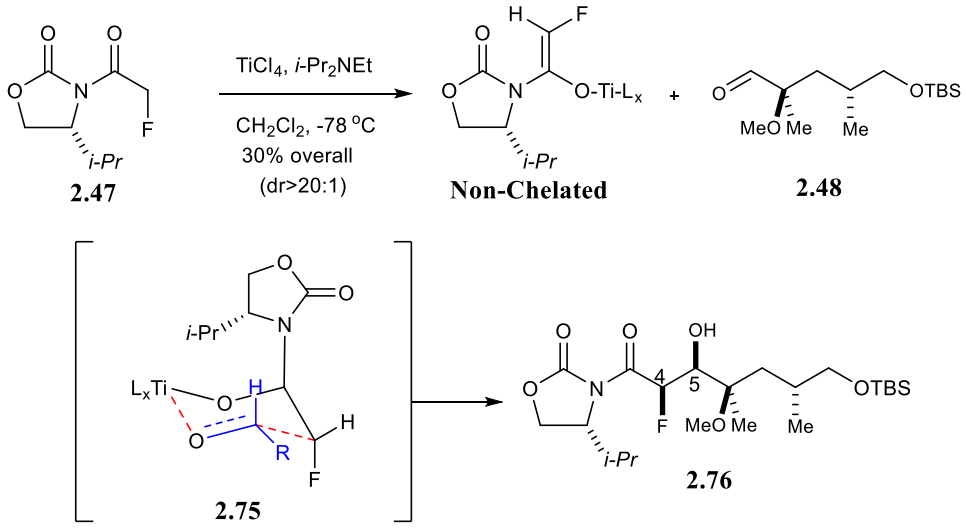
Scheme 2.19 Determination of absolute stereochemistry at C6



The most commonly used strategy for the preparation of fluorinated compound remains fluorine substitution at a late stage using fluorinating reagents. Hence, we were interested in applying a new approach. Herein, we put our efforts towards the preparation of fluorinated intermediate through stereoselective titanium-mediated aldol additions. This aldehyde **2.48** was subjected to a two carbon chain elongation step by applying a stereoselective Lewis acid mediated aldol addition. The best result for the aldol reaction was achieved when $TiCl_4$ and DIPEA were used for enolization with excellent stereoselectivity. The results may reflect the involvement of a nonchelating chair like transition state as proposed by Pridgen *et al*.[42] Unfortunately, the reactions generally failed to go to completion even after extended reaction times and isolated yields did not go beyond 30% (BRSM 50%, >20:1 dr). Many reaction conditions including varying Lewis acid [$TiCl_4$, Bu_2BOTf, and $Sn(OTf)_2$], temperature (-78 °C to RT), base (DIPEA, TMEDA, LDA, and Et_3N) and solvents (THF and DCM) were screened to improve the yield;

unfortunately, none of them resulted in any significant improvement. Limitations were handling $TiCl_4$ and having robust α-chiral tertiary aldehyde. The stereochemistry on C4 and C5 were introduced via fluoro aldol reaction.

Scheme 2.20 Aldol addition of **2.47** and **2.48** to afford **2.76**



Fluorine effect on the coupling pattern of NMR proved that our desired compound had been made. Proton, carbon, and fluorine NMR spectra were obtained. Each NMR spectra showed splitting patterns like this: ^{1}H NMR at δ 6.12(dd, $^{1}J_{HF}$= 48.5 Hz and $J_{2.3}$ = 1.6 Hz) , ^{13}C NMR at δ 167.7 (d, $^{1}J_{CF}$= 187.5 Hz), δ 89.1 (d, $^{2}J_{CF}$= 25 Hz), and δ 73.5 (d, $^{2}J_{CF}$ = 25 Hz), 19 F NMR at δ -207.77 (dd, $^{1}J_{HF}$ = 48.4 and $J_{2.3}$ 30.5 Hz). The splitting patterns matched those reported by Pridgen *et al*.[42]

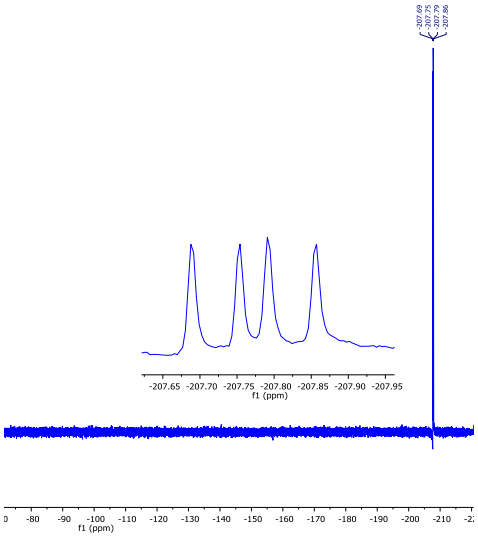


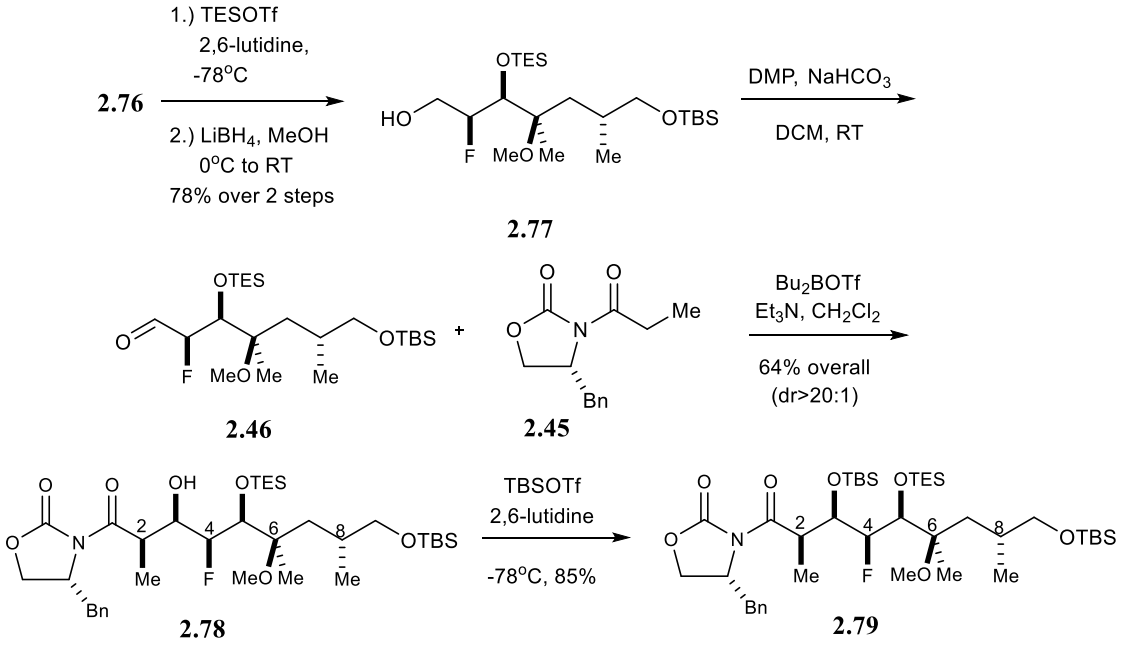
Figure 2.4 Coupled ^{19}F NMR

Protection of **2.76** as the TES ether followed by reductive auxiliary removal with $LiBH_4$ delivered alcohol **2.77**, which was oxidized with the Dess-Martin periodinane. Aldehyde **2.46** was used directly in the Evans aldol reaction with (*R*)-4-benzyl-3-propionyl-2-oxazolidinone (**2.45**) which set the configuration of the C2 and C3 carbon to furnish *syn* aldol product **2.78**. Protection of the C3 alcohol with TBSOTf and 2,6-lutidine delivered **2.79**, which represents the most advanced intermediate in hand to date.

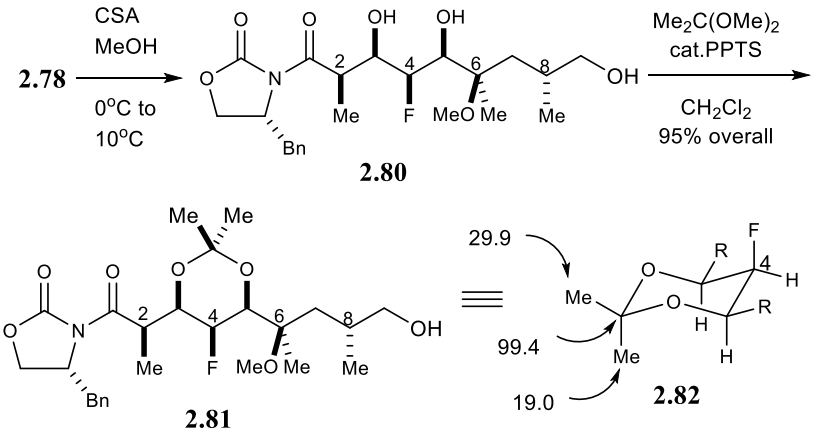
Confirmation of the stereochemistry of **2.78** began by preparing acetonide **2.81** in two steps. In accordance with Rychnovsky-Evans acetonide analysis, the ^{13}C NMR spectrum of the **2.81** contained the signals at 19.0 ppm and 29.9 ppm for the methyls of the acetonide and 99.4 ppm for the ketal carbon. Resonances from the acetonide NMR were consistent with those found in a chair conformation **2.82**, which is indicative of the newly formed 1,3-*syn*-diol disposition.[67,68] Additional evidence from the ^{1}H NMR spectrum of

2.81, particularly the vicinal (^{3}J) coupling constants in **2.81** H3-H4 and H4-H5, was complicated by the 4-fluorine atom.

Scheme 2.21 Synthesis of fragment **2.79**



Scheme 2.22 Acetonide analysis of **2.78**



2.5.3.3 Future directions and Proposed Synthesis of 2.1b

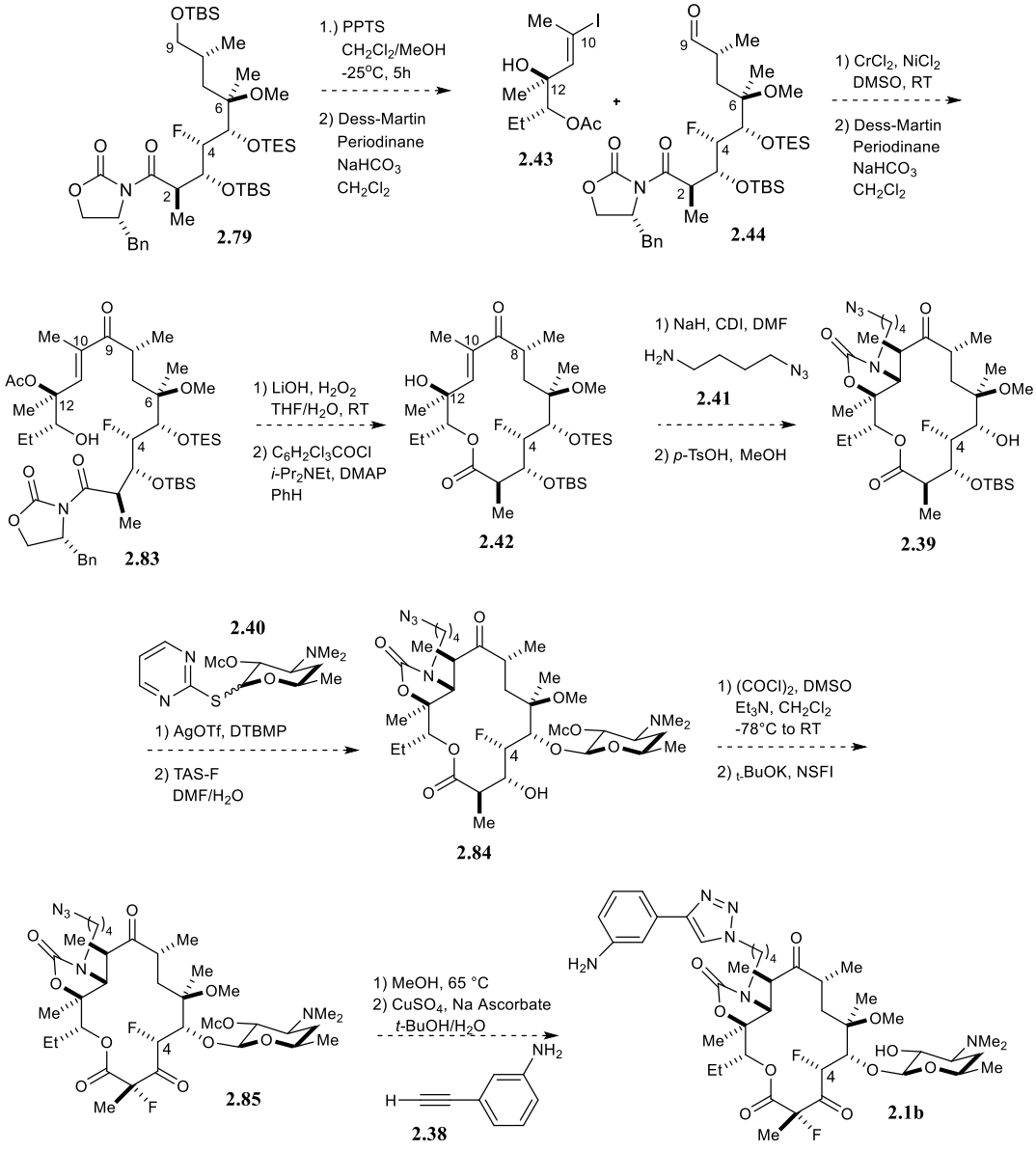
Chemoselective removal of the primary TBS group and Dess-Martin oxidation will furnish **2.44.** The synthesis of vinyl iodide **2.43** was established throughout the desmethyl synthesis series (see Chapter 1). Iodide **2.43** has been accomplished in 12 steps from commercially available methacrolein in 3.2% overall yield.[1,64,69]

Following the successful route developed for 4-desmethyl telithromycin, an intermolecular NHK reaction would be employed to join fragments **2.44** and iodide **2.43** to furnish **2.83**. Following auxiliary removal with LiOOH, the Yamaguchi macrolactonization protocol will be employed to prepare macrolactones **2.42**. The remainder of the synthesis features reactions used to prepare desmethyl analogs (see Chapter 1). It was observed that once the macrolactone stage has been reached in the synthesis, the remaining steps proceed largely without incident. Accordingly, Baker cyclization of **2.42** with NaH, CDI, and 4-azidobutylamine **2.41** followed by acid-mediated TES removal at O-5 will access acceptor **2.39**.

Glycosylation with desosamine donor **2.40** under Woodward's conditions and TBS removal of O3 will give **2.84**. If donor **2.40** is not compatible with the C2 fluoro group, recourse to *p*-tolyl thioglycosides will be made since Optimer demonstrated they could glycosylate the 5-OH position bearing the C2-fluoro moiety.[2] Swern[70] or Corey-kim[71] oxidation of **2.84** will furnish a β-keto ester intermediate; site-selective deprotonation with *t*-BuOK and reaction with NFSI will stereoselectively install the C2-fluoro moiety in **2.85**

ndgame for (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1b**) will consist of carbonate removal and the Click reaction with 3-ethynyl aniline (**2.38**).

Scheme 2.23 Proposed completion of the synthesis of (*R*)-4-fluoro-4-desmethyl solithromycin (**2.1b**) from **2.79**



2.6 Conclusion

In summary, progress was made toward the synthesis of (*R*)-4-fluoro-4-desmethyl solithomycin (**2.1b)** wherein partners **2.43** and **2.79** were synthesized utilizing a key asymmetric fluoroaldol reaction based on the Evans oxazolidinone. However, the synthesis was paused at this stage due to a change in research objectives and are discussed in Chapters 3 and 4.

References

1. Glassford, I.; Lee, M.; Wagh, B.; Velvadapu, V.; Paul, T.; Sandelin, G.; DeBrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Amdrade, R. B. *ACS Med. Chem. Lett.* **2014**, *5*, 1021–1026.

2. Liang, C. H.; Yao, S.; Chiu, Y. H.; Leung, P. Y.; Robert, N.; Seddon, J.; Sears, P.; Hwang, C. K.; Ichikawa, Y.; Romero, A. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 1307–1310.

3. Rostovtsev, V. V.; Green, L. G.; Fokin, V. V.; Sharpless, K. B. *Angew. Chemie - Int. Ed.* **2002**, *41*, 2596–2599.

4. Llano-Sotelo, B.; Dunkle, J.; Klepacki, D.; Zhang, W.; Fernandes, P.; Cate, J. H. D.; Mankin, A. S. *Antimicrob. Agents Chemother.* **2010**, *54*, 4961–4970.

5. Moissan, H. *Compt. Rend.* **1886**, *102*, 1534.

6. Cruikshank, A. J.; Cowper, G.; Grummitt, W. E. *Can. J. Chem.* **1956**, *34*, 214–219.

7. Müller, K.; Faeh, C.; Diederich, F. *Science.* **2007**, *317*, 1881–1886.

8. O'Hagan, D. *Chem. Soc. Rev.* **2008**, *37*, 308–319.

9. Dunitz, J. D.; Taylor, R. *Chem. - A Eur. J.* **1997**, *3*, 89–98.

10. Howard, J. A. K.; Hoy, V. J.; O'Hagan, D.; Smith, G. T. *Tetrahedron.* **1996**, *52*, 12613–12622.

11. Dunitz, J. D. *ChemBioChem.* **2004**, *5*, 614–621.

12. Fröhlich, R.; Rosen, C. T.; Meyer, Oliver, G. J.; Rissanen, K.; Haufe, G. *J. Mol. Struct.* **2006**, *787*, 50–62.

13. Olsen, J. A.; Banner, D. W.; Seiler, P.; Sander, U. O.; D'Arcy, A.; Stihle, M.; Müller, K.; Diederich, F. *Angew. Chemie. Int. Ed.* **2003**, *42*, 2507–2511.

14. Olsen, J. A.; Banner, D. W.; Seiler, P.; Wagner, B.; Tschopp, T.; Obst-Sander, U.; Kansy, M.; Müller, K.; Diederich, F. *ChemBioChem* **2004**, *5*, 666–675.

15. Olsen, J.; Seiler, P.; Wagner, B.; Fischer, H.; Tschopp, T.; Obst-Sander, U.; Banner, D. W.; Kansy, M.; Müller, K.; Diederich, F. *Org. Biomol. Chem.* **2004**, *2*, 1339–1352.

16. Bondi, A. *J. Phys. Chem.* **1964**, *68*, 441–451.

17. O'Hagan, D.; Rzepa, H. S. *Chem. Commun.* **1997**, 7, 645–652.

18. Goodman, L.; Gu, H.; Pophristic, V. *J. Phys. Chem. A.* **2005**, *109*, 1223–1229.

19. Hunter, L. *Beilstein J. Org. Chem.* **2010**, *6*, 1–14.

20. Smart, B. E. *J. Fluor. Chem.* **2001**, *109*, 3–11.

21. Harper, D. B.; O'Hagan, D. *Nat. Prod. Rep.* **1994**, *11*, 123–133.

22. Liang, T.; Neumann, C. N.; Ritter, T. *Angew. Chem. Int. Ed.* **2013**, *52*, 8214–8264.

23. Gingras, M. *Tetrahedron Lett.* **1991**, *32*, 7381–7384.

24. Jeffery, T. *Tetrahedron Lett.* **1992**, *33*, 1989–1992.

25. Martínez, A. G.; Osío, J. B.; Rys, A. Z.; Subramanian, L. R. *Synlett* **1993**, 587–588.

26. Markovskij, L.N.; Pashinnik, V. E.; Kirsanov, A. V. *Synthesis.* **1973**, 787–789.

27. Middleton, W. J. *J. Org. Chem.* **1975**, *40*, 574–578.

28. Rosenbrook, W.; Riley, D. A.; Lartey, P. A. *Tetrahedron Lett.* **1985**, *26*, 3–4.

29. Hudlicky, M. *Organic Reactions*. 1988, 513–637.

30. Kozikowski, A. P.; Fauq A. H.; Rusnak. J. M.; *Tetrahedron Lett.* **1989**, *30*, 3365–3368.

31. Lal, G. S.; Pez, G. P.; Pesaresi, R. J.; Prozonic, F. M.; Cheng, H. *J. Org. Chem.* **1999**, *64*, 7048–7054.

32. Lal, G. S.; Lobach, E.; Evans, A. *J. Org. Chem.* **2000**, *65*, 4830–4832.

33. Singh, R. P.; Twamley, B.; Shreeve, J. M. *J. Org. Chem.* **2002**, *67*, 1918–1924.

34. White, J. M.; Tunoori, A. R.; Turunen, B. J.; Georg, G. I. *J. Org. Chem.* **2004**, *69*, 2573–2576.

35. Umemoto, T.; Singh, R. P.; Xu, Y.; Saito, N. *J. Am. Chem. Soc.* **2010**, *132*, 18199–18205.

36. Zhou, C.; Li, J.; Lu, B.; Fu, C.; Ma, S. *Org. Lett.* **2008**, *10*, 581–583.

37. Davis, F. A.; Han, W. *Tetrahedron Lett*. **1991**, *32*, 1631–1634.

38. Davis, F. A.; Han, W.; Murphy, C. K. *J. Org. Chem.* **1995**, *60*, 4730–4737.

39. Nie, J.; Zhu, H. W.; Cui, H. F.; Hua, M. Q.; Ma, J. A. *Org. Lett.* **2007**, *9*, 3053–3056.

40. Davis, F. A.; Han, W. *Tetrahedron Lett*. **1992**, *33*, 1153–1156.

41. Davis, F. A.; Kasu, P. V. N. *Tetrahedron Lett.* **1998**, *39*, 6135–6138.

42. Pridgen, L. N.; Abdel-Magid, A. F.; Lantos, I.; Shilcrat, S.; Eggleston, D. S. *J. Org. Chem.* **1993**, *58*, 5107–5117.

43. Evans, D. A.; Sjogren, E. B.; Bartroli, J.; Dow, R. L. *Tetrahedron Lett.* **1986**, *27*, 4957–4960.

44. Evans, D. A.; Ellman, J. A.; Dorow, R. L. *Tetrahedron Lett.* **1987**, *28*, 1123–1126.

45. Bender, D. A. E. and S. L. *Tetrahedron Lett.* **1986**, *27*, 799–802.

46. Myers, A. G.; Mckinstry, L.; Barbay, J. K.; Gleason, J. L. *Tetrahedron Lett.* **1998**, *39*.

47. Myers, A. G.; Barbay, J. K.; Zhong, B. *J. Am. Chem. Soc.* **2001**, *123*, 7207–7219.

48. Bissantz, C.; Kuhn, B.; Stahl, M. *J. Med. Chem.* **2010**, *53*, 5061–5084.

49. Celmer, W. D. *Pure Appl. Chem.* **1971**, *28*, 413–453.

50. Huisgen, R. *Proc. Chem. Soc.* **1961**, 357–397.

51. Fernandes, P.; Martens, E.; Bertrand, D.; Pereira, D. *Bioorg. Med. Chem.* **2016**, *24*, 6420–6428.

52. Baker, W. R.; Clark, J. D.; Stephens, R. L.; Kim, K. H. *J. Org. Chem.* **1988**, *53*, 2340–2345.

53. Inanaga, J.; Hirata, K.; Saeki, H.; Katsuki, T.; Yamaguchi, M. *Bull. Chem. Soc. Jpn.* **1979**, 1989–1993.

54. Venkatraman, L.; Aldrich, C. C.; Sherman, D. H.; Fecik, R. a. *J. Org. Chem.* **2005**, *70*, 7267–7272.

55. Evans, D.; Bartroli, J.; Shih, T. L. *J. Am. Chem. Soc.* **1981**, *103*, 2109–2127.

56. Smith, A. B.; Mesaros, E. F.; Meyer, E. A. *J. Am. Chem. Soc.* **2006**, *128*, 5292–5299.

57. Umemoto, T.; Tomizawa, G. *Bull. Chem. Soc. Jpn*. 1986, 3625–3629.

58. Stace, B. C. S.; G. J. *J. Chem. Soc.* **1948**, 1773–1779.

59. Shiina, I.; Kubota, M.; Ibuka, R. *Tetrahedron Lett.* **2002**, *43*, 7535–7539.

60. Valeur, E.; Bradley, M. *Chem. Soc. Rev.* **2009**, *38*, 606–631.

61. Kuethe, T. J.; Beutner, L. G. *Org. Synth.* **2009**, *86*, 92.

62. VanRheenen, V.; Kelly, R. C.; Cha, D. Y. *Tetrahedron Lett.* **1976**, *17*, 1973–1976.

63. Kolb, H. C.; VanNieuwenhze, M. S.; Sharpless, K. B. *Chem. Rev.* **1994**, *94*, 2483–2547.

64. Velvadapu, V.; Glassford, I.; Lee, M.; Paul, T.; Debrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2012**, *3*, 211–215.

65. Dess, D. B.; Martin, J. C. *J. Org. Chem.* **1983**, *48*, 4155–4156.

66. González, Á.; Aiguadé, J.; Urpí, F.; Vilarrasa, J. *Tetrahedron Lett.* **1996**, *37*, 8949–8952.

67. Evans, D. A.; Rieger, D. L.; Gage, J. R. *Tetrahedron Lett.* **1990**, *31*, 7099–7100.

68. Rychnovsky, D. S.; Griesgraber, G.; Schlegel, R. *J. Am. Chem. Soc.* **1994**, *116*, 2623–2624.

69. Wagh, B.; Paul, T.; Glassford, I.; Debrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2012**, *3*, 1013–1018.

70. Omura, K.; Swern, D. *Tetrahedron* **1978**, *34*, 1651–1660.

71. Corey, E. J.; Kim, C. U. *J. Am. Chem. Soc.* **1972**, *94*, 7586–7587.

CHAPTER 3

MACROLIDE ANTIBIOTICS WITH DESOSAMINE MODIFICATIONS

3.1 Introduction

Amino sugars wherein of one or more hydroxyl groups have been replaced with an amine are an essential class of carbohydrates found abundantly in nature as monosaccharides. Moreover, they often contribute to a diverse range of biological activities.[1] D-Desosamine (3,4,6-trideoxy-3-dimethylamino-D-*xylo*-hexo-pyranose, **3.1**) is an amino sugar found in a number of macrolide antibiotics including erythromycin (**3.2**), clarithromycin (**3.3**), and azithromycin (**3.4**). It has been identified as critical in the molecular recognition (i.e., binding) of the macrolide to the bacterial ribosome thus conferring its antibacterial activity (Figure 3.1).[2]

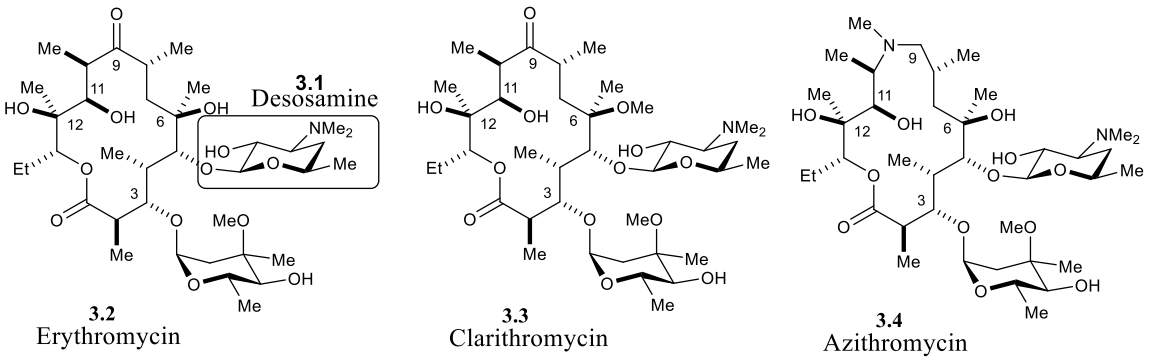


Figure 3.1 Macrolide antibiotics with a desosamine moiety

The desosamine moiety within the macrolides is crucial for its interactions with the peptidyl transferase center of 23S ribosomal RNA. The crystal structures of the 50S ribosomal subunit of the eubacterium *Deinococcus radiodurans* complexed with the clinically useful antibiotic erythromycin (**3.2**) revealed that 2'-hydroxyl group of the desosamine moiety forms hydrogen bonds with nitrogen atoms of adenosine 2058 and 2059 in the peptidyl transferase center. Additionally, the protonated form of 3'- dimethylamino group of desosamine was found to interact with the backbone oxygen of guanosine 2505 in the peptidyl transferase center through ionic interactions.[3]

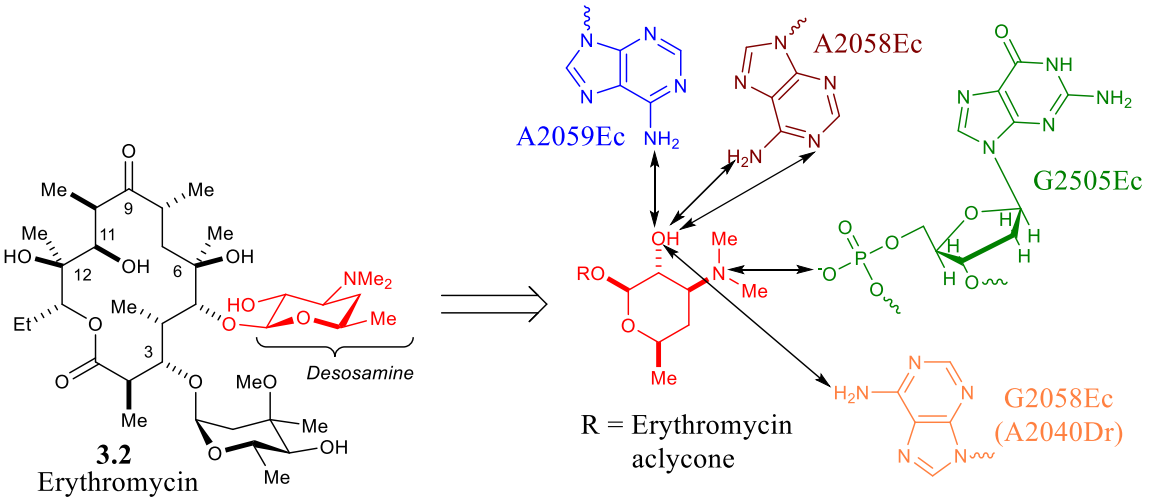


Figure 3.2 Interaction of erythromycin and desosamine within the peptidyl transferase cavity

3.2 Prior Syntheses of Desosamine

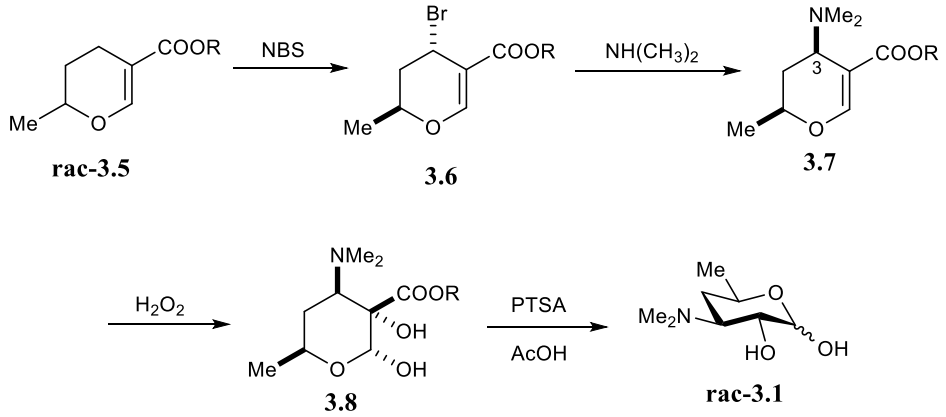
The biological significance of desosamine, coupled with its structural complexity (i.e., three stereogenic centers on the pyranose scaffold, not including the anomeric center)

has attracted the attention of various laboratories. A number of syntheses have been reported, and those approaches will be discussed in this section.

3.2.1 Desosamine Synthesis by Korte (1962)

In 1962, Korte reported a racemic synthesis of desosamine starting from 6-methyl-5,6-dihydro-4H-pyran-3-carboxylate (**3.5**) in four steps (Scheme 3.1).[4] Allylic bromination with NBS followed by the treatment with aqueous dimethylamine gave **3.7** with the amine substituting at the desired C-3 position (Scheme 3.1). Dihydroxylation with hydrogen peroxide followed by decarboxylation gave desosamine (**3.1**).

Scheme 3.1 Korte's approach

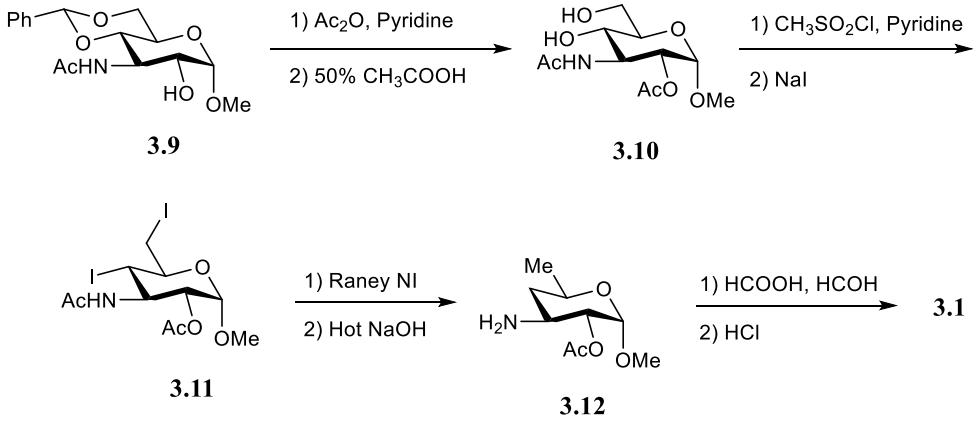


3.2.2 Desosamine Synthesis by Richardson (1964)

In 1964, Richardson reported a synthesis of D-desosamine from methyl 3-acetamido-4,6-*O*-benzylidene-3-deoxy-α-D-glucopyraminoside (**3.9**) that proceeded in eight steps (Scheme 3.2).[5] Acetylation of **3.9** with a mixture of hot acetic anhydride and

pyridine in dimethylformamide gave the 2-*O*-acetate, which was subjected to acidic hydrolysis to remove the benzylidene acetal and furnish diol **3.10**. Treatment of **3.10** with methanesulfonyl chloride followed by NaI gave bis-iodide **3.11**. Subsequent reduction with Raney nickel catalyst and saponification afforded methyl 3,4,6-trideoxy-3-dimethylamino-α-D-*xylo*-hexopyranoside (**3.12**). *N*-methylation under Eschweiler-Clarke conditions (i.e., formic acid and formaldehyde) and hydrolysis of the methyl acetal with aq. HCl furnished **3.1**.

Scheme 3.2 Richardson's approach



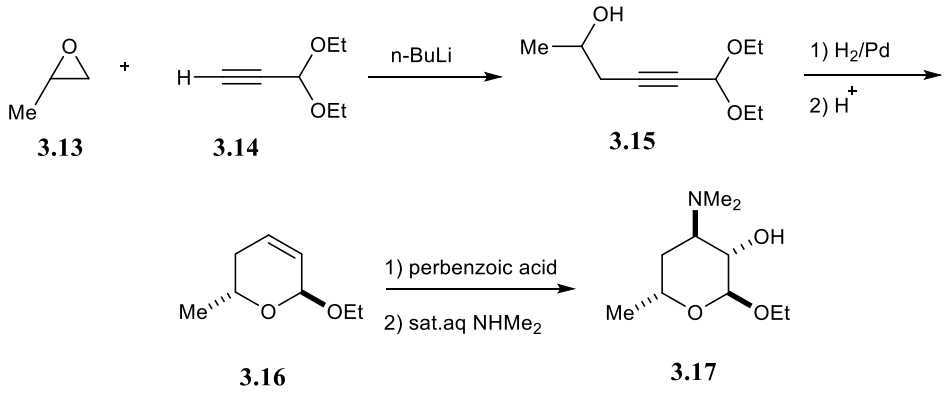
3.9 **3.10** **3.11** **3.12**

3.2.3 Ethyl Desosaminide Synthesis by Newman (1974)

In 1964, Newman reported ethyl desosaminide (i.e., the ethyl glycoside of desosamine) in four steps (Scheme 3.3).[6] 3,3-Diethoxy-1-propyne (**3.14**) reacted with propylene oxide (**3.13**) to give 1,1-diethoxy-5-hydroxyhex-2-yne (**3.15**) in 60% yield. Direct conversion of **3.15** to dihydropyran **3.16** was accomplished by partial reduction of

the alkyne to the *Z*-alkene, followed by acid-mediated catalysis. Epoxidation of **3.16** with perbenzoic acid and subsequent treatment with saturated aqueous dimethylamine gave **3.17**.

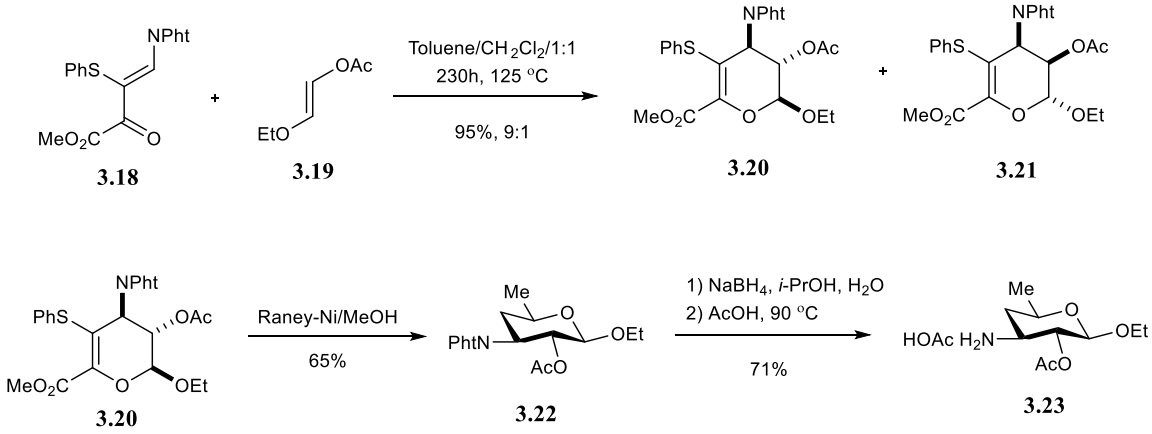
Scheme 3.3 Newman's approach



3.2.4 Ethyl Desosaminide Synthesis by Tietze (1990)

In 1990, Tietze employed a hetero-Diels-Alder reaction to access the pyranose framework of desosamine (Scheme 3.4).[7] Heating a solution of racemic phenylthiol-activated enamino ketone **3.18** and 2-ethoxyvinylacetate **3.19** yielded cycloadducts **3.20** and **3.21** with the major product (9:1 *endo*/*exo*) possessing the desired 3,4-*trans* stereochemistry. The phenylthio substituent in dihydropyran **3.20** was reduced with Raney nickel followed by removal of phthalimido and acetyl groups afforded the acetic acid salt of ethyl desosaminide (**3.23**).

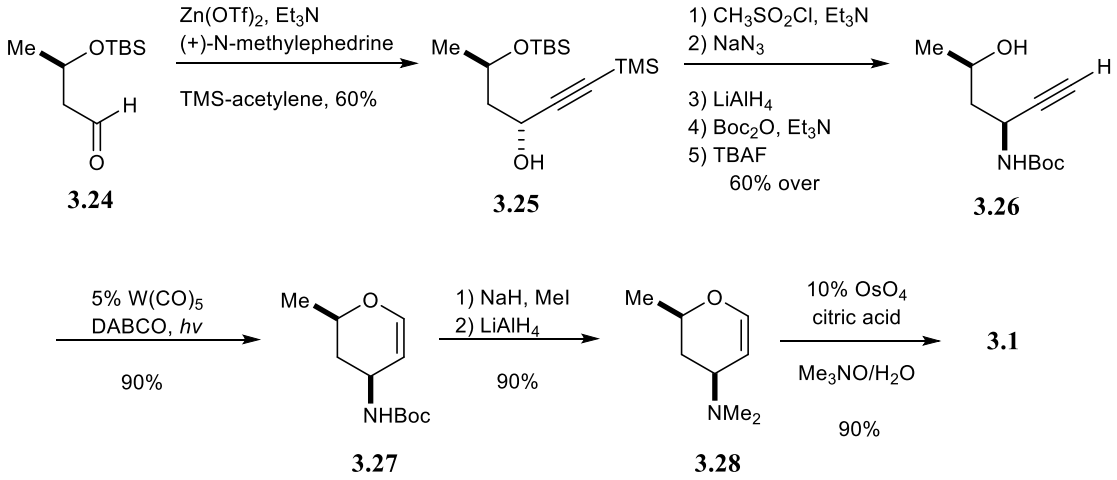
Scheme 3.4 Tietze's approach



3.2.5 Desosamine synthesis by McDonald (2004)

David and McDonald reported a ten step synthesis of D-desosamine in 2004 (Scheme 3.5).[8] This route employed (*R*)-3-*tert*-butyldimethylsiloxy butanal (**3.24**) as the starting material. The addition of TMS-acetylene to **3.24** Carreira's asymmetric alkynylation methodology delivered the secondary alcohol **3.25**.[9] Mesylation of the newly formed alcohol and reaction with sodium azide followed by $LiAlH_4$ reduction incorporated the amine functionality. Acylation of the amine and removal of silyl blocking groups provided alkynol **3.26**. Application of McDonald's tungsten carbonyl-catalyzed cycloisomerization of the corresponding amino alkynol gave **3.27** in 90% yield. Methylation and reduction established C-3 dimethylamine functionality in **3.28**. Dihydroxylation of the olefin using modified Sharpless protocol gave D-desosamine (**3.1**).

Scheme 3.5 McDonald's approach



3.2.6 Desosamine synthesis by Andrade (2005)

In 2005, the Andrade group published a five step route to D-desosamine employing methyl α-D-glucopyranoside (**3.29**) as starting material (Scheme 3.6).[10] Compound **3.29** was treated with sulfuryl chloride in a mixture of pyridine and chloroform, followed by the addition of NaI afforded bis-chloride **3.30**. Reduction with Raney nickel furnished diol **3.31**. Under Mitsunobu conditions, the diol was transformed into 2,3-anhydrosugar **3.32**. Subsequent treatment with aqueous dimethylamine afforded C-3 and C-2 methyl regioisomers in a 6:1 ratio wherein the major C-3, methyl desosaminide (**3.33**), was hydrolyzed under the standard acidic condition, then basic workup with Amberyst A-26(OH) to deliver D-desosamine (**3.1**).

New desosamine analogs were introduced by regioselective ring opening at the C3' position with secondary amines. At the time, these analogs were inaccessible from natural

desosamine. The ring-opening reaction was conducted with pyrrolidine, piperidine, and morpholine in a highly regioselective manner. Exclusively, C3 amino analogs **3.34**–**3.36** were prepared in good yields.

Scheme 3.6 Andrade's approach

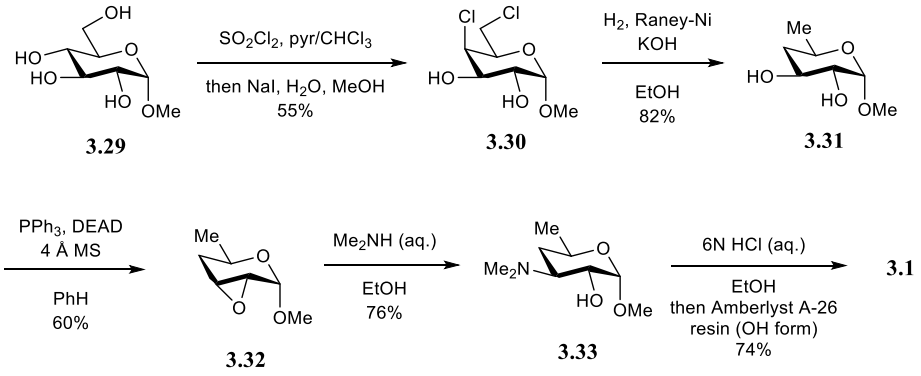
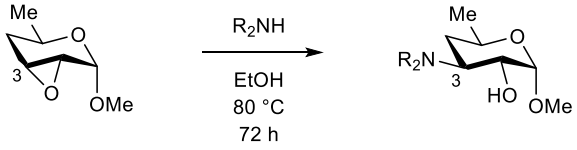
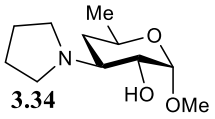
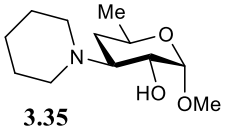
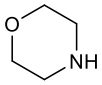
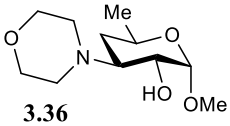


Table 3.1. C3 amino desosamine analogs

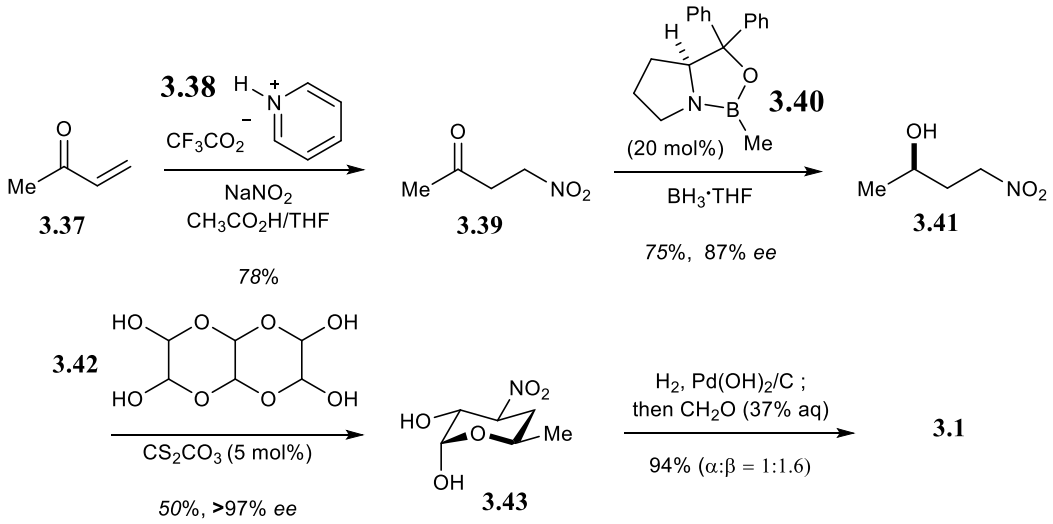


Entry	R_2NH	Product	Yield(%)
1		 **3.34**	86
2		 **3.35**	83
3		 **3.36**	78

3.2.7 Desosamine Synthesis by Myers (2015)

Most recently, Zhang and Myers reported a concise route to D-desosamine from methyl vinyl ketone in 4 steps (Scheme 3.7).[11] Conjugate addition of sodium nitrite to methyl vinyl ketone (**3.37**) delivered 4-nitro-2-butanone (**3.39**). Careful addition of solid pyridinium trifluoroacetate (**3.38**) prevented forming the by-product, 4-acetoxy-2-butanone. Corey-Bakshi-Shibata enantioselective reduction afforded secondary alcohol **3.41** through the slow addition of the ketone to catalyst **3.40** followed by acidic workup. The addition of an aqueous solution of cesium carbonate to a mixture of glyoxal trimer dehydrate (**3.42**) and nitro alcohol **3.41** generated α-D-nitropyranose **3.43** as a white powder. The stereochemistry of **3.43** was confirmed by X-ray crystallography. Hydrogenolysis of **3.43** reduced the nitro group and carried out the reductive alkylation to afford D-desosamine (**3.1**).

Scheme 3.7 Myers's approach



3.3 Clarithromycin

3.3.1 Background

Clarithromycin (i.e., 6-*O*-methyl erythromycin) was invented by Taisho Pharmaceuticals in 1980.[12] Clarithromycin (**3.3**) is derived from erythromycin (**3.2**) by semi-synthesis and features a 14-membered macrocyclic lactone ring attached to two sugar moieties, cladinose on the C3 position and desosamine (**3.1**), in addition to an *O*-methyl ether at the C6 position instead of the hydroxyl group. This modification resulted in greater acid stability than erythromycin, thus improving antimicrobial and pharmacokinetics properties. Clarithromycin is active against aerobic and anaerobic Gram-positive and Gram-negative bacteria. However, the lack of activity against most Gram-negative bacteria is likely due to its inability to penetrate the bacterial cell wall. Clarithromycin is active against many Gram-positive bacterial pathogens. It is used for the treatment of pharyngitis/tonsillitis, acute bacterial exacerbation, acute maxillary sinusitis, skin infections, pneumonia, and with other drugs (proton pump inhibitors and metronidazole) as a triple therapy for treatment of *Helicobacter pylori* infection.[13]

3.3.2. Desosamine Modifications on the Clarithromycin

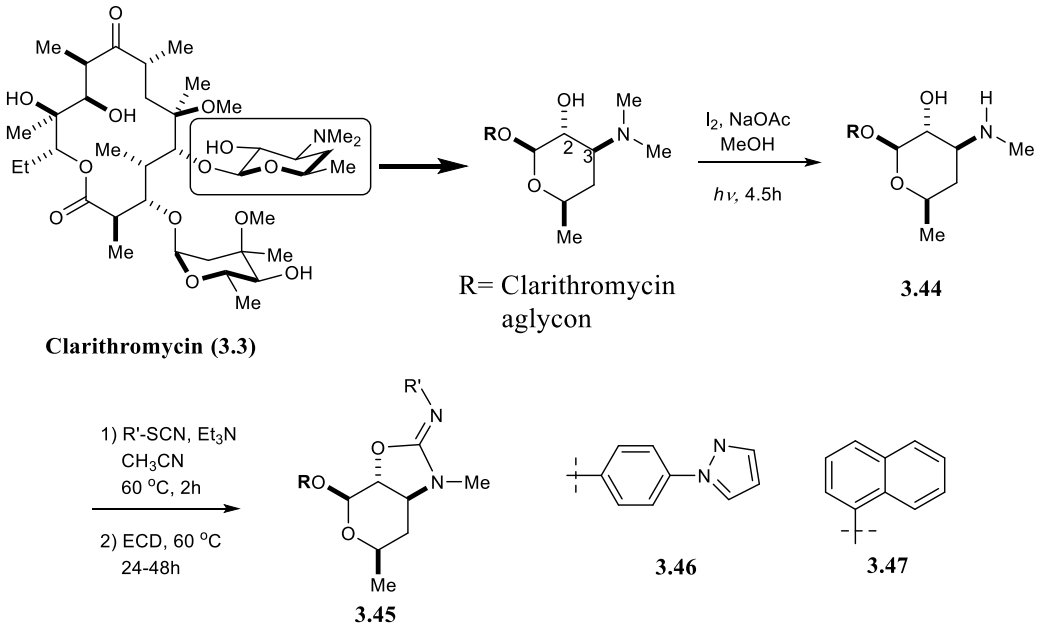
Many innovations have occurred since the discovery of clarithromycin (**3.3**) in 1980. All involved the exploration of chemical space to address physicochemical properties and/or resistance, in addition to the development of intellectual property. One of the most accessible sites in clarithromycin is the cladinose.[14–16] The addition of extra

functional groups or simple removal of cladinose followed by either acylation of oxidation has expanded the family of analogs. Additional innovations include oxime functionalization on the C9 ketone,[17–19] Beckmann-mediated expansion of the ring size from 14 to 15,[20] and derivatization of the C11 and C12 alcohol groups to either access a C11,12 cyclic carbonate or oxazolidinone moieties.[21] Finally, desosamine modifications on clarithromycin have been reported, and those are discussed below.

3.3.2.1 Synthesis of *N'*-substituted 2-Imino-1,3-oxazolidines

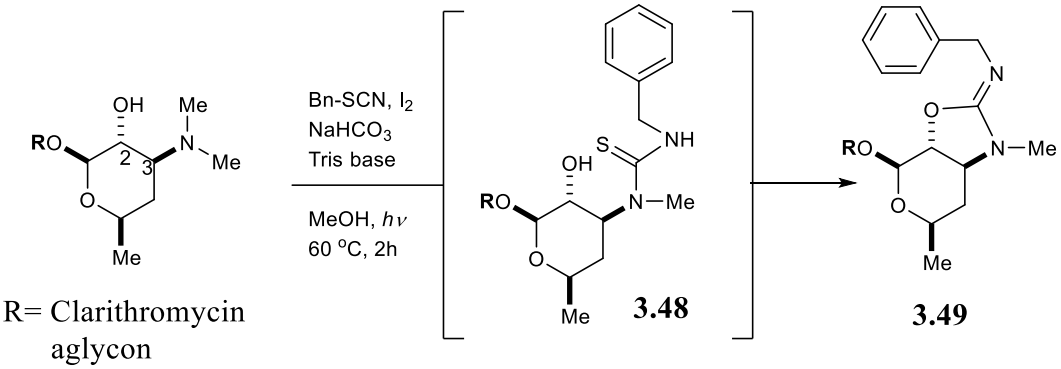
In 2011, Kragol and co-workers reported the synthesis of fused desosamine analogs wherein the vicinal C2/C3 amino alcohol was bridged via a 2-imino-1,3-oxazolidine moiety without altering the macrolactone ring.[20] Oxidative *N*-methylation of clarithromycin (**3.3**) was accomplished by treatment with $NaOAc/I_2$ and irradiation to afford 3'-*N*-desmethyl clarithromycin (**3.44**). The reaction of **3.44** with various substituted isothiocyanates for 2 h followed by the dehydrating agent *N*-(3-dimehtylaminopropyl)-*N*'-ethyl carbodiimide hydrochloride (EDC) afforded the desired *N*'-aryl-2-imino-1,3-oxazolidines **3.46** and **3.47** fused to the desosamine ring with reasonable yields.

Scheme 3.8 Synthesis of *N'*-substituted 2-imino-1,3-oxazolidines



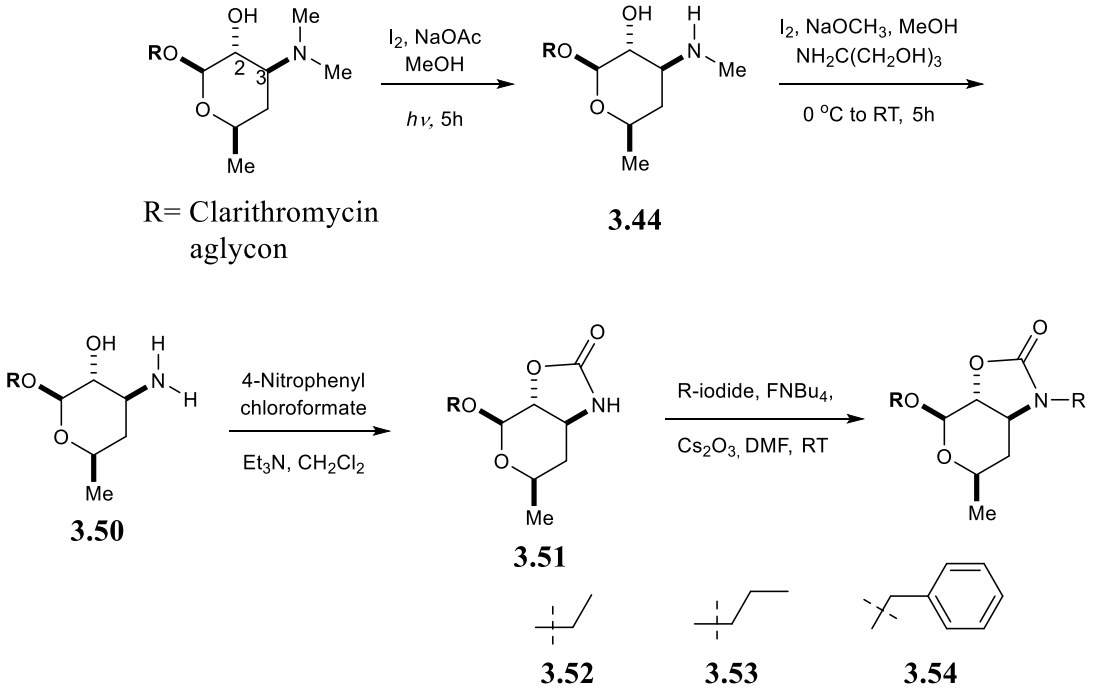
Optimization of the reaction conditions resulted in a novel one-pot tandem reaction for the construction of desosamine bearing a *N*'-benzyl-2-imino-1,3-oxazolidine. First, demethylation of the 3'-amino substituent was followed by the formation of thiourea intermediate **3.49**. Next, a cyclization ensued to generate the *N*'-benzyl-2-imino-1,3-oxazolidine heterocycle. The reagent mixture contained iodine, sodium bicarbonate as a base, tris(hydroxymethyl)aminomethane (Tris base) for quenching the produced formaldehyde, and methanol as solvent. Slightly larger excesses of iodine (6 equiv) and isothiocyanate (5 equiv) were needed but the yield was comparable to stepwise procedures.

Scheme 3.9 Tandem one-pot synthesis of *N*-benzyl-2-imino-1,3-oxazolidine **3.49**.



In 2012, the synthesis of *N*-methyl substituted 1,3-oxazolidin-2-one analogs of desosamine was achieved without altering the macrolide ring.[22] This was an extension of Kragol's previous research. First, the *N*-methyl group in clarithromycin was removed with NaOAc/I_2 to afford **3.44**; however, the second *N*-methyl group proved more challenging to remove. Ultimately, this reaction was achieved using NaOMe to give primary amine **3.50**. Treatment of 3'-*N*,*N*-didesmethyl desosamine (**3.50**) with 4-nitrophenyl chloroformate formed an intermediary 3'-(4-nitrophenyl)carbamate, which cyclized with the 2'-OH to form 1,3-oxazolidin-2-one **3.51**. *N*-alkylation of **3.15** to furnish **3.52-3.54** was realized in the presence of tetrabutylammonium iodide and cesium carbonate. Significantly diminished antibacterial activity of the novel macrolides products was observed by the formation of 1,3-oxazolidin-2-one, as well as *N*-substituted-1,3-oxazolidin-2-ones, which is consistent with the aforementioned requirements for antibacterial activity.

Scheme 3.10 Synthesis of 1,3-oxazolidin-2-one derivatives of desosamine

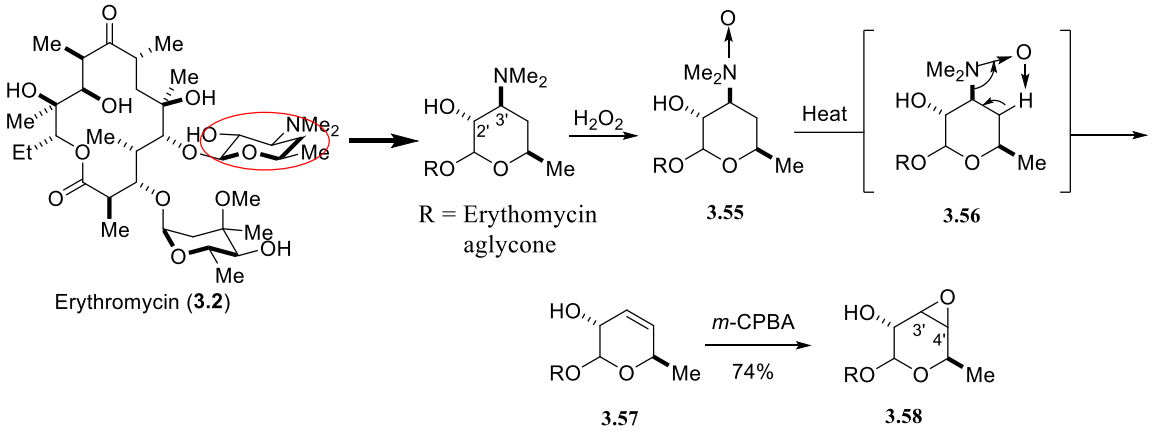


3.4 Epoxide Formation on Desosamine

3.4.1 Cope Elimination

In 1971, Jones reported the preparation of epoxide formation on the desosamine of erythromycin.[23] Erythromycin *N*-oxide (**3.55**) was prepared with dilute hydrogen peroxide followed by recrystallization from methanol/diethyl ether. The *N*-oxide upon heating without solvent for 6 h at 150 °C, was converted into 3'-de(dimethylamino)-3',4'-dehydroerythromycin (**3.57**), as shown in Scheme 3.11. The pyrolytic elimination of *N*,*N*-dimethylhydroxylamine from the erythromycin *N*-oxide (**3.55**) led to the corresponding allylic alcohol via a Cope elimination.[24] Oxidation of the alkene with *m*-chloroperbenzoic acid (MCPBA) gave epoxide **3.58**.

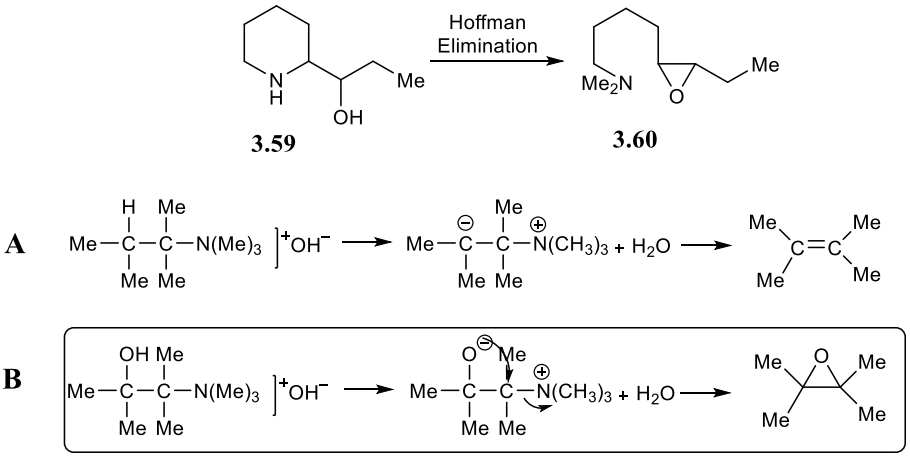
Scheme 3.11 Jones's approach



3.4.2 The Hofmann Elimination Reaction

The Hofmann elimination is an historic reaction that can convert an amine bearing a β-hydrogen to an alkene using methyl iodide and silver oxide under thermal conditions. Thus, the elimination reactions of quaternary ammonium halide salts in the presence of Ag_2O are exemplary Hofmann eliminations.[25] Since the counter anion in quaternary ammonium salts is a halide, ion metathesis to the more basic hydroxide ion with silver oxide is critical. The resulting hydroxide salt then is heated to effect the elimination of the tertiary amine. In 1956, the formation of oxygen-containing heterocycles was reported by Hofmann elimination.[26] Employing Hofmann conditions, epoxide formation was observed in the presence of a neighboring (vicinal) alcohol group (Scheme 3.12).

Scheme 3.12 Hofmann reaction



3.5 Present Study

3.5.1. Motivation

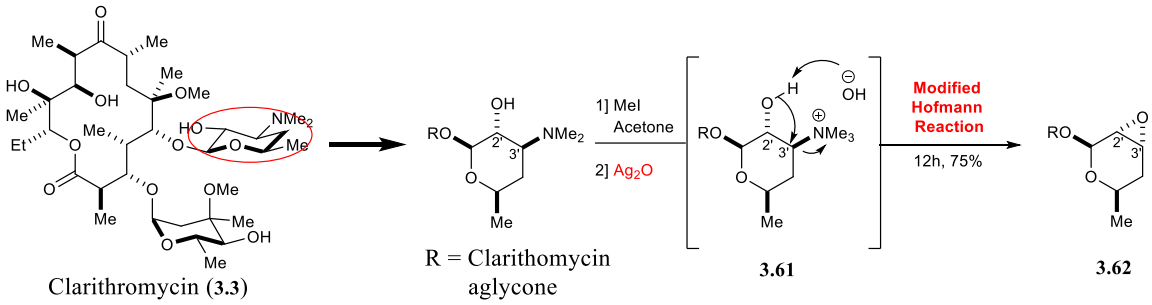
Since desosamine plays pivotal role in the biological activity of macrolide antibiotics, efforts have been directed at both synthesizing and modifying this residue to conduct structure-activity relationship studies. Despite progress in the step-efficient syntheses of desosamine shown above, the need to remove the natural sugar (deglycosylate) and reglycosylate to access novel analogs is cumbersome. The focused modification to the desosamine residue on the macrolide (e.g., clarithromycin) is highly desirable. While such modifications have been reported, they have resulted in analogs lacking antibacterial activity. A goal of the Andrade group was to retain the desosamine moiety and access a new class of desosamine analogs specifically at the 3-amino group. The incorporation of a range of amine substituents into desosamine and biological evaluation of the novel macrolide analogs was therefore pursued.

3.5.2 Desosamine Modification Through a Modified Hofmann Elimination

As previously discussed, the Andrade group reported a concise synthesis of D-desosamine and analogs thereof at the C3' position. However, this method would require an inefficient deglycosylation-reglycosylation protocol. To avoid this, accessing a desosamine residue bearing the 2,3-epoxide was pursued. It was reasoned that employing the Hofmann elimination reaction conditions could accomplish this goal. The first step toward this goal was accessing the quarternary ammonium salt **3.61** by reacting clarithromycin with methyl iodide. In order to make homogenous solution, acetone was used as a solvent. Monitoring of the reaction via TLC to consume the starting material was critical to avoid low yields.

With **3.61** was in hand, several Hofmann reaction conditions were screened (Scheme 3.13). At low temperature, the epoxidation did not proceed. Despite prolonged reaction times, the outcome did not change. Many conditions were screened including varying temperature, solvent, and bases to optimize the reaction. To increase the temperature, toluene was employed as solvent. The reaction was run with microwave irradiation and in a sealed tube. The standard base for Hofmann reaction, silver (I) oxide, was the most effective amongst others. The reaction time was adjusted with TLC monitoring. Finally, the Hofmann elimination reaction when applied to clarithromycin (**3.3**) allowed gram-scale access to 2,3-epoxide **3.62** in 75% yield (Scheme 3.13). Full characterization of 2D NMR analysis was employed to confirm the structure of **3.62**.

Scheme 3.13 Modified Hofmann reaction



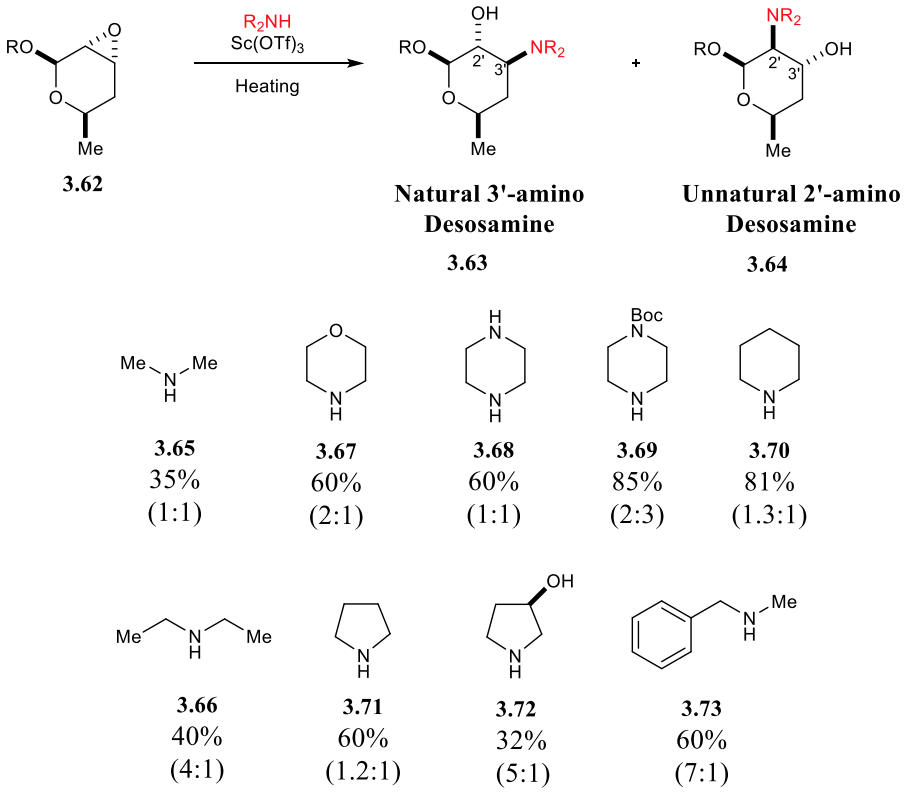
	Solvent	Base	Temperature
Trials	MeOH, DCM, H_2O, DMF, Toluene	Ag_2O, Ag_2CO_3, DBU, AgOTf, Et_3N, DIPEA, Basic Al_2O_2	65 °C to 120 °C, Microwave

3.5.3 Epoxide Ring-opening

With successful access to the requisite 2,3-anhydro target **3.62**, attention was directed at introducing new amines at the C3' position. To test the method and confirm the structure of **3.62**, dimethylamine was the first reactant because the resulting ring-opened product would be clarithromycin (**3.3**). Based on the ring-opening methodology of secondary amines and 2,3-anhydrosugars related to desosamine (see Table 1.1 above), regioselective ring-opening would deliver only natural 3'-amino desosamine **3.63**. Surprisingly, the reaction with dimethylamine was not regioselective. In fact, both natural 3'-amino desosamine **3.63** and unnatural 2'-amino desosamine were isolated in a 1:1 ratio. We reasoned the addition of a Lewis acid would facilitate the ring-opening and perhaps steer the reaction to the desired 3'-amino regioisomer. Screening reaction conditions

ultimately revealed that heating **3.62** with a secondary amine and 10 mol% $Sc(OTf)_3$ gave a mixture of 3'-amino analog **3.63** and novel 2'-amino analog **3.64**. Using those conditions did not change the 1:1 regiochemistry of the reaction with dimethylamine. The reaction temperature was varied based on amine's volatility. Most secondary amine reactions were conducted in toluene under reflux at 120 °C except for dimethylamine (65 °C).

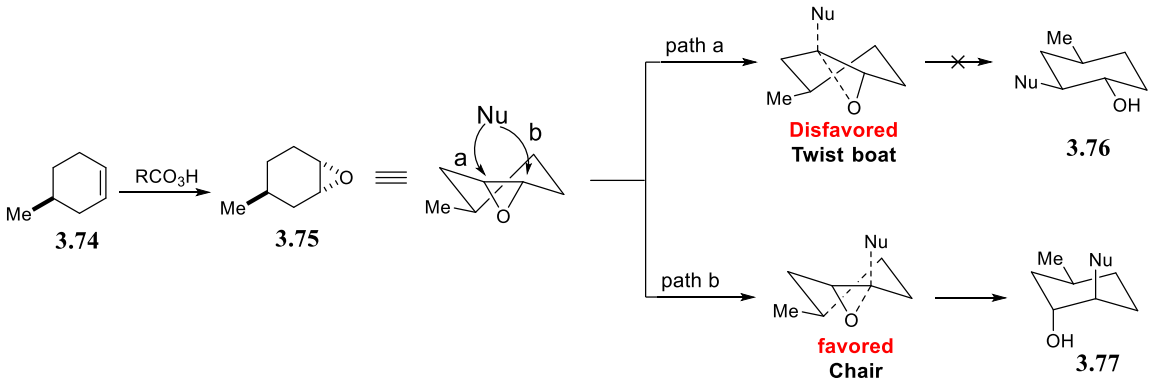
Scheme 3.14 Amines prepared with ratios (C3'/C2')



3.5.4 Rationale for Regioselectivity

In 1951, Fürst and Plattner formulated the "rule for the bi-axial opening of the epoxide ring."[27] The opening of an epoxide ring always takes place in such a way that the nucleophile and the alcohol group in the product are *trans*-oriented (Fürst–Plattner rule). Route B scheme 3.15, in which the transition state is chair like, yielding product **3.77**, will be favored over route A, in which the transition state is boat like. The boat conformer, the initial product in route B, will readily change to the chair conformer **3.76**. However, the thermodynamically stable product was not observed with cyclohexene oxide system. Route B represents "diaxial" opening of the epoxide ring and is energetically preferred over route A, the so-called "equatorial" opening.

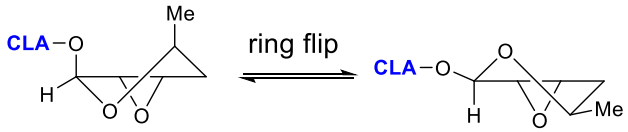
Scheme 3.15 Fürst–Plattner rule



Epoxide ring-opening in a system with a rigid conformation, for example in a steroid framework, takes place under the Fürst–Plattner rule preferentially. However, epoxide ring-opening in systems with a flexible conformation can proceed via additional

pathways to afford regioisomeric products. The desosamine epoxide on the clarithromycin has a flexible conformation and can exist in two forms by ring flip (Figure 3.3).[28]

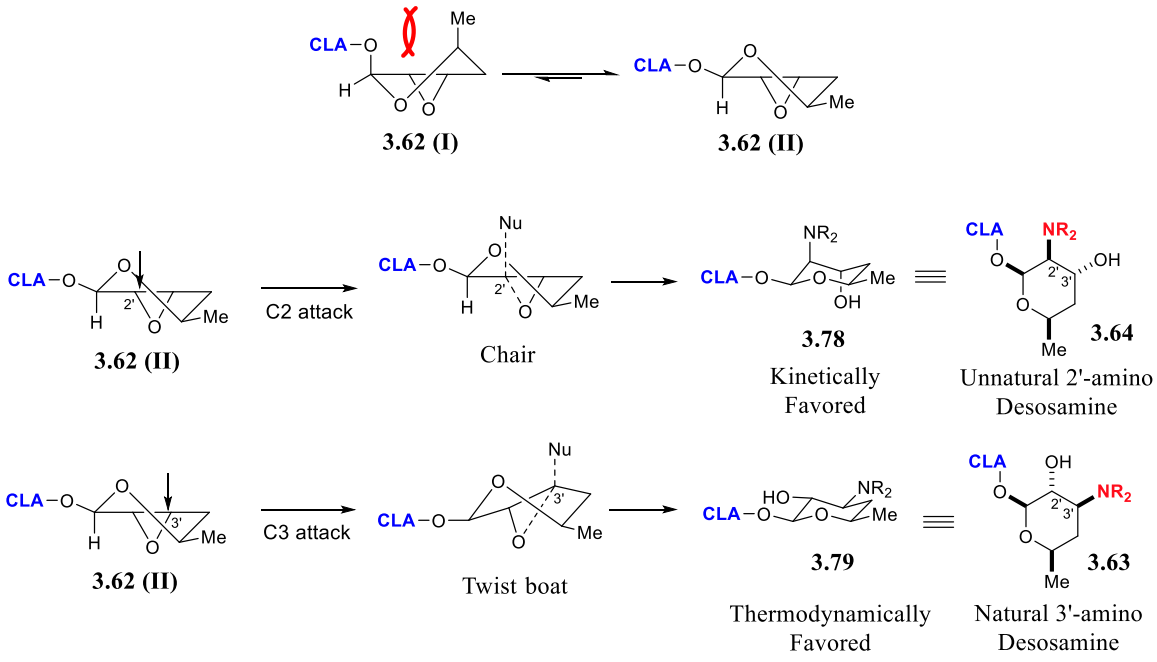
Figure 3.3 Two conformations of desosamine epoxide



3.62 (I) **3.62 (II)**

If it is accepted that axial attack is the rule, either form may react and the products can then ring flip into their most stable conformation. From this point of view, it is unnecessary to postulate "exceptions" to Fürst and Plattner's rule. There are two conformations of epoxide [**3.62(I)** and **3.62(II)**] via ring flip. However, **3.62(I)** conformation was not preferred because of the repulsive 1,3-diaxial steric interaction between the large macrolide ring and the methyl group. The reaction of desosamine epoxide **3.62(II)** with dimethyl amine gave C2/C3 substituted products with 1:1 ratio whereas with bulkier amines, the C3 substituted product **3.63** was formed as a major product (Scheme 3.14). This is probably due to a simple steric effect. The formation of a diaxial product necessitates attack at C2 and in the case of methyl benzyl amine, such an attack may well be subject to steric hindrance by the adjacent bulky macrolactone ring. Therefore, it attacks C3 which is farther away and less sterically hindered which gives C3 natural desosamine. This is consistent with the size of the secondary amine. The bigger the amine, the more C3 attack would happen.

Scheme 3.16 Rationale for observed regioselectivity



The stereochemistry of the opening of epoxy compounds is determined by S_N2 processes. The stereochemical requirements for an S_N2 mechanism arise in the formation of a *trans*-coplanar transition state as a result of an axial attack by the nucleophilic reagent on one of the carbon atoms of the epoxide with subsequent inversion of the configuration of the center being attacked.

3.6 Biological Evaluation of Novel Clarithromycin Analogs

With desosamine analogs in hand, minimum inhibitory concentrations (MIC) were determined against five strains of *Escherichia coli* (Table 3.1) and four strains of *Staphylococcus aureus* (Table 3.2). Unnatural dimethyl desosamine analog **3.65(C'2)** and

natural *N*-methyl-*N*'-benzylamine desosamine analog **3.73(C'3)** were comparable with clarithromycin. Unnatural (*R*)-3-hydroxyl pyrrolidine desosamine analog **3.72(C'2)** showed better activity than clarithromycin against the *Staphylococcus aureus* wild type strain. This meant having different secondary amine on 2' position explored new binding space to have the equal or better potent as clarithromycin.

Table 3.2 MIC analysis of novel clarithromycin analogs against *Escherichia coli*

MIC(µg/ml)	*Escherichia coli*				
Compound #	DK	DK 3535	DK 2058G	SQ 171	SQ 171 2058G
CLA **3.65a**	4	4	8	>128	>128
3.62	>128	>128	>128	>128	>128
3.65b (C2′)	4	4	16	>128	>128
3.66a (C3′)	>128	>128	>128	>128	>128
3.66b (C2′)	>128	>128	>128	>128	>128
3.67a(C3′)	>128	>128	>128	>128	>128
3.67b (C2′)	>128	>128	>128	>128	>128
3.68a (C3′)	>128	>128	>128	>128	>128
3.68b (C2′)	>128	>128	>128	>128	>128
3.69a (C3′)	>128	>128	>128	>128	>128
3.69b (C2′)	>128	>128	>128	>128	>128
3.70a (C3′)	>128	>128	>128	>128	>128
3.70b (C2′)	>128	>128	>128	>128	>128
3.71a (C3′)	64	64	64	>128	>128

3.71b (C2′)	>128	>128	>128	>128	>128
3.72a (C3′)	64	64	128	>128	>128
3.72b(C2′)	16	16	8	>128	>128
3.73a (C3′)	4	4	16	>128	>128
3.73b (C2′)	>128	>128	>128	>128	>128

Table 3.3 MIC analysis of novel clarithromycin analogs against against *Staphylococcus aureus*

MIC(µg/ml)	*Staphylococcus aureus*			
Compound #	SA ATCC WT	UNC 14 A2058U	UCN 17 A2058G	UCN 18 A2059G
CLA **3.65a**	2	>512	>512	>512
3.62	>512	>512	>512	>512
3.65b (C2′)	4	>512	>512	>512
3.66a (C3′)	512	>512	>512	>512
3.66b (C2′)	512	>512	>512	>512
3.67a(C3′)	>512	>512	>512	>512
3.67b (C2′)	>512	>512	>512	>512
3.68a (C3′)	>512	>512	>512	>512
3.68b (C2′)	8	>512	>512	>512
3.69a (C3′)	>512	>512	>512	>512
3.69b (C2′)	>512	>512	>512	>512
3.70a (C3′)	>512	>512	>512	>512
3.70b (C2′)	128	>512	>512	>512
3.71a (C3′)	32	>512	>512	>512
3.71b (C2′)	128	>512	>512	>512

3.72a (C3′)	64	>512	>512	>512
3.72b(C2′)	<0.125	>512	>512	>512
3.73a (C3′)	2	>512	>512	>512
3.73b (C2′)	>512	>512	>512	>512

3.7 Conclusion

Previous desosamine modifications at the 3'-amino position have been limited to monosubstitution via sequential dealkylation/*N*-alkylation. To date, the Andrade group has prepared a small library of clarithromycin analogs **65**–**73** by our $Sc(OTf)_3$-catalyzed ring-opening of epoxide **3.62** with secondary amines in good to excellent yields and varying ratios of regioisomeric C3'/C2' products. This approach markedly reduces the steps, time, and cost involved in preparing novel desosamine-modified analogs. Significantly, this novel route enables the first synthesis of *N*, *N'*-disubstituted desosamine analogs from epoxide **3.62**. Since clarithromycin is the most common starting material for semi-synthesis of macrolides, this method can be utilized to prepare novel analogs to address the problem of antibacterial resistance.

3.8 References

1. Hudlicky, T.; Entwistle, D. A.; Pitzer, K. K.; Thorpe, A. J. *Chem. Rev.* **1996**, *96*, 1195–1220.

2. Jelić, D.; Antolović, R. *Antibiotics* **2016**, *5*, 29.

3. Schlünzen, F.; Zarivach, R.; Harms, J.; Bashan, A.; Tocilj, A.; Albrecht, R.; Yonath, A.; Franceschi, F. *Nature* **2001**, *413*, 814–821.

4. Issleib, K. *Tetrahedron* **1984**, *18*, 261.

5. Richardson, C. A. *J. Chem. Soc.* **1964**, 5364–5370.

6. Newman,.H. *J. Org. Chem.* **1964**, *29*, 1461–1468.

7. Tietze, L. F.; Haqtiel, U. *Tetrahedron Lett.* **1990**, *31*, 1697–1700.

8. Davidson, M. H.; McDonald, F. E. *Org. Lett.* **2004**, *6*, 1601–1603.

9. Frantz, D. E.; Fässler, R.; Carreira, E. M. *J. Am. Chem. Soc.* **2000**, *122*, 1806–1807.

10. Velvadapu, V.; Andrade, R. B. *Carb.res* **2008**, *343*, 145–150.

11. Zhang, Z.; Fukuzaki, T.; Myers, A. G. *Angew. Chemie - Int. Ed.* **2016**, *55*, 523–527.

12. Morimoto, S.; Takahashi, Y.; Watanabe, Y. S. O. *J. Antibiot.* **1984**, *37*, 187–189.

13. Rodvold, K. a. *Clin. Pharmacokinet.* **1999**, *37*, 385–398.

14. Kapić, S.; Ipić Paljetak, H.; Alihodić, S.; Antolović, R.; Eraković Haber, V.; Jarvest, R. L.; Holmes, D. J.; Broskey, J. P.; Hunt, E. *Bioorg. Med. Chem.* **2010**, *18*, 6569–6577.

15. Liang, J. H.; Wang, Y. Y.; Wang, H.; Li, X. L.; An, K.; Xu, Y. C.; Yao, G. W. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 2880–2883.

16. Zhu, D.; Xu, Y.; Liu, Y.; Chen, X.; Zhao, Z.; Lei, P. *Bioorg. Med. Chem. Lett.* **2013**, *23*, 6274–6279.

17. Morimoto, S.; Moriguchih, I. *Chem. Pharm. Bull.* **1994**, *42*, 1088–1095.

18. Nam, G.; Kang, T. W.; Shin, J. H.; Choi, K. Il. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 569–572.

19. Brunet, E.; Muñoz, D. M.; Parra, F.; Mantecón, S.; Juanes, O.; Rodríguez-Ubis, J. C.; Carmen Cruzado, M.; Asensio, R. *Tetrahedron Lett.* **2007**, *48*, 1321–1324.

20. Vujasinović, I.; Marušić Ištuk, Z.; Kapić, S.; Bukvić Krajačić, M.; Hutinec, A.; Dilović, I.; Matković-Čalogović, D.; Kragol, G. *European J. Org. Chem.* **2011**, *13*, 2507–2518.

21. Letourneau, N.; Vimal, P.; Klepacki, D.; Mankin, A.; Melman, A. *Bioorg. Med. Chem. Lett.* **2012**, *22*, 4575–4578.

22. Jakopović, I. P.; Krajačić, M. B.; Škugor, M. M.; Štimac, V.; Pešić, D.; Vujasinović, I.; Alihodžić, S.; Paljetak, H. Č.; Kragol, G. *Bioorg. Med. Chem. Lett.* **2012**, *22*, 3527–3530.

23. Jones, P. H. Erythromycin ester derivatives, US 3598805 A. **1971**.

24. Cope, C. A.; Ciganek, E. *Org. Synth.* **1959**, 4, 40–43.

25. Hofmann, V. A. W. *Ann. der Chemie und Pharm.* **1851**, *78*, 253–286.

26. Hans Werner Bersch, G. H. *Arch. Pharm.* **1956**, *289*, 673–676.

27. Fürst, A.; Plattner, P. A. *Helv. Chim. Acta* **1949**, *32*, 275–283.

28. Newth, H. F. *Q. Rev. Chem. Soc* **1959**, *13*, 30–47.

CHAPTER 4

THE DEVELOPMENT OF LEAD COMPOUNDS FOR RIBOSOME-TEMPLATED *IN SITU* CLICK CHEMISTRY

4.1 Solithromycin

4.1.1 Discovery of Solithromycin

Despite the fact that many macrolide and ketolide (i.e, 3-keto macrolide) candidates have failed to reach late stage clinical development, many pharmaceutical companies have expended considerable efforts to develop a lead macrolide that was as potent as or better than telithromycin. Hundreds of macrolides and ketolides were made and tested by Optimer pharmaceuticals. They synthesized a novel 5-membered 1,2,3-triazole ring with copper Click chemistry, which delivered metabolic stability as an advantage. Cempra pharmaceuticals licensed the entire macrolide program from Optimer to develop a lead candidate. There were three possible candidates OP-1068, OP1055, the des-2-fluorine analog of OP-1068, and a third molecule that had only a phenyl group at the terminus of the side chain (Figure 4.1). OP-1068 (code CEM-101), which later was named solithromycin, emerged as the most potent erythromycin-based congener in both *in vitro* and animal infection models, including activity against telithromycin-resistant bacterial strains.[1]

4.1.2 Binding Study of Solithromycin to Bacterial Ribosomes

The binding conformation of solithromycin to bacterial ribosomes has been confirmed by X-ray crystallography (Figure 4.2).[1,2] The side chain aniline and 1,2,3-

triazole provided multiple ribosomal interactions (B) that were not observed previously. The desosamine sugar of solithromycin showed the same manner of interaction as other macrolides (e.g., erythromycin and clarithromycin). The 2-fluorine of solithromycin also interacts with the ribosome which provides a third interaction site and contributes to improved pharmacokinetics (D).

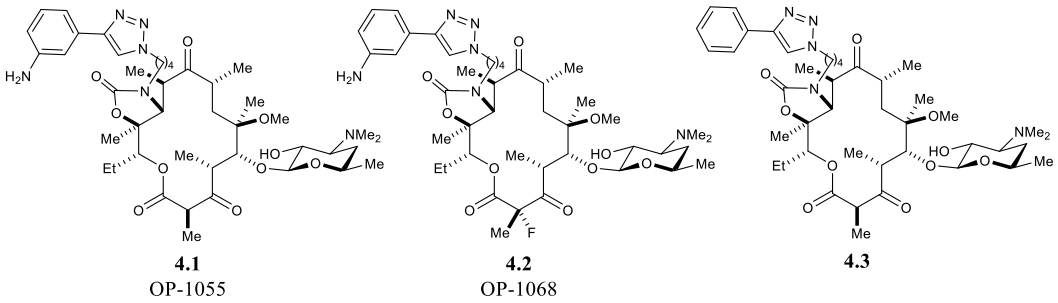


Figure 4.1 Chemical structure of solithromycin (OP-1068) and OP-1055

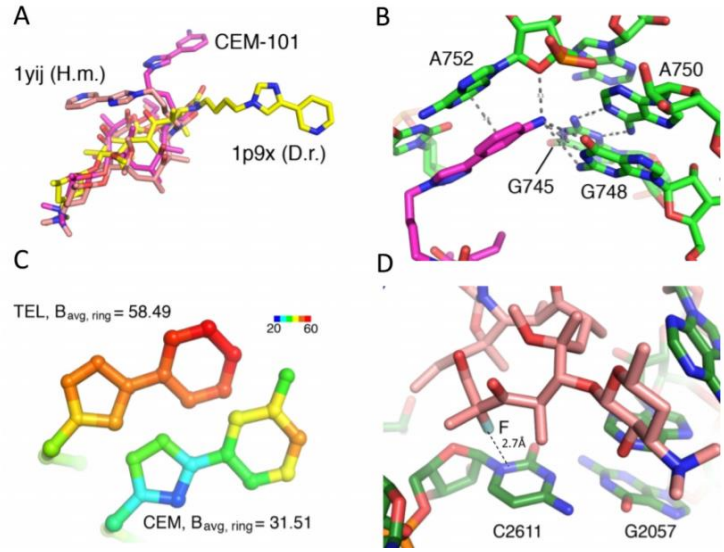
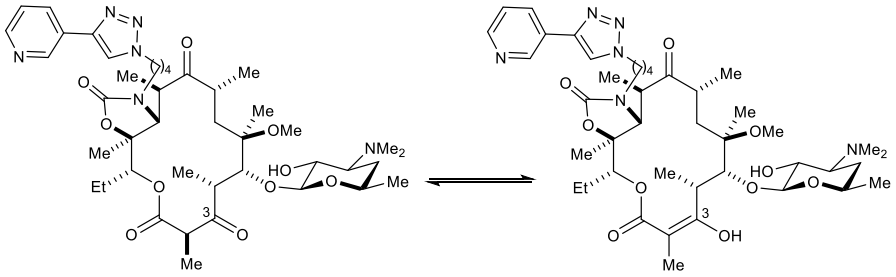


Figure 4.2. Crystallographic structure of solithromycin bound to the *E. coli* ribosome at 3.2 Å. (A) Solithromycin (pink) and telithromycin (yellow) positions of the macrolactone ring and the alkyl aryl side chain in interactions with the ribosome; (B) Solithromycin alkyl aryl side chains interactions with the ribosome; (C) Solithromycin and telithromycin interactions with the ribosome; (D) Proximity of solithromycin's fluorine to base pair 2611-G2057 of the ribosome

Previously, the enolization from C3-keto was observed with other ketolides. The presence of the 2-fluorine precludes enolization and provides enhanced activity against telithromycin-resistant strains and pharmacokinetics (Figure 4.3). Solithromycin is a fourth generation macrolide and the first fluoroketolide which is in phase 3 clinical trial.[3]



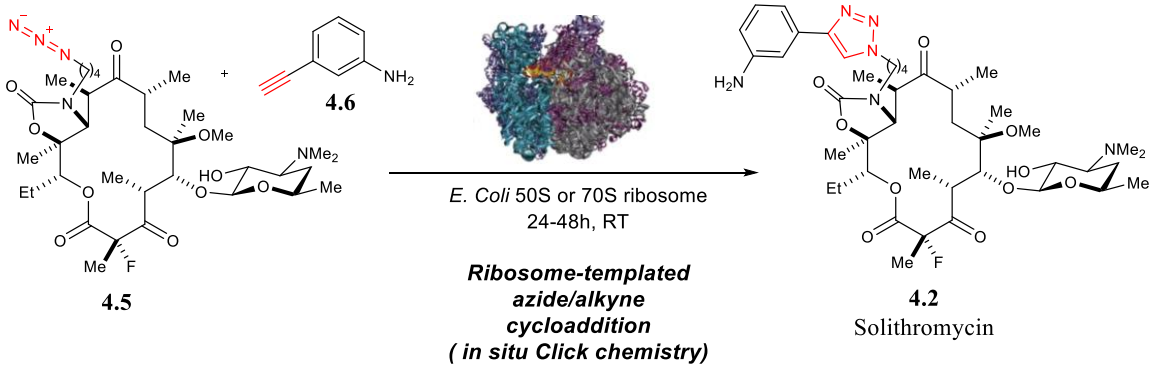
4.4a Telithromycin **4.4b**

Figure 4.3 Equilibrium between keto (**4.4a)** and enol (**4.4b)** forms in telithromycin

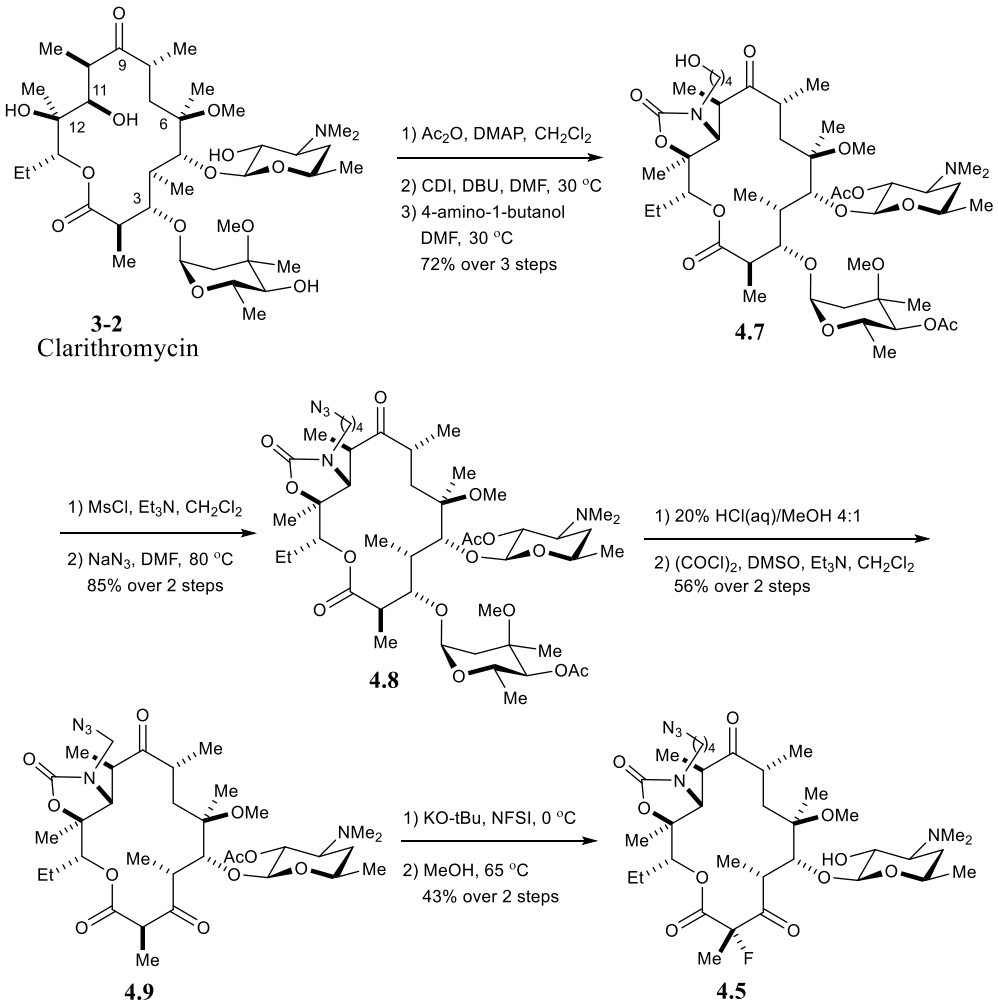
4.2 Ribosome-Templated *In Situ* Click

In 2016, the Andrade laboratory reported the first ribosome-templated *in situ* Click reaction with *E. coli* 70S ribosomes or 50S subunits.[4] To demonstrate the concept, solithromycin (**4.2**) was synthesized by incubating either *E. coli* 70S ribosomes or 50S subunits with macrolide-functionalized azide **4.5** and 3-ethynylaniline (**4.6**) in tris(hydroxymethyl)-aminomethane (Tris) buffer at room temperature for 24-48 hours (Scheme **4.1**). The azide **4.5** was prepared from the clarithromycin (Scheme **4.2**). First, five steps were employed for the protection of alcohols on the sugars and installing the butylazide side chain via Baker cyclization. Acidic removal of the 3-*O*-cladinose sugar followed by oxidation provided C3 ketone moiety. Fluorination on C2 with *t*-BuOK and NFSI followed by methanolysis of the C2' acetate furnished macrolide azide **4.5**.

Scheme 4.1 Ribosome-templated azide-alkyne cycloaddition (*in situ* Click chemistry)



Scheme 4.2 Synthesis of azide **4.5**



The result showed >10-fold more *syn* and *anti*-**4.2** than incubation with buffer solution alone. Determination of the retention time and quantity of *anti*-isomer **4.2** and *syn*-isomer **4.10** by using extracted ion chromatograms (EIC) method was used. The retention time was determined by correlation with an authentic sample. An authentic sample of *anti*-isomer **4.2** was made using a copper-catalyzed click reaction whereas *syn*-**4.10** was prepared by the thermal Huisgen cycloaddition. The ratio of *anti*-isomer **4.2** and *syn*-isomer **4.10** without ribosome was approximately 1:1; However, the ratio of *anti*-isomer **4.2** and *syn*- isomer **4.10** ribosome templated *in situ* Click reaction was 1:2, which showed a selectivity.

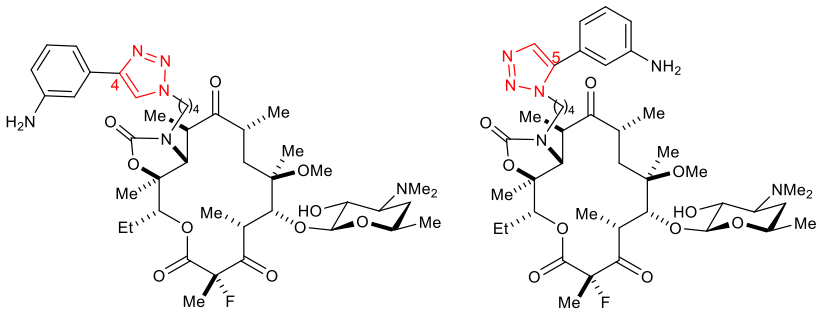


Figure 4.4 Possible regioisomeric *anti*- and *syn*-triazoles

This novel method summarized in Figure 4.5 wherein the tighter binder (macrolide azide, **A**) docks first to the ribosome followed by a second fragment (alkyne, **B**). The resultant proximity of the reactive groups markedly lowered the activation energy barrier leading to irreversible triazole formation (red line). Thermodynamically, the ribosome (target) pays the entropic penalty of bringing the two reactants together. Accordingly, the

result of this experiment was a mixture of triazole products wherein the major products formed were also the best inhibitors.

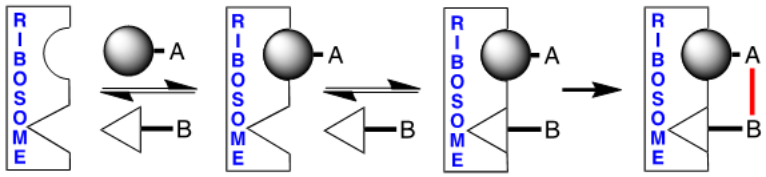


Figure 4.5 Ribosome-templated *in situ* Click approach to novel antibiotics

The successful completion of the ribosome-templated *in situ* Click method in the binary mode (i.e., one alkyne/one azide) was extended to a six-component mode (i.e., five alkynes/one azide) and ultimately led to a sixteen-component mode (i.e., fifteen alkynes/one azide). As was the case in the one- and five-alkyne experiments, the fifteen-alkyne experiment was a good predictor of lead compounds that bind better, showed significant triazole formation in the assay, and possessed lower MIC values (i.e., were more potent in antibacterial assays). A strength of the *in situ* Click methodology is that it eliminates the laborious need to synthesize countless numbers of potential drug candidates, thus saving substantial resources.

The application of *in situ* Click chemistry toward the discovery of novel macrolide antibiotics first required the synthesis of suitable azide and aryl alkyne reactants. Alkyne partners were procured by commercial vendors or chemical synthesis. We targeted two logical, validated positions to tether the side chains, specifically N11 on the macrolactone and N3’ of desosamine.

4.3 Present study

4.3.1 Synthesis of (*E*)-Dehydro Solithromycin Analogs

4.3.1.1 Rationale

Researchers at Abbott discovered that an aryl-alkyl side-chain at the C11 position A-66173 (**4.11**) and A-66005 (**4.12**) or C4" position on cladinos A-60565 (**4.13**) improved the activity against inducible *Streptococcus pyogenes* or constitutive *Streptococcus pyogenes* compared to clarithromycin (Figure 4.6).[5]

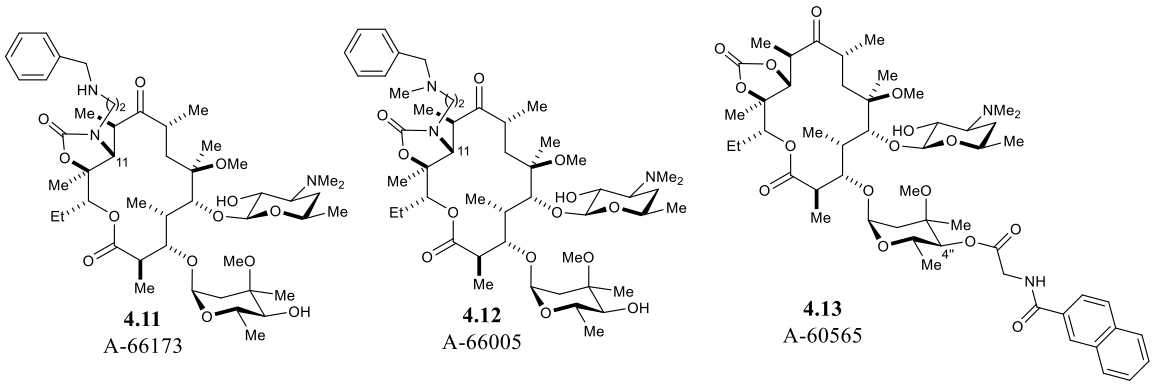


Figure 4.6 The structure of A-66173, A-66005, and A-60565

Later, scientists at Hoechst-Marion-Roussel (Aventis) and at Abbott also reported macrolide analogs with aryl-alkyl side chains at the C-11 position.[6,7] Most of the work focused on the structure and length of side chains, aryl or heteroaryl groups and C11,12 cyclic carbonates or carbamates (oxazolidinones). The greatest potency observed in this lattermost class was realized with aryl groups attached to a butyl carbamate.[8] Extensive optimization resulted in compound HMR-3647, which possessed a pyridyl-imidazole side chain and showed 10-fold improved potency over other analogs. HMR-3647 was later

renamed telithromycin. Based on these successes, three N11-tethered ketolides went on to clinical trials.[1,8,9]

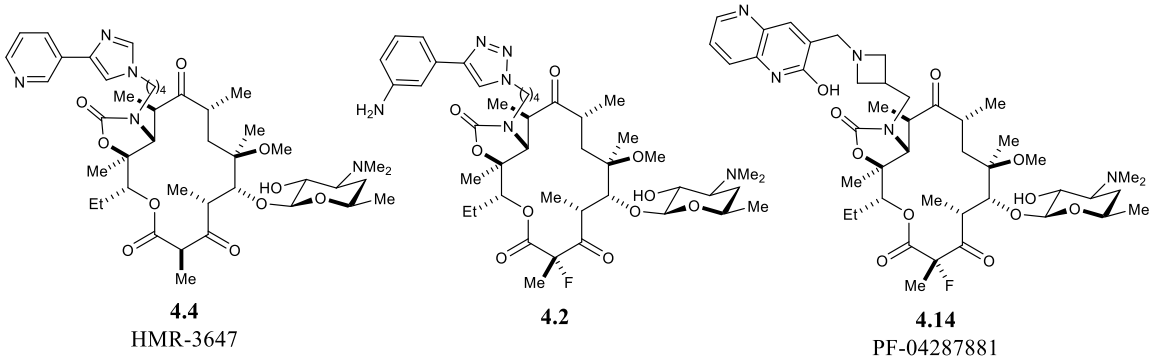


Figure 4.7 Clinical candidates of N11-tethered ketolides

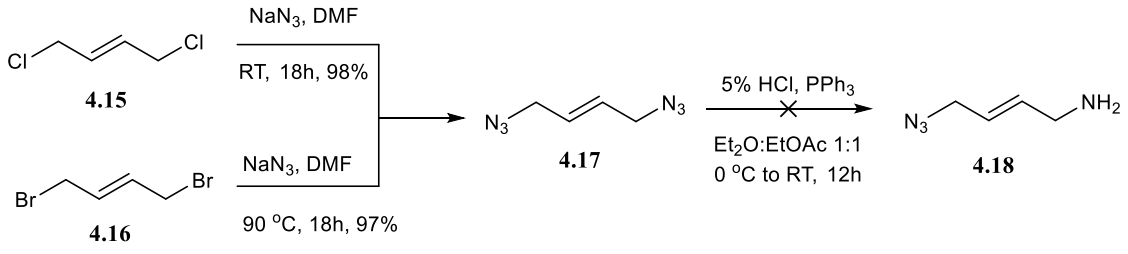
The N-11 site had been the most utilized; moreover, extensive structure-activity relationships (SAR) revealed a four-carbon tether was ideal, which is also consistent with X-ray studies (*vide supra*). While keeping the four-carbon chain length, we reasoned that installing an *E*-alkene (i.e., dehydro side chain) between C2 and C3 of the butyl side chain would change the overall energy of binding. Ernesto Freire had looked at the thermodynamic signatures (i.e., binding enthalpy and entropy) of various classes of drugs (e.g., statins and anti-HIV drugs) in order to explain and understand overall binding affinity.[10] Since the binding affinity is a combination of the binding enthalpy and entropy, favorable contributions of both to binding prove to have a high binding affinity. The favorable enthalpy associated with the formation of hydrogen bonds and van der Waals contacted via perfect geometric fit such as the distance and angle. However, the conformational entropy change was almost always unfavorable, as the binding process

involves the loss of conformational degrees of freedom. The use of an unsaturated side chain would conformationally preorganize the bi-aryl side chain in order to avoid the entropic penalty and thus favorably contribute to the overall binding.

4.3.1.2 Synthesis of (*E*)-Dehydro Side Chain

To test the hypothesis that an unsaturated side chain would improve binding, the synthesis of the (*E*)-dehydro side chain was required. With the *in situ* Click method, it was envisioned that azide **4.5** could be applied to this project with (*E*)-dehydro side chain. Accordingly, (*E*)-1,4-dichloro-2-butene or (*E*)-1,4-dibromo-2-butene was employed as starting material, which reacted with sodium azide at room temperature or 90 °C in DMF to give the diazide **4.15** in quantitative yield. Due to the instability and hazardous nature of this intermediate, it was used directly without further purification in the next step. Reduction of a single azido group with the Staudinger reaction despite modifying the stoichiometry was unsuccessful in accessing desired product **4.18**.[11]

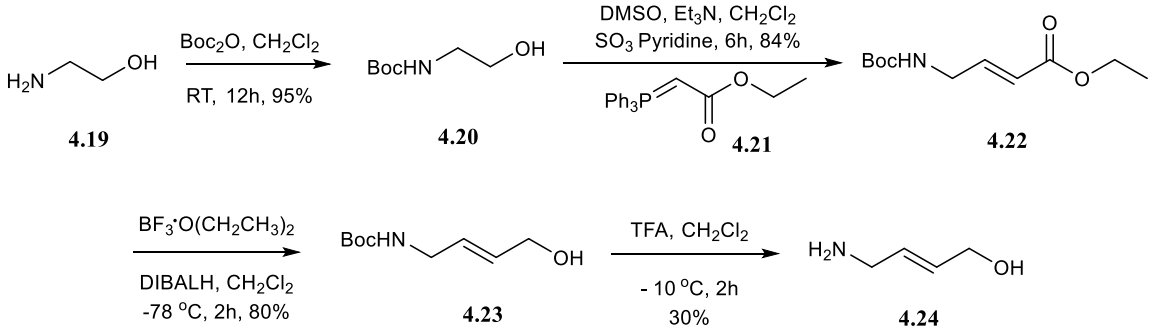
Scheme 4.3 First route of (*E*)-4-azidobut-2-en-1-amine **4.18**



At this stage, recourse to a more conservative and safer synthetic plan was made. The nitrogen in 2-aminoethanol was protected with a *tert*-butyl carbonate (Boc) group. A

one-pot Parikh-Doering oxidation/Wittig olefination procedure was used to generate **4.22** in 84% overall yield (E/Z ratio = 97:3), which contains all of the carbon atoms required.[12] A combination of DIBAL-H with $BF_3 \cdot OEt_2$ was demonstrated to be a useful reagent for the selective 1,2-reduction of α,β-unsaturated esters.[13] Thus, the selective 1,2-reduction of **4.22** followed by deprotection of Boc group gave the unsaturated amino butanol TFA salt. Basifying this salt to form the free amine was a challenge. The compound was difficult to isolate. However, passing the TFA salt solution through basic alumina and column chromatography gave the desired product **4.24** in low yields.

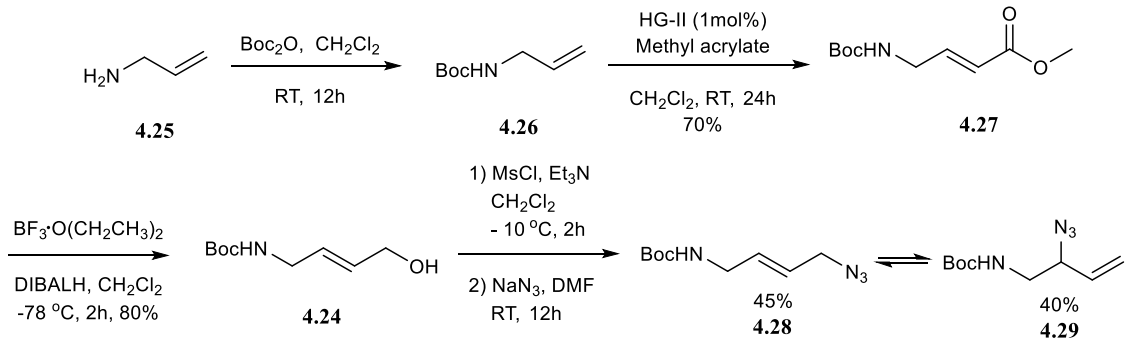
Scheme 4.4 Second route of (*E*)-4-azidobut-2-en-1-amine



To avoid tedious separation of triphenylphosphine oxide from the Wittig reaction, the route was slightly modified. It was envisioned that the cross-metathesis reaction between Boc-protected allyl amine **4.26** could access to methyl ester **4.27**. The reaction of methyl acrylate and **4.26** in the presence of Hoveyda-Grubbs second-generation catalyst gave **4.27** in 70% yield. The enoate was reduced to the allylic alcohol using previously discussed conditions. Conversion of the alcohol into the mesylate was followed by

treatment with NaN_3, which generated two regioisomers of Boc protected azido butene: **4.28** and **4.29**.

Scheme 4.5 Modified route of (*E*)-4-azidobut-2-en-1-amine

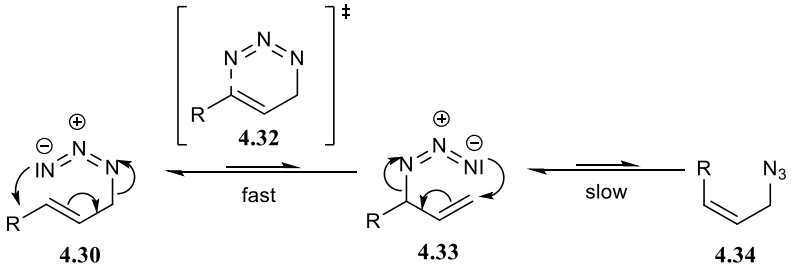


A straightforward mechanism to rationalize the routes to these two regioisomers includes an S_N2 reaction to produce **4.28** and an S_N2' process to generat **4.29**. Unfortunatley, these isomers were inseparable with countless column chromatography. A survey of the literature was enlightening. In 1960, the allylic azide rearrangement was reported, and was found that allylic azides equilibrate very rapidly.[14] The azide and the olefin groups are engaged in the dynamic [3,3]-sigmatropic equilibration process, which complicates but not necessarily precludes the isolation of **4.28** (Scheme 4.6).

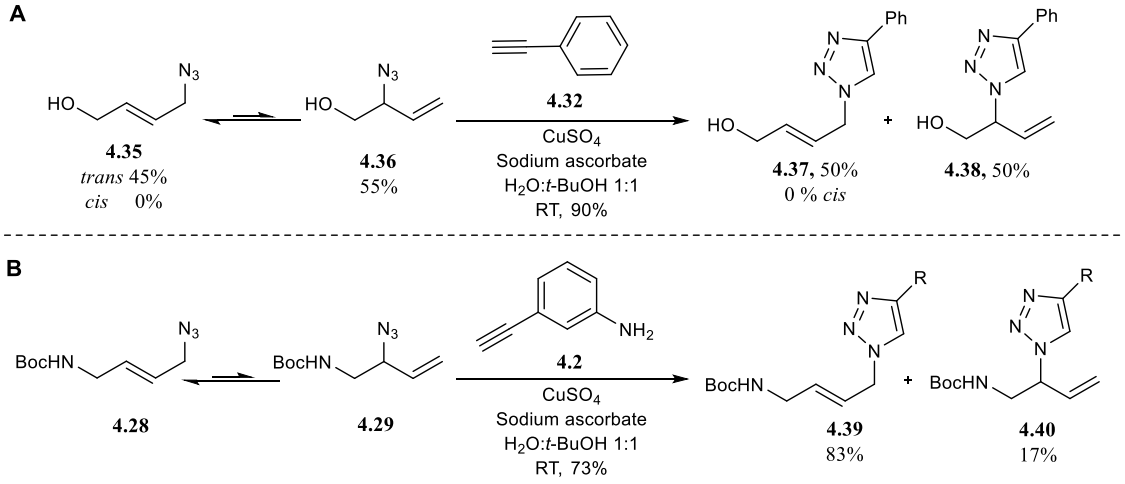
In 2005, Sharpless examined both steric and electronic influence on the reactivity of azides and olefins.[15] Primary vs secondary azide allylic systems were studied. The copper-catalyzed Click reaction did not distinguish well between the primary and secondary azide regioisomers, which showed the very similar composition of the resulting mixture of triazole products from the starting materials (Scheme 4.7, A). After several purification processes, pure **4.28** could be isolated. Subsequently, the Click reaction was

applied to the mixture of two isomers to resolve the regioisomers. The major component was the primary isomer (Scheme 4.7, B).

Scheme 4.6 Rearrangement of allylic azides



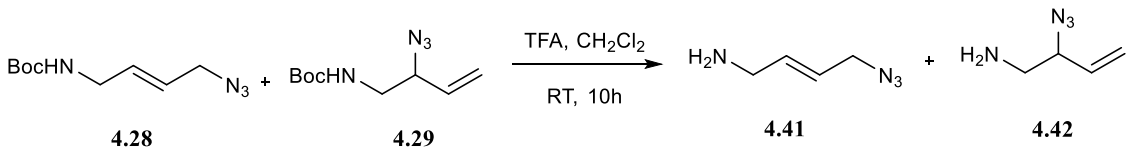
Scheme 4.7 Cu(I)-catalyzed cycloaddition of allylic azides



Trifluoroacetic acid was added to a mixture of two isomers to remove Boc group. After numerous trials to make free amine from the TFA salt, basifying the salt with ammonium hydroxide was the best condition. The TFA salt was dissolved in dichloromethane then NH_4OH was added at 0 °C to basify by monitoring pH. Once it was the optimal pH 9, the organic layer was isolated without aqueous workup and subjected to

the purification step. After quick column chromatography to get rid of the minor isomer, **4.41** was carried to the next step.

Scheme 4.8 Synthesis of (*E*)-4-azidobut-2-en-1-amine

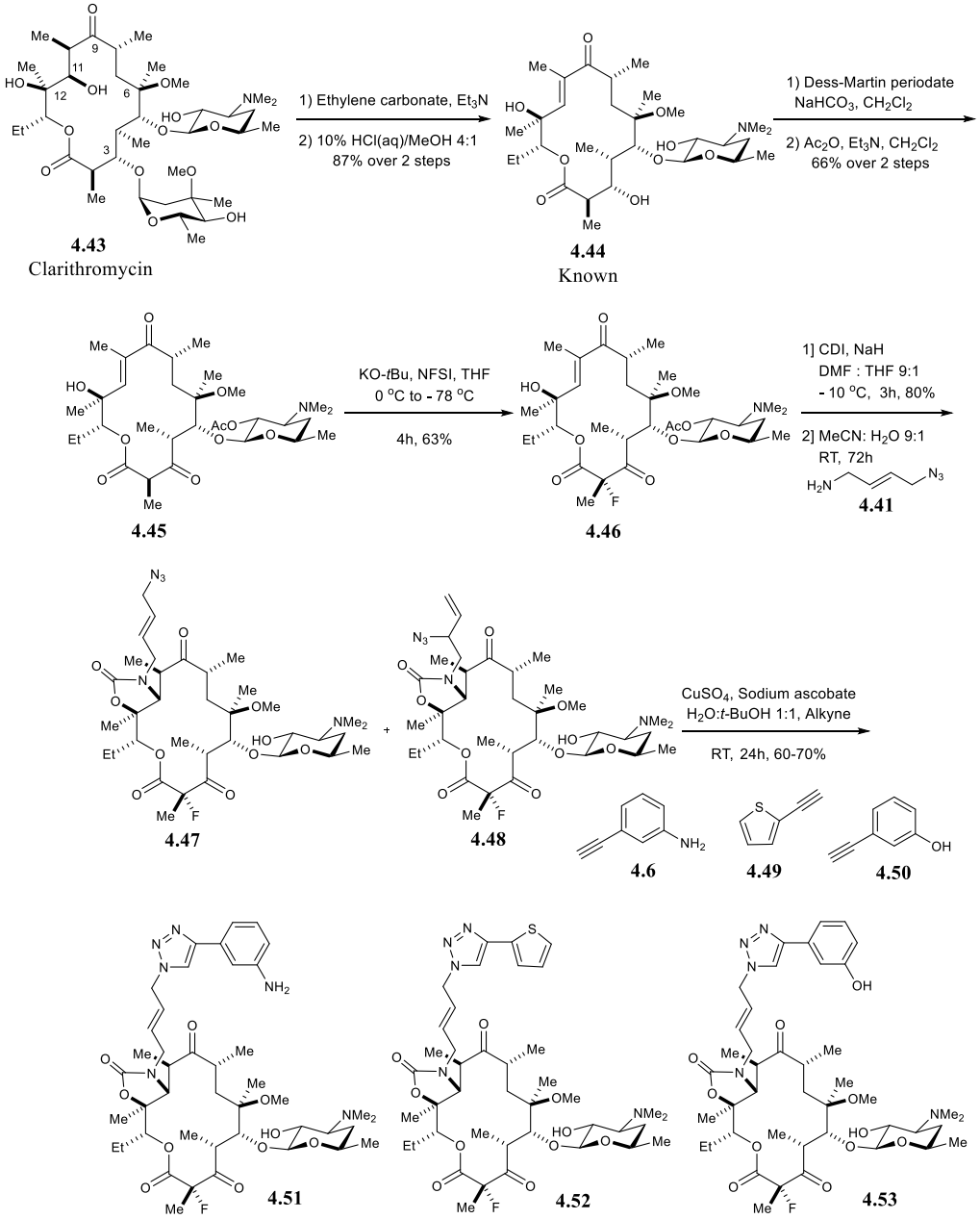


4.28 **4.29** **4.41** **4.42**

4.3.1.3 Synthesis of (*E*)-Dehydro Solithromycin Analogs

Cognizant of the allylic azide rearrangement, a new strategy for the synthesis of the solithromycin azide was put forward. Since azido side chain **4.41** could not be installed at an earlier stage (Scheme 4.2), it was reasoned that late-stage would rectify the problem. Thus, treatment of clarithromycin (**4.43**) with ethylene carbonate in refluxing triethylamine followed by dilute aqueous acid afforded enone **4.44** in good yield.[16] Increasing the amount of acid used from 4% to 10% aqueous acid reduced the reaction time from overnight to 2 h. Oxidation of the C-3 alcohol with the Dess-Martin periodinane, followed by acetylation of the 2′-hydroxy group gave **4.45**. Introducing the C-2 fluorine required optimization. At 0 °C, significant deprotection of the acetate group was observed with 1 equiv of KO-*t*Bu whereas lowering the temperature to -78 °C was not suitable to generate the C-2 anion. However, conducting anion generation at 0 °C followed by the addition of NFSI at -78 °C furnished the desired product along with 10% of acetate deprotection product. With the fluorine installed on the C-2 position, the Baker cyclization was used to install the oxazolidinone. The C-12 tertiary alcohol was activated with CDI, followed by reaction

Scheme 4.9 Synthesis of (*E*)-dehydro solithromycin **4.51-4.53**



with (*E*)-4-azidobut-2-en-1-amine to effect cyclization and acetate removal on the 2´-hydroxy group to furnish **4.47**. During the cyclization, the allylic azide rearrangement

generated minor product **4.48**. The standard Cu(I)-catalyzed Click reaction of **4.47** with three aromatic alkynes generated (*E*)-dehydro solithromycin analogs **4.51**-**4.53**.

4.3.1.4 Biological Evaluation of Analogs

With unsaturated analogs in hand, minimum inhibitory concentration (MIC) assays were determined against five strains of *Escherichia* coli (Table 4.1) and four strains of *Staphylococcus aureus* (Table 4.2). Most of these analogs were comparable with solithromycin (**4.2**); however, **4.51** showed better activity than **4.2** on *Staphylococcus aureus* strain. This meant that the unsaturated side chain favorably contributed to the overall binding.

Table 4.1 MIC assays with *Escherichia coli*

MIC(μg/ml)	*Escherichia coli*				
Compound #	DK	DK 3535	DK 2058G	SQ 171	SQ 171 2058G
SOL **4.2**	2	4	4	>32	>32
4.51	2	2	2	>32	>32
4.52	2	2	2	>32	>32
4.53	2	2	2	>32	>32

Table 4.2 MIC assays with *Staphylococcus aureus*

MIC(μg/ml)	*Staphylococcus aureus*			
Compound #	SA ATCC WT	UNC 14 A2058U	UCN 17 A2058G	UCN 18 A2059G

SOL **4.2**	<0.0625	8	4	4
4.51	<0.0625	2	0.125	<0.0625
4.52	<0.0625	16	8	8
4.53	<0.0625	8	8	4

4.3.2 Synthesis of N11-Functionalized Solithromycin Analogs that Engage in Hydrogen Bonding and π-Stacking

4.3.2.1 Rationale

The presence of an aryl group such as aniline on the side chain was extremely important for ketolides to overcome resistance. Changing the biaryl side chain tuned the activity of telithromycin and ultimately led to solithromycin. Specifically, the triazole-aniline group of solithromycin was inspired by the imidazole-pyridine moiety of telithromycin. Both side chains engage in π-stacking interactions with the uracil−adenine pair (A752 and U2609) in the binding site of the *E. coli* ribosome (Figure 4.8). More precisely, the aniline fragment stacks with A752 whereas the triazole stacked with U2609. However, the amino group in the solithromycin aniline also engages in hydrogen bonding. As a donor, it hydrogen bonds with O-4' of A752 and O-6 of G748; as an acceptor, it hydrogen bonds with N-1 of G748.[2] These additional hydrogen bonding contributions to binding help rationalize why solithromycin is the best-in-class ketolide antibiotics.

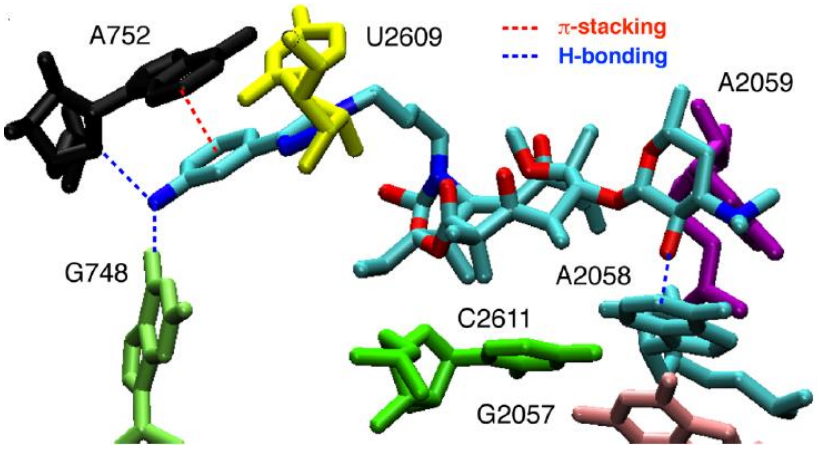


Figure 4.8 Structure of solithromycin in the binding site of *E. coli* 70S ribosome

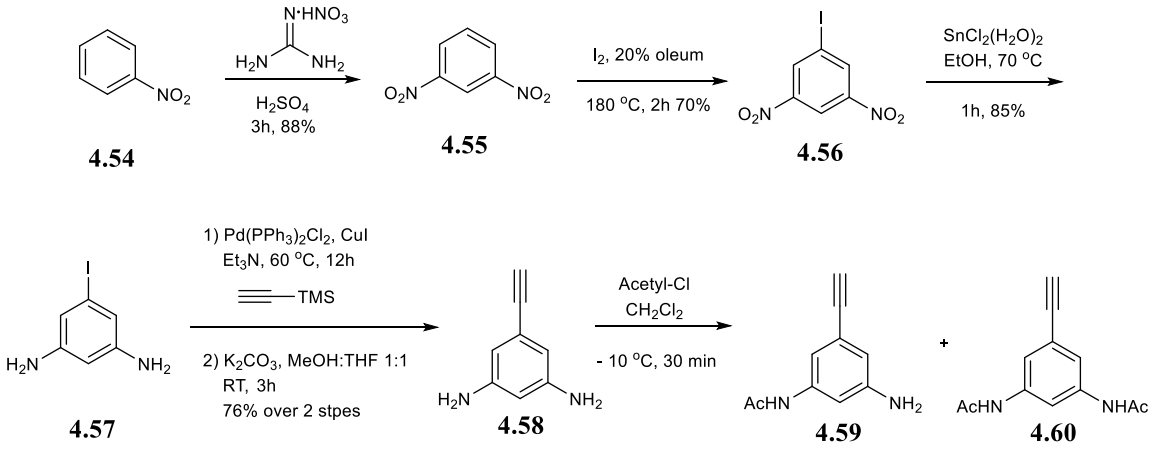
It was not apparent what aryl group could replace this aniline to enhance the stacking and hydrogen bonding interactions with 23S rRNA residues A752 and G748. Paying attention to the single bond connection between aniline and triazole inspired the introduction of a new functional group on the C5 position. Even though the single bond rotated the other functional group would compensate the loss of binding. Other analogs of solithromycin were explored via Click chemistry derived from non-commercial alkynes to study their structure-activity relationships.

4.3.2.2 Synthesis of Alkynes and Solithromycin Analogs

As a starting point, the nitro compound was believed to be suitable intermediate for preparing amine via reduction. Exploring various reports on nitration reactions led to the implementation of guanidinium nitrate in sulfuric acid for nitration of aromatic compounds.[17] With guanidinium nitrate, the nitration went smoothly to generate 1,3-dinitro-benzene (**4.55**) in the present of the strongly deactivating group in the nitration

substrate. The next challenge was the installation of a good leaving group such as bromo or iodo on the aromatic molecules containing deactivating substituents. Bromination with N-bromosuccinimide (NBS) in the presence of sulfuric acid successfully generated 1-bromo-3,5-dinitrobenzene; however, it was not suitable for the Sonogashira coupling to install the requisite alkyne functionality for subsequent Click chemistry. This resulted in a change to a better leaving group such as iodo. Direct iodination was conducted with iodine and sulfuric acid under reflux. 1-Iodo-3,5-dinitrobenzene (**4.56**) was prepared by reacting I_2 in the presence of 20% oleum.[18] The nitro groups were then simultaneously reduced to amino groups by tin(II) chloride to generate 1-Iodo-3,5-diaminobenzene (**4.57)**.[19] Lastly, a terminal alkyne was installed via Sonogashira coupling followed by a deprotection of the trimethylsilyl group yielded alkyne **4.58**.[20]

Scheme 4.10 Synthesis of bis-amino alkynes

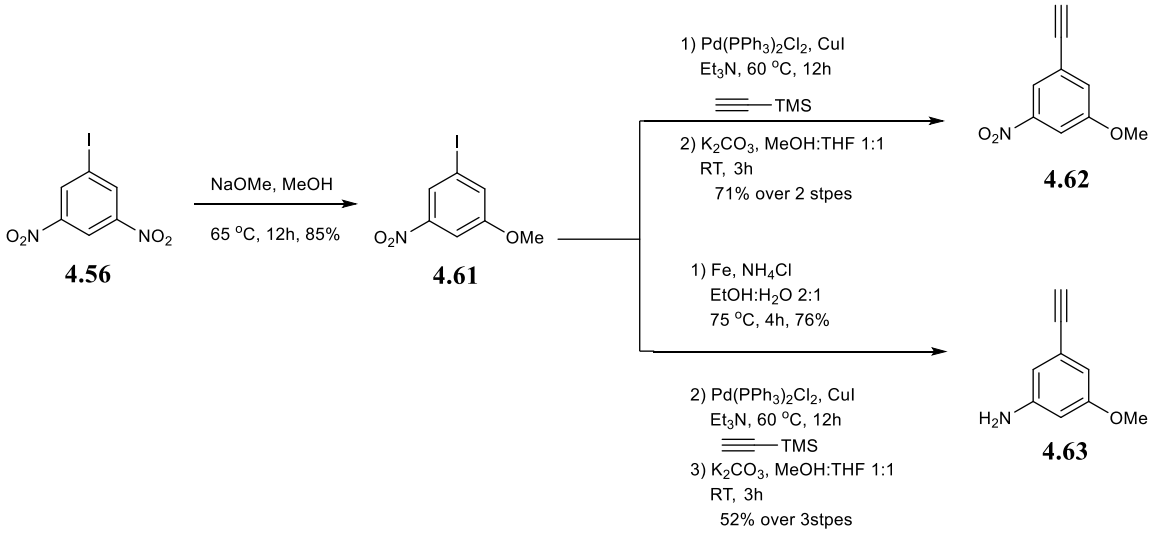


On the other hand, due to adverse effects of hydrogen bonding such as solvation, which could reduce the efficacy of potential analogs, further synthetic modifications were

implemented on alkyne **4.58** (Scheme 4.10). Protecting either one or both amino groups via acetylation generated mono- and bis-acylated amine, respectively. Both acetylated compounds were achieved in one pot, and bis-acylated alkyne **4.60** was the major product. Accessing alkynes **4.58**–**4.60** was essential for studying the contribution of hydrogen bonding provided by the amino group to binding affinity.

Expanding the scope of hydrogen bonding interactions required new functionalities other than the amino group that could have varied effects on the net forces such as methoxy or hydroxy groups. An extensive literature search revealed that the hydroxyl group could be accessed via a methoxy precursor by acidic hydrolysis. As in the previous synthetic approach, the nitro functionality served as a masked amino group. Moreoever, nitro groups have played a critical role in various drugs combating bacterial resistance (e.g., the 5-nitroimidazole class of antibacterials), which further encouraged us to implement those nitro compounds derived in our synthetic pathway.[21]

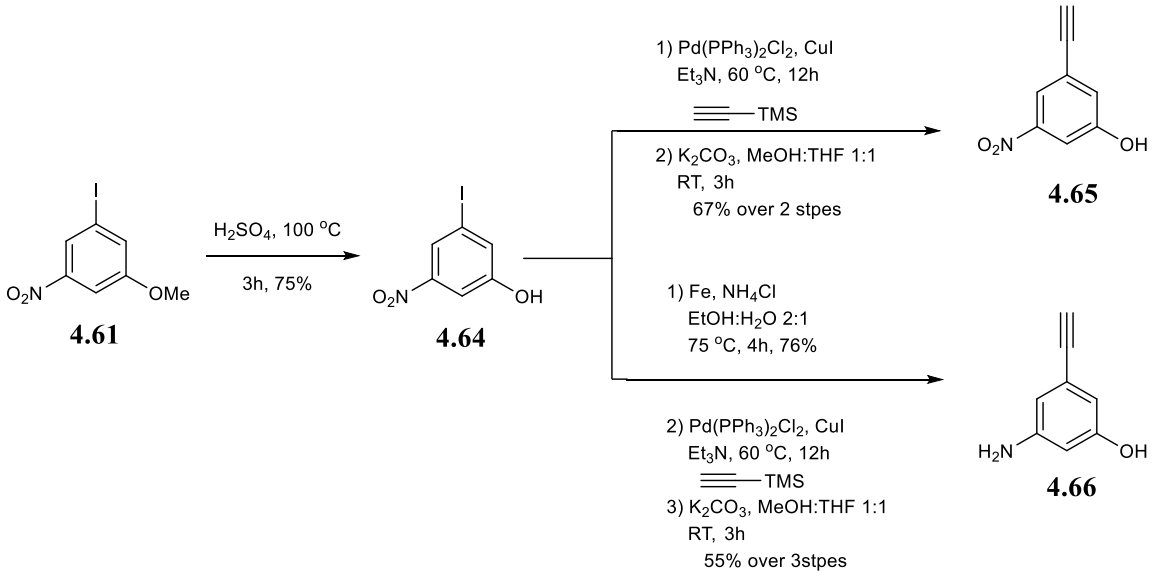
Scheme 4.11 Synthesis of methoxy alkynes



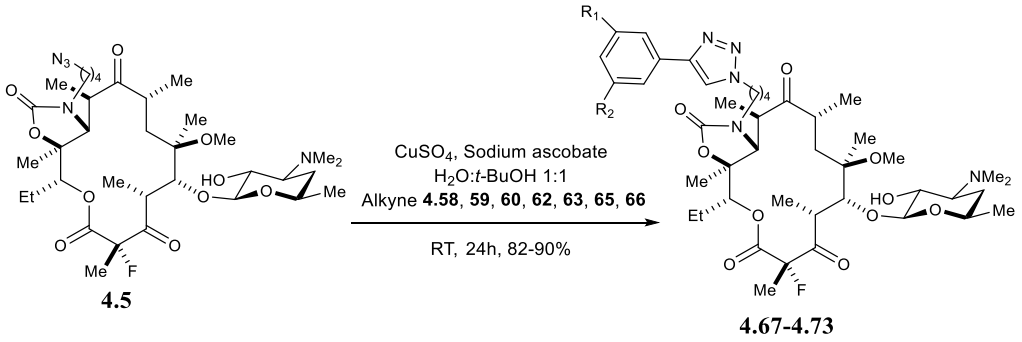
Treatment of **4.56** with sodium methoxide in methanol under reflux yielded 1-iodo-3-methoxy-5-nitrobenzene (**4.61**), which was subjected to Sonogashira coupling conditions then removal of the trimethylsilyl group to yield **4.62** in good yields (Scheme 4.11).[22] 5-Methoxy-3-ethynyl aniline (**4.63**) was prepared by Fe-mediated reduction prior to the Sonogashira coupling.

Iodide **4.61** was dissolved in concentrated sulfuric acid to generate 3-iodo-5-nitrophenol (**4.64**), which was subjected to the same synthetic route as methoxy alkynes to prepare the phenolic alkynes **4.65** and **4.66**.[23] Click chemistry was then employed to the synthesized alkynes **4.58, 59, 60, 62, 63, 65**, and **66** to prepare a library of solithromycin analogs, **4.67**–**4.73** (Scheme 4.13).

Scheme 4.12 Synthesis of phenolic alkynes



Scheme 4.13 Cu(I)-catalyzed cycloaddition with synthesized alkynes



4.5 **4.67-4.73**

4.3.2.3 Biological Evaluation of Analogs

Minimum inhibitory concentrations (MIC) assays were determined against five strains of *Escherichia* coli (Table 4.3) and four strains of *Staphylococcus aureus* (Table 4.4). Analog **4.70** which contained the nitro and methoxy moieties showed better activity than solithromycin against DK 3535, DK2058G and SQ 171 strains. The nitro and hydroxy functional groups showed better activity against the DK strain which enhanced H-bonding and π-stacking. Most of these analogs were inferior to solithromycin except wild-type *Staphylococcus aureus* strain.

Table 4.3 MIC analysis of *Escherichia coli*

	MIC(μg/ml)	*Escherichia coli*				
Alkyne #	Compound #	DK	DK 3535	DK 2058G	SQ 171	SQ 171 2058G
4.6	SOL **4.2**	2	4	4	>32	>32
4.58	**4.67**	4	4	8	32	>32
4.59	**4.68**	4	4	4	>32	>32
4.60	**4.69**	8	16	8	>32	>32

4.62	**4.70**	4	2	2	8	N/A
4.63	**4.71**	4	4	4	32	>32
4.65	**4.72**	0.5	8	4	32	>32
4.66	**4.73**	4	4	4	32	>32

Table 4.4 MIC analysis of *Staphylococcus aureus*

	MIC(µg/ml)	*Staphylococcus aureus*			
Alkyne #	Compound #	SA ATCC WT	UNC 14 A2058U	UCN 17 A2058G	UCN 18 A2059G
4.6	SOL **4.2**	<0.0625	8	4	4
4.58	**4.67**	0.03125	32	8	16
4.59	**4.68**	0.03125	>128	128	128
4.60	**4.69**	0.0625	>128	>128	128
4.62	**4.70**	0.25	64	32	32
4.63	**4.71**	0.008	64	16	16
4.65	**4.72**	<0.0625	16	4	4
4.66	**4.73**	0.016	32	32	32

4.3.3 Synthesis of Solithromycin Analogs with Biaryl Side Chain

4.3.3.1 Rationale

As shown previously, desosamine is a key component for macrolide antibiotic activity. In 2010, Rib-X Pharmaceuticals introduced a modification on the C3' position of desosamine via the Click reaction (Figure 4.9).[24]

As a key structural element, Rib-X analog **4.74** with the desosamine modification contains a triazole fragment connected via a two-carbon linker to the desosamine nitrogen of clarithromycin. Inspired by Rib-X, two-carbon and three-carbon linkers bearing azides connected via the desosamine and generate leads **4.75** and **4.76**. An overlay of these two analogs with the Rib-X candidate revealed overlapping of chemical space. Thus, the synthesis of these two analogs was undertaken.

4.3.3.2 Synthesis of Bis-Click Analogs

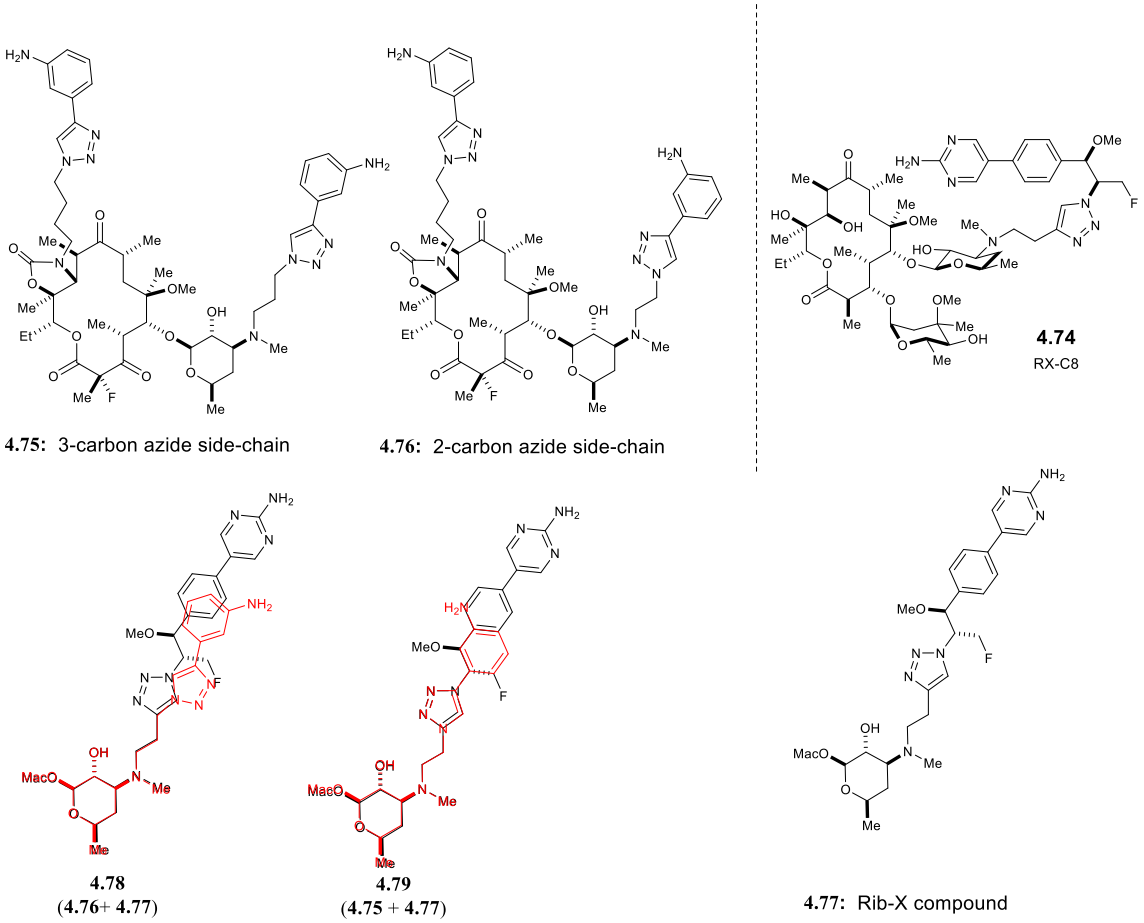
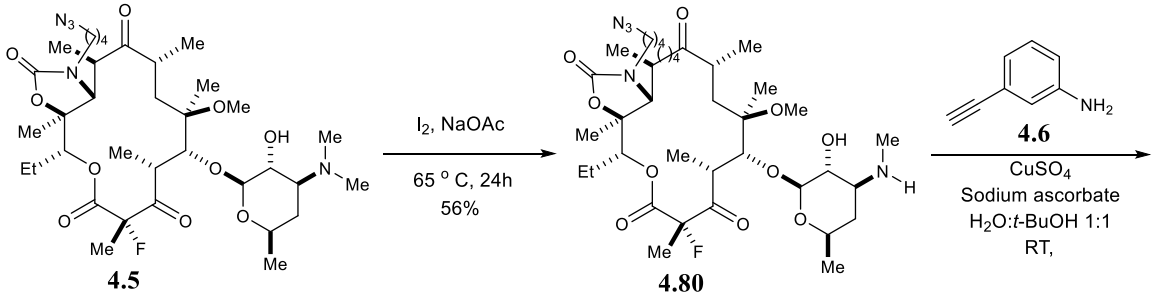
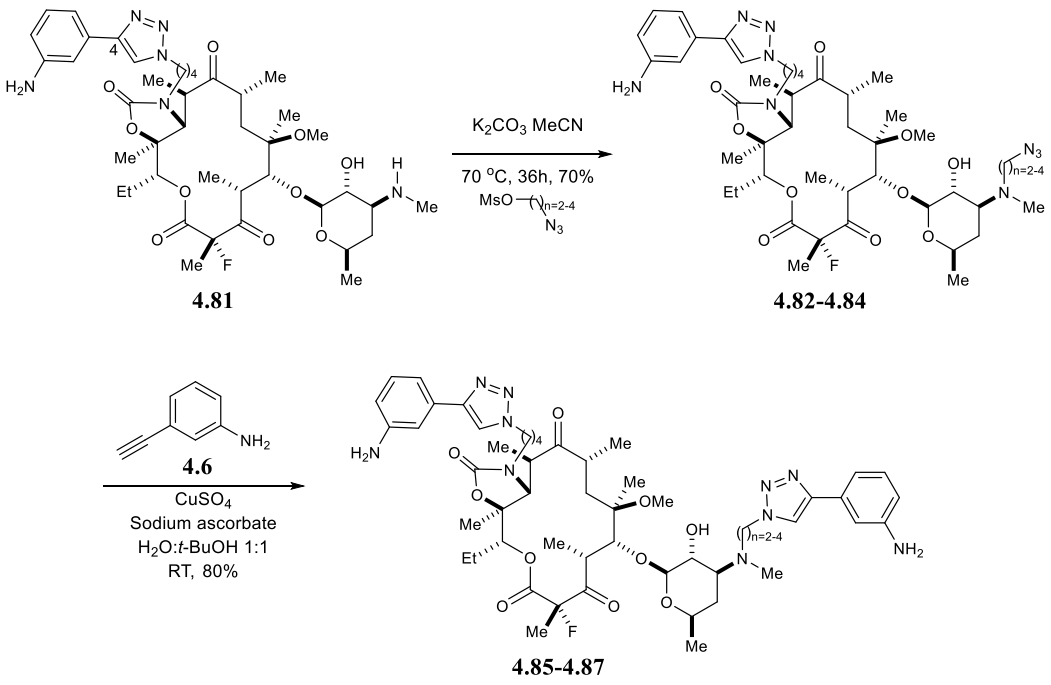


Figure 4.9. Desosamine targeting approach from Rib-X

The Andrade lab has reported a nine-step, gram-scale synthesis of macrolide tethered azide **4.5** at N11 (Scheme 2) from commercial clarithromycin. To access the requisite bis-azide, selective demethylation on *N*-methyl desosmaine was considered. The best reaction conditions for this transformation was iodine with a strong base such as sodium methoxide under basic pH. Retaining basic conditions (pH 8) was the key to success for this reaction. At low pH the reaction did not proceed whereas decomposition of the product was observed at pH above 9. Since the aniline side chain was optimal at N-11, it was not modified and introduced via the Click reaction. Next, the side chain on desosamine was installed by alkylation of the C-3' position with ω-azido mesylates. Two-, three-, and four-carbon linkers were introduced bearing a terminal azide functionality. With desosamine azides in hand the Click reaction was used to generate novel desosamine-modified solithromycin analogs. The reactions proceeded smoothly with moderate yields. The stage was now set to identify the optimal linker length between the desosamine residue and secondary sites in the ribosome that could establish additional contacts with the ketolide.

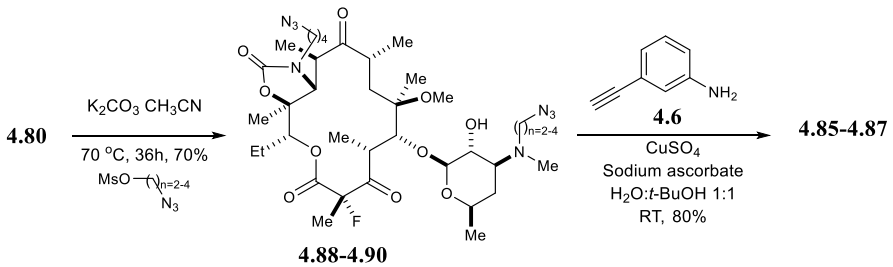
Scheme 4.14. Desosamine targeting approach





Application of desosamine linker chain strategy with the ribosome-templated azide-alkyne cycloaddition approach would be the use of bis-azide moieties **4.88-4.90** on macrolide ring. To date, the use of a two-side chain strategy has not been reported. To access the requisite bis-azide, the same reaction route without having side chain on N-11 was carried out. With the bis-azide compound **4.88-4.90** in hand, a test Click reaction was conducted to generate the same product **4.85-4.87** as Scheme 14.

Scheme 4.15. Test reaction of the bis-Click approach



4.3.3.3 Biological Evaluation of Bis-Click Analogs

Minimum inhibitory concentrations (MIC) assays were determined against five strains of *Escherichia coli* (Table 4.5) and four strains of *Staphylococcus aureus* (Table 4.6). All of the analogs tested were inferior to solithromycin (**4.2**). The MIC data failed to provide a clear picture of the optimal chain length.

Table 4.5 MIC assay with *Escherichia coli*

MIC(μg/ml)	*Escherichia coli*				
Compound #	DK	DK 3535	DK 2058G	SQ 171	SQ 171 2058G
SOL **4.2**	2	4	4	>32	>32
4.85 (C2)	>32	>32	>32	>32	>32
4.86 (C3)	4	4	16	>32	>32
4.87(C4)	8	8	>32	>32	>32

Table 4.6 MIC assay with *Staphylococcus aureus*

MIC(μg/ml)	*Staphylococcus aureus*			
Compound #	SA ATCC WT	UNC 14 A2058U	UCN 17 A2058G	UCN 18 A2059G
SOL **4.2**	<0.0625	8	4	4
4.85 (C2)	0.5	>128	>128	>128
4.86 (C3)	1	>128	>128	>128
4.87(C4)	1	>128	>128	>128

4.4. Conclusion

Described herein is the successfully development of lead compounds for *in situ* Click chemistry, in addition to have various clicked analogs of solithromycin. *In situ* Click chemistry will inform optimal fragments and linker lengths. We will optimize conditions for the bis-azide variant first using a five-alkyne mixture with saturated/unsaturated side chain solithromycin azide. The number of products in the bis-azide *in situ* Click ($4n^2 + 4n$) where n=# of alkynes is markedly higher than the mono-azide. The reported step-efficient routes will enable the rapid synthesis and hit validation based on LC-MS analysis.

4.5 References

1. Fernandes, P.; Martens, E.; Bertrand, D.; Pereira, D. *Bioorg. Med. Chem.* **2016**, *24*, 6420–6428.
2. Llano-Sotelo, B.; Dunkle, J.; Klepacki, D.; Zhang, W.; Fernandes, P.; Cate, J. H. D.; Mankin, A. S. *Antimicrob. Agents Chemother.* **2010**, *54*, 4961–4970.
3. Evrard-Todeschi, N.; Gharbi-Benarous, J.; Gaillet, C.; Verdier, L.; Bertho, G.; Lang, C.; Parent, A.; Girault, J. P. *Bioorg. Med. Chem.* **2000**, *8*, 1579–1597.
4. Glassford, I.; Teijaro, C. N.; Daher, S. S.; Weil, A.; Small, M. C.; Redhu, S. K.; Colussi, D. J.; Jacobson, M. A.; Childers, W. E.; Buttaro, B.; Nicholson, A. W.; MacKerell, A. D.; Cooperman, B. S.; Andrade, R. B. *J. Am. Chem. Soc.* **2016**, *138*, 3136–3144.
5. Fernandes, P. B.; Baker, W. R.; Freiberg, L. A.; Hardy, D. J.; McDonald, E. J. *Antimicrob. Agents Chemother.* **1989**, *33*, 78–81.
6. Agouridas, C.; Denis, A.; Auger, J. M.; Benedetti, Y.; Bonnefoy, A.; Bretin, F.; Chantot, J. F.; Dussarat, A.; Fromentin, C.; D'Ambrières, S. G.; Lachaud, S.; Laurin, P.; Le Martret, O.; Loyau, V.; Tessot, N. *J. Med. Chem.* **1998**, *41*, 4080–4100.
7. Griesgraber, G.; Or, Y. S.; Chu, D. T. W.; Nilius, A. M.; Johnson, P. M.; Flamm, R. K.; Henry, R. F.; Plattner, J. J.; Road, A. P.; Park, A. **1995**, *49*, 465–477.
8. Denis, A.; Agouridas, C.; Auger, J.; Benedetti, Y.; Bonnefoy, A.; Bretin, F.; Chantot, J.; Dussarat, A.; Fromentin, C.; Gouin, S.; Ambri, D.; Lachaud, S.; Laurin, P.; Martret, O. Le; Loyau, V. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 3075–3080.
9. Magee, T. V.; Ripp, S. L.; Li, B.; Buzon, R. A.; Chupak, L.; Dougherty, T. J.;

Finegan, S. M.; Girard, D.; Hagen, A. E.; Falcone, M. J.; Farley, K. A.; Granskog, K.; Hardink, J. R.; Huband, M. D.; Kamicker, B. J.; Kaneko, T.; Knickerbocker, M. J.; Liras, J. L.; Marra, A.; Medina, I.; Nguyen, T. T.; Noe, M. C.; Obach, R. S.; O'Donnell, J. P.; Penzien, J. B.; Reilly, U. D.; Schafer, J. R.; Shen, Y.; Stone, G. G.; Strelevitz, T. J.; Sun, J.; Tait-Kamradt, A.; Vaz, A. D. N.; Whipple, D. A.; Widlicka, D. W.; Wishka, D. G.; Wolkowski, J. P.; Flanagan, M. E. *J. Med. Chem.* **2009**, *52*, 7446–7457.

10. Freire, E. *Drug Discov Today.* **2008**, *13*, 869–874.
11. Gololobov, Y. G.; Zhmurova, I. N.; Kasukhin, L. F. *Tetrahedron* **1981**, *37*, 437–472.
12. Tosatti, P.; Horn, J.; Campbell, A. J.; House, D.; Nelson, A.; Marsden, S. P. *Adv. Synth. Catal.* **2010**, *352*, 3153–3157.
13. Moriwake, T.; Hamano, C., S.; Miki, D.; Saito, S. *Chem. Lett.* **1986**, 815–818.
14. Gagneuz, A.; Winstein, S.; Young, W. G. *J. Am. Chem. Soc.* **1960**, *82*, 5956–5957.
15. Feldman, A. K.; Colasson, B.; Sharpless, K. B.; Fokin, V. V. *J. Am. Chem. Soc.* **2005**, *127*, 13444–13445.
16. Griesgraber, G.; Kramer, M. J.; Elliott, R. L.; Nilius, A M.; Ewing, P. J.; Raney, P. M.; Bui, M. H.; Flamm, R. K.; Chu, D. T.; Plattner, J. J.; Or, Y. S. *J. Med. Chem.* **1998**, *41*, 1660–1670.
17. Ramana, M. M. V; Malik, S. S.; Parihar, J. A. *Tetrahedron Lett.* **2004**, *45*, 8681–8683.
18. Arotsky, J.; Butler, R.; Darby, A. C. *J. Chem. Soc.* **1970**, 1480–1485.

19. Lux, J.; Chan, M.; Elst, L. Vander; Schopf, E.; Mahmoud, E.; Laurent, S.; Almutairi, A. *J. Mater. Chem. B. Mater. Biol. Med.* **2013**, *1*, 6359–6364.

20. Sonogashira, K. *J. Organomet. Chem.* **2002**, *653*, 46–49.

21. Moraski, G. C.; Thanassi, J. A.; Podos, S. D.; Pucci, M. J.; Miller, M. J. *J. Antibiot.* **2011**, *64*, 667–671.

22. Jorgensen, W. L.; Bollini, M.; Thakur, V. V.; Domaoal, R. A.; Spasov, K. A.; Anderson, K. S. *J. Am. Chem. Soc.* **2011**, *133*, 15686–15696.

23. Henry, H.; Wignall, J. S. *J. Chem. Soc.* **1926**, 2077–2079.

24. Hanselmann, R.; Job, G. E.; Johnson, G.; Lou, R.; Martynow, J. G.; Reeve, M. M. *Org. Process Res. Dev.* **2010**, *14*, 152–158.

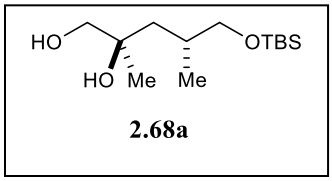
CHAPTER 5

EXPERIMENTAL SECTION

General Methods. All reactions containing moisture or air sensitive reagents were performed in oven-dried glassware under nitrogen or argon. *N*,*N*-Dimethylformamide, tetrahydrofuran, toluene and dichloromethane were passed through two columns of neutral alumina prior to use. Anhydrous dimethyl sulfoxide (DMSO) was purchased from Sigma-Aldrich and subjected to three cycles of freeze-pump-thaw before use. Pyridine, 2,6-lutidine, acetone, *i*-Pr_2NEt, *i*-Pr_2NH and Et_3N were distilled from CaH_2 prior to use. Molecular sieves (4Å) were activated by flame-drying under vacuum prior to use. AgOTf was purchased from Sigma-Aldrich and azeotropically dried with dry toluene prior to use. Compounds **2.49 (**Smith, A. B.; Mesaros, E. F.; Meyer, E. A. *J. Am. Chem. Soc.* **2006**, *128*, 5292–5299), **2.72 (**González, Á.; Aiguadé, J.; Urpí, F.; Vilarrasa, J. *Tetrahedron Lett.* **1996**, *37*, 8949–8952), **4.5 (**Glassford, I.; Teijaro, C. N.; Daher, S. S.; Weil, A.; Small, M. C.; Redhu, S. K.; Colussi, D. J.; Jacobson, M. A.; Childers, W. E.; Buttaro, B.; Nicholson, A. W.; MacKerell, A. D.; Cooperman, B. S.; Andrade, R. B. *J. Am. Chem. Soc.* **2016**, *138*, 3136–3144), **4.44 (**Griesgraber, G.; Kramer, M. J.; Elliott, R. L.; Nilius, A M.; Ewing, P. J.; Raney, P. M.; Bui, M. H.; Flamm, R. K.; Chu, D. T.; Plattner, J. J.; Or, Y. S. *J. Med. Chem.* **1998**, *41*, 1660–1670), **4.55 (**Ramana, M. M. V; Malik, S. S.; Parihar, J. A. *Tetrahedron Lett.* **2004**, *45*, 8681–8683), **4.56** (Arotsky, J.; Butler, R.; Darby, A. C. *J. Chem. Soc.* **1970**, 1480–1485), **4.57** (Lux, J.; Chan, M.; Elst, L. Vander; Schopf, E.; Mahmoud, E.; Laurent, S.; Almutairi, A. *J. Mater. Chem. B. Mater. Biol. Med.* **2013**, *1*, 6359–6364), **4.61** (Jorgensen, W. L.; Bollini, M.; Thakur, V. V.; Domaoal, R. A.; Spasov, K. A.; Anderson, K. S. *J. Am. Chem. Soc.* **2011**, *133*, 15686–15696) were prepared

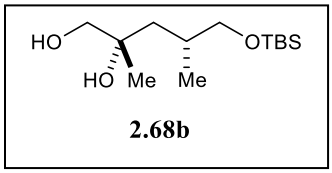
according to known literature procedures. All other reagents were purchased from commercial sources and used without further purification. All solvents for work-up procedures were used as received. Flash column chromatography was performed according to the procedure of Still (Still, W. C.; Kahn, M.; Mitra, A. *J. Org. Chem.* **1978**, *43*, 2923) using ICN Silitech 32-63 D 60Å silica gel with the indicated solvents. All HF reactions are performed in Nalgene containers. Thin layer chromatography was performed on Merck 60 F254 silica gel plates. Detection was performed using UV light, $KMnO_4$ stain, iodine chamber, PMA stain and subsequent heating. ^{1}H and ^{13}C NMR spectra were recorded at the indicated field strength in $CDCl_3$ at rt. Chemical shifts are indicated in parts per million (ppm) downfield from tetramethylsilane (TMS, $\delta = 0.00$) and referenced to the CDCl3. Splitting patterns are abbreviated as follows: s (singlet), d (doublet), bs (broad singlet), bd (broad doublet), t (triplet), q (quartet) and m (multiplet).

5.1 Chapter 2: Progress Toward the Synthesis of (*R*)-4-Fluoro-4-Desmethyl-Solithrolycin



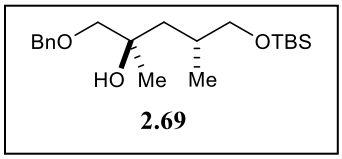
2.68a

Dihydroxylation 2.68: TBSCl (4.48 g, 29.74 mmol) and imidazole (4.05 g, 59.49 mmol) were added sequentially to a solution of alcohol **2.49** (2.72 g, 23.80 mmol) in CH_2Cl_2 (205 mL) at 0 °C. The reaction mixture was stirred for 2h while warming to room temperature. H_2O (100 mL) was added, and the reaction mixture was extracted with CH_2Cl_2 (3 x 100 mL). The organic layers were washed with brine (100 mL) and filtered over Na_2SO_4. The solvent was removed under reduced pressure and dried under high vacuum, and dissolved in a mixture of *t*-BuOH/THF/H_2O (3:2:1). *N*-Methylmorpholine *N*-oxide (13.94 g, 118.98 mmol) was added to **2.67** (5.44 g, 23.80 mmol). A catalytic amount of OsO_4 (4% wt solution in H_2O, 1.51 mL) was added dropwise to the solution. The reaction was stirred at rt for 12 h. The reaction was quenched by adding 40% Na_2SO_3 solution (100 mL). The reaction mixture was extracted using CH_2Cl_2 (3 x 100 mL). The combined organic phases were dried over Na_2SO_4 and concentrated under vacuum. The residue was purified by flash column chromatography eluting with 0–5% MeOH/CH_2Cl_2 to give 3.12 g of **2.68a** (75%, dr = 2:1) as a colorless oil. $[\alpha]^{23}_D$ +12.3 (c 1.0, $CHCl_3$); IR (neat) 3356, 2971, 2160, 1750, 1457, 1377, 1264, 1161, 1056, 1002, 952, 837, 735, 703 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.53 (dd, *J* = 9.8, 4.1 Hz, 1H), 3.36 – 3.20 (m, 3H), 1.93 (s, 1H), 1.61 (dd, *J* = 14.6, 8.2 Hz, 1H), 1.32 (dd, *J* = 14.6, 3.1 Hz, 1H), 1.10 (s, 3H), 0.89 – 0.79 (m, 12H), 0.03 (d, *J* = 1.7 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 71.8, 71.4, 70.1, 44.5, 31.4, 25.9 (2C) , 23.5, 19.5, 18.3, -5.5 (3C); HRMS (ESI) calc'd for $C_{13}H_{30}O_3Si$ + Na = 285.1862, found 285.1854.



2.68b

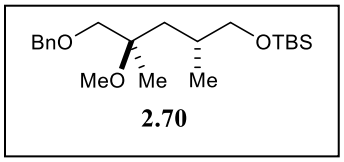
$[\alpha]^{23}_D$ +14.7 (c 1.1, $CHCl_3$); IR (neat) 3355, 2954, 2928, 2857, 2161, 1463, 1387, 1255, 1045, 1005, 939, 834, 814, 774, 737, 667 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.59 – 3.42 (m, 2H), 3.40 – 3.22 (m, 2H), 1.85 (s, 1H), 1.59 (dd, *J* = 14.6, 7.0 Hz, 1H), 1.51 (dd, *J* = 14.6, 4.1 Hz, 1H), 1.16 (s, 3H), 0.93 – 0.81 (m, 12H), 0.07 (s, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 72.2, 70.1, 69.5, 45.0 (2C), 31.7, 26.0, 25.8, 25.2, 19.4, 18.5, -5.4 (3C); HRMS (ESI) calc'd for $C_{13}H_{30}O_3Si$ + Na = 285.1862, found 285.1857.



2.69

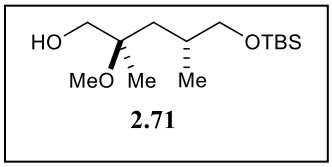
Benzyl ether 2.69: A solution of diol **2.68a** (2.44 g, 9.29 mmol) in THF (10 mL) was cannulated into a suspension of NaH (60%, 743 mg, 18.57 mmol) in THF (93 mL) at 0 °C. The resulting mixture was stirred for 20 min at 0 °C before adding BnBr (1.74g, 10.2 mmol). The solution was allowed to gradually warm to rt and after 12 hours cooled back to 0 °C and slowly quenched with H_2O until the bubbling of H_2 ceased. The mixture was then diluted with H_2O (30 mL) and extracted with EtOAc (3 x 50 mL). The combined organic layers were then washed with H_2O (2 x 30 mL), brine (30 mL) and dried over Na_2SO_4. The solvent was removed under reduced pressure and the product purified by flash column chromatography eluting with 0-10% EtOAc in hexanes to afford 2.13 g (65%) of **2.69**. $[\alpha]^{23}_D$ + 11.8 (c 1.2, $CHCl_3$); IR (neat) 3365, 2954, 2928, 2857, 1255, 1076, 1006, 939, 834, 774, 738. 667 cm-1; ^1H NMR (500 MHz, $CDCl_3$) δ 7.24 (m, 5H), 4.50 (d, *J* = 3.4 Hz, 2H), 3.85 (s, 1H), 3.47 (dd, *J* = 9.8, 4.7 Hz, 1H), 3.32 – 3.15 (m, 3H), 1.86 (s, 1H), 1.56 (dd, *J* = 14.5, 7.5 Hz, 1H), 1.43 (dd, *J* = 14.3, 4.6 Hz, 1H), 1.13 (s, 3H), 0.83 (s, 12H), -0.00 (d, *J* = 1.9 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 138.7, 128.5, 128.4, 127.7 (2C), 127.6,

79.1, 73.5, 71.6, 69.9, 44.5, 31.4, 26.0 (2C), 24.2, 23.7, 19.5, 18.4, -5.4, -5.3; HRMS (ESI) calc'd for $C_{20}H_{36}O_3Si$ + Na = 375.2331, found 375.2334.



2.70

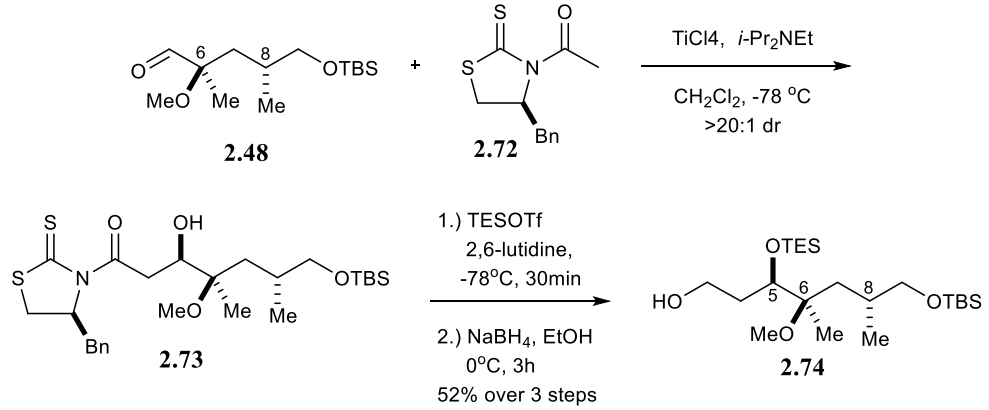
Methyl Ether 2.70: To a solution of **2.69** (6 g, 11.72 mmol) in CH_2Cl_2 (145 mL) was added 2,6-DTBMP (24.0 g, 117.21 mmol) and MeOTf (11.5 g, 70.32 mmol). After 48 h, the reaction was quenched by adding sat $NaHCO_3$ (100 mL) and stirred for 15 mins. MeOH (100 mL) was added and stirred for 30 min. The reaction mixture was then diluted with Et_2O (250 mL) and the aqueous layer was extracted with Et_2O (2 x 150 mL). The combined organic layers were washed with brine (200 mL), dried (Na_2SO_4) and filtered. The solvent was evaporated under reduced pressure, and the residue was purified by flash column chromatography eluting with hexanes to recover the 2,6-DTBMP then 0–5% EtOAc/hexanes to give 316 mg of **2.70** (75%) as a colorless oil. $[\alpha]^{23}_D$ +12.3 (c 1.2, $CHCl_3$); IR (neat) 2928, 2855, 2160, 2032, 1462, 1361, 1250, 1168, 1081, 834, 774, 735, 698, 667 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.35 – 7.27 (m, 5H), 4.59 – 4.43 (m, 2H), 3.46 (ddd, *J* = 31.3, 9.7, 5.8 Hz, 1H), 3.37 – 3.28 (m, 3H), 3.22 (s, 3H), 1.76 (td, *J* = 6.7, 4.3 Hz, 1H), 1.62 (ddd, *J* = 19.1, 14.6, 4.3 Hz, 1H), 1.29 (ddd, *J* = 35.5, 14.6, 6.9 Hz, 1H), 1.19 (s, 3H), 0.95 (dd, *J* = 6.7, 4.7 Hz, 3H), 0.89 (s, 9H), 0.03 (s, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ ^{13}C NMR (126 MHz, $CDCl_3$) δ 138.6 (2C), 128.4, 127.7 (2C), 127.6, 75.0, 74.8, 73.5, 73.4, 69.1, 49.6, 38.0, 37.8, 31.4, 26.1, 21.3, 18.9, 18.8, 18.5, -5.2 (2C); HRMS (ESI) calc'd for $C_{21}H_{38}O_3Si$ + Na = 389.2488, found 389.2501.

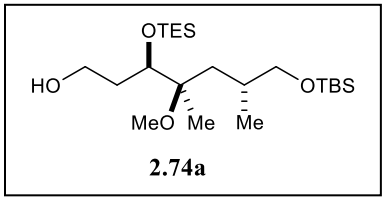


2.71

Alcohol 2.71: To a solution of ether **2.70** (1.52 g, 4.13 mmol) in EtOH (41 mL) was added 10% Pd/C (88 mg, 0.83 mmol) under an atmosphere of H_2. The reaction was monitored by TLC for 4–8 h. The reaction mixture was filtered through a Celite plug that had been previously washed with EtOAc. The solvent was concentrated under reduced pressure, and the residue was purified by flash column chromatography eluting with 0–10% EtOAc/hexanes to afford 913.42 mg (80%) of **2.71** as a colorless oil. $[\alpha]^{23}_{D}$ –1.2 (c 0.96, $CHCl_3$); IR (neat) 3373, 2954, 2928, 2857, 2160, 1472, 1387, 1255, 1076, 1002, 939, 834, 774, 738, 667 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.43 (dd, *J* = 11.4, 4.7 Hz, 1H), 3.37 (dd, *J* = 9.6, 5.8 Hz, 2H), 3.30 (dd, *J* = 9.6, 7.5 Hz, 1H), 3.15 (s, 3H), 2.46 (s, 1H), 1.78 – 1.66 (m, 1H), 1.63 (dd, *J* = 14.8, 3.9 Hz, 1H), 1.21 (dd, *J* = 14.8, 6.4 Hz, 1H), 1.11 (s, 3H), 0.87 (d, *J* = 6.8 Hz, 3H), 0.84 (s, 9H), -0.00 (s, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 77.6, 69.4, 67.1, 49.3, 37.4, 31.1, 26.2, 26.1, 26.0, 20.9, 19.1, 18.6, -5.3 (2C); HRMS (ESI) calc'd for $C_{14}H_{32}O_3Si$ + Na = 299.2018, found 299.2019.

Scheme 2.19 Determination of absolute stereochemistry at C6

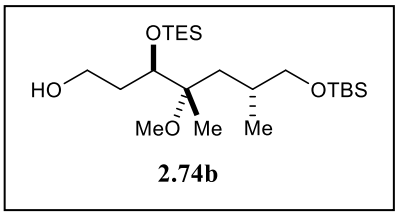




2.74a

Stereochemical correlation of 2.74a: DMSO (133 mg, 1.70 mmol) was added dropwise to a solution of oxalyl chloride (104 mg, 0.82 mmol) in CH_2Cl_2 (6.8 mL) at -78 °C. The solution was stirred for 20 min, then alcohol **2.71** (187 mg, 0.68 mmol) in CH_2Cl_2 (2 mL) was cannulated into the solution and stirred for 45 min at -78 °C. Et_3N (172 mg, 1.70 mmol) was then added and the solution allowed to warm to rt over 1 h. H_2O (7 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 10 mL). The combined organic layers were washed with brine (15 mL) and removed under reduced pressure. The product **2.48** was redissolved in Et_2O (30 mL) and passed through a plug of silica washed with Et_2O (3 × 50 mL). The solvent was removed under reduced pressure and azeotropically dried with toluene (3 × 50 mL). The product was dried under high vacuum for 3 h before taking directly to the next step. To a solution of acetyl thiazolidine thione **2.72** (169 mg, 0.68 mmol) in CH_2Cl_2 (2 mL) at 0 °C was added $TiCl_4$ (0.81 mg, 0.81 mmol) and the yellow slurry was stirred for 5 min. Freshly distilled *i*-Pr_2NEt (105 mg, 0.81 mmol) was slowly added at 0 °C to give a red solution, which was stirred for an additional 20 min at 0 °C. The reaction mixture was cooled to -78 °C, and aldehyde **2.48** (186 mg, 0.68 mmol) was added dropwise. After stirring at -78 °C for 1 h, the mixture was warmed to 0 °C over 3 h. The reaction mixture was quenched by the addition of saturated NH_4Cl (5 mL) and stirred for 5 min at 0 °C. The reaction mixture was extracted with CH_2Cl_2 (30 mL). The combined organic layers were washed with brine solution (20 mL), dried (Na_2SO_4) and filtered. The solvent was concentrated under reduced pressure, and the residue was purified by flash column chromatography eluting with EtOAc/hexanes (1:4) to afford 179 mg (50%) of **2.73** as a yellow oil. 2,6-Lutidine (72.8 mg, 0.68 mmol) followed by TESOTf (0.17 mg, 0.51

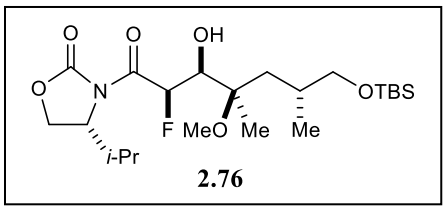
mmol) were added to a solution of **2.73** (179 mg, 0.34 mmol) in CH_2Cl_2 (2 mL) at -78 °C and stirred for 1h. Sat'd aq. $NaHCO_3$ (5 mL) was added and the mixture extracted with CH_2Cl_2 (3 × 10 mL). The combined organic layers were washed with brine (5 mL), filtered over Na_2SO_4 and the solvent removed under reduced pressure. The product was purified by flash column chromatography eluting with 0−5% EtOAc/hexanes to afford 187 mg (86%) of TES protected product as a colorless oil. $NaBH_4$ (44 mg, 1.2 mmol) was added to a solution of the above material above (187 mg, 0.29 mmol) dissolved in EtOH (1 mL) at 0 °C. The reaction mixture was maintained at 0 °C for 30 min then warmed to rt over 60 min. The reaction was quenched with H_2O (3 mL), and the reaction mixture was stirred until both phases were clear. The aqueous phase was extracted with Et_2O (3 × 10 mL), and the combined organic layers were washed with brine and dried over Na_2SO_4. The product was purified by flash column chromatography eluting with 0−5% EtOAc/hexanes to afford 98 mg (80%) of **2.74** as a colorless oil. Spectral data (i.e., ^1H and ^{13}C NMR) were fully consistent those reported in reference (see Ref. 24 in Chapter 1).



2.74b

Stereochemical correlation of 2.74b: The procdure was the same as **2.74a**: $[\alpha]^{23}_D$ +7.9 (c 1.1, $CHCl_3$); IR (neat) 3349, 2929, 2875, 1642, 1376, 1251, 1166, 1062, 835, 775, 737, 667 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 3.85 – 3.77 (m, 2H), 3.61 (td, *J* = 6.7, 3.6 Hz, 1H), 3.47 – 3.41 (m, 1H), 3.36 (ddd, *J* = 9.6, 6.5, 0.9 Hz, 1H), 3.28 (s, 3H), 1.80 (dq, *J* = 19.7, 6.2 Hz, 2H), 1.78 (m, 1H), 1.62 – 1.51 (m, 1H), 1.36 (dd, *J* = 14.6, 6.9 Hz, 1H), 1.17 (s, 3H), 0.95 (ddd, *J* = 9.1, 7.5, 6.2 Hz, 12H), 0.91 – 0.81 (m, 9H), 0.62 (m, 6H), δ 0.03 (s, *J* = 1.0 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 80.1, 75.9, 68.8, 59.6, 50.5, 38.0,

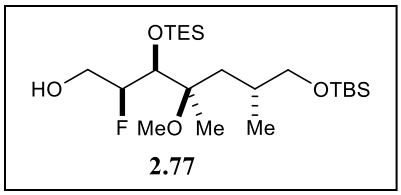
36.1, 31.7, 26.1, 18.8, 18.6, 18.5, 7.2 (2C), 5.5 (2C), -5.2 (2C); HRMS (ESI) calc'd for $C_{22}H_{50}O_4Si_2$ + Na = 457.3145, found 457.3117.



2.76

Fluoro Aldol 2.76: The Dess-Martin periodinane (1.23 g, 2.91 mmol) and $NaHCO_3$ (650 mg, 7.76 mmol) were added to a solution of **2.71** (535.4 mg, 1.94 mmol) in CH_2Cl_2 (20 mL). The reaction mixture was stirred at rt for 2 h. Sat'd aq. $NaHCO_3$ (10 mL), sat'd aq. Na_2SO_3 (10 mL), and H_2O (20 mL) were added, and the reaction mixture was stirred for 30 min before extracting with CH_2Cl_2 (3 × 20 mL). The combined organic layers were washed with brine (50 mL) and filtered over Na_2SO_4. The solvent was removed under reduced pressure. The crude product was redissolved in Et_2O (50 mL) and passed through a plug of silica followed by washing with Et_2O (3 x 50 mL). The solvent was removed under reduced pressure, azeotropically dried with toluene (3 x 15 mL), and placed under high vacuum. This material was used directly without further purification. $TiCl_4$ (1.94 mg, 1.94 mmol) was added to a solution of **2.47** in CH_2Cl_2 (8.8 mL) at -78 °C under a nitrogen atmosphere producing a thick yellow slurry. After 5 min, diisopropylamine (274 mg, 2.12 mmol) was added, and the resulting dark solution was stirred at -78 °C for 1.5 h. Aldehyde **2.48** (531.5 mg, 1.94 mmol) in CH_2Cl_2 (5 mL) was cannulated into the solution, and the reaction mixture was stirred at -78 °C for 1.5 h. The reaction was quenched with 5% citric acid (10 mL), and the aqueous layer was extracted with Et_2O (4 x 15 mL). The combined organic layers were filtered over Na_2SO_4. The solvent was removed under reduced pressure and the product was purified by flash column chromatography eluting with 0-30% EtOAc in hexanes to give 282.9 mg (30%, BRSM 53%) of **2.76** as a colorless

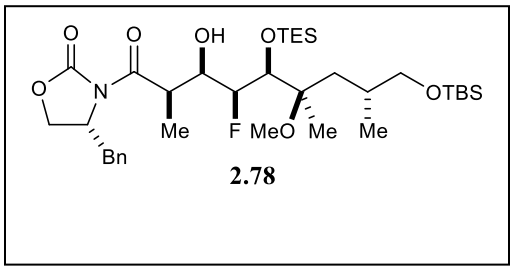
oil: $[\alpha]^{23}_D$ –37.1 (c 1.1, $CHCl_3$); IR (neat) 3476, 2956, 2929, 2856, 2360, 2160, 2034, 1978, 1782, 1720, 1464, 1389, 1302, 1256, 1208, 1102, 836, 775, 713, 668 cm-1; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.12 (dd, *J* = 48.5, 1.6 Hz, 1H, CHF), 4.41 (s, 1H), 4.30 (d, *J* = 8.4 Hz, 1H), 4.27 (d, *J* = 2.6 Hz, 1H), 4.01 – 3.85 (m, 1H), 3.38 (dd, *J* = 9.6, 6.1 Hz, 1H), 3.32 (dd, *J* = 9.6, 6.1 Hz, 1H), 3.19 (s, 3H), 2.87 (d, *J* = 8.0 Hz, 1H), 2.53 (td, *J* = 7.0, 3.6 Hz, 1H), 1.84 – 1.74 (m, 2H), 1.31 (d, *J* = 1.5 Hz, 3H), 1.21 (s, 1H), 0.91 (dd, *J* = 8.6, 6.8 Hz, 6H), 0.85 (d, *J* = 3.6 Hz, 12H), -0.00 (d, *J* = 1.1 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 167.7, 154.0, 89.6 (C-F), 88.1(C-F), 79.0, 73.5, 68.9, 64.4, 59.3, 49.8, 38.2, 31.4, 29.8, 28.0, 26.1, 19.6, 19.0 (2C), 18.5, 18.1, 14.4, -5.3 (2C); ^{19}F NMR (471 MHz, Chloroform-*d*) δ -207.77 (dd, *J* = 48.4, 30.5 Hz); HRMS (ESI) calc'd for $C_{22}H_{42}FNO_6Si$ + Na = 486.2663, found 486.2658.



2.77

Alcohol 2.77: 2,6-Lutidine (26.8 mg, 0.25 mmol) followed by TESOTf (50.2 mg, 0.19 mmol) were added to a solution of **2.73** (59 mg, 0.13 mmol) in CH_2Cl_2 (2 mL) at -78 °C and stirred for 1h. Sat. $NaHCO_3$ (5 mL) was added and the mixture extracted with CH_2Cl_2 (3 x 10 mL). The combined organic fractions were washed with brine (5 mL), filtered over Na_2SO_4 and the solvent removed under reduced pressure. The product was purified by flash column chromatography eluting with 0-5% EtOAc in hexanes to afford 63 mg (86%) of TES protected product as a colorless oil. MeOH (0.005 mL, 0.12 mmol) was added to a solution of the above product (62.5 mg, 0.018 mmol) in THF (1 mL) at 0 °C, followed by $LiBH_4$ (0.24 ml, 0.12 mmol). The reaction mixture was maintained at 0 °C for 30 min and at rt for 60 min. An aqueous solution of NaOH (112 mg in 1.25 mL H_2O) was

added, and the mixture was stirred until both phases were clear. The aqueous phase was further extracted with Et_2O (3 × 10 mL) and the combined organic layers were washed with brine and dried over Na_2SO_4. The product was purified by flash column chromatography eluting with 0-5% EtOAc in hexanes to afford 44 mg (90%) of **2.77** as a colorless oil. $[\alpha]^{23}_D$ –7.2 (c 0.96, $CHCl_3$); IR (neat) 3312, 2952, 2877, 2857, 2160, 2032, 1978, 1462, 1361, 1250, 1083, 1006, 836, 775, 739, 668 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 4.78 – 4.37 (m, 1H, CHF), 3.97 – 3.60 (m, 3H), 3.36 (ddd, *J* = 40.8, 9.7, 6.4 Hz, 2H), 3.20 (s, 3H), 2.76 – 2.41 (m, 1H), 1.78 (td, *J* = 6.8, 4.4 Hz, 1H), 1.62 (dd, *J* = 14.9, 4.4 Hz, 1H), 1.44 (dd, *J* = 14.9, 7.2 Hz, 1H), 1.25 (d, *J* = 1.2 Hz, 5H), 0.98 – 0.93 (m, 12H), 0.89 (s, 9H), 0.73 – 0.53 (m, 6H), 0.04 (s, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 95.1 (C-F), 93.7 (C-F), 80.0, 76.4, 76.3, 68.9, 63.3, 63.1, 49.4, 36.6, 31.6, 26.1, 20.0, 18.7, 18.5, 7.1, 6.9, 6.8, 5.5, 5.3, 5.1, -5.3, -5.2; HRMS (ESI) calc'd for $C_{22}H_{49}FNO_4Si_2$ + Na = 475.3051, found 475.3055.

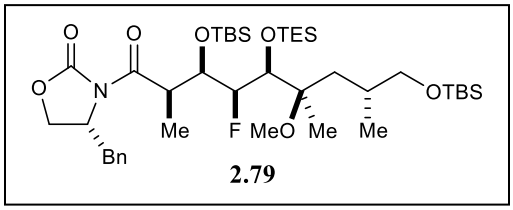


2.78

Aldol 2.78: The Dess-Martin periodinane (102.43 mg, 0.24 mmol) and $NaHCO_3$ (54.1 mg, 0.544 mmol) was added to a solution of **2.77** (73 mg, 0.16 mmol) in CH_2Cl_2 (2 mL). The solution was stirred at rt for 2 h. Sat'd aq. $NaHCO_3$ (5 mL), sat'd aq. Na_2SO_3 (5 mL) and H_2O (10 mL) were added to the reaction vessel and stirred for 30 min before extracting with CH_2Cl_2 (3 x 15 mL). The combined organic layers were washed with brine (30 mL) and filtered over Na_2SO_4. The solvent was removed under reduced pressure. The product was redissolved in Et_2O (30 mL) and passed through a plug of silica washed with Et_2O (3 x 30 mL). The solvent was removed under reduced pressure and azeotropically dried with toluene (3 x 15 mL). The product was dried under high vacuum. This material was used

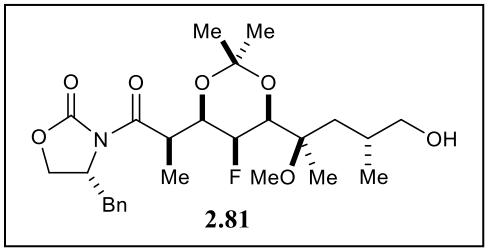
directly without further purification. Et_3N (24.2 mg, 0.24 mmol) was added dropwise to a solution of (*R*)-4-benzyl-3-propionyl-2-oxazolidinone **2.45** (37.56 g, 0.16 mmol) and Bu_2BOTf (0.21 mL, 0.22 mmol, 1 M in CH_2Cl_2) in CH_2Cl_2 (2 mL). The solution changed from red to yellow and was subsequently cooled to -78 °C. Aldehyde **2.46** in CH_2Cl_2 (2 mL) was cannulated into the solution and stirred at -78 °C for 20 min and then at 0 °C for 1 h. Phosphate buffer (pH 7, 0.2 M aq. Na_2HPO_4:0.1 M aq. citric acid, 82:18, 1.14 mL) and MeOH (2.29 mL) was added to the reaction mixture. The solution becomes cloudy and a solution of MeOH:30% H_2O_2 (2:1, 2.28 mL) was added and stirred at 0 °C for 1 h. The solution was then concentrated under reduced pressure and the remaining aqueous layer was extracted with EtOAc (3 x 15 mL). The combined organic layers were washed with sat. $NaHCO_3$ (20 mL), brine (20 mL) and filtered over Na_2SO_4. The solvent was removed under reduced pressure and the product was purified by flash column chromatography eluting with 0-30% EtOAc in hexanes to give 70 mg (64%) of **2.78** as a colorless oil. $[\alpha]^{23}_D$ –53.0 (c 0.23, $CHCl_3$); IR (neat) 3313, 2953, 2929, 2856, 2450, 2160, 2031, 1978, 1784, 1456, 1384, 1349, 1250, 1210, 1097, 1007, 969, 836, 815, 775, 740, 703 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.41 – 7.11 (m, 5H), 4.67 (td, *J* = 6.4, 3.3 Hz, 1H), 4.55 – 4.38 (m, 1H), 4.31 – 4.09 (m, 3H), 3.99 (q, *J* = 7.2 Hz, 1H), 3.88 (dd, *J* = 17.3, 6.6 Hz, 1H), 3.46 (dd, *J* = 9.7, 6.1 Hz, 1H), 3.38 – 3.32 (m, 2H), 3.27 (s, 3H), 3.24 (s, 1H), 2.78 (dd, *J* = 13.4, 9.5 Hz, 1H), 2.00 – 1.79 (m, 1H), 1.71 (dd, *J* = 14.8, 4.5 Hz, 1H), 1.40 (d, *J* = 6.9 Hz, 3H), 1.27 (s, 3H), 1.03 – 0.80 (m, 21H), 0.63 (q, *J* = 7.9 Hz, 6H), 0.06 (d, *J* = 4.3 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.6, 152.8, 135.4, 129.6, 129.1, 127.5, 95.2 (C-F), 93.8 (C-F), 80.1 (2C), 71.8, 71.7, 68.9, 66.4, 66.2, 55.4, 49.7, 40.5, 40.4 (2C), 38.0, 36.3, 32.1, 31.7,

29.9, 26.1, 20.3, 19.0, 18.5, 14.6, 14.4, 7.1, 5.3, -5.2; HRMS (ESI) calc'd for $C_{35}H_{62}FNO_7Si_2$ + Na = 706.3941, found 706.3925.



2.79

Oxazolidinone 2.79: To a solution of **2.78** (65 mg, 0.14 mmol) in CH_2Cl_2 (1 mL) at 0 °C was added 2,6-lutidine (30 mg, 0.28 mmol) followed by TBSOTf (56 mg, 0.21 mmol). The reaction mixture was stirred for 30 min at this temperature. Sat'd aq. $NaHCO_3$ (5 mL) was added, and the reaction mixture was extracted with CH_2Cl_2 (3 x 10 mL). The combined organic layers were washed with brine (5 mL), filtered over Na_2SO_4, and the solvent was concentrated under reduced pressure. The residue was purified by flash column chromatography eluting with 0–5% EtOAc/hexanes to afford 560 mg (95%) of **2.79** as a colorless oil: $[\alpha]^{23}_D$ –15.5 (c 0.60, $CHCl_3$); IR (neat) 2953, 2929, 2856, 2450, 2160, 2031, 1978, 1784, 1456, 1384, 1349, 1250, 1210, 1097, 1007, 969, 836, 815, 775, 740, 703 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.36 – 7.27 (m, 3H), 7.23 – 7.18 (m, 2H), 4.69 (dtd, *J* = 9.3, 5.7, 3.4 Hz, 1H), 4.55 – 4.42 (m, 2H), 4.16 (d, *J* = 5.8 Hz, 2H), 3.81 (dd, *J* = 25.8, 1.4 Hz, 1H), 3.73 (q, *J* = 6.8 Hz, 1H), 3.40 (dd, *J* = 9.6, 5.3 Hz, 1H), 3.29 – 3.21 (m, 2H), 3.11 (s, 3H), 2.75 (dd, *J* = 13.4, 9.5 Hz, 1H), 1.71 (d, *J* = 14.0 Hz, 2H), 1.42 (dd, *J* = 15.5, 9.5 Hz, 1H), 1.31 (d, *J* = 7.1 Hz, 3H), 1.19 (d, *J* = 1.4 Hz, 3H), 1.00 – 0.85 (m, 34H), 0.67 – 0.48 (m, 6H), 0.14 (m, 6H), 0.04 (d, *J* = 3.6 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.0, 153.1, 135.4, 129.6, 129.1, 127.5, 92.8 (C-F), 91.3 (C-F), 80.2, 74.5, 74.3, 70.8, 70.6, 69.3, 66.3, 55.2, 53.8, 48.4, 40.2 (2C), 38.1, 35.9, 31.7, 29.9, 26.4, 26.2, 19.4 (2C), 18.8, 18.5, 17.4, 12.0, 7.3, 5.5, -3.9 (2C), -4.9, -5.2, -5.3; HRMS (ESI) calc'd for $C_{41}H_{76}FNO_7Si_3$ + Na = 820.4806, found 820.4788.

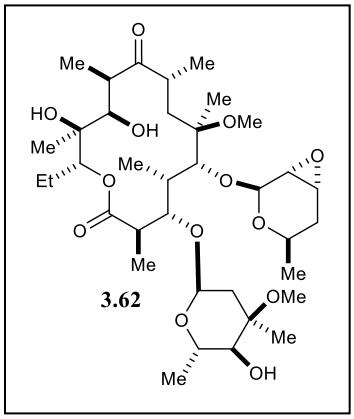


2.81

Acetonide analysis of 2.81: Camphorsulfonic acid (10.2 mg, 0.044 mmol) was added to a solution of aldol **2.78** (15 mg, 0.022 mmol) in MeOH (1 mL) at 0 °C. The reaction mixture was stirred for 2 h. The solvent was concentrated under reduced pressure, and the crude product was redissolved in EtOAc (5 mL). The organic layer was washed with sat. $NaHCO_3$ (5 mL), brine (5 mL) and filtered over Na_2SO_4. The solvent was removed under reduced pressure and the product purified by flash column chromatography eluting with 0-40% EtOAc in hexanes to afford 5 mg (88%) **2.80** as a colorless oil. The crude triols were dissolved in CH_2Cl_2, then 2,2-dimethoxypropane (0.014mg, 0.014mmol) and a catalytic amount of pyridinium *p*-toluenesulfonate were added. The reaction mixture was stirred for 3 h then filtered through a plug of cotton. The solvent was concentrated under reduced pressure, and the residue was purified by flash column chromatography eluting with 0–10 % EtOAc/hexanes to afford 4 mg (75%) of **2.81** as a colorless oil. $[\alpha]^{23}_D$ –30.0 (c 0.20, $CHCl_3$); IR (neat) 2981, 2443, 2360, 2160, 2031, 1978, 1781, 1700, 1684, 1437, 1383, 1351, 1261, 1206, 1156, 1078, 1049, 763, 750 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.36 – 7.32 (m, 2H), 7.31 – 7.28 (m, 1H), 7.23 – 7.17 (m, 2H), 4.73 – 4.67 (m, 1H), 4.53 (dt, *J* = 48.9, 1.0 Hz, 1H), 4.30 – 4.24 (m, 1H), 4.19 (dd, *J* = 9.1, 2.5 Hz, 1H), 4.11 – 4.00 (m, 2H), 3.83 (dd, *J* = 35.2, 1.1 Hz, 1H), 3.55 (dt, *J* = 6.7, 3.9 Hz, 1H), 3.42 (t, *J* = 5.9 Hz, 1H), 3.34 (s, 3H), 3.28 (ddd, *J* = 11.2, 7.6, 4.3 Hz, 1H), 3.22 (dd, *J* = 13.4, 3.5 Hz, 1H), 2.79 (dd, *J* = 13.4, 9.4 Hz, 1H), 1.96 – 1.88 (m, 1H), 1.75 – 1.67 (m, 1H), 1.57 (d, *J* = 3.4 Hz, 1H), 1.48 (s, 6H), 1.35 (d, *J* = 6.6 Hz, 3H), 1.32 (s, 3H), 0.93 (d, *J* = 7.0 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.9, 152.5, 135.2, 129.6, 129.1, 127.6, 99.4, 83.5 (C-F), 82.0 (C-F), 78.8,

75.6, 72.7, 69.0, 66.3, 55.1, 51.3, 40.9, 39.0, 38.0, 31.4, 29.9, 20.1, 19.0, 18.4, 15.2; HRMS (ESI) calc'd for $C_{26}H_{38}FNO_7$ + Na = 518.2525, found 518.2544.

5.2 Chapter 3: Macrolide Antibiotics with Desosamine Modifications



3.62

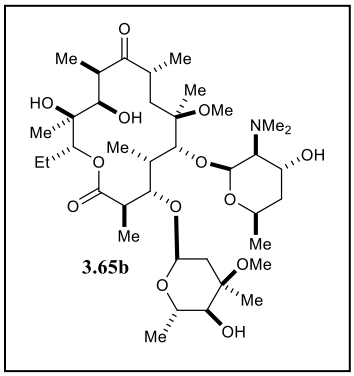
Clarithromycin-Epoxide 3.62: MeI (2.0 g, 13.37 mmol) was added to a solution of clarithromycin (**3.3**) (2.00 g, 2.67 mmol) in acetone (50 mL) at 0 °C, and the reaction mixture was warmed to rt at which time a solid precipitate was formed. The solution was cooled to 0 °C and stirred an additional hour. The reaction mixture was concentrated under reduced pressure to remove solvent and MeI. The solid was filtered off, washed with EtOAc (3 x 50 mL), and dried under vacuum and used directly in the next step without further purification. Ag_2O (3.08 g, 13.4 mmol) was added to a solution of clarithromycin ammonium salt **3.61** (2.37 g, 2.67 mmol) in toluene (26.7 mL) in a sealed tube, which was heated to 120 °C for 15 h. The reaction was cooled to rt, filtered through a Celite pad, and washed with EtOAc (3 x 50 mL). The combined organic phases were concentrated under vacuum. The residue was purified by flash column chromatography eluting with 8–13% acetone/hexane to give 711.72 mg of **3.62** (60%) as a yellow foam. $[\alpha]^{23}_D$ –87.5 (c 1.1, $CHCl_3$); IR (neat) 3473, 2974, 2443, 2159, 2031, 1978, 1730, 1690, 1377, 1346, 1169, 1129, 1049, 1034, 1021, 905, 832, 738 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.07 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.92 (d, *J* = 5.0 Hz, 1H), 4.91 (s, 1H), 3.99 – 3.93 (m, 2H), 3.77 – 3.70 (m, 2H), 3.67 (d, *J* = 6.7 Hz, 1H), 3.39 (ddd, *J* = 11.0, 6.3, 2.9 Hz, 1H), 3.35 (s, 3H), 3.33 – 3.29 (m, 1H), 3.18 (d, *J* = 4.2 Hz, 2H), 3.04 (s, 3H), 3.01 – 2.96 (m, 2H), 2.87 (ddd, *J* = 14.5, 8.4, 4.8 Hz, 1H), 2.61 – 2.54 (m, 1H), 2.37 (dd, *J* = 15.3, 0.9 Hz, 1H), 2.19 (d, *J* = 10.8 Hz, 1H), 2.02 – 1.88 (m, 3H), 1.81 – 1.71 (m, 1H), 1.68 – 1.53 (m, 3H), 1.47 (ddd, *J* = 14.3, 11.1, 7.2 Hz, 1H), 1.39 (s, 3H), 1.27 (d, *J* = 6.2 Hz, 3H), 1.25 – 1.18 (m, 6H), 1.16

– 1.10 (m, 12H), 1.03 (d, J = 7.5 Hz, 3H), 0.84 (t, J = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ175.7, 98.0, 96.4, 83.4, 78.4, 78.2, 78.1, 76.8, 74.4, 72.7, 69.2, 65.7, 62.0, 53.9, 51.8, 50.8, 49.4, 45.4, 45.1, 39.0, 39.0, 37.1, 35.0, 32.7, 21.6, 21.2, 21.1, 19.7, 18.9, 18.1, 16.2, 16.1, 12.5, 10.7, 9.6; HRMS (ESI) calc'd for $C_{36}H_{62}O_3$ + Na = 725.4088, found 725.4077.

General procedure for the epoxide ring-opening of 3.62 with secondary amines

A solution of **3.62** (1.0 equiv) in toluene (3 mL) was treated with the secondary amine (5.0 equiv) and $Sn(OTf)_3$ (0.05 equiv) at rt. The reaction mixture was heated to 120 °C and stirred for 24-48 h at this temperature. After cooling to rt, the reaction mixture was diluted with EtOAc (10 mL) and washed with H_2O (5 mL). The layers were separated and the aqueous phase was extracted with EtOAc (2 x 5 mL). The combined organic phases were dried over Na_2SO_4, filtered, and concentrated under reduced pressure. The residue was purified by flash column chromatography eluting with MeOH/CH_2Cl_2/NH_4OH (5:95:0.1) to afford the corresponding natural 3'-amino desosamine and unnatural 2'-amino desosamine analogs **3.65-3.73** as foams.

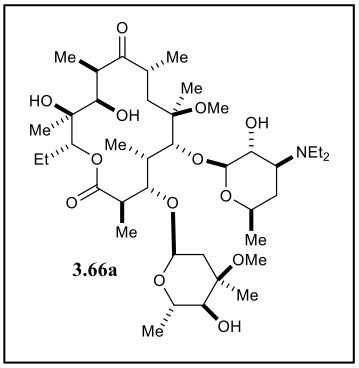
Clarithromycin 3.65a (prepared by reaction with *N,N*-dimethylamine): Spectral data were fully consistent with those reported in Ref. 12 in Chapter 3.



3.65b

Clarithromyin analog 3.65b: $[\alpha]^{23}_D$ –65.7 (c 0.8, $CHCl_3$); IR (neat) 3470, 2970, 2943, 2839, 1730, 1690, 1425, 1380, 1265, 1214, 1110, 1052, 749, 669 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.05 (dd, J = 11.1, 2.3 Hz, 1H), 4.92 (d, J = 4.7 Hz, 1H), 4.44 (d, J = 7.2 Hz, 1H), 4.03 – 3.93 (m, 2H), 3.79 – 3.71 (m,

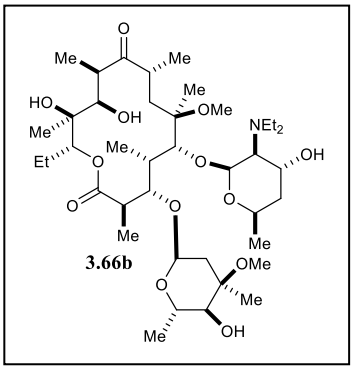
2H), 3.66 (d, *J* = 7.3 Hz, 1H), 3.54 – 3.37 (m, 2H), 3.32 (s, 3H), 3.23 – 3.14 (m, 2H), 3.08 – 2.93 (m, 5H), 2.88 (dd, *J* = 9.2, 7.2 Hz, 1H), 2.61 – 2.51 (m, 1H), 2.45 (s, 1H), 2.36 (dd, *J* = 15.3, 1.1 Hz, 1H), 2.31 (s, 6H), 2.18 (d, *J* = 10.1 Hz, 1H), 1.96 – 1.86 (m, 2H), 1.86 – 1.79 (m, 1H), 1.70 (dd, *J* = 14.8, 2.1 Hz, 1H), 1.58 (dd, *J* = 15.1, 5.0 Hz, 1H), 1.52 – 1.42 (m, 1H), 1.40 (s, 3H), 1.30 (d, *J* = 6.2 Hz, 3H), 1.25 (s, 3H), 1.21 (dd, *J* = 11.8, 6.7 Hz, 6H), 1.13 (d, *J* = 1.8 Hz, 3H), 1.12 – 1.06 (m, 9H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.2, 176.0, 102.9, 96.2, 81.0, 78.6, 78.5, 78.1, 74.4, 72.9, 71.1, 69.2, 68.3, 65.8, 65.1, 50.8, 49.6, 45.4, 45.2, 40.5, 39.5, 39.3, 37.4, 35.0, 29.9, 29.0, 21.6 (2C), 21.2, 19.9, 18.9, 18.2, 16.2, 16.1, 12.5, 10.8, 9.3; HRMS (ESI) calc'd for $C_{38}H_{70}NO_{13}$ + H = 748.4847, found 748.4812.



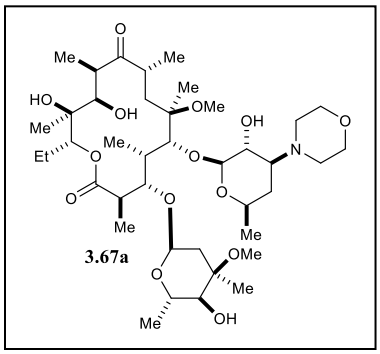
3.66a

Clarithromyin analog 3.66a: $[\alpha]^{23}_D$ –61.2 (c 0.7, $CHCl_3$); IR (neat) 3473, 2978, 2438, 2159, 2030, 1730, 1690, 1457, 1346, 1265, 1215, 1169, 1108, 1073, 1052, 995, 749, 667 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.05 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.93 (d, *J* = 4.8 Hz, 1H), 4.46 (d, *J* = 7.2 Hz, 1H), 4.07 – 3.92 (m, 2H), 3.79 – 3.70 (m, 2H), 3.65 (d, *J* = 7.2 Hz, 1H), 3.46 (t, *J* = 8.6 Hz, 1H), 3.33 (s, 3H), 3.17 (d, *J* = 11.3 Hz, 2H), 3.06 – 2.94 (m, 5H), 2.93 – 2.83 (m, 1H), 2.72 – 2.47 (m, 3H), 2.42 – 2.27 (m, 3H), 2.14 (d, *J* = 10.3 Hz, 1H), 1.96 – 1.78 (m, 2H), 1.76 – 1.54 (m, 5H), 1.53 – 1.41 (m, 1H), 1.40 (s, 3H), 1.30 (d, *J* = 6.2 Hz, 3H), 1.25 (s, 3H), 1.20 (dd, *J* = 6.7, 3.9 Hz, 6H), 1.11 (dd, *J* = 4.2, 2.8 Hz, 12H), 1.04 (t, *J* = 7.0 Hz, 6H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.3, 176.0, 103.0, 96.1, 81.0, 78.5, 78.4, 78.2, 74.4, 72.9, 70.6, 69.2, 65.8, 62.1, 50.8, 49.6, 45.5, 45.2, 43.4, 39.5, 39.3, 37.3, 35.0,

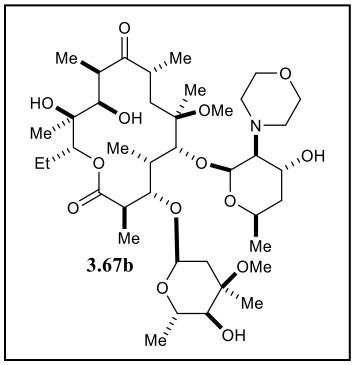
32.1, 30.7, 29.9, 24.0, 21.6, 21.2, 19.9, 18.8, 18.2, 16.2, 16.1, 14.7, 12.5, 10.8, 9.2; HRMS (ESI) calc'd for $C_{40}H_{74}NO_{13}$ + H = 776.5160, found 776.5195.



Clarithromyin analog 3.66b: $[\alpha]^{23}_{D}$ –85.8 (c 1.0, $CHCl_3$); IR (neat) 3470, 2977, 2446, 2160, 2031, 1730, 1665, 1457, 1377, 1345, 1265, 1215, 1169, 1105, 1075, 1053, 995, 748, 666 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.05 (dd, *J* = 11.1, 2.3 Hz, 1H), 5.01 (d, *J* = 2.9 Hz, 1H), 4.90 (d, *J* = 4.6 Hz, 1H), 4.06 (ddd, *J* = 12.5, 7.8, 4.5 Hz, 1H), 3.96 (s, 2H), 3.83 – 3.72 (m, 2H), 3.62 (d, *J* = 7.2 Hz, 1H), 3.30 (s, 3H), 3.21 (s, 1H), 3.07 – 2.93 (m, 6H), 2.80 (ddt, *J* = 21.3, 14.3, 7.2 Hz, 1H), 2.67 – 2.56 (m, 1H), 2.49 (s, 1H), 2.33 (dd, *J* = 15.1, 1.4 Hz, 1H), 1.92 (ddd, *J* = 14.3, 7.6, 2.3 Hz, 1H), 1.84 (t, *J* = 7.4 Hz, 1H), 1.80 – 1.71 (m, 1H), 1.67 (td, *J* = 13.9, 10.3 Hz, 1H), 1.55 (ddd, *J* = 14.4, 10.6, 3.9 Hz, 2H), 1.47 (ddd, *J* = 14.3, 11.1, 7.1 Hz, 1H), 1.41 (s, 3H), 1.30 (d, *J* = 6.3 Hz, 3H), 1.26 – 1.16 (m, 10H), 1.16 – 1.09 (m, 9H), 1.02 (d, *J* = 7.4 Hz, 6H), 0.96 (d, *J* = 7.4 Hz, 3H), 0.85 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.0, 99.8, 96.3, 82.4, 78.5, 78.4, 77.9, 76.8, 74.4, 73.0, 69.2, 66.0 (2C), 61.2, 50.7, 49.6, 46.0, 45.5, 45.1, 39.6, 39.5, 37.5, 36.2, 35.0, 29.9, 21.8, 21.6, 21.2, 20.3, 18.6, 18.2, 16.3, 16.1, 14.7, 12.4, 10.8, 9.4; HRMS (ESI) calc'd for $C_{40}H_{74}NO_{13}$ + H = 776.5160, found 776.5183.

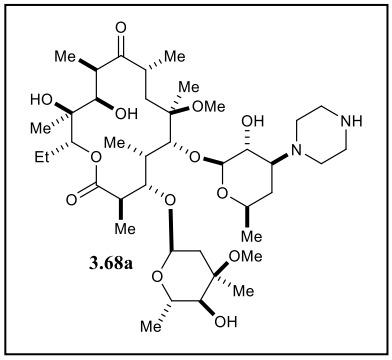


Clarithromyin analog 67a: $[\alpha]^{23}_{D}$ –74.5 (c 0.6, $CHCl_3$); IR (neat) 3462, 2971, 2446, 2159, 2031, 1977, 1729, 1664, 1457, 1378, 1345, 1216, 1167, 1111, 1052, 751, 668 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.05 (dd, *J* = 11.0, 2.3 Hz, 1H), 4.92 (d, *J* = 4.8 Hz, 1H), 4.46 (d, *J* = 7.2 Hz, 1H), 4.05 – 3.93 (m, 2H), 3.78 – 3.74 (m, 2H), 3.72 (d, *J* = 6.3 Hz, 2H), 3.70 – 3.65 (m, 3H), 3.49 (ddd, *J* = 10.7, 6.1, 2.0 Hz, 1H), 3.32 (s, 3H), 3.24 (dd, *J* = 10.4, 7.2 Hz, 1H), 3.19 – 3.16 (m, 1H), 2.72 (d, *J* = 11.3 Hz, 2H), 2.62 – 2.52 (m, 1H), 2.45 (d, *J* = 10.9 Hz, 3H), 2.36 (d, *J* = 15.2 Hz, 1H), 2.16 (d, *J* = 10.1 Hz, 1H), 1.96 – 1.87 (m, 2H), 1.83 (dd, *J* = 14.7, 11.8 Hz, 1H), 1.75 – 1.66 (m, 2H), 1.58 (dd, *J* = 15.2, 5.1 Hz, 1H), 1.47 (ddd, *J* = 14.3, 11.1, 7.1 Hz, 1H), 1.40 (s, 3H), 1.30 (d, *J* = 6.2 Hz, 3H), 1.27 – 1.21 (m, 8H), 1.20 (d, *J* = 7.2 Hz, 3H), 1.13 – 1.07 (m, 12H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.2, 176.0, 102.8, 96.2, 81.0, 78.6, 78.5, 78.1, 74.4, 72.9, 70.2, 69.2, 68.9, 67.5, 66.6, 65.9, 50.8, 49.7, 49.0, 45.4, 45.2, 39.4, 39.3, 37.4, 35.0, 30.4, 29.8, 21.6, 21.6, 21.2, 19.9, 18.9, 18.2, 16.2, 16.1, 12.5, 10.8, 9.2; HRMS (ESI) calc'd for $C_{40}H_{72}NO_{14}$ + H = 790.4953, found 790.4977.



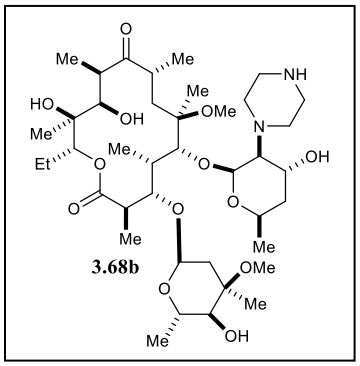
Clarithromyin analog 3.67b: $[\alpha]^{23}_{D}$ –71.9 (c 0.7, $CHCl_3$); IR (neat) 3447, 2971, 2446, 2159, 2031, 1977, 1733, 1457, 1378, 1345, 1216, 1168, 1111, 1053, 999, 754, 668 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.06 (dd, *J* = 11.1, 2.2 Hz, 1H), 5.01 (s, 1H), 4.89 (d, *J* = 4.4 Hz, 1H), 4.07 (dq, *J* = 12.8, 6.4 Hz, 2H), 3.97 (s, 2H), 3.79 – 3.73 (m, 2H), 3.71 – 3.54 (m, 5H), 3.29 (s, 3H), 3.21 (s, 1H),

3.07 – 2.93 (m, 6H), 2.89 – 2.77 (m, 2H), 2.66 – 2.50 (m, 2H), 2.39 – 2.28 (m, 1H), 2.02 – 1.75 (m, 3H), 1.67 (p, *J* = 12.4, 10.9 Hz, 2H), 1.56 (dd, *J* = 15.0, 4.8 Hz, 1H), 1.52 – 1.44 (m, 0H), 1.43 (s, 3H), 1.30 (d, *J* = 6.3 Hz, 3H), 1.23 (s, 4H), 1.19 (dd, *J* = 8.3, 6.7 Hz, 7H), 1.16 – 1.11 (m, 9H), 0.97 (d, *J* = 7.4 Hz, 3H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.9, 96.4, 84.1, 78.7, 78.4, 77.9, 76.8, 74.3, 73.1, 69.6, 69.2, 68.1, 66.8, 66.0, 64.8, 52.8, 50.8, 49.5, 45.4, 45.2, 39.8, 39.4, 37.5, 36.6, 35.0, 29.8, 21.7, 21.6, 21.2, 20.2, 18.5, 18.3, 16.3, 16.1, 12.4, 10.8, 9.9; HRMS (ESI) calc'd for $C_{40}H_{72}NO_{14}$ + H = 790.4953, found 790.4933.

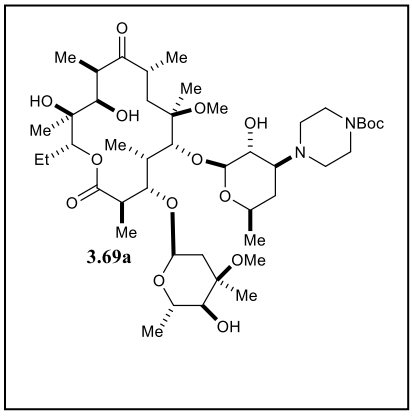


3.68a

Clarithromyin analog 3.68a: $[\alpha]^{23}_D$ –68.0 (c 1.3, $CHCl_3$); IR (neat) 3447, 2971, 2936, 2446, 2159, 2031, 1977, 1734, 1684, 1457, 1378, 1168, 1052, 1010, 758, 668 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.04 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.91 (d, *J* = 4.9 Hz, 1H), 4.44 (d, *J* = 7.3 Hz, 1H), 3.98 (dt, *J* = 12.5, 6.2 Hz, 1H), 3.75 (t, *J* = 4.7 Hz, 2H), 3.65 (d, *J* = 7.3 Hz, 1H), 3.55 – 3.40 (m, 1H), 3.31 (s, 3H), 3.22 (dd, *J* = 10.4, 7.2 Hz, 1H), 3.05 – 2.82 (m, 9H), 2.74 (ddd, *J* = 10.4, 6.8, 3.1 Hz, 2H), 2.56 (dq, *J* = 12.2, 5.6 Hz, 1H), 2.51 – 2.29 (m, 4H), 2.00 – 1.76 (m, 3H), 1.74 – 1.61 (m, 2H), 1.57 (dd, *J* = 15.2, 5.1 Hz, 1H), 1.46 (ddd, *J* = 14.3, 11.1, 7.1 Hz, 1H), 1.39 (s, 3H), 1.29 (d, *J* = 6.2 Hz, 4H), 1.26 – 1.16 (m, 12H), 1.14 – 1.05 (m, 13H), 0.83 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.1, 176.0, 102.8, 96.2, 80.9, 78.6, 78.5, 78.1, 74.4, 72.8, 70.3, 69.2, 69.0, 66.7, 65.9, 50.8, 49.6, 49.1, 46.2, 45.4, 45.2, 39.4, 39.3, 37.3, 35.0, 30.6, 29.8, 21.6, 21.6, 21.1, 19.9, 18.8, 18.2, 16.4, 16.1, 12.4, 10.7, 9.2; HRMS (ESI) calc'd for $C_{40}H_{72}N_2O_{13}$ + H = 789.5113, found 789.5122.

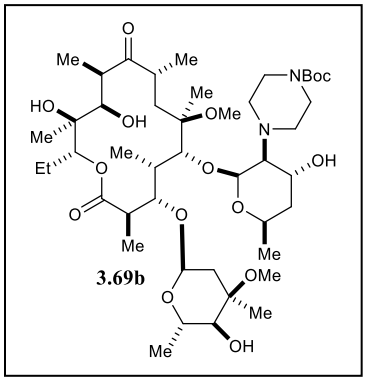


Clarithromyin analog 3.68b: $[\alpha]^{23}_{D}$ –46.0 (c 0.4, $CHCl_3$); IR (neat) 3447, 2971, 2446, 2159, 2031, 1977, 1730, 1684, 1457, 1378, 1168, 1110, 1069, 1052, 1000, 906, 758, 648 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 5.00 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.92 (d, *J* = 2.4 Hz, 1H), 4.83 (d, *J* = 4.4 Hz, 1H), 4.07 – 3.96 (m, 2H), 3.93 – 3.80 (m, 2H), 3.73 – 3.64 (m, 2H), 3.51 (d, *J* = 7.1 Hz, 1H), 3.23 (s, 3H), 3.00 – 2.87 (m, 7H), 2.85 – 2.71 (m, 6H), 2.59 – 2.47 (m, 1H), 2.45 (d, *J* = 2.7 Hz, 1H), 2.26 (d, *J* = 15.0 Hz, 1H), 1.93 – 1.70 (m, 2H), 1.67 – 1.55 (m, 1H), 1.52 – 1.40 (m, 2H), 1.37 (s, 3H), 1.24 (d, *J* = 6.2 Hz, 4H), 1.18 (d, *J* = 8.1 Hz, 7H), 1.09 (tt, *J* = 14.1, 7.9 Hz, 16H), 0.90 (d, *J* = 7.3 Hz, 3H), 0.79 (t, *J* = 7.3 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.9, 100.9, 96.4, 85.1, 79.1, 78.4, 77.7, 74.4, 73.3, 69.3, 67.0, 66.2, 64.4, 53.6, 50.7, 49.5, 45.4, 45.0, 39.6, 39.3, 37.5, 36.6, 35.1, 32.1, 29.9, 21.7, 21.6, 21.2, 20.1, 18.5, 18.4, 16.3, 16.0, 14.3, 12.5, 10.8, 10.0; HRMS (ESI) calc'd for $C_{40}H_{72}N_2O_{13}$ + H = 789.5113, found 789.5139.



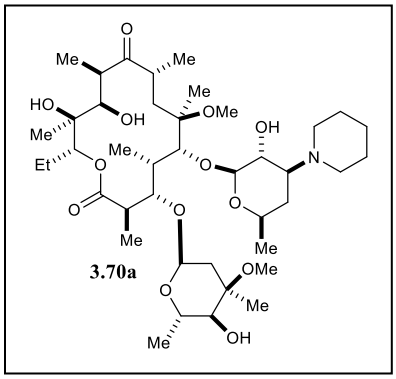
Clarithromyin analog 3.69a: $[\alpha]^{23}_{D}$ –45.4 (c 0.9, $CHCl_3$); IR (neat) 3447, 2971, 2934, 2446, 2159, 2030, 1977, 1734, 1696, 1614, 1457, 1366, 1248, 1169, 1125, 1072, 1054, 1002, 756, 668 cm-1; ^1H NMR (500 MHz, $CDCl_3$) δ 5.07 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.94 (d, *J* = 4.8 Hz, 1H), 4.48 (d, *J* = 7.2 Hz, 1H), 4.04 – 3.94 (m, 2H), 3.80 – 3.74 (m, 2H), 3.69 (d, *J* = 7.2 Hz, 1H), 3.54 – 3.41 (m, 5H), 3.34 (s, 3H), 3.28 – 3.23 (m, 1H), 3.20 (d, *J* = 0.9 Hz, 1H), 3.12 – 2.98 (m, 5H), 2.91 (dt, *J* = 9.4, 7.4 Hz, 1H), 2.73

– 2.64 (m, 2H), 2.60 (ddd, *J* = 11.7, 6.9, 1.9 Hz, 1H), 2.48 (ddd, *J* = 13.4, 10.4, 3.7 Hz, 1H), 2.43 – 2.35 (m, 1H), 2.18 (d, *J* = 10.1 Hz, 1H), 1.93 (ddt, *J* = 10.0, 4.8, 2.6 Hz, 2H), 1.86 (dd, *J* = 14.8, 11.8 Hz, 1H), 1.76 – 1.66 (m, 2H), 1.66 – 1.57 (m, 2H), 1.54 – 1.49 (m, 0H), 1.48 (s, 9H), 1.42 (s, 3H), 1.32 (d, *J* = 6.3 Hz, 3H), 1.27 (s, 4H), 1.23 (dd, *J* = 9.6, 6.7 Hz, 6H), 1.16 – 1.10 (m, 12H), 0.86 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.0, 154.7, 102.7, 96.2, 81.0, 80.0, 78.6, 78.5, 78.1, 76.8, 74.4, 72.9, 70.4, 69.2, 69.0, 66.5, 65.9, 50.8, 49.7, 48.5, 45.4, 45.2, 39.4, 39.3, 37.3, 35.0, 30.6, 29.8, 28.5, 21.6, 21.5, 21.1, 19.9, 18.8, 18.2, 16.1(2C), 12.4, 10.7, 9.2; HRMS (ESI) calc'd for $C_{45}H_{80}N_2O_{15}$ + H = 889.5637, found 889.5614.

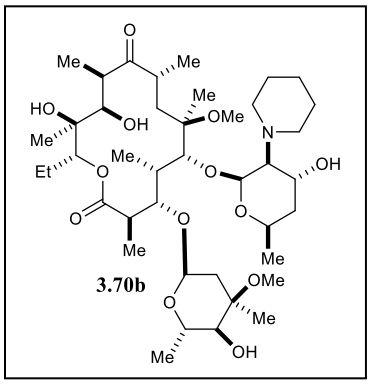


Clarithromyin analog 69b: $[\alpha]^{23}_D$ –67.3 (c 0.7, $CHCl_3$); IR (neat) 3462, 2971, 2446, 2159, 2031, 1978, 1734, 1684, 1457, 1419, 1366, 1247, 1168, 1111, 1052, 1008, 755, 668 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.06 (dd, *J* = 11.0, 2.2 Hz, 1H), 5.00 (d, *J* = 2.5 Hz, 1H), 4.89 (d, *J* = 4.3 Hz, 1H), 4.06 (dd, *J* = 9.1, 6.2 Hz, 2H), 3.97 (s, 2H), 3.80 – 3.71 (m, 2H), 3.58 (d, *J* = 7.2 Hz, 1H), 3.29 (s, 8H), 3.21 (s, 1H), 3.07 – 2.89 (m, 4H), 2.87 – 2.68 (m, 2H), 2.62 – 2.44 (m, 3H), 2.32 (dd, *J* = 15.2, 1.4 Hz, 1H), 1.98 – 1.80 (m, 2H), 1.71 (dd, *J* = 36.1, 13.3 Hz, 2H), 1.54 (td, *J* = 15.0, 6.0 Hz, 2H), 1.48 – 1.45 (m, 1H), 1.43 (d, *J* = 2.4 Hz, 12H), 1.30 (d, *J* = 6.3 Hz, 3H), 1.23 (s, 3H), 1.19 (dd, *J* = 6.7, 5.0 Hz, 6H), 1.15 (s, 3H), 1.11 (dd, *J* = 7.0, 5.5 Hz, 6H), 0.95 (d, *J* = 7.5 Hz, 3H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.0, 175.9, 154.9, 100.8, 96.4, 84.0, 79.6, 78.6, 78.4, 77.9, 74.4, 73.0, 69.2, 66.7, 66.0, 64.7, 52.3, 52.0, 50.8, 49.6, 45.4, 45.2, 39.8, 39.4, 37.5,

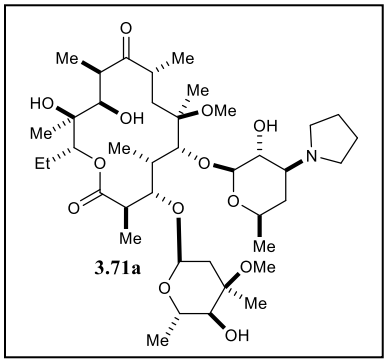
36.8, 36.0, 35.0, 29.9, 28.6, 21.7, 21.6, 21.2, 20.2, 18.5, 18.3, 16.3, 16.1, 12.4, 10.7, 9.8; HRMS (ESI) calc'd for $C_{45}H_{80}N_2O_{15}$ + H = 889.5637, found 889.5653.



Clarithromyin analog 3.70a: $[\alpha]^{23}_D$ –55.8 (c 1.1, $CHCl_3$); IR (neat) 3447, 2971, 2447, 2159, 2031, 1730, 1684, 1457, 1378, 1248, 1345, 1169, 1109, 1072, 1052, 906, 729, 647 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 5.04 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.92 (d, *J* = 4.8 Hz, 1H), 4.44 (d, *J* = 7.2 Hz, 1H), 4.05 – 3.92 (m, 2H), 3.76 (td, *J* = 4.5, 1.3 Hz, 2H), 3.65 (d, *J* = 7.4 Hz, 1H), 3.46 (t, *J* = 8.3 Hz, 1H), 3.32 (s, 3H), 3.24 – 3.14 (m, 2H), 3.03 (s, 3H), 3.02 – 2.97 (m, 2H), 2.91 – 2.85 (m, 1H), 2.65 (s, 2H), 2.57 (ddd, *J* = 11.5, 6.9, 2.0 Hz, 1H), 2.35 (t, *J* = 13.3 Hz, 4H), 2.16 (d, *J* = 10.1 Hz, 1H), 1.95 – 1.85 (m, 2H), 1.85 – 1.79 (m, 1H), 1.75 – 1.64 (m, 2H), 1.62 – 1.53 (m, 2H), 1.53 – 1.46 (m, 1H), 1.44 (dd, *J* = 7.2, 3.9 Hz, 1H), 1.40 (s, 3H), 1.29 (d, *J* = 6.2 Hz, 3H), 1.24 (s, 4H), 1.20 (t, *J* = 6.7 Hz, 6H), 1.11 (td, *J* = 5.7, 5.1, 2.8 Hz, 11H), 0.84 (t, *J* = 7.3 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.3, 176.0, 103.0, 96.2, 80.8, 78.6, 78.5, 78.1, 76.7, 74.4, 72.8, 70.2, 69.2, 67.1, 65.8, 50.8, 50.1, 49.7, 45.4, 45.2, 39.4, 39.3, 37.3, 35.0, 30.2, 29.8, 26.8, 24.8, 21.6, 21.2, 20.0, 18.8, 18.2, 16.2, 16.1 (2C), 12.5, 10.8, 9.1; HRMS (ESI) calc'd for $C_{41}H_{74}NO_{13}$ + H = 788.5160, found 788.5133.

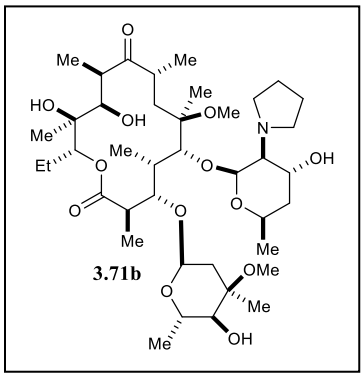


Clarithromyin analog 3.70b: $[\alpha]^{23}_{D}$ –53.2 (c 0.2, $CHCl_3$); IR (neat) 3447, 2971, 2446, 2159, 2031, 1997, 1733, 1684, 1457, 1378, 1216, 1168, 1110, 1052, 999, 908, 755, 667 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.06 (dd, *J* = 11.0, 2.3 Hz, 1H), 4.98 (d, *J* = 2.7 Hz, 1H), 4.90 (dd, *J* = 4.9, 1.3 Hz, 1H), 4.20 – 4.02 (m, 2H), 3.98 (s, 1H), 3.92 (ddd, *J* = 11.5, 6.1, 2.6 Hz, 1H), 3.81 – 3.70 (m, 2H), 3.58 (d, *J* = 7.3 Hz, 1H), 3.29 (s, 3H), 3.21 (d, *J* = 0.9 Hz, 1H), 3.08 – 2.92 (m, 4H), 2.83 (dd, *J* = 9.2, 7.3 Hz, 1H), 2.79 (d, *J* = 14.9 Hz, 2H), 2.61 (ddd, *J* = 11.3, 7.0, 2.3 Hz, 1H), 2.57 – 2.45 (m, 2H), 2.33 (dd, *J* = 15.1, 1.4 Hz, 1H), 1.92 (ddd, *J* = 14.3, 7.6, 2.3 Hz, 1H), 1.87 – 1.81 (m, 1H), 1.80 – 1.75 (m, 1H), 1.75 – 1.69 (m, 2H), 1.57 (d, *J* = 4.8 Hz, 1H), 1.55 – 1.46 (m, 6H), 1.44 (s, 3H), 1.39 (t, *J* = 6.2 Hz, 2H), 1.30 (d, *J* = 6.2 Hz, 3H), 1.22 (s, 3H), 1.19 (dd, *J* = 9.4, 6.8 Hz, 6H), 1.16 – 1.11 (m, 9H), 0.97 (d, *J* = 7.5 Hz, 3H), 0.85 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.9, 100.9, 96.4, 85.1, 79.1, 78.4, 77.7, 74.4, 73.3, 73.1, 69.3, 67.0, 66.2, 64.4, 53.6, 50.7, 49.5, 45.4, 45.0, 39.6, 39.3, 37.5, 36.6, 35.1, 32.1, 29.9, 21.7, 21.6, 21.2, 20.1, 18.5, 18.4, 16.3, 16.0, 14.3, 12.5, 10.8, 10.0.; HRMS (ESI) calc'd for $C_{41}H_{74}NO_{13}$ + H = 788.5160, found 788.5122.



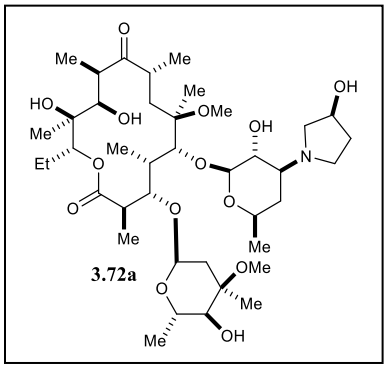
Clarithromyin analog 3.71a: $[\alpha]^{23}_{D}$ –12.9 (c 0.3, $CHCl_3$); IR (neat) 3447, 2980, 2159, 2031, 1997, 1730, 1684, 1457, 1375, 1345, 1215, 1168, 1109, 1053, 996, 751, 668 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.05 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.93 (d, *J* = 4.8 Hz, 1H), 4.45 (d, *J* = 7.3 Hz, 1H), 4.04

– 3.95 (m, 2H), 3.79 – 3.72 (m, 2H), 3.67 (d, *J* = 7.3 Hz, 1H), 3.55 – 3.44 (m, 1H), 3.33 (s, 3H), 3.20 (d, *J* = 15.9 Hz, 2H), 3.07 – 2.95 (m, 5H), 2.92 – 2.84 (m, 1H), 2.69 (s, 4H), 2.62 – 2.53 (m, 3H), 2.37 (d, *J* = 15.2 Hz, 1H), 2.19 (d, *J* = 10.3 Hz, 1H), 1.98 – 1.88 (m, 2H), 1.87 – 1.80 (m, 1H), 1.79 – 1.66 (m, 6H), 1.58 (dd, *J* = 15.2, 5.0 Hz, 1H), 1.52 – 1.44 (m, 1H), 1.41 (s, 3H), 1.30 (d, *J* = 6.1 Hz, 3H), 1.27 – 1.17 (m, 14H), 1.16 – 1.07 (m, 13H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.0, 102.9, 96.3, 80.9, 78.6, 78.1, 77.7, 76.8, 74.4, 72.8, 72.5, 69.2, 68.8, 65.9, 61.5, 50.8, 49.7, 47.8, 45.4, 45.2, 39.5, 39.3, 37.4, 35.1, 29.9, 23.7, 21.6, 21.2, 19.9, 18.9, 18.2, 16.2, 16.1, 14.3, 12.5, 10.8, 9.3; HRMS (ESI) calc'd for $C_{40}H_{72}NO_{13}$ + H = 774.5004, found 774.4977.

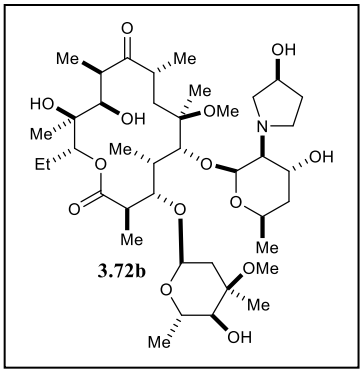


Clarithromyin analog 3.71b: $[\alpha]^{23}_D$ –56.3 (c 0.6, $CHCl_3$); IR (neat) 3447, 2971, 2446, 2364, 2159, 2031, 1997, 1734, 1700, 1684, 1457, 1378, 1345, 1169, 1054, 999, 756, 668 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.11 (d, *J* = 2.8 Hz, 1H), 5.06 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.89 (d, *J* = 4.5 Hz, 1H), 4.06 (dd, *J* = 15.7, 8.1 Hz, 2H), 3.96 (s, 1H), 3.80 – 3.72 (m, 2H), 3.64 (d, *J* = 7.2 Hz, 1H), 3.30 (s, 3H), 3.20 (s, 1H), 3.04 (s, 4H), 3.00 – 2.93 (m, 1H), 2.82 (dd, *J* = 8.9, 7.1 Hz, 1H), 2.68 – 2.55 (m, 1H), 2.32 (d, *J* = 14.9 Hz, 1H), 1.97 – 1.76 (m, 6H), 1.73 – 1.65 (m, 1H), 1.63 – 1.57 (m, 1H), 1.55 – 1.46 (m, 1H), 1.45 (s, 3H), 1.31 (d, *J* = 6.2 Hz, 3H), 1.28 – 1.17 (m, 12H), 1.17 – 1.08 (m, 9H), 0.97 (d, *J* = 7.4 Hz, 3H), 0.85 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 175.9, 96.4, 84.5, 78.5, 78.2, 77.7, 74.3, 72.9, 69.2, 67.1, 66.4, 50.8, 49.5, 45.4, 45.0, 39.8, 39.4, 37.6, 35.6, 35.0, 29.9, 23.6, 23.6,

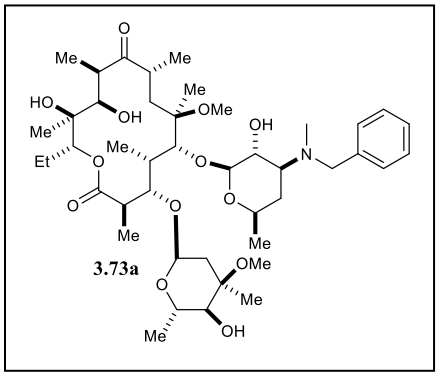
21.7, 21.2, 21.1, 21.0, 20.4, 18.6, 18.2, 16.3, 16.0, 12.4, 10.8, 10.7, 10.7, 10.1; HRMS (ESI) calc'd for $C_{40}H_{72}NO_{13}$ + H = 774.5004, found 774.4965.



Clarithromyin analog 3.72a: $[\alpha]^{23}_{D}$ -50.0 (c 0.3, $CHCl_3$); IR (neat) 3447, 2971, 2159, 2031, 1717, 1653, 1457, 1378, 1215, 1168, 1052, 1009, 999, 752, 668 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 5.05 (dd, *J* = 11.2, 2.2 Hz, 1H), 4.92 (d, *J* = 4.8 Hz, 1H), 4.45 (d, *J* = 7.3 Hz, 1H), 4.37 (s, 1H), 3.99 (d, *J* = 8.1 Hz, 2H), 3.76 (t, *J* = 4.7 Hz, 2H), 3.67 (d, *J* = 7.1 Hz, 1H), 3.52 (dd, *J* = 9.7, 5.0 Hz, 1H), 3.32 (s, 4H), 3.19 (s, 1H), 3.08 – 2.95 (m, 5H), 2.92 – 2.68 (m, 3H), 2.58 (dd, *J* = 11.6, 7.0 Hz, 1H), 2.36 (d, *J* = 15.1 Hz, 1H), 2.25 – 2.07 (m, 2H), 1.99 – 1.86 (m, 1H), 1.80 (q, *J* = 13.3, 12.9 Hz, 2H), 1.68 (dd, *J* = 14.9, 2.1 Hz, 1H), 1.59 (dd, *J* = 15.1, 5.0 Hz, 1H), 1.53 – 1.43 (m, 1H), 1.40 (s, 3H), 1.30 (d, *J* = 6.1 Hz, 3H), 1.27 – 1.18 (m, 10H), 1.15 – 1.10 (m, 9H), 1.07 (d, *J* = 7.4 Hz, 3H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 221.2, 175.9, 102.7, 96.2, 81.1, 78.5, 78.5, 78.0, 77.8, 74.4, 72.9, 72.6, 70.9, 69.2, 68.6, 66.0, 61.3, 57.1, 50.8, 49.6, 47.4, 45.4, 45.2, 39.4, 39.2, 37.4, 35.0, 34.7, 31.2, 29.9, 21.6, 21.5, 21.2, 19.9, 18.9, 18.2, 16.1, 12.5, 10.7, 9.4; HRMS (ESI) calc'd for $C_{40}H_{72}NO_{14}$ + H = 790.4953, found 790.4978.

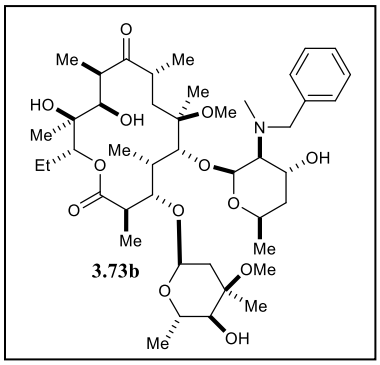


Clarithromyin analog 3.72b: $[\alpha]^{23}_{D}$ –67.8 (c 0.5, $CHCl_3$); IR (neat) 3447, 2971, 2441, 2159, 2031, 1997, 1734, 1700, 1684, 1457, 1374, 1216, 1168, 1053, 1002, 758, 666 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 5.05 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.92 (d, *J* = 4.7 Hz, 1H), 4.46 (d, *J* = 7.1 Hz, 1H), 4.39 (d, *J* = 6.6 Hz, 1H), 3.98 (d, *J* = 4.8 Hz, 2H), 3.75 (t, *J* = 4.8 Hz, 2H), 3.67 (d, *J* = 7.0 Hz, 1H), 3.52 (dt, *J* = 7.1, 3.8 Hz, 1H), 3.32 (s, 4H), 3.19 (s, 1H), 3.03 (s, 4H), 2.96 – 2.76 (m, 2H), 2.65 – 2.51 (m, 1H), 2.34 (d, *J* = 15.1 Hz, 1H), 2.18 (dt, *J* = 14.2, 7.5 Hz, 1H), 1.99 – 1.86 (m, 2H), 1.86 – 1.76 (m, 2H), 1.66 (dd, *J* = 14.7, 2.1 Hz, 1H), 1.59 (dd, *J* = 15.1, 4.9 Hz, 1H), 1.46 (dt, *J* = 10.9, 7.0 Hz, 1H), 1.39 (s, 3H), 1.29 (d, *J* = 6.1 Hz, 3H), 1.27 – 1.18 (m, 14H), 1.13 (q, *J* = 3.2 Hz, 9H), 1.06 (d, *J* = 7.5 Hz, 3H), 0.86 (dt, *J* = 14.7, 6.8 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 221.2, 175.9, 102.6, 96.2, 81.2, 78.6, 78.5, 78.0, 74.4, 72.9, 72.5, 70.6, 69.2, 68.4, 66.1, 61.8, 57.6, 50.8, 49.6, 45.3, 39.4, 39.1, 37.4, 35.1, 34.5, 32.1, 29.8, 29.5, 22.8, 21.6, 21.4, 21.2, 19.9, 18.8, 18.1, 16.1, 14.3, 12.5, 10.8, 9.5; HRMS (ESI) calc'd for $C_{40}H_{72}NO_{14}$ + H = 790.4953, found 790.4977.



Clarithromyin analog 3.73a: $[\alpha]^{23}_{D}$ –58.2 (c 0.8, $CHCl_3$); IR (neat) 3442, 2977, 2446, 2364, 2159, 2031, 1977, 1730, 1664, 1457, 1374, 1345, 1215, 1169, 1107, 1074, 1053, 995, 752, 668 cm^{-1}; ^1H NMR (500 MHz $CDCl_3$) δ 7.35 – 7.26 (m, 5H).δ 5.05 (dd, *J* = 11.1, 2.3 Hz, 1H), 4.91 (d, *J* = 4.9 Hz, 1H), 4.41 (d, *J* = 7.1 Hz, 1H), 4.00 – 3.89 (m, 2H), 3.79 – 3.70 (m, 3H), 3.62 (d, *J* = 7.2 Hz, 1H), 3.54 (s, 1H), 3.48 – 3.37 (m, 2H), 3.29 (dd, *J* = 10.2, 7.1 Hz, 1H), 3.18 (s, 1H), 3.12 (s, 3H), 3.05 – 2.94 (m,

5H), 2.91 – 2.82 (m, 1H), 2.54 (dddd, *J* = 31.9, 16.0, 8.6, 2.8 Hz, 2H), 2.32 (d, *J* = 15.3 Hz, 1H), 2.24 (s, 3H), 2.05 (d, *J* = 10.2 Hz, 1H), 1.95 – 1.80 (m, 3H), 1.78 – 1.68 (m, 2H), 1.55 (dd, *J* = 15.2, 5.1 Hz, 1H), 1.51 – 1.44 (m, 1H), 1.40 (s, 3H), 1.28 (d, *J* = 6.3 Hz, 3H), 1.26 – 1.22 (m, 4H), 1.19 (d, *J* = 15.5 Hz, 6H), 1.14 – 1.10 (m, 9H), 1.07 (d, *J* = 7.5 Hz, 3H), 0.84 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 221.2, 175.9, 138.9, 129.0, 128.6, 127.5, 102.9, 96.1, 81.1, 78.5, 78.4, 78.1, 76.7, 74.4, 72.7, 70.9, 69.2, 68.8, 65.8, 63.8, 58.1, 50.8, 49.6, 45.4, 45.2, 39.4, 39.3, 37.4, 37.0, 35.0, 29.4, 21.6, 21.6, 21.2, 20.0, 18.8, 18.2, 16.2, 16.1, 12.5, 10.8, 9.2; HRMS (ESI) calc'd for $C_{44}H_{74}NO_{13}$ + H = 824.5160, found 824.5153.



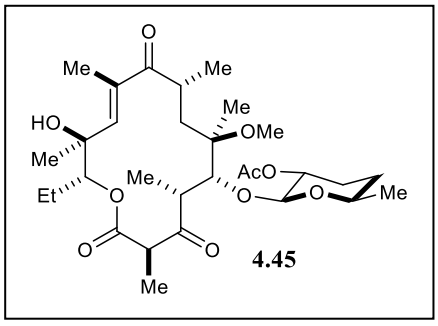
3.73b

Clarithromyin analog 3.73b: $[\alpha]^{23}_D$ –87.0 (c 0.7, $CHCl_3$); IR (neat) 3462, 2971, 2936, 2446, 2368, 2159, 2031, 1977, 1734, 1696, 1457, 1378, 1286, 1168, 1054, 755, 668 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.29 (m, 5H), 5.13 – 4.99 (m, 2H), 4.91 (dd, *J* = 4.9, 1.4 Hz, 1H), 4.20 – 4.05 (m, 3H), 3.98 (s, 2H), 3.82 – 3.74 (m, 2H), 3.64 (d, *J* = 7.2 Hz, 1H), 3.28 (s, 3H), 3.22 (d, *J* = 0.9 Hz, 1H), 3.05 (s, 3H), 3.04 – 2.99 (m, 2H), 2.88 – 2.81 (m, 1H), 2.61 (ddd, *J* = 11.5, 6.8, 2.1 Hz, 1H), 2.53 (d, *J* = 9.9 Hz, 1H), 2.45 (s, 3H), 2.34 (dd, *J* = 15.2, 1.4 Hz, 1H), 1.93 (ddd, *J* = 14.2, 7.6, 2.3 Hz, 1H), 1.90 – 1.78 (m, 3H), 1.78 – 1.71 (m, 1H), 1.57 (dt, *J* = 15.0, 5.6 Hz, 2H), 1.53 – 1.47 (m, 1H), 1.45 (s, 3H), 1.31 (d, *J* = 6.3 Hz, 3H), 1.23 (s, 3H), 1.20 (dd, *J* = 6.8, 3.2 Hz, 6H), 1.17 (s, 3H), 1.13 (t, *J* = 6.5 Hz, 6H), 1.00 (d, *J* = 7.4 Hz, 3H), 0.85 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.0, 141.3, 128.4, 126.9, 100.6, 96.4, 83.4, 78.6, 78.5, 77.9, 76.8, 74.4, 73.0, 70.2, 69.2, 66.5, 66.0, 63.7, 60.8, 50.8, 49.5, 45.5, 45.2, 41.2, 39.8, 39.5, 37.5, 36.7, 35.0, 29.9, 21.8, 21.6, 21.2, 20.3, 18.6, 18.3,

16.3, 16.1, 12.4, 10.8, 9.7; HRMS (ESI) calc'd for $C_{44}H_{74}NO_{13}$ + H = 824.5160, found 824.5160.

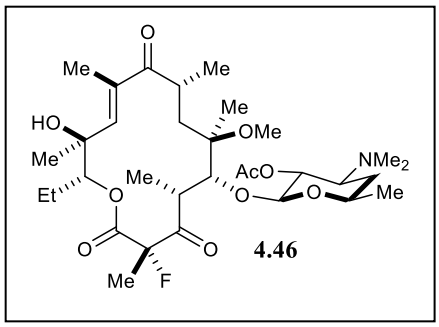
5.3 Chapter 4: THE DEVELOPMENT OF LEAD COMPOUNDS FOR RIBOSOME-TEMPLATED *IN SITU* CLICK CHEMISTRY

5.3.1 Synthesis of (*E*)-dehydro solithromycin analogs



Enone Ketone 4.45: Dess-Martin periodinane (1.23 g, 2.91 mmol) and $NaHCO_3$ (650 mg, 7.76 mmol) were added to a solution of **4.44** (1.025.6 g, 1.94 mmol) in CH_2Cl_2 (20 mL). The solution was stirred at rt for 2 h. Sat'd aq. $NaHCO_3$ (10 mL), sat'd aq. Na_2SO_3 (10 mL) and H_2O (20 mL) were added to the reaction vessel and stirred an additional 30 min before extracting with CH_2Cl_2 (3 x 20 mL). The combined organic layers were washed with brine (50 mL) and filtered over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was directly used to the next step. A solution of C3 ketone compound, acetic anhydride (396.1 mg, 3.88 mmol), and triethylamine (392.6 mg, 3.88 mmol) in CH_2Cl_2 (10 ml) was stirred at room temperature for 12h. Then the organic layer was washed with 100ml portions of saturated aqueous $NaHCO_3$, H_2O, and brine and filtered over Na_2SO_4. The solvent was removed under reduced pressure, purified by flash column chromatography eluting with 0–5% MeOH/CH_2Cl_2 to give 711.72 mg of **4.45** (65%) as a yellow foam. $[\alpha]^{23}_D$ +57.3 (c 1.24, $CHCl_3$); IR (neat) 3019, 2160, 2027, 1742, 1710, 1456, 1374, 1215, 1163, 1106, 1060, 998, 749, 837, 668 cm^{-1}; ^1H NMR (500 MHz $CDCl_3$) δ 6.57 (s, 1H), 4.97 (dd, *J* = 9.9, 2.9 Hz, 1H), 4.70 (dd, *J* = 10.5, 7.6 Hz, 1H), 4.32 (d, *J* = 7.6 Hz, 1H), 4.10 (d, *J* = 8.3 Hz, 1H), 3.71 (q, *J* = 6.9 Hz, 1H), 3.50 (ddt, *J* = 12.4, 6.4, 3.1 Hz, 1H), 3.19 – 3.08 (m, 1H), 3.06 – 2.98 (m, 1H), 2.83 (s, 3H), 2.69 – 2.57 (m, 1H), 2.26 (s, 1H), 2.22 (s, 6H), 2.02 (s, 3H), 1.99 (d, *J* = 1.3 Hz, 3H), 1.96 – 1.90 (m, 1H), 1.81 (dd, *J* = 14.4,

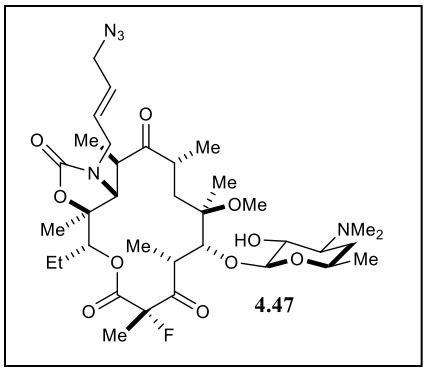
6.9 Hz, 1H), 1.71 (ddd, *J* = 13.0, 4.5, 1.9 Hz, 1H), 1.60 – 1.49 (m, 1H), 1.45 (s, 3H), 1.33 (d, *J* = 6.9 Hz, 3H), 1.30 – 1.24 (m, 3H), 1.21 (d, *J* = 6.2 Hz, 3H), 1.11 (dd, *J* = 13.1, 7.1 Hz, 6H), 0.90 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 207.2, 169.9, 142.3, 138.9, 101.9, 83.4, 81.4, 78.8, 78.4, 75.3, 73.4, 71.6, 69.2, 63.6, 56.1, 51.3, 50.5, 47.2, 40.7, 40.2, 38.5, 30.5, 22.5, 22.1, 21.5, 21.1, 19.0, 14.9, 14.2, 14.1, 13.6, 11.0; HRMS (ESI) calc'd for $C_{32}H_{53}NO_{10}$ + Na = 634.3567, found 634.3573.



4.46

2-Fluoroketolide 4.46. *t*-BuOK (4.44 mL of 1 M in THF, 4.44 mmol) was added dropwise to a solution of **4.45** (2.262 g, 3.7 mmol) in THF (37 mL) at 0 °C under an inert atmosphere and stirred for 30 minutes. The solution was subsequently cooled to -78 °C. NFSI (1.283 g, 4.07 mmol) in THF (10 mL) was cannulated into the solution, and the reaction mixture was stirred at -78 °C for 2 h. Sat'd aq. NH_4Cl (10 mL) was added and the mixture extracted with EtOAc (3 x 20 mL). The combined organic layers were washed with brine (20 mL) and dried over sodium sulfate. The solvent was removed under reduced pressure, and the residue was purified by flash column chromatography eluting with 0–5% MeOH/CH_2Cl_2 to give 1.47 g (63%) of **4.46** as a yellow foam. $[\alpha]^{23}_D$ +33.1 (c 0.98, $CHCl_3$); IR (neat) 2974, 2160, 2031, 1742, 1669, 1456, 1374, 1215, 1105, 1060, 751, 668 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.39 (d, *J* = 1.7 Hz, 1H), 4.95 (dd, *J* = 9.6, 3.1 Hz, 1H), 4.65 (dd, *J* = 10.6, 7.4 Hz, 1H), 4.29 (d, *J* = 7.4 Hz, 1H), 3.89 (dd, *J* = 9.9, 1.6 Hz, 1H), 3.41 (dddd, *J* = 20.3, 10.1, 7.3, 4.7 Hz, 2H), 2.88 (s, 1H), 2.68 (s, 1H), 2.64 – 2.58 (m, 1H), 2.56 (d, *J* = 6.9 Hz, 4H), 2.16 (s, 8H), 2.00 (s, 4H), 1.93 (ddd, *J* = 14.4, 7.5, 3.0 Hz, 1H), 1.89 –

1.85 (m, 4H), 1.76 (dd, *J* = 14.1, 9.7 Hz, 1H), 1.71 – 1.60 (m, 6H), 1.55 (ddt, *J* = 14.5, 9.7, 7.2 Hz, 1H), 1.42 (s, 4H), 1.23 (td, *J* = 12.7, 11.0 Hz, 1H), 1.14 (dd, *J* = 12.6, 6.3 Hz, 10H), 1.08 (d, *J* = 6.9 Hz, 3H), 0.87 (t, *J* = 7.3 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 207.2, 203.2, 169.8, 166.3, 166.1, 142.2, 138.3, 101.7, 98.9, 97.2, 81.3, 80.2, 78.9, 73.1, 71.6, 69.1, 63.3, 49.2, 40.6 (2C), 30.6, 24.5, 24.3, 23.1, 22.4, 21.4, 21.0, 19.8, 19.6, 14.4, 13.5, 10.9; HRMS (ESI) calc'd for $C_{32}H_{52}FNO_{10}$ + H = 630.3654, found 630.3635.



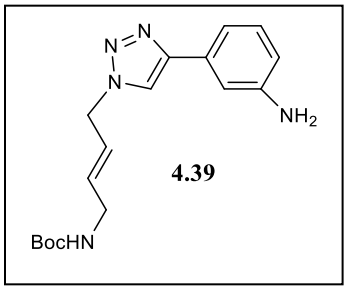
4.47

Azide 4.47: 60% NaH in oil (38 mg, 0.95 mmol) was added to a solution of **4.46** (150 mg, 0.24 mmol) and CDI (103.5 mg, 0.64 mmol) in DMF/THF (2.38 mL, 10:1) at -20 °C. The solution was stirred for 45 min while warming to 0 °C. Sat'd aq. $NaHCO_3$ (2 mL) was added dropwise, and the reaction mixture was extracted with EtOAc (3 × 5 mL). The combined organic layers were washed with brine (20 mL) and dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified by flash column chromatography eluting with 0−5% MeOH in CH_2Cl_2 to afford an acyl imidazole intermediate. After dissolving the acyl imidazole in MeCN/H_2O (2 mL, 9:1), amine **4.41** (85.2 mg, 0.76 mmol) was added, and the reaction mixture stirred at rt for 72 h. The solvent was removed under reduced pressure, and the residue was purified by flash column chromatography eluting with 0−5% MeOH/CH_2Cl_2 to give 67 mg (61%) of **4.47** as a yellow foam. [□]$^{23}_D$ +7.7 (c 1.12, solvent); IR (neat) 2972, 2940, 2444, 2360, 2160, 2101, 2033, 1978, 1757, 1710, 1457, 1379, 1261, 1223, 1162, 1108, 1052, 1003, 979, 756, 668 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.83 – 5.77 (m, 2H), 5.43 – 5.34 (m, 1H),

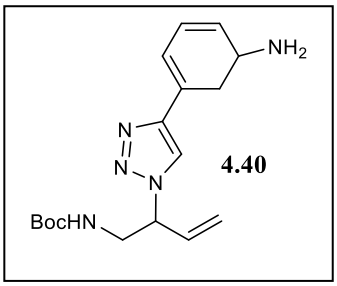
5.12 (dt, *J* = 10.6, 3.4 Hz, 1H), 4.88 (dd, *J* = 10.5, 2.6 Hz, 1H), 4.39 – 4.34 (m, 1H), 4.31 (dd, *J* = 7.2, 4.0 Hz, 2H), 4.25 – 4.18 (m, 1H), 4.07 (ddd, *J* = 16.2, 10.2, 3.2 Hz, 2H), 3.85 – 3.67 (m, 4H), 3.63 – 3.49 (m, 4H), 3.44 (s, 1H), 3.18 (dd, *J* = 10.2, 7.2 Hz, 2H), 3.10 (q, *J* = 6.9 Hz, 2H), 2.67 – 2.57 (m, 4H), 2.53 (s, 3H), 2.48 – 2.41 (m, 2H), 2.27 (s, 10H), 1.98 (ddtt, *J* = 18.2, 10.7, 7.6, 4.3 Hz, 2H), 1.88 (dd, *J* = 14.5, 2.8 Hz, 2H), 1.79 (dd, *J* = 21.4, 6.5 Hz, 5H), 1.71 – 1.59 (m, 9H), 1.55 (d, *J* = 12.8 Hz, 1H), 1.50 (d, *J* = 3.8 Hz, 5H), 1.39 – 1.15 (m, 26H), 1.00 (dd, *J* = 16.2, 6.9 Hz, 5H), 0.88 (td, *J* = 7.5, 4.6 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.9, 203.2, 166.3, 157.1, 133.9, 129.7, 127.1, 121.1, 104.4, 98.7, 82.4, 80.9, 78.7, 78.6, 70.5, 69.8, 66.0, 62.4, 61.4, 52.3, 49.5, 44.8, 40.4, 39.8, 39.4, 28.3, 25.3, 22.3, 21.3, 19.8, 18.0, 15.2, 14.8, 14.1, 10.5; HRMS (ESI) calc'd for $C_{35}H_{56}FN_5O_{10}$ + H = 726.4084, found 726.4056.

Experimental Procedure for Copper(I)-catalyzed Click Reactions:

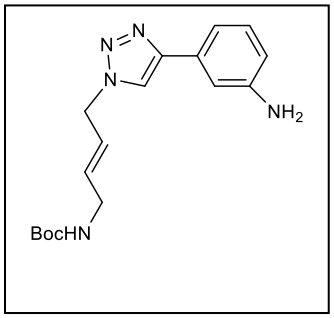
Azide **4.28 or 4.47** (1 equiv), $CuSO_4$ (0.02 equiv), (+)-sodium L-ascorbate (0.1 equiv), and alkyne **4.6, 4.49-4.50** (2 equiv) in 1:1 H_2O:*t*-BuOH (0.05 M) was stirred at rt for 24 hours. H_2O (2 mL) was added and the mixture extracted with EtOAc (3 × 5 mL). The combined organic layers were washed with brine (5 mL), dried over Na_2SO_4, and the solvent removed under reduced pressure. The residue was purified by flash column chromatography with a Combiflash instrument (MeOH/DCM (1% NH^4OH) 0–10%) to give triazoles **4.39-4.40** and **4.51-4.53** (70-90% yield).



(*E*)-alkene 4.39 : IR (neat) 2954, 2939, 2876, 2160, 2031, 1700, 1463, 1366, 1250, 1156, 1077, 835, 739 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.69 (s, 1H), 7.23 (dd, *J* = 2.4, 1.6 Hz, 1H), 7.18 (t, *J* = 7.7 Hz, 1H), 7.12 (dt, *J* = 7.6, 1.2 Hz, 1H), 6.64 (ddd, *J* = 7.9, 2.4, 1.1 Hz, 1H), 5.80 (d, *J* = 4.6 Hz, 1H), 4.95 (dt, *J* = 5.5, 1.3 Hz, 2H), 3.78 (t, *J* = 4.7 Hz, 5H), 1.43 (s, 9H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 155.8, 148.2, 147.0, 133.3, 131.6, 129.9, 124.3, 119.6, 116.2, 115.0, 112.3, 79.8, 51.7, 41.7, 28.5; HRMS (ESI) calc'd for $C_{17}H_{23}N_5O_2$ + H = 330.1930, found 330.1923.

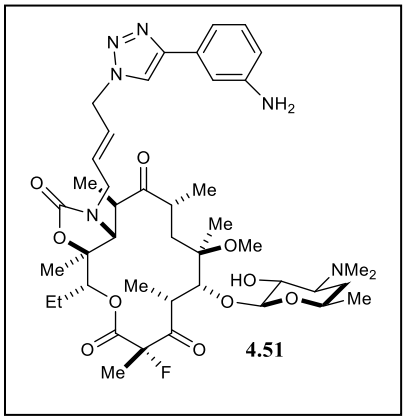


Terminal alkene 4.40: IR (neat) 2954, 2939, 2876, 2160, 2031, 1700, 1463, 1366, 1250, 1156, 1077, 835, 739 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.72 (s, 1H), 7.22 (t, *J* = 1.9 Hz, 1H), 7.20 – 7.09 (m, 2H), 6.68 – 6.60 (m, 1H), 6.04 (ddd, *J* = 17.2, 10.4, 6.8 Hz, 1H), 5.35 (d, *J* = 10.4 Hz, 1H), 5.25 (d, *J* = 17.2 Hz, 1H), 5.21 – 5.09 (m, 2H), 3.78 (dddd, *J* = 20.2, 14.4, 10.7, 5.1 Hz, 4H), 1.39 (s, 9H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 156.0, 147.8, 147.1, 133.5, 131.4, 129.9, 119.9, 116.0, 115.1, 112.2, 80.0, 63.5, 43.9, 29.8, 28.4; HRMS (ESI) calc'd for $C_{17}H_{23}N_5O_2$ + H = 330.1930, found 330.1933.

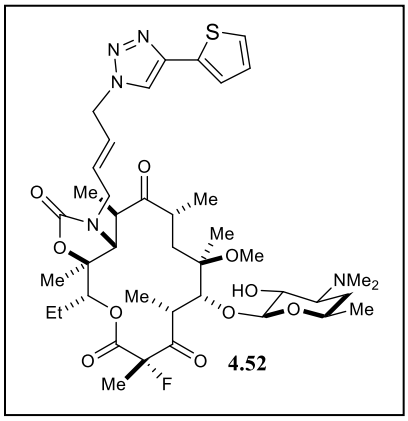


(*E*)-alkene 4.39 : IR (neat) 2954, 2939, 2876, 2160, 2031, 1700, 1463, 1366, 1250, 1156, 1077, 835, 739 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.69 (s, 1H), 7.23 (dd, *J* = 2.4, 1.6 Hz, 1H), 7.18 (t, *J* = 7.7 Hz, 1H), 7.12 (dt, *J* = 7.6, 1.2 Hz, 1H), 6.64 (ddd, *J* = 7.9, 2.4, 1.1 Hz, 1H), 5.80 (d, *J* = 4.6 Hz, 1H), 4.95

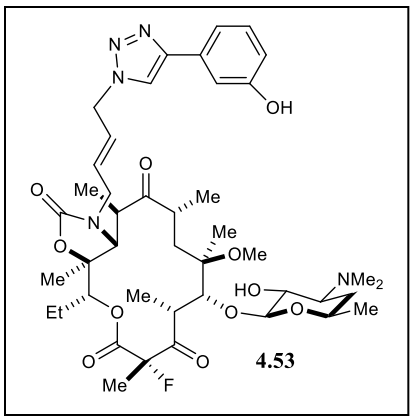
(dt, *J* = 5.5, 1.3 Hz, 2H), 3.78 (t, *J* = 4.7 Hz, 5H), 1.43 (s, 9H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 155.8, 148.2, 147.0, 133.3, 131.6, 129.9, 124.3, 119.6, 116.2, 115.0, 112.3, 79.8, 51.7, 41.7, 28.5; HRMS (ESI) calc'd for $C_{17}H_{23}N_5O_2$ + H = 330.1930, found 330.1923.



Triazole 4.51: $[\alpha]^{23}_D$ + 13.5 (c 1.12, solvent); IR (neat) 2980, 2940, 2360, 2341, 1750, 1710, 1669, 1603, 1457, 1379, 1261, 1162, 1108, 1052, 1003, 979, 756, 668 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.85 (s, 1H), 7.31 (t, *J* = 1.9 Hz, 1H), 7.20 (dd, *J* = 16.0, 8.3 Hz, 2H), 6.64 (d, *J* = 7.5 Hz, 1H), 5.92 (q, *J* = 5.5 Hz, 2H), 4.98 (t, *J* = 4.3 Hz, 2H), 4.89 (d, *J* = 10.7 Hz, 1H), 4.39 (dd, *J* = 15.8, 5.1 Hz, 1H), 4.31 (d, *J* = 7.3 Hz, 1H), 4.26 – 4.18 (m, 1H), 4.04 (d, *J* = 10.5 Hz, 1H), 3.54 (t, *J* = 8.5 Hz, 3H), 3.45 (s, 1H), 3.23 (dd, *J* = 10.2, 7.4 Hz, 1H), 3.10 (q, *J* = 7.0 Hz, 1H), 2.61 (d, *J* = 14.1 Hz, 3H), 2.51 (s, 3H), 2.34 (s, 7H), 2.17 (s, 2H), 1.96 (dd, *J* = 14.4, 7.4 Hz, 1H), 1.91 – 1.85 (m, 1H), 1.79 (d, *J* = 21.4 Hz, 3H), 1.71 (d, *J* = 13.3 Hz, 1H), 1.67 – 1.56 (m, 1H), 1.51 (s, 4H), 1.37 – 1.11 (m, 20H), 1.02 (d, *J* = 6.9 Hz, 4H), 0.84 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 217.0, 203.4, 203.1, 157.1, 148.3, 147.0, 131.8, 131.4, 129.8, 126.3, 119.7, 116.4, 114.9, 112.5, 104.4, 82.6, 81.0, 78.7, 78.5, 70.5, 69.8, 66.0, 61.5, 52.0, 49.4, 44.9, 44.8, 41.0, 40.4, 39.8, 39.4, 29.9, 28.5, 28.3, 25.5, 25.3, 22.2, 21.3, 19.9, 18.0, 15.2, 14.8, 14.1, 10.6; HRMS (ESI) calc'd for $C_{43}H_{63}FN_6O_{10}$ + H = 843.4668, found 843.4648.



Triazole 4.52: $[\alpha]^{23}_{D}$ +9.0 (c 0.32)); IR (neat) 3301, 2443, 2160, 2031, 1978, 1757, 1669, 1603, 1534, 1487, 1392, 1370, 1313, 1260, 1172, 1072, 1006, 822, 746, 692 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.81 (s, 1H), 7.44 (dd, *J* = 3.6, 1.2 Hz, 1H), 7.28 (dd, *J* = 5.1, 1.2 Hz, 1H), 7.06 (dd, *J* = 5.1, 3.6 Hz, 1H), 5.97 – 5.91 (m, 1H), 4.97 (t, *J* = 5.3 Hz, 2H), 4.88 (dd, *J* = 10.4, 2.3 Hz, 1H), 4.38 (dd, *J* = 15.8, 5.8 Hz, 1H), 4.30 (d, *J* = 7.3 Hz, 1H), 4.22 (dd, *J* = 15.8, 4.9 Hz, 1H), 4.06 (dd, *J* = 10.7, 1.7 Hz, 1H), 3.53 (dt, *J* = 14.9, 5.7 Hz, 3H), 3.45 (s, 1H), 3.18 (dd, *J* = 10.2, 7.3 Hz, 1H), 3.10 (q, *J* = 6.9 Hz, 1H), 2.66 – 2.57 (m, 0H), 2.52 (s, 3H), 2.48 – 2.41 (m, 1H), 2.27 (s, 6H), 2.25 (s, 1H), 1.97 (ddd, *J* = 14.6, 7.6, 2.5 Hz, 1H), 1.89 (dd, *J* = 14.5, 3.0 Hz, 1H), 1.78 (d, *J* = 21.4 Hz, 4H), 1.70 – 1.59 (m, 2H), 1.55 (d, *J* = 13.3 Hz, 1H), 1.51 (s, 4H), 1.34 (s, 3H), 1.31 (d, *J* = 7.1 Hz, 4H), 1.29 – 1.21 (m, 8H), 1.21 – 1.15 (m, 4H), 1.02 (d, *J* = 6.9 Hz, 4H), 0.85 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 217.0, 203.0, 157.1, 143.3, 133.3, 131.6, 127.6, 126.4, 125.0, 124.4, 119.2, 104.4, 82.6, 80.9, 78.8, 78.5, 70.5, 69.8, 66.0, 61.6, 52.0, 49.5, 45.0, 44.8, 41.0, 40.8, 40.4, 39.8, 39.4, 29.9, 28.3, 25.5, 25.3, 22.3, 21.3, 19.9, 18.0, 15.1, 14.8, 14.1, 10.6; HRMS (ESI) calc'd for $C_{41}H_{60}FSN_5O_{10}$ + H = 834.4123, found 835.4141.

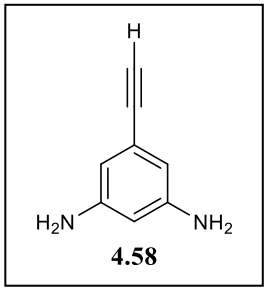


Triazole 4.53 : $[\alpha]^{23}_{D}$ +51.2 (c 0.16, solvent); IR (neat) 2932, 2442, 2362, 2160, 2033, 1989, 1757, 1457, 1275,1261, 1051, 1003, 764, 750, 668 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.88 (s, 1H), 7.61 (dt, *J* = 7.9, 1.2 Hz, 1H), 7.40 (dd, *J* = 2.5, 1.5 Hz, 1H), 7.37 – 7.28 (m, 1H), 6.87 (ddd, *J* = 8.2, 2.4, 1.0 Hz, 1H), 5.99 (dt, *J* = 15.7, 5.8 Hz, 1H), 5.91 – 5.82 (m, 1H), 5.18 – 5.08 (m, 1H), 5.00 – 4.86 (m, 2H), 4.53 (dd, *J* = 16.7, 7.0 Hz, 1H), 4.33 – 4.19 (m, 2H), 4.01 (dd, *J* = 10.7, 1.7 Hz, 1H), 3.67 – 3.49 (m, 1H), 3.48 (s, 1H), 3.22 – 3.07 (m, 2H), 2.66 – 2.59 (m, 1H), 2.54 (s, 3H), 2.50 – 2.40 (m, 1H), 2.27 (s, 7H), 2.00 (ddt, *J* = 15.6, 8.1, 4.1 Hz, 1H), 1.96 – 1.88 (m, 1H), 1.85 (s, 2H), 1.81 (s, 2H), 1.72 – 1.61 (m, 1H), 1.53 (s, 4H), 1.37 – 1.16 (m, 21H), 1.04 (d, *J* = 7.0 Hz, 3H), 0.87 (dt, *J* = 12.7, 7.0 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 166.4, 157.1, 156.7, 148.2, 132.1, 130.5, 124.9, 119.5, 118.1, 115.3, 112.9, 104.5, 82.6, 81.1, 79.0, 78.9, 77.7, 76.3, 70.5, 69.9, 66.0, 61.9, 52.1, 49.2, 44.8, 44.5, 41.4, 40.4, 39.9, 39.5, 29.9, 28.3, 25.5, 25.3, 22.2, 21.3, 19.8, 18.0, 15.4, 14.8, 14.2, 10.6; HRMS (ESI) calc'd for $C_{43}H_{62}FN_5O_{11}$ + H = 844.4508, found 844.4531.

5.3.2 Synthesis of N11-Functionalized Solithromycin Analogs that Engage in Hydrogen bonding and π-stacking

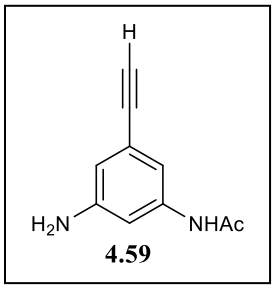
Experimental Procedure for Alkyne Synthesis by Sonogashira Coupling/TMS Deprotection:

CuI (0.04 equiv), $Pd(PPh_3)_2Cl_2$ (0.02 equiv), and aryl iodide precursor were were dissolved in anhydrous Et_3N. Trimethylsilyl acetylene (1.5 equiv) was added dropwise at 0 °C. The solution was allowed to gradually warm to rt. After 12 hours, Et_3N was removed via reduced pressure. The crude mixture was filtered over Celite and washed successively with CH_2Cl_2. The combined organic layers were washed with brine and dried over anhydrous Na_2SO_4. The solvent removed under reduced pressure, and the crude product was dried under vacuum before use in the subsequent step. To a solution of TMS-alkyne (1equiv) in MeOH/THF (1:1, 0.25 M) was added K_2CO_3 (0.5 equiv) and the mixture was stirred for 3–12 hours at rt under N_2. The reaction mixture was concentrated *in vacuo*, diluted with CH_2Cl_2, washed with H_2O, brine and dried over Na_2SO_4. The filtrate was concentrated under reduced pressure, and the residue was purified by flash column chromatography eluting with acetone/hexanex 0–40% to give alkynes **4.58, 59, 60, 62, 63** and **66** (70-90% yield).



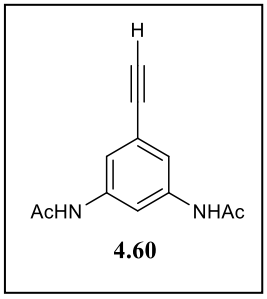
4.58

5-ethynylbenzene-1,3-diamine (4.58): IR (neat) 3421, 3310, 3281, 3207, 2162, 2105, 1625, 1593, 1487, 1353, 1192, 992, 869, 832, 711 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.25 (d, *J* = 2.1 Hz, 2H), 5.99 (t, *J* = 2.1 Hz, 1H), 3.59 (s, 4H), 2.96 (s, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 147.5, 123.5, 109.7, 102.7, 84.3, 76.0; HRMS (ESI) calc'd for $C_8H_8N_2$ + H = 133.0760, found 133.0759.



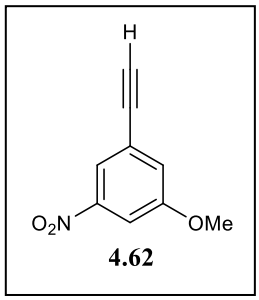
4.59

N-(3-amino-5-ethynylphenyl)acetamide (**4.59**): IR (neat) 3299, 1674, 1592, 1538, 1423, 1264, 896, 848, 732, 703 cm^{-1}; ^{1}H NMR (400 MHz, MeOD) δ 7.00 (t, *J* = 2.0 Hz, 1H), 6.92 (t, *J* = 1.7 Hz, 1H), 6.54 (dd, *J* = 2.2, 1.4 Hz, 1H), 3.32 (s, 1H), 2.07 (s, 3H); ^{13}C NMR (101 MHz, MeOH) δ 171.6, 149.7, 140.7, 124.4, 115.3, 114.1, 108.5, 84.8, 77.4, 23.9; HRMS (ESI) calc'd for $C_{10}H_{10}N_2O$ + H = 175.0866, found 175.0863.



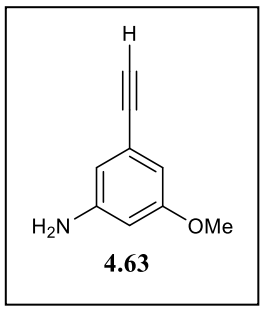
4.60

N,N'-(5-ethynyl-1,3-phenylene)diacetamide (**4.60**): IR (neat) 3294, 3090, 2924, 2406, 1647, 1584, 1462, 1383, 1275, 1177, 1088, 980, 882, 641 cm^{-1}; ^{1}H NMR (400 MHz, MeOD) δ 7.83 (t, *J* = 2.0 Hz, 1H), 7.42 (d, *J* = 2.0 Hz, 2H), 3.45 (s, 1H), 2.10 (s, 6H); ^{13}C NMR (126 MHz, MeOD) δ 171.8, 140.5, 124.3, 119.8, 113.00, 84.0, 78.6, 23.9; HRMS (ESI) calc'd for $C_{12}H_{12}N_2O_2$ + Na = 239.0796, found 239.0797.



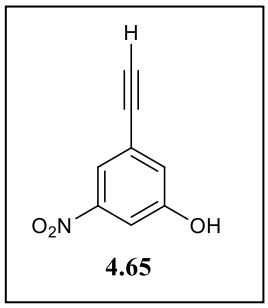
4.62

1-ethynyl-3-methoxy-5-nitrobenzene (**4.62**): IR (neat) 3293, 2979, 2159, 2030, 1976, 1724, 1572, 1529, 1373, 1356, 1014, 996, 868, 774, 692 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.93 (dd, *J* = 2.0, 1.2 Hz, 1H), 7.72 (t, *J* = 2.3 Hz, 1H), 7.30 (dd, *J* = 2.5, 1.3 Hz, 1H), 3.89 (d, *J* = 0.8 Hz, 4H), 3.19 (s, 1H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 160.1,149.2, 124.5, 124.0, 119.5, 109.1, 81.3, 79.7, 56.2; HRMS (ESI) calc'd for $C_9H_7NO_3$ + H = 178.0499, found 178.0504.



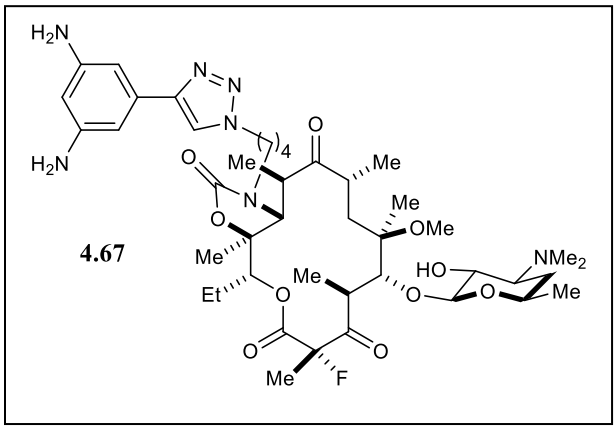
4.63

3-ethynyl-5-methoxy-aniline (4.**63**): IR (neat) 3450, 3371, 3286, 2104, 1620, 1588, 1434, 1199, 1168, 1051, 933, 837, 680 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.45 (ddd, *J* = 8.0, 2.2, 1.3 Hz, 2H), 6.23 (t, *J* = 2.2 Hz, 1H), 3.75 (s, 4H), 3.00 (s, 1H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 160.6, 147.7, 123.5, 111.8, 107.6, 102.5, 84.0, 76.4, 55.4; HRMS (ESI) calc'd for $C_9H_7NO_3$ + H = 178.0499, found 178.0493.



4.65

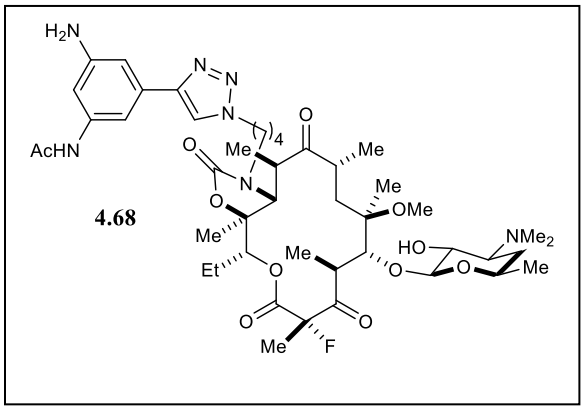
3-ethynyl-5-nitrophenol (**4.65**): IR (neat) 3425, 3301, 3091, 2981, 2423, 1616, 1515, 1482, 1284, 1155, 1091, 871, 781, 669 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.92 (t, *J* = 1.7 Hz, 1H), 7.68 (t, *J* = 2.2 Hz, 1H), 7.26 (d, *J* = 2.8 Hz, 2H), 5.49 (s, 1H), 3.19 (s, 1H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 156.2, 149.2, 125.0, 124.8, 119.8, 111.4, 81.1, 80.0; HRMS (ESI) calc'd for $C_8H_5NO_3$ + H = 164.0348, found 164.0345.



4.67

Triazole 4.67: $[\alpha]^{23}_{D}$ +9.0 (c 0.53, solvent); IR (neat) 2984, 2160, 1750, 1603, 1426, 1381, 1265, 1162, 1167, 1078, 1654, 1002, 735, 703 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.74 (s, 1H), 6.64 (d, *J* = 2.0 Hz, 2H), 6.02 (t, *J* = 2.0 Hz, 1H), 4.88 (dd, *J* = 10.3, 2.5 Hz, 1H), 4.39 (ddt, *J* = 15.1, 13.7, 7.0 Hz, 2H), 4.31 (d, *J* = 7.3 Hz, 1H), 4.05 (dd, *J* = 10.7, 1.7 Hz, 1H), 3.76 (dt, *J* = 13.8, 6.7 Hz, 1H), 3.63 (dt, *J* = 14.0, 7.7 Hz, 1H), 3.56 – 3.50 (m, 2H), 3.43 (s, 1H), 3.22 (dd, *J* = 10.2, 7.2 Hz, 1H), 3.11

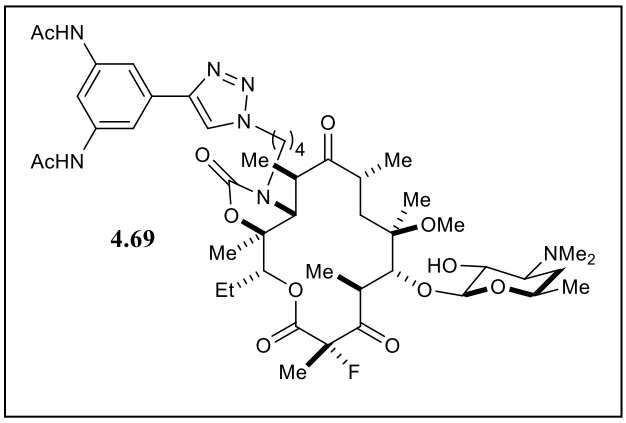
(q, *J* = 7.0 Hz, 3H), 2.64 – 2.57 (m, 1H), 2.54 (s, 3H), 2.46 (t, *J* = 10.5 Hz, 1H), 2.33 (s, 6H), 1.97 (dddd, *J* = 15.4, 14.0, 9.6, 6.2 Hz, 3H), 1.86 (dd, *J* = 14.5, 2.9 Hz, 1H), 1.78 (d, *J* = 21.4 Hz, 3H), 1.73 – 1.61 (m, 3H), 1.55 – 1.51 (m, 1H), 1.50 (s, 4H), 1.33 (s, 3H), 1.30 (d, *J* = 7.1 Hz, 4H), 1.27 – 1.21 (m, 6H), 1.18 (d, *J* = 6.9 Hz, 3H), 1.00 (d, *J* = 7.0 Hz, 4H), 0.88 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.7, 203.2, 166.7, 166.5, 157.4, 148.1 (2C), 132.7, 119.8, 104.2, 103.8, 101.7, 98.8, 97.1, 82.3, 81.0, 78.7, 70.4, 69.6, 66.1, 61.3, 49.9, 49.4, 44.7, 42.9, 41.0, 40.4, 39.7, 39.4, 29.8, 28.6, 27.7, 25.5, 25.3, 24.5, 22.3, 21.3, 19.9, 18.0, 15.2, 14.9, 13.9, 10.7; HRMS (ESI) calc'd for $C_{43}H_{66}FN_7O_{10}$ + H = 860.4933, found 860.4917



4.68

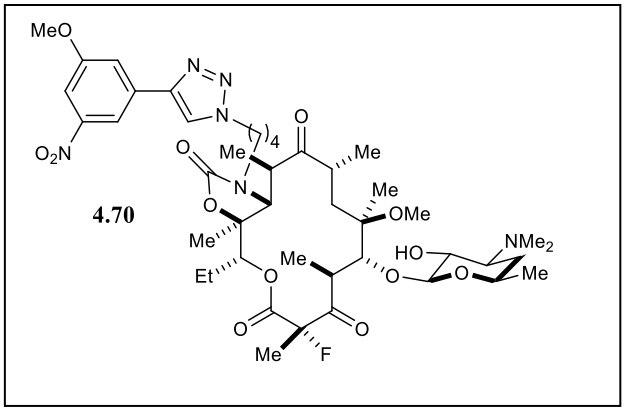
Triazole 4.68: $[\alpha]^{23}_D$ +10.0 (c 1.03); IR (neat) 2977, 2160, 1750, 1708, 1620, 1571, 1425, 1369, 1264, 1161, 1107, 1078, 1052, 1002, 978, 733, 702 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.92 (s, 1H), 7.79 (s, 1H), 7.40 (t, *J* = 2.0 Hz, 1H), 6.99 (t, *J* = 1.7 Hz, 1H), 6.96 (t, *J* = 1.8 Hz, 1H), 4.88 (dd, *J* = 10.4, 2.6 Hz, 1H), 4.44 (ddd, *J* = 14.3, 8.0, 6.5 Hz, 1H), 4.34 (dt, *J* = 14.1, 7.4 Hz, 1H), 4.28 (d, *J* = 7.2 Hz, 1H), 4.05 – 3.98 (m, 2H), 3.78 (dt, *J* = 13.9, 6.8 Hz, 1H), 3.63 (dt, *J* = 14.5, 7.4 Hz, 1H), 3.59 – 3.48 (m, 1H), 3.42 (s, 1H), 3.17 (dd, *J* = 10.3, 7.3 Hz, 1H), 3.10 (q, *J* = 7.0 Hz, 1H), 2.60 (ddd, *J* = 12.3, 7.1, 3.1 Hz, 1H), 2.51 (s, 3H), 2.44 (ddd, *J* = 12.3, 10.1, 3.9 Hz, 1H), 2.26 (s, 7H), 2.12 (s, 4H), 2.02 – 1.91 (m, 3H), 1.87 (dd, *J* = 14.5, 2.9 Hz, 1H), 1.78 (d, *J* = 21.3 Hz, 3H), 1.70 – 1.60 (m, 4H), 1.49 (s, 4H), 1.35 – 1.27 (m, 7H), 1.21 (dd, *J* = 13.1, 6.1 Hz, 17H), 0.99 (d, *J* = 6.9

Hz, 3H), 0.86 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.7, 204.0, 166.7, 157.4, 148.0, 139.8, 132.2, 119.9, 108.3, 107.1, 106.2, 104.4, 98.7, 97.1, 82.3, 81.1, 78.9, 78.8, 70.4, 69.8, 66.0, 61.5, 50.0, 49.4, 44.7, 43.0, 41.1, 40.4, 39.7, 39.4, 29.8, 28.4, 27.6, 25.5, 25.3, 24.9, 24.6, 22.3, 21.3, 19.9, 18.0, 15.3, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{45}H_{68}FN_7O_{11}$ + Na = 924.4859, found 924.4837.

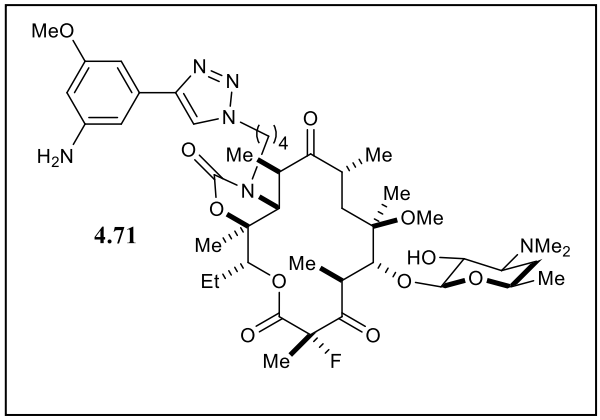


4.69

Triazole 4.69: $[\alpha]^{23}_D$ + 9.5 (c 1.0); IR (neat) 2981, 2161, 2035, 1748, 1695, 1623, 1575, 1418, 1368, 1261, 1219, 1162, 1078, 1057, 924, 868, 750, 667; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.45 (s, 2H), 7.92 (t, *J* = 1.9 Hz, 1H), 7.80 (s, 1H), 7.71 (d, *J* = 1.9 Hz, 2H), 4.86 (dd, *J* = 10.4, 2.6 Hz, 1H), 4.37 (dq, *J* = 14.8, 7.0 Hz, 2H), 4.28 (d, *J* = 7.3 Hz, 1H), 4.02 (dd, *J* = 10.7, 1.6 Hz, 1H), 3.74 (ddd, *J* = 13.9, 8.3, 5.4 Hz, 1H), 3.60 (dt, *J* = 14.6, 7.7 Hz, 1H), 3.52 (dtd, *J* = 14.1, 6.6, 6.1, 2.8 Hz, 2H), 3.40 (s, 1H), 3.18 (dd, *J* = 10.2, 7.3 Hz, 1H), 3.08 (q, *J* = 7.0 Hz, 1H), 2.64 – 2.55 (m, 1H), 2.51 (s, 3H), 2.48 – 2.44 (m, 1H), 2.26 (s, 7H), 2.11 (s, 6H), 1.96 – 1.89 (m, 3H), 1.86 (dd, *J* = 14.6, 2.9 Hz, 1H), 1.76 (d, *J* = 21.4 Hz, 3H), 1.63 (dddd, *J* = 25.8, 11.7, 5.7, 3.3 Hz, 4H), 1.53 – 1.49 (m, 1H), 1.48 (s, 4H), 1.33 – 1.26 (m, 6H), 1.25 – 1.18 (m, 5H), 1.16 (d, *J* = 6.9 Hz, 3H), 0.99 (d, *J* = 6.9 Hz, 3H), 0.85 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.6, 203.6, 203.4, 169.2, 166.5, 166.4, 157.4, 147.3, 139.3, 131.8, 120.5, 112.9, 111.1, 104.4, 98.7, 97.0, 82.3, 81.0, 78.7 (2C), 70.5, 69.7, 65.9, 61.3, 50.0, 49.3, 44.7, 43.0, 41.0, 40.3, 39.7, 39.3, 29.8, 28.3, 27.6, 25.5, 25.3, 24.6, 24.4, 22.2, 21.3, 19.9, 18.0, 15.2, 14.8, 13.9, 10.6; HRMS (ESI) calc'd for $C_{47}H_{70}FN_7O_{12}$ + H = 944.5145, found 944.5170.

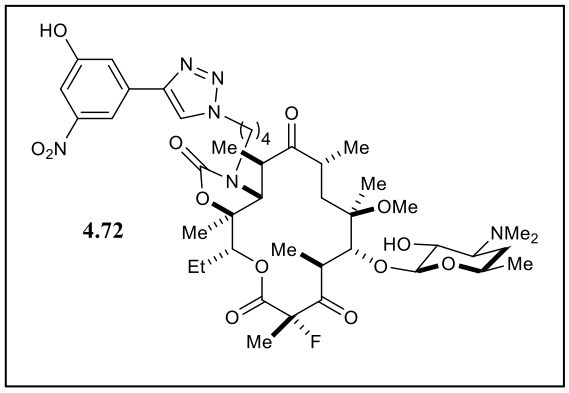


Triazole 4.70: $[\alpha]^{23}_{D}$ +30.0 (c 1.26); IR (neat) 2970, 2926, 2850, 1752, 1709, 1642, 1533, 1457, 1379, 1345, 1249, 1163, 1108, 1078, 1052, 1003, 922, 751; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.16 (s, 1H), 8.02 (s, 1H), 7.85 (d, *J* = 2.3 Hz, 1H), 7.67 (d, *J* = 2.4 Hz, 1H), 4.89 – 4.77 (m, 1H), 4.45 (t, *J* = 7.4 Hz, 4H), 4.30 (d, *J* = 7.3 Hz, 1H), 4.04 (d, *J* = 10.7 Hz, 1H), 3.94 (s, 3H), 3.75 (dt, *J* = 13.6, 6.5 Hz, 1H), 3.63 (dt, *J* = 14.6, 7.8 Hz, 1H), 3.52 (q, *J* = 6.5, 5.8 Hz, 2H), 3.41 (s, 1H), 3.21 (dd, *J* = 10.2, 7.2 Hz, 1H), 3.10 (d, *J* = 7.2 Hz, 1H), 2.74 – 2.44 (m, 5H), 2.31 (s, 7H), 2.16 (d, *J* = 4.1 Hz, 1H), 2.07 – 1.91 (m, 5H), 1.91 – 1.82 (m, 1H), 1.82 – 1.60 (m, 7H), 1.49 (s, 4H), 1.38 – 1.10 (m, 18H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.93 – 0.74 (m, 5H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.9, 202.6, 166.8, 160.7, 157.4, 149.7, 145.8, 133.4, 121.0, 117.3, 113.0, 108.4, 104.2, 98.9, 96.9, 82.3, 80.7, 78.7, 70.4, 69.5, 65.9, 61.0, 56.2, 50.1, 49.3, 44.7, 42.8, 41.0, 40.2, 39.7, 39.3, 29.8, 28.6, 27.7, 25.5, 25.1, 24.3, 22.3, 21.3, 19.9, 18.0, 15.2, 14.8, 13.9, 10.6; HRMS (ESI) calc'd for $C_{44}H_{65}FN_6O_{13}$ + H = 905.4672, found 905.4706.



Triazole 4.71: $[\alpha]^{23}_{D}$ +34.5 (c 0.42); IR (neat) 2980, 2884, 2360, 1749, 1708, 1600, 1558, 1457, 1381, 1261, 1215, 1163, 1053, 1000, 956, 748, 667; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.79 (s, 1H), 6.82 (p, *J* = 1.5 Hz, 2H), 6.25 – 6.13 (m, 1H), 4.87 (dd,

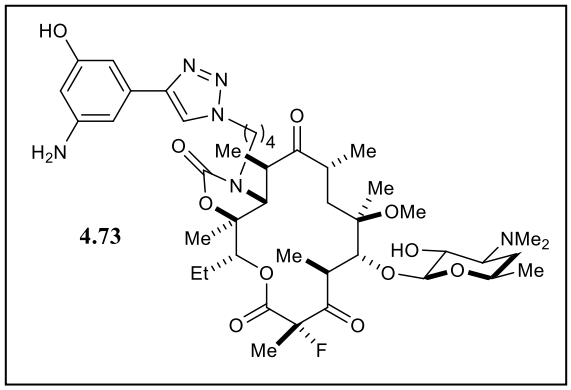
J = 10.3, 2.6 Hz, 1H), 4.41 (dd, *J* = 8.3, 6.5 Hz, 2H), 4.30 (d, *J* = 7.2 Hz, 1H), 4.12 – 4.00 (m, 1H), 3.82 (s, 3H), 3.79 – 3.71 (m, 1H), 3.63 (dt, *J* = 14.3, 7.6 Hz, 1H), 3.57 – 3.48 (m, 3H), 3.43 (s, 1H), 3.18 (t, *J* = 8.8 Hz, 1H), 3.11 (q, *J* = 6.9 Hz, 1H), 2.55 (s, 3H), 2.45 (t, *J* = 10.8 Hz, 1H), 2.27 (s, 7H), 2.17 (d, *J* = 2.3 Hz, 1H), 2.03 – 1.92 (m, 3H), 1.91 – 1.83 (m, 2H), 1.80 (d, *J* = 2.1 Hz, 2H), 1.75 (d, *J* = 2.1 Hz, 2H), 1.72 – 1.59 (m, 6H), 1.50 (s, 4H), 1.36 – 1.27 (m, 7H), 1.26 – 1.14 (m, 10H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.87 (td, *J* = 7.3, 2.2 Hz, 4H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.7, 202.9, 166.5, 161.2, 157.4, 148.2, 132.8, 120.0, 105.8, 104.4, 101.4, 101.3, 99.0, 96.9, 82.3, 80.9, 78.7, 70.5, 69.8, 66.0, 61.2, 55.5, 49.9, 49.4, 44.7, 42.9, 41.0, 40.4, 39.7, 39.4, 29.8, 28.3, 27.7, 25.5, 25.3, 24.4, 22.3, 21.3, 19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{44}H_{67}FN_6O_{11}$ + Na = 897.4750, found 897.4754.



4.72

Triazole 4.72: [α] $^{23}_{D}$ +17 (c 0.5); IR (neat) 3019, 2980, 1749, 1627, 1529, 1457, 1348, 1215, 1053, 978, 747, 668; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.17 – 8.13 (m, 1H), 8.03 (s, 1H), 7.90 (dd, *J* = 2.4, 1.4 Hz, 1H), 7.69 (t, *J* = 2.2 Hz, 1H), 4.86 (dd, *J* = 10.2, 2.6 Hz, 1H), 4.57 – 4.40 (m, 2H), 4.30 (d, *J* = 7.3 Hz, 1H), 4.02 (dd, *J* = 10.7, 1.7 Hz, 1H), 3.82 (dt, *J* = 13.7, 6.7 Hz, 1H), 3.66 (dt, *J* = 14.6, 7.7 Hz, 1H), 3.60 – 3.48 (m, 2H), 3.43 (s, 1H), 3.18 (dd, *J* = 10.2, 7.3 Hz, 1H), 3.12 (q, *J* = 7.0 Hz, 1H), 2.62 (ddd, *J* = 12.4, 6.9, 2.9 Hz, 1H), 2.51 (s, 3H), 2.49 – 2.42 (m, 1H), 2.27 (s, 7H), 2.06 – 1.92 (m, 3H), 1.88 (dd, *J* = 14.5, 2.9 Hz, 1H), 1.77 (d, *J* = 21.4 Hz, 3H), 1.66 (dddt, *J* = 21.8, 17.6, 14.5, 7.3 Hz, 4H), 1.51

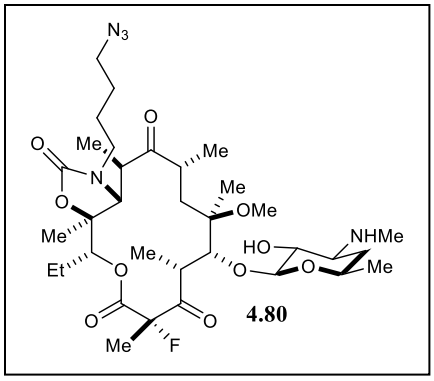
(s, 5H), 1.35 – 1.27 (m, 7H), 1.22 (d, *J* = 6.0 Hz, 4H), 1.21 – 1.15 (m, 4H), 1.01 (d, *J* = 7.0 Hz, 3H), 0.85 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.8, 204.1, 166.8, 158.0, 157.7, 149.9, 146.0, 133.2, 121.1, 119.2, 112.6, 110.4, 104.4, 98.7, 97.1, 82.6, 80.9, 78.9, 70.6, 69.9, 65.9, 61.5, 50.3, 49.3, 44.7, 43.0, 41.1, 40.4, 39.8, 39.4, 28.4, 27.6, 25.4, 25.3, 24.6, 22.3, 21.3, 19.9, 18.0, 15.3, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{43}H_{63}FN_6O_{13}$ + H = 891.4515, found 891.4515.



4.73

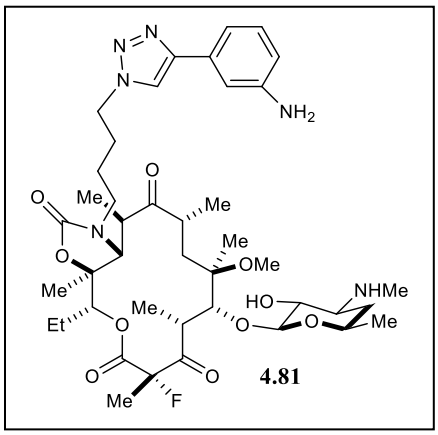
Triazole 4.73: [α] $^{23}_D$ +21.7 (c 0.62); IR (neat) 2990, 2883, 2360, 2341, 1750, 1599, 1457, 1381, 1283, 1261, 1215, 1053, 1002, 978, 748, 667; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.77 (s, 1H), 6.79 (d, *J* = 27.3 Hz, 2H), 6.23 (s, 1H), 4.89 (dd, *J* = 10.6, 2.5 Hz, 1H), 4.51 – 4.40 (m, 1H), 4.32 (dd, *J* = 24.6, 7.1 Hz, 2H), 4.09 – 3.98 (m, 1H), 3.80 (dt, *J* = 13.7, 6.7 Hz, 1H), 3.70 – 3.47 (m, 3H), 3.43 (s, 1H), 3.14 (dq, *J* = 29.7, 7.1 Hz, 2H), 2.60 (t, *J* = 10.8 Hz, 1H), 2.56 – 2.41 (m, 4H), 2.27 (s, 6H), 2.04 – 1.85 (m, 3H), 1.85 – 1.74 (m, 3H), 1.74 – 1.58 (m, 4H), 1.50 (s, 4H), 1.35 – 1.12 (m, 16H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.87 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.8, 204.3, 166.7, 166.5, 157.9, 148.5, 132.6, 120.0, 105.2, 104.3, 103.8, 102.2, 98.9, 96.9, 82.3, 81.0, 78.9, 78.8, 70.5, 69.8, 65.9, 61.5, 50.0, 49.4, 44.7, 43.0, 41.1, 40.7, 39.7, 39.4, 28.3, 27.5, 25.5, 25.3, 24.7, 22.3, 21.3, 19.9, 18.0, 15.3, 14.9, 14.0, 10.6; HRMS (ESI) calc'd for $C_{43}H_{65}FN_6O_{11}$ + H = 861.4774, found 861.4785.

5.3.3 Synthesis of Solithromycin Analogs with Biaryl Side Chains



Demethylation 4.80: NaOAc (52mg, 0.25 mol) and I_2 were (63 mg, 0.25 mmol) were added sequentially to a solution of **4.5 (**152 mg, 0.21 mmol) in MeOH/H_2O (2.2 mL, 10:1). The reaction mixture was heated under gentle reflux. During the course of the reaction, volatiles were slowly distilled off, and a 2N NaOH solution (X mL) was added every 15 min to adjust the pH to 8. Additional charges of iodine (63 mg, 0.25 mmol) were added after 1 h and 1.5 h. Upon completion of the reaction, the reaction mixture was cooled to rt. The mixture was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with brine (20 mL) and dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified by flash column chromatography eluting with 0−10% MeOH with 1% of NH_4OH to give 85 mg (57%, BRSM 72%) of **4.80** as a yellow foam. $[\alpha]^{23}_D$ +57.3 (c 1.24, $CHCl_3$); IR (neat) 2971, 2360, 2341, 2160, 2097, 2029, 1978, 1756, 1700, 1457, 1374, 1261, 1235, 1061, 1002; ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.87 (dd, *J* = 10.4, 2.5 Hz, 1H), 4.31 (d, *J* = 7.5 Hz, 1H), 4.10 (dd, *J* = 10.8, 1.7 Hz, 1H), 3.73 – 3.62 (m, 1H), 3.55 (dddd, *J* = 15.6, 7.3, 4.9, 3.3 Hz, 3H), 3.42 (s, 1H), 3.36 – 3.22 (m, 2H), 3.15 – 3.03 (m, 2H), 2.68 – 2.59 (m, 1H), 2.57 (s, 3H), 2.47 (ddd, *J* = 11.6, 9.6, 4.3 Hz, 1H), 2.42 (s, 3H), 2.04 – 1.92 (m, 2H), 1.81 (s, 2H), 1.78 (d, *J* = 2.8 Hz, 1H), 1.75 (s, 1H), 1.68 – 1.56 (m, 6H), 1.53 (dd, *J* = 11.3, 3.3 Hz, 1H), 1.49 (s, 3H), 1.34 (s, 3H), 1.25 (dd, *J* = 14.2, 6.6 Hz, 6H), 1.18 (d, *J* = 6.8 Hz, 3H), 1.09 (dt, *J* = 13.0, 11.4 Hz, 1H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.88 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.5, 202.9, 166.5, 157.2, 103.5, 99.0, 96.9, 82.1, 80.3, 78.7, 78.7, 74.5, 69.5,

61.0, 51.1, 49.3, 44.7, 43.2, 40.8, 39.7, 37.4, 33.2, 26.3, 25.5, 25.3, 24.5, 22.3, 21.1, 19.8, 18.1, 15.3, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{34}H_{56}FN_5O_{10}$ + H = 714.4089, found 714.4078.



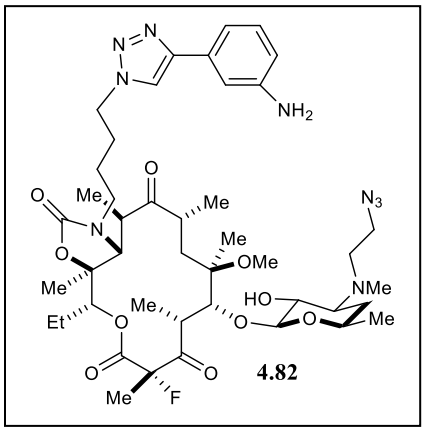
4.81

Secondary amine 4.81: $[\alpha]^{23}_D$ +11.8 (c 0.89, $CHCl_3$); IR (neat) 2979, 2524, 2159, 2030, 1732, 1530, 1374, 1259, 1215, 1124, 1079, 999, 868, 765, 692; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.80 (s, 1H), 7.28 – 7.27 (m, 1H), 7.21 – 7.13 (m, 2H), 6.64 (ddd, *J* = 7.6, 2.4, 1.4 Hz, 1H), 4.87 (dd, *J* = 10.4, 2.5 Hz, 1H), 4.41 (td, *J* = 7.2, 1.8 Hz, 2H), 4.30 (d, *J* = 7.5 Hz, 1H), 4.08 (dd, *J* = 10.7, 1.6 Hz, 1H), 3.75 (tt, *J* = 12.8, 5.9 Hz, 1H), 3.66 – 3.60 (m, 1H), 3.55 (ddtd, *J* = 21.9, 14.1, 7.0, 3.1 Hz, 2H), 3.42 (s, 1H), 3.17 (dd, *J* = 9.8, 7.5 Hz, 1H), 3.09 (q, *J* = 7.0 Hz, 1H), 2.64 – 2.56 (m, 1H), 2.54 (s, 3H), 2.46 (s, 3H), 1.99 (dddd, *J* = 23.1, 16.0, 7.6, 5.6 Hz, 4H), 1.82 – 1.73 (m, 4H), 1.65 (dddt, *J* = 29.0, 14.6, 10.4, 6.9 Hz, 3H), 1.49 (s, 4H), 1.32 (s, 3H), 1.28 – 1.20 (m, 7H), 1.17 (d, *J* = 7.0 Hz, 4H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.87 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.5, 202.5, 166.6, 157.3, 148.0, 147.0, 131.8, 129.8, 119.8, 116.2, 114.9, 112.5, 103.5, 98.8, 97.2, 82.2, 80.4, 78.7, 73.9, 69.3, 61.2, 60.8, 49.9, 44.6, 42.9, 40.8, 39.7, 39.4, 36.9, 32.9, 29.8, 27.7, 25.4, 24.4, 22.3, 21.1, 19.9, 18.1, 15.3, 14.8, 13.9, 10.6; HRMS (ESI) calc'd for $C_{42}H_{63}FN_6O_{10}$ + Na = 853.4487, found 853.4499.

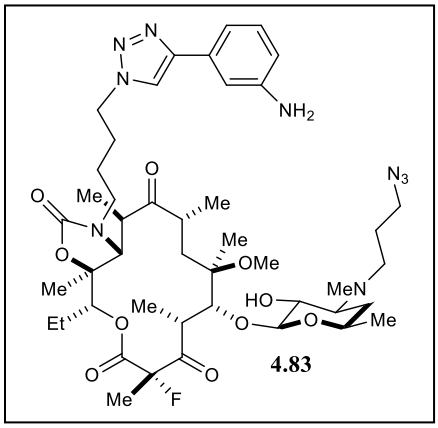
General Experimental Procedure for Alkylation:

A mixture of secondary amine precursor **4.81** (1.0 equiv), powdered K_2CO_3 (1.5 equiv), and ω-azido mesylate (1.5 equiv) in CH_3CN (0.2 M) was stirred at 70 °C for 24-48 h. The

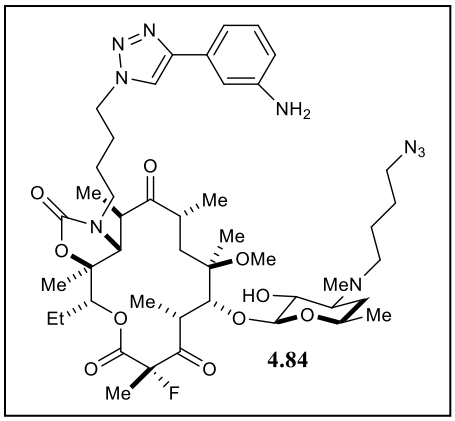
reaction mixture was cooled to 50 °C, and H_2O (3 mL) was added. The mixture was partitioned between EtOAc (10 mL) and H_2O (5 mL). The organic layer was washed with brine (20 mL) and dried over Na_2SO_4. The solvent was removed under reduced pressure, and the residue was purified by flash column chromatography eluting with 0−5% MeOH with 1% of NH_4OH to give products **4.82-4.84** (60-70% yield).



Azide 4.82: $[\alpha]^{23}_D$ +12.3 (c 0.98, $CHCl_3$); IR (neat) 2980, 2159, 2030, 1728, 1520, 1455, 1329, 1215, 1161, 1017, 999, 869, 748, 666; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.80 (s, 1H), 7.28 – 7.26 (m, 1H), 7.19 – 7.14 (m, 2H), 6.64 (dt, *J* = 7.1, 2.2 Hz, 1H), 4.87 (dd, *J* = 10.3, 2.5 Hz, 1H), 4.41 (td, *J* = 7.2, 1.4 Hz, 2H), 4.31 (d, *J* = 7.2 Hz, 1H), 4.06 (dd, *J* = 10.7, 1.6 Hz, 1H), 3.80 – 3.71 (m, 1H), 3.67 – 3.59 (m, 1H), 3.58 – 3.49 (m, 2H), 3.40 (d, *J* = 26.6 Hz, 4H), 3.10 (d, *J* = 7.0 Hz, 1H), 2.78 (d, *J* = 9.6 Hz, 1H), 2.65 – 2.56 (m, 1H), 2.54 (s, 3H), 2.32 (s, 3H), 1.97 (ddd, *J* = 12.3, 9.0, 6.1 Hz, 3H), 1.87 (dd, *J* = 14.4, 2.9 Hz, 1H), 1.80 (s, 1H), 1.75 (s, 1H), 1.72 – 1.59 (m, 6H), 1.56 – 1.51 (m, 1H), 1.50 (s, 4H), 1.34 – 1.28 (m, 7H), 1.26 – 1.22 (m, 4H), 1.18 (d, *J* = 6.9 Hz, 3H), 1.00 (d, *J* = 7.1 Hz, 3H), 0.87 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.7, 203.1, 166.5, 157.4, 148.0, 147.0, 131.9, 129.8, 119.8, 116.3, 114.9, 112.5, 104.1, 98.9, 96.9, 82.3, 80.9, 78.7, 70.6, 69.6, 66.4, 61.2, 52.8, 49.9, 49.4, 44.7, 42.9, 41.0, 39.3, 37.2, 30.0, 29.8, 27.7, 25.5, 25.3, 24.4, 22.3, 21.2, 19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{44}H_{66}FN_9O_{10}$ + Na = 922.4814, found 922.4815.

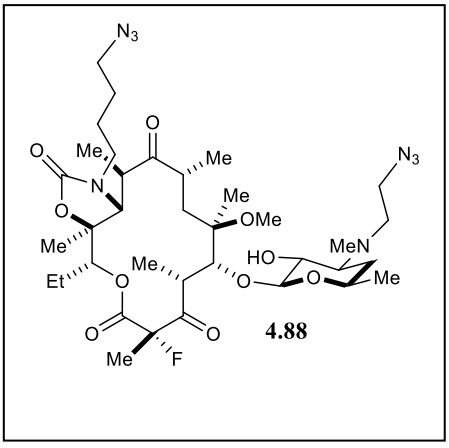


Azide 4.83: $[\alpha]^{23}_{D}$ +8.6 (c 0.81, $CHCl_3$); IR (neat) 2974, 2938, 2887, 2159, 2097, 2031, 1977, 1756, 1457, 1377, 1168, 1003, 833; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.80 (d, *J* = 2.8 Hz, 1H), 7.29 – 7.27 (m, 1H), 7.21 – 7.15 (m, 2H), 6.65 (dt, *J* = 7.0, 2.2 Hz, 1H), 4.88 (dd, *J* = 10.5, 2.5 Hz, 1H), 4.42 (td, *J* = 7.2, 1.5 Hz, 2H), 4.30 (d, *J* = 7.2 Hz, 1H), 4.05 (dd, *J* = 10.8, 1.7 Hz, 1H), 3.83 – 3.71 (m, 3H), 3.63 (dt, *J* = 14.5, 7.7 Hz, 1H), 3.53 (dq, *J* = 15.1, 6.6, 5.0 Hz, 2H), 3.46 – 3.30 (m, 4H), 3.21 (dd, *J* = 10.1, 7.3 Hz, 1H), 3.10 (t, *J* = 7.0 Hz, 1H), 2.68 – 2.57 (m, 2H), 2.54 (d, *J* = 3.7 Hz, 3H), 2.50 – 2.34 (m, 1H), 2.24 (s, 3H), 2.03 – 1.84 (m, 4H), 1.83 – 1.59 (m, 11H), 1.50 (s, 4H), 1.36 – 1.28 (m, 6H), 1.24 (q, *J* = 6.0, 4.7 Hz, 7H), 1.21 – 1.14 (m, 3H), 1.01 (d, *J* = 6.9 Hz, 3H), 0.88 (t, *J* = 7.3 Hz, 4H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.7, 203.1, 166.7, 157.4, 148.0, 147.0, 131.9, 129.8, 119.8, 116.3, 114.9, 112.5, 104.3, 99.0, 96.9, 82.3, 80.9, 78.7, 70.4, 69.8, 66.2, 61.2, 50.5, 49.9, 49.5, 49.4, 44.7, 42.9, 41.0, 39.7, 39.4, 37.0, 29.8, 29.4, 27.8, 27.5, 24.4, 22.3, 21.3, 19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{45}H_{68}FN_9O_{10}$ + Na = 936.4965, found 936.4929.



Azide 4.84: $[\alpha]^{23}_{D}$ +5.1 (c 0.86, $CHCl_3$); IR (neat) 2980, 2438, 2159, 2031, 1977, 1751, 1436, 1379, 1265, 1215, 1077, 1051, 752, 668; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.80 (s, 1H), 7.28 (t, *J* = 1.6 Hz, 1H), 7.21 – 7.15 (m, 3H), 6.65 (dt, *J* = 7.2, 2.1 Hz, 1H), 4.88 (dd, *J* = 10.6, 2.5 Hz, 2H), 4.42 (td, *J* = 7.3, 2.3

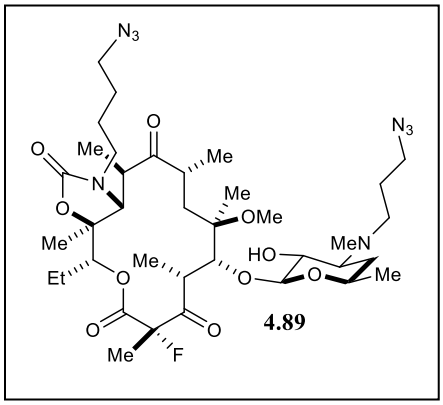
Hz, 3H), 4.30 (d, *J* = 7.2 Hz, 1H), 4.15 – 4.03 (m, 2H), 3.80 – 3.71 (m, 5H), 3.63 (dt, *J* = 13.9, 7.7 Hz, 2H), 3.57 – 3.46 (m, 3H), 3.45 – 3.41 (m, 3H), 3.30 (td, *J* = 6.5, 2.9 Hz, 3H), 3.19 (dd, *J* = 10.2, 7.2 Hz, 1H), 3.11 (q, *J* = 6.9 Hz, 2H), 2.60 (dt, *J* = 12.1, 8.1 Hz, 2H), 2.54 (d, *J* = 4.6 Hz, 4H), 2.48 (d, *J* = 9.6 Hz, 1H), 2.34 (dd, *J* = 12.8, 7.2 Hz, 1H), 2.23 (s, 3H), 1.97 (ddd, *J* = 12.6, 9.0, 6.2 Hz, 5H), 1.90 – 1.83 (m, 1H), 1.81 – 1.47 (m, 20H), 1.36 – 1.28 (m, 8H), 1.24 (dd, *J* = 11.6, 5.9 Hz, 7H), 1.19 (dd, *J* = 7.0, 3.8 Hz, 4H), 1.01 (dd, *J* = 6.8, 1.5 Hz, 5H), 0.88 (t, *J* = 7.4 Hz, 5H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.7, 203.1, 166.7, 157.4, 148.0, 147.0, 131.9, 129.8, 119.8, 116.3, 114.9, 112.5, 104.4, 98.8, 97.1, 82.3, 81.0, 78.7, 70.4, 69.8, 66.1, 61.2, 52.9, 51.5, 49.9, 49.4, 44.8, 42.9, 41.0, 39.7, 39.4, 37.0, 29.3, 27.8, 26.7, 25.6, 25.5, 24.4, 22.3, 21.3, 19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{46}H_{70}FN_9O_{10}$ + H = 928.5308, found 928.5306.



4.88

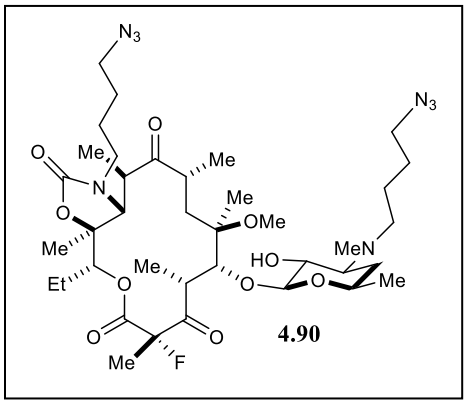
Bis-azide 4.88: $[\alpha]^{23}_D$ +19.1 (c 0.93, $CHCl_3$); IR (neat) 2971, 2360, 2341, 2160, 2097, 2029, 1978, 1756, 1700, 1457, 1374, 1261, 1235, 1061, 1002; ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.86 (dd, *J* = 10.6, 2.5 Hz, 1H), 4.32 (d, *J* = 7.2 Hz, 1H), 4.07 (d, *J* = 10.6 Hz, 1H), 3.67 (q, *J* = 7.0, 6.5 Hz, 1H), 3.54 (ddd, *J* = 15.1, 12.4, 6.2 Hz, 2H), 3.45 – 3.16 (m, 6H), 3.10 (q, *J* = 7.4 Hz, 1H), 2.78 (dt, *J* = 12.6, 6.3 Hz, 1H), 2.65 – 2.49 (m, 6H), 2.31 (s, 3H), 1.97 (ddd, *J* = 14.4, 7.6, 2.4 Hz, 1H), 1.87 (dd, *J* = 14.5, 2.8 Hz, 1H), 1.78 (d, *J* = 21.3 Hz, 3H), 1.72 – 1.46 (m, 11H), 1.38 – 1.15 (m, 13H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.88 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.7, 203.2, 166.6, 157.2, 104.1, 98.9, 96.8, 82.1, 80.8, 78.7, 70.6, 69.6, 66.4, 61.0, 52.7, 51.1, 49.5, 49.3, 44.8, 43.2, 40.9, 39.7, 39.4, 37.1, 29.9, 26.3, 25.5,

25.3, 24.5, 22.3,19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{36}H_{59}FN_8O_{10}$ + H = 783.4411, found 783.4413.

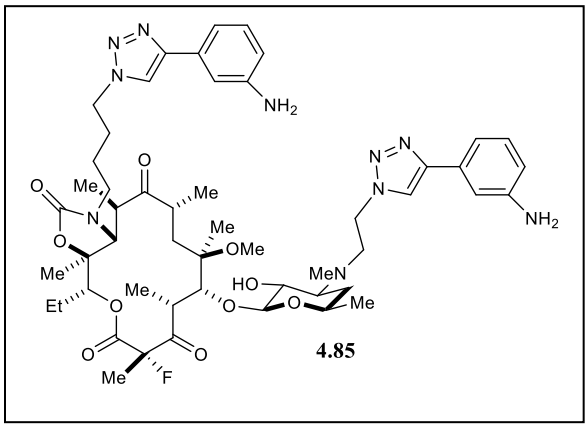


4.89

Bis-azide 4.89: $[\alpha]^{23}_D$ +27.8 (c 0.31, $CHCl_3$); IR (neat) 2971, 2358, 2341, 2160, 2097, 2029, 1978, 1756, 1690, 1457, 1374, 1261, 1238, 1061, 1002; ^{1}H NMR (500 MHz, $CDCl_3$) δ 4.87 (dd, *J* = 10.4, 2.5 Hz, 1H), 4.31 (d, *J* = 7.2 Hz, 1H), 4.07 (dd, *J* = 10.7, 1.7 Hz, 1H), 3.68 (dt, *J* = 13.5, 6.5 Hz, 1H), 3.60 – 3.48 (m, 3H), 3.43 (d, *J* = 3.9 Hz, 1H), 3.41 – 3.37 (m, 1H), 3.36 – 3.25 (m, 3H), 3.21 (dd, *J* = 10.2, 7.2 Hz, 1H), 3.10 (q, *J* = 7.0 Hz, 1H), 2.63 (dtd, *J* = 11.0, 7.0, 3.0 Hz, 2H), 2.58 (s, 3H), 2.54 – 2.46 (m, 1H), 2.40 (dt, *J* = 13.1, 6.7 Hz, 1H), 2.24 (s, 3H), 1.97 (dtd, *J* = 15.3, 7.6, 2.7 Hz, 1H), 1.88 (dd, *J* = 14.5, 2.9 Hz, 1H), 1.80 (s, 2H), 1.75 (d, *J* = 6.3 Hz, 3H), 1.70 – 1.52 (m, 9H), 1.49 (s, 3H), 1.35 (s, 3H), 1.30 (d, *J* = 7.1 Hz, 3H), 1.27 – 1.22 (m, 5H), 1.19 (d, *J* = 6.9 Hz, 3H), 1.00 (d, *J* = 7.0 Hz, 3H), 0.89 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.7, 203.2, 166.6, 157.2, 104.3, 98.7, 97.1, 82.2, 80.9, 78.7, 70.5, 69.8, 66.2, 61.06, 51.4, 50.5, 49.5, 44.8, 44.5, 43.2, 41.0, 39.7, 39.4, 37.0, 29.4, 27.5, 26.4, 25.5, 25.3, 24.5, 22.3, 19.9, 18.1, 15.2, 14.9, 13.9, 10.8; HRMS (ESI) calc'd for $C_{37}H_{61}FN_8O_{10}$ + Na = 819.4392, found 819.4397.

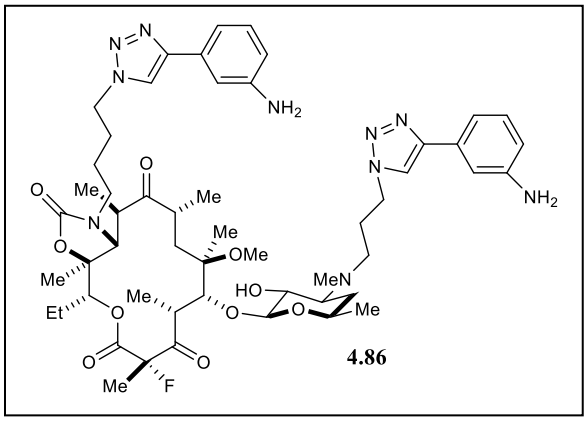


Bis-azide 4.90: $[\alpha]^{23}_D$ +18.3 (c 1.1, $CHCl_3$); IR (neat) 2971, 2360, 2341, 2160, 2097, 2029, 1978, 1756, 1700, 1457, 1364, 1261, 1235, 1061, 1002^{1}H NMR (400 MHz, $CDCl_3$) δ 4.87 (dd, *J* = 10.4, 2.5 Hz, 1H), 4.30 (d, *J* = 7.2 Hz, 1H), 4.06 (d, *J* = 10.5 Hz, 1H), 3.67 (q, *J* = 7.0, 6.5 Hz, 1H), 3.62 – 3.45 (m, 4H), 3.42 (s, 1H), 3.29 (t, *J* = 6.3 Hz, 4H), 3.19 (dd, *J* = 10.1, 7.2 Hz, 1H), 3.10 (q, *J* = 6.9 Hz, 1H), 2.64 (d, *J* = 5.9 Hz, 0H), 2.58 (s, 4H), 2.54 – 2.44 (m, 1H), 2.39 – 2.28 (m, 1H), 2.23 (s, 3H), 1.97 (ddt, *J* = 16.5, 8.9, 4.5 Hz, 1H), 1.88 (dd, *J* = 14.5, 2.9 Hz, 1H), 1.78 (d, *J* = 21.4 Hz, 3H), 1.72 – 1.44 (m, 17H), 1.37 – 1.14 (m, 14H), 1.00 (d, *J* = 6.9 Hz, 3H), 0.88 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.7, 202.9, 166.6, 157.2, 104.4, 98.9, 96.9, 82.1, 81.0, 78.7, 70.3, 69.8, 66.0, 61.0, 52.8, 51.4, 51.1, 49.3, 44.8, 43.2, 40.9, 39.7, 39.4, 37.0, 29.3, 26.6, 26.4, 25.5, 25.3, 24.5, 22.3, 21.3, 19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{38}H_{63}FN_8O_{10}$ + H = 811.4729, found 811.4759.



Bis-triazole 4.85: $[\alpha]^{23}_D$ +4.8 (c 0.88, $CHCl_3$); IR (neat) 2979, 2524, 2159, 2030, 1732, 1530, 1374, 1263, 1215, 1124, 1079, 999, 868, 765, 692; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.80 (s, 1H), 7.75 (s, 1H), 7.27 (d, *J* = 2.0 Hz, 1H), 7.24 (t, *J* = 2.0 Hz, 1H), 7.20 – 7.15 (m, 3H), 7.10 (dt, *J* = 7.6, 1.3 Hz, 1H), 6.65 (ddd, *J* = 9.3, 7.8, 2.2 Hz, 2H), 4.87 (dd, *J* = 10.5, 2.6 Hz, 1H), 4.49 – 4.37 (m, 4H), 4.25 (d, *J* = 7.2 Hz, 1H), 4.03 (dd, *J*

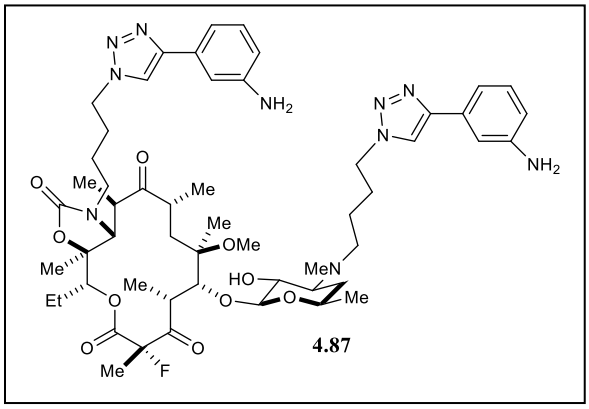
= 10.7, 1.7 Hz, 1H), 3.76 (dq, *J* = 13.6, 8.5, 6.9 Hz, 5H), 3.62 (dt, *J* = 14.6, 7.8 Hz, 1H), 3.49 (dtt, *J* = 12.1, 7.3, 4.3 Hz, 2H), 3.42 (s, 1H), 3.17 (dd, *J* = 10.2, 7.2 Hz, 1H), 3.09 (dq, *J* = 12.8, 6.7 Hz, 2H), 2.99 (s, 1H), 2.92 (dt, *J* = 12.8, 6.1 Hz, 1H), 2.60 (ddq, *J* = 13.5, 6.5, 4.0 Hz, 1H), 2.53 (s, 3H), 2.51 – 2.45 (m, 1H), 2.32 (s, 3H), 2.03 – 1.92 (m, 3H), 1.83 (dd, *J* = 14.5, 2.9 Hz, 1H), 1.77 (d, *J* = 21.3 Hz, 3H), 1.72 – 1.59 (m, 10H), 1.49 (s, 4H), 1.30 (d, *J* = 9.0 Hz, 4H), 1.23 (dd, *J* = 12.5, 6.4 Hz, 7H), 1.18 (d, *J* = 6.8 Hz, 3H), 1.00 (d, *J* = 7.0 Hz, 3H), 0.87 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.7, 203.0, 166.7, 157.4, 148.1, 148.0, 147.0, 131.9, 131.5, 129.9, 129.8, 120.3, 119.8, 116.3, 116.2, 115.1, 114.9, 112.5, 112.4, 104.1, 98.7, 97.1, 82.3, 80.8, 78.7, 70.6, 69.4, 66.0, 61.2, 53.9, 49.9, 49.4, 44.7, 42.9, 41.0, 39.7, 39.4, 37.1, 30.3, 29.8, 27.7, 25.4, 25.3, 24.4, 22.3, 21.2, 19.9, 18.1, 15.2, 14.9, 13.9, 10.6; HRMS (ESI) calc'd for $C_{52}H_{73}FN_{10}O_{10}$ + H = 1017.5573, found 1017.5569.



4.86

Bis-triazole 4.86 $[\alpha]^{23}_D$ +12.5 (c 0.47, $CHCl_3$); IR (neat) 2979, 2524, 2160, 2030, 1732, 1530, 1374, 1259, 1220, 1124, 1080, 999, 868, 765, 692; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.81 (s, 1H), 7.74 (s, 1H), 7.28 (s, 2H), 7.18 (q, *J* = 7.5 Hz, 3H), 7.12 (d, *J* = 7.6 Hz, 1H), 6.69 – 6.63 (m, 2H), 4.88 (d, *J* = 10.2 Hz, 1H), 4.50 – 4.38 (m, 4H), 4.30 (d, *J* = 7.2 Hz, 1H), 4.05 (d, *J* = 10.6 Hz, 1H), 3.75 (d, *J* = 6.3 Hz, 1H), 3.64 (q, *J* = 7.3, 6.8 Hz, 1H), 3.52 (d, *J* = 17.3 Hz, 3H), 3.43 (s, 1H), 3.21 (t, *J* = 8.8 Hz, 1H), 3.11 (d, *J* = 7.1 Hz, 1H), 2.60 (s, 2H), 2.54 (s, 4H), 2.36 (s, 1H), 2.24 (s, 3H), 2.11 (s, 3H), 1.97 (t, *J* = 7.7 Hz, 3H), 1.90 – 1.74 (m, 4H), 1.69 (d, *J* = 14.0 Hz, 3H), 1.58 (s, 17H), 1.51 (s, 4H), 1.35 –

1.29 (m, 7H), 1.22 (dq, *J* = 18.3, 9.3, 6.8 Hz, 11H), 1.01 (d, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 7.3 Hz, 4H); ^{13}C NMR (101 MHz, $CDCl_3$) δ 216.7, 203.0, 157.4, 148.0, 147.0, 131.5, 129.9, 129.8, 120.3, 119.8, 116.3, 116.2, 115.1, 114.9, 112.5, 112.4, 104.1, 96.9, 82.3, 80.8, 78.7, 70.6, 69.5, 68.4, 68.3, 66.0, 61.2, 54.9, 53.8, 50.9, 49.9, 49.4, 48.9, 44.7, 42.9, 39.7, 39.4, 37.0, 30.2, 30.1, 29.8, 27.7, 25.5, 25.2, 24.4, 22.3, 21.2, 18.1, 15.2, 14.8, 13.9, 13.7, 10.6; HRMS (ESI) calc'd for $C_{53}H_{75}FN_{10}O_{10}$ + H = 1031.5730, found 1031.5725.



4.87

Bis-triazole 4.87 $[\alpha]^{23}_D$ +8.3 (c 1.12, $CHCl_3$); IR (neat) 2985, 2524, 2159, 2030, 1732, 1530, 1374, 1259, 1215, 1130, 1079, 999, 868, 765, 692; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.80 (s, 1H), 7.70 (s, 1H), 7.28 – 7.27 (m, 1H), 7.22 – 7.17 (m, 3H), 7.12 (dt, *J* = 7.6, 1.3 Hz, 1H), 6.69 – 6.62 (m, 2H), 4.87 (dd, *J* = 10.7, 2.4 Hz, 1H), 4.41 (tdd, *J* = 7.0, 3.5, 1.4 Hz, 4H), 4.29 (d, *J* = 7.3 Hz, 1H), 4.05 (dd, *J* = 10.8, 1.6 Hz, 1H), 3.83 – 3.70 (m, 2H), 3.63 (dt, *J* = 14.3, 7.7 Hz, 1H), 3.52 (ddt, *J* = 15.5, 11.5, 5.3 Hz, 2H), 3.43 (s, 1H), 3.20 (dd, *J* = 10.2, 7.2 Hz, 1H), 3.10 (q, *J* = 6.9 Hz, 1H), 2.58 (dd, *J* = 11.6, 7.0 Hz, 2H), 2.54 (s, 3H), 2.49 (d, *J* = 10.0 Hz, 1H), 2.39 – 2.29 (m, 1H), 2.21 (s, 3H), 2.04 – 1.90 (m, 5H), 1.86 (dd, *J* = 14.4, 2.8 Hz, 1H), 1.78 (d, *J* = 21.4 Hz, 3H), 1.73 – 1.56 (m, 3H), 1.51 (d, *J* = 7.5 Hz, 5H), 1.35 – 1.27 (m, 6H), 1.27 – 1.20 (m, 9H), 1.17 (d, *J* = 6.9 Hz, 4H), 1.00 (d, *J* = 7.0 Hz, 3H), 0.87 (t, *J* = 7.4 Hz, 4H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 216.7, 203.1, 202.8, 166.7, 166.5, 157.4, 148.2, 148.0, 147.1, 147.0, 131.9, 131.6, 129.9, 129.8, 119.9, 119.6, 116.3, 116.1, 115.1, 114.9, 112.5, 112.3, 104.3, 82.3, 81.0, 78.7, 70.4, 69.7,

66.1, 61.2, 50.1, 49.9, 49.4, 44.7, 42.9, 41.0, 39.7, 39.4, 37.0, 29.8, 29.5, 27.9, 27.8, 25.4, 25.2, 24.43, 22.3, 21.2, 19.9, 18.1, 15.2, 14.9, 13.92, 10.6; HRMS (ESI) calc'd for $C_{54}H_{77}FN_{10}O_{10}$ + H = 1045.5886, found 1045.5882.

BIBLIOGRAPHY

Agmon, I.; Amit, M.; Auerbach, T.; Bashan, A.; Baram, D.; Bartels, H.; Berisio, R.; Greenberg, I.; Harms, J.; Hansen, H. A. S.; Kessler, M.; Pyetan, E.; Schluenzen, F.; Sittner, A.; Yonath, A.; Zarivach, R. *FEBS Lett.* **2004**, *567*, 20–26.

Agouridas, C.; Denis, A.; Auger, J. M.; Benedetti, Y.; Bonnefoy, A.; Bretin, F.; Chantot, J. F.;

Dussarat, A.; Fromentin, C.; D'Ambrières, S. G.; Lachaud, S.; Laurin, P.; Le Martret, O.; Loyau, V.; Tessot, N. *J. Med. Chem.* **1998**, *41*, 4080–4100.

Allen, N. E. *Antimicrob. Agents Chemother.* **1977**, *11*, 669–674.

Arotsky, J.; Butler, R.; Darby, A. C. *J. Chem. Soc.* **1970**, 1480–1485.

Baker, W. R.; Clark, J. D.; Stephens, R. L.; Kim, K. H. *J. Org. Chem.* **1988**, *53*, 2340–2345.

Barber, J. *Magn. Reson. Chem.* **1991**, *29*, 740–743.

Bashan, A.; Agmon, I.; Zarivach, R.; Schluenzen, F.; Harms, J.; Berisio, R.; Bartels, H.; Franceschi, F.; Auerbach, T.; Hansen, H. A. S.; Kossoy, E.; Kessler, M.; Yonath, A. *Mol. Cell.* **2003**, *11*, 91–102.

Bender, D. A. E. and S. L. *Tetrahedron Lett.* **1986**, *27*, 799–802.

Bissantz, C.; Kuhn, B.; Stahl, M. *J. Med. Chem.* **2010**, *53*, 5061–5084.

Blair, J. M. A.; Webber, M. A.; Baylay, A. J.; Ogbolu, D. O.; Piddock, L. J. V. *Nat. Rev. Microbiol.* **2015**, *13*, 42–51.

Bondi, A. *J. Phys. Chem.* **1964**, *68*, 441–451.

Brockmann, H.; Henkel, W. *Naturwissenschaften.* **1951**, *37*, 284–288.

Brunet, E.; Muñoz, D. M.; Parra, F.; Mantecón, S.; Juanes, O.; Rodríguez-Ubis, J. C.; Carmen Cruzado, M.; Asensio, R. *Tetrahedron Lett.* **2007**, *48*, 1321–1324.

Celmer, W. D. *Pure Appl. Chem.* **1971**, *28*, 413–453.

Cope, C. A.; Ciganek, E. *Org. Synth.* **1959**, 4, 40–43.

Corey, E. J.; Kim, C. U. *J. Am. Chem. Soc.* **1972**, *94*, 7586–7587.

Cruikshank, A. J.; Cowper, G.; Grummitt, W. E. *Can. J. Chem.* **1956**, *34*, 214–219.

Davis, F. A.; Han, W. *Tetrahedron Lett*. **1991**, *32*, 1631–1634.

Davis, F. A.; Han, W.; Murphy, C. K. *J. Org. Chem.* **1995**, *60*, 4730–4737.

Davis, F. A.; Han, W. **1992**, *33*, 1153–1156.

Davis, F. A.; Kasu, P. V. N. *Tetrahedron Lett.* **1998**, *39*, 6135–6138.

Davidson, M. H.; McDonald, F. E. *Org. Lett.* **2004**, *6*, 1601–1603.

Denis, A.; Agouridas, C.; Auger, J.; Benedetti, Y.; Bonnefoy, A.; Bretin, F.; Chantot, J.; Dussarat, A.; Fromentin, C.; Gouin, S.; Ambri, D.; Lachaud, S.; Laurin, P.; Martret, O. Le; Loyau, V. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 3075–3080.

Dess, D. B.; Martin, J. C. *J. Org. Chem.* **1983**, *48*, 4155–4156.

DjokiC, S.; Kobrehel, G.; Lazaravski, G.; Lopotar, N.; Z. T. *J. Chem. Soc. Perkin. Trans. I.* **1986**, 1881–1886.

Dunitz, J. D.; Taylor, R. *Chem. - A Eur. J.* **1997**, *3*, 89–98.

Dunitz, J. D. *ChemBioChem.* **2004**, *5*, 614–621.

Evans, D. A.; Sjogren, E. B.; Bartroli, J.; Dow, R. L. *Tetrahedron Lett.* **1986**, *27*, 4957–4960.

Evans, D. A.; Ellman, J. A.; Dorow, R. L. *Tetrahedron Lett.* **1987**, *28*, 1123–1126.

Evans, D.; Bartroli, J.; Shih, T. L. *J. Am. Chem. Soc.* **1981**, *103*, 2109–2127.

Evans, D. A.; Rieger, D. L.; Gage, J. R. *Tetrahedron Lett.* **1990**, *31*, 7099–7100.

Evrard-Todeschi, N.; Gharbi-Benarous, J.; Gaillet, C.; Verdier, L.; Bertho, G.; Lang, C.; Parent, A.; Girault, J. P. *Bioorg. Med. Chem.* **2000**, *8*, 1579–1597.

Feldman, A. K.; Colasson, B.; Sharpless, K. B.; Fokin, V. V. *J. Am. Chem. Soc.* **2005**, *127*, 13444–13445.

Fernandes, P. B.; Baker, W. R.; Freiberg, L. A.; Hardy, D. J.; McDonald, E. J. *Antimicrob. Agents Chemother.* **1989**, *33*, 78–81.

Fernandes, P.; Martens, E.; Bertrand, D.; Pereira, D. *Bioorg. Med. Chem.* **2016**, *24*, 6420–6428.

Frantz, D. E.; Fässler, R.; Carreira, E. M. *J. Am. Chem. Soc.* **2000**, *122*, 1806–1807.

Freire, E. *Drug Discov Today.* **2008**, *13*, 869–874.

Fröhlich, R.; Rosen, C. T.; Meyer, Oliver, G. J.; Rissanen, K.; Haufe, G. *J. Mol. Struct.* **2006**, *787*, 50–62.

Fürst, A.; Plattner, P. A. *Helv. Chim. Acta* **1949**, *32*, 275–283.

Gardner, A. D.; Chain, E. *Br J Exp Path.* **1942**, *23*, 123–127.

Gagneuz, A.; Winstein, S.; Young, W. G. *J. Am. Chem. Soc.* **1960**, *82*, 5956–5957.

Glassford, I.; Lee, M.; Wagh, B.; Velvadapu, V.; Paul, T.; Sandelin, G.; DeBrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Amdrade, R. B. *ACS Med. Chem. Lett.* **2014**, *5*, 1021–1026.

Glassford, I.; Teijaro, C. N.; Daher, S. S.; Weil, A.; Small, M. C.; Redhu, S. K.; Colussi, D. J.; Jacobson, M. A.; Childers, W. E.; Buttaro, B.; Nicholson, A. W.; MacKerell, A. D.; Cooperman, B. S.; Andrade, R. B. *J. Am. Chem. Soc.* **2016**, *138*, 3136–3144.

Gingras, M. *Tetrahedron Lett.* **1991**, *32*, 7381–7384.

Gololobov, Y. G.; Zhmurova, I. N.; Kasukhin, L. F. *Tetrahedron* **1981**, *37*, 437–472.

González, Á.; Aiguadé, J.; Urpí, F.; Vilarrasa, J. *Tetrahedron Lett.* **1996**, *37*, 8949–8952.

Griesgraber, G.; Or, Y. S.; Chu, D. T. W.; Nilius, A. M.; Johnson, P. M.; Flamm, R. K.; Henry, R. F.; Plattner, J. J.; Road, A. P.; Park, A. **1995**, *49*, 465–477..

Griesgraber, G.; Kramer, M. J.; Elliott, R. L.; Nilius, A M.; Ewing, P. J.; Raney, P. M.; Bui, M.

H.; Flamm, R. K.; Chu, D. T.; Plattner, J. J.; Or, Y. S. *J. Med. Chem.* **1998**, *41*, 1660–1670.

Hanselmann, R.; Job, G. E.; Johnson, G.; Lou, R.; Martynow, J. G.; Reeve, M. M. *Org. Process Res. Dev.* **2010**, *14*, 152–158.

Hans Werner Bersch, G. H. *Arch. Pharm. (Weinheim).* **1956**, *289*, 673–676.

Henry, H.; Wignall, J. S. *J. Chem. Soc.* **1926**, 2077–2079.

Hofmann, V. A. W. *Ann. der Chemie und Pharm.* **1851**, *78*, 253–286.

Hudlicky, M. *Organic Reactions*. **1988**, 513–637.

Hudlicky, T.; Entwistle, D. A.; Pitzer, K. K.; Thorpe, A. J. *Chem. Rev.* **1996**, *96*, 1195–1220.

Huisgen, R. *Proc. Chem. Soc.* **1961**, 357–397.

Inanaga, J.; Hirata, K.; Saeki, H.; Katsuki, T.; Yamaguchi, M. *Bull. Chem. Soc. Jpn.* **1979**, 1989–1993.

Issleib, K. *Tetrahedron* **1984**, *18*, 261.

Jakopović, I. P.; Krajačić, M. B.; Škugor, M. M.; Štimac, V.; Pešić, D.; Vujasinović, I.; Alihodžić, S.; Paljetak, H. Č.; Kragol, G. *Bioorg. Med. Chem. Lett.* **2012**, *22*, 3527–3530.

Jeffery, T. *Tetrahedron Lett.* **1992**, *33*, 1989–1992.

Jelić, D.; Antolović, R. *Antibiotics* **2016**, *5*, 29.

Jones, P. H. Erythromycin ester derivatives, US 3598805 A. **1971**.

Jorgensen, W. L.; Bollini, M.; Thakur, V. V.; Domaoal, R. A.; Spasov, K. A.; Anderson, K. S. *J. Am. Chem. Soc.* **2011**, *133*, 15686–15696.

Kapić, S.; Ipić Paljetak, H.; Alihodić, S.; Antolović, R.; Eraković Haber, V.; Jarvest, R. L.; Holmes, D. J.; Broskey, J. P.; Hunt, E. *Bioorg. Med. Chem.* **2010**, *18*, 6569–6577.

Katz, L.; Ashley, G. W. *Chem. Rev.* **2005**, *105*, 499–527.

Kolb, H. C.; VanNieuwenhze, M. S.; Sharpless, K. B. *Chem. Rev.* **1994**, *94*, 2483–2547.

Kozikowski, A. P.; Fauq A. H.; Rusnak. J. M.; *Tetrahedron Lett.* **1989**, *30*, 3365–3368.

Kuethe, T. J.; Beutner, L. G. *Org. Synth.* **2009**, *86*, 92.

Kurath, P.; Jones, P. H.; Egan, R. S.; Perun, T. J. *Experientia.* **1971**, *27*, 362.

Lal, G. S.; Pez, G. P.; Pesaresi, R. J.; Prozonic, F. M.; Cheng, H. *J. Org. Chem.* **1999**, *64*, 7048–7054.

Lal, G. S.; Lobach, E.; Evans, A. *J. Org. Chem.* **2000**, *65*, 4830–4832.

LeMahieu, R. A.; Carson, M.; Kierstead, R. W.; Fern, L. M.; Grunberg, E. *J. Med. Chem.* **1974**, *17*, 953–956.

Letourneau, N.; Vimal, P.; Klepacki, D.; Mankin, A.; Melman, A. *Bioorg. Med. Chem. Lett.* **2012**, *22*, 4575–4578.

Liang, T.; Neumann, C. N.; Ritter, T. *Angew. Chem. Int. Ed.* **2013**, *52*, 8214–8264.

Liang, C. H.; Yao, S.; Chiu, Y. H.; Leung, P. Y.; Robert, N.; Seddon, J.; Sears, P.; Hwang, C. K.; Ichikawa, Y.; Romero, A. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 1307–1310.

Llano-Sotelo, B.; Dunkle, J.; Klepacki, D.; Zhang, W.; Fernandes, P.; Cate, J. H. D.; Mankin, A. S. *Antimicrob. Agents Chemother.* **2010**, *54*, 4961–4970.

Liang, J. H.; Wang, Y. Y.; Wang, H.; Li, X. L.; An, K.; Xu, Y. C.; Yao, G. W. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 2880–2883.

Lux, J.; Chan, M.; Elst, L. Vander; Schopf, E.; Mahmoud, E.; Laurent, S.; Almutairi, A. *J. Mater. Chem. B. Mater. Biol. Med.* **2013**, *1*, 6359–6364.

Magee, T. V.; Ripp, S. L.; Li, B.; Buzon, R. A.; Chupak, L.; Dougherty, T. J.; Finegan, S. M.;

Girard, D.; Hagen, A. E.; Falcone, M. J.; Farley, K. A.; Granskog, K.; Hardink, J. R.; Huband, M. D.; Kamicker, B. J.; Kaneko, T.; Knickerbocker, M. J.; Liras, J. L.; Marra, A.; Medina, I.; Nguyen, T. T.; Noe, M. C.; Obach, R. S.; O'Donnell, J. P.; Penzien, J. B.; Reilly, U. D.; Schafer, J. R.; Shen, Y.; Stone, G. G.; Strelevitz, T. J.; Sun, J.; Tait-Kamradt, A.; Vaz, A. D. N.; Whipple, D. A.; Widlicka, D. W.; Wishka, D. G.; Wolkowski, J. P.; Flanagan, M. E. *J. Med. Chem.* **2009**, *52*, 7446–7457.

Markovskij, L.N.; Pashinnik, V. E.; Kirsanov, A. V. *Synthesis.* **1973**, 787–789.

Martínez, A. G.; Osío, J. B.; Rys, A. Z.; Subramanian, L. R. *Synlett* **1993**, 587–588.

Middleton, W. J. *J. Org. Chem.* **1975**, *40*, 574–578.

Moissan, H. *Compt. Rend.* **1886**, *102*, 1534.

Moraski, G. C.; Thanassi, J. A.; Podos, S. D.; Pucci, M. J.; Miller, M. J. *J. Antibiot.* **2011**, *64*, 667–671.

Morimoto, S.; Takahashi, Y.; Watanabe, Y. S. O. *J. Antibiot. (Tokyo).* **1984**, *37*, 187–189.

Morimoto, S.; Moriguchih, I. *Chem. Pharm. Bull.* **1994**, *42*, 1088–1095.

Moriwake, T.; Hamano, C., S.; Miki, D.; Saito, S. *Chem. Lett.* **1986**, 815–818.

Müller, K.; Faeh, C.; Diederich, F. *Science.* **2007**, *317*, 1881–1886.

Myers, A. G.; Mckinstry, L.; Barbay, J. K.; Gleason, J. L. *Tetrahedron Lett.* **1998**, *39*.

Myers, A. G.; Barbay, J. K.; Zhong, B. *J. Am. Chem. Soc.* **2001**, *123*, 7207–7219.

Nam, G.; Kang, T. W.; Shin, J. H.; Choi, K. Il. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 569–572.

Newman,.H. *J. Org. Chem.* **1964**, *29*, 1461–1468.

Newth, H. F. *Q. Rev. Chem. Soc* **1959**, *13*, 30–47.

Nie, J.; Zhu, H. W.; Cui, H. F.; Hua, M. Q.; Ma, J. A. *Org. Lett.* **2007**, *9*, 3053–3056.

O'Hagan, D.; Rzepa, H. S. *Chem. Commun.* **1997**, 7, 645–652.

O'Hagan, D. *Chem. Soc. Rev.* **2008**, *37*, 308–319.

Olsen, J. A.; Banner, D. W.; Seiler, P.; Sander, U. O.; D'Arcy, A.; Stihle, M.; Müller, K.;

Diederich, F. *Angew. Chemie. Int. Ed.* **2003**, *42*, 2507–2511.

Olsen, J. A.; Banner, D. W.; Seiler, P.; Wagner, B.; Tschopp, T.; Obst-Sander, U.; Kansy, M.; Müller, K.; Diederich, F. *ChemBioChem* **2004**, *5*, 666–675.

Olsen, J.; Seiler, P.; Wagner, B.; Fischer, H.; Tschopp, T.; Obst-Sander, U.; Banner, D. W.; Kansy, M.; Müller, K.; Diederich, F. *Org. Biomol. Chem.* **2004**, *2*, 1339–1352.

Omura, K.; Swern, D. *Tetrahedron* **1978**, *34*, 1651–1660.

Pridgen, L. N.; Abdel-Magid, A. F.; Lantos, I.; Shilcrat, S.; Eggleston, D. S. *J. Org. Chem.* **1993**, *58*, 5107–5117.

Ramana, M. M. V; Malik, S. S.; Parihar, J. A. *Tetrahedron Lett.* **2004**, *45*, 8681–8683.

Rodvold, K. a. *Clin. Pharmacokinet.* **1999**, *37*, 385–398.

Richardson, C. A. *J. Chem. Soc.* **1964**, 5364–5370.

Rosenbrook, W.; Riley, D. A.; Lartey, P. A. *Tetrahedron Lett.* **1985**, *26*, 3–4.

Ross, D. B. *N. Engl. J. Med.* **2007**, *356*, 1601–1604.

Rostovtsev, V. V.; Green, L. G.; Fokin, V. V.; Sharpless, K. B. *Angew. Chemie - Int. Ed.* **2002**, *41*, 2596–2599.

Rychnovsky, D. S.; Griesgraber, G.; Schlegel, R. *J. Am. Chem. Soc.* **1994**, *116*, 2623–2624.

Schlünzen, F.; Zarivach, R.; Harms, J.; Bashan, A.; Tocilj, A.; Albrecht, R.; Yonath, A.; Franceschi, F. *Nature* **2001**, *413*, 814–821.

Shain, C. S.; Amsden, G. W. *Ann. Pharmacother.* **2002**, *36*, 452–464.

Shiina, I.; Kubota, M.; Ibuka, R. *Tetrahedron Lett.* **2002**, *43*, 7535–7539.

Singh, R. P.; Twamley, B.; Shreeve, J. M. *J. Org. Chem.* **2002**, *67*, 1918–1924.

Sonogashira, K. *J. Organomet. Chem.* **2002**, *653*, 46–49.

Smart, B. E. *J. Fluor. Chem.* **2001**, *109*, 3–11.

Smith, A. B.; Mesaros, E. F.; Meyer, E. A. *J. Am. Chem. Soc.* **2006**, *128*, 5292–5299.

Stace, B. C. S.; G. J. *J. Chem. Soc.* **1948**, 1773–1779.

Tietze, L. F.; Haqtiel, U. *Tetrahedron Lett.* **1990**, *31*, 1697–1700.

Tosatti, P.; Horn, J.; Campbell, A. J.; House, D.; Nelson, A.; Marsden, S. P. *Adv. Synth. Catal.* **2010**, *352*, 3153–3157.

Tu, D.; Blaha, G.; Moore, P. B.; Steitz, T. A. *Cell*. **2005**, *121*, 257–270.

Umemoto, T.; Tomizawa, G. *Bull. Chem. Soc. Jpn*. 1986, 3625–3629.

Umemoto, T.; Singh, R. P.; Xu, Y.; Saito, N. *J. Am. Chem. Soc.* **2010**, *132*, 18199–18205.

Valeur, E.; Bradley, M. *Chem. Soc. Rev.* **2009**, *38*, 606–631.

VanRheenen, V.; Kelly, R. C.; Cha, D. Y. *Tetrahedron Lett.* **1976**, *17*, 1973–1976.

Velvadapu, V.; Andrade, R. B. *Carb.res.* **2008**, *343*, 145–150.

Velvadapu, V.; Paul, T.; Wagh, B.; Glassford, I.; Debrosse, C.; Andrade, R. B. *J. Org. Chem.* **2011**, *76*, 7516–7527.

Velvadapu, V.; Glassford, I.; Lee, M.; Paul, T.; Debrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2012**, *3*, 211–215.

Venkatraman, L.; Aldrich, C. C.; Sherman, D. H.; Fecik, R. a. *J. Org. Chem.* **2005**, *70*, 7267–7272.

Vujasinović, I.; Marušić Ištuk, Z.; Kapić, S.; Bukvić Krajačić, M.; Hutinec, A.; Dilović, I.; Matković-Čalogović, D.; Kragol, G. *European J. Org. Chem.* **2011**, *13*, 2507–2518.

Wagh, B.; Paul, T.; Glassford, I.; Debrosse, C.; Klepacki, D.; Small, M. C.; MacKerell, A. D.; Andrade, R. B. *ACS Med. Chem. Lett.* **2012**, *3*, 1013–1018.

Weisblum, B. *Antimicrob. Agents Chemother.* **1995**, *39*, 577–585.

White, J. M.; Tunoori, A. R.; Turunen, B. J.; Georg, G. I. *J. Org. Chem.* **2004**, *69*, 2573–2576.

Woodward, R. B. *Angew. Chem.* **1957**, *69*, 50–58.

Xuan, R.; Oh, H. S.; Lee, Y.; Kang, H. Y. *J. Org. Chem.* **2008**, *73*, 1456–1461.

Zhang, Z.; Fukuzaki, T.; Myers, A. G. *Angew. Chemie - Int. Ed.* **2016**, *55*, 523–527.

Zhou, C.; Li, J.; Lu, B.; Fu, C.; Ma, S. *Org. Lett.* **2008**, *10*, 581–583.

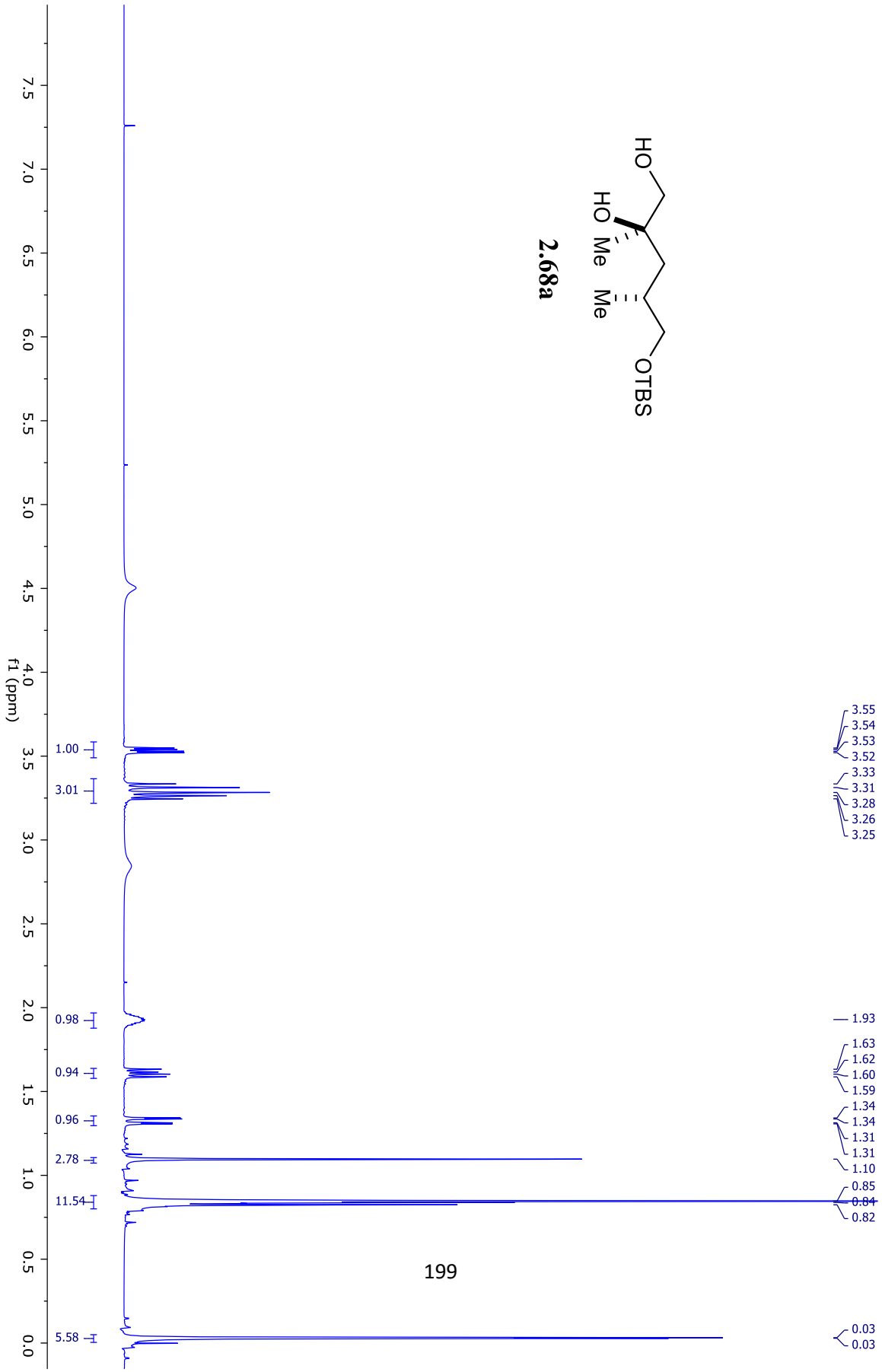
Zhu, D.; Xu, Y.; Liu, Y.; Chen, X.; Zhao, Z.; Lei, P. *Bioorg. Med. Chem. Lett.* **2013**, *23*, 6274–6279.

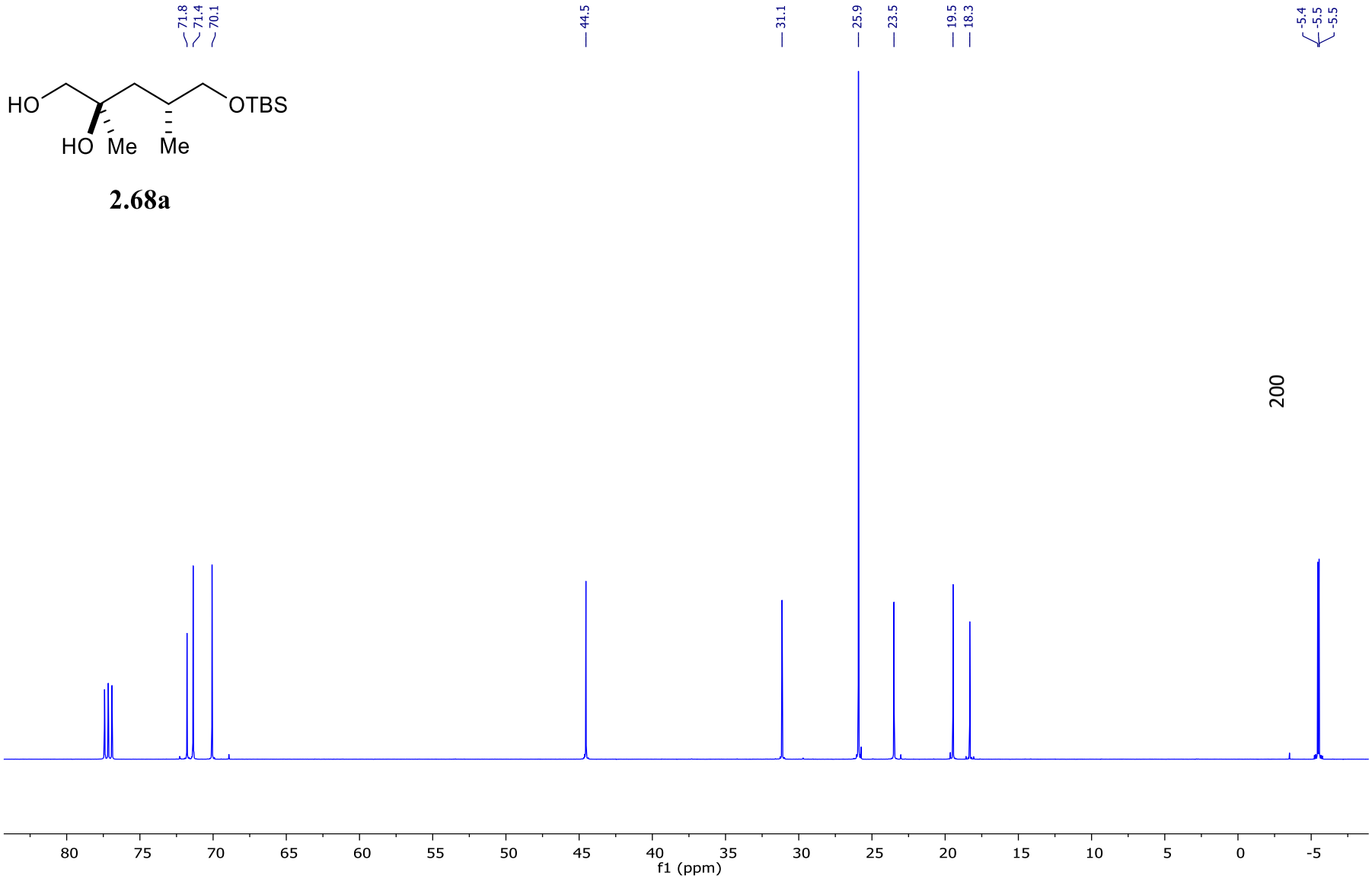
Zimmerman, H. E. *Acc. Chem. Res* **1987**, *20*, 263–268.

APPENDIX A1

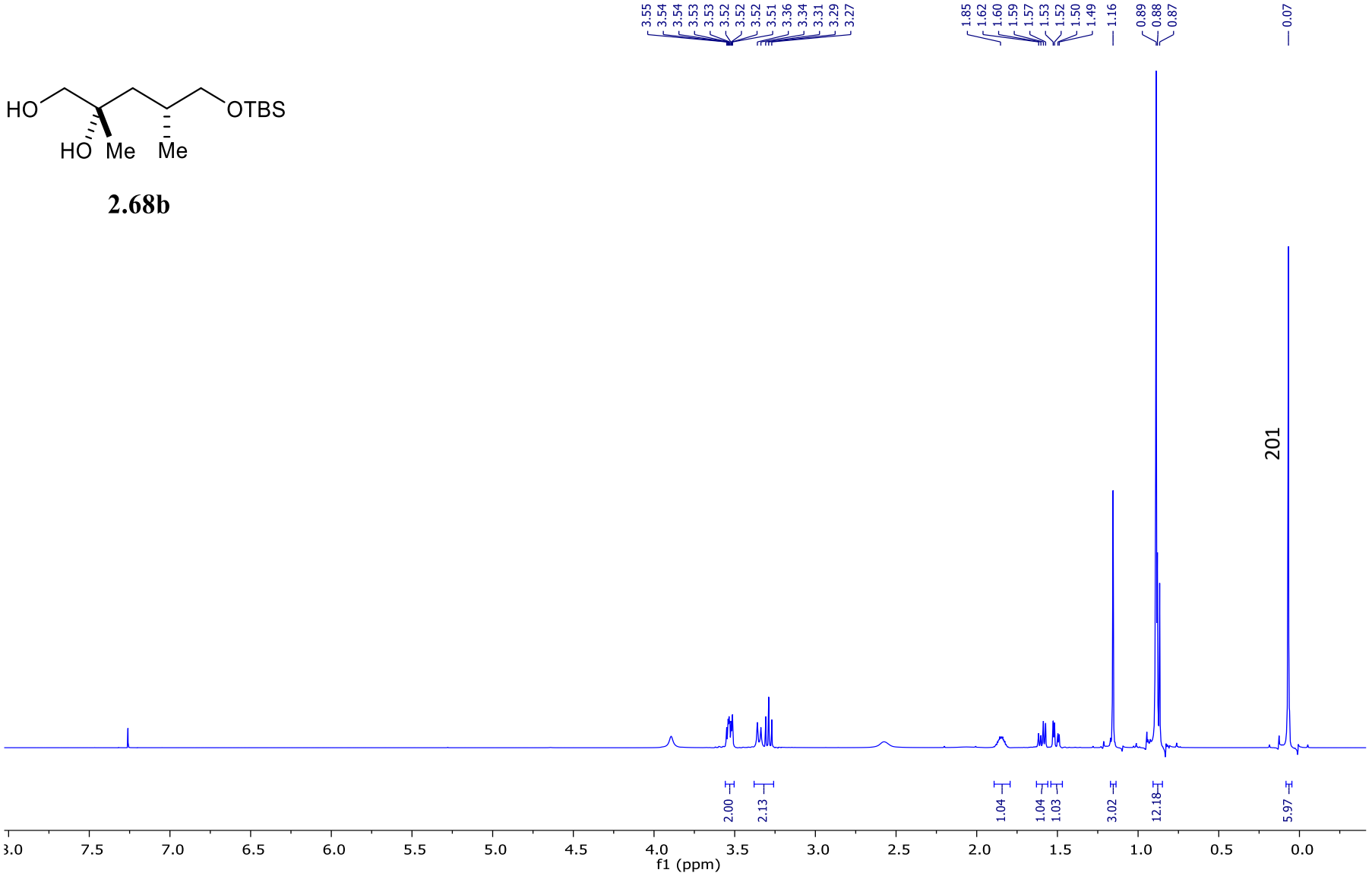
NMR SPECTRA OF COMPOUNDS IN

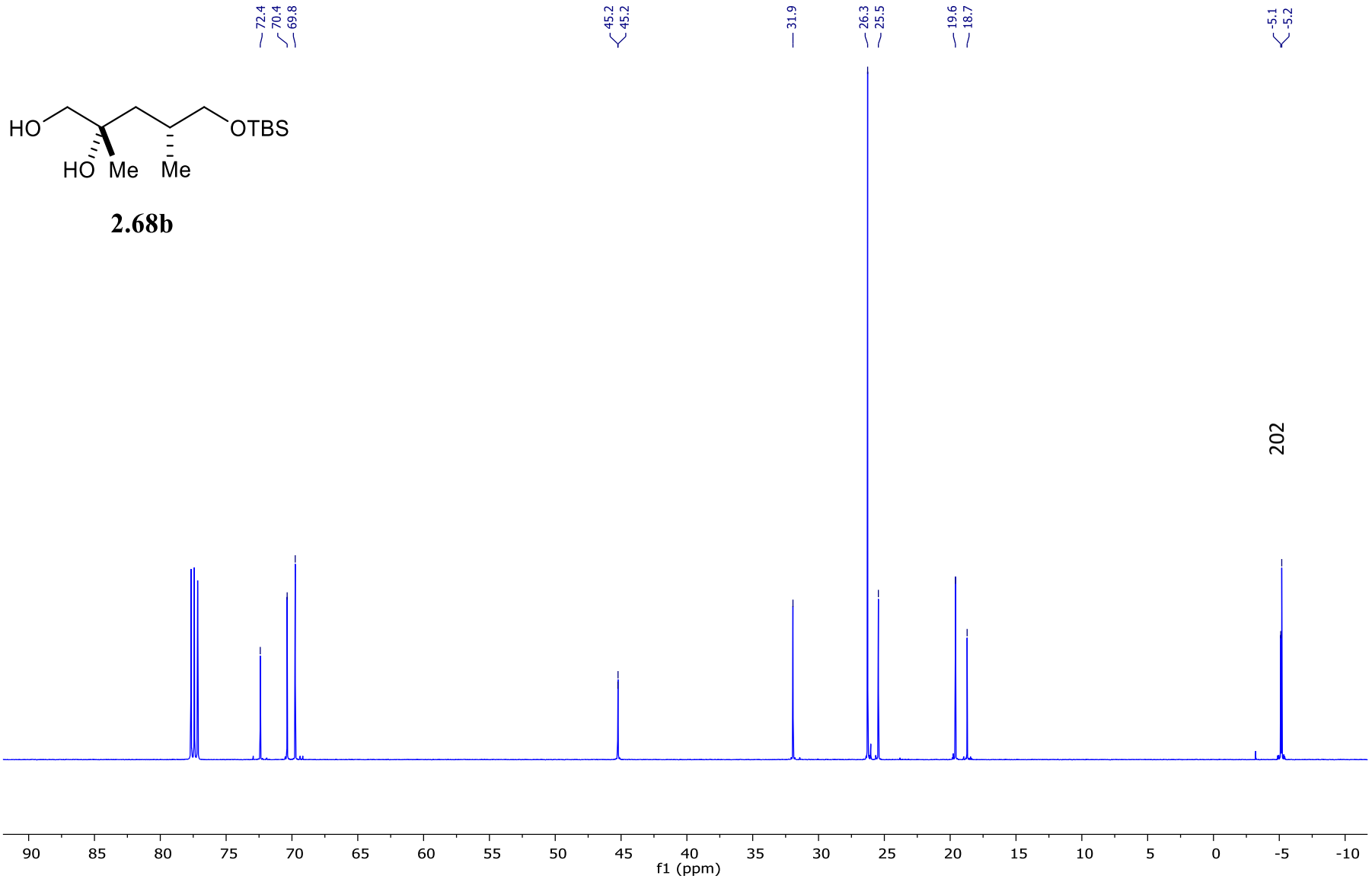
CHAPTER 2

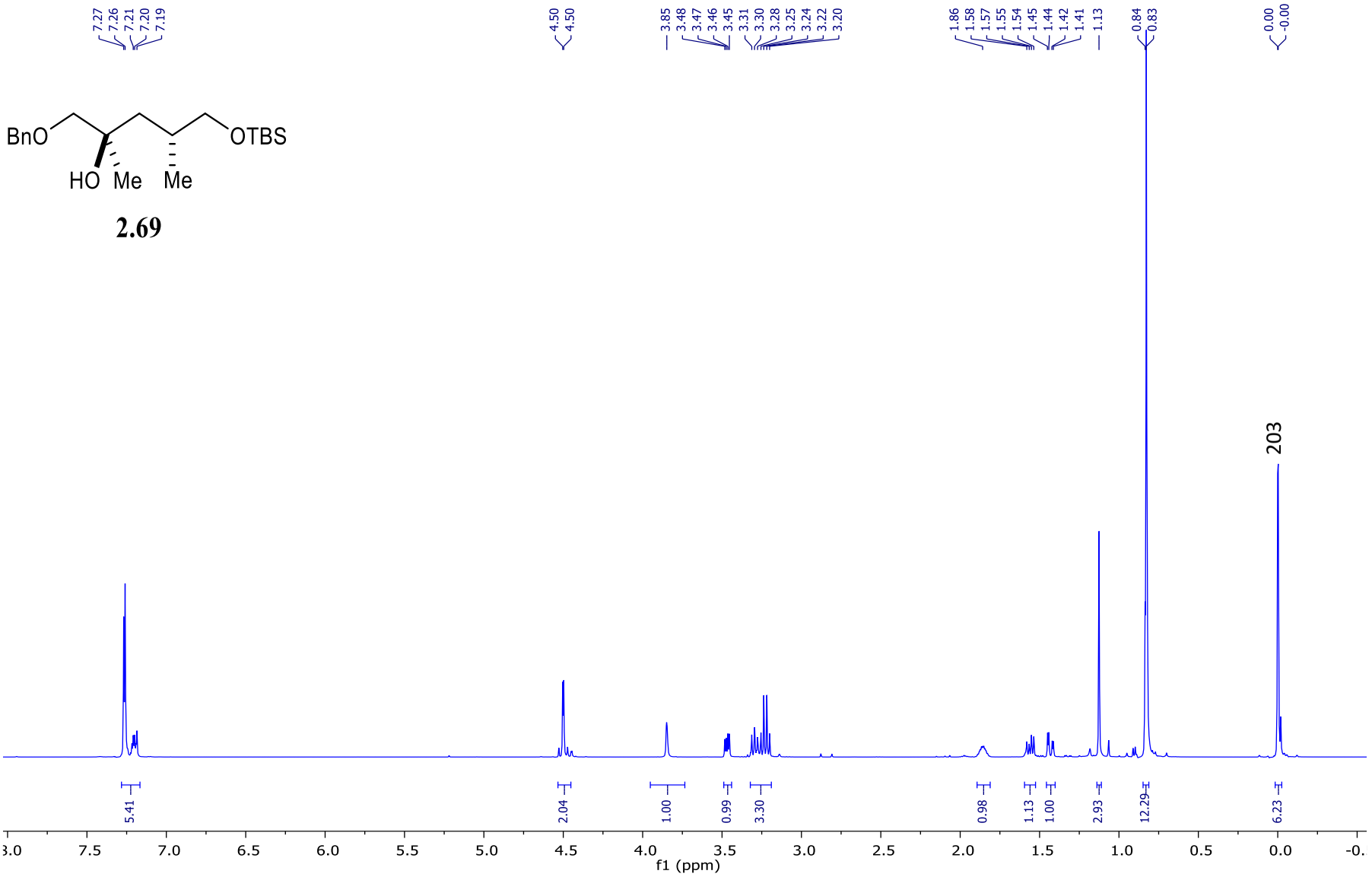

HO
HO
Me
Me
OTBS
2.68a
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
1.00
3.01
0.98
0.94
0.96
2.78
11.54
5.58
3.55
3.54
3.53
3.52
3.33
3.31
3.28
3.26
3.25
1.93
1.63
1.62
1.60
1.59
1.34
1.34
1.31
1.31
1.10
0.85
0.84
0.82
0.03
0.03


71.8
71.4
70.1
44.5
31.1
25.9
23.5
19.5
18.3
-5.4
-5.5
-5.5
HO
OTBS
HO
Me
Me
200
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
-5
f1 (ppm)

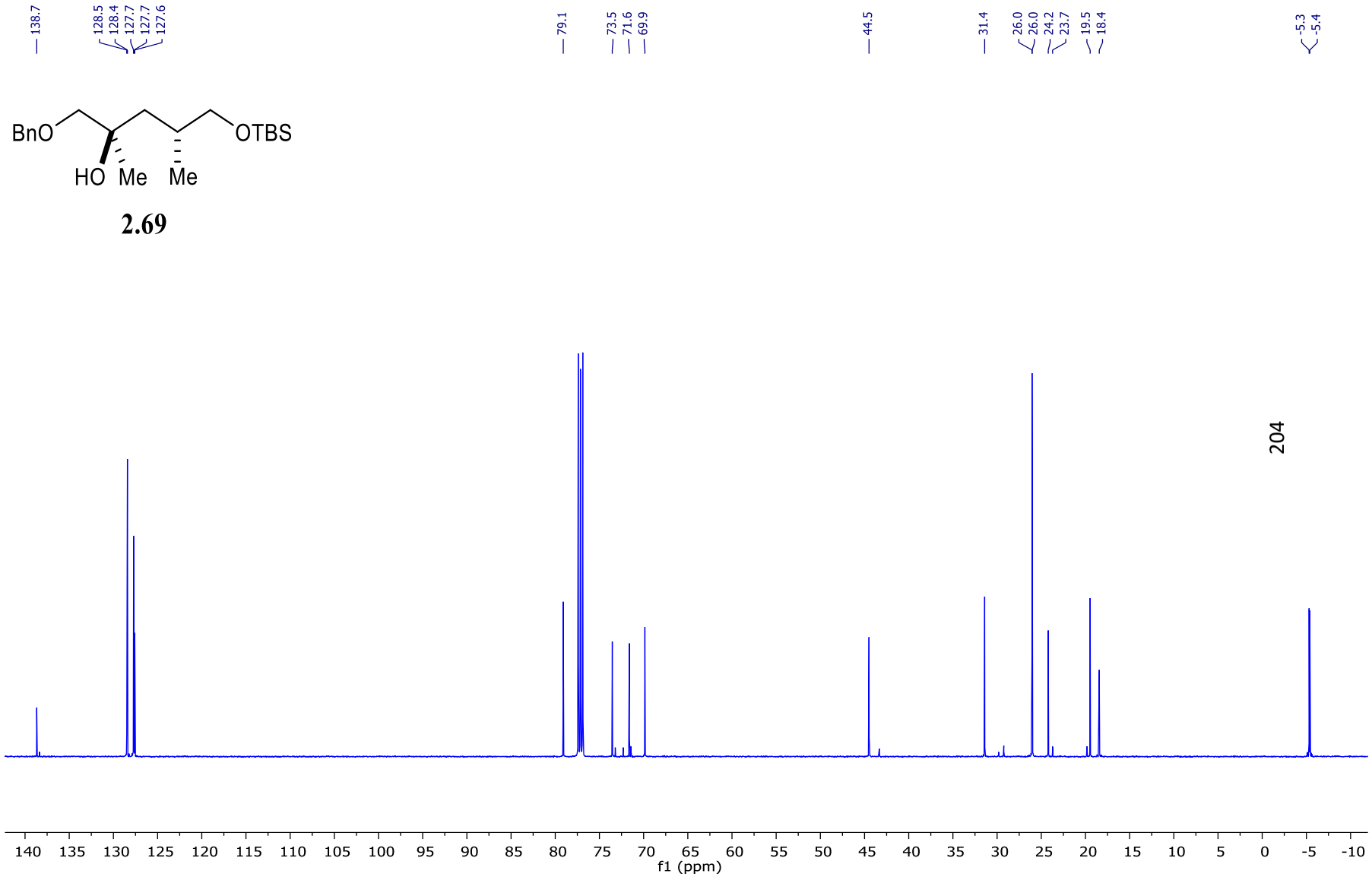
2.68a

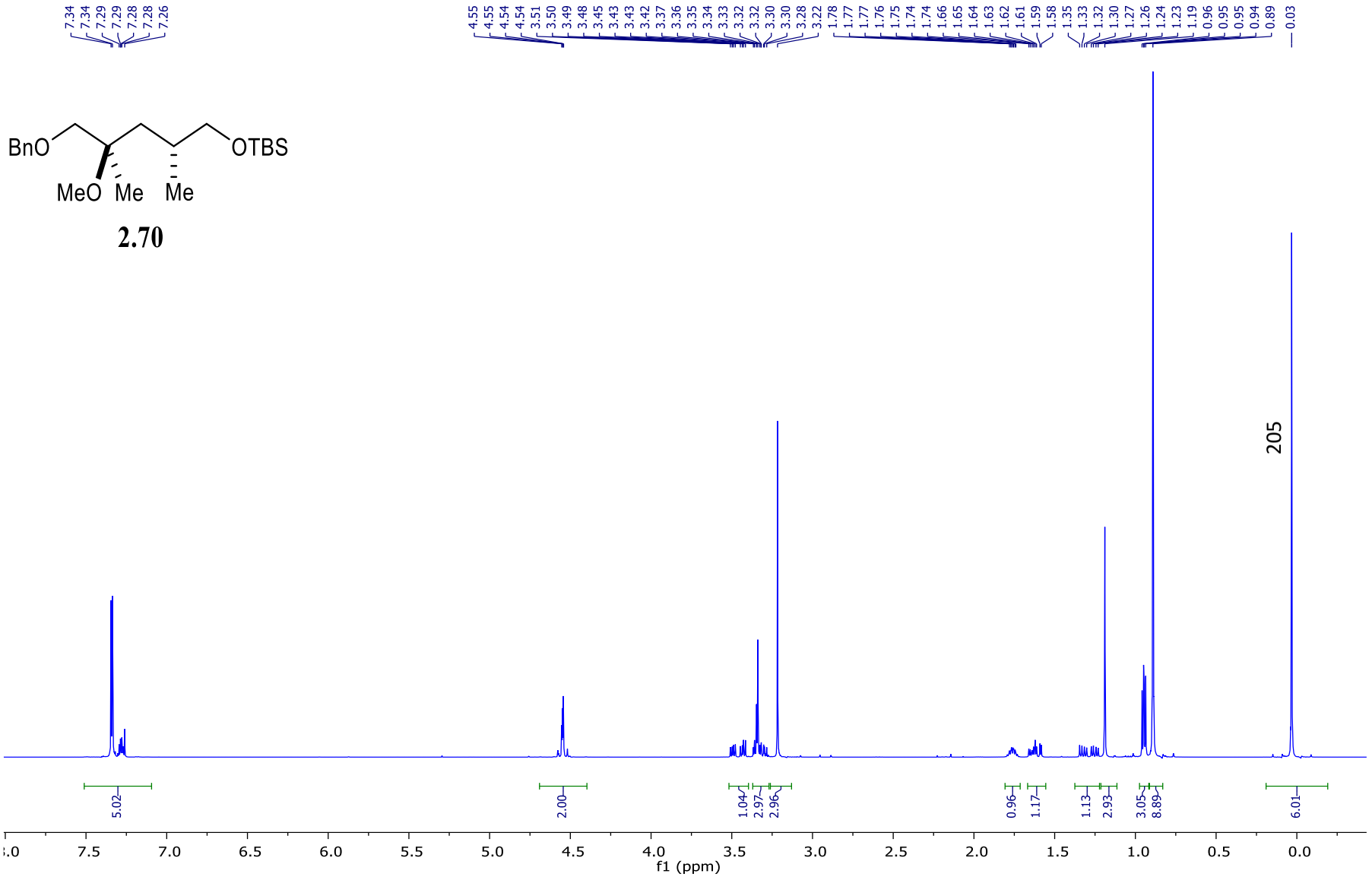

HO
OTBS
HO
Me
Me
2.68b
3.55
3.54
3.54
3.53
3.53
3.52
3.52
3.52
3.51
3.36
3.34
3.31
3.29
3.27
1.85
1.62
1.60
1.59
1.57
1.53
1.52
1.50
1.49
1.16
0.89
0.88
0.87
0.07
201
2.00
2.13
1.04
1.04
1.03
3.02
12.18
5.97
f1 (ppm)

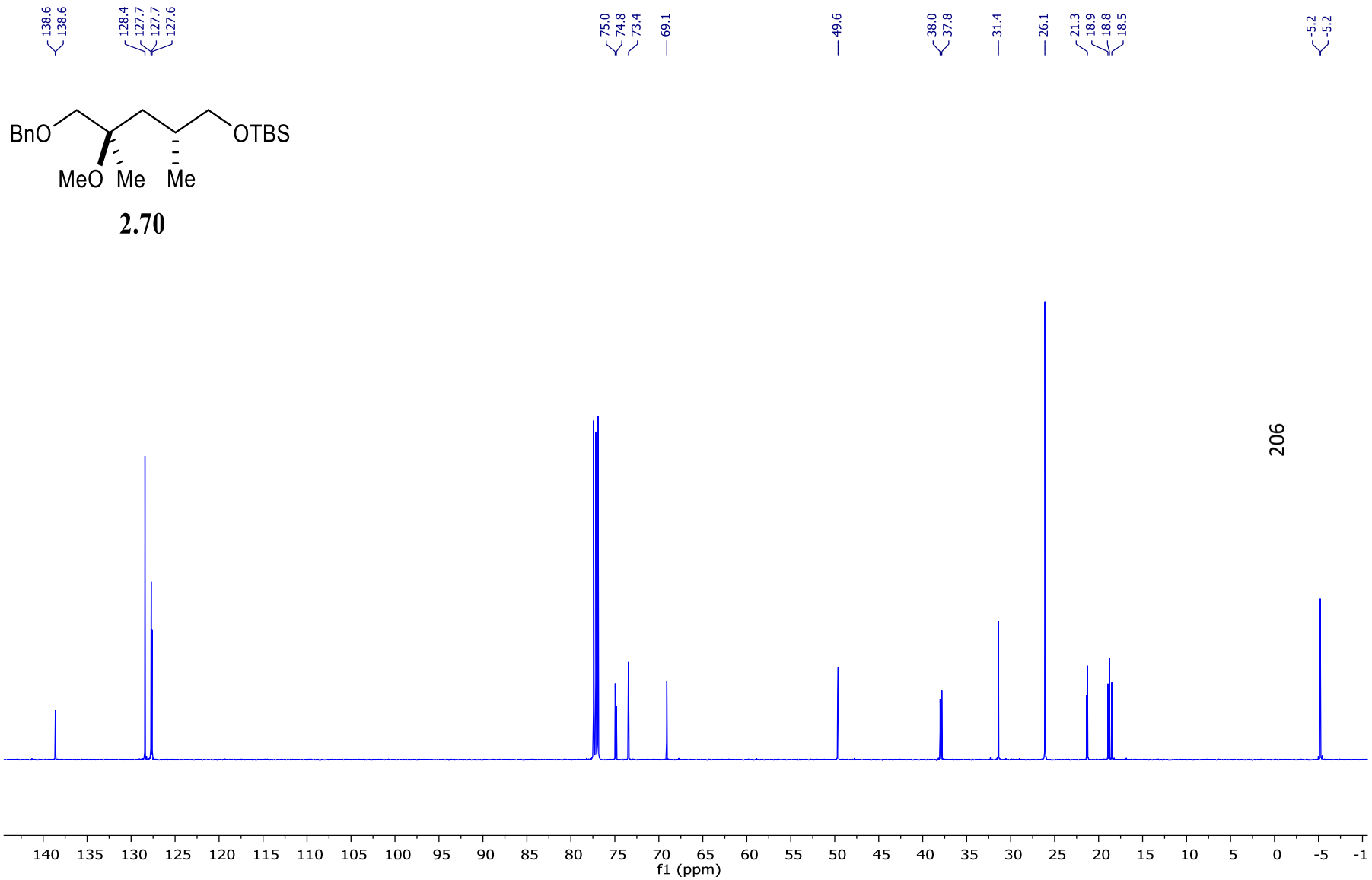

72.4
70.4
69.8
45.2
45.2
31.9
26.3
25.5
19.6
18.7
-5.1
-5.2
HO
OTBS
HO
Me
Me
2.68b
202
90
85
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
-5
-10
f1 (ppm)


BnO
OTBS
HO
Me
Me
7.27
7.26
7.21
7.20
7.19
4.50
4.50
3.85
3.48
3.47
3.46
3.45
3.31
3.30
3.28
3.25
3.24
3.22
3.20
1.86
1.58
1.57
1.55
1.54
1.45
1.44
1.42
1.41
1.13
0.84
0.83
0.00
-0.00
203
5.41
2.04
1.00
0.99
3.30
0.98
1.13
1.00
2.93
12.29
6.23
f1 (ppm)

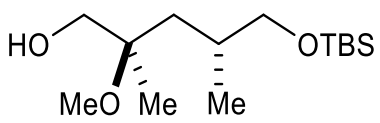
2.69


138.7
128.5
128.4
127.7
127.7
127.6
79.1
73.5
71.6
69.9
44.5
31.4
26.0
26.0
24.2
23.7
19.5
18.4
-5.3
-5.4
BnO
OTBS
HO
Me
Me
2.69
204
140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40 35 30 25 20 15 10 5 0 -5 -10
f1 (ppm)


BnO
OTBS
MeO
Me
Me
2.70
f1 (ppm)

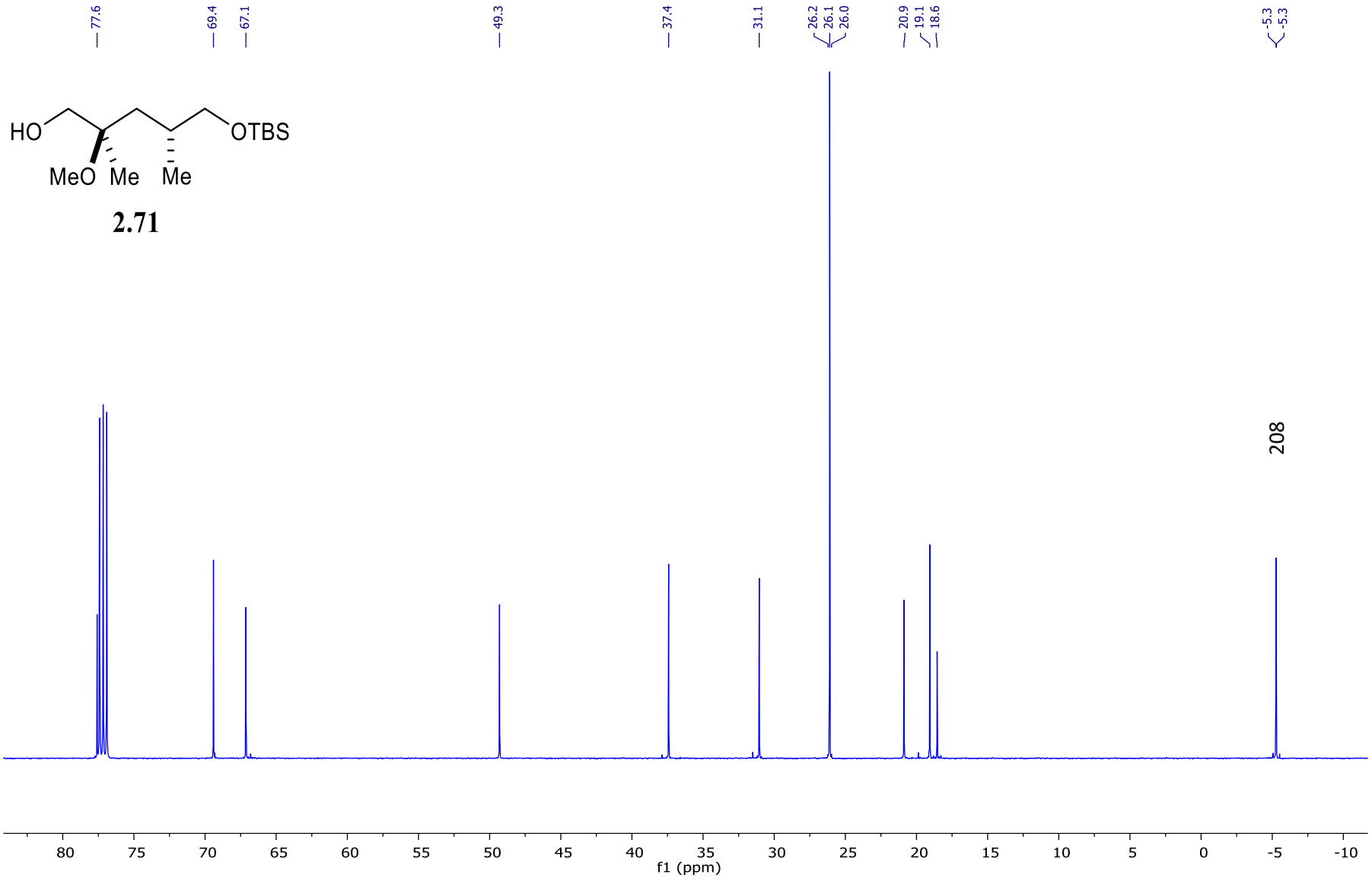

138.6
138.6
128.4
127.7
127.7
127.6
75.0
74.8
73.4
69.1
49.6
38.0
37.8
31.4
26.1
21.3
18.9
18.8
18.5
-5.2
-5.2
BnO
OTBS
MeO
Me
Me
206
140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40 35 30 25 20 15 10 5 0 -5 -1
f1 (ppm)

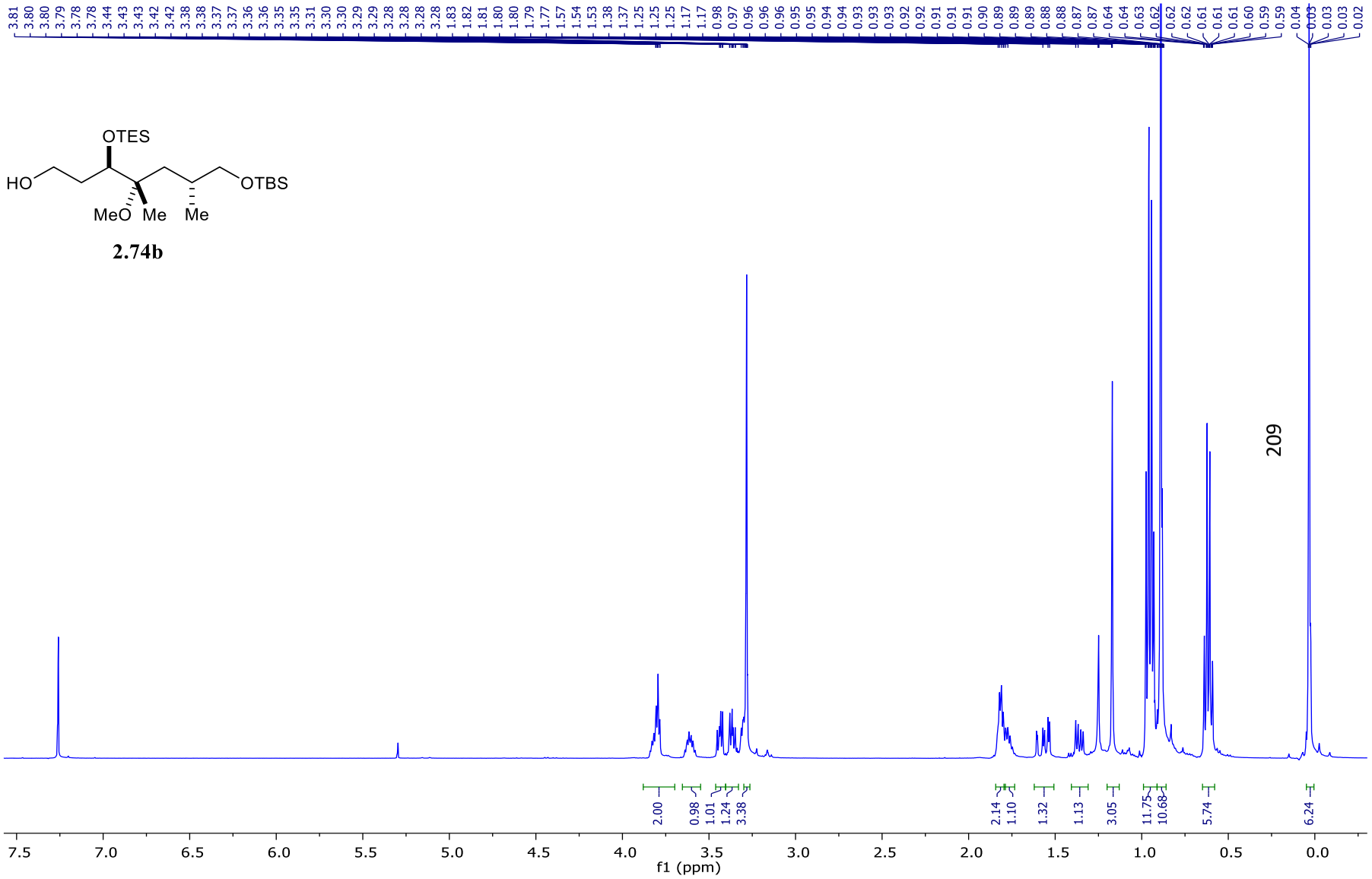
2.70

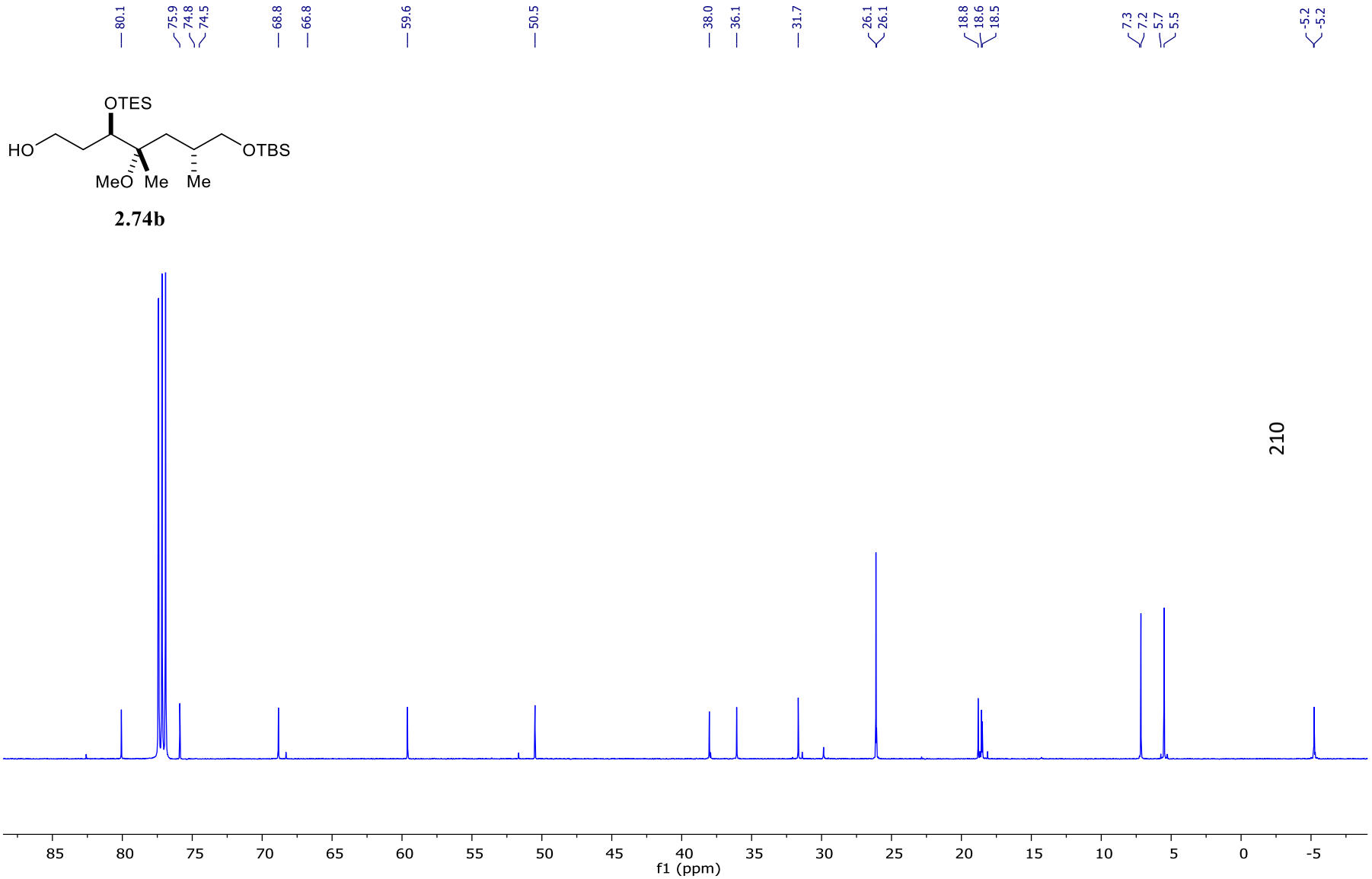

HO
OTBS
MeO
Me
Me

2.71

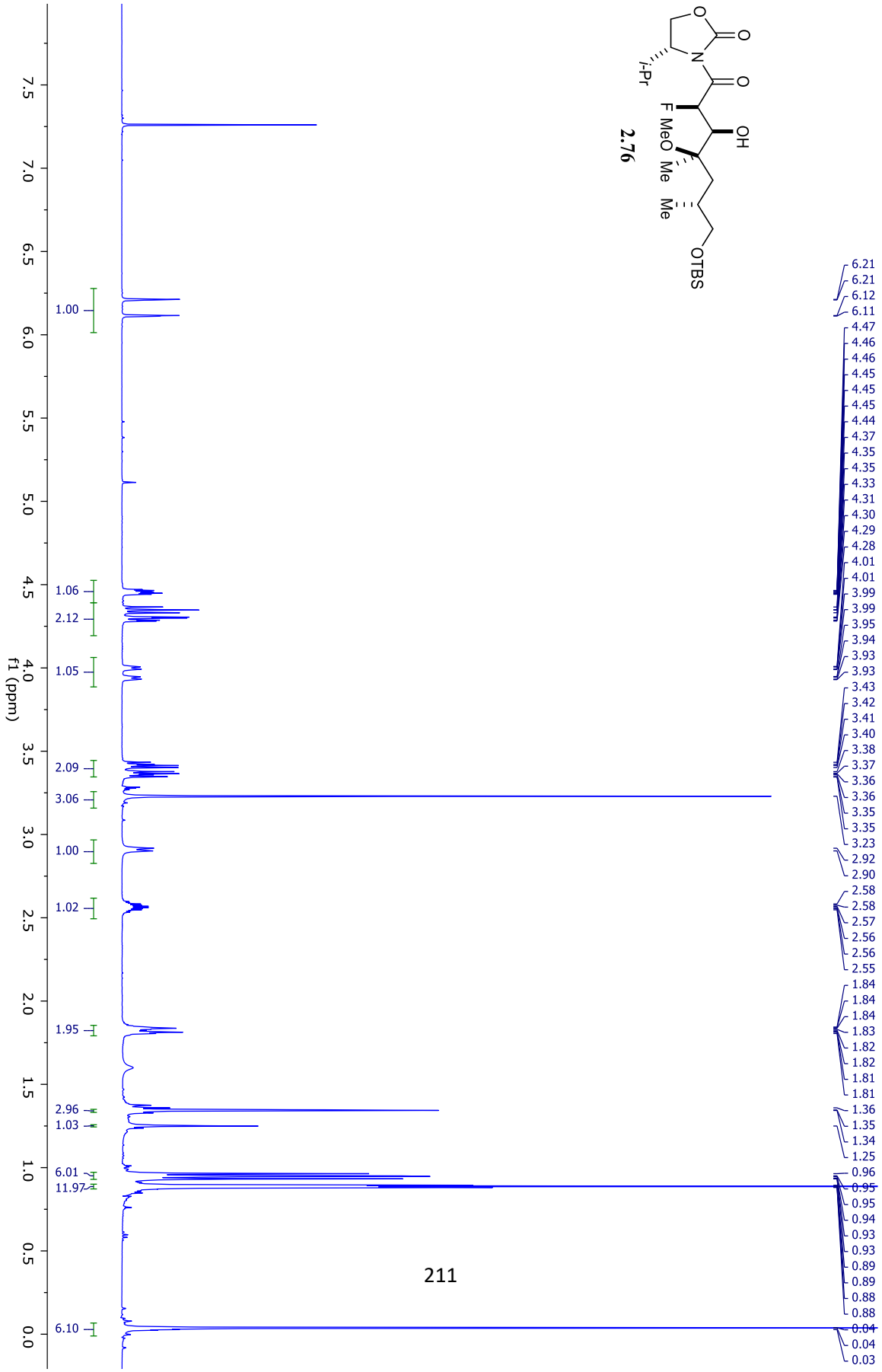
3.49 3.48 3.47 3.46 3.44 3.43 3.42 3.42 3.40 3.36 3.35 3.34 3.33 3.20 3.20 2.51 1.79 1.79 1.79 1.79 1.78 1.78 1.78 1.77 1.77 1.77 1.77 1.76 1.76 1.76 1.76 1.69 1.69 1.66 1.66 1.28 1.27 1.26 1.24 1.16 0.93 0.92 0.90 0.89 0.89 0.06 0.06 0.05 0.04 0.04 0.04
207
1.08 2.10 1.09 2.96 1.00 1.04 1.15 1.18 3.09 3.11 9.04 5.99
7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)


77.6
69.4
67.1
49.3
37.4
31.1
26.2
26.1
26.0
20.9
19.1
18.6
-5.3
-5.3
HO
OTBS
MeO
Me
Me
2.71
208
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
-5
-10
f1 (ppm)


OTES
HO
OTBS
MeO
Me
Me
2.74b
209
2.00
0.98
1.01
1.24
3.38
2.14
1.10
1.32
1.13
3.05
11.75
10.68
5.74
6.24
f1 (ppm)

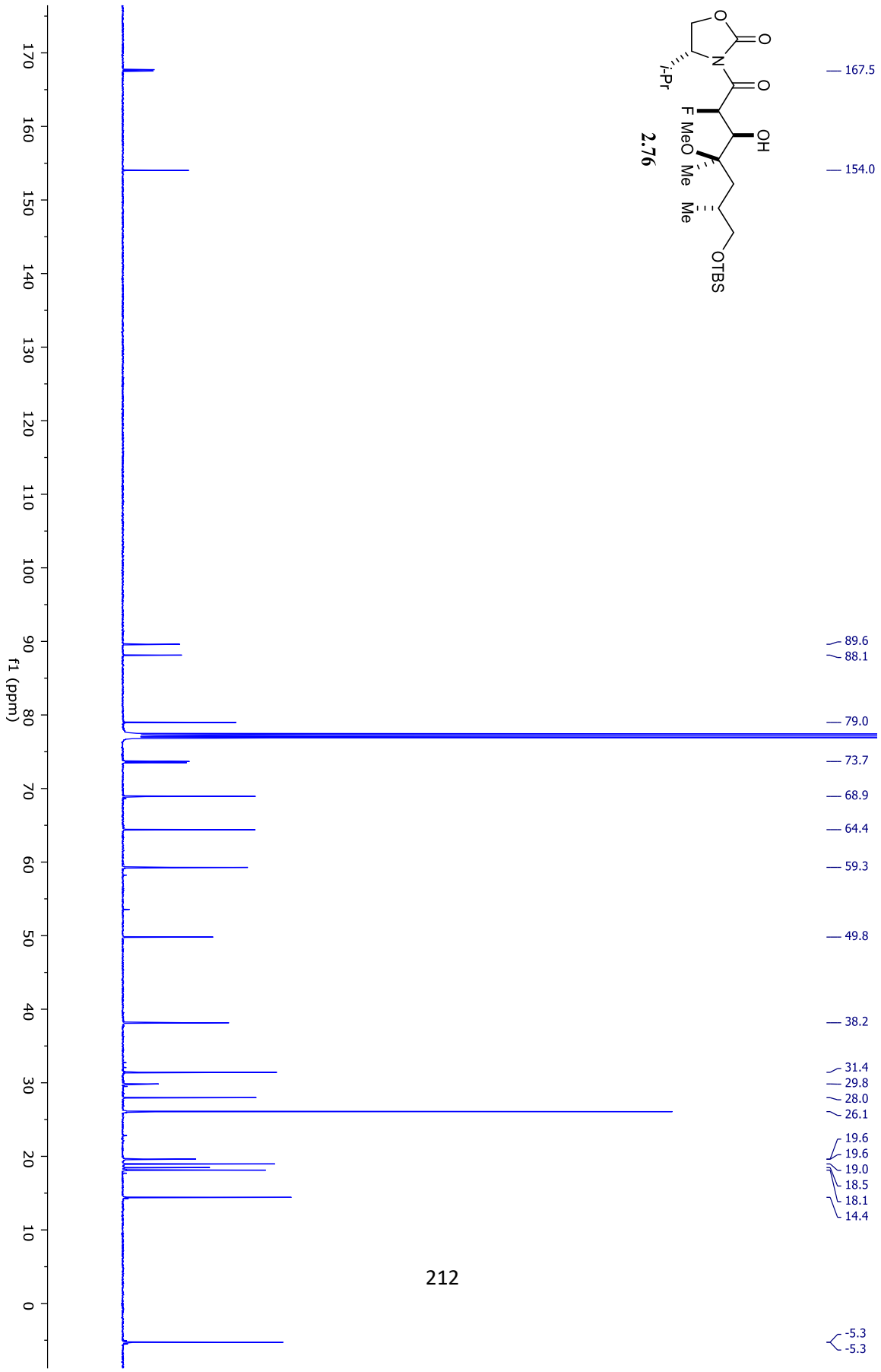

80.1
75.9
74.8
74.5
68.8
66.8
59.6
50.5
38.0
36.1
31.7
26.1
26.1
18.8
18.6
18.5
7.3
7.2
5.7
5.5
-5.2
-5.2
OTES
HO
OTBS
MeO
Me
Me
210
85
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
-5
f1 (ppm)

2.74b

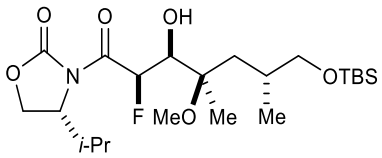

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
1.00
1.06
2.12
1.05
2.09
3.06
1.00
1.02
1.95
2.96
1.03
6.01
11.97
6.10
6.21
6.21
6.12
6.11
4.47
4.46
4.46
4.45
4.45
4.45
4.44
4.37
4.35
4.35
4.33
4.31
4.30
4.29
4.28
4.01
4.01
3.99
3.99
3.95
3.94
3.93
3.93
3.43
3.42
3.41
3.40
3.38
3.37
3.36
3.36
3.35
3.35
3.23
2.92
2.90
2.58
2.58
2.57
2.56
2.56
2.55
1.84
1.84
1.84
1.83
1.82
1.82
1.81
1.81
1.36
1.35
1.34
1.25
0.96
0.95
0.95
0.94
0.93
0.93
0.89
0.89
0.88
0.88
0.04
0.04
0.03

O
O
N
O
i-Pr
O
OH
F
MeO
Me
Me
OTBS

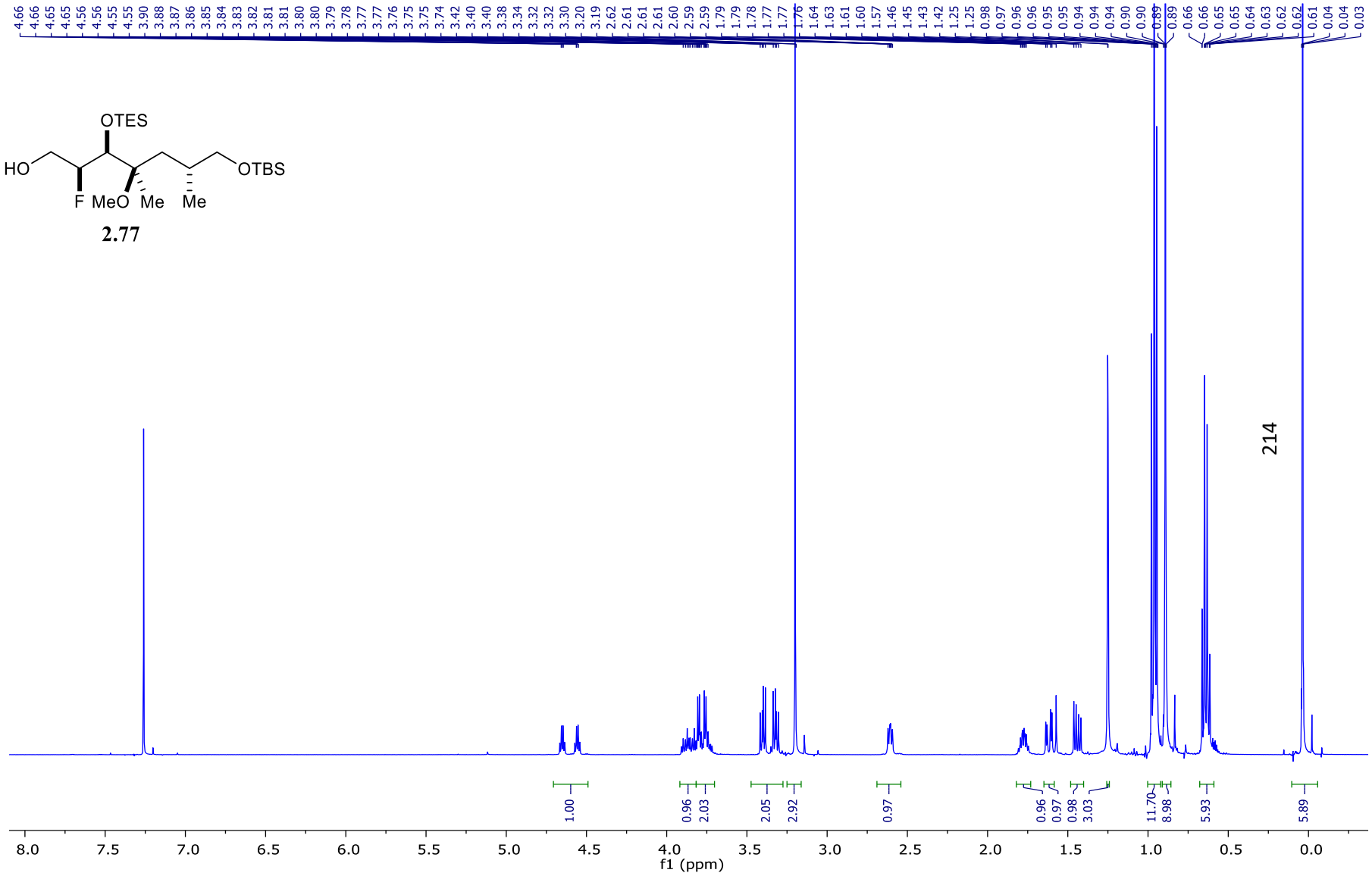
2.76

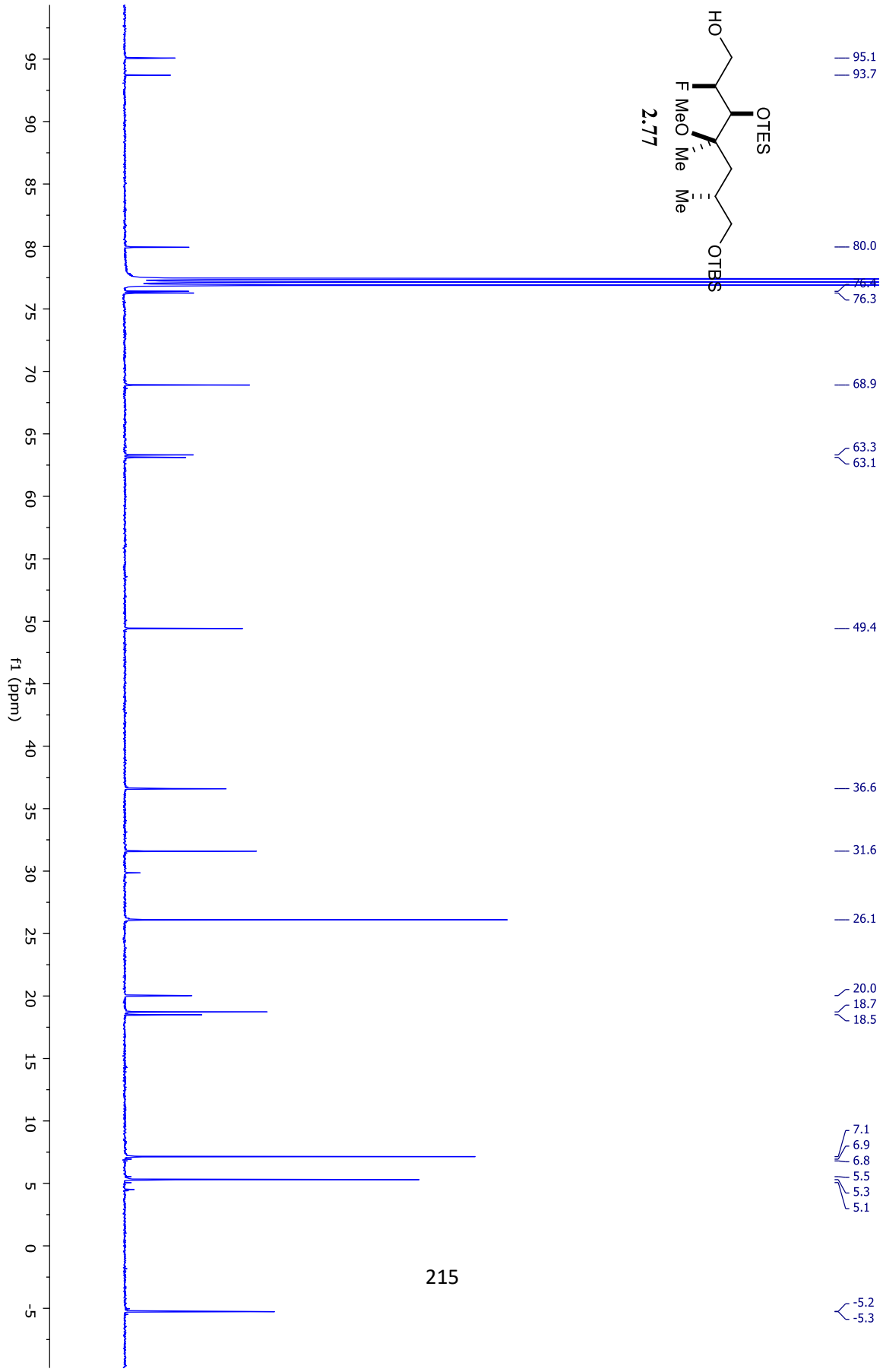

f1 (ppm)
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
i-Pr
F MeO Me Me
OTBS
OH
2.76
167.5
154.0
89.6
88.1
79.0
73.7
68.9
64.4
59.3
49.8
38.2
31.4
29.8
28.0
26.1
19.6
19.6
19.0
18.5
18.1
14.4
-5.3
-5.3

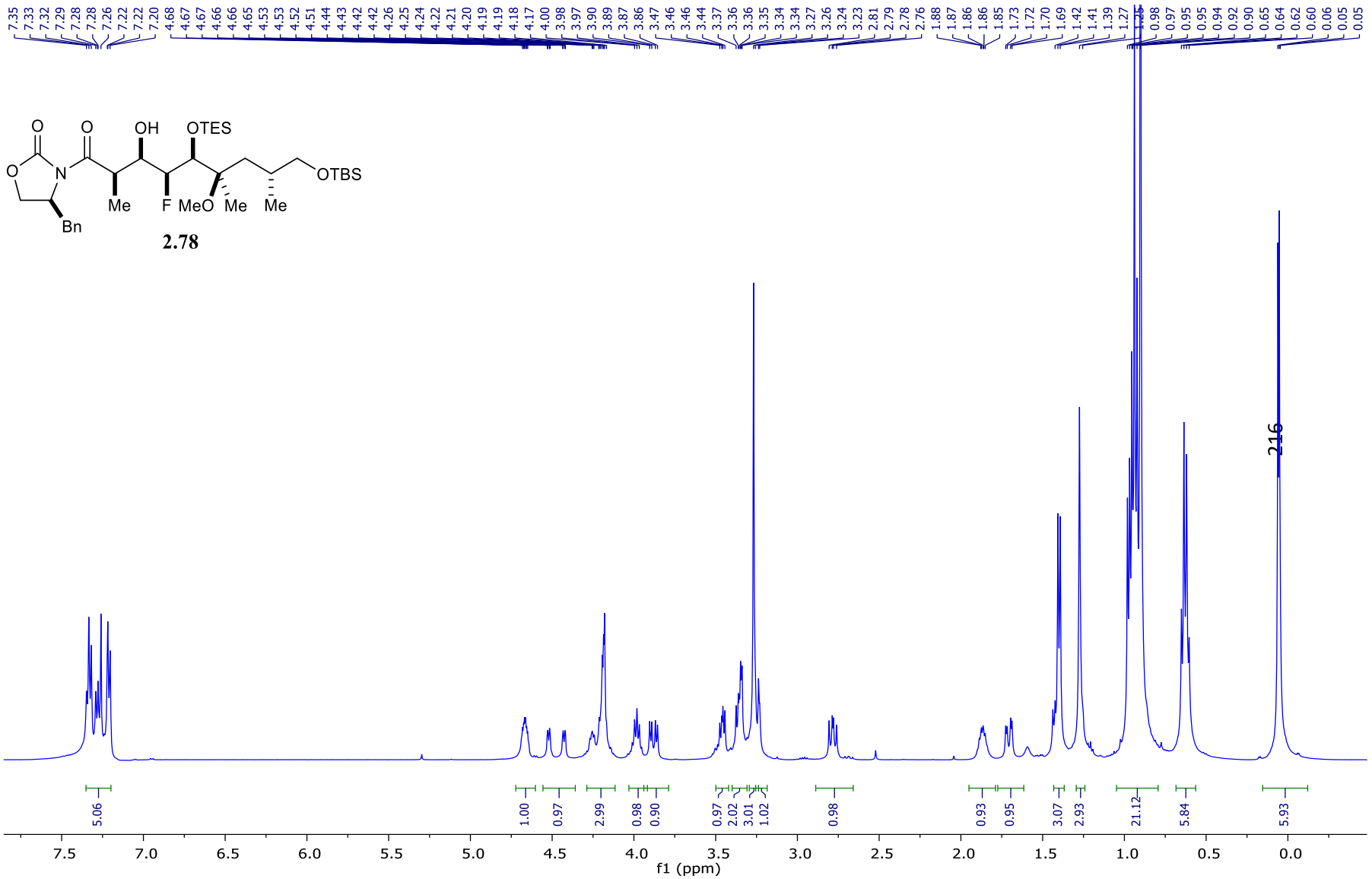
Fluorine NMR (Coupled)

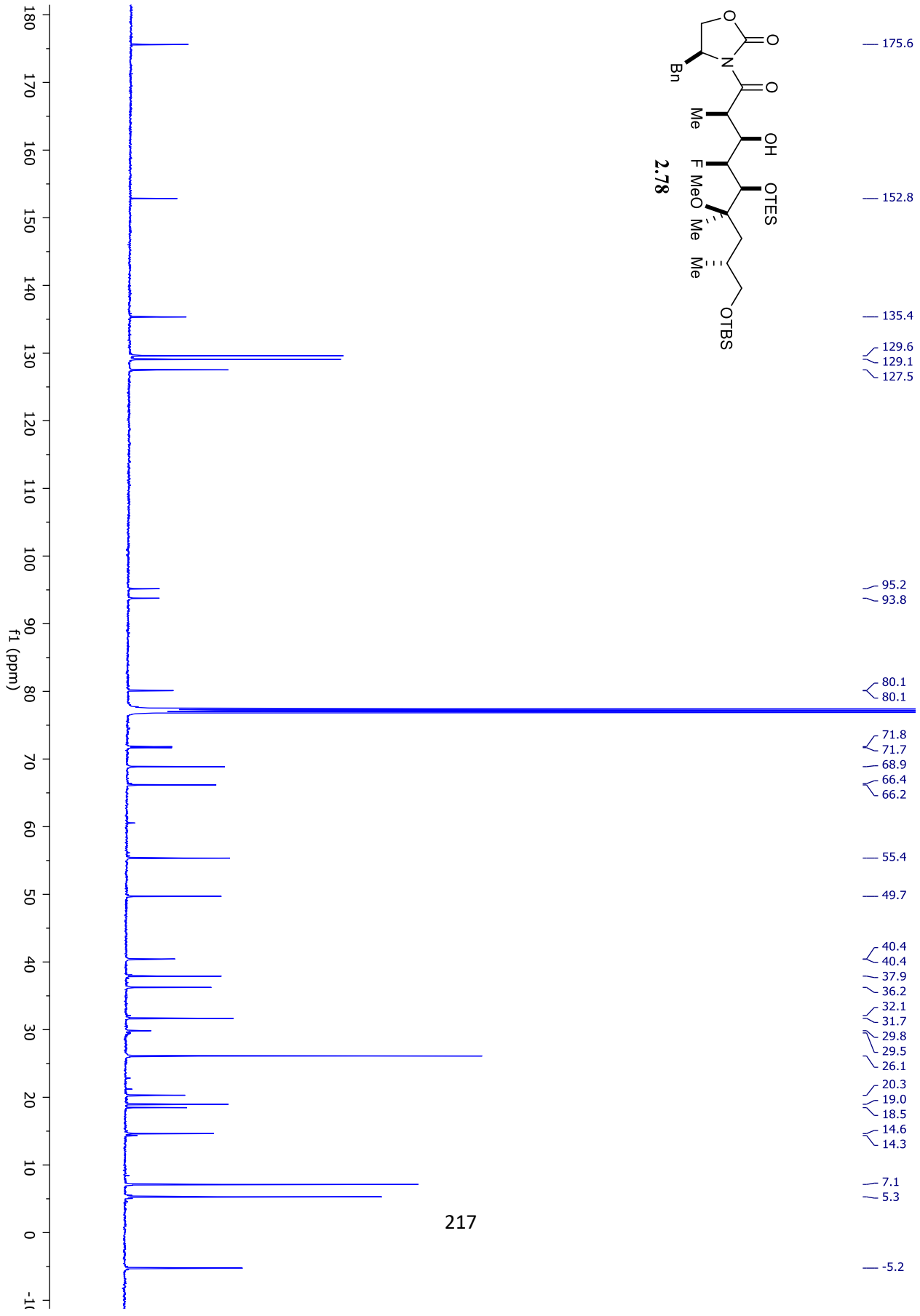


2.76

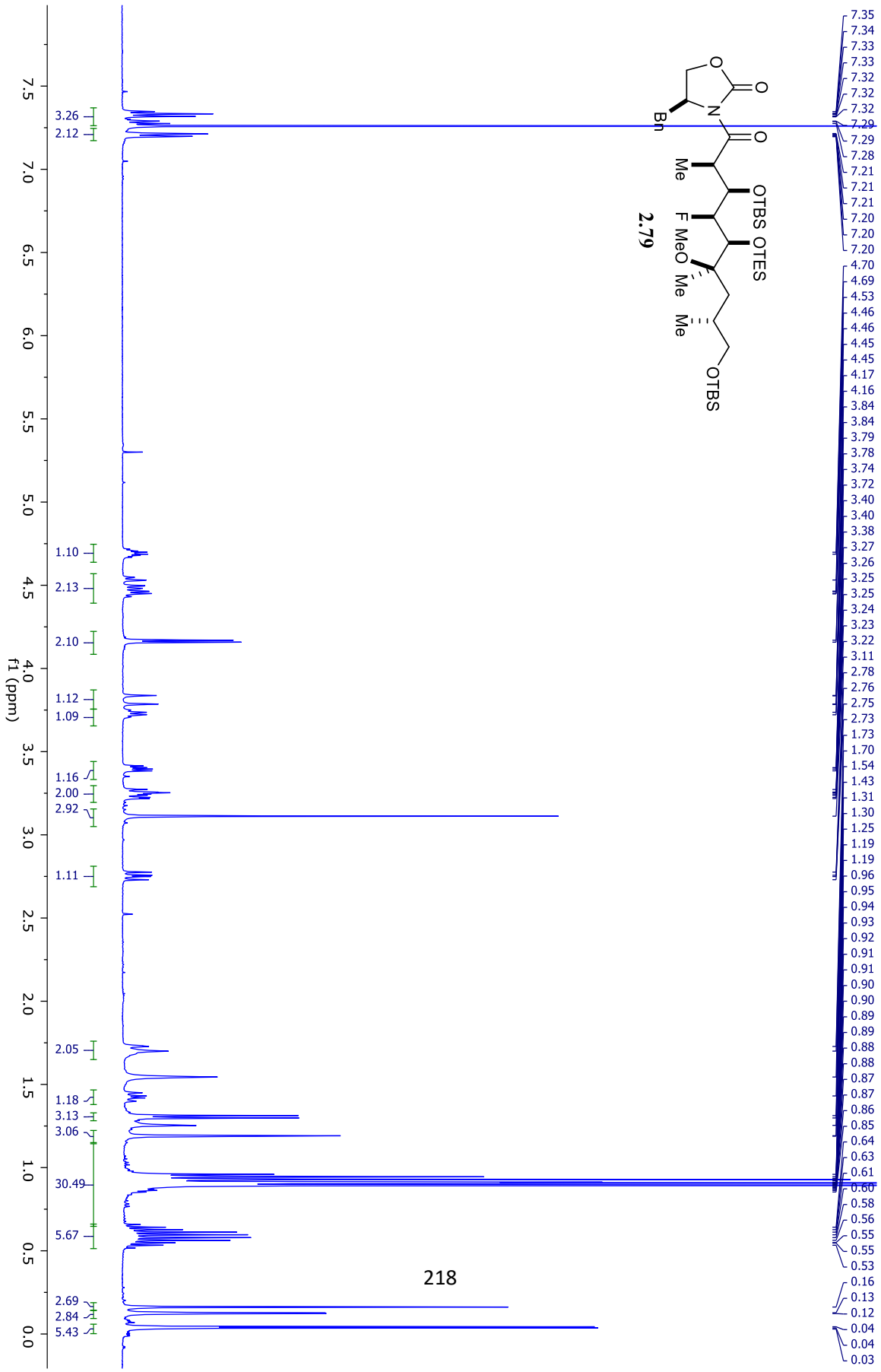

OTES
HO
OTBS
F MeO Me Me
2.77
214
1.00
0.96
2.03
2.05
2.92
0.97
0.96
0.97
0.98
3.03
11.70
8.98
5.93
5.89
f1 (ppm)

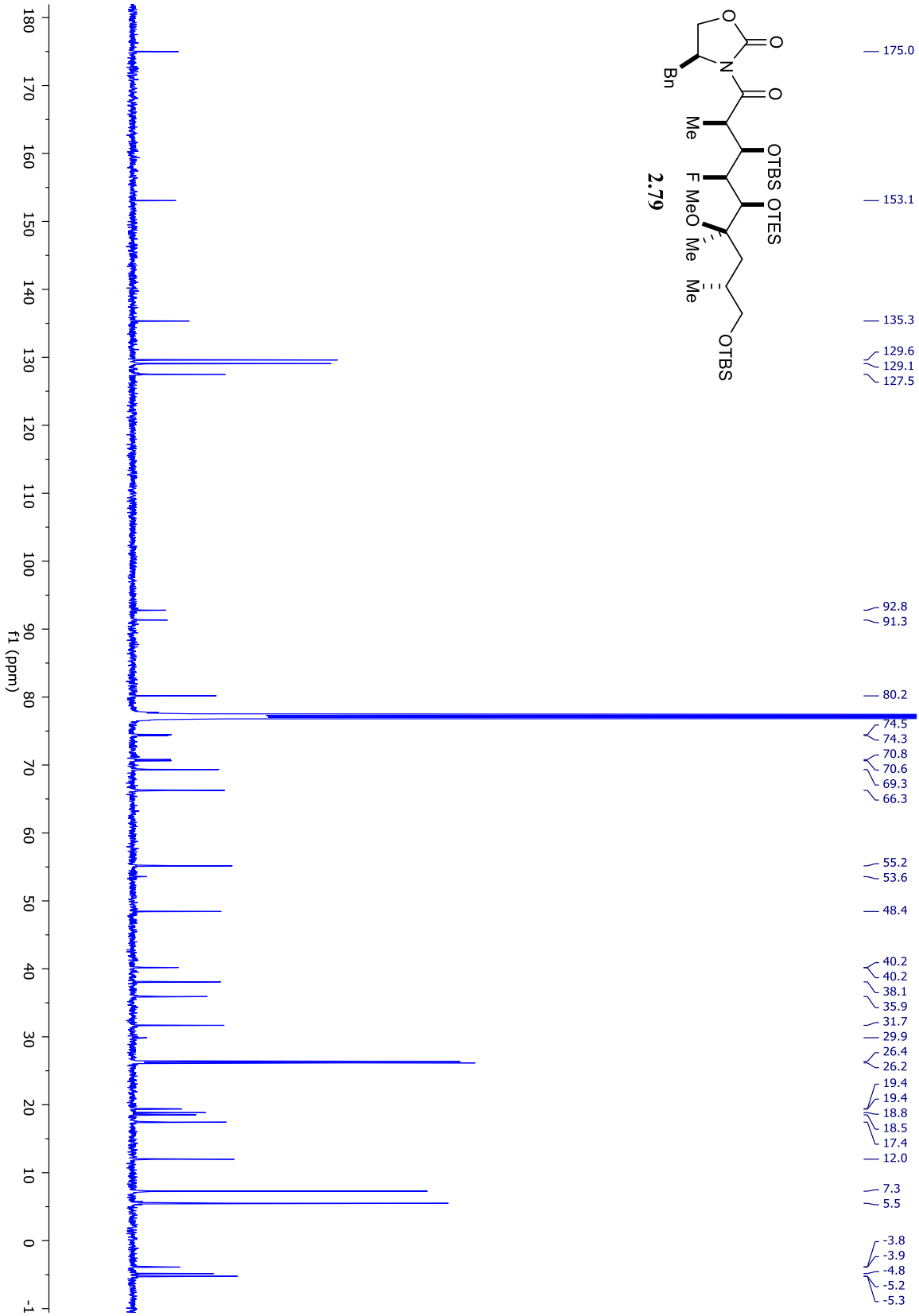

HO
OTES
F MeO Me Me
OTBS
2.77
95.1
93.7
80.0
76.4
76.3
68.9
63.3
63.1
49.4
36.6
31.6
26.1
20.0
18.7
18.5
7.1
6.9
6.8
5.5
5.3
5.1
-5.2
-5.3
f1 (ppm)

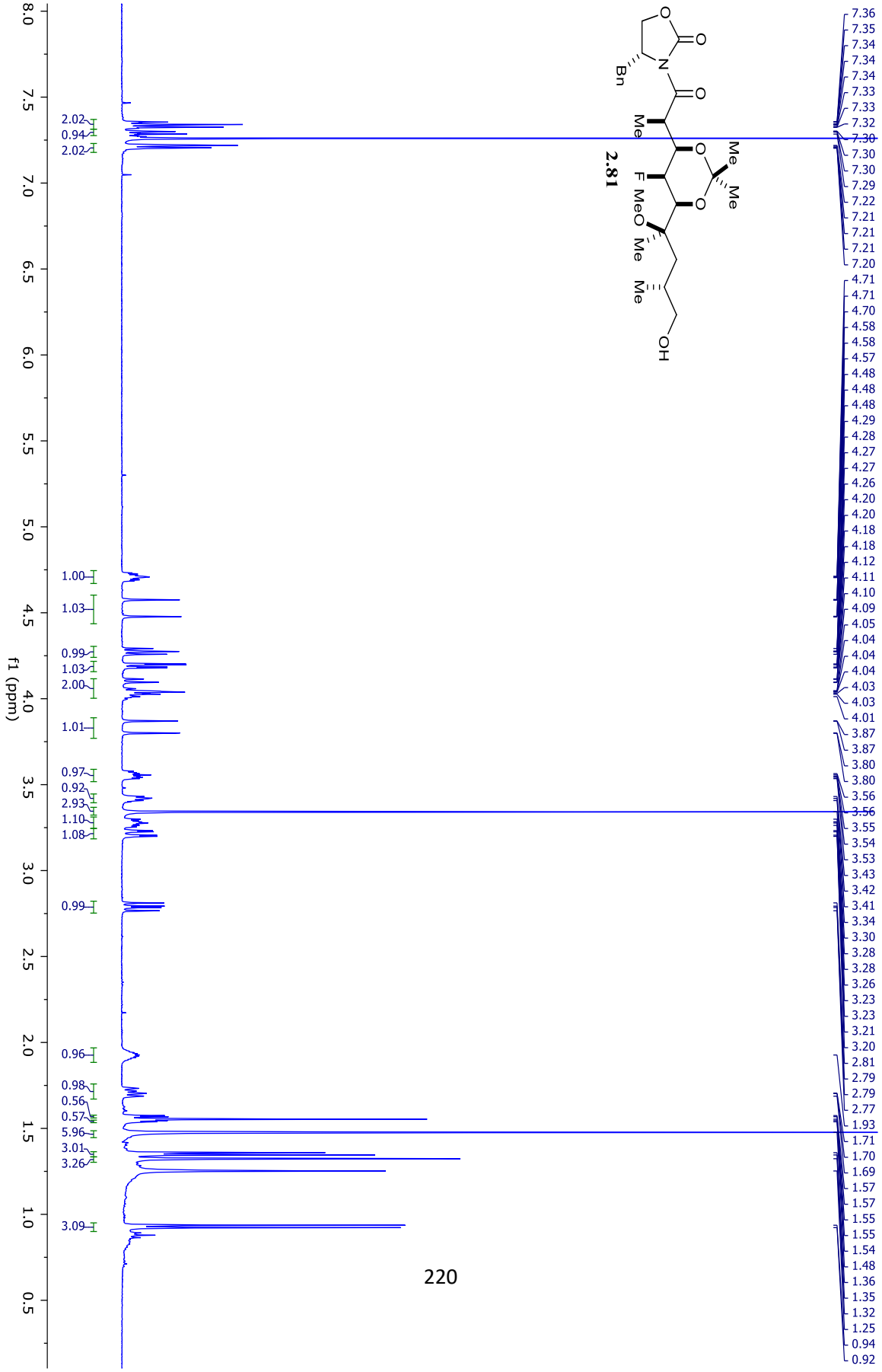

7.35 7.33 7.32 7.29 7.28 7.28 7.26 7.22 7.22 7.20
4.68 4.67 4.67 4.66 4.66 4.65 4.53 4.53 4.52 4.51 4.44 4.43 4.42 4.42 4.26 4.25 4.24 4.22 4.21 4.20 4.19 4.19 4.18 4.17 4.00 3.98 3.97 3.90 3.89 3.87 3.86 3.47 3.46 3.46 3.44 3.37 3.36 3.36 3.35 3.34 3.34 3.27 3.26 3.24 3.23 2.81 2.79 2.78 2.76
1.88 1.87 1.86 1.86 1.85 1.73 1.72 1.70 1.69 1.42 1.41 1.39 1.27 1.25 0.98 0.97 0.95 0.95 0.94 0.92 0.90 0.65 0.64 0.62 0.60 0.06 0.05 0.05
O
O
OH
OTES
O
N
OTBS
Me
F
MeO
Me
Me
Bn
2.78
216
5.06
1.00
0.97
2.99
0.98
0.90
0.97
2.02
3.01
1.02
0.98
0.93
0.95
3.07
2.93
21.12
5.84
5.93
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

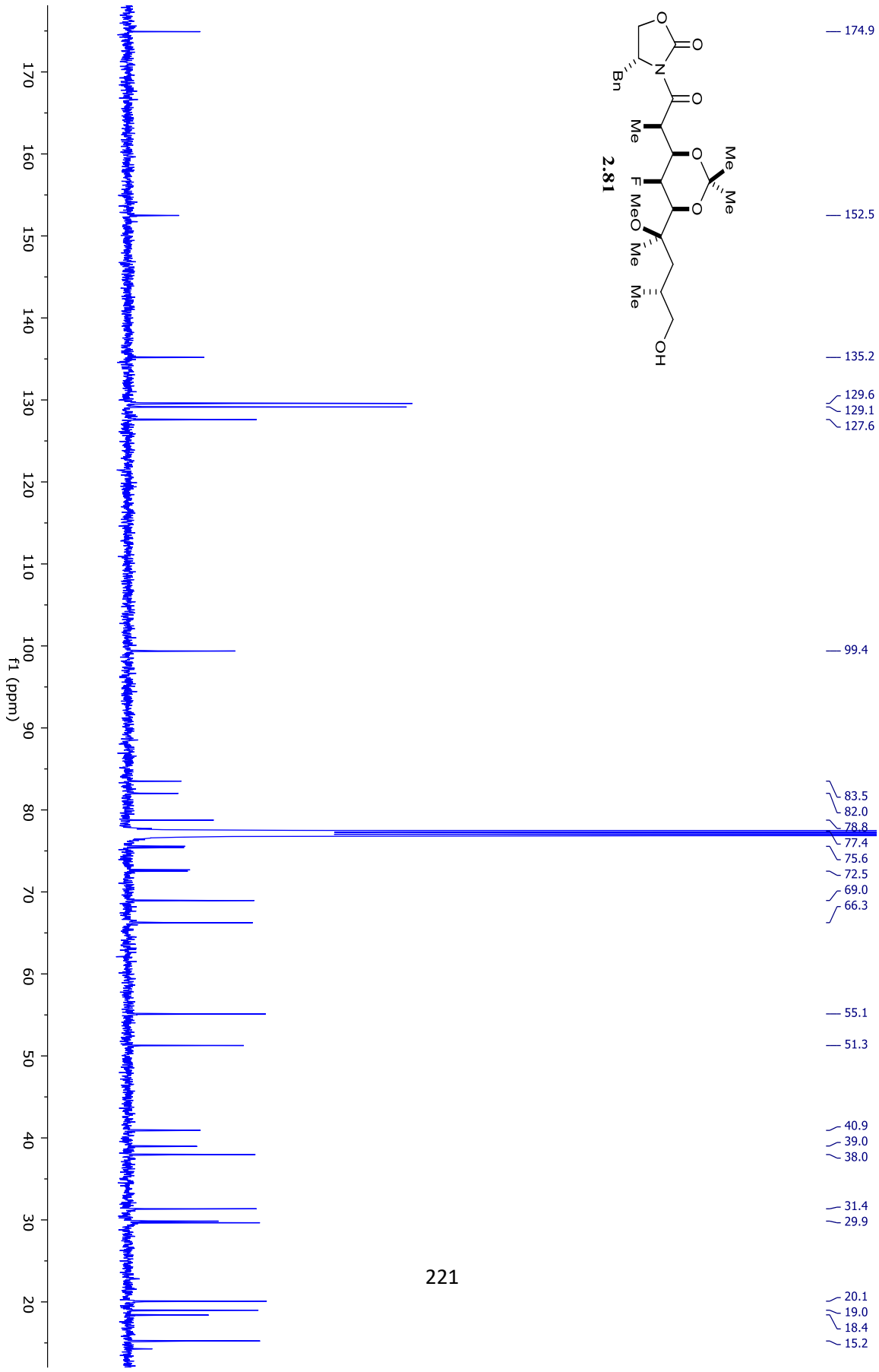

f1 (ppm)
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-1
175.6
152.8
135.4
129.6
129.1
127.5
95.2
93.8
80.1
80.1
71.8
71.7
68.9
66.4
66.2
55.4
49.7
40.4
40.4
37.9
36.2
32.1
31.7
29.8
29.5
26.1
20.3
19.0
18.5
14.6
14.3
7.1
5.3
-5.2
O
O
N
Bn
O
OH
OTES
Me
F
MeO
Me
Me
OTBS

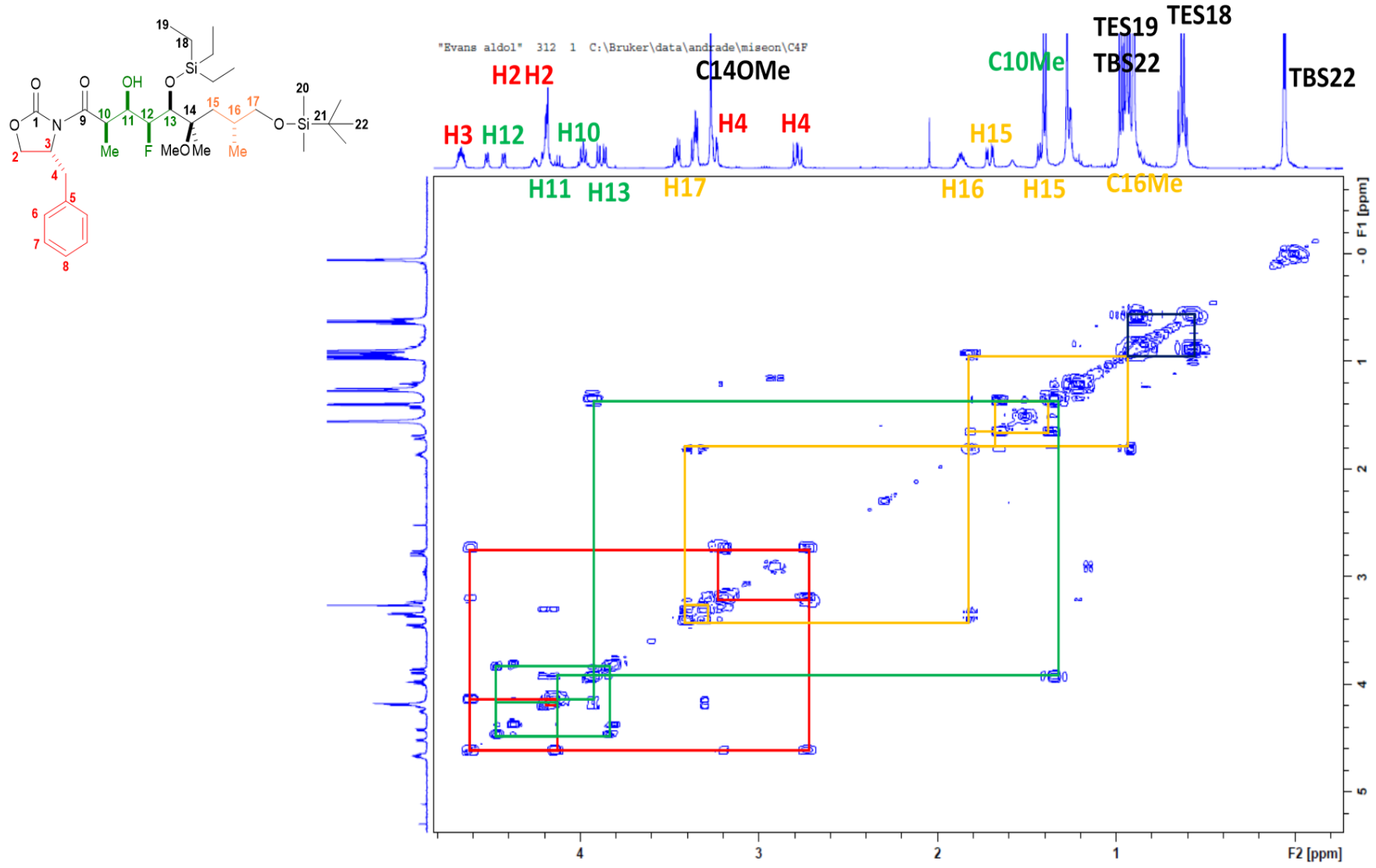
2.78

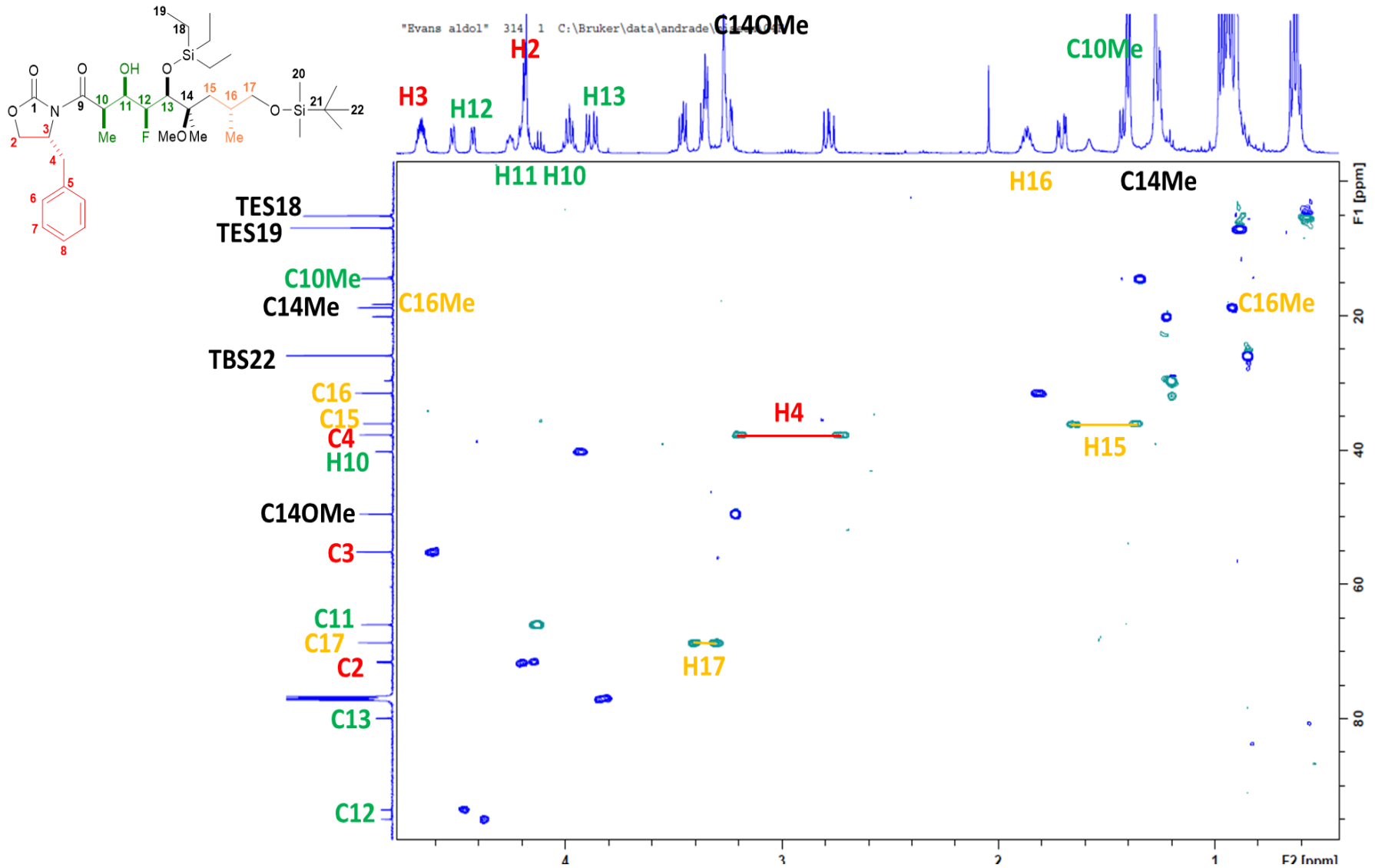

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
3.26
2.12
1.10
2.13
2.10
1.12
1.09
1.16
2.00
2.92
1.11
2.05
1.18
3.13
3.06
30.49
5.67
2.69
2.84
5.43
Bn
O
O
N
O
Me
OTBS OTES
F MeO Me Me
OTBS
2.79

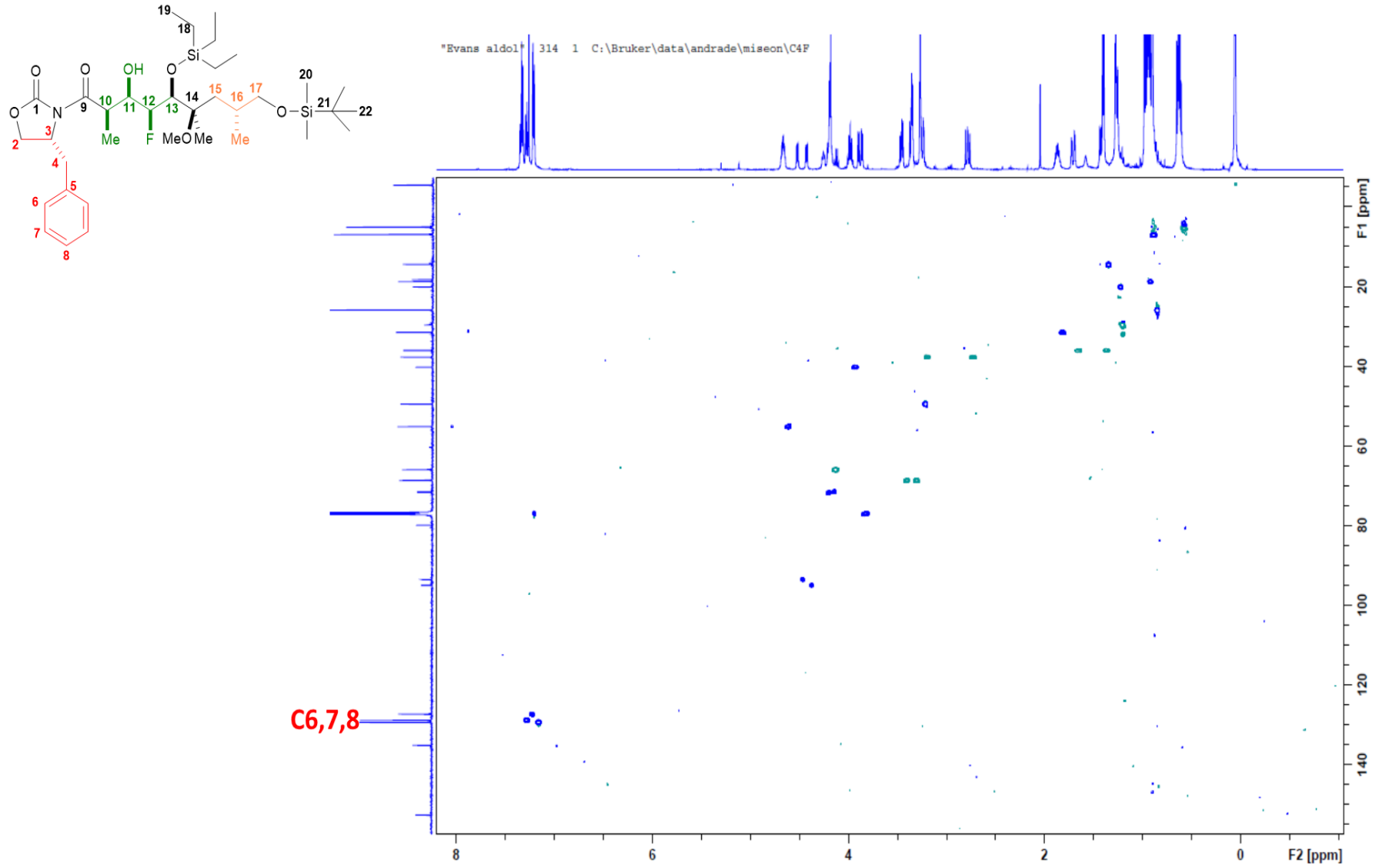

2.79
Bn
Me
F
MeO
Me
Me
OTBS
OTBS
OTES
f1 (ppm)
175.0
153.1
135.3
129.6
129.1
127.5
92.8
91.3
80.2
74.5
74.3
70.8
70.6
69.3
66.3
55.2
53.6
48.4
40.2
40.2
38.1
35.9
31.7
29.9
26.4
26.2
19.4
19.4
18.8
18.5
17.4
12.0
7.3
5.5
-3.8
-3.9
-4.8
-5.2
-5.3

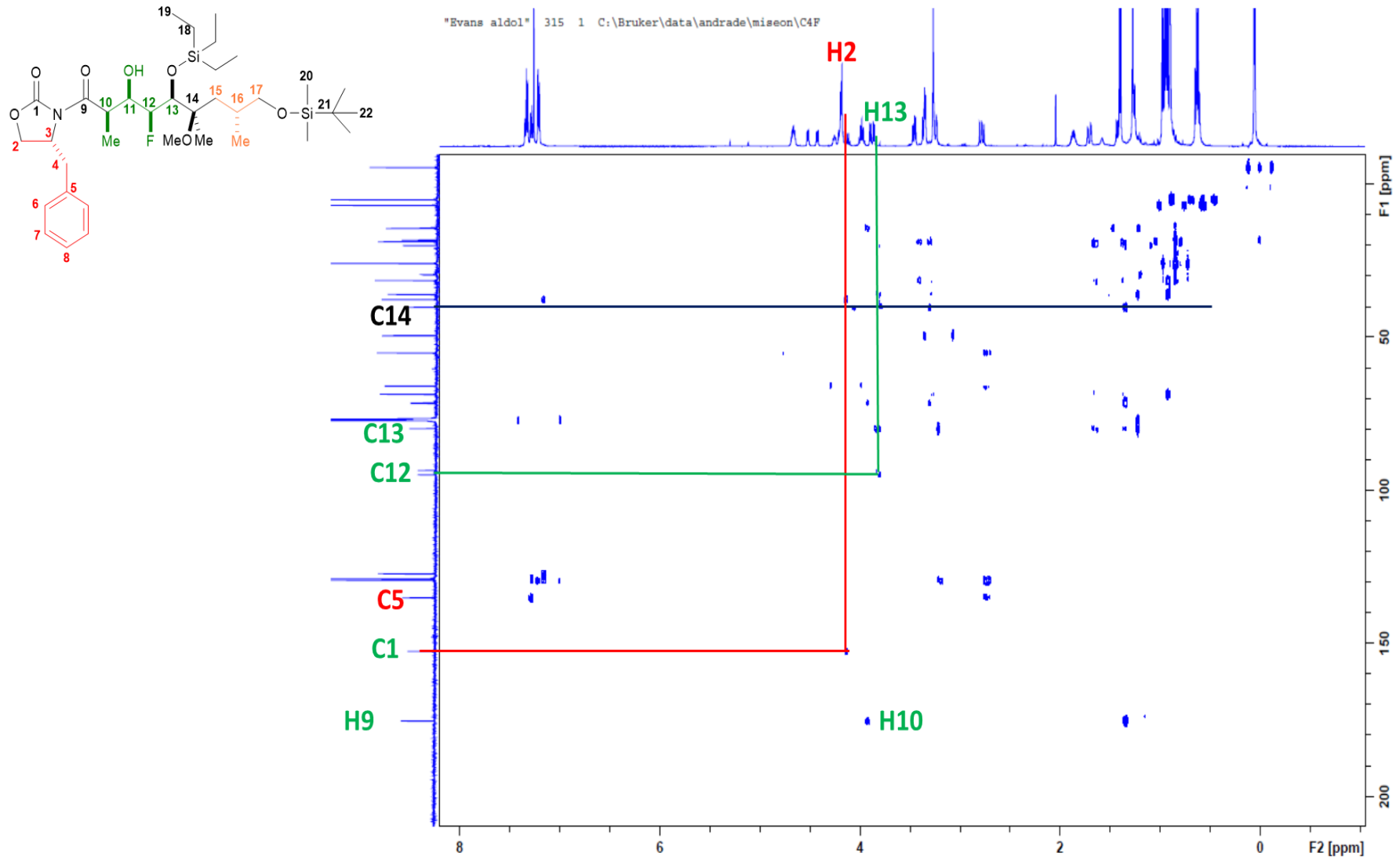

f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
2.02
0.94
2.02
1.00
1.03
0.99
1.03
2.00
1.01
0.97
0.92
2.93
1.10
1.08
0.99
0.96
0.98
0.56
0.57
5.96
3.01
3.26
3.09
Bn
Me
F
MeO
Me
Me
Me
Me
Me
OH
2.81


174.9
152.5
135.2
129.6
129.1
127.6
99.4
83.5
82.0
78.8
77.4
75.6
72.5
69.0
66.3
55.1
51.3
40.9
39.0
38.0
31.4
29.9
20.1
19.0
18.4
15.2
f1 (ppm)
2.81


"Evans aldol" 312 1 C:\Bruker\data\andrade\miseon\C4F
H2 H2
C14OMe
C10Me
TES19
TES18
TBS22
TBS22
H3 H12
H10
H4
H4
H15
H11 H13
H17
H16
H15
C16Me
F1 [ppm]
F2 [ppm]
Me
F
MeO Me
Me
OH


"Evans aldol" 314 1 C:\Bruker\data\andrade\
C14OMe
H2
H3
H12
H13
C10Me
H11 H10
H16
C14Me
TES18
TES19
C10Me
C14Me
C16Me
C16Me
TBS22
C16
C15
C4
H10
H4
H15
C14OMe
C3
C11
C17
C2
H17
C13
C12
F1 [ppm]
F2 [ppm]

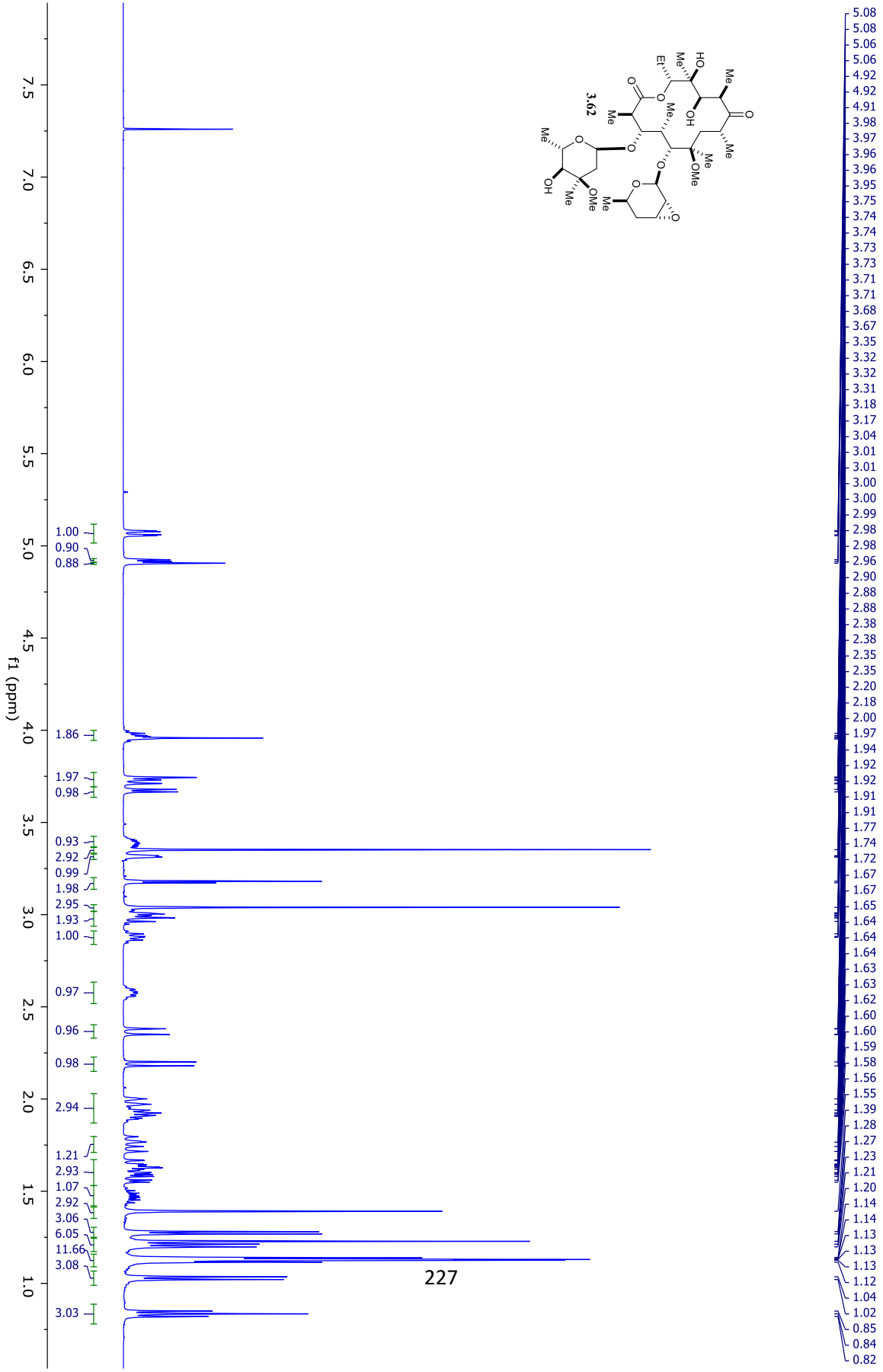

"Evans aldol" 314 1 C:\Bruker\data\andrade\miseon\C4F
C6,7,8
F2 [ppm]
F1 [ppm]

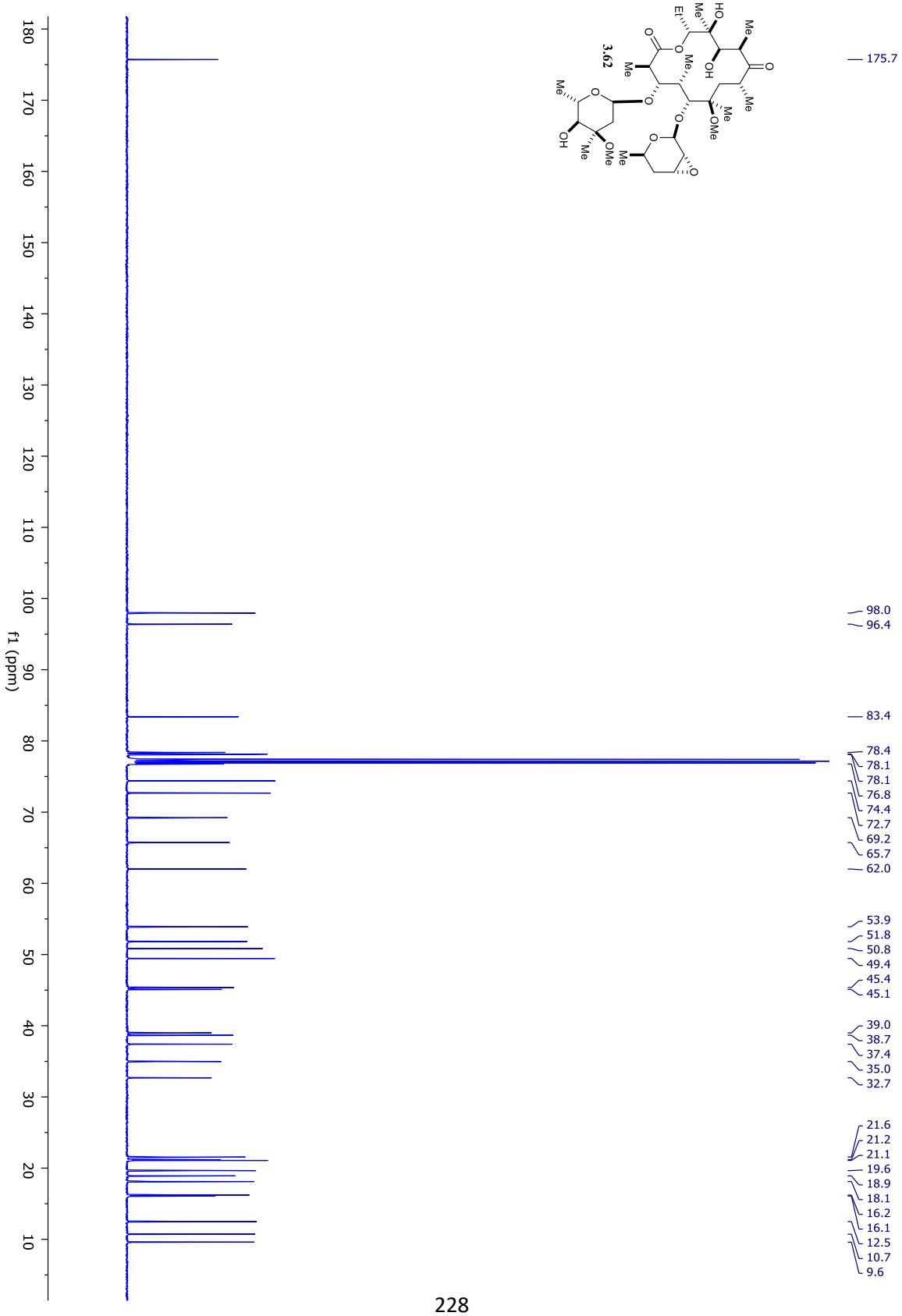

"Evans aldol" 315 1 C:\Bruker\data\andrade\miseon\C4F
H2
H13
C14
C13
C12
C5
C1
H9
H10
F1 [ppm]
F2 [ppm]
8
6
4
2
0
50
100
150
200
Me
F
MeO
Me
Me
OH

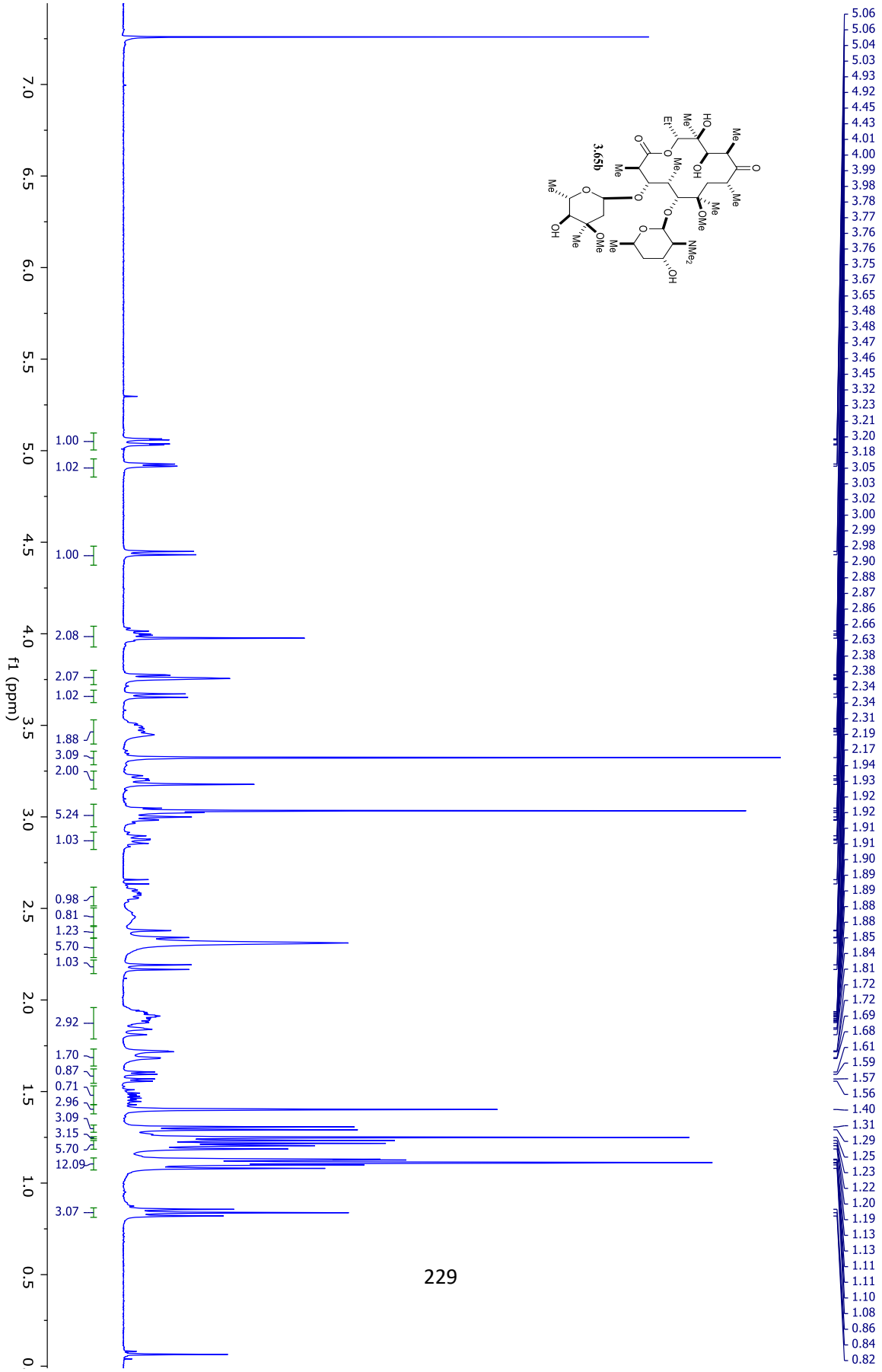
APPENDIX A2

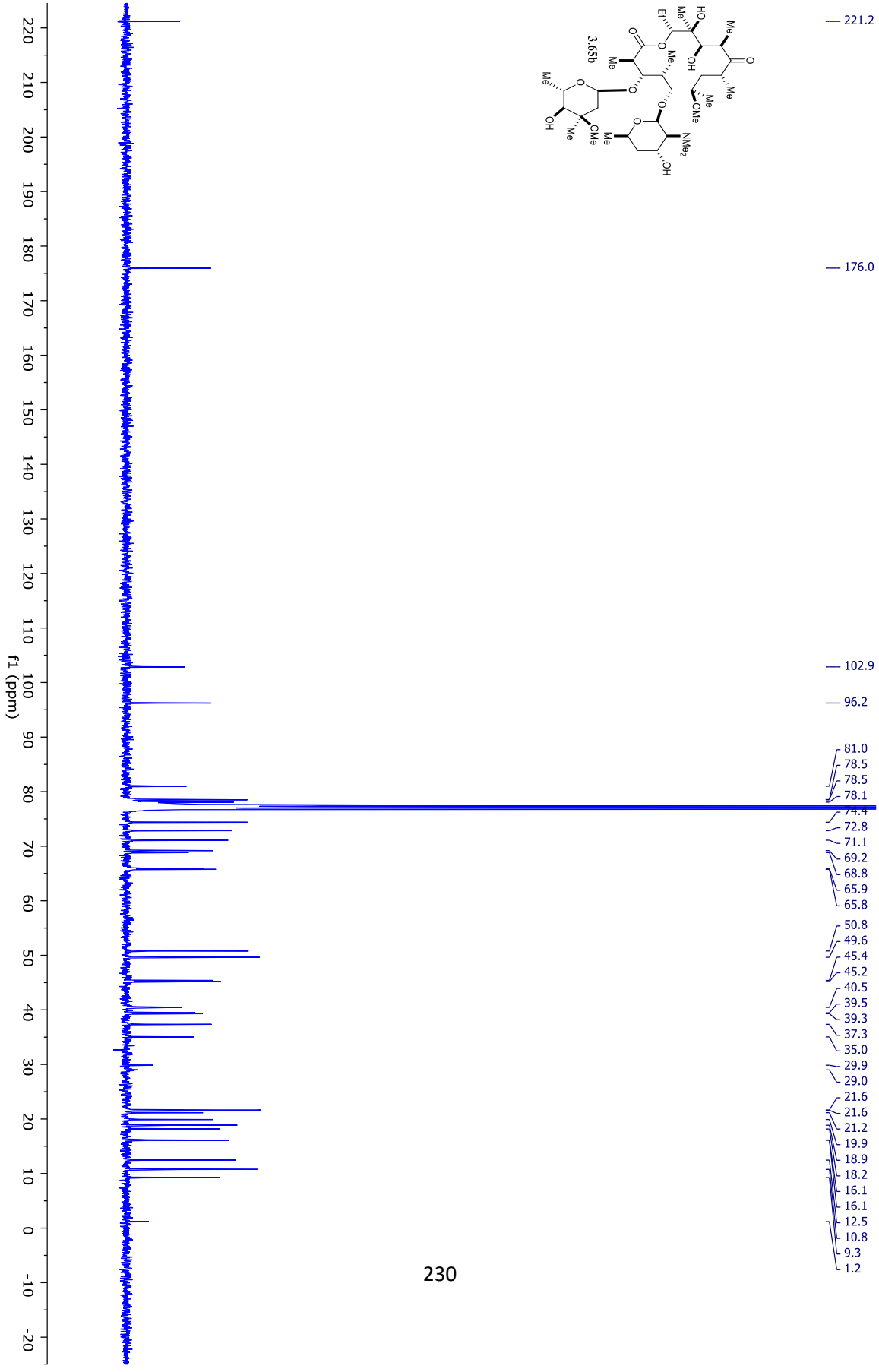
NMR SPECTRA OF COMPOUNDS IN

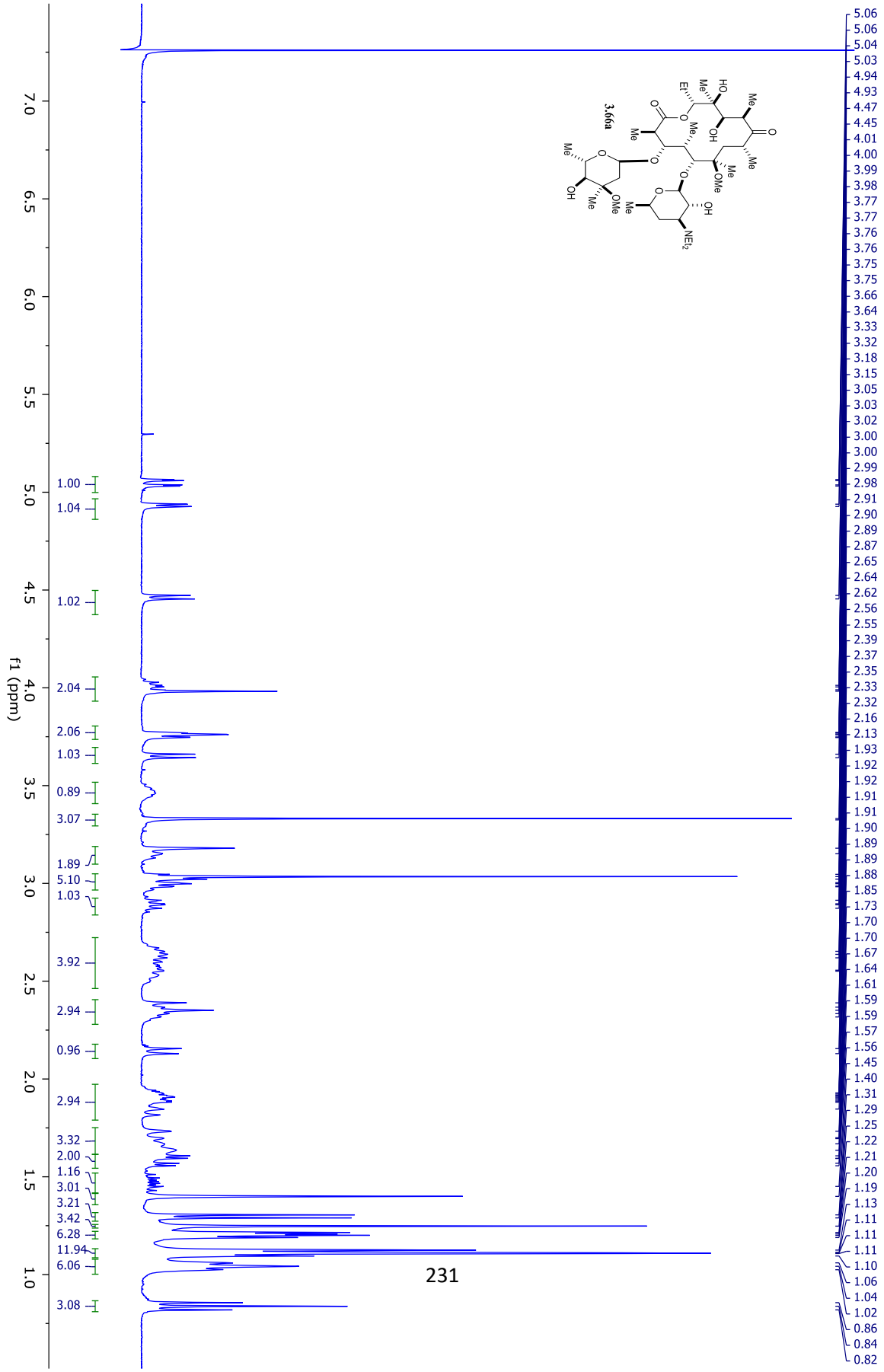
CHAPTER 3

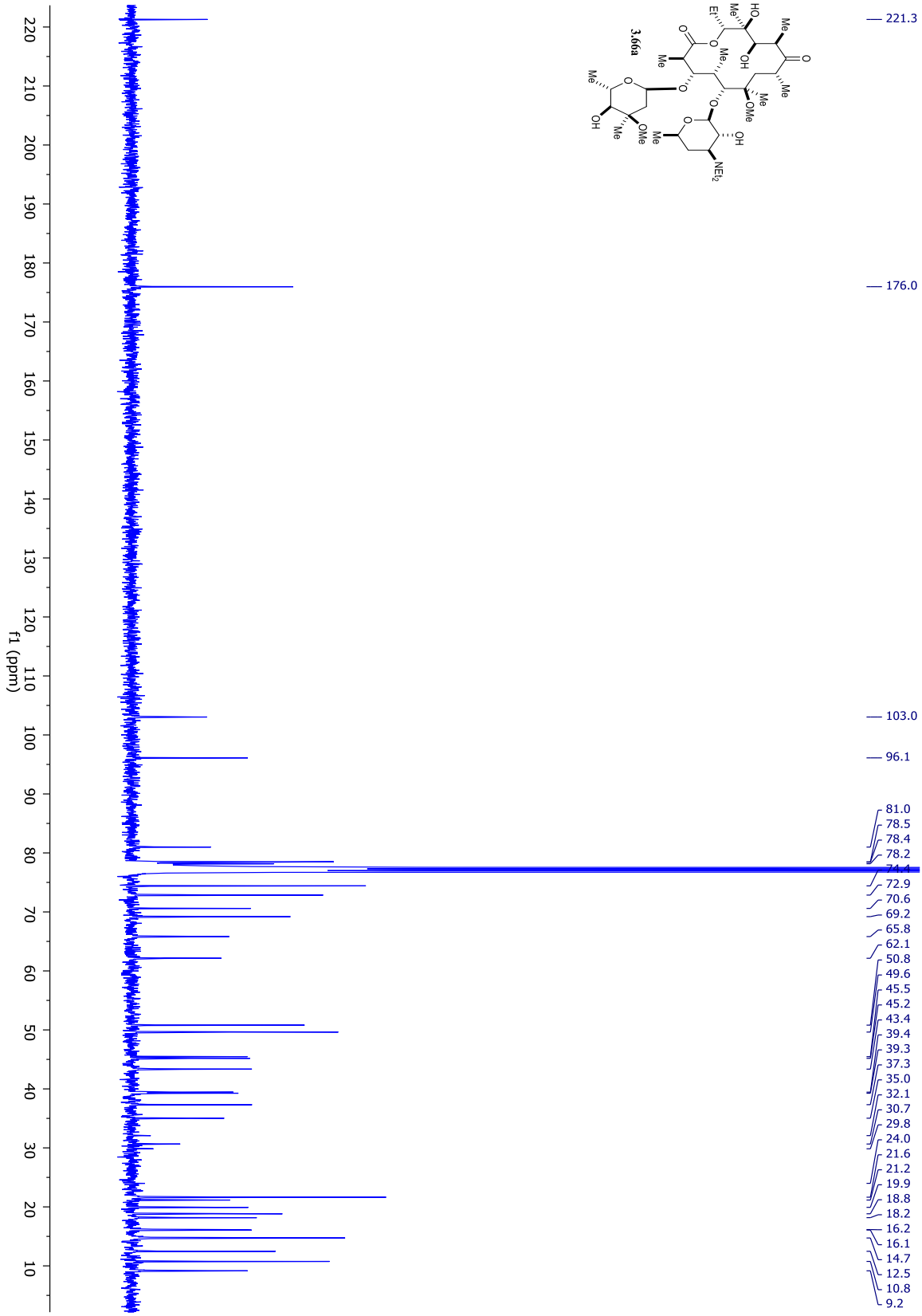

3.62
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
1.00
0.90
0.88
1.86
1.97
0.98
0.93
2.92
0.99
1.98
2.95
1.93
1.00
0.97
0.96
0.98
2.94
1.21
2.93
1.07
2.92
3.06
6.05
11.66
3.08
3.03
5.08
5.08
5.06
5.06
4.92
4.92
4.91
3.98
3.97
3.96
3.96
3.95
3.75
3.74
3.74
3.73
3.73
3.71
3.71
3.68
3.67
3.35
3.32
3.32
3.31
3.18
3.17
3.04
3.01
3.01
3.00
3.00
2.99
2.98
2.98
2.96
2.90
2.88
2.88
2.38
2.38
2.35
2.35
2.20
2.18
2.00
1.97
1.94
1.92
1.92
1.91
1.91
1.77
1.74
1.72
1.67
1.67
1.65
1.64
1.64
1.64
1.63
1.63
1.62
1.60
1.60
1.59
1.58
1.56
1.55
1.39
1.28
1.27
1.23
1.21
1.20
1.14
1.14
1.13
1.13
1.13
1.12
1.04
1.02
0.85
0.84
0.82

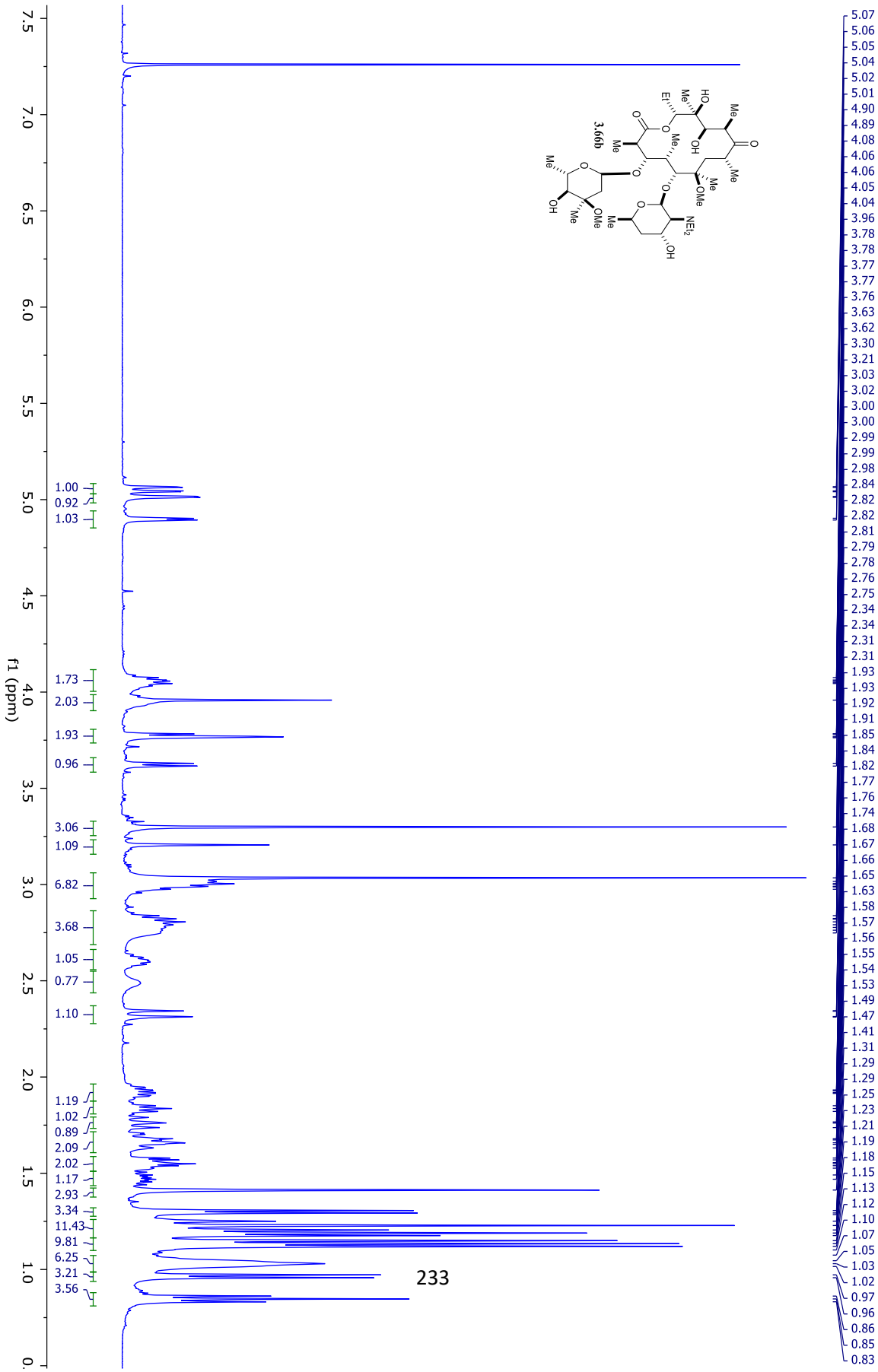

3.62
f1 (ppm)
175.7
98.0
96.4
83.4
78.4
78.1
78.1
76.8
74.4
72.7
69.2
65.7
62.0
53.9
51.8
50.8
49.4
45.4
45.1
39.0
38.7
37.4
35.0
32.7
21.6
21.2
21.1
19.6
18.9
18.1
16.2
16.1
12.5
10.7
9.6

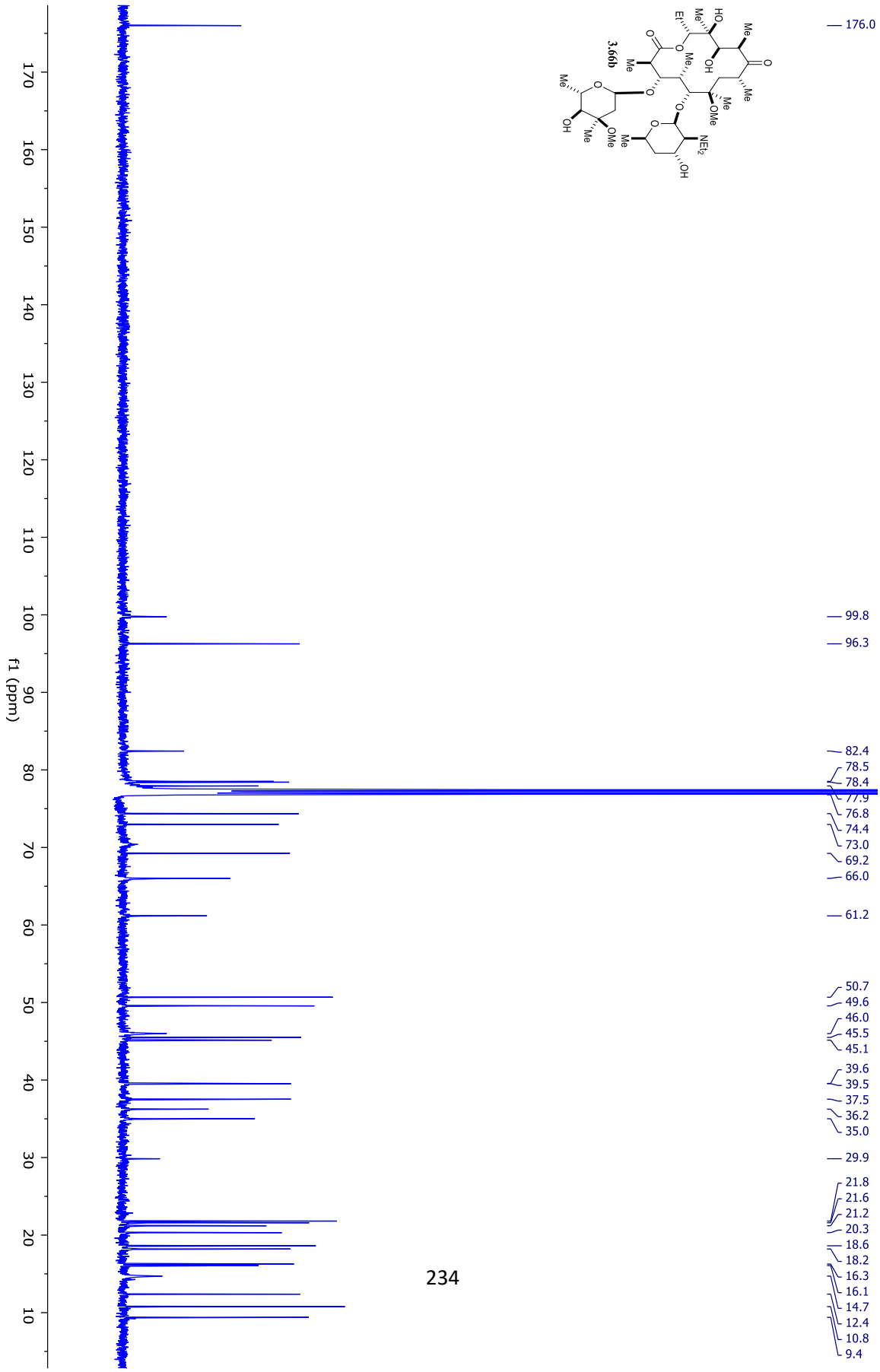

3.65b
f1 (ppm)
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0
1.00
1.02
1.00
2.08
2.07
1.02
1.88
3.09
2.00
5.24
1.03
0.98
0.81
1.23
5.70
1.03
2.92
1.70
0.87
0.71
2.96
3.09
3.15
5.70
12.09
3.07

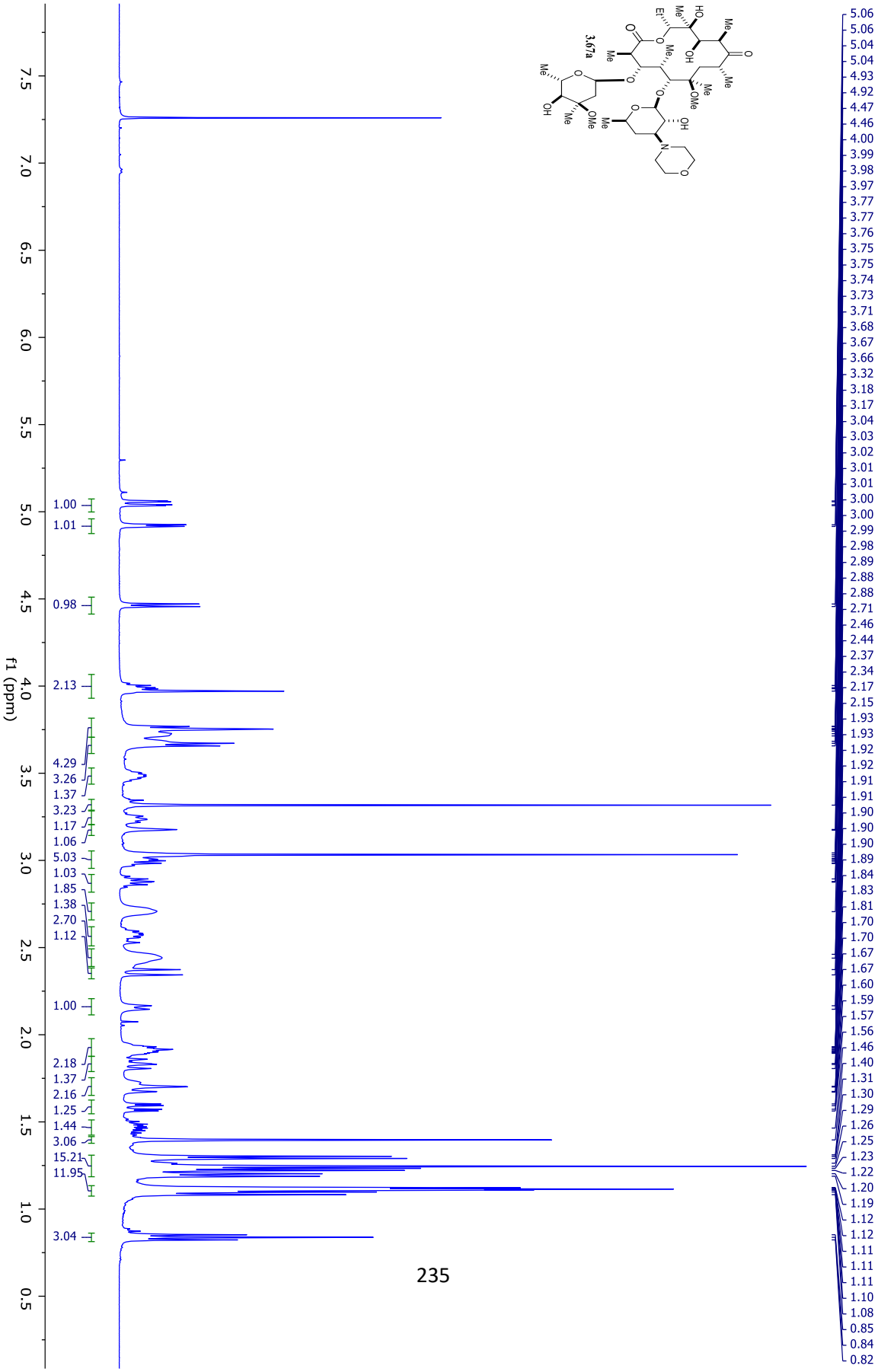

3.65b
221.2
176.0
102.9
96.2
81.0
78.5
78.5
78.1
74.4
72.8
71.1
69.2
68.8
65.9
65.8
50.8
49.6
45.4
45.2
40.5
39.5
39.3
37.3
35.0
29.9
29.0
21.6
21.6
21.2
19.9
18.9
18.2
16.1
16.1
12.5
10.8
9.3
1.2
f1 (ppm)

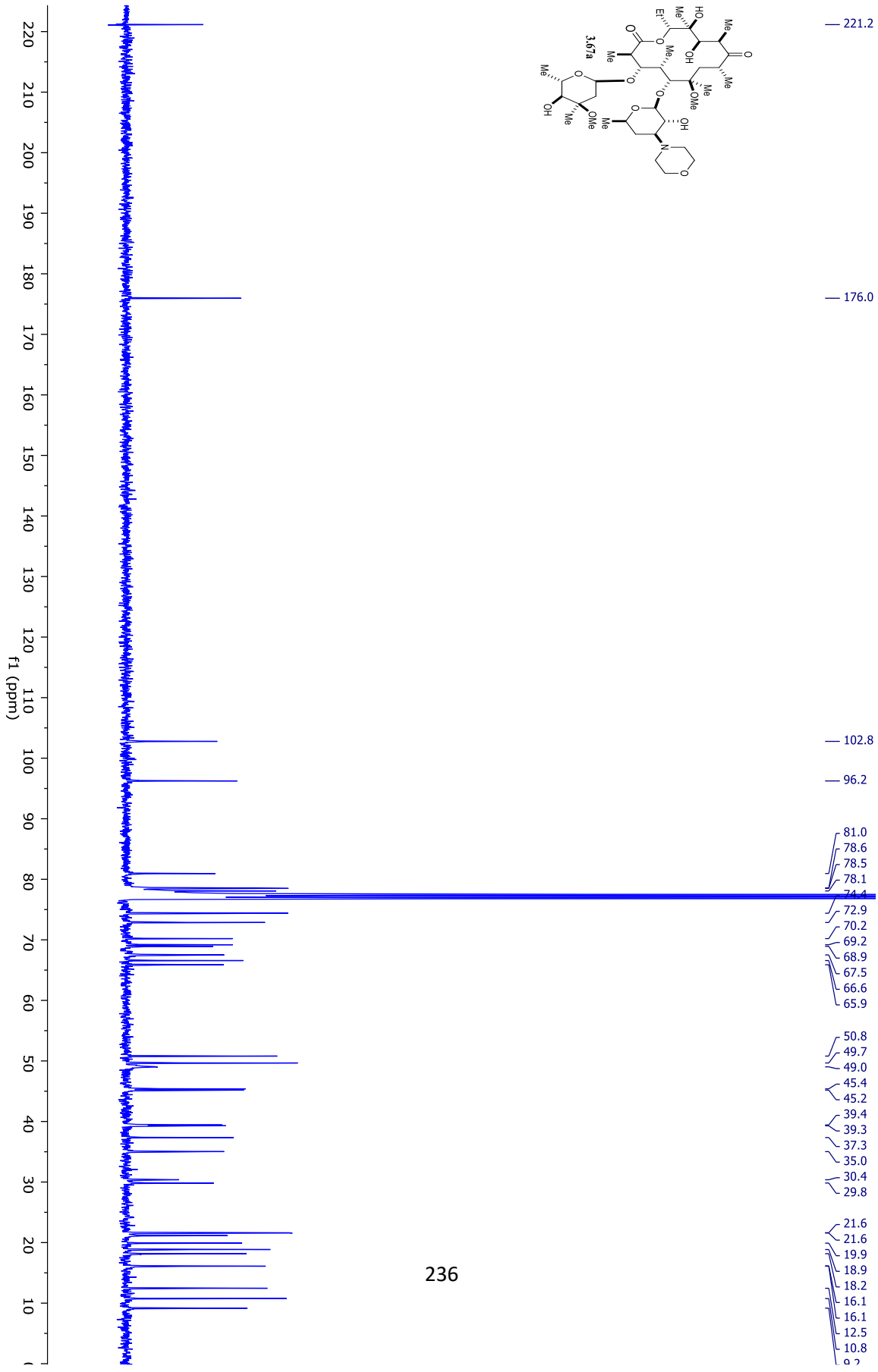

3.66a
f1 (ppm)
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
1.00
1.04
1.02
2.04
2.06
1.03
0.89
3.07
1.89
5.10
1.03
3.92
2.94
0.96
2.94
3.32
2.00
1.16
3.01
3.21
3.42
6.28
11.94
6.06
3.08
5.06
5.06
5.04
5.03
4.94
4.93
4.47
4.45
4.01
4.00
3.99
3.98
3.77
3.77
3.76
3.76
3.75
3.75
3.66
3.64
3.33
3.32
3.18
3.15
3.05
3.03
3.02
3.00
3.00
2.99
2.98
2.91
2.90
2.89
2.87
2.65
2.64
2.62
2.56
2.55
2.39
2.37
2.35
2.33
2.32
2.16
2.13
1.93
1.92
1.92
1.91
1.91
1.90
1.89
1.89
1.88
1.85
1.73
1.70
1.70
1.67
1.64
1.61
1.59
1.59
1.57
1.56
1.45
1.40
1.31
1.29
1.25
1.22
1.21
1.20
1.19
1.13
1.11
1.11
1.11
1.10
1.06
1.04
1.02
0.86
0.84
0.82

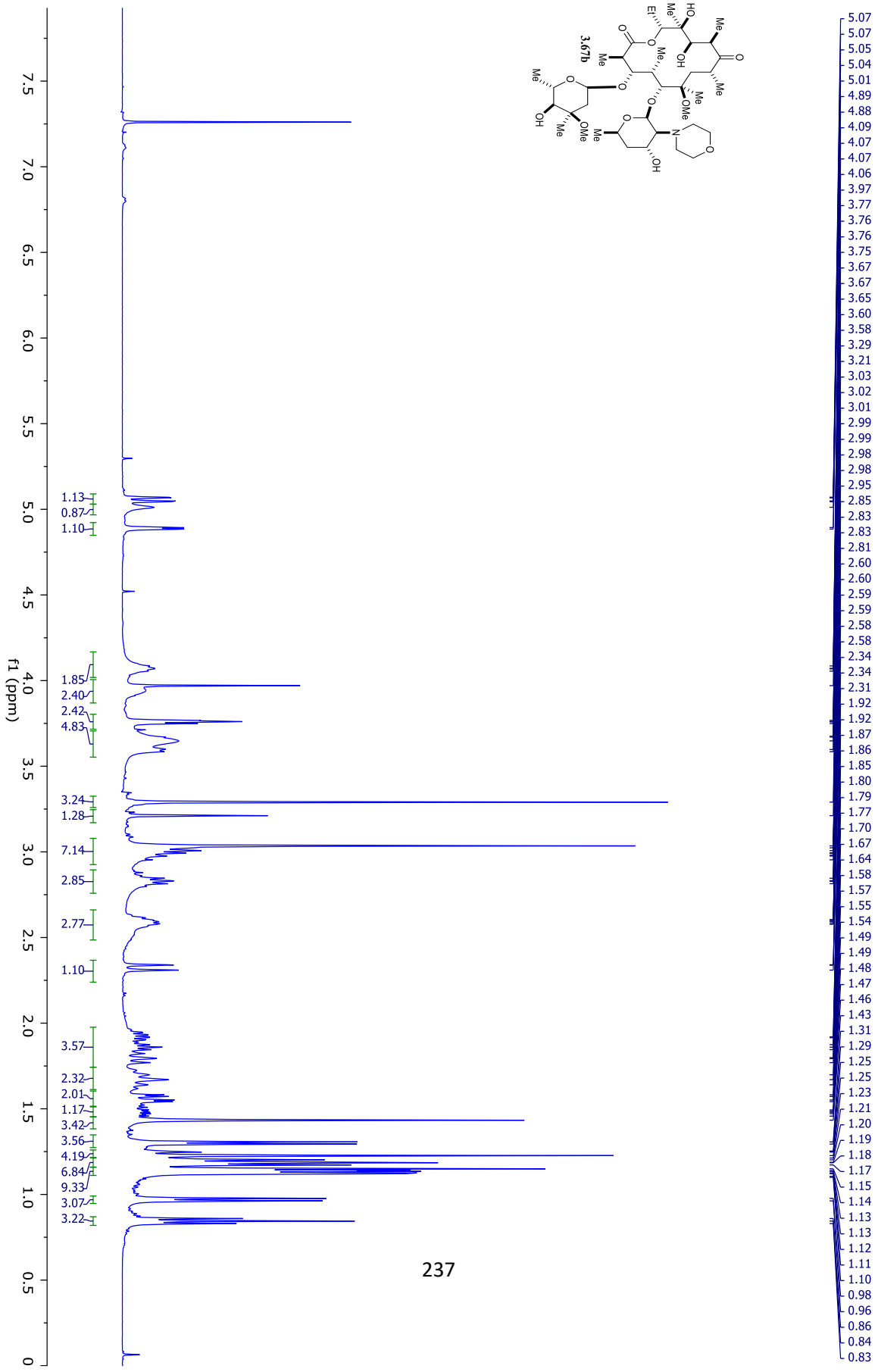

3.66a
f1 (ppm)
221.3
176.0
103.0
96.1
81.0
78.5
78.4
78.2
72.9
70.6
69.2
65.8
62.1
50.8
49.6
45.5
45.2
43.4
39.4
39.3
37.3
35.0
32.1
30.7
29.8
24.0
21.6
21.2
19.9
18.8
18.2
16.2
16.1
14.7
12.5
10.8
9.2

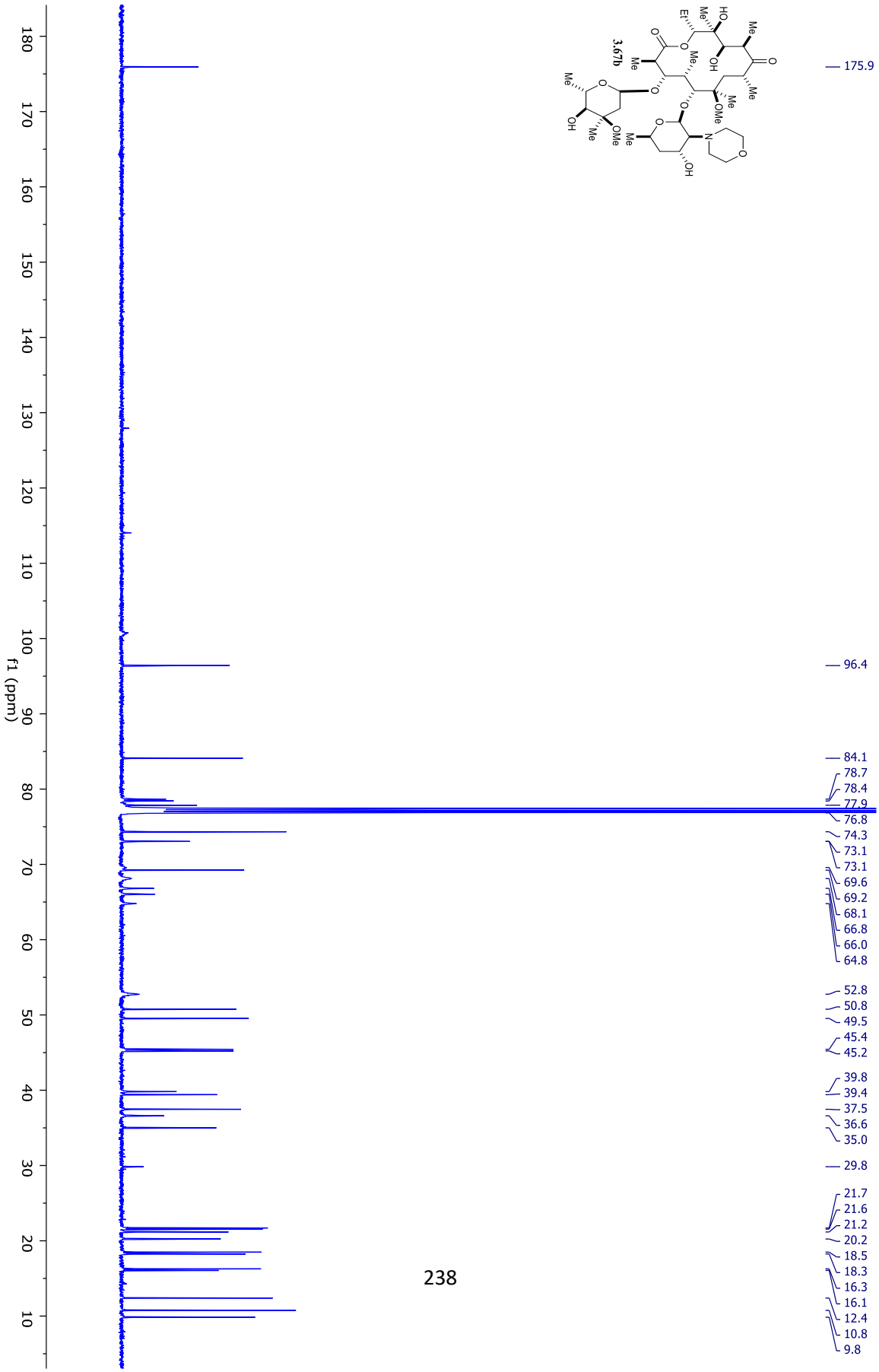

3.66b
f1 (ppm)
1.00
0.92
1.03
1.73
2.03
1.93
0.96
3.06
1.09
6.82
3.68
1.05
0.77
1.10
1.19
1.02
0.89
2.09
2.02
1.17
2.93
3.34
11.43
9.81
6.25
3.21
3.56

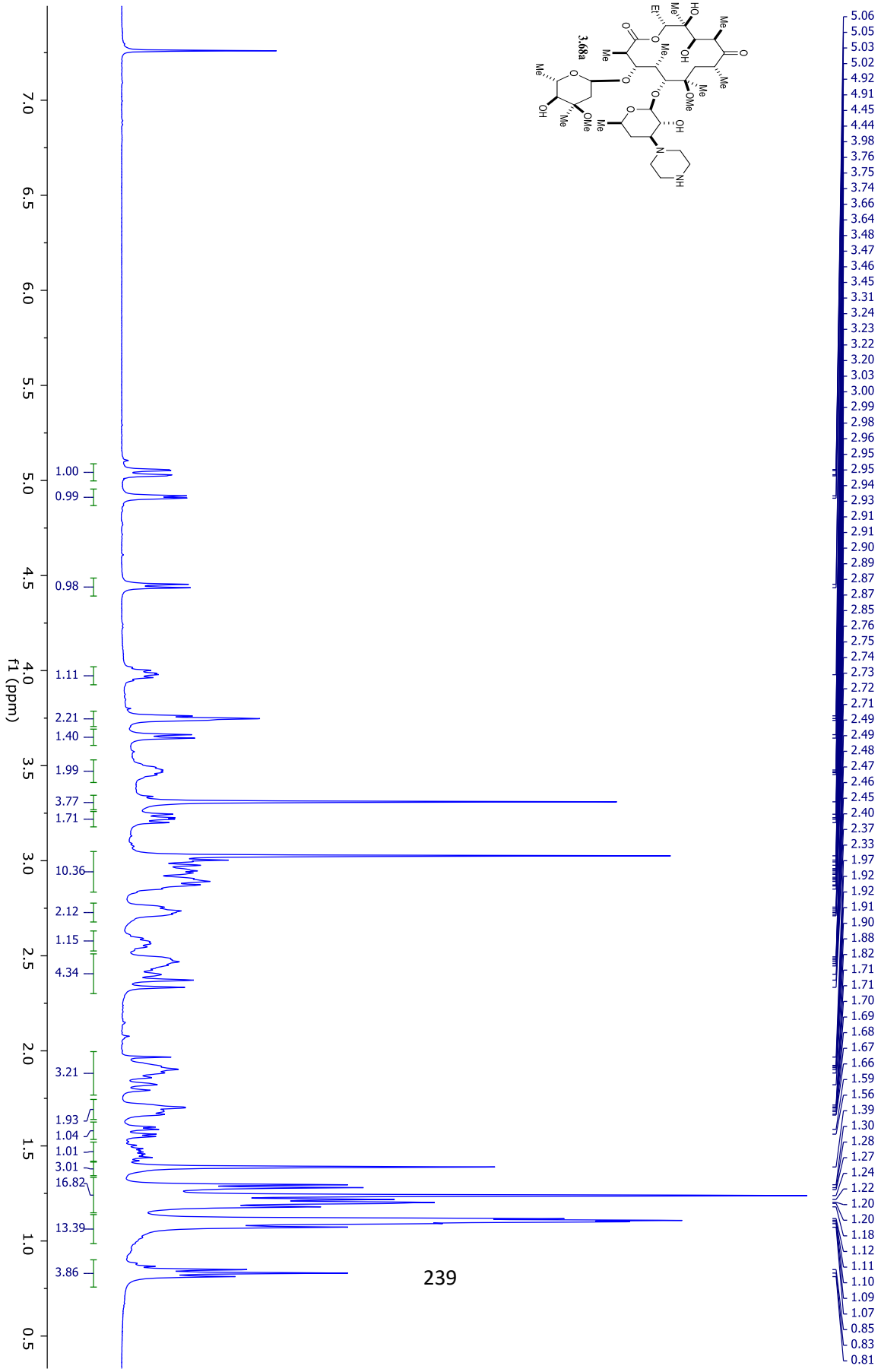

3.66b
f1 (ppm)
176.0
99.8
96.3
82.4
78.5
78.4
77.9
76.8
74.4
73.0
69.2
66.0
61.2
50.7
49.6
46.0
45.5
45.1
39.6
39.5
37.5
36.2
35.0
29.9
21.8
21.6
21.2
20.3
18.6
18.2
16.3
16.1
14.7
12.4
10.8
9.4

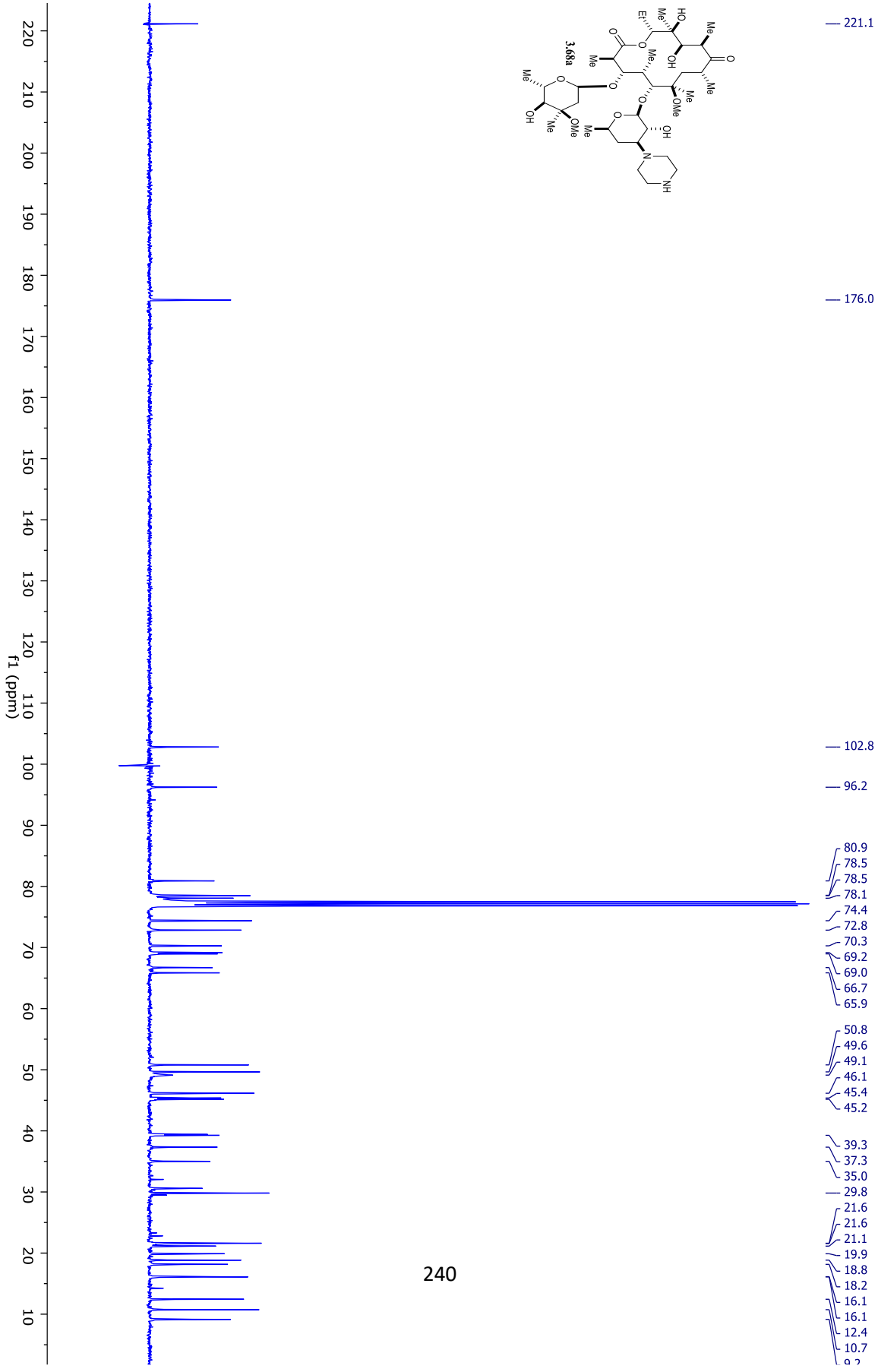

3.67a
f1 (ppm)
1.00
1.01
0.98
2.13
4.29
3.26
1.37
3.23
1.17
1.06
5.03
1.03
1.85
1.38
2.70
1.12
1.00
2.18
1.37
2.16
1.25
1.44
3.06
15.21
11.95
3.04

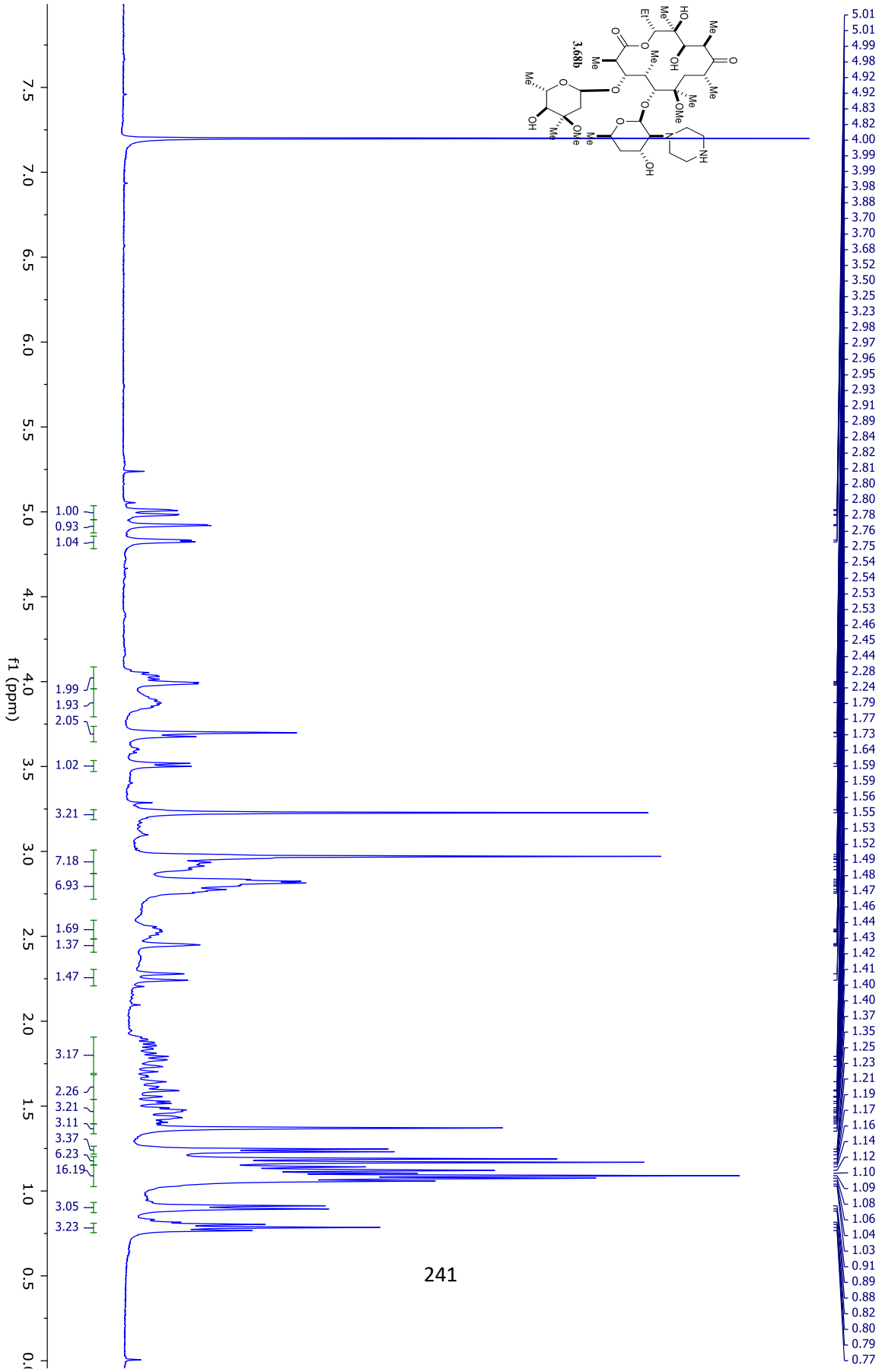

3.67a
f1 (ppm)
221.2
176.0
102.8
96.2
81.0
78.6
78.5
78.1
74.4
72.9
70.2
69.2
68.9
67.5
66.6
65.9
50.8
49.7
49.0
45.4
45.2
39.4
39.3
37.3
35.0
30.4
29.8
21.6
21.6
19.9
18.9
18.2
16.1
16.1
12.5
10.8

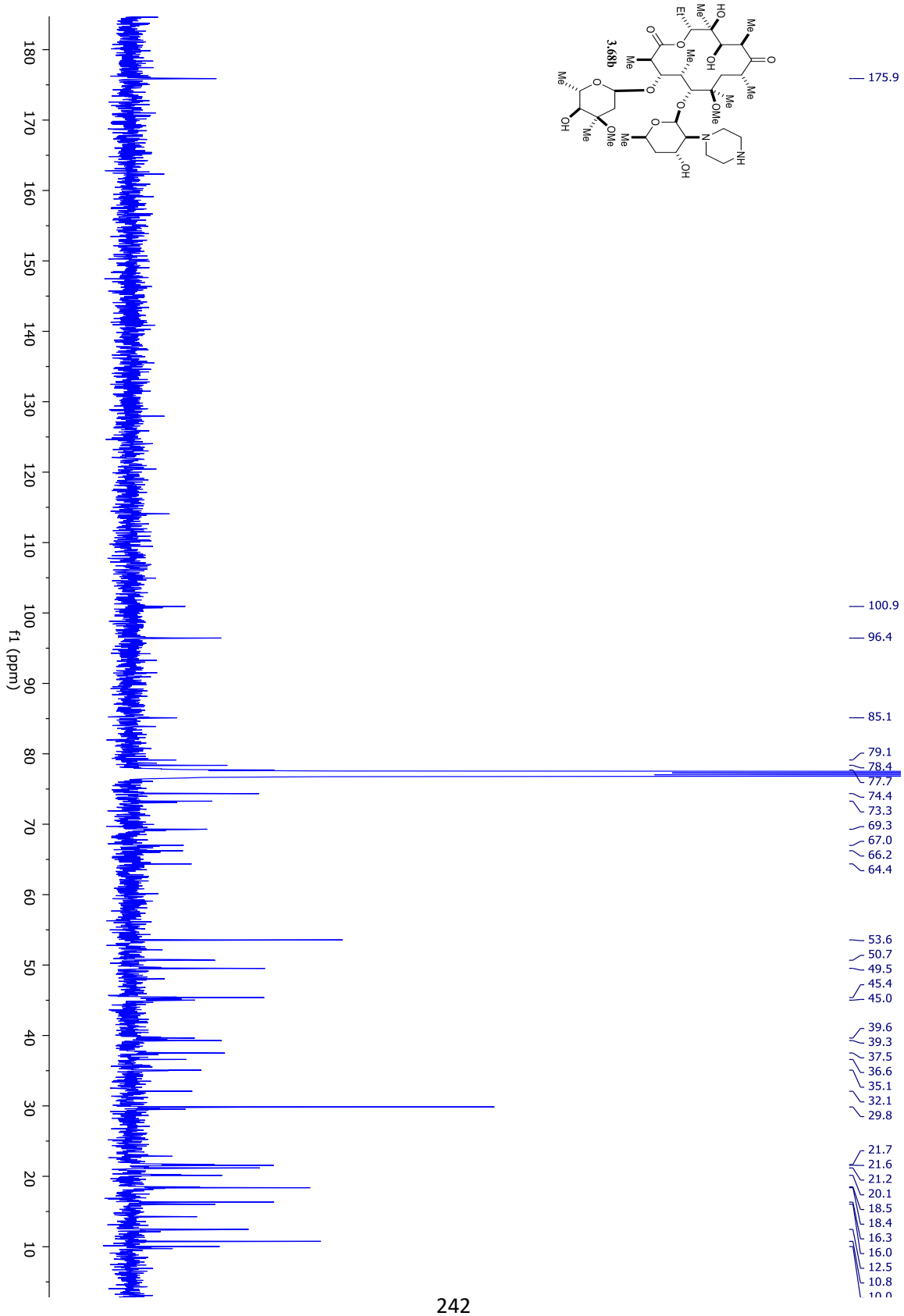

3.67b
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0
1.13
0.87
1.10
1.85
2.40
2.42
4.83
3.24
1.28
7.14
2.85
2.77
1.10
3.57
2.32
2.01
1.17
3.42
3.56
4.19
6.84
9.33
3.07
3.22
5.07
5.07
5.05
5.04
5.01
4.89
4.88
4.09
4.07
4.07
4.06
3.97
3.77
3.76
3.76
3.75
3.67
3.67
3.65
3.60
3.58
3.29
3.21
3.03
3.02
3.01
2.99
2.99
2.98
2.98
2.95
2.85
2.83
2.83
2.81
2.60
2.60
2.59
2.59
2.58
2.58
2.34
2.34
2.31
1.92
1.92
1.87
1.86
1.85
1.80
1.79
1.77
1.70
1.67
1.64
1.58
1.57
1.55
1.54
1.49
1.49
1.48
1.47
1.46
1.43
1.31
1.29
1.25
1.25
1.23
1.21
1.20
1.19
1.18
1.17
1.15
1.14
1.13
1.13
1.12
1.11
1.10
0.98
0.96
0.86
0.84
0.83

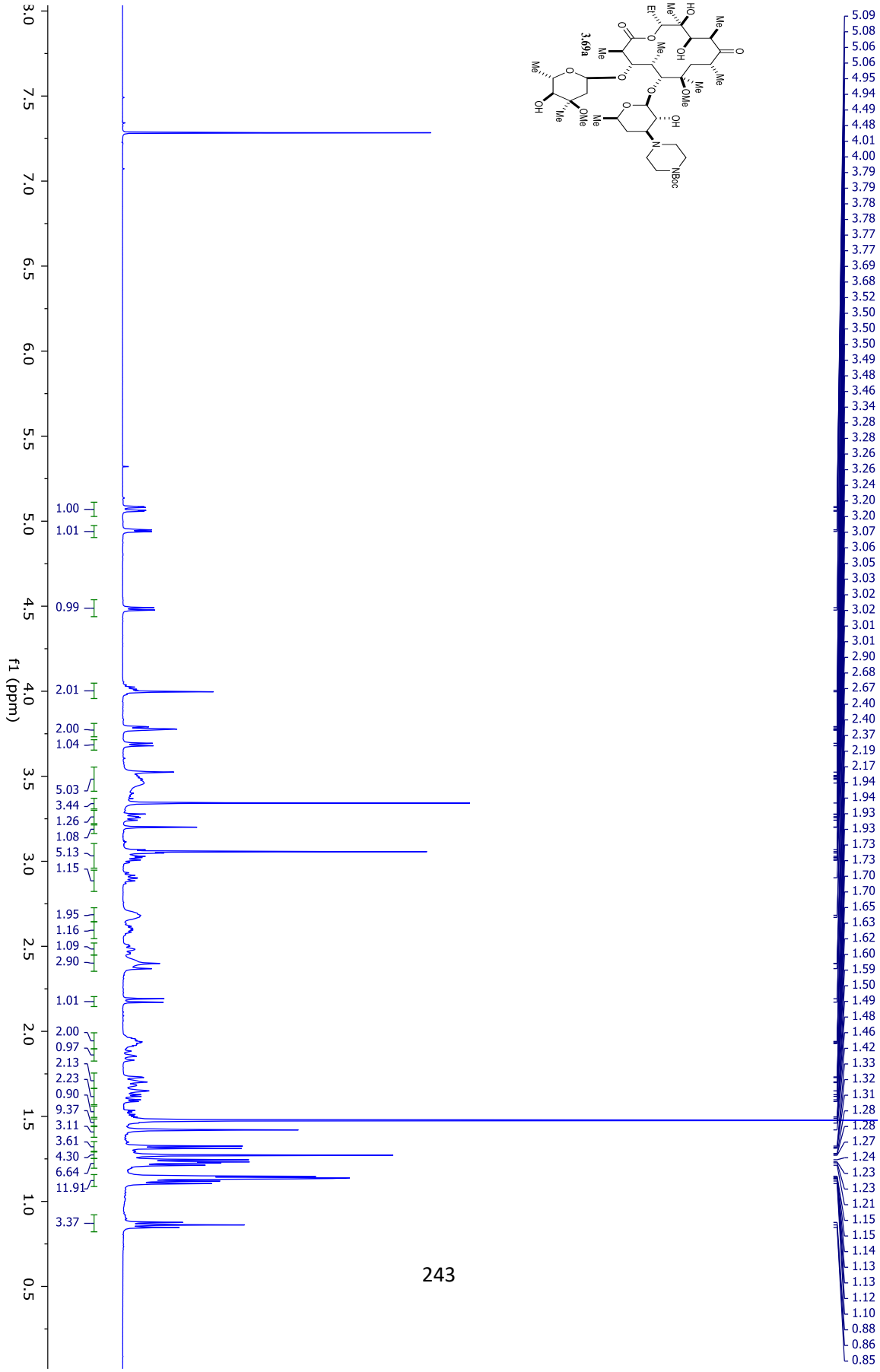

3.67b
f1 (ppm)
175.9
96.4
84.1
78.7
78.4
77.9
76.8
74.3
73.1
73.1
69.6
69.2
68.1
66.8
66.0
64.8
52.8
50.8
49.5
45.4
45.2
39.8
39.4
37.5
36.6
35.0
29.8
21.7
21.6
21.2
20.2
18.5
18.3
16.3
16.1
12.4
10.8
9.8

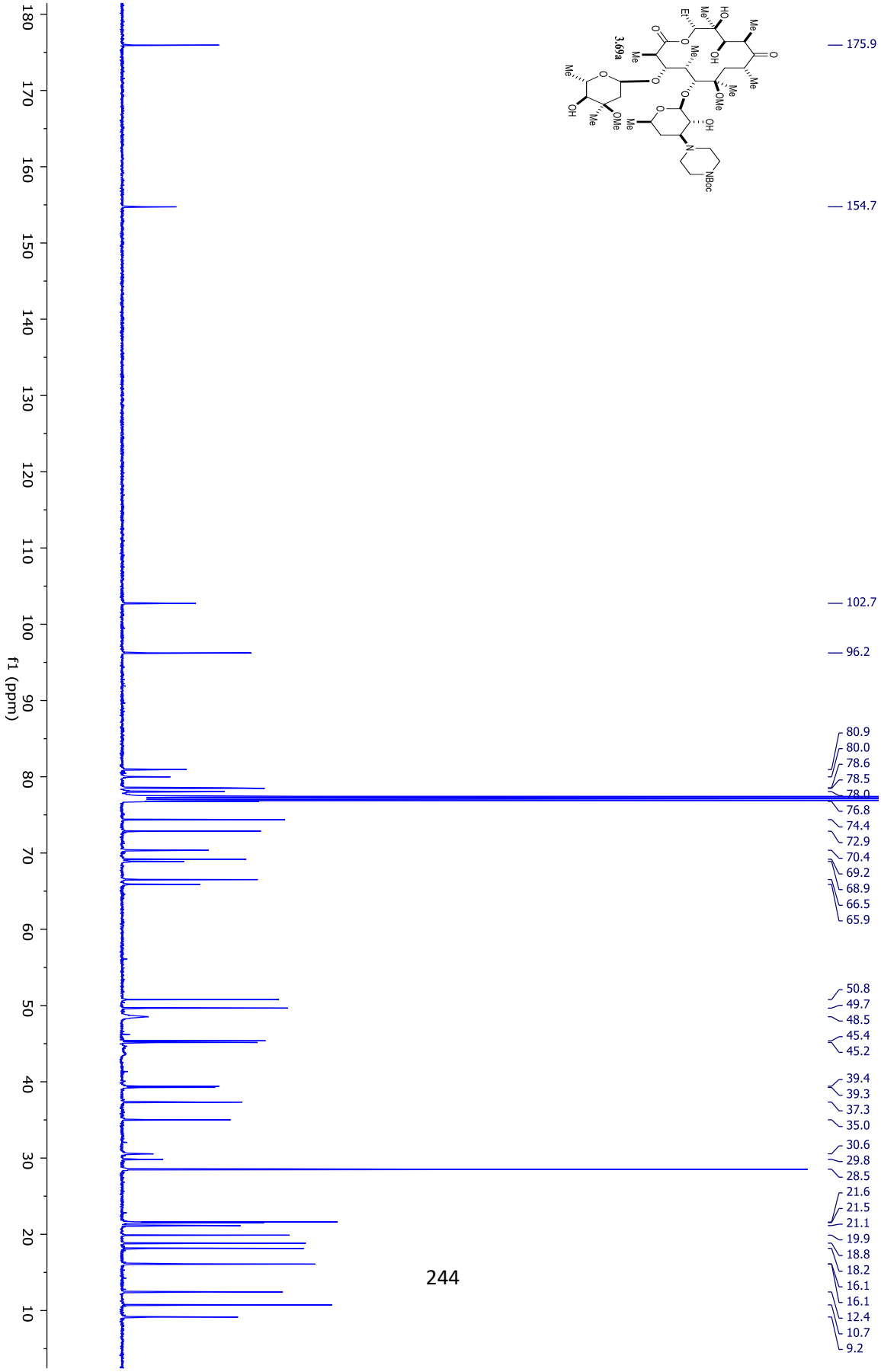

3.68a
f1 (ppm)
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
0.99
0.98
1.11
2.21
1.40
1.99
3.77
1.71
10.36
2.12
1.15
4.34
3.21
1.93
1.04
1.01
3.01
16.82
13.39
3.86

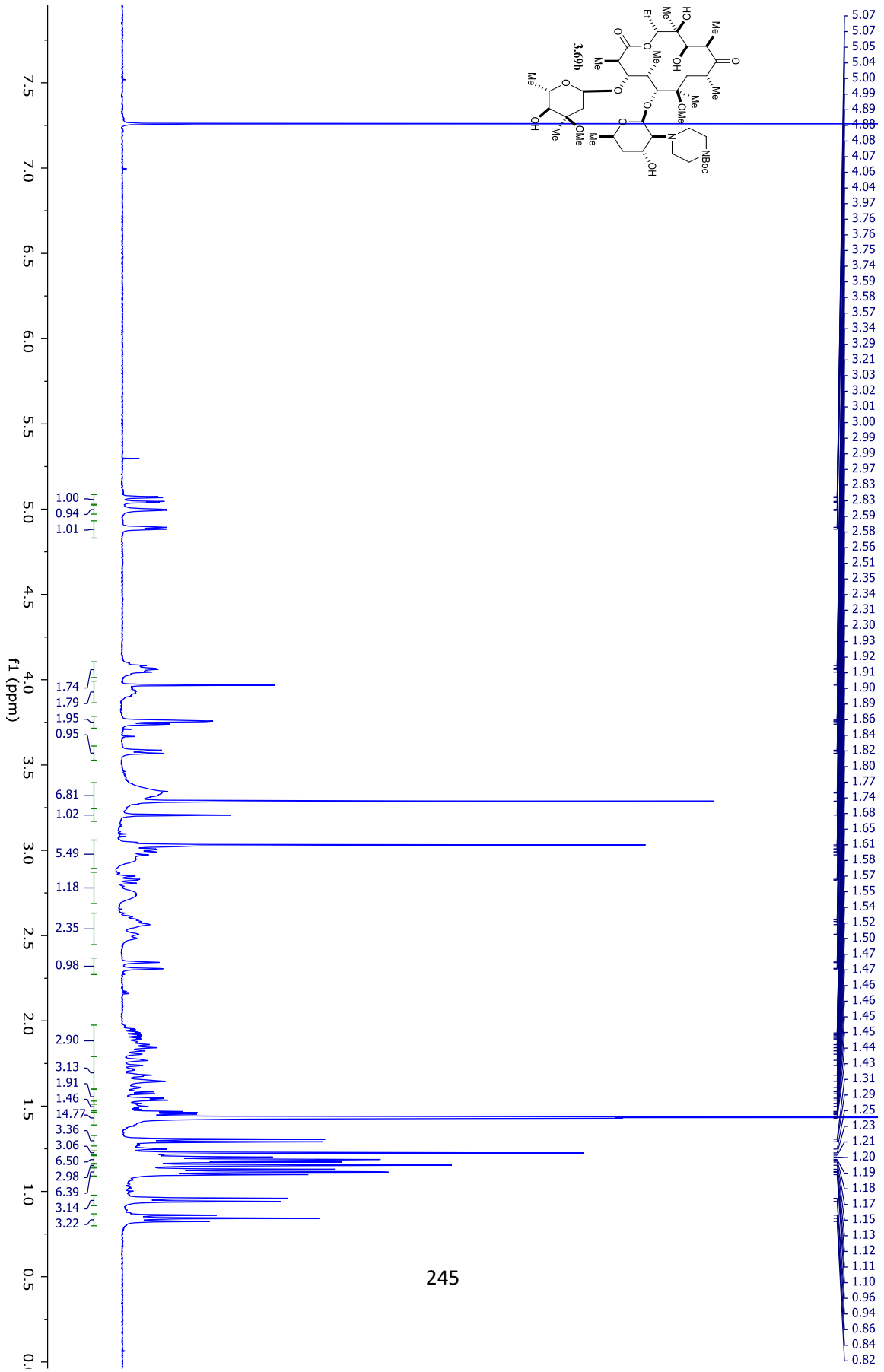

3.68a
f1 (ppm)
221.1
176.0
102.8
96.2
80.9
78.5
78.5
78.1
74.4
72.8
70.3
69.2
69.0
66.7
65.9
50.8
49.6
49.1
46.1
45.4
45.2
39.3
37.3
35.0
29.8
21.6
21.6
21.1
19.9
18.8
18.2
16.1
16.1
12.4
10.7

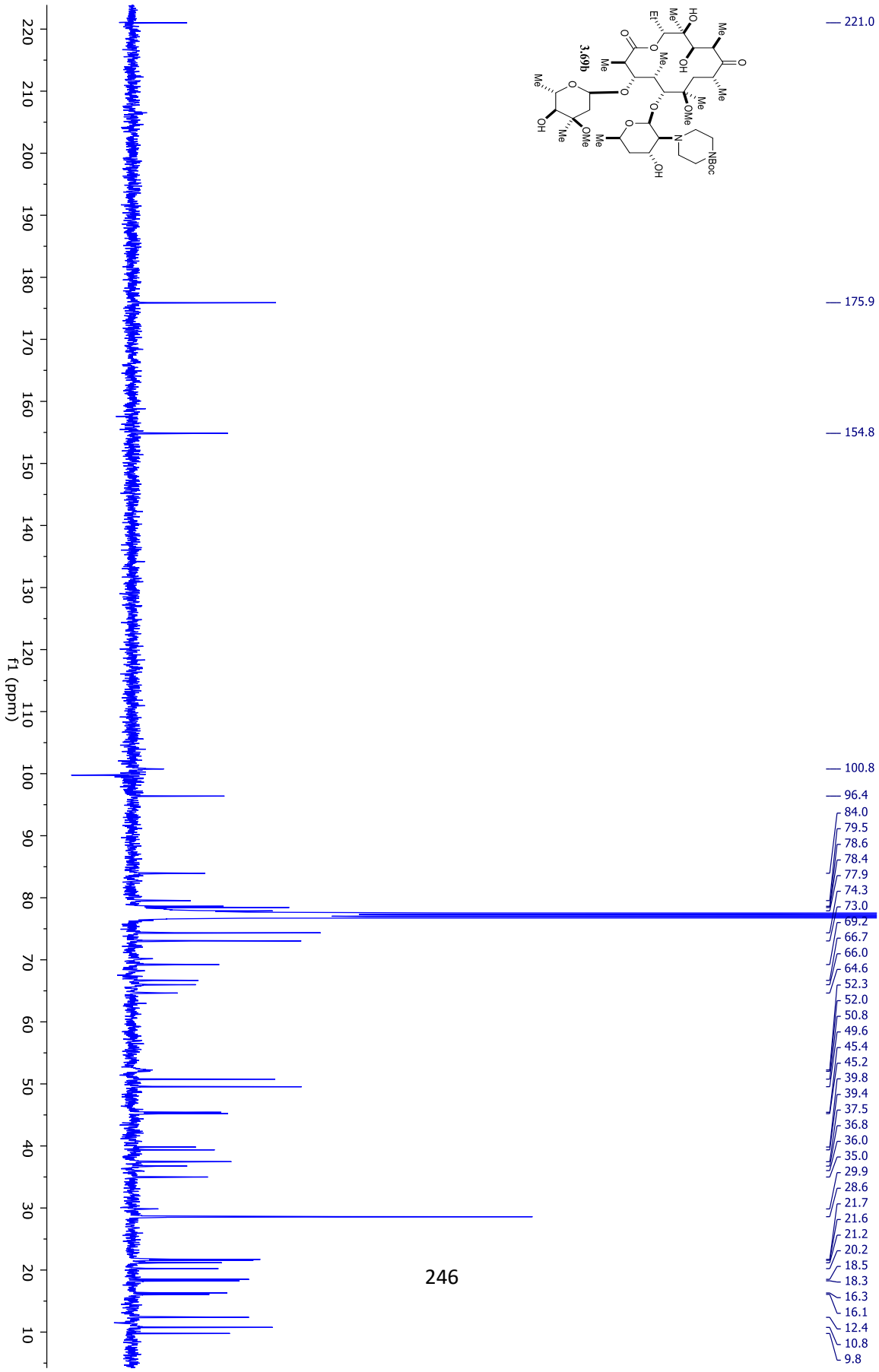

3.68b
f1 (ppm)
1.00
0.93
1.04
1.99
1.93
2.05
1.02
3.21
7.18
6.93
1.69
1.37
1.47
3.17
2.26
3.21
3.11
3.37
6.23
16.19
3.05
3.23

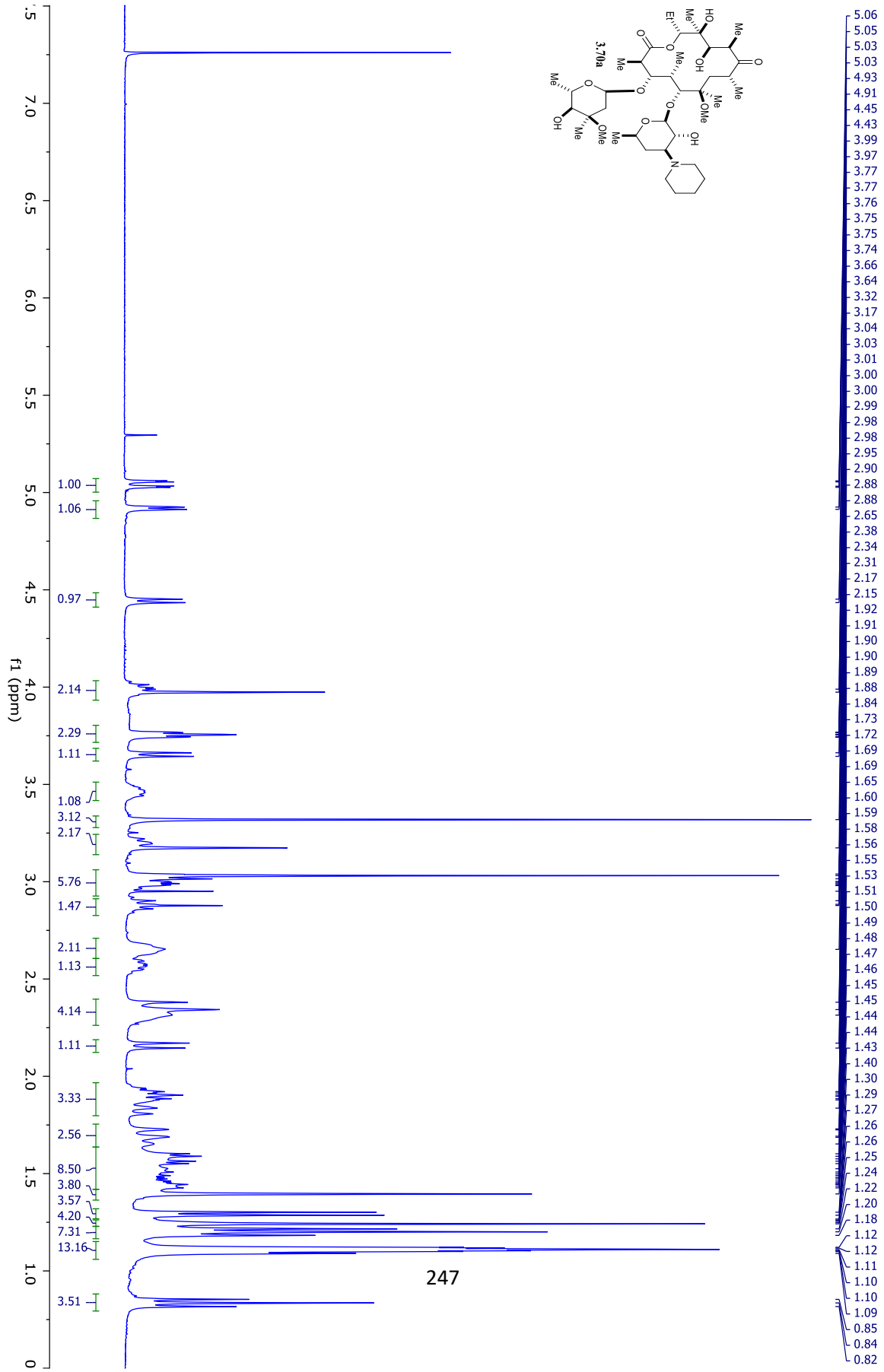

3.68b
f1 (ppm)
175.9
100.9
96.4
85.1
79.1
78.4
77.7
74.4
73.3
69.3
67.0
66.2
64.4
53.6
50.7
49.5
45.4
45.0
39.6
39.3
37.5
36.6
35.1
32.1
29.8
21.7
21.6
21.2
20.1
18.5
18.4
16.3
16.0
12.5
10.8

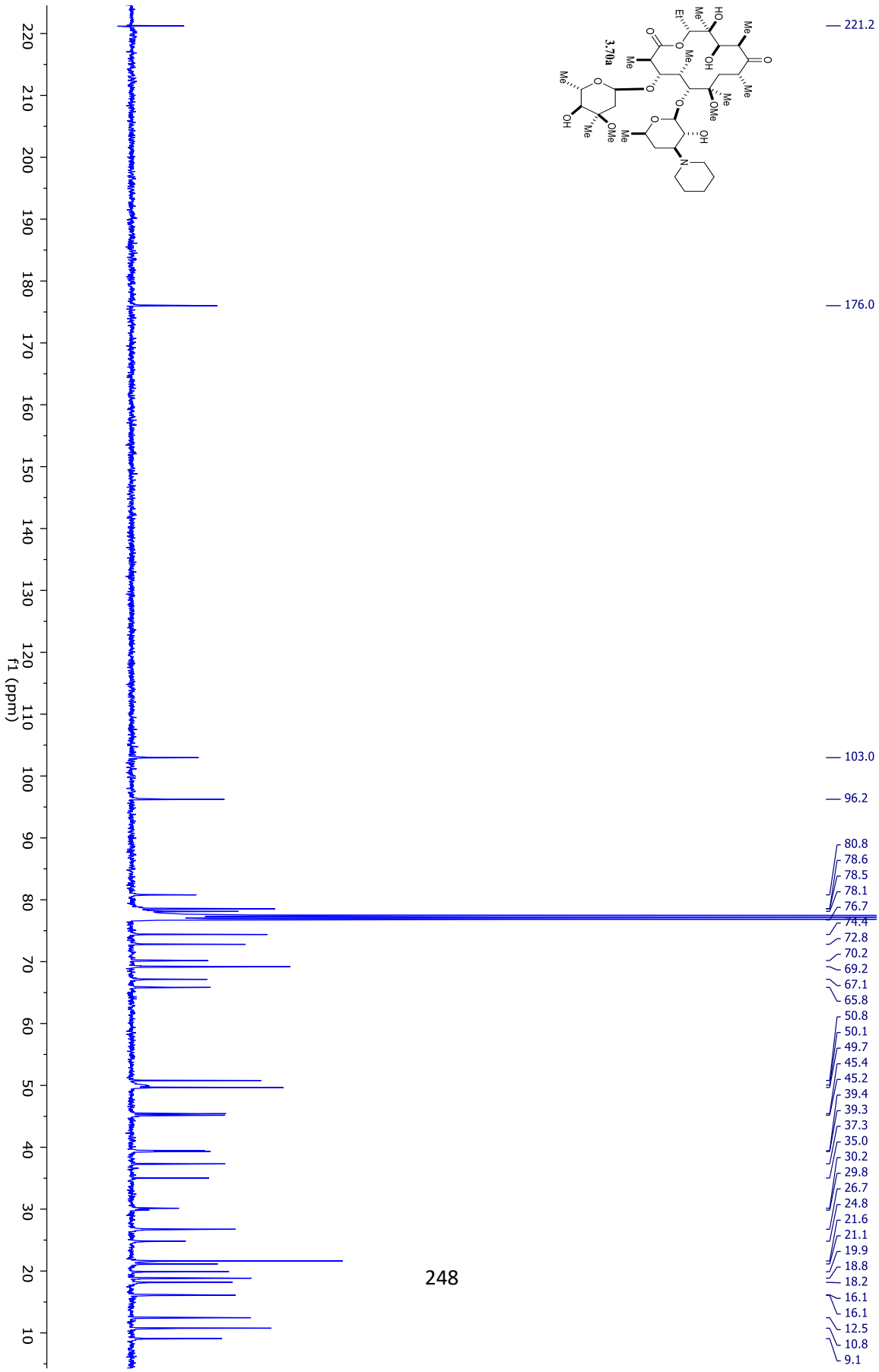

3.69a
f1 (ppm)
1.00
1.01
0.99
2.01
2.00
1.04
5.03
3.44
1.26
1.08
5.13
1.15
1.95
1.16
1.09
2.90
1.01
2.00
0.97
2.13
2.23
0.90
9.37
3.11
3.61
4.30
6.64
11.91
3.37

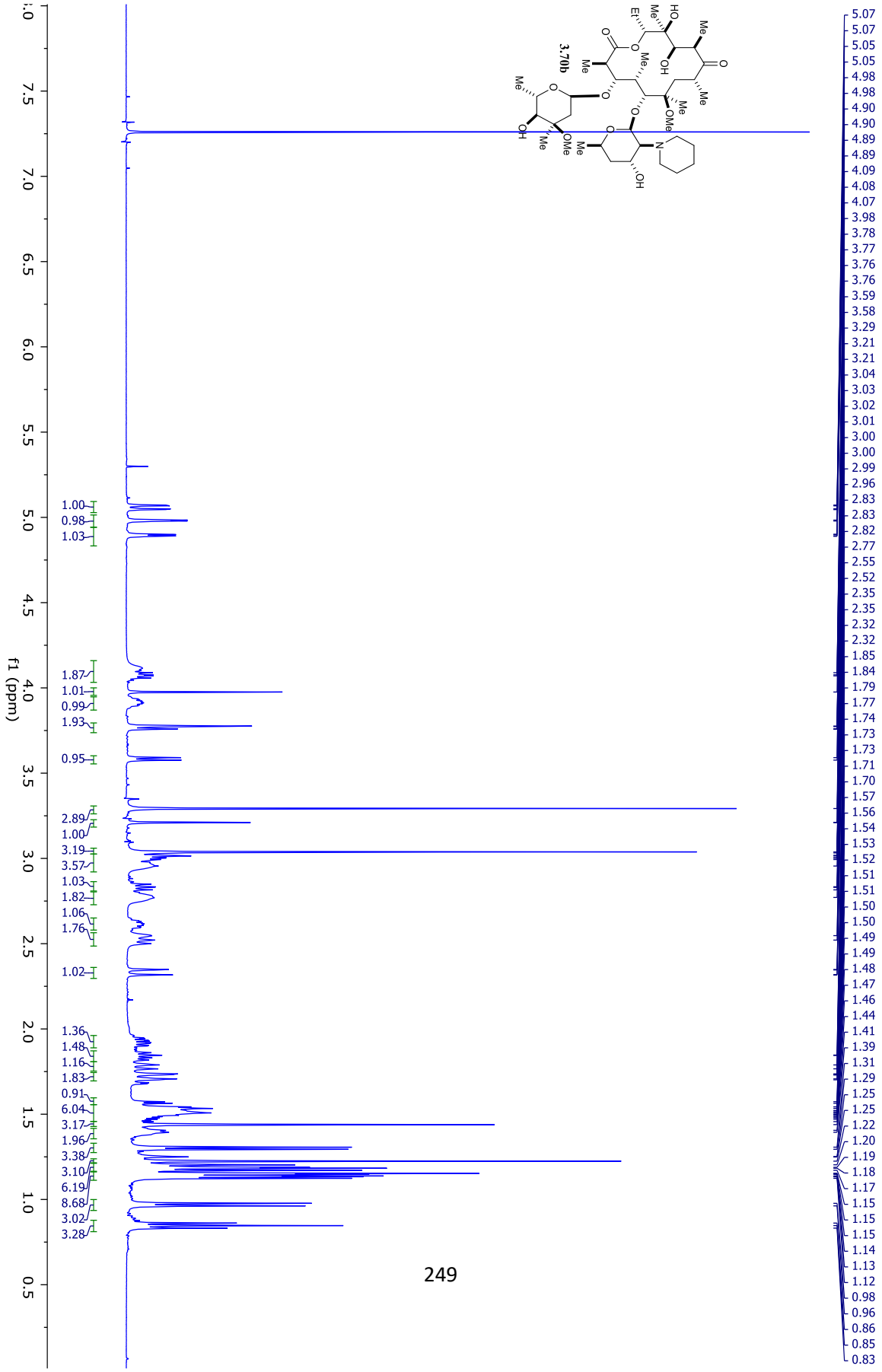

3.69a
f1 (ppm)
175.9
154.7
102.7
96.2
80.9
80.0
78.6
78.5
78.0
76.8
74.4
72.9
70.4
69.2
68.9
66.5
65.9
50.8
49.7
48.5
45.4
45.2
39.4
39.3
37.3
35.0
30.6
29.8
28.5
21.6
21.5
21.1
19.9
18.8
18.2
16.1
16.1
12.4
10.7
9.2

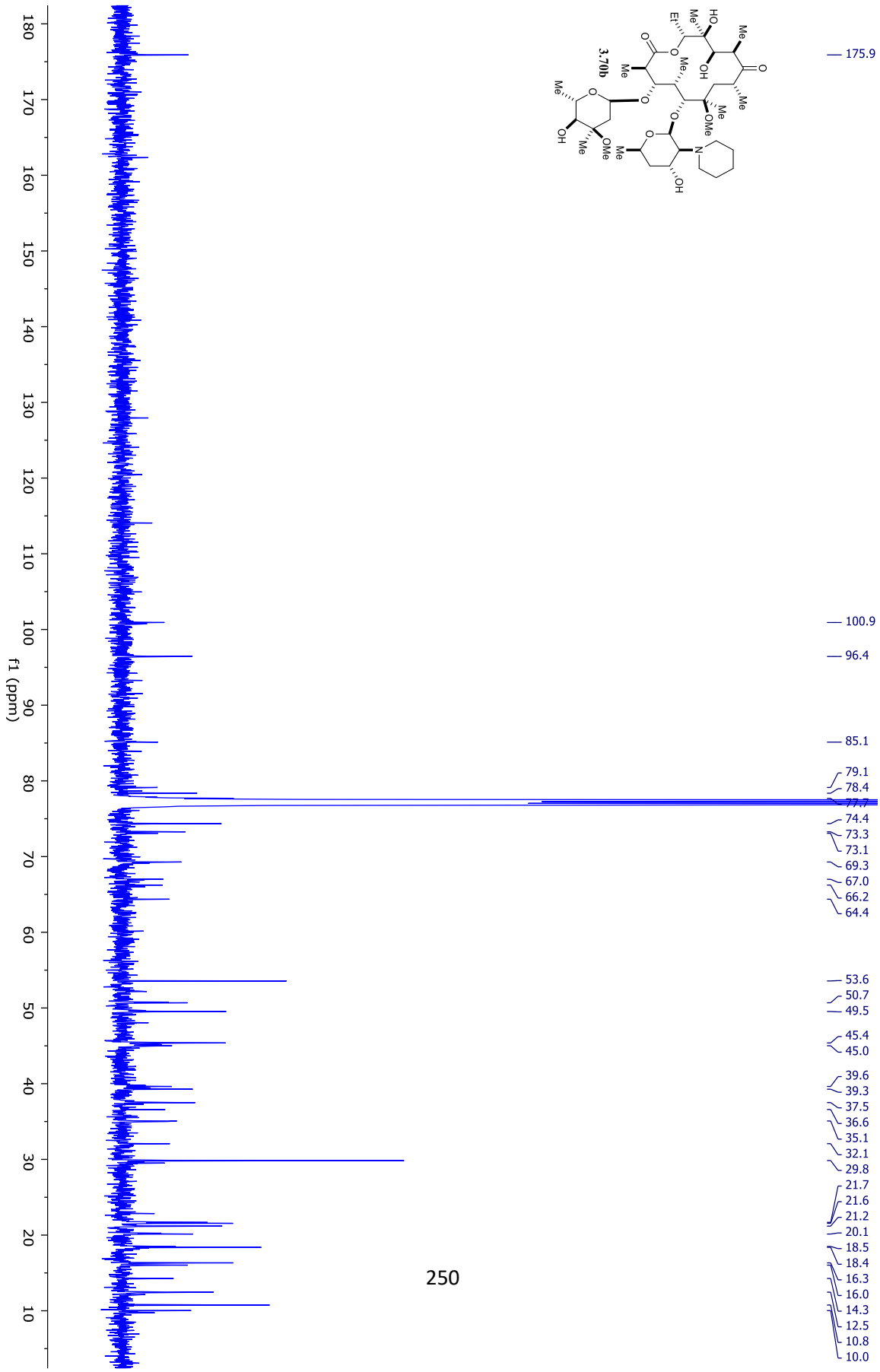

3.69b
f1 (ppm)
1.00
0.94
1.01
1.74
1.79
1.95
0.95
6.81
1.02
5.49
1.18
2.35
0.98
2.90
3.13
1.91
1.46
14.77
3.36
3.06
6.50
2.98
6.39
3.14
3.22

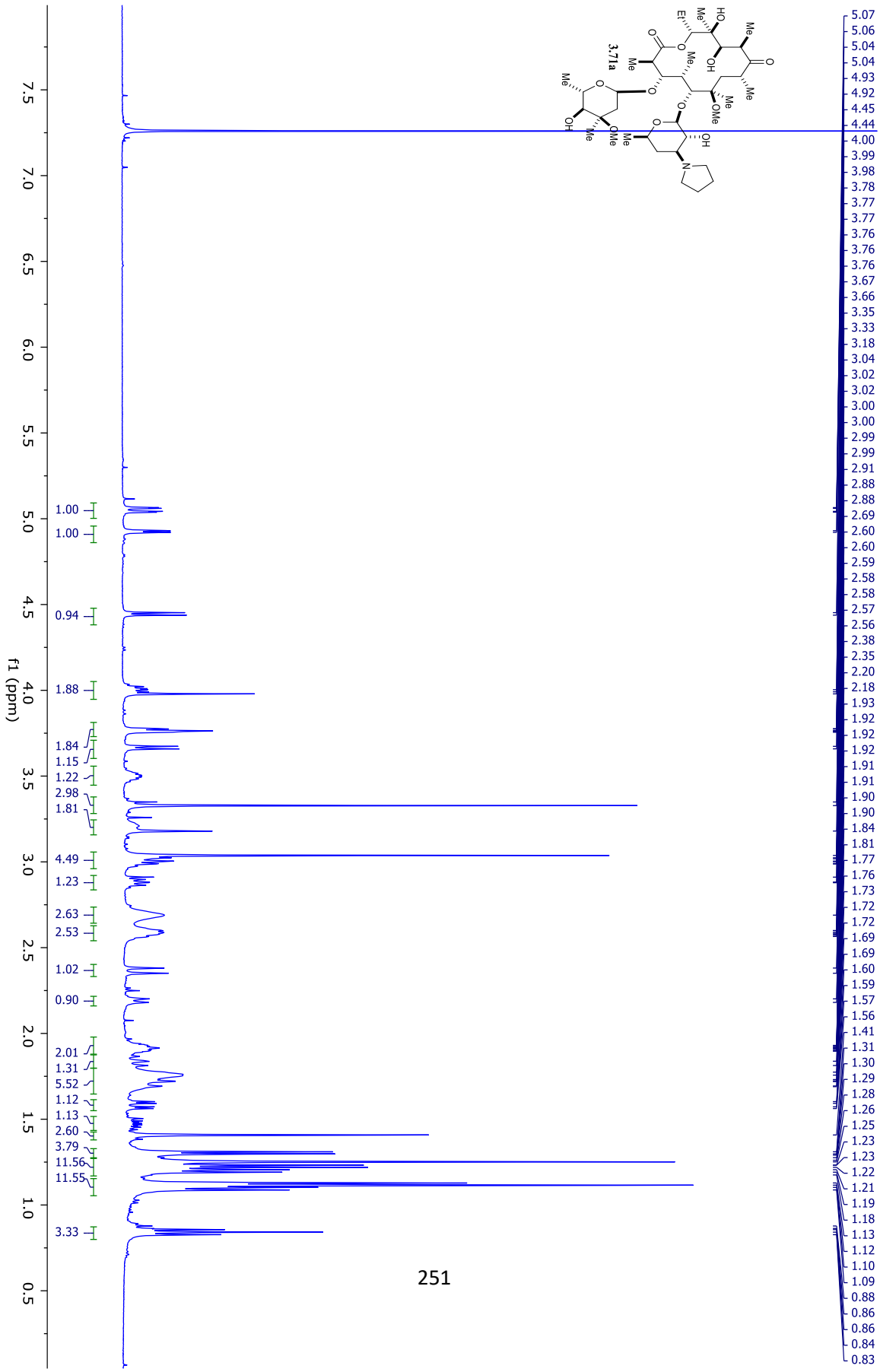

3.69b
f1 (ppm)
221.0
175.9
154.8
100.8
96.4
84.0
79.5
78.6
78.4
77.9
74.3
73.0
69.2
66.7
66.0
64.6
52.3
52.0
50.8
49.6
45.4
45.2
39.8
39.4
37.5
36.8
36.0
35.0
29.9
28.6
21.7
21.6
21.2
20.2
18.5
18.3
16.3
16.1
12.4
10.8
9.8

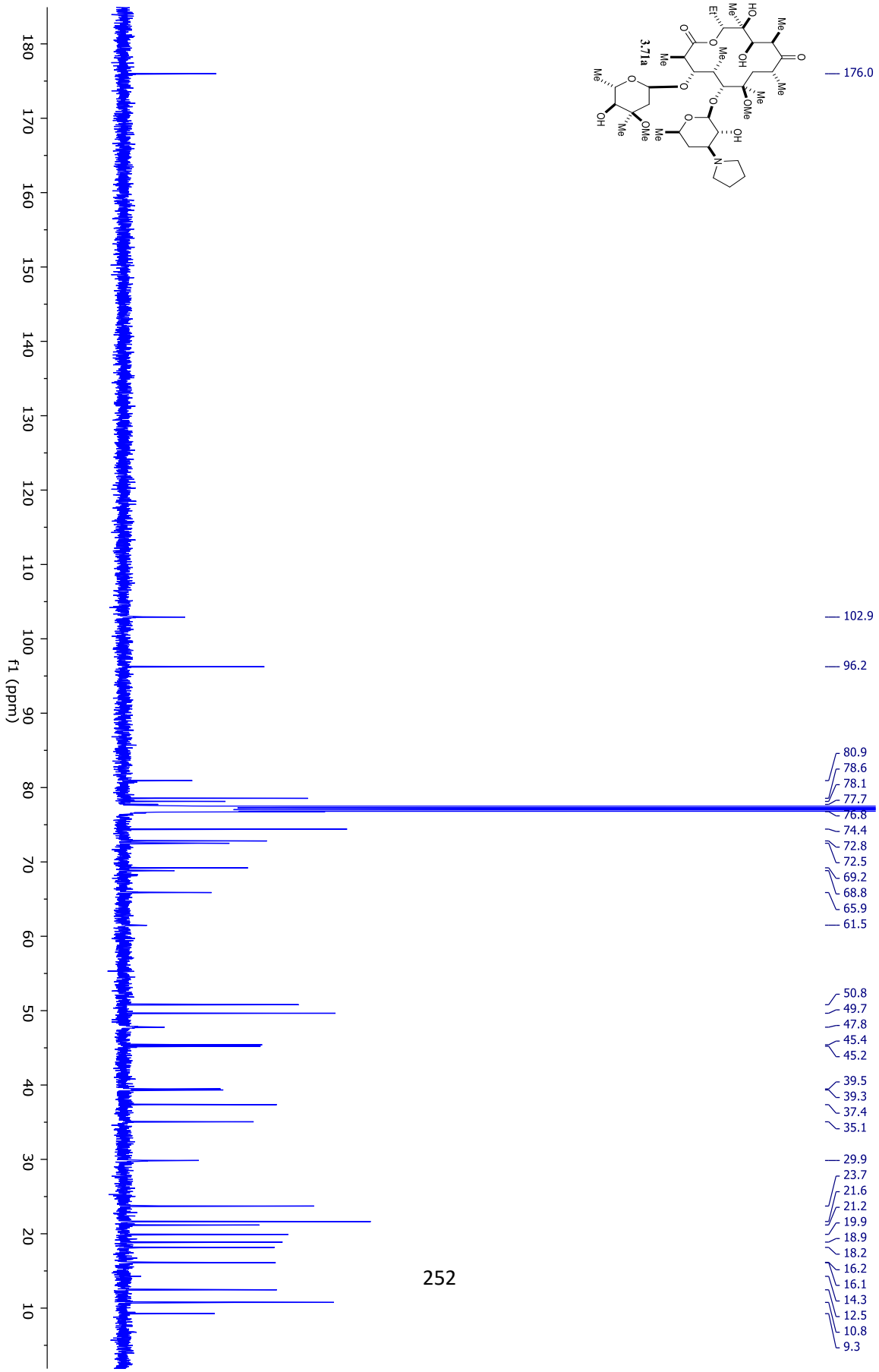

3.70a
f1 (ppm)
1.00
1.06
0.97
2.14
2.29
1.11
1.08
3.12
2.17
5.76
1.47
2.11
1.13
4.14
1.11
3.33
2.56
8.50
3.80
3.57
4.20
7.31
13.16
3.51

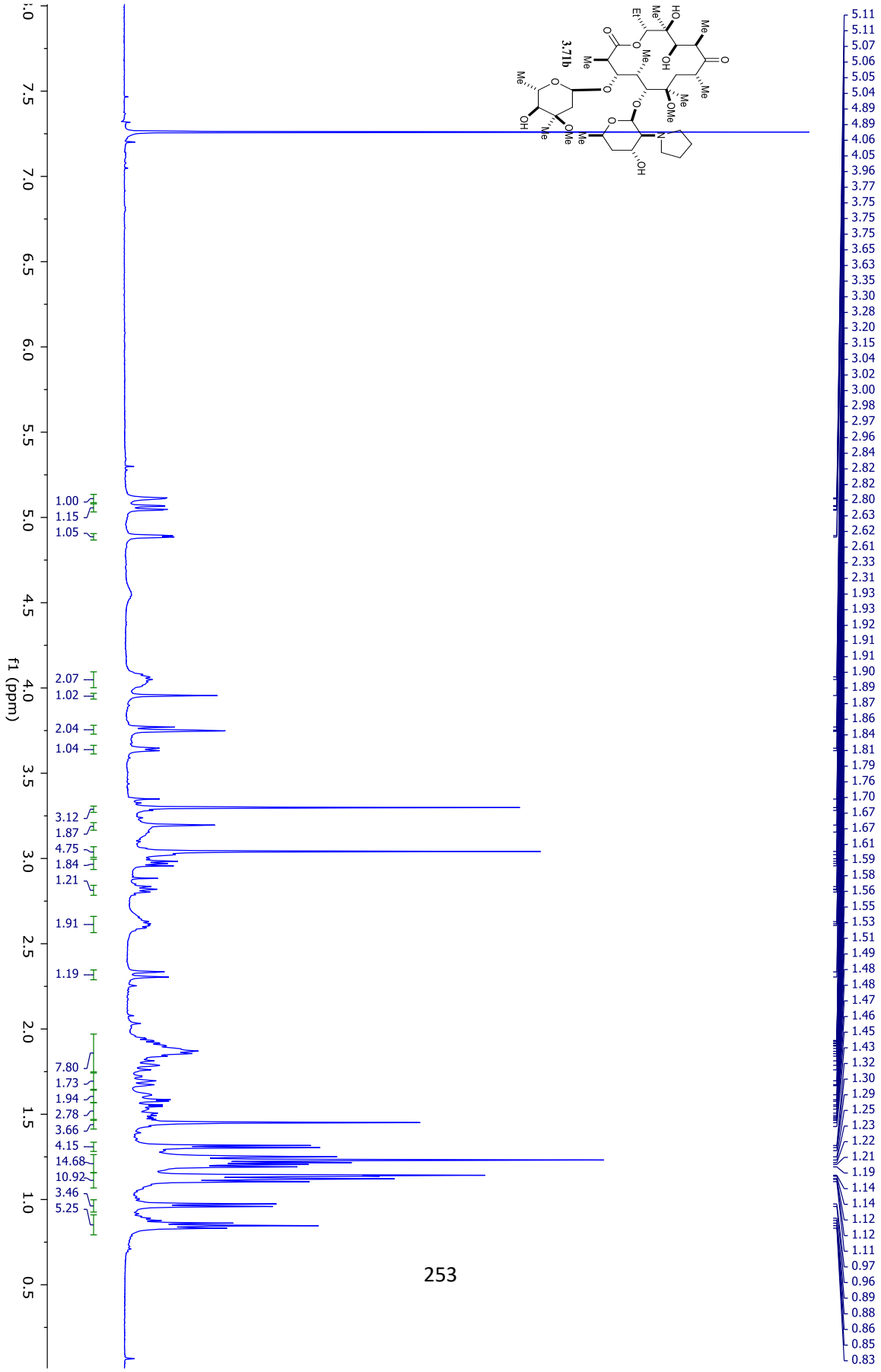

3.70a
221.2
176.0
103.0
96.2
80.8
78.6
78.5
78.1
76.7
74.4
72.8
70.2
69.2
67.1
65.8
50.8
50.1
49.7
45.4
45.2
39.4
39.3
37.3
35.0
30.2
29.8
26.7
24.8
21.6
21.1
19.9
18.8
18.2
16.1
16.1
12.5
10.8
9.1
f1 (ppm)

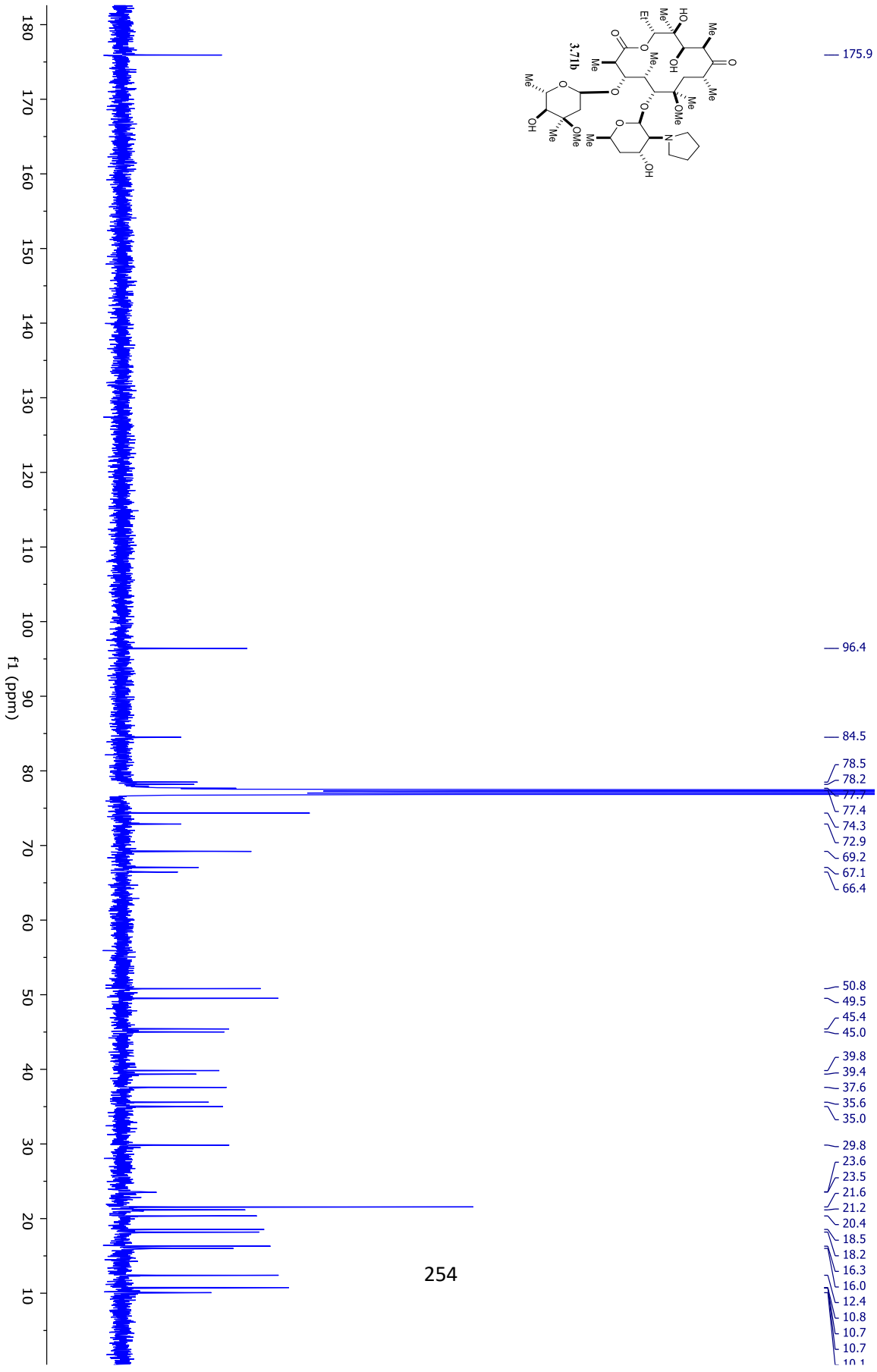

3.70b
f1 (ppm)
1.00
0.98
1.03
1.87
1.01
0.99
1.93
0.95
2.89
1.00
3.19
3.57
1.03
1.82
1.06
1.76
1.02
1.36
1.48
1.16
1.83
0.91
6.04
3.17
1.96
3.38
3.10
6.19
8.68
3.02
3.28

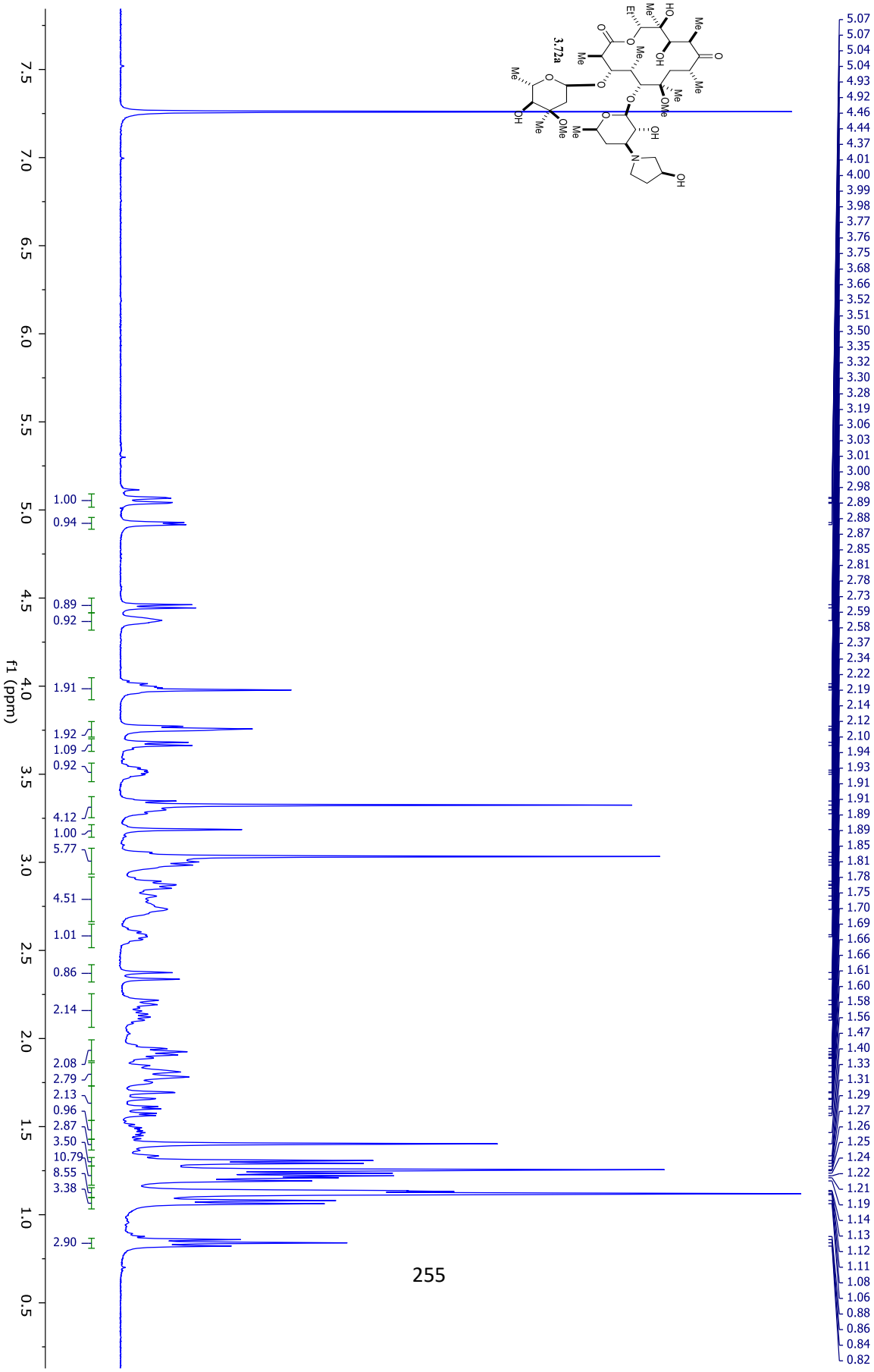

3.70b
175.9
100.9
96.4
85.1
79.1
78.4
77.7
74.4
73.3
73.1
69.3
67.0
66.2
64.4
53.6
50.7
49.5
45.4
45.0
39.6
39.3
37.5
36.6
35.1
32.1
29.8
21.7
21.6
21.2
20.1
18.5
18.4
16.3
16.0
14.3
12.5
10.8
10.0
f1 (ppm)

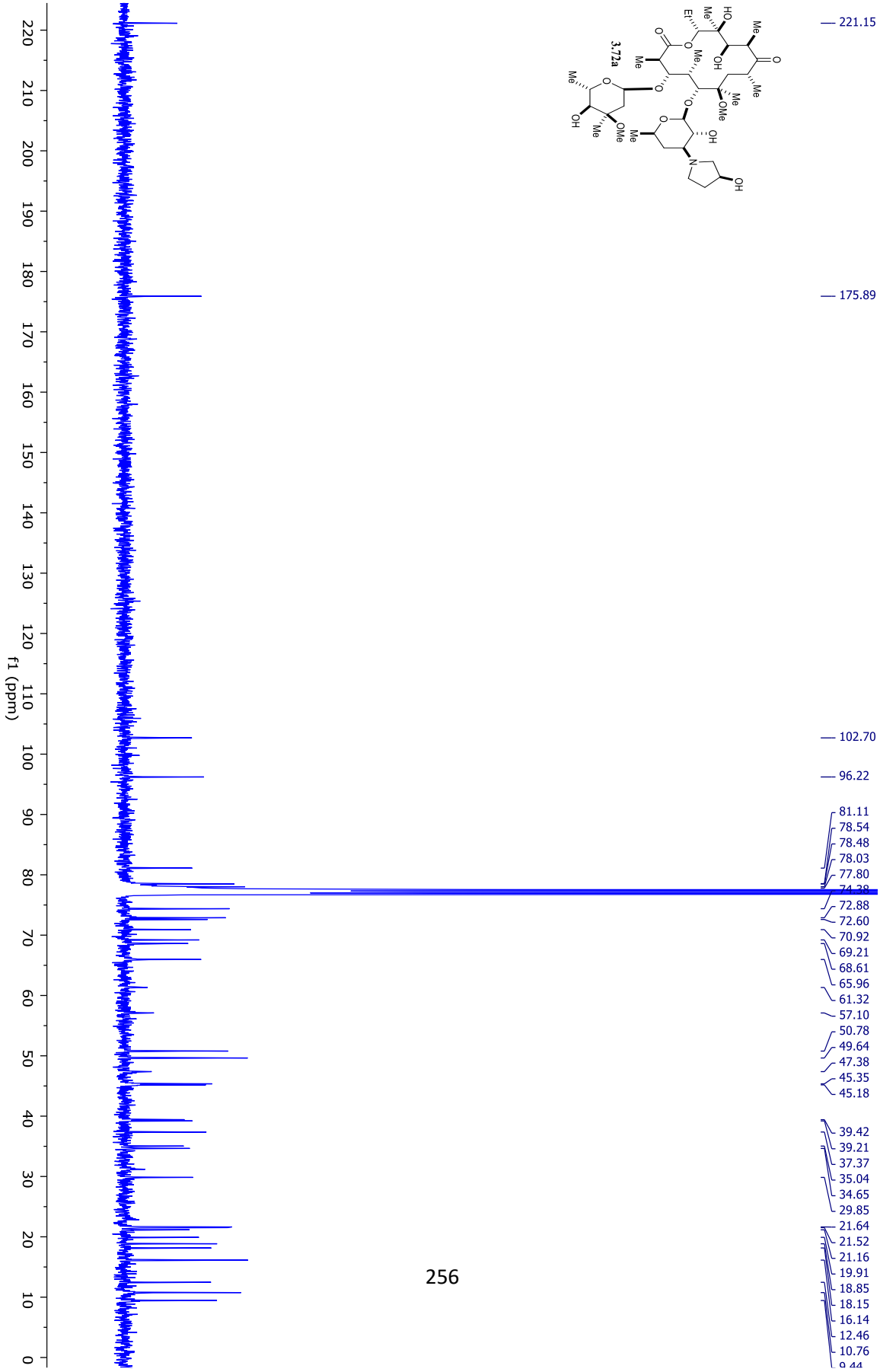

3.71a
f1 (ppm)
1.00
1.00
0.94
1.88
1.84
1.15
1.22
2.98
1.81
4.49
1.23
2.63
2.53
1.02
0.90
2.01
1.31
5.52
1.12
1.13
2.60
3.79
11.56
11.55
3.33

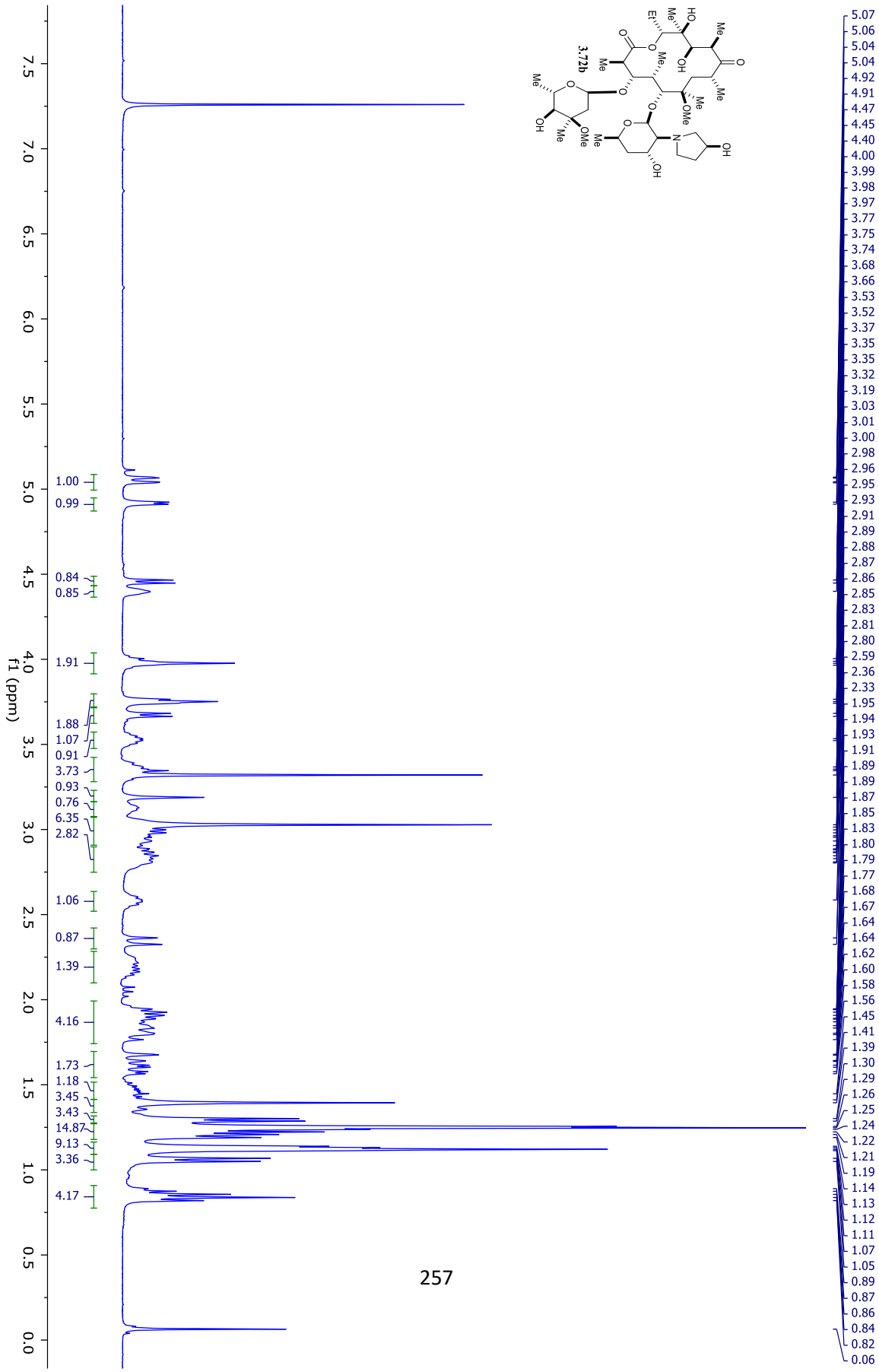

3.71a
f1 (ppm)
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
176.0
102.9
96.2
80.9
78.6
78.1
77.7
76.8
74.4
72.8
72.5
69.2
68.8
65.9
61.5
50.8
49.7
47.8
45.4
45.2
39.5
39.3
37.4
35.1
29.9
23.7
21.6
21.2
19.9
18.9
18.2
16.2
16.1
14.3
12.5
10.8
9.3

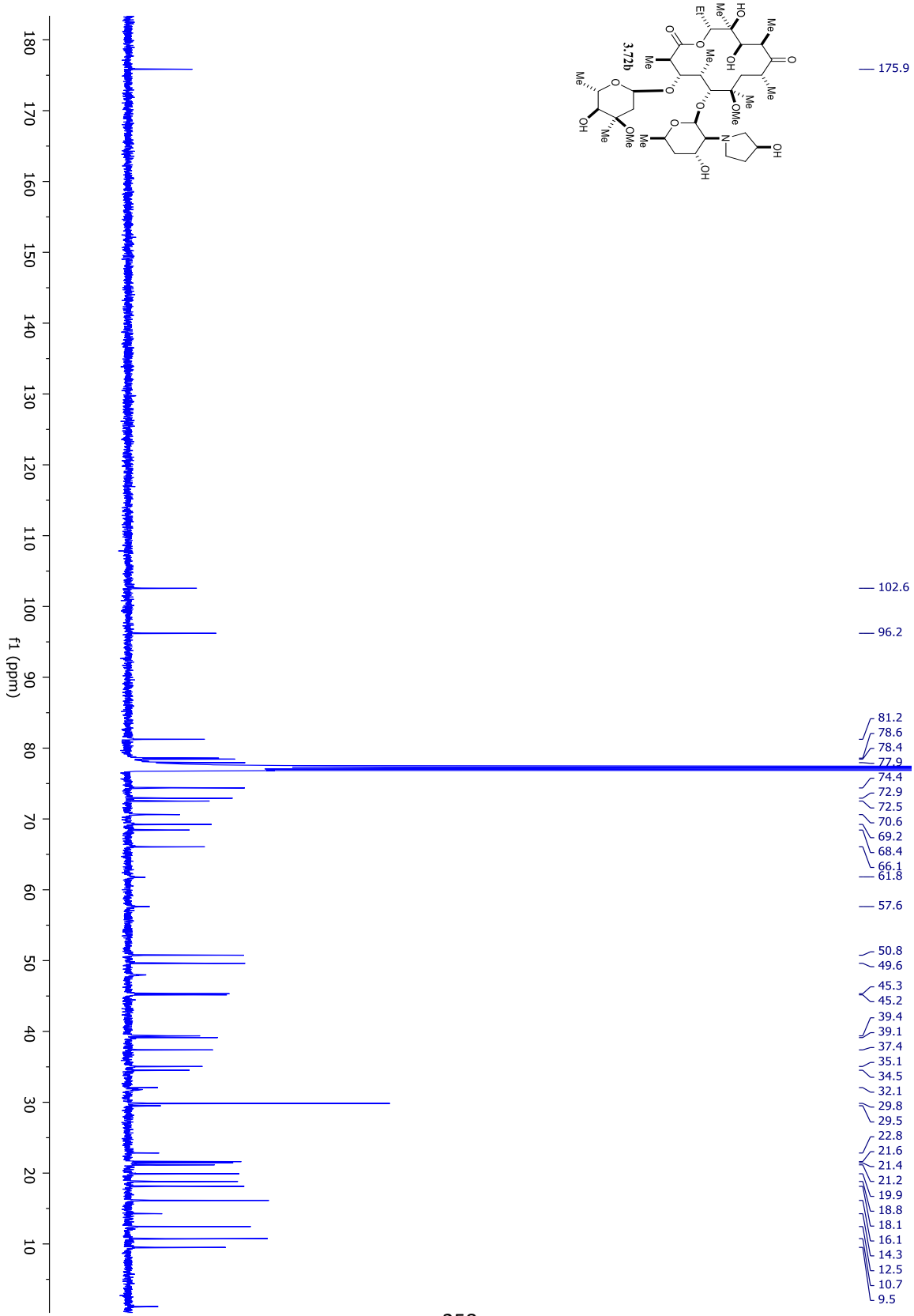

3.71b
f1 (ppm)
1.00
1.15
1.05
2.07
1.02
2.04
1.04
3.12
1.87
4.75
1.84
1.21
1.91
1.19
7.80
1.73
1.94
2.78
3.66
4.15
14.68
10.92
3.46
5.25

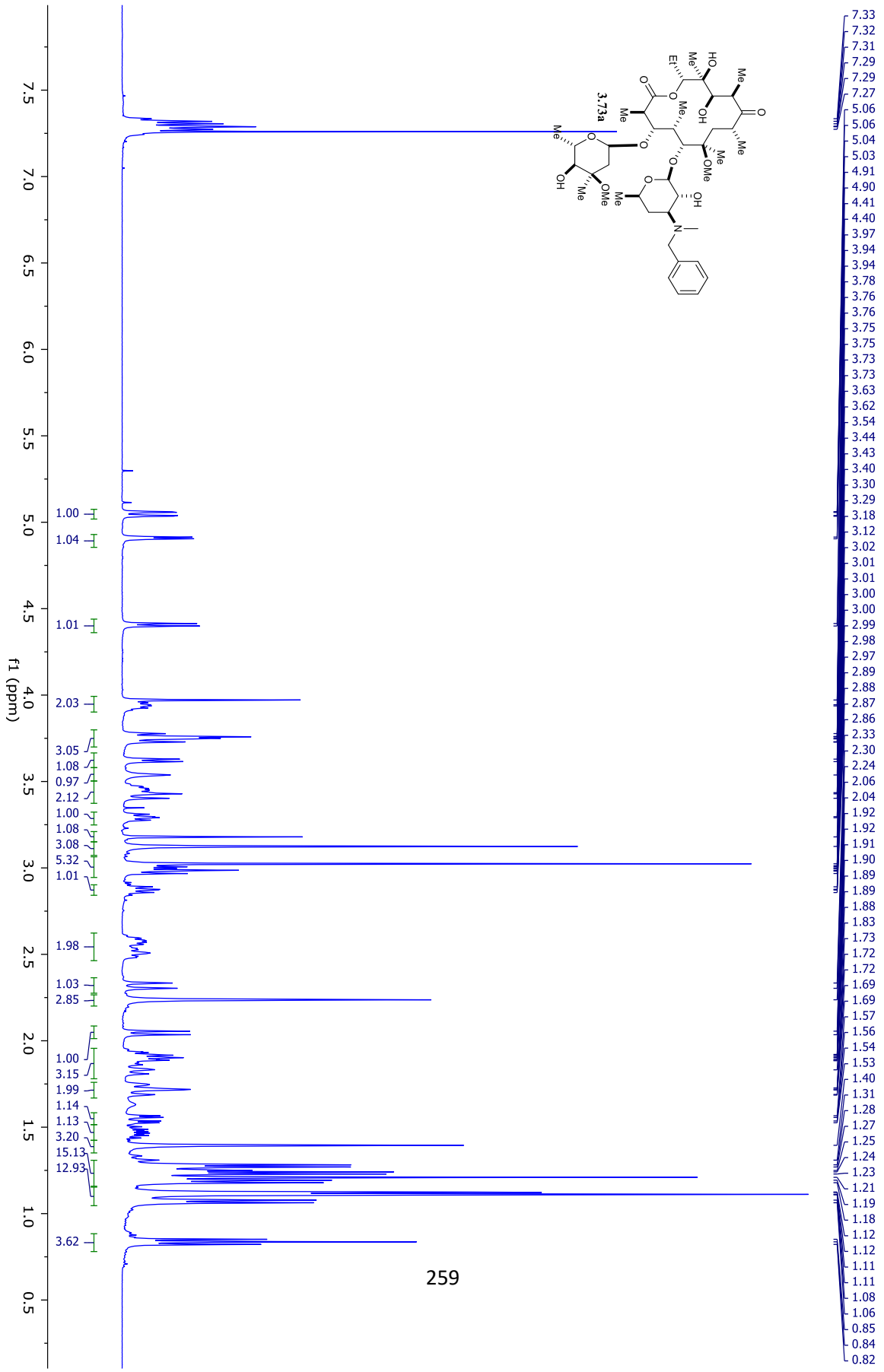

3.71b
f1 (ppm)
175.9
96.4
84.5
78.5
78.2
77.4
74.3
72.9
69.2
67.1
66.4
50.8
49.5
45.4
45.0
39.8
39.4
37.6
35.6
35.0
29.8
23.6
23.5
21.6
21.2
20.4
18.5
18.2
16.3
16.0
12.4
10.8
10.7
10.7

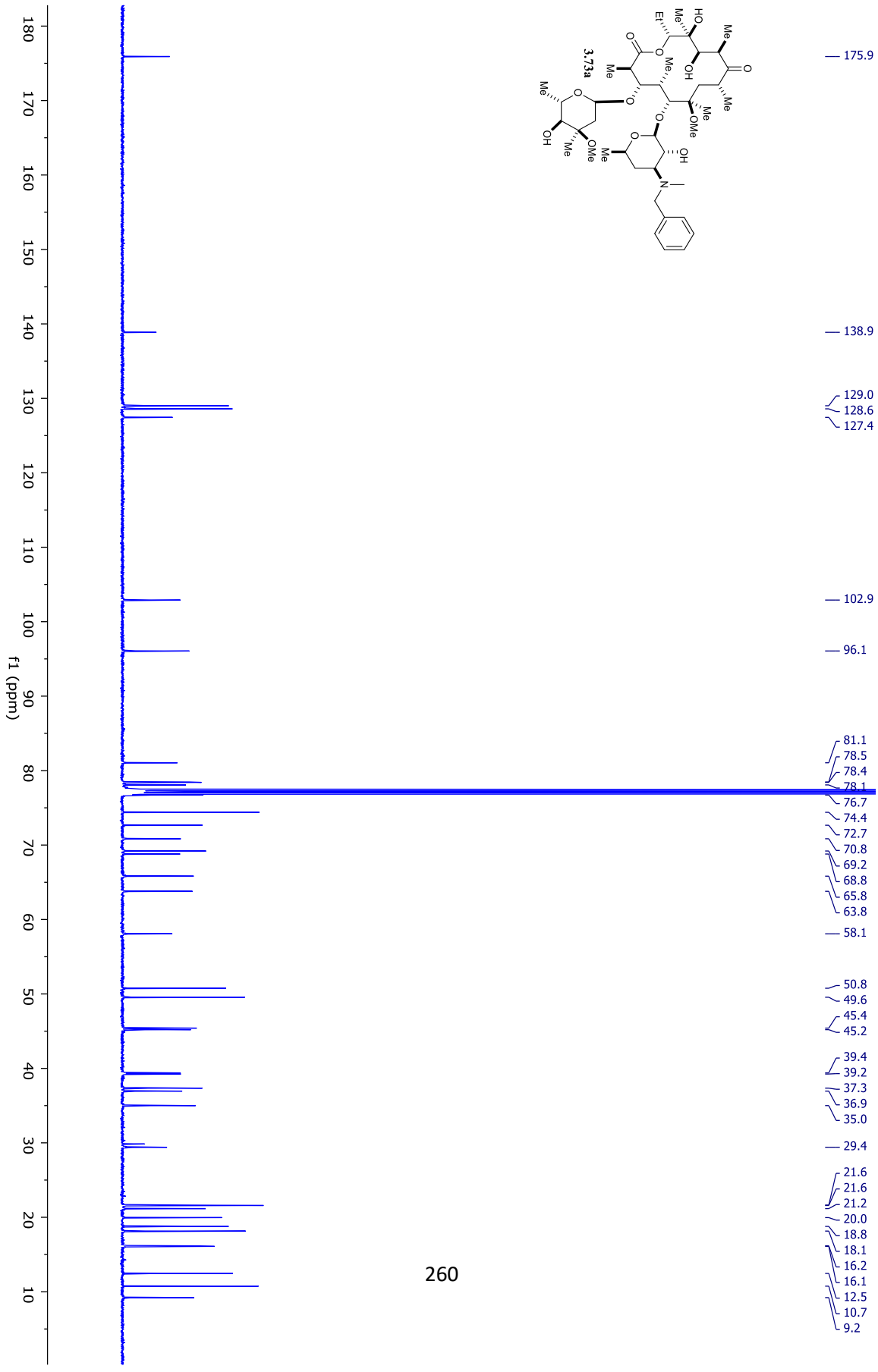

3.72a
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
0.94
0.89
0.92
1.91
1.92
1.09
0.92
4.12
1.00
5.77
4.51
1.01
0.86
2.14
2.08
2.79
2.13
0.96
2.87
3.50
10.79
8.55
3.38
2.90

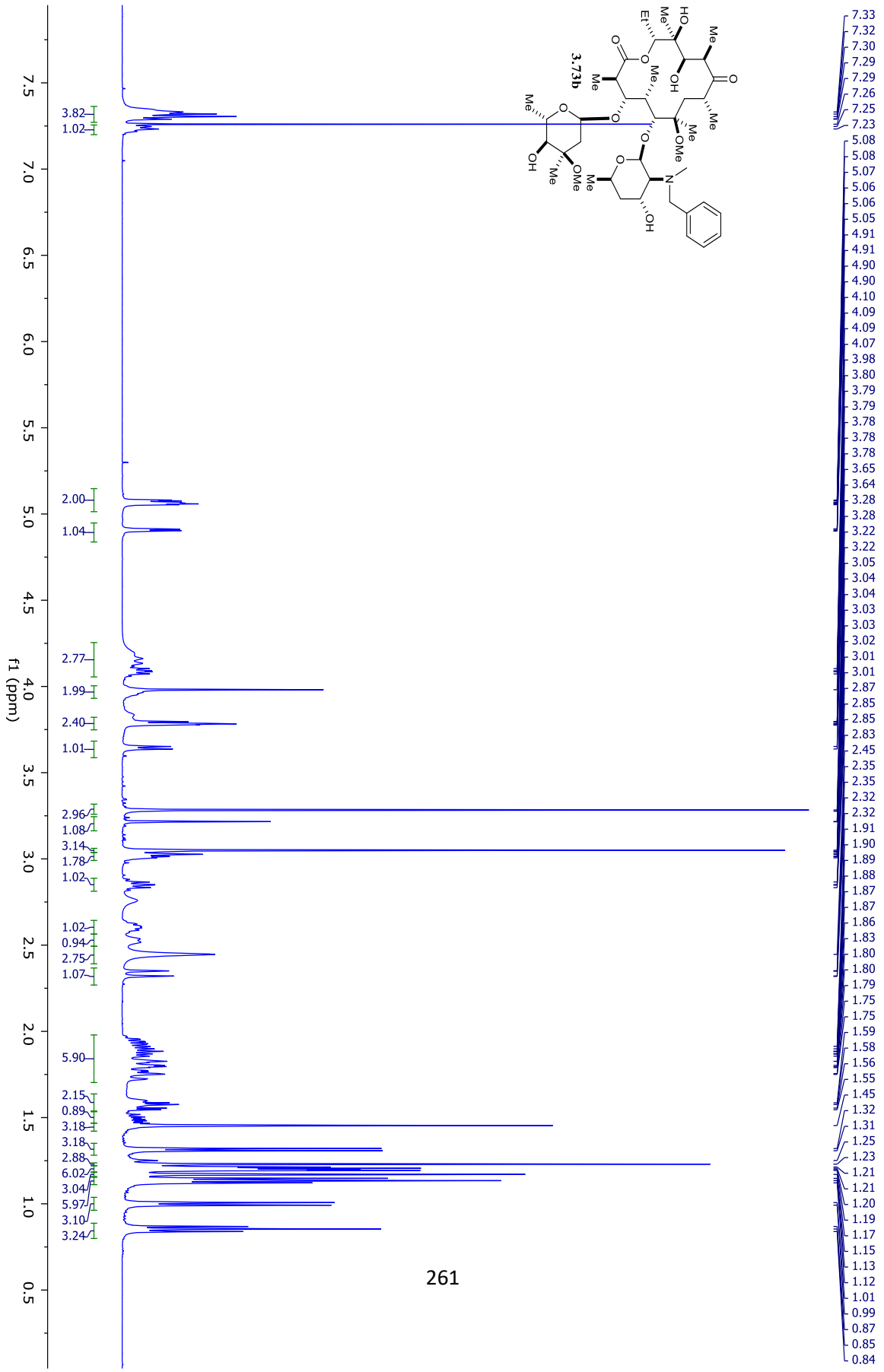

3.72a
f1 (ppm)
221.15
175.89
102.70
96.22
81.11
78.54
78.48
78.03
77.80
74.38
72.88
72.60
70.92
69.21
68.61
65.96
61.32
57.10
50.78
49.64
47.38
45.35
45.18
39.42
39.21
37.37
35.04
34.65
29.85
21.64
21.52
21.16
19.91
18.85
18.15
16.14
12.46
10.76
9.44

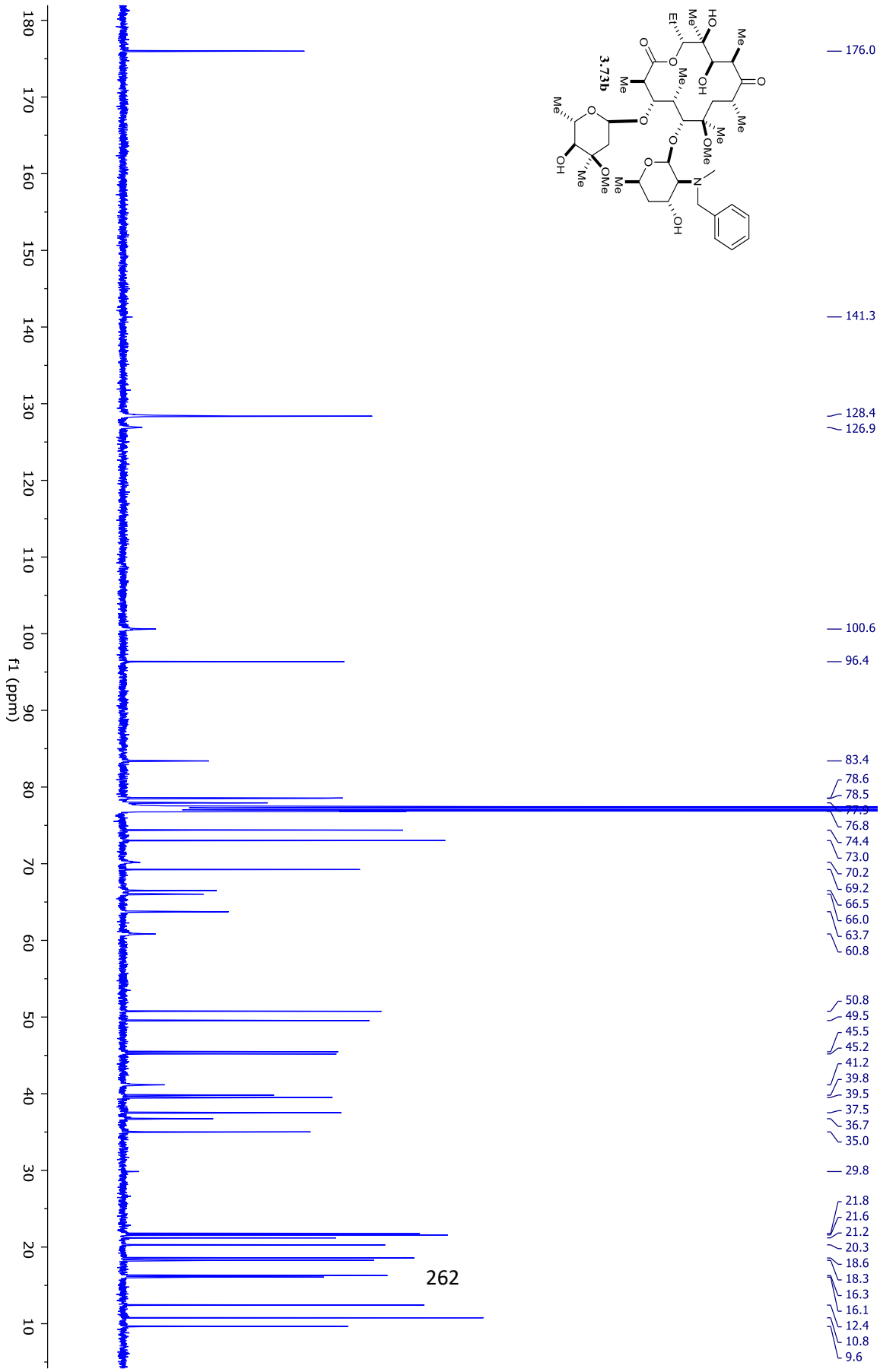

3.72b
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
1.00
0.99
0.84
0.85
1.91
1.88
1.07
0.91
3.73
0.93
0.76
6.35
2.82
1.06
0.87
1.39
4.16
1.73
1.18
3.45
3.43
14.87
9.13
3.36
4.17

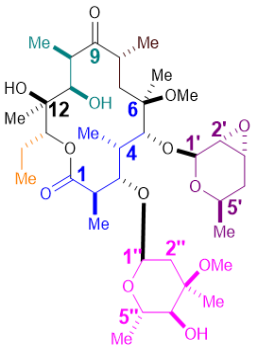

3.72b
f1 (ppm)
175.9
102.6
96.2
81.2
78.6
78.4
77.9
74.4
72.9
72.5
70.6
69.2
68.4
66.1
61.8
57.6
50.8
49.6
45.3
45.2
39.4
39.1
37.4
35.1
34.5
32.1
29.8
29.5
22.8
21.6
21.4
21.2
19.9
18.8
18.1
16.1
14.3
12.5
10.7
9.5

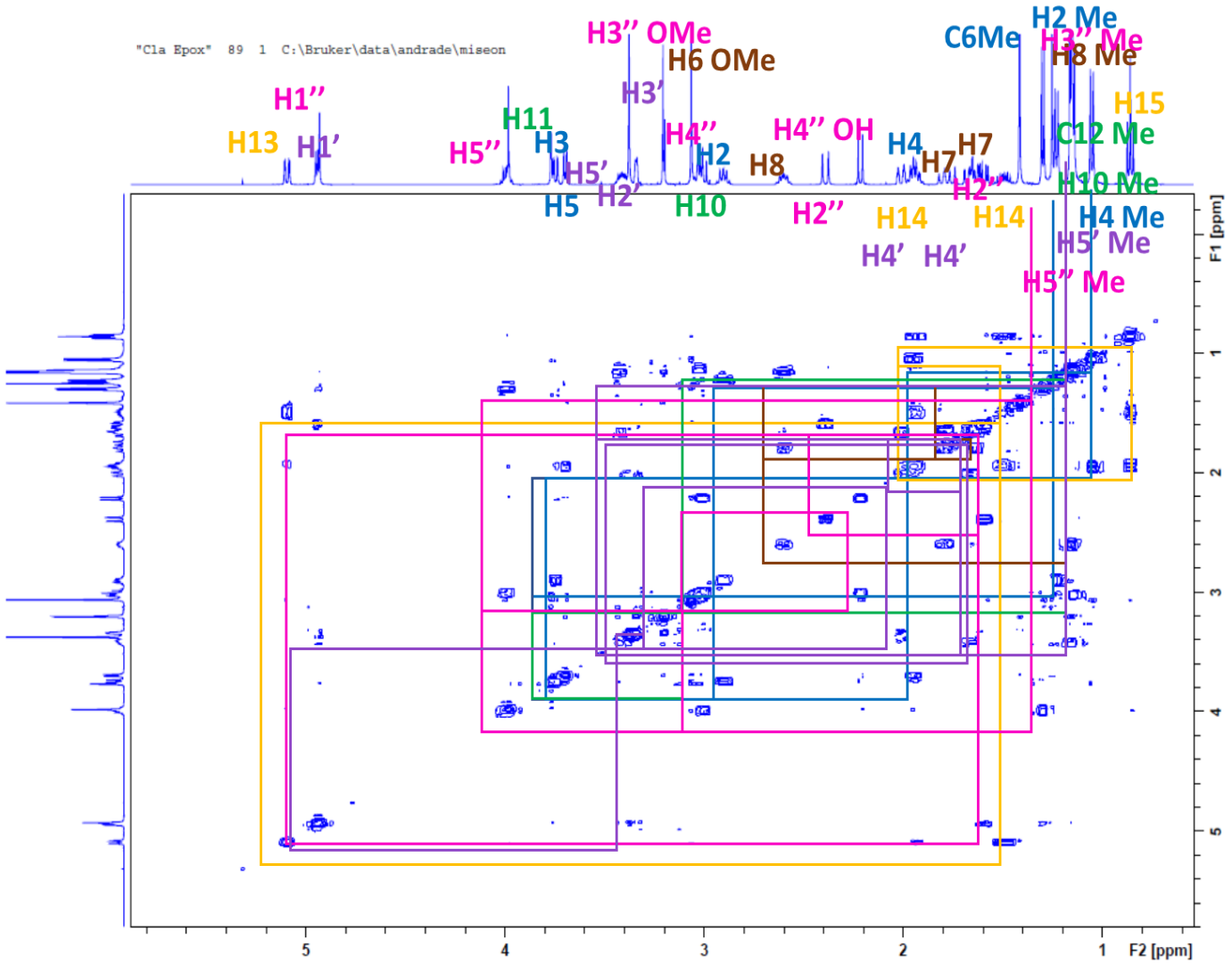

3.73a
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.04
1.01
2.03
3.05
1.08
0.97
2.12
1.00
1.08
3.08
5.32
1.01
1.98
1.03
2.85
1.00
3.15
1.99
1.14
1.13
3.20
15.13
12.93
3.62

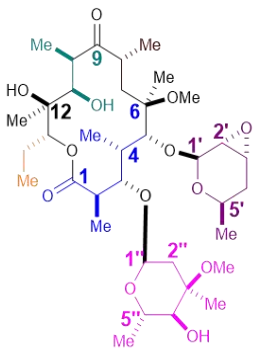

3.73a
f1 (ppm)
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
175.9
138.9
129.0
128.6
127.4
102.9
96.1
81.1
78.5
78.4
78.1
76.7
74.4
72.7
70.8
69.2
68.8
65.8
63.8
58.1
50.8
49.6
45.4
45.2
39.4
39.2
37.3
36.9
35.0
29.4
21.6
21.6
21.2
20.0
18.8
18.1
16.2
16.1
12.5
10.7
9.2

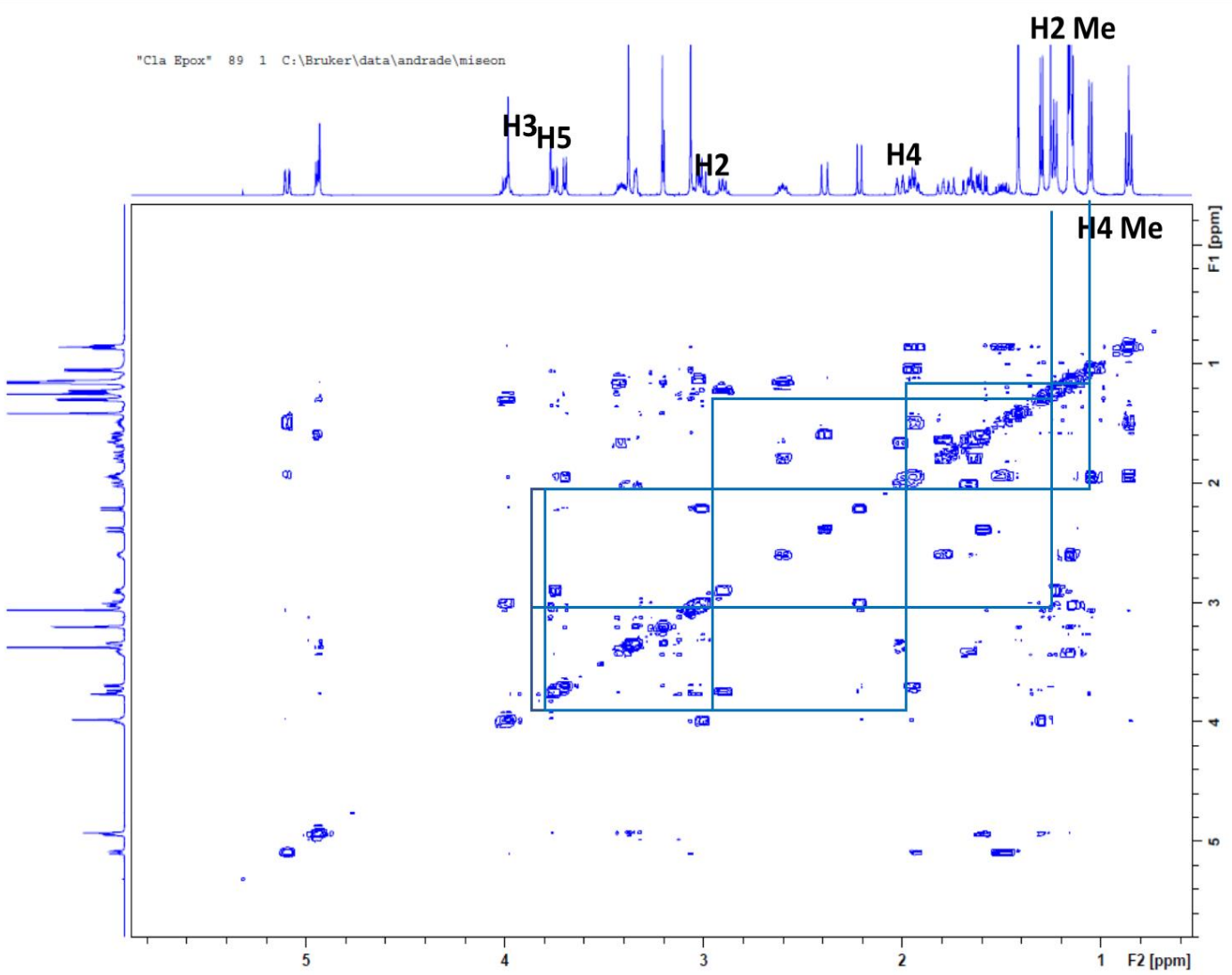

3.73b
f1 (ppm)

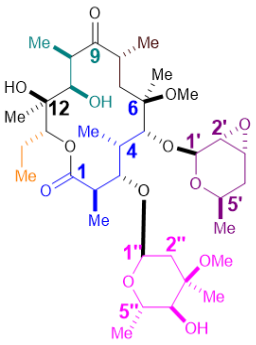

3.73b
f1 (ppm)
176.0
141.3
128.4
126.9
100.6
96.4
83.4
78.6
78.5
76.8
74.4
73.0
70.2
69.2
66.5
66.0
63.7
60.8
50.8
49.5
45.5
45.2
41.2
39.8
39.5
37.5
36.7
35.0
29.8
21.8
21.6
21.2
20.3
18.6
18.3
16.3
16.1
12.4
10.8
9.6

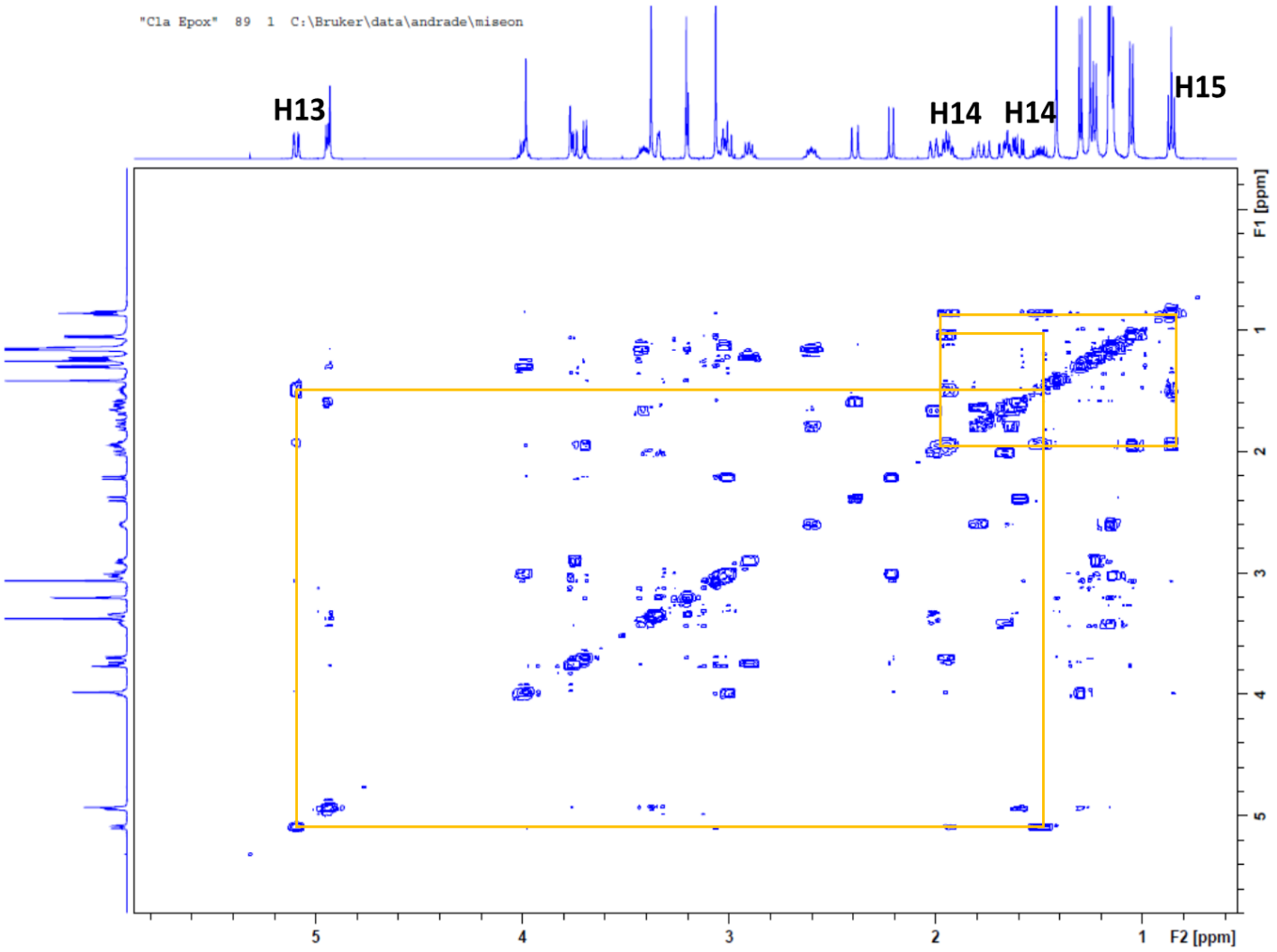

O
Me
Me
9
HO
Me
12
OH
6
Me
OMe
Me
4
1'
2'
O
O
1
O
O
5'
Me
Me
Me
1''
2''
OMe
O
Me
5''
OH
Me

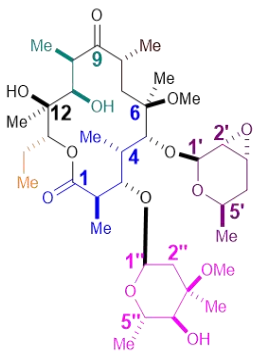

"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H3'' OMe
H6 OMe
H2 Me
C6Me
H3'' Me
H8 Me
H15
H1''
H13
H1'
H11
H5''
H3
H5'
H3'
H4''
H2
H8
H4'' OH
H4
H7
H7
C12 Me
H10 Me
H5
H2'
H10
H2''
H14
H14
H4 Me
H4'
H4'
H5' Me
H5'' Me
F1 [ppm]
F2 [ppm]
1
2
3
4
5

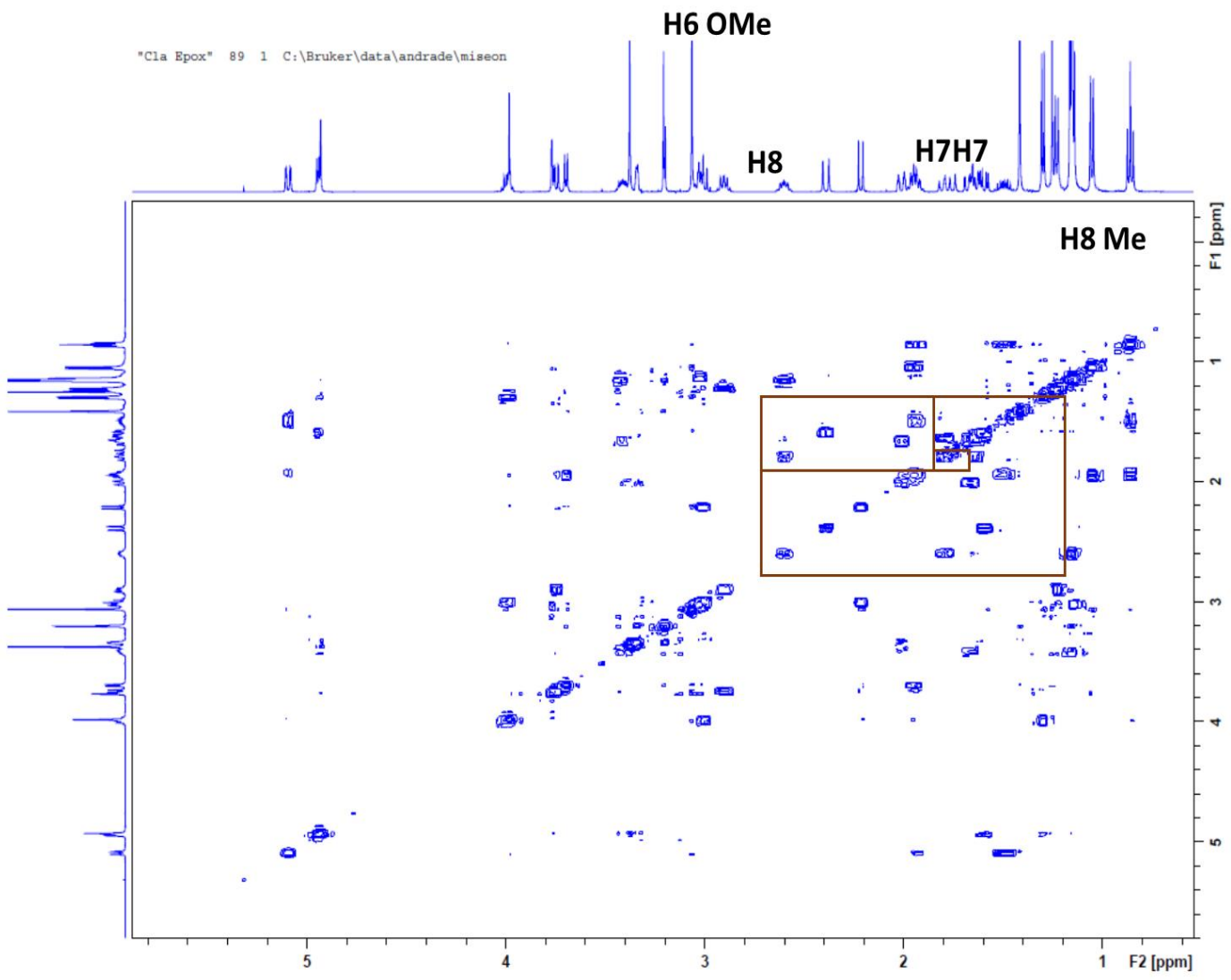

O
Me
Me
9
Me
HO
OMe
12
OH
6
Me
Me
4
1'
2'
O
O
1
O
O
5'
Me
Me
1''
2''
OMe
O
Me
5''
Me
OH

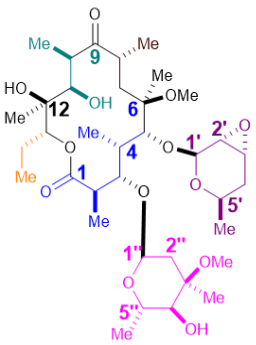

"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H2 Me
H3
H5
H2
H4
H4 Me
F1 [ppm]
F2 [ppm]
1
2
3
4
5

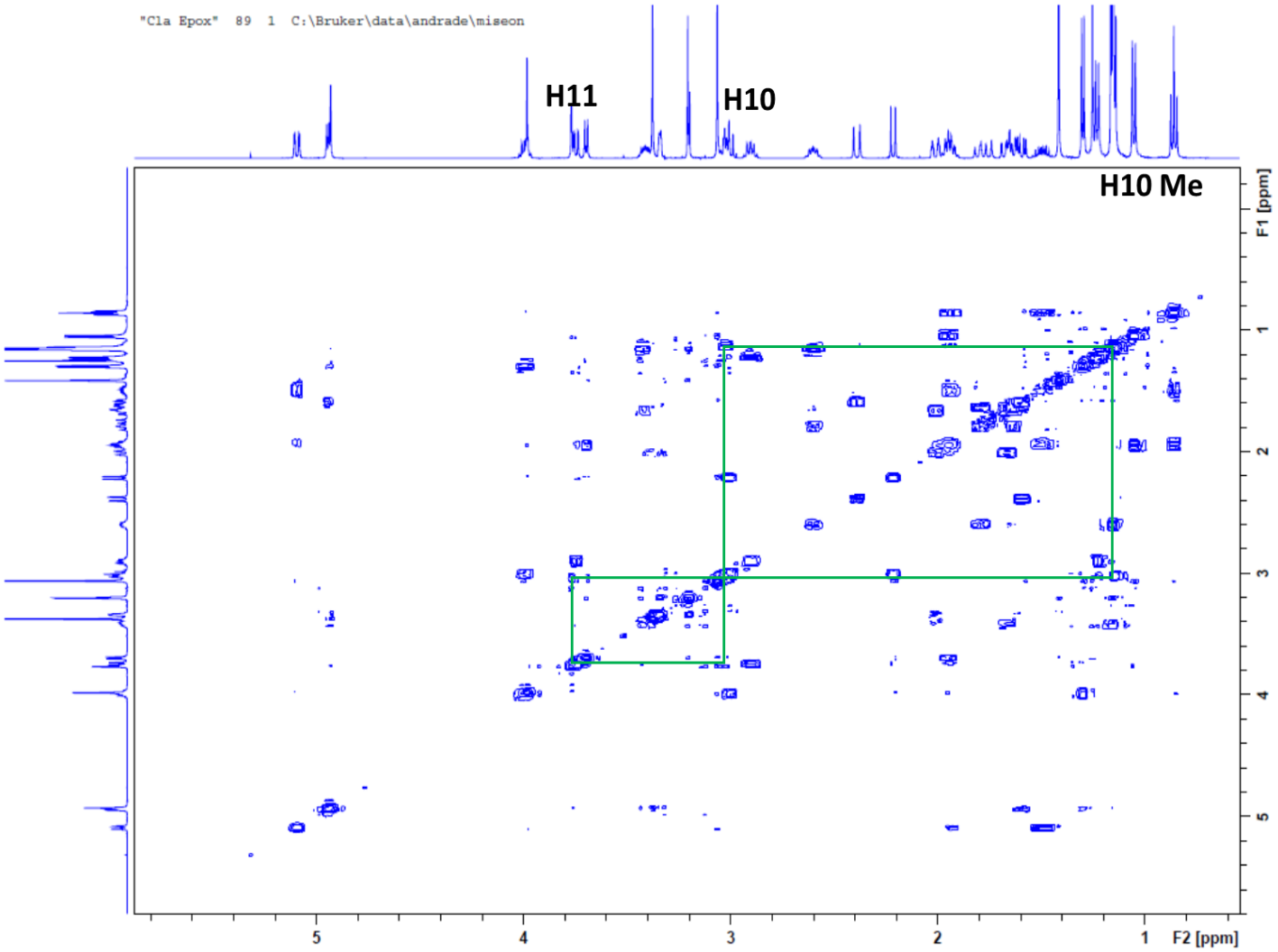

O
Me
9
Me
Me
OMe
HO
12
OH
6
Me
Me
4
2'
O
1'
O
Me
1
O
O
5'
Me
Me
1''
2''
O
OMe
Me
5''
Me
OH

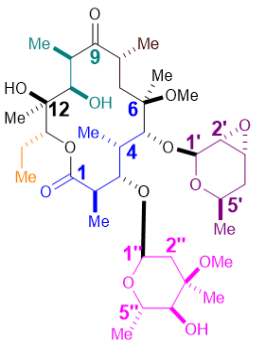

"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H13
H14
H14
H15
F1 [ppm]
F2 [ppm]
1
2
3
4
5

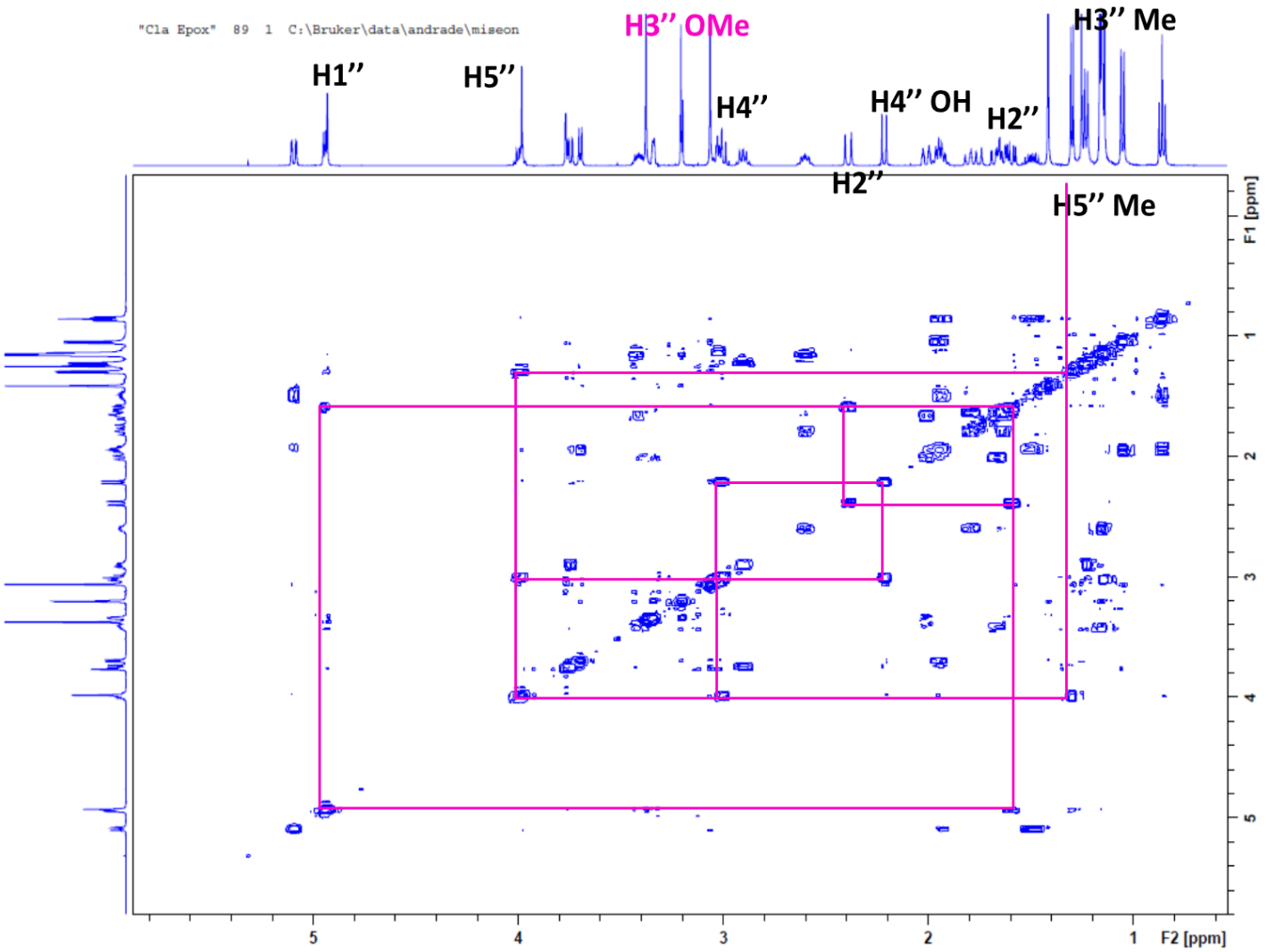

O
Me
9
Me
Me
HO
OMe
12
OH
6
Me
Me
O
4
1'
2'
O
1
O
O
Me
5'
Me
Me
1''
2''
OMe
O
Me
5''
Me
OH

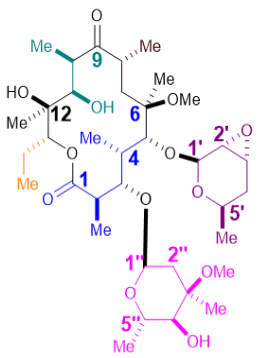

H6 OMe
"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H8
H7H7
H8 Me
F1 [ppm]
F2 [ppm]
1
2
3
4
5

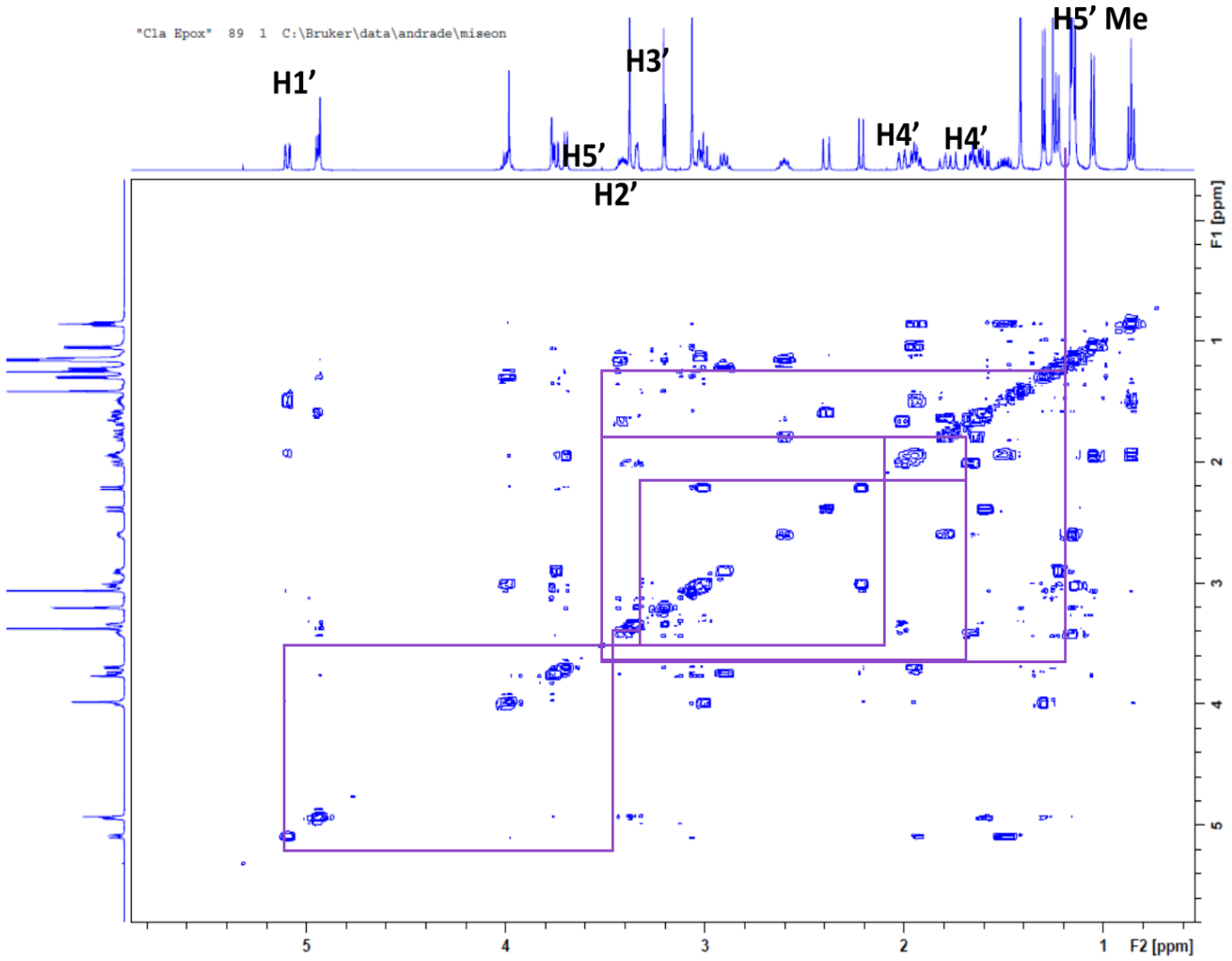

O
Me
Me
9
Me
OMe
HO
12
OH
6
Me
Me
2'
O
1'
O
4
O
1
O
Me
Me
5'
Me
1"
2"
OMe
O
Me
5"
Me
OH

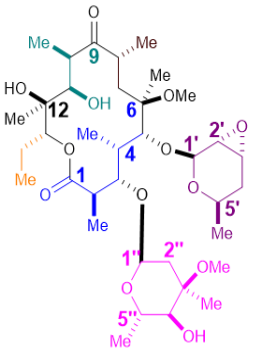

"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H11
H10
H10 Me
F1 [ppm]
F2 [ppm]
1
2
3
4
5

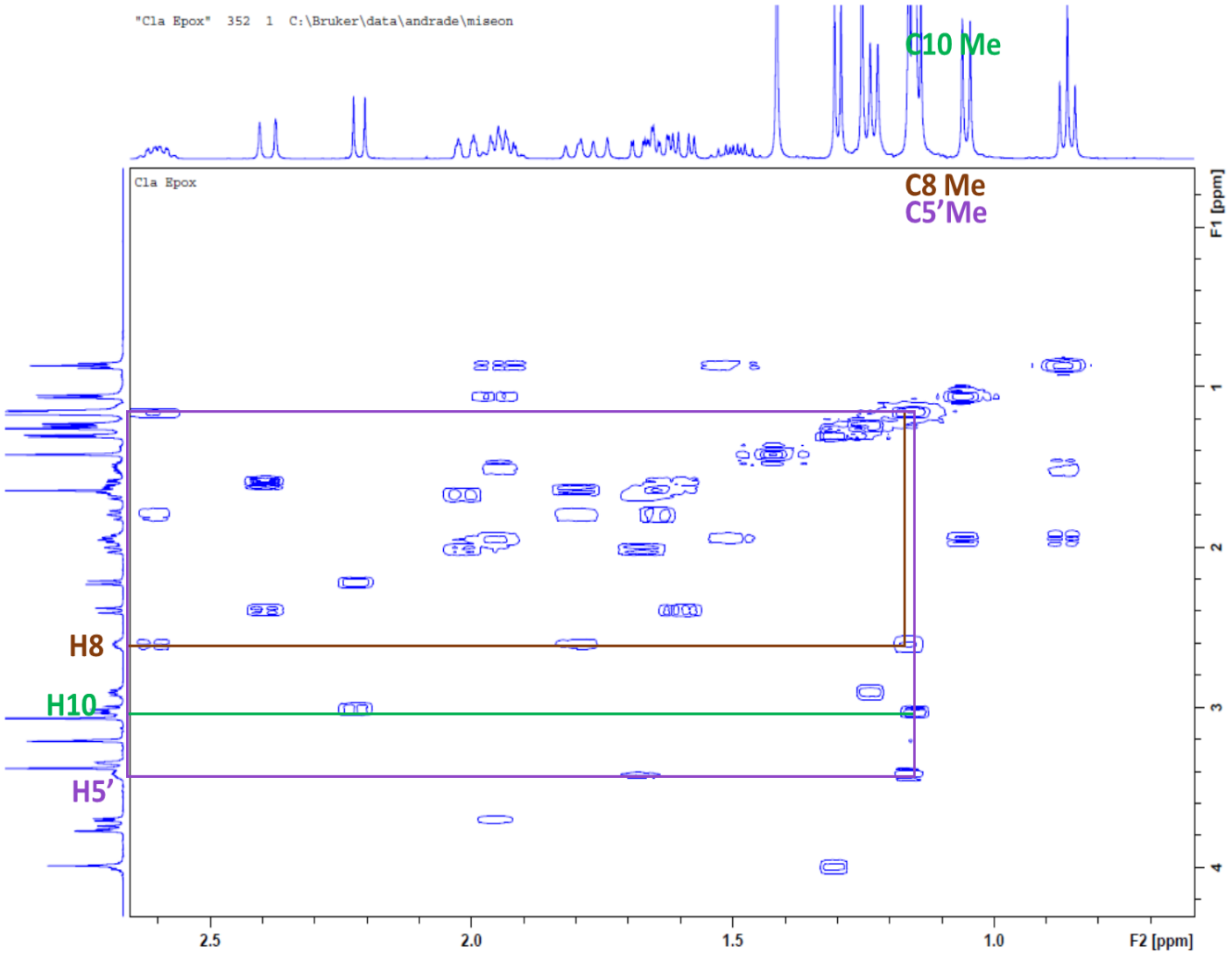

O
Me
9
Me
Me
HO
12
OH
6
OMe
Me
Me
4
1'
2'
O
Me
1
O
O
5'
Me
Me
1''
2''
OMe
O
Me
5''
Me
OH

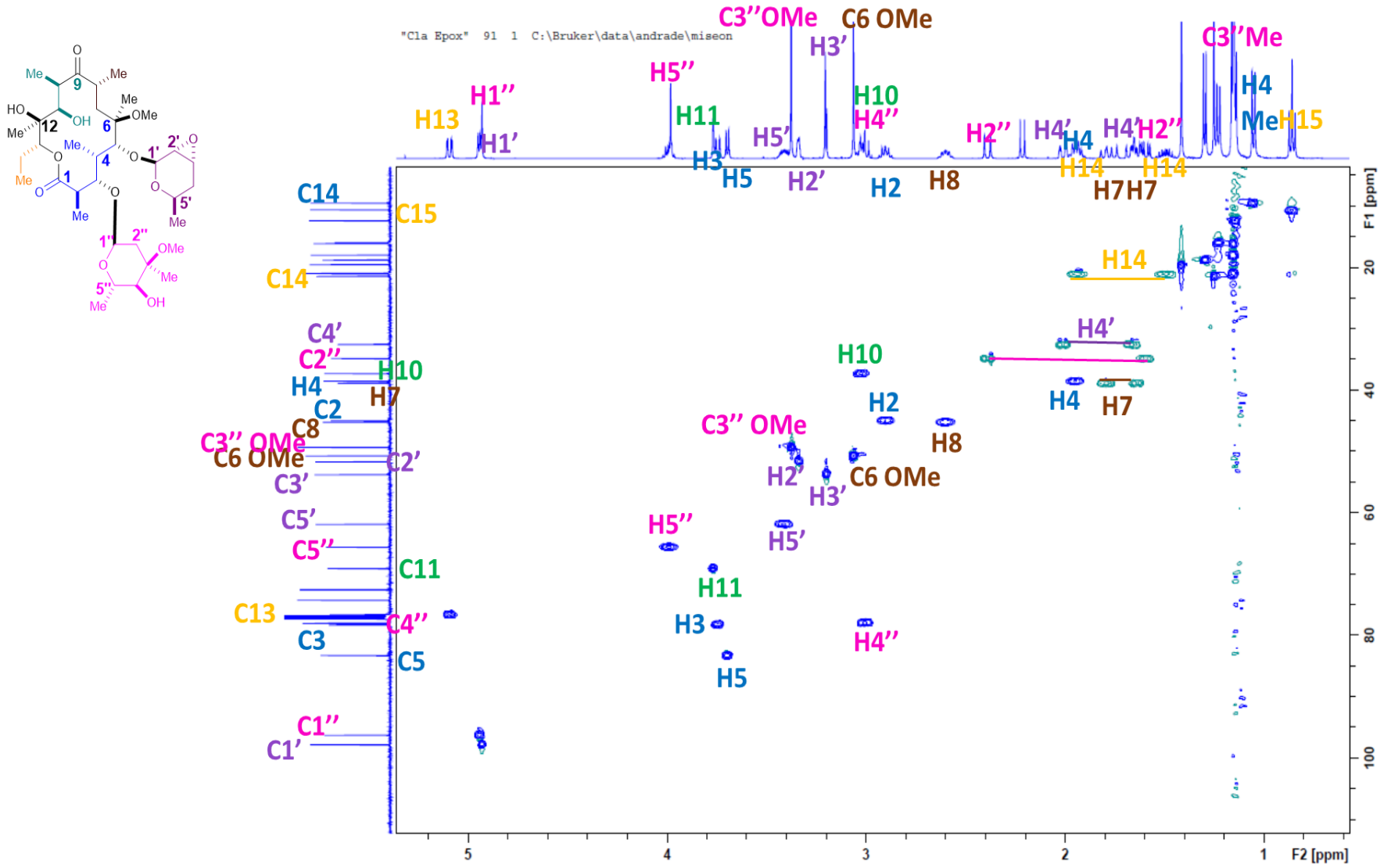

"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H1''
H5''
H3'' OMe
H4''
H4'' OH
H2''
H3'' Me
H5'' Me
F1 [ppm]
F2 [ppm]
1
2
3
4
5

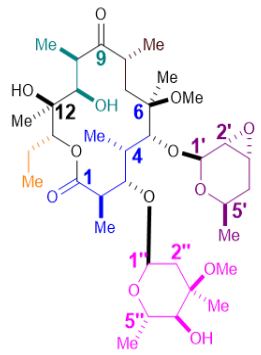

O
Me
Me
9
Me
HO
OMe
12
OH
6
Me
Me
2'
O
1'
4
O
O
1
Me
O
5'
Me
Me
1''
2''
OMe
O
Me
5''
Me
OH

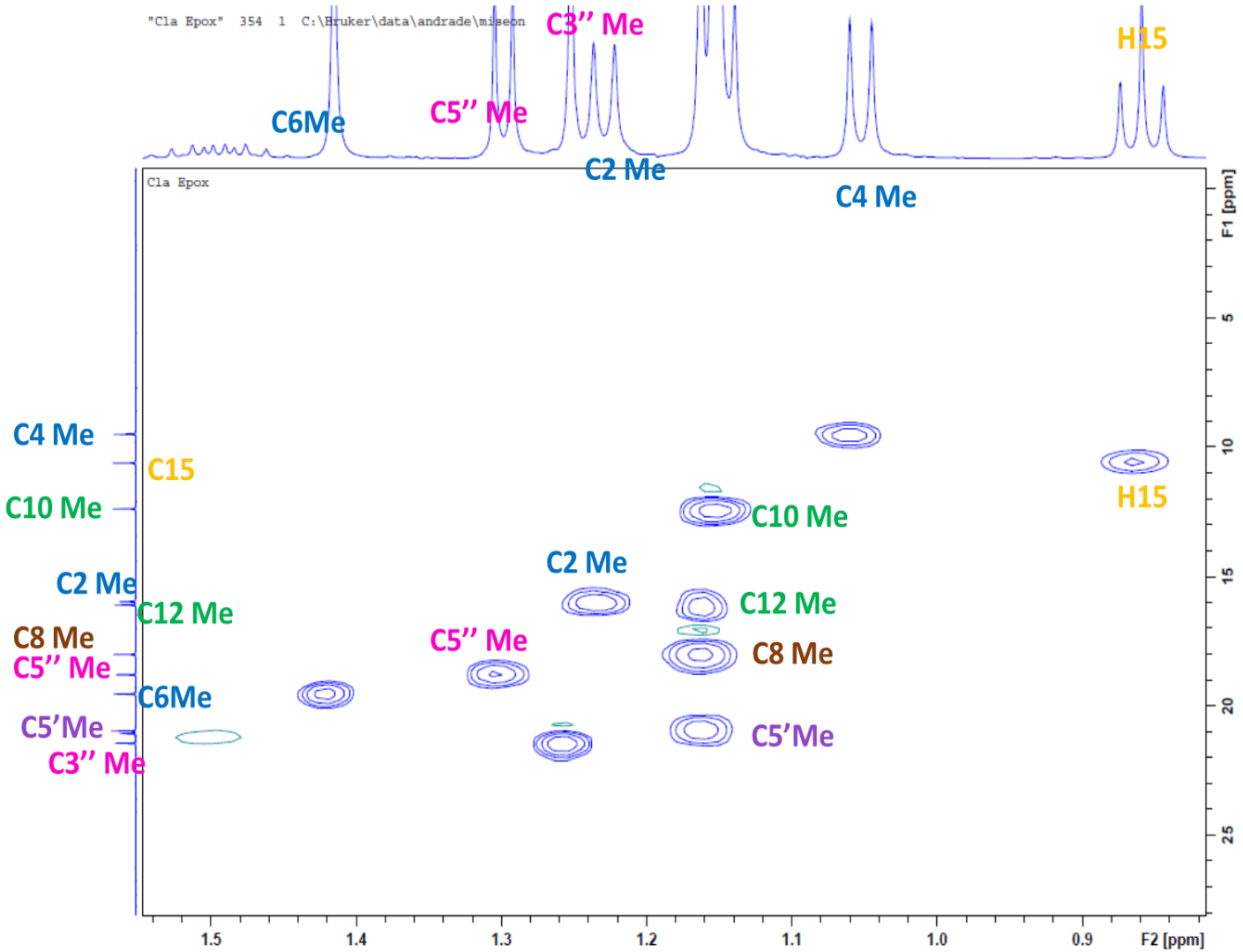

"Cla Epox" 89 1 C:\Bruker\data\andrade\miseon
H1'
H5'
H3'
H2'
H4'
H4'
H5' Me
F1 [ppm]
F2 [ppm]
1
2
3
4
5

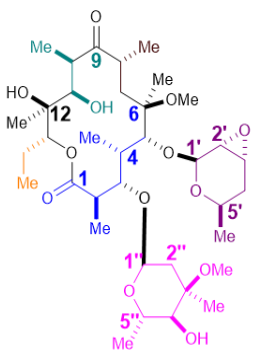

O
Me
Me
9
Me
HO
12
OH
6
OMe
Me
Me
4
1'
2'
O
Me
1
O
O
5'
Me
Me
1''
2''
OMe
O
Me
5''
Me
OH

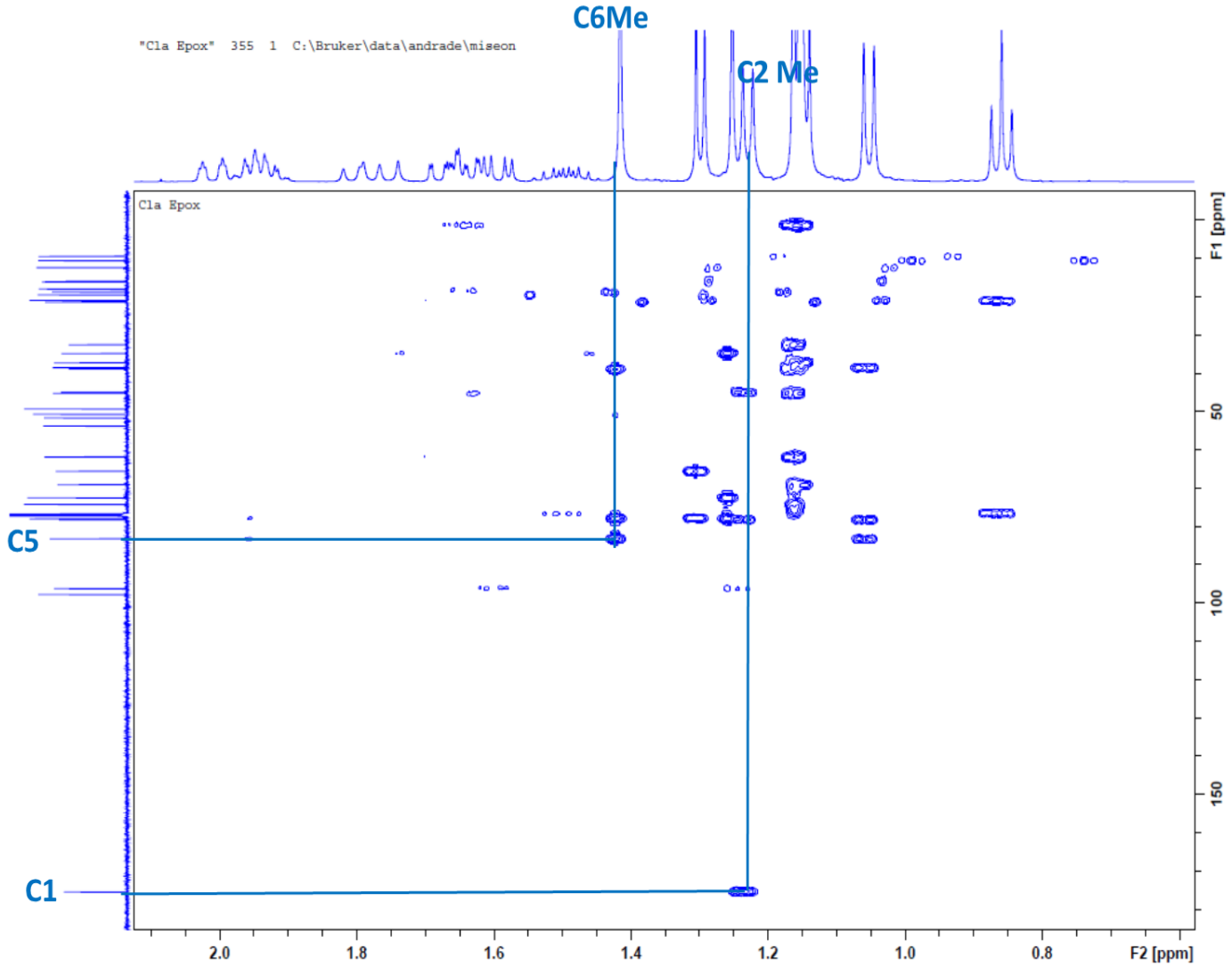

"Cla Epox" 352 1 C:\Bruker\data\andrade\miseon
C10 Me
Cla Epox
C8 Me
C5'Me
H8
H10
H5'
F1 [ppm]
F2 [ppm]
2.5
2.0
1.5
1.0
1
2
3
4

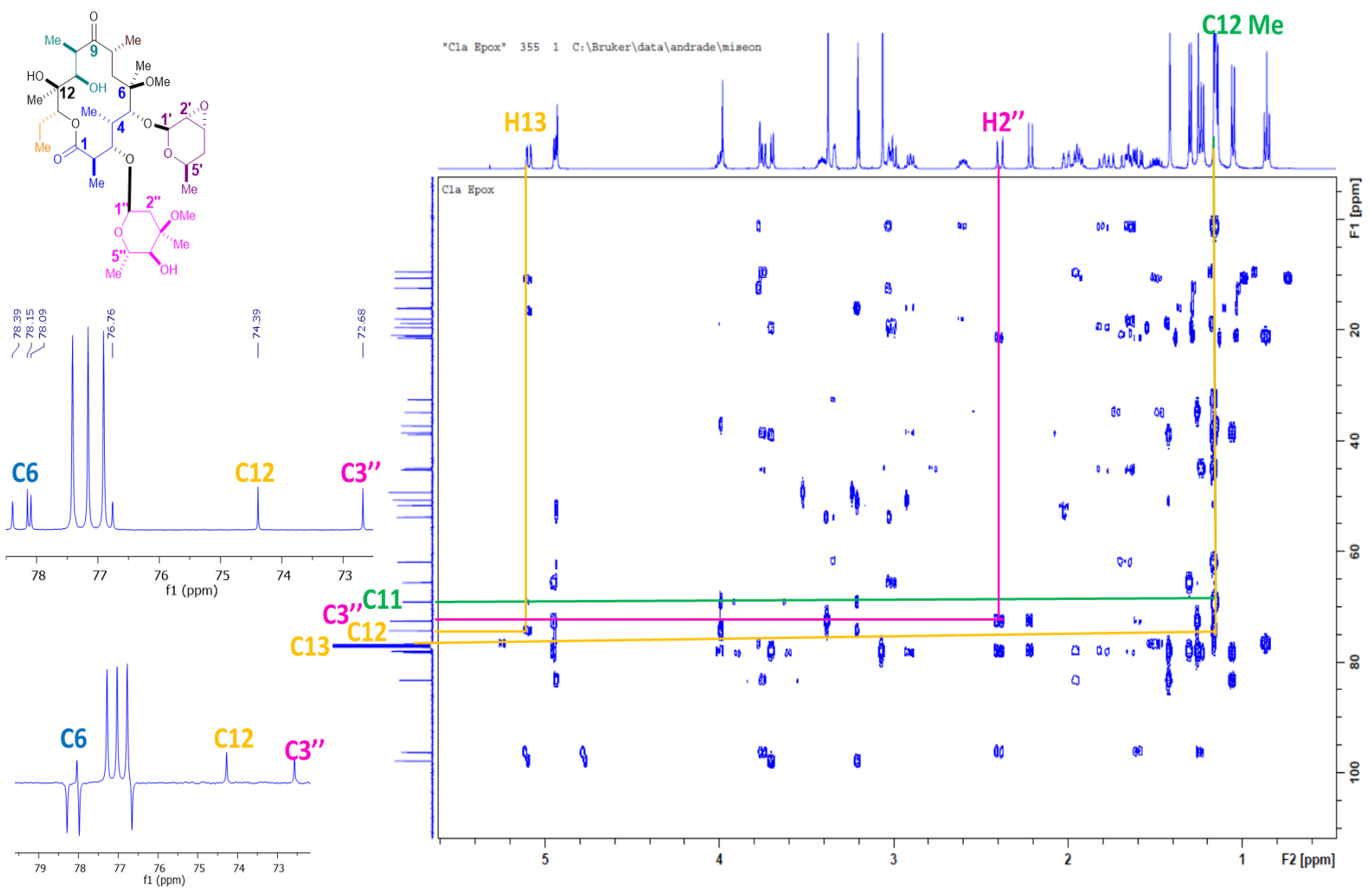

"Cla Epox" 91 1 C:\Bruker\data\andrade\miseon
C3''OMe
C6 OMe
H3'
H5''
H1''
H13
H1'
H11
H10
H4''
H5'
H2''
H4'
H4
H4'
H2''
C3'Me
H4
MeH15
H3
H5
H2'
H2
H8
H14
H14
H7H7
H14
H4'
H10
H2
H4
H7
C3'' OMe
H8
H2'
C6 OMe
H3'
H5''
H5'
H11
H3
H4''
H5
C14
C15
C14
C4'
C2''
H10
H4
H7
C2
C8
C3'' OMe
C6 OMe
C2'
C3'
C5'
C5''
C11
C13
C4''
C3
C5
C1''
C1'
F1 [ppm]
F2 [ppm]
5
4
3
2
1
20
40
60
80
100


O
Me
Me
9
Me
HO
12
OH
6
OMe
Me
Me
4
2'
O
1'
O
1
O
O
Me
Me
5'
Me
1''
2''
OMe
O
Me
5''
OH
Me


"Cla Epox" 354 1 C:\Bruker\data\andrade\miseon
C3'' Me
H15
C6Me
C5'' Me
C2 Me
C4 Me
Cla Epox
C4 Me
C15
C10 Me
C2 Me
C12 Me
C8 Me
C5'' Me
C6Me
C5'Me
C3'' Me
C10 Me
C2 Me
C12 Me
C5'' Me
C8 Me
C5'Me
H15
F1 [ppm]
5
10
15
20
25
1.5
1.4
1.3
1.2
1.1
1.0
0.9
F2 [ppm]


O
Me
Me
9
Me
HO
OMe
12
OH
6
Me
Me
O
4
O
1'
2'
O
1
O
O
Me
Me
5'
Me
1''
2''
OMe
O
Me
5''
Me
OH

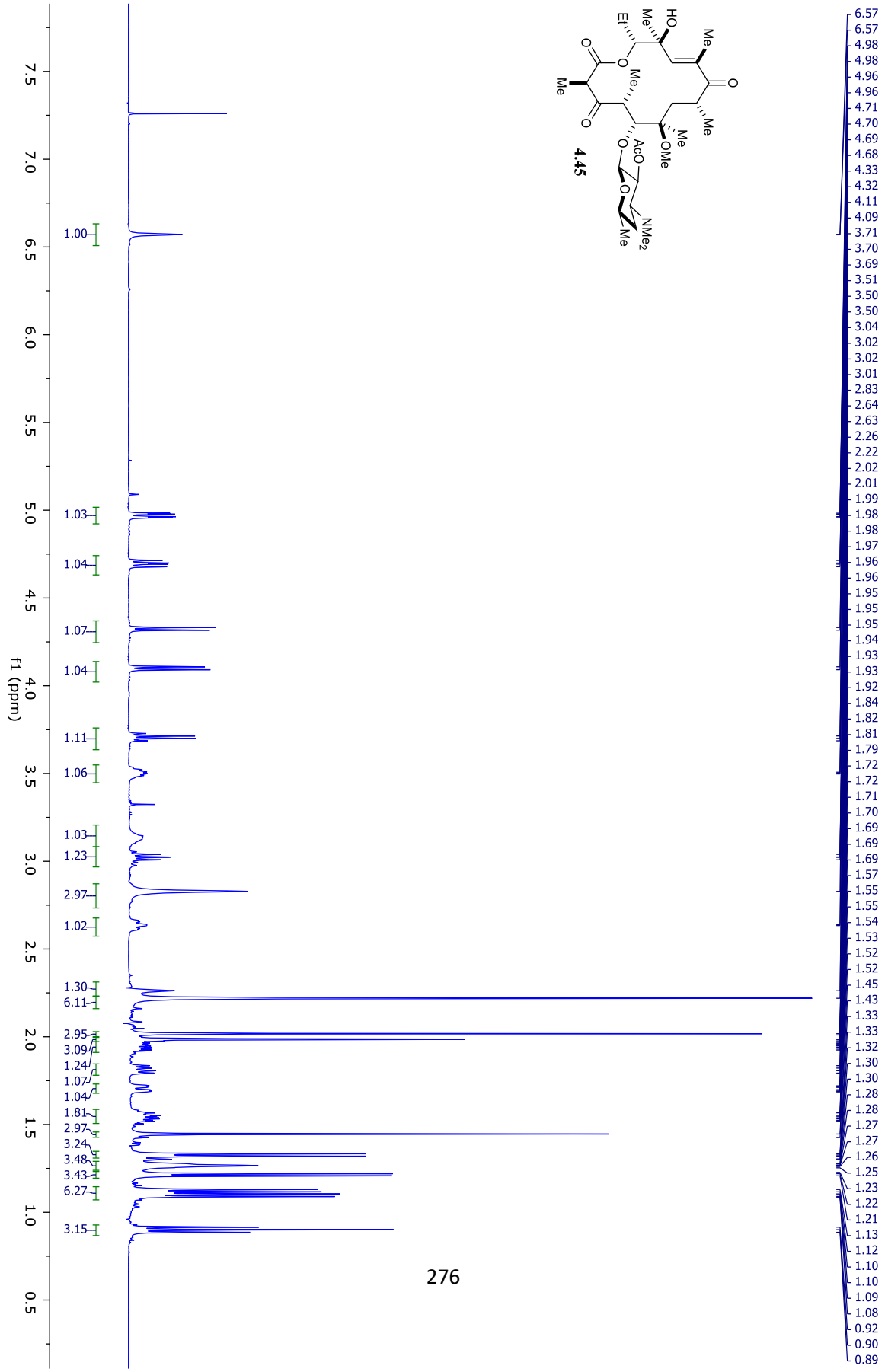

C6Me
C2 Me
"Cla Epox" 355 1 C:\Bruker\data\andrade\miseon
Cla Epox
C5
C1
F1 [ppm]
50
100
150
2.0
1.8
1.6
1.4
1.2
1.0
0.8
F2 [ppm]

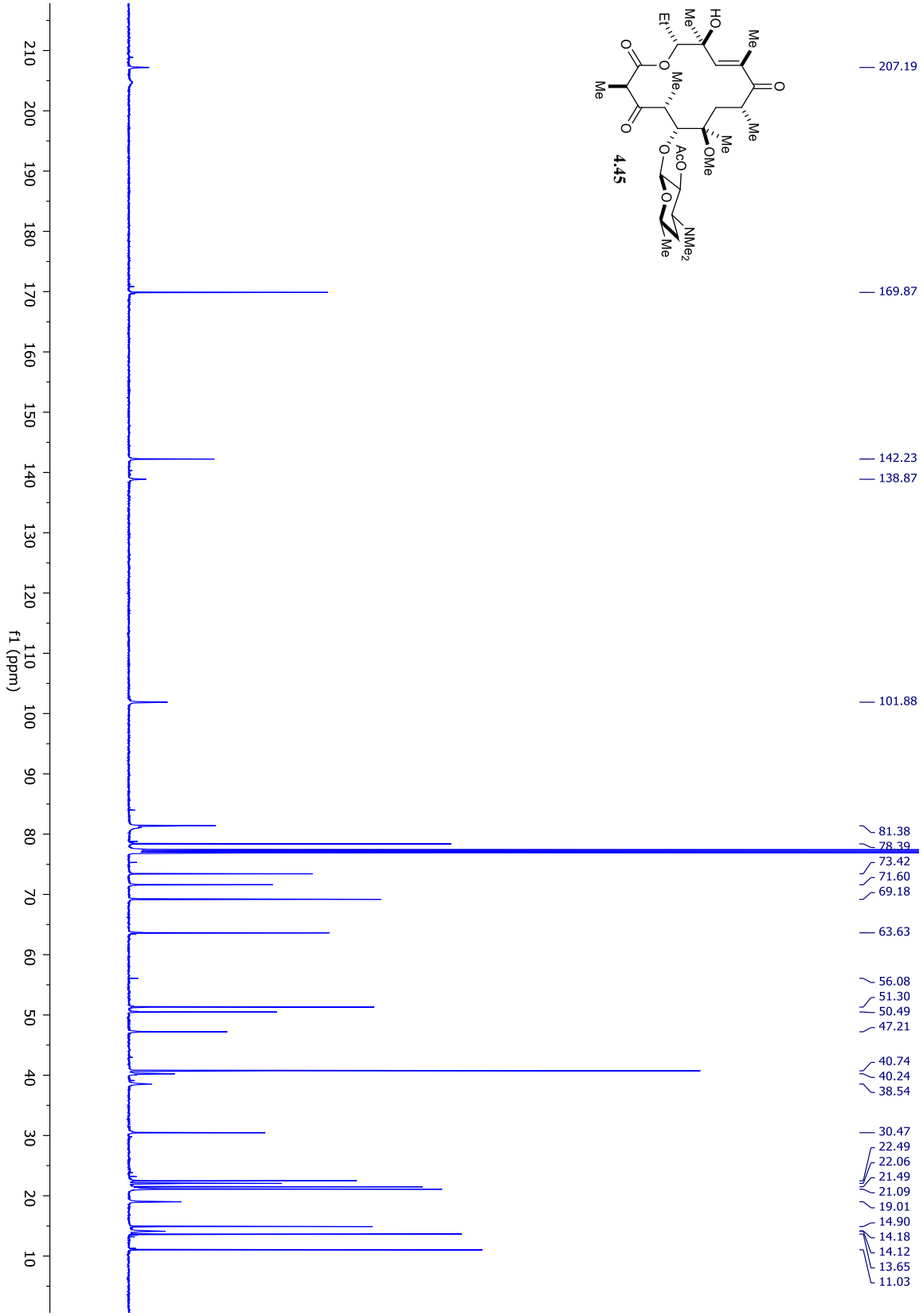

C12 Me
"Cla Epox" 355 1 C:\Bruker\data\andrade\miseon
H13
H2''
Cla Epox
F1 [ppm]
F2 [ppm]
C11
C3''
C12
C13
78.39
78.15
78.09
76.76
74.39
72.68
C6
C12
C3''
f1 (ppm)
C6
C12
C3''
f1 (ppm)

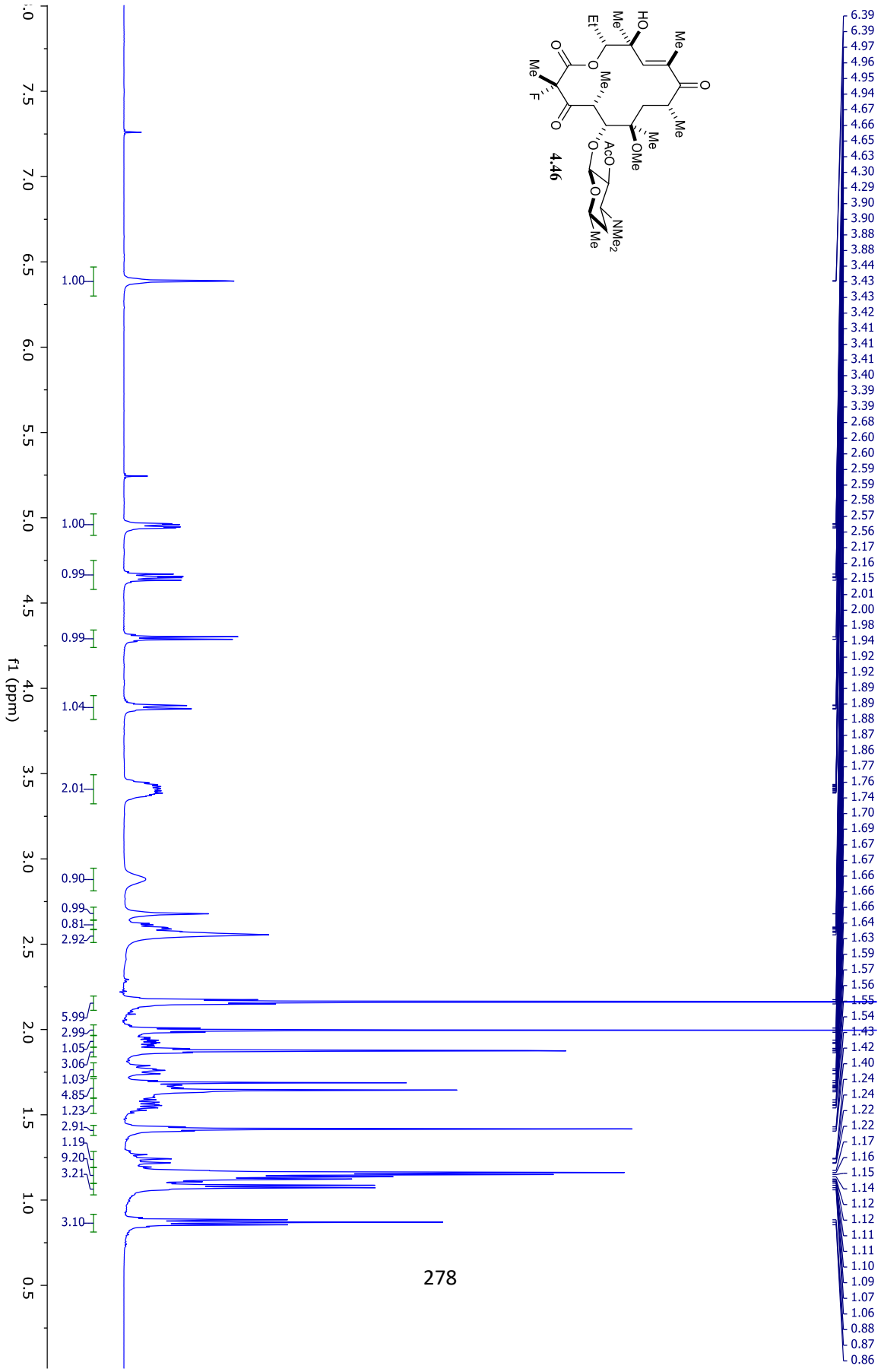
APPENDIX A3

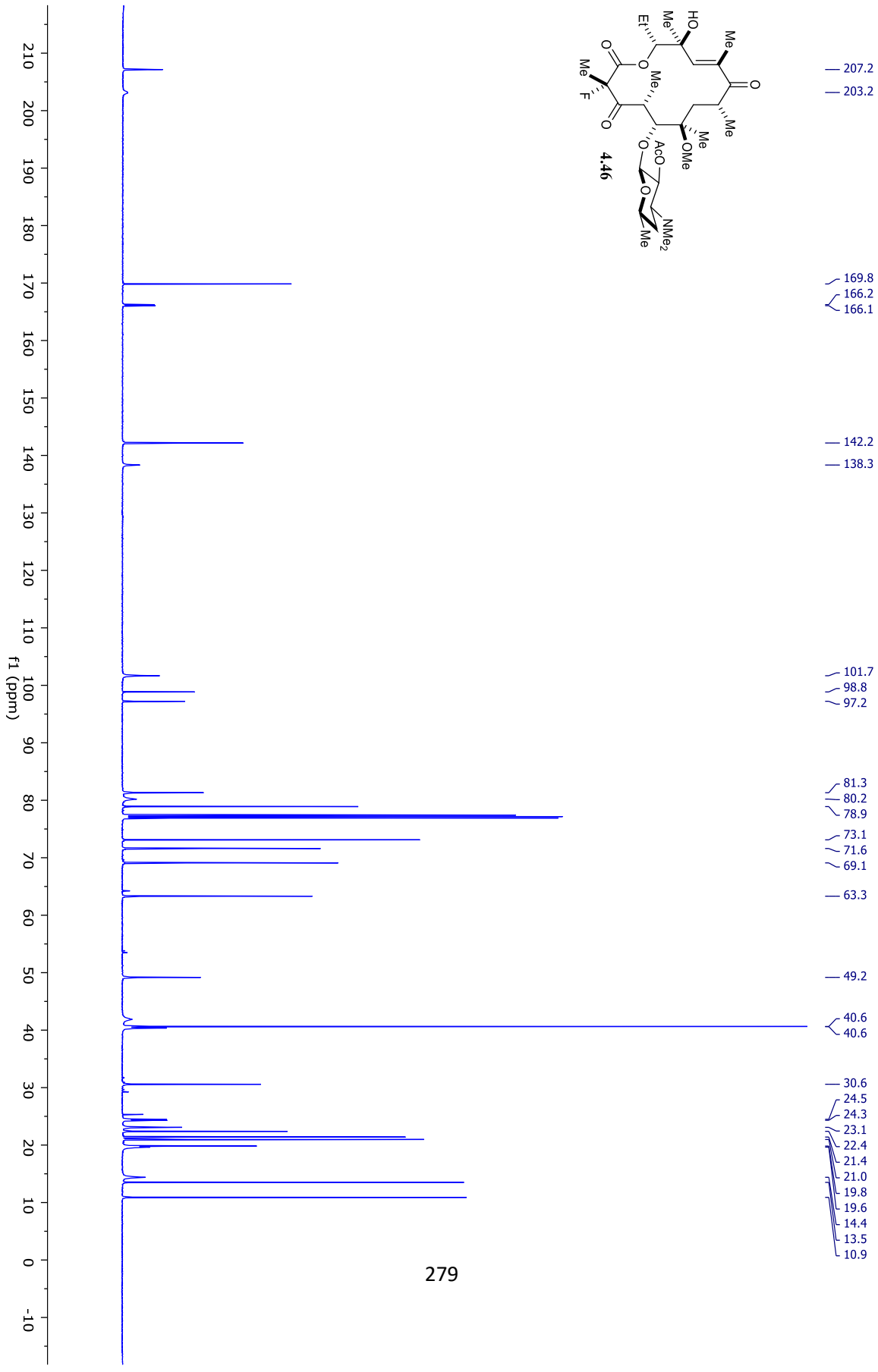
NMR SPECTRA OF COMPOUNDS IN

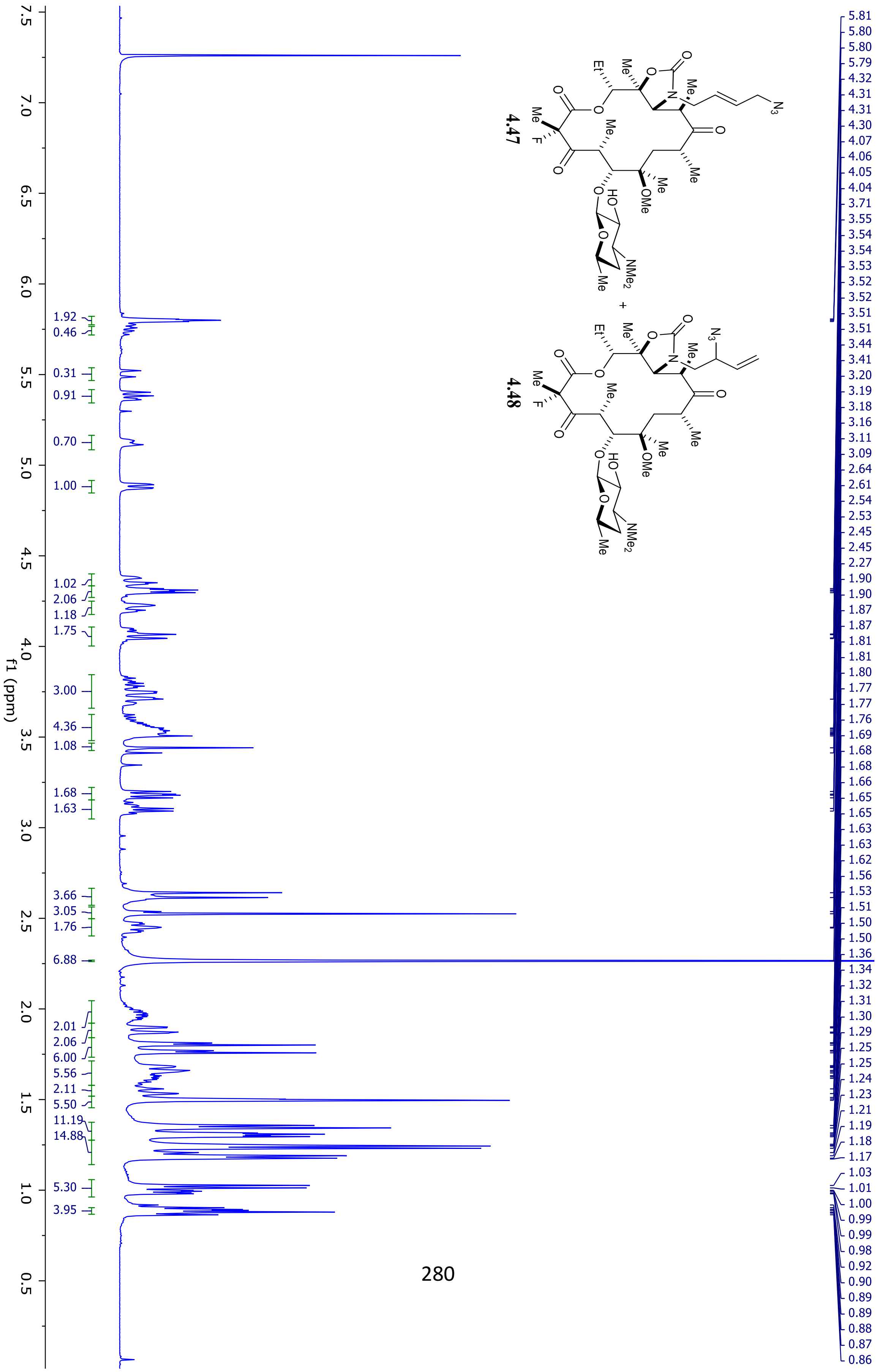
CHAPTER 4

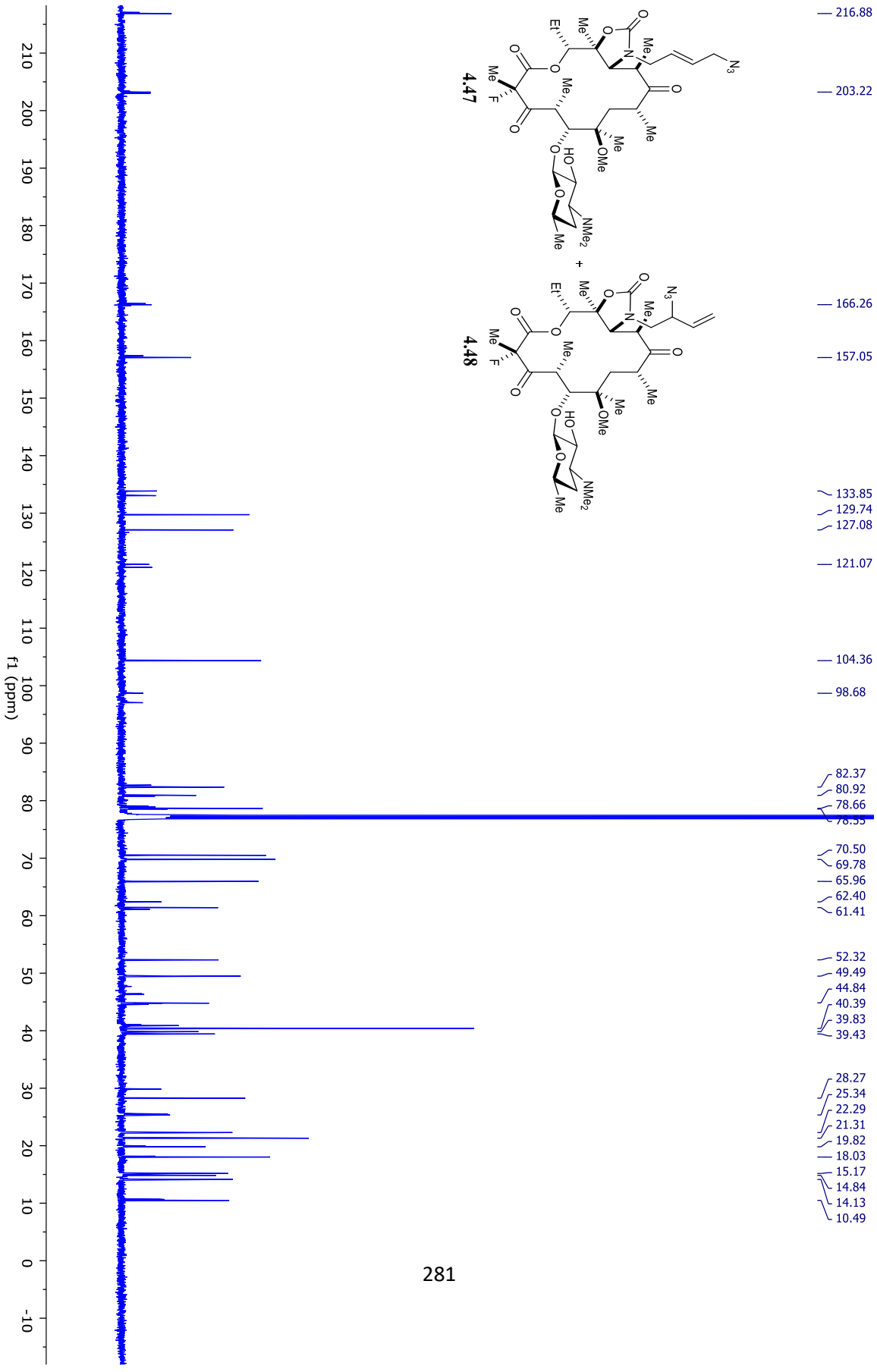

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.03
1.04
1.07
1.04
1.11
1.06
1.03
1.23
2.97
1.02
1.30
6.11
2.95
3.09
1.24
1.07
1.04
1.81
2.97
3.24
3.48
3.43
6.27
3.15
6.57
6.57
4.98
4.98
4.96
4.96
4.71
4.70
4.69
4.68
4.33
4.32
4.11
4.09
3.71
3.70
3.69
3.51
3.50
3.50
3.04
3.02
3.02
3.01
2.83
2.64
2.63
2.26
2.22
2.02
2.01
1.99
1.98
1.98
1.97
1.96
1.96
1.95
1.95
1.95
1.94
1.93
1.93
1.92
1.84
1.82
1.81
1.79
1.72
1.72
1.71
1.70
1.69
1.69
1.69
1.57
1.55
1.55
1.54
1.53
1.52
1.52
1.45
1.43
1.33
1.33
1.32
1.30
1.30
1.28
1.28
1.27
1.27
1.26
1.25
1.23
1.22
1.21
1.13
1.12
1.10
1.10
1.09
1.08
0.92
0.90
0.89
4.45
HO
Me
Et
Me
O
Me
Me
Me
OMe
Me
AcO
O
O
NMe2
Me

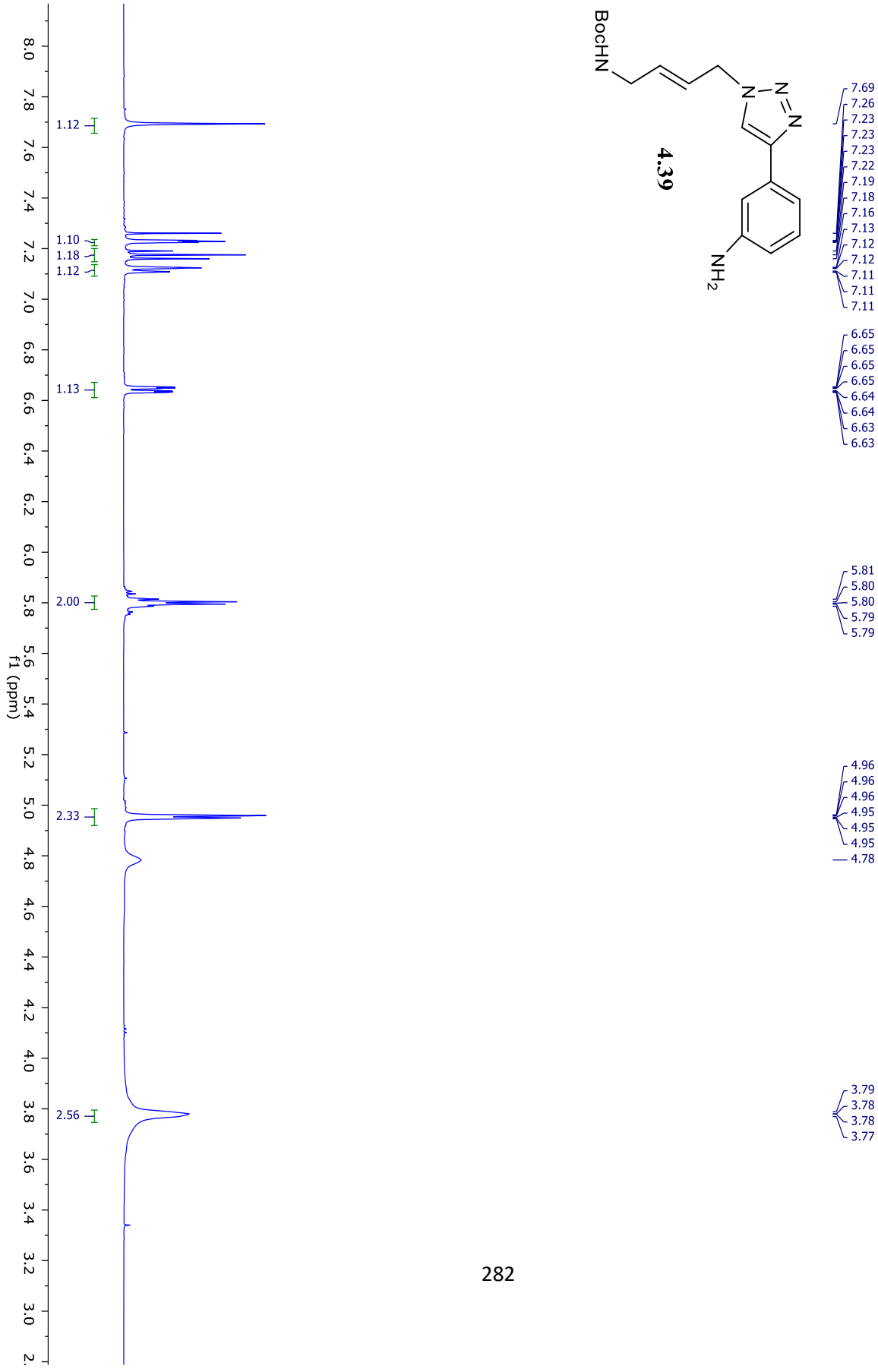

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
207.19
169.87
142.23
138.87
101.88
81.38
78.39
73.42
71.60
69.18
63.63
56.08
51.30
50.49
47.21
40.74
40.24
38.54
30.47
22.49
22.06
21.49
21.09
19.01
14.90
14.18
14.12
13.65
11.03
4.45

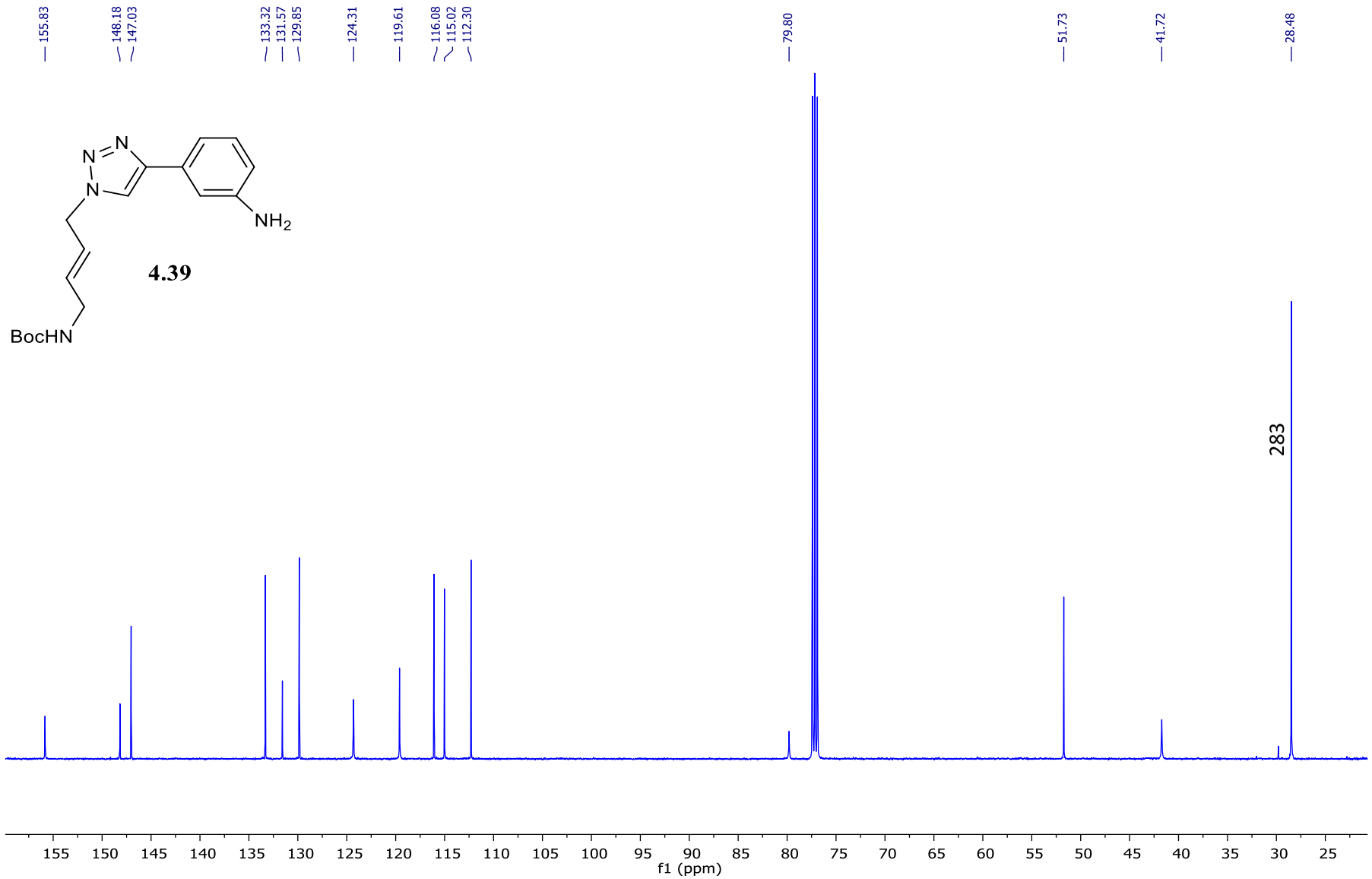

f1 (ppm)
1.00
1.00
0.99
0.99
1.04
2.01
0.90
0.99
0.81
2.92
5.99
2.99
1.05
3.06
1.03
4.85
1.23
2.91
1.19
9.20
3.21
3.10
4.46

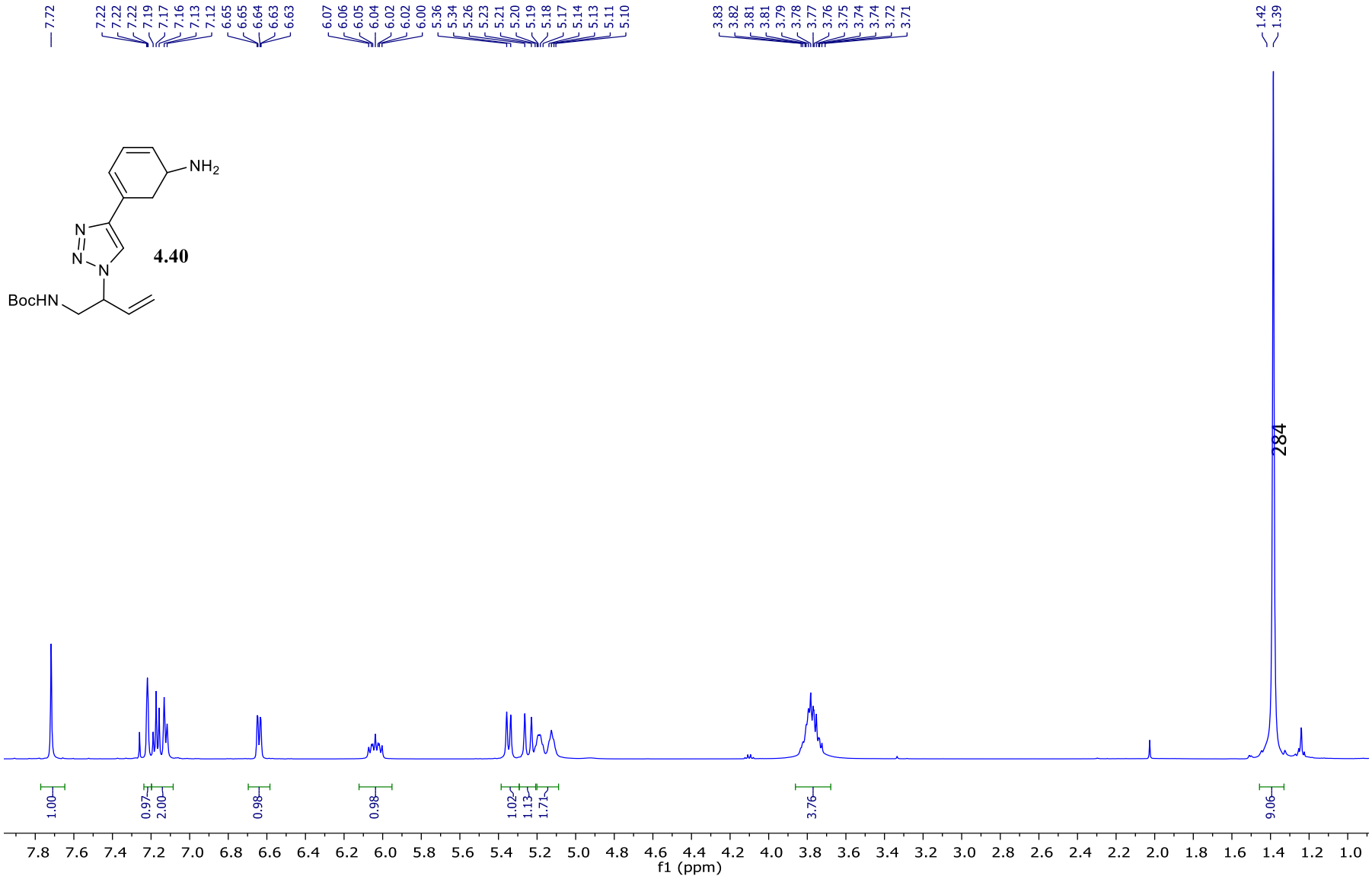

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
207.2
203.2
169.8
166.2
166.1
142.2
138.3
101.7
98.8
97.2
81.3
80.2
78.9
73.1
71.6
69.1
63.3
49.2
40.6
40.6
30.6
24.5
24.3
23.1
22.4
21.4
21.0
19.8
19.6
14.4
13.5
10.9
4.46

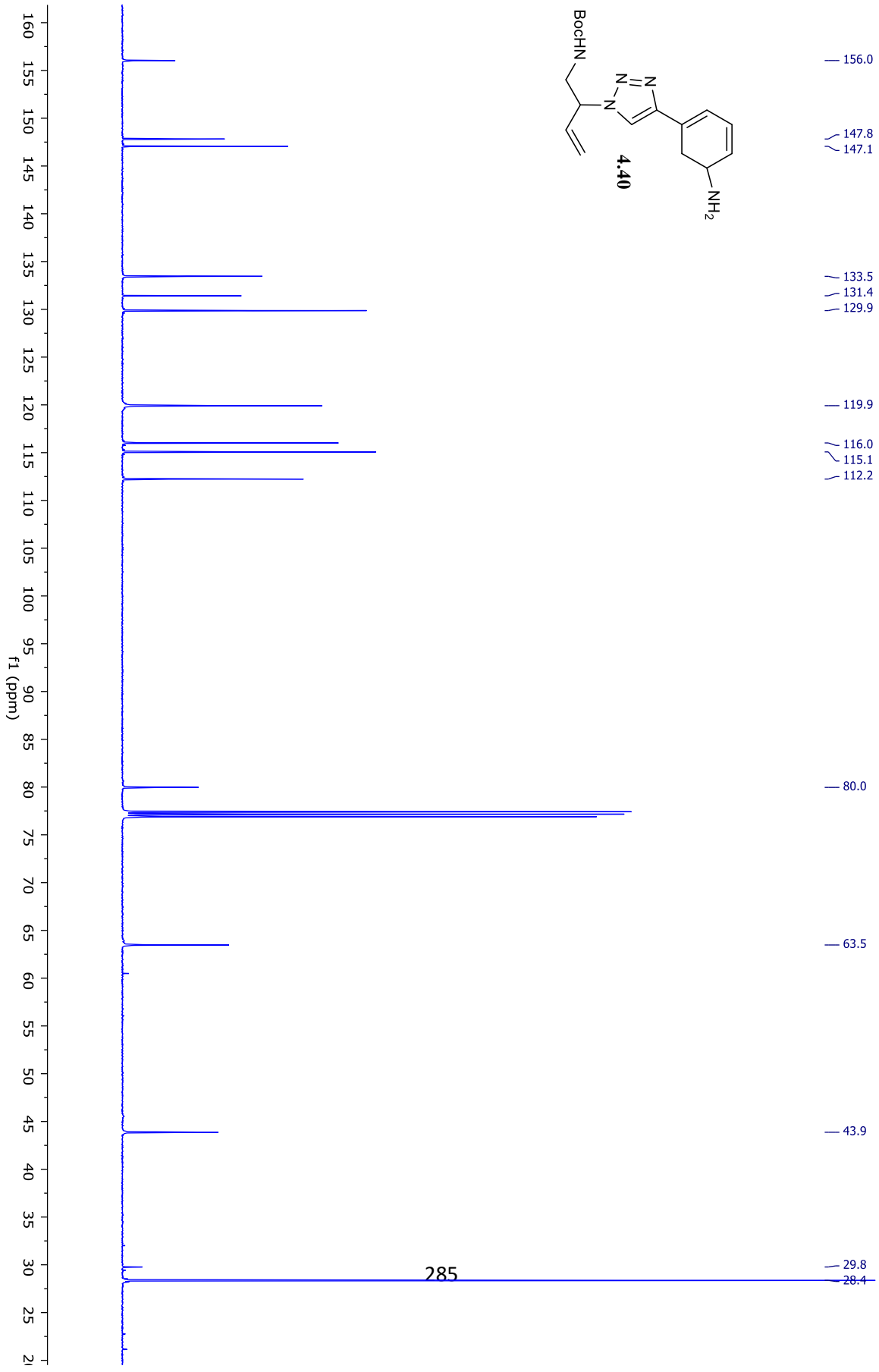

f1 (ppm)
4.47
4.48

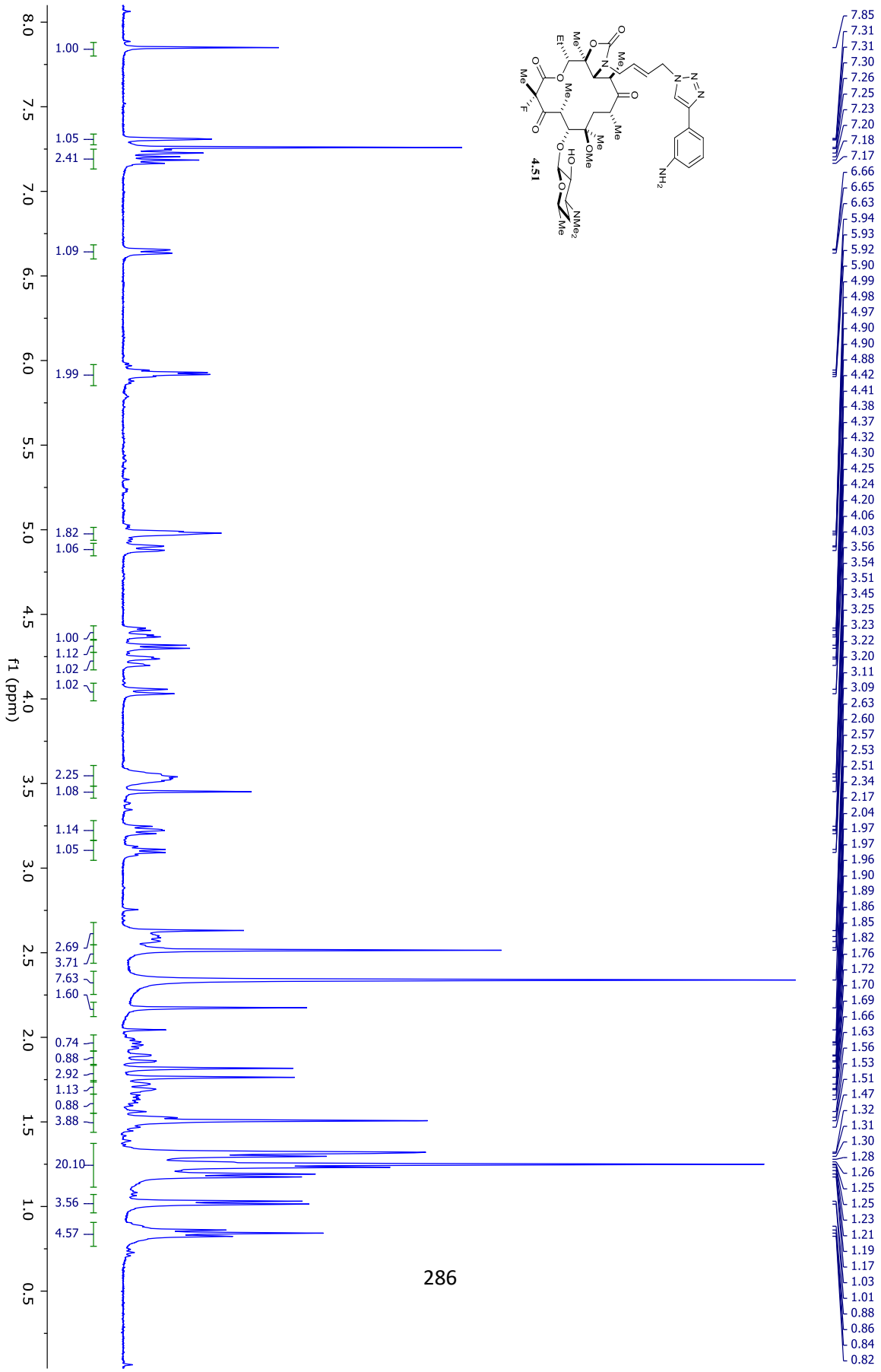

4.47
4.48
216.88
203.22
166.26
157.05
133.85
129.74
127.08
121.07
104.36
98.68
82.37
80.92
78.66
78.55
70.50
69.78
65.96
62.40
61.41
52.32
49.49
44.84
40.39
39.83
39.43
28.27
25.34
22.29
21.31
19.82
18.03
15.17
14.84
14.13
10.49
f1 (ppm)

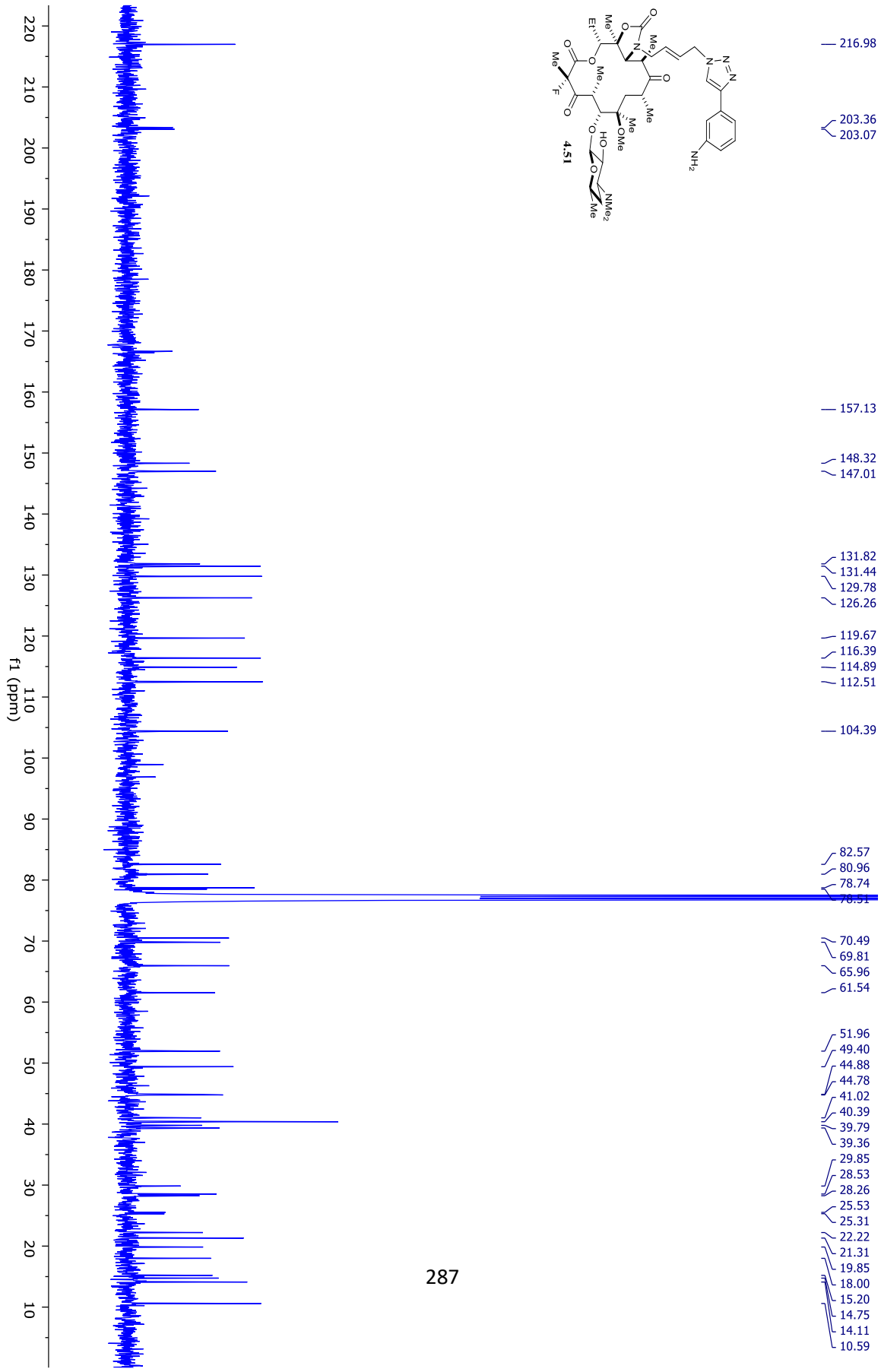

BocHN
N
N
N
NH2
4.39
7.69
7.26
7.23
7.23
7.23
7.22
7.19
7.18
7.16
7.13
7.12
7.12
7.11
7.11
7.11
6.65
6.65
6.65
6.65
6.64
6.64
6.63
6.63
5.81
5.80
5.80
5.79
5.79
4.96
4.96
4.96
4.95
4.95
4.95
4.78
3.79
3.78
3.78
3.77
1.12
1.10
1.18
1.12
1.13
2.00
2.33
2.56
f1 (ppm)

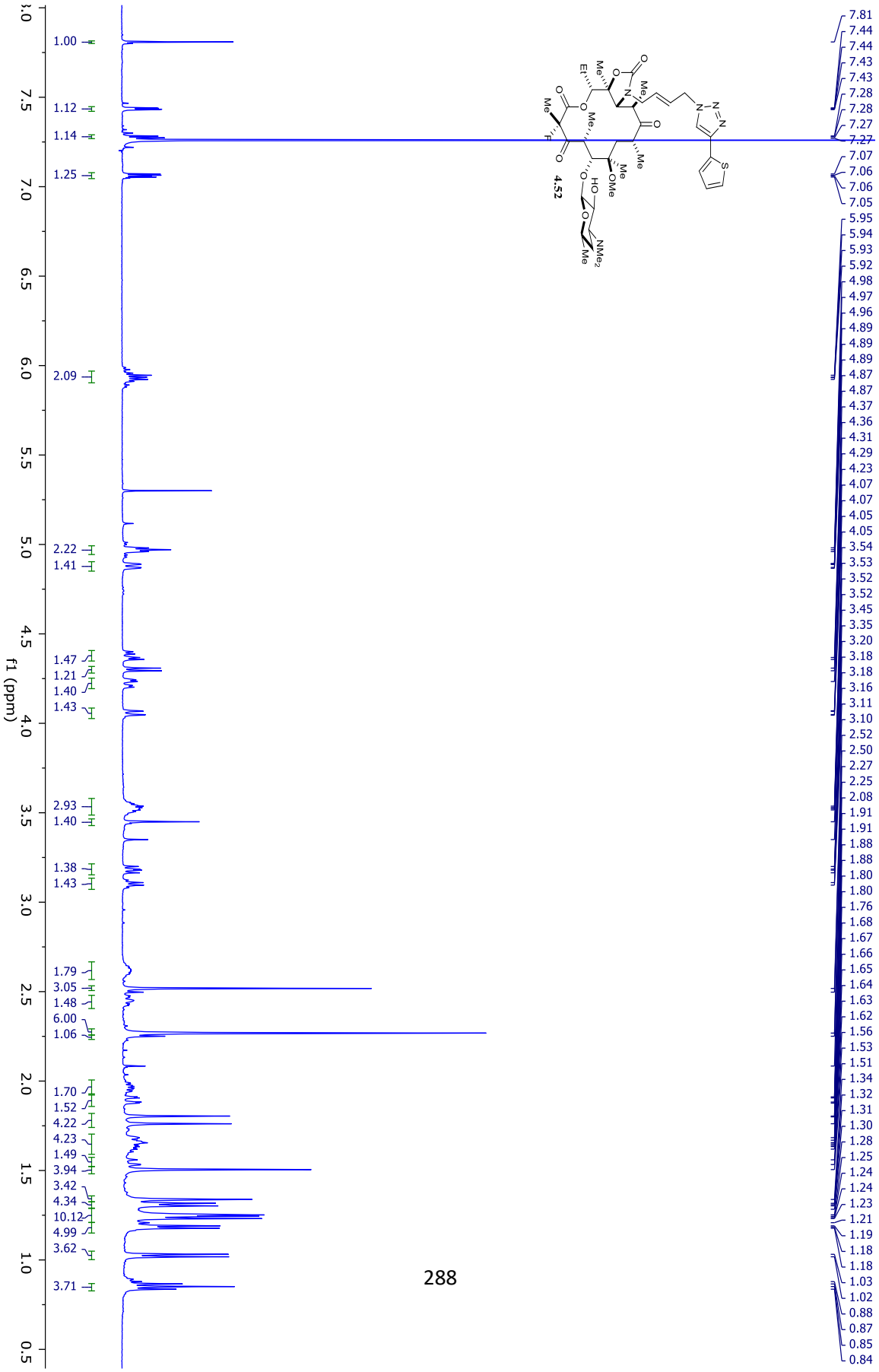

155.83
148.18
147.03
133.32
131.57
129.85
124.31
119.61
116.08
115.02
112.30
79.80
51.73
41.72
28.48
283
N
N
N
NH2
BocHN
4.39
155
150
145
140
135
130
125
120
115
110
105
100
95
90
85
80
75
70
65
60
55
50
45
40
35
30
25
f1 (ppm)

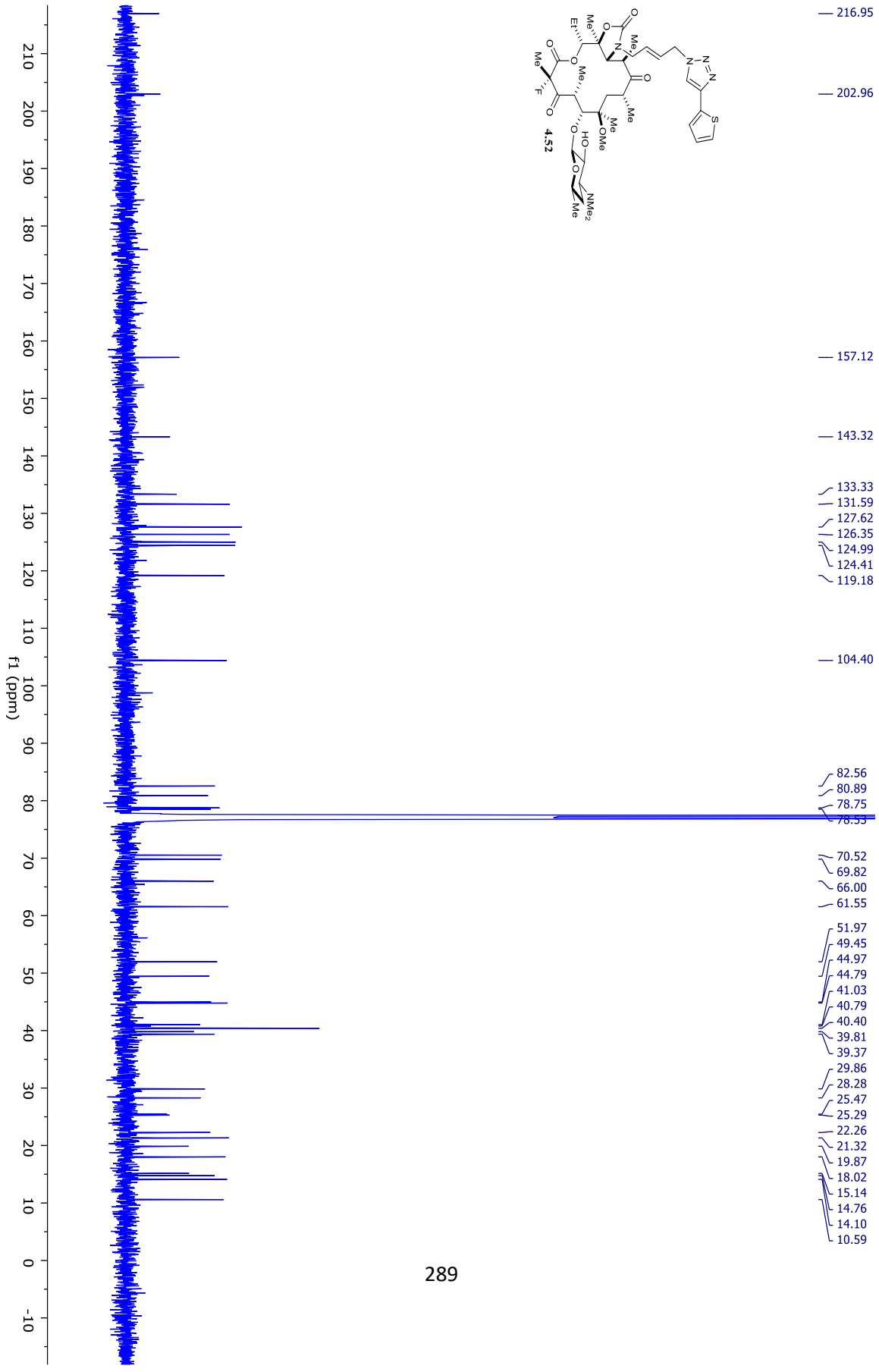

7.72
7.22
7.22
7.22
7.19
7.17
7.16
7.13
7.12
6.65
6.64
6.63
6.63
6.07
6.06
6.05
6.04
6.02
6.02
6.00
5.36
5.34
5.26
5.23
5.21
5.20
5.19
5.18
5.17
5.14
5.13
5.11
5.10
3.83
3.82
3.81
3.81
3.79
3.78
3.77
3.76
3.75
3.74
3.74
3.72
3.71
1.42
1.39
NH2
N
N
N
BocHN
4.40
284
1.00
0.97
2.00
0.98
0.98
1.02
1.13
1.71
3.76
9.06
7.8 7.6 7.4 7.2 7.0 6.8 6.6 6.4 6.2 6.0 5.8 5.6 5.4 5.2 5.0 4.8 4.6 4.4 4.2 4.0 3.8 3.6 3.4 3.2 3.0 2.8 2.6 2.4 2.2 2.0 1.8 1.6 1.4 1.2 1.0
f1 (ppm)

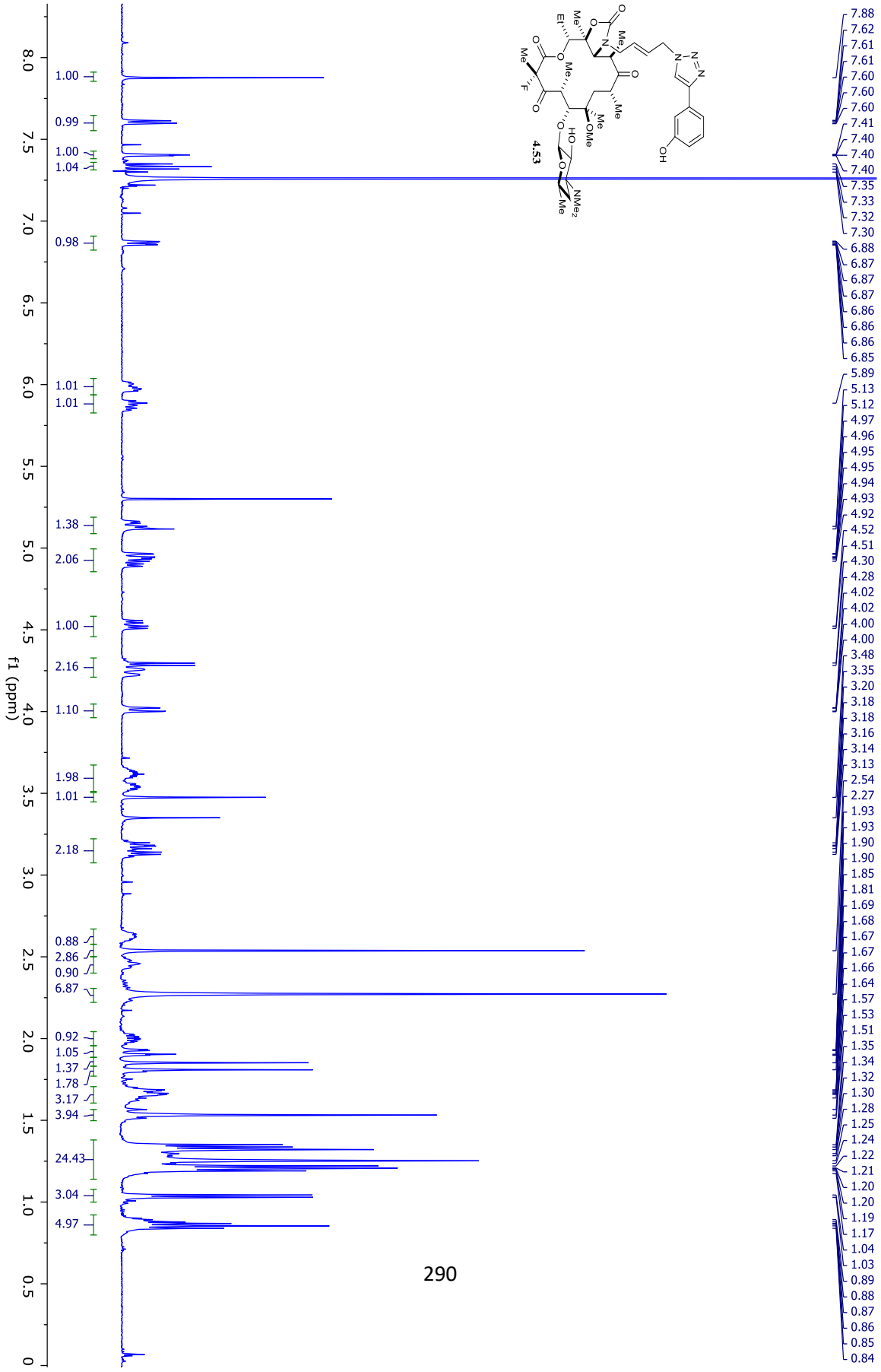

BocHN
N
N
N
NH2
4.40
156.0
147.8
147.1
133.5
131.4
129.9
119.9
116.0
115.1
112.2
80.0
63.5
43.9
29.8
28.4
f1 (ppm)

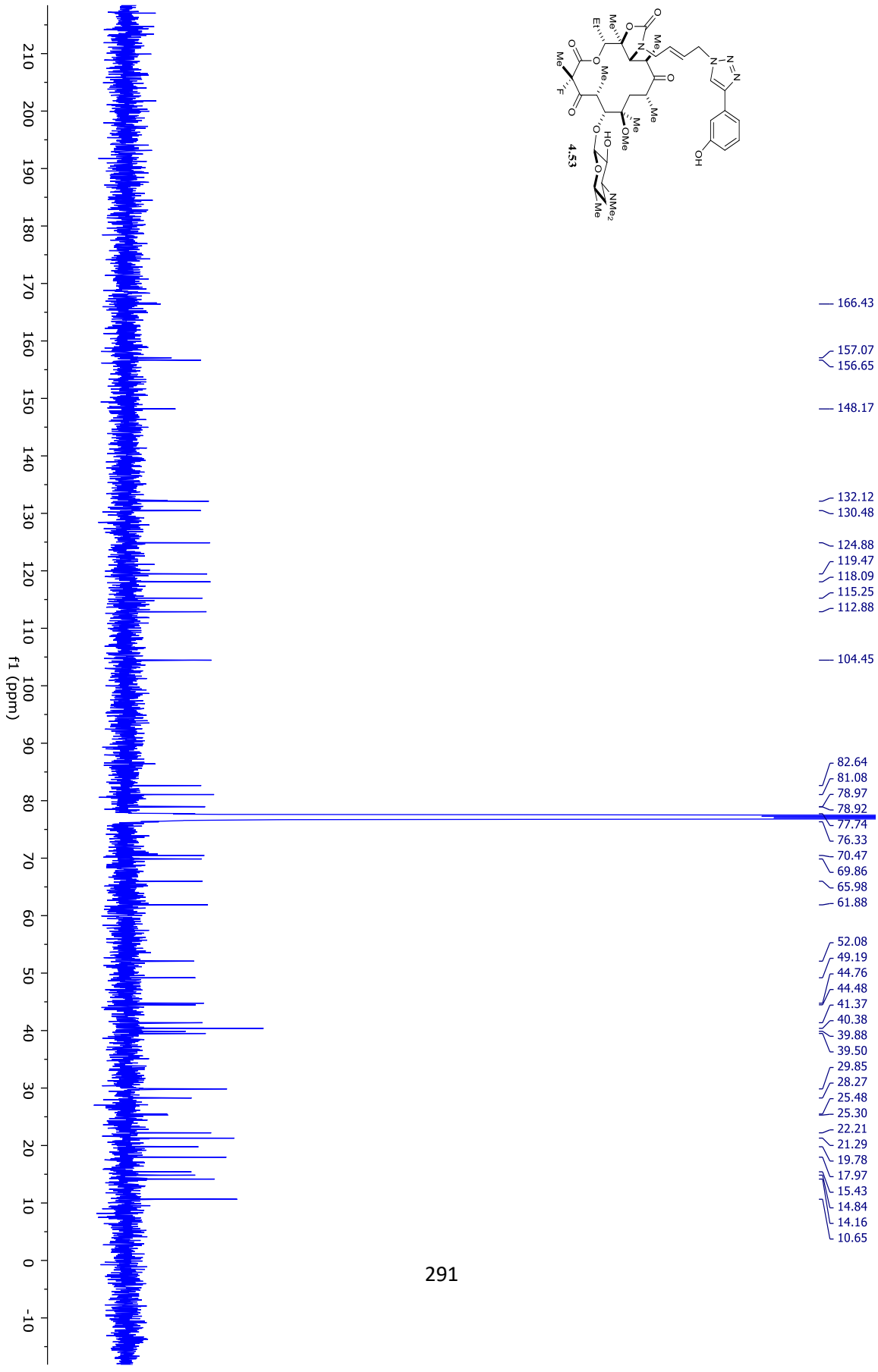

f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.05
2.41
1.09
1.99
1.82
1.06
1.00
1.12
1.02
1.02
2.25
1.08
1.14
1.05
2.69
3.71
7.63
1.60
0.74
0.88
2.92
1.13
0.88
3.88
20.10
3.56
4.57
4.51
7.85
7.31
7.31
7.30
7.26
7.25
7.23
7.20
7.18
7.17
6.66
6.65
6.63
5.94
5.93
5.92
5.90
4.99
4.98
4.97
4.90
4.90
4.88
4.42
4.41
4.38
4.37
4.32
4.30
4.25
4.24
4.20
4.06
4.03
3.56
3.54
3.51
3.45
3.25
3.23
3.22
3.20
3.11
3.09
2.63
2.60
2.57
2.53
2.51
2.34
2.17
2.04
1.97
1.97
1.96
1.90
1.89
1.86
1.85
1.82
1.76
1.72
1.70
1.69
1.66
1.63
1.56
1.53
1.51
1.47
1.32
1.31
1.30
1.28
1.26
1.25
1.25
1.23
1.21
1.19
1.17
1.03
1.01
0.88
0.86
0.84
0.82

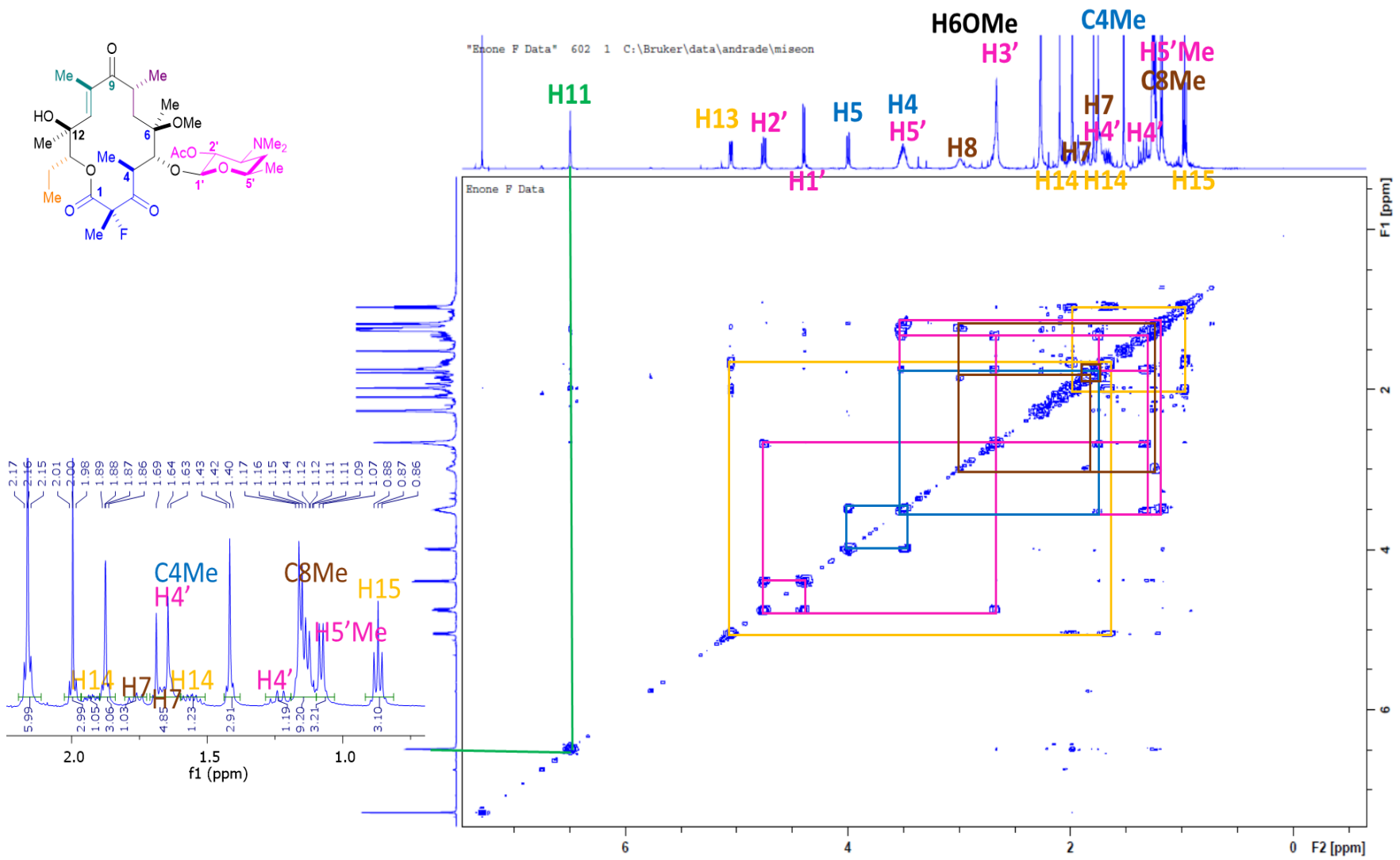

f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.98
203.36
203.07
157.13
148.32
147.01
131.82
131.44
129.78
126.26
119.67
116.39
114.89
112.51
104.39
82.57
80.96
78.74
78.51
70.49
69.81
65.96
61.54
51.96
49.40
44.88
44.78
41.02
40.39
39.79
39.36
29.85
28.53
28.26
25.53
25.31
22.22
21.31
19.85
18.00
15.20
14.75
14.11
10.59
4.51

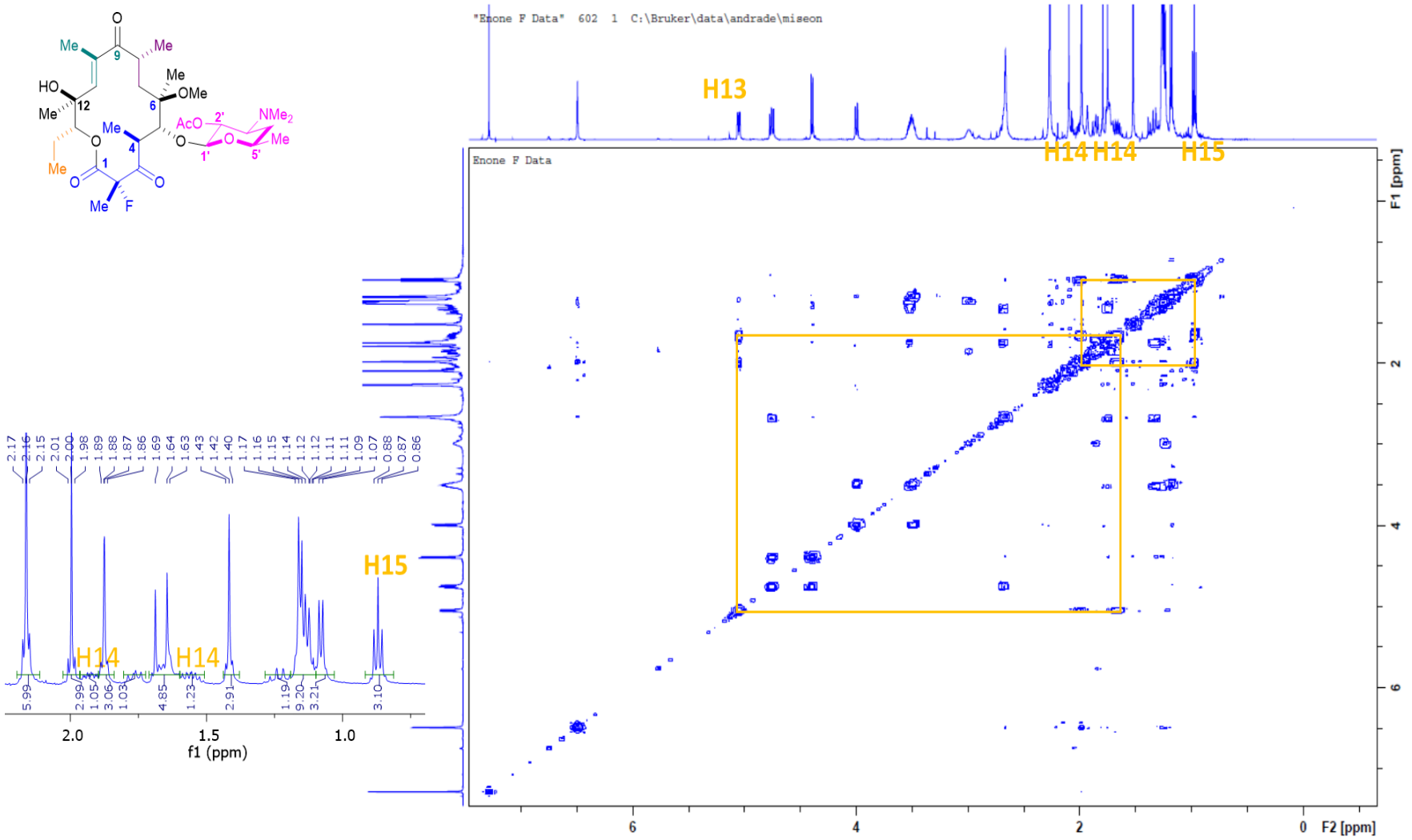

f1 (ppm)
4.52

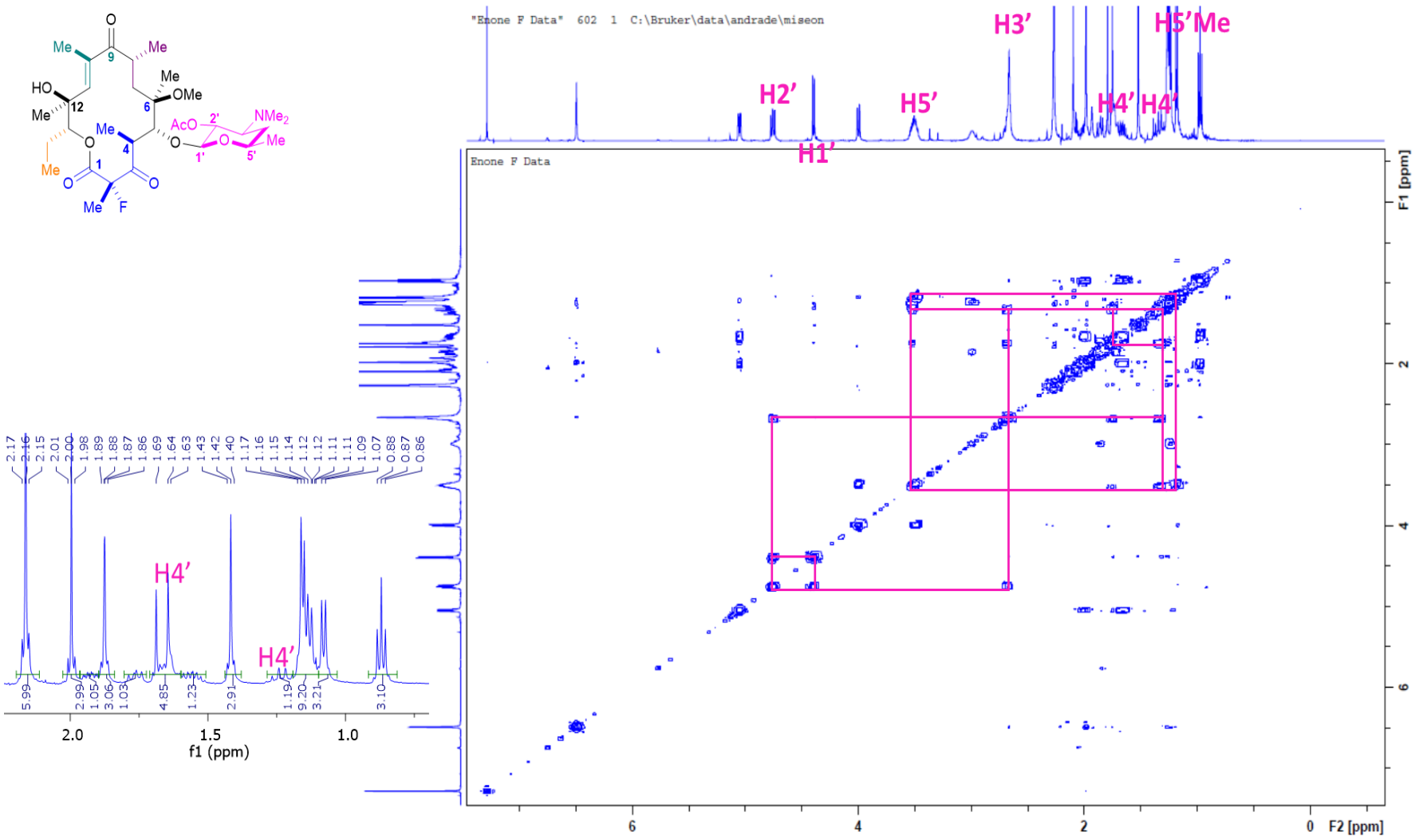

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
216.95
202.96
157.12
143.32
133.33
131.59
127.62
126.35
124.99
124.41
119.18
104.40
82.56
80.89
78.75
78.53
70.52
69.82
66.00
61.55
51.97
49.45
44.97
44.79
41.03
40.79
40.40
39.81
39.37
29.86
28.28
25.47
25.29
22.26
21.32
19.87
18.02
15.14
14.76
14.10
10.59
4.52

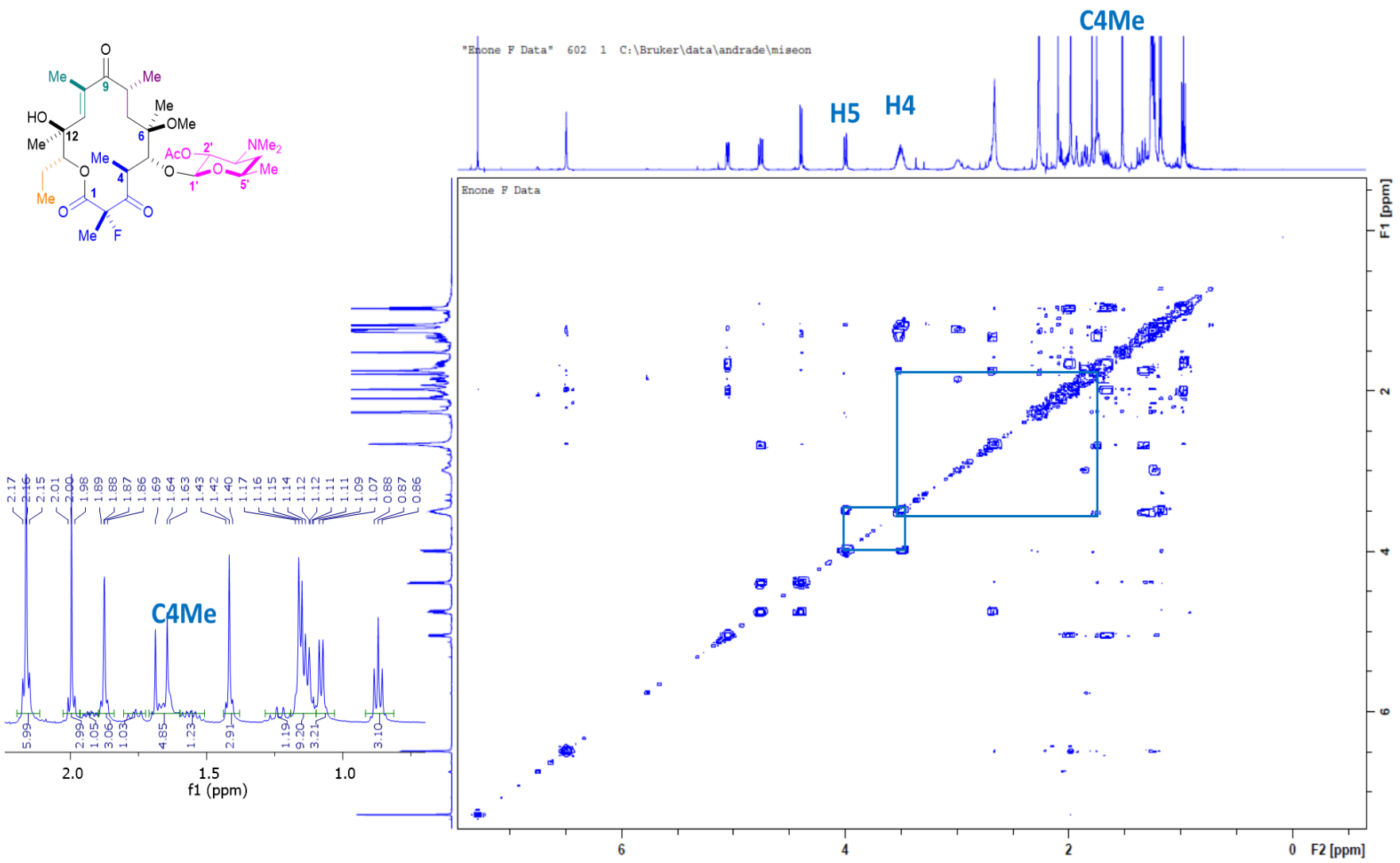

f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0
1.00
0.99
1.00
1.04
0.98
1.01
1.01
1.38
2.06
1.00
2.16
1.10
1.98
1.01
2.18
0.88
2.86
0.90
6.87
0.92
1.05
1.37
1.78
3.17
3.94
24.43
3.04
4.97
4.53
7.88
7.62
7.61
7.61
7.60
7.60
7.60
7.41
7.40
7.40
7.40
7.35
7.33
7.32
7.30
6.88
6.87
6.87
6.87
6.86
6.86
6.86
6.85
5.89
5.13
5.12
4.97
4.96
4.95
4.95
4.94
4.93
4.92
4.52
4.51
4.30
4.28
4.02
4.02
4.00
4.00
3.48
3.35
3.20
3.18
3.18
3.16
3.14
3.13
2.54
2.27
1.93
1.93
1.90
1.90
1.85
1.81
1.69
1.68
1.67
1.67
1.66
1.64
1.57
1.53
1.51
1.35
1.34
1.32
1.30
1.28
1.25
1.24
1.22
1.21
1.20
1.20
1.19
1.17
1.04
1.03
0.89
0.88
0.87
0.86
0.85
0.84

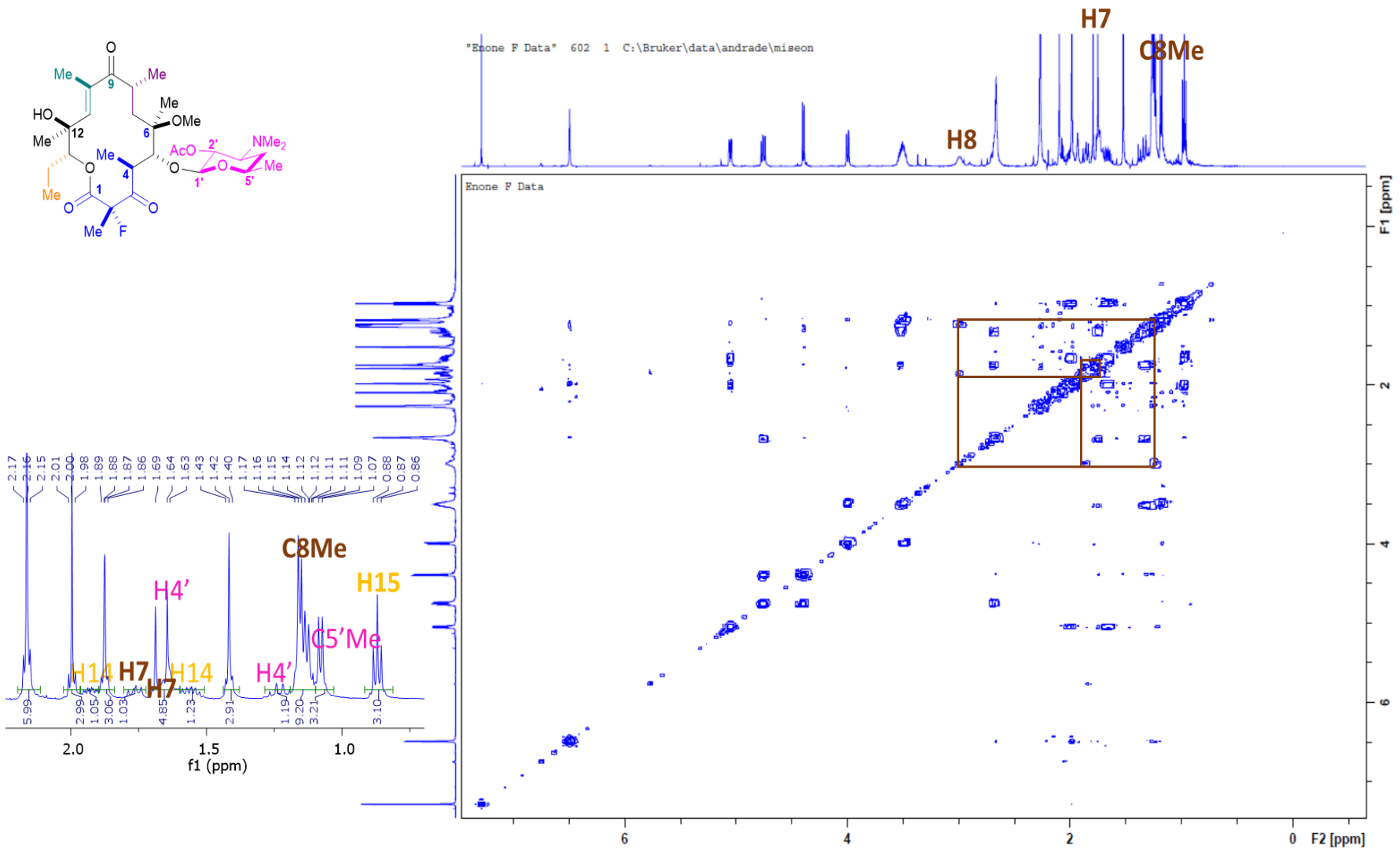

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
166.43
157.07
156.65
148.17
132.12
130.48
124.88
119.47
118.09
115.25
112.88
104.45
82.64
81.08
78.97
78.92
77.74
76.33
70.47
69.86
65.98
61.88
52.08
49.19
44.76
44.48
41.37
40.38
39.88
39.50
29.85
28.27
25.48
25.30
22.21
21.29
19.78
17.97
15.43
14.84
14.16
10.65
4.53

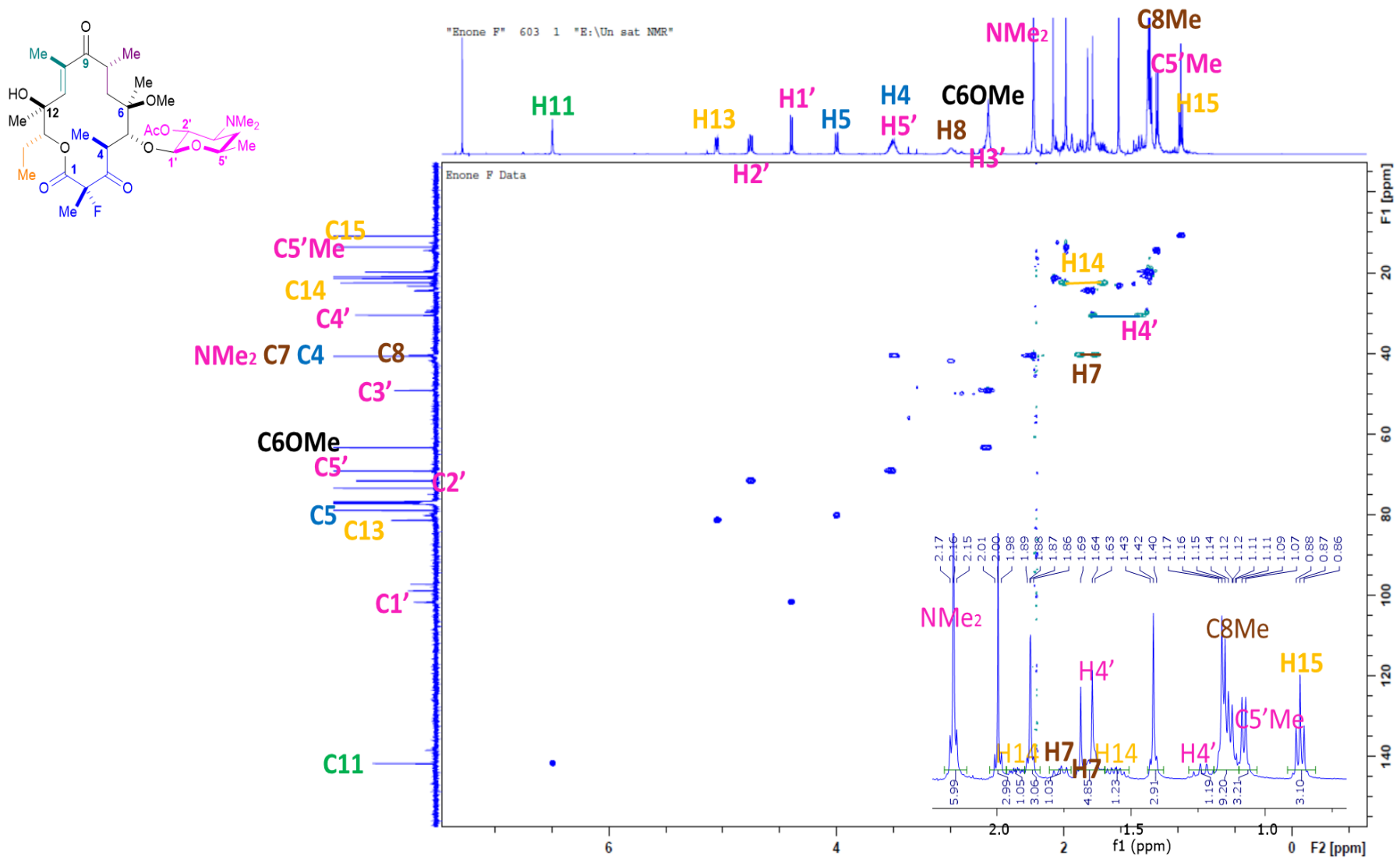

"Enone F Data" 602 1 C:\Bruker\data\andrade\miseon
H6OMe
C4Me
H3'
H5'Me
C8Me
H11
H13
H2'
H5
H4
H5'
H8
H7
H4'
H4'
H7
H1'
H14
H14
H15
Enone F Data
F1 [ppm]
F2 [ppm]
C4Me
H4'
C8Me
H15
H5'Me
H14
H7
H7
H14
H4'
f1 (ppm)

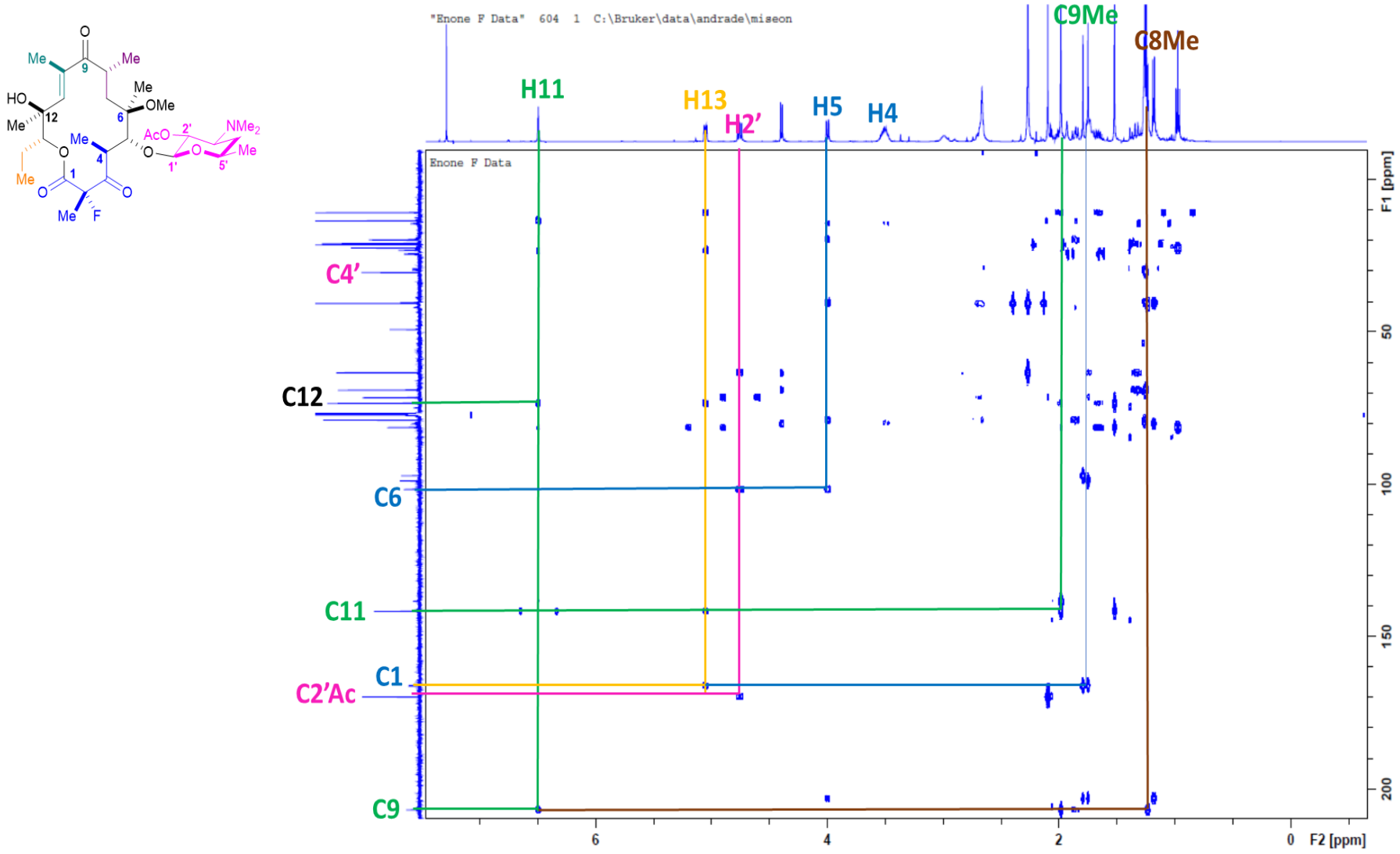

"Enone F Data" 602 1 C:\Bruker\data\andrade\miseon
H13
H14 H14
H15
Enone F Data
F1 [ppm]
F2 [ppm]
H15
H14
H14
f1 (ppm)

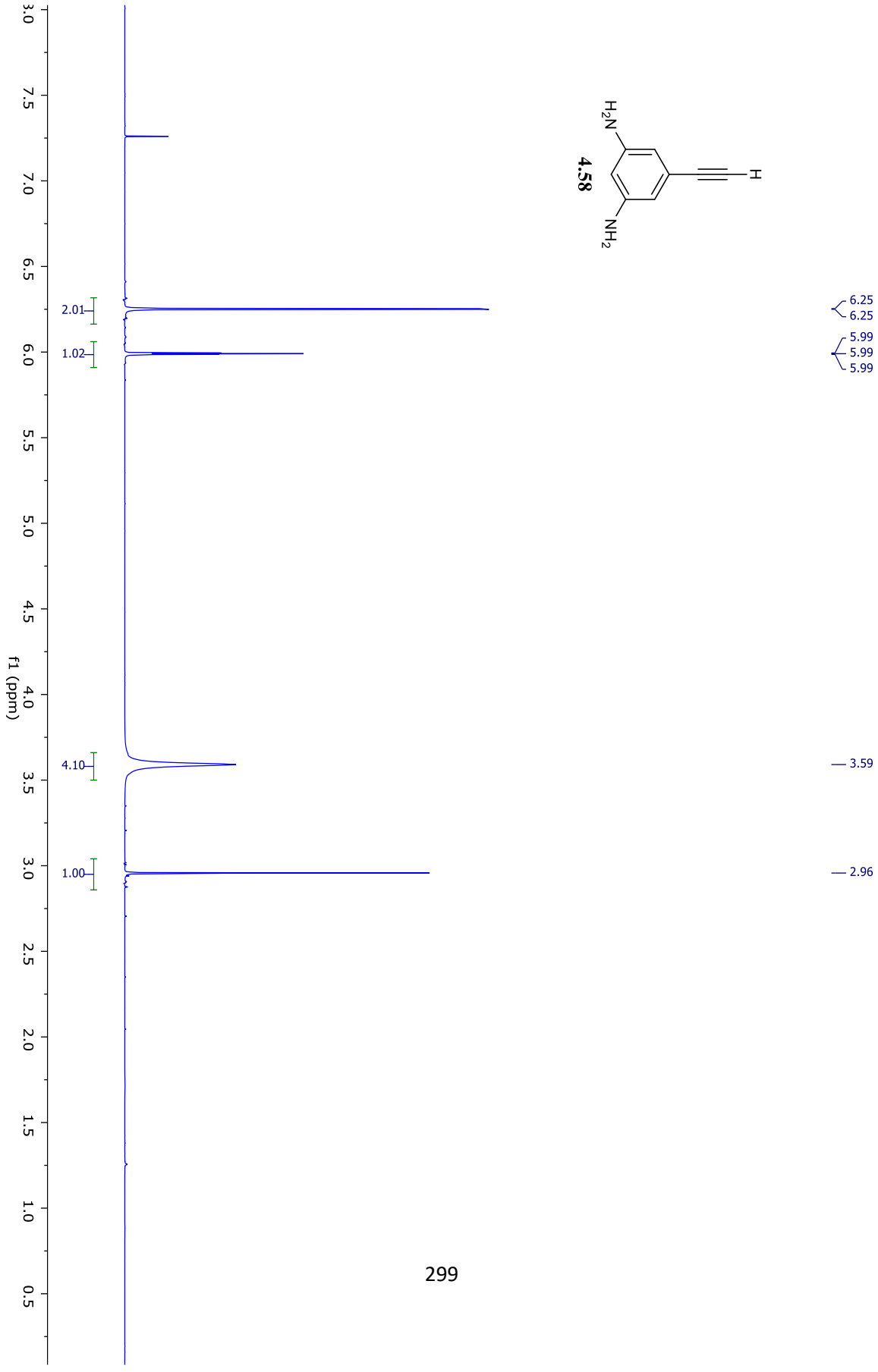

"Enone F Data" 602 1 C:\Bruker\data\andrade\miseon
H3'
H5'Me
H2'
H5'
H4'
H4'
H1'
Enone F Data
F1 [ppm]
F2 [ppm]
H4'
H4'
f1 (ppm)

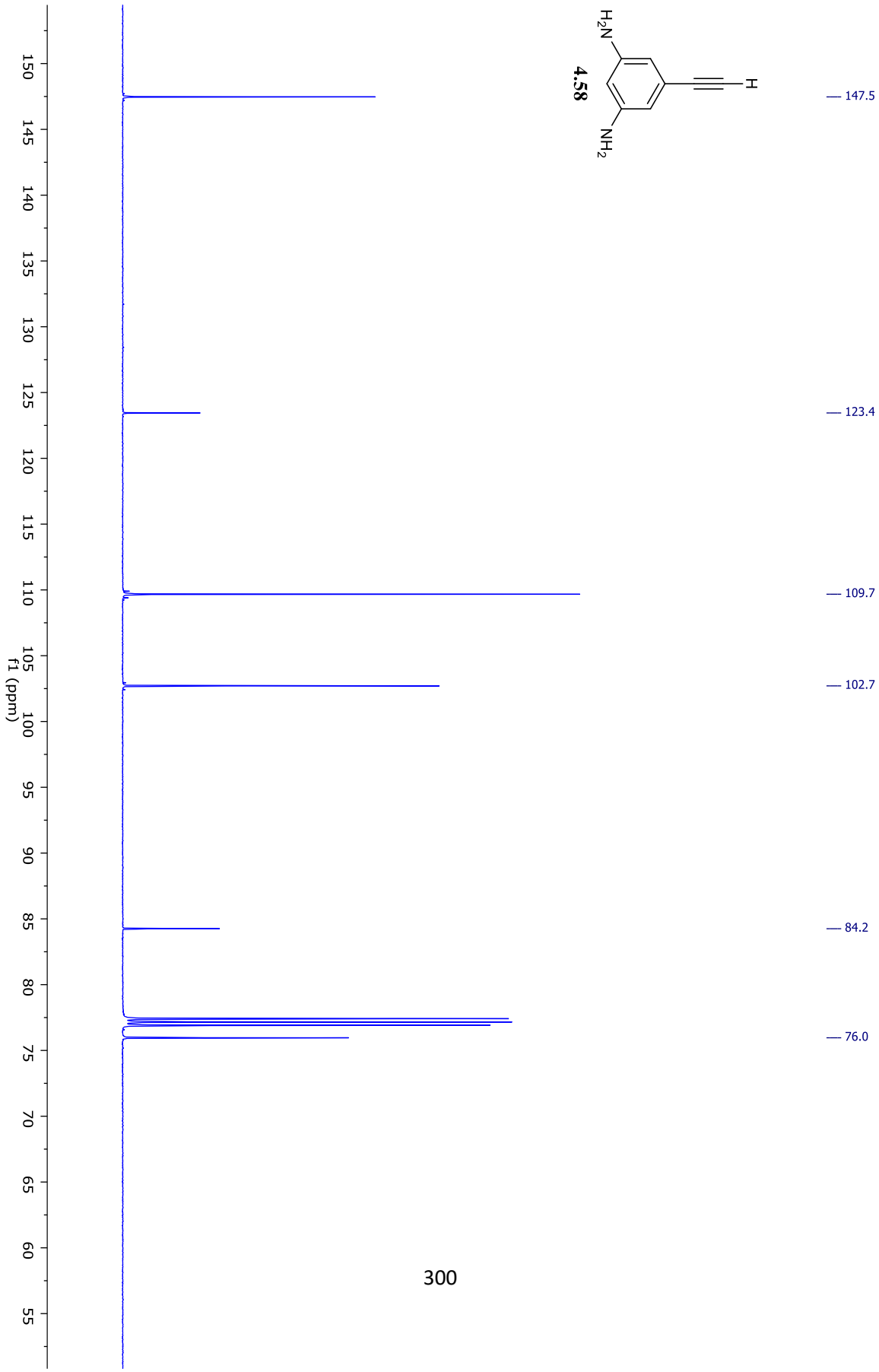

C4Me
"Enone F Data" 602 1 C:\Bruker\data\andrade\miseon
H5
H4
Enone F Data
F1 [ppm]
F2 [ppm]
C4Me
f1 (ppm)

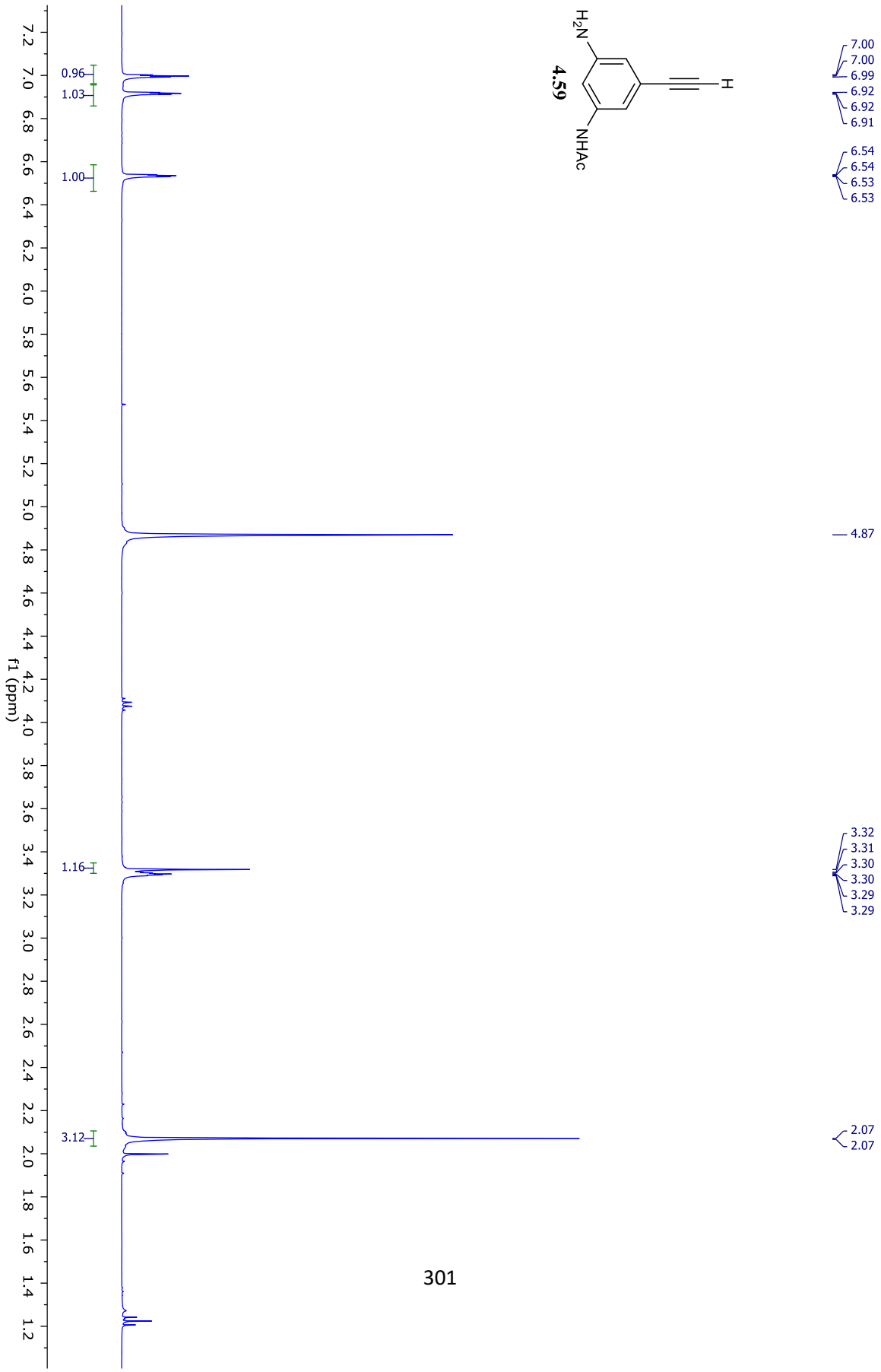

"Enone F Data" 602 1 C:\Bruker\data\andrade\miseon
H7
C8Me
H8
Enone F Data
F1 [ppm]
F2 [ppm]
C8Me
H15
H4'
C5'Me
H14
H7
H7
H14
H4'
f1 (ppm)

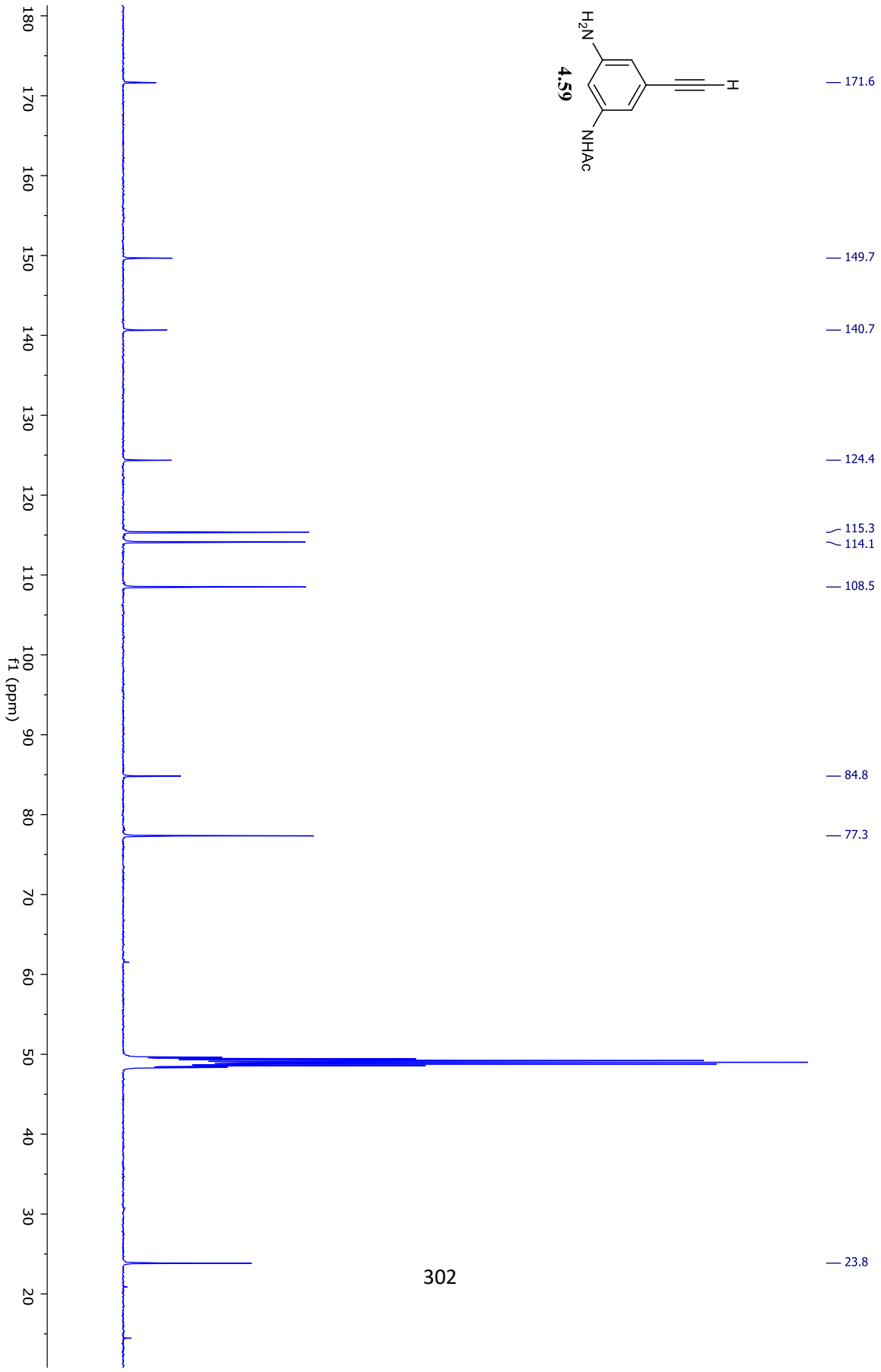

"Enone F" 603 1 "E:\Un sat NMR"
Enone F Data
NMe2
C8Me
C5'Me
H15
H11
H13
H1'
H5
H4
H5'
H8
C6OMe
H3'
H2'
H14
H4'
H7
C15
C5'Me
C14
C4'
NMe2 C7 C4
C8
C3'
C6OMe
C5'
C2'
C5
C13
C1'
C11
F1 [ppm]
f1 (ppm)
F2 [ppm]

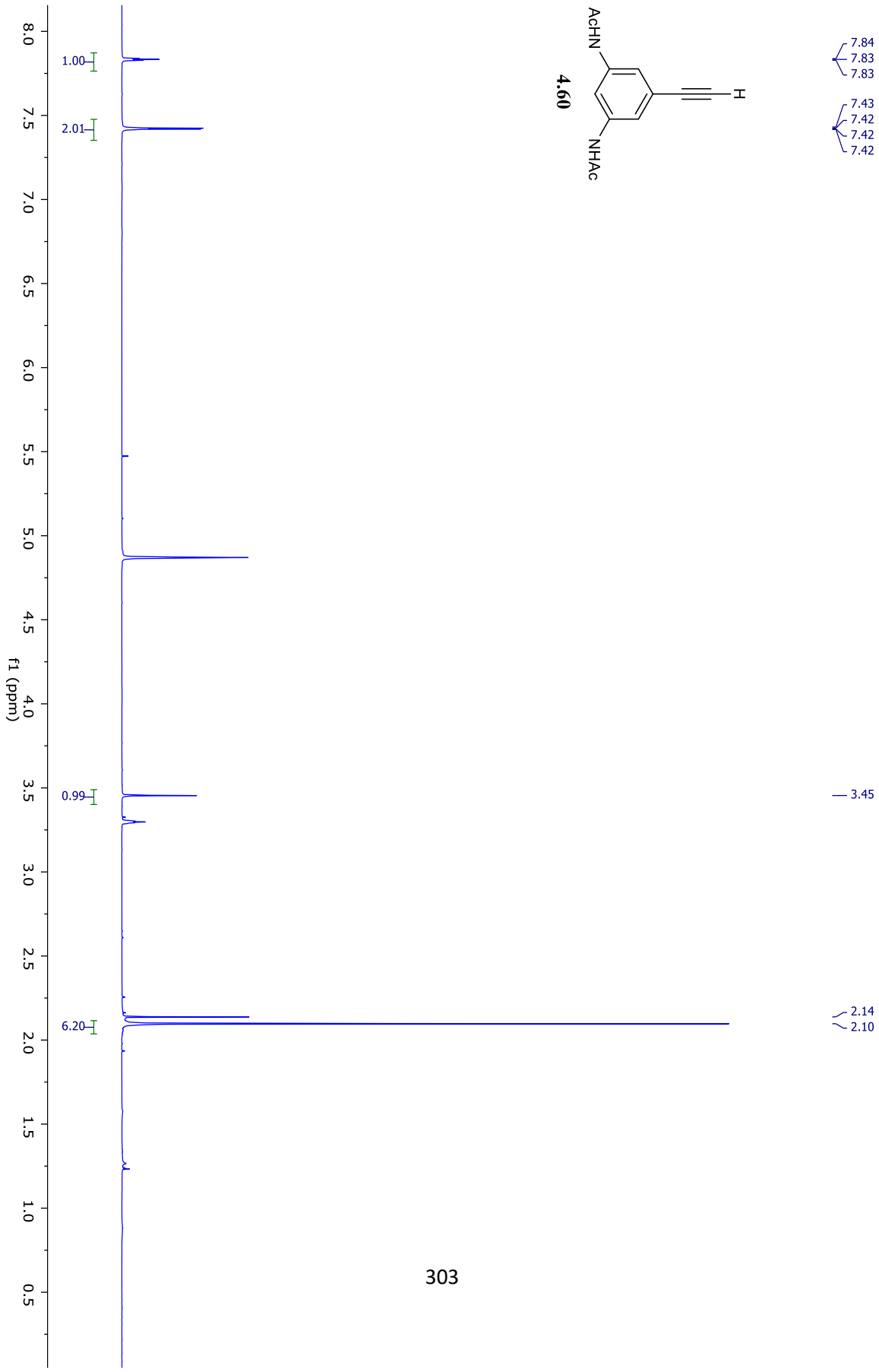

"Enone F Data" 604 1 C:\Bruker\data\andrade\miseon
Enone F Data
C9Me
C8Me
H11
H13
H2'
H5
H4
C4'
C12
C6
C11
C1
C2'Ac
C9
F1 [ppm]
F2 [ppm]
6
4
2
0
50
100
150
200

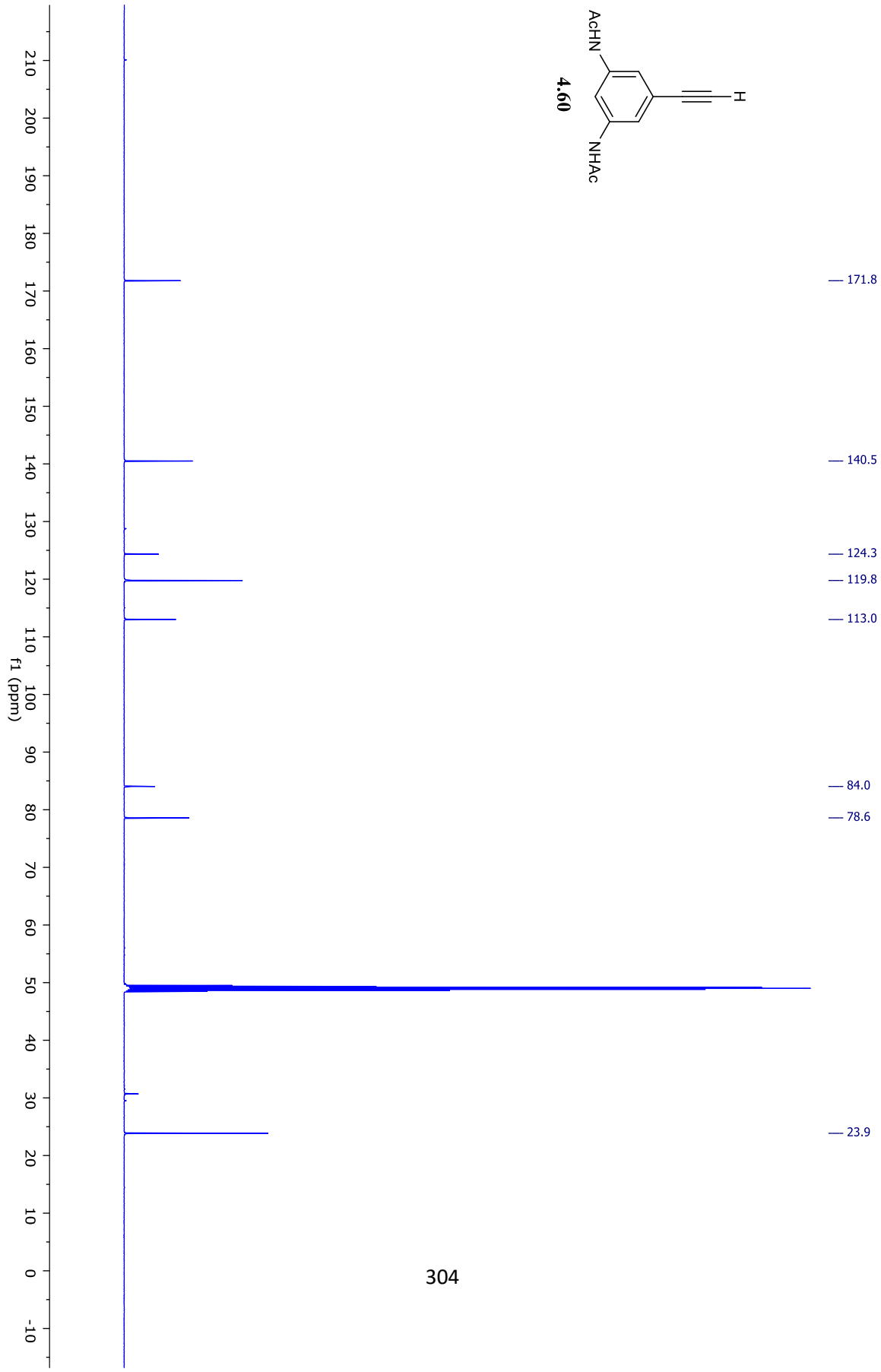

4.58
H2N
NH2
H
f1 (ppm)
3.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
2.01
1.02
4.10
1.00
6.25
6.25
5.99
5.99
5.99
3.59
2.96

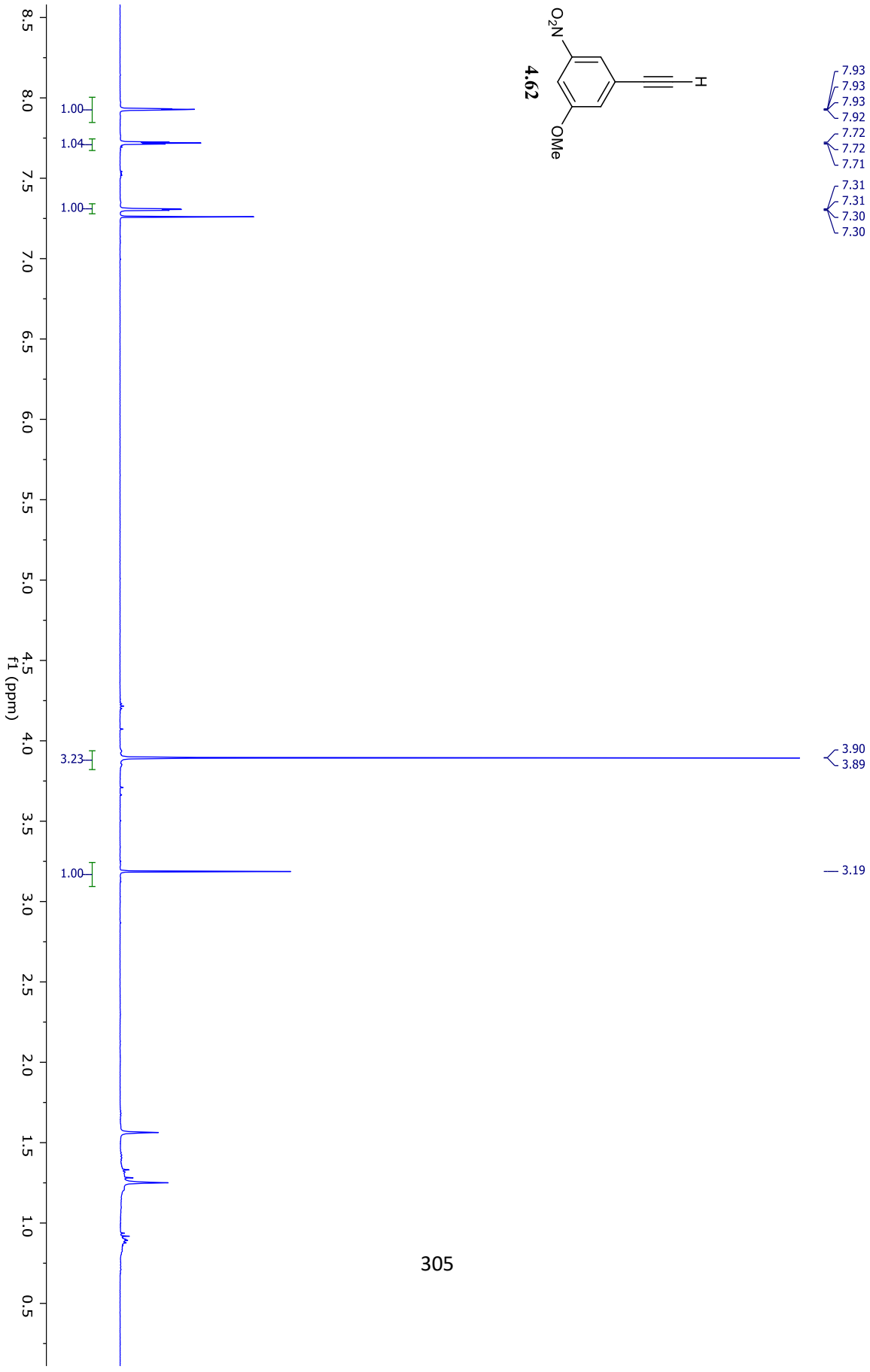

4.58
H2N
NH2
H
147.5
123.4
109.7
102.7
84.2
76.0
f1 (ppm)
150
145
140
135
130
125
120
115
110
105
100
95
90
85
80
75
70
65
60
55

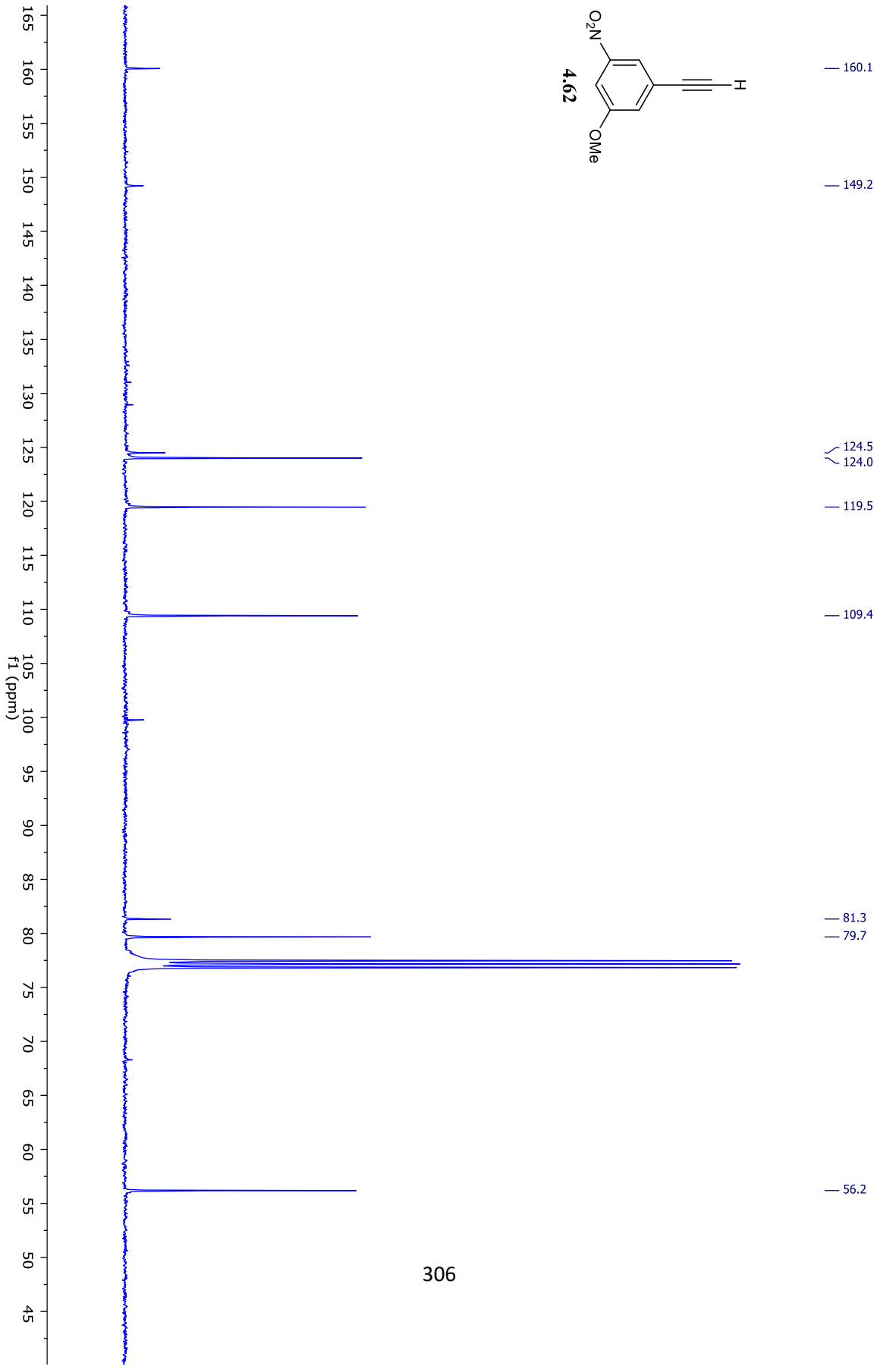

4.59
H2N
NHAc
H
7.00
7.00
6.99
6.92
6.92
6.91
6.54
6.54
6.53
6.53
4.87
3.32
3.31
3.30
3.30
3.29
3.29
2.07
2.07
0.96
1.03
1.00
1.16
3.12
f1 (ppm)

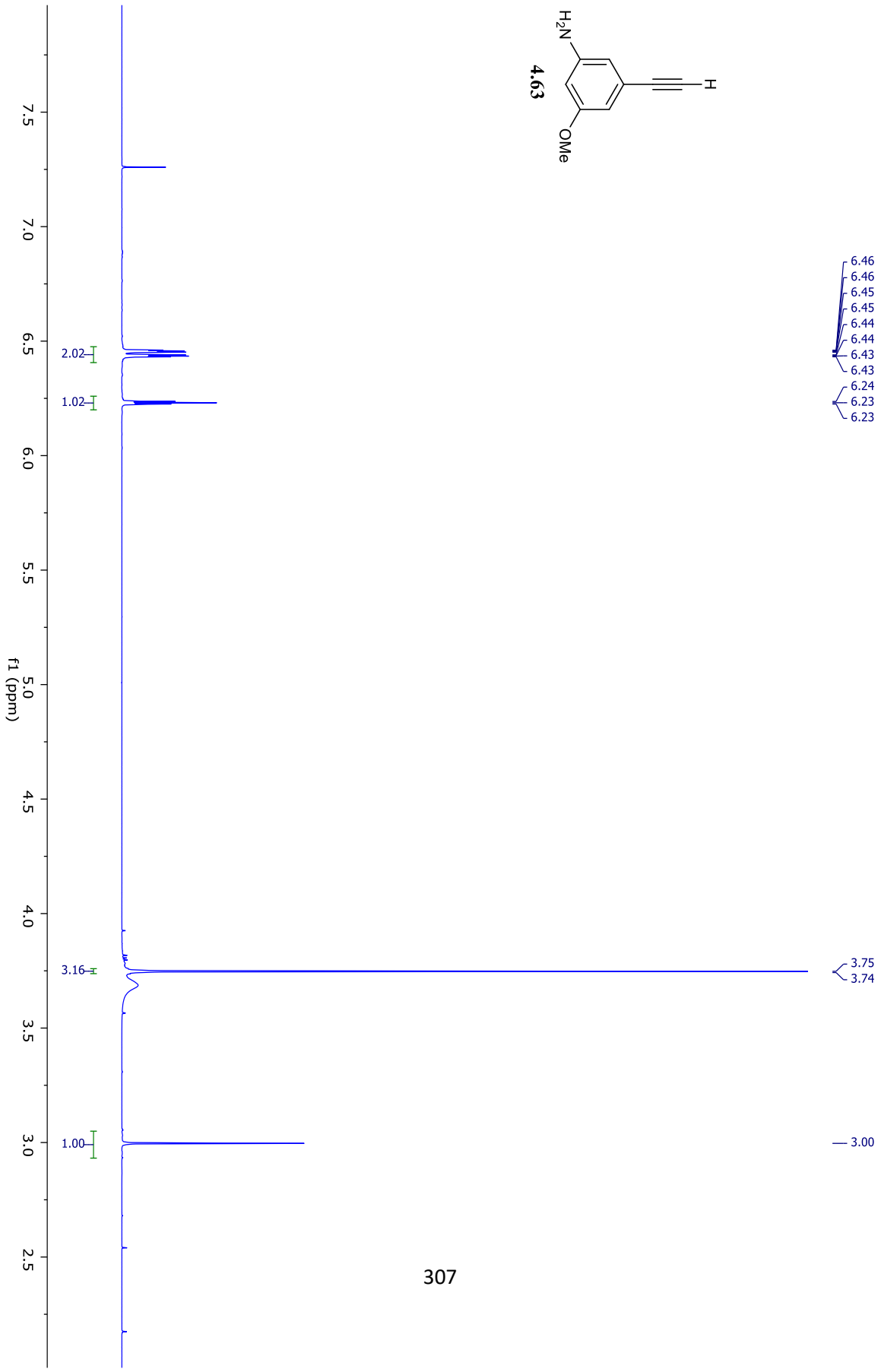

H2N
NHAc
H
4.59
171.6
149.7
140.7
124.4
115.3
114.1
108.5
84.8
77.3
23.8
f1 (ppm)
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20

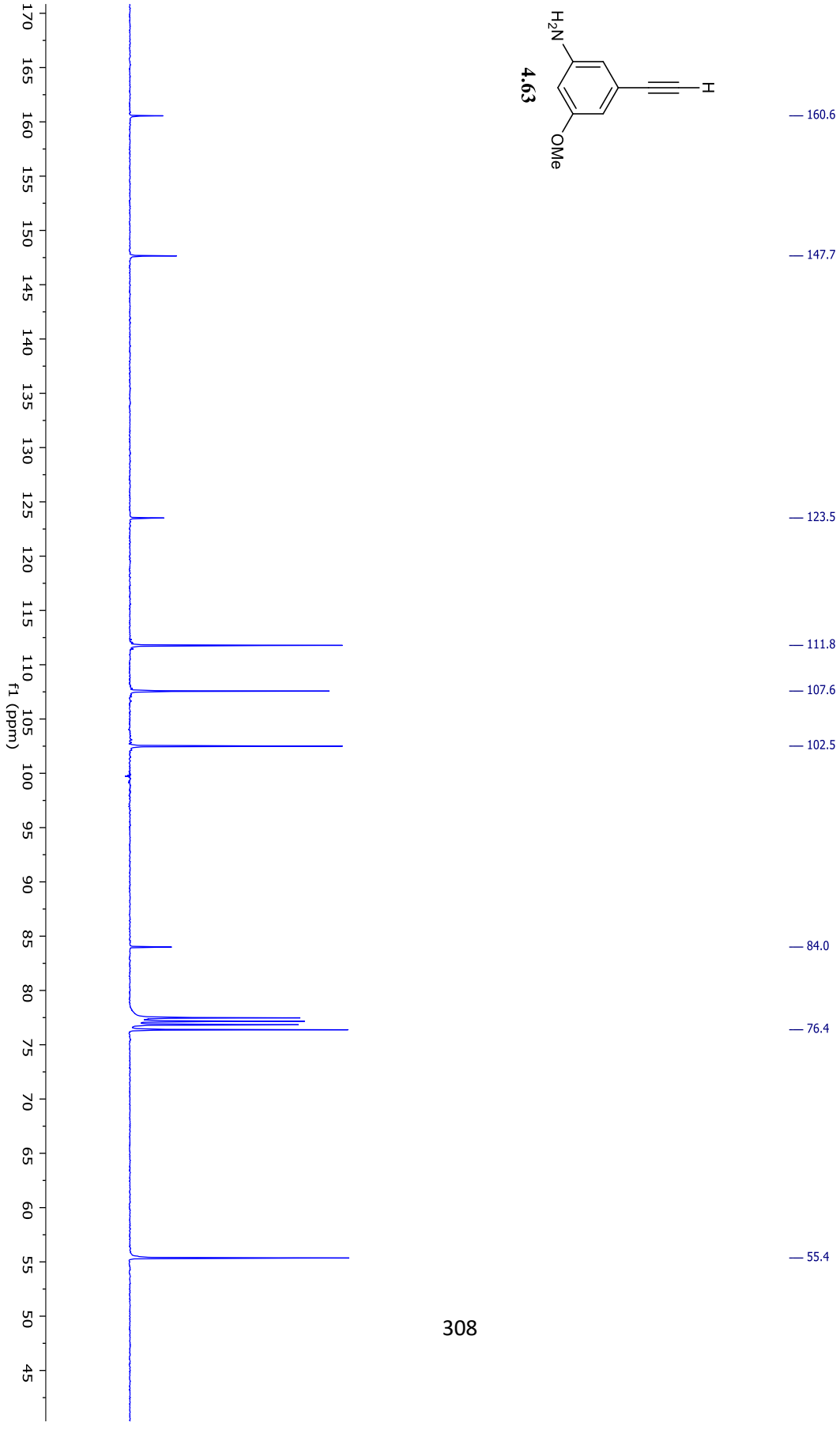

AcHN
NHAc
H
4.60
7.84
7.83
7.83
7.43
7.42
7.42
7.42
3.45
2.14
2.10
1.00
2.01
0.99
6.20
f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5

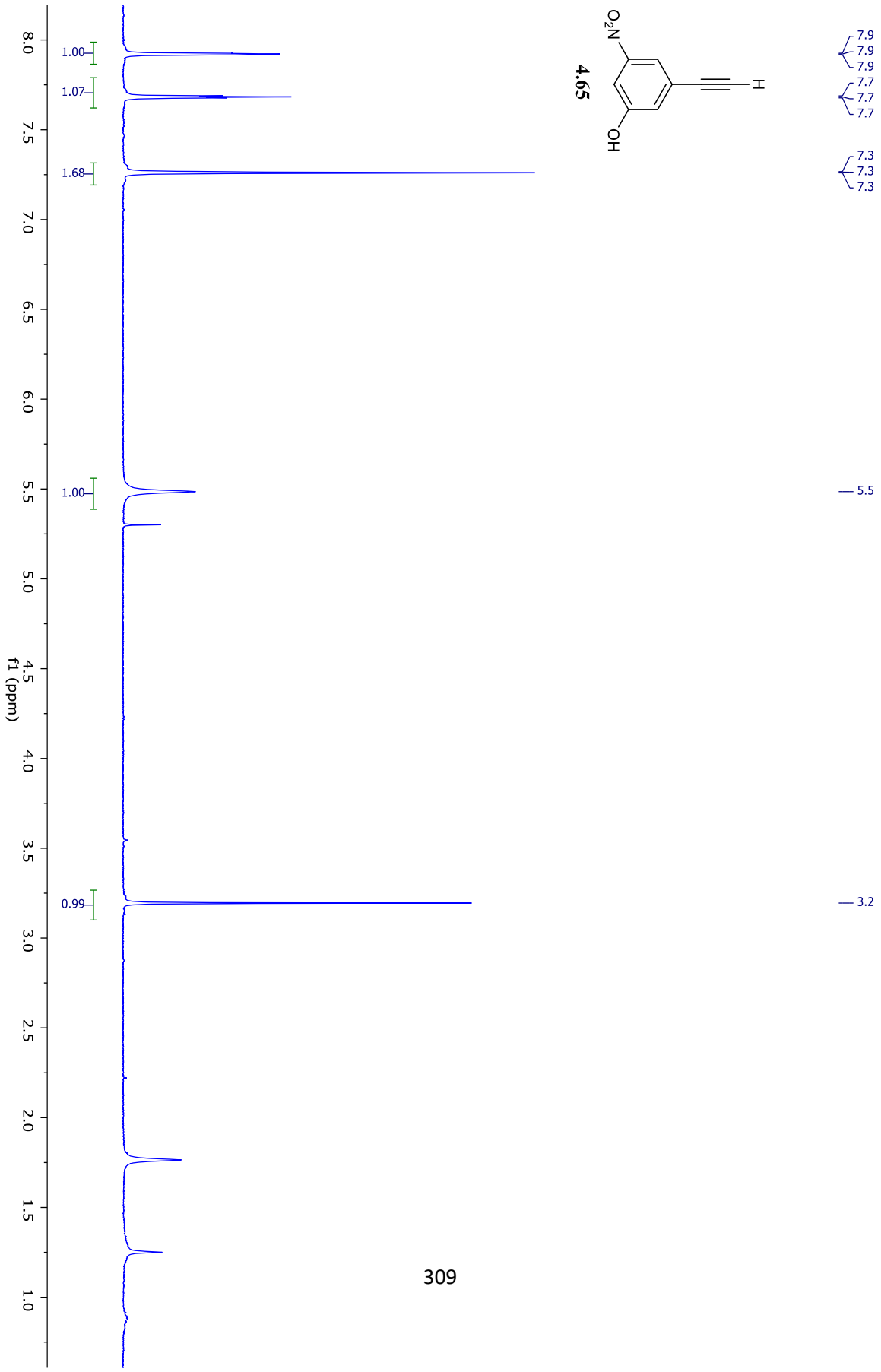

AcHN
NHAc
H
4.60
f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
171.8
140.5
124.3
119.8
113.0
84.0
78.6
23.9

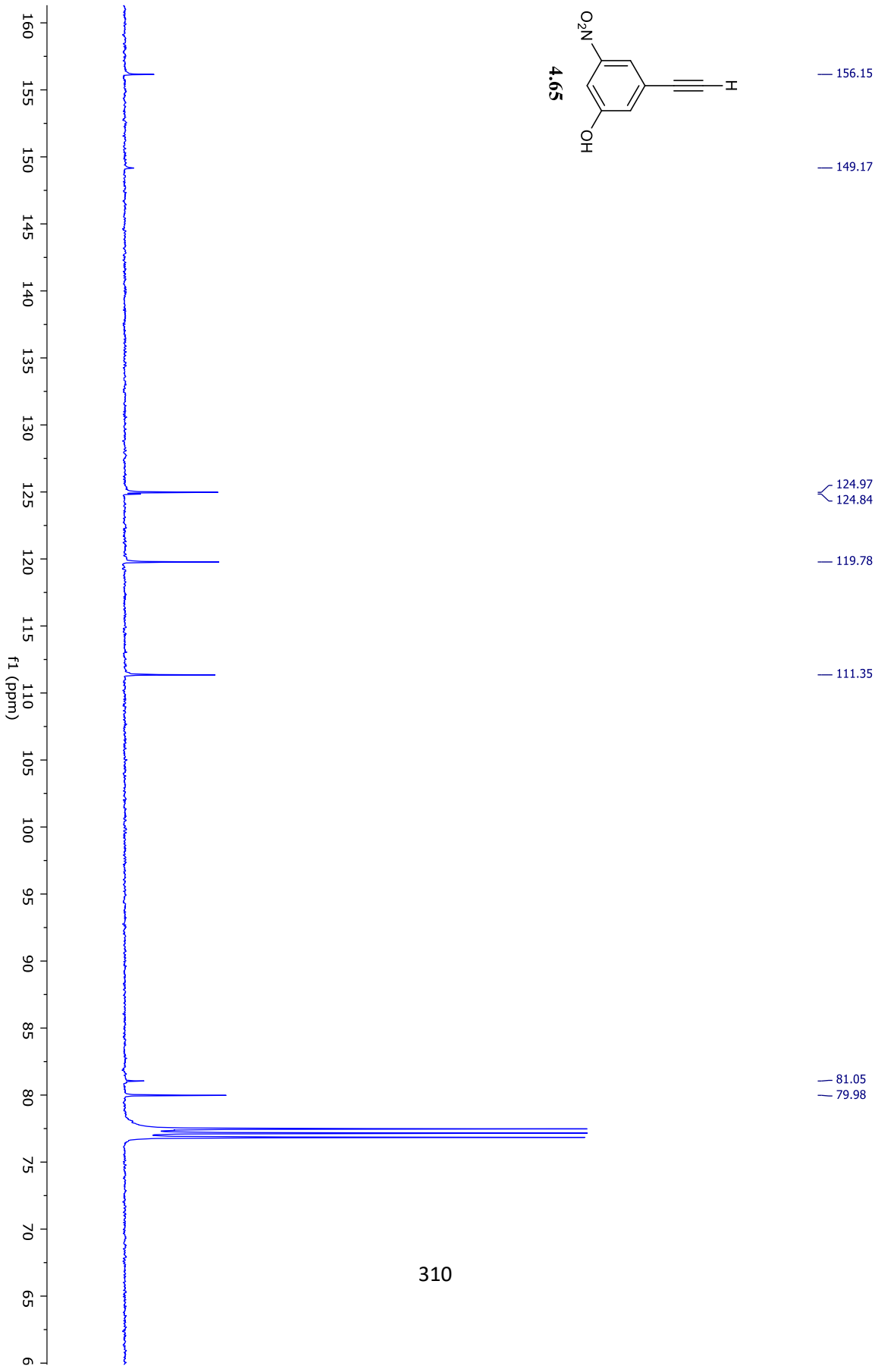

4.62
O2N
OMe
H
7.93
7.93
7.93
7.92
7.72
7.72
7.71
7.31
7.31
7.30
7.30
3.90
3.89
3.19
1.00
1.04
1.00
3.23
1.00
f1 (ppm)

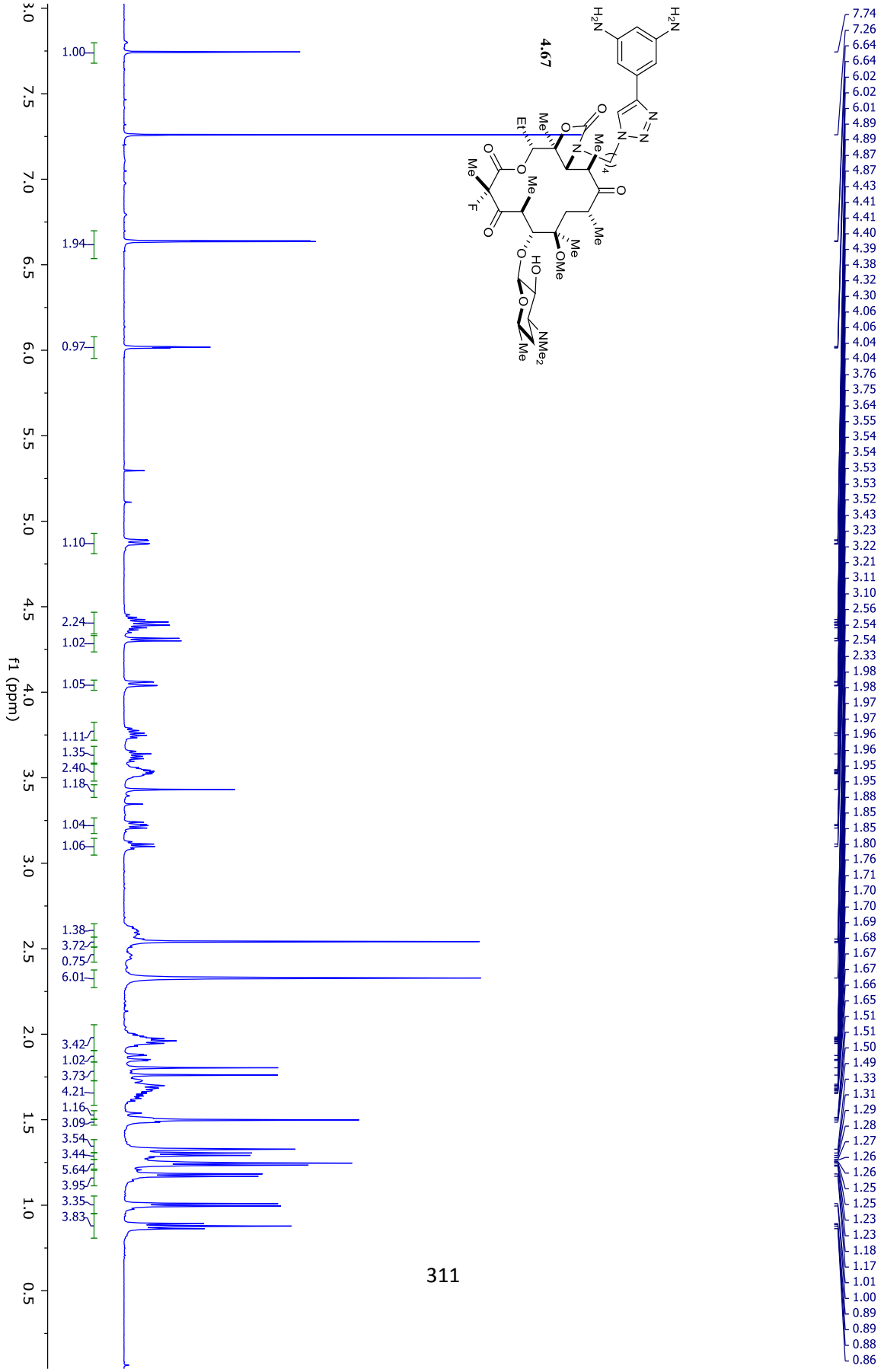

O2N
OMe
H
4.62
160.1
149.2
124.5
124.0
119.5
109.4
81.3
79.7
56.2
f1 (ppm)

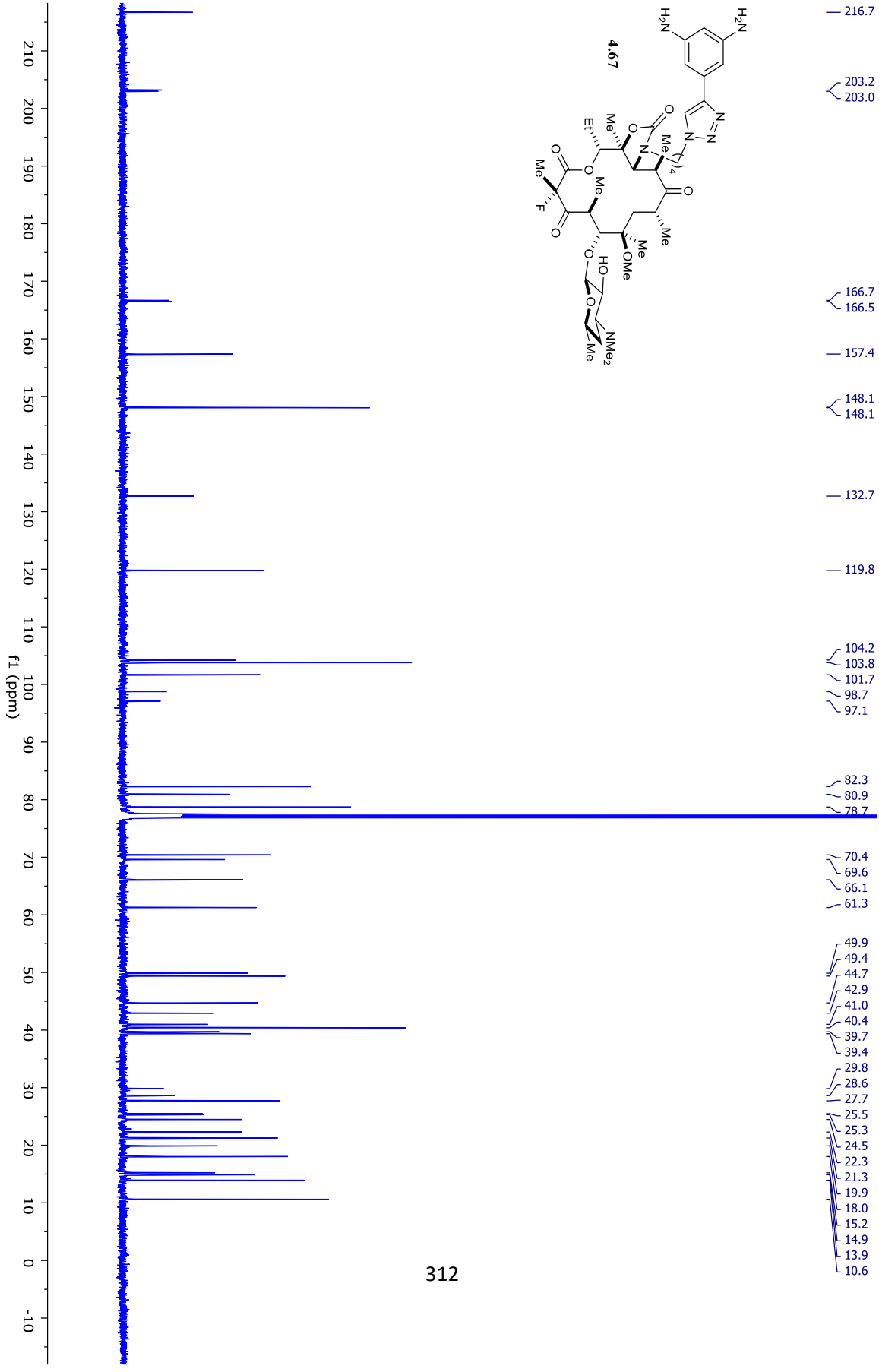

4.63
H2N
OMe
H
f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.02
1.02
3.16
1.00
6.46
6.46
6.45
6.45
6.44
6.44
6.43
6.43
6.24
6.23
6.23
3.75
3.74
3.00

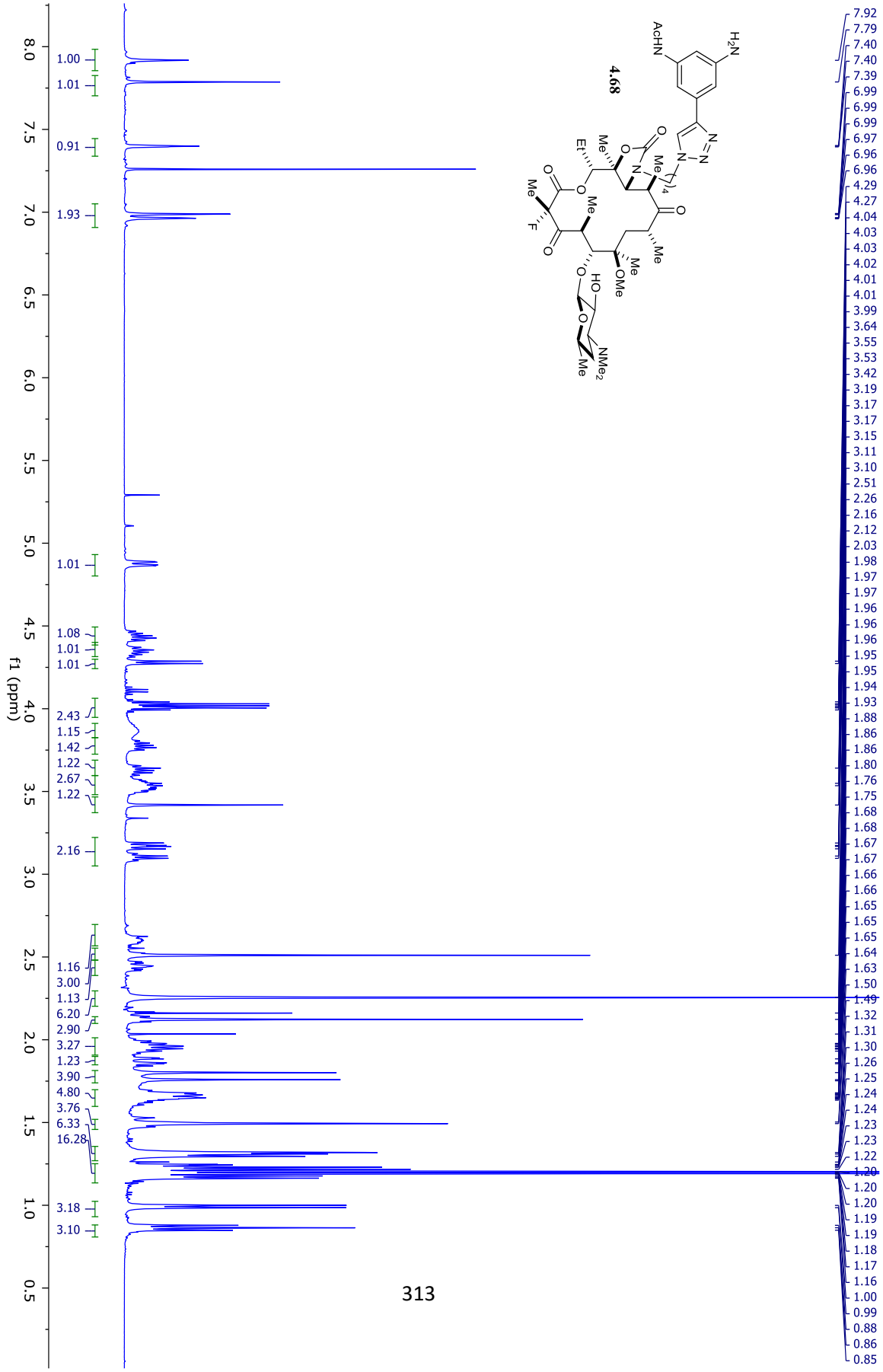

H2N
OMe
H
4.63
160.6
147.7
123.5
111.8
107.6
102.5
84.0
76.4
55.4
f1 (ppm)

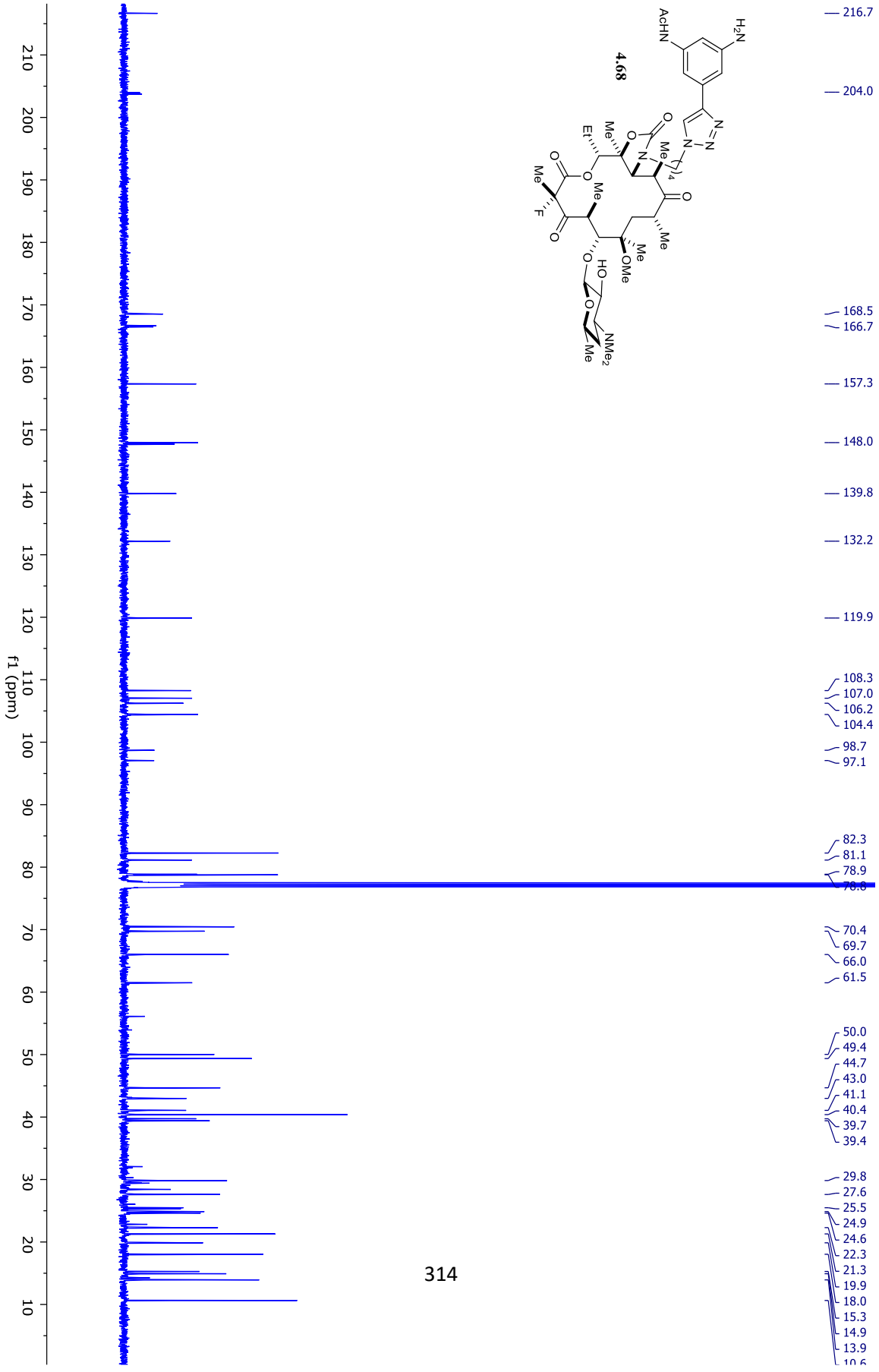

4.65
O2N
OH
H
1.00
1.07
1.68
1.00
0.99
7.9
7.9
7.9
7.7
7.7
7.7
7.3
7.3
7.3
5.5
3.2
f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0

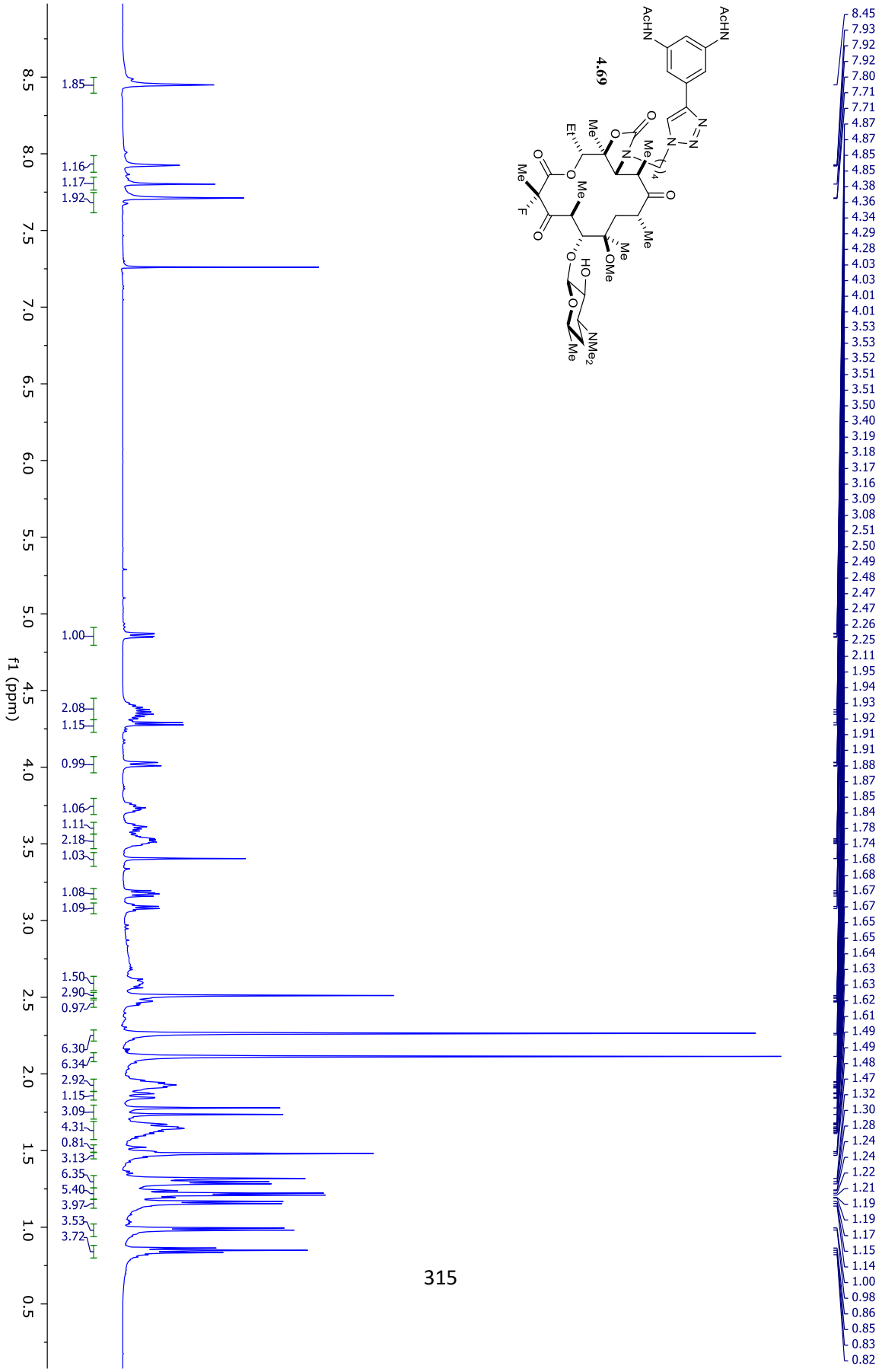

4.65
O2N
OH
H
156.15
149.17
124.97
124.84
119.78
111.35
81.05
79.98
f1 (ppm)
160
155
150
145
140
135
130
125
120
115
110
105
100
95
90
85
80
75
70
65
60

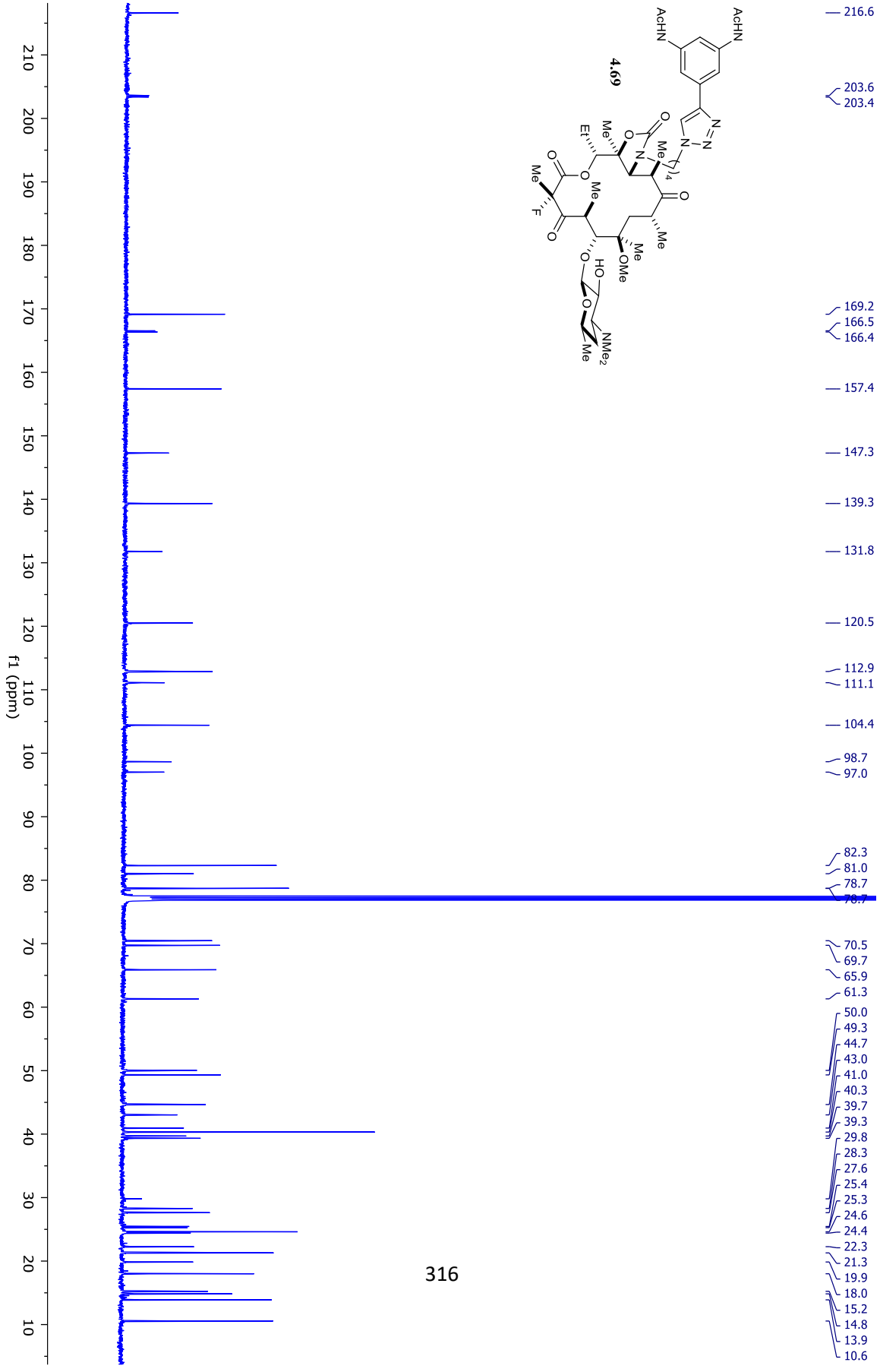

4.67
f1 (ppm)
3.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.94
0.97
1.10
2.24
1.02
1.05
1.11
1.35
2.40
1.18
1.04
1.06
1.38
3.72
0.75
6.01
3.42
1.02
3.73
4.21
1.16
3.09
3.54
3.44
5.64
3.95
3.35
3.83
7.74
7.26
6.64
6.64
6.02
6.02
6.01
4.89
4.89
4.87
4.87
4.43
4.41
4.41
4.40
4.39
4.38
4.32
4.30
4.06
4.06
4.04
4.04
3.76
3.75
3.64
3.55
3.54
3.54
3.53
3.53
3.52
3.43
3.23
3.22
3.21
3.11
3.10
2.56
2.54
2.54
2.33
1.98
1.98
1.97
1.97
1.96
1.96
1.95
1.95
1.88
1.85
1.85
1.80
1.76
1.71
1.70
1.70
1.69
1.68
1.67
1.67
1.66
1.65
1.51
1.51
1.50
1.49
1.33
1.31
1.29
1.28
1.27
1.26
1.26
1.25
1.25
1.23
1.23
1.18
1.17
1.01
1.00
0.89
0.89
0.88
0.86

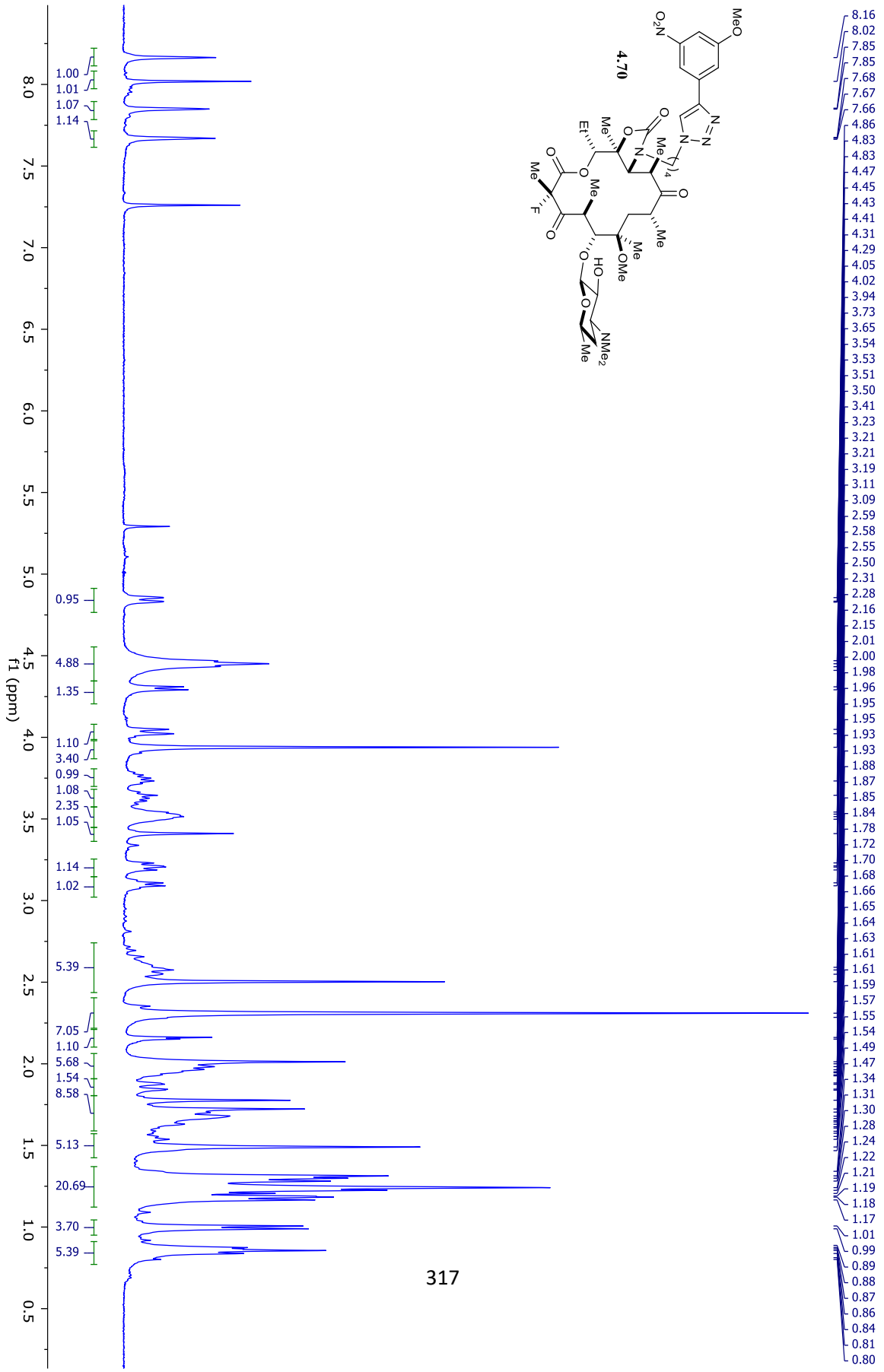

4.67
f1 (ppm)
216.7
203.2
203.0
166.7
166.5
157.4
148.1
148.1
132.7
119.8
104.2
103.8
101.7
98.7
97.1
82.3
80.9
78.7
70.4
69.6
66.1
61.3
49.9
49.4
44.7
42.9
41.0
40.4
39.7
39.4
29.8
28.6
27.7
25.5
25.3
24.5
22.3
21.3
19.9
18.0
15.2
14.9
13.9
10.6

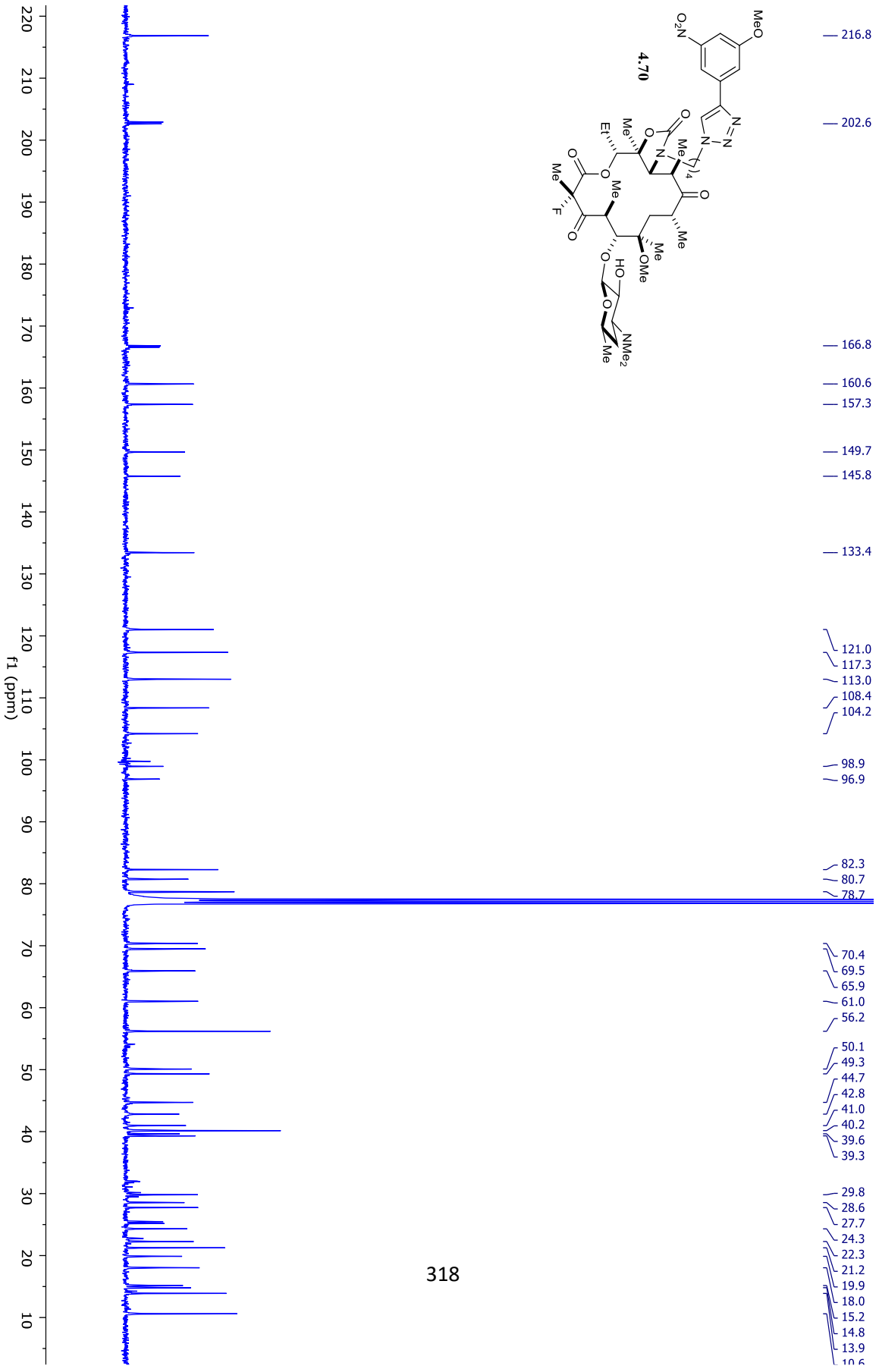

4.68
f1 (ppm)

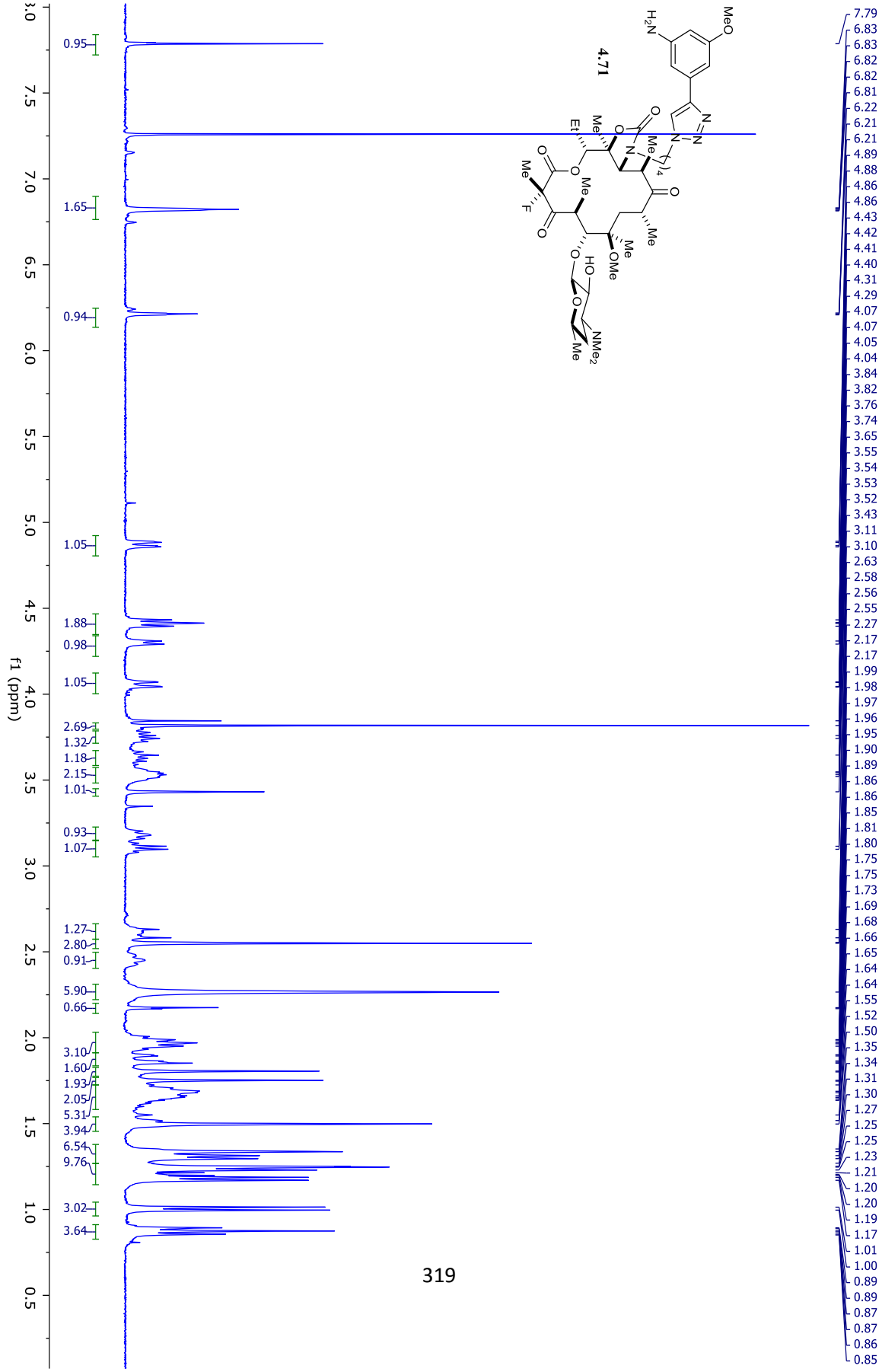

4.68
AcHN
H2N
N
N
N
Me
Et
Me
Me
Me
F
Me
Me
Me
OMe
HO
NMe2
Me
f1 (ppm)
216.7
204.0
168.5
166.7
157.3
148.0
139.8
132.2
119.9
108.3
107.0
106.2
104.4
98.7
97.1
82.3
81.1
78.9
78.6
70.4
69.7
66.0
61.5
50.0
49.4
44.7
43.0
41.1
40.4
39.7
39.4
29.8
27.6
25.5
24.9
24.6
22.3
21.3
19.9
18.0
15.3
14.9
13.9

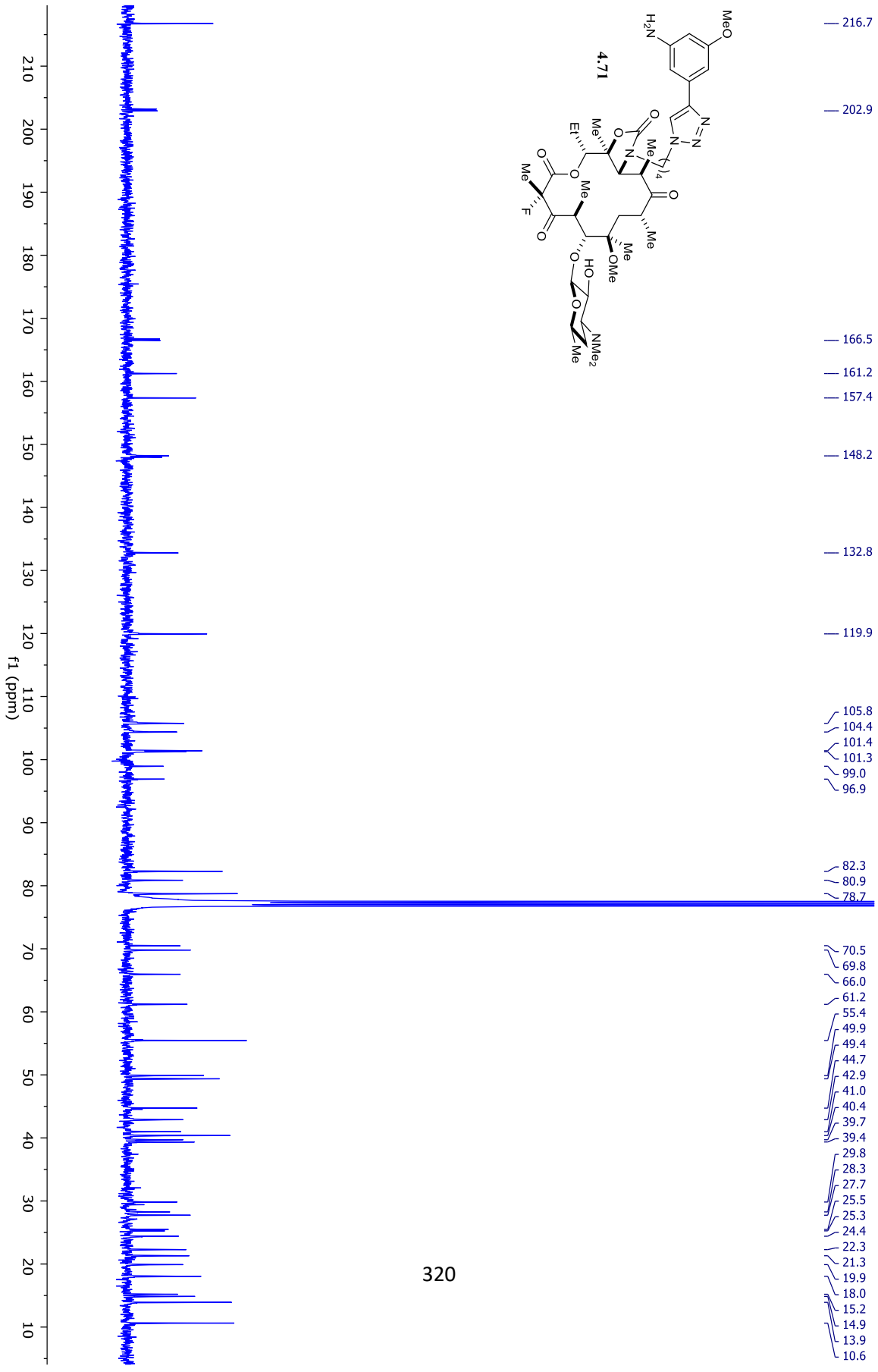

4.69
AcHN
AcHN
f1 (ppm)
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.85
1.16
1.17
1.92
1.00
2.08
1.15
0.99
1.06
1.11
2.18
1.03
1.08
1.09
1.50
2.90
0.97
6.30
6.34
2.92
1.15
3.09
4.31
0.81
3.13
6.35
5.40
3.97
3.53
3.72
8.45
7.93
7.92
7.92
7.80
7.71
7.71
4.87
4.87
4.85
4.85
4.38
4.36
4.34
4.29
4.28
4.03
4.03
4.01
4.01
3.53
3.53
3.52
3.51
3.51
3.50
3.40
3.19
3.18
3.17
3.16
3.09
3.08
2.51
2.50
2.49
2.48
2.47
2.47
2.26
2.25
2.11
1.95
1.94
1.93
1.92
1.91
1.91
1.88
1.87
1.85
1.84
1.78
1.74
1.68
1.68
1.67
1.67
1.65
1.65
1.64
1.63
1.63
1.62
1.61
1.49
1.49
1.48
1.47
1.32
1.30
1.28
1.24
1.24
1.22
1.21
1.19
1.19
1.17
1.15
1.14
1.00
0.98
0.86
0.85
0.83
0.82

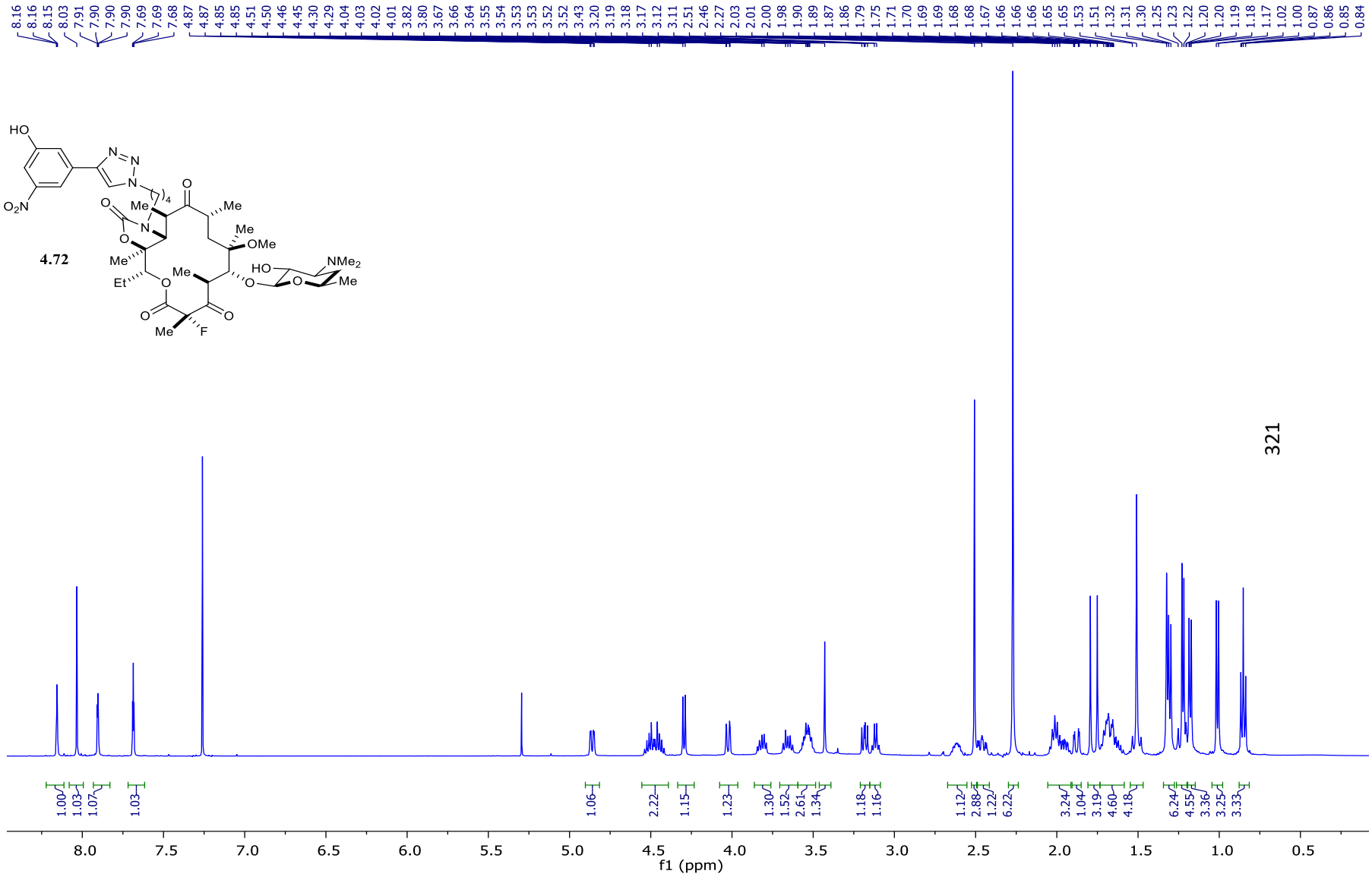

4.69
AcHN
AcHN
f1 (ppm)
216.6
203.6
203.4
169.2
166.5
166.4
157.4
147.3
139.3
131.8
120.5
112.9
111.1
104.4
98.7
97.0
82.3
81.0
78.7
70.5
69.7
65.9
61.3
50.0
49.3
44.7
43.0
41.0
40.3
39.7
39.3
29.8
28.3
27.6
25.4
25.3
24.6
24.4
22.3
21.3
19.9
18.0
15.2
14.8
13.9
10.6

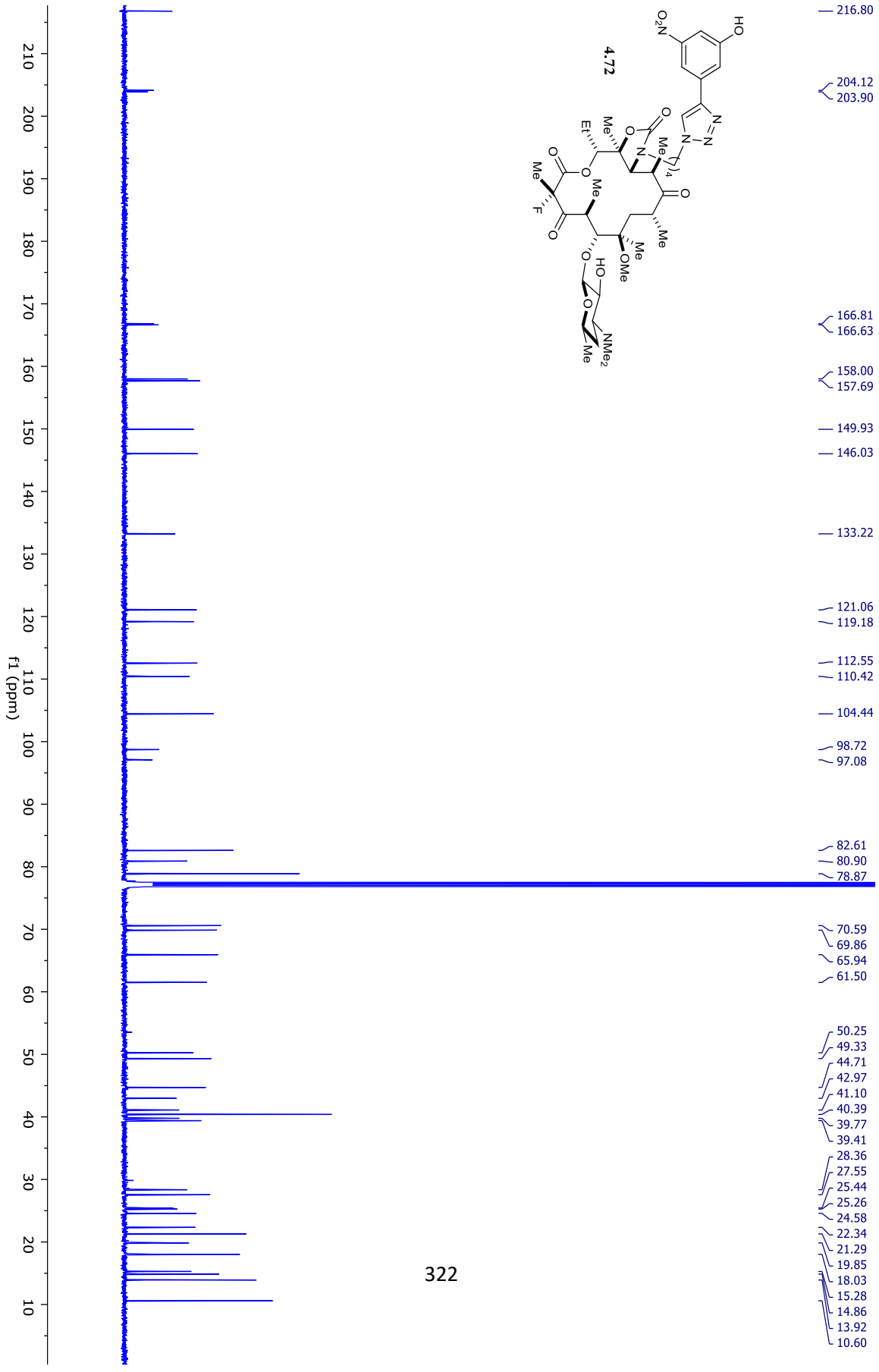

4.70
f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.01
1.07
1.14
0.95
4.88
1.35
1.10
3.40
0.99
1.08
2.35
1.05
1.14
1.02
5.39
7.05
1.10
5.68
1.54
8.58
5.13
20.69
3.70
5.39
8.16
8.02
7.85
7.85
7.68
7.67
7.66
4.86
4.83
4.83
4.47
4.45
4.43
4.41
4.31
4.29
4.05
4.02
3.94
3.73
3.65
3.54
3.53
3.51
3.50
3.41
3.23
3.21
3.21
3.19
3.11
3.09
2.59
2.58
2.55
2.50
2.31
2.28
2.16
2.15
2.01
2.00
1.98
1.96
1.95
1.95
1.93
1.93
1.88
1.87
1.85
1.84
1.78
1.72
1.70
1.68
1.66
1.65
1.64
1.63
1.61
1.61
1.59
1.57
1.55
1.54
1.49
1.47
1.34
1.31
1.30
1.28
1.24
1.22
1.21
1.19
1.18
1.17
1.01
0.99
0.89
0.88
0.87
0.86
0.84
0.81
0.80

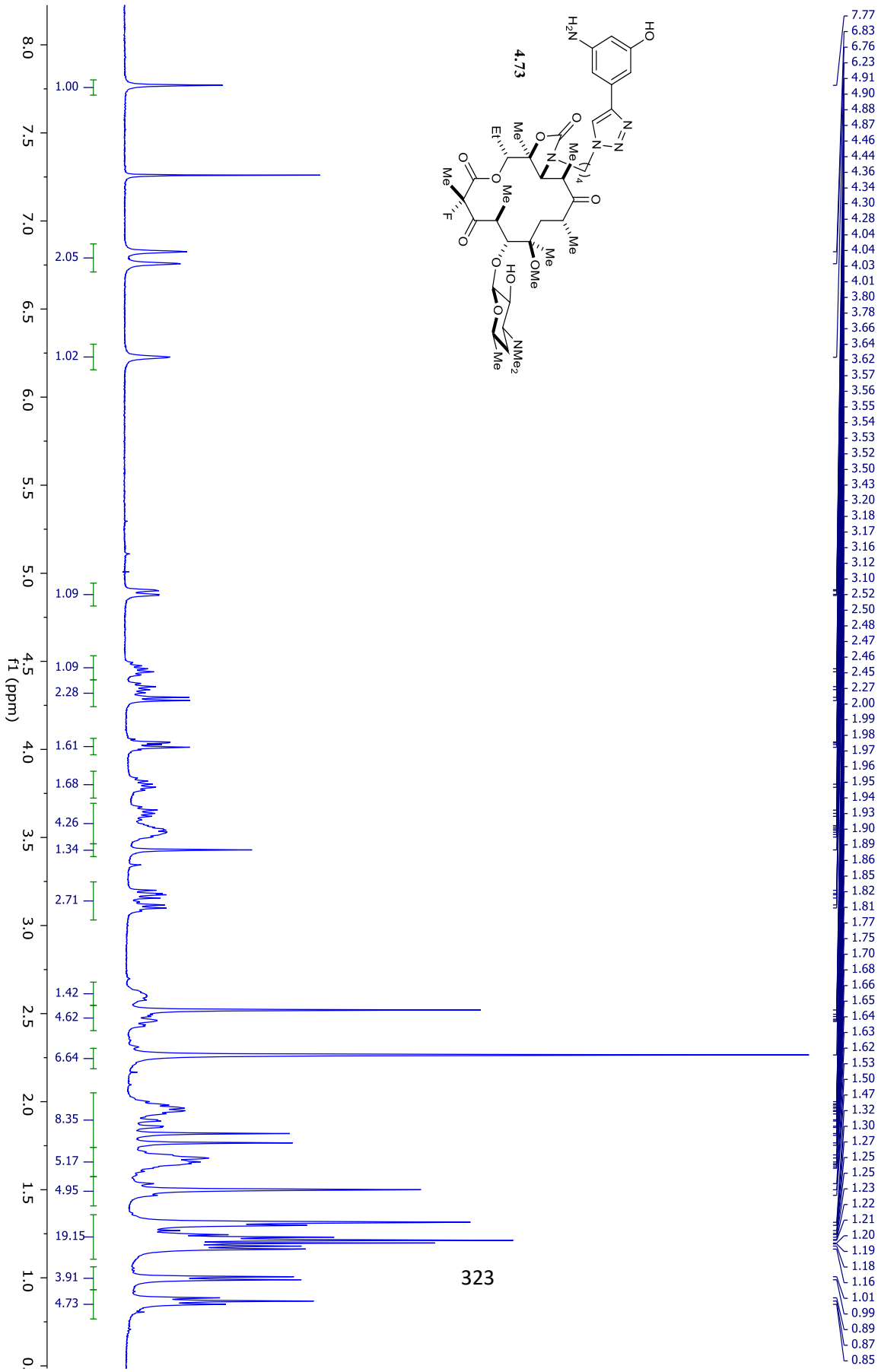

4.70
f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.8
202.6
166.8
160.6
157.3
149.7
145.8
133.4
121.0
117.3
113.0
108.4
104.2
98.9
96.9
82.3
80.7
78.7
70.4
69.5
65.9
61.0
56.2
50.1
49.3
44.7
42.8
41.0
40.2
39.6
39.3
29.8
28.6
27.7
24.3
22.3
21.2
19.9
18.0
15.2
14.8
13.9

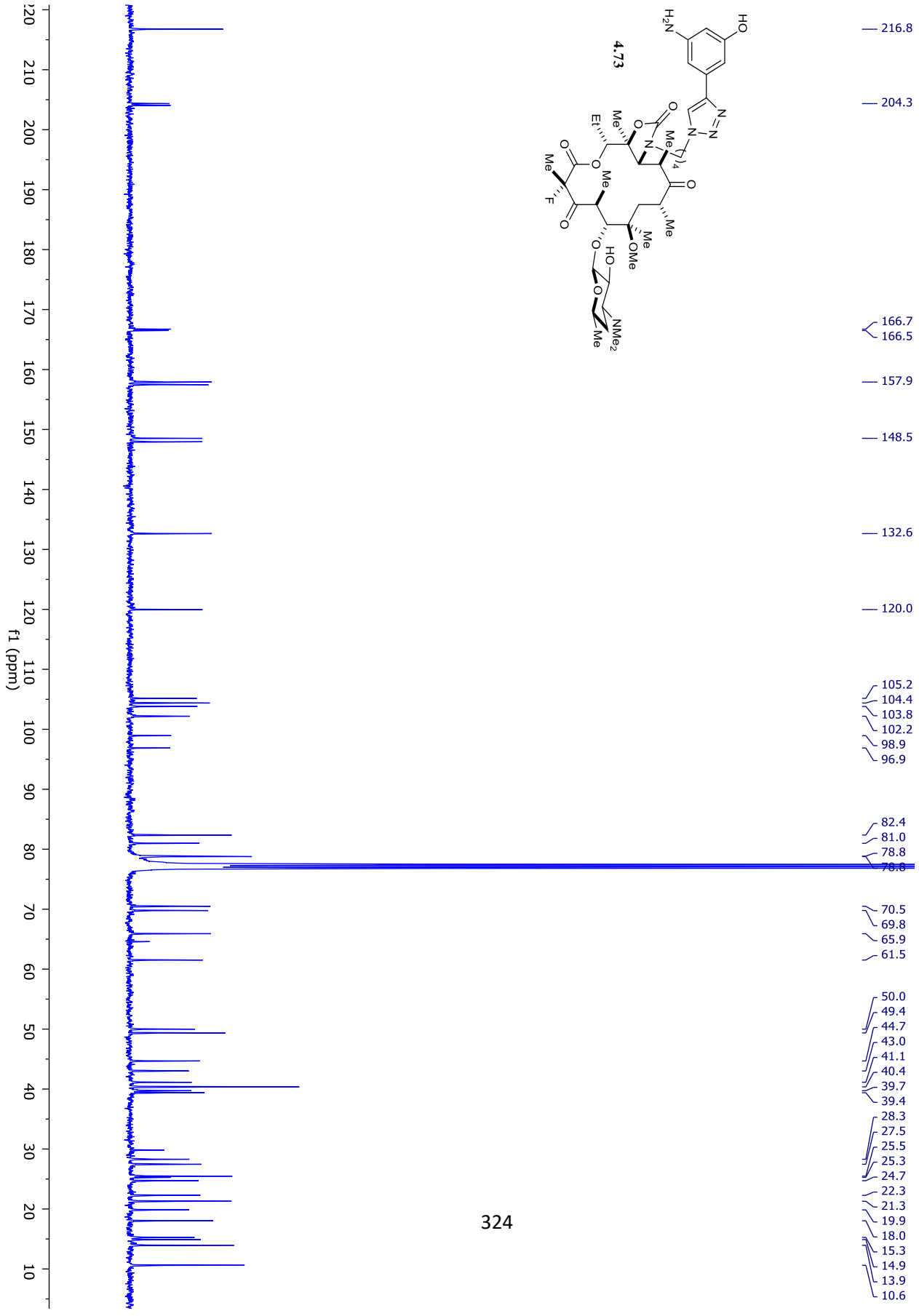

4.71
f1 (ppm)

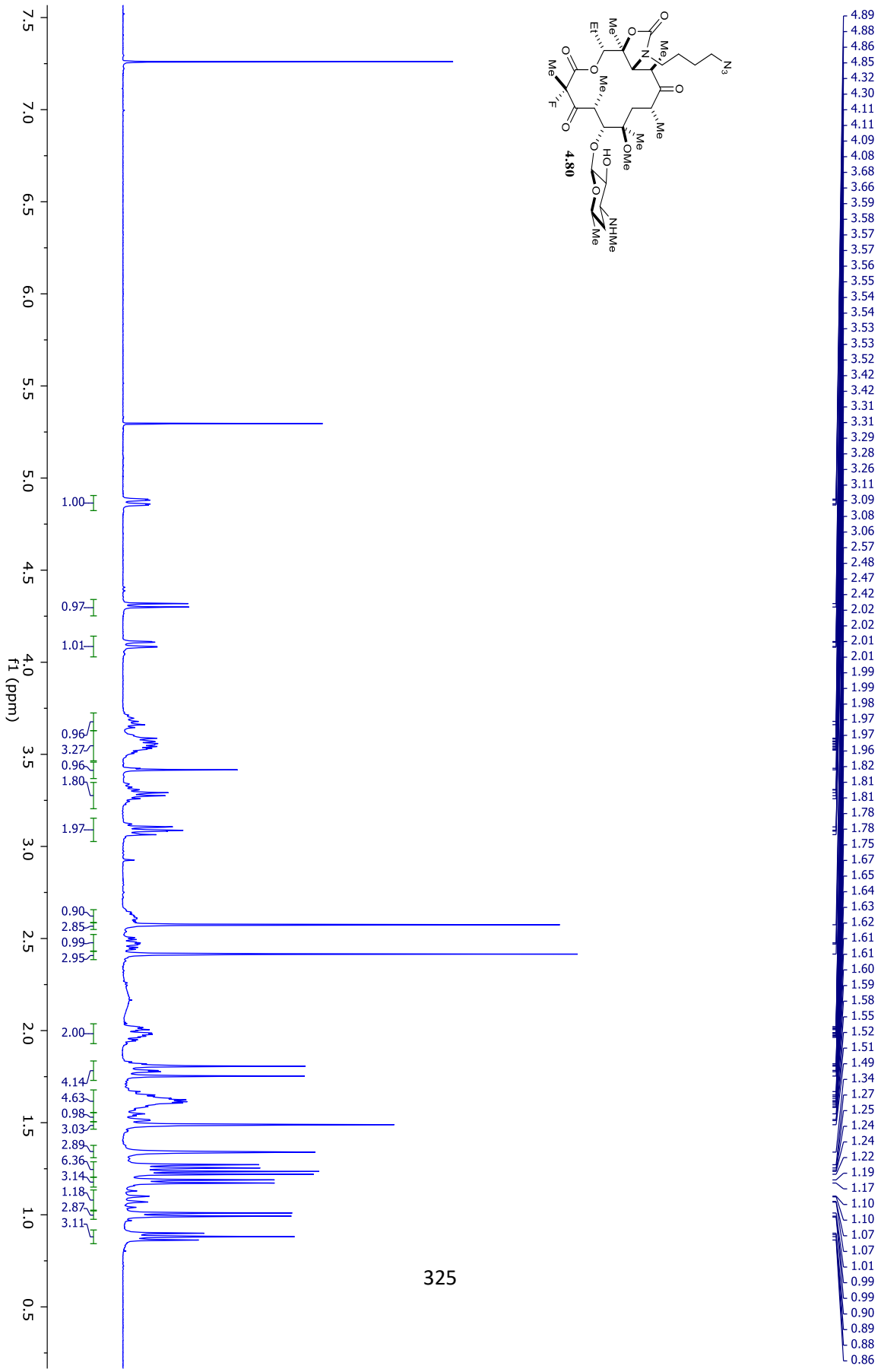

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
202.9
166.5
161.2
157.4
148.2
132.8
119.9
105.8
104.4
101.4
101.3
99.0
96.9
82.3
80.9
78.7
70.5
69.8
66.0
61.2
55.4
49.9
49.4
44.7
42.9
41.0
40.4
39.7
39.4
29.8
28.3
27.7
25.5
25.3
24.4
22.3
21.3
19.9
18.0
15.2
14.9
13.9
10.6
4.71
MeO
H2N
NMe2
OMe
HO
Me
Et
F


8.16 8.16 8.15 8.03 7.91 7.90 7.90 7.90 7.69 7.69 7.68 4.87 4.87 4.85 4.85 4.51 4.50 4.46 4.45 4.30 4.29 4.04 4.03 4.02 4.01 3.82 3.80 3.67 3.66 3.64 3.55 3.54 3.53 3.53 3.52 3.52 3.43 3.20 3.19 3.18 3.17 3.12 3.11 2.51 2.46 2.27 2.03 2.01 2.00 1.98 1.90 1.89 1.87 1.86 1.79 1.75 1.71 1.70 1.69 1.69 1.68 1.68 1.67 1.66 1.66 1.66 1.65 1.65 1.53 1.51 1.32 1.31 1.30 1.25 1.23 1.22 1.20 1.20 1.19 1.18 1.17 1.02 1.00 0.87 0.86 0.85 0.84
HO
O2N
N
N
N
4
O
O
Me
N
O
Me
Me
Me
OMe
Me
Et
O
Me
HO
NMe2
O
O
Me
O
O
Me
F
4.72
321
1.00 1.03 1.07 1.03 1.06 2.22 1.15 1.23 1.30 1.52 2.61 1.34 1.18 1.16 1.12 2.88 1.22 6.22 3.24 1.04 3.19 4.60 4.18 6.24 4.55 3.36 3.25 3.33
8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5
f1 (ppm)

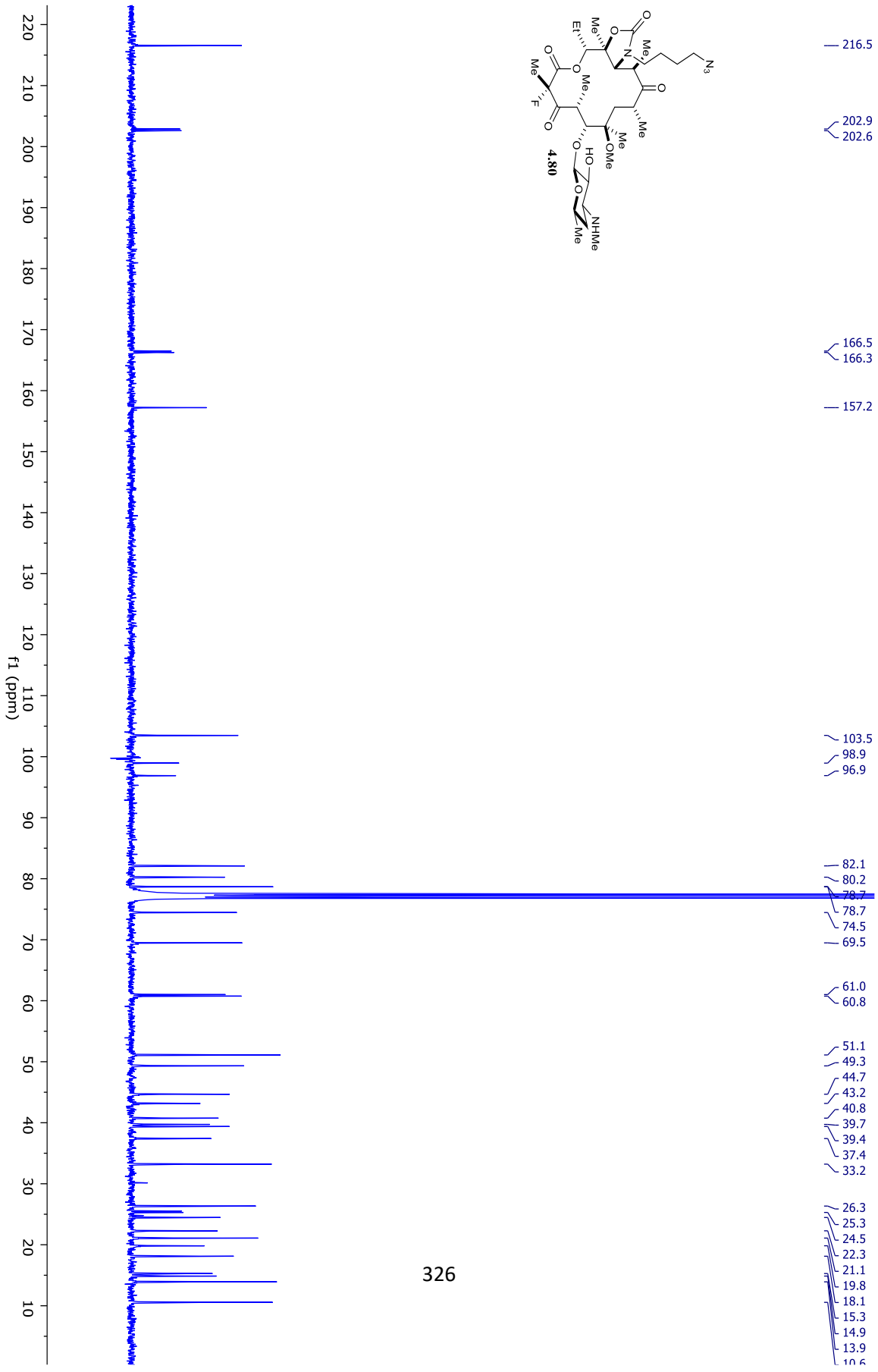

4.72
f1 (ppm)
216.80
204.12
203.90
166.81
166.63
158.00
157.69
149.93
146.03
133.22
121.06
119.18
112.55
110.42
104.44
98.72
97.08
82.61
80.90
78.87
70.59
69.86
65.94
61.50
50.25
49.33
44.71
42.97
41.10
40.39
39.77
39.41
28.36
27.55
25.44
25.26
24.58
22.34
21.29
19.85
18.03
15.28
14.86
13.92
10.60

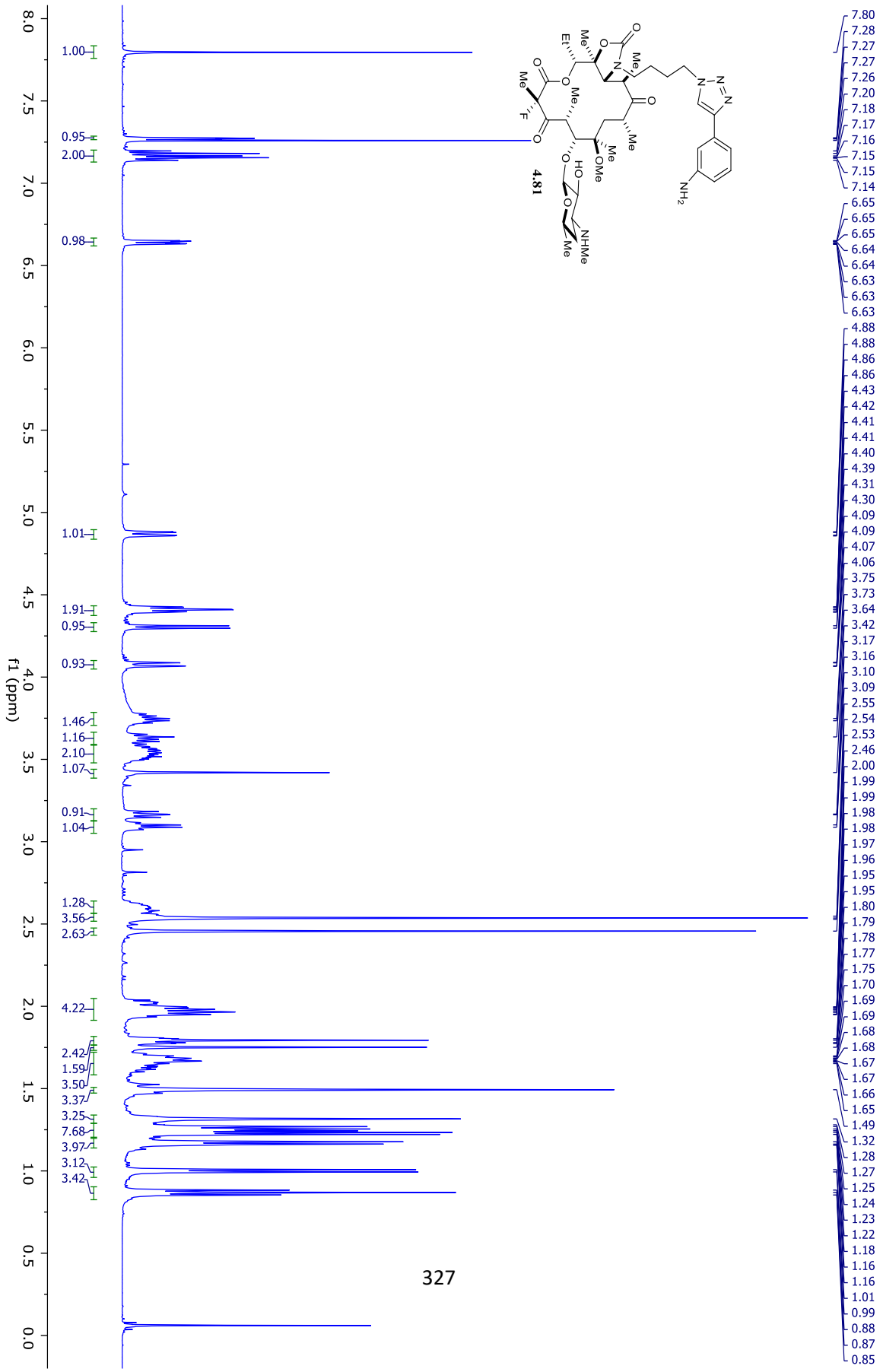

4.73
f1 (ppm)
1.00
2.05
1.02
1.09
1.09
2.28
1.61
1.68
4.26
1.34
2.71
1.42
4.62
6.64
8.35
5.17
4.95
19.15
3.91
4.73

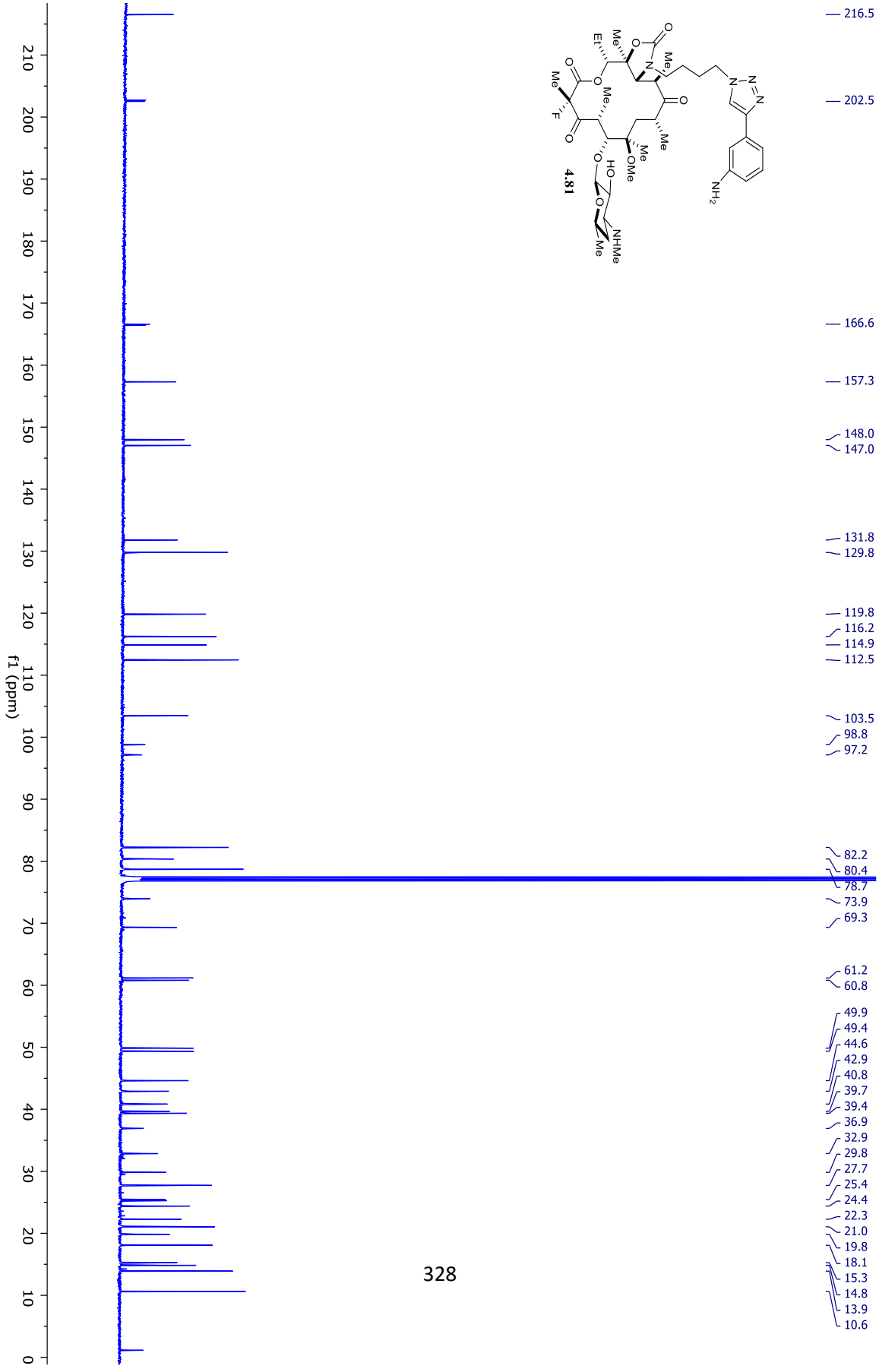

4.73
216.8
204.3
166.7
166.5
157.9
148.5
132.6
120.0
105.2
104.4
103.8
102.2
98.9
96.9
82.4
81.0
78.8
78.6
70.5
69.8
65.9
61.5
50.0
49.4
44.7
43.0
41.1
40.4
39.7
39.4
28.3
27.5
25.5
25.3
24.7
22.3
21.3
19.9
18.0
15.3
14.9
13.9
10.6
f1 (ppm)

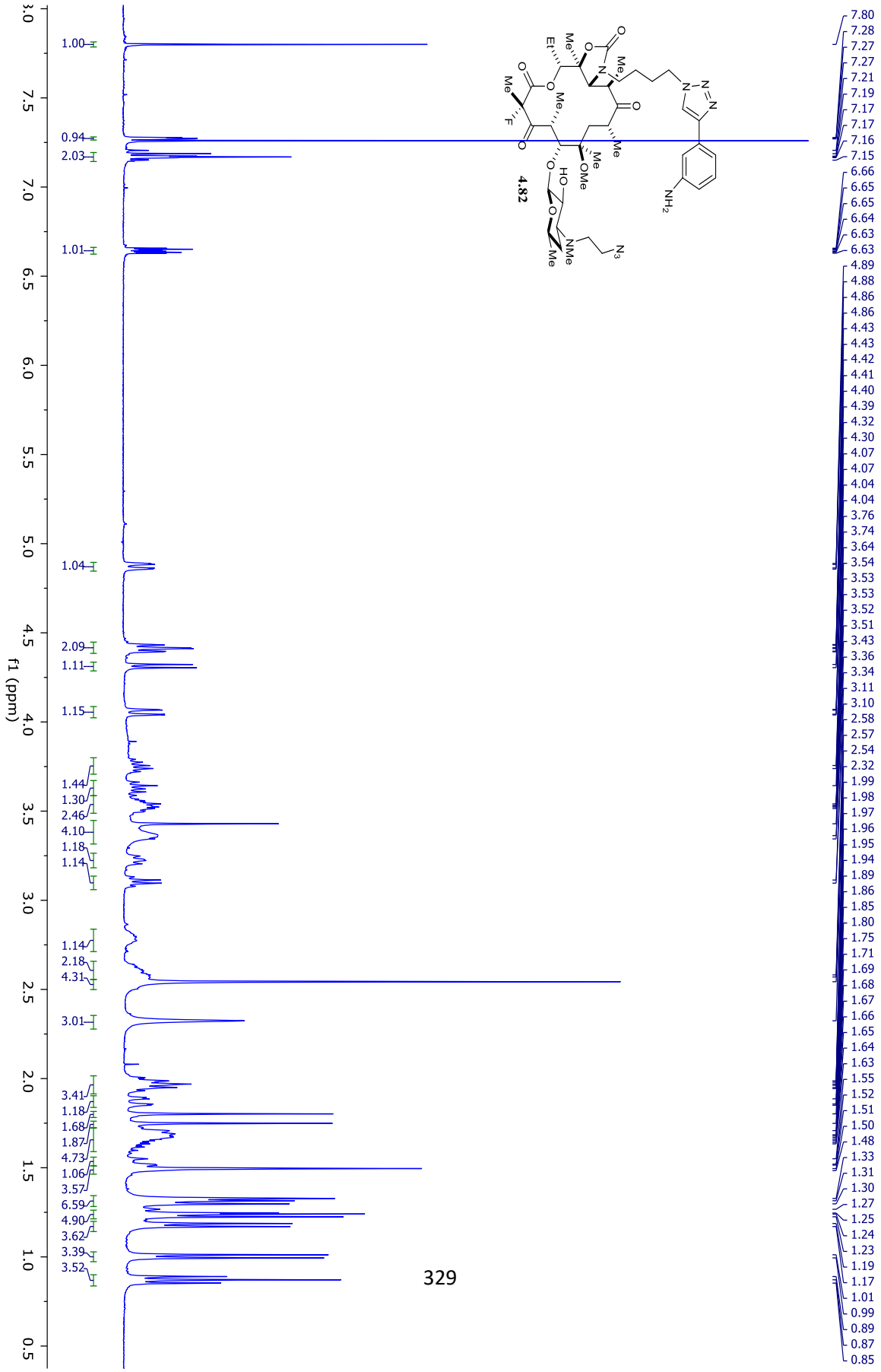

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
0.97
1.01
0.96
3.27
0.96
1.80
1.97
0.90
2.85
0.99
2.95
2.00
4.14
4.63
0.98
3.03
2.89
6.36
3.14
1.18
2.87
3.11
4.89
4.88
4.86
4.85
4.32
4.30
4.11
4.11
4.09
4.08
3.68
3.66
3.59
3.58
3.57
3.57
3.56
3.55
3.54
3.54
3.53
3.53
3.52
3.42
3.42
3.31
3.31
3.29
3.28
3.26
3.11
3.09
3.08
3.06
2.57
2.48
2.47
2.42
2.02
2.02
2.01
2.01
1.99
1.99
1.98
1.97
1.97
1.96
1.82
1.81
1.81
1.78
1.78
1.75
1.67
1.65
1.64
1.63
1.62
1.61
1.61
1.60
1.59
1.58
1.55
1.52
1.51
1.49
1.34
1.27
1.25
1.24
1.24
1.22
1.19
1.17
1.10
1.10
1.07
1.07
1.01
0.99
0.99
0.90
0.89
0.88
0.86

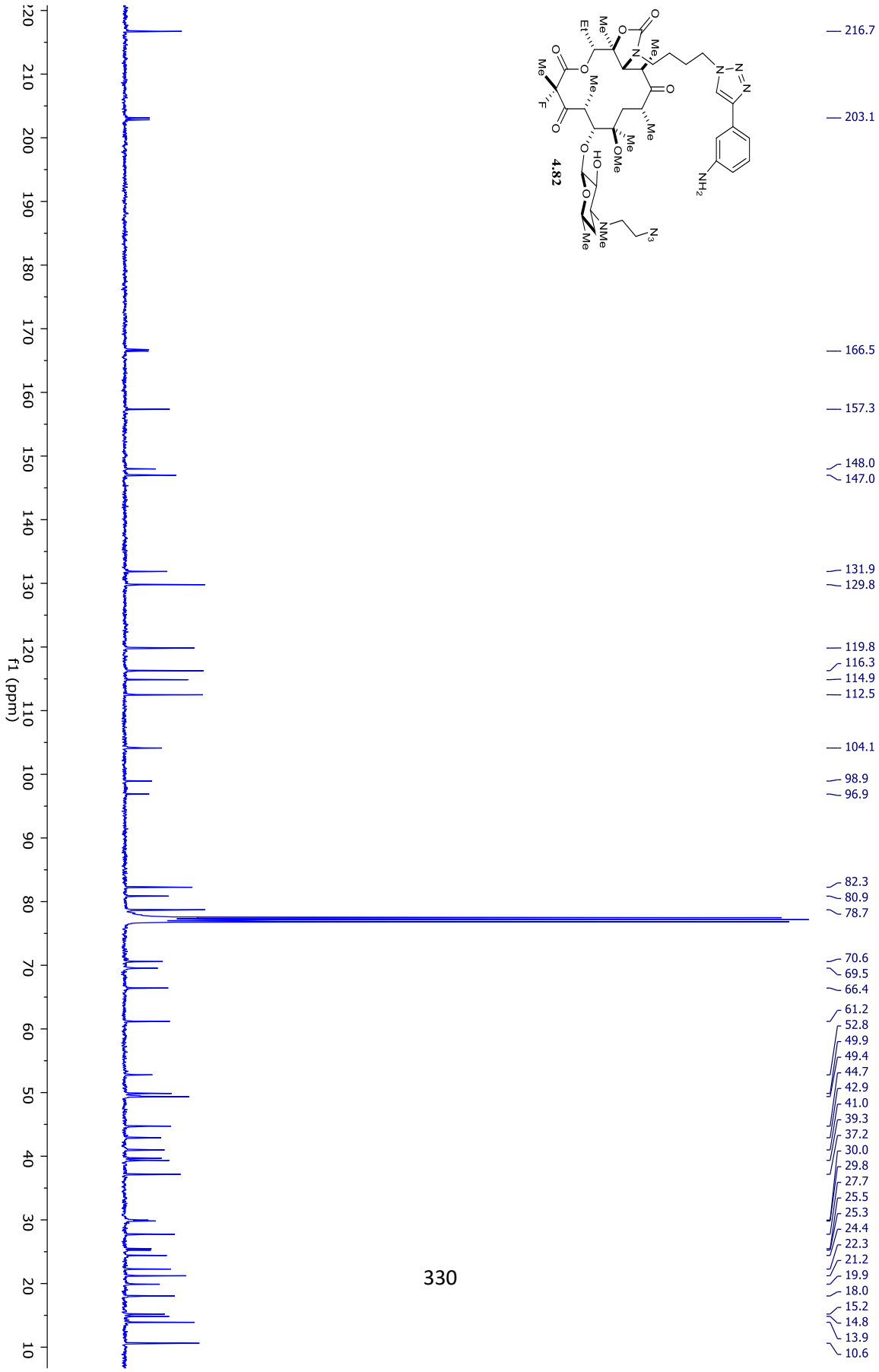
4.80

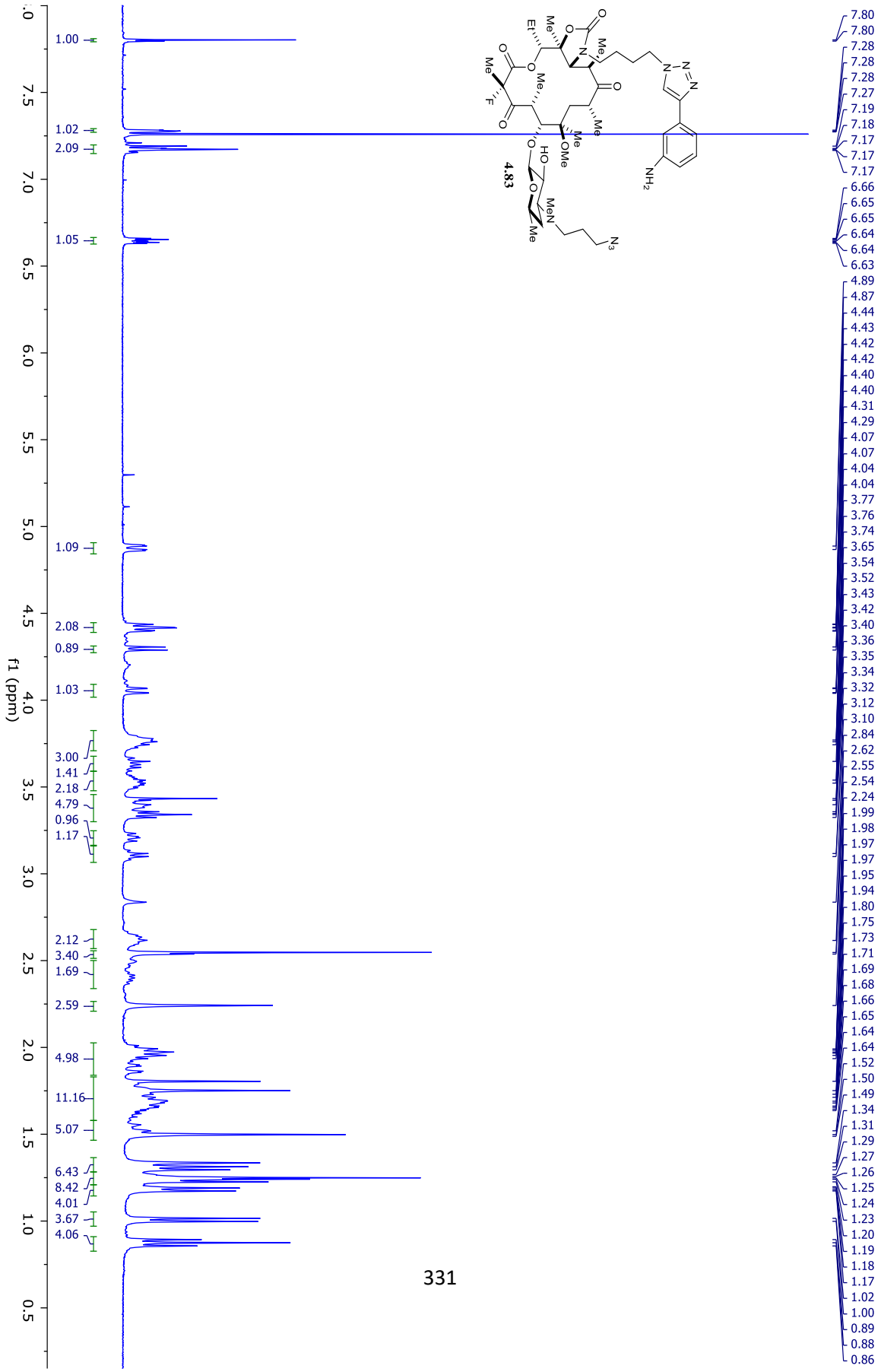

f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.5
202.9
202.6
166.5
166.3
157.2
103.5
98.9
96.9
82.1
80.2
78.7
74.5
69.5
61.0
60.8
51.1
49.3
44.7
43.2
40.8
39.7
39.4
37.4
33.2
26.3
25.3
24.5
22.3
21.1
19.8
18.1
15.3
14.9
13.9
4.80

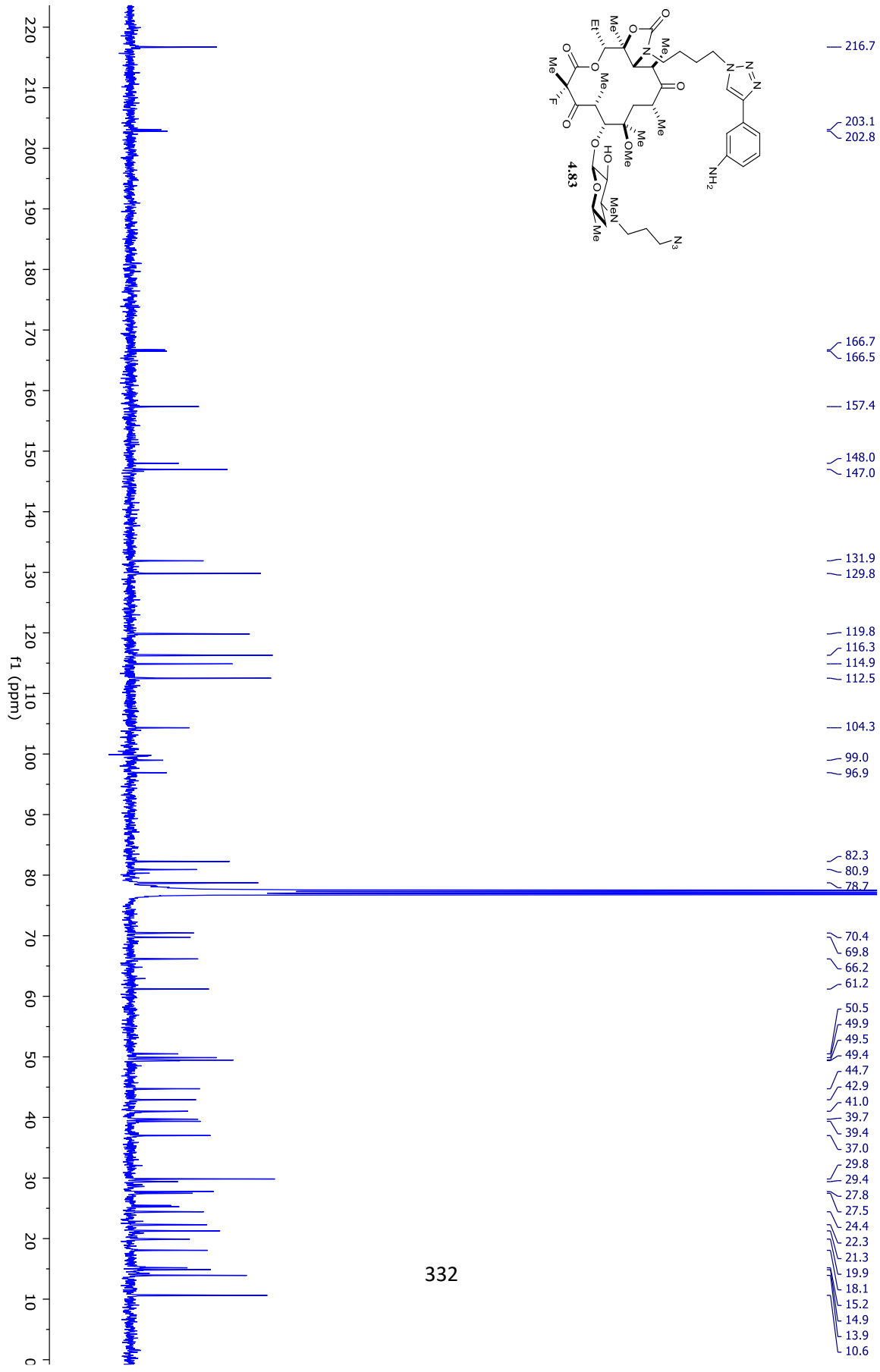

f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
1.00
0.95
2.00
0.98
1.01
1.91
0.95
0.93
1.46
1.16
2.10
1.07
0.91
1.04
1.28
3.56
2.63
4.22
2.42
1.59
3.50
3.37
3.25
7.68
3.97
3.12
3.42
4.81
7.80
7.28
7.27
7.27
7.26
7.20
7.18
7.17
7.16
7.15
7.15
7.14
6.65
6.65
6.65
6.64
6.64
6.63
6.63
6.63
4.88
4.88
4.86
4.86
4.43
4.42
4.41
4.41
4.40
4.39
4.31
4.30
4.09
4.09
4.07
4.06
3.75
3.73
3.64
3.42
3.17
3.16
3.10
3.09
2.55
2.54
2.53
2.46
2.00
1.99
1.99
1.98
1.98
1.97
1.96
1.95
1.95
1.80
1.79
1.78
1.77
1.75
1.70
1.69
1.69
1.68
1.68
1.67
1.67
1.66
1.65
1.49
1.32
1.28
1.27
1.25
1.24
1.23
1.22
1.18
1.16
1.16
1.01
0.99
0.88
0.87
0.85

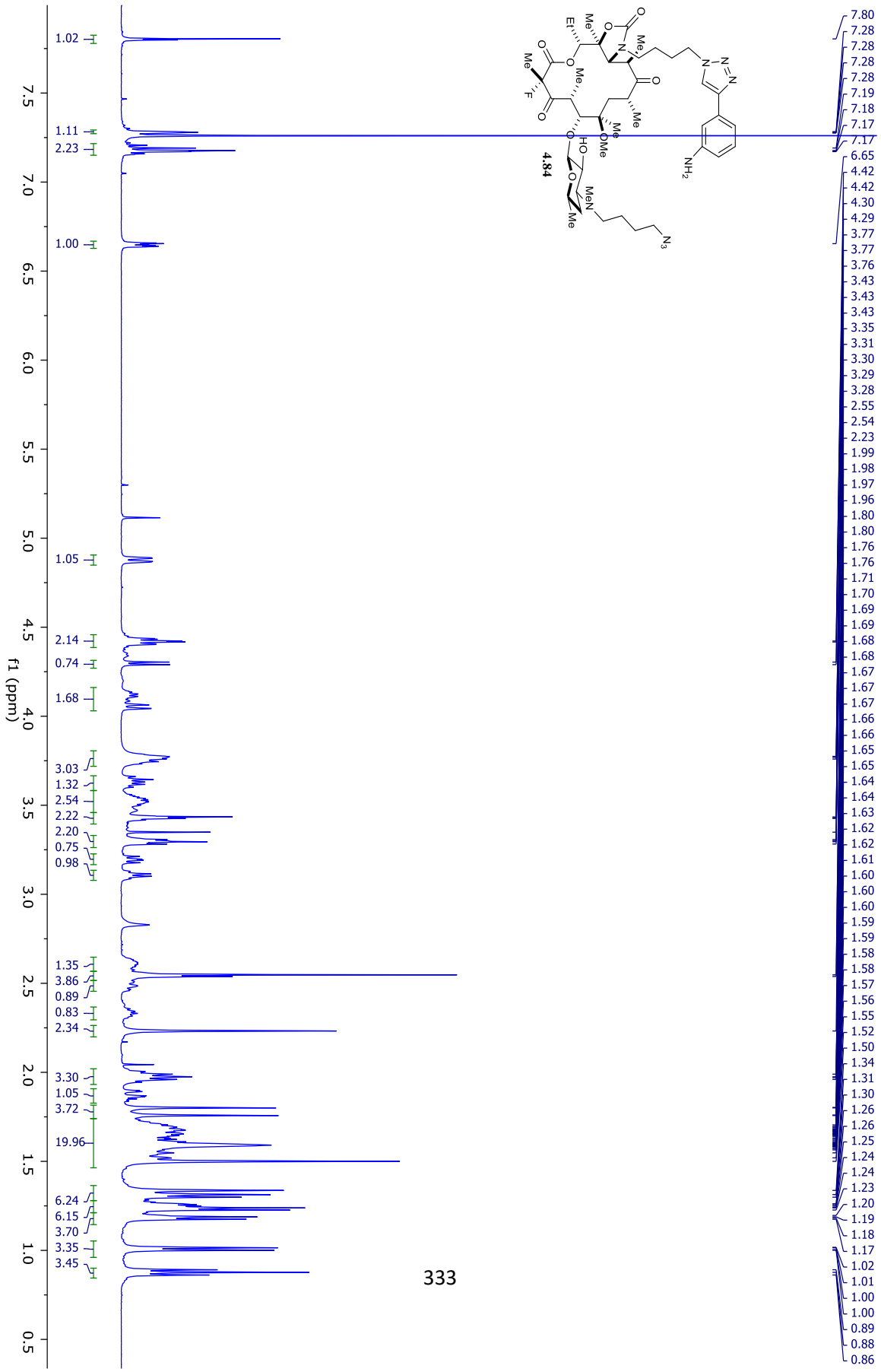

f1 (ppm)
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140
150
160
170
180
190
200
210
216.5
202.5
166.6
157.3
148.0
147.0
131.8
129.8
119.8
116.2
114.9
112.5
103.5
98.8
97.2
82.2
80.4
78.7
73.9
69.3
61.2
60.8
49.9
49.4
44.6
42.9
40.8
39.7
39.4
36.9
32.9
29.8
27.7
25.4
24.4
22.3
21.0
19.8
18.1
15.3
14.8
13.9
10.6
4.81

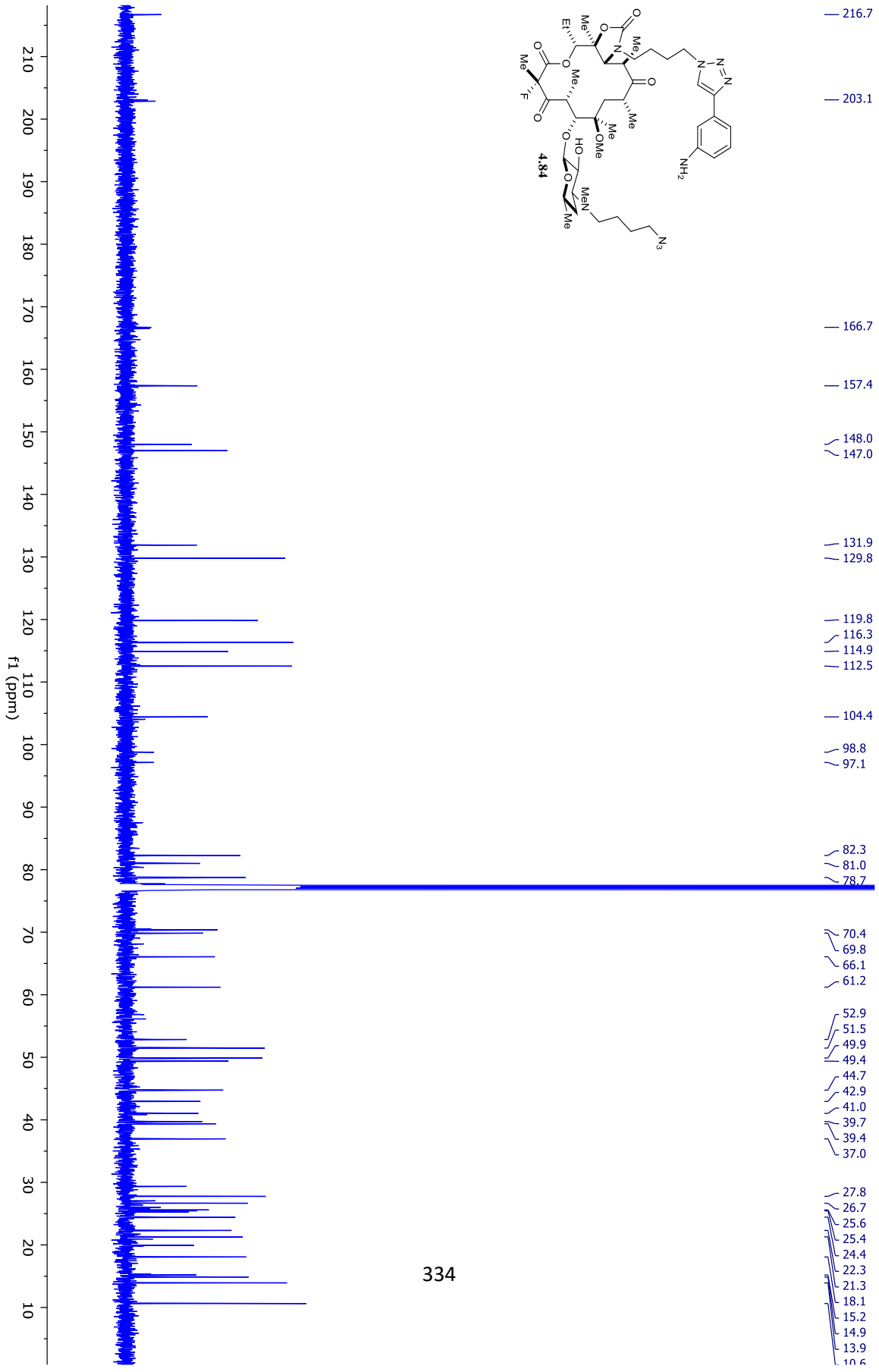

f1 (ppm)
4.82

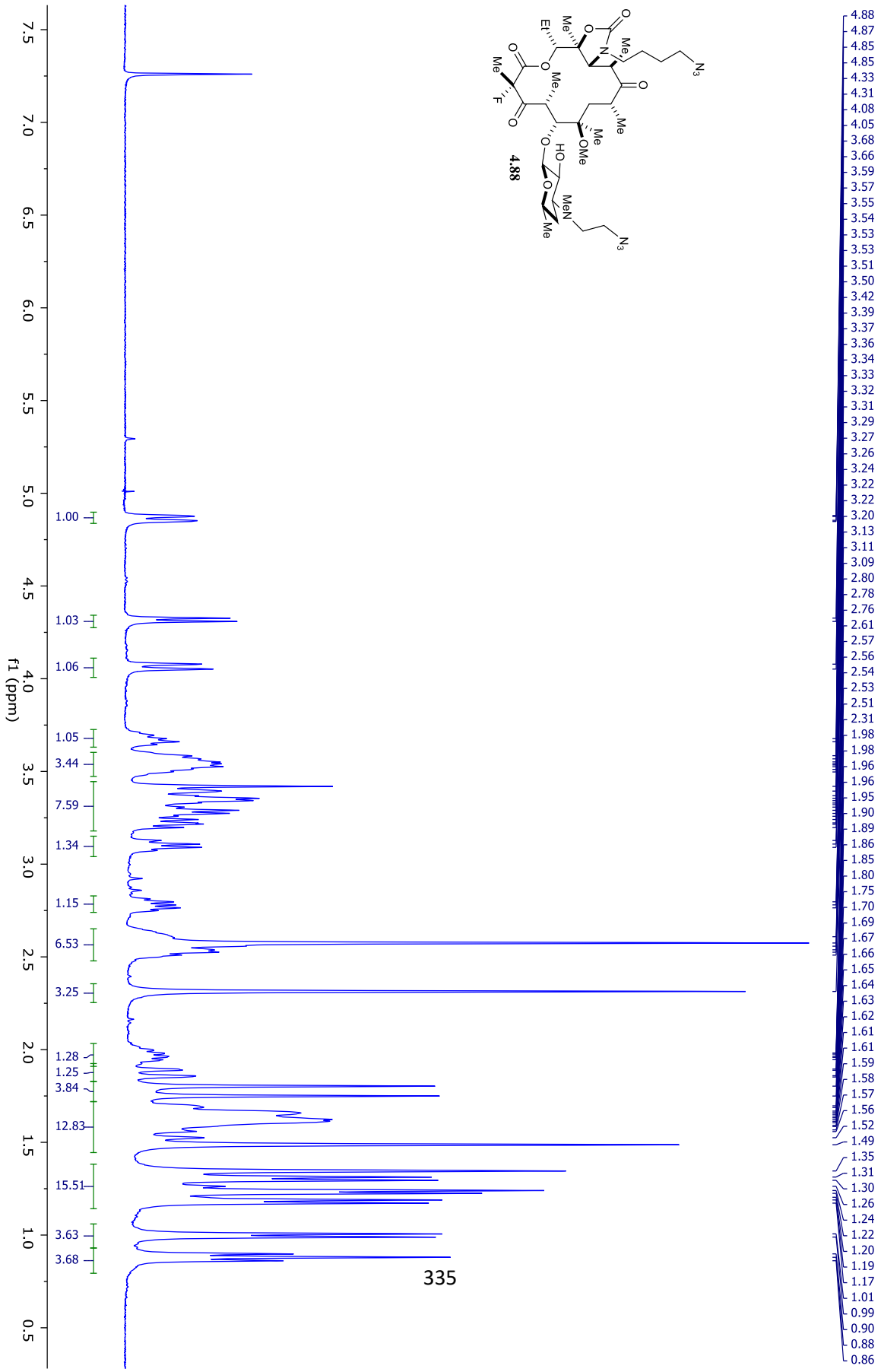

4.82
216.7
203.1
166.5
157.3
148.0
147.0
131.9
129.8
119.8
116.3
114.9
112.5
104.1
98.9
96.9
82.3
80.9
78.7
70.6
69.5
66.4
61.2
52.8
49.9
49.4
44.7
42.9
41.0
39.3
37.2
30.0
29.8
27.7
25.5
25.3
24.4
22.3
21.2
19.9
18.0
15.2
14.8
13.9
10.6
f1 (ppm)

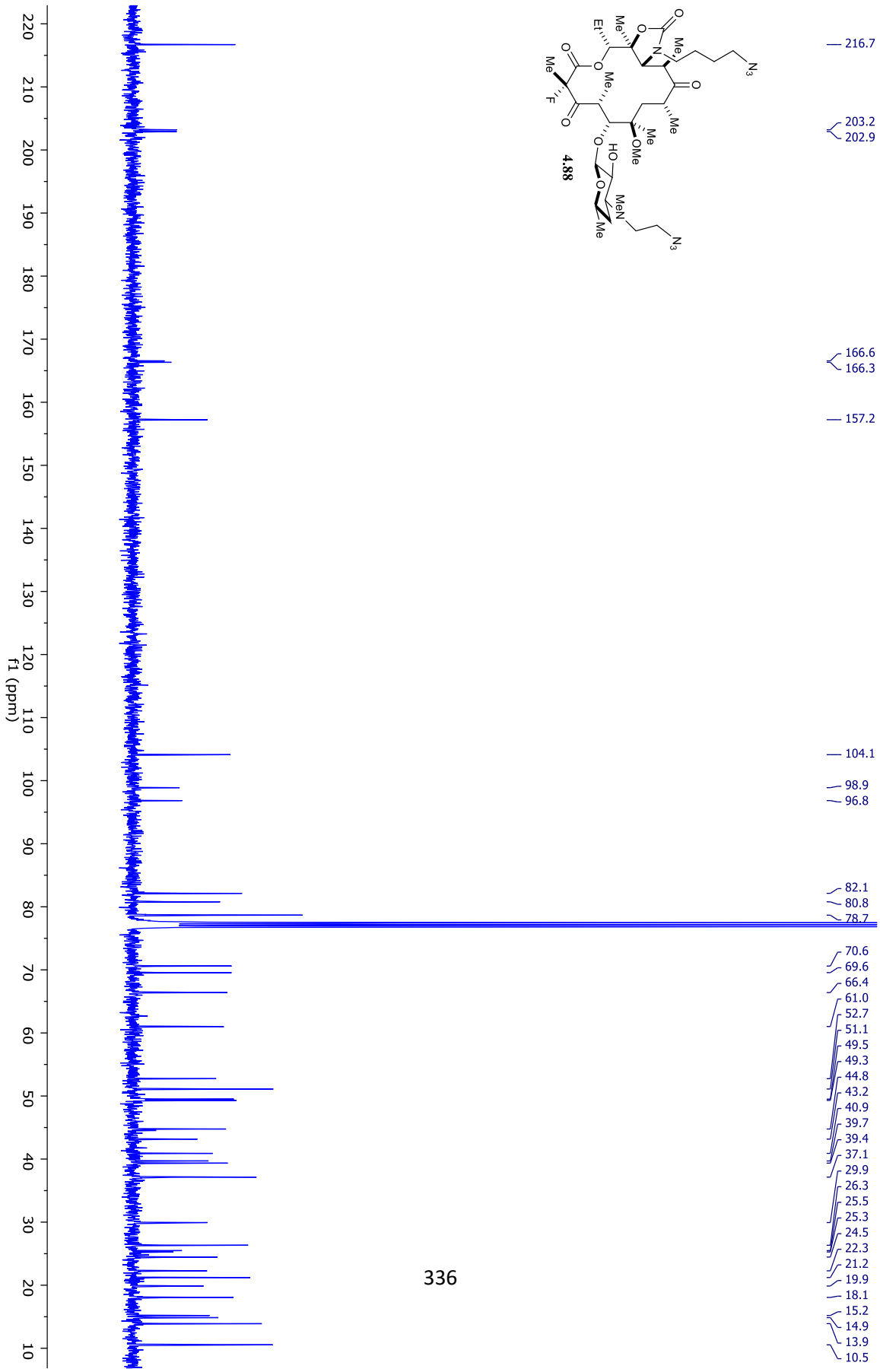

f1 (ppm)
1.00
1.02
2.09
1.05
1.09
2.08
0.89
1.03
3.00
1.41
2.18
4.79
0.96
1.17
2.12
3.40
1.69
2.59
4.98
11.16
5.07
6.43
8.42
4.01
3.67
4.06
4.83
7.80
7.28
7.27
7.19
7.18
7.17
6.66
6.65
6.64
6.63
4.89
4.87
4.44
4.43
4.42
4.40
4.31
4.29
4.07
4.04
3.77
3.76
3.74
3.65
3.54
3.52
3.43
3.42
3.40
3.36
3.35
3.34
3.32
3.12
3.10
2.84
2.62
2.55
2.54
2.24
1.99
1.98
1.97
1.95
1.94
1.80
1.75
1.73
1.71
1.69
1.68
1.66
1.65
1.64
1.52
1.50
1.49
1.34
1.31
1.29
1.27
1.26
1.25
1.24
1.23
1.20
1.19
1.18
1.17
1.02
1.00
0.89
0.88
0.86

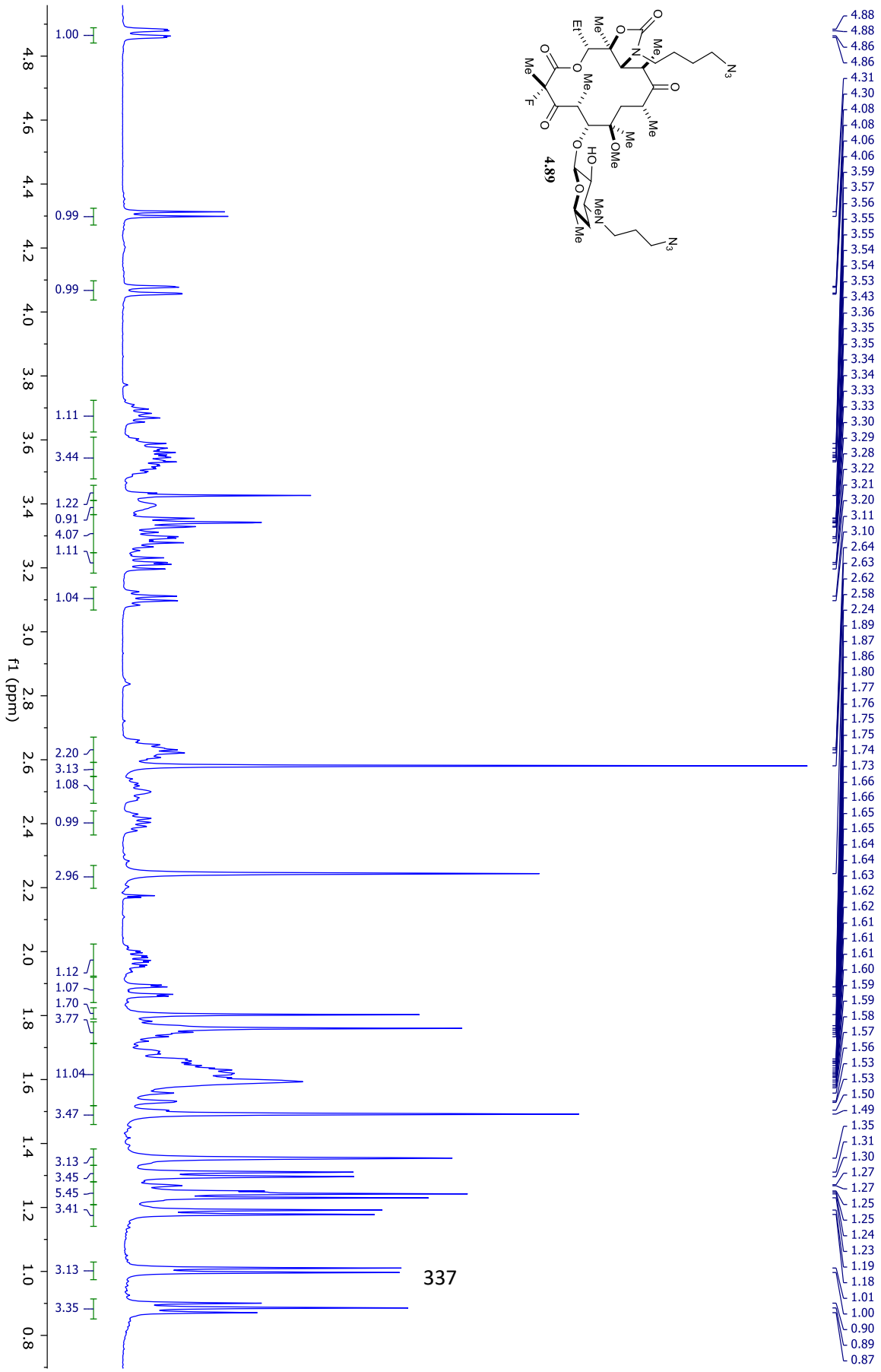

f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
216.7
203.1
202.8
166.7
166.5
157.4
148.0
147.0
131.9
129.8
119.8
116.3
114.9
112.5
104.3
99.0
96.9
82.3
80.9
78.7
70.4
69.8
66.2
61.2
50.5
49.9
49.5
49.4
44.7
42.9
41.0
39.7
39.4
37.0
29.8
29.4
27.8
27.5
24.4
22.3
21.3
19.9
18.1
15.2
14.9
13.9
10.6
4.83

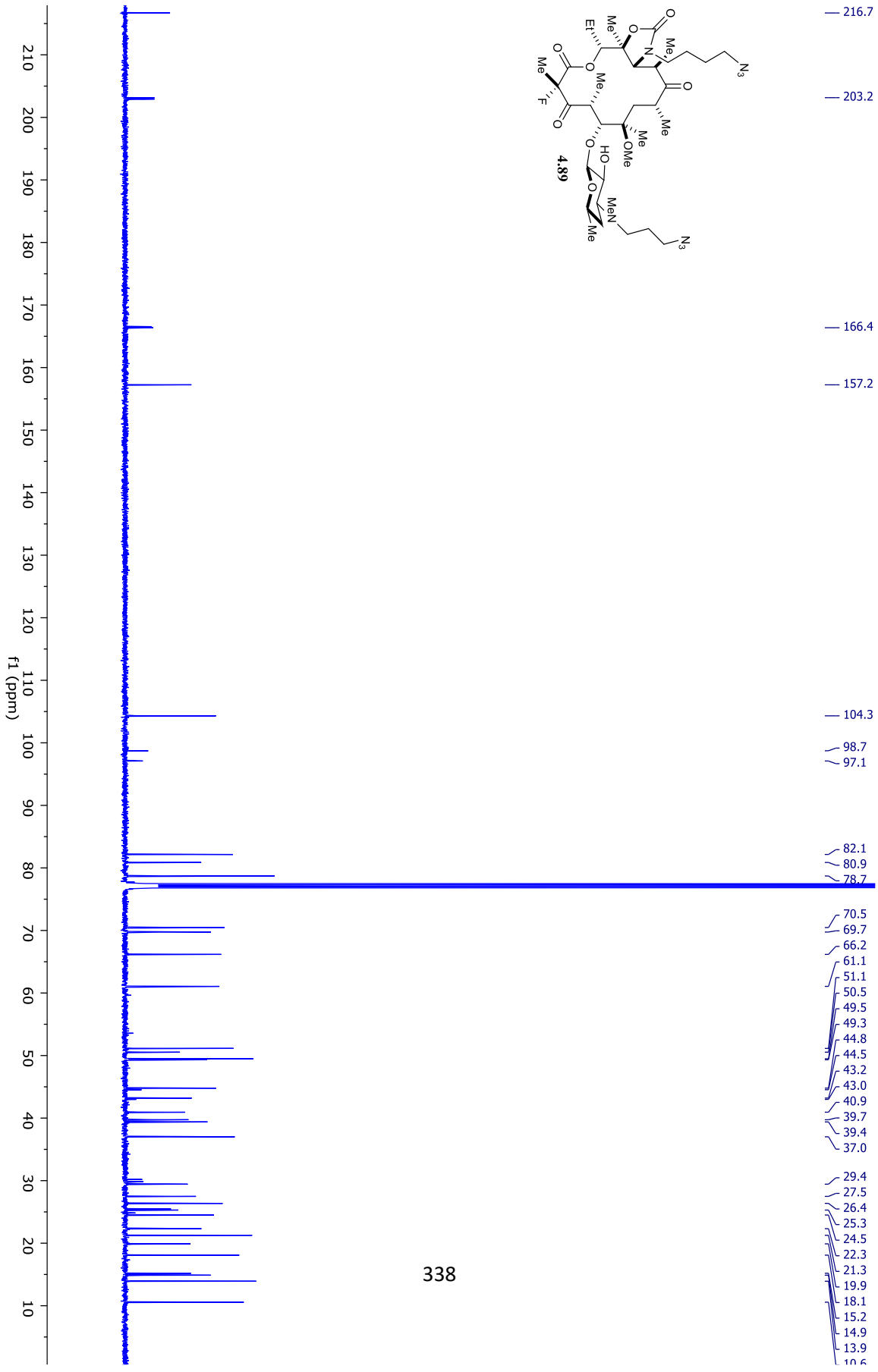

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.02
1.11
2.23
1.00
1.05
2.14
0.74
1.68
3.03
1.32
2.54
2.22
2.20
0.75
0.98
1.35
3.86
0.89
0.83
2.34
3.30
1.05
3.72
19.96
6.24
6.15
3.70
3.35
3.45
4.84
7.80
7.28
7.28
7.28
7.28
7.19
7.18
7.17
7.17
6.65
4.42
4.42
4.30
4.29
3.77
3.77
3.76
3.43
3.43
3.43
3.35
3.31
3.30
3.29
3.28
2.55
2.54
2.23
1.99
1.98
1.97
1.96
1.80
1.80
1.76
1.76
1.71
1.70
1.69
1.69
1.68
1.68
1.67
1.67
1.67
1.67
1.66
1.66
1.65
1.65
1.64
1.64
1.63
1.62
1.62
1.61
1.60
1.60
1.60
1.59
1.59
1.58
1.58
1.57
1.56
1.55
1.52
1.50
1.34
1.31
1.30
1.26
1.26
1.25
1.24
1.24
1.23
1.20
1.19
1.18
1.17
1.02
1.01
1.00
1.00
0.89
0.88
0.86

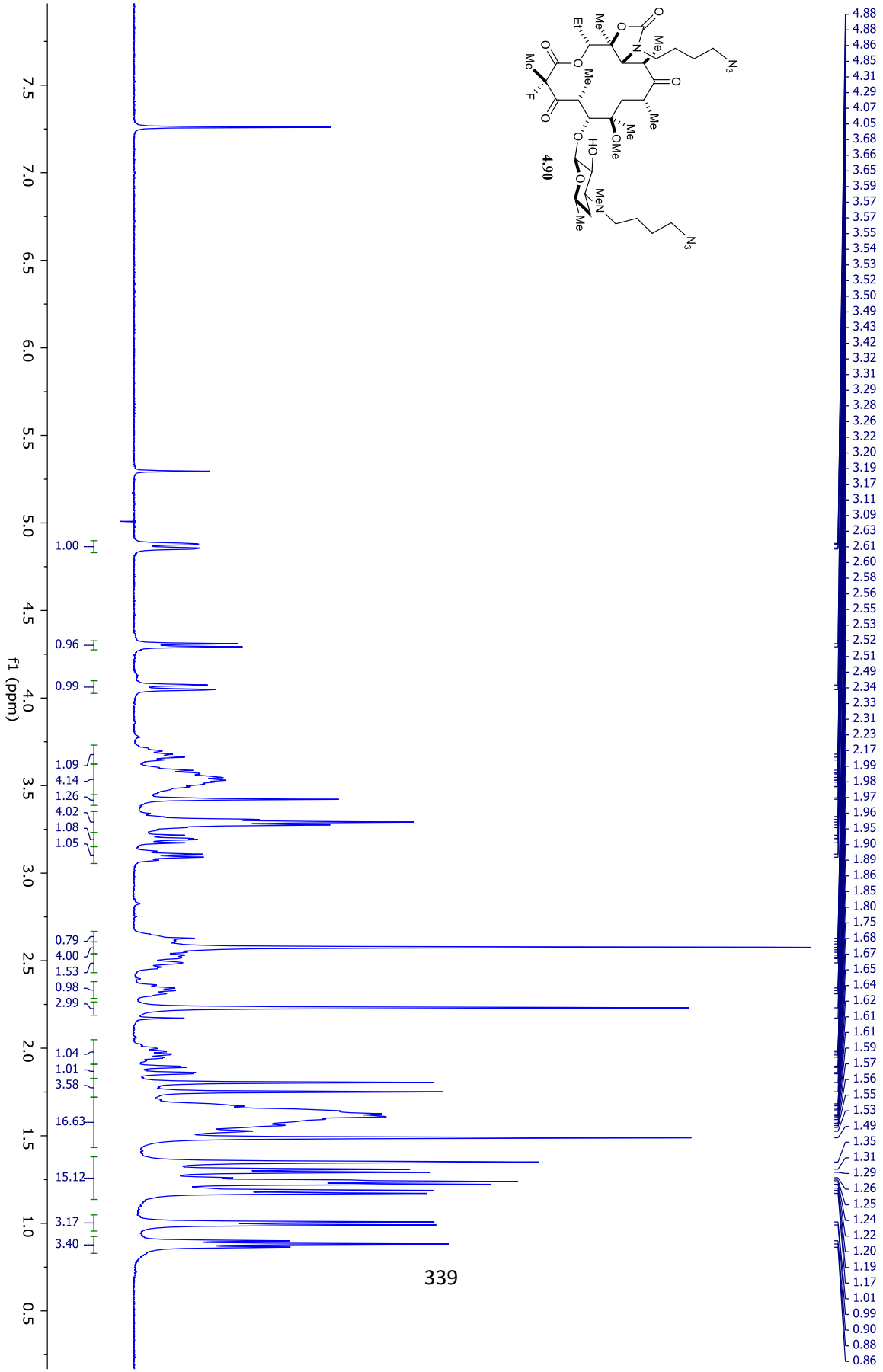

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
203.1
166.7
157.4
148.0
147.0
131.9
129.8
119.8
116.3
114.9
112.5
104.4
98.8
97.1
82.3
81.0
78.7
70.4
69.8
66.1
61.2
52.9
51.5
49.9
49.4
44.7
42.9
41.0
39.7
39.4
37.0
27.8
26.7
25.6
25.4
24.4
22.3
21.3
18.1
15.2
14.9
13.9
10.6
4.84

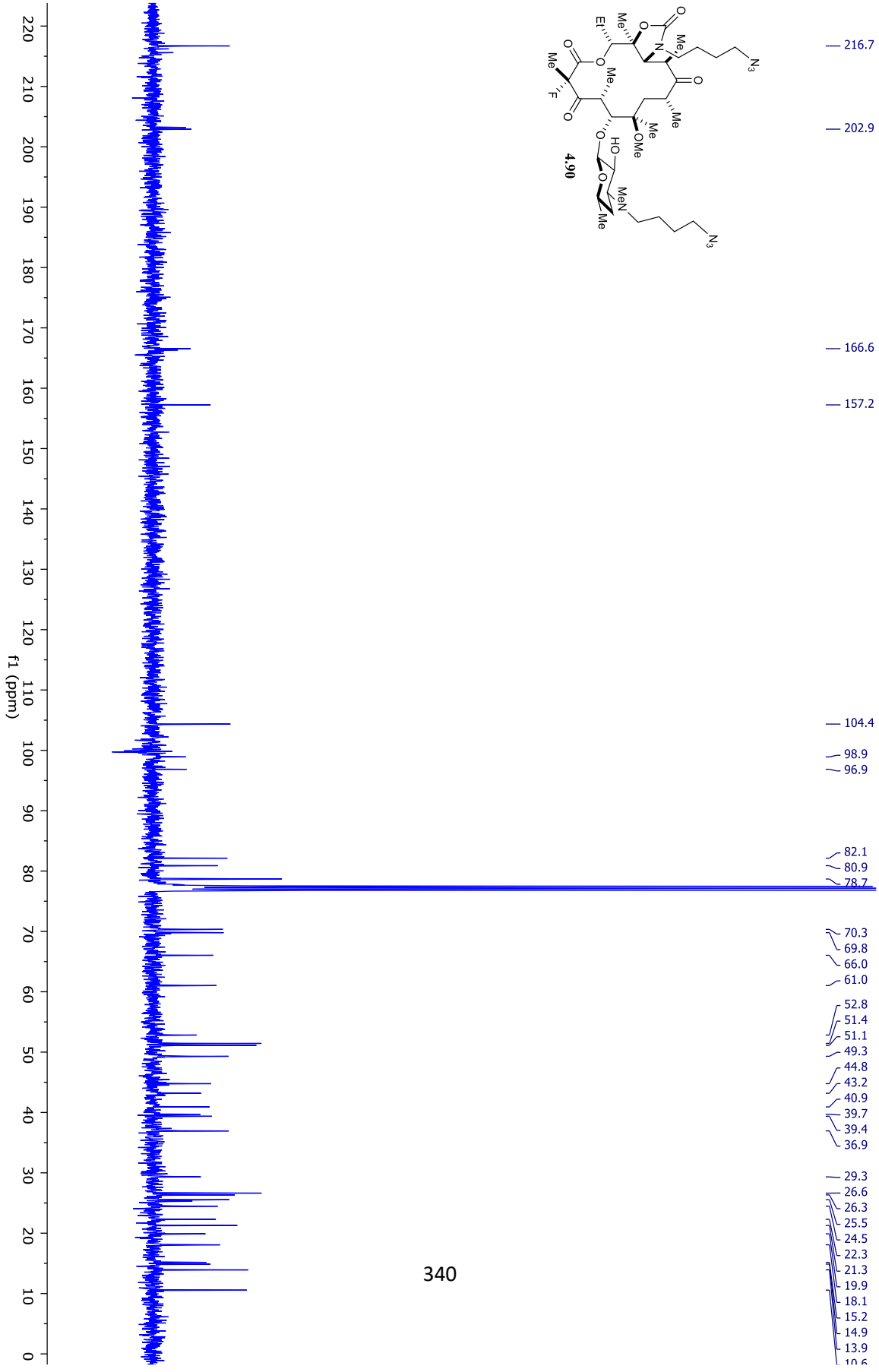

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.03
1.06
1.05
3.44
7.59
1.34
1.15
6.53
3.25
1.28
1.25
3.84
12.83
15.51
3.63
3.68
4.88
4.87
4.85
4.85
4.33
4.31
4.08
4.05
3.68
3.66
3.59
3.57
3.55
3.54
3.53
3.53
3.51
3.50
3.42
3.39
3.37
3.36
3.34
3.33
3.32
3.31
3.29
3.27
3.26
3.24
3.22
3.22
3.20
3.13
3.11
3.09
2.80
2.78
2.76
2.61
2.57
2.56
2.54
2.53
2.51
2.31
1.98
1.98
1.96
1.96
1.95
1.90
1.89
1.86
1.85
1.80
1.75
1.70
1.69
1.67
1.66
1.65
1.64
1.63
1.62
1.61
1.61
1.59
1.58
1.57
1.56
1.52
1.49
1.35
1.31
1.30
1.26
1.24
1.22
1.20
1.19
1.17
1.01
0.99
0.90
0.88
0.86
4.88

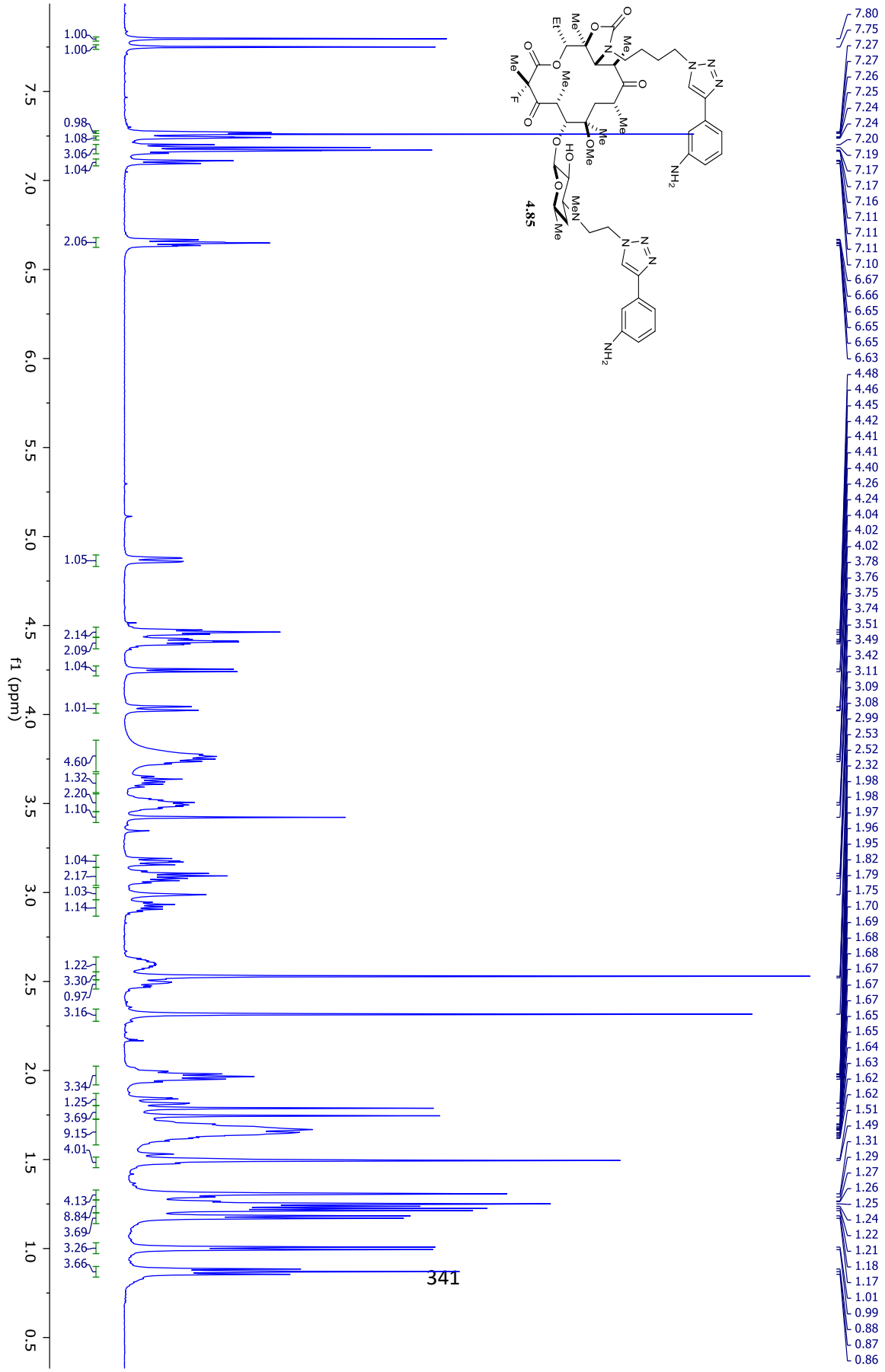

f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
203.2
202.9
166.6
166.3
157.2
104.1
98.9
96.8
82.1
80.8
78.7
70.6
69.6
66.4
61.0
52.7
51.1
49.5
49.3
44.8
43.2
40.9
39.7
39.4
37.1
29.9
26.3
25.5
25.3
24.5
22.3
21.2
19.9
18.1
15.2
14.9
13.9
10.5
4.88

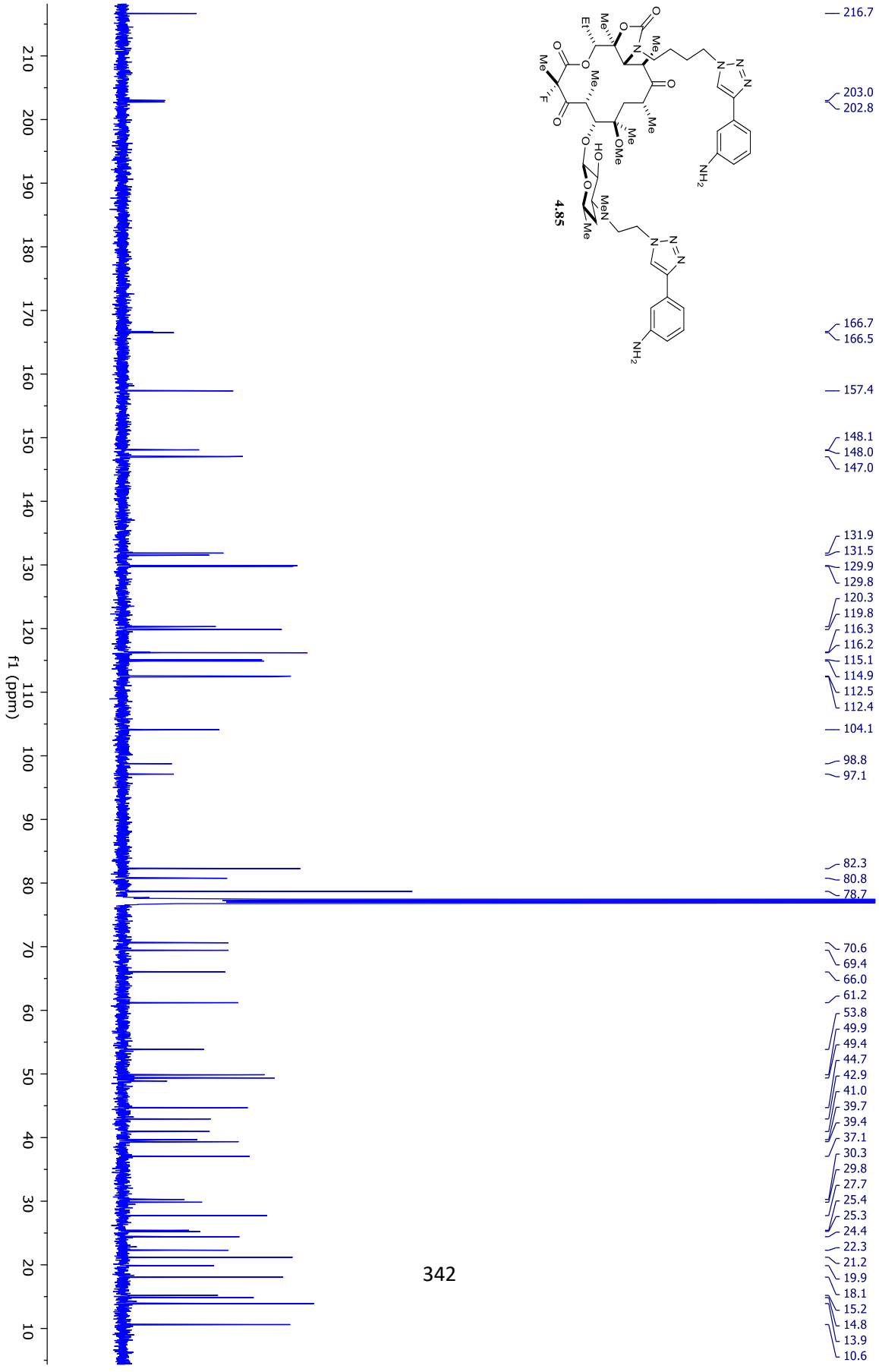

f1 (ppm)
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
1.00
0.99
0.99
1.11
3.44
1.22
0.91
4.07
1.11
1.04
2.20
3.13
1.08
0.99
2.96
1.12
1.07
1.70
3.77
11.04
3.47
3.13
3.45
5.45
3.41
3.13
3.35
4.89
4.88
4.88
4.86
4.86
4.31
4.30
4.08
4.08
4.06
4.06
3.59
3.57
3.56
3.55
3.55
3.54
3.54
3.53
3.43
3.36
3.35
3.35
3.34
3.34
3.33
3.33
3.30
3.29
3.28
3.22
3.21
3.20
3.11
3.10
2.64
2.63
2.62
2.58
2.24
1.89
1.87
1.86
1.80
1.77
1.76
1.75
1.75
1.74
1.73
1.66
1.66
1.65
1.65
1.64
1.64
1.63
1.62
1.62
1.61
1.61
1.61
1.60
1.59
1.59
1.58
1.57
1.56
1.53
1.53
1.50
1.49
1.35
1.31
1.30
1.27
1.27
1.25
1.25
1.24
1.23
1.19
1.18
1.01
1.00
0.90
0.89
0.87

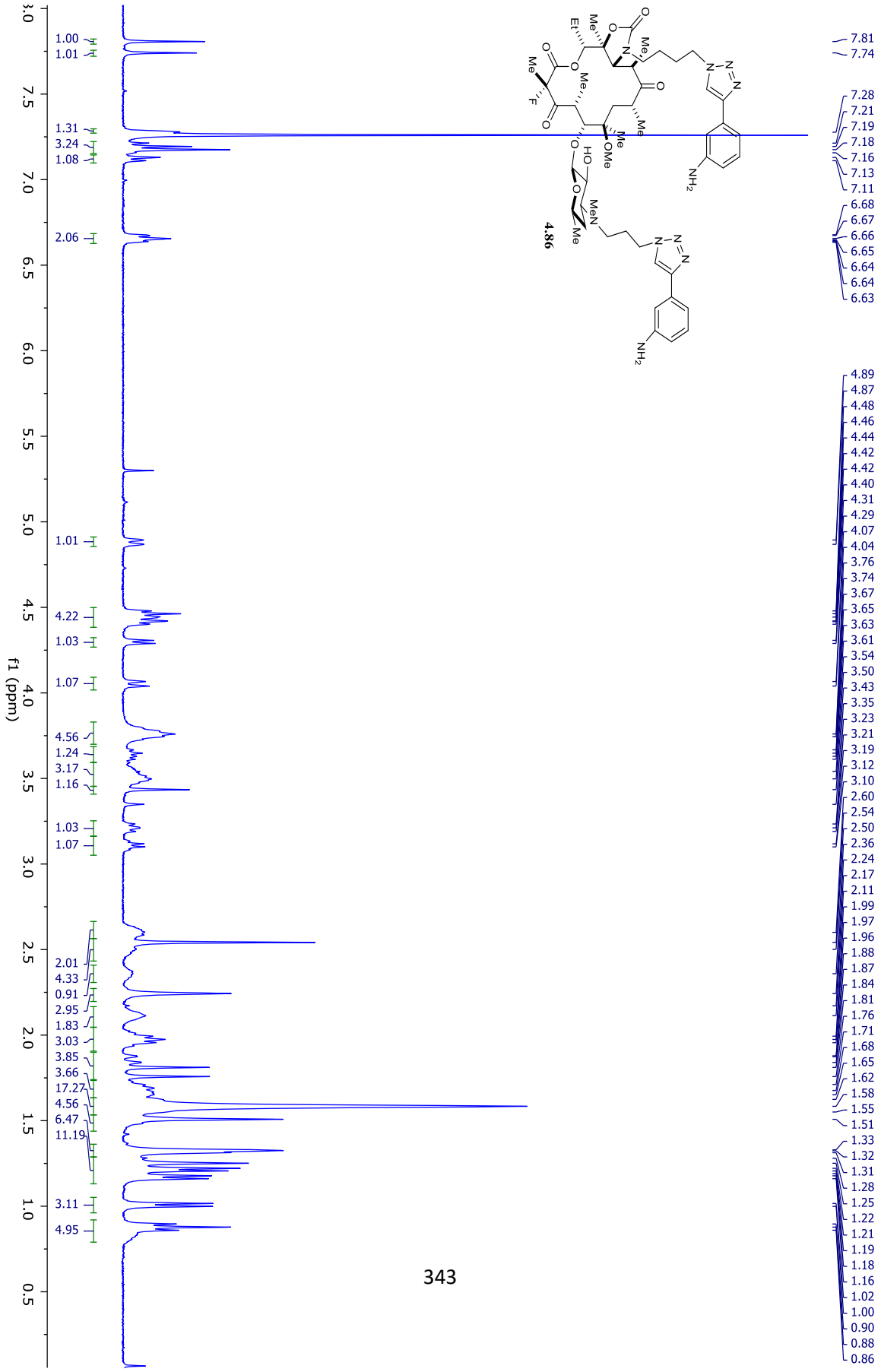

f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
203.2
166.4
157.2
104.3
98.7
97.1
82.1
80.9
78.7
70.5
69.7
66.2
61.1
51.1
50.5
49.5
49.3
44.8
44.5
43.2
43.0
40.9
39.7
39.4
37.0
29.4
27.5
26.4
25.3
24.5
22.3
21.3
19.9
18.1
15.2
14.9
13.9
10.6
4.89

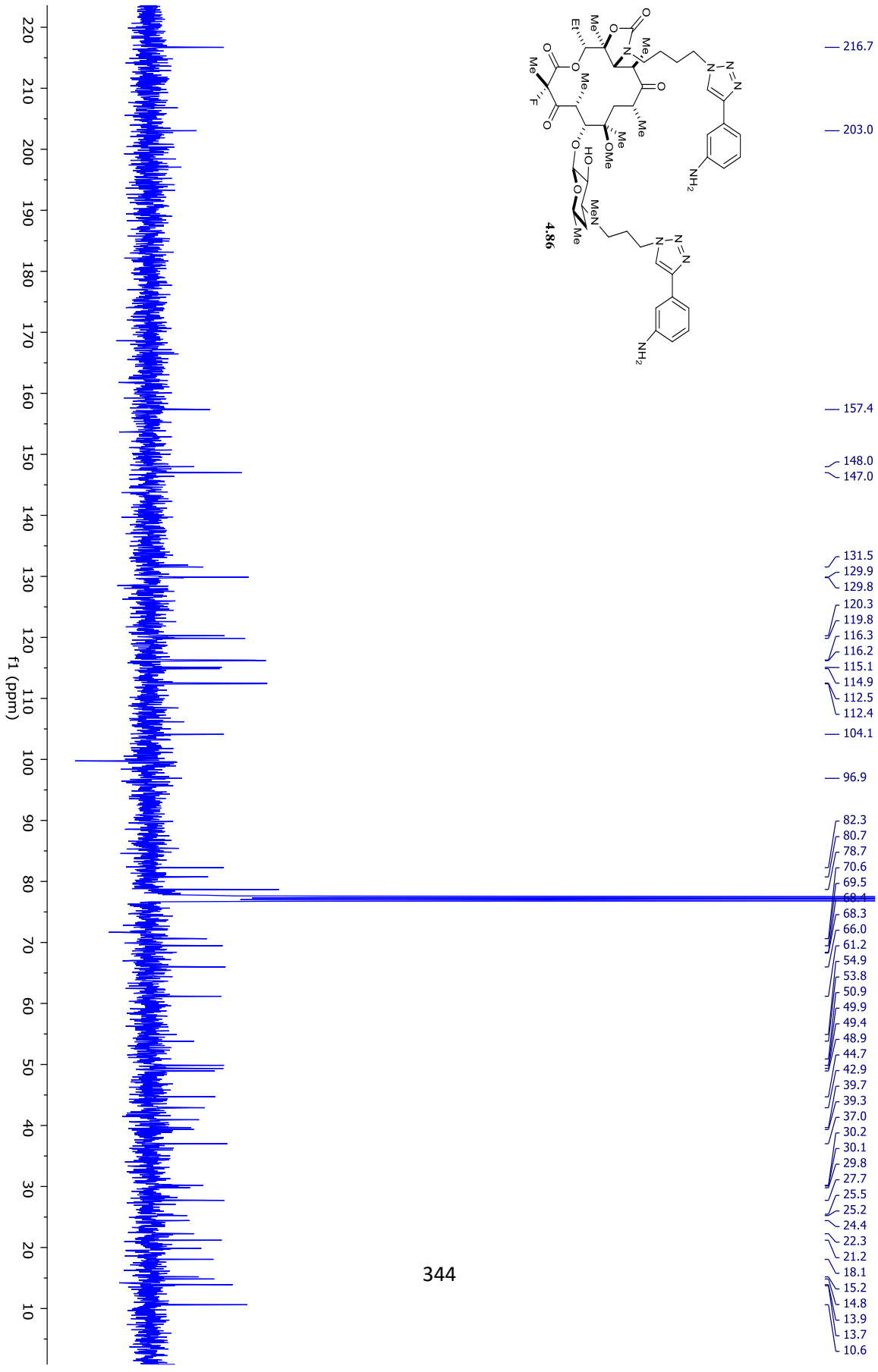

f1 (ppm)
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
0.96
0.99
1.09
4.14
1.26
4.02
1.08
1.05
0.79
4.00
1.53
0.98
2.99
1.04
1.01
3.58
16.63
15.12
3.17
3.40
4.90

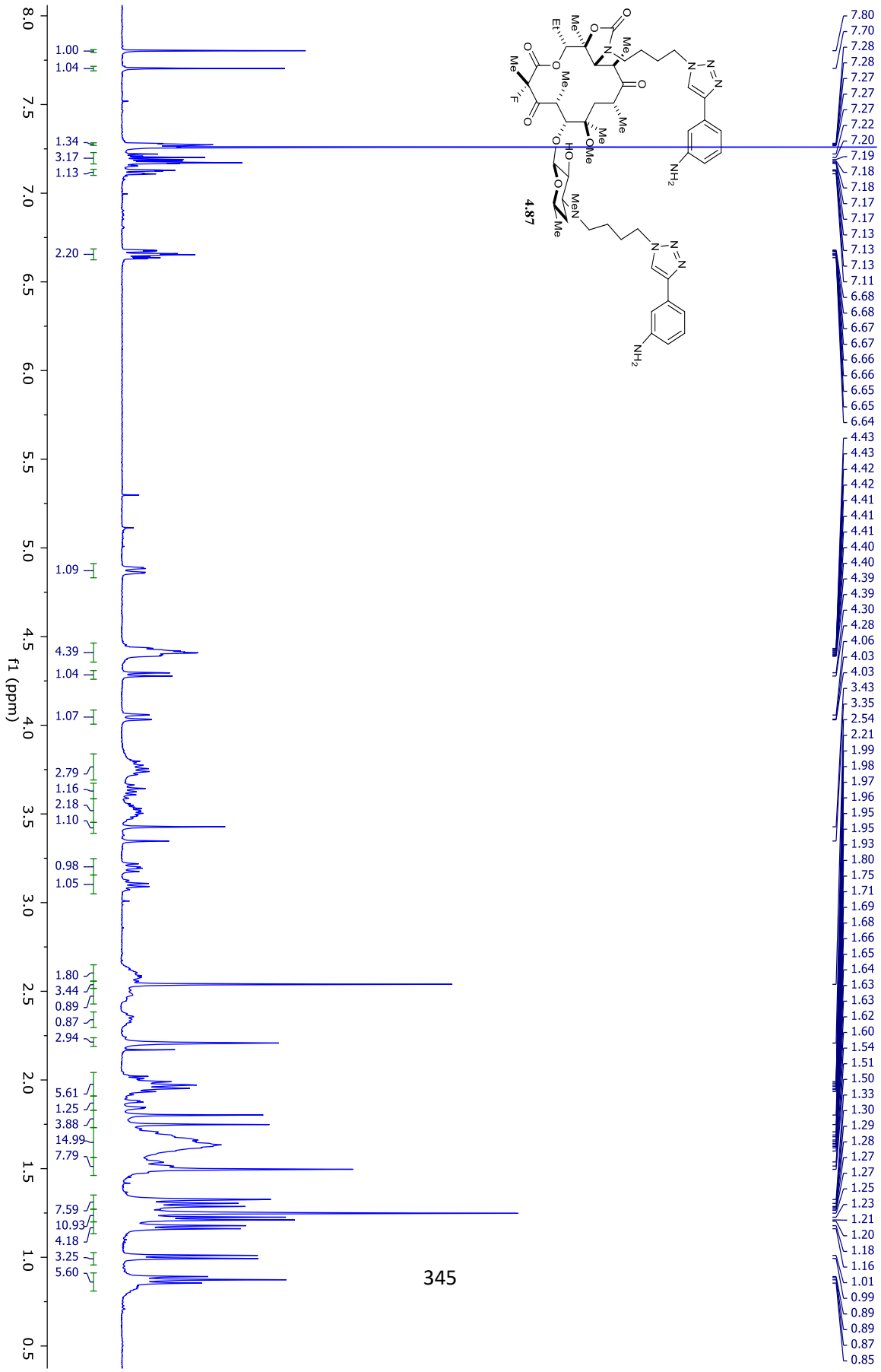

f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
216.7
202.9
166.6
157.2
104.4
98.9
96.9
82.1
80.9
78.7
70.3
69.8
66.0
61.0
52.8
51.4
51.1
49.3
44.8
43.2
40.9
39.7
39.4
36.9
29.3
26.6
26.3
25.5
24.5
22.3
21.3
19.9
18.1
15.2
14.9
13.9
10.6
4.90

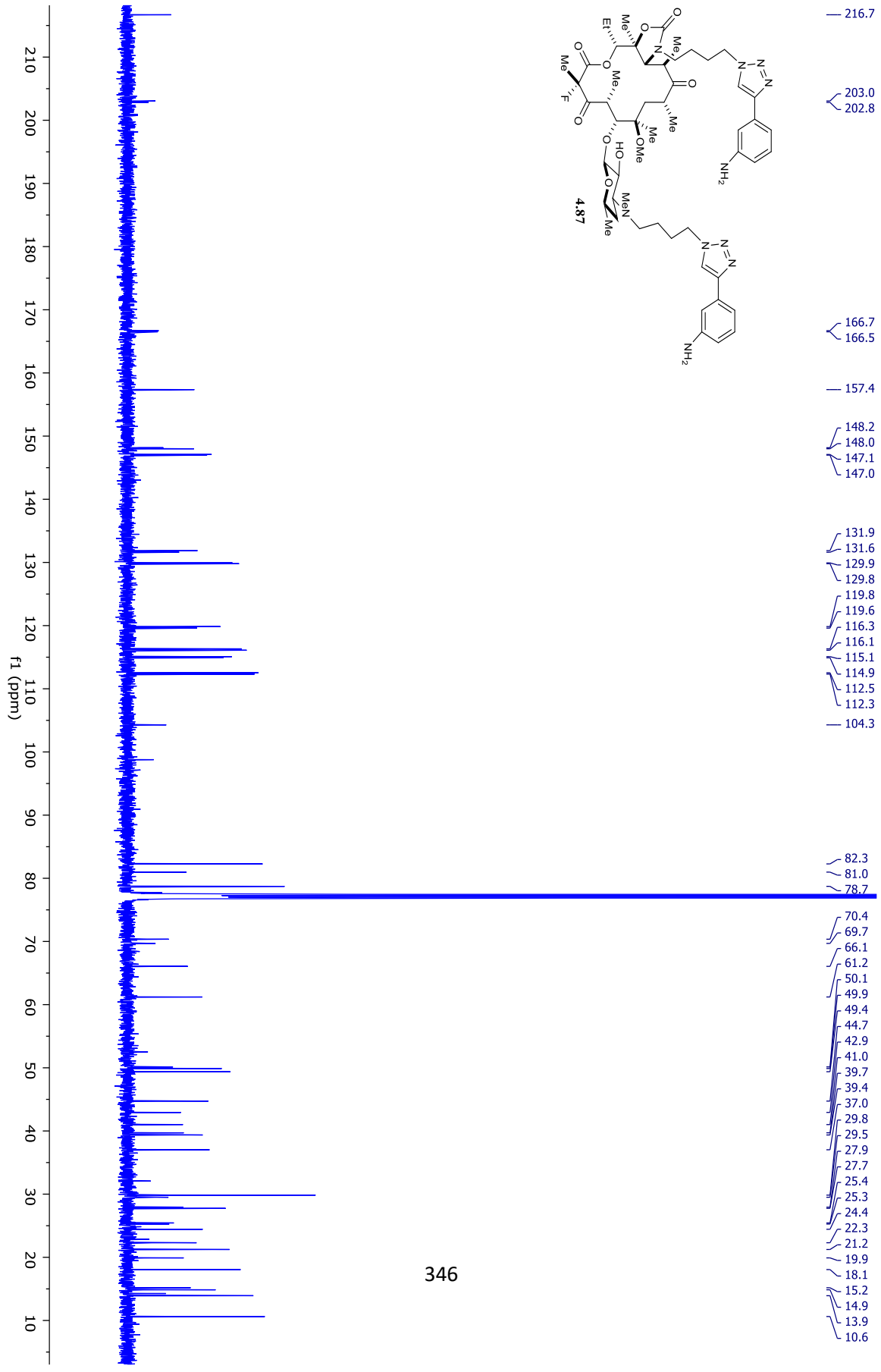

4.85
f1 (ppm)


f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
203.0
202.8
166.7
166.5
157.4
148.1
148.0
147.0
131.9
131.5
129.9
129.8
120.3
119.8
116.3
116.2
115.1
114.9
112.5
112.4
104.1
98.8
97.1
82.3
80.8
78.7
70.6
69.4
66.0
61.2
53.8
49.9
49.4
44.7
42.9
41.0
39.7
39.4
37.1
30.3
29.8
27.7
25.4
25.3
24.4
22.3
21.2
19.9
18.1
15.2
14.8
13.9
10.6
4.85


f1 (ppm)
4.86
1.00
1.01
1.31
3.24
1.08
2.06
1.01
4.22
1.03
1.07
4.56
1.24
3.17
1.16
1.03
1.07
2.01
4.33
0.91
2.95
1.83
3.03
3.85
3.66
17.27
4.56
6.47
11.19
3.11
4.95
7.81
7.74
7.28
7.21
7.19
7.18
7.16
7.13
7.11
6.68
6.67
6.66
6.65
6.64
6.64
6.63
4.89
4.87
4.48
4.46
4.44
4.42
4.42
4.40
4.31
4.29
4.07
4.04
3.76
3.74
3.67
3.65
3.63
3.61
3.54
3.50
3.43
3.35
3.23
3.21
3.19
3.12
3.10
2.60
2.54
2.50
2.36
2.24
2.17
2.11
1.99
1.97
1.96
1.88
1.87
1.84
1.81
1.76
1.71
1.68
1.65
1.62
1.58
1.55
1.51
1.33
1.32
1.31
1.28
1.25
1.22
1.21
1.19
1.18
1.16
1.02
1.00
0.90
0.88
0.86


f1 (ppm)
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
203.0
157.4
148.0
147.0
131.5
129.9
129.8
120.3
119.8
116.3
116.2
115.1
114.9
112.5
112.4
104.1
96.9
82.3
80.7
78.7
70.6
69.5
68.4
68.3
66.0
61.2
54.9
53.8
50.9
49.9
49.4
48.9
44.7
42.9
39.7
39.3
37.0
30.2
30.1
29.8
27.7
25.5
25.2
24.4
22.3
21.2
18.1
15.2
14.8
13.9
13.7
10.6
4.86


f1 (ppm)
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1.00
1.04
1.34
3.17
1.13
2.20
1.09
4.39
1.04
1.07
2.79
1.16
2.18
1.10
0.98
1.05
1.80
3.44
0.89
0.87
2.94
5.61
1.25
3.88
14.99
7.79
7.59
10.93
4.18
3.25
5.60
4.87
7.80
7.70
7.28
7.28
7.27
7.27
7.27
7.22
7.20
7.19
7.18
7.18
7.17
7.17
7.13
7.13
7.13
7.11
6.68
6.68
6.67
6.67
6.66
6.66
6.65
6.65
6.64
4.43
4.43
4.42
4.42
4.41
4.41
4.41
4.40
4.40
4.39
4.39
4.30
4.28
4.06
4.03
4.03
3.43
3.35
2.54
2.21
1.99
1.98
1.97
1.96
1.95
1.95
1.93
1.80
1.75
1.71
1.69
1.68
1.66
1.65
1.64
1.63
1.63
1.62
1.60
1.54
1.51
1.50
1.33
1.30
1.29
1.28
1.27
1.27
1.25
1.23
1.21
1.20
1.18
1.16
1.01
0.99
0.89
0.89
0.87
0.85


f1 (ppm)
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
216.7
203.0
202.8
166.7
166.5
157.4
148.2
148.0
147.1
147.0
131.9
131.6
129.9
129.8
119.8
119.6
116.3
116.1
115.1
114.9
112.5
112.3
104.3
82.3
81.0
78.7
70.4
69.7
66.1
61.2
50.1
49.9
49.4
44.7
42.9
41.0
39.7
39.4
37.0
29.8
29.5
27.9
27.7
25.4
25.3
24.4
22.3
21.2
19.9
18.1
15.2
14.9
13.9
10.6
4.87